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Sarah Butler and Fiona Bird

June 2010





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#### Author(s):

Sarah Butler – PhD Student, La Trobe University

Fiona Bird – Senior Lecturer, La Trobe University

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### Monitoring the macroinvertebrates and soft sediments in the Marine National Parks in Western Port

Sarah Butler Fiona Bird

La Trobe University, Melbourne

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### EXECUTIVE SUMMARY

Western Port Bay in Victoria has three Marine National Parks (MNPs), Yaringa, French Island and Churchill Island MNPs. The Marine National Parks in Western Port were created to protect areas of seagrass beds, saltmarsh, mangroves, deep channels, soft-sediments and mudflats (Parks Victoria 2007). Specifically, Yaringa MNP protects intertidal mudflats, mangrove and saltmarsh areas; French Island MNP protects seagrass beds, intertidal mudflats, soft-sediment beds, some deep channels, mangroves and saltmarsh; and Churchill Island MNP protects intertidal mudflats, saltmarsh, seagrass beds and deep channels.

With the declaration of Marine National Parks in Victoria in 2002, a Strategic Management Plan was written for 2003-2010 (Parks Victoria 2003). This Strategic Management Plan outlined the importance of environmental research and monitoring in filling knowledge gaps about many facets of the MNPs in Victoria. In rocky shore Marine National Parks and Sanctuaries around Victoria, several long term monitoring programs have already been well established and provide useful information about the communities that exist in these areas (see Edmunds *et al.* 2004). The same level of monitoring has not been developed for soft-sediment habitats in MNPs in Victoria.

In the MNPs in Western Port, the soft-sediment habitats are key environments as feeding grounds for migratory shore birds and many fish species (Peterson 1977; Howard and Lowe 1984; Edgar and Shaw 1995; Dann 1981, 1999) and are also important for nutrient cycling because of the invertebrates that inhabit them. The intertidal soft-sediment habitats or mudflats of the MNPs in Western Port have not been intensively studied and information about the types of invertebrates and even fish species that are found within the MNPs in Western Port has been inferred from sites outside of the MNPs (Plummer *et al.* 2003; Park Victoria 2007). This presents a significant gap in knowledge about many of the components of the MNPs in Western Port.

The purpose of the current study was to investigate the macroinvertebrates and sediment conditions of the soft-sediment habitats in the MNPs in Western Port to try to fill some of these gaps in knowledge. This report investigates whether after four and five years of protection the macroinvertebrate communities found within the MNPs in Western Port are significantly different to the macroinvertebrate communities found outside of the MNPs.

It was found that there was no significant difference in the species richness and diversity of macroinvertebrates between sites within and external to the MNPs. Instead MNPs seemed to be representative of the species richness and diversity found throughout Western Port. This is important as management will need to consider bay wide processes that influence the benthic invertebrates in the MNPs in Western Port as well as local factors.

There were significant overall differences in macroinvertebrate abundances (multivariate differences) and sediment properties between the three MNP sites and between MNP sites and external sites. The results provide evidence that some differences between these areas do exist although these differences may be more subtle than linear increases or decreases in the number of species or diversity of organisms and sediment properties. Subtle differences between MNP and external sites do provide incentive for continued effort to protect the MNPs from human disturbance because MNPs may facilitate further changes in macroinvertebrate communities that will protect biodiversity of benthic habitats throughout Victoria. This is one of the main goals of the MNPs.

A subset of key variables, potentially useful for monitoring, was identified in the current study. These key variables included the ghost shrimps *Trypaea australiensis* and *Biffarius arenosus*, the crab *Macrophthalmus latifrons*, the polychaeate worm *Lumbrineris* sp. and sediment properties total organic content (TOC) and substrate temperature. Although these key variables require further investigation they provide an efficient starting point for monitoring the macroinvertebrate communities and sediment properties that are key features

of the MNPs in Western Port, Victoria. It is suggested that further studies focussing on these key variables and their interaction or function with other MNP components will promote our understanding of the delicate marine environment in these areas. Species lists of all macroinvertebrates found in the MNPs in this study are also included in this report.

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### 1 INTRODUCTION

Marine protected areas in Victoria were created in 2002 as a representative system of the many unique habitats found throughout southern Australia. These marine protected areas include Marine National Parks and Sanctuaries that are both 'no-take' areas, completely protecting the marine environment of removal of any component, in particular fishes and invertebrates. In Victoria these Marine National Parks and Sanctuaries include areas of rocky reefs, pelagic waters, sandy beaches, subtidal sandy and muddy seabeds, and intertidal mudflats and soft-sediments (Parks Victoria 2003).

Western Port Bay in Victoria has three Marine National Parks (MNPs), Yaringa, French Island and Churchill Island MNPs. The MNPs in Western Port were created to protect areas of seagrass beds, saltmarsh, mangroves, deep channels, soft-sediments and mudflats (Parks Victoria 2007). Specifically, Yaringa MNP protects intertidal mudflats, mangrove and saltmarsh areas; French Island MNP protects seagrass beds, intertidal mudflats, soft-sediment beds, some deep channels, mangroves and saltmarsh; and Churchill Island MNP protects intertidal mudflats, soft-sediment beds, some deep channels, mangroves and saltmarsh; and Churchill Island MNP protects intertidal mudflats, soft-sediment beds, saltmarsh, seagrass beds and deep channels.

With the declaration of Marine National Parks in Victoria, a Strategic Management Plan was written for 2003-2010 (Parks Victoria 2003). This Strategic Management Plan outlined five major themes that are important components of achieving the vision of the MNPs in Victoria. These themes are: Protecting natural values; Protecting and recognizing cultural values; Community engagement; Recreation, tourism and visitor management; and Environmental research and monitoring. The environmental research and monitoring theme is an important component in filling knowledge gaps in many facets of the MNPs in Victoria. The main objective of environmental research and monitoring, as outlined in the strategic management plan, is 'to provide information on the status of natural values and threatening processes and to determine the nature and magnitude of trends over time' (Parks Victoria 2003, p.100). Research and monitoring will also provide information for many of the other themes, such as education, recreation, community involvement and cultural values.

The challenge of developing a monitoring tool for MNPs in Victoria and particularly in Western Port, underpins the research in the report. It is clear that appropriate monitoring tools are needed to assess various components of the marine environment in the MNPs of Victoria to facilitate a deeper understanding of the environments being protected. In particular, gaps in knowledge of fauna and various habitats within the MNPs need to be addressed so that management can be guided by rigorous scientific research. Monitoring programs have been established for some of the intertidal and subtidal rocky reef habitats within Marine National Parks in Victoria (Edmunds *et al.* 2003, 2004; Hart *et al.* 2005) to try to fill these knowledge gaps. However in soft-sediment environments, particularly in the Marine National Parks in Western Port, Victoria, no such monitoring programs have been established.

Rapid assessment techniques are desirable for monitoring because they can reduce the time and money required. In soft-sediment environments this is particularly true because sampling is often difficult and can create disturbance. Rapid assessment techniques are also desirable for long term monitoring because techniques that are easy to use and are efficient, are likely to be sustainable over time. There are several types of rapid assessment techniques (*e.g.* indicators or surrogates). However the general premise of rapid assessment is to reduce the effort involved in monitoring.

# 1.1 Monitoring in Marine Protected Areas: previous examples and expectations

Marine Protected Areas (MPAs) and their 'no-take' management strategy require monitoring to establish if the removal of disturbances, such as fishing, leads to changes in the environment. Due to the complicated and political process of establishing a MPA, before, after, control, impact studies (BACI) are often not available for MPAs (Barrett *et al.* 2007). Focussed monitoring prior to MPA declaration is often not established although this would be the ideal approach. Consequently monitoring a protected area often involves comparing some attribute of the protected area, such as species richness or diversity, over time and with external sites to assess whether the protected areas maintain greater species richness, diversity and/ or abundances of species of interest (Quinn *et al.* 1993; Man *et al.* 1995; Halpern & Warner 2002).

Despite the lack of strict experimental studies such as BACI, many studies comparing sites inside and external to MPAs have given good evidence that MPAs are effective in managing marine environments. Edgar and Barrett (1999) compared sites inside and outside of four of the marine reserves in Tasmania from the time of declaration in 1991. Six years after declaration, their results showed clear increases in the size and number of fishes, invertebrate and algal species inside the marine reserves, suggesting that these areas were successful in protecting many species. Of particular interest were the increases in the densities of large fishes (> 325 mm length), the bastard trumpeter (*Latridopsis forsteri*) and the rock lobster (*Jasus edwardsii*). These species are all commercially important species and increases in their numbers gives evidence that marine reserves, as a management tool, can enhance fish stocks.

Similarly, in Leigh Marine Reserve in New Zealand, the effectiveness of the protection of commercially and recreationally important fish species within the reserve was monitored in a number of ways (Shears & Babcock 2003). Over time, the recovery of macroalgae species from over grazing by the sea urchin *Evechinus chloroticus*, was recorded. The detection of a reduction in sea urchins and an increase in macroalgae biomass gave evidence that the number of fish species (predators of the sea urchin) was increasing within the boundaries of the reserve. Spatially, the increase in macroalgae and decline in sea urchins within the Leigh Marine Reserve was not the same as sites outside of the reserve. Sites outside of the marine reserve remained dominated by sea urchins and barren of macroalgae (Shears and Babcock 2003).

In the Fagatele Bay National Marine Sanctuary, American Samoa, monitoring of fish and coral species allowed the detection of damages caused by major disturbances, such as hurricanes and the invasion of the crown-of-thorns starfish within the sanctuary (Green et al. 1999). This monitoring program also recorded the temporal recovery of coral and fish species after these disturbances which provided vital information to improve the management strategies. Long term monitoring is essential as it can reveal changes in an ecosystem that are not apparent in short term studies. In Tasmania (as mentioned earlier) Edgar and Barrett (1999) gave good scientific evidence that there were increases in the abundances and sizes of a number of species in four marine reserves six years after declaration. In 2007, ten years of data for these marine reserves was analysed and it was suggested that after ten years the changes in the number and size of particular species was much more variable, slow and complex than originally suggested after six years (Barrett et al. 2007). Changes in the number and size of species were also species-specific and thought to be dependent on pressures external to the Marine Reserves. These results highlight the importance of establishing long term monitoring to assess management programs such as MPAs.

Not only is monitoring important for assessing whether the objectives of a MPA are being met, but it can also be important for detecting changes in other key ecosystem processes. In Finland, the recovery of aquatic flora and macroinvertebrates in streams that had been

converted to channels for timber transport was observed by monitoring and comparing the changes, in this case the increase, in abundance of macroinvertebrates at impacted and non-impacted streams (Muotka *et al.* 2002). Long term monitoring of fish assemblages off the Arrabida rocky coast in Portugal, showed that rapid changes in community composition were interspersed with slower changes in community composition in response to changes in climatic conditions, similar to what might be caused by climate change (Henriques *et al.* 2007). Monitoring of benthic nematode assemblages off the coast of Brittany, France, showed negative changes and a subsequent recovery in community composition following the Amoco Cadiz oil spill in 1978 (Gourbault 1987). The changes in community structure due to the oil spill would not have been observed without a monitoring program already being in place.

# 1.2 The need for monitoring strategies in Western Port, Victoria

In Western Port, Victoria, the establishment of three Marine National Parks (MNPs) has resulted in the need to develop monitoring tools to assess two specific goals of MNPs:

- 1. Whether these parks are different, after establishment, to sites outside of the MNPs in community composition given that human impact is reduced in the MNPs.
- 2. Whether the MNPs are meeting their objectives, such as conserving marine biodiversity (Parks Victoria 2007).

There is also a lack of detailed scientific information about the organisms and site characteristics in Western Port that can help to inform management strategies (Carey *et al.* 2007). In particular, there is limited information about the benthic invertebrates inhabiting the MNPs in Western Port even though they are of great importance as a food resource for the many migratory shore bird species that use this RAMSAR site, for feeding and breeding (Parks Victoria 2007).

Benthic macroinvertebrates are thought to be ideal for monitoring soft-sediment environments as they are in direct contact with the habitat (burrowing in the sediment) and often respond to changes in the sediment environment (Pocklington & Wells 1992). Furthermore, benthic macro-invertebrates have been successfully used to monitor differences between sites and changes in sites over time (see Giangrande *et al.* 2005; Smith 2005; Hirst 2008 for examples). In many studies the condition of an area has been assessed by measuring the biodiversity of an area. Common measures of biodiversity are species richness and species diversity. Generally it is thought that higher species richness and diversity is associated with good ecological condition compared to areas of lower species richness. Therefore by comparing the species richness and diversity of benthic macroinvertebrates in soft-sediment habitats within and external to the Marine National Parks in Western Port, some assessment about the condition of the Marine National Parks can be made.

Alternatively, multivariate statistical approaches using macroinvertebrate community composition may be useful in identifying whether MNPs, facilitate differences in overall macroinvertebrate assemblages compared with sites outside of the MNPs. If differences in the macroinvertebrates exist, these may be tracked over time (monitored) to identify if these differences become more or less pronounced (Pik *et al.* 2002). In particular, increases in macroinvertebrates that are reduced due to bait collection or trampling outside of the MNPs may be useful in assessing if the no take strategy in these areas is successful.

Time and funding restraints prohibit the collection and monitoring of all of the important components of soft-sediment environments in the MNPs of Western Port. In the past, collection and assessment of too many variables has been a major downfall of monitoring

ecosystems and has resulted in monitoring programs that have been too ambitious (Hellawell 1991). Along with this, the aim of collecting many variables is often not clearly stated. This has often resulted in the collection of a large amount of useless data. Long term monitoring of a subset of important variables that directly link to the aims of monitoring would be more favourable in assessing the effectiveness of protecting soft-sediment environments within MNPs in Western Port.

#### 1.3 Aims

The aim of the current study was to compare the MNPs of Western Port with external sites in Western Port to investigate any key differences in the species and/ or sediment properties that make the MNPs unique. It was expected that the number of species, the number of individuals and diversity would be higher in the MNP sites as these are protected from human disturbances. Other hypotheses have been proposed that show that the opposite may be true. For example, there might be an increase in diversity external to the MNPs or with increasing disturbance (see the ecological disturbance hypothesis, Connell 1978). However, the hypothesis proposed in this thesis aligns with other studies of MPAs from around the world, many of which show an increase in the number of individuals and diversity after the establishment of the protected area (for examples see Edgar and Barrett 1999; Shears and Babcock 2003, Langlois et al. 2005). It was also expected that the sediment properties within the MNPs would be representative of sediments found throughout Western Port. A comparison between sites from each of the three MNPs in Western Port also aimed to identify differences in invertebrate species and sediment properties between the MNPs. Finally, the current study aimed to use a multivariate approach to identify macroinvertebrates and/ or sediment properties that are important biological components of these habitats. Any subset of variables identified via the multivariate approach may provide a tool for monitoring changes in the benthic environment over time. Changes in a subset of variables can alert managers to changes in processes or human impacts within Western Port MNPs and give evidence that the MNPs in Western Port are effective in conserving the soft-sediment environments.

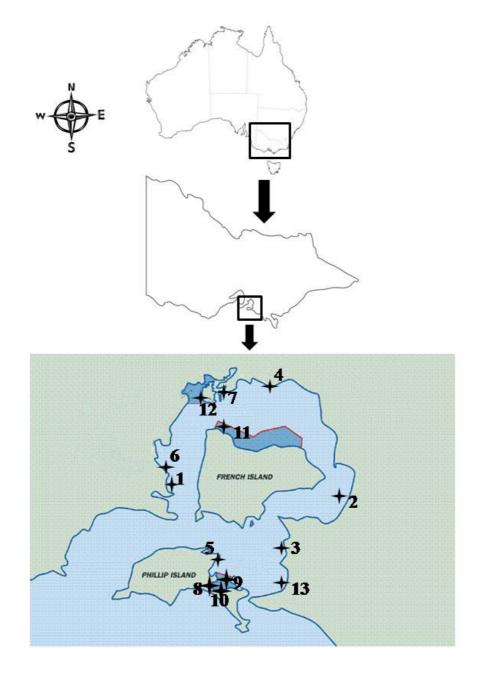
### 2 METHODS

#### 2.1 Study sites

Sampling was conducted in March, April and May 2006 at ten sites around Western Port, Victoria. Sites included Stony Point, Lang Lang, Coronet Bay, Tooradin, Rhyll, Crib Point, Warneet and three within Marine National Parks (MNPs): Churchill Island MNP, French Island MNP and Yaringa MNP (Figure 2.1). Each site was sampled on one day at low tide. Sites were generally open mudflat, either flat or very gently sloping (<1 degree elevation). Sites at Stony Point, Tooradin, Rhyll and Warneet were within 150 m of heavily used boat ramps, while Crib Point, Lang Lang, and Coronet Bay were in the same area as boat ramps but >150 m from any heavy use by humans. Churchill Island MNP, French Island MNP and Yaringa MNP were situated away from boating activities and had limited access/ use by humans.

In 2007, the number of sites within MNPs was increased to five and the number of sites outside the MNPs was decreased to six sites because the focus of the study was shifted to a comparison of inside/ outside of MNPs. In March, April and May 2007, Coronet Bay, Tooradin, Rhyll, Crib Point, Warneet, Bass River, Churchill Island MNP (three sites separated by > 1 km), French Island MNP and Yaringa MNP were sampled. Stony Point and Lang Lang were replaced by Bass River in 2007 because Bass River is in the southern region of Western Port and this balanced the number of sites outside of MNPs (Coronet Bay, Rhyll and Bass River) with the three sites within Churchill Island MNP in this region of the bay. Three sites outside of MNPs (Tooradin, Warneet and Crib Point) were situated in the northern region of Western Port along with a single site sampled in each of Yaringa MNP and French Island MNP. The plan was to sample three sites in French Island MNP in 2007 to balance the number of northern sites in and out of MNPs. However for this study, the mud/sand flats in French Island MNP were only accessible by boat with the assistance of Parks Victoria rangers. A combination of bad weather and inappropriate low tides restricted sampling to only one site in 2007.

All sites were typical mudflats of Western Port, some containing small stands of seagrass (*Heterozostera tasmanica* and/ or *Zostera muelleri*) and in some cases fringed by stands of the grey mangrove *Avicennia marina*.



**Figure 2.1** Location of Western Port, Victoria, Australia, showing the sites sampled in 2006 and 2007. Dark blue shades show Marine National Parks in Western Port. Sites are represented by the numbers: 1 – Stony Point, 2 – Lang Lang, 3 – Coronet Bay, 4 – Tooradin, 5 – Rhyll, 6 – Crib Point, 7 – Warneet, 8 – Churchill MNP site 1, 9 – Churchill MNP site 2, 10 – Churchill MNP site 3, 11 – French Island MNP, 12 – Yaringa MNP.

#### 2.2 Sampling procedure

At each site three transects (labelled transect 1, transect 2 and transect 3) were marked out running from higher on the shore towards the low tide mark. Transects were 30 m long and spaced 20 m apart. Along each transect, three large cores (15 cm diameter, 40 cm depth) were taken at 10 m intervals (total of nine large cores per site), and ten small cores (5 cm diameter, 10 cm depth) were taken at 3 m intervals (total of 30 small cores per site) (Figure 2.2).

In the field all cores, large and small, were sieved to collect the invertebrates contained within them. The large cores were sieved through a 1mm sieve size and invertebrates were collected using forceps and immediately place in vials containing 100% ethanol. Ethanol of 100% was used instead of the standard 70% as it was hoped that specimens would be used for future for genetic analysis in another project. All small cores were sieved using 1 mm and 0.5 mm mesh sieves in a series. The samples remaining in the sieves were flushed into plastic zip-lock bags with seawater and 4% formalin was added for preservation of the samples. In the laboratory, macroinvertebrates from large cores were identified to species with the aid of a dissecting microscope. The sediment collected from small cores was searched for smaller macroinvertebrates (~5 mm) under a dissecting microscope and identified to species. The number of each species and the number of individuals in each core was recorded.

Temperature, porosity, organic carbon content, sediment particle size, chlorophyll *a* concentration, pH of the sediment, redox potential and seagrass cover (vegetation cover) were all measured at 10 m, 20 m and 30 m points along each of the three transects (total of nine measurements/ samples per site) (Figure 2.1). Temperature was taken using a standard mercury thermometer (0-40 °C) inserted into the sediment to 5-10 cm depth. Samples to measure porosity were collected using a 60 ml syringe with the end removed and 20 ml of surface sediment was extracted and placed in a plastic zip-lock bag. Sediment cores of 5 cm diameter and 10-15 cm depth were collected for organic carbon content and particle size analysis. Samples for chlorophyll *a* concentration were collected by taking a teaspoon full of sediment from the sediment surface, wrapping this in aluminium foil and freezing on site using dry ice. The pH of the sediment was tested with a standard CSIRO soil pH testing kit (see website: www.inoculo.com.au), and percentage seagrass cover (percentage of vegetation cover) was measured using a 25 x 25 cm quadrat divided into 100 smaller squares. Redox potential was measured using a Calomel reference redox meter, and readings of redox were taken from the top few centimetres of sediment and at 10 cm depth.

Pore water samples were taken at 10 m and 20 m along transect 1 and transect 2 for analysis of total Nitrogen (N) and Phosphorus (P), soluble N and P, and ammonia. Pore water was collected using a 60 ml syringe, with 10-20 cm of aquarium tubing and ceramic airstone (2 cm long and 1cm diameter) attached as described in Winger and Lasier (1991). For each sample, 50 ml for total N and P, 50 ml for soluble N and P, and 100 ml for ammonia of pore water was extracted at the two sampling points along transect 1 and transect 2. The samples for soluble N and P were filtered through glass fibre filter paper to retain soluble particles <  $0.5\mu$ m. All pore water samples were kept in ice in the field and later frozen for storage. All pore water samples were sent frozen to Deakin University Water Quality Laboratory, Warrnambool, for analysis.

In 2007, sampling methods were identical to 2006 except that the sediment variable pH was also omitted as it was found to be constant throughout Western Port at the scale that the CSIRO soil testing kit could detect. Perhaps a finer resolution would be more important or detect differences between sites but because this was not measured in 2006 it was not measured in 2007.

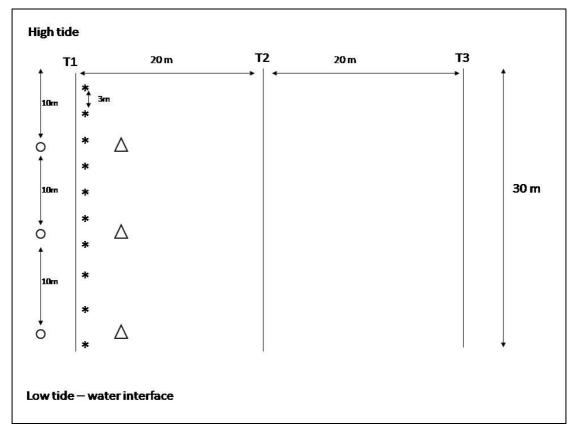


Figure 2.2 Layout of sampling area at each site. T1-3 represent transects 1-3, O represent large \*

cores, \* represent small cores and  $\Delta$  represent sediment samples.

#### 2.3 Analysis of sediment samples

Samples for porosity were weighed (wet weight) and then dried in an oven for 48 h at 100 °C. Dry weight was then measured and the difference between the wet weight and dry weight was calculated and divided by the initial volume (20 ml) to determine porosity.

Sediment cores collected for organic carbon content and sediment particle size were thoroughly mixed in a container and 30 g and 100 g (wet weight) sub-samples were separated for carbon content and sediment particle size respectively. Sediment collected for organic carbon content was dried in an oven for 48 h at 100 °C then weighed (dry weight) and placed in a combustion oven for 3.5 h at 500 °C. Combusted weight was measured and the difference between dry weight and combusted weight was calculated to give the proportion of organic carbon in each sample.

Sediment collected for sediment particle size was dried in an oven for 48 h at 100 °C. Dried samples were then shaken in a nested sieve series (2000, 1000, 500, 250, 125, 63  $\mu$ m) for 10 minutes at an amplitude of 2.5 using a Fritsch GmbH vibratory sieve shaker "Analysette 3". The sediment in each sieve was weighed and the proportion of each sediment particle size class was calculated by dividing the weight of each sediment size class by the total weight of the sample sieved (see Contessa & Bird 2004).

Chlorophyll *a* concentration was determined by placing 1 g of sediment (wet weight) into a pre-weighed and labelled test tube and adding 12.6 ml of acetone. This was mixed thoroughly and chilled in a darkened ice bath for 72 hrs. After 72 hrs, the supernatant was transferred into a large centrifuge tube containing 1.4 ml of distilled water and mixed to create a 90% chlorophyll extract solution. The solution was then centrifuged for 5 mins at

3000 rpm. Sediment remaining in the test tubes was dried at 100 °C for 48 hrs. After centrifuging, the solution containing extracted chlorophyll was analysed using a spectrophotometer at wavelengths of 664, 647 and 630 nm ( $E_{664}$ ,  $E_{647}$ ,  $E_{630}$ ). The concentration of chlorophyll *a* in the samples was then calculated using the equation (Equation 1) in Jeffrey and Humphrey (1975) for mixed chlorophyll samples in 90% acetone extract.

$$\mu g \text{ chl } a = 11.85 E_{664} - 1.54 E_{647} - 0.08 E_{630} \tag{1}$$

Sediment samples from 2006 and 2007 were analysed in exactly the same way.

# 2.4 Univariate analysis of macro-invertebrates and sediment properties at all sites

The number of species, the number of individuals and Simpson's diversity index were compared for each site in 2006 and 2007 using a two-way ANOVA using the statistical package SPSS 15.0. Independent factors were year and site. The sediment properties; substrate temperature, proportion of 250  $\mu$ m sediment particle size, proportion of < 63  $\mu$ m sediment particle size, porosity, chlorophyll *a* concentration, total organic content (TOC), redox at 10 cm depth, percentage of vegetation cover, concentration of total nitrogen (N), concentration of total phosphorus (P), concentration of soluble reactive phosphate (react P), concentration of oxidised N and concentration of ammonia were also compared between sites for 2006 and 2007 and again tested using a two-way ANOVA in SPSS version 15.0 with year and site as independent factors.

Assumptions of a two-way ANOVA were tested and it was found that even after a number of transformations both the assumptions of normality and homogeneity of variances were not met, although usually one of these two assumptions was met. Despite this, a two-way ANOVA was used as these tests tend to be robust to variation in the assumptions of normality and/ or homogeneity of variances (Quinn & Keough 2002). To reduce the risk of obtaining a type I error due to not meeting the assumptions, a more conservative significance level of 0.01 was used instead of 0.05.

Excel 2007 was used to plot the mean number of species, mean number of individuals and the mean Simpson's diversity index and the mean of each sediment property for each site in 2006 and 2007. Excel 2007 was also used to create tables showing the number of species collected from each site in 2006 and 2007.

# 2.5 Multivariate analysis of macro-invertebrates and sediment properties all sites

The multivariate statistical analysis package PRIMER (Clarke & Gorley 2001) was used to compare macroinvertebrate community composition between samples around Western Port in 2006 and 2007 to identify any differences between the MNP sites and sites external to MNPs.

Macroinvertebrate abundance data was square root transformed to reduce the effect of dominant species on the analysis. A ranked similarity matrix was conducted on data for 2006 and 2007 data, using Bray-Curtis similarity measures. Statistical tests to assess the differences in community composition between MNP sites and external sites in 2006 and 2007 were done using a one-way analysis of similarities (ANOSIM) (Clarke & Gorley 2001).

The species that contributed to 50 percent of the average (dis)similarities between MNP sites and external sites were identified using the similarities percentages procedure SIMPER (Clarke 1993). A non-metric multidimensional scaling (MDS) ordination was conducted for the square root transformed invertebrate abundance data in PRIMER. Bray-Curtis similarity measures were used and sites within and external to MNP were compared. The data from the MDS were graphed in Excel 2007.

Euclidean distance measures were used to create a ranked similarity matrix of sediment properties for 2006 and 2007. Sediment properties included in the multivariate analysis were; percentage of < 63 µm sediment particle size (<63 µm sediment size), total organic carbon content (TOC), chlorophyll *a*, porosity, redox at 10 cm depth, and percentage vegetation cover. Euclidean distance measures were also used to create a ranked similarity matrix of nutrient concentrations for 2006 and 2007. Nutrient measures included in the analysis included total nitrogen (total N), total phosphorus (total P), soluble reactive phosphate (reactive P), oxidised nitrogen (oxidised N), and ammonia. Data of TOC, total N, total P, reactive P and oxidised N were log-transformed and all nutrients data was normalised prior to the calculation of the Euclidean distance similarity matrices. A one-way ANOSIM was used to statistically test the differences between samples from MNP and external sites in 2006 and 2007 in their sediment composition and nutrient concentrations separately.

The differences in macroinvertebrate abundances between MNP and external sites were compared with the differences between sediment properties between MNP and external sites using Spearman's rank correlation in the BVSTEP routine in PRIMER (Clarke & Gorley 2001). This correlation compared the Bray-Curtis (square root transformed) similarity matrix of macroinvertebrate abundance data and the Euclidean distance (normalised) similarity matrix of sediment property data (Clarke & Gorley 2001). This was done for 2006 and 2007 data. BVSTEP gave the sediment properties that 'best' explained the (dis)similarities between MNP sites and external sites in macroinvertebrate abundances. The correlation between sediment properties and macroinvertebrate abundances was tested statistically using a random permutations test in BVSTEP to give a correlation coefficient (rho) and a significance value (p value).

The "stress" value is given on each MDS ordination plot. This value indicates how well the ordination given represents true relationships between data points. An acceptable stress value is under 0.2 or as low as possible (Clarke & Warwick 2001). Some of the values in the current study are 0.2 or slightly higher but all are closer to 0.2 than 0.3. Therefore, it is accepted that the MDS ordinations plots in this study are useful in representing the data that were collected.

#### 2.6 Multivariate analyses of macroinvertebrate abundances and sediment properties in MNP sites only

A comparison of the three MNP sites was done to test if sites within MNPs were distinctly different from each other in terms of the macroinvertebrate community composition and/ or sediment properties. Similarity matrices for macroinvertebrate abundances and sediment properties were calculated using the same transformations and distance measures discussed in Section 2.3. A one-way ANOSIM was used to compare the macroinvertebrate abundances (Bray-Curtis similarities, square root transformed) and sediment properties (Euclidean distances, normalised data) between MNP sites in 2006 and 2007. The species that contributed to 50 percent of the average (dis)similarities between MNP sites were identified using the similarities percentages procedure SIMPER (Clarke 1993).The macroinvertebrates and sediment properties were then compared in 2006 and 2007 using the BVSTEP routine in the BEST procedure in PRIMER (Clarke & Gorley 2001). The BVSTEP routine was used to identify the sediment properties that could explain any

differences between sites based on macroinvertebrate abundances. Plots of the MDS ordinations were created in Excel 2007 using the co-ordinates calculated in PRIMER for non-parametric MDS to compare the differences in macroinvertebrate abundances and the differences in sediment properties between MNP sites in 2006 and 2007 separately.

# 2.7 Multivariate assessment of the subset of key variables identified from the initial multivariate analysis

The multivariate analyses given so far identify a number of variables that best explain significant differences between MNP and external sites and between MNP sites in terms of the macroinvertebrates and sediment properties collected in the current study (see results sections 3.2 and 3.3). In order to identify variables that may be used for long term monitoring, only the variables that are found to contribute to all of these comparisons (MNP sites with external sites and between MNP sites only) were investigated further. The subset of key variables that contributed to all multivariate differences between sites were: four species, B. arenosus, B. lepte, M. latifrons, Lumbrineris sp., and two sediment properties, TOC and < 63 µm sediment particle size. The species *Trypaea australiensis* and the sediment property substrate temperature were also included in the subset as they were found to be important contributors to the differences between MNP sites and external sites. In total there were eight variables considered important for differences between MNP and external sites and between MNP sites. Therefore, further analysis involved using only six of these eight key variables as two of the six key variables (*B. lepte* and  $< 63 \mu m$  sediment particle size) do not meet the criteria of monitoring variables being easy and efficient to sample. *Barantolla lepte* is a small polychaete worm in the common family Capitellidae (1-2 mm wide and ~ 10mm long). The features of this polychaete worm essential in determining the species are usually only distinguishable by using a compound microscope (Glasby et al. 2000). The sediment particle size < 63  $\mu$ m requires sorting sediment (wet and/ or dry sieving) in the laboratory to extract only sediment particles of this size (Buchanan 1984). Both identification of B. lepte and sorting of  $< 63 \mu m$  sediment particles can be both tedious and time consuming and also require expertise, not conducive to sampling efficiently to monitor areas long term. Therefore, these two variables are left out of further analysis to assess whether the four common key variables and T. australiensis and substrate temperature identified here could represent differences in sediment properties and macroinvertebrates between MNP sites and external sites and between MNP sites only.

A similarity matrix was constructed using only the six remaining key variables. Euclidean distances were used to create the similarity matrices. Total organic content was transformed with a natural log to bring the data closer to normality, although not necessarily to be completely normally distributed. Species abundances were square root transformed, again to reduce the effect of very dominant species. All data was standardised as they were of different measurements (*i.e.* number of individuals per core for species and g.g<sup>-1</sup> of dry weight for TOC). A two-way ANOSIM was then used to test the differences between MNP sites and external sites based on all six key variables. Subsequent one-way ANOSIM, for 2006 and 2007 (separately) were done to test whether the six key variables could show significant differences between MNP sites and external sites from one year to the next. MDS ordinations using the six key variables were calculated in PRIMER for 2006 and 2007 separately and the co-ordinates were plotted using Excel 2007.

# 2.8 Univariate comparison of key variables between the MNP sites

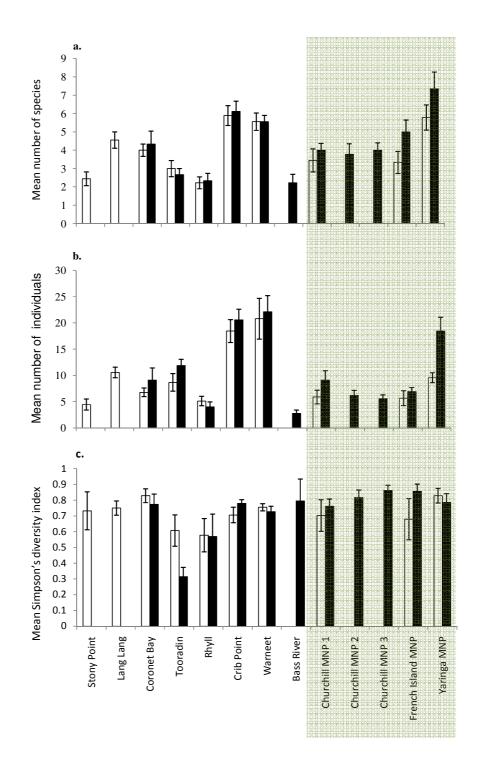
A comparison of the numbers of key species within the MNPs and external to the MNP sites was made to see if trends exist that may be used for future monitoring of these variables. The number of individuals per m<sup>2</sup> of *Macrophthalmus latifrons*, *Lumbrineris* sp., *Biffarius arenosus*, and *Trypaea australiensis* were plotted for Coronet Bay, Tooradin, Rhyll, Crib Point, Warneet, Churchill Island MNP site 1, French Island MNP and Yaringa MNP for 2006 and 2007 separately using Excel 2007. Significant differences between the number of individuals of the key species at each site was tested using a two-way ANOVA. As explained earlier in this chapter (Section 2.2) the assumptions of a two-way ANOVA were tested and it was found that even after transformations the assumptions of normality and homogeneity of variances were not met. Despite this, a two-way ANOVA was used as these tests tend to be robust to variation in the assumptions of normality and/ or homogeneity of variances (Quinn & Keough 2002). Again, to reduce the risk of obtaining a type I error due to not meeting the assumptions, a more conservative significance level of 0.01 was used instead of 0.05.

### 3 RESULTS

# 3.1 Univariate analysis of macroinvertebrates and sediment properties at all sites

Two-way ANOVA showed there was no significant interaction effect found between years (2006 and 2007) and sites and there was no significant difference in the number of species between years (p > 0.01). There was, however, a significant difference in the number of species found between sites (F = 0.388, df = 12, p < 0.001). Yaringa MNP, Crib Point and Warneet consistently had a greater number of species than other sites (Figure 3.1). Tukey's post hoc analysis showed that there were no consistent differences between Marine National Park (MNP) sites and other sites around Western Port with a number of significant differences between both external and MNP sites and between MNP sites (Table 3.1, Figure 3.1a).

Similarly there was no significant interaction effect found between years and sites in the number of individuals. There was, however, a significant difference in the number of individuals collected each year at a site (F = 8.625, df = 1, p = 0.004) with a greater number of individuals being present in 2007 than 2006. A significant difference in the number of individuals collected at each site (F = 18.872, df = 12, p < 0.001) was also found with Coronet Bay, Warneet and Yaringa having a greater number of individuals (Figure 3.1b). As with the number of species collected at each site, there were no consistent significant differences between MNP sites and external sites in the number of individuals collected but many significant differences between various sites, external and MNP sites (Table 3.2, Figure 3.1b).



**Figure 3.1** The mean number of species a. the mean number of individuals b. and mean Simpson's diversity c. per core (15 cm diameter X 40 cm depth) collected at each site in Western Port, Victoria, in 2006 (open bars) and 2007 (filled bars). Sites within Marine National Parks are shaded. Error bars show one standard error.

**Table 3.1** Statistical results of the two-way ANOVA used to test the difference between the mean number of species at each site sampled in Western Port in 2006 and 2007. Post Hoc pairwise site comparisons are given for significant pairwise comparison. Non-significant comparisons are not shown. Significant results are given in bold.

	F	df	р
Mean no. species			
between years	0.099	1	0.097
between sites	0.388	12	<0.001
sites*years	0.899	7	0.508
Pairwise comparisons			
Stony Point vs Crib Point			<0.001
Stony Point vs Warneet			<0.001
Stony Point vs Yaringa MNP			<0.001
Lang Lang vs Rhyll			0.002
Coronet Bay vs Rhyll			0.001
Tooradin vs Crib Point			<0.001
Tooradin vs Warneet			<0.001
Tooradin vs Yaringa MNP			<0.001
Rhyll vs Crib Point			<0.001
Rhyll vs Warneet			<0.001
Rhyll vs Churchill Is. MNP site 1			0.025
Rhyll vs Churchill Is. MNP site 2			0.026
Rhyll vs French Is. MNP			0.007
Rhyll vs Yaringa MNP			<0.001
Crib Point vs Bass River			<0.001
Crib Point vs Churchill Is. MNP site 1			0.018
Warneet vs Bass River			<0.001
Bass River vs Yaringa MNP			<0.001
Churchill Is. MNP site 1 vs Yaringa MNP			<0.001
French Is. MNP vs Yaringa MNP			0.024

Two-way ANOVA showed that there was a significant interaction effect in the Simpson's diversity index between years and sites (F = 5.229, df = 7, p < 0.001, Figure 3.1), making it difficult to determine if there are any true differences in diversity between sites only or years only. At some sites (*e.g.* Crib Point) diversity was greater in 2007 whereas at other sites (*e.g.* Coronet Bay) diversity was greater in 2006 (Figure 3.1c). No post hoc tests were carried out for the Simpson's diversity index between sites due to the significant interaction effect. A significant interaction effect here means we cannot easily determine whether significant differences are due to the differences between years or the differences between sites. As with the number of species and the number of individuals collected at each site, there does not seem to be any consistency in differences between the Simpson's diversity index found in the MNP sites and the external sites (Figure 3.1c).

Appendix 1 shows the species and the number of individuals of these species that were collected from each site in 2006 and 2007. In general, differences between sites seem to be due to small and inconsistent differences in the number of species that are only represented by a few individuals (see Appendix 1, amphipods and polychaete worms for examples). Other differences between sites are most likely due to the relative abundance of the more

dominant macroinvertebrates which varies from very abundant to only a few individuals (see Appendix 1, decapod crustaceans *Biffarius arenosus, Macrophthalamus latifrons, Trypaea australiensis* for examples). A number of species are more abundant in the MNPs than in any of the external sites, and some species are restricted solely to the MNPs. These species include *Alpheus richardsoni, Paragrapsis* sp., *Paratanaidae* sp., *Armandia* MoV sp. 282, *Musculista senhousia*, and *Phoronopsis albomaculata*, and are all found in very low numbers.

A two-way ANOVA of each sediment property showed that there was a significant interaction effect for all sediment properties between sites and years (p<0.01, Table 3.3) except for porosity, chlorophyll a and percentage vegetation cover (p > 0.01, Table 3.3). In addition, all sites, even those without strong interaction effects, showed significant differences between sediment properties between years (Table 3.3). The only sites with no significant difference found between years were chlorophyll *a* and percentage of vegetation cover (p > 0.01, Table 3.3). As with the number of species, individuals and the Simpson's diversity at each site, there were no consistent differences between the sediment properties between MNP sites and external sites (Figure 3.2, 3.3, 3.4). Post Hoc tests revealed some interesting differences between MNP and external sites. Yaringa MNP had significantly greater vegetation cover (seagrass) than all other sites (p < 0.01) except Churchill Island MNP site 1, Rhyll and Stony Point (Figure 3.2d). A significantly higher chlorophyll *a* concentration was found in sediments of Churchill Island MNP (sites 2 and 3) compared with all other sites (p < 0.01, Figure 3.2b) and significantly higher levels of TOC were found than at other sites except Churchill Island MNP site 1, Tooradin, Rhyll and Bass River (p < 0.01, Figure 3.3d). Yaringa MNP also had a higher concentration of ammonia (Figure 3.4e) than at Coronet Bay, Rhyll and French Island MNP. However these differences were not significant at p < 0.01 but at were significant at p < 0.05. In section 2.2, it is explained that only significance at p < 0.01 would be accepted due to the assumptions of normality and homogeneity of variances not being met.

**Table 3.2** Statistical results of the two-way ANOVA used to test the difference between the mean number of individuals at each site sampled in Western Port in 2006 and 2007. Pairwise comparisons are given for significant pairwise comparisons; non-significant pairwise results are not shown. Significant results are given in bold.

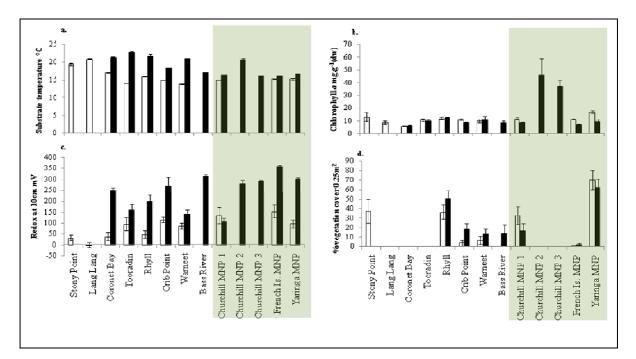
	F	df	р
Mean no. individuals			
between years	8.625	1	0.004
between sites	18.872	12	<0.001
sites*years	1.337	7	0.236
Pairwise comparisons			
Stony Point vs Lang Lang			0.002
Stony Point vs Tooradin			0.001
Stony Point vs Crib Point			<0.001
Stony Point vs Warneet			<0.001
Stony Point vs Yaringa MNP			<0.001
Lang Lang vs Rhyll			0.001
Lang Lang vs Bass River			<0.001
Coronet Bay vs Rhyll			0.048
Coronet Bay vs Crib Point			<0.001
Coronet Bay vs Warneet			<0.001
Coronet Bay vs Bass River			<0.001
Rhyll vs Tooradin			<0.001
Rhyll vs Crib Point			<0.001
Rhyll vs Warneet			<0.001
Rhyll vs Yaringa MNP			<0.001
Crib Point vs Tooradin			<0.001
Crib Point vs Bass River			<0.001
Crib Point vs Churchill Is. MNP site 1			<0.001
Crib Point vs Churchill Is. MNP site 2			<0.001
Crib Point vs Churchill Is. MNP site 3			<0.001
Crib Point vs French Is. MNP			<0.001
Warneet vs Tooradin			<0.001
Warneet vs Bass River			<0.001
Warneet vs Churchill Is. MNP site 1			<0.001
Warneet vs Churchill Is. MNP site 2			<0.001
Warneet vs Churchill Is. MNP site 3			<0.001
Warneet vs French Is. MNP			<0.001
Bass River vs Tooradin			<0.001
Bass River vs Churchill Is. MNP site 1			0.018
Bass River vs Yaringa MNP			<0.001
Churchill Is. MNP site 1 vs Yaringa MNP			0.016
Churchill Is. MNP site 2 vs Yaringa MNP			<0.001
Churchill Is. MNP site 3 vs Yaringa MNP			<0.001
French Is. MNP vs Yaringa MNP			<0.001

**Table 3.3** Statistical results of the two-way ANOVA comparing each sediment property and nutrient property measured at each site in years 2006 and 2007. Significant results are given in bold.

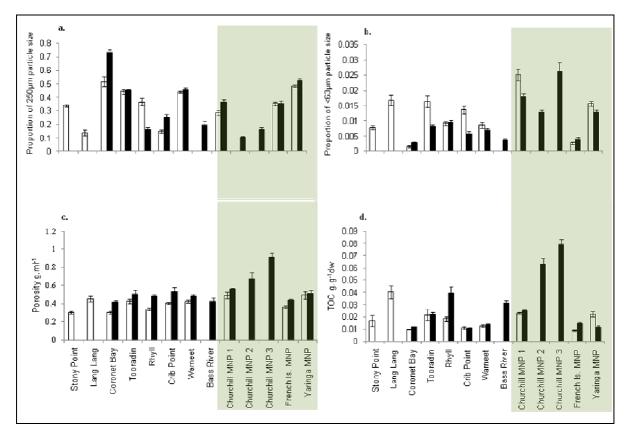
	F	df	р
Substrate temperature			
between years	1736.92	1	<0.001
between sites	176.625	12	<0.001
sites*years	103.124	7	<0.001
Medium sand (250µm)			
Between years	2.208	1	0.139
Between sites	72.272	12	<0.001
Sites*years	12.671	7	<0.001
Fine sediment (<63µm)			
between years	8.038	1	0.005
between sites	79.72	12	<0.001
sites*years	10.737	7	<0.001
Porosity			
between years	47.263	1	<0.001
between sites	17.143	12	<0.001
sites*years	1.732	7	0.105
Chlorophyll a			
between years	1.453	1	0.23
between sites	13.039	12	<0.001
sites*years	0.391	7	0.907
Total organic content (TOC)			
between years	6.081	1	0.015
between sites	48.84	12	<0.001
sites*years	8.104	1	<0.001
Redox at 10cm depth			
between years	168.734	1	<0.001
between sites	12.398	12	<0.001
sites*years	9.719	7	<0.001
% vegetation cover			
between years	0.304	1	0.583
between sites	20.781	12	<0.001
sites*years	1.444	7	0.191
Total N			
between years	5.257	1	0.023
between sites	7.321	12	<0.001
sites*years	4.799	7	<0.001

#### Table 3.3 continued

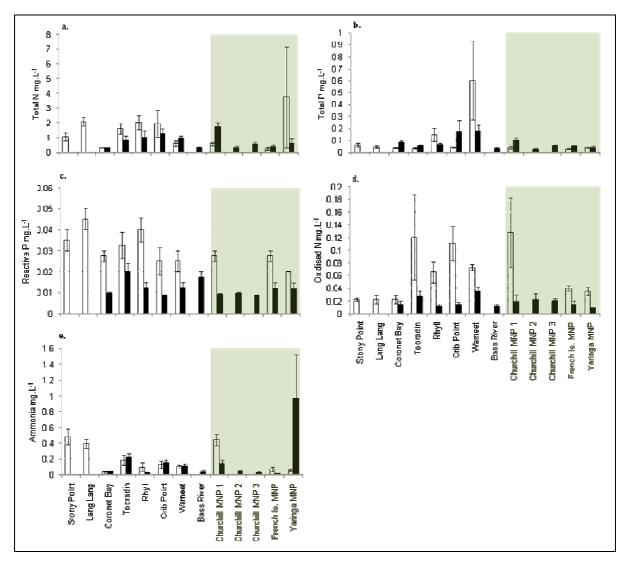
	F	df	р
Total P			
between years	2.306	1	0.131
between sites	17.147	12	<0.001
sites*years	14.779	7	<0.001
Soluble reactive P			
between years	733.347	1	<0.001
between sites	18.925	12	<0.001
sites*years	6.016	7	<0.001
Oxidised N			
between years	294.055	1	<0.001
between sites	19.733	12	<0.001
sites*years	7.959	7	<0.001
Ammonia			
between years	30.079	1	<0.001
between sites	78.363	12	<0.001
sites*years	21.709	7	<0.001
sites*years	6.016	7	<0.001



**Figure 3.2** Measures of various sediment properties a. substrate temperature, b. chlorophyll *a*, c. redox potential at 10cm and d. % vegetation cover at each site sampled in Western Port in 2006 (open bars) and 2007 (closed bars). Error bars are ± standard error. Shaded sites are Marine National Parks.



**Figure 3.3** Measures of various sediment properties a. proportion of 250  $\mu$ m particle size, b. proportion of <63 $\mu$ m particle size, c. porosity and d. total organic content (TOC) at each site sampled in Western Port in 2006 (open bars) and 2007 (closed bars). Error bars are ± standard error. Shaded sites are Marine National Parks



**Figure 3.4** Measures of various nutrients a. total N, b. total P, c. soluble reactive P, d. oxidised N and e. ammonia at each site sampled in Western Port in 2006 (open bars) and 2007 (closed bars). Error bars are  $\pm$  standard error. Shaded sites are Marine National Parks.

### 3.2 Multivariate analysis of macroinvertebrates and sediment properties at all sites

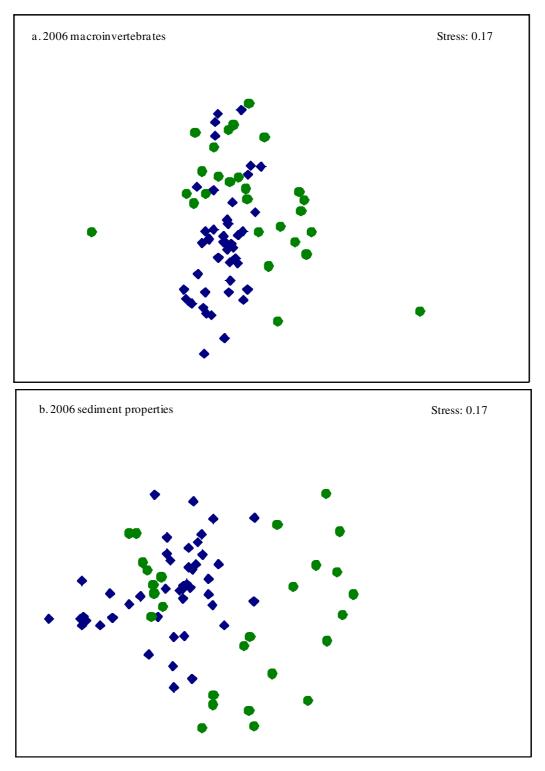
One-way ANOSIM showed that there was a significant difference in the overall macroinvertebrate abundances between MNP sites and external sites in 2006 (Global R = 0.373, p = 0.001) (Figure 3.5) and in 2007 (Global R = 0.230, p = 0.001) (Figure 3.6). Similarity percentages analysis (SIMPER) showed that there was an average dissimilarity of 78.11% between MNP sites and external sites. Five species, the ghost shrimps *Biffarius arenosus* and *Trypaea australiensis*, the polychaetes *Barantolla lepte* and *Lumbrineris* sp., and the crab *Macrophthalmus latifrons*, were found to contribute to 50% of the average dissimilarity in overall macroinvertebrate abundances between MNP and external sites.

A one-way ANOSIM also showed that there were significant differences in the sediment properties between MNP and external sites in 2006 (Global R = 0.328, p = 0.001) (Figure 3.5) and 2007 (Global R = 0.360, p = 0.001) (Figure 3.6). No significant difference in nutrient concentrations was found between MNP sites and external sites in 2006 (Global R = 0.051, p = 0.167) or 2007 (Global R = 0.041, p = 0.272), so nutrient concentrations variables were left out of subsequent multivariate comparisons with macroinvertebrates.

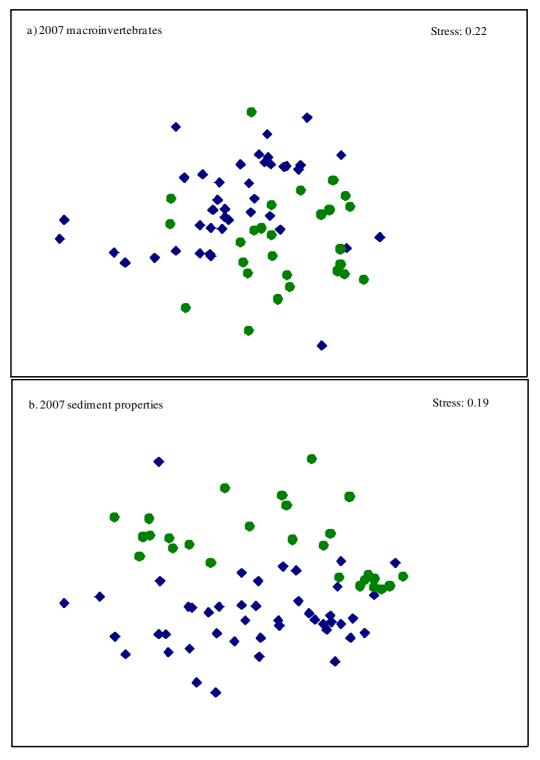
Comparisons of the macroinvertebrate abundances and sediment properties using BVSTEP analysis showed that in 2006 there was a significant correlation between the similarities in macroinvertebrate abundances and similarities in sediment properties between MNP sites and external sites (Spearman's rho = 0.458, p = 0.001) (Figure 3.5). The BVSTEP analysis also showed that total organic content (TOC) could explain the similarities in macroinvertebrate abundances better than all other sediment properties (rho = 0.458).

A significant correlation was also found in 2007 between the similarities in macroinvertebrate abundances and the similarities in sediment properties between MNP sites and external sites (Spearman's rho = 0.309, p = 0.001) (Figure 3.6). In 2007, however, three sediment properties were found to explain the similarities in macroinvertebrate abundances better than any other sediment property (rho = 0.309). These three sediment properties were TOC, substrate temperature and the proportion of < 63  $\mu$ m sediment particle size.

The relatively small Spearman's rank coefficient in 2006 (rho = 0.458) and in 2007 (rho = 0.309) suggests that although there is a correlation between the macroinvertebrates and the sediment properties at these sites in each year the sediment properties, TOC, < 63  $\mu$ m particle size and substrate temperature only explain about 31 - 46% of the similarities in macroinvertebrate abundances between MNP sites and external sites. This is evident in Figures 3.5 and 3.6 where a number of samples from MNP are more similar to samples from external sites (closer or overlapping blue circles and green diamonds on Figure 3.5 and 3.6) than other samples from MNP sites. This suggests that there are some similarities between samples from MNP and external sites in community composition of macroinvertebrates and sediment properties, and the grouping of samples into MNP and external sites does not always reflect the biggest differences between the samples.



**Figure 3.5** MDS ordinations of a. Bray-Curtis similarity matrix for macroinvertebrate abundances (square root transformed) showing MNP sites (circles) and external sites (diamonds) and b. Euclidean distance matrix for sediment properties (natural log transformation of TOC) showing MNP sites (closed circles) and external sites (closed diamonds) for 2006 samples only. The two-dimensional stress values indicate how well the ordinations represent true relationships between data points.



**Figure 3.6** MDS ordinations of a. Bray-Curtis similarity matrix for macroinvertebrate abundances (square root transformed) showing MNP sites (circles) and external sites (diamonds) and b. Euclidean distance matrix for sediment properties (natural log transformation of TOC) showing MNP sites (circles) and external sites (diamonds) for 2007 samples only. The two-dimensional stress values indicate how well the ordinations represent true relationships between data points.

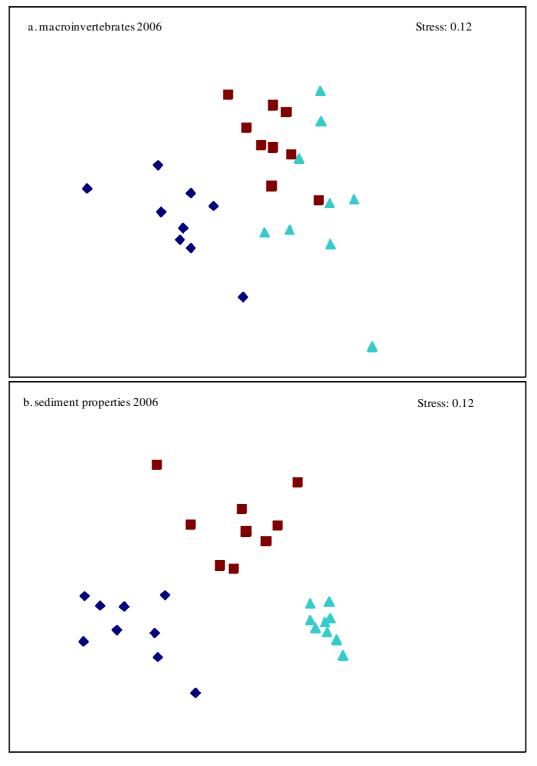
# 3.3 Multivariate analysis of macroinvertebrates and sediment properties in marine national parks (MNPs) only

When MNP sites were compared to each other only, a significant difference in the macroinvertebrate abundances between MNP sites was found in 2006 (one-way ANOSIM, Global R = 0.577, p = 0.001, Figure 3.7a) as well as a significant difference in the sediment properties between MNP sites (one-way ANOSIM, Global R = 0.997, p = 0.001, Figure 3.7b). A significant difference in the macroinvertebrate abundances (one-way ANOSIM, Global R = 0.577, p = 0.001, Figure 3.8a) and the sediment properties (one-way ANOSIM, Global R = 0.773, p = 0.001, Figure 3.8b) between MNP sites was also found in 2007.

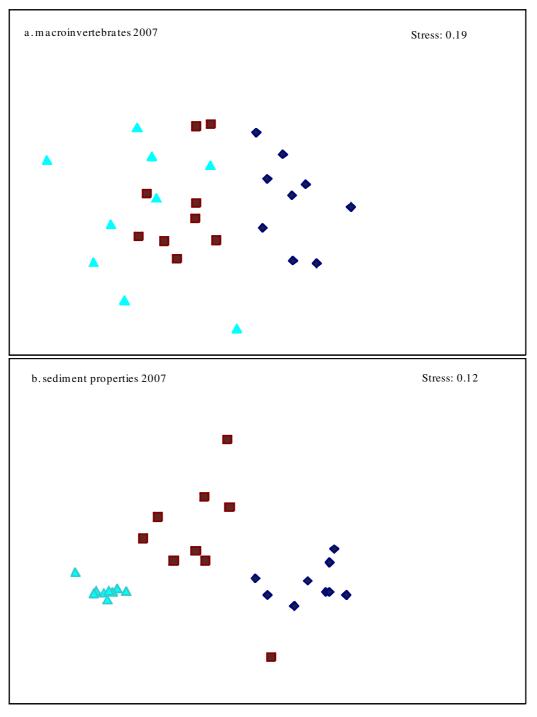
The statistical procedure, SIMPER, showed that there were eight species in 2006 that contributed to differences in the overall macroinvertebrate community composition between MNP sites. The SIMPER analysis showed that Churchill Island MNP and French Island MNP had an average dissimilarity of 93.70 % and *Biffarius arenosus*, *Phoronopsis albomaculata*, *Macrophthalamus latifrons* and *Tellina deltoides* were the species that contributed to 50% of this 93.70% dissimilarity. Churchill Island MNP and Yaringa MNP had an average dissimilarity of 85.90% and *B. arenosus*, *P. albomaculata*, *Barantolla lepte*, *Lumbrineris* sp. and *M. latifrons* contributed to 50% of this 85.90% dissimilarity. Finally, in 2006 there was an average dissimilarity between French Island MNP and Yaringa MNP of 70.80% and *B. lepte*, *M. latifrons*, *Lumbrineris* sp., *B. arenosus*, *Alpheus richardsoni* and *Australonereis elulersi* contributed to 50% of this 70.80% dissimilarity.

In 2007, the SIMPER procedure showed that there were again eight species that contributed to differences in macroinvertebrate community composition between MNP sites. The analysis showed that Churchill Island MNP and French Island MNP had an average dissimilarity of 83.38% and that *M. latifrons, B. arenosus, P. albomaculata, B. lepte* and *Lumbrineris* sp. contributed to 50% of this dissimilarity. Churchill Island MNP and Yaringa had an average dissimilarity of 75.36% and *B. lepte, B. arenosus, M. latifrons, P. albomaculata* and *Lumbrineris* sp. contributed to 50% of this dissimilarity. Finally in 2007, there was an average dissimilarity of 69.07% between French Island MNP and Yaringa MNP and *B. lepte, B. arenosus, M. latifrons, P. albomaculata, Lumbrineris* sp., Sipunculan sp. 2 and *Armandia* sp. MoV 282 contributed 50% to this dissimilarity.

A comparison between the sediment properties and the macroinvertebrate abundances in 2006 using the BVSTEP routine found that there was a significant correlation between the similarities in macroinvertebrates and the similarities in sediment properties between MNP sites (Spearman's rho = 0.697, p = 0.001, Figure 3.7). The sediment property that could best explain the differences in macroinvertebrate abundances between MNP sites was TOC (rho = 0.697). In 2007, BVSTEP again showed that there was a significant correlation between the similarities in sediment properties and the similarity in macroinvertebrate abundances between MNP sites (Spearman's rho = 0.536, p = 0.001, Figure 3.8). TOC and redox potential at 10 cm depth were the two sediment properties in 2007 that could best explain the differences in macroinvertebrate abundances between MNP sites.



**Figure 3.7** MDS ordinations of a. Bray-Curtis similarity matrix of macroinvertebrate abundances (square root transformed) and b. Euclidean distance matrix of sediment properties (natural log transformation of TOC, all normalised) for 2006 samples. Diamonds represent Churchill MNP, triangles represent French Island MNP and squares represent Yaringa MNP. The two-dimensional stress values indicate how well the ordinations represent true relationships between data points.



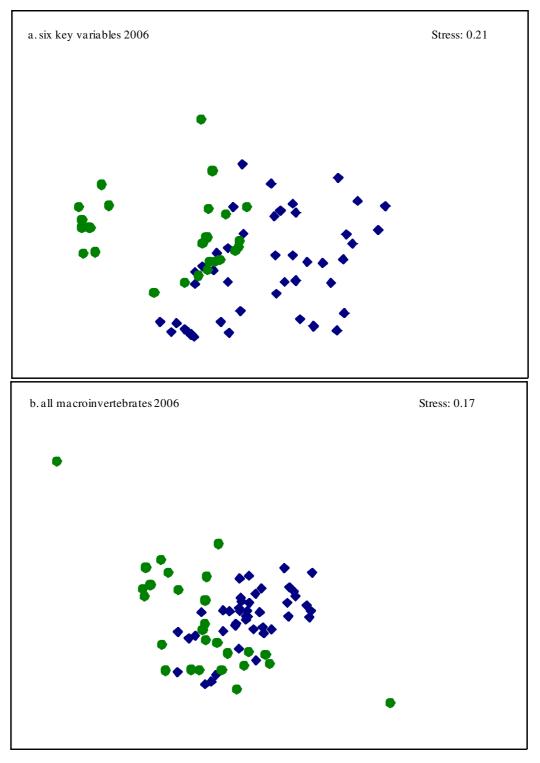
**Figure 3.8** MDS ordinations of a. Bray-Curtis similarity matrix of macroinvertebrate abundances (square root transformed) and b. Euclidean distance matrix of sediment properties (natural log transformation of TOC, all normalised) for 2007 samples. Diamonds represent Churchill MNP, triangles represent French Island MNP and squares represent Yaringa MNP. The two-dimensional stress values indicate how well the ordinations represent true relationships between data points.

#### **3.4** Assessing the subset of key variables

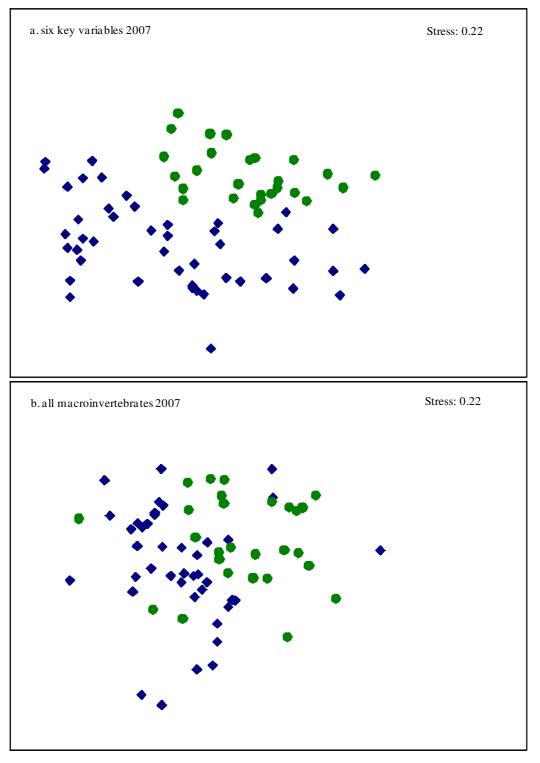
As explained in the methods (section 2.5), a subset of variables as identified throughout the analysis of all variables was statistically analysed to see if this subset represented the same differences between sites as seen with the full dataset. The rationale is given in detail in section 2.5. Briefly, eight variables (species *Trypaea australiensis, Biffarius arenosus, Macrophthalmus latifrons, Lumbrineris* sp., *Barantolla lepte* and sediment properties TOC, substrate temperature, fine sediment particles) were identified as important in contributing to all differences between MNP and external sites and between MNP sites only in 2006 and 2007. Of these eight, six key variables (species *Trypaea australiensis, Biffarius arenosus, Macrophthalmus latifrons, Lumbrineris* sp., and sediment properties TOC and substrate temperature) were chosen because they are easy to sample. *Barantolla lepte* and fine sediment particles were excluded because they are difficult and time consuming to process. The results comparing the differences in these key variables between MNP and external sites and MNP sites only are presented here.

The subset of key variables showed differences between MNP and external sites similar to that found for the complete data set in 2006 (one-way ANOSIM Global R = 0.191, p = 0.001) (Figure 3.9) and in 2007 (one-way ANOSIM Global R = 0.0.353, p = 0.001) (Figure 3.10). A significant correlation was found between the similarity matrix of the subset of variables and the similarity matrix for all macroinvertebrate abundances in both 2006 (RELATE, Spearman's rho = 0.475, p = 0.001) and in 2007 (RELATE, Spearman's rho = 0.427, P = 0.001). This gives statistical evidence that differences between MNP and external sites shown with all of the macroinvertebrate data can be represented well by only a subset of key variables. The differences between MNP and external sites were the same with only the subset of variables as they were with the full data set of macroinvertebrate abundances.

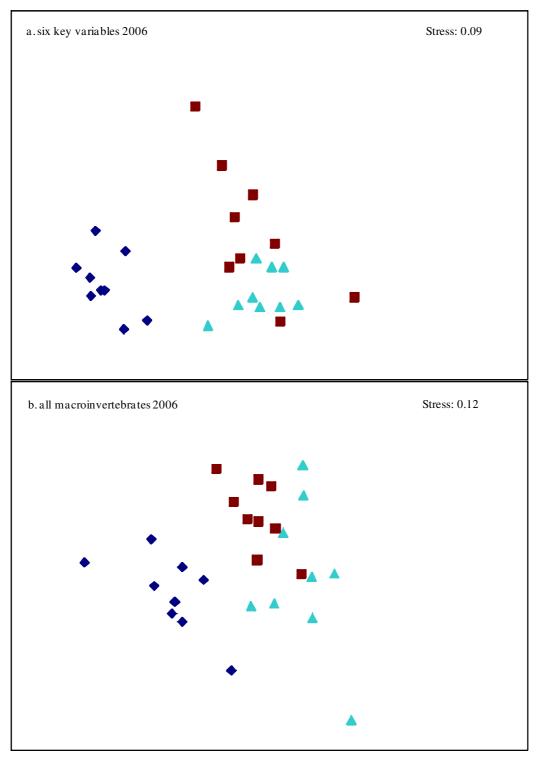
A comparison of MNP sites using the subset of key variables also showed that there was a significant difference between MNP sites in 2006 (one-way ANOSIM, Global R = 0.717, p = 0.001) (Figure 3.11) and in 2007 (one-way ANOSIM, Global R = 0.518, p = 0.001) (Figure 3.12). A significant correlation was found between the similarity matrix of the subset of variables and the similarity matrix of all macroinvertebrate abundance data in 2006 (RELATE, Spearman's rho = 0.624, p = 0.001) and in 2007 (RELATE, Spearman's rho = 0.527, p = 0.001). Again, this supports the findings that the differences between MNP sites in the subset of variables and the differences between MNP sites in all macroinvertebrates abundances are the same.



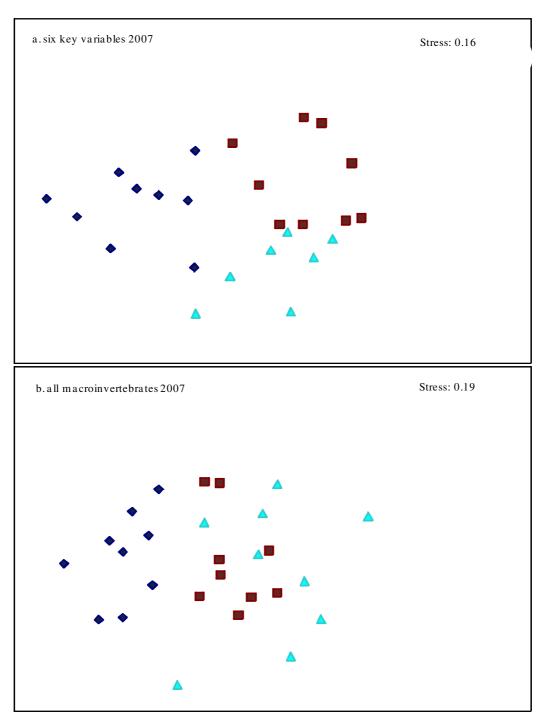
**Figure 3.9** MDS ordinations of a. Euclidean distance matrix of six key variables in 2006 and b. Bray-Curtis similarity matrix of macroinvertebrate abundances (square root transformed) in 2006. MNP sites are represented by circles and external sites are represented by diamonds. The two-dimensional stress values indicate how well the ordinations represent true relationships between data points.



**Figure 3.10** MDS ordinations of a. Euclidean distance matrix of six key variables in 2007 and b. Bray-Curtis similarity matrix of macroinvertebrate abundances (square root transformed) in 2007. MNP sites are represented by circles and external sites are represented by diamonds. The two-dimensional stress values indicate how well the ordinations represent true relationships between data points.



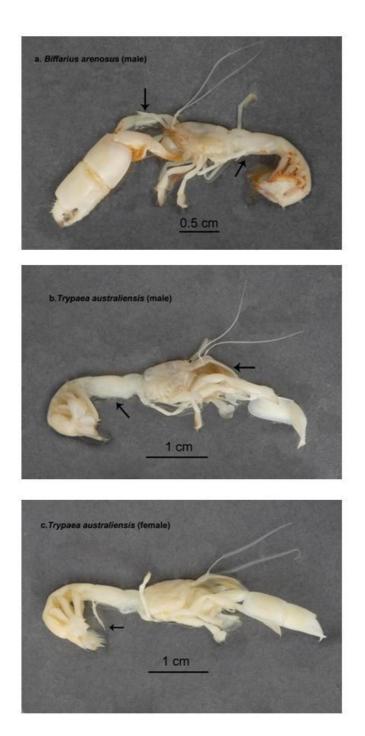
**Figure 3.11** MDS ordinations of a. Euclidean distance matrix of the six key variables (normalised) in 2006 and b. Bray-Curtis similarity matrix of macroinvertebrate abundances (square root transformed) in 2006. Diamonds represent Churchill MNP, triangles represent French Island MNP and squares represent Yaringa MNP samples. The two-dimensional stress values indicate how well the ordinations represent true relationships between data points.



**Figure 3.12** MDS ordinations of a. Euclidean distance matrix of the six key variables (normalised) in 2007 and b. Bray-Curtis similarity matrix of macroinvertebrate abundances (square root transformed) in 2007. Diamonds represent Churchill MNP, triangles represent French Island MNP and squares represent Yaringa MNP samples. The two-dimensional stress values indicate how well the ordinations represent true relationships between data points.

#### **3.5 Description of the key variables**

Figures 3.13, 3.14 and 3.15 show photographs of each of the species identified in the subset of key variables. These species are potentially useful for long term monitoring in the softsediment environments of Western Port. The ghost shrimp Biffarius arenosus is shown in Figure 3.13a. Figure 3.13b shows a male *Trypaea australiensis* with the highly setose lower margin of the antennule peduncle highlighted with an arrow. This highly setose antennule is a taxonomic feature of this species. Another arrow shows the lack of a second pleopod, which was used to distinguish between males and females. A female T. australiensis is shown in Figure 3.13c with the second pleopod highlighted with an arrow. These two ghost shrimp species are common thoughout Western Port (Coleman and Poore 1980). Macrophthalamus latifrons is a common shore crab inhabiting intertidal soft-sediment areas (Phillips et al. 2006) (Figure 3.14). Figure 3.14a shows a dorsal view of this crab and its easily identifiable shape and Figure 3.14b shows the ventral side of a male of *M. latifrons*. Lumbrineris sp. is a detritivorous polychaete worm found in many soft-sediment environments (Wilson et al. 1998). The lateral view of the anterior end of the worm is shown in Figure 3.15a and position of the mouth is highlighted with an arrow. The ventral view of the anterior end of *Lumbrineris* sp. is shown in Figure 3.15b again with the mouth highlighted. The dark areas around the mouth are caused by the black chitinous jaws inside the mouth which are sometimes seen everted from the mouth on the pharynx (as in Figure 3.15c). The shape of the jaws is a defining feature of this species.

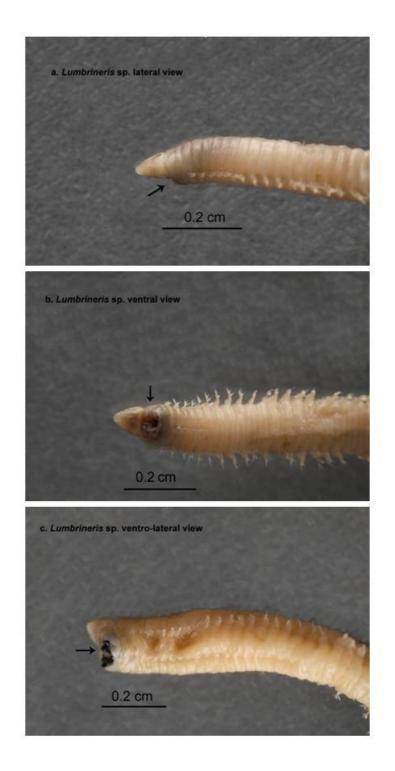


**Figure 3.13** Photographs of a. *Biffarius arenosus* male showing simple antennules (downwards facing arrow) and lack of  $2^{nd}$  pleopods (upwards facing arrow), b. *Trypaea australiensis* male showing heavily setose anntennule (horizontal arrow) and lack of  $2^{nd}$  pleopods (upward facing arrow), c. *T. australiensis* female showing presence of  $2^{nd}$  pleopods (horizontal arrow). Scale bars are given showing *B. arenosus* specimen is much smaller than the two *T. australiensis* specimens.



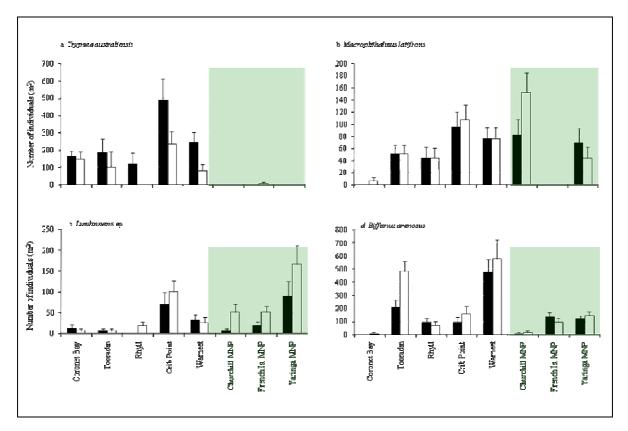


Figure 3.14 Photographs of a. *Macrophthalmus latifrons* dorsal view and b. *M. latifrons* male ventral view. Scale bars are shown.



**Figure 3.15** Photographs of a. *Lumbrineris* sp. lateral view with arrow showing mouth, b, c. *Lumbrineris* sp. ventral and lateral views with arrow pointing to jaws which are slightly everted. All photos show scale bars.

A simple comparison of the number of individuals of the key species in Western Port is given in Figures 3.16. A comparison of the key sediment properties is not discussed here because this is done earlier in section 3.1. Figure 3.16 shows the differences in the density of the key species between sites and may be used to compare with future collections of these key variables as part of a monitoring program. Statistical analysis of this data using a two-way ANOVA shows that there are significant differences between sites for each species (T. australiensis F = 10.931, df = 7, p < 0.001; M. latifrons F = 10.040, df = 7, p < 0.001; *Lumbrineris* sp. F = 9.200, df = 7, p < 0.001; *B. arenosus* F = 20.615, df = 7, p < 0.001, Figures 3.16a-d). There is also a significant differences between the number of T. *australiensis* between years (F = 8.949, df = 1 p = 0.003). There is no significant difference in the number of *M. latifrons*, *Lumbrineris* sp. and *B. arenosus* between years (p > 0.01). No significant interaction effect was found between years and sites for any of the key species (p > 0.01). Post Hoc comparisons showed that there were no consistent differences in the number of each species between MNP sites and sites external to the MNP. However, some general trends can be drawn from Figure 3.16. In general, many more individuals of T. australiensis and B. arenosus were found at sites external to the MNP in Western Port (Figure 3.16a, d). In contrast, there seemed to be more individuals of Lumbrineris sp. found in the MNP sites than sites external to MNPs. There was no general pattern in the number of M. latifrons with a number of individuals being found at every site, except French Island and Coronet Bay.



**Figure 3.16** The four key species identified as important discriminators between the MNP sites and between external sites. Figure 3.17a shows *Trypaea australiensis*, Figure 3.17b shows *Macrophthalmus latifrons*, Figure 3.17c shows *Lumbrineris* sp. and Figure 3.17d shows *Biffarius arenosus*. Solid bars represent data from 2006 and open bars represent data from 2007. Areas shaded highlight the Marine National Park sites. Error bars are one standard error.

# 4 DISCUSSION

The Marine National Parks (MNPs) in Victoria were declared in 2002 to protect extensive intertidal mudflats, mangroves, seagrass beds, saltmarsh areas, soft-sediments and deep channels (Parks Victoria 2003). With the establishment of MNPs in Victoria, came a need to monitor their effectiveness in protecting various aspects of the MNPs' flora and fauna. In the current study, the focus has been to compare the community composition of macroinvertebrates that inhabit the soft-sediment areas and the sediment properties within the MNPs of Western Port, Victoria with areas external to the MNPs in Western Port. This approach was taken to provide biogeochemical information about the MNPs and to address some of the basic monitoring needs for MNPs in Western Port, Victoria.

# 4.1 Differences between MNP sites and external sites and differences between the three MNP sites

In the current study it was found that the sites sampled within the Marine National Parks (MNPs) in Western Port were different from other sites in Western Port and different from one another in terms of the macroinvertebrate communities and sediment properties. However, the differences between MNP and external sites were generally inconsistent. It was expected that the number of species, the number of individuals and the species diversity would be significantly higher within the MNP sites than at the external sites (Halpern & Warner 2002; 2003). This was not the case as the number of species, the number of individuals and the species diversity within the three MNPs sampled was found to be significantly different to some sites in Western Port but not others. In addition, the significant differences were not always the same with some cases revealing that the MNP sites had higher numbers of macroinvertebrate species and individuals.

Similar results were found for the range of sediment properties sampled both within the MNPs and external to the MNPs. Although there were differences in the sediment properties between some MNP sites and some external sites the differences were not the same for all comparisons. For example, while a higher total organic content (TOC) and chlorophyll *a* concentration was found at Churchill Island MNP sites 2 and 3 compared with external sites, no significant differences in the TOC or the chlorophyll *a* concentrations were found between Churchill Island MNP site 1, French Island MNP or Yaringa MNP sites and external sites. Interestingly, nutrients within and external to the MNPs were highly variable between years and did not show any consistency with which MNP and external sites could be compared. This supports various studies that have suggested that marine nutrient levels can fluctuate daily or with changes in invertebrate activity (for examples see Bulthuis *et al.* 1984 and Webb and Eyre 2004), and therefore, sampling once a year, as in the current study, may not be accurate enough to detect any consistent patterns.

There were a number of species that were predominantly collected in the MNP sites and rarely collected outside of MNPs. These were *Alpheus richardsoni, Paragrapsis* sp., Paratanaidae sp., *Armandia* MoV sp. 282, *Musculista senhousia*, and *Phoronopsis albomaculata*. However, there is very little biological information that supports why they might be found only in these areas. All of these species were represented by very low numbers of individuals and may be present elsewhere but not detected in the current study. Previously, *Alpheus richardsoni* was found near Tooradin (north), Lang Lang (east), Hastings (west) and near Rhyll (south) in two benthic surveys of Western Port, one conducted in 1973 and the other conducted in 1985 (see Coleman 1985). Therefore, it is possible that this species has declined in areas outside of MNPs due to human disturbance and is now more commonly present in sites within the MNPs. Further study on this species would be needed to confirm this.

Although trends in species numbers and sediment properties between MNP sites and external sites were not found, multivariate analysis did show that MNP sites had a significantly different overall macroinvertebrate community composition and overall sediment properties when compared to external sites. These significant differences were in some cases relatively small (R = 0.2 - 0.6). As was seen with the univariate comparison of the number of species and the number of individuals between sites, the low R values in multivariate analysis may be a reflection of the large amount of variation between sites. Muotka *et al.* (2002) found that the change in invertebrate communities from assemblages associated with previously disturbed or restored areas to assemblages associated with natural areas of stream beds (*i.e.* restoration rate) was very gradual. If changes in macroinvertebrates in soft-sediments in Western Port are also gradual, then the differences between MNP sites and external sites may not be highly distinct, as is apparent in the current study (low R values). There may be some graduation between external sites and MNP sites making the significant differences between the two small.

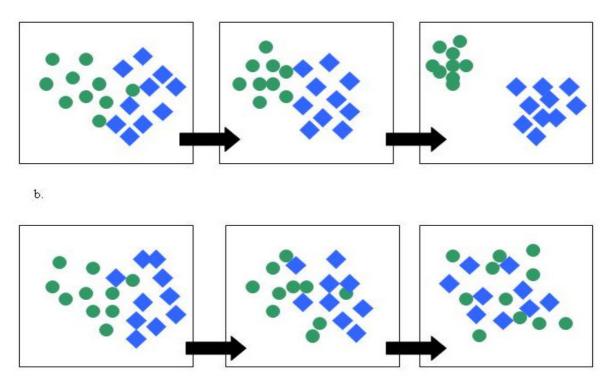
Barrett et al. (2007) found that ten years after declaration, responses to MPAs in Tasmania were species-specific, often complex and varied with external factors surrounding the MPA boundary. Although, they found that some species increased in size and abundance with the boundaries of some MPAs, other species showed no difference. This lack of change was particularly true of species that were not common in commercial or recreational fish catches. If fishes or invertebrates were generally not affected by fishing pressure then they tended to show no response to the protection of the marine reserve. The species that were sampled for in the current study in Western Port, apart from the ghost shrimps, Trypaea australiensis and Biffarius arenosus are not commonly collected for bait or fishing purposes. Therefore, they may not show large changes in their abundance due to the MPAs. Also in Western Port, the areas sampled in the MPAs were on shallow intertidal mudflats that are generally difficult to access and present a barrier to bait collection, human disturbance or fishing nearby. This supports Barrett et al. (2007), who suggests that the level of pressure outside of a particular MPA could have an impact on any changes that would be observed within a MPA. If the MPA was in an area of low fishing pressure, bait collection or human disturbance there would be no obvious changes in the species abundance or size within the MPA.

Despite these small significant differences, multivariate differences between MNP and external sites were evident for both macroinvertebrate assemblages and sediment properties and were consistent between years 2006 and 2007. The significant differences between MNP sites and external sites found in the current study, could be statistically explained by the abundances or concentrations of a subset of key variables (the species *Trypaea australiensis*, *Biffarius arenosus*, *Macrophthalamus latifrons*, *Barantolla lepte*, *Lumbrineris* sp., and the sediment properties substrate temperature, <  $63\mu$ m sediment particle size and total organic content (TOC)). As explained in the results section, *Barantolla lepte* and <  $63\mu$ m sediment particle size, although important variables, were omitted from the subset of key variables for further analysis because they were not suitable candidates for a simplistic monitoring tool (see section 2.5 for details).

The final subset of key variables (the species *Trypaea australiensis*, *Biffarius arenosus*, *Macrophthalamus latifrons, Lumbrineris* sp., and the sediment properties substrate temperature, TOC) were found to reflect differences in macroinvertebrate abundances between the MNP and external sites. The ability of the six key variables to discriminate between the three MNP sites and between the MNP and external sites suggests that in combination, these six variables may be a useful tool for long term monitoring purposes. In the current study it has been shown, that these six key variables could show us the same differences between sites that were shown when all variables were analysed. In order to use these key variables for long term monitoring, temporal samples should be collected to create a long term series of MDS plots, using ANOSIM to compare MNP sites with external sites as was done in the current study. From this it will be possible to determine whether sites within the MNPs are becoming more similar to each other, remaining the same or becoming more similar to external sites. Figure 4.1 shows two hypothetical changes that may be detected

with a long term monitoring program using this approach. A third alternative is that the differences between the sites would not change over time. Figure 4.1a shows the hypothetical case where sites within MNPs becoming more similar to each other and more different to sites external, whereas Figure 4.1b shows MNPs becoming more similar to external sites. This monitoring would be effective in assessing whether the MNPs in Western Port have any affect on changes in the abundance of these large macroinvertebrates and important sediment properties over time and this may indicate changes in other facets of the ecosystem such as community composition.

Gaining further information on these key variables is an important starting point for monitoring in these areas. In particular, these variables may provide a baseline data set that could be used to monitor any changes in the overall soft-sediment environments over time (Smith *et al.* 2007). A large change in any one of these variables may indicate a change in other ecological properties (including community composition) and more detailed sampling can be initiated in response to identification of that change.



**Figure 4.1** Two hypothetical scenarios that may be determined through the use of multivariate analysis, using PRIMER, MDS and ANOSIM. a) shows Marine National Parks (MNP) sites (•) becoming more different to external sites (•), while b) shows MNP sites becoming more similar to external sites.

#### 4.2 The ecological importance of the key variables

The six variables identified as potential monitoring variables are all important components of the ecological processes occurring within the soft-sediment environment. There have been many studies suggesting that ghost shrimps, *Biffarius arenosus* and *Trypaea australiensis* are focal species in soft-sediment environments with the potential to be indicators of species richness, diversity and/ or key sediment properties (Posey *et al.* 1991; Nicholls 2002). It is evident from the current study that the ghost shrimps do in fact contribute strongly to the community composition of macroinvertebrates in Western Port, and the sediment

a.

environment. When ghost shrimps were assessed in combination with other key variables, using multivariate analyses the differences between sites in Western Port can be shown. Ghost shrimps are also important as 'ecosystem engineers' because they can change the sediment environment which can lead to changes in community composition (Berkenbusch and Rowden 2003; Berkenbusch 7 Rowden 2007). They are also important as a food source for migratory shorebirds and commercially important fish species (Robertson 1977) Therefore, sampling ghost shrimps in the subset of key variables would improve knowledge about many processes between benthic invertebrates and other components of softsediment environments in Western Port.

*Macrophthalamus latifrons* is a common ocypodid crab of mudflats of Western Port, Andersons Inlet and Corner Inlet in Victoria, but is not found elsewhere in Victoria or Australia (Phillips *et al.* 2006). It is commonly found burrowing in soft-sediment intertidal areas. There is limited literature on how this species interacts with the soft-sediment environment or other species, but what is known is that it is a very important food resource for a number of internationally important shore bird species. The double-banded plover, *Charadrius bicinctus*, was found to mainly feed on this species in the mudflats around Churchill Island (Churchill Island MNP) and near Rhyll in Western Port (Dann 1991). The rednecked stint (*Calidris ruficollis*) and the curlew sandpipers (*Calidris ferruginea*) were also found to consume *M. latifrons* in Western Port, with up to 33% and 15% of their diets respectively consisting of *M. latifrons* (Dann 1999). Dann (1999) also showed that *M. latifrons* breeds in summer and abundance of this crab tends to be higher in autumn and winter. Further research on this species will provide not only supporting information for the monitoring tool, but also valuable biological information on a very important food resource for Western Port's internationally important shorebirds.

The polychaete worm *Lumbrineris* sp. is generally thought to be a detritivore in soft-sediment environments (Glasby *et al.* 2000). There is very little known about the biology of *Lumbrineris* sp. in Australia. A number of species of this genus have been reported in benthic studies around the world. In Port Phillip Bay, Victoria, one species of the genus *Lumbrineris* (*Lumbrineris* sp. MoV 322) was found to be common in soft-sediments in both a study conducted in the 1970s and in a study in the early 1990s (Wilson *et al.* 1998). In northern Australia, *Lumbrineris* sp. was found to be one of the species initially colonising an area of soft-sediments directly after severe flooding. Although many studies have collected this species and have found it in benthic communities there is very little evidence of its interaction with the soft-sediment. One example where this is not the case is in Japan, where *Lumbrineris* sp. was found to be sensitive to contaminants, occurring only in communities away from contaminant sources (Belan 2003). The sensitivity of members of this genus to contamination supports the further research of this species for the purposes of monitoring. Further study of this species in Western Port would increase the understanding of the role of this species in structuring soft-sediment communities in Western Port.

Organic matter (total organic content) is a food source for many organisms in soft-sediment environments (de Vaugelas 7 Buscail 1990; Ziebis *et al* 1996; Kerr & Corfield 1998; Edgar 2001). In the 1970s, a study in Western Port suggested that the majority of organic matter in the system arose from decaying seagrass and therefore higher concentrations of organic matter were found closer inshore, on beaches or mudflats (Gibbs *et al.* 1976). This study also suggested that another major source of organic carbon in Western Port was input from rivers. TOC is known to limit production (Edgar 1994), abundance (Ford *et al.* 1999) and distribution (Kerr & Corfield 1998) of benthic invertebrate species. Increases in organic content due to human impacts, such as land runoff, can also significantly change the community composition of benthic invertebrates (Widdicombe & Austen 2001). Pearson and Rosenberg (1978) showed that generally there is a decrease in the number of suspension feeders and an increase in the number of deposit feeders with increasing organic enrichment levels. The clearly documented link between TOC and invertebrate communities supports the inclusion of this variable in the monitoring tool in Western Port. Changes in TOC levels would indicate potential changes in food resources for organisms in the MNPs. Substrate temperature, although a discriminating variable for comparing MNP and external sites, was shown to only really differ between years when only MNP sites were compared. This key variable may need further investigation to assess whether it is closely associated with the macroinvertebrate fauna. Perhaps this variable is limited to discrimination between samples collected at different times and not truly between different locations. Collecting all macroinvertebrates on the same day at a number of different locations and recording the temperature differences between sites would allow clarification of whether the sites can be different in temperatures at the exact same sampling time or if the temperature is only different at different sampling times.

As a multivariate subset, this study shows that the key variables can discriminate between MNP and external sites well. However, when the number of species and the amount of each sediment property are compared separately at each site, there were no statistically consistent differences between sites within and external to the MNPs. General trends showed that there were less ghost shrimps, T. australiensis and B. arenosus, and more Lumbrineris sp. found within the MNP than externally. It was expected that the number of ghost shrimps and other species, such as Lumbrineris sp., would be higher in the MNP due the lack of human pressure, particularly bait collection and trampling. This was supported by the general trend for more Lumbrineris sp. to be found in MNP sites. The opposite trend for the ghost shrimp species can be explained by a number of hypotheses. For example, protection of commercial fisheries species through the establishment of MPAs can result in an increase in these fish species and a subsequent decrease in their prey species (Langlois et al. 2005). Ghost shrimps are a common prey item for many commercially important fish species, such as the King George Whiting (Robertson 1977) and therefore, if MNP protection of the King George Whiting in Western Port has resulted in an increase in their numbers, a subsequent decline in ghost shrimps may have occurred. Alternatively, the MNP sites in Western Port were chosen to represent extensive areas of mangrove and seagrass. It has been shown that burrowing by large macroinvertebrates can be inhibited by the structural presence of seagrass and seagrass roots, and therefore, these areas may never have been ideal habitat for ghost shrimps (Brenchley 1982).

As there is no specific data on the macroinvertebrate fauna from the sites within the MNPs prior to their establishment it is difficult to assess whether the trends observed here existed prior to this study or whether the establishment of the MNPs in Western Port has resulted in these differences in key species. Ideally, it would be useful to compare changes in the abundances/ concentrations of all of the six key variables both inside and outside of the MNP over time (*i.e.* monthly). This would support or refute the findings here that show that these six species are useful in showing differences between MNP and external sites and whether the differences between MNP and external sites change over time. It is evident, however, from the current results that the subset of key variables identified are worth further investigation as they are ecologically important components of soft-sediment environments. These six key variables are a good starting point for further refinement of a long term monitoring strategy for the MNPs of Western Port.

# 5 CONCLUSIONS

The MNPs in Victoria were declared in 2002 (Parks Victoria 2003), only four years prior to the current study. It is possible that changes to the sediment environment and the macroinvertebrates that inhabit the sediments, in response to a change in management (*i.e.* no take zones), have not yet occurred. Anthropogenic pressures on intertidal soft-sediment communities outside of MNP areas have been well documented (Hailstone & Stephenson 1961; Wynberg & Branch 1994; Contessa & Bird 2004) and these impacts are thought to cause changes to macrobenthic communities. In particular, bait collection can greatly reduce or change the numbers of benthic species and individuals (Wynberg & Branch 1994). Perhaps more significant differences in communities within the MNPs of Western Port compared with sites outside of the MNPs will be evident after a longer period of time. Over time, changes in community composition may become more apparent as disturbance by bait collectors continues in external sites but remains prohibited within MNP sites. Alternatively, the subtle differences between MNP sites and external sites shown in the current study may be the extent of the difference that will ever be seen for MNPs in Western Port. It is possible that the protection of the MNPs does not directly impact the invertebrates because bay wide processes, such as currents, tidal exchange or pollution are more influential on the distribution of macroinvertebrates and sediment properties in Western Port. Furthermore, Skilleter et al. (2005) showed that macroinvertebrate communities can remain relatively stable even after extended periods of human pressure, such as bait collection and trampling. With growing environmental awareness in our community, management of our coastline both within and external to the MNPs might mean that in future the differences between these areas may be minimal.

In the current study, significant multivariate differences were found in macroinvertebrate abundances and sediment properties between the three MNP sites and between MNP and external sites. The results provide evidence that some differences between these areas do exist although these differences may be more subtle than linear increases or decreases in the number of species or diversity of organisms and sediment properties. Subtle differences between MNP and external sites do provide incentive for continued effort to protect the MNPs from human disturbance because MNPs may facilitate further changes in the macroinvertebrate communities that will protect biodiversity of benthic habitats throughout Victoria. This is one of the main goals of the MNPs. Despite the need for further investigation, the subset of key variables identified in the current study provide an efficient starting point for monitoring the macroinvertebrate communities and sediment properties that are key features of the MNPs in Western Port, Victoria. Further studies focussing on these key variables and their interaction or function with other MNP components will promote our understanding of the delicate marine environment in these areas.

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# **APPENDIX 1**

### A1.1 Species abundance

The abundance of species collected in Yaringa Marine National Park, French Island Marine National Park and Churchill Island Marine National Park

	CHURCHILL IS. MNP 2006	CHURCHILL IS. MNP 2007	FRENCH IS. MNP 2006	FRENCH IS. MNP 2007	YARINGA MNP 2006	YARINGA MNP 2007
Total no. of species per site	12	11	14	21	19	24
Total no. of individuals per site	4037	4065	4063	4063	4076	4098
Decapod crustaceans						
Alpheus richardsoni	1	0	0	1	4	3
Bellidilia laevis	0	0	0	1	1	0
Biffarius arenosus	1	2	21	15	19	23
Ebalia crassipes	0	0	0	0	0	3
Grapsid sp.	1	0	0	0	0	0
Macrophthalamus latifrons	13	24	0	0	11	7
Trypaea australiensis	0	0	0	1	0	0
Amphipods						
Aora mortoni	0	0	0	0	1	0
Melita group sp. 1	0	0	0	0	3	1
Photis sp.	0	0	0	0	1	1
Tipimegus dinjerrus	0	0	0	1	0	0
Tethygeneia megalophthalma	0	0	0	0	0	1

#### Appendix 1 continued

	CHURCHILL IS. MNP 2006	CHURCHILL IS. MNP 2007	FRENCH IS. MNP 2006	FRENCH IS. MNP 2007	YARINGA MNP 2006	YARINGA MNP 2007
Isopods						
Eurydice binda	0	0	0	1	0	0
Natatolana pellucida	0	0	0	1	0	0
Paratanaids						
Paratanaidae sp.	0	0	0	4	0	2
Polychaete worms						
Armandia sp. Mov 282	0	0	2	5	1	0
Australonereis elulersi	0	0	4	4	0	0
Barantolla lepte	0	7	7	8	12	43
Ceratocephale setosa	0	0	3	1	0	0
Goniada antipoda	0	0	1	0	1	0
Glycera ovigera	0	0	0	1	4	0
Glycinde sp. Mov 1403	1	0	0	2	2	0
Lumbrineris sp.	1	8	3	8	14	26
Magelonidae sp.1	0	3	1	0	0	0
Maldane sarsi	4	2	0	0	0	1
Microspio granulate	0	0	1	0	0	0
Nephtys australiensis	2	1	1	0	4	3
Phyllodoce sp. Mov 2876	1	2	0	1	0	0
Poecilochaetus sp. Mov 627	0	0	1	0	0	0
Polycirrus tesselatus	0	0	0	0	1	3
Prionospio aucklaudica	0	1	0	0	0	0
Scalibregma inflatum	0	0	0	0	0	1

#### Appendix 1 continued

MNP 2006	CHURCHILL IS. MNP 2007	FRENCH IS. MNP 2006	FRENCH IS. MNP 2007	YARINGA MNP 2006	YARINGA MNP 2007
0	0	0	1	1	0
0	0	3	0	0	0
0	0	0	0	0	1
0	0	1	0	2	6
0	0	0	0	1	0
0	0	0	1	3	30
0	0	0	2	0	1
0	0	0	0	0	3
7	5	0	0	0	2
0	0	0	1	0	0
0	0	0	0	0	0
2	0	0	0	0	1
0	0	0	0	0	1
0	0	0	0	0	1
19	27	2	2	0	2
0	1	0	0	0	0
	0 0 0 0 0 0 0 0 0 0 0 7 0 0 0 2 0 0 0 19	0 0   0 0   0 0   0 0   0 0   0 0   0 0   0 0   0 0   0 0   0 0   0 0   0 0   7 5   0 0   0 0   2 0   0 0   12 0   0 0   19 27	0 0 0   0 0 3   0 0 0   0 0 1   0 0 0   0 0 0   0 0 0   0 0 0   0 0 0   0 0 0   0 0 0   0 0 0   0 0 0   0 0 0   0 0 0   0 0 0   10 0 0   10 0 0   10 0 0   10 0 0   10 0 0   10 0 0   11 0 0   11 0 0   11 0 0   11 0 0   11 0 0   11 0 0   11 0 0	0   0   1     0   0   3   0     0   0   0   0     0   0   0   0     0   0   1   0     0   0   1   0     0   0   0   0     0   0   0   0     0   0   0   0     0   0   0   0     0   0   0   1     0   0   0   1     0   0   0   2     0   0   0   0     7   5   0   0     0   0   0   1     0   0   0   0     2   0   0   0     0   0   0   0     0   0   0   0     0   0   0   0     0   0   0	0   0   0   1   1     0   0   3   0   0     0   0   0   0   0     0   0   0   0   0     0   0   0   0   0     0   0   1   0   2     0   0   0   0   1     0   0   0   0   1     0   0   0   1   3     0   0   0   1   3     0   0   0   0   0     1   0   0   0   0     0   0   0   0   0     7   5   0   0   0     0   0   0   0   0     1   0   0   0   0     0   0   0   0   0     0   0   0   0   0

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