# The Impact of a Sugar By-products Effluent on the Beach Meiofauna at Sezela Beach, KwaZulu-Natal, South Africa

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## ABSTRACT

Beach meiofauna were chosen as environmental indicators to investigate the impact of Illovo sugar by-products effluent. The effluent is pumped through a 20 cm diameter pipeline into surf zone at Sezela beach on the coast of KwaZulu-Natal, South Africa. Meiofaunal communities were considered appropriate indicators as they are relatively stable both qualitatively and quantitatively on a seasonal and year to year basis. Most meiofauna also do not have planktonic stages in their life cycles, respond rapidly to pollution due to their fast generation times, and they are often abundant with high species diversity in habitats which are subject to considerable natural physical and chemical fluctuations. In this particular study there was a specific concern about trace amounts of furfural in the effluent. Furfural has been used as the active ingredient in a product designed to kill parasitic nematodes in crop fields. A large proportion of the beach meiofauna consists of nematodes.

Eight stations were sampled for meiofauna along the beaches at Sezela on 7 different occasions. Seasonal effects on meiofauna and meiofaunal recovery during the period when the factory was not pumping effluent to sea was assessed. Samples were taken on the following dates: 4 July 2000 (winter); 30 August 2000 (winter); 13 December 2000 (spring); 26 January 2001 (summer); 8 March 2001 (summer); 9 April 2001 (autumn); and 2 January 2002 (summer). PRIMER (Plymouth Routines in Multivariate Ecological Research) was used for statistical analysis and included various univariate indices such as species richness, species diversity and evenness. These indices were then analysed using one-way ANOVA to determine any significant difference between sites over the 7 sampling periods and between the different seasons. Clustering and Ordination multivariate analyses were carried out on the community data and physico/chemical data to determine community patterns and relate them to the effluent and environmental data. The Nematode/Copepod ratio was also calculated. Meiofauna were analysed at major taxa level, as well as to nematode feeding groups and harpacticoid copepod and annelid family level, to determine if analysis to major taxa level is adequate as an indicator of pollution impact.

The analyses indicated a possible degree of impact at stations close to the effluent discharge when effluent was being pumped to sea and a recovery was noted at the station closest to the discharge when effluent was not being discharged and analysis was conducted to the major taxonomic rank only. No improved resolution was achieved by analysing some of the meiofaunal major taxa to family level or different feeding groups. The analysis of the Nematode/Copepod ratio was shown to correspond with the multivariate analyses, however, this ratio could not reveal the severity of the impact where both nematodes and harpacticoids i.e. total meiofauna had been reduced by adverse conditions. The physical and chemical

variables that showed the greatest correlation with the meiofaunal community patterns were sediment grain size, dissolved oxygen and salinity. There was a very strong positive correlation between Kjeldahl nitrogen in the interstitial waters and total numbers of meiofauna. This and the relationship with salinity may have suggested other possible sources of influence such as enrichment from the three estuaries in the area as well as a storm water drain located 150m north of the effluent discharge. A seasonal effect was observed with increased meiofauna numbers in autumn, but this was possibly influenced by the periods when effluent was not being pumped to sea.

## PREFACE

The research work described in this dissertation was carried out in the School of Biological and Conservation Sciences, University of Kwa Zulu-Natal, Durban, from July 2000 to December 2006, under the supervision of Professor John A. Cooke.

These studies represent original work by the author and have not otherwise been submitted in any form for any degree or diploma to any tertiary institution. Where use has been made of the work of others it is duly acknowledged in the text.

Signed:_____ Name:_____ Date:_____

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# 1. INTRODUCTION

## 1.1 Meiofauna

The study of meiofauna started many years before the term meiofauna (or meiobenthos) was proposed and the earliest meiofauna studies focused on the discovery and description of new taxa (Higgins & Thiel, 1988). It was in the 1920s that scientists first started to seriously study the animals that live in the interstitial water found in marine, estuarine and freshwater sediments (Swedmark, 1964). The term 'meiobenthos' was introduced and defined in 1942 by Mare in her account of the benthos of muddy substrates off Plymouth, England (Coull & Giere, 1988; Dye & Furstenburg, 1981). The term 'meiofauna' is derived from the Greek *meio* meaning 'smaller' and refers to the fauna that are smaller than the lower size limit for macrofauna (which has been defined as animals retained on a 1 mm sieve). This group was therefore, originally defined by size and included animals retained on a 0.045 mm sieve but smaller than 1.0 mm in body length (Dye & Furstenburg, 1981; Higgins & Thiel, 1988). The term is now more restricted to benthos (animals on or near the sea floor) and phytal fauna (animals living on plants), but not for planktonic organisms, which are treated as a separate category (Hulings & Gray, 1971). The meiofauna are not a homogeneous ecological group and occupy a wide diversity of habitats from freshwater to marine environments, from high on the beach to the bottom of the deepest oceans and they are found in the finest muds to the coarsest shell gravels. Other meiofaunal habitats include rooted vegetation, moss, macroalgal fronds and various animal structures such as coral crevices, worm tubes and echinoderm spines.

The intertidal beach meiofauna are the main focus of this study, where animal size is determined by the fact that they live in the interstitial spaces between the particles of the substrate. Most casual observers are unaware of the existence of the meiofauna although they form common animal communities which inhabit beaches throughout the world. The meiofauna of beaches includes species belonging to the Nematoda, Turbellaria, Annelida, Kinorhyncha, Rotifera, Gastrotricha, Acarina (mites), Tardigrada and Crustacea (Nicholas & Hodda, 1999). McIntyre (1971) uses the term 'permanent members' for species belonging to the meiofauna during the whole of their life cycle. The meiofauna also includes a large number of temporary members, including larvae and juveniles stages of species that, as adults, belong to the macrofauna.

Originally the terms macrofauna and meiofauna were considered as just arbitrary size divisions of the metazoan benthos determined by different collecting methods. On sandy beaches however meiofauna differ from macrofauna not only in size but also in the ecological niche it occupies. Meiofaunal communities are usually much more diverse than macrofauna and they are controlled by different environmental factors. Highly exposed beaches with coarse sediment tend to have abundant meiofauna and sparse resident macrofauna populations (Hooge, 1999; McLachlan, 1977) as meiofauna are less sensitive to greater exposure to strong wave action and coarser substrata than the macrofauna (McLachlan *et al.*, 1981; Rodriguez *et al.*, 2003). The macrofauna and the meiofauna of sandy beaches can comprise two separate faunal communities which have little overlap or exchange of energy ( McLachlan, 1977 and McLachlan & Erasmus, 1983).

## 1.2 Ecology of sandy beach meiofauna

The major process controlling the habitat of beach meiofauna, the interstitial climate, is the filtration of sea water through the sand. This is determined both by the wave action and by sediment properties (Brown & McLachlan, 1990). Progressing from steep reflective to flat dissipative beaches, the filtered volumes can range from 100 to less than 1 $m^3$ $m^{-1}$ $day^{-1}$, while

the residence times of this water in the interstitial sand can vary over the range 1 hour to more than 100 days (McLachlan *et al.*, 1985). In summary, coarse-grained, steep reflective beaches have large volumes of sea water flushing rapidly through them while fine grained, flat dissipative beaches receive less water, which percolates through the substrate more slowly.

The water input concentrates organic material in the sand and as a chemical extreme, organic input can exceed oxygen supply and the sediment becomes deoxygenated, developing strong chemical gradients where three vertical layers can be distinguished. There is an oxygenated layer at the surface where elements such as nitrogen and sulphur occur in their oxidised states ($NO_3$, $SO_4$). Below this is a transition zone, where the changeover occurs from oxidising to reducing conditions and oxygen levels are reduced causing reduced states of N and S ($NH_4$, $H_2S$). The third layer is characterised by toxic reduced compounds. These three layers can appear as yellow, grey and black sand respectively, the black colour being due to iron sulphides. Interstitial fauna are concentrated in the top layer and anaerobic microbial processes predominate at depth. Towards the physical extreme, water and oxygen inputs exceed organic input and the interstices remain open as high energy capillaries (McLachlan *et al.*, 1979a). McLachlan (1980) proposed a simple rating system for defining sandy beaches in relation to exposure. The parameters on which this index is based are: wave action, sand particle size and beach slope, sediment oxidation depth of reducing layers and macrofauna burrows. Table 1.1 shows how scores are obtained from the various parameters and then a rating of exposure for a particular beach from 0 to 20 can be obtained with 0 being the least exposed and 20 the most extreme of exposure.

Table 1.1
Rating scheme for assessing the degree of exposure of sandy beaches (from McLachlan, 1980)

| Parameter | Rating | score |
|---|---|---|
| Wave action: | Practically absent | 0 |
| | Variable, light to moderate, wave height seldom exceeds 0.5m | 1 |
| | Continuous, moderate, wave height seldom exceeds 1m | 2 |
| | Continuous, heavy, wave height mostly exceeds 1m | 3 |
| | Continuous, extreme, wave height never less than 1.5m | 4 |
| Surf zone width: | Very wide, waves first break on bars | 0 |
| | Moderate, waves usually break 50-150m from shore | 1 |
| | Narrow, large waves break on beach | 2 |
| % very fine sand: (63-125$\mu$m) | >5% | 0 |
| | 1-5% | 1 |
| | <1% | 2 |

| Median particle Diameter ($\mu$m) | | Slope of intertidal zone >1/10 | 1/10-1/15 | 1/15-1/25 | 1/25-1/50 | <1/50 |
|---|---|---|---|---|---|---|
| >710 | (>0.5Ø[1]) | 5 | 6 | 7 | 7 | 7 |
| 500-710 | (1.0-0.5Ø) | 4 | 5 | 6 | 7 | 7 |
| 350-450 | (1.5-1.0Ø) | 3 | 4 | 5 | 6 | 7 |
| 250-350 | (2.0-1.5Ø) | 2 | 3 | 4 | 5 | 6 |
| 180-250 | (2.5-2.0Ø) | 1 | 2 | 3 | 4 | 5 |
| 180 | (>2.5Ø) | 0 | 0 | 1 | 2 | 3 |

| | | |
|---|---|---|
| Depth of reducing layers (cm): | 0 – 10 | 0 |
| | 10 – 25 | 1 |
| | 25 – 50 | 2 |
| | 50 – 80 | 3 |
| | >80 | 4 |
| Stable macrofaunal burrows: | Present | 0 |
| | Absent | 1 |
| | Highest exposure | 20 |
| | Lowest exposure | 0 |

On the basis of total score beaches may be rated in exposure categories as shown in Table 1.2:

[1] Particle size is often classified according to the Wentworth scale, in phi units, where $\text{Ø} = -\log_2$ diameter (mm).

Table 1.2
Exposure categories of beaches rated according to total score from Table 1.1. (from McLachlan, 1980)

| Score | Beach type | Description |
|---|---|---|
| 1 - 5 | Very sheltered | Virtually no wave action; shallow reduced layers; abundant macrofaunal burrows |
| 6 – 10 | Sheltered | Little wave action; reduced layers present; usually some macrofaunal burrows |
| 11 – 15 | Exposed | Moderate to heavy wave action; reduced layers deep if present; usually no macrofaunal burrows |
| 16 – 20 | Very exposed | Heavy wave action; no reduced layers; macrofauna only of tough motile forms |

This rating scale should be workable under most conditions likely to occur on sandy beaches. It cannot, however, replace the experienced eye and is intended rather to assist in obtaining uniformity in exposure ratings between different parts of the world (McLachlan, 1980).

Exposed sandy shores are usually characterised by benthic microalgae and phytoplankton as the main primary producers, while attached macroalgae are missing. In addition to microalgae, the food web is based on dissolved and particulate organic matter as detritus and carrion, with the latter being of relatively minor importance (Brown & McLachlan, 1990). A study carried out on a beach on the west coast of South Africa by Koop & Griffiths (1982) revealed that bacteria accounted for around 87% of the annual production, with meiofauna and macrofauna making up 10 and 3% respectively. Despite the rather low contribution to the productivity of the beach as a whole the macro- and meiofauna are of key functional importance in the initial process of fixing particulate organic material and making it available for mineralization by bacteria in the interstitial environment (Gheskiere *et al.*, 2006). With the normal rate of organic loading in nature the beach animal communities remain in equilibrium with the supply of material from the ocean. If the organic loading into the surf is increased then a new point of equilibrium may be reached with a higher biological activity. This additional organic loading could come either from rivers and seepage of ground water after rains or from an organic pollution source such as an effluent pipeline. The ecosystem can absorb a considerable load of organic material but the supply of oxygen is usually the limiting condition (Rodriguez, 2003). Beaches can become overloaded with organic material, with subsequent breakdown of aerobic processes, and the onset of anaerobic conditions. Many meiofaunal species graze directly on bacterial populations, maintaining the bacteria in a state of active growth and thus contribute significantly to the organic decomposition process. If pollutants prove not only to affect the species composition of the meiofauna, but also to

reduce the metabolic rate of the meiofauna, one result could be a long-term accumulation of organic and toxic materials (Tietjen, 1982).

Kwa Zulu-Natal (KZN) south-coast beaches experience heavy wave action characterized by plunging waves, which break on the beach. At the low water mark these beaches drop steeply into gullies, which are bordered by offshore bars. The wave heights may exceed 2 m and the beaches can be described as dangerous for swimming (Dye *et al.*, 1981). Meiofauna are well represented on all KZN beaches and very high biomass values may be obtained from some of the seemingly most inhospitable beaches. Ocean beaches subjected to large waves, remain much less frequently studied than bay and estuarine beaches with relatively small waves. This is most likely due to the substantial logistical difficulties associated with studying high-energy beaches (McLachlan & Erasmus, 1983; Nicholas & Hodda, 1999).

Meiofauna are considered temporary if they are larval stages of macrofaunal forms. In finer sediments and more sheltered areas, temporary meiofauna may be particularly abundant during certain seasons but in more dynamic situations, where all the meiofauna are truly interstitial, temporary forms are usually rare. The dominant taxa of sandy beach meiofauna are nematodes and harpacticoid copepods, where nematodes usually represent the greatest number of species, whereas harpacticoid copepods seldom have more than 5 to 10 species on any one beach. Other important groups include turbellarians, oligochaetes, mystacocarids, gastrotrichs, ostracods, acarina and tardigrades (Brown & McLachlan, 1990).

Meiofauna, particularly harpacticoid copepods, are significant bioindicators of ecological disturbance. Despite numerous uncertainties which remain in the autecology and feeding habits of many species, harpacticoid copepods can be divided into several ecological groups. Sandy, muddy, phytophilous (living on plant surfaces) and eurytopous (wide range of habitats) species can be distinguished (Bodin, 1988). Among sandy species, there are epipsammic types which generally remain at the surface of the sand, endopsammic types, whose appendages have strong spines which allow them to dig through the sand by shifting the grains, and mesopsammic (interstitial) types, which are vermiform (long and slender), have simplified appendages and carry reduced numbers of eggs, allowing them to move between sand grains (usually in coarser sands) without any need to shift the grains (Bodin, 1988). Interstitial meiofauna are near the lower limits of body size for metazoans and have reduced cell numbers and display simple organisation and sizes may be reduced to as little as 0.2 mm total length. As a result of low cell numbers, gamete production is low and consequently few eggs are carried by female harpacticoids in berry (carrying eggs below the abdomen) which aids in their interstitial life style (Brown & McLachlan, 1990). Some species are characteristic of estuarine muds, and tolerate both lower salinity (euryhaline) and higher organic matter content which commonly occurs in estuaries. Phytophilous species usually live among algae or are linked to the presence of stranded algae and plant detritus. So called eurytopous species can develop in most habitat types. They are generally euryhaline (tolerate a range of salinities), eurythermal (tolerate a range of temperatures) and are tolerant to the higher silt/clay fraction and to higher organic matter content (Bodin, 1988). The interstitial mesopsammic forms are found on coarser sands e.g. on beaches.

Most studies of meiofaunal feeding have been conducted in the laboratory. These studies were reviewed by Coull (1988). Tubellaria prey on other meiofauna and oligochaetes feed on detritus and bacteria. Nematodes suspected of feeding on bacteria because of their narrow tubular bucal morphology ingested more bacteria than diatoms when given a choice, and those nematodes suspected of feeding on diatoms because of having a buccal cavity armed with small or moderately sized teeth, ingested more diatoms than bacteria (Coull, 1988). Nematodes and copepods are known to extrude mucus to trap bacteria and the bacterial/mucus mixture is ingested. Meiofauna-macrofauna interactions are apparently absent from coarse-grained exposed beaches but occur more frequently on more fine grained sheltered shores (Brown & McLachlan, 1990). The most extensive work on trophic level and

biological interactions has been done by Reise (1985) on the sand flats of the Wadden Sea. These were fine grained sands and Reise found marked effects of macrofaunal predation on meiofauna and of macrofaunal burrows with the irrigation of the sediment and its subsequent effects on the meiofauna.

Reise (1985) showed that numerous small fish and invertebrates become active predators at high tide. Three predators he investigated were a gobiid fish (*Pomatoschistus*), juvenile shore crabs (*Carcinus*), and brown shrimp (*Crangon*). These predators sort the surface sand and remove juvenile macrofauna and meiofauna mainly from the top 1 cm of the sediment. Juvenile crabs significantly reduced the numbers of nematodes, turbellaria and harpacticoid copepods in surface sediments. Experimentally excluding all predators from the sediment using mesh cages, he found nematode numbers doubled after two months. The caging experiments had less of an affect on nematodes however than on juvenile macrofauna where total abundance of juvenile polychaetes and cockles were four times higher in the cages as opposed to the control sites. He considered the small size and deeper vertical distribution of meiofauna to be protection mechanisms shielding them from predation by macrofauna.

Reise (1985) also demonstrated several effects of macrofaunal burrows and feeding actions on the sediments, which promote meiofauna community development. Increased numbers of meiofauna around macrofaunal burrows resulted from irrigation and oxygenation, but further to this, excretory or secretory products from the macrofauna may enhance bacterial growth, which in turn promoted meiofauna. This activity might also result in the release of nutrients and promote better diatom growth on the sediment, thereby also providing more available food for the meiofauna. Meiofauna are known as important consumers of primary production (De Troch *et al.*, 2006).

Reise (1985) demonstrated significant effects of burrows of the lugworm, *Arenicola*, on the meiofauna, the presence of which generally increased meiofaunal densities. However deposit feeders such as *Arenicola, Callianassa* and others, ingest sediment containing meiofauna and thus feed directly on the latter. In summary there is still a large gap in knowledge on the meiofaunal food web and meiofauna-macrofauna interactions on coarse high energy sandy shores and further studies are needed on this aspect of meiofaunal ecology (Rodriguez *et al.*, 2003; Menn, 2002 and Moreno *et al.*, 2006).

## 1.3 Meiofauna as pollution indicators

Biological indicators are biological variables which can be used to make judgements of the effects of a pollutant on the environment. Bioindicator organisms can be used for the identification and qualitative and quantitative determination of pollutants and have been classified as being sensitive or accumulative (Conti & Cecchetti, 2001). Sensitive biomonitors may be of the observational type based on morphological changes, changes in abundance or behaviour of organisms related to environmental variables; or based on chemical or physiological changes such as alterations in the activity of different enzyme systems or processes such as photosynthetic or respiratory activities. The accumulative type, have the ability to store contaminants in their tissues and are used for the integrative measurement of such contaminants in the environment through bioaccumulation from the surrounding environment (Conti & Cecchetti, 2001). Cortet *et al.* (1999) describe a bioindicator as ideally needing to fulfil certain requirements such as: playing an important role in the functioning of the ecosysytem; being widely distributed, common and easy to sample; being relatively robust so as not to be killed at very low levels of pollutants; have measurable responses such as pollutant concentration in tissues or disturbances in growth and fertility; and should have reproducible responses such that they produce similar responses to the same levels of pollutant exposure at different sites.

Hierarchical levels of biological indicators range from the biochemical (subcellular) to the ecosystem level. Examples of biochemical and physiological indicators include changes in enzyme activity (biochemical) and in respiratory metabolism (physiological) as a result of exposure to a toxicant (Rosenberg & Resh, 1996). Biomarkers are indicators that are measurable molecular and biochemical changes which occur after exposure to toxic substances (Cortet *et al.,* 1999). An example is a class of metalloproteins called metallothioneins which are synthesised by an organism in response to heavy metal accumulation in the cells. These metalloproteins bind with excess free metal cations present in the cytosol and detoxify excess metal penetration into the cell and protect cell structures from non-specific interactions with heavy metal cations (Viarengo *et al.*, 1999). Due to their inducibility to heavy metals, metallothioneins are usually considered an important specific biomarker to detect organism response to inorganic pollutants such as Cd, Hg, Cu, Zn, etc. present in the environment (Viarengo *et al.*, 1999). Bioindicator organisms that have been employed in the application of metallothioneins as biomarkers are fish (Roy & Bhattacharya, 2006), molluscs (Marie *et al.*, 2006) and plants (Cozza *et al.*, 2006). The measurement of intra-cellular DNA-damage induction by a contaminant is another example of a sub-cellular bioindicator. Martin *et al*. (2005) conducted genotoxicity assays on earthworm tissues exposed to benzo(a)pyrene and lindane, and found that these assays could facilitate hazard identification within terrestrial ecosystems.

Indicators at the level of the individual organism may involve morphological deformities, altered behaviour, life history such as survival and growth or may involve measurements of bioaccumulation of toxic substances in the tissues of a particular species (Rosenberg & Resh, 1996). For example, abnormalities in Chironomid mouthparts have often been used to monitor the quality of sediments in freshwater environments (Martinez *et al.,* 2003; Meregalli *et al.*, 2001). It is believed that deformities develop during larval molting due to hormonal disturbance in the development of the bucal structures during the molting process and that many chemicals can mimic the hormones that regulate the molting process (Meregalli *et al.*, 2001). One of the most important hormones regulating the molting process is ecdysone and Meregalli *et al.* (2001) used a laboratory bioassay to demonstrate that the endocrine disruptor 4-*n*-nonylphenol increases the frequency of mouthpart deformities in chironomids. The quantification of the scale of characteristic mouthpart deformities in natural chironomid populations could provide a relatively inexpensive bioassay for certain pollutants, such as heavy metals.

However, for such bioassays to be a practical reality, there is first a need for adequate demonstration, usually experimental, of cause/effect relationships between specific pollutants and the deformities or biological effects (Martinez *et al.,* 2003). Three examples of such studies are given below. Cadmium and copper treated sediments induced deformities in *Chironomus tentans* at significantly higher proportions than control sediments (Martinez *et al.,* 2003). Imposex related studies have used neogastropods as indicator organisms. Imposex is a genital disorder, wherein male sex organs, notably a penis and a vas deferens, are superimposed onto the female of gonochoristic gastropods and is induced in gastropods primarily by tributyltin (TBT) compounds which are widely used in antifouling paints for ships (Vishwakiran *et al.,* 2006). Changes in liver and kidney tissue of the fish *Channa punctatus* have been used as indicators of arsenic toxicity (Roy & Bhattacharya, 2006).

Deviations in normal behaviour in response to a specific pollutant has also been used as an indicator of pollution. Many amphibians are good indicators of pesticide contamination in the environment. In *Rana temporaria* a low level of the insecticide cypermethrin (1µg $l^{-1}$) in an aquatic environment evoked a pronounced inhibition of body growth of the tadpoles, and aberrant behaviour such as tail kinking and the consequent twirling behaviour caused greater hazards of predation as it diminished the capability of the tadpoles to escape predators (Greulich & Pflugmacher, 2003). It is an important point in ecology that acute lethality tests (LC50) used to determine the concentration of a toxicant that is lethal to 50 % of individuals

of a particular experimental population after a specific exposure duration are useful for generating guidelines to protect against physiological death but they ignore 'ecological death' that may occur after much lower toxicant exposures; even if animals are not overly harmed by a contaminant they may be unable to function in an ecological context if their normal behaviour is altered (Scot & Sloman, 2004). Since behaviour serves as a link between physiological and ecological processes, it may be ideal for studying environmental pollutant effects as environmental contamination measured in natural ecosystems often occurs at concentrations well below those causing significant direct mortality (Scot & Sloman, 2004).

Life history endpoints such as survival and growth can also be used as an environmetal indicator (Rosenberg & Resh, 1996). Greulich & Pfugmacher (2003) studied the influence of the pyrethroid insecticide cypermethrin on the hatching success, mortality and deformities in development, duration of metamorphosis, and growth of the amphibian *Rana arvalis* tadpoles exposed at various life stages. Eggs were harmed significantly by exposure to different concentrations of cypermethrin depending on exposure time. Individuals exposed to cypermethrin in early life stages such as eggs and newly hatched tadpoles metamorphosed earlier than in the corresponding control. However, exposure of the tadpoles throughout their whole development prolonged the metamorphosis. Bejaro *et al.* (2004) used a meiobenthic copepod bioassay with *Amphiascus tenuiremis* to test the toxicity of sediments with urban-related contaminants such as polycyclic aromatic hydrocarbons (PAHs), metals and pesticide mixtures. Significant effects were found on reproductive outputs and that reproductive end points rather than adult survivorship were more sensitive to effects of contaminated sediments. Therefore endpoints relative to controls suggested a high risk to long term *A tenuiremis* population maintenance.

A sentinel species according to Martin *et al.* (2005) should be ubiquitous, sedentary, abundant and sufficiently long lived with the capacity to be reasonably tolerant to toxicants that bioaccumulate. The use of sentinel organisms as bioaccumulators of metals and organic contaminants such as insecticides and polychlorinated biphenyls (PCBs) provides a number of advantages over the direct, chemical analysis of contaminants in water or sediments. For example, this approach can provide a time-integrated indication that the contaminant is bioavailable, and can warn that other parts of the food web and the ecosystem may be affected (Rosenberg & Resh, 1996). Sessile benthic molluscs are used all around the world as quantitative biological indicators for monitoring chemical contaminants in marine environments (Nakhle *et al.*, 2006).

At the population level indicator taxa have been used to classify the degree of pollution in an aquatic ecosystem by determining the tolerance or sensitivity of a taxon to a given pollutant (Rosenberg & Resh, 1996). An example of a tolerant taxon is the opportunistic polychaete *Capitella capitata*, which proliferates after increases in organic matter (Giangrande *et al*., 2005). Species from the family Syllidae such as *Brania pusilla, Salvatoria clavata, Eusyllis lamelligra* and species of the genus *Exogone* occurring on hard bottom habitats have proved to be useful indicators that react in the opposite way to *Capitalla capitata* by being highly sensitive to pollution or other kinds of stress, and decreasing in numbers of species and individuals or completely disappearing from habitats (Giangrande *et al.*, 2005). Other examples from the major taxonomic groups within the meiofauna particularly concern different sensitivities to lack of oxygen and to chemical toxicity. The crustacean meiofauna in general seems to be the most quickly affected by hydrocabons which cause reductions in copepods, ostracods and nauplii, (Carmen *et al.*, 2000). Lee *et al.* (2001) found that copper pollution reduced harpacticoid numbers significantly. Harpacticoid copepods were found by Murrell & Fleeger (1989) to be sensitive to hypoxia and unable to survive these conditions for more than a few days while Bodin (1988) and Carmen *et al.* (2000) found them to be more sensitive to both the lack of oxygen and to hydrocarbon toxicity than nematodes. Copepods have been assumed, generally, to be more sensitive to the effects of most pollutants than nematodes (Raffaelli & Mason, 1981).

In contrast to the subcellular, individual and population level measurement, community level measurements have been the most widely performed for marine environmental monitoring and are considered by many to be the most important level of ecological organisation for such measurements (Warwick, 1993 and Martin & Richards, 1995). According to Attrill & Depledge (1997), investigations at the community level have a number of advantages over assessments targeted at lower levels of organisation. Firstly, such investigations are the most ecologically relevant as alterations in community structure can be extrapolated to the health of the ecosystem through changes such as in the food web and competition/predation. Second, investigations at lower levels (eg. as in most toxicity tests) tend to focus on a single species response whereas the community provides a multi-species response often covering a wide taxonomic range with a range of sensitivities to any given contaminant. Toxicity tests tend to concentrate on species that survive well under laboratory conditions (eg. *Mytilus edulis, Daphnia magna, Carcinus maenas*) and that have a relatively high tolerance to contamination and are not necessarily the most relevant to the natural situation, or the species that will cause community level effects (Attrill & Depledge, 1997).

Studies involving benthic communities have progressively replaced the biology of single indicator species, with the effect of stress being measured by utilizing multispecies assemblages, and examining changes in abundance of sets of species (Giangrande *et al.,* 2005). Together with this approach, in discriminating between sites or sampling times, multivariate methods have been shown to be very useful (Warwick & Clarke, 1991). However, community level investigations have several limitations. They are often unable to distinguish between natural variations and effects caused by anthropogenic factors. Natural variability creates 'noise' in the system, and anthropogenic influences must be of a certain magnitude to be distinguishable from natural variability (Martin & Richardson, 1995). In this kind of study the identification of organisms at species level within communities represents the greatest constraint in terms of both time and costs, so that reliable use of a reduced taxonomic resolution has been an important development in the practical assessment of pollution changes (Giangrande *et al.,* 2005). Some studies have shown that little information is lost by working at a higher taxonomic level (e.g. Family or even Phylum), and there are theoretical reasons and empirical evidence that in this way community responses may still be easily detected (Warwick, 1988 & 1993; Gyedu-Ababio, 1999 and Danovaro *et al.,* 1995). In marine systems aggregation to higher taxonomic levels is possible due to the large number of phyla (Attrill & Depledge, 1997). Warwick (1988) analysed the benthic invertebrate data obtained before and after the Amoco Cadiz oil tanker disaster off the coast of Brittany, and demonstrated that patterns of community change following the oil spill were equally apparent at species and phylum level. Only five phyla were used (Annelida, Crustacea, Echinodermata, Mollusca, others grouped as one group), which would require a low level of expertise to detect an impact. Using higher taxonomic levels also allows different geographical areas, which may have naturally different species assemblages but similar ranges of phyla and families, to be compared (Warwick & Clarke, 1993a). This approach, called Taxonomic Sufficiency (TS), completely bypasses the importance of indicative species (Giangrande *et al.,* 2005). However, Martin and Richardson (1995) believe that a wide range of approaches from both chemistry and biology are necessary to relate contaminat concentrations and their potential impacts. Martin and Richardson (1995) state that, an assessment of the acceptability of a complex waste discharge into a marine system requires the following approaches: "An accurate chemical analysis of the major compartments (water, air, tissue, sediments) to establish the presence, concentrations, loading and exposure durations for chemical contaminats. Toxicity and biomarker tests to establish that exposures, biological responses, or effects are caused by the released contaminants; and ecological field assessments to document the extent and duration of the resulting impacts or to document recovery in those instances where a contaminant release scheme has been modified."

Increasing use is being made of meiofaunal communities and population densities of particular species as indicators of pollution on sandy beaches, because they occur in numbers

sufficient for valid statistical analysis, which is not always so for larger animals (Hennig *et al.,* 1982). Other advantages of meiofauna include their small size and high density allowing quantitative sampling; high turnover and thus rapid response to disturbance; lack of larval dispersion and continuous reproduction *in situ* thus giving a relatively stable temporal baseline; sensitive to changes in environmental conditions (Higgins & Thiel, 1988; Mirto & Danovaro, 2004 and Vassallo *et al.,* 2006). Meiofauna normally have high species richness and therefore offers the possibility that trends in this community parameter can be used. Furthermore, it is possible that working at a higher taxonomic level than species may convey sufficient information to detect real differences in meiofaunal community structure. Sublethal effects causing reductions in fecundity or growth can manifest themselves in measured structural parameters of the community in a shorter time in the meiofauna due to the faster generation times (Moore & Bett, 1989).

Disadvantages of using meiofauna as pollution indicators according to Warwick (1993) include the fact that their taxonomy is considered difficult and the identification of almost all meiofaunal taxa to species level presents difficulties even in well studied habitats in Europe and North America. In many other parts of the world the fauna is almost completely unknown. However, there are factors that mitigate against this problem such as the robustness of community analysis to the use of taxonomic levels higher than species and the wide distribution of most meiofaunal genera. Community responses of the meiofauna to pollution are not well documented, so there is not an extensive body of information in the literature against which particular case histories or sites such as the study reported here, can be evaluated (Gheskiere *et al.,* 2005; Menn, 2002; Moreno *et al.,* 2006 and McLachlan *et al.,* 1977).

Taxon diversity of the meiofauna phyla has been proposed as a possible tool for the assessment of pollution effects by Herman *et al.* (1985). Species determination can only be done realistically by a group of specialists in the various major taxa (Heip *et al.*, 1988). Taxon diversity is lower in polluted conditions; this is caused mainly by the disappearance of some rare taxa e.g. in the Ostracoda, Gastrotricha, Halacarida, Hydrozoa, Tardigrada (Vinx & Heip, 1991). Amjad & Gray (1983) also found a decrease in the number of meiofauna taxa along an organic enrichment gradient. Gyeu-Ababio *et al.* (1999) stated that at present, there is little empirical evidence to suggest that ecologically-similar species, belonging to the same genus or family, respond differently to pollution effects. In other words species within the same family may have similar reactions to specific pollutants. Also, there are strong indications that pollution effects are detectable at even higher taxonomic levels than genus or family (Warwick, 1988). Thus when assessing the impacts of pollution, it may not be necessary to work at the species level (Heip *et al*., 1988; Warwick, 1988; Danovaro *et al*., 1995). Factors that influence the occurrence and abundance of species (both natural and pollution factors) may still be recognised from monitoring based on higher taxonomic levels (Herman & Heip, 1988).

Spatial patterns in community structure have also been examined in terms of functional diversity as opposed to taxonomic diversity. For example, studies have been done where nematodes have been divided into feeding types instead of taxonomic groups (Netto *et al.*, 1999). The functional role, in terms of feeding type, for each species can be deduced from the physiognomic characters of the buccal cavity. The four feeding groups proposed by Wieser (1953a) were:

1A. Species with no buccal cavity, or a narrow tubular one, regarded as " selective deposit feeders" which ingest bacterial-sized particles.

1B. Species with a large buccal cavity, but unarmed with teeth are regarded as "non-selective deposit feeders".

2A. Species with a buccal cavity armed with small or moderately sized teeth are regarded as "epigrowth" or diatom feeders.

2B. Species with large teeth or jaws are considered the "predator/omnivore" group.

The Nematode/Copepod ratio (N/C) proposed by Raffaelli and Mason (1981) as a tool for pollution monitoring using meiobenthic organisms has provoked much discussion and controversy in the literature. They compared the response of nematodes and copepods to organic pollution in intertidal areas along the British coast. They sampled to a depth of 35 cm and found that the ratio of total nematode to total copepod densities of all individuals of all species in these two groups was highest where sewage pollution was most obvious. In particular, an increase in the abundance of deposit feeding nematodes was noted, while the copepods decreased in number. Deposit feeding nematodes (1A and 1B of Wieser (1953)) take advantage of a higher biomass of bacteria and detritus caused by organic pollution and extremely high densities of meiobenthos, especially nematodes can occur.

The proposal of Raffaelli and Mason (1981) of this very simple method for pollution monitoring, stimulated further research. Most studies were carried out on organically-enriched beaches along the British coasts. Within these areas the N/C ratio increased in response to the presence of large quantities of organic wastes (Warwick, 1981; Raefaelli, 1982; Lambshead, 1984 and Shields & Anderson, 1985). Similar observations were made in the Oslofjord in Norway by Amjad and Grey (1983).

The N/C ratio also increased with decreasing particle size, but ratios from polluted sites were always extremely high. Ratios from clean beaches were low and always less than 100, even for muddy sites. Amjad & Gray (1983) sampled the Oslofjord and found mean ratios of 125.4 ± 54.1 SD in the high organic pollution zone near Oslo and mean ratios of 19.6 ± 14.3 SD in the zone they considered unpolluted. Raffaelli (1982) sampled 17 sandy beaches on the Scottish coast with varying degrees of sewage pollution and found mean ratios of 179.5 ± 140 SD where the organic input into the study area was six times that normally found in Scottish waters. At beaches classified from occasionally polluted to relatively clean by traditional water quality measures, Raffaelli (1982) found mean ratios of 62.8 ± 58 SD and at sites considered unpolluted he found mean ratios of 3 ± 1.5 SD. All intertidal sites with fine as well as coarse sediments with ratios exceeding 100 were polluted with sewage (Amjad & Gray, 1983 and Raffaelli, 1982). Some sublittoral ratios from unpolluted sites were relatively high ranging from 8 to 46, but never approached the very high values characteristic of polluted intertidal areas. The sublittoral N/C ratios also increased with depth (Amjad & Gray, 1983).

Coull *et al*. (1981) thoroughly discussed the validity of the N/C ratio and pointed out that spatial and temporal variations, as well as other ecological processes such as predation could alter the ratio. These authors stated that the complex meiofaunal community structure should not be reduced to a single ratio. Platt *et al.* (1984) and Lambshead (1986) also raised the possibility that nematode and copepod populations may be influenced independently by various ecological factors, including pollution, and that the simple N/C ratio is inadequate in that it is difficult to relate to environmental parameters. They found it to over simplify a highly complex set of relationships. The N/C ratio is directly influenced by granulometry, which affects nematodes and copepods in different ways, nematodes preferring mud and copepods sand (McLachlan *et al.,* 1981; Warwick, 1981 and Vinx & Heip, 1991). According to McLachlan *et al.* (1981), proportions of nematodes decrease and harpacticoids increase with increasing particle size above the range of 0.2 to 0.9 mm. From these studies it is suggested by McLachlan *et al.* (1981) that nematodes should disappear above a mean particle size of 1.34 mm and harpacticoids should disappear below 0.07 mm particle size.

Warwick (1981) proposed a refinement of the ratio where the number of copepods are compared to the number of group 2A nematodes (epigrowth or diatom feeders) only, as this is the group that depends on the same food source as the copepods. If copepods are more sensitive to the effects of pollution than nematodes, then changes in the proportion of copepods relative to type 2A nematodes (Warwick, 1981) might be a useful indicator to separate the effects of pollution from those caused by changes in other environmental variables. Warwick (1981) stated that the scatter of ratio values recorded by Raffaelli &

Mason (1981) was too wide and that the ratio had little precision beyond the observation that grossly polluted beaches had values over 100, that on such beaches pollution and its effects were easily observed and that if meiofauna are to be used in a monitoring programme they must respond to pollution before it becomes visually obvious. Warwick (1981) suggested that pollution might be indicated by N/C ratios of around 40 for fine sediments and 10 for sands when only using the type 2A nematodes. These values are much lower than the values of over 100 suggested by Raffaelli and Mason (1981).

Coull and Wells (1981) found no relationship between the N/C ratio and pollution when sampling mud flats in New Zealand. However this was probably due to the fact that they sampled sediments to 1-2 cm depth as they believed that the meiofauna were restricted to the upper 1 to 2 cm of oxidized sediment only and therefore would have missed a large percentage of the nematodes. Data from sub-littoral muds suggest that nematodes can penetrate to depths below 5cm due to their ability to exist anaerobically for long periods (McLachlan *et al.*, 1977). Reise (1985) found a mean number of 39 nematodes and no copepods per 10 $cm^{-3}$ between 5 and 15 cm deep in the anoxic layer on sand flats in Konigshafen, island of Sylt. Gee *et al*. (1985) stated that in organically-enriched sublittoral soft sediments the N/C ratio was unreliable as a biomonitoring tool. They found in their mesocosm experiments, in which sediment grain size and other environmental factors such as temperature and depth were standardized, that the N/C ratio was inversely related to organic pollution. This was due to the fact that a few opportunistic species of harpacticoid copepods that live on or above the sediment surface were able to thrive in the high dose treatments even though the burrowing and interstitial species disappeared completely.

Moore and Pearson (1986) also found an enhancement of copepod density resulting from sewage pollution. They sampled in the Firth of Clyde on the west coast of Scotland where the operating authority was licensed to dump up to $1.55 \times 10^6$ wet tonnes of sewage per year into a 6 $km^2$ disposal area at a depth of 70-80 m. This had led to a sludge depth of 15 cm in the centre of the dumpsite. The overlying water however was found to be fully oxygenated throughout the area and extremely high numbers of an opportunistic species of harpacticoid copepod, *Bulbamphiascus imus*, was found to flourish at the centre of the dumping ground. This species has been observed in culture to make frequent excursions to the sediment surface, where a high oxygen tension is readily available (Moore & Pearson, 1986). In Amjad and Gray's (1983) study in Oslofjord low numbers of copepods and high N/C ratios corresponded with low oxygen levels in the overlying waters. Raffaelli and Mason's (1981) study was of polluted beaches where at low tide when the sediments are exposed and there is no oxygen-rich overlying water, the harpacticoid copepods would have no means of avoiding conditions of low oxygen and high sulphide which might develop (Moore & Pearson, 1986). Moore and Pearson (1986) concluded that the N/C ratio is mainly determined by the availability of high dissolved oxygen levels to the copepod fauna. Similarly Travizi (2000) found that a high N/C ratio seems to be a useful indicator of anoxic stress conditions and not organic enrichment.

Raffaelli (1987) discussed the variable behaviour of the N/C ratio in organic pollution studies and concluded that differences in the habitat requirements of nematodes, mesopsammic and epi-/endosammic copepods affected the responses of these groups to organic pollution. It was found by Raffaelli (1987) that in the sublittoral environment, epipsammic copepods sometimes increase in response to organic pollution while nematodes decrease. The early studies of the N/C ratio by Raffaelli and Mason (1981) were done on sandy beaches where they had positive results and therefore this tool may be appropriate for this environment (Moore & Pearson, 1986). Here one is dealing mainly with mesopsammic copepods and organic pollution is rarely severe enough on high energy beaches to produce a significant decline in nematodes (Raffaelli, 1987). Shiells and Anderson (1985) proposed a possible improvement to the ratio whereby only interstitial species are included, so that only those animals occupying the same micro-habitat are compared.

In terms of oil and diesel contamination on beaches the N/C ratio appeared to have some merit. On beaches near the Amoco Cadiz wreck, Bodin (1988) showed that just after the Amoco Cadiz oil spill a considerable fall in densities occurred, particularly among the harpacticoid copepoda, and an increase of the N/C ratio, which indicated that copepods were more sensitive than nematodes both to hydrocarbon toxicity and anoxia. Bodin (1988) suggested that for a pollution impact survey of the meiofauna, it may be sufficient to study only harparcticoid copepods but gave a warning against the simplistic use of the N/C ratio, especially for long term studies. Carman *et al*. (1997) examined the direct and indirect effects of diesel-contaminated sediments on microalgae, meiofauna and meiofauna-microalgae trophic interactions. Grazing on microalgae by copepods was reduced in high diesel treatments, primarily because of high copepod mortality. Nematode grazing rates increased significantly in high diesel treatments, indicating possible competition for microalgae between copepods and nematodes. However total grazing on microalgae was reduced and the large increase in microalgae observed was likely a consequence of reduced total meiofaunal grazing. Microalgal activity was possibly also stimulated in high diesel concentration treatments. The elimination of copepods by high PAH (polycyclic aromatic hydrocarbons) caused the N/C ratio to increase significantly.

In laboratory experiments on the effects of organic enrichment on meiofauna, Sandulli & de Nicola-Giudici (1989) found a general reduction in all the faunal groups with high organic loading. Reduction in the total numbers and species richness of meiofauna abundance after contamination with hydrocarbons has also been demonstrated in the field (Danovaro *et al.*, 1995 and Ansari & Ingole, 2002). Therefore an impacted site could have a N/C ratio well below 100 due to both groups being in low abundance. On high energy, medium sand beaches in South Africa, it was found that it was possible for perturbations to increase or decrease the ratio, or even leave it unaffected while significantly altering the densities of both nematodes and copepods in the same direction (Platt *et al.,* 1984).

Lee *et al.* (2001) assessed the use of the N/C ratio in the monitoring of metal pollution on high-energy beaches and found that metal enrichment generally drives down both species diversity and density of individuals of all meiofaunal taxonomic groups. Thus, the N/C ratios in impacted areas are based on very low numbers of organisms and, therefore, small changes in the density of either taxa could have a pronounced effect on the ratio. Lee *et al.* (2001) found that the ratio was not a good predictor of metal pollution due to the generally low densities of meiofauna on impacted beaches and suggested that harpacticoid copepod densities may be a better indicator for broad-based surveys, where the dominant pollutant is expected to be metals.

In conclusion it appears that an increase in the N/C ratio may indicate effects of pollution if that pollutant results in decreased oxygen levels (Moore & Pearson, 1986 and Travizi, 2000). Copepods have been found to be more sensitive to hydrocarbon toxicity than nematodes (Bodin, 1988; Carmen *et al.,* 1997) and more sensitive to some pesticides eg. atrazine, a widely used herbicide (Bejarano *et al.,* 2005). Where a pollutant is equally toxic to both nematodes and copepods then the N/C ratio would be unaffected as there would be a reduction in density and diversity of all taxonomic groups (Ansari & Ingole, 2002; Danovaro *et al.,* 1995; Lee *et al.,* 2001 and Nicola-Giudici, 1989). High organic pollution in sublittoral environments where high oxygen levels in the overlying water are maintained may result in a lower N/C ratio due to the proliferation of opportunistic species of harpacticoid copepods (Gee *et al.,* 1985 and Moore & Pearson, 1986). Organic enrichment was indicated by an increase in the N/C ratio in Raffaelli and Masons' (1981) study where, during low tide and subsequent sediment exposure to the atmosphere, there was no oxygen rich overlying water to prevent the reduction in opportunistic harpacticoid copepods as suggested above. However as indicated by McLachlan *et al.* (1981), on extremely coarse grained beaches nematode numbers decrease and harpacticoid copepods dominate. In such a case large interstitial spaces between the sand grains would also facilitate greater oxygenation of the sediment and thus a high organic

loading would have less of an effect on the N/C ratio. The N/C ratio may therefore be useful in detecting pollution in well defined environments and its usefulness should be assessed in relation to what is known about the type of habitat, pollutant, and other factors especially oxygen levels.

## 1.4 This Study

The purpose of this study was to measure the impact of the Illovo Sugar by-products effluent on the marine environment at Sezela. Illovo Sugar By-products (ISBP) currently discharges about 3100 $m^3$ of effluent per day during the sugar cane harvesting season, via a surf-zone discharge 200 m north of the mouth of the Sezela River. The effluent is a mild acetic acid with a trace of furfuraldehyde. Furfural is the liquid aldehyde obtained by distilling acid-digested sugar cane by-products. The pH of the effluent is generally around 2.8, and the chemical oxygen demand (COD) is about 16000 mg/l. Under normal conditions of sand movement, the pipe end is positioned about 50 m offshore on a rocky seabed. The effluent pipeline of approximately 20 cm diameter runs across the beach, between rocks into the surf zone. Some of the effluent is released back towards the shore by wave action. Sandy beach meiofaunal communities were used as indicators of environmental degradation. Meiofauna were also appropriate in this situation as trace amounts of furfural occurs in the effluent, and furfural is used as the active ingredient in a product, Crop Guard, developed by Illovo Sugar to kill parasitic nematodes in crop fields. Furfural reacts with the cuticle of the nematode causing suffocation (www.cropguard.co.za). Generally the largest proportion of the beach meiofauna consists of nematodes (Brown & McLachlan, 1990). Reduced diversity of taxonomic groups and reduced total numbers of meiofauna would therefore be expected close to the effluent outfall. The N/C ration may react in either direction or remain unchanged depending on the sensitivities of the two groups to the effluent.

Analyses were also carried out for a range of physical and chemical variables and for an assessment of organic enrichment. These analyses included both interstitial waters and sediments. Chemical oxygen demand and nitrogen were measured for the interstitial waters and the sediments, and dissolved oxygen, pH, salinity and ammonia were measured for interstitial waters.

# 2. MATERIALS & METHODS

## 2.1 Study Area

Eight stations were sampled for meiofauna along the beaches at Sezela on 7 different occasions. This sampling frequency and timing was chosen to assess any seasonal effects on meiofauna, and meiofaunal changes during the period when the factory was not pumping effluent to sea. The seasons were chosen according to the meteorological definition as being four equal length periods of 3 months each. This definition points to 1 December being the beginning of summer and 1 June being the beginning of winter (Alpert *et al.*, 2004). Therefore autumn would be the 1st March to the end of May and spring would be the 1st September to the endof November. This definition more closely reflects periods of differing environmental temperatures than the astronomical definition (Alper *et al.*, 2004) Table 2.1 shows the sampling dates and when effluent was being pumped to sea.

Table 2.1
Dates when samples were collected

| Sampling date | Season | Active effluent discharge | Period of discharge prior to sampling |
|---|---|---|---|
| 4th July 2000 | Winter | yes | 2 months |
| 30th August 2000 | Winter | yes | 3 months |
| 13th December 2000 | Summer | yes | 7 months |
| 26th January 2001 | Summer | yes | 1 month |
| 8th March 2001 | Autumn | no | no discharge for 1 month |
| 9th April 2001 | Autumn | no | no discharge for 2 months |
| 2nd January 2002 | Summer | yes | 1 week (following a shut down of 5 days) |

The samples were taken on the following dates: 4 July 2000 (winter); 30 August 2000 (winter); 13 December 2000 (summer); 26 January 2001 (summer); 8 March 2001 (autumn); 9 April 2001 (autumn); and 2 January 2002 (summer). There was a short shutdown over Christmas from 23rd December 2000 to 27th December 2000, after which there was a short start up until the 1st February 2001 when the factory closed until the 18th of April 2001. Therefore the March and April 2001 sampling occasions were during the closed period. The beach was rated according to McLachlans' proposed rating system (McLachlan, 1980) for defining sandy beaches in relation to exposure shown in Table 1.1. Table 2.2 shows how the beach at Sezela was scored and the total for Sezela beach came to 14 giving it an exposure rating of 'exposed' (see Table 1.2).

Table 2.2
Exposure rating scores for Sezela beach (see Table 1.1; from McLachlan, 1980).

| Parameter | Rating | | | | | score |
|---|---|---|---|---|---|---|
| Wave action: | Continuous, heavy, wave height mostly exceeds 1m | | | | | **3** |
| Surf zone width: | Moderate, waves usually break 50-150m from shore | | | | | **1** |
| % very fine sand: (63-125$\mu$m) | <1% | | | | | **2** |
| Median particle Diameter ($\mu$m) | | Slope of intertidal zone >1/10 | 1/10-1/15 | 1/15-1/25 | 1/25-1/50 | <1/50 |
| 500-710 | (1.0-0.5Ø) | 4 | **5** | 6 | 7 | 7 |
| Depth of reducing layers (cm): | | 25 – 50 | | | | **2** |
| Stable macrofaunal burrows: | | Absent | | | | **1** |
| | | Highest exposure | | | | 20 |
| | | Lowest exposure | | | | 0 |
| | | Sezela exposure | | | | **14** |

The location of the sampling stations at Sezela, are shown in Figure 2.1 and an aerial photograph of the area is shown in Plate 2.1. There were three stations to the north of the effluent pipeline, N1, N2 and N3, four stations to the south, S1, S2, S3 and S4, and one station at the effluent discharge point, S0. These stations are described below:

- **Station N3** is located in the first sandy bay south of the Mkumbane river. The beach at this point has a steep slope and the sand is coarse-grained. The beach is backed by primary dunes vegetated by *Scaevola plumieri*.

- **Stations N2 and N1** are less steep than N3 and there are numerous rocks at the waters edge. An eroded runoff channel from a storm water drain was evident at Station N1.

- **Station S0** is at the pipe discharge point next to high rocks, and the discharge pipe is laid through a small gully on the south edge of these rocks (Plate 2.2). This station is located at the southern end of a prominent set of high rocks, and consequently undergoes periods of accretion and erosion (see Plates 2.2 and 2.3).

- **Stations S1 to S4** are all located in a long sandy bay extending south towards the Mdesingane River. Station **S2** is influenced by the Sezela River (the estuary was closed during all 7 sampling periods) and station **S4** is influenced by the Mdesingane River, which was flowing into the sea during all 7 sampling periods. The bay is bordered by a promontory of rocks south of the Mdesingane River, beyond station S4.

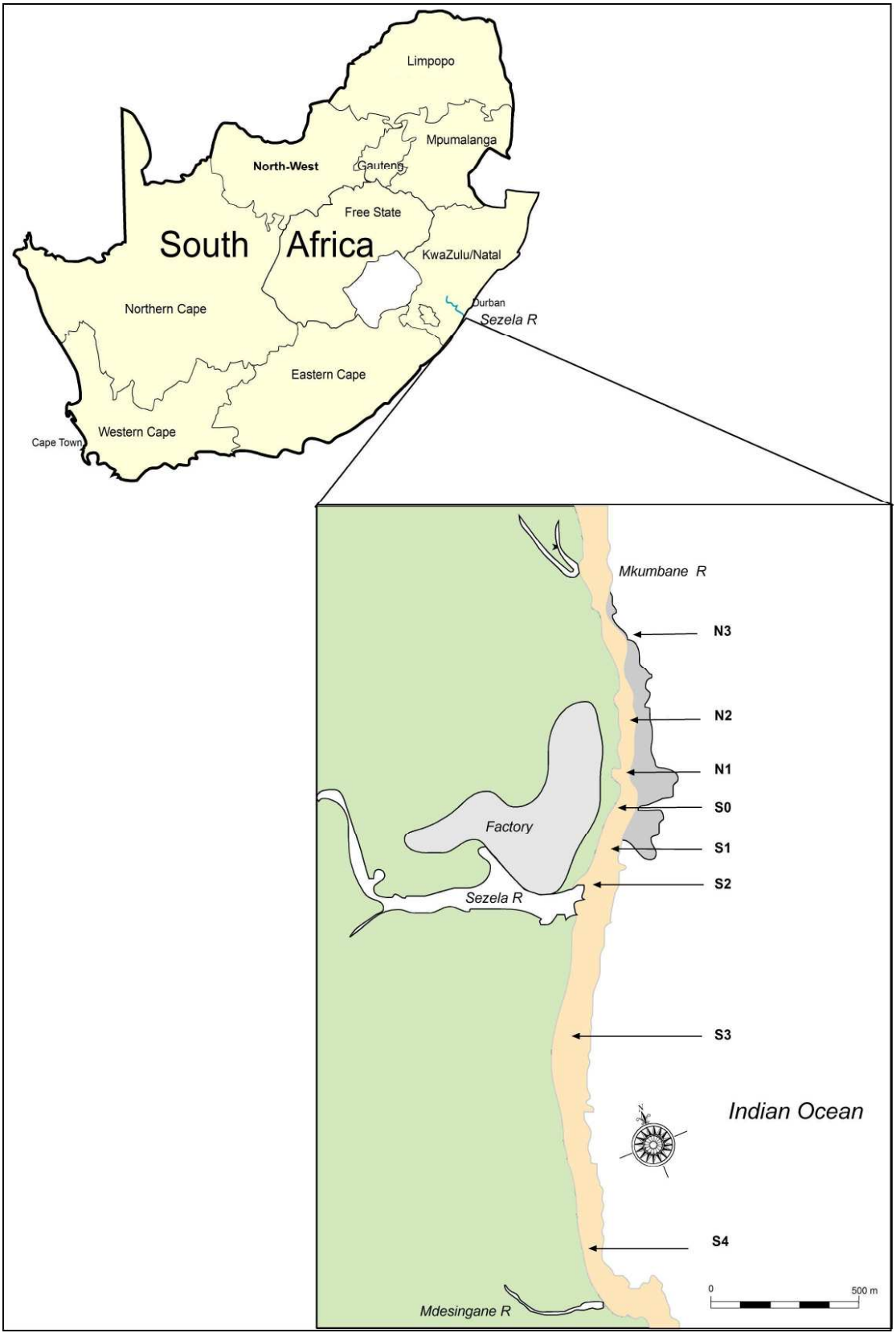


Figure 2.1. Map showing the sampling sites for the Sezela beach study.

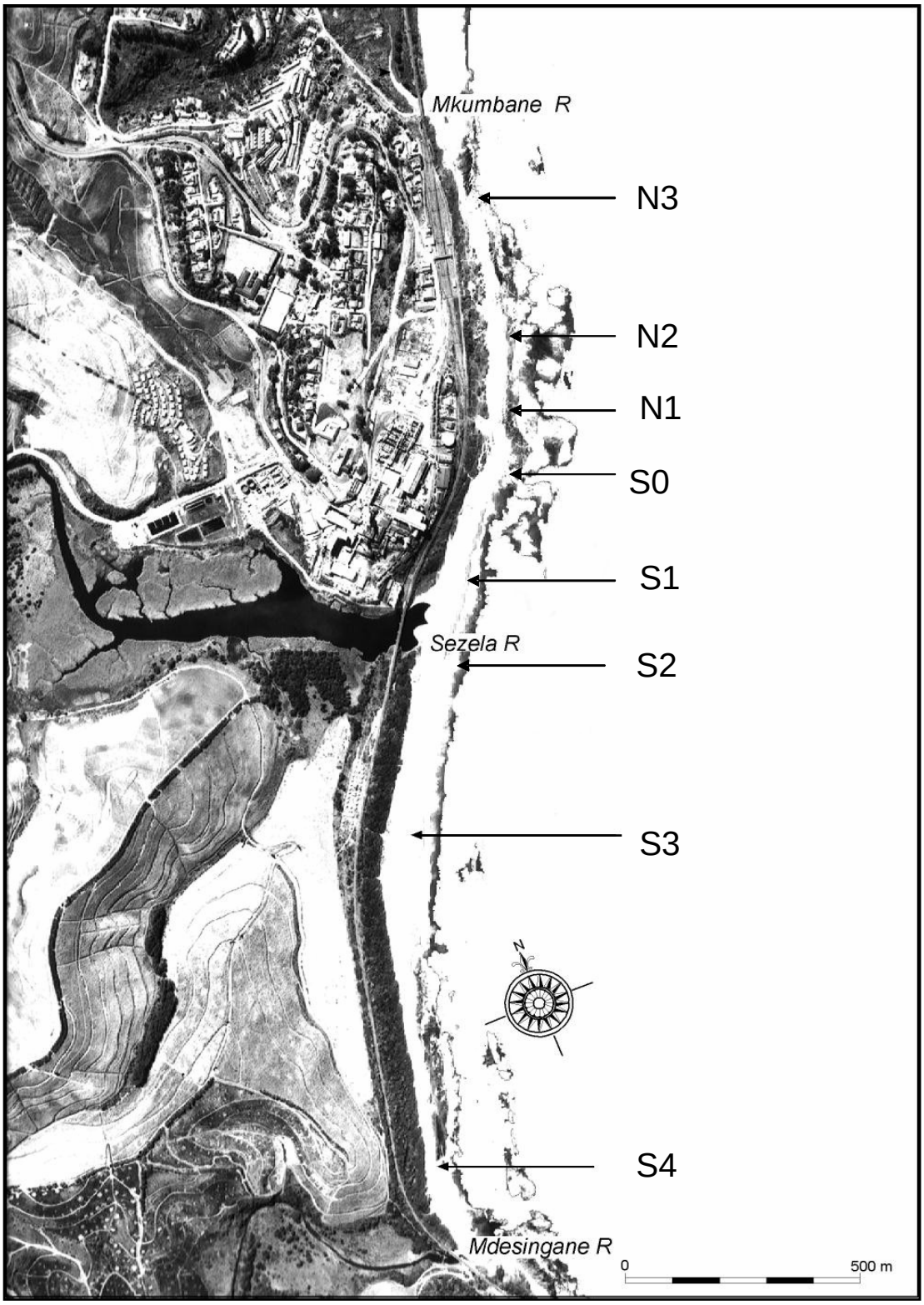


Plate 2.1. Map of the study area at Sezela beach

Plate 2.2 Station S0 where the effluent pipeline enters the sea.

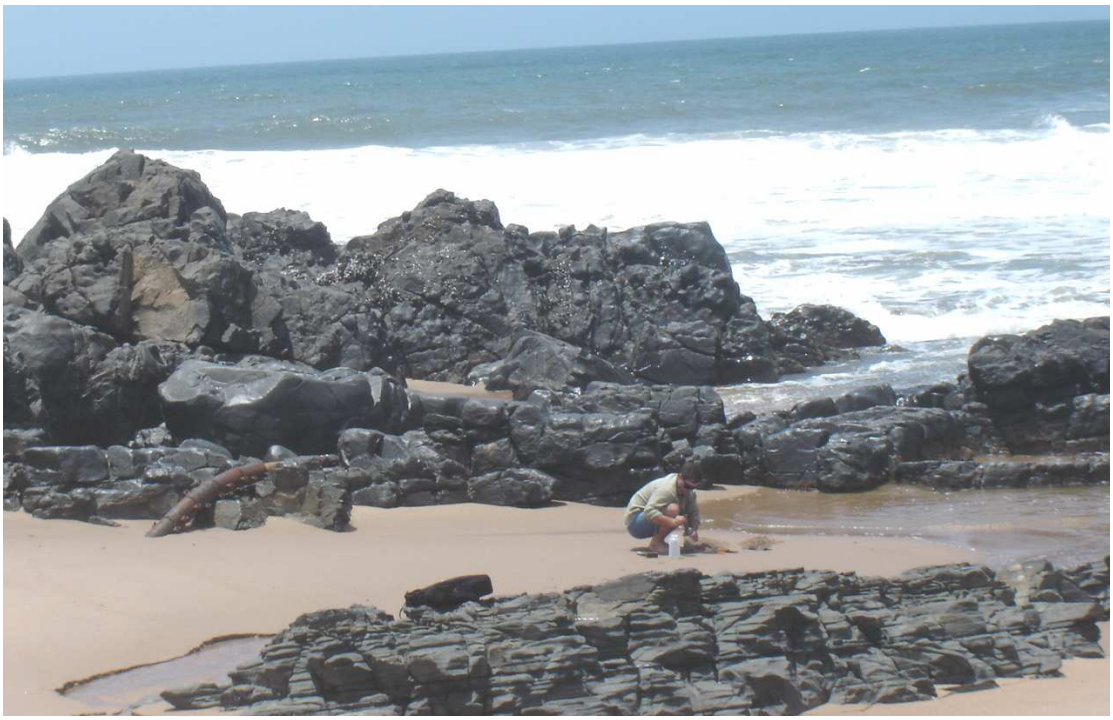

Plate 2.3 Samples being collected at station S0

## 2.2 Meiofauna sampling and counting techniques

Each station was sampled in the swash zone at low spring tide. Each sample consisted of two duplicate 200 ml sand cores (Plate 2.4) taken to a depth of 20 cm, which were combined into one sample for counting. Of the eight stations sampled, four replicates were taken at one of the stations at each sampling time to determine the variability that occurred within that station. For the last two sampling times, all the stations were sampled in at least triplicate and each replicate was counted separately. Replicates were not taken at seven of the stations for the first five sampling trips due to time and financial constraints. It was felt that it was more important to sample on more occasions to obtain more seasonal data than to have more replicates and fewer sampling occasions.

The samples were returned to the laboratory two hours after collection and preserved in 5% formalin solution. The meiofauna were extracted using a modified Oostenbrink separator (Plate 2.5) (Fricke, 1979) and a 45 micron sieve (Plate 2.6) and the sieved meiofauna were washed from the sieve into 100 ml bottles with 70% alcohol. Counts were made of each meiofaunal group distinguishable at a 63 X magnification by concentrating them on a 45 micron sieve and washing them into a 25 ml tray designed to fit onto the stage of a compound microscope. The tray was scanned to count each meiofaunal group distinguishable at a 63 X magnification. The nematodes were divided into the 4 feeding groups and the harpacticoid copepods and the annelids were divided into families. Copepod nauplii, which are the juvenile life stages of Copepods, were counted as a separate group as it was not possible to distinguish what type of copepod they were e.g. Calanoid, Harpacticoid, Cyclopoida. Nauplii of other groups such as the annelids were not observed in the meiofauna. The taxonomic groups counted are given in Table 2.2. Spreadsheets of the data for animals grouped into higher taxonomic levels (Table 2.3) were also prepared so that comparative analyses of the data could be carried out to determine whether analysing to lower taxonomic levels is necessary to determine the extent of an environmental impact at Sezela beach.

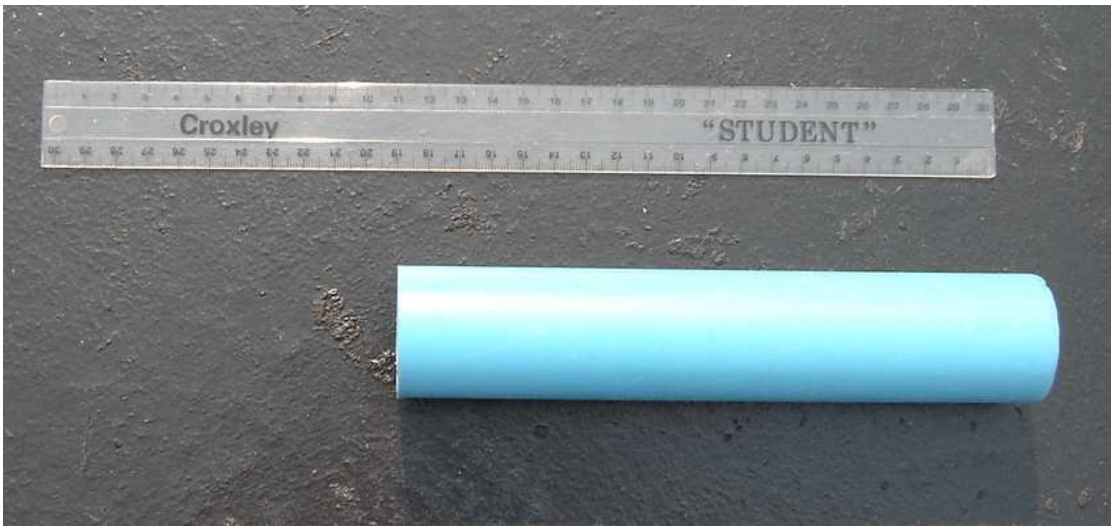


Plate 2.4 Corer used to collect meiofauna samples. Depth: 20 cm, diameter: 3.6 cm, volume: 200 ml.

Plate 2.5 Oostenbrink separator (Fricke, 1979).

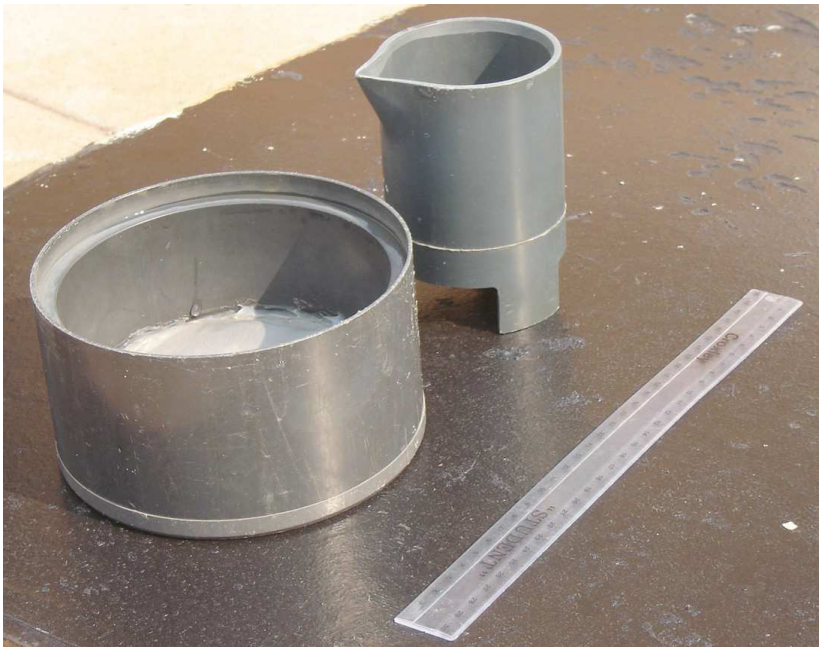

Plate 2.6 45 micron sieves used to collect the meiofauna from the elutriate produced by the Oostenbrink separator.

Table 2.2

Twenty eight animal and protistan groups counted from each Sample

| PHYLUM | CLASS | ORDER | FAMILY | FEEDING GROUP |
|---|---|---|---|---|
| Platyhelminthes | Turbellaria | | | |
| Nematoda | | | | 1A; Selective deposit feeders (Bacterial feeders) |
| | | | | 1B: Non-selective deposit feeders |
| | | | | 2A: Epigrowth or diatom feeders |
| | | | | 2B: Predators/Omnivores |
| Annelida | Oligochaeta | | | |
| | Polychaeta | | Dynophilidae | |
| | | | Hesionidae | |
| | | | Nerillidae | |
| | | | Pisionidae | |
| | | | Polygordiidae | |
| | | | Protodrilidae | |
| | | | Saccocirridae | |
| | | | Syllidae | |
| Tardigrada | | | | |
| Arthropoda | Arachnida | Acarina | Halacaridae | |
| Subphylum: | | | | |
| Crustacea | Ostracoda | | | |
| | Copepoda | Harpacticoida | Canuellidae | |
| | | | Cylindropsyllidae | |
| | | | Darcythompsoniidae | |
| | | | Paramesochridae | |
| | | | Tachidiidae | |
| | | | Tisbidae | |
| | | Cyclopoida | | |
| | Copepod nauplii (ie. Juvenile copepods counted as one group) | | | |
| | | Amphipoda | | |
| | | Isopoda | | |
| Sarcomastigophora | | | | |

Table 2.3
14 animal and protistan groups used for analysis to a higher taxonomic level.

| PHYLUM | CLASS | ORDER |
|---|---|---|
| Platyhelminthes | Turbellaria | |
| Nematoda | | |
| Rotifera | | |
| Gastrotricha | | |
| Kinorhyncha | | |
| Annelida | Oligochaeta | |
| | Polychaeta | |
| Tardigrada | | |
| Arthropoda | Arachnida | Acarina |
| Sub phylum Crustacea | Ostracoda | |
| | Copepoda | Harpacticoida |
| | Copepod nauplii | |
| | Malacostraca | Isopoda |
| Sarcomastigophora | | |

## 2.3 Sampling and analysis of physical and chemical variables

The eight stations were sampled separately for physical and chemical analysis at the same time as the samples taken for biological purposes. Separate sediment samples were taken at each station and analysed for grain size. The methods used are described by Leuci (1998) and included wet sieving, dry sieving and a settling tube. 100 $cm^3$ of sediment was collected at each station and 50 $cm^3$ was used from each sample for grain size analysis. This was wet sieved using a 63 µm sieve to separate the mud fraction from the sand and gravel. The sand and gravel fractions remain in the sieve and the mud fraction was collected after passing through the sieve. The gravel and sand were transferred to a pre-weighed beaker and the mud fraction funnelled into another pre-weighed beaker. The two beakers with sediment were placed under infrared lamps and dried at 50 $^oC$ and the dry weight recorded. The sand and gravel fractions were then dry sieved through a 2 mm sieve to separate the gravel from the sand, and the two separated fractions were then weighed. The size distribution of the sand-sized particles was measured using a settling tube. Sediment particle sizes were categorized as percentage gravel (> 2 mm), very coarse sand (1 - 2 mm), coarse sand (0.5 - 1 mm); medium sand (0.25 – 0.5 mm); fine sand; (0.125 – 0.25 mm); very fine sand (0.063 – 0.125 mm) and mud (0 < 0.063 mm).

Interstitial water samples were collected at each station by digging down to the water table. For the remaining 50 ml of sediment collected as well as for interstitial water samples collected at each station at each sampling time, analysis for a range of physical and chemical parameters for indications of organic enrichment were carried out. These included salinity, pH, Chemical Oxygen Demand from permanganate (CODmn) in milligrams per litre, dissolved oxygen (DO) in milligrams per litre, ammonia ($NH_3$) in micrograms per litre and kjeldahl nitrogen (Kjel N) in micrograms per litre for the waters. For the remaining 50 $cm^3$ of sediment, CODmn in milligrams per gram and kjel N in micrograms per gram were also measured for the sediments. CODmn is defined as the amount of oxygen needed to oxidise the organic and inorganic material in a sediment sample.

The salinity (ppt or %o), pH and DO (mg $l^{-1}$) were measured using a YSI Model 57 Oxygen Meter. The methods employed to analyse the rest of these parameters were in accordance with standard methods proposed by Watling (1981) for use in South African marine pollution surveys and samples were analysed 48 hours after returning them to the laboratory. For chemical oxygen demand from permanganate (CODmn) from sediment analysis approximately 1 gram of sample was weighed out and placed in a 250 $cm^3$ Erlenmeyer flask. 10 $cm^3$ of N/80 Potassium Permanganate and 0.5 $cm^3$ 0f 33% Sodium Hydroxide was added to the Erlenmeyer flask together with 100 $cm^3$ of deionised water. This mixture was heated in a water bath for 30 minutes and then cooled to room temperature. 25 $cm^3$ of Magnesium Sulphate solution and a spatula full of Potassium Iodide was added and this mixture was titrated to a starch (blue to clear end point) with 0.01 N Sodium Thiosulphate. The results were reported as mg $g^{-1}$ CODmn. For the Kjeldahl nitrogen analysis of sediments 0.1 to 0.3 grams of sample was weighed out into a TKN digestion tube. 2 ml of 1:1 Sulphuric acid and a few bumping stones were added to the tube and heated at 360 degrees Celsius for 30 minutes. After the digestion 30 g $l^{-1}$ of Sodium Hydroxide solution was added to the digestate to neutralize the acid mixture and then the ammonia concentration in this solution was determined calorimetrically. The results were reported as µg $g^{-1}$ (NH3 – N). For the analysis of CODmn and Kjeldahl nitrogen in the interstitial waters the same methods as above were used except 1 gram of sample was replaced by 1 ml of sample and the results were reported as µg $l^{-1}$. An aliquote of sample was place in an auto-analyser for the analysis of ammonia.

## 2.4 Univariate statistical analysis of meiofaunal data

A variety of different indices were calculated to measure various attributes of the community structure in the samples. These included the total number of taxa (S), the total number of individuals (N), species richness- Margalef's index (d), species diversity- Shannon-Wiener diversity index (H'), and species equitability- Pielou's evenness index (J) (Clarke & Warwick, 1994a).

Margalef's index of richness (d) incorporates the total number of individuals (N) and is a measure of the number of species (S) present for a given number of individuals:

$$d = (S-1)/\log N$$

The Shannon-Wiener diversity index:

$$H' = - \Sigma_i \, pi(\log pi)$$

where *pi* is the proportion of the total count arising from the *i*th species. This incorporates both the species richness and equitability components.
Equitability was expressed as Pielou's evenness index:

$$J' = H'(\text{observed})/H'\text{max}$$

where H'max is the maximum possible diversity which would be achieved if all species were equally abundant (= log S).

The above indices were calculated using the PRIMER statistical package developed by Plymouth Marine Laboratories. This software is extensively used in marine benthic monitoring and pollution impact assessment studies (Clarke & Warwick, 1994a). The Nematode/Copepod ratio was also calculated for each sampling occasion at each station.

The above indices were statistically analysed by analysis of variance (ANOVA) to determine any significant difference between sites over the 7 sampling periods and between the different

seasons when samples were taken. The software used was the Sigma Stat 3.1 package. Where the data failed to meet the statistical assumptions of parametric ANOVA techniques, namely normality, the data was $\log_{10}$ transformed to meet these assumptions. Sigma Stat uses the Kolmogorov-Smirnov test (with Lilliefors' correction) to test for normality. Where the normality tests failed even after transformation, the Kruskal-Wallis procedure was used to conduct analysis of variance by ranks. When the null hypothesis of no difference was rejected at a probability $P<0.05$, differences of ranks were compared using a pair-wise multiple comparisons procedure (Student-Newman-Keuls Method).

## 2.5 Multivariate statistical analysis of meiofaunal data

### 2.5.1 Overview of analysis

The statistical analysis of animal community data from sampling of soft sediment benthos has been extensively discussed (Clarke, 1993; Clark & Ainsworth, 1993 and Clark & Warwick 1994b). The main objective of such analysis of the field data is to display community patterns through clustering and to link these patterns to environmental (e.g. sediment particle size, pH, etc.) and pollution variables through ordination. If possible a zone of impact related to the effluent can be identified based on a modified meiofauna community.

The raw meiofauna counts were subjected to multivariate analysis using the PRIMER statistical package. Multivariate methods were used that included an analysis of similarity (ANOSIM) as well as hierarchical clustering (CLUSTER) and multi-dimensional scaling (MDS), these methods are used to explore the degree of similarity between sampling stations as reflected in their community structure. Analysis was first carried out for the last two sampling occasions where at least three replicates were taken at each site. This was carried out to backup the findings of the ANOSIM test. That is if replicates within stations are significantly more similar to one another than to replicates from different stations according to the ANOSIM test, then replicates within sampling stations should tend to group together with a greater similarity to one another than with replicates from different sampling stations, within a CLUSTER analysis. Further to this, effluent was not being pumped to sea on the 9$^{th}$ April 2001 whereas it was on the 2$^{nd}$ January 2002 so a comparison between sampling times relating to effluent discharge could also be made. Thereafter CLUSTER analysis and MDS ordinations were generated for all the samples using mean counts per station. A SIMPER analysis was then carried out to identify the taxa which were the most responsible for the groupings observed in the CLUSTER and MDS analyses. The above analyses were conducted for samples where all taxonomic groups were identified (Table 2.2) and for samples where simplified data with respect to counts only of higher taxonomic levels (Table 2.3). Then a BIO-ENV analysis was run which links the biotic multivariate patterns to the abiotic multivariate environmental patterns and indicates which abiotic or environmental variables are the most significant for the observed biotic community pattern. These analyses are described more fully in the following sections.

### 2.5.2 Similarity matrix and ANOSIM

All these methods start explicitly from a matrix of similarity coefficients computed between every pair of samples**.** The coefficient is an algebraic measure of how close the abundance levels are for each species, averaged over all species, and defined such that 100 % represents total similarity and 0 % complete dissimilarity. However, most matrices in benthic invertebrate survey data have more than half of the data entries as zeros. Field *et al.* (1982) adopted a measure for comparing samples, which is not affected by joint absences and

sufficiently robust for marine biological data, yet giving more weight to abundant species than to rare ones. This is the Bray-Curtis coefficient and has the form:

$$Sjk = 100\{1- \sum \mid Yij - Yik \mid \Big/ \sum (Yij + Yik)\} \quad \text{(Field } \textit{et. al.} \text{ 1982)}$$

Sjk represents the percent similarity between samples j and k
Yij represents the i*th* species in the j*th* sample
Yik represents the i*th* species in the k*th* sample
As at least three replicates were taken at all the sites for the last two sampling occasions (9th April 2001 and 2nd January 2002), it was possible to test the null hypothesis that there are no differences in community composition between the eight stations. A test statistic can be computed, reflecting between-site and within-site variability. This test was an analysis of similarity (ANOSIM) and is built on a simple permutation procedure, applied to the Bray-Curtis similarity matrix which is used for the ordination analysis and classification of samples.

If $\bar{r}_w$ is defined as the average of all rank similarities among replicates within stations, and $\bar{r}_b$ is the average of rank similarities arising from all pairs of replicates between different stations, then the suitable test statistic is:

$$R = (\bar{r}_b - \bar{r}_w) / (M/2) \text{ (Clarke \& Green, 1988)}$$

Where M = n(n - 1)/2 and n is the total number of replicates under consideration. This denominator constant is chosen so that:

- *R* can never technically lie outside the range (-1,1);
- *R* = 1 only if all replicates within sites are more similar to each other than any replicates from different sites;
- *R* is approximately zero if the null hypothesis is true, so that the similarities between and within sites will be the same on average.
- A negative value would indicate that replicates from different sites are more similar to each other than replicates within sites and should only happen if for example the labelling of samples was mixed up by accident.

To test whether the calculated *R* statistic is significantly different from zero it is recalculated under permutations. If the labels identifying which replicates belong to which sites are randomly mixed up and *R* recalculated and the process repeated a number of times (*T*), then the significance level can be calculated. If the null hypothesis is true that there is no difference between sites then there will be little effect on average to the value of *R* after reshuffling the sample labels.

If only *t* of the *T* simulated values of *R* are as large, or larger than the observed *R* then the null hypothesis can be rejected at a significance level of:

$$100(t + 1) / (T + 1) \text{ \% (Clarke \& Green, 1988)}$$

Therefore if none of the *T* simulated *R* values are equal to or greater than the observed *R* (*t* = 0) and 999 simulations were done, then there is a probability of less than 1 in 1000 that the null hypothesis is true and we can therefore reject the null hypothesis at a value of $\rho < 0.1$ % ($p < 0.001$).

### 2.5.3 Cluster and MDS Ordinations

CLUSTER analysis results in a dendrogram which groups the stations hierarchically according to their Bray-Curtis measure of similarity. MDS uses a two-dimensional scatter plot

to depict relative similarities between stations. These two methods are complimentary and are frequently used to corroborate one another. They provide an objective method for recognising and describing trends.

For the CLUSTER and MDS analyses the raw data were square root transformed. With untransformed data a MDS plot can be distorted when species in a sample have a strong degree of spatial clustering (Clarke & Green, 1988). At the other extreme, an analysis which places weight on a taxon that occurs in low numbers is highly susceptible to the "noise" introduced by the presence of a rare taxa. The practical choice is therefore often between a moderate "root" and fairly severe "root-root" transformation, which retains the hard-won quantitative information but downplays species dominance (Clark & Warwick, 1994a).

This study uses square-root transformation:

$$Yij = \sqrt{X}ij \text{ (Field } \textit{et. al.,} \text{ 1982)}$$

Yij = the transformed value of the entry in the ith row and jth column of the data matrix, i.e. the abundance for the ith species in the jth sample.

For CLUSTER analysis a dendrogram is constructed from the percentage similarities in the matrix using the hierarchical agglomerative method. A similarity matrix is the starting point from which the samples are successively combined into groups and the groups into larger clusters starting with the highest mutual similarities and then gradually lowering the similarity level at which groups are formed. One of the axes represents the full set of samples and the other axis defines a similarity level at which two samples or groups of samples are considered to have combined. This is a particularly appropriate representation in cases where the samples are expected to divide into well defined groups, for example if structured or limited by some discontinuous environmental factors (Clark & Warwick, 1994a).

It is important to employ an additional method of presentation such as an ordination technique to show individual relationships between samples and the environmental variables. This is a more appropriate representation when the samples do not group in a well-defined manner and the community is responding to abiotic gradients which are more continuous. Multidimensional scaling (MDS) was used to create a "map" or configuration of the samples constructed in a specified number of dimensions. The distance between points on a plot is a measure of their relative degree of similarity or dissimilarity, e.g. if sample A has a higher degree of similarity to sample B than it does to sample C, then sample A will be placed closer to sample B than it is to sample C in the ordination plot (Gray *et. al.,* 1988). Agreement between the cluster analysis and the ordination strengthens belief in the common conclusions of both.

The success of a 2-dimensional MDS can be measured by comparing the stress value with that of higher dimensions (Clarke & Warwick, 1994a). In theory, stress increases with reducing dimensionality of the ordination, as well as with increasing quantity of data, and the lower the stress value the more accurate the representation of samples in the MDS. The following function calculates the stress value of a 2-dimensional ordination plot:

$$\text{Stress} = \Sigma j \, \Sigma k \, (djk - d\hat{}jk)^2 / \Sigma j \, \Sigma k \, djk^2$$

d^jk is the distance predicted from a fitted regression line or plot of distance against dissimilarity for the n(n – 1)/2 pairs of dissimilarity percentage values that correspond to the dissimilarity percentage from the Bray-Curtis matrix. The djk is the actual distance between the ith and jth sample points on the MDS ordination plot. Therefore if d^jk = djk the stress is zero. Guide values for 2-dimensional ordinations, using the stress function above is as follows (Field *et al.,* 1982):

- Stress value of < 0.05 gives a very good representation with no prospect of misinterpretation;
- Stress value of < 0.1 corresponds to a good ordination with no real prospect of a misleading interpretation;
- Stress value of < 0.2 still gives a potentially useful 2-dimensional picture, though for values at the upper end of this scale too much reliance should not be placed on the detail of the plot;
- Stress value of > 0.3 indicates that the points are close to being arbitrarily placed in the 2-dimensional ordination space (Clark & Warwick, 1994a).

The current studies used 2-dimensions with 20 repetitions of the analysis to ensure that results converge to an optimal configuration.

### 2.5.4 SIMPER analysis

For different sample groups identified as a result of a cluster analysis or an MDS ordination, an important requirement is to identify which taxa primarily account for the observed assemblage differences. By looking at the overall percentage contribution each species makes to the average dissimilarity between two groups, one can list species in decreasing order of importance in discriminating two sets of samples. This is accomplished by the SIMPER routine in PRIMER (Clarke, 1993).

### 2.5.5 BIO-ENV

A BIO-ENV procedure in PRIMER for linking the biotic multivariate patterns to the abiotic multivariate environmental patterns was run. The principle here is that if the suite of environmental variables responsible for structuring the community were known, then samples having rather similar values for these variables would be expected to have rather similar species composition, and an ordination based on this abiotic information would group sites in the same way as for the biotic MDS plot. If key or limiting environmental variables are omitted (not measured), the match between the two plots will be poor.

Rank similarity matrices are generated for the biotic and abiotic ordinations. Two possible matching coefficients are defined between the elements of the respective rank similarity matrices (ri; i = 1,…,N) and (si; i = 1,…,N), where N = n(n-1)/2 and n is the number of samples. The following equation is the weighted Spearman or harmonic rank correlation (Clarke & Ainsworth, 1993):

$$p_w = 1 - c\sum_{i=1}^{N}\frac{(ri - si)^2}{(ri + si)}$$

The constant terms are defined such that, in both cases, p lies in the range (-1, 1), with the value of p = -1 and p = +1 corresponding to the cases where the two sets of ranks are in complete negative agreement or complete positive agreement. Algebraic manipulation shows that c=6/N(N-1). Values of p around zero correspond to the absence of any match between the two patterns, but typically p will be positive as a negative value is unlikely to be attained in practice because of the constraints inherent in a similarity matrix where there is either no similarity between samples or some positive percentage similarity (Clarke & Ainsworth, 1993). Combinations of environmental variables are considered at steadily increasing levels of complexity in seeking a good match in the biotic and abiotic matrices. The closer Pw is to 1 the greater will be the positive correlation of that particular combination of abiotic variables with the biotic community pattern.

### 2.5.5 Nematode/Copepod ratio

The Nematode/Copepod ratio (proposed by Raffaelli & Mason (1981)) for each sample from each sampling occasion was also calculated to see if there was any relationship between sites shown to be impacted using the PRIMER analysis and high N/C ratio values. This was done by dividing the number of individuals of Nematoda by the number of individuals of Harpacticoida from each sample. A one way analysis of variance (ANOVA) was then carried out to test for significant differences between stations over the seven sampling occasions in terms of the N/C ratio.

### 2.5.6 Correlation analysis

Standard product moment correlation coefficients were calculated using Statgraphics version 3.0 between all the physical and chemical variables and the total number of animals, numbers of taxa and the Nematode/Copepod ratio. Then simple regressions were carried out to determine if the correlations were significant for each of the physical and chemical variables against the biological data.

# 3. RESULTS

## 3.1 One-way ANOVA for univariate indices

A one-way ANOVA was carried out to test for significant differences between stations using the complete data set for the seven sampling periods (n = 105) in terms of the following indices:

- S = total number of taxa recorded per sample
- N = mean total number of animals recorded per sample
- d = Margalef's species richness
- J'= Pielou's evenness index
- H'= Shannon-Wiener diversity

The differences in the mean values for the number of taxa (S) per station are shown in Figure 3.1(a) and were greater than would be expected by chance ($P<0.05$) where the F statistic was 2.684 with P=0.020. From pair-wise multiple comparison procedures (Student-Newman-Keuls Method) there was a significant difference between Stations S4 and S2 (P=0.026). Station S4 had the highest number of taxa. It was the furtherest station from the effluent outfall (about 1.5 km) whereas Station S2 was within 400 m of the outfall and in a bay were effluent was observed being pushed ashore by wave action on the first, second, fourth and seventh sampling occasions (4$^{th}$ July 2000, 30$^{th}$ August 2000, 26$^{th}$ January 2001 and 2$^{nd}$ January 2002).

The numbers found for total meiofauna per 10 $cm^2$ and to a depth of 20 cm ranged from 45 to 4414. Nematodes ranged from 21 to 777 and harpacticoid copepods ranged from 0 to 1121 (Appendix 1). Twenty two out of 56 samples had mean numbers of Harpacticoid copepods below 10. Five of the S0 and S2 stations, four of the N1 stations, three of the N3 stations, two of the S1 and N2 stations and one of the S3 stations. Stations S0 and S2 had the most frequent occurrence of low harpacticoid numbers and Station S0 had the lowest numbers of nematodes and lowest total numbers on one occasion, this suggested that these two stations were most disturbed. Only Station S4 (which had greatest distance from the outfall) maintained relatively high total numbers and numbers of taxa throughout the study period. For the mean total number of animals (N), the data was $\log_{10}$ transformed to meet the statistical assumptions of normality in order to proceed with a one-way ANOVA. Figure 3.1(b) shows differences between stations with the untransformed data where one can see that Station S4 had the highest number of animals. The differences in the mean values for total numbers (N) were greater than would be expected by chance ($P<0.05$); the F statistic was 3.936 with P=0.002. The pair-wise multiple comparison procedures revealed that Station S4 was significantly different to four other stations, namely S2, S0, N3 and N1 with P values of 0.003, 0.006, 0.009 and 0.011 respectively. Here the highest numbers of animals were found at Station S4 and the lowest number at S2.

Figure 3.1(c) shows the differences in mean species richness (d) between stations and there appears to be reduced species richness at stations closer to the effluent outlet (S0, S1 and S2) compared to those further away but there was no statistically significant difference between the stations with an F statistic of 1.419 and P=0.220.

Figure 3.1(d) shows the differences in the mean evenness (J') between stations. There was a significant difference ($P<0.050$) between the mean values among the sampling stations which was greater than would be expected by chance with F = 2.483 and P=0.029. The pair-wise multiple comparison procedures, however, did not reveal any significant difference between any of the stations.

The differences in the mean index of diversity (H') is shown in Figure 3.1(e). There was no significant difference found between these means with F = 2.175 and P=0.530.

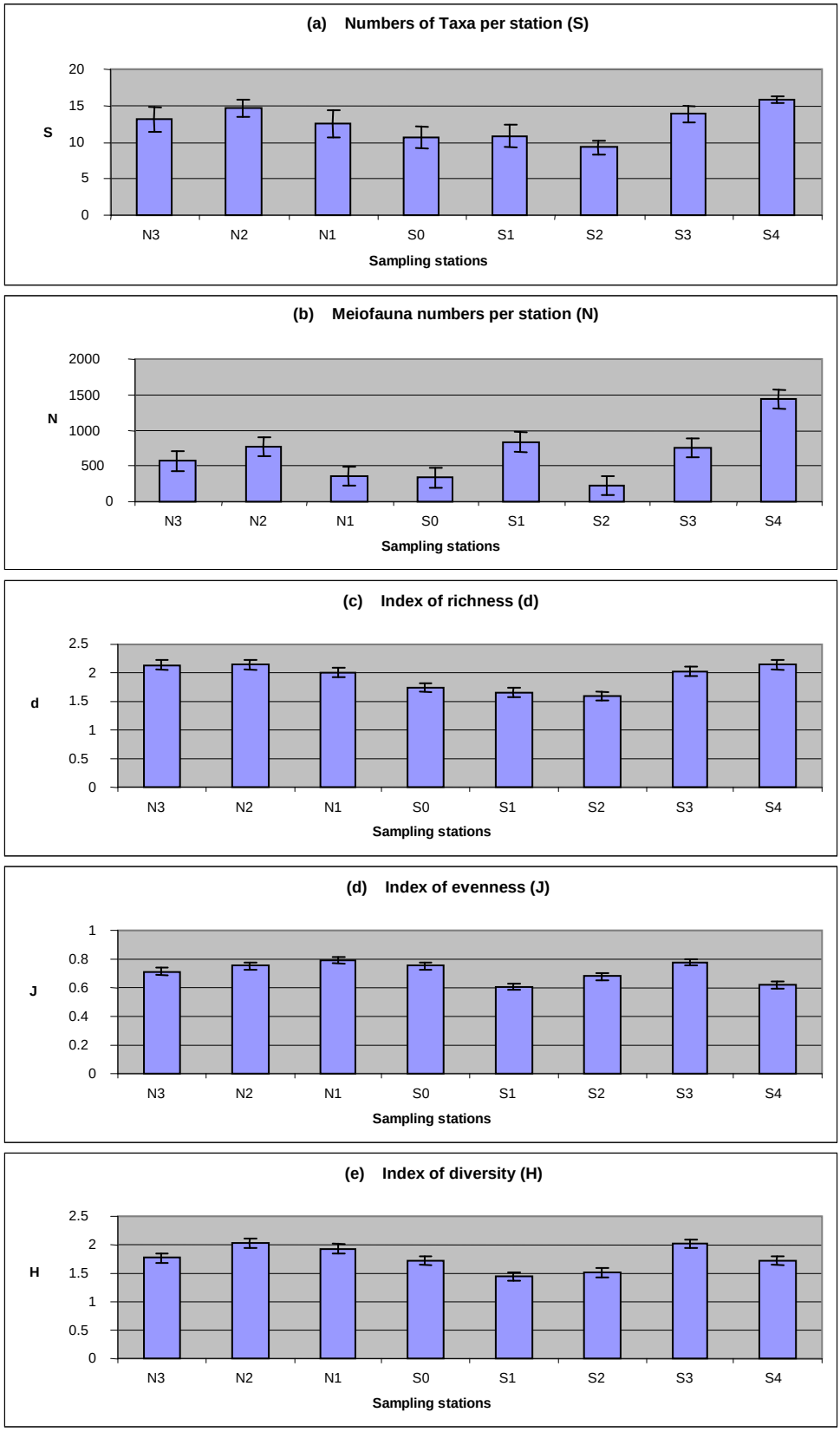


Figure 3.1 Univariate indices used to measure attributes of community structure in relation to sampling stations. (a) number of taxa, (b) meiofauna numbers, (c) index of richness (d) index of evenness and (e) index of diversity. Bars indicate the standard error of the mean.

The differences in the mean values for the number of taxa (S) per season are shown in Figure 3.2(a) and were greater than would be expected by chance ($P<0.05$) where the F statistic was 7.382 with P=0.001. From pair-wise multiple comparison procedures (Student-Newman-Keuls Method) there was a significant difference between autumn and winter (P=0.001), between autumn and summer (P=0.029) and between summer and winter (P=0.039).

For the mean total number of animals (N), the data needed to be $\log_{10}$ transformed to meet the statistical assumptions of normality in order to proceed with a one-way ANOVA. Figure 3.2(b) shows apparent differences between seasons with the untransformed data where one can see that the highest number of animals was recorded in autumn. However there were no significant differences found in the mean values for total numbers (N) where the F statistic was 2.481 with P=0.089.

Figure 3.2(c) shows the differences in the mean species richness (d) between the seasons. There was a statistically significant difference between the seasons with an F statistic of 7.700 and $P<0.001$. From pair-wise multiple comparison procedures (Student-Newman-Keuls Method) it was evident that there was a significant increase in autumn compared to summer (P=0.003). Autumn was also significantly higher than winter (P=0.001P) but there was no significant difference between summer and winter.

For the mean index of evenness (J) shown in Figure 3.2(d) the normality test failed and no transformation could be found that would meet the assumptions of normality, so the Kruskal-Wallis procedure was used to conduct an analysis of variance by ranks. No significant difference was found with P=0.948 and H=0.107.

For the mean diversity index (H) shown in Figure 3.2(e) there were no significant differences between seasons with P=0.184 and F=1.723.

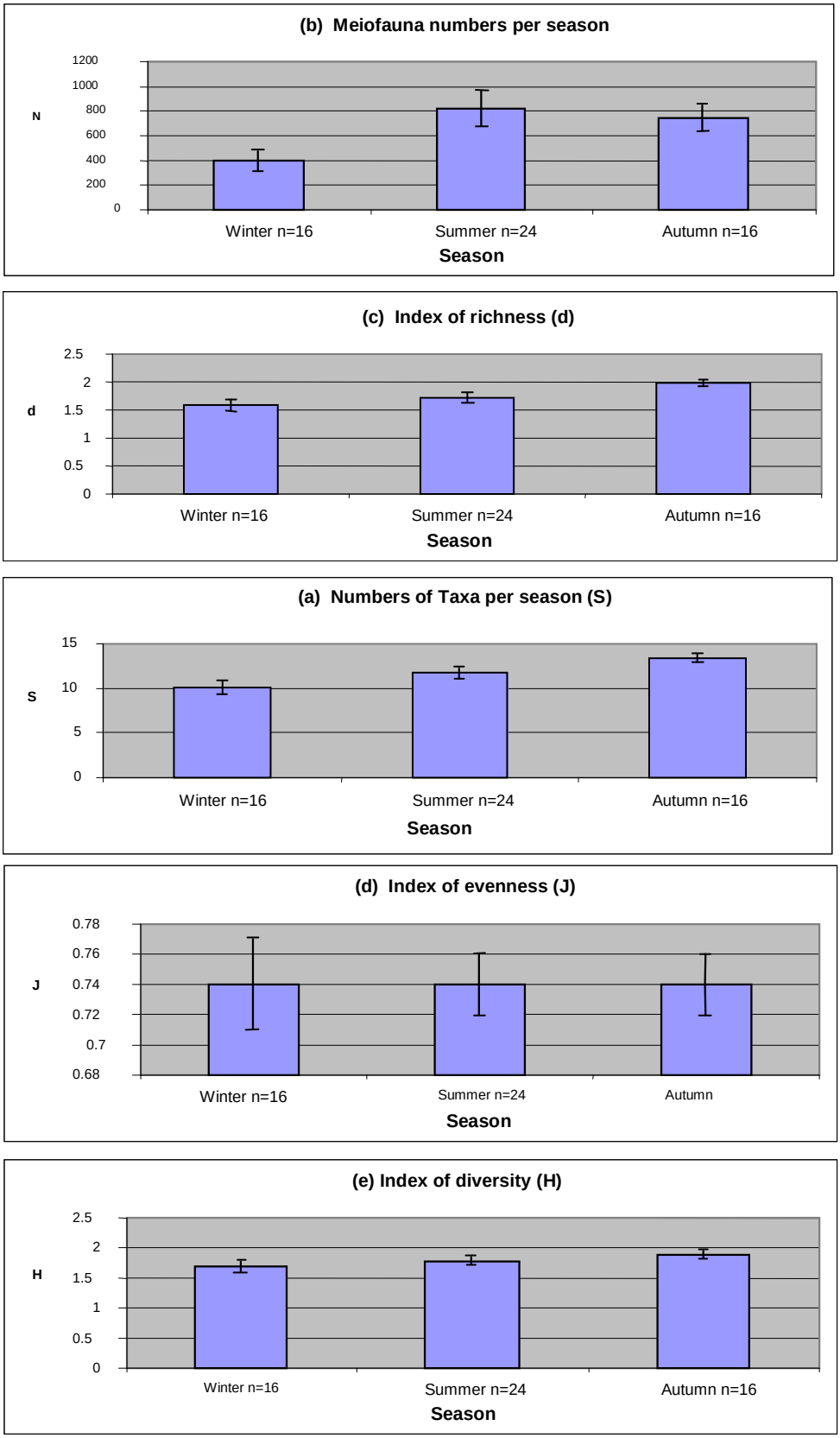


Figure 3.2 Univariate indices used to measure attributes of community structure in relationto season. (a) number of taxa, (b) meiofauna numbers, (c) index of richness, (d) index of evenness and (e) index of diversity. Bars indicate the standard error of the mean.

## 3.2 Analysis of similarity (ANOSIM)

An analysis of similarity (ANOSIM) was done for the last two sample data sets of the 9th April 2001 and 2nd January 2002 to determine whether the similarity between replicates within stations was significantly greater than similarities between replicates from different stations. These two sampling occasions were chosen because at least three replicates were taken for every station on these days. The null hypothesis (Ho) to be tested was that there were no differences in community composition between the different sampling stations and different sampling times. The replicates from each of the 8 stations were grouped and both sampling occasions were analysed together making a comparison of 16 groups (i.e. 8 stations x 2 sampling times). The groups were analysed using all the taxa identified as well as using only the major taxa to determine the differences if any of analyzing only to the major taxon level. The global R statistic value for all taxa identified was found to be 0.948 with an associated significance level of $p < 0.1$ % ($p < 0.001$). That means that the number of permuted statistics greater than or equal to global R was zero. The global R statistic value for animals identified to higher taxonomic levels was 0.914 with an associated significance level of $p < 0.1$ % ($p < 0.001$) and therefore also meaning no permuted statistics were greater than or equal to the global R statistic. The above results lead to the rejection of the null hypothesis and demonstrate that there is a significantly greater similarity between replicates within a sampling station than between replicates from different sampling stations and that there is very little difference in the result obtained whether the meiofauna is identified to major taxonomic groups or to lower taxa.

## 3.3 Cluster, MDS ordination and SIMPER analysis of the replicated data sets for the 9th April 2001 and 2nd January 2002

Cluster and MDS analyses were applied to the meiobenthic community data for the last two sample sets when at least three replicates were taken (see section 3.2). These plots investigated whether replicates within sampling stations tend to group together with a greater similarity than the replicates from different stations, and further, that any spatial and temporal trends may be related to effluent discharge as effluent was not being pumped to sea on the 9th April 2001 whereas it was on the 2nd January 2002. Cluster and MDS analyses explore the degree of similarity between stations as reflected in their meiofaunal community structure. Cluster analysis results in a dendrogram, which groups the stations hierarchically according to their Bray-Curtis measure of similarity. MDS uses a two-dimensional scatter plot to depict relative similarities between stations.

A cluster plot for all taxa identified to the lower taxonomic rank, as well as the respective MDS ordination plot were generated. Samples were labeled such that the symbol vi indicated the sixth sample set (9/04/2001) and the symbol vii indicated the seventh sample set (2/01/2002). The letters a, b, c and d after the sample site name indicated which replicate it was. From the dendrogram in Figure 3.3 replicates within a station (shown by small brackets) tended to group together with greater similarity to one another than with replicates from different stations. The dendrogram in Figure 3.3 also divided the data set into two distinct groups shown as Group 1 and Group 2 indicated by the large brackets. Group 2 divided further into Groups 2A and 2B. The corresponding MDS for the dendrogram in Figure 3.3 is shown in Figure 3.4. The Group 2A samples are coloured red and the Group 2B samples are coloured green to

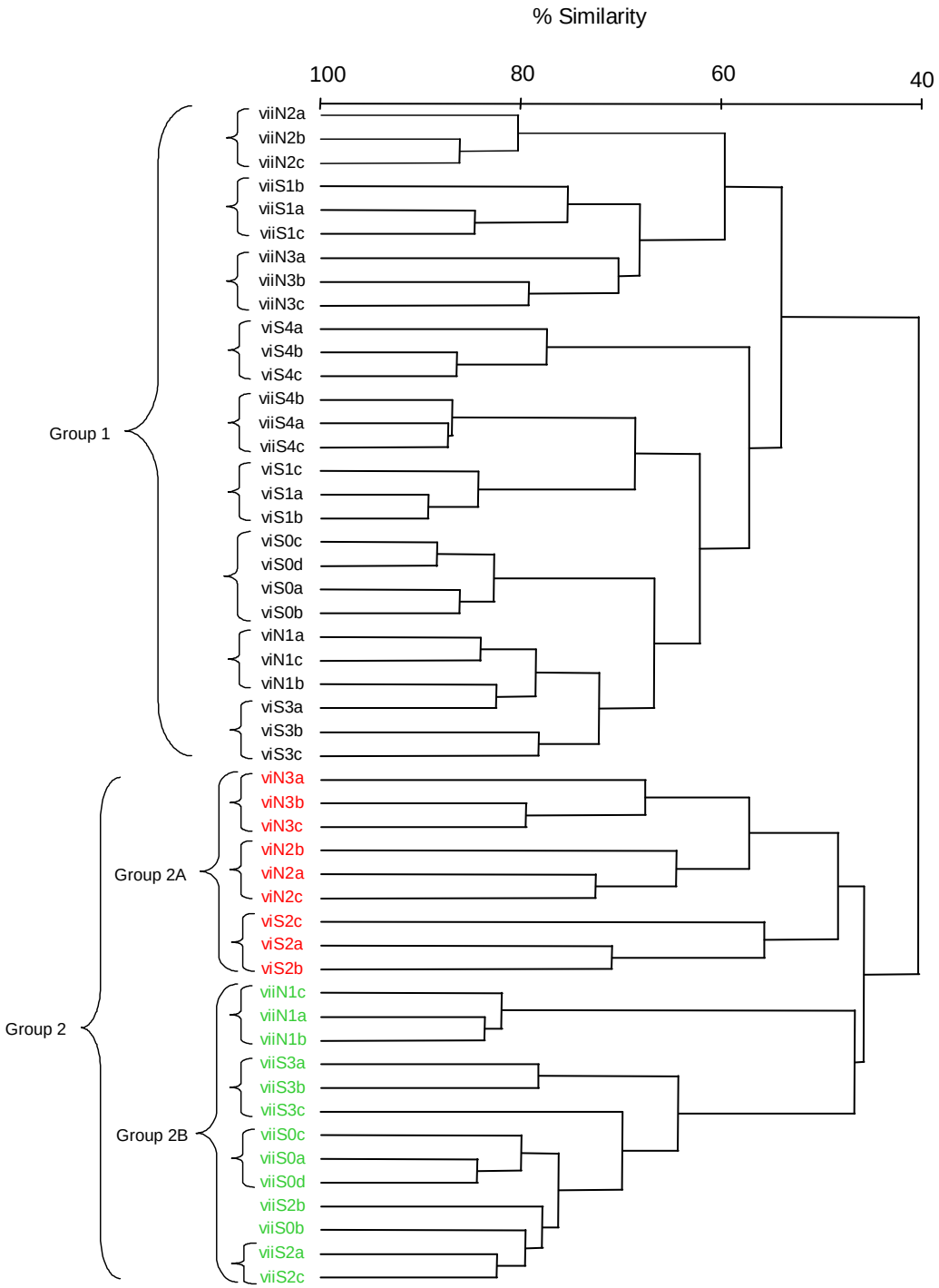


Figure 3.3. Dendrogram showing hierarchical relationships between meiofauna samples identified to the lowest taxonomic rank (Table 2.2) taken at Sezela beach in April 2001 (vi) and January 2002 (vii). Similarity was computed using the Bray-Curtis coefficient and square root transformations.

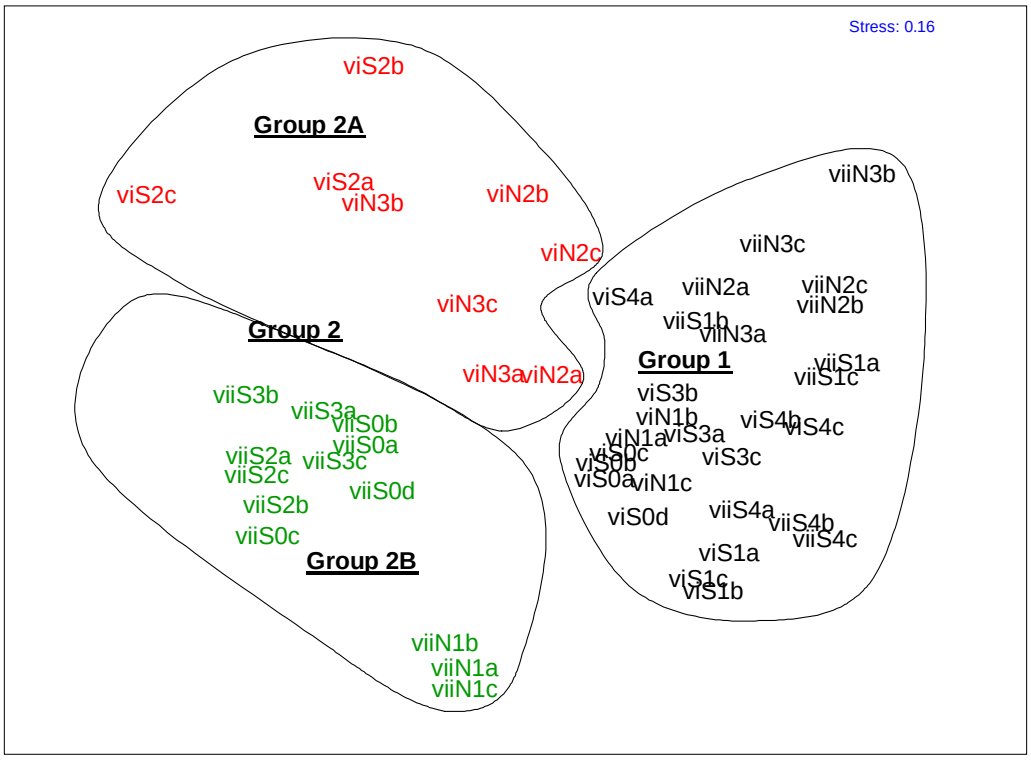


Figure 3.4. MDS plot of meiofauna samples identified to the lowest taxonomic rank (Table 2.3) for the last two sample sets taken at Sezela beach in April 2001 (vi) and January 2002 (vii). The circled groups correspond with the groups in Figure 3.3.

emphasise the distinction between Groups 1, 2A and 2B. The stress value was 0.16 which is lower than 0.2 (see section 2.5.3) and this analysis therefore in Figure 3.4 offers a potentially useful picture of the relationships between samples.

SIMPER analyses were carried out for these dendrograms to determine which taxa were primarily responsible for influencing the sample groupings revealed by the cluster analysis. There were two main groups in Figure 3.3 shown as Group 1 and Group 2. Table 3.1 shows results which compare Groups 1 and 2 (dissimilarity of 59%) and it is evident that Group 1 had a much higher abundance of all taxa except for the Sarcomastigophora which had a higher average abundance in Group 2. Table 3.2 shows the taxa most responsible for the similarity (60%) within Group 1. A number of crustacean groups played an important role in the similarity of the samples in Group 1 namely copepod nauplii, ostracods and the family Paramesochridae. Table 3.3 shows the taxa most responsible for the similarity within Group 2. The main contributors to their similarity (52%), was the greater relative abundance of the nematode feeding groups and the turbellarians. It was concluded that the lower diversity of crustacean groups and the overall lower average abundance observed in the Group 2 samples resulted in the separation of the two groups.

Group 2 was further subdivided into Groups 2A and 2B (Figure 3.3). Table 3.4 compares the Groups 2A and 2B (dissimilarity of 54%) and it was evident that the main contributions to their dissimilarity was the higher abundances of sarcomastigophorans in Group 2B compared to zero abundance in Group 2A, which had much higher abundances of all the crustacean groups such as ostracods, copepod nauplii, Cylindropsyllidae, Tisbidae and Paramesochridae than Group 2B. The taxa that contributed to the similarity (55%) of the samples in Group 2A

are shown in Table 3.5 where the higher abundances of 1A and 2B nematodes and ostracods and low abundances of turbellarians contributed largely to their similarity. Table 3.6 shows the taxa most responsible for the similarity (62%) of the Group 2B samples which had higher relative abundances of 1B, 2A and 1A nematodes, the turbellarians and sarcomastigophorans.

Table 3.1
SIMPER analysis of Groups 1 and 2 depicted in Figure 3.3 (meiofauna identified to lowest taxonomic rank). Average dissimilarity = 59.72%

| Taxa | Group 1 Av.Abund. | Group 2 Av.Abund. | Av.Diss. | Diss./SD | Contrib.% | Cum.% |
|---|---|---|---|---|---|---|
| Copepod nauplii | 320 | 10.3 | 7.75 | 1.39 | 13 | 13 |
| Paramesochridae | 145 | 2.68 | 5.73 | 1.39 | 9.6 | 22.6 |
| Saccocirridae | 231 | 19.9 | 5.7 | 0.93 | 9.55 | 32.1 |
| Ostracoda | 150 | 17.7 | 5.1 | 1.15 | 8.53 | 40.7 |
| Cylindropsyllidae | 135 | 3.95 | 4.71 | 1.14 | 7.89 | 48.6 |
| Gastrotricha | 68.6 | 0.95 | 3.84 | 1.51 | 6.43 | 55 |
| Nematoda 1B | 113 | 33.9 | 3.28 | 1.26 | 5.49 | 60.5 |
| Nematoda 1A | 98.3 | 34.3 | 2.85 | 1.31 | 4.77 | 65.2 |
| Oligochaeta | 50.8 | 0.77 | 2.43 | 0.84 | 4.08 | 69.3 |
| Nematoda 2A | 53.7 | 23 | 2.38 | 1.09 | 3.98 | 73.3 |
| Sarcomastigophora | 16.4 | 24.9 | 2.27 | 0.9 | 3.81 | 77.1 |
| Nematoda 2B | 43.4 | 15.8 | 2.13 | 1.25 | 3.57 | 80.7 |
| Acarina | 22.9 | 9.59 | 2.13 | 1.74 | 3.56 | 84.2 |
| Turbellaria | 34 | 24.3 | 1.98 | 1.45 | 3.31 | 87.6 |
| Tisbidae | 19.7 | 3.95 | 1.95 | 1.15 | 3.27 | 90.8 |

Table 3.2
SIMPER analysis showing the taxa most responsible for the similarity of the samples depicted as Group 1 of the dendrogram in Figure 3.3 (meiofauna identified to lowest taxonomic rank). Average similarity = 60.04%

| Taxa | Av. Abund. | Av. Sim. | Sim./SD | Contrib.% | Cum.% |
|---|---|---|---|---|---|
| Copepod nauplii | 319.79 | 8.22 | 3.19 | 13.69 | 13.7 |
| Nematoda 1B | 112.89 | 6.61 | 2.62 | 11.01 | 24.7 |
| Nematoda 1A | 98.25 | 6.45 | 3.18 | 10.75 | 35.5 |
| Ostracoda | 150.21 | 6.05 | 2 | 10.07 | 45.5 |
| Paramesochridae | 144.64 | 5.28 | 1.5 | 8.79 | 54.3 |
| Saccocirridae | 230.61 | 3.6 | 0.89 | 6 | 60.3 |
| Nematoda 2B | 43.36 | 3.58 | 2.12 | 5.97 | 66.3 |
| Nematoda 2A | 53.68 | 3.57 | 1.94 | 5.94 | 72.2 |
| Turbellaria | 33.96 | 3.46 | 2.61 | 5.77 | 78 |
| Cylindropsyllidae | 135.07 | 3.28 | 0.93 | 5.47 | 83.5 |
| Gastrotricha | 68.61 | 3.2 | 1.26 | 5.34 | 88.8 |
| Acarina | 22.86 | 3.04 | 2.61 | 5.06 | 93.9 |

Table 3.3
SIMPER analysis showing the taxa most responsible for the similarity of the samples depicted as Group 2 of the dendrogram in Figure 3.3 (meiofauna identified to lowest taxonomic rank). Average similarity = 52.87%

| Taxa | Av. Abund. | Av. Sim. | Sim./SD | Contrib.% | Cum.% |
|---|---|---|---|---|---|
| Nematoda 1A | 34.3 | 10.5 | 2.98 | 19.9 | 19.9 |
| Nematoda 1B | 33.9 | 9.83 | 2.14 | 18.6 | 38.5 |
| Nematoda 2A | 23 | 6.16 | 1.35 | 12.8 | 51.3 |
| Turbellaria | 24.3 | 6.16 | 1.22 | 11.7 | 63 |
| Nematoda 2B | 15.8 | 5.88 | 1.5 | 11.1 | 74.1 |
| Ostracoda | 17.7 | 3.58 | 0.93 | 6.78 | 80.9 |
| Sarcomastigophora | 24.9 | 2.33 | 0.5 | 4.41 | 85.3 |
| Acarina | 9.59 | 1.85 | 0.66 | 3.51 | 88.8 |
| Copepod nauplii | 10.3 | 1.61 | 0.63 | 3.04 | 91.8 |

Table 3.4
SIMPER analysis of Groups 2A and 2B depicted in Figure 3.3 (meiofauna identified to lowest taxonomic rank). Average dissimilarity = 54.24%

| Taxa | Group 2A Av. Abund. | Group 2B Av. Abund. | Av.Diss. | Diss./SD | Contrib.% | Cum.% |
|---|---|---|---|---|---|---|
| Sarcomastigophora | 0 | 42.2 | 6.43 | 1.36 | 11.9 | 11.9 |
| Ostracoda | 37.2 | 4.15 | 5.03 | 1.72 | 9.27 | 21.1 |
| Nematoda 2A | 9 | 32.7 | 4.68 | 1.55 | 8.63 | 29.8 |
| Turbellaria | 7 | 36.3 | 4.53 | 1.96 | 8.35 | 38.1 |
| Saccocirridae | 4.11 | 30.9 | 4.06 | 0.84 | 7.49 | 45.6 |
| Nematoda 1B | 25.7 | 39.6 | 3.94 | 1.17 | 7.27 | 52.9 |
| Copepod nauplii | 22 | 2.23 | 3.5 | 1.23 | 6.45 | 59.3 |
| Nematoda 1A | 40.3 | 30.2 | 3.12 | 1.5 | 5.75 | 65.1 |
| Nematoda 2B | 24.8 | 9.54 | 3.04 | 1.14 | 5.6 | 70.7 |
| Acarina | 2 | 14.9 | 2.99 | 1.02 | 5.51 | 76.2 |
| Cylindropsyllidae | 8.44 | 0.85 | 2.5 | 0.99 | 4.61 | 80.8 |
| Tisbidae | 9 | 0.38 | 2.37 | 1.09 | 4.38 | 85.2 |
| Paramesochridae | 5.11 | 1 | 1.69 | 0.94 | 3.11 | 88.3 |
| Gastrotricha | 2 | 0.23 | 1.16 | 0.87 | 2.14 | 90.4 |

Table 3.5
SIMPER analysis showing the taxa most responsible for the similarity of the samples depicted as Group 2A of the dendrogram in Figure 3.3 (meiofauna identified to lowest taxonomic rank). Average similarity = 55.05%

| Taxa | Av. Abund. | Av. Sim. | Sim./SD | Contrib.% | Cum. % |
|---|---|---|---|---|---|
| Nematoda 1A | 40.3 | 10 | 2.62 | 18.2 | 18.2 |
| Ostracoda | 37.2 | 8.96 | 2.2 | 16.3 | 34.5 |
| Nematoda 2B | 24.8 | 8.79 | 2.03 | 16 | 50.4 |
| Turbellaria | 7 | 6.03 | 2.69 | 11 | 61.4 |
| Nematoda 1B | 25.7 | 5.69 | 1.5 | 10.3 | 71.7 |
| Copepod nauplii | 22 | 3.16 | 0.79 | 5.75 | 77.5 |
| Cylindropsyllidae | 8.44 | 2.9 | 1.06 | 5.26 | 82.8 |
| Nematoda 2A | 9 | 2.21 | 0.79 | 4.01 | 86.8 |
| Tisbidae | 9 | 1.75 | 0.59 | 3.19 | 90 |
| Saccocirridae | 4.11 | 1.61 | 0.82 | 2.92 | 92.9 |

Table 3.6
SIMPER analysis showing the taxa most responsible for the similarity of the samples depicted as Group 2B of the dendrogram in Figure 3.3 (meiofauna identified to lowest taxonomic rank). Average similarity = 62.53%

| Taxa | Av. Abund. | Av. Sim. | Sim./SD | Contrib.% | Cum. % |
|---|---|---|---|---|---|
| Nematoda 1B | 39.6 | 13.4 | 5.13 | 21.4 | 21.4 |
| Nematoda 2A | 32.7 | 11.7 | 4.25 | 18.7 | 40.4 |
| Nematoda 1A | 30.2 | 10.8 | 3.43 | 17.3 | 57.4 |
| Turbellaria | 36.3 | 7.62 | 1.08 | 12.2 | 69.6 |
| Sarcomastigophora | 42.2 | 6.9 | 1.19 | 11 | 80.6 |
| Nematoda 2B | 9.54 | 4.24 | 1.33 | 6.78 | 87.4 |
| Acarina | 14.9 | 2.52 | 0.68 | 4.02 | 91.4 |

A CLUSTER plot for taxa identified to the major taxonomic rank as well as the respective MDS ordination were also generated in a similar way to that above. From the dendrogram in Figure 3.5 replicates within a station (shown by small brackets) tended to group together with greater similarity to one another than with replicates from different stations. The dendrogram in Figure 3.5 also divided the data set into two distinct groups shown as Group 1 and Group 2 by the large brackets. Group 1 was further divided into Groups 1A and 1B. The corresponding MDS for the dendrogram in Figure 3.5 is shown in Figure 3.6. The Group 2 stations are coloured green to emphasise the distinction between Groups 1 and 2. The Group 1B samples are coloured red to show their distinction from the Group 1A samples. The stress value was 0.12 which is lower than 0.2 (see section 2.5.3) and therefore offers a potentially useful picture of the relationships between samples.

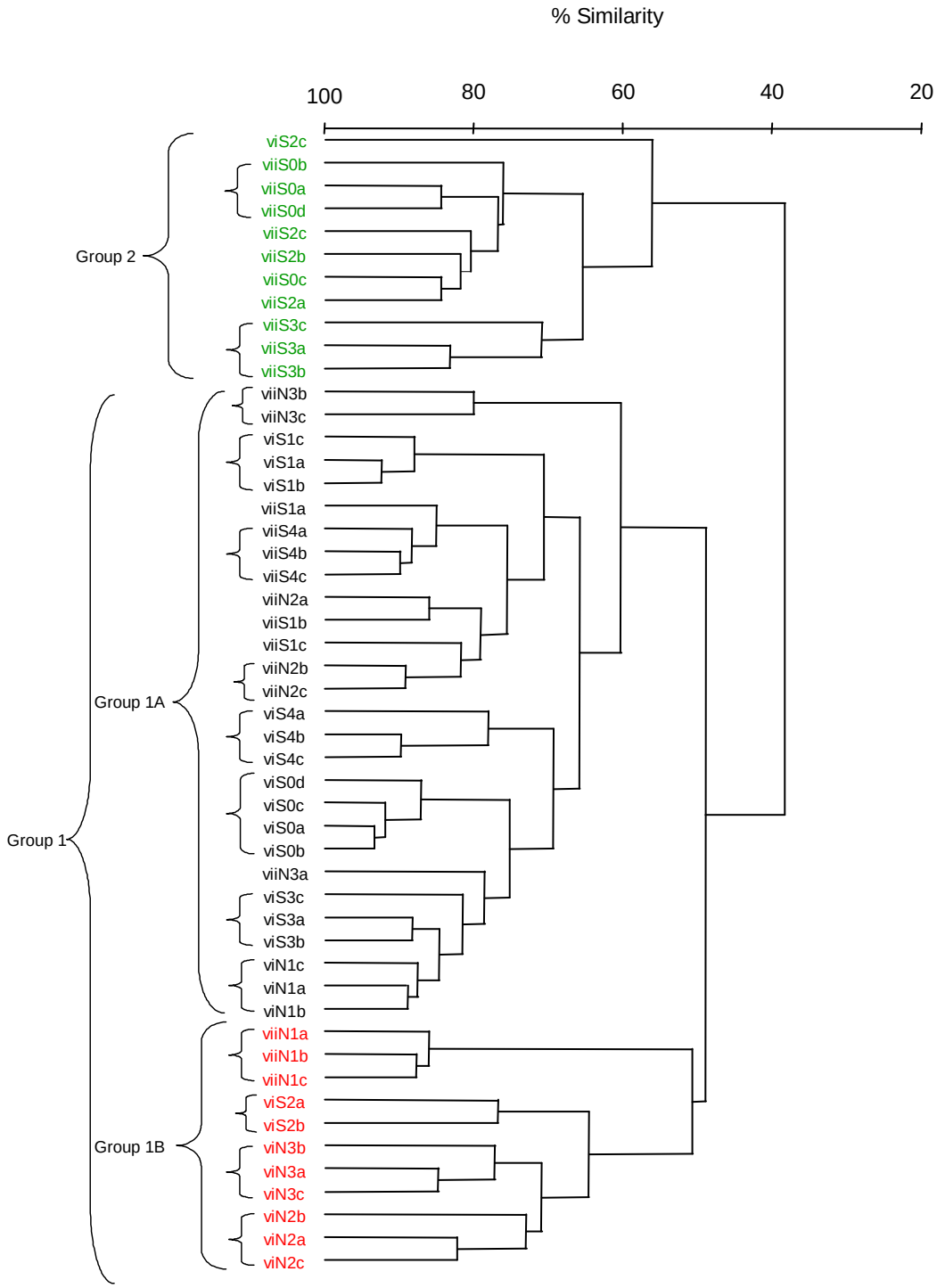


Figure 3.5. Dendrogram showing hierarchical relationships between meiofauna samples taken at Sezela beach in April 2001 (vi) and January 2002 (vii) and identified to the major taxonomic rank (Table 2.3). Similarity was computed using the Bray-Curtis coefficient and Square root transformations.

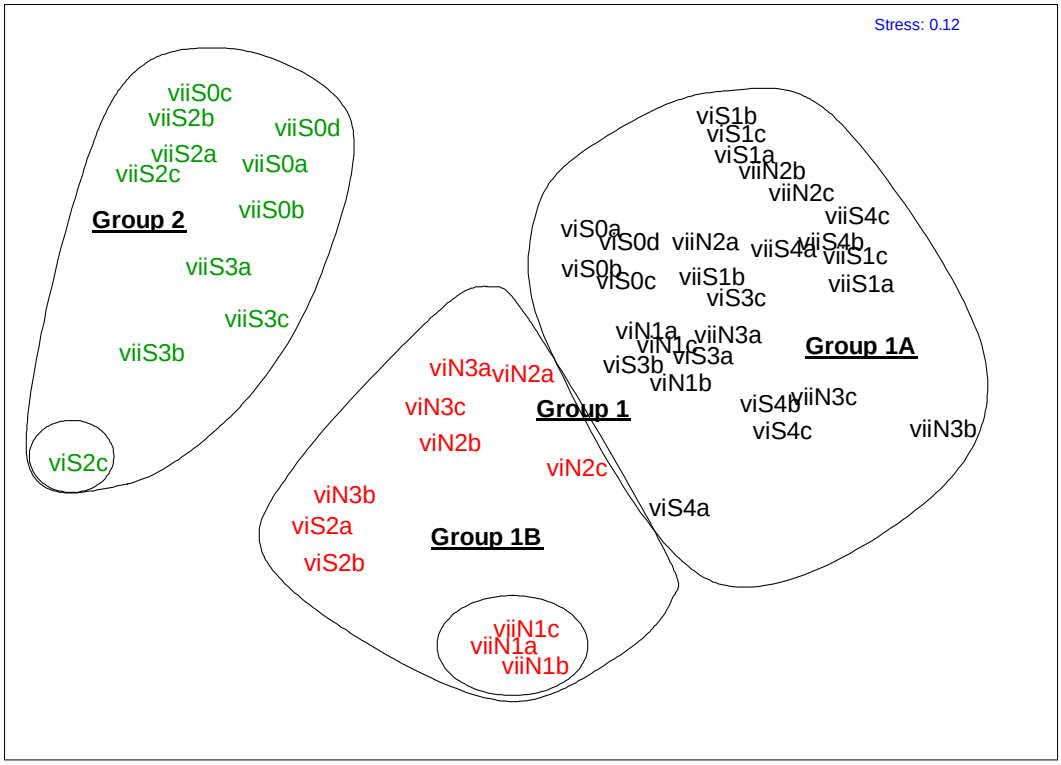


Figure 3.6. MDS plot of meiofauna samples for the last two sample sets taken in April 2001 (vi) and January 2002 (vii) identified to major taxonomic rank (Table 2.3). The circled groups correspond with the groups Figure 3.5.

SIMPER analyses were done for these dendrograms to determine which taxa were primarily responsible for influencing the sample groupings revealed by the cluster analysis. Table 3.7 compares Groups 1 and 2 shown in Figure 3.5 (dissimilarity of 61 %) and it is evident that Group 1 had a much higher abundance of all taxa except for the sarcomastigophorans and turbellarians which had higher average abundances in Group 2. Table 3.8 shows the taxa most responsible for the similarity (60%) within Group 1. A number of crustacean groups play an important role in the similarity of the samples in Group 1 namely harpacticoids, copepod nauplii and ostracods. Table 3.9 shows the taxa most responsible for the similarity within Group 2. The main contributors to Group 2 similarity (69%), was the greater relative abundance of the nematodes, turbellarians and sarcomastigophorans. It was concluded that the lower diversity of crustacean groups and the overall lower average abundance observed in the Group 2 samples resulted in the separation of the two groups.

Group 1 was further subdivided into Groups 1A and 1B (Figure 3.5). Table 3.10 compares the Groups 1A and 1B (dissimilarity of 51%) and it was evident that the main contributions to their dissimilarity was the higher abundances of harpacticoids, copepod nauplii, annelids, nematodes, ostracods, gastrotrichs, turbellarians and acarinas in Group 1A. The taxa most responsible for the similarity of the samples in Group 1A are shown in Table 3.11. High abundances of nematodes, harpacticoids, copepod nauplii and annelids contributed the most to this group. Table 3.12 shows the taxa contributing the most to the similarity (62.4%) of the Group 1B samples in Figure 3.5 which were the nematodes, ostracods, annelids and harpacticoids but in lower abundances relative to the Group 1 samples.

Table 3.7
SIMPER analysis of Groups 1 and 2 depicted in Figure 3.5 (meiofauna anaysed to the major taxonomic rank). Average dissimilarity = 61.74%

| Taxa | Group 1 Av.Abund. | Group 2 Av.Abund. | Av.Diss. | Diss./SD | Contrib.% | Cum.% |
|---|---|---|---|---|---|---|
| Annelida | 246 | 1.18 | 10.2 | 1.4 | 16.5 | 16.5 |
| Harpacticoida | 226 | 2.36 | 10.1 | 1.87 | 16.4 | 33 |
| Copepod nauplii | 235 | 0.82 | 9.51 | 1.43 | 15.4 | 48.4 |
| Ostracoda | 117 | 2 | 7.7 | 1.5 | 12.5 | 60.8 |
| Sarcomastigophora | 12.2 | 48.4 | 5.62 | 1.25 | 9.1 | 69.9 |
| Nematoda | 246 | 118 | 5.46 | 1.29 | 8.84 | 78.8 |
| Gastrotricha | 49.7 | 0.27 | 4.15 | 1.18 | 6.72 | 85.5 |
| Turbellaria | 25.6 | 44.2 | 3.61 | 1.12 | 5.84 | 91.3 |

Table 3.8
SIMPER analysis showing the taxa responsible for the similarity of the samples depicted as Group 1 of the dendrogram in Figure 3.5 (meiofauna analysed to the major taxonomic rank). Average similarity = 60.3%

| Taxa | Av. Abund. | Av. Sim. | Sim./SD | Contrib.% | Cum. % |
|---|---|---|---|---|---|
| Nematoda | 245 | 15.5 | 3.15 | 25.8 | 25.8 |
| Harpacticoida | 226 | 9.77 | 1.73 | 16.2 | 42 |
| Annelida | 245 | 8.55 | 1.76 | 14.2 | 56.2 |
| Copepod nauplii | 235 | 8.05 | 1.8 | 13.4 | 69.5 |
| Ostracoda | 117 | 8.01 | 2.09 | 13.3 | 82.8 |
| Turbellaria | 25.6 | 3.74 | 1.7 | 6.21 | 89 |
| Acarina | 21 | 3.65 | 1.42 | 6.05 | 95 |

Table 3.9
SIMPER analysis showing the taxa most responsible for the similarity of the samples depicted as Group 2 of the dendrogram in Figure 3.4 (meiofauna analysed to the major taxonomic rank). Average similarity = 69.74%

| Taxa | Av. Abund. | Av. Sim. | Sim./SD | Contrib.% | Cum.% |
|---|---|---|---|---|---|
| Nematoda | 118 | 35 | 6.28 | 50.6 | 50.6 |
| Turbellaria | 44.2 | 17.6 | 4.43 | 25.5 | 76 |
| Sarcomastigophora | 48.4 | 10.2 | 1.19 | 14.8 | 90.8 |

Table 3.10
SIMPER analysis of Groups 1A and 1B depicted in Figure 3.5 (meiofauna analysed to the major taxonomic rank). Average dissimilarity = 51.11%

| Taxa | Group 1A Av Abund. | Group 1B Av. Abund. | Av. Diss. | Diss./SD | Contrib. % | Cum. % |
|---|---|---|---|---|---|---|
| Harpacticoida | 307 | 19.6 | 9.63 | 2 | 18.9 | 18.9 |
| Copepod nauplii | 319 | 19.8 | 8.58 | 1.26 | 16.8 | 35.6 |
| Annelida | 324 | 43.9 | 8.2 | 1.19 | 16 | 51.7 |
| Nematoda | 304 | 95.8 | 6.5 | 1.39 | 12.7 | 64.4 |
| Ostracoda | 150 | 33.4 | 4.96 | 1.02 | 9.7 | 74.1 |
| Gastrotricha | 68.6 | 1.64 | 4.69 | 1.5 | 9.18 | 83.3 |
| Turbellaria | 34 | 4.45 | 2.93 | 1.69 | 5.73 | 89 |
| Acarina | 22.9 | 16.2 | 2.61 | 1.82 | 5.11 | 94.1 |

Table 3.11
SIMPER analysis showing the taxa responsible for the similarity of the samples depicted as Group 1A of the dendrogram in Figure 3.5 (meiofauna analysed to the major taxonomic rank). Average similarity = 69.29%

| Taxa | Av.Abund. | Av. Sim. | Sim./SD | Contrib.% | Cum. % |
|---|---|---|---|---|---|
| Nematoda | 304 | 15.2 | 3.46 | 21.9 | 21.9 |
| Harpacticoida | 307 | 13.4 | 3.39 | 19.3 | 41.2 |
| Copepod nauplii | 319 | 10.5 | 3.34 | 15.2 | 56.4 |
| Annelida | 324 | 9.6 | 2.07 | 13.9 | 70.2 |
| Ostracoda | 150 | 7.7 | 2.11 | 11.1 | 81.3 |
| Turbellaria | 34 | 4.47 | 2.5 | 6.46 | 87.8 |
| Gastrotricha | 68.6 | 4.1 | 1.26 | 5.92 | 93.7 |

Table 3.12
SIMPER analysis showing the taxa responsible for the similarity of the samples depicted as Group 1B of the dendrogram in Figure 3.5 (meiofauna analysed to the major taxonomic rank). Average similarity = 62.4%

| Taxa | Av. Abund. | Av. Sim. | Sim./SD | Contrib. % | Cum. % |
|---|---|---|---|---|---|
| Nematoda | 95.8 | 22.3 | 5.43 | 35.7 | 35.7 |
| Ostracoda | 33.4 | 11.4 | 2.5 | 18.2 | 53.9 |
| Annelida | 43.9 | 8.64 | 1.56 | 13.9 | 67.8 |
| Harpacticoida | 19.6 | 6.43 | 1.07 | 10.3 | 78.1 |
| Copepod nauplii | 19.8 | 5.61 | 1.21 | 9 | 87.1 |
| Acarina | 16.2 | 3.65 | 0.75 | 5.9 | 92.9 |

When comparing the analysis using taxa identified to the lower rank with that of identification to the major taxonomic rank it was evident that all the samples that grouped together as Group 1 in Figure 3.3 (lower taxonomic rank) also group together as Group 1A in Figure 3.5 (major taxonomic rank). However, the samples that made up Group 2A in Figure 3.3 and grouped as Group 2 in that figure move to Group 1 in Figure 3.5. This occurs when combining the nematode feeding groups, the annelid families and the harpacticoid copepod families as single groups. The samples in Group 2B in Figure 3.3 remain virtually the same similarity to the Group 1 samples from both Figures 3.3 and 3.5. This appears to be due to the low numbers of annelids and harpacticoids in this group so that on combining their families would have little effect on their similarity to the Group 1 samples in Figure 3.3. The only exceptions were the replicate viS2c and the replicates viiN1a, viiN1b and viiN1c which changed groups and are indicated by circles in the MDS plot in Figure 3.6. The viiN1a, b and c replicates had relatively higher numbers of annelid and harpacticoid families than the viS2c replicate causing their movement into the other groups when these families are combined into higher taxonomic ranks (see Appendix Tables A1.7 and A1.6).

When comparing the MDS plots in Figures 3.4 and 3.6 the relationship between the samples that are circled as Group 1 (Figure 3.4) and Group 1A (Figure 3.6) and the rest of the samples does not appear to be extremely different between the two plots suggesting little difference in analysing samples to lower taxonomic ranks as apposed to the major taxonomic rank.

## 3.4 Analysis of all data sets for seven sampling times

Having established that replicates within sampling stations were significantly more similar to one another than replicates between different stations from the analysis of similarity (ANOSIM, section 3.2), Bray-Curtis similarity plots and MDS ordinations were generated for all the sampling occasions using mean numbers of animals per sampling site for each sampling time. The fact that replicates within sampling stations were significantly more similar to one another than replicates between different stations from the analysis of similarity for the last two sample data sets does not imply that it is true for all the sampling occasions, but this could not be tested as replicates for all the stations from the other data sets were not taken. The dendrogram showing station similarities for the study area, for all sampling periods where animals were identified to the lowest taxonomic rank is depicted in Figure 3.7. Two main groups of samples are derived. Group 1 contained all the S4 samples (greatest distance from the effluent discharge) and Group 2 contained all the S0 samples (station closest to the effluent discharge). The next two stations closest to the effluent discharge, namely N1 and S1 also occurred mostly in Group 2. Station N1 (approx. 150 m north of the discharge) occurred once in Group 1 on the sixth sampling period when effluent was not being pumped to sea. Station S1 (approx. 200 m south of the discharge) occurred twice in Group 1 of which one occurrence was during the sixth sampling period when effluent was not being pumped to sea. Station S2 was within 400m of the outfall and in a bay were effluent was observed being pushed ashore by wave action on the first, second, fourth and seventh sampling occasions (4$^{th}$ July 2000, 30$^{th}$ August 2000, 26$^{th}$ January 2001 and 2$^{nd}$ January 2002). This station occurred in Group 2 only. Station N2 (approx.300 m north of the discharge occurred five times in Group 1 and only twice in Group 2 as did Station S3 about 750 m south of the discharge. Station N3, about 600 m north of the discharge occurred four times in Group 1 and three times in Group 2.

The Multidimensional scaling (MDS) ordination relating to the Bray-Curtis similarity plot shown in Figure 3.7 is shown in Figure 3.8. The stations coloured red and underlined correspond with the Group 2 stations in Figure 3.7. The groups are not very distinct from each other when looking at the ordination in Figure 3.8 alone. However it did have a relatively high stress value of 0.18.

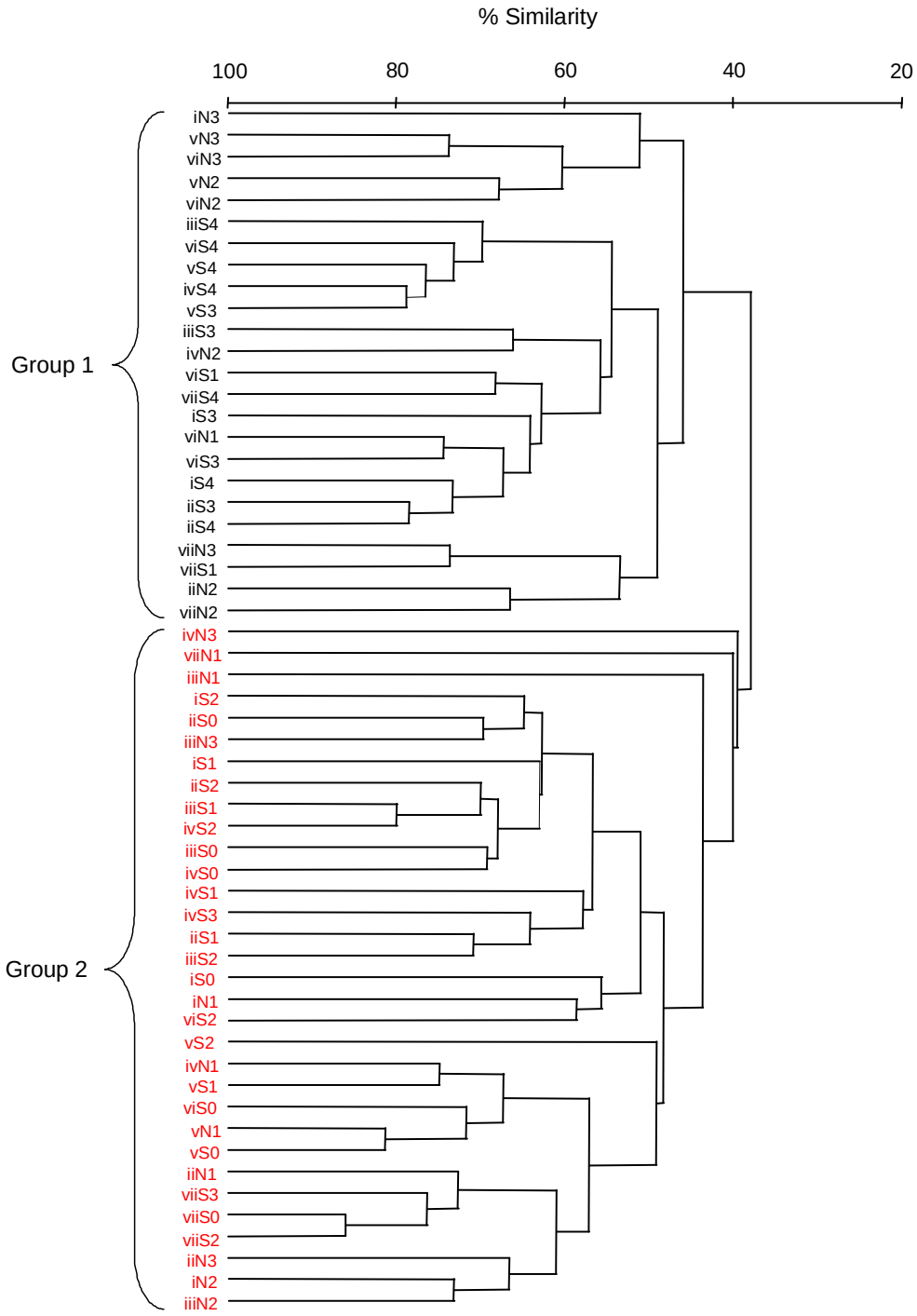


Figure 3.7. Dendrogram showing hierarchical relationships between meiofauna samples identified to the lowest taxonomic rank (Table 2.2) taken at Sezela beach. Similarity was computed using the Bray-Curtis coefficient and square root transformations. The Roman numerals i to vii indicate the sample date of which there were seven (i to vii).

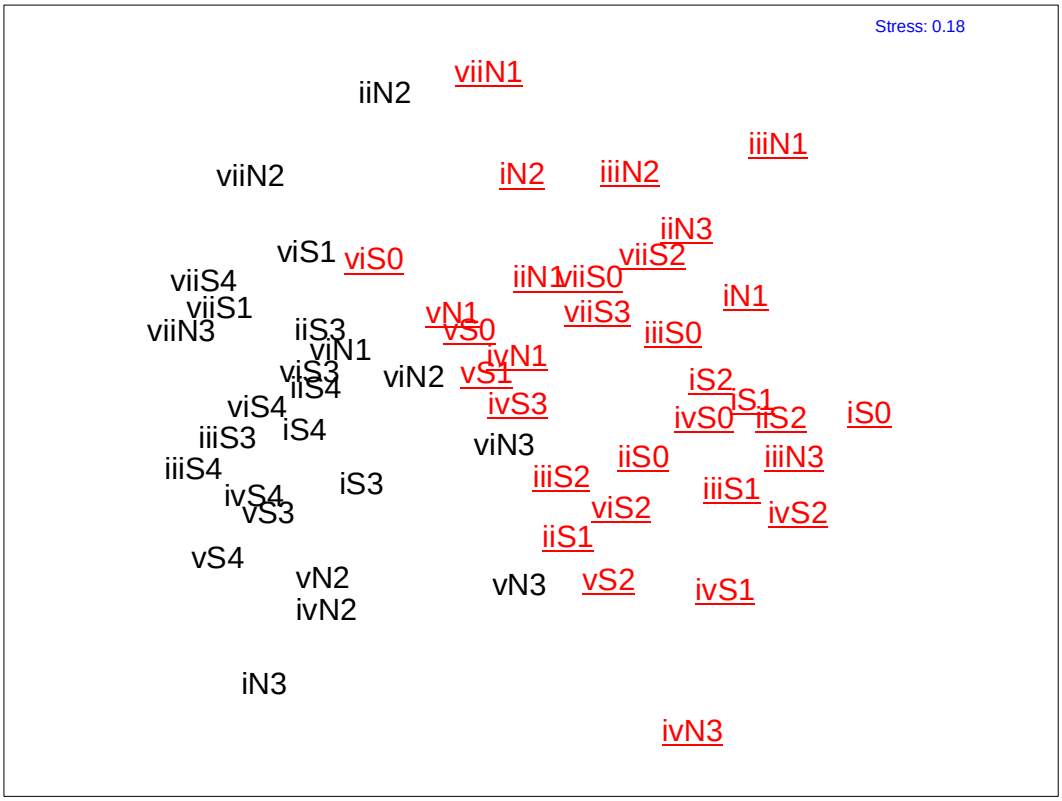


Figure 3.8. MDS plot of all meiofauna samples identified to the lowest taxonomic rank (Table 2.2). Stations in red and underlined correspond with the Group 2 samples in Figure 3.7.

SIMPER analyses were done to determine which taxa were primarily responsible for influencing the sample groupings revealed in the dendrogram in Figure 3.7 ( samples analysed to lowest taxonomic rank). The two groups depicted in the dendrogram are compared in Table 3.13. The average dissimilarity between the two groups is shown to be 61.9 %. The taxa contributing the most to the dissimilarity between the two groups are placed in descending order of their percentage contribution in Table 3.13. It is clear from Table 3.13 that the high abundance of four crustacean taxa (copepod nauplii, ostracods, Cylindropsyllidae and Paramesochridae) in Group 1 was the main contributing factor distinguishing it from Group 2. In fact, the average abundances of all of the taxa listed in Table 3.13 were greater for Group 1 than for Group 2 except for the 2B nematodes (predator/omnivore group) and the turbellarians. Table 3.14 shows the taxa contributing the most to the similarity of samples depicted as Group 1 of the dendrogram in Figure 3.7. Table 3.14 shows that the main contributors to the similarity of this group of samples was the four crustacean taxa mentioned above and 1 nematode feeding group (1A), the selective deposit feeders. Table 3.15 shows the taxa contributing the most to the similarity of samples depicted as Group 2 of the dendrogram in Figure 3.6 and it is evident from Table 3.15 that all the nematode feeding groups and the tubellarians were the main contributors to the similarity of this group of samples.

Table 3.13
SIMPER analysis of Groups 1 and 2 depicted in Figure 3.7 (all taxa analysed)
Average dissimilarity = 61.9%

| Taxa | Group 1 Av Abund. | Group 2 Av. Abund. | Av. Diss. | Diss./SD | Contrib. % | Cum. % |
|---|---|---|---|---|---|---|
| Copepod nauplii | 243 | 10.1 | 6.95 | 1.32 | 11.2 | 11.2 |
| Ostracoda | 224 | 10.3 | 6.91 | 1.08 | 11.2 | 22.4 |
| Cylindropsyllidae | 124 | 3.11 | 5.37 | 1.45 | 8.68 | 31.1 |
| Paramesochridae | 100 | 17.3 | 4.68 | 1.32 | 7.56 | 38.6 |
| Saccosirridae | 157 | 8.63 | 4.47 | 0.78 | 7.22 | 45.9 |
| Gastrotricha | 62.6 | 1.79 | 3.9 | 1.53 | 6.3 | 52.2 |
| Nematoda 1B | 63.1 | 36 | 3.17 | 1.44 | 5.12 | 57.3 |
| Nematoda 2B | 39.2 | 52.4 | 2.7 | 1.14 | 4.37 | 61.6 |
| Nematoda 1A | 86.3 | 61.8 | 2.67 | 1.3 | 4.32 | 66 |
| Tisbidae | 26 | 3.38 | 2.52 | 0.91 | 4.07 | 70 |
| Nematoda 2A | 27.9 | 26.2 | 2.13 | 1.28 | 3.45 | 73.5 |
| Oligochaeta | 39.7 | 1.75 | 2.11 | 0.76 | 3.41 | 76.9 |
| Turbellaria | 30.9 | 31.8 | 2.05 | 1.21 | 3.32 | 80.2 |
| Acarina | 16.5 | 4.83 | 1.98 | 1.4 | 3.19 | 83.4 |
| Sarcomastgophora | 18.6 | 14.2 | 1.95 | 0.78 | 3.15 | 86.5 |
| Nerillidae | 28.1 | 1.44 | 1.53 | 0.61 | 2.48 | 89 |
| Tachiidae | 14.1 | 0 | 1.42 | 0.64 | 2.29 | 91.3 |

Table 3.14
SIMPER analysis showing the taxa which most responsible for the similarity of the samples depicted as Group 1 of the dendrogram in Figure 3.7 (all taxa analysed).
Average similarity = 55.22%

| Taxa | Av. Abund. | Av.Sim. | Sim./SD | Contrib. % | Cum. % |
|---|---|---|---|---|---|
| Copepod nauplii | 243 | 7.82 | 2.53 | 14.2 | 14.2 |
| Nematoda 1A | 86.3 | 6.48 | 3.38 | 11.7 | 25.9 |
| Ostracoda | 224 | 6.33 | 1.26 | 11.5 | 37.4 |
| Cylindropsyllidae | 124 | 4.9 | 1.28 | 8.87 | 46.2 |
| Paramesochridae | 100 | 4.5 | 1.63 | 8.15 | 54.4 |
| Nematoda 1B | 63.1 | 3.89 | 1.5 | 7.04 | 61.4 |
| Nematoda 2B | 39.2 | 3.86 | 2.54 | 7 | 68.4 |
| Gastrotricha | 62.6 | 3.61 | 1.36 | 6.53 | 74.9 |
| Turbellaria | 30.9 | 3.27 | 2.6 | 5.93 | 80.9 |
| Nematoda 2A | 27.9 | 2.25 | 1.33 | 4.07 | 84.9 |
| Saccociridae | 157.1 | 2.11 | 0.63 | 3.82 | 88.8 |
| Acarina | 16.5 | 1.93 | 1.22 | 3.49 | 92.2 |

Table 3.15
SIMPER analysis showing the taxa most responsible for the similarity of the samples depicted as Group 2 of the dendrogram in Figure 3.7(all taxa analysed).
Average similarity = 49.78%

| Taxa | Av.Abund. | Av. Sim. | Sim./SD | Contrib. % | Cum. % |
|---|---|---|---|---|---|
| Nematoda 1A | 61.8 | 12 | 2.72 | 24.2 | 24.2 |
| Turbellaria | 31.8 | 8.49 | 1.42 | 17.1 | 41.3 |
| Nematoda 2B | 52.4 | 8.4 | 1.32 | 16.9 | 58.1 |
| Nematoda 2A | 26.2 | 6.05 | 1.75 | 12.2 | 70.3 |
| Nematoda 1B | 36 | 5.38 | 1.16 | 10.8 | 81.1 |
| Ostracoda | 10.3 | 1.87 | 0.66 | 3.75 | 84.9 |
| Copepod nauplii | 10.1 | 1.73 | 0.68 | 3.48 | 88.3 |
| Sarcomastigophora | 14.2 | 1.46 | 0.47 | 2.93 | 91.3 |

The dendrogram showing station similarities for the study area for all sampling trips where animals were identified to the highest taxonomic rank (Table 2.3) is depicted in Figure 3.9. Two main groups are shown as in Figure 3.7. Groups 1 and 2 from both figures share most of the same samples except in Figure 3.7, samples iiN2, vN3 and viN2 occur in Group 1 whereas they move to Group 2 in Figure 3.9 (indicated by boxes) and sample viS0 occurs in Group 2 in Figure 3.7 but moves to Group 1 in Figure 3.9 (indicated by box). This indicates that overall there was little difference in the Bray-Curtis analysis whether meiofauna were identified to major taxa or to a lower taxonomic rank or feeding group (Table 2.2).

The mutidimensional scaling (MDS) ordination which relates to the Bray-Curtis similarity plot shown in Figure 3.9 is shown in Figure 3.10. The stations coloured red and underlined correspond with the Group 2 stations in Figure 3.9. This figure is very similar to Figure 3.8 where taxa were analysed to the lower taxonomic rank except for the stations that moved to different groups as indicated by the boxes.

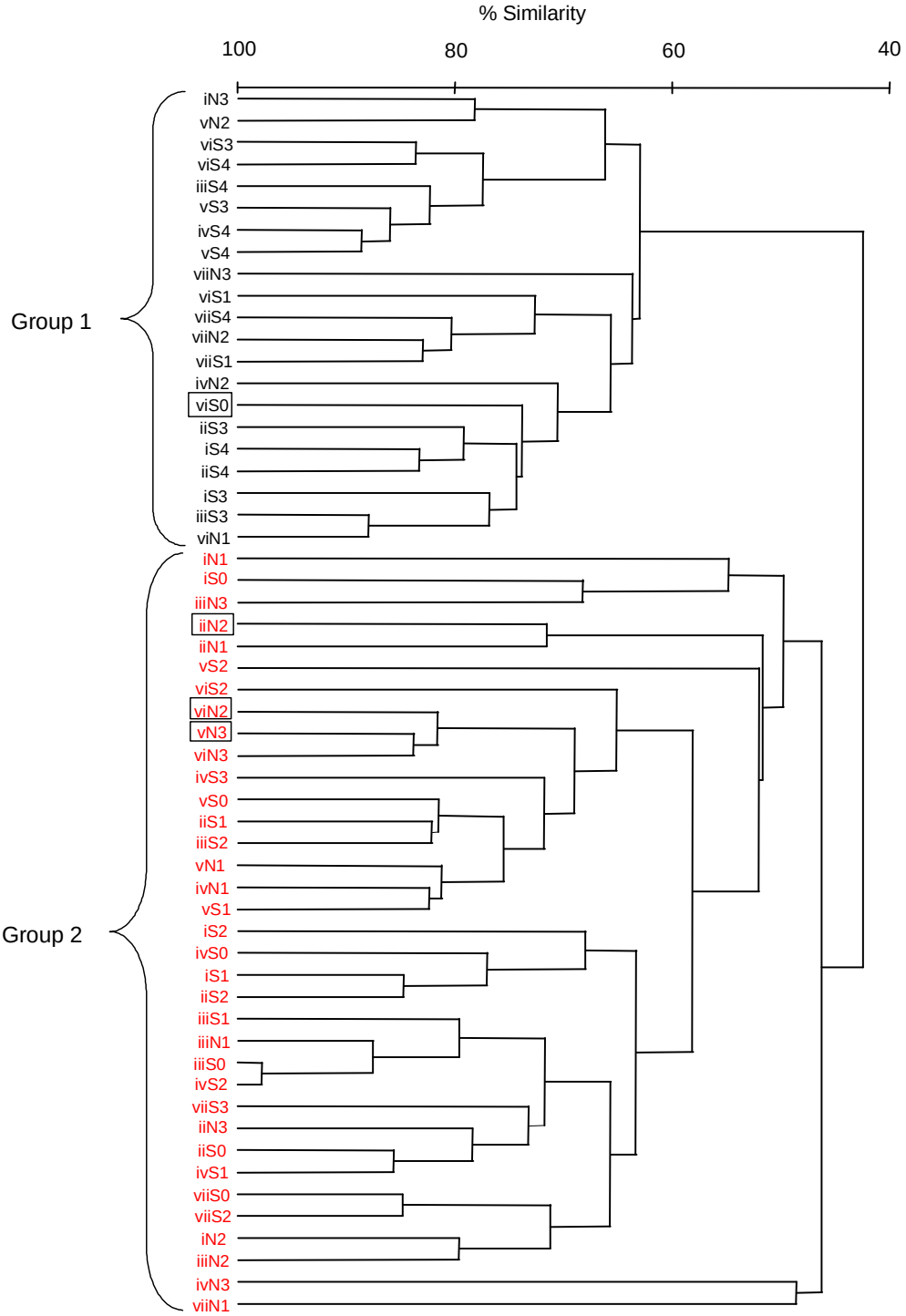


Figure 3.9**.** Dendrogram showing hierarchical relationships between meiofauna samples taken at Sezela beach and identified to the major taxonomic rank (Table 2.3). Similarity was computed using the Bray-Curtis coefficient and Square root transformations. The Roman numerals i to vii indicate the sample date of which there were seven (i to vii).

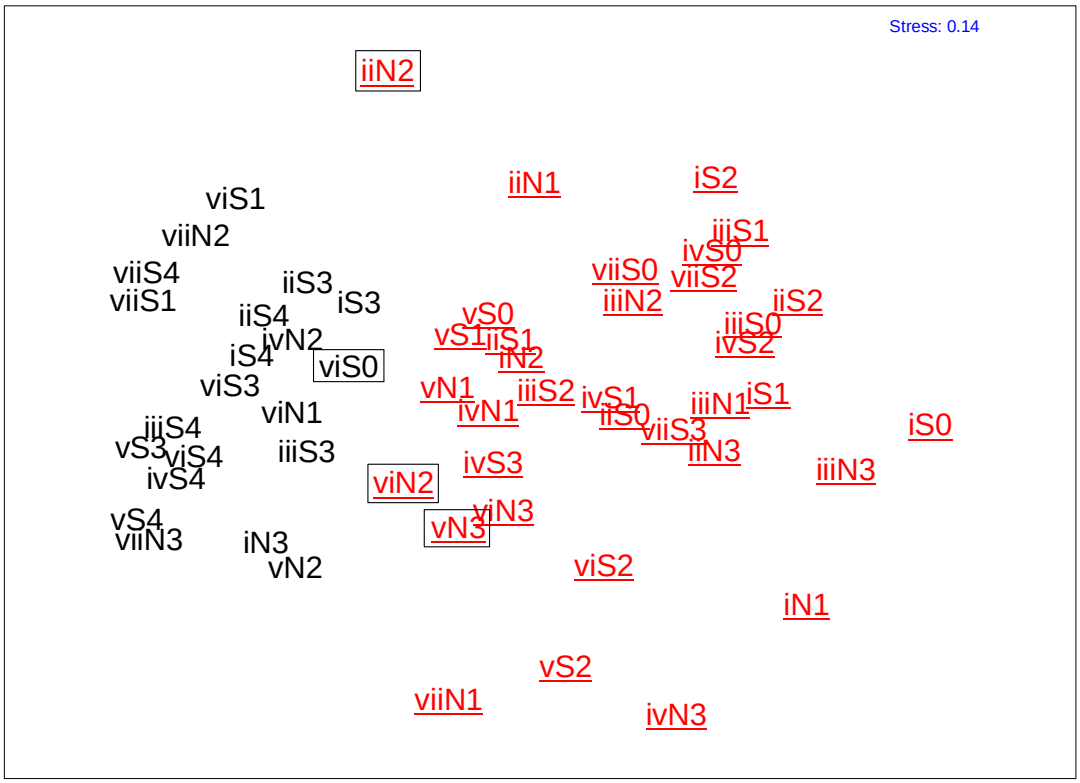


Figure 3.10. MDS plot of all meiofauna samples identified to the highest taxonomic rank (Table 2.3). Stations in red and underlined correspond with the Group 2 samples in Figure 3.9. The boxed samples are those that changed groups after being analysed to the higher taxonomic rank.

The two groups depicted in the dendrogram in Figure 3.9 (analysed to highest taxonomic rank) are compared by SIMPER analysis in Table 3.16. Here again as in Table 3.13 it is the crustacean taxon Harpacticoida that contributed the most (21.69 %) to the dissimilarity between the two groups. In Table 3.13, the harpacticoid families that contributed highly to the dissimilarity of the two groups were Cylindropsyllidae and Paramesochridae. The average abundance of all the taxa in Table 3.16 is much higher for Group 1 than for Group 2. Table 3.17 shows the taxa most responsible for the similarity between the samples of Group 1. The harpacticoid copepods contributed the highest percent (27.97%) for this group while in Table 3.18 the nematodes and turbellarians contributed the most to the similarity of Group 2 (73.85 % and 14.7 % respectively).

Table 3.16
SIMPER analysis of Groups 1 and 2 depicted in Figure 3.9 (analysed to highest taxonomic rank). Average dissimilarity = 74.05%

| Taxa | Group 1 Av. Abund. | Group 2 Av. Abund. | Av. Diss. | Diss./SD | Contrib.% | Cum.% |
|---|---|---|---|---|---|---|
| Harpacticoida | 256 | 15.5 | 16.1 | 1.54 | 21.7 | 21.7 |
| Nematoda | 229 | 165 | 12.4 | 1.07 | 16.8 | 38.5 |
| Ostracoda | 201 | 9.97 | 12.2 | 0.68 | 16.5 | 55 |
| Annelida | 216 | 10.1 | 11.7 | 0.78 | 15.8 | 70.8 |
| Copepod nauplii | 222 | 6.88 | 11.2 | 0.92 | 15.1 | 85.9 |
| Gastrotricha | 53 | 3.84 | 3.47 | 0.86 | 4.69 | 90.6 |

Table 3.17
SIMPER analysis showing the taxa most responsible for the similarity of samples depicted as Group 1 of the dendrogram in Figure 3.9 (analysed to highest taxonomic rank). Average similarity = 44.16%

| Taxa | Av. Abund. | Av. Sim. | Sim./SD | Contrib.% | Cum.% |
|---|---|---|---|---|---|
| Harpacticoida | 256 | 12.4 | 1.37 | 28 | 28 |
| Nematoda | 229 | 11.8 | 1.65 | 26.8 | 54.8 |
| Copepod nauplii | 222 | 6.36 | 1.32 | 14.4 | 69.2 |
| Annelida | 216 | 5.12 | 0.81 | 11.6 | 80.8 |
| Ostracoda | 201 | 4.82 | 0.65 | 10.9 | 91.7 |

Table 3.18
SIMPER analysis showing the taxa most responsible for the similarity of the samples depicted as Group 2 of the dendrogram in Figure 3.7 (analysed to highest taxonomic rank). Average similarity = 49.49%

| Taxa | Av. Abund. | Av. Sim. | Sim./SD | Contrib.% | Cum.% |
|---|---|---|---|---|---|
| Nematoda | 165 | 36.6 | 2.4 | 73.9 | 73.9 |
| Turbellaria | 33.9 | 7.27 | 0.92 | 14.7 | 88.6 |
| Harpacticoida | 15.5 | 1.71 | 0.56 | 3.46 | 92 |

## 3.5 The Nematode/Copepod ratio

A one way analysis of variance (ANOVA) was carried out to test for significant differences between stations over the seven sampling periods in terms of the N/C ratio. The data needed to be $\log_{10}$ transformed to meet the statistical assumptions of normality in order to proceed with a one-way ANOVA. Figure 3.11 shows differences between stations with the untransformed data where one can see that Station S4 had the lowest N/C ratio. Higher ratios occurred closer to the outfall. There was a large degree of variability in the N/C ratio at stations closer to the outfall and lower variability at stations further away such as Stations N3, S3 and S4 which had generally lower N/C ratios throughout as shown in Figure 3.12. All stations had low N/C rations during the April 2001 sampling occasion. The differences in the mean values for the N/C ratios were greater than would be expected by chance ($P<0.05$) indicating a statistically significant difference, were the F statistic was 4.805 with $P<0.001$. The pair-wise multiple comparison procedures revealed that Station S4 was significantly different to four other stations, namely S2, S0, N1 and S1 with P values of 0.002, 0.002, 0.003 and 0.032 respectively. Station S3 was also revealed to be significantly different to three other stations namely S2, S0 and N1 with P values of 0.033, 0.035 and 0.045 respectively. These two stations were the greatest distance from the effluent outfall.

The Nematode/Copepod (N/C) ratio was used to test whether it supported the findings of the groupings in the multivariate analyses. For sampling stations and times where replicates were taken, the mean counts of Nematodes and Harpacticoid copepods were used. If one compares the N/C ratios in Table 3.19 to the way the samples group as Groups 1 and 2 in Figures 3.7 and 3.9, it is evident that the majority of samples with ratios below 4.5 cluster in Group 1 and the majority of samples with a ratio above 4.5 cluster in Group 2. Figure 3.7 is the dendrogram for all taxa identified and the samples marked in red and underlined in Table 3.19 indicate the samples found in Group 2 of Figure 3.7. In Group 1 only two samples have a N/C ratio greater than 4.5, namely viN3 and iiN2 indicated with circles in Table 3.19. In Group 2 there were only 3 samples with a ratio less than 4.5, namely ivS3, viS2 and viS0 indicated by boxes in Table 3.19. Figure 3.9 is the dendrogram for major taxa identified. Here the two samples with higher ratios from Group 1 (viN3 and iiN2) in Figure 3.7 move into Group 2 in Figure 3.9 and the sample viS0 found in Group 2 in Figure 3.7 moved into Group 1 in Figure 3.9. Therefore all samples found in Group 1 of Figure 3.9 had ratios below 4.5 and only two samples namely viS2 and ivS3 had ratios below 4.5 in Group 2. The N/C ratio values are strongly aligned with the groupings shown in the dendrograms in Figures 3.7 and 3.9. There was a slightly stronger agreement between the N/C ratio and the groupings in the dendrogram when the meiofauna was identified to major taxa level (Figure 3.9). This was expected as the Nematode feeding groups and the Harpacticoid families are each grouped as a single taxon in the multivariate analysis. However the ratio would not be very efficient in revealing impacted stations when the abundance of both Nematodes and Harpacticoids has been reduced by adverse conditions. This was evident at Station viS2 where the total numbers of animals ranged from 66 to 104 which was the lowest of all sampling stations (Table A1.6) and a low N/C ratio of 2 was recorded.

Table 3.19
Nematode /Copepod ratios for the 8 stations and 7 sampling dates

| Sampling dates | N3 | N2 | N1 | S0 | S1 | S2 | S3 | S4 |
|---|---|---|---|---|---|---|---|---|
| (i) 4th July 2000 | 0.3 | 20.5 | 46 | 21 | 13 | 79 | 0.6 | 0.72 |
| (ii) 30th August 2000 | 39 | 321 | 44.8 | 17.7 | 4.7 | 31 | 1.4 | 0.6 |
| (iii) 13th December 2000 | 48 | 40.5 | 116 | 122 | 262 | 4.6 | 1 | 0.4 |
| (iv) 26th January 2001 | 7.5 | 0.7 | 45.4 | 106 | 11.6 | 122 | 1.1 | 1.6 |
| (v) 8th March 2001 | 2.6 | 0.4 | 17.5 | 14.1 | 29.9 | 107 | 0.8 | 0.4 |
| (vi) 9th April 2001 | 19.8 | 2.6 | 0.6 | 3.6 | 2.5 | 2 | 2.2 | 0.8 |
| (vii) 2nd January 2002 | 0.2 | 4.5 | 40.7 | 88 | 0.7 | 161 | 25.6 | 0.4 |

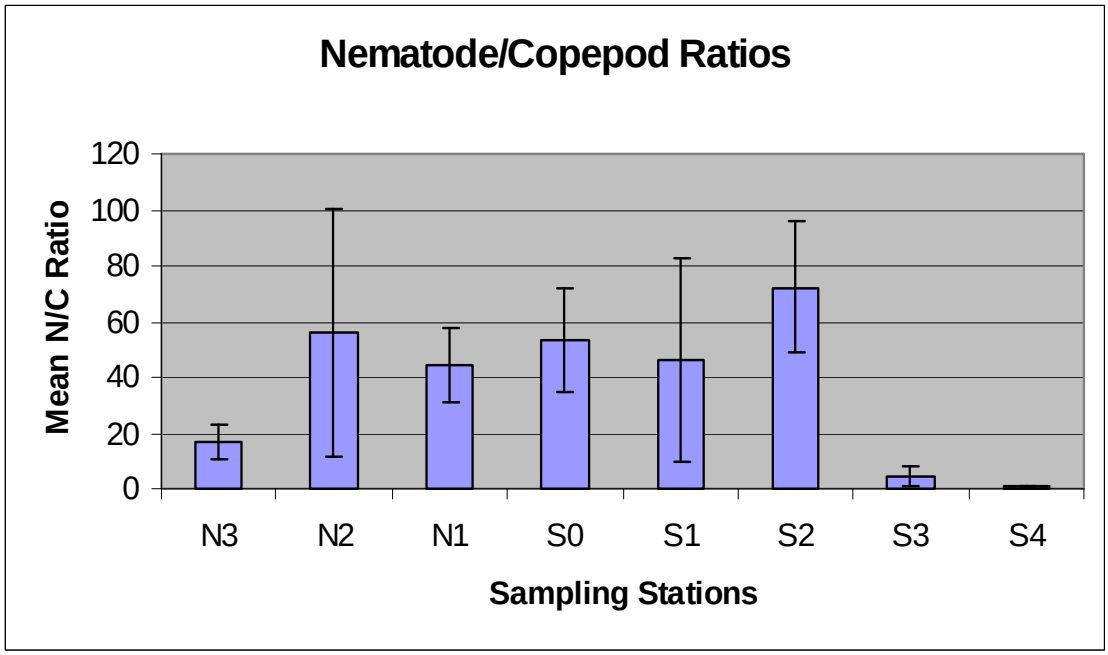


Figure 3.11. Mean Nematode/Copepod ratios for the whole study period for each site. Bars indicate standard errors (n=7).

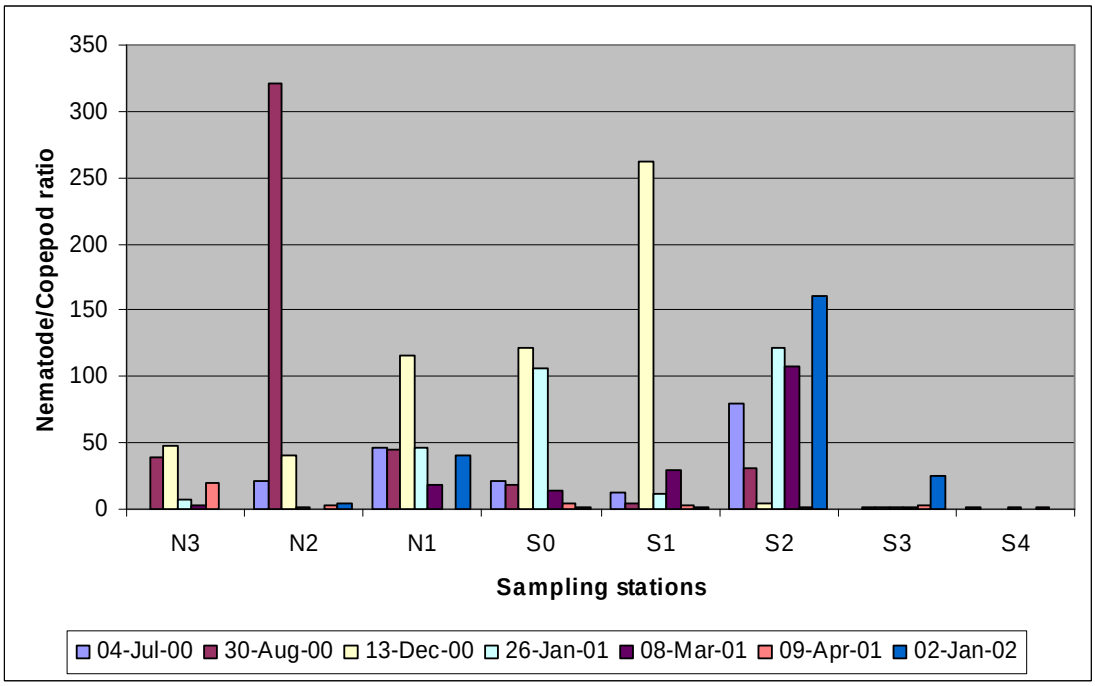


Figure 3.12 Nematode/Copepod ratios for each of the seven sampling periods.

## 3.6 Physical and Chemical assessment of the environment

The results of the chemical analyses and sediment grain size analyses are in Appendix 2 in Tables A2.1 to A2.21.

A BIO-ENV procedure was run with PRIMER, which selects combinations of environmental variables that best relate to the observed biological pattern of the CLUSTER and MDS analysis. The combination of salinity, dissolved oxygen and mean sediment grain size was shown to contribute the most to the observed biological pattern with a P value of 0.1. For a perfect match, P = 1 and for no correlation, P = 0, therefore the correlation between the measured variables and the abiotic data was not very strong.

Scatter plots were generated to compare three biological variables (Total meiofauna numbers, numbers of taxa and the nematode/copepod ratio) with a number of physical and chemical variables. A regression analysis was then done to test the significance of these relationships and the results are ahown in Table 3.20.

From Table 3.20 the strongest correlation was found between total numbers of meiofauna and nitrogen in the interstitial waters with a product moment correlation coefficient of 0.96. The regression analysis revealed a $P < 0.01$ indicating a significant positive relationship between these two variables. The R-squared statistic indicates that the model as fitted explains 91.87% of the variability in total numbers of meiofauna. This suggests a strong influence of organic input on meiofauna numbers at Sezela. The scatter plot of the correlation is shown in Figure 3.12.

Table 3.20
Regression analyses between the physical and chemical variables and three biological variables: Total numbers, number of taxa and the Nematode/Copepod ratio. The P values in bold and underlined indicate significant relationships.

| | Sed. Size | COD-W | COD-sed | Kjel-W | Kjel-sed | DO | pH | Salinity | Ammonia | % gravel | Fine sand |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **Total No.s** | | | | | | | | | | | |
| P value | **0.0043** | **0.0023** | 0.6226 | **0** | **0.0252** | 0.5506 | 0.4662 | 0.1357 | **0.003** | **0.0379** | **0.0768** |
| Correlation coefficient | -0.376 | 0.399 | 0.0672 | 0.959 | 0.299 | -0.082 | 0.0993 | 0.202 | 0.3898 | -0.2781 | 0.2384 |
| R-squared | 14.12% | 15.89% | 0.45% | 91.87% | 8.93% | 0.66% | 0.99% | 4.07% | 15.20% | 7.74% | 5.68% |
| Stand.error of Est. | 493.747 | 488.64 | 531.59 | 151.89 | 508.44 | 531.02 | 530.16 | 521.83 | 490.643 | 511.763 | 517.432 |
| **No. of Taxa** | | | | | | | | | | | |
| P value | 0.1193 | 0.529 | **0.0027** | 0.5616 | 0.1193 | **0.052** | 0.969 | **0.0873** | 0.5819 | 0.2158 | 0.2248 |
| Correlation coefficient | 0.2105 | 0.0859 | -0.394 | 0.0792 | 0.211 | 0.261 | 0.0053 | 0.231 | 0.0752 | 0.168 | -0.165 |
| R-squared | 4.43% | 0.74% | 15.53% | 0.63% | 4.43% | 6.82% | 0.00% | 5.32% | 0.56% | 2.82% | 2.72% |
| Stand.error of Est. | 3.937 | 4.012 | 3.791 | 4.015 | 3.94 | 3.887 | 4.027 | 3.918 | 4.016 | 3.97 | 3.972 |
| **Nem/Cop ratio** | | | | | | | | | | | |
| P value | **0.0385** | 0.5805 | 0.3692 | 0.3486 | 0.297 | 0.3841 | 0.8813 | **0.01** | 0.4448 | 0.186 | 0.7597 |
| Correlation coefficient | -0.2773 | -0.075 | 0.1223 | -0.128 | -0.142 | -0.119 | -0.02 | -0.3417 | -0.1042 | -0.1793 | -0.0418 |
| R-squared | 7.69% | 0.57% | 1.50% | 1.63% | 2.01% | 1.41% | 0.04% | 11.68% | 1.09% | 3.22% | 0.17% |
| Stand.error of Est. | 61.341 | 63.663 | 63.366 | 63.323 | 63.199 | 63.395 | 63.832 | 60.002 | 63.498 | 62.81 | 63.789 |

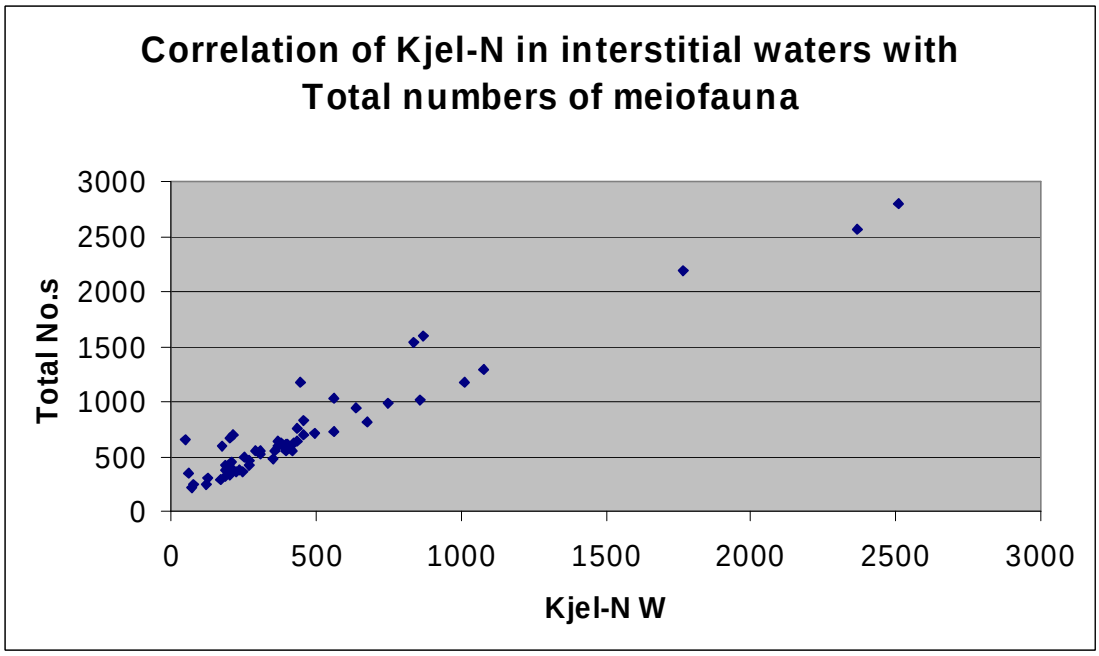


Figure 3.12. Scatter plot of the product moment correlation coefficient of 0.96 between Kjel-N of the interstitial waters and total numbers of meiofauna.

There were other significant correlations noted for other variables (P values in bold in Table 3.20). Total numbers of meiofauna had a significant negative correlation with mean sediment size and with % gravel. It also had a significant positive correlation with COD in the interstitial waters, nitrogen in the sediment, ammonia, fine sand and nitrogen in the interstitial waters as mentioned above. Higher COD and nitrogen and ammonia values suggest higher organic enrichment which was resulting in increased total numbers of meiofauna.

A significant negative correlation between numbers of meiofauna taxa and COD in the sediment was revealed (Table 3.20). There was also a significant positive correlation between numbers of taxa and dissolved oxygen and salinity. Reduced salinity suggests fresh water input from ground water and the estuaries in the area. All these factors suggest that organic enrichment may increase meiofauna numbers but reduce diversity.

There was a significant negative correlation between the Nematode/Copepod ratio and mean sediment grain size and salinity suggesting increased numbers of harpacticoid copepods with increased grain size and increased salinity.

# 4. DISCUSSION

In studies of unpolluted and unperturbed sandy beaches, Coull (1988) gives numbers of total meiofauna per 10 $cm^2$ to range from 60 to 2250, nematodes ranged from 35 to 1328 and harpacticoid copepods ranged from 10 to 502. In studies of four sandy beaches in the Eastern Cape, McLachlan (1977) found numbers of total meiofauna per 10 $cm^2$, at the low water mark to range from 97 to 1320. Nematodes ranged from 71 to 220 and harpacticoid copepods ranged from 14 to 1014. The numbers found for this study for total meiofauna per 10 $cm^2$ and to a depth of 20 cm ranged from 45 to 4414, nematodes ranged from 21 to 777 and harpacticoid copepods ranged from 0 to 1121. Brown and McLachlan (1990) stated that beaches can display numbers between 50 per 10 $cm^2$ and 3000 per 10 $cm^2$ depending on physical and chemical gradients with the highest values from intermediate beaches with reasonable organic inputs.

Total numbers of meiofauna were significantly correlated with nitrogen in the interstitial waters ($P < 0.01$) (Table 3.20) suggesting that one of the strongest influences on meiofauna numbers at Sezela to be related to organic input. The pattern of high kjeldahl-N and dense interstitial fauna on a beach is usually associated with organic pollution (Oliff *et al.*, 1967). The notion that organic enrichment was influencing the total numbers of meiofauna was also supported by significant (Table 3.20) relationships between total numbers and COD and ammonia in the interstitial waters and nitrogen in the sediments (see Table 3.20).

A significant negative correlation was found between total numbers of meiofauna and mean grain size and % gravel and a significant positive correlation with fine sand. According to Gheskiere *et al.*. (2005b) sediment grain size is one of the main factors influencing the distribution of meiofauna and higher meiofauna densities are expected in coarser and therefore more oxygenated sands. However the beaches they studied had median grain sizes ranging from 0.375 up to 0.509 mm. At Sezela beach the mean particle size ranged from 0.26 mm to 1.23 mm but 64% of the samples had mean grain sizes above 0.5 mm and according to a conceptual model by Brown & Mclachlan (1990) shown in Figure 4.1, both interstitial harpacticoids and nematodes reduce in abundance above 0.5 mm grain size. This then explains the negative correlation between total numbers and mean grain size and % gravel for this study.

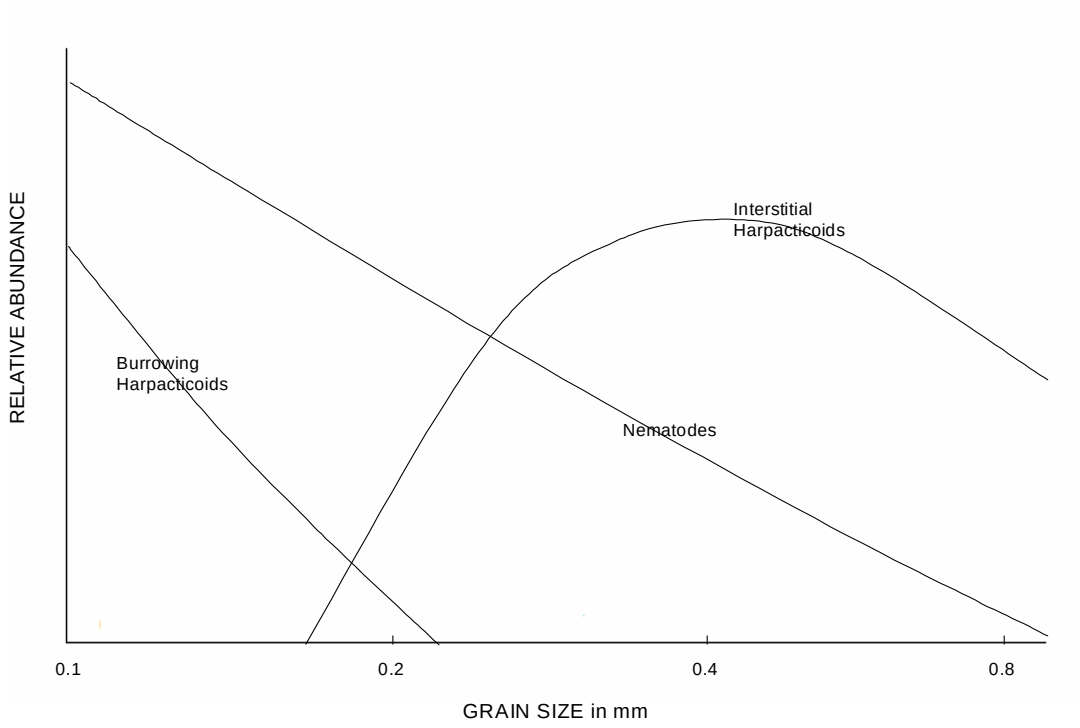


Figure 4.1 Conceptual model of response of sandy-beach meiofauna to a particle size gradient (Brown and Mclachlan, 1990).

In terms of total numbers the only sample below 50 was Station iS0 (closest to the out fall) at 45. Almost any disturbance was found by Coull & Palmer (1984) to result in an immediate decrease in the abundance and diversity of all the meiofauna indicating a possible disturbance here by the effluent. In terms of nematode numbers Station iS0 had the lowest at 21 with ivN3 also low at 30. Low harpacticoid copepod numbers were also a major feature of the data with 40 of the replicate samples having numbers below 10. Stations S0 and S2 had the highest frequency of samples <10 at 8 and 9 samples respectively. Station N1 had 7, Station S1 had 5, Station S3 had 3 and Station N3 had 6 samples. Crustacean groups, particularly the harpacticoid copepods are generally considered to be the most sensitive groups to pollution impact (Sarkka, 1995; Carmen *et al.*, 2000 & Lee *et al.*, 2001). The above evidence seems to indicate that all stations may have been affected at some stage except for Station S4 which was the greatest distance from the effluent outfall. It also suggests that Stations S0 and S2 were possibly the most heavily impacted by the effluent. However, the large range of numbers found for this and other studies reflects a large natural variability in numbers which occurs in relation to natural fluctuations in wave height, sand movement and organic input from the marine environment and terrestrially from fresh water sources such as estuaries, storm water drains and ground water. Further to this the strong correlation of meiofauna numbers with nitrogen and COD of interstitial waters, suggests that the variability in numbers cannot be directly linked to the effluent discharge unless it represents the only source of organic input. Other studies have also shown that the variability of natural conditions effects meiofauna population dynamics making it difficult to relate pollution to the observed fluctuations. Ansari & Ingole (2002) investigated the short and long term effects of a fuel oil spill from a grounded ship "*M V Transporter"* on meiofauna of a sandy beach of the central west coast of India and found that the effects of the oil spill were confounded with seasonal monsoon effects and beach dynamics. Large seasonal variations in the occurrence of meiofauna occurs in Indian beaches due to the south west monsoons and the authors found that the

synchronization of the monsoons with the maximum concentration of petroleum hydrocarbons in the sediment made it difficult to determine the effect of the oil spill on the meiofauna. They found that meiofauna were highly resilient in nature and recovered quickly, and thus long term effects were also difficult to detect. After the *Agip Abruzza* oil spill Danovaro *et al.* (1995) also found meiofauna to be highly resilient recovering after only 2 weeks and a few weeks later reaching a level almost indistinguishable from pre-pollution conditions.

From the analysis of variance (ANOVA) of a number of univariate measures, a significant difference was found between Station S4 and Station S2 in terms of the number of taxa present and also between Station S4 and Stations S2, S0, N1 and N3 in terms of total numbers with Station S4 having the highest (Figure 3.1(a) and (b)). If it was the effluent causing the reduced numbers at Stations S0, S2, N1 and N3 then Station S4 was possibly being the least impacted by the effluent. Reduced numbers and taxa are indicative of impacted sites (Coull & Palmer, 1984). Reduction in the total and species level meiofauna abundance after contamination with hydrocarbons has also been demonstrated both in the natural habitat and in laboratory experiments (Danovaro *et al.,* 1995 and Ansari & Ingole, 2002). In a laboratory study Sandulli & De Nicola Giudici (1989) found a net decline in all meiofauna taxa proportional to the applied organic load of sewage sludge. However, this may have been partly related to factors imposed by experimental conditions as reduced numbers also occurred in their controls. They state that in a field situation where recruitment can occur the organic load equivalent to their lowest treatment of 900 µg C $g^{-1}$ may have resulted in faunal enhancement. The strong positive correlation of meiofauna numbers with nitrogen in the interstitial waters for this study supports this idea. It would also have been worth assessing the sediment organic content at each station for this study for comparison.

SIMPER analyses to establish which taxa were responsible for the various groupings in all of the CLUSTER and MDS analyses revealed a higher average abundance of most taxa in the Group 1 samples compared to the Group 2 samples from Figures 3.3, 3.5, 3.7 and 3.9. As mentioned for the ANOVA between stations in terms of total numbers, almost any disturbance results in an immediate decrease in the abundance and diversity of the meiofauna (Coull, 1988; Sandulli & De Nicola Giudici, 1989; Danovaro *et al.,* 1995 and Ansari & Ingole, 2002). Therefore the Group 2 samples were possibly being impacted on by the effluent. A rapid recovery of harpacticoid copepods from the sixth sampling occasion was noted at Station viS0. This may be expected as effluent was not being pumped to sea at that time and meiofauna are known to recover rapidly from disturbance due not only to active or passive migration, but also to high intrinsic population growth potential ie. short generation times (Alongi, 1985). Changes in community structure can take place over a time span of weeks to months rather than years (Heip *et al.,* 1988). Further to this members of all the major meiobenthic taxa have been found in the water column, and at least the better swimmers among harpacticoid copepods and turbellarians may actively leave the sediment and disperse by tidal currents (Armonies, 1990). The role of passive processes in meiobenthic copepod dispersal have been emphasized by Palmer (1988) to dominate in more hydrodynamically rigorous areas such as beaches. However Kern (1990) proposed that active behaviour is more important in determining copepod dispersal and, ultimately copepod abundance in the sediment. If population density is too high, food levels too low, potential mates relatively unavailable, or it is otherwise unacceptable, a copepod could swim out of the sediment to be dispersed by currents or reduce behaviours that tend to help resist being entrained by flow (Armonies, 1988; Commito & Tita, 2002). Once in the water column copepods will largely be passively transported by currents until they are deposited in a new patch. This would enhance recovery of meiofauna in depleted areas. Gheskiere *et al.,* (2006) used a front end loader to experimentally clean litter from a Belgian beach in order to assess the effects of this on the meiofauna. The top 5 cm of sediment were removed in this process after which they found that the drop in meiofaunal densities caused by mechanical beach cleaning recovered to initial values after the next high tide. Gheskiere *et al.,* (2006) assumed that recolonization occurred

via passive vertical migration forced by the upcoming tide, from the underlying sediment layers.

From the ANOVA between seasons of a number of univariate indices a significant difference was found between summer and winter, autumn and winter and between summer and autumn in terms of the numbers of taxa present (Figure 3.1(a)).The highest numbers of taxa were recorded in autumn which coincided with the factories closed season when no effluent was being pumped to sea. Reduced numbers of taxa in winter would have been affected by lower temperatures as most meiofauna communities exhibit some seasonality, with greatest abundances in the warmer months (Coull, 1987; Brown & McLachlan, 1990). A significant difference was found between autumn and summer in terms of species richness with the highest species richness recorded in autumn (Figure 3.2(c)). This is opposite to what would be expected at a pristine site (Coull, 1987; Brown & McLachlan, 1990) and the higher species richness in autumn could be related to the absence of effluent being pumped to sea during the autumn sampling run. Taxon diversity of the meiofauna phyla has been proposed as a possible tool for the assessment of pollution effects by Herman *et al.* (1985). Taxon diversity is lower in pollution conditions; this is caused mainly by the disappearance of some sensitive taxa e.g. in the ostracods, gastrotrichs, halacarids, hydrozoans and tardigrades (Vinx & Heip, 1991). Therefore the above effect of higher species richness in autumn may have been caused by the lack of effluent discharge at the time and not seasonally. No significant differences between seasons were found in terms of the other univariate indices analysed and are therefore not discussed further.

The results indicated little difference in relationships between stations whether analysing the meiofauna to a lower or higher taxonomic rank. For the analysis of the replicated data sets for the 9$^{th}$ April 2001 and 2$^{nd}$ January 2002, the samples labeled as Group 1 in Figure 3.3 (identified to lowest taxonomic rank) all group together again as Group 1A in Figure 3.5 where the samples were analysed to the major taxonomic rank. Groups 2A and 2B in Figure 3.3 remain mostly the same with only four samples swopping positions between groups in Figure 3.5 (analysis to major taxonomic rank) and Group 2A from Figure 3.3 moves into Group 1 in Figure 3.5 as Group 1B. A SIMPER analysis revealed that the cause for this was the combining of the nematode feeding groups, the annelid families and the harpacticoid families as single taxa. The greater numbers of these taxa make these samples less similar to the Group 2 samples and more similar to the Group 1 samples in Figure 3.5. Further to this when comparing the MDS plots for the analysis to lower taxonomic rank (Figure 3.4) and analysis to the higher taxonomic rank (Figure 3.6), the relationship between the three groups of samples formed in the CLUSTER analyses does not appear to be very different except for the four samples that changed groups as indicated by the small circles in Figure 3.6.

The analysis of all the data sets for the seven sampling times revealed the same conclusion with little difference between the analysis to the lower or higher taxonomic ranks. For the CLUSTER analyses shown in Figures 3.7 and 3.9, two main groups are shown (Groups 1 and 2). The only difference between the two analyses was that four stations changed groups. Station viS0 moved to Group 1 and Stations iiN2, viN2 and vN3 move to Group 2 (indicated by boxes). This is also apparent when comparing the two MDS plots in Figures 3.8 (lowest taxonomic rank) and 3.10 (highest taxonomic rank). Here too, the stations that moved to different groups are indicated by boxes.

In the sea phyletic diversity is extremely high and Gray *et al.* (1990) found that both species and phyla showed sequential responses to stressors. Recent studies suggest that taxon richness reflects the specific diversity and that little information appears to be lost at higher taxonomic levels (Mirto & Danovaro, 2003). Warwick (1988) found strong indications that pollution effects are detectable at even higher taxonomic levels than genus or family. Factors that influence the occurrence and abundance of species (both natural and anthropogenic factors) may still be recognised from monitoring based on higher taxonomic levels (Herman & Heip,

1988). Therefore, working at higher taxonomical levels than the species is believed to convey sufficient information (Moore & Bett, 1989) and is confirmed in this study.

However, in contrast to the observed general similarity between the analyses of samples to lower and higher taxonomic ranks, the movement of Station viS0 into Group 1 in Figure 3.9, indicates a possible recovery at the pipe station when the samples are analysed to the major taxonomic rank. It has been suggested that changes in community structure also occur due to natural variables which may mask the effects of pollution and multivariate analyses based on higher taxa may more closely reflect gradients of contamination or stress than those based on species data, the latter being more affected by 'nuisance' environmental variables (Warwick, 1988a & 1988b and Heip *et al.*, 1988). In many situations this is due to changes in natural environmental variables from place to place or time to time which may result in species replacement, since species are normally adapted to rather narrow ranges of environmental conditions. This may confound any change in pattern due to the perturbation under investigation. However natural environmental variables, such as water depth or sediment granulometry, may not alter the proportions of major taxa present, and if there is a degree of coherence among species in these higher taxa with respect to their response to a perturbation, the response will be more evident above the natural environmental noise (Warwick, 1993). If one looks at the raw data in Appendix 1 (Tables A1.1 to A1.7), it is apparent that Station S0 had extremely low numbers of harpacticoid copepods throughout the study period (range 0 to 21) with the exception of a considerable increase on the sixth sampling occasion of numbers ranging from 185 - 226 harpacticoids (Table A1.6). This appears to be a progression towards recovery after only a couple of weeks of effluent stoppage, which is hidden by the analysis to lower taxa and is possibly indicative of what Warwick (1988a & 1988b) terms 'nuisance' variables where natural variables effect the community structure at lower taxonomic levels and complicate the detection of pollution effects. It may be possible that a pioneer species within the family Paramesochridae initially colonises an area after the cause of the negative impact has ceased as it may be more resilient to the residual pollutants in the sediments. Therefore the signal of environmental recovery in this case seems to be more visible when analysis is made of major taxa. According to Somerfield & Clark (1995) whatever taxonomical level the analysis is carried out to, interpretable results are possible, especially if the pattern of community change is marked and the response to a pollution event can be more clear-cut at higher taxonomic levels than the species level (e.g. Family, order and/or Phylum).

An analysis of similarity (ANOSIM) of the replicated data sets for the 9$^{th}$ April 2001 and 2$^{nd}$ January 2002, showed that the ranked similarities between replicates within a station were significantly greater than the ranked similarities of replicates between different stations. The global R statistic values for all taxa identified as well as major taxa identified was 0.948 and 0.914 respectively with an associated significance level of $P<0.001$ for both analyses. This suggests that sites were still highly distinguishable from one another whether the meiofauna was identified to major taxonomic ranks or divided into lower taxa. This was also evident in the CLUSTER analyses of these two data sets, where replicates within sites clustered closely together, as shown by the small brackets in Figure 3.3 (analysed to lowest taxonomic rank) and Figure 3.5 (analysed to major taxonomic rank). According to Sandulli & Pinckney (1999) meiofauna are patchily distributed on tidal sand flats in correlation with microalgae which are also patchily distributed and constitute a primary food source for a wide variety of meiofaunal organisms such as nematodes, copepods, protistans, oligochaetes, turbellarians, polychaetes and amphipods. However the high similarity of replicates within stations for this study suggests low patchiness per station within the swash zone on high energy beaches. Moreno *et al.* (2006) suggested that on sandy beaches autotrophic primary production was negligible and stronger correlations exist between meiofauna, bacteria, and organic matter. It has been suggested that the main function of the beach interstitial system is the processing of organic materials flushed into the sand (Brown & McLachlan, 1990). Dissolved and particulate organic matter, is mineralized by a food chain with heterotrophic bacteria as its base (Brown & McLachlan 1990). Ciliates, nematodes and harpacticoid copepods feed on particulate

organic matter and bacteria and tubelarians and predatory nematodes feed on other meiofauna (Reise, 1985). Therefore most beaches function as biological filters that mineralize organic materials and have no direct trophic interactions with other food chains (Brown & McLachlan, 1990).

The SIMPER analyses of the replicated data sets for the 9th April 2001 and 2nd January 2002, revealed that only the sarcomastigophorans had a higher abundance in Group 2 samples from Figures 3.3 and 3.5 shown in Tables 3.1 and 3.7. It is known that some Foraminifera (a most conspicuous order within the Sarcomastigophora) are more tolerant to heavy metals than nematodes (Gustafsson *et al.*, 2000) and previous work has shown that some Foraminifera species thrive in organically enriched, oxygen-depleted environments (Gooday *et al.*, 2000). Sarcomastigophorans are tolerant of reducing conditions and feed on bacteria, diatoms, other protozoans and even metazoans (Brown & McLachlan, 1990). However the relationships between free-living protozoa and meiofauna are virtually unknown (Alongi, 1985). Certain species of sarcomastigophorans may therefore be good indicators of some types of anthropogenic perturbations.

For both analyses to lower and higher taxonomic ranks of the last two replicated data sets, the nematodes, turbellarians and sarcomastigophoran numbers played a major role in the similarities of the Group 2 samples. The relative abundance of the nematods and turbellarians and the reduced numbers of crustacean taxa, were the main contributors to the similarity of the Group 2 samples for the analysis of all data sets for all seven sampling times. For the analysis to lower taxonomic ranks for all the data sets, only the 2B nematodes (predatory) and tubellarians had higher abundances in Group 2 than in Group 1 (Table 3.13). Higher population densities of large predatory/omnivorous nematodes are normally associated with heavily organically polluted areas and/or physically disturbed systems (Netto *et al.*, 1999 and Danovaro, 2000). Turbellarians are also known to be voracious predators (Danovaro, 2000 and Gray & Rieger, 1971) and their greater relative abundance may also be indicative of organic pollution, filling a similar niche to the 2B nematodes. Nematodes are also assumed to be quite resistant to sediment organic enrichment and the resulting reducing conditions, and some species are permanently found in suboxic sediments (Mirto *et al.*, 2000).

A number of crustacean groups contributed to the similarity of the Group 1 samples in the analysis of the last two sample sets (Table 3.2) namely copepod nauplii, ostracods and harpacticoid copepods of the family Paramesochridae. This was also true for the Group 1 samples in the analysis of all the data sets where higher numbers of copepod nauplii, ostracods, Cylindropsyllidae and Paramesochridae played a major role in their similarity. Crustacean groups are generally considered to be the most sensitive groups to pollution impacts particularly the harpacticoid copepods (Sarkka, 1995; Carman *et al*, 2000 and Lee *et al*, 2001) and so Group 1 possibly consisted of samples being less influenced by the effluent.

In assessing the Nematode/Copepod ratio (N/C), the study of Sezela beach indicated a ratio of over 4.5 for the majority of samples that cluster as Group 2 (Figures 3.7 and 3.9). Warwick (1981) suggested that pollution might be indicated by N/C ratios of around 40 for fine sediments and 10 for sands. These values are much lower than the values of over 100 suggested by Raffaelli & Mason (1981). **A**n impacted site could have a low N/C ratio due to both groups being in low abundance such as Station viS2 collected on the sixth sampling occasion. This station had low numbers of all the taxa with a mean total number of animals of 83.67 and a low N/C ratio of 2. But this is the only station that stands out in this manner.

From the regression analyses the N/C ratio had a significant negative correlation with mean sediment grain size and salinity (Table 3.10). This is as might be expected as according to McLachlan *et al.* (1981), proportions of nematodes decrease and harpacticoids increase with increasing particle size above the range of 0.2 to 0.9 mm. Following this general pattern he suggested that nematodes should disappear above a mean particle size of 1.34 mm and

harpacticoids should disappear below 0.07mm particle size. The average South African beach has a median particle diameter of 0.35 mm (or 0.285 mm if the coarsest southern Natal beaches are ignored) and nematodes and harpacticoids in sediments of median particle diameter of 0.33 mm should respectively make up 38 % of the meiofauna (McLachlan *et al.*, 1981). A negative correlation of the N/C ratio with salinity suggests greater numbers of harpacticoid copepods in response to less influence from freshwater runoff and the influence of the nearby estuaries.

Raffaelli (1987) discussed the variable behaviour of the N/C ratio in organic pollution studies. It was concluded that differences in the habitat requirements of nematodes, mesobenthic and epi-/endobenthic copepods affected the responses of these groups to organic pollution. It was found in the sublittoral environment that epi-benthic copepods sometimes increase in response to organic pollution while nematodes decrease. The first studies of the N/C ratio by Raffaelli and Mason (1981) were done on sandy beaches where they had positive results and therefore this tool may be appropriate for this environment. Here one is dealing mainly with mesobenthic copepods and organic pollution is rarely severe enough on high energy beaches to produce a significant decline in nematodes (Raffaelli, 1987). The results for this study had some merit in terms of the use of the N/C ratio but in situations where both nematodes and harpacticoids ie total meiofauna (Coull & Palmer, 1984; Sandulli & De Nicola Giudici, 1989; Danovaro *et al.*, 1995; Lee *et al.*, 2001 and Ansari & Ingole, 2002) had been reduced by adverse conditions the ratio could not reveal the impact such as at Station viS2.

From an ANOVA of the Nematode/Copepod ratios, between all the stations, Stations S4 and S3 were found to be significantly different to Stations S2, S0 and N1 with Station S4 also being significantly different to Station S1. The lower ratios at the stations further away from the effluent outfall possibly indicated less impact from the effluent at these stations (Figure 3.11).

The BIO-ENV procedure that relates environmental variables to the observed biological pattern revealed that a combination of sediment grain size, dissolved oxygen and salinity contributed the most to the community patterns observed from the MDS plots in Figures 3.8 and 3.10. However the correlation between these measured variables and the biotic data was not very strong with $P = 0.1$.

Sediment grain size is one of the main factors influencing the distribution of meiofauna (Gheskiere *et al.*, 2005b). Meiofauna distribution is related to the degree of drainage and oxygenation of the sediment and abundance drops off drastically in reduced layers of the sediment (McLachlan & Erasmus, 1983). Therefore larger particle size would be expected to correlate with increased oxygen concentrations. From the regression analyses numbers of meiofauna taxa showed a significant positive correlated with dissolved oxygen and a significant negative correlation with COD in the sediments (Table 3.20) signifying higher diversity in less enriched and higher oxygenated sediments. At Sezela beach the mean particle size ranged from 0.26 mm to 1.23 mm. In sands above 0.2 mm the meiofauna is usually entirely interstitial and as most open sandy beaches have grain sizes in the range 0.2 to 0.5 mm, interstitial nematodes and harpacticoids are almost always dominant (McLachlan & Erasmus, 1983). Sezela beach tended towards the coarse side of the range and therefore the correlation of sediment grain size with dissolved oxygen would not be expected to have a marked impact on the meiofauna unless some other unmeasured variable was contributing to the biological community changes.

Salinity variations as a factor in influencing the observed biological pattern probably resulted from the proximity of the three estuaries and storm water drain in the area which were possibly adding to the observed variability in the meiofauna communities on the beach. Benthic communities in brackish water have lower densities and fewer species than either pure marine or pure freshwater communities (Gheskiere *et al.*, 2005b) and from the regression

analyses numbers of taxa showed a significant positive correlation with salinity (Table 3.20). Other variables such as enrichment, insecticides from farmlands etc. could also lead to toxicity and possibly lower oxygen levels.

Rodriguez *et al.* (2003) states that the concentration of interstitial oxygen is one of the most relevant physical factors affecting the presence of meiofauna. The crustacean taxa had greatly reduced average abundances in Group 2 samples. They are known to be highly mobile and normally occur in well oxygenated environments and generally appear to be highly sensitive to hypoxic conditions, surviving only a few hours in oxygen-deficient water (Josefson & Widbom, 1988). However, the Mdesingane river is opposite Station S4 which was shown to be very stable with high numbers of all taxa, especially crustacean groups throughout the study period. Station S4 was the greatest distance from the effluent discharge (1.5 km) suggesting that the effluent possibly played a significant role in the variability of the meiofauna communities at the other stations. However, as explained by Clarke & Warwick, (1994), linking the above variables to the biological pattern cannot demonstrate for certain that those variables are causing the observed biological pattern. Causality is only ever demonstrable by manipulative field or laboratory experiments, since the real causal variables, affecting the biology, may not have been measured but be strongly correlated with one or more of the variables which were measured.

Due to the lower diversity of crustacean groups and lower average abundance observed in the Group 2 samples in the CLUSTER analyses, it was concluded that this group possibly represents samples negatively impacted by the effluent. However the BIO-ENV procedure weakly linked (P = 0.1) grain size, dissolved oxygen and salinity as the main measured variables responsible for the observed biological pattern. The salinity aspect suggested other possible sources of pollutants such as organic enrichment from the nearby estuaries and the storm water drain located opposite Station N1. Enriched sites typically result in a depletion of oxygen which, if not replenished, can cause a crash in the population, particularly the crustaceans. Increased N/C ratios coincided mostly with the Group 2 samples from the CLUSTER analyses except were reduction in total meiofauna (nematodes and copepods) reduced the ability of the ratio to predict any negative effects.

The notion that anthropogenic environmental variables may induce community responses at a higher taxonomic level than natural environmental variables is worth further exploration, as it could lead to the solution of one of the major problems of interpretation of benthic community data in respect to pollution effects (Warwick, 1988). The effects of the effluent become confounded by beach dynamics (Ansari & Ingole, 2002) especially at a high energy beach like Sezela with a constant alteration of wind direction. Several investigators have envisioned meiofauna variability to be caused by a continuing disturbance/recolonization process (Murrell & Fleeger, 1989). On the KwaZulu-Natal coast the wind directions alternate between north east and south west resulting in periodically alternating along shore currents which would spread the effluent north and south along the beach from the source of the outlet. It appeared that the effluent was impacting at Station S0 during periods of discharge as the pipe is relatively short and effluent was observed being pushed back by the surf onto the beach at Station S0 which is located between two ridges of rock that run into the sea thus concentrating the effluent between them. All the stations showed indications of possibly being influenced by the effluent at some stage except for Station S4 which was the greatest distance (1.5 km) from the discharge point. Here the meiofauna community remained very stable throughout the study period. From this study, increased numbers of turbellarians and sarcomastigophorans appears to be indicative of disturbance and reduced oxygen appears to be the main chemical factor influencing the meiofauna.

Lack of sample replication during the first five sampling occasions could lead to reduced confidence in the validity of the above conclusions and future studies should include replication on all sampling occasions. Increased sampling frequency would also improve the

determination of seasonal variability and total organic content and chlorophyll-a concentrations should be measured in the sediments to support conclusions made about enrichment and oxygen depletion.

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**APPENDIX 1: Meiofauna Counts.**

Table A1.1
Sezela beach meiofauna per 200 $cm^3$ sediment sample 4th July 2000

| | N3 | N2 | N1 | S0 | S1a | S1b | S1c | S1d | S2 | S3 | S4 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Turbellaria | 4 | 4 | 7 | 20 | 60 | 62 | 68 | 43 | 128 | 64 | 48 |
| Nematoda 1A | 32 | 146 | 16 | 4 | 14 | 35 | 27 | 28 | 50 | 72 | 137 |
| Nematoda 1B | 12 | 28 | 9 | 7 | | 4 | 4 | 12 | | | 4 |
| Nematoda 2A | | 27 | 13 | 4 | 14 | 12 | 7 | 12 | 8 | | 4 |
| Nematoda 2B | | 4 | 8 | 6 | 4 | 10 | 11 | 8 | 21 | 20 | 46 |
| Rotifera | | | 2 | | | | | | 4 | | |
| Gastrotricha | 2 | 2 | 6 | | | | | | 6 | 38 | 42 |
| Kinorhyncha | | | | | | | | | 4 | | |
| Annelida: | | | | | | | | | | | |
| Pisionidae | 20 | | | | | | | | | | |
| Protodrilidae | 24 | | | 2 | 2 | 4 | | | | 11 | 52 |
| Nerillidae | 6 | 32 | | | | | | | | | |
| Hesionidae | | | | | | | | | | | |
| Syllidae | | | | | | | | | | | |
| Saccocirridae | | | | | | | | | | 101 | |
| polygordiidae | | | | | | | | | | | |
| Dynophilidae | | | | | | | | | | | |
| Oligochaeta | | | | | | | | | | | |
| Tardigrada | | | 8 | | 2 | | | | | | 6 |
| Acarina | 2 | 1 | | 2 | | | | | | 15 | 8 |
| Ostracoda | 176 | | 3 | | 2 | | | 4 | | | 84 |
| Copepod nauplii | 36 | 16 | | | 2 | | | | 4 | 28 | 240 |
| Harpacticoida: | | | | | | | | | | | |
| Peltidiidae | | | | | | | | | | | |
| Laophontidae | | | | | | | | | | | |
| Cylindropsyllidae | | | | | 4 | 4 | 2 | 4 | | 73 | 160 |
| Ectinosomatidae | | | | | | | | | | | |
| Tachidiidae | | | | | | | | | | | |
| Tisbidae | 148 | 8 | | | | | | | | | 2 |
| Paramesochridae | 12 | 2 | | | | | | | | 80 | 104 |
| Isopoda | | | | | | | | | | | 50 |
| Sarcomastigophora | | 72 | 4 | | | | | | 6 | | 8 |
| Total numbers | 474 | 342 | 76 | 45 | 104 | 131 | 119 | 111 | 231 | 502 | 995 |
| Total No. of Taxa | 12 | 12 | 10 | 7 | 9 | 7 | 6 | 7 | 9 | 10 | 16 |

Table A1.2
Sezela beach meiofauna per 200 $cm^3$ sediment sample 30th August 2000

| | N3 | N2 | N1 | S0 | S1a | S1b | S1c | S1d | S2 | S3 | S4 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Turbellaria | 24 | 110 | 86 | 28 | 62 | 10 | 8 | 42 | 100 | 103 | 16 |
| Nematoda 1A | 38 | 330 | 106 | 43 | 51 | 54 | 54 | 22 | 30 | 236 | 117 |
| Nematoda 1B | 22 | 156 | 68 | 4 | | | | | | 12 | 12 |
| Nematoda 2A | 15 | 128 | 44 | 4 | | 4 | 2 | 4 | 2 | 28 | 20 |
| Nematoda 2B | 3 | 28 | 51 | 55 | 148 | 102 | 286 | 380 | 32 | 36 | 42 |
| Rotifera | 2 | | | | | | | | | | |
| Gastrotricha | | 112 | | | | | 2 | 2 | | 26 | 74 |
| Kinorhyncha | | | | | | | | | | | |
| Annelida: | | | | | | | | | | | |
| Pisionidae | | | | | | | | | | | |
| Protodrilidae | | | | 14 | | | | 2 | | | |
| Nerillidae | 6 | 100 | | | | | | | | | |
| Hesionidae | 4 | | | | | | | | | | |
| Syllidae | | 8 | | | | | | | | | |
| Saccocirridae | | 12 | 12 | | | | | | | 43 | 2 |
| polygordiidae | | | | | | | | | | | 2 |
| Dynophilidae | | | | | | | | | | | |
| Oligochaeta | 2 | 64 | | | | 8 | | | 2 | | |
| Tardigrada | | 16 | 32 | | | | | | 2 | | 6 |
| Acarina | | 9 | | | | | | | | 8 | 4 |
| Ostracoda | | | 2 | | | 2 | | | | | 9 |
| Copepod nauplii | | 20 | 9 | 5 | 8 | 40 | 4 | 4 | | 132 | 103 |
| Harpacticoida: | | | | | | | | | | | |
| Peltidiidae | | | | | | | | | | | |
| Laophontidae | | | | | | | | | | | |
| Cylindropsyllidae | 2 | 2 | 2 | | | | | 16 | | 78 | 121 |
| Ectinosomatidae | | | | | | | | | | 4 | |
| Tachidiidae | | | | | | | | | | 36 | 53 |
| Tisbidae | | | | | | | | | | | |
| Paramesochridae | | | 4 | 6 | 14 | 16 | 80 | 100 | 2 | 108 | 168 |
| Isopoda | | | | | 2 | | | | | 26 | 74 |
| Sarcomastigophora | 28 | 304 | 67 | 4 | 4 | | | | | | 2 |
| Total numbers | 146 | 1399 | 483 | 163 | 289 | 236 | 436 | 572 | 170 | 876 | 825 |
| Total No. of Taxa | 11 | 15 | 12 | 9 | 7 | 8 | 7 | 9 | 7 | 14 | 17 |

Table A1.3
Sezela beach meiofauna per 200 $cm^3$ sediment sample 13th December 2000

| | N3 | N2 | N1 | S0 | S1 | S2 | S3a | S3b | S3c | S3d | S4 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Turbellaria | 18 | 24 | 20 | 46 | 38 | 42 | 20 | 6 | 6 | 18 | 22 |
| Nematoda 1A | 28 | 142 | 100 | 52 | 56 | 52 | 116 | 48 | 42 | 40 | 62 |
| Nematoda 1B | | 8 | 16 | 20 | 4 | | | | 4 | 4 | |
| Nematoda 2A | 2 | 12 | | 28 | 4 | 6 | | 4 | 14 | 11 | |
| Nematoda 2B | 18 | | | 22 | 198 | 72 | 16 | 20 | 13 | 9 | 40 |
| Rotifera | | | | | | | 2 | | | | 2 |
| Gastrotricha | | 6 | | | | 2 | 152 | 64 | 38 | 4 | 122 |
| Kinorhyncha | | | | | | | | | | | |
| Annelida: | | | | | | | | | | | |
| Pisionidae | | | | | | 4 | | | | 4 | |
| Protodrilidae | | | | | | | | | 4 | 14 | |
| Nerillidae | | 4 | | | | | | | | | |
| Hesionidae | | | | | | | | | | | |
| Syllidae | | | | | | | | | | | |
| Saccocirridae | | | | 2 | | 4 | 4 | 19 | | | 14 |
| polygordiidae | | | | | | | | | | | |
| Dynophilidae | | | | | | | 8 | | | | |
| Oligochaeta | | 2 | 16 | 2 | | | | | | | 4 |
| Tardigrada | | | | | | | | | | | |
| Acarina | | | | | | | 6 | 4 | 10 | 2 | |
| Ostracoda | | | | | | 2 | 62 | 57 | 28 | 90 | 812 |
| Copepod nauplii | 2 | | | | | 16 | 86 | 41 | 141 | 38 | 86 |
| Harpacticoida: | | | | | | | | | | | |
| Peltidiidae | | | | | | | | | | | |
| Laophontidae | | | | | | | | | | | |
| Cylindropsyllidae | | 2 | | | | 26 | 120 | 115 | 80 | 40 | 120 |
| Ectinosomatidae | | | | | | | | 25 | 12 | 28 | |
| Tachidiidae | | | | | | | 32 | 36 | 96 | 16 | 40 |
| Tisbidae | | | | | | | 16 | 64 | 104 | 76 | |
| Paramesochridae | | 2 | | | | 2 | 52 | 10 | 64 | 32 | 128 |
| Isopoda | | | | | | | | | | | 8 |
| Sarcomastigophora | 2 | 72 | | | | | 4 | | | | 6 |
| Total numbers | 70 | 274 | 152 | 172 | 300 | 228 | 696 | 513 | 656 | 426 | 1466 |
| Total No. of Taxa | 6 | 10 | 4 | 7 | 5 | 11 | 15 | 14 | 15 | 16 | 14 |

Table A1.4
Sezela beach meiofauna per 200 $cm^3$ sediment sample 26th January 2001

| | N3 | N2 | N1a | N1b | N1c | N1d | S0 | S1 | S2 | S3 | S4 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Turbellaria | 4 | 8 | 20 | 21 | 8 | 5 | 112 | 22 | 44 | 16 | 20 |
| Nematoda 1A | 5 | 158 | 53 | 126 | 85 | 81 | 12 | 8 | 20 | 40 | 80 |
| Nematoda 1B | 4 | 5 | 23 | 36 | 31 | 14 | 9 | | | 14 | 52 |
| Nematoda 2A | 6 | 3 | 30 | 65 | 50 | 18 | 11 | 4 | 2 | 19 | 5 |
| Nematoda 2B | 15 | 40 | 105 | 136 | 99 | 103 | 74 | 150 | 111 | 11 | 29 |
| Rotifera | 61 | | | | | 2 | | | | | 4 |
| Gastrotricha | | | 6 | 19 | 10 | 17 | 6 | | | | 21 |
| Kinorhyncha | | | | | | | | | | | |
| Annelida: | | | | | | | | | | | |
| Pisionidae | 5 | 11 | | | 4 | 2 | | | | | |
| Protodrilidae | 8 | 12 | | | | | | | | 2 | 25 |
| Nerillidae | | | 2 | | | 2 | | | | | 2 |
| Hesionidae | | | | | | | | | | | |
| Syllidae | | | | | | | | | | | |
| Saccocirridae | | | | | | | 2 | 25 | | 6 | |
| polygordiidae | | | | | | | | | | | |
| Dynophilidae | | | | | | | | | | | 10 |
| Oligochaeta | | 182 | 6 | 9 | 1 | 13 | | | 3 | | |
| Tardigrada | | | | | | | | | | | |
| Acarina | | | 2 | 10 | | | | | | | |
| Ostracoda | | | 6 | 4 | 7 | 15 | 2 | | | 5 | 1251 |
| Copepod nauplii | 8 | 118 | 2 | 8 | 21 | 7 | | | | 26 | 86 |
| Harpacticoida: | | | | | | | | | | | |
| Peltidiidae | | | | | | | | | | | |
| Laophontidae | | | | | | | | | | | |
| Cylindropsyllidae | | 44 | | 4 | 4 | | | 14 | | 4 | 30 |
| Ectinosomatidae | 4 | 13 | | | | | | | | | |
| Tachidiidae | | 89 | | | | | | | | | 25 |
| Tisbidae | | 113 | 10 | 28 | 26 | 6 | | | | | |
| Paramesochridae | | 25 | | 2 | 3 | 2 | | | | 71 | 48 |
| Isopoda | | | | | | | | | | | 11 |
| Sarcomastigophora | 6 | 7 | 2 | 10 | 6 | 12 | | 4 | | 5 | |
| Total numbers | 126 | 828 | 267 | 478 | 355 | 299 | 228 | 227 | 180 | 219 | 1699 |
| Total No. of Taxa | 11 | 15 | 13 | 14 | 14 | 15 | 8 | 7 | 5 | 12 | 16 |

Table A1.5
Sezela beach meiofauna per 200 $cm^3$ sediment sample 8th March 2001

| | N3a | N3b | N3c | N3d | N2 | N1 | S0 | S1 | S2 | S3 | S4 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Turbellaria | 5 | 8 | 4 | 8 | 4 | 21 | 71 | 20 | | 24 | 10 |
| Nematoda 1A | 32 | 22 | 16 | 42 | 13 | 182 | 184 | 303 | 85 | 62 | 22 |
| Nematoda 1B | 14 | 14 | 8 | 14 | 22 | 121 | 94 | 46 | 8 | 39 | 28 |
| Nematoda 2A | 4 | 10 | 10 | 10 | 5 | 24 | 35 | 45 | 6 | 12 | 4 |
| Nematoda 2B | 66 | 41 | 59 | 79 | 16 | 41 | 39 | 145 | 196 | 108 | 11 |
| Rotifera | | | 2 | 2 | 3 | | | | 4 | 2 | |
| Gastrotricha | | | | | | | 4 | 5 | | | 10 |
| Kinorhyncha | | | | | | | | | | | |
| Annelida: | | | | | | | | | | | |
| Pisionidae | | | | | 4 | | | 6 | | | |
| Protodrilidae | 4 | | | 4 | | 6 | | 19 | | 22 | 16 |
| Nerillidae | | | | | | 1 | 4 | | | | 3 |
| Hesionidae | | | | | | | | | | | 6 |
| Syllidae | | | | | | | | | | | |
| Saccocirridae | | | | | | | | | | | |
| polygordiidae | 12 | 20 | 18 | 12 | 11 | | | | | | |
| Dynophilidae | | | | | | | | | | | |
| Oligochaeta | | | | | | 16 | 2 | | | | 4 |
| Tardigrada | 4 | | | | | | | | | | |
| Acarina | 6 | | 14 | 7 | | 4 | | 11 | 4 | 15 | |
| Ostracoda | 55 | 23 | 46 | 30 | 63 | 31 | 18 | 4 | 22 | 1220 | 1336 |
| Copepod nauplii | | | 12 | 9 | 156 | 30 | 19 | 8 | | 85 | 29 |
| Harpacticoida: | | | | | | | | | | | |
| Peltidiidae | | | | | | | | | | | |
| Laophontidae | 25 | 2 | 6 | 8 | 12 | | | | | | 16 |
| Cylindropsyllidae | 16 | 8 | 4 | | 65 | | | | | 281 | 128 |
| Ectinosomatidae | 44 | | 10 | 44 | | | 2 | | | | |
| Tachidiidae | | | | | | | | | | | |
| Tisbidae | | 2 | | | 32 | 10 | 6 | 4 | | 4 | 9 |
| Paramesochridae | | | | | 28 | 11 | 13 | 14 | 2 | 8 | 4 |
| Isopoda | | | | | | | | | | 28 | 15 |
| Sarcomastigophora | | | | | | 30 | | 2 | 2 | | |
| Total numbers | 287 | 150 | 209 | 269 | 434 | 528 | 491 | 632 | 329 | 1910 | 1651 |
| Total No. of Taxa | 13 | 10 | 13 | 13 | 14 | 14 | 13 | 14 | 9 | 14 | 17 |

Table A1.6
Sezela beach meiofauna per 200 $cm^3$ sediment sample 9th April 2001

| | N3a | N3b | N3c | N2a | N2b | N2c | N1a | N1b | N1c | S0a | S0b | S0c | S0d | S1a | S1b | S1c | S2a | S2b | S2c | S3a | S3b | S3c | S4a | S4b | S4c |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Turbellaria | 4 | 6 | 6 | 5 | 4 | 8 | 16 | 4 | 20 | 44 | 32 | 40 | 34 | 104 | 90 | 66 | 8 | 8 | 14 | 12 | 8 | 34 | 2 | 16 | 4 |
| Nematoda 1A | 89 | 22 | 26 | 112 | 60 | 12 | 44 | 20 | 45 | 213 | 192 | 145 | 216 | 74 | 79 | 179 | 8 | 6 | 28 | 44 | 90 | 120 | 44 | 125 | 62 |
| Nematoda 1B | 88 | 7 | 20 | 45 | 30 | 29 | 48 | 50 | 60 | 229 | 240 | 212 | 241 | 70 | 42 | 66 | 2 | 10 | | 112 | 139 | 159 | 21 | 16 | 24 |
| Nematoda 2A | 23 | | 6 | 35 | | 6 | 32 | 12 | 43 | 200 | 6 | 79 | 292 | 45 | 62 | 100 | 9 | | 2 | 5 | 5 | 19 | 8 | 12 | |
| Nematoda 2B | 39 | 35 | 80 | 5 | 8 | 6 | 21 | 20 | 22 | 9 | | 20 | 28 | 86 | 120 | 126 | 18 | 14 | 18 | 66 | 90 | 154 | 10 | 21 | 10 |
| Rotifera | | | | | | | | | | | | | | | | | | | | | | | | | |
| Gastrotricha | | | | | 8 | 3 | 24 | 62 | 46 | | | 8 | 2 | 59 | 39 | 40 | 5 | 2 | | 52 | 32 | 126 | 6 | 24 | 38 |
| Kinorhyncha | | | | | | | 4 | | | | | | | | | | | | | 8 | | | | | |
| Annelida: | | | | | | | | | | | | | | | | | | | | | | | | | |
| Pisionidae | 4 | | | | 4 | 4 | | | | | | 6 | 4 | | | | | | | | | | | | |
| Protodrilidae | 5 | | | | | 2 | | | 15 | | | | | | | | | | | | | | | | |
| Nerillidae | | | | 4 | | 2 | 4 | 10 | 8 | | 4 | 8 | 8 | | 2 | | | | | | 3 | | 2 | 6 | 2 |
| Hesionidae | | | | | | | | | 2 | | | | | | | | | | | | | | | | |
| Syllidae | | | | | | | | | | | | | | | | | | | | | | | | | |
| Saccocirridae | 6 | 2 | 6 | 4 | 4 | 15 | 71 | 51 | 56 | 12 | 12 | 17 | 33 | 1296 | 1710 | 1120 | | | | 56 | 25 | 70 | 25 | 50 | 62 |
| polygordiidae | | | | | | | | | | | | | | | | | | | | | | | | | |
| Dynophilidae | | | | | | | | | | | | | | | | | | | | | | | | | |
| Oligochaeta | | | | 4 | | 5 | | | | 14 | 2 | | | 24 | 16 | | 4 | 4 | | | 6 | | | | |
| Tardigrada | 4 | | | | 4 | | | | | 4 | 5 | | | | | 2 | | | | | | | | | |
| Acarina | 2 | 3 | 7 | | | | 16 | 18 | 32 | 4 | 6 | 6 | 16 | 25 | 25 | 33 | 4 | 2 | | 34 | 15 | 50 | 18 | 32 | 54 |
| Ostracoda | 48 | 31 | 68 | 32 | 80 | 60 | 60 | 50 | 25 | 30 | 35 | 31 | 18 | 16 | 13 | 2 | 4 | 10 | 2 | 90 | 74 | 72 | 425 | 856 | 568 |
| Copepod nauplii | 15 | 7 | 12 | 65 | | 88 | 136 | 92 | 54 | 41 | 54 | 70 | 70 | 56 | 52 | 120 | 11 | | | 101 | 54 | 200 | 30 | 108 | 146 |
| Harpacticoida: | | | | | | | | | | | | | | | | | | | | | | | | | |
| Peltidiidae | | | | | | | | | | | | | | | | | | | | | | | | | |
| Laophontidae | 4 | | | | | 2 | | | | | | | | | | | | | | | | | | | |
| Cylindropsyllidae | 3 | 5 | 8 | | | 12 | | 41 | | 3 | 2 | | | 65 | 14 | 28 | 2 | 44 | 2 | 48 | 16 | 80 | 41 | 149 | 192 |
| Ectinosomatidae | | | | | 4 | | 8 | | | | | | | 16 | 12 | | | | | | 3 | | | | |
| Tachidiidae | | | | | | | | | | | | | | | | | | | | | 12 | 36 | | | |
| Tisbidae | | | | 24 | 20 | 28 | 30 | 18 | 36 | 21 | 13 | 20 | 21 | 8 | | | 5 | 4 | | | | | 3 | | |
| Paramesochridae | 2 | | | 20 | 20 | 4 | 159 | 134 | 212 | 168 | 170 | 167 | 205 | 160 | 56 | 56 | | | | 124 | 54 | 92 | 15 | 31 | 12 |
| Isopoda | | 2 | | | | | | | | | | | | | | | | | | 6 | 6 | 2 | 6 | 24 | 30 |
| Sarcomastigophora | | | | | | | 4 | | | | | | 6 | | | | | | | | | | | | 2 |
| Total numbers | 336 | 120 | 239 | 355 | 246 | 286 | 677 | 582 | 676 | 992 | 773 | 829 | 1194 | 2104 | 2332 | 1938 | 80 | 104 | 66 | 758 | 632 | 1214 | 656 | 1470 | 1206 |
| Total No. of taxa | 15 | 10 | 10 | 12 | 12 | 17 | 16 | 14 | 15 | 14 | 14 | 14 | 15 | 15 | 15 | 13 | 12 | 10 | 6 | 14 | 17 | 14 | 15 | 14 | 14 |

Table A1.7
Sezela beach meiofauna per 200 $cm^3$ sediment sample 2nd January 2002

| | N3a | N3b | N3c | N2a | N2b | N2c | N1a | N1b | N1c | S0a | S0b | S0c | S0d | S1a | S1b | S1c | S2a | S2b | S2c | S3a | S3b | S3c | S4a | S4b | S4c |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Turbellaria | 22 | 8 | 13 | 50 | 48 | 20 | | | | 40 | 96 | 38 | 57 | 46 | 36 | 40 | 50 | 60 | 75 | 28 | 8 | 20 | 46 | 64 | 32 |
| Nematoda 1A | 62 | 32 | 40 | 78 | 299 | 164 | 14 | 16 | 20 | 26 | 39 | 39 | 40 | 34 | 30 | 80 | 10 | 44 | 16 | 54 | 60 | 14 | 84 | 100 | 56 |
| Nematoda 1B | 31 | 30 | 40 | 64 | 200 | 212 | 18 | 36 | 30 | 46 | 30 | 23 | 74 | 134 | 85 | 212 | 39 | 35 | 52 | 26 | 41 | 65 | 140 | 104 | 180 |
| Nematoda 2A | 32 | 25 | 21 | 22 | 54 | 60 | 9 | 20 | 32 | 30 | 31 | 30 | 58 | 16 | 26 | 30 | 42 | 16 | 28 | 52 | 21 | 56 | 80 | 105 | 112 |
| Nematoda 2B | 50 | 26 | 9 | 12 | 14 | 12 | 5 | 4 | | 16 | 18 | 2 | 26 | 54 | 60 | 70 | 16 | 8 | 8 | 6 | | 15 | 20 | 56 | 28 |
| Rotifera | 21 | | | | | | | | | | | | | | | | | | | | | | | | 4 |
| Gastrotricha | 11 | | | 126 | 240 | 230 | | | | 3 | | | | 61 | 126 | 400 | | | | | | | 28 | 25 | 116 |
| Kinorhyncha | | | | | | | | | | | | | | | | | | 4 | | | | | | | |
| Annelida: | | | | | | | | | | | | | | | | | | | | | | | | | |
| Pisionidae | | 12 | | | | | | | | | | | | | 6 | | | | | | | | | | |
| Protodrilidae | | | | | | | | | | | | | | | | | | | | | | | | | |
| Nerillidae | 32 | 12 | 4 | 315 | 246 | 413 | 4 | | 12 | | | | | 6 | | | | | | | | | | | |
| Hesionidae | | | | 15 | 50 | 12 | | | | | | | | | | | | | | | | | | | |
| Syllidae | | | | | | | | | | | | | | | | | | | | | | | | | |
| Saccocirridae | | | | 3 | | | 125 | 101 | 162 | | | 2 | 6 | 35 | 15 | 25 | | 5 | | | | | 553 | 400 | 760 |
| polygordiidae | | | 4 | 3 | | | | | | | | | | | | | | | | | | | | | |
| Dynophilidae | | | | | | | | | | | | | | | | | | | | | | | | | |
| Oligochaeta | 50 | 49 | 51 | 104 | 136 | 90 | | | | | | | | 380 | 141 | 360 | | | | | | | | | |
| Tardigrada | 8 | | | | | | | | | | 2 | 4 | | | | | 6 | 4 | | 6 | | | 14 | 5 | 12 |
| Acarina | 25 | 4 | 25 | 16 | 64 | 21 | 48 | 60 | 52 | 2 | 10 | 5 | 2 | 42 | 5 | 30 | 2 | | | | | 12 | 6 | 5 | 13 |
| Ostracoda | 132 | 108 | 170 | 69 | 109 | 160 | 6 | 12 | 16 | 2 | 3 | | 2 | 213 | 59 | 351 | | | 4 | | | 9 | 121 | 209 | 140 |
| Copepod nauplii | 288 | 3199 | 876 | 150 | 150 | 400 | 8 | 8 | 4 | 3 | 2 | | 2 | 756 | 425 | 608 | | | | 2 | | | 206 | 280 | 232 |
| Harpacticoida: | | | | | | | | | | | | | | | | | | | | | | | | | |
| Peltidiidae | | | | | 2 | 2 | | | | | | | | | | | | | | | | | | | |
| Laophontidae | | | | | | | | | | | | | | | | | | | | | | | | | |
| Cylindropsyllidae | 244 | 861 | 432 | | | 40 | | | | | 2 | | | 804 | 47 | 264 | 2 | | | 3 | 4 | | 90 | 128 | 193 |
| Ectinosomatidae | 4 | | | | | | | | | | | | | 6 | 8 | | | | | | | | | | |
| Tachidiidae | | | | | | | | | | | | | | | | | | | | | | | 12 | 96 | 15 |
| Tisbidae | | 32 | 36 | 99 | 56 | 101 | 3 | | | 2 | | | | 25 | 8 | 24 | | | | | | | | | |
| Paramesochridae | 12 | 16 | 12 | 8 | 16 | 8 | 2 | | | 2 | | | | 36 | 8 | 32 | | | | 5 | 4 | | 342 | 832 | 913 |
| Isopoda | | | | | | | | | | | | | | | | | | | | | | | | | |
| Sarcomastigophora | 3 | | | 20 | 241 | 170 | 4 | | 12 | 136 | 18 | 121 | 114 | | 6 | 6 | 48 | 31 | 55 | 2 | 3 | 4 | | | |
| Total numbers | 1027 | 4414 | 1733 | 1154 | 1925 | 2115 | 246 | 257 | 340 | 308 | 251 | 264 | 381 | 2648 | 1091 | 2532 | 215 | 207 | 238 | 184 | 141 | 195 | 1742 | 2409 | 2806 |
| Total No. of Taxa | 17 | 14 | 14 | 17 | 16 | 17 | 12 | 8 | 9 | 12 | 11 | 9 | 10 | 16 | 17 | 15 | 9 | 9 | 7 | 10 | 7 | 8 | 14 | 14 | 15 |

**Appendix 2: Chemistry and sediment grain size results.**

Table A2.1
Chemical analysis of interstitial water samples taken on 4th July 2000

| Station | CODmn mg $l^{-1}$ | Kjel-N µg $l^{-1}$ | DO mg $l^{-1}$ | pH | Salinity ‰ | Ammonia µg $l^{-1}$ |
|---|---|---|---|---|---|---|
| N3 | 0.14 | 75 | 7.31 | 8 | 28.6 | 69 |
| N2 | 0.11 | 127 | 3.42 | 7.8 | 28 | 69 |
| N1 | 0.16 | 370 | 1.83 | 7.8 | 28.8 | 89 |
| S0 | 0.26 | 394 | 1.32 | 7.4 | 29.1 | 30 |
| S1 | 0.16 | 204 | 3.21 | 7.5 | 26.9 | 82 |
| S2 | 0.12 | 58 | 2.7 | 7.8 | 20.7 | 106 |
| S3 | 0.12 | 396 | 5.51 | 7.9 | 28.4 | 63 |
| S4 | 0.2 | 749 | 6.72 | 8 | 28.5 | 142 |

Table A2.2
Chemical analysis of interstitial water samples taken on 30th August 2000

| Station | CODmn mg $l^{-1}$ | Kjel-N µg $l^{-1}$ | DO mg $l^{-1}$ | pH | Salinity ‰ | Ammonia µg $l^{-1}$ |
|---|---|---|---|---|---|---|
| N3 | 0.45 | 402 | 4 | 8.1 | 31.2 | 79 |
| N2 | 0.41 | 204 | 4.11 | 7.6 | 19 | 63 |
| N1 | 0.37 | 186 | 4.32 | 7.6 | 20.8 | 59 |
| S0 | 1.22 | 2509 | 2.83 | 7.8 | 30.2 | 166 |
| S1 | 0.53 | 235 | 4.22 | 7.8 | 29 | 33 |
| S2 | 0.69 | 255 | 3.11 | 7.7 | 24.1 | 131 |
| S3 | 0.49 | 69 | 4.2 | 7.6 | 31.3 | 44 |
| S4 | 0.24 | 206 | 3.52 | 7.7 | 31 | 28 |

Table A2.3
Chemical analysis of interstitial water samples taken on 13th December 2000

| Station | CODmn mg $l^{-1}$ | Kjel-N µg $l^{-1}$ | DO mg $l^{-1}$ | pH | Salinity ‰ | Ammonia µg $l^{-1}$ |
|---|---|---|---|---|---|---|
| N3 | 0.47 | 560 | 2.57 | 7.7 | 35.1 | 35 |
| N2 | 0.98 | 456 | 5.04 | 7.4 | 32.5 | 130 |
| N1 | 0.54 | 311 | 5.23 | 7.9 | 35 | 11.8 |
| S0 | 0.4 | 310 | 7.8 | 7.4 | 34.8 | 35 |
| S1 | 0.69 | 457 | 7.9 | 7.9 | 34.9 | 35 |
| S2 | 0.4 | 433 | 8.39 | 7.9 | 34.9 | 106 |
| S3 | 0.54 | 268 | 9.38 | 7.9 | 35 | 11 |
| S4 | 0.76 | 426 | 8.69 | 7.7 | 33.6 | 134 |

Table A2.4
Chemical analysis of interstitial water samples taken on 26th January 2001

| Station | CODmn | Kjel-N | DO | pH | Salinity | Ammonia |
|---|---|---|---|---|---|---|
| | mg $l^{-1}$ | µg $l^{-1}$ | mg $l^{-1}$ | | ‰ | µg $l^{-1}$ |
| N3 | 0.98 | 228 | 4.56 | 7.8 | 33.7 | 34 |
| N2 | 0.98 | 289 | 4.28 | 7.7 | 33.3 | 178 |
| N1 | 0.75 | 267 | 4.6 | 7.8 | 31.6 | 51 |
| S0 | 1.28 | 47 | 4.46 | 7.6 | 29.4 | 414 |
| S1 | 0.75 | 352 | 3.81 | 7.8 | 34.6 | 27 |
| S2 | 0.6 | 211 | 5.25 | 7.9 | 34.4 | 41 |
| S3 | 1.2 | 246 | 6.05 | 7.9 | 25.6 | 31 |
| S4 | 1.65 | 418 | 4 | 8.1 | 34.8 | 24 |

Table A2.5
Chemical analysis of interstitial water samples taken on 8th March 2001

| Station | CODmn | Kjel-N | DO | pH | Salinity | Ammonia |
|---|---|---|---|---|---|---|
| | mg $l^{-1}$ | µg $l^{-1}$ | mg $l^{-1}$ | | ‰ | µg $l^{-1}$ |
| N3 | 0.8 | 170 | 6.02 | 7.1 | 34.7 | 36 |
| N2 | 0.66 | 186 | 3.89 | 7.5 | 27.5 | 29 |
| N1 | 0.8 | 186 | 3.15 | 7.7 | 32.4 | 129 |
| S0 | 1.15 | 444 | 2.87 | 7.1 | 34.4 | 626 |
| S1 | 1.07 | 216 | 3.43 | 6.9 | 34.6 | 365 |
| S2 | 0.88 | 178 | 3.7 | 7.8 | 34.2 | 215 |
| S3 | 1.01 | 123 | 5.56 | 7.8 | 34.6 | 34 |
| S4 | 0.78 | 394 | 5.09 | 7.7 | 34.5 | 70 |

Table A2.6
Chemical analysis of interstitial water samples taken on 9th April 2001

| Station | CODmn | Kjel-N | DO | pH | Salinity | Ammonia |
|---|---|---|---|---|---|---|
| | mg $l^{-1}$ | µg $l^{-1}$ | mg $l^{-1}$ | | ‰ | µg $l^{-1}$ |
| N3 | 0.38 | 435 | 7.9 | 7.9 | 35.3 | 190 |
| N2 | 0.25 | 369 | 8.1 | 7.4 | 35.3 | 98 |
| N1 | 0.3 | 493 | 4.44 | 7.5 | 35.3 | 129 |
| S0 | 0.46 | 357 | 5.83 | 7.7 | 35.2 | 98 |
| S1 | 0.38 | 562 | 4.74 | 7.6 | 35.2 | 251 |
| S2 | 0.38 | 871 | 8 | 7.9 | 35.1 | 631 |
| S3 | 0.21 | 834 | 5.51 | 7.5 | 35.2 | 606 |
| S4 | 0.13 | 196 | 7.8 | 7.9 | 35.3 | 67 |

Table A2.7
Chemical analysis of interstitial water samples taken on 2nd January 2002

| Station | CODmn mg $l^{-1}$ | Kjel-N µg $l^{-1}$ | DO mg $l^{-1}$ | pH | Salinity ‰ | Ammonia µg $l^{-1}$ |
|---|---|---|---|---|---|---|
| N3 | 0.22 | 677 | 5.85 | 8.1 | 35.3 | 37 |
| N2 | 1.17 | 1015 | 6.32 | 8 | 35.6 | 28.8 |
| N1 | 0.66 | 861 | 2.79 | 8 | 35.5 | 28.8 |
| S0 | 0.95 | 380 | 1.19 | 7.8 | 35.3 | 49.3 |
| S1 | 1.69 | 1079 | 6.3 | 8 | 35.3 | 28.8 |
| S2 | 0.51 | 636 | 1.89 | 7.7 | 19.4 | 53.5 |
| S3 | 0.51 | 2365 | 2.26 | 7.8 | 34.1 | 45.2 |
| S4 | 4.48 | 1766 | 5.45 | 7.9 | 35.3 | 251 |

Table A2.8
Chemical analyses of sediment samples taken on 4th July 2000

| Station | COD(mn) mg $g^{-1}$ | Kjel N µg $g^{-1}$ |
|---|---|---|
| N3 | 0.089 | 32.7 |
| N2 | 0.028 | 21.2 |
| N1 | 0.085 | 39.5 |
| S0 | 0.09 | 37 |
| S1 | 0.085 | 39.2 |
| S2 | 0.076 | 39.9 |
| S3 | 0.088 | 41.2 |
| S4 | 0.066 | 32.2 |

Table A2.9
Chemical analyses of sediment samples taken on 30th August 2000

| Station | COD(mn) mg $g^{-1}$ | Kjel N µg $g^{-1}$ |
|---|---|---|
| N3 | 0.051 | 23.5 |
| N2 | 0.044 | 31.2 |
| N1 | 0.057 | 39.7 |
| S0 | 0.064 | 38.6 |
| S1 | 0.064 | 48.4 |
| S2 | 0.059 | 26.1 |
| S3 | 0.064 | 43.4 |
| S4 | 0.04 | 23.3 |

Table A2.10
Chemical analyses of sediment samples taken on 13th December 2000

| Station | COD(mn) mg $g^{-1}$ | Kjel N µg $g^{-1}$ |
|---|---|---|
| N3 | 0.103 | 27.1 |
| N2 | 0.11 | 8.3 |
| N1 | 0.06 | 28.7 |
| S0 | 0.108 | 17.2 |
| S1 | 0.079 | 19.8 |
| S2 | 0.083 | 28.8 |
| S3 | 0.077 | 64.1 |
| S4 | 0.074 | <1.0 |

Table A2.11
Chemical analyses of sediment samples taken on 26th January 2001

| Station | COD(mn) mg $g^{-1}$ | Kjel N µg $g^{-1}$ |
|---|---|---|
| N3 | 0.036 | 27.1 |
| N2 | 0.042 | 24.8 |
| N1 | 0.027 | 33.4 |
| S0 | 0.036 | 30.8 |
| S1 | 0.036 | 36.1 |
| S2 | 0.027 | 23.2 |
| S3 | 0.018 | 25.9 |
| S4 | 0.028 | 25.7 |

Table A2.12
Chemical analyses of sediment samples taken on 8th March 2001

| Station | COD(mn) mg $g^{-1}$ | Kjel N µg $g^{-1}$ |
|---|---|---|
| N3 | 0.038 | 19.3 |
| N2 | 0.028 | 33.6 |
| N1 | 0.053 | 30.3 |
| S0 | 0.074 | 25.8 |
| S1 | 0.124 | 12.9 |
| S2 | 0.064 | 25.2 |
| S3 | 0.115 | 19.6 |
| S4 | 0.001 | 24.4 |

Table A2.13
Chemical analyses of sediment samples taken on 9th April 2001

| Station | COD(mn) mg $g^{-1}$ | Kjel N µg $g^{-1}$ |
|---|---|---|
| N3 | <0.01 | 40.5 |
| N2 | <0.01 | 47.0 |
| N1 | <0.01 | 21.8 |
| S0 | <0.01 | 26.5 |
| S1 | <0.01 | 146 |
| S2 | <0.01 | 23.6 |
| S3 | <0.01 | 17.1 |
| S4 | <0.01 | 16.1 |

Table A.14
Chemical analyses of sediment samples taken on 2nd January 2002

| Station | COD(mn) mg $g^{-1}$ | Kjel N µg $g^{-1}$ |
|---|---|---|
| N3 | 0.046 | 18.9 |
| N2 | 0.054 | 46.8 |
| N1 | 0.027 | 22.8 |
| S0 | 0.067 | 33.4 |
| S1 | 0.059 | 101 |
| S2 | 0.051 | 36.3 |
| S3 | 0.076 | 58.1 |
| S4 | 0.108 | 75.5 |

Table A2.15
Particle size distributions for sediment samples taken on 4th July 2000

| Station | %Gravel >2mm | %Very coarse 1-2mm | %Coarse 0.5-1mm | %Medium 0.25-0.5mm | %Fine 0.125-0.25mm | %Very fine 0.063-0.125 mm | %Mud <0.063 mm | Mean size mm |
|---|---|---|---|---|---|---|---|---|
| N3 | 7.04 | 58.52 | 25.26 | 6.21 | 2.97 | 0 | 0 | 1.23 |
| N2 | 8.21 | 40.12 | 37.79 | 12.02 | 1.8 | 0.06 | 0 | 0.98 |
| N1 | 0.96 | 5.08 | 13.66 | 39.64 | 40.19 | 0.47 | 0 | 0.31 |
| S0 | 0.36 | 3.64 | 20.7 | 48.68 | 26.38 | 0.24 | 0 | 0.35 |
| S1 | 0.47 | 17.34 | 33.02 | 35.09 | 13.98 | 0.11 | 0 | 0.52 |
| S2 | 0.04 | 8.55 | 29.33 | 40.88 | 21.04 | 0.16 | 0 | 0.42 |
| S3 | 1.1 | 0.6 | 8.69 | 41.3 | 47.81 | 0.5 | 0 | 0.26 |
| S4 | 2.3 | 55.88 | 19.9 | 21.02 | 0 | 1 | 0 | 0.86 |

Table A2.16
Particle size distributions for sediment samples taken on 30th August 2000

| Station | %Gravel >2mm | %Very coarse 1-2mm | %Coarse 0.5-1mm | %Medium 0.25-0.5mm | %Fine 0.125-0.25mm | %Very fine 0.063-0.125 mm | %Mud <0.063 mm | Mean size mm |
|---|---|---|---|---|---|---|---|---|
| N3 | 4 | 24.59 | 57.2 | 13.21 | 0.7 | 0.4 | 0 | 0.65 |
| N2 | 1.11 | 2.6 | 30.69 | 63.29 | 1.91 | 0.4 | 0 | 0.36 |
| N1 | 1.8 | 0.61 | 30.3 | 65.01 | 2 | 0.29 | 0 | 0.36 |
| S0 | 0 | 2.7 | 31.1 | 55 | 10.81 | 0.39 | 0 | 0.33 |
| S1 | 0.29 | 7.38 | 38 | 49.2 | 5.11 | 0.01 | 0 | 0.4 |
| S2 | 0.9 | 12.8 | 42.58 | 40.21 | 3.11 | 0.3 | 0 | 0.46 |
| S3 | 3.51 | 18.31 | 46.9 | 29.7 | 1.58 | 0 | 0 | 0.56 |
| S4 | 16.3 | 14.39 | 42.21 | 25.31 | 1.49 | 0.3 | 0 | 0.75 |

Table A2.17
Particle size distributions for sediment samples taken on 13th December 2000

| Station | %Gravel >2mm | %Very coarse 1-2mm | %Coarse 0.5-1mm | %Medium 0.25-0.5mm | %Fine 0.125-0.25mm | %Very fine 0.063-0.125 mm | %Mud <0.063 mm | Mean size mm |
|---|---|---|---|---|---|---|---|---|
| N3 | 4.01 | 21 | 50.9 | 22.7 | 0.79 | 0.51 | 0 | 0.6 |
| N2 | 5 | 30.89 | 52.5 | 10.71 | 0.7 | 0.3 | 0 | 0.7 |
| N1 | 1.01 | 28.4 | 70.29 | 0 | 0.6 | 0 | 0 | 0.66 |
| S0 | 0 | 4.21 | 62.2 | 33.11 | 1.49 | 0 | 0 | 0.44 |
| S1 | 1 | 10.79 | 53.51 | 33.1 | 1.5 | 0.1 | 0 | 0.48 |
| S2 | 3.01 | 29.69 | 58.3 | 9.1 | 0 | 0.4 | 0 | 0.67 |
| S3 | 5 | 29.68 | 54.12 | 10.8 | 0 | 0.6 | 0 | 0.69 |
| S4 | 1 | 12.79 | 73.31 | 11.4 | 0.5 | 1 | 0 | 0.67 |

Table A2.18
Particle size distributions for sediment samples taken on 26th January 2001

| Station | %Gravel >2mm | %Very coarse 1-2mm | %Coarse 0.5-1mm | %Medium 0.25-0.5mm | %Fine 0.125-0.25mm | %Very fine 0.063-0.125 mm | %Mud <0.063 mm | Mean size mm |
|---|---|---|---|---|---|---|---|---|
| N3 | 1.53 | 48.75 | 46.68 | 2.96 | 0.08 | 0 | 0 | 0.76 |
| N2 | 2.23 | 51.83 | 42.77 | 2.89 | 0.25 | 0.03 | 0 | 1.1 |
| N1 | 0.59 | 20.64 | 38.23 | 35.46 | 4.91 | 0.17 | 0 | 0.6 |
| S0 | 0.2 | 9.61 | 27.41 | 58.49 | 4.58 | 0.01 | 0 | 0.5 |
| S1 | 0.18 | 11.28 | 50.17 | 35.78 | 2.58 | 0 | 0 | 0.57 |
| S2 | 0.18 | 22.5 | 58.33 | 17.38 | 1.57 | 0.04 | 0 | 0.76 |
| S3 | 0.44 | 25.93 | 56.69 | 15.26 | 1.66 | 0.03 | 0 | 0.79 |
| S4 | 1.4 | 3.73 | 18.67 | 60.73 | 15.4 | 0.07 | 0 | 0.38 |

Table A2.19
Particle size distributions for sediment samples taken on 8th March 2001

| Station | %Gravel >2mm | %Very coarse 1-2mm | %Coarse 0.5-1mm | %Medium 0.25-0.5mm | %Fine 0.125-0.25mm | %Very fine 0.063-0.125 mm | %Mud <0.063 mm | Mean size mm |
|---|---|---|---|---|---|---|---|---|
| N3 | 0.28 | 15.07 | 58.64 | 25.1 | 0.99 | 0 | 0 | 0.64 |
| N2 | 6.64 | 31.78 | 34.31 | 25.21 | 2.06 | 0 | 0 | 0.82 |
| N1 | 0.28 | 18.76 | 62.15 | 16.87 | 1.89 | 0.06 | 0 | 0.72 |
| S0 | 0.28 | 11.16 | 56.1 | 30.67 | 1.71 | 0.08 | 0 | 0.55 |
| S1 | 1.81 | 14.07 | 38.75 | 39.68 | 5.62 | 0.06 | 0 | 0.55 |
| S2 | 0.63 | 13.15 | 42.21 | 38.07 | 5.9 | 0.04 | 0 | 0.55 |
| S3 | 4.14 | 29.01 | 43.45 | 20.49 | 2.87 | 0.04 | 0 | 0.79 |
| S4 | 6.2 | 26.01 | 43.1 | 20.69 | 3.96 | 0.04 | 0 | 0.78 |

Table A2.20
Particle size distributions for sediment samples taken on 9th April 2001

| Station | %Gravel >2mm | %Very coarse 1-2mm | %Coarse 0.5-1mm | %Medium 0.25-0.5mm | %Fine 0.125-0.25mm | %Very fine 0.063-0.125 mm | %Mud <0.063 mm | Mean size mm |
|---|---|---|---|---|---|---|---|---|
| N3 | 1.48 | 16.01 | 47.25 | 33.73 | 1.54 | 0 | 0 | 0.64 |
| N2 | 1.19 | 26.98 | 58.65 | 11.82 | 1.35 | 0 | 0 | 0.83 |
| N1 | 0.67 | 18.43 | 65.11 | 14.84 | 0.94 | 0 | 0 | 0.74 |
| S0 | 0 | 5.59 | 51.72 | 36.94 | 5.59 | 0.15 | 0 | 0.52 |
| S1 | 0.09 | 9.06 | 44.25 | 38.74 | 7.87 | 0 | 0 | 0.51 |
| S2 | 0 | 0.89 | 30.25 | 58.93 | 9.81 | 0.12 | 0 | 0.41 |
| S3 | 0.06 | 6.65 | 30.68 | 51.53 | 11.01 | 0.08 | 0 | 0.46 |
| S4 | 0 | 4.4 | 29.75 | 52.28 | 12.03 | 1.54 | 0 | 0.42 |

Table A2.21
Particle size distributions for sediment samples taken on 2nd January 2002

| Station | %Gravel >2mm | %Very coarse 1-2mm | %Coarse 0.5-1mm | %Medium 0.25-0.5mm | %Fine 0.125-0.25mm | %Very fine 0.063-0.125 mm | %Mud <0.063 mm | Mean size mm |
|---|---|---|---|---|---|---|---|---|
| N3 | 1.57 | 53.18 | 44.71 | 0.46 | 0.07 | 0 | 0 | 0.79 |
| N2 | 1.93 | 27.77 | 64.87 | 5.17 | 0.21 | 0.04 | 0 | 0.65 |
| N1 | 1.6 | 43.01 | 52.7 | 2.43 | 0.19 | 0.08 | 0 | 0.73 |
| S0 | 0.39 | 9.58 | 47.14 | 38.09 | 4.6 | 0.2 | 0 | 0.44 |
| S1 | 0.12 | 5.54 | 29.01 | 58.96 | 6.31 | 0.06 | 0 | 0.36 |
| S2 | 0.87 | 5.08 | 20.65 | 54.16 | 19 | 0.24 | 0 | 0.33 |
| S3 | 0.7 | 11.29 | 30.94 | 37.25 | 19.59 | 0.23 | 0 | 0.4 |
| S4 | 0.72 | 7.06 | 21.69 | 48.05 | 22.26 | 0.22 | 0 | 0.34 |