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# Factors affecting meiofaunal community structure in the Pina Basin, an urbanized embayment on the coast of Pernambuco, Brazil

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Intertidal meiofauna were collected from various locations around the Pina Basin, Recife, Brazil. The basin is a shallow restricted environment which receives large quantities of untreated sewage, and is therefore polluted, eutrophic and hypoxic. The meiofauna were identified to major taxa (nematodes, copepods, polychaetes, oligochaetes, turbellarians, gastrotrichs, rotifers, ostracods, acari, and juvenile amphipods and bivalves) and counted. Subsamples of nematodes were extracted and identified to the level of genus. The nematode community structure was typical of organically-enriched intertidal areas world-wide. Various environmental measurements were made, including sediment structure, salinity, dissolved oxygen and biochemical oxygen demand in the overlying water. Of these the latter two were shown to be most closely linked to variation in meiobenthic community structure. A much closer link was shown between variation in nematode assemblage structure, biochemical oxygen demand in the overlying water and gravel content of the sediment.

## INTRODUCTION

Recife is situated on the coast of the State of Pernambuco (Figure 1), an area with a warm wet tropical climate. The city, home to 1.4 million (municipal area) and 3.3 million (metropolitan area) people, has built up around the Pina Basin. This is a complex estuary, separated from the open ocean by a natural reef dyke of calcareous sandstone, into which flow the Capibaribe, Tejipió, Jiquiá, Jordão and Pina Rivers. The basin is nearly 3.6 km long and varies between 0.26 and 0.86 km in width, with a total area of 2.02 km$^2$. It has a maximum depth of 4.5 m, and a semi-diurnal tide with a maximum amplitude of 3.5 m. Salinity within the basin ranges from 0.4 to 37 psu, and water temperature between 27 and 32°C. The rivers entering the basin flow through urban areas with no, or only poor, sanitation where they pick up a range of pollutants, in particular high levels of nutrients in domestic sewage derived from Boa Viagem, Pina and the centre of Recife. As a result of the constant influx of nutrients the Pina basin is hypereutrophic and organically polluted. Despite this the area has enormous biological potential, and molluscs, crustaceans and fish are exploited by the low-income population.

Hypoxia affects large areas of marine waters all over the world, the severity, frequency and spatial scale of hypoxic events are increasing, and as a result of human population growth and global warming the problem of hypoxia is likely to become worse in the years to come (Wu, 2002). Meiofauna are useful indicators of anthropogenic perturbation in aquatic ecosystems (see review by Coull & Chandler, 1992), and under hypoxic conditions there is a general tendency for meiobenthos to increase in importance relative to macrobenthos (Wu, 2002). Josefson & Widbom (1988) hypothesized from evidence in the literature that meiofauna are less sensitive to hypoxia than macrofauna. As hypoxic periods are often symptomatic of eutrophic conditions their findings were used (Austen & Wibdom, 1991) to suggest that meiofauna may be of limited use in the monitoring of such conditions.

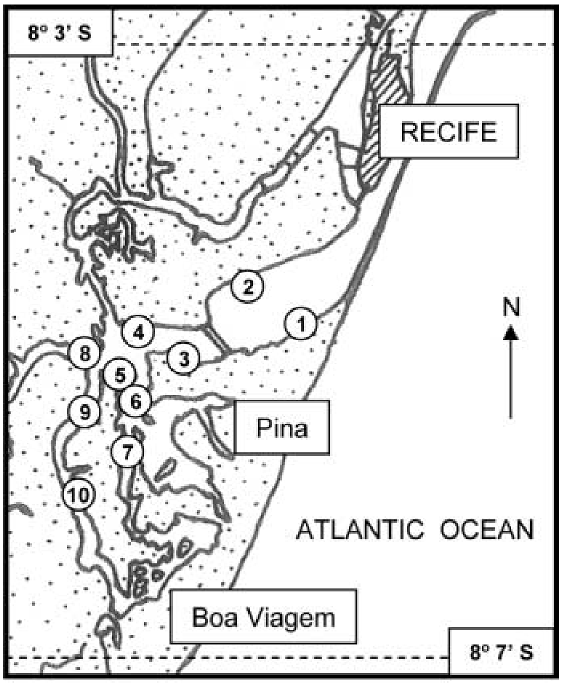

**Figure 1.** Map of the Recife area of the coast of Pernambuco, showing the sampling locations (1–10) in the Pina Basin. The opening to the sea is at the top right.

In this study we examine variation in the community structure of free-living nematodes, identified to the level of genus, and of meiofauna identified to major groups, within the Pina Basin. Relationships between meiofaunal variation, at both levels of taxonomic discrimination, and environmental variables, including variables chosen to reflect hypoxia and organic pollution, are examined and compared.

## MATERIALS AND METHODS

Samples were collected on 27 September 1999 from ten stations along the length of the Pina Basin (Figure 1). At each station three sampling points were selected at random on the mid-shore. At each sampling location a core for meiofaunal analysis was collected using a 2.7 cm internal diameter sawn-off hypodermic syringe inserted to a depth of 2.0 cm, transferred to a plastic pot and fixed with 4% formaldehyde in seawater. The amount of 100 g of sediment was collected for organic and granulometric analysis and a water sample was collected for the determination of salinity, dissolved oxygen (DO) and biochemical oxygen demand (BOD). Salinity was determined by Morh-Knudse's, and DO by Winkler's, method (Strickland & Parsons, 1965). Biochemical oxygen demand was determined according to the standard methods for the examination of waste-water (APHA, 1965). Organic matter in sediments was determined by calcination (Haynes, 1982).

For granulometric analysis sediment samples were air-dried prior to being dried in an oven at 90°C for 24 h. After drying, samples were gently disaggregated and then manually quartered with a spatula. Weighed samples were passed through a standard series of sieves (apertures of 2, 1, 0.5, 0.25, 0.125 and 0.0625 mm), and the retained fractions reweighed. The proportion of silt and clay in the samples was calculated as the difference between the total weight of the sample and the sum of the sediment fractions retained on the sieves.

Samples for meiofaunal analysis were gently washed through 0.5 and 0.045 mm meshes. The fraction retained on the smaller mesh was thinly spread on 200-square Dolffus plates and abundances of major taxa were determined using a binocular dissecting microscope. For the identification of nematodes 50 specimens were picked out of each sample at random and transferred to glycerol using a modification of the evaporation technique described by

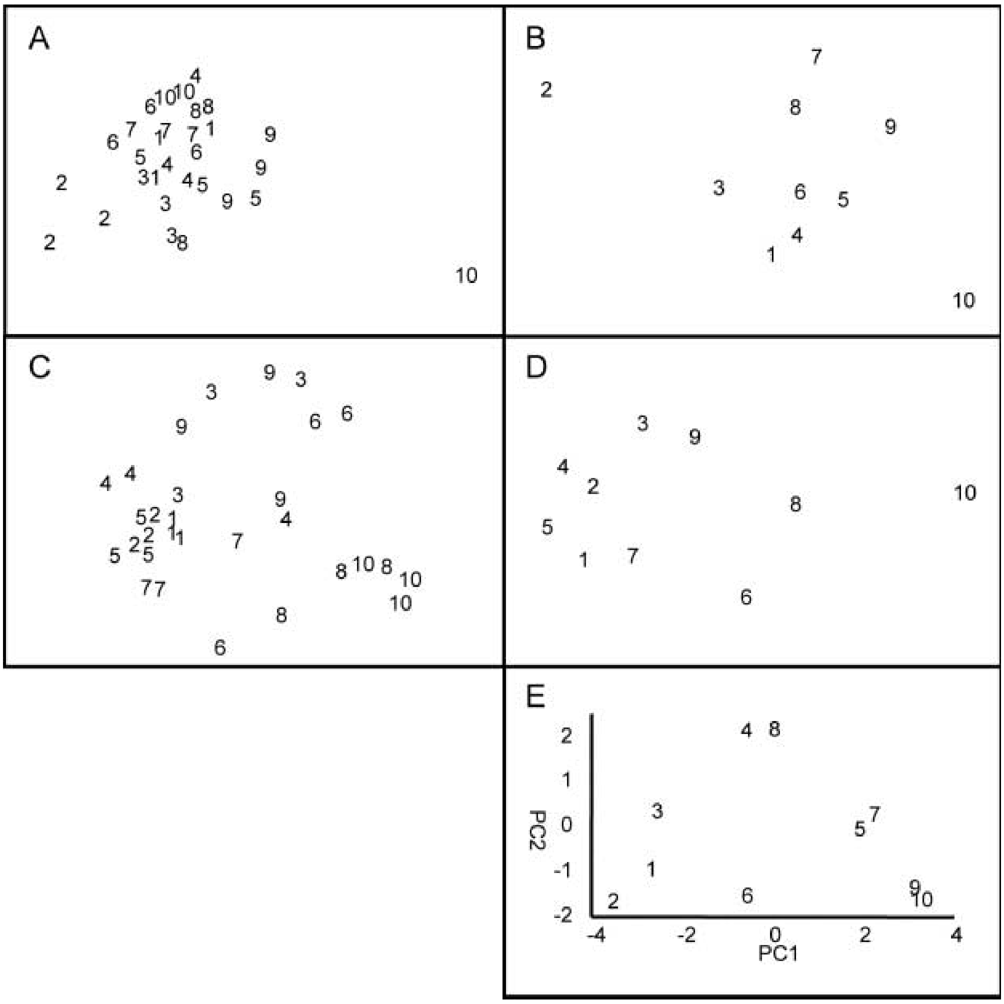

**Figure 2.** Ordination plots: (A) multidimensional scaling (MDS) ordination of samples based on Bray–Curtis similarities calculated from standardized, fourth-root transformed nematode abundances; (B) MDS ordination of sites, as (A) but calculated using the average abundances of nematodes in the three replicates from each site; (C) as (A), but calculated using abundances of major groups; (D) as (B), but calculated using abundances of major groups; (E) principal components analysis ordination of sites based on measured environmental variables.

Warwick et al. (1998). Subsequently ten specimens at a time were transferred to a drop of anhydrous glycerol placed within a pre-prepared paraffin wax ring on a microscope slide, a coverslip was added, and the slide was carefully placed on a hotplate to gently melt the wax and seal the slide. Specimens were identified to genera under a compound microscope using the pictorial keys of Warwick et al. (1998).

For the majority of statistical analyses non-parametric multivariate techniques included in PRIMER (Plymouth Routines In Multivariate Ecological Research) version 5 (see www.primer-e.com) were used.

## RESULTS

### *Nematodes*

Nematode abundances within samples ranged from 35 to 3339 individuals.10 cm$^{-2}$. A total of 38 genera were recognized. Of these 17 were rare, occurring in one or two samples only.

As a fixed number of individuals within each sample was identified the data are not suitable for a full quantitative treatment as the effective sample size varies. In particular calculating diversity indices that measure changes in species richness and evenness would be misleading. To cope with varying sample size data were standardized prior to multivariate analyses. As dominance within samples varied widely (Simpson's dominance index 1-$\lambda'$ ranged from 0.33 to 0.78) replicate nematode abundances were fourth-root transformed prior to analysis. Intersample similarities were then calculated using the Bray–Curtis similarity coefficient. One-way analysis of similarities (ANOSIM) (Global $R=0.342$, $P<0.001$) confirms that the nematode community structure differs significantly between stations. The degree of replication (three replicates per station) was not sufficient for pairwise comparisons between stations to achieve significance at $P=0.05$, although the differences between stations can be visualized in the multidimensional scaling (MDS) plot (Figure 2A) in which some evidence of a weak gradient from stations nearer to the open sea (1, 2, 3) to those deep within the basin (7, 9, 10) is apparent. In order to examine relationships between nematode community structure and environmental variables, and variation in these between stations, nematode abundances from each station were averaged. With the removal of within-station variability MDS shows a clearer pattern of variation (Figure 2B), and may be compared to an ordination of stations based on correlation-based principal components analysis (PCA, Figure 2E) of sites based on measured environmental variables (Table 1). Principal components 1 and 2 explain 83% of the variation in the data. Variables scoring high on axis 1 are mean particle size, per cent silt/clay and per cent organic matter, and variables scoring low are salinity, sorting, per cent gravel and sand, temperature, BOD and DO. Variables scoring high on axis 2 are salinity, mean particle size, per cent silt/clay, temperature, BOD and DO, and variables scoring low are sorting and per cent gravel and sand. Stations are clearly separated on the basis of their locations throughout the basin.

BIOENV was used to determine the subsets of environmental variables which provide the closest matches with the averaged nematode community structure (Table 2). No single variable was highly correlated with the variation in community structure. The best correlation (Spearman's $\rho=0.74$) was with four variables, namely sorting, per cent gravel, temperature and BOD. Variables which were included in all the ten best combinations were per cent gravel and BOD. Variables which were not among the ten best combinations were salinity and per cent sand, and per cent silt/clay was only included in one combination.

Similarity percentages analysis (SIMPER) was used to determine the genera contributing to observed differences between stations. Genera contributing to the first cumulative 40% dissimilarity between stations included the comesomatids *Sabatieria* and *Comesoma*, linhomoeids *Metalinhomoeus*, *Linhomoeus* and *Terschellingia*, xyalids *Daptonema* and *Theristus*, monhysterids *Diplolaimella* and *Diplolaimelloides*, desmodorid *Spirinia*, chromodorids *Prochromadorella* and *Innocuonema*, cyatholaimids *Paracanthonchus*, *Pomponema* and *Marylynnia* and oncholaimids *Viscosia*, *Oncholaimus* and *Prooncholaimus*.

### *Major groups*

Total meiofaunal abundances within samples varied between 112 and 3423 individuals.10 cm$^{-2}$. Major taxa identified were nematodes, copepods, polychaetes,

**Table 1.** *Values of environmental variables measured at each of the sites in the Pina Basin.*

| Site | Salinity (psu) | MPS (Phi) | Sorting | Gravel % | Sand % | Silt+clay % | OM % | Temp. °C | BOD mg l$^{-1}$ h$^{-1}$ | DO ml l$^{-1}$ |
|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 27.9 | 1.69 | 1.26 | 0.0 | 89.1 | 11.0 | 0.13 | 28.9 | 3.18 | 2.24 |
| 2 | 27.9 | 1.60 | 1.86 | 18.5 | 69.7 | 11.8 | 0.05 | 28.9 | 3.18 | 2.24 |
| 3 | 30.0 | 2.38 | 1.92 | 0.0 | 67.6 | 32.4 | 0.73 | 28.8 | 4.03 | 2.84 |
| 4 | 30.0 | 4.28 | 0.85 | 0.3 | 17.1 | 82.6 | 0.17 | 28.8 | 4.03 | 2.84 |
| 5 | 24.9 | 4.41 | 0.60 | 0.0 | 15.1 | 85.0 | 1.36 | 28.6 | 3.04 | 1.89 |
| 6 | 24.9 | 2.96 | 1.62 | 0.4 | 58.7 | 40.9 | 0.52 | 28.6 | 3.04 | 1.89 |
| 7 | 24.9 | 4.48 | 0.32 | 0.0 | 4.3 | 95.7 | 1.20 | 28.6 | 3.04 | 1.89 |
| 8 | 30.0 | 4.41 | 0.80 | 0.0 | 15.9 | 84.1 | 1.07 | 28.8 | 4.03 | 2.84 |
| 9 | 26.3 | 4.44 | 0.78 | 0.4 | 11.0 | 88.6 | 1.70 | 28.4 | 0.46 | 1.32 |
| 10 | 26.3 | 4.37 | 0.61 | 0.0 | 16.8 | 83.2 | 1.70 | 28.4 | 0.46 | 1.32 |

MPS, mean particle size; OM, organic matter; Temp., water temperature; BOD, biochemical oxygen demand; DO, dissolved oxygen.

**Table 2.** *Summary of BIOENV results. Environmental variables (✓) contributing to subsets providing the ten 'best' matches ($\rho$=Spearman's rank correlation) with standardized, fourth-root tansformed nematode and meiofaunal average abundances across sites.*

| $\rho$ | Salinity | MPS | Sorting | Gravel | Sand | Silt+clay | OM | Temp. | BOD | DO |
|---|---|---|---|---|---|---|---|---|---|---|
| Nematodes | | | | | | | | | | |
| 0.741 | | | ✓ | ✓ | | | | ✓ | ✓ | |
| 0.737 | | | ✓ | ✓ | | | | | ✓ | |
| 0.730 | | | ✓ | ✓ | | | ✓ | ✓ | ✓ | |
| 0.721 | | | | ✓ | | ✓ | | | ✓ | |
| 0.716 | | | | ✓ | | | ✓ | | ✓ | |
| 0.715 | | ✓ | ✓ | ✓ | | | | ✓ | ✓ | |
| 0.714 | | ✓ | | ✓ | | | | | ✓ | |
| 0.714 | | | ✓ | ✓ | | | ✓ | | ✓ | |
| 0.712 | | | ✓ | ✓ | | | ✓ | | ✓ | ✓ |
| 0.711 | | ✓ | | ✓ | | | | | ✓ | ✓ |
| Major taxa | | | | | | | | | | |
| 0.230 | | | | | | | | ✓ | ✓ | |
| 0.228 | | | | | | | | ✓ | ✓ | ✓ |
| 0.199 | | | | | | | ✓ | ✓ | ✓ | |
| 0.192 | | | | | | | | | ✓ | |
| 0.192 | | | | | | | | | ✓ | ✓ |
| 0.191 | | | | | | | | ✓ | | |
| 0.189 | ✓ | | | | | | | ✓ | ✓ | |
| 0.189 | ✓ | | | | | | | ✓ | ✓ | ✓ |
| 0.169 | | | | | | | ✓ | ✓ | | |
| 0.167 | ✓ | | | | | | | ✓ | | |

MPS, mean particle size; OM, organic matter; Temp., water temperature; BOD, biochemical oxygen demand; DO, dissolved oxygen.

oligochaetes, turbellarians, gastrotrichs, rotifers, ostracods, acari, and juvenile amphipods and bivalves. Nematodes were numerically dominant, followed by polychaetes. Copepods and oligochaetes occurred in most samples, but in lower numbers.

Replicate abundances of major groups were fourth-root transformed prior to a similar suite of analyses to that applied to the abundances of nematode genera. The assemblage structure was significantly different between stations (1-way ANOSIM Global R=0.54, $P<0.001$, Figure 2C), with inter-station differences being more marked (higher Global R) than for nematode genera, and with the main separation appearing to be between stations 8 and 10, and to a lesser extent 6, from those nearer to the open sea. Variation in the average community structure across stations (Figure 2D) clarifies the gradient in community structure, which was most highly correlated with two variables (temperature and BOD) although the correlation (Spearman's $\rho$=0.230) was considerably lower than for the nematode genera (Table 2). None of the variables relating to sediment structure (mean particle size, sorting, percentages of different sized particles) contributed to any of the ten best combinations; BOD, temperature or both contributed to all of the combinations.

Taxa contributing to the first 50% dissimilarity between stations included oligochaetes, gastrotrichs, polychaetes, copepods, turbellaria, and nematodes. Despite their numerical dominance and the fact they contribute 34 to 100% of within-station similarities, differences in nematode abundances were relatively less important in discriminating between stations.

## DISCUSSION

The densities of total meiofauna and individual meiofaunal groups reported here are broadly in line with those reported in intertidal marine sediments from a variety of locations where the fauna has been collected on sieves with comparable mesh sizes to those used in the present study. Thirty-eight genera of nematodes were recorded from intertidal sediments in the Pina Basin. This is many fewer than the number of nematode genera found in detritus-rich intertidal mangroves in Malaysia (89 genera: Somerfield et al., 1998), or in sediments subject to seasonal hypoxia in the Venice lagoon (63 genera: Villano & Warwick, 1995), but is a higher diversity than the 23 species of nematodes found in muddy sediments in a sheltered bay on the Swedish west coast influenced by seasonal hypoxia in the bottom water (Hendelberg & Jensen, 1993). Somerfield et al. (1998) found approximately 40% of the meiofauna identified to species (nematodes and copepods) in their study to be rare and less than 25% of species to be common or abundant, figures very much in line with the findings of this study. In addition, the genera found in the Pina Basin are found in intertidal sediments world-wide. In light of these facts there is no evidence that the meiofaunal communities in the Pina Basin are very different to those already studied in other parts of the world, and therefore they should be expected to respond to changes in environmental conditions in similar ways.

Some nematode assemblages are only found in anoxic and hypoxic conditions, and the families and species typical of such assemblages are highly adapted and specific to that environment, but these were not present in the assemblages found in this study. The genera contributing to differences between stations include, however, genera known to respond to factors related to eutrophication and low oxygen levels. For example, some *Sabatieria* species are tolerant of long periods of anoxia, and Oncholaimids thrive in disturbed and heavily enriched areas. Decaying marsh vegetation and mangrove leaves are often dominated by *Diplolaimella* and *Diplolaimelloides*. These two genera are also associated with brackish-water environments.

In this study, although the assemblages of both major taxa, and nematodes identified to the level of genus, provide evidence that factors relating to organic enrichment, hypoxia and sewage pollution are important determinants of community structure, the nematodes provide a much closer match with variations in environmental conditions. Although this study may be added to those others which are seen as supporting the idea that environmental changes may be adequately monitored by examining changes in meiofaunal communities at the level of major taxa, this would be an oversimplification. In field comparisons of organically polluted and unpolluted sites responses at the level of higher taxa are highly variable whereas in field studies where meiobenthos has been identified to lower taxonomic levels, such as genera or species, the response is more predictable (Coull & Chandler, 1992). The nematode communities found in this study are similar to those found elsewhere, may easily be identified to genus using modern pictorial keys, and the ecology of many genera is reasonably well known. With these facts in mind, we recommend that programmes which set out to monitor the effects of reduced oxygen conditions on meiobenthic communities attempt to identify the fauna to lower taxonomic levels.

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