This manuscript is contextually identical with the following published paper:

Czeglédi I., Sály P., Takács P., Dolezsai A., Nagy S.A., Erős T. (2016) The scales of variability of stream fish assemblages at tributary confluences. - AQUATIC SCIENCES 78:(4) pp.641-654. DOI: 10.1007/s00027-015-0454-z

The original published pdf available in this website:

http://link.springer.com/article/10.1007%2Fs00027-015-0454-z

1 **The scales of variability of stream fish assemblages**

2 **at tributary confluences**

3

4

5 Czeglédi I.[1], Sály P.[2], Takács P.[2], Dolezsai A.[2], Nagy S.A.[1], Erős T.[2]

6 [1]University of Debrecen, Department of Hydrobiology

7 Egyetem tér 1., H-4032 Debrecen, Hungary

8 [2]Balaton Limnological Institute, MTA Centre for Ecological Research

9 Klebelsberg K. u. 3., H-8237 Tihany, Hungary

10

11

12 *Corresponding author:

13 Tibor ERŐS

14 Balaton Limnological Institute, MTA Centre for Ecological Research

15 Klebelsberg K. u. 3., H-8237 Tihany, Hungary

16 Tel.: +36 87 448 244

17 Fax.: +36 87 448 006

18 E-mail address: eros.tibor@okologia.mta.hu

19 **Running head:** organization of stream fishes at tributary confluences

20

Abstract

Tributary confluences play an important role in the dispersal of organisms, and consequently, in shaping regional scale diversity in stream networks. Despite their importance in dispersal processes, little is known about how ecological assemblages are organized in these habitats. We studied the scales of variability of stream fish assemblages over three seasons using a hierarchical sampling design, which incorporated three tributaries, three sites at the mouth of each tributary and using four sampling units at each site. We found strong scale dependent variability in species richness, composition and relative abundance. Most of the variation was accounted for by the interactive effect of season, between stream and between site effects, while habitat structure of the sampling units had a relatively minor role. Species richness showed a continuous decrease from the mainstem river in most cases, while species composition and relative abundance changed less consistently along the longitudinal profile. Consequently, we found that not only the junctions presented a strong filter on the species pool, but some species were filtered out if they passed this critical habitat bottleneck. Overall, our results suggest high variability in fish assemblages across multiple scales at tributary confluences. Environmental management should take a more critical care on the filtering role of tributary confluences in species dispersal, for better understanding patterns and processes in the branches of dendritic stream networks.

Keywords: stream networks; tributaries; ecotones; habitat bottlenecks; environmental filtering; dispersal

44   Introduction

45   A central task of community ecology is to disentangle how environmental filtering processes
46   and biotic interactions select the assembly of local communities from the regional species
47   pool (Vellend 2010). The act of environmental filtering tends to be the most prominent at the
48   interface of those habitat patches which largely differ in their environmental conditions
49   (ecotones). Characterizing how species disperse through these habitat bottlenecks and
50   distribute themselves among patches is important, among others, for a more regional scale
51   understanding of the organization of ecological communities (Risser 1995).

52   In dendritic stream networks tributary confluences (or junctions) provide one of the most
53   characteristic examples of within stream ecotones (Ward and Wiens 2001). Here, abrupt
54   changes often happen in width, depth, sediment characteristics and water quality along the
55   longitudinal profile of the stream (Rice et al. 2008). Tributary confluences can also increase
56   physical heterogeneity (e.g. in water and sediment characteristics) in the recipient river which
57   have important consequences in channel and floodplain morphology (Benda et al. 2004; Rice
58   et al. 2006). Confluences thus serve as key habitat structures, which may influence dispersal
59   processes for stream organisms in both directions (i.e. from the tributary to the main channel
60   and *vica versa*). However, despite their importance relative to their small spatial extent,
61   ecological studies in stream networks concentrated mainly on understanding patterns and
62   processes in the branches and not in the junctions (sensu Grant et al. 2007).

63   Studies which would specifically focus on the role of confluences considered mainly changes
64   in the composition of the main river assemblages below tributary mouth sections (see e.g.
65   Rice et al. 2001; Kiffney et al. 2006; Milesi and Melo 2014). Studies, which would have
66   specifically considered the upstream filtering role of tributary mouths are especially rare (but
67   see e.g. Grenouillet et al. 2004; Hitt and Angermeier 2008), and in most cases used only
68   indirect evidence for the permeability vs. resistance function of the tributary-mainstem
69   ecotone. For stream macroinvertebrates Beckmann et al. (2005) highlighted that not only
70   tributaries influence the composition and assemblage structure of the main river, but that
71   mainstem assemblages can intensively use the tributary mouth for habitat. For stream fish
72   assemblages Thornbrugh and Gido (2010) showed an abrupt change in assemblage
73   composition between mainstem river sites and tributary sample sites above confluences,
74   followed by a gradual taxonomic change with increasing upstream distance. Overall, these
75   studies suggest that confluences play a strong filter on the species pool of mainstem rivers and
76   has an overarching role on assemblage organization of tributary streams.

77   Stream systems show extreme heterogeneity at a hierarchy of spatial scales from microhabitat
78   to the catchment (Hildrew and Giller 1994; Poff 1997). Determining the scales at which
79   stream organisms varies the most is essential for understanding the predictability of
80   assemblages and providing implications for management (Heino et al. 2004, Erős and
81   Schmera 2010; Ligeiro et al. 2010). Variability of stream assemblages can be especially high
82   at tributary confluences due to the importance of these habitats (i.e. crossroad function) in the
83   dispersal of organism in the dendritic network. Quantifying the importance of scale related
84   variability at tributary confluences is thus important for understanding the variability of

85   assemblages upstream in the branches. For example, if one finds large between stream
86   variability already at the tributary mouth section, this could (partly) explain the differences in
87   the composition of assemblages among the branches, at least for those stream organisms,
88   which dispersal is restricted exclusively to within stream movement. However, the relative
89   role of different scales (e.g. between vs within stream effects, mesohabitat level heterogeneity
90   within sites) is largely unknown on the assembly of stream organisms at tributary
91   confluences.

92   In this study, we examine the filtering role of confluences on stream fish assemblages. Our
93   goal was to determine the major scales of assemblage variation at tributary mouth sections.
94   Specifically, we used a hierarchical (i.e. nested) sampling design to assess the role of season,
95   stream identity (i.e. between stream differences), spatial position at the tributary mouth
96   section (between reach differences), and habitat structure (between sampling unit or
97   mesohabitat level differences) on the structure of fish assemblages using survey data from
98   three tributary mouth sections of a lowland medium size river. Such studies have importance
99   for a more complete understanding of the role of the junctions in the organization of
100 assemblages in stream networks.

101

102 Materials and Methods

103 Study area

104 Our study streams were the 33.3 km long Hajagos (catchment area: 236.9 km^2), the 57.1 km
105 long Gerence (310.3 km^2) and the 45.4 km long Sokoroi (349 km^2) streams. These streams are
106 the right side tributaries of the Marcal River, a 100.5 km long lowland river, which is situated
107 in North-Western Hungary. At the middle and downstream part of the Marcal, the average
108 wet with is 10–15 m and the mean annual discharge is 6.8 m^3 s^{-1}. The tributary mouths of the
109 three studied streams are on average 20.3 km from each other along the Marcal. In their
110 middle and downstream sections, the streams run on an agricultural landscape. They have of
111 lowland character, with low flow, fine substrate (predominantly sand, silty sand, and fine and
112 coarse gravel) and with no clear riffle-pool structure. At the most downstream (i.e. tributary
113 mouth) section the mean channel width, velocity and depth varied between 2.89 and 5.19 m,
114 6.1 and 13.4 cm s^{-1}, 43.5 and 88.1 cm, respectively (see Table 1 for details). Emergent (e.g.
115 reed *Phragmites australis,* broadleaf and narrowleaf cattail *Typha latifolia* and *Typha*
116 *angustifolia,* sweet-grass *Glyceria sp.*) and floating (yellow water-lily *Nuphar lutea,*
117 duckweeds *Lemna sp.*) macrophytes can be abundant at some sites.

118 Fish and habitat sampling

119 We chose three sampling sites per stream. Their downstream end point situated at 0 m, 500 m
120 and 1000 m from the tributary mouth (Fig. 1). Each site was 200 m long and were further
121 divided into four 50 m long sampling units, to examine the effect of habitat structure on fish
122 assemblage characteristics at a finer scale. We sampled fishes and characterized the habitat
123 during three sampling periods in 2013: (1) spring (end of April, early May), (2) summer (early

124 July), and (3) autumn (early October). This sampling effort yielded a total of 108 samples (3
125 seasons × 3 streams × 3 sites per stream × 4 habitat units per site).

126 We surveyed the 200 m long sites by wading, single pass electrofishing using a backpack
127 electrofishing gear (IG200/2B, PDC, 50-100 Hz, 350-650 V, max. 10 kW; Hans Grassl
128 GmbH, Germany). The 50 m long sampling units were block netted before electrofishing,
129 with nets of 2 mm mesh size anchored with chain to the bottom. Pulsating direct current with
130 a frequency of 75–100 Hz and a voltage of 200–300 V was used. The 2 m long catcher rod
131 had a ring shaped anode with a diameter of 40 cm equipped with a net (mesh size 6 mm). The
132 sampling crew consisted of 3–4 people: the electrofisher operator who effectively caught the
133 fish and handled the machine, a netter who helped to catch escaping or unseen fish and one or
134 two helpers who carried the buckets on the bank, took care of the fish and measured their
135 standard length. The team electrofished the whole stream width while moving slowly
136 upstream and trying to catch all fish in the study area. All fish were placed in large containers
137 filled with water and released back to the stream after identification and length measurement.

138 Transect based measurements of habitat data were taken at each 50 m long unit after fish
139 sampling. At each unit, four transects were placed perpendicular to the main axis of the
140 stream at equal distances from each other. We measured wet width of the channel with a tape
141 measure, depth and current velocity with a meter stick and a water velocity meter (FP101
142 Global Flow Probe, Global Water Instrumentation Inc., Gold River, CA, USA), respectively,
143 at five equally spaced points along each transect. We also recorded aquatic vegetation type
144 and substrate type (see Table 1) at every transect point. We calculated the mean values of the
145 aforementioned environmental variables and the percentage occurrence of the categorical
146 variables (i.e., substrate type and aquatic vegetation type) to characterise habitat features at
147 the sample unit scale.

148 We also used fish monitoring data from the Marcal River for comparing similarities and
149 differences in fish assemblages between the mainstem Marcal and the tributary fish
150 assemblages. The river was sampled from a boat using the same electrofishing device and its
151 accessories as was used for the tributary streams. To allow effective manuovering in this
152 shallow and vegetated river, a small rubber boat (Yamaha 300S) was used. The crew
153 comprised two persons: one for catching the fish with the hand-held anode (2.5 m long pole
154 with a net of 40 cm diameter, mesh size 6 mm) and one for driving the boat. Two 400 m long
155 reaches (altogether 800 m long sections) were electrofished in the vicinity (i.e. within 5 km)
156 of each tributary at our standard monitoring sites in this river. Continuous electrofishing was
157 carried out, by dipping the anode into the water at approximately 2 m long intervals and
158 pulling the anode toward the boat, while moving slowly downstream with the flow and
159 controlling the boat by an oar if it was necessary. As for the tributary streams fish were
160 released back to the river after identification and length measurement.

161

162

163 Statistical analysis

164 We used linear models in a nested design to assess the effect of (1) seasons, (2) tributary
165 streams within seasons, (3) sites within tributary streams and seasons, and (4) habitat data
166 nested within seasons and tributary streams on (1) species richness and (2) ecological
167 dissimilarity of the assemblages compared to the main river. For dissimilarity index the
168 Jaccard and the Bray-Curtis indices were used for species composition and relative abundance
169 data (see Legendre and Legendre 1998), respectively.

170 We used principal component analysis on the variance-covariance matrix of the recorded
171 habitat data (PCA) to characterize the physical structure of the habitat units, and used the first
172 three component scores (hereafter habitat PC scores) to quantify habitat data in the nested
173 model. The advantage of this procedure is that it reduces the number of the original habitat
174 variables which usually are correlated with each other. The correlation between the habitat
175 variables could be a nuisance factor in linear models, which phenomenon is known as
176 multicollinearity (Graham 2003; Dormann et al. 2013). Here, the first three principal
177 components of the PCA explained 24.4%, x% and Y% of the variance, which justify their
178 inclusion in the models. In PC1, habitat units with the strongest negative loadings were
179 relatively deep and wide with relatively high velocity, silty-sand substrate and relatively dense
180 submerged macrophyte vegetation. Units with strong positive loadings were relatively
181 shallow with low velocity and relatively dense emergent vegetation (Table 5). In PC2 the
182 habitat gradient was mainly characterized by differences in substrate composition, and
183 especially sandy silt (negative loadings) and fine gravel (positive loadings). Although
184 biologically less interpretable, abiotic differences between habitat units along PC3 were
185 further refined by substrate composition and differences in vegetation structure (Table 6).

186 For a more complete understanding of confluence effects, we examined changes in species
187 richness, and dissimilarity in species composition (Jaccard index) and relative abundance
188 (Bray-Curtis index) from the mainstem Marcal River for each stream and in each season.
189 Spearman correlation tests were used to test whether the values of these assemblage level
190 variables showed significant decay with rank distance upstream from the mainstem at the
191 level of habitat units (n=12 in each test).

192 Then, focusing exclusively on the tributaries, we used principal coordinate analysis (PCoA) to
193 reveal patterns in species composition and relative abundance between seasons, streams and
194 sites. Here, data from individual habitat units were pooled for the ease of visualization. The
195 Jaccard and the Bray-Curtis indices were used for species composition and relative
196 abundance, respectively. Finally, we used nested permutational analysis of variance
197 (PERMANOVA) to identify the scales at which species composition and relative abundance
198 of the tributary fish assemblages varied the most. As with the PCoA, the Jaccard and the
199 Bray-Curtis indices were used for species composition and relative abundance, respectively.

200 All data analyses were performed in the R statistical environment (R Core Team 2013).
201 PERMANOVA was conducted with 'adonis' function of the R package named vegan
202 (Oksanen et al. 2013).

203

Results

205 Most species that occurred in the mainstem river appeared also in the tributaries (Table 2). In
206 fact, only large bodied and/or strongly riverine species did not appear in the tributaries such as
207 asp (*Aspius aspius*), barbel (*Barbus barbus*), European catfish (*Silurus glanis*), zingel (*Zingel*
208 *zingel*). However, these species were rather rare in the mainstem river as well.

209 Species richness varied significantly at a variety of sampling scales in the tributaries (Table
210 3). Most of the variation was related to stream (36.4%; P<0.001) and site level variability
211 (19.3%; P<0.001), although seasonal differences also proved to be significant (7.5%;
212 P<0.001). Species richness distributed relatively evenly amongstreams and sites in spring
213 (Fig. 2), while it tended to decrease upstream in both summer and autumn. Species richness
214 also varied among streams according to their upstream-downstream position in the mainstem
215 in both summer and autumn. The most downstream Sokoroi stream had the highest species
216 richness, while the most upstream Hajagos had the lowest. Interestingly, habitat structure (i.e.
217 PC1-3) did not significantly influence species richness, although the overall contribution of
218 the biologically interpretable most significant environmental gradients (i.e. PC1-3) were in
219 fact higher than the effect of season (14.7%).

220 Jaccard dissimilarity of species composition from the mainstem showed similar influence of
221 sampling scales to species richness (Table 3). Most of the variation was related to stream
222 (39.0%; P<0.001) and site level variability (23.6%; P<0.001), although seasonal differences
223 (3.2%; P<0.05) and the effect of habitat structure (i.e. PC2 6.7%; P=0.026) also proved to be
224 significant. Dissimilarity from the mainstem was more even among streams and sites in spring
225 (Fig. 3), while it tended to increase upstream among sites in the tributaries and among streams
226 along their upstream-downstream position in the mainstem, both summer and autumn. For
227 Bray-Curtis dissimilarity of relative abundance data seasonal changes proved to be the most
228 influential (54.3%; P<0.001). Clearly, dissimilarity from the mainstem increased from spring
229 to autumn (Fig. 3), while the effect of hierarchical spatial levels proved to be low, albeit
230 significant at the stream (6.4%; P=0.003) and site (12.4%; P<0.001) levels.

231 Species richness showed a continuous decrease from the mainstem river in most seasons and
232 streams (Fig. 4; Table 4). However, species composition and relative abundance changed less
233 consistently along the longitudinal profile. Dissimilarity in species composition generally
234 increased from the mainstem in both summer and autumn, with the exception of the Sokoroi
235 stream. On the contrary, dissimilarity in relative abundance showed a variety of patterns. For
236 example, it decreased in the Gerence stream in both spring and summer, while it increased in
237 the Sokoroi stream in autumn.

238 Species composition of the streams and sites within streams was determined by several
239 species (Fig. 5). However, no clear pattern in assemblage composition was found regarding
240 seasonal, between stream or between site differences. PERMANOVA analysis (Table 5)
241 confirmed these findings and showed that relatively equal amount of variation of species
242 composition was related to seasonal (14.6%; P<0.001) and to between stream (22.4%;
243 P<0.001) and between site (13.7%; P<0.001) effects. Habitat structure also significantly
244 influenced species richness (i.e. PC2 6.5%; P=0.018), and the overall contribution of the

biologically interpretable most significant environmental gradients (i.e. PC1-3) was in fact
higher (15.8%) than the effect of season. Relative abundance data were mostly influenced by
three dominant species (i.e. roach *Rutilus rutilus*, bitterling *Rhodeus sericeus*, pike *Esox lucius*; Fig. 5). Here, the greatest amount of variation in fish abundance data was related to the
site scale (20.7%; $P<0.001$), although seasonal (12.6%; $P<0.001$) and stream level (17.8%;
$P<0.001$) effects, and the effect of individual environmental gradients (PC1 7.1%; $P=0.007$;
PC2 6.4%; $P=0.022$; PC3 6.4%; $P=0.021$) also proved to be significant (Table 5).

Discussion

In this study we examined the scales of variation of stream fishes at tributary confluences and
found that the majority of assemblage variation could be explained by the joint effect of
different hierarchical (spatial and temporal) scales. In general, seasonal, between stream and
site level effects proved to be the most important in explaining assemblage structure, whereas
physical attributes of the sample units (50 m long each) had a relatively minor role. These
results on the importance of season, spatial positioning in the stream network (between stream
effects) and positioning within a stream (between site effects) may suggest that dispersal
processes had critical importance for fish assemblage organization at tributary confluences,
and will be discussed below in more detail.

Most of the studies that considered the role of spatial processes (e.g. dispersal limitation,
neutral effects) in stream networks used a metacommunity perspective and examined the
relative role of environmental factors and spatial positioning of the sampling sites at large
spatial extents (i.e. within and among different subcatchments) (e.g. Mykrä et al. 2007; Erős
et al. 2012). The logical consensus of these studies is that the role of spatial effects decreases
at smaller spatial extents (Heino et al. 2012; Muneepeerakul et al. 2008). However, it has been
also shown that spatial processes like dispersal limitation can be very important at smaller
extents too, for example within and between branches of the dendritic network (Fagan 2002;
Erős and Schmera 2010). Our field study shows that between stream effects can be significant
even among streams with relatively similar environmental and topographic characteristic and
spatial positioning along the mainstem river, which, to our knowledge, has not been addressed
in other studies.

Between stream effects were high for species richness and dissimilarity of species
composition from the mainstem river (Table 3). Specifically, we found a reverse pattern
between species richness and dissimilarity in species composition from the mainstem. Species
richness was the highest in the Sokoroi and lowest in the Hajagos stream, while dissimilarity
in species composition from the mainstem was the lowest in the Sokoroi and highest in the
Hajagos, at least in summer and autumn. Lower species richness and higher dissimilarity from
the mainstem in species composition suggest lower colonization potential of the Hajagos from
the Marcal River. Confluences thus served as natural habitat bottlenecks for stream fishes in a
stream specific manner. The results may suggest the role of stochastic effects in the
colonization of tributary mouths by different species from the mainstem and/or suggest the

285 role of tributary spatial position in the mainstem river (see Osborne and Wiley 1992), even if
286 the tributaries were relatively close to each other.

287 Temporal variability also had some legacy on between stream differences in richness and
288 dissimilarity of species composition from the mainstem. These differences were relatively
289 minor in spring and showed consistent pattern in summer and autumn. Higher water levels in
290 spring could diminish between stream differences in habitat availability of the junctions for
291 mainstem fishes, since alluvium bars (i.e. sand and gravel bars) at the mouth may hinder
292 upstream migration at low water periods. Higher movement activity of fish during spring
293 (spawning migrations) may also dampened differences among the tributaries. Dissimilarity
294 from the mainstem in relative abundance data supports this argument on seasonality.
295 Dissimilarity values were the lowest in spring and the highest in autumn. In fact, dissimilarity
296 from the mainstem was most related to variation among seasons for relative abundance data
297 Table 3), reflecting seasonal differences in habitat availability and/or in movement activity for
298 the dominant species (Gorman 1986; Roberts and Hitt 2010).

299 At a lower hierarchical level, all assemblage attributes varied significantly between sites,
300 which further indicates the importance of spatial processes for stream fishes at very small
301 spatial extents (here within a 1 km long section). Species richness generally decreased
302 upstream at least in summer and autumn (Fig. 2), while dissimilarity in species composition
303 increased from the mainstem. To our knowledge no study examined differences in fish
304 assemblages from the mainstem at such a small spatial extent, but these data and studies at
305 larger spatial extents (Hitt and Angermeier 2008; Thornbrugh and Gido 2010) indicate that
306 not only the junctions present a strong filter on the species pool, but that some species are
307 filtered out even if they passed through this most critical habitat bottleneck. Dissimilarity in
308 relative abundance from the mainstem was less related to variation between sites, which,
309 compared with results on composition, indicates the effect of some dominant species on these
310 results. Some habitat generalist species, like roach, bitterling and bleak (*Alburnus alburnus*),
311 which are dominant both in the mainstem river and in the tributaries were abundant at all sites
312 and could increase assemblage similarity at the site level, similarly to their effect at the stream
313 level. Patterns in distance decay from the mainstem (Fig. 4) confirmed site level changes, and
314 indicated the most consistent pattern between seasons and streams for species richness (i.e.
315 decrease in richness upstream) and the least consistent patterns for relative abundance data.

316 Analyses which did not consider similarities and differences from the mainstem, but focused
317 on assemblage variability in the tributaries exclusively (i.e. PCoA and PERMANOVA),
318 showed that relatively equal amount of variation was related to season, and between stream
319 and between site variation for both compositional and relative abundance data. These results
320 suggest strong context dependency in fish assemblage organization at tributary confluences,
321 which may harden generalizations across sites, streams and seasons (see Heino et al., 2012 for
322 a larger scale study). The relatively high amount of variation explained (66.4–71.0%) suggest
323 that the joint consideration of seasonal effects and between and within stream positioning of
324 the sampling sites is critically important for better understanding assemblage organization,
325 beside the consideration of habitat structure.

For the lowest hierarchical level (i.e. sampling unit scale), we found that habitat structure had low importance relative to seasonal, between stream, and between site variation. Habitat structure is clearly the most important factor (group) for the organization of stream fishes (Matthews 1998; Jackson et al. 2001). It seems however that larger scale factors (e.g. permeability of the junctions, dispersal ability of species) can also significantly influence the species pool of lower hierarchical levels (i.e. species composition and relative abundance of fishes at the meso-habitat level) and in case of relatively homogenous habitat units, they can override the effect of habitat structure (see Grönroos and Heino 2012 for a study on stream macroinvertabrates). In this lowland system, the units had relatively similar physical characteristics regarding width, flow, depth and substrate, although the type and coverage of macrovegetation provided some heterogeneity, which could influence the distribution of fish to some extent.

In conclusion, we found strong scale dependent variability in species richness, composition and relative abundance of stream fish assemblages at tributary confluences. Most of the variation was accounted for by the interactive effect of season, between stream and between site effects, while habitat structure of the sampling units (i.e. mesohabitat level structure) had a lower role. The results suggest that dispersal processes (e.g. permeability, dispersal limitation and movement activity of fishes) have critical importance on the assembly of stream fishes at very small spatial extents, which may strongly determine fish assemblages more upstream in the branches of the dendritic network. From an applied perspective revitalization projects often focus on enhancing instream habitat quality for the biota of streams (Lepori et al. 2005; Palmer et al. 2010). We suggest that environmental management of stream networks should take a more critical care on the filtering role of tributary confluences in species dispersal, for better understanding patterns and processes in the branches of dendritic stream networks.

Literature

Beckmann MC, Schöll F, Matthaei CD (2005) Effects of increased flow in the main stem of the River Rhine on the invertebrate communities of its tributaries. Freshwater Biol 50:10–26

Benda L, Poff NL, Miller D, Dunne T, Reeves G, Pess G, Pollock M (2004) The network dynamics hypothesis: How channel networks structure riverine habitats. Bioscience 54:413–427

Dormann CF, Elith J, Bacher S, Buchmann C, Carl G, Carré G, García Marquéz JR, Gruber B, Lafourcade B, Leitão PJ, Münkemüller T, McClean C, Osborne PE, Reineking B, Schröder B, Skidmore AK, Zurell D, Lautenbach S (2013) Collinearity: a review of methods to deal with it and a simulation study evaluating their performance. Ecography 36(1):27–46

Erős T, Schmera D (2010) Spatio-temporal scaling of biodiversity and the species-time relationship in a stream fish assemblage. Freshwater Biol 55:2391–2400

Erős T, Olden JD, Schick RS, Schmera D, Fortin M (2012) Characterizing connectivity relationships in freshwaters using patch-based graphs. Landscape Ecol 27:303–317

Fagan WF (2002) Connectivity, fragmentation, and extinction risk in dendritic metapopulations. Ecology 83:3243–3249

Gorman OT (1986) Assemblage organization of stream fishes: the effect of rivers on adventitious streams. Am Nat 128:611–616

Graham MH (2003) Confronting multicollinearity in ecological multiple regression. Ecology 84:2809–2815

Grant EHC, Lowe WH, Fagan WF (2007) Living in the branches: population dynamics and ecological processes in dendritic networks. Ecol Lett 10:165–175

Grenouillet G, Pont D, Seip KL (2004) Within-basin fish assemblage structure: the relative influence of habitat versus stream spatial position on local species richness. Can J Fish Aquat Sci 61:93–102

Grönroos M, Heino J (2012) Species richness at the guild level: effects of species pool and local environmental conditions on stream macroinvertebrate communities. J Anim Ecol 81:679-691

Heino J, Louhi P, Muotka T (2004) Identifying the scales of variability in stream macroinvertebrate abundance, functional composition and assemblage structure. Freshwater Biol 49:1230–1239

Heino J, Grönroos M, Soininen J, Virtanen R, Muotka T (2012) Context dependency and metacommunity structuring in boreal headwater streams. Oikos 121:537–544

Hildrew AG, Giller PS (1994) Patchiness, species interactions and disturbance in the stream benthos. In Giller, A.G., A. G. Hildrew & D. G. Rafaelli (eds) Aquatic Ecology: scale, pattern and process, 34th Symposium of the British Ecological Society, pp. 21-62. Blackwell, Oxford.

Hitt NP, Angermeier PL (2008) Evidence for fish dispersal from spatial analysis of network topology. J N Am Benthol Soc 27:304–320

Jackson DA, Peres-Neto PR, Olden JD (2001) What controls who is where in freshwater fish communities – the roles of biotic, abiotic and spatial factors. Can J Fish Aquat Sci 58:157–170

Kiffney PM, Greene CM, Hall JE, Davies JR (2006) Tributary streams create spatial discontinuities in habitat, biological productivity, and diversity in mainstem rivers. Can J Fish Aquat Sci 63:2518–2530

410 Legendre P, Legendre L (1998) Numerical ecology. 2nd English Edition. Elsevier Science,
411   Amsterdam: BV. xv+853 p.

412 Lepori F, Palm D, Brannas E, Malmqvist B (2005) Does restoration of structural
413   heterogeneity in streams enhance fish and macroinvertebrate diversity? Ecol Appl
414   15:2060–2071

415 Ligeiro R, Melo AS, Callisto M (2010) Spatial scale and the diversity of macroinvertebrates
416   in a Neotropical catchment. Freshwater Biol 55:424–435

417 Matthews WJ (1998) Patterns in freshwater fish ecology. Chapman & Hall, New York

418 Milesi SV, Melo AS (2014) Conditional effects of aquatic insects of small tributaries on
419   mainstream assemblages: position within the network matter. Can J Fish Aquat Sci 71:1–
420   9

421 Muneepeerakul, R., Bertuzzo, E., Lynch, H. J., Fagan, W. F., Rinaldo, A., and Rodriguez-
422   Iturbe, I. (2008) Neutral metacommunity models predict fish diversity patterns in
423   Mississippi-Missouri basin. Nature 453:220-222

424 Mykrä H, Heino J, Muotka T (2007) Scale related patterns in the spatial and environmental
425   components of stream macroinvertebrate variation. Global Ecol Biogeogr 16:149–159

426 Oksanen J, Blanchet FB, Kindt R, Legendre P, Minchin PR, O'Hara RB, Simpson GL,
427   Solymos P, Stevens MHH, Wagner H (2013) vegan: Community Ecology Package. R
428   package version 2.0-10. \url{http://CRAN.R-project.org/package=vegan}.

429 Osborne LL, Wiley MJ (1992) Influence of tributary spatial position on the structure of
430   warmwater fish communities. Can J Fish Aquat Sci 49:671–681

431 Palmer MA, Menninger HM, Bernhardt E (2010) River restoration, habitat heterogeneity and
432   biodiversity: a failure of theory or practice? Freshwater Biol 55:205–222

433 Poff NL (1997). Landscape filters and species traits: towards mechanistic understanding and
434   prediction in stream ecology. J N Am Benthol Soc 16:391–409

435 R Core Team (2013) R: A language and environment for statistical computing. R Foundation
436   for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0. \url{http://www.R-
437   project.org}.

438 Rice SP, Greenwood MT, Joyce CB (2001) Tributaries, sediment sources, and the
439   longitudinal organisation of macroinvertebrate fauna along river systems. Can J Fish
440   Aquat Sci 58:824–840.

441 Rice SP, Ferguson RI, Hoey TB (2006) Tributary control of physical heterogeneity and
442   biological diversity at river confluences. Can J Fish Aquat Sci 63:2553–2566

443 Rice SP, Kiffney P, Greene C, Pess GR (2008) The ecological importance of tributaries and
444   confluences. In: Rice SP, Roy AG, Rhoads BL River Confluences, Tributaries and the
445   Fluvial Network. John Wiley & Sons, New York, pp 209–242

446 Risser PG (1995) The status of the science examining ecotones. Bioscience 322:318–325

447 Roberts JH, Hitt NP (2010) Longitudinal structure of stream fish communities: evaluating
448   conceptual models with temporal data. Am Fish S S 73:281–299

449 Thornbrugh DJ, Gido KB (2010) Influence of spatial positioning within stream networks on
450   fish assemblage structure in the Kansas River basin, USA. Can J Fish Aquat Sci 67:143–
451   156

452 Vellend M (2010) Conceptual synthesis in community ecology. Q Rev Biol 85(2):183–206

453   Ward JV, Wiens JA (2001) Ecotones of riverine ecosystems: role and typology, spatio-
454   temporal dynamics, and river regulation. Ecohydrol Hydrobiol 1:25–36
455

Table 1. The average habitat characteristics of the 200 m long sample sites in the Hajagos (H), Gerence (G) and Sokoroi (S) streams. Each mean value is based on the pooled mean data of seasonal (i.e. spring, summer, autumn) transect based surveys (see methods for details).

	Hajagos stream			Gerence stream			Sokorói stream		
	H1	H2	H3	G1	G2	G3	S1	S2	S3
Width (m)	4.90 (1.35)	4.00 (0.17)	3.98 (0.38)	5.19 (1.14)	3.56 (0.62)	2.89 (0.79)	4.98 (0.76)	3.90 (0.21)	3.66 (0.47)
Depth (cm)	72.7 (25.9)	50.7 (10.3)	43.5 (7.8)	76.0 (22.4)	44.8 (18.0)	47.3 (19.8)	88.1 (10.1)	71.9 (10.1)	54.6 (17.0)
Velocity (cm s^{-1})	8.1 (4.9)	7.7 (7.7)	10.4 (8.8)	6.1 (4.9	13.4 (12.6)	9.7 (6.2)	6.6 (4.4)	9.8 (4.6)	7.5 (6.1)
Silty sand (%)	49.1 (23.5)	38.7 (23.4)	28.1 (7.0)	46.3 (18.4)	47.6 (28.2)	81.9 (13.4)	81.3 (23.5)	77.1 (28.0)	56.3 (16,4)
Sand (%)	0.4 (1.4)	-	-	-	1.1 (1.7)	3.2 (9.6)	2.1 (4.5)	4.6 (5.4)	8.3 (12.3)
Fine gravel (%)	30.1 (16.7)	16.1 (10.8)	25.2 (13.2)	35.6 (15.5)	25.9 (16.3)	12.4 (11.9)	8.3 (12.3)	18.3 (23.0)	30.8 (21.7)
Coarse gravel (%)	8.9 (10.3)	45.2 (15.8)	46.7 (10.9)	11.4 (17.9)	25.4 (25.2)	2.5 (3.2)	8.3 (12.3)	-	4.6 (7.8)
Stone (%)	9.6 (12.9)	-	-	5.4 (14.4)	-	-	-	-	-
Rock (%)	1.9 (5.8)	-	-	1.3 (3.1)	-	-	-	-	-
Emergent plant (%)	52.3 (40.1)	61.9 (40.8)	67.7 (35.8)	23.7 (21.6)	27.5 (26.5)	53.1 (22.2)	15.6 (9.2)	26.6 (10.9)	69.2 (24.0)
Submerged plant (%)	24.9 (31.0)	22.9 (34.1)	17.1 (26.7)	2.1 (5.8)	15.2 (18.2)	9.8 (14.8)	14.0 (6.8)	16.7 (11.2)	6.5 (7.5)
Floating leaved plant (%)	4.4 (6.0)	8.5 (15.9)	4.3 (8.2)	20.6 (35.6)	0.1 (0.3)	-	17.1 (27.8)	3.1 (6.1)	-
Filamentous algae (%)	-	-	-	-	0.2 (0.4)	-	4.6 (6.9)	13.3 (20.6)	2.2 (3.6)

Table 2. The fish species, species codes and the relative abundance of species collected in the mainstem Marcal River (M) and the Sokoroi (S), Gerence (G) and Hajagos (H) tributary streams.

Species name	Species code	Relab (M) (%)	Relab (S) (%)	Relab (G) (%)	Relab (H) (%)
Abramis brama (Linnaeus, 1758)	abrbra	0.094	0.247	-	0.191
Alburnus alburnus (Linnaeus, 1758)	albalb	40.508	5.890	2.476	4.498
Ameiurus melas (Rafinesque, 1820)	amemel	0.399	0.038	-	0.383
Aspius aspius (Linnaeus, 1758)	aspasp	0.141	-	-	-
Ballerus ballerus (Linnaeus, 1758)	balbal	0.047	1.102	0.667	-
Barbatula barbatula (Linnaeus, 1758)	ortbar	0.023	-	0.334	-
Barbus barbus (Linnaeus, 1758)	barbar	0.141	-	-	-
Blicca bjoerkna (Linnaeus, 1758)	blibjo	1.739	2.451	7.322	12.344
Carassius gibelio (Bloch, 1782)	cargib	0.352	0.418	-	2.967
Chondrostoma nasus (Linnaeus, 1758)	chonas	0.094	-	0.018	-
Cobitis elongatoides (Băcescu & Maier, 1969)	cobelo	0.235	1.045	0.474	0.287
Cyprinus carpio (Linnaeus, 1758)	cypcar	0.023	0.019	-	-
Esox lucius (Linnaeus, 1758)	esoluc	2.867	4.275	1.247	7.464
Gobio obtusirostris (Valenciennes, 1842)	gobgob	-	-	0.246	-
Gymnocephalus cernuus (Linnaeus, 1758)	gymcer	-	-	0.018	-
Lepomis gibbosus (Linnaeus, 1758)	lepgib	0.258	0.038	1.826	0.096
Leuciscus idus (Linnaeus, 1758)	leuidu	1.386	3.040	0.158	1.627
Leuciscus leuciscus (Linnaeus, 1758)	leuleu	0.752	0.817	1.212	0.670
Misgurnus fossilis (Linnaeus, 1758)	misfos	-	0.247	0.228	2.584
Neogobius fluviatilis (Pallas, 1814)	neoflu	0.728	0.171	1.159	0.957
Neogobius melanostomus (Pallas, 1814)	neomel	1.269	0.095	1.054	1.053
Perca fluviatilis (Linnaeus, 1758)	perflu	1.621	3.648	1.018	0.191
Phoxinus phoxinus (Linnaeus, 1758)	phopho	-	-	0.035	-
Proterorhinus semilunaris (Pallas, 1814)	prosem	1.692	2.812	0.544	3.923
Pseudorasbora parva (Temminck & Schlegel, 1842)	psepar	-	0.399	0.105	2.105
Rhodeus sericeus (Pallas, 1776)	rhoser	11.302	46.114	58.472	25.359
Rutilus rutilus (Linnaeus, 1758)	rutrut	31.790	23.580	18.262	28.134
Scardinius erythrophthalmus (Linnaeus, 1758)	scaery	0.705	0.836	0.070	0.957
*Silurus glanis (*Linnaeus, 1758)	silgla	0.211	-	-	-
Squalius cephalus (Linnaeus, 1758)	squcep	1.081	2.641	3.020	4.115
Tinca tinca (Linnaeus, 1758)	tintin	0.023	0.038	-	0.096
Vimba vimba (Linnaeus, 1758)	vimvim	0.470	0.038	0.035	-
Zingel zingel (Linnaeus, 1758)	zinzin	0.047	-	-	-

Table 3. Summary results of the linear models (nested design) for species richness, and dissimilarity of species composition and relative abundance of fishes from the mainstem Marcal River.

Source of variation	d.f.	MS	*F*	*P*	% variance
Species richness					
Season	2	42.19	10.62	<0.001	7.5
Stream (within season)	6	68.30	17.19	<0.001	36.4
Site (within season and stream)	9	24.19	6.09	<0.001	19.3
Habitat PC1 (within season and stream)	9	6.85	1.72	0.102	5.5
Habitat PC2 (within season and stream)	9	7.13	1.79	0.087	5.7
Habitat PC3 (within season and stream)	9	4.33	1.09	0.384	3.5
Residuals	63	3.97			22.2
Dissimilarity in species composition					
Season	2	0.03	5.00	0.010	3.2
Stream (within season)	6	0.12	20.26	<0.001	39.0
Site (within season and stream)	9	0.05	8.19	<0.001	23.6
Habitat PC1 (within season and stream)	9	0.01	1.95	0.061	5.6
Habitat PC2 (within season and stream)	9	0.01	2.31	0.026	6.7
Habitat PC3 (within season and stream)	9	0.00	0.60	0.792	1.7
Residuals	63	0.01			20.2
Dissimilarity in relative abundance					
Season	2	1.36	95.77	<0.001	54.3
Stream (within season)	6	0.05	3.76	0.003	6.4
Site (within season and stream)	9	0.07	4.88	<0.001	12.4
Habitat PC1 (within season and stream)	9	0.02	1.56	0.147	4.0
Habitat PC2 (within season and stream)	9	0.01	0.65	0.750	1.7
Habitat PC3 (within season and stream)	9	0.02	1.30	0.253	3.3
Residuals	63	0.01			17.9

Table 4. Spearman correlation tests between species richness and rank distance from the junction, dissimilarity in species composition (Jaccard index) and rank distance from the junction, and relative abundance (Bray-Curtis index) and rank distance from the junction for each stream and season (n=12 in each test).

Species richness

	Spring		Summer		Autumn	
	R^2	P	R^2	P	R^2	P
Sokoroi	-0.79	0.002	-0.52	0.080	-0.61	0.037
Gerence	-0.69	0.013	-0.79	0.002	-0.76	0.004
Hajagos	-0.12	0.704	-0.54	0.067	-0.89	<0.001

Species composition

	Spring		Summer		Autumn	
	R^2	P	R^2	P	R^2	P
Sokoroi	-0.45	0.138	0.09	0.770	0.34	0.276
Gerence	0.30	0.338	0.80	0.002	0.83	<0.001
Hajagos	0.29	0.367	0.69	0.013	0.80	0.002

Relative abundance

	Spring		Summer		Autumn	
	R^2	P	R^2	P	R^2	P
Sokoroi	-0.60	0.039	-0.37	0.236	0.85	<0.001
Gerence	-0.71	0.009	-0.71	0.009	0.04	0.914
Hajagos	-0.04	0.914	0.25	0.430	0.03	0.931

Table 5. Summary results of the PERMANOVA analyses for species composition and relative abundance data.

Source of variation	d.f.	MS	*F*	*P*	% variance
Dissimilarity in species composition					
Season	2	1.90	13.70	<0.001	14.6
Stream (within season)	6	0.97	6.99	<0.001	22.4
Site (within season and stream)	9	0.39	2.85	<0.001	13.7
Habitat PC1 (within season and stream)	9	0.16	1.16	0.154	5.6
Habitat PC2 (within season and stream)	9	0.19	1.35	0.018	6.5
Habitat PC3 (within season and stream)	9	0.11	0.78	0.938	3.7
Residuals	63	0.14			33.6
Dissimilarity in relative abundance					
Season	2	1.47	13.75	<0.001	12.6
Stream (within season)	6	0.69	6.44	<0.001	17.8
Site (within season and stream)	9	0.54	5.01	<0.001	20.7
Habitat PC1 (within season and stream)	9	0.18	1.71	0.007	7.1
Habitat PC2 (within season and stream)	9	0.17	1.55	0.022	6.4
Habitat PC3 (within season and stream)	9	0.17	1.55	0.021	6.4
Residuals	63	0.11			29.0

Captions to figures

Fig. 1 Scheme of the study design using three tributary streams (Sokoroi, Gerence, Hajagos) and three sites in each stream in the Marcal River system, North Western, Hungary. The sites were situated 0, 500 and 1000 m from the tributary mouth. Numbers indicate individual habitat units (50 m long each)

Fig. 2 Mean species richness (±S.D.) of the sampling units (50 m long each) at three different sites in the Sokoroi, Gerence and Hajagos streams of the Marcal River in spring, summer and autumn

Fig. 3 Mean dissimilarity (±S.D.) of the sampling units (50 m long each) from the mainstem Marcal River for (a) species composition (Jaccard index) and (b) relative abundance (Bray-Curtis index) data of fishes at three different sites in the Sokoroi, Gerence and Hajagos streams in spring, summer and autumn

Fig. 4 Changes in (a) species richness, and dissimilarity in (b) species composition (Jaccard index) and (c) relative abundance (Bray-Curtis index) data from the mainstem Marcal River at the level of the sampling units (50 m long each) in the Sokoroi, Gerence and Hajagos streams in spring, summer and autumn

Fig. 5 Principal Coordinate Analyses (PCoA) summarizing the variation across sites in three tributaries of the Marcal River for (a) species composition and (c) relative abundance data, and their associated species loadings (b and d). For species code abbreviations see Table 2

Appendix

Table 6. Pearson correlation matrix of the abiotic variables and the first three principal component axes (PC1, PC2, PC3). Lower and upper diagonals show correlations and their significance values, respectively.

	PC1	PC2	PC3	Width (m)	Depth (cm)	Velocity (cm s⁻¹)	Silty sand (%)	Sand (%)	Fine gravel (%)	Coarse gravel (%)	Stone and rock (%)	Emergent plant (%)	Submerged plant (%)	Floating leaved plant (%)	Filamentous algae (%)
PC1	-	1.000	1.000	<0.001	<0.001	<0.001	<0.001	0.001	0.151	<0.001	0.054	<0.001	<0.001	0.137	0.424
PC2	<0.001	-	1.000	<0.001	0.002	0.091	<0.001	0.027	<0.001	0.001	<0.001	0.447	0.483	<0.001	0.006
PC3	<0.001	<0.001	-	0.331	<0.001	<0.001	<0.001	<0.001	0.194	<0.001	0.827	0.59	<0.001	0.007	<0.001
Width (m)	-0.603	0.55	0.095	-	<0.001	0.281	0.342	0.003	0.011	0.279	0.002	<0.001	0.038	0.541	0.627
Depth (cm)	-0.794	0.297	0.334	0.732	-	0.085	0.016	0.05	0.789	<0.001	0.002	<0.001	0.001	0.661	0.162
Velocity (cm s-1)	-0.614	-0.164	-0.489	0.105	0.167	-	0.238	0.014	0.646	0.807	0.586	<0.001	<0.001	0.003	0.054
Silty sand (%)	-0.4	-0.76	0.404	-0.092	0.231	0.115	-	0.709	<0.001	<0.001	0.013	0.02	0.509	0.035	0.177
Sand (%)	0.306	-0.213	0.394	-0.282	-0.189	-0.236	-0.036	-	0.284	0.168	0.558	0.317	0.264	0.33	<0.001
Fine gravel (%)	0.139	0.731	0.126	0.243	0.026	-0.045	-0.587	-0.104	-	0.579	0.913	0.548	0.059	0.005	0.007
Coarse gravel (%)	0.375	0.305	-0.73	-0.105	-0.367	-0.024	-0.703	-0.134	-0.054	-	0.725	0.03	0.389	0.362	0.208
Stone and rock (%)	-0.186	0.355	-0.021	0.293	0.297	-0.053	-0.237	-0.057	0.011	-0.034	-	0.966	0.617	0.514	0.44
Emergent plant (%)	0.792	-0.074	-0.052	-0.347	-0.567	-0.501	-0.223	0.097	0.058	0.209	0.004	-	<0.001	0.145	0.209
Submerged plant (%)	-0.67	-0.068	-0.417	0.2	0.316	0.609	0.064	-0.108	-0.182	0.084	0.049	-0.568	-	0.038	0.72
Floating leaved plant (%)	0.144	0.484	0.258	0.06	0.043	-0.288	-0.203	-0.095	0.266	0.089	-0.063	-0.141	-0.2	-	0.072
Filamentous algae (%)	0.078	0.263	0.535	-0.047	0.135	-0.186	-0.131	0.346	0.26	-0.122	-0.075	-0.122	-0.035	0.174	-

Species richness

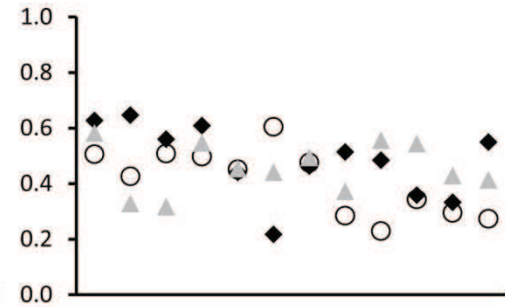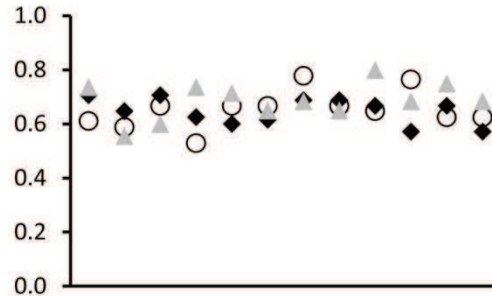(a) **Species composition** (b) **Relative abundance**
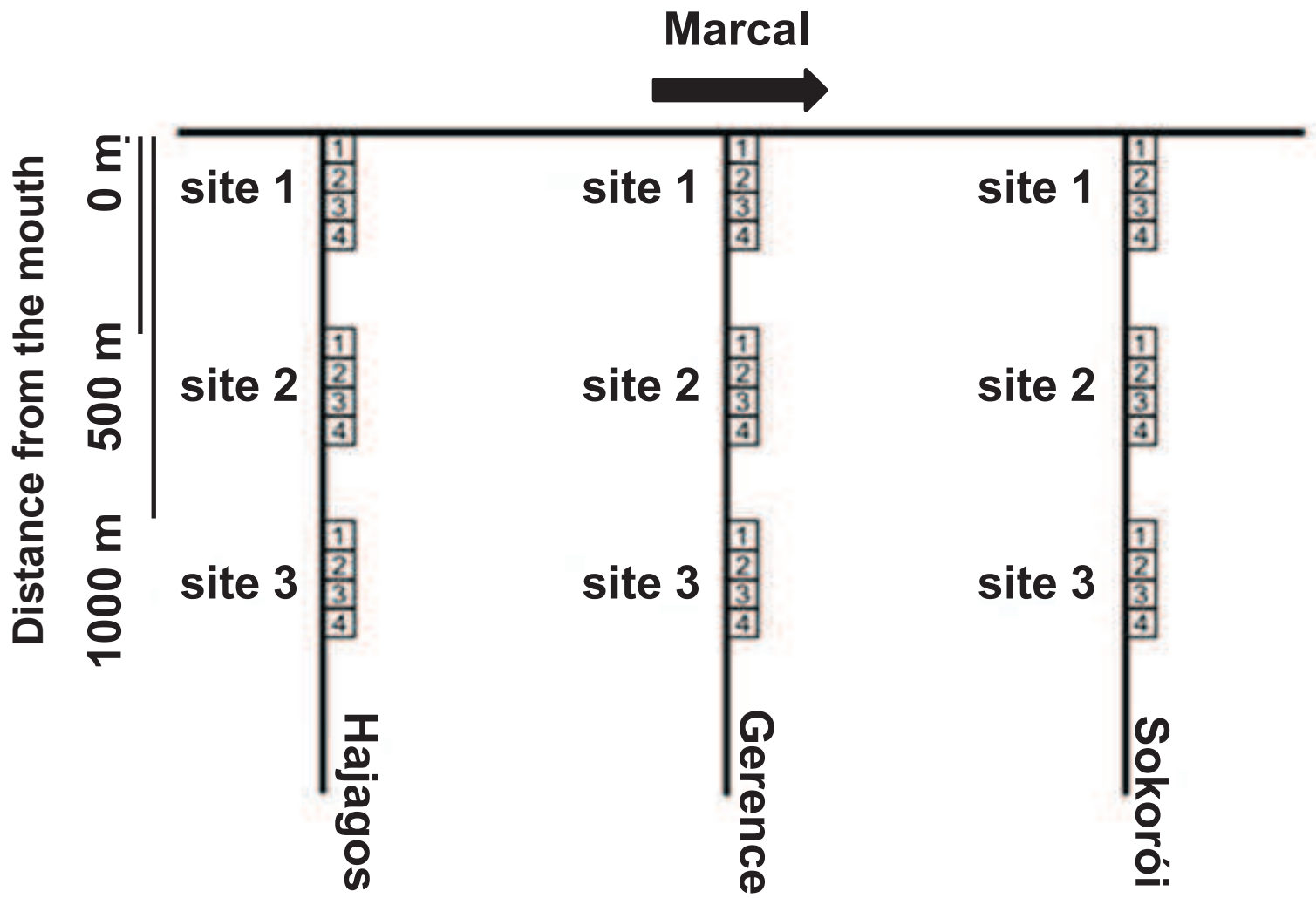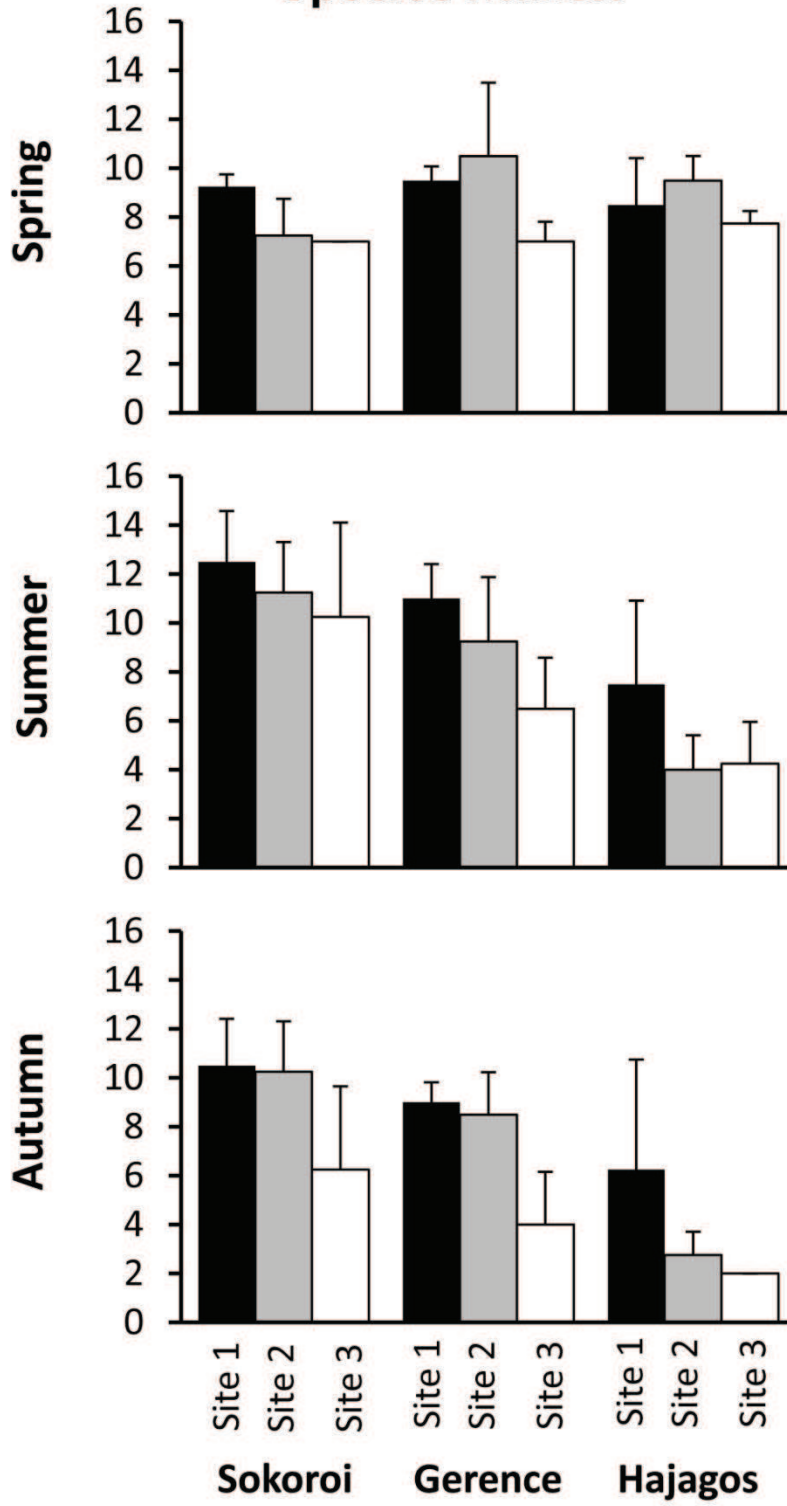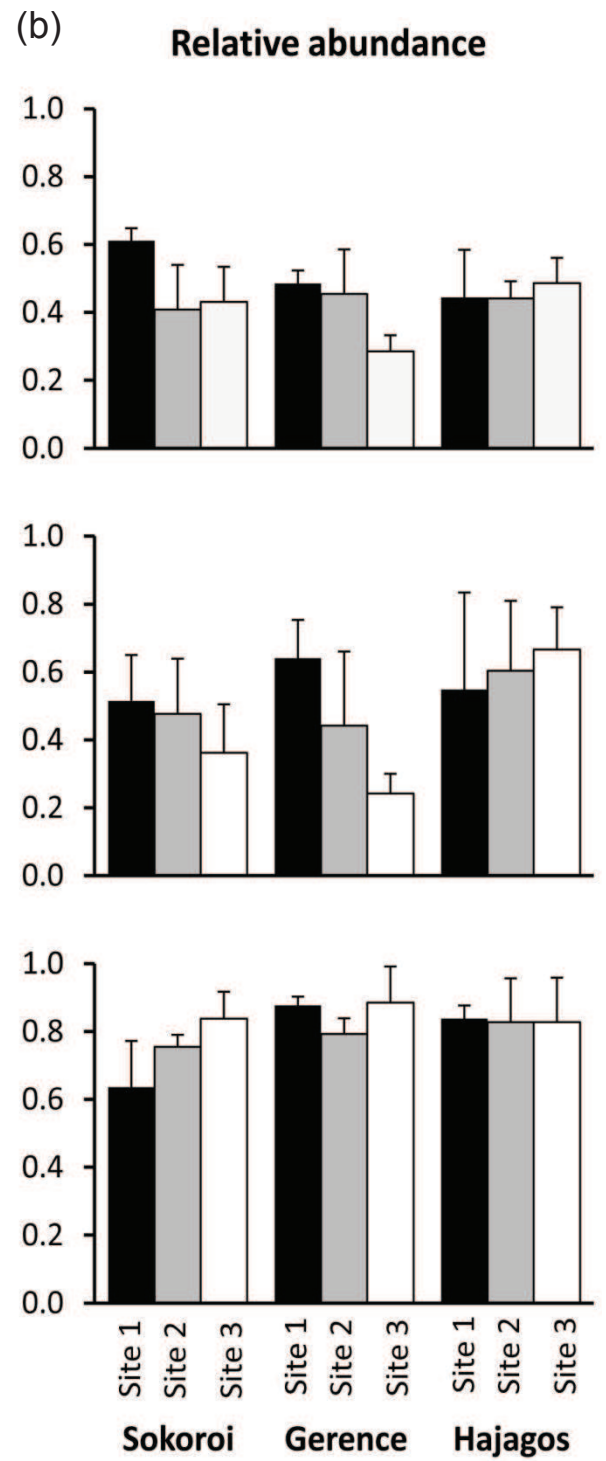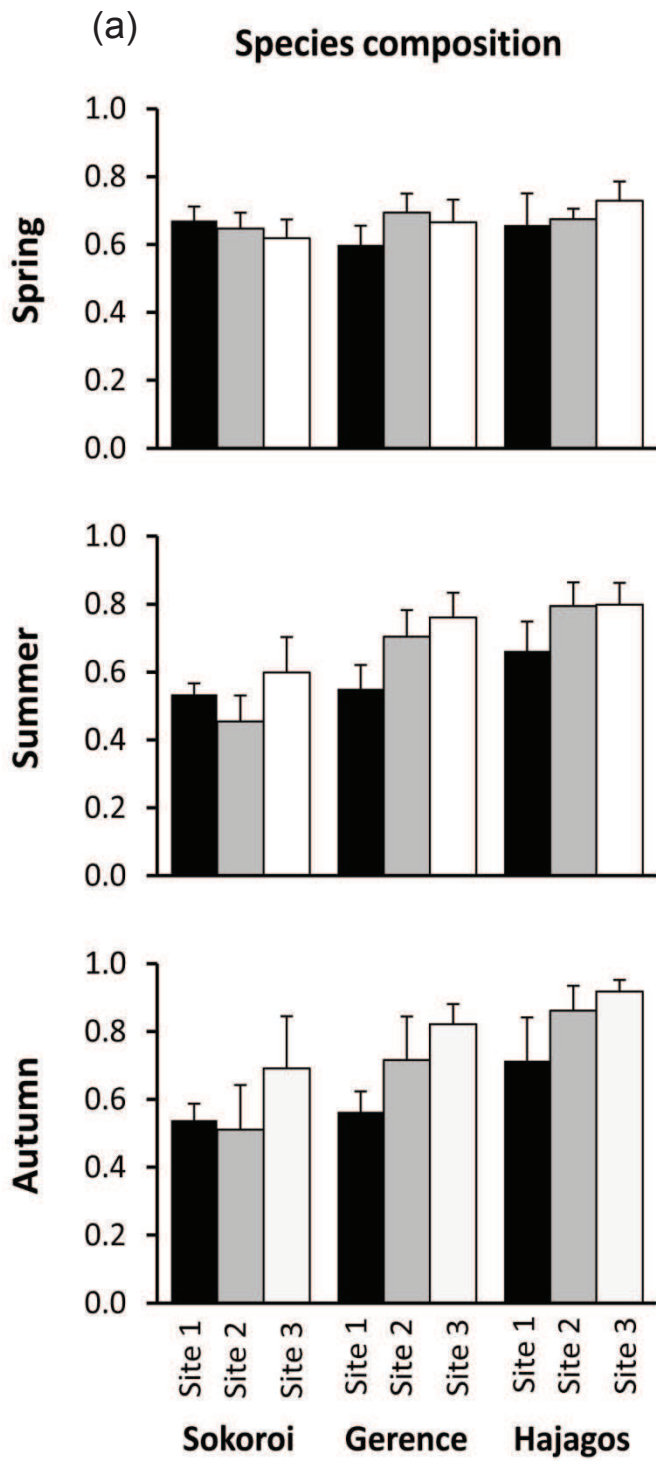
Species richness **Species composition** **Relative abundance**
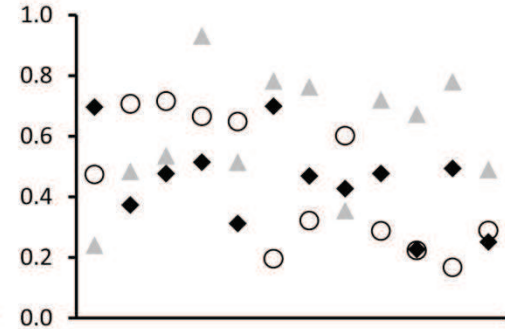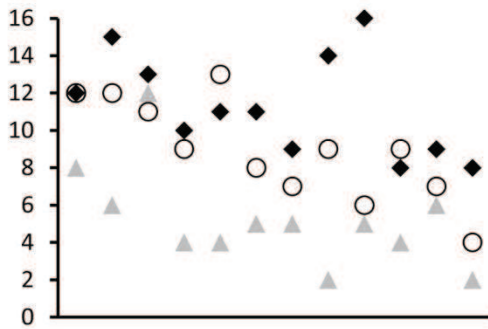
◆ Sokoroi ○ Gerence ▲ Hajagos

Spring
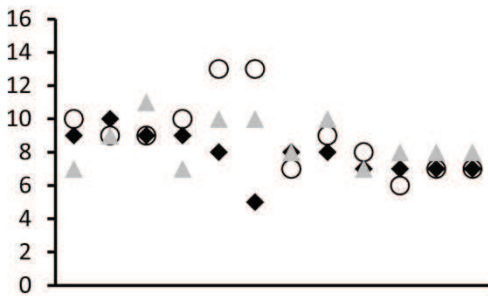
Summer
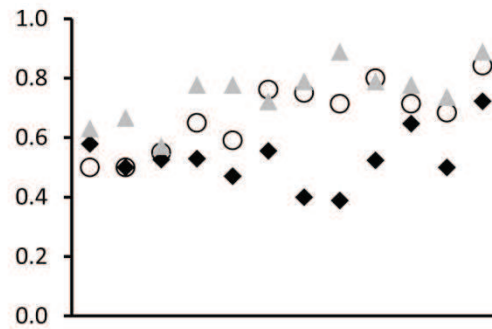
Autumn
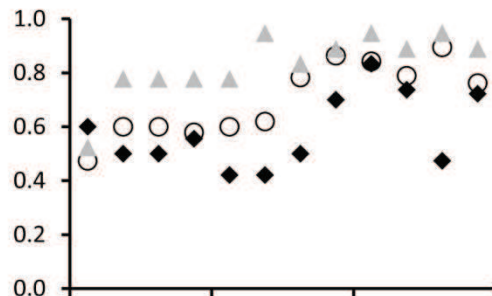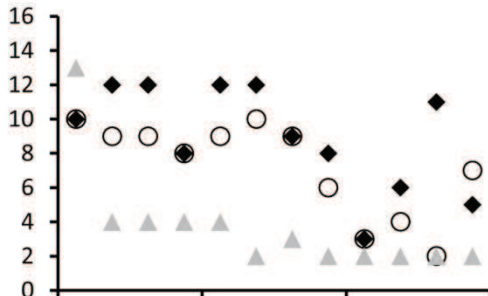
1st 2nd 3rd 4th 1st 2nd 3rd 4th 1st 2nd 3rd 4th
Site1   Site2   Site3

(a)

PCoA axis 2 (9.32%)

PCoA axis 1 (14.19%)

SPRING
AUTUMN
SUMMER

- ● Sokoroi, Site 1
- ■ Sokoroi, Site 2
- ◆ Sokoroi, Site 3
- ● Gerence, Site 1
- ■ Gerence, Site 2
- ◆ Gerence, Site 3
- ● Hajagos, Site 1
- ■ Hajagos, Site 2
- ◆ Hajagos, Site 3

(b)

PCoA axis 2 (13.11%)

PCoA axis 1 (16.83%)

AUTUMN
SUMMER
SPRING

- ● Sokoroi, Site 1
- ■ Sokoroi, Site 2
- ◆ Sokoroi, Site 3
- ● Gerence, Site 1
- ■ Gerence, Site 2
- ◆ Gerence, Site 3
- ● Hajagos, Site 1
- ■ Hajagos, Site 2
- ◆ Hajagos, Site 3

(c)

PCoA axis 2 (9.32%)

PCoA axis 1 (14.19%)

m psepar
prosem
gobgob
tintin
cargib phor
pcar
rhoser
amemel
gymcer
rutrut
ch vimvim
blibjo
leuidu
albalb
lepgib
abrbra
neomel
leuleu
esoluc
neoflu
cobelo
scaery
squcep
perflu

(d)

PCoA axis 2 (13.11%)

PCoA axis 1 (16.83%)

esoluc
amemel
cargib
abrbra
scaery
perflu
squcep
prosem
cobel
rhoser
le ph
neoflu
s
vimvim
pcar
gobgob
misfos
psepar eu lb
leuidu
rutrut