A new substrate for sampling deep river macroinvertebrates

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Abstract

We compared macroinvertebrate communities colonising multiplate samplers constructed from perspex or tempered hardboard (wood) with an alternative artificial substrate constructed from folded coconut fibre matting (coir) enclosed in nylon netting. Substrates were incubated for 62 days over January to March 2007 at six sites over 240 km along the Waikato River. The three substrates supported similar numbers of invertebrate taxa (27 - 29 taxa), but coir samples contained 71% of total invertebrate numbers from all substrates combined, compared with <17% for each type of multiplate sampler. Coir faunas were heavily dominated by the hydrobiid snail *Potamopyrgus* (84 % of numbers), and this taxon along with the amphipod *Paracalliope* comprised 58 - 66 % of invertebrates on both types of multiplate samplers. Analysis of a Bray-Curtis matrix suggested statistically significant differences in percent community composition between coir samplers and each type of multiplate sampler over the late summer study period. Densities per cm^3 of Oligochaeta, Mollusca, and "other worms" (Platyhelminthes, Rhabdocoela, Nemertea and Hirudinea combined) were significantly higher in coir samples than one or both of the multiplate samplers. Results suggest coir samplers may provide a useful supplement to multiplate samplers for deep river invertebrate studies by collecting a different range of taxa, including those favouring cover and characteristic of depositional environments.

Keywords: Hester-Dendy — multiplate sampler — artificial substrate — coir — large river — Waikato — New Zealand.

Introduction

Sampling macroinvertebrates in large rivers is hampered by the physical difficulties and dangers associated with accessing deep-flowing water, and the complexity of habitats that occur within them. Such environments can be characterised by distinct species assemblages (Bournard *et al.* 1998; deDrago *et al.* 2004; Strayer *et aL* 2006), and in New Zealand at least these

communities are poorly-studied (see Collier & Lill 2008). In New Zealand and internationally, there is increasing interest from resource managers and society in the ecological assessment and rehabilitation of large rivers, with recognition that management efforts require an improved understanding of temporal dynamics and spatial patterns of their biological communities (e.g., Schweiger *et at* 2005; Flotemersch *et al.* 2006).

Bioassessment studies of non-wadeable streams and rivers are increasing in number, but methods to monitor macroinvertebrates in these systems are not as well-advanced as for wadeable streams (Blocksom & Flotemersch 2005). A wide range of approaches has been used to sample macroinvertebrate faunas in deep-water habitats, including dredges (Bournard *et al.* 1998), grab (Thorp 1992), core (Boubee 1977), or air-lift (Carter 2000; Neale *et al.* 2006) samplers, as well as artificial substrates (see Merritt *et al.* 1984). Artificial substrates have varied in construction, including those made of hardboard plates (e.g., Hester & Dendy 1962; Hall 1982), or inorganic substrates such as cement, glass or porcelain spheres (De Pauw *et al.* 1993; Hall 1982; Pashkevich et al. 1996; Roby et al. 1978), concrete "cupcakes" or mattress blocks (Troelstrup & Hergenrader 1990; Way *etal.* 1995), rock baskets (Rabeni & Gibbs 1978; Courtemanch 1984), and "conservation webbing" (Voshell & Simmons 1977). Vegetation mimics have also been used; Suren (1991) used artificial moss analogues to distinguish habitat effects of bryophytes in alpine streams, whereas Linklater (1995) and Collier *et al.* (2006) employed plastic strips as artificial substrates to separate physical and trophic effects of entrained leaves.

Artificial substrates offer the advantage of eliminating the influence of substrate variability on macroinvertebrate faunas (Boothroyd & Stark 2000), and enable quantitative sampling or comparisons of a standard-sized sample when this is more important than obtaining a comprehensive species list (Boothroyd & Dickie 1989). Although multiplate samplers are typically only colonised by a subset of the species present at a particular site (Boothroyd & Stark 2000), they may support a more diverse and even range of invertebrates than macrophytes in lowland streams (Collier 2004). When comparing different types of samplers in a range of riverbank habitats, Blocksom & Flotemersch (2005) reported that faunal composition on multiplate samplers effectively represented the prevailing abiotic conditions even though macroinvertebrate metrics derived from multiplate and net samples differed greatly. Death (2000) concluded that artificial substrates are a powerful biomonitoring tool to examine substrate-invertebrate interactions experimentally, and should be used more often in studies of New Zealand streams.

One possible approach to increasing the range of species collected by artificial substrates is to develop alternative substrate types that offer contrasting microhabitat conditions. With this in mind, we evaluated the performance of folded coconut-fibre matting (coir) enclosed in nylon netting, compared to more conventional multiplate samplers modified from the design of Hester & Dendy (1962). We hypothesised that the complex mesh of natural fibres making up coir matting would attract a different suite of species by providing a greater variety of spaces, cover and feeding surfaces, and a more depositional environment than traditional plates. We deployed coir and multiplate samplers constructed from perspex or tempered hardboard (wood) in triplicate at six sites down the Waikato

River over summer 2007, and compared invertebrate communities colonising these different substrates. Thus we were able to compare organic and inorganic multiplate samplers, and organic samplers made up of multiple wooden plates and folded coconut fibre.

Methods

Sampling sites

Substrates were deployed at six sites on the Waikato River, North Island, between Huka Falls and Rangariri, covering a river distance of around 240 km and corresponding to the same sites used by Davenport (1981). The Huka site (2778918E, 6278661N) was above the falls on the true right side in a backwater area where there was some circulation of water along a steep bank. The river was deep at this site (> 5 m) and clarity was high with luxuriant growths of macrophytes visible. Riparian vegetation consisted of native species dominated *by* manuka *(Leptoipermum scoparium).* The Ohakuri site (2779481E, 6306108N) was c. 350 m below Ohakuri Dam in a backwater area on the true right with

circulating water fed by the fast-flowing main channel. Depth was around 2-3 m and riparian vegetation comprised mainly native species, although willows *(Salix* sp.) on the river edge grew out over the water and provided convenient anchoring sites for substrate deployment. Bottom materials appeared to be mainly cobbles. The Narrows site (2716817E, 6370859N) was above Hamilton City in a gorge section of the river dominated on the true right bank by riparian willows and poplars with roots that extended into the river. Depth at the river edge was around 1-2 m. The Horotiu site (2704864E, 6387048N) comprised a shallow shelf that extended out into the river from the true right bank with riparian poplar trees. The Ngaruawahia site (2699295F, 6391545N) was on the true left bank c, 350 m below the confluence with Wa5pa River which dominated the now and increased turbidity at this site. Riparian willows grew into the channel and water depth was around 2 m. The most downstream site was below the Rangariri Bridge (2698676E, 6416834N) on the true right bank where rank pasta grass dominated riparian vegetation along with
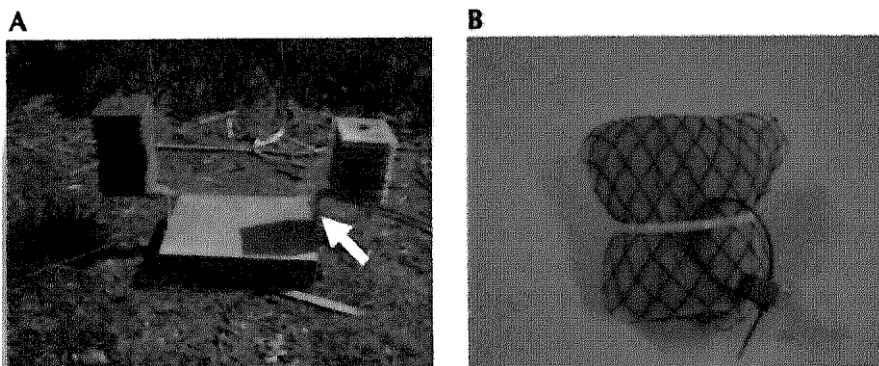

Plate 1. A. Multiplate samplers constructed from hardboard (left) and perspex (right) showing position of samplers in relation to the concrete paver. Arrow indicates attachment point for the coir sampler. B. Coir sampler viewed from bottom showing the weight on a plastic tie, the black coarse mesh enveloping the folded coir mat, and the 0.5 mm mesh which covers the lower half of the coir sampler to prevent loss of macroinvertebrates during removal.

the occasional willow tree. The substrate was mainly sand with abundant growths of aquatic macrophytes dominated by *Ceratophyllum demersum.*

Artificial substrates

Both multiplate samplers consisted of 15 square plates measuring 7.6 x 7.6 cm secured through the middle on a stainless steel rod with variable spacing between plates of 3 to 6 mm (Plate 1A). The perspex plates were 2-3 mm thick and had their upper and lower surfaces roughened. The wooden plates were made of 4 mm tempered hardboard that had been soaked in water until any colour from leaching was not visible. The plates on each sampler provided a colonisable area (including edges) of 0.18 m^2 (perspex) or 0.19 m^2 (wood), the difference reflecting the greater thickness of hardboard plates. In volumetric terms the multiplate samplers occupied 578 and 751 cm^3, respectively, including the spaces between plates.

A coir sampler (Plate 1B) comprised a 30 x 25 cm piece of matting comprising a woven mesh of multiple coconut fibres about 0.3 mm in diameter made up of 40-45 % lignin, 32-43 % cellulose, 3-4 % pectin and 1 % ash (Lekha 2004). The coir was folded into a 14 x 10 cm rectangular shape, and placed on a 15 x 15 cm square of 0.5 mm mesh nylon netting intended to prevent loss of macroinvertebrates as water drained through the bottom during retrieval. The coir and drainage netting were enveloped in 20 mm mesh netting and secured around the middle with a plastic tie. A 30 g stainless steel weight was attached beneath the drainage netting to ensure the sampler was correctly oriented when retrieved. In volumetric terms a coir sampler typically occupied 700 cm^3.

Deployment and retrieval

The two types of multiplate samplers were deployed in pairs on metal pegs bolted to the ends of concrete pavers (23 x 16 x 4 cm). The pegs elevated the samplers 6 cm above the pavers (Plate 1A); one coir sampler was tied through one of the pegs on each paver. Three sets of samplers were deployed at each site by tying a rope through the pegs on either end of each brick so that they could be lowered with the multiplate samplers oriented upwards, and retrieved with minimal disturbance. Substrates were deployed where water was flowing, and were at or near the bottom except at the Huka site where the water was very deep and the bricks rested on a submerged shelf. Deployment depths varied depending on the characteristics of the site and ranged from <1 m at Horotiu to around 5 m at Huka. Deployments spanned a 62-day period from 18-19 January to 21-22 March 2007.

On retrieval, the pavers and attached samplers were gently pulled out of the water using the ropes that had been tied off to nearby vegetation. A total of 15 coir, 14 wooden plate, and 14 perspex plate samplers were retrieved with minimal disturbance. Triangular hand-nets (0.5 mm mesh) were placed around each substrate before they were removed from the water to capture any invertebrates dislodged during transfer to land. Any material caught in the net along with the intact substrates were placed in ice-cream containers with a little river water, and then transported on ice to the laboratory.

Macroinvertebrate sample processing and analysis

In the laboratory, multiplate samplers were disassembled and individual plates were lightly rubbed and rinsed to dislodge

any invertebrates. Coir samples were unfolded and repeatedly washed until the elutriate appeared clean. All material from each sampler was passed through a 0.5 mm sieve and stored in 70 % isopropynol until invertebrates could be picked out on a white tray for identification. Level of taxonomic resolution was mostly to genus for Insecta, Crustacea and Mollusca, whereas most other taxa were identified to family.

Non-metric multidimensional scaling (NMDS) (Primer 6.1.2) was conducted on a Bray-Curtis similarity matrix of percent abundance invertebrate data (square-root transformed). This was followed by a two-way crossed Analysis of Similarities (ANOSIM) to investigate differences among sites and substrates. SIMPER analysis was also conducted using Bray-Curtis similarity of percent abundance data excluding the Ohakuri and Huka sites which were heavily dominated by one taxon and differed significantly in composition from the lower river sites (see Results). Numbers of major invertebrate groups and total invertebrates were standardised by the volume of each sampler type (see above), and compared using Analysis of Variance (Systat v. 11) on ranked data (Conover & Iman 1981), with substrate type as a factor and site as a covariate since sites were fixed and all substrates were deployed at all sites. Pairwise comparisons among substrate types were made using Tukey test.

Results

Taxonomic richness

A total of 38 macroinvertebrate taxa was found on all substrates deployed at the six sites over summer (Appendix I). Of these taxa, 29 were found in coir samples, 29 on perspex substrates, and 27 on wood substrates. Coir samples were colonised by 5 taxa not found on other samplers, compared to 5 and 2 taxa not found in coir samples but present on perspex or wood plates, respectively (Appendix I). Acarina and *Paratya* were found on both multiplate samplers but were absent from coir samples. Taxonomic richness averaged 5.6 taxa in Ohakuri and Huka samples, compared to 8.8 taxa for lower river samples (all substrates and sites combined).
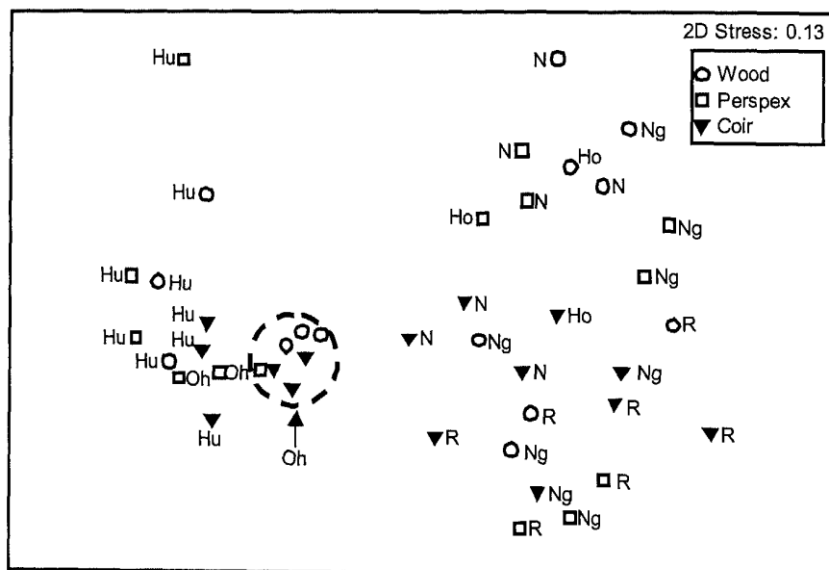
Community composition

Coir faunas were heavily dominated by the hydrobiid snail *Potamopyrgus antipodarum* (84 %), which occurred mostly at Ohakuri where 4,451 were found in one sample. This taxon along with the amphipod *Paracalliope* dominated numbers on both multiplate samplers (58-66 % of total numbers). Tanytarsini midge larvae were relatively abundant on perspex samplers (19 % of numbers cf < 4 % on wooden plates or coir samples). The hydropsychid caddisfly *Aoteapsyche* was also relatively common on multiplate samplers, in particular the wooden plates where it comprised > 19 % of total numbers compared to 6 % on the perspex plates and 0.1 % in the coir samples. In contrast, platyhelminth flatworms were relatively abundant on perspex samplers (> 10 % of numbers cf < 4 % on wooden plates or coir samples).

The NMDS analysis based on percent abundance data had a stress value of 0.13 indicating a good representation of the data in two dimensions (Figure 1). The coir samples showed a similar spread to the multiplate samples along axis 1 of the ordination, but a narrower spread along axis 2 (Figure 1). ANOSIM indicated significant differences among substrates (Global R = 033, P = 0.001). Pairwise comparisons using ANOSIM indicated wooden and perspex multiplate samples

were similar overall in terms of community composition, whereas both types of multiplate samples were significantly different to coir samples. ANOSIM also indicated significant differences among sites (Global R = 0.75, P < 0.01). The upper river sites, Huka and Ohakuri, separated from the downstream sites along Axis 1 irrespective of substrate type. Indeed pairwise comparisons indicated significant differences among all sites, except for Horotiu which was statistically similar to Narrows, Ngaruawahia, and Rangariri in terms of community composition in late summer. A NMDS of presence-absence data provided a poor representation of the data in two dimensions *(stress* = 0.22; graph not shown), although ANOSIM indicated similar differences observed for the percent abundance analysis.

SIMPER analysis, excluding the upper Ohakuri and Huka sites, indicated comparable community similarity within wood, perspex, and coir samples (average Bray-Curtis similarity = 44.9, 46.5 and 52.8, respectively).

Dissimilarity levels between substrate types ranged from 51.8 to 53.7 for paired substrate type comparisons from the lower river. *Aoteapsyche* and *Cura* made the greatest contributions to dissimilarity between the two multiplate samplers, with relative abundances being higher on wood and perspex, respectively. Relative abundances were similar on both these substrate types for other taxa contributing at least 5 % to the dissimilarity, although Tanytarsini and Dalyellidae were relatively more common on perspex (Table 1). Coir samples were distinguished from both perspex and wood samples in the SIMPER analysis by lower relative abundances of *Aoteapsyche* and higher percentages of indeterminate Nemertea taxa, *Potamopyrgus* and *Paracalliope* (Table 1). *Cura* was relatively more common on coir compared to wood, but the reverse was observed when coir samplers were compared to perspex. Coir

Figure 1. Non-metric multidimensional scaling plot of invertebrate community composition (percent abundance) on three substrate types deployed at six sites along Waikato River. Hu, Huka; Oh, Ohakuri; N; Narrows; Ho, Horotiu; Ng, Ngaruawahia; R, Rangariri.

Table 1. Results of a SIMPER analysis with taxa ordered by their contribution to average, dissimilarity (Av.Diss.) comparing substrate types for taxa contributing > 5 %to the dissimilarity Indet. = Indeterminate.

A. Wood vs. Perspex	Wood %	Perspex %	Av.Diss.	% Contribution	Cumulative %
Aoteapsyche	3.48	2.63	6.84	13.19	13.19
Cura	2.05	3.80	6.68	12.90	26.09
Nemertea indet.	1.65	1.85	4.52	8.72	34.81
Paracalliope	5.65	5.33	4.19	8.08	42.89
Potamopyrgus	2.68	2.33	3.85	7.44	50.33
Tanytarsini	0.74	1.57	3.66	7.07	57.39
Dalyellidae	0.56	1.08	2.62	5.05	62.45
B. Wood vs. Coir	**Wood %**	**Coir %**	**Av.Diss.**	**% Contribution**	**Cumulative %**
Aoteapsyche	3.48	0.19	7.61	14.17	14.17
Cura	2.05	3.05	5.75	10.72	24.89
Potamopyrgus	2.68	3.71	5.36	9.98	34.87
Nemertea indet.	1.65	3.23	5.31	9.89	44.76
Paracalliope	5.65	5.94	4.04	7.52	52.28
C. Perspex vs. Coir	**Perspex %**	**Coir %**	**Av.Diss.**	**% Contribution**	**Cumulative %**
Cura	3.8	3.05	6.17	11.74	11.74
Nemertea indet.	1.85	3.23	5.67	10.79	22.53
Aoteapsyche	2.63	0.19	5.38	10.23	32.76
Potamopyrgus	2.33	3.71	5.16	9.82	42.58
Paracalliope	5.33	5.94	4.38	8.33	50.92
Tanytarsini	1.57	0.74	3.57	6.79	57.71
Dalyellidae	1.08	0.75	2.64	5.02	62,73

samples supported fewer Tanytarsini and Dalyellidae than perspex samples (Table 1). The oligochaetes *Branchiura,* Naididae and indeterminate taxa were among those contributing to 90 % of the dissimilarity between wood and coir samples, as was *Branchiura* for perspex and coir samples, but these taxa contributed < 2 % each to the dissimilarity between substrate types.

Density

Coir samples contained 71 % of total invertebrate numbers over all substrates combined, compared with 13 % and 16 % for the perspex and wooden multiplate samples, respectively. Mean densities adjusted for sampler volume were higher for the main insect groups in wood samples and higher for total invertebrate numbers in coir samples,

but these differences were not statistically significant (Figure 2). Similarly there was no significant difference among substrates for densities of Crustacea which was heavily dominated by *Paracalliope*. In contrast, coir samplers supported significantly higher densities of Mollusca (mainly *Potamopyrgus)* than the wood samples, and more Oligochaeta and "other worms" (Platyhelminthes, Rhabdocoela, Nemertea and Hirudinea combined) than either multiplate sampler.

Discussion

All types of artificial, substrates tested in this study were colonised by similar numbers of macroinvertebrate taxa, but the composition of communities and the abundances of certain groups

differed. The taxonomic richness found in multiplate and coir samples over the summer period (27-29 taxa) was close to the 31 taxa found on similar perspex multiplate samplers deployed over August-November in the Ohinemuri River, Waikato, by Boothroyd & Dickie (1987). Davenport (1981) reported 38 taxa from multiplate substrates deployed at the same sites in the Waikato River over one year, with most taxa being found at the four lower river sites (17-28 per site). Davenport found no pattern with distance downstream, although he did observe higher diversity in the lower river in general compared with two sites in the upper Waikato River.

This upper-lower river separation was also evident in the present study, both in terms of community composition and taxa richness.

Differences in community composition observed in the ordination analysis suggested that sampler configuration had a greater effect on the colonising fauna than whether substrates were constructed from organic or inorganic materials. The same five taxa *(Aoteapsyche, Cura, Potamopyrgus,* indeterminate Nemertea and *Paracalliope)* accounted for over 50 % of the dissimilarity between coir and either type of multiplate sampler, with *Aoteapsyche* clearly favouring multiplate samplers over coir, and wood over perspex.
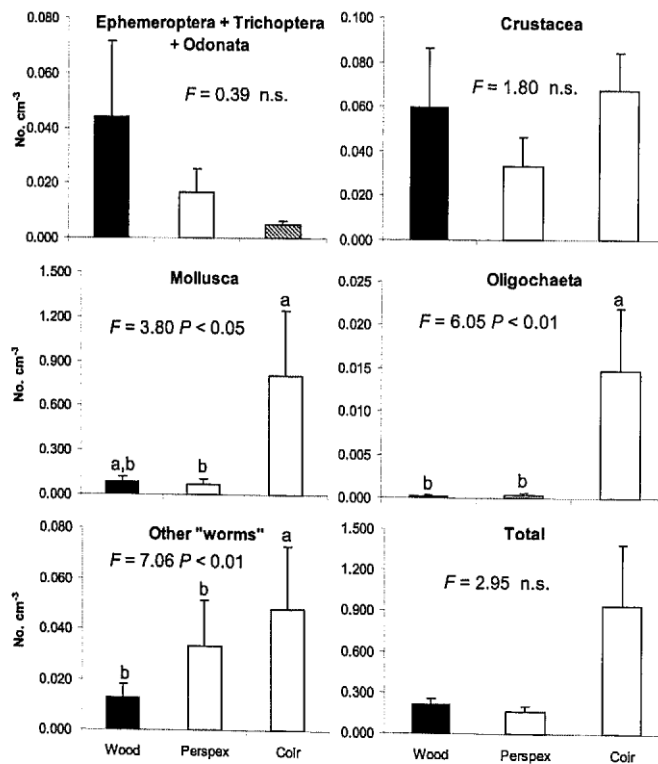

Figure 2. Density (mean + 1 SE adjusted for cm' of sampler including interstitial spaces) of major invertebrate groups and total invertebrates on wood and perspex multiplate samplers and coir samplers deployed at six sites along Waikato River over summer 2007. F values indicate significance of substrate as a factor in an Analysis of Variance using site as a covariate. For significant effects, bars with the same letter above are not significantly different (Tukey test).

It is possible that variations in surface roughness providing attachment sites for hydropsychiid nets may have affected the abundance of this taxon. In support of this hypothesis, Way *et al.* (1995) reported marked effects of smooth versus grooved surfaces on concrete artificial substrates, with hydropsychiid caddisflies preferring grooved substrates for retreat and net construction. In contrast, *Cura* occurred in higher relative abundances on perspex plates than on wood plates or coir, suggesting an avoidance of organic material by flatworms. This pattern may partly reflect the suitability of flat surfaces for mobility, coupled with the possibility that toxic materials were still being released from the hardboard and may have influenced colonisation by taxa that have a high proportion of their body area in direct contact with substrate surfaces. Leaching of hardboard plates prior to introduction was intended to minimise any effect of this, and the overall similarity in the NMDS between perspex and wood multiplate samplers suggests any influence of continued leaching was negligible at the community level.

It is unclear why *Potamopyrgus* numbers were so high in coir samples compared to plates which might be expected to provide a more suitable surface for colonisation by algae on which snails often graze. However, *Potamopyrgus* may also rasp surfaces of organic material such as submerged leaves, presumably ingesting microflora associated with organic matter decomposition (Collier & Winterbourn 1986), and they can also be common on wood and macrophytes (Death 2000). In addition, it may be that the coir samplers provided a refuge from predation or that the sediment associated with these samplers contained trace nutrients important for snail nutrition (Broekhuizen *et al.* 2001). We observed that the mesh of the coir matting entrained large amounts of fine sediment, which may have contributed to the significantly higher densities of oligochaetes and other non-insect taxa that are otherwise not commonly encountered (e.g., Nemertea and Rhabdocoela). Taxa colonising coir samplers deployed elsewhere have included the crayfish *Paranephrops planifrons* (KJC & MH unpubl. data) suggesting this substrate configuration may also be suitable for taxa that prefer cover (Parkyn 2000).

In summary, the present study suggests that macroinvertebrate communities colonising coir matting differed in composition and abundance of some groups from those colonising multiplate samplers over late summer in the Waikato River. This difference may reflect the higher habitat complexity of coir samplers providing cover for some taxa, and the depositional environment within the fibre matting. Coir samplers may therefore be a useful adjunct to multiplate samplers where the aim is to document the invertebrate biodiversity present in deep-water environments.

Acknowledgements

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Appendix I. Numbers of macroinvertebrate taxa found on three substrate types at six Waikato River sites combined. +, found only in coir samples. X, present but unable to enumerate.

	Wood N = 14	Perspex N = 14	Coir N = 15
Ephemeroptera			
Zephlebia			
Trichoptera			
Aoteapsyche	425	115	7
Oxyethira	24	20	14
Paroxyethira	6	7	17
Polyplectropus		1	
Triplectides +			2
Odonata			
Hemicordulia +			1
Xanthocnemis	8	9	11
Hemiptera			
Microvelia		1	
Diptera			
Chironomini	3	1	
Orthocladiinae	4	5	5
Tanytarsini	51	343	12
Crustacea			
Amarinus +			3
Calanoida			4
Ostracoda +			2
Paracalliope	587	579	688
Paratya	1	2	
Phreatogammarus	37	4	8

	Wood	Perspex	Coir
Tanaidacea	3		3
Acarina	1	3	
Mollusca			
Ferrissia	13	7	7
Gyraulus	4	1	10
Latia		1	
Physa	28	11	134
Planorbella		1	
Potamopyrgus	882	571	8,271
Pseudosuccinea		2	
Oligochaeta			
Branchiura			9
Enchytraeidae		1	127
Naididae	1	4	2
Indeterminate	1	1	17
Platyhelminthes			
Cura	55	195	338
Rhabdocoela			
Dalyellidae	12	19	8
Nemertea			
Prostoma	26		7
Indeterminate	34	49	139
Hirudinea			
Clossiphonlidae	5	7	11
Cnidaria			
Chlorohydra	4	3	1
Bryozoa	X	X	X
Total numbers	2,254	2,023	9,910