# BIOLOGICAL MONITORING OF MACROFAUNA IN ARTIFICIAL UNITS OF HABITAT ON INTERTIDAL ROCKY SHORES IN BEROWRA CREEK

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**EXECUTIVE SUMMARY:** Intertidal and subtidal benthic animals are particularly useful for measuring anthropogenic environmental impacts because they are diverse, relatively sedentary and responsive to environmental perturbations. Colonization of Artificial Units of Habitat (AUHs) by these organisms is a useful tool for measuring biodiversity or environmental impacts because AUHs eliminate much of the natural physical variability in habitats among different places. Thus, the assemblages that colonize AUHs are less influenced by natural physical conditions of their immediate surroundings, e.g. grain-size of sediments, or topographic complexity of the rock-surface and, thus, more comparable among locations that differ naturally in features of habitat.

This research programme has extended the study of the macrobenthos colonizing AUHs in mangroves in Berowra Creek (Chapman and Underwood, 2004) to that colonizing AUHs on nearby rocky shores. AUHs in six locations, three considered by Hornsby Council to be subjected to large amounts of urban run-off and three subjected to less run-off, were sampled four times in 2004.

The AUHs developed more diverse assemblage of invertebrates than were found in the AUHs in the mangrove forests. This could be due to characteristics of the different habitats, but could also be confounded by the study of mangroves being in 2003-2004 and that of rocky shores in 2004, or due to the locations of the mangroves being deeper within the embayments than were the rocky shores.

Analyses of the data did do not identify any definitive patterns that could be attributable to differences in water-quality from catchments with little or more urban run-off. This was true at the scale of the entire assemblage and for the diversity and abundances of particular taxa.

From these results and those from the previous study (Chapman and Underwood, 2004), modifications to this project have been recommended for further discussion with Hornsby Council.

## INTRODUCTION

Intertidal macrobenthic animals are frequently used to measure environmental impacts because they are diverse and numerous. In marine and estuarine systems in New South Wales, benthic samples typically consist of hundreds of individuals from dozens of families or phyla. They include predators, grazers, detrital and filter-feeders and may be direct developers, or have larvae with different degrees of dispersal. They are therefore a microcosm of different life-histories, which will respond to different disturbances, such as contamination, changes to temperature or turbidity, etc., in different ways. In addition, they do not move very far and therefore individuals cannot generally move away from sites that are disturbed.

Anthropogenic disturbances can affect the physiological state of the animals, which may result in changes of processes, such as rates of growth (Tablado et al., 1994; Ng and Keough, 2003). More commonly, however, disturbances affect recruitment (Johnston and Keough, 2000, 2002) or mortality (Bryan et al., 1986; Johnston and Keough, 2000) and, therefore, are most easily identified as differences in the numbers and types of animals found in disturbed or undisturbed sites (numerous papers in MEPS, 1988; Smith, 1994, 1996; Chapman et al. 1995).

The spatial and temporal scales necessary to measure diversity and abundances of macrobenthos have been well documented, although benthic invertebrates are notoriously patchy from place to place (Morrisey et al., 1992a; Thrush et al., 1994; Underwood and Chapman, 1996; Hewitt et al., 1998, 2001; Chapman and Tolhurst, 2004). For animals living in sediments, the physical and biochemical condition of the sediment itself, can influence the numbers and types of animals found in them (e.g. Austen et al., 2002; Bolam et al., 2004; Chapman and Tolhurst, 2004). Characteristics of intertidal and subtidal sediment vary naturally from place to place and time to time (Gray, 1974; Morrisey et al., 199a, b; Chapman and Tolhurst, 2004), in addition to potentially being affected by any disturbances. This means that large amounts of replication are necessary to measure changes in fauna caused by anthropogenic disturbances.

The use of artificial units of habitat (AUHs; e.g. pot-scourers, as used by Gee and Warwick (1996) in a world-wide study of marine biodiversity), can create similar physical and chemical structure of habitat in different places (e.g. Hall et al., 2000). Therefore, any differences from place to place in the animals that live in AUHs are likely to be due to differences in which animals arrive into and/or leave the units. They will therefore provide information about ambient environmental conditions that affect rates of recruitment (e,g, whether there are larvae or adults in the water-column, or adults available to breed), or rates at which animals die or leave. Although these animals are relatively sedentary if conditions are suitable, some do enter the water column and drift to new sites when conditions become unsuitable (Cummings et al., 1995).

This research programme is investigating the intertidal macrobenthos in Berowra Creek to test the general model that urban run-off into the creek detrimentally influences the assemblages living there. The specific hypothesis being tested is that there would be differences in fauna among places considered by Council to be subjected to large volumes of urban run-off when compared to places which are considered to have little urban run-off. In 2003-2004, macrobenthos colonizing AUHs were compared between three catchments with large amounts of urban run-off (Sam's Creek, Joe's Craft Bay and Bujwa Bay) and three catchments with less urban run-off (Mount Orient, Kimmerikong Bay and Donnybrook Bay). These AUHs were placed in two sites in each location under the canopy of mangrove trees. In addition, natural assemblages in nearby sediments of the mangrove forests were sampled for comparison with the assemblages colonizing the AUHs.

The AUHs developed a diverse assemblage of invertebrates, which, although not the same as that that occurred naturally in the sediments, was appropriate for detecting environmental impacts. Because of the reduced amount of time needed to process samples from AUHs compared to natural sediments, it was also a cost-effective sampling method. In addition, the assemblages in the AUHs showed smaller amounts of small-scale spatial and temporal ecological pattern ("noise") at scales 1 – 2 m (within sites), ≈ 10 m (between sites) and among locations with similar types of run-off. This should make those assemblages likely to detect smaller impacts than would be detected using natural sediments.

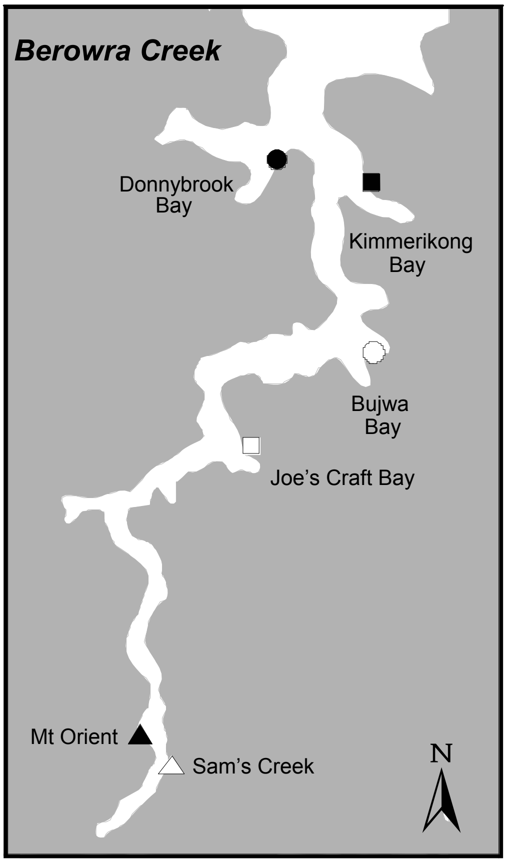


**Figure 1.** Study locations in Berowra Creek; empty symbols represent potentially impacted locations; solid symbols represent control locations.

Nevertheless, the assemblages did not show patterns that could be attributable to differences in water-quality between catchments with little or more urban run-off, although some of the data were indicative of such effects. Each location developed a significantly different assemblage and there was a strong upstream-downstream gradient, despite the uniformity of habitat provided. Some of the graphs suggested minor effects of run-off, but, because all locations differed, no differences could be attributed unambiguously to run-off. All individual taxa were extremely patchy and variable in abundances and the only strong trend was for a smaller number of taxa (mixes of species, families, phyla) in locations with urban run-off.

The original contract with Hornsby Council was to sample mangrove forests and rocky shores in alternate years over a 4-year period Despite the lack of any strong signal in the first year of the study, after consultation with the Council, the requirement was to sample rocky shores, as planned, in 2004. There are very few intertidal rocky shores in Berowra Creek, particularly in the catchments of concern. They are also very small and mainly occupied by oysters and fine green filamentous algae. After further consultation, it was therefore agreed that AUHs would be used on intertidal boulders in the same locations as used in the previous study (Chapman and Underwood, 2004).

## MATERIALS AND METHODS

Five AUHs were deployed in each of two sites (± 30 m apart) in six locations; Mount Orient, Kimmerikong Bay and Donnybrook Bay (little urban run-off) and Sam's Creek, Joe's Craft Bay and Bujwa Bay (considered to have greater urban run-off; Figures 1 and 2). The AUHs were attached directly onto the rock-surface with screws, with at least 1 m between adjacent AUHs (Figure 3). They were collected after approximately 1 month. AUHs were deployed approximately May-Jun, 2004, June-July, 2004, October-November, 2004 and November-December, 2004.

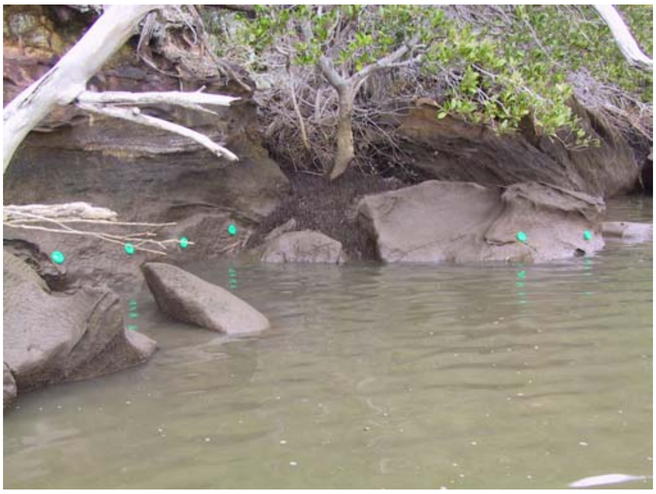

**Figure 2.** Deployment of the AUHs in one of the sites at Bujwa Bay.

Samples were collected by carefully removing the AUH and placing it into a plastic bag in the field. In addition to the assemblage in the AUH, a diverse assemblage of animals had colonized the rock beneath the scourer. Subsamples of this were also incorporated into each sample. In the laboratory, each sample was preserved in 7 % formalin. Prior to being sorted under the microscope, the animals were washed out of the unravelled scourer. If it was estimated that a sample would take more than two hours to sort because of the large numbers of animals present, it was randomly subsampled to a maximum of a 2-hour sorting time. Animals were sorted to a mixed level of resolution, depending on taxonomic expertise and the diversity within the different taxa (as in Chapman and Underwood, 2004).

**Figure 3.** Close-up view of the AUHs *in situ*.

## RESULTS

### Patterns in assemblages

The taxa found in these scourers in each location are summarized in Appendix 1. Only unidentified juvenile crabs, four species of amphipods, three species of isopods, copepods, the mussel, *Xenostrobus securis*, the gastropod, *Assiminea* spp., adult and larval insects and nemerteans were found in all locations. There were between 30 (Donnybrook Bay) and 55 (Mount Orient) taxa per site (of 83 taxa) and no obvious pattern of difference between locations with more or less urban run-off.

To analyses suites of species using multivariate analyses, it is general to reduce any analysis to no more than two factors, because of the complexity of understanding multivariate interactions (Clarke, pers. comm..). Therefore, the hypothesis that assemblages in the two sites in each location would be similar was first tested to determined whether data could be pooled across sites, to test for potential impact, using locations as a second nested factor. For each time separately, the two sites in each location were compared using ANOSIM (Clarke, 1993) based on Bray-Curtis measures of dissimilarity calculated from untransformed data. The Bray-Curtis dissimilarity measure (Bray and Curtis, 1957) gives a measure of differences in assemblages between any two samples, summarizing information about the species present and their relative abundances into a single index. These indices can then be used to measure how assemblages differ among samples, *e.g.* from place to place or time to time. ANOSIM provides a test statistic (*R*) which measures the average difference in an assemblage between sets of samples, compared to the variability of assemblages within these sets. The analyses are based on the ranked data, rather than the dissimilarity values directly and are thus tests of relative differences among samples, rather than absolute differences. The probability level (*P*) is the probability of getting *R* from any set of data if the null hypothesis were true, i.e. the samples came from the same population. Values of $P < 0.05$ indicate that the sets of samples are significantly different (with a 5 % chance of Type I error; Underwood, 1997).

The data were also analysed using npMANOVA (Anderson, 2001), which is a similar procedure but which analyses the Bray-Curtis measures directly, rather than their ranked differences. Because they gave similar results, only the ANOSIM measures are reported here.

There were generally few significant differences between assemblages in the two sites in each location, except for Donnybrook Bay, when sites differed on all occasions (Table 1). In all other locations, sites were generally quite similar.

**Table 1.** Probability levels for comparisons of assemblages in AUHs between sites in each location from ANOSIM calculated from Bray-Curtis dissimilarities using untransformed data; BU – Bujwa Bay, JB – Joe's Craft Bay, SC – Sam's Creek, DB – Donnybrook Bay, KB – Kimmerikong Bay, MO – Mount Orient.

| Times | Between sites | | | | | |
|---|---|---|---|---|---|---|
| | BU | JB | SC | DB | KB | MO |
| Time 1 | > 0.25 | > 0.05 | **< 0.01** | **<0.01** | > 0.05 | > 0.25 |
| Time 2 | > 0.25 | > 0.05 | ND | **< 0.01** | > 0.05 | **< 0.05** |
| Time 3 | > 0.25 | **< 0.05** | **< 0.01** | **< 0.01** | > 0.25 | > 0.25 |
| Time 4 | **< 0.01** | > 0.25 | > 0.05 | **< 0.05** | **< 0.05** | > 0.25 |

The species that each contributed more than 10% to differences between sites for those locations and times of sampling when sites did differ significantly are summarized in Table 2. A value of 10 % is an arbitrary cut-off value that identifies those species that show very large spatial variability, while excluding the majority of the assemblage. Of the 83 taxa, only 8 contributed considerably to this small-scale variation within locations and, of these, only the mussel, *X. secures* and Amphipod sp. 1, were consistently important among locations.

Therefore, because sites were generally not different and only few of the taxa contributed to differences between sites when significant differences were found, the potentially impacted locations were compared to the controls, using a 2-factor nested npMANOVA, with Impact/Control as a fixed factor and Locations as a second nested factor, with *n* = 10, ignoring sites as a factor. This was done to reduce the analyses to two factors, while maximising the number of permutations possible for tests of significance. (ANOSIM was not used because it does not allow enough permutations for a test of significance for the main effect). These analyses were done for each time separately and either including all taxa, or omitting *X. secures* and Amphipod sp. 1 (to reduce differences between sites). For Time 2, data from Mount Orient and Sam's Creek were omitted because all of the scourers from one site at Sam's Creek were lost.

**Table 2.** The taxa that contributed more than 10 % to differences between sites (and the percentage of their contribution) for locations and sampling times where sites were significantly different; BU – Bujwa Bay, JB – Joe's Craft Bay, SC – Sam's Creek, DB – Donnybrook Bay, KB – Kimmerikong Bay, MO – Mount Orient.

| | BU | JB | SC | DB | KB | MO |
|---|---|---|---|---|---|---|
| *X. securis* | 30% (T4) | 12% (T3) | | 12% (T4) | 26% (T4) | 10% (T2) |
| Amphipod sp. 1 | | 15% (T3) | 18% (T1) | | 10% (T4) | 27% (T2) |
| | | | 15% (T3) | 33% (T2) | | |
| Amphipod sp. 3 | | | | 11% (T1) | | |
| | | | | 15% (T3) | | |
| | | | | 19% (T4) | | |
| Amphipod sp. 58 | | | 11% (T1) | | | |
| Isopod sp. 2 | | | | 28%(T1) | | |
| | | | | 17% (T2) | | |
| Isopod sp. 5 | | | 16% (T1) | | | |
| Isopod sp. 8 | | | | 17% (T3) | | |
| Insect larvae | | | 11% (T1) | | | 12% (T2) |

Although all analyses showed significant differences in assemblages among locations, there were no consistent differences between assemblages in potentially impacted locations and those in control locations (all *P* values > 0.05). This is clearly illustrated in Figure 4, where, at no time of sampling, did the potentially impacted locations (empty symbols) plot separately from the control locations (filled symbols).

Environmental impacts can also be identified as differences in temporal or spatial variability in assemblages (Underwood, 1991). It has been suggested that increased variability reflects increased levels of disturbance (Warwick and Clarke, 1993), although some studies show that disturbance can decrease variability (Chapman et al., 1995). Decreased variability could certainly be expected if populations in the disturbed locations are very depressed so there are very small abundances of animals.

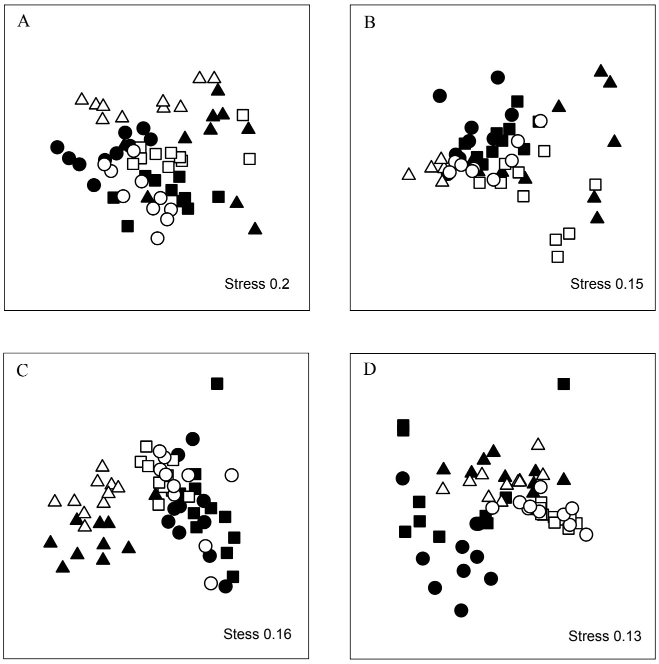


**Figure 4.** nMDS plots of assemblages in scourers for Mount Orient (▲), Sam's Creek (△), Kimmerikong Bay (■), Joe's Craft Bay (□) Donnybrook Bay (●) and Bujwa Bay (○) for 4 times of sampling (A to D).

To test the model(s) that these assemblages respond to increased urban run-off by changing either their degree of spatial or temporal variability, the average Bray-Curtis measures of dissimilarity were calculated for 3 spatial scales and one temporal scale. These were (a) among scourers within each site, (b) between sites (averaged across all locations and times for the potentially impacted and control locations, respectively), (c) among locations (averaged across all times for the potentially impacted and control locations, respectively) and (d) among times (averaged across all locations for the potentially impacted and control locations, respectively).

Model (a) was tested formally by analysing the average within-site variability for each location and time, using a 3 factor analysis of variance. Although there were no significant differences among between potentially impacted and control locations ($P > 0.05$), there was less variability within the potentially impacted sites (mean 45 % dissimilarity) compared to the control sites (mean 53 % dissimilarity), between sites in the potentially impacted locations (53 %) compared to the control locations (66 %) and among the potentially impacted locations (63 %) than among the control locations (74 %). This is indicated in Figure 5. The points that represent the control locations (**▲,■, ●**) plot closer together than the points that represent the potentially impacted locations (△, □, ○**).**

Temporal variability in the assemblages was similar in both sets of locations; 73 % and 75 % for the controls and potentially impacted locations, respectively.

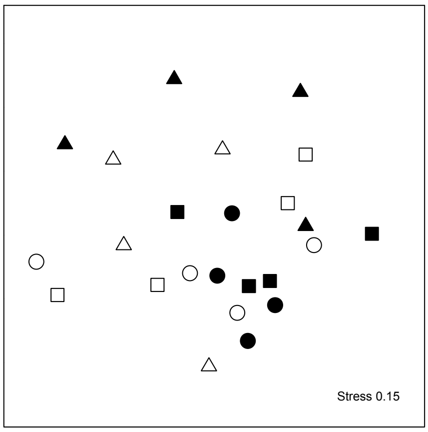


**Figure 5.** nMDS plots of the average assemblages in AUHs for Mount Orient (▲), Sam's Creek (△), Kimmerikong Bay (■), Joe's Craft Bay (□) Donnybrook Bay (●) and Bujwa Bay (○) for 4 times of sampling; each point represents the assemblage in one location at one time.

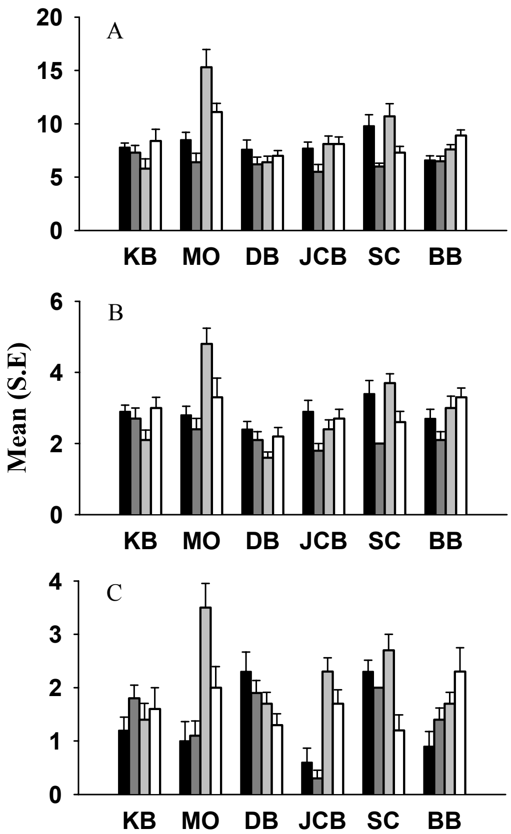


**Figure 6.** Mean (S.E.) number of taxa, species of amphipods and species of isopods in scourers for each location and each time of sampling, represented by the four bars; BU – Bujwa Bay, JB – Joe's Craft Bay, SC – Sam's Creek, DB – Donnybrook Bay, KB – Kimmerikong Bay, MO – Mount Orient.

## Patterns in individual taxa

The most diverse taxa in the AUHs were amphipods and isopods (Appendix 1). To test the model that the numbers of types of these and the number of all taxa differed between locations with or without large amount of urban run-off and that these patterns were consistent through time, the numbers of types of each of these taxa were analysed using 4-factor analyses of variance (Factor 1, Time, random; Factor 2, much/little run-off, fixed and orthogonal; Factor 3, locations, random and nested in Factor 2; Factor 4, sites, random and nested in Factor 3; $n$ = 5). The analyses are presented in Appendix 2.

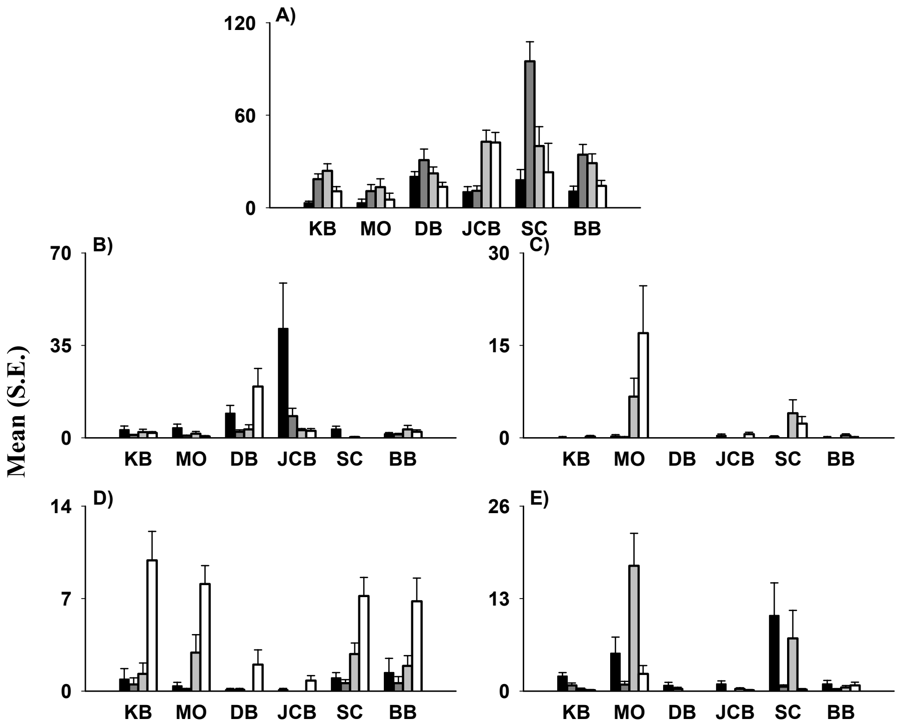


**Figure 7.** Mean (S.E.) number of (A) Amphipod 1, (B) Amphipod 3, (C) Amphipod 5, (D) Amphipod 21 and (E) Amphipod 58 in AUHs for each location and each time of sampling, represented by the four bars; BU – Bujwa Bay, JB – Joe's Craft Bay, SC – Sam's Creek, DB – Donnybrook Bay, KB – Kimmerikong Bay, MO – Mount Orient.

Because of the complex sampling design with times, locations and sites random factors, tests of the main effect of run-off/no run-off can only be done if interaction terms can be pooled. Although the number of taxa, the number of species of amphipods and the number of species of isopods did not vary through time at any spatial scale (Appendix 2) and pooling could proceed (as per Underwood, 1997), there was still no significant effect of much/little run-off. All groups showed variability from time to time in their numbers at the scale of sites and/or locations, but there were no main effects of much run-off *versus* little urban run-off.

There were also no clear patterns in abundances (Figure 6) that warranted further analyses (e.g. binomial tests, as done in Chapman and Underwood, 2004).

To evaluate any effects of run-off on abundances of individual taxa, the most widespread and relatively abundant species (generally identified as morphospecies *sensu* Oliver and Beattie, 1996) or larger taxonomic groups were similarly analysed. They were selected if they had a mean density > 1 per AUH and were present in at least 10 % of the AUHs. These included 5 (morpho)species of amphipods, 3 (morpho)species of isopods, juvenile crabs, insect larvae and the mussel, *Xenostrobus securis* (Appendix 3).

Abundances were very patchy (Figures 7 – 9), although most taxa were found in reasonable densities in most of the places at most of the times. Some taxa were really abundant at only one of the times, e.g. juvenile crabs at Time 1 (Figure 9C), or Amphipod 21 (Figure 7D) and *X. securis* at Time 4 (Figure 9A).

No taxon showed any significant differences between locations with more urban run-off (on average) and those with less run-off (Appendix 3), although many showed considerable temporal variation at the scale of sites and/or locations. Not only did these analyses show non-significant effects of run-off, the Mean Square estimates for this term were very small. There was therefore no indication of any effect that should be investigated using any other analyses.

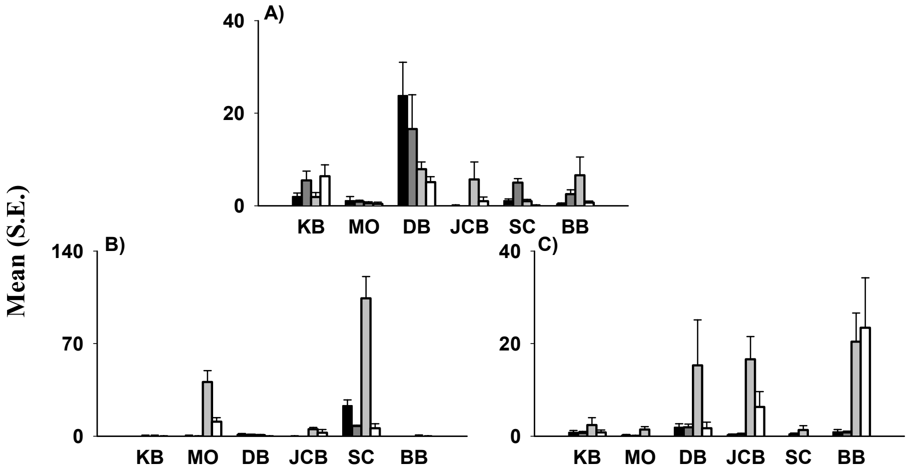


**Figure 8.** Mean (S.E.) number of (A) Isopod 2, (B) Isopod 5 and (C) Isopod 8 in AUHs for each location and each time of sampling, represented by the four bars; BU – Bujwa Bay, JB – Joe's Craft Bay, SC – Sam's Creek, DB – Donnybrook Bay, KB – Kimmerikong Bay, MO – Mount Orient.

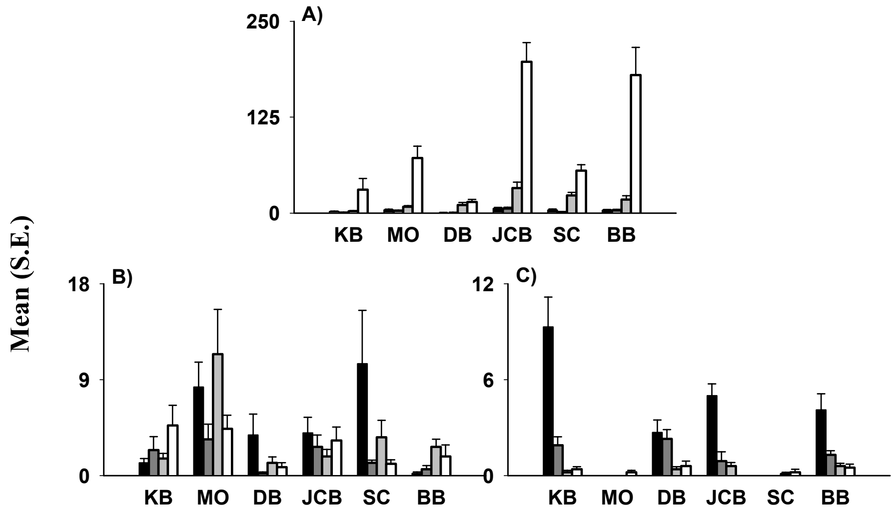


**Figure 9.** Mean (S.E.) number of (A) the mussel, *Xenostrobus securis*, (B) insect larvae and (C) juvenile crabs in AUHs for each location and each time of sampling, represented by the four bars; BU – Bujwa Bay, JB – Joe's Craft Bay, SC – Sam's Creek, DB – Donnybrook Bay, KB – Kimmerikong Bay, MO – Mount Orient.

## DISCUSSION

AUHs, in the form of plastic pot-scourers placed on rocky boulders along the edge of Berowra Creek developed very diverse assemblages of invertebrates, with about 70 % more taxa than colonized similar AUHs in the mangrove forest in 2003-2004 (Appendix 4). Both assemblages included a wide range of broad taxa (polychaetes, gastropods, bivalves, amphipods, etc.), but in this study there were more representatives of a broad range of taxa.

There are three possible explanations for differences in these assemblages. First, AUHs on rocky shores may be colonized by more taxa because more taxa are attracted to AUHs on shores than in mangroves and/or fewer taxa die in AUHs on rocky shores. If true, this would suggest that AUHs on rocky shores are very good substrata to measure local diversity of benthic macrofauna.

Second, the pattern could, however, be due to differences in the locations of the two types of habitats. The mangrove forests used in 2003-2004 (Chapman and Underwood, 2004) were chosen to be, wherever possible, at the heads of the embayments. This is where the mangrove forests are most extensive and where run-off was reported to be most concentrated (J. Grove, Hornsby Council). There were no hard substrata on which to attach the AUHs at similar distances into the embayments and near the mangrove forests. Therefore, these AUHs were attached to boulders, which were closer to the main stream of Berowra Creek. Therefore, differences in colonization between the two sets of AUHs could simply have been due to proximity of the AUHs to the main flow of water through Berowra Creek.

Third, the two studies were done in different years and, therefore, differences in colonization could simply have been due to different supplies of larvae in the two years. This seems, however, less likely to be the explanation because similar patterns were found in the four different time periods sampled each year.

As in the study in mangroves in 2003-2004 (Chapman and Underwood, 2004), the assemblages colonizing these AUHs did not show any definitive patterns that could be attributable to variations in water-quality from catchments with little or more urban run-off. This was true for the proportion of taxa present in the different locations, measures of the entire assemblage, diversity of amphipods and isopods and abundances of the most widespread and abundant organisms. This was supported by multivariate and univariate analyses and visual inspection of trends.

Unlike the AUHs in the mangroves, where there was a strong upstream-downstream gradient in the assemblages (Chapman and Underwood, 2004), assemblages in the AUHs in this study generally overlapped considerably (except for Mount Orient and Sam's Creek at Time 3; Figure 4C), although analyses showed some significant differences among some locations within each group at some times. Along with the large diversity of fauna colonizing these AUHs, the lack of a strong upstream-downstream gradient in these assemblages shows that they should be a good "indicator" of anthropogenic impact, by minimizing natural variability ("noise") among locations, thus making it more likely to detect statistically any impact. This justifies using AUHs instead of natural samples.

At the scale of diversity or abundance of individual taxa, all analyses were complicated by small-scale variation in abundances of individual taxa, as is the norm for the benthos in soft sediments (e.g. Morrisey et al., 1992a, b; Thrush et al., 1994; Chapman and Tolhurst, 2004). The data were analysed with Time as a single factor, rather than dividing the times into Summer and Winter, each with two times of sampling. This was done because there were no clear seasonal patterns in most of the data.

Treating Time as a single factor simplified the analyses, allowing pooling to get a test for Much/Little Run-off (Underwood, 1997). Although this test was not very powerful, the magnitudes of the Mean Square estimates for this term in all of the analyses indicated no major increase in variance due to this factor. Therefore, not only was there no significant effect of run-off in any analyses of any measures, there was no indication from the magnitudes of these measures, or from any of the graphs, that there was potentially an impact that was not being detected. Thus, despite the large diversity of fauna, the spatial replication of sampling at scales from metres to kilometres, the numerous analyses and the fact that there were four independent tests of these hypotheses in this study, there was no evidence of impact from run-off.

The only measure that did indicate some pattern was the smaller (albeit not significantly so) variation among AUHs, particularly within and between sites in the potentially impacted locations. Impacts can alter spatial variability (or the amount of patchiness; e.g. Warwick and Clarke, 1993) and temporal variability (or the amount that populations change through time; e.g. Underwood, 1991). If these changes to natural variance are large enough, they can potentially affect sustainability of biodiversity, but there is inadequate information at this stage to speculate about the importance of the differences found here, especially when the effect of the impact (if there was one) was to reduce, not increase, variability.

The lack of any strong patterns in this study and in Chapman and Underwood (2004) have many explanations:

(1) Despite current understanding by Hornsby Council, there are no differences in urban run-off among these locations, so there is no contamination for the fauna to respond to. This seems unlikely if water-quality has been measured with a rigorous sampling design.

(2) Although run-off does differ among catchments as predicted, the benthos is only affected under extreme conditions and rapidly recovers. If this is correct, it is necessary to identify those extreme events and ensure that they do not start to occur so frequently that recovery cannot proceed before the next event occurs.

(3) Although run-off does differ among catchments as predicted, it does not affect the benthic ecology of the estuary. In this case, it will be necessary to determine at what levels run-off will affect the benthos and ensure that it does not reach those levels.

(4) Fourth, the scale of impact may actually be larger than predicted, so that the locations identified as control locations are actually as impacted as the locations with excess urban run-off. In this case, there is no difference between potentially impacted and control locations, because all, rather than none, are impacted. This may be the case if the major environmental problems in Berowra Creek are from other sources and/or run-off from the small urban catchments are having widespread effects.

## RECOMMENDATIONS

In the light of these results, the following recommendations need discussion for the rest of this project in 2005-2006.

1. The investigation of effects of water-quality in Berowra Creek on the benthos continue with the use of AUHs because they cheaply, easily and reliably sample a diverse benthic fauna, which will show impacts if there are any.
2. We should continue with the AUHs being attached to hard substrata (as in this study) because these developed more diverse assemblages.
3. As suggested in Chapman and Underwood (2004), the spatial components of this study should be increased at the expense of the temporal components. For example, sampling only twice a year would allow two control locations to be sampled for each of the three potentially impacted locations (i.e. replacing 4 times x 4 locations with 2 times x 9 locations) with little increase in funding. The choice of further sites must depend on information about water-quality to be provided by Hornsby Council.
4. Alternatively, for 2005, there could be less effort into identifying impacts from these catchments and more focus on other potential sources of contamination or disturbances. For example, if Berowra Creek is impacted at a large spatial scale, leading to the lack of any differences between what were perceived as impacted and control locations, then the study needs to compare Berowra Creek with external reference waterways. In addition, a broad survey, aimed at characterizing the fauna at many different locations along the creek, rather than to contrast specific potentially impacted and control locations, may also assist in pinpointing where any problem spots may be.
5. We therefore propose a meeting with the Council to discuss the direction forward in this project. This was suggested in 2004, but it was decided by Hornsby Council to wait until the results of the research in 2004 were available. A meeting is now needed to clarify the results so far, the issues of concern and the most cost-effective way forward to increase the "ecological return" for expenditure.

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## APPENDIX 1

The list of taxa and their occurrence in scourers deployed on rocky shores during 2004 in each of the locations.

| Phylum | Class | Order | Family | Organism | Presence in location | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | BU | JB | SC | DB | KB | MO |
| Annelida | Oligochaeta | | | | X | X | X | X | | X |
| | Polychaeta | | Capitellidae | | | X | X | | X | X |
| | | | Chrysopetalidae | | | X | | | | |
| | | | Cirratulidae | | | X | | | | |
| | | | Nereididae | | X | X | X | X | X | X |
| | | | Sabellidae | | | | X | | | X |
| | | | Spionidae | | | X | X | | | X |
| | | | Spirorbidae | | X | X | X | | | X |
| | | | Syllidae | | X | | | X | X | |
| Crustacea | Malacostraca | Decapoda | | Crabs | X | X | X | X | X | X |
| | | | Caprellidea | Amphipod 8 | | | | | X | X |
| | | | Gammaridae | Grey gammarid | | X | X | | X | X |
| | | | Corophiidae | Amphipod 3 | X | X | X | X | X | X |
| | | | | Amphipod 7 | | | X | | | X |
| | | | | Amphipod 40 | | | | | | X |
| | | | | Amphipod 43 | | X | | | | X |
| | | | | Amphipod 54 | | X | X | | | X |
| | | | | Amphipod 61 | X | | X | | | X |
| | | | Ischyroceridae | Amphipod 47 | | | | | | X |
| | | | Eusiridae | Amphipod 4 | | | | | X | |
| | | | | Amphipod 59 | | | X | | | |
| | | | Gammaridae | Amphipod 5 | X | X | X | | X | X |
| | | | | Amphipod 21 | X | X | X | X | X | X |
| | | | | Amphipod 41 | | | | | X | |
| | | | | Amphipod 58 | X | X | X | X | X | X |
| | | | Hyalidae | Amphipod 12 | | | | | | X |
| | | | Phoxocephalidae | Amphipod 9 | | | | | | X |
| | | | Talitridae | Amphipod 1 | X | X | X | X | X | X |
| | | | Hyperiidae | Amphipod 28 | | | | | | X |
| | | | | Unidentified juveniles | X | X | X | | X | X |
| | | Cumacea | | | | | X | | | |
| | | Isopoda | | Isopod 2 | X | X | X | X | X | X |
| | | | | Isopod 3 | X | | | X | | |
| | | | | Isopod 4 | X | X | X | | | X |
| | | | | Isopod 5 | X | X | X | X | X | X |
| | | | | Isopod 6 | | | | | X | |
| | | | | Isopod 7 | | X | X | X | X | X |
| | | | | Isopod 8 | X | X | X | X | X | X |
| | | | | Isopod 9 | X | X | X | | | X |
| | | | | Isopod 12 | X | X | | | | X |
| | | | | Isopod 13 | | | X | | X | X |
| | | | | Isopod 14 | X | | X | | | X |
| | | | | Isopod 15 | | | | X | | |

| **Phylum** | **Class** | **Order** | **Family** | **Organism** | Presence in location | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | BU | JB | SC | DB | KB | MO |
| | | | | Isopod 25 | X | X | X | | | X |
| | | | | Unident.ified isopods | | | | X | X | |
| | | Tanaidacea | | | X | X | | | X | X |
| | Maxillipoda | Cirripedia | Tetraclitidae | *Tesseropora rosea* | | | | | X | |
| | | | | Unidentified barnacles | X | | | X | X | |
| | | Copepoda | | | X | X | X | X | X | X |
| | | Ostracoda | | | | | | | | X |
| Echinodermata | Asteroidea | | Asteriidae | | | | | | X | |
| | Ophiuroidea | | | | X | | | | | |
| Mollusca | Bivalvia | | | | | | X | X | X | X |
| | | | Erycinidae | *Arthritica helmsii* | X | | X | | | X |
| | | | | *Lasaea australis* | | X | X | | | |
| | | | Laternulidae | *Laternula* spp. | | | | X | X | |
| | | | Mytilidae | *Xenostrobus securis* | X | X | X | X | X | X |
| | | | Neculidae | *Leionucula* spp. | | | X | | | X |
| | Gastropoda | | Anabathronidae | *Amphithalamus* spp. | | X | | | | |
| | | | Assiminidae | *Assiminea* spp. | X | X | X | X | X | X |
| | | | Buccinidae | *Nassarius* spp. | | | | | | X |
| | | | Cingulopsidae | *Pseudopisinna gregaria* | | | X | | | |
| | | | Eatoniellidae | *Crassitoniella flammea* | | | | | | X |
| | | | | *Eatoniella atropurpurea* | X | | | | | |
| | | | Hydrobiidae | *Ascorhis* spp. | | | | | | X |
| | | | | *Tatea* spp. | X | | X | | | |
| | | | Littorinidae | *Bembicium auratum* | | X | | X | X | |
| | | | Rissoellidae | *Rissoella* spp. | | | X | | | X |
| | | | Unidentified | Gastropod 26 | | X | X | | | X |
| | | | | Gastropod 66 | | | | | X | |
| | | | | Gastropod 117 | | X | | X | | |
| | | | | Gastropod 118 | | | X | | | |
| | | | | Gastropod 161 | | | | | | X |
| | | | | Gastropod 171 | | | | X | | |
| | | | Acmaeidae | *Patelloida mufria* | | | X | | | |
| | | | | Unidentified | | X | X | X | | X |
| Arthropoda | Insecta | | | Adult insect | X | X | X | X | X | X |
| | | | | Insect larvae | X | X | X | X | X | X |
| | | | | Collembolid | X | X | X | | X | X |
| | Chelicerata | Arachnida | | Spider | | X | | | | X |
| | | | | Mite | X | X | X | X | | |
| Nemertea | | | | | X | X | X | X | X | X |
| Platyhelminthes | | | | Unidentified | X | X | X | X | X | X |

**APPENDIX 2**

Analyses of the mean number of types of the most important taxa and the mean number of widespread and important taxa in scourers in each of the four locations sampled four times in 2003; ***** = *P* < 0.05, ****** = *P* < 0.01, ******* = *P* < 0.001.

| **Source** | | **No. taxa** | | **Spp. amphipods** | | **Spp. isopods** | |
|---|---|---|---|---|---|---|---|
| | **df** | **MS** | ***F*** | **MS** | ***F*** | **MS** | ***F*** |
| Time = T | 3 | 80.1 | 2.04 | 7.33 | 1.86 | 8.91 | 1.59 |
| Run-off = R | 1 | 10.4 | NT | 0.04 | NT | 0.82 | NT |
| Locations(R) = L(R) | 4 | 80.1 | NT | 9.00 | NT | 4.30 | NT |
| Sites(L(R)) = S(L(R)) | 6 | 22.1 | 2.43 | 0.72 | 1.08 | 1.86 | 2.06 |
| T x R | 3 | 1.9 | 0.05 | 1.58 | 0.40 | 0.73 | 0.13 |
| T x L(R) | 12 | 39.2 | 4.31** | 3.95 | 5.93*** | 5.60 | 6.20*** |
| T x S(L(R)) | 18 | 9.1 | 1.72* | 0.67 | 0.78 | 0.90 | 1.10 |
| Residual | 192 | 5.3 | | 0.85 | | 0.82 | |
| Test for R against L(R) after eliminating T x R | | | 0.13 | | 0.00 | | 0.19 |
| Cochrans *C* | | 0.14** | | 0.11** | | 0.08 | |
| Transform | | None | | None | | None | |

## APPENDIX 3

Analyses of the mean number of all taxa that were present with mean density > 1 and were found in > 10 5 of all scourers; ***** = *P* < 0.05, ****** = *P* < 0.01, ******* = *P* < 0.001.

| **Source** | | **Amphipod 1** | | **Amphipod 3** | | **Amphipod 5** | | **Amphipod 21** | |
|---|---|---|---|---|---|---|---|---|---|
| | **df** | **MS** | ***F*** | **MS** | ***F*** | **MS** | ***F*** | **MS** | ***F*** |
| Time = T | 3 | 7.40 | 4.17 | 3.72 | 3.74* | 1.98 | 2.29 | 13.38 | 23.84*** |
| Run-off = R | 1 | 10.51 | NT | 0.00 | NT | 0.00 | NT | 0.63 | NT |
| Locations(R) = L(R) | 4 | 5.02 | NT | 7.66 | NT | 3.25 | NT | 4.56 | NT |
| Sites(L(R)) = S(L(R)) | 6 | 2.03 | 4.94** | 1.82 | 4.35** | 0.57 | 2.66 | 0.42 | 2.56 |
| T x R | 3 | 0.04 | 0.02 | 0.56 | 0.57 | 0.08 | 0.09 | 0.31 | 0.55 |
| T x L(R) | 12 | 1.77 | 4.32** | 0.99 | 2.38* | 0.86 | 4.00** | 0.56 | 3.45** |
| T x S(L(R)) | 18 | 0.41 | 1.23 | 0.42 | 1.23 | 0.22 | 1.04 | 0.16 | 0.62 |
| Residual | 192 | 0.33 | | 0.34 | | 0.21 | | 0.26 | |
| Test for R against L(R) | | | | | | | | | |
| after eliminating T x R | | | 2.09 | | 0.00 | | 0.00 | | 0.14 |
| Cochrans *C* | | 0.08 | | 0.08 | | 0.20** | | 0.06 | |
| Transform | | $X^{0.5}$ | | $X^{0.5}$ | | $X^{0.5}$ | | $X^{0.5}$ | |

| **Source** | | **Amphipod 58** | | **Isopod 2** | | **Isopod 5** | | **Isopod 8** | |
|---|---|---|---|---|---|---|---|---|---|
| | **df** | **MS** | ***F*** | **MS** | ***F*** | **MS** | ***F*** | **MS** | ***F*** |
| Time = T | 3 | 4.75 | 3.41 | 2.26 | 2.76 | 10.69 | 3.58* | 3.75 | 5.26* |
| Run-off = R | 1 | 0.48 | NT | 13.90 | NT | 4.12 | NT | 2.31 | NT |
| Locations(R) = L(R) | 4 | 5.05 | NT | 9.56 | NT | 24.33 | NT | 5.07 | NT |
| Sites(L(R)) = S(L(R)) | 6 | 0.45 | 1.80 | 1.57 | 4.33* | 1.83 | 5.52** | 1.14 | 2.13 |
| T x R | 3 | 0.21 | 0.15 | 0.67 | 0.82 | 0.87 | 0.29 | 2.40 | 3.37 |
| T x L(R) | 12 | 1.39 | 5.62*** | 0.82 | 2.27 | 2.99 | 9.03*** | 0.71 | 1.33 |
| T x S(L(R)) | 18 | 0.25 | 0.81 | 0.36 | 1.06 | 0.33 | 1.70* | 0.54 | 1.15 |
| Residual | 192 | 0.31 | | 0.34 | | 0.19 | | 0.47 | |
| Test for R against L(R) | | | | | | | | | |
| after eliminating T x R | | | 0.10 | | 1.45 | | 0.17 | 0.46 | |
| Cochrans *C* | | 0.10 | | 0.10 | | 0.11 | | 0.10 | |
| Transform | | $X^{0.5}$ | | $X^{0.5}$ | | $X^{0.5}$ | | $X^{0.5}$ | |

| **Source** | | **Crabs** | | **Insect larvae** | | ***X. securis*** | |
|---|---|---|---|---|---|---|---|
| | **df** | **MS** | ***F*** | **MS** | ***F*** | **MS** | ***F*** |
| Time = T | 3 | 4.79 | 4.04* | 1.07 | 1.24 | 39.90 | 47.31*** |
| Run-off = R | 1 | 0.02 | NT | 0.13 | NT | 24.05 | NT |
| Locations(R) = L(R) | 4 | 6.12 | NT | 3.91 | NT | 3.70 | NT |
| Sites(L(R)) = S(L(R)) | 6 | 0.37 | 1.62 | 2.22 | 3.00* | 0.63 | 1.47 |
| T x R | 3 | 0.53 | 0.44 | 0.16 | 0.20 | 2.71 | 3.21 |
| T x L(R) | 12 | 1.19 | 5.21*** | 0.81 | 1.10 | 0.84 | 1.98 |
| T x S(L(R)) | 18 | 0.23 | 1.21 | 0.74 | 2.18** | 0.43 | 1.45 |
| Residual | 192 | 0.19 | | 0.34 | | 0.29 | |
| Test for R against L(R) | | | | | | | |
| after eliminating T x R | | | 0.00 | | 0.03 | | 6.50 |
| Cochrans *C* | | 0.07 | | 0.05 | | 0.13* | |
| Transform | | $X^{0.5}$ | | $X^{0.5}$ | | $X^{0.55}$ | |

**APPENDIX 4**

The list of taxa and their occurrence in scourers deployed in mangroves in 2003 and on rocky shores in 2004.

| Phylum | Class | Order | Family | Organism | Presence in Habitat | |
|---|---|---|---|---|---|---|
| | | | | | Mangrove | Rocky shore |
| Nemertea | | | | | X | X |
| Annelida | Oligochaeta | | | | X | X |
| | Polychaeta | | Capitellidae | | | X |
| | | | Chrysopetalidae | | | X |
| | | | Cirratulidae | | | X |
| | | | Nereididae | | X | X |
| | | | Sabellidae | | | X |
| | | | Spionidae | | | X |
| | | | Spirorbidae | | X | X |
| | | | Syllidae | | X | X |
| Crustacea | | | | Other crustacean | X | |
| | Malacostraca | Decapoda | | Crabs | X | X |
| | | Amphipoda | Caprellidea | | X | |
| | | | | Amphipod 8 | | X |
| | | | Corophiidae | Amphipod 3 | X | X |
| | | | | Amphipod 7 | | X |
| | | | | Amphipod 40 | X | X |
| | | | | Amphipod 43 | | X |
| | | | | Amphipod 54 | | X |
| | | | | Amphipod 61 | | X |
| | | | Ischyroceridae | Amphipod 47 | | X |
| | | | Eusiridae | Amphipod 4 | | X |
| | | | | Amphipod 59 | | X |
| | | | Gammaridae | Grey gammarid | X | X |
| | | | | Amphipod 5 | X | X |
| | | | | Amphipod 21 | | X |
| | | | | Amphipod 41 | X | X |
| | | | | Amphipod 58 | | X |
| | | | Hyalidae | Amphipod 12 | | X |
| | | | Phoxocephalidae | Amphipod 9 | X | X |
| | | | Talitridae | Amphipod 1 | X | X |
| | | | Hyperiidae | Amphipod 28 | | X |
| | | | | Unident. Amhipod | X | |
| | | Cumacea | | | | X |
| | | Isopoda | | Isopod 2 | X | X |
| | | | | Isopod 2b | X | |
| | | | | Isopod 3 | X | X |
| | | | | Isopod 4 | X | X |
| | | | | Isopod 5 | X | X |
| | | | | Isopod 6 | X | X |
| | | | | Isopod 7 | | X |
| | | | | Isopod 8 | X | X |
| | | | | Isopod 9 | X | X |

| Phylum | Class | Order | Family | Organism | Presence in Habitat | |
|---|---|---|---|---|---|---|
| | | | | | Mangrove | Rocky shore |
| | | | | Isopod 12 | X | X |
| | | | | Isopod 13 | | X |
| | | | | Isopod 14 | X | X |
| | | | | Isopod 15 | | X |
| | | | | Isopod 25 | | X |
| | | | | Unident. isopod | X | X |
| | | Tanaidacea | | | X | X |
| | Maxillipoda | Cirripedia | Tetraclitidae | *Tesseropora rosea* | | X |
| | | | | Unident. barnacle | | X |
| | | Copepoda | | | | X |
| | | Ostracoda | | | | X |
| Echinodermata | Asteroidea | | Asteriidae | | | X |
| | Ophiuroidea | | | | | X |
| Mollusca | Bivalvia | | | Unident. Bivalve | X | X |
| | | | Erycinidae | *Arthritica helmsii* | X | X |
| | | | | *Lasaea australis* | | X |
| | | | Laternulidae | *Laternula* spp. | X | X |
| | | | Mytilidae | *Xenostrobus securis* | X | X |
| | | | Neculidae | *Leionucula* spp. | | X |
| | Gastropoda | | | Unident. gastropod | | X |
| | | | Anabathronidae | *Amphithalamus* spp. | X | X |
| | | | Assiminidae | *Assiminea* spp. | X | X |
| | | | Buccinidae | *Nassarius* spp. | | X |
| | | | Cingulopsidae | *Pseudopisinna gregaria* | | X |
| | | | Eatoniellidae | *Crassitoniella flammea* | | X |
| | | | | *Eatoniella atropurpurea* | | X |
| | | | Hydrobiidae | *Ascorhis victoriae* | X | |
| | | | | *Ascorhis* spp. | X | X |
| | | | | *Tatea* spp. | X | X |
| | | | Littorinidae | *Bembicium auratum* | X | X |
| | | | Rissoellidae | *Rissoella* spp. | | X |
| | | | | Gastropod 26 | | X |
| | | | | Gastropod 66 | X | X |
| | | | | Gastropod 117 | | X |
| | | | | Gastropod 118 | | X |
| | | | | Gastropod 161 | X | X |
| | | Pulmonata | Amphibolidae | *Salinator solida* | | X |
| | | | Ellobiidae | *Ophicardelus ornatus* | X | |
| Arthropoda | Insecta | | | Adult insect | X | X |
| | | | | Insect larvae | X | X |
| | | | | Collembolid | X | X |
| | Chelicerata | Arachnida | | Spider | X | X |
| | | | | Mite | | X |
| | | | | Pycnogonida | X | |
| | | | | Acari | X | |