

# Freshwater ecology: quantitative macroinvertebrate sampling in soft-bottomed streams

Version 1.0



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## Synopsis

The protocol described here is based upon that described by Stark et al. (2001)<sup>1</sup> as being an appropriate minimum requirement. Quantitative sampling of soft-bottomed, wadeable New Zealand streams is designed to produce high-precision estimates of the population density of macroinvertebrates at a sampling location. Consequently, this data requires greater cost, time and resources to acquire and is only justified in situations such as threatened species monitoring or research where density effects are of interest. There are no limits on the metrics and analyses which can be performed on quantitative count data; see the 'Introduction to macroinvertebrate monitoring in freshwater ecosystems' (docdm-724991) to consider appropriate metrics to meet specific objectives. Always consult a biometrician or experienced freshwater ecologist to confirm that your design and methods are appropriate to meet your objectives.



Figure 1. A good example of a soft-bottomed stream dominated by growths of *Egeria* sp.

This protocol details a quantitative method to sample macrophytes in soft-bottomed streams. Rather than providing an estimate of individuals per unit area, the method provides density per unit volume (wet or dry) of macrophytes. Quantitative sampling of invertebrate communities in macrophytes requires no specialised equipment and most soft-bottomed streams will have macrophytes present. Different macrophyte species may have distinct assemblages, therefore the

<sup>1</sup> <http://www.cawthron.org.nz/coastal-freshwater-resources/downloads/protocols-full-manual.pdf>

species used for collection should be standardised and the composition of macrophyte communities noted. It is also worth noting the condition of macrophytes (e.g. senescent, flowering or covered in epiphytic algae). Sampling effort should be standardised such that results are comparable. Stark et al. (2001) recommend that a sampling effort three times greater than that required for hard-bottomed streams should be employed. Thus, an area of 3 m<sup>2</sup> is required to characterise soft-bottomed stream communities. Furthermore, it is recommended that four replicate samples should be taken to provide reliable estimates of variance in invertebrate densities.

Sampling is performed moving upstream using a kick-net. Select the correct species of macrophyte to be sampled and move the net over the tips of the growth before breaking off the required amount. Four replicate samples are recommended. 1.5 L to 2 L of weed is equivalent to approximately 100 g wet weight of the top 20–30 cm of the plant. Each replicate sample should be placed in a separate bucket and rinsed thoroughly. Shaking of a sealed bucket effectively dislodges many individuals. After each rinse, pour dislodged invertebrates onto a 0.5 mm sieve and transfer into a sample pottle.

Each sample should have preservative (usually 70% ethanol) added as soon as possible. A unique identifying code must be clearly marked on the lid and on a slip of waterproof paper inside the pottle (Fig. 2). Pottles should also be marked with the location, date and the field operative names.

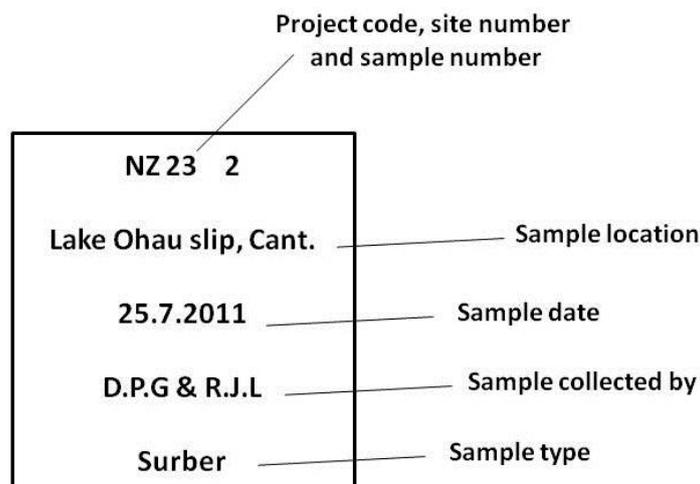


Figure 2. An example wet label with the required information to identify each sample should be written in pencil on waterproof paper.

Water should be drained from the remaining plant material which is then weighed using a field balance. The wet weight of weed associated with each replicate sample should be noted. If greater precision is required, plant material may be returned to the laboratory dried thoroughly and weighed.

Samples should be transported to a laboratory for storage prior to processing. Processing methods are detailed in Stark et al. (2001), but require experienced taxonomists to oversee the process.

## Assumptions

- The sample is representative of the wider macroinvertebrate population.
- Sampling effort and duration is standardised across all sample sites.

## Advantages

- Quantitative sampling provides high-precision information about the richness and composition of macroinvertebrate communities.
- Allows the user to isolate a known volume of macrophytes for the sample.

## Disadvantages

- Generates a greater number of replicate samples than a semi-quantitative method. There will be a significant increase in time and cost of processing.
- Unsuitable where substrates macrophyte growths are not present. In this situation it is recommended to sample known areas of woody debris if available.

## Suitability for inventory

This technique is not suitable for inventory which does not require quantitative estimates of population size. Collection and processing of quantitative macrophyte samples for inventory would be a considerable waste of time and resources. An exception to this might be in the case of sampling for threatened species that have a high affinity for macrophytes.

## Suitability for monitoring

This method is suitable for monitoring where the density of individuals is considered to be of interest. If high-precision estimates of population density are not important, consider using a more efficient semi-quantitative sampling or processing method.

## Skills

Field observers will require:

- Basic training in stream macroinvertebrate and habitat sampling
- Basic outdoor and river-crossing skills
- A reasonable level of fitness

Study design, sample processing and quality control are specialised processes that require input from a freshwater specialist.

## Resources

Quantitative sampling of soft-bottomed streams may be carried out by a single field operative. However, in the interests of safety it is recommended that sampling is done by teams of at least two people.

Standard equipment includes:

- Waterproof notebook or field data sheets
- Pencil
- Permanent marker pen
- Wet labels
- Waders or gumboots, dependent on stream depth
- GPS and map

Specialist equipment required:

- Kick-net sampler (0.1 m<sup>2</sup>, 0.5 mm mesh)
- 10 litre bucket with lid
- Field balance capable of 5 g precision
- Ziploc plastic bags (if plant material to be dried)
- Sieve or sieve bucket
- Plastic sample containers (usually 500–1000 ml volume)
- Preservative (usually 70% ethanol)

## Minimum attributes

Consistent measurement and recording of these attributes is critical for the implementation of the method. Other attributes may be optional depending on your objective. For more information refer to [‘Full details of technique and best practice’](#).

DOC staff must complete a ‘Standard inventory and monitoring project plan’ (docdm-146272).

The more information that is collected at each site, the more thorough and complete will be any interpretation of the biological data collected. However, some basic information should be recorded with each sample collected:

- Substrate composition
- Riparian vegetation
- Stream width
- Stream depth
- Stream velocity

- Periphyton and macrophyte community composition

It is also commonplace to collect basic water chemistry information where possible. Temperature (°C), electrical conductivity ( $\mu\text{S}$ ), pH and dissolved oxygen may all be measured by handheld meters to inform biological data. Some basic habitat and sites notes are also worthwhile, e.g. the occurrence of stock at the site or evidence of recent flooding. The '[Stream habitat assessment field sheet](#)' (docdm-761873) is a good guide to the basic information that can be collected without recourse to specialised equipment or processing in a laboratory. Basic training in the use of this habitat sheet or a thorough perusal of Harding et al. (2009) is required before use of this habitat assessment sheet.<sup>2</sup> As with all visual and qualitative assessments it is important to standardise collection protocols within a group of field observers or within a particular project. There is considerable opportunity for user bias with this method of habitat assessment.

## Data storage

It is quite likely that biological sample processing will be outsourced to an accredited laboratory. During sample processing, data is conventionally recorded on a hardcopy data sheet prior to transfer to an electronic format. Hardcopy sheets will be clearly marked with the details of the project and identity of samples. The format of hardcopy data sheets is normally columns representing samples and rows for each species or taxa group. Data should be entered into an electronic media in the same format to avoid confusion (see 'Stream invertebrate data sheet example'—docdm-761858). Electronic data sheets should contain all the information required to identify each sample, and any habitat or water chemistry data that was collected simultaneously may be appended on a separate worksheet within the electronic file (usually Excel).

It is important that habitat and water chemistry data are entered in a comparable format to biological data, i.e. columns as sites, and this should be done as soon as possible by the field operative so that details are fresh. All hardcopies of habitat data and notes should be labelled and stored in a project file and retained.

All electronic files should have a notes sheet which details any relevant information for future users. In particular each user, beginning with the field operative who enters the data, should record details of any changes to the data, including when and why they were made. It is also recommended to retain a single version of the data which has undergone quality control and may not be altered. All analysis is performed on copies of this master sheet.

Forward copies of completed survey sheets to the survey administrator, or enter data into an appropriate spreadsheet as soon as possible. Collate, consolidate and store survey information securely, also as soon as possible, and preferably immediately on return from the field. The key steps here are data entry, storage and maintenance for later analysis, followed by copying and data backup for security.

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<sup>2</sup> <http://www.cawthron.org.nz/coastal-freshwater-resources/downloads/stream-habitat-assessment-protocols.pdf>

Summarise the results in a spreadsheet or equivalent. Arrange data as 'column variables'—i.e. arrange data from each field on the data sheet (date, time, location, plot designation, number seen, identity, etc.) in columns, with each row representing the occasion on which a given survey plot was sampled.

If data storage is designed well at the outset, it will make the job of analysis and interpretation much easier. Before storing data, check for missing information and errors, and ensure metadata are recorded.

Storage tools can be either manual or electronic systems (or both, preferably). They will usually be summary sheets, other physical filing systems, or electronic spreadsheets and databases. Use appropriate file formats such as .xls, .txt, .dbf or specific analysis software formats. Copy and/or backup all data, whether electronic, data sheets, metadata or site access descriptions, preferably offline if the primary storage location is part of a networked system. Store the copy at a separate location for security purposes.

## Analysis, interpretation and reporting

Seek statistical advice from a biometrician or suitably experienced person prior to undertaking any analysis.

The invertebrate data derived from macrophyte sampling are either semi-quantitative fixed counts or more commonly full counts of all individuals. They are high-precision estimates of population size. There is no limit to the indices or analyses that can be produced with this data. Common basic indices calculated from this data are:

- Taxa richness
- Richness of Ephemeroptera, Plecoptera and Trichoptera (EPT) taxa or % EPT abundance
- Macroinvertebrate Community suite of indices for soft-bottomed streams (MCI-sb), Semi-Quantitative Macroinvertebrate Community Index for soft-bottomed streams (SQMCI-sb) (Stark & Maxted 2007) and the Quantitative Macroinvertebrate Community Index for soft-bottomed streams (QMCI-sb) (Stark & Maxted 2007)

### Taxa richness

Taxa richness is simply the number of taxa that were found at a site and is routinely used in studies of ecosystem condition, restoration monitoring and research. Sites may be compared in terms of taxa richness provided the sampling effort and taxonomic resolution at each site is standardised. If groups of sites are to be compared, e.g. forest streams versus grassland streams, then it is important that equal numbers of each site type have been sampled. If this assumption is violated the degree of difference must be noted or comparisons will require rarefaction and a biometrician should be consulted (Magurran 2004). If sample numbers and effort are balanced, i.e. equal, then basic Analyses of Variance (ANOVA) or *t*-tests can be used to compare between the mean values for habitat types. Alternatively, instead of comparing richness between groups, a gradient approach may be used whereby the richness of taxa at each site is compared to the value for an

environmental condition at that site. Such a correlative approach is more appropriate when sites do not fit into meaningful groupings.

## EPT richness

EPT richness is the number of taxa which are members of the Ephemeroptera (mayfly), Trichoptera (caddis fly) and Plecoptera (stonefly) orders and is commonly used for ecosystem condition monitoring. Many of the species within these groups require undisturbed habitats and so this metric may be more sensitive to impacts than taxa richness alone. Accordingly, EPT richness may be presented as a proportion of total richness, e.g. % EPT.

## MCI-sb

The Macroinvertebrate Community Index (MCI) was initially proposed by Stark (1985) to assess organic enrichment in the stony riffles of New Zealand streams and rivers. A variant index for soft bottomed streams (MCI-sb) was created by Stark & Maxted (2007) which uses the same formula as the MCI but different tolerance values for individual taxa. Each taxa is assigned a score (1–10) which represents its tolerance to pollution. The MCI-sb score for a sample is calculated thus:

$$= 20 \sum a_i / S$$

Where  $a_i$  is the MCI-sb tolerance score for the  $i^{\text{th}}$  taxon and  $S$  is the total number of taxa. Taxon tolerance scores can be found in Table 3.

MCI values range from 0–200, which may be interpreted in terms of water quality according to Table 1. The same analyses and assumptions apply as for taxa richness and EPT richness. All comparisons should be made with reference to habitat data.

Table 1. Interpretation of MCI, QMCI and SQMCI values from stony riffles (after Boothroyd & Stark 2000).

Interpretation	MCI	QMCI & SQMCI
Clean water	> 120	> 6.00
Doubtful quality of possible mild pollution	100–119	5.00–5.99
Probable moderate pollution	80–99	4.00–4.99
Probable severe pollution	< 80	< 4.00

Coded abundance and fixed count data provide rough estimates of the relative numbers of the different taxa and so provide the ability to calculate an additional index—the Semi-Quantitative Macroinvertebrate Community Index (SQMCI-sb). If coded abundance data are received in alpha code form they may be converted to numerical form according to Table 2. Like the MCI-sb, SQMCI-sb is designed to be calculated from kick-net samples collected over a standardised area (0.3–0.6 m<sup>2</sup>), but unlike the MCI-sb, SQMCI-sb scores range from 0–10. The SQMCI-sb is calculated thus:

$$= \sum (c_i a_i) / M$$

Where  $c_i$  is the coded abundance of individuals in the  $i^{\text{th}}$  taxon and M is the coded abundance total number of individuals. Scores may be interpreted in terms of water quality according to Table 1 and are directly comparable with QMCI-sb scores, but not MCI-sb. The same analyses and assumptions apply as for taxa richness and EPT richness. All comparisons should be made with reference to habitat data.

Table 2. Abundance classes, count ranges and coded abundance used for the calculation of SQMCI-sb scores. Abundance class may be converted to coded abundance for the purposes of analysis. (Reproduced from Stark 1998.)

Abundance class	Counts	Coded abundance
R—rare	1–4	1
C—common	5–19	5
A—abundant	20–99	20
VA—very abundant	100–499	100
VVA—very very abundant	500+	500

The real value of full count data is in allowing the calculation of the QMCI-sb which is the quantitative variant of the MCI-sb. The QMCI-sb is calculated thus:

$$= \sum (n_i a_i) / N$$

Where  $n_i$  is the number of individuals in the  $i^{th}$  taxon and N is the total number of individuals. Scores may be interpreted in terms of water quality according to Table 1 and are directly comparable with SQMCI-sb scores, but not MCI-sb.

## Community composition

Quantitative macroinvertebrate data may also be used to compare the abundance of groups of taxa between sites or examine changes in the dominant taxa at a site. Relative or absolute abundance of different taxa groups are commonly displayed as a stacked bar graph where each column represents a location or sampling event and the column is divided vertically according to the proportional or absolute abundance of major taxa groups. Taxa groupings can be defined according to the objectives of the study, but conventionally approximate the major orders, such as Ephemeroptera, Trichoptera, Mollusca and other. An example of a stacked bar graph is shown in [‘Case study A’](#). A further basic descriptive technique for comparing invertebrate communities between sites/occasions would be to list the five most abundant taxa.

It is commonplace to provide a number of these summary statistics, such as richness and abundance of taxa along with habitat summary data, prior to any more complicated analyses in order to ‘set the scene’ for the reader.

There are numerous indices and statistical techniques used for describing richness and diversity (a function of the number of both taxa and individuals) which are available. However, an experienced biometrician / freshwater ecologist should be consulted before applying these techniques. The best overview of available statistical measures of diversity may be found in Magurran (2004). Further, ‘multivariate’ techniques are also available for investigating differences in entire communities, often in relation to accompanying habitat data; however, these techniques require an experienced practitioner.

The majority of collation and calculation described here can be performed in a basic spreadsheet package such as Excel, although there are a variety of commercial and freeware packages available to calculate summary statistics and perform more in-depth analyses. However, beyond the basic descriptive statistics, such as richness, MCI and summary plots, the user will require specific training and experience.

Table 3. Recommended minimum level of macroinvertebrate identification (based on Stark 1998; Winterbourn et al. 2000) with associated MCI, SQMCI and QMCI tolerance values.

<b>INSECTA</b>		<b>Neuroptera</b>		<b>Trichoptera (Cont.)</b>	
<b>Ephemeroptera</b>		<i>Kempynus</i>	5	<i>Hydrobiosella</i>	9
<i>Acanthophlebia</i>	7	<b>Diptera</b>		<i>Hydrobiosis</i>	5
<i>Ameletopsis</i>	10	<i>Aphrophila</i>	5	<i>Hydrochorema</i>	9
<i>Arachnocolus</i>	8	<i>Austrosimulium</i>	3	<i>Kokiria</i>	9
<i>Atalophlebioides</i>	9	<i>Calopsectra</i>	4	<i>Neurochorema</i>	6
<i>Austroclima</i>	9	Ceratopogonidae	3	Oeconesidae	9
<i>Coloburiscus</i>	9	<i>Chironomus</i>	1	<i>Olinga</i>	9
<i>Deleatidium</i>	8	<i>Corynoneura</i>	2	<i>Orthopsyche</i>	9
<i>Ichthybotus</i>	8	<i>Cryptochironomus</i>	3	<i>Oxyethira</i>	2
<i>Isotrachus</i>	8	<i>Culex</i>	3	<i>Paroxyethira</i>	2
<i>Mauiulus</i>	5	Culicidae	3	<i>Philorheithrus</i>	8
<i>Neozephlebia</i>	7	Dolichopodidae	3	<i>Plectrocnemia</i>	8
<i>Nesameletus</i>	9	Empididae	3	<i>Polypsectopus</i>	8
<i>Oniscigaster</i>	10	Ephydriidae	4	<i>Psilochorema</i>	8
<i>Rallidens</i>	9	<i>Eriopterini</i>	9	<i>Pycnocentrella</i>	9
<i>Siphlaenigma</i>	9	<i>Harrisius</i>	6	<i>Pycnocentria</i>	7
<i>Zephlebia</i>	7	<i>Hexatomini</i>	5	<i>Pycnocentrodes</i>	5
<b>Plecoptera</b>		<i>Limonia</i>	6	<i>Rakiura</i>	10
<i>Acroperla</i>	5	<i>Lobodiamesa</i>	5	<i>Tiphobiosis</i>	6
<i>Austroperla</i>	9	<i>Maoridiamesa</i>	3	<i>Triplectides</i>	5
<i>Cristaperla</i>	8	<i>Mischoderus</i>	4	<i>Triplectidina</i>	5
<i>Halticoperla</i>	8	<i>Molophilus</i>	5	<i>Zelolessica</i>	10
<i>Megaleptoperla</i>	9	Muscidae	3	<b>Lepidoptera</b>	
<i>Nesoperla</i>	5	<i>Nannochorista</i>	7	<i>Hygraula</i>	4
<i>Spaniocerca</i>	8	<i>Neocurupira</i>	7	<b>Collembola</b>	6
<i>Spaniocercoides</i>	8	<i>Neoscatella</i>	7	ACARINA	5
<i>Stenoperla</i>	10	<i>Nothodixa</i>	5	<b>CRUSTACEA</b>	
<i>Taraperla</i>	5	<i>Orthocladinae</i>	2	Amphipoda	5
<i>Zelandobius</i>	5	<i>Parochlus</i>	8	Copepoda	5
<i>Zelandoperla</i>	10	<i>Paradixa</i>	4	Cladocera	5
<i>Megaloptera</i>		<i>Paralimnophila</i>	6	Isopoda	5
<i>Archichauliodes</i>	7	<i>Paucispinigera</i>	6	Ostracoda	3
<b>Odonata</b>		Pelecorhynchidae	9	<i>Paranephrops</i>	5
<i>Aeshna</i>	5	<i>Peritheates</i>	7	<i>Paratya</i>	5
<i>Antipodochlora</i>	6	Podonominae	8	<i>Tanaidacea</i>	4
<i>Austrolestes</i>	6	<i>Polypedilum</i>	3	<b>MOLLUSCA</b>	

<i>Hemicordulia</i>	5	Psychodidae	1	<i>Ferrissia/Grunlachia</i>	3
<i>Xanthocnemis</i>	5	Sciomyzidae	3	<i>Gyraulus</i>	3
<i>Procordulia</i>	6	Stratiomyidae	5	<i>Hyridella</i>	3
<b>Hemiptera</b>		Syrphidae	1	<i>Latia</i>	3
<i>Anisops</i>	5	Tabanidae	3	<i>Lymnaea/ Austropeplia</i>	3
<i>Diaprepocoris</i>	5	Tanypodinae	5	<i>Melanopsis</i>	3
<i>Microvelia</i>	5	<i>Tanytarsini</i>	3	<i>Physa</i>	3
<i>Sigara</i>	5	<i>Tanytarsus</i>	3	<i>Physastra</i>	5
<b>Coleoptera</b>		Thaumaleidae	9	<i>Potamopyrgus</i>	4
<i>Antiporus</i>	5	<i>Zelandotipula</i>	6	Sphaeriidae	3
<i>Berosus</i>	5	<b>Trichoptera</b>		OLIGOCHAETA	1
Dytiscidae	5	<i>Alloecentrella</i>	9	HIRUDINEA	3
Elmidae	6	<i>Aoteapsyche</i>	4	PLATYHELMINTHES	3
<i>Homeodytes</i>	5	<i>Beraeoptera</i>	8	NEMATODA	3
Hydraenidae	8	<i>Confluens</i>	5	NEMATOMORPHA	3
Hydrophilidae	5	<i>Conuxia</i>	8	NEMERTEA	3
<i>Liodessus</i>	5	<i>Costachorema</i>	7	<b>COELENTERATA</b>	
Ptilodactylidae	8	<i>Edpercivalia</i>	9	Hydra	3
<i>Rhantus</i>	5	<i>Ecnomidae/Zelandotipula</i>	8		
Scirtidae	8	<i>Helicopsyche</i>	10		
Staphylinidae	5	<i>Hudsonema</i>	6		

## Case study A

### Case study A: the influence of substrate type and physico-chemical conditions on macroinvertebrate faunas and biotic indices of some lowland Waikato, New Zealand, streams

#### Synopsis

This study used quantitative sampling techniques, but analysis methods would suit semi-quantitative data except where noted. Data used with permission of Kevin Collier (Collier et al. 1998). If your sampling is quantitative then more emphasis may be placed on estimates of abundance and your study will be able to detect subtle shifts in community composition. In a semi-quantitative context it is relative, or proportional, abundance and indices of richness which are of primary interest.

Lowland streams in agricultural areas often appear degraded because of high turbidity, extensive bank erosion, high macrophyte abundance and silt-laden beds. However, these soft-bottomed streams do contain macroinvertebrate populations which can be sampled in a quantitative or semi-quantitative manner. Collier et al. (2008) compared inorganic substrates, macrophyte and invertebrate communities between four different soft-bottomed streams. The format of data presented here is descriptive and highlights the major differences between streams. More in-depth analysis can be found in the original paper. However, the primary findings of this study are that soft-bottomed streams can be highly variable in terms of macrophyte species present and cover, and in

the invertebrate communities that occupy both the macrophytes and inorganic sediment. Accordingly, in a soft-bottomed stream, whether your sampling is quantitative or semi-quantitative, it is important to estimate or measure the cover of macrophytes and sample the different macrophyte species and other habitat types in any stream under consideration. Collier et al. (1998) also showed that woody debris was an important substrate in soft-bottomed streams, particularly for sensitive EPT taxa. However, there is currently no recommended protocol for quantitative sampling of woody debris in New Zealand, but see Grouns et al. (1999).

## Objectives

- To compare the inorganic substrates, macrophyte and invertebrate communities in four soft-bottom streams in the Waikato.

## Sampling design and methods

### Study area

Streams in the lowlands of Waikato drain some of the most extensively used landscapes in New Zealand. Soils in this region are predominately yellow-brown loams and yellow-brown pumice overlying sedimentary rocks with volcanics and some peat basins. The combination of erodible soils and intensive land use has resulted in a considerable sedimentation of many streams which, due to their low gradient, do not flush sediments during high flow events.

### Habitat assessments

Ten evenly spaced transects were established across reaches. At each transect, size and composition of bed substrates were estimated by classifying 10 particles recorded at evenly spaced points across the transects. Classifications were 'small' wood (< 10 cm diameter), 'large' wood ( $\geq$  10 cm diameter), roots, or inorganic particle size classes: < 2 mm, sand; 2–4 mm, fine gravel; 4–8 mm, small gravel; 8–16 mm, small–medium gravel; 16–32 mm, medium–large gravel; 32–64 mm, large gravel; 64–128 mm, small cobble; 128–256 mm, large cobble; > 256 mm, boulder.

### Invertebrate and macrophyte sampling

Five invertebrate samples from both macrophytes and inorganic substrates were collected at randomly chosen locations in each study reach using a 0.25 mm mesh kick-net. For each inorganic substrate sample an area equivalent to 0.1 m<sup>2</sup> was washed into the net. Grab samples of macrophytes were made into a net held downstream. Each macrophyte sample was washed in a bucket of water to dislodge invertebrates, shaken to remove excess water and divided according to species (of macrophyte), and weighed with a spring balance. All invertebrate samples were picked live on a white tray. When taxa were particularly common and easy to identify a randomly selected quarter was counted and counts were adjusted for total area (of tray). All invertebrates were then stored in 70% isopropyl alcohol and identified under a binocular microscope.

## Results

The Waitoa stream bed was entirely composed of sand and silt, while the Kāniwhaniwha and Waihou stream beds had approximately 50% coverage of silts and sand (Fig. 3). In the Kāniwhaniwha Stream the rest of the bed was covered with woody debris, while the Waihou Stream had woody debris, bedrock and some gravels. The Ōhinemuri Stream had the most diverse bed substrates including woody debris, bedrock, cobbles, gravel and about 15% silt and sand.

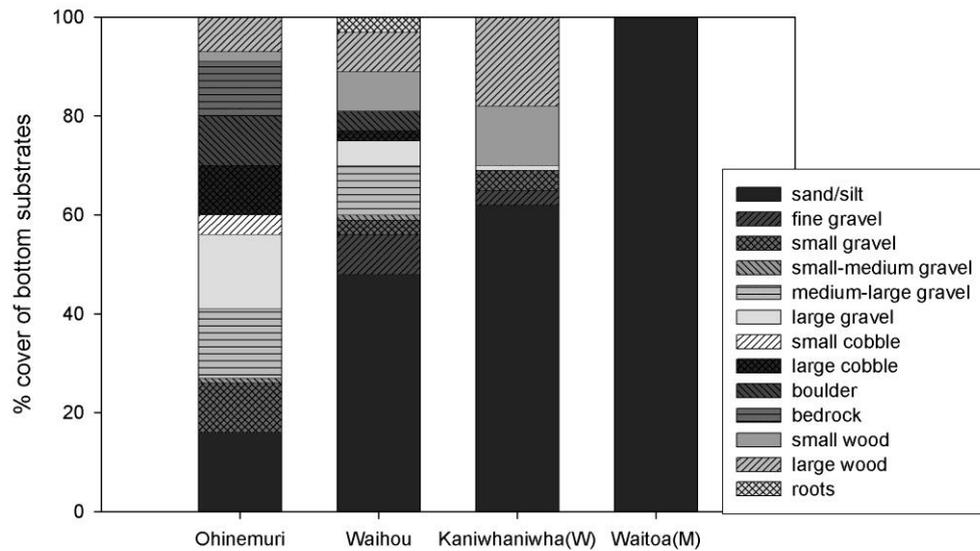


Figure 3. Percentage of bottom substrates classified according to 10 size groups for inorganic material, and large wood ( $\geq 10$  cm diameter), small wood ( $< 10$  cm diameter), or roots.

Macrophyte communities in the streams were quite variable. *Potamogeton* sp. was present at all sites except the Kāniwhaniwha Stream. The Ōhinemuri Stream had the most diverse macrophyte community, possibly due to the diversity of inorganic substrate types found in this stream.

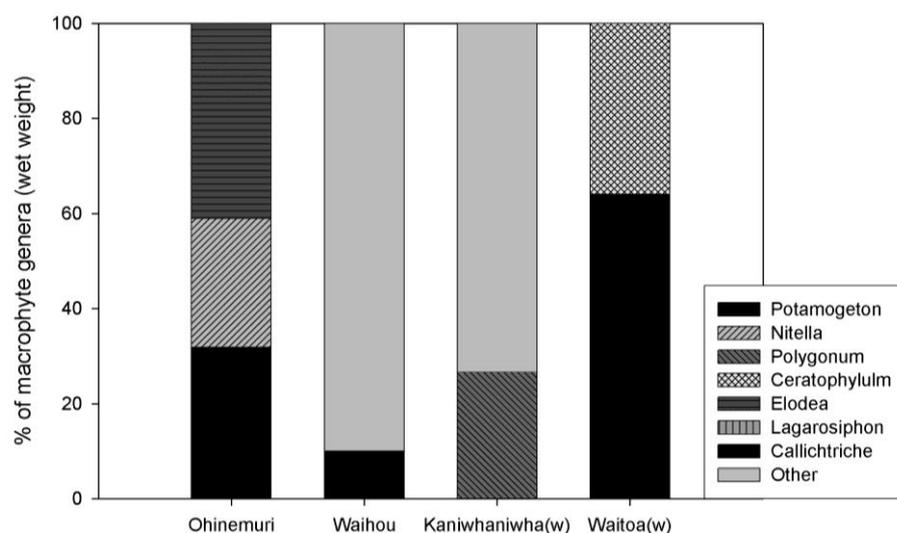


Figure 4. Percentage of different macrophyte genera (wet weight) collected from five locations at four lowland streams in the Waikato.

A total of 106 invertebrate taxa were recorded during this survey. Most were Trichoptera (32 taxa), Diptera (22) and Ephemeroptera (16). Plecoptera, Mollusca and Coleoptera made up 5–7 taxa each. Only the dipteran *Austrosimulium* sp. and the mollusc *Potamopyrgus antipodarum* were found at all sites. An average of 27 taxa were found at each site.

Macroinvertebrate communities on inorganic substrates in the Ōhinemuri, Kāniwhaniwha and Waitoa streams were proportionally dominated by the mollusc *Potamopyrgus antipodarum* (Fig. 5). The Waihou Stream contained *P. antipodarum* but was proportionally dominated by Dipteran, Coleopteran and mayfly taxa. However, in general, Ephemeroptera, Trichoptera and Plecoptera (EPT taxa) were relatively uncommon on inorganic substrates except in the Waihou Stream.

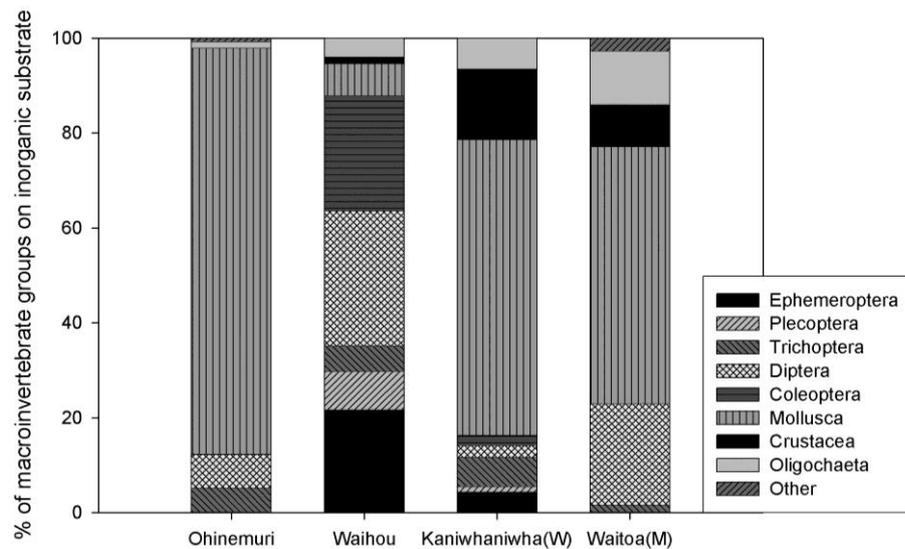


Figure 5. Percentage of different macroinvertebrate groups collected from inorganic substrates at four lowland stream sites.

On macrophytes there were some similarities between invertebrate communities but also some marked differences (Fig. 6). The Ōhinemuri and Kāniwhaniwha stream macrophyte dwelling invertebrate communities were similarly dominated by *P. antipodarum*. However, in the Waitoa Stream, Diptera replaced *P. antipodarum* as the most common taxa. In the Waihou Stream, invertebrates on macrophytes were composed of > 50% EPT taxa, some Diptera, and the ubiquitous snail *P. antipodarum*.

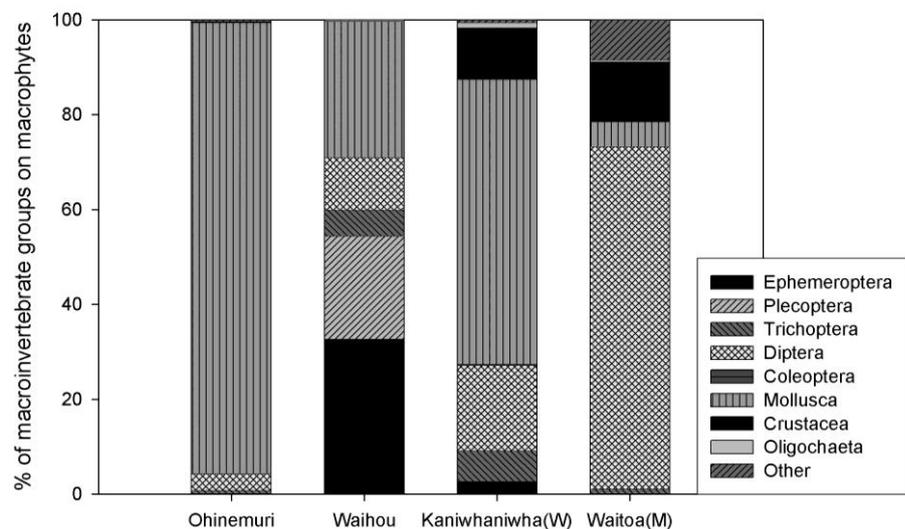


Figure 6. Percentage of different macroinvertebrate groups collected from macrophytes at four lowland stream sites.

Between streams the total abundance of invertebrates on macrophytes and inorganic sediments followed a very similar pattern (Fig. 7). Abundances were greatest in the Ōhinemuri Stream and lowest in the Waihou Stream. It is not appropriate to compare the abundances between macrophytes and inorganic substrates because the sampling methods and units are different.

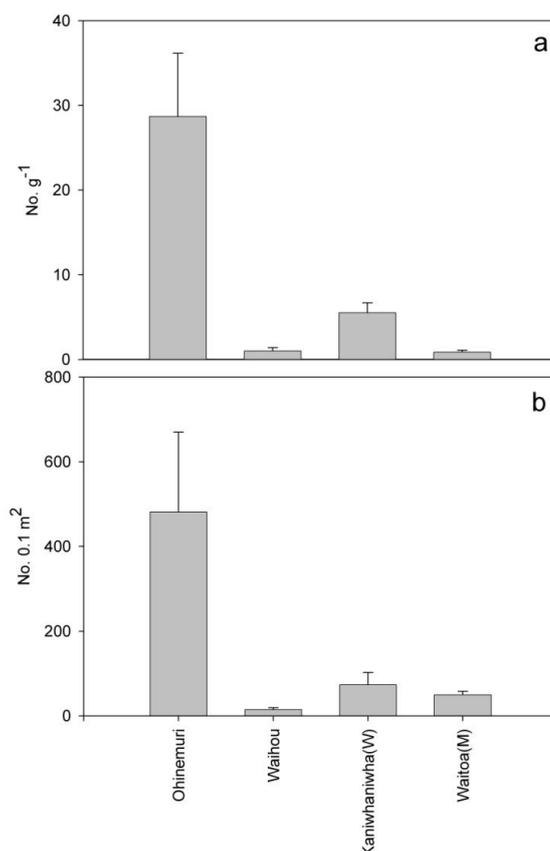


Figure 7. Mean (+ 1 SE,  $n = 5$ ) abundance of total invertebrates from **a**) macrophytes samples (per g wet weight) and **b**) inorganic sediment samples (0.1 m<sup>2</sup>).

## Limitations and points to consider

This case study utilises a method and protocol for sampling both inorganic substrates and macrophytes in soft-bottomed streams that are very similar to ones described in this Toolbox module. All of the graph and presentation techniques described are applicable to both quantitative and semi-quantitative data. However, it is important to acknowledge the limitations of your data. If your sampling is quantitative then more emphasis may be placed on estimates of abundance and your study will be able to detect subtle shifts in community composition. In a semi-quantitative context it is relative, or proportional, abundance and indices of richness which are of primary interest.

## References for case study A

Collier, K.J.; Wilcock, R.J.; Meredith, A.S. 1998: Influence of substrate type and physic-chemical conditions on macroinvertebrate faunas and biotic indices of some lowland Waikato, New Zealand, streams. *New Zealand Journal of Marine and Freshwater Research* 32: 1–19.

Growns, J.E.; King, A.J.; Betts, F.M. 1999: The snag bag: a new method for sampling macroinvertebrate communities on large woody debris. *Hydrobiologia* 405: 67–77.

## Full details of technique and best practice

A complete and detailed guide to this technique can be found in Stark et al. (2001).

Protocol:

1. Ensure that the sampling net is clean.
2. Approach sample site by moving upstream through the waterway. Determine plant species to be sampled. Consistency in plant species is important for comparisons between sites, although not always possible. Standardise the depth/velocity conditions of sampling points, where possible.
3. Collect replicate samples ( $n \geq 4$ ) of submerged macrophyte tips (approx. 100 g wet weight of top 20–30 cm of plant, which is equivalent to 1.5–2 L of weed) by moving net upstream into macrophyte bed and breaking off required portion of plant material. Place each replicate sample in a separate bucket. Rinse net thoroughly between replicates.
4. Add approx. 1 L of clean water to each bucket and firmly attach lid. Shake bucket vigorously (20x) to detach invertebrates from macrophyte material.
5. Pour dislodged macroinvertebrates and detritus through a 0.5 mm sieve. Rinse each sample twice more in a similar manner.
6. With the aid of a wash bottle, transfer material retained on the sieve to a plastic container.
7. Add preservative. Aim for a preservative concentration in the sample container of 70–80% (i.e. allow for the water already present). Be generous.
8. Place a sticky label on the side of the sample container and record the site code/name, date, and replicate number (if applicable) using a permanent marker. Write on the label when it is dry and do not rely on a label on the pottle lid! Place a waterproof label inside the container. Screw the lid on tightly.
9. Note the sample type, collector's name and preservative used on the field data sheet.
10. Drain the plant material of excess water (leave to stand in sieve for 2 minutes) and then weigh to the nearest 5 g using a spring balance. If greater precision is required place plant samples in labelled plastic bags and return to laboratory for drying (70°C for at least 24 hrs) and weighing.

11. Record wet weight of macrophyte material associated with each replicate sample. Also record the species and condition (i.e. senescent, flowering, covered in epiphytes) for the macrophyte bed from which the sample was taken.

## References and further reading

- Boothroyd, I.K.G.; Stark, J.D. 2000: Use of invertebrates in monitoring. Pp. 344–373 in Collier, K.J.; Winterbourn, M.J. (Eds): *New Zealand stream invertebrates: ecology and implications for management*. New Zealand Limnological Society, Christchurch.
- Growns, J.E.; King, A.J.; Betts, F.M. 1999: The snag bag: a new method for sampling macroinvertebrate communities on large woody debris. *Hydrobiologia* 405: 67–77.
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<http://www.cawthron.org.nz/coastal-freshwater-resources/downloads/stream-habitat-assessment-protocols.pdf>
- Magurran, A.E. 2004: *Measuring biological diversity*. Wiley-Blackwell, London. 260 p.
- Stark, J.D. 1985: A macroinvertebrate community index of water quality for stony streams. *Water & Soil Miscellaneous Publication 87*. National Water and Soil Conservation Authority, Wellington.
- Stark, J.D. 1998: SQMCI: a biotic index for freshwater macroinvertebrate coded abundance data. *New Zealand Journal of Marine and Freshwater Research* 32: 55–66.
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<http://www.cawthron.org.nz/coastal-freshwater-resources/downloads/protocols-full-manual.pdf>
- Stark, J.D.; Maxted, J.R. 2007: A biotic index for New Zealand's soft-bottomed streams. *New Zealand Journal of Marine and Freshwater Research* 41: 43–61.

## Appendix A

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

docdm-724991	Introduction to macroinvertebrate monitoring in freshwater ecosystems
docdm-146272	Standard inventory and monitoring project plan
docdm-761873	Stream habitat assessment field sheet
docdm-761858	Stream invertebrate data sheet example