
Effects of Water Based Drilling Muds on Recolonization of Sandy Soft Bottom Communities

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Forord

Denne oppgaven ble utført ved Biologisk institutt i perioden 2009-2010 som en del av prosjektet “Parameterisation of the Environmental Impacts on Bottom Fauna of Water-based Drilling Fluids and Cuttings – Field and Meso-cosm Experiments” (PEIOFF-FAME), med støtte fra Norsk forskningsråd.

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Abstract

This study is part of a larger research project, Parameterisation of the Environmental Impacts on Bottom Fauna of Water-based Drilling Fluids and Cuttings – Field and Mesocosm Experiments (PEIOFF-FAME). The goal was to investigate the impact of drill cuttings when drilling with water-based drilling fluids on recolonization of a benthic ecosystem. Drill cuttings from oil and gas installations contain either oil-based, synthetic or water-based muds. Today only cuttings from water-based muds are allowed to be discharged. Drill cuttings from water-based muds are expected to cause only minimal damage to the biota surrounding the installations offshore, but this statement has not been tested experimentally in the field.

My approach was a field experiment where defaunated sandy sediment treated with water-based cuttings was deployed at the seafloor as substrate for settling benthic larvae.

Test sediment was sampled in the Oslofjord in March 2007. Drill cuttings were added in a pattern of 0, 6 and 24 mm top layer in the boxes. Four experimental frames were deployed at 60 m depth on 21st of March and recaptured 6 months later on 24th of September. The data were investigated by univariate and multivariate statistical techniques.

The polychaet *Ophelina acuminata* showed a significant decline in abundance as a function of layer thickness of drill cuttings, and there was an overall negative trend in recolonization with treatment. The echinuran *Echiurus echiurus* showed a weak positive trend which was close to significant. A weak positive trend was also found for number of taxa and for the Hurlbert's rarefaction diversity index. There was no grouping of the boxes of test sediment as function of treatment, but there was a clear grouping as a function of frame. The test sediment had a markedly larger grain size than the drill cuttings, while the grain size in the fine material was similar to the drill cuttings. Analysis of the oxygen penetration depth showed a weak negative trend as a function of added drill cuttings.

Settling communities may not be as sensitive as established communities. However, since there was a negative trend as a function of added drill cuttings it is not impossible that natural variation covers an effect of the drill cuttings in the field.

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Chapter 1

Introduction

1.1 Characteristics of drilling muds and cuttings

Drilling mud is a mixture of clay, chemicals, water or oil. There are three types of drilling muds; water based (WBM), oil based (OBM) and synthetic based (SBM). The mud has several important functions when drilling for oil. It lubricates and cools the drilling bit during drilling and it also brings mass from the drilling to the surface. The drilling mud will also prevent that the wall in the drilling hole collapses and it keeps the pressure in the well under control. If the weight of the drilling muds is too low, the pressure in the well can push oil or gas to the surface (blowout). If the weight of the drilling muds is too high, the mud can disappear in to the reservoir and close the pores (OLF, 2009). Other functions of drilling mud are: seal permeable formations of the borehole, suspend cuttings when circulation is interrupted such as when adding a new piece of drillpipe, support part of the weight of the drillstring through buoyancy, and ensure the securing of important information about the formation being drilled to permit its successful evaluation (Hinwood et al, 1994).

A type of weight material is used to apply counter-pressure in the process. Common materials are barite, ilmenite, hematite and brines where barite and ilmenite are the most common types. Barite (BaSO_4) is a mineral consisting of barium sulphate. Barium is inactive, but may have a negative effect on biota if the concentrations become high (Olsgard and Gray, 1995). Barite is more or less polluted with Mercury and Lead. It is possible to clean the Barite to remove the toxic substances. The other most common material is ilmenite (FeTiO_3). Since both ilmenite and barite are inactive they make useful tracers of dispersion and transport of discharges related to drilling activities such as drilling muds and cuttings (Neff, 2005; OLF, 2009). About 1000 products available for formulation of

drilling fluids and the total number of ingredients in most drilling fluids is in the range of 8-12 (Holdway, 2002).

Drill cuttings are particles of crushed rock created by the grinding action of the drill bit as it penetrates the earth and is brought to the surface. The diameter is usually less than 1 cm (Neff, 2005; OLF, 2009). Drill cuttings (only water-based) are released into the environment after separation from the mud on the platform (Davies et al, 1984).

There are no general restrictions on the release of drill cuttings from water based muds (WBM) into the environment. The reason for this policy is that bringing the drill cuttings to shore will give higher releases in the air and demand more space for storage. Some areas are protected; no drilling fluid or drill cuttings are allowed to be released in the Barents sea and Lofoten and the areas around (Klif, 2009)(Norwegian Climate and Pollution Agency). Drill cuttings from topholes can normally be released in the Barents sea, under the condition that the release does not contain substances with unacceptable environmental properties and only in areas where the potential harm on vulnerable environmental components are considered low. Klif has determined that it is important to do thorough research on possible effects of future releases of cleaned cuttings with oil content less than one weightprocent (Klif, 2009). There is a zero release policy for substances harmful to the environment, and drill cuttings are part of this policy. Practically, this means that drill cuttings from water based muds are released, except in vulnerable ecosystems. The two other types of cuttings are not allowed released other than under particularly demanding conditions (Klif, 2010).

The release of drill cuttings spreads different types of contaminants to the bottom of the sea. Around many of the installations there are high heavy metal concentrations with negative biological effects; copper, cadmium and zinc (Olsgard and Gray, 1995). The heavy metals in drill cuttings are likely to be distributed in the same manner as Ba (Re-naud et al, 2008). Total hydrocarbon (THC) concentrations show a clear decline over time in the field in the surface layer of the sediment, which is probably because of discontinuation of OBM use in 1991 (Klif, 2010; Olsgard and Gray, 1995). When it comes to fauna, monitoring programmes have been performed since the mid 1970s and annually since 1985. Many of the oil fields have been sampled and the fauna examined by univariate and multivariate methods, looking at both biological and environmental factors through classifications and ordinations (Olsgard and Gray, 1995). Previous results suggest that the communities surveyed are partly structured by depth related factors (organic content, grain size), but there is some indication that disturbance makes up a much of the secondary axis for most of the fields with installations. In multivariate analysis (Multi Dimensional Scaling, MDS) of observations of benthic fauna beneath and around explo-

ration wells (later oil platforms), stations are often grouped regardless of their distance from the installation, which suggests that other factors that are specific to each field are mostly responsible for structuring communities (Olsen et al, 2007). It is therefore useful to look at results from field experiments where it is easier to rule out confounding factors that are specific to each field.

Discharge of solid waste (drill cuttings) from offshore drilling operations is often contaminated by an organic phase from the mud to facilitate drilling. When sinking, most of the cuttings will place themselves near the installation, and can stay in the environment for many years (Schaanning and Bakke, 2006). Dispersion of particles from drill cuttings are greatly influenced by their particle size and the prevailing current regime. The distribution usually follows the currents, often producing an ellipsoidal distribution at the seafloor. However, it is believed that cuttings from oil-based mud drilling, fall more directly to the seabed compared to WBM and SBM as a result of agglomeration (Davies et al, 1984). It is therefore common to find deposits of oil based drilling muds released before 1993 when cuttings from OBM with oil content $>1\%$ was prohibited to release on the Norwegian shelf (Schaanning and Bakke, 2006).

Since the beginning of the Norwegian oil adventure several surveys have been executed in the field to monitor the fauna around the different installations (Olsgard and Gray, 1995; Gray et al, 1999; Renaud et al, 2008). Historically, most drilling operations in the North Sea have used WBM. However, in some drilling operations it is difficult to use WBM primarily because of hole instability caused by the swelling of water-absorbing rock. Problems of this type can be greatly alleviated by using mud suspended in an oil base instead of water (Davies et al, 1984).

1.1.1 Water based drilling muds (WBM)

Originally, all drilling muds used in drilling operations were in an aqueous solution, but these were later replaced by oil-based because it was preferable in drilling operations (Olsgard and Gray, 1995). Water based drilling muds (WBM) consist of fresh or salt water containing a weight agent (usually barite: BaSO_4), clay or organic polymers, and various inorganic salts, inert solids, and organic additives to modify the physical properties of the mud so that it functions optimally (Olsgard and Gray, 1995). Ingredients list for water based mud can be divided into 18 categories:

Weighting materials	Alkalinity, pH-control additives	Filtrate reducers
Viscosifiers		Flocculants
Thinners, dispersants	Bactericides	Foaming agents

Lost circulation materials	Emulsifiers	Temperature stability agents
Pipe-freeing agents	Defoamers	Lubricants
Calcium reducers	Shale control inhibitors	
Corrosion inhibitors	Surface-active agents	

(Neff, 2005)(and references therein). Water based drilling muds also contain several metals, the ones of greatest concern because of their toxicity and/or abundance in drilling muds include arsenic, nickel, chromium, barite, cadmium, copper, iron, lead, mercury and zinc (Neff, 1987; Neff et al, 2000). A typical discharge of drill cuttings from WBM will contain between 5% and 25% drilling muds discharge after passing through the solids control equipment on the platform. Drill cuttings produced during drilling with WBM may contain a small amount of petroleum hydrocarbons. These may originate from spotting fluids and lubricants added to the mud, or from geological strata penetrated by the drill (Neff, 2005). Water based drilling muds contain a appreciably amount of organic matter, and one important ingredient is glycol. This substance is highly degradable, and with low toxicity (Schaanning et al, 2008). Degradation may suppress H₂S and make the environment highly anoxic (M Schaanning, 2010, pers. com). WBM are more fine grained (than OBM) and can be expected to lead to a wider dispersion of barite (Olsgard and Gray, 1995). Because of the quick dispersion, cuttings from WBM do not affect the environment in the same way as cuttings from OBM and SBM (Neff, 2005).

1.1.2 Oil based drilling muds (OBM)

Oil based drilling muds contain a refined petroleum product, usually diesel fuel, mineral oil or a parafin mixture (Neff, 2005). In the beginning, OBMs contained diesel oil. This was later exchanged with mineral oil, mainly because of the work conditions for the workers at the oil platforms. Mineral oil did not significantly improve the environmental conditions (Bakke et al, 1986). Drill cuttings from OBM with an oil content of maximum 6 % on Norwegian sector was before 1st of January 1993 permitted to release during drilling operations. After 1st of January 1993 intentional discharge of oil-contaminated cuttings was prohibited on the Norwegian continental shelf (Gray et al, 1999). Under drilling conditions where the technical properties of OBM are needed for safety or operational reasons, OBM may be used after approval of the Norwegian authorities (Klif, 2009), for instance when drilling in shale formations (Neff, 2005). The cuttings from OBM that are allowed to be released must have a oil content below 1 % (Berge, 1993). If OBM cuttings are used it will pass through treatment facilities such as shale shakers, desanders, desilters and mud cleaners to separate the cuttings from the mud and maintain

the desired mud formulation (Davies et al, 1984). With the discharges of OBM, appreciably amounts of hydrocarbons and heavy metals were released into the environment. High oil concentrations were found close to some of the major OBM operations (both diesel and alternative mud users), typically between 1000 to 10000 times background within 250 m of the platform. The concentrations fall steeply, generally reaching background levels within 3000 m. The extent of biological effects from oil-based mud cuttings is greater than the extent from water-based mud cuttings. Beyond the area of physical smothering, the effects of oil-based mud cuttings may be because of organic enrichment of the sediment and/or the toxicity of certain fractions of the oils used, such as aromatic hydrocarbons. It is not possible from the present available results to distinguish between the ecological effects of diesel mud and alternative base mud (Davies et al, 1984).

The amount of drill cuttings released from OBM between 1983 and 1992 are estimated to be around 300,000 tonnes distributed on average 92 wells per year. Heights of the cuttings piles varied between <2m to 15m, with the most cuttings piles being less than 2m or 7-15m tall (Cripps et al, 1998).

1.1.3 Synthetic based drilling muds (SBM)

To replace OBM, synthetic based muds (SBM) were developed. They are contaminated with organic fluids such as ethers, esters and olefins that were meant to replace the mineral oil in OBM (Schaanning and Bakke, 2006). Common substances in SBMs are olefins, esters, ethers, polyalphaolefins, glycols, glycerins and glucosides. These chemicals are intended to make the muds having the advantages of oil muds but with the handling and disposal characteristics of water muds (Caenn and Chillingar, 1996). In Norway, synthetic-based drilling muds were used in the period around 1990-2000. Around 2000 it was forbidden to use SBM with organic content $> 1\%$ because of the effects on the environment. Benthic effects of SBM were recorded up to 500 m from the platform (Jensen et al, 1999). SBM contains little substances harmful to the environment, but the high organic content leads in many cases to anoxia and bad conditions for the benthic fauna (Neff et al, 2000).

1.2 Recolonization of benthic fauna

One can assume that studies on recolonisation of contaminated sediments provide relevant information about species tolerance of contaminants (Trannum et al, 2004). Recolonization and succession in soft sediments have been studied extensively (see (Gray, 1981;

Probert, 1984; Thrush, 1991)). Dominance in the early phase of recolonization appears to be determined by the availability of benthic species/larvae at the time the habitat was made available (Grassle and Grassle, 1974; Pearson and Rosenberg, 1978). Dense aggregations of polychaete tubes are often considered to stabilize sediments by altering the characteristics of near-bed waterflow and have been shown to be particularly important in affecting early stages of succession (Sanders et al, 1962; Fager, 1964; Gallagher et al, 1983; Levin, 1985). Timing of initial colonization seems to be an important factor that controls development of experimental populations, since postlarvae and juveniles are available as potential colonizers change depending on the season of the year (Diaz-Castañeda et al, 1993). Initial recolonization after defaunation in marine soft bottom sediment is predominant by opportunistic species with r-selected life-history traits, such as capitellid and spionid polychaetes. Species termed opportunists have evolved life-history characteristics such as rapid dispersal and high reproductive rates that allow them to locate and colonize disturbed patches rapidly so that these species occur early in succession. Other species which are better resource competitors invade later and displace the opportunists only to be displaced by succeeding colonists themselves (Thistle, 1981).

Certain qualities characterize species that are typical in the initial phase of recolonization; (1) opportunistic (many reproductions per year, high recruitment, rapid development, early colonizers, high death rate), (2) small, (3) sedentary, (4) deposit feeders (mostly surface feeders) and (5) brood protection (lecithotrophic larvae). Average life span, generation time and population growth rate set the pace of population processes (Zajac et al, 1998)

Biological interactions become more important in the later successional stages, and accumulation of toxic metabolites may also become a limiting factor. The abundant initial colonizers may often be replaced at a later successional stage (Grassle and Grassle, 1974; Connel and Slatyer, 1977; McCall, 1977).

Some biotic processes influence the process of recolonization. Facilitation comprises interactions in which one group of organisms enhances the establishment of another. Inhibition results in groups of organisms preventing or significantly reducing the establishment of another group. This may occur via competition for resources such as food and/or space. Predation can also be added to the list (Zajac et al, 1998).

Hydrodynamics also affect the distribution of food resources which may have a critical role in shaping successional dynamics in soft-sediment habitats (Thistle, 1981).

The mode of recolonization (e.g., contribution of larval vs. post-larval dispersal) of disturbed habitats appears to be scale dependent (Gunther, 1992). Thus, understanding how the spatial scale of recolonization influences this mode is important in developing

realistic models of patch and community dynamics (Smith and Brumsickle, 1989; Thrush et al, 1996). As the spatial scale of disturbances increases, the duration of successive recovery should increase (Zajac et al, 1998).

The experimental environment facilitates the survival of young organisms on defaunated test plots. There is no competition and lower rates of predation when compared with the natural environment, and there is a high content of organic matter which favours the settlement of deposit feeders (Zajac et al, 1998). In recolonization experiments with defaunated sediments the abundance will increase to a certain point, reach peak after some time, followed by a decline in number of individuals. The number of species shows a similar trend (Lu and Wu, 2000).

1.3 Settling of benthic larvae

“Settlement“ is the process by which planktonic larva moves toward the substratum, explores, attaches to the substratum, and begins its benthic life (Quian, 1999). Settling of benthic invertebrate larvae is an important part of the recolonization process. Disturbances such as the release of drill cuttings can possibly be a disturbance that can influence this process. The larval and juvenile stages are considered the most vulnerable stages of marine invertebrates, and might be particularly vulnerable to pollution (Woodin, 1976; Jablonski and Lutz, 1983), in this context from drill cuttings. The larvae of opportunistic species normally have little or no selectivity in their substratum requirements (Pearson and Rosenberg, 1978).

Larval development can be split into to groups; planktonic and benthonic, and some species brood larvae to different extents and release them into the plankton for various periods of time (Olive and Clark, 1978). We have a good understanding of the settling of benthic invertebrate larvae, but the planktonic phase of benthic larval organisms is less known (Eckman, 1996). Life cycles of most benthic marine invertebrates species include microscopic, free-living dispersive stages that may be feeding (planktotrophic) or non-feeding (lecithotrophic) (Pechenik, 1999). Some controlled experiments have been carried through to learn more about the settlement stage in the life cycle of benthic organisms. Species of marine invertebrates with a planktonic larval stage differentiates into a planktotrophic trochophore and then a metatrochophore (Marsden et al, 1990). The planktonic phase of invertebrate larvae may last from minutes to months. (Pawlik, 1992). Just before metamorphosis and settlement, the larvae become demersal, moving slowly along the bottom. Some species show a clear preference for certain habitats (Marsden et al, 1990).

It is possible for post-larvae to move on mudflats, although it is usually assumed that postlarvae are only capable of moving short distances (Thrush et al, 1996; Smith and Brumsickle, 1989). The process for polychaet larvae settlement is a dynamic event because the larvae can leave one site and select another for settlement (Quian, 1999). However, sooner or later the larvae has to settle because it will eventually metamorphose. Interaction between the larvae and the substratum will therefore determine the site of larval settlement on small spatial scales and may determine postsettlement mortality of larvae (Quian, 1999) (and references therein). This interaction can be affected by biological, physical or chemical factors, such as community structures, presence or absence of natural inducers released by conspecific individuals, biofilms, prey species, or sympatric species (Quian, 1999) (and references therein). One author suggests that chemical cues from adults or adult sites in the form of dissolved material may induce orientation behaviour by presettlement larvae (Burke, 1986). Several compounds can induce settlement in marine larval polychaetes; (1) juvenile hormones, (2) free fatty acids, (3) polysaccharides, (4) proteins and small peptides, (5) amino acids, (6) inorganic ions and (7) neurotransmitters (Quian, 1999) (and references therein). Water currents and flow dynamics may determine both vertical and horizontal distribution of larvae in a water column. Swimming and adhesive behaviour is of some importance if the larvae are moved near the substratum by currents (Quian, 1999). Video observations of competent larvae have shown that the animal swim primarily on the horizontal plane, about a centimeter above the bed, frequently testing the substratum by swimming down to the bottom and swimming away in the absence of an appropriate cue. This is the first demonstration (to the authors knowledge) that infaunal species can actively select a preferred habitat in a realistic, turbulent flow (Butman et al, 1988).

The following factors has been shown to inhibit the successfull settlement of some benthic larvae; oxygen depletion (Arntz, 1977), sediment instability (Rhoads and Young, 1970; Rhoads et al, 1977) and pollution (Bellan et al, 1972). Drill cuttings have properties that can influence the settling of larvae and possibly cause such conditions to develop in benthic communities.

After settling, postsettlement mortality and emigration can determine the success of the larvae that initiate metamorphosis (Watzin, 1983; Luckenbach, 1984; Eyster and Pechenik, 1987)

1.4 Previous related studies

Effect of Barite (BaSO_4) on development of estuarine communities has been studied in a laboratory experiment. The authors found that large quantities of this compound might adversely affect the colonizing of benthic animals (Tagatz and Tobia, 1978). An experiment with different level of exposure from drill cuttings on larva was executed to observe the effect on larval development, where the largest levels of drill cuttings added showed lower densities and fewer species (Menzie, 1984). Field experiments on benthic recolonization and chemical changes in response to various types and amounts of cuttings, both water based and oil based have been done in Raunefjorden, western Norway. The fauna was greatly affected by the OBM drill cuttings, although effects on WBM drill cuttings were not present (Bakke et al, 1986). The effects on defaunated sediment contaminated with crude oil was studied in two Norwegian fjords with unequal eutrophication status. The unpolluted Raunefjord in Western Norway was affected by the oil with lower densities, caused by toxic response to the oil directly leading to increased mortality (Berge, 1990). In an experiment with treated drill cuttings little effects were observed on recolonization of benthic communities, but severe effects on oil-based cuttings with high oil content (15-20%) were observed (Berge, 1993). Assemblages of recruiting soft-sediments contaminated by petroleum hydrocarbon were significantly affected in a field experiment at Casey Station, Antarctica (Stark et al, 2003). Effects of WBM cuttings were observed in a mesocosm experiment in established soft-bottom communities (Trannum et al, 2009)(also in PEIOFF-FAME). A study of effects of WBM cuttings in the field with its current composition is lacking.

1.5 Objective of this study

This study is part of a larger research project, Parameterisation of the Environmental Impacts on Bottom Fauna of Water-based Drilling Fluids and Cuttings – Field and Mesocosm Experiments (PEIOFF-FAME), and some results or measurements from other parts of the experiment have therefore been included wherever necessary. The main objective of PEIOFF-FAME are to provide quantitative results on effects of WBM drill cuttings discharges on bottom fauna through new mesocosm- and field experiments, together with existing results and literature observations and quantitative observations on the most important factors relevant for a realistic parameterisation of the ERMS- model (Environmental Risk Management System) (Olsgard et al, 2005).

The main objectives of this study are (through a field experiment):

- to assess the relationship between the dose of WBM cuttings and the effects on the benthic ecosystem; faunal composition, diversity, individual species, groups of species or ecological groups.
- to investigate if change in environmental variables such as oxygen penetration depth, grain size and total organic carbon can explain possible negative effects of the drill cuttings (Olsgard et al, 2005).

This part of the study was done on coarse sediment (sand), another was done on fine sediment (clay). A third part had coarse sediment (sand) as “treatment” on fine sediments and fine sediment (clay) as “treatment” on coarse sediment as controls, on order to look for particularly for the effect of grain size. In this experiment the boxes without treatment (only sand) serve as controls and will be treated as controls. The null hypothesis tested is that there is no decline in abundance or diversity as a function of the added WBM drill cuttings thickness layer in the experiment. The results from the experiments will in itself be highly relevant for the future management of drilling activities in temperate, boreal and arctic waters (Olsgard et al, 2005).

Chapter 2

Materials and Methods

2.1 Test sediment, eksperimental design and fieldwork.

Sediment samples were collected at two locations at 116 and 96 m depth in the outer part of inner Oslofjord (59,643°N/10,629°E, /59,652°N/ 10,6213°E) representatives of a fine and coarse sediment with a 0.1 m² Van veen grab on the 3rd and 5th of March 2007 (figure 2.1). After the collection, the sediment was stored in 120-L PVC boxes for a maximum of 3 days at 8-10°C. All sediments were mixed separately in batches of 30 L in a cement mixer for 1 hour each. After mixing, a 10-cm-thick layer was filled into 0.1-m² propene plastic boxes (29 x 32 x 13cm) and frozen at -20°C for at least 5 days for additional defaunation of the sediments and to avoid loss of sediment during deployment. The drill cuttings used in this experiment is water based, with ilmenite as weight material and contains glycol. These cuttings were used in the Barents sea before disposed on shore on a disposal site.

A total of 64 boxes with sediment, with or without cuttings were placed into four separate 1.5 x 1.5 m² aluminium frames (figure 2.2), with 16 boxes in each frame. The experiment started on the 21st of March 2007 when the frames were deployed at 60 m depth at an unpolluted location in the Oslofjord just outside Norwegian Institute of Water Research's (NIVA) research station, Solbergstrand (figure 2.1). The four frames were placed at two different sites (in the same area as the test sediment sampling) at each side of the fjord to avoid pseudoreplication. Frame A and B and C and D were placed at the same location. The experimental frames were positioned about 20 cm above the seabed.

The PVC-boxes contained either sandy silt or clay. The drill cuttings were added in a 6 or 24 mm top layer (table 2.1). Sandy silt and clay were also added in 6 or 24 mm top layer and the rest were controls. The marked boxes (table 2.1) are the samples included

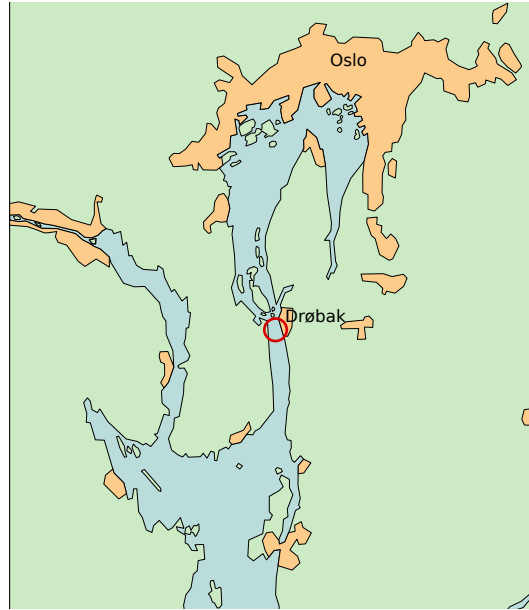


Figure 2.1: Map of the Oslofjord; the red circle marks the approximate area for the experiment and the ambient grab samples. Modified after Finn Bjorklid.

Table 2.1: Setup for the frames in the experiment. All the frames had the same configuration. (S-sand, C-drill cuttings, F-fine, 6mm and 24mm layer of drill cuttings). The frames marked with (*) are the types of treatments in this master thesis.

F	S(*)	FF	FF24
FS6	FS24	FC6	FC24
SF6	SF24	SS6	SS24
SC6(*)	SC24(*)	F	S(*)

in this master thesis from three of the four initial frames. In total there were initially 6 control boxes (either with sand or fine material), 3 boxes with a 6mm layer of treatment (either cuttings, clay or sand) and 3 boxes with a 24mm layer of treatment. All the boxes were placed at random in each frame. Aluminium bars screwed to the handles of each frame held the boxes in position in the frames.

After 6 months, on the 24th of September, each underwater buoy was recaptured (figure 2.3) and (figure 2.4). When the frames were brought up frame C came up in a tilted position so that the content did not stay in place and this frame had to be excluded from the experiment. Two ambient (Southern and Northern) grab samples taken from the adjacent seabed are also included in the material to enable faunal comparisons with the experimental material. Ambient samples are collected to give a picture of the fauna and of the

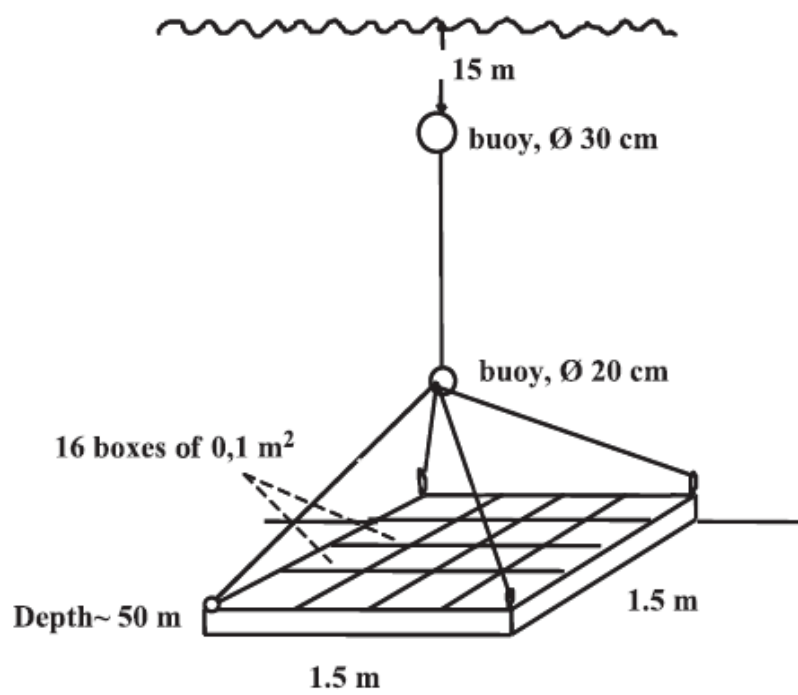


Figure 2.2: Schematic illustration of one of the experimental frames (Trannum et al, 2004)



Figure 2.3: Picture of the recapturing of one of the frames from the bottom at the experimental site. Each frame had a curtain with a mechanism that closed the curtain when the frame was pulled up from the bottom. The function of the curtain is to make sure that the material stays in place. Photo: Frode Olsgard with permission

potential of colonisers present in the area (Olsgard, 1999) The sediments from the boxes and the grab samples were for practical reasons washed through 1 and 0.5 mm sieves with round holes for macrofaunal analysis. The residues from the 1 and 0.5mm sieves were later pooled and treated together in the analysis of the results. The sieve residues were fixed in 4% buffered formaldehyde and stained with Rose Bengal according to Eleftheriou and Moore (2005).

2.2 Species identification

In the laboratory the samples were washed on a 0.5mm sieve to remove the formaldehyde and excess sediment. The fauna was sorted in 5 groups: Annelida, Crustacea, Mollusca, Echinodermata and “Varia”. The animals were identified to the lowest taxonomic level possible and preserved with 75% ethanol. Faunal abundances were enumerated for each sample. 1 of the 12 samples from the experiment seemed not to be fixed with formaldehyde because no animals were found.

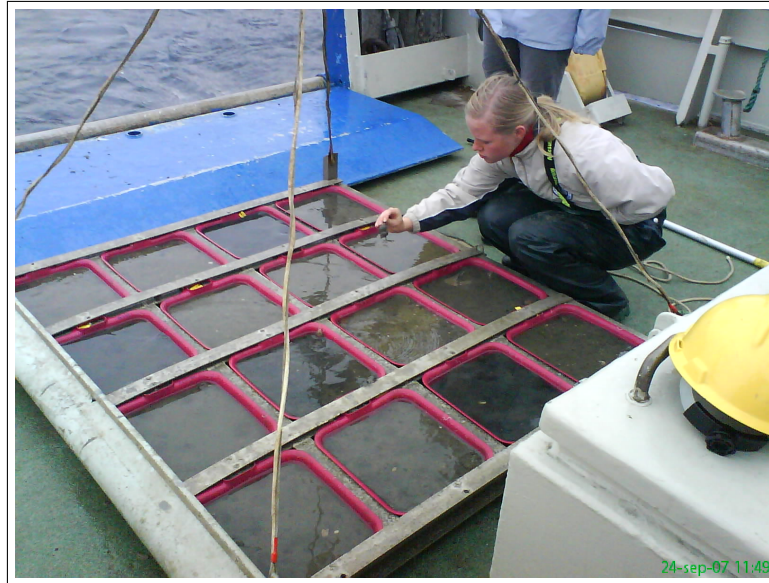


Figure 2.4: Picture of one of the frames on deck after recapturement. Student in deep concentration labeling the boxes. Photo: Frode Olsgard with permission

2.3 Grain size analysis and oxygen penetration

Grain size was measured for the coarse and fine sediment and for the drill cuttings before the frames were deployed. The sediment was sampled with a spoon and put in a plastic bag for further analysis. This is usually done with a corer, but since the sediment was mixed this was not necessary. The samples were split into two fractions, 1mm (silt) and 0.063 (clay) and separated by wet sieving. The sediment was then dried at 60°C until dry. The dried sample of the fraction >0.063 was weighed and shaken for 10 minutes through a nest of graded sieves (2, 1, 0.5, 0.25, 0.125 and 0.063 mm. For 2.2 g dryweight of the material of the <0.063 mm fraction, 50 ml distilled water was added and 1 drop (0.06 g) dispersant (Calgon corresponding to 1-3% of the samples dry weight) was added. The mixture was then treated in ultrasonic bath for 10 minutes. The solution was finally analysed in a Sedigraph 5000 at 33.3 °C.

The oxygen penetration depth was measured after retrieval of the frames with a Clark-type oxygen electrode. The oxygen penetration depth defines the thickness of the oxic zone in sediments. Example of the range of penetration depth is from mm to less than 10 cm on the continental margin (Wei-Jun and Sayles, 1996). The oxygen penetration depth is here defined as the sediment depth having $> 5\%$ oxygen saturation, and was calculated from the measured profiles (Trannum et al, 2009).

2.4 Data analysis

The faunal observations were analysed by univariate and multivariate techniques. For each sample (ambient grab samples and experimental samples), univariate measures included total number of individuals (N) (abundance) and total number of taxa (S) (richness).

Several diversity indices were calculated. It is common to use several measures of diversity in the same investigation. The different ways of calculating diversity interpret the fauna composition in different ways (Olsgard, 1995).

Shannon's diversity index (exp H') is given by

$$H' = - \sum_i p_i \log(p_i)$$

where p_i is the proportion of the total count (or biomass etc) arising from the i th species (Clarke and Warwick, 2001), (Shannon and Weaver, 1963). The Shannon's index is sensitive for rare species (Olsgard, 1995).

Simpson's diversity (1-Lambda) (Simpson, 1949) has a number of forms

$$\begin{aligned} \lambda &= \sum p_i^2 \\ 1 - \lambda &= 1 - (\sum p_i^2) \\ \lambda' &= \sum_i \frac{N_i(N_i - 1)}{N(N - 1)} \\ 1 - \lambda' &= 1 - \sum_i \frac{N_i(N_i - 1)}{N(N - 1)} \end{aligned}$$

where N_i is the number of individuals of species i and λ is the probability that any two individuals from the sample, chosen at random, are from the same species (λ is always < 1) (Clarke and Warwick, 2001). Simpson's diversity is a *dominance* index, in the sense that its largest values correspond to assemblages whose total abundance is dominated by one, or a very few of the species present (Olsgard, 1995).

Pielou's evenness index (J') (Pielou, 1966) is given by

$$J' = H' / H'_{\max} = H' / \log S$$

where S is the number of species and H'_{max} is the maximum possible value of Shannon diversity, i.e. that which would be achieved if all species were equally abundant (namely, $\log S$). (Clarke and Warwick, 2001). Pielou's evenness index measures how even the individuals are distributed between the species (Olsgard, 1995).

Hurlbert's rarefaction (Sanders, 1968; Hurlbert, 1971) is given by

$$ES_n = \sum_{i=1}^S \frac{[1 - (N - N_i)!(N - n)!]}{(N - N_i - n)!N!}.$$

The method can be used to project back from the counts of total species (S) and individuals (N), how many species (ES_n) would have been 'expected' if we had observed a smaller number (n) of individuals (Clarke and Warwick, 2001). Hurlbert's rarefaction is a graphical method for describing diversity. According to Klif (earlier SFT) guide for classification of environmental state the community (here: the box) is considered unaffected, in equilibrium and the state is classified as "good" when the ES_{100} -value is over 18.5, while lower values can indicate influence from pollution or some kind of disturbance (Olsgard, 1995). Minitab version 15 was used to make the box plots of the diversity indices.

Regression analysis were carried out for abundance of the ten most abundant species from the experimental boxes, feeding groups, total abundance, total number of species and diversity indices against the layer thickness of drill cuttings with the statistical program R. The faunal counts were log-transformed in order to attain equal spread.

Because it was desirable to see if the drill cuttings would affect the function of ecological groups of animals, the fauna was divided into the following feeding groups; (1) suspension/filter feeders, (2) surface deposit feeders, (3) subsurface deposit feeders, (4) carnivore/omnivore, (5) scavengers, (6) scrapers/grazers, (7) dissolved matter/symbionts, (8) parasites/commensals, (9) large detritus/sandlickers (see appendix for table with list of feeding mode of each species, table A.5 and table A.3 for the compiled list). The species got a score for the different feeding modes; 0, 1, 2, or 3 depending on how much the species is one or another of the categories. If a species fit equally well into two groups it was assigned to both groups. The traits for the feeding groups were acquired from the NIVA-database.

Multivariate analysis were carried out with nonparametric methods in the PRIMER-package (Plymouth Routines In Multivariate Ecological Research) (Clarke, 1993; Clarke and Warwick, 2001). To analyse for similarities in community structure Multi Dimen-

sional Scaling (MDS) based on Bray-Curtis similarity measure was executed given by

$$S_{jk} = \frac{\sum_{i=1}^S |X_{ij} - X_{ik}|}{\sum_{i=1}^S (X_{ij} + X_{ik})}$$

where X_{ij} and X_{ik} are the numbers of individuals of the species i at station j (Olgard, 1995). Similarities were calculated based on the fourth root counts. The purpose of the MDS is to construct a “map” or configuration of the samples, in a specified number of dimensions (Clarke and Warwick, 2001). Cluster analysis was carried out and aim to find “natural groupings” of samples such that samples within a group are more similar to each other, than samples in different groups. It is possible to test for significance in the MDS-ordination, the ANOSIM procedure in the PRIMER-package. Since there were only three replicates this was not considered useful.

Chapter 3

Results

The raw data (environmental variable measurements and species abundances) are provided in the appendix A.

3.1 Grain size analysis and oxygen penetration

The median grain size for sand, clay and drill cuttings was $65.0\mu\text{m}$, $8.8\mu\text{m}$ and $10.9\mu\text{m}$ respectively. The sediments had a pelite-fraction ($<63\mu\text{m}$) of 39, 57 and 74% for coarse (fine sand), fine (silt) and the drill cuttings (silt) respectively. Although there was a clear tendency of lower oxygen the more drilling cuttings were added, there was no statistical difference in oxygen penetration depth between the treatments, measured at the end of the experiment (table 3.1).

Table 3.1: Oxygen penetration depth in cm, sediment depth having $> 5\%$ oxygen saturation. A, B and D are the three frames, S = sand, C = cuttings and the 6 and 24 are the layer thickness of drill cuttings.

	A	B	D	Mean
S	3.7	2.2	3.5	3.1
SC6mm	3.2	3.4	1.4	2.7
SC24mm	2.0	-	2.8	2.4

3.2 Univariate analysis

A total number of 3574 animals belonging to a total of 130 species were counted in the 11 boxes and the two ambient grab samples. Around 2/3 of the taxa (84 out of 130) were identified to the species level. Annelida (Polychaeta) was by far the dominant group, comprising 88% of the individuals and 51 % of the taxa. Crustaceans, molluscs and echinoderms made up the remainder of the samples, in addition the group “Varia” which included the phyla Cnidaria, Echiura, Sipunculida, Nemertinea and Nematoda (table 3.2).

There was a slight increase in the average number of individuals in each of the treatments. There were on average 345 individuals in the controls, while the numbers for 24mm mud and 6mm mud was 280 and 246, respectively (table 3.3). Maximum and minimum number of individuals for each treatment are also found in table 3.3. The 6mm treatment had 19% less animals than the controls and the 24mm treatment had 29% less animals than the controls on average and the 24mm treatments had 12% less animals than the controls. The average number of taxa in the three treatments was about equal (table 3.3), but the average number of species in the ambient samples was lower.

For many of the species and feeding groups there is a negative trend as a function of the layer thickness, but this trend is not statistically significant (figure 3.2 and 3.3). However, there are exceptions. *Ophelina acuminata* ($p = 0.009$) (figure 3.2) shows a significant decline for the number of individuals as a function of the layer thickness of added drilling cuttings. *Echiurus echiurus* shows a positive trend ($p = 0.09$) for number of individuals as the layer thickness of drill cuttings increased that is close to significant (figure 3.2). Linear regression on the feeding groups (carnivore/omnivore, suspension feeders, subsurface deposit feeder, surface deposit feeders and suspension/subsurface deposit feeders) does not show significant p-values (figure 3.3).

Table 3.2: The total number of individuals and taxa within each phylum in all the samples (in the ambient samples and the experimental boxes), and the percentage of individuals and taxa that each phylum made up of the total abundance and species richness. Both abundance and number of taxa are absolute values.

	Annelida	Crustacea	Mollusca	Echinodermata	Varia
Total no. of individuals	3160	75	98	39	204
Total no. of taxa	66	31	19	6	8
% of the individuals	88.37	2.1	2.74	1.09	5.7
% of the taxa	50.8	23.7	14.6	4.6	6.2

Table 3.3: Average abundance and average number of taxa per box in the different treatments. Averages are used to enable comparisons between the treatments.

Abundance (N)	Controls	6mm	24mm	Ambient
Max	570	453	276	149
Average	345	280	246	135.5
Min	248	186	193	122
Taxa (S)	Controls	6mm	24mm	Ambient
Max	35	44	45	25
Average	33.6	33.7	36.3	24.5
Min	30	26	29	24

3.2.1 Faunal diversity

Shannon's diversity index ranged from 1.7 to 2.7, Pielou's evenness from 0.51 to 0.82, Simpson's from 0.69 to 0.89 and Hurlbert's rarefaction ranged from 15.22 to 24.7 (table A.1 in appendix A and figure 3.1). Only two of the boxes had values below 18.5 for Hurlbert's rarefaction and both of these were surprisingly from the controls. Pielou's evenness index show a significant difference between the groups; there is a higher evenness in the established community at the experimental site, but also surprisingly higher (though not significant) evenness in the 24mm treatment compared to the 6mm treatment. There is a wider range for the numbers for the diversity indices in the controls compared with the ambient samples and the treatments. There are no significant results in the regression analysis on the diversity indices (figure 3.4). Hurlbert's rarefaction show a weak positive trend as a function of increasing thickness layer of drill cuttings.

3.2.2 Faunal Composition

The ten most abundant taxa for each of the treatments and the ambient samples are listed in table 3.4. Most of the species are polychaetes with except for a few, one Echiuran (*Echiurus echiurus*), some Ophiurids and Calanoids. The most abundant taxon in the experiment was the polychaet *Polydora caulleryi*, which comprised from 25% to almost 30% of the abundance in the treatments, followed by *Pseudopolydora pausibranchiata*, *Heteromastus filiformis* and *Prionospio steenstrupi*. The ambient samples has a clearly different composition with several species not found in the experiment (table 3.4). The ten most abundant taxa in each of the samples are listed in figure A.2, in appendix A. There is a tendency for more dominance in the controls and the ambient samples, where the ten

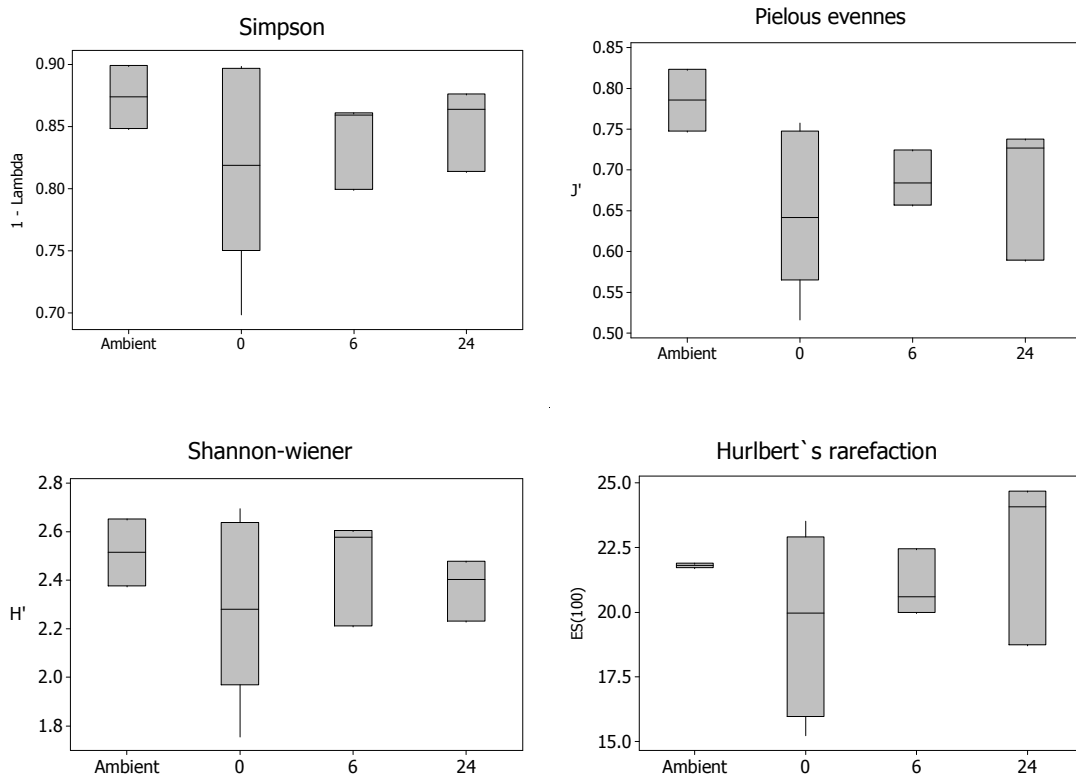


Figure 3.1: Simpsons diversity ($1-\text{Lambda}$), Pielou's evenness index (J'), Shannons diversity exp (H') and Hurlbert's rarefaction (ES_{100}) for the ambient samples and the experimental boxes. Box-plots with median, range, 1st(Q1) and 3rd(Q3) quartiles indicated.

Table 3.4: Ten most abundant species in each of the three treatments; controls (S), 6mm cuttings on sand (SC6) and 24 mm cuttings on sand (SC24), as well as the ambient grab samples.

(a) Ambient			(b) Sand (S)		
Species	N	% of total abundance	Species	N	% of total abundance
<i>Prionospio steenstrupi</i>	39.5	29.2%	<i>Polydora caulleryi</i>	102	29.6%
<i>Pseudopolydora paucibranchiata</i>	16	11.8%	<i>Pseudopolydora pausibranchiata</i>	69.6	20.2%
<i>Prionospio fallax</i>	11	8.1%	<i>Prionospio steenstrupi</i>	44.2	12.8%
<i>Myriochele oculata</i>	9.5	7%	<i>Heteromastus filiformis</i>	23.4	6.8%
<i>Nematoda indet</i>	9	6.6%	<i>Prionospio fallax</i>	14.2	4.1%
<i>Spiophanes kroeyeri</i>	8	5.9%	<i>Ophelina acuminata</i>	19	5.5%
<i>Calanoida indet</i>	6.5	4.8%	<i>Echiurus echiurus</i>	12.6	3.7%
<i>Prionospio cirrifera</i>	6.5	4.8%	<i>Eteone longa/flava</i>	5.2	1.5%
<i>Chaetozone setosa</i>	5.5	4.1%	<i>Spiophanes kroeyeri</i>	4.2	1.2%
<i>Levinsenia gracilis</i>	4	3%	<i>Antinoella sarsi</i>	4.2	1.2%
Total	115.5	85.2%	Total	298.67	86.55%

(c) 6mm added drill cuttings (SC6)			(d) 24mm added cuttings (SC24)		
Species	N	% of total abundance	Species	N	% of total abundance
<i>Polydora caulleryi</i>	74.3	26.5%	<i>Polydora caulleryi</i>	62	25.2%
<i>Pseudopolydora pausibranchiata</i>	65.7	23.5%	<i>Pseudopolydora pausibranchiata</i>	48.3	19.7%
<i>Prionospio steenstrupi</i>	24.7	8.8%	<i>Heteromastus filiformis</i>	24.7	10%
<i>Heteromastus filiformis</i>	23	8.2%	<i>Prionospio steenstrupi</i>	24.3	9.9%
<i>Echiurus echiurus</i>	17	6.1%	<i>Echiurus echiurus</i>	19.3	7.9%
<i>Ophelina acuminata</i>	8.7	3.1%	<i>Prionospio fallax</i>	6	2.4%
<i>Gattyana cirrosa</i>	5.5	2%	<i>Ophiuridea indet</i>	4.3	1.8%
<i>Ophiuroidea indet</i>	4.7	1.7%	<i>Gattyana cirrosa</i>	4	1.6%
<i>Eteone longa/flava</i>	4.3	1.6%	<i>Nephtys pente</i>	3.3	1.4%
<i>Nephtys pente</i>	4.3	1.6%	<i>Eteone longa/flava /Antinoella sarsi</i>	3	1.2%
Total	232.2	82.9%	Total	199.3	82%

most abundant species make up 85 and 86 percent of total abundance respectively, while in the two treatments the numbers are 82 and almost 83, respectively. There is no evidence that the ten species with highest abundance contributes more to the total abundance in any of the treatments. The different boxes has a similar composition in the ten most abundant species, and in all but two treatments the top two species are the same. The number of unique species to each of the ambient grab samples (11), controls (12) , 6mm mud (12) and 24 mm mud (11) is about the same. 9% of the total number of species were only found in the ambient samples.

3.3 Multivariate analysis

The MDS ordination did not show any clear clustering of the samples as a function of treatment. The stress value is 0.13. It is generally accepted that a value below 0.2 is required for a reasonable representation of the overall faunal pattern in the MDS ordination (Clarke, 1993). The MDS ordination with the macrofaunal counts from the ambient grab

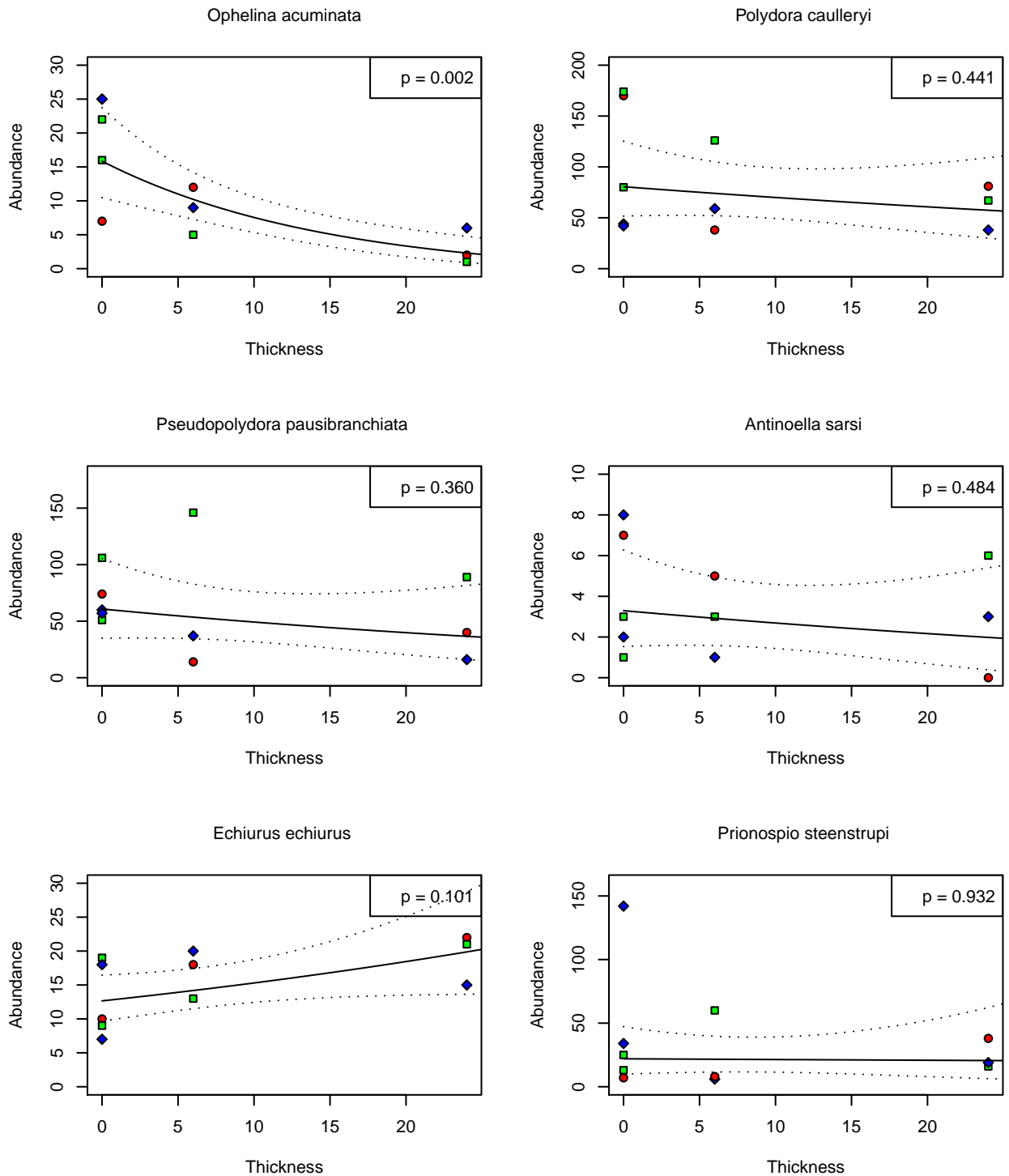


Figure 3.2: Linear regression on log transformed faunal counts for the ten most abundant species. Frame A = red circles, frame B = green squares and frame D = blue diamonds. Pointwise 95% confidence bands for the regression lines are marked with dotted lines.

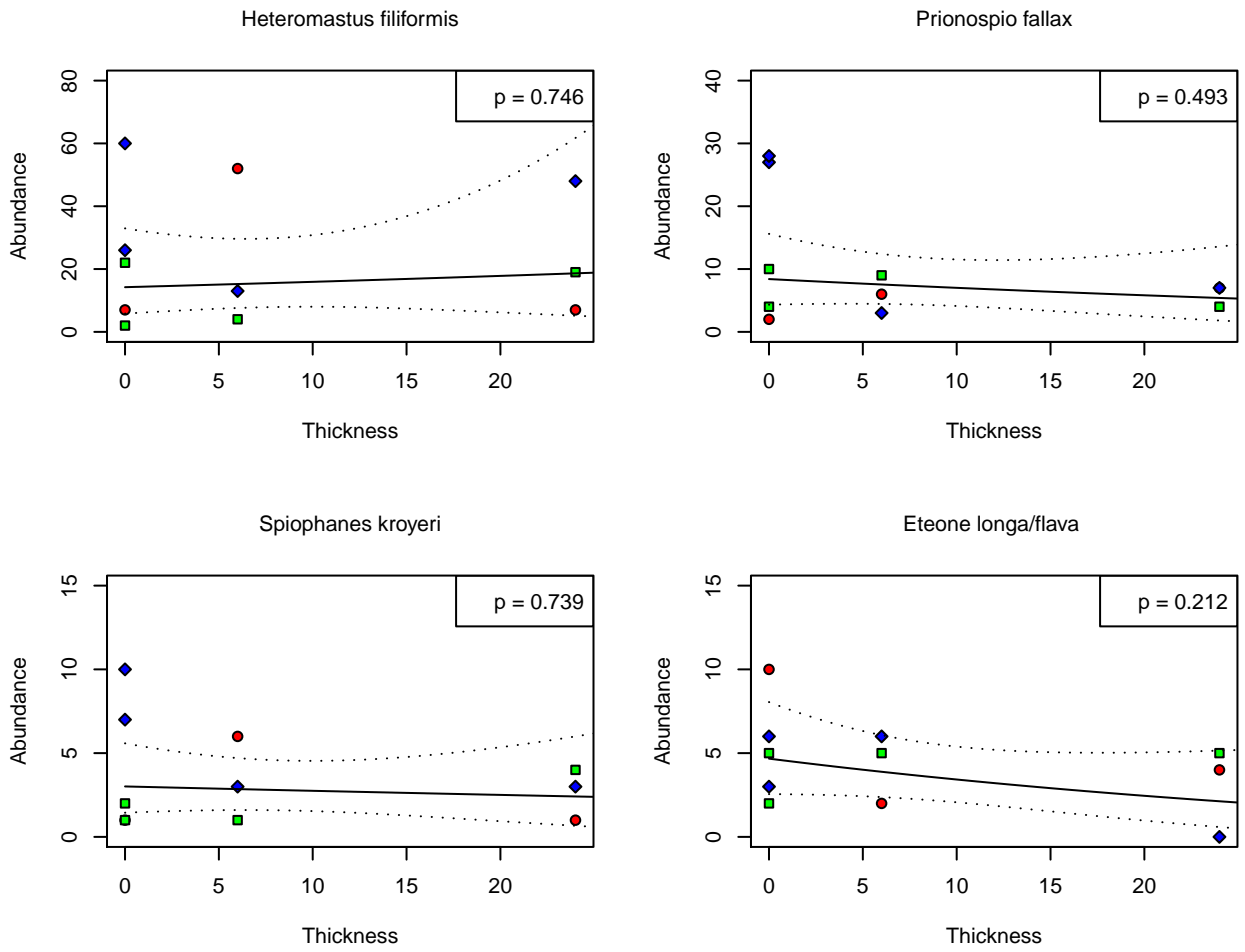


Figure 3.2: (continued)

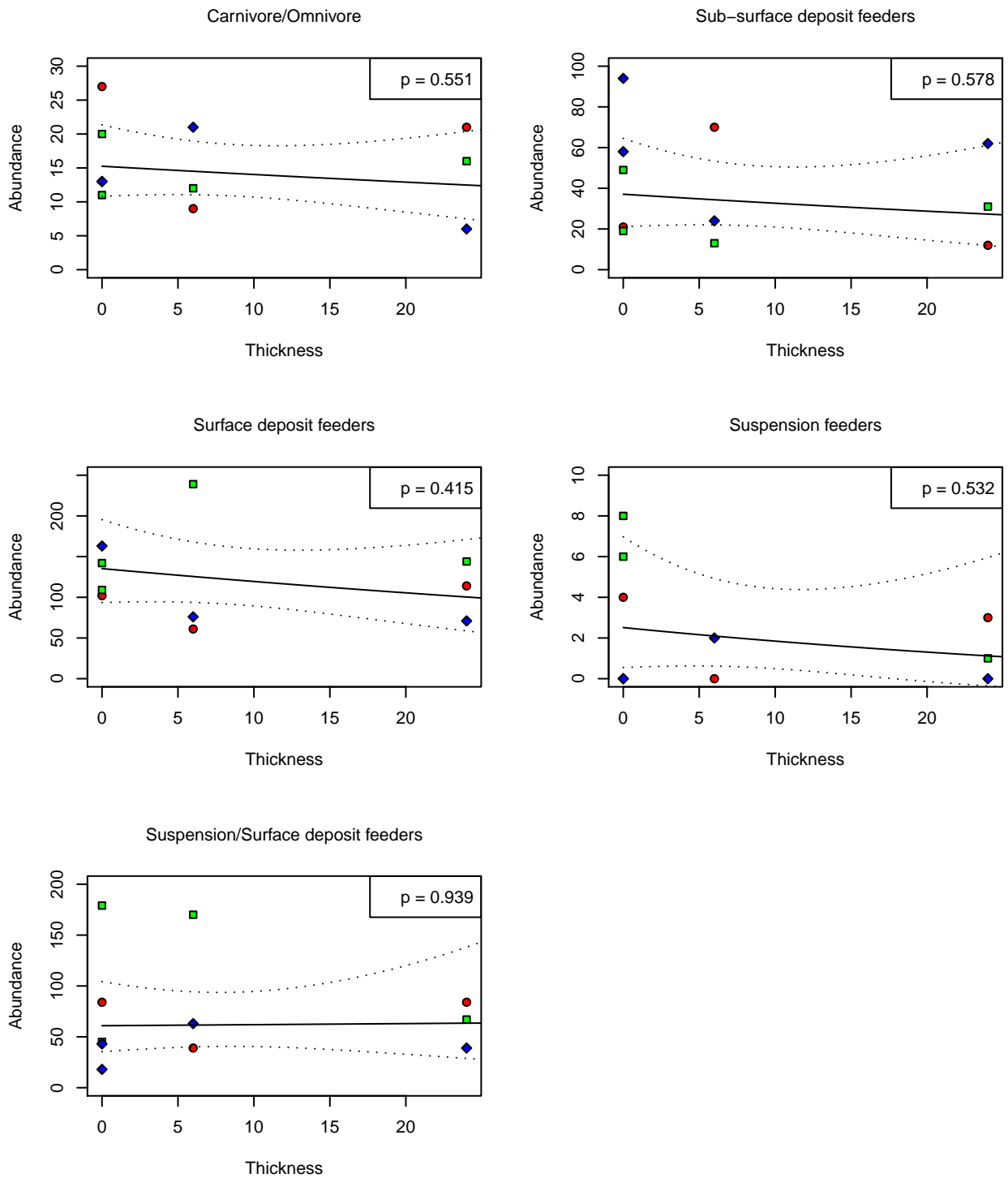


Figure 3.3: Linear regression on log transformed faunal counts on feeding groups. The frames are indicated in the same way as in figure 3.2. Pointwise 95% confidence bands for the regression lines are marked with dotted lines.

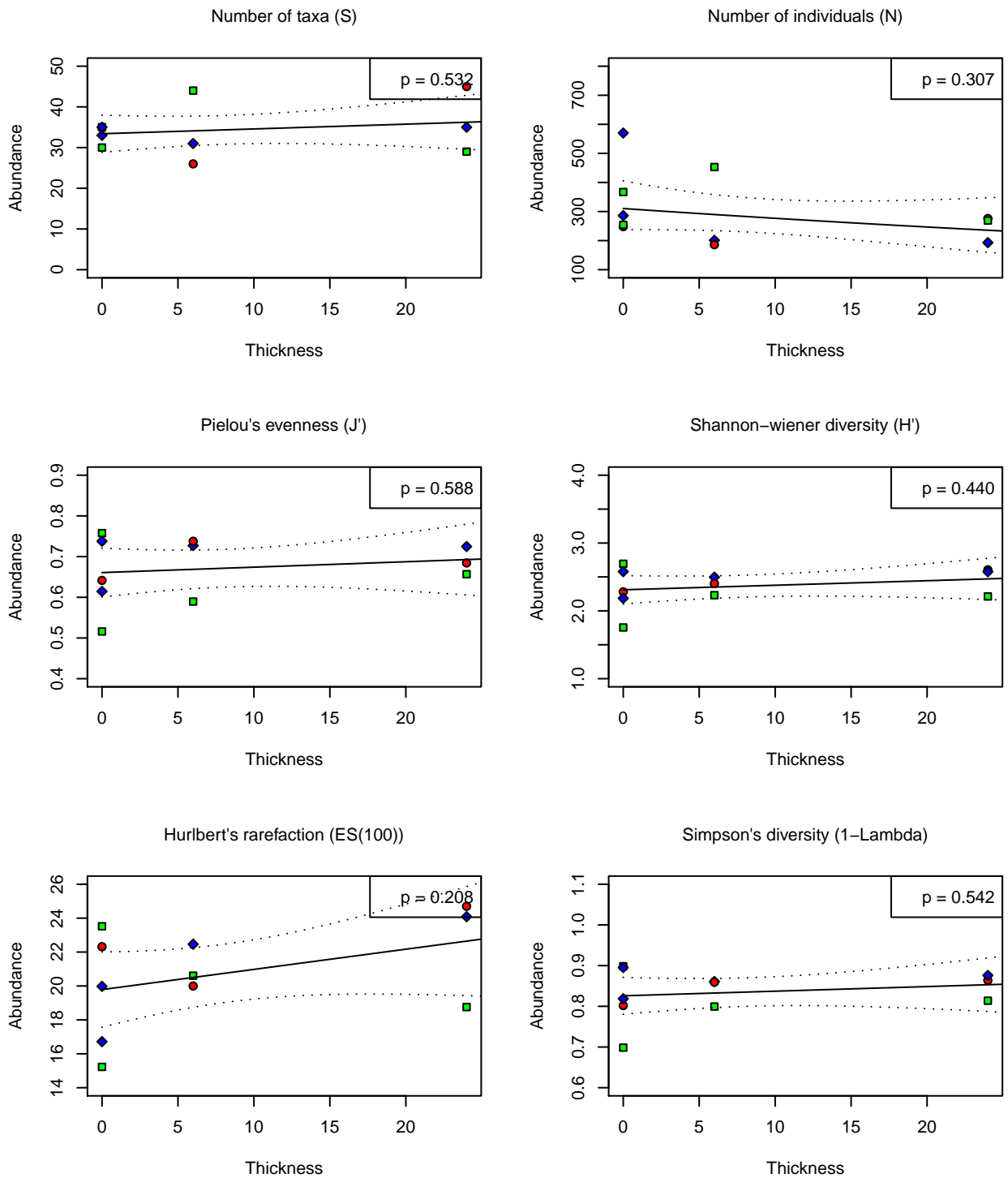


Figure 3.4: Linear regression for number of taxa (S), number of individuals (N)(log-transformed) and on the diversity indices. The frames are indicated in the same way as in figure 3.2. Pointwise 95% confidence bands for the regression lines are marked with dotted lines.

samples combined with the counts from the experiment show two distinct groups, one group consisting of the two grab samples and one group consisting of the experimental boxes (figure B.2, in appendix B). This shows a clear difference in the two subsets of samples. MDS ordination as a function of frame show that the boxes in each of the frames are grouped more closely than the boxes for each treatment (figure 3.6). MDS ordination as a function of treatment show no grouping, the boxes are randomly distributed in the plot (figure 3.5). The outlines show that none of the boxes from the different treatments are grouped more closely than the others.

MDS ordination as a function of frame without the two most abundant species, *Polydora caulleryi* and *Pseudopolydora pausibranchiata* was executed (figure B.3, appendix B). Without the two most abundant species the effect of frame changes, but is still present. In this plot frame A is more scattered, but frame B and D have about the same clustering. The same ordination was done as a function of treatment, which resulted in a different community structure, but no grouping of the frames according to treatment (figure B.1, in appendix B). To investigate the possibility that the two most abundant species were responsible for the frame effect another MDS was executed with only the two most abundant species, *Polydora caulleryi* and *Pseudopolydora pausibranchiata* (figure B.4, in appendix B). This plot shows no grouping of the samples neither according to frame nor according to treatment.

Cluster analysis based on Bray-Curtis similarities show the same groupings as the MDS as a function of frame (figure 3.7); the B and D frames are two almost separate groups with one box from frame D more closely related to frame B.

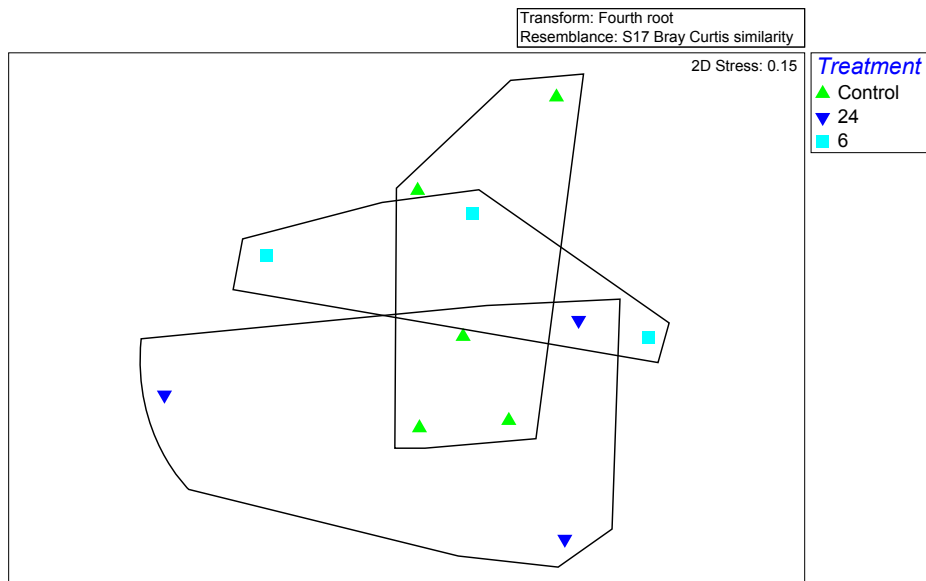


Figure 3.5: Multi dimensional scaling (MDS) based on fourth root transformed counts from species abundances of the 11 experimental boxes as a function of thickness of drill cuttings. The treatments are outlined for illustration.

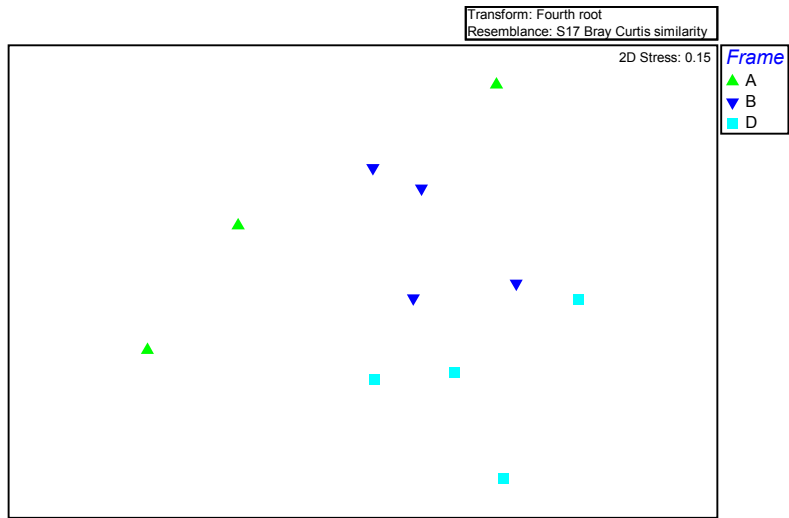


Figure 3.6: Multi dimensional scaling based on fourth root transformed faunal counts from species abundances of the 11 experiemental boxes as a function of frame.

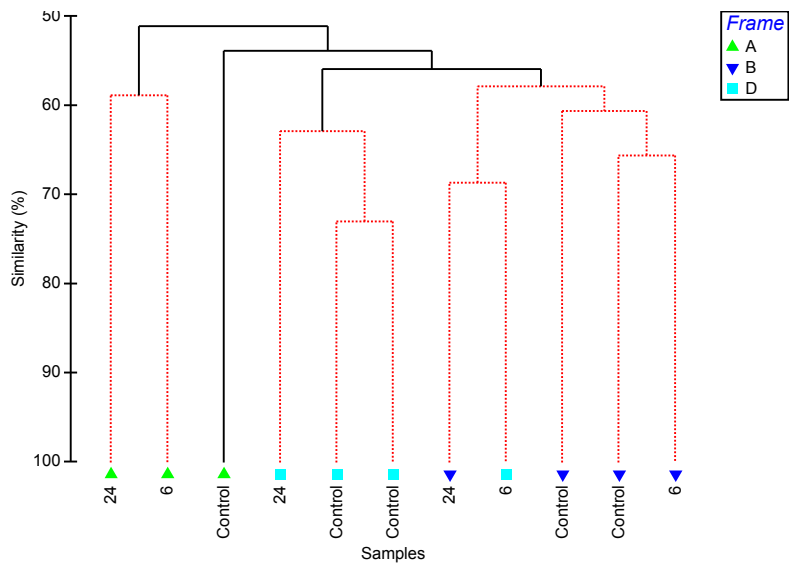


Figure 3.7: Hierarchical agglomerative clustering with group-average linking of the 11 boxes in the experiment, with Bray-Curtis similarities on fourth root transformed counts for species abundances. Red split = not statistically different species composition.

Chapter 4

Discussion

4.1 Effects of drill cuttings on benthic communities

Multivariate analysis of the faunal counts show no significant effects of the drill cuttings as a function of thickness layer of the cuttings added. The MDS analysis shows no grouping as a function of the treatment (figure 3.5). The regression analysis shows a weak tendency to lower abundance as a function of the thickness layer of the drill cuttings.

This study provides no evidence that settling communities are sensitive to WBM cuttings. This is in contrast to a related mesocosm experiment (Trannum et al, 2009)(also in PEIOFF-FAME) who found effects of WBM drill cuttings. These results indicates that living communities “buried” by WBM drill cuttings are more sensitive to this kind of contamination. The two types of experiments are widely different; in a recolonization experiment the sediment is defaunated, whereas in the mesocosm experiment the purpose is to imitate already established communities that are “buried” in drill cuttings. It is not unlikely that natural variation might cover negative effects of the drill cuttings in the field, since there was an effect in the mesocosm experiment, negative effects in the other part of the experiment (fine sediment) and a negative tendency in my part of the experiment.

My results are in compliance with findings of Daan and Mulder (1993). They found no adverse effects of WBM-cuttings one year after dumping of WBM cuttings, even as close as 25 m from the former discharge site. Recent field and laboratory studies tend to confirm these results. Benthic fauna are not harmed by drill cuttings from WBM, since the exposure from drill cuttings from WBM in a oil field is of short duration and the cuttings are rapidly diluted. Impacts of WBM are limited to 100 m within the platform and recovery is well within one year. However, effects are more severe if released to; coastal areas, deep-water environments or low-energy habitats (Neff, 2005). In many of

the cases where effects of WBM have been observed one has not been able to attribute these effects to WBM with certainty, because of previous drilling with OBM in the same area (Daan and Mulder, 1993).

Bakke et al (1986) performed an experiment with four types of drill cuttings; WBM, LAC (low aromatic oil based), DOC (diesel oil based mud, washed with diesel oil) and BRI-cuttings (cuttings from drilling with diesel oil based mud compressed into solid briquettes). As expected there were large differences in response between the oil and non-oil cuttings, with significant effects only in the sediment treated with oil based cuttings. There was a slightly stronger negative trend in this experiment compared to my experiment (Bakke et al, 1986). With the restrictions for OBM that came in 1993 there was a need to improve the technical properties of WBM, since OBM was preferred due to its superior properties. It is reasonable to assume that the composition of WBM has changed because of this need. The stronger trend in Bakke et al (1986)'s experiment can possibly be explained by the fact that the composition of WBM might have changed (T Bakke 2010, pers. com). My experiment differed in having higher number of species per box at the same stage (after 6 months). There were on average 34 species per box in my experiment as opposed to about 22 per box in Bakke et al (1986). One possible explanation for this difference is that there were fewer species identified to the species level (T Bakke 2010, pers. com.).

Berge (1990) also did an experiment in the Oslofjord, but this was with sediments treated with crude oil. He did the experiment in two fjords with unequal eutrophication status, Raunefjord in western Norway in addition to the Oslofjord. The eutrophicated Oslofjord was little affected by the oil, while there was a clear reduction of species in Raunefjord. The possible explanation given for this result is that communities with few species, low diversity, high dominance and high production rates are more stress tolerant (they have a higher resilience) than more complex systems (Jernløv and Rosenberg, 1976). From these criteria the fauna found in my experiment does not have the characteristics for a resilient community, although the Oslofjord is described as one by the author above. The fauna in the ambient samples has many species, high diversity and low dominance. No measurements for production were made in my experiment, hence there is no value for this factor. The difference between the fauna in Berge (1990) and my experiment can be explained by the change in eutrophication status in the Oslofjord since 1990 (Magnusson et al, 1997). It is difficult to predict how resilient the Oslofjord is today. However, there has been an overall improvement of the pollution situation in the Oslofjord since 1990. The fauna in the outer Oslofjord was surveyed by Niva in 2008, and the report shows a positive trend for the fauna in the outer Oslofjord, although Shannon-Wiener diversity

index (H') and Hurlbert's rarefaction ($ES(n)$) has not changed systematically. One station in the survey is located right before the Oslofjord widens (Walday et al, 2009). This station is located further out in the fjord from the experimental site. However, assuming sometimes inward directed currents the, community in my experiment may have received some of its recruits from the outer part of the Drøbak strait. 70% of the species present in the recruiting assemblages were not found in the adjacent seabed, indicating significant nonlocal recruitment.

Another factor is that the experimental site in Berge (1990)'s experiment was in the inner Oslofjord (Berge et al, 1987), which is known to be more polluted than the outer part. The lacking effect from exposure of oil in the experiment in 1990 may not be comparable with the experiment with WBM cuttings from 2007 in the same fjord, because of the improved environmental conditions and the following positive trend for the fauna. However, the improved conditions might have made the fauna in the Oslofjord less robust today present, so that it would be more likely to observe an effect of the drill cuttings.

4.1.1 Effects on faunal diversity

The diversity indices show that there is no effect of the drill cuttings on the diversity (figure 3.1 and 3.4). The only index with significant differences are the Pielou's evenness index, where the ambient grab samples have a higher evenness. This is expected since the ambient samples are from mature communities. It is more surprising that the other diversity indices does not differ more, since a mature community is expected to have a higher diversity (Margalef, 1963). The values for Hurlbert's rarefaction are slightly higher in the samples with 24 mm added drill cuttings, which indicates a positive relationship between the added cuttings and the number of species, but this is not statistically significant. This result is unexpected and difficult to explain. Such high values are associated with a healthy fauna (Molvær et al, 1997), which is an indication that the community is not polluted and not affected by the drill cuttings added.

4.1.2 Effects on faunal composition

The composition of the most abundant species (figure 3.4) supports that the drill cuttings have no significant effect on the settling of the benthic community in my experiment, since the composition of the ten most abundant species are similar in the treatments and the control. The species *Polydora caulleryi* and *Pseudopolydora pausibranchiata* were the two most abundant species in all except for two boxes. The rest of the species varied more.

Ophelina acuminata was the only species with significantly lower abundance as a function of cuttings added. According to literature this species is sensitive to pollution (Rygg, 1985) and it is not surprising that we see a negative effect of the drill cuttings on this species.

Polychaeta was the most abundant group in the experiment. This is in compliance with other studies of recruiting soft-sediment assemblages (Berge, 1990; Olgard, 1999; Trandum et al, 2004). They found that polychaetes was the most abundant group, independent of depth and habitat and treatment.

Polydora Caulleryi was the most dominating species in many of the boxes in my experiment, independent of treatment. This species is known to often be one of the first colonizers in a succession. *Polydora* has a flexible life history strategy and a short life cycle which makes it a suitable colonist. (Gray and Elliott, 2009).

As mentioned in section 1.3, the larvae of opportunistic species have little preferences for substratum for settlement. Members of the family Spionidae, often the genus *Polydora* are the first to settle. Many of the most abundant species in this experiment are known to be opportunistic, which could partly explain the lack of difference between the test boxes and the control boxes. If the experiment had lasted longer, it is possible that larvae of more K-selected species would react differently to the exposure of drill cuttings. If “conditions” fail to improve (in the case where drill cuttings could affect K-selected species), the r-selected species may not be replaced by K-selected species (Gray and Elliott, 2009). It is possible that a negative effect of the drill cuttings would appear at a later stage.

4.2 Effects of frame location

The MDS ordination shows a weak but clear grouping as a function of frame (figure 3.6). Some of the plots in the regression analysis stand out because the controls have a larger range in the observations (figure 3.2, 3.3 and 3.4). This applies to the following plots: *Prionospio steenstrupi*, *Prionospio fallax*, *Spiophanes kroyeri*, *Eteone longa/flava*, sub-surface deposit feeders, suspension feeders, total abundance (N) and Pielou’s evenness index. It looks like there is a pattern in the observations, because there is a preponderance of observations with high abundance by blue diamonds (= frame D). Frame D was the frame that was placed on the opposite side of the fjord, away from the frame A and B. A possible explanation for the preponderance may be a greater supply of larvae on the side of the fjord where frame D was placed. One possible explanation is that the supply of larvae for frame D could have come more from the outer Oslofjord than for frame A and B. Hydrodynamics and natural heterogeneity can partly be responsible for this difference

(Bourget and Harvey, 1998; Morrisey et al, 1992).

When removing the two most abundant species (both overall and most of the boxes), *Polydora caulleryi* and *Pseudopolydora pausibranchiata* from the MDS-ordination, the effect of frame became much weaker (figure B.1, in appendix B). To test if these two species alone were responsible for the effect of frame, another MDS-plot with only these two species was made (figure B.4, in appendix B). This MDS did not show an effect of frame, hence these two species cannot be accounted for the frame effect in the original MDS-plot.

4.3 Environmental variables as explanatory factors

4.3.1 Total organic carbon (TOC)

Since total organic carbon for the drill cuttings and the test sediment were not measured in this experiment, values from (Trannum et al, 2009) and (Olsgard, 1995) are used for comparison. The same type of drill cuttings were used in my experiment as in Trannum's mesocosm experiment and the samples for the test sediment was taken in the same area as one of the stations in Olsgard's survey from 1993. The TOC measured in the drill cuttings was 0.8 % (Trannum et al, 2009) and the TOC value from the corresponding station in (Olsgard, 1995) was 1.9 %. The measured values for TOC are not particularly different and both of the values are well within the criteria for a healthy bottom fauna determined by Klif (earlier SFT) (Molvær et al, 1997).

4.3.2 Grain size

The sandy sediment and the drill cuttings differ in grain size, while the fine sediment and the drill cuttings are only slightly different in sediment grain size. If grain size is an important factor for settling and recolonization after disturbance from WBM drill cuttings, we should have seen an effect on the fauna in the experiment. Grain size is considered an important factor in structuring benthic communities (Grebmeier et al, 1989). The authors found that lower diversity correlated with an increase in fine sand fractions. However, my results does not correspond with these findings. Preliminary results from the fine sediments show an effect from the drill cuttings on the fauna (Trannum, unpublished results). Based on the grain size factor it is unexpected that the results show an effect in the fine sediment, while there was only seen a negative effect of the drill cuttings on one species in the coarse sediment (my part of the experiment).

The level of no observable effect, PNEC is a central part of the EIF (Environmental Impact Factor) which again is a part of ERMS (Environmental Risk Management System) that is developing for offshore drilling activities. Default values for PNEC are at present used to estimate effects of drill cuttings (Olsgard et al, 2005). All PNEC-values are at currently independent of the biota present in the sediment. A 21% change in median grain size is the PNEC-value determined by Leung et al (2005). The change in grain size in my experiment is more than the PNEC-value of 21% change (a reduction from 65m μ to 10.9 m μ). It is also more than the Hazardous level (HL50), which is 17.8 μ m determined by (Smit et al, 2008). On the basis of these numbers and the results from the whole experiment it is surprising that no effect of the drill cuttings was found, particularly when we know from literature that grain size is an important for the settlement of benthic larvae (Grebmeier et al, 1989).

4.3.3 Oxygen penetration

There is no significant reduction of the oxygen penetration depth as a function of increasing layer thickness of drill cuttings (table 3.1), although there is a decreasing trend. In mesocosm experiments with WBM cuttings, Trannum et al (2009) found that the oxygen penetration depth decreased as a function of layer thickness of the drill cuttings because of oxygen depletion. In their experiment, the test sediment boxes (2.1%) had a higher TOC content than the boxes with drill cuttings (0.8%). The author suggest that the organic carbon present in the cuttings was more degradable than the TOC in the test boxes. In addition, decay of dead fauna in the treatments with drill cuttings may have contributed to the decreased oxygen penetration depth (Trannum et al, 2009). It is possible that the oxygen level in the boxes in my experiment with drill cuttings was initially lower, but this situation might have gradually improved in the field because of the water currents circulating oxygen to the boxes (H Trannum 2010, pers. com.).

4.4 Conclusions

No clear relationship between the dose of WBM cuttings and the effect on settling of the benthic ecosystem on coarse sediments was found, but there was a clear negative trend. It is not impossible that natural variation covers an effect of the drill cuttings in the field. The environmental variables explained to some extent this result. Oxygen penetration depth supported this result, but grain size deviated with an unexpected result, i.e. more significant effects on fine sediment despite the fact that the fine sediments and the drill

cuttings have a more similar grain size than the cuttings and the coarse sediment. Other unexpected results that are difficult to explain are the higher variation in abundances in the control trays and the higher diversity (Hurlbert's rarefaction) in the treatments with drill cuttings. It is possible that the picture will change when all the results from the experiment is in place. It was not possible to include the rest of the results because of the limited time frame of the master thesis. For further research it would be interesting to do an experiment in a location that is known to have sensitive species. This would be highly relevant since it is not yet settled whether or not oil should be recovered from the vulnerable areas in Northern Norway.

4.5 Limitations

Fundamental to ecology is how populations, communities, and the processes that influence them vary with changes in spatial and temporal scale. This is important to have in mind, because field experiments can only feasibly be conducted at small spatial scales (Thrush et al, 1996). The time frame is also usually a limiting factor.

To what extent can we use small scale experiments to predict larger scale responses? Zajac et al (1998) suggests that this may not be possible, because of the mix of factors controlling successive processes at different spatial scales may be fundamentally different. However, definitive causal relationships between the presence of contaminants and their effects can only be shown by manipulative experiments (Underwood and Peterson, 1988). Manipulative field experiments are particularly well suited for studying recruitment of benthic communities. This is becoming an increasingly more common method for studying processes and variables that influence biological patterns of distribution in soft-sediments (Olsgard, 1999; Trannum et al, 2004; Olsgard et al, 2005).

Potential errors can be made in several stages in the process. The experimental design may potentially contain weaknesses; four frames were deployed on two different locations not far from each other, far enough to be true replicates, but close enough that an effect of frame/location should be avoided. A weak frame effect was present, but this does not necessarily mean that there is a weakness in the set-up of the experiment. The result may be an indication that even on such small scales, the communities have a different composition. This may be because of different recruitment potential (Morrisey et al, 1992). The dead fauna in the sediment collected for the experiment was not removed, and could potentially have influenced recolonisation. Another possible weakness of the experiment is that we cannot be sure that the thickness of the drill cuttings stayed the same throughout the experiment.

The frames were placed 20 cm above the seabed, but since most macrofauna is dispersed as pelagic larva, this will probably not represent a major obstacle for colonization through settlement. Previous studies on recolonization show that pelagic larval recruitment accounts for between 70% and 90 % of all individuals (McCall, 1977; Santos and Simon, 1980; Diaz-Castañeda et al, 1993).

One could possibly argue that the experiment should have lasted longer to observe effects on a more mature community. Bakke et al (1986) showed that benthic communities can be initially moderately affected by drill cuttings from OBM and then after two years come to a total collapse. The reference and the WBM followed one another through the experiment and as in my experiment there was no significant effect of the WBM drill cuttings. Since there was a longer time frame in this experiment it was more suitable to predict development in the community over time (Bakke et al, 1986). A scenario similar to that for OMB cuttings is probably not likely with WBM cuttings, even with a longer time frame since they are known to have less effect on benthic fauna (Daan and Mulder, 1993; Olsgard and Gray, 1995; Neff, 2005).

The identification process is also a stage where potential mistakes can be made. As I am an unexperienced taxonomist, there is a potential risk that I might make mistakes, especially in the beginning. Although not systematically, a significant amount of reidentification was carried through in cases where it was clear that specimens were misidentified. A lot of the identification work was also controlled by a supervisor. Possible errors made during identification can have serious consequences for the result and the reliability of the experiment. Some errors will affect the result, others will not. Mislabeling an entire species will not affect the result because statistics are independent on species names. It will only affect the result when looking for explanations for the result in the ecology of species or groups of species. Mislabeling some specimens of a species could affect the result if this applies to many animals. In my results, the chance that there are large errors is considered low, because of the consistency in the results from the whole experiment. The result from the ten most abundant species in all the boxes (figure A.2, in appendix A) can possibly indicate that there are no large errors in the identification because of the overall consistency. The Capitellides may be a possible source for errors in the identification. They are very small and it is difficult to see all the setae. Since there is a large predominance of *Heteromastus filiformis* and little deviation from the fine material it is most likely correct on the coarse material as well.

There were only taken two ambient grab samples. This is obviously too few replicates to do proper statistics, but the ambient samples are still included in the box plots for illustration of the difference between the adjacent seabed and the experimental community.

Both parametric and non-parametric statistics are used in the analysis of the fauna in the experiment. Non-parametric methods are based on fewer assumptions and it is not possible to make a direct comparison of the results. However, it is possible to discuss the validity of the assumptions for both the parametric and non-parametric methods. Hence; how well the parametric method fits for the purpose and the corresponding: how well the non-parametric method acts for the purpose. This will not be discussed here as it is beyond the scope of this thesis.

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Appendix A

Tables

Table A.1: Total number of taxa (S), total number of individuals (N), Shannon's diversity exp (H'), Simpson's diversity (1-Lambda') and Pielou's evenness index per for all the boxes and the ambient grab samples.

Sample	S	N	J'	H'(loge)	ES(100)	1-Lambda'
AMBTN	25	149	0.8237	2.651	21.733	0.8991
AMBTS	24	122	0.7478	2.377	21.902	0.8483
SA1	35	248	0.6415	2.281	22.316	0.8019
SB2	35	254	0.7577	2.694	23.52	0.8984
SB3	30	367	0.516	1.755	15.228	0.6985
SD4	35	570	0.6145	2.185	16.717	0.8186
SD5	33	286	0.7377	2.579	19.977	0.8954
SC24A1	45	276	0.6844	2.605	24.699	0.8639
SC24B2	29	269	0.6568	2.212	18.756	0.8138
SC24D3	35	193	0.7247	2.577	24.087	0.876
SC6A1	44	453	0.5895	2.231	20.606	0.7994
SC6B2	31	201	0.7271	2.497	22.461	0.8609
SC6D3	26	186	0.7377	2.403	19.999	0.8592

Table A.2: Tables of the ten most numerically abundant taxa in each of the experimental boxes in the experiment.

(a) B16S			(b) B2S		
Species	N	% of total abundance	Species	N	% of total abundance
<i>Pseudopolydora pausibranchiata</i>	51	20.1	<i>Polydora caulleryi</i>	170	46.3
<i>Polydora caulleryi</i>	44	17.3	<i>Pseudopolydora pausibranchiata</i>	106	28.9
<i>Prionospio steenstrupi</i>	25	9.8	<i>Ophelina acuminata</i>	16	4.4
<i>Heteromastus filiformis</i>	22	8.7	<i>Prionospio steenstrupi</i>	13	3.5
<i>Ophelina acuminata</i>	22	8.7	<i>Echiurus echiurus</i>	9	2.5
<i>Echiurus echiurus</i>	19	7.5	<i>Trochochaeta multisetosa</i>	9	2.5
<i>Prionospio fallax</i>	10	3.9	<i>Corbula gibba</i>	8	2.2
<i>Gattyana cirrosa</i>	7	2.8	<i>Eteone longa/flava</i>	5	1.4
<i>Nephtys pente</i>	5	2.0	<i>Prionospio fallax</i>	4	1.1
<i>Nephtys longosetosa</i> /			<i>Nephtys pente</i> /		
<i>Mediomastus fragilis</i> /			<i>Glycera alba</i> /		
<i>Gastropoda</i> indet	4	1.6	<i>Heteromastus filiformis</i> /		
Total	209	82.3	<i>Myriochele oculata</i> /		
			<i>Spiophanes kroyeri</i> /		
			<i>Prionospio cirrifera</i>	2	0.5
			Total	342	93.2

(c) D2S			(d) A16S		
Species	N	% of total abundance	Species	N	% of total abundance
<i>Pseudopolydora pausibranchiata</i>	57	19.9	<i>Polydora caulleryi</i>	80	32.3
<i>Polydora caulleryi</i>	42	14.7	<i>Pseudopolydora pausibranchiata</i>	74	29.8
<i>Prionospio steenstrupi</i>	34	11.9	<i>Eteone longa/flava</i>	10	4.0
<i>Prionospio fallax</i>	28	9.8	<i>Echiurus echiurus</i>	10	4.0
<i>Heteromastus filiformis</i>	26	9.1	<i>Ophelina acuminata</i>	7	2.8
<i>Ophelina acuminata</i>	25	8.7	<i>Heteromastus filiformis</i>	7	2.8
<i>Echiurus echiurus</i>	18	6.3	<i>Prionospio steenstrupi</i>	7	2.8
<i>Antinoella saris</i>	8	2.8	<i>Antinoella sarsi</i>	7	2.8
<i>Spiophanes kroyeri</i>	7	2.5	<i>Trochochaeta multisetosa</i>	4	1.6
<i>Scalibregma inflatum</i>	4	1.4	<i>Spionidae</i> indet/ <i>Capitellidae</i> indet	4	1.6
Total	249	87.1	Total	206	83.1

(e) D16S			(f) D13SC6		
Species	N	% of total abundance	Species	N	% of total abundance
<i>Polydora caulleryi</i>	174	30.5	<i>Heteromastus filiformis</i>	52	28.0
<i>Prionospio steenstrupi</i>	142	24.9	<i>Polydora caulleryi</i>	38	20.4
<i>Pseudopolydora pausibranchiata</i>	60	10.5	<i>Echiurus echiurus</i>	18	9.7
<i>Heteromastus filiformis</i>	60	10.5	<i>Pseudopolydora pausibranchiata</i>	14	7.5
<i>Prionospio fallax</i>	27	4.7	<i>Ophelina acuminata</i>	12	6.5
<i>Ophelina acuminata</i>	25	4.4	<i>Prionospio steenstrupi</i>	8	4.3
<i>Spiophanes kroyeri</i>	10	1.6	<i>Spiophanes kroyeri</i>	6	3.2
<i>Prionospio cirrifera</i>	8	1.4	<i>Antinoella sarsi</i>	5	2.7
<i>Echiurus echiurus</i>	7	1.2	<i>Chaetozone setosa</i>	4	2.2
<i>Eteone longa/flava</i>	6	1.1	<i>Scalibregma inflatum</i>	3	1.6
Total	519	91.1	<i>Nephtys pente</i>	3	1.6
			Total	163	87.6

Table A.2: (continued)

(g) A13SC6			(h) B13SC6		
Species	N	% of total abundance	Species	N	% of total abundance
<i>Polydora caulleryi</i>	146	32.2	<i>Polydora caulleryi</i>	59	29.4
<i>Pseudopolydora pausibranchiata</i>	126	27.8	<i>Pseudopolydora pausibranchiata</i>	37	18.4
<i>Prionospio steenstrupi</i>	60	13.3	<i>Echiurus echiurus</i>	20	10.0
Ophiuridea indet	14	3.1	<i>Heteromastus filiformis</i>	13	6.5
<i>Echiurus echiurus</i>	13	2.9	<i>Gattyana cirrosa</i>	11	5.5
<i>Prionospio fallax</i>	9	2.0	<i>Ophelina acuminata</i>	9	4.5
<i>Corbula gibba</i>	7	1.6	<i>Eteone longa/flava</i>	6	3.0
<i>Ampelisca tenuicornis</i>	5	1.1	<i>Prionospio steenstrupi</i>	6	3.0
<i>Ophelina acuminata</i>	5	1.1	<i>Nephtys pente</i>	5	2.5
<i>Eteone longa/flava</i> <i>Nephtys pente</i>	5	1.1	<i>Trochochaeta multisetosa</i>	4	2.0
Total	390	86.1	Total	170	84.6

(i) A14SC24			(j) B14SC24		
Species	N	% of total abundance	Species	N	% of total abundance
<i>Polydora caulleryi</i>	81	29.4	<i>Pseudopolydora pausibranchiata</i>	89	33.1
<i>Pseudopolydora pausibranchiata</i>	40	14.5	<i>Polydora caulleryi</i>	67	24.9
<i>Prionospio steenstrupi</i>	38	13.8	<i>Echiurus echiurus</i>	21	7.8
<i>Echiurus echiurus</i>	22	8.0	<i>Heteromastus filiformis</i>	19	7.1
Ophiuridea indet	12	4.4	<i>Prionospio steenstrupi</i>	16	6.0
<i>Gattyana cirrosa</i>	11	4.0	Capitellidae indet	9	3.4
<i>Nephtys pente</i>	8	2.9	<i>Antinoella sarsi</i>	6	2.2
<i>Heteromastus filiformis</i>	7	2.5	<i>Eteone longa/flava</i>	5	1.9
<i>Prionospio fallax</i>	7	2.5	<i>Spiophanes kroyeri</i>	4	1.5
<i>Eteone longa/flava</i>	4	1.5	<i>Prionospio fallax</i>	4	1.5
Total	230	83.3	Total	240	89.2

(k) D14SC24		
Species	N	% of total abundance
<i>Heteromastus filiformis</i>	48	24.9
<i>Polydora caulleryi</i>	38	19.7
<i>Prionospio steenstrupi</i>	19	9.8
<i>Pseudopolydora pausibranchiata</i>	16	8.3
<i>Echiurus echiurus</i>	15	7.8
Anomiidae indet	7	3.6
<i>Prionospio fallax</i>	7	3.6
<i>Ophelina acuminata</i>	6	3.1
<i>Capitella capitata</i>	3	1.6
<i>Spiophanes kroyeri</i>	3	1.6
Total	162	83.9

Table A.3: Abundances for the feeding groups in all the boxes.

Feeding	S_A1	S_B1	S_B2	S_D1	S_D2	SK24_A	
unknown/other	8	14	4	9	4	26	
<i>Carnivore/omnivore</i>	27	20	11	13	13	21	
Scavenger/carnivore/omnivore	2	5	2	2	1	8	
<i>DF</i>	21	49	19	94	58	12	
<i>suspension/SDF</i>	84	45	179	180	43	84	
SDF/DF						2	
<i>SDF</i>	102	109	142	270	163	114	
<i>Suspension</i>	4	6	8	0	0	3	
Feeding	SK24_B	SK24_D	SK6_A	SK6_B	SK6_D	sum	
unknown/other	8	12	32	9	3	129	
<i>Carnivore/omnivore</i>	16	6	12	21	9	169	
Scavenger/carnivore/omnivore	1	1	5	5	1	20	
<i>DF</i>	31	62	13	24	70	453	
<i>suspension/SDF</i>	67	39	170	63	39	953	
SDF/DF						2	
<i>SDF</i>	144	71	239	76	61	1491	
<i>Suspension</i>	1	0	19	2	0	43	

Table A.4: Complete species list

Species_name	AMBTN_G1	AMBTS_G1	S_A1	S_B1	S_B2	S_D1	S_D2	SK24_A	SK24_B	SK24_D	SK6_A	SK6_B	SK6_D
Hydrozoa indet								1					
Anthozoa indet		1											
Hexacorallia	1												
Nemertinea indet	3					2	1	1			1		
Nematoda indet	1	13											
Polychaeta indet		1											
<i>Antinoella sarsi</i>	7			3	1	2	8		6	3	3	1	5
<i>Eunoe nodosa</i>											1		
<i>Gattyana cirrosa</i>				7	1			11		1		11	
Polynoidae indet		1										1	
<i>Eteone longaflava</i>	1		10	2	5	6	3	4	5		5	6	2
<i>Phyllodoce groenlandica</i>			3	3	1				2		1		1
<i>Phyllodoce maculata</i>								1					
<i>Phyllodoce mucosa</i>				1		1						1	
<i>Phyllodoce rosea</i>													
<i>Sige fusigera</i>						2							
<i>Nereimyra punctata</i>					1								
<i>Nereis pelagica</i>								1				1	
<i>Nephtys caeca</i>		1											
<i>Nephtys hombergii</i>		2	3	1	1	1		3	1	1	1	1	
<i>Nephtys longosetosa</i>			4									1	
<i>Nephtys pente</i>		2	2	5	2	2	1	8	1	1	5	5	3
Spharodoridae indet						1							
<i>Glycera alba</i>					2	1	1		2	1			1
<i>Gonioda maculata</i>		3											
Dorvilleidae indet								1		1			
<i>Scoloplos armiger</i>										1			
<i>Levinsenia gracilis</i>													
Paronidae indet									1				
<i>Trochochaeta multisetosa</i>			4	1	9	1	1	1	1		2	4	
<i>Polydora caulleryi</i>			80	44	170	174	42	81	67	38	126	59	38
<i>Polydora</i> sp								1					
<i>Prionospio cirrifera</i>	7	6		2	2	8	3	1	3	2	4		
<i>Prionospio fallax</i>	15	7	2	10	4	27	28	7	4	7	9	3	6

Table A.4: (continued)

Species name	AMBTN_G1	AMBTS_G1	S_AI	S_B1	S_B2	S_D1	S_D2	SK24_A	SK24_B	SK24_D	SK6_A	SK6_B	SK6_D
<i>Prionospio multibranchiata</i>							7			1			
<i>Prionospio</i> sp									1				
<i>Prionospio steenstrupi</i>	38	40	7	25	13	142	34	38	16	19	60	6	8
<i>Pseudopolydora paucibranchiata</i>	12	20	74	51	106	60	57	40	89	16	146	37	14
<i>Spio filicornis</i>	1		1	1	1	1	1					2	
Spionidae indet			4			3	2			1			
<i>Spiophanes bombyx</i>						4	1			2			
<i>Spiophanes kroeyeri</i>	8		1	1	2	10	7	1	4	3	1	3	6
<i>Spiophanes</i> sp											2		
<i>Magelona</i> sp	1												
<i>Chaetozone setosa</i>													
<i>Chaetozone setosa</i>	8		3			5	2	1	3	1	2	3	4
<i>Diploclirrus glaucus</i>													
<i>Pherusa flabellata</i>													
<i>Pherusa plumosa</i>													
<i>Scalibregma inflatum</i>	1			2	1	2	4	1		1	1	1	3
<i>Ophelina acuminata</i>	3		7	22	16	25	25	2	1	6	5	9	12
<i>Ophelina cylindricaudata</i>						2	1						
<i>Ophelina modesta</i>								1			3		
<i>Capitella capitata</i>			1	1		5	1	1		3			
Capitellidae indet			4						9				2
<i>Heteromastus filiformis</i>	4		7	22	2	60	26	7	19	48	4	13	52
<i>Mediomastus fragilis</i>				4		1	1	1	1		2	2	1
<i>Myriochele oculata</i>						2	2		1	2	1	1	1
<i>Pectinaria koreni</i>	12		7				1		2			1	1
<i>Pectinaria</i> indet													
<i>Ampharete lindstroemi</i>			1										
<i>Ampharete lindstroemi</i>													
Ampharetidae indet													2
<i>Anobothrus gracilis</i>	3												
<i>Anobothrus gracilis</i>	2		1										
<i>Sabellides octocirrata</i>								1					
<i>Sosane sulcata</i>													1
<i>Neoamphitrite</i> sp			2										1
<i>Polychaerus norvegicus</i>											2		
<i>Terbellidae</i> indet			1			1	1	3					1
<i>Terbellides stroemi</i>								1					
<i>Jasminetra caudata</i>				1							3	1	

Table A.4: (continued)

Species_name	AMBTN_GI	AMBTS_GI	S_A1	S_B1	S_B2	S_D1	S_D2	SK24_A	SK24_B	SK24_D	SK6_A	SK6_B	SK6_D
Sabellidae indet			1	1				1					
Serpulidae indet				4							3		
Gastropoda indet											1	2	
Polyplocophora indet	1							2			3	2	1
Bivalvia indet													
<i>Nucula cf. sulcata</i>										1			
<i>Nuculoma tenuis</i>										1			
<i>Yoldiella philippiana</i>											1		
<i>Mytilus edulis</i>				2									
<i>Chlamys septemradiatus</i>													
<i>Chlamys</i> sp													
<i>Chlamys striatum</i>				1	1	1			1	1		1	
Anomiidae indet						1	1		2	7			
<i>Thyasira</i> sp			2	2						1		1	
<i>Acanthocardia echinata</i>											1		
<i>Parvicardium minimum</i>											1		
<i>Abra nitida</i>						5		1		1	2		1
<i>Abra</i> sp								1					
<i>Kelliella abyssicola</i>												1	
<i>Corbula gibba</i>				2	8			3	1		7	1	
<i>Hiatella arcica</i>											1		
Calanoidea indet	10	3						1					
Cyclopoidea indet	4							2			1		
<i>Nebalia bipes</i>							1						
Cumacea indet											2		
<i>Eudorella cf. truncatula</i>								1					
Amphipoda indet			1								1		
Gammaridea indet			1				3						
Lysianassidae indet						2							
<i>Ampelisca brevicornis</i>		1	1										
<i>Ampelisca cf. typica</i>			1										
<i>Ampelisca macrocephala</i>			1	1									
<i>Ampelisca</i> sp													
<i>Ampelisca tenuicornis</i>											1		
<i>Cheirocratus sundewalli</i>			2				1			1	5		

Table A.4: (continued)

Species_name	AMBTN_GI	AMBTS_GI	S_A1	S_B1	S_B2	S_D1	S_D2	SK24_A	SK24_B	SK24_D	SK6_A	SK6_B	SK6_D
Melittidae indet				3						1			
<i>Westwoollia caecula</i>										2			1
Phoxocephalidae indet								2					
<i>Podocerus cf. falcatus</i>										1			
Caridea indet			1				1						
Decapoda indet								2					
<i>Lebbeus polaris</i>				1									
<i>Athanas nitescens</i>				1									
<i>Euaulus gainmarthi</i>				1									
<i>Euaulus pustulosus</i>									1			4	2
<i>Hippolyte varians</i>									1				1
Hippolytidae indet			1	3				1					1
<i>Thorulus cranchii</i>								1					
<i>Pandalus montaguui</i>								1					
<i>Philocheras bispinosus</i>	2												
Nephtropidae indet									2				
<i>Carcinus maenas</i>			1					1					
<i>Golfiggia cf. minuta</i>	1												
<i>Phaseolton strombi</i>		1											
<i>Echirus echirus</i>			10	19	9	7	18	22	21	15	13	20	18
Asteroidea indet				1									
Crinoidea indet								1					
Ophiuroidea indet	1					2	1	12		1	14		
<i>Ophiura cf. albida</i>				1									
Echinoidea indet							1	1			1		
<i>Echinoacartium cordatum</i>	1												
Sum	149	122	248	254	367	570	286	276	269	193	453	201	186

Table A.5: Feeding mode of each species. The categories No info and Other are placed together in the final table. DF = deposit feeders, SDF = subsurface deposit feeders.

Feeding	Species
no info	Hydrozoa indet
no info	Anthozoa indet
no info	Hexacorallia
Carnivore/omnivore	Nemertinea indet
no info	Nematoda indet
other	Polychaeta indet
carnivore/omnivore (largedetrituslicker)	<i>Antinoella sarsi</i>
Carnivore/omnivore	<i>Eunoe nodosa</i>
Carnivore/omnivore(parasite/commensal)	<i>Gattyana cirrosa</i>
no info	Polynoidae indet
carnivore/omnivore	<i>Eteone longa/flava</i>
carnivore/omnivore	<i>Phyllodoce groenlandica</i>
carnivore/omnivore	<i>Phyllodoce maculata</i>
carnivore/omnivore	<i>Phyllodoce mucosa</i>
carnivore/omnivore	<i>Phyllodoce rosea</i>
no info	<i>Sige fusigera</i>
other	<i>Nereimyra punctata</i>
no info	<i>Nereis pelagica</i>
Scavenger/carnivore/omnivore	<i>Nephtys caeca</i>
Carnivore/omnivore	<i>Nephtys hombergii</i>
Carnivore/omnivore(scavenger)	<i>Nephtys longosetosa</i>
Scavenger/carnivore/omnivore	<i>Nephtys pente</i>
no info	Spharodoridae indet
Carnivore/omnivore	<i>Glycera alba</i>
Carnivore/omnivore	<i>Goniada maculata</i>
no info	Dorvilleidae indet
DF	<i>Scoloplos armiger</i>
DF(SDF)	<i>Levinsenia gracilis</i>
no info	Paraonidae indet
suspension/SDF	<i>Trochochaeta multisetosa</i>
suspension/SDF	<i>Polydora caulleryi</i>
suspension/SDF	<i>Polydora</i> sp
SDF(suspension)	<i>Prionospio cirrifera</i>
SDF(suspension)	<i>Prionospio fallax</i>
SDF(suspension)	<i>Prionospio multibranchiata</i>
SDF(suspension)	<i>Prionospio steenstrupi</i>
SDF(suspension)	<i>Pseudopolydora paucibranchiata</i>
SDF	<i>Spio filicornis</i>
SDF(suspension)	Spionidae indet
SDF(suspension)	<i>Spiophanes bombyx</i>
SDF(suspension)	<i>Spiophanes kroeyeri</i>
SDF(suspension)	<i>Spiophanes</i> sp
SDF(suspension)	<i>Magelona</i> sp
SDF	<i>Chaetozone setosa</i>
SDF(largesandlicker)	<i>Diplocirrus glaucus</i>
no info	<i>Pherusa flabellata</i>
SDF(largesandlicker)	<i>Pherusa plumosa</i>
DF(suspension)	<i>Scalibregma inflatum</i>

Table A.5: (Continued)

DF	<i>Ophelina acuminata</i>
DF	<i>Ophelina cylindricaudata</i>
DF	<i>Ophelina modesta</i>
DF	<i>Capitella capitata</i>
DF	Capitellidae indet
DF	<i>Heteromastus filiformis</i>
DF	<i>Mediomastus fragilis</i>
SDF	<i>Myriochele oculata</i>
DF(largesandlicker)	<i>Pectinaria koreni</i>
no info	Pectinariidae indet
SDF	<i>Ampharete lindstroemi</i>
SDF	Ampharetidae indet
SDF	<i>Anobothrus gracilis</i>
SDF	<i>Sabellides octocirrata</i>
SDF	<i>Sosane sulcata</i>
no info	<i>Neoamphitrite</i> sp
SDF(dissolvedmatter/symbionts)	<i>Polycirrus norvegicus</i>
SDF	Terebellidae indet
SDF	<i>Terebellides stroemi</i>
Suspension(SDF)	<i>Jasmineira caudata</i>
Suspension	Sabellidae indet
no info	Serpulidae indet
no info	Gastropoda indet
no info	Polyplacophora indet
no info	Bivalvia indet
DF	<i>Nucula cf. sulcata</i>
DF	<i>Nuculoma tenuis</i>
no info	<i>Yoldiella philippiana</i>
no info	<i>Mytilus edulis</i>
no info	<i>Chlamys septemradiatus</i>
no info	<i>Chlamys</i> sp
no info	<i>Chlamys striatum</i>
no info	Anomiidae indet
DF	<i>Thyasira</i> sp
suspension	<i>Acanthocardia echinata</i>
suspension	<i>Parvicardium minimum</i>
suspension/SDF	<i>Abra nitida</i>
other	<i>Abra</i> sp
no info	<i>Kelliella abyssicola</i>
suspension	<i>Corbula gibba</i>
no info	<i>Hiatella arctica</i>
no info	<i>Nebalia bipes</i>
no info	Cumacea indet
no info	<i>Eudorella cf. truncatula</i>
no info	Amphipoda indet
no info	Gammaridea indet
no info	Lysianassidae indet
suspension	<i>Ampelisca brevicornis</i>
suspension	<i>Ampelisca cf. typica</i>
suspension	<i>Ampelisca macrocephala</i>

Table A.5: (Continued)

suspension	<i>Ampelisca</i> sp
suspension	<i>Ampelisca tenuicornis</i>
SDF	<i>Cheirocratus sundewalli</i>
no info	Melitidae indet
SDF	<i>Westwoodilla caecula</i>
SDF/DF	Phoxocephalidae indet
no info	<i>Podocerus cf. falcatus</i>
no info	Caridea indet
no info	Decapoda indet
no info	<i>Lebbeus polaris</i>
no info	<i>Athanas nitescens</i>
no info	<i>Eualus gaimardii</i>
no info	<i>Eualus pusiolus</i>
no info	<i>Hippolyte varians</i>
no info	Hippolytidae indet
no info	<i>Thoralus cranchii</i>
no info	<i>Pandalus montagui</i>
no info	<i>Philocheras bispinosus</i>
no info	Nephropidae indet
no info	<i>Carcinus maenas</i>
suspension/SDF	<i>Golfingia cf. minuta</i>
other	<i>Phascolion strombi</i>
no info	<i>Echiurus echiurus</i>
no info	Asteroidea indet
no info	Crinoidea indet
other	Ophiuroidea indet
no info	<i>Ophiura cf. albida</i>
no info	Echinoidea indet
SDF/DF	<i>Echinocardium cordatum</i>

Table A.6: Grain size sand: values from cumulative curve (interpolated).

Percentiles	Size(mm)	Phi (Ø)
95	0.4084	1.29
90	0.2273	2.14
84	0.1692	2.56
75	0.1169	3.10
50	0.0650	3.94
25	0.0051	7.61
16	0.0016	9.25
10	0.0005	10.83
5	0.0004	11.47

Table A.7: Sediment grain size composition (% dry weight).

Clay	17.08%
Silt	21.92%
Pelite	39.00%
Very fine sand	9.31%
Fine sand	29.54%
Medium sand	14.09%
Coarse sand	4.33%
Very coarse sand	2.25%
Total sand	59.52%
Pebbles	1.48%
Cobbles	0.00%
Gravel	1.48%

Table A.8: Median grain size for the cuttings: values from cumulative curve (interpolated).

Percentiles	Size(mm)	Phi (Ø)
95	0.4569	1.13
90	0.1799	2.47
84	0.0802	3.64
75	0.0335	4.90
50	0.0109	6.52
25	0.0046	7.75
16	0.0028	8.5
10	0.0015	9.38
5	0.0004	11.13

Table A.9: Sediment grain size composition for the drill cuttings (% dry weight).

Clay	11.43%
Silt	62.81%
Pelite	74.25%
Very fine sand	7.69%
Fine sand	5.73%
Medium sand	4.44%
Coarse sand	3.32%
Very coarse sand	2.69%
Total sand	23.88%
Pebbles	1.88%
Cobbles	0.00%
Gravel	1.88%

Table A.10: Median grain size for the fine sediment: values from cumulative curve (interpolated).

Percentiles	Size(mm)	Phi (\emptyset)
95	0.1551	2.69
90	0.1111	3.17
84	0.0915	3.45
75	0.0684	3.87
50	0.0088	6.82
25	0.0009	10.11
16	0.0005	11.11
10	0.0004	11.44
5	0.0003	11.72

Table A.11: Sediment grain size composition for the fine sediment (% dry weight).

Clay	33.11%
Silt	23.97%
Pelite	57.08%
Very fine sand	15.12%
Fine sand	21.46%
Medium sand	4.31%
Coarse sand	1.55%
Very coarse sand	0.48%
Total sand	42.92%
Pebbles	0.00%
Cobbles	0.00%
Gravel	0.00%

Appendix B

Figures

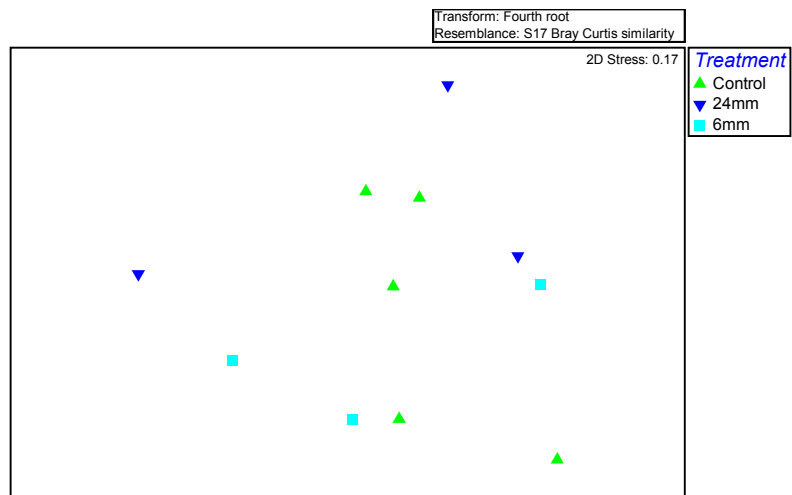


Figure B.1: Multi Dimensional Scaling (MDS) based on fourth root faunal counts, as a function of treatment without the two most abundant species, *Polydora caulleryi* and *Pseudopolydora pausibranchiata*.

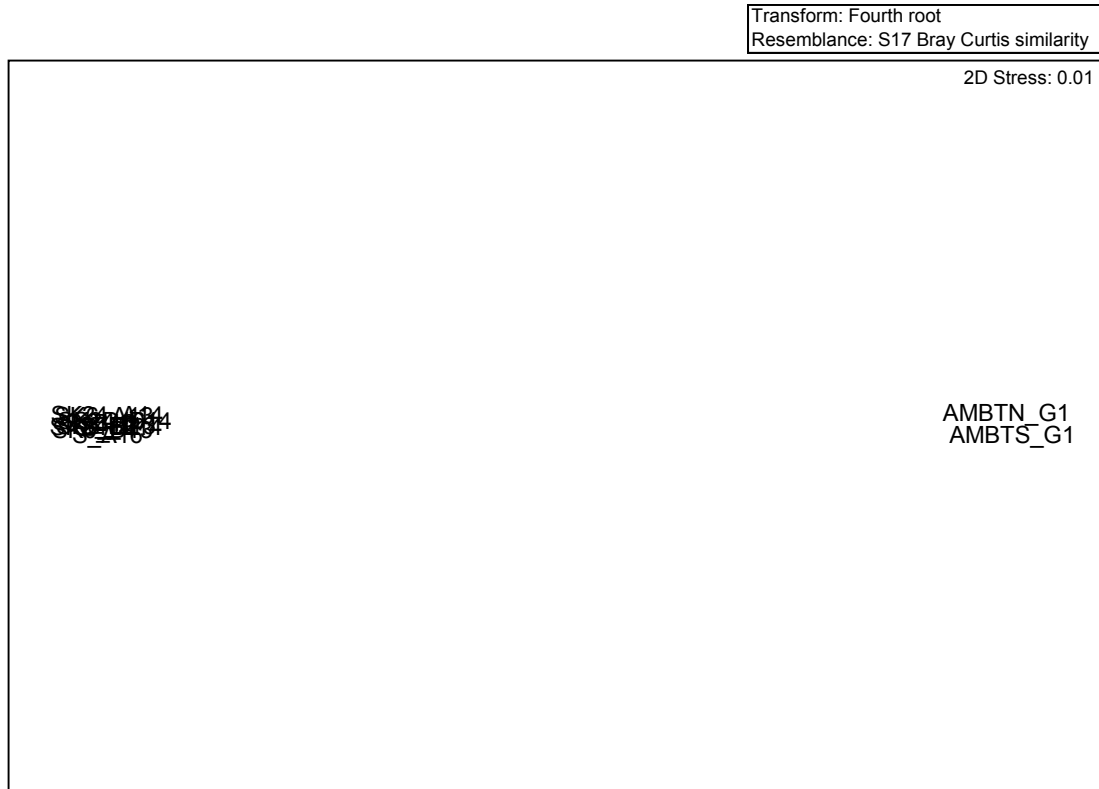


Figure B.2: Multi Dimensional Scaling (MDS) based on fourth root faunal counts, of the 11 experimental boxes and the ambient grab samples.

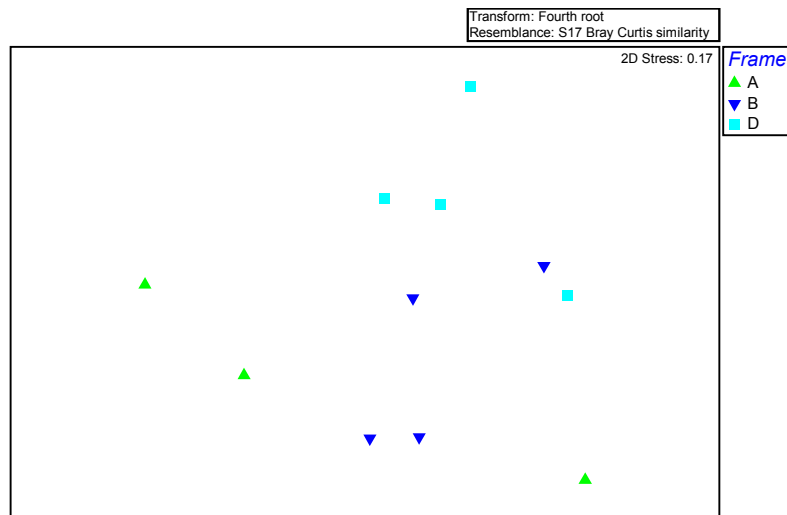


Figure B.3: Multi dimensional scaling (MDS) based on fourth root faunal counts, of the 11 experimental boxes, leaving out the two ambient grab samples, as a function of frame, without the two most abundant species, *Polydora caulleryi* and *Pseudopolydora Pausibranchiata*.

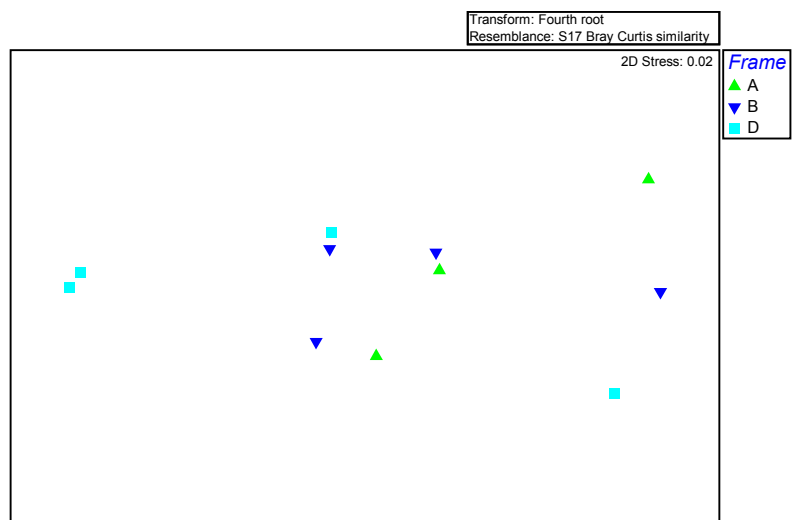


Figure B.4: Multi Dimensional Scaling (MDS) based on fourth root faunal counts, with only the two most abundant species, *Polydora caulleryi* and *Pseudopolydora pausibranchiata*.