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Biodegradation of Esters
and Olefins in Drilling
Mud Deposited on Arctic
Soft-bottom Communities
in a Low-temperature
Mesocosm

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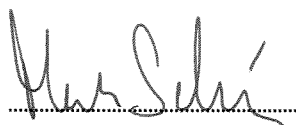
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Abstract


Discharge of chemicals from off-shore drilling operations represent a matter of concern for the oil industry and pollution control authorities world-wide. As the industry expands towards higher latitudes and greater depths, mud chemicals attached to bore hole cuttings may become deposited on sediments with ambient temperatures below 0°C. In order to assess the fate of the most recently developed ester- and olefin-based chemicals under such conditions, a comparative study was performed in the soft-bottom laboratory at Marine Research Station Solbergstrand on sediment communities transferred from Arctic and temperate deep fjord locations. The sediments were initially treated with four levels of cuttings (zero, low, medium and high dose) and maintained at simulated seabed conditions at -0.5 and 7°C, respectively. Biodegradation and effects on the sediment environment were assessed from observations of concentration change of organic phase and barium in the sediment, oxygen consumption rates, redox potentials and the macrobenthic community structure. During the three months experimental period, increasing rates of oxygen consumption in the Arctic communities showed adaptation to degradation of the chemicals at rates no less than those observed in the Oslofjord communities. The Arctic communities appeared, however, more sensitive to sulphide toxicity produced in high dose ester treatments, and no evidence was found to support the hypothesis that better availability to anoxic degraders would yield shorter sea-bed remediation times of areas contaminated with esters as compared to areas contaminated with C₁₄-C₁₈ olefins.

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Mud Deposited on Arctic Soft-bottom
Communities in a Low-temperature Mesocosm

PREFACE

The work described in this report has been performed on request from Saga petroleum a/s. The experimental work was performed in arctic and temperate mesocosms at Solbergstrand Marine Research Station (MRS) during the period April-June 1997. Sampling and transportation of 8x150kg sediment communities from Finnmark to Solbergstrand, was organised as a separate project (O-97014).

Einar Johannessen, Tom Tellefsen, Gjermund Bahr at Akvaplan-NIVA, and the crews at M/S Rusken, Honningsvåg, and F/F Trygve Braarud, Oslo, shared the responsibilities that the field work could be performed without irrevocable problems and that samples could be transferred from the two locations with a minimum of disturbance of the biological communities. *Ingegerd Rustad*, SINTEF Industrial Chemistry was subcontracted to perform the chemical analyses of barium. Klimanord AS built and delivered the cooling compressor for the Arctic mesocosm.

At MRS, *Oddbjørn Pettersen* was responsible for test set-up and daily maintenance of the experiment. *Morten Wilbergh* implemented instrumentation for monitoring of experimental environment and control of the cooling system. Extraction and GC-analyses of the organic phase was performed by *Gro Prestbakmo, Helle Juul Rasmussen* and *Svein Ingar Semb* at the chemical laboratory and biological samples were prepared by *Unni Efraimsen, Randi Romstad, Lise Tveiten* and *Pirkko Rygg* at the biological laboratory. *Rainer Lichtenthaler* and *Brage Rygg* were responsible for base fluid and biological analyses, respectively. Quality assurance was performed by *Torgeir Bakke*. Finally, thanks to *Birger Bjerkeng* for contributions to statistical analyses and preparation of Appendix 1.

Oslo, 15.12.1997

Morten T. Schaanning

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EXECUTIVE SUMMARY

Outline of the experiment

A simulated seabed study was designed to compare the fate of olefin (LAO) and ester (FOE) based drilling mud on cuttings deposited at sub-zero temperatures. Sediment communities were transferred from an arctic location, using a 50x50cm box-corer, and maintained in benthic chambers at sub-zero temperatures in the soft-bottom mesocosm at Marine Research Station Solbergstrand. For comparison, another set of communities were transferred from an Oslofjord location and maintained at the ambient temperature of 6-8°C. At time zero of the experiment, each chamber was subdivided in four 24x24cm sections and treated with, respectively, low, medium and high dose of laboratory prepared cuttings. The fourth section was left untreated for control. During the following three months, disappearance of the two organic phases and barium were determined from GC and X-ray analyses on initial and final sediment samples. Biodegradation rates were assessed from bi-weekly determination of oxygen consumption, and effects on redox potentials and macrofauna communities were compared after three months exposure to the cuttings.

Collection and maintenance of test communities

Test communities were sampled using a 0.25m² USNEL steel box corer fitted with an internal acrylic liner. A bottom plate was attached to the liners before removing the samples from the box corer and the overlying water was drained off to reduce erosion of the sediment water interface during transport and handling. In the mesocosm, each liner holding a 48x48x30cm seabed sample was sealed with a lid and fitted with an internal current generator and a flow through system for continuous exchange of the 10 cm layer of water overlying the sediment. This sampling technique allowed transfer of seabed samples and establishment of benthic chambers designed for optimum experimental performance with negligible disturbance of natural sediment stratification and biological habitats.

Eight communities were sampled 12.02.97 at 212m depth in the Oslofjord and transferred to the mesocosm laboratory within less than 12 hours after collection. In the mesocosm they were maintained under a continuous flow of seawater from 60m depth until initiation of the experiment 06.04.97.

A suitable arctic location was chosen in the inner part of Porsangen, Finnmark, in a basin separated from the main fjord by sills at 30-40m depth. Regular hydrographic surveys of the basin, performed in 1993, had revealed weak density stratification during winter and temperatures below zero from January until June. During our survey, temperatures ranged from -1.15°C at the surface to -1.35°C in the bottom water and the salinities were almost non-variable throughout the watercolumn (33.99 ± .05 PSU). The existing data indicated seasonal deep-water renewal, presumably driven by cold off-shore winds and ice-formation during winter. Thus, eight test communities adapted to the low temperatures were collected 11.03.97, from 120m depth. The communities were carefully sealed, insulated and transported to Solbergstrand, where they arrived less than 36 hours after collection.

13.03.97, two samples from each location were processed for biological sampling (zero samples). The remaining 12 boxes were converted to benthic chambers, continuously supplied with separate flows of seawater from 60m depth in the fjord adjacent to the research station. The arctic samples were kept submersed in a separate tray maintained at -0.5°C by circulating cooling liquid through a copper circuit in the tray water. In addition, the

continuous supply of fjord water to each of the arctic chambers was precooled through tube coils submersed in the tray water. Thus, whereas the Oslofjord communities were maintained at the ambient fjord water temperature of 6-8°C, the arctic communities were maintained in the same water cooled to temperatures below 0°C.

Throughout the experimental period, the salinity of the source water did not vary beyond a range of 33-35 PSU, and apart from short periods of sampling or inspection, the experimental hall was kept in dim light to avoid primary production.

Preparation and addition of “cuttings”

Two different mud samples were prepared by Anchor/M-I Drilling Fluids A.S., Forus. One was based on a mixture of ¹⁴C and ¹⁶C α-olefins (LAO). The other was based on a mixture of saturated and unsaturated esters produced from natural, marine fatty acids (FOE).

Based on the concentration of base fluid (ca. 35% by wght.) given by Anchor/M-I, aliquots of the mud samples were diluted with dried marine sediment to yield a simulated discharge product (“cuttings”) with a base fluid content of 10%.

On day zero, each of the benthic chambers were subdivided into four identical 24x24cm sections by insertion of 5 mm acrylic walls to a sediment depth of 15 cm. The “cuttings” were suspended in sea water and added to the surface of each section via sedimentation through a small column of overlying water. The four sections in each chamber were treated with 0, 5-7, 50 and 200 mg (wet wght.) of the respective suspension. These additions corresponded to nominal loads of 0, 0.5, 5 and 20 mgOP·cm⁻² (OP = Organic Phase) or nominal thickness of “cuttings” layer of 0, 0.1, 1 and 4 mm.

Initial analyses of strategic samples showed the presence of 0.42±0.22 mgOP·cm⁻² in low dose sections, 4.33±0.69 mgOP·cm⁻² in medium dose sections and 19.0±2.1 mgOP·cm⁻² in high dose sections. This was reasonably consistent with the concentrations predicted from the amounts of mud added and the mud composition given by Anchor/M-I. The data gave no reason to differentiate initial concentrations, neither between the two chemicals nor between the two environments.

Statistical analyses

The experimental design allowed three-factor ANOVA analyses to be performed on any parameter measured in the various sediment sections. The factor *Environment* had two levels (Oslofjord and Arctic), the factor *Substance* had two levels (Olefin and Ester), and the factor *Dose* had four levels (Control, Low, Medium and High). Chamber effects were taken into consideration by nesting each of the twelve chambers as a random variable within *Environment* and *Substance*. The analyses were performed using JMP® Statistical Software (SAS Institute Inc.).

Cross-contamination

The differential treatment of the four sections within each chamber represented a potential problem with regard to spreading of contaminated cuttings from higher into lower dosed sections. However, concentration of base fluids were less than analytical detection limits in five of the six randomly selected control sections sampled four days after addition of cuttings. In the six'th sample, traces of esters (0.018 mgOP·cm⁻²) were determined, but hardly distinguishable from the back-ground of naturally occurring fatty acids. Thus, the initial

samples showed that cross-contamination during test set-up was negligible. Neither did barium determined in final samples reveal any evidence for cross-contamination during the experimental period.

Disappearance of barium

Background concentrations of barium were 0.6-0.7 mg g⁻¹ (dry wght.) in the arctic sediment, slightly higher in the Oslofjord. Within the standardised 0-3 cm sampling depth interval, the concentrations corresponded to 0.8-1.0 mgBa cm⁻² at both locations. In the treated sediments, after subtraction of these background concentrations, mean initial barium was 0.54 (n=6), 3.62 (n=6) and 13.7 (n=2) mg cm⁻², respectively, in low, medium and high dose treatments. In the final samples (n=12), the corresponding concentrations and estimated standard deviations were 0.50±0.14, 3.56±0.76 and 12.5±1.6 mg cm⁻². This showed that negligible amounts of barium had disappeared from the test sediments during the 3 months experimental period.

Bioturbation

Several individuals of the star-fish *Ctenodiscus crispatus* were present in all sections of the arctic sediments. The disc-shaped organism had a diameter of 2-3cm. Numerous tracks revealed considerable horizontal mobility within the surface layer. The species was, therefore, a potentially important bioturbator present in the arctic sediment only, and rapid mixing between added cuttings and chamber sediments as well as the lack of formation of bacterial mats in the Arctic sediments, were obviously related to the activity of this species.

However, at the end of the experimental period, no significant decrease of barium was found within the 0-3 cm layer, and sectioned core samples showed that ≥99% of the cuttings particles were still present within the 0-1 cm layer. This distribution was also found in core samples from the Oslofjord. Thus, in spite of the activity of *Ctenodiscus*, downwards mixing of cuttings particles to depths beyond 1 cm had been negligible. Nevertheless, it was concluded that bioturbative dilution of cuttings had occurred within the 0-1 cm layer of the arctic sediments and that biodegradation of the organic phase might have been enhanced by this process.

Disappearance of organic phase

In the low dose treatments, the olefins were not detectable in the Oslofjord sediments and esters were present in trace amounts, only (0.005 mgOP cm⁻²). This result was important, showing that given a low initial dose and sufficient time, both of these drilling fluids will disappear completely from the sediment. In the arctic sediment, 0.03mg cm⁻² of the ester and 0.11 mg cm⁻² of the olefins were still present by the end of the experimental period. Corresponding to 7% and 26% of the initial concentration, disappearance was somewhat slower in the arctic environment.

A similar pattern was found in the medium dose treatments, but a larger fraction of the added chemicals had remained present. In the high dose treatments, as much as 64% of the ester and 76% of the olefin had remained present in the Oslofjord sediment. Normalised against barium, 74% was found to remain of both organic phases. So far, the results were consistent, showing that the relative disappearance of both fluids decreased with increasing loads and decreasing temperatures, and that the olefin tended to disappear more slowly than the ester. Also, the recovery of 79% of the added ester (Ba-normalised 84%) in the high dose arctic sediment, was consistent with these trends. However, the recovery of only 61% (Ba-normalised 74%) of the olefins in the arctic high dose sediment was surprisingly low.

The ANOVA analyses on disappearance showed strong dominance of the *Dose* factor. Neither *Environment* nor *Substance* contributed significantly to the variance observed in the final concentrations. However, the probability ($p=0.014$) of the *DoseEnvironmentSubstance* factor indicated that the rate of disappearance had been affected by complex three-ways interactions between the local environment and the type and amount of drilling fluids added.

A rather extraordinary behaviour of *Ctenodiscus* in one of the arctic chambers treated with esters, represented a possible clue to the understanding of these observations. The sediment surface in the medium and high dose sections had turned black from precipitated ferrous sulphide and several individuals of the species had escalated from their normal buried position, lying upside down on the surface, facing mouth and respiratory organs towards the oxygenated chamber water. Also, all tracks, which were present in all other sections of the arctic sediments, had vanished. Obviously, the *Ctenodiscus* had become immobilised by sulphide toxicity. Furthermore the concentrations determined seemed to confirm that disappearance of esters had been particularly small in the affected sections.

Half-lives

The experiment was not designed for analysing time trends of the concentration data. Nevertheless half-lives were calculated from exponential regression analyses of initial and final OP:Ba concentration ratios. The analyses showed significant decrease of the ratios in all treatments. For the medium dose ester treatments, half lives of 57 days were found for the Oslofjord and 58 days for Porsangen. In the high dose treatments, half-lives increased to 198 and 340 days, respectively. Similarly the half-lives for the olefin:Ba ratios, increased with dose from 65 to 206 days in the Oslofjord and from 92 to 205 days in Porsangen sediments.

The half-life of 65 days for the medium dose olefin treatment was consistent with previously determined LAO:Ba half-lives of 45-76 days (95% confidence interval at an initial load of 4 mgOP cm⁻²), but 57 days for the ester was high compared to the previously determined confidence interval of 15-22 days at an initial load of 6 mg cm⁻². Probably, the rapid enzymatic hydrolyses of the ester, yielding rapid initial decline of concentrations, was not properly accounted for by the present design omitting all intermediate samples between time zero and three months.

The strong dose dependency of the half-life was confirmed. The surprisingly short LAO:Ba half-life in arctic high dose sediments was merely a result of the concentration data as discussed above.

Sediment oxygen consumption

Sediment oxygen consumption (SOC) was measured bi-weekly on whole chambers integrating the four dose levels. Thus, the structure of the SOC-data, was different from the other parameters which were measured in each treatment, but only at two or three occasions.

In the Oslofjord chambers, SOC varied about a level of approximately 500 $\mu\text{molO}_2\text{m}^{-2}\text{h}^{-1}$, interrupted by a broad peak which culminated at about day 60. Initially, SOC was lower in the Arctic than in the Oslofjord chambers. However, the rates increased slowly during the experimental period and towards the end of the experimental period (day 75), the SOC rates in several of the Arctic chambers crossed over the rates in the Oslofjord chambers. This indicated slow adaptation of the Arctic degrader community to the added chemicals.

The most remarkable feature of the oxygen consumption data, was the absence of any difference between the ester and olefin treatments. For the whole period, 839-1023 mmolO₂m⁻² was consumed in the three arctic chambers treated with olefins as compared to 764-956 mmolO₂m⁻² for the three ester chambers. In the Oslofjord chambers, the corresponding results were 830-1383 mmolO₂m⁻² for the olefins and 1053-1287 mmolO₂m⁻² for the esters.

A three-factor ANOVA analyses was performed also on the SOC data, by replacement of *Dose* with *Time* as the third factor in addition to *Environment* and *Substance*. The analyses showed four significant effects ($p \leq 0.05$). The *Environment* with an F-ratio of 65.6 was the most important factor controlling the rates of oxygen consumption. *Chamber* effects were found with an F-ratio of 11.1, and also *Time* and *Environment**Time* interactions were significant, whereas *Substance* with an F-ratio of 1.7 had no significant impact on the oxygen consumption rates.

Low temperatures may not be the only element responsible for the *Environmental* effect. The *Chamber* effect revealed inherent differences between separate chambers, most likely a result of inherent differences with regard to number of individuals (biomass). Similarly, the effect of *Environment* may have been an inherent effect relating to the generally higher individual numbers in the Oslofjord communities.

Redox potentials

A characteristic difference was shown to prevail between the vertical profiles of redox potentials in the sediments from the two locations. In the arctic sediment, a consistent redox-cline was observed within the 1-3 cm depth interval in all initial profiles as well as in control sections measured 1 and 3 months after addition. A similar transformation was found in the Oslofjord sediment, but the decrease was less sharp and occurred at various depths below 4 cm. The characteristic redox profile of the Arctic sediment appeared to be maintained by the activities of two macrobenthic species, the surface bioturbator *Ctenodiscus crispatus* and the tube-dwelling polychaete *Spiochaetopterus typicus*. The bioturbator maintained potentials of about 200 mV within the surface layer, whereas an upwards flux of pore water maintained by the polychaete assemblies at 20cm depth, might explain the potentials of about 0mV throughout the analysed sub-surface layer.

30 days after addition of cuttings, effects were observed in ester treatments from both locations. In the Oslofjord chambers, lowered redox potentials were only observed beneath a few white spots which had appeared in the high dose sections. In the Porsangen chambers, neither white spots nor any other miscoloured sediments were observed in any treatment. Nevertheless, duplicate profiles revealed lowered redox potentials within the 0-1cm depth interval in medium and high dose treatments. In the ester treatments, the observed effects were stronger than in the olefin treatments.

During final sampling, redox potentials were determined at 0.5cm depth at three different locations in each of the 48 sections. ANOVA analyses showed significant ($p \leq 0.05$) effects of all three factors as well as two-ways *Dose**Environment* and *Dose**Substance* and three-ways *Dose**Environment**Substance* interaction effects. *Dose* could account for most of the variation (F ratio = 50.3) followed by *Substance* (F ratio = 33.4) and *Environment* (F ratio = 21.2). Chamber effects were not significant. The data showed that the redox potentials were more strongly lowered by esters than by olefins, and more easily lowered in Arctic than in Oslofjord sediments. Potentials indicating the presence of hydrogen sulphide were only observed in some of the ester treatments.

Macrofauna communities

All animals retained after washing the final sediment from each section through a 1 mm mesh size sieve, were sorted into main groups. Most groups were determined to species level. In the Oslofjord samples, 56-460 individuals of 14-43 different species were identified in each sample. Diversities (H) ranged from 2.93 to 4.03. Thus, practically, all sections could be classified as a class II ("good") benthic environment. In the arctic sediment, the fauna was less diverse showing 53-132 individuals of 14-23 different species. The corresponding range of diversities of 1.98-3.52 was consistent with the diversities determined in zero-samples sieved two days after sampling on the arctic location. Thus, natural, biogeographical variation was concluded to account for the major difference between the macrobenthic communities from the two locations.

ANOVA analyses showed significant effect of *Dose* on the Shannon-Wiener diversity index. The effect was not very strong, accounting for an F ratio of 3.9 ($p=0.02$) as compared to the F ratio of 19.5 for the *Environment*. The latter was however an inherent effect resulting from the different diversities at the two source locations, whereas the dose effect was a result of experimental treatments. The results of the ANOVA analyses was confirmed by MDS-plots showing that the fauna in different treatments within each chamber were in general more similar than the fauna in replicate treatments in different chambers.

The fact that significant chamber effects were found both with regard to sediment oxygen consumption and total number of individuals suggested a coupling between total sediment respiration and the biomass of the macrobenthic community. Linear regression analyses of total sediment oxygen consumption versus number of individuals, showed a positive correlation, but the significance level was low. The significance level improved, however, strongly after rejection of one or two of the twelve observations.

In spite of up to 4x higher doses, and clear effects on sediment oxygen consumption and redox potentials, the effects on macrobenthic communities in the present experiment were small compared to those observed in a previous test. A major fraction of the added base fluids were, however, still present in the sediments and the growth of bacteria mats in Oslofjord sediments as well as the immobilisation of *Ctenodiscus* in one of the arctic chambers, clearly indicated that effects were about to come. Probably, three months exposure was too short for manifestation of structural effects in the macrobenthic communities.

Conclusions

Throughout the three months experimental period, temperatures were maintained close to 7°C in chambers transferred from the Oslofjord and -0.5°C in chambers transferred from the Arctic.

Base fluid disappearance rates were not significantly different in the two environments. Esters tended to disappear faster than the olefins, but not in the high dose Arctic treatments.

Initial oxygen consumption rates were lower in Arctic than in Oslofjord chambers, but increasing trends and cross-over towards the end of the experimental period showed adaptation of the Arctic degrader community to the added chemicals. Carbon-fluxes may have been more important than temperatures in explaining inherent biological differences between the two locations.

With regard to effects during the degradation event, the tendency of the esters to stimulate sulphate reducing bacteria and indications found that functionally, the Arctic communities

were more sensitive to sulphide toxicity than the Oslofjord communities, would tend to disfavour discharge of esters in Arctic environments.

No evidence was found to support the hypothesis that rapid anoxic degradation would yield shorter sea-bed remediation times of areas contaminated with esters as compared to areas contaminated with LAO-olefins. On the contrary, indications were found that maintenance of bioturbation in sediments heavily contaminated with LAO-olefins were more important with regard to sea-bed remediation than the availability of the esters to sulphate reducing bacteria.

1. INTRODUCTION

At about 1990, concern with regard to environmental effects of drill cuttings deposited on the seabed led to restrictions on off-shore discharges of cuttings containing mud based on mineral oil (Dicks *et al.*, 1986/87, Neff *et al.*, 1987, Reiersen *et al.*, 1989, Bakke *et al.*, 1989b, Gray *et al.*, 1990). Since then new drilling muds have been formulated in which mineral oil has been replaced with various organic phases. Discharge of cuttings from drilling operations in which the new muds have been applied, has been permitted on a case to case basis (Reiersen, 1990). In order to improve the basis of such decisions, an experimental procedure, frequently referred to as a simulated seabed test, has been developed at NIVA's Marine Research Station Solbergstrand (MRS).

In those tests, cuttings contaminated with various drilling muds are added in thin layers on top of mixed or undisturbed marine sediments. Biodegradation and effects on redoxpotentials and macrobenthic communities are studied for experimental periods of 3-9 months. The sediments are continuously flushed with sea water supplied from 60m depth in the Oslofjord. Thus, all experiments so far have been performed at the ambient salinity and temperature of the fjord water, ranging 32-35 PSU and 6-11°C, respectively.

As the industry expands towards higher latitudes and greater depths, cuttings may become deposited on sediments with ambient temperatures down to the freezing point of sea water. In such environments, weathering processes, metabolic rates, bacterial abundancies and macrobenthic community responses may differ from those observed at higher temperatures in communities from the Oslofjord. Therefore, it was not only decided to perform a test at temperatures below zero, but also to transfer undisturbed sediment communities from an arctic location to the Solbergstrand laboratory.

Most of the current knowledge on fate of hydrocarbons in arctic environments, has been derived from studies focused on problems related to accidental spills of mineral oil. Such studies have shown that hydrocarbon-utilising bacteria appear to be ubiquitous in marine environments (Arhelger *et al.*, 1977, Pedersen *et al.*, 1979, Leahy and Colwell, 1990) and bacterial responses in arctic environments have not been found to be much different from those described in more temperate regions (Eimhjellen, 1982, Thingstad and Martinussen, 1991). Thus, the potential exist for biodegradation of mineral oil in arctic environments. It appears likely that this conclusion also applies for the mineral oil substitutes in drilling muds.

In spite of the documented efficiencies of bacteria adapted to low temperatures, biodegradation of actual and simulated spills of crude oil in Arctic environments has been found to proceed slowly (Gulliksen and Taasen, 1982, Haines and Atlas, 1982, Boehm *et al.*, 1987). Higher viscosity at low temperatures may slow down weathering and microbial attack, and long lag periods before degradation have been related to slow evaporation of toxic components (Atlas and Bartha, 1972, Atlas, 1975, Payne *et al.*, 1991). In these respects, mineral oil substitutes in cuttings deposited on sublittoral sediments may possibly perform better than crude oil or diesel deposited in shore-line environments. Frequently, other environmental factors such as the availability of oxygen and nutrients have been considered more important than low temperatures as limiting factor for degradation rates (Atlas, 1985, Bartha and Atlas, 1987, Macdonald and Bewers, 1996).

In addition to differences with regard to microbial degradation rates, arctic macrobenthic communities may respond to hydrocarbon pollution, in different ways than those observed in

communities from lower latitudes (e.g. Sanders *et al.*, 1980). Adsorption to clay particles and rapid transfer to benthic communities was found after an oil spill at Svalbard (Senstad, 1979). Two years after the accident, concentrations were still elevated. However, due to the poor natural fauna at the spill site, few effects could be observed on macrobenthic organisms (Gulliksen and Taasen, 1982). Reported impacts from other spills in northern environments vary from severe (Wikander, 1982) to small (Kingston *et al.*, 1995). The thorough investigations after the *Exxon Valdez* accident revealed large effects on shore-line communities, but also strong recovery potentials through recruitment from unaffected areas (Stoker *et al.*, 1992, cit.). No indications have been found that hydrocarbons would have different effects on benthic animals in arctic as compared to temperate regions (Evenset and Hansen, 1994).

Thus, current knowledge is to a large extent derived from studies addressing chronic or accidental pollution from mineral oil hydrocarbons and remediation of shallow littoral and ice-edge environments. Reports addressing sub-littoral benthic communities were scarce and even less is known about the new organic phases. Furthermore, in the aquatic environment, even within the arctic region, sub-zero temperatures rarely prevail. Therefore, considerable efforts were taken to perform the present seabed simulation experiment on soft bottom communities transferred from an arctic environment with ambient temperatures below zero. The transplanted sediments were treated with muds based on a ^{14}C - ^{16}C olefin, and an ester produced from natural, marine fatty acids. Thus, two of the most recent and easily degradable organic phases were chosen for the experimental work.

2. MATERIAL AND METHODS

2.1 TEST PRINCIPLES

The test principles have been developed through several experiments on biodegradation and effects of mineral oil and synthetic base fluids on cuttings deposited on test sediments. The idea is to establish a series of replicate experimental systems, which are maintained in easily accessible indoor basins. Each system is referred to as a benthic chamber (Figure 1) and the degradation environment inside the chambers is made to resemble the conditions at the discharge site as closely as suitable to the purpose.

The chambers have an area of 48 x 48 cm and a height of 35 cm. The four walls (frame) of the chambers are dismantled and fitted into the steel box of the USNEL box corer. In the field, the steel box and frame is lowered to a seabed penetration depth of 25-30 cm. On reversal of the winch, the spade digs into the sediment below the sample and the entire 48x48xca.25cm section of the seabed is lifted to the surface. Onboard the boat, a bottom plate is inserted between the spade and the frame before dismantling frame and sample from the steel box. In order to prevent sediment erosion and spilling during handling and transportation to the soft bottom laboratory, the overlying water is drained off and the chambers are sealed with temporary lids.

At Solbergstrand the chambers are placed in larger trays continuously flushed with seawater supplied via pipe-line from 60m depth in the Oslofjord adjacent to the research

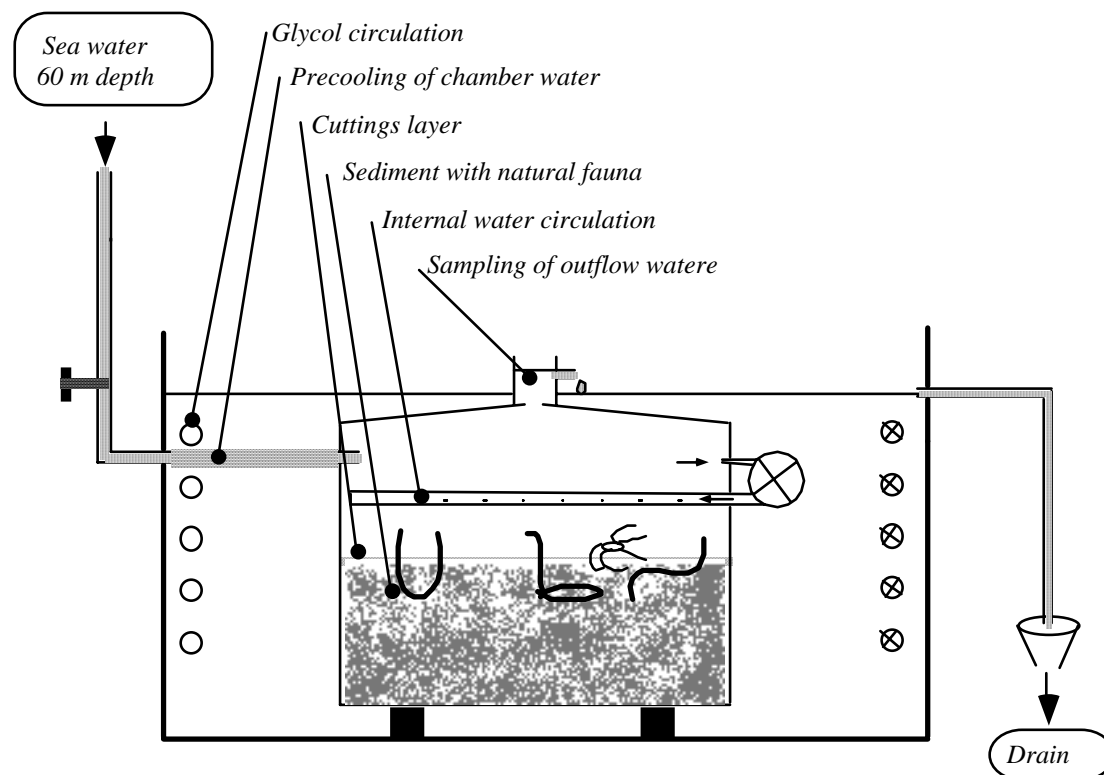


Figure 1. Schematic drawing of experimental unit.

station. Then the chambers are stored for the period of time required for experimental preparations and adaptation of sediment communities to the experimental environment. The present set-up in the soft-bottom laboratory is schematically shown in Figure 3.

2.2 FIELD WORK

2.2.1 Oslofjord

For the present experiment eight box core samples were collected from RV Trygve Braarud 12.02.97 at 212 m depth in the Oslofjord at a station located at 59°36:120 N, 10°38:257 E. Within 12 hours, the samples were transferred to Solbergstrand and submersed in two trays continuously flushed with water from 60m depth. Thus, the samples were stored for the two months required for the remaining preparations of the experiment.

2.2.2 Porsangen

Hydrography of the arctic location

Roddenessjøen (Raaddenjargsjøen) is an approximately 25km long and 5km wide basin located in the inner part of Porsangen, Finnmark. The basin is enclosed by land in north, east and south and a chain of islands in the west. The maximum depth of the sounds connecting the basin and the main fjord is uncertain, but map and hydrographic data indicated a sill depth at 30-35m. Data from hydrographic surveys available on the net (<http://lupus.nfh.uit.no>) (Appendix 3) showed salinities between 33.5 and 34.0 PSU, weak density stratification and temperatures below 0°C during winter, rarely increasing to above +2°C during the warmest months. During the present survey (Figure 2), the temperature increased slightly with depth from -1.35°C at the surface to -1.15°C in the bottom water. The very weak density stratification (ref. vertical profile of sigma-t in Figure 2) showed that any increase of the density of the surface water by cooling, evaporation or ice formation, might easily trigger vertical circulation of this watercolumn. During winter such processes may provide frequent renewal of the basin deep water and supply oxygen for the benthic community.

Sampling and transportation

Immediately after the Oslofjord survey the equipment was shipped to Honningsvåg and reloaded to the tugboat "Rusken". Eight box core samples were collected at 120m depth in the deepest part of Roddenessjøen, at 70°12:54N, 25°15:02 E. The samples were collected 11.03.97 between 9:00 and 16:00 using the box corer as described above.

The samples were distributed onto four "euro-palls" with wooden frames. For protection during transportation, the overlying water was carefully drained off and the samples were sealed with a lid, insulated with 5cm polystyrene sheets and wrapped in a water-resistant cover. At Russeneset the samples were reloaded from "Rusken" to a motor truck and driven to the airport in Alta. Here, the samples were stored over-night in a non-heated warehouse. The next day, the cargo had to be divided on two separate flights leaving Alta at 12:00 and 15:30. Thus the last containers arrived at Fornebo, Oslo, at 18:35 and further by truck to Solbergstrand at which the samples arrived at 21:00, less than 36 hours after sampling on the field location. Six of the boxes were immediately placed in the refrigerator tray constructed for the purpose and flushed with pre-cooled water from 60m depth in the Oslofjord. Cold seawater was added to the remaining two boxes, which were sieved for biological sampling on the following day.

On arrival, the sediment surface was covered by a few mm brown slurry. Remnants of

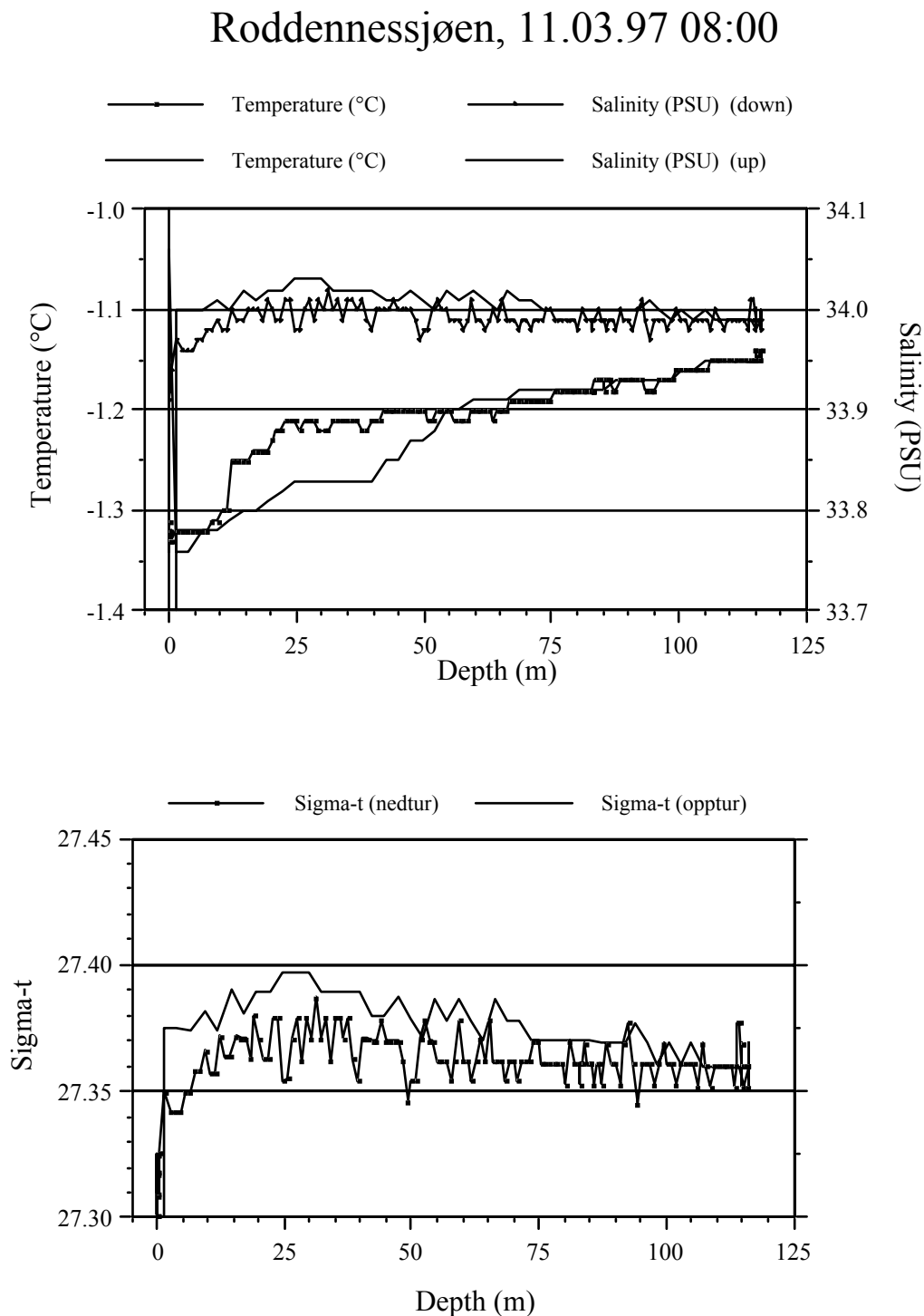


Figure 2. Vertical profiles of temperature, salinity and density recorded on a “Gyttre” recorder mounted on the USNEL box corer during field work in Porsangen.

splattered slurry on the inside of the lid covering the chambers revealed some physical disturbance during air and truck transportation. However, below the surface slurry, the clay sediment had remained firm, and no indications were found that the benthic community had suffered during transportation. During the first night at Solbergstrand the slurry disappeared

and numerous tracks of cushion stars and tubes of sediment-dwelling polychaetes extending into the overlying water revealed diverse biological activity in the chambers.

During sampling, the sediment had a temperature of -1°C . Air temperatures remained below or close to 0°C during sampling and transportation in Finnmark, as well as in the warehouse at Alta airport. Air temperatures increased to $10\text{-}15^{\circ}\text{C}$ during air transportation and after arrival in Oslo. On arrival at Solbergstrand temperatures of $2\text{-}3^{\circ}\text{C}$ and a concentration of oxygen of 5 mg l^{-1} were measured inside the slurry layer covering the sediment samples. It appears not likely, that the small temperature increase during transportation would have had any severe impact on the health of the test communities.

2.3 TEST SET-UP

The test set-up is shown in Figure 3. The samples from the southern location were placed in two trays (ca $1\text{x}3\text{x}0.7\text{m}$) continuously flushed with SSW-60 (Solbergstrand Sea Water - 60m depth), which was continuously supplied from the Oslofjord adjacent to the research station.

The samples from Porsangen were placed in a temperature stabilised water bath constructed for the purpose. The bath (ca $1\text{x}5\text{x}0.7\text{m}$) was filled with the same source water (SSW-60) cooled to experimental temperatures by circulating cooling liquid (glycol) through $9\text{ }1/2''$ copper tubes attached to the walls of the tray. The total copper surface was 4.3 m^2 . To improve heat transfer and temperature uniformity, small circulating pumps were placed between the sediment chambers.

Cooling was provided by a 2 kW compressor with an internal shunt circuit which allowed for 40-100% load variation. The cooling liquid from the compressor was kept at -4°C with a normal return temperature about -1.5°C . Temperature of the water bath was controlled by circulating the cooling liquid from the compressor, through a 100 litre buffer tank, followed by a three-way valve controlled by a PID regulator receiving input signals from a temperature sensor placed in the water bath. This allowed very narrow temperature control (normally better than 0.1°C).

Two six-channel peristaltic pumps maintained separate flows of seawater (SSW-60) from the header tank through each chamber. In the cold tray, the water from the header tank was precooled through coils of 1.77 mm (ID) polyethylene tubes submersed in the tray water. Thus, throughout the experimental period, the chamber water was continuously renewed with a turnover time ranging from 12 to 32 hours, depending on oxygen consumption rates.

A laminar type internal circulation system was maintained by submersed, aquarium pumps driving water through a perforated pipe positioned along one side of the chamber. The timer control of the pumps were set to 15 minutes on and 45 minutes off. The pumps generated characteristic current velocities of $5\text{-}10\text{ cm sec}^{-1}$. No visible resuspension of cuttings or sediments were ever observed to result from the internal circulation system.

2.4 MONITORING OF TEST ENVIRONMENT

Temperatures and oxygen in the SSW-60 source water was regularly measured in the header for determination of oxygen consumption. These measurements are shown in Figure 4. Figure 5 shows temperatures determined in the cold tray outside the chambers, whereas temperatures measured inside the chambers are shown in Figure 6.

During test preparations the temperature in the tray water was set at -0.5°C . When the samples arrived, temperatures were about 3°C and declining (Figure 5). Unfortunately, technical problems with a valve in the glycol circulation system, caused a temporary

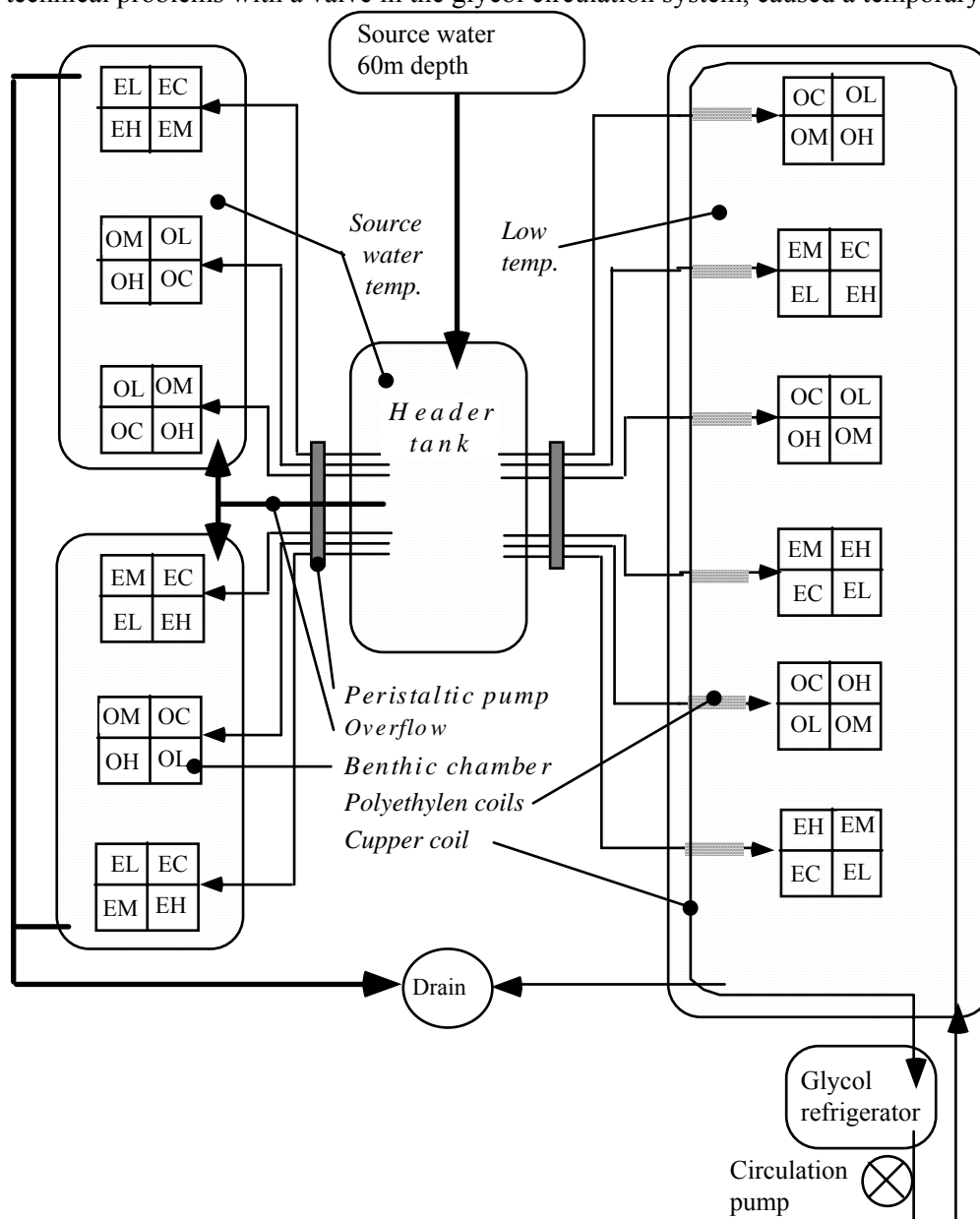


Figure 3. Schematic drawing of experimental set-up. Arrows show the flow of seawater through trays and chambers as well as the separate, closed circulation of glycol. Treatments are shown by the two letters in each section of the experimental chambers: E=Ester, O=Olefin, C=Control, L=Low, M=Medium and H=High dose.

increase of temperatures before the set temperature of -0.5°C was obtained 20 days before addition of the cuttings. The maintenance of this temperature level was controlled daily until the temperature sensor was connected on day 13. Figure 5 shows temperatures between -0.5 and -1.5°C throughout most of the remaining experimental period. A temporary rise of temperatures to $+0.5^{\circ}\text{C}$ occurred after removal of surface insulation material for sampling and inspection on day 35. In the afternoon 29.05.97 the glycol refrigerator failed to auto-restart after an interruption of the current supply. Within 7:00 the next morning, the temperatures in the tray had reached as high as $+2.3^{\circ}\text{C}$ before the refrigerator was restarted manually. Later that day, temperatures up to $+2^{\circ}\text{C}$ were recorded inside the chambers (Figure 6). No

indications were found, and it did not appear likely, that the short exposures to elevated temperatures had had any effects on any other experimental parameters.

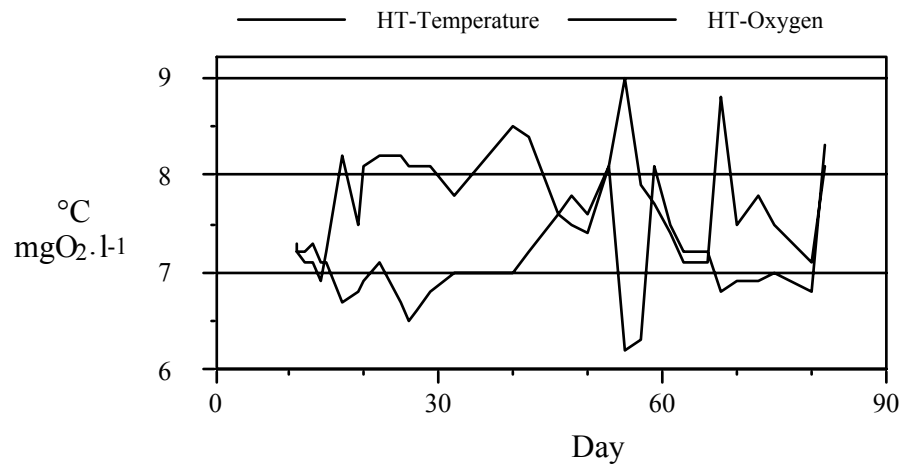


Figure 4. Temperature and oxygen in source water for the experiment (measured in header tank).

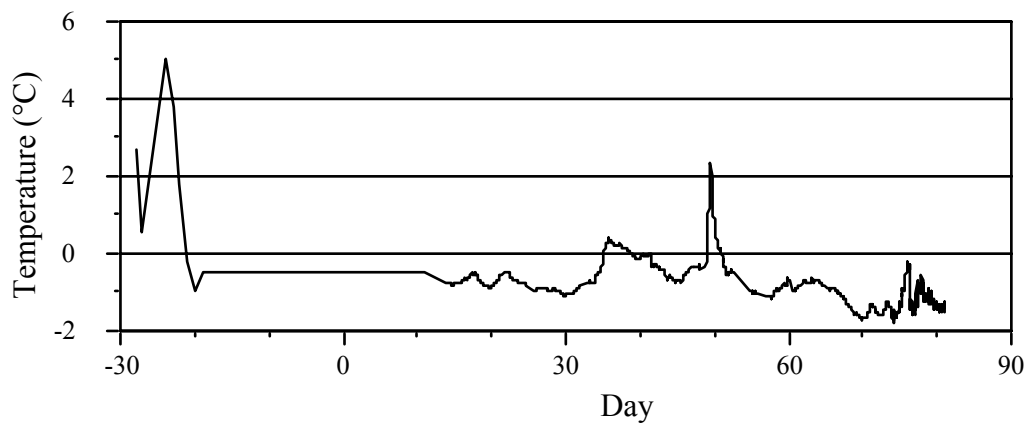


Figure 5. Temperatures in the water bath of the chambers from Porsangen. Accurate measurements were performed daily during the first week after sample arrival. During test preparations and set-up the set level of -0.5°C was regularly controlled. After day 13, temperatures were recorded hourly and stored on a data logger.

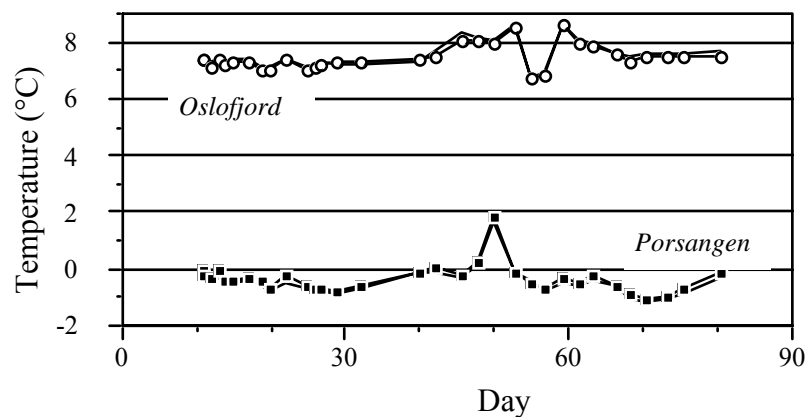


Figure 6. Mean temperatures in experimental chambers from the Oslofjord (upper curve) and Porsangen (lower curve).

However, shortly after this event, natural variations in the fjord caused a temporary drop of temperatures in the source water from 8.1 to 6.2°C (Figure 4) as well as inside the chambers maintained at Oslofjord temperatures (Figure 6). Simultaneously, the concentration of oxygen in the header tank, increased from 7.5 to 9.0 mgO₂l⁻¹. In this experiment, the header tank was too small to damp such fluctuations and because of the time lag between the header tank and the outlet from each chamber such events may lead to an overestimation of the oxygen consumption at the beginning and an underestimation towards the end of the event (ref. ch. 3.2).

2.5 ADDITION OF CUTTINGS

The objectives of the initial treatment was to obtain an evenly distributed layer of cuttings contaminated with three levels of the respective drilling fluids. A medium dose of approximately 5 mgOP cm⁻² corresponded to the level most frequently used in previous tests. In addition, a low dose of 0.5 mgOP cm⁻² and a high dose of approximately 20 mgOP cm⁻² was planned.

Two samples labelled “Nova Tec Mud” and “Eco Green Mud” were received from Anchor/M-I Drilling Fluids A.S. medio march 1997. The respective compositions of the two samples were stated in the lab-report enclosed in APPENDIX 4. Cuttings were prepared in the laboratory at MRS by mixing the mud samples with non-contaminated marine clay sediment which had been dried at 90°C and crushed through a 2x2 mm mesh size sieve. Thorough mixing was done manually using a knife. The laboratory prepared cuttings were then allowed to age for six days in a cool, dark place. On 10.04.97 seawater (SSW-40) was added and two homogeneous slurries were prepared using a steel whirl mixer. Aliquots of the slurries were weighed and added to the experimental surfaces.

The addition was done by dividing each chamber into four 24x24cm sections using 15 cm high, cross frames made up from 5 mm acrylic sheets. The frames were inserted to a sediment depth of 1 cm and the water level in the trays were lowered a few cm below the edge of the chambers. Thus, ca 10 cm high columns of water were enclosed on top of each sediment section. During gentle stirring, the slurries were poured into the enclosed watercolumns. According to the scheme shown in Table 1, the four sections (a, b, c and d) in each chamber (nos. 1-12) were treated with zero, 5, 50 or 200 mg of the slurry containing either olefin- or ester-based muds. The four levels are referred to as control, low, medium and high dose, respectively.

The chambers were then left for particle settling until the next day. Then, the walls were pushed slowly into the sediment to leave only 1-2 cm of the wall above the sediment-water interface and the circulation pumps were initiated. Thus mobile animals had the possibility to move from one section into another.

2.6 SAMPLING PROGRAMME

The sampling programme is shown in Table 1.

2.6.1 Added cuttings

Samples of the suspensions of seawater and cuttings were drawn before, during and after addition to the chambers and analysed for water content, barium and organic phase.

Table 1. Treatment and sampling of test sediments.

Source	Ch.	Sec.	Treatment	Sediment samples						Redox potentials				
				Initial			Final			Initial for profile	Day 35 for profile	Final at 2mm	at 20 mm	for profile
				0-3 cm OP	Ba	Core Ba	0-3 cm OP	Ba	Core Ba					
	1	a	Ester low				1	1		9	3	3	6	
	"	b	" con	1		4		1		9	9	3	3	
	"	c	" high			4	1	1		9	18	3	3	6
	"	d	" med	1	1		1	1			18	3	3	12
	2	a	Olefin med				1	1			9	3	3	
	"	b	" low	1	1		1	1			9	3	3	
O	"	c	" high				1	1		9	9	3	3	
S	"	d	" con				1	1		9	9	3	3	6
L	3	a	Olefin low	1	1		1	1			9	3	3	
O	"	b	" med				1	1			9	3	3	
F	"	c	" con				1	1			9	3	3	
J	"	d	" high				1	1			9	3	3	
O	4	a	Ester med	1	1		1	1				3	3	
R	"	b	" con				1	1				3	3	
D	"	c	" low				1	1				3	3	
E	"	d	" high				1	1				3	3	
N	5	a	Olefin med				1	1				3	3	
	"	b	" con	1				1		9		3	3	
	"	c	" high				1	1		9		3	3	
	"	d	" low	1	1		1	1				3	3	
	6	a	Ester low				1	1			9	3	3	6
	"	b	" con	1				1		9	9	3	3	6
	"	c	" med	1	1		1	1			9	3	3	6
	"	d	" high				1	1		9	18	3	3	
	7	a	Olefin con	1				1			9	3	3	4
	"	b	" low				1	1			9	3	3	
	"	c	" med	1	1		1	1	4		9	3	3	
	"	d	" high				1	1	3		9	3	3	
	8	a	Ester med				1	1				3	3	
	"	b	" con	1				1		9		3	3	3
	"	c	" low	1	1		1	1				3	3	
	"	d	" high				1	1		9		3	3	
	9	a	Olefin con	1				1		9		3	3	
	"	b	" low				1	1		9		3	3	
P	"	c	" high				1	1				3	3	
O	"	d	" med	1	1		1	1				3	3	
R	10	a	Ester med				1	1			9	3	3	
S	"	b	" high				1	1		9	9	3	3	
A	"	c	" con				1	1			9	3	3	4
N	"	d	" low	1	1		1	1			9	3	3	
G	11	a	Olefin con			4	1	1			9	3	3	4
E	"	b	" high			4	1	1			9	3	3	
N	"	c	" low				1	1			9	3	3	
	"	d	" med	1	1		1	1			9	3	3	
	12	a	Ester high				1	1			9		3	
	"	b	" med				1	1			9	2	3	8
	"	c	" con				1	1			9		3	
	"	d	" low	1	1		1	1			9		3	
Total number of analyses:				18	12	16	42	48	7	117	315	134	144	71

2.6.2 Sediment samples

In the present study, in addition to the determination of initial and final concentrations of the organic phase, it was an objective to reveal any cross-contamination between adjacent sections during addition and to determine the extent of downwards mixing of cuttings particles during the experimental period. In order to meet the various objectives a strategic sampling programme was designed for the initial sampling which encountered 18 analyses of the organic phase and 12 analyses of barium in sediment samples drawn on 14.04.97 (Table 1). In addition barium was determined in 16 core samples drawn from control and high dose treatments from both locations. Water content was determined in all samples.

The initial sampling programme allowed determination of the organic phase in all sections, using at least one of the three methods:

1. analyses of organic phase in sediment samples drawn after sedimentation of cuttings,
2. analyses of barium in sediment samples and calculation of the organic phase from the OP:Ba ratio or
3. calculation from the amount of slurry added.

Final sediment samples were collected 3.07.97, 89 days after addition of cuttings. 42 samples including all sections treated with contaminated cuttings and six non-treated control sections, were analysed for barium and organic phase. In addition barium was determined in 7 core samples drawn from two different treatments.

Most of the sediment samples were pooled from three separate cores (ID \approx 15 mm) drawn at random locations within each section. The top 0-3 cm section was cut off and used for analyses of organic phase and barium. For bioturbation assessment, core samples (ID \approx 13 mm) were drawn from a few treatments and sectioned at 1, 3, 5 and 7 cm. Those samples were analysed for barium only.

2.6.3 Redox potentials

Vertical profiles of redox potentials were determined in control and treated sections as shown in Table 1. In general, potentials were recorded at the sediment water interface (0cm) and at 1 cm depth intervals down to 8 cm or down to a depth at which no further change of potential was observed. During the final sampling, potentials were recorded at 0.5 and 20 mm depth at three randomly chosen locations in each section.

2.6.4 Oxygen consumption

Sediment oxygen consumption in each chamber was determined at 2-3 days intervals throughout the experimental period between the initial and final sampling.

2.6.5 Macrofauna

By the end of the experiment, the benthic fauna in each section was collected by washing the sediments on a 1 mm mesh size sieve. The animals from the preserved samples were then sorted into main taxonomic groups and further identified to species level. In a few cases, identification was only done to a higher taxon. Below, those taxons will also be treated and referred to as species. Shannon-Wiener (H) (Shannon & Weaver 1963) and Hurlberts (ES₁₀₀) (Hurlbert 1971) diversity indices were determined.

End-of-experiment faunas in all twelve chambers (48 sections) from the Oslofjord and Porsangen were analysed. No animals were found in sample 3B from the Oslofjord. The reason for this is unknown, but it is unlikely that this in any way could have been related to the treatment (olefin, medium dose). Probably some error has occurred during sampling, conservation or treatment.

2.7 PRIMARY SAMPLE HANDLING AND CALCULATIONS

2.7.1 Sediment samples

The set-up of the experiment implicated that the cuttings and contaminants were present in a thin layer at the sediment-water interface. Thus concentrations measured against sediment mass (wet or dry), will be crucially dependent on the sediment depth at which the core-sample is cut off. This depth may vary intentionally or unintentionally, and concentration units such as $\text{mg}\cdot\text{kg}^{-1}$ dry sediment will vary accordingly. In the present test the half-life and mass balance calculations depend on estimates of the total amount of organic phase present within each chamber or below a given sediment area, and the preferred units were such as $\text{mg}\cdot\text{chamber}^{-1}$ or $\text{mg}\cdot\text{cm}^{-2}$.

Total wet weight of the samples were determined during sampling. In the laboratory, the drilling fluids were extracted from a sub-sample of the wet sediment. The concentration of drilling fluids was calculated from:

Equation 2.1
$$C_a = I_{GC} \cdot M_s / M_{GC} \cdot n \cdot A_{core}$$

in which:

- C_a = concentration of organic phase ($\text{mgOP}\cdot\text{cm}^{-2}$)
- I_{GC} = integrated GC peak area, corrected for reagent blank (mgOP in extract)
- M_{GC} = mass of sub-sample for extraction (g wet sediment)
- M_s = total mass of sediment sample (g wet sediment)
- n = number of cores
- A_{core} = area of each core sample

Following this procedure errors resulting from inaccurate core sectioning and false assumptions of mass-volume ratios in the sediment were eliminated. The sampled area was calculated from accurate measurements of the core diameter using a sliding calliper.

2.7.2 Oxygen consumption

Oxygen consumption was determined by successive measurements of concentration of oxygen in the inlet water in the header tank and in the outlet water in the sampling cell on top of each chamber (Figure 1), using WTW oximeter and electrodes. The flow of water through each separate chamber was measured gravimetrically after collection of outflow water for at least 4 minutes.

Thus, the sediment oxygen consumption SOC, was calculated from the equation:

Equation 2.2
$$\text{SOC} = (C_i - C_o) \cdot F \cdot 10^3 / A \cdot M_{O_2}$$

in which

- SOC is the sediment oxygen consumption ($\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$)
- C_i is the concentration of oxygen in the water entering the chamber ($\text{mg}\cdot\text{kg}^{-1}$)
- C_o is the concentration of oxygen in the water leaving the chamber ($\text{mg}\cdot\text{kg}^{-1}$)

F is the flow of water through the chamber ($\text{kg}\cdot\text{h}^{-1}$)
A is the area of the chamber (m^2)
 M_{O_2} is the molecular weight of oxygen = $32 \text{ g}\cdot\text{mol}^{-1}$

2.7.3 Redox potentials

Redox potentials (E_h) were determined *in situ* by insertion of a Radiometer P101 platinum electrode into the submersed sediments. Potentials were measured against a Ag|AgCl reference electrode hanging in the overlying water. Before each series of measurements, the redox circuit was checked in a ZoBell Fe(II)-Fe(III) redox-buffer solution. The E_h of the samples were obtained by adding the half-cell potential of the reference electrode, to the potential recorded on the Pt-electrode.

A standard deviation of ± 33 mV has been calculated from repeated measures of E_h at a depth 1 cm below the sediment-water interface in similarly treated sediments kept in different chambers. This range may represent the between chamber reproducibility of E_h .

2.8 CHEMICAL CHARACTERISATION AND ANALYSES

2.8.1 Chemical characterisation

Eco Green ester

The Eco Green base fluid (Figure 7) characterised qualitatively in a previous report (Schaanning *et al.*, 1996). Mass spectral identification, partly based on library search, revealed that the components consisted of isopropyl-esters of fatty acids of varying chain lengths and varying degree of unsaturation. It was confirmed that the product consisted of esters produced from naturally derived fatty acids (fish oil). The product was not found to be different from the Ancogreen ester tested in a previous study (Schaanning *et al.*, 1996).

Novatec

The Nova tec olefins (Figure 8) were confirmed to consist of a mixture of C_{14} and C_{16} with a small ($\leq 5\%$) contribution from C_{18} decenes. The base fluid was not found to be different from the Ultidril product first described by Oreld (1995), and used in two previous studies (Schaanning, 1995, Schaanning *et al.*, 1996).

2.8.2 Chemical analyses

Analytical methods

Work-up procedure, sediments

Wet sediment samples weighing 1-5 g were homogenised and placed in a soxhlet tube. Internal standards, 1-dodecen (for *Novatec* samples) and ethyloctanoate (for *Eco Green* samples) were added. Tubes were refluxed with 100 ml methanol for 2.5 hours to remove water. The methanol was decanted and the samples refluxed with 150 ml dichloromethane over night (min. 16 h). The methanol extract was diluted with 100 ml distilled water and extracted twice with 40 ml dichloromethane. The dichloromethane extracts were combined, washed twice with 50 ml water and dried over sodium sulphate for min. 16 h. Finally, the extracts were evaporated to a suitable volume (5-25 ml) and analysed by gas liquid chromatography (GC).

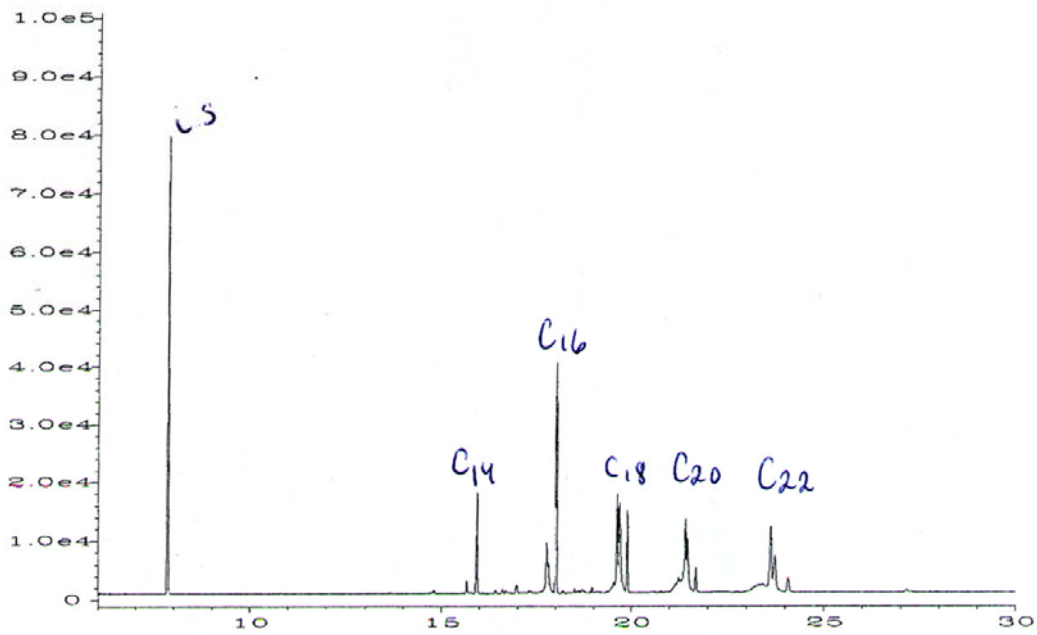


Figure 7 *Eco Green* ester extracted from initial sediment sample. (i.s. = internal standard).

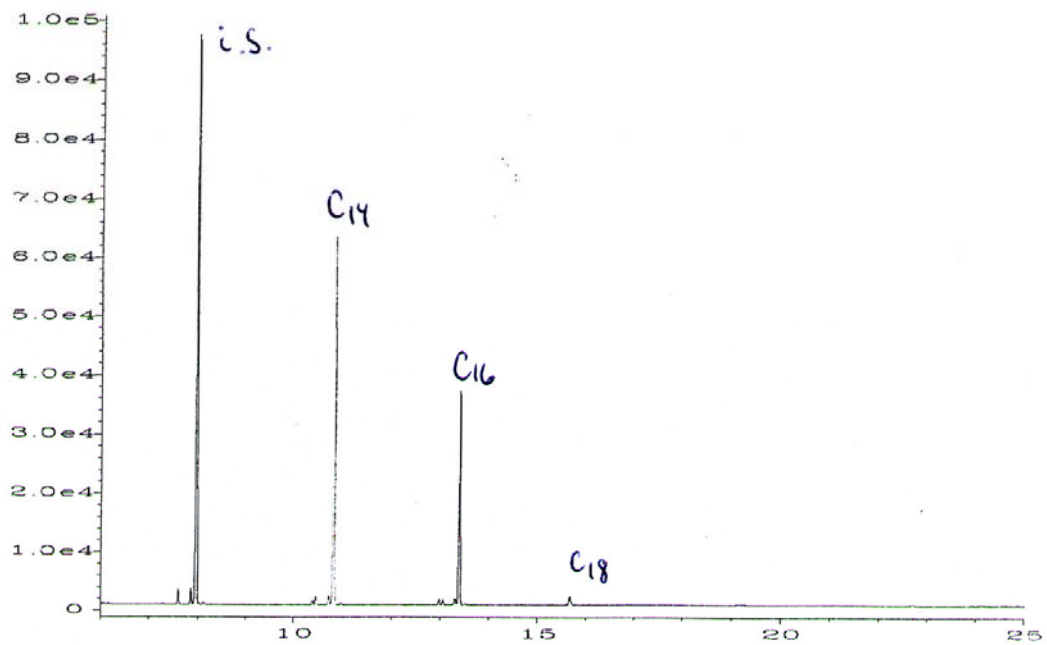


Figure 8 *Nova Tec* olefins extracted from initial sediment sample. (i.s. = internal standard).

Determination by GC-FID

Quantitation of olefin and ester based components was carried out by measuring the flame ionisation detector (FID) response of the area of the components of interest. This area was compared to the corresponding response of known amounts of the internal standards.

Quantitation of SMO was carried out by measuring the area of the flame ionisation detector response of the components. This area was compared to the corresponding response of known amounts of SMO as an external standard.

The gas chromatographic analyses were carried out under the following conditions, identical for all five components:

Gas chromatograph : HP 5890II with autosampler HP 7673
Column : 12.5 m, 0.20 mm i.d., fused silica, cross-linked with dimethyl silicone
Detector : Flame ionisation detector.
Total flow : 5.0 psi
Split : 9.0 ml/min 1:10
Column flow : 0.9 ml/min
Septum purge : 1.5 ml/min
Carrier gas : Hydrogen
Injection vol. : 1 µl
Temperatures:
Column : 50°C, 1 min-10°C/min-250°C, 10 min. total: 31 min.
Detector : 325°C
Injector : 258°C
Data system : HP-Chem station.

Quality assurance

Equipment and reagents

Trace analysis requires control of the background levels of chemicals and equipment.

The following chemicals were used:

- Dichloromethane, Rathburn HPLC-grade.
- Methanol, Merck p.a.
- Deionized water
- Sodium sulphate, Merck for org. trace analysis.

Most of the equipment was rinsed with acetone and heated at 600°C over night. Some equipment, such as soxhlet and graduated flasks were rinsed with dichloromethane three times before use.

The analytical procedure was controlled for possible contamination by analysing procedural blanks.

The instrument was regularly calibrated during the period of analysis using appropriate standards. The validity of the standard curve was verified by analysing control standards. These controls were required to be within +/- 10 % of the expected values for acceptance.

Accuracy

For all components, an internal standard was added to the sediment samples prior to the extraction, in order to compensate for possible losses during the preparation.

The accuracy of the methods has been checked by analysing sediment samples with known amounts of the components. An average recovery of > 95% was obtained after work up and analysis of two replicates pr. component.

Reproducibility

The reproducibility of the analytical procedure has been determined by repeated analyses of samples taken early in the experiment. A relative standard deviation of $\pm 2\%$ was obtained for all five components.

3. RESULTS

3.1 SEDIMENT SAMPLES

3.1.1 Vertical distribution of barium

The vertical distribution of barium in six cores sectioned in 0-1, 1-3, 3-5 and 5-7 cm depth intervals is shown on a logarithmic scale in Figure 9. The two control cores were drawn at the start of the experiment from chambers 1 (Oslofjord) and 11 (Porsangen). Simultaneously, two high dose cores were drawn from the same chambers. By the end of the experimental period, cores were drawn from medium and high dose section in chamber 7 (Porsangen). The figure shows that most of the added barium was recovered within the 0-1cm section. A small fraction was found within the 1-3cm section of the high dose treatments. Below 3 cm, the concentration of barium remained at background level in all samples. Thus, the core samples gave no evidence for any downwards transport of cuttings particles to depths below the 0-3cm sampling depth for the organic phase.

In control sections sampled at the end of the experimental period, the concentration of barium in the Oslofjord sediment (0-3cm) of $0.832 \pm 0.026 \text{ mgBa g}^{-1}$ dry wght. (n=6) was slightly higher than the initial concentration of $0.785 \text{ mgBa g}^{-1}$. Also in Porsangen, the final concentration of $0.693 \pm 0.071 \text{ mgBa/gTS}$ (n=6) in control sections was slightly higher than the initial concentration of 0.635 mgBa/gTS . Some post-depositional cross-contamination was expected to result from the internal water circulation and the possibility of animal migration into control sections. However, relative to the amounts added, the increase observed during the experimental period was negligible. Neither did the absence of any surface enrichment of the initial control samples (as shown by the straight vertical profiles in Figure 9), reveal any need for taking cross-contamination into account in mass balance calculations on the organic phase.

The fact that traces of the organic phase was only found in one of the six control sections ($0.018 \text{ mgOP cm}^{-2}$ in section 8b) confirmed that cross-contamination during test set-up was negligible.

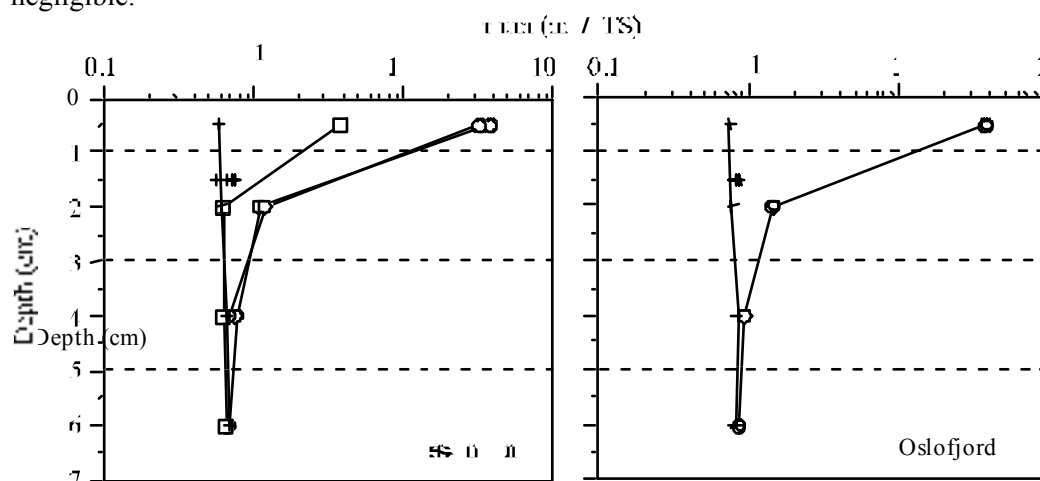


Figure 9. Vertical profiles of barium in core samples of control (crosses) and high dose (circles) sediments taken at the beginning of the experimental period. In the Porsangen sediment, profiles were also determined in one high dose and one medium dose section at the end of the study. Two clusters of six crosses show that the final concentrations in 0-3cm section of control samples had not increased relative to the initial profile.

3.1.2 Disappearance of organic phase

The three subsamples taken of each of the two slurries gave a mean concentration of the organic phase of 108 mgOP g^{-1} (d.wght.) and a standard deviation of 6.7 mgOP g^{-1} , and correspondingly, a concentration \pm one standard deviation of barium of $78 \pm 5.4 \text{ mgBa g}^{-1}$ (n=6) and an OP:Ba ratio of 1.39 ± 0.04 (n=6).

Average aliquots of 6.4g, 50,0g and 200,0g wet slurry was weighed out and added to each of the low, medium and high dose sections, respectively. Multiplying the added aliquots with the 44.4% dry weight content of the slurries and dividing with the section area of 564 cm^2 , the respective loads were calculated as shown in Table 2. Disregarding relatively large standard deviations in the low dose treatments, Table 2 shows that the observed initial concentrations were reasonably consistent with the amounts added. The variation between replicate samples was too large to allow meaningful differentiation, neither between sediments from the two locations nor between sediments treated with the two types of chemicals. Thus initial concentrations were taken to be $0.42 \pm 0.22 \text{ mgOP cm}^{-2}$ in all low dose treatments, and $4.33 \pm 0.69 \text{ mgOP cm}^{-2}$ in all medium dose treatments. In the high dose treatments an initial concentration of $19.0 \pm 2.1 \text{ mgOP cm}^{-2}$ was calculated from the observed barium enrichment in the two core samples (ref. Table 1) and the OP:Ba ratio in the added material.

Table 2. Composition of slurry (mg g^{-1} d.wght.), additions (mg cm^{-2}) and observed initial concentrations (mg cm^{-2}) in replicate sediment samples of the 0-3cm layer. Half of the n number of analyses were performed on olefins, the other half on esters.

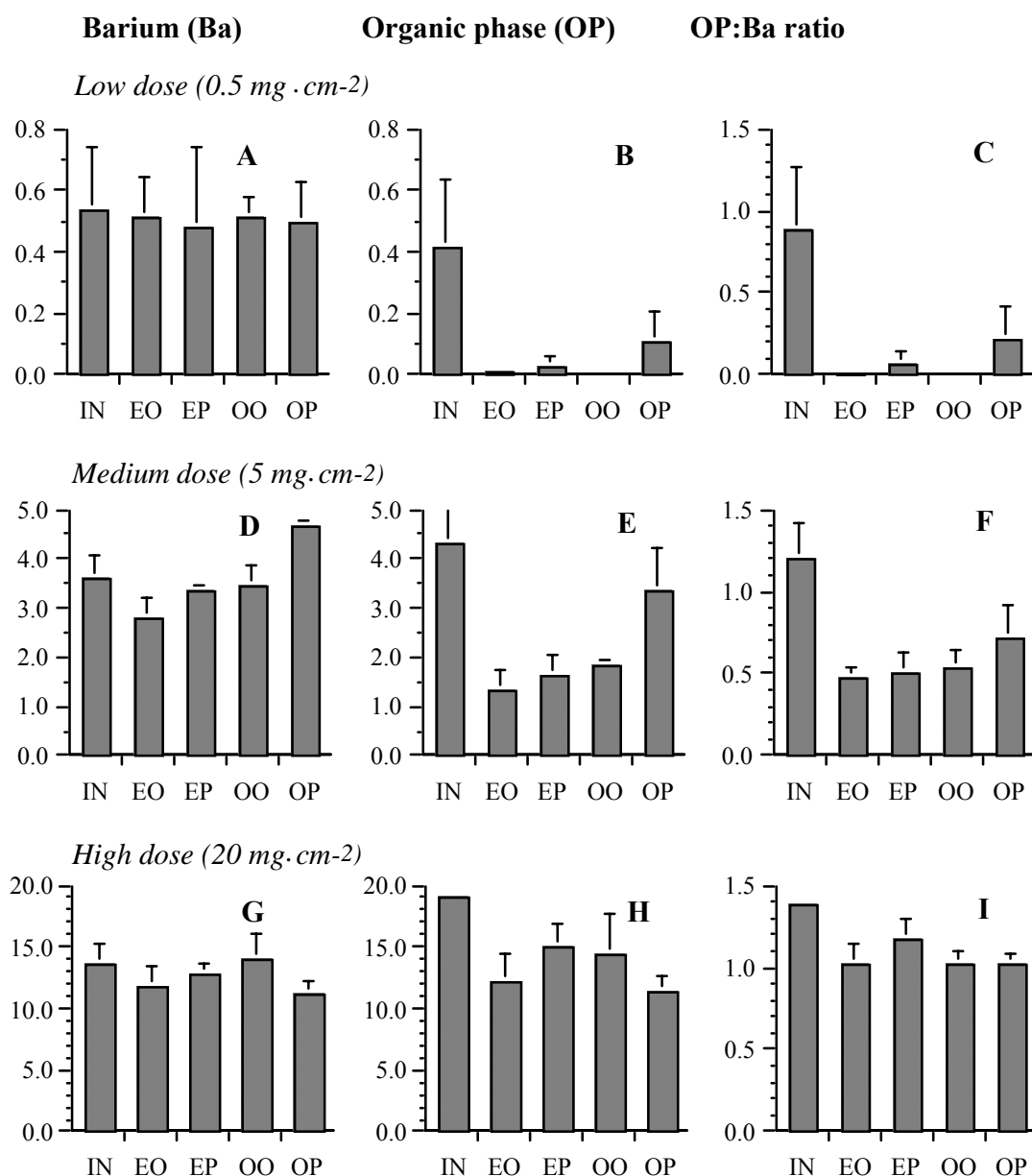
	n	OP \pm std.dev	Ba \pm std.dev.	OP:Ba \pm std.dev.
Slurry (mg g^{-1})	6	108 ± 6.7	78 ± 5.4	1.39 ± 0.04
Added low (6.4g)		0.55 ± 0.09	0.40 ± 0.07	1.39 ± 0.04
Observed low	6	0.42 ± 0.22	0.54 ± 0.20	0.88 ± 0.40
Added med. (50g)		4.25 ± 0.01	3.07 ± 0.01	1.39 ± 0.04
Observed medium	6	4.33 ± 0.69	3.62 ± 0.45	1.21 ± 0.21
Added hi. (200g)		17.0 ± 0.1	12.3 ± 0.1	1.39 ± 0.04
Observed high	2	na	13.7 ± 1.5	-
Estimated high		19.0	-	1.39

Table 3. Mean final concentration and standard deviation of esters and olefins in three replicate sections of sediment transferred from Oslofjord and Porsangen (mgOP cm^{-2}).

	Ester		Olefin	
	Oslofjord	Porsangen	Oslofjord	Porsangen
Low dose (0.5)	$.005 \pm .004$	$.031 \pm .027$	nd	$.111 \pm .096$
Medium dose (5)	$1.34 \pm .41$	$1.66 \pm .38$	$1.83 \pm .13$	$3.35 \pm .87$
High dose (20)	12.2 ± 2.1	15.1 ± 1.8	14.4 ± 3.2	11.5 ± 1.0

Table 4. Fraction (%) of initial concentration of organic phase and OP:Ba ratios remaining in sediment 89 days after addition of cuttings.

	E S T E R				O L E F I N			
	Oslofjord		Porsangen		Oslofjord		Porsangen	
	OP	OP:Ba	OP	OP:Ba	OP	OP:Ba	OP	OP:Ba
Low dose	1	1	7	8	0	0	26	25
Medium dose	31	39	38	41	42	44	77	60
High dose	64	74	79	84	76	74	61	74

**Figure 10. Concentrations of barium (left), organic phase (middle) and OP:Ba ratios (right) in sediments from Porsangen and Oslofjord. The five columns represent (from left to right) initial value (IN), Ester treatment in Oslofjord sediment (EO), and correspondingly Ester Porsangen (EP), Olefin Oslofjord (OO) and Olefin Porsangen (OP). Vertical bars show one standard deviation (n=3).**

Final concentrations of organic phase for each treatment and location are given in Table 3 and relative to initial concentrations in Table 4. In Figure 10, the initial concentrations and ratios from Table 2 are compared to all final observations (Ba, Op and OP:Ba ratios) for each treatment and location.

Figure 10A-C, and Table 3 shows that in low dose treatments, both organic phases were depleted in the final samples of the Oslofjord sediment. In the arctic sediments, 0.03 mg cm^{-2} of the ester and 0.11 mg cm^{-2} of the olefin had remained present (Table 3). These concentrations corresponded to 7% and 26%, respectively, of the initial concentration (Table 4). The maintenance of the concentrations of barium, confirmed that the barite particles were still present in the sediment, but stripped off the organic phases. Apparently, the degradation of both organic phases had been slowed down in the arctic sediments, and olefins to a greater extent than the esters.

In the medium dose treatments (Figure 10D-F) organic phases were present in all samples collected at the end of the experimental period. Concentrations had, however, clearly declined leaving 1.34 mg cm^{-2} (31%) of the ester and 1.83 mg cm^{-2} (42%) of the olefin in Oslofjord sediment and 1.66 mg cm^{-2} (38%) of the ester and 3.35 mg cm^{-2} (77%) of the olefins in the arctic sediment. The high concentration of olefins remaining in the arctic sediment was modified to some extent by the high barium content in the samples, yielding a reduction of the OP:Ba ratio to 60% rather than 77%. Nevertheless, as in the low dose treatments, the disappearance of both organic phases had been slowed down in the arctic sediments, and olefins to a greater extent than the esters.

Also in the high dose treatments, the final samples had clearly lower concentrations of esters, olefins and OP:Ba ratios as compared to the initial samples (Figure 10H,I). For the esters, final concentrations of 12.2 mg cm^{-2} in the Oslofjord sediments and 15.1 mg cm^{-2} in the arctic sediments corresponded to remaining concentrations of 64% and 79% respectively. Thus, for the ester, data were consistent showing a clear decrease of concentrations as well as ester:Ba ratios, in all sediments. The disappearance was slightly less in the arctic sediments as compared to the Oslofjord sediment. The relative disappearance decreased, however, strongly with increasing dose.

In the high dose olefin treatments, the final concentrations of $11.5 \text{ mg OP cm}^{-2}$ in the arctic were lower than the concentrations of $14.4 \text{ mg OP cm}^{-2}$ observed in the Oslofjord sediment. Considering the OP:Ba ratios final values were identical in the two environments (Table 4). This result was different from the results obtained in all ester treatments as well as the low and medium dose olefin treatment.

3.1.3 Regression analyses

In previous studies, first order kinetics have been found most appropriate for the description of the loss of drilling fluids with time. The general form of a first order reaction is:

$$\text{Equation 3.1} \quad C = C_0 \cdot 10^{-kt}$$

in which:

C = concentration at time t

C_0 = initial concentration

t = time

k = rate constant

from Equation 3.1 it can be shown that if the half-life, τ , is the time at which $C = C_0/2$, then $\tau = 0.302/k$.

Exponential regression analyses were performed on initial and final OP:Ba concentration ratios in medium and high dose treatments. In Table 5, the results are compared to the results of previous regression analyses performed on more suitable data with monthly sampling over a six months experimental period with medium dose treatments of Oslofjord communities from the same location (Schaanning *et al.*, 1996).

The table shows that the short half-life previously determined for the ester was not confirmed in the present analyses. This may, however, be primarily attributed to the lack of proper time series in the present experiment giving little credit to the rapid initial disappearance of the ester. The medium dose olefin treatment in the Oslofjord sediment yielded, however, half-life and confidence interval reasonably consistent with the results of the previous study. The data also shows, very clearly, the strong dependence of half-life on dose level. Else, the variations between treatments shown in final concentrations of the organic phase is also reflected in the half-lives given in Table 5.

Table 5. Comparison of exponential regression analyses performed on OP:Ba ratios in a previous study with proper 0-6 months time series with corresponding analyses on initial and final (day 89) OP:Ba ratios in the medium and high dose treatments of the present study. Columns show initial dose, correlation coefficients (r^2), probabilities of no decrease (p), half-life (τ) and the corresponding 95% confidence interval.

Treatment	Initial conc. mgOP cm ⁻²	Corr. r^2	Prob. p	Half-life τ	95% conf. limits	
					lower τ	upper τ
<i>Previous study</i>						
Ester	4.6	0.957	0.0001	18	15	22
Olefin	2.6	0.930	0.0001	56	45	76
<i>This study</i>						
Ester Oslofjord	4.3	0.974	0.000	57	46	73
“ Porsangen	“	0.915	0.003	58	41	101
“ Oslofjord	19	0.849	0.009	198	125	478
“ Porsangen	“	0.667	0.047	340	172	16906
Olefin Oslofjord	4.3	0.949	0.001	65	49	96
“ Porsangen	“	0.799	0.016	92	54	301
“ Oslofjord	19	0.904	0.004	206	142	376
“ Porsangen	“	0.946	0.001	205	154	307

3.2 OXYGEN CONSUMPTION

All results on sediment oxygen consumption (SOC) are shown in Table 6 and Figure 11 - Figure 14. As shown, most clearly in Figure 14, more oxygen had been consumed in Oslofjord than in Porsangen chambers, but hardly any difference was found between ester and olefin treatments.

3.2.1 Fluctuations

Throughout most of the experimental period, the rates fluctuated quite strongly between

Table 6. Sediment oxygen consumption rates (minimum, 25% quartile, median, 75% quartile and maximum) and total oxygen consumption for the 89 days experimental period in each chamber and averaged over similarly treated chambers.

Treatment	Ch.	min	25%	median $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$	75%	max.	Total $\text{mmol}\cdot\text{m}^{-2}$
O	ester	1	69	332	556	708	1255
S	“	4	125	474	625	783	1275
L	“	6	79	560	709	839	1303
O	“	all	69	473	622	800	1303
F							
J	olefin	2	293	417	600	710	1143
O	“	3	139	314	473	533	748
R	“	5	274	554	769	931	1189
D	“	all	139	414	564	736	1189
P	ester	8	152	351	481	607	1005
O	“	10	145	314	518	633	1587
R	“	12	192	239	334	481	958
S	“	all	145	311	453	574	1190
A							
N	olefin	7	165	336	420	517	929
G	“	9	179	346	470	681	976
E	“	11	66	334	431	485	1186
N	“	all	66	337	444	533	1186

300 and 600 $\mu\text{molO}_2\cdot\text{m}^{-2}\cdot\text{h}^{-1}$ in the Porsangen chambers and slightly higher, between 400 and 800 $\mu\text{molO}_2\cdot\text{m}^{-2}\cdot\text{h}^{-1}$ in the Oslofjord chambers (Figure 11, Figure 12, and quartiles in Table 6). Because of the long ($\geq 24\text{h}$) residence times of the chamber water, oxygen consumption measured during periods with increasing concentration of oxygen in the supply water will tend to be overestimated, and oxygen consumption measured during periods with decreasing concentration of oxygen will tend to be underestimated. Probably, the fluctuations result primarily from small fluctuations in the concentration of oxygen in the source water. Averaged over longer periods, the concentration of oxygen in the source water will be constant, and temporary errors in SOC-rates resulting from increasing concentration of oxygen will compensate for the errors resulting from decreasing concentrations.

However, successive events in the uptake of oxygen in the sediments may also contribute to some of the fluctuations. In previous studies, two successive peaks in SOC rates have frequently been observed to occur 1-3 months after addition of medium dose easily degradable chemicals, somewhat later in higher dosed chambers. In this experiment, the SOC was an integrated measure of four differently dosed sediment sections. Therefore, successive events in low and medium dose sections may have contributed to some of the variations, but masked by the general fluctuations.

A major fluctuation was observed between day 40 and day 70 (Figure 11 and Figure 12). In several chambers oxygen consumption increased to maximum rates on day 55. The maxima were succeeded by a very consistent minimum on day 66. This major fluctuation was preceded by an increase of the concentrations of oxygen in the header tank, culminating at 9.0 $\text{mgO}_2\cdot\text{l}^{-1}$ on day 55, followed by a decrease culminating in a minimum of 7.1 $\text{mgO}_2\cdot\text{l}^{-1}$ on day 66 (Figure 4). Probably then, some of the fluctuation was an artefact of natural variations in source water quality. Because of the short duration of the peaks in the Porsangen chambers, a major influence from the hydrographic event cannot be ruled out. The broader peaks in the Oslofjord chambers, showed a persistent increase of sediment metabolism and the timing of the event fitted well with peaks observed in previous experiments. Assuming that the peaks

represented the first culmination of microbial succession in the high dose sections and that the second culmination was denied by experiment termination, the present observations would be consistent with previously observed trends of oxygen consumption rates for LAO and FOE.

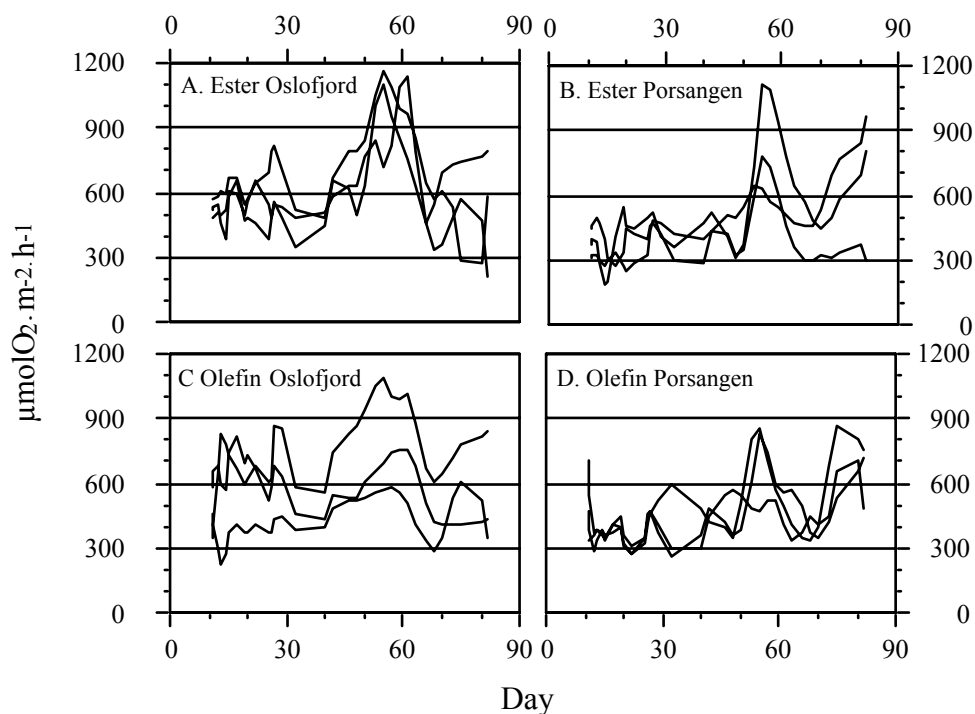


Figure 11. Rates of sediment oxygen consumption in each chamber treated with esters (A and B) and olefins (C and D). The data were smoothed using a three point binomial function.

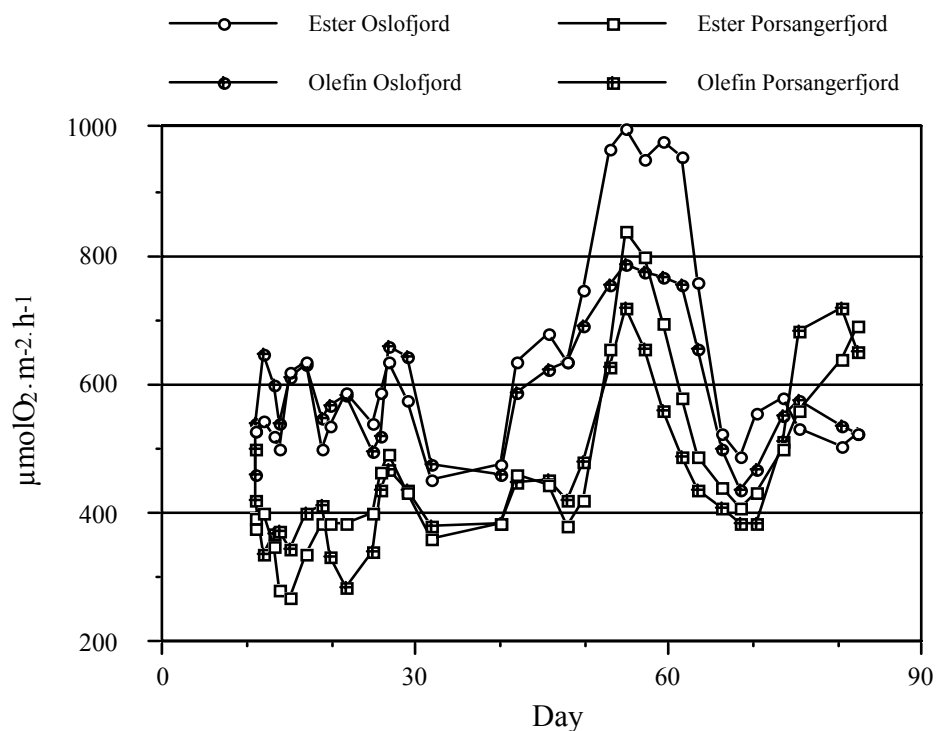


Figure 12. Rates of sediment oxygen consumption in each treatment (mean of three chambers, no smoothing).

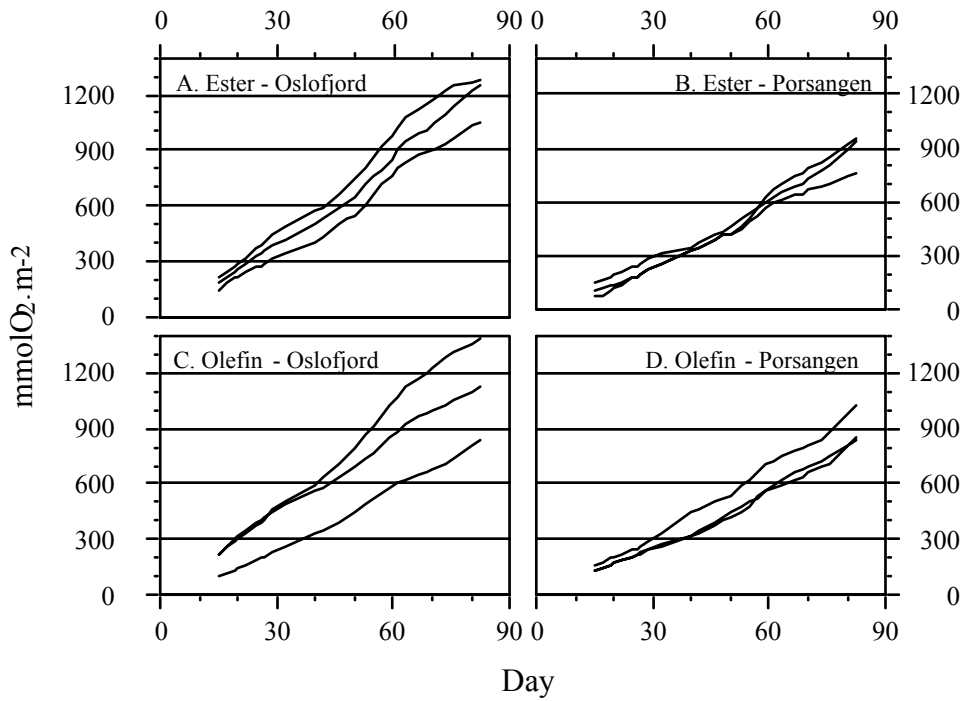


Figure 13. Cumulative sediment oxygen consumption in each chamber treated with esters (A and B) and olefins (C and D).

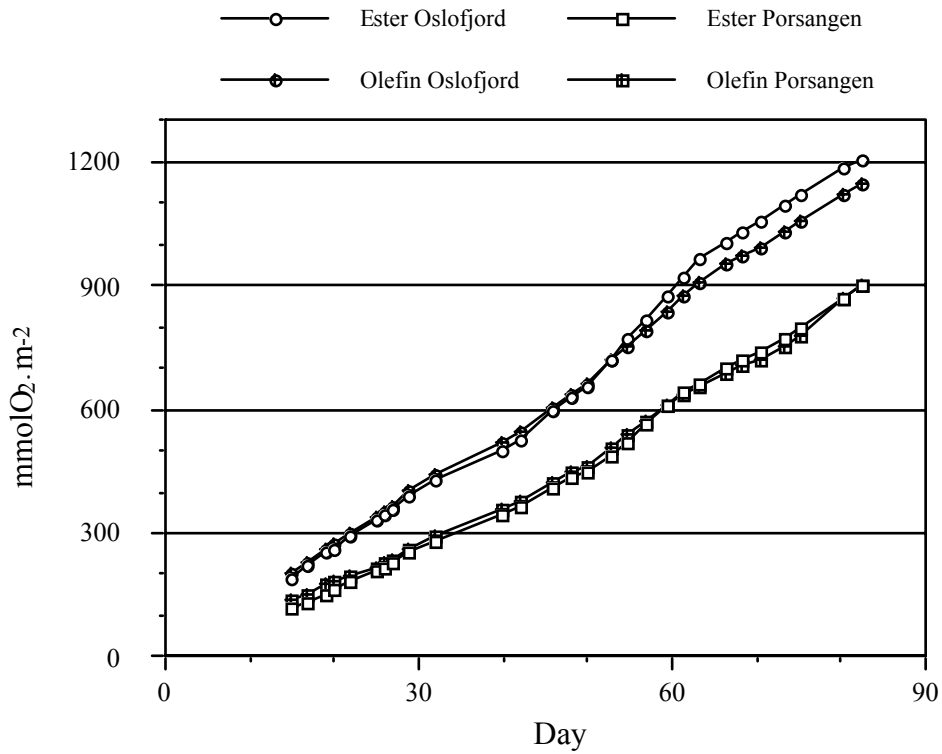


Figure 14. Cumulative sediment oxygen consumption in each treatment (mean of three chambers).

3.2.2 Trends

At the end of the experiment 60-80% of the added chemicals were still present in the high dose sections (Table 3). This showed that even though most of the material may have been degraded in the low dose sections, the degradation event was still in an early stage in the high dose sections which accounted for 78% of the chemicals added to each chamber. It follows that most of the oxygen consumed in the present experiment represent an early phase of the degradation event in the high dose sections. Thus, the lower rates of oxygen consumption observed in the Porsangen as compared to the Oslofjord sediment (Figure 14) might be an initial feature. As shown in Figure 12, SOC rates in the Porsangen chambers increased from initial values $200 \mu\text{molO}_2\text{m}^{-2}\text{h}^{-1}$ lower than the rates in the Oslofjord chambers, to similar and even higher rates towards the end of the experimental period. This gradual increase and eventual cross-over indicated that the degrader communities in the Porsangen chambers adapted to the material added on day zero.

3.2.3 Dose response relationship

Averaged over the entire sediment surface, the present load corresponded to 5.5 mgOP cm^{-2} . This was similar to the load of $2.5\text{-}5.0 \text{ mgOP cm}^{-2}$ added to olefin and ester treated chambers in the previous experiment. However, in the previous test the dose was evenly distributed over the sediment surface providing whereas in the present test oxygen consumption was probably dominated by the processes in the high dose sections in which the layer thickness and total load was approximately 4x higher than in the previous test. As shown in Bakke *et al.*, 1989b, the increase of oxygen consumption with increasing dose of mineral oil reached a limit at high dose treatments. If a non-linear relationship exists also for the present chemicals, the oxygen consumed in the present experiment should be smaller than the oxygen consumed in the previous test.

By the termination of the present experiment, total sediment oxygen consumption in Oslofjord chambers was close to $1200 \text{ mmolO}_2\text{m}^{-2}$ (Figure 14). In previous experiments (Schaanning, 1995, Schaanning *et al.*, 1996) oxygen consumption during the 0-3 months time interval was $1700\text{-}2000 \text{ mmolO}_2\text{m}^{-2}$ for the $^{14}\text{C}\text{-}^{16}\text{C}$ olefins and $1900\text{-}2400 \text{ mmolO}_2\text{m}^{-2}$ for the fish oil esters. Clearly, in the present test on Oslofjord sediment, less oxygen had been consumed than in previous tests confirming that the non-linear relationship found for mineral oil also applies for the present chemicals. Thus, the data indicated that biodegradation will be significantly delayed or slowed down when the load increases from ca 5 to ca 20 mg cm^{-2} , corresponding to an increase of nominal thickness of the cuttings layer from 1 to 4 mm.

3.3 REDOX POTENTIALS

Vertical profiles of redox potentials measured *in situ* in selected chambers and treatments 5, 35 and 92 days after addition of cuttings are shown in Figure 15 and Figure 17. During final sampling, three replicate measurements were taken at 0.5 and 2 cm depth in each section. In general, the data from 2cm depth confirmed the potentials recorded at 0.5cm, but in Porsangen sediments, the 2cm data were more variable because of the RPD transition (see below) at this depth. Thus, only the 0.5 cm data (Figure 16) will be considered below. All measurements are given in Appendix 5 and details on experimental design and statistical methods using the redox data set as example, are given in Appendix 1.

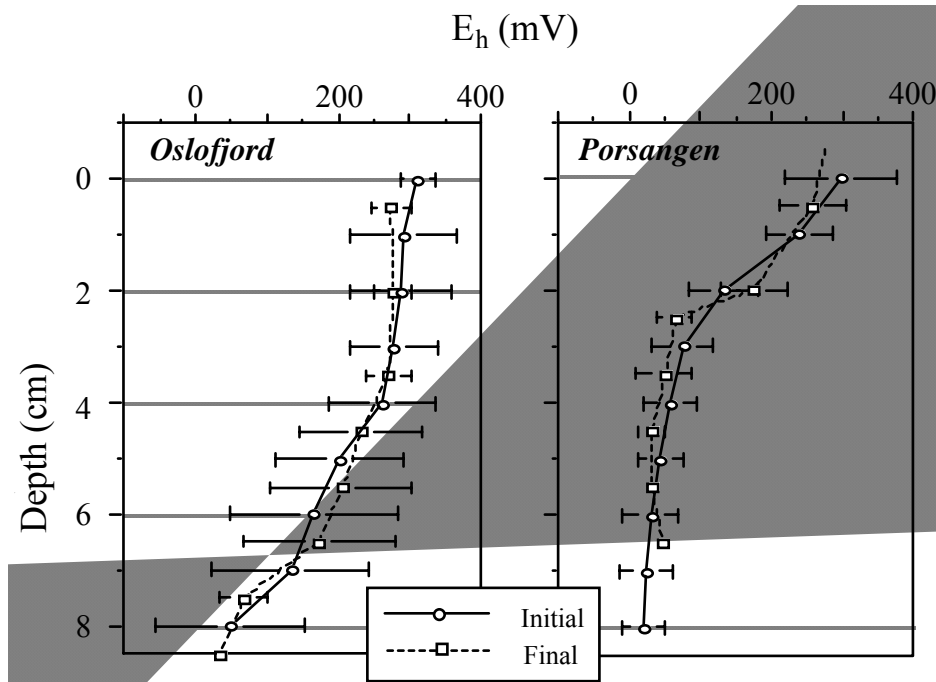


Figure 15. Vertical profiles of redox potentials in control sediment 5 days (initial) and 92 days (final) after addition of cuttings.

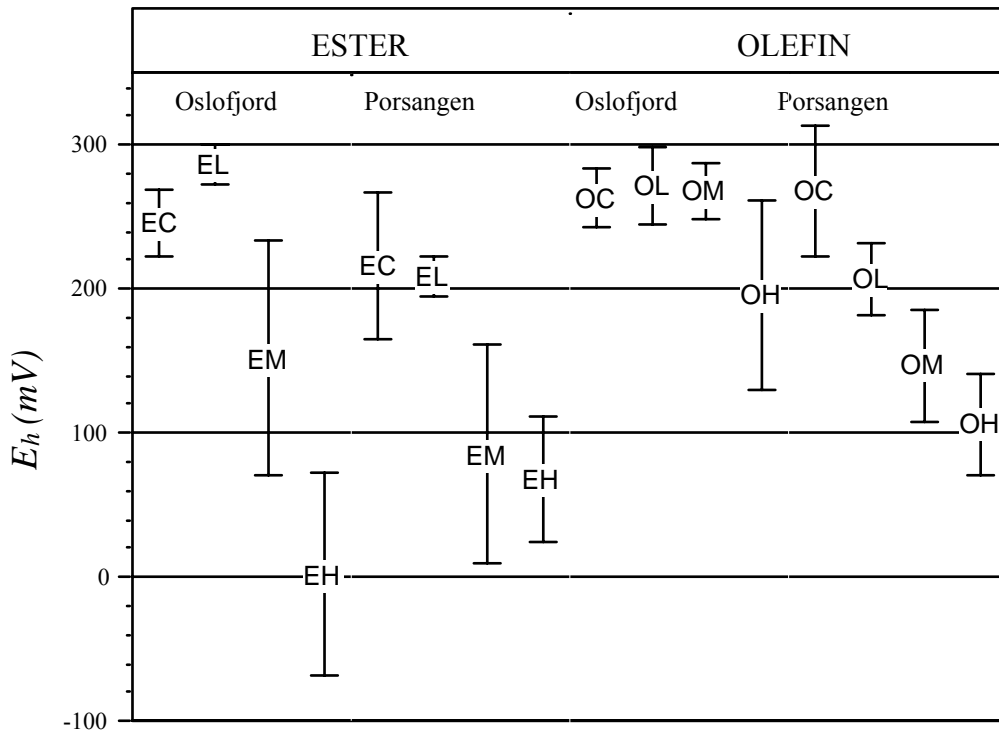


Figure 16. Mean E_h and standard deviation ($n=9$) in each treatment at the end of the experimental period. E=ester, O= Olefin, C=Control, L=Low, M=Medium and H=High.

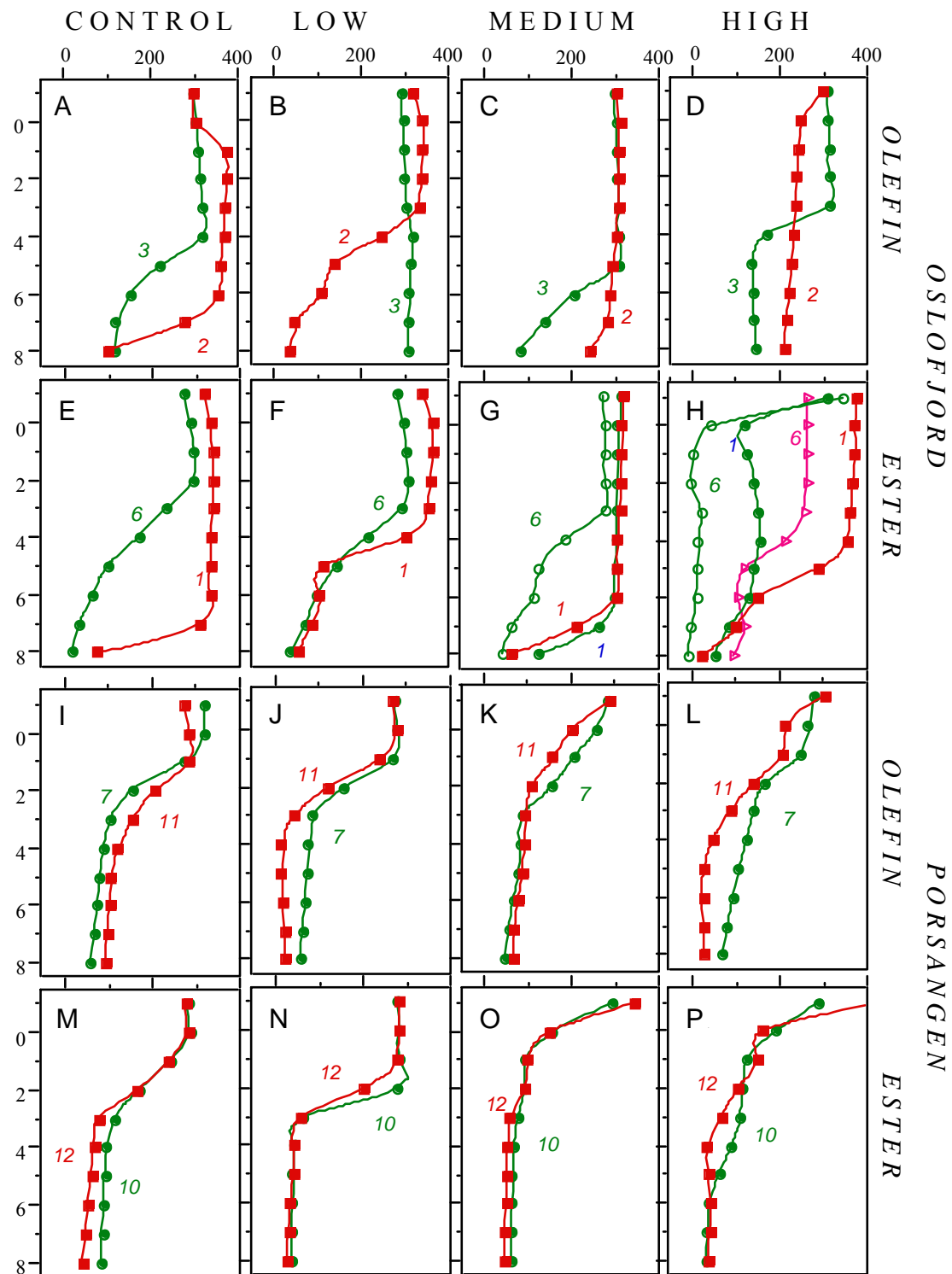


Figure 17. Variation of E_h (x-axis, mV) with depth (y-axis, cm) in all treatments, 35 days after addition of cuttings. Numbers show the chamber in which the profile was measured. Plate H shows potentials measured below normally coloured areas of the sediment surface (squares and triangles) and below white spots (circles), respectively.

3.3.1 Vertical profiles

The vertical profiles frequently revealed a Redox Potential Discontinuity (RPD) layer, within which the E_h decreased from a characteristic near-surface value of 300 mV to a characteristic deeper value of 100 mV. The surface values were consistent with potentials controlled by the presence of O_2 or electroactive redox-couples in equilibrium with oxygen (Bågander and Niemistö, 1978), whereas the sub-surface values were consistent with redox couples involving ammonia or reduced forms of iron and manganese. Potentials $\leq -0.1V$, which are characteristic for the presence of hydrogen sulphide (Bågander and Niemistö, 1978) were never observed in any control section. In the Porsangen sediment, the discontinuity was consistently present at about 2 cm depth (Figure 15 and Figure 17 I-P). In the Oslofjord sediment, the RPD occurred at various depths, was less sharp and occasionally absent within the sampled 0-8 cm layer (Figure 15 and Figure 17 A-H). In control sediments from both locations, the major features of the redox profiles were maintained throughout the experimental period (Figure 15).

On day 35, a decrease of the E_h had occurred within the top layer of medium and high dose sections in Porsangen sediments (Figure 17K,L,O,P). The decrease in the ester treatments (Figure 17O,P) was larger than the decrease in the olefin treatments (Figure 17K,L). No clear effects were found in Oslofjord sediments treated with olefins (Figure 17B-D). In the ester treatments, however, white spots had appeared on the sediment surface in the high dose treatments. In two chambers (nos. 1 and 6), profiles were taken in the middle of such spots (Figure 17H, circles) as well as in areas where no spots were observed (Figure 17H, squares and triangles). Obviously, the potentials measured beneath the spots had been lowered, whereas no clear effects were observed beneath areas where no visible change had occurred. During measurements, fragments of a biofilm in the spot area attached to the sensor surface and were carried downwards on the tip of the electrode. Therefore, the potentials recorded 2-3 cm and more below such biofilms, may be severely underestimated. Conclusively, redox effects were clearly observed below bacteria mats in ester high dose treatments of the Oslofjord sediments on day 35, but the effects may have been restricted to a more shallow depth interval than those indicated in Figure 17H.

3.3.2 Trends

In Figure 18, initial and final redox potentials of the present experiment are compared with the six months trends observed in the previous experiment. The figure shows that after three months, the potentials in the medium dosed chambers of the present experiment were higher than the potentials observed in similarly dosed sediments of the previous experiment. In the high dose sections, the potentials were very similar to those observed in the medium dosed sediments of the previous test. It appears, therefore, that the redox-lowering in the present experiment lags behind the lowering observed previously. This was most likely due to inherent differences in the source sediments. Thus the sediments used for the previous test were collected in May 1995, shortly after the normal sedimentation of the phytoplankton spring bloom adding to the detritus from an extraordinary large flooding of the rivers entering the Oslofjord area. Therefore, the sediments in the first test may have been more enriched

with labile organic carbon and the processes balancing the redox-potentials in the surface layer may have been more easily over-run by the addition of cuttings material.

3.3.3 Final potentials at 0.5 cm depth

During final sampling, the redox potentials at 0.5 cm depth in the Oslofjord sediments were clearly lowered in medium and high dose ester treatments and in high dose olefin treatments. In the Porsangen sediment, both chemicals had clear effects in medium and high dose treatments. Really low potentials showing the presence of hydrogen sulphide were, however, only observed in ester treatments.

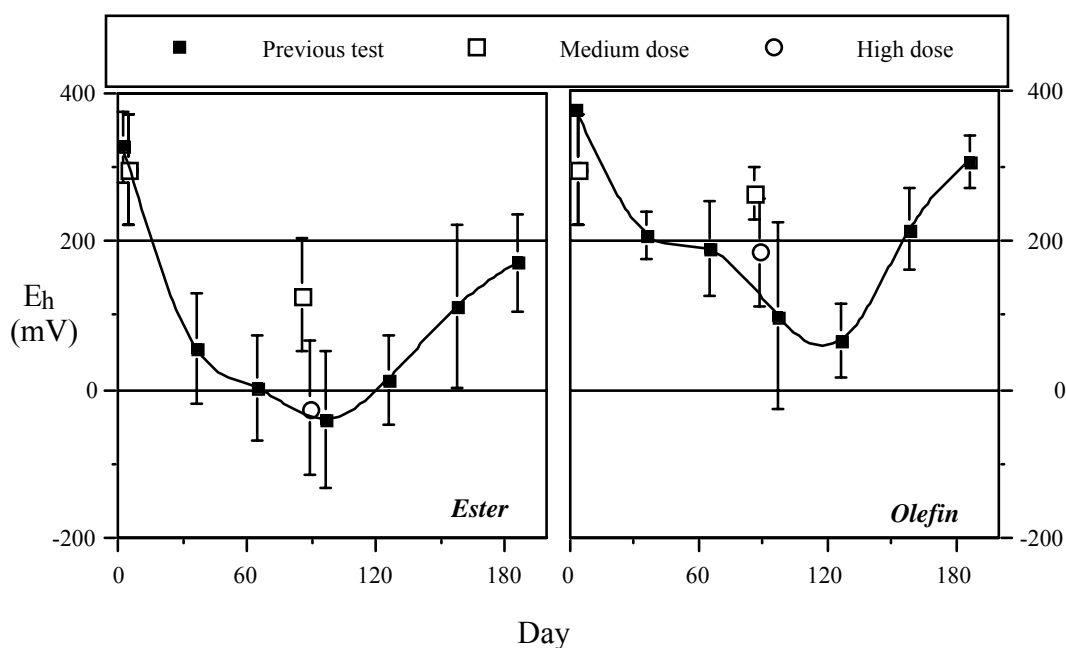


Figure 18. Initial and final redox potentials at 2 cm depth in medium and high dose sections of the present experiment (Oslofjord sediments), compared to the development of redox potentials at 1.5cm depth in sediments treated with medium doses of the same ester and a similar olefin. Vertical bar = \pm one standard deviation.

3.4 MACROFAUNA

The results are shown in Table 7-Table 8 and Figure 19-Figure 22.

The plot of number of individuals (N) vs. number of species (S) revealed large density variations in the Oslofjord fauna in control as well as in treated sections (Figure 19A,B). Roughly, low N corresponded with low S and high N corresponded with high S. The diversities plotted versus the redox potential at 0.5 cm depth (Figure 20) ranged between 3 and 4. According to Norwegian classification criteria for natural fjord locations (Molvær *et al.*, 1997), this was consistent with a class II (“good”) environment. However, the redox potentials were low in several sections, particularly in medium and high dose ester treatments and white bacteria mats had colonised parts of the sediment surface. A time-lag would be expected between the effects on redox potentials from oxygen depletion and bacterial production of hydrogen sulphide and manifestation of sulphide toxicity effects in the macrofauna community structure. In a previous experiment (Schaanning *et al.*, 1996), redox effects were observed three months after addition of cuttings and reduced diversities were found in the corresponding communities sampled after six months exposure. Considering also the large fractions of chemicals remaining for degradation in the medium and high dose sections (Table 4), effects on diversities would most likely have occurred if longer exposure had been allowed.

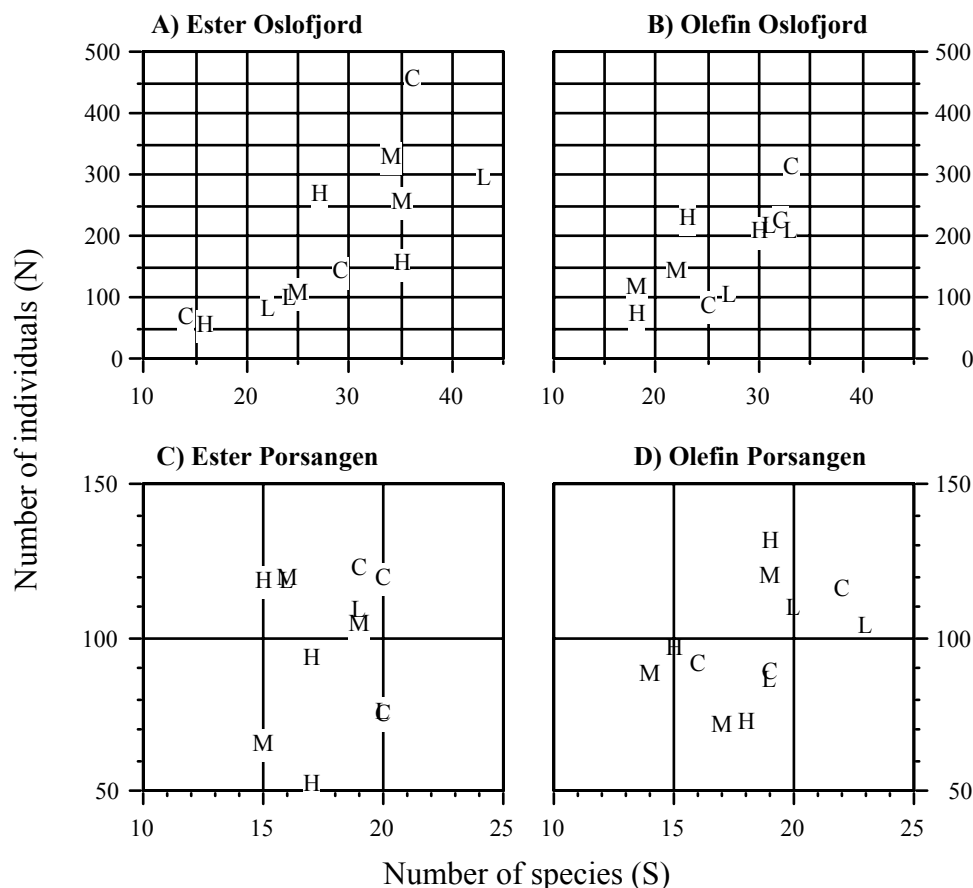


Figure 19. Plot of macrofauna Number of individuals vs. number of species in each chamber after 3 months exposure to various levels of olefin and ester based cuttings. C = control, L = low dose, M = medium dose, H = high dose.

Compared to the Oslofjord communities, the Porsangen communities were less variable and counted fewer species and individuals (Figure 19C,D). High numbers of the polychaete *Spiochaetopterus typicus* dominated in all samples from Porsangen and the diversities were relatively low ranging 2,5-3,5 in control samples (Figure 20C,D and Table 8). Preliminary analyses of communities sampled on arrival at MRS, two days after the survey in Porsangen, revealed diversities similar to those observed by the end of the experimental period. The samples were collected in a pristine area, remote from any evident anthropogenic influence and the diversities were not beyond the expected natural variation. Thus, the lower diversities in Porsangen sediments were considered a result of natural, biogeographical variation, not fit for the standard classification system derived from empirical data on more southern locations.

A rather dramatic event was observed in one of the Porsangen chambers treated with esters. In this chamber (no 12) the colour of the sediment surface in the medium and high dose sections had changed from grey-olive to dark grey-black and all tracks after the star-fish *Ctenodiscus crispatus* had disappeared. Several individuals of the star-fish had left their normal, buried position and were lying on top of the sediment surface with mouth and gills faced upwards into the water column. Low redox potentials confirmed the smell of H₂S in the pore water. In the overlying water, however, an oxygen concentration of 7.2 mg/l was measured to confirm that the continuous supply of water had not been disturbed.

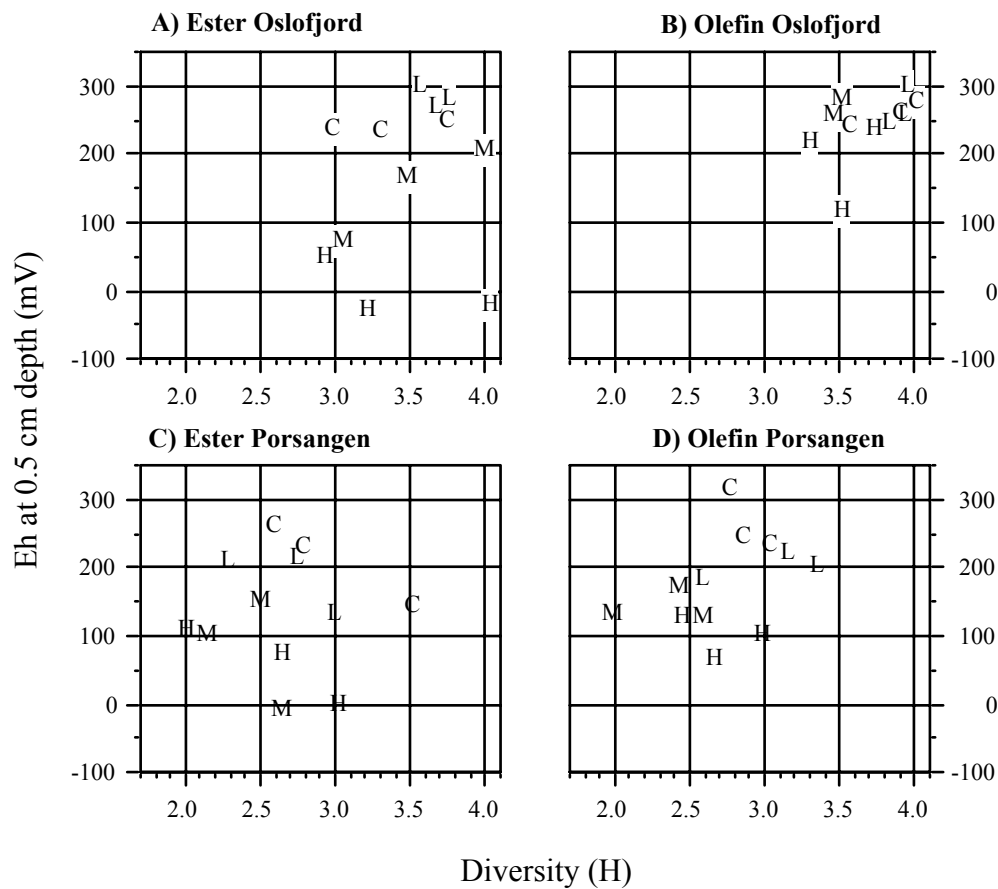


Figure 20. Plot of macrofauna diversity (H) vs. E_h at 0.5 cm depth in each chamber after 3 months exposure to various levels of olefin and ester based cuttings. C = control, L = low dose, M = medium dose, H = high dose.

Obviously, the anomalous behaviour of the star-fish was an emergency reaction to avoid toxic H_2S in the pore water and to obtain oxygen directly from the chamber water. The animals were all alive during sampling and registered as part of the benthic community.

In the MDS plots (Figure 21, Figure 22) the dissimilarities between the fauna types are shown as distances between the plots of the chamber codes. In the Oslofjord samples (Figure 21), differently treated sections from the same chamber tended to be closer to each other (more similar) than similarly treated sections from different chambers. Thus, the natural variation between each box core sample seemed to be larger than the variation produced by experimental treatments. Among the Porsangen samples (Figure 22), no clear similarity patterns could be detected neither by chambers nor by treatments.

The plot of diversity vs. E_h (Figure 20C,D) revealed a separation between control and low

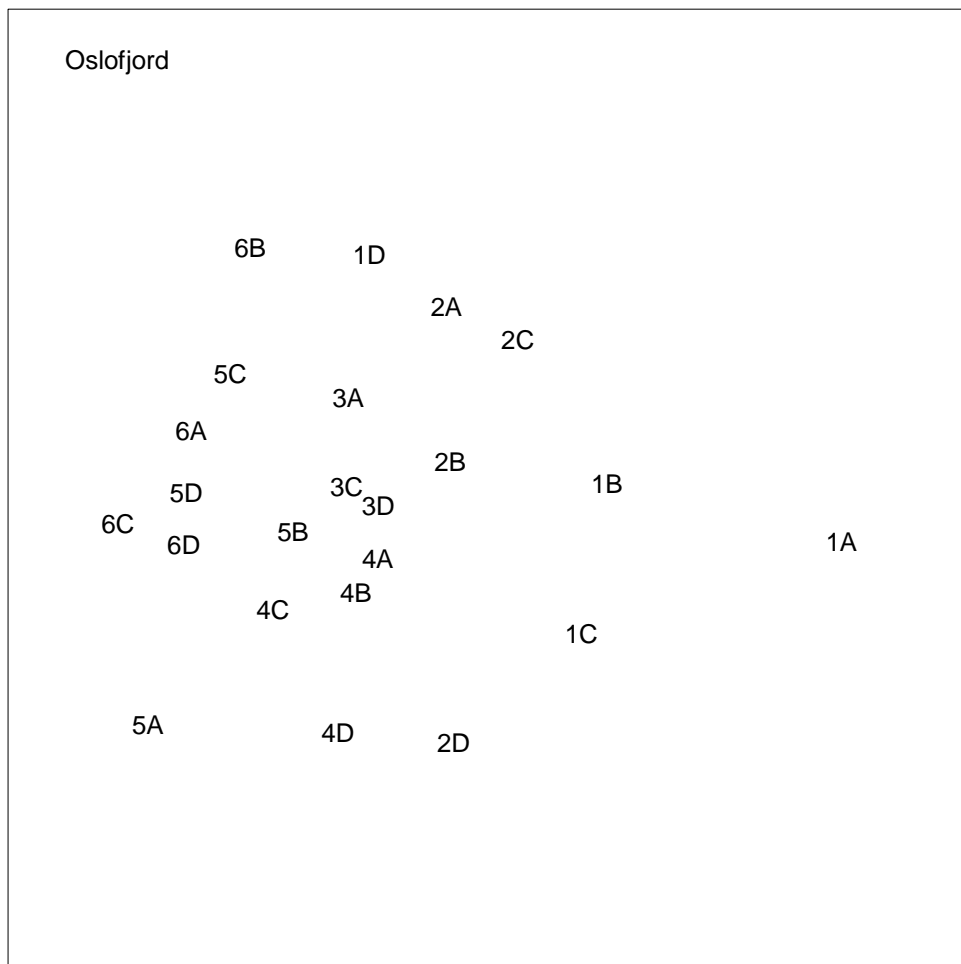


Figure 21. MDS-plot of fauna dissimilarity between the samples from Oslofjorden (the figures and letters are box and quadrant numbers respectively)

dose treatments to the upper right and medium and high dose treatments to the lower left. Thus, at medium and high dose levels, both olefin and ester based cuttings had clear effects on, at least, some of the communities from Porsangen. This tend to suggest that the arctic communities were more sensitive to the present contamination than were the Oslofjord communities. It may be assumed that the Arctic communities would have a lower background concentration of hydrocarbons than the Oslofjord samples (c.f. Bakke *et al.*, 1989, Schaanning *et al.*, 1996), which thus by adaptation might be less sensitive to cuttings treatments. The difference is, however, more likely a result of redox-change and may depend primarily on time of exposure, i.e. the arctic communities were more rapidly affected by the cuttings than were the Oslofjord communities.

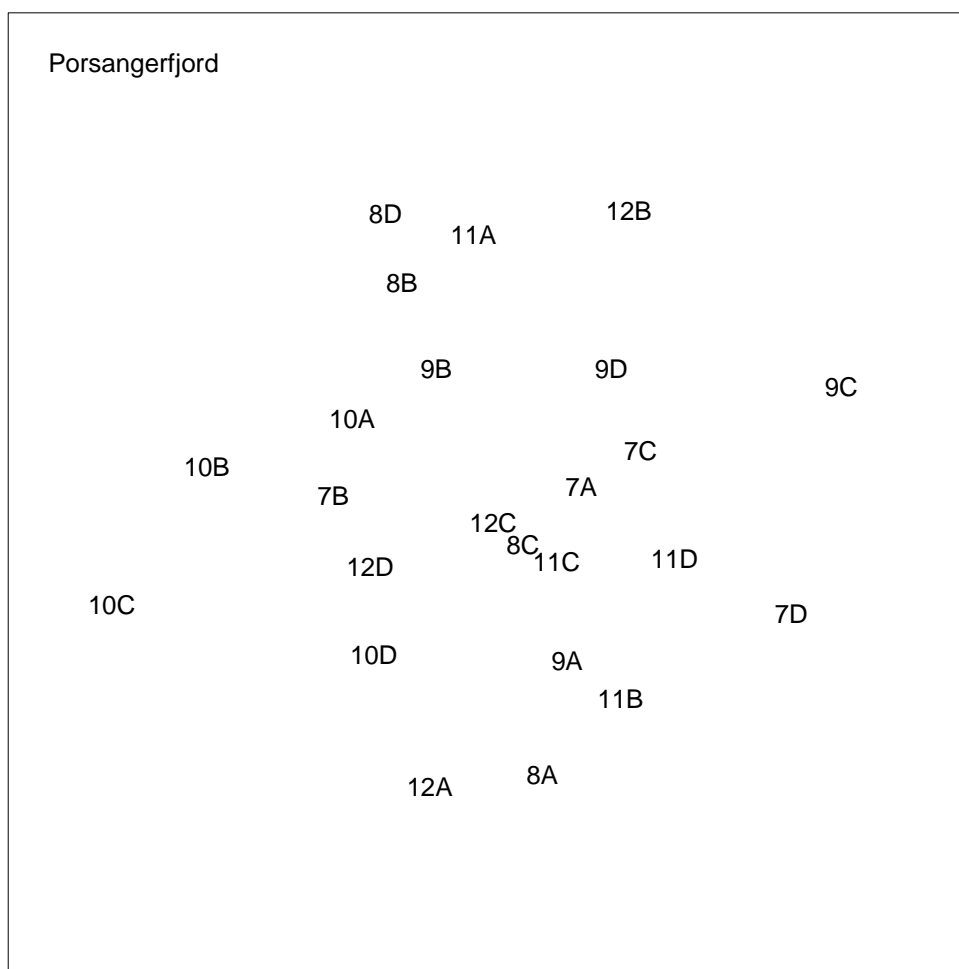


Figure 22. MDS-plot of fauna dissimilarity between the samples from Porsangen (the figures and letters are box and quadrant numbers respectively)

Table 7. Treatment types and levels, Eh values and fauna-parameter values in the samples from Oslofjorden.

Ch.no.	Section	Organic phase	Level	Number of species	Number of individuals	Diversity (H)	Diversity (ES ₁₀₀)
1	A	Ester	Low	22	84	3.56	
1	B	Ester	Control	29	146	3.75	24.6
1	C	Ester	High	16	56	3.21	
1	D	Ester	Medium	35	260	4.00	24.7
2	A	Olefin	Medium	22	146	3.47	18.3
2	B	Olefin	Low	31	219	3.95	22.7
2	C	Olefin	High	23	232	3.52	17.4
2	D	Olefin	Control	33	313	3.90	22.0
3	A	Olefin	Low	33	210	3.83	24.5
3	B	Olefin	Medium	-	-	-	-
3	C	Olefin	Control	32	227	3.57	22.8
3	D	Olefin	High	30	210	3.31	21.4
4	A	Ester	Medium	34	332	3.06	19.1
4	B	Ester	Control	36	460	3.30	19.3
4	C	Ester	Low	43	297	3.66	24.9
4	D	Ester	High	27	273	2.93	17.3
5	A	Olefin	Medium	18	119	3.52	17.1
5	B	Olefin	Control	25	88	4.02	
5	C	Olefin	High	18	76	3.72	
5	D	Olefin	Low	27	107	3.94	26.3
6	A	Ester	Low	24	101	3.75	23.9
6	B	Ester	Control	14	70	2.98	
6	C	Ester	Medium	25	112	3.48	23.5
6	D	Ester	High	35	158	4.03	29.0

Table 8. Treatment types and levels, Eh values and fauna-parameter values in the samples from Porsangen

Ch. no.	Section	Organic phase	Level	Number of species	Number of individuals	Diversity (H)	Diversity (ES ₁₀₀)
7	A	Olefin	Control	19	89	2.86	
7	B	Olefin	Low	23	104	3.15	22.4
7	C	Olefin	Medium	19	121	2.43	17.1
7	D	Olefin	High	19	132	2.44	16.2
8	A	Ester	Medium	19	105	2.5	18.6
8	B	Ester	Control	20	120	2.59	18.3
8	C	Ester	Low	16	119	2.28	15
8	D	Ester	High	15	119	2	13.9
9	A	Olefin	Control	22	116	3.03	21
9	B	Olefin	Low	20	110	2.58	19.2
9	C	Olefin	High	18	73	2.67	
9	D	Olefin	Medium	14	88	1.98	
10	A	Ester	Medium	16	120	2.15	14.8
10	B	Ester	High	17	94	2.65	
10	C	Ester	Control	19	123	2.78	17.3
10	D	Ester	Low	19	109	2.74	18.4
11	A	Olefin	Control	16	92	2.77	
11	B	Olefin	High	15	97	2.99	
11	C	Olefin	Low	19	87	3.34	
11	D	Olefin	Medium	17	72	2.59	
12	A	Ester	High	17	53	3.02	
12	B	Ester	Medium	15	66	2.65	
12	C	Ester	Control	20	75	3.52	
12	D	Ester	Low	20	76	2.99	

3.5 STATISTICAL ANALYSES

In Appendix 1, a statistical model was developed for treatment of the redox potentials measured at the end of the experimental period. In this approach, the experiment was designed to study effects of three factors.

1. *Environment* was the two sediment types including differences with regard not only to temperature, but also to biological communities, particle size, organic carbon and other inherent differences in the sediments from the two locations.
2. *Substance* was the two mud types including differences with regard not only to olefin or ester, but also to various mud additives. However, no indications were found that the additives had any effects on parameters measured during the present test.
3. *Dose* was the four different aliquots of 0, 5, 50 and 200 mg slurry added to each section of the various chambers. The aliquots corresponded to additions of 0, 0.5, 5 and 20 mg cm⁻² of the organic phase, but included also similar gradients of accompanying mud components and control sediment matrix.

The model was used to assess the effects of the three factors on final concentrations of organic phase (disappearance), redox potentials and community structure (number of individuals, number of species and diversity). In addition, the model was modified to assess effects on oxygen consumption. This parameter was measured at chamber level, only. Thus, the effects of *Dose* on oxygen consumption rates could not be assessed. However, oxygen consumption rates were measured bi-weekly throughout the

Table 9. *Environment, Substance, Dose* three-factor ANOVA analyses on final concentration of organic phase (C_{final}), redox potentials at 0.5 cm depth (E_h) and the macrofauna parameters N (number of individuals), S (number of species) and H (diversity index). Shaded F ratios show cases for which the probabilities were ≤ 0.05 .

	C_{final}	N	S	H	$E_{h, \text{final}}$
<i>Model</i>					
n (number of obs.)	42	48	48	48	47
DF (model)	24	24	24	24	24
DF (error)	17	23	23	23	22
r^2 (adjusted)	0.95	0.63	0.37	0.44	0.86
F ratio	35.86	4.36	2.14	2.51	12.39
Probability (p)	<.0001	0.0004	0.036	0.0153	<.0001
<i>Effects (F ratios)</i>					
E; Environment (DF=1)	0.00	22.73	18.05	19.50	21.20
S; Substance (DF=1)	0.09	0.95	0.36	0.10	33.40
D; Dose (DF=3)	266.6	0.84	2.01	3.94	50.34
E *S (DF=1)	1.12	1.13	2.18	0.61	1.69
E*D (DF=3)	0.25	0.59	0.20	0.17	4.69
S *D (DF=3)	1.12	1.67	1.47	1.24	6.87
E*S*D (DF=3)	4.75	1.20	1.88	1.46	4.33
Chamber (E, S) (DF=9)	0.97	6.70	1.04	1.89	2.10

Table 10. *Environment, Substance, Time* three-factor ANOVA analyses on sediment oxygen consumption rates. Shaded F ratios show cases for which the probabilities were ≤ 0.05 .

	SOC
<i>Model</i>	
n (number of obs.)	383
DF (model)	139
DF (error)	243
r^2 (adjusted)	0.52
F ratio	4.02
Probability (p)	<.0001
<i>Effects (F ratios)</i>	
E; Environment (DF=1)	65.56
S; Substance (DF=1)	1.65
T (Time) (DF=3)	8.51
E *S (DF=1)	0.38
E*T (DF=3)	2.51
S *T (DF=3)	0.90
E*S*T (DF=3)	0.44
Chamber (E, S) (DF=9)	11.11

experimental period. Thus, the three-ways ANOVA could be applied for this parameter as well, replacing the factor *Dose* with the factor *Time*.

Results of the ANOVA-analyses performed using the JMP® Statistical Software (SAS Institute Inc.) are shown in Table 9 and Table 10. The p-values ≤ 0.036 showed that the probabilities were equal to or less than 36/1000 that the variations in the respective effect-parameter occurred as a result of chance alone. Thus, the applied model could explain a significant proportion of the variation in all parameters analysed.

3.5.1 Disappearance

The mean concentrations given in ch. 3.1.2 frequently confirmed the expected result that the ester would disappear more rapidly than the olefin and that both chemicals would disappear more rapidly in the Oslofjord than in the Arctic sediment. The ANOVA analyses on final concentrations (Table 9) did, however, not confirm statistically significant effects neither from *Environment*,

Substance nor any of the two-ways interaction terms. The large F-ratio of 266.6 showed that *Dose* was the dominating factor explaining most of the variation in the final concentrations. This simply means that the more we add on day zero, the more remains in the sediment after three months.

However, in addition to this rather obvious statement, the analyses did reveal significant three-ways interactions ($F=4.75$) which showed that the combination of *Environment* and *Substance* was important with regard to how *Dose* affected the final concentration. Some unexpectedly rapid degradation of olefins was observed in the high dose arctic treatments. Obviously the temperature was not the most important component of the *Environment* factor. More likely, the bioturbating activity of *Ctenodiscus crispatus*, which was present in Porsangen sediment only, was a key factor which could enhance biodegradation in the Arctic chambers. Inhibition of this bioturbation was evident in one of the chambers (no. 12) treated with esters, within which all *Ctenodiscus* individuals had surfaced to avoid H_2S in the pore water. The fact that the final concentration of 17.1 mg cm^{-2} in chamber 12 was higher than in the replicate chambers (13.6 mg cm^{-2} in ch. 10, 14.6 mg cm^{-2} in ch. 8) seemed to confirm that biodegradation had been slower in sediments where *Ctenodiscus* activity had been inhibited by sulphide toxicity.

Thus, complex interactions between substance, dose and the local macrobenthic community may affect degradation rates in ways which may be difficult to predict. With regard to the evaluation of the present chemicals, it appears important to note that at high doses, chemicals highly available to sulphate reducing bacteria, may impose a stronger negative feed-back on degradation than substances with less tendency to degrade via anaerobic pathways.

3.5.2 Macrofauna

F-ratios of 18-23 (Table 9) showed that *Environment* could explain most of the variation in the macrofauna parameters. F-ratios ≤ 1 for *Substance* showed that type of chemical had no significant impact on the macrofauna parameters, whereas *Dose* had significant effects on diversity (F-ratio = 3.9). Interaction effects were not found.

Preliminary analyses of box-core samples collected on day 0, revealed differences between Porsangen and Oslofjord communities similar to the difference found at the end of the experimental period. Thus, the high F-ratios for *Environment* was a result of inherent community differences and must not be confused with treatment effects. In addition to the inherent difference between the two locations, high F ratios revealed differences between chambers, in particular with regard to number of individuals. The chamber-effect was also revealed in the MDS-plots (Figure 21 and Figure 22) and was interpreted as an inherent difference resulting from patchiness at the field locations.

Thus, the *Dose* effect on diversity was the only significant effect on the macrofauna community found to result from the various treatments of this experimental set-up. Whether the cuttings were based on olefins or esters was not important. This might seem contradictory to the proliferation of hydrogen sulphide in some of the ester-chambers and the obvious effects on *Ctenodiscus* behaviour. Probably, the sulphide had not been present for a sufficiently long period to affect the macrobenthic community structure.

3.5.3 E_h

The high values of r^2 in Table 9 showed that redox potentials and final concentrations were the two parameters best explained by the model. However, whereas the concentration data responded almost exclusively to *Dose*, the redox potentials showed significant contributions from all three experimental factors as well as significant two-ways E^*D and three-ways E^*D^*S interactions. The highest F-ratio (50.3) was found for *Dose*, showing that the more of the cuttings added, the lower the E_h . Secondly, the F-ratio of 33.4 for *Substance* showed that at similar dose and environment, sediments treated with esters will have lower E_h than sediments treated with olefins. Finally, the F-ratio of 21.2 for *Environment* showed that the final E_h was significantly different in similarly treated sediments from the two environments. The direction of the difference was towards lower potentials in the Porsangen sediment. Initial redox potentials determined at 0 and 1 cm depth were high in both environments (Figure 15), but the high potentials were restricted to a more narrow 0-2cm depth interval in the Porsangen sediment. Thus, it appears that upwards displacement of the shallow redox-cline may explain the apparently higher sensitivity of the Porsangen sediment to processes lowering the redox potential.

Both C_{final} and $E_{h, final}$ revealed the presence of complex three-ways interactions between substance, dose and the local environment. According to the above discussion, the absence of corresponding interactions in the macrofauna parameters (Table 9) may primarily result from the time-lag between microbial degradation and effects on the macrofauna level.

3.5.4 SOC

Because the model was different, the effects on SOC cannot be directly compared with the other parameters. Effects of *Dose* on SOC could not be assessed in the present test, but previous tests have shown that dose is a primary factor affecting respiration rates in sediments treated with drilling chemicals. Previous tests have also revealed large differences between different chemicals and a 19-24% higher total sediment oxygen consumption in chambers treated with FOE as compared to chambers treated with LAO. Therefore it may seem surprising that the F-ratio of 1.65 for *Substance* (Table 10) was so much smaller than the F-ratio of 65 for *Environment*. The different treatment of the chambers in the present as compared to previous experiment may have acted to reduce the effect of *Substance* on SOC. The distribution of 79% of the total load (which was similar in the two experiments) over 1/4 of the chamber area, should implicate a less aerobic degradation environment for most of the chemicals added in the present experiment. That this difference would favour biodegradation of the olefins appears controversial, but tended to confirm the unexpected results on disappearance of olefins in the high dose Porsangen sediments.

Anyhow the F-ratios in Table 10 showed that the difference between LAO and FOE with regard to stimulated sediment metabolism was rather marginal as compared to the importance of the local depositional environment. Whether this was an effect of lower temperatures, different organic back-ground enrichment or different benthic communities remains unclear, but significant chamber effects for SOC (F-ratio = 11.1) and number of individuals (F-ratio = 6.7), indicated the importance of inherent differences between individual chambers. The possible link between SOC and inherent macrobenthic biomass will be further discussed below.

4. DISCUSSION

4.1.1 Biogeochemical interactions in the source-sediments

In general, observed differences between the Oslofjord and Porsangen chambers may result from inherent differences in seabed samples, as well as from the difference with regard to experimental temperatures. The macrobenthic communities in the Porsangen sediments were characterised by, fewer species, fewer individuals and lower diversity indices. This does not necessarily imply that the arctic communities were functionally different from the Oslofjord communities. Some indications on functional differences were, however, found and their importance to experimental parameters are discussed below.

In the Porsangen sediment, dense assemblies of tubes of the polychaete *Spiochaetopterus typicus* were found at about 20 cm depth. Most of the tubes were abandoned, but some inhabited tubes penetrated the overlying sediment layer to leave an open end 1-2cm above the sediment surface. Irrigation through these tubes may have important effects on pore water dynamics and biological and geochemical processes in the overlying sediment, probably primarily at depths intermediate between the redox-cline and the depth of the tube assembly. No indications were found on interactions between the degradation of drill cuttings and the tube assemblies.

An obvious difference between the two source sediments, was the persistency of the redox-cline at 2 cm depth in the Porsangen samples (Figure 15, Figure 17 i,j,m,n). The decrease from ca 300 mV above to ca 0 mV below the cline, was similar at both locations, but in the Oslofjord the transformation occurred at larger and more variable depths (Figure 15, Figure 17 a,b,c,e,f,g). Bågander and Niemistö (1978) described three clusters of E_h -values. The lower cluster from -0.1 to -0.3V, was controlled by reduced sulphur. No observations of E_h in the present, untreated source sediments belonged to the latter group. The upper cluster between 0.2 and 0.4V was controlled by redox-couples including or influenced by oxygen and nitrate, whereas the Fe^{2+}/Fe^{3+} redox couple was thought to control intermediate potentials between 0.0 and 0.2V. If the polychaete assemblies maintain a continuous flow of seawater through the tubes for respiration and discharge at 20 cm depth in the sediment, upwards advection of pore water must result. A slow upwards flow of pore water is likely to become rapidly depleted in oxygen. If also sulphate reduction may be limited by the availability of organic carbon, the Fe^{2+}/Fe^{3+} redox couple may be left to control redox potentials at the intermediate level, as actually observed throughout the subsurface layer of the Porsangen sediment (Figure 15). The sharp transformation at 2 cm would represent the interface between the upwards advection of pore water and the more random bioturbative activities of *Ctenodiscus crispatus* which sustain oxygen control and high redox potentials within the 0-2cm surface layer. Thus the very characteristic vertical profile of redoxpotentials in the Porsangen sediment appeared to be controlled by the activities of these two macrobenthic species.

4.1.2 Test duration

In the previous test (Schaanning *et al.*, 1996), Shannon Wiener diversity indexes were clearly lowered in Oslofjord sediments treated with amounts of esters corresponding to the present medium dose level. Thus, it might seem surprising that no clear effects were found on the indexes calculated for the present communities, not even in the high dose treatments which had received a 4x higher loading. Effects were however, observed both with regard to oxygen

consumption rates and redox potentials. Such effects would be expected to precede changes at the macrofauna level, and the behaviour of the *Ctenodiscus* in chamber 12 (Ch. 2.6.5), indicated that effects were about to become manifest, also at the macrofauna level. Furthermore, 31-84% of the added chemicals were still present in the high and medium dose sediments (Table 4), providing a considerable potential for further development of effects. It appears therefore, that the three months duration of the present experiment, was too short for significant development of effects in the macrofauna community.

The remaining of a major fraction of the added organic chemicals was a problem, also with regard to interpretation of the oxygen consumption rates, and particular with regard to the development of SOC in the Arctic chambers. The trend appeared very consistent but the large fluctuations tended to obscure the significance of the cross-over observed less than two weeks before the termination of the experiment. A prolonged test period might have clarified whether the cross-over was a permanent feature, a temporary peak or merely a result of random fluctuations. The latter appeared not very likely. If the cross-over had been proven to be a permanent feature, it would have been a very strong evidence confirming the hypothesis that the potential for biodegradation in Arctic communities, after a few months of adaptation, was no less than the potential for biodegradation in more temperate communities. On the other hand, further development of effects on key species in the Arctic macrofauna might have given an opposite effect on biodegradation rates.

It appears that the test period may have been adequate with regard to the low dose sections, acceptable with regard to medium dose, but too short for proper elucidation of the biodegradation event in the high dose sections and consequently too short for proper elucidation of whole chamber respiration rates.

4.1.3 Biodegradation in the Arctic sediment

Even though the *Environment* factor could not explain much of the variance in the final concentrations of esters and olefins (Table 9), the mean concentrations remaining in the Arctic sediment three months after addition were frequently higher than the concentrations remaining in correspondingly treated Oslofjord sediments (Table 11). However, the difference was only significant in the medium dose olefin treatment, and did not apply to the concentrations normalised against barium. The right-hand column of Table 11, shows that

Table 11: Difference (Porsangen - Oslofjord) between means of concentrations of organic phase and barium and OP:Ba ratios in low, medium and high dose treatments. Shading show differences which were significant at 95% (Tukey-Kramer HSD comparison for all pairs at each dose level (n=12)).

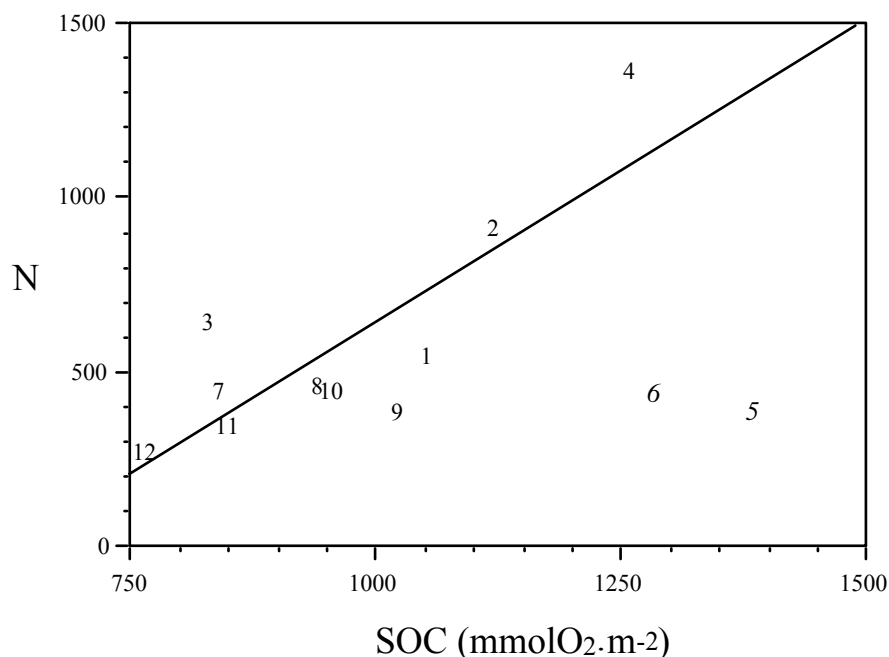
	$\Delta[\text{OP}]$	$\Delta[\text{Ba}]$	$\Delta([\text{OP}]:[\text{Ba}])$
<i>Low dose</i>			
Olefin	0.11	-0.02	0.22
Ester	0.03	-0.03	0.06
<i>Medium dose</i>			
Olefin	1.52	1.21	0.18
Ester	0.32	0.55	0.03
<i>High dose</i>			
Olefin	-2.89	-2.70	-0.002
Ester	2.86	1.01	0.15

OP:Ba ratios were never significantly different in sediments from the two environments. Thus, the concentration data gave no statistically significant evidence to support the hypothesis of slower degradation in Arctic sediments at -0.5°C as compared to temperate sediments at $6-8^{\circ}\text{C}$.

Oxygen consumption rates were clearly lower in the Arctic as compared to the Oslofjord sediments (Figure 14). The respiration rate cross-over observed towards the end of the three-months period, showed, however, that the difference may have been an initial effect resulting from slower adaptation rather than lower long-term potential for biodegradation

Table 12. Linear regression analyses of total sediment oxygen consumption vs. number of individuals (N = a + b·SOC).

	a	b	n	r ² (adj.)	p
All chambers	-53	0.6	12	0.07	0.208
Ch. 5 omitted	-502	1.08	11	0.29	0.051
Ch. 5&6 omitted	-1105	1.75	10	0.63	0.004

**Figure 23. Number of individuals (N) versus total sediment oxygen consumption (SOC) for the experimental period. The plotted regression line applies to ten chambers omitting nos. 5 and 6 (see Table 12).**

of the two chemicals. Slower adaptation might result both from smaller inherent degrader population size and/or longer generation times. Only the latter would be a clear temperature effect. Micro-organisms are generally assumed to account for most of the oxygen consumed in a sediment sample, but a positive correlation might be expected between inherent biomass or population size of micro- and macro-organisms. A regression analyses was therefore performed to elucidate the possible correlation between macrobenthic population size and total sediment respiration. As shown in Figure 23 and Table 12 a positive correlation was found. The correlation was however, not significant unless the data from one or two chambers were omitted. Thus, the data gave some support to the idea that a larger macrobenthic population is frequently coupled with higher sediment oxygen consumption. Such coupling might explain the similarity between the results of the three-ways ANOVA for SOC (Table 9) and N (Table 10).

If significant effects of *Environment* resulted primarily from inherently richer communities consuming more oxygen in the Oslofjord sediment, and the significant chamber-effects resulted primarily from large variations in number of individuals and oxygen consumption

between separate chambers, the lower rates of oxygen consumption in the Arctic sediments, appeared to be an initial effect resulting from inherent differences in the benthic community rather than from biochemical processes proceeding more slowly at the sub-zero temperatures. After addition of cuttings, the inherently smaller degrader populations in the Arctic sediment will grow to yield oxygen consumption rates similar to those of the Oslofjord communities. Thus, the present investigation indicated that initially different rates of sediment metabolism were primarily limited by the availability of organic carbon and the similarity of the rates observed at the end of the study showed that the potential for degradation of these chemicals was not different in Arctic as compared to more temperate sediment environments. Thus, the previously documented presence and efficiency of arctic hydrocarbon degrader bacteria (Pedersen *et al.*, 1979, Thingstad and Martinussen, 1991) was shown to apply for the presently applied esters and olefins as well. The similarity of bacterial efficiencies were confirmed by the similarity of concentrations of chemicals remaining in the sediments from the two environments and the concentration data gave no evidence that the slow remediation frequently found for oil spills in Arctic shore-line environments (Gulliksen and Taasen, 1982, Haines and Atlas, 1982, Boehm *et al.*, 1987) will apply to the present chemicals in drill cuttings deposited in sub-littoral environments even at sub-zero temperatures.

5. CONCLUSIONS

1. Throughout the three months experimental period, temperatures were maintained close to 7°C in chambers transferred from the Oslofjord and -0.5°C in chambers transferred from the Arctic.
2. Disappearance rates were not significantly different in the two environments. Esters tended to disappear faster than the olefins, but not in the high dose Arctic treatments.
3. Concentrations of olefins as well as esters decreased to detection limits in low dose Oslofjord sediments.
4. Initial oxygen consumption rates were lower in Arctic than in Oslofjord chambers, but increasing trends and cross-over towards the end of the experimental period showed adaptation of the Arctic degrader community to the added chemicals.
5. The biogeochemistry of the Arctic sediments appeared strongly influenced by the activities of two macrobenthic species, the surface bioturbator *Ctenodiscus crispatus* and the tube-dwelling polychaete *Spiochaetopterus typicus*. Immobilisation of the surface bioturbator by hydrogen sulphide produced in one of the high dose ester treatments, appeared to have negative feed-back on biodegradation rates.
6. With regard to effects during degradation event, the tendency of the esters to stimulate sulphate reducing bacteria and the fact that the Arctic communities appeared to be less sulphide tolerant than the Oslofjord communities, would tend to disfavour discharge of esters in Arctic environments.
7. Carbon-fluxes may be more important than temperatures in explaining biological differences between the Arctic and the Oslofjord location.
8. No evidence was found to support the hypothesis that rapid anaerobe degradation would yield shorter sea-bed remediation times of areas contaminated with esters as compared to areas contaminated with LAO-olefins. On the contrary, indications were found that maintenance of bioturbation in sediments heavily contaminated with LAO-olefins might be more important to sea-bed remediation than the availability of the esters to sulphate reducing bacteria.
9. A prolonged test period might have clarified the full impact of the high dose treatments on the macrobenthic communities and consolidated the evidence for the high biodegradation potential of the Arctic communities.

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APPENDIX 1
STATISTICAL ANALYSES OF REDOX POTENTIALS
Birger Bjerkeng, NIVA

Description of design

The complete experimental design has 4 treatment factors: *Environment*, *Substance*, *Dose* and *Depth* and 4 sizes of experimental units: *Chamber*, *Section*, *Profile* (i.e. below a given point on the surface of the sediment) and finally single *Point of measurement*.

The first treatment factor, *Environment*, with $E=2$ fixed levels *Oslofjord* and *Porsangen*, represents the properties of the two sediment types collected from two geographical locations and the different experimental conditions they were subject to, each assigned to a separate experimental basin under different temperature. Thus, the effects of basin and temperature are confounded with effect of natural environment or sediment type, and do not enter the analysis as separate factors.

The second treatment factor, *Substance*, with $S=2$ fixed levels *Ester*, *Olefin*, was assigned to the largest experimental unit in the design, the chambers of sediment. Each substance was randomly assigned to 3 of the 6 chambers of each sediment type. *Chamber* thus enters the analysis as a random factor, nested with $C=3$ chamber within each combination of Environment and Substance.

The third treatment factor, *Dose*, with $D=4$ fixed levels *Control*, *Low*, *Medium* and *High*, was assigned randomly to the next size of experimental unit, which are the 4 *Sections* each chamber was subdivided in. *Section* does not enter the analysis as an explicit factor, because there is only one section per chamber for each dose. The effect of within-chamber variation between sections is confounded with the random interaction between Dose and Chamber.

Redox potential was measured in $P=3$ profiles in each section, and this is the next smallest experimental unit, entering the design as replication within Environment*Substance*Dose. In each profile, measurements were done at $Z=2$ *Depths*, 0.5 and 2 cm. Depth is assigned to the smallest experimental unit, i.e. the single measurement point, and represents a so-called repeated measure on the profiles. This takes account of the fact that individual measurements are not done on separate and independent entities, but are paired with two values from each profile.

Options for statistical analysis

In principle one could analyse the data from both depths together with depth as a factor as described above. However, a preliminary inspection by plots (Figure 1-1) show that the values for the two depths are well correlated, but with a striking differences between the two sediment types both in the mean and the variation of the difference between depths. For sediment from the Oslofjord the two depths give almost identical values for most points, independent of Substance and Dose treatments. For a number of observations there is a slight difference of around 20 between the depths. Three of 134 data points stand out as outliers, with a difference between the two depths in the range 70-130. The outliers are from chamber 1 and 6, where a large depth effect was noted in depth profiles below white surface spots. In contrast to this, the Porsangen data show much larger differences between the two depths in general, and also a larger variability of the difference. This is in accordance with the results of the vertical profiles discussed in chapter 3.3.1. These differences between the two sediment types speaks against analysing the depth effect as part of the general analysis of variance. It is obvious that analysing the depth effect as part of a multifactor analysis of variance can only be of interest for the Porsangen data.

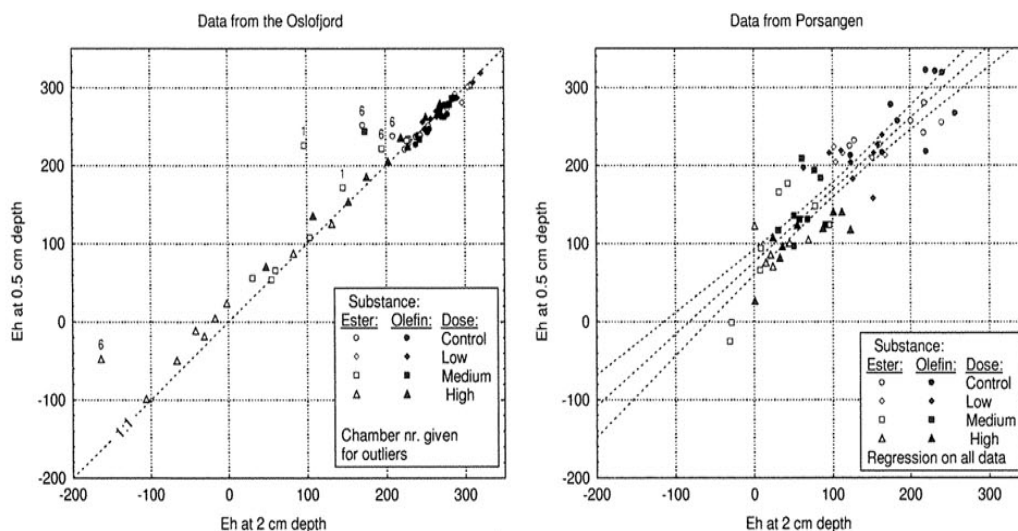


Figure 1-1. Correlation between Redox potential (Eh) at 0.5 and 2 cm

Thus, the most natural option is to analyse for the effects of main interest, namely environment, substance and dosage, on data from one depth only. The 2 cm data have the advantage of being balanced, but the effect of environments and interactions including environment could be masked by variations in position of the depth of the redoxcline. The data from 0.5 cm are believed to represent best the effects of the additions for both environments or sediment types. However, this data set is not quite balanced; most of the 0.5 cm data for chamber 12 (Porsangen, Ester addition) are missing. Special care must be taken when analysing unbalanced data, to avoid testing special and complex contrasts that are difficult to interpret.

We have chosen to do the analysis on the 0.5 cm data. We might have used these data as they are, with only 2 chambers instead of 3 for one treatment combination. However, this would mean throwing away the information available in the 2 cm data about the differences between chamber 12 and the other two Porsangen chambers with ester addition. At 2 cm sediment depth chamber 12 has generally lower values than chamber 8 and 10. This is also the case for the few data that exists for 0.5 cm from this chamber. Because of this we have chosen to complete the 0.5 cm data set as described below.

Estimating missing data for 0.5 cm depth

The 0.5 cm data are completed with estimates based on the 2 cm data for chamber 12 by using a depth correction based on the other two chambers of Porsangen sediment with ester addition (chambers 8 and 10). The data for these chambers can be analysed as a design with two fixed treatment factors: *Dose* ($D=4$ levels) and *Depth* ($Z=2$ levels), and different sizes of experimental units: *Chamber* as random block with $X=2$ levels, *Section* as the whole plot (1,...,4) within Chamber, assigned randomly to the Doses, with $P=3$ replicate *Profiles* within each section, and depth as a repeated measure, assigned to the basic experimental unit, which is each measurement *point*. The ANOVA model for these data can be written:

$$Eh_{cdzp} = \mu + \varepsilon_c + \gamma_d + \tau_z + (\gamma\tau)_{dz} + \varepsilon_{cd} + \varepsilon_{cz} + \varepsilon_{cdz} + \varepsilon_{(cd)p} + \varepsilon_{(cd)zp}$$

where ϵ_c , ϵ_{cd} , ϵ_{cz} , ϵ_{cdz} , $\epsilon_{(cd)p}$ and $\epsilon_{(cd)zp}$ are the different error (random variation) terms for chamber (c), dose (d), depth (z), profile (p) within sections and random interactions of these terms, with respective variances σ_c^2 , σ_{cd}^2 , σ_{cz}^2 , σ_{cdz}^2 , $\sigma_{(cd)p}^2$ and σ_z^2 .

The resulting mixed model has the following variance components:

	Source of Variation	Degr. of Freedom	Expected Mean Square	Error term
ϵ_c	Error(Chamber)	X-1	$Z\sigma_p^2 + DZP \cdot \sigma_c^2$	$\epsilon_{(cd)p}$
γ_d	Dose	D-1	$Z\sigma_p^2 + ZP \cdot \sigma_{cd}^2 + CZP \sum_d \gamma_d^2 / (D-1)$	ϵ_{cd}
τ_z	Depth	Z-1	$\sigma_z^2 + DP \cdot \sigma_{cz}^2 + CDP \sum_z \tau_z^2 / (Z-1)$	ϵ_{cz}
ϵ_{cd}	Error(Chamber·Dose)	(X-1)·(D-1)	$Z\sigma_p^2 + ZP \cdot \sigma_{cd}^2$	$\epsilon_{(cd)p}$
ϵ_{cz}	Error(Chamber·Depth)	(X-1)·(Z-1)	$\sigma_z^2 + DP \cdot \sigma_{cz}^2$	$\epsilon_{(cd)zp}$
$(\gamma\tau)_{dz}$	Dose·Depth	(D-1)·(Z-1)	$\sigma_z^2 + P \cdot \sigma_{cdz}^2 + CP \cdot \frac{\sum_{dz} (\gamma\tau)_{dz}^2}{(D-1)(Z-1)}$	ϵ_{cdz}
ϵ_{cdz}	Error(Chamber·Dose·Depth)	(X-1)·(D-1)·(Z-1)	$\sigma_z^2 + P \cdot \sigma_{cdz}^2$	$\epsilon_{(cd)zp}$
$\epsilon_{(cd)p}$	Error(Chamber·Dose·Profile)	X·D·(P-1)	$Z\sigma_p^2$	
$\epsilon_{(cd)zp}$	Error(residual)	X·D·(Z-1)·(P-1)	σ_z^2	

and the corresponding ANOVA table is:

	df	MS	F	p
Error(Chamber)	1	5,292	7.09	0.017
Dose	3	64,796	18.87	0.019
Depth	1	64,240	95.17	0.065
Error(Chamber·Dose)	3	3,434	4.60	0.017
Error(Chamber·Depth)	1	675	1.20	0.290
Dose·Depth	3	450	0.47	0.723
Error(Chamber·Dose·Depth)	3	953	1.69	0.209
Error(Chamber·Dose·Profile)	16	747		
Error(residual)	16	563		

The depth effect is not strongly significant, with $p=0.065$, but can still be used as the best possible estimate of a mean depth correction. There are no significant interaction terms involving depth, and consequently no reason to apply different depth corrections for different doses. The means plot below (Figure 1-2) shows a fairly uniform difference between the two depths across doses.

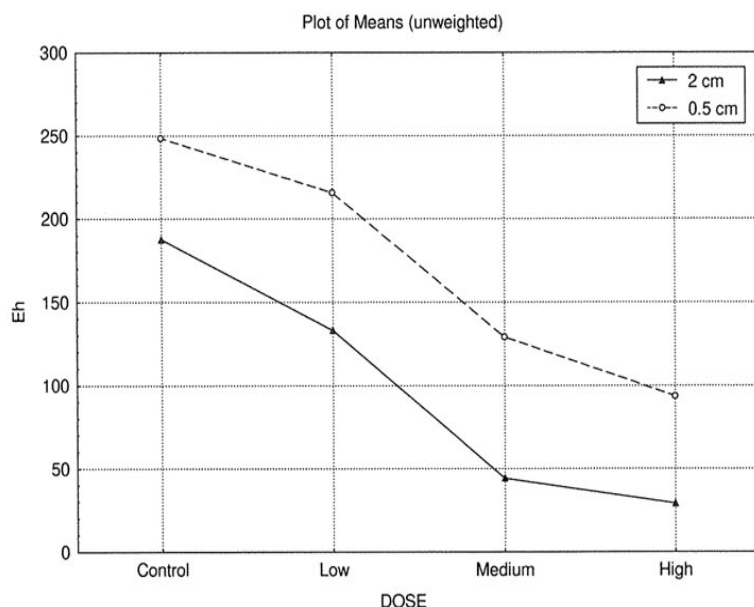


Figure 1-2. Means of Eh in Porsangen sediment chambers with complete data sets at both depths for different doses of ester addition.

The mean difference between the two depths based on these data is $Eh_{0.5cm} - Eh_{2cm} = 73$. The missing 0.5 cm data can be estimated by adding 73 to the 2 cm data. This produces a complete data set for 0.5 cm. Completing the data in this way can generally be expected to increase interaction effects including environment and or substance. This should be kept in mind when drawing conclusions from the result. The degrees of freedom have not been reduced in the analysis, since the converted 2 cm data contribute degrees of freedom.

Analysis of the completed data set from 0.5 cm depth

The ANOVA model for data from a single depth over both environments can be written as a mixed model, with both fixed and random factors:

$$Eh_{escdp} = \mu + \alpha_e + \beta_s + (\alpha\beta)_{es} + \varepsilon_{(es)c} + \gamma_d + (\alpha\gamma)_{ed} + (\beta\gamma)_{sd} + (\alpha\beta\gamma)_{esd} + \varepsilon_{(es)cd} + \varepsilon_{(escd)p}$$

where:

- μ = overall mean
- α_e = fixed effect of environment $e=1,2$
- β_s = fixed effect of substance $s=1,2$
- $(\alpha\beta)_{es}$ = interaction between environment and substance
- $\varepsilon_{(es)c}$ = random variation (σ^2_c) between chambers within combination of environment and substance $c=1,\dots,3$
- γ_d = fixed effect of dose $d=1,\dots,4$
- $(\alpha\gamma)_{ed}$ = interaction between environment and dose
- $(\beta\gamma)_{sd}$ = interaction between substance and dose
- $(\alpha\beta\gamma)_{esd}$ = interaction between environment, substance and dose
- $\varepsilon_{(es)cd}$ = random variation (σ^2_{cd}) between sections within chamber
- $\varepsilon_{(escd)p}$ = random variation (σ^2_p) between points within sections $p=1,\dots,3$

Note that there are no interaction terms between Chamber and Environment or Substance, since Chamber is nested within the combination of these two factor. The basic experimental unit is now observation profile (p), which is now equivalent to single point, as within-section replicate.

The variance components of the various effects and error terms according to the standard mixed model are given in the table below:

Effect or Error	Degrees of Freedom	Expected Mean Square
α_e	$(E-1)$	$\sigma^2 + P \cdot \sigma_{cd}^2 + DP \cdot \sigma_c^2 + SCDP \frac{\sum_e \alpha_e^2}{(E-1)}$
β_s	$(S-1)$	$\sigma^2 + P \cdot \sigma_{cd}^2 + DP \cdot \sigma_c^2 + ECDP \frac{\sum_s \beta_s^2}{(S-1)}$
$(\alpha\beta)_{es}$	$(E-1)(S-1)$	$\sigma^2 + P \cdot \sigma_{cd}^2 + DP \cdot \sigma_c^2 + CDP \frac{\sum_{es} (\alpha\beta)_{es}^2}{(E-1)(S-1)}$
$\varepsilon_{(es)c}$	$ES(C-1)$	$\sigma^2 + P \cdot \sigma_{cd}^2 + DP \cdot \sigma_c^2$
γ_d	$D-1$	$\sigma^2 + P \cdot \sigma_{cd}^2 + ESCP \cdot \frac{\sum_d \gamma_d^2}{(D-1)}$
$(\alpha\gamma)_{ed}$	$(E-1)(D-1)$	$\sigma^2 + P \cdot \sigma_{cd}^2 + SCP \cdot \frac{\sum_{ed} (\alpha\gamma)_{ed}^2}{(E-1)(D-1)}$
$(\beta\gamma)_{sd}$	$(S-1)(D-1)$	$\sigma^2 + P \cdot \sigma_{cd}^2 + ECP \cdot \frac{\sum_{sd} (\beta\gamma)_{sd}^2}{(S-1)(D-1)}$
$(\alpha\beta\gamma)_{esd}$	$(E-1)(S-1)(D-1)$	$\sigma^2 + P \cdot \sigma_{cd}^2 + CP \cdot \frac{\sum_{esd} (\alpha\beta\gamma)_{esd}^2}{(E-1)(S-1)(D-1)}$
$\varepsilon_{(es)cd}$	$ES(C-1)(D-1)$	$\sigma^2 + P \cdot \sigma_{cd}^2$
$\varepsilon_{(escd)p}$	$ESCD(P-1)$	σ^2

The analysis of the 0.5 cm data according to this model gives the following ANOVA table:

Source of Variation		df	MS	F	p-level	p-level , rank tests
Chamber analysis:						
Environment	α_e	1	79853	8.09	.0217	0.002*
Substance	β_s	1	120236	12.18	.0082*	0.011
Environment*Substance	$(\alpha\beta)_{es}$	1	15109	1.53	.251	0.233
Error (Chamber)	$\varepsilon_{c(es)}$	8	9871	2.57	0.035*	0.0528
Section analysis:						
Dose	γ_d	3	194809	50.73	$1.5 \cdot 10^{-10}***$	$3.5 \cdot 10^{-10}***$
Environment*Dose	$(\alpha\gamma)_{ed}$	3	17146	4.465	.0125	0.0025*
Substance*Dose	$(\beta\gamma)_{sd}$	3	26865	6.996	.001524*	0.0091*
Environment*Substance*Dose	$(\alpha\beta\gamma)_{esd}$	3	16310	4.247	.015295	0.024
Error (Section)	$\varepsilon_{(es)cd}$	24	3840	3.857	.000001***	0.000001
Error (Residual)	$\varepsilon_{(escd)p}$	96	995.5			
Total:		132				

The results indicate significance of most of the various effects and interactions. In particular, there is a very significant contribution to experimental error from variation between whole sections of equal Environment*Substance levels, common to the three measurement points ($p=0.000001$) and also signs of variation between chambers ($p=0.035$) in addition to that expected from the random variation on the Section level. Using the expressions for the expected mean squares, we can estimate the different error terms to have standard deviations $\sigma=31.5$ (residual), $\sigma_{cd}=30.8$ (section), and $\sigma_c=22.4$ (chamber).¹

The results are based on the assumption that residuals are normally distributed and with homogeneous variances across factors. Although the variance analysis is fairly robust against violations of these assumption, they should be examined.

Normal distribution plots of residuals show that the normality assumption is not met, however, this is normally not serious. Besides, a parallel analysis were done on the ranks of values, leading to quite similar results, as is seen in the right-most column of the table above. According to Montgomery (1991) this often means that one can rely on the analysis of the original data.

The ordinary tests for non-homogeneous variances (Bartlett's test, Levene's test) strongly indicate that the variance differs between groups ($p < 5 \cdot 10^{-6}$). This fact in itself is not so important, but a plot of standard deviations against means indicate that the variance is much smaller for mean values above 250 than for lower means, and this could mean that the significance of effects are somewhat exaggerated. Also, it can be noted from Figure 1-1 that for the Oslofjord sediment data there is larger within-treatment variation for high doses (triangles) than for the other doses, while this tendency is not as clear in the Porsangen data.

To have an experiment-wise significance level of 0.05 for testing N fixed effects simultaneously, we can look at the p values sorted by value, and apply a limiting value $1-0.95^{1/(N-k+1)}$ to the k 'th smallest p value. Applying this criterion to the 7 fixed order effects in the table above, we can conclude that all effects and interactions except the *Environment*Substance* interaction are significant, with an overall significance level of 5 %. The estimated mean squares of the different effects and interactions can be calculated based on the ANOVA table and the expressions for the expected mean squares, as squared sums of effects divided by degrees. For the Dose effect, for example, the mean square is estimated as $\bar{\gamma}^2 = \sum_d \gamma_d^2 / (D-1) = (MS_d - MS_{(es)cd}) / (ESCP)$. The results for the significant fixed-effect terms are:

Effect	Mean square
Environment	$\bar{a}^2 = 972$
Substance	$\bar{b}^2 = 1533$
Dose	$\bar{\gamma}^2 = 3978$
Environment*Dose	$\overline{(\alpha\gamma)}_{..}^2 = 739$
Substance*Dose	$\overline{(\beta\gamma)}_{..}^2 = 1279$
Environment*Substance*Dose	$\overline{(\alpha\beta\gamma)}_{...}^2 = 1385$

The interaction terms are fairly large compared to the Environment and Substance effects. The general conclusion is that the response is complex, so one must be cautious in making inferences about the main effects of *Environment* or *Substance*. Both of the two-way interaction terms including *Dose* are also significant.

¹ This is based on the so-called unrestricted mixed model, where Error(Chamber) is compared to Error(Section).

Figure 1-3 shows a half-normal plot of residual standard deviation. Tests indicate that the residual variance is significantly heterogeneous, and the normal probability plot indicate 45 as an upper limit estimate to the residual standard deviation. Using this upper limit would add 1030 to the error terms used in the tests, and increase the p values somewhat, but would not make a large difference on the results

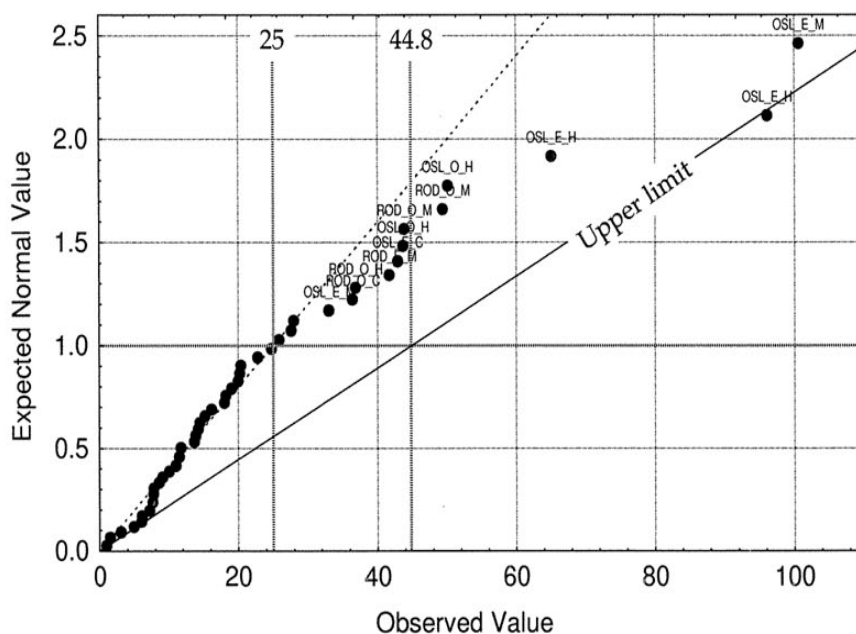


Figure 1-3. Half-normal plot of residual within-section standard deviation of Eh at 0.5 cm depth

The estimated means for the different treatments are shown in Figure 1-4. Some conclusions that can be drawn from this plot are:

For the both types of sediments and substances there appears to be a *dose* effect.

For sediment from Porsangen the two substances, *Medium* and *High Dose* have about the same effect, with Eh lowered about 150 compared to the controls for both substance, while *Low dose* are intermediate. There is further a consistent difference between substances, in that *Ester* addition leads to lower Eh values than *Olefins* by about 50. This is also the case for the control, where no substance were added, indicating that the control section is not unaffected by the additions to the other sections in each chamber.

For the Oslofjord sediment, the profiles across Doses are quite different. Addition of Olefin has much weaker effect than for the Porsangen sediment, and only for the High Dose. The addition of Ester, on the other hand, shows a much stronger effect with increasing dose, both compared with the Olefin curve, and compared with the ester curve for Porsangen sediment.

Figure 1-4 shows an interaction plot for the 0.5 cm data with Dose/Response curves for the mean effect of Ester and Olefin on the two sediment types. In the Oslofjord sediment, Ester seems to have a much larger effect on redox potential than Olefin, while the Porsangen sediment is about equally affected by both substances, intermediate between the two Dose/Response curves for the Oslofjord sediment.

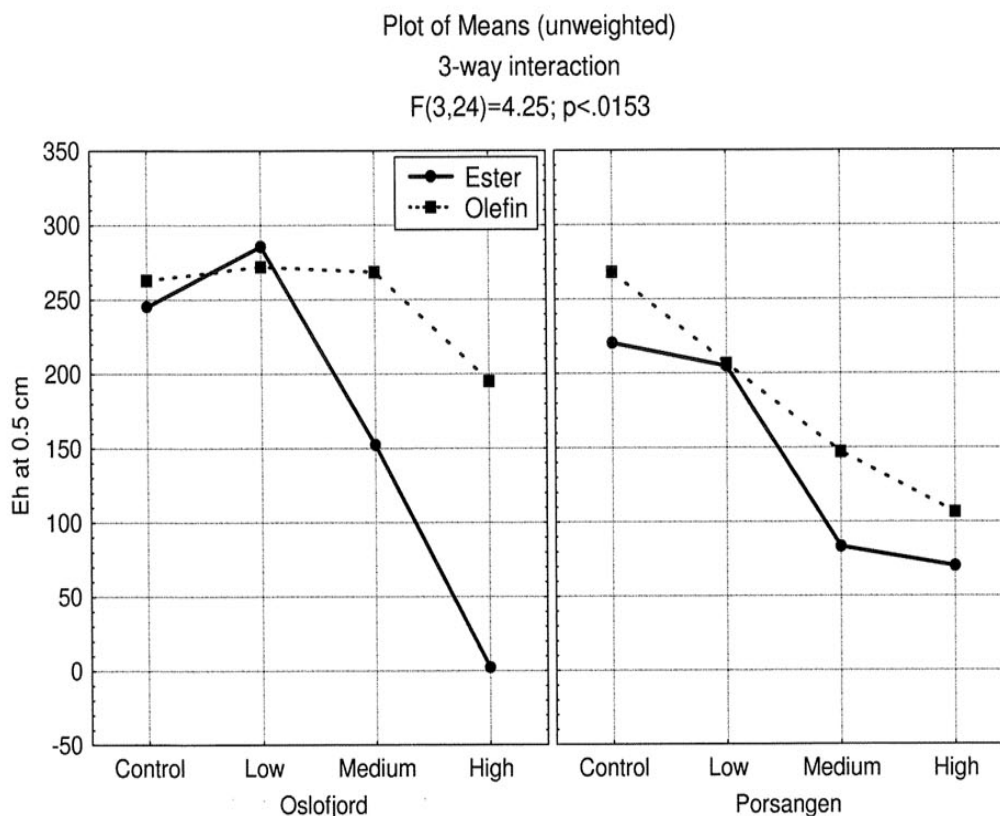


Figure 1-4. Estimated mean Eh at 0.5 cm depth for different treatments

The experimental procedure cannot completely rule out the possibility that the Control sections might be affected by the addition of Ester or Olefin to the other sections within the same chamber. To check this, a separate analysis was performed on the 0.5 cm data from the Control sections of the different chambers, with Environment and Substance as fixed factors, and Chamber as random factor nested within Environment*Substance as before. The result is given in the ANOVA table below.

		Effect		Error		F	p-level
		df	MS	df	MS		
Environment	Fixed	1	870.25	8	3800.4	0.229	0.645
Substance	Fixed	1	9571.36	8	3800.4	2.52	0.151
Environment * Substance	Fixed	1	2010.03	8	3800.4	0.529	0.488
Chamber	Random	8	3800.4	24	418.2	9.09	0.000011
Error (residual)		24	418.2				

There is a strongly significant random variation between chambers compared to the residual between-points variance, but no significant effects of Environment and/or Substance when tested against the variation between Chambers. *Thus, there is no reason to reject the assumption that the controls are unaffected by additions to the other sections.*

APPENDIX 2
ANALYSES OF BARIUM

Laboratory report. 4pp.

Ingegerd Rustad, SINTEF



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1997-08-22

Oppdrag nr.:
664070.70

Prøveserie.:
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Oppdragets tittel:

ANALYSE AV BARIUM I SEDIMENTPRØVER

Innledning

Prøvene ble mottatt i mai og juli 1997 for analyse Barium.

Prøvebeskrivelse

Prøvesettet besto av 95 sedimentprøver, merket som vist i Tabell 1.

Eksperimentelt

Prøvene ble tørket ved 110 °C og analysert ved hjelp av røntgenfluorescensspektrometri (XRF).



Resultat

Resultatet er gitt i Tabell 1.

Tabell 1

SINTEF Nr.	Prøve	Ba (Vekt%)
1997-474-1	Slurry Ester I	7,29
1997-474-2	Slurry Ester II	7,53
1997-474-3	Slurry Ester III	7,34
1997-474-4	Slurry Olefin I	8,40
1997-474-5	Slurry Olefin II	8,38
1997-474-6	Slurry Olefin III	8,28
1997-474-7	1d	0,379
1997-474-8	2b	0,131
1997-474-9	3a	0,139
1997-474-10	4a	0,337
1997-474-11	5d	0,159
1997-474-12	6c	0,370
1997-474-13	7c	0,335
1997-474-14	8c	0,114
1997-474-15	9d	0,467
1997-474-16	10d	0,083
1997-474-17	11d	0,355
1997-474-18	12d	0,089
1997-474-19	1c1	3,82
1997-474-20	1c2	0,148
1997-474-21	1c3	0,093
1997-474-22	1c4	0,085
1997-474-23	11b1	0,074
1997-474-24	11b2	0,112
1997-474-25	11b3	0,079
1997-474-26	11b4	0,068
1997-474-27	1b1	3,88
1997-474-28	1b2	0,078
1997-474-29	1b3	0,082
1997-474-30	1b4	0,080
1997-474-31	11a1	0,059
1997-474-32	11a2	0,060
1997-474-33	11a3	0,067
1997-474-34	11a4	0,068
1997-474-35	3/7 7a	0,071
1997-474-36	3/7 9a	0,057
1997-474-37	3/7 12a	1,021
1997-474-38	3/7 1b	0,083




1997-474-39	3/7 5b	0,084
1997-474-40	3/7 6b	0,081
1997-474-41	3/7 8b	0,075
1997-474-42	3/7 11b	0,826
1997-474-43	3/7 12b	0,293
1997-474-44	3/7 11C	0,098
1997-474-45	3/7 12C	0,065
1997-474-46	3/7 11d	0,460
1997-474-47	3/7 12d	0,086
1997-474-48	9/7 7c 0-1	0,397
1997-474-49	9/7 7c 1-3	0,063
1997-474-50	9/7 7c 3-5	0,063
1997-474-51	9/7 7c 5-7	0,066
1997-474-52	9/7 7d 0-1	3,35
1997-474-53	9/7 7d 1-3	0,120
1997-474-54	9/7 7d 3-5	0,069
1997-474-55	1a ester	0,146
1997-474-56	1c ester	1,155
1997-474-57	1d ester	0,401
1997-474-58	4a ester	0,262
1997-474-59	4b ester	0,088
1997-474-60	4c ester	0,113
1997-474-61	4d ester	0,936
1997-474-62	6a ester	0,119
1997-474-63	6c ester	0,330
1997-474-64	6d ester	1,029
1997-474-65	8a ester	0,317
1997-474-66	8c ester	0,123
1997-474-67	8d ester	1,007
1997-474-68	10a ester	0,295
1997-474-69	10b ester	0,961
1997-474-70	10c ester	0,075
1997-474-71	10d ester	0,089
1997-474-72	2a olefin	0,471
1997-474-73	2b olefin	0,124
1997-474-74	2c olefin	1,586
1997-474-75	2d olefin	0,082
1997-474-76	3a olefin	0,131
1997-474-77	3b olefin	0,379
1997-474-78	3c olefin	0,081
1997-474-79	3d olefin	1,033
1997-474-80	5a olefin	0,452
1997-474-81	5c olefin	1,360
1997-474-82	5d olefin	0,138



1997-474-83	7b olefin	0,113
1997-474-84	7c olefin	0,401
1997-474-85	7d olefin	0,979
1997-474-86	9b olefin	0,094
1997-474-87	9c olefin	0,903
1997-474-88	9d olefin	0,394
1997-474-89	11a olefin	0,073
1997-474-90	1b n.a.	0,083
1997-474-91	5b n.a.	0,081
1997-474-92	6b n.a.	0,086
1997-474-93	7a n.a.	0,063
1997-474-94	8b n.a.	0,064
1997-474-95	9a n.a.	0,065

Med hilsen
SINTEF Kjemi


Nina Gjøs
Laboratorieleder
Miljøteknologi og analyse


Ingegerd Rustad
Prosjektleder

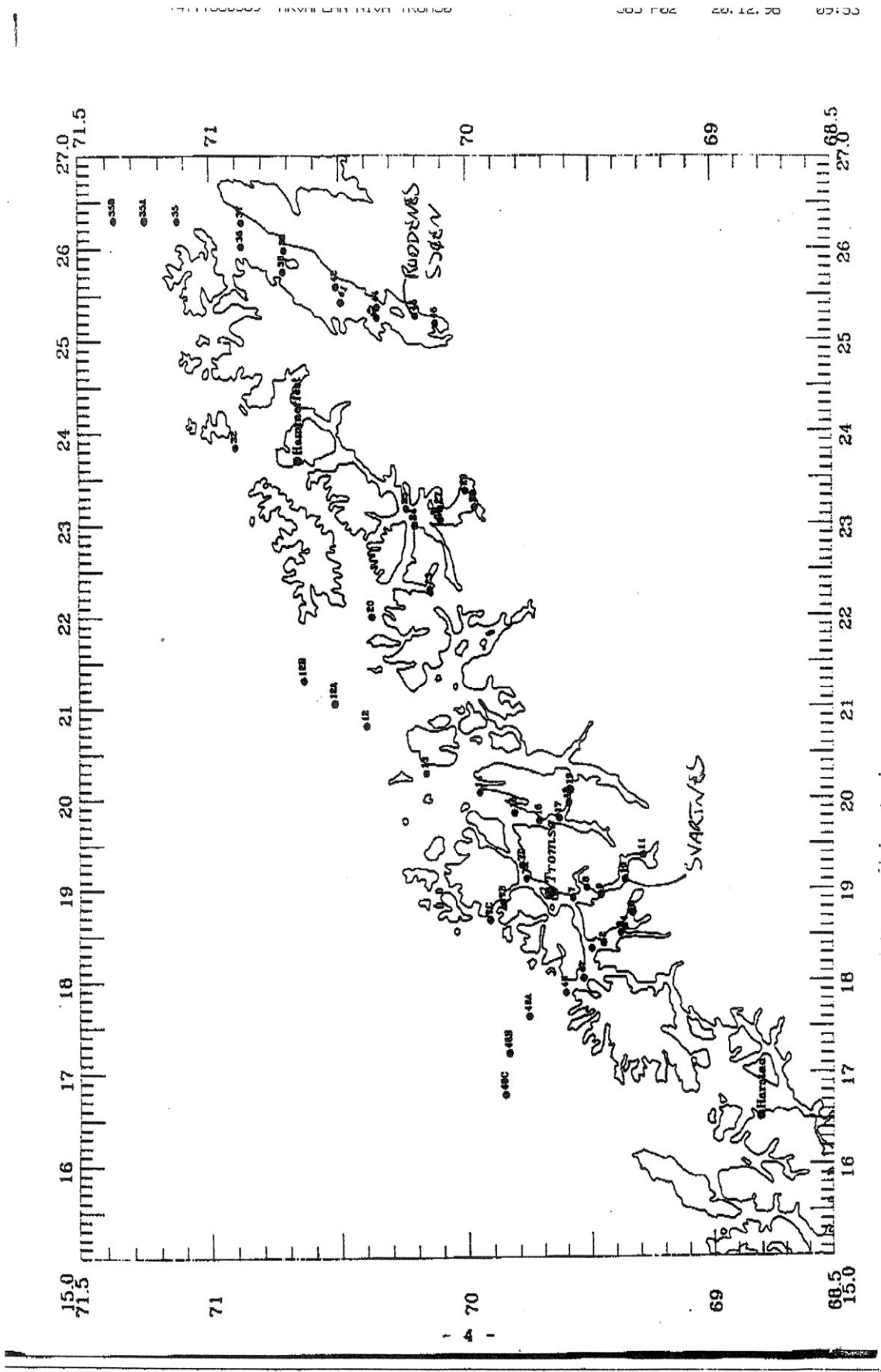
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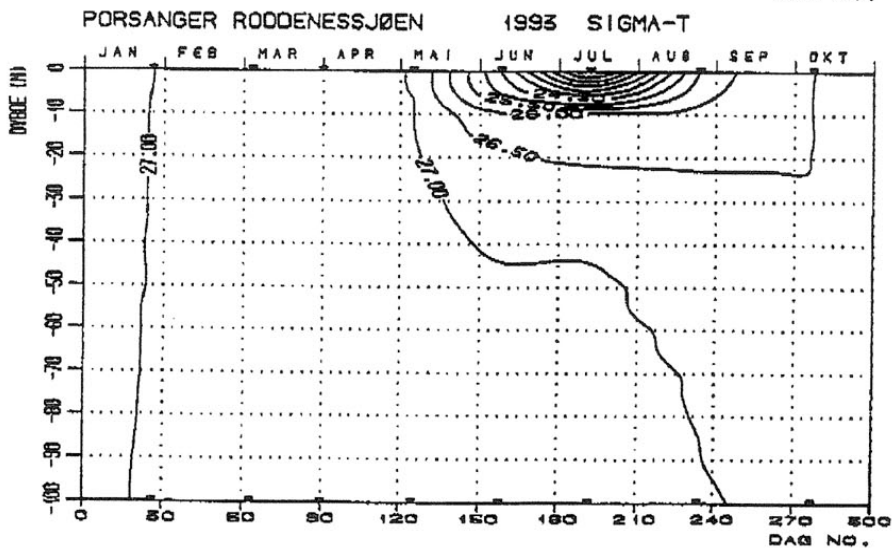
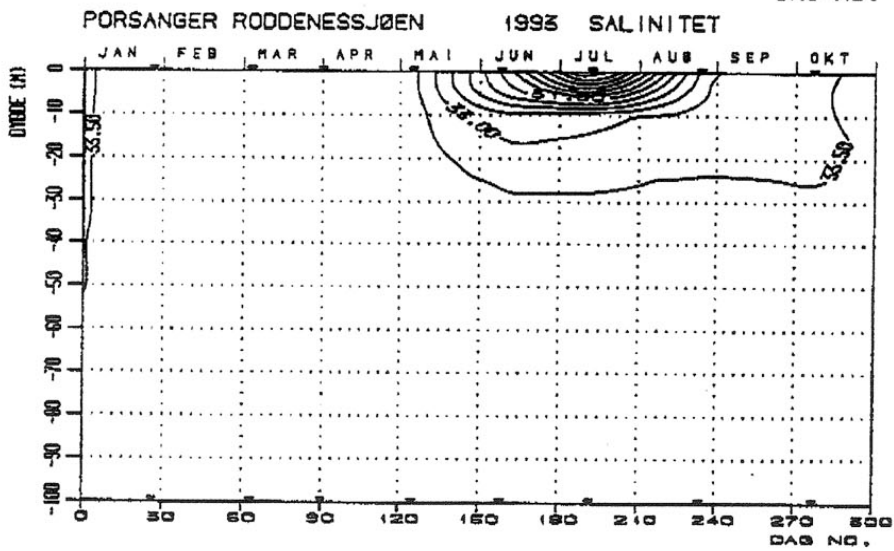
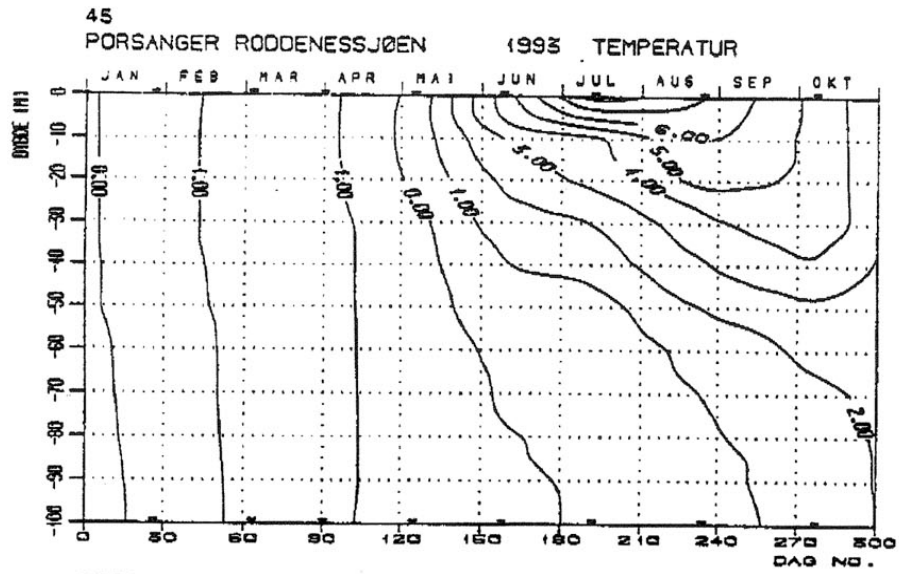
Resterende prøvemateriale oppbevares på SINTEF Industriell kjemi i 6 måneder etter at oppdraget er utført om ikke annet avtales med oppdragsgiver. Analyseresultater rapportert i dette dokument er frembragt ved analyse av de anførte prøver i den stand de ble mottatt ved SINTEFs analyselaboratorium. SINTEF tar intet ansvar for oppdragsgivers bruk av resultatene eller for konsekvenser av slik bruk. *Delvis* kopiering av denne rapport er ikke tillatt uten skriftlig samtykke fra SINTEF.

APPENDIX 3

SEASONAL VARIATIONS OF TEMPERATURE AND SALINITY IN RODDENESSJØEN IN 1993

Source: University of Tromsø, Havmiljødata
(<http://lupus.nfh.uit.no>)





APPENDIX 4

DRILLING FLUID FORMULATIONS AND TEST RESULTS

Laboratory report. 3pp.

Anchor/M-I Drilling Fluids A.S



Anchor Drilling Fluids a.s
Gamle Forusvei 43
Phone +47 51 57 73 00
Fax +47 51 57 06 05

Eastern Hemisphere Technology Centre

Laboratory Report

Lab Job Reference : LJ1103

Lab Request Reference : LR-052-97 HH

Issued to : Harry Hoset
Issued by : Anne Myrvold
Approved by : Wray Curtis *A.W.C.*

Date issued: 12 th. March 1997

Background : Saga Petroleum / NIVA want to test two different mud systems, using ester (Eco Green) and LAO (Nova Tec) as baseoils, for biodegradation.

Objectives: To mix one litre of each according to standard formulation at 1,60 SG and with an oil/water ratio at 75/25.

Checked rheology after mixing.

Specification: 3 rpm \pm 10.

The samples are to be sent to NIVA - Oslo (attn. Morten Schaanning).

Enclosures : Drilling fluid formulation and test results (2)

Discussion: The ester - based mud got high 3 rpm readings, due to the high water content in the mud (75/25). This result is not believed to affect the result of the biodegradation test.

Conclusion: To be completed by originator.

Anchor Drilling Fluids

DRILLING FLUID FORMULATIONS AND TEST RESULTS

SAMPLE DETAILS

FLUID SYSTEM	NOVA TEC
DESCRIPTION	Sample for biodegradation test (NIVA)
TEST DATE	9 th. march 1997
TESTED BY	Anne Myrvold
SAMPLE VOLUME	1000 ml

SAMPLE COMPOSITION

SAMPLE NUMBER

PRODUCT	UNITS	1													
Nova Tec B	ml	544,7													
VG-69	g	8													
Lime	g	28,6													
Nova mul IO	ml	28,6													
Water	ml	182,1													
CaCl2	g	59,7													
Barite	g	874,7													
Novamod IO	ml	2,5													
PERIOD AGED	HOURS														
TEMPERATURE	°C														
DYNAMIC/STATIC	D/S														
RHEOLOGY:	TEMP.	°C	50												
600 RPM			52												
300 RPM			34												
200 RPM			26												
100 RPM			19												
6 RPM			9												
3 RPM			8												
GELS 10"	Pa		5,5												
GELS 10'	Pa		8												
APPARENT VISC.	m Pa s		26												
PLASTIC VISC.	m Pa s		18												
YIELD POINT	Pa		8												
FILTRATION															
TEMPERATURE	°C														
DELTA PRESSURE	psi														
HTHP FLUID LOSS	mls														
WATER IN FILTRATE	mls														
OIL / WATER RATIO	% / %		75/25												
ELECTRIC STABILITY	VOLTS		755												
SPECIFIC GRAVITY			1,6												
COMMENTS:															

Anchor/M-I Drilling Fluids

DRILLING FLUID FORMULATIONS AND TEST RESULTS

SAMPLE DETAILS

FLUID SYSTEM	ECO GREEN
DESCRIPTION	Sample for biodegradation test (NIVA)
TEST DATE	9 th. march 1997
TESTED BY	Anne Myrvold
SAMPLE VOLUME	1000 ml

SAMPLE COMPOSITION

SAMPLE NUMBER

PRODUCT	UNITS	1															
Eco Green B	ml	553,3															
Eco Green P	ml	20															
Eco Green S	ml	12															
Eco Green F	g	4															
Eco Green Vis	g	3															
Water	ml	193,1															
CaCl2	g	28,9															
Barite	g	861,8															
PERIOD AGED	HOURS																
TEMPERATURE	°C																
DYNAMIC/STATIC	D/S																
RHEOLOGY:	TEMP.	°C	50														
600 RPM			151														
300 RPM			96														
200 RPM			74														
100 RPM			51														
6 RPM			32														
3 RPM			30														
GELS 10"		Pa	16,5														
GELS 10'		Pa	19,5														
APPARENT VISC.		m Pa s	75,5														
PLASTIC VISC.		m Pa s	55														
YIELD POINT		Pa	20,5														
FILTRATION																	
TEMPERATURE		°C															
DELTA PRESSURE		psi															
HTHP FLUID LOSS		mls															
WATER IN FILTRATE		mls															
OIL / WATER RATIO		% / %	75/25														
ELECTRIC STABILITY		VOLTS	907														
SPECIFIC GRAVITY			1,6														
COMMENTS:																	

APPENDIX 5
MACROBENTHOS SPECIES LISTS

Species and number of individuals in the Oslofjord samples

GRUPPENAVN	FAMILIENAVN	ARTSNAVN	1A	1B	1C	1D	2A	2B	2C	2D	3A	3B	3C	3D
ANTHOZOA		Anthozoa indet	1			2	1		2	2	2		1	1
ANTHOZOA		Pennatulacea indet												1
ANTHOZOA	Cerianthidae	Cerianthus lloydi Gosse	8	31	9	27	26	25	42	39	21		15	19
NEMERTINEA		Nemertinea indet	4	11	3	4	1	6		2	6		4	4
POLYCHAETA	Amphinomidae	Paramphionome jeffreysii (McIntosh 1868)												
POLYCHAETA	Polynoidae	Harmothoe sp	3	2		1		1	1	1	2		2	1
POLYCHAETA	Sigalionidae	Leanira tetragona (Oersted 1844)					1			1				
POLYCHAETA	Sigalionidae	Pholoe anoculata Hartmann 1965												
POLYCHAETA	Sigalionidae	Pholoe minuta (Fabricius 1780)												
POLYCHAETA	Phyllodoctidae	Phyllodoctidae indet						1						
POLYCHAETA	Hesionidae	Gyptis rosea (Malm 1874)							1					
POLYCHAETA	Hesionidae	Kefersteinia cirrata (Keferstejn 1862)												
POLYCHAETA	Hesionidae	Nereimyra punctata (O.F.Mueller 1788)	1	3	1	2	13	3	10					3
POLYCHAETA	Pilargidae	Synelmis klatti (Friedrich 1950)												
POLYCHAETA	Nereidae	Ceratocephale loveni Malmgren 1867	1	1	1	1	1	8	3	12	2		1	1
POLYCHAETA	Onuphidae	Onuphis fiordica Fauchald 1974												
POLYCHAETA	Onuphidae	Onuphis quadricuspis M.Sars 1872					1				1		1	1
POLYCHAETA	Lumbrineridae	Lumbrineris sp								1				
POLYCHAETA	Orbinidae	Scoloplos armiger (O.F.Mueller 1776)	1											
POLYCHAETA	Paraonidae	Paradoneis lyra (Southern 1914)			1	1	2	3	1	4			3	1
POLYCHAETA	Paraonidae	Paraonis gracilis (Tauber 1879)			1	1	2	2	1	2	1			
POLYCHAETA	Spionidae	Prionospio cirrifera Wiren 1883	1										2	
POLYCHAETA	Spionidae	Pseudopolydora sp				2					1		1	
POLYCHAETA	Spionidae	Spiophanes kroeyeri Grube 1860	2	1	2	2	2	1		2	3		6	9
POLYCHAETA	Cirratulidae	Caulleriella sp	2	1		3					1			
POLYCHAETA	Cirratulidae	Chaetozone setosa Malmgren 1867												3
POLYCHAETA	Cirratulidae	Tharyx sp	1								2			
POLYCHAETA	Cossuridae	Cossura longocirrata Webster & Benedict							1					
POLYCHAETA	Flabelligeridae	Brada sp	1											
GRUPPENAVN	FAMILIENAVN	ARTSNAVN	1A	1B	1C	1D	2A	2B	2C	2D	3A	3B	3C	3D

GRUPPENAVN	FAMILIENAVN	ARTSNAVN	1 A	1 B	1 C	1 D	2 A	2 B	2 C	2 D	3 A	3 B	3 C	3 D
BIVALVIA	Thyasiridae	Thyasira ferruginea (Forbes)	3	3	1	10	27	20	31	17	19		18	12
BIVALVIA	Thyasiridae	Thyasira obsoleta (Verrill & Bush)		2		5	3	8	10	8	3		7	6
BIVALVIA	Thyasiridae	Thyasira pygmaea (Verrill & Bush)		8	4	22	8	13	23	16	3		2	
BIVALVIA	Thyasiridae	Thyasira sarsi (Philippi 1845)		3	2	9	1		4					
BIVALVIA	Lasaeidae	Montacuta ferruginosa (Montagu 1803)												
BIVALVIA	Astartidae	Astarte elliptica Brown 1827						1		2	3		3	4
BIVALVIA	Cardiidae	Parvicardium minimum (Philippi 1836)	2	1		3								
BIVALVIA	Tellinidae	Macoma calcarea (Gmelin 1790)												
BIVALVIA	Scrobiculariidae	Abra nitida (Mueller 1789)		3	1	6				2	5		9	1
BIVALVIA	Kelliellidae	Kelliella miliaris (Philippi 1844)	1	2	2	22	1	2	15	18			4	1
BIVALVIA	Cuspidariidae	Cuspidaria obesa (Loven 1846)				2								2
OSTRACODA	Cypridimidae	Philomedes liljeborgi G.O.Sars				1		7		7	1			
OSTRACODA	Cypridae	Macrocypis minna (Baird)												
CUMACEA	Diastylidae	Leptostylis cf. longimana G.O.Sars												
TANAIDACEA		Tanaidacea indet		1										
AMPHIPODA	Melittidae	Ertopisa elongata Bruzeius	2	1	1	2	1	3		4	1			
DECAPODA		Decapoda indet												
SIPUNCULIDA		Golfingia sp												
SIPUNCULIDA		Onchnesoma steenstrupi Koren & Danielssen	1	2			2	1		2	4		6	4
SIPUNCULIDA		Phascoscolion strombi (Montagu 1804)												
SIPUNCULIDA		Sipunculida indet												
OPHIUROIDEA	Amphilepididae	Amphilepis norvegica Ljungman												
OPHIUROIDEA	Ophiuridae	Ophiura sp												1
ECHINOIDEA	Brissidae	Brissopsis lyrifera (Forbes)												
ASCIDIACEA		Ascidiacea indet		4										
VARIA		Ubestemt indet												
VARIA		Vermiformis indet												1
ANTHOZOA		Anthozoa indet												
ANTHOZOA		Pennatulacea indet												1
ANTHOZOA	Cerianthidae	Cerianthus lloydi Gosse	1	1										1

GRUPPENAVN	FAMILIENAVN	ARTSNAVN	4A	4B	4C	4D	5A	5B	5C	5D	6A	6B	6C	6D
NEMERTINEA		Nemertinea indet	10	14	8	17	23	7	12	11	9	6	14	8
POLYCHAETA	Amphinomidae	Paramphinoe jeffreysii (McIntosh 1868)	1	7	3	6	8	7	2	2	4	4	3	3
POLYCHAETA	Polynoidae	Harmothoe sp				1		1		1		2		1
POLYCHAETA	Sigalionidae	Leanira tetragona (Oersted 1844)	2	1	1			1						2
POLYCHAETA	Sigalionidae	Pholoe anoculata Hartmann 1965	1											2
POLYCHAETA	Sigalionidae	Pholoe minuta (Fabricius 1780)		1										
POLYCHAETA	Phyllodoceidae	Phyllodoceidae indet												
POLYCHAETA	Hesionidae	Gyptis rosea (Malm 1874)												
POLYCHAETA	Hesionidae	Kefersteimia cirrata (Keferstein 1862)	4	1	4	4	2	2	4	5	3	1	4	4
POLYCHAETA	Hesionidae	Nereimyra punctata (O.F.Mueller 1788)		1										
POLYCHAETA	Pilargiidae	Synelmis klatti (Friedrich 1950)	7	5	10	3		3	2					2
POLYCHAETA	Nereidae	Ceratocephale loveni Malmgren 1867					4	1	1					
POLYCHAETA	Onuphidae	Onuphis frordica Fauchald 1974			1			4	3	2				
POLYCHAETA	Onuphidae	Onuphis quadricuspis M.Sars 1872		1		1					1			
POLYCHAETA	Lumbrineridae	Lumbrineris sp												
POLYCHAETA	Orbiniidae	Scoloplos armiger (O.F.Mueller 1776)								1				
POLYCHAETA	Paronidae	Paradoneis lyra (Southern 1914)	2	3	1			3		1	1	2	1	1
POLYCHAETA	Paronidae	Paraonis gracilis (Tauber 1879)									1			1
POLYCHAETA	Spionidae	Prionospio cirrifera Wiren 1883	1	1										2
POLYCHAETA	Spionidae	Pseudopolydora sp	1	1	1									1
POLYCHAETA	Spionidae	Spiophanes kroeyeri Grube 1860	4	4	6	3	1	1		1	1	1	1	4
POLYCHAETA	Cirratulidae	Cauleriella sp			1		1	4	2	2				1
POLYCHAETA	Cirratulidae	Chaetozone setosa Malmgren 1867	1	1	2	1					2			1
POLYCHAETA	Cirratulidae	Tharyx sp												
POLYCHAETA	Cossuridae	Cossura longocirrata Webster & Benedict			2									1
POLYCHAETA	Flabelligeridae	Brada sp												
POLYCHAETA	Flabelligeridae	Brada villosa (Rathke 1843)					1			1				
POLYCHAETA	Flabelligeridae	Pherusa sp			1									
POLYCHAETA	Capitellidae	Heteromastus filiformis (Claparede 1864)	96	88	58	105	1	5	2	5	5	4	21	13
POLYCHAETA	Maldanidae	Euclymeninae indet			2									

GRUPPENAVN	FAMILIENAVN	ARTSNAVN	4 A	4 B	4 C	4 D	5 A	5 B	5 C	5 D	6 A	6 B	6 C	6 D
POLYCHAETA	Maldanidae	Rhodine gracilior Tauber 1879	1											
POLYCHAETA	Maldanidae	Rhodine loveni Malmgren 1865			2	1							1	2
POLYCHAETA	Oweniidae	Myriochele oculata Zaks 1922	1	1	2	1								
POLYCHAETA	Ampharetidae	Amage auricula Malmgren 1865							1	1			1	1
POLYCHAETA	Ampharetidae	Melinna cristata (M.Sars 1851)		3	1	1		2					1	1
POLYCHAETA	Ampharetidae	Melythasides laubieri Desbruyeres 1978								1				
POLYCHAETA	Ampharetidae	Mugga wahrbergi Eliason 1955												
POLYCHAETA	Ampharetidae	Sosane sulcata Malmgren 1865		1	2									
POLYCHAETA	Terebellidae	Proclea graffii (Langerhans 1884)			2									
POLYCHAETA	Trichobranthidae	Terebellides stroemi M.Sars 1835	1		1	2				2				2
POLYCHAETA	Trichobranthidae	Trichobranthus roseus (Malm 1874)												
POLYCHAETA	Sabellidae	Euchone sp		1	1	1							1	
POLYCHAETA	Sabellidae	Sabellidae indet	3	10	7		1			1	3	3		4
POLYCHAETA	Serpulidae	Serpula vermicularis Linne 1767	1										2	4
OLIGOCHAETA		Oligochaeta indet		2	1					2			2	4
OPISTHBRANCHIA		Nudibranchia indet												1
OPISTHBRANCHIA		Opisthobranchia indet	1	6						2				3
CAUDOFOVEATA		Caudofoveata indet	1		1	4	2							
BIVALVIA	Nuculidae	Nucula sulcata (Bronn 1831)												
BIVALVIA	Nuculidae	Nucula tumidula (Malm)	4	17	5	3	15	18	7	27	3		1	1
BIVALVIA	Nuculidae	Nucula turgida Leckenby & marshall		1		1	5							1
BIVALVIA	Nuculidae	Nuculoma tenuis (Montagu)	7	21	13	11		2	1	1	4	3	11	6
BIVALVIA	Nuculanidae	Yoldiella lucida (Loven 1846)	5	1	1									1
BIVALVIA	Nuculanidae	Yoldiella sp												
BIVALVIA	Pectinidae	Delectopecten vitreus (Gmelin 1789)								3				
BIVALVIA	Thyasiridae	Thyasira croulinensis (Jeffreys)											1	
BIVALVIA	Thyasiridae	Thyasira equalis (Verrill & Bush)	120	176	101	73	15	12	5	9	32	30	31	50
BIVALVIA	Thyasiridae	Thyasira ferruginea (Forbes)	9	20	7	9	18	2	8	11	6		2	7
BIVALVIA	Thyasiridae	Thyasira obsoleta (Verrill & Bush)	3	13	1	3	5		3	2	3	1	2	2
BIVALVIA	Thyasiridae	Thyasira pygmaea (Verrill & Bush)	16	29	11	15	8	2	10	5	1		3	11

GRUPPENAVN	FAMILIENAVN	ARTSNAVN	4 A	4 B	4 C	4 D	5 A	5 B	5 C	5 D	6 A	6 B	6 C	6 D
BIVALVIA	Thyasiridae	Thyasira sarsi (Philippi 1845)												
BIVALVIA	Lasaeidae	Montacuta ferruginosa (Montagu 1803)												
BIVALVIA	Astartidae	Astarte elliptica Brown 1827		1		1								1
BIVALVIA	Cardiidae	Parvicardium minimum (Philippi 1836)			1									
BIVALVIA	Tellinidae	Macoma calcarea (Gmelin 1790)	1											
BIVALVIA	Scrobiculariidae	Abra nitida (Mueller 1789)	20	12	19	1				9	4	1		8
BIVALVIA	Kelliellidae	Kelliella militaris (Philippi 1844)	2	10	2	3	7	4	10	4	2	3	1	2
BIVALVIA	Cuspidariidae	Cuspidaria obesa (Loven 1846)		1										
OSTRACODA	Cypridiniidae	Philomedes liljeborgi G.O.Sars	2		7			1						
OSTRACODA	Cypridae	Macrocypis minna (Baird)												
CUMACEA	Diastylidae	Leptostylis cf. longimana G.O.Sars												
TANAIDACEA		Tanaidacea indet												
AMPHIPODA	Melitidae	Eriopisa elongata Bruzelius	1		1					3	1	4		3
DECAPODA		Decapoda indet						1						
SIPUNCULIDA		Golfingia sp												
SIPUNCULIDA		Onchomesoma steenstrupi Koren & Danielssen	1		1	1		2	2	2	1	6	5	3
SIPUNCULIDA		Phascolion strombi (Montagu 1804)						1						
SIPUNCULIDA		Sipunculida indet								1				
OPHIUROIDEA	Amphilepididae	Amphilepis norvegica Ljungman												2
OPHIUROIDEA	Ophiuridae	Ophiura sp												
ECHINOIDEA	Brissidae	Brissopsis lyrifera (Forbes)												
ASCIDIACEA		Asciacea indet												1
VARIA		Ubestemt indet												
VARIA		Vermiformis indet	1											

Species and number of individuals in the Porsangerfjord samples

Gruppe	Familie	Art	7A	7B	7C	7D	8A	8B	8C	8D	9A	9B	9C	9D	12A	12B	12C	12D
HYDROZOA		Hydrozoa indet																1
NEMERTINEA		Nemertinea indet	2	1	1	1	3	1	4	1	2	2	2	2				3
POLYCHAETA		Polychaeta indet	1															1
POLYCHAETA	Polynoidae	Harmothoe sp			1					1								1
POLYCHAETA	Hesionidae	Kefersteinia cirrata (Keferstein 1862)										1						
POLYCHAETA	Syllidae	Syllidae indet										1						
POLYCHAETA	Nephtyidae	Aglaophamus cf. malmgreni Theel 1879	1	1	4	1	3	7	3	2	4	2	2	2	3			4
POLYCHAETA	Nephtyidae	Nephtys sp			1													3
POLYCHAETA	Nephtyidae	Nephtys ciliata (O.F.Mueller 1776)	1	1	1	1	2	1	1	1	1	2	1	2	1			1
POLYCHAETA	Nephtyidae	Nephtys paradoxa Malm 1874										1						1
POLYCHAETA	Lumbrineridae	Lumbrineris sp	8	9	12	14	7	6	5	3	8	8	7	4	2	3	5	4
POLYCHAETA	Dorvilleidae	Dorvilleidae indet		1														
POLYCHAETA	Orbiniidae	Scoloplos armiger (O.F.Mueller 1776)								2								
POLYCHAETA	Apistobranchidae	Apistobranchus tullbergi (Theel 1879)	1	2							1	1	1	1	1			1
POLYCHAETA	Paraonidae	Aricidea sp				1						1						
POLYCHAETA	Paraonidae	Paraonis gracilis (Tauber 1879)		1														1
POLYCHAETA	Chaetopterididae	Spiochaetopterus typicus M.Sars 1856	44	44	71	74	63	69	73	80	58	64	40	59	25	35	25	38
POLYCHAETA	Cirratulidae	Cauleriella sp																
POLYCHAETA	Cirratulidae	Chaetozone setosa Malmgren 1867	1	1	2	3	4	4	4	2	1	1	1	1	1	1	1	1
POLYCHAETA	Cirratulidae	Cirratulidae indet			1							1	1	1				1
POLYCHAETA	Cossuridae	Cossura longocirrata Webster & Benedict										1						1
POLYCHAETA	Opheliidae	Ophelina acuminata Oersted 1843		1				1	1	1	1							
POLYCHAETA	Capitellidae	Heteromastus filiformis (Claparede 1864)	1	1	3	1	1	2	2	2	2	1	1	1	1	1		3
POLYCHAETA	Maldanidae	Euclymeninae indet	1	1	3	2	1	4	1	3	2	2	1	1	1			2
POLYCHAETA	Maldanidae	Maldane sarsi Malmgren 1865	1	3	3			3	2	1	3	4	1	1	2	1		4
POLYCHAETA	Maldanidae	Maldanidae indet																1
POLYCHAETA	Oweniidae	Myriochele oculata Zaks 1922	8	13	6	7	2	9	11	7	6	7	4	9	2	3	9	5

Gruppe	Familie	Art	7A	7B	7C	7D	8A	8B	8C	8D	9A	9B	9C	9D	12A	12B	12C	12D
POLYCHAETA	Pectinariidae	<i>Pectinaria hyperborea</i> (Malmgren 1865)	4	1	1	1	4	2	1	3	7	2			1	3	1	1
POLYCHAETA	Ampharetidae	<i>Sabellides borealis</i> M.Sars 1856					2	1							2			1
POLYCHAETA	Terebellidae	<i>Amphitritinae</i> indet																
POLYCHAETA	Terebellidae	<i>Laphania boeckii</i> Malmgren 1866																1
POLYCHAETA	Terebellidae	<i>Neoamphitrite groenlandica</i> Malmgren																
POLYCHAETA	Terebellidae	<i>Polycirrus</i> sp																
POLYCHAETA	Trichobranchidae	<i>Terebellides stroemi</i> M.Sars 1835	1				2	1	2	1	2	2		3			3	1
POLYCHAETA	Sabellidae	<i>Sabellidae</i> indet	1	4				1										1
PROSOBRANCHIA	Turridae	<i>Oenopota violacea</i> (Mighels & Adams)																2
CAUDOFOVEATA		<i>Caudofoveata</i> indet	4															
BIVALVIA	Nuculidae	<i>Nuculoma corticata</i> (Moeller)	2				1				2		1					
BIVALVIA	Nuculanidae	<i>Nuculana permula</i> (Mueller 1776)	2			1		1										
BIVALVIA	Nuculanidae	<i>Yoldia amygdalea</i> Valenciennes 1846	1	4	1	1	2	1	1	1	2	2	1	1	1	1	3	4
BIVALVIA	Nuculanidae	<i>Yoldiella fraterna</i> Verrill & Bush					1					1					1	1
BIVALVIA	Nuculanidae	<i>Yoldiella lenticula</i> (Mueller 1842)	4	8	9	15	6	5	8	10	5	6	4	2	4	6	1	3
BIVALVIA	Nuculanidae	<i>Yoldiella</i> sp				2	3	1			2	1						
BIVALVIA	Pectinidae	<i>Chlamys cf. tigrina</i> (Mueller)	1															
BIVALVIA	Thyasiridae	<i>Thyasira equalis</i> (Verrill & Bush)			1	1	1	1	1	1	1	1	3	1	1		2	2
BIVALVIA	Thyasiridae	<i>Thyasira pygmaea</i> (Verrill & Bush)				1	1						1					
SCAPHOPODA	Siphonodontalitiidae	<i>Siphonodontalium lobatum</i> (G.B.Sowerby II)												1				
CUMACEA	Leuconidae	<i>Eudorella emarginata</i> Kroeyer	1	1	1	4			1	1	1	1						1
CUMACEA	Leuconidae	<i>Leucon nasica</i> (Kroeyer)				1												
CUMACEA	Diastylidae	<i>Brachydiastylis resima</i> (Kroeyer 1846)					1	1	1	1	1	1			1		2	
AMPHIPODA	Ampeliscidae	<i>Haploops tubicola</i> Lijeborg																
DECAPODA	Hippolytidae	<i>Eualus gaimardii</i> (H.Milne Edwards)																
ASTEROIDEA	Goniopectinidae	<i>Ctenodiscus crispatus</i> (Bruz.)	5	1	1	1	1	1	3	1	3	3			3	4	3	4
ASCIDIACEA		<i>Ascidiacea</i> indet																1
VARIA		<i>Ubestem</i> indet																1
VARIA		<i>Vermiformis</i> indet																1

APPENDIX 6.
ANALYSES OF SEDIMENT SAMPLES

Appendix Table: Analyses of initial sediments samples.

IDENTIFICATION				ANALYSES				CALCULATED PARAMETERS			
Date	Section	Tr.ment	Depth	Sample g w.wght	Dry wght. %	Ba mg/g	OP mg/g	OP mg/cm2	Excess Ba mg/g	Exc. Ba mg/cm2	OP:exc.Ba ratio
14.04.97	1b	EC	0-3 cm	22.62	25.1	na	0.000	0.000	-	-	-
14.04.97	1d	EM	0-3 cm	22.79	23.9	3.79	4.254	4.377	3.005	3.092	1.416
14.04.97	2b	OL	0-3 cm	22.70	23.9	1.31	0.310	0.318	0.525	0.538	0.591
14.04.97	3a	OL	0-3 cm	23.10	26.2	1.39	0.235	0.268	0.605	0.690	0.388
14.04.97	4a	EM	0-3 cm	23.13	27.8	3.37	3.467	4.208	2.585	3.138	1.341
14.04.97	5b	OC	0-3 cm	22.46	21.9	na	0.000	0.000	-	-	-
14.04.97	5d	OL	0-3 cm	22.16	22.3	1.59	0.408	0.381	0.805	0.752	0.507
14.04.97	6b	EC	0-3 cm	23.54	28.9	na	0.000	0.000	-	-	-
14.04.97	6c	EM	0-3 cm	15.98	27.9	3.70	4.288	5.412	2.915	3.679	1.471
14.04.97	7a	OC	0-3 cm	24.88	29.0	na	0.000	0.000	-	-	-
14.04.97	7c	OM	0-3 cm	24.65	29.4	3.35	2.496	3.411	2.715	3.711	0.919
14.04.97	8b	EC	0-3 cm	23.62	27.0	na	0.015	0.018	-	-	-
14.04.97	8c	EL	0-3 cm	23.32	31.6	1.14	0.638	0.888	0.505	0.703	1.263
14.04.97	9a	OC	0-3 cm	22.47	25.8	na	0.000	0.000	-	-	-
14.04.97	9d	OM	0-3 cm	22.12	26.3	4.67	4.483	4.925	4.035	4.433	1.111
14.04.97	10d	EL	0-3 cm	24.23	26.0	0.83	0.225	0.268	0.195	0.232	1.156
14.04.97	11d	OM	0-3 cm	24.28	27.5	3.55	2.874	3.620	2.915	3.671	0.986
14.04.97	12d	EL	0-3 cm	23.90	25.6	0.89	0.344	0.396	0.255	0.294	1.347
<i>Core samples</i>											
14.04.97	1c-core	EH	0-3 cm	-	-	-	-	-	-	14.722	-
14.04.97	11b-core	OH	0-3 cm	-	-	-	-	-	-	12.643	-
14.04.97	1b-core	EC	0-3 cm	-	-	-	-	-	-	-0.017	-
14.04.97	11a-core	OC	0-3 cm	-	-	-	-	-	-	-0.044	-
14.04.97	1c1	EH	0-1 cm	3.86	26.4	38.20	-	-	37.415	14.186	-
14.04.97	1c2	EH	1-3 cm	6.96	29.7	1.48	-	-	0.695	0.536	-
14.04.97	1c3	EH	3-5 cm	7.52	34.7	0.93	-	-	0.145	0.141	-
14.04.97	1c4	EH	5-7 cm	5.84	37.7	0.85	-	-	0.065	0.053	-
14.04.97	11b1	OH	0-1 cm	4.00	21.6	38.80	-	-	38.165	12.265	-
14.04.97	11b2	OH	1-3 cm	6.34	33.0	1.12	-	-	0.485	0.378	-
14.04.97	11b3	OH	3-5 cm	6.78	36.5	0.79	-	-	0.155	0.143	-
14.04.97	11b4	OH	5-7 cm	7.89	43.7	0.68	-	-	0.045	0.058	-
14.04.97	1b1	EC	0-1 cm	3.20	26.6	0.74	-	-	-0.045	-0.014	-
14.04.97	1b2	EC	1-3 cm	6.62	25.6	0.78	-	-	-0.005	-0.003	-
14.04.97	1b3	EC	3-5 cm	6.76	30.3	0.82	-	-	0.035	0.027	-
14.04.97	1b4	EC	5-7 cm	7.32	35.7	0.80	-	-	0.015	0.015	-
14.04.97	11a1	OC	0-1 cm	3.61	22.8	0.59	-	-	-0.045	-0.014	-
14.04.97	11a2	OC	1-3 cm	6.63	34.5	0.60	-	-	-0.035	-0.030	-
14.04.97	11a3	OC	3-5 cm	7.54	38.8	0.67	-	-	0.035	0.038	-
14.04.97	11a4	OC	5-7 cm	6.66	46.0	0.68	-	-	0.045	0.051	-

Appendix Table: Analyses of final sediments samples.

-----IDENTIFICATION-----				-----ANALYSES-----				-----CALCULATED PARAMETERS-----			
Date	Section	Tr.ment	Depth	Sample g w.wght	Dry wght. %	Ba mg/g	OP mg/g	OP mg/cm2	Excess Ba mg/g	Exc. Ba mg/cm2	OP:exc.Ba ratio
03.07.97	1b	EC	0-3 cm	23.432	23.052	0.830	na	-	0.045	0.046	-
03.07.97	1a	EL	0-3 cm	22.511	22.783	1.460	0.000	0.000	0.675	0.653	0.000
03.07.97	1c	EH	0-3 cm	24.098	24.959	11.550	12.490	14.178	10.765	12.219	1.160
03.07.97	1d	EM	0-3 cm	22.924	22.957	4.010	1.801	1.789	3.225	3.203	0.558
03.07.97	2a	OM	0-3 cm	23.168	22.481	4.710	1.708	1.679	3.925	3.858	0.435
03.07.97	2b	OL	0-3 cm	23.433	21.948	1.240	0.000	0.000	0.455	0.442	0.000
03.07.97	2c	OH	0-3 cm	24.231	23.628	15.860	16.712	18.058	15.075	16.288	1.109
03.07.97	2d	OC	0-3 cm	23.448	22.214	0.820	0.000	0.000	0.035	0.034	0.000
03.07.97	3a	OL	0-3 cm	23.926	24.173	1.310	0.000	0.000	0.525	0.573	0.000
03.07.97	3b	OM	0-3 cm	23.809	25.528	3.790	1.640	1.881	3.005	3.447	0.546
03.07.97	3c	OC	0-3 cm	24.000	26.174	0.810	0.000	0.000	0.025	0.030	0.000
03.07.97	3d	OH	0-3 cm	24.651	28.801	10.330	9.652	12.932	9.545	12.789	1.011
03.07.97	4a	EM	0-3 cm	24.421	27.790	2.620	0.784	1.004	1.835	2.350	0.427
03.07.97	4b	EC	0-3 cm	23.752	22.915	0.880	0.000	0.000	0.095	0.098	0.000
03.07.97	4c	EL	0-3 cm	24.387	26.785	1.130	0.005	0.006	0.345	0.425	0.014
03.07.97	4d	EH	0-3 cm	24.600	25.791	9.360	8.318	9.960	8.575	10.268	0.970
03.07.97	5a	OM	0-3 cm	23.604	18.319	4.520	2.363	1.929	3.735	3.048	0.633
03.07.97	5b	OC	0-3 cm	23.453	20.022	0.840	na	-	0.055	0.049	-
03.07.97	5c	OH	0-3 cm	23.810	22.328	13.600	12.124	12.164	12.815	12.857	0.946
03.07.97	5d	OL	0-3 cm	23.359	19.999	1.380	0.000	0.000	0.595	0.525	0.000
03.07.97	6a	EL	0-3 cm	24.018	24.386	1.190	0.008	0.008	0.405	0.448	0.019
03.07.97	6b	EC	0-3 cm	25.115	25.218	0.810	na	-	0.025	0.030	-
03.07.97	6c	EM	0-3 cm	24.149	24.483	3.300	1.092	1.219	2.515	2.806	0.434
03.07.97	6d	EH	0-3 cm	24.831	29.561	10.290	9.059	12.549	9.505	13.167	0.953
03.07.97	7a	OC	0-3 cm	24.828	27.485	0.710	na	-	0.075	0.097	-
03.07.97	7b	OL	0-3 cm	25.242	27.599	1.130	0.125	0.164	0.495	0.651	0.252
03.07.97	7c	OM	0-3 cm	25.105	28.348	4.010	3.084	4.142	3.375	4.533	0.914
03.07.97	7d	OH	0-3 cm	23.808	26.637	9.790	9.936	11.892	9.155	10.957	1.085
03.07.97	8a	EM	0-3 cm	25.031	27.907	3.170	1.454	1.916	2.535	3.342	0.573
03.07.97	8b	EC	0-3 cm	25.282	28.145	0.750	na	-	0.115	0.154	-
03.07.97	8c	EL	0-3 cm	32.953	28.063	1.230	0.033	0.043	0.595	0.779	0.056
03.07.97	8d	EH	0-3 cm	25.254	29.957	10.070	10.229	14.604	9.435	13.471	1.084
03.07.97	9a	OC	0-3 cm	25.228	28.144	0.570	na	-	-0.065	-0.087	-
03.07.97	9b	OL	0-3 cm	25.238	28.227	0.940	0.000	0.000	0.305	0.410	0.000
03.07.97	9c	OH	0-3 cm	25.590	30.335	9.030	8.381	12.278	8.395	12.299	0.998
03.07.97	9d	OM	0-3 cm	25.302	29.853	3.940	1.699	2.422	3.305	4.711	0.514
03.07.97	10a	EM	0-3 cm	25.543	31.075	2.950	0.819	1.226	2.315	3.468	0.354
03.07.97	10b	EH	0-3 cm	24.593	29.275	9.610	9.976	13.555	8.975	12.195	1.112
03.07.97	10c	EC	0-3 cm	24.543	26.698	0.750	0.000	0.000	0.115	0.142	0.000
03.07.97	10d	EL	0-3 cm	25.135	28.444	0.890	0.000	0.000	0.255	0.344	0.000
03.07.97	11a	OC	0-3 cm	24.412	25.756	0.730	0.000	0.000	0.095	0.113	0.000
03.07.97	11b	OH	0-3 cm	25.136	29.209	8.260	7.442	10.311	7.625	10.565	0.976
03.07.97	11c	OL	0-3 cm	24.920	26.589	0.980	0.135	0.169	0.345	0.431	0.393
03.07.97	11d	OM	0-3 cm	24.619	25.718	4.600	2.918	3.487	3.965	4.738	0.736
03.07.97	12a	EH	0-3 cm	25.413	28.330	10.210	12.597	17.116	9.575	13.010	1.316
03.07.97	12b	EM	0-3 cm	25.416	29.148	2.930	1.305	1.825	2.295	3.209	0.569
03.07.97	12c	EC	0-3 cm	24.391	29.237	0.650	0.000	0.000	0.015	0.020	0.000
03.07.97	12d	EL	0-3 cm	25.465	28.930	0.860	0.036	0.050	0.225	0.313	0.161
<i>Core samples</i>											
03.07.97	7c	OM	0-3 cm	-	-	-	-	na	0.000	1.105	-
03.07.97	7d	OH	0-3 cm	-	-	-	-	na	0.000	12.786	-
03.07.97	7c	OM	0-1 cm	1.921	23.222	3.970	-	na	3.335	1.109	-
03.07.97	7c	OM	1-3 cm	3.687	32.786	0.630	-	na	-0.005	-0.005	-
03.07.97	7c	OM	3-5 cm	3.986	37.662	0.630	-	na	-0.005	-0.006	-
03.07.97	7c	OM	5-7 cm	2.813	43.191	0.660	-	na	0.025	0.023	-
03.07.97	7d	OH	0-1 cm	1.899	26.411	33.500	-	na	32.865	12.288	-
03.07.97	7d	OH	1-3 cm	3.726	31.749	1.200	-	na	0.565	0.498	-
03.07.97	7d	OH	3-5 cm	3.685	37.160	0.690	-	na	0.055	0.056	-

Appendix Table. Analyses of slurry samples.

Date	Slurry	D. wght %	OP mg/g d.wght.	Ba	OP:Ba ratio
10.04.97	E1	44.62	104.7	72.9	1.436
10.04.97	E2	47.40	101.3	75.3	1.345
10.04.97	E3	46.40	103.9	73.4	1.415
10.04.97	O1	43.68	112.1	84.0	1.334
10.04.97	O2	42.00	117.7	83.8	1.404
10.04.97	O3	42.42	115.1	82.8	1.390