BIOLOGICAL EFFECTS OF WASHED OBM DRILL CUTTINGS DISCHARGED ON THE DUTCH CONTINENTAL SHELF

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This study was commissioned by the North Sea Directorate (RWS) and carried out in 1989

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SUMMARY AND CONCLUSIONS

Since 1985, biological investigations have been carried out on the effects of discharges of contaminated drill cuttings at offshore installations in the Dutch part of the North Sea. This research programme, which is called 'Boorspoeling' (= drilling muds), includes a number of studies on the spatial distribution of drill cuttings, usually contaminated with oil-based muds (OBM), and their effects on the benthic fauna around drill sites. The project is coordinated by the North Sea Directorate of the Dutch Ministry of Transport and Public Works and carried out by TNO (chemical research) and NIOZ (biological research). The present report deals with the biological results of fieldwork and experiments performed in 1989. The chemical results will be published in a TNO- report (VAN HET GROENEWOUD, in prep.).

In 1989 the attention focussed on the location 'L5-5', where an explorative drilling had taken place in the autumn of 1988. During drilling, OBM were utilized and the drill cuttings were washed by means of a recently developed technique, before they were dumped on the seabed. The central question underlying the research programme was to what extent such a washing procedure contributes to a reduction in contamination of the seabed and, thus, to a reduction of adverse effects on the benthic community. Some concrete aims are formulated in the introduction.

Fieldwork took place in May and included sediment sampling at 9 stations around the abandoned drilling location. The results show that the distribution pattern of oil contaminants was quite different from that observed during earlier surveys at drilling locations where no washing procedure was applied. In the close vicinity of the discharge point oil concentrations were much lower. At larger distance, however, still elevated contamination levels were found, even at 5000 m. Hence, a plot of oilconcentrations against increasing distance to the discharge point showed a relatively weak slope, and, consequently, no evident decrease in the intensity of observed effects could be detected along the sampled transect. At first sight, hardly any effects were observed in the area between 250 m and 750 m from the location. The total fauna abundance was rather high in this area. On closer analysis of the distribution patterns of some very sensitive species, however, it appeared that the area of environmental stress extended to over 750 m from the discharge point.

Experimental investigations were conducted on sediment cores, collected in September 1989 at stations at 25 m, 250 m and 5000 m from location L5-5. On these 'boxcosms', which were incubated in the laboratory under semi-natural conditions for 3

months, behaviour and mortality were studied of some introduced test species. The results confirm that the functioning of some test species seemed to be stressed by the presence of oil in the sediment, not only in the 25-m cores, but also in the 250-m cores. Major evidence for this was obtained from mortality rates in *Echinocardium cordatum*.

The present data suggest that, with the application of a washing procedure, a reduction is achieved in the intensity of adverse effects within a few hundred meter from the discharge point. There is, however, no indication that the extent of the area that is subjected to environmental stress is reduced.

The results and conclusions of the 1989 study at L5-5 may be summarized as follows:

- 1. The macrofauna was substantially impoverished only at the nearest station, 25 m from the discharge point. This reduction in macrofauna can only partly be explained by OBM contamination. It is also possible that the macrofauna close to the discharge point could have suffered from smothering, due to the accumulation of cuttings discharged during the first phase of the drilling procedure, when no OBM but WBM were employed.
- 2. At a distance of 250 m the only unmistakable effect of oil pollution observed was the presence of the opportunist Capitella capitata. However, a deviant species assemblage in the area between 250 and 750 m, which became manifest by enhanced densities of some naturally abundant species, might indicate environmental stress.
- 3. It is almost certain that environmental stress stretches over a distance exceeding 750 m. This is indicated by the abundance patterns of 2 commensal species, Montacuta ferruginosa and the sea urchin, Echinocardium cordatum. At least one of these species must be extremely sensitive to OBM contamination, as is explained in detail in the discussion.
- 4. The sensitiveness to OBM of members of the genus Montacuta, or of their host species (sea urchins), provides a starting point for alternative and less time-consuming methods of biological field monitoring in the future. In the analyses of macrofauna samples and in the evaluation of the resulting data, aimed to assess the spatial extent of environmental stress, the attention could be restricted to these sensitive indicator species.
- 5. Echinocardium cordatum functioned appropriately as test species in the boxcosm experiments. Observed lethal and sublethal effects were in agreement with earlier findings on the response of this species to OBM contamination of the sediment.
- Echinocardium suffered considerable mortality in contaminated sediment of both the 25-m and the

- 250-m stations, at oil concentrations ranging from 28 to 935 mg kg⁻¹ dry sediment. In sediment of the 5000-m reference station mortality was almost nil
- 7. In 2 other test species (Nucula turgida and Corystes cassivelaunus) a response was observed, but could not be quantified. A third species (Amphiura filiformis) was present as natural infauna of the boxcosms and occurred in variable numbers in the sediment tested. Hence, mortality among introduced specimens could not be estimated reliably.
- 8. Mortality of the natural infauna in the boxcosms did not provide a suitable comparative criterion in assessing a dose-effect relationship. Estimated mortality rates were not significantly related to oil concentrations in the sediment.
- 9. The present data suggest that, with the application of a washing procedure, a reduction is achieved in the intensity of adverse effects within a few hundred meter from the discharge point. There is, however, no indication that the extent of the area that is subjected to environmental stress is reduced. To get insight in long-term effects of discharges of washed OBM cuttings it will be necessary to carry out one or more follow-up surveys at L5-5.

SAMENVATTING EN CONCLUSIES

Sedert 1985 wordt op de Noordzee, op het Nederlandse deel van het Continentaal Plat, het onderzoeksproject 'Boorspoeling' uitgevoerd. Het programma omvat een aantal ecotoxicologische studies naar de ruimtelijke verspreiding van veelal met olie verontreinigd boorgruis, dat geloosd wordt vanaf boorplatforms, en de effekten daarvan op de bodemfauna rond deze lokaties. Het projekt wordt gecoördineerd door de Direktie Noordzee van het ministerie van Verkeer en Waterstaat en uitgevoerd door het TNO, dat het chemische werk uitvoert, en het NIOZ, dat verantwoordelijk is voor het biologische onderzoek.

In het voorliggende rapport wordt verslag gedaan van het vijfde deel-projekt, dat in 1989 in het kader van 'Boorspoeling' plaats vond. Het gaat hier om de biologische resultaten van veldwerk en experimenten. De chemische resultaten zijn te vinden in een TNO-rapport (VAN HET GROENEWOUD, in prep.).

In 1989 ging de aandacht uit naar het platform 'L5-5', waar tijdens de boring (een half jaar voor de veldsurvey) gebruik gemaakt was van boorvloeistoffen op olie-basis (OBM), en waar het opgeboorde gruis volgens een nieuw ontwikkelde methode een wassing had ondergaan, alvorens het op de zeebodem geloosd werd. De vraagstelling die bij het onderzoek centraal stond was in hoeverre een dergelijke wasprocedure bijdraagt tot een verminderde belasting van de zeebodem met verontreinigd materiaal en tot een reduktie van negatieve effekten op de bodemfauna. Enkele specifieke deelvragen zijn in de inleiding geformuleerd.

Veldwerk vond plaats in mei en betrof bemonstering van de zeebodem op een 9-tal stations rond de inmiddels verlaten boorlokatie. De resultaten laten duidelijk andere verspreiding van contaminanten zien dan het algemene beeld, dat bekend was van eerder bezochte platforms, waar het boorgruis geen wassing had ondergaan. In de direkte omgeving van het lozingspunt werden aanzienlijk lagere olieconcentraties in het sediment aangetroffen dan bij lokaties waar ongewassen boorgruis was geloosd. Op grote afstand, zelfs op 5000 m, werd echter nog steeds verontreiniging geconstateerd. Als gevolg van deze met toenemende afstand zeer geleidelijke afname in de concentraties van olie, manifesteerde een afnemende intensiteit van effekten op de bodemfauna zich dan ook weinig opvallend. Op 250 tot 750 m waren op het eerste gezicht nauwelijks effekten aantoonbaar. De totale fauna abundantie was hier zelfs relatief groot. Een nadere analyse van het verspreidingspatroon van enkele zeer gevoelige soorten laat echter zien, dat het gebied waarover de fauna stress ondervindt als gevolg van de verontreiniging zich uitstrekt tot op een afstand van meer dan 750 m van het lozingspunt.

Experimenteel werk werd uitgevoerd met sedimentkernen verzameld in september 1989 (bijna 1 jaar na lozing) op 25, 250 en 5000 m van lokatie L5-5. Op deze 'boxcosms', die in het laboratorium onder semi-natuurlijke omstandigheden werden geïncubeerd, werd gedrag en mortaliteit onder enkele uitgezette proefdiersoorten gedurende 3 maanden bestudeerd. De resultaten bevestigen, dat op 250 m de bodemfauna wel degelijk stress ondervindt van de in het sediment aanwezige olie. Met name sterfte onder Echinocardium cordatum wees hierop.

De huidige gegevens duiden er op, dat het toepassen van een wasprocedure leidt tot een vermindering van de intensiteit van negatieve effekten binnen enkele honderden meters van het platform. Vooralsnog is er echter geen aanwijzing, dat de omvang van het gebied dat als gevolg van lozingen blootgesteld wordt aan 'environmental stress' hiermee ook gereduceerd wordt.

Samengevat leiden de resultaten tot de volgende conclusies:

- 1. Een aanzienlijk verarmde macrofauna werd alleen aangetroffen op het dichtstbijzijnde station, 25 m van het lozingspunt. De reduktie in macrofauna kan hier maar ten dele aan OBM-verontreiniging worden toegeschreven. Mogelijk heeft dicht bij het lozingspunt verstikking ook een belangrijke rol gespeeld. Dit verschijnsel zou verklaard kunnen worden uit opeenhoping van cuttings, die geloosd werden tijdens de eerste fase van de boring, waarin geen OBM maar WBM werd toegepast.
- 2. Op 250 m vormde de aanwezigheid van de opportunistische soort Capitella capitata de enige onmiskenbare aanwijzing, dat hier van een effect van olie-verontreiniging sprake was. Wellicht echter mag in de zone tussen 250 en 750 m een afwijkende fauna-opbouw, die zich met name manifesteerde in verhoogde dichtheden van een aantal van nature al min of meer abundante soorten, opgevat worden als een teken van 'environmental stress'.
- 3. Het is zeer waarschijnlijk, dat environmental stress zich uitstrekt tot voorbij 750 m, gezien het verspreidingspatroon van 2 in symbiose met elkaar levende soorten, nl. Montacuta ferruginosa en de zeeëgel, Echinocardium cordatum. Voor de stelling, dat het hier om soorten gaat waarvan er tenminste één uiterst gevoelig moet zijn voor OBM contaminatie, wordt in de discussie uitvoerig argumentatie gegeven.
- 4. De geconstateerde OBM-gevoeligheid van soorten behorend tot het genus *Montacuta*, ofwel die van hun gastheer-soorten (zeeëgels), biedt een aanknopingspunt voor een alternatieve, minder tijdrovende aanpak van biologische veldmonitoring in de toekomst. Immers, bij de analyse van macro-

- fauna-monsters en de evaluatie van de resultaten daarvan met betrekking tot het vaststellen van de ruimtelijke omvang van environmental stress, zou de aandacht zich kunnen beperken tot die gevoelige indicator-soorten.
- Echinocardium cordatum voldeed als testdier in de boxcosm-opstelling goed. Waargenomen lethale en sublethale effekten waren in overeenstemming met eerder gepubliceerde data over de respons van de soort op sedimentverontreiniging met OBM.
- 6. De mortaliteit onder Echinocardium was zowel in verontreinigd sediment van het 25-m station als dat van het 250-m station (bij olie-concentraties van 28 tot 935 mg·kg⁻¹ droog sediment) aanzienlijk. In sediment verzameld op 5000 m van het lozingspunt was de sterfte verwaarloosbaar klein.
- 7. Bij 2 andere geteste soorten (Nucula en Corystes) was een respons waarneembaar, maar niet te kwantificeren. Een derde, vaak talrijke soort (Amphiura) kwam van nature in wisselende aantallen in

- het geteste sediment voor, waardoor mortaliteit onder de uitgezette exemplaren niet betrouwbaar kon worden geschat.
- 8. Mortaliteit onder de natuurlijke infauna in de boxcosms bood geen bruikbaar aanknopingspunt als criterium voor het vaststellen van een dosis-effekt relatie. De mate waarin gedurende het experiment sterfte optrad was niet significant gecorreleerd met olie-concentraties in het sediment.
- 9. De huidige gegevens duiden er op, dat het toepassen van een wasprocedure leidt tot een vermindering van de intensiteit van negatieve effekten binnen enkele honderden meters van het platform. Vooralsnog is er echter geen aanwijzing, dat de omvang van het gebied dat als gevolg van lozingen blootgesteld wordt aan 'environmental stress' hiermee ook gereduceerd wordt. Teneinde inzicht te krijgen in lange-termijn effekten van lozingen van gewassen boorgruis, is het noodzakelijk één of meer follow-up surveys bij L5-5 uit te voeren.

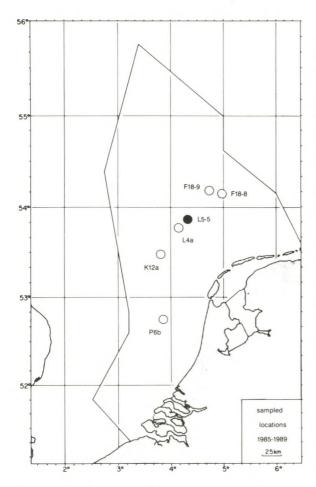


Fig. 1. Position of drill site L5-5. Open circles refer to drilling locations investigated earlier. Solid line: border of the Dutch part of the continental shelf.

1 INTRODUCTION

1.1 GENERAL PART

Since the late 1960's, when in the North Sea the first explorative drillings were performed for exploitable sources of fossil fuels, the intensity of drilling activities has strongly expanded. These increasing efforts went along with a fast technological development. Methods applied in the drilling process were brought to perfection and more sophisticated drilling muds came into use. The improved composition of these muds made them appropriate not only to cool the drilling bit in hard layers of rock, but also to prevent erosion of deep salt layers. Initially only water-based muds (WBM) were used. At the end of the seventies, however, oil-based muds (OBM) were introduced. The oil incorporated in these muds often strongly increased the toxicity of the material, in consequence of its high content of aromatic hydrocarbons (e.g.

diesel oil). Due to tightened regulations the muds currently employed are prepared with oil with a low aromatic content, but still they are more or less toxic. Sieving procedures aimed to regain as much as possible of the OBM from the cuttings before they are dumped do not prevent the disposal of more or less toxic drill grit. Since, together with dumpings of drill cuttings on the sea floor, still large amounts of adhering OBM are discharged, their impact on the marine environment is a source of concern. Earlier investigations in the Dutch part of the continental shelf have shown that significantly enhanced concentrations of oil could be detected in the sediment around drilling locations, within a few hundred meter from the discharge point (VAN HET GROENEWOUD, 1991). These elevated concentrations generally involved evident adverse effects on the benthic fauna (MULDER et al., 1987, 1988; DAAN et al., 1990). Similar observations are numerous for other parts of the North Sea (e.g. DICKS, 1982; GRAY, 1982; DAVIES et al., 1984, 1989; MATHESON, 1986; KINGSTON, 1987; GRAY et al., 1990).

Since 1988 a new technique is applied to reduce the oil content of drill cuttings (according to the new restriction on discharges of OBM at offshore installations to less than 100 g oil.kg⁻¹ dry material). When the total amount of cuttings per drilling location remains the same, a smaller amount of oil discharged would be expected. The technique implies that the sieved cuttings are washed before discharge. A characteristic feature of these so-called 'washed' drill cuttings is the finer grain size.

This report deals with the results of a study on biological effects of discharged washed drill cuttings. The research (carried out in 1989) focussed on the location L5-5, situated in the 'Frisian Front' area of the North Sea (see GEE et al., 1991), i.e. in the transition zone between sand and silty sediment (Fig. 1). More information about drilling location L5-5 is given in Table 1.

The programme included a macrobenthic field survey and boxcosm experiments, to study the functioning of the natural macrofauna and of some introduced test species in intact sediment sections collected around L5-5 and contaminated with various amounts of OBM cuttings. This biological research was carried out in close cooperation with the Netherlands Organisation for Applied Scientific Research (TNO, dept. Den Helder). TNO performed the chemical analyses to assess OBM-contamination levels in the sediment.

The aims of the research programme were the following:

- to develop, evaluate and select suitable methods for a field monitoring system
- to detect the extent of the contaminated area and of the biological effects involved
- 3. to investigate dose-effect relationships

TABLE 1
Information on drilling location L5-5

Position:	53°48′33.1′′ N
	04°20′54.4″ E
Area:	Transition zone between fine sand (South) and silty sediment (North); silt fraction ($<$ 63 μ m) is \approx 15%; depth \approx 41 m
Drilling activities:	1 well drilled with low-aromatic OBM, July-November 1988
Emission:	Dry rock: 336 m ³ or 891 tonnes WBM: 1241 m ³ or 308 tonnes (excl. water)
	OBM: 101 m ³ or 148 tonnes oil: 44 tonnes
Platform:	removed after drilling
Survey:	May 1989
Boxcosms:	September 1989 - December 1989

The fieldwork, which was carried out in the same way as earlier surveys at 5 other drilling locations on the Dutch Continental Shelf, provides appropriate data to describe effects on the benthic community on a spatial as well as a temporal scale. The aim of the experimental boxcosm study was particularly to assess dose-effect relationships.

1.2 ACKNOWLEDGEMENTS

This project was the fifth in a series that is carried out in the framework of a research programme on biological effects of discharges of contaminated drill cuttings. The programme, running since 1985, is performed under contract with and responsibility of the Ministry of Transport and Public Works (RWS, North Sea Directorate), the Ministry of Housing, Physical Planning and the Environment (VROM) and the Ministry of Economic Affairs (EZ, State Supervision of Mines). The project was coordinated by the working group 'Monitoring Offshore Installations', in which participated:

Dr. W. Zevenboom (RWS, North Sea Directorate), chairwoman

Drs. H.R. Bos (RWS, North Sea Directorate), secretary

Drs. S.A. de Jong (RWS, North Sea Directorate), secretary

Drs. K. Meijer (VROM)

Ir. L. Henriquez (EZ, State Supervision of Mines)
Ir. P.J.M. van der Ham (EZ)

Drs. E. Stutterheim (RWS, Tidal Waters Division)
Dr. R. Jacobs (Oil Companies, NOGEPA)
Dr. J.M. Marquenie (Oil companies, NOGEPA)

H.J. van het Groenewoud (TNO) Drs. M.A. van Arkel (NIOZ) M. Mulder (NIOZ) Dr. R. Daan (NIOZ)

Thanks are due to captain, crew and the RWS employees on board of the R.V. 'Mitra' for their assistance in the fieldwork. I. Schweimler and A. Keijser assisted in sorting the samples and T. Kuip and R. Lakeman took care of the experimental setup. Drs. J. van der Meer and H.J. Witte advised and assisted in statistical analyses. B. Bak improved the English. A. Bol, J. Mulder and N. Krijgsman took care of the lay-out and H. Hobbelink made the coverdesign.

2 METHODS

2.1 FIELD SURVEY

2.1.1 POSITIONING

The location L5-5 was drilled in the autumn of 1988 and abandoned afterwards, so there is no platform present any more. Therefore, the position of the discharge point had to be traced merely by navigational techniques. R.V. Mitra, equipped with advanced apparatus like 'side scan', 'dynamic positioning' and underwater video-cameras (mounted on the 'ROHP', i.e. Remote Operating Hoisting Platform), was ideal for this purpose.

Sampling stations were chosen along a crossshaped transect. One axis of the transect ran parallel with the residual current direction, the other axis perpendicular to the first one (Fig. 2). The outermost stations, at 5 km from the discharge point, were assumed to represent a reference situation to which possible spatial effects could be related.

2.1.2 SAMPLING

Bottom samples were collected with a 0.2 m² Van Veen grab, 10 samples per station. From each sample small duplicate cores (diameter 28 mm, depth 10 cm) were taken for chemical and grainsize analyses (N.B. grainsize analyses are performed to confirm that the natural sediment composition is more or less homogeneous in the investigated area). The pooled sediment samples of a station were thoroughly homogenised and immediately frozen at -20°C until later analysis in the laboratory (see VAN HET GROENEWOUD, in prep.). Then the contents of the grab were washed through a sieve (mesh size 1 mm) and the residual macrofauna was preserved in a 6% neutralized formaldehyde solution.

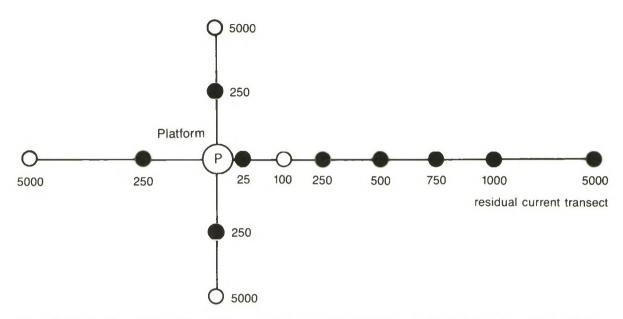


Fig. 2. Positions of the sampled stations (distance to platform in m). Solid circles: samples analysed; open circles: samples not analysed.

2.1.3 TREATMENT OF SAMPLES IN THE LABORATORY

In the laboratory the samples were stained with Bengal rose and sorted under a stereomicroscope. Then, molluscs, crustaceans, polychaetes and echinoderms were identified and counted on species level. (In this report the following acronyms are used: POL=Polychaeta, MOL=Mollusca, CRU=Crustacea, ECH=Echinodermata). Remaining taxa were not further identified and only recorded at higher taxonomic levels. When specimens were broken by handling, only heads were counted.

Not all samples were analysed. In Fig. 2 those stations are indicated where 6 samples per station were analysed.

Oil analyses were performed using the GCMS technique (gas chromatograph mass spectrometer). Concentrations of the fractions of alkanes (C_9 - C_{32}), unidentified complex matter (UCM) and 'other components' were quantified. Methods and results are presented in detail by VAN HET GROENEWOUD (in prep.). Data on oil concentrations are available for the stations at the residual current transect only.

2.1.4 STATISTICAL PROCEDURES

In samples collected over a gradient in pollution there is usually a gradual change in the abundance of individual species over the sampled transect. Statistical analyses of the data are required to obtain objective criteria to decide whether such changes in faunal abundance are significant or not. Such analyses have been performed on both species level and community level. The methods applied have been described in detail in an earlier report (DAAN et al., 1990) and are summarized below.

2.1.4.1 INDIVIDUAL SPECIES (LOGIT REGRESSION)

Logit regression (JONGMAN et al., 1987) is based on a mathematical model for presence-absence data of individual species in samples collected over a transect with a gradient in pollution. The basic idea is that, if the contaminant involved affects the chance of survival of a certain species, the frequency of occurrence of this species will increase or decrease along the transect. Thus, the probability (p) of a species being present in a sample will change along the transect. Logit regression provides a test to decide either that p increases or decreases significantly with distance to the platform, or that p just fluctuates at random.

Logit regression was applied to those species of which at least 20 specimens were found. For stations at the residual current transect the log-transformed distance to the platform was used as the explanatory variable. This transformation was applied for the following reason: according to FINNEY (1978) the response generated by a toxic substance is proportionally related to log c, in which c=concentration of the contaminant. Since the distance (d) to the discharge point is, in theory, inversely proportional to log c (d \approx -log c), effects will usually be inversely proportional to the distance. In such a situation the real distance will therefore act as a suitable explanatory

variable. The distribution of oil contaminants around L5-5, however, suggests that the relation between c and d is better described by: $\log d \approx -\log c$ (see Fig. 3). Therefore the log-transformed distance may provide a more appropriate explanatory variable. For stations at the perpendicular transect the real distance was first multiplied by 1.5, in view of an ellipsoid distribution pattern to be expected around the discharge point (O'REILLY, 1989). Then the same log-transformation was applied.

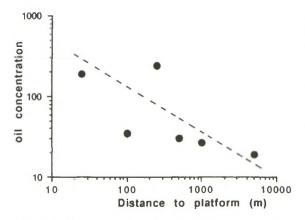


Fig. 3. Sediment contamination levels at the residual current transect of L5-5. Oil concentrations in mg·kg⁻¹ dry sediment. (Data from VAN HET GROENEWOUD, in prep.). The regression line was fit by eye.

2.1.4.2 MACROBENTHIC COMMUNITY: RELATIVE ABUNDANCE

This method provides a measure of the mean relative abundance of all identified species at each station compared to the other stations around a location. If e.g. at a certain station most species occur in lower numbers than at other stations, the relative macrofauna abundance at this station will be attributed a low value. Computation of relative macrofauna abundance is based on a ranking procedure. For all of the individual species the mean density is considered at each of the (n) analysed stations. Per species a rank is attributed to each of the stations, i.e. the rank is 1 for the station with the lowest density and n for the station with the highest density. When this procedure is completed for all species a mean rank can be computed for each station. Differences in mean ranks between stations can be tested for significance by applying analysis of variance and a least significant difference test (see SOKAL & ROHLF, 1981).

2.2 BOXCOSM EXPERIMENTS

The boxcosms employed were sediment cores collected in the first week of September 1989 on the residual current transect of location L5-5. Three stations were chosen, at distances of 25 m, 250 m and 5000 m from the discharge point. All stations were first sampled with a Reineck boxcorer (round cores, diameter 30 cm, depth ≈ 40 cm), to determine the benthic fauna composition at the start of the experiment. Per station 10 cores weren taken and sieved immediately on deck on a 1 mm mesh screen. The samples were preserved in 6% formalin for later analysis in the laboratory. Then the sediment sections to be used in the boxcosms were collected: 5 at each of the 25-m and 5000-m stations and 4 at the 250-m station. These intact boxcores were taken with a (modified) Scripps corer. The stainless steel boxes (50x50x60 cm) were furnished with a mica-teflon coating. The Scripps corer tended to dig about 40 cm deep, thus collecting the major sediment layers inhabited by the benthic infauna. On board the cores were placed in waterproof plywood cases with cooling water and fixed in sand. The water on top of the cores was regularly changed during the transport to the laboratory. After transport the complete cases were placed in an indoor basin and incubated at ≈ 16°C. During the period of incubation (3 months), the temperature was gradually lowered to ≈11°C, according to in situ temperatures. The water on the cores was continuously replaced with filtered and O₂-saturated water from the Wadden Sea, with salinity varying between 27 and 31‰. Apart from inspections, incubation always took place continuously in the dark. During the period of incubation the boxcosms were inspected daily for mortality and activity of test animals and natural infauna. Some small crabs were removed to minimize mortality by predation.

The boxcosms were stocked with test animals in the third week of September. Four species were chosen: Echinocardium cordatum (ECH) and Amphiura filiformis (ECH) were already used in earlier experiments and Nucula turgida (MOL) has been found susceptible to high contamination levels during earlier field surveys (see DAAN et al., 1990). Corystes cassivelaunus (CRU) was chosen to explore its response to sediment contamination merely because of its suitable size (≈4 cm) and its active way of life. In the natural situation this species generally occurs in densities too low to detect any response by its distribution pattern. It was expected that a possible response might be detected by observing its behaviour. On 12 cores, 4 of each station, adult Echinocardium (20 specimens per box) and Nucula (30 per box) were introduced and on 6 of these cores, 2 of each station, also Amphiura (85 per box). Amphiura later appeared (when the Reineck samples were analysed) to be present as natural infauna of the boxcosms, but in variable numbers. Hence, it will not be possible to give a reliable esti-

TABLE 2

The benthic fauna at L5-5. Percentage of occurrence of each species in the total number of analysed samples (54).

POLYCHAETA		Owenia fusiformis	40.74	Eudorella truncatula	9.26
		Myriochele heeri	5.56	Iphinoe trispinosa	3.70
Aphrodita aculeata	33.33	Lanice conchilega	44.44	Diastylis bradyi	44.44
Harmothoe lunulata	3.70	Lagis koreni	5.56	Cirolana borealis	3.70
Harmothoe longisetis	7.41	Pectinaria auricoma	55.56	Ione thoracica	1.85
Harmothoe spec. juv.	1.85	Sosane gracilis	3.70	Melita obtusata	1.85
Gattyana cirrosa	68.52	Terebellides stroemi	3.70	Orchomenella nana	9.26
Pholoe minuta	88.89			Ampelisca brevicornis	1.85
Sthenelais limicola	46.30	MOLLUSCA		Ampelisca tenuicornis	16.67
Eteone longa	3.70			Ampelisca spec. juv.	3.70
Anaitides mucosa	1.85	Nucula turgida	18.52	Amphilochus spec	1.85
Ophiodromus flexuosus	44.44	Thyasira flexuosa	12.96	Cheirocratus sundevalli	1.85
Gyptis capensis	57.41	Montacuta ferruginosa	11.11	Bathyporeia guilliamsoniana	1.85
Synelmis klatti	31.48	Mysella bidentata	85.19	Bathyporeia elegans	5.56
Exogone hebes	18.52	Arctica islandica	11.11	Harpinia antennaria	11.11
Nereis longissima	16.67	Acanthocardia echinata	3.70	Apherusa spec	1.85
Nereis spec. juv.	12.96	Dosinia Iupinus	25.93	Perioculodes longimanus	29.63
Nephtys hombergii	62.96	Venus striatula	57.41	Aora typica	5.56
Nephtys incisa	18.52	Mysia undata	25.93	Caprella spec	3.70
Nephtys cirrosa	14.81	Abra alba	35.19		
Nephtys spec. juv.	40.74	Gari fervensis	1.85	ECHINODERMATA	
Glycera rouxii	72.22	Cultellus pellucidus	51.85		
Glycera alba	20.37	Mya spec. juv.	3.70	Astropecten irregularis	5.56
Glycera spec. juv.	24.07	Corbula gibba	44.44	Amphiura filiformis	98.15
Glycinde nordmanni	59.26	Thracia convexa	33.33	Amphiura chiajei	12.96
Goniada maculata	88.89	Cingula nitida	72.22	Ophiura albida	7.41
Lumbrineris latreilli	10 000	Turritella communis	37.04	Ophiura spec. juv.	57.41
Lumbrineris fragilis	57.41	Natica alderi	24.07	Echinocardium cordatum	11.11
Driloneris filum	1.85	Retusa truncatula	1.85	Lomioudiani oordata	
Orbinia sertulata	3.70	Retusa umbilicata	1.85	OTHER TAXA	
Paraonis spec	11.11	Cylichna cilindracea	87.04		
Spio filicornis	3.70	Cynonia China Ca	01.0	Nemertinea	98.15
Polydora quillei	3.70	CRUSTACEA		Hydrozoa	3.70
Spiophanes kroyeri	1.85	OTTO MOLA		Turbellaria	12.96
Spiophanes bombyx	88.89	Pagurus bernhardus	1.85	Phoroniden	33.33
Scolelepis foliosa	1.85	Porcellana longicornis	1.85	Harp copepoda	7.41
Magelona alleni	5.56	Porcellana spec. juv.	1.85	Parasitaire copepoda	11.11
Chaetopterus variopedatus	57.41	Macropipus spec. juv.	1.85	Oligochaeta	11.11
Tharyx marioni	24.07	Pinnotheres pisum	1.85	Holothuroidea	48.15
Chaetozone setosa	64.81	Ebalia cranchii	27.78	Sagitta spec	9.26
Diplocirrus glaucus	74.07		1.85	Echiurida	37.04
Scalibregma inflatum	1.85	Cancer pagurus	9.26	Sipinculida	94.44
	7.41	Corystes cassivelaunus	9.26 5.56	-	94.44
Ophelina acuminata		Upogebia deltaura		Ascidiacea	9.20
Capitella capitata	12.96	Callianassa subterranea	92.59		
Notomastus latericeus	55.56	Decapoda larven	38.89		
Heteromastus filiformis	38.89	Nebalia bipes	14.81		

mate of the mortality among the introduced *Amphiura* (see chapter 3.2.4). Specimens of *Corystes* were introduced in the remaining 2 boxcosms (20 per box), *i.e.* one of 25 m and one of 5000 m. The 25-m box was, on visual inspection, severely polluted. However, oil data for these 2 boxcosms are lacking. For *Echinocardium*, *Nucula* and *Amphiura* the time was recorded during which the animals burrowed before they finally disappeared in the sediment. In *Corystes* it was meaningless to do so, because of its unpredictable behaviour.

At termination of the experiments, from all boxcosms (except the 2 *Corystes* boxes) 10 samples of the upper 10 cm sediment were collected with a tube corer (diameter 25 mm) for oil analyses (see VAN HET GROENEWOUD, in prep.). Then the sediment of the boxcosms was washed through a 1 mm sieve to collect the macrofauna, including the introduced test animals.

3 RESULTS

3.1 FIELD SURVEY MAY 1989

3.1.1 GENERAL DESCRIPTION

Analysis of a total number of 54 samples from 9 stations resulted in 96 identified species. In Table 2 their percentual occurrence in the samples is summarized.

The fauna in the area was dominated by the polychaete *Lumbrineris latreilli* and the echinoderm *Amphiura filiformis* (Table 11, appendix). On average *Lumbrineris* accounted for 33% of the total macrofauna, *Amphiura* for 39%. *Lumbrineris* occurred in numbers ranging from 240 to 870 ind·m $^{-2}$. Densities at the 25-m station were not different, but the species seemed to be particularly abundant at the 250-m stations. At the stations $\geqslant 250$ m from the platform *Amphiura* densities fluctuated between 320 and 1130 ind·m $^{-2}$ and their share in the total macrofauna abundance between 28 and 54%. At the 25-m station, however, *Amphiura* densities were remarkably low (19 ind·m $^{-2}$) and represented only 4% of the macrofauna.

A majority of the other relatively abundant species (≥10 ind·m⁻²) showed a similar trend as *Amphiura*. *Pholoe minuta*, *Glycera rouxii*, *Spiophanes bombyx*, *Diplocirrus glaucus*, *Cylichna cilindracea* and *Callianassa subterranea* all showed reduced densities at the 25-m station and *Mysella bidentata* was totally absent there. Only *Goniada maculata* showed a different trend. Two species (*Pholoe minuta* and *Spiophanes bombyx*) seemed to occur in remarkably enhanced numbers especially at the 250-m stations.

As a result of the reduced densities of *Amphiura* and most of the other abundant species, the total macrofauna abundance was considerably lower (470 ind·m $^{-2}$) at 25 m from the platform than at all other stations. On the other hand, total macrofauna densities were high at the perpendicular 250-m stations (1970-2150 ind·m $^{-2}$) compared to the stations at the r.c. transect (840-1890 ind·m $^{-2}$). The species richness at the 25-m station was also low (39) compared to the other stations (46-62).

3.1.2 PRESENCE-ABSENCE DATA: LOGIT REGRESSION

Logit regression was applied to 34 species where at least 20 specimens were found (Table 3). The hypothesis Ho that frequency of occurrence in the samples was not dependent on the distance between sampling station and platform was rejected at the 5% level in 10 species, in favour of the alternative, viz. gradients being significantly present. In 3 species such a gradient was even significant at the 0.1% level. Two species showed a significant negative gradient, i.e. increasing densities towards the platform. All other species that showed a significant trend occurred in increasing densities with increasing distance from the platform. Table 3 shows that the total number of positive trends, including those that were not significant, was considerably higher (25) than the number of negative trends (10). This indicates, once more, that most species occurred in reduced numbers in the vicinity of the discharge point.

TABLE 3

List of species for which density gradients were tested by logit regression. Sign of the gradient (+/-) and significance level are indicated: +=increasing densities off the location; -=decreasing densities off the location; o=no gradient.

	sign	signif level (%)
Aphrodite aculeata	-	n.s.
Gattyana cirrosa	+	1
Pholoe minuta	+	0.1
Sthenelais limicola	+	n.s.
Ophiodromus flexuosus	-	5
Gyptis capensis	-	n.s.
Glycera rouxii	+	n.s.
Glycinde nordmanni	+	5
Goniada maculata	-	5
Lumbrineris latreilli	0	-
Lumbrineris fragilis	+	5
Spiophanes bombyx	+	n.s.
Chaetopterus variopedatus	+	0.1
Chaetozone setosa	+	n.s.
Diplocirrus glaucus	+	n.s.
Notomastus latericeus	+	n.s.
Heteromastus filiformis	+	n.s.
Owenia fusiformis	+	n.s.
Lanice conchilega	+	n.s.
Pectinaria auricoma	+	0.5
Mysella bidentata	+	0.1
Venus striatula	+	n.s.
Mysia undata	+	n.s.
Nephtys hombergii	-	n.s.
Abra alba	+	n.s.
Cultellus pellucidus	-	n.s.
Corbula gibba	+	n.s.
Cingula nitida	-	n.s.
Turritella communis	+	n.s.
Cylichna cilindracea	+	n.s.
Ebalia cranchii	-	n.s.
Callianassa subterranea	+	n.s.
Diastylis bradyi		n.s.
Amphiura filiformis	+	5

3.1.3 RELATIVE MACROFAUNA ABUNDANCE

The relative macrofauna abundance was obviously the lowest at the 25-m station (Fig. 4). Analysis of variance revealed highly significant differences (0.1% level) in mean ranks of the different stations. It seems that not only the 25-m station differed with other stations. At 1000 m relative abundance was also apparently low, particularly when compared to the 250-m stations at the perpendicular transects. The low abundance at 1000 m cannot be explained from the observed oil concentration at that site. A least significant difference test (LSD-test, SOKAL & ROHLF, 1981) was applied to all pairs (36) of means to detect whether they were significantly different. The result was that among the 36 pairs of stations mutually compared, 21 were significantly different at the 5%

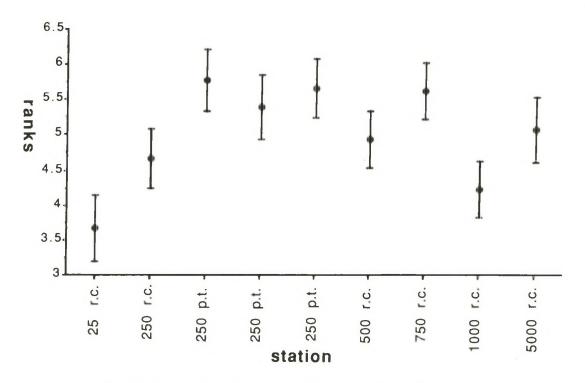


Fig. 4. Relative macrofauna abundance at L5-5 (mean ranks and 95% confidence limits).

TABLE 4
Statistical significance (LSD-test) of differences in relative macrofauna abundance between sampled stations. R=residual current transect; P=perpendicular transects. Significance level (%) indicated.

					(/ 0 / 1.				
	25 (R)	250 (R)	250 (P)	250 (P)	250 (P)	500 (R)	750 (R)	1000 (R)	5000 (R)
(R) 25 m			7						
(R) 250 m	0.5			7					
(P) 250 m	0.5	0.5			7				
(P) 250 m	0.5	5	n.s.			7			
(P) 250 m	0.5	0.5	n.s.	n.s.			٦		
(R) 500 m	0.5	n.s.	5	n.s.	5			1	
(R) 750 m	0.5	0.5	n.s.	n.s.	n.s.	5			7
(R) 1000 m	n.s.	n.s.	0.5	0.5	0.5	5	0.5		
(R) 5000 m	0.5	n.s.	5	n.s.	n.s.	n.s.	n.s.	5	

level (Table 4). The number of 21 rejections of H_0 (*i.e.* mean ranks of mutually compared stations are equal) is much higher than expected, if H_0 were true for all stations, since among 36 tests at the 5% level maximally 4 will statistically lead to rejection. This clearly indicates that the majority of the differences detected do not concern Type-1 errors (*i.e.* rejection of H_0 when it is true). Moreover, at the 0.5% level still 15 differences between stations were significant. The 25-m station differed from all other stations, with the exception of the 1000-m station. On the other hand, the 1000-m station differed from the other stations, but not from the 250-m r.c. station.

3.1.4 ABUNDANCE PATTERNS OF 'SENSITIVE' AND 'OPPORTUNISTIC' SPECIES

In the spatial distribution of the macrofauna around the discharge point, the abundance patterns of sensitive and opportunistic species are of particular importance. In a previous report (DAAN et al., 1990) a list of 37 species was presented, which had shown susceptibility to OBM contamination in at least 3 OBM surveys by their relatively low densities, or even absence, in the vicinity of platforms. A similar list referred to 4 opportunistic species that were found to be particularly abundant in the vicinity of platforms. Table 5 shows to what extent the listed sensitive and opportunistic species revealed a distribution pattern that was indicative of a gradient in OBM contamination of the sediment at L5-5. Thirteen species were too scantily distributed to identify such a gradient. Five species did not display any gradient, and 19 sensitive species showed reduced densities (or were absent) at the 25-m station. The gradient suggested by the distribution of these species seems, however, to cover only a small area around the platform, since the densities of most species did not further increase beyond 250 m. The presence of a known opportunistic species (Capitella capitata, 10 ind·m-2) at the 25-m station obviously indicates sediment contamination there. A few specimens of this species were also found at two 250-m stations.

3.1.5 DOSE-EFFECT RELATIONSHIPS

An evaluation of the spatial variation in the macrobenthic community in relation to sediment contamination levels can be based only on those stations where both macrofauna abundance and contamination levels were assessed. In Fig. 3 data on oil concentrations in the sediment on the residual current transect are plotted against distance from the platform. The figure shows that strongly elevated concentrations were found at 25 m and 250 m, whereas slightly (but when compared to background level ≈ 2 mg oil·kg⁻¹ dry sediment, significantly) enhanced concentrations were observed at all other stations.

TABLE 5

Evaluation of the abundance patterns of 41 species, which earlier have been described as either 'sensitive' or 'opportunistic'. +: abundance pattern is indicative of a sensitive species; —: abundance pattern is indicative of an opportunist =: abundance pattern does not indicate a response; ?: numbers found too low to be indicative (Note that the qualifications are based on the abundance patterns of the individual species and not on presence-absence data as used in logit regression).

used in logit	regression).
A. Sensitive species	
Montacuta ferruginosa	+ (NB <20 specimens)
Scalibregma inflatum	?
Pholoe minuta	+
Amphiura filiformis	+
Echinocardium cordatum	+ (NB < 20 specimens)
Mysella bidentata	+
Nephtys hombergii	+
Lumbrineris latreilli	=
Chaetozone setosa	+
Owenia fusiformis	+
Nucula turgida	?
Gattyana cirrosa	+
Harpinia antennaria	?
Lagis koreni	?
Glycinde nordmanni	+
Cylichna cilindracea	+
Harmothoe longisetis	?
Callianassa subterranea	+
Magelona papillicornis	? (species not found)
Tellina fabula	? (species not found)
Natica alderi	?
Spiophanes bombyx	+
Ophiodromus flexuosus	=
Notomastus latericeus	+
Lumbrineris fragilis	+
Amphiura chiajei	?
Leucothoe incisa	? (species not found)
Chaetopterus variopedatus	+
Thamarioni	?
Ophiura albida	?
Gyptis capensis	=
Lanice conchilega	+
Perioculodes longimanus	?
Diplocirrus glaucus	+
Abra alba	=
Turritella communis	=
Sthenelais limicola	+
B. Opportunistic species	
Nereis longissima	?
Capitella capitata	- (NB < 20 specimens)
0	

? (species not found)

Spio filicornis

Anaitides groenlandica

The relationship between observed adverse effects on the macrobenthic community and sediment contamination levels is not quite clear. Indeed, at the 25-m station all defined effects (DAAN et al., 1990) were observed, as was to be expected in view of the high contamination level (190 mg oil-kg-1 dry sediment) measured here. However, at 250 m, where oil concentrations were even higher, observed faunistic features were hardly indicative of the presence of oil. In fact only the presence of some specimens of Capitella capitata suggested sediment contamination. Slight indications of the presence of oil in the sediment were further provided by the absence (or reduced densities) of some sensitive species (Callianassa subterranea, Mysella bidentata, Montacuta ferruginosa and Echinocardium cordatum) and low relative macrofauna abundance. These indications should, however, not be regarded as decisive. If, on the other hand, the distribution of Montacuta and Echinocardium was indicative of sediment pollution, the effects of this pollution would seem to stretch over 750 m, since these species were found almost exclusively at large distances (1000 and 5000 m) from the platform (see Table 11).

For the discrepancy between observed effects at 25 and 250 m and the corresponding sediment contamination levels, the following explanations are suggested:

- a. Inhomogeneity in the distribution pattern of the oil. Within a series of grab samples oil concentrations may be extremely variable (Davies et al., 1989; VAN HET GROENEWOUD, 1991). The degree of patchiness may largely determine the intensity of effects. The high mean contamination level at 250 m may be the result of a few patches of high oil concentrations (oil concentrations were determined in pooled subsamples originating from 10 Van Veen Grabs, of which 6 were used for macrofauna analysis). Patchiness in the distribution of the oil is indicated by the low concentration at 100 m compared to 25 m and 250 m.
- b. Effects at 25 m may be the accumulated result of OBM contamination and smothering, due to large amounts of drill cuttings dumped before the discharge of washed drill cuttings. Water-based drilling fluids (WBM) are often used during the first phase of the drilling. The material dumped during this period may have accumulated in the immediate vicinity of the platform.

3.1.6 SUMMARY OF THE FIELD SURVEY RESULTS

The results of the survey at L5-5 may be summarized as follows:

 An accumulation of strong adverse effects on the macrobenthic community became manifest at the 25-m station only. The observed effects were

- a. a number of sensitive species occurred in reduced densities
- b. >50% of the abundant species occurred in reduced densities
- c. a dominant species (Amphiura filiformis) occurred in reduced density
- d. overall macrofauna abundance was reduced
- e. relative macrofauna abundance was reduced
- f. species richness was reduced
- g. an opportunistic species (Capitella capitata) occurred in enhanced numbers
- 2. In spite of elevated oil concentrations at the 250-m residual current station, the macrofauna seemed hardly to be affected here. Only the presence of an opportunistic species was plainly indicative of sediment pollution. The 250-m stations at the perpendicular transects did not seem to be affected.
- The 5000-m station did not strictly speaking represent a true reference station, since oil concentrations were, although low, above background level (≈2-3 mg·kg⁻¹ dry sediment).
- 4. The 1000-m station was rather poor in macrofauna, apparently due to other factors than sediment contamination. There is no other drilling location particularly in the vicinity of the 1000-m station. A possible but unverifiable explanation might be that the area around the 1000-m station had been visited by beam trawlers before macrobenthos sampling took place.
- The distribution patterns of only 2 species (Montacuta ferruginosa and Echinocardium cordatum) were indicative of a larger area (>750 m) being affected. Even at 5000 m, however, their numbers were low.

3.2 BOXCOSM EXPERIMENTS

3.2.1 GENERAL REMARKS

At first sight, the sediment surface of cores of the 3 stations looked different. In the 5000-m cores the sediment surface displayed numerous funnels, tubes, holes and cones (≈ 10 cones per core). Signs of oil contamination were absent. Biological activity was confirmed by the observation of repeated emission of sediment clouds from the holes. The sediment of the 25-m cores was obviously less rich in relief. Fewer holes and tubes could be seen and the number of cones was about 5 per core. In 4 cores black blotches of oil appeared at the sediment surface. The cores of the 250-m station had a more or less intermediate appearance. On 2 cores a small blotch of oil was visible. During the experimental period small white colonies of the sulphur bacterium Beggiatoa mirabilis developed on the oil patches. The occurrence of this bacteria is indicative of the presence of organic substrate and of anoxic conditions (e.g. WIESSNER, 1981).

TABLE 6

Boxcosm experiment. Initial fauna abundance of the sediment used in the boxcosms. Numbers of species and individuals (except unidentified taxonomic groups) in 10 Reineck samples per station.

	Number of species	Total number of individuals
5000 m	67	1156
250 m 25 m	63 50	1865 692

Oil analyses performed after termination of the experiment (VAN HET GROENEWOUD, in prep.) revealed that oil concentrations in the sediment of the 5000-m cores were at background level (2-3 mg oil-kg⁻¹ dry sediment). In the cores of the other 2 stations the concentrations were rather variable, ranging from 28 to 215 mg-kg⁻¹ in the 250-m boxes and from 31 to 935 mg-kg⁻¹ in the 25-m boxes.

3.2.2 SURVIVAL OF THE NATURAL INFAUNA

An estimate of the survival rate of the natural infauna in the boxcosms is obtained by comparing the abundance of living fauna present at the end of the experiment with estimates of the initial macrofauna abundance. Estimates of the initial abundance are based on the Reineck box samples that were taken simultaneously, when the sediment sections for the boxcosms were collected. The sediment depth sampled by the Reineck corer is similar to that in the boxcosms (35- 50 cm).

Together the 3x10 Reineck samples collected at the 3 stations yielded 83 identified species. Numbers of species and total numbers of individuals found at each station are listed in Table 6. The number of species was low at the 25-m station compared to 250 m and 5000 m. Total macrofauna densities were also low at the 25-m station but particularly high at the 250-m station, largely due to high densities of the 2 dominant species *Lumbrineris latreilli* and *Amphiura filiformis*.

A comparative estimate of mortality in individual species is only possible for species that were more or less abundant at the start of the experiment. In Table 7 those species are listed of which at least 2 specimens were found per Reineck sample (2 specimens per Reineck sample corresponds to 27 specimens per m²). The estimated mean initial density, the mean final density and the estimated mortality are given. The data show that mortality occurred among all species, but it was rather variable between species. Only 3 species (Lumbrineris latreilli, Callianassa subterranea and Amphiura filiformis) were initially abundant at all stations. Statistical significance of differences in mortality between the boxcosms of the 3 stations was tested for these species by two-way analysis of variance, after transformation by log(n+1) to stabilize variances. The same statistical procedure was performed for the pooled counts of the 3 species and for the pooled counts of all other species. The results summarized in Table 8 show that overall mortality during incubation was significant at the 5% level in all species. Differences between numbers found in the sediment of the 3 stations were also significant in all species except Lumbrineris latreilli. However, an interaction effect could not be

TABLE 7

Boxcosm experiment-natural infauna. Densities (n·m⁻²) of 11 abundant species and of 4 taxonomic groups before and after incubation. Densities of taxonomic groups are exclusive of dominant species (*Lumbrineris latreilli* and *Amphiura filiformis*). Estimated percentual mortalities are indicated. Oil contents in parentheses (mg·kg⁻¹ dry sediment).

	5000 m (2-3)			250	250 m (29-215)			25 m (31-935)		
	start	end	% mort	start	end	% mort	start	end	% mort	
Lumbrineris latreilli	283	129	53	461	382	15	368	238	34	
Callianassa subterranea	93	70	23	73	44	38	39	17	56	
Amphiura filiformis	690	171	74	1390	532	58	251	119	62	
Gattyana cirrosa	44	22	47	38	18	52	< 27			
Chaetopterus variopedatus	47	33	27	54	35	32	< 27			
Mysella bidentata	34	13	61	96	44	53	< 27			
Pholoe minuta	< 27			48	11	76	< 27			
Heteromastus filiformis	< 27			35	2	94	< 27			
Abra alba	< 27			30	1	96	< 27			
Cingula nitida	< 27			39	14	64	< 27			
Philine scabra	< 27			< 27			31	18	61	
Polychaeta	308	149	50	360	125	62	125	57	54	
Mollusca	132	100	22	248	126	48	123	68	43	
Crustacea	175	94	45	145	55	61	91	22	75	
Echinodermata	41	26	35	22	8	64	19	6	69	

among all species, but it was rather variable between species. Only 3 species (Lumbrineris latreilli, Callianassa subterranea and Amphiura filiformis) were initially abundant at all stations. Statistical significance of differences in mortality between the boxcosms of the 3 stations was tested for these species by two-way analysis of variance, after transformation by log(n+1) to stabilize variances. The same statistical procedure was performed for the pooled counts of the 3 species and for the pooled counts of all other species. The results summarized in Table 8 show that overall mortality during incubation was significant at the 5% level in all species. Differences between numbers found in the sediment of the 3 stations were also significant in all species except Lumbrineris latreilli. However, an interaction effect could not be found. In other words, differences in mean mortality between boxcosms of the 3 stations were not significant. A plausible explanation might be that possible impact of oil on mortality rates was masked by the high variability in oil concentrations. Therefore an additional statistical procedure was performed (analysis of covariance) to assess whether varying oil concentrations may explain the variance in final densities in the boxcosms. When the density of a species in a box sample is assumed to depend on place (station), time (before or after incubation) and, after incubation, on the oil concentration in the boxcosms, the following expression may represent a model which can be tested by analysis of covariance (SOKAL & ROHLF, 1981):

$$\underline{Y}_{ijk} = \mu + c_i \cdot \alpha_i + c_j \cdot \beta + c_j \cdot \gamma \cdot z_{ijk} + \underline{\epsilon}_{ijk}$$

where:

 Y_{ijk} =log(density at station i, at time j in the k^{th} sample)

 μ = mean density of the population

 α_i = place effect of station i

 β = time effect (effect of incubation)

 $\gamma \cdot z_{ijk}$ =effect of oil concentration in the sediment, with

z_{ijk} =log(oil concentration)

 $\frac{\epsilon}{-}_{ijk}$ = random deviation

ci and ci are dummy variables

This model was tested (at the 5% level) for each of the species and groups listed in Table 8. The results were similar to those listed in this table, *i.e.* in none of the species or groups of species the variance in oil concentrations could explain a significant part of the variance in final densities. It is concluded therefore that, from a statistical point of view, there is no indication that the elevated contamination levels in the 25-m and 250-m boxcosms did affect the mortality rates of the natural infauna, neither those of individual species nor those of larger taxonomic groups.

TABLE 8

Significance of differences in densities between stations, mortality and of the interaction effect, i.e. station dependent mortality, tested by analysis of variance. (ANOVA) * : significant at the 5% level; n.s. : not significant.

	difference between stations	mortality	interaction
Lumbrineris latreilli	*	n.s.	n.s.
Callianassa subterranea	*		n.s.
Amphiura filiformis		*	n.s.
Pooled counts of the above 3 species	•	*	n.s.
Pooled counts of all other species	•		n.s.

3.2.3 BURROWING BEHAVIOUR OF TEST ANIMALS

When the test animals were introduced in the boxcosms, their burrowing times were recorded (Fig. 5). The burrowing time of Amphiura was apparently not affected by the presence of oil in the sediment. After 20 min 90-95% had burrowed in all boxcosms and burrowing was finished within 40-80 min in all boxcosms. In Nucula burrowing times varied more between the boxcosms, but there was no indication that their behaviour was affected by sediment contamination. Individual differences in a delay before the animals became active, after they were introduced, did consistently influence the burrowing times. Also the behaviour of Echinocardium was hardly affected. Only a few specimens that were placed on black oil patches in the 25-m and 250-m boxes kept creeping around for some time till finally they dug in.

During the incubation period effects of sediment pollution became evident by the behaviour of *Echinocardium*. In the 5000-m boxes the animals remained under the sediment for almost the whole period (Table 10). In the contaminated 25-m and 250-m boxes, however, the animals frequently reappeared at the sediment surface, till finally they stopped burrowing. As Table 10 shows, the mean percentage of living animals observed on top of the sediment varied between 5 and 57%.

3.2.4 MORTALITY OF TEST ANIMALS

Mortality among the introduced test species could be assessed only in Corystes, Nucula and Echino-

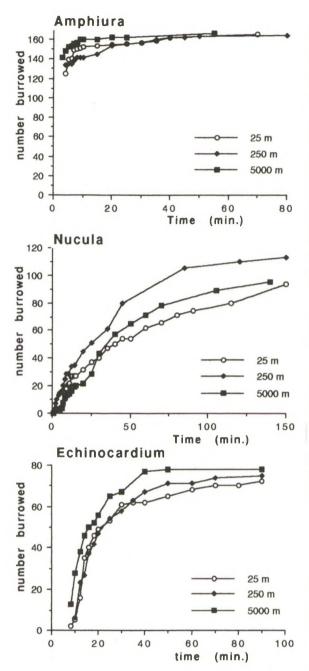


Fig. 5. Burrowing times of 3 test species in sediment from 25 m, 250 m and 5000 m. Numbers of specimens introduced per boxcosm: *Amphiura*: 85, *Nucula*: 30, *Echinocardium*: 20.

cardium. For Amphiura it was not possible because of its variable numbers initially present among the natural infauna in the sediment of the boxcosms.

All the 20 specimens of *Corystes* survived the experimental period in the 25-m box. In the 5000-m box one specimen died. Hence, it would seem that there

was no adverse effect of oil on this species. It is remarkable, however, that at termination of the experiment the animals were found at a depth of maximally 10-15 cm in the sediment of the 25-m box, whereas in the 5000-m box most animals were recovered at 20-30 cm.

On average *Nucula* showed highest mortality in the 25-m sediment and lowest mortality in the 5000-m sediment (Table 9). However, the variability in mortality between different cores of each station was high compared to the differences in mortality between stations. The variability in mortality may be partly explained by differences in sediment contamination levels. Fig. 6, showing relative mortality rates plotted against log-transformed oil concentrations, indeed suggests that a positive correlation exists between mortality rate and contamination, notwithstanding the poor fit of the linear regression model illustrated.

The most obvious trend in mortality was found in *Echinocardium*. Table 10 shows that mortality of the test animals in the reference cores of 5000 m was almost nil compared to that in the 25-m and 250-m

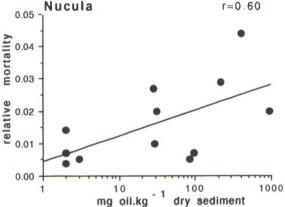


Fig. 6. Boxcosm experiment. Plot of relative mortality of *Nucula* against oil concentrations in the sediment. Relative mortality is defined as ($\log n_0 - \log n^*$)/t, in which $n_0 = \text{initial}$ number of specimens (30), $n^* = \text{final}$ number of specimens and t = time (11 weeks).

cores. Remarkably, the mortality was even higher in the 250-m sediment than in the 25-m sediment. Since the animals usually appear at the sediment surface before dying, mortality could be recorded during daily inspections (Fig. 7). In the 250-m cores 50% of the animals died within 3 weeks. A positive relationship between relative mortality rates and oil concentrations in the individual boxcosms is once more evident from Fig. 8. Although the linear model fitted does not provide a quantitatively reliable dose-effect relationship, this plot may again illustrate the existence of such a relationship. In addition to mortal-

TABLE 9
Boxcosm experiment. Mortality of Nucula turgida. Number of specimens introduced per boxcosm=30.

25 m				250 m		5000 m			
Вох	number recovered	mortality (%)	Box	number recovered	mortality (%)	Вох	number recovered	mortality (%)	
1	18	40	11	23	23	6	27	10	
2	18	40	12	25	17	7	21	30	
3	10	67	13	15	50	8	25	17	
4	26	13	14	14	53	9	26	13	
mean	18	40		19	36		25	17	

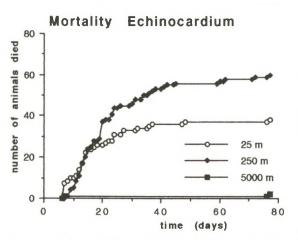


Fig. 7. Mortality in *Echinocardium* in sediment cores from 25 m (31 - 935 mg oil-kg⁻¹ dry sediment), 250 m (28 - 215 mg-kg⁻¹) and 5000 m (2 - 3 mg-kg⁻¹).

concentrations in the individual boxcosms is once more evident from Fig. 8. Although the linear model fitted does not provide a quantitatively reliable dose-effect relationship, this plot may again illustrate the existence of such a relationship. In addition to mortality, loss of spines was recorded in both dead and surviving animals. Loss of spines generally occurs on the ventral side of *Echinocardium* when the animals are in bad condition. Table 10 shows that this sub-

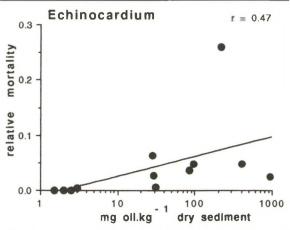


Fig. 8. Boxcosm experiment. Plot of relative mortality of *Echinocardium* against oil concentrations in the sediment. Relative mortality is defined as (log n* - log n₀)/t, in which n₀=initial number of specimens (20), n*=final number of specimens and t=time (11 weeks).

lethal effect became manifest in most dead specimens, but that it was also frequently observed in the surviving animals of the contaminated boxcosms.

4 DISCUSSION

The central question underlying the present study was to what extent washing procedures as applied at

TABLE 10

Boxcosm experiment L5-5. Mortality, loss of spines and behaviour of *Echinocardium cordatum*. Number of specimens introduced per boxcosm=20. *: The fraction of the living animals that were, averaged over the whole incubation period, on top of the sediment.

		5000 m			250 m				25 m			
Core nr	6	7	8	9	11	12	13	14	1	2	3	4
Mortality (%)	0	0	0	10	50	70	80	100	45	15	70	60
Mean loss of spines on ventral side in dead animals	-	-	-	0	45	60	90	80	45	0	35	45
Idem in surviving animals (%)	0	0	0	1	5	20	35	-	10	1	10	10
Animals on top of the sediment between 29.9 and 51.2 (%)*	0	0.4	0	0.1	7	30	11	57	22	7	5	32
Oil contents mg/kg dry weight	2	2	2	3	29	97	28	215	935	31	409	85

location L5-5 contribute to a reduction of adverse effects on the benthic community. Answering this question is complicated by the fact that the spatial distribution of oil contaminants after discharges of washed material is more diffuse than oil distribution patterns after traditional discharges. As shown by VAN HET GROENEWOUD (in prep.) the contamination levels in the immediate vicinity (≤100 m from the platform) are considerably less elevated than was generally observed at OBM-locations where no washing procedure was applied. At such locations contamination levels were found readily to exceed 1000 mg-kg-1 dry sediment (compare the locations L4a, K12a and F18.9, VAN HET GROENEWOUD, 1991). On the other hand it seems that, at L5-5, significantly enhanced oil concentrations (above background level ≈2 mg oil·kg-1 dry sediment) occurred over a much wider area. During the May survey elevated concentrations were assessed even at 5000 m (19 mg oil-kg⁻¹ dry sediment). Therefore, the 5000-m station should, in fact, not be regarded as a true reference station. The more diffuse an environmental disturbance, the more diffuse will be the spatial distribution of resulting effects. Effects on the benthic community, which is characterized by spatial density patterns of individual species that are seldom homogeneous, will thus be more difficult to distinguish.

The field data obtained during the May survey showed strong adverse effects only at 25 m, though the highest oil concentrations were assessed at 250 m. At 250 m the only evident effect observed was, so far, the occurrence of Capitella capitata. It is not clear why at the high contamination level detected, which generally has been found to involve clear signs of stress (DAAN et al., 1990), strong effects were not observed. A possible explanation may be a different distribution of contaminants in the sediment, e.g. the oil in the sediment may be more patchily distributed at 250 m than at 25 m. Moreover it should be stressed that effects observed are not necessarily induced merely by oil contamination. In fact such effects are the cumulative result of all kinds of disturbances to which the benthic fauna is subjected. As suggested in chapter 3.1.5 the effect of smothering, due to preceding discharges of WBM cuttings, might have substantially contributed to the impoverishment of the macrofauna close to the discharge point.

Although strong adverse effects were not found at 250 m and farther off, this does not imply that environmental stress was restricted to the area in the immediate vicinity of the discharge point. When the data are evaluated according to the scenario outlined by GRAY et al. (1990), there are indications that stress may have influenced the species assemblage in the area between 250 m and 750 m. These authors suggested that commonly used criteria to describe ef-

fects of environmental pollution (e.g. dominance by opportunists or reduction of species richness) are based on features that appear rather late in the sequence of responses to stressors (see also GRAY, 1989). Changes in the species assemblage should evidently be considered more decisive at lower contamination levels. Their scenario suggests that moderate environmental disturbance 'gives advantage to some species which increase abundance, and leads to eradication of some rare species, whereby different species are able to colonize in low numbers.' As a consequence this could lead to a slight increase in faunal abundance and species richness. Indeed, we have seen that several of the rather abundant species occurred in highest densities in the area between 250 m and 750 m (e.g. Pholoe minuta, Nephtys hombergii, Goniada maculata, Lumbrineris latreilli, Spiophanes bombyx, Chaetozone setosa, Diplocirrus glaucus, Cingula nitida, Cylichna cilindracea and Amphiura filiformis) and macrofauna abundance and species richness were highest at 250-m stations at perpendicular transects. Although this faunal increase seems to fit in the scenario of a changing species assemblage, it is doubtful whether this trend should be considered as a response to environmental stress, since none of the species mentioned above showed a density trend consistently related to sediment contamination levels. Moreover, an inspection of the data collected by us during 9 preceding OBM surveys (see MULDER et al., 1987, 1988 and DAAN et al. 1990) revealed that none of these species showed a consistent trend of increased numbers at moderate contamination levels. A more substantial indication of stress in the area between 250 m and 750 m is the observation that some species, which are apparently highly sensitive, seem to have disappeared here. GRAY et al. (1990) indeed suggested such responses in some rare species, but they did not mention particular species by name, probably because eradication of rare species is generally not statistically verifiable. There is, however, much evidence now that species belonging to the genus Montacuta are extremely sensitive to oil contamination. In an earlier report (DAAN et al., 1990) Montacuta ferruginosa ranked first in a list of species that were found to be consistently absent in the vicinity of platforms. The list was based on data obtained during 10 surveys to 4 different OBM locations. The present survey at L5-5 provides convincing evidence that the earlier observed patterns in the spatial distribution of *Montacuta* were not accidental. Fig. 9 may illustrate this but some brief comments will be appropriate:

The locations L4a, L5-5 and F18.9 are situated in the silty area, where *Montacuta ferruginosa* is generally less abundant than in the more sandy area where P6b and K12a are situated. At L4a, a baseline

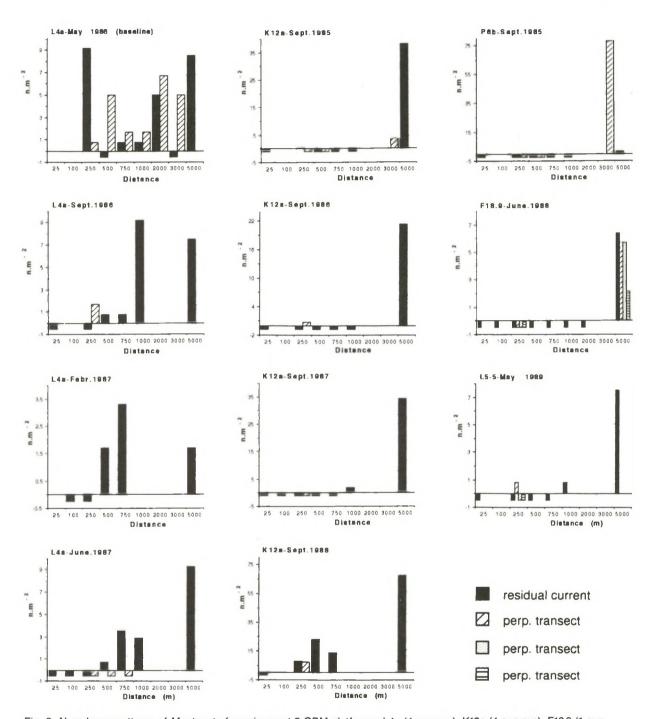


Fig. 9. Abundance patterns of *Montacuta ferruginosa* at 5 OBM platforms: L4a (4 surveys), K12a (4 surveys), F18.9 (1 survey), P6b (1 survey) and L5-5 (1 survey). Zero values are shown below the x-axis.

study (before OBM drilling, May 1986) failed to find a pattern in the distribution of Montacuta. In September of the same year, during drilling activities, a pattern indicating sensitiveness became visible. In February and May of 1987, the species was found only at 500 m and farther off. At K12a the distribution of the species was, during 3 consecutive years (1985, 1986 and 1987), almost completely restricted to stations at 3000 m and 5000 m, whereas in 1988 it seemed to recolonize areas closer to the platform, which was in accordance with the overall impression that a phase of recovery had set in. Still the densities at 5000 m were highest. At P6b and F18.9 (both locations —with heavy loads of pollution— were visited only once) Montacuta was exclusively found at 3000 m and 5000 m. This general pattern of low densities or even absence within ≈ 1000 m was confirmed now by the observation at L5-5, where several specimens were found only at the 5000-m station (though oil concentrations were still elevated there).

An inspection of the data presented by GRAY et al. (1990) surprisingly reveals that another member of the genus, viz. Montacuta substriata, was abundant around EKOFISK only outside a radius of 2 to 3 km, whereas this species was absent within 500 m. At ELDFISK M. substriata was found only outside a radius of 1 to 2 km.

The aggregated data available now on distribution patterns of *Montacuta* species around drilling platforms evidently suggest high susceptibility to OBM contamination and indicate that, at L5-5, the area of environmental stress stretches over a radius of 750 m.

If, in the future, the distribution pattern of Montacuta was utilized as a most sensitive criterion to assess the radius of environmental stress around drilling platforms, the scenario of field research could be adapted to this purpose. In this context it is worth noting that known identification keys consider members of the genus Montacuta as commensals of echinoids. M. ferruginosa was found to be a commensal of Echinocardium cordatum and M. substriata of Spatangus purpureus (e.g. NORDSIECK, 1969; TEB-BLE, 1976). It is not clear yet to what extent this interspecific relationship is absolute, but according to BERGMAN & DUINEVELD (1990) the distribution of M. ferruginosa in the Southern North Sea is restricted to those areas in which E. cordatum occurs. At L5-5 there was also a clear relationship between the presence-absence patterns of the 2 species (Table 11), the latter species being represented only by relatively large specimens (20 to 42 mm). It seems plausible that the presence of *Montacuta* depends on the occurrence of larger echinoid specimens on which it lives attached to the anal spines. If this is true, it is conceivable that Montacuta might be indirectly indicative of sediment contamination and that, in fact, the presence-absence pattern of larger specimens of the host species would be the direct response to sediment contamination. This idea is supported by the results of the boxcosm experiments, in which adult *E. cordatum* proved to be strongly sensitive to sediment contamination. If this hypothesis turns out to be true, scenarios for field monitoring will become much simpler.

The functioning of the natural infauna in the box-cosms did not reveal significant differences in relative mortality. Indeed, crustaceans and molluscs seemed to suffer somewhat higher mortality in the contaminated cores, but for polychaetes and echinodermes this was only true when the dominant species (Lumbrineris latreilli and Amphiura filiformis) were left out of consideration. Moreover, overall mortalities were not significantly different.

In agreement with the results of the field survey, the initial fauna abundance in the 25-m boxcosms was low compared to that of the 250-m boxcosms, though the oil concentrations in the 25-m cores were in the same range as found in the 250-m cores. However, mortality rates of the natural infauna were not different, neither on species level nor on community level. This once more supports the idea that initial discharges of WBM cuttings might have suffocated a considerable part of the natural infauna at 25 m.

In contrast to the findings concerning the natural infauna, clear responses were observed in the introduced test animals. However, not all responses observed were sensitive enough to serve as a suitable criterion for active biological monitoring. In Corystes there was no lethal response. Only its burrowing depth was obviously reduced in the severely polluted 25-m core to which it was transferred. Nucula showed enhanced mortality in polluted cores, but its background mortality was too high and also too variable to base a reliable test on. The outstanding test species again appeared to be Echinocardium cordatum, which supports the earlier findings of DAAN et al. (1990) and ADEMA (1991). To average contamination levels ranging from 29 to 935 mg oil kg-1 dry sediment it responded consistently by elevated mortality, loss of spines and stressed burrowing behaviour. The slight mortality in one of the reference cores (3 mg oil-1) should, as yet, not be considered an effect induced by contamination. ADEMA (1991) suggested that the 'first observed effect concentration' was about 20 mg·-1, which seems in very good agreement with our observations. Moreover, this may explain why, during the field survey, no (adult) Echinocardium were found within 750 m from the platform.

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APPENDIX

Table 11. Data location L5-5 (abandoned), survey May 1989.

Number of analysed samples per station: 6.

Number of identified species.

Mean densities (n.m-2).

Tot. number of ind. per m2 per station.

Number of samples () in which species are present.

r.c.= residual current transect.

Distance to platform (m) Direction of transect	25 r 77°	. с	250 r 77°	·. C.	250 167°		250 257°	
POLYCHAETA								
Aphrodita aculeata	5.0	(3)	. 8	(1)	. 8	(1)	3.3	(2)
Harmothoe lunulata	→ →				1.7	(2)		
Harmothoe longisetis					1.7	(1)		
Harmothoe spec. juv.		(0)		/ 4 \		121	. 8	(1)
Gattyana cirrosa	2.5	(2)	5.0	(4)	5.8	(3)	4.2	(3)
Pholoe minuta	. 8	(1)	15.0	(6)	45.8 4.2	(6) (5)	45.8	(6)
Sthenelais limicola	. 8	(1)	2.5	(1)	4.2	(2)	. 8	(3)
Eteone longa Anaitides mucosa					. 8	(1)	. 0	(1)
Ophiodromus flexuosus	4.2	(5)	1.7	(2)	2.5	(3)	7.5	(5)
Gyptis capensis	3.3	(4)	4.2	(3)	5.0	(3)	4.2	(3)
Synelmis klatti	. 8	(1)	. 8	(1)	. 8	(1)	1.7	(2)
Exogone hebes	. 0	(1)	1.7	(1)	2.5	(2)	. 8	(1)
Nereis longissima			. 8	(1)	. 8	(1)		(- /
Nereis spec. juv.				(- /	1.7	(2)		
Nephtys hombergii	2.5	(3)	8.3	(5)	5.8	(5)	5.8	(4)
Nephtys incisa		(-)		,	1.7	(1)	1.7	(2)
Nephtys cirrosa	1.7	(1)	. 8	(1)	. 8	(1)	1.7	(2)
Nephtys spec. juv.	1.7	(2)	5.8	(3)	5.8	(4)	2.5	(3)
Glycera rouxii	2.5	(3)	5.0	(4)	5.8	(2)	20.8	(5)
Glycera alba			1.7	(2)	4.2	(2)		
Glycera spec. juv.	1.7	(2)	2.5	(3)	. 8	(1)	3.3	(3)
Glycinde nordmanni	. 8	(1)	3.3	(2)	5.0	(5)	5.0	(5)
Goniada maculata	10.0	(6)	7.5	(5)	10.0	(4)	15.8	(6)
Lumbrineris latreilli	316.7	(6)	676.7	(6)	626.7	(6)	865.8	(6)
Lumbrineris fragilis			3.3	(3)	5.0	(4)	9.2	(6)
Driloneris filum					. 8	(1)		
Orbinia sertulata						(0)		
Paraonis spec.		(1)			2.5	(2)		
Spio filicornis	. 8	(1)			. 8	(1)		
Polydora guillei							. 8	(1)
Spiophanes kroyeri Spiophanes bombyx	6.7	(4)	66.7	(6)	104.2	(6)	86.7	(6)
Scolelepis foliosa	0.7	(4)		(0)	104.2	(0)	.8	(1)
Magelona alleni	. 8	(1)						(1)
Chaetopterus variopedatus	1.7	(1)	2.5	(3)	2.5	(3)	1.7	(1)
Tharyx marioni		(- /		(5)	. 8	(1)	2.5	(2)
Chaetozone setosa			5.8	(5)	11.7	(6)	10.8	(6)
Diplocirrus glaucus	. 8	(1)	20.0	(6)	17.5	(6)	10.8	(5)
Scalibregma inflatum		, ,	. 8	(1)		, ,		, ,
Ophelina acuminata				, ,				
Capitella capitata	10.0	(4)	2.5	(2)	. 8	(1)		
Notomastus latericeus	. 8	(1)	4.2	(4)	1.7	(2)	10.0	(6)
Heteromastus filiformis			1.7	(2)	2.5	(2)	5.0	(3)
Owenia fusiformis	. 8	(1)	1.7	(2)	2.5	(3)	2.5	(3)
Myriochele heeri		(0)		(0)		163	1.7	(-)
Lanice conchilega	1.7	(2)	1.7	(2)	8.3	(6)	1.7	(1)
Lagis koreni				/1)	. 8	(1)		
Pectinaria auricoma Sosane gracilis			. 8	(1)	13.3	(6)		
Terebellides stroemi					. 8	(1)	. 8	(1)
Terebellines struemi					. 0	(1)	. 0	(1)

Table 11. Data location L5-5 (abandoned), survey May 1989.

Number of analysed samples per station: 6.

Number of identified species.

Mean densities (n.m-²).

Tot. number of ind. per m² per station.

Number of samples () in which species are present.
r.c.= residual current transect.

Distance to platform (m) Direction of transect	250 347°	500 r.c. 77°	750 r.c. 77°	1000 r.c. 77°	5000 r.c. 77°
POLYCHAETA					
Aphrodita aculeata Harmothoe lunulata Harmothoe longisetis Harmothoe spec. juv. Gattyana cirrosa Pholoe minuta Sthenelais limicola Eteone longa Anaitides mucosa Ophiodromus flexuosus Gyptis capensis Synelmis klatti Exogone hebes Nereis longissima Nereis spec. juv. Nephtys hombergii	2.5 (3)8 (1) 12.5 (4) 27.5 (6) 4.2 (2)8 (1) 5.8 (5) 1.7 (2) .8 (1) 4.2 (3) 3.3 (3)	1.7 (2) 14.2 (5) 15.8 (6) 2.5 (3) 8 (1) 8.3 (3) 2.5 (3) 1.7 (2) 1.7 (2) 6.7 (5)	2.5 (3) 16.7 (6) 15.0 (6) 4.2 (4) 2.5 (3) 5.8 (4) 1.7 (2)8 (1) 1.7 (2) 7.5 (4)	1.7 (2)8 (1) 5.0 (4) 8.3 (5) 5.0 (4) .8 (1) 4.2 (3) 2.5 (2) 2.5 (3) 1.7 (2) 3.3 (2)	.8 (1)8 (1) 18.3 (6) 14.2 (6) 2.5 (2) 1.7 (1) 5.0 (4) 2.5 (2) 2.5 (3) 1.7 (1) .8 (1) 3.3 (3)
Nephtys incisa Nephtys cirrosa Nephtys spec. juv. Glycera rouxii Glycera alba Glycera spec. juv. Glycinde nordmanni Goniada maculata Lumbrineris latreilli Lumbrineris fragilis Driloneris filum	1.7 (2) 4.2 (3) 15.8 (6) 2.5 (3) 2.5 (2) 3.3 (3) 10.8 (6) 436.7 (6) 9.2 (4)	2.5 (3) 3.3 (3) 10.0 (5) .8 (1) 2.5 (3) 10.0 (6) 438.3 (6) 5.8 (4)	2.5 (2) .8 (1) .8 (1) 12.5 (5) .8 (1) 10.0 (5) 10.8 (6) 515.8 (6) 6.7 (4)	3.3 (3) 10.8 (5) 	1.7 (2) 10.0 (4) 2.5 (2) 1.7 (1) 5.8 (4) 6.7 (3) 353.3 (6) 9.2 (5)
Orbinia sertulata Paraonis spec. Spio filicornis Polydora guillei Spiophanes kroyeri	1.7 (2)	.8 (1)	.8 (1) .8 (1) .8 (1)		.8 (1)
Spiophanes bombyx Scolelepis foliosa Magelona alleni Chaetopterus variopedatus Tharyx marioni Chaetozone setosa Diplocirrus glaucus Scalibregma inflatum	40.0 (6) 5.8 (3) 3.3 (3) 9.2 (5) 10.8 (6)	24.2 (6) 15.0 (5) .8 (1) 9.2 (5) 19.2 (5)	46.7 (6) 21.7 (6) 4.2 (4) 9.2 (5) 6.7 (5)	10.8 (3) 3.3 (3) .8 (1) .8 (1) 15.8 (5)	39.2 (5) 1.7 (2) 16.7 (6) .8 (1) 1.7 (2) 2.5 (1)
Ophelina acuminata Capitella capitata Notomastus latericeus Heteromastus filiformis Owenia fusiformis Myriochele heeri Lanice conchilega Lagis koreni Pectinaria auricoma Sosane gracilis Terebellides stroemi	.8 (1) 5.0 (4) 4.2 (3) 6.7 (4) 7.5 (4) .8 (1) 7.5 (5)	2.5 (3) 3.3 (4) 2.5 (2) 2.5 (1) 3.3 (1) 7.5 (6)	1.7 (2) 5.8 (4) 5.0 (3) 2.5 (1) 1.7 (2) 4.2 (4) .8 (1) 6.7 (5)	3.3 (3) 1.7 (1) 1.7 (2) 3.3 (2) 3.3 (3)	.8 (1) 3.3 (3) 3.3 (3) 5.0 (4) 2.5 (2) 4.2 (4)

Table 11 continued.

Astropecten irregularis

, 40 14 22 001141114441								
Distance to platform (m) Direction of transect	25 r 77°	· . c	250 r 77°	.С.	250 167°		250 257°	
MOLLUSCA								
Nucula turgida Thyasira flexuosa Montacuta ferruginosa Mysella bidentata Arctica islandica Acanthocardia echinata	 		1.7 10.8 2.5	(2) (5) (3)	1.7 .8 43.3	(2) (1) (6)	1.7 1.7 50.0	(2) (1) (6)
Dosinia lupinus Venus striatula Mysia undata Abra alba Gari fervensis	2.5	(1)	. 8 9 . 2 8 . 3 1 . 7	(1) (5) (3) (1)	2.5 5.8 2.5 7.5	(2) (3) (3) (4)	4.2 9.2 3.3 4.2	(3) (5) (3) (3)
Cultellus pellucidus Mya spec. juv. Corbula gibba Thracia convexa Cingula nitida	2.5 9.2	(2)	7.5 5.8 .8 4.2	(3) (4) (1) (2)	8.3 .8 10.0 .8	(5) (1) (4) (1) (4)	7.5 5.0 1.7 5.8	(3) (4) (1) (4)
Turritella communis Natica alderi Retusa truncatula Retusa umbilicata Cylichna cilindracea	. 8 1 . 7 6 . 7	(1) (2)	5.8 .8 .8 14.2	(4) (1) (1) (5)	9.2 1.7 40.0	(5) (2) (6)	.8 2.5 10.0	(1) (3)
CRUSTACEA		(.)		(-,		(-)		(/
Pagurus bernhardus Porcellana longicornis Porcellana spec. juv. Macropipus spec. juv.	. 8	(1)	 . 8	(1)			 . 8	(1)
Pinnotheres pisum Ebalia cranchii Cancer pagurus Corystes cassivelaunus	. 8	(1)	 . 8	(1)	1.7 .8	(2)	5 . 8 . 8	(4)
Upogebia deltaura Callianassa subterranea Decapoda larven Nebalia bipes Eudorella truncatula	8.3 8.3 	(5) (4) (1)	7.5 2.5 2.5 .8	(4) (2) (1) (1)	21.7 1.7 .8	(5) (2) (1)	13.3 4.2 1.7	(6) (3) (2) (1)
Iphinoe trispinosa Diastylis bradyi Cirolana borealis Ione thoracica	. 8 4 . 2 	(1)	3.3	(2)	2.5	(3)	4.2	(4)
Melita obtusata Orchomenella nana Ampelisca brevicornis Ampelisca tenuicornis Ampelisca spec. juv.	. 8 3 . 3 . 8	(1) (3) (1)	. 8 . 8	(1)	. 8 1.7	(1)		
Amphilochus spec. Cheirocratus sundevalli Bathyporeia guilliamsoniana Bathyporeia elegans	=======================================		 . 8 	(1)	=======================================		. 8 	(1)
Bathyporeia tenuipes Harpinia antennaria Apherusa spec. Perioculodes longimanus Aora typica	2.5	(3)	 1.7	(1)	1.7 3.3	(2)	. 8 2 . 5	(1)
Caprella spec. ECHINODERMATA					. 8	(1)		
- CONTRODERIMIN								

Table	1 1	CODI	inuea.

Table II continued.					
Distance to platform (m) Direction of transect	250 347°	500 r.c. 77°	750 r.c. 77°	1000 r.c. 77*	5000 r.c. 77°
MOLLUSCA					
Nucula turgida Thyasira flexuosa Montacuta ferruginosa Mysella bidentata Arctica islandica Acanthocardia echinata Dosinia lupinus Venus striatula Mysia undata Abra alba Gari fervensis Cultellus pellucidus Mya spec. juv. Corbula gibba Thracia convexa Cingula nitida Turritella communis Natica alderi Retusa truncatula Retusa umbilicata Cylichna cilindracea	.8 (1) .8 (1) 77.5 (6) 2.5 (3) .8 (1) 1.7 (2) 5.0 (4) .8 (1) 2.5 (3) 6.7 (6) .8 (1) 2.5 (2) 2.5 (3) 18.3 (5) .8 (1) .8 (1) .8 (1) .8 (1) .9 (1)	.8 (1) 47.5 (6)8 (1) 4.2 (2) 2.5 (2) 2.5 (2) 1.7 (2) 2.5 (3) 19.2 (6) 1.7 (2) 2.5 (2) 2.5 (2) 2.5 (8)	2.5 (2) .8 (1) 95.0 (6) .8 (1) 3.3 (3) 5.8 (4) 1.7 (2) .8 (1) 4.2 (4) 5.0 (5) 7.5 (4) 5.8 (3) .8 (1) 18.3 (6)	.8 (1) 13.3 (6) 1.7 (1) 1.7 (2) 5.0 (5) 12.5 (5)8 (1) .8 (1) 8.3 (5) 19.2 (6)	2.5 (2) 1.7 (2) 7.5 (4) 48.3 (5) .8 (1) 6.7 (5) 1.7 (2) .8 (1) 1.7 (1) 3.3 (3) 2.5 (3) 5.8 (4) 5.0 (3) .8 (1) 6.7 (4)
CRUSTACEA					
Pagurus bernhardus Porcellana longicornis Porcellana spec. juv. Macropipus spec. juv. Pinnotheres pisum Ebalia cranchii Cancer pagurus Corystes cassivelaunus Upogebia deltaura Callianassa subterranea Decapoda larven Nebalia bipes Eudorella truncatula Iphinoe trispinosa Diastylis bradyi Cirolana borealis Ione thoracica Melita obtusata Orchomenella nana Ampelisca brevicornis Ampelisca spec. juv. Amphilochus spec. Cheirocratus sundevalli Bathyporeia guilliamsoniana Bathyporeia guilliamsoniana Bathyporeia tenuipes Harpinia antennaria Apherusa spec. Perioculodes longimanus Aora typica Caprella spec.		1.7 (2) 38.3 (6) 1.7 (2)	8 (1)8 (1)8 (1)8 (1)5 (2)7 .7 (2)7 .8 (1)	.8 (1) 16.7 (6) .8 (1)8 (1)8 (1)8 (1)8 (1)8 (1)8 (1)8 (1)8 (1)8 (1)8 (1)8 (1)8 (1)8 (1)8 (1)8 (1)	1.7 (2) 1.7 (2) 1.8 (1)
ECHINODERMATA					
Astropecten irregularis	.8 (1)		.8 (1)		.8 (1)

Table 11 continued.

Distance to platform (m) Direction of transect	25 r 77°	`. C	250 r	·. c .	250 167°		250 257°	
Amphiura filiformis Amphiura chiajei Ophiura albida Ophiura spec. juv. Echinocardium cordatum	19.2 3.3	(5)	385.0 .8 9.2	(6) (1) (3)	766.7 2.5 8.3	(6) (2) (3)	735.8 .8 10.8	(6) (1) (5)
OTHER								
Nemertinea Hydrozoa Turbellaria Phoroniden	P p	(5)	P P 	(6) (1)	P 2.5 p	(6) (2) (2)	. 8	(6) (1)
Harp. copepoda Parasitaire copepoda Oligochaeta Holothuroidea Sagitta spec. Echiurida Sipinculida Ascidiacea	1.7 .8 .8 .8	(1) (2) (1) (1) (3)	P 2.5	(2) (2) (2) (6)	.8 P 3.3 1.7 3.3 75.0	(1) (2) (4) (2) (2) (6)	5.0 5.8 103.3	(4) (5) (6)
Tot. nr. of ind. (n.m-²) Nr. of identified species P=present, not counted	470 39		2.5 1381 55	(3)	1969		. 8 2148 52	(1)

Table 11 continued.

Distance to platform (m) Direction of transect	250 347°	500 r.c. 77°	750 r.c. 77°	1000 r.c. 77°	5000 r.c. 77°
Amphiura filiformis Amphiura chiajei Ophiura albida Ophiura spec. juv. Echinocardium cordatum	1133.3 (6) 1.7 (1) 4.2 (3)	715.0 (6) .8 (1) 2.5 (2) 2.5 (3)	847.5 (6) 1.7 (2) 4.2 (4)	330.8 (6) 14.2 (5) .8 (1)	320.0 (6) .8 (1) 1.7 (2) 6.7 (5)
Nemertinea Hydrozoa Turbellaria Phoroniden Harp. copepoda Parasitaire copepoda Oligochaeta Holothuroidea Sagitta spec. Echiurida Sipinculida Ascidiacea	P (6) P (1) .8 (1) P (3) .8 (1) .8 (1) P (1) 2.5 (3) 106.7 (6) .8 (1)	P (6)8 (1) P (1) .8 (1) 3.3 (3) 2.5 (2) .8 (1) .8 (1) 73.3 (6)	P (6) P (5) .8 (1) P (1) 3.3 (3) .8 (1) 9.2 (5) 81.7 (6)	P (6) 2.5 (3) 2.5 (1) 47.5 (6)	P (6) 1.7 (2) P (4) .8 (1) 2.5 (3) 2.5 (3) 101.7 (6)
Tot. nr. of ind. (n.m-²) Nr. of identified species P= present, not counted	2113 62	1585 51	1893 58	844 46	1121 60

Table 12. Boxcosms L5-5, natural infauna. Mean densities $(n.m^{-2})$ at the start of the incubation period (Sept. 1989) and at the end (Dec. 1989).

		00 -	24	FO -		25 m
	start	00 m end	start	50 m end	start	end
POLYCHAETA	Start	end	Start	end	start	ena
. 02 / 0111/2						
Aphrodita aculeata	2.8			5.0	8.5	3.2
Harmothoe lunulata		. 8				
Harmothoe longisetis		-				1.6
Harmothoe spec. juv.					1.4	
Gattyana cirrosa	43.7	22.4	38.0	18.0	4.2	4.0
Pholoe minuta	25.4	11.2	47.9	11.0		1.6
Sthenelais limicola	4.2	1.6	4.2	1.0	1.4	
Anaitides groenlandica			1.4			
Anaitides mucosa			1.4			
Anaitides maculata			1 4	2.0	1.4	1 6
Ophiodromus flexuosus	4.2		1.4	2.0	1.4	1.6
Gyptis capensis	7.0	8.0	8.5	2.0	9.9	1.6
Synelmis klatti	2.8	4.0	9.9	7.0	12.7 5.6	6.4
Exogone hebes	5.6	2.4	12.7		1.4	
Nereis longissima	8.5 4.2	3.2	7.0	4.0	1.4	. 8
Nereis spec. juv.	4.2	4.8	4.2	4.0	1.4	2.4
Nephtys hombergii	4.2	. 8	2.8	4.0		. 8
Nephtys incisa Nephtys spec. juv.	1.4	2.4	2.0	1.0		. 0
Glycera rouxii	8.5	11.2	5.6	4.0	2.8	4.8
Glycera alba		2.4	2.8	1.0		
Glycera spec. juv.	4.2	. 8	2.0	5.0	7.0	4.0
Glycinde nordmanni	4.2	. 8		1.0	1.4	
Goniada maculata	9.9	2.4	8.5	2.0		1.6
Lumbrineris latreilli	283.1	128.8	460.6	382.0	367.6	237.6
Lumbrineris fragilis	2.8	4.0	7.0	5.0	7.0	2.4
Driloneris filum						. 8
Paraonis spec.	1.4		1.4			
Poecilochaetus serpens	1.4					
Polydora pulchra				1.0		
Polydora guillei	4.2	8.8				. 8
Spiophanes bombyx	18.3		16.9	3.0	9.9	
Scolelepis foliosa				1.0		. 8
Magelona papillicornis	7.0		1.4		1.4	
Chaetopterus variopedatus	46.5	32.8	53.5	35.0	9.9	12.8
Tharyx marioni	2.8					
Chaetozone setosa	1.4		2.8	1.0	1.4	
Diplocirrus glaucus	4.2		15.5	3.0	2.8	1.6
Scalibregma inflatum	5.6		7.0	1.0	8.5	. 8
Ophelina acuminata	1.4	. 8	2.8			
Capitella capitata			4.2	1.0	4.2	
Notomastus latericeus	25.4	2.4	18.3	2.0		. 8
Heteromastus filiformis	19.7	1.6	35.2	2.0	1.4	
Owenia fusiformis	8.5	2.4	12.7	1.0	1.4	. 8
Myriochele heeri	1.4	. 8	1.4		2.0	
Lanice conchilega	5.6	7.2	1.4		2.8	
Lagis koreni	4.2		2.8			
Pectinaria auricoma	2.8	6.4	1.4	1.0		
Amphicteis gunneri			15.5		11.3	
Sosane gracilis Lysilla loveni	1.4		1.4		11.3	
Terebellides stroemi	1.4		1.4		1.4	. 8
recepetitues attoemi	1.4		1.4		1.4	. 0
MOLLUSCA						
Nucula turgida	4.2	4.0	1.4	2.0		3.2
Thyasira flexuosa	2.8	2.4	2.8	3.0		2.4
Lepton squamosum	1.4	6.4				

Table 12 continued						
Table 12 Continued	50	00 m	2	50 m		25 m
	start	end	start	end	start	end
Montacuta ferruginosa	12.7	14.4				
Mysella bidentata	33.8	12.8	95.8	44.0	2.8	. 8
Arctica islandica		. 8	1.4			
Acanthocardia echinata	7.0		2.8		4.2	
Dosinia lupinus			1.4		1.4	4.0
Venus striatula	2.8	1.6		4.0	11.3	. 8
Mysia undata	4.2	. 8	2.8	4.0	1.4	. 8
Mactra corallina					1.4	
Spisula spec. juv.	7.0	1.6	8.5		11.3	
Abra alba	2.8	1.6	29.6	1.0	12.7	
Gari fervensis		. 8				
Cultellus pellucidus	8.5	1.6	8.5	1.0	9.9	. 8
Mya truncata	1.4					
Corbula gibba	2.8	3.2	5.6	8.0		5.6
Thracia convexa	1.4	1.6	5.6	2.0	2.8	3.2
Cingula nitida	12.7	24.8	39.4	14.0	25.4	14.4
Turritella communis	11.3	10.4	9.9	4.0	4.2	2.4
Natica alderi		4.8	2.8	9.0	1.4	8.8
Buccinum undatum						. 8
Cylichna cilindracea	12.7	7.2	18.3	10.0	1.4	1.6
Philine scabra	1.4		11.3	21.0	31.0	18.4
CRUSTACEA						
	15.5	15.0	0.1 1		0 5	2.4
Processa parva	15.5	15.2	21.1	6.0	8.5	2.4
Pontophilus trispinosus	2.8					
Macropipus holsatus		1 6			7.0	
Macropipus spec. juv.	4.2	1.6	5.6	3.0	7.0	. 8
Ebalia cranchii	2.8	. 8	2.8	2.0	2.8	. 8
Corystes cassivelaunus		. 8				
Upogebia stellata	2.8	1.6	1 . 4			. 8
Upogebia deltaura	2.8	3.2				. 0
Upogebia spec. juv.	5.6		2.8 73.2	44.0	39.4	16.8
Callianassa subterranea	93.0	69.6	1.4	44.0	39.4	10.0
Nebalia bipes			1.4		2.8	
Schistomysis ornata			1.4			
Eudorella truncatula	2.8		4.2		2.8	
Diastylis bradyi	2.0				7.0	
Diastylis spec. juv.		. 8				. 8
Ione thoracica	4.2	. 0	5.6			
Ampelisca brevicornis	19.7		14.1		15.5	
Ampelisca tenuicornis	8.5		7.0		2.8	
Ampelisca spec. juv.	1.4					
Bathyporeia tenuipes	1.4					
Harpinia antennaria Perioculodes longimanus	1.4				1.4	
Aora typica	5.6		2.8			
Microdeutopus spec.	1.4					
Caprella spec.			1.4			
ECHINODERMATA						
Astropecten irregularis						2.4
Amphiura filiformis	690.1	171.2	1390.1	532.0	250.7	119.2
Amphiura chiajei	1.4		7.0			
Ophiura texturata		1.6				
Ophiura albida	7.0	2.4	4.2		1.4	
Ophiura spec. juv.	5.6	1.6	11.3	8.0	14.1	4.0
Asteroidea spec. juv.					1.4	
Echinocardium cordatum	26.8	15.2			2.8	

Table 12 continued							
	500	00 m	25	0 m	25 m		
OTHER TAXA	start	end	start	end	start	end	
Nemertinea	Р	Р	Р	Р	Р	Р	
Hydrozoa					P		
Turbellaria		. 8		1.0			
Phoroniden	P	P	Р	P	P	P	
Harp. copepoda			1.4				
Parasitaire copepoda	2.8		8.5				
Holothuroidea	2.8	4.0	9.9	2.0	7.0	1.6	
Sagitta spec.	8.5		7.0		8.5		
Echiurida		5.6	8.5	3.0	1.4		
Sipunculida	136.6	72.8	78.9	19.0	7.0	7.2	
Cerianthus spec.				3.0			
Ascidiacea			1.4				
Foraminiferida			2.8				
Tot. numbers of ind.	1778	745	2757	1257	1003	527	
Nr. of identified species P= present, not counted	67	51	63	44	50	43	

Table 13. Locations investigated in 1985, 1986, 1987, 1988 and 1989.

Location	Position	Orilling activities	Area	Survey	Remarks	Report	
P6b	52°42′?′′N 03°50′?′′E	1 well, drilled with low-tox OBM with high aromatic content; drilled in April-June'85	Erosion zone, sand silt fraction (<63 µm) < 1%	Sept. 85		Mulder et al., 1987	
F18/8	54°05′30′′N 04°59′15′′E	1 Well drilled with W8M in April 1986	Sedimentation area, silt fr. 10 - 20% depth 40 m. Deserted location	May/June 186		Mulder et al., 1987	
L4a	53*43'40''N	Previous drilling with	Transition zone,	May '86	'Baseline' (only	Mulder et al., 1988	
	04°06′15′′E	WBM in Jan. 85; drilling of 2 wells in AugOct. 1986 with low-aromatic	fine sand with silt (≈ 15%)	Sept.'86	WBMs discharged) Drilling not yet	Mulder et al., 1988	
		OBM; tot. emission: 912	depth 35 m. Platform present	Febr. 187	finished	Daan et al., 1990	
		tonnes. OBM: 555 tonnes; oil:178 tonnes	· · · · · · · · · · · · · · · · · · ·	the state of the s	June'87	Additional photo- survey (Sept. 187)	Daan et al., 1990
K12a	53'28'36.2''N 63'47'19.4''E	5 Wells drilled diesel and low-tox OBM, between	Transition zone,	Sept. '85		Mulder et al., 1987	
	03 47 13.4 E	Febr. 183 and Nov. 184: tot. emission: 2278 tonnes. OBM: 1082 tonnes;	silt (5 - 10%) depth 28 m.	Sept. 186 Sept. 187	Additional photo- survey	Mulder et al., 1988 Daan et al., 1990	
		oil: 393 tonnes		Sept.'88	4 boxcosms	Daan et al., 1990	
F18/9	54°06′09.8′′N 04°45′25.1′′E	1 Well drilled with OBM in May'87; tot. emission: 1912 tonnes; OBM: 819 tonnes; oil: 283 tonnes	Sedimentation area, silt (10 - 20%); depth 42 m. Deserted location	June 188	17 boxcosms (collected Sept. 88)	Daan et al., 1990	
L5-5	53°48′33.1′″N 04°20′54.4′″E	1 Well drilled with low- aromatic OBM, July-Nov. 1988; tot. emission: 1347 tonnes. WBM: 308 tonnes; OBM: 148 tonnes; oil: 44 tonnes	Transition zone, fine sand and silt (≈15 %), depth ≈41 m), Deserted location	May '89	14 boxcosms (collected Sept. 89)	This report	

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