



Impact of Thin Deposits of Terrigenous Clay on Benthic Communities

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Impact of Thin Deposits of Terrigenous Clay on Benthic Communities

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1 Executive Summary

This study assessed the impact of the deposition of thin layers of terrigenous clay on estuarine communities and ecosystem processes. It is one of a series of studies commissioned by ARC to provide information on the threats to estuarine ecology posed by changes in land use.

Potential ecological threats include loss of sensitive species, changes in biodiversity, reduced oxygenation of surficial sediment, shifting microbial activity, diminished light levels and restricted photosynthesis within the sediments and interference with animal feeding processes across the sediment surface.

These issues are focused into three major questions that were addressed by field experiments:

1. What is the critical depth of thin (<1 cm) clay deposits that cause chronic effects?
2. How do benthic communities in different habitats respond to thin clay deposits?
3. How does frequent deposition of thin layers of terrigenous clay affect benthic communities?

The experimental sites encompassed a variety of common habitats that cover a range of hydrodynamic conditions, sediment properties and benthic community composition.

Results showed that thin clay deposits ranging from 3 – 7 mm depth have an impact on benthic communities, with negative effects increasing with the depth of deposited clay. However, these responses were not of the magnitude recorded from earlier studies in Okura estuary which described the impact of > 2 cm deposits of clay.

The response of the macrobenthic community varied with habitat, with the subtidal habitat, in particular, showing little response to the experimental additions of clay. Nevertheless, community analysis indicated that repeated additions of 3 mm layers of clay over a 6-month period had a cumulative effect on the macrobenthic community.

Of the biogeochemical sediment properties measured variations in response dependent on habitat and timing of clay deposition were observed. At some sites and some times we recorded ecologically important changes in nutrient fluxes and in the abundance of microphytes in the sediment.

It is clear from the experiments that the frequency of disturbance and thus the time available for recovery between disturbance events is critical in assessing whether the deposition of thin layers of terrigenous clay pose a threat of broad-scale degradation to the Whitford embayment.

Low load depositional events that produce thin layers of clay are likely to be more frequent and to cover a larger area than rare catastrophic events. It will be important to use the catchment and hydrodynamic modelling to assess the risk of low-load events depositing clay in different parts of the embayment. The experiments

presented in this report have provided us with information that will enable us to refine the rules for assessing the risk of ecological change as a result of sediment inputs.

2 Introduction

Estuarine environments are rich in both structural and biological diversity and play an important role in the functioning of coastal ecosystems (Heip et al. 1995). Although these environments are characterised by large fluctuations in the quantity and quality of suspended sediment in their waters (Navarro & Widdows, 1997), changes in land-use and modification of coastlines due to human development have increased rates of sedimentation and changed the areal extent of depositional environments in estuaries (Edgar & Barrett, 2000).

The Auckland Region is a rapidly expanding area with a current population of 1.1 million, which is projected to reach two million by the year 2050. To contain this expanding population, the Auckland region is undergoing continual urban and semi-rural development, and expansion of the infrastructure that is necessary to support this development. The urban development process usually results in higher than usual amounts of storm-water contamination by sediment, which eventually ends up in the region's rivers, estuaries and coastal ecosystems. Similarly, episodic events such as landslides, extreme rain events and flooding can result in catastrophic deposition of sediments and elevated suspended sediment concentrations. There is growing recognition that sediments pose a threat to the biodiversity of estuaries and coastal areas (Gray 1997) by profoundly influencing the structure and function of macrobenthic communities (Ellis et al. 2000).

The ARC is determining the risk that the urban development process poses to the receiving environment of the Whitford Embayment in a series of studies.

- Catchment modelling will determine the potential for sediment entry into the three estuaries entering the embayment.
- Hydrodynamic modelling will investigate dispersal within and through the system, concentrating on potential areas for increases in sediment deposition and suspended sediment concentrations.
- Studies of mangrove and salt marsh communities of the estuaries and fringing environment of the larger embayment will determine their sensitivity to increased deposition and the effect they may have on depositional patterns.
- Laboratory and field experiments will determine macrofaunal species, communities and habitats sensitive to sediment deposition.
- Laboratory and field experiments will determine the potential for sublethal effects of increased suspended sediment concentrations on the macrofauna.

This study deals with identifying the impacts of the deposition of thin layers of terrigenous clay on macrobenthic populations and communities as well as on various biogeochemical processes.

Estuaries and tidal inlets are particularly vulnerable to increased levels of sedimentation, as they act as natural retention systems. Accelerated deposition of

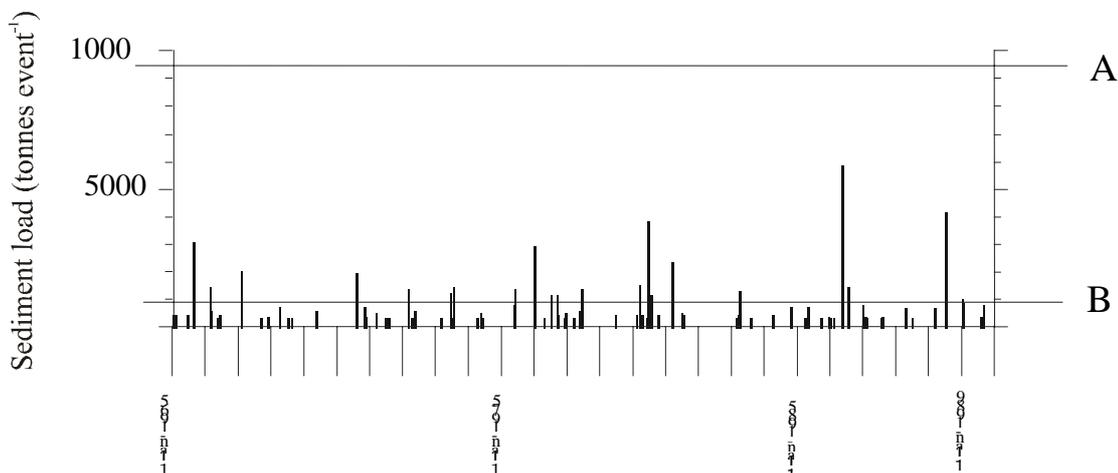
land-derived sediment leads to habitat modification and impacts on estuarine ecology by directly and indirectly killing, displacing, or damaging components of the macrobenthic community, resulting in changes to the abundance and distribution of benthic organisms. The magnitude of impact depends on spatial and temporal scales and the frequency of depositional events. Ecological repercussions can be both short-term catastrophic and long-term chronic in nature.

Recent field and laboratory studies have examined the catastrophic effects of sudden sediment deposits on macrofauna communities in Okura estuary (Norkko et al. 1999, Nicholls et al. 2000). From these studies it is evident that terrigenous sediment deposits deeper than 2-3 cm and which cover the underlying sediment for 5-7 days will smother natural sediments, turn it anaerobic and kill all resident macrofauna. With the exception of highly mobile crustaceans (mud crabs and shrimps), common estuarine animals are unable to move through thick sediment deposits, and even if animals are not killed before the sediment is removed, individual animals are sufficiently stressed to be more susceptible to sub-lethal effects such as predation.

Modelling predictions for Okura estuary document the risk of impact and frequency of such catastrophic sedimentation events (Cooper et al. 1999, Stroud et al. 1999). Accordingly, critical catchment loads that result in sediment deposits of at least 2 cm depth are infrequent and event-driven. This is illustrated by the catchment modelling conducted for the Okura estuary study.

Figure 1 presents the predicted sediment loads coming off the Okura catchment under existing land use conditions (Fig 18 in Stroud et al. 1999). On this we have superimposed the critical sediment load required to exceed a 2 cm depositional event (A) and a 2-3 mm event (B) in one of Okura estuary's sub-environments (S4 from Table 5 Stroud et al. 1999). The catchment modelling predicts that there has been insufficient load to generate a 2 cm depositional event in this sub-environment, whereas a 2-3 mm event is predicted to have occurred 21 times over the modelled period. Although the immediate impact will be associated with sediment deposition in specific sub-environments, these small events are more likely to cover a broader expanse of the estuary. Thus it is important that we consider the ecological effects of these non-catastrophic depositional events, as they will occur more frequently and more extensively.

Fig. 1. Modelled sediment load derived from the catchment of Okura Estuary under present land-use. "A" is the critical load needed to produce a 2 cm deep depositional event in the S4 sub-environment. "B" is the critical load needed to produce a 2-3 mm deep depositional event. See Stroud et al. (1999).



However, no information is currently available regarding the potential effects of thin clay deposits on benthic communities. Even though there is a general understanding that potential impacts would be less catastrophic, the long-term consequences are unknown. Whilst thin layers of clay may not result in smothering and mortality of all infauna, more subtle changes may occur. Potential ecological responses include both functional and structural changes to benthic communities. These could include loss of sensitive species, changes in biodiversity, reduced oxygenation of surficial sediment, shifting microbial activity to anaerobic processes, diminished light levels and restricted photosynthesis by microphytobenthos (microalgae), and interference with feeding processes across the sediment surface (see also Gibbs et al. 2001; Hatton et al. 2001; Norkko et al. 2001).

The current lack of information provided the impetus for the present study, which presents an assessment of the potential impact of thin layers of land-derived sediments on estuarine organisms. It is one of a suite of investigations currently conducted by NIWA (commissioned by ARC) to assess the risks of changes in land use on the Whitford embayment. This work is also supported by NIWA's research programme "Effects of Sediments on Estuarine Ecosystems" funded by the Foundation for Research Science and Technology.

This report presents the results of field studies designed to assess the potential risk of thin layers of terrigenous clay (< 1 cm deep) to the benthic community in the Whitford embayment. Through a series of field experiments we investigated ecological effects on macrofauna community structure and ecosystem dynamics, and addressed three key questions:

- What is the critical depth of thin (<1 cm) clay deposits that cause chronic effects?
- How do benthic communities in different habitats respond to thin clay deposits?

- How does frequent deposition of thin layers of terrigenous clay affect benthic communities?

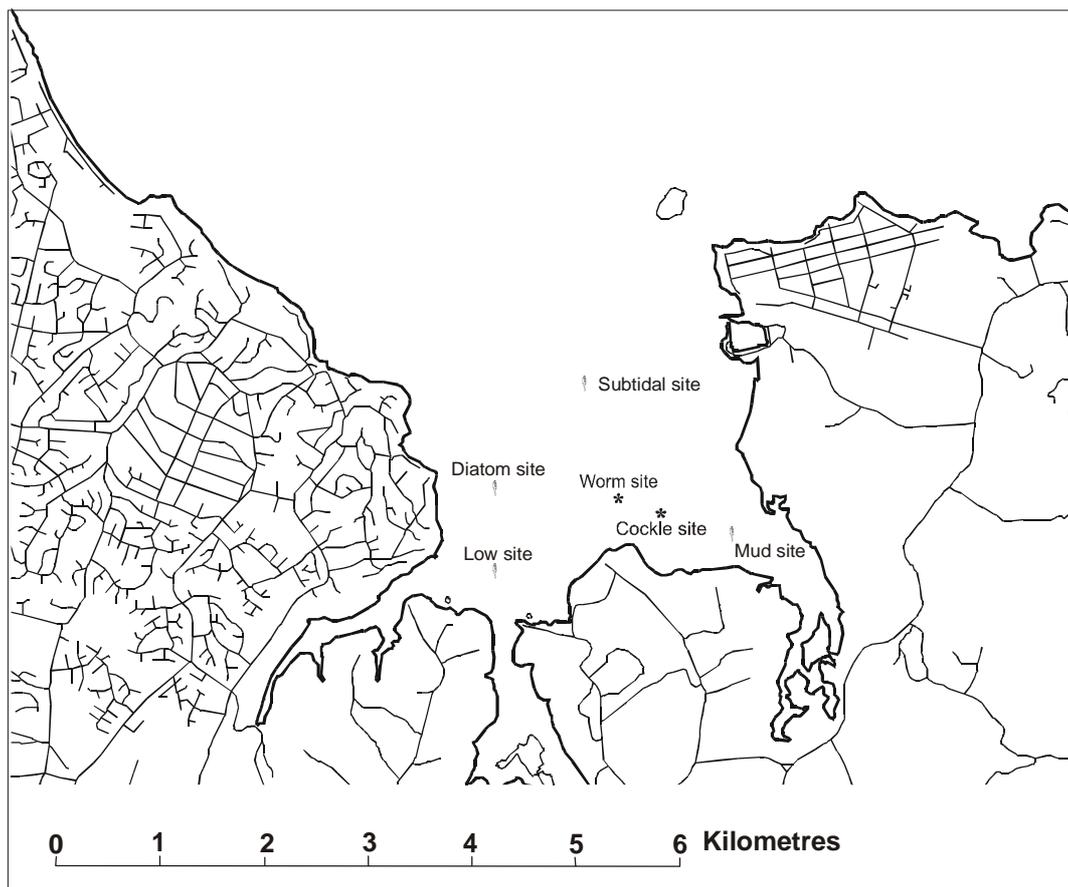
We addressed these questions through three field experiments, which allowed us to generalise effects across a wide variety of habitats and benthic communities. In this report, findings from the three-fold study are presented consecutively.

3 Methods

3.1 General experimental set-up

In order to assess ecological effects across different habitats, one subtidal and five intertidal sites were selected within the Whitford embayment (Fig. 2). Based on preliminary information from the habitat survey of the Whitford embayment (NIWA report due for submission to Manukau City Council in 2002), experimental sites were chosen to represent a variety of common habitats that cover a range of hydrodynamic conditions, sediment properties and benthic community composition.

Figure 2: Location of study sites in the Whitford embayment.



Terrigenous clay was obtained from earthworks at the Sandstone landfill site. Clay was broken up and mixed into a slurry at a ratio of clay to seawater of 33:67. The clay slurry was stored over night before re-mixing and transport to the experimental sites. The clay mix was deposited at different thickness onto the sediment surface of circular experimental plots of $\sim 4.5\text{m}^2$.

3.2 Field experiment 1: (What is the critical depth of thin (<1 cm) clay deposits that cause chronic effects?)

Two experimental sites were selected at mid-tide level, approximately 200 m apart and exposed to similar hydrodynamic conditions. The two sites were, however, markedly different in their community composition. The Cackle site was dominated by cockles *Austrovenus stutchburyi*, whereas polychaete worms and the wedge shell *Macomona liliiana* dominated the benthic community at the Worm site (Fig. 2).

Experimental plots were arranged in a Latin square design with a distance between plots of 10 m (from midpoint to midpoint). In order to account for any possible effects due to differences in tidal height and inundation, each row was placed parallel to the shoreline.

Five different treatments (different thickness of terrigenous clay) were applied to the experimental plots at both sites on the 7th of November 2000. Treatment levels were chosen to reflect a range of thin sediment layers less than 1 cm deep and included 0 (Control), 1, 3, 5, and 7 mm clay depth. Each treatment was replicated four times, with the exception of 7 mm treatments, which had two and three replicates at the Cackle and Worm site, respectively.

Experimental plots were established by pushing thin sheet metal rings into the sediment surface (to ~5 cm depth) to provide a bund for the clay mixture. The appropriate quantity of clay slurry was then carefully sprayed onto the sediment surface of each plot. The clay was left to settle over two tidal cycles, before the rings were removed and the sampling initiated (Photo 1 and 2).

Photo 1. Tide inundating the sandflat and submerging recently created experimental plots.



Photo 2. Treatment plot one tide after clay deposition.



Each experimental plot was sampled four times over the ten day experimental period. Sediment cores (2.4 cm diameter, 2 cm depth) were taken on day 1, 3, 7 and 10, to measure particle size, organic content, chlorophyll *a* and pore water ammoniacal nitrogen ($\text{NH}_4\text{-N}$ which includes NH_3 ; hereafter referred to as $\text{NH}_4\text{-N}$). For each variable, paired core samples were taken and combined prior to analysis. The sampling location within each plot was recorded on each occasion to avoid re-sampling of the same area on subsequent visits. Sediment samples for organic content, chlorophyll *a* and $\text{NH}_4\text{-N}$ analysis were immediately chilled and kept in the dark. Three measurements of the depth of deposited clay and any overlying sand (moved over the plot by sediment bedload transport) were taken from one of the sediment cores.

Sediment cohesiveness was determined at three haphazard positions within each plot using a shear vane and a penetrometer, which respectively measure the lateral and vertical force required to move surface sediment (i.e., top 5 mm). At the same time, photographs of each plot were taken to document visible surface characteristics and activities of large macrofauna on the sediment surface. Ten days after the initial deposition of clay, two replicate cores (13 cm diameter, 15 cm depth) were collected for macrofauna analysis, and the depth of the clay and any overlying sand layers was recorded for each core. This sampling marked the termination of the experiment.

3.3 Field experiment 2: (How do benthic communities in different habitats respond to thin clay deposits?)

The second experiment included the Cackle site used in Field experiment 1 and three additional sites (Fig. 2). Accordingly, the Low site was positioned relatively low on the shore in relation to tidal height, the Diatom site was in an area previously determined as having high sediment chlorophyll *a* concentrations, and the Mud site was characterised by very fine-grained, muddy sediment at the transition into the mud/mangrove habitat (Craggs et al. 2001).

The experiment was initiated on the 16th January 2001. At each site, experimental plots were arrayed in a randomised block design, with controls located in the middle of each block. An appropriate quantity of clay slurry was added to experimental plots to produce clay layers of 0 (Control), 1, and 5 mm depth. Each treatment level was replicated four times. The treatment levels of 1 and 5 mm were chosen to reflect two thin clay layers that were within the tested range in Field experiment 1, and that were shown in that experiment to have an ecological effect. The application procedure followed that described for Field experiment 1. However, we noted the pH of the clay slurry in the first experiment was low (~ pH 4). To negate any potential effects that might be due to the acid nature of the clay (see Section 4) the clay mixture was buffered to pH 7.2 (with an aqueous solution of sodium-hydroxide, NaOH). A field experiment was also conducted to assess the duration and spatial extent of low pH associated with the deposition of the unbuffered clay slurry used in Field experiment 1 (see section below).

Physical and biogeochemical characteristics of the sediment were determined after 1, 3, 7 and 9 days. From each plot, one sediment core (5 cm diameter, 2 cm depth) was collected for sediment grain size and organic content analysis, and paired sediment cores (2.4 cm diameter, 2 cm depth) were combined for chlorophyll *a* analysis. The cohesiveness of surface sediment was measured at three haphazard positions in each plot using a shear vane and a penetrometer as described for Field experiment 1. Photographs of each plot were taken to visually record the sediment surface of each plot. Nine days after the initial application of clay, the experiment was terminated and two replicate macrofaunal cores (13 cm diameter, 15 cm depth) were taken from each experimental plot.

3.3.1 Assessing nutrient flux across the sediment-water interface

At the Low and the Diatom sites, sealed chambers were deployed to measure the nutrient exchange between the sediment and overlying water in each experimental plot. Oxygen and nutrient levels measured in the samples reflect biogeochemical processes associated with each plot. Chambers consisted of clear plastic domes (14.5 cm diameter, 8 cm height) with an attached reservoir bag and a fastened and lockable sampling port (Photo 3). The chambers were sealed to avoid mixing between incoming seawater and water in the chambers. In order to keep the water volume within each chamber constant, the exact amount of water removed from each chamber during sampling was replenished from a reservoir bag, which had been filled with seawater at the beginning of the experiment.

Photo 3. Chamber and attached reservoir bag at the beginning of the experiment.



Starting 3 h before high tide on the 18th of January 2001, chambers and reservoir bags were filled with incoming seawater and were carefully placed into each experimental plot (one chamber per plot). Water samples (60 ml) were taken from each chamber on four occasions over the tidal cycle. Initial samples were collected when tidal inundation over the site was approximately 20 cm deep, a second sample was taken one hour later, when the tidal height was approximately 1 m. The chambers were then left over high tide and re-sampled at similar water levels on the out-going tide. Dissolved oxygen (DO) was immediately measured in each sample using a calibrated Yellow Springs Instrument (YSI) BOD probe (0.05 ppm accuracy). Subsequently, water samples were filtered (Whatman GF/C glass fibre filter) and stored frozen pending analysis for NH₄-N and nitrate (NO₃-N).

3.3.2 pH experiment

Another separate component of this field experiment examined the pH chemistry of unbuffered clay in the estuarine environment. Due to the acidic nature of the clay (pH 3-4), we examined the pH level of a 3 mm clay layer in relation to neutralisation by seawater. The pH level of the sediment was measured in a 1.2 m² experimental plot and an adjacent control area near the Diatom site by using a pH microelectrode (Diamond General, tip diameter 0.9 mm) and a micromanipulator. Measurements were taken at 5 mm intervals to a total depth of 30 mm, immediately after the application of clay, and 1, 2 and 10 tidal cycles later.

3.3.3 Subtidal habitat

Apart from assessing ecological effects across different intertidal habitats, this part of the field study also examined the ecological effects of clay deposits on a subtidal benthic community. Although the experimental design of the subtidal study followed the experimental protocol at the intertidal sites, the former was considered as a

separate component within Field experiment 2. This is due to the large contrast in hydrodynamic and sediment characteristics and differences in biota between intertidal and subtidal environments, which preclude a meaningful direct comparison between both types of estuarine environment.

A site was chosen for the subtidal experiment based on underwater video and diver observations made as part of the fieldwork for the Whitford embayment habitat survey (NIWA report due for submission to Manukau City Council in 2002). The Subtidal site (Fig. 2) was typical of the shallow subtidal environment that dominates the bay. We chose an area just below the sand dollar zone (*Fellaster zelandiae*), with a relatively low density of crab burrows. Crab burrow density tends to increase with water depth and, at the same time, the sediment becomes muddier. The site was in approximately 2 m of water and the substrate consisted of a fine layer of recently resuspended flocculent matter overlying fine sand.

To assist underwater navigation in low visibility the experimental lay out used in the subtidal experiment was different from the design used on the intertidal flats. We used a randomised block design, with each block forming the corner of a rectangle and each block separated by at least 5 m. The experimental plots were the same size as those used intertidally (~ 4.5m²) and replication and treatment levels were the same as those used in Field experiment 2. Experimental plots that were to be covered with clay were banded by metal rings pushed into the sediment surface. Plastic sheeting was bound to these rings to enclose the surface of the experimental plot. The appropriate quantity of clay slurry was then injected into the enclosed space above the ring, to achieve clay layers of 1 and 5 mm depth. After a settling period of 1-2 h, the rings and sheeting were removed.

Sediment cores (5 cm diameter, 2 cm depth) for grain size, organic content and chlorophyll *a* analysis were taken by divers on day 1, 3, and 7. The site was videoed, and observations were made of the clay and sediment surface. The high water content and non-cohesive nature of the sediment surface meant that we were unable to measure sediment geotechnical properties using the penetrometer or shear vane. Macrofaunal samples were collected by coring (10 cm diameter, 14 cm depth) at the end of the experiment, on day 7.

3.4 Field experiment 3: (How does frequent deposition of thin layers of terrigenous clay affect benthic communities?)

The third field experiment investigated ecological effects of frequent clay deposition, and was initiated in conjunction with Field experiment 1, on 7th of November 2000. It was conducted at both the Cackle and the Worm site and followed the same experimental set-up as Field experiment 1. At monthly intervals, over a period of six months, a 3 mm layer of clay was deposited on the same four experimental plots at each site. Each month, paired sediment cores (2.4 cm diameter, 2 cm depth) were collected and pooled for particle size, organic content, and chlorophyll *a* analysis prior to the re-application of clay mixture. The depth of the clay and any sand layers were measured from one of the sediment cores. At the same time, sediment cohesiveness

was measured (shear vane and penetrometer) and the surface of each experimental plot was photographed. The same sampling procedure was applied at the end of the field experiment, on the 11th of April 2001. In addition, two replicate macrofaunal cores (13 cm diameter, 15 cm depth) were collected and the depth of deposited clay and any overlying sand was recorded for each core.

3.5 Laboratory analysis

Macrofauna samples were sieved on 0.5 mm mesh, preserved in 70% isopropyl alcohol and stained with 0.2% Rose Bengal. Macrofaunal specimens were sorted and identified to the lowest taxonomic level practicable. The most common bivalves (*Austrovenus stutchburyi* and *Macomona liliana*) were measured and grouped into three different size-classes, small (<5 mm), medium (5-15 mm) and large (>15 mm).

Chlorophyll *a* was extracted from sediment samples by boiling the freeze-dried sediment in 95% ethanol. The extract was processed using a spectrophotometer. An acidification step was used to separate degradation products from chlorophyll *a* (Sartory 1982).

Organic content was determined by change in mass after drying the sediment at 60°C for 48 h and combusting in a muffle furnace for 5.5 h at 400°C. Samples for particle size analysis were pre-treated by digesting sediments in 6% hydrogen peroxide for 48 h to remove organic matter, and dispersed using Calgon. Subsequently, % volumes for sediment fractions (gravel, coarse, medium and fine sand, silt and clay) were determined by wet-sieving (particle size > 63µm) and by using a Galai particle analyser (Galai Cis - 100; Galai Productions Ltd., Midgal Haemek, Israel)(particle size < 63µm).

Water samples collected from the chambers were analysed for NH₄-N and NO₃-N following the seawater methods of Grasshoff et al. (1983) using an Alphkem series 500 seawater auto-analyser. In this report, NO₃-N is taken to include any nitrite nitrogen (NO₂-N).

Photographs from each plot were examined for the visual appearance of the clay layer and distinct biological features on the sediment surface.

3.6 Statistical analysis

We used a variety of univariate and multivariate statistical procedures to determine patterns in macrobenthic community structure and the significance of variation apparent in the experiment.

Univariate analysis started with the calculation of the five most common taxa of the macrofauna community at each site in each treatment and the total number of individuals and taxa found in each plot. These were then used as variables in the univariate analysis. For Field experiments 1 and 2, regression models were developed within a generalized linear modelling framework (McCullagh & Nelder 1989), using model criteria to select for appropriate error structures and link functions. Linear

regressions were applied to relate macrofaunal densities and sediment characteristics to clay depth. For Field experiment 1 significant differences between the sites meant that the sites were analysed separately. Partial regression analysis was used to account for the considerable spatial variability observed across and along the sandflat within each site, and results presented in this report exclude within-site spatial effects. For Field experiment 2, initial models were developed that included site-treatment interaction terms. Where significant interactions were detected the data was split to reveal treatment effects. Also in Field experiment 2, due to the smaller area encompassed by each experimental site, we did not need to account for within-site spatial variation so partial regression was not needed. For Field experiment 3, paired t-tests (or non-parametric equivalents if appropriate) were performed on the independent experiments conducted in November 2000 and April 2001.

Initial multivariate analysis for Field experiments 1 and 3 was carried out on the mean data from each treatment with a down weighting of rare taxa by correspondence analysis (see Legendre & Legendre 1998 for a full description of multivariate procedures). For Field experiment 1, canonical correspondence analysis was subsequently applied to this data using clay depth and all sediment characteristics as explanatory variables. This enabled us to identify which environmental variables accounted for most variation in macrobenthic community composition. For Field experiment 3, the significance of decreasing overlap in community composition over time was tested using distance-based redundancy analysis. For Field experiment 2, we used a classification procedure based on Bray-Curtis similarities calculated from raw data to assess differences in the community composition between sites and treatment. The significance of differences was assessed using Analysis of Similarities (Clarke 1993).

4 Clay weathering and ion exchange

Clay minerals are crystalline solids, typically less than 1 μm in diameter and are generally produced by terrestrial weathering of igneous rocks (such as basalt or granite). Clays are composed of layered sheets of aluminium silicate, in which oxygen atoms surround a central silicon atom. The four most abundant clay types (illite, kaolinite, montmorillonite and chlorite) differ in the number and layering of these sheets. The exterior surfaces of these aluminium silicate sheets possess a small negative charge, due to the electronegativity of the peripheral oxygen atoms. The overall effect is a negative surface charge of the clay mineral, which causes it to attract and adsorb cations, primarily hydrogen, alkali, and earth alkali metal ions. The degree to which a specific clay mineral can bind or exchange cations with its surroundings depends on the relative ion concentrations, the ability of a particular ion to compete for an adsorption site and the cation exchange capacity (CEC) of the clay mineral. For example, montmorillonite and illite have very high CECs, whereas kaolinite and glauconite have lower ones. New Zealand brown soils, which are also found around the Whitford/Howick area, commonly contain substantial percentages of illite (mica) and montmorillonite (vermiculite), although kaolinitic components often predominate. (Hewitt 1998; Rijkse and Hewitt 1995; Soil Bureau 1968).

During chemical weathering, hydrogen and hydroxide ions in interstitial water react with the rocks' crystalline lattice, resulting in breakage of some cation-oxygen bonds. This hydrolysis reaction is greatly accelerated in the presence of acids (such as present in rainwater) and can lead to a significant adsorption of hydrogen ions by the clay matrix. When the clay comes into contact with river water (whose cation content is higher than that of rainwater), incorporation of cations (K^+ , Ca^{2+} , and trace metal ions) and exchange with hydrogen ions (H^+) can occur.

Upon delivery to the estuary, clay minerals react with seawater. Most cation exchange occurs in estuaries, due to the large difference in cation concentrations between river and seawater. As riverborne clay materials enter seawater, adsorbed potassium and calcium are displaced by sodium and magnesium, because the Na^+/K^+ and $\text{Mg}^{2+}/\text{Ca}^{2+}$ ratios are higher in seawater than in river water. If the clay reacts directly with seawater, Na^+ and Ca^{2+} can directly replace H^+ . Release of hydrogen ions, if remaining unbuffered, can exert toxic effects on organisms living in it. This was tested in Field experiment 2.

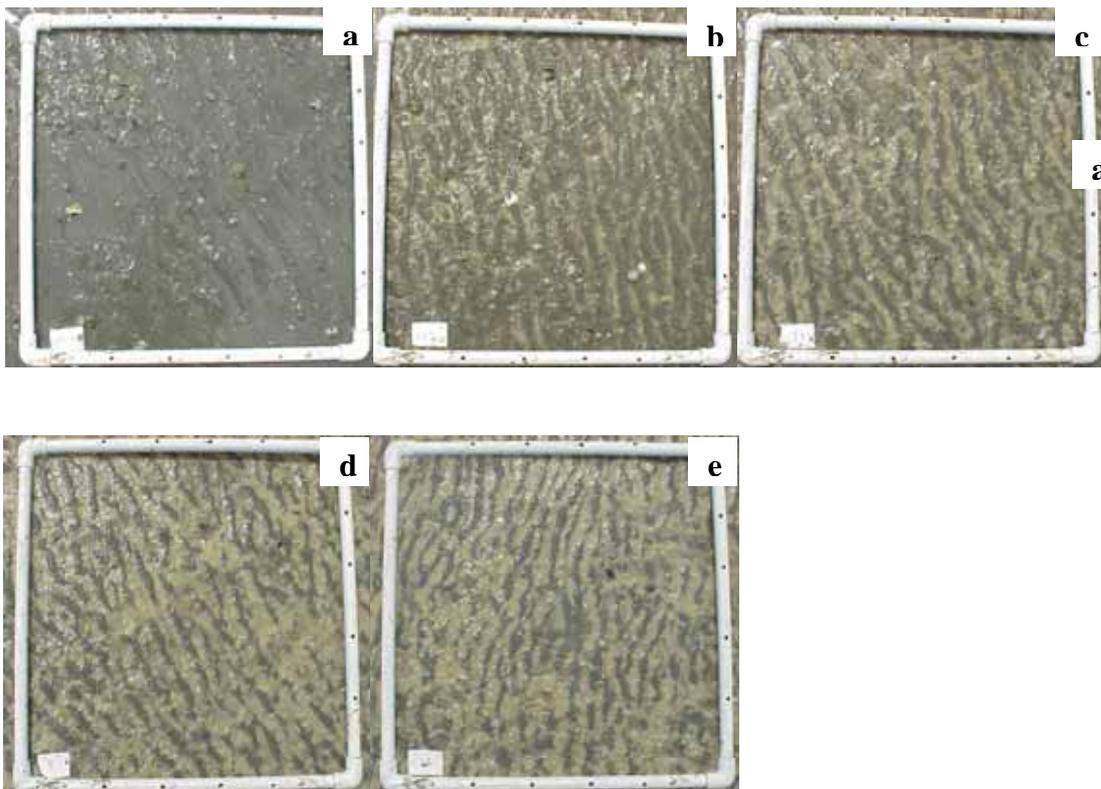
5 Results

5.1 Field experiment 1: (What is the critical depth of thin (<1 cm) clay deposits that cause chronic effects?)

5.1.1 Physical and biogeochemical sediment characteristics

The deposited clay slurry quickly formed a cohesive layer on the surface of the sediment. Its presence was clearly visible as a light-coloured homogenous layer in each plot, which appeared to change little over time (Photo 4).

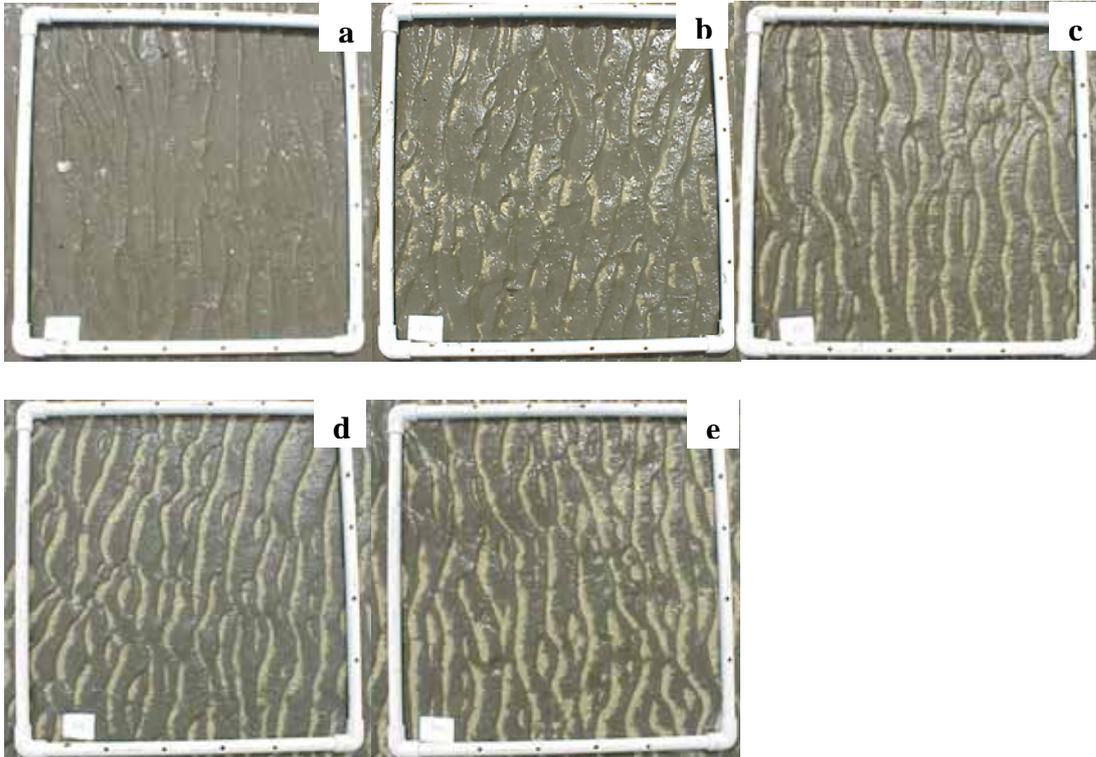
Photo 4. Experiment 1: Cockle Site



Experiment 1: Cockle site one tide after the application of the clay to experimental plots. The control plot (a) shows little sign of sand ripples indicating that the movement of cockles and other infauna is influencing the appearance of the sediment surface. Experimental plots are sequentially ordered in increasing clay depth (b – e). Note all experimental plots clearly show bedload-transported sands sitting on the terrigenous clay. The movement of cockles across the sediment surface is already

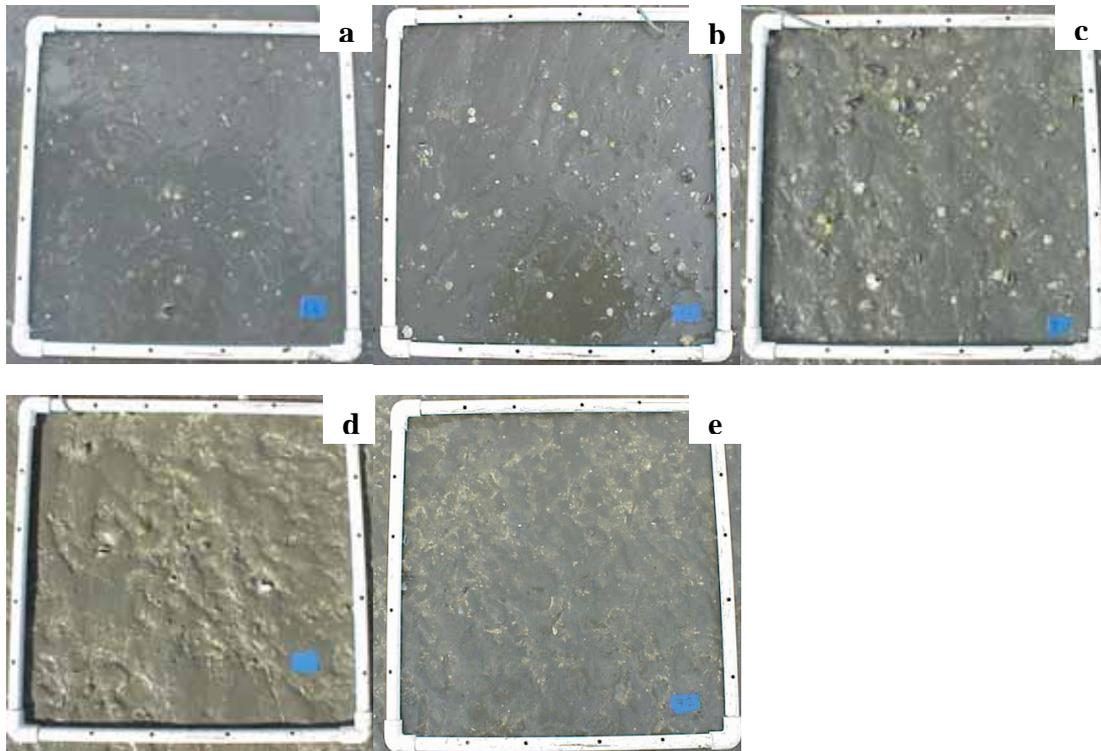
influencing the 1 mm clay layer. Photo quadrat (0.25m²), note orientation of individual photos is not necessarily consistent.

Experiment 1 Worm Site



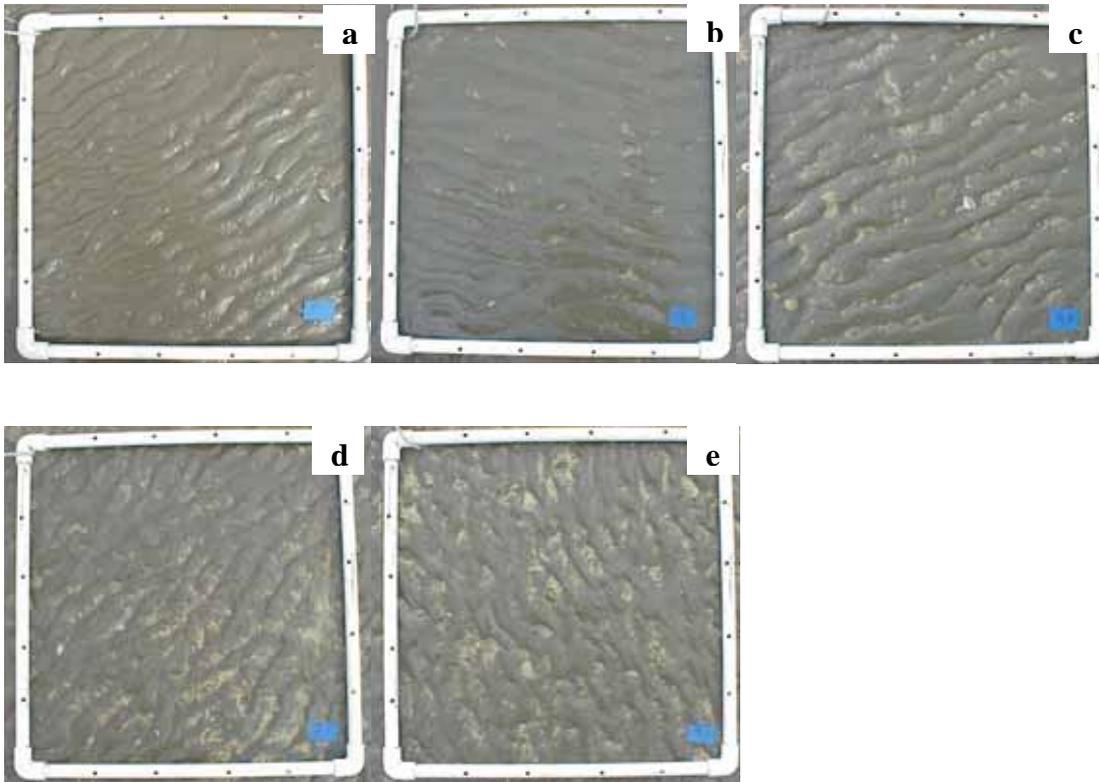
Experiment 1: Worm site one tide after the application of the clay to experimental plots. The control plot (a) shows sharp and well-defined sand ripples indicative of a recent wave event. Experimental plots are sequentially ordered in increasing clay depth (b – e). All experimental plots clearly show bedload-transported sands sitting on the terrigenous clay. Photo quadrat (0.25m²), note orientation of individual photos is not necessarily consistent.

Experiment 1 Cockle site



Experiment 1: Cockle site 10 days after the application of the clay to experimental plots. The control plot (a) shows well-defined sand ripples indicative of a recent wave event. Experimental plots are sequentially ordered in increasing clay depth (b – e). Not all experimental plots show signs of the clay deposit, the cockle movement has facilitated the erosion from the 1 and 3 mm plots. Recently dead cockles can be seen in the remaining clay layer of the 5 and 7 mm plots. Photo quadrat (0.25m²), note orientation of individual photos is not necessarily consistent.

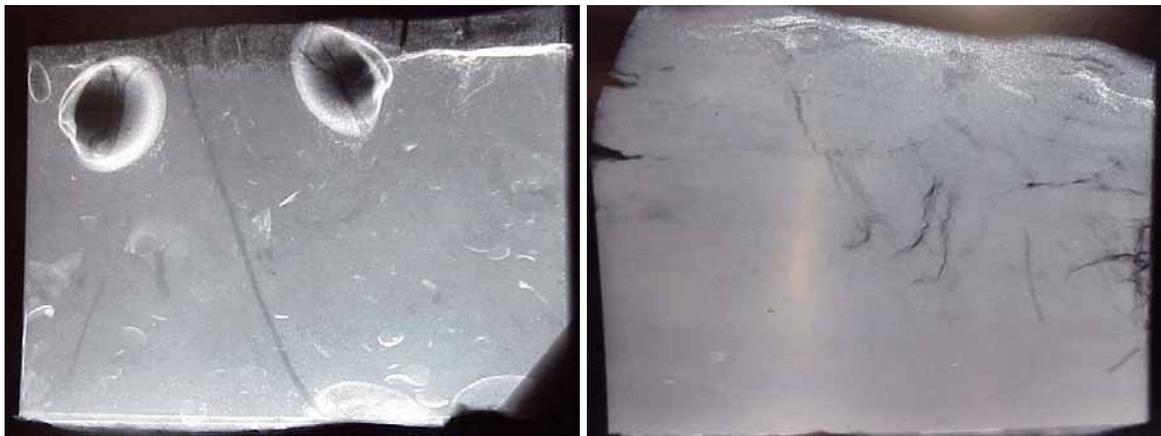
Experiment 1 Worm site



Experiment 1: Worm site 10 days after the application of the clay to experimental plots. The control plots show more rounded and less well defined sand ripples, indicative of lower wave energy. Experimental plots are sequentially ordered in increasing clay depth (b – e). All experimental plots still show some clay underlying the sand ripples, although the quantity increases with the depth originally deposited. Holes and smearing of the clay layer reflect erosion and bioturbation. Photo quadrat (0.25m²), note orientation of individual photos is not necessarily consistent.

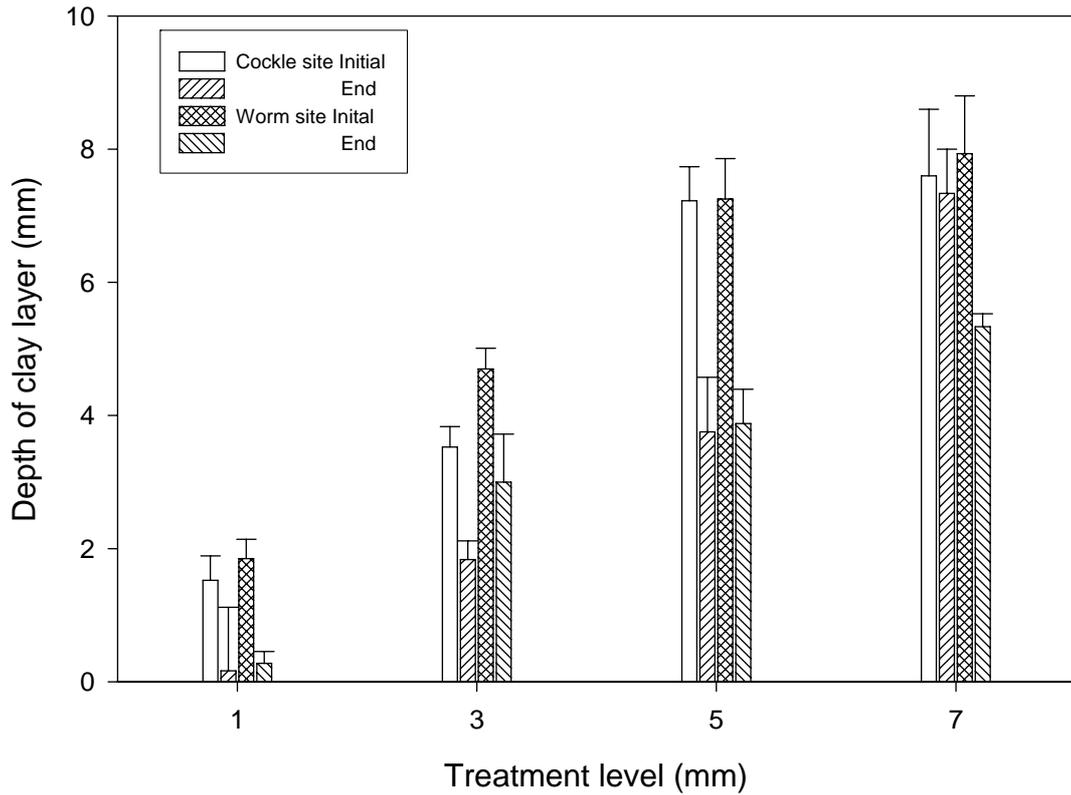
On occasion, some of the experimental plots became partially covered by sand, but this did not appear to cause a break-up of the clay layers. After ten days, clay deposits were still obvious in most of the experimental plots, with the exception of the 1 mm treatments. There, the clay layers showed signs of erosion and biological reworking (Photo 5).

Photo 5. X-radiograph sediment from the Cockle site (left). Note the bright band just below the clay layer on the sediment surface, this is a particle tracer - the animal burrow running up through the sediment has subducted the tracer down to the bottom of the image. X-radiograph from the Worm site (right). Tracer and clay sitting on the sediment surface with little indication of burrowing activity.



The thickness of the clay layers decreased over the study period (Fig. 3) and, apart from the 1 mm treatments, there was still at least 50% of the initial clay depth remaining at the end of the experiment. Differences between the target and the actual depth of deposited clay on day 1 were due to the nature of the intertidal sandflats, where the surface of the sediment contains many small natural depressions and elevations, even on a mm-scale. Statistical analysis confirmed that differences at both sites between treatments were significant at the beginning and end of the experiment (both $P = 0.0001$). This demonstrates that terrigenous clay layers of only a few mm deep can persist for at least ten days.

Figure 3. Depth of the clay layer (mm) in the experimental plots at the Cackle and Worm site at the beginning (Initial) and end (End) of the experiment. Values are expressed as the mean \pm standard error.



Physical sediment characteristics of the control plots changed little over time and were dominated by a high proportion of fine sand, with a small amount of silt and clay (Table 1). Not surprisingly, in the experimental plots at both sites, the proportion of fine sand in the sediment decreased with increasing clay depth and the proportion of silt and clay increased with clay depth. Ten days after the initiation of the experiment, trends of increasing clay content with treatment levels were still apparent.

Table 1: Volumetric composition (%; Mean \pm SE) of the surficial sediment at the start (Day 1) and end (Day 10) of the experiment. C = control; 1, 3, 5, 7 mm experimental treatment (depth of clay layer).

	Sediment particle size composition %%				
Cockle site					
DAY 1	C	1mm	3mm	5mm	7mm
Clay (0-3.9 μ m)	2.8 \pm 1.0	4.6 \pm 0.2	7.4 \pm 0.6	10.5 \pm 2.5	9.0 \pm 0.1
Silt (3.9-63 μ m)	0.1 \pm 0.0	0.3 \pm 0.1	0.6 \pm 0.1	0.5 \pm 0.2	0.9 \pm 0.5
Fine sand (63-250 μ m)	95.6 \pm 1.2	93.5 \pm 0.6	90.3 \pm 0.8	88.0 \pm 2.8	89.5 \pm 0.7
Medium sand (250-500 μ m)	0.3 \pm 0.1	0.4 \pm 0.2	0.3 \pm 0.1	0.4 \pm 0.1	0.2 \pm 0.0
Coarse sand (500-2000 μ m)	0.4 \pm 0.1	0.2 \pm 0.1	0.2 \pm 0.0	0.1 \pm 0.0	0.1 \pm 0.0
Gravel (>2000 μ m)	0.8 \pm 0.1	0.9 \pm 0.3	1.2 \pm 0.6	0.3 \pm 0.2	0.2 \pm 0.1
DAY 10					
Clay (0-3.9 μ m)	4.4 \pm 1.2	4.3 \pm 0.9	6.0 \pm 1.3	7.0 \pm 1.1	13.5 \pm 4.0
Silt (3.9-63 μ m)	0.3 \pm 0.0	0.4 \pm 0.1	0.4 \pm 0.1	2.3 \pm 1.3	0.8 \pm 0.2
Fine sand (63-250 μ m)	91.2 \pm 2.0	89.6 \pm 4.6	87.5 \pm 3.6	88.5 \pm 1.4	84.4 \pm 4.8
Medium sand (250-500 μ m)	0.1 \pm 0.1	0.3 \pm 0.1	0.3 \pm 0.1	0.4 \pm 0.0	0.6 \pm 0.2
Coarse sand (500-2000 μ m)	0.2 \pm 0.1	0.1 \pm 0.0	0.2 \pm 0.0	0.2 \pm 0.1	0.3 \pm 0.1
Gravel (>2000 μ m)	3.8 \pm 2.5	5.3 \pm 3.8	5.7 \pm 4.3	1.6 \pm 1.4	0.4 \pm 0.3
Worm site					
DAY 1					
Clay (0-3.9 μ m)	1.5 \pm 0.7	1.4 \pm 0.4	4.1 \pm 0.5	5.3 \pm 1.0	6.0 \pm 0.7
Silt (3.9-63 μ m)	0.1 \pm 0.1	0.1 \pm 0.0	0.4 \pm 0.1	1.5 \pm 1.0	0.4 \pm 0.1
Fine sand (63-250 μ m)	98.2 \pm 0.8	97.4 \pm 0.4	94.8 \pm 0.8	92.4 \pm 1.1	93.1 \pm 0.8
Medium sand (250-500 μ m)	0.0 \pm 0.0	0.3 \pm 0.1	0.2 \pm 0.1	0.1 \pm 0.0	0.1 \pm 0.0
Coarse sand (500-2000 μ m)	0.0 \pm 0.0	0.1 \pm 0.0	0.0 \pm 0.0	0.1 \pm 0.1	0.0 \pm 0.0
Gravel (>2000 μ m)	0.2 \pm 0.2	0.6 \pm 0.3	0.3 \pm 0.1	0.6 \pm 0.4	0.4 \pm 0.2
DAY 10					
Clay (0-3.9 μ m)	2.4 \pm 0.5	4.5 \pm 1.9	5.4 \pm 0.7	10.1 \pm 3.0	8.2 \pm 2.4
Silt (3.9-63 μ m)	0.4 \pm 0.1	1.0 \pm 0.5	0.9 \pm 0.2	0.9 \pm 0.2	1.0 \pm 0.2
Fine sand (63-250 μ m)	96.9 \pm 0.4	85.2 \pm 8.8	93.6 \pm 0.9	88.2 \pm 3.2	89.6 \pm 2.2
Medium sand (250-500 μ m)	0.0 \pm 0.0	8.9 \pm 8.9	0.1 \pm 0.0	0.2 \pm 0.1	0.4 \pm 0.1
Coarse sand (500-2000 μ m)	0.0 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0	0.1 \pm 0.0
Gravel (>2000 μ m)	0.3 \pm 0.2	0.4 \pm 0.2	0.0 \pm 0.0	0.4 \pm 0.4	0.6 \pm 0.6

Assessment of sediment cohesiveness showed little difference in shear strength between the sites (Table 2). Note that penetrometer readings were not taken at the Cockle site due to the high density of cockle shells in the sediment. At the start of the experiment, values showed large differences between experimental treatments at both sites. However, after ten days the difference between experimental treatments had decreased, although trends of decreasing sediment cohesiveness with increased clay thickness were still apparent.

Table 2: Sediment cohesiveness and biogeochemical characteristics (Mean \pm SE) of the surficial sediment. Results from linear regression at the start (Day 1) and end (Day 10) of the experiment are also given (significant values in bold). C = control; 1, 3, 5, 7 mm experimental treatment (depth of clay layer).

Cockle site							
DAY 1	C	1mm	3mm	5mm	7mm	P	r
Shear strength (kg cm ⁻²)	1.4 \pm 0.2	1.6 \pm 0.2	0.4 \pm 0.2	0.3 \pm 0.3	0.0 \pm 0.0	0.000	0.6681
Penetrometer (kg cm ⁻²)							
Organic content (%)	0.8 \pm 0.0	1.02 \pm 0.1	1.04 \pm 0.1	1.21 \pm 0.2	0.99 \pm 0.1	0.030	0.511
Chlorophyll <i>a</i> (μ g cm ⁻²)	56.3 \pm 6.3	60.2 \pm 5.0	53.8 \pm 1.9	53.8 \pm 6.4	53.3 \pm 1.0	0.670	0.01
NH ₄ -N (μ g cm ⁻²)	36.8 \pm 29.1	28.1 \pm 15.4	14.4 \pm 4.1	11.9 \pm 7.3	4.2 \pm 0.0	0.149	0.148
DAY 10							
Shear strength (kg cm ⁻²)	0.5 \pm 0.0	0.5 \pm 0.0	0.4 \pm 0.0	0.4 \pm 0.0	0.3 \pm 0.1	0.000	0.6228
Penetrometer (kg cm ⁻²)							
Organic content (%)	1.1 \pm 0.1	1.0 \pm 0.0	1.1 \pm 0.0	1.2 \pm 0.0	1.2 \pm 0.2	0.141	0.1481
Chlorophyll <i>a</i> (μ g cm ⁻²)	52.4 \pm 3.8	52.2 \pm 7.2	53.4 \pm 4.6	50.6 \pm 6.2	40.2 \pm 3.8	0.009	0.0085
NH ₄ -N (μ g cm ⁻²)	9.5 \pm 1.8	24.5 \pm 13.3	73.0 \pm 61.7	24.7 \pm 8.7	9.0 \pm 0.3	0.681	0.01345
Worm site							
DAY 1							
Shear strength (kg cm ⁻²)	1.1 \pm 0.1	0.8 \pm 0.1	0.1 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0	0.000	0.6909
Penetrometer (kg cm ⁻²)	3.6 \pm 0.2	3.1 \pm 0.2	1.4 \pm 0.3	0.1 \pm 0.1	0.0 \pm 0.0	0.000	0.7265
Organic content (%)	0.6 \pm 0.1	0.6 \pm 0.0	0.7 \pm 0.0	0.8 \pm 0.0	0.8 \pm 0.0	0.736	0.0078
Chlorophyll <i>a</i> (μ g cm ⁻²)	25.6 \pm 1.3	25.9 \pm 1.7	31.5 \pm 2.2	27.6 \pm 2.2	33.2 \pm 3.9	0.042	0.0391
NH ₄ -N (μ g cm ⁻²)	8.8 \pm 2.5	5.08 \pm 0.3	5.2 \pm 0.6	3.7 \pm 0.3	2.7 \pm 2.7	0.005	0.42128
DAY 10							
Shear strength (kg cm ⁻²)	0.5 \pm 0.0	0.6 \pm 0.0	0.5 \pm 0.0	0.4 \pm 0.0	0.3 \pm 0.0	0.0001	0.8448
Penetrometer (kg cm ⁻²)	3.0 \pm 0.1	3.0 \pm 0.3	1.9 \pm 0.4	0.6 \pm 0.1	0.5 \pm 0.0	0.0001	0.8658
Organic content (%)	0.6 \pm 0.0	0.81 \pm 0.0	0.8 \pm 0.0	0.93 \pm 0.1	0.97 \pm 0.03	0.000	0.54484
Chlorophyll <i>a</i> (μ g cm ⁻²)	41.4 \pm 3.7	50.0 \pm 5.0	45.3 \pm 3.1	45.6 \pm 3.7	33.16 \pm 2.33	0.275	0.1985
NH ₄ -N (μ g cm ⁻²)	11.2 \pm 2.5	5.1 \pm 2.3	7.8 \pm 2.3	4.8 \pm 1.5	9.40 \pm 3.30	0.585	0.02352

The organic content of the sediment was higher at the Cockle than at the Worm site, but both sites showed low values with no major differences between experimental treatments or between the start and end of the experiment (Table 2). The proportion of organic matter in the experimental plots generally increased slightly with the depth

of the clay layer, and by the end of the study this relationship was significant at both sites.

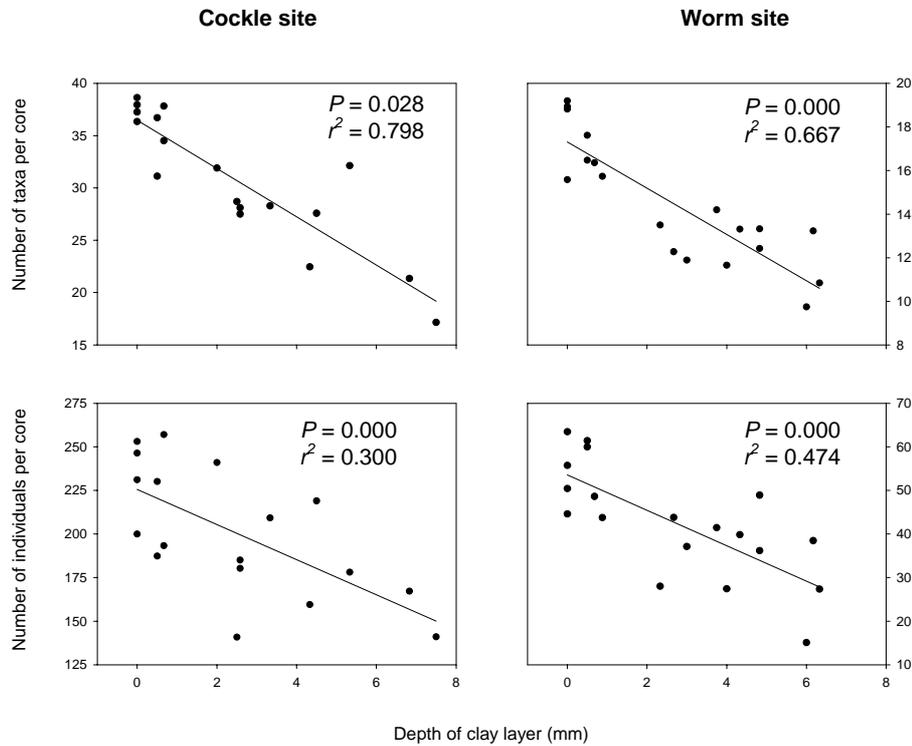
Sediment chlorophyll *a* concentrations were initially about twice as high at the Cockle site, however, at the end of the experiment the two sites differed by about 20 %. After ten days, sediment chlorophyll *a* decreased with increasing clay depth, but this relationship was only significantly at the Cockle site (Table 2).

The sediment pore water $\text{NH}_4\text{-N}$ content was also different between both sites at the start of the experiment, again the Cockle site recorded the highest concentrations (Table 2). At this site, the $\text{NH}_4\text{-N}$ level in the control plots was more than four times higher than at the Worm site. Nevertheless, both sites exhibited a negative relationship between $\text{NH}_4\text{-N}$ and clay depth. The decline in $\text{NH}_4\text{-N}$ content was much greater at the Cockle site, where levels in 7 mm treatments dropped to 11% of those in the control plots. At the Worm site, sediment $\text{NH}_4\text{-N}$ decreased to 30% of the control values. After ten days, control plots at both sites contained much less $\text{NH}_4\text{-N}$ and the previously observed difference between sites was less apparent.

5.1.2 Responses of the macrofauna community

Although the two sites were of similar sediment grain size, the macrobenthic community, like many of the sediment biogeochemical and geotechnical properties, was markedly different. The macrofaunal community at both sites clearly responded to the deposition of thin layers of clay over the 10 days of the experiment. Both number of taxa and individuals exhibited a significant negative relationship with clay depth (Fig. 4). Note the data presented in Fig. 4 (and Fig. 5) are derived from the partial regression models and thus corrected for spatial variation within each of the experimental sites. Although there was no evidence of mass mortality, there was a marked decline in both community measures. The level of impact depended on the depth of the clay layer, but even low treatment levels had a perceptible effect on both parameters. Accordingly, lowest values were recorded in 7 mm experimental plots, where values declined to about 50% of the number of taxa and individuals in the control plots. These patterns are remarkably consistent between sites despite the much higher density and diversity at the Cockle site.

Figure 4: Relationship between the number of taxa and individuals and clay depth (mm) at the Cockle and Worm sites at the end of the experiment. Note the different scales (on the y-axis) used at each site.

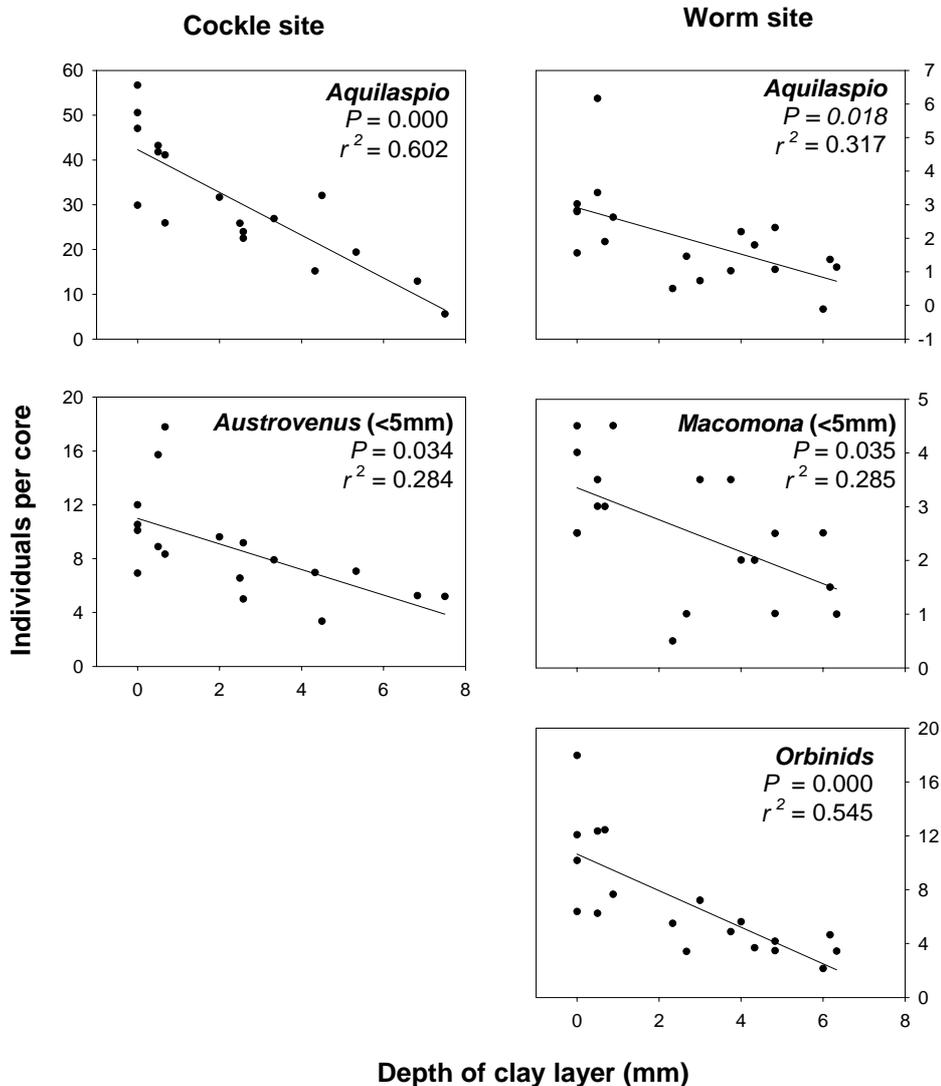


The benthic communities were dominated by different taxa at each site (Table 3). Within each community, individual species responded to an increase in clay depth with a decrease in abundance. In particular, the spionid polychaete worm *Aquilaspio aucklandica* and small-sized (<5 mm) wedge shells *Macomona liliana* significantly declined at both sites (Fig. 5). In addition, the number of small-sized (<5 mm) *Austrovenus stutchburyi* diminished at the Cockle site and the abundance of the two orbinid polychaetes (*Orbinia papillosa* and *Scoloplos cylindifer*) declined with increasing clay depth at the Worm site. Note, we have presented the total abundance of orbinids and nereids only, as it is difficult to identify juvenile individuals of either taxa to species level. None of the common species from either site demonstrated a significant trend of increasing abundance with the addition of clay.

Table 3: Rank order of the five most abundant taxa found in the control plots at the Cockle and Worm sites at the end of the experiment. Means and standard errors are presented for each taxa, based on density per core.

		Mean \pm SE
Cockle site	<i>Aquilaspio aucklandica</i>	25.1 \pm 5.0
	<i>Nucula hartvigiana</i>	20.1 \pm 2.8
	<i>Austrovenus stutchburyi</i>	16.9 \pm 4.0
	Orbinids	14.7 \pm 2.4
	<i>Chamaesipho columna</i>	12.2 \pm 0.4
	Number of taxa	27.0 \pm 2.4
	Number of individuals	161.4 \pm 30.3
Worm site	<i>Aonides oxycephala</i>	18.1 \pm 2.9
	<i>Macomona liliana</i>	9.6 \pm 1.7
	Orbinids	8.7 \pm 1.7
	<i>Aquilaspio aucklandica</i>	2.4 \pm 0.5
	<i>Paracalliope</i>	2.2 \pm 0.5
	Number of taxa	16.0 \pm 1.3
	Number of individuals	48.8 \pm 3.2

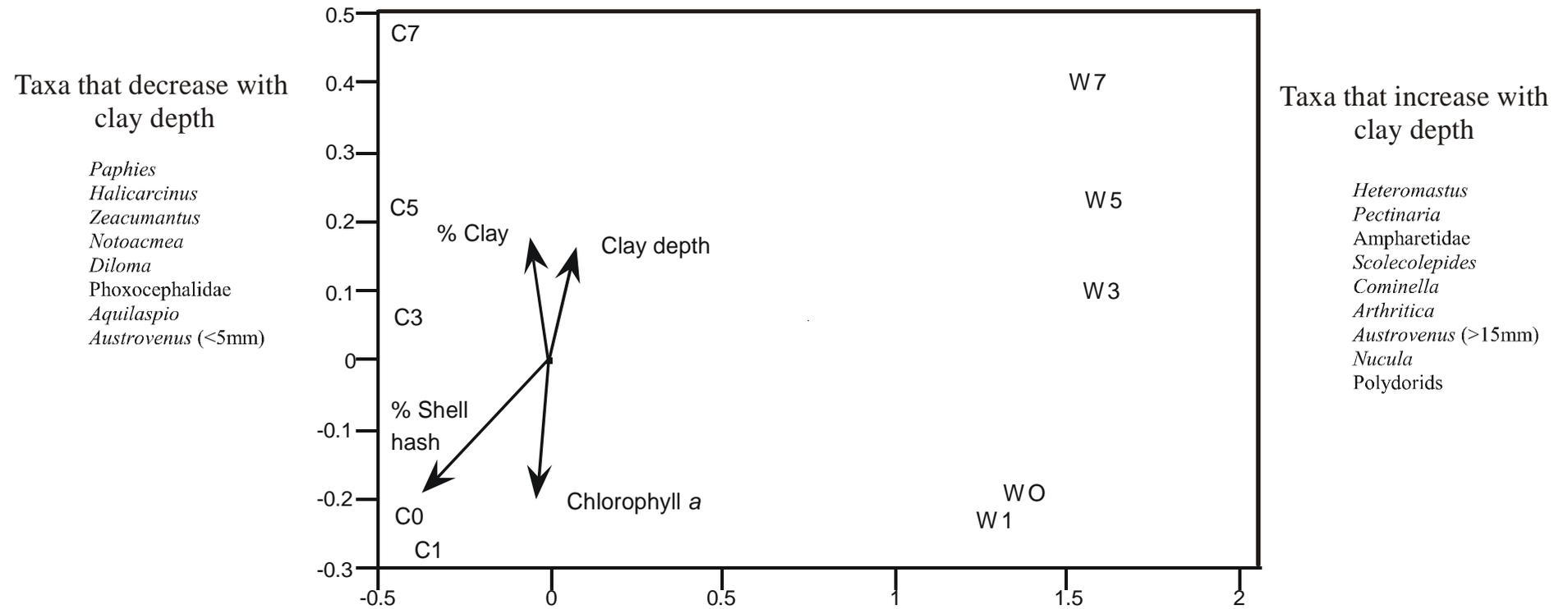
Figure 5: Relationship between individual common taxa and clay depth (mm) at the Cackle and Worm sites at the end of the experiment. Note the different scales (on the y-axis) used at each site.



Correspondence analysis revealed consistent changes in macrofaunal community structure at the Cackle and Worm site in relation to clay depth (Figure 6). The first two axes of the ordination accounted for 47.8% and 35.7% of the variability in the macrofauna data respectively. Axis 1 separates the composition of the benthic community at the two sites, while a clear separation along axis 2 with increasing clay depth is apparent for both sites. Differences in community structure increased with clay depth, and assemblages in 7 mm treatment plots were the most dissimilar to those in control plots. As well as providing information on the distribution of experimental treatments in the ordination space, the correspondence analysis also allowed us to identify individual species that are tending to change abundance relative to the different depth of clay. Species tending to increase or decrease in abundance

with increasing clay depth are indicated in Figure 6. This information, along with a previous study (Norkko et al. 2001), provides an indication of how macrobenthic communities could change in the Whitford embayment with increasing sedimentation events. Finally, to confirm that the spread of the data was due to the clay additions rather than other unknown spurious factors, we used canonical correspondence analysis to identify which of the suite of environmental variables were important in accounting for the spread of the experimental sites in the ordination space. The four environmental factors (Figure 6) produced a significant relationship with the ordination of macrobenthic community data ($P = 0.026$) and accounted for 67% of the variability in community composition. None of the available environmental variables are particularly good at discriminating between the two sites. The best discriminator was the presence of shell fragments in the sediment, which was highest at the site with high cockle densities. However, clear discrimination of the experimental treatments was given by sediment chlorophyll *a* concentration, sediment clay content and clay depth. Chlorophyll *a* concentration decreased with increasing level of clay addition, whereas sediment clay content increased with treatment level. As a consequence, the presence and depth of clay appears to be the underlying factor, prompting changes in community composition. Although the magnitude of the effect increased with clay depth, findings from this Field experiment clearly show that even very thin clay deposits have an impact on benthic communities.

Figure 6: Ordination plot of macrofauna community composition at the Cocker (C) and Worm (W) sites for each treatment level (0 to 7mm) at the end of the experiment. The arrows indicate the direction and importance environmental variables revealed by canonical correspondence analysis. Changes in abundance of individual taxa with clay depth are illustrated on either side of the ordination plot.



5.2 Field-experiment 2. (How do benthic communities in different habitats respond to thin clay deposits?)

5.2.1 Physical and biogeochemical sediment characteristics

The appearance and characteristics of clay layers in this field experiment were very similar to those in Field experiment 1 (Photo 6). Accordingly, visual differences between treatments persisted throughout the experiment, with the 1 mm clay layers changing the most over time due to erosion and reworking (Figure 7). At the end of the experiment (Day 9), 5 mm treatments at each site were significantly different to the controls and 1 mm treatments (General linear models, $P < 0.05$ for all sites).

Figure 7. Depth of the clay layer (mm) in the experimental plots at the Cockle (C), Low (L), Mud (M) and Diatom (D) sites at the beginning (Initial) and end (End) of the experiment. Values are expressed as the mean \pm standard error.

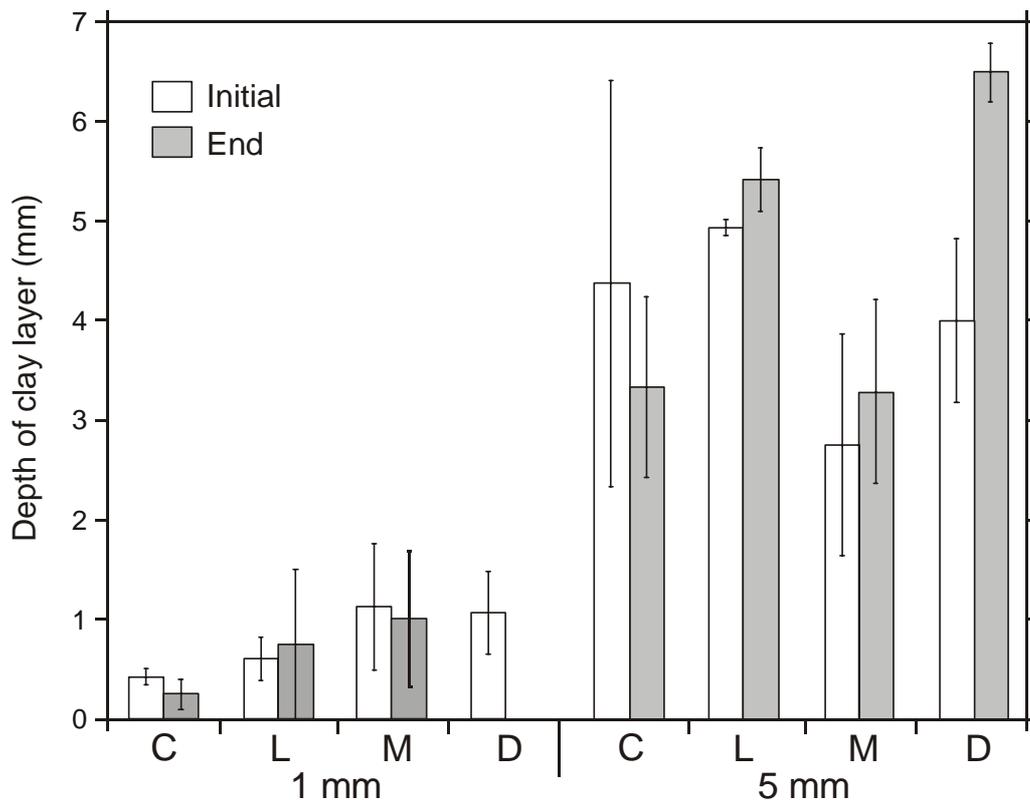
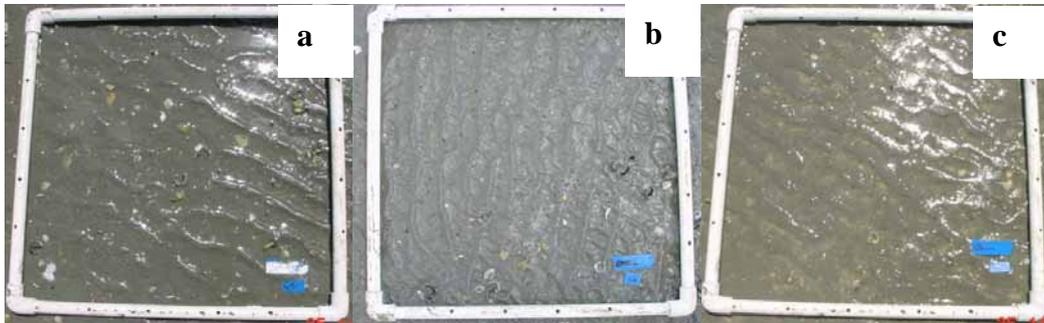
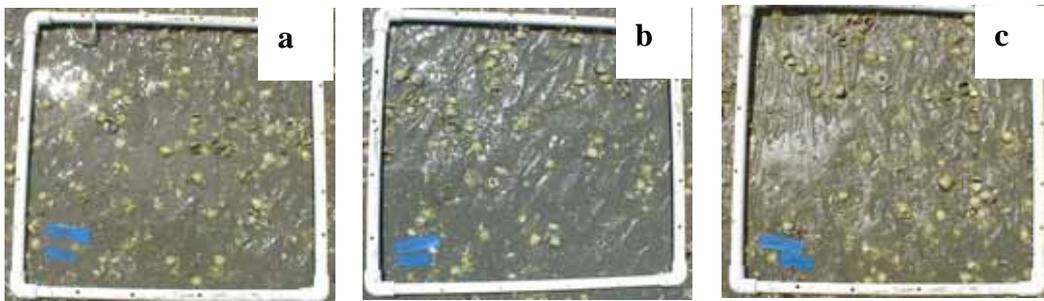


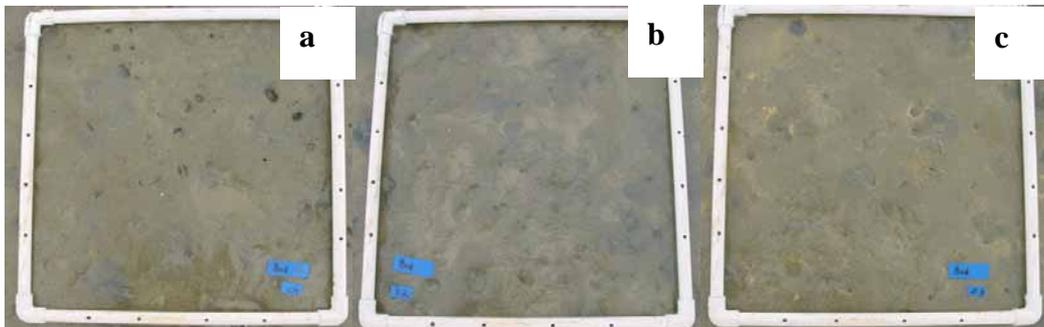
Photo 6. Experiment 2 Cockle site



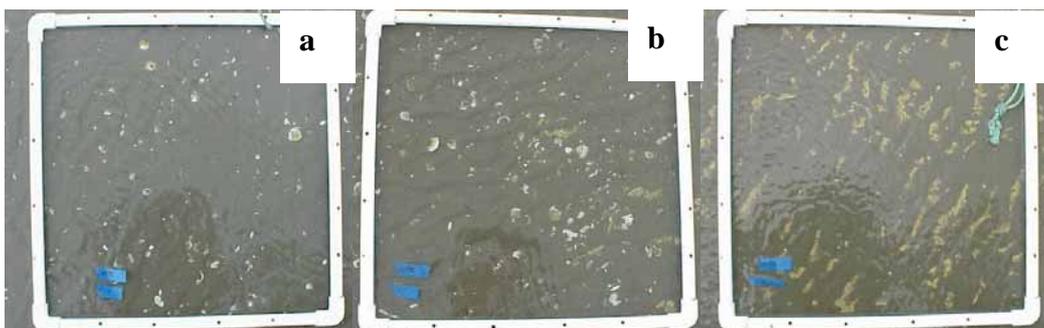
Experiment 2 Low site



Experiment 2 Mud site



Experiment 2 Diatom site



Experiment 2. Photo-quadrat showing differences between the four intertidal sites in the 3 experimental treatments (a, control; b, 1 mm; c, 5 mm). Photographs were taken at the end of the experiments, 9 days after the clay application. Large quantities of shell are apparent in the Cockle, Low and Diatom sites, while mud crab burrows are visible at the Mud site. Variations in the size and structure of sand ripples at the different sites provide a clue as to variations in wave climate. Terrigenous clay is visible in the 5 mm plots (c) except at the Low site. Photo quadrat (0.25m²), note orientation of individual photos is not necessarily consistent.

The sediment grain size distribution of the control plots was dominated by fine sand in all four intertidal habitats (Table 4). However, at the Mud site, the natural sediment in the control plots was about 25% clay. The amount of clay in the experimental plots increased with clay depth in all habitats, except at the Mud site. It is likely that we did not detect a change in the proportion of clay due the relatively high background level at this site.

Table 4: Volumetric composition (%; Mean \pm SE) of the surficial sediment at the end (Day 9) of the experiment. C = control; 1, 3, 5, 7 mm experimental treatment (depth of clay layer).

	Cockle site			Low site		
	C	1mm	5mm	C	1mm	5mm
Clay (0-3.9 μ m)	3.7 \pm 0.8	3.9 \pm 1.0	5.4 \pm 0.7	2.2 \pm 0.8	1.2 \pm 0.3	4.8 \pm 1.0
Silt (3.9-63 μ m)	0.4 \pm 0.2	0.5 \pm 0.1	0.7 \pm 0.1	0.3 \pm 0.1	0.2 \pm 0.1	0.9 \pm 0.4
Fine sand (63-250 μ m)	94.8 \pm 1.2	94.6 \pm 0.8	86.1 \pm 5.9	86.7 \pm 1.0	85.9 \pm 1.9	81.9 \pm 1.1
Medium sand (250-500 μ m)	0.2 \pm 0.1	0.2 \pm 0.0	0.4 \pm 0.1	7.8 \pm 0.8	8.2 \pm 1.1	8.0 \pm 0.5
Coarse sand (500-2000 μ m)	0.1 \pm 0.1	0.1 \pm 0.0	0.2 \pm 0.0	0.7 \pm 0.1	0.5 \pm 0.2	0.8 \pm 0.1
Gravel (>2000 μ m)	0.7 \pm 0.5	0.7 \pm 0.3	7.1 \pm 6.1	2.2 \pm 0.7	3.9 \pm 2.1	3.5 \pm 0.8
		Mud site			Diatom site	
		%			%	
Clay (0-3.9 μ m)	23.0 \pm 1.6	23.2 \pm 2.3	22.8 \pm 2.4	1.2 \pm 0.6	1.3 \pm 0.3	3.0 \pm 1.0
Silt (3.9-63 μ m)	0.8 \pm 0.2	0.6 \pm 0.1	0.8 \pm 0.2	0.2 \pm 0.1	0.2 \pm 0.1	0.49 \pm 0.2
Fine sand (63-250 μ m)	75.9 \pm 1.6	75.8 \pm 2.4	76.2 \pm 2.7	69.1 \pm 0.9	96.2 \pm 0.6	92.2 \pm 1.9
Medium sand (250-500 μ m)	0.2 \pm 0.1	0.3 \pm 0.1	0.2 \pm 0.1	1.9 \pm 0.3	2.1 \pm 0.3	2.3 \pm 0.2
Coarse sand (500-2000 μ m)	0.0 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0	0.2 \pm 0.1	0.1 \pm 0.0	0.2 \pm 0.1
Gravel (>2000 μ m)	0.0 \pm 0.0	0.0 \pm 0.0	0.0 \pm 0.0	0.3 \pm 0.3	0.2 \pm 0.1	1.8 \pm 0.9

Differences between sites were also reflected in organic and chlorophyll *a* content of the sediment. The amount of organic matter in the sediment was generally very low at all sites (Table 5). Organic content was highest at the Mud site. Chlorophyll *a* content of the sediment was highest at the Low site, and lowest at the Diatom site (Table 5). It is worth noting that the Diatom site was initially selected as a habitat

distinguished by its high chlorophyll *a* content. From the results of this part of the study, it appears that the high level of chlorophyll *a*, which we previously detected at this site, was a temporary phenomenon, and most likely related to a spring microalgae bloom. Neither organic content nor chlorophyll *a* exhibited a significant trend with clay depth.

Table 5: Sediment cohesiveness and biogeochemical characteristics (Mean \pm SE) of the surficial sediment at the end (Day 9) of the experiment. C = control; 1, 3, 5, 7 mm experimental treatment (depth of clay layer).

	Cockle site			Low site		
	C	1mm	5mm	C	1mm	5mm
Shear strength (kg cm ⁻²)	0.1 \pm 0.0	0.1 \pm 0.0	0.1 \pm 0.0	0.1 \pm 0.02	0.1 \pm 0.0	0.1 \pm 0.0
Penetrometer (kg cm ⁻²)	2.7 \pm 0.4	3.1 \pm 0.3	2.7 \pm 0.5	3.0 \pm 0.1	3.0 \pm 0.4	2.5 \pm 0.4
Organic content (%)	0.5 \pm 0.0	0.4 \pm 0.0	0.4 \pm 0.1	0.5 \pm 0.1	0.5 \pm 0.2	0.7 \pm 0.0
Chlorophyll <i>a</i> (μ g cm ⁻²)	60.5 \pm 4.2	56.6 \pm 4.3	64.1 \pm 6.0	123.3 \pm 3.8	124.7 \pm 3.9	114.1 \pm 3.0
	Mud site			Diatom site		
	C	1mm	5mm	C	1mm	5mm
Shear strength (kg cm ⁻²)	0.08 \pm 0.01	0.1 \pm 0.0	0.1 \pm 0.0	0.1 \pm 0.0	0.1 \pm 0.0	0.0 \pm 0.0
Penetrometer (kg cm ⁻²)	1.52 \pm 0.13	1.5 \pm 0.1	1.3 \pm 0.1	1.8 \pm 0.2	1.9 \pm 0.1	0.5 \pm 0.1
Organic content (%)	1.6 \pm 0.0	1.7 \pm 0.1	1.7 \pm 0.3	0.5 \pm 0.1	0.4 \pm 0.0	0.4 \pm 0.1
Chlorophyll <i>a</i> (μ g cm ⁻²)	73.2 \pm 5.7	70.0 \pm 1.7	64.0 \pm 2.8	49.8 \pm 2.7	41.5 \pm 1.7	46.2 \pm 2.3

Measurements of sediment cohesiveness showed some variability in penetrometer readings, but similar shear strength was noted at all sites (Table 5). Although generally low across all habitats, penetrability was approximately twice as high at the Cockle and Low sites compared to the other sites. Apart from the Diatom site, where penetrability decreased considerably with increasing clay depth, there were no clear trends with treatment level.

5.2.2 Responses of the macrofauna community

Across the different habitats, the macrofauna community showed an obvious response to thin deposits of clay over the course of the study. Statistical analysis, which accounted for treatment effects within and across sites, revealed that there was a decline in the number of taxa and individuals with increasing clay depth. Even though values for the number of taxa differed between sites (Table 6), this community measure declined with increasing clay depth ($P = 0.0074$), and this effect was consistent across all habitats. The decrease in the number of individuals, on the other hand, varied within habitats, and was significant at the Cockle, Low and Diatom sites ($P = 0.047$, $P = 0.042$ and $P = 0.03$, respectively).

Table 6: Rank order of the five most abundant taxa found in the control plots at all four sites at the end of the experiment. Means and standard errors are presented for each taxa, based on density per core.

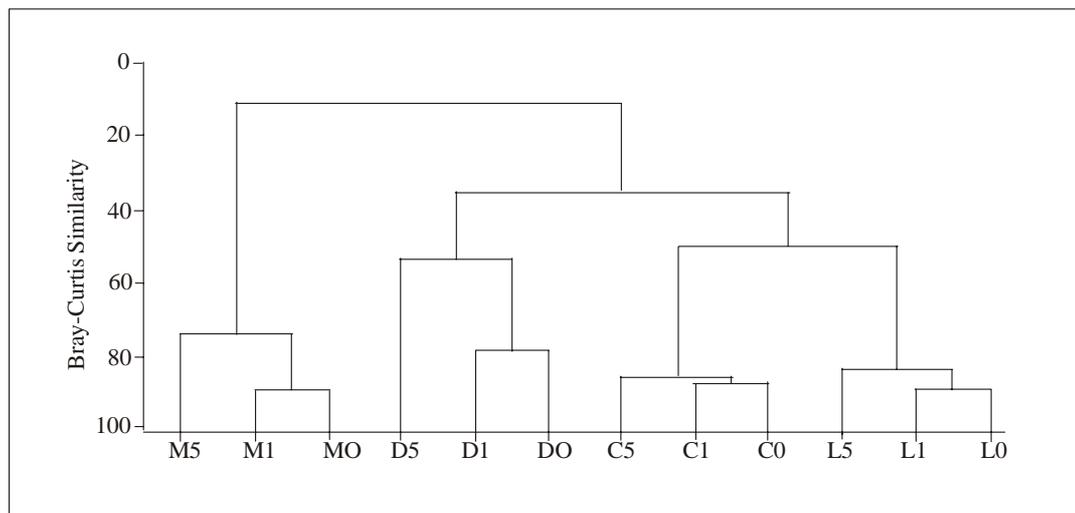
		Mean \pm SE	
Cockle site	<i>Nucula hartvigiana</i>	16.9 \pm 2.4	
	<i>Austrovenus stutchburyi</i>	14.0 \pm 1.5	
	<i>Aquilaspio aucklandica</i>	6.2 \pm 3.0	
	<i>Macomona liliana</i>	6.2 \pm 0.3	
	<i>Anthopleura aureoradiata</i>	3.9 \pm 0.6	
	Number of taxa	15.1 \pm 0.6	
	Number of individuals	57.0 \pm 2.3	
Low site	<i>Nucula hartvigiana</i>	25.9 \pm 4.8	
	<i>Chamaesipho columna</i>	25.0 \pm 0.4	
	<i>Notoacmea helmsi</i>	18.7 \pm 4.0	
	<i>Anthopleura aureoradiata</i>	17.6 \pm 2.4	
	<i>Austrovenus stutchburyi</i>	15.0 \pm 1.6	
	Number of taxa	20.5 \pm 0.8	
	Number of individuals	144.6 \pm 10.6	
Diatom site	<i>Macomona liliana</i>	8.1 \pm 1.1	
	<i>Austrovenus stutchburyi</i>	6.0 \pm 1.1	
	<i>Aonides oxycephala</i>	4.5 \pm 1.7	
	<i>Anthopleura aureoradiata</i>	3.2 \pm 0.6	
	<i>Colurostylis lemurum</i>	2.1 \pm 0.5	
	Number of taxa	13.6 \pm 1.0	
	Number of individuals	36.1 \pm 4.4	
Mud site	Orbinid	7.7 \pm 1.6	
	<i>Aquilaspio aucklandica</i>	3.0 \pm 0.9	
	<i>Arthritica bifurca</i>	1.5 \pm 1.3	
	<i>Heteromastus filiformis</i>	1.2 \pm 0.4	
	Number of taxa	7.4 \pm 0.4	
	Number of individuals	15.9 \pm 1.9	

The bivalve *Austrovenus stutchburyi* was the only species common at all sites and its abundance declined significantly and consistently across all sites ($P = 0.0441$).

Within each habitat, some of the dominant species exhibited a significant decrease in abundance with increasing clay depth. At the Low site, the grazing limpet *Notoacmea helmsi* declined significantly ($P = 0.0069$). The abundance of the spionid polychaete *Aonides oxycephala* and the sea cucumber *Trochodota dendyi* at the Diatom site declined with an increase in clay depth ($P = 0.0381$ and $P = 0.0462$, respectively). At the Mud site, the overall abundance of individual taxa was generally lower than at the other sites. Here, the small bivalve *Arthritica bifurca* decreased with increasing clay depth ($P = 0.0259$).

Cluster analysis of community data from all intertidal sites revealed the magnitude of change in community composition between the different treatment levels in the different habitats (Figure 8). In particular, the community composition in 5 mm treatments was distinctly different to that in control and 1 mm plots. This pattern in community structure was consistent across all habitats, but varied in magnitude between sites: At the Cockle and Low sites the difference in community composition was small (< 5%), but it increased at the Mud (~15%) and the Diatom site (~25%). Therefore, even though community structure differed significantly between habitats (ANOSIM, all $P < 0.001$) each macrofauna community responded similarly to the deposition of clay. This shows that thin deposits of clay have an impact on macrofauna communities across different habitats. Community measures and abundance of individual species decreased and the overall community composition shifted distinctly in relation to the deposition of clay. While this response was unilateral across different sites, the magnitude of effects depended on the type of the community and habitat.

Figure 8. Cluster analysis of macrofauna data from all sites (M = Mud, D = Diatom, C = Cockle, L = Low site) showing differences in community composition in relation to clay depth (0, 1 and 5mm).



5.3 Subtidal habitat

Apart from differences in tidal regime and physical characteristics, the Subtidal site was markedly different from the intertidal habitats. Here, the sediment had a high clay content (Gibbs et al. 2001), and appeared to be very mobile.

The chlorophyll *a* content of the sediment showed no significant change with clay depth and ranged from 18.8 $\mu\text{g cm}^{-2}$ to 18.5 $\mu\text{g cm}^{-2}$ between control and treatment plots. Similarly, sediment organic content varied little, ranging from 1.3% in controls to 1.1% in 5 mm treatments.

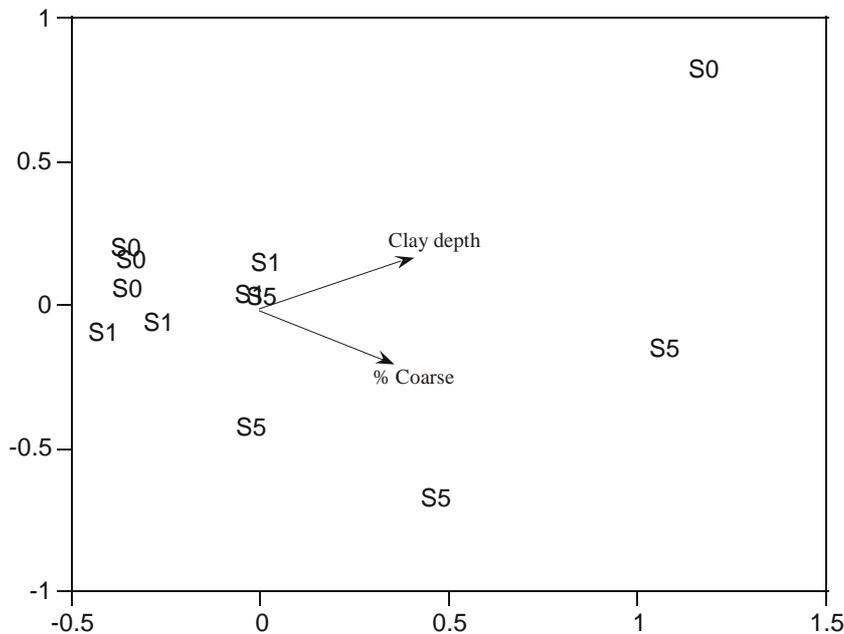
The macrofauna at this site was indicative of an environment with predominantly fine sediment (Table 7). All of the common species found here are also common in the muddy subtidal sediments of Mahurangi estuary (Cummings et al. 2001).

Table 7: Rank order of the five most abundant taxa found in the control plots at the Subtidal site at the end of the experiment. Means and standard errors are presented for each taxa, based on density per core.

		Mean \pm SE
Subtidal site	<i>Theora lubrica</i>	6.0 \pm 1.4
	Ostracoda	5.8 \pm 2.2
	Phoxocephalidae	3.2 \pm 0.9
	Tanaidae	3.1 \pm 0.5
	<i>Cossura</i> sp.	2.2 \pm 0.2
	Number of taxa	12.2 \pm 0.9
	Number of individuals	28.7 \pm 5.0

Due to wave action and reworking by crabs, the clay layers in the subtidal plots were quickly broken up into smaller patches and buried. As a consequence, experimental treatments only had little effect. Only one of the common species, the invasive bivalve, *Theora lubrica*, showed higher densities in experimental plots than in the controls. Multivariate analysis showed that sites were oriented along axis 1 relative to treatment levels, but one aberrant control plot precluded a uniform trend in macrofauna community change in relation to clay depth (Figure 9).

Figure 9: Ordination plot of macrofauna community data at the Subtidal site (S) for individual plots at each treatment level (0 to 5 mm). The arrows indicate how the community changes in relation to environmental variables as detected by canonical correspondence analysis.

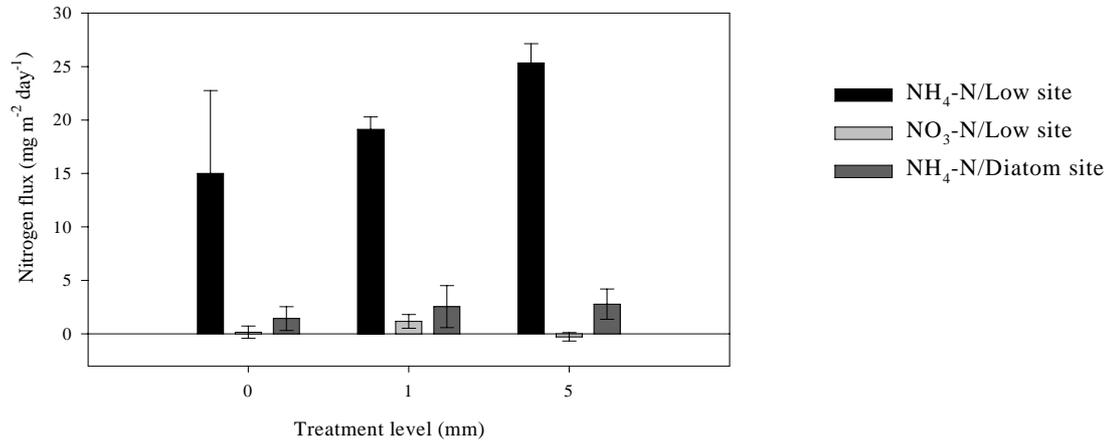


5.4 Assessing nutrient flux across the sediment-water interface

$\text{NH}_4\text{-N}$ is released into the sediment by the decomposition of organic matter and excretion by sediment fauna. Two bacterial processes are important in transforming $\text{NH}_4\text{-N}$ and thus are directly relevant to the nutrient flux across the sediment-water interface. Nitrification occurs under aerobic conditions, wherein bacteria transform $\text{NH}_4\text{-N}$ to nitrate ($\text{NO}_3\text{-N}$). In contrast, denitrification is an anaerobic process, in which $\text{NO}_3\text{-N}$ gets converted to N_2 gas. As the level of oxygen in the environment regulates both processes, they are closely linked to photosynthesis and oxygen production by benthic algae (microphytobenthos) in the sediment and sediment permeability. Microphytes require $\text{NH}_4\text{-N}$ to grow; its uptake and the resulting increase in growth enhance photosynthesis rates and therefore lead to elevated levels of oxygen in the sediment. Due to very low $\text{NH}_4\text{-N}$ concentrations in the overlying water, excess $\text{NH}_4\text{-N}$ diffuses from the sediment into the water column.

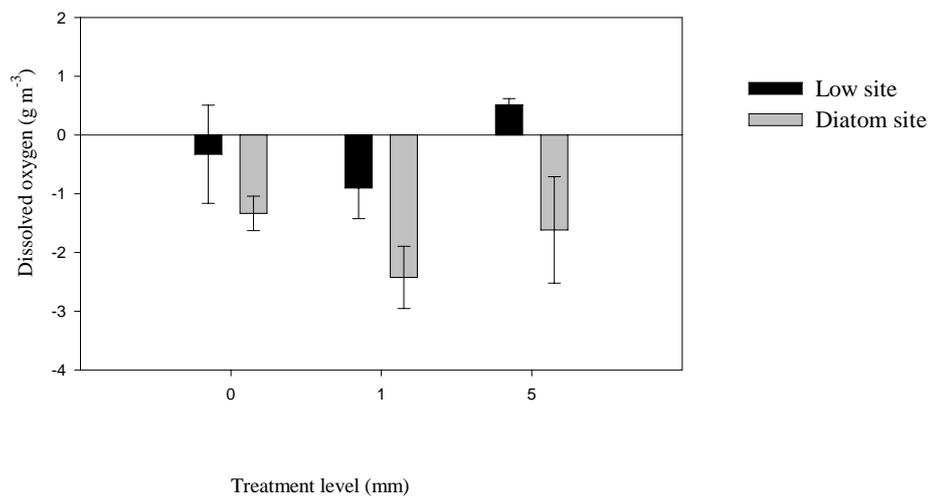
The deposition of thin clay layers on the sediment surface impacted on this diffusion process. In particular at the Low site, release rates of $\text{NH}_4\text{-N}$ increased with clay depth (Figure 10).

Figure 10: Sediment nitrogen regeneration flux rates (Mean \pm SE) in the chambers showing increasing release rates of NH₄-N with clay depth at both sites. The difference in flux rate between sites reflects the difference in sediment substrate.



NH₄-N release rates at the Low site increased by 27 and 69% respectively in the 1 mm and 5 mm treatments relative to the controls. This is indicative of a decrease in NH₄-N uptake by microphytes of 60-100% in the treatments. The reduction in oxygen production and microphyte growth was reflected in reduced NO₃-N release rates at the Low site. Although some nitrification occurred in the 1 mm treatments, negative release rates for NO₃-N clearly showed denitrification in 5 mm treatments. Dissolved oxygen measurements supported these findings (Fig. 11). The loss of dissolved oxygen from the 5 mm treatments showed that oxygen consumption exceeded its production, indicating the lack of photosynthesis by the microphytes at the Low site (Figure 11). Although the same processes occurred at the Diatom site, the sandier substrate was more easily flushed by the tide, resulting in much lower sediment NH₄-N release rates. Mixing of the clay into these coarser grained sands also allowed the microphytes to migrate into the photic zone and continue to photosynthesise and produce oxygen at all treatments at the Diatom site (Fig. 11).

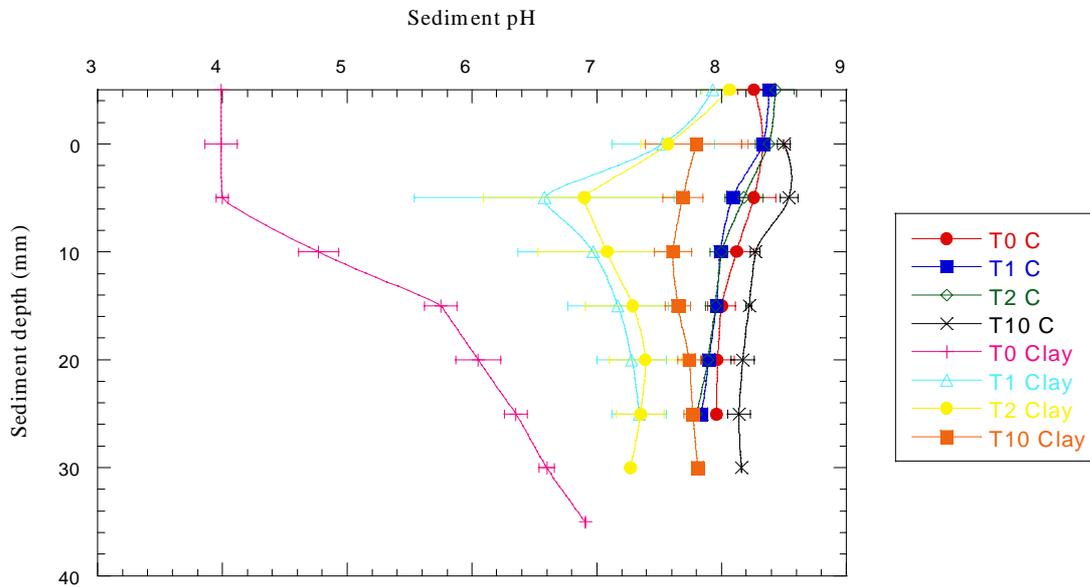
Figure 11: Changes in dissolved oxygen (DO) concentrations (Mean \pm SE) in the chambers showing oxygen production in the 0 and 1 mm treatments, and consumption in the 5 mm treatments at the Low site. Oxygen was produced at all treatment levels at the Diatom site.



5.5 pH experiment

The pH experiment showed that clay deposits of 3 mm depth might initially acidify the underlying sediment (Figure 12). Immediate differences in pH-values between the control area and the experimental plot ranged from ~ 4 pH-units in the top 5 mm of the sediment to ~ 1 pH-unit at a sediment depth of 30 mm. One to two tidal cycles after the deposition of clay, pH-values returned to a characteristic level of pH 7-8 of the sediment. It is worth noting, however, that the pH profiles in the experimental plot retained a slightly more acidic signature relative to controls for at least 10 days. Given the nature and mobility of the organisms living in the sediment, the potential to adversely affect animals with the rapid deposition of terrigenous clay will be restricted to a few surface dwellers that are unable to burrow into the sediment.

Figure 12: Sediment pH-profiles of the 3 mm treatment (Clay) and adjacent control area (C) at the Diatom site. Measurements (Mean \pm SE) were taken immediately after the application of clay (T = 0) and 1, 2 and 10 tidal cycles later (T=1, 2, and 10, respectively).



5.6 Field experiment 3. (How does frequent deposition of thin layers of terrigenous clay affect benthic communities?)

5.6.1 Physical and biogeochemical sediment characteristics

Physical changes associated with frequency deposition of thin layers of clay showed some potential for long-term change in the physical characteristics of these sandflat habitats. For example, while the depth of the clay layer decreased over the interval between applications, clay was being worked into the sediment matrix at both sites (Photo 7) increasing the overall sediment clay content in treatments relative to controls (Table 8). There are also indications of changes in sediment cohesive properties (penetration and shear stress), but long-term changes in sediment organic matter or chlorophyll *a* content were not observed.

Photo 7. Experiment 3. Cockle site – 10 days after clay deposition



a) Control plot - November



b) 3 mm clay plot



c) Control plot - April



d) 3 mm clay plot

Experiment 3. Photos of the experimental plots prior to sampling in November 2000 and April 2001. Terrigenous clay, although not forming a layer smothering the underlying sandflat, is still visible in the 3 mm plots. Photo quadrat (0.25m²), note orientation of individual photos is not necessarily consistent.

Experiment 3. Worm site – 10 days after clay deposition



a) Control plot - November



b) 3 mm clay plot



c) Control plot - April



d) 3 mm clay plot

Experiment 3. Photos of the experimental plots prior to sampling in November 2000 and April 2001. Terrigenous clay, although not forming a layer smothering the underlying sandflat, is still visible in the 3 mm plots. Photo quadrat (0.25m²), note orientation of individual photos is not necessarily consistent.

Table 8: Physical characteristics (Mean \pm SE) measured in the 3 mm and control plots in November 2000, and in plots subjected to monthly additions of clay from November 2000 to April 2001. *P*-values are the result of paired t-tests to assess the significance of differences between treatment and control plots.

Cockle Site						
	November 2000			April 2001		
	3 mm Plot Mean \pm SE	Control Plot Mean \pm SE	<i>P</i>	3 mm Plot Mean \pm SE	Control Plot Mean \pm SE	<i>P</i>
Clay depth (mm)	2.4 \pm 1.0	0.0 \pm 0.0	0.001	0.5 \pm 0.3	0.0 \pm 0	.016
Chlorophyll <i>a</i> ($\mu\text{g cm}^{-2}$)	53.4 \pm 4.6	52.3 \pm 3.8	0.862	229.1 \pm 10.5	202.8 \pm 18.8	.267
Clay content (%)	0.4 \pm 0.1	0.3 \pm 0.1	0.432	0.8 \pm 0.0	0.4 \pm 0.0	.002
Organic content (%)	1.0 \pm 0.1	1.1 \pm 0.1	0.288	0.9 \pm 0.1	1.1 \pm 0.3	.550
Shear strength (kg cm^{-2})	0.4 \pm 0.0	0.5 \pm 0.0	0.055	0.8 \pm 0.03	0.8 \pm 0.0	.239

Worm Site						
	November 2000			April 2001		
	3 mm Plot Mean \pm SE	Control Plot Mean \pm SE	<i>P</i>	3 mm Plot Mean \pm SE	Control Plot Mean \pm SE	<i>P</i>
Clay depth (mm)	3.0 \pm 0.0	0.0 \pm 0	0.002	3.1 \pm 0.0	0 \pm 0	0.016
Chlorophyll <i>a</i> ($\mu\text{g cm}^{-2}$)	45.3 \pm 3.1	41.4 \pm 3.7	0.444	158.6 \pm 8.2	163.2 \pm 17.6	0.821
Clay content (%)	0.9 \pm 0.2	0.4 \pm 0.1	0.065	1.0 \pm 0.2	0.4 \pm 0.1	0.039
Organic content (%)	0.8 \pm 0.0	0.6 \pm 0.0	0.001	1.0 \pm 0.0	0.8 \pm 0.0	0.008
Penetrometer (kg cm^{-2})	3.0 \pm 0.6	4.7 \pm 0.2	0.045	3.3 \pm 0.3	0.8 \pm 0.3	0.314
Shear strength (kg cm^{-2})	0.5 \pm 0.0	0.6 \pm 0.0	0.003	0.9 \pm 0.0	1.4 \pm 0.1	0.001

5.6.2 Responses of the macrofauna community

The species that were most common at the two sites during the first experiment were generally common at the end of the experiment (Table 9). Some slight changes in rank order occurred as a result of seasonal recruitment over the duration of the experiment. Comparing the control and treatment plots at the end of the experiment did not reveal major shifts in the rank order of dominant species. However, there are substantially different densities for the common species in controls and treatments.

Table 9: Rank order of the five most abundant taxa found in the control plots in November 2000, and the control and treatment plots in April 2001. Means and standard errors are presented for each taxa, based on density per core.

Cockle Site	Control plots, November	Mean \pm SE	Control plots, April 2001	Mean \pm SE	Experimental plots, April	Mean \pm SE
	<i>Aquilaspio aucklandica</i>	25.1 \pm 5.0	<i>Austrovenus stutchburyi</i>	17 \pm 3.6	<i>Aquilaspio aucklandica</i>	14.6 \pm 2.5
	<i>Nucula hartvigiana</i>	20.1 \pm 2.8	<i>Aquilaspio aucklandica</i>	16.2 \pm 4.2	<i>Nucula hartvigiana</i>	11.6 \pm 2.6
	<i>Austrovenus stutchburyi</i>	16.9 \pm 4.0	<i>Nucula hartvigiana</i>	15.5 \pm 4.1	<i>Austrovenus stutchburyi</i>	11.0 \pm 2.9
	Orbinids	14.7 \pm 2.4	<i>Macomona liliana</i>	8.6 \pm 1.5	Nereid	7.8 \pm 2.7
	<i>Chamaesipho columna</i>	12.2 \pm 0.4	<i>Chamaesipho columna</i>	7.8 \pm 1.1	<i>Chamaesipho columna</i> / <i>Anthopleura aureoradiata</i>	6.2 \pm 1.4/6.2 \pm 1.1
	Number of taxa	27.0 \pm 2.4	Number of taxa	11.5 \pm 0.5	Number of taxa	8.6 \pm 0.3
	Number of individuals	161.4 \pm 30.3	Number of individuals	108.5 \pm 16.5	Number of individuals	80.5 \pm 8.9
Worm Site	<i>Aonides oxycephala</i>	18.1 \pm 2.9	<i>Aonides oxycephala</i>	5.6 \pm 0.7	<i>Aonides oxycephala</i>	19.0 \pm 4.2
	Orbinids	8.7 \pm 1.7	<i>Macomona liliana</i>	10.4 \pm 1.6	<i>Macomona liliana</i>	6.8 \pm 3.0
	<i>Macomona liliana</i>	9.6 \pm 1.7	Nereid	2.1 \pm 1.0	Nereid	5.2 \pm 1.8
	<i>Paracalliope</i>	2.2 \pm 0.5	<i>Paracalliope</i>	1.9 \pm 1.3	Orbinids	2.7 \pm 0.7
	<i>Aquilaspio aucklandica</i>	2.4 \pm 0.5	<i>Aquilaspio aucklandica</i>	1.2 \pm 0.3	<i>Heteromastus filiformis</i>	2.7 \pm 2.4
	Number of taxa	16.0 \pm 1.3		8.0 \pm 0.7		8.3 \pm 0.6
	Number of individuals	48.8 \pm 3.2		46.0 \pm 1.0		32.3 \pm 5.5

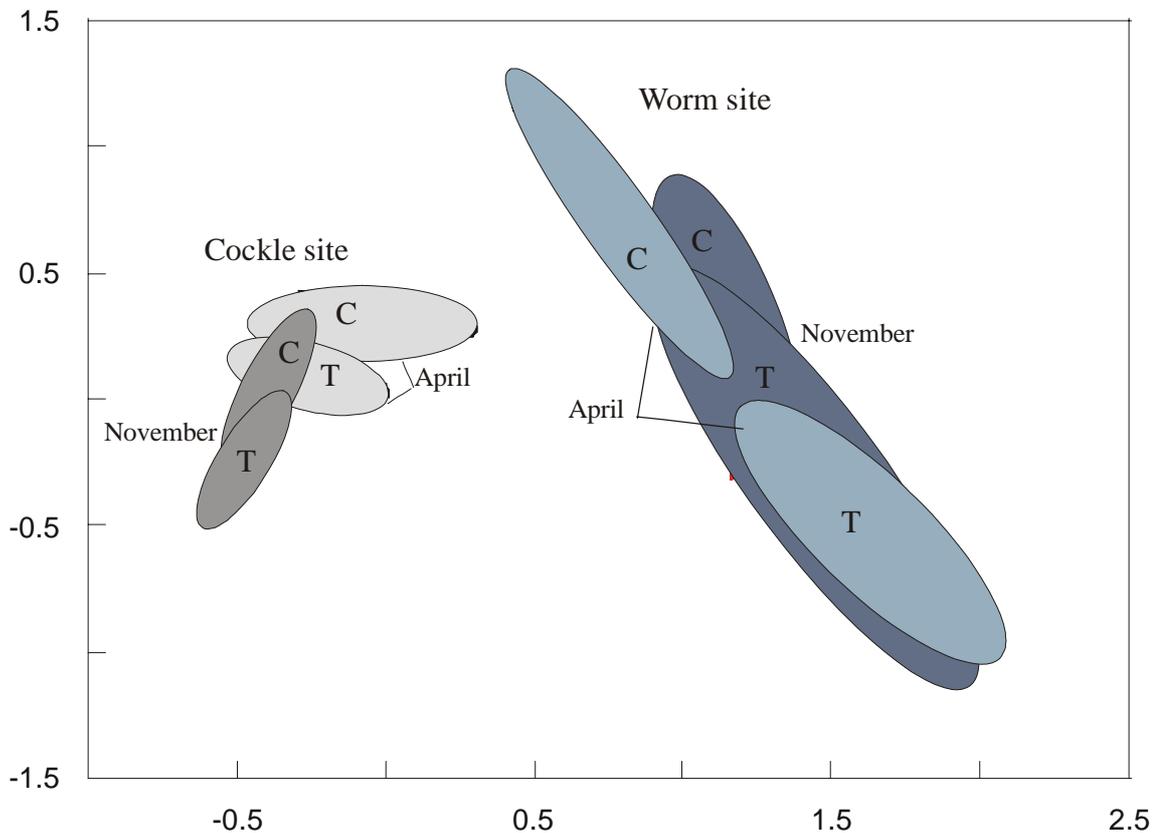
We used paired t-tests, or if appropriate non-parametric equivalents, to assess the significance of changes in the density of common taxa and number of individuals and number of taxa (Table 10). These tests did not reveal a clear pattern of increase in the strength of the effect over the duration of the experiment. This is almost certainly because the abundance of common species has changed over the experimental sites between November 2000 and April 2001. The low number of significant effects is also a product of the low level of replication for these experiments (n = 4). Nevertheless, the response of the macrofauna community to clay deposits was uniform, and only one variable that demonstrated a significant effect showed a response of increased abundance in the clay treatments. The higher density of *Macomona* in the treatments at the Cackle site in November 2000 was driven by higher numbers of large (>15 mm) individuals.

Table 10: The significance of differences between treatment and control plots at the Cackle and Worm sites in November 2000 and April 2001. *P*-values resulting from paired t-tests or Wilcoxon-sign rank tests on dominant taxa, number of individuals and number of taxa.

	Cackle Site		Worm Site	
	November 2000	April 2001	November 2000	April 2001
<i>Austrovenus stutchburyi</i>	0.593	0.224	-	-
<i>Nucula hartvigiana</i>	0.703	0.472	-	-
<i>Macomona liliana</i>	0.189	0.0328	0.384	0.0036
<i>Aquilaspio aucklandica</i>	0.283	0.750	0.055	0.052
Orbinids	0.631	0.810	0.048	0.537
<i>Aonides oxycephala</i>	-	-	0.511	0.173
Nereid	1.0	0.361	0.937	0.484
<i>Heteromastus filiformis</i>	-	-	0.068	0.380
<i>Anthopleura aureoradiata</i>	0.940	0.443		
<i>Paracalliope</i>	-	-	0.631	0.323
<i>Chamaesipho columna</i>	0.731	0.404		
Number of individuals	0.528	0.262	0.009	0.085
Number of taxa	0.409	0.016	0.067	0.049

Similar to the results in Field experiment 1, ordination of the benthic community data clearly separates the two experimental sites (Figure 13). In this graph we have identified the area of the ordination space occupied by data from the controls and treatments at the end of Field experiment 1 (November 2000) and 5 months later in April 2001, after the plots had been subjected to monthly applications of 3 mm layers of clay.

Figure 13: Ordination plot of macrofauna community composition at the Cockle and Worm sites at the beginning (November) and end (April) of Field experiment 3. The differences in community composition increased with repeated exposure to clay at both sites. C = Control, T = Treatment.



At the Cockle site, there is some overlap between the control and experimental data and no evidence of a significant difference in community composition ($P = 0.57$). Although the difference between treatment and control plots was not significantly different in April 2001 ($P = 0.29$) the degree of overlap has decreased. This provides some evidence, albeit weak, that repeated application of clay tends to increase the difference in community structure over time. More clear-cut results are apparent in the data from the Worm site, with no significant difference between samples collected from the clay deposition and control plots in November 2000 ($P = 0.28$), but complete separation of these plots by April ($P = 0.014$). These patterns are confirmed by comparing the Bray-Curtis similarities (a measure of similarity in community composition) in the treatment and control plots at the two sites over time. For the Cockle site, the treatment and control plots had a similarity of 71% in November 2000, which decreased by 24 - 62% by April 2001. For the Worm site, the treatment and control plots had a similarity of 58% in November 2000, which decreased by 36 - 40% by April 2001. These results indicate that the benthic community did not have time to completely recover over the monthly intervals between the clay applications, and that frequent additions of only 3 mm layers of clay are having a cumulative effect on the nature of the macrobenthic community. However, the magnitude of the effect was very dependent on the resident community, as illustrated by the differences in the strength of effect in the two sites used in this experiment.

6 Discussion: implications and conclusions

Through a series of field experiments we investigated the ecological effects of the deposition of thin layers of terrigenous clay on macrofaunal community structure and ecosystem dynamics. Thin layers of clay did not produce the catastrophic and long-term response that we observed as a result of thick (> 2 cm) clay deposits in earlier experiments in Okura estuary (Norkko et al. 1999). Studies in the Whitford embayment show that the macrobenthic community and biogeochemical variables responded to the addition of < 1 cm layers of clay, and that the clay layers were only detectable for up to 1 month (pers. obs.) In this section of the report, we briefly discuss the general patterns of response apparent in the three experimental studies, before drawing some conclusions as to the implications of these types of sedimentation events to the ecology of the embayment. It is important to note that the full implications of these findings will only be able to be determined when relevant information on the hydrodynamics and catchment modelling is available.

6.1 General observation on clay layers and biogeochemical parameters

We noted that the clay material, even in layers 1 - 3 mm deep, adhered to the seafloor and eroded away slowly. In Field experiment 1, we recorded about a 50% reduction in clay depth for clay layers 3 to 7 mm thick over the 10 days of the experiment. Animals burrowing up through these thin layers of clay, and moving over the sediment surface were important in facilitating the removal of clay from the experimental plots. On the intertidal flats a very important process that influences changes in the experimental plots was the bedload transport of sand over the deposited clay layer. This process helps to incorporate the clay into the underlying sediment matrix as seen by the increased fraction of clay size particles. Similar patterns across all the intertidal sites were also apparent in Field experiment 2, with the 1 mm clay layers changing the most over time due to erosion and reworking. The movement of surficial sediment by crabs and wave action were particularly important in mobilising the clay deposits at the Subtidal site. Irrespective of clay depth, these physical and biological processes rapidly reworked the deposited clay.

The resident macrobenthic community and the local wave climate will influence the potential for long-term changes in the muddiness of sediments. However, results from Field experiment 3 indicate that the frequency of clay deposition is also an important factor in determining changes to the physical characteristics of the sandflat habitats.

Of the biogeochemical sediment properties routinely measured throughout the experiments, sediment chlorophyll *a* showed a consistent response of decreasing concentration with increasing clay depth in Field experiment 1. However, this response was not repeated in Field experiments 2 and 3. Chlorophyll *a* decreases, because the clay layer restricts light penetration of the sediment surface and thus shuts down or reduces primary production. However, the time-scale of response in

standing stock (as measured by chlorophyll *a*) vs production is not known. Depending on the species of microphytobenthos smothered by clay, we may expect some variability in response in terms of how long these tiny plants can survive the absence of light. Long-term changes in sediment chlorophyll *a* concentration were not apparent in Field experiment 3, probably because microphytobenthos were transported by sediment bedload into the experimental plots and re-established in the one-month interval between clay deposition and sampling.

Changes in sediment pore-water ammoniacal nitrogen (including $\text{NH}_4\text{-N}$ and NH_3) concentrations were investigated. NH_3 (ammonia) is an important pathway for nitrogen cycling within the sediments and is the form of nitrogen preferentially taken up by the microphytobenthos. Ammonia is also highly toxic to macrofauna, particularly juvenile life-stages. Differences in the pore-water ammoniacal nitrogen concentration were apparent between the two sites used in Field experiment 1, where concentrations were about four times higher at one site (Cockle site). The Cockle site exhibited an 11% decrease of pore-water ammoniacal nitrogen concentration across the range of treatments while at the Worm site, sediment ammoniacal nitrogen decreased by 30%. However, the biogeochemical processes contributing to ammoniacal nitrogen concentration within the sediment pore water are not static, as demonstrated by the changes in concentrations in the control plots of Field experiment 1 over time. Thus we should be cautious in determining the environmental consequences of short-term shifts in isolation from the other ecological processes occurring within the sediments. Nevertheless, these processes occurring within the sediment are tightly coupled as indicated by the response to the flux chamber experiment conducted as part of Field experiment 2. Although pore-water ammoniacal nitrogen concentrations were low over the main intertidal sandflat of the Whitford embayment due to the low organic content of the sediments, the flux chamber experiment showed increased release rates of ammoniacal nitrogen from the sediments covered with clay. This is probably due to the decrease in photosynthesis by the microphytobenthos. The ammoniacal nitrogen diffuses across the sediment-water interface into the water column, and potentially has a negative effect on macrofauna attempting to recolonise the disturbed area.

6.2 Response of the macrobenthic community

In Field experiment 1 we saw clear patterns of about a 50% decrease in macrofaunal abundance and diversity with increasing clay depth. This pattern was confirmed by the multivariate analysis that revealed overall changes in community composition. This was a remarkably consistent pattern despite there being large differences in species composition, abundance and diversity between the two experimental sites. There was no evidence of complete defaunation of the sediments as we have seen in previous studies (Norkko et al. 1999, Nicholls et al. 2000).

Across the different intertidal habitats studied in Field experiment 2, the macrofauna community also showed a clear response of decreasing numbers of taxa with increasing clay depth. The decrease in the number of individuals occurred at all but the Mud site. Cluster analysis of community data from all intertidal sites studied in Field

experiment 2 demonstrated that the different sites had different macrobenthic communities that resulted in some overall variation in response to the additions of clay. We anticipated that the macrofauna community at the Mud site would show the weakest response, due to high ambient levels of clay in this habitat and the anticipated concomitant tolerance of resident taxa. However, the weakest response were detected at the Cockle and Low sites, while the Diatom site showed the strongest response (as indicated by the large variability in community composition between experimental treatments). Despite the magnitude of the effect being site-specific, all sites indicated some differences in community composition in the 5 mm experimental plots in relation to controls.

Results of Field experiment 3 indicated that repeated additions of 3 mm layers of clay over a 6-month period had a cumulative effect on the macrobenthic community. The community did not have time to completely recover over the monthly intervals between the clay applications. The magnitude of this effect was very dependent on the resident community.

The subtidal experiment resulted in less clear-cut patterns than noted for the intertidal sites. This was due to wave action and reworking by crabs, which quickly broke up the clay layers in the treatment plots. Multivariate analysis showed that sites were oriented relative to treatment levels, but one aberrant control plot precluded a uniform trend in macrofauna community change in relation to clay depth. However, we need to interpret this result carefully as it may not reflect community responses to much larger areal disturbances. Furthermore, the surficial clay apparent in the subtidal region of the Whitford embayment may well indicate that the area has already been subjected to clay depositions. The relatively non-sensitive taxa comprising the resident macrobenthic community may be a reflection of this disturbance.

A variety of polychaete worms (the spionid *Aquilaspio aucklandica* and the orbinids *Orbinia papillosa* and *Scoloplos cylindifer*) and the juvenile life-stages (i.e., individuals <5 mm long) of the common bivalves, *Macomona liliana* and *Austrovenus stutchburyi*, showed clear negative responses to clay additions. It is tempting to speculate on the mechanisms behind these patterns. With the exception of the orbinids, these taxa feed on microphytobenthos at the sediment-water interface and thin layers of terrigenous clay may adversely affect their feeding. Most of the common taxa from these experiments demonstrated trends of decreasing abundance with the addition of clay. However, the multivariate data also allowed us to identify the less common taxa that showed either an increase or decrease in abundance with clay additions (see Figure 6).

The cockle *Austrovenus stutchburyi* was found to be sensitive to additions of clay in Field experiments 1 and 2. The sensitivity of this species to elevated levels of suspended sediment was investigated in another study (Hatton et al. 2001). The small limpet *Notoacmea helmsi* (which feeds by grazing on diatom film found on hard surfaces such as cockle shells), also declined in abundance at the Low site, while the bivalve *Arthritica bifurca* decreased in abundance at the Mud site. The spionid polychaete *Aonides oxycephala* and the sea cucumber *Trochodota dendyi* were negatively affected by clay depth at the Diatom site. In Field experiment 3, tests did not reveal a clear pattern of increasing strength of effect over the duration of the

experiment. Contributing factors that must be taken into account when considering this result are the species-specific seasonal changes in abundance and the low power of the statistical test to detect significant differences.

In our investigations of the possible impact of the deposition of thin layers of terrigenous clay onto the surface of sediments of the Whitford embayment we asked 3 general questions:-

Question	Answer
What is the critical depth of thin (<1 cm) clay deposits that cause chronic effects?	3 mm layers can change the abundance of common taxa and macrobenthic community structure over 10 days.
How do benthic communities in different habitats respond to thin clay deposits?	Response varied with habitat type but not consistently. The nature of this is specific to the variable being tested . But all habitats are responding to additions of clay.
How does frequent deposition of thin layers of terrigenous clay affect benthic communities?	There are indications of the cumulative affect of frequent additions of clay on macrobenthic communities, cumulative effects on sediment properties such as chlorophyll <i>a</i> concentration are not apparent.

These studies have provided a lot of general information on the immediate effects of the deposition of thin layers of terrigenous clay, as well as many details and subtleties in response. Nevertheless, it is important to consider the nature of these experiments before we try and draw conclusions on the potential long-term and broad-scale effects.

7 Conclusions

Identifying subtle changes in macrofauna response to small additions of clay is challenging. We tried to focus our experiments on questions that are relevant to assessing impacts and to reaching general conclusions about how these impacts may pose a risk to the long-term integrity of macrobenthic communities in different parts of the Whitford embayment. Thus we designed our experiments to encompass different habitats, some of which were difficult to work in, and different time scales. Inevitably this constrained how much effort could be put into any single study and thus restricted the level of replication employed. Low replication reduces the statistical power of tests to identify differences between treatments. This implies that any significant trends apparent in these experiments are a result of major changes, and it is highly likely that greater replication would have detected even more effects. Thus we should be cautious in assuming that not detecting a significant change implies there really is no change. The latter point is particularly important if management decisions are to be based on the precautionary principle.

To some extent we have limited the impact of low replication on the interpretation of the results of Field experiments 1 and 2 by using gradient approaches in the design and analysis of these experiments, and the use of various multivariate analysis techniques. These approaches consider the simultaneous changes in all of the communities represented at different sites or in different treatments. Although complex, these techniques are sensitive as they reflect change in the aggregated response of the overall community and thus integrate effects across species with varying sensitivities.

Two possible problems with the experimental approach that must be considered are potential artefacts associated with clay addition procedures and the spatial scale of the experimental plots. Cation exchange processes in the clay particle matrix occur when mixing with seawater. This is of concern because of the potential to significantly lower pH. To investigate this we initiated a specific study to assess the significance of this response within the experimental context. Although the pH of the clay slurry added to the sediments was initially low, it was quickly buffered by the surrounding seawater and reached ambient conditions within 2 tidal cycles. Given the nature and mobility of the organisms living in the sediment, the potential to adversely affect animals with the rapid deposition of terrigenous clay will be restricted to a few surface dwellers that are unable to burrow into the sediment or move away from the affected areas. Moreover, the similarity in response in Field experiment 1 (using unbuffered clay slurry) and Field experiment 2 (using clay slurry buffered to ambient pH) also indicated that the pH drop associated with the experimental application of clay did not represent a significant artefact. In fact the terrigenous clay deposited on estuarine flats during "natural" sedimentation events may be delivered to the estuary by a variety of processes, not all of which result in clay particles being well mixed, and thus buffered by seawater, prior to deposition on the seafloor.

A more important consideration in terms of potential experimental artefacts is the discrepancy between the areal extent of our experimental plots (~ 4.5 m²) and the

much larger areas potentially impacted during an actual sedimentation event (100 – 1000's m²). Scaling between these two is not simply a matter of multiplying-up the experimental results and must be considered carefully. The rate of erosion of the deposited clay is likely to be independent of the area of the deposited patch. However, changes in biogeochemical processes may be considerably underestimated in small experimental plots, due to ground water transport below the clay layer. The bedload transport of ambient sediment and associated fauna and flora over the plots will also be scale dependent. These two processes are likely to reduce the magnitude of experimental effects and speed up recovery rates in our small plots. Thus we should consider these experiments as conservative in their estimate of the magnitude of effects.

A very clear result from this study, in particular Field experiment 3, is that the frequency of disturbance, and thus the time available for recovery between disturbance events, is critical in assessing whether the deposition of these thin layers of terrigenous clay pose a threat of broad-scale degradation to the Whitford embayment.

As illustrated in the Introduction, we can expect low load depositional events that produce thin layers of clay to be more frequent and to cover a larger area than larger catastrophic events. In particular, we would expect that the frequency of events would be higher in upper estuary and channel margin habitats. In the example from the Okura study presented in the Introduction, catchment modelling based on current land-use practice predicts 21, 2-3 mm depositional events occurred on an upper estuary sandflat over the 35 year weather record. This frequency of disturbance is unlikely to detrimentally impact the ecology of that sandflat. However, for the Whitford embayment these risks can only be predicted when catchment and hydrodynamic modelling has been completed. Only when this information is available for different land-use scenarios will we be able to most appropriately assess the risk to the Whitford embayment. Field experiment 3 indicates that even 3 mm depositions at monthly intervals can result in cumulative changes to macrobenthic communities. It will be important to use the catchment and hydrodynamic modelling to develop an iterative assessment of the risk of low-load events depositing clay in different parts of the embayment. Although this task cannot be completed yet, these experiments have provided us with information that enables us to refine the "rules" for assessing the risk of ecological change as a result of sediment inputs.

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