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Effects of Development on Zero-order Streams in the Waikato Region

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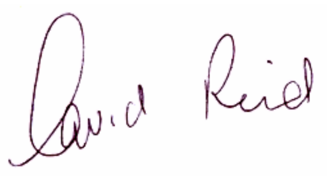

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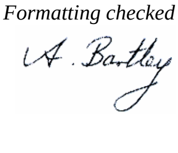

Executive Summary

Headwater streams are the smallest stream channels in a stream network. They are often referred to as "zero-order", rather than first-order, streams because they typically do not appear on 1:50 000 topographic maps. Zero-order streams account for a high proportion of the total channel length in a stream network, and represent the closest association between the terrestrial and aquatic environments in that network. Studies in New Zealand and overseas have shown that zero-order streams often have unique habitat conditions supporting a distinctive aquatic invertebrate community, and the ecological functions they perform may have a strong influence on the health of the wider stream network. Thus, the first aim of the present study was to determine some of the biological values and ecological functions of zero-order streams compared to those of larger first-order streams, and to quantify the effects of land use on those values and functions. Because they are small, numerous and easy to pipe or infill, zero-order streams are particularly vulnerable to the effects of development. Therefore the second aim of our study was to design a decision support framework for quantifying compensation if land development requires the loss of a zero-order stream by piping or degradation by a change in land use.

The study included sites in four levels of land-use stress – native forest catchments (lowest stress), pasture catchments with thick riparian forest surrounding the stream, pasture catchments with thin riparian forest, and open pasture with no riparian forest (highest stress). In each land-use type, three sites were sampled in a perennially-flowing zero-order habitat and three in a perennially-flowing first-order habitat downstream. The study was replicated in Pokeno, representing low topographic relief country, and in Whatawhata, representing steep hill country. At each site benthic macro-invertebrates were collected and the ecological functions of a 50 m reach were assessed using the non-biological functions of the Stream Ecological Valuation (SEV).

Results showed that macro-invertebrate communities were as diverse, or nearly as diverse, in perennial zero-order streams as in first-order streams, though in steep country, EPT (Ephemeroptera, Plecoptera and Trichoptera) richness was significantly lower in zero-order streams. All major taxonomic groups were well represented in zero-order streams. We did not find a consistent difference in community composition between flowing zero-order and first-order streams. Though some taxa were found only in zero-order streams, they occurred rarely and we cannot discount the possibility that further sampling in first-order streams may reveal their presence there. This result is consistent with a previous study of Waikato headwater (zero-order) streams (Parkyn et al. 2006b) which found that among three types of headwater habitat (wet mud, isolated pools and flowing water), only isolated pools and wet mud habitat had significantly different invertebrate communities to nearby higher-order stream habitats. It appears that a significant change in stream invertebrate communities occurs at the point where flow ceases and the aquatic habitat is reduced to pools or mud. Our study, being conducted in perennially-flowing zero-order streams, did not capture this difference. We did not find any taxa in zero-order streams that are unknown in first-order streams. However, since many taxa of

New Zealand aquatic invertebrates are not described beyond genus level, we cannot discount the possibility that there may be some unique species in zero-order streams.

We found no difference in the ability of flowing zero-order vs. first-order streams to perform the non-biological ecological functions of the SEV.

Land-use stress was a significant predictor of macro-invertebrate diversity, explaining 19% and 31% of variability in Shannon diversity at Pokeno and Whatawhata respectively, 10% and 52% of variability in total richness at Pokeno and Whatawhata respectively, and 21% and 50% of variability in EPT (Ephemeroptera, Plecoptera and Trichoptera) richness at Pokeno and Whatawhata respectively. The following table summarises the biological values that would be lost if a typical zero-order stream in a particular land use and topographic type were piped or infilled. It also indicates the change in biological values if a stream catchment or riparian zone were converted from one land use to another (abbreviations: NF native forest; RF Thick wide buffer of riparian forest with dense understorey; RF Thin narrow buffer or riparian trees with little or no understorey; P pasture):

	Lowland				**Steep hill country**			
	NF	**RF Thick**	**RF Thin**	**P**	**NF**	**RF Thick**	**RF Thin**	**P**
Total richness	20	21	27.3	15	26.3	21.7	22.7	11.7
±std deviation	±1	±2.6	±4.6	±1	±5.5	±6.0	±1.5	±0.58
EPT richness	4.7	3.7	5.3	2.0	8.0	6.0	4.3	1.0
±std deviation	±1.2	±0.6	±2.51	±1	±1	±4.6	±1.2	±1
Shannon diversity	2.03	2.11	2.43	1.64	2.08	1.98	1.87	1.15
±std deviation	±0.30	±0.35	±0.20	±0.58	±0.59	±0.48	±0.21	±0.25

Land-use stress was also strongly correlated with non-biological SEV (nb-SEV) mean score for both zero-order and first-order sites, explaining 41% of variability in nb-SEV score in low-topography country and 76% of variability in steep country. The following table summarises the non-biological ecological functions that would be lost if a typical zero-order stream were piped or infilled, and the changes that would occur if a stream catchment were converted from one land use to another (abbreviations are: NFR natural flow regime, CFP connectivity to floodplain, CSM connectivity for species migrations, CGW connectivity to groundwater, WTC water temperature control, DOM dissolved oxygen maintenance, OMI organic matter input, IPR instream particle retention, DOP decontamination of pollutants, FPR floodplain particle retention, FSH fish spawning habitat, HAF habitat for aquatic fauna, FFI fish fauna intact, IFI invertebrate fauna intact, ABI aquatic biodiversity intact, RVI riparian vegetation intact):

	Lowland				Steep hill country			
SEV function	**NF**	**RF Thick**	**RF Thin**	**P**	**NF**	**RF Thick**	**RF Thin**	**P**
NFR	0.77	1.00	0.90	0.90	0.75	0.90	0.65	0.72
± std deviation	±0.14	±0	±0.09	±0.09	±0.17	±0.09	±0.17	±0.25
CFP	0.45	0.40	0.30	0.82	0.43	0.55	0.37	0.62
± std deviation	±0.23	±0	±0.09	±0.2	±0.25	±0.15	±0.2	±0.19
CGW	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
± std deviation	±0	±0	±0	±0	±0	±0	±0	±0
WTC	0.76	0.78	0.71	0.46	0.78	0.76	0.68	0.56
± std deviation	±0.04	±0.02	±0.02	±0.1	±0.01	±0.01	±0.09	±0.12
DOM	1.00	0.38	0.74	0.37	1.00	0.83	0.79	0.8
± std deviation	±0	±0.21	±0.31	±0.1	±0	±0.3	±0.36	±0.35
OMI	0.84	0.88	0.67	0.06	0.92	0.83	0.48	0.24
± std deviation	±0.16	±0.11	±0.07	±0.1	±0.03	±0.04	±0.29	±0.25
IPR	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
± std deviation	±0	±0	±0	±0	±0	±0	±0	±0
DOP	0.89	0.9	1.00	0.79	0.93	0.70	0.79	0.69
± std deviation	±0.1	±0.05	±0	±0.15	±0.06	±0.12	±0.22	±0.29
FPR	0.54	0.41	0.29	0.64	0.39	0.58	0.31	0.45
± std deviation	±0.22	±0.09	±0.07	±0.05	±0.12	±0.11	±0.18	±0.11
FSH	0.35	0.31	0.40	0.56	0.52	0.50	0.35	0.44
± std deviation	±0.23	±0.17	±0.2	±0.34	±0.02	±0.06	±0.19	±0.18
HAF	0.97	0.76	0.70	0.49	0.99	0.85	0.67	0.53
± std deviation	±0.04	±0.04	±0.13	±0.09	±0.01	±0.14	±0.11	±0.08
RVI	0.96	0.86	0.54	0.36	0.98	0.84	0.47	0.42
± std deviation	±0.08	±0.13	±0.05	±0.01	±0.04	±0.11	±0.10	±0.10
mean nb-SEV score	0.79	0.72	0.69	0.62	0.81	0.78	0.63	0.62
±std deviation	±0.02	±0.03	±0.05	±0.04	±0.04	±0.03	±0.07	±0.04

In order to compensate for loss of a zero-order stream reach by piping or infilling, we recommend remediation of a degraded zero-order site that has similar characteristics and is as nearby as possible to the impacted site. In this study, first-order sites appeared to have similar biological values and ecological functions to zero-order sites, suggesting that a first-order mitigation site may be substituted for a zero-order one. However, we advise caution in substituting a first-order stream, as our knowledge of zero-order streams is still limited. In particular, this study was limited to perennially-flowing zero-order habitats; zero-order sites that include non-flowing habitats (e.g., pools or wet mud) for part or all of the year may have different fauna, therefore compensation for loss of a stream with seasonally non-flowing habitats should require remediation of an equivalent stream. The mitigation site should be located as close as possible to healthy (e.g., forested) stream reaches, as these can provide a source of aquatic invertebrates to re-colonise the remediated stream.

In order to calculate the length of stream that must be remediated as compensation for the loss of non-biological functions in a flowing zero-order stream, we recommend the formula for environmental compensation ratio (ECR) adopted by Auckland Regional Council (Rowe et al. 2008). The function scores in the above table may be used as a guide to calculate future SEV scores for the impact and remediation sites, noting that in this study NFR, CFP, FPR and FSH scores for native forest were artificially low due to site-specific factors. To reduce the chance of overall loss of biodiversity, we recommend calculating a second ECR based on EPT richness. The ECR used should be greater of the two.

1. Introduction

1.1 The importance of headwater streams

Though often overlooked, headwater streams are numerically an important part of stream networks. In North America, studies have estimated that headwater streams (in these cases first- and second-order streams) account for 80% of the total length of a stream network (Wipfli et al. 2007). In Auckland, Storey and Wadwha (2009) estimated that perennial streams too small to be included on 1:50 000 topographic maps totalled 7200 km, representing 44% of perennial stream length in the region. In that study, intermittent (seasonally dry) streams totalled 4500 km of the stream network.

Headwater streams have gained increasing attention in recent years, with several North American and Mediterranean studies highlighting their local and landscape level importance (Freeman et al. 2007, Meyer et al. 2007, Wipfli et al. 2007, Maasri et al. 2008). These studies have shown that headwater streams may harbour unique species of aquatic invertebrates because they differ physically and chemically from higher-order streams and because they can act as refugia, protecting invertebrates from competitors, predators and high-flow events (Meyer et al. 2007). Certain species rely strongly on headwaters, either to increase survivorship of particular life stages, or to increase the connectivity between populations (Freeman et al. 2007). On a landscape scale headwaters are more variable than higher-order streams in the habitat conditions they provide, resulting in more variable invertebrate communities and therefore increased regional biodiversity (Meyer et al. 2007). Degradation of headwater streams by urbanisation may eliminate taxa unique to headwaters more than taxa common to downstream reaches (Smith and Lamp, 2008).

Headwater streams have a strong influence on the downstream river network, and indeed are considered crucial for sustaining the structure, function productivity and biodiversity of downstream ecosystems (Wipfli et al. 2007). As a refuge for aquatic invertebrates, headwater streams may provide a source of colonists to downstream reaches following disturbance events such as floods (Meyer et al. 2007). Having a closer relationship with the surrounding terrestrial environment than higher-order streams, they receive large fluxes of materials and energy from the land, including nutrients, woody debris and invertebrate prey, which they export downstream, to subsidise downstream productivity (Wipfli et al. 2007). They are also effective in processing terrestrial materials, and eutrophication of downstream receiving waters has been known to result when large-scale alteration of headwaters has occurred (Freeman et al. 2007)

1.2 Zero-order streams in Auckland and Waikato

In the Auckland and Waikato regions, there have been two studies comparing the fauna of zero-order headwater streams to those in adjacent higher-order perennial streams (Parkyn et al. 2006a,b). Both of these studies involved sampling invertebrates on a longitudinal profile between the stream source and its confluence with a higher-order stream. Along this profile, three main habitat types were sampled – wet mud, isolated pools and flowing water. The former two habitat types were truly non-perennial, having no flow in summer and/or in winter, but the flowing water habitat type included short reaches that remained flowing during summer. Therefore, while the Waikato study refers to these streams as “non-perennial”, and the Auckland study as “headwaters”, they are referred to here as “zero-order streams” to acknowledge the mix of perennial and non-perennial habitats. Both studies covered a wide geographic area including streams on different geological types. The Waikato study included sites in four River Environment Classification (REC) classes that together account for 75% of the region’s mapped stream length. Whereas the Waikato study was confined to native forest streams, the Auckland study included streams in different land-use types (open pasture catchments, pasture catchments with riparian forest, and native forest catchments).

Both studies found that zero-order streams harboured high taxa richness. In the Waikato, taxon richness and EPT richness were lowest in the mud habitat and were slightly, but significantly, lower in isolated pools and flowing zero-order stream reaches than in higher-order streams. In Auckland, only the mud habitat had significantly lower richness and EPT richness than higher-order streams, and the highest richness was found in flowing zero-order reaches. As well as harbouring high taxa richness, each zero-order site contained some taxa that were not found in the adjacent higher-order stream. Therefore, when zero-order streams were added to the higher-order stream, overall biodiversity increased. However, in the Auckland study, a multivariate analysis showed that the invertebrate communities of zero-order flowing and isolated pool habitats were not significantly different to the community of adjacent higher-order stream; only the mud habitat had a significantly different community. In the Waikato study, the mud and isolated pools communities were significantly different to the higher-order stream and flowing zero-order habitat communities, but flowing zero-order habitats did not have significantly different invertebrate composition to higher-order streams.

The Auckland study concluded that zero-order streams contain specialist species that do not commonly occur in higher-order streams. The Waikato study concluded that including zero-order streams in assessments of biodiversity could more than double the number of taxa found.

In the Auckland study, the effects of land-use type were greater than the effects of habitat type on taxon richness, EPT richness and community composition. Pasture streams had lower taxon richness and EPT richness than native forest and riparian forest streams. Furthermore, land-use affected the differences between habitat types in zero-order streams: in native forest streams there was little difference in total richness between mud, isolated pools and flowing water habitats, but in pasture streams, mud habitats had lower richness than pools and flowing water habitats.

1.3 Knowledge gaps and aims of this study

The studies by Parkyn et al. (2006a,b) have shown the biodiversity values of zero-order headwater streams, and highlighted the differences in taxonomic composition between these and adjacent higher-order streams. In the Waikato, these results were shown only for native forest streams, whereas in Auckland, results included streams in different land-use types (pasture, riparian forest and native forest). Therefore the first aim of this study was to complement these studies by determining the biodiversity values and taxonomic distinctiveness of zero-order streams in different land-use types in Waikato.

As well as biological values, overseas studies have also emphasised the ecological functions that headwaters provide to the stream network (Freeman et al. 2007; Wipfli et al. 2007). In New Zealand, the Waikato study focused exclusively on invertebrate communities and basic parameters of their aquatic habitat. The Auckland study, although it included aspects of hydrology and water quality (McKergow et al. 2006, Sukias and Nagels 2006), did not examine these using an ecological function approach. Therefore the second aim of this study was to identify the ecological functions provided by zero-order streams, and quantify these relative to higher-order streams.

1.4 Stream Ecological Valuation

The SEV, or Stream Ecological Valuation (Rowe et al. 2008), quantifies the ecological functions of stream reaches. It considers sixteen different functions that stream reaches provide to the stream network, and rates each function according to how well it is fulfilled by the particular stream reach.

The stream functions fall into four categories (Table 1). Hydraulic functions include measures of the flow regime (how close it is to the natural hydrologic regime) and measures of how well the stream is connected to groundwater, flood plains and other stream reaches. Biogeochemical functions include the ability of the stream to maintain suitable levels of oxygen, temperature and organic matter, and its ability to decontaminate pollutants. Habitat provision functions consider the suitability of the stream habitat for fish and invertebrates. Biodiversity functions measure the ability of the stream to maintain healthy fish and invertebrate fauna.

Rowe et al. (2008) also provide a method for quantifying environmental compensation required when a stream reach is impacted by development, e.g., through piping or infilling. Environmental compensation is assumed to involve remediation works on a degraded stream reach nearby, and the method calculates the length of stream that must be remediated, given the degree of ecological degradation on the stream being impacted. SEV scores without the biological functions (IFI, FFI and ABI) are used throughout, due to the difficulty of predicting the biological responses to remediation works. First the current SEV score is calculated for the reach to be impacted (SEV_i-C) and for the proposed environmental compensation site (SEV_m-C). Then the "best potential SEV" score is calculated for the same two sites (SEV_i-P and SEV_m-P respectively), using predicted function scores and assuming that best practice remediation works have been used at each site. An "impact SEV" score is calculated for the reach to be developed (SEV_i-I), using predicted outcomes of the development on each SEV function. Finally, the environmental compensation ratio (ECR) is calculated using the following formula:

$$ECR = [(SEV_i\text{-}P - SEV_i\text{-}I)/(SEV_m\text{-}P - SEV_m\text{-}C)] \times 1.5$$

The multiplication factor of 1.5 is used to compensate for the time lag between performing stream remediation works and realising the ecological benefits of such works. The ECR formula has been applied successfully in Auckland streams but to date has not been tested elsewhere. The use of non-biological functions of SEV makes it suitable for the present study, in which only the non-biological functions were used.

Table 1: Ecological functions comprising the Stream Ecological Valuation (SEV).

Function category	Function abbreviation	Function name
Hydraulic	NFR	natural flow regime
Hydraulic	CFP	connectivity to floodplain
Hydraulic	CSM	connectivity for species migrations
Hydraulic	CGW	connectivity to groundwater
Biogeochemical	WTC	water temperature control
Biogeochemical	DOM	dissolved oxygen maintenance
Biogeochemical	OMI	organic matter input
Biogeochemical	IPR	instream particle retention
Biogeochemical	DOP	decontamination of pollutants
Biogeochemical	FPR	floodplain particle retention
Habitat provision	FSH	fish spawning habitat
Habitat provision	HAF	habitat for aquatic fauna
Biodiversity	FFI	fish fauna intact
Biodiversity	IFI	invertebrate fauna intact
Biodiversity	ABI	aquatic biodiversity intact
Biodiversity	RVI	riparian vegetation intact

1.5 Definitions

In this study zero-order streams are defined as the smallest channels in the stream network, within about a hundred metres of where the stream channel is initiated. Because stream networks expand and contract between dry and wet seasons, such reaches may or may not be intermittent, depending on the hydrological properties of the underlying aquifer.

This definition was adopted to focus on reaches similar in size to the flowing water habitats studied by Parkyn et al. (2006a,b), though the flowing water habitats in those studies partly dried out during summer whereas our sites did not. According to the Strahler scheme, the stream reaches in the present study are, strictly speaking, first-order, however because they typically do not appear on 1:50 000 topographic maps, we refer to them as “zero-order”. In this study, zero-order reaches were compared with reaches known or assumed to have perennial flow. These reaches are referred to as “first-order” as typically they are downstream of the confluence of two zero-order streams.

2. Methods

2.1 Sites

Two Waikato study areas were selected in contrasting topographic types. The first area, around Pokeno (175°1'E, 37°15'S), just south of the Bombay Hills at the northern boundary of the Waikato Region, represents low topography. In this area, streams selected for study were mostly in catchments with slopes <5°, though a few were in catchments with slopes up to 20° (Fig. 1). Roughly half the study reaches drain into the Whakapipi Stream, which flows into the Waikato River 2 km southwest of Tuakau (Fig. 2); most of the remaining reaches drain into the Pokeno and Helenslee Streams, which merge just east of Pokeno, flowing into the Mangatawhiri River and then into the Waikato River south of Pokeno (about 12 km upstream of the Whakapipi). Two study reaches drain directly into the Mangatawhiri River east of Pokeno. The underlying geology around Pokeno is largely volcanic ashes (Mo and Vo in New Zealand Land Resource Inventory, Landcare Research NZ Ltd.), with a few sites on bedded sandstone (Sb) and one site (NF1 zero-order) located on greywacke (Gw).

The second study area, Whatawhata Research Station (175°15'E, 37°47'S), south of the Hakarimata Range and about 16 km west of Hamilton, represents steep country. In this area, streams selected for study were mostly in steep (>30°) "v"-shaped catchments, though a few of the first-order reaches were in rolling (17–20°) catchments with narrow floodplains adjacent to the stream channel (Fig. 1). All the stream reaches were within 6 km of each other, two draining into the Tunaeke Stream, two into the Karakariki Stream, two into the Whakakai Stream and the rest draining into the Kiripaka Stream. All of these streams are tributaries of the Waipa River, joining the river over a distance of 5 km (Fig. 3). The underlying geology at Whatawhata is mostly sedimentary sandstones and siltstones (greywacke and argillite) laid down in the Mesozoic, which are overlain by yellow brown earth soils (Kaawa hill soil, an Ochreptic Hapludult, and the Waingaro steepland soil, an Umbric Dystrochrept). Patches of overlying volcanic ash remain in less steep parts of the catchments and these have formed yellow brown loam soils (Dunmore silt loam, a Typic Hapludand) (Quinn and Stroud 2002). The pasture area was converted from native forest c. 80 years ago and is intensively stocked with sheep and cattle (Quinn and Stroud 2002).

In each of these two areas, stream reaches were chosen at sites representing a gradient in land-use intensity from native forest to open pasture. "Native forest" sites were defined as those where most of the catchment upstream was covered in intact native forest. "Thick riparian forest" sites had forested buffer strips 5-20 m wide on each side of the stream. Typically native understorey was intact and stock access was restricted. "Thin riparian forest" had buffer strips 1-5 m wide, consisting of trees but typically little or no native understorey. In most of these sites, stock had access to the stream. At "pasture sites", near-stream vegetation consisted only of grasses and occasional shrubs. Stock access to the stream was unrestricted in almost all cases. In each land-use type, three replicate pairs of sites were selected, each pair consisting of a zero-order and a first-order reach on the same stream (Table 2). In Pokeno, where it was more difficult to find suitable first-order sites on the same stream as the zero-order sites, each first-order site was located on a stream as nearby as possible to the corresponding zero-order site.

In the original study design, the zero-order sites were intended to be intermittent, i.e., cease flowing during the driest part of the year. However, a 1-in-100 year drought during late summer 2008 meant that intermittent streams in Pokeno remained dry during the sampling period, and hence were not suitable for sampling. Therefore in Pokeno the smallest perennial channels were selected as zero-order sites. At Whatawhata the smallest zero-order sites proved not to be intermittent, therefore in this area too, the zero-order sites selected had perennial flow.

Table 2: Layout of sampling sites in each of the two study areas (Whatawhata and Pokeno). In the thick riparian forest first-order category, only two replicates were collected due to a lack of suitable sites.

		Increasing land-use stress →			
		native forest	**thick riparian forest**	**thin riparian forest**	**open pasture**
Stream order	zero-order	3 replicates	3 replicates	3 replicates	3 replicates
	first-order	3 replicates	2 replicates	3 replicates	3 replicates

2.2 Field methods

At each site, field data collection consisted of surveys using the non-biological parts of the Stream Ecological Valuation (SEV) methodology (Rowe et al. 2008) and sampling of benthic invertebrates. A standard 50 m reach of stream was defined for data collection at each site.

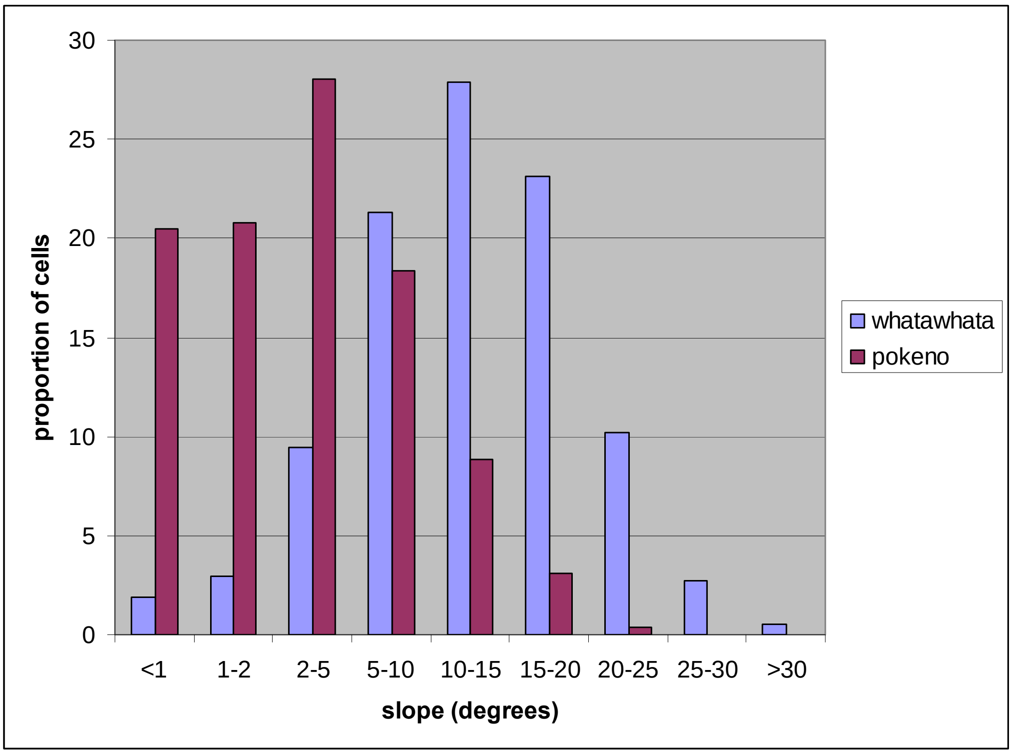

Figure 1: Topography of the two study areas, as shown by a slope raster in Geographic Information Systems (GIS). Proportion of cells refers to proportion of study area.

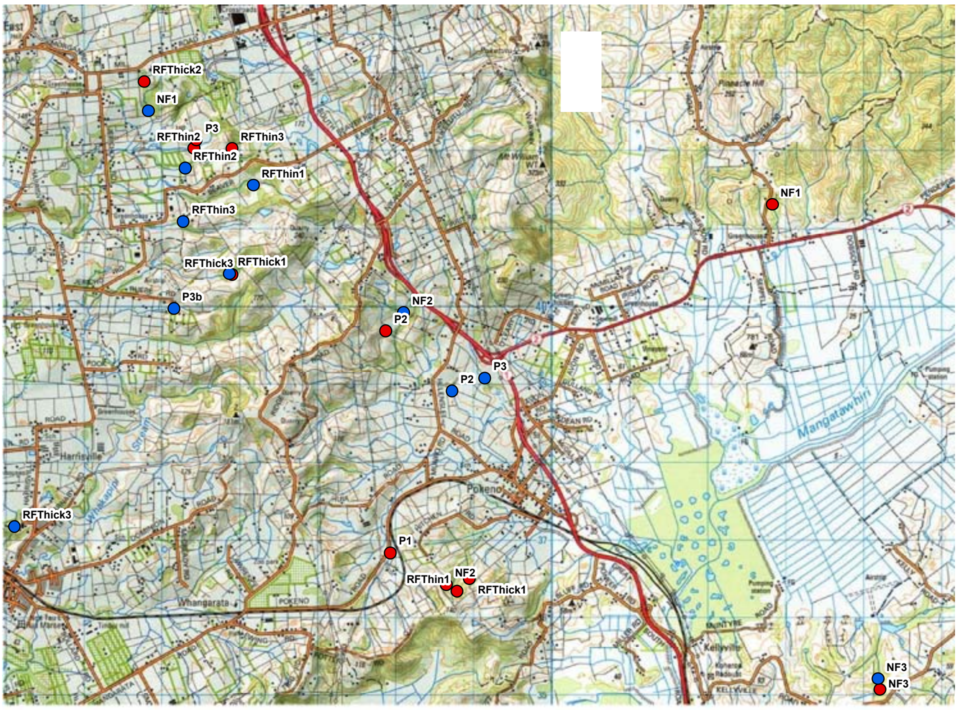

Figure 2: Map of the Pokeno study area showing locations of sample sites. Red dots are zero-order sites and blue dots are first-order sites.

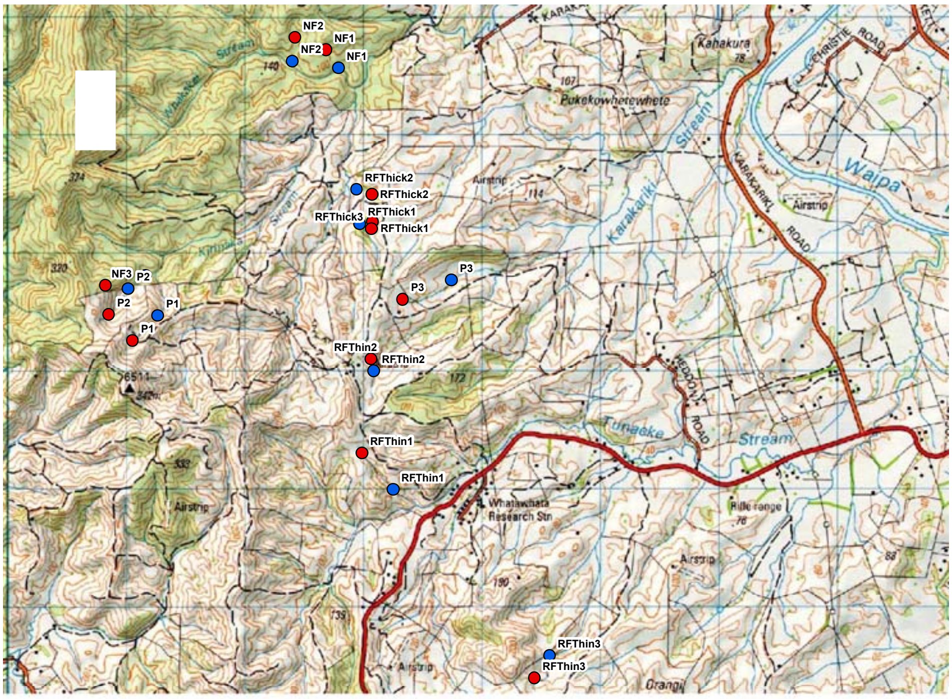

Figure 3: Map of the Whatawhata study area showing locations of sample sites. Red dots are zero-order sites and blue dots are first-order sites.

2.2.1 Stream Ecological Valuation (SEV)

For this study, the non-biological functions of the SEV (called nb-SEV hereafter) were used as an ecological function score. The biological functions omitted from the full SEV were FFI (fish fauna intact), IFI (invertebrate fauna intact) and ABI (aquatic biodiversity intact). Because we were focusing on zero-order streams, which do not connect with aquatic habitats upstream, we also omitted CSM (connectivity for species migrations), leaving 12 of the 16 functions to produce the nb-SEV mean score.

nb-SEV functions are derived from one or more variables, most of which are assessed in the field, though some are derived from maps. Field-assessed variables include the physical dimensions of the stream and its floodplain, stream bed particle sizes, instream organic matter and plant growth, water flow types, artificial channel modifications, retention of organic matter, bank erosion, barriers to fish migration and riparian vegetation characteristics (see Rowe et al. (2008) for a full list of the variables and the stream characteristics they are based on). All of these can be determined using visual assessments and simple equipment such as measuring rods. Retention of organic matter involves releasing 20 plastic triangles (leaf analogues) across the width of the stream channel and recording the distance travelled (and the structures they were retained on) after they have stopped moving.

Data analysis

The nb-SEV variables were weighted and combined in a mathematical equation to produce a score between 0 and 1 for each function. The final nb-SEV score was an average of the 12 individual function scores, and provided an overall measure of the functional health of the stream. Some nb-SEV variables can only be calculated with respect to Reference sites. Reference sites, considered close to pristine, also provide a measure of the maximum score that can reasonably be achieved in a particular location. In this survey, we separated zero-order from first-order streams, and for each of the two stream orders designated the three native forest replicates as reference sites.

2.2.2 Benthic macro-invertebrates

Benthic aquatic macro-invertebrates were sampled by kicking stream bed sediments over approximately 0.1 m^2 (sampling effort standardised among samples), and the disturbed area swept with a 250-μm mesh hand net. Five such samples, located to cover all available habitat types in the 50 m reach, were pooled.

In the lab, invertebrates were counted using protocol P2 “200 Individual Fixed Count with Scan for Rare Taxa” from Stark et al. (2001). The benthic sample was spread evenly across a gridded tray, and invertebrates removed from each grid square in turn. After 200 invertebrates had been removed, remaining invertebrates from the current grid square were collected, and the number of grid squares sorted in this way was recorded as a proportion of the total. Collected insects were identified to genus (except for some Diptera for which family was the lowest possible taxonomic level) using the keys of Towns and Peters (1996), Winterbourn et al. (2006) and Smith and Ward (2007); other invertebrates identified to the lowest possible taxonomic level. The abundance of each invertebrate taxon was then multiplied by the proportion of the total sample that had been sorted, in order to estimate the total abundance of each taxon in the sample. Finally, the full sample was scanned for rare taxa and the raw abundance of each new taxon found was added to the numbers derived above.

Data analysis

To assess alpha (within-habitat) biodiversity of invertebrates, three metrics were used. Taxon richness is a simple count of all the taxa present in a sample. Healthy stream ecosystems typically support a higher number of taxa than very degraded ones. However, slight land-use stress in a stream catchment may result in higher taxon richness as the slightly increased light and nutrients in the stream increase instream productivity (a subsidy-stress gradient; Quinn, 2000). Therefore we could expect taxon richness to increase with slight land-use stress (or slightly impaired ecological function), then decline as land-use stress intensifies. EPT richness is a simple count of the genera present belonging to the orders Ephemeroptera (mayflies), Plecoptera (stoneflies) and Trichoptera (caddisflies). In streams where these orders predominate, they tend to be favoured where land-use stress is lower, thus higher EPT richness reflects better stream ecological functioning. Shannon diversity, like total richness, counts all taxa in the habitat, but also incorporates relative abundance of the taxa, thus it is higher for samples that have a greater number of taxa present and more even abundances among taxa. Poorly-functioning stream ecosystems tend to be dominated by high abundances of a few taxa, whereas better-functioning ones support more even abundances among a range of taxa. Therefore higher Shannon diversity typically is associated with better ecological functioning. Zero-order and first-order sites were compared on the basis of these univariate measures using Wilcoxon signed-rank tests, paired t-tests and Analysis of variance (ANOVA) in SPSS™ v11 software.

To test whether the fauna of zero-order sites is distinct from that of first-order sites, two statistical approaches were used. First, we tested whether the number of additional taxa when a zero-order site was added to a first-order site was greater than the number of additional taxa when a first-order site was added to a first-order site. This was a randomisation test, in which the number of taxa added when each zero-order sample was added in turn to each first-order sample from the same area and land-use type was recorded (total of 22 combinations, since zero-order/first-order pairs from the same stream were not included). Then the number of taxa added when each first-order sample was added to each other first-order sample from the same area and land-use type was similarly recorded (total of 20 combinations). The two datasets were then compared using a t-test.

The second approach was to assess beta diversity (turnover of taxa between habitats), using Bray-Curtis similarity to compare sites pairwise. Bray-Curtis is commonly used in ecological studies as it has fewer drawbacks than most other similarity measures, and for presence-absence data, is mathematically equivalent to Sorenson's similarity index, a popular measure of beta diversity (Clarke and Warwick, 2001). Based on the matrix of Bray-Curtis pairwise similarities, differences between sites were displayed using non-parametric Multi-Dimensional Scaling (MDS), and differences between site groups (stream orders and land-use types) were tested for statistical significance using Analysis of Similarities (ANOSIM). The similarity percentages (SIMPER) routine was then used to identify the taxa that most strongly distinguished between groups of sites. These analyses were performed using Primer 6™ software (Primer-E Ltd.).

Relationships between land-use stress (substituting dummy variables 1-4), ecological function (nb-SEV score), taxon richness, EPT richness and Shannon diversity were determined using linear regression and multiple linear regression with SPSS™ v11 software. To determine whether (and which) individual nb-SEV functions differed significantly between zero-order and first-order streams, discriminant function analysis (DFA) was used with SPSS v11™ software.

To determine whether there were significant differences between zero-order and first-order streams with regard to any of the individual functions that make up the nb-SEV score, a permutational multivariate analysis of variance (Anderson 2005) was used.

3. Results

3.1 Physical characteristics of study sites

The zero-order streams were all small (<0.7 m wide) and shallow (<0.10 m) with discharge <3.5 Ls^{-1} (Table 3). First-order streams were slightly larger, up to 1.4 m wide, 0.32 m deep and with discharge up to 4.0 Ls^{-1}. Overall, the stream bed of zero-order and first-order streams in both areas was dominated by sand and silt, but both areas and stream orders showed high variability, each having some sites dominated by bedrock or with a moderate abundance of cobbles.

Table 3: Physical characteristics of study streams (averaged over all land-use types and replicates, followed by range in parentheses). *Pokeno zero-order site P1 omitted from these statistics as flow was elevated by recent rain on the day of sampling.

		Pokeno		**Whatawhata**	
		zero-order	**first-order**	**zero-order**	**first-order**
Average mid-channel depth (m)		0.04 (0.01-0.10)*	0.12 (0.05-0.26)	0.03 (0.01-0.05)	0.09 (0.02-0.32)
Average width (m)		0.45 (0.2-0.7)*	0.89 (0.4-1.4)	0.37 (0.2-0.5)	0.57 (0.3-1.1)
Discharge (Ls^{-1})		0.50 (0.01-3.4)*	10.9 (0.22-3.21)	0.2 (0.03-0.3)	0.73 (0.06-4.0)
Substrate (% cover)	sand/silt (<2mm)	76 (22-100)	78 (24-100)	33 (9-99)	39 (11-98)
	small gravel (2-8mm)	2 (0-8)	1 (0-4)	16 (0-29)	13 (0-19)
	small-medium gravel (8-16mm)	2 (0-14)	1 (0-6)	13 (1-28)	10 (0-24)
	medium gravel (16-32mm)	3 (0-9)	2 (0-10)	9 (0-22)	10 (1-22)
	large gravel (32-64mm)	5 (0-13)	2 (0-9)	7 (0-19)	7 (0-21)
	small cobble (64-128mm)	2 (0-11)	1 (0-4)	3 (0-10)	3 (0-10)
	large cobble (128-256mm)	1 (0-4)	1 (0-11)	1 (0-2)	1 (0-6)
	boulder (>256mm)	2 (0-14)	4 (0-24)	1 (0-6)	2 (0-10)
	bedrock	7 (0-54)	9 (0-51)	18 (0-49)	16 (0-36)

3.2 Biodiversity values of Waikato zero-order streams

Zero-order streams in both Pokeno and Whatawhata had diverse benthic macro-invertebrate communities (Figs. 4 and 5). Most taxonomic groups were well-represented in zero-order sites, and overall there was little difference in taxonomic richness between the two stream orders. Statistical tests showed that at Pokeno, Shannon diversity, total richness and EPT richness of the zero-order sites were not significantly different from those of the first-order sites (Wilcoxon non-parametric paired samples t-test n=11, Z=-0.711, p=0.477; n=11 Z=-0.205, p=0.837; n=11, Z=-0.845, p=0.398 respectively). At Whatawhata, Shannon diversity was not significantly different (Wilcoxon paired samples t-test n=11, Z=-1.423, p=0.155), but total richness and EPT richness were significantly lower (Wilcoxon paired samples t-test n=11, Z=-2.247, p=0.025; n=11, Z=-2.142, p=0.032 respectively) in the zero-order sites than in the first-order sites (zero-order sites had an average of 4.8 EPT genera per sample whereas first-order sites had an average of 7.9 genera per sample).

A number of taxa were found in zero-order streams but not in first-order streams, and vice versa (Fig. 6). This suggests that zero-order streams may have a distinctive fauna that adds diversity to the wider stream network. However, the taxa that were present only in zero-order sites occurred rarely, most of them just once among 3 replicate sites. Since they did not occur consistently among zero-order stream samples, their absence from first-order stream samples could have been due to chance. The randomisation test showed that in Pokeno, adding a zero-order sample to a first-order sample did not add significantly more taxa to the total taxon count than adding a first-order sample (t=0.909, df=40, p=0.369). In Whatawhata, adding a zero-order sample to a first-order sample added significantly fewer taxa to the total taxon count than adding a first-order sample (t=2.595, df=40, p=0.013). When the same analysis was repeated using only EPT taxa, the results were very similar to those for all taxa (t=0.593, df=40, p=0.557 at Pokeno; t=3.021, df=40, p=0.004 at Whatawhata). This suggests that differences between the fauna of zero-order and first-order samples are due to chance rather than due to habitat differences between zero-order and first-order sites.

To further determine whether zero-order sites had a distinctive fauna, the presence-absence invertebrate data were analysed using Multi-Dimensional Scaling (Fig. 7). A two-way crossed ANOSIM, with land-use type and stream order as the two factors, was used to test whether any differences in faunal composition were due to chance alone. At Pokeno there was no significant difference in faunal composition between zero-order and first-order sites (Global R=-0.056, significance level=67.3%), i.e., there was no evidence for a distinctive fauna in zero-order streams across the four land-use types. At Whatawhata there was a significant difference (Global R=0.145, significance level=12.3%), however this was due to differences in relative abundance and absence of taxa from zero-order sites, not to the presence of distinctive zero-order stream taxa. Among the 10 taxa distinguishing most strongly between zero-order and first-order sites (according to SIMPER analysis; Table 4), only two (*Polyplectropus* and Ptilodactylidae) occurred more frequently in zero-order sites. Furthermore, these two taxa, and four others (Paraleptamphopidae, Sphaeriidae, *Neozephlebia scita* and Collembola) ranked in the top 20 taxa by SIMPER, were not absent from first-order sites, but just less frequent.

Therefore, while some taxa in zero-order samples were absent from first-order samples, these taxa did not occur consistently enough to demonstrate conclusively that there are distinctive taxa in headwater perennial streams of different sizes. It is possible that further sampling in first-order sites may show they occur there also.

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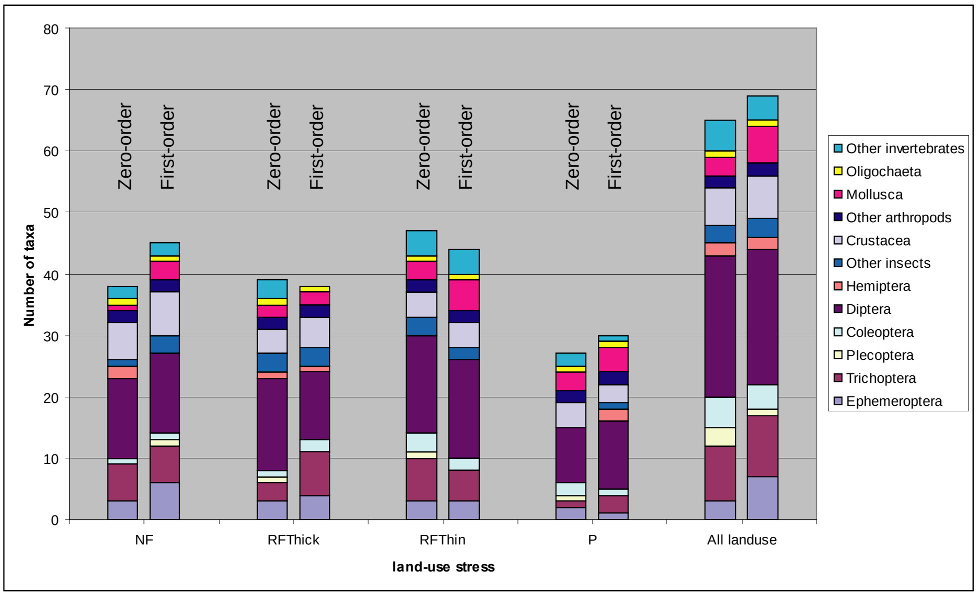

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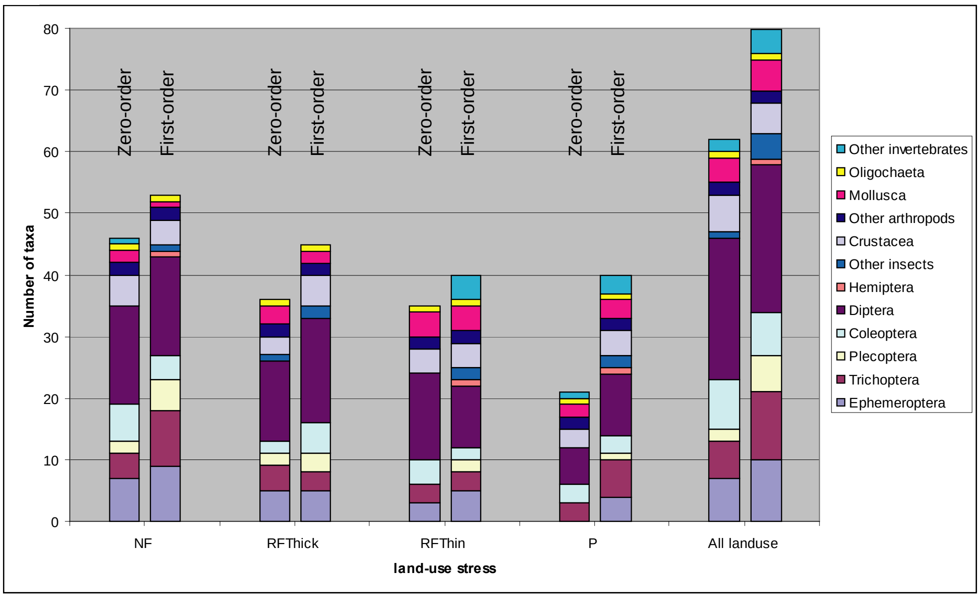

Figure 4: Taxonomic richness per land-use type in zero-order and first-order sites at a) Pokeno and b) Whatawhata. Data from all 3 replicates combined.

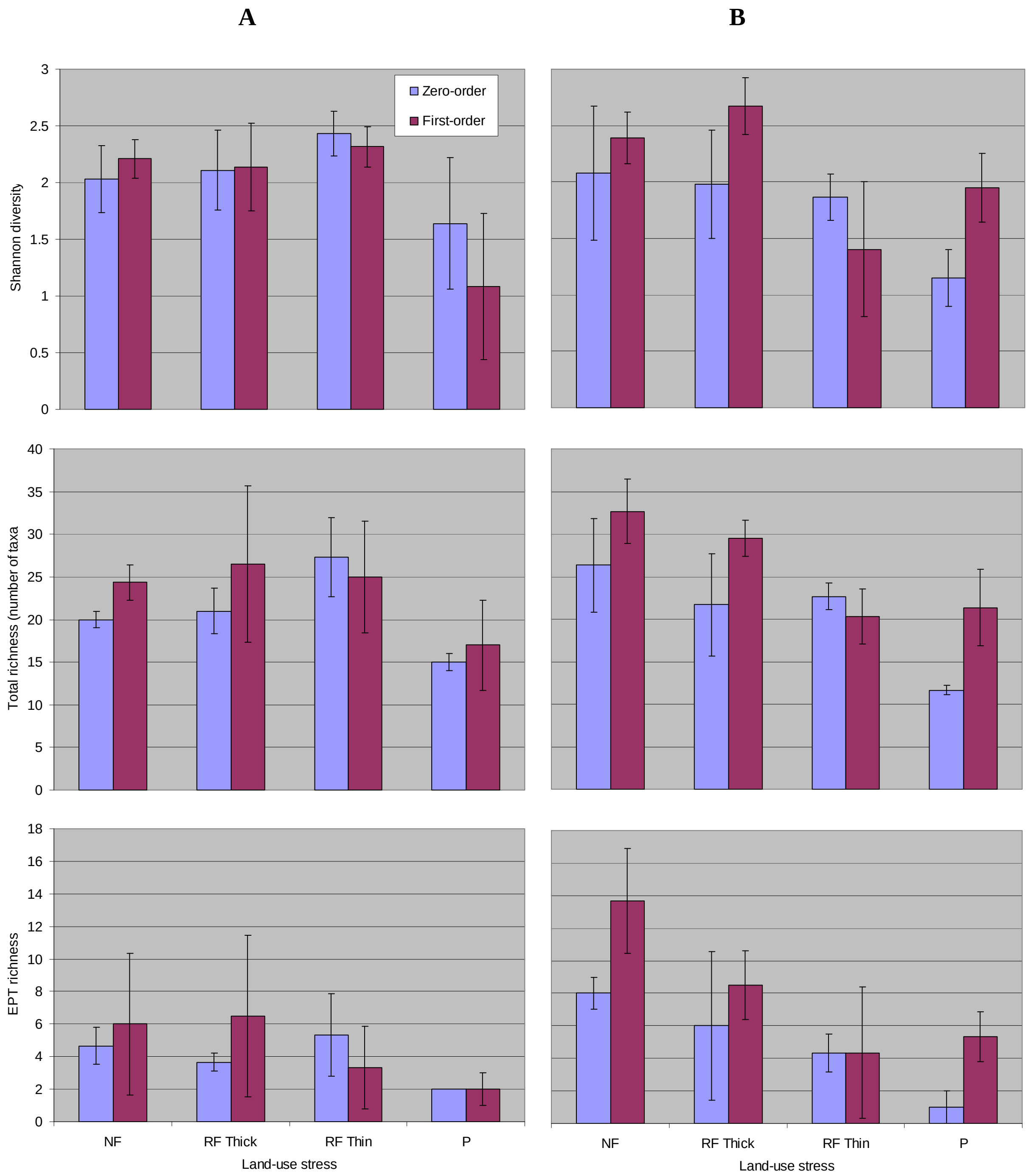

Figure 5: Shannon diversity, total richness and EPT richness (number of taxa belonging to Ephemeroptera, Plecoptera and Trichoptera) per sample in zero-order and first-order sites at a) Pokeno and b) Whatawhata. Bars are averages of 3 replicates (± 1 standard deviation).

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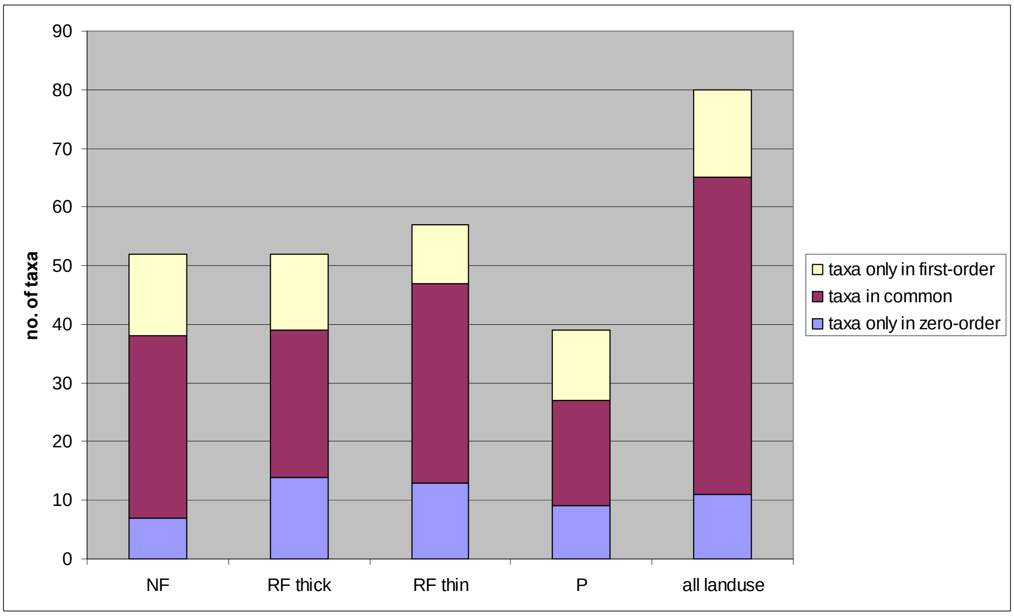

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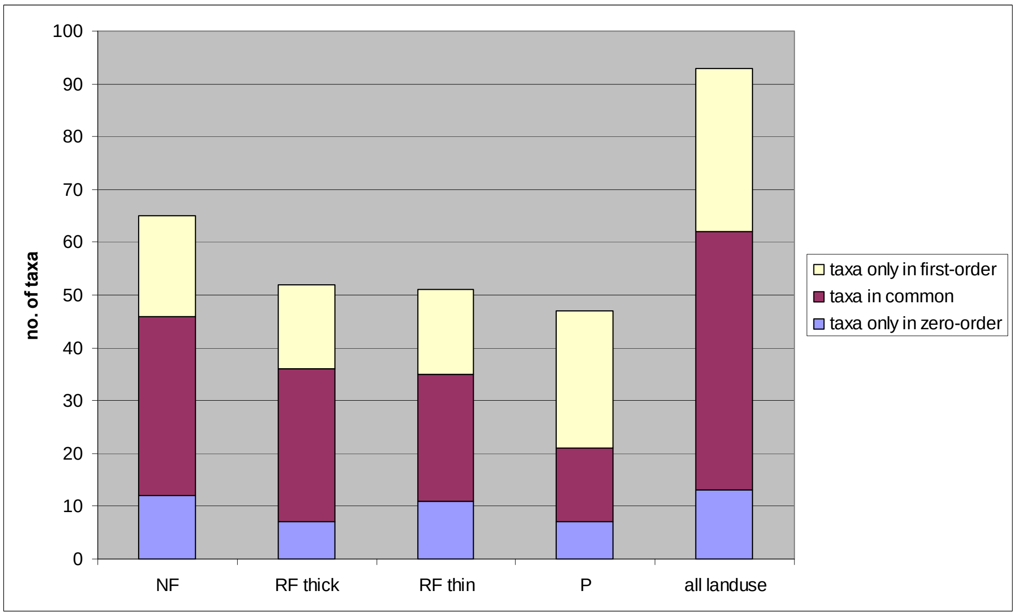
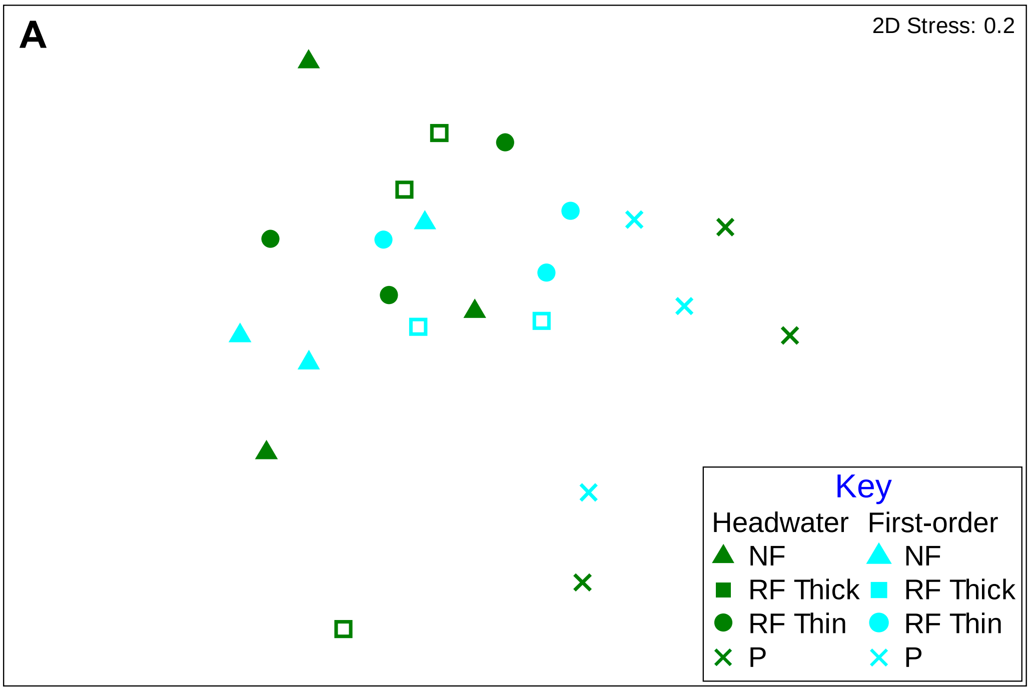


Figure 6: Number of taxa found only in zero-order or first-order sites and those in common, at a) Pokeno and b) Whatawhata.



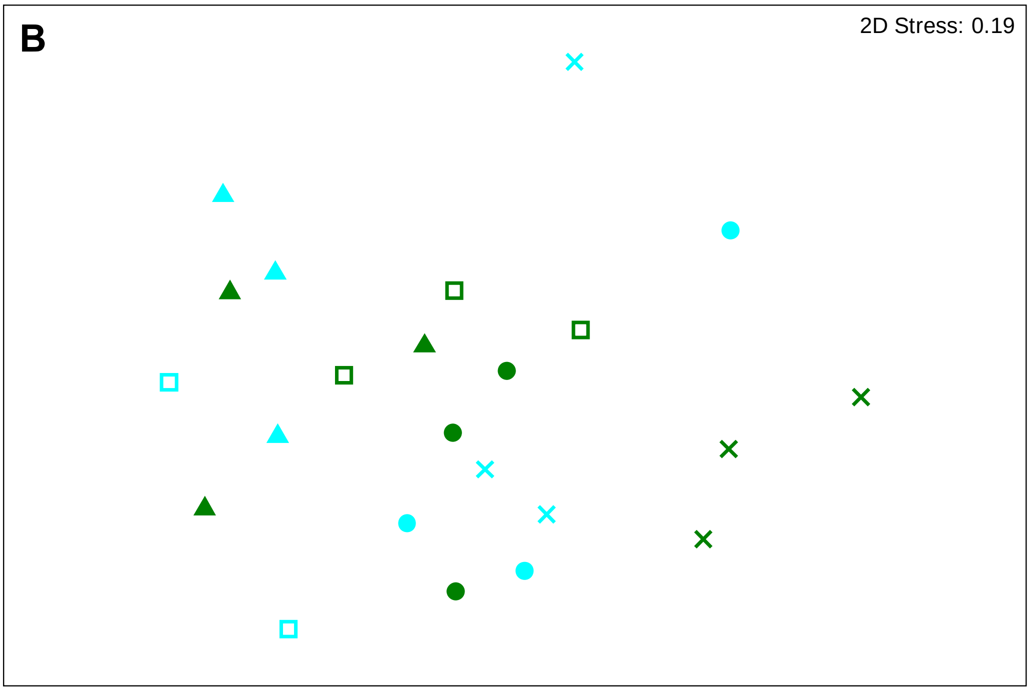


Figure 7: Non-parametric multi-dimensional scaling (MDS) two-dimensional plots of presence-absence invertebrate data from a) Pokeno and b) Whatawhata. MDS was based on Bray-Curtis similarity measure.

Table 4: SIMPER analysis comparing zero-order and first-order stream sites at Whatawhata, based on presence-absence invertebrate data. Taxa are listed in order of how strongly and consistently they contributed to the difference in taxonomic composition between zero-order and first-order sites. Taxa that occurred more frequently in zero-order streams than first-order sites are shown in italics.

Taxon	Zero-order freq. of occurrence	First-order freq. of occurrence	Average Dissimilarity	Dissimilarity/SD	Contribution%
Deleatidium	0.17	0.73	1.44	1.12	2.92
Paralimnophila	0.33	0.55	1.40	1.13	2.85
Polyplectropus	*0.58*	*0.45*	*1.31*	*1.09*	*2.65*
Coloburiscus	0.17	0.55	1.25	0.94	2.53
Ptilodactylidae	*0.50*	*0.45*	*1.19*	*1.03*	*2.41*
Orthopsyche	0.58	0.64	1.18	0.89	2.40
Polypedilum	0.67	0.73	1.17	0.82	2.38
Paradixa	0.42	0.82	1.17	0.93	2.37
Hexatomini:other	0.58	0.64	1.13	0.89	2.30
Elmidae	0.25	0.55	1.13	0.97	2.29
Paraleptamphopidae	*0.83*	*0.55*	*1.06*	*0.84*	*2.16*
Zephlebia	0.75	0.91	1.06	0.73	2.14
Talitridae	0.25	0.36	1.02	0.84	2.08
Oeconesidae/*Oeconesus*	0.25	0.36	1.01	0.80	2.04
Platyhelminthes	0.08	0.36	0.89	0.68	1.81
Scirtidae	0.25	0.27	0.89	0.82	1.80
Sphaeriidae	*0.42*	*0.18*	*0.87*	*0.73*	*1.77*
Corynoneura	0.00	0.36	0.85	0.71	1.72
Neozephlebia scita	*0.58*	*0.36*	*0.84*	*0.78*	*1.70*
Collembola	*0.92*	*0.64*	*0.80*	*0.65*	*1.62*

3.3 Relationships among invertebrate biodiversity, land-use stress, ecological function and stream order

3.3.1 Ecological function, stream order and land-use stress

Ecological function (as measured by the mean of the non-biological SEV functions) did not change significantly between zero-order and first-order streams in either Pokeno or Whatawhata. This was demonstrated by paired t-tests of zero-order vs first-order sites (df=10, t=-0.969 p=0.355 at Pokeno; df=10, t=-0.179, p=0.861 at Whatawhata).

Mean non-biological SEV score was only a little higher in the native forest sites than in other land use types (Fig. 8 and Table 5). Nevertheless, ecological function was significantly correlated with land-use stress. At Pokeno, land-use stress explained 80% of variability in nb-SEV score among zero-order sites, but among first-order sites land-use stress explained only 14% of the variability (Fig. 8a). The reasons for the low value of the latter were the high variability among first-order sites (especially RFThick sites, which included a hard-bottom and a soft-bottom site), and the low slope of the correlation between nb-SEV score vs. land-use stress. At Whatawhata land-use stress explained 73 and 79% of the variability in nb-SEV score among zero-order and first-order sites respectively (Fig. 8b).

A multiple regression combining land-use stress and stream order confirmed that land-use stress had a strong effect on nb-SEV score (41% of variability and 76% of variability explained at Pokeno and Whatawhata respectively) whereas stream order had minimal effect (3% and <0.1% variability explained at Pokeno and Whatawhata respectively; Table 6a and b respectively). Multiple regression on the entire data set, combining land-use stress, stream order and topography, showed that land-use stress explained 59% of the variability in nb-SEV score among all samples (Table 6c). In contrast, stream order explained only 0.3% and topography only 3.6% of the variability.

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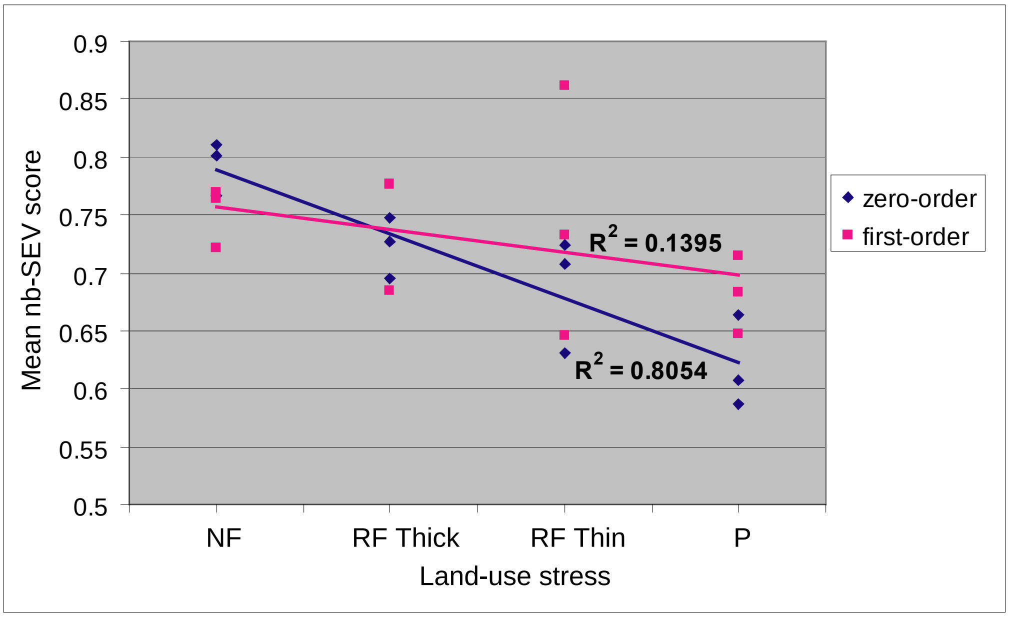

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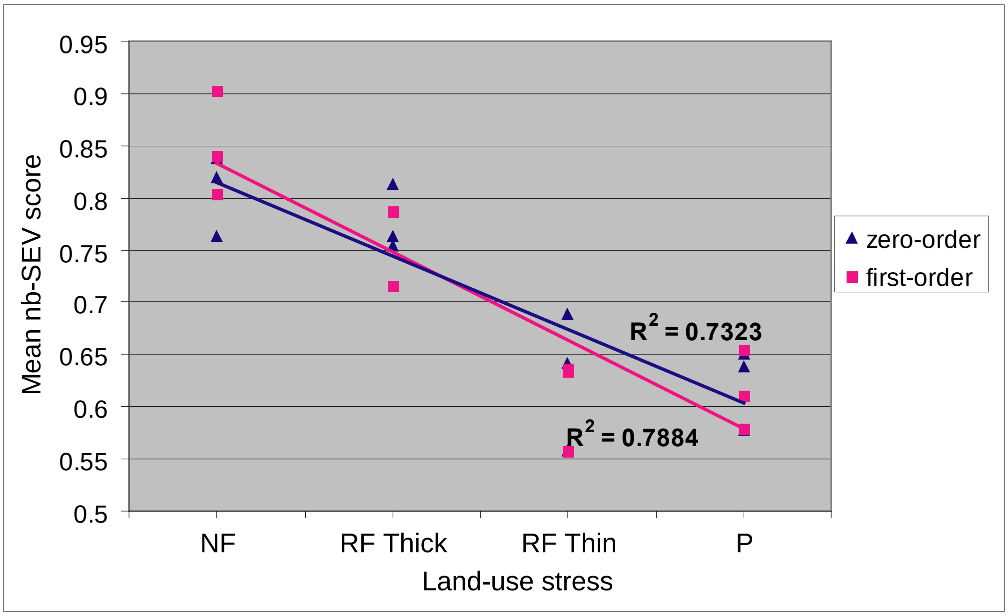

Figure 8: Relationships between mean nb-SEV score and land-use stress for the two stream orders at a) Pokeno and b) Whatawhata. Trend lines and R^2 values were obtained by using "dummy variables" 1-4 for increasing land-use stress (native forest to pasture).

Table 5: Mean nb-SEV scores for zero-order and first-order streams in Pokeno (lowland) and Whatawhata (steep hill country) sites in four levels of land-use stress. Abbreviations: NF (native forest), RF Thick (thick riparian forest), RF Thin (thin riparian forest), P (pasture).

	Lowland				Steep hill country			
	NF	RF Thick	RF Thin	P	NF	RF Thick	RF Thin	P
Zero-order ± std deviation	0.79 ±0.02	0.72 ±0.03	0.69 ±0.05	0.62 ±0.04	0.81 ±0.04	0.78 ±0.03	0.63 ±0.07	0.62 ±0.04
First-order ± std deviation	0.75 ±0.03	0.73 ±0.07	0.75 ±0.11	0.68 ±0.03	0.85 ±0.05	0.75 ±0.05	0.61 ±0.04	0.62 ±0.04

Table 6a: Summary of results for multiple regression, testing for effect of land-use stress and stream order (zero-order vs. first-order) on non-biological SEV score at Pokeno. Abbreviations: sr^2 is the square of the semi-partial correlation coefficient (i.e., the proportion of the variation in Shannon diversity that is explained by each factor); b is the unstandardised slope of each factor, β is the standardised slope; asterisk indicates β is statistically significant; p is the probability of β occurring due to chance alone; R2 is the proportion of the variation in Shannon diversity that is explained by the full model.

Variable	raw correlation	sr^2	b	Std error of b	β	p
Land-use stress	-0.645	0.412	-0.038	0.010	-0.649*	0.001
Stream order	0.162	0.031	0.023	0.022	0.175	0.306
		R^2	df	F		p
Full model		0.447	2,20	8.07		0.003

Table 6b: Summary of results for multiple regression, testing for effect of land-use stress and stream order (zero-order vs. first-order) on non-biological SEV score at Whatawhata. Abbreviations as above.

Variable	raw correlation	sr^2	b	Std error of b	β	p
Land-use stress	-0.871	0.757	-0.077	0.010	-0.870*	<0.001
Stream order	-0.032	<0.001	-0.003	0.022	-0.015	0.895
		R^2	**df**	**F**		**P**
Full model		0.758	2,20	31.324		<0.001

Table 6c: Summary of results for multiple regression, testing for effect of land-use stress, stream order (zero-order vs. first-order) and topography (Pokeno vs. Whatawhata) on non-biological SEV score at Whatawhata. Abbreviations as above.

Variable	raw correlation	sr^2	b	Std error of b	β	p
Land-use stress	-0.764	0.585	-0.058	0.007	-0.765*	<0.001
Stream order	0.044	0.003	0.010	0.017	0.059	0.554
Topography	-0.060	0.036	-0.010	0.017	-0.060	0.546
		R^2	**df**	**F**		**P**
Full model		0.590	3,42	20.152		<0.001

3.3.2 Individual non-biotic ecological functions, stream order and land-use stress

The mean nb-SEV score of the zero-order native forest sites was lower than expected due to several functions with particularly low scores (Table 7). These were NFR (natural flow regime), CFP (connectivity to floodplain), FPR (floodplain particle retention) and FSH (fish spawning habitat). NFR was low because of a high proportion of bank length showing erosion from flood flows. CFP, FPR and FSH were low mainly because of very narrow floodplains and low frequency of flooding.

A two-way crossed multivariate permutational ANOVA (Permanova), with stream order and land-use stress as independent factors, was used to test whether there were differences between zero-order and first-order streams and between levels of land-use stress with regard to any of the ecological functions that make up the nb-SEV scores (RFThick sites were omitted due to a missing data point). Whatawhata and Pokeno sites were analysed separately. Results of the Permanova showed no significant difference in non-biotic ecological functions between zero-order and first-order streams at either Pokeno (df=1,17; F=1.2982; p=0.25) or Whatawhata (df=1,17; F=0.4819; p=0.75). There were significant differences between land-use stresses in each area (df=2,17; F=1.5986; p=0.18 and df=2,17; F=0.5416; p=0.81 respectively), but there was no significant interaction between land-use stress and stream order.

To confirm the results of the Permanova, a univariate two-way ANOVA was performed on each nb-SEV function in turn, with land-use stress and stream order as independent factors. Two nb-SEV functions, CGW (connectivity to groundwater) and IPR (instream particle retention), were omitted as they scored 1 at all sites without exception. Out of the 20 ANOVAs performed on the remaining ten nb-SEV functions in two areas (Pokeno and Whatawhata), only one test (natural flow regime in Pokeno) showed a significant difference in a nb-SEV function between zero-order and first-order streams (df=1,18; F=10.173; p=0.008). This difference was due to slightly greater bank erosion among zero-order streams than first-order streams and very low variance in bank erosion among the first-order streams.

Table 7: Individual ecological function scores of zero-order streams in low and steep topography of different land-use types. These are the values that would be lost if a zero-order stream were piped or infilled.

	Lowland				Steep hill country			
SEV function	**NF**	**RF Thick**	**RF Thin**	**P**	**NF**	**RF Thick**	**RF Thin**	**P**
NFR	0.77	1.00	0.90	0.90	0.75	0.90	0.65	0.72
± std deviation	±0.14	±0	±0.09	±0.09	±0.17	±0.09	±0.17	±0.25
CFP	0.45	0.40	0.30	0.82	0.43	0.55	0.37	0.62
± std deviation	±0.23	±0	±0.09	±0.2	±0.25	±0.15	±0.2	±0.19
CGW	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
± std deviation	±0	±0	±0	±0	±0	±0	±0	±0
WTC	0.76	0.78	0.71	0.46	0.78	0.76	0.68	0.56
± std deviation	±0.04	±0.02	±0.02	±0.1	±0.01	±0.01	±0.09	±0.12
DOM	1.00	0.38	0.74	0.37	1.00	0.83	0.79	0.8
± std deviation	±0	±0.21	±0.31	±0.1	±0	±0.3	±0.36	±0.35
OMI	0.84	0.88	0.67	0.06	0.92	0.83	0.48	0.24
± std deviation	±0.16	±0.11	±0.07	±0.1	±0.03	±0.04	±0.29	±0.25
IPR	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
± std deviation	±0	±0	±0	±0	±0	±0	±0	±0
DOP	0.89	0.9	1.00	0.79	0.93	0.70	0.79	0.69
± std deviation	±0.1	±0.05	±0	±0.15	±0.06	±0.12	±0.22	±0.29
FPR	0.54	0.41	0.29	0.64	0.39	0.58	0.31	0.45
± std deviation	±0.22	±0.09	±0.07	±0.05	±0.12	±0.11	±0.18	±0.11
FSH	0.35	0.31	0.40	0.56	0.52	0.50	0.35	0.44
± std deviation	±0.23	±0.17	±0.2	±0.34	±0.02	±0.06	±0.19	±0.18
HAF	0.97	0.76	0.70	0.49	0.99	0.85	0.67	0.53
± std deviation	±0.04	±0.04	±0.13	±0.09	±0.01	±0.14	±0.11	±0.08
RVI	0.96	0.86	0.54	0.36	0.98	0.84	0.47	0.42
± std deviation	±0.08	±0.13	±0.05	±0.01	±0.04	±0.11	±0.10	±0.10
mean nb-SEV score	0.79	0.72	0.69	0.62	0.81	0.78	0.63	0.62
±std deviation	±0.02	±0.03	±0.05	±0.04	±0.04	±0.03	±0.07	±0.04

3.3.3 Biodiversity and land-use stress

In zero-order sites, Shannon diversity, total richness and EPT richness of the benthic macroinvertebrate communities decreased between native forest and pasture (Fig. 5), but the relationship was not linear in all cases. At Whatawhata a steady decline in all three biodiversity metrics with increasing land-use stress could be discerned; however, at Pokeno only EPT richness declined steadily whereas Shannon diversity and total richness remained more or less constant between native forest and thin riparian vegetation, and declined only between thin riparian vegetation and pasture. The response of Shannon diversity and total richness at Pokeno may be an example of a subsidy-stress gradient (Section 2.2.2). Linear regression slopes of biodiversity metrics vs. land-use stress (Table 8) were highly significant for zero-order streams at Whatawhata but not significant for zero-order streams at Pokeno. EPT richness appeared to be slightly more sensitive than Shannon diversity to the effects of land-use stress. Land-use stress appeared to have a stronger effect on biodiversity at Whatawhata than at Pokeno.

In order to use all the available data, invertebrate data from first-order streams and zero-order streams were combined, and multiple regression was used to separate the effects of land-use stress and stream order. At Pokeno, land-use stress was significantly correlated with Shannon diversity and EPT richness, though not with total richness (Tables 9a-11a). At Whatawhata, land-use stress was highly correlated with all three biodiversity metrics (Tables 9b-11b). In these multiple regressions, stream order overall had a weak effect on biodiversity metrics, explaining <4% of variability in biodiversity metrics at Pokeno and up to 15% at Whatawhata.

To determine the influence of topography, all invertebrate data from Pokeno and Whatawhata were combined in a multiple regression with three factors – land-use stress, stream order and topography (Tables 9c-11c). Topography was not significant in explaining variability in any of the three diversity metrics. It explained about as much variability as stream order for Shannon diversity, much less variability than stream order for total richness, and slightly more variability than stream order for EPT richness.

Table 8: Summary of results for regressions, testing for effects of land-use stress on Shannon diversity, total richness and EPT richness in zero-order streams at Pokeno and Whatawhata. Abbreviations: b is the unstandardised slope of the factor, β is the standardised slope; p is the probability of β occurring due to chance alone; asterisk indicates regression is statistically significant at p<0.05; R^2 is the proportion of the variation in biodiversity metric that is explained by land-use stress.

		Intercept	b	Std error of b	β	R^2	F	p
Pokeno	Shannon diversity	2.263	-0.0847	0.116	-0.224	0.050	0.530	0.483
	Total richness	23.000	-0.867	1.367	-0.197	0.039	0.402	0.540
	EPT richness	5.500	-0.633	0.454	-0.404	0.163	1.947	0.193
Whatawhata	Shannon diversity	2.494	-0.291	0.107	-0.652*	0.425	7.390	0.022*
	Total richness	31.333	-4.300	1.199	-0.750*	0.563	12.866	0.005*
	EPT richness	10.500	-2.267	0.581	-0.777*	0.604	15.231	0.003*

Table 9a: Summary of results for multiple regression, testing for effects of land-use stress and stream order (zero-order vs first-order) on Shannon diversity at Pokeno. Abbreviations: sr^2 is the square of the semi-partial correlation coefficient (i.e., the proportion of the variation in Shannon diversity that is explained by each factor); b is the unstandardised slope of each factor, β is the standardised slope; asterisk indicates β is statistically significant; p is the probability of β occurring due to chance alone; R^2 is the proportion of the variation in Shannon diversity that is explained by the full model.

Variable	raw correlation	sr^2	b	Std error of b	β	p
Land-use stress	-0.440	0.194	-0.201	0.091	-0.438*	0.040
Stream order	-0.129	0.014	-0.126	0.208	-0.120	0.553
		R^2	df	F		p
Full model		0.208	2, 20	2.626		0.097

Table 9b: Summary of results for multiple regression, testing for effects of land-use stress and stream order (zero-order vs first-order) on Shannon diversity at Whatawhata. Abbreviations as above.

Variable	raw correlation	sr^2	b	Std error of b	β	p
Land-use stress	-0.547	0.306	-0.267	0.085	-0.553*	0.005
Stream order	0.259	0.073	0.297	0.195	0.271	0.142
		R^2	**df**	**F**		**p**
Full model		0.373*	2, 20	5.943		0.009

Table 9c: Summary of results for multiple regression, testing for effects of land-use stress, stream order (zero-order vs first-order) and topography (Whatawhata vs Pokeno) on Shannon diversity. Abbreviations as above.

Variable	raw correlation	sr^2	b	Std error of b	β	p
Land-use stress	-0.493	0.245	-0.234	0.063	-0.495*	0.001
Stream order	0.070	0.006	0.086	0.143	0.080	0.552
Topography	-0.078	0.006	-0.083	0.143	-0.078	0.563
		R^2	**df**	**F**		**p**
Full model		0.256	3,42	4.812		0.006

Table 10a: Summary of results for multiple regression, testing for effects of land-use stress and stream order (zero-order vs first-order) on total richness at Pokeno. Abbreviations as above.

Variable	raw correlation	sr^2	b	Std error of b	β	p
Land-use stress	-0.318	0.104	-1.563	1.008	-0.322	0.137
Stream order	0.188	0.038	2.147	2.294	0.194	0.361
		R^2	**df**	**F**		**p**
Full model		0.139	2, 20	1.611		0.224

Table 10b: Summary of results for multiple regression, testing for effects of land-use stress and stream order (zero-order vs first-order) on total richness at Whatawhata. Abbreviations as above.

Variable	raw correlation	sr^2	b	Std error of b	β	p
Land-use stress	-0.712	0.518	-4.283	0.778	-0.720*	<0.001
Stream order	0.373	0.151	5.248	1.772	-0.388*	0.008
		R^2	df	F		p
Full model		0.658*	2, 20	19.208		<0.001

Table 10c: Summary of results for multiple regression, testing for effects of land-use stress, stream order (zero-order vs first-order) and topography (Whatawhata vs Pokeno) on total richness. Abbreviations as above.

Variable	raw correlation	sr^2	b	Std error of b	β	p
Land-use stress	-0.530	0.287	-2.923	0.664	-0.536*	<0.001
Stream order	0.287	0.089	3.697	1.511	0.298*	0.019
Topography	0.095	0.009	1.174	1.509	0.095	0.441
		R^2	df	F		p
Full model		0.378	3,42	8.525		<0.001

Table 11a: Summary of results for multiple regression, testing for effects of land-use stress and stream order (zero-order vs first-order) on EPT richness at Pokeno. Abbreviations as above.

Variable	raw correlation	sr^2	b	Std error of b	β	p
Land-use stress	-0.462	0.214	-1.047	0.446	-0.464*	0.029
Stream order	0.069	0.006	0.404	1.016	0.078	0.695
		R^2	df	F		p
Full model		0.220	2, 20	2.814		0.084

Table 11b: Summary of results for multiple regression, testing for effects of land-use stress and stream order (zero-order vs first-order) on EPT richness at Whatawhata. Abbreviations as above.

Variable	raw correlation	sr^2	b	Std error of b	β	p
Land-use stress	-0.700	0.501	-2.606	0.497	-.708*	<0.001
Stream order	0.367	0.145	3.194	1.131	.381*	0.010
		R^2	**df**	**F**		**p**
Full model		0.636*	2, 20	17.449		<0.001

Table 11c: Summary of results for multiple regression, testing for effects of land-use stress, stream order (zero-order vs first-order) and topography (Whatawhata vs Pokeno) on EPT richness. Abbreviations as above.

Variable	raw correlation	sr^2	b	Std error of b	β	p
Land-use stress	-0.565	0.325	-1.826	0.359	-0.570*	<0.001
Stream order	0.235	0.061	1.799	0.818	0.247*	0.033
Topography	0.304	0.092	2.217	0.817	0.304	0.01
		R^2	**df**	**F**		**p**
Full model		0.472	3,42	12.529		<0.001

3.3.4 Summary of relationships among biodiversity values, ecological function and land-use stress: a basis for decision-making for environmental compensation

Table 12 shows the biodiversity values, and Table 7 the ecological functions, that are likely to be lost if a zero-order stream in a particular land-use type in lowland or steep country area is piped or the land-use stress is increased. These two tables can be used as a basis for making decisions about environmental compensation for loss or degradation of a zero-order stream.

Table 12: Summary of biodiversity values belonging to zero-order streams in low and steep topography of different land-use types. These are the values that would be lost if a zero-order stream were piped or infilled.

	Lowland				Steep hill country			
	NF	RF Thick	RF Thin	P	NF	RF Thick	RF Thin	P
Total richness ±std deviation	20 ±1	21 ±2.6	27.3 ±4.6	15 ±1	26.3 ±5.5	21.7 ±6.0	22.7 ±1.5	11.7 ±0.58
EPT richness ±std deviation	4.7 ±1.2	3.7 ±0.6	5.3 ±2.51	2.0 ±1	8.0 ±1	6.0 ±4.6	4.3 ±1.2	1.0 ±1
Shannon diversity ±std deviation	2.03 ±0.30	2.11 ±0.35	2.43 ±0.20	1.64 ±0.58	2.08 ±0.59	1.98 ±0.48	1.87 ±0.21	1.15 ±0.25

4. Discussion

4.1 Distinctiveness of zero-order stream fauna

In the light of previous studies of headwater streams, we were surprised to find so little difference in the macro-invertebrate community between zero-order and first-order streams. The lack of difference in the three biodiversity metrics between zero-order and first-order streams was similar to the results of Parkyn et al. (2006a,b), which included perennial and non-perennial habitats. However, the lack of a distinct invertebrate community between zero-order and first-order streams initially appeared at odds. In this study, ANOSIM showed no significant difference in the community composition of zero-order vs. first-order streams at Pokeno, whereas at Whatawhata ANOSIM distinguished zero-order from first-order streams only by the absence of certain taxa from the zero-order streams. While some taxa were found only in zero-order sites, the randomisation test showed that adding a zero-order site to a first-order site did not increase the total taxon count any more than adding a second first-order site. Therefore, statistically, zero-order streams could not be shown to have a distinct fauna, and it is possible that the taxa that appeared unique to zero-order streams in fact occur (rarely) across a range of headwater streams.

A closer examination of the previous studies in Auckland and Waikato (Parkyn et al. 2006a,b) shows that our result was not as different from those studies as it first appeared. In both the Auckland and the Waikato studies, several habitat types in zero-order streams were sampled, including flowing water, isolated pools and wet mud. In the Auckland study, only the mud habitat had a significantly different invertebrate composition to the higher-order stream habitat according to MDS and ANOSIM, isolated pools and flowing zero-order stream habitats being indistinguishable from the higher-order stream habitat. In Waikato study (Parkyn et al. 2006b), both the mud and the isolated pools habitats had a significantly different invertebrate community to the higher-order stream habitat, but the flowing zero-order stream habitat did not. Therefore it appears that a significant change in stream invertebrate communities occurs at the point where flow ceases and the aquatic habitat is reduced to pools or mud. Our study, being conducted in perennially-flowing zero-order streams, did not capture this difference.

We believe that the conclusion of Parkyn et al. (2006b) that “inclusion of non-perennial stream sampling could more than double the overall estimate of biodiversity within stream systems” is somewhat overstated. Parkyn et al. reached this conclusion by comparing the total taxa richness of the combined sample from three zero-order habitats and an adjacent higher-order habitat (a total of 3.1 m^2) to the taxa richness of a single 1 m^2 sample from the higher-order stream habitat. However, the higher richness of the combined sample is at least partly due to a larger sampling area, and not necessarily to a different faunal composition in the zero-order habitats. This can be demonstrated in two steps. First, simply increasing the sampled area of the higher-order stream habitat by combining the four higher-order stream samples in the survey increases the total richness of that habitat. If this is done, and the same done with samples from all the zero-order habitats, the zero-order habitats increase the total taxon count by 38%. This is high, but not as high as the "more than double" stated by Parkyn et al. (2006b), and it still relies on a limited sample size (n=4). Second, a randomisation test, equivalent to that in the present study, shows that adding a zero-order site to a higher-order site does not increase the taxa richness by a greater amount than does adding a second higher-order site to the first. Therefore, as in the present study, it is possible that all of the same taxa found in the zero-order streams would also be found in higher-order streams if sampling effort in higher order streams were increased.

It should be noted that all of the taxa found by Parkyn et al. (2006b) in zero-order and non-perennial stream habitats are known to occur in higher-order or perennial streams. However, in that study, as in the present one, conclusions are limited by the current taxonomic knowledge of New Zealand aquatic invertebrates. Many New Zealand taxa have been described only to genus or higher level, thus it is possible that zero-order and non-perennial streams harbour different species of genera found in higher-order and perennial streams.

Other New Zealand studies of non-perennial habitats have found only taxa that are known also from perennial waters. Storey and Quinn (2008), Wissinger et al. (2009) and Larned et al. (2007) found that taxa distinguishing intermittent from perennial samples were rare, but not unknown, in perennial streams. This is in contrast with overseas studies, which have found some unique taxa in non-perennial streams (e.g., Dieterich and Anderson 2000). The difference between New Zealand and these other countries may be real, based on our different evolutionary history, or may be an artefact arising from gaps in the taxonomy of New Zealand aquatic invertebrates. Lack of taxonomic resolution among the New Zealand fauna limits the conclusions we can draw in this study, and we caution that future work may reveal taxa or genetic variants that are unique to zero-order streams.

4.2 Ecological functions of zero-order streams

Mean non-biological SEV scores of the zero-order native forest sites (0.79±0.02 and 0.81±0.04 for Pokeno and Whatawhata respectively) were lower than those of typical native forest streams (0.944; Rowe et al. 2008). The functions with particularly low scores that lowered the mean nb-SEV score of the native forest sites were NFR (natural flow regime), CFP (connectivity to floodplain), FPR (floodplain particle retention) and FSH (fish spawning habitat). NFR was low because of a high proportion of bank length showing erosion from flood flows. The other functions were low mainly because of very narrow floodplains and low frequency of flooding. These results probably reflect the fact that zero-order streams are positioned near the tops of catchments. An assumption of SEV is that streams higher in catchments will flood less frequently than those further down, and users of SEV are recommended to give a lower flood frequency score to streams higher in the landscape (Rowe et al. 2008). Near the tops of catchments, stream valleys also tend to be steeper, generally resulting in greater erosion (hence lower NFR score) and narrower floodplains (hence lower CFP, FPR and FSH scores) for lower-order streams.

Although we believe that the low NFR, CFP, FPR and FSH scores in the zero-order streams were due to their high position in the catchment, we found little difference in the ecological functions between zero-order and first-order streams in this study. Among twelve ecological functions in two topographic areas, the only one function (natural flow regime) in Pokeno showed a significant difference between zero-order and first-order streams. The reason for this lack of significant difference is probably that the first-order sites in this study were quite close to the zero-order sites. The native forest sites in Rowe et al. (2008), while still low-order, were probably further from the stream source than the first-order sites in this study, explaining the greater development of flood plains, higher flood frequency and lower flood-induced bank erosion.

A number of overseas studies have stressed the closer ecological connections between terrestrial and aquatic habitats in headwaters compared to higher-order streams (e.g., Wipfli et al. 2007, Freeman et al. 2007, Meyer et al. 2007). There may be several reasons why such differences did not appear in our comparison of zero-order and first-order sites. First, some of the nb-SEV functions focus on the effects of development, particularly urban development, and would not be expected to differ between zero-order and higher-order streams. Some functions, such as depth, canopy cover, diversity of flow types and instream particle retention, would be expected to differ between zero-order and higher order streams, but in our study the difference between zero-order and first-order streams was too small to affect the scores of these functions. Note that in many published studies, headwaters include first-order and even second-order streams (Freeman et al. 2007), therefore comparisons in these studies are at a different scale. Finally, some functions of headwater streams, such as the amount of stream bed organic matter, that might be expected to differ between zero- and first-order streams are not expressed strongly in nb-SEV, and some, such as refuge for aquatic invertebrates, are not included at all. Therefore, although our study showed no significant differences in ecological functions between perennial zero-order and first-order streams, we advise caution in treating these as equivalent.

In this study the mean non-biological SEV scores of the zero-order native forest streams were not as high as expected compared with those in other land-use types. The mean scores of native forest streams were lowered by the functions NFR, CFP, FPR and FSH, which scored lower in native forest than in other land uses. This is probably an artefact reflecting the present pattern of land use. At both Pokeno and Whatawhata, native forest has remained mostly in steep-sided gullies whereas flatter land has been cleared for pasture. Steep gullies may be expected to show greater erosion from flooding (lowering NFR) and narrower flood plains (lowering CFP, FPR and FSH) than flat land. Therefore the lower scores of NFR, CFP, FPR and FSH in native forest are most likely due to site-specific factors and do not indicate that these functions improve if native vegetation is cleared.

Tables 12 and 7 summarise the biological values and ecological functions, respectively, of zero-order streams in different land-use types and topographic areas. The standard deviations of these values are moderately large because only three replicates were used for each estimate. Standard deviations could have been reduced by combining samples from lowland and steep hill country, as in many cases there were no significant differences between these. However, because certain combinations of land-use type and topographic area did show significant differences, we believe more accurate estimates of the values were achieved by keeping all land-use types and topographic areas separate.

4.3 Offsetting loss of biodiversity and ecological function in zero-order streams

If a zero-order stream is piped, infilled or the land-use stress in the catchment is increased, biodiversity values and ecological functions of the stream (the impact stream) will be lost or degraded. To offset this loss, a degraded stream, similar to the impact stream and as physically close to it as possible, should be remediated. Remediation means that the biodiversity values and ecological functions of the degraded stream should be improved through measures such as riparian planting. To determine the length of stream to be remediated, we recommend calculating an environmental compensation ratio according to the procedure outlined in Rowe et al. (2008). Note that although ecological functions may be predicted broadly from land-use type and topographic area (Table 7), an environmental compensation ratio for loss of a zero-order stream should not be derived by simply using the values in Table 7 for the corresponding land-use type. Rather, a full nb-SEV assessment, based on field-derived data, should be performed. Rowe et al. (2008) describe certain caveats and defaults for calculating ECR. We recommend these and add the following:

1. If the impact on a stream is an increase in land-use stress, the values in Table 7 can be used as a guide to predict the final ecological function values in the impacted stream. However, the scenario must be realistic. Although in Table 7 some function scores for native forest streams are lower than those for riparian forest or pasture (due to site specific factors), it is not expected that a reduction in forest cover in the catchment or riparian zone will cause the score of any function to increase.

2. Similarly, at the mitigation site, the values in Table 7 can be used as a guide to predict the final ecological function values of the remediated stream. Although in Table 7 some function scores for native forest are lower than those for riparian forest or pasture (due to site specific factors), it is not expected that an increase in forest cover in the catchment or riparian zone will cause any function score to decrease.

3. Spring-fed streams should be given high scores for water temperature control (WTC) and lower than average scores for flood frequency, due to the constancy of spring-fed streams with respect to temperature and flow.

4. To reduce the probability of a net loss in biodiversity values, we recommend calculating an additional environmental compensation ratio (ECR), based on a biodiversity metric, in a manner equivalent to the SEV. The same formula for ECR can be used, substituting EPT richness of the aquatic macro-invertebrate community in place of SEV score. If the impact is an increase in land-use stress, the values in Table 12 can be used as a guide to predict the final EPT richness in the impacted stream. The ECR used should be the greater of that based on SEV and that based on EPT richness.

5. Since our knowledge of zero-order streams is still partial, mitigation ideally should be “like for like”, i.e., a zero-order stream remediated for a zero-order stream lost or degraded. The data in this study indicate that remediation of a perennial first-order stream may be equivalent to remediation of a perennial zero-order stream as compensation for loss or degradation of a perennial zero-order stream. However, this conclusion is subject to several limitations discussed in Section 4.1 and 4.2. Further, since all the sites sampled in this study were perennial, we cannot extend our conclusions to non-perennial stream reaches. If the impacted site has intermittent flow (isolated pools and

wet mud) then the remediated site also should have intermittent flow. Similarly, if the impacted site has perennial flow, the remediated site also should have perennial flow.

6. The mitigation site should be as near as possible to other stream reaches that are in good ecological health, e.g., those that are surrounded by native forest. This is because the healthy stream reach will likely act as a source of aquatic invertebrates. Invertebrates will colonise the remediated reach via aerial colonisation or upstream movement and thus greatly increase the likelihood of an improvement in the aquatic biodiversity (Quinn et al. 2009). If a first-order mitigation site is used because no zero-order site can be found, we recommend locating the mitigation site downstream of a forested stream reach, if possible, since invertebrates colonise via downstream drift more readily than by aerial or upstream movement. In this situation, the remediation works ideally should extend to connect with the forested stream reach. If that is not possible, the length of poor-quality stream habitat between the remediated reach and the forested reach should be minimised.

5. Summary and conclusions

1. Macro-invertebrate communities were as diverse, or nearly as diverse, in zero-order streams as in first-order streams, though in steep country EPT richness was significantly lower. All major taxonomic groups were well-represented in zero-order streams. We did not find a consistent difference in community composition between zero-order and first-order streams. Though some taxa were found only in zero-order streams, they occurred rarely and we cannot discount the possibility that further sampling in first-order streams may reveal their presence there also. Similarly, some taxa found only in first-order streams may be found by further sampling in zero-order streams.

2. No difference was found in the performance of non-biological SEV functions in zero-order vs. first-order streams, except for greater bank erosion in lowland zero-order streams. Zero-order native forest streams in this study had lower nb-SEV scores than first- or second-order native forest streams in Rowe et al. (2008). This may reflect that zero-order tend to occur in steeper, more incised valleys than first- and second-order streams.

3. In zero-order sites, Shannon diversity, total richness and EPT richness of the benthic macroinvertebrate communities decreased between native forest and pasture. At Whatawhata there was a significant linear relationship between biodiversity metrics and land-use stress, but at Pokeno these linear relationships were not significant, as only pasture sites had lower biodiversity values.

4. Land-use stress explained 19% and 31% of variability in Shannon diversity at Pokeno and Whatawhata respectively; 10% and 52% of variability in total richness at Pokeno and Whatawhata respectively; and 21% and 50% of variability in EPT richness at Pokeno and Whatawhata respectively.

5. nb-SEV mean score was strongly correlated with land-use stress in both steep and low-topography country, across both zero-order and first-order sites. Lower scores in native forest streams than in pasture streams for some ecological functions were probably due to site-specific factors (in particular, steeper valleys) rather than a true difference between land-use types.

6. Land-use stress explained 41% and 76% of variability in non-biological SEV score at Pokeno and Whatawhata respectively, when these two areas of contrasting topography were analysed separately. When data from both

topographic types were combined, land-use stress explained 59% of variability in nb-SEV whereas topography explained only 4%. Stream order (zero-order vs. first-order) explained only 3% of variation in nb-SEV at Pokeno, and less than 1% at Whatawhata or both areas combined.

7. The biodiversity values and ecological functions expected for zero-order streams in different land-use types and topographic areas are summarised in Table 12. These values can be used as a guide for calculating future nb-SEV values of impacted and remediated sites if a zero-order stream is piped or land-use stress is increased. Our data suggest that either zero-order or first-order streams may be remediated as compensation for loss of a zero-order stream. However, zero-order streams may have values and functions not captured in this study, thus where possible a zero-order stream should be remediated as compensation for impacts to a zero-order stream. The mitigation site should be located as close as possible to healthy streams that can act as sources of invertebrate colonists.

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8. Appendix 1

Individual ecological function scores (±1 standard deviation) of zero-order and first-order streams in low and steep topography of different land-use types. These are the values that would be lost if a zero-order stream were piped or infilled.

	Stream order	Land- use stress	NFR	CFP	CGW	WTC	DOM	OMI	IPR	DOP	FPR	FSH	HAF	RVI
Lowland	**Zero**	**NF**	0.77 ±0.14	0.45 ±0.23	1.00 ±0	0.76 ±0.04	1.00 ±0	0.84 ±0.16	1.00 ±0	0.89 ±0.1	0.54 ±0.22	0.35 ±0.23	0.97 ±0.04	0.96 ±0.08
		RFThick	1.00 ±0	0.40 ±0	1.00 ±0	0.78 ±0.02	0.38 ±0.21	0.88 ±0.11	1.00 ±0	0.90 ±0.05	0.41 ±0.09	0.31 ±0.17	0.76 ±0.04	0.86 ±0.13
		RFThin	0.90 ±0.09	0.30 ±0.09	1.00 ±0	0.71 ±0.02	0.74 ±0.31	0.67 ±0.07	1.00 ±0	1.00 ±0	0.29 ±0.07	0.40 ±0.2	0.70 ±0.13	0.54 ±0.05
		P	0.90 ±0.09	0.82 ±0.2	1.00 ±0	0.46 ±0.1	0.37 ±0.1	0.06 ±0.1	1.00 ±0	0.79 ±0.15	0.64 ±0.05	0.56 ±0.34	0.49 ±0.09	0.36 ±0.01
	First	**NF**	0.95 ±0.09	0.18 ±0.12	1.00 ±0	0.76 ±0.03	0.75 ±0.44	0.83 ±0.02	1.00 ±0	0.95 ±0.1	0.26 ±0.22	0.49 ±0.05	0.90 ±0.14	0.96 ±0.08
		RFThick	0.93 ±0.11	0.25 ±0	1.00 ±0	0.78 ±0.04	0.59 ±0.58	0.89 ±0.06	1.00 ±0	0.88 ±0.17	0.26 ±0.08	0.37 ±0.27	0.83 ±0.22	1.00 ±0
		RFThin	1.00 ±0	0.70 ±0.3	1.00 ±0	0.69 ±0.02	0.52 ±0.42	0.59 ±0.21	1.00 ±0	0.99 ±0.01	0.61 ±0.21	0.55 ±0.13	0.65 ±0.12	0.65 ±0.16
		P	1.00 ±0	0.95 ±0.09	1.00 ±0	0.43 ±0.05	0.49 ±0.22	0.11 ±0.1	1.00 ±0	0.92 ±0.13	0.76 ±0.12	0.72 ±0.25	0.43 ±0.03	0.36 ±0.01

	Stream order	Land- use stress	NFR	CFP	CGW	WTC	DOM	OMI	IPR	DOP	FPR	FSH	HAF	RVI
Steep hill country	**Zero**	**NF**	0.75 ±0.17	0.43 ±0.25	1.00 ±0	0.78 ±0.01	1.00 ±0	0.92 ±0.03	1.00 ±0	0.93 ±0.06	0.39 ±0.12	0.52 ±0.02	0.99 ±0.01	0.98 ±0.04
		RFThick	0.90 ±0.09	0.55 ±0.15	1.00 ±0	0.76 ±0.01	0.83 ±0.3	0.83 ±0.04	1.00 ±0	0.70 ±0.12	0.58 ±0.11	0.50 ±0.06	0.85 ±0.14	0.84 ±0.11
		RFThin	0.65 ±0.17	0.37 ±0.2	1.00 ±0	0.68 ±0.09	0.79 ±0.36	0.48 ±0.29	1.00 ±0	0.79 ±0.22	0.31 ±0.18	0.35 ±0.19	0.67 ±0.11	0.47 ±0.1
		P	0.72 ±0.25	0.62 ±0.19	1.00 ±0	0.56 ±0.12	0.80 ±0.35	0.24 ±0.25	1.00 ±0	0.69 ±0.29	0.45 ±0.11	0.44 ±0.18	0.53 ±0.08	0.42 ±0.1
	First	**NF**	0.90 ±0.09	0.58 ±0.29	1.00 ±0	0.77 ±0.01	1.00 ±0	0.92 ±0.03	1.00 ±0	0.92 ±0.14	0.54 ±0.25	0.57 ±0.05	0.99 ±0.01	1.00 ±0
		RFThick	0.93 ±0.11	0.45 ±0.07	1.00 ±0	0.76 ±0.03	0.61 ±0.55	0.85 ±0.13	1.00 ±0	1.00 ±0	0.37 ±0.1	0.35 ±0.25	0.82 ±0.09	0.87 ±0
		RFThin	0.65 ±0.17	0.38 ±0.42	1.00 ±0	0.64 ±0.07	0.80 ±0.35	0.49 ±0.37	1.00 ±0	0.59 ±0.15	0.33 ±0.33	0.31 ±0.22	0.67 ±0.15	0.45 ±0.1
		P	0.90 ±0.09	0.35 ±0.22	1.00 ±0	0.57 ±0.08	0.74 ±0.44	0.14 ±0.15	1.00 ±0	0.57 ±0.21	0.28 ±0.13	0.78 ±0.21	0.60 ±0.08	0.44 ±0.11