Visually determined stream mesohabitats influence benthic macroinvertebrate assessments in headwater streams

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Abstract Mesohabitat components such as substrate and surface flow types are intimately related to benthic macroinvertebrate assemblages in streams. Visual assessments of the distribution of these components provide a means of evaluating physical habitat heterogeneity and aid biodiversity surveys and monitoring. We determined the degree to which stream site and visually assessed mesohabitat variables explain variability (i.e., beta-diversity) in the relative abundance and presence-absence of all macroinvertebrate families and of Ephemeroptera, Plecoptera, and Trichoptera (EPT) genera. We systematically sampled a wide variety of mesohabitat arrangements as they occured in stream sites. We also estimated how much of the explanation given by mesohabitat was associated with substrate or surface flow types. We performed variation partitioning to determine fractions of explained variance through use of partial redundancy analysis (pRDA). Mesohabitats and stream sites explained together from 23 to 32 % of the variation in the four analyses. Stream site explained 8–11 % of that variation, and mesohabitat variables explained 13–20 %. Surface flow types accounted for >60 % of the variation provided by the mesohabitat component. These patterns are in accordance with those obtained in previous studies that showed the predominance of environmental variables over spatial location in explaining macroinvertebrate distribution. We conclude that visually assessed mesohabitat components are important predictors of assemblage composition, explaining significant amounts of beta-diversity. Therefore, they are critical to consider in ecological and biodiversity assessments involving macroinvertebrates.

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Introduction

Stream systems are arranged in a hierarchical spatial structure in which fluvial patterns and processes can be evaluated (Frissel et al. 1986; Allan 2004). We can assess physical habitat and the associated biological assemblages at multiple spatial scales, ranging from microhabitat (centimeters) to catchment (kilometers) (Wiens 2002; Carbonneau et al. 2012). In this hierarchical arrangement, studies at the mesohabitat scale are of fundamental importance for understanding the distribution of aquatic organisms (Beisel et al. 2000). Such

studies range between microhabitats (organic and inorganic substrates) and habitat types (riffles and pools) (Armitage et al. 1995; Armitage and Pardo 1995; Kemp et al. 2000). Mesohabitats are defined subjectively as apparently uniform and visually distinct habitat units (Armitage et al. 1995; Armitage and Pardo 1995) promoted by interaction between hydrological and geomorphological forces (Tickner et al. 2000; Jähnig et al. 2009). Visual assessments of the distribution of mesohabitats provide a means of evaluating the physical habitat heterogeneity of stream reaches and are important components in biodiversity and bioassessments (Boyero 2003; Kubíková et al. 2012).

Previous authors have demonstrated the usefulness of mesohabitats as ecological study units in streams (Pardo and Armitage 1997; Beisel et al. 2000; Tickner et al. 2000). Armitage and Cannan (1998) identified a number of mesohabitat types characterized by particular macroinvertebrate assemblages. Organic (roots, macrophytes, marginal plants) and inorganic (silt, sand, gravel, cobbles) substrates are considered key mesohabitat components. In addition to substrates, surface flows (riffles, glides, pools) are important mesohabitat elements in characterizing benthic macroinvertebrate assemblages (Buss et al. 2004; Reid and Thoms 2008). Rigorous biomonitoring protocols account for these characteristics when assessing stream conditions using macroinvertebrates (Barbour et al. 1999; Hering et al. 2004; Hughes and Peck 2008).

Biological variations among sites are often described in terms of their beta-diversity, as initially proposed by Whittaker (1960). Beta-diversity contrasts with the analysis of alpha-diversity, which is the amount of diversity (e.g., taxonomic richness) at a site (Jurasinski et al. 2009; Anderson et al. 2011). Despite the preponderance of studies describing patterns of local aquatic diversity (Clarke et al. 2010), beta-diversity is an important component characterizing diversity distribution across riverscapes (Wiens 2002; Stendera and Johnson 2005; Ligeiro et al. 2010). Because of differences in physical habitat characteristics, water quality, colonization history, and frequency and magnitude of disturbances, different stream sites may have different biological assemblages (Melo and Froelich 2001; Hughes et al. 2010; Ligeiro et al. 2010; Kovalenko et al. 2012). Actually, stream macroinvertebrate assemblages are driven by factors acting at multiple spatial scales (Bonada et al. 2008; Heino 2011). The metacommunity concept explicitly recognizes the importance of the regional context for explaining local assemblages and the variation among them (Leibold et al. 2004; Thompson and Townsend 2006; Brown et al. 2011). Therefore, in addition to mesohabitat differences, differences in site location help us to understand assemblage variation (Legendre et al. 2005; Landeiro et al. 2012). Some multivariate analyses decompose the relative influence of environmental (e.g., mesohabitat characteristics) and spatial factors (e.g., site location) (Legendre et al. 2005; Costa and Melo 2008), which help us define what is important in structuring assemblages. Such analyses, when based on partial canonical correspondence analysis (CCA) or partial redundancy analyses (pRDAs), are referred as the "raw data approach" (Legendre et al. 2005) and are concerned with the explanation of variation in assemblage composition among sites ("explaining beta-diversity," as defined by Tuomisto and Ruokolainen (2006)).

To improve and implement monitoring programs focused on rehabilitating and protecting water bodies, it is necessary to understand which factors influence benthic macroinvertebrate assemblages at different spatial scales (Roque et al. 2010; Siqueira et al. 2012). Understanding the associations between benthic macroinvertebrate assemblages and the different mesohabitat units that comprise stream sites is essential for predicting and evaluating the effects of human pressures on aquatic ecosystems (Principe et al. 2007; Moya et al. 2011). The visual assessment of mesohabitat components is a common practice in ecological assessments and biomonitoring programs. These qualitative methods are described in many field protocols (e.g., Hering et al. 2004; Peck et al. 2006), and they are important for assessing habitat characteristics in a reasonable amount of time (Hughes and Peck 2008). Although several studies have demonstrated that different substrate or surface flow types result in different assemblage compositions (Wang et al. 2006; Duan et al. 2008; Reid and Thoms 2008), most studies do not analyze both components together or do not consider a wide range of substrate and surface flow types, thereby poorly representing natural patterns. Therefore, the objective of this study was to evaluate how visually assessed mesohabitat components explain variation in the composition (i.e., beta-diversity) of macroinvertebrate assemblages using a dataset covering a wide range of mesohabitat typologies (i.e., substrate and surface flow types). We hypothesized that mesohabitat variation strongly influences macroinvertebrate composition, independent of the stream site location.

Methods

Study area

We studied stream sites in the upper São Francisco Basin, southeastern Brazil (Minas Gerais state) (44° 30′ 0″ W–46° 0′ 0″, 17° 0′ 0″ S–19° 30′ 0″ S). The study area is in the Cerrado biome, the second most extensive biome of the Neotropics, covering originally almost 20 % of Brazil (Wantzen 2003). The constant anthropogenic threat to this biome, together with its high biodiversity and number of endemic species, makes the Cerrado a global biodiversity "hotspot" (Myers et al. 2000). The Cerrado climate is defined by a rainy season (October to March) and a dry season (April to September) with annual precipitation of 1,200 to 1,800 mm and mean daily temperature ranging from 22 to 27 °C.

Field sampling and laboratory procedures

We sampled sites in September 2010, during the end of the dry season, when discharge is constant, habitats are more exposed, and macroinvertebrate abundances are usually high (Bispo et al. 2001). We randomly selected 12 wadeable stream sites belonging to different microbasins in the upper São Francisco Basin. The catchments of these stream sites were good to slightly altered by agriculture and pasture, and there was no evidence of major anthropogenic alterations in the stream beds, banks, or riparian zones.

At each stream site, we established a longitudinal reach equal to 40 times the mean wetted width (or a minimum length of 150 m) and collected 11 equidistant benthic macroinvertebrate samples via a systematic zig-zag pattern along the site (Peck et al. 2006; Hughes and Peck 2008). The samples were kept separate for subsequent identifications and analyses. We used a D-frame kick net (30-cm mouth width, 0.09-m^2 sampled area, 500-μm mesh) to collect the samples, taking care to clean the net between samples, in this way avoiding collection contamination.

We fixed the samples with 10 % formalin and took them to the laboratory, where the macroinvertebrates were sorted by eye and identified. The identification was primarily done to family level using identification keys (Pérez 1988; Merritt and Cummins 1996; Fernández and Domínguez 2001; Costa et al. 2006; Mugnai et al. 2010). Hydracarina, Oligochaeta, Bivalvia, Nematoda, and Hirudinea were not identified

to family. Those groups represented together <5 % of the total number of individuals and were considered in the analyses together with the families. Therefore, we will call hereafter the whole set of taxa found as families. Organisms of the insect orders Ephemeroptera, Plecoptera, and Trichoptera (EPT) were identified to genus with the help of the same identification keys. This allowed us to interpret results at two taxonomic levels: total families and EPT genera.

At each of the 11 benthic sampling stations, we measured current velocity and visually classified the mesohabitats according to substrate and surface flow types following Peck et al. (2006). The predominant substrate was classified as fine (silt and clay), sand, gravel, cobble, boulder, or leaf pack. The surface flow type was classified as pool, slow glide, fast glide, riffle, or rapid (Tables 1 and 2).

Data analysis

The variation-partitioning method proposed by Borcard et al. (1992) decomposes the variation of taxonomic composition data into independent components: pure spatial, pure environmental, shared by spatial and environmental, and undetermined. According to Borcard et al. (1992), the environmental component can be represented by any habitat variable able to determine changes in the biological assemblage (Borcard et al. 1992; Legendre et al. 2005). The spatial component, based on geographic coordinates or site location, estimates the contribution of spatial variation in structuring assemblages, filtering out the effects of spatial correlation on the environmental variables (Peres-Neto and Legendre 2010).

In our study, the environmental component was represented by the categorical mesohabitat variables (the

Table 1 Description of substrates sampled (adapted from Peck et al. 2006)

Substrates	Size range	Description
Fines	<0.06	Silt and clay
Sand	>0.06 to 2 mm	Smaller than ladybug size but visible as particles
Gravel	>2 to 64 mm	Ladybug to tennis ball size
Cobble	>64 to 250 mm	Tennis ball to basketball size
Boulder	>250 mm	Basketball to car size
Leaf pack		

Table 2 Description of surface flow types sampled (adapted from Peck et al. 2006). Mean current velocity (m s^{-1}) and standard deviation (±SD) of each surface flow type obtained through field measurements

Surface low types	Description	Current velocity m s^{-1} (±SD)
Pool	Surface form appears stationary, smooth surface, usually deep	0
Glide	Very little surface turbulence, water moving slowly, unbroken surface	Slow 0.02 (±0.04)
(slow and fast)		Fast 0.21 (±0.16)
Riffle	Water moving, with small ripples, waves and eddies, waves not breaking	0.43 (±0.26)
Rapid	Water movement rapid and turbulent, surface with intermittent white water and breaking waves	0.69 (±0.38)

various substrate and surface flow types determined visually). We treated the mesohabitat components (substrates and surface flows) as independent variables because we did not observe a dependent relation between them in the field (i.e., a variety of substrate types were found in different flow types). Because nearby streams in Cerrado microbasins can harbor very dissimilar macroinvertebrate assemblages (Ligeiro et al. 2010), we were more interested in the among-site variation than in the variation in geographic distances among sites. Therefore, the spatial component was represented by the site identities and treated as a categorical variable in the analyses (site 1 to site 12) (following Costa and Melo 2008). The macroinvertebrate abundance data were Hellinger transformed (Legendre and Gallagher 2001) and treated as relative abundances. This transformation provides unbiased and accurate estimates of the explained fraction of variance through use of pRDA (Peres-Neto et al. 2006).

A first variation partitioning yielded four fractions of explained total variation: (i) variation due only to environmental factors, (ii) variation due only to spatial factors, (iii) variation shared between environmental and spatial factors, and (iv) the variation that remained unexplained. The fractions were expressed as the percentage of explanation of the total variation. A second variation partitioning was conducted with only the environmental factors and produced three fractions: (i) variation explained purely by substrate variables, (ii) variation explained purely by surface flow type variables, and (iii) the variation shared between them. These fractions were expressed as the percentage of explanation given by the environmental variation fraction in the first partition.

We assessed benthic macroinvertebrate assemblage composition in two ways: relative abundance and presence-absence. Presence-absence data concerns only the incidence of taxa in the sites, being particularly valuable in biodiversity assessments and conservation studies. On the other hand, relative abundance data details aspects of dominance among taxa. The two different measures of composition are important for understanding biological patterns (Anderson et al. 2011). Taxonomic resolution was evaluated as total assemblage families and EPT genera. These different assessments resulted in four separate analyses for both total variation and environmental variation alone. In both partitions, the explained fractions were interpreted via adjusted R^2. Given the influence of the number of variables and sample sizes in the analyses, the adjusted R^2 provides an unbiased estimation of the explained fractions (Peres-Neto et al. 2006). We performed significance tests of the fractions (spatial and environmental fractions in the first set of partitions, surface flow and substrate types in the second set of partitions) with 1,000 permutations at a significance level of 0.05, to test if each fraction contributed significantly to explaining assemblage distribution. The analyses were all performed in *R* (R Development Core Team 2013), through the vegan (Oksanen et al. 2013) and dummies (Brown 2011) packages.

Results

In the 12 sites, we collected a total of 27,333 organisms distributed in 65 families; 8,493 individuals were EPT, represented by 58 genera. The most abundant families were Chironomidae (32 % of the total families), Simuliidae (20 %), and Baetidae (10 %). Of the total EPT identified, 23 % were

Traveryphes (Ephemeroptera), 12 % were *Americabaetis* (Ephemeroptera), and 9 % were *Chimarra* (Trichoptera).

Total variation explained

The total explained variation (sum of environmental and spatial fractions) was 32 % for family relative abundances and 23 % for family presence-absence data. Similar values were found when considering only EPT genera: 30 and 24 %, respectively, for relative abundance and presence-absence data (Fig. 1). All fractions of explained variation (spatial and environmental in the first partitioning, surface flow and substrate types in the second partitioning) were significant ($p < 0.001$).

The environmental component explained 20 % of the total variation of the relative abundances of families and 13 % of their presence-absence. Regarding EPT genera, environmental data explained 17 % of relative abundances and 14 % of presence-absence variation (Fig. 1). The fraction explained purely by the spatial component was lower than that explained by the environmental component: 11 and 8 % for relative abundances and presence-absence data, respectively, when considering both families and EPT genera analyses.

Spatial and environmental components shared low percentages of explained variation. In all cases (families and EPT genera relative abundances and presence-absence data), approximately 2 % of the shared variation was explained (Fig. 1). The unexplained percentage was 68 and 77 % for the relative abundance and presence-absence of families, respectively, and 70 and 76 % for

the relative abundance and presence-absence of EPT genera, respectively.

Decomposition of the environmental variation

The second partition was performed to evaluate the separate effects of substrate and surface flow types on the total explanation given by the environmental fraction (Fig. 2). Surface flow types explained most of the environmental variation in family composition: 61 % for relative abundances and 48 % for presence-absence versus 22 and 31 % given by substrate types, respectively. Likewise, for EPT genera, surface flow types explained 68 % of the relative abundance variation and 61 % of the presence-absence variation versus 14 and 21 % for the substrate types, respectively (Fig. 2). Percentages of shared explanation between surface flow and substrate types for the relative abundances and the presence-absence were 21 and 17 % for families, respectively, compared to 18 % for both relative abundance and presence-absence of EPT genera.

Discussion

Total variation explained

Our analyses allowed us to evaluate the percentage of variation explained by environmental variables (mesohabitat) and the spatial component (stream site identities) and to distinguish the amount of environmental variation explained purely by substrate, purely by

Fig. 1 Fractions of variation explaining variability of **a** abundance and **b** presence-absence of families and EPT genera, expressed in percentages (%). *[S]* indicates the fraction of variation explained by the spatial factor, stream site identities; *[E]* is the fraction of variation explained purely by the environmental factor, mesohabitat components; *[S+E]* is the fraction shared between environmental and spatial factors; and *[R]* is the residual fraction that is not explained by any factor evaluated
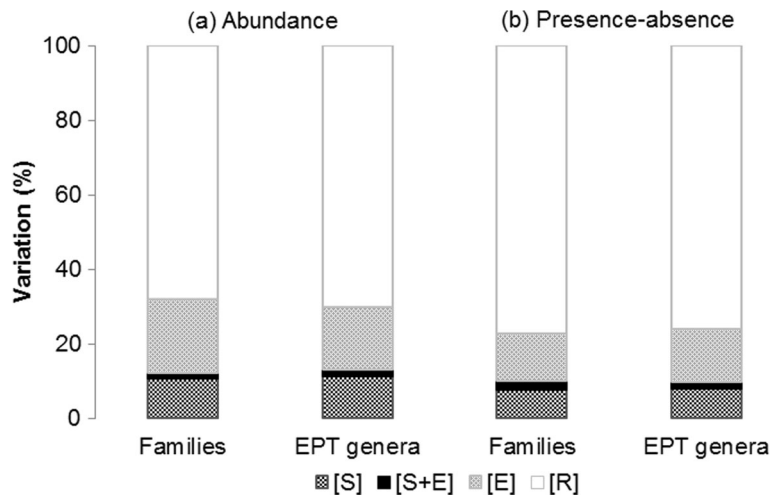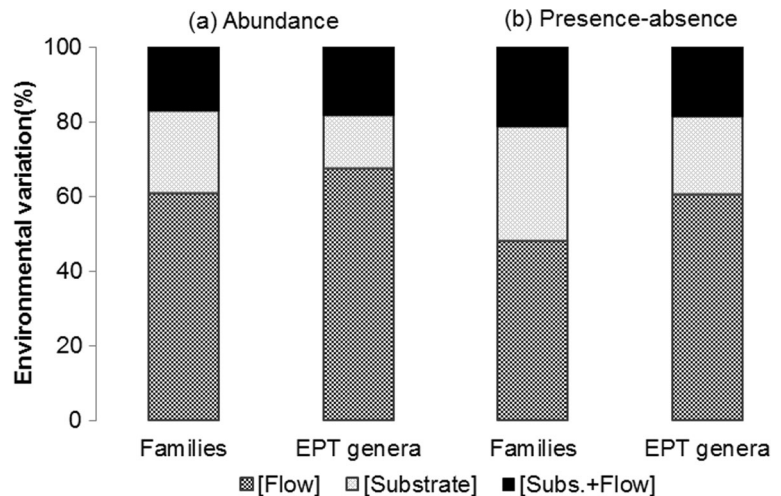
Fig. 2 Fractions of the pure environmental factor explaining the variation of **a** abundance and **b** presence-absence of families and EPT genera, expressed in percentages (%) of the total mesohabitat variation

surface flow type, and that shared between the two. We observed that environmental and spatial factors together explained 23 to 32 % of the variation in the four analyses. The percentage of variance explained when analyzing relative abundance was usually greater than that of presence-absence data. In comparison with presence-absence data, abundance data provide more complete information regarding taxonomic composition, assemblage size, and relative abundances of taxa (Beisner et al. 2006). Similar results were observed in studies of fish assemblages (Sály et al. 2011), bird assemblages (Cushman and McGarigal 2004), and fish, zooplankton, phytoplankton, and bacteria assemblages (Beisner et al. 2006).

We consider the total explanation given by the models substantial, given that we only analyzed substrate and surface flow variables. Actually, several potential explanatory environmental variables were intentionally not included in our analyses so that we could focus on the explanation provided by the mesohabitat variables that are easily assessed visually. Others have shown the importance of additional environmental variables on structuring macroinvertebrate assemblages such as stream size and microhabitat features (Ligeiro et al. 2013), depth, channel width (Mykrä et al. 2007; Jiang et al. 2010; Moya et al. 2011), riparian vegetation structure (Rios and Bailey 2006), coarse particulate organic matter (Hepp et al. 2012), and periphyton (Schneck et al. 2011). Still, even when considering a great number and variety of explanatory variables, the amounts of explanation might not be high due to unmeasured controlling variables, complex spatial

interactions, and stochastic and nondeterministic fluctuations in the assemblages (Borcard et al. 1992; Heino 2005). Many studies using the variation-partitioning approach have found amounts of explanation less than 30 % (Johnson and Goedkoop 2002; Beisner et al. 2006; Mykrä et al. 2007; Landeiro et al. 2012; Grönroos et al. 2013).

The visual assessment of mesohabitat explained more of the variation than stream site identities, corroborating our initial hypothesis. The different macroinvertebrate taxa usually occur in particular mesohabitats partly because food availability, habitat heterogeneity, and refuge are driven by the interaction of substrate and surface flow (Principe et al. 2007). The importance of these environmental factors for explaining variation in macroinvertebrate assemblages has been reported in other studies in streams (Johnson et al. 2004; Ligeiro et al. 2010; Moya et al. 2011; Hepp et al. 2012; Landeiro et al. 2012; Ligeiro et al. 2013), rivers (Angradi et al. 2006; Jiang et al. 2010), and lakes (Johnson and Goedkoop 2002; Johnson et al. 2004).

The relative importance of mesohabitat and spatial factors for explaining macroinvertebrate variation can be translated in terms of beta-diversity. We found that mesohabitat factors were of greater importance in generating beta-diversity in macroinvertebrate assemblages than stream site identities. This means that assemblages occupying the same habitat types in different sites are likely to be more similar to each other than to assemblages occupying different habitat types in the same site. Costa and Melo (2008) obtained similar results when analyzing beta-diversity of macroinvertebrates in

Neotropical streams. Although they reported greater total explanation (sum of environmental + spatial fractions), the proportional amounts of variation explained by environmental and spatial components were similar between their study and ours. The differences in the total amounts of explanation likely result from different study designs. They used a sample-balanced study on four microhabitats in three Neotropical stream sites. We used a greater combination of factors (substrate and surface flow types) sampled in a systematic manner at 12 randomly selected sites. Our methodology aims to better represent natural patterns but also generates more noise in the analysis.

Although providing less explanation, the spatial component (among-site variation) significantly increased the explanation of macroinvertebrate assemblage composition. Sály et al. (2011) also observed that addition of a spatial factor increased the explanation of fish assemblage composition. The percentage of shared fractions between environmental and spatial factors was low (~2 % in the four analyses). This can be attributed to the low structuring effect of the site with respect to environmental descriptors in our study (Borcard et al. 1992), i.e., the different types of substrate and surface flow were not associated with particular sites. It also indicates that environmental and spatial factors may act separately in structuring benthic assemblages (Hepp et al. 2012).

Environmental variation explained

In the second partition, surface flow type explained >60 % of the variation in the environmental component in most cases. The response of EPT assemblages was slightly more sensitive than families to surface flow type (61 and 68 % for relative abundances and presence-absence data, respectively). Surface flow types are of fundamental importance to biota, modifying physical habitat, setting levels of hydraulic stress, and influencing concentrations of dissolved gases and nutrients (Reid and Thoms 2008). Pastuchová et al. (2008), studying only EPT assemblages, concluded that surface flow types were the most important variable affecting the distribution of EPT genera. Duan et al. (2008) also found that surface flow types discriminated macroinvertebrate assemblages. According to Newson and Newson (2000), surface flow types represent the hydraulic conditions of the riverbed, making them important components for ecological studies. Actually, many macroinvertebrate assessments target sampling to a particular surface flow type (e.g., riffle) to reduce the effects of interhabitat variation so that they can better assess water quality degradation (Wang et al. 2006; Gerth and Herlihy 2006).

The proportion explained by substrate types was smaller but still important in explaining variation in macroinvertebrate assemblages. Many other variation-partitioning studies have indicated the importance of substrates as a predictor of benthic macroinvertebrate composition in different ecosystems (Rempel et al. 2000; Johnson and Goedkoop 2002; Johnson et al. 2004; Mykrä et al. 2007; Jiang et al. 2010). Substrate types regulate habitat complexity, food availability, and refuge against predators and flow disturbance (Vinson and Hawkins 1998; Verdonschot 2001; Allan and Castillo 2007). The different substrate types also affect periphyton colonization and the amount of organic matter that accumulates in the streambed (Graça et al. 2004; Hepp et al. 2012). Minshall (1984) considered substrate as the primary factor driving the abundance and distribution of aquatic insects. However, he emphasized the need to investigate the interaction of substrate with other abiotic factors, such as hydraulic components. Principe et al. (2007) also concluded that, more than each factor separately, the mutual consideration of mesoscale geomorphological and hydrological conditions explains benthic macroinvertebrate assemblage structure.

Conclusions

The variation of mesohabitat factors within a given site was more important in generating beta-diversity in assemblages than the variation among sites. The relative importance of visually assessed substrate and surface flow variables explaining macroinvertebrate composition has been rarely evaluated in previous studies. Although surface flow explained a greater proportion of this variation, substrate also contributed significantly. Consequently, mesohabitat heterogeneity is of key importance for sustaining biological diversity and hydrogeomorphological integrity in headwater streams. We demonstrated through our analyses that beta-diversity can be maximized when sampling designs incorporate the widest possible combination of mesohabitat types (versus focusing on a single mesohabitat type), in this way enhancing macroinvertebrate assessments.

References

Allan, J. D. (2004). Landscape and riverscapes: the influence of land use on river ecosystems. *Annual Reviews of Ecology, Evolution and Systematics, 35*, 257–284.

Allan, J. D., & Castillo, M. M. (2007). *Stream ecology: structure and function of running waters. 2nd edn*. Dordrecht: Springer.

Anderson, M. J., Crist, T. O., Chase, J. M., Vellend, M., Inouye, B. D., Freestone, A. L., et al. (2011). Navigating the multiple meanings of β diversity: a roadmap for the practicing ecologist. *Ecology Letters, 14*, 19–28.

Angradi, T. R., Schweiger, E. W., & Bolgrien, D. W. (2006). Inter-habitat variation in the benthos of the Upper Missouri River (North Dakota, USA): implications for great river bioassessment. *River Research and Applications, 22*, 755–773.

Armitage, P. D., & Cannan, C. E. (1998). Nested multi-scale surveys in lotic systems: tools for management. In G. Bretschko & J. Helesic (Eds.), *Advances in River Bottom Ecology* (pp. 293–314). Leiden: Backhuys Publishers.

Armitage, P. D., & Pardo, I. (1995). Impact assessment of regulation at the reach level using macroinvertebrate information from mesohabitats. *Regulated Rivers: Research & Management, 10*, 147–158.

Armitage, P. D., Brown, A., & Pardo, I. (1995). Temporal constancy of faunal assemblages in 'mesohabitats'— application to management? *Archiv für Hydrobiologie, 133*, 367–387.

Barbour, M. T., Gerritsen, J., Snyder, B. D., & Stribling, J. B. (1999). *Rapid bioassessment protocols for use in streams and wadeable rivers: periphyton, benthic macroinvertebrates and fish* (2nd ed.). Washington, DC: EPA 841-B-99–002. Office of Water, US Environmental Protection Agency.

Beisel, J. N., Usseglio-Polatera, P., & Moreteau, J. C. (2000). The spatial heterogeneity of a river bottom: a key factor determining macroinvertebrate communities. *Hydrobiologia, 422*(423), 163–171.

Beisner, B. E., Peres-Neto, P. R., Lindström, E. S., Barnett, A., & Longhi, M. L. (2006). The role of environmental and spatial processes in structuring lake communities from bacteria to fish. *Ecology, 87*, 2985–2991.

Bispo, P. C., Oliveira, L. G., Crisci, V. L., & Silva, M. M. (2001). A pluviosidade como fator de alteração da entomofauna bentônica (Ephemeroptera, Plecoptera e Trichoptera) em córregos do Planalto Central do Brasil. *Acta Limnologica Brasiliensia, 13*, 1–9.

Bonada, N., Rieradevall, M., Dallas, H., Davis, J., Day, J., Figueroa, R., et al. (2008). Multi-scale assessment of macro-invertebrate richness and composition in Mediterranean-climate rivers. *Freshwater Biology, 53*, 772–788.

Borcard, D., Legendre, P., & Drapeau, P. (1992). Partialling out the spatial component of ecological variation. *Ecology, 73*, 1045–1055.

Boyero, L. (2003). The quantification of local substrate heterogeneity in streams and its significance for macroinvertebrates assemblages. *Hydrobiologia, 499*, 161–168.

Brown, C. (2011). Dummies: create dummy/indicator variables flexibly and efficiently. R package version 1.5.4. http://CRAN.R-project.org/package=dummies.

Brown, B. L., Swan, C. M., Auerbach, D. A., Grant, E. H. C., Hitt, N. P., Maloney, K. O., et al. (2011). Metacommunity theory as a multispecies, multiscale framework for studying the influence of river network structure on riverine communities and ecosystems. *Journal of the North American Benthological Society, 30*, 310–327.

Buss, D. F., Baptista, D. F., Nessimian, J. L., & Egler, M. (2004). Substrate specificity, environmental degradation and disturbance structuring macroinvertebrate assemblages in neotropical streams. *Hydrobiologia, 518*, 179–188.

Carbonneau, P., Fonstad, M. A., Marcus, W. A., & Dugdale, S. J. (2012). Making riverscapes real. *Geomorphology, 137*, 74–86.

Clarke, A., Macnally, R., Bond, N., & Lake, P. S. (2010). Conserving macroinvertebrate diversity in headwater streams: the importance of knowing the relative contributions of α and β diversity. *Diversity and Distributions, 16*, 725–736.

Costa, S. S., & Melo, A. S. (2008). Beta diversity in stream macroinvertebrate assemblages: among-site andamong-microhabitat components. *Hydrobiologia, 598*, 131–138.

Costa, C., Ide, S., & Simonka, C. E. (2006). *Insetos Imaturos: Metamorfose e Identificação*. Ribeirão Preto: Holos.

Cushman, S. A., & McGarigal, K. (2004). Patterns in the species–environment relationship depend on both scale and choice of response variables. *Oikos, 105*, 117–124.

Duan, X., Wang, Z., & Tian, S. (2008). Effect of streambed substrate on macroinvertebrate biodiversity. *Frontiers of Environmental Science & Engineering in China, 2*, 122–128.

Fernández, H. R., & Domínguez, E. (2001). *Guia para la determinación de los artrópodos bentônicos sudamericanos*. San Miguel de Tucumán: Universidad Nacional de Tucumán.

Frissell, C. A., Liss, W. J., Warren, C. E., & Hurley, M. D. (1986). A hierarchical framework for stream habitat classification: viewing streams in a watershed context. *Environmental Management, 10*, 199–214.

Gerth, W. J., & Herlihy, A. T. (2006). Effect of sampling different habitat types in regional macroinvertebrate bioassessment surveys. *Journal of the North American Benthological Society, 25*, 501–512.

Graça, M. A. S., Pinto, P., Cortes, R., Coimbra, N., Oliveira, S., Morais, M., et al. (2004). Factors affecting macroinvertebrate

richness and diversity in Portuguese streams: a two-scale analysis. *International Review of Hydrobiology, 89*, 151–164.

Grönroos, M., Heino, J., Siqueira, T., Landeiro, V. L., Kotanen, J., & Bini, L. M. (2013). Metacommunity structuring in stream networks: roles of dispersal mode, distance type and regional environmental context. *Ecology and Evolution, 3*, 4473–4487.

Heino, J. (2005). Functional biodiversity of macroinvertebrate assemblages along major ecological gradients of boreal headwater streams. *Freshwater Biology, 50*, 1578–1587.

Heino, J. (2011). A macroecological perspective of diversity patterns in the freshwater realm. *Freshwater Biology, 56*, 1703–1722.

Hepp, L. U., Landeiro, V. L., & Melo, A. S. (2012). Experimental assessment of the effects of environmental factors and longitudinal position on alpha and beta diversities of aquatic insects in a neotropical stream. *International Review of Hydrobiology, 97*, 157–167.

Hering, D., Moog, O., Sandin, L., & Verdonschot, P. F. M. (2004). Overview and application of the AQEM assessment system. *Hydrobiologia, 516*, 1–20.

Hughes, R. M., & Peck, D. V. (2008). Acquiring data for large aquatic resource surveys: the art of compromise among science, logistics, and reality. *Journal of the North American Benthological Society, 27*, 837–859.

Hughes, R. M., Herlihy, A. T., & Kaufmann, P. R. (2010). An evaluation of qualitative indexes of physical habitat applied to agricultural streams in ten U.S. States. *Journal of the American Water Resources Association, 46*, 792–806.

Jähnig, S., Brunzel, S., Lorenz, A., & Hering, D. (2009). Restoration effort, habitat mosaics, and macroinvertebrates —does channel form determine community response? *Aquatic Conservation: Marine and Freshwater Systems, 19*, 157–169.

Jiang, X. M., Xiong, J., Qiu, J. W., Wu, J. M., Wang, J. W., & Xie, Z. C. (2010). Structure of macroinvertebrate communities in relation to environmental variables in a subtropical Asian river system. *International Review of Hydrobiology, 95*, 42–57.

Johnson, R. K., & Goedkoop, W. (2002). Littoral macroinvertebrate communities: spatial scale and ecological relationships. *Freshwater Biology, 47*, 1840–1854.

Johnson, R. K., Goedkoop, W., & Sandin, L. (2004). Spatial scale and ecological relationships between the macroinvertebrate communities of stony habitats of streams and lakes. *Freshwater Biology, 49*, 1179–1194.

Jurasinski, G., Retzer, V., & Beierkuhnlein, C. (2009). Inventory, differentiation, and proportional diversity: a consistent terminology for quantifying species diversity. *Oecologia, 159*, 15–26.

Kemp, J. L., Harper, D. M., & Crosa, G. A. (2000). The habitat-scale ecohydraulics of rivers. *Ecological Engineering, 16*, 17–29.

Kovalenko, K. E., Thomaz, S. M., & Warfe, D. M. (2012). Habitat complexity: approaches and future directions. *Hydrobiologia, 685*, 1–17.

Kubíková, L., Simon, O. P., Tichá, K., Douda, K., Maciak, M., & Bílý, M. (2012). The influence of mesoscale habitat conditions on the macroinvertebrate composition of springs in a geologically homogeneous area. *Freshwater Science, 31*, 669–679.

Landeiro, V. L., Bini, L. M., Melo, A. S., Pes, A. M. O., & Magnusson, W. E. (2012). The roles of dispersal limitation and environmental conditions in controlling caddisfly (Trichoptera) assemblages. *Freshwater Biology, 57*, 1554–1564.

Legendre, P., & Gallagher, E. D. (2001). Ecologically meaningful transformations for ordination of species data. *Oecologia, 129*, 271–280.

Legendre, P., Borcard, D., & Peres-Neto, P. R. (2005). Analyzing beta diversity: partitioning the spatial variation of community composition data. *Ecological Monographs, 75*, 435–450.

Leibold, M. A., Holyoak, M., Mouquet, N., Amarasekare, P., Chase, J. M., Hoopes, M. F., et al. (2004). The metacommunity concept: a framework for multi-scale community ecology. *Ecology Letters, 7*, 601–613.

Ligeiro, R., Melo, A. S., & Callisto, M. (2010). Spacial scale and the diversity of macroinvertebrates in a Neotropical catchment. *Freshwater Biology, 55*, 424–435.

Ligeiro, R., Hughes, R. M., Kaufmann, P. R., Macedo, D. R., Firmiano, K. R., Ferreira, W. R., et al. (2013). Defining quantitative stream disturbance gradients and the additive role of habitat variation to explain macroinvertebrate taxa richness. *Ecological Indicators, 25*, 45–57.

Melo, A. S., & Froelich, C. G. (2001). Evaluation of methods for estimating macroinvertibrate species richness using individual stones in tropical streams. *Freshwater Biology, 46*, 711–721.

Merritt, R. W., & Cummins, K. W. (1996). *An introduction to the aquatic insects of North America* (3rd ed.). Dubuque: Kendall/Hunt Publishing Company.

Minshall, G. W. (1984). Aquatic insect-substratum relationships. In V. H. Resh & D. M. Rosenberg (Eds.), *The ecology of aquatic insects* (pp. 358–400). New York: Prager.

Moya, N., Hughes, R. M., Dominguez, E., Gibon, F. M., Goita, E., & Oberdorff, T. (2011). Macroinvertebrate-based multimetric predictive models for measuring the biotic condition of Bolivian streams. *Ecological Indicators, 11*, 840–847.

Mugnai, R., Nessimian, J. L., & Baptista, D. F. (2010). *Manual de identificação de macroinvertebrados aquáticos do Estado do Rio de Janeiro*. Rio de Janeiro: Technical Books.

Myers, N., Mittermeier, R. A., Mittermeier, C. G., da Fonseca, G. A. B., & Kent, J. (2000). Biodiversity hotspots for conservation priorities. *Nature, 403*, 853–858.

Mykrä, H., Heino, J., & Muotka, T. (2007). Scale-related patterns in the spatial and environmental components of stream macroinvertebrate assemblage variation. *Global Ecology and Biogeography, 16*, 149–159.

Newson, M., & Newson, C. (2000). Geomorphology, ecology and river channel habitat: mesoscale approaches to basin-scale challenges. *Progress in Physical Geography, 24*, 195–217.

Oksanen, J., Blanchet, F. G., Kindt, R., Legendre, P., Minchin, P. R., O'Hara, R. B., Simpson, G. L., Solymos, P., Henry, M., Stevens, H., Wagner, H. (2013). Vegan: Community Ecology Package. R package version 2.0-8. http://CRAN.Rproject.org/package=vegan.

Pardo, I., & Armitage, P. D. (1997). Species assemblages as descriptors of mesohabitats. *Hydrobiologia, 344*, 111–128.

Pastuchová, Z., Lehotský, M., & Grešková, A. (2008). Influence of morphohydraulic habitat structure on invertebrate communities (Ephemeroptera, Plecoptera and Trichoptera). *Biologia, 63*, 720–729.

Peck, D. V., Herlihy, A. T., Hill, B. H., Hughes, R. M., Kaufmann, P. R., Klemm, D. J., et al. (2006). *Environmental monitoring and assessment program-surface waters: Western Pilot Study field operations manual for wadeable streams*. Washington, DC: EPA/620/R-06/003. USEPA.

Peres-Neto, P. R., & Legendre, P. (2010). Estimating and controlling for spatial structure in the study of ecological communities. *Global Ecology and Biogeography, 19*, 174–184.

Peres-Neto, P. R., Legendre, P., Dray, S., & Borcard, D. (2006). Variation partitioning of species data matrices: estimation and comparison of fractions. *Ecology, 87*, 2614–2625.

Pérez, G. R. (1988). *Guía para es estudio de los macroinvertebrados acuáticos del Departamento de Antioquia*. Fondo Fen: Colombia/Colciencias, Universidad de Antioquia, Colombia.

Principe, R. E., Raffaini, G. B., Gualdoni, C. M., Oberto, A. M., & Corigliano, M. C. (2007). Do hydraulic units define macroinvertebrate assemblages in mountain streams of central Argentina? *Limnologica – Ecology and Management of Inland Waters, 37*, 323–336.

R Development Core Team (2013). R: a language and environment for statistical computing (3.0.1). R Foundation for Statistical Computing, Vienna, Austria. http://www.R-project.org.

Reid, M. A., & Thoms, M. C. (2008). Surface flow types, near-bed hydraulics and the distribution of stream macroinvertebrates. *Biogeosciences, 5*, 1043–1055.

Rempel, L. L., Richardson, J. S., & Healey, M. C. (2000). Macroinvertebrate community structure along gradients of hydraulic and sedimentary conditions in a large gravel-bed river. *Freshwater Biology, 45*, 57–73.

Rios, S. L., & Bailey, R. C. (2006). Relationship between riparian vegetation and stream benthic communities at three spatial scales. *Hydrobiologia, 55*, 153–160.

Roque, F. O., Siqueira, T., Bini, L. M., Ribeiro, M. C., Tambosi, L. R., Ciocheti, G., et al. (2010). Untangling associations between chironomid taxa in Neotropical streams using local and landscape filters. *Freshwater Biology, 55*, 847–865.

Sály, P., Takács, P., Kiss, I., Bíró, P., & Erős, T. (2011). The relative influence of spatial context and catchment- and site-scale environmental factors on stream fish assemblages in a human-modified landscape. *Ecology of Freshwater Fish, 20*, 251–262.

Schneck, F., Schwarzbold, A., & Melo, A. S. (2011). Substrate roughness affects stream benthic algal diversity, assemblage composition, and nestedness. *Journal of the North American Benthological Society, 30*, 1049–1056.

Siqueira, T., Bini, L. M., Roque, F. O., Couceiro, S. R. M., Trivinho-Strixino, S., & Cottenie, K. (2012). Common and rare species respond to similar niche processes in macroinvertebrate metacommunities. *Ecography, 35*, 183–192.

Stendera, S. E. S., & Johnson, R. K. (2005). Additive partitioning of aquatic invertebrate species diversity across multiple spatial scales. *Freshwater Biology, 50*, 1360–1375.

Thompson, R., & Townsend, C. (2006). A truce with neutral theory: local deterministic factors, species traits and dispersal limitation together determine patterns of diversity in stream invertebrates. *Journal of Animal Ecology, 75*, 476–484.

Tickner, D., Armitage, P., Bickerton, M. A., & Hall, K. A. (2000). Assessing stream quality using information on mesohabitat distribution and character. *Aquatic Conservation: Marine & Freshwater Ecosystem, 10*, 179–196.

Tuomisto, H., & Ruokolainen, K. (2006). Analyzing or explaining beta diversity? Understanding the targets of different methods of analysis. *Ecology, 87*, 2697–2708.

Verdonschot, P. F. (2001). Hydrology and substrates: determinants of oligochaete distribution in lowland streams (The Netherlands). *Hydrobiologia, 158*, 249–262.

Vinson, M. R., & Hawkins, C. P. (1998). Biodiversity of stream insects: variation at local, basin and regional scales. *Annual Review of Entomology, 43*, 271–293.

Wang, L., Weigel, B. W., Kanehl, P., & Lohman, K. (2006). Influence of riffle and snag habitat specific sampling on stream macroinvertebrate assemblage measures in bioassessment. *Environmental Monitoring and Assessment, 119*, 254–273.

Wantzen, K. M. (2003). Cerrado streams— characteristics of a threatened freshwater ecosystem type on the tertiary shields of South America. *Amazoniana, 17*, 485–502.

Whittaker, R. H. (1960). Vegetation of the Siskiyou Mountains, Oregon and California. *Ecological Monographs, 26*, 1–80.

Wiens, J. A. (2002). Riverine landscapes: taking landscape ecology into the water. *Freshwater Biology, 47*, 501–515.