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Preliminary report on macroinvertebrate assemblages of Pyrites and Goodmans Creek in relation to the management of Merrimu Reservoir: patterns in 2013 in early stages of flow manipulation

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Technical Report

14.8

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Preliminary report on macroinvertebrate assemblages of Pyrites and Goodmans Creek in relation to the management of Merrimu Reservoir: patterns in 2013 prior to flow manipulation

Report to: Melbourne Water

Published: Dec 2014

Cite as: *Walsh, C.J., Bloink, C & Hehir, G. 2014. Preliminary report on macroinvertebrate assemblages of Pyrites and Goodmans Creek in relation to the management of Merrimu Reservoir: patterns in 2013 in early stages of flow manipulation. Melbourne Waterway Research-Practice Partnership Technical Report 14.8. Waterway Ecosystem Research Group, The University of Melbourne.*

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Reviewed by: Bill Moulden, Edward Tsyrlin

Cover photo: Pyrites Creek Site PCER5 – approximately 6m downstream of Merrimu Reservoir.

Summary

Environmental flow management requires understanding of the degree to which the natural flow regime must be replicated to protect ecological structure and function of streams. A basic requirement, particularly in the application of environmental flow management to intermittent streams, is a robust understanding of ecological responses to different aspects of the flow regime.

This report presents an analysis of macroinvertebrate assemblage composition in two intermittent streams, Pyrites and Goodmans creeks, in the early stages of an environmental flow program in the lower reaches of Pyrites Creek. Both creeks have segments with augmented flows that are used as water supply conduits, and Pyrites Creek has a large water supply reservoir, which greatly reduces the variability and volume of flow downstream, while producing a small permanent flow through leakage for several kilometres downstream. We assess the nature of variation in assemblage composition, between the two creeks, along each creek, and among the segments with differing flow regimes. Specifically, we aimed to assess the likely effects on assemblage composition of a) the use of the streams as water supply conduits and b) Merrimu reservoir (Pyrites Creek only) in its pre-2014 flow management. Finally, we considered appropriate analytical approaches for assessing changes resulting from environmental flow releases from Merrimu reservoir now and in the future.

Pyrites Creek macroinvertebrate assemblage composition downstream of the reservoir was distinct from all other sites. The permanently flowing sites downstream of the reservoir were in substantially worse condition than most other sites, with a much greater dominance of scrapers (algal grazers) than other sites. This suggests a shift in trophic functioning of the stream from a riparian-organic-matter-dominated food-web to an algal-dominated one. The higher abundance of scrapers (primarily several snail families and notonemourid stoneflies) and other disturbance-tolerant families was accompanied by an absence or low abundance of a range of sensitive families.

In the Pyrites Creek segment further downstream, in which the small permanent flow transitioned into intermittency with reduced flows, the macroinvertebrate assemblage showed greater similarity to upstream, intermittent sites.

Intermittent sites with augmented flows differed in assemblage condition from other intermittent sites and had higher family richness. The most upstream intermittent sites were more similar to the Pyrites Creek sites downstream of the reservoir, than to other upstream sites, which we hypothesize is a result of those sites having shorter and less reliable wet periods or being at a later stage of the annual drying cycle than others.

The dominance of scrapers downstream of the reservoir suggests that a strong, detectable change in assemblage composition to an assemblage more dominated by riparian inputs of organic matter is possible, if environmental flow releases adequately mimic the magnitude and frequency of high-flow events capable of scouring biofilms. However, transfer rates limit capacity to use flows to scour the stream bed and remove the stands of *Typha* that have colonized the channel downstream of the reservoir. Thus changes to assemblage composition are likely to be limited.

The large seasonal variability in intermittent streams such as these, present challenges for robust monitoring of changes resulting from management actions. As much temporal replication of sampling as possible is recommended. Spatial autocorrelation among sites points to the need to use Bayesian statistical methods for future analyses, comparing 2013 assemblage patterns with those in future years following longer periods of ongoing environmental flow management.

Introduction

The adoption of environmental flow management for the protection of freshwater and estuarine ecosystems has grown steadily over recent decades (Postel and Richter 2003), with a growing consensus on scientific approaches to ensure their efficacy (Poff et al. 2010). A common principle in environmental flow management is that the structure and function of rivers and streams are largely determined by the nature of the historic flow regime, and that ecosystem protection requires replication of the important facets of that regime (Poff et al. 2010). A basic need in determining those facets and the degree to which they need to be replicated, is a robust understanding of ecological responses to the flow regime.

The importance of flow regime in the structuring of biological communities in intermittent streams is well understood (e.g. Boulton and Lake 1992a; Brooks 1998), but the application of environmental flow regimes to such streams to retain their intermittent nature is not common. This report presents an analysis of macroinvertebrate assemblage composition in two intermittent[1] streams, Pyrites and Goodmans creeks, in the early stages of an ongoing environmental flow program. The creeks partly rise in the Lerderderg State Park and on private land, and Pyrites Creek drains to Merrimu Reservoir, which is used primarily supply drinking water to Melton and Bacchus Marsh. Segments of both streams are used as conduits of water from other catchments. As a result the streams vary longitudinally in flow regime (Fig. 1, Table 1).

Environmental flows were released from the reservoir July–November 2012 and over a similar period in 2013. This report is a preliminary assessment of the biological data collected in December 2013 (prior to a final fresh release in that year) to support and monitor the environmental flow program. Melbourne Water commissioned Ecology Australia to sample macroinvertebrate assemblages along the creeks in December 2013. These data are to be used to assess the influence of current flow regimes on in-stream assemblages, and to serve as baseline data for the assessment of future environmental flow management.

The current flow management practices and diversion/storage infrastructure of the creeks differentiates segments of each stream into four hydrologic classes (Fig. 1, Table 1). The flow regimes of the uppermost reaches remain largely intact (ephemeral natural). The segment of each creek used as a water supply conduit carries increased volumes of water and remain flowing for up to 2 months longer than the upstream segments (ephemeral enhanced). Immediately downstream of the reservoir, leakage results in a small permanent flow for several kilometres downstream in Pyrites Creek (permanent). Downstream of the permanent segment of Pyrites Creek and downstream of the ephemeral enhanced segment of Goodmans Creek, the length of the annual flow season, flow volume and peak flows (Pyrites Creek) are reduced by abstractions (ephemeral reduced).

In the reaches of Pyrites Creek downstream of Merrimu reservoir, environmental releases of 2012 and 2013 increased flow variability, although not to the same degree as the natural and enhanced reaches. In the ephemeral reduced reach of Pyrites Creek, environmental releases also increased the duration of the flow period in dry years. Currently, the ecological objective of the environmental releases is to improve the diversity and abundance of macroinvertebrate communities within the affected reaches and the region, by providing a 'drought refuge' in dry years. If necessary, these objectives will change in response to the findings of this monitoring program.

In this report, a preliminary analysis of assemblage composition along the creeks and among the four hydrologic classes is presented to inform future analysis of the data. Primary questions to be addressed by the analyses are:

- What is the nature of variation in assemblage composition: between the two creeks, along each creek, and among the four hydrologic classes?

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[1] The streams in this study are intermittent: the hydrologic classes used in this draft report refer to most reaches as ephemeral. This should be changed for subsequent considerations.

[2] Current attenuated forest cover (AF) and attenuated imperviousness (AI) (sensu Walsh and Webb 2013) were estimated using the values calculated by Walsh and Webb (2013: from 2006 aerial imagery) for reaches of the 12,380 stream segments in Melbourne Water's DCI dataset (Grace Detailed-GIS Services 2012). As sites generally fell along each stream segment (and the AF and AI estimates were calculated for the bottom of each segment), we used the mapping tool at http://urbanstreams.net/tools/bugmodels/ that allows recalculation of AF following removal or addition of patches of forest cover to estimate AF for each site. (AI was very low in all sites, but its value was interpolated between segments for each site). The preparation of this report was used as a basis for developing a new tool to allow users to conduct similar analyses in the future. The tool will be located at

- What are the likely effects on assemblage composition of a) the use of the streams as water supply conduits and b) Merrimu reservoir in its pre-2014 flow management?
- What are appropriate analytical approaches for assessing changes to Pyrites Creek resulting from future environmental flow releases from Merrimu Reservoir?

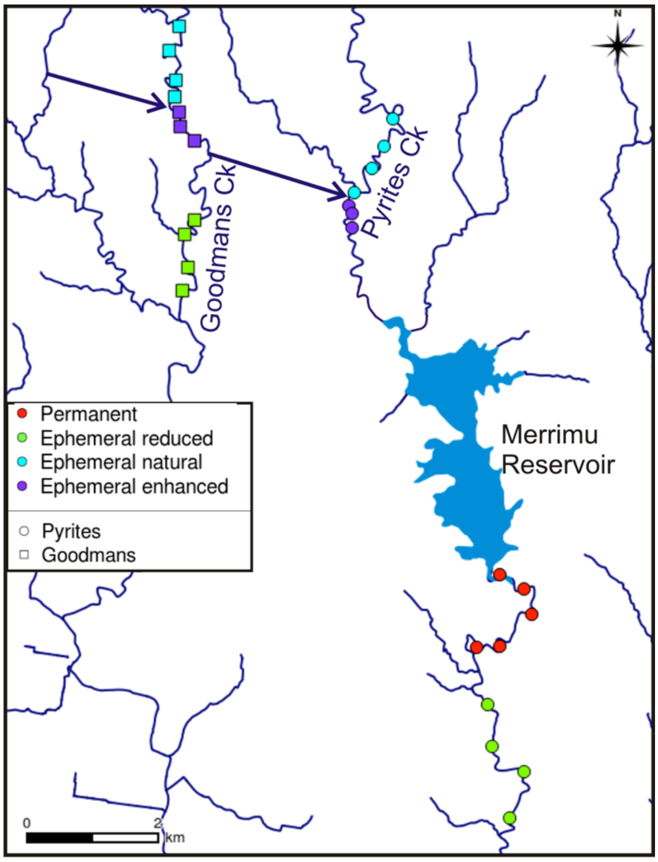
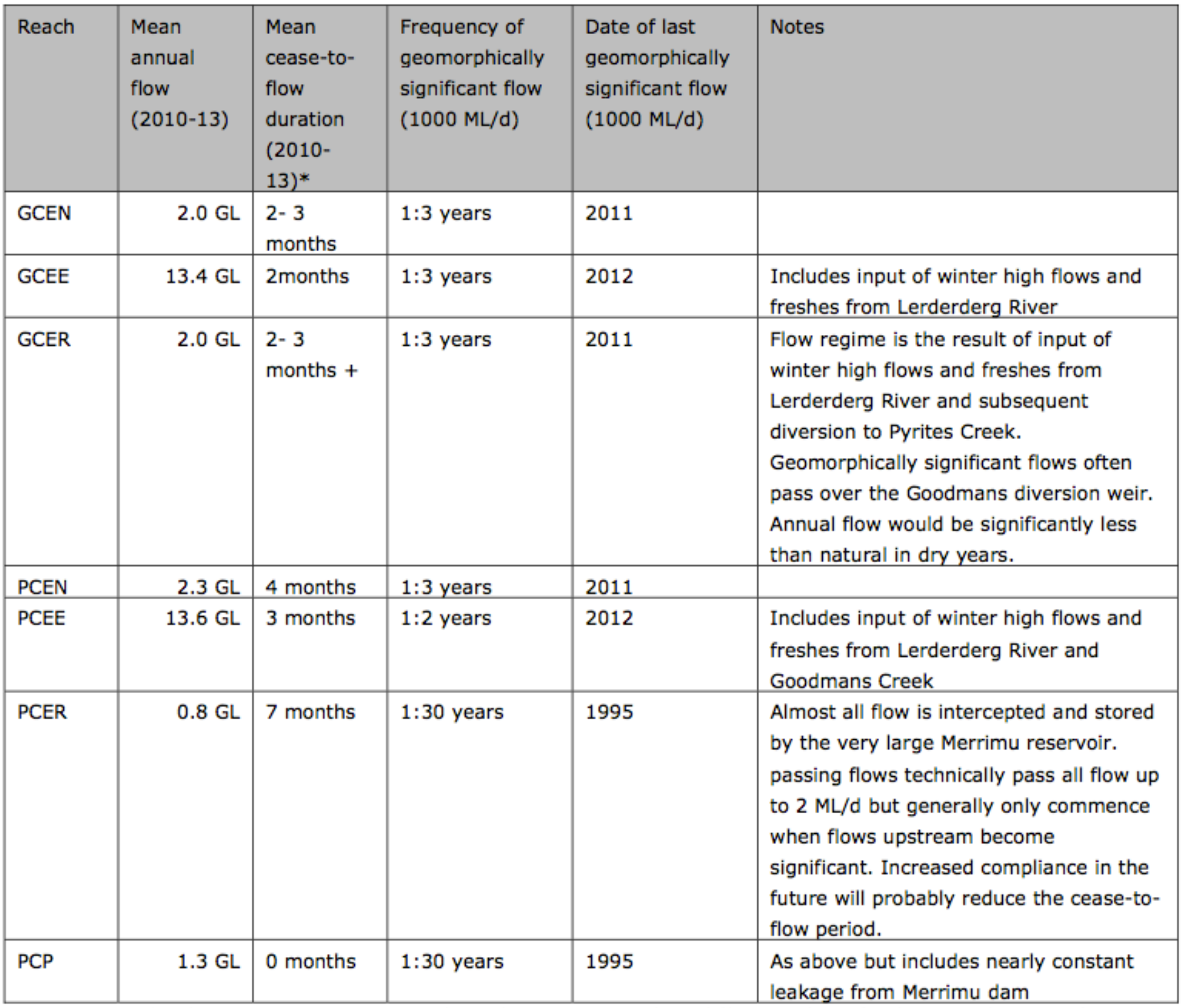


Fig. 1. Map of the 28 sites sampled for macroinvertebrates in Pyrites and Goodmans creeks. The symbol conventions (used throughout this report) separate sites into hydrologic classes. 'Ephemeral natural' sites in the headwaters of both streams have neither augmented nor abstracted flows. 'Ephemeral enhanced' sites are in sections of the streams used as water supply conduits for Merrimu Reservoir, diverting water through tunnels between catchments (black arrows). 'Ephemeral reduced' sites are downstream of the water supply diversion (on Goodmans Creek) and the reservoir (on Pyrites Creek). Immediately downstream of the reservoir on Pyrites Creek, leakage results in a permanently flowing reach, with reduced temporal variability. Discrete boundaries exist between all flow regime reaches except Pyrites Creek 'permanent' and Pyrites Creek 'ephemeral reduced', where an arbitrary reach was determined on the basis of flow observations in consultation with Melbourne Water.



Reach	Mean annual flow (2010-13)	Mean cease-to-flow duration (2010-13)*	Frequency of geomorphically significant flow (1000 ML/d)	Date of last geomorphically significant flow (1000 ML/d)	Notes
GCEN	2.0 GL	2- 3 months	1:3 years	2011	
GCEE	13.4 GL	2months	1:3 years	2012	Includes input of winter high flows and freshes from Lerderderg River
GCER	2.0 GL	2- 3 months +	1:3 years	2011	Flow regime is the result of input of winter high flows and freshes from Lerderderg River and subsequent diversion to Pyrites Creek. Geomorphically significant flows often pass over the Goodmans diversion weir. Annual flow would be significantly less than natural in dry years.
PCEN	2.3 GL	4 months	1:3 years	2011	
PCEE	13.6 GL	3 months	1:2 years	2012	Includes input of winter high flows and freshes from Lerderderg River and Goodmans Creek
PCER	0.8 GL	7 months	1:30 years	1995	Almost all flow is intercepted and stored by the very large Merrimu reservoir. passing flows technically pass all flow up to 2 ML/d but generally only commence when flows upstream become significant. Increased compliance in the future will probably reduce the cease-to-flow period.
PCP	1.3 GL	0 months	1:30 years	1995	As above but includes nearly constant leakage from Merrimu dam

Table 1. Description of flow regime in each stream reach. GC, PC = Goodmans and Pyrites creeks, respectively; EN, EE, ER = ‘Ephemeral Natural’, ‘Enhanced’, ‘Reduced’ respectively; P = ‘Permanent’ (Source: Melbourne Water).

Methods

Sampling and processing methods

Two samples were collected from each of 28 sites (Fig. 1) using rapid bioassessment methods (EPA Victoria 2003) by kick-sampling from riffles and sweep sampling from pool edges, between 2 and 17 December 2013. In six sites (four of the permanently flowing sites on Pyrites, and one of the ephemeral reduced sites on each creek), no sample-able riffle was present, and two edge samples were collected.

All samples were preserved in 70% ethanol and subsampled in the laboratory according the method of Walsh (1997). 10% of each sample was first sorted, then, if 300 individual specimens had not been collected, it was further subsampled to achieve 300 specimens. In addition, each whole sample was scanned and any further taxa missed in the subsampling process were identified and noted as scanned specimens.

Specimens were identified to family (sub-family for Chironomidae) where possible. Damaged or immature specimens that could not be confidently identified to family were identified as precisely as possible and noted as ambiguous.

Several taxonomic changes were made to the dataset to be consistent with the analyses of Walsh and Webb (2013). Nematoda were excluded, as they were recorded rarely and are likely to be missed in many samples. Recent divisions of the odonate families Aeshnidae and Corduliidae were not used. Temnocephala are obligate commensals on crayfish. Although they have been recorded on large atyid species, they are not associated with *Paratya australiensis*, the only atyid recorded in this region. They are thus certainly associated with parastacid crayfish in these study streams. Thus all records of Temnocephala were recoded as Parastacidae.

Although bulk RBA samples as described above are generally considered qualitative, they do represent a standard sampling effort, so we report (log-transformed) abundance as a measure of number per unit effort. To calculate abundance, counts of all subsampled specimens (including ambiguously identified specimens but excluding scanned specimens), were divided by the subsampled fraction of the sample.

To assess compositional similarity, only non-ambiguous specimens were considered and both subsampled and scanned specimens were included. Abundance of scanned specimens was their raw count.

To assess family richness, both ambiguous and scanned specimens were excluded from consideration.

After an initial assessment of differences between edge and riffle samples, analyses were conducted on combined sample pairs. The abundances of all families in the pair of samples taken at each site were summed (or converted to presence-absence, depending on the analysis).

Statistical analysis

Non-metric multidimensional scaling based on Bray-Curtis dissimilarity (Minchin 1987) was used to derive two-dimensional ordinations of samples or sample pairs to permit a visual representation of the similarity among samples. Two ordinations were calculated: one using presence-absence data, and the other using log(x+1)-transformed data.

The families whose transformed densities or occurrence were correlated ($P \leq 0.0001$) with the ordination space were identified using the function envfit (Oksanen et al. 2013). This function finds the direction in the ordination space towards which each taxon abundance changes most rapidly and to which it is maximally correlated with the ordination configuration. The significance of the family vectors was determined by a permutation test ($n = 9999$).

Family richness for each sample pair was calculated by rarefaction. We randomly subsampled the data from each sample pair to 200, 400 and 600 individuals and calculated richness for each subsample. This randomization was repeated 1000 times to calculate a mean richness, expressed as the number of families expected for 200 (EF_{200}), 400 (EF_{400}) and 600 (EF_{600}) individuals.

Two biotic indices were also calculated to provide a synoptic assessment of stream condition:

- SIGNAL (Stream Invertebrate Grade Number-Average Level), based on pollution-sensitivity grades of stream invertebrate families (Chessman 1995) using the scoring variant of (EPA Victoria 2003).
- LUMaR (Land-Use Macroinvertebrate Response), based on 60 models of family distribution across the Melbourne region (Walsh and Webb 2013).

For both indices, the predictive models of Walsh and Webb (2013) were used to compare observed scores with those predicted for the sites given their current forest cover and urban land use[2]. Lower than predicted scores would be suggestive of impacts on stream condition other than loss of forest or catchment urbanization: the only two human impacts used by Walsh and Webb in their models. In addition, LUMaR predictions allow diagnosis of the families driving differences between observed and predicted scores.

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[2] Current attenuated forest cover (AF) and attenuated imperviousness (AI) (sensu Walsh and Webb 2013) were estimated using the values calculated by Walsh and Webb (2013: from 2006 aerial imagery) for reaches of the 12,380 stream segments in Melbourne Water's DCI dataset (Grace Detailed-GIS Services 2012). As sites generally fell along each stream segment (and the AF and AI estimates were calculated for the bottom of each segment), we used the mapping tool at http://urbanstreams.net/tools/bugmodels/ that allows recalculation of AF following removal or addition of patches of forest cover to estimate AF for each site. (AI was very low in all sites, but its value was interpolated between segments for each site). The preparation of this report was used as a basis for developing a new tool to allow users to conduct similar analyses in the future. The tool will be located at http://urbanstreams.net/tools/lumar.

To assess if compositional differences were explained by differences in the dominance of different functional traits among macroinvertebrates, each family was assigned to a functional feeding group (using classifications determined by Chessman 1986; Boulton and Lake 1992b; Gooderham and Tsyrlin 2002), and abundances of scrapers (algal/biofilm grazers), shredders and predators were calculated.

Each variable derived from the data (ordination scores, abundances, family richness, SIGNAL, LUMaR, and abundance of families identified as influential either by envfit or LUMaR, and abundance of functional feeding groups) was plotted against stream distance downstream from the most upstream site (estimated manually using the DCI stream layer: Grace Detailed-GIS Services 2012), with different symbols for each hydrologic class of site (Fig. 1). No statistical models for differences between classes are developed in this report, but trends are described to inform the development of models to be used following the collection of further samples.

Results

Initial NMDS analysis revealed that a large proportion of the variation in assemblage composition among samples was between riffle and edge samples. Despite this high degree of variability between habitats, patterns of compositional similarity among groups was similar for edge samples and for riffle samples (Fig 2), with the Pyrites Creek sites downstream of the reservoir being distinct from sites upstream and from Goodmans Creek sites. Sweep samples from the permanently flowing sites immediately downstream of the reservoir were less similar to sweep samples from upstream and Goodmans Creek sites than were those from the ephemeral reduced sites further downstream. Insufficient riffle samples were collected downstream of the reservoir to make a similar inference about riffle samples (where riffles were absent, two edge samples were collected).

Because of the large variability in composition between habitats within sites and the similar patterns among sites, inter-site patterns in assemblage composition could arguably be more robustly assessed (with less within-site 'noise') using the combined data from the pair of samples from each site. As the primary purpose of this analysis is to assess difference in assemblages among sites, all subsequent analyses were conducted using the combined data from pairs of samples at each site.

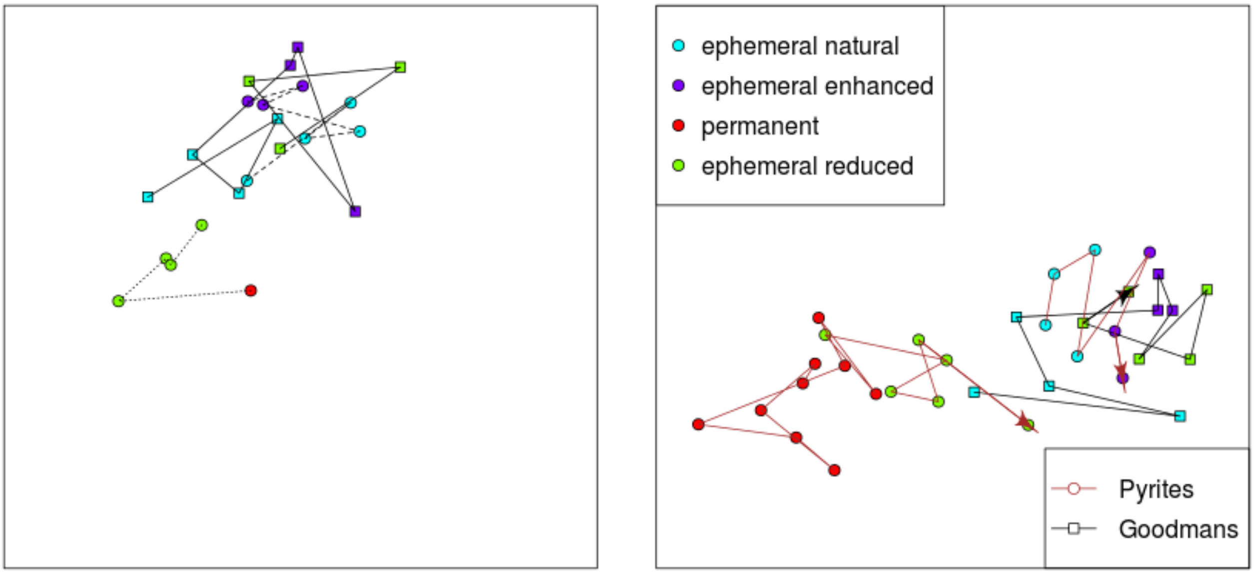

Fig. 2. NMDS of macroinvertebrate samples from 28 sites in Pyrites and Goodmans creeks, based on presence-absence data. Sites are connected to the next downstream site by arrows for each stream, with the arrows for Pyrites Creek being interrupted by Merrimu Reservoir. Both plots are extracted from the same NMDS (Stress = 0.17), but the plot on the left shows only riffle samples and the plot on the right shows only sweep samples.

Patterns attributable to Merrimu Reservoir

The strongest differentiation of sample-pair groups was between Pyrites Creek sites downstream of the reservoir and all other sites. All sites downstream of the reservoir were distinct from other sites, but the permanently flowing sites immediately downstream of the reservoir were less similar to upstream Pyrites and Goodmans Creek sites than were the ephemeral reduced sites further downstream (Figs 3, 4). The patterns of similarity of sites were similar for presence-absence data (Fig. 3) and for log-transformed abundance data (Fig. 4), suggesting that the patterns are driven as much, and in a similar way, by differences in species occurrences as by differences in abundances. Total abundances per sample pair did

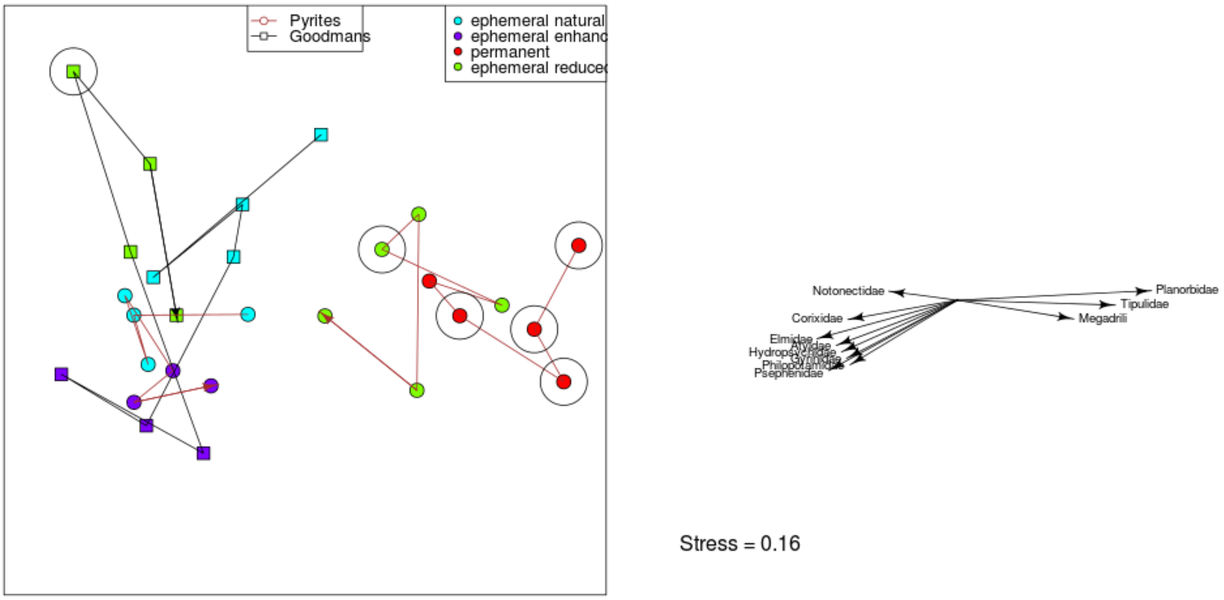

Fig. 3. NMDS of macroinvertebrate sample pairs from 28 sites in Pyrites and Goodmans creeks, based on presence-absence data. Sites are connected to the next downstream site by arrows for each stream, with the arrows for Pyrites Creek being interrupted by Merrimu Reservoir. Sites at which the sample pair consisted of two edge samples are circled. The 11 families whose patterns of occurrence were highly correlated with the ordination are indicated by vectors in the directions that their occurrence increases most rapidly.

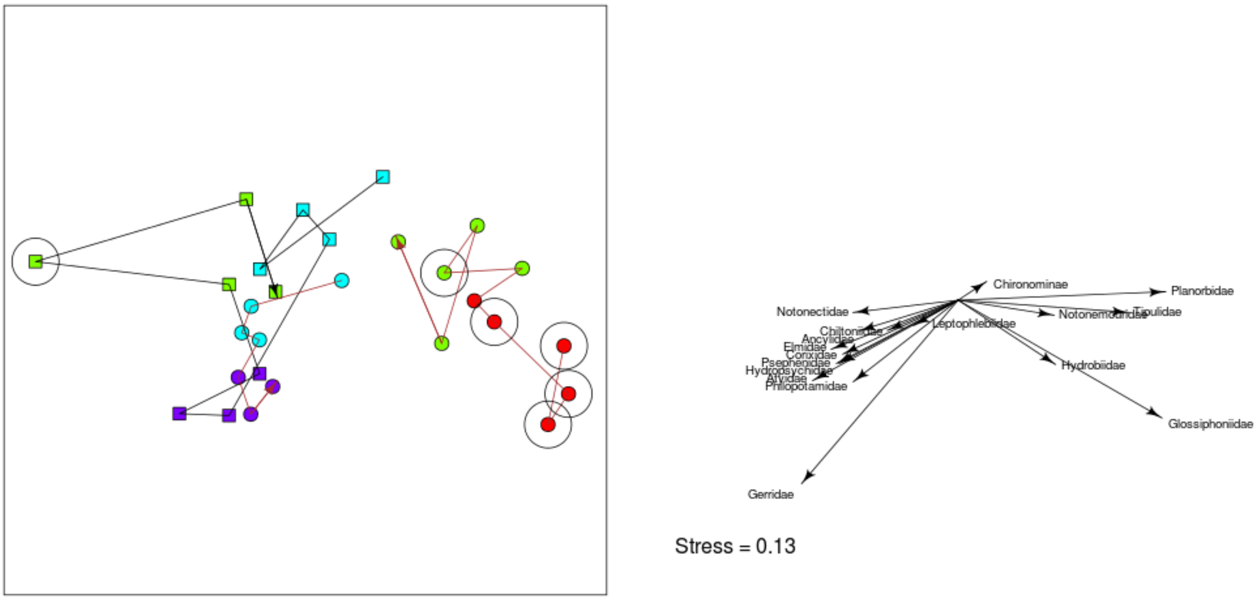

Fig. 4. NMDS of macroinvertebrate sample pairs from 28 sites in Pyrites and Goodmans creeks, based on log(x + 1)-transformed abundance data. Conventions are as for Fig. 3. For this ordination there were 17 families that were highly correlated with ordination space.

not differ strongly among sites, with no clear difference between downstream of the reservoir and other sites (Fig. 5A). Similarly, no strong differences in total family richness per sample pair were evident (Fig. 5B), however there were differences evident in other richness measures.

Sites downstream of the reservoir were in substantially worse condition than most other sites as indicated by 2 biotic indices: LUMaR and SIGNAL (Fig. 6). Lower LUMaR scores, in particular, indicate changes in family composition from what would be expected in these sites in the absence of human impacts. SIGNAL and particularly LUMaR suggest that all sites are in worse condition than would be expected given existing catchment development and forest clearance, and that the divergence from expected is even greater for sites downstream of the reservoir (Fig 5).

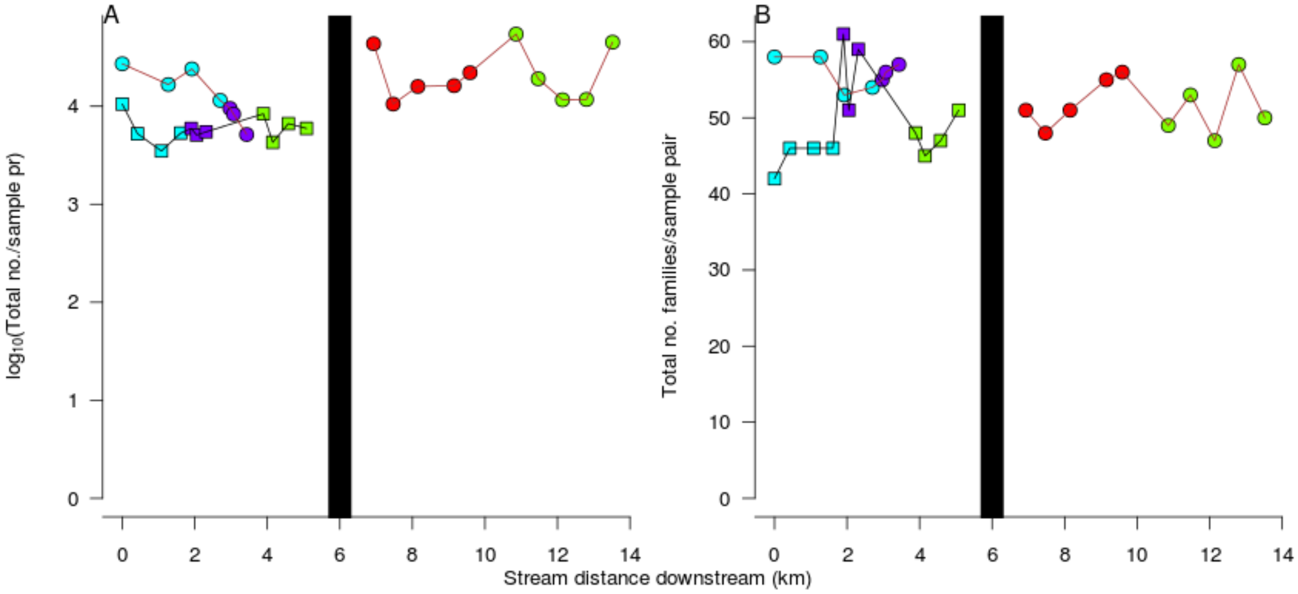

Fig. 5. Trends in total macroinvertebrate abundance and total family richness per sample pair of with distance downstream along Goodmans and Pyrites Creeks. Symbol conventions as in Figs 3 and 4. The solid bar at 6 km downstream from the uppermost Pyrites Creek site represents Merrimu Reservoir.

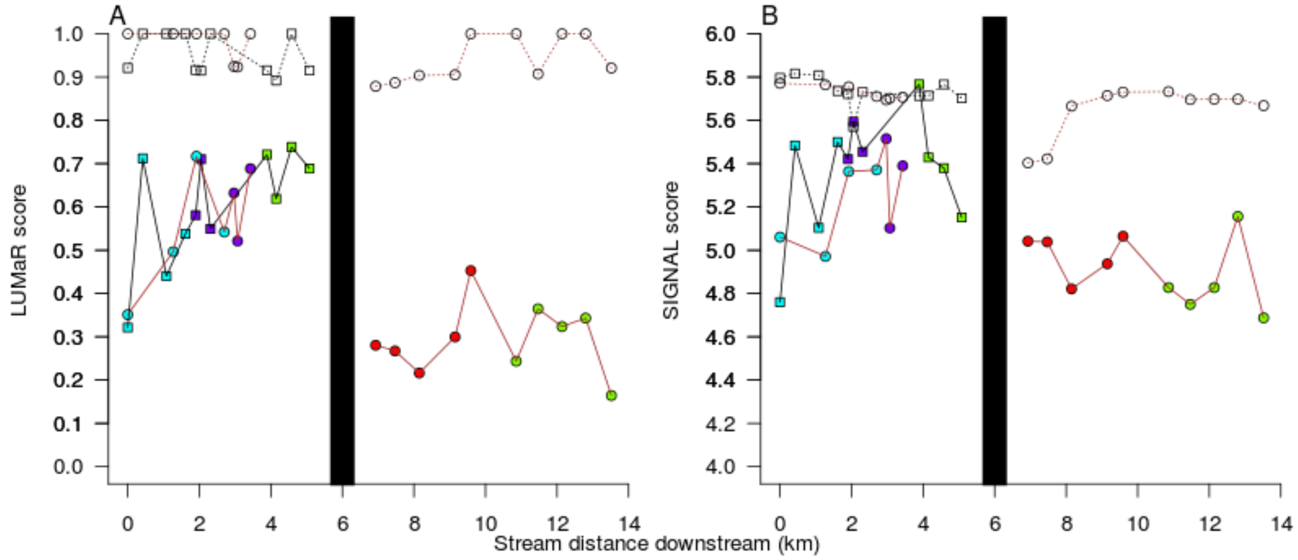

Fig. 6. Trends in two biotic indices with distance downstream along Goodmans and Pyrites Creeks. Symbol conventions as in Fig. 5. The open symbols and dotted lines show the LUMaR and SIGNAL scores predicted by the models of Walsh and Webb (2013) accounting for current forest cover and catchment urbanization for each site. (In almost all cases, the observed scores were lower than predicted.)

The shift in composition downstream of the reservoir is driven by a shift in the trophic structure of the macroinvertebrate assemblage, with a much greater abundance of scrapers (algal grazers) downstream of the reservoir (Fig. 7A). The increase in scraper abundance is dominated by snails (Planorbidae, Physidae and Lymnaeidae, Fig. 7C, G, H) and notonemourid stoneflies (Fig. 7F). Other families that increase downstream of the reservoir—megadrili worms and glossiphonid leeches (Fig. 7B, D) share with Physidae and Lymnaiedae the tendency to be associated with degraded sites. Conversely, Notonemouridae and Tipulidae (Fig. 7E) tend to be more sensitive to catchment disturbance.

The dissimilarity of sites downstream of the reservoir from other sites was also driven by the absence of a number of families (Fig. 8), including elmid beetles, which are also scrapers, and atyid shrimp, which, while not scrapers, do graze on algal biofilms. Comparison of the differences between the expected assemblage under current forest conditions and the observed assemblages points to the lower LUMaR scores downstream of the reservoir, were driven by absence of a range of sensitive families—scirtid, gyrinid and elmid beetles (Fig. 8B, D, E), hydropsychid caddisflies (Fig. 9C), and atyid shrimp (Fig. 8A), but also by unexpected occurrences of macrophyte-associated families such as glossiphonid leeches, and lymnaeid snails (Fig. 7D, H). The presence of invasive physid snails in sites upstream and downstream of the reservoir was unexpected (by the model of Walsh and Webb 2013), given the low degree of riparian clearing along the streams. Physids thus contributed to the relatively low observed LUMaR scores in all sites, but not to the even lower scores observed downstream of the reservoir.

Recovery downstream of the reservoir

The more downstream, ephemeral reduced sites of Pyrites Creek tended to be more similar to sites upstream of the reservoir and to sites in Goodmans Creek (Figs. 3, 4), which is likely driven by the reduced numbers of Planorbidae (Fig. 7C), Tipulidae (Fig. 7E) and Notonemouridae (Fig. 7F), and increased numbers of Notonectidae (Fig. 8C).

Patterns attributable to augmented flows in the 'ephemeral enhanced' streams

The MDS plots show the ephemeral enhanced sites to group separately from the ephemeral reduced and ephemeral natural sites, in the same direction along the second MDS axis as the permanent sites in lower Pyrites Creek (Fig. 4). No difference in assemblage condition as indicated by LUMaR or SIGNAL was evident (Fig. 6). However several families showed increased abundances in ephemeral enhanced sites, but two of these—hydrobiid snails and aeshnid dragonflies—also showed increased abundances in permanent sites downstream of the reservoir (Fig. 9A, B). These similarities between ephemeral enhanced and permanent sites could explain the similarity between the two groups of sites suggested by the log(x+1) MDS (Fig. 4), that was less evident in the presence-absence MDS (Fig. 3).

Five other families—ancylid limpets, Psephenidae ('water penny' beetles), Corixidae (water boatmen), and philopotamid and hydropsychid caddisflies (Fig 8C–G)—showed an increase in abundance in ephemeral enhanced sites compared to surrounding sites, and a contrasting absence in permanent sites downstream of the reservoir (although the absence downstream of the reservoir is likely driven by a lack of riffles). This increased abundance of a number of families likely resulted in an increased evenness in abundance of families, which was reflected in an increased family richness when sample size is controlled (Fig. 9H: showing EF400, which showed similar trends to EF200 and EF600).

Longitudinal effects

The most upstream site in each creek was the most similar to the degraded sites downstream of the reservoir (Figs. 3, 4). This similarity is reflected by both LUMaR and SIGNAL scores for the sites, which were similarly low to the sites downstream of the reservoir (Fig. 5). LUMaR scores broadly increase with distance downstream in both Goodmans and Pyrites Creek upstream of the reservoir. The most upstream sites are marked by an absence of families that are also absent from sites downstream of the reservoir—Atyidae, Gyrinidae (Fig 7A, E), Ancylidae, Corixidae, Psephenidae, and Hydropsychidae (Fig. 9C,D, E, G)—and large numbers of families that are also abundant in sites downstream of the reservoir—Physidae and Tipulidae (Fig. 7E, G).

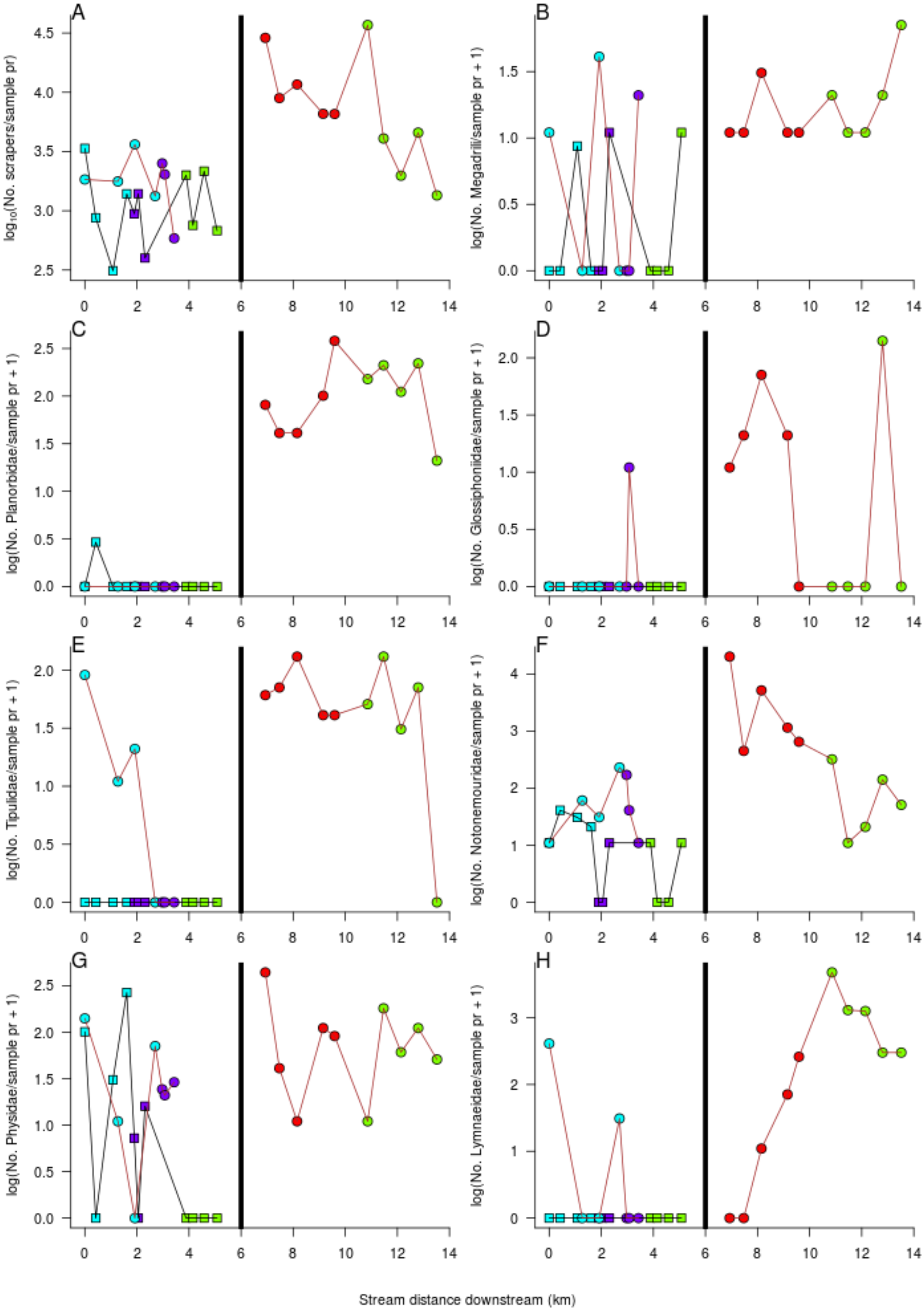

Fig. 7. Functional feeding group and families that were more abundant downstream of Merrimu Reservoir than in other sites. Trends with distance downstream along Goodmans and Pyrites Creeks in abundance of scrapers and 7 families (B. Megadrili oligochaete worms, C. Planorbid snails [scrapers], D. Glossiphonid leeches, E. Tipulid fly larvae, F Notonemourid stoneflies [scrapers], G. Physid snails [scrapers], and H. Lymnaeid snails [scrapers]). Symbol conventions as in Fig 4

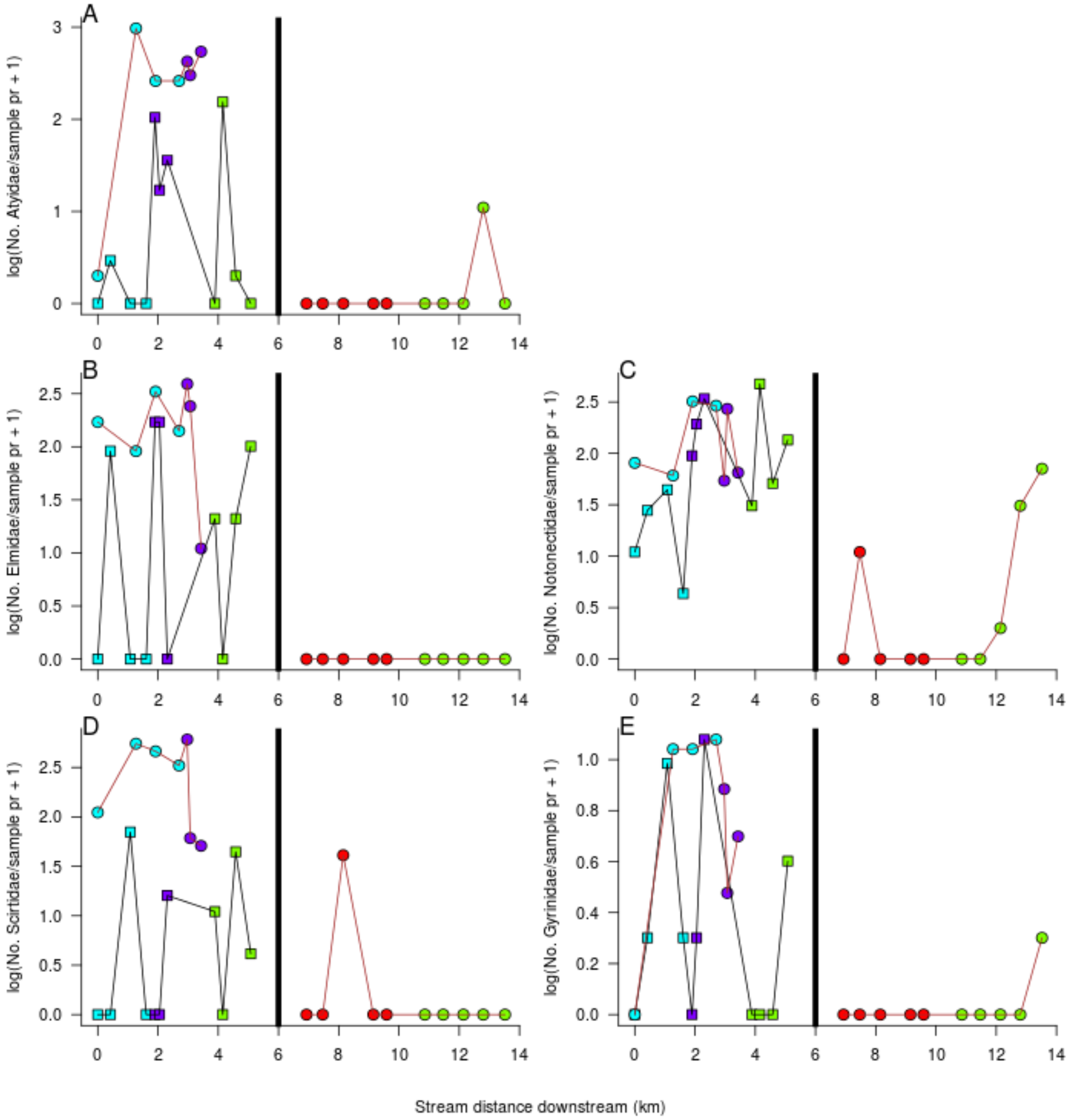

Fig. 8. Families that were less abundant downstream of Merrimu Reservoir than in other sites. Trends with distance downstream along Goodmans and Pyrites Creeks in abundance of A. Atyid shrimps, B. Elmidae (riffle beetles), C. Notonectidae (backswimmers), D. Scirtid beetles, E. Gyrinidae (whirligig beetles). Symbol conventions as in Fig 4

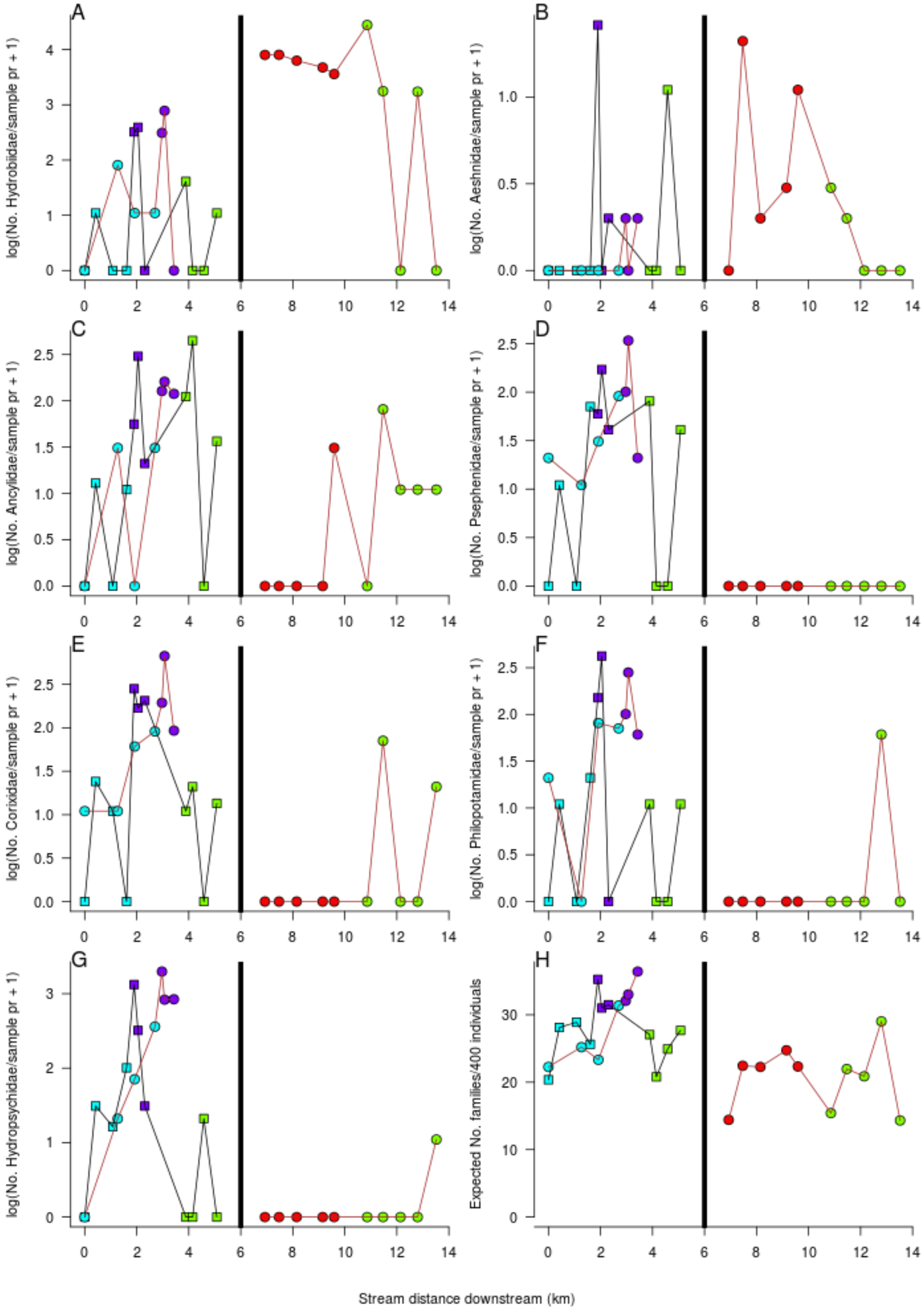

Fig. 9. Families that were more abundant in ephemeral enhanced sites than surrounding sites, and expected family richness for a count of 400 individuals, which showed a similar trend. Trends with distance downstream along Goodmans and Pyrites Creeks in abundance of A. Hydrobiid snails, B. Aeshnid dragonflies, C. Ancylid limpets, D. Psephenidae (water penny beetles), E. Corixidae (water boatmen), F. Philopotamid caddisflies, G. Hydropsychid caddisflies, H. Expected family richness for 400 individuals. Symbol conventions as in Fig 4

Discussion

Intermittent streams of this region are characterized by substantial variation in assemblage composition over the annual wetting and drying cycle, typically with an initial gradual increase in densities, first of scrapers and later of shredders and predators, followed by a rapid decline in densities as pools dry (Boulton and Lake 1992a). It is possible that the reduced condition of the most upstream sites, as indicated by LUMaR and SIGNAL, could be a result of those sites being at a later stage of the drying cycle or earlier stage of the wetting cycle at the time of sampling than were sites located further downstream. The increased family richness of assemblages in the ephemeral enhanced sites, with their slightly longer periods of flow (Table 1) suggests that assemblages in these sites could have an extended period of community assembly before the decline of the drying period, or than the community assembly is aided by improved retention of and ultimately connectivity with refuge pools. Additionally, the increased flow contributes to increased diversity of microhabitats associated with increased depth, surface area and hydraulic variability. In Pyrites Creek, the 'ephemeral enhanced' sites were characterized by higher channel flow status (i.e. a greater proportion of available channel and riffle substrate was inundated), higher dissolved oxygen (8.4–9.6 mg/L in 'ephemeral enhanced sites compared with 6.6–7.6 mg/L in 'ephemeral natural' sites), higher pH (8.3–8.6 compared with 7.0–7.7). Similar differences were noted between the 'ephemeral natural' and ephemeral enhanced sections of Goodmans Creek

Seasonal variation in assemblage composition could explain the lower than predicted scores for SIGNAL and LUMaR. It is likely that intermittent streams, such as these, were under-represented in the data used by Walsh and Webb (2013) in developing their models, since permanently flowing streams comprised the majority of Melbourne Water's macroinvertebrate monitoring program (E.Tsyrlin pers. comm.). Future model development using additional data from intermittent streams such as those of this study could allow improved predictions in the future. However, the abundance of a range of 'weedy' (sensu Walsh and Webb 2013: families that respond positively to one or more human impacts) and invasive families in all of the sites of this study, point to some level of degradation not related to intermittency. It is possible that current assemblage composition could be influenced by past land-use legacies, such as grazing, but particularly mining, which persisted in the study catchments until the 1940s. Regardless of the cause, the large degree of variation in assemblage composition among 'ephemeral natural' sites suggests a need for greater within season temporal replication of sampling to allow robust inference of causes of longer-term change.

Macroinvertebrate assemblages of intermittent streams are strongly controlled by their flow disturbance regimes (Brooks 1998), and the small leaking release from the reservoir creating the permanent downstream section of Pyrites Creek, greatly reduces both the variability of flow and the frequency of disturbance. It is likely that such a flow regime would promote habitat changes via sediment accumulation, together with algal and macrophyte growth, driving the observed prevalence of scrapers in the macroinvertebrate community. The relative lack of forest (particularly on the western banks) upstream of sites near the reservoir, and the resultant reduction in allochthonous organic matter inputs, could also contribute to the dominance of algae as the basis of the food web in these reaches. However, LUMaR and SIGNAL scores point to changes in assemblage composition far beyond what would be expected as a result of reduced upstream riparian forest alone.

There are marked differences in habitat between the downstream Pyrites Creek reaches ('permanent' and 'ephemeral reduced') and all other reaches that appear to be the direct result of sediment accumulation. The sediment accumulation appears to have resulted in subsequent excessive (choking) growth of the channel with reeds (*Typha* spp. and to a lesser extent *Phragmites australis*)-greater than 90% cover of the available channel at all sites except the two most downstream sites (PCER4 and PCER5: 6590%)—which likely exacerbates subsequent sediment accumulation. The natural streambed appears to have been progressively buried as a result of this process- 95-100% cover of the streambed consists of silt between the reservoir and site PCER3, and the reduction in substrate and depth variability appears to have substantially reduced the diversity of available habitats and microhabitats for macroinvertebrates.

The influence of the marked habitat differences on the macroinvertebrate community assemblages is expected to be considerable, however the 2013 macroinvertebrate assemblage data provides hope that environmental flow releases that are sufficient to scour algae and biofilms from the stream-bed may result in a shift in macroinvertebrate assemblages from scraper-dominated assemblages, to an increased abundance of other functional feeding groups. Such assemblages should be more similar to the ephemeral sections upstream. Valve constraints preclude geomorphically significant environmental or operational releases that could rework the bed and remove in-stream vegetation (B. Moulden pers. comm.), so substantial scouring and remediation of downstream Pyrites Creek instream habitat to a state approaching that of the other reaches, would require a natural event large enough to overtop the Merrimu Reservoir spillway. Notwithstanding the marked habitat differences, the strong, consistent differences in assemblage composition downstream of the reservoir detected in this analysis suggest that the potential for detecting differences could be high. It should be noted that the absence of baseline data from before the commencement of environmental releases in mid 2012 reduces the robustness of inferences possible from the study.

Some recommendations

The data for many families suggest the sites are closely spaced enough to be exhibiting spatial autocorrelation, thus presenting analytical challenges. The non-independence of the sites calls for a Bayesian approach (to account for spatial autocorrelation and complex interactive effects) to analyse trends for subsequent stages of the project; when the aim will be to assess if environmental flow releases from the reservoir have changed the assemblage composition of sites downstream and in which way.

It would be worth separating the distinguishing between the invasive *Potamopyrgus antipodarum* and the diverse group of indigenous hydrobiid snails (Ponder et al. 1993) during sample identification. This will be particularly important if downstream of the reservoir is dominated by *P. antipodarum*, and upstream by indigenous species.

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