IMPACTS OF TRAWL FISHERIES ON MARINE BENTHIC BIOGEOCHEMISTRY

PHILIP PERCIVAL

UNIVERSITY OF
NEWCASTLE

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ABSTRACT

Fishing is widely recognised as the largest anthropogenic impact on coastal marine ecosystems. Bottom trawling is both widespread and a major impacting activity with documented effects on target species, populations of non-target species, food web dynamics and habitat features. In this thesis the effects of trawling on the benthic biogeochemistry of the central west North Sea were investigated. Two areas with similar sediment properties were identified for comparative analysis of North Sea sediments from trawled and untrawled areas. A combination of field and complimentary microcosm experiments were employed to examine the impacts of commercial trawling on the assemblage of benthic macrofauna, distributions of dissolved nutrients and chromophoric dissolved organic matter (CDOM).

The infauna in trawled areas consisted of species, the size of which were on average ~36 % smaller than those from untrawled areas, whereas total mean macrofaunal abundance was ~60 % greater in trawled sediments. These size and abundance differences had a profound affect on bioturbation. Natural densities of bioturbators from untrawled sediments increased benthic fluxes relative to an abiotic situation by up to 81 % NH_4^+, 197 % PO_4^{3-}, 96 % NO_2^- and 33 % NO_3^-. Fauna from trawled sediments gave flux values that lay between untrawled faunal fluxes and fluxes from controls without fauna.

In addition to the long-term alteration in sediment biogeochemistry resulting from altered faunistic composition the direct impact of trawl gear produced an enhanced sediment efflux for NH_4^+ (475 % greater than the background flux) and NO_2^- (26 % greater than the background flux). In contrast, PO_4^{3-} influx, observed in systems without trawl impacts changed to a net efflux following trawl disturbance (-15 %). In contrast, NO_3^-, displayed a net efflux in the control systems, yet, a decrease of -1.0 % was apparent following heavy trawling. Altered flux rates persisted for > 48 hours. During the main fishing season nutrient profiles were perturbed down to ~ 4 cm in sediments from trawled sites. Nutrient concentrations in the top 4 cm of trawled sediments were characterised by a relatively homogenous surface sediment layer in which concentrations were significantly lower than concentrations in untrawled sediments.

This thesis has demonstrated that the impacts of commercial trawl fisheries on benthic biogeochemistry in the North Sea are of a significant magnitude and scale to affect local and potentially regional nutrient dynamics.

Contents:

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The Earth's surface is covered with 140 million square miles of ocean...welcome to my world!

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Chapter 1: Introduction

1.1 Biogeochemical environment: The North Sea

The North Sea ecosystem is a heavily impacted system subject to intense fishing disturbances. It has a historical record of benthic trawling dating back for centuries, yet the last century witnessed a rapid development in the efficiency, intensity and extent of bottom fishing (Frid et al. 2001). Despite its relatively small size the North Sea is highly productive, representing approximately 5% of the global catch while only representing approximately 1.5% of the surface area of the global ocean (Heessen 1988).

The North Sea has a surface area of ~ 575,300 km^2, a volume of 42,294 km^2 and a mean depth of 74 m (including the Skagerrak), representing only $1/600^{th}$ of the global ocean's area and $1/50^{th}$ of the average ocean depth (Otto et al. 1990). Largely surrounded by continental Europe, this 'coastal sea' displays transitional features between coastal and oceanic dynamics and properties. The North Sea therefore can generally be classified as a marginal sea because its circulation is governed mainly by internal dynamics and the shallow depth largely dictates its properties (Howarth et al. 1994). The areas surrounding the North Sea are important contributors of substances that can affect oceanographic conditions. From the surrounding coastlines the North Sea receives ~ 370 km^3 freshwater annually, rising to > 900 km^3 if the Baltic contribution is included, thus giving a ratio of 1:60 (ICES 1983) in the annual input of water. This freshwater discharge is concentrated in the winter from British and continental shores and during the summer from Norway (Otto et al. 1990).

High volume input areas of water enter the North Sea from the north, around Scotland, and from the south-west, through the Dover Straits. These inputs aid the formation of a cyclonic circulation. A prevalent south-west wind also helps induce this anti-clockwise circulation which creates well defined current distributions near the coastlines (Prandle et al. 1994). Consequently the total flushing time of the North Sea can reach up to 3 years (residence time) of the water entering from around Scotland (Prandle et al. 1994).

Tidal dynamics within the North Sea are the single most dominant process (Otto et al. 1990). Tidal effects are ecologically significant because they are the controlling factor for; horizontal and vertical turbulent mixing, transport and disturbance to the sea bed (Tett et al. 1994). Running north to south in the North Sea, along the central main axis, an increase in tidal strength occurs due to a general shallowing (Otto et al. 1990). These increasing tidal currents, with decreasing latitude, consequently undergo interactions with the bottom (Huntley et al. 1994). Therefore, tidal energy is dissipated in turbulent motion through bottom friction. This creates turbulence and promotes vertical mixing. As a result, southern regions of the North Sea are well mixed and high stress is carried down into the benthic boundary layer (Otto et al. 1990). Central areas display a transitional zone where complete mixing occurs, yet bottom stress is relatively low (Otto et al. 1990). In the north North Sea, where depths are generally deeper, bottom friction decreases and pockets of stratification develop (Otto et al. 1990). However, although year to year variations can have a marked effect, an underlying pattern where the majority of the North Sea is vertically mixed for the greater part of the year occurs (Otto et al. 1990). As a result, a permanent pycnocline cannot develop and, as precipitation ≠ evaporation,

salinity values are relatively stable (34 – 35), except for localised estuarine inputs (Becker et al. 1983).

1.2 Sediments

In general, the North Sea seabed consists of sediment, while rocky outcrops are generally restricted to coastal areas. The southern regions tend to be dominated by sand (~ 2.5 phi), whereas central and northern regions typically have muddy (~ 3.5 phi) sediments. Chemical and physical transformations of sediments following deposition are collectively termed diagenesis (Hallberg 1992). Organic matter in marine sediments undergoes extensive diagenesis after burial as a result of microbial oxidation of organic carbon (Berner 1980). The significance of benthic degradation of organics, is that it can provide a source of regenerated nutrients in the form of nitrates and phosphates that might otherwise be limiting for phytoplankton production (Nixon et al. 1976, Rowe et al. 1977). Klump and Martens (1983) discovered that the benthic system was crucial in controlling pelagic primary production in a coastal marine basin. Although the amount of benthic nutrient release was temperature dependant, Klump and Martens (1983) found the benthos to supply greater than 50% of the nutrient requirements of plankton. The importance of a benthic nutrient supply is inversely related to the depth of the overlying water column (Jorgensen 1983). Therefore, the controlling mechanisms on mineralization rates and release in the relatively shallow North Sea are of particular interest.

The generally recognised diagenetic reaction sequence of organic matter degradation in marine sediments is shown in Table 1.1 It must be noted however, that following the utilisation of oxygen (O_2), the boundaries between

reactions may overlap and less efficient oxidants may be used simultaneously or out of sequence to gain the greatest thermodynamic advantage.

Particulate organic matter (POM) deposited to the sediment subsequently becomes available for breakdown processes which occur in specific zones of different electron acceptors required to drive progressive microbially mediated oxidation reactions (Froelich et al. 1979). The oxidation of organic matter occurs in an energy efficient manner. That is, there is a generally recognised diagenetic pathway such that bacterial oxidation of organics into inorganic compounds uses the available oxidant that yields the greatest free energy change per mole of organic carbon oxidised (Froelich et al. 1979). During this sequence, the oxidant alters when enough of this preferred species has been transformed to drive down the redox potential low enough that microbial organisms switch to the next most efficient utilisable oxidant (Chester 2000). Diagenesis therefore follows a general sequence where successive oxidants are utilised: oxygen > nitrate ≥ manganese > iron oxides > sulphate.

Table 1.1 The generalised diagenetic sequence of bacterially mediated organic matter degradation reactions in marine sediments. Relative measurements of C : N : P are presented as variables (x, y, and z) (after Froelich et al. 1979).

1, Aerobic respiration	$(CH_2O)x(NH_3)y(H_3PO_4)z + (x+2y)O_2 \rightarrow$ $x\,CO_2 + (x+y)H_2O + yHNO_3 + zH_3PO_4$
2, Nitrate reduction	$[(CH_2O)x(NH_3)y(H_3PO_4)z] + 4xNO_3^- \rightarrow$ $x\,CO_2 + 3xH_2O + 4xHCO_3^- + 2xN_2 + 5yNH_3 + 5zH_3PO_4$
3, Manganese reduction	$[(CH_2O)x(NH_3)y(H_3PO_4)z] + 4xMnOOH + 7xCO_2 + xH_2O \rightarrow$ $8x\,HCO_3^- + 4xMn^{2+} + 7yNH_3 + 7zH_3PO_4$
4, Iron reduction	$[(CH_2O)x(NH_3)y(H_3PO_4)z] + 4xFe(OH)_3 + 7xCO_2 \rightarrow$ $8x\,HCO_3^- + 3xH_2O + 4xFe^{2+} + 2yNH_3 + 2zH_3PO_4$
5, Sulphate reduction	$[(CH_2O)x(NH_3)y(H_3PO_4)z] + xSO_4^{2-} \rightarrow$ $2xHCO_3^- + xH_2S + 2yNH_3 + 2zH_3PO_4$
6, Methane reduction	$[(CH_2O)x(NH_3)y(H_3PO_4)z] \rightarrow$ $x\,CO_2 + xCH_4 + 2yNH_3 + 2zH_3PO_4$
7, Fermentation	$[(CH_2O)x(NH_3)y(H_3PO_4)z] \rightarrow$ $x\,CH_3CH_2COOH + xCH_3COOH + 2xCH_3CH_2OH + 3xCO_2 + xH_2$ $+ 12yNH_3 + 12zH_3PO_4$

The redox status within the interstitial water of sediments is fundamental in controlling the rate of many chemical changes (Libes 1992). Aerobic and anaerobic conditions therefore have a profound effect on the distributions and depth profiles of chemical constituents and transformations within sediments (Schlesinger 1991). The balance between microbial O_2 demand for oxidation and the rate at which O_2 can be supplied to porewaters is affected by grain size, porosity, activity of benthic invertebrates, natural physical disturbances and the direct impact from trawlers (Pilskaln et al. 1998). As a result, the depth of the surface oxic layer may vary spatially and temporally. Its presence however

exerts a profound influence on the transport of solutes across the sediment-water interface (Berner 1976).

Ammonium (NH_4^+) ions can only be oxidised by O_2 and thus NH_4^+ concentrations usually increase in deeper anoxic sediments (Nedwell *et al.* 1999). The build up of strong concentration gradients cause NH_4^+ to diffuse upwards to surficial oxic sediments (Blackburn 1988). Diffusive fluxes to the water column are then dependant upon the extent to which NH_4^+ is nitrified to nitrate (NO_3^-) (Henriksen and Kemp 1988). Nitrification is the step-wise oxidation of NH_4^+ to NO_3^- with nitrite (NO_2^-) as an intermediate (Henriksen and Kemp 1988). Aerobic bacteria are responsible for carrying out nitrification (Froelich et al. 1979). Therefore, notwithstanding disturbance or bioturbation, nitrification only occurs at the sediment surface (Nedwell et al. 1983). Deeper O_2 penetration can act as a potential nitrification barrier for NH_4^+ and facilitate NO_3^- production and flux to the water column (Billen 1982). High deposition can cause high C:N ratios which favour heterotrophic bacteria, causing NH_4^+ nitrifiers to be out-competed and under such circumstances the oxic layer may be depleted and facilitate NH_4^+ export (Nixon 1981). Alternately however, the process of nitrification can be inhibited by high levels of organic matter (Nedwell et al. 1999). Immediately below the oxic layer, and around faunal burrows, nitrification is often coupled to denitrification through complex interconnected pathways that undergo a multitude of redox transformations (Nedwell et al. 1983). At the onset of anoxia NO_3^- acts as the terminal electron acceptor for organic matter oxidation (Koike and Sorensen 1988). This process is therefore limited by NO_3^- produced via nitrification and organic substrates. The principal pathways of denitrification reduce NO_3^- to NO_2^- and then to gaseous nitrous

oxide (N_20) and nitrogen (N_2) in anaerobic environments. Further fermentative reactions may also act to reduce NO_2^- to NH_4^+ (ammonification) (Henriksen and Kemp 1988).

Biologically available phosphate (PO_4^{3-}) in marine sediments is regenerated from organic matter degradation by common source reactions with ammonium (Table 1). Soluble fractions of phosphorous generally increase with depth because PO_4^{3-}, associated with oxidised Fe(III) hydroxides, is liberated under anoxic conditions (Slomp et al. 1998). However, a surficial oxic sediment horizon can act as a solubility barrier where soluble PO_4^{3-} diffusing to the sediment surface is co-precipitated with iron and maintained within the sediment for further transformations (Sundby et al. 1992). This is of consequence to pelagic ecosystem dynamics as PO_4^{3-} is readily utilised by phytoplankton and would contribute to primary production in overlying waters (Klump and Martens 1983). Only at times when there is a distinct lack of an oxic layer (< 1mm) or through physical sediment resuspension can soluble PO_4^{3-} produced in sediments be mobilised (Slomp et al. 1998).

Recent studies have highlighted another pathway through which NH_4^+ can be oxidised under anoxic conditions (Dalsgaard et al. 2003, Thamdrup and Dalsgaard 2002). Under this "Anammox" reaction, bacteria utilise NO_2^- in a one-to-one pairing with NH_4^+ to form N_2 gas, thus separating this reaction from denitrification. Figure 1.1 highlights all the major pathways by which organic matter transformations occur in marine sediments.

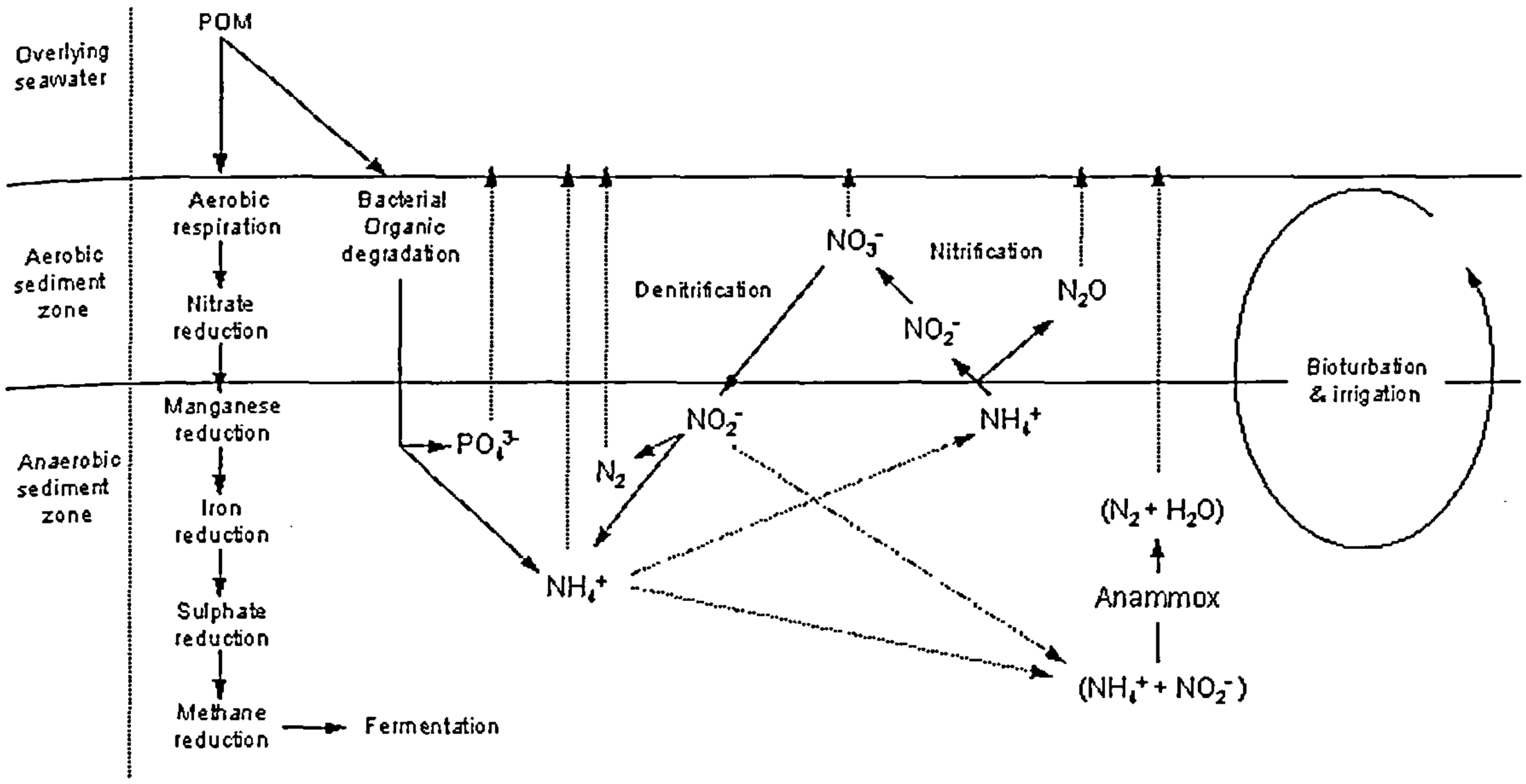

Figure 1.1. Schematic of the generalised sediment diagenetic sequence and principal chemical pathways for bacterial transformations of organic matter.

The biogeochemical processes occurring in sediments described so far generally have been idealised in terms of vertical succession processes of organic matter degradation, changes in redox status and diffusional transport. In reality the situation is more spatially and temporally complex because benthic macrofauna and physical disturbances create a vertical mosaic of chemical microenvironments that can have profound effects on sediment biogeochemistry. Faunal tubes, burrowing and sediment resuspension all affect particle and solute transport, the supply of organic-rich compounds and biogenic transformations (Kristensen 1988).

The physical structure of infaunal tubes and burrows effectively acts to increase the overall surface area across which solute exchange can take place (Aller 1988). Irrigation of those tubes increases the flux of solutes vertically through the sediment (Rhoads 1974). Consequently, the depth to which O_2 can

penetrate is greatly enhanced, while also providing a transport route to the overlying water for dissolved constituents of organic matter degradation from deep within the sediment. Burrowing activity of benthic macroinvertebrates, however, is responsible for particle reworking and redistribution of reactive organic matter, potentially promoting microbial production and hence associated biogenically mediated reactions (Aller and Yingst 1978). For example, Widdicombe and Austen (1998) found bioturbation by the heart urchin *Brissopsis lyrifera* increased surficial sediment oxygenation, resulting in decreased denitrification while increasing phosphate binding. Repeatedly, studies have shown faunal tubes and bioturbation increase O_2 penetration thereby altering nitrification – denitrification reactions which suggests that bioturbation can exert a regulatory effect on benthic nutrient cycling (Aller 1982, Hines et al. 1982, Hines and Jones 1985). Consequently, infaunal assemblages and physical impacts, that can stimulate regeneration processes may play a significant role in nutrient dynamics and the transfer of nutrients across the sediment-water interface.

Many models and theoretical treatments address the maintenance of diversity within marine communities. The following section briefly introduces some of the more established concepts. The intermediate disturbance hypothesis suggests that at low levels of disturbance, a few competitively superior species will dominate communities resulting in low levels of diversity (Connell 1978). At intermediate disturbance levels, frequent removal of these species will provide space for others, resulting in high diversity. At very high levels of disturbance only a few very tolerant or opportunistic species will occur (Connell 1978).

Physical disturbance appears to play a role in structuring communities with limited disturbance promoting biodiversity (Begon et al. 1996). The basic tenet of the competitive exclusion principle is that similar species exist in competition for available resources (Grime 1973). Species are in competition with one another for such requirements as habitat and food. Those species that create a competitive advantage will flourish at the expense of the less competitive. Competition coefficients quantify the magnitude of the competitive effect of one species on another. If two species are perfect competitors, if they have the same competition coefficients, one of the two species will become extinct. Species that coexist even in the face of competition must not be perfect competitors. Therefore species may utilise different food sources, habitats, or times of activity to avoid competition.

The dynamic equilibrium hypothesis (Huston 1979) seeks to explain how disturbance and productivity influence species richness. The model predicts that when the opposing forces of disturbance and productivity are in dynamic equilibrium, diversity will be high (Huston 1994). This model also predicts that temporal synchrony of disturbances across an otherwise homogenous area creates heterogeneity. This increases diversity by creating patches of different seral stages containing different suites of species.

1.3 Trawling

Trawl activity occurs on every major continental shelf and represents the most widespread anthropogenic impact in marine ecosystems (Jennings and Kaiser 1998, Dayton et al. 1995). Benthic trawls are designed to catch those

species that live on, or are associated with the seabed (Collie et al. 2000). Consequently, trawls are dragged through the seabed to stimulate target species into the net and maximise the catch. Within the North Sea, two major bottom trawl practices take place; otter trawling (typically in central and northern regions) and beam trawling (predominantly in the south) (Jennings et al. 2000), with otter and beam trawls representing 44 and 51 percent of total towed gears respectively (Jennings et al. 2000).

1.4 *Beam trawls*

The beam trawl, as the name suggests, consists of a rigid beam supported at each end by beam shoes (Fig 1.2). The headline of the net is attached along the beam while the sweep of the net and ground gear are fastened to the shoes (Jennings and Kaiser 1998). Consequently, the mouth of the net is continually maintained open and its size is dictated by the length of the beam and height of the shoes. The shoes are designed to act as sledges, passing over the surface of the seabed to distribute the weight and prevent the trawl sinking into the sediment (Jennings et al. 2001). Modern bean trawls can typically have beams reaching 20m in length and weighing up to 10 tonnes (Beek et al. 1990).

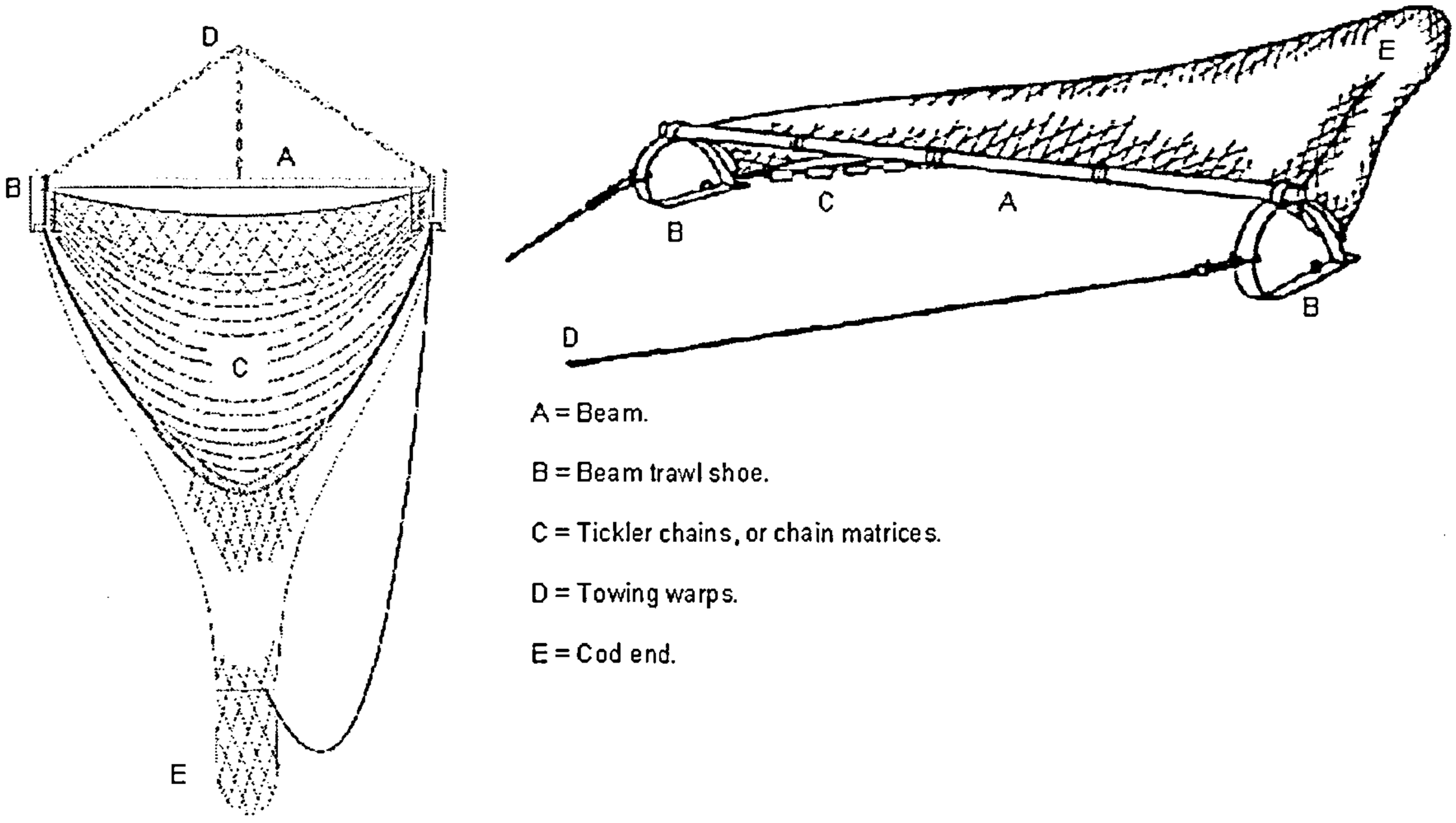

Figure 1.2. Diagram of a typical beam trawl used in North Sea fisheries. A = the trawl beam, B = the beam shoes, C = tickler chains or chain matrices, D = towing warps, and E = the net's cod end.

Because beam trawls specifically target species buried in the sediment, the ground gear of beam trawls often comprise 'tickler chains' or a chain matrix attached between the base of the shoes, designed to disturb the seabed and stimulate benthic fauna into the net (Cruetzberg et al. 1987). Dependent on the target species and sediment characteristics, different configurations of tickler chains are used. It is not uncommon for large trawlers to use up to 20 chains, each designed to scour successive layers of sediment (Jennings et al. 2001).

1.5 Otter trawls

Otter trawls are the most widespread and common type of trawl used in world fisheries (Watling and Norse 1998), and are used almost exclusively in

the study areas of the current thesis. Therefore, from hereon in the focus of the thesis concerns on the impact of otter trawls.

A typical otter trawl is shown in figure 2. Otter trawls comprise two otter boards or doors, linked by the ground gear (Jennings and Kaiser 1998). The doors maintain the net open because they are towed at an oblique angle across the sediment (Fig 1.3) (Jones 1992). Therefore, unlike beam trawls, the size of an otter trawl is not restricted by rigid structures, thus the horizontal opening of the net can exceed 200m (Ocean Studies Board 2002). The tow speed is critical for determining the aperture of the net mouth, while floats attached along the headline and weighted ground gear maintain a vertical opening in the net (Jennings et al. 2001).

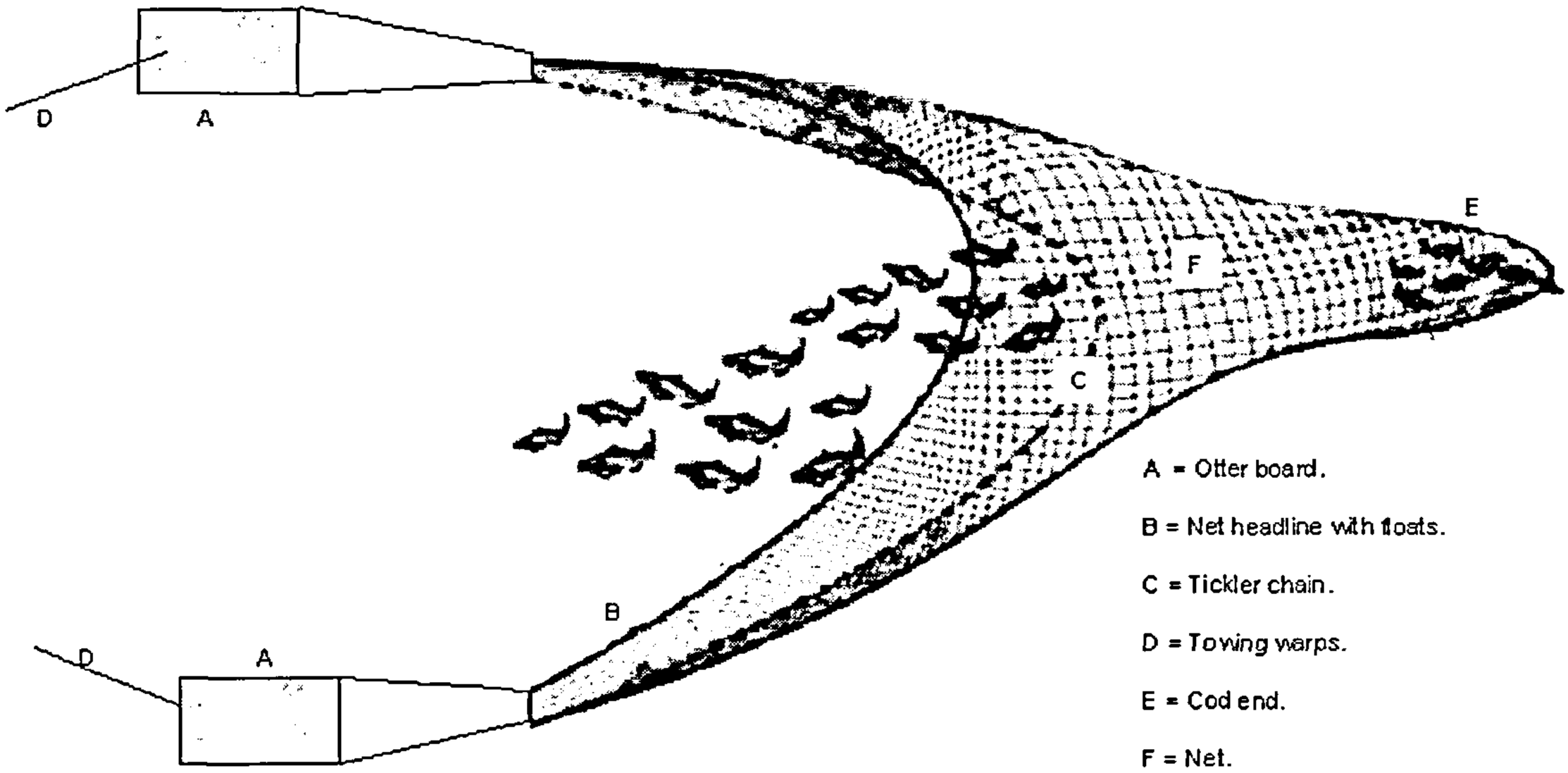

Figure 1.3. Diagram of a typical otter trawl used in North Sea fisheries. A = otter board, B = headline with floats, C = tickler chain, D = towing warps, E = cod end, and F = net; (adapted from Effects of Fishing Gear on the Seafloor of New England)

The otter doors can weigh several tonnes in air, and can penetrate up to 30 cm in muddy sediments (Jones 1992). Depending on the target species and

substrate type, ground gear may comprise only a single tickler chain with rubber bobbins. However, more commonly, otter trawls use multiple tickler chains, chain matrices, which further penetrate the sediment, or moulded rubber and steel discs (> 50 cm diameter and > 10 kg) (Jennings and Kaiser 1998) that are designed to fish in structurally complex habitats.

1.6 Bottom Trawl impact

A major unavoidable consequence of bottom trawling is impact by the trawl gear on the benthos. Each time trawl gear passes over the seabed, successive sediment layers are mobilised. The depth of penetration, and hence the magnitude of disturbance that occurs, is determined by the number of tickler chains, sediment and habitat characteristics and the relationship between the weight of the trawl and the tow speed (Fonteyne 2000). However, regardless of the depth to which mobile bottom gears dig into the seabed, even the most conservative estimates of trawl gear penetration have the potential to exert a profound and direct effect on:

- Mortality of benthic organisms from direct contact with trawl gear;
- Reduction and modification of habitat and structural complexity by altering physical habitat structures and removing large epifaunal species;
- Fish stocks, through removal of target species and alterations to recruitment;
- Resuspension and mobilisation of sediment and infaunal organisms;
- Increase in food for benthic scavengers;

- Burial of organisms by displaced sediment;
- Excavation and exposure of infaunal species.

All of the above effects are likely to be compounded if repeated trawl impacts occur over the same area of seabed. The consequential effects from these direct impacts may be less obvious, yet indirect effects may be more important in structuring benthic communities (Frid et al. 2001) and include:

- Altered benthic community composition and structure;
- Altered food web and trophic interactions;
- Diversion of energy from growth and/or reproduction for non-mortality effects such as repairing damaged tissue or rebuilding burrow structures.
- Altering sediment-water fluxes of dissolved constituents;
- Alterations to the sediment redox status (with concomitant consequences for microbial processes);
- Sediment regeneration rates from organic matter degradation.

The direct ecosystem effects of trawling have become a major focus of environmental concern (for reviews see Collie et al. 2000 and Johnson 2002). However, there have been fewer studies on the impacts of trawling on the indirect effects (for example Brylinsky et al. 1994, Jennings et al. 2002, Pilskaln et al. 1998).

1.7 North Sea Fisheries

Vessels operating in the North Sea can typically reach lengths of up to 80 metres with engines in excess of 3000 HP (Fishing News, 1997 - 1999). Within the North Sea demersal fishery, which mainly comprises beam and otter trawls, the major commercial species targeted are cod, plaice, sole, *Nephrops norvegicus*, haddock, whiting and monkfish (Anon 1998). Several countries that border the North Sea have significant stakes in North Sea fisheries including the UK, Denmark, Netherlands, Belgium, France and Germany which account for an estimated 21,300 fishers and 9,800 vessels and a combined gross landing value of £550 million in 1998 (Anon 1998, MAFF 1997). Total UK North Sea landings however, were valued at £293 million in 1998, with ~ 29% attributable to the central North Sea (Anon 1998, MAFF 1997). Logic dictates that because of such a prolonged and intense activity, North Sea fisheries must have had and continues to have a marked and wide ranging impact on the benthic ecosystem.

1.8 Gaps in our Knowledge

Much recent research has focused on the direct impact of bottom trawling on benthic organisms. This has identified mortality to infaunal organisms, reduction in habitat complexity, alterations to fish stocks and the potential release of limiting nutrients while acknowledging the indirect effects of fishing (see above). However, while the cycling of inorganic nutrients between the sediment and overlying waters in response to naturally occurring physical and biological phenomena is well studied, and for some regions is well characterised, the extent to which benthic biogeochemical cycling can be impacted by anthropogenic disturbance is much less clear. One such common

disturbance, commercial trawling, has recently been identified, but as yet it is rather poorly studied in respect of its impact on benthic biogeochemistry (but see Duplisea et al. 2001, Pilskaln et al. 1998, Percival and Frid 2000).

The broad aim of this thesis is to determine the impact of commercial trawling on benthic biogeochemistry and consequently the impact on the coupled benthic-pelagic biogeochemical system through enhanced sediment-water exchange. Specific aims of this thesis are:

1: To determine the impact of benthic trawling on macrofaunal communities from trawled and untrawled areas of the North Sea for mud and sand sediment types.

2: To investigate the effects of trawl disturbance on porewater nutrient profiles in the central west North Sea from untrawled, trawled, and immediately trawled sediments.

3: To evaluate the effects of trawling on North Sea sediment nutrient dynamics from *in situ* measurements and in laboratory based microcosm experiments.

4: To determine what effect potential changes in the composition of benthic macrofaunal assemblages may have on benthic nutrient dynamics from trawled and untrawled areas.

In order to understand the dynamics of nutrients in real fishing grounds of the central west North Sea, the impact on benthic community structure from trawling was established (Chapter 2). Having characterized the nature of the impact on the fauna it was necessary to understand how the fauna and trawl impacts affect sediment chemistry. In order to achieve this, a coring technique was developed (Chapter 3). Once the technique was established, it was applied to help quantify the faunal and trawl impacts on sediment chemistry (chapter 4).

Once the effects on sediment chemistry had been assessed, the specific impact of trawl disturbance on nutrient cycling between the sediment and water column needed to be established (Chapter 5). Finally, knowing what effect trawling has on faunal and nutrient dynamics, indirect effects through potentially altered faunal assemblages were determined (chapter 6). These impacts and the concomitant effects on benthic biogeochemistry form the main subject matter of this thesis.

Chapter 2:

Effect of commercial trawling on the benthic infauna of two sediment types in the North Sea: Faunal comparison with untrawled areas.

2.1 Abstract

There is considerable discrepancy between the results of studies on trawl disturbance on macrofaunal assemblages. Many studies have employed experimental trawling techniques that impose an unnatural level of fishing on an area. These approaches lack spatial and temporal clarity and confound the interpretation of data because the reference areas have often already been modified through disturbance. Therefore, while an emergent pattern is evident, specific responses of fauna at a scale representative of commercial fisheries is lacking. This study examines the impact of benthic trawling on macrofaunal communities from trawled and 'virgin' untrawled areas of the North Sea for mud and sand sediment types. A known, productive and long established fishing ground was sampled to provide data from areas impacted at levels consistent with commercial fishing operations. Untrawled areas (sand and mud) within the fishing ground were identified for seasonal comparative analysis of macrofaunal assemblages.

Multivariate analysis revealed distinct differences in the macrofaunal assemblages in trawled and untrawled areas. In support of the existing paradigm, trawling reduced infaunal species size. Measures of species abundance and species richness however, were higher in trawled areas than at untrawled reference sites. A higher incidence of scavenging species occurred

within disturbed areas. With respect to broad taxonomic groups, polychaetes and echinoderms were more abundant in trawled areas. The polychaetes *Chaetozone setosa*, *Harmothoe imbricata* and *Nephtys* spp were found to be resistant to disturbance as their abundances were higher in the trawled area. In contrast, *Scoloplos armiger* and *Terebellides stroemi* appeared to be sensitive to fishing because their abundance decreased in trawled areas. Finally it is concluded that trawling within the North Sea is likely to maintain the benthos in a permanently altered state.

2.2 Introduction

Fishing activity using mobile gears occurs on every major continental shelf (Collie at al. 2000) and is recognised as the dominant human impact in marine environments (Jennings and Kaiser 1998). Many studies have focused on the impact of trawling on the benthos (for reviews see Collie et al. 2000 and Johnson 2002) because trawls are designed to catch those species that live on or have associations with benthic habitats (Collie et al. 2000). Trawls are therefore dragged over the seabed and often seek to maximise their catch by the use of specific features that penetrate the sediment, thus stimulating target species into the net (Jennings and Kaiser 1998). The depth of penetration into the substratum is governed by trawl and sediment characteristics (Piet et al. 2000). The direct ploughing action of trawl gear through the sediment may cause profound physical affects to infaunal species. Therefore, numerous studies, employing various approaches, have investigated the impact of mobile fishing gears on benthic invertebrate community structure. However, while there has been a consensus on the basic principle that dragged trawl gear can alter

benthic community structure and composition (Collie et al. 2000), there are considerable differences in the nature of the changes that occur.

The extent of the current paradigm of trawl impact experiments is documented by Collie et al. (2000). They undertook a meta-analysis of 57 different investigations (39 publications) on the effects of bottom fishing on benthic faunal communities. The analysis confirmed that the response was broadly consistent between studies. However, the magnitude of the response varied between gear and habitat type. Specifically, Collie et al. (2000) concluded that the response of benthic fauna is greatest following initial fishing disturbances. Fishing impact was shown to be the most pronounced in stable habitats, in terms of both removal of species (> 50%) and recovery time. Whereas areas subjected to a fishing intensity > 3 times annually (i.e. the North Sea) were likely to exhibit a permanently altered faunal assemblage. Collie et al. (2000) concluded that small-scale pulse or push experimental fishing in areas that have already been fished does not allow predictive conclusions to be drawn. They also noted that the modifications inflicted on faunal assemblages by fishing disturbance can mask and lessen the response and recovery displayed in subsequent experimental studies because long-lived sensitive species have already been removed and recolonisation would occur predominantly through immigration. Consequently, the present resident faunal community (i.e. often sampled as controls in before/after, control/impact (BACI) type experiments) is not representative of the original assemblage (Collie et al. 2000). Collie et al. (2000) advocate that future studies should dispense with short-term, small-scale experimental trawling and focus on fishery scale impacts that are compared with designated protected areas.

Despite the recommendations of Collie et al. (2000) recent studies on the impact of fishing on the benthos still predominantly undertake experimental trawling and consequently discrepancies in the findings are still occurring. For example Drabsch et al. (2001) conducted a BACI design experiment to assess the impact on benthic infauna of experimental otter trawling. The study was replicated in an area reported to have been exposed to little or no trawling for 15 years. Drabsch et al. (2001) showed a decrease in total abundance, yet it was not attributable to trawling due to high variability within the controls. Drabsch et al. (2001) concluded the affect of otter trawl activity on infauna to be within the range of natural variation. The study by Drabsch et al. (2001) was conducted in shallow water, ~ 20 m deep in coastal locations in the Gulf of St Vincent, Australia. Shallow water locations are commonly used for experimental trawling as they pose fewer logistical sampling problems (Emerson et al. 1984), however an unfortunate consequence is that these areas typically display high levels of natural disturbance which may mask trawl effects (Brylinsky et al. 1994, Kendrick et al. 1998). Furthermore, the haphazard method in which sites were selected resulted in different sediment types at each area. Differing sediment types typically support different benthic communities, affecting community composition (Freeman and Rogers 2003), and this may explain the inconsistent results Drabsch et al. (2001) found following trawl impact.

The effect of trawling on benthic fauna in a sheltered sea loch that had been closed to fishing for ~ 25 years was examined by Tuck et al. (1998). A treatment area (~ 2 km x 0.5 km) was trawled monthly for 16 months with modified rock hopper gear (without a net). In contrast to Drabsch et al. (2001), results from Tuck et al. (1998) revealed species richness to increase, while

diversity and evenness decreased following trawling. While no significant change occurred in total abundance or biomass, the polychaete *Chaetozone setosa* was found to be most resistant to trawl disturbance as its abundance increased post disturbance. However, the conclusions of this study may not provide wide-ranging transferable information at spatial scales consistent with commercial fisheries. The study by Tuck et al. (1998) consisted of an isolated environment that was not spatially replicated. The loch was semi-enclosed by a sill that only permitted surface exchange with an adjoining estuary, yet helped retain high levels of domestic sewage discharged into the loch (Tuck et al. 1998). The apparent increase in the polychaete *C. setosa* from trawl activity may therefore have occurred as a result of the loch's high anthropogenic nutrient level, a phenomenon shown to stimulate polychaete proliferation in other marine areas (Kroncke et al. 1998). Scavengers, migrating into the relatively small treatment area to feed on moribund organisms, may also have biased the results of Tuck et al. (1998).

In contrast, a BACI type study conducted by Sparks-McConkey and Watling (2001) showed *C. setosa*, among other species, to be sensitive to trawling. Sparks-McConkey and Watling (2001) used a short-term (4 trawl tows during one ebb tide) experimental otter trawl manipulation to quantify trawl impact on an area that had been closed to trawling for ~ 20 years. Immediately post trawl disturbance, species richness, abundance and diversity decreased. This trend of a short-term decrease in benthic biomass following experimental trawling is well documented (for example, Bergman and Hup 1992, Kenchington et al. 2001, Moran and Stephenson 2000, Prena et al. 1999).

The studies outlined above are in well defined, localised areas, and are perhaps not directly applicable to areas used for commercial fisheries purposes. Recent studies (summarised in Collie et al. 2000) generally agree that the macrofaunal mortality produced by direct physical contact with trawl gear is typically greater on larger and more fragile species, which tend to have lower intrinsic rates of population increase than smaller species (Jennings et al. 2001). Therefore, the initial trawl impact can be predicted to cause the most profound reduction in biomass in stable sediment habitats (Tuck et al. 1998). However, depending on the frequency of disturbance, trawling may lead to long-term changes in the benthic assemblage of macrofauna (Jennings et al. 2002). In repeatedly trawled areas investigators have documented a shift from large bodied, long lived species to those displaying a smaller size spectra and an increased productivity (Jennings et al. 2001).

The diversity in these findings on the impacts on benthic infauna from mobile fishing highlights the difficulty in quantifying the impact of trawling on the benthos. Differing trawl regimes have varying effects due to the type, weight and tow speed of ground gear. The type of substratum can also affect the severity of a trawl impact. Temporal and spatial variations in the benthos occur in response to disturbance events such as wave action and tidal currents (Hall et al. 1993a). Separating the effects of trawling from such inherent natural variability is often difficult. Another problem is that almost every area that is amenable to trawling will at some point have been trawled to some degree (Engel and Kvitek 1998). Therefore the lack of viable controls for experimental comparison may limit our ability to discern the real affect trawling has had on benthic infaunal assemblages.

Post-trawl recovery periods vary considerably between studies, ranging from a month, or less, to decades (Hutchings 2000). Benthic community recovery varies depending on the stability and structural complexity of the habitat and the frequency of the disturbances, whether natural or anthropogenic. However, it is likely that recovery rates of mobile species from experimental trawl studies are significantly influenced by immigration from the surrounding areas. Brey (1999) reported that reproduction of macrofauna could not occur within most recovery times documented in current experimental literature because increased body size is correlated with slower growth. Thrush et al. (1996) found that recolonisation and immigration were lower following large scale disturbances as compared to smaller scale disturbances. This suggests that experimental trawling studies may not disturb on scales representative of commercial fisheries and recovery rates may be biased by immigration.

Following Collie et al. (2000), an alternative approach is to sample within a known, productive and long established fishing ground. This method should yield clear interpretable data as the infaunal community sampled will be the product of years of trawl disturbance at real commercial fisheries scale and intensity. An untrawled area for comparative analysis is then required. Unfortunately it is well documented that in intensively fished areas such as the North Sea, there is a distinct lack of unfished control sites (Collie et al. 2000). However, fishers are notoriously non-random in selecting the grounds they trawl (Rijnsdorp et al. 1998). Therefore, fishing effort is not homogeneously distributed and fishers will avoid areas with obstructions and rough ground that could damage their trawl gear (Kaiser 1998).

Liaising with local fishers can thus provide vital information on areas that are avoided owing to bad ground and obstacles surrounding an otherwise fishable area. As a result, local knowledge of ICES statistical rectangle 39E8 helped identify two areas that are untrawled due to the presence of a wreck and rocky reefs. These areas were acoustically mapped and this revealed areas between these features that could be accessed with sampling equipment, but which were too small for the deployment of commercial fishing gear. This allowed two sites, adjacent to existing fishing grounds, to be examined in a fashion that avoids the problems inherent in experimental trawl studies.

This study examines the impact of benthic trawling on macrofaunal communities from trawled and untrawled areas of the North Sea. Mud and sand sediment types were studied and a sampling strategy that accounted for variations within a season was employed. Variations in benthic community structure were assessed using species richness, abundance, diversity and multi-dimensional scaling to test the hypothesis that trawling affects benthic macrofaunal assemblages. It was further predicted that direct trawling induced mortality removes larger long-lived species allowing the proliferation of smaller species.

2.3 Method

Trawlable sediments within ICES statistical rectangle 39E8 are typically of two types; mean particle size < 177 µm (referred to as sand) and mean particle size < 88 µm (referred to as mud) (Wentworth 1922). Within areas of each sediment type, trawled and untrawled sites were identified. Following the

approach outlined above, discussions with local fishermen revealed areas that were avoided by trawlers (Fig 2.1). The untrawled sites remain undisturbed due to the presence of a wreck and rocky reefs that were well known to local fishers and prevented the deployment of commercial trawl gear. It is acknowledged that with only one untrawled area for each sediment type the design is pseudoreplicated. Yet, I contend that the nature of sampling 'virgin' areas of the sea bed combined with a statistical approach to assessing differences between treatments allows robust conclusions to be drawn.

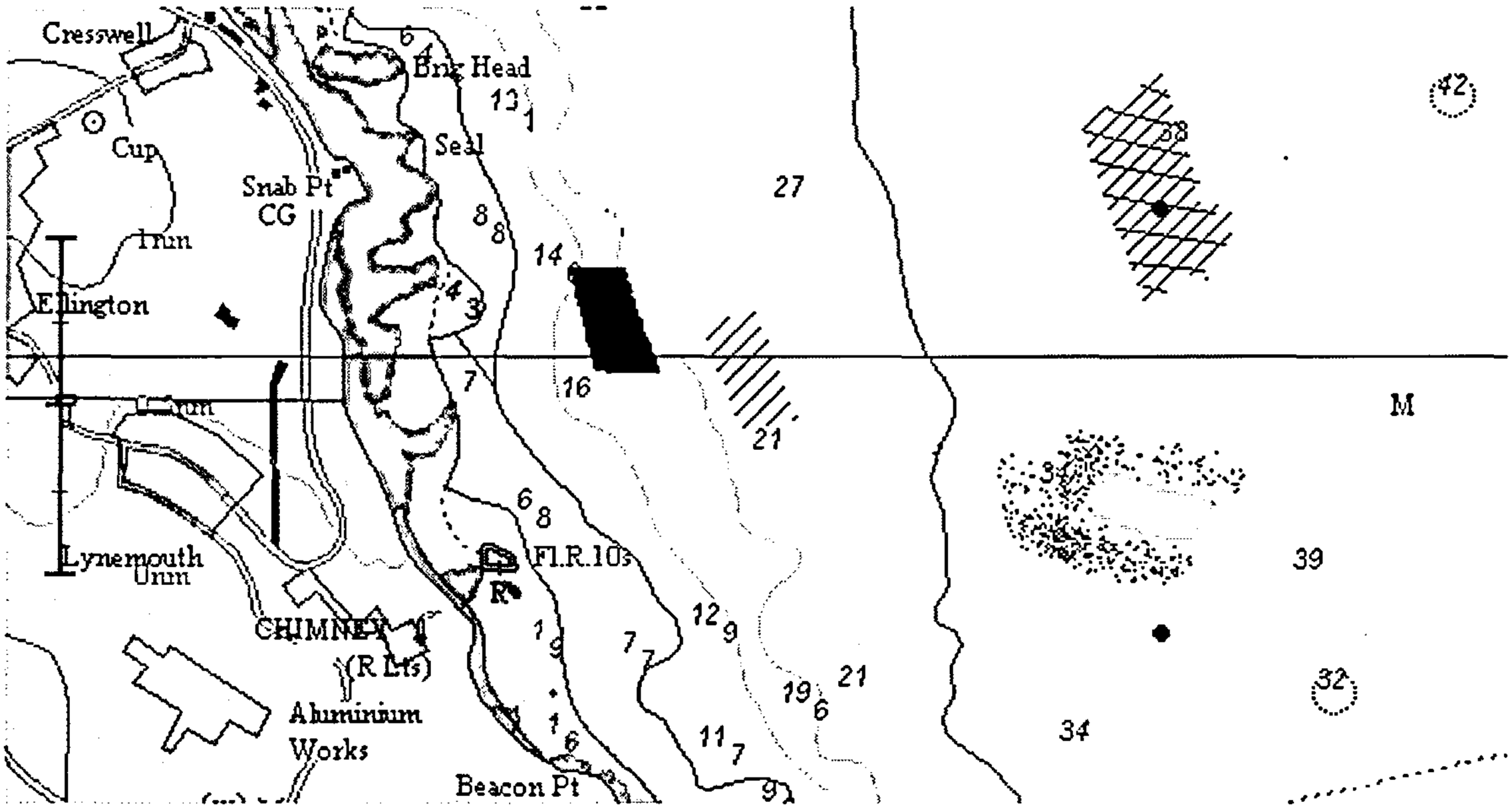

Figure 2.1. Coastal outline of north-east England and sampling sites within the central-west North Sea: Untrawled mud (grey box): $55^0$12.64'N $01^0$27.20'W; trawled mud (hatched line box): $55^0$13.55'N $01^0$27.28'W; Untrawled sand (black box): $55^0$13.15'N $01^0$30.09'W; trawled sand (diagonal line box): $55^0$13.00'N $01^0$29.42'W.

At each of the four sites, six sediment samples were collected on cruises of RV *Bernicia,* using a van Veen grab ($0.1m^2$). Five samples were taken for macrofaunal analysis and the sixth was for sediment analysis. Samples were collected during winter (1/2/2001) and summer (13/8/2001). In order to incorporate different sediment types, while maintaining an untrawled control, the

sand and mud sites were located at different depths. The mud sites were at ~ 40m depth while the sand sites were ~ 30m deep.

The samples were immediately sieved onboard (0.5mm mesh aperture). Retained macrofauna were transferred to sample jars containing 4% formaldehyde and Rose Bengal stain and returned to the laboratory for analysis. Samples reserved for sediment analysis were sub-sampled with a perspex corer, maintained in the dark and frozen in the laboratory until analysis.

2.3.1 Laboratory analysis: Basic sediment characteristics

Sediment grain size was determined using standard gravity sedimentation and mechanical agitation (Buchanan 1984). Porosity was determined by weight loss on drying at 80^0C. Organic matter was measured following wet digestion with hydrogen peroxide in order to account for the possible presence of mineral carbon (coal) (Buchanan 1984).

2.3.2 Macrofaunal analysis

Samples were allowed at least 48 hours for fixing and staining to occur. Samples were then re-sieved (0.5mm mesh aperture) to remove excess stain and formaldehyde. The remaining filtrate was transferred into shallow white trays and sorted by hand to remove all macrofauna. Specimens were then preserved in 70% ethanol before being identified to species level, where possible, and enumerated. Each species was given a size classification derived from the literature (Hayward and Ryland 1990a, Hayward and Ryland 1990b, Hayward and Ryland 1998). Analysis proceeded based on the sizes for each

species (species size) and the mean size of an individual from each sample, i.e. weighted for the abundance of each species within each sample, as follows.

$$sample\ species\ size = \frac{\sum(mean\ length\ of\ an\ individual\ species * species\ abundance)}{total\ abundance}$$

Reported literature values of species size were deemed the most suitable method of determining size as specimens may be damaged during the collection and sorting phases. As a result whole undamaged specimens were inconsistent in the samples and would have biased size calculations.

Comparisons of community structure were made between trawled and untrawled sites for both sediment types and seasons. Differences in species richness, diversity and abundance were examined using a non-parametric one-way Kruskal-Wallis statistical test. Primer (Plymouth Routines in Multivariate Ecological Research) was used to create non-metric Multi-dimensional Scaling (MDS) plots to demonstrate variations in community structure within and between the sites based on Bray-Curtis similarities of each sample (Clarke & Warwick 1994). The faunal datasets were fourth root transformed to reduce the influence of numerically rare species. Evenness and diversity measures were taken from the primer out-put. Measures of diversity range from simple species richness, which can be either the number of species in a sample or number of individuals in a sample through to complex indices which take account of relative abundance of these (e.g. Shannon Wiener index) . Species richness is proportionate to sample size, thus larger samples allow for more species.

Species richness therefore is sensitive to sample size. Evenness is how even the individuals are distributed between species, with high diversity resulting in an even sample and low dominance. The Shannon statistic used by primer is the combination of species richness and evenness.

Unfortunately a potential limitation of this study is spatial pseudoreplication, but this has been overcome by addressing and implementing several study design features identified by the meta-analysis of Collie et al. (2000). Previous studies have shown that fishing can have an affect, however, this study sets to discover in the real world what that affect is. In order to do this it is important to work at real levels and therefore a commercial ground was sampled to provide a more appropriate spatial and temporal scale of trawl disturbance. This was then compared to closed areas. Each closed area was large enough to overcome problem artefacts from the surrounding features but not influenced by fishing effects and therefore removed the need to disentangle affects from a gradient of varying trawl disturbances.

2.4 Results

2.4.1 Sediment analysis

Analysis of sediment characteristics revealed no significant differences between respective trawled and untrawled sites for grain size, organic content (sand: W = 6, p = 0.081; mud: W = 6, p = 0.081) and porosity (sand: W = 15.0, p = 0.08; mud: W = 12, p = 0.66). Grain size values did not vary significantly between seasons with the sand sites displaying a median grain size of 2.5 phi whereas the mud sites had a grain size of 3.5 phi. Consequently I was confident that any differences were due to benthic disturbance.

2.4.2 Sand macrofaunal community structure

Within trawled sand sediments *Magelona mirabilis* and *Tellimya ferruginosa* were the most abundant species during summer and winter respectively. The bivalve *Nucula nitidosa* was the most abundant in untrawled summer samples, whereas *Magelona mirabilis* was the most common species in untrawled sediments during winter.

Comparison of total species abundance between trawled and untrawled sand sediments revealed that greater abundances occurred in trawled sediments (Fig 2.2). These differences were significant during the summer ($H = 3.94$, $P = 0.047$).

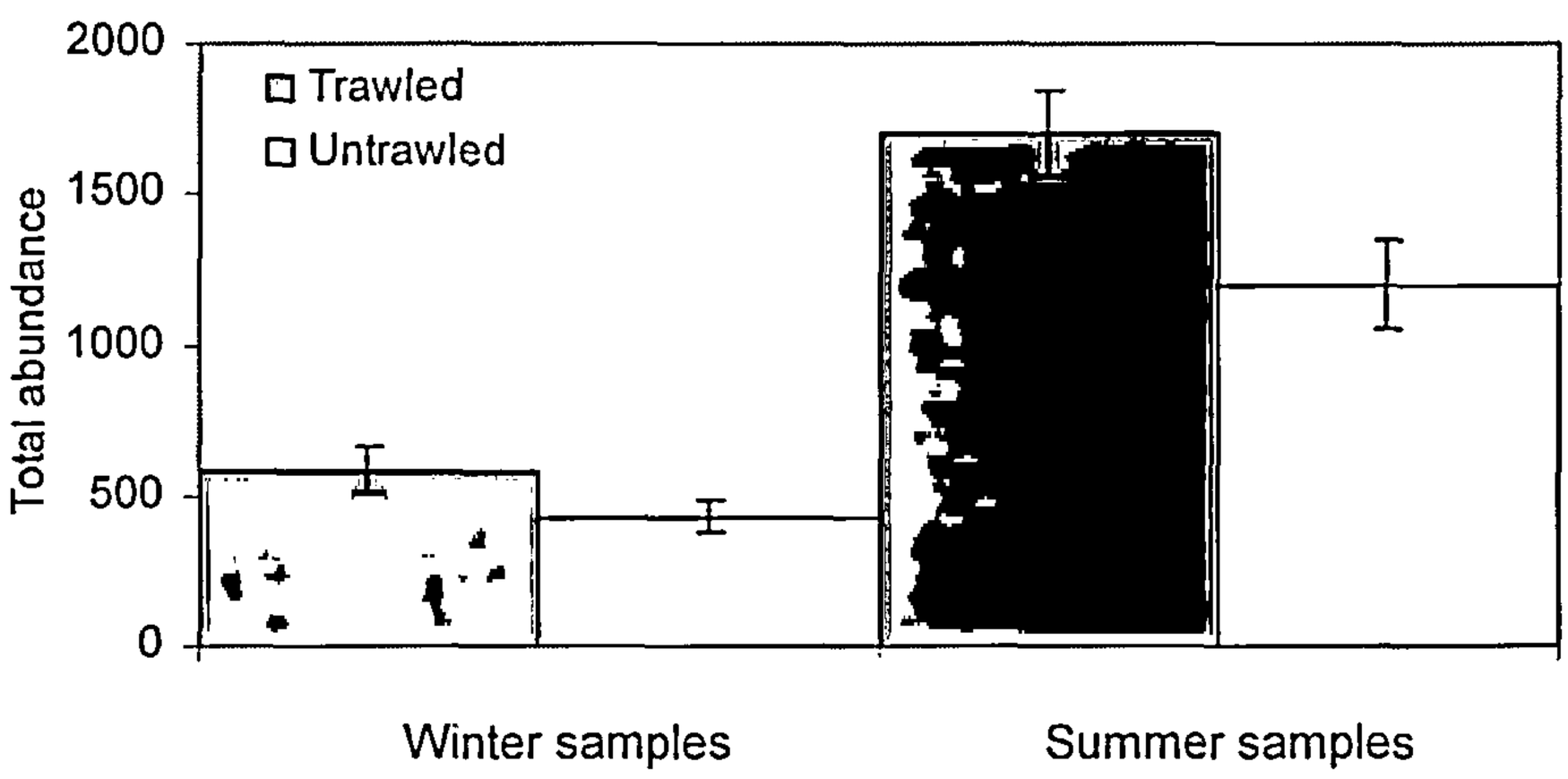

Figure 2.2. Total mean species abundance ($0.1m^2$) within sand sediments from trawled (solid) and untrawled (blank) areas during winter (1/2/01) and summer (13/8/01), ± standard error.

Based on the literature derived maximum sizes, for sand sediments the size of infaunal species recorded were greater within the untrawled samples than the trawled sample, at the same time of year (Fig 2.3). The species present in trawled sand had (maximum sizes) on average 33.6 and 53.5 % smaller, within winter and summer sand, than the species in the untrawled sample.

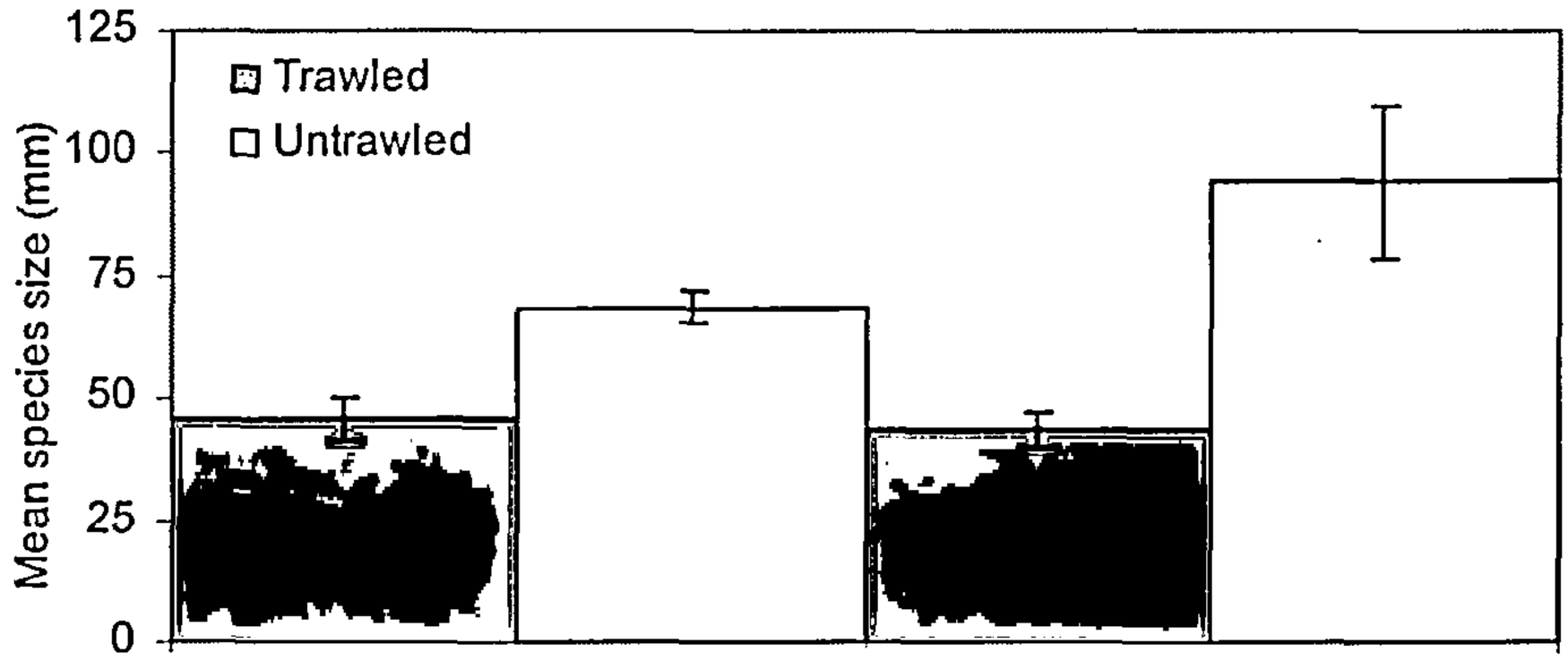

Figure 2.3. Mean maximum size (mm) for species of macrofauna within sand sediments (pooled samples) from trawled (solid) and untrawled (blank) areas during winter (1/2/01) and summer (13/8/01), ± standard error.

The mean 'maximum' body size of a member of the assemblage (i.e. weighted by abundance) was more pronounced in the summer (Fig 2.4), with percentage decreases in trawled compared to untrawled fauna sizes of; 36.4 % (winter) and 61.2 % (summer).

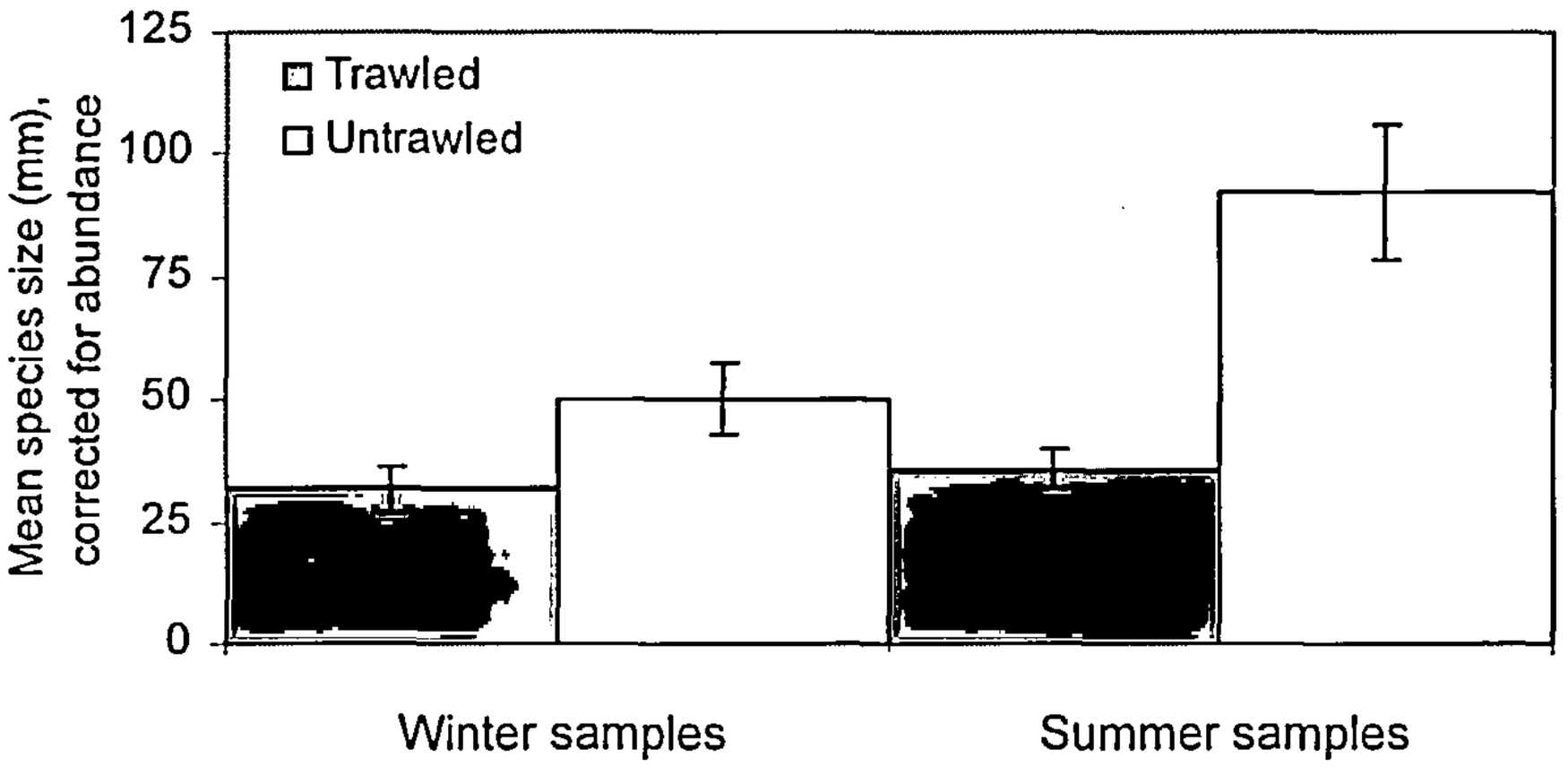

Figure 2.4. Mean size (mm), weighted by abundance, of macrofauna (pooled) within sand sediments from trawled (solid) and untrawled (blank) areas during winter (1/2/01) and summer (13/8/01), ± standard error.

Increased species richness was apparent within the trawled sand sites, irrespective of season, when compared to the respective untrawled area (Fig 2.5). However, significant differences in species richness only occurred

between trawled and untrawled sand sediments in the summer (H = 6.82, P = 0.009).

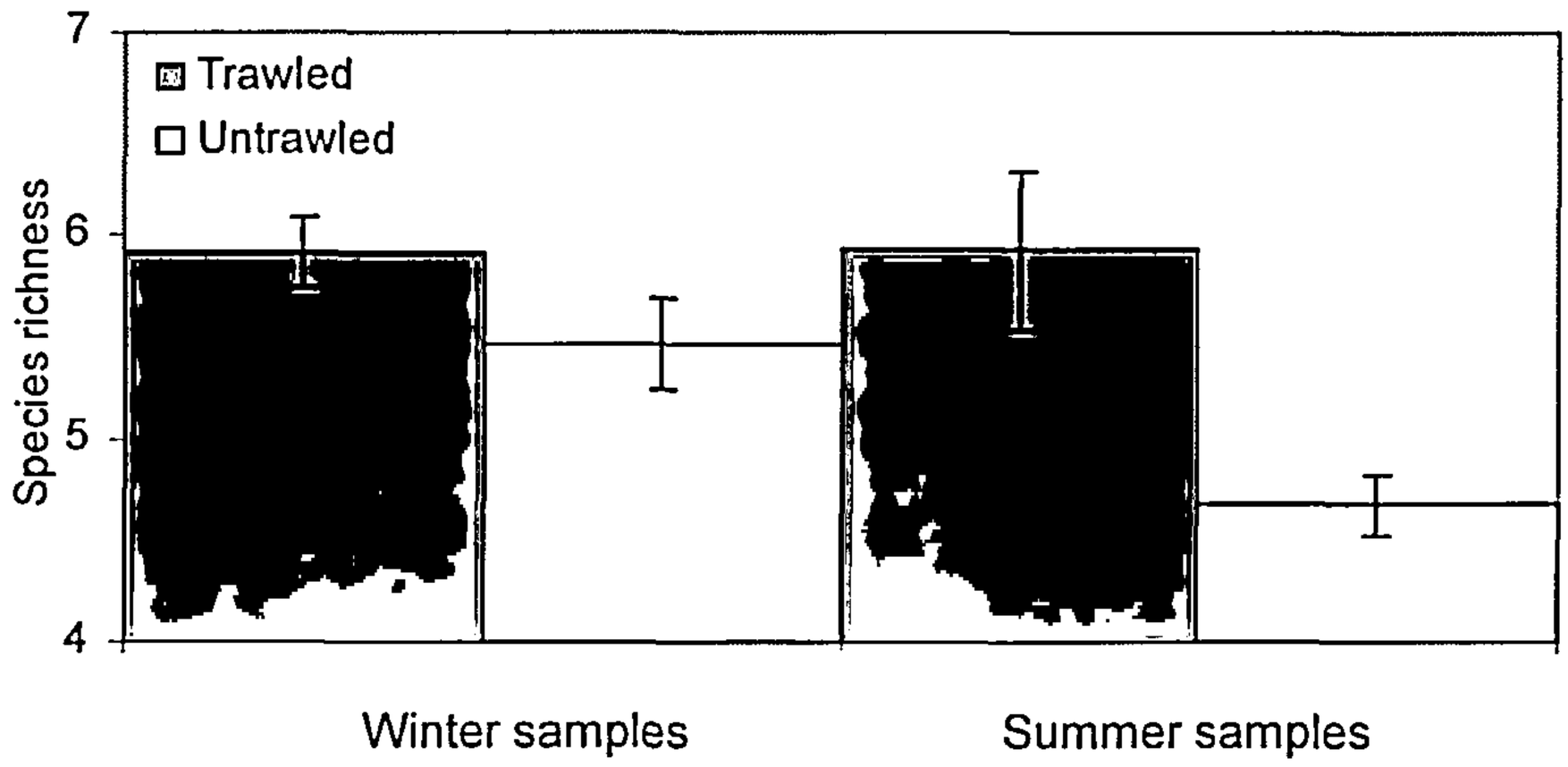

Figure 2.5. Mean species richness (total number of species) within sand sediments from trawled (solid) and untrawled (blank) areas during winter (1/2/01) and summer (13/8/01), ± standard error.

A significantly greater degree of diversity (as measured by the Shannon-Weiner index) of macrofauna occurred in trawled sand sediments compared to the respective untrawled samples (winter sand, H = 5.77, P = 0.016, summer sand, H = 6.82, P = 0.009) (Fig 2.6).

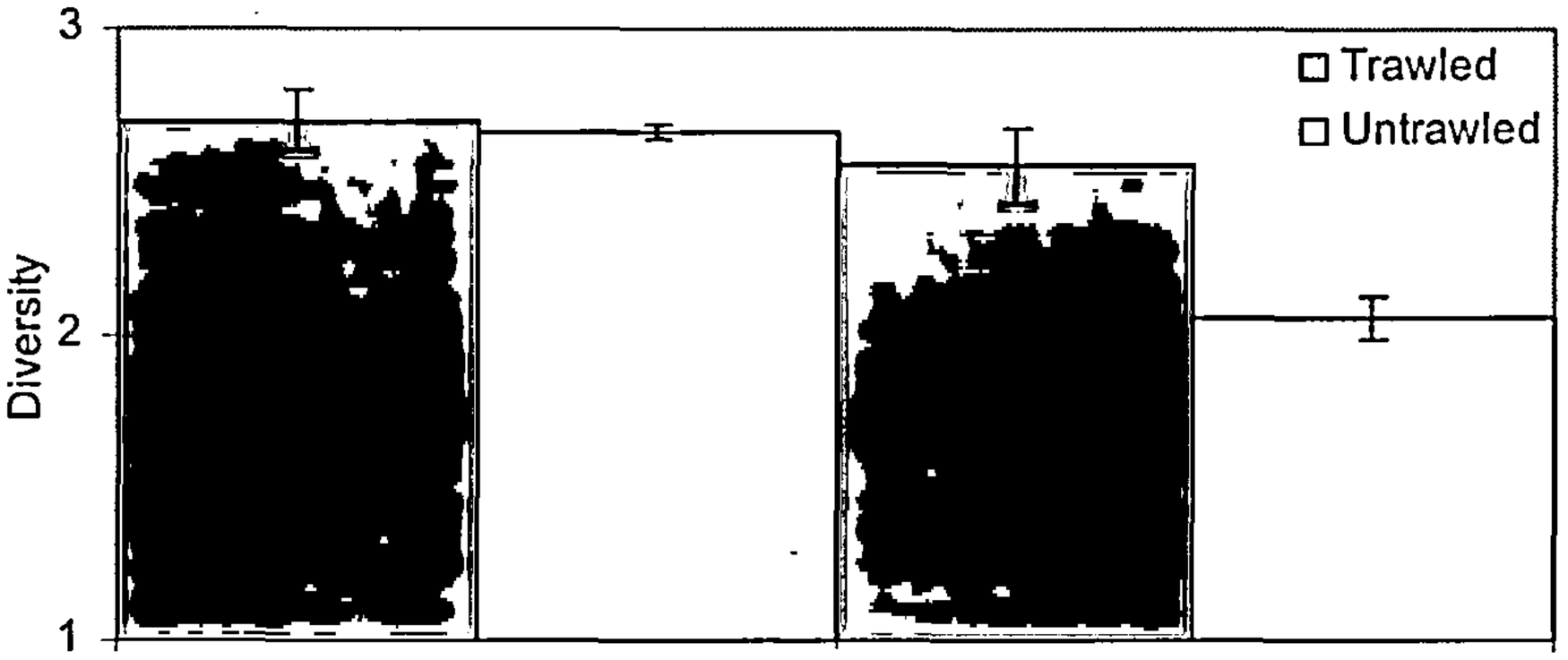

Figure 2.6. Species diversity index (calculated with the Shannon statistic) within sand sediments from trawled (solid) and untrawled (blank) areas during winter (1/2/01) and summer (13/8/01), ± standard error.

MDS ordinations (fourth root transformation) revealed clear statistical differences in community composition between trawled and untrawled areas for

sand sediments (Fig 2.7 and 2.8) (ANOSIM winter sand trawled v's untrawled p < 0.01, r = 0.996; summer sand trawled v's untrawled p < 0.01, r = 0.884). Within the winter sand macrofaunal community the species most responsible for differences between trawled and untrawled areas (calculated as; average dissimilarity / standard deviation) were *Phoronis muelleri, Eunice harassi* and *Capitella capitella* (Table 2.1). The species responsible for differences in the sand community during the summer were *Mya truncata, Dosinia exoleta* and *Venus casina* (Table 2.1). Seasonal differences in the macrofaunal community composition occurred within the sand sediments (Fig 2.9)

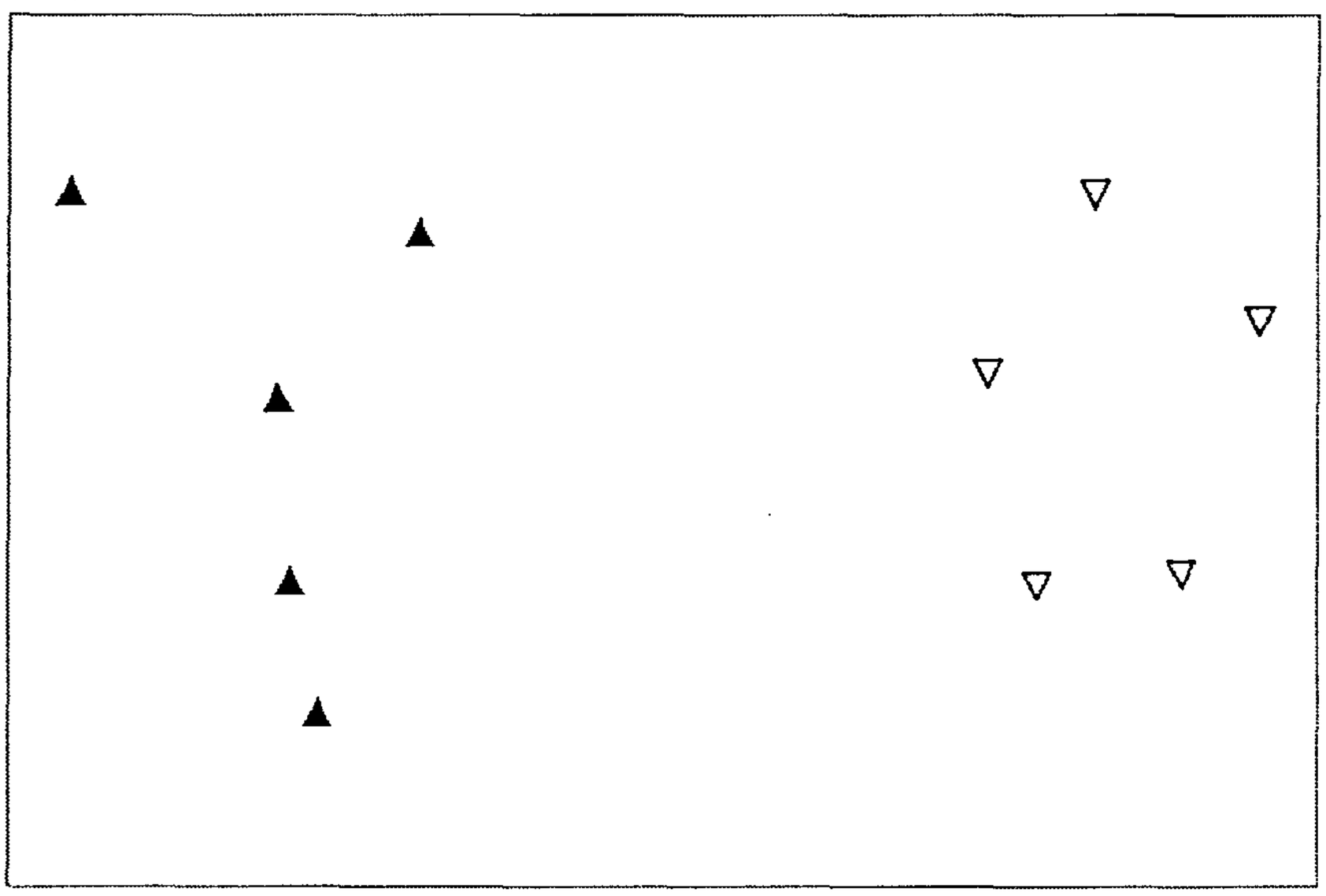

Figure 2.7. Nonparametric multidimensional scaling of transformed (fourth root) total species composition data from trawled (▲) and untrawled (▽) sand sediments during winter (1/2/01). Stress = 0.04.

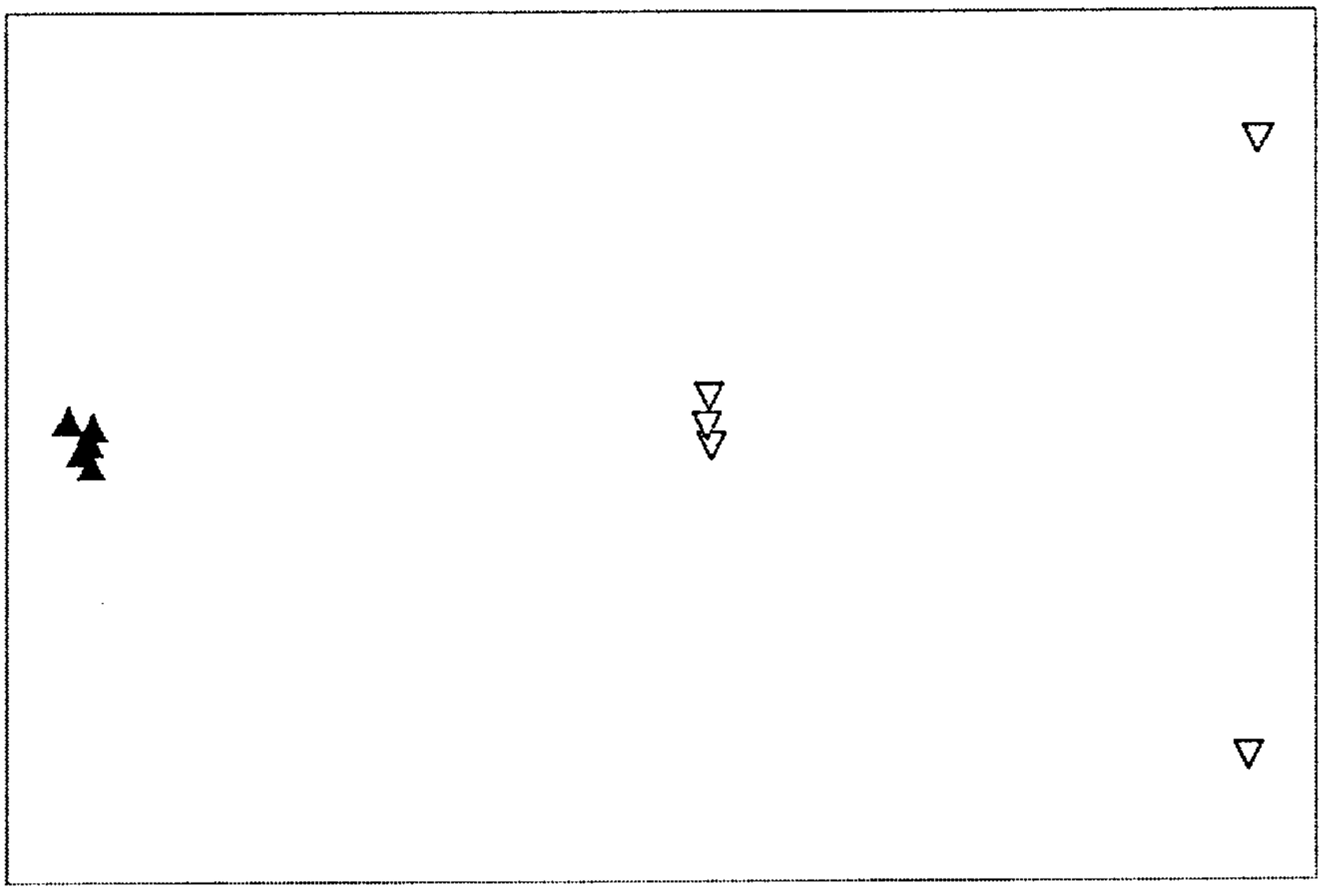

Figure 2.8. Nonparametric multidimensional scaling of transformed (fourth root) total species composition data from trawled (▲) and untrawled (▽) sand sediments during summer (13/8/01). Stress = 0.01.

Table 2.1. SIMPER results highlighting the species most responsible for dissimilarity between groups in sand sediments.

Season	Species	Mean proportion difference in abundance (trawled –untrawled)	Average dissimilarity / standard deviation
Winter	*Capitella capitella*	-1.0	16.28
Winter	*Phoronis muelleri*	6.2	10.44
Winter	*Eunice harassi*	-3	10.23
Summer	*Mya truncata*	32.0	14.53
Summer	*Dosinia exoleta*	3.8	13.44
Summer	*Venus casina*	32.2	12.49

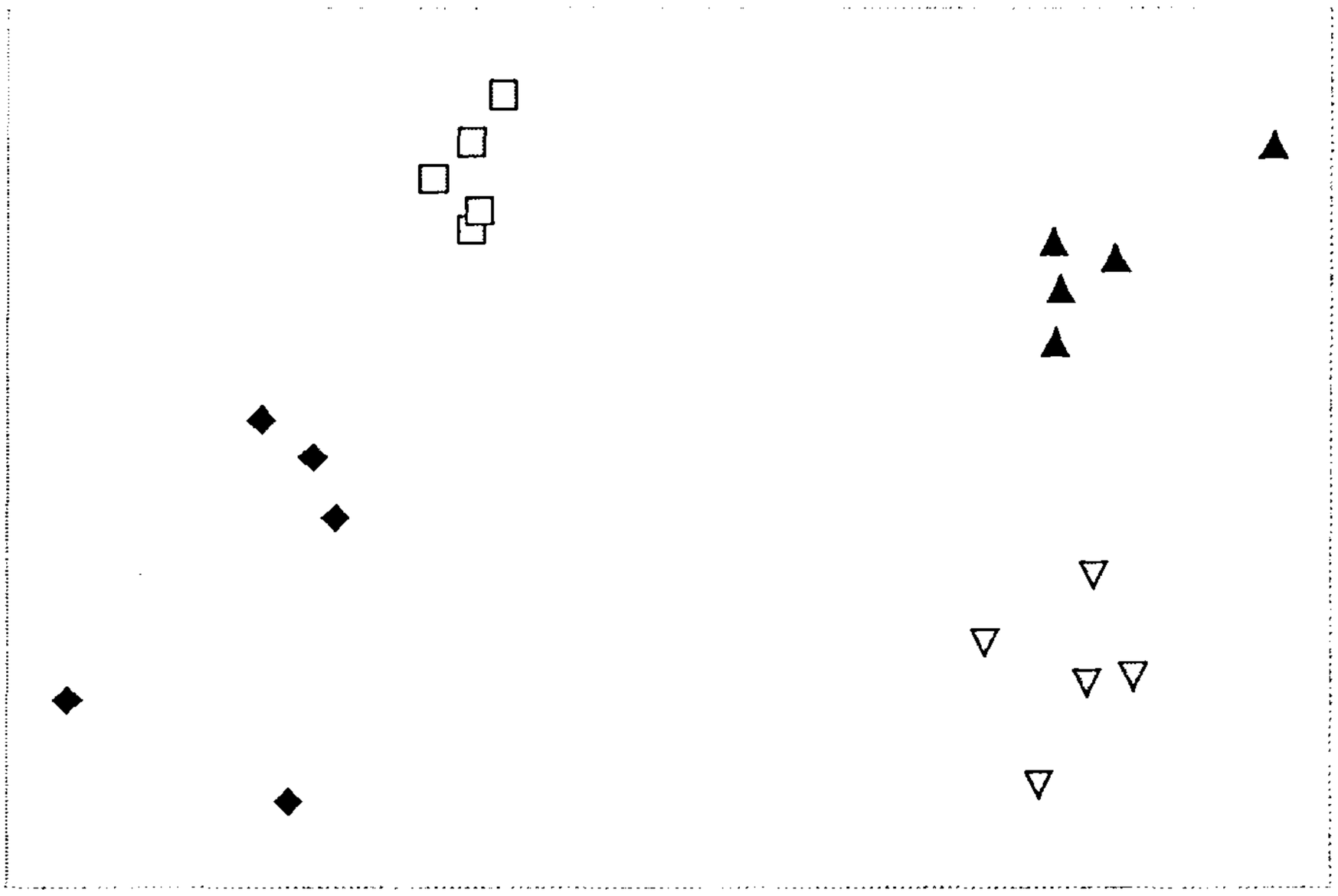

Figure 2.9. Nonparametric multidimensional scaling of transformed (fourth root) total species composition data from trawled and untrawled sand sediments throughout the year. Stress = 0.01. Trawled (1/2/01) = (▲), untrawled (1/2/01) = (▽), trawled (13/8/01) = (□) and untrawled (13/8/01) = (◆).

2.4.3 Mud macrofaunal community structure

The most abundant organisms in trawled mud areas were Maldanidae (polychaete) and *Amphiura chiajei* for summer and winter respectively. In untrawled mud samples *A. chiajei* was the most common species during summer while *T. ferruginosa* was the most common species during winter.

When comparisons were made between trawled and untrawled mud samples during summer and winter, total abundance was higher within each trawled sample (Fig 2.10), but these differences were only significant during summer ($H = 6.82$, $P = 0.009$).

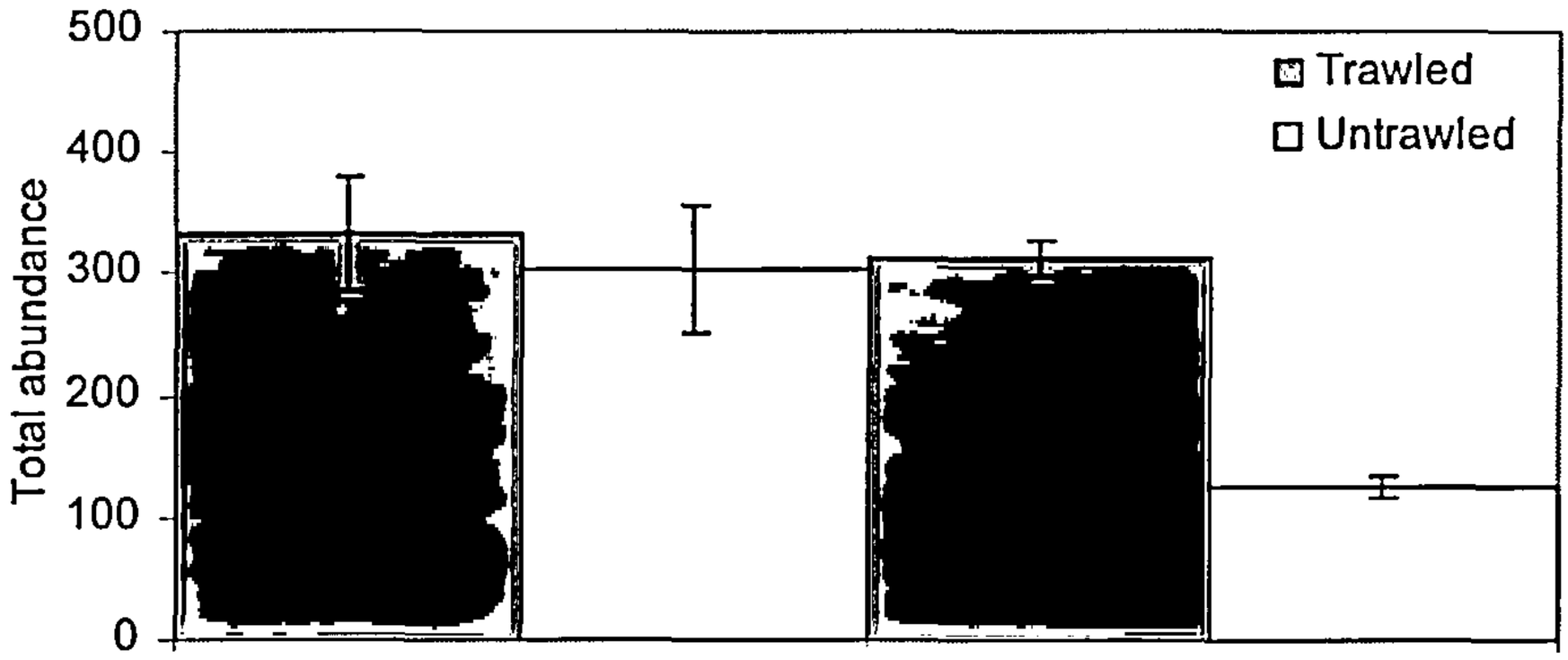

Figure 2.10. Total mean species abundance (pooled samples 0.1m^2) within mud sediments from trawled (solid) and untrawled (blank) areas during winter (1/2/01) and summer (13/8/01), ± standard error.

Analysis of the size of infaunal species present revealed a greater mean size for species to occur within the untrawled samples with respect to the reciprocal (same time of year) trawled sample (Fig 2.11). Species in the trawled samples were on average 31.1 % and 25.1 % smaller, for winter mud and summer mud samples respectively.

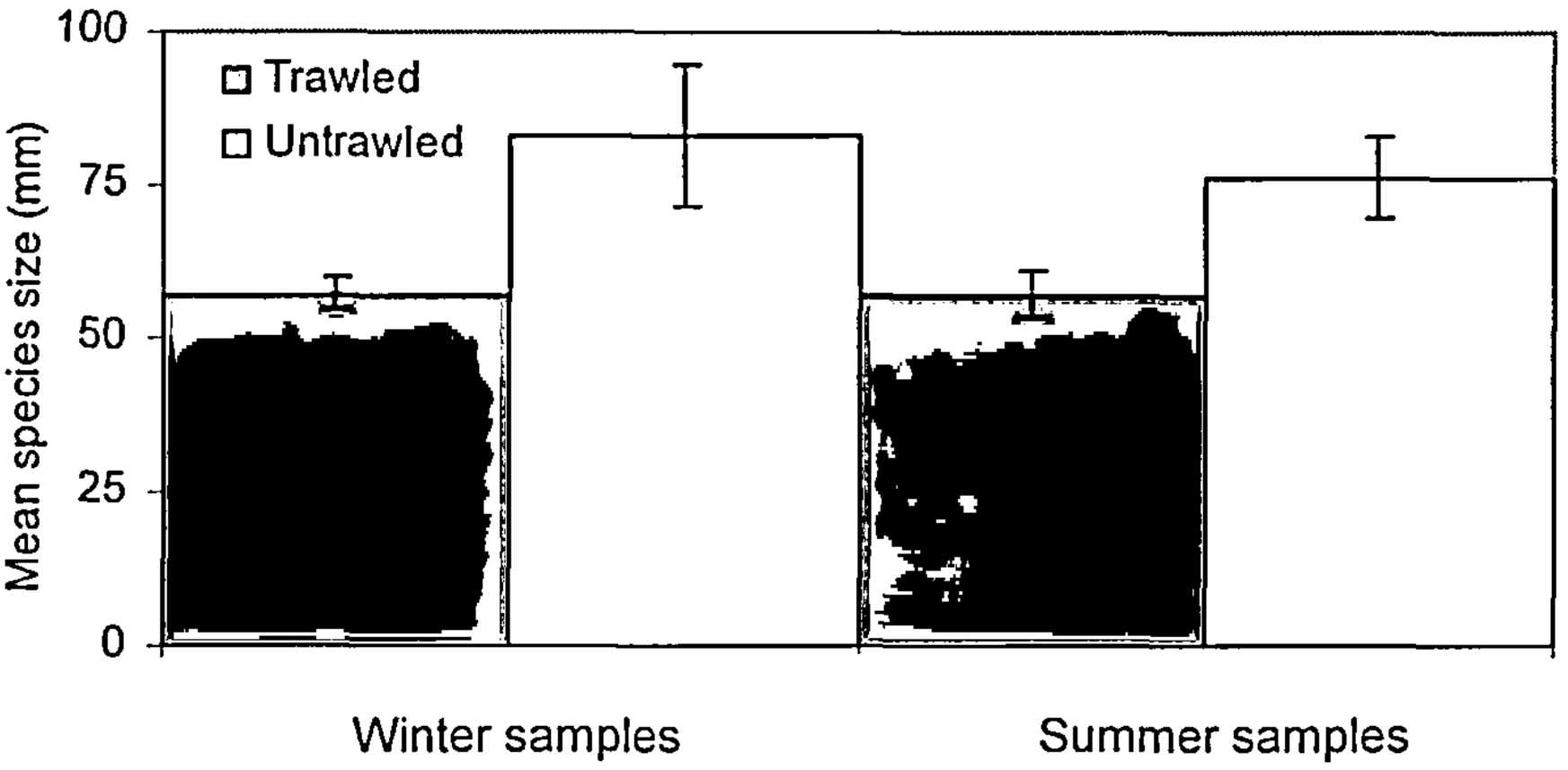

Figure 2.11. Mean species size (mm) of macrofauna (pooled) within mud sediments from trawled (solid) and untrawled (blank) areas during winter (1/2/01) and summer (13/8/01), ± standard error.

Within summer mud samples, a pattern of decreased individual size (weighted by abundance) was displayed by the trawled infauna (Fig 2.12).

Percentage differences between trawled and untrawled areas were 12 % (winter mud) and 5.6 % (summer mud).

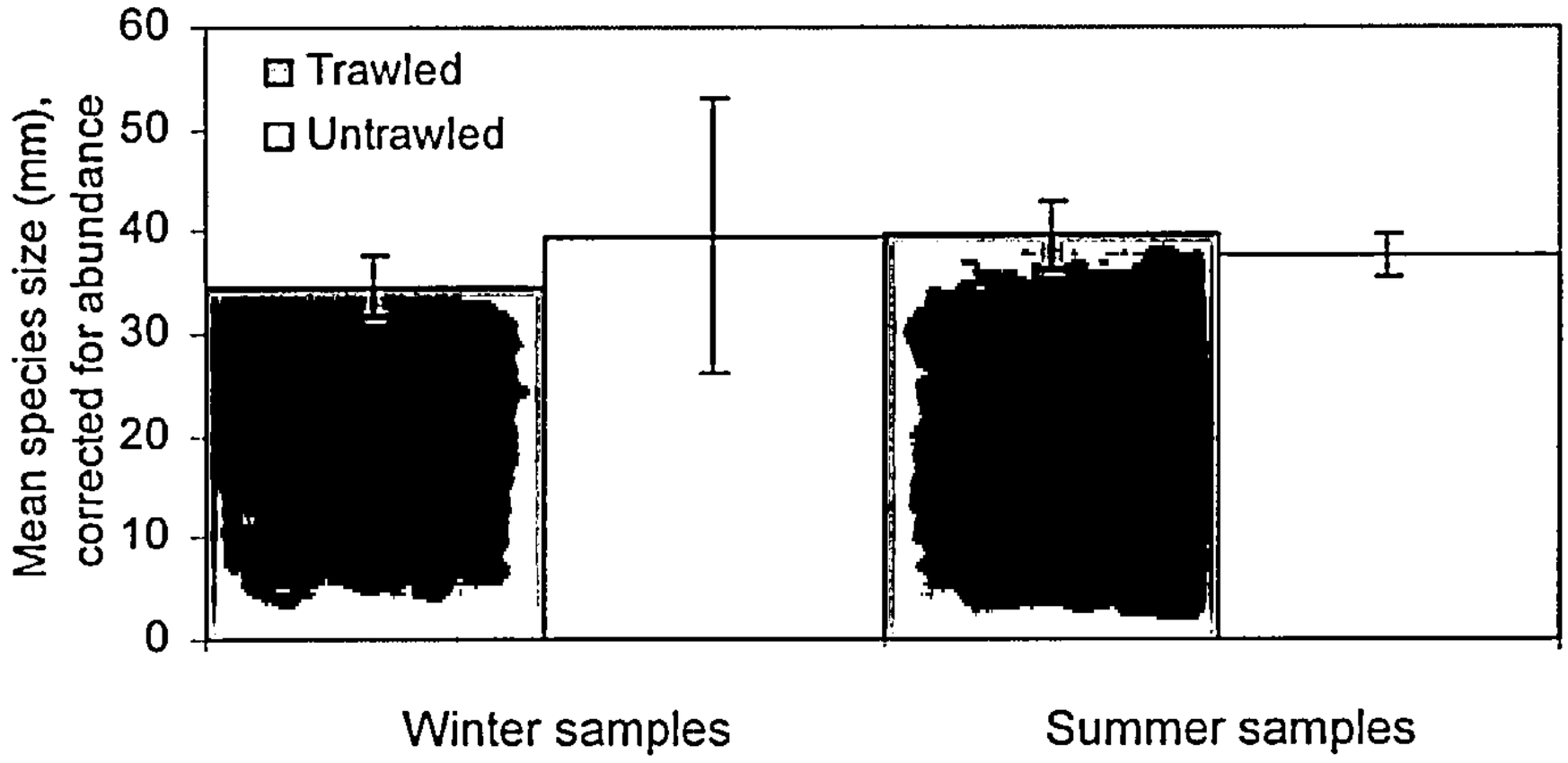

Figure 2.12. Mean species size (mm), corrected for abundance (pooled), of macrofauna within mud sediments from trawled (solid) and untrawled (blank) areas during winter (1/2/01) and summer (13/8/01), ± standard error.

Increased species richness was exhibited within the trawled mud sites when compared to the untrawled area during the same season (Fig 2.13). Species richness was significantly different for mud sediments during the winter ($H = 5.77$, $P = 0.016$).

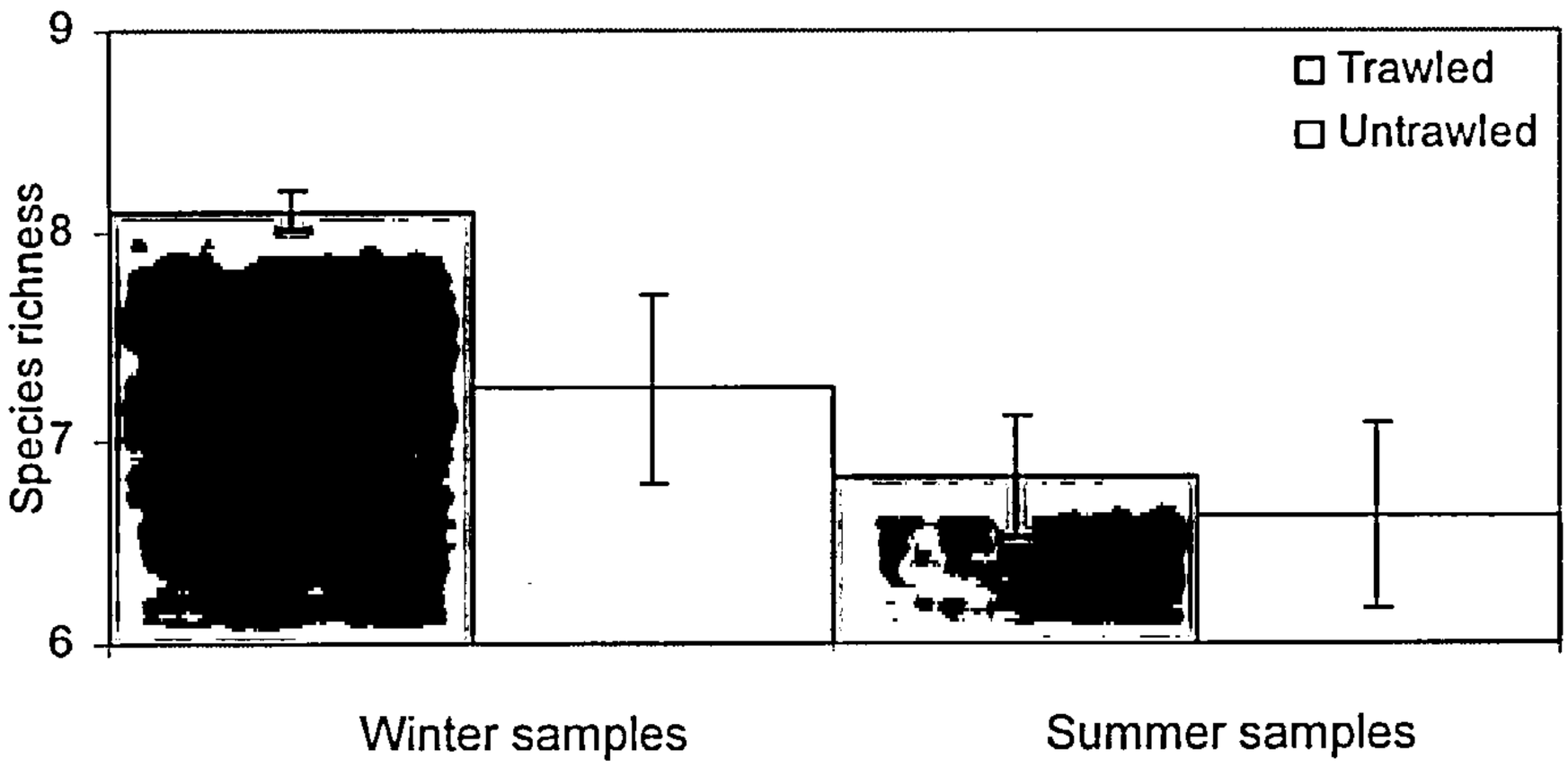

Figure 2.13. Mean species richness (total number of species) within mud sediments from trawled (solid) and untrawled (blank) areas during winter (1/2/01) and summer (13/8/01), ± standard error.

Macrofaunal diversity was greater and only significantly different in trawled mud sediments during winter (Fig 2.14), (H = 6.82, P = 0.009).

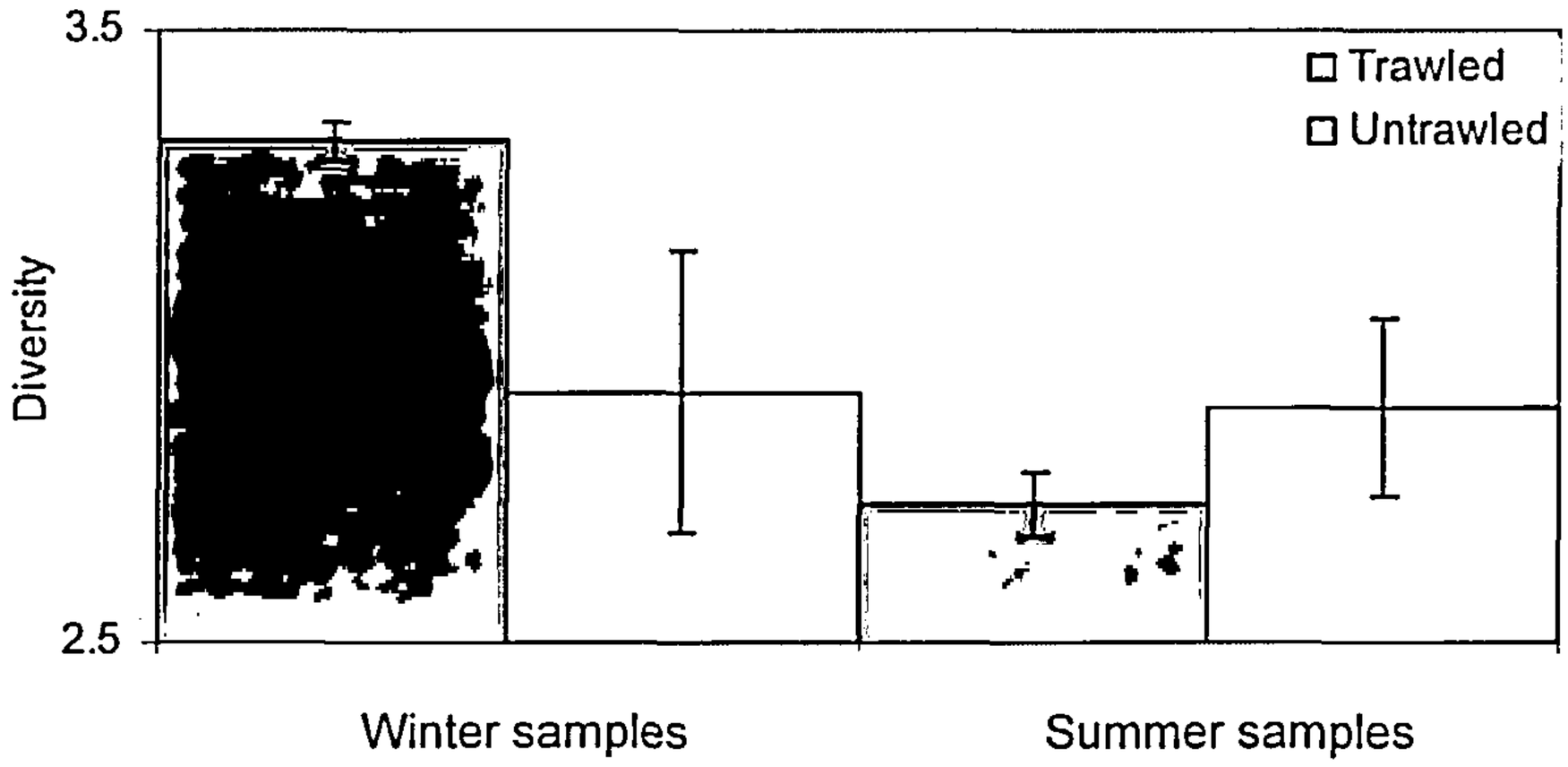

Figure 2.14. Species diversity index (calculated with the Shannon statistic) within mud sediments from trawled (solid) and untrawled (blank) areas during winter (1/2/01) and summer (13/8/01), ± standard error.

MDS ordinations (fourth root transformation) of community structure revealed statistically different results within mud samples between trawled and untrawled areas during winter and summer (Fig 2.15 and 2.16) (ANOSIM winter mud trawled v's untrawled $p < 0.01$, $r = 0.584$; summer mud trawled v's untrawled $p < 0.01$, $r = 0.72$). Species within the macrofaunal community contributing the most to differences between trawled and untrawled muddy sediments during winter were *Capitella capitella,* Hoplonemertea spp and *Mya truncata* (Table 2.2). The species causing the differences within summer muddy sediments were Maldanidae spp, *Ampelisca brevicornis* and *Amphiura filiformis* (Table 2.2). Clear differences within community composition are also apparent between seasons and trawl impact (Fig 2.17).

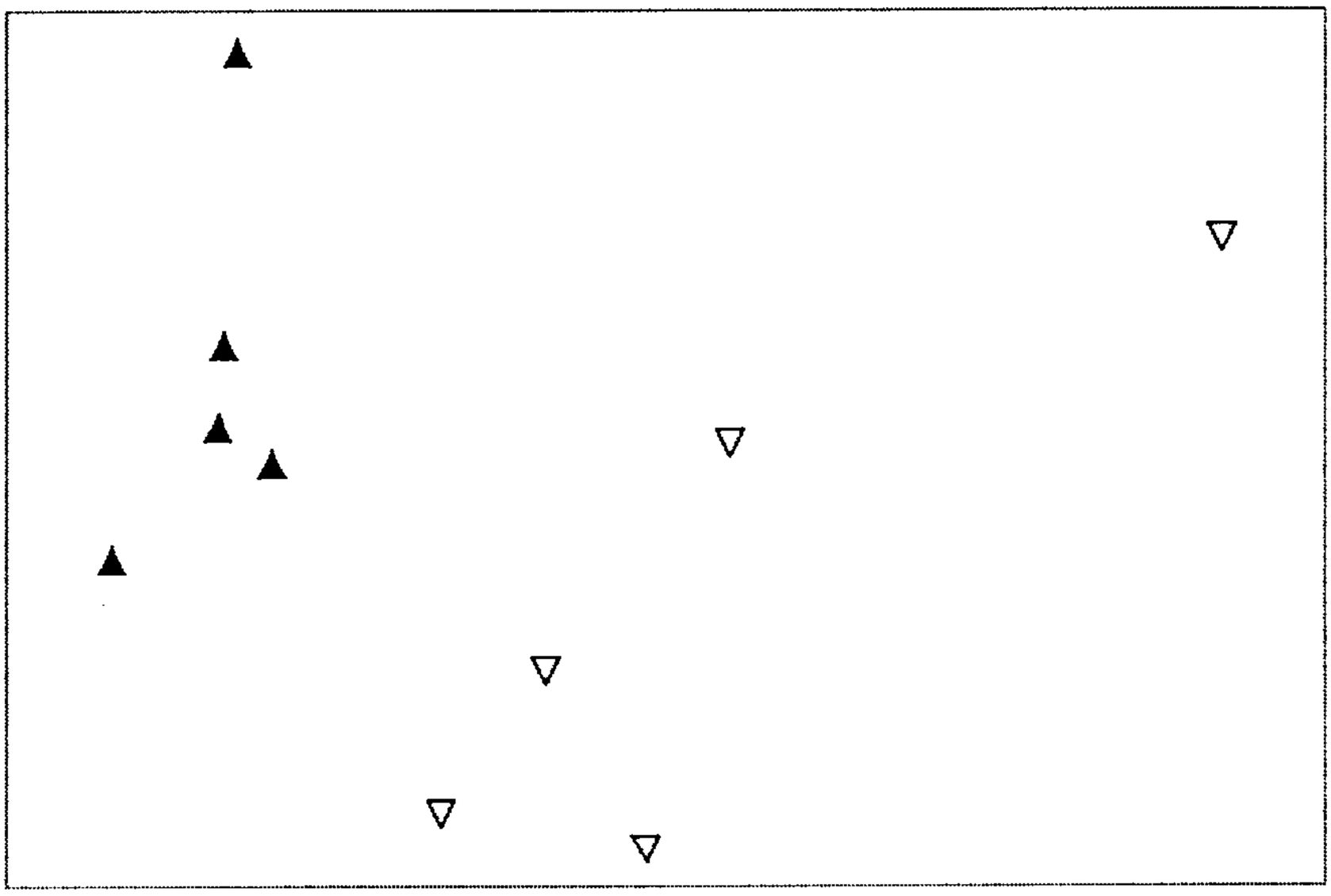

Figure 2.15. Nonparametric multidimensional scaling of transformed (fourth root) total species composition data from trawled (▲) and untrawled (▽) mud sediments during winter (1/2/01). Stress = 0.07.

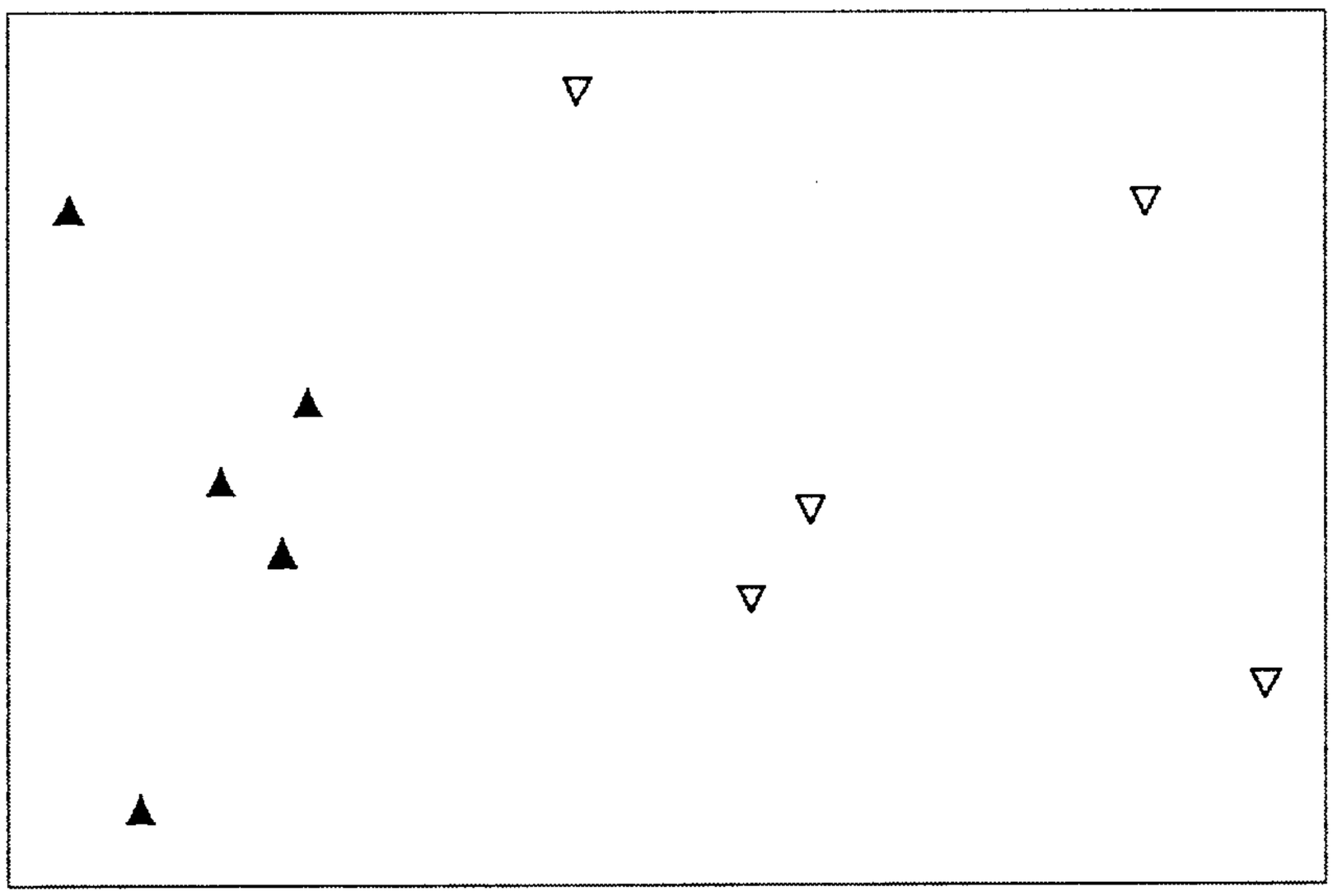

Figure 2.16. Nonparametric multidimensional scaling of transformed (fourth root) total species composition data from trawled (▲) and untrawled (▽) mud sediments during summer (13/8/01). Stress = 0.07.

Table 2.2. SIMPER results highlighting the species most responsible for dissimilarity between groups in mud sediments.

Season	Species	Mean proportion difference in abundance (trawled –untrawled)	Average dissimilarity / standard deviation
Winter	*Capitella capitella*	-2.4	8.35
Winter	Hoplonemertea spp	1.4	7.63
Winter	*Mya truncata*	1.2	1.88
Summer	Maldanidae spp	99.2	3.39
Summer	*Ampelisca brevicornis*	3.4	2.26
Summer	*Amphiura filiformis*	3.8	2.12

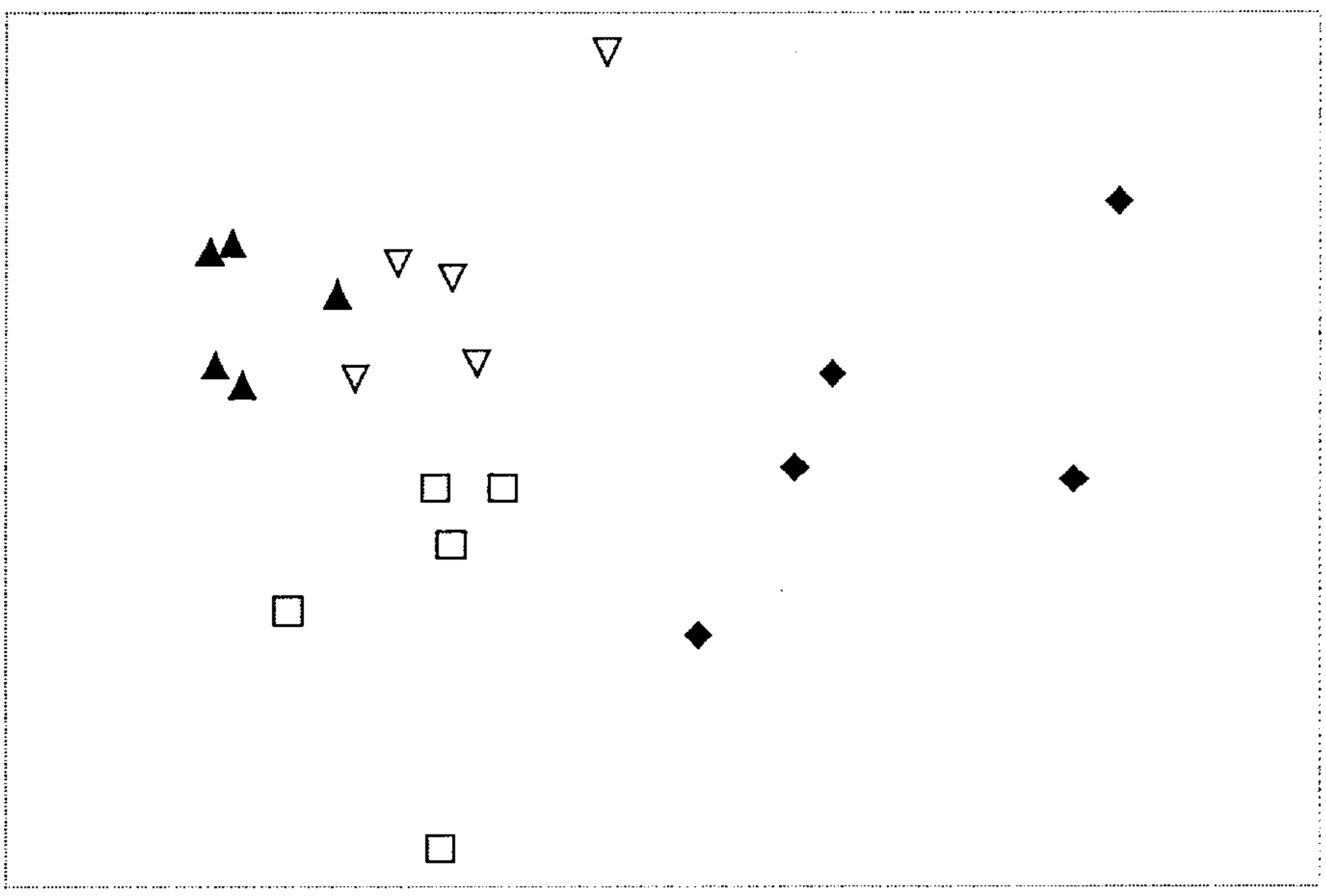

Figure 2.17. Nonparametric multidimensional scaling of transformed (fourth root) total species composition data from trawled and untrawled mud sediments throughout the year. Stress = 0.04. Trawled (1/2/01) = (▲), untrawled (1/2/01) = (▽), trawled (13/8/01) = (□) and untrawled (13/8/01) = (◆).

2.5 Discussion

The sediment data demonstrated that trawled and untrawled sites within each sediment type displayed similar physical properties. Therefore, any disturbances to trawled and untrawled fauna did not affect sediment characteristics. This concurs with the study of Tuck et al. (1998), who also reported trawl disturbance to have no influence on sediment properties. More importantly, differing sediment types profoundly influence the structure of benthic assemblages (Freeman and Rogers 2003). However, as the natural disturbance regime and sediment type was consistent between trawled and untrawled areas it can be assumed that the inherent natural variability in marine infaunal assemblages would be the same.

This study set out to limit natural physical variations between sites and avoid difficulties in interpretation arising from experimental trawling inflicting an unnatural level of fishing disturbance, thus creating spatial and temporal confounding. Other studies have attempted to assess the impact of trawling on macrobenthic infauna by imposing experimental trawling on areas that had already been modified by previous trawl impacts (Bergman and Hup 1992, Jennings and Kaiser 1998, Kaiser 1998). Even comparisons of fauna from trawled areas with fauna from 'protected areas' declared by regulatory authorities cannot fully account for the real impact of trawling as 'protected areas' were often popular fishing grounds before restrictions came into effect (Poiner et al. 1998). Nevertheless, any level of fishing can have severe effects, as it is often the initial impact that has the largest effect on the infauna (Ocean Studies Board 2002). Unfortunately, there is also scope for 'protected areas' to be fished illegally because, unless they are tracked by the relatively recent

development of satellite monitoring systems, fishing areas have to be inferred through log book and over flight data (Rjinsdorp et al. 1998). Consequently, trawling still occurs in protected areas (Poiner et al. 1998). Therefore studies employing this technique can lack distinct and pristine controls for comparison. In the present study however, the untrawled area remains unfished due to the choice of the fishers and not through legislation. Consequently, it is highly unlikely that the untrawled area had been exposed to trawling. Due to the reasons outlined above it is reasonable to ascribe any differences in macrofaunal community structure to trawl disturbance.

The results of this study concur with the initial theoretical predictions and provide evidence that commercial scale trawling has a profound effect on the resident macrofauna of the central west North Sea. Multivariate analysis showed pronounced differences in the infaunal community structure within sand and mud sediments, between trawled and untrawled sites throughout the year. The sampled trawled sites are long established fishing grounds specifically targeted by north east England's fishing fleet. Therefore the scale and frequency of disturbance argue against the observed faunal response being purely a result of an initial response of the impacted infauna or immigration from surrounding areas. Consistently, between sand and mud sediments and between seasons, albeit only within one year, trawling was shown to increase species richness and species abundance. Trawling impact however, resulted in a reduction in species size.

Trawling disturbance has effectively created a scenario in which large numbers of relatively small species predominate in trawled areas and small numbers of large species predominate in untrawled areas. The significant

differences in species size and abundance between trawled and untrawled areas may have important consequences for ecosystem functioning. The differing physiologies of these species will affect sediment burrowing and irrigation processes because bioturbation activity increases with the log of the organism's size (Wheatcroft et al. 1990). Disturbed areas are also likely to exhibit a greater occurrence of mobile species that scavenge the sediment surface. Therefore the differing life strategies of macrofaunal assemblages between trawled and untrawled areas will inevitably have implications for benthic nutrient dynamics (Chapter 6).

Large bioturbating species have also been shown to act as "ecosystem engineers" that are instrumental in setting diversity and community structure within macrobenthic assemblages (Coleman and Williams 2002). Removal of species that can structure the architecture of habitat and thus alter its complexity or influence the biogeochemistry of sediments could have significant effects on local biodiversity and important water-sediment processes. These large organisms are particularly vulnerable to trawling and thus overfishing can create trophic cascades that cause a decline in such species (Coleman and Williams 2002). The loss of these large species could also indirectly affect community structure and compound the direct effects of trawling.

The data suggest that winter conditions could have influenced some of the community structure parameters at the shallower sand sites. Although differences in total abundance and species richness occurred seasonally between trawled and untrawled sand areas, these differences were only significantly different during summer. Therefore, because the sand sites were located at a shallower depth, increased storm and wave action during winter

may have impacted the larger, more vulnerable untrawled fauna. However, the indirect effect of winter storms may be more responsible for reducing variation between trawled and untrawled areas and be attributable to a passive redistribution of fauna. Furthermore, the mud sites displayed significant differences in species richness and diversity during the winter. At this time trawl effort is high and likely to be the dominant form of disturbance in structuring benthic assemblages at the deeper trawled mud site (> 40m depth). At such depths, the influence of the natural disturbance regime would be reduced and thus suggests that trawling alone lead to an altered benthic macrofauna assemblage.

Overall, the taxonomic groups which showed a consistent increase in abundance in association with trawl disturbance were echinoderms and polychaetes. Lindley et al. (1995) suggested North Sea fishing to have a positive effect on echinoderm populations. It has been suggested in other investigations that echinoderms aggregate in disturbed areas to scavenge in response to increased food availability in the form of damaged organisms (Kaiser and Spencer 1994). However, it must be noted that not all echinoderms and polychaetes will display an increase with trawling as specific functional types within these phyla will be more adapted to trawl impacts. Although the overall proportion of polychaetes was greater in trawled areas (excluding winter sand), *Terebellides stroemi* and *Scoloplos armiger* abundance declined. A decrease in the abundance of *T. stroemi* and *S. armiger* following disturbance is consistent with the findings of Tuck et al. (1998). *S. armiger* was discovered to be sensitive to high sedimentation rates and burial following trawling, whereas

the large body size of *T. stroemi* made it susceptible to high mortality from the direct impact of the trawl gear.

Differing modes of colonisation occurring at the trawled and untrawled sites can be discerned. At the untrawled sites the recruitment mechanism was based on life history succession traits of the resident fauna. Albeit the sand site underwent a short-lived influx of the opportunistic scavenging species *Chaetozone setosa, Harmothoe imbricata* and *Nephtys* spp during the winter, the main resident fauna persisted. Hence the untrawled area exhibited lower diversity, lower abundance and was dominated by those species with a larger body size. These traits are common within low intensity disturbance environments because competition and reproductive succession are controlling factors in structuring the community (Sparks-McConkey and Watling 2001, Thrush et al. 1996). Immigration is attributed as a mode of recruitment in experimental trawl studies (Sparks-McConkey and Watling 2001). However, such mobility dependent immigration is only effective over relatively small spatial scales (Thrush and Roper 1988). Furthermore, as the rate of recovery is scale dependent, the frequency of disturbance in a commercially trawled ground will likely exceed the time required for reproductive succession to achieve full assemblage recovery (trawl disturbance > 3 times annually, based on conservative estimates of recovery in sand sediments from meta-analysis of 57 trawl impact observations by Collie et al. 2000). As a result, recolonisation dynamics in trawled areas would favour those species with trawl resistant strategies and employ a combination of increased reproductive out-put and smaller body size approaches. Therefore, as long as trawling persists, the

trawled areas are likely to be in a permanently altered state compared to untrawled fauna.

Chapter 3: Sediment Core method development

3.1 Sampling procedure methods: considerations and solutions

The primary objective was to implement a sampling strategy in order to obtain a sample that would yield an accurate representation of sediment stratification over depth, together with a sample of overlying water. This required the collection of undisturbed cores to examine the nutrient dynamics within the overlying water, surficial (oxic) sediment, sub-surface (hypoxic) sediment porewaters and those processes occurring across each boundary. Due to the strong redox gradients, which typically occur in sediments and that can have a controlling effect on sediment chemistry (Schink 1989), the corer was designed to be gas tight and of non-contaminating material to prevent the contribution of analytes to the sample. Therefore major considerations included the possible ingress of air into the sample and the prevention of light penetration. Sampling anoxic/hypoxic sediments presents particular sampling difficulties. The introduction of oxygen can have a profound effect on the nature and amount of dissolved organic matter (Hall et al. 1996). Bacterial transformation of organic matter, which ultimately leads to the production of inorganic nutrients within sediments, relies on specific redox boundaries (Van Duyl et al. 1993). More specifically, it is on the rate at which these transformations take place that oxygen exerts a control (Sun et al. 1997). It is well documented that early diagenesis within marine sediments occurs through a series of reactions that are mediated by progressively lower free energy yield derived from oxidation of organic matter (Froelich et al. 1979). Therefore the sampling strategy, storage and analysis procedure must occur without the ingress of oxygen.

The reactions and transformations that take place during sediment diagenesis therefore demand appropriate sampling and handling procedures in order to eliminate errors. In order to create a sampling strategy that was fully amenable to these diverse requirements for sediment and pore water analysis that could be used in marine environments >40m, a viable coring method was developed.

3.2 Core sampling and core modification

The analysis employed dictated that the cores be removable to allow multiple samples to be taken that could be maintained under *in situ* conditions. Therefore, the sampling system included a corer with removable core tubes, a cover plate and flange for attachment to a modified Haps coring frame (Kanneworff and Nicolaisen 1973), with a fitted piston (Fig 3.1 & 3.2) and glove box for laboratory analysis. The Haps corer frame was made from stainless steel and in order to main stability during a swell, was mounted with lead weights (Fig 3.2). Metal or acrylic cores, traditionally used for coring, were replaced with core tubes manufactured from Acrylonitrile Butadiene Styrene (ABS). ABS provided a strong durable material that could withstand a high degree of compression and impact, yet was relatively easy to manufacture to exact specifications. The modified core sleeves allowed for multiple cores to be taken that could be sealed gas-tight tight to prevent any oxidation artefacts occurring during transportation and analysis.

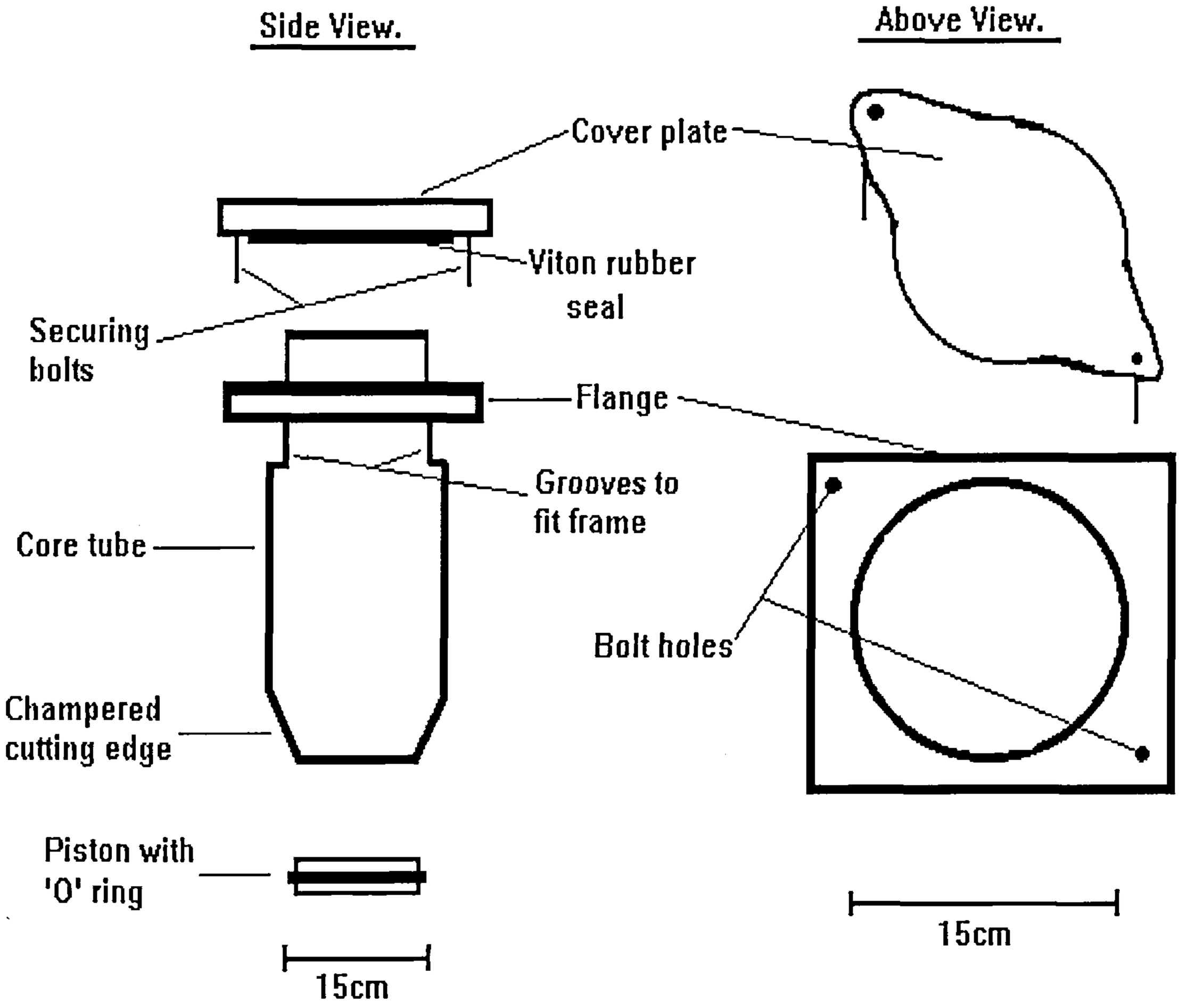

Figure 3.1. Sediment corer for gas-tight transfer of an undisturbed sediment core

3.3 Gas Chromatography

A gas chromatograph (GC) method was employed to test how gas tight the cores were. Numerous studies have reported CH_4 production from sediments (Borowski et al. 1999, Crill and Martens 1983, Martens 1982, Reed et al. 2002). Therefore high CH_4 concentrations may occur within sediment cores and is easily detectible in laboratory air (> 1.8 ppm CH_4 in air). As a result it was deemed appropriate to gas test for CH_4. An empty core tube was sealed and tapped through the cover plate with two Nupro two-way stop cocks. Immediately prior to analysis the core tube was thoroughly flushed with

industrial grade 99.998% minimum oxygen free nitrogen (OFN) gas to expel all trace of laboratory air. A positive pressure of OFN was maintained during flushing to prevent any ingress of air. Following flushing the stopcock valves were sealed to create a fixed volume of OFN within the core tube.

The Shimadzu GC-8A system used for analysis employed flame ionisation detection operated at 120^0C with the carrier gas flow at $25cm^3min^{-1}$. Chromatographic separation was at 60^0C on a 2m x 2mm i.d. Porapak-Q® (80-100 mesh) column. The separation utilises an ultra high purity N_2 carrier which is pre-dried with a molecular sieve 5A and passed through an oxygen trap (Upstill-Goddard et al. 1996). Analytical detection limits, measured as X2 noise at baseline were 0.02 parts per million by volume methane (CH_4).

To facilitate sample removal, the gas volume within the core was maintained through displacement of the sample through one stopcock, into gas-tight 100ml glass syringes, with the simultaneous replacement of OFN via the second stopcock. Sub-samples (~50ml) were taken hourly for six hours following an initial sample taken immediately following the establishment of the OFN fixed volume within the core. The initial sample was used to create an OFN baseline. Aliquots of the samples were immediately injected into the GC. Any air seepage into the core would be detected as a CH_4 peak above the N_2 baseline. This method facilitated minimal and rapid sample handling, thus minimising the potential for air contamination. GC analysis of the initial and all subsequent samples exhibited no CH_4 peak. Precision was 0.02 parts per million by volume methane. Therefore I was confident that a gas tight core method for remote coring had been created.

3.4 Deployment and retrieval

Careful operation of the core frame was needed to retrieve an intact, undisturbed core with a layer of overlying water. Initially, anchorage of the research vessel was required to maintain position over the selected site. The frame and coring assembly was winched at a controlled rate (~ 0.5 ms^{-1}) until they were suspended approximately 2 m above the sediment surface (determined by echo sounder). The rate of deployment was then reduced to approximately 1 m / min^{-1} in order for the frame to be placed on the seabed with minimal disturbance.

Throughout deployment to the sea bed and core penetration, the weighted frame maintained a horizontal position (i.e. the core tube was vertical), thus allowing water to pass through the core tube. The flow of water through the core tube ensured that cover plate would rise up and 'float' above the rim of the core. As a result any water retained in the core would be overlying water and not surface water trapped in the core.

Once on the sediment surface the axle release allowed the steering axle to pass through the guide plate (Fig 3.2). The weighted axle was allowed approximately 30 – 40 minutes to slowly drive down the core and penetrate the sediment in order to take a core of the required depth (~15-20 cm sediment depth; ~10 cm overlying water). After this time the winch wire was made taught to prevent further penetration. The action of tensioning the wire acted to trigger the release mechanism and activate the closing plate. The chamfered edge on the bottom of the core served a dual purpose. Not only did the edge aid penetration into the sediment, but allowed the closing plate to fit exactly under the core and create a seal around the bottom of the core tube.

Once penetration ceased the "floating" cover plate was designed to fall gravimetrically onto the rim of the core tube. Thus, at the top of the core a gas-tight seal was created against the ABS cover plate, fitted with viton rubber, and the core tube rim (Fig 3.1). This seal occurred prior to removal of the core from the sediment and consequently trapped the water overlying the sediment and minimised disturbance to the core during transport back onto the deck of the research vessel. Once on board, the cover plate was further secured by bolting the cover plate to the flange (Fig 3.1).

The bottom of the core, sealed during recovery by the closing plate, was sealed gas-tight by an exact fit machined piston with a viton rubber 'O' ring (Fig 3.1). All viton seals were smeared with a thin film of silicone grease to prevent any sediment particles that could get trapped between the seal and core, reducing the efficiency of the seal. A thin metal plate (2mm) machined to precisely contain the piston was positioned between the closing plate and core tube prior to removal from the core frame to minimise oxygen penetration into the core. Following removal, the piston was positioned directly under the opening of the core and fitted into place. At no time was the bottom of the core directly exposed to the air. Cores were then placed in a water bath maintained at *in situ* bottom temperature and returned to the laboratory for immediate analysis.

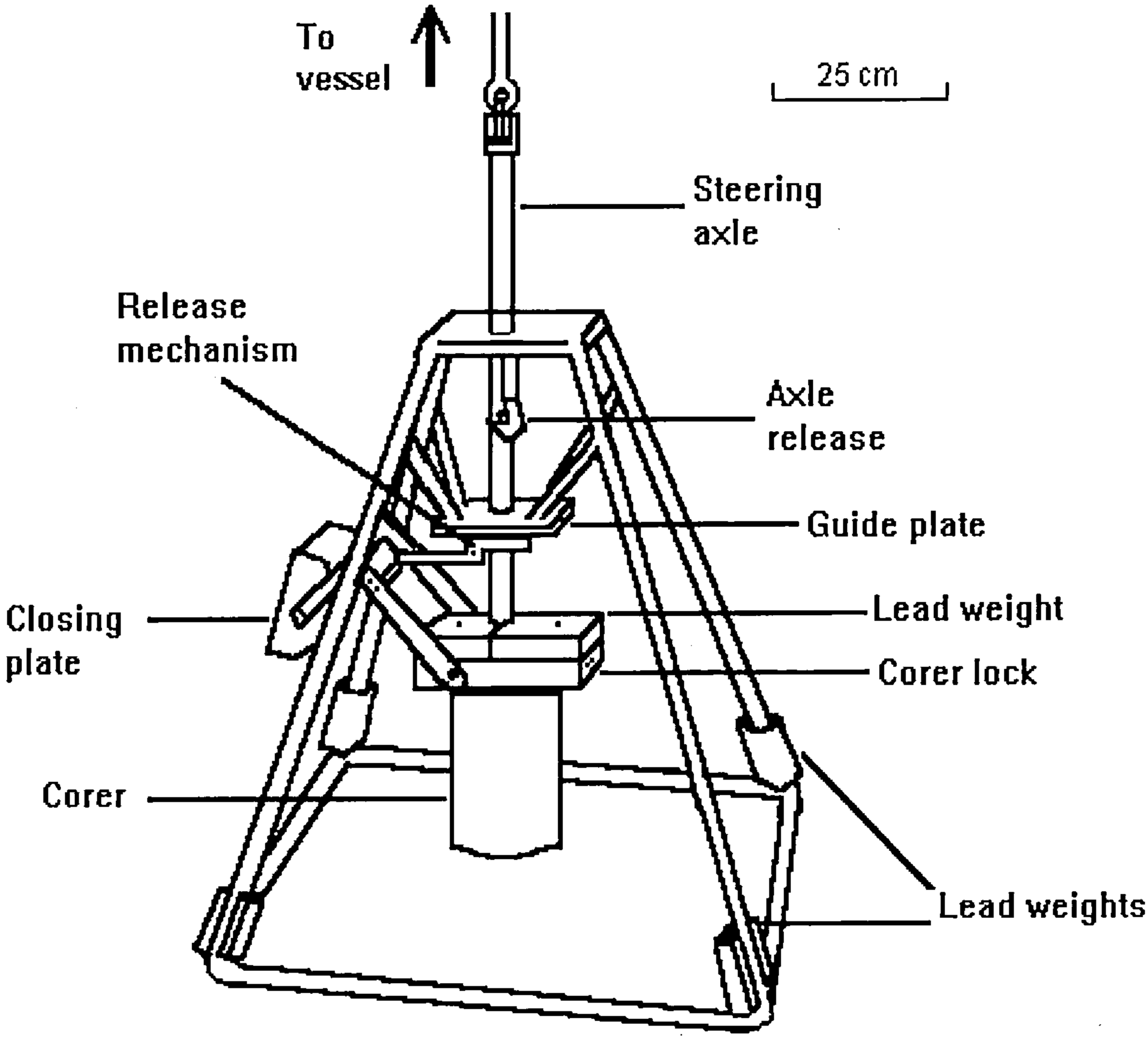

Figure 3.2. Modified frame support for benthic cores.

3.5 The glove box

Laboratory handling of the cores was undertaken under an inert atmosphere within a purpose built glove box (Fig 3.3). The glove box (internal dimensions = 50 cm deep, 100 cm x 75 cm) was constructed of clear perspex and had two front glove/sampling ports, a top access hatch and attachment for a jacking cradle and core tube (Fig 3.3). Prior to core attachment, the glove box was flushed with OFN gas. The flow of OFN was maintained at a pressure

above atmospheric during all handling procedures to ensure that any leakage from the glove box would be outward and prevent any influx of air.

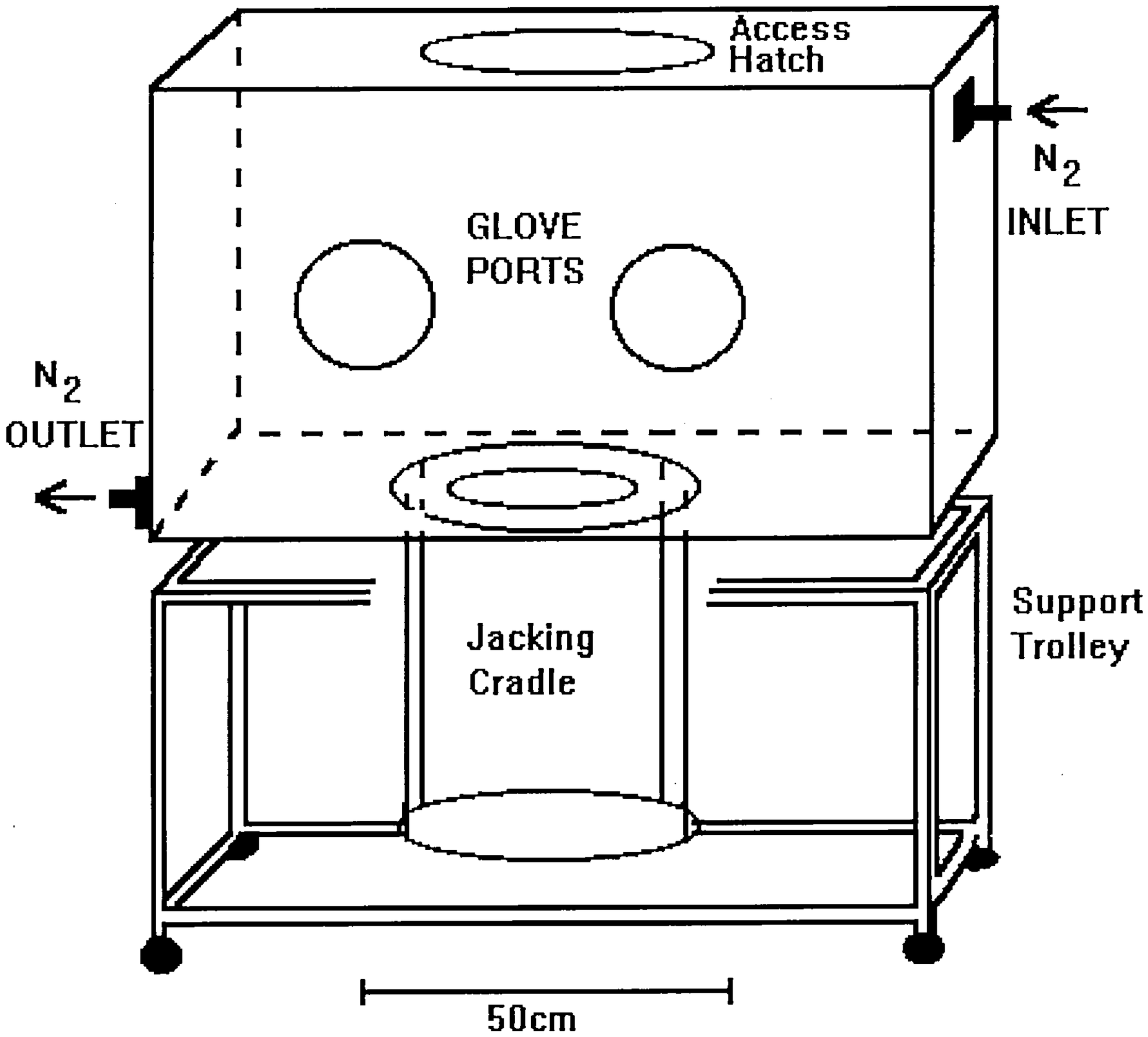

Figure 3.3. Glove box for sediment core sectioning under an inert atmosphere.

3.6 Core attachment to glove box

The following refers to figure 3.4. Cores to be analysed were attached to the jacking cradle (A) by a rubber seal around the corer (B). The cradle was then offered up to the central opening of the glove box from below and attached to twelve permanently fixed bolts (C), with lockdown nuts (D) to the base of the glove box (E). When secured, a gas-tight seal was created against a viton rubber gasket (F). Once attached to the glove box the corer cover plate was sealed within a removable airlock (G). The airlock was secured before the sediment core tubes were attached, with a gas-tight viton rubber gasket (H) and secured with twelve threaded enclosure lockdown screws (I). Following cradle attachment, the corer piston (J) was supported with a modified platform scissor jack (K). The airlock (G) was then thoroughly flushed with nitrogen by use of the inlet (L) and outlet (M) valves. Following thorough flushing with the N_2, the airlock was removed and retained within the glove box. Flat-top nuts were then attached to the tops of the exposed bolts (C) to prevent fouling of the gloves. Once the airlock had been removed, the corer was further secured, against a viton rubber gasket (N), to the cradle by the flange (O), and the cover plate (P) removed.

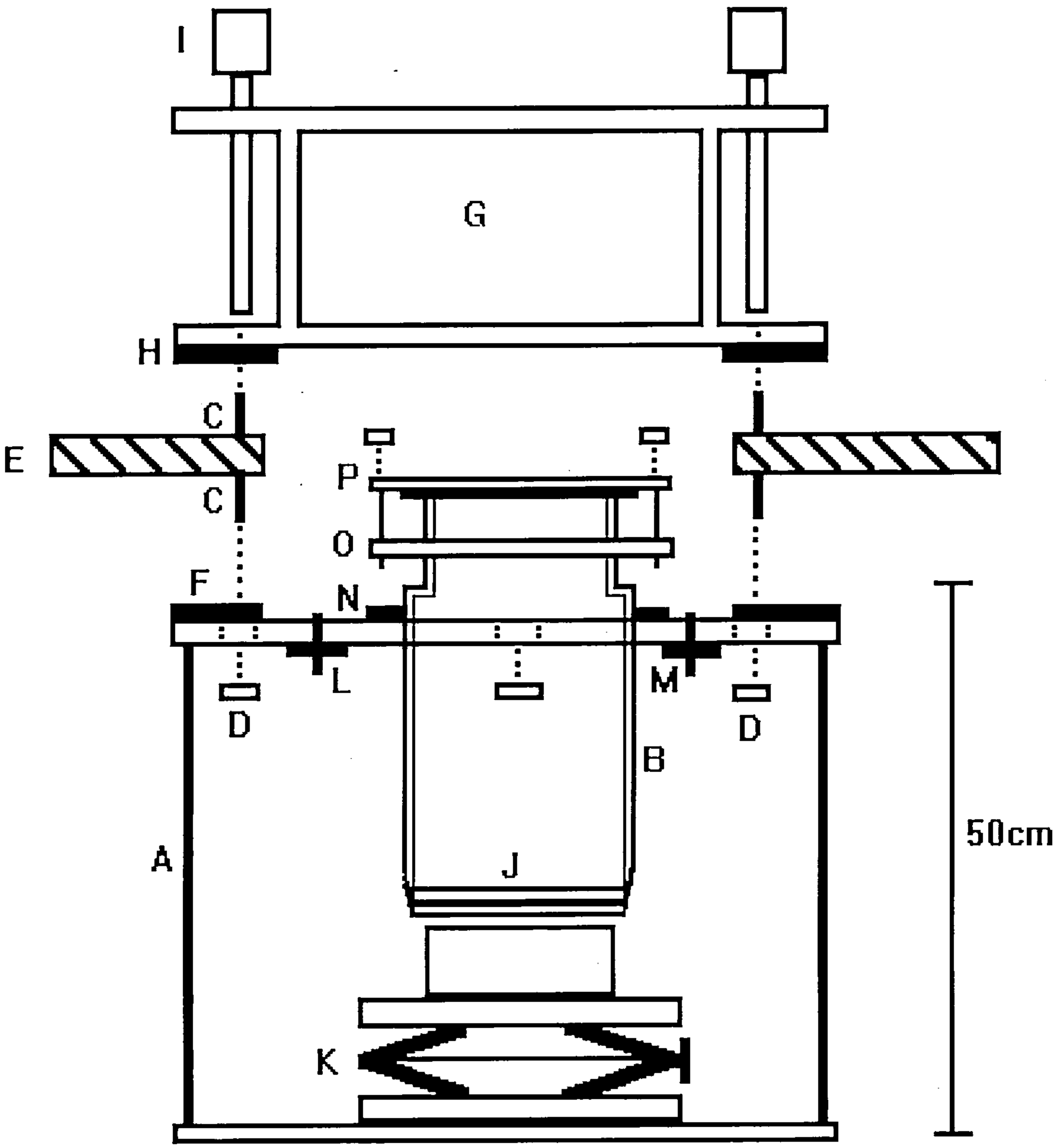

Figure 3.4. Attachment of corer to glove box and jacking cradle.

3.7 Core sectioning

Following the removal of the cover plate, water overlying the core was extracted with disposable pipettes. Care was taken when pipetting to prevent

disturbance to the sediment-water interface. The overlying water was then filtered to remove any particulates (type HA Millipore 0.45µm filter aperture), stored in polypropylene bottles and frozen within one hour of extraction.

By means of the jack, the sediment core was raised within the corer. Successive 1 cm sediment slices were removed using cheese wire and nylon spatulas. Each slice was placed into 200ml polypropylene centrifuge bottles and stored inside the glove box until the end of core handling activities (1 hour maximum). Due to the possibility of oxygen artefacts occurring during the fitting of the piston, the bottom ~5cm of sediment in the core was discarded.

3.8 Porewater extraction

Following removal, via the airlock, of the sediment filled centrifuge bottles the sealed samples were centrifuged at ambient temperature (± 0.5°C) using a refrigerated Sigma 3-series centrifuge at 5500 min^{-1} for 40 minutes to expel the porewater. Once centrifuged, the samples were re-introduced to the inert atmosphere of the glove box. Bottles were then carefully opened, without disturbance, in order to minimise possibility of sediment re-suspension. The porewater was then extracted using disposable 3.5 ml pasteur pipettes, filtered (type HA Millipore 0.45µm filter aperture), transferred to sealable sample vials and maintained in the dark until analysis. All samples reserved for nutrient analysis were immediately frozen and were analysed within two weeks. Following extraction and preparation, the porewater samples maintained in the dark underwent spectrophotometric analysis within three days. Appropriate storage protocols for nutrient samples have been the focus of many studies

(Aminot and Kerouel 1995, Dore et al. 1996, Kirkwood 1992, MacDonald and McLaughlin 1982). The contribution of analytes to the sample from or nutrient adsorption to the sample container is a common problem. Freezing of filtered samples can help combat this problem, especially for PO_4^{3-} and NO_3^-. Although there is evidence that freezing alters nutrient concentrations it is commonly within the precision of analysis (Dore et al. 1996).

3.9 Nutrient analysis

Nutrient analysis of NH_4^+, NO_3^-, NO_2^- and PO_4^{3-} was carried out using a Skalar Sim^{Plus} autoanalyser, following a standard protocol involving segmented flow analysis with colourimetric detection. In this technique, air segments introduced into the sample / reagent lines separate the samples and the samples are mixed with reagent aliquots creating a coloured complex, the absorbance of which is measured spectrophotometrically. Calibration is by using of standard solutions. Precision was determined by the standard deviation of multiple analyses (n = 6) of a standard containing 1 µM total NO^{3-} + NO_2^-, 1 µM PO_4^{3-}, 0.5 µM NO_2^- and 1 µM NH_4^+. Analytical precision was 0.18 NO^{3-} + NO_2^-, 0.2 PO_4^{3-}, 0.02 NO_2^- and 0.3 NH_4^+. The limits of detection of the nutrient analyser used were approximately 50 nM for NO_2^-, 50 nM for NO_3^-, 50 nM for PO_4^{3-} and 100 nM for NH_4^+. Detection limits were assessed by two times the standard deviation of the baseline blank. Following detection the signal was transferred to a computer and interpreted as a peak above that of a Milli-Q water baseline. Milli-Q water is produced via reverse osmosis (Millipore, Milli-RO), and then deionised using a carbon filter cartridge finally irradiated using a UV lamp. Nutrient concentrations were calculated from comparing sample

peaks to those from a series of serial dilutions of a known standard stock. These standards were run before and following sample runs to account for any baseline drift or sensitivity shifts.

Small sample volumes extracted from the porewaters dictated that an autosampler (8 ml sample vial volume) was needed to introduce samples into the reaction manifold. The sediment reservoir is generally regarded as one of the largest nutrient pools in the marine environment with sediment dynamics and dictates that some nutrient concentrations can rapidly increase with sediment depth (Marinelli et al. 1998). Therefore, due to the expected high nutrient concentrations, samples were initially diluted with Milli-Q to prevent saturation of the detectors while maintaining a high sensitivity. Dilution also ensured that possible errors arising from refractive index differences between saltwater and the Milli-Q baseline were eliminated. A salinity correction was therefore not needed. The use of an autosampler also ensured a constant time for reactions to occur and reduced handling time and exposure to the surrounding air, a potential source of contamination (Mee, 1986).

Careful and accurate operation and maintenance of the analyser and associated materials ensured the integrity of the nutrient analyses made during this project. Peristaltic pump tubes were replaced after 50 hours of analyser use. All glassware was thoroughly soaked in a 10% hydrochloric acid and rinsed with Milli-Q water prior to use. All stock solutions, standards and dilutions were made using analytical grade Milli-Q water. Air displacement pipettes were always maintained vertically and regularly gravimetrically checked to prevent contamination between samples and maintain accurate operation. Aminot and Kerouel (1996) identified the inaccurate preparation of calibration solutions to

be a major source of error in nutrient analysis. Therefore, single salt stock solutions were routinely made every six months for PO_4^{3-} (100 mM), every two months for NO_2^- (100 mM), NO_3^- (100 mM) and monthly for NH_4^+ (10 mM). Replacement stock standards were cross-checked with the old stock to eliminate preparation errors. Standard solutions were made daily by serial dilution from stock solutions using Milli-Q water.

3.9.1 NO_2^-

NO_2^- analysis followed the procedure outlined by Hansen and Koroleff (1999). In this, NO_2^- ions react with an acidic sulphanilamide solution forming a diazonium compound, which subsequently reacts with α-naphthylethylenediamine dihydrochloride (NEDD) to form a red/purple azo dye. The dye absorbance was measured at 540nm.

3.9.2 NO_3^-

The sum of NO_3^- and NO_2^- was determination using a modification of the method of Brewer and Riley (1965), which is based on cadmium reduction. Initially NO_3^- ions were reduced to NO_2^- ions using a reduction column. This consisted of a 1 metre copper cadmium wire in an ammonium chloride solution (pH ~ 8.5). The efficiency of the reduction column was periodically checked with comparisons between the respective peak heights of two equimolar NO_3^- and NO_2^- standards. Mean NO_3^- and NO_2^- reduction efficiency was 100.5 ± 1.3%, (n = 6). NO_3^- concentration was obtained by subtracting the NO_2^- concentration from the sum of NO_3^- plus NO_2^- concentration.

3.9.3 PO_4^{3-}

The analytical method is based on determination of soluble reactive phosphorous (i.e. ortho-phosphate) PO_4^{3-} in seawater (Skalar, 1996a). PO_4^{3-} ions react with ammonium molybdate, catalysed by antimony potassium tartrate. The resultant yellow coloured compound is formed and is subsequently reduced by ascorbic acid to a phosphomolybdenum-blue compound. Addition of acetone to the ammonium molybdate reagent prevents absorption of the phosphomolybdenum-blue compound by the system tubing. The compound absorbance is then determined at 880nm using the spectrophotometer. In order to prevent a competitive reaction from silicate ions the pH needs to be <1. The presence of a silicate reaction was tested for by calculating the percentage peak height of 1µM silicate/1µM PO_4^{3-} and was always negligible (<1%). Throughout analysis phosphorous free surfactants were used, for example FFD-6 (Woodward 1994).

3.9.4 NH_4^+

Analysis was by a modified Berthelot salicylate/di-chloro-s-triazine-2, 4, 6-trione sodium salt dihydrate (DDT) method (Skalar, 1996b). NH_4^+ ions are chlorinated with a DDT reagent and tri-sodium-citrate/NaOH buffer at a reaction pH of 10.6. The resulting formation of monochloramine reacts with a sodium nitroprusside/salicylate solution to form an idophenol-blue compound, the absorbance of which was detected at 660nm. Salt precipitation in the systems tubes has been observed in other studies (Laima 1992) but did not compromise the current study as sample dilution with Milli-Q and high ammonium concentrations maintained a high signal to noise ratio.

3.9.5 Dissolved organic matter (DOM)

Spectrophotometric analysis of porewaters at 350nm detects DOM in the form of Chromophoric dissolved organic matter (CDOM) (Kitidis 2002). Light induced transformations can act to accelerate photochemical changes of DOM (Tranvik and Bertilsson 2001). Nelson and Guarda (1995) advocate that the effect of bacterial degradation on organic matter is negligible if samples are maintained in the dark for periods < 4 days. Therefore spectrophotometric analysis followed porewater extraction, however, as the samples had been refrigerated during centrifugation, the samples were stored for a period (> 24, but < 36 hours) in the dark to thermally equilibrate, thus preventing any anomalies due to differences in thermal artefacts between the reference cell and sample. The UV spectra were obtained with a double beam UV-visible UviKon 923, spectrophotometer (Kontron Instruments, range = 190-900 nm) at room temperature. The limits of detection were 0.001 absorbance units, giving a resultant absorption coefficient of 0.043 m^{-1} for 10mm pathlengh cuvettes. All samples were analysed using high optical density, 10mm quartz cuvettes. Prior to analysis of samples, a blank reference spectrum containing analytical grade Milli-Q water was obtained. To account for Raman scattering by water molecules, the reference blank was subtracted from all samples to set the baseline value (Green and Blough, 1994). Sample spectra were examined over a wavelength range of 250-800nm. Consequently, the absorption coefficient at 350nm could be calculated.

Chapter 4:

The impact of trawling on inorganic nutrient porewater profiles in the central west North Sea.

4.1 Abstract

The effects of trawl disturbance on porewater nutrient profiles were investigated in the central west North Sea. Sediment cores from trawled and untrawled sites were examined monthly for one year (April 2001 – March 2002) with additional cores taken directly behind and in the track of a commercial trawler. Concentrations of dissolved constituents (NO_2^-, NO_3^-, PO_4^{3-}, NH_4^+) and CDOM in porewaters, and sediment properties (% organic content, grain size and porosity) were quantified. During the main fishing season (October – March inclusive) significant differences in nutrient concentrations existed in the top 4 cm of the sediment between trawled and untrawled areas. Porewaters from the trawled site displayed distinct zones of surface homogenisation with relatively low nutrient concentrations. Below 5 cm nutrient concentrations increased with depth. The gradient of increasing NH_4^+ and PO_4^{3-} with depth was greater in untrawled compared to trawled sediments, with immediately trawled sediments displaying the highest concentrations and steeper gradients. Immediately following a trawl disturbance, surficial NO_2^- and NO_3^- concentrations were greatly enhanced, although they declined rapidly with depth. Recovery of nutrient concentrations following trawl disturbance through diffusion could not account for NH_4^+ and PO_4^{3-} profiles. However, the formation of a temporary nepheloid layer and other biogeochemical processes are discussed as mechanisms for exposing what were deep anoxic sediments to oxygen rich

surface waters following sediment excavation by trawl gear and the consequential alteration of benthic nutrient dynamics.

4.2 Introduction

In shallow estuarine and intertidal environments, exchange of particles and solutes between surficial sediments and the overlying water column can affect nitrogen and oxygen cycling (Lohse et al. 1993). Such sediment-water interactions are important biogeochemical pathways that can affect whole system processes, including primary production (Bertuzzi et al. 1997). Shallow water and estuarine areas often have relatively high concentrations of particulate organic matter (POM) incorporated into the sediment (Nedwell et al. 1999). While dependant on several physical factors and the varying time lags between burial and regeneration, a significant fraction of POM is ultimately remineralised (Seiki et al. 1991). Benthic remineralisation of POM has important impacts on water-column processes (Rowe et al. 1975).

Organic matter deposition creates an oxygen demand in the benthic layer due to respiration by aerobic bacteria and oxidation of reduced components produced by anaerobic metabolism (Froelich et al. 1979). Sedimentary remineralisation generates NH_4^+ and PO_4^{3-} that once released, via diffusion or physical mixing to overlying waters, provide bio-available nutrients critical for phytoplankton growth (Nixon et al. 1976, Rowe et al. 1977). Physical mixing can create constantly changing redox boundaries and development of micro-niche environments that enable a constant coupling between aerobic and anaerobic dependent processes. Altered redox conditions are likely to effect organic degradation and nutrient mineralization rates. Logistically these

environments can provide readily available study sites, where samples can be taken at low tide without disturbing the sediment surface.

However, in contrast to estuarine and intertidal environments, shelf sea sediments, while being recognised as areas of high nutrient remineralisation (Rutgers Van Der Loeff 1980a), are an understudied component of nutrient cycling. These sediments potentially play a critical role in structuring and controlling water-column nutrient dynamics. The transformation of organic debris into dissolved inorganic nutrients within shelf sediments creates steep concentration gradients between the sediment and water-column through which nutrients diffuse (Hammond et al. 1985). Temperate shelf seas typically display a regular seasonal cycle of natural physical, chemical and biological processes that control rates of nutrient regeneration and release (Rutgers Van Der Loeff 1980b).

Seasonal fluctuations in the relative amount of organic matter transported to shelf sediments, temperature variations and natural disturbance can control the remineralisation of dissolved constituents (Parsons et al. 1984). Deposition rates dictate the amount of POM available for remineralisation while temperature and oxygen influence the rate of transformations. Storms and increased wave action can increase the physical disturbance to the benthos and alter O_2 input and nutrient exchange during winter (Fanning et al. 1982). It is reasonable to predict that NH_4^+ and PO_4^{3-} would exhibit relatively low concentrations within surficial porewaters and increase with depth through anaerobic degradation of organic matter (Nedwell et al. 1999). In contrast, a steady decline of both NO_2^- and NO_3^- in sediment porewaters could be expected

due to utilisation of O_2 at increasing depth within the sediment (Ogilvie et al. 1997).

Alterations to the rate of organic matter transformation and release of dissolved constituents from this benthic 'reservoir' of nutrients may have local and wider reaching consequences to primary production. The North Sea with its high level of trawling activity is of considerable interest in this regard since the impact of trawling could effectively alter the chemical and biological properties of sediments. The direct physical disturbance to the seabed caused by the fishing gear effectively mobilises the sediment surface, releasing bound up nutrients (Percival and Frid 2000). Fanning et al (1982) calculated sediment nutrient release from the Gulf of Mexico could be augmented by > 200%, following the resuspension of the surface 1mm of sediment by storm disturbance. These large, pulsed, releases of nutrients were shown to accelerate nutrient turnover and increase primary productivity (Fanning et al. 1982). Physical disruption to sediments also alters redox conditions and mixes organic matter and O_2 rich water deeper into sediments (Pilskaln et al. 1998). As a consequence of high trawl activity, such sediments may therefore end up in a permanent state of perturbation where nutrient regeneration and release rates are altered (chapter 5).

In areas of trawl activity, large benthic organisms are susceptible to relatively high mortality rates (Lindeboom and de Groot 1998). Consequently, within areas of high fishing effort the benthic species that persist are trawl resistant. This term refers to organisms that can cope with the repeated direct physical impacts. Faunal assemblages within these environments tend to be dominated by species with a smaller body size and higher reproductive output

than species in undisturbed areas (Jennings et al. 2001). The altered biological activity associated with this modified faunal assemblage may further change nutrient dynamics.

This chapter examines benthic nutrient dynamics within untrawled sediments from the North Sea and compares them to trawled sediments with similar sediment characteristics over a full annual cycle. Finally this chapter aims to examine porewater profiles immediately following trawling and compare them to untrawled and trawled sediment profiles to demonstrate any impact on porewater chemistry from trawling.

4.3 Methods

Two sites in the North Sea, approximately 10 km off England's north east coast were sampled monthly from April 2001 to March 2002 (Fig 4.1). This region of the North Sea receives relatively low inputs of anthropogenic nutrients (Clark and Frid 2001). Both areas were located within a heavily fished area, ICES statistical sub-rectangle 39E8 (annual average = >35,000 hours fishing effort, 1997-2001 inclusive, data provided by Department for Environment, Food and Rural Affairs). Isolated within 39E8 was a smaller area that remained untrawled, yet was a viable site for comparison as it consisted of sediment with identical characteristics as the trawled area. This area remained untrawled due to the presence of a wreck and rocky reef that effectively enclosed an area too small for the deployment of commercial towed fishing gear.

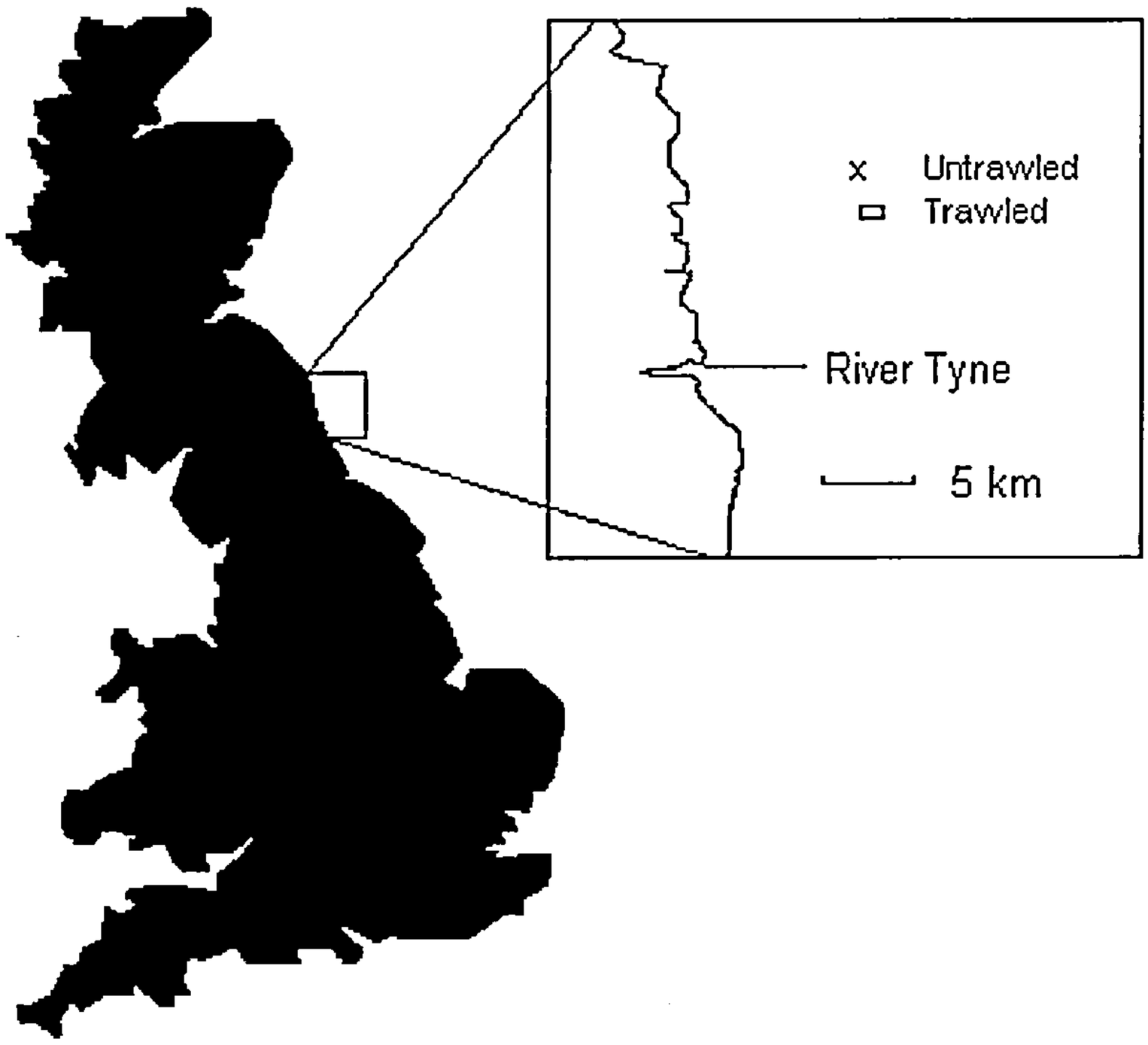

Figure 4.1. Coastal outline of north-east England and sampling sites within the central-west North Sea. Untrawled site (x) and trawled area (box) illustrated (untrawled = 55^{0}12.64'N 01^{0}27.20'W, trawled area centred on = 55^{0}13.55'N 01^{0}27.28'W).

A sediment core from the trawled and untrawled sites was taken monthly for a year (for full details Chapter 3). Trawled sediments however, were collected from a known fishing ground but where the timing of trawling event was unknown. An Additional of sample was taken in February 2002, at a time of high fishing effort (Appendix 1). These latter samples comprised a core from the untrawled and trawled sites and a sample taken behind and in the track of a commercial trawler. Immediately trawled sediments were ~ 15 mins after fishing. It was critical to take immediately trawled samples early in the calendar year. (i.e. February), because at this time relatively low insolation and reduced temperatures result in low levels of primary production. Consequently benthic productivity is also relatively low as little POM is transported to the benthos (Parsons et al. 1984). At this time benthic macrofaunal activity is also reduced,

which would otherwise lead to increased bioturbation, with subsequent impacts on benthic nutrient cycling (Widdicombe and Austen 1998). Thereby, reducing the need to disentangle inherent physical and chemical processes and thus isolating the specific impact of trawling.

Cores were taken with a modified corer capable of retrieving undisturbed cores at depth (for full details Chapter 3). Once collected, cores were placed in a water bath maintained at *in situ* bottom temperature and returned to the laboratory for immediate analysis. Subsequent core handling was in an inert atmosphere of N_2 within a glove box. Water overlying the core was extracted using disposable pipettes. Care was taken when pipetting to prevent disturbance to the sediment-water interface. The overlying water was then filtered to remove any particulates (0.45µm, type HA, Millipore) and stored in polypropylene bottles prior to nutrient analysis. By means of a jack and piston, the sediment core was raised within the corer. Successive sediment slices were removed using cheese wire and nylon spatulas. Each slice was placed into 200ml polypropylene centrifuge bottles. In order to preclude the possibility of oxygen ingress during the raising of the core piston, the bottom ~5cm of sediment was always discarded.

The sealed sample bottles were centrifuged at 5500 revolutions min^{-1} for 40 minutes to expel the porewater (Sigma 3-series centrifuge). The bottles were then re-introduced to the inert atmosphere of the glove box and carefully opened in order to preclude disturbance to the sediment-water interface. The porewater was then extracted using disposable pipettes and transferred to sealable sample vials. Following extraction and preparation, porewaters underwent nutrient and spectrophotometric analysis.

4.3.1 Nutrient analysis

Concentrations of NO_2^-, NO_3^-, NH_4^+ and PO_4^{3-} were determined with an automated nutrient analyser (Skalar Sim^{Plus}), following standard protocols (Brewer and Riley 1965; Mantoura and Woodward 1983; Kirkwood 1989). Analytical precisions ± 1% were routinely achieved for all methods (see chapter 3 for full details).

4.3.2 Spectrophotometric analysis

UV spectra were obtained with a double beam UV-visible spectrophotometer (UviKon 923, Kontron Instruments, range = 190-900 nm) at room temperature, following standard protocols (for full details chapter 3). Sample spectra were examined over a wavelength range 250-800nm enabling the absorption coefficient at 350nm to be calculated for quantification of chromophoric dissolved organic matter (CDOM). The limits of detection were 0.001 absorbance units, giving a resultant absorption coefficient of 0.043 m^{-1} for 10mm pathlengh cuvettes.

4.3.3 Seasonal profiling

Following nutrient and spectrophotometric analysis, NO_2^-, NO_3^-, NH_4^+ PO_4^{3-} and CDOM results were profiled for trawled and untrawled sediments for each sampling occasion. Porewater profiles were subsequently categorised by season (based on air temperature) (winter = December, January and February; spring = March, April and May; summer = June, July and August and autumn = September, October and November), depth zone (above and below 4 cm) and

trawled or untrawled. This allowed each factor to be statistically assessed using a General Linear Model (GLM) with subsequent Tukey pairwise comparisons.

Profiles generated from the cores taken during February to give a comparison of untrawled and trawled against immediately trawled sediment porewaters were analysed using a one-tailed z test.

4.4 Results

4.4.1 Sediment characteristics

Sediments collected from each site did not vary significantly in porosity (%) (Fig 4.2), grain size (median particle size = 3.5 phi, (Wentworth 1922) or percentage organic content (trawled sediment organic content mean = 3.2 ± 1.0 % and untrawled sediment organic content mean = 2.8 ± 0.8 %; Kruskal-Wallis, W = 6, p = 0.081) between systems.

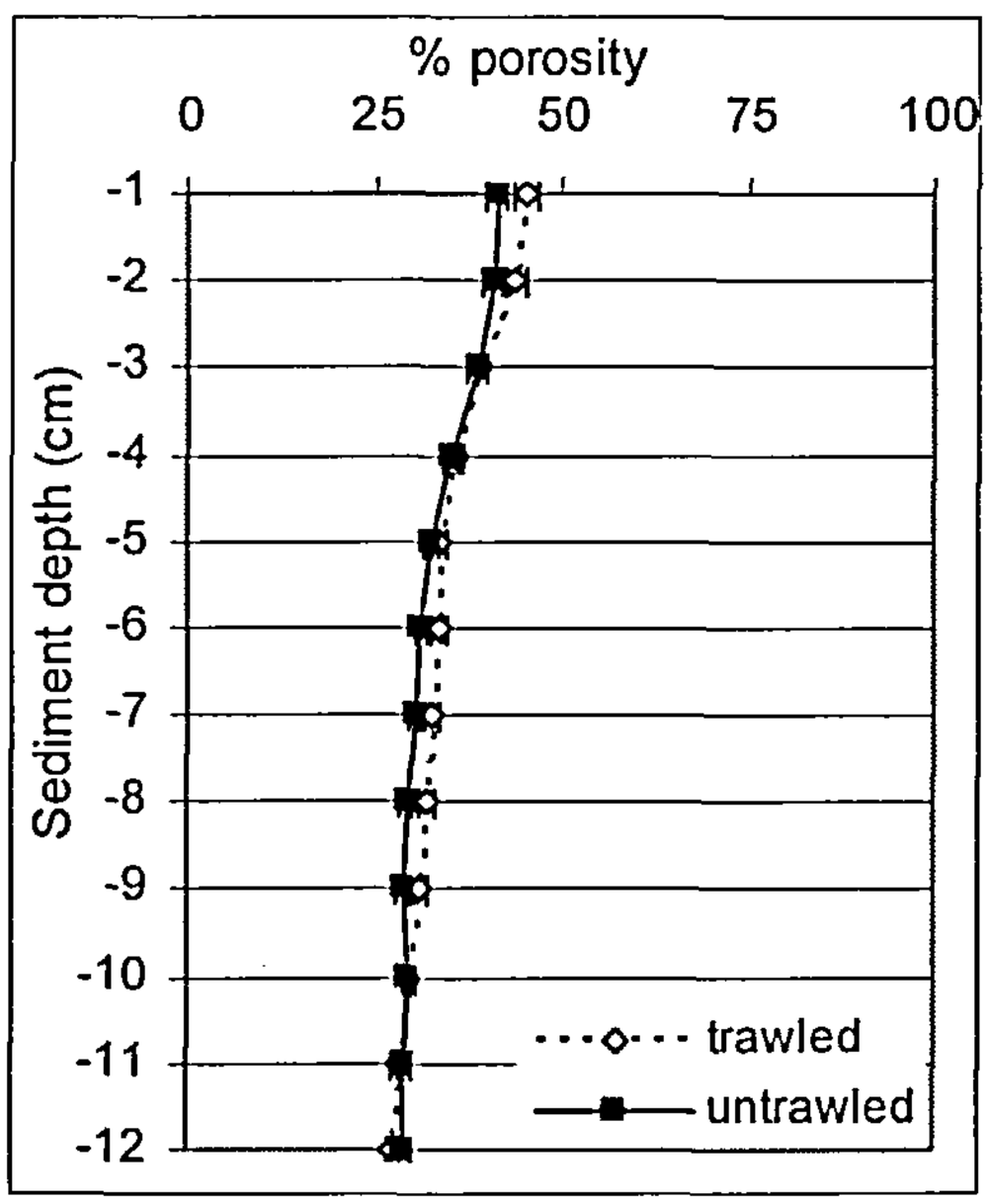

Figure 4.2. Percentage porosity profiles from trawled (broken line and diamond) and untrawled (line and solid square) sites.

4.4.2 Seasonal porewater profiling

Porewater profiles of NO_2^-, NO_3^-, NH_4^+, PO_4^{3-} and CDOM are summarised in Table 4.1. Throughout the year porewater NO_2^- concentrations from trawled and untrawled sediments were enhanced compared to the overlying water (Fig 4.3). Within untrawled sediments NO_2^- varied seasonally (~ 0.5 - ~ 4µmol L^{-1}), with the highest concentrations occurring during April. Untrawled NO_2^- concentrations at this time were significantly greater than NO_2^- concentrations within the trawled core during spring (GLM Tukey, untrawled spring vs trawled spring, $T = 5.8635$, $P < 0.001$). Clear monthly distinctions between NO_2^- concentrations within trawled sediments however, only extended from January to August. NO_2^- within trawled porewaters exhibited a homogenous distribution in September through December (Fig 4.3). Trawled sediments exhibited the highest NO_2^- during the summer (~ 3µmol L^{-1} during July) and summer concentrations were significantly different to winter and spring concentrations (GLM Tukey, trawled summer vs winter, $T = 4.0908$, $P < 0.001$; trawled summer vs spring, $T = 3.3585$, $P < 0.05$).

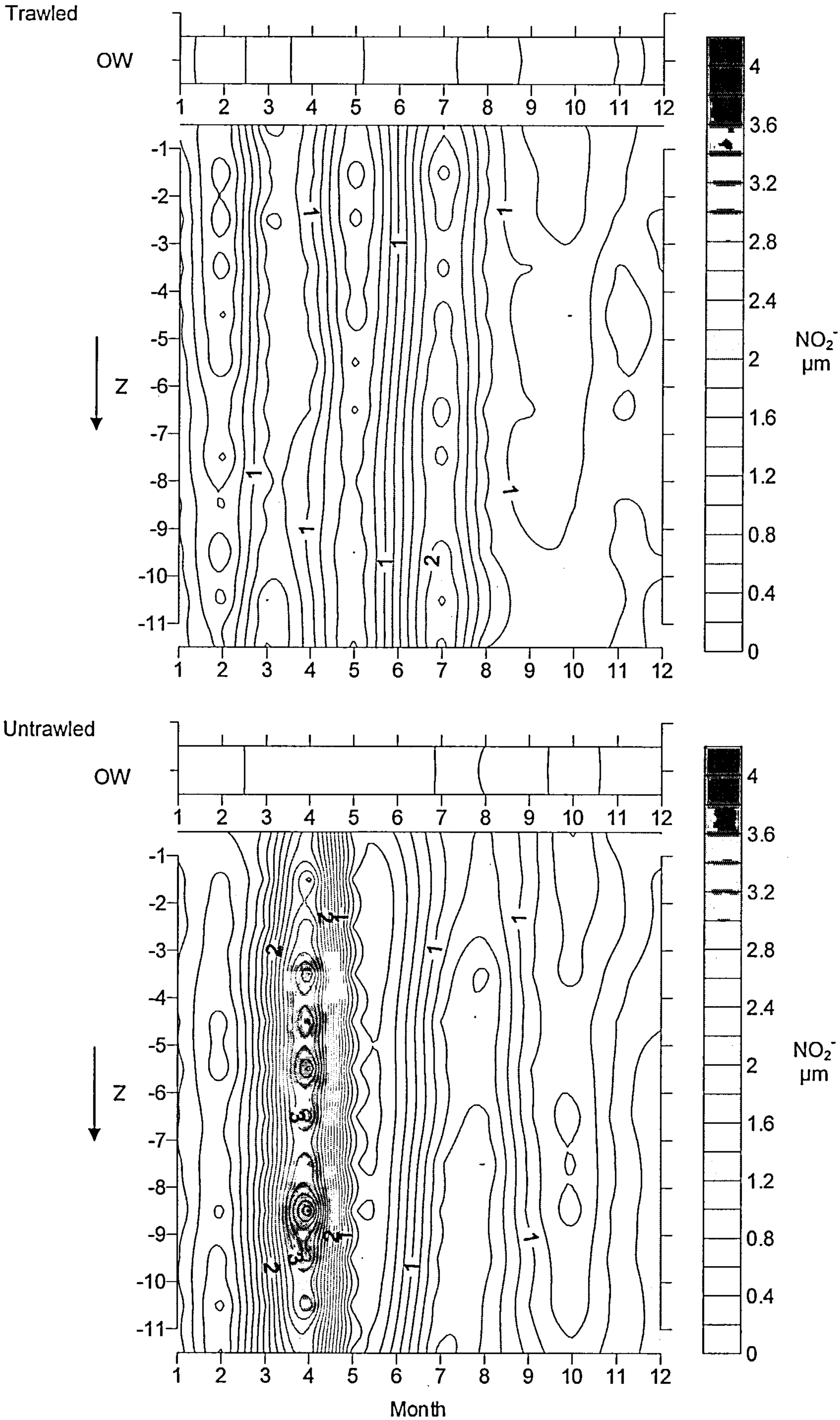

Figure 4.3. Seasonal sediment porewater and overlying water (OW) profiles of NO_2^- from trawled and untrawled sediments within ICES statistical rectangle 39 E8. Z = depth (cm).

NO_3^- within the overlying waters was elevated compared to porewaters for trawled and untrawled sediments throughout the year (Fig 4.4). Untrawled sediments displayed discrete micro-niches of increased NO_3^- porewater concentration between September and June. Throughout this period surficial concentrations were greater than sub surface (>4 cm) levels. During July and August however, low NO_3^- concentrations were present throughout the entire depth profile. Within trawled sediments NO_3^- was elevated within surficial sediments (~ 3 cm) from October through to May (Fig 4.4). Surface NO_3^- was significantly different to concentrations below 4 cm (GLM Tukey, depth > 4 cm vs depth < 4 cm, $T = -5.069$, $P < 0.001$). From June through to October profiles of porewater NO_3^- were un-stratified and maintained a relatively low concentration.

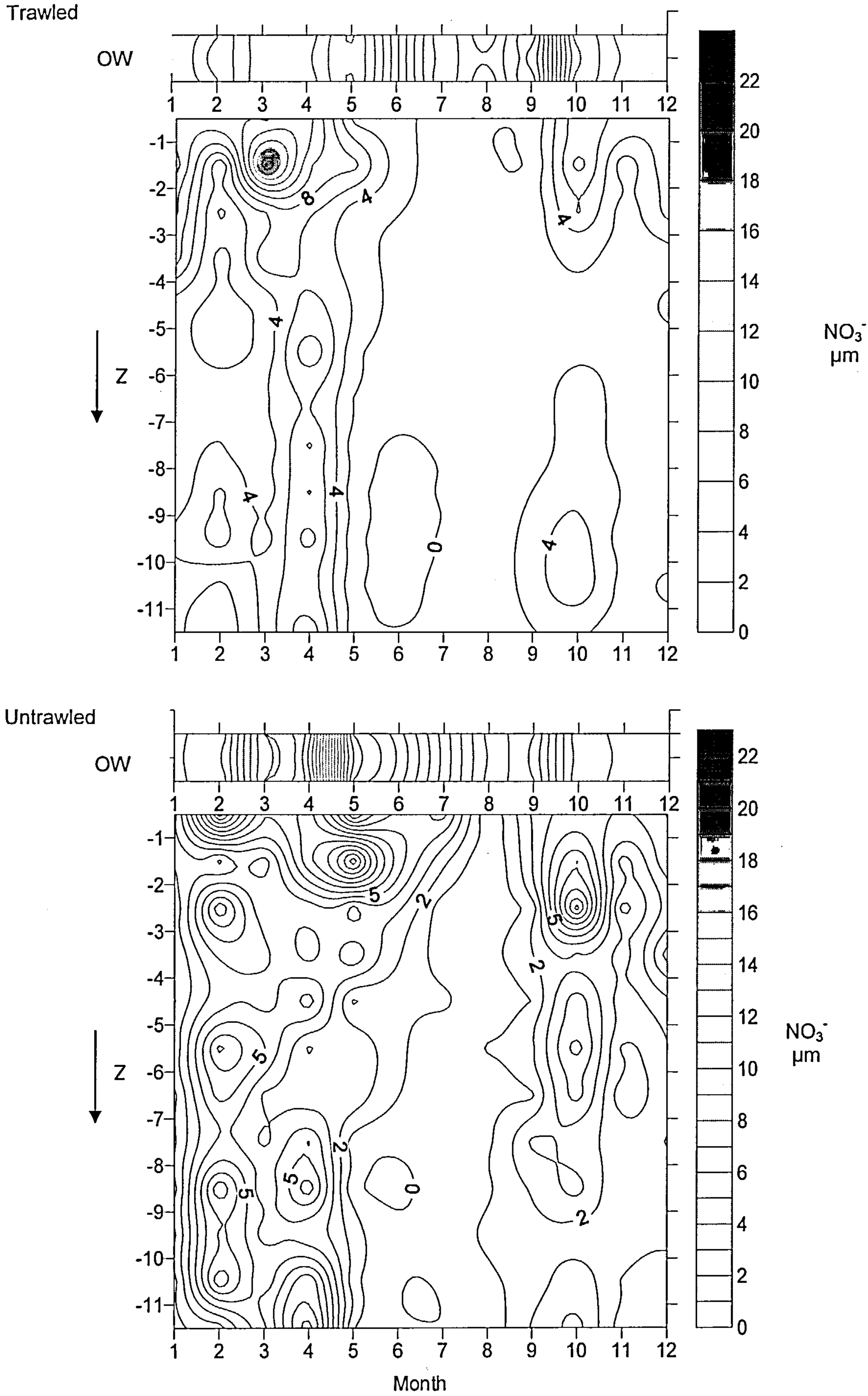

Figure 4.4. Seasonal sediment porewater and overlying water (OW) profiles of NO_3^- from trawled and untrawled sediments within ICES statistical rectangle 39 E8. Z = depth (cm).

Porewater PO_4^{3-} from trawled and untrawled sediments was greater than overlying water concentrations throughout the year (Fig 4.5). The interstitial waters of both sediments displayed a trend of increasing concentration with sediment depth (Fig 4.5). Untrawled sediments, between September and May were vertically stratified. High PO_4^{3-} concentrations occurred in the untrawled samples during July. In contrast to untrawled sediments, trawled sediments displayed reduced vertical stratification and the low PO_4^{3-} levels during the summer (Fig 4.5). As a result, summer PO_4^{3-} concentrations were significantly different to concentrations during all other seasons (GLM Tukey, summer vs winter, $T = 4.2401$, $P < 0.001$; summer vs spring, $T = 3.9385$, $P < 0.001$; summer vs autumn, $T = -53.565$, $P < 0.05$). Trawled samples exhibited a homogenised surface layer down to ~ 4 cm with relatively low PO_4^{3-} concentrations. Below this depth interstitial PO_4^{3-} was more stratified and significantly greater than surface concentrations (GLM Tukey, depth > 4 cm vs depth < 4 cm, $T = 5.895$, $P < 0.001$).

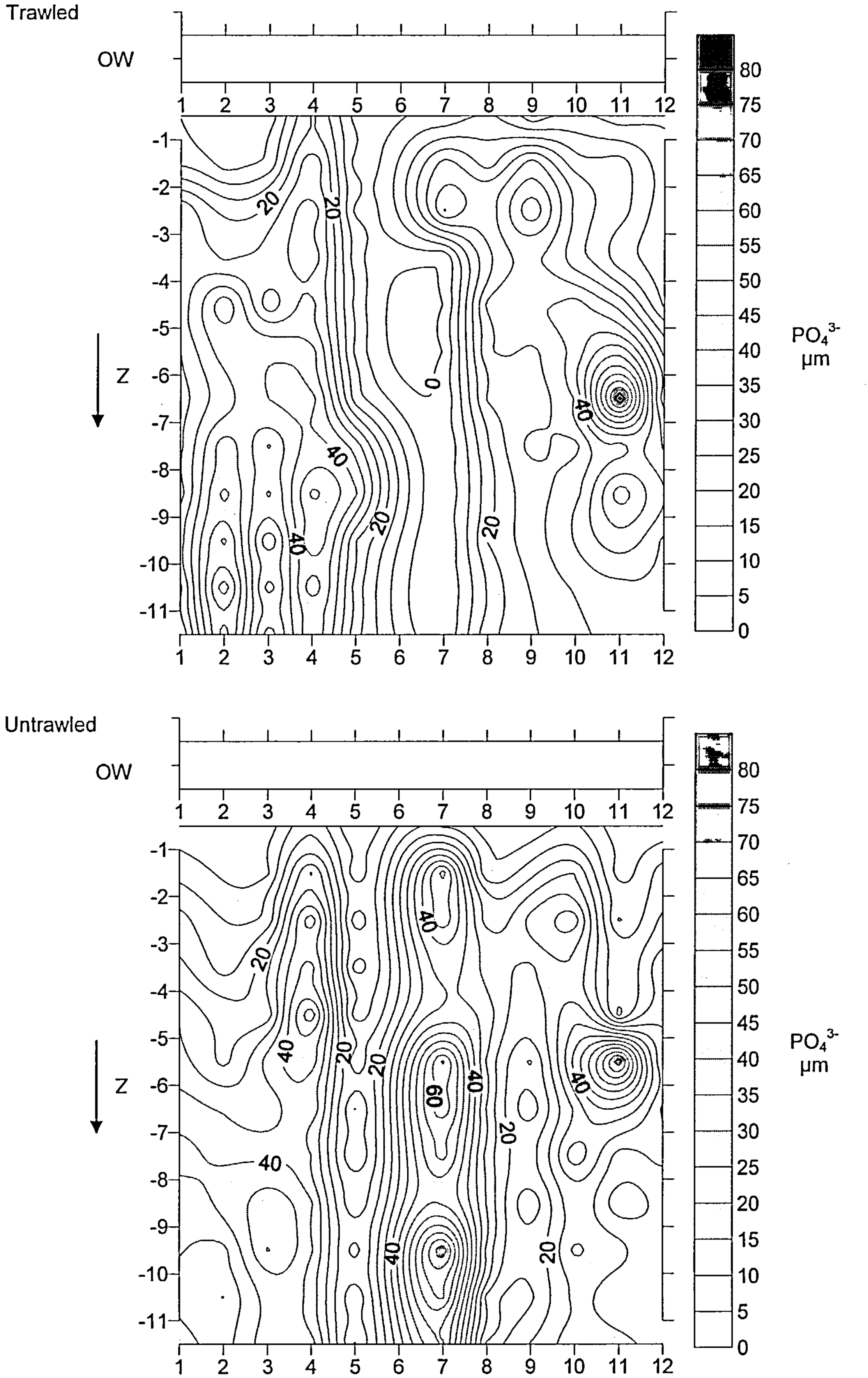

Figure 4.5. Seasonal sediment porewater and overlying water (OW) profiles of PO_4^{3-} from trawled and untrawled sediments within ICES statistical rectangle 39 E8. Z = depth (cm).

NH_4^+ in the overlying water was lower than porewater concentrations for untrawled and trawled sediments (Fig 4.6). Generally NH_4^+ increased with depth for both sediment types. A greater degree of monthly variation of porewater NH_4^+ occurred in untrawled sediments. Winter and spring however were the only seasons to display significant differences in NH_4^+ between depths (GLM Tukey, winter depth > 4 cm vs depth < 4 cm, $T = 4.7159$, $P < 0.001$; spring depth > 4 cm vs depth < 4 cm, $T = 4.3698$, $P < 0.05$). Trawled sediments displayed a homogenous surface layer (~ 4 - 5 cm) with relatively lower NH_4^+ concentrations compared to untrawled sediments (Fig 4.6). Below this depth however, trawled sediments increased in concentration above those exhibited by untrawled sediments. Concentration of NH_4^+ were significantly different over depth during each season for trawled porewater profiles (GLM Tukey, winter depth > 4 cm vs depth < 4 cm, $T = 4.368$, $P < 0.05$; spring depth > 4 cm vs depth < 4 cm, $T = 6.194$, $P < 0.001$; summer depth > 4 cm vs depth < 4 cm, $T = 3.4981$, $P < 0.05$; autumn depth > 4 cm vs depth < 4 cm, $T = 7.441$, $P < 0.001$).

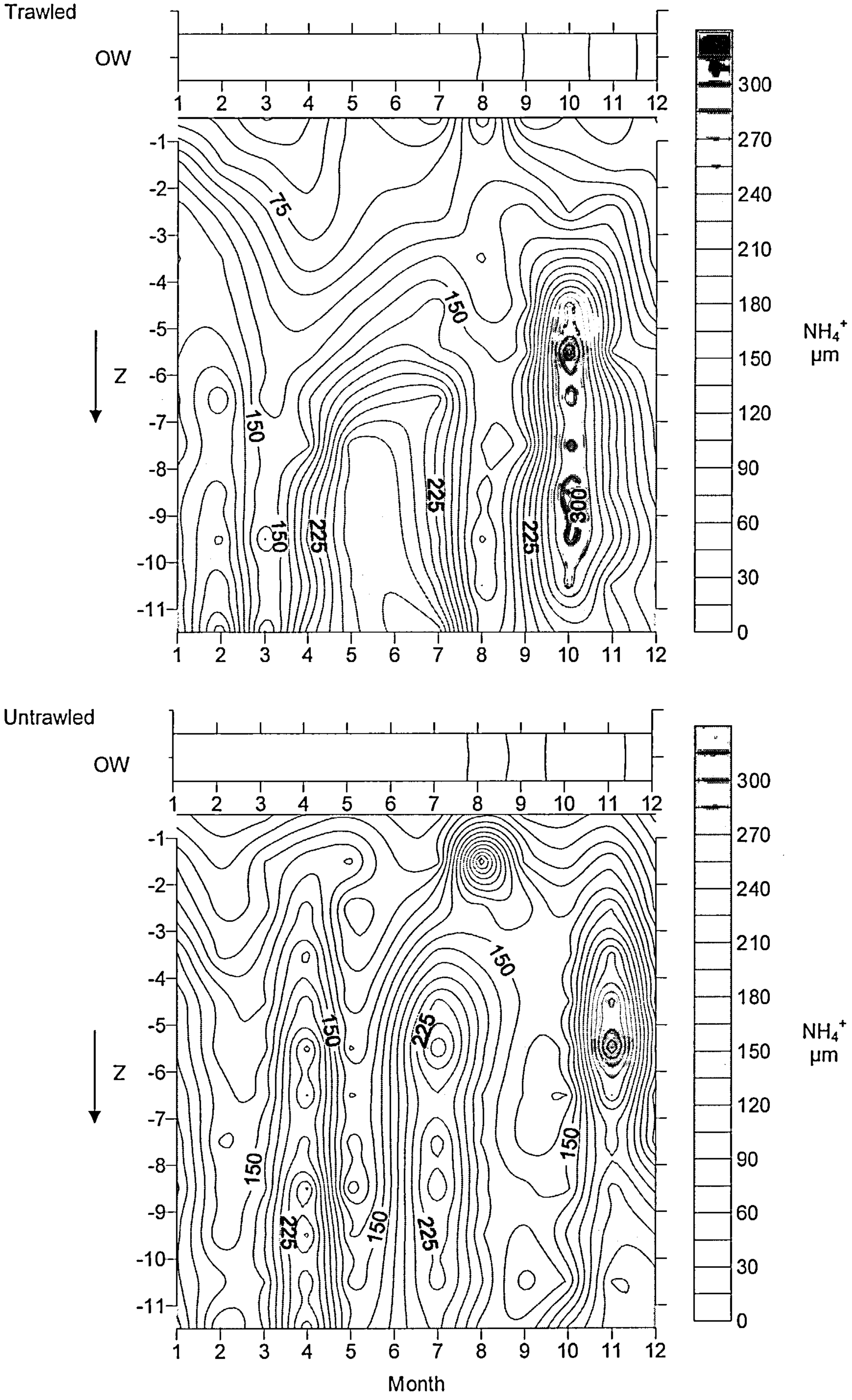

Figure 4.6. Seasonal sediment porewater and overlying water (OW) profiles of NH_4^+ from trawled and untrawled sediments within ICES statistical rectangle 39 E8. Z = depth (cm).

While a comparable pattern of CDOM was present within the porewaters of trawled and untrawled sediments, the magnitude of, and depth changes in CDOM varied (Fig 4.7). CDOM within the overlying water was consistently lower than in untrawled and trawled sediment porewaters. Near surface (< 2cm) porewaters of all samples also typically displayed low CDOM levels (Fig 4.7). Below the surface the level of CDOM increased rapidly with increasing depth reaching a maximum between 3 and 7 cm. After reaching the CDOM maximum, levels generally declined with depth throughout the remaining core porewaters.

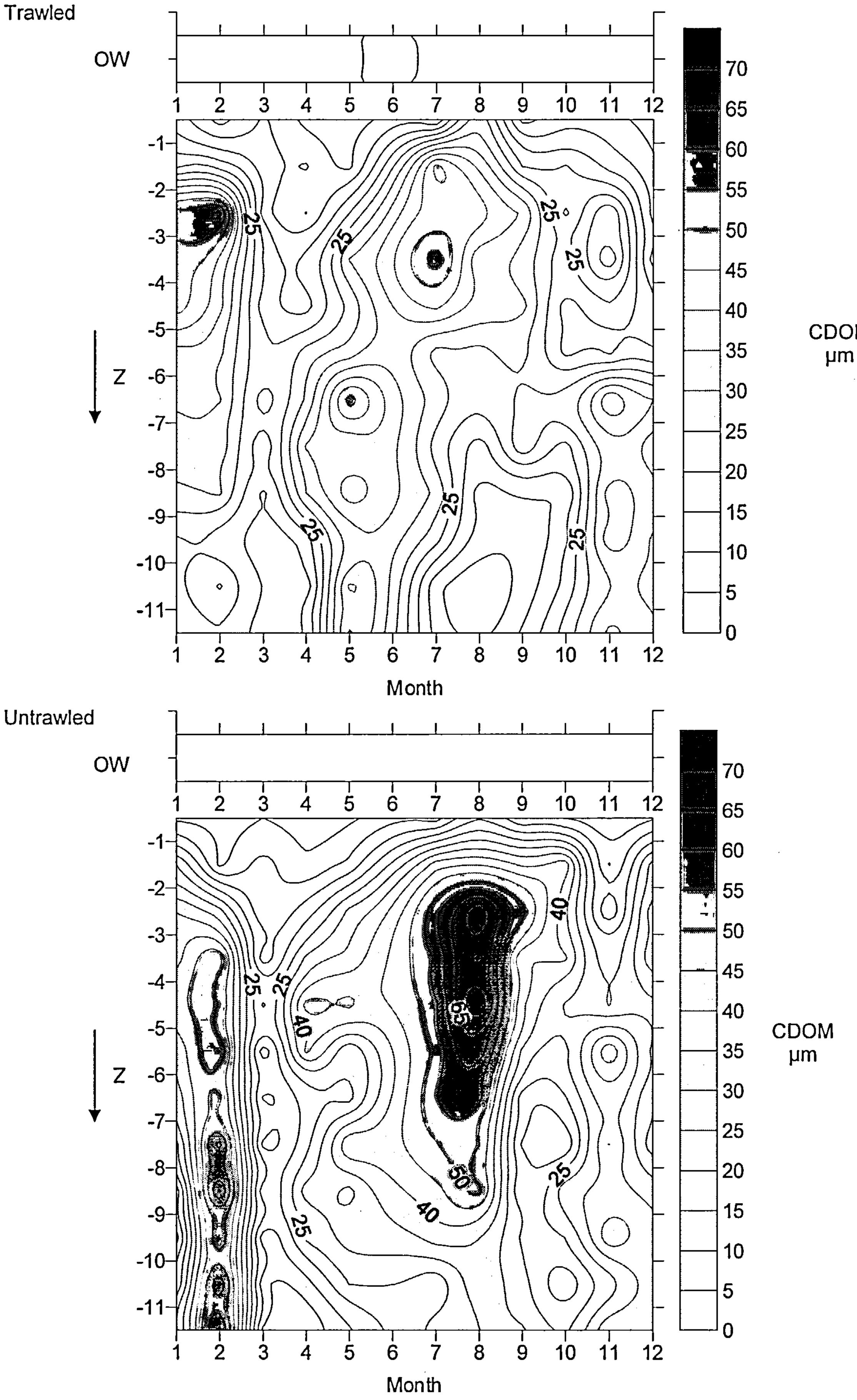

Figure 4.7. Seasonal sediment porewater and overlying water (OW) profiles of CDOM from trawled and untrawled sediments within ICES statistical rectangle 39 E8. Z = depth (cm).

Table 4.1. Summary tables of trawled (a) and untrawled (b) porewater profiles, plus overlying water, for NO_2^-, NO_3^-, NH_4^+, PO_4^{3-} and CDOM.

a

Trawled					
Nutrient	Overlying water		Porewater		
	Seasonal	Higher / lower than porewater	Seasonal	Peak	Stratified? where / when
NO_2^-	No	Lower	Not markedly	Jul / Aug	No
NO_3^-	Yes	Higher	Yes	Apr	Yes, ~ 3 cm, Oct - May
PO_4^{3-}	No	Lower	Yes	Nov	Yes, 4 cm, Aug - May
NH_4^+	No	Lower	Yes	Oct	Yes, ~ 4 – 5 cm, Jan - Dec
CDOM	No	Lower	No	Feb / May / Aug	Yes, ~ 2cm, seasonally

b

Untrawled					
Nutrient	Overlying water		Porewater		
	Seasonal	Higher / lower than porewater	Seasonal	Peak	Stratified? where / when
NO_2^-	No	Lower	Yes	Apr	No
NO_3^-	Yes.	Higher	No	Feb	Yes, ~ 4 cm Sep - Jun
PO_4^{3-}	No	Lower	Yes	Jul	Some, down to 11cm, Jan - Feb
NH_4^+	No	Lower	Yes.	Nov	Yes, down to 11 cm, Feb & May
CDOM	No	Lower	No	Feb / May / Aug	Yes, ~ 2cm, seasonally

4.4.3 Comparison of untrawled, trawled and immediately post trawled sediments

A general pattern of increasing concentration with depth was apparent for NH_4^+ and PO_4^{3-} within untrawled, trawled and immediately trawled sediments (Fig 4.8). The porewater concentration of NH_4^+ and PO_4^{3-} was greatest in immediately trawled and least in untrawled sediments. NH_4^+ and PO_4^{3-} within immediately trawled porewaters was significantly different to trawled and untrawled porewaters at every depth (Appendix 2). While there were differences in porewater NH_4^+ and PO_4^{3-} between the three sediments, corresponding concentrations within the overlying water were similar to surficial sediment porewater concentrations. In contrast, NO_2^- and NO_3^- concentrations in the overlying water were enhanced immediately following trawling (Fig 4.8), and were significantly different to the water overlying trawled and untrawled sediments (Appendix 2). NO_2^- and NO_3^- perturbations occurred within the top 2 – 3 cm of the sediment between untrawled, trawled and immediately trawled sediments, with NO_3^- being significantly different at 2 cm (Appendix 2). Both trawled and immediately trawled concentrations were greater than in the untrawled sediments (Fig 4.8). Below 3 cm NO_2^- and NO_3^- concentrations within the three sediments were relatively similar.

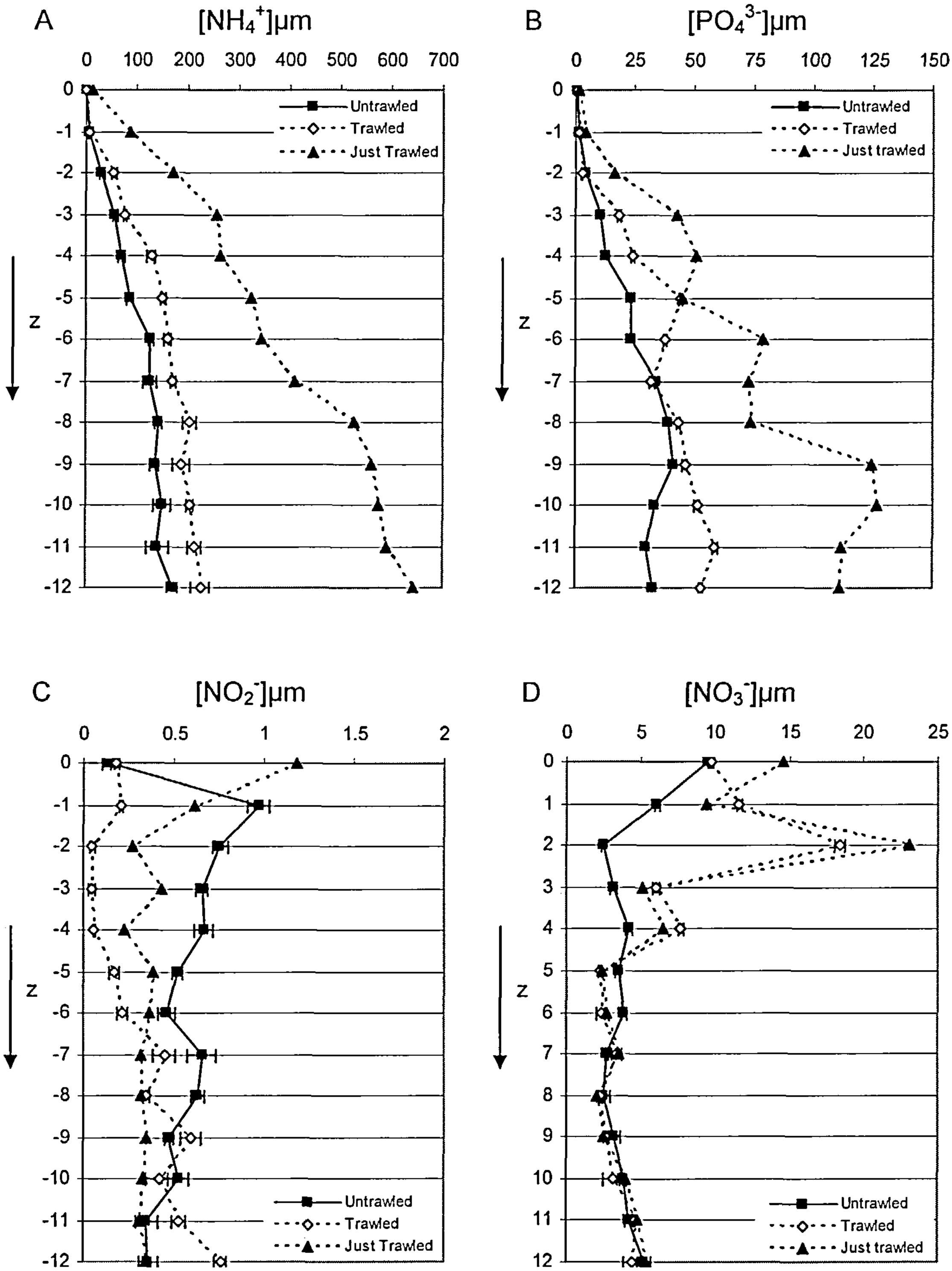

Figure 4.8. Comparison of porewater nutrient concentrations within untrawled (line and solid square), trawled (broken line and diamond) and immediately trawled (broken line and solid triangle) areas (± standard deviation). Stoiciometry of dissolved constituents, NH_4^+ (A), PO_4^{3-} (B), NO_2^- (C) and NO_3^- (D). All cores were collected on 19/2/2002 from untrawled (55^0:12.66N; 001^0:27.23W), trawled (55^0:13.54N; 001^0:27.10W) and immediately trawled sediments (approximately 15-30 minutes after the ground was trawled by a commercial trawler at 55^0:13.58N; 001^0:26.72W). Z = depth (cm).

4.5 Discussion

Notwithstanding seasonal variability at each site, it is clear that there are distinct differences between the nutrient profiles for each site (Fig 4.8). Porewater constituents from trawled and untrawled sediments displayed vertical stratification and seasonal variability. Benthic environments within temperate regions are likely to respond to certain key factors including; direct human induced disturbance, natural physical disturbance, O_2 input and the depth of its penetration, temperature, POM transport to the sediment and size specific bioturbation and macrofaunal mortality that influence biogeochemical processes (Oppenheimer 1960, Parsons et al. 1984). These are difficult to disentangle in the field considering marine sediments have the ability to differ biogenically and chemically over relatively small spatial and temporal scales (Rhoads 1974, Hines et al. 1982). In this study the proximity of the sites and lack of any differences in sediment characteristics should ensure that these natural factors occurred at both sites to the same extent. Therefore, any differences are likely to occur from direct trawl impacts or indirectly through altered faunal assemblages.

NO_3^- concentrations within surficial interstitial waters of trawled sediments were elevated during winter and spring, the periods of high fishing effort (Fig 4.4 and 4.9). Trawling is likely to have extended the penetration of oxygenated bottom water through the direct ploughing action of ground gear (Mayer et al. 1991, Pilskaln et al. 1998). However, during June through September, when bottom temperatures were high and fishing effort was reduced (Fig 4.9), NO_3^- was barely detected in trawled sediments. Summer temperatures during this period are likely to increase the demand for O_2 through

enhanced microbial oxidative metabolism (Clavero et al. 2000). As a result anaerobic conditions may dominate because any available O_2 would be quickly utilised through intense heterotrophic activity (Clavero et al. 2000). Under such circumstances other electron acceptors, including NO_3^-, are utilised (Berner 1980). The elevated NO_3^- concentrations in the overlying water during the summer, compared to porewater concentrations, also supports the idea that nitrate respiration occurred in the sediment as this process would continually transform and thus remove NO_3^-. Furthermore, trawling can reduce the natural level of sediment reworking (bioturbation) through size specific mortality of the benthos (see Chapters 2 and 6). Notwithstanding the periodic pulse of O_2 from the direct ploughing action of the trawl, O_2 normally transported into the sediment via bioturbation would be limited. This further supports the results as NO_3^- within untrawled sediments proliferated deeper throughout the year in all but one month which coincided with the highest bottom temperature (Fig 9). Untrawled fauna could therefore be acting to counter and offset enhanced summer oxidative consumption by transporting relatively O_2 rich overlying waters through the sediment and maintaining an oxic sediment horizon.

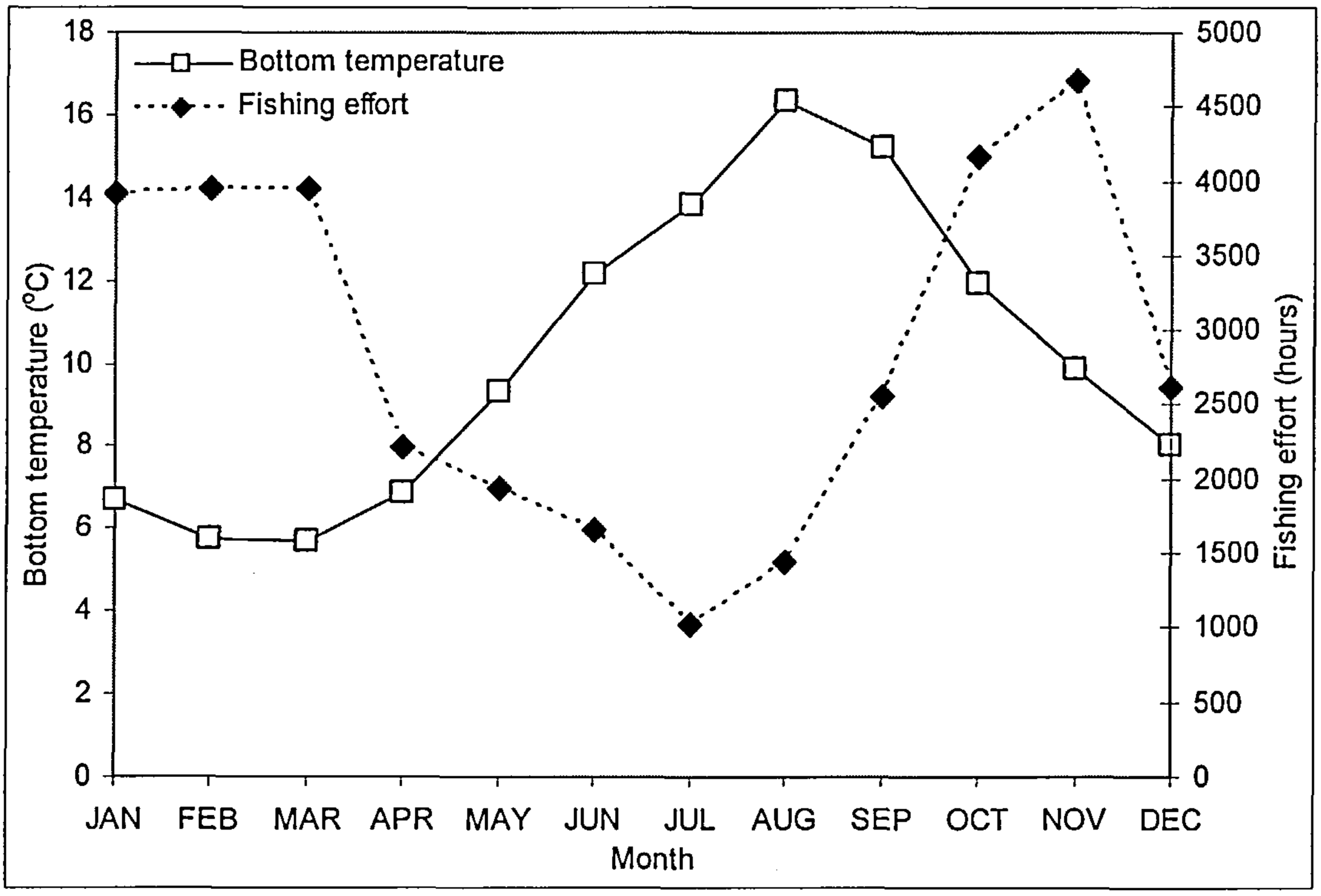

Figure 4.9. Seasonal benthic dynamics of average temperature (y axis, line and square) and average fishing effort (z axis, broken line and solid diamond) within ICES statistical rectangle 39 E8, for 1999-2001 inclusive.

As NO_3^- was depleted during summer it should hold that anoxia was high. However, NO_2^- concentrations within trawled porewaters were highest during summer. This suggests that nitrate reduction dominated and NO_2^- was the major end product. Respiratory metabolism of NO_3^- leads to denitrification processes which consequently produces NO_2^- (Koike and Sorensen 1988). Keiskamp et al. (1991) also discovered low NO_3^- due to diminished nitrifying activity during summer anoxia in coastal marine sediments as a result of reduced O_2 penetration. Coupled nitrification – denitrification processes could account for the observed reduced summer NO_3^- concentration in untrawled, compared to trawled, interstitial porewaters.

In general terms, a trend of increasing concentration with increasing sediment depth was observed for PO_4^{3-} in trawled and untrawled porewaters.

This can be explained by the liberation of soluble PO_4^{3-} from phosphorus associated with oxidised Fe(III) hydroxides which occurs within deeper anoxic conditions where free O_2 is exhausted (Slomp et al. 1998). Sediment anoxia occurred in trawled areas during summer. PO_4^{3-} concentrations under these conditions were at their lowest. The lack of a distinct sediment oxic horizon may lead to rapid diffusion of PO_4^{3-} across the sediment-water interface (Zwolsman 1994). It has been shown that PO_4^{3-} flux from the sediment is highest during summer when mineralization is most rapid due to high temperatures (Clavero 2000). A consequence of a rapid and prolonged efflux of PO_4^{3-} could be depletion within the interstitial waters greater than organic matter degradation could replenish a significant amount of PO_4^{3-}. Slomp et al. (1998) demonstrated that an oxic layer of <1mm would cause rapid efflux of porewater PO_4^{3-}. Therefore during the summer, when sediment O_2 concentrations are generally low (Nedwell et al. 1983), trawled sediments may act as a source of PO_4^{3-}. This is of significance as PO_4^{3-} is readily utilised by phytoplankton and consequently could contribute to primary production in overlying waters (Klump and Martens 1983). At other times of the year trawl activity could well have acted to maintain enough of an oxic input to regulate PO_4^{3-} flux and transformations. At times of high fishing effort (Fig 4.9), a distinct zone between relatively homogenous surficial sediments, of low concentration, and deeper stratified layers, with higher concentrations increasing with depth occurred. This zone was apparent at approximately 4 cm, a depth to which trawl ground gear can penetrate muddy sediments and could transport O_2 rich water. Bioturbation within untrawled sediments evidently was sufficient to create a surficial oxidising layer throughout the year. Under such oxic conditions a fraction of the soluble PO_4^{3-} would be co-

precipitated with iron and maintained within the sediment for further transformations (Sundby et al. 1992). Untrawled sediments with relatively higher bioturbation created a mosaic of PO_4^{3-} micro-environments. Microenvironments were seasonal because during winter (January – March) when bioturbation activity would be minimal due to low temperature, a stratified pattern of PO_4^{3-} was evident.

NH_4^+ in trawled sediment porewaters displayed a profile with a homogenous surface layer (~4 cm) with lower concentrations than untrawled sediments. Obviously this mimics the pattern expressed by PO_4^{3-} and can be attributed to the same direct impact of the trawl gear. Again, below ~4 cm, NH_4^+ concentrations rapidly increased with depth where anoxic conditions would likely proliferate (Nedwell et al. 1999). Reducing conditions would favour bacterial degradation of organics to NH_4^+ consequently giving high concentrations in deeper sediments (Nedwell et al. 1999). Biological activity within untrawled sediments is likely to be greater than in trawled areas. It must be noted that; trawled sediments commonly have a high incidence of opportunistic species that scavenge on a food source in the form of moribund organisms following trawl disturbance, yet these organisms generally dwell on the sediment surface and have a limited impact on bioturbation (Kaiser and Spencer 1994). Therefore, untrawled NH_4^+ profiles displayed a higher degree of heterogeneity. January through to March was the only exception, and again suggests a decline in organism activity in response to low temperatures, resulting in stratified sediments. Of course, as for PO_4^{3-}, the 'potential extra nutrient' available to primary production due to trawling will not all reach the

overlying water as some will be oxidised and precipitate out (PO_4^{3-}), and undergo ion exchange (NH_4^+ and PO_4^{3-}).

Immediately following a trawl event, the measured porewater nutrients were altered beyond the patterns expressed within untrawled and trawled profiles. Nutrient concentrations at all other times of the year were compared to these values and did not exceed the immediate impact of a trawler. Therefore, this demonstrates the direct impact of a trawl event (although likely to be short-term) to be greater than natural seasonal variations.

Similar patterns were expressed in NH_4^+ and PO_4^{3-} porewater profiles. Initial concentrations were relatively low and increased with depth. As outlined above, depth increases typically exhibit a shift from oxidising to reducing conditions and thus allows for the step-wise breakdown of complex organic material into inorganic forms of nitrogen (NH_4^+) and phosphorous (PO_4^{3-}) (Froelich et al. 1979). The higher bioturbation that would occur in untrawled sediments if the fauna were larger burrowing species, compared to trawled, would elevate sedimentary O_2 input and therefore explain the shallower slope of nutrient increase with depth in the untrawled profile (Hines and Jones 1985). Immediately following trawling however, NH_4^+ and PO_4^{3-} concentrations were enhanced at all depths. This could suggest a very rapid recovery of dissolved constituents, the proliferation of highly reducing conditions or other phenomena. However, when a diffusion coefficient calculation was employed (Equation 1) it was found that NH_4^+ would have required at least 48 hours to exceed a 1 cm (z = 1.0386 cm) vertical diffusion. Therefore it is highly unlikely that rapid recovery was occurring.

$$Z = \sqrt{Dt}\ ;$$

Equation 1. Diffusion coefficient equation based on the diffusional rate of NH_4^+. Where Z = depth to which NH_4^+ could be gained / lost by diffusion through the sediment in time period t (seconds). D = the approximate sedimentary diffusion rate of NH_4^+ ($6*10^{-6}$ cm^{-2} S^{-1}).

Immediately following a trawl event extensive sediment excavation and mobilisation is likely to occur to the depth of gear penetration (Churchill 1997). It is therefore possible that a temporary nepheloid layer was created before resettlement and / or horizontal transport of sediment particles. As the core equipment was deployed directly behind the trawler, the corer may have passed through the fluidised layer. Therefore the corer would have sampled what were deeper pre-trawled and hence probably reduced sediments, and help explain the concentration increases of NH_4^+ and PO_4^{3-}. An extended oxic layer, typically expected from trawl disturbance, would only subsequently form and the results of this investigation suggest this to require > 15mins in winter sea temperatures. Surface perturbations in NO_2^- and NO_3^- concentrations were evident immediately after trawling. It is possible that O_2 at the sediment-water interface acted to stimulate intense coupling of nitrification and denitrification (Henriksen and Kemp 1988, Koike and Sorensen 1988). Such rapid shifts between nitrification and denitrification may act to alter NO_2^- and NO_3^- concentrations but may not have had enough time for subsequent impacts on NH_4^+ and PO_4^{3-}.

Spectrophotometric analysis of marine porewaters detects between 30 – 60% of dissolved organic carbon (DOC) in the form of CDOM and therefore it is an appropriate proxy for the amount of dissolved organic matter within the samples (Kitidis 2002). The pattern of DOC within sediment porewaters indicated three interrelated processes to have occurred. Initially DOC within

surficial sediments (~ 0 - 1 cm) exhibited similar levels to those in overlying waters. POM deposited on the sediment would inevitably undergo a temporal lag before being incorporated into the sediment where oxidising bacterial populations would initiate POM breakdown transformations (Parsons et al. 1984). Following this lag period, DOC concentration exhibited an almost linear increase with depth from ~ 1 – 7 cm, the production phase. Redox conditions favouring oxidants that yield a high free energy change per mole of organic carbon mineralised combined with a large supply of organic matter at this depth support high levels of DOC production (Kitidis 2002). DOC production therefore exceeded the rate at which it could be consumed. At depths greater than ~ 7 cm DOC declined with depth. At such depths anaerobic conditions probably dominated and thereby limited the production of DOC and consequently enabled DOC to be bacterially consumed, thus regenerating nutrients (consumption > production) (Schlesinger 1991, Kitidis 2002). We have shown NH_4^+ and PO_4^{3-} concentrations to increase with depth where anaerobic conditions could facilitate their release from degradation transformations. Thus, DOC levels within sediment porewaters reflected three distinct phases; 1, Lag phase; 2, Production phase; and 3, Consumption phase.

It has been clearly demonstrated that trawl activity affects early diagenetic processes. Not only do these impacts occur immediately following trawling but the effects have been shown to persist in trawled areas, when compared to untrawled control areas that exhibited similar sediment characteristics.

Chapter 5:

The impact of trawling on the nutrient dynamics of North Sea sediments: A microcosm investigation.

5.1 Abstract

The effects of trawling on North Sea sediment nutrient dynamics were examined in a series of nine 30cm deep, 37.5cm x 21cm laboratory microcosms. North Sea sediments were collected from a fishing ground, located approximately 6km off the U.K coast, centred on $55^0$13.55'N $01^0$27.28'W, placed into the microcosms and allowed to stabilise for a period of ten days. Simulated trawling events were carried out by dragging a section of ground gear through the sediment. Fishing intensity corresponded to moderate (trawled every other day) and heavy (trawled every day) trawl frequencies which were compared to untrawled control systems. Nutrient concentrations (NH_4^+, PO_4^{3-}, NO_2^- and NO_3^-) were measured at hourly intervals for the first three hours following each disturbance event, and then every 4 hours for the next 98 hours. The results were used to estimate the benthic nutrient fluxes for each treatment.

Trawling impacted the fluxes of all of the measured nutrients. An enhanced sediment efflux that was observed for NH_4^+ (475 % greater than the background flux) and NO_2^- (26 % greater than the background flux) persisted > 4 days. A PO_4^{3-} influx in the untrawled systems was reversed to a net efflux following trawl disturbance (-15 %). NO_3^-, displayed a net efflux in the control systems yet, this was reduced under intensive trawling and reversed to an influx of 1.0 % greater than the background flux. Altered fluxes were seen to persist between consecutive trawl events (i.e. >48 hours). These results imply that in

heavily fished areas of the North Sea benthic nutrient fluxes can be significantly modified, with potential implications for phytoplankton abundance and composition.

5.2 Introduction

Continental shelf sediments are often organically rich (0.1 – 10 % organic carbon by weight), which can give rise to high productivity (Hensen et al. 1998). The breakdown products of microbially-mediated organic matter oxidation include inorganic nutrients. These and other by-products, generated during oxidative reduction (Reimers et al. 2001), accumulate in the sediment porewaters until their concentrations become limited by coupled-inorganic mineralization/sorption reactions, diffusional benthic exchange, bioturbation and other benthic disturbance, including current and tidal disturbances (Bale at al. 1985, McCave 1976, Lavery et al. 2001). Benthic nutrient regeneration via these mechanisms can contribute significantly to the productivity of the overlying waters (Gibbs et al. 2002, Hines et al. 1982, Maksymowska-Brossard and Piekarek-Jankowska. 2001). For example, Percival (this thesis, chapter 6) reported that bioturbation increased benthic nutrient fluxes by up to; 81 % NH_4^+, 197 % PO_4^{3-}, 96 % NO_2^- and 33 % NO_3^-, when compared to diffusional exchange rates without macrofauna. In areas that are intensively trawled, the additional physical disruption of the sediment surface can further enhance sediment-water exchange, thereby increasing the potential impact on regional primary production (Pilskaln et al. 1998). Studies have shown that in areas of high trawl activity, benthic remineralisation can contribute more than half of the photosynthetic nutrient requirement (Klump and Martens 1987).

In addition to the enhancement of benthic fluxes (Klump and Martens 1983), the physical sediment mixing caused by natural processes and trawling gear can result in the downward mixing of relatively fresh (labile) organic matter and oxygen rich surface waters, giving rise to changes in the redox status of the sediment (Mayer et al. 1991). The amount of organic matter and the availability of oxygen are critical factors in controlling remineralisation (Hall et al. 1996). Bacterially mediated transformation of organic matter ultimately controls the amount of regenerated products (Van Duyl et al. 1993). Oxygen utilisation is more critical in controlling the rate of organic breakdown because oxygen based respiratory pathways are the most efficient route to degrade organic material (Sun et al. 1997). Diagenetic transformations occur though the most energy efficient route. As a result, compounds undergo bacterial oxidation, using the oxidant, which yields the greatest free energy change per mole of organic carbon (Froelich et al. 1979). Once the oxidant has been consumed microbial organisms switch to the next most efficient oxidant (Froelich et al. 1979).

The thermodynamic sequence of energy yield favours aerobic respiration. However, once O_2 levels fall below a critical threshold anaerobic respiration occurs in the most energetically favourable pathway with NO_3^-, MnO_2, $Fe(OH)_3$ and SO_{42-} being successively utilised (Froelich et al. 1979). Therefore, the interrelationships between aerobic and anaerobic processes are crucial in controlling the breakdown of organic matter with some compounds resistant to degradation under strictly anaerobic conditions (Schink 1989). Although redox changes and organic enhancement due to trawling may be temporary, they may nevertheless enhance subsequent transformations of organic material and hence modify the rate of sediment-water nutrient exchange.

The release of nutrients to the overlying water provides a reactive fraction that is readily available for photosynthesis. Denitrifying bacteria, associated with deeper hypoxic sediments, convert remineralised nitrogenous forms into N_2 gas, which is then lost from the system and hence is biologically unavailable (Seitzinger 1988). However, following a trawl event this process is impeded because the pore waters are liberated into the oxygen rich water column.

In this chapter, I examine the contribution of trawling to benthic nutrient dynamics in the central west North Sea. The North Sea accounts for less than 0.2% of the area of the global ocean, yet is one of the worlds most heavily fished seas, contributing ~5% of the global fish catch (Heessen 1988). Fishing activity typically occurs in waters that are shallower than 60m (Lindeboom and de Groot 1998). Hence benthic nutrient regeneration resulting from trawl disturbance could contribute directly to primary production.

I simulated trawling events of various intensities in a series of laboratory microcosms and estimated associated benthic fluxes of nutrients from measured temporal changes in nutrient concentrations in overlying water. Whilst it is recognised that microcosm experiments are somewhat removed from the "real" situation they are valuable in specifically isolating cause and effect treatments that would otherwise be confounded in the natural environment by a multitude of factors.

5.3 Methods

5.3.1 Sediment sampling and microcosm set up

Surface sediment samples, defined as muddy-sand (median sediment particle size = 3.5 phi), were collected on cruises of RV *Bernicia*, from an established fishing ground in the central west North Sea (~100 m^2 centred on $55^0$13.55'N $01^0$27.28'W, Fig 5.1). Fishing effort data (fishing hours) were provided by the UK Department for Environment, Food and Rural Affairs (DEFRA) (Clark and Frid 2001). Sediments were collected from a known trawled area in order to limit the possible effect on nutrient release through altered bioturbation rates as a result of generally larger size classes of macrofauna associated with untrawled areas (see chapter 6). Sediments were collected with a van Veen grab (0.1 m^2) and transferred with minimal disturbance, to a series of 30cm (deep) x 37.5cm x 21cm black PVC coated polycarbonate microcosms. The purpose of the PVC coating was to prevent light penetration. The microcosms were maintained under Bernicia's continuous water flow system and transported to the Laboratory's aquaria suite. All samples were returned to the laboratory within 3 hours of collection.

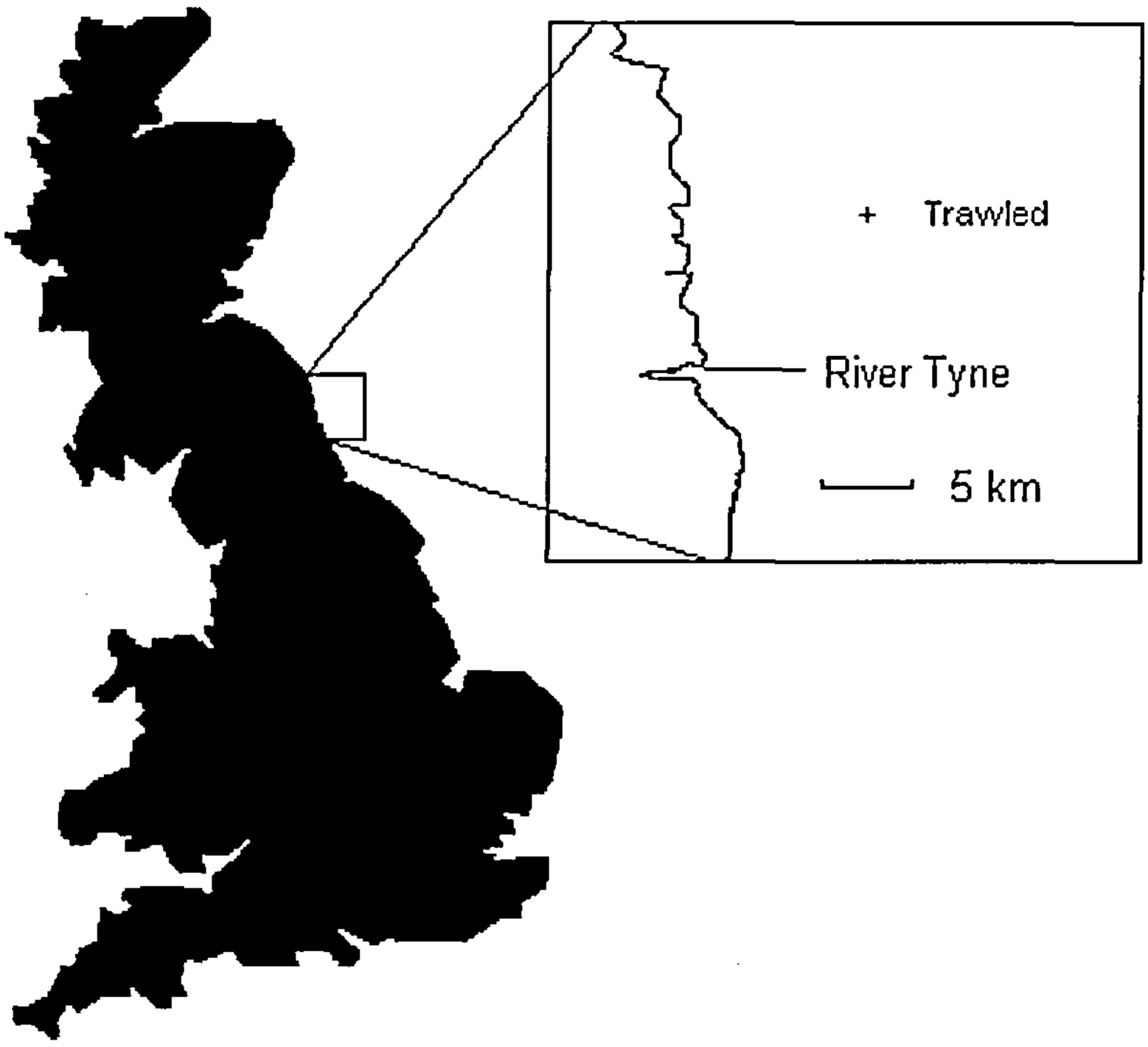

Figure 5.1. Coastal outline of north-east England and sampling site within the central-west North Sea. Trawled (+) sediment site illustrated (55^{0}13.55'N 01^{0}27.28'W).

In the laboratory the microcosms were placed under a continuous flow of filtered seawater, at coastal salinity values (34.5 ± 0.5) and *in situ* (8.5 ± 0.5 ^{0}C) temperature. The arrangement gave approximately 12cm of overlying water for each microcosm. The seawater for the system was drawn from a coastal inlet pipe, gravimetrically settled and sand filtered to remove detritus. The flow rate was maintained at ~45 $l hr^{-1}$ and allowed to run to waste. Water circulation and aeration rates were controlled using a 45 degree splash plate positioned 4cm above the water surface on to which the continuous water flow was directed. This arrangement allowed oxygen to be introduced to the system while the flow was maintained without visible resuspension of the sediment.

Following their establishment, the microcosms were left for 10 days to allow stabilisation of chemical and biological processes. This time period was

determined to allow any redox microenvironments, which may have been created during the set-up period, to dissipate but before excessive consumption of the sediments organic matter content could occur.

5.3.2 Experimental treatments and nutrient sampling

One hour prior to experimentation, three replicate samples were taken directly from the continuous flow water source and analysed for NO_2^-, NO_3^-, PO_4^{3-} and NH_4^+ in order to establish background concentrations in the overlying water. The continuous water flow system was then shut off and remained so for the duration of the experiment. This incubation was deemed necessary in order to prevent the removal of nutrients fluxing between the sediment water interface.

Three replicate microcosms were established for each of three treatments; controls (no trawl simulation), "moderate trawl effort" (trawled every other day) and "high trawl effort" (trawled every day). Trawl frequencies were selected to mimic those of ICES statistical rectangle 39E8. Specific fishing grounds within this area are often targeted for periods of up to four days. Within this time period the area may be visited daily until the fisher moves to a new area (Catchpole, pers.com.).

In order to simulate a trawl disturbance event, a section of ground gear comprising a tickler chain (a steel-link chain designed to penetrate the sea-bed and cause an escape response in benthic dwelling fish) was dragged through the sediment, in a single pass at approximately 1.5 ms^{-1}. The average penetration depth of the ground gear was ~ 4 cm and extended to a maximum of 6 cm into the muddy sediment.

Three replicate samples of overlying water were collected immediately following each trawl simulation and then hourly for the next three hours. Subsequent samples were collected at four hourly periods until the next trawl simulation (high trawl effort = every 24 hours; moderate trawl effort = every 48 hours).

On each sampling occasion 40ml of overlying water was carefully withdrawn from each microcosm using acid cleaned 50ml plastic syringes and 0.45µm filtered (Millipore) into gas tight polypropylene bottles. Samples were immediately frozen for subsequent nutrient analysis.

5.3.3 Nutrient analysis

Concentrations of NO_2^-, NO_3^-, NH_4^+ and PO_4^{3-} were determined with an automated nutrient analyser (Skalar San ^{Plus}), following standard protocols (Brewer and Riley 1965; Mantoura and Woodard 1983; Kirkwood 1989). Analytical precisions ± 1% were routinely achieved for all methods (see chapter 3).

5.3.4 Sediment characterisation

At the end of each experiment, three replicate core samples (7cm diameter x 15cm deep) were collected from each microcosm and analysed for grain size, organic matter content and porosity. Sediment grain size was determined using standard gravity sedimentation (Buchanan 1984). Organic matter content was determined by wet digestion in order to account for the possible presence of mineral carbon (coal) (Buchanan 1984). Porosity was determined by weight loss on drying at 80^0C.

5.4.5 Calculation of flux change

Benthic nutrient fluxes (F; μmol m^{-2} d^{-1}) within the microcosms were estimated using the following equation:

$$\frac{((c_2 - c_1) * v)}{a \div (t_2 - t_1)},$$

where c = concentration (μmol L^{-1}), v = volume (L), a = area (m) and t = time (hr). Changing concentrations within the control systems over time were modelled with a line of best fit. Control flux rates were then calculated from the slope of the fitted line. Instantaneous fluxes were calculated from the concentration preceding a trawl event. Fluxes between trawl events were calculated using the slope of the line fitted on those values between trawl impacts and subtracting the concentration following a trawl event from the concentration preceding the next trawl event or end of the experiment.

In order to assess the significance of trawling on benthic nutrient dynamics, the untrawled nutrient fluxes were first scaled to annual rates for the area within 39E8 accessible to trawling (approximately 50% or 1545.5 km^2), hereinafter referred to as the background mean flux. DEFRA fishing effort data, for 39E8 for the period 1999-2001 inclusive, was averaged to give an annual measure of fishing effort. Annual fishing data were calculated from a conservative estimate of the average total ground gear wing spread (total area covered) and tow speed, 10.6m and 5.5 km/hr respectively, to give the annual area covered by otter trawl ground gear by the trawl fleet within 39E8. A second calculation was made to account for the area covered solely by trawl doors, based on total trawl door width of 2.2m. Although otter trawl doors penetrate deeper into the sediment this calculation would give a conservative estimate of

the impact of trawl doors because the simulated trawl impacts penetrated to a depth representative of ground gear only (~ 4 cm). The percentage change in flux as a result of trawling was estimated from appropriately scaled (trawled flux * area trawled – control flux for trawlable area of 39E8) nutrient fluxes measured following simulated trawl events within the experimental systems. These calculations were based on heavily trawled, intermediately trawled and untrawled sediments for the total area covered and trawl doors.

5.5 Results

There were no significant differences between nutrient concentrations in the inflows and any of the systems (one-way ANOVA; NO_2^- F=0.24, p=0.867; NO_3^- F=2.49, p=0.134; NH_4^+ F=1.17, p=0.379; PO_4^{3-} F=0.36, p=0.784). In other words the aquaria suite had not altered (through the possible build up of organic matter) and had maintained the microcosms under *in situ* conditions. Consequently I was confident that initial nutrient concentrations were not influenced by aquarium conditions and subsequent results were due to the experimental treatments.

5.5.1 Sediment characteristics

Sediments collected from each microcosm did not vary significantly in porosity (Kruskal-Wallis, W=6, p=0.081), .grain size (median particle size = 3.5 phi/muddy sand for all systems) or percentage organic content (Kruskal-Wallis, W=6, p=0.081) between systems.

5.5.2 Microcosm nutrient concentrations and fluxes

In the following section negative fluxes refer to a net influx whilst positive fluxes refer to sediment efflux. In the control system's overlying water, NO_2^- increased steadily throughout the 98 hour duration of the experiment from 0.2 (± 0.02) to 0.5 (± 0.1) µmol L^{-1} (Fig 5.2a), corresponding to a benthic flux of 8.5 (± 1.0) µmol m^{-2} d^{-1} (Table 5.1). The first trawl event of both the moderately and heavily impacted systems gave instantaneous concentration increases of approximately 1.5 and 1.0 µmol respectively (Fig 5.2b and c). The corresponding instantaneous fluxes were 166 (± 55) and 111 (± 7.5) µmol m^{-2} hr^{-1} (Table 5.1). Conversely, the NO_2^- concentration decreased at the second trawl impact. In both the moderate and high trawl effort systems, reductions in concentration at the second impact created negative instant fluxes of -17.5 (± 7.0) and -7.0 (± 1.0) µmol m^{-2} hr^{-1}. Nonetheless, in the moderately trawled system post disturbance flux rates, displayed similar values (22.0 (± 1.0) and 21.0 (± 1.5) µmol m^{-2} d^{-1}) (Table 5.1). The NO_2^- concentration in the heavily trawled system declined at the final two trawl simulations by 0.3 (± 0.1) µmol L^{-1} (Fig 5.2c), resulting in influx values of -19.0 (± 0.5) and -12.0 (± 1.5) µmol m^{-2} hr^{-1}. However, while the final three trawl events caused an instantaneous nutrient influx, the post trawl daily flux rates all remained positive with successive rates of 3.0 (± 1.0); 1.0 (± 2.0); 8.0 (± 1.5) and 18.0 (± 1.5) µmol m^{-2} d^{-1}.

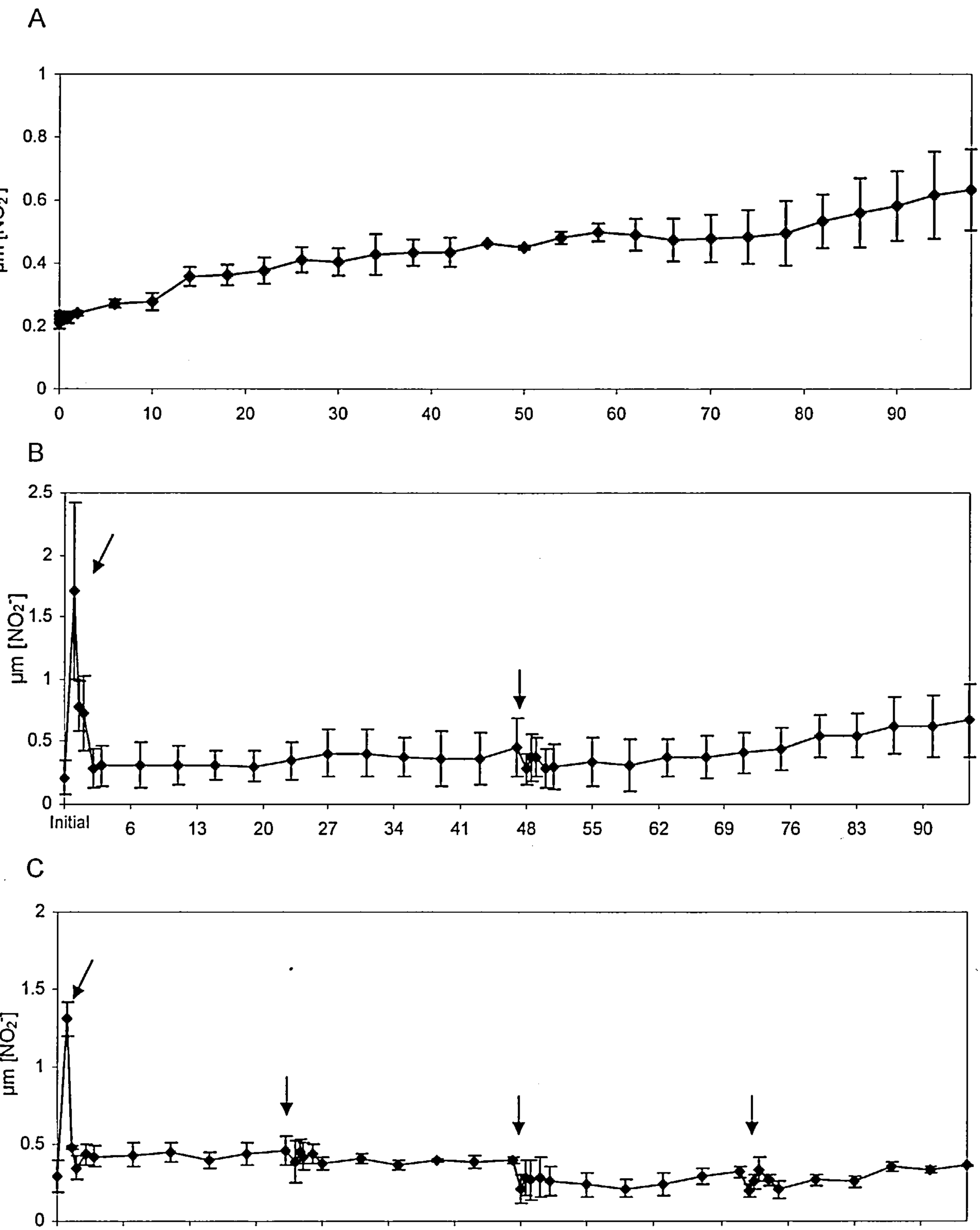

Figure 5.2. Concentration of nitrite (µmol L^{-1}) within microcosms exposed to different trawling regimes ± standard deviation: A, Control (without trawling), B, Moderate trawl effort (trawled every other day) and C, heavy trawl effort (trawled every day) frequencies. (↓ denotes a disturbance event).

Table 5.1. Summary of NO_2^-, NO_3^-, PO_4^{3-} and NH_4^+ concentrations (µmol L^{-1}) within experimental microcosms for control, moderate and heavy trawl effort ± standard deviation. Calculated instant and post disturbance flux vales presented in µmol m^{-2} hr^{-1} and µmol m^{-2} d^{-1} respectively. Min and max indicate the minimum and maximum concentrations attained during the experiment. Initial refers to the first pre-trawl system concentration and final refers to the last sampled concentration.

Treatment	Nutrient	Nutrient Concentration µmol L^{-1}				Control µmol $m^{-2}d^{-1}$	Instant flux immediately after trawling µmol m^{-2} hr^{-1}				Post trawl impact flux rates µmol m^{-2} d^{-1}			
		Initial	Final	Min	Max		1^{st} impact	2^{nd} impact	3^{rd} impact	4^{th} impact	1^{st} post impact	2^{nd} post impact	3^{rd} post impact	4^{th} post impact
Without trawling	NO_2^-	0.2 ± 0.02	0.5 ± 0.1	0.2 ± 0.02	0.5 ± 0.1	8.5 ± 1	-	-	-	-	-	-	-	-
Moderate trawl effort	NO_2^-	0.2 ± 0.2	0.5 ± 0.5	0.2 ± 0.2	1.5 ± 0.7	-	166 ± 54.5	-18 ± 7	-	-	22 ± 1	21 ± 1	-	-
Heavy trawl effort	NO_2^-	0.3 ± 0.1	0.4 ± 0.01	0.2 ± 0.05	1.5 ± 0.1	-	111 ± 7.5	-7 ± 1	-19 ± 0.5	-12 ± 1	3 ± 1	1 ± 2	8 ± 1	18 ± 1
Without trawling	NO_3^-	2 ± 0.5	10 ± 1.5	2 ± 0.5	10 ± 1.5	176 ± 19	-	-	-	-	-	-	-	-
Moderate trawl effort	NO_3^-	4.5 ± 1.5	1.0 ± 0.5	0.5 ± 1	4.5 ± 1.3	-	-235 ± 29	-47 ± 16	-	-	-71 ± 9	14 ± 6	-	-
Heavy trawl effort	NO_3^-	2 ± 0.1	0.2 ± 0.05	0.2 ± 0.06	2.5 ± 0.3	-	-53 ± 38	-5 ± 7	35 ± 11	33 ± 9	-80 ± 6	-29 ± 1	1.5 ± 1	0.5 ± 1
Without trawling	PO_4^{3-}	2.5 ± 0.5	1 ± 0.2	1 ± 0.2	3 ± 0.5	-49 ± 10.5	-	-	-	-	-	-	-	-
Moderate trawl effort	PO_4^{3-}	2.5 ± 0.5	0.5 ± 0.2	0.5 ± 0.02	2.5 ± 0.5	-	-109 ± 19	140 ± 33	-	-	-97 ± 7	-42 ± 5	-	-
Heavy trawl effort	PO_4^{3-}	2.5 ± 0.2	0.5 ± 0.1	0.5 ± 0.02	4 ± 0.5	-	-65 ± 14	163 ± 28	200 ± 44	308 ± 51	-104 ± 8	-148 ± 24	-19 ± 6	-2 ± 1
Without trawling	NH_4^+	1 ± 0.5	2.5 ± 0.5	1 ± 0.1	2.5 ± 0.5	21 ± 4.5	-	-	-	-	-	-	-	-
Moderate trawl effort	NH_4^+	1.5 ± 0.5	16 ± 1.5	1.5 ± 0.5	17.5 ± 2	-	439 ± 27	740 ± 166	-	-	192 ± 14	85 ± 36	-	-
Heavy trawl effort	NH_4^+	2 ± 0.5	32 ± 1.5	2 ± 0.5	39 ± 2	-	358 ± 19	1136 ± 145	976 ± 82	1587 ± 141	270 ± 30	183 ± 26	40 ± 26	9 ± 11

The control microcosm exhibited a net gain in NO_3^-, increasing to 10.0 (± 1.5) µmol L^{-1} (Fig 5.3a) at the end of the experiment. The resultant flux was 176.5 (± 19.0) µmol m^{-2} d^{-1} (Table 5.1). Within the moderately disturbed systems, the NO_3^- concentration decreased immediately following both disturbance events 4.5 (± 1.5) to 2.5 (± 1.0) and 1.5 (± 0.5) to 1.0 (± 0.5) µmol L^{-1} respectively (Fig 5.3b), consequently creating instantaneous flux values of -235.0 (± 29.0) and -46.5 (± 15.5) µmol m^{-2} hr^{-1}. The flux rate following the first trawl event maintained an influx flux of -71.0 (± 9.5) µmol m^{-2} d^{-1} to the second trawl event (Table 5.1). Whereas, after the second trawl event an efflux to the end of the experiment 13.5 (± 6.0) µmol m^{-2} d^{-1} occurred. The heavily trawled system exhibited a similar pattern of decreasing NO_3^- concentration at the first and second trawl simulations 2.0 (± 0.1) to 1.5 (± 0.5) and 1.0 (± 0.2) to 1.0 (± 0.5) µmol L^{-1} (Fig 5.3c). However, while the magnitude of the instantaneous flux values declined after the second disturbance, the fluxes following the first and second disturbance were negative -53.0 (± 38.0) and -5.0 (± 6.5) µmol m^{-2} hr^{-1}. Interestingly, the third and fourth trawl events elevated the NO_3^- concentration by 0.5 µmol. The instantaneous flux rates at the third and forth trawl events were 35.0 (± 11.0) and 33.5 (± 8.5) µmol m^{-2}n hr^{-1} respectively. Again the magnitude of the subsequent flux rates following the later two trawls declined at 1.5 (± 1.0) and 0.5 (± 1.0) µmol m^{-2} d^{-1}.

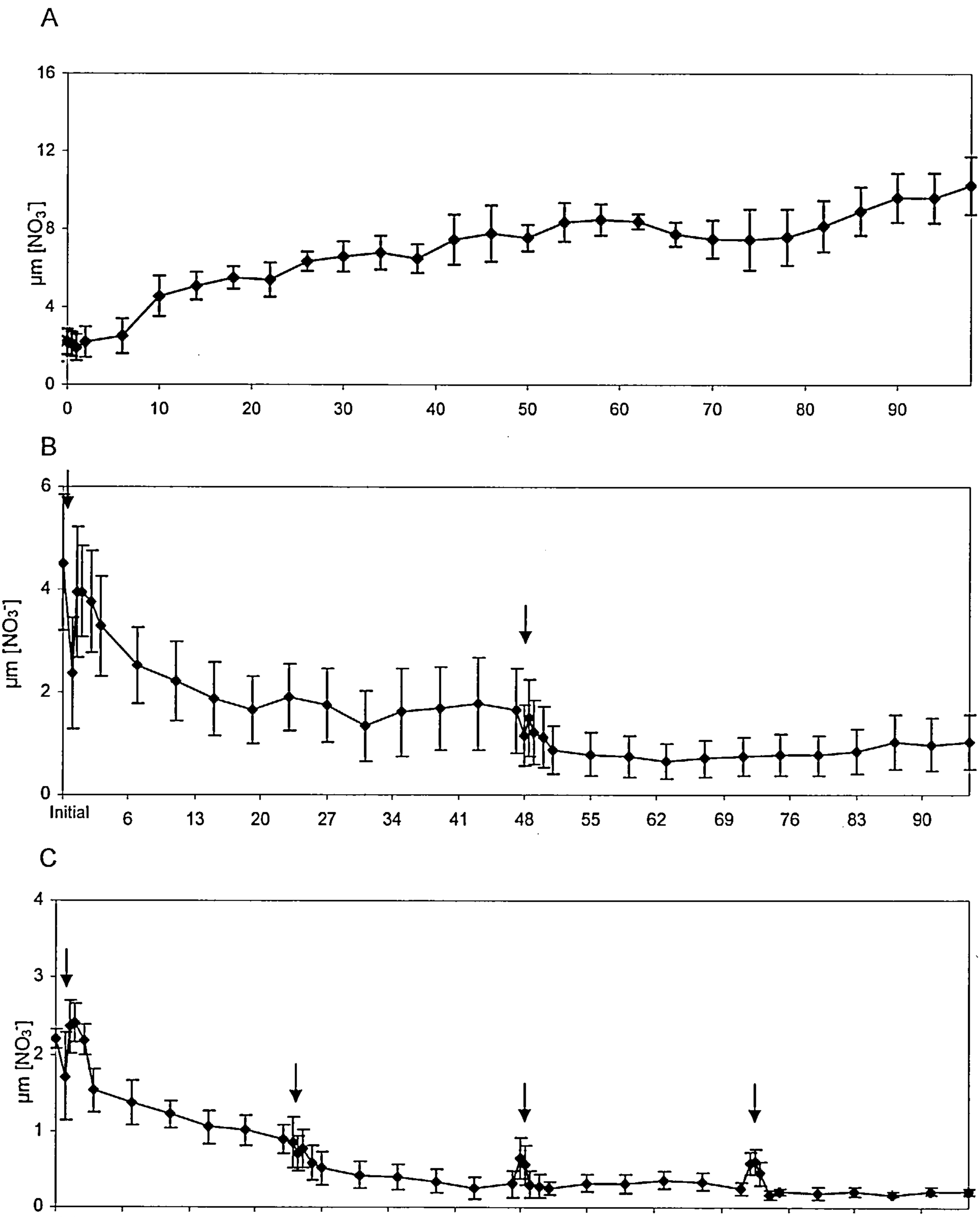

Figure 5.3. Concentration of nitrate (µmol L^{-1}) within microcosms exposed to different trawling regimes ± standard deviation: A, Control (without trawling), B, Moderate trawl effort (trawled every other day) and C, heavy trawl effort (trawled every day) frequencies. (↓ denotes a disturbance event).

The control system displayed a PO_4^{3-} decline from 2.5 (± 0.5) to 1.0 (± 0.2) µmol L^{-1} (Fig 5.4a) with a flux rate of -49.0 (± 10.5) µmol m^{-2} d^{-1}. The moderately trawled system exhibited a decrease in PO_4^{3-} concentration at the first trawl simulation of 2.5 (± 0.5) to 1.5 (± 0.1) µmol L^{-1} (Fig 5.4b). This resulted in a negative flux of -109.0 (± 19.0) µmol m^{-2} hr^{-1} (Table 5.1). At the second disturbance however, the PO_4^{3-} concentration immediately increased by 1.5 µmol L^{-1}. The instant flux stimulated by this second trawl event was 139.5 (± 33.5) µmol m^{-2} hr^{-1}. The flux rates during the periods preceding, and following, the second trawl event were both negative at -96.5 (± 7.0) and -41.5 (± 5.5) µmol m^{-2} d^{-1} respectively. The heavily trawled system also exhibited a decline in concentration at the first trawl event of 0.5 µmol (Fig 5.4c). The subsequent three trawl events stimulated increasingly elevated PO_4^{3-} concentrations of 1.5; 2.0 and 3.0 µmol within the microcosms (Fig 5.4c). The corresponding instantaneous fluxes associated with each of these were 163.5 (± 28.0); 200.5 (± 43.5) and 307.5 (± 51.0) µmol m^{-2} hr^{-1} (Table 5.1). From three hours after a trawl event to the next trawl event or the end of the experiment all flux rates were negative with values of -104.0 (± 8.0); -147.5 (± 24.0); -18.5 (± 6.0) & -2.5 (± 1.0) µmol m^{-2} d^{-1} respectively.

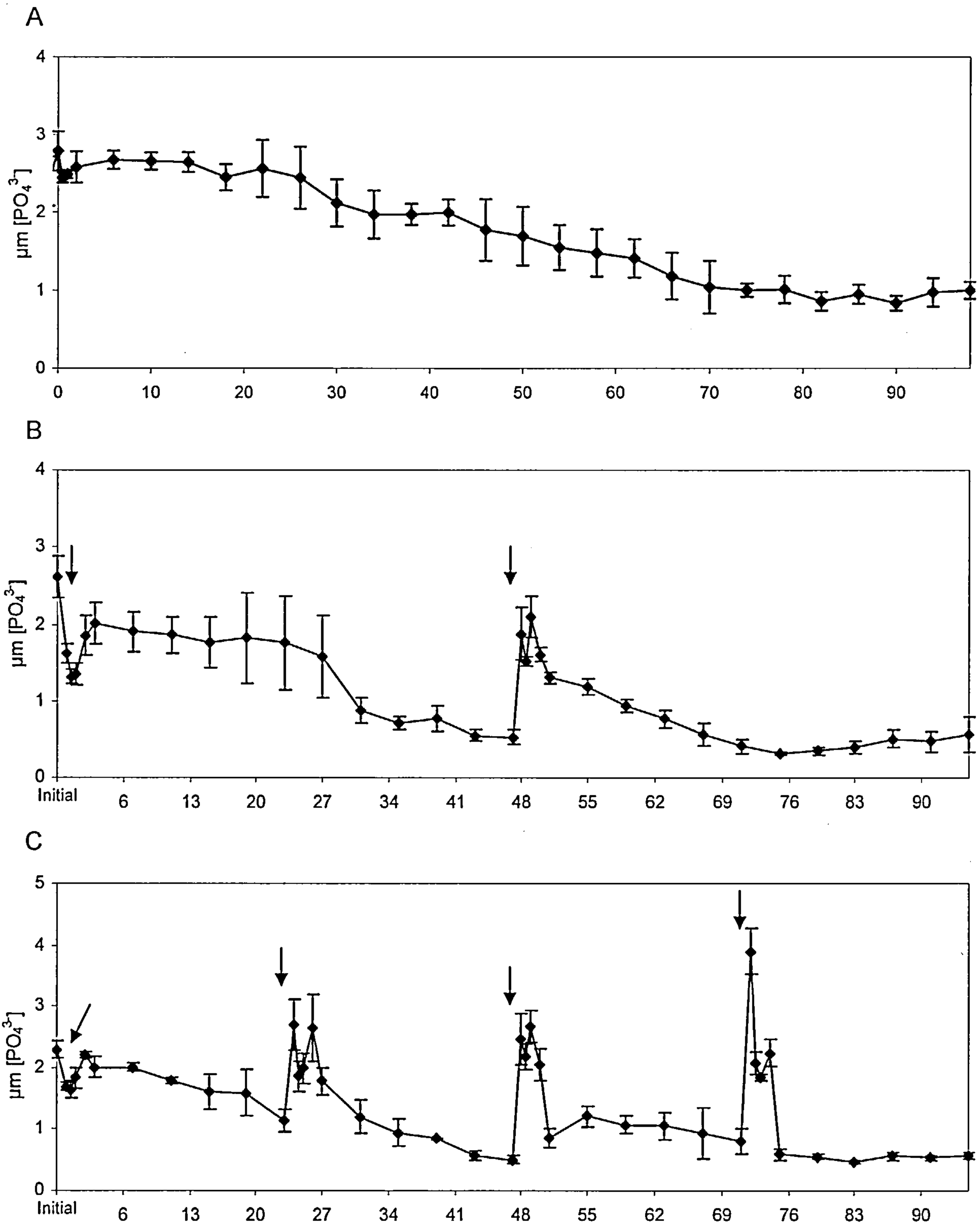

Figure 5.4. Concentration of phosphate (µmol L^{-1}) within microcosms exposed to different trawling regimes ± standard deviation: A, Control (without trawling), B, Moderate trawl effort (trawled every other day) and C, heavy trawl effort (trawled every day) frequencies. (↓ denotes a disturbance event).

The NH_4^+ concentrations within the non-trawled controls steadily increased over the duration of the experiment from 1.0 (± 0.5) to 2.5 (± 0.5) µmol L^{-1} (Fig 5.5a), giving a resultant flux rate of 21.0 (± 4.5) µmol m^{-2} d^{-1} (Table 5.1). Within those microcosms exposed to a moderate level of simulated trawling, the NH_4^+ concentration rose steeply following both trawl events from 1.5 (± 0.5) to 5.5 (± 0.5) µmol L^{-1} immediately following the first disturbance and 10.0 (± 1.0) to 17.0 (± 1.0) µmol L^{-1} after 48 hours (Fig 5.5b). The two trawl simulations produced fluxes of 439.0 (± 27.5) and 739.5 (± 165.5) µmol m^{-2} hr^{-1} (Table 5.1). Following a brief period of stabilisation after each trawl event, the subsequent flux rates, while both maintained values above that displayed in the control system, declined at 192.0 (± 14.0) and 84.5 (± 36.5) µmol m^{-2} d^{-1} respectively. A similar amplitude of increase in NH_4^+ concentration was also displayed by the first two trawl events in the heavily trawled system (trawl event one from 2.0 (± 0.5) to 5.0 (± 0.02) µmol L^{-1}; trawl event two 7.5 (± 0.5) to 18.0 (± 0.5) µmol L^{-1}) (Fig 5.5c). The third and forth simulated trawl impacts also increased the NH_4^+ concentration by 9.5 and 16.0 µmol respectively, and stimulated large efflux rates of 976.0 (± 82.0) and 1586.5 (± 141.0) µmol m^{-2} hr^{-1}. Yet, the magnitude of each flux preceding a trawl simulation, while maintaining a positive flux, decreased with values of 270.0 (± 30.0); 182.5 (± 25.5); 40.5 (± 26.5) and 9.5 (± 11.5) µmol m^{-2} d^{-1}. This successive decline in NH_4^+ concentration preceding trawling may have resulted from a build up of NH_4^+ within the microcosm.

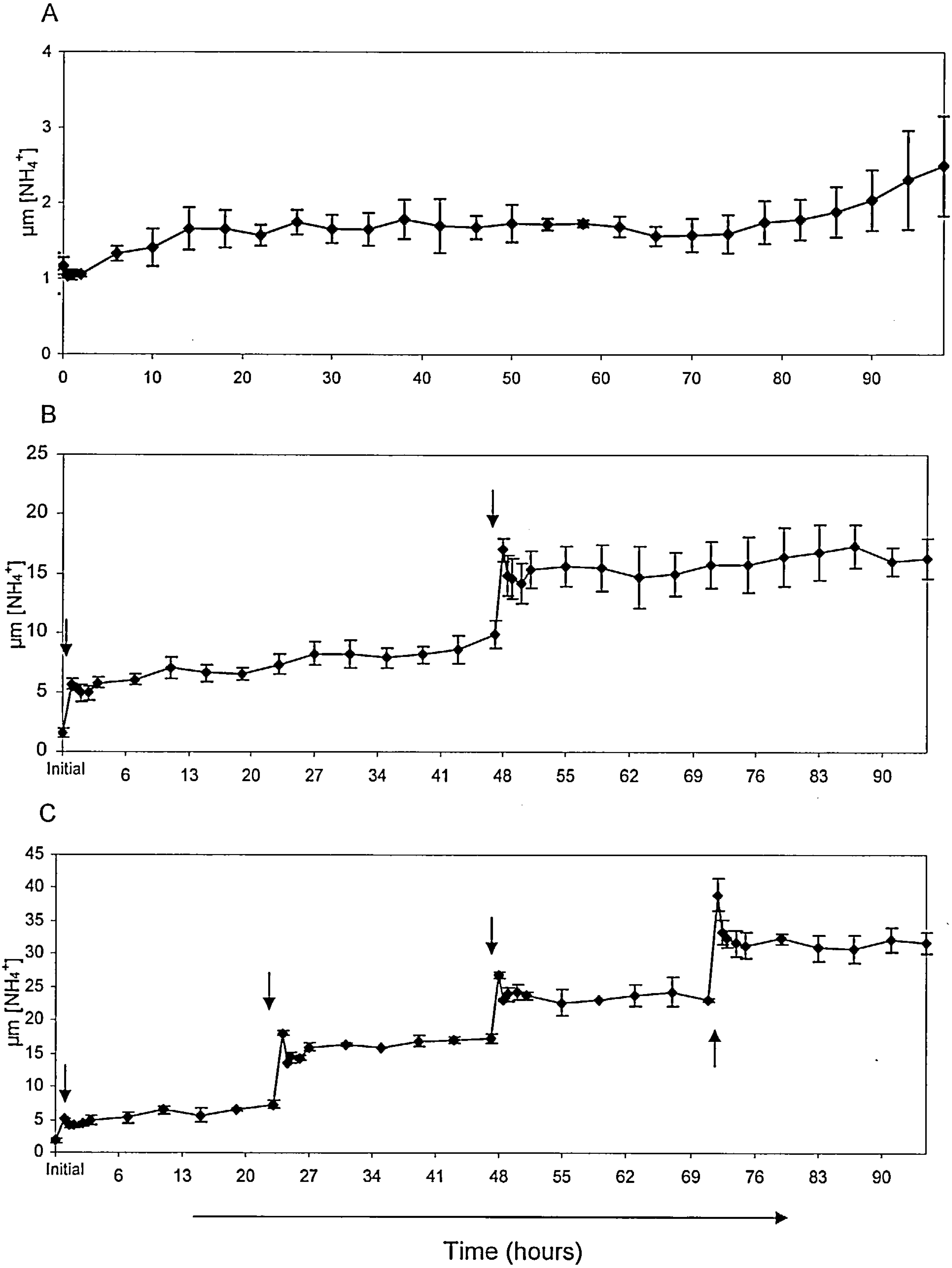

Figure 5.5. Concentration of ammonium (µmol L^{-1}) within microcosms exposed to different trawling regimes ± standard deviation: A, Control (without trawling), B, Moderate trawl effort (trawled every other day) and C, heavy trawl effort (trawled every day) frequencies. (↓ denotes a disturbance event).

5.5.3 Summary of results

In the control systems NO_2^-, NO_3^- and NH_4^+ all showed time dependent concentration increases, whereas PO_4^{3-} decreased. Initial disturbance to sediments yielded an increase in NO_2^-, while all subsequent trawl impacts caused a reduction of NO_2^- fluxes. All post disturbance NO_2^- flux rates were positive. At the first two trawl events NO_3^- decreased. The following two trawl events within the heavily trawled microcosm resulted in elevated NO_3^- fluxes.

The magnitude of post trawl NO_3^- flux declined after each successive impact. PO_4^{3-} initially decreased at the first trawl event, yet displayed an elevated flux at each subsequent disturbance. An influx of PO_4^{3-} occurred between every trawl event. Following each trawl event NH_4^+ instantly increased and maintained an elevated flux to the next trawl event for both intermediate and heavily trawled treatments.

5.5.4 The flux from trawling

Estimating the annual change in nutrient flux as a result of trawl activity suggests that increasing trawl frequency reduced the sediment flux of NO_2^- compared to the annual background flux for any trawl frequency and total ground gear and trawl doors (Table 5.2). Therefore, a greater sediment efflux of NO_2^- occurred from a low frequency of trawl disturbance.

A net NO_3^- efflux occurred from undisturbed sediments, whereas, any level of disturbance by trawl doors or total ground gear led to a change in the direction of NO_3^- flux across the sediment-water interface (Table 5.2). NO_3^- displayed the greatest change from the background flux at intermediate levels of fishing intensity.

The PO_4^{3-} flux from intermediate trawling was not significantly greater (5.0 % increase) than the annual background flux (Table 5.2). Following the initial impact from the total ground gear to previously untrawled sediments the flux increased to 676.0 % greater than the annual background flux (Table 5.2). However, in contrast, a repeated heavy frequency of trawling caused a PO_4^{3-} efflux from the sediment, thus reversing the direction of flux across the sediment-water interface.

NH_4^+ flux increased with increasing fishing intensity for both trawl doors and total wing spread (Table 5.2). The flux from the impact of trawl doors resulted in an additional fluxes for either fishing intensity, reaching a maximum of 41.5 % above the background flux for heavily trawled sediments. When considering the total ground gear impact to the sediment however, the flux increase from trawling was greater than the background flux for intermediately and heavily trawled sediments with percentages of 151.5 and 475.0 % respectively (Table 5.2).

Table 5.2. The percentage change in nutrient flux arising from trawl disturbance relative to the annual background flux from undisturbed sediments within ICES statistical rectangle 39E8. Percentage comparisons are made for three scenarios: previously untrawled sediments (pristine sediments), intermediately (trawled every other day) and heavily trawled (trawled everyday) sediments. Percentage calculations are based on total trawl door and total ground gear widths. Negative numbers denote a reversal of flux across the sediment-water interface compared to the flux direction displayed in untrawled control systems.

	% nutrient flux from trawling corrected for the annual untrawled background mean flux for...					
Nutrient	Initial impact to pristine sediments		Intermediately disturbed sediments		Heavily disturbed sediments	
	and two trawl ground gears ...					
	Total ground gear	Trawl doors	Total ground gear	Trawl doors	Total ground gear	Trawl doors
NO_2^-	38.0	3.5	48.5	4.0	26.0	2.0
NO_3^-	-2.5	-0.2	-4.0	-0.5	-1.0	-0.1
PO_4^{3-}	676.0	0.5	5.0	0.5	-15.0	-1.5
NH_4^+	6.5	5.5	151.5	13.0	475.0	41.5

5.6 Discussion

The enhancement of sediment-water nutrient exchange through bioturbation and natural physical disturbance by tides, waves and currents, is well known for a range of nearshore and coastal environments (Blackley and Rickards 1992, Kendrick et al. 1998, Widdicombe and Austen 1998). At some sites in shelf seas however, trawl activity could be the most intense mechanism of benthic disturbance (Watling and Norse 1998).

Otter trawls are the most important fishing gear used by the UK fleet and between 1990-1995 comprised ~44% of the total international demersal fishing effort of the North Sea (Jennings et al. 2000) and are exclusively used in the area of the field site.

It is important to recognise however, that in the field situation the ground gear used *in situ* often comprises heavy chain bobbins and doors that penetrate deeper into the sediment, releasing deeper pore waters than the single tickler chain used here. Hence the microcosm estimates of benthic exchange given here are likely to be minimum estimates for the true situation.

Following a trawl simulation in the microcosms, nutrient fluxes increased relative to non-trawled controls, with the elevated flux persisting until either the next trawl event, or to the end of the experiment (i.e. >48 hours). Interestingly, the degree of flux enhancement was similar for moderate and high trawl frequencies. In other words, any modification of the benthic nutrient dynamic following a trawl impact persisted even if the time to the next impact was extended, as was the case in the moderately trawled system. However, the magnitude of the fluxes between successive trawl events declined. It is unlikely that the disturbance frequency exceeded the rate of organic matter degradation

supply because at successive trawl simulations the response in nutrient concentration and flux was step-wise and increased at each disturbance for NH_4^+ and PO_4^{3-}. Therefore the data suggest that these altered fluxes can be sustained over long periods and not exhaust supply. The instantaneous flux following trawl activity and subsequent post disturbance fluxes also exhibited similar patterns between corresponding trawl impacts for intermediate and heavily trawled systems. Once released, this pulse of nutrients will be potentially available for subsequent bacterial transformation and primary production.

Blackburn (1997) predicted that the nutrient contribution to overlying waters and primary producers from surficial (< 2.4 cm) sediment resuspension would be insignificant. He predicted that the same quantity of nutrients would be released from the sediment and that only the timing of release would be affected by sediment resuspension. He concluded that a resuspension depth of ≥ 2.4 cm would be required to make a significant transfer to the overlying water. The ground gear of bottom benthic trawls is known to penetrate to depths up to 6 cm (De Groot 1995) while trawl doors have been recorded penetrating up to 30 cm (Krost 1990; Jones 1992). A representative mean depth of penetration in the field is 4cm (Krost 1990). The present study clearly demonstrated a significant increase in the quantity of some nutrients liberated into the water column from a mean penetration depth of 4 cm. Studies have also stated that the magnitude of nutrient release is ultimately dependent on the amount and rate of organic degradation (Val Klump and Martens 1987). This is unquestionably a major factor, yet the rate of degradation is usually controlled by oxygen exposure (Berelson et al. 2002). Trawl gear penetration into the

sediment extends beyond Blackburn's (1997) 2.4 cm depth, but more importantly, penetrates deeper into those hypoxic sediments required to yield a significant effect. As a result, it is likely that nitrification and denitrification processes are stimulated and consequently remineralisation pathways are altered. Thus, trawl-impacted sediments can potentially contribute to the magnitude of nutrients transported to the overlying waters as well as altering the timing of release.

Once a trawl event occurred the subsequent flux rate was altered compared to the non-trawled control sediments. Not only did trawl disturbance lead to the efflux of nutrients from the sediment but the resuspension also acted to scavenge some nutrients from the water column. This influx was apparent for PO_4^{3-} and NO_3^- between trawl impacts. Conversely, fluxes of NO_2^- and NH_4^+ effluxed from the sediment. NH_4^+ displayed the biggest release from the sediment of all examined nutrients. This may hold particular significance to the planktonic ecosystem, as NH_4^+ is the most biologically available nutrient utilised by phytoplankton (L'Helguen et al. 1996). We have shown PO_4^{3-} influx to occur in undisturbed control sediments and between trawl events. However, an instantaneous efflux of PO_4^{3-} did occur during trawl simulations. Therefore trawl activity is effectively reversing the direction of flux across the sediment-water interface.

Porewater nutrients resuspended into the water column are susceptible to advective transport (Churchill 1989). Within the North Sea this is likely to be driven by the dominant counter clockwise residual circulation (Otto et al. 1990). Radach and Lenhart (1995) stated that the inflow of nutrients from the northwest was almost balanced by the northeast outflow. This would suggest

one or a combination of processes occurring. Firstly, the nutrients entering the North Sea could be utilised by primary production and replaced by new and/or remineralised nutrient fractions. Secondly, additional nutrient inputs, from new and regenerated sources, could be carried and concentrated into the middle of the central North Sea by the cyclonic current. This would create a flow of nutrients from the coast towards central regions. As trawl activity can be intense in central North Sea areas, a gyre effect could also act to reduce transport of those trawl induced nutrients and consequently enhance primary production in the central North Sea (Radach and Lenhart 1995). The circular current would transport a residual amount of nutrients, from continental estuarine input, to the northeast to balance out the northwest inflow.

As PO_4^{3-} is also readily utilised by plankton (Karlson 1989), trawl induced PO_4^{3-} fluxes could contribute to productivity in overlying waters. Consequently, this shift could act to modify local 'Redfield' nutrient ratios of 16Si:16N:1P. Such shifts in local nutrient ratios can effectively be a limiting factor for some diatoms and lead to altered abundance and/or composition of phytoplankton (Redfield et al. 1963). These changes in nutrient structure inevitably hold consequences for the ecosystem which could potentially stimulate the production of toxic phyoplankton populations (Justic et al. 1995).

Intense fishing activity in shelf seas has been shown to contribute approximately 50% of the nutrient requirements of primary production (Pilskaln et al. 1998). Therefore, nutrients released from trawling have potentially important implications for regional nutrient budgets. As central west North Sea fishing is seasonal (high trawl frequency between September and April), it follows that trawl-induced nutrient inputs also exhibit a similar seasonal pattern.

Spring and autumn productivity blooms are typical in the North Sea (Otto et al. 1990). Concentrated intense trawl activity within strict spatial (determined by the fishers preferred fishing grounds) and temporal scales could however exert a regional affect on nutrient dynamics. The addition of large quantities of trawl-induced nutrients could act to extend the spring and autumn blooms, increasing overall production. We have demonstrated high fishing intensity to reverse the flux of some nutrients between the sediment and water column.

Scaling up our results to the trawl impacted sediments of ICES statistical rectangle 39E8 (based on calculations from raw fishing effort hours data, average gear width and tow speed) gave annual fluxes of NO_2^-, NO_3^-, PO_4^{3-} and NH_4^+ of approximately 50×10^{-11}, 10×10^{-13}, -28×10^{-12} and 12×10^{-12} (μmol km^{-2} yr^{-1}) respectively. The percentage flux attributable to trawl ground gear, under the heavy fishing scenario, compared to the annual background flux was 26.0 % for NO_2^-, -1.0% for NO_3^-, -15.0 PO_4^{3-} and 475.0% for NH_4^+. Negative values represent a shift in the direction of flux across the sediment-water interface. As a result, NO_3^- was taken up by, and PO_4^{3-} was released from, the sediment following trawling in contrast to the respective efflux and influx exhibited by these nutrient species in undisturbed sediments.

Within the North Sea, fishers tend to target specific areas, and trawl frequencies can be as high as several times daily. Although different trawlers often target the same area, a realistic time period for one trawler to target an area is approximately four days before moving on to another area (Catchpole, pers. com.). The results of this study therefore, suggest that during periods of intense fishing, North Sea sediments in fished areas may be in a continuous state of perturbation.

It is clear that trawl-induced nutrients can potentially have significant affects on shelf sea environments and both benthic and pelagic ecosystems. This study has confirmed that trawl activity supplies a substantial amount of remineralised nutrients to the pelagic zone while also altering the magnitude and direction of flux across the sediment-water interface. The study of season-wide nutrient dynamics between trawled and untrawled areas remains an important component of research on the effects of trawling on benthic biogeochemistry (see chapter 4).

This study has increased our knowledge of how benthic nutrient dynamics are affected by trawling activity. Certain areas however still need attention. Methods need to be established that could take *in situ* measurements over a large temporal scale, without equipment damage by on-going trawl activity. Further trawl impact studies concerned with benthic biogeochemistry and concomitant consequences to infaunal populations are needed (see chapter 6).

Chapter 6:

To what extent do trawling induced changes in macrobenthic assemblages affect benthic nutrient dynamics?

6.1 Abstract

The effects of changes in the composition of benthic macrofaunal assemblages on benthic nutrient dynamics were examined in a series of laboratory microcosm experiments using North Sea sediments collected from trawled ($55^0$13.55'N $01^0$27.28'W) and untrawled ($55^0$12.64'N $01^0$27.20'W) areas. The principal bioturbating species were first identified and removed from the sediments, and then reintroduced in strictly controlled numbers in order to manipulate macrofaunal density. The species present in the untrawled fauna were 25.1 % bigger than those in the trawled, while mean macrofaunal abundance was 59.8 % greater in trawled sediments. This means that the greater bioturbation activity of the large species in untrawled areas may be compensated for by greater numbers of individuals in trawled areas. To quantify this microcosms comprising the following treatments were established: no macrofauna, natural macrofaunal density (times 1), times 4 and times 8 densities, for both trawled and untrawled sediments and were overlayed with a continuous water flow of filtered coastal seawater. Following a stabilisation period, of 10 days, the continuous flow water system was turned off and inorganic macronutrients (NH_4^+, PO_4^{3-}, NO_3^- and NO_2^-) were measured hourly for 45 hours and used to derive benthic nutrient fluxes. Compared to treatments without bioturbators, natural densities increased the flux of all measured nutrients across the sediment-water interface by up to 81 % NH_4^+, 197 % PO_4^{3-},

96 % NO_2^- and 33 % NO_3^-. The magnitude of flux increased from molecular diffusion rates through trawled to untrawled fauna at natural density levels. NH_4^+ and NO_3^- flux rates increased with increasing density of fauna. PO_4^{3-} fluxes declined at each faunal density increase towards the diffusional flux rate in systems without macrofauna. The maximum NO_2^- flux occurred at times 4 natural density levels (88 (± 4) µmol m^{-2} d^{-1}). Untrawled and trawled fauna treatments displayed a similar pattern of flux rate increase in relation to increasing density for NH_4^+, PO_4^{3-} and NO_3^-. However, the magnitude of flux rate (calculated from the slope of the plotted linear regression line) was greater in untrawled treatments when compared to the same faunal density in trawled treatments.

6.2 Introduction

Benthic fluxes of inorganic nutrients resulting from the remineralisation of organic matter have recently become a major focus for international research because of the potential contribution to nutrient budgets and hence primary production (Pennifold and Davis 2001). This potentially significant nutrient supply has stimulated numerous investigators to assess the role of regenerated nitrogen and phosphorous fluxes in the overall nutrient supply in estuaries, enclosed bays and intertidal areas (Rao and Berner 1993, McCaffrey et al. 1980, Morin and Morse 1999, Rizzo and Christian 1996, Rutgers Van der Loeff 1980a). Benthic flux rates are influenced by a combination of chemical, biological and physical processes and interactions (Hines and Jones 1985), yet, the influence of macrofaunal communities on shelf sea benthic fluxes has often been overlooked or inadequately accounted for. Direct *in situ* measurements of

sediments and the prediction of ion fluxes using diffusive modelling of the sediment, cannot fully account for the role of faunal communities, as they fail to isolate the biogenic contribution to the fluxes. However, while microcosms may be limited spatially, laboratory systems do allow the strict control of environmental conditions and easy access for manipulation of experimental treatments where organism interactions can be isolated and their influence on biogeochemical cycling determined.

For estuarine, near shore, intertidal and shallow water sediments, numerous studies have been carried out on modelling / understanding the role of the biota in porewater chemistry and benthic fluxes (Hines et al. 1982, Davey and Watson 1995, Cermelj et al 1997, see Nedwell et al. 1999 for a review). However, shelf sea environments are less studied. An unfortunate consequence of estuarine and intertidal areas is that they are often influenced by fluxes of anthropogenic nutrients and terrestrial run off (Fichez et al. 1992). As a result, measured flux rates in these environments may not be applicable to shelf areas. Other studies concerned with benthic disturbance have primarily focused on the role of physical disturbances in the form of tidal currents and waves (Blackley and Rickards 1992, Kendrick et al. 1998). However, it is now recognised that in certain areas the main physical factor impacting the benthos is trawling (Frid et al. 1999, Percival and Frid 2000). Shelf sediments are often intensively trawled and can be highly productive (Pilskaln et al. 1998), thus providing the potential for significant modification of the overall nutrient regime which could impact primary production locally or regionally. Within the Gulf of Maine, for example, Pilskaln et al. (1998) has shown that the potential contribution from trawled sediments to the nutrient regime of overlying water is up to 50 %.

The central and northern North Sea accounts for approximately 5 % of the global fishing catch (Heessen 1988), hence, North Sea fishing activity can be regarded as intense (Jennings et al. 2000). Typically, North Sea fishing grounds are shallower than 60m, and are not significantly anthropogenically impacted by estuarine run off (Lindeboom and de Groot 1998). It is within these areas where altered benthic nutrient regime may be biogeochemically important.

There are two main aspects to consider immediately following a trawl impact; the physical impact to the substrate and the impact on benthic fluxes. Recent studies on the effects of fishing have addressed the direct physical impact of trawl gear (Watling and Norse 1998, Chapter 5). Key effects shown to date recognise the impact of trawl gear on the benthic habitat in terms of reducing physical structure and complexity (Watling and Norse 1998, Turner et al. 1999). The periodic disturbance to the seabed by trawl gear can also directly impact benthic fluxes (Percival and Frid 2000). Following disturbance, sediment and those nutrients remineralised within the sediment are resuspended and typically carried by horizontal transport (Watson and Frickers 1995). Consequently porewater nutrients are released in a pulse, while the direct ploughing effect of the trawl gear potentially exposes deeper hypoxic sediments to oxygen rich surface waters. This may further alter nutrient regeneration, as remineralisation rates of many nutrient species are dependent on specific redox conditions and the availability of organic compounds (Wilken et al. 1990). To date however, there has been only limited work on benthic biogeochemistry that may be affected indirectly through faunal alterations (Widdicombe and Austen 1998).

Bioturbator function is crucial in maintaining bioturbation output; yet, studies have shown bioturbation activity to also be dependant on the size of the organism, with larger individuals of the same species having increased burrowing activity (Wheatcroft et al. 1990). In benthic environments where natural levels of disturbance are low, the community can often be dominated by a few, slow growing, large species (Jennings and Kaiser 1998) and as a result have a high bioturbation output. Kaiser et al. (2002) demonstrated that changes to the species composition and/or abundance of faunal communities have the potential to impact benthic recycling rates and fluxes of inorganic macronutrients. Therefore any activity that can alter body size structure could significantly affect nutrient stoichiometry in sediments.

Trawling can reasonably be expected to be one such activity that can potentially alter benthic size distributions due to its direct ploughing action through the sediment. Trawl activity has been shown to cause a loss of larger species from the direct impact of the trawl gear (Kaiser et al. 2000). Disturbance induced mortality is generally size dependent, within and between species (Jennings et al. 2001). Size and abundance variations within benthic macrofauna have been investigated in mud and sand sediments (see Chapter 2). It was estimated the mean size of untrawled organisms to be larger by an average of 43.5 % (sand) and 28.0 % (mud) when compared to trawled sediments that displayed similar sediment characteristics. Trawled sediments have also been shown to exhibit a 29.0 % (sand) and 33.4 % (mud) increase in abundance of macrofauna (Chapter 2). Recent studies have also shown the composition of macrofaunal assemblages to be significantly altered due to the impact of mobile fishing gears because trawl-induced mortality is often

positively correlated with the size of the organism (Kaiser et al. 2000, Jennings et al. 2001). Engel and Kvitek (1998) concluded that in areas of repeated trawl activity, assemblages tend to shift from those dominated by large, slow reproducing species, to ones comprising smaller organisms with faster reproductive rates. The impact of trawling will therefore have impacts on bioturbation activity which will consequently affect the biogenic contribution to nutrient recycling. Therefore an important question is whether infaunal assemblages within trawled sediments contribute a different flux of nutrients in terms of quantity and mix of nutrient species between the sediment-water interface compared to untrawled fauna. Precise density manipulations of trawled and untrawled macrofauna along with comparisons to gradient driven diffusive fluxes are needed to elucidate the specific contribution to benthic nutrient dynamics from infaunal assemblages.

In this investigation the contribution to benthic nutrient dynamics of macrofauna from both trawled and untrawled areas of the central North Sea is assessed. It specifically aims to isolate and quantify any changes reflected in benthic nutrient fluxes as a result of the known effect trawling has on altering the assemblage of benthic organisms (see Chapter 2).

6.3 Method

6.3.1 Field sites

Untrawled and regularly trawled areas of the North Sea were identified using fishing data (provided by the UK Department for Environment, Food and Rural Affairs). The untrawled site, centred on 55^{0}12.64'N 01^{0}27.20'W, covers an area approximately 100m^2 and comprises a U-shaped area surrounded by

shipwrecks and a rocky reef (Fig 6.1). These physical obstructions prevent the use of towed fishing gears. To avoid any anomalies caused by altered benthic flow dynamics due to the presence of these features, samples were collected at the centre of the untrawled area (Hall et al. 1993b). The nearby (~ 2 km) regularly trawled area was centred on $55^{0}13.55'N$ $01^{0}27.28'W$.

6.3.2 Feasibility studies

Evaluation of the relative roles of infaunal organisms in controlling sediment nutrient distributions required their removal by sieving in some experiments. Such procedures have the potential to significantly modify benthic redox status, with implications for nutrient distributions and fluxes. In order to assess this, feasibility trials were conducted in which two systems, each comprising three replicate microcosms, were established using sediments from trawled and untrawled areas. Following a ten-day stabilisation period nutrient concentrations were measured and fluxes estimated (see below for nutrient methods). The duration of each trial sampling period was 45 hours. Each system was then sieved (500μm) to remove all macrofaunal species. Organisms were identified, to family level, and those known to have a bioturbation, siphon extension and/or tube building capacity were selected and returned to the sediment (Appendix 3) (Hayward et al. 1996, Hayward and Ryland 1998). This was done in order that the reduced fraction of retained macrofauna would preserve/maintain the majority of bioturbating activity and physically stimulate sediment-water nutrient exchange. Following a further ten-day stabilisation period, nutrient concentrations were again measured, fluxes estimated and compared to the initial control values.

Sediment and infaunal collections for the feasibility trials and experimental treatments were carried out during the summer of 2001 when the organisms were at their most active because of high bottom temperatures (Landen and Hall 1998).

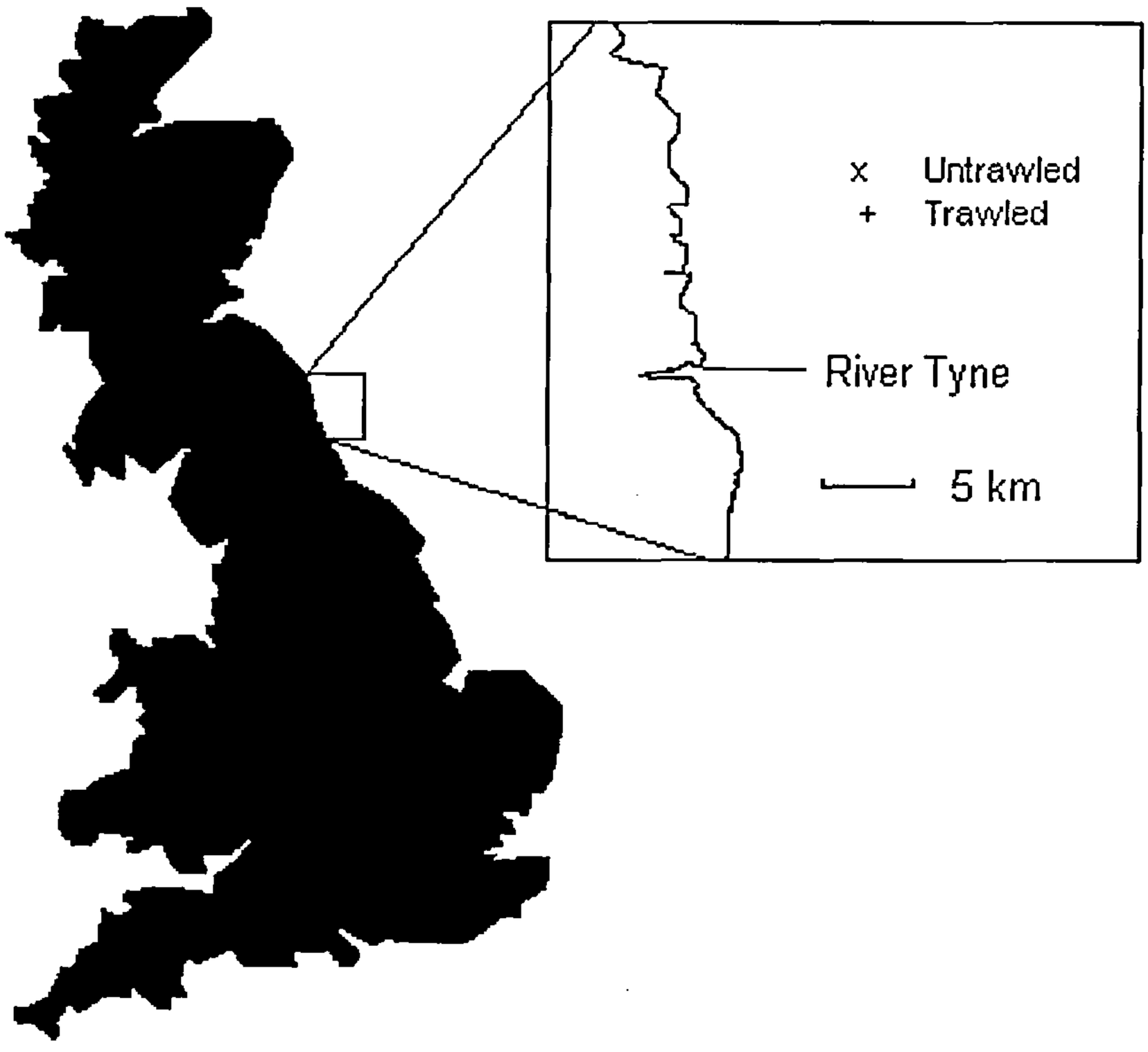

Figure 6.1. Coastal outline of north-east England and sampling sites within the central-west North Sea. Untrawled (x) and trawled (+) sediment sites illustrated (untrawled = 55^{0}12.64'N 01^{0}27.20'W, trawled = 55^{0}13.55'N 01^{0}27.28'W).

6.3.3 Sediment handling and set up procedure

Sediment samples (0.1 m^2) were collected from both sites with a van Veen grab and transferred on board, with minimal disturbance, into a series of 30 cm deep, 37.5 cm x 21 cm covered microcosms. These were maintained under the research vessels' continuous water flow system and transported to

the laboratory. The total elapsed time from sampling to microcosm establishment in the laboratory never exceeded 3 hours.

Within the laboratory a regulated continuous flow of filtered seawater overlay each sediment system. The water flow was maintained at a rate of approximately 45 lhr^{-1} and allowed to run to waste. Water circulation and aeration rates were controlled using a 45 degree splash plate, positioned 4cm above the water surface, on to which the continuous water flow was directed. This allowed oxygen to be introduced into the system and the inflow was maintained at a rate below that of visible resuspension of sediment. *In situ* temperature (14.5^{0}C +/- 0.5^{0}C) and ambient light were maintained throughout with use of the temperature regulated laboratory aquarium. All microcosm systems were allowed a ten-day stabilisation period, in order for the chemical and biological processes to reach equilibrium prior to any experimental procedure.

The difference between nutrient concentrations and fluxes in the natural community and the sieved community were assessed using a one-way ANOVA with Tukey. Comparisons between nutrient concentrations from the natural community and the sieved community were shown to be not significant. As a result, it can be assumed that the selection of active bioturbators and the time period selected for stabilisation were satisfactory for the manipulated biological community and chemical processes to re-establish.

6.3.4 Experimental treatments

On the 15 August 2001 6 microcosms were established as above using sediment from the untrawled and six from the trawled area. Within 24 hours, a

further 60 grab samples were taken (30 untrawled, 30 trawled) and immediately sieved (500µm mesh). Densities were estimated by taking the mean number of individual bioturbators that were retained on the sieve for the untrawled and trawled sediments and scaled to appropriate density levels for the density manipulation treatments. Retained organisms were maintained in aerated seawater filled containers during transportation to the laboratory and added to the appropriate microcosms for density manipulation treatments. Sediment from three of the sieved grabs was retained and used to establish microcosms without macrofauna for measurement of diffusive nutrient fluxes. Seven separate experimental treatments were carried out with three replicate microcosms for each with an experimental duration of 45 hours (see Table 1 for list and abbreviations used). Density manipulations were employed to specifically isolate infaunal assemblage driven fluxes.

Table 6.1. Experimental treatments for trawled and untrawled sediments without fauna, natural density fauna, four times the natural density and eight times the natural density of fauna from trawled and untrawled areas. Symbol refers to the text abbreviation by which each treatment is referred to.

	Treatment	Symbol
1	molecular diffusion of nutrients (sediments without macrofauna)	(MD)
2	Natural density of untrawled macrofauna	(NU)
3	Natural density of trawled macrofauna	(NT)
4	X4 the natural density of untrawled macrofauna	(4U)
5	X4 the natural density of trawled macrofauna	(4T)
6	X8 the natural density of untrawled macrofauna	(8U)
7	X8 the natural density of trawled macrofauna	(8T)

6.3.5 Sampling procedure: Water sampling

Prior to the start of each microcosm experiment, replicate samples were taken from the continuous flow seawater system, for the analysis of NO_2^-, NO_3^-, NH_4^+ and PO_4^{3-}. These were subsequently compared to field bottom water and

initial microcosm concentrations in order to rule out any possible contamination arising from within the system. One hour prior to experimentation, the water flow system was shut off and remained so for the duration of the experiment. Overlying water from within each microcosm was sampled over a 45-hour period using acid cleaned, 50ml plastic syringes, filtered through 0.45µm filters (Millipore type HA) into gas tight polypropylene bottles and immediately frozen for subsequent nutrient analysis.

6.3.6 Nutrient analysis

Concentrations of NO_2^-, NO_3^-, NH_4^+ and PO_4^{3-} were determined with an automated nutrient analyser (Skalar Sim^{plus}), following standard protocols (Brewer and Riley 1965; Mantoura and Woodard 1983; Kirkwood 1989). Following analysis, any differences in nutrient concentrations between treatments were assessed using a one-way ANOVA with subsequent Tukey procedure. Analytical precisions ± 1 % were routinely achieved for all methods. For full details of nutrient analysis refer to chapter 3.

6.3.7 Sediment characterisation

At the end of each experiment, replicate core samples were collected from each microcosm and analysed for grain size, percentage organic matter and porosity. Sediment particle size was determined using a standard gravity sedimentation technique (Buchanan 1984). To account for the possible presence of mineral carbon (coal), organic matter was determined by wet digestion (Buchanan 1984). Porosity was determined by weight loss on drying (80^0C).

6.3.8 Biomass calculation

After each experiment all macrofaunal organisms were removed using a 500µm mesh sieve. These organisms were then decalcified using a 10% formic acid solution for 48 hours. Following 48 hours, the samples were blotted dry and weighed to give biomass in grams.

6.4 Results

6.4.1 Sediment characteristics

Sediments collected from trawled and untrawled areas did not vary significantly in porosity (W = 12, p = 0.66), grain size (median grain size = 3.5 phi (Wentworth 1922)) or percentage organic content (W = 6, p = 0.081).

6.4.2 Inflow system and initial nutrient concentrations

The analysis of inflowing water used to check for possible contamination within the water flow system showed no significant nutrient concentration differences to the field (correction factor applied), laboratory flow through supply or initial concentrations within each microcosm for all nutrients tested (Appendix 4). Initial microcosm concentrations of NH_4^+ ranged from ~ 2 to ~ 3µmol L^{-1}, all PO_4^{3-} concentrations were < 1.5µmol L^{-1}, the NO_2^- concentration was approximately 0.25µmol L^{-1} and initial NO_3^- ranged ~ 4 to ~ 5µmol L^{-1}. Therefore the results can be attributed to the differences arising from the treatments.

6.4.3 Microcosm nutrient concentrations

The NH_4^+ concentrations in the molecular diffusion (MD) controls remained constant during all experiments (Fig 6.2). All natural density (NU and

NT) microcosms exhibited NH_4^+ concentrations slightly above MD levels throughout the experiments, reaching ~ 4 (± 0.2) µmol after 45 hours (Fig 6.2). The concentrations within the times 4 density manipulations (4U and 4T) increased approximately linearly through the experiment reaching ~ 9µmol L^{-1} at the end of the experiment (Fig 6.2). NH_4^+ concentrations within the times 8 untrawled density (8U) displayed the same pattern expressed by the times 4 density systems up to 37 hours, but then increased from ~ 9 to ~ 13.5µmol L^{-1} after 45 hours (Fig 6.2). Within the times 8 trawled system (8T) the concentration rose steeply, (approximately 1µmol $L^{-1}hr^{-1}$) between 1-5 hours, and (approximately 0.5µmol $L^{-1}hr^{-1}$) between 13 - 21 hours, after which time concentrations levelled out at ~ 11.5µmol L^{-1} to 45 hours. As the experiment progressed beyond 5 hours NH_4^+ the diverging concentrations between different treatments became statistically different. At each subsequent sampling time beyond 5 hours, the number of treatment concentrations that were significantly different increased (Appendix 4).

The pattern of different treatments diverging with time occurred for all nutrient species. However the time and number of treatments that diverged was varied and a full statistical summary is included (Appendix 4a).

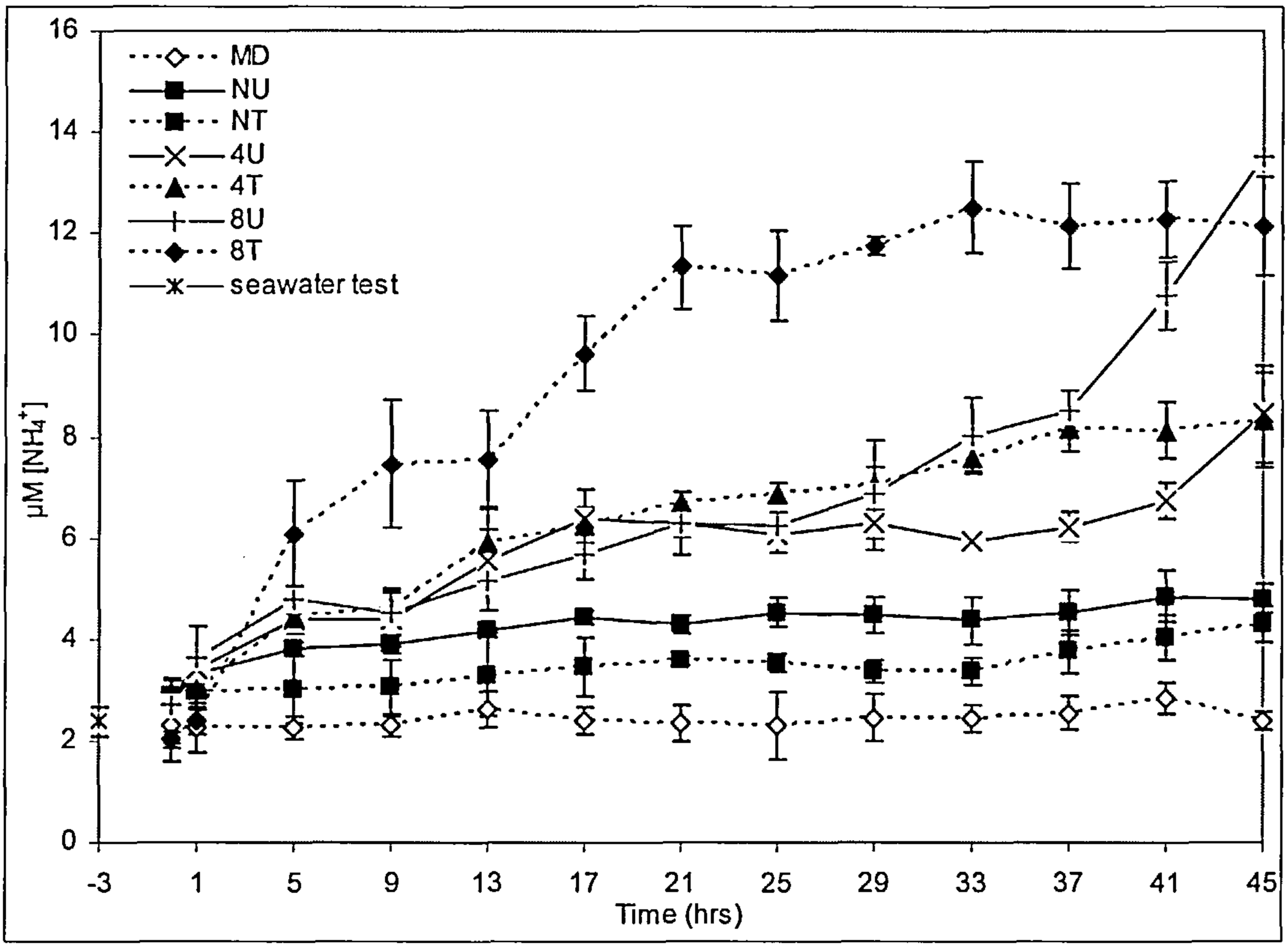

Figure 6.2. Microcosm NH_4^+ concentrations for each of the seven treatments (molecular diffusion, natural untrawled density of fauna, natural density of trawled fauna, times four untrawled density, times four trawled density, times eight untrawled fauna and times eight trawled fauna) and the *in situ* seawater test (denoted by x at time -3).

NO_3^- exhibited the greatest increase, to ~ 18µmol L^{-1} after 45 hrs within the MD controls (Fig 6.3). The microcosms containing NU and NT, while maintaining lower concentrations than MD, also displayed a steady increase over the duration of the experiment to ~ 16µmol L^{-1} (Fig 3). Both 4U and 4T NO_3^- concentrations initially increased and then remained relatively constant at ~ 7µmol L^{-1} throughout the experiment (Fig 6.3). 8U and 8T, however, displayed a decline in NO_3^- concentration over the experiment. After 45 hours both system concentrations were ~ 1µmol L^{-1}, a decline of ~ 3µmol L^{-1} (Fig 6.3). Although there was some variability in the rate of increase, a general trend of NO_3^- concentrations diverging in each density treatment, that became significantly different over time, was apparent (Appendix 4b).

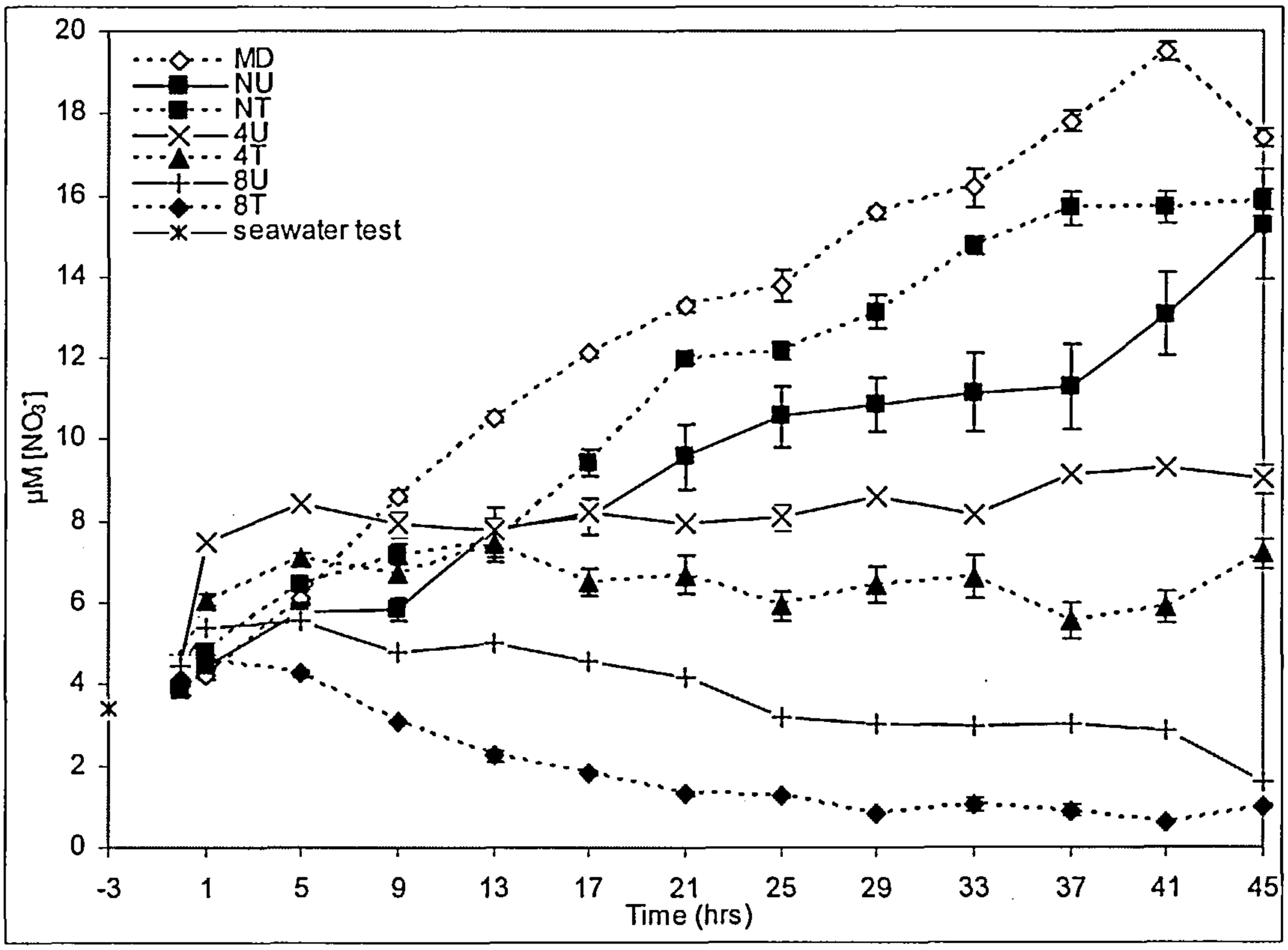

Figure 6.3. Microcosm $N0_3^-$ concentrations for each of the seven treatments (molecular diffusion, natural untrawled density of fauna, natural density of trawled fauna, times four untrawled density, times four trawled density, times eight untrawled fauna and times eight trawled fauna) and the *in situ* seawater test (denoted by x at time -3).

The MD controls were the only systems to exhibit a net gain in PO_4^{3-} concentration, increasing to ~ 4µmol L^{-1} by the end of the experiment (Fig 6.4). PO_4^{3-} for 8U fluctuated throughout the experiment but displayed no significant change from initial concentrations (Fig 6.4). The 8T treatment declined in concentration and was below that displayed by 8U (to ~ 2.0µmol L^{-1} after 45 hours). The 4U and 4T microcosm concentrations declined at a similar rate, maintaining a < 0.5µmol L^{-1} difference over the experiment, to ~ 1.5µmol L^{-1} at the end of the experiment (Fig 6.4). NU & NT displayed the greatest concentration decrease, both following a similar pattern of a steady decline (within 0.5µmol L^{-1} of each other) throughout the duration of the experiment to ~

0.75 after 45 hours (Fig 6.4). Significant differences in PO_4^{3-} concentrations between treatments occurred after 9 hours. At each subsequent sampling time, the number of treatments that were significantly different increased (Appendix 4c).

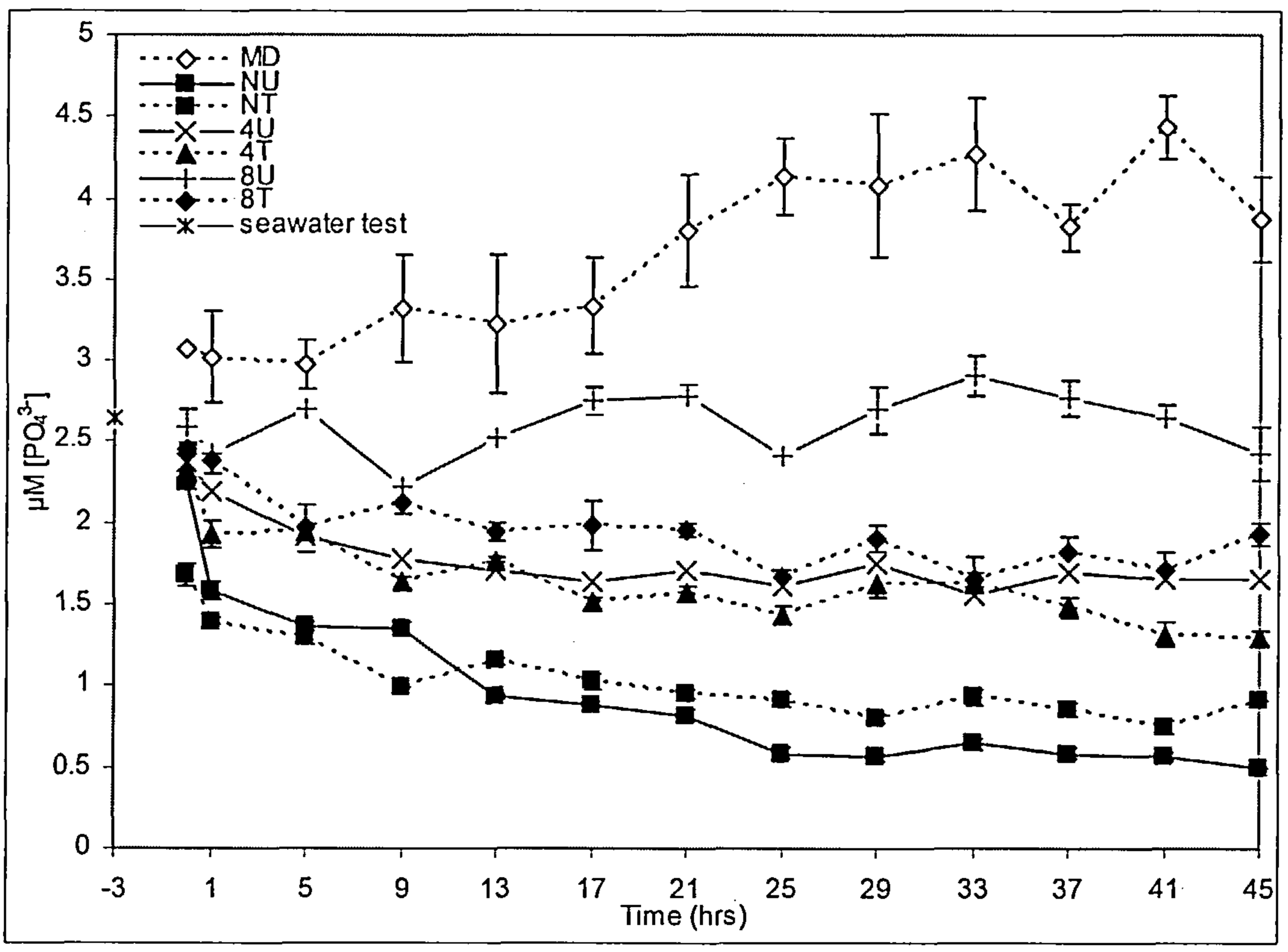

Figure 6.4. Microcosm PO_4^{3-} concentrations for each of the seven treatments (molecular diffusion, natural untrawled density of fauna, natural density of trawled fauna, times four untrawled density, times four trawled density, times eight untrawled fauna and times eight trawled fauna) and the *in situ* seawater test (denoted by x at time -3).

The concentration of NO_2^- within the MD systems displayed only a slight increase to ~ 1µmol L^{-1} after 13 hours (Fig 6.5). After this time the NO_2^- concentration maintained a steady decline to ~ 0.5µmol L^{-1} after 45 hours. NO_2^- concentrations within the 8U and 8T microcosms displayed a small increase throughout the experiment to ~ 0.6µmol L^{-1} (Fig 6.5). All natural density and times 4 density NO_2^- concentrations steadily increased over the duration of the experiment to ~ 2.0µmol L^{-1} (Fig 6.5). Within these trends; however, statistically

significant differences between concentrations within treatment microcosms occurred after 45 hours (between 8U and 8T, One-way ANOVA with Tukey, 8U vs 8T, f = 10.67, P < 0.001).

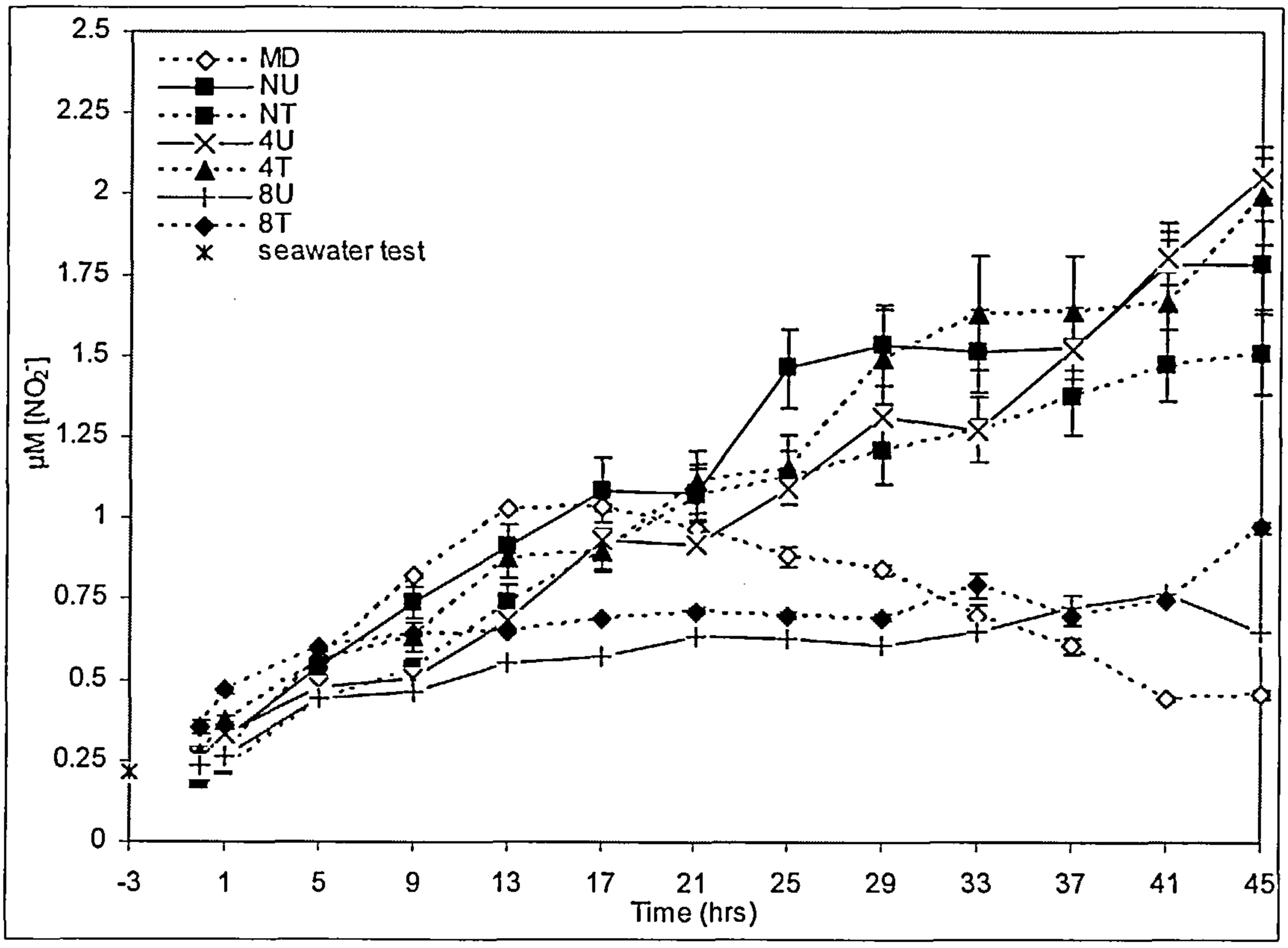

Figure 6.5. Microcosm $N0_2^-$ concentrations for each of the seven treatments (molecular diffusion, natural untrawled density of fauna, natural density of trawled fauna, times four untrawled density, times four trawled density, times eight untrawled fauna and times eight trawled fauna) and the *in situ* seawater test (denoted by x at time -3).

6.4.4 Treatment summary

An emergent pattern of diverging nutrient concentrations over time occurred for each nutrient species (Fig 6.2, 6.3, 6.4, and 6.5). These divergent patterns related to changes in macrofaunal density. NH_4^+ and NO_3^- systems displayed a similar pattern of diverging nutrient signal in relation to increasing macrofaunal density (Fig 6.2 and 6.3). Within the PO_4^{3-} and NO_2^- systems however, the pattern was reversed and the nutrient signal increased away from molecular diffusion rates in relation to decreasing macrofauna density (Fig 6.4

and 6.5). Therefore PO_4^{3-} and NO_2^- displayed increasing concentration changes, from the MD control systems to times eight then times four and finally natural density levels (Fig 6.4 and 6.5).

6.4.5 Mean daily nutrient fluxes (average flux over 24hrs)

If the rate of efflux / influx is related to bioturbation, and bioturbation increases with body size, then if the fauna in the untrawled area have a larger body size than in the trawled area it would be predicted that increasing efflux / influx with increasing density of fauna was higher in untrawled than trawled systems (untrawled flux > trawled flux). However, a second prediction can also be established. At higher manipulated macrofauna densities the high abundance increases in trawled sediments may be sufficient to increase the flux from trawled fauna above fluxes from untrawled sediments (trawled flux > untrawled flux).

The addition of macrofauna did cause increased fluxes away from diffusional flux rates. At natural density levels, increased fluxes rates were apparent from sediments containing untrawled fauna when compared to sediments containing trawled fauna (Fig 6.6). However, following density increases of X4 and above, the trawled fauna increased the magnitude of flux away from the MD rates compared to the same untrawled macrofaunal density for NH_4^+ and NO_3^-.

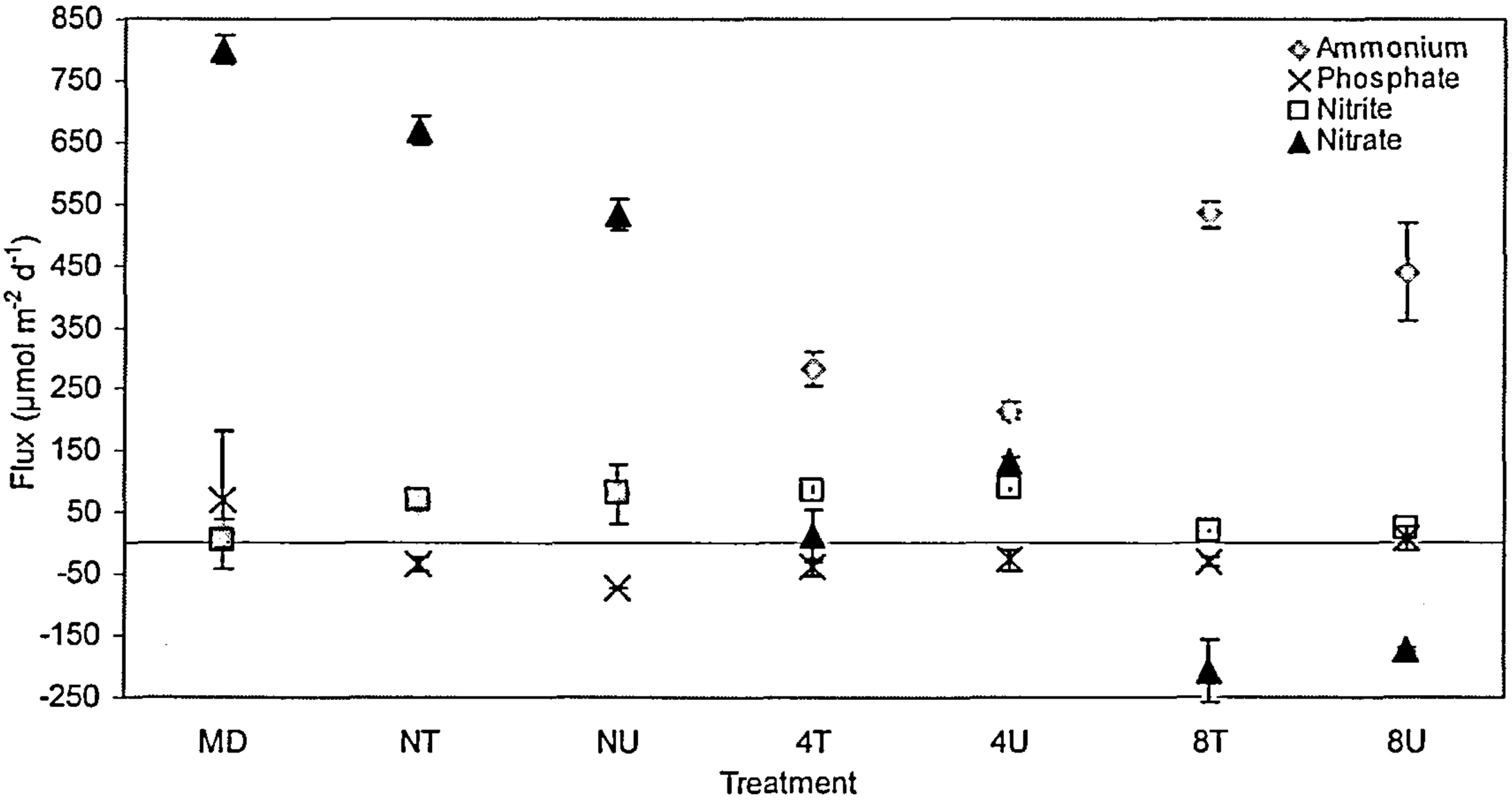

Figure 6.6. Daily mean, nutrient flux (µmol m^{-2} d^{-1}) (± standard deviation) for experimental treatments. MD = molecular diffusion, NU = natural untrawled density of fauna, NT = natural density of trawled fauna, 4U = times four untrawled density, 4T = times four trawled density, 8U = times eight untrawled fauna and 8T = times eight trawled fauna.

When the treatments are plotted in order of increasing density and biomass, it is interesting to note that a general trend of increasing flux with increasing density was apparent for NH_4^+ (Fig. 6.7 and 6.8). However, the rate of increase was greater for treatments containing untrawled fauna (Fig. 6.7 and 6.8). NO_2^- was greater in the untrawled systems compared to same level of trawled macrofaunal density (Fig. 6.9 and 6.10). The flux of NO_3^- declined within untrawled and trawled systems at each density increase (Fig. 6.11 and 6.12). PO_4^{3-} exhibited the greatest flux away from MD in NU and NT treatments (Fig. 6.13 and 6.14). As faunal density increased, within the trawled and untrawled treatments, the rate of PO_4^{3-} flux moved towards the MD flux. The magnitude of this flux rate was greater in systems containing untrawled macrofauna (Fig. 6.13 and 6.14).

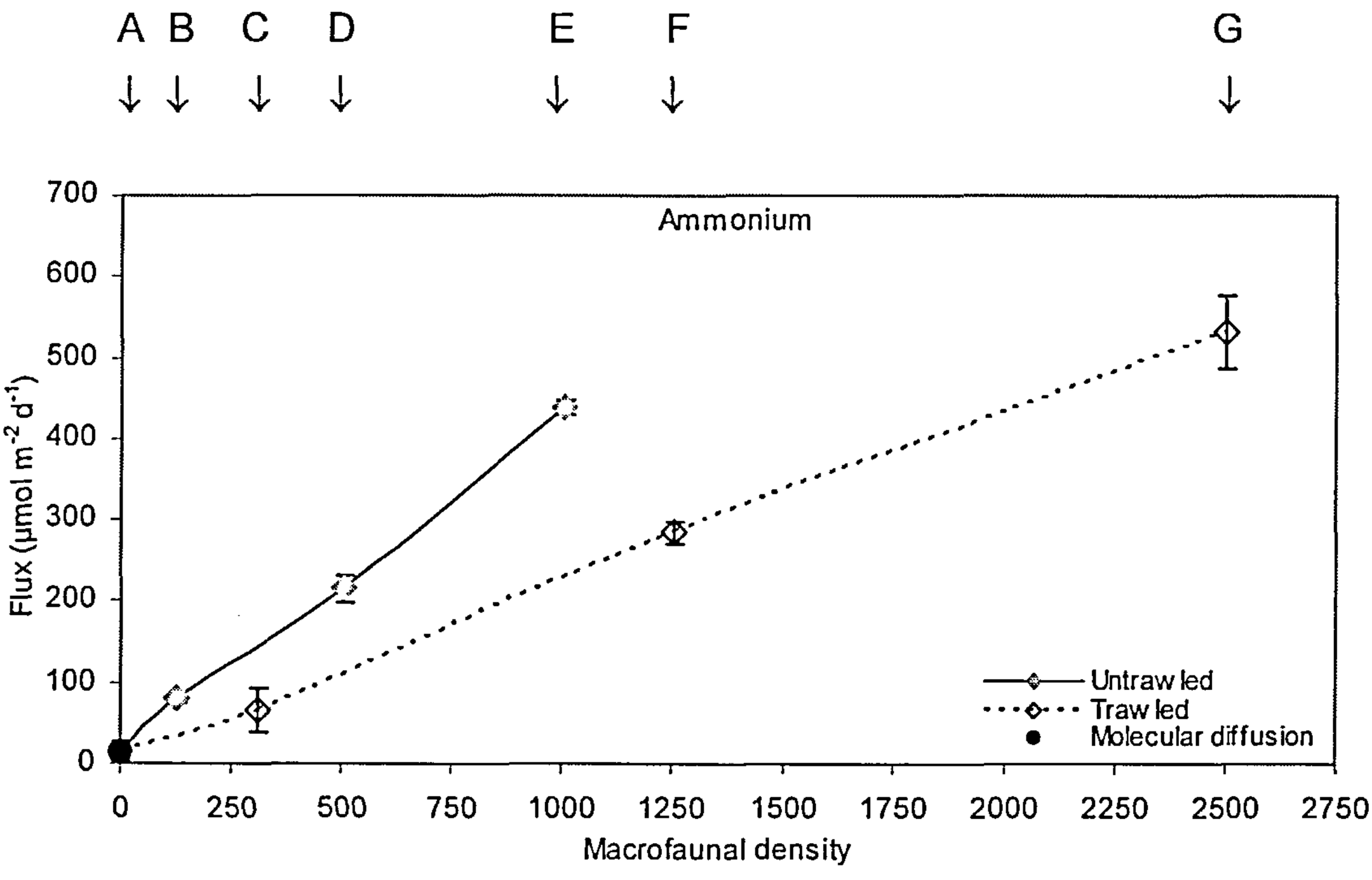

Figure 6.7. Macrofaunal density ($0.1m^2$) against ammonium flux (µmol m^{-2} d^{-1}) (± standard deviation). A = molecular diffusion, B = natural untrawled density of fauna, C = natural density of trawled fauna, D = times four untrawled density, E = times eight untrawled fauna, F = times four trawled density and G = times eight trawled fauna.

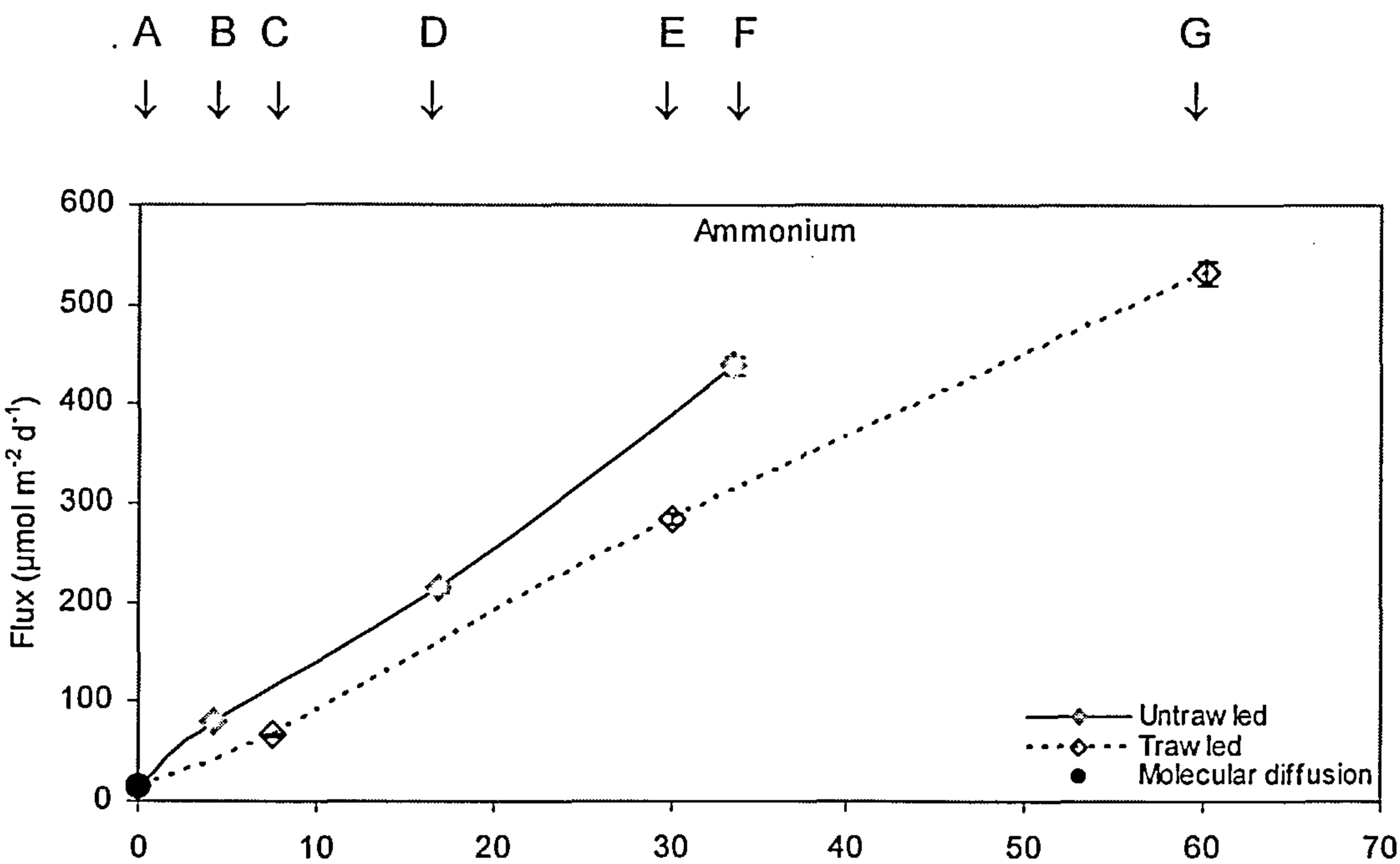

Figure 6.8. Macrofaunal biomass (g) against ammonium flux (µmol m^{-2} d^{-1}) (± standard deviation). A = molecular diffusion, B = natural untrawled density of fauna, C = natural density of trawled fauna, D = times four untrawled density, E = times four trawled fauna, F = times eight untrawled density and G = times eight trawled fauna.

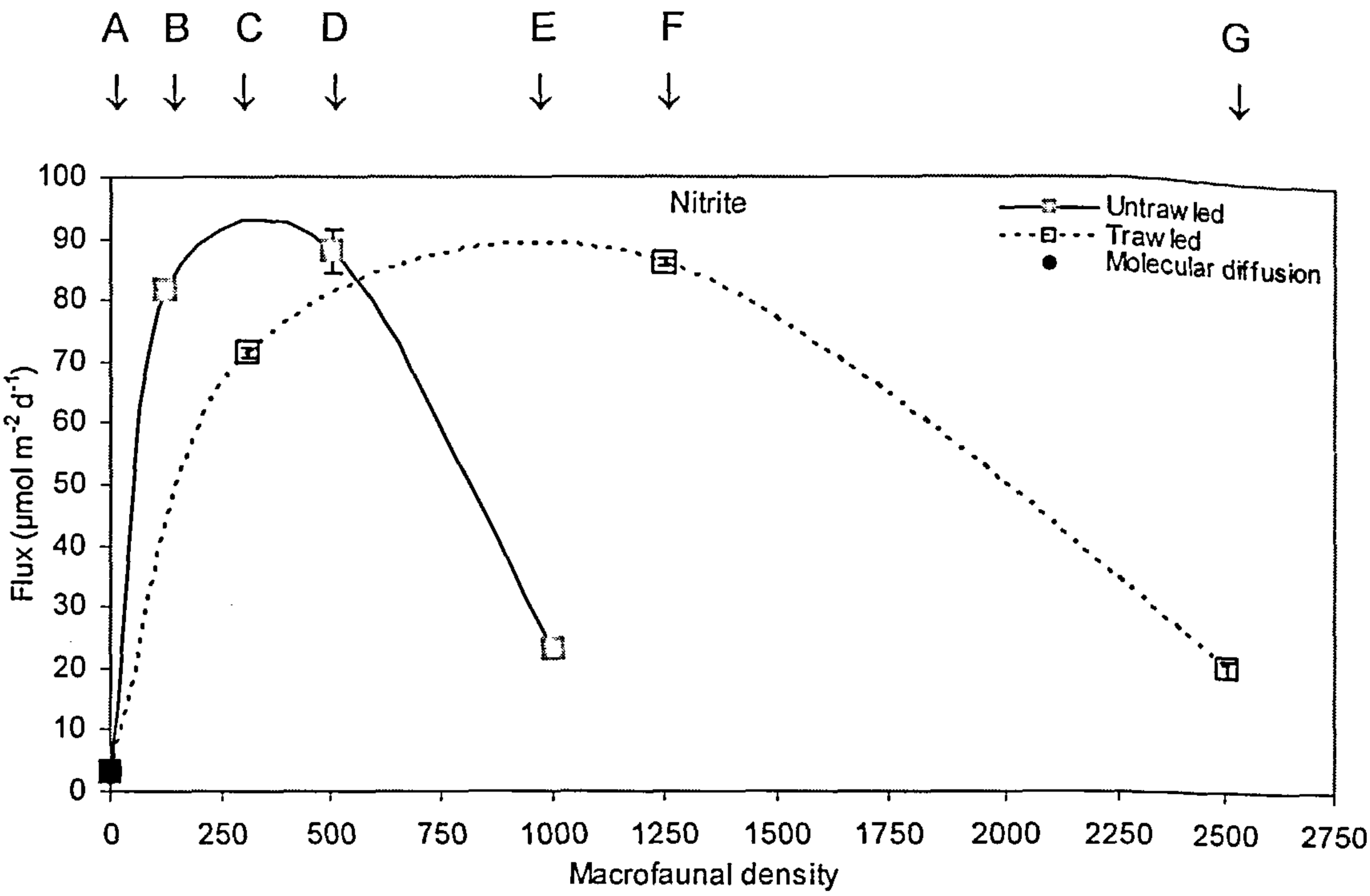

Figure 6.9. Macrofaunal density ($0.1m^2$) against nitrite flux (µmol m^{-2} d^{-1}) (± standard deviation). A = molecular diffusion, B = natural untrawled density of fauna, C = natural density of trawled fauna, D = times four untrawled density, E = times eight untrawled fauna, F = times four trawled density and G = times eight trawled fauna.

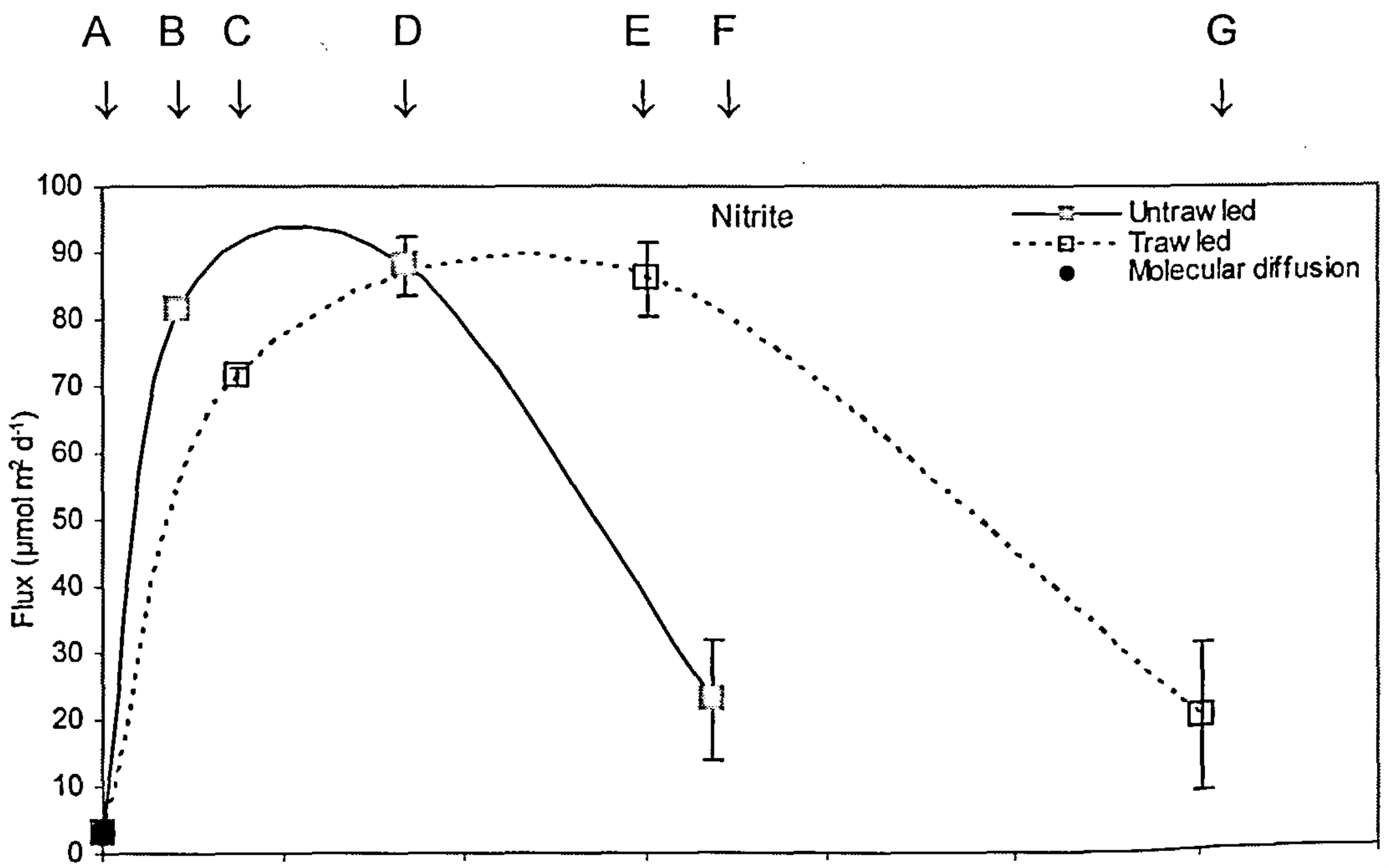

Figure 6.10. Macrofaunal biomass (g) against nitrite flux (µmol m^{-2} d^{-1}) (± standard deviation). A = molecular diffusion, B = natural untrawled density of fauna, C = natural density of trawled fauna, D = times four untrawled density, E = times four trawled fauna, F = times eight untrawled density and G = times eight trawled fauna.

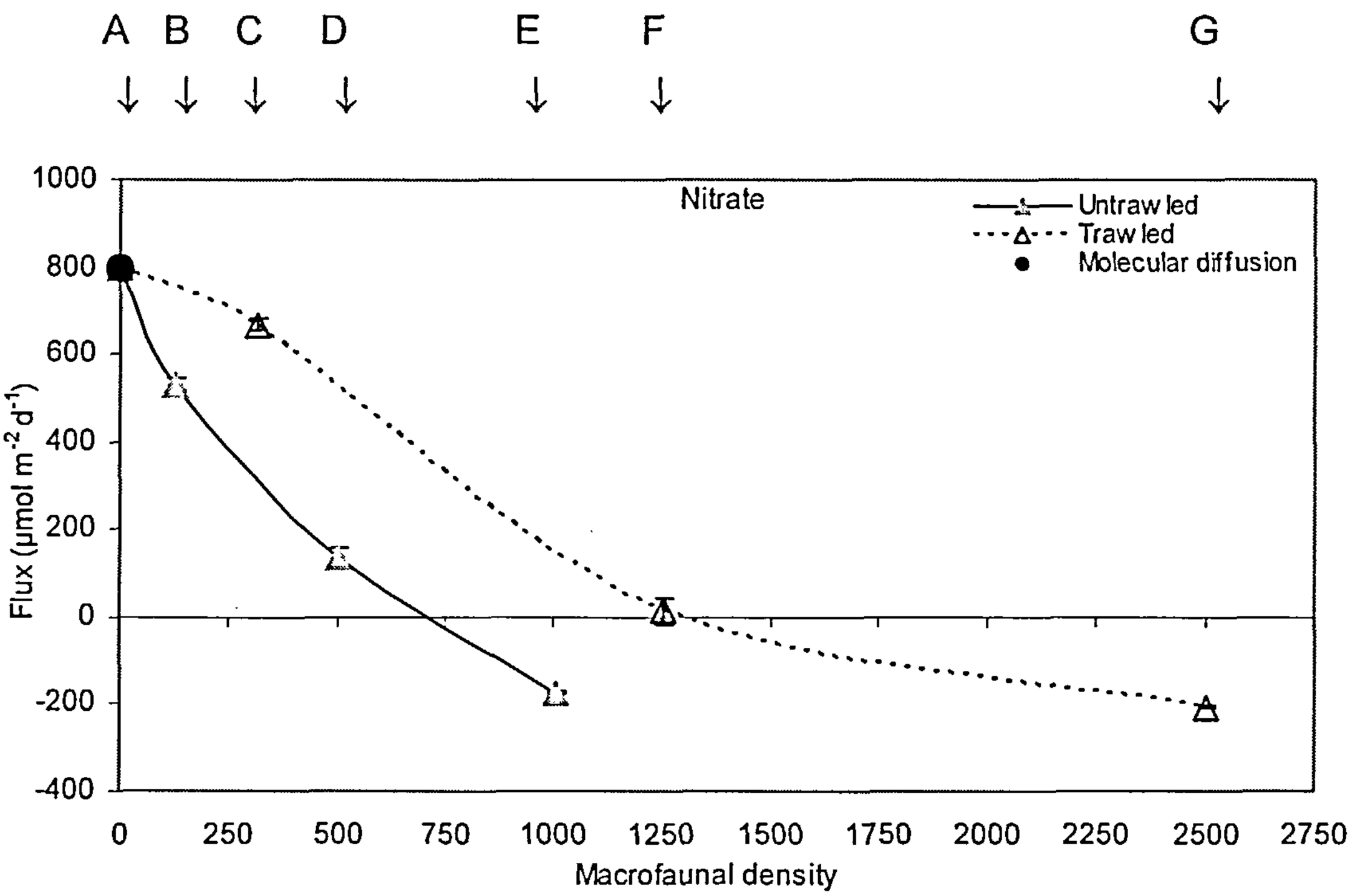

Figure 6.11. Macrofaunal density ($0.1m^2$) against nitrate flux (µmol m^{-2} d^{-1}) (± standard deviation). A = molecular diffusion, B = natural untrawled density of fauna, C = natural density of trawled fauna, D = times four untrawled density, E = times eight untrawled fauna, F = times four trawled density and G = times eight trawled fauna.

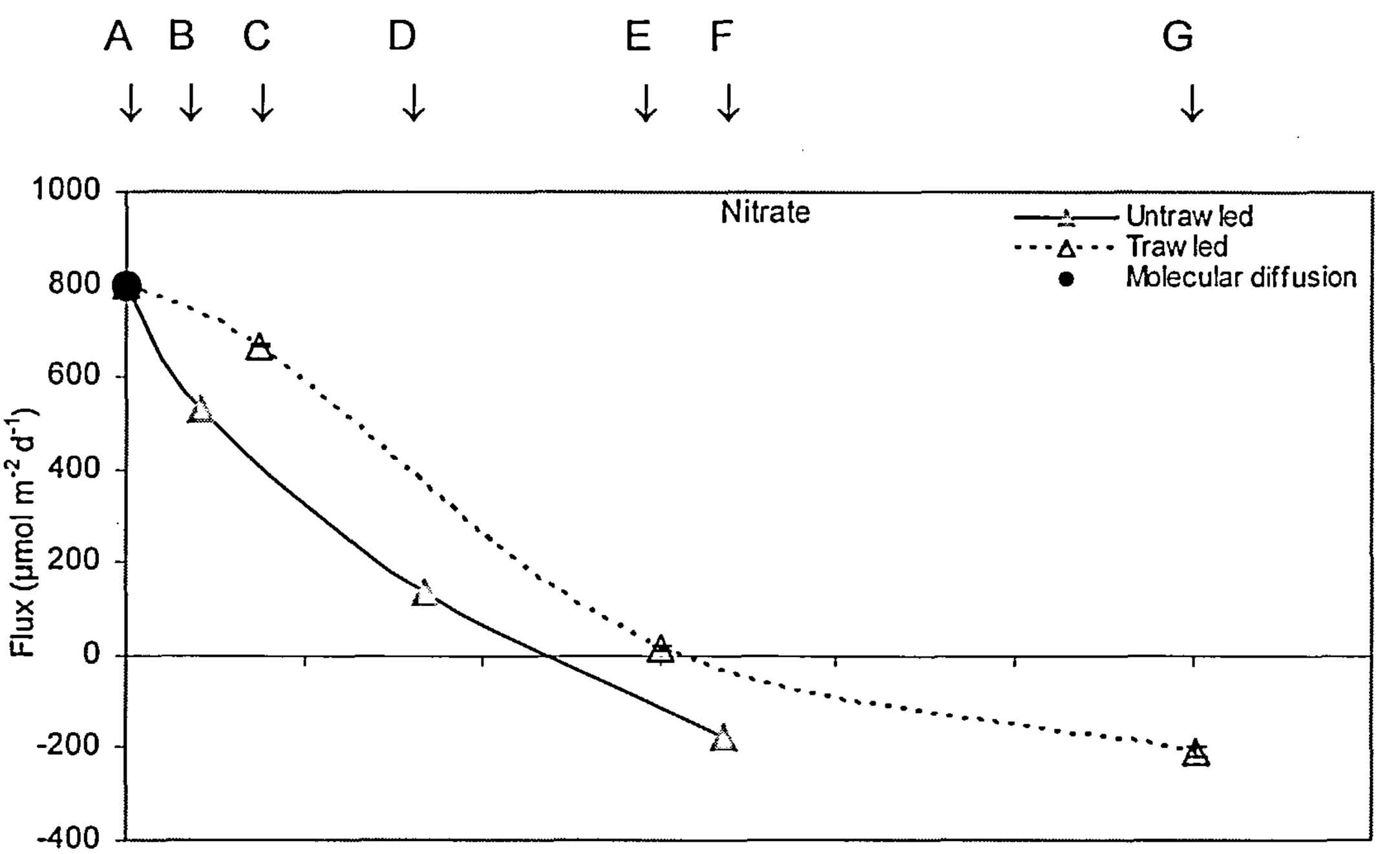

Figure 6.12. Macrofaunal biomass (g) against nitrate flux (µmol m^{-2} d^{-1}) (± standard deviation). A = molecular diffusion, B = natural untrawled density of fauna, C = natural density of trawled fauna, D = times four untrawled density, E = times four trawled fauna, F = times eight untrawled density and G = times eight trawled fauna.

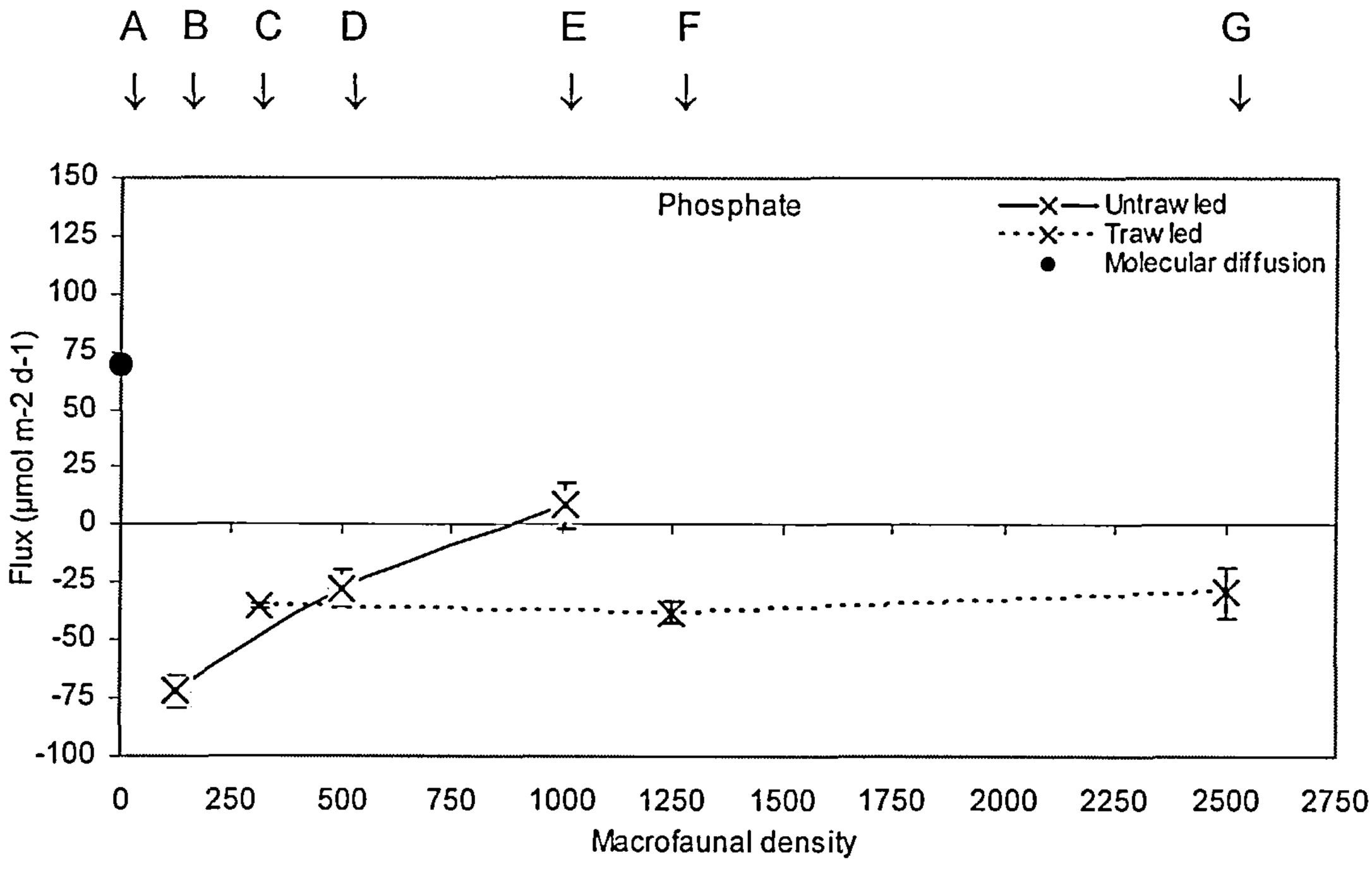

Figure 6.13. Macrofaunal density ($0.1m^2$) against phosphate flux (µmol m^{-2} d^{-1}) (± standard deviation). A = molecular diffusion, B = natural untrawled density of fauna, C = natural density of trawled fauna, D = times four untrawled density, E = times eight untrawled fauna, F = times four trawled density and G = times eight trawled fauna.

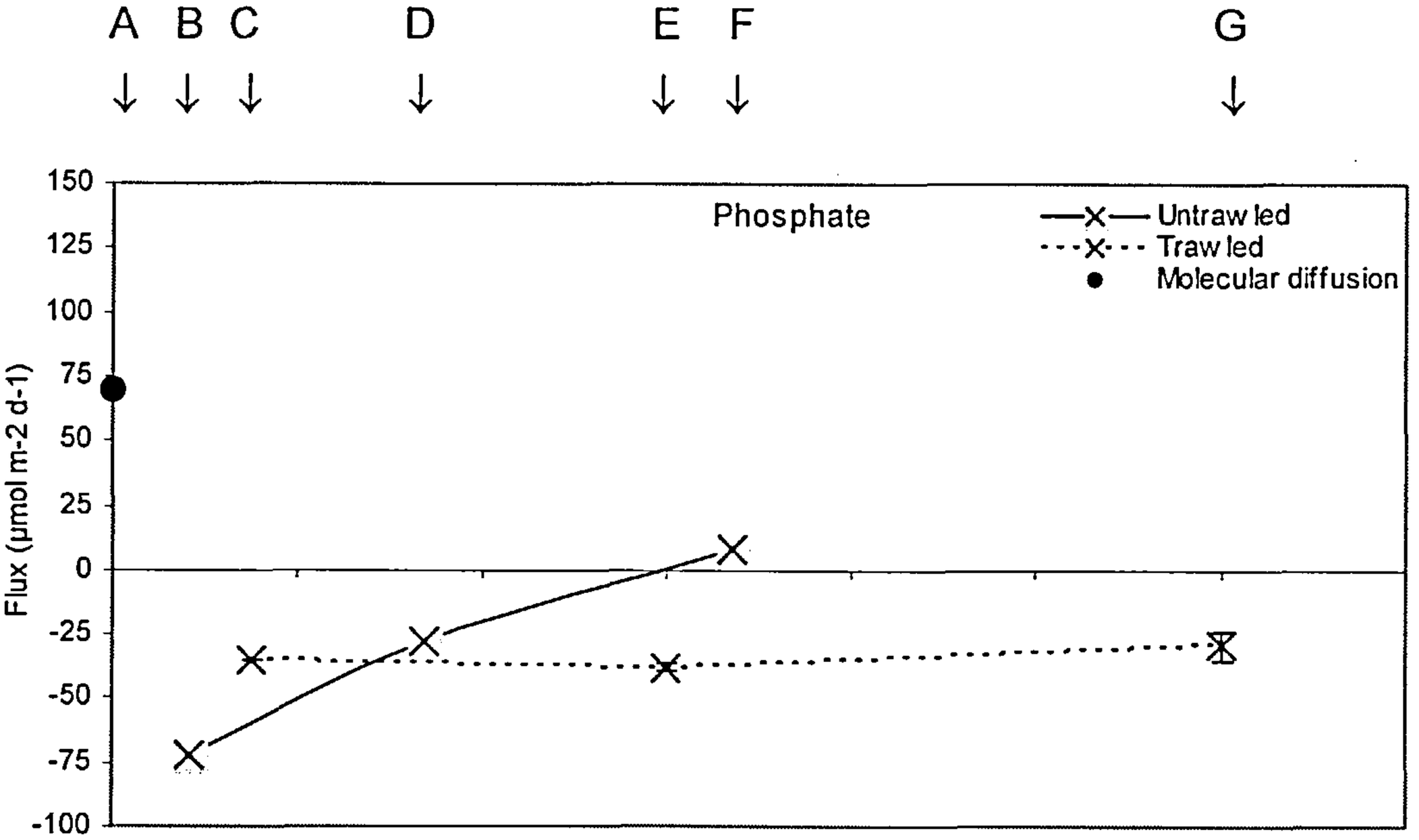

Figure 6.14. Macrofaunal biomass (g) against phosphate flux (µmol m^{-2} d^{-1}) (± standard deviation). A = molecular diffusion, B = natural untrawled density of fauna, C = natural density of trawled fauna, D = times four untrawled density, E = times four trawled fauna, F = times eight untrawled density and G = times eight trawled fauna.

6.5 Discussion

Bioturbation increased the magnitude of the four nutrient fluxes across the sediment-water interface above gradient driven diffusion fluxes (by between 77 - 97 % increase NH_4^+, 88 - 197 % PO_4^{3-}, 85 - 97 % NO_2^- and 20-487 % NO_3^-) irrespective of the assemblage of benthic organisms. Unquestionably, untrawled and trawl impacted macrofauna affect benthic chemistry differently. The presence of bioturbators can actively promote the transport of dissolved constituents across the sediment-water interface through a combination of active ploughing, tube building and vertical siphon extension through surficial sediments (Barbanti et al. 1992). This concept of a faunal contribution altering benthic fluxes compared to molecular diffusion is not new. Many studies (Hines et al. 1982, Barbanti et al. 1992, Schaffner et al. 1997, Widdicombe and Austen 1998, Berg et al. 2001, Duplisea et al. 2001) have shown bioturbation to be a factor in modifying benthic chemistry. However, these studies often focus on species specific bioturbation activity or include a bioturbation factor within a mathematical model (Berg et al. 2001, Duplisea et al. 2001). Yet, we have shown flux rates to differ in relation to the indirect impact of fishing gear altering the benthic faunal assemblage.

While it is difficult to isolate the specific bioturbation contribution from each species within a community, it is likely that both the size and number of benthic macrofaunal species are the key factors responsible for producing the nutrient signal. Recent studies have highlighted a significant shift in the assemblage of benthic organisms that are exposed to frequent trawl disturbances (Kaiser et al. 2000). As a result of repeated trawl impact, the size dependent mortality of benthic organisms can cause assemblage shifts from

ones comprising large, long-lived species, to smaller, faster reproducing species (see also chapter 2). Trawled communities therefore contain a greater number of smaller individuals that can utilise a fast reproductive output as a trawl resistance measure. Investigators have calculated bioturbation activity to increase with the log of mean organism size (Wheatcroft et al. 1990). Therefore, it would suggest that the larger organisms within untrawled sediments, although potentially fewer in number than species within trawled sediments, would exert a greater level of bioturbation activity. As a result nutrient flux across the sediment-water interface could also be enhanced. Obviously the nature of these microcosm experiments excludes the impact from very large scavenging macrofauna which can migrate over relatively large areas and can influence. However, the veracity of these results are preserved as they provide data on the permanent resident faunal community and ecosystem engineer species that structure and influence habitat and biogeochemical processes. The results of this study support this notion at natural macrofaunal density levels increased the mean daily nutrient flux of NH_4^+, NO_3^-, NO_2^- and PO_4^{3-} through trawled, to untrawled fauna. For example NO_2^- (µmol m^{-2} d^{-1}) increased from an average MD flux of 3.0 (± 1 standard deviation) to 71.5 (± 1) from sediments with trawled fauna to the highest flux of 81.5 (± 1) from sediments containing untrawled fauna (see Fig 6 for details of other nutrient species).

The presence, absence or altered density of macrofauna each revealed different effects on the measured nutrient species. The between NT and UT treatment differences observed could generally be explained by an increased surficial oxic layer as a result of bioturbation at the natural density of

macrofauna. Density manipulated treatments (times 4 and times 8), however, are likely to be subjected to a combination of competing processes.

Reducing sedimentary environments release soluble fractions of phosphate from degradation of phosphorous containing iron oxyhydroxides or via adsorption – de-sorption mechanisms (Moshiri and Crumpton 1978). Once released PO_4^{3-} will be transported across the sediment-water interface via passive diffusion into the overlying water column (Watanabe and Tsunogai 1984). This is undoubtedly the case in the molecular diffusion control treatments and has been demonstrated by other investigators (Widdicombe and Austen 1998). Bioturbated sediments however, acted to scavenge dissolved phosphate. Bioturbation increases the potential for oxygen to penetrate into anoxic sediment layers and consequently could cause phosphate sorption and co-precipitation with metal hydroxides to occur (Widdicombe and Austen 1998). This is consistent with the hypothesis that bioturbation activity, and hence oxygen penetration, increases with body size, as PO_4^{3-} exhibited a greater loss from solution in sediments containing untrawled fauna. However at organism densities greater than natural levels, PO_4^{3-} concentration and flux reduced. Increased organism densities are likely to increase re-working in deeper sediments. Consequently, a two-way transport pathway may act to carry oxygen, but also oxidised phosphate minerals, to depth while transporting soluble biologically available PO_4^{3-} into the water column. As shown above, increased oxygen would remove more PO_4^{3-} as it is co-precipitated out of solution, however, precipitated phosphate oxy-hydroxides would re-dissolve within deeper anoxic layers and flux out of the sediment. As a result the two competing processes could counter balance one another, thus maintaining the

PO_4^{3-} concentration and flux between systems without macrofauna and natural bioturbation levels.

If we assume the MD control systems exhibit relatively hypoxic conditions because of a lack of biological reworking and deeper O_2 mixing (Nedwell and Walker 1995), then NO_3^- levels could seem high. However, in some systems NO_3^- is likely to proliferate as it is the thermodynamically stable form of inorganic nitrogen in aerobic seawater (Spencer 1975). Under such conditions NO_3^- efflux could be stimulated if the overlying seawater contained relatively high O_2 concentrations (Cowan and Boynton 1996). Only a small surface micro-oxic layer would be needed for nitrification to occur and release NO_3^- to the water column (Lohse et al. 1993). Distinct (due to a lack of bioturbation) oxic, hypoxic and anoxic layers down the sediment column could aid NO_3^- formation and account for the relatively high average NO_3^- flux of 800 (± 13 standard deviation) µmol m^{-2} d^{-1} within the MD system.

As macrofaunal density increased, a general trend of increasing NH_4^+ flux was shown. An inverse relationship between NH_4^+ and NO_3^- indicated a net reduction in nitrification because NO_3^- decreased while NH_4^+ proliferated. Interestingly, as density / bioturbation increased it should hold that O_2 would be introduced to the sediment system and thus nitrification should persist. Bioturbation activity cannot create strictly homogenous sediment layers and therefore numerous redox micro-environments would occur (Forster 1996). Therefore, coupled nitrification / denitrification processes would simultaneously occur and under the circumstances of this experiment denitrification seemed to be the dominant process. Bioturbation into deeper sediment would potentially carry O_2 deeper, yet, there would likely be a lag period, depending on

temperature and number of nitrifying bacteria, before oxidative reactions could take place (Cavari 1977). A physical transport route for dissolved constituents would be temporarily created by increased burrowing activity at increased densities. The relatively high concentrations of NH_4^+ at depth from organic decomposition would use this pathway to flux out of the sediment. This explains the high NH_4^+ efflux values at increased macrofaunal density (i.e. trawled sediments had higher total abundance and therefore density compared to untrawled sediments). Temperature increases during the summer, leading to oxygen removal, are also likely to compound this effect in the field (Hines et al. 1982).

Another explanation for high fluxes of, NH_4^+ and NO_3^-, relating to higher density in trawled sediments is a potential conflict between bioturbation and bioirrigation. In the field density will alter due to the frequency and severity of disturbance impacts. As we have shown, total abundance increased in trawled sediments and the major dominating species was the Maldanidae polychaetes. High indices of polychaetes have been recorded in other heavily trawled areas (Tuck et al. 1998). However, while bioturbation has been shown to increase with increasing size, bioirrigation has been shown to increase with increasing density (Sanders et al. 2000). It could therefore be possible that at high macrofauna density levels where a high percentage of the community is dominated by polychaetes bioirrigation stimulates another major transport route of dissolved constituents across the sediment-water interface.

Increased densities of macrofauna are also likely to consume large quantities of meiofauna (Tita et al. 2000). Short generation rates and the likelihood to be resuspended instead of direct mortality could give meiofauna

increased trawl resistance (Schratzberger et al. 2002). Meiofauna are known to influence microbial and organic matter dynamics (Hakenkamp and Morin 2000). Changes in the quality and availability of organic matter, as well as microbe biotic activity, have the potential to alter inorganic nutrients. This may have a limiting effect on microbial transformations and further explain the reduced nutrient levels at macrofaunal densities above natural. Microbial limitation also supports the notion that at higher densities, bioturbation serves as a transport route for dissolved constituents. At natural levels of bioturbation it is likely that a chemical and biological equilibrium is achieved which becomes altered and achieves a new equilibrium with manipulated densities of larger organisms. Thus, indicating that trawling can indirectly have a significant affect on benthic biogeochemistry.

Muddy sediments, by their nature, suggest stable sediments and limited disturbance by physical events such as storms and wave action. Consequently such sediments typically display a "rich" macrofaunal community that is not adapted to frequent disturbance. This "rich" macrofaunal community however is not maintained by stability but by the creation of spatial temporal mosaics. These mosaics are generated and maintained by the patchy distribution of larger bioturbating species, including the feeding behaviour of large mobile predators e.g. whales, rays and crabs. However, due to the size and mobility of these predators they were precluded in the microcosms. Trawling can destroy the organisms responsible for maintaining these mosaics (see Chapter 2) and can therefore affect environmental heterogeneity and infaunal diversity, thus having a profound affect on the exchange of nutrients across the sediment-water interface.

Chapter 7: General discussion and concluding remarks

As early as the beginning of the 20th century, fishers were exerting a profound effect on the North Sea, representing an annual removal of around 10% of the total standing stock (Daan et al. 1990). In the last decade or so the effects of fishing on a range of ecosystem properties have been demonstrated. For the most part these have centred on how the direct effects of fisheries on populations ramify through food web dynamics (Jennings et al. 2002) and on the direct impacts on habitat features (Watling and Norse 1998, Collie et al. 2000). The broad aim of this thesis was to determine the effect of commercial trawling on benthic biogeochemistry. In this chapter the total body of findings presented in the previous chapters are considered to provide the overall conclusions of the work. Following this, a number of recommendations for future research based on these findings are made.

Many of the studies on the effects of trawling on the benthos have suggested that trawl impacts cause a shift from few large bodied, slow reproducing species to a proliferation of small bodied species with a high reproductive output (Jennings et al. 2001). This study has unequivocally demonstrated that trawling can have a significant impact on benthic nutrient dynamics. One important aspect of this impact was a measurable change in the distributions of benthic fauna (chapter 2). This was marked and predicated across a whole range of metrics of the system, but ultimately were attributable to a shift from fewer large bodied species to more numerous small bodied species following trawling on a commercial scale.

A method for extracting *in situ,* undisturbed sediment cores for porewater analysis was developed (chapter 3), and this method helped elucidate clear

differences between the porewater profiles of NO_2^-, NO_3^-, NH_4^+, PO_4^{3-} and CDOM for trawled and untrawled sediments (chapter 4). In particular, trawling was shown to temporarily increase the depth of O_2 penetration, associated with mixing of surficial sediment layers. Consequently, trawled sediments were characterised by a relatively homogenous surface sediment layer (~ 4cm) where nutrient concentrations were elevated for NO_2^- and NO_3^-, relative to untrawled sediments, yet were lower for NH_4^+ and PO_4^{3-} as they were liberated into overlying waters following trawl disturbance. It has been suggested that sediment resuspension only alters the timing of porewater nutrient release and holds little significance for primary production (Blackburn 1997). However, the depth of penetration by trawl gear extends into hypoxic sediment layers, transporting O_2 rich surface waters and thus alters nitrification and denitrification processes. Consequently, remineralisation pathways are stimulated and the magnitude of nutrient regeneration is changed.

A direct consequence of the altered nutrient dynamic due to trawling was the modification of sediment-water nutrient exchange (chapter 5). High trawl frequencies did not exhaust the supply of regenerated nutrients to bottom waters, and even low frequency trawl disturbances maintained an altered post trawl flux rate between impacts. Following any frequency (above the low frequency) of trawl disturbance sediment efflux of NO_2^- and NH_4^+ was enhanced relative to an untrawled background flux. Nutrient fluxes across an untrawled sediment-water interface for NO_3^- and PO_4^{3-} were into the sediment. However, following trawl disturbance these fluxes were reversed, i.e. net sediment efflux. These altered fluxes, although relatively short-term, persisted for > 48 hours. If these trawl induced pulses of nutrient release immediately following an impact

are scaled to the average annual area trawled in ICES statistical rectangle 39E8, the percentage increase relative to the annual background flux from undisturbed sediments, results in additional sediment effluxes of 676% and 475% for PO_4^{3-} and NH_4^+ respectively. NH_4^+ is significant to biological production as it is the most biologically available form of nitrogen while PO_4^{3-} is also essential for photosynthesis (L'Helguen et al. 1996). However, the addition of any anthropogenic nutrient source has the potential to modify phytoplankton abundance and composition (Redfield et al. 1963). As a result, eutrophication and/or toxic blooms have been stimulated which affect ecosystem processes (Justic et al. 1995).

The larger untrawled fauna increased the sediment-water exchange rates of nutrients relative to trawled fauna (chapter 6). As chapter 2 demonstrated the faunal assemblage was altered in trawled sediments and therefore, unlike the immediate pulsed release of nutrients following trawling, faunal induced fluxes are long-term and would persist throughout the year. Manipulated densities of fauna revealed fluxes of NH_4^+ increase with increasing faunal density, while the PO_4^{3-} and NO_3- fluxes declined. NO_2^- fluxes were highest for an intermediate level (times 4) of faunal density. These differences are explained by potential conflicts between increased organism size and bioturbation (Wheatcroft et al. 1990) in untrawled (relatively low organism density systems) versus increased bioirrigation in high density trawled systems with relatively smaller organisms (Sanders et al. 2000). These processes have been shown to have a regulatory effect on biogenically mediated remineralisation processes (Hines and Jones 1985).

It can be concluded that bottom trawling can have measurable effects, both direct and indirect on the benthic biogeochemistry of the North Sea. Direct effects include the immediate release of porewater nutrients to the overlying water and the removal of large infaunal organisms. Indirect effects include assemblage shifts in faunal communities and the alteration of diagenetic processes, including changes to the sediment redox status with resultant alterations to nitrification and denitrification processes and concomitant consequences for organic matter degradation rates.

Other studies have shown that benthic processes contribute over 50% of the nutrient requirements of primary producers (Klump and Martens 1987, Pilskaln et al. 1998). Stratification within the area of study is limited and only briefly occurs in some years during summer (chapter 1). Therefore, trawl induced nutrient fluxes in the North Sea will be in areas of high insolation and thus can be easily assimilated by primary producers and directly contribute to production. Following this assumption, it is reasonable to predict that a significant proportion of trawl induced nutrients will be mixed throughout the water column of the current study site. Consequently, the contribution of nutrients based on the findings of this investigation result in additional increases of 99% NH_4^+ and 230% PO_4^{3-} for each m^3 of water column overlying every m^2 of sediment that is trawled. These additional inputs are of ecological importance because the dynamics of the North Sea ecosystem are primarily bottom up controlled (Clark 2001). In other words primary production drives ecosystem components and therefore as trawling influences this, trawl activity inevitably affords significant ecological and biogeochemical value. If this assumption holds true then the scale of trawl fisheries in the North Sea would hold significance to

nutrient cycling. The magnitude of these anthropogenic inputs inevitably provides scope for regional eutrophication.

7.1 Future investigations

This thesis has provided valuable data on the relationships between nutrient dynamics and biology, how they react under trawl disturbance and has identified some of the key effects. Further studies need to be carried out in order to develop predictive approaches estimating the significance of trawl disturbance and in establishing the need for, and practicality of management measures. The following are a number of issues that have arisen from or remain unresolved during this study and require further work.

Measures of the susceptibility of the North Sea ecosystem to long-term additions of trawl induced nutrient sources: The pulsed release of nutrients following trawling can potentially lead to increased productivity (including the extension of spring/autumn bloom in temperate regions), nuisance blooms or even the release of toxins and pollutants. Therefore, the definitive end point of trawl stimulated nutrient additions needs to be established. Isotopic labelling of trawl derived nutrients may help to track and elucidate local and regional impacts of a pulsed release of nutrients.

The proportion of trawl induced nutrients utilised by plankton: With respect to plankton uptake of nutrients, the fraction of nutrients from regenerated sediment sources that are utilised by primary producers needs to be established. Labelled primary and secondary nutrient sources would enable

trawl-induced nutrients to be accurately quantified in planktonic blooms and would compliment the study outlined above.

Additional organic carbon incorporated in sediments following trawl activity: Knowledge of the amounts of organic matter transported to the sediment and any additional sediment inputs that would help drive degradation, and ultimately regeneration of inorganic nutrients would be valuable as this would determine other routes by which trawling alters benthic nutrient release and does not just alter the timing of sediment-water interface exchange. Studies have highlighted the potential of trawling to alter the amount of organic matter that is transported to and physically mixed with different redox horizons in the sediment (for example, Mayer et al. 1991, Pilskaln et al. 1998). However, these effects and how they affect remineralisation pathways have yet to be quantified.

7.2 Management implications

These future studies, as well as the data from this thesis are needed to compliment and enhance the current structure and knowledge of ecosystem dynamics. For example, the OSPAR commission's marine monitoring system seeks to assess the impacts of human induced activities that can directly or indirectly alter the natural attributes of marine ecosystems (OSPAR 1995). Their joint assessment and monitoring programme (JAMP) relies on high quality interpretable data on sources of nutrient enrichment which may lead to eutrophication in order to evaluate changes, causes, implications and identify the source of any impact/input that will require the attention of policy-makers and management groups. Another example where the value of the data

generated from this thesis could be of benefit is the continued development of the European Regional Seas Ecosystem Model (ERSEM). Within the ERSEM framework, data are needed to prescribe the initial ecosystem situation within the North Sea and for driving and validating the ecosystem model within annual cycles of the main state variables to enable a comparison of model outcome with observed system behaviour. Therefore these and other existing ecosystem and biogeochemical models would benefit from such data and help provide a framework from which management decisions could be made that incorporate a predictive and holistic approach.

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Appendix

Appendix 1. Date, position, ground type and depth of core sampling.(*) denotes a core taken directly in the trawl tracks of a commercial trawler, approximately 15-30 minutes after the ground was trawled.

Date	Position	Ground type	Water depth (M)
9/1/2002	55^0:13.63N; 001^0:27.37W	Trawled	41.5
9/1/2002	55^0:12.69N; 001^0:27.14W	Untrawled	42.1
19/2/2002	55^0:12.66N; 001^0:27.23W	Untrawled	40.3
19/2/2002	55^0:13.54N; 001^0:27.10W	Trawled	40.8
19/2/2002	55^0:13.58N; 001^0:26.72W	*Trawled	41.1
29/2/2002	55^0:13.64N; 001^0:27.19W	Trawled	40.0
29/2/2002	55^0:12.66N; 001^0:27.14W	Untrawled	40.6
13/3/2002	55^0:13.61N; 001^0:27.36W	Trawled	40.5
13/3/2002	55^0:12.65N; $001^0$27.11W	Untrawled	42.1
24/4/2001	55^0:13.43N; 001^0:27.11W	Trawled	44.0
24/4/2001	55^0:12.70N; 001^0:27.18W	Untrawled	44.3
30/5/2001	55^0:13.45N; 001^0:27.14W	Trawled	41.2
30/5/2001	55^0:12.71N; 001^0:27.14W	Untrawled	39.6
19/7/2001	55^0:13.61N; 001^0:27.43W	Trawled	38.6
19/7/2001	55^0:12.68N; 001^0:27.15W	Untrawled	40.0
17/8/2001	55^0:13.63N; 001^0:27.28W	Trawled	39.7
17/8/2001	55^0:12.68N; 001^0:27.25W	Untrawled	40.2
18/9/2001	55^0:13.43N; 001^0:27.13W	Trawled	38.6
18/9/2001	55^0:12.64N; 001^0:27.22W	Untrawled	38.9
16/10/2001	55^0:13.71N; 001^0:27.40W	Trawled	38.2
16/10/2001	55^0:12.70N; 001^0:27.19W	Untrawled	39.6
16/11/2001	55^0:13.55N; 001^0:27.28W	Trawled	39.3
16/11/2001	55^0:12.64N; 001^0:27.14W	Untrawled	42.2
14/12/2001	55^0:13.59N; 001^0:27.33W	Trawled	39.6
14/12/2001	55^0:12.66N; 001^0:27.21W	Untrawled	40.0

Appendix 2. Z-test statistic results for comparison of immediately trawled porewaters against trawled and untrawled sediment porewaters. Where if Z is greater than the critical value of 1.6449 the null hypothesis is rejected and the porewaters are significantly different. (*) denotes a significantly result.

Depth into sediment	z value for nutrient species:			
	Phosphate	Ammonium	Nitrite	Nitrate
Overlying water	2.400052*	63.4453*	14.04348*	30.90523*
1	10.72795*	155.8072*	0.113531	0.797583
2	32.11009*	85.69449*	-0.52568	10.46303*
3	31.96138*	139.1214*	0.365225	1.028806
4	31.55634*	69.80962*	-0.59879	1.025746
5	7.78137*	83.69902*	0.246705	-1.57628
6	41.76171*	113.6588*	0.175852	-1.05562
7	74.14236*	125.8043*	-1.60584	1.605742
8	47.27139*	148.4469*	-1.06693	-1.4861
9	110.0279*	178.4647*	-1.64263	-1.63888
10	65.6771*	171.1534*	-1.2477	1.502517
11	41.0732*	154.4681*	-0.99637	1.450333
12	49.56744*	191.476*	-1.1223	1.600375

Appendix 3. (a)Taxonomic family groups for those organisms identified within sediment samples that are known bioturbators, can extend siphons or have tube building capacities. (b) Total species list.

a

Ampeliscidae	Ampharetidae	Amphiuridae	Aphroditidae	Astartidae	Bodotriidae
Capitellidae	Chaetopteridae	Cirratulidae	Crangonidae	Cucumeriidae	Dentaliidae
Diastylidae	Emplectonematidae	Eunicidae	Flabelligeridae	Glyceridae	Golfingiidae
Halicampidae	Harrimaniidae	Hesionidae	Leptonidae	Leucinidae	Leucosiidae
Lucinidae	Luidiidae	Mactridae	Magelonidae	Maldanidae	Montacutidae
Myidae	Naticidae	Nephropidae	Nephtyidae	Nereidae	Nuculanidae
Nuculidae	Opheliidae	Ophiolepidae	Orbiniidae	Oweniidae	Parthenopidae
Pectinariidae	Phyllodocidae	Phyramidellidae	Portunidae	Priapulidae	Psammobiidae
Retusidae	Sabellidae	Scalibregmidae	Solecurtidae	Spatangidae	Spionidae
Synaptidae	Tanaidacea	Tellinidae	Terebellidae	Thraciidae	Veneridae

b

Acanthocardia echinata	*Achelia echinata*	*Ampharete grubei*	Amphiura spp	*Amphiura chiajei*
Amphiura filiformis	*Ampelisca brevicornis*	*Anaitides maculata*	*Angulus squalidus*	*Angulus tenuis*
anopla spp	*Apseudes latreillii*	*Astarte sulcata*	*Bathyporeia guilliamsoniana*	*Bodotria arenosa arenosa*
Brachystomia scalaris	*C.zaddachi*	*Callista chione*	*Carcinus maenas*	*Capitella capitella*
Chaetodermomorpha spp	*Chaetopterus variopedatus*	*Chaetozone setosa*	*Clausinella fasciata*	*Corystes cassivelaunus*
Crangon allmanni kinahan	*Dentalium entalis*	*Diastylis rathkei typica*	*Diastylis rugosa*	*Dodecaceria concharum*
Dosinia exoleta	*Dosinia lupinus*	*Ebalia tuberosa*	*Echinocardium cordatum*	Egg
Emplectonema echinoderma	*Eteone foliosa*	*Eteone longa*	*Eteone picta*	*Euclymere lumbricoides*
Eunice harassi	*Flatworm*	*Gari fervensis*	*Gattyana cirrosa*	*Glycera gigantea*
Glycera tridactyla	*Golfingia vulgaris vulgaris*	*Goniada maculata*	*Halcampa chrysanthellum*	*Harmothoe imbricata*
Harpinia antennaria	Harrimaniidae spp	Hoplonemertea spp	*Kefersteinia cirrata*	*Laetmatonice filicornis*
Lanice conchilega	*Laonice cirrata*	*Lepidonotus squamatus*	*Lepton squamosum*	*Leptosynapta inhaerens*
Leucothoe spinicarpa	*Liocarcinus pusillus*	*Lucinella divaricata*	*Luidia sarsi*	*Lumbrineris latreilli*
Lutaria lutraria	*Mactra corallina*	*Mactra stultorum*	*Magelona mirabilis*	*Malacoceros fuliginosus*
Maldanidae spp	Maldane sarsi	Mya truncata	Myrtea spinifera	Nebalia bipes
Nemertea spp	*Neoamphitrite figulus*	*Nephrops norvegicus*	Nephtys spp	*Nereimyra punctata*
Nereis spp	*Nereis longissima*	*Nicomache lumbricalis*	*Nuculana minuta*	*Nucula nitidosa*
Obtusella intersecta	Oligochaeta spp	*Ophelia bicornis*	*Ophelina acuminata*	*Ophiodromus flexuosus*
Ophiura affinis	*Ophiura ophiura*	*Orbinia latrelli*	*Orchomere nanus*	*Owenia fusiformis*
Pawsonia saxicola	*Peachia cylindrica*	*Pectinaria auricoma*	*Pectinaria belgica*	*Pharus legumen*
Phascolion strombus strombus	*Phaxas pellucidus*	*Pherusa plumosa*	*Phoronis muelleri*	*Phtisica marina*
Phyllodoce laminosa	Phyllodocidae spp	Poecilochaetidae spp	*Polinices fuscus*	*Polinices polianus*
Priapulus caudatus	*Pseudocuma longicornis*	*Retusa obtusa*	*Retusa truncatula*	*Rhodine loveni*
Sabella pavonina	*Scalibregma inflatum*	*Scoloplos armiger*	*Skeneopsis planorbis*	*Spiophanes bombyx*
Spio filicornis	Spionidae spp	*Stenothoe monoculoides*	*Sthenelais boa*	*T.helgolandica*
Tellimya ferruginosa	*Terebellides stroemi*	*Tetrastemma ambiguum*	*Thracia pubescens*	*Travisia forbesii*
Turbellaria spp	*Venus casina*			

Appendix 4. One-way ANOVA with subsequent TUKEY pairwise test (correction factor applied). Where MD = molecular diffusion of nutrients (sediments without macrofauna); NU = Natural density of untrawled macrofauna; NT = Natural density of trawled macrofauna; 4U = times 4 the natural density of untrawled macrofauna; 4T = times 4 the natural density of trawled macrofauna; 8U = times 8 the natural density of untrawled macrofauna and 8T = times 8 the natural density of trawled macrofauna.

a

Ammonium				Level of significance	
time	factor	versus	factor	f value	p value
9	8T	vs.	MD, NU, NT, 4U, 4T, 8U	9.24	< 0.001
13	8T	vs.	MD, NU, NT	7.70	< 0.001
17	4U	vs.	MD, NT	27.59	< 0.001
17	4T	vs.	MD, NT	27.59	< 0.001
17	8U	vs.	MD	27.59	< 0.001
17	8T	vs.	MD, NU, NT, 4U, 4T, 8U	27.59	< 0.001
21	4U	vs.	MD, NT	43.14	<0.001
21	4T	vs.	MD, NU, NT	43.14	<0.001
21	8U	vs.	MD, NT	43.14	<0.001
21	8T	vs.	MD, NU, NT, 4U, 4T, 8U	43.14	<0.001
25	4U	vs.	MD, NT	34.09	<0.001
25	4T	vs.	MD, NU, NT	34.09	<0.001
25	8U	vs.	MD, NT	34.09	<0.001
25	8T	vs.	MD, NU, NT, 4U, 4T, 8U	34.09	<0.001
29	4U	vs.	MD, NT	32.03	< 0.001
29	4T	vs.	MD, NT	32.03	< 0.001
29	8U	vs.	MD, NT	32.03	< 0.001
29	8T	vs.	MD, NU, NT, 4U, 4T, 8U	32.03	< 0.001
33	4U	vs.	MD, NT	45.47	< 0.001
33	4T	vs.	MD, NU, NT	45.47	< 0.001
33	8U	vs.	MD, NU, NT	45.47	< 0.001
33	8T	vs.	MD, NU, NT, 4U, 4T, 8U	45.47	< 0.001
37	4U	vs.	MD	35.98	< 0.001
37	4T	vs.	MD, NU, NT	35.98	< 0.001
37	8U	vs.	MD, NU, NT, 4U	35.98	< 0.001
37	8T	vs.	MD, NU, NT, 4U, 4T, 8U	35.98	< 0.001
41	4U	vs.	MD	31.57	< 0.001
41	4T	vs.	MD, NU, NT	31.57	< 0.001
41	8U	vs.	MD, NU, NT, 4U	31.57	< 0.001
41	8T	vs.	MD, NU, NT, 4U, 4T	31.57	< 0.001
45	4U	vs.	MD, NU, NT	37.06	< 0.001
45	4T	vs.	MD, NU, NT	37.06	< 0.001
45	8U	vs.	MD, NU, NT, 4U, 4T	37.06	< 0.001
45	8T	vs.	MD, NU, NT, 4U, 4T	37.06	< 0.001

b

Nitrate				Level of significance	
time	factor	versus	factor	f value	p value
1	4U	vs.	MD, NU, NT, 8T	4.38	< 0.05
5	4U	vs.	NU, 8U, 8T	5.81	< 0.05
5	8T	vs.	4T	5.81	< 0.05
9	8T	vs.	MD, NT, 4U	5.26	< 0.05
13	8U	vs.	MD	5.40	< 0.05
13	8T	vs.	MD, NU, NT, 4U	5.40	< 0.05
17	4T	vs.	MD	10.48	< 0.001
17	8U	vs.	MD	10.48	< 0.001
17	8T	vs.	MD, NU, NT, 4U	10.48	< 0.001
21	8U	vs.	MD, NT	7.92	< 0.001
21	8T	vs.	MD, NU, NT, 4U	7.92	< 0.001
25	4T	vs.	MD	11.71	< 0.001
25	8U	vs.	MD, NU, NT	11.71	< 0.001
25	8T	vs.	MD, NU, NT, 4U	11.71	< 0.001
29	4T	vs.	MD	11.57	< 0.001
29	8U	vs.	MD, NU, NT	11.57	< 0.001
29	8T	vs.	MD, NU, NT, 4U	11.57	< 0.001
33	4T	vs.	MD	9.59	< 0.001
33	8U	vs.	MD, NT	9.59	< 0.001
33	8T	vs.	MD, NU, NT	9.59	< 0.001
37	4U	vs.	MD	11.12	< 0.001
37	4T	vs.	MD, NT	11.12	< 0.001
37	8U	vs.	MD, NT	11.12	< 0.001
37	8T	vs.	MD, NU, NT	11.12	< 0.001
41	4U	vs.	MD	15.08	< 0.001
41	4T	vs.	MD, NT	15.08	< 0.001
41	8U	vs.	MD, NU, NT	15.08	< 0.001
41	8T	vs.	MD, NU, NT, 4U	15.08	< 0.001
45	4T	vs.	MD	10.74	< 0.001
45	8U	vs.	MD, NU, NT	10.74	< 0.001
45	8T	vs.	MD, NU, NT	10.74	< 0.001

c

Phosphate				Level of significance	
time	factor	versus	factor	f value	p value
9	MD	vs.	NU, NT	7.72	< 0.001
13	MD	vs.	NU, NT, 4U, 4T	10.63	< 0.001
17	MD	vs.	NU, NT, 4T	9.83	< 0.001
21	MD	vs.	NU, NT, 4U, 4T, 8T	19.36	< 0.001
25	MD	vs.	NU, NT, 4U, 4T, 8T	9.40	< 0.001
29	MD	vs.	NU, NT, 4U, 4T, 8T	14.09	< 0.001
33	MD	vs.	NU, NT, 4U, 4T, 8T	24.29	< 0.001
33	MD	vs.	MD, NU, NT	24.29	< 0.001
37	MD	vs.	NU, NT, 4U, 4T, 8T	19.74	< 0.001
37	8U	vs.	NU	19.74	< 0.001
41	MD	vs.	NU, NT, 4U, 4T, 8U, 8T	6.23	< 0.001
45	MD	vs.	NU, NT, 4T	6.23	< 0.001