

# Marine: soft sediment sampling for infaunal communities



Version 1.0

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### Disclaimer

This document contains supporting material for the Inventory and Monitoring Toolbox, which contains DOC's biodiversity inventory and monitoring standards. It is being made available to external groups and organisations to demonstrate current departmental best practice. DOC has used its best endeavours to ensure the accuracy of the information at the date of publication. As these standards have been prepared for the use of DOC staff, other users may require authorisation or caveats may apply. Any use by members of the public is at their own risk and DOC disclaims any liability that may arise from its use. For further information, please email [biodiversitymonitoring@doc.govt.nz](mailto:biodiversitymonitoring@doc.govt.nz)

## Synopsis

Below is a brief summary of the rationale and possible monitoring objectives associated with this methodology, but for full details of how to perform sediment core surveys please refer to '[Full details of technique and best practice](#)'.

Soft sediment cores are used to measure population (e.g. abundance) and/or community (e.g. species richness or diversity) metrics of soft sediment infaunal assemblages within estuarine sand/mudflats. They can give a good indication of the health and ecological condition of sedimentary environments (e.g. whether they are polluted by toxins and/or impacted by increased sedimentation) (Robertson et al. 2002), which can be used to aid future management and conservation decisions. In particular, certain species that occupy sand/mudflat areas have specific tolerances to toxins (i.e. heavy metal contaminants) or the nutrient and % fine mud content of the sediment and the water column (Borja et al. 2000; Gibbs & Hewitt 2004; Borja & Muxika 2005). For example, pipi (*Paphies australis*) are intolerant of high sediment mud content, and therefore tend to only be found in estuaries that are relatively sandy (see [Case study B](#) and [Case study C](#)). Conversely, the amphipod *Paracorophium excavatum* is tolerant of a wide range of conditions and is a primary coloniser of estuarine habitats; therefore, it is a good indicator of disturbance (Ford et al. 1999; Robertson & Stevens 2012).

The most common method for surveying soft sediment infaunal communities is to take sediment cores and to sieve out the individuals contained within them. Sampling usually consists of three components: (1) locating and establishing monitoring sites and core extraction; (2) core sieving to separate out the individuals from the sediment; and (3) identifying and counting individuals within the core. Given a sufficient number of core samples, this method can provide a good estimate of the biotic communities living within the sediments, as most of the species are relatively sedentary and are therefore likely to be observed. Commonly, species of biological and social importance, such as cockles, are also measured and their size distribution determined. This can give information about the status of their stocks as well as their population demographics.

The arrangement of sites, number of cores per site, frequency of sampling, sieve mesh size and nature of sample processing will be determined by the objectives of the monitoring study. Typically, sampling is repeated annually and can be used in long-term monitoring. Common objectives include monitoring the status of estuarine environments in response to changing catchment usage, the effects of effluent discharge into estuarine environments, and changing water flow regimes.

A robust sampling design should complement sediment core surveys with the collection of data describing the physical environment (e.g. organic and nutrient content, heavy metal concentration, grain size composition of the sediment) so that changes to the biological community can be attributed to a specific effect, which will aid future management and conservation decisions. In addition, taxonomic identification and counts of organisms should be quality controlled to ensure that it is consistent with previous surveys (see '[Quality control](#)').

## Assumptions

- The taxa of interest can be detected and identified with sufficient accuracy for the research or survey objectives.
- Observer effort and skills are similar across sites, locations and/or sampling occasions.
- Sites are representative of the wider estuarine environment or of the gradient or effect that is to be determined (if not accompanied by physical information gathered from the same locations and times).
- Sites and cores are statistically independent.

## Advantages

- Reliable indicators of sedimentary environment.
- Can be used in long-term monitoring.
- Requires only basic equipment.
- If processing is done in the field, sampling is non-destructive (i.e. individuals are returned to the site of collection alive).
- Similar methods are currently employed in a wide variety of estuaries nationwide, so there is a large body of pre-existing data that can be used in cross-estuary comparisons and to quantify the level of natural variability in communities through time.
- Methods are recognised and used internationally so that comparisons with sites outside New Zealand are also possible.
- Sampling is easily repeatable over time.
- Size distributions and demographics of large conspicuous species (e.g. cockles) can also be determined using this method.
- The method is amenable to the collection of covariate data regarding the physical environment.
- The method is relatively inexpensive to employ as no specialist equipment is required, and costs are likely to be predominantly determined by the number of hours required to process samples and to carry out the fieldwork.

## Disadvantages

- Time-consuming to process and record all information.
- Species identification may require expert knowledge.
- Sampling times are restricted by tidal cycle.
- Depending on sieve size, certain species will be lost/not-accounted for, so there is a trade-off between time and quality of data collected.

- Frequent monitoring may negatively affect the state of monitoring sites, particularly if samples are not processed *in situ*.
- Frequent monitoring could also introduce a degree of bias into the monitoring data collected, giving a false impression of the changing state of the estuary.

## Suitability for inventory

- Inventories of individual sites with homogeneous habitat may be possible—previous studies have concluded that very few if any new species are found when sampling more than 30 cores within relatively homogeneous sites (Thrush et al. 1988; Pridmore et al. 1990).
- For sites with high habitat heterogeneity, species inventories are difficult because taxa are likely to differ between microenvironments associated with tidal elevations and toxin, nutrient, freshwater and saltwater influx. In this instance, sediment cores are not recommended as a method for inventory.

## Suitability for monitoring

- This method is suited for the monitoring of estuarine condition given known species sensitivities to toxins, nutrients and muddiness (Thrush et al. 2003; Anderson et al. 2007).
- Changes in species composition can be strongly associated with physical changes to the estuarine environment, making this method suitable for identifying remedial management actions.

## Skills

Soft sediment sampling requires a relatively high level of expertise.

Pre-survey:

- Survey design skills for determining the number of replicates, stratification (if any) and placement of replicates, and what variables are to be recorded
- GIS knowledge for the planning of field locations and sites
- Transfer of site coordinates to portable GPS

In the field:

- Ability to identify, count, size and (if applicable) sex individuals and record any other variables of interest along transects
- The skills to record and securely manage data
- Use of portable GPS
- Good fitness level

Data analysis:

- Familiarity with basic statistics
- Familiarity with statistical package (*R* recommended)
- Appropriate storage of data

## Resources

Survey work requires a minimum of two people, one taking samples and potentially identifying and counting species and the other as note taker and sample labeller/depositor. If more than three, it is advisable to split work forces into teams of two people to achieve maximum efficiency.

Field equipment includes:

- Waders, or wetsuit booties as footwear (if users don't have waders then it may be worth bringing something for surveyors to kneel on)
- Sunscreen, hat, insect repellent and plenty of snacks and water
- Wet weather gear and warm items of clothing as weather can change quickly
- Sled for dragging gear
- Large box and tie-downs for placement of gear on sled
- Shovel
- Corer (hollow piece of plastic tubing, most commonly 13 cm in diameter by 15 cm long in intertidal applications; however, see '[Full details of technique and best practice](#)' as this may vary depending on the research question)
- Sharpies or permanent markers for sample labelling
- Sieve (0.5 mm or 1 mm; however, see '[Full details of technique and best practice](#)' as this may vary depending on the research question)
- Sample jars containing 70% isopropyl alcohol for storage and preservation of sieved core contents
- Bucket (for gathering water to perform sieving)
- Waterproof notepad, pre-prepared data sheets and pencil for note-taking and sample recording
- Calipers for recording the sizes of indicator species (e.g. shell width of cockles)
- At least 4 bamboo stakes for marking site corners (recommended additional stakes for marking placement of blocks within sites, and also locations of core samples)
- 2 measuring tapes (at least 100 m long for measuring out site extent)
- GPS unit for site location
- Additional ziplock bags for samples of unknown species for lab identification
- ID guides to aid in species identification
- Waterproof camera

## Minimum attributes

Consistent recording and measurement of the following attributes is critical for the implementation of the method. Depending on the research question(s), other attributes may be required.

DOC staff must complete a 'Standard inventory and monitoring project plan' (doccm-146272).<sup>1</sup>

### Survey meta data

- Observer and recorder
- Date and time
- Estuary and site name and coordinates
- Coordinates of each core and the block it came from within the site
- Photograph of the general appearance of the site

### Core data

- Location of transect within the site (e.g. depth)
- Transect (replicate) number
- Depth at start and end of transect
- Time at start and end of transect
- Data for the variables of interest to the survey objectives (e.g. species counts, size measurements)
  - Individuals should be identified to species level and counted.
  - When species cannot be identified they should be pooled into family or genus level groups and counted. Alternatively, unidentified species can be counted and classified into operational taxonomic unit (OTU; e.g. *Abra* sp1, *Abra* sp2, etc.) with a few labelled specimens set aside for later identification by a specialist.

### Optional attributes

- For the area surrounding each core, the presence and % cover of surface vegetation and macroalgae as well as presence and abundance of surface dwelling animals (e.g. snails) or signs, such as crab burrow holes.
- Photographs of the outer appearance of each core (prior to sieving) so that additional attributes related to sediment condition can be qualitatively determined.
- Measurement of redox potential depth (RPD) based on colour of sediment.
- Samples of sediment for: (1) grain size analysis; (2) nutrient analysis; (3) heavy metal contaminant analysis; and (4) determination of sediment organic content. It is recommended to take these samples adjacent to each core, although in homogenous sites, fewer

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<sup>1</sup> <http://www.doc.govt.nz/Documents/science-and-technical/inventory-monitoring/im-toolbox-standard-inventory-and-monitoring-project-plan.doc>

representative samples might be sufficient. Samples should be labelled indicating date collected, site name and block within the site.

- Samples of shellfish (e.g. cockles) for tests of heavy metal contaminants and bacterial infection.
- Sizes of cockles, pipi and other large and/or conspicuous species, such as crabs.
- Physical analyses of sediment composition may include determination of:
  - % mud, % sand and % gravel (i.e. grain size analysis).
  - Nitrates, nitrites, total nitrogen, total phosphorous, organic carbon and chlorophyll-a content of sediment.
  - Concentrations of heavy metals including cadmium, chromium, copper, nickel, lead, zinc and mercury.
- Concentrations of heavy metals and indicators of bacterial infection (related to effluent discharge) within shellfish tissue.

## Data storage

DOC is currently developing a national database to hold and provide access to data collected from marine reserve monitoring in New Zealand. The aims of the database are to:

- Support consistent standards in national marine reserve monitoring programmes for marine environmental quality
- Coordinate and optimise marine reserve monitoring in New Zealand
- Provide a high quality monitoring dataset for New Zealand's marine reserves

Once operational, this methodology will be updated with a description of how to lodge data within the national database. In the interim, data should be recorded within the spreadsheets associated with this methodology. It is essential that all raw data sheets are completed, digitised and backed up on external hard drives. Raw data and associated metadata should be entered into databases/spreadsheets in a standardised format. This should include metadata stored in a separate sheet, and a sheet containing sampling data collected during the monitoring programme stored in one 'brick' of data that can be continually updated as more surveys in that monitoring programme are carried out.

The first spreadsheet/database should include metadata associated with the survey (as shown in Figure 1). Where relevant, this should include site, species and sampling unit metadata.

Additionally, the metadata should also include a description of the monitoring objectives and any information that will allow someone unfamiliar with the monitoring to interpret the data and reproduce the monitoring methodology.

Data should be entered into subsequent spreadsheets/databases in a logical format (as shown in Figure 1 for future reference). Data should be arranged such that each row represents an individual core with the corresponding data regarding time, location and species abundances arranged into columns. Ideally, all data should be located within a single database to facilitate ease of access..

For internal DOC monitoring, information pertaining to each survey within a marine reserve and resultant data/reports should be entered into the Marine Protected Area Monitoring and Research (MPAMAR) datasheet ('MPAMAR metadata: national'—doccm-1163829) so there is an easily accessible account of the survey.

	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T	U	V	W	X	Y	Z	AA	
1							Group >	Nematoda	Polychaeta		Oligochaeta	Gastropoda		Bivalvia	Crustacea													
2	Date	Estuary	Site	Lat-NZMG	Lon-NZMG	Block	Species >	Nematoda	<i>Aglaophamus macroura</i>	<i>Hermonastus filiformis</i>	<i>Perinereis valvata</i>	<i>Scolecopides bennhami</i>	<i>Terebellidae</i> sp. 1	Oligochaeta	<i>Cominella glandiformis</i>	<i>Diloma subrostrata</i>	<i>Haminoea zelandiae</i>	<i>Notoacmea helmsi</i>	<i>Zedunculus lutulentus</i>	<i>Austrovenus stutchburyi</i>	<i>Macromona liliana</i>	<i>Nucula hartvigiana</i>	<i>Amphipoda</i> sp. 1	<i>Helice crassa</i>	Total Species	Total Individuals	Notes	
3	21/01/2012	Porirua	Por-A	6009488	2666477	1		0	0	0	5	3	0	0	0	7	5	4	0	3	0	2	0	0	7	29		
4	21/01/2012	Porirua	Por-A	6009500	2666482	2		0	0	0	0	0	0	6	0	0	2	0	0	0	0	0	0	0	0	2	8	
5	21/01/2012	Porirua	Por-A	6009518	2666481	3		0	6	0	5	8	4	0	4	4	0	0	0	0	0	0	0	0	6	7	8	44
6	21/01/2012	Porirua	Por-A	6009534	2666492	4		3	0	0	0	0	0	3	0	5	0	0	0	0	0	3	4	6	6	24		
7	21/01/2012	Porirua	Por-A	6009533	2666500	5		0	5	4	4	4	4	3	0	0	0	9	5	0	0	0	0	0	0	7	34	
8	21/01/2012	Porirua	Por-A	6009518	2666497	6		0	3	0	0	0	3	0	2	0	0	5	1	0	6	0	1	0	7	21		
9	21/01/2012	Porirua	Por-A	6009505	2666497	7		6	0	0	0	5	0	0	0	0	0	5	0	0	5	4	0	0	0	5	25	
10	21/01/2012	Porirua	Por-A	6009484	2666489	8		0	0	0	0	0	7	0	0	0	2	5	6	0	4	0	0	0	0	5	24	Surface algal mat
11	21/01/2012	Porirua	Por-A	6009488	2666498	9		0	0	0	0	0	0	7	5	2	6	0	0	0	0	0	0	0	6	5	26	
12	21/01/2012	Porirua	Por-A	6009525	2666514	10		0	3	5	0	0	0	9	8	0	0	0	0	0	0	5	0	0	0	5	30	
13	21/01/2012	Porirua	Por-B	6009488	2666477	1		12	0	0	0	13	9	17	0	0	0	0	0	0	16	0	5	0	6	72		
14	21/01/2012	Porirua	Por-B	6009500	2666482	2		0	0	12	4	8	13	10	14	0	0	0	12	0	12	12	0	0	9	97		
15	21/01/2012	Porirua	Por-B	6009518	2666481	3		0	13	11	5	13	0	0	0	0	0	10	0	0	0	0	0	0	7	6	59	
16	21/01/2012	Porirua	Por-B	6009534	2666492	4		0	14	0	0	0	8	8	10	0	14	13	6	11	6	0	0	9	10	99		
17	21/01/2012	Porirua	Por-B	6009533	2666500	5		8	0	0	10	0	0	9	11	6	0	9	12	0	0	11	9	0	9	85		
18	21/01/2012	Porirua	Por-B	6009518	2666497	6		5	17	0	9	0	0	9	0	0	10	0	0	8	0	0	0	6	7	64		
19	21/01/2012	Porirua	Por-B	6009505	2666497	7		9	11	0	0	0	0	9	6	0	10	8	7	0	0	12	10	7	10	89		
20	21/01/2012	Porirua	Por-B	6009484	2666489	8		0	11	0	0	4	10	12	0	7	0	0	13	0	16	15	3	13	10	104	Lots of crab holes	
21	21/01/2012	Porirua	Por-B	6009488	2666498	9		0	14	10	9	10	9	0	0	0	10	0	0	6	4	8	0	13	10	93	Lots of crab holes	
22	21/01/2012	Porirua	Por-B	6009525	2666514	10		0	11	0	0	0	0	16	7	0	12	0	0	9	11	11	10	0	8	87		
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Figure 1. Example layout and format of a datasheet resulting from sediment core surveys. Separate data classes are arranged into columns and separate samples (i.e. cores) are arranged in rows.

## Analysis, interpretation and reporting

Seek advice from a statistician or suitably experienced person prior to undertaking any analysis. Ideally, statistical advice should be sought prior to any data collection to ensure that the design of the data collection is robust and suitable for answering the question at hand. For quality control the data should be checked for unlikely abundances of organisms, and errors in data entry.

### Data analyses

The type of analysis most applicable to the data will largely be determined by the research question, and whether additional supporting information (such as physical condition of sediment) has been collected or is available. However, the analysis types detailed below will likely be applicable in many cases.

## Populations—univariate analyses

A first step would be to calculate the mean and standard error (SE) (among cores within sites) of individual species densities as an indication of the variation among sites. These can then be graphed to visualise these differences. If data are to be compared among sediment cores of different sizes (i.e. among these data and data collected by regional councils that have adopted an alternative core size), then all data should be converted to densities by dividing the observed abundances by the volume ( $\text{Volume} = \pi r^2 d$ , where  $r$  is the radius, or half the core diameter, and  $d$  is the core depth) prior to any further analysis.

For rigorous statistical comparisons within estuaries (i.e. differences among sites) univariate tests (tests on individual species abundances) could include:

1. One-way analysis of variance (ANOVA) (assumptions of normally distributed errors and homogeneity of variance will need to be tested for, and, if necessary, transformations may need to be applied) (Underwood 1997)
2. Simple factorial generalised linear models (GLMs) (McCullagh & Nelder 1989) (GLMs can account for error distributions other than Gaussian and so are more flexible than ANOVAs)
3. Calculation of 95% confidence intervals for each site (based on SE, or more robustly using bootstrap routines to determine confidence intervals) (Cummings 2012) and comparing these limits among sites.

These methods could also be applied to individual sites through time.

For comparisons across estuaries (assuming similar data collection, or standardising data to account for differences in sampling routine) a mixed effects ANOVA with fixed effect of estuary and nested random effects for sites within estuary would be appropriate (Underwood 1997). Again, the assumptions of ANOVA will need to be tested for and data transformed accordingly.

If accompanying data are available then correlation or regression analyses, including linear models (LM), generalised linear models (GLM) and generalised additive models (GAM), can be used to identify the relationship between species abundances/richness/diversity, and physical attributes of the environment such as heavy metal concentrations, % mud content or nutrient content of the sediment.

## Community—multivariate analyses

The simplest community analyses involve the calculation of indices that combine information from all of the species observed. Data can be summarised into species richness and biodiversity (e.g. Shannon–Wiener index, Simpson's index) metrics, which can be plotted and compared among sites. Exploration of other metrics of biotic community compositions can also be calculated, such as the AZTI (AZTI-Tecnalia Marine Research Division, Spain) Marine Biotic Index (AMBI) (however, there are some limitations to its application—Robertson & Stevens 2010, 2012).

Rigorous statistical analyses can be applied to the entire species dataset (rather than performing analyses on each species/species group) using multivariate approaches, but these often require more skill and care in model fitting. These analyses can be more sensitive to changes in entire community composition and have been used extensively on soft sediment data (Clarke & Ainsworth 1993; Anderson et al. 2007). Examples include permutational multivariate analysis of variance (PERMANOVA) (Anderson 2001), distance based linear modelling (Legendre & Anderson 1999; Anderson et al. 2008) and principal components analyses (PCA) (Clarke & Warwick 2001; Clarke & Gorley 2006). Several excellent manuals/tutorials are available for multivariate analyses (Clarke & Warwick 2001; Clarke & Gorley 2006; Anderson et al. 2008).

Visualisation of multivariate (whole community) patterns can be achieved using a variety of methods. Examples include principal components analysis (PCA) and non-metric multidimensional scaling (nMDS) (Clarke & Warwick 2001; Clarke & Gorley 2006).

Again, a statistician should be consulted if these methods are to be used, as they can be sensitive to particular aspects of the data, and the transformations that are applied to it.

## Interpretation

Interpretation of results should be performed with the assistance of a statistician as well as consideration of the major driving forces operating within the system. At this stage it should be determined whether the goals of the original data collection have been achieved and whether the data are sufficient to answer those questions outlined prior to the initial surveys.

## Reporting

Reporting will largely be governed by the duration of the monitoring and data collection. If data collection is ongoing, regular reports should be submitted at 3- to 5-year intervals, whereas for short term (< 2 years in duration) data collection, reports should be submitted within a year of the final data collection.

## Case study A

### **Case study A: Regional Estuary Monitoring Programme—Southern Firth of Thames and Raglan Harbour, Environment Waikato (Singleton 2010)**

#### Synopsis

The Regional Estuary Monitoring Programme was initiated by Environment Waikato in 2001 to monitor changes to the biological and physical attributes of the estuarine environment within the Firth of Thames and Raglan (Whāingaroa) harbours. This monitoring programme provides a good illustration of the use of sediment core sampling and complementary surveys of physical sediment characteristics to monitor infaunal communities.

## Objectives

- To capture temporal changes in intertidal sediment characteristics and benthic macrofaunal communities. This information can then be used to distinguish changes to the estuaries as a consequence of catchment modification and human activities within the estuary from those changes that would be expected as a result of natural variability.

## Sampling design and methods

- Five 100 × 100 m sites were established in the mid-tidal elevation in each of the Firth of Thames and Raglan Harbour in 2001.
- In each estuary, two sites were sampled at 3-monthly intervals while the remaining three sites were sampled every 6 months. Following 2009, all sites were sampled on a 6-monthly basis.
- Each site was split into 12 equally sized blocks (33 × 25 m) and a core measuring 13 cm in diameter and driven to 15 cm deep was extracted from a random position within each block.
- To reduce spatial autocorrelation and interdependence between samples, sediment cores were not taken from positions within a radius of 5 m of each other. Furthermore, to avoid sampling locations that had been affected by previous surveys, samples were not taken from locations within 5 m of a previous sampling position.
- Cores were sieved using a 0.5 mm mesh size, and all remaining items in the sieve were preserved in 70% isopropyl alcohol.
- Macrofauna were sorted and identified in the laboratory. The abundance of 26 indicator species (2 amphipod, 6 bivalve, 1 cumacean, 2 gastropod, 1 cnidarian and 14 polychaete species) was determined. Bivalves were measured and assigned into large and small size classes (except pipi and wedge shells that were assigned into small, medium and large size classes). Other individuals were identified to the lowest taxonomic level and counted.
- Samples of surficial sediment were also obtained from every other block within a site, combined, and homogenised for analyses of sediment characteristics. These included grain size composition, organic carbon and nitrogen content and chlorophyll-a content (for a detailed description of the preparation and analysis techniques see Turner 2001 and Singleton 2010).

## Results

- Spatial differences between the two estuaries were exhibited as a higher abundance of polychaetes and crustaceans in the Firth of Thames (Figure 2), whereas bivalves and gastropods were more abundant in Raglan Harbour (Figure 3).
- Over the period 2008–2009 the infaunal communities were relatively stable at all of the sampled sites.
- Sediment characteristics were also relatively similar over the same time.

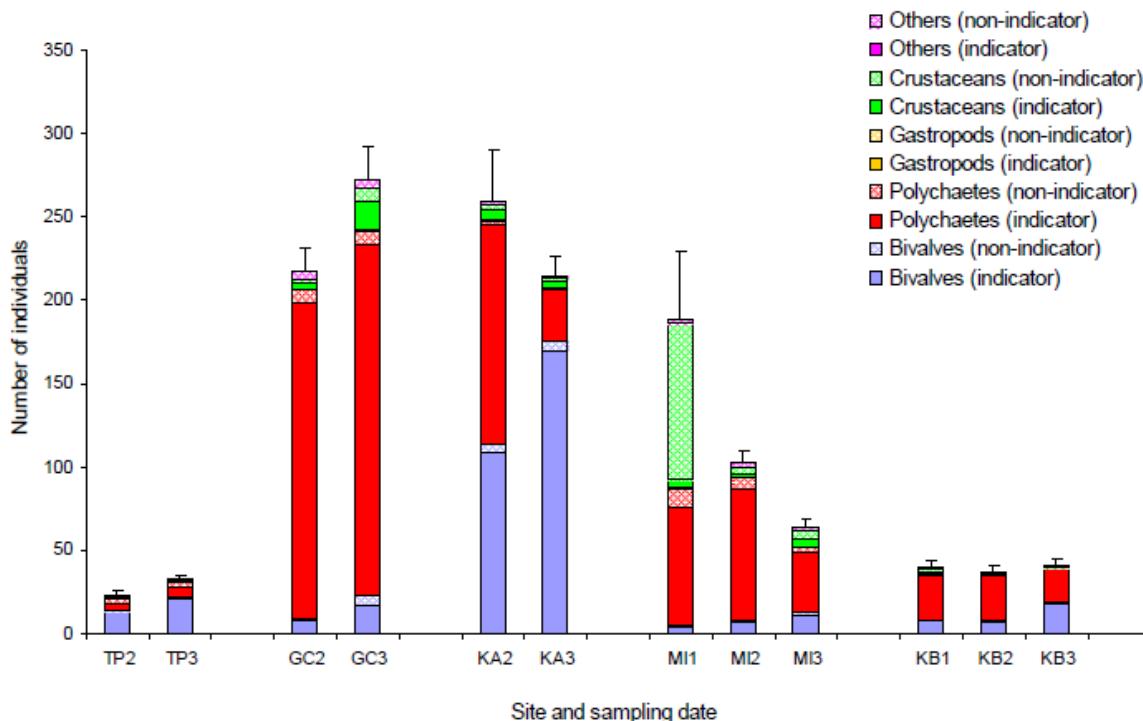


Figure 2. Mean ( $\pm 1$  SE) number of individuals and taxonomic composition of communities in the southern Firth of Thames between July 2008 and April 2009. X-axis label represents sites (TP = Te Puru, GC = Gun Club, KA = Kaiaua, MI = Miranda, KB = Kuranui Bay) and sampling dates (1 = July 2008, 2 = October 2008, 3 = Apr 2009). Taken from Singleton (2010).

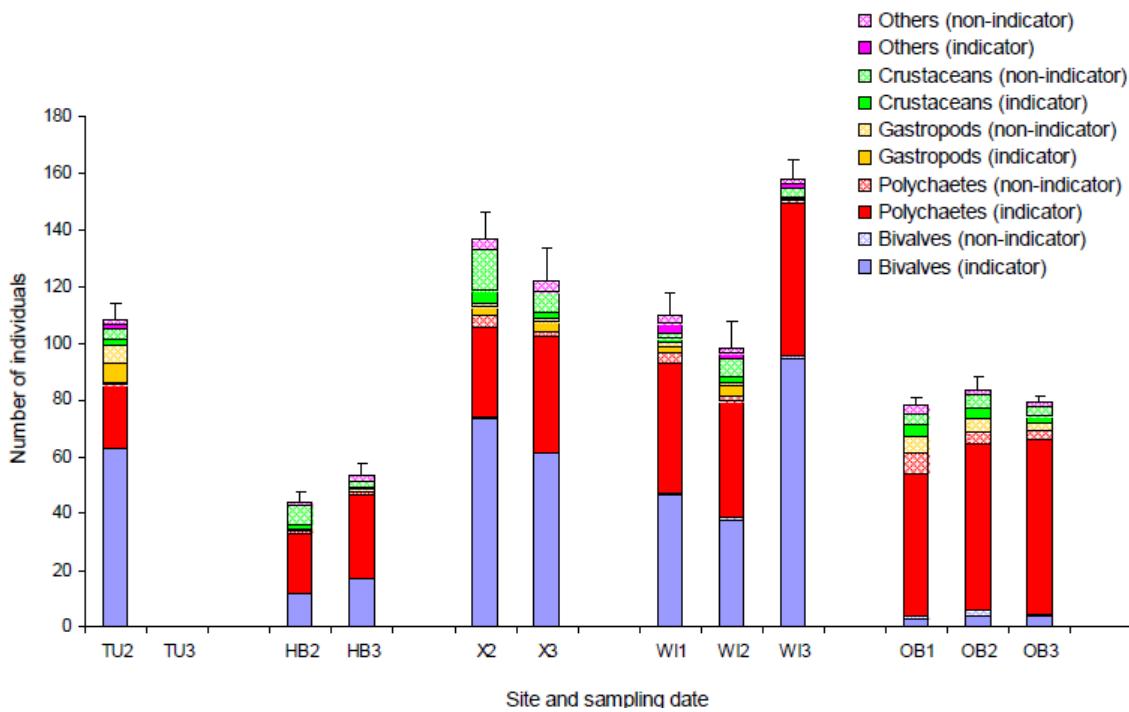


Figure 3. Mean ( $\pm 1$  SE) number of individuals and taxonomic composition of communities in Whāingaroa Harbour between July 2008 and April 2009. X-axis label represents sites (TU = Te Puna Point, HB = Hāroto Bay, X = Ponganui Creek, WI = Whatitirinui Island, OB = Ōkete Bay) and sampling dates (1 = July 2008, 2 = October 2008, 3 = Apr 2009). Taken from Singleton (2010).

## Limitations and points to consider

- Sampling was extensive in each estuary and the sampling design was robust accounting for possible spatial autocorrelation prior to sampling.
- With frequent sampling there is a greater chance that previous surveys can influence survey results through re-sampling locations that are recovering from removal of individuals in the previous survey. However, adequate controls are in place (not sampling within 5 m of these locations) to guard against this.
- As this is a general monitoring programme aiming to identify levels of natural variability, the sites were assigned to random locations within each estuary. Given a more focused research question, sites may be placed differently to identify changes associated with external pressures (e.g. close to sewer or freshwater influx).
- The collection of physical information, in addition to the biological data, provides more evidence to support the view that there were few changes to the estuarine environment between 2008 and 2009, and any observed changes would be within the limits of natural variability.
- Data quality is likely to be of a high standard as several quality control measures were in place (see '[Full details of technique and best practice](#)' for a description of control measures).

## References for case study A

- Singleton, N. 2010: Regional Estuary Monitoring Programme (REMP) data report: benthic macrofauna communities and sediments—July 2008 to April 2009. Southern Firth of Thames and Whaingaroa (Raglan) Harbour. *Environment Waikato Internal Series 2010/37*. Hamilton, Waikato Regional Council (Environment Waikato).
- Turner, S. 2001: Monitoring the region's estuaries: intertidal sand-flat benthic communities. *Environment Waikato Internal Series 2000/11*. Hamilton, Waikato Regional Council (Environment Waikato).
- Turner, S.; Carter, N. 2004: Regional Estuary Monitoring Programme: benthic macrofauna communities—April 2001 to April 2002. Southern Firth of Thames and Whaingaroa (Raglan) Harbour. *Environment Waikato Internal Series 2004/08*. Hamilton, Waikato Regional Council (Environment Waikato).
- Turner, S.; Gibberd, B.; Crozier, J. 2002: Regional Estuary Monitoring Programme—pilot study. *Environment Waikato Internal Series 2002/02*. Hamilton, Waikato Regional Council (Environment Waikato).

## Case study B

### Case study B: fine scale monitoring of highly eutrophic arms in the New River Estuary, Southland, 2011/2012 (Robertson & Stevens 2012)

#### Synopsis

Surveys in the New River Estuary were carried out between 2001 and 2005 and again in 2010 as part of a long-term monitoring programme. This estuary is bordered by a mix of vegetation and land uses, with the effects of dairy farming being a particular concern. The estuary has a wide range of habitats, but has lost areas through drainage and reclamation. Additionally, treated wastewater is discharged into the estuary causing blooms of macroalgae and elevated bacterial concentrations. Further, saltmarsh areas have been seriously degraded, which makes the area more susceptible to eutrophication and sedimentation.

This case study illustrates the use of sediment core monitoring and how it can be used to identify areas with degraded communities by quantifying the abundance of species with different mud/organic enrichment tolerances. It also illustrates how this method can be used to answer specific questions about an estuary as further monitoring sites were surveyed in 2011/2012 in response to the rapid degradation of sites within low-flow areas of the estuary.

#### Objectives

- To establish a baseline representation of the biological community and the conditions within the estuary, as well as an indication of the levels of natural variability one might expect through time. Surveys in 2010 were a follow-up to see how the state of the estuary had progressed in the intervening years. 2011/2012 surveys aimed to quantify the state of heavily degraded eutrophic sites, which were identified in 2010 and added to the monitoring programme, and to examine whether these sites supported similar communities to those previously monitored.

#### Sampling design and methods

- Three 60 × 30 m sites were established at a mid-low tidal elevation in New River Estuary in 2001, and a further two sites were added to the monitoring programme in 2011 in locations that were severely degraded.
- Between 2001 and 2005, sites were sampled on an annual basis, with follow-up surveys at 5-yearly intervals. Highly degraded sites have been sampled annually since 2011/2012.
- Each site was split into 12 equally sized blocks (15 × 10 m) and a core measuring 13 cm in diameter and driven to 15 cm deep was extracted from a random position within 10 randomly chosen blocks. Additional cores were also extracted to examine the redox potential depth (related to oxygenation and anoxic conditions) and the appearance of the sediment.
- Cores were sieved using a 0.5 mm mesh size, and all remaining items in the sieve were preserved in 70% isopropyl alcohol.

- Macrofauna were sorted and identified in the laboratory.
- The Mud Tolerance Biotic Coefficient (MTBC) was used to combine information from the entire biological community into easily interpretable values that give an indication of whether the community displays an affinity towards species that prefer muddy conditions (for further information see Gibbs & Hewitt 2004).
- Samples of surficial sediment were collected for analyses of sediment characteristics. These included grain size composition, total organic carbon, total nitrogen, total phosphorous and trace metal concentrations (cadmium, chromium, copper, nickel, lead and zinc) (for a detailed description of the preparation and analysis techniques see Robertson & Stevens 2010, 2012).
- Sedimentation rates were also measured by burying plates within the sediment and measuring the depth of sediment at successive times.
- Surface-dwelling organisms were also recorded using surface quadrats.

## Results

- Baseline surveys (2001–2005) indicated that the physical condition of the estuary was characterised by high sand content and low to moderate organic enrichment with biological communities that reflected this (Figure 4).
- Sedimentation rates between 2005 and 2010 were very high, and sites have become muddier and less oxygenated. Species that are intolerant of mud and display a strong sand preference were reduced in abundance, or in the case of pipi (*Paphies australis*), completely absent in the 2010 surveys.
- This has been accompanied by an increase in the abundance of some mud-tolerant species (Figure 4).
- Although there have been changes to the abundance of sand/mud-tolerant species, the MTBC is very low to low (higher indicating a switch to a community dominated by mud-tolerant species and a decline in ecosystem function) for these sites due to their high sand content (Figure 5).
- Conversely, the eutrophic sites were dominated by deep anoxic mud with very high sedimentation rates.
- Invertebrate communities at these sites were dominated by surface-feeding, mud- and organic enrichment tolerant species of crustaceans, bivalves and gastropods (Figure 4).
- The MTBC for these sites indicates communities that are dominated by mud-tolerant species (Figure 5).
- The sediment physical characteristics displayed signs of serious degradation with much higher nutrients, organic content and heavy metal contamination than the other monitoring sites.

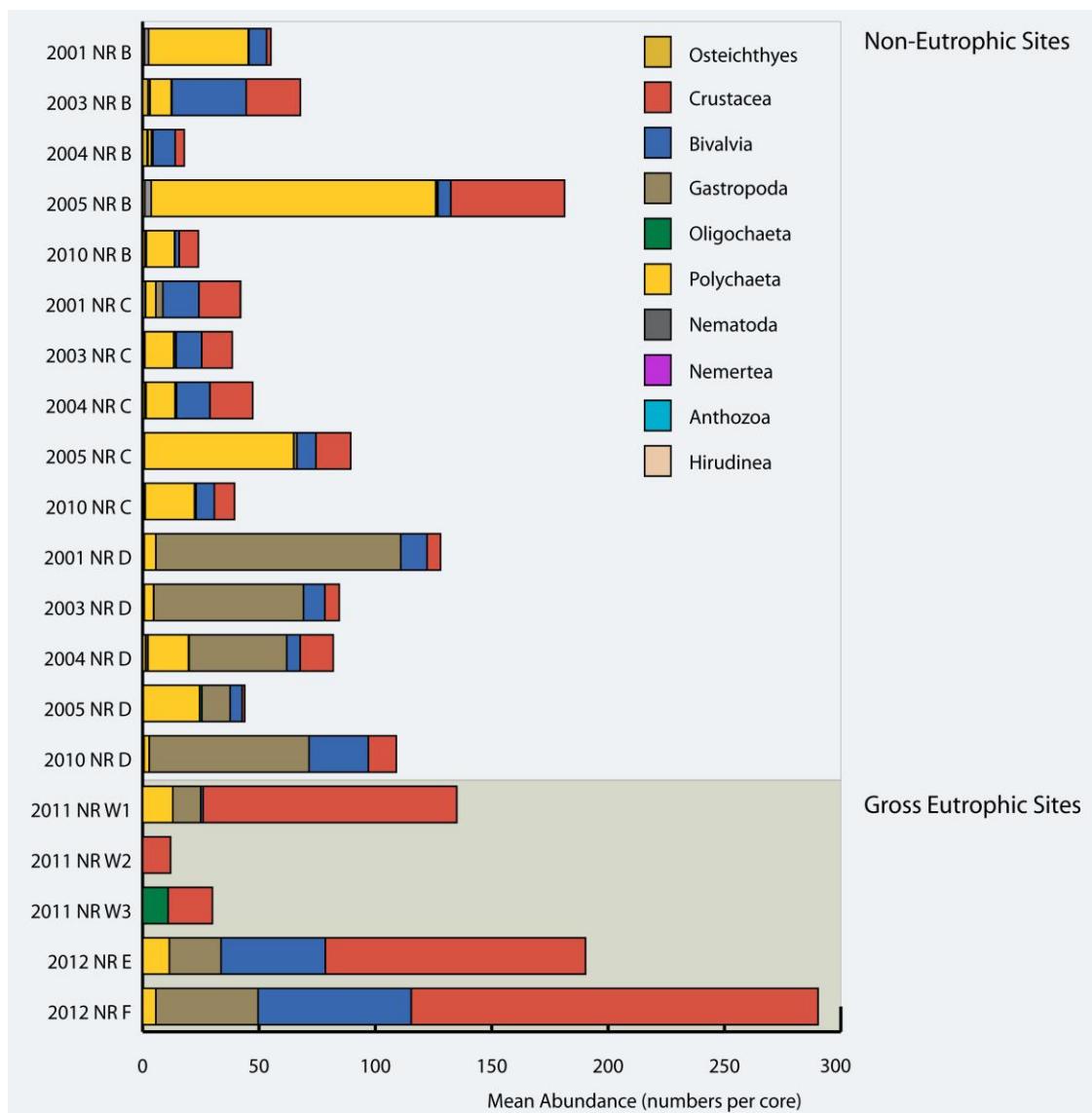


Figure 4. Mean abundance of major infauna groups from sites sampled within the New River Estuary between 2001 and 2012. Taken from Robertson & Stevens (2012).

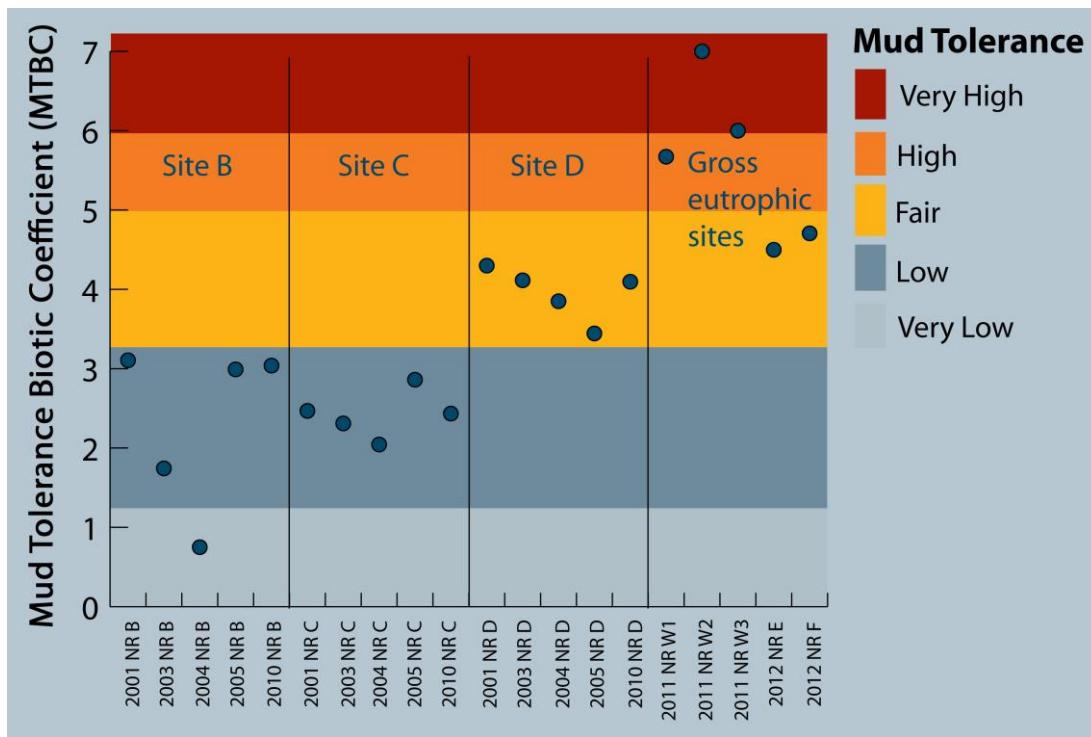


Figure 5. MTBCs calculated based on the composition of the community and its relative preference for muddy conditions for each of the New River Estuary monitoring sites between 2001 and 2012. Sites B–D are the original monitoring sites, while the remainder are the sites added in 2011/2012 to monitor the eutrophic conditions within the estuary. Adapted from Robertson & Stevens (2012).

## Limitations and points to consider

- Surveys on the physical condition of a site and the MTBC helped explain differences in biological condition among sites and over time.
- The MTBC usefully combines information from an entire community into a metric with defined standards that can be used in conservation and management.
- Care must be taken in interpreting these metrics as there is significant natural variation among estuaries, which may confound interpretation of the defined standards. To help inform management actions, these indices should be accompanied by additional information, such as sediment characteristics and known modifications to catchment and estuarine conditions.
- Collection of baseline sediment core data was needed to demonstrate the decline in pipi abundance between 2005 and 2010.
- This estuary has undergone significant and rapid degradation over the past 5 years with conditions much worse than previously observed. However, sampling at the eutrophic sites prior to this degradation would have quantified this decline further.

## References for case study B

- Borja, A.; Franco, J.; Perez, V. 2000: A marine biotic index to establish the ecological quality of soft bottom benthos within European estuarine and coastal environments. *Marine Pollution Bulletin* 40: 1100–1114.
- Borja, A.; Muxika, H. 2005: Guidelines for the use of AMBI (AZTI's Marine Biotic Index) in the assessment of benthic ecological quality. *Marine Pollution Bulletin* 50: 787–789.
- Gibbs, M.; Hewitt, J. 2004: Effects of sedimentation on macrofaunal communities: a synthesis of research studies for ARC. Technical Paper 264. NIWA Client Report: HAM2004-060.
- Robertson, B.; Stevens, L. 2010: New River Estuary—intertidal fine scale monitoring 2009/10. Prepared for Environment Southland. 35 p.
- Robertson, B.; Stevens, L. 2012: New River Estuary—fine scale monitoring of highly eutrophic arms 2011/2012. Prepared for Environment Southland. 30 p.

## Case study C

### **Case study C: temporal variation in benthic estuarine assemblages of the Auckland Region (Anderson et al. 2007)**

#### Synopsis

Auckland Regional Council established a monitoring programme in seven estuaries between 2000 and 2004. Through regular monitoring of these estuaries, the communities that occupy them, the levels of natural variability and the environmental affinities of the most abundant species have been determined.

#### Objectives

Two objectives of the study were to:

- Identify levels of natural variability through time within each of the estuaries
- Identify whether any environmental variables can be used to explain variation among sites

Further information regarding the other objectives of the study is presented in Anderson et al. 2007.

#### Sampling design and methods

- Sampling was performed in seven estuaries within the Auckland region:
  - Pūhoi (monitoring established in 2002)
  - Waiwera (monitoring established in 2002)
  - Ōrewa (monitoring established in 2002)

- Ōkura (monitoring established in 2000)
- Mangemangeroa (monitoring established in 2002)
- Tūranga (monitoring established in 2004)
- Waikōpua (monitoring established in 2004)
- Within each estuary, ten 50 × 25 m sites located on intertidal sand/mudflats ranging from -0.6 to 1.6 m tidal height were established along a gradient from the mouth of the estuary to its head. Therefore, sampling is along defined gradients of sediment inputs, salinity, tidal height and water flow.
- Each site is monitored four times per year with sampling before and after heavy rainfall in each season.
- Six replicate sediment cores (13 cm in diameter by 15 cm deep) were extracted from random coordinates within each site.
- Sediment cores were sieved using a 0.5 mm mesh and all remaining contents were preserved in 70% isopropyl alcohol.
- Indicator bivalve species (*Austrovenus stutchburyi*, *Macomona liliana* and *Paphies australis*) were measured into three size classes.
- Adjacent to each core a 20 mL syringe was pushed into the sediment to a depth of 2 cm. These six samples per site were combined and homogenised and the grain size fractions (% mud, % sand etc.) determined.
- Sediment traps were also deployed to monitor the sediment inputs into each site.

## Results

- Estuary-wide changes in community composition through time were evaluated by obtaining each species' average abundance across the 60 cores collected from each estuary at each time. These average community data were visualised using non-metric multidimensional scaling (nMDS).
- The magnitude of temporal variation within estuaries was relatively small compared to spatial variation among estuaries (Figure 6). Most of the variation in the data comes from differences among sites within estuaries, likely associated with the known environmental gradient that these sites are arranged upon (see Anderson et al. 2007).
- A canonical analysis of principal coordinates (CAP) was used to identify linear combinations of species abundances (principal component axes) which have the strongest correlation with % mud content (Anderson & Willis 2003). This allowed the identification of species that characterise muddy habitats vs. those in sandy habitats, and how the community composition changes along this gradient (Figure 7).

*MDS of faunal data averaged for each Time x Estuary combination*

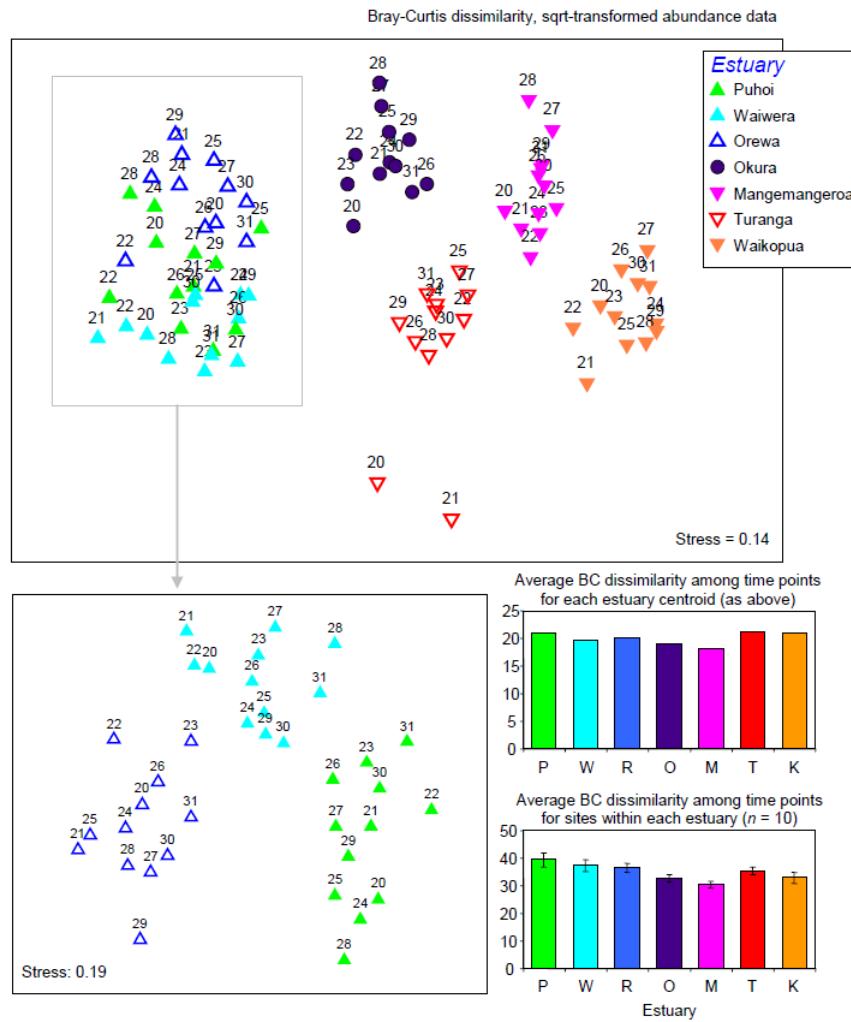


Figure 6. nMDS plots illustrating the average community at each estuary over 12 time points. The separation of points is indicative of the relative difference in average community between those estuaries/times. Different time points for each estuary tend to be clustered closely within a specific portion of the plot, whereas estuaries tend to be separated by much greater differences. This is indicative of much greater spatial than temporal variability in communities among these estuaries. Taken from Anderson et al. (2007).

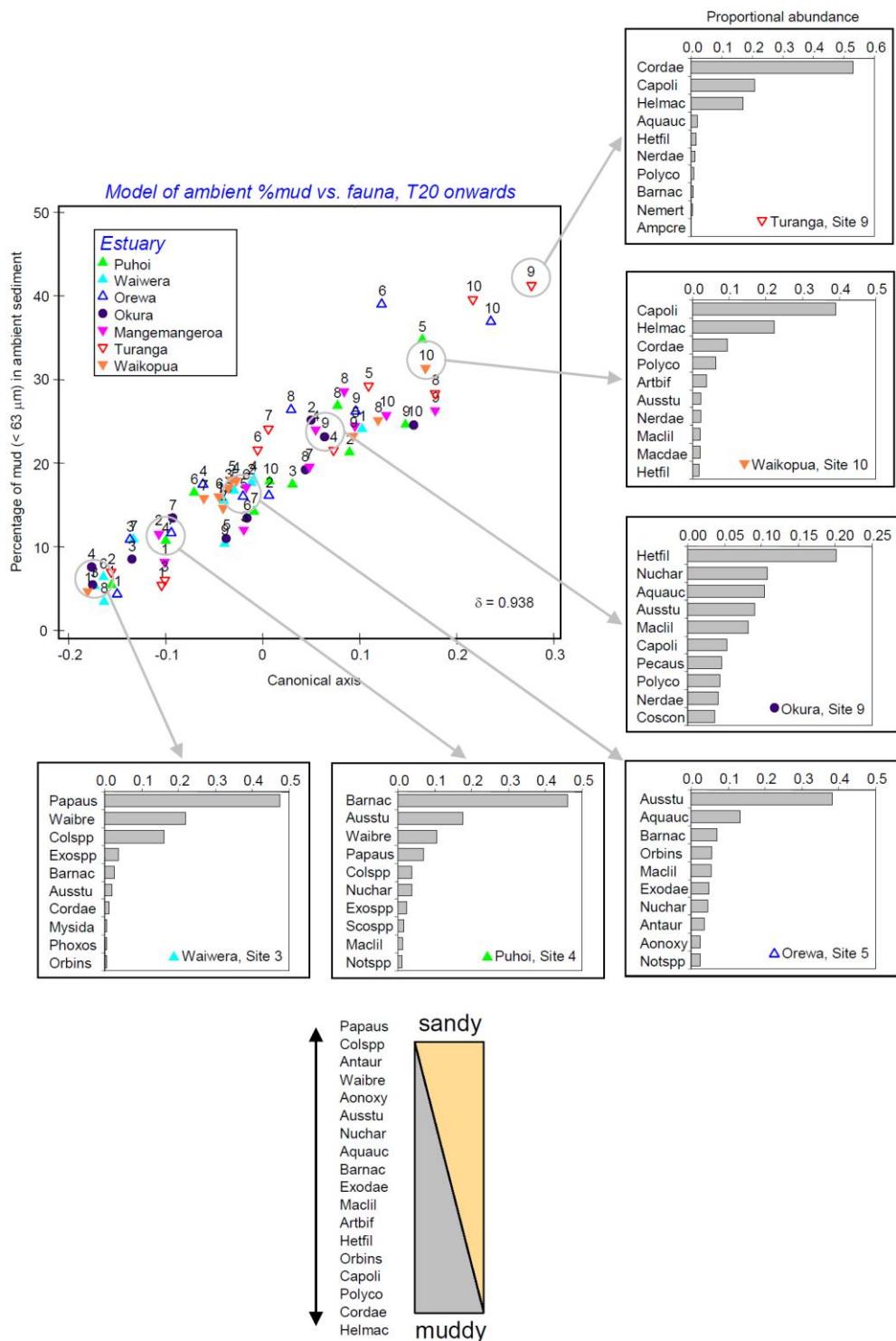


Figure 7. Results of the CAP analysis relating community differences to % mud. Side plots show the relative abundance of the most common species for selected points along this environmental gradient. The lower figure gives an indication of species turnover and preferential location along this gradient. Taken from Anderson et al. (2007).

## Limitations and points to consider

- This case study provides an indication of some questions that can be answered with, and the methods of data analysis that can be applied to, multivariate/community data.
- Although communities were very similar over short time periods, longer time-series are required to assess the effects of multi-year weather patterns such as El Niño/La Niña.
- The sampling occurred before and after rainfall, but communities tended not to differ significantly between these times, potentially due to the low rainfall limit ( $> 15$  mm in 24 hours) set as a trigger being not sufficient to cause changes in estuarine communities. Consequently, subsequent sampling was changed so that sampling would occur after more severe rainfall to try to identify rainfall effects.

## References for case study C

- Anderson, M.J.; Pawley, M.D.; Ford, R.B.; Williams, C.L. 2007: Temporal variation in benthic estuarine assemblages of the Auckland region. Prepared by UniServices for Auckland Regional Council. *Auckland Regional Council Technical Publication 348*. 102 p.
- Anderson, M.J.; Willis, T.J. 2003: Canonical analysis of principal coordinates: a useful method of constrained ordination for ecology. *Ecology* 84: 511–525.
- ter Braak, C.J.F. 1985: Correspondence analysis of incidence and abundance data: properties in terms of a unimodal response model. *Biometrics* 41: 859–873.

## Full details of technique and best practice

The exact survey/monitoring design will be governed by the research question, but the following text details the techniques and general survey design to be used when surveying soft sediment communities (also see the very detailed description of methodologies given in Robertson et al. 2002, which most of the following is based on).

### Survey design

Monitoring preparation includes developing a robust survey design, including prior consultation with experts/statisticians, to ensure the design meets the requirements to answer the research question. The following aspects need to be incorporated:

- Identification of monitoring objectives
- Statement of clear outcomes of the surveys and how they relate to the original monitoring objectives
- Determining which variables are of interest for measurement, how they are to be measured, and how the data are to be recorded (see Box 1)
- Determining the number of sites to be surveyed within the survey location, and where they are to be situated (Box 2)

- Determining the number of quadrats to be sampled within each site and their spatial arrangement (Box 2)
- Determining a survey schedule to ensure that data are collected as required over the lifetime of the study
- creation of habitat maps to identify suitable sites, and stratification of sampling among habitat types within the estuary.
- Making sure all gear is ready and datasheets are printed.
- Production of sheets of random numbers to identify sediment core locations.
- Appropriate taxonomic expertise is available to aid in species identification.

Several sites within an estuary should be identified to ensure findings are representative of the entire estuary and in line with the research question.

Site size should be c.  $30 \times 60$  m, although the exact size will be determined by local geography and the research question (Figure 8). Bamboo canes or stakes can be used to mark the four corners.

Sites should be relatively homogeneous, unvegetated, representative of the wider area, approximately uniform in elevation, and should not encompass water channels (some of these criteria may not apply depending on the research question). Habitat mapping of the estuary will significantly help with site placement and may be a prerequisite in some cases. Once a suitable site is laid out, the GPS coordinates of the four corners should be recorded.

Sediment cores and sediment samples should be collected using a stratified random sampling regime. This involves splitting the site into equal-sized blocks, with the number of blocks being equal to the number of core samples. A sediment core is then taken from each block at a randomly determined position. For example, in the case illustrated in Figure 8 the site measures  $30 \times 60$  m and 12 sediment cores are required. The site is split into blocks measuring  $10 \times 15$  m in a  $3 \times 4$  grid arrangement using the measuring tapes to mark out the blocks. Within each block a random pair of coordinates, with the X coordinate being between 0 and 10, and the Y coordinate between 0 and 15, are generated or read off of a pre-prepared sheet of random point coordinates. This determines the location of the sediment core.

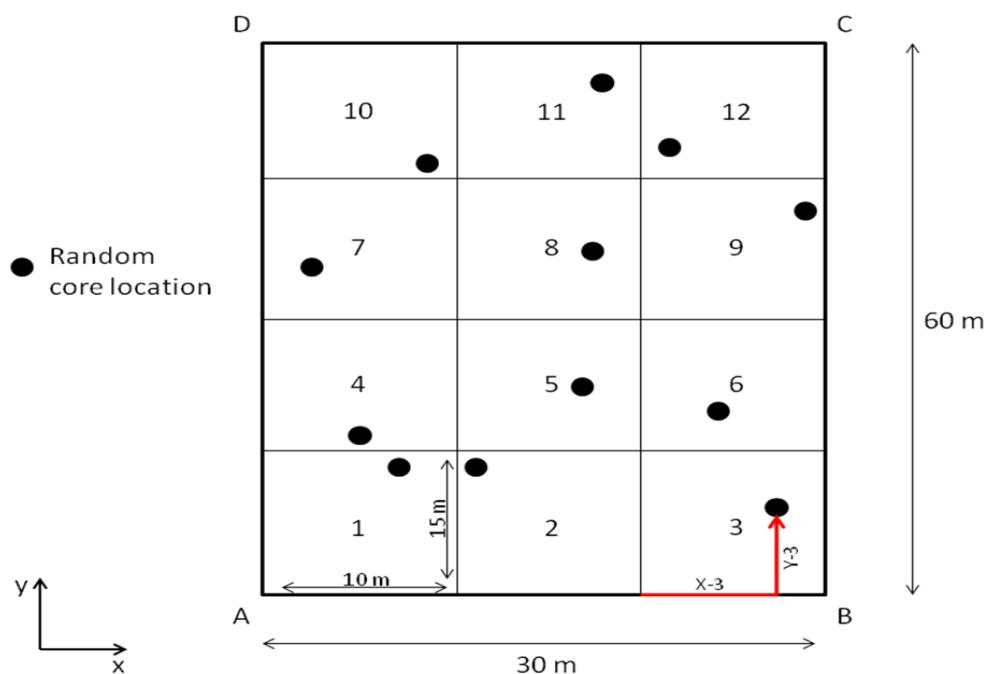


Figure 8. Schematic layout of a site and the determination of core locations. A–D indicate the four corners of the site whose GPS coordinates should be recorded; 1–12 indicate the blocks within the site which the sediment cores are extracted from. X-3 and Y-3 indicate the location of the core within block 3, determined by generating random numbers between 0 and 10 for X and 0 and 15 for Y.

## Sampling procedure

Following the determination of a clear and robust survey design, the following steps outline a typical process for conducting a transect survey.

Standard cores which are used in council monitoring programmes are cylinders of 13 cm in diameter. Corers are usually constructed from a combination of metal and an inert material (plastic or polycarbonate) to prevent chemical reaction between the corer and the sediments. These corers are pushed into the sediment to a depth of 15 cm in intertidal applications, extracting a total volume of 1990 cm<sup>3</sup>, but may be deeper in subtidal applications depending on the depth of infaunal communities. Exact core depth should be driven by the research question. For example, examining the abundance of a near-surface dwelling species in response to distance from effluent discharge would only require cores to be driven to the maximum depth that this species occupies. This would reduce processing time compared to deeper cores, while still returning the same quantity and quality of data. Deeper cores provide a more complete view of the estuarine community and would therefore be recommended for general surveillance monitoring.

Cores can then be extracted from the mudflat by digging underneath the core on one side and lifting it out of the surrounding mud. Care should be taken to level off the sediment on the bottom edge of the core to make sure sediment volumes are approximately equal among samples. The sediment can then be transferred from the plastic corer to a sieve to be processed.

## Processing of samples

The sediment from the core is sieved off by pouring seawater collected nearby over it. The sieve mesh size will determine which species/individuals will be retained and should be determined by the research question. Commonly used mesh sizes are 0.5 mm and 1 mm. A 0.5 mm mesh size sieve will retain more of the small individuals and smaller species than a 1 mm mesh, but will take longer to sieve and sample processing time will be longer. For example, surveying the size distribution of cockles throughout an estuary would not require a 0.5 mm mesh as it would increase sieving and processing time without increasing the level of information gained. However, for a general overview of the estuarine community, a finer mesh will retain more individuals and more species, and so finer meshes are more appropriate for surveillance monitoring.

Processing the samples in the field by identifying and counting all of the individuals left after sieving can be time-consuming and may limit the number of samples a survey team can collect in a day. However, it has the advantage that individuals can be directly returned to the estuary, reducing the potential for negative effects of repeated surveys. If identification and counting of all individuals is to be performed in the field, then care should be taken to return organisms to as natural an environment as possible. For example, organisms should be buried under a shallow layer of mud (c. 5 cm) to protect them from predation by birds, but not too deep as some organisms only survive in the near-surface layer.

An alternative to processing in the field is transferring the organisms into sample pots containing 70% ethanol for preservation. This allows individuals to be identified and counted later in a laboratory setting, or to be sent to experts for identification (Robertson et al. 2002; Anderson et al. 2007). All sample pots should be labelled with the date and corresponding site and block number to enable sample matching later on.

Large, charismatic and/or target species, such as cockles (*Austrovenus stutchburyi*), pipi (*Paphies australis*) and crabs (*Helice crassa*), should be measured. Sizes should be measured to the nearest millimetre using calipers as the width of the shell (from side to side, not back to front, see Figure 9) at its widest point. If these species are particularly abundant, or if measurement is done in the field, then assignment of individuals into size classes (e.g. 0–5 mm, 5–10 mm, 10–15 mm etc.) using a simple scale bar with markings at the class boundaries (Figure 9) will significantly speed up the measurement process.

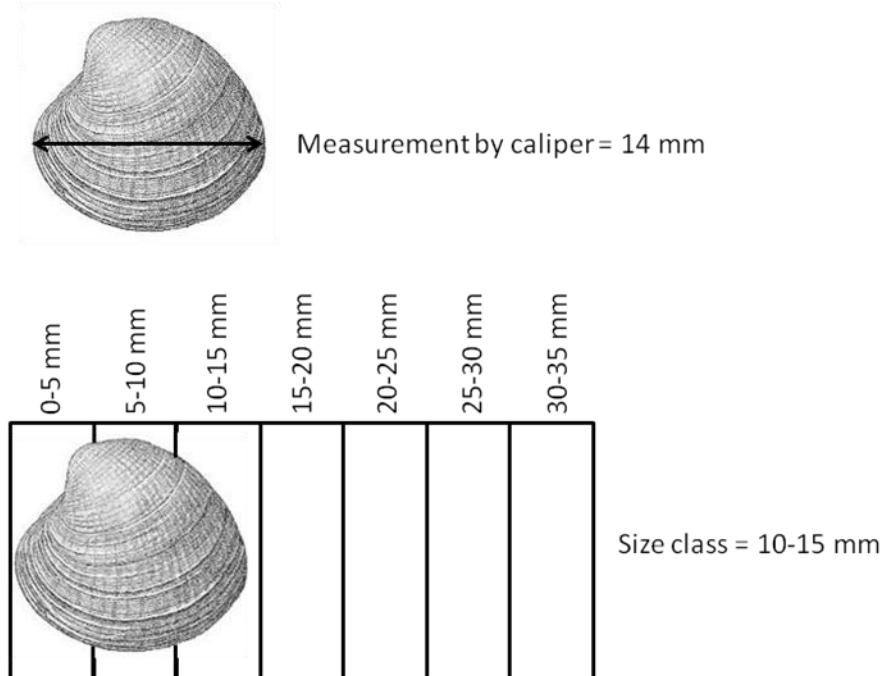


Figure 9. Correct methods for measuring shellfish. In both methods the size is determined as the maximum width of the cockle shell. Measurement of individuals into size classes can be quicker as the scale (as illustrated) can be drawn onto a plastic board or slate, preferably with a raised edge at one side so that individuals can be pressed against it for easier alignment.

## Sampling considerations

Length of time in the field may be determined by the extent of sampling to be carried out, but will more likely be determined by the length of time sites are accessible at low tide. It would be advisable to access the sites before low tide (i.e. as the tide is going out), sample throughout low tide and return before or as the tide is rising, making sure all safety precautions are adhered to. Survey teams should know the exact timing of the tides and adhere to strict time limits in the field, even if full sampling is not achieved within the time limit as tides can rise quickly creating potentially dangerous conditions.

Frequent monitoring (i.e. at intervals less than a year) may negatively affect the state of monitoring sites, particularly if samples are not processed *in situ* (i.e. organisms are destructively sampled). This could introduce a degree of bias into the monitoring data collected, giving a false impression of the changing state of the estuary. This can be avoided by recording the GPS locations of previous cores and avoiding those locations (plus a 5 m buffer zone) in the next survey (Turner et al. 2002; Singleton 2010). Alternatively, sites could be surveyed on a rotating basis to allow communities to recover from sampling.

These surveys should ideally be accompanied by analyses of the concentration of sediment contaminants (e.g. heavy metals such as arsenic, copper, chromium, zinc and organic contaminants such as polycyclic aromatic hydrocarbons and insecticides), sediment composition (including grain size analyses), redox potential depth, and nutrient and organic content of the sediment. For the above, samples of sediment should be taken at random positions within each site

block (i.e. using the same sampling protocol as sediment coring). As the surface layer of sediment is most relevant to recent sedimentary history, samples of sediment should only be taken from the top 2 cm. The analyses will usually require specialist skill and equipment; therefore, samples may have to be processed externally. The resulting information can be used to identify the drivers of changes in the biological community.

## Safety

Safety is paramount during any survey activity. The safety recommendations below are provided as general guidance, but it is imperative that the survey leader understands all risks associated with the activity, always uses caution, and develops a Safety Plan for the survey activity and location (DOC staff should use RiskManager, and non-Departmental staff should consult WorkSafe New Zealand's 4-step risk management<sup>2</sup> or their own organisation's safety plans). Safety Plans should include resources (e.g. equipment, boats, communication, support, personal protective equipment), environmental hazards or considerations (e.g. remoteness, surf zones), personnel (experience, training, physical and mental fitness), weather and mission complexity. Following a thorough safety briefing, all team members should read and then sign the Safety Plan.

Specifically, the survey must be planned so that:

- A minimum of two people make up the survey team
- All personnel are operating within the limits of their training and experience
- The magnitude and complexity of the survey are relevant for the planned duration of the survey

## Quality control

Quality control measures should be used to ensure that data quality is consistent with previous surveys. These measures are briefly described here (see Singleton 2010 for a full description). Quality control measures should be employed to ensure that: (1) the number of organisms found in each core is accurately identified and counted, and (2) individuals are identified to the correct taxonomic group and counted correctly.

The quality control measure for counting errors involves complete re-sorting (separating biological material from shell hash and non-living biological material) and counting of the sieved contents of a subset of the samples (Singleton 2010 recommends a ratio of 1 in 6 samples) following the original count. This should be performed by an experienced sorter, other than the original sorter.

The sorting efficiency percentage is:

$$\frac{\# \text{ organisms originally sorted}}{\# \text{ organisms originally sorted} + \# \text{ organisms found in re-sort}} \times 100$$

<sup>2</sup> <http://www.worksafe.govt.nz/worksafe/hswa/health-safety/how-to-manage-work-risks>



If sorting efficiency is greater than 95%, then no action is suggested. However, if less than 95%, all samples should be re-sorted.

Once this quality control criterion has been met, organisms can be identified to species or species-group level and counted. Quality control of species identification and counting is performed by an experienced recorder, other than the original researcher, recounting and identifying all individuals within a sample (ratio of 1 in 6 samples) of the previously processed cores.

The identification efficiency percentage is:

$$\frac{\text{# organisms in recount} - \text{number of errors}}{\text{# organisms in recount}} \times 100$$

Where the number of errors is defined as the number of individuals misidentified/miscounted and can include the following errors:

- Counting errors (e.g. 6 individuals of a species instead of 7)
- Identification errors (e.g. identifying species X as species Y)
- Unrecorded species errors (e.g. not recording species X when it is present)
- Recording errors (e.g. recording species X as species Y on the sheet)
- Individuals overlooked (e.g. organisms that were missed)

If the identification efficiency is less than 90%, all samples are rechecked for the above errors. An example of quality assurance is given in Figure 10. Once quality assurance has been satisfied, the data along with associated metadata and quality assurance data should be entered into spreadsheets/databases.

	A	B	C	D	E	F	G	H	I	J	K
1	Date	21-Jan-12									
2	Original sorter/recorder	Jo Smith									
3	Experienced sorter/recorder	Pete Jones									
4			Percent sorting efficiency			Species identification and counting					
5	Site	Sample	Original number	Resort number	% Sorting efficiency		Recount number	Number of errors	% Identification efficiency		
6	Por-A	2	156	1	99.36		157	12	92.36		
7	Por-A	5	169	7	96.02		176	15	91.48		
8	Por-A	8	126	0	100.00		126	4	96.83		
9	Por-B	3	189	6	96.92		195	7	96.41		
10	Por-B	4	169	8	95.48		177	15	91.53		
11	Por-B	7	136	1	99.27		137	6	95.62		
12											
13											
14											
15											

Figure 10. Example layout and format of quality assurance for sorting efficiency percentage and species identification and counting for a subset of sediment cores.

## References and further reading

- Anderson, M.J. 2001: A new method for non-parametric multivariate analysis of variance. *Austral Ecology* 26: 32–46.
- Anderson, M.J.; Gorley, R.N.; Clarke, K.R. 2008: PERMANOVA+ for PRIMER: Guide to software and statistical methods. PRIMER-E, Plymouth, UK.
- Anderson, M.J.; Pawley, M.D.; Ford, R.B.; Williams, C.L. 2007: Temporal variation in benthic estuarine assemblages of the Auckland region. Prepared by UniServices for Auckland Regional Council. *Auckland Regional Council Technical Publication 348*. 102 p.
- Anderson, M.J.; Willis, T.J. 2003: Canonical analysis of principal coordinates: a useful method of constrained ordination for ecology. *Ecology* 84: 511–525.
- Borja, A.; Franco, J.; Perez, V. 2000: A marine biotic index to establish the ecological quality of soft bottom benthos within European estuarine and coastal environments. *Marine Pollution Bulletin* 40: 1100–1114.
- Borja, A.; Muxika, H. 2005: Guidelines for the use of AMBI (AZTI's Marine Biotic Index) in the assessment of benthic ecological quality. *Marine Pollution Bulletin* 50: 787–789.
- Clarke, K.R.; Ainsworth, M. 1993: A method of linking multivariate community structure to environmental variables. *Marine Ecology Progress Series* 92: 205.
- Clarke, K.R.; Gorley, R.N. 2006: PRIMER v.6: User manual/tutorial. PRIMER-E, Plymouth, UK.
- Clarke, K.R.; Warwick, R.M. 2001: Change in marine communities: an approach to statistical analysis and interpretation. 2nd edition. PRIMER-E, Plymouth, UK.
- Cumming, G. 2012: Understanding the new statistics: effect sizes, confidence intervals, and meta-analysis. Routledge, New York.
- Ford, R.B.; Thrush, S.F.; Probert, P.K. 1999: Macrofaunal colonisation of disturbances on an intertidal sandflat: the influence of season and buried algae. *Marine Ecology Progress Series* 191:163–174.
- Gibbs, M.; Hewitt, J. 2004: Effects of sedimentation on macrofaunal communities: a synthesis of research studies for ARC. *Technical Paper 264*. NIWA Client Report: HAM2004-060.
- Legendre, P.; Anderson, M.J. 1999: Distance-based redundancy analysis: testing multispecies responses in multifactorial ecological experiments. *Ecological Monographs* 69: 1–24.
- McCullagh, P.; Nelder, J.A. 1989: Generalized linear models (Vol. 2). Chapman and Hall, London.

Pridmore, R.D.; Thrush, S.F.; Hewitt, J.E.; Roper, D.S. 1990: Macrofaunal community composition of six intertidal sandflats in Manukau Harbour, New Zealand. *New Zealand Journal of Marine and Freshwater Research* 24: 81–96.

Robertson, B.M.; Gillespie, P.A.; Asher, R.A.; Frisk, S.; Keeley, N.B.; Hopkins, G.A.; Thompson, S.J.; Tuckey, B.J. 2002: Estuarine environmental assessment and monitoring: a national protocol. Part A: Development, Part B: Appendices, and Part C: Application. Prepared for supporting councils and the Ministry for the Environment. Sustainable Management Fund Contract No. 5096. Part A: 93 p. Part B: 159 p. Part C: 40 p plus field sheets.

Robertson, B.; Stevens, L. 2010: New River Estuary—intertidal fine scale monitoring 2009/10. Prepared for Environment Southland. 35 p.

Robertson, B.; Stevens, L. 2012: New River Estuary—fine scale monitoring of highly eutrophic arms 2011/2012. Prepared for Environment Southland. 30 p.

Singleton, N. 2010: Regional Estuary Monitoring Programme (REMP) data report: benthic macrofauna communities and sediments—July 2008 to April 2009. Southern Firth of Thames and Whaingaroa (Raglan) Harbour. *Environment Waikato Internal Series 2010/37*. Hamilton, Waikato Regional Council (Environment Waikato).

ter Braak, C.J.F. 1985: Correspondence analysis of incidence and abundance data: properties in terms of a unimodal response model. *Biometrics* 41: 859–873.

Thrush, S.F.; Hewitt, J.E.; Norkko, A.; Nicholls, P.E.; Funnell, G.A.; Ellis, J.I. 2003: Habitat change in estuaries: predicting broad-scale responses of intertidal macrofauna to sediment mud content. *Marine Ecology Progress Series* 263: 101–112.

Thrush S.F.; Pridmore R.D.; Hewitt J.E.; Roper D.S. 1988: Design of an ecological monitoring programme for the Manukau Harbour. Water Quality Consultancy Report No. 7099. Auckland Regional Water Board, Auckland.

Turner, S. 2001: Monitoring the region's estuaries: intertidal sand-flat benthic communities. *Environment Waikato Internal Series 2000/11*. Hamilton, Waikato Regional Council (Environment Waikato).

Turner, S.; Carter, N. 2004: Regional Estuary Monitoring Programme: benthic macrofauna communities—April 2001 to April 2002. Southern Firth of Thames and Whaingaroa (Raglan) Harbour. *Environment Waikato Internal Series 2004/08*. Hamilton, Waikato Regional Council (Environment Waikato).

Turner, S.; Gibberd, B.; Crozier, J. 2002: Regional Estuary Monitoring Programme—pilot study. *Environment Waikato Internal Series 2002/02*. Hamilton, Waikato Regional Council (Environment Waikato).

Underwood, A.J. 1997: Experiments in ecology: their logical design and interpretation using analysis of variance. Cambridge University Press, Cambridge, UK.

## Appendix A

The following Department of Conservation documents are referred to in this method:

doccm-1163829 MPAMAR metadata: national

doccm-146272 Standard inventory and monitoring project plan