Influence of Human Pressures on Large River Structure and Function

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By K.J.Collier[1], J.E. Clapcott[2] and R.G. Young[2]

[1], Centre for Biodiversity and Ecology Research
Department of Biological Sciences
School of Science and Engineering
The University of Waikato
Private Bag 3105
Hamilton, New Zealand

Email: kcollier@waikato.ac.nz

[2], Cawthron Institute
Private Bag 2
Nelson, New Zealand

THE UNIVERSITY OF WAIKATO
Te Whare Wānanga o Waikato

CBER University of Waikato
Centre for Biodiversity and Ecology Research

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Reviewed by:

David P. Hamilton
University of Waikato
Affiliation

Approved for release by:

Brendan Hicks
University of Waikato
Affiliation

Executive summary

A large river study was conducted as part of the Cross Departmental Research Pool (CDRP) ecological integrity project to (i) provide an overview of the macroinvertebrate faunas of large rivers, including those in deep-water habitats, and (ii) to elucidate links between these faunas, river function and anthropogenic stressors. Eleven sites on 6th-order or 7th-order rivers were sampled; four in the South Island and seven in the North Island. We measured (i) macroinvertebrate communities colonising wood, riffles (where present), littoral habitats (<1 m deep) and deepwater habitats (>1.5 m deep) (ii) ecosystem metabolism using a single-station open-channel approach based on natural changes in dissolved oxygen concentration over a 24-hour period, and (iii) wood and cellulose breakdown. Relationships were investigated between these response variables and reach-scale assessments of habitat quality, underlying upstream and segment environmental variables provided in the Freshwater Environments of New Zealand (FWENZ) database, and anthropogenic pressure variables provided by the Waters of National Importance (WONI) database.

Sampling sites grouped together based primarily on location using FWENZ variables, reflecting spatial differences in underlying environmental pattern, whereas WONI pressure variables grouped sites with limited pressure together, and other sites according to different stressor profiles (e.g., nitrogen enrichment, impervious surfaces, dams, geothermal inputs, coal deposits). Sites with limited pressure also grouped together based on habitat quality components, with the amount of large wood evident in the channel, lateral connectivity and the prevalence of off-channel habitats driving separation along a secondary axis.

Macroinvertebrate percent abundance data revealed a distinction between Waikato sites and other sites irrespective of habitat sampled, reflecting the influence of low channel gradient and the absence of riffle habitat at all Waikato sites. Sites with riffles and minimal impact tended to support different macroinvertebrate community composition than impacted large river sites. Segment riparian shade and average-weighted natural cover appeared to be the main factors explaining macroinvertebrate community patterns, after taking account of whether riffle habitat was present or absent.

Natural vegetation cover was a significant predictor variable for Shannon diversity, whereas this variable along with total nitrogen concentration and probability of brown trout occurrence were significant predictor variables for the richness and relative abundance of sensitive aquatic insect groups (Ephemeroptera, Plecoptera and Trichoptera (excluding Hydroptilidae) - EPT). These metrics showed linear relationships with certain stressor variables rather than defined thresholds

for the sites sampled. Analysis of replicate deepwater and littoral samples for sites with riffles at the upper and lower ends of the land-use stressor gradient sampled suggested that non-EPT compositional measures (i.e., based on other insects, Crustacea, Mollusca, Oligochaeta) may provide assessments of impact response independently of habitat type.

Overall, macroinvertebrate communities in these large rivers appeared to respond to anthropogenic stressors in a similar way expected for smaller streams, although interpretation was limited by the number of sites sampled. While some conventional macroinvertebrate metrics, such as EPT richness and % EPT abundance, appeared to be strongly influenced by anthropogenic stressors, the Macroinvertebrate Community Index (MCI) did not appear to be as effective for large rivers where multiple habitats were sampled and riffles were sometimes absent. Our results suggest that sampling of multiple habitats was required to accurately document the biodiversity of large river macroinvertebrate communities, and that metrics derived from groups more common in large river environments (e.g., Crustacea) may provide useful additions to other metrics for documenting large river health. Insufficient functional data were available to draw strong conclusions about the relationship between structural and functional metrics in large rivers.

1. Introduction

Large rivers are the ultimate integrators of upstream activities, yet, compared to wadeable streams, little is known about the ways their benthic biology or functional processes respond to anthropogenic stressors. 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. Distinctive features of large river environments include transverse asymmetry across the channel (Bournaud *et al*. 1998), the occurrence of islands, side-arms and large tributary junctions (Thorp, 1992; Cellot, 1996; Kiffney *et al*. 2006), and extensive floodplain areas that periodically provide habitat (Puckridge *et al*. 1998; Tockner and Stanford 2002). This diversity of habitats suggests that a range of sampling methods is necessary to document the biological diversity of large river environments. Other studies have demonstrated that such environments can be characterised by distinct species assemblages (Bournard *et al*. 1998; deDrago 2004; Strayer *et al*. 2006). Assessing river processes can involve measuring rates at specific habitats (Bott *et al.* 1985; Fellows *et al*. 2009) or of the whole ecosystem (Young *et al.* 1996; Uehlinger 2006).

Large rivers have undergone significant modification globally over the last few hundred years, often brought about by a combination of large-scale de-snagging operations to facilitate navigation (Harmon *et al.* 1986), impoundment and flow regulation for hydroelectricity generation (Ligon *et al.* 1995), truncation of floodplain interactions for flood control purposes (Bayley 1991; Kroon and Ansell 2006), and the spread of alien species that often proliferate in these environments (Thorp and Casper 2003; Tempero *et al*. 2006). There is increasing interest from resource managers and society in the ecological condition 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 al. 2005; Flotemersch *et al.* 2006) and their ecosystem processes (e.g., Gawne *et al.* 2007; Fellows *et al.* 2009).

The large river study, conducted as part of the Cross Departmental Research Pool (CDRP) ecological integrity project, was intended to provide an overview of the macroinvertebrate faunas of large rivers, including those in deep-water habitats, and to elucidate links between these, river function and anthropogenic stressors. For the purposes of this study, a large river is defined as 6^{th} order or larger, and we selected sampling sites from the top 60 rivers nationally in terms of catchment size. Specific aims of the CDRP large rivers study were to:

1. Investigate responses of macroinvertebrate communities to stressor gradients in North and South Island sites relative to other factors, including underlying environmental variables and local habitat quality;
2. Determine differences in macroinvertebrate community composition and structure between habitats of a selection of New Zealand's largest rivers, including littoral (within 5 m of river's edge, <1 m deep) and deepwater (>5 m from edge, 1-6 m deep) zones;
3. Evaluate the response of some of the commonly used macroinvertebrate metrics developed for wadeable rivers in relation to stressor gradients in large river environments; and
4. Measure river metabolism parameters and cellulose and wood decomposition, and define relationships between these, the prevailing stressor gradient and macroinvertebrate community indices.

2. Methods

Sampling sites

Eleven sites on 6th-order or 7th-order rivers were sampled as part of this study; four in the South Island and seven in the North Island (Fig. 1; Table 1). These included three sites draining predominantly native forested catchments with limited pressures (Motu, Karamea and Mokihinui) to serve as benchmarks against which to compare other sites in terms of upstream land cover. Although some mining activity apparently occurred in the Mokihinui catchment according to the Waters of National Importance (WONI) pressure dataset, there is little evidence of acid mine drainage as can occur for other rivers of this region (see Floeder and Spigel 2008). All sites were outside of the tidal influence except, as it transpired, for the Mokau site which was influenced by tidally-induced water level variation during sampling. Most sites had relatively unmodified flow regimes except for the Waikato River site.

Summary statistics for selected variables from the Freshwater Environments of New Zealand (FWENZ) and WONI datasets for these sampling sites were compared with national data from 6th- to 8th-order rivers compiled from available regional and central government agencies, research institutes and universities (Scarsbrook 2008). Of these 572 river reaches, 186 had invertebrate presence/absence data associated with them. Although the method of collection and habitats sampled at these sites is not known, it is probable that sampling was done only to wadeable depth using Surber nets or kick nets. Generally, the sites sampled as part of the present study were similar to the national dataset sites in terms of flow statistics, slope, summer air temperatures, upstream geology (excluding peat), and local habitat and

sediment conditions (Table 2). Average minimum daily air temperature, shade levels, nitrogen concentrations, proportion of catchment in native vegetation, and upstream lake and peat influences were markedly (>20%) lower than the national dataset average, whereas upstream rain days and the proportion of upstream indigenous forest were higher. In terms of the WONI variables, impervious area and effects of dams, coal, geothermal inputs and mines were on average higher in the sampled dataset, predicted total nitrogen concentrations and the probability of brown trout capture were lower and natural cover and summary pressure variables appeared similar (Table 2).

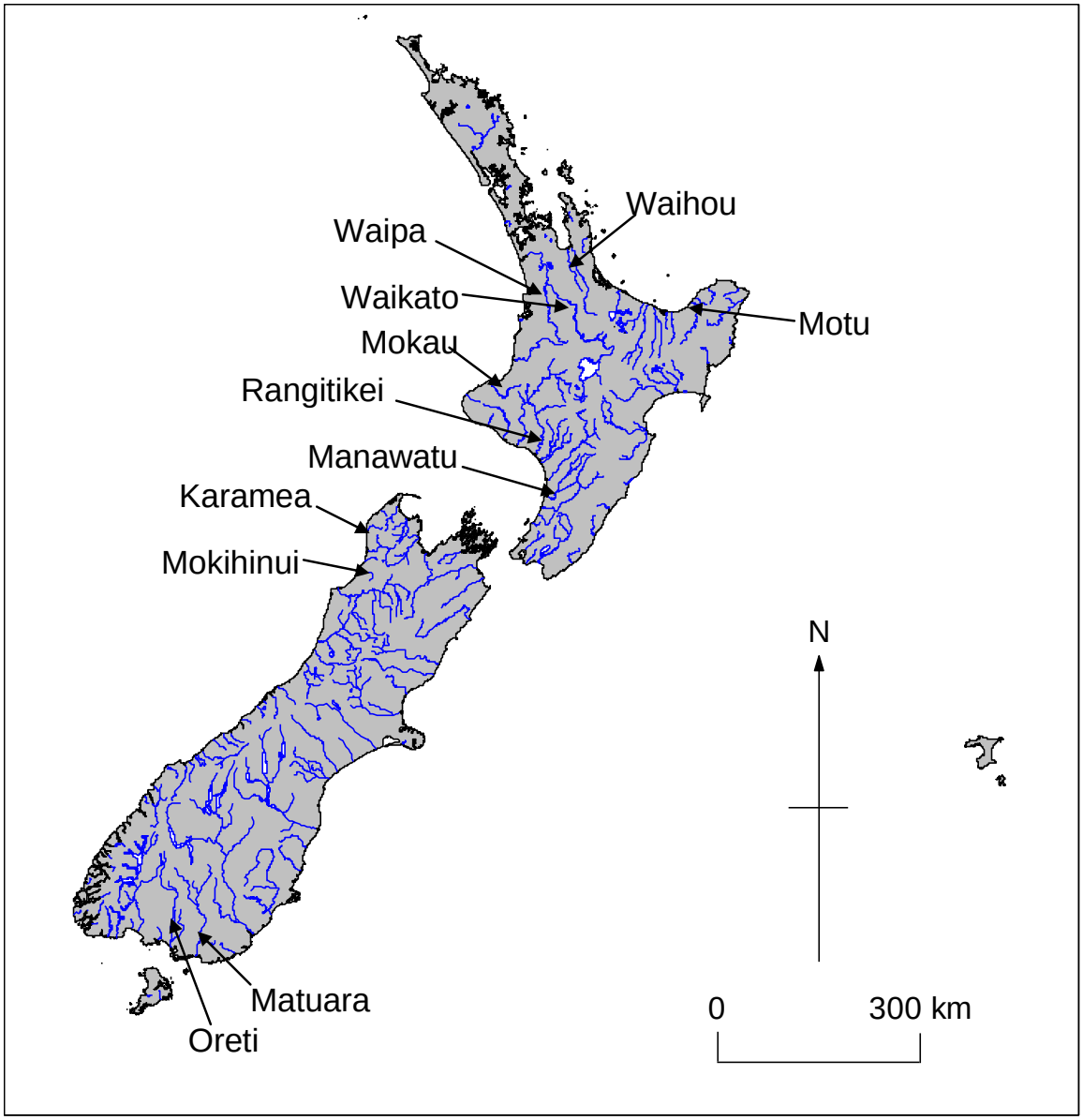


Figure 1: Location of large river sampling sites on 6th-and 7th-order rivers. The Waihou, Waipa, Waikato and Mokau sites were on low gradient rivers without riffles, whereas riffles were present at the other sites.

Table 1: Locations and sampling times of the large river sites.

Sites	Location	NZReachID	Easting	Northing	Date sampled	Riffle sample	Comments
North Island							
Motu	SH Bridge	4005116	2917653	6360615	16/11/2007	11/02/2008	
Manawatu	Opiki	7041173	2719425	6082635	27/11/2007	28/02/2008	
Rangitikei	Bulls	7035612	6110479	2713541	26/11/2007	28/02/2008	*n* = 4 for airlift and littoral
Mokau	Awakau Rd	3043108	2660012	6238601	22/11/2007	NA	Tidally-influenced
Waipa	Ngaruawahia	3015397	2699520	6389554	19/11/2007	NA	No airlift samples (too deep)
Waikato	Botanic Gardens	3018187	2713160	6374738	20/11/2007	NA	
Waihou	Te Aroha	3013115	2749995	6402365	21/11/2007	NA	
South Island							
Mataura	Seaward Downs	15059190	2186653	5415962	8/12/2007	21/12//07	
Oreti	Lumsden Cableway	15058642	2145685	5422805	7/12/2007	21/12/2007	
Mokihinui	Seddonville	12006282	2424396	5962351	13/12/2007	13/12/2007	*n* = 3 for airlift and littoral
Karamea	Arapito	12003364	2438964	5993157	12/12/2007	20/12/2007	

Table 2: Summary statistics for the national large rivers dataset (6^{th} - 8^{th} order; n = 573) compiled as part of the CDRP project from the Freshwater Environments of New Zealand (FWENZ) and Waters of National Importance (WONI) datasets and the large rivers sampled as part of this study (n = 11). % difference is derived from differences in means between this dataset and the national dataset divided by the national dataset value (negative values indicate sampled mean lower than national dataset; differences >20% in bold). See Appendices 1 and 2 for key to variable abbreviations.

Variable	National large river data set						Large rivers sampled						% diff.
	Mean	50th-%ile	70th-%ile	90th-%ile	Min.	Max.	Mean	50th-%ile	70th-%ile	90th-%ile	Min.	Max.	(mean)
FWENZ													
SEGFLOW	84.51	37.62	74.72	245.71	0.04	576.94	94.15	81.23	95.06	122.96	40.39	243.73	11
SEGLOWFLOW	25.85	8.72	17.80	87.18	0.01	279.41	23.77	15.99	21.57	32.83	4.65	99.20	-8
SEGLFLOW4T	1.90	1.77	2.08	3.06	1.00	4.09	2.10	2.03	2.18	2.41	1.54	3.16	11
SEGFLOWSTA	0.24	0.21	0.29	0.41	0.01	0.49	0.22	0.21	0.27	0.30	0.10	0.41	-5
SEGJANAIRT	16.72	17.00	17.50	18.60	12.80	19.60	17.18	17.40	18.20	18.70	14.00	18.80	3
SEGMINTNOR	-0.31	-0.11	0.55	1.17	-4.20	3.26	0.77	0.67	0.81	1.66	0.17	1.79	**-345**
SEGRIPSHAD	0.04	0.01	0.03	0.10	0.00	0.67	0.02	0.00	0.00	0.08	0.00	0.15	**-42**
SEGSLOPESQ	1.12	1.06	1.14	1.31	1.00	2.86	1.03	1.02	1.04	1.05	1.00	1.14	-8
SEGLOGN	-0.33	-0.31	-0.08	0.14	-1.32	0.83	-0.09	-0.02	0.21	0.25	-0.79	0.31	**-72**
USDAYSRAIN	16.56	13.00	18.02	31.20	1.60	101.40	21.09	15.79	19.41	59.10	3.70	60.40	**27**
USAVGTNORM	-2.43	-2.22	-1.43	-0.29	-7.75	1.33	-1.53	-1.22	-0.36	0.05	-3.60	0.55	**-37**
USAVGSLOPE	17.92	17.32	22.59	28.16	2.38	32.01	16.05	13.21	17.77	26.45	7.84	29.47	-10
USNATIVE	0.58	0.64	0.80	0.91	0.04	1.00	0.46	0.36	0.43	0.99	0.19	0.99	**-21**
USINDIGFOR	0.21	0.14	0.27	0.57	0.00	0.99	0.31	0.16	0.23	0.78	0.06	0.83	**47**
USCALCIUM	1.45	1.41	1.67	1.98	0.58	2.45	1.51	1.61	1.64	1.81	1.03	1.98	4
USPHOSPHOR	2.38	2.40	3.00	3.12	1.03	4.26	2.07	1.93	2.05	3.11	1.45	3.24	-13
USHARDNESS	3.15	3.22	3.53	3.82	1.65	4.27	2.98	2.78	3.04	3.79	2.25	4.22	-5
USPEATPC	0.00	0.00	0.00	0.01	0.00	0.35	0.00	0.00	0.00	0.00	0.00	0.02	**-30**
USLAKEPC	0.02	0.00	0.00	0.08	0.00	0.25	0.01	0.00	0.00	0.00	0.00	0.08	**-60**
LOCSED	3.87	4.00	4.20	4.60	1.00	5.30	3.56	3.70	4.00	4.10	2.20	4.30	-8
LOCHAB	4.08	4.10	4.30	4.50	2.00	5.20	3.92	3.90	4.10	4.20	3.40	4.20	-4

Variable	National large river data set						Large rivers sampled						% diff.
	Mean	50th-%ile	70th-%ile	90th-%ile	Min.	Max.	Mean	50th-%ile	70th-%ile	90th-%ile	Min.	Max.	(mean)
WONI													
A_WT_IMPER	0.00	0.01	0.02	0.03	-9.99	0.56	0.02	0.02	0.03	0.03	0.00	0.03	**862**
A_WT_NATCO	0.52	0.53	0.70	0.88	0.00	1.00	0.47	0.37	0.47	0.99	0.19	0.99	-10
LOGNCONC	-0.33	-0.31	-0.08	0.14	-1.32	0.83	-0.09	-0.02	0.21	0.25	-0.79	0.31	**-72**
DAMEFFECT	0.63	0.01	0.30	1.55	0.00	9.41	0.91	0.00	0.15	0.48	0.00	9.13	**43**
COALEFFECT	0.13	0.00	0.00	0.04	0.00	4.67	0.54	0.00	0.00	1.95	0.00	3.96	**306**
GEOTHEFFEC	0.05	0.00	0.00	0.00	0.00	1.84	0.11	0.00	0.00	0.00	0.00	1.17	**97**
MINEFFECT	0.04	0.00	0.00	0.03	0.00	1.33	0.09	0.00	0.00	0.03	0.00	0.99	**115**
SALTRU_RES	0.46	0.43	0.69	0.90	0.00	0.99	0.26	0.26	0.35	0.49	0.02	0.58	**-44**
AVERAGE_SU	0.39	0.38	0.49	0.63	0.00	0.93	0.45	0.42	0.52	0.67	0.11	0.79	16
MIN_SUM	0.24	0.21	0.31	0.51	0.00	0.74	0.21	0.17	0.23	0.59	0.00	0.68	-12

Macroinvertebrate communities

Macroinvertebrates were collected from 2 - 4 habitat types, depending on availability, over an approximately 500-m long reach (except Rangitikei where shallow water limited boat access). Wood samples consisted of a composite collection from five pieces of wood brushed into a D-frame net (0.5 mm mesh) at all sites. Riffles were present at seven sites and were sampled by turning over and brushing stones upstream of the net. Replicate paired air-lift and littoral samples (n = 3 - 5, number depending on availability of suitable habitat; see Table 1) were collected at approximately every 100 m along the sampling reach. Littoral sampling involved sweeping and brushing accessible substrates along river edges into the D-frame net. A coin was tossed to randomly determine which bank was sampled at each transect. The airlift sampler was deployed from a boat in deeper water (>1.5 m deep); it consisted of sections of tube (10.5 cm internal diameter) linked together to the desired depth with a slanted top section covered by 0.5 mm mesh metal netting at the top end and a sample bag (0.5 mm mesh) at the bottom end. Compressed air was forced from dive tanks through hoses which vented at the bottom of the tube after it was lowered onto the riverbed. The tube was held in place by ropes attached to a collar at the bottom of the tube; these ropes were used to hold the tube vertically and move it up and down, with the help of an operator holding the upper section, to dislodge bottom material which was then caught by the rising bubble and belched into the sample bag. This method sampled sand-gravel substrates effectively, but had limited effectiveness on cobbles, boulders and bedrock (although many taxa were still obtained from coarse substrates; see Results).

Macroinvertebrate samples were preserved in c.70% isopropyl alcohol. Processing consisted of spreading the sample across a white tray, and randomly selecting grid squares from which invertebrates were picked. Grid squares were processed sequentially until ≥200 invertebrates were obtained or the entire sample had been processed. Unprocessed parts of the sample were searched for additional unrecorded taxa. A range of diversity (Margalef, Simpson, Shannon, total number of taxa, rarified number of taxa), evenness (Pielou), compositional (EPT (Ephemeroptera, Plecoptera and Trichoptera excluding Hydroptilidae) taxa richness and percent abundance) and tolerance (Macroinvertebrate Community Index; MCI) metrics were calculated from the macroinvertebrate data.

Functional indicators

Recent work has provided a methodology for including functional indicators into holistic assessments of stream integrity (e.g., Young *et al*. 2008), and such indicators now form part of regular river health assessments in Australia (Fellows *et al*. 2006). Functional indicators such as river metabolism and organic matter breakdown rates measure ecosystem processes and therefore complement structural indicators such as macroinvertebrate community metrics. Recent work in New Zealand suggests that functional indicators respond to increasing land-use pressures (Clapcott *et al*. 2009).

Two different organic substrates were deployed at each site to assess decomposition rates; pre-weighed birchwood (*Betula platyphylla* Sukaczev) coffee stirrer sticks and strips of cotton cloth. Rates of decomposition provide a measure of the potential for rivers to provide valuable ecosystem services, such as organic matter and nutrient processing. Stick assays were conducted as described in Clapcott *et al*. (2009). Briefly, sticks were weighed and five groups of five sticks were deployed at each site for three months. Each group was weighed down to keep it submerged close to the river bed. Following retrieval, sticks were gently washed and re-weighed. A set of control sticks was oven-dried to determine the proportional difference between air-dry weight and oven-dry weight, which averaged 90% (range 89 - 90%). This correction factor was used to estimate initial oven-dry weights for the sticks that were deployed. Exponential decay coefficients for wood were determined using the equation presented in Petersen and Cummins (1974) with degree days as the time variable (k_{dd}). Water temperature was recorded every 15 minutes throughout the three month deployment period by a Hobo pendant logger (Onset, Massachusetts, USA) at each site.

To measure cellulose decomposition potential, a cotton strip assay was conducted as described in Clapcott *et al*. (2009). Briefly, five replicate strips of unbleached cotton fabric were deployed at each site for seven days. Each strip was attached by nylon thread to a metal stake and weighed down close to the stream bed. Following retrieval, cotton strips were gently washed and dried, frayed to a width of 35 mm (100 threads), and the tensile strength (kg) of each length of strip was measured on a motorised tensometer (Sundoo, Whenzhou, China). The initial tensile strength of the strips was determined using a set of control strips that were soaked in tap water for one day, and then frozen and processed in the same way as the other strips. The loss of tensile strength was reported in terms of exponential decay coefficients in the same way as the wooden stick data.

Ecosystem metabolism is a measure of how much carbon is produced and consumed in river ecosystems, and provides an indication of how "well-balanced" a river is, especially in terms

of supporting river food webs. The combination of primary production and ecosystem respiration was estimated using the single-station open-channel approach which requires measurement of the natural changes in dissolved oxygen concentration at the site over at least a 24-hour period (Owens 1974; Young and Huryn 1996). Oxygen concentration and temperature were recorded once every 15 minutes using a D-Opto logger (Zebra-tech, Nelson, New Zealand) attached to a metal stake and deployed in the thalweg at each site. Light recordings provide an indication of day length for the calculation of metabolism and were gathered using an Odyssey light logger (Dataflow, Christchurch, New Zealand), set to record photoactive radiation every 15 minutes and also attached to the metal stake. Average depth upstream of sampling sites was calculated using at least five measurements of depth at each of five cross-sections spaced at regular intervals upstream of the stake to cover the local variation in channel morphology.

Metabolism values were calculated using a spreadsheet model described in Young and Collier (2009). Briefly, mean daily ecosystem respiration (ER), re-aeration coefficient (*k*) and the oxygen deficit (D) were determined using the night-time regression method (Owens 1974). These values were then used to determine gross photosynthetic rate over the sampling interval using:

$$GPP_t = dO/dt + ER - kD$$

where: GPP_t is the gross photosynthetic rate (g O_2 m^{-3} s^{-1}) over the time interval t of measurement (every 15 minutes). Daily gross primary production (GPP) was estimated as the integral of all temperature-corrected photosynthetic rates during daylight (g O_2 m^{-3} d^{-1}) (Wiley *et al.* 1990). Areal estimates (g O_2 m^{-2} d^{-1}) were obtained by multiplying the volume based estimates by average reach depth (m) which allowed comparison among sites with different depths.

Habitat measurements

A qualitative assessment of river habitat condition (Habitat score) was made by scoring 10 attributes along the 500-m sampling reach on a scale of 1 to 20 (1 lowest quality; 20 highest quality): lateral connectivity, off-channel habitats (such as side-arms, backwaters, connected wetlands etc), riparian vegetation composition, riparian vegetation width in terms of buffering from surrounding land use, bank stability, large wood abundance, submerged macrophyte cover, fine sediment deposition, mid-river substrate size, and turbidity. Water depth was measured at airlift sampling points using a hand-held depth sounder (Speedtech Depthmate), and estimated at littoral sampling points. The proportion of substrates sampled for each

littoral sample was also estimated according to seven inorganic particle sizes (bedrock – silt), and the organic categories of wood, roots and macrophytes (see Table 3).

Statistical analysis

Separate principal component analyses (PCA) were conducted in *PC-ORD v.6* on transformed (arc sine square-root, log or log x+1) segment, upstream and local FWENZ variables (downstream variables were excluded as they have little bearing on macroinvertebrate distribution), as well as site eastings and northings, WONI pressure variables, and habitat variables. The variables SegLogN, SegRipShade, USIndigFor and USNative (see Appendix 1) were retained in the FWENZ analysis, although it is recognised that these variables are more likely to represent anthropogenic pressures rather than underlying conditions. The PCA used a cross-products correlation matrix and distance based biplots for calculating variable scores. Macroinvertebrate community data were converted to percent abundances (deep and littoral replicates combined). Species recorded as "rare" (i.e., not in the initial 200+ count) were allocated a value of 0.5 before conversion to percentage. Data were analysed using non-metric multidimensional scaling with a Bray-Curtis similarity matrix following 4^{th}-root transformation (*Primer-E v.6.1.2*).

Biota and/or Environmental Matching was conducted under the BEST routine in *Primer* using the BV-step algorithm which conducts a stepwise search over all trial variables. This method uses a normalised Euclidean distance matrix to select environmental variables "best explaining" community pattern, by maximising the Spearman rank correlation between their respective resemblance matrices. Three analyses were conducted; firstly including all variables (excluding the summary pressure variables *Min_Sum* and *Av_Sum*), secondly with the WONI pressure variables forced into the analysis, and thirdly with habitat variables along with FWENZ and WONI variables. Gradient (high or low reflecting the presence or absence of riffles) and habitat type sampled (littoral, deep, wood, riffle) were included as dummy variables.

Spearman rank correlation analyses were used to explore relationships between macroinvertebrate metrics and functional indicators. Spearman rank was also used to explore relationships between macroinvertebrate community metrics, with r_s = 0.7 being used as a cut-off to identify highly correlated metrics. Regression trees were then used to explore the influence of environmental factors on key metrics showing least redundancy. This analysis was run with the TREES routine in *Systat v.11* using the least squares loss function. The maximum number of splits was set at 22 with a minimum count of three allowed at any node. The minimum proportion reduction in error for the tree allowed at any split and the minimum

split value at any node were set at 0.05. TREES analysis was run using all variables (FWENZ, WONI, habitat), along with dummy variables. TREES analysis was also run on the national large river compilation dataset for MCI, total taxa richness and percent EPT richness, allowing a maximum for four splits. To enable comparisons with the large rivers sampled as part of the present study, these metrics were calculated from combined substrate samples (excluding the Waipa site where only two habitats could be sampled).

Stepwise linear regression analyses were performed using metrics derived from combined wood, littoral and deepwater samples (all sites excluding Waipa) as the dependent variable, and Habitat score, *Natcover*, *LogN* and *Imperv* as the independent variables (all log transformed) with gradient (high or low) as a dummy variable. Highly skewed variables (i.e., those with a large number of zero values such as *Coaleffect*, *Mineeffect* etc.) were omitted from the analysis as they were not normally distributed. The probability for inclusion was set at $P < 0.05$

Finally, General Linear Models (GLM; *Systat v.11.1*) were used to compare least impacted sites with riffles (Karamea, Mokihinui and Motu; *LogN* < -0.3) with most impacted sites with riffles (Manawatu, Oreti, Mataura; *LogN* > 0.16) using ANOVA on replicate littoral and deepwater samples ($n = 5$ except for Mokihinui where $n = 3$). This analysis enabled us to explore the interaction between spatial variation (within and between habitats and sites) and two levels of anthropogenic impact independent of marked differences in river gradient. Dependent variables were selected to represent the composition of major invertebrate groups: (i) arcsine square-root transformed % abundance of EPT, non-EPT insects, Crustacea, Mollusca, Oligochaeta, and (ii) untransformed richness of total taxa, Trichoptera and Diptera. Main effects tested for were habitat (littoral, deep) and degree of impact (high, low), with the interaction between these variables used to determine whether responses to impact were different among habitats. Sites were also nested within degree of impact classes to determine how much variation was due to site differences.

Table 3: Depth, substrates sampled (littoral) and habitat scores for the 11 large river sampling sites. The maximum possible habitat score was 200.

	Airlift sample depth (m)	Littoral sample depth (m)	Littoral sample substrates (%)										Habitat score
			Bedrock	Boulder	Large cobble	Small Cobble	Gravel	Sand	Silt	Wood	Roots	Macrophyte	
North Island													
Motu	1.7	0.3	0	0	14	24	53	9	0	0	0	0	136
Manawatu	4.2	0.6	20	0	0	0	10	34	10	24	2	0	40
Rangitikei	1.7	0.6	0	13[1]	10	23	34	5	6	10	0	0	109
Mokau	3.5	0.7	0	0	0	0	0	0	22	24	2	52	58
Waipa	NA	0.6	0	0	0	0	0	0	32	30	30	8	52
Waikato	2.7	0.7	40	0	0	0	16	2	0	12	16	14	93
Waihou	1.7	0.7	0	0	0	0	0	1	21	3	9	66	49
South Island													
Mataura	1.6	0.4	0	0	0	10	25	3	16	0	6	40	92
Oreti	1.7	0.5	0	0	0	4	72	4	4	2	12	2	87
Mokihinui	1.6	0.4	0	13	60	20	3	3	0	0	0	0	146
Karamea	1.9	0.5	6	37	29	20	6	1	0	0	1	0	122

[1], concrete rip rap

3. Results

Environmental variables

The first two PCA axes of FWENZ, WONI and habitat variables explained 64 - 68% of the variation in the dataset (Table 4). The FWENZ ordination indicated spatial groupings driven primarily by location, with Westland, Southland and non-tidal Waikato sites occurring in close proximity to each other in 2-dimensional ordination space (Fig. 2). Westland sites were most strongly associated with *Natcover* (as was the Motu site) and upstream average slope and hardness of the underlying rock in the catchment, whereas most Waikato sites appeared related to upstream peat (Waihou, Waipa) or lake (Waikato) influences. Southland sites appeared to be influenced by phosphorus-bearing surface rocks (and inversely related to several hydrological variables), whereas the Manawatu and Mokau sites occurred along gradients apparently related to climate, calcium-bearing rocks in the catchment and local land-use intensity as indicated by SegLogN.

WONI pressure variables grouped sites with limited pressure to the left of axis 1. Sites apparently influenced by *LogN* or *Imperv* (reflecting urban development; but see Discussion) grouped towards the bottom right of the ordination, whereas the potential effect of dams, geothermal inputs and to a lesser extent coal deposits, contributed to the grouping of the Mataura and Waikato sites at the top right of the ordination. The Motu site was strongly associated with the summary variables of individual pressures (*Av_Sum* and *Min_Sum*), partly reflecting lower *Natcover* and intermediate *LogN* compared to the other sites with minimal pressure. The PCA of habitat score components also grouped sites with limited pressure to the left of the ordination along axis 1, with Waikato, Mataura, Rangitikei and Oreti intermediate, and Mokau, Waihou, Waipa and Manawatu to the far right. The distribution of sites along axis 2 appeared to be driven by the amount of large wood in the channel, lateral connectivity and the prevalence of off-channel habitats.

Table 4: Results of the Principal Components Analysis for FWENZ, WONI and habitat variables.

Axes	Eigenvalue	% of variance	Cum. % variance	Eigenvalue
		FWENZ + location		
1	8.508	38.671	38.671	3.691
2	6.081	27.639	66.310	2.691
3	2.992	13.599	79.908	2.191
		WONI		
1	3.955	39.551	39.551	2.929
2	2.460	24.596	64.147	1.929
3	1.884	18.845	82.992	1.429
		Habitat		
1	6.061	55.096	55.096	3.020
2	1.512	13.746	68.841	2.020
3	1.235	11.224	80.065	1.520

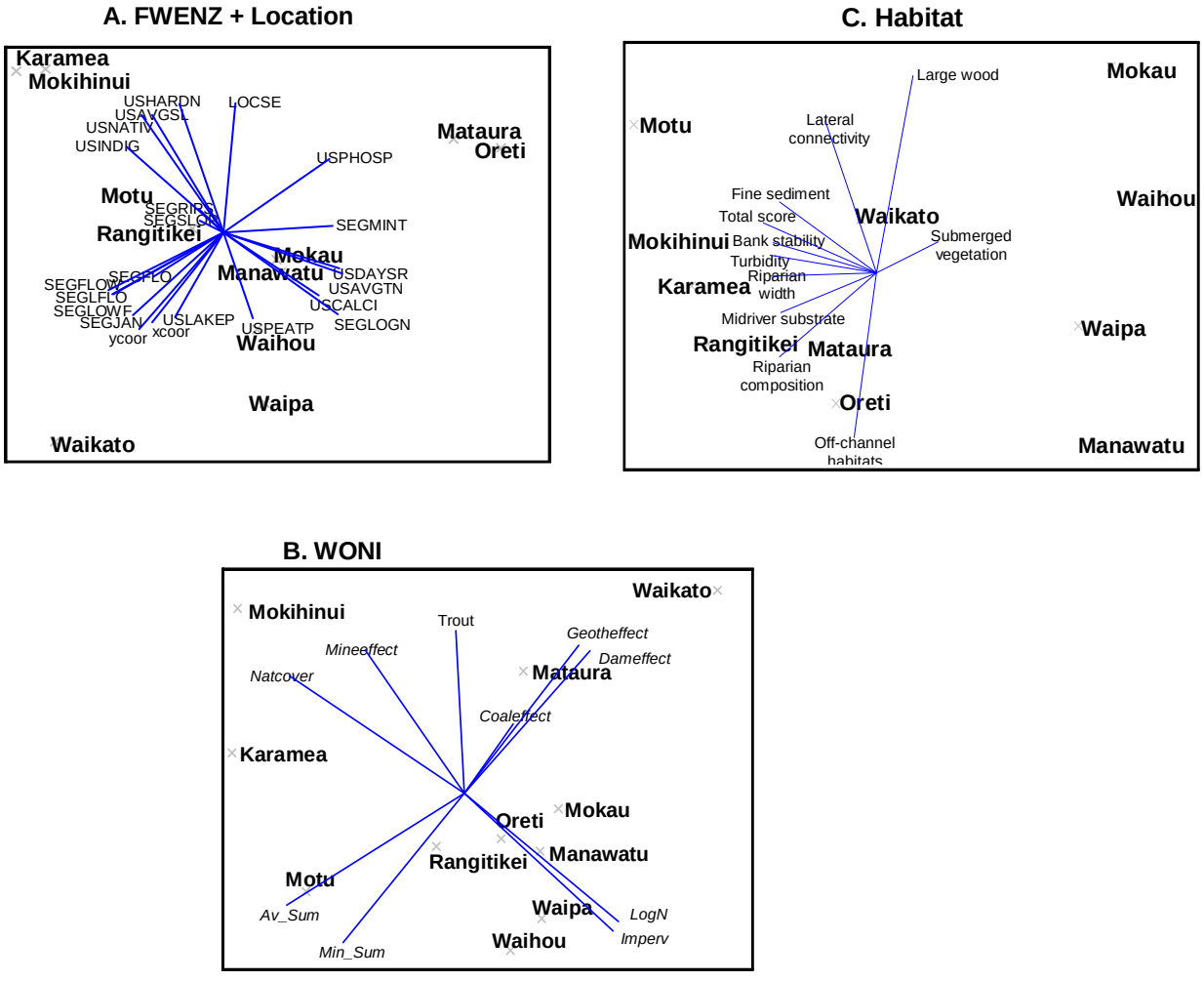


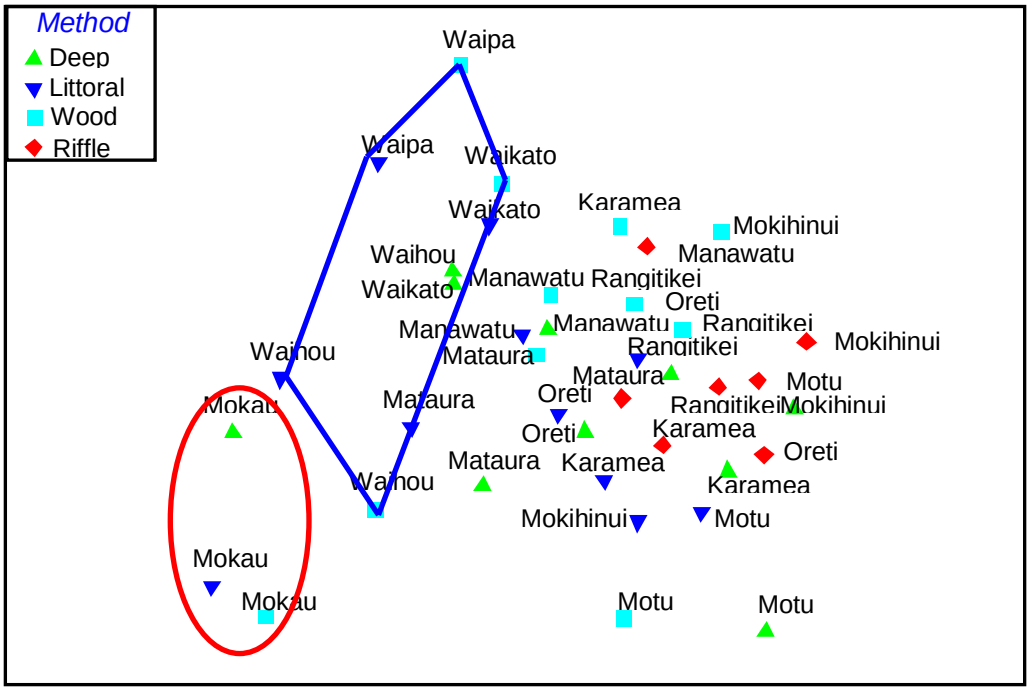
Figure 2: Principal component plots along axes 1 and 2 showing relationship between large river sampling sites (indicated by "X"; see also Table 3), and FWENZ, WONI and habitat variables. The longer the biplot lines, the stronger the relationship. See Appendices 1 and 2 for key to abbreviations of FWENZ and WONI variables.

Community composition

Non-metric multidimensional scaling ordinations of macroinvertebrate community percent abundance data indicated a distinction between Waikato sites on the left of the 2-dimensional ordination plot, and other sites irrespective of whether samples were taken from littoral, deep or wood habitats (riffles were not present at the Waikato sites) (Fig. 3A). A further separation along axis 2 occurred within the Waikato sites with all samples from the tidal Mokau River falling to the bottom left of the ordination. In contrast, riffle samples tended to occur towards the right of the ordination suggesting the distribution of sites along axis 1 was driven by channel gradient. When the low gradient Waikato sites were removed from the ordination, sites with minimal impact tended to cluster together irrespective of substrate type (Fig. 3B).

The best solution provided by the BV-Step analysis, with all sites and variables available, yielded a correlation coefficient of 0.72 using the dummy variable gradient (riffles present or absent, reflected in the separation of the Waikato sites), segment riparian shade (FWENZ) and *Natcover* (WONI). Adding in the qualitative assessments of local habitat increased the correlation coefficient from 0.72 to 0.75 with the following predictor variables: substrate type, gradient, segment riparian shade, upstream average slope, percent upstream peat in the catchment, local habitat (still, backwater, pool, run, riffle, rapid, cascade), *Saltru_res*, lateral connectivity, off-channel habitats, riparian vegetation composition, large wood abundance and mid-river substrate. When all the WONI pressure variables were forced into the analysis, the correlation coefficient reduced to 0.50 with the most parsimonious solutions provided by the WONI variables plus gradient, segment riparian shade, percentage of peat in the upstream catchment, and the reach-scale predictors of local habitat and local sediment (mud, sand, fine gravel, coarse gravel, cobble, boulder, bedrock).

A.



B.

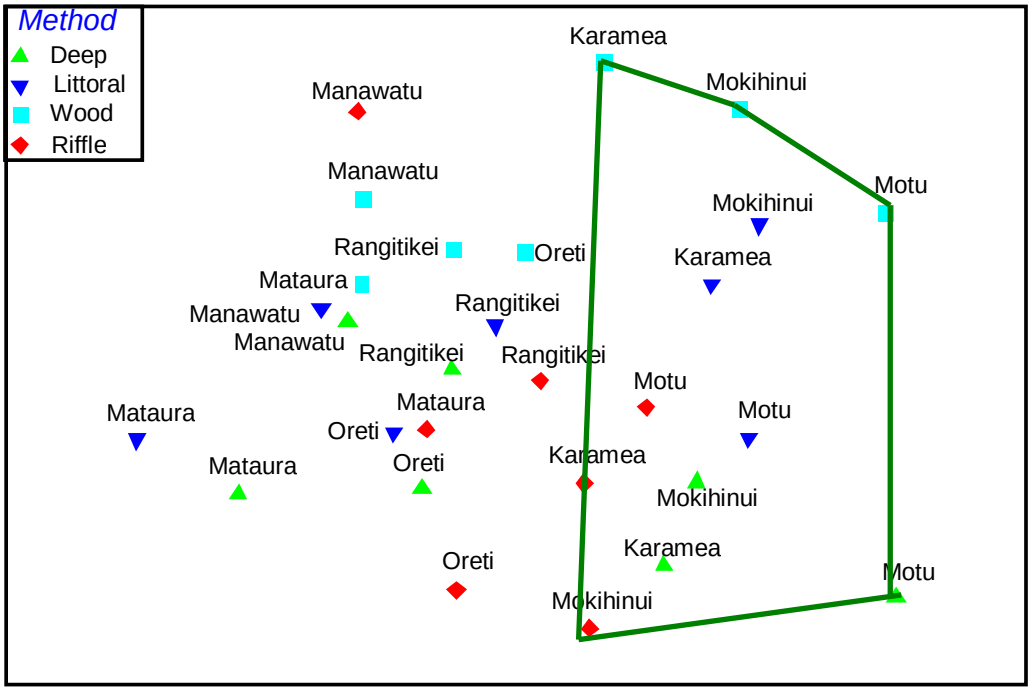


Figure 3: Non-metric multidimensional scaling plots of macroinvertebrate community percent abundance data for (A) all samples (ellipse envelopes tidal Mokau sites and polygon envelopes other low gradient Waikato sites), and (B) moderate gradient sites with riffles (polygon envelopes sites with minimal disturbance). Stress values = 0.19 (A) and 0.20 (B).

Macroinvertebrate metrics

Inter-relationships among the various diversity, compositional and tolerance metrics evaluated suggested that three metrics were providing distinct information, as indicated by rank intercorrelation coefficients (r_s) <0.7: Shannon diversity, EPT taxa richness and % EPT abundance (Table 5). MCI was also included in subsequent analyses to compare its utility. This metric was originally developed only for stony streams and may not be applicable to the non-stony substrates sampled. Although the soft-bottomed MCI was developed for this purpose in wadeable streams, its utility in large river settings is largely untested, and thus the original MCI was used to enable direct comparisons across all substrate types and sites.

Regression trees for these metrics suggested that only % EPT abundance was sensitive to habitat type with riffle samples splitting from littoral, wood and deepwater samples which appeared related to predicted local sediment conditions and the effect of dams (Fig. 4). EPT richness at the sampling sites was also associated with the effect of dams for both analyses, whereas second level splits identified the extent of phosphorus-bearing rocks. Shannon diversity appeared most strongly influenced by local habitat (runs vs. backwater and still habitats) followed by the extent of upstream native vegetation cover. MCI values were associated with the influence of lateral connectivity, followed by predicted segment low flow and upstream catchment area in peat (Fig. 4).

The TREES analysis of the national large river compilation suggested that flow and climatic variables were influencing taxa richness, whereas predicted upstream nitrogen concentrations and upstream natural cover were influencing % EPT richness, and two thresholds of natural vegetation cover were identified for MCI (Fig. 5). Comparisons with these metrics were made for all habitats combined for the sites sampled as part of the present study (excluding the Waipa where only two habitat types were sampled), with presence or absence of riffles as a dummy variable. The resulting TREES (data not shown) identified only one split for each metric, with weighted average of proportional cover of local habitat (still, backwater, pool, run, riffle, rapid, cascade) associated with taxa richness, upstream indigenous forest cover with % EPT richness, and upstream rain days with MCI.

Linear regression analyses indicated moderate to high explanatory power (adjusted multiple R^2 = 0.52 - 0.91) between three key metrics and pressure and habitat variables (Table 6). *Natcover* was a significant predictor variable for Shannon diversity, whereas this variable along with *LogN* and probability of brown trout occurrence were significant predictor variables for both EPT metrics. Habitat score was also significant for % EPT abundance which had the highest adjusted R^2 value of the three metrics, with highest coefficients for *Natcover* and *LogN* (Table 6). Regressions with MCI were not statistically significant.

Spatial variation in taxonomic groups

The GLM analysis provided insights into the significance of spatial scale based on replicate sampling of deepwater and littoral habitats for sites with riffles classified into contrasting impact classes. Degree of impact (low vs. high) had a significant effect on the composition and richness of major macroinvertebrate groups in littoral and deepwater habitats of these rivers (Appendix 3). In contrast habitat type (littoral vs. deep) had significant effects only for % Crustacea (representing greater dominance in littoral habitats), and total and Trichoptera richness (reflecting higher richness in littoral habitats). Habitat type responded differently to degree of impact for % EPT, and total, Trichoptera and Diptera richness suggesting that non-EPT compositional measures (other insects, Crustacea, Mollusca, Oligochaeta) may provide assessments of impact response independently of habitat type. Sites accounted for significant amounts of variability within impact groups for all compositional and richness measures, except for % Crustacea (Appendix 3).

Table 5: Spearman rank intercorrelation matrix among metrics evaluated reflecting macroinvertebrate community diversity, evenness, composition and tolerance. Bold values indicate r_s >0.7 which was used to define "strong" relationships.

	Margalef diversity (D)	Pielou evenness (J)	Rarified richness	Shannon diversity (*H*)	Simpson diversity	No. of taxa	EPT* richness	% EPT* richness	% EPT* abundance
Pielou evenness (J)	0.06								
Rarified richness (*n* = 127)	**0.81**	0.45							
Shannon diversity (*H*)	0.56	**0.79**	**0.82**						
Simpson diversity	0.38	**0.87**	0.68	**0.96**					
No. of taxa	**0.93**	-0.08	0.67	0.46	0.30				
EPT richness	**0.78**	0.08	0.68	0.49	0.39	**0.75**			
% EPT richness	0.30	0.16	0.34	0.25	0.22	0.19	**0.72**		
% EPT abundance	0.20	0.02	0.21	0.06	0.07	0.15	0.54	0.54	
MCI	0.42	0.15	0.49	0.34	0.30	0.32	**0.77**	**0.82**	0.51

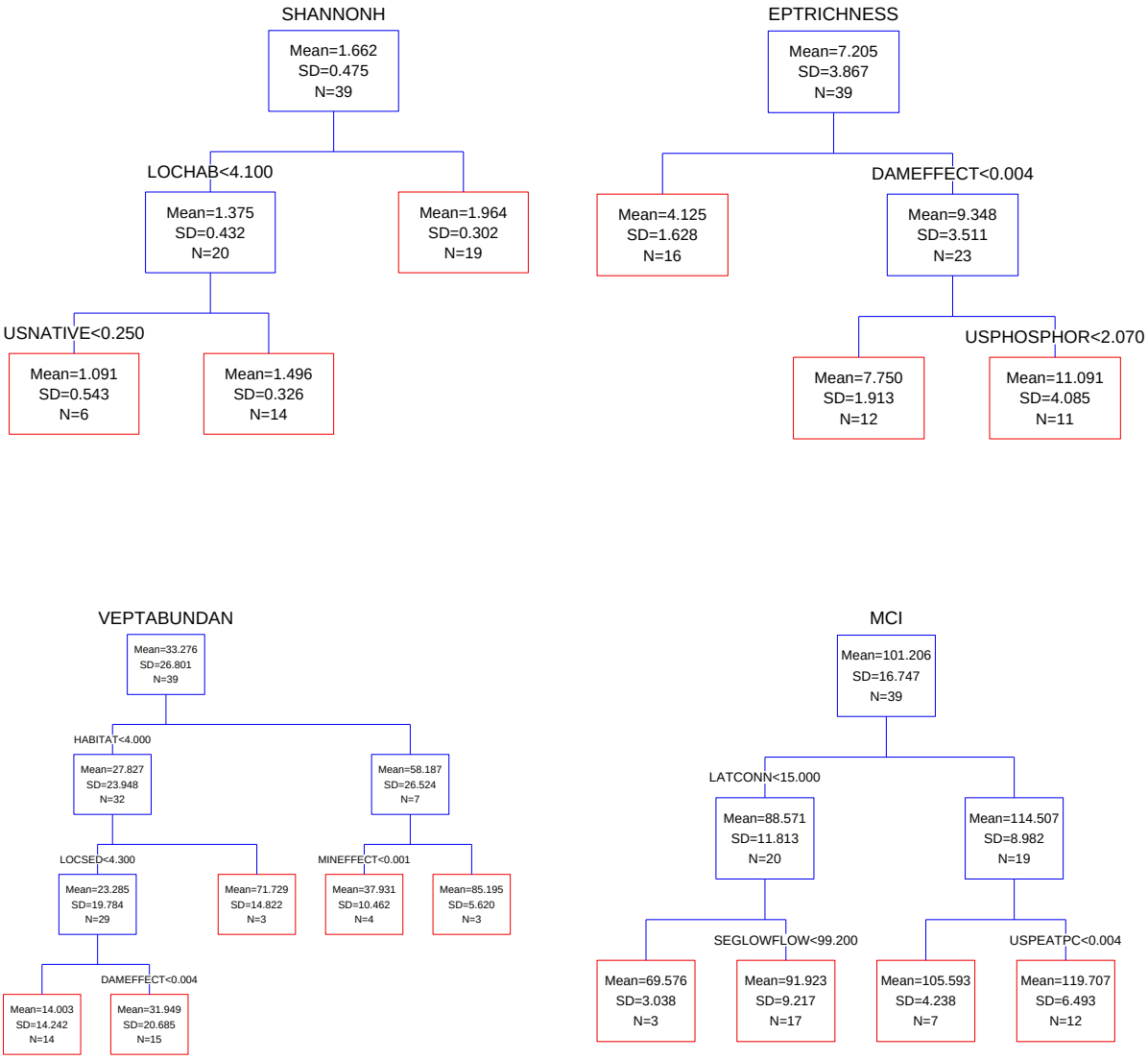


Figure 4: Regression TREES for key invertebrate metrics (Shannon diversity, EPT richness, % EPT abundance (VEPTABUNDAN)) and Macroinvertebrate Community Index (MCI) for individual habitat types from the present study using all FWENZ, WONI and habitat variables, with dummy variables for habitat type (riffle, littoral, deepwater, wood) and gradient (riffles present or absent).

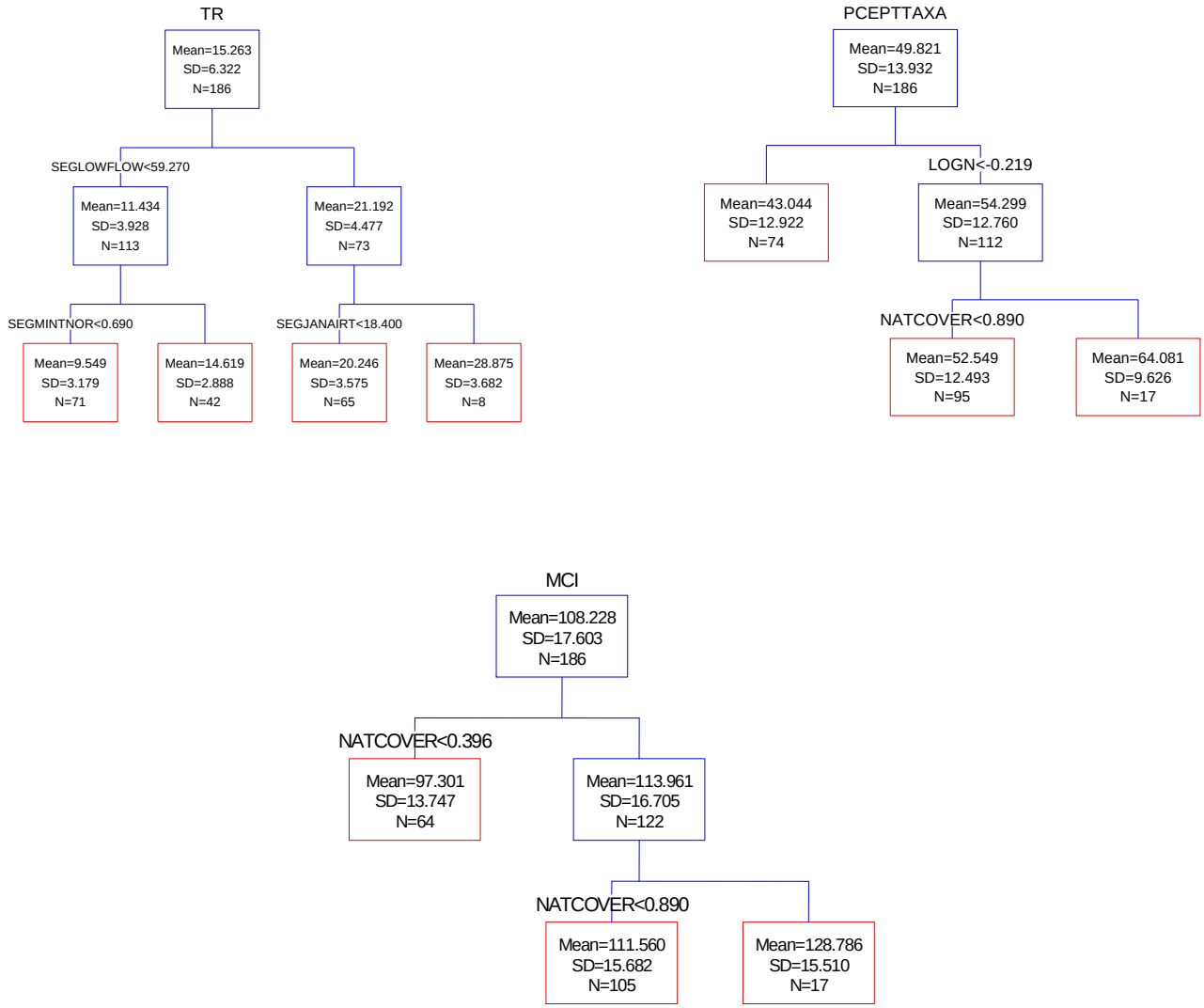


Figure 5: Regression TREES for invertebrate metrics calculated from presence/absence data in the national compilation of large rivers (*n* = 186) using stream order (6 - 8), and FWENZ and WONI (excluding summary pressure) variables.

Table 6: Linear regression analysis results for relationships between macroinvertebrate metrics from combined wood, littoral and deepwater samples, and selected pressure variables (n = 10). Regression analyses for MCI were not statistically significant. Waipa samples were omitted because no deepwater samples were collected there. See Appendix 1 for key to WONI abbreviations.

Effect	Coefficient	Std Error	Std Coef	t	P(2 Tail)
Shannon diversity: Multiple R: 0.759; Squared multiple R: 0.576; Adjusted squared multiple R: 0.523; Standard error of estimate: 0.064					
CONSTANT	0.148	0.050	0.000	2.939	0.019
Natcover	0.185	0.056	0.759	3.294	0.011
EPT richness: Multiple R: 0.942; Squared multiple R: 0.887; Adjusted squared multiple R: 0.831; Standard error of estimate: 0.100					
CONSTANT	0.215	0.198	0.000	1.088	0.319
Natcover	1.608	0.306	2.496	5.255	0.002
LogN	1.101	0.286	1.815	3.853	0.008
Saltru_res	-0.651	0.167	-0.594	-3.897	0.008
% EPT abundance: Multiple R: 0.974; Squared multiple R: 0.948; Adjusted squared multiple R: 0.906; Standard error of estimate: 0.106					
CONSTANT	-1.832	0.532	0.000	-3.444	0.018
Habitat score	1.018	0.324	0.552	3.147	0.025
Natcov	2.341	0.353	2.552	6.634	0.001
LogN	2.190	0.303	2.534	7.221	0.001
Saltru_res	-0.848	0.184	-0.543	-4.606	0.006

Relationships with functional indicators

Metabolism measurements of the large rivers assessed ranged from GPP of 1.07 g O_2 m^{-2} d^{-1} in the Rangitikei River to two orders of magnitude higher in the nearby Manawatu River, which also had the highest ER (Table 7). ER was low (<5 g O_2 m^{-2} d^{-1}) in the Motu, Waikato and Karamea Rivers. As expected stick decomposition was slower than cotton breakdown in rivers where both substrates were retrieved. Stick decomposition was lowest in the Manawatu and Rangitikei, followed by the Motu, Mokihinui and Mokau, and was highest in the Waikato. Cotton breakdown was lowest in the Mokau and Motu, and highest in the Mataura and Karamea. The patterns observed did not seem to be related to catchment development, such that catchments with minimal disturbance (Motu, Karamea, Mokihinui) often spanned the range of values observed. Interpretation of relationships between functional indicators and indices of macroinvertebrate health and habitat quality were limited due to low samples size. Significant relationships were detected between GPP and Habitat score, and between cotton strip decay rate per degree day (k_{dd}) and MCI (Table 8), but these correlations were driven by outlying points (Waikato for cotton k_{dd} and Manawatu for GPP). Investigation of relationships between functional indicators and large river macroinvertebrate metrics were hampered in this study by the low sample size.

Table 7: Gross primary production (GPP), ecosystem respiration (ER) and decay rates for wooden sticks and cotton strips (k per day adjusted for degree days) for nine of the large river sampling sites. ND = no data.

	GPP (g O_2 m^{-2} d^{-1})	ER (g O_2 m^{-2} d^{-1})	Stick k_{dd}	Cotton k_{dd}
North Island				
Motu	1.23	4.26	0.00007	0.00027
Manawatu	107.11	65.24	0.00003	ND
Rangitikei	1.07	28.98	0.00004	0.00090
Mokau	ND	ND	0.00010	0.00020
Waikato	3.00	3.64	ND	0.01133
South Island				
Mataura	5.88	11.72	ND	0.00185
Oreti	7.97	10.27	ND	0.00074
Mokihinui	7.44	22.65	0.00008	0.00053
Karamea	5.26	3.86	0.00013	0.00181

Table 8: Spearman rank correlation coefficients between four macroinvertebrate community metrics, habitat score and measures of river metabolism (gross primary production (GPP; g O_2 m^{-2} d^{-1}) and ecosystem respiration (ER; g O_2 m^{-2} d^{-1}), and stick and cotton processing (*k* per day adjusted for degree days). * = P <0.05**.**

	GPP (n = 8)	ER (n = 8)	Stick k_{dd} (n = 6)	Cotton k_{dd} (n = 8)
Shannon	-0.34	-0.20	0.38	-0.24
EPT* richness	-0.38	-0.38	0.68	-0.53
%EPT*	-0.45	-0.49	0.20	-0.50
MCI	-0.29	-0.19	0.68	-0.88*
Habitat score	-0.77*	-0.63	0.32	-0.17

4. Discussion

The principal aim of this study was to determine the nature of any relationships between anthropogenic stressor variables, in particular upstream native cover, nitrogen concentrations and impervious area, on macroinvertebrate communities in large rivers. We were curious to find out whether relationships between these variables, and also functional indicators, were similar to those observed in wadeable streams. Impervious areas in contributing catchments were low in this study, as in the national large river compilation (Table 2), suggesting that urban influences are unlikely to be major pressures to consider for large river environments in New Zealand in general. Rather, pressure gradients are more likely related primarily to other land cover types and ensuing effects of nutrient enrichment, although abrupt changes such as the presence of dams were also implicated in this study. We sampled only a small proportion of large rivers nationally, and because of their individual physical characteristics they did not always closely reflect the "typical" stressor profiles of large rivers generally as indicated by a national large river dataset compilation.

Environmental variables

Classification of sites using FWENZ variables (PCA analysis) revealed strong geographic groupings, particularly for Westland, Southland and North Island sites, indicating spatial differences in underlying catchment, segment and reach-scale patterns. While this separation was driven in part by upstream land cover, which reflects anthropogenic disturbance, underlying variables relating to upstream geology, slope, climate and segment flow characteristics also appeared to play a significant role. Greater statistical distance among North Island sites compared to South Island sites probably reflects finer-scale variations in geology and hydrology, particularly peat and also lake influences which in part reflect anthropogenic effects of upstream dams.

As expected the WONI analysis clearly discriminated among the sites sampled according to measures of anthropogenic pressure. Oreti, Mokau, Rangatikei, Manawatu, Waipa and Waihou rivers were most strongly associated with *LogN* and *Imperv*, whereas Karamea and Mokihinui were most strongly associated with *Natcover*. Qualitative assessments of habitat quality also discriminated sites with low pressure suggesting a relationship between catchment ”intactness” and reach-scale habitat quality, although low channel gradient contributing to the establishment of depositional zones and more submerged vegetation may have influenced any relationship between catchment condition and habitat quality for some of the Waikato sites.

Relationships with macroinvertebrate communities

Channel gradient, as reflected by the presence of riffles, appeared to have a strong bearing on the composition of macroinvertebrate communities in the NMDS analysis, although within higher gradient sites there was a clear separation among sites with minimal pressure (Karamea, Mokihinui and Motu) compared to other more impacted sites. Indeed, inclusion of a dummy variable accounting for channel gradient (riffles present absent), along with segment riparian shade and upstream natural cover provided high explanatory power in the BV-step analysis, with little increase in power provided by reach-scale habitat quality. This result suggests that, although local habitat conditions tend to be better at sites with minimal pressure, larger-scale variables can be used to account for this effect on macroinvertebrate community composition in large rivers. Moreover, the full complement of WONI pressure variables was not required to provide high explanatory power in macroinvertebrate community composition, reflecting in part the over-riding influence of land use (in terms of vegetation cover and modelled *LogN* concentration as a surrogate for land-use intensity) as a

modifying variable at the sites studied, and also potentially the antagonistic responses of various WONI variables coupled with the low number of sampling sites.

An average of 31 taxa was found across all sites sampled in the present study, compared to 15 taxa in the national large river compilation. Sampling methods are not known for the national large river dataset, but they are unlikely to have been as comprehensive as in the present study, highlighting the value of sampling multiple habitats to determine biodiversity values. However, of the set of three macroinvertebrate metrics identified as showing least redundancy, only % EPT abundance showed a clear effect of habitat type, with riffle faunas separating from the other habitats in the TREES analysis. This is not surprising given that many EPT species are known to be dependent on fast water velocities. Of the pressure variables of particular interest in this study (note that *Imperv* was low for all sites), neither *Natcover* nor *LogN* were distinguished in the hierarchical regression tree analysis as key pressure variables. However, upstream indigenous cover was identified for Shannon diversity in habitats predicted to be dominated by runs or slower water when all sampled habitats were considered separately (Fig. 4). This result differed from the TREES analysis of the national large rivers dataset (Fig. 5) which identified *LogN* as the primary split for % EPT richness, and *Natcover* as associated with this metric and MCI. The contrasting results between these two analyses could partly reflect the fact that sites included in the national large river compilation had habitat typically dominated by riffles (i.e., median LOCHAB = 4.1) whereas those in the sampled dataset were typically dominated by runs. The EPT and MCI metrics are likely to be most applicable to riffle faunas, suggesting that habitat-specific metrics may need to be developed for large rivers without riffles and that knowledge of the location of stressor effects may be necessary for interpreting results (e.g., a riverside discharge plume may affect mainly littoral habitats).

Despite these differences evident in the TREES analysis, *Natcover* was a significant predictor variable in the linear regression analysis for Shannon diversity, EPT richness and % EPT abundance, with *LogN* providing significant predictive power for the last two metrics. The difference between the TREES and linear regression analyses suggests that relationships between macroinvertebrate metrics and these pressure variables were more linear than being regulated by defined thresholds for the sites sampled (see Appendix 4). In contrast, no significant predictor variables were identified for MCI, suggesting this metric may not be suitable for large rivers where multiple habitat types are sampled. Rather, as shown in the GLM analysis, non-EPT compositional measures involving other insects, Crustacea, Mollusca and Oligochaeta may yield metrics that respond to land-use pressure independently of habitat type.

Functional measures

Functional indicators suggest that large rivers exhibit a wide range in rates of metabolism and organic matter processes. In particular, rates of metabolism in Manawatu River are extremely high compared to the literature and values recorded at 86 smaller streams at a similar time (Clapcott *et al.* 2009). Whilst there were insufficient data to conduct robust statistics, exploration of a broader national dataset (R Young, Cawthron Institute, unpubl. data) suggests few predictive relationships between anthropogenic pressures and one-off measures of functional indicators. It is likely that, like hydrological variables, a greater understanding of temporal dynamics is necessary to assess trends in river metabolism, due to their time integrating characteristics. Furthermore, the limited relationship between functional and structural indicators in large rivers is similar to that observed in smaller systems (Clapcott *et al.* 2009).

5. Conclusions

Macroinvertebrate communities in these large rivers appear to respond to anthropogenic stressors in a similar way expected for smaller streams, although interpretation was limited by the number of sites sampled. While some conventional macroinvertebrate metrics, such as EPT richness and % EPT abundance, appeared to be strongly influenced by anthropogenic stressors, MCI did not appear to be as effective for large rivers where multiple habitats were sampled and/or riffles were absent. Our results suggest that sampling of multiple habitats is required to accurately document the biodiversity of large river macroinvertebrate communities, and that metrics derived from groups more common in large river environments (e.g., Crustacea) may provide a useful addition to other metrics for documenting large river health.

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8. Appendices

Appendix 1: *FWENZ variables*

Upstream/catchment scale predictors

USDaysRainGT25: days/month with rainfall greater than 25 mm in the upstream catchment to indicate the likely frequency of elevated flows;

USAvgTNorm: average air temperature (^{o}C) in the upstream catchment, normalised with respect to SegJanAirT, with negative values indicating colder (higher elevation) headwaters than average, given the segment temperature, and positive values indicating warmer (lower elevation) headwaters;

USAvgSlope: average slope in the upstream catchment (^{o}), describes catchment-driven modification of flow variability;

USNative: area with indigenous vegetation (proportion) in upstream catchment;

USIndigFor: area with indigenous forest (proportion) in upstream catchment;

USCalcium: calcium concentrations in surface rocks;

USPhosphorus: average phosphorus concentrations available in surface rocks;

USHardness: average hardness of underlying rocks, variation in geological substrates which affects flow variability;

USLake: area of lake in catchment (proportion), describes local buffering of river flows in the upstream catchment by lakes;

USPeat: area of peat in catchment (proportion), describes local buffering of river flows in the upstream catchment by wetlands;

Segment scale predictors

SegFlow: mean annual 7-day low flow (m^3/sec), derived from hydrological models;

SegLowFlow: mean annual flow (m^3/sec), derived from hydrological models;

SegFlowStability: ratio of annual low flow / annual mean flow;

SegFlow4th Root: 4^{th} root transformed mean annual 7-day low flow $(\text{low flow} + 1)^{0.25}$;

SegJanAirT: summer (January) air temperature (^{o}C);

SegMinTNorm: average minimum daily air temperature (^{o}C) normalised with respect to SegJanAirT, negative values indicate strongly seasonal climates and positive values indicate weakly seasonal climates;

SegRipShade: riparian shading (proportion), the likely degree of riparian shading, derived by using national, satellite image-based vegetation classification to identify riparian shading in each segment, with the degree of shading then estimated from river size and expected vegetation height;

SegSlopeSqrt: square-root (+1) transformed segment slope (^{o}), derived from GIS calculation using length and difference between upstream and downstream elevation for each segment;

SegLogN: $\log_{10}$ total nitrogen concentration, stream nitrogen load as estimated from CLUES' a leaching model combined with a regionally-based regression model, implemented within a catchment framework;

Reach scale predictors

LocHab: weighted average of proportional cover of local habitat using categories of: 1–still; 2–backwater; 3–pool; 4–run; 5–riffle; 6–rapid; 7–cascade, predicted from a boosted regression tree model;

LocSed: weighted average of proportional cover of bed sediment using categories of: 1–mud; 2–sand; 3–fine gravel; 4–coarse gravel; 5–cobble; 6–boulder; 7–bedrock, predicted from a boosted regression tree model.

Appendix 2: *WONI Pressure variables*

Natcover: the proportional natural vegetation cover in the planning unit;

LogN: log10 total nitrogen concentration, range from -4.1 (very low concentrations) to 3.1 (very high concentrations), based on CLUES, a regionally-based regression model implemented within a catchment framework;

Imperv: proportional cover of impervious surfaces in the upstream catchment (proportion) ranging from 0-1, supplied by D. Brown, Department of Conservation. The proportional cover of impervious surfaces for the immediate catchment was calculated and traversed downstream and an area weighted average for the upstream catchment was calculated;

Saltru_res: predicted probability of capture for Salmo trutta (brown trout);

Dameffect: downstream effects of dams/barriers on species populations. Flow weighted calculation of upstream dam effects and their progressive dilution downstream as flow increases with input from undammed tributaries. Locations of known dams were supplied by Department of Conservation;

Mineffect: mineral mine point discharges;

Geotheffect: point discharges of human extracted geothermal water;

Coaleffect: coal mine point discharges;

Av_Sum: pressure indices calculated from individual pressure factors (average);

Min_Sum: pressure indices calculated from individual pressure factors (minimum)

Appendix 3: *Results of General Linear Model analysis for % composition and taxonomic richness of major macroinvertebrate groups at large river sites with riffles.*

Impact = high (> 0.16) or low (< -0.3) *LogN*; Habitat = littoral or deepwater.

%EPT N: 56 Multiple R: 0.774 Squared multiple R: 0.599

Source	Sum-of-Squares	df	Mean-Square	F-ratio	P
IMPACT	0.858	1	0.858	10.966	0.002
HABITAT	0.312	1	0.312	3.989	0.051
IMPACT*HABITAT	0.361	1	0.361	4.615	0.037
SITE(IMPACT)	3.814	4	0.954	12.190	0.000
Error	3.755	48	0.078		

% Non-EPT Insecta N: 56 Multiple R: 0.763 Squared multiple R: 0.583

Source	Sum-of-Squares	df	Mean-Square	F-ratio	P
IMPACT	2.238	1	2.238	31.089	0.000
HABITAT	0.012	1	0.012	0.164	0.687
IMPACT*HABITAT	0.024	1	0.024	0.338	0.564
SITE(IMPACT)	2.782	4	0.696	9.663	0.000
Error	3.455	48	0.072		

% Mollusca N: 56 Multiple R: 0.615 Squared multiple R: 0.378

Source	Sum-of-Squares	df	Mean-Square	F-ratio	P
IMPACT	1.574	1	1.574	18.510	0.000
HABITAT	0.019	1	0.019	0.221	0.641
IMPACT*HABITAT	0.011	1	0.011	0.124	0.726
SITE(IMPACT)	0.893	4	0.223	2.626	0.046
Error	4.083	48	0.085		

% Crustacea N: 56 Multiple R: 0.582 Squared multiple R: 0.339

Source	Sum-of-Squares	df	Mean-Square	F-ratio	P
IMPACT	0.394	1	0.394	12.294	0.001
HABITAT	0.199	1	0.199	6.216	0.016
IMPACT*HABITAT	0.006	1	0.006	0.187	0.667
SITE(IMPACT)	0.168	4	0.042	1.311	0.279
Error	1.539	48	0.032		

% Oligochaeta N: 56 Multiple R: 0.804 Squared multiple R: 0.647

Source	Sum-of-Squares	df	Mean-Square	F-ratio	P
IMPACT	1.771	1	1.771	41.855	0.000
HABITAT	0.000	1	0.000	0.001	0.981
IMPACT$*HABITAT	0.004	1	0.004	0.089	0.766
SITE(IMPACT)	1.897	4	0.474	11.208	0.000
Error	2.031	48	0.042		

Total richness N: 56 Multiple R: 0.681 Squared multiple R: 0.464

Source	Sum-of-Squares	df	Mean-Square	F-ratio	P
IMPACT	63.375	1	63.375	8.947	0.004
HABITAT	56.719	1	56.719	8.007	0.007
IMPACT*HABITAT	105.433	1	105.433	14.884	0.000
SITE(IMPACT)	77.313	4	19.328	2.729	0.040
Error	340.021	48	7.084		

Trichoptera richness N: 56 Multiple R: 0.769 Squared multiple R: 0.591

Source	Sum-of-Squares	df	Mean-Square	F-ratio	P
IMPACT	11.482	1	11.482	7.005	0.011
HABITAT	14.144	1	14.144	8.629	0.005
IMPACT*HABITAT	36.001	1	36.001	21.965	0.000
SITE(IMPACT)	54.918	4	13.729	8.377	0.000
Error	78.672	48	1.639		

Diptera richness N: 56 Multiple R: 0.821 Squared multiple R: 0.674

Source	Sum-of-Squares	df	Mean-Square	F-ratio	P
IMPACT	80.667	1	80.667	66.075	0.000
HABITAT	1.548	1	1.548	1.268	0.266
IMPACT*HABITAT	6.190	1	6.190	5.071	0.029
SITE(IMPACT)	35.882	4	8.971	7.348	0.000
Error	58.600	48	1.221		

Appendix 4: *Relationships between the two main WONI pressure variables and MCI and % EPT taxa richness for the national large rivers data compilation (closed circles) and the 11 sites sampled in the present study (open squares).*