

How pondscapes function: Connectivity matters for biodiversity even across small spatial scales in aquatic metacommunities

Barbara Barta¹, Attila Szabó², Beáta Szabó¹, Robert Ptacnik³, Zsófia Horváth¹, and Csaba Vad¹

¹Centre for Ecological Research

²Swedish University of Agricultural Sciences

³WasserCluster Lunz

June 2, 2023

Abstract

Habitat loss and fragmentation are growing global threats to natural habitats and their networks, posing significant challenges to biodiversity conservation. Among the most vulnerable ecosystems, ponds stand out due to their small sizes exhibiting global declines in numbers and extent. While it is generally agreed that connectivity in habitat networks is crucial for sustaining biodiversity, the effect of connectivity on biodiversity patterns over small-scaled habitat networks has so far received less attention given the general assumption that metacommunities lack spatial structuring on small scales. In this study, we tested whether this holds for multiple passively and actively dispersing organism groups in a well-delineated pond metacommunity of 54 bomb crater ponds situated within 1 km. We investigated the influence of space and environment on species richness and metacommunity structure in these ponds, which share similar age, size, and shape and are subject to strong environmental gradients, making it an ideal study system. We specifically examined the impact of network centrality on species richness and eigenvector-based spatial arrangement on metacommunity structure across different organism groups, including prokaryotes, microeukaryotes, zooplankton, macroinvertebrates, and amphibians. We found that while environmental filtering is the primary driver of community dynamics, there is also a significant spatial signal, particularly for passively dispersing groups, demonstrating the role of the central-peripheral connectivity gradient. These findings highlight the importance of studying and protecting ponds as parts of a network rather than focusing on individual ponds.

How pondscapes function: Connectivity matters for biodiversity even across small spatial scales in aquatic metacommunities

Abstract

Habitat loss and fragmentation are growing global threats to natural habitats and their networks, posing significant challenges to biodiversity conservation. Among the most vulnerable ecosystems, ponds stand out due to their small sizes exhibiting global declines in numbers and extent. While it is generally agreed that connectivity in habitat networks is crucial for sustaining biodiversity, the effect of connectivity on biodiversity patterns over small-scaled habitat networks has so far received less attention given the general assumption that metacommunities lack spatial structuring on small scales. In this study, we tested whether this holds for multiple passively and actively dispersing organism groups in a well-delineated pond metacommunity of 54 bomb crater ponds situated within 1 km. We investigated the influence of space and environment on species richness and metacommunity structure in these ponds, which share similar age, size, and shape and are subject to strong environmental gradients, making it an ideal study system. We specifically examined the impact of network centrality on species richness and eigenvector-based spatial arrangement on metacommunity structure across different organism groups, including prokaryotes, microeukaryotes, zoo-

plankton, macroinvertebrates, and amphibians. We found that while environmental filtering is the primary driver of community dynamics, there is also a significant spatial signal, particularly for passively dispersing groups, demonstrating the role of the central-peripheral connectivity gradient. These findings highlight the importance of studying and protecting ponds as parts of a network rather than focusing on individual ponds.

Keywords: multi-group, spatial network, centrality, zooplankton, microbe, invertebrate, dispersal

Introduction

Connectivity in habitat networks is a major driver of local and regional biodiversity patterns via species dispersal (Taylor *et al.* , 1999; Leibold *et al.* , 2004; Fletcher *et al.* , 2016). Among discrete habitat patches, such as ponds, connectivity is frequently approximated as a function of distance, where the highest levels of dispersal are assumed among neighbouring habitats (Calabrese & Fagan, 2004). Across small spatial scales, i.e., up to a few hundred metres, dispersal levels are generally assumed to be high enough to efficiently counteract local extinction events (Langenheder & Lindström, 2019), thereby contributing to efficient species sorting. Within a few meters, e.g., in rock pool metacommunities, occasional biotic homogenisation may occur via mass effects, resulting from extremely high dispersal rates (overspill; Vanschoenwinkel *et al.* , 2007). The relevance of these processes, however, decays rapidly with increasing distance and most natural metacommunities are rather assembled along a gradient of intermediate to low dispersal, depending on the actual spatial scale, landscape connectance, and species traits (Leibold & Chase, 2018). While there is a general agreement on the importance of sufficient connectivity in habitat networks for the sustenance of biodiversity, its above-mentioned scale and context dependence and their effect on biodiversity patterns remain lesser understood.

Pondscapes are regional networks of ponds clustered in a landscape (Biggs *et al.* , 1994; Boothby, 1997; Baguette *et al.* , 2013). Since the introduction of the term, it has been frequently argued that ponds function as habitat networks, e.g., by serving as stepping stones for aquatic taxa (Pereira, Segurado, & Neves, 2011), and that their value for biodiversity lies in their numbers (Oertli *et al.* , 2002; Martínez-Sanz *et al.* , 2012). Pondscapes are shrinking worldwide, which increases the isolation between the remaining habitats (Thornhill *et al.* , 2018). These changes are predicted to result in biodiversity loss both at the local (Horváth *et al.* , 2019; Holmes *et al.* , 2020) and the landscape scale (Horváth *et al.* , 2019). This calls for a better understanding of connectivity within their remaining networks, both for conservation and efficient restoration measures. Graph-based measures can be important connectivity metrics to understand the functioning of habitat networks (Urban & Keitt, 2001), and for such analyses, well-delineated networks of pools and ponds could provide ideal test cases. Surprisingly though, such explicit network analyses involving ponds are still scarce (exceptions include e.g., Thornhill *et al.* , 2018; Godet & Clauzel, 2021) including their utilisation for metacommunity theory in understanding drivers of biodiversity patterns (Borthagaray, Berazategui, & Arim, 2015; Cunillera-Montcusí *et al.* , 2021).

While there is an upsurge in studies focusing on the role of spatial configuration within pond networks in shaping the structure and dynamics of aquatic metacommunities, there is a significant knowledge gap in understanding the role of pond configuration in networks over smaller spatial scales (within a few km, where most of the dispersal events supposedly happen). Most of the previous works come from networks over large distances, which at the same time did not capture all ponds in the landscape (Soininen *et al.* , 2007; Florencio *et al.* , 2014; Gálvez *et al.* , 2023). While these studies are very informative on distance-based community similarities and general community patterns, they cannot offer a full view of the functioning of the habitat network per se. A central-peripheral connectivity gradient in a pondscape might underlie the main biodiversity gradients, with central ponds hosting more species or playing a relatively more important role (Borthagaray, Berazategui, & Arim, 2015; Cunillera-Montcusí *et al.* , 2020; Holmes *et al.* , 2020). However, these patterns are largely masked in such study designs and can only reliably be revealed in pondscapes where the entire network is considered for the calculation of network properties (i.e., measures of centrality).

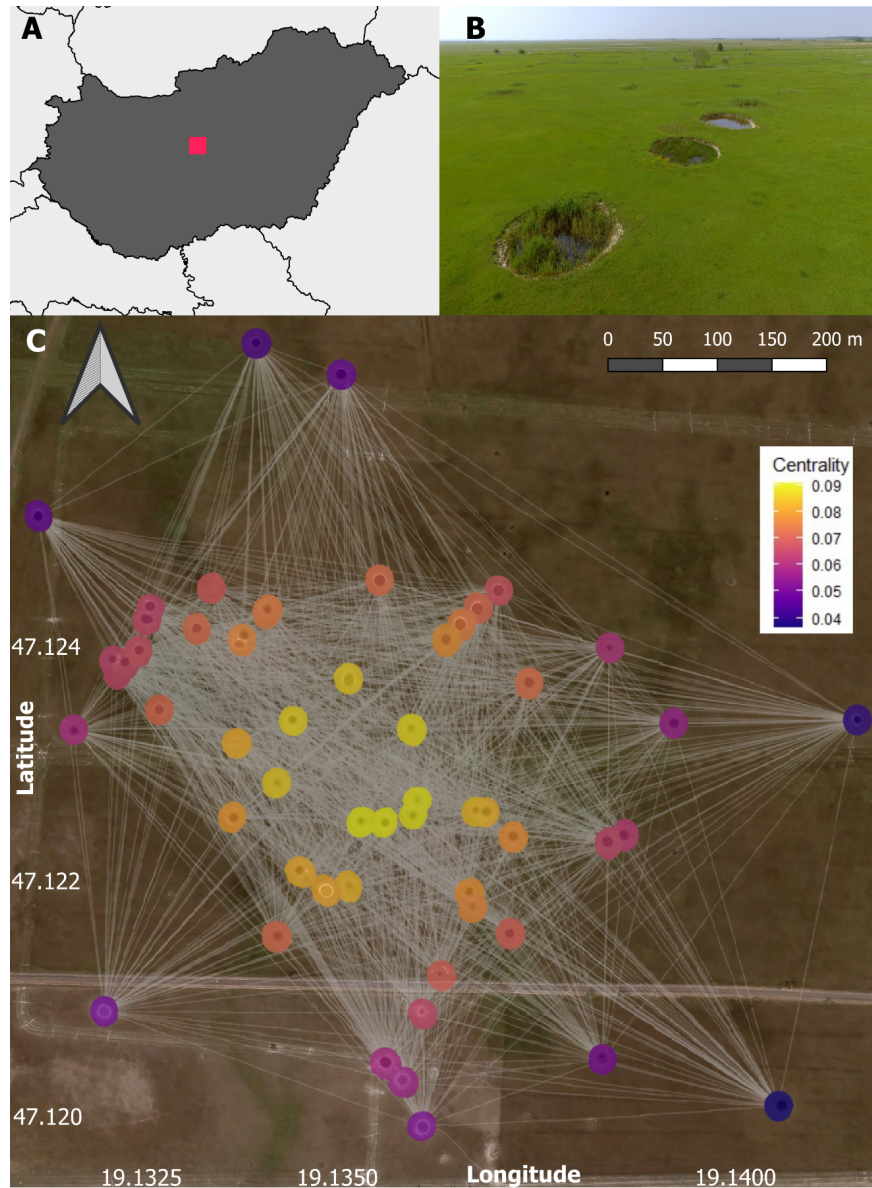
In addition to spatial distances, the dispersal capacity of organisms among habitat patches also varies according to key traits such as dispersal mode and body size. Passive dispersers, such as microscopic organisms, rely on dispersal vectors, such as wind or animals (Bilton, Freeland, & Okamura, 2001; Mony *et al.* 2022). Their dispersal potential is expected to decrease with body or propagule size (Bie *et al.* , 2012). Active dispersers include organisms with life stages that allow movement outside of water independent of vectors, such as amphibians or flying insects. As these groups can actively select suitable habitat patches, their distribution patterns are expected to be predominantly determined by the local environmental conditions rather than spatial processes (Heino, 2013). In contrast to passive dispersers, the dispersal ability of active dispersers is expected to increase with body size (Alzate & Onstein, 2022; Cote *et al.* , 2022). While small-bodied chironomids mostly stay within 200 m of their emergence site (Khan 2012), larger aquatic heteropterans disperse readily over to 1.6 km (Briers 1998; Choi & Kim 2009), and most amphibian species cover distances up to a few kilometres (Smith & Green, 2005).

Small passive dispersers and active, good dispersers are assumed to disperse well over large distances and thus their communities are not expected to show spatial patterns across small spatial scales related to dispersal limitation. Consequently, small scales stretching over only a few kilometres are often neglected in metacommunity studies. Nonetheless, studying community patterns at small spatial scales could still be informative: large passive dispersers may display spatial patterns across small scales due to low dispersal ability (Cottenie & De Meester, 2003; Cottenie *et al.* , 2003) and even active dispersers might show patterns from limited dispersal due to territorial behaviour (McCauley, 2010) or differences in the surrounding matrix (Gall, Chaput-Bardy, & Husté, 2017). Even so, recent studies tend to concentrate on a single or a limited number of organism groups, while multi-group studies, covering a wide range of taxonomic groups with diverse dispersal traits are still scarce, even though they have been increasing in recent years (e.g. Bie *et al.*, 2012, De Marco *et al.*, 2014, Gálvez *et al.*, 2023). Such studies are crucial as they can corroborate the overall functioning of connectivity networks within pondscapes by highlighting congruent patterns across diverse taxa.

The aim of the current study was to investigate how the relative position of ponds in a well-delineated pond network affects local species richness and metacommunity structure in metacommunities of multiple organism groups, encompassing a variety of dispersal traits. As a model system, we used a cluster of closely-spaced bomb crater ponds, with over 50 habitats situated on a spatial scale of a few hundred meters. We specifically tested whether local species richness scales positively with the centrality of the ponds in a graph-based approach. In addition, we quantified the extent to which spatial configuration predicts metacommunity structure based on eigenvector analysis. We expected to detect spatial signals despite the small spatial extent, linked to the presumed higher dispersal rates among more central ponds, which should result in a gradient of biodiversity along the gradient of connectivity. Further, we also predicted that the strength of spatial signals will vary between organism groups linked to body size and dispersal mode as key traits. Specifically, we expected weaker spatial signals in the richness and community structure of taxa assumed to be good dispersers (i.e., especially for large active and small passive taxa) and stronger for groups more constrained due to larger size or lower motility.

Materials and Methods

Study area



A network of 112 bomb crater ponds forming a well-delineated pondscape relatively isolated from other waterbodies is situated in the Kiskunság area, Central Hungary (Fig. 1). These ponds were likely created during World War II by mistargeted bombing on a sodic meadow. They are saline waters mostly dominated by sodium carbonates and hydrocarbonates, and despite being the same age and in close proximity to each other, they vary in many of their environmental and morphological characteristics including hydroperiod, surface area, depth and vegetation cover (Vadet *al.* , 2017). The ponds are not physically connected thus dispersal is expected to occur via wind, animal vectors or active movement of the organisms. *Figure 1. A) Location of the study region near Apaj in Hungary. B) Drone photo of a smaller part of the bomb crater pond*

network. C) Location of the 54 bomb crater ponds with their closeness centrality scores indicated by a colour gradient (Google Earth Pro, 2014)

Sample collection and environmental data

Fieldwork was carried out between 7 and 9 May 2014. We surveyed all ponds holding water (i.e., omitting the smallest and already dry ephemeral habitats), resulting in a total of 54 ponds. The average distance between these ponds is 285 m and all habitats are situated within a radius of 0.5 km (min. and max. distances between the centre point of ponds: 9–863 m). A range of physical and chemical parameters was recorded on site including water depth (cm), diameter (m), and open surface area (%), emergent and submerged vegetation (%). Conductivity ($\mu\text{S cm}^{-1}$) and pH were measured using a field multimeter (Eutech CyberScan PCD 650). Per pond, a total of 10 L of water was collected and mixed from 10 randomly chosen points, 1 L of this composite water sample was taken for water chemistry analysis and community sequencing of pro- and microeukaryotes. 1–50 mL (depending on turbidity) of this water sample was filtered through a nitrocellulose membrane filter (\varnothing 47 mm, 0.22 μm pore size). These filters were stored at -20°C until DNA extraction. Concentrations of total phosphorus (TP, mg L^{-1}), total suspended solids (TSS, mg L^{-1}), dissolved inorganic nitrogen (DIN, mg L^{-1}), calcium (Ca^{2+} , mg L^{-1}), and chlorophyll *a* (*Chl-a* $\text{}\mu\text{g L}^{-1}$) were measured in the lab as detailed in Vadet *et al.*, (2017).

To collect zooplankton samples, 10 L of water was randomly collected from the open water and sieved through a 45- μm mesh. Macroinvertebrates were sampled using sweep netting standardised to 3 minutes (frame-size: 0.25x0.25 m^2 , mesh size: 500 μm), for which all microhabitats present in a pond were included. Zooplankton and macroinvertebrate samples were preserved in 70% ethanol for further analysis. For the amphibian survey, hand netting maximised to 15 minutes, visual searches, and dip netting for tadpoles and newt larvae were applied. All identified specimens were released back to their habitat. For a more detailed description of the study area and sampling procedures, see Vad *et al.* (2017)

Sample processing and community datasets

The isolation of community DNA from the membrane filters was carried out using the PowerSoil® DNA Isolation Kit (MO BIO Laboratories Inc.). Prokaryotic 16S and microeukaryotic 18S rRNA gene amplification and sequencing were performed by LGC Genomics (Berlin, Germany). For gene amplification, the following primer pairs were applied: EMBf 515F (GTGYCAGCMGCCGCGGTAA, Parada *et al.*, 2016) – EMBr 806R (GGACTACNVGGGTWCTAAT, Apprill *et al.*, 2015) for the V4 region of the prokaryotic 16S rRNA gene and UnivF-1183mod (AATTTGACTCAACRCGGG) – UnivR-1443mod (GRGCATCACAGACCTG) (Ray *et al.*, 2016) for the V7 region of the eukaryotic 18S rRNA gene. Sequencing was carried out on an Illumina MiSeq platform. For a detailed description of the entire procedure, see Szabó *et al.* (2022). Amplicon readsets were analysed using mothur v1.43.0 (Schloss *et al.*, 2009) involving, sequence processing, taxonomic assignments, and OTU picking with the MiSeq SOP as a reference (<http://www.mothur.org/wiki/MiSeq-SOP>; Kozich *et al.*, 2013, downloaded on 12th November 2020). Additional quality filtering steps were implemented to minimize the presence of sequence artefacts. These steps included the adjustment of the deltaq parameter to 10 in the 'make.contigs' command, primer removal from both ends of the sequences, chimera identification and removal using the mothur-implemented version of VSEARCH and the exclusion of singleton reads. De-noising was carried out using mothur's 'pre.cluster' command with the suggested 2 bp difference cutoff. Read alignment and taxonomic assignment were performed using the ARB-SILVA SSU Ref NR 138 reference database with a minimum bootstrap confidence score of 80 (Quast *et al.*, 2012). Non-primer-specific taxonomic groups ('Chloroplast', 'Mitochondria' and 'unknown') were excluded from the dataset.

OTUs were selected at the 99% similarity threshold. Taxonomic assignment of 18S rRNA gene OTUs was performed using the 'classify.otu' command with the PR2 v4.12.0 reference database (Guillou *et al.*, 2012). 18S rRNA gene OTUs assigned to taxa Streptophyta, Metazoa, Ascomycota, and Basidiomycota were ex-

cluded from the microeukaryotic dataset. For the taxonomic assignment of prokaryotic OTUs, the TaxAss software (Rohwer *et al.*, 2018) was used with default parameters and the FreshTrain (15 June 2020 release) and ARB-SILVA SSU Ref NR 138 databases. Subsequently, both 16S and 18S OTU sets were rarefied to the read number of the sample having the lowest sequence count. Samples BC40 and BC105 were excluded from the eukaryotic dataset due to a low read count after filtering.

Zooplankton was identified to the lowest possible taxonomic level by microscopic analysis (generally to species), except for bdelloid rotifers which were treated as a single group. Macroinvertebrates were also identified to the lowest possible taxonomic level (generally to family, genus and species levels, depending on the higher taxa) using relevant identification keys. As passively dispersing macroinvertebrates were represented only by four taxa (Vad *et al.*, 2017), they were excluded from the subsequent analyses. For a detailed description of the sample processing and the list of identified taxa, see Vad *et al.*, (2017).

We focused on the following organism groups in our study (Table S1; Supporting information): prokaryotes, phototrophic microeukaryotes, heterotrophic microeukaryotes, crustacean zooplankton (copepods and cladocerans), rotifer zooplankton, dipterans, other macroinvertebrates, and amphibians-reptilians. For community matrices of prokaryotes and microeukaryotes, we used the rarefied 16S and 18S OTU tables. The 18S dataset was split into phototrophs and heterotrophs based on phyla using the ‘phyloseq’ package v1.36.0 (McMurdie & Holmes, 2013) in R 4.1.0 (R Core Team, 2021). Phototrophs mostly included groups capable of photosynthesis, while heterotrophs consisted of the remaining groups. Dipterans were treated separately from other macroinvertebrates as they are considered weak active dispersers (Bilton, Freeland, & Okamura, 2001; Heino, 2013). Other macroinvertebrates included all actively dispersing groups considered intermediate or strong aerial dispersers with flying adults according to Heino (2013).

Statistical analysis

In order to assess the relative importance of environmental and spatial predictors in explaining patterns of taxonomic richness and community similarity, we carried out three sets of variance partitioning analyses using the *varpart* function of the ‘vegan’ package (Oksanen *et al.*, 2020), where either network position (in the case of taxonomic richness) or spatial distance (in the case of taxonomic richness and community similarity) was included as the spatial predictor. We used data from all 54 ponds in all three cases, except for phototrophic and heterotrophic microeukaryotes for which data was available from 52 ponds only. In the environmental dataset, we excluded highly correlated (i.e., Pearson’s $r > 0.75$) environmental variables (turbidity, percentage of reed cover) prior to the analyses. Secchi depth and submerged vegetation cover were also excluded, as Secchi depth delivers the same information as TSS but on a less precise scale, while the percentage of submerged vegetation cover almost exclusively contained zeros. Our final environmental dataset thus contained 10 variables, which were tested for normality and those deviating from it (Shapiro-Wilk test $p < 0.05$) were transformed (conductivity, pH, depth - untransformed; DIN, TP, TSS - natural log; Ca^{2+} , surface area - square root; chlorophyll-*a* - $\log(x+1)$; open surface area - cube transformed).

First, we analysed how taxonomic richness is predicted by the local environment and the relative position of ponds within the habitat network. For this, partial multiple linear regressions for each studied organism group were carried out using the *rda* function of the ‘vegan’ package (Oksanen *et al.*, 2020). As a response variable, richness was included either as OTU (pro- and microeukaryotes) or taxonomic richness (other groups), but hereinafter, we refer to these as ‘taxonomic richness’ for simplicity. Taxonomic richness data of crustacean zooplankton and amphibians-reptilians were square-root transformed to normalise model residuals (inspected via diagnostic plots), while untransformed values were used in the other organism groups. As environmental predictors, we used the first two axes of a Principal Component Analysis (PCA) constructed for the environmental variables, following z-score standardisation. For all subsequent analyses, site scores of PC1 and PC2 axes (explaining >65% of the total variation, Fig. S1, Supporting information) were extracted and used as environmental data. As a spatial predictor, we calculated the closeness centrality index of each pond (Fig. 1 C) based on the spatial distance matrix of all ponds using the ‘igraph’, ‘fields’ and ‘reshape2’ packages (Csárdi & Nepusz, 2006; Wickham, 2007; Nychka *et al.*, 2021). To test for the possible significance

of the unique effects of environment and centrality, a permutation test (2000 permutations) was run for each partial multiple linear regression model using the function *anova* .

We run a second set of variance partitioning (partial multiple linear regression models) for each organism group separately to test if and how taxonomic richness is affected by the environmental variables (PC1 and PC2 axes) and the number of neighbouring ponds. In these models, environmental variables were included as one set of predictors the same way as described above, and the number of ponds within a radius with increasing distance from each local pond over a scale of 0-800 m, with 10 m increments was involved as the spatial predictor. The unique effects of space and environment were calculated the same way as in the first set of variance partitioning 80 times in total for each organism group. The unique effect of space along threshold distance (cut-off distance for calculating pond numbers) was plotted with the help of General Additive Models (GAMs, formula = $y \sim s(x, k=5)$) for each taxonomic group separately with the package ‘ggplot2’ (Wickham, 2016).

Third, we tested how metacommunity structure is predicted by the local environment and spatial configuration. Since we did not have abundance data for all organism groups, statistical analyses were run on presence-absence community data for comparability. Taxa occurring in less than three ponds were omitted given their minor contribution to the overall similarity in the community dataset. Then, any pond which contained no taxa was removed. Dissimilarity matrices were calculated based on Sørensen dissimilarity with the *vegdist* function of the ‘vegan’ package (Oksanen *et al.* , 2020). Here, the initial pool of environmental predictors was represented by the 10 transformed environmental predictors, while the spatial predictors were Moran’s Eigenvector Maps (MEM). These were constructed based on the spatial distance matrix using the *dbMEM* function from the ‘adespatial’ package (Dray *et al.* , 2022) and only the 15 positive MEMs were retained (Bauman *et al.* , 2018; Borcard, Gillet, & Legendre, 2018; Fig. S2; Supporting information). To select significant variables of each set of explanatory variables (environment and space), we applied stepwise selection both for the transformed environmental variables and for the positive MEM eigenvectors using the *ordistep* function (direction=both, perm.max=2000) of the ‘vegan’ package (Oksanen *et al.* , 2020). These selection steps were done separately for each organism group resulting in different sets of retained environmental variables and MEMs (Table S2, Supporting information). We carried out a set of partial distance-based redundancy analyses (dbRDA) using the *capscale* function of the ‘vegan’ package (Oksanen *et al.* , 2020) followed by significance testing with a permutation test (2000 permutations) using the function *anova* .

All statistical analyses were carried out in R 4.1.0 (R Core Team, 2021) and figures were created using the ‘ggplot2’ (Wickham, 2016), ‘pubbr’ (Kassambara, 2020) and ‘car’ packages (Fox & Weisberg, 2019).

Results

Taxonomic richness

Environment (represented by the first two PCA axes) was predominant in explaining variation in taxonomic richness in all organism groups (Fig. 2). It varied between 10.6 and 52.2%, and was statistically significant in almost all organism groups except for phototrophic and heterotrophic microeukaryotes. The effect of relative spatial position (closeness centrality) was also relevant for some taxa varying between 1.9 and 9.3% explained variation. It was the highest and statistically significant in rotifers (9.3%, $F_{(1,50)}=9.08$, $p=0.004$) and crustaceans (1.9%, $F_{(1,50)}=3.94$, $p=0.047$), while only marginally significant in prokaryotes (2.8%, $F_{(1,50)}=3.05$, $p=0.091$) and dipterans (3.4%, $F_{(1,50)}=3.30$, $p=0.068$; Table S3 and S4, Supporting Information). The relationship between taxonomic richness and centrality (with the environment partialled out) was positive in all these groups (Fig. S3, Supporting information).

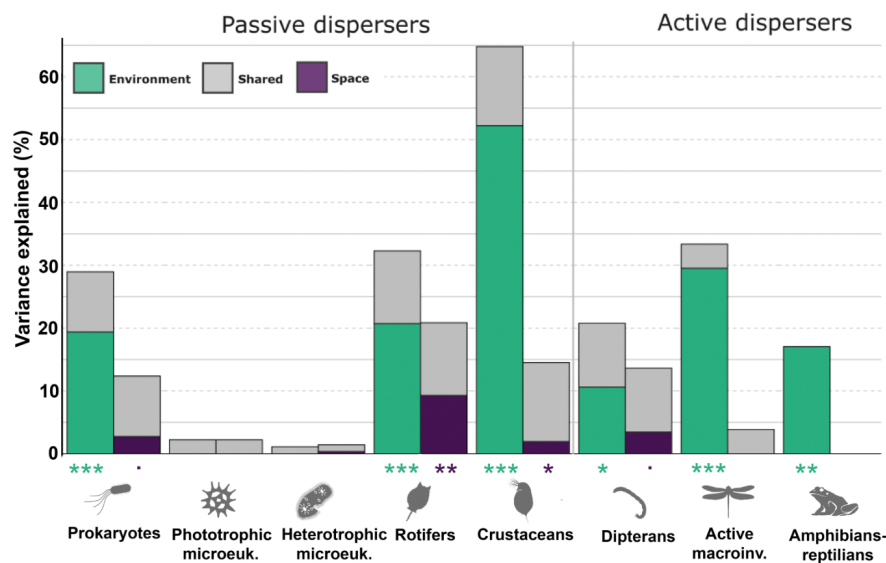


Figure 2. The variance of taxonomic richness explained by environment (PC1 and PC2 axes) and space (closeness centrality) of the studied organism groups of both passive and active dispersers, ordered according to body size within the two categories. The shared portion of the explained variance (grey shading) is shown at the top of the bars of both space and environment. $p < 0.1$, $*p < 0.05$, $**p < 0.005$, $***p < 0.001$

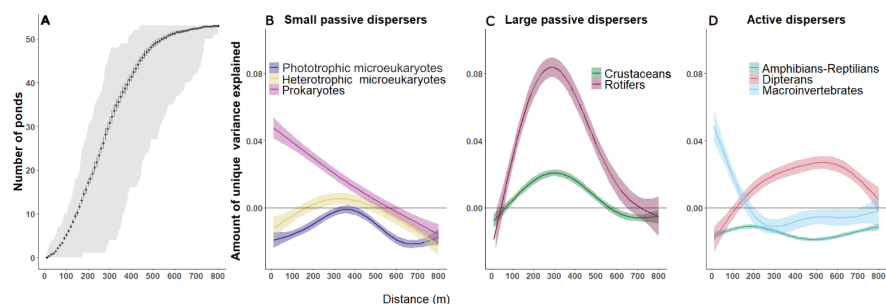


Figure 3. A) The mean number of neighbouring ponds (+/- SE) around each pond within an increasing radius (cut-off distance), where grey shading shows the full range between minimum and maximum values. B-D) Variograms with the amount of unique variance of taxonomic richness explained by the number of ponds within a radius against each cut-off distance, grouped according to dispersal traits.

With increasing radius size around the focal pond, the number of ponds within the radius increased rapidly until around 550 meters where the curve saturates, indicating the most dense part of the pond cluster (Fig. 3 A). Accordingly, the number of ponds above this distance did not explain a considerable amount of variance in taxonomic richness in any of the studied groups (Fig. 3 B-D). At the same time, there was considerable variance in pond numbers within the scales of 150-550 m (Fig. 3 A), with related spatial signals in the local taxonomic richness of four groups (prokaryotes, rotifer and crustacean zooplankton, dipterans Fig. 3 B-D). In both zooplankton groups, the relationship between pond number and taxonomic richness was strongest within a circle radius between 200 and 400 metres, while it was 300-700 metres for dipterans. As the variation explained by the environment did not show a similar spatial structuring, this suggests that indeed neighbouring pond densities are the major driver of the observed pattern (Fig. S4).

Metacommunity structure

In metacommunity structure, the overall pattern was similar to taxonomic richness with the environment explaining more variance than space in most organism groups (Fig. 4). The amount of unique variance explained by the environment varied between 10.1 and 36.4% and it was statistically significant in all organism groups. The unique effect of space varied between 0.7 and 15.6%, being the highest in rotifers and lowest in prokaryotes (Table S5 and S6, Supporting information). It was statistically significant for prokaryotes ($F_{(3,43)}=1.14$, $p=0.0475$), heterotrophic microeukaryotes ($F_{(5,40)}=1.21$, $p=0.026$), rotifers ($F_{(7,41)}=1.95$, $p<0.001$), crustaceans ($F_{(5,44)}=1.70$, $p=0.004$), and dipterans ($F_{(2,45)}=1.96$, $p=0.004$).

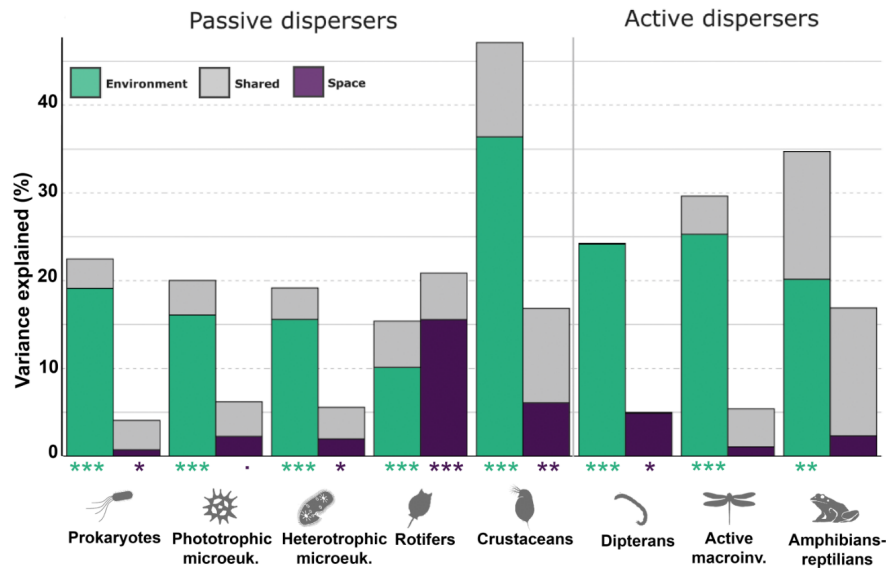


Figure 4. The proportion of unique variance explained by environment (selected environmental variables) and space (selected MEMs) in the community composition data of the various organism groups of both passive and active dispersers, ordered according to body size within the two categories. The shared portion of the explained variance (grey shading) is shown at the top of the bars of both space and environment.

. $p<0.1$, * $p<0.05$, ** $p<0.005$, *** $p<0.001$

Discussion

Our results supported the importance of connectivity for multiple organism groups in a delineated habitat network covering a small spatial scale of a few hundred metres. We found that pond taxonomic richness scales with network position and that spatial configuration has an imprint on metacommunity structure across multiple taxonomic groups. The local environment explained a larger share of the variance in both richness and metacommunity structure than space, indicating a predominant role of species sorting, which is not surprising given the existence of strong environmental gradients (e.g., salinity, vegetation cover). Despite this, we found a clear indication that space also played an important role in structuring the metacommunity of the bomb crater pond network with such a small spatial extent. This held for most of the studied organism groups and for both taxonomic richness and metacommunity structure. Overall, our results highlight the importance of dense habitat networks in sustaining biodiversity. Moreover, the various organism groups differed in the amount of variance explained in their richness and composition by space, and it was likely related to their dispersal abilities according to our expectations.

While metacommunity processes are highly context-dependent, species sorting is expected to be predominant over short spatial scales, especially for microbes (Bie *et al.* , 2012; Hanly & Mittelbach, 2017; Langenheder & Lindström, 2019; Mony *et al.* , 2022). In line with this, we found clear evidence for the predominance of species sorting for all organism groups, but with slightly different environmental variables being relevant for each group. Overall, conductivity, pH, and TSS were among the most important environmental predictors, which is in line with data from similar saline temporary waters from the region (Horváth *et al.* , 2014; Horváth, Vad, & Ptacnik, 2016; Márton *et al.* , 2023). But while there was a strong environmental signal underlying community patterns, a significant spatial effect also emerged in multiple groups. These included the larger-bodied passively dispersing rotifer and crustacean zooplankton and the weak flyer dipterans, suggesting some level of dispersal limitation in their case.

The evidence for spatial structuring has been confirmed with complementary analyses, as we both tracked similarities based on spatial eigenvectors and explored the predictive role of relative spatial position (closeness centrality). The different sets of analyses gave consistent results, revealing the importance of spatial effects in the richness and metacommunity structure of the three organisms groups expected to be the weakest dispersers (i.e., large passive dispersers and weak active dispersers). For metacommunity structure, the first five MEM eigenvectors were the most frequently selected spatial explanatory variables across the organism groups. These correspond to the largest eigenvalues indicating coarse-scaled spatial structuring. Furthermore, multiple of these significant MEM eigenvectors (MEM 3, 4, and 5) illustrated the main spatial structuring between central and peripheral sites, similar to our results on centrality and richness. These overall indicated an important network effect in several organism groups, where richness is enhanced by a more central position of a local habitat via a higher number of surrounding patches, presumably related to the higher frequency of dispersal.

Overall, the richness and community structure of prokaryotes and microeukaryotes showed only weak spatial structuring, which is in line with other studies on spatial patterns in prokaryotes across various spatial scales (Beisner *et al.* , 2006; Van Der Gucht *et al.* , 2007; Bie *et al.* , 2012; Padial *et al.* , 2014). These taxa are easily dispersed by the wind even across vast distances (Smith *et al.* , 2013; Mony *et al.* , 2020) or via zoochory at small spatial scales (Lindström & Langenheder, 2012; Szabó *et al.* , 2022). Similarly, we did not find any evidence for spatial structuring in communities of actively flying macroinvertebrates (excluding dipterans), amphibians and reptilians. This was in line with our expectation, as these groups are unlikely to show patterns related to dispersal limitation due to their abilities to move over distances larger than in our study system (Ficetola *et al.* , 2004; Smith & Green, 2005; Heino, 2013; Florencio *et al.* , 2014; Godet & Clauzel, 2021). At the same time, spatial signals in zooplankton richness and community structure were evident. These indicated some level of dispersal limitation at the spatial scale of our study in a passively dispersing group with a large body compared to microbes. This finding aligns with previous studies that showed the importance of pond centrality for *Daphnia* metapopulation structure over a comparable spatial scale (Holmes *et al.* , 2020) and the role of connectivity in structuring zooplankton metacommunities over larger scales of hundreds of square kilometers (Cottenie *et al.* , 2003; Soininen *et al.* , 2007).

Therefore, connectivity is important in pondscales even across small spatial scales for at least some members of the metacommunity. The different groups of organisms that make up a metacommunity differ in their traits and the scales relevant to them in terms of spatial processes. Therefore, in multi-group studies, it is worth considering small spatial scales even if some groups are not expected to show spatial patterns. Additionally, the varying responses of the different groups indicate different structuring processes, i.e., the small but significant spatial effect on community structure may indicate a certain level of mass effect for microeukaryotes, while the largest-bodied actively dispersing groups with the best dispersal abilities showed efficient species sorting at the same spatial scale. In several groups, the effect of connectivity was found to be more important when metacommunity structure was considered compared to taxonomic richness indicating that more connected patches do not necessarily hold more species but connectivity within the pondscape is an important determinant of metacommunity structure.

There remains a large portion of unexplained variation in taxonomic richness and metacommunity structure

for all groups, similar to other metacommunity studies (Soininen *et al.* , 2007; Vanormelingen *et al.* , 2008; Bie *et al.* , 2012). Our analyses were based on an exhaustive dataset of the environmental variables, and three complementary metrics of spatial configuration and both environmental and spatial variables were well-represented making their effects less likely to be underestimated. At the same time, biotic interactions (competition, grazing, predation, mutualism, parasitism etc.) are likely to play a role in metacommunity structuring and may mask the effects of space or environment (Van De Meutter, Stoks and De Meester, 2008; Verreydt *et al.*, 2012, Mony *et al.*, 2022). Apart from such trophic relationships, interspecific competition at the same trophic level can also affect spatial patterns (Thompson *et al.*, 2020; Guzman *et al.*, 2022). Finally, our study is only a snapshot in a presumably highly dynamic system and temporal aspects may need to be explored along with historical processes (Vyverman *et al.* , 2007; Vanormelingen *et al.* , 2008; Thompson *et al.* , 2020; Guzman *et al.* , 2022). Dormant eggs and other resting stages integrate past dispersal events and this could have masked recent dispersal events in passive dispersers in our study (Wisnoski, Leibold, & Lennon, 2019; Holyoak, Caspi, & Redosh, 2020; Wisnoski & Shoemaker, 2022). Incorporating these effects in future studies would provide novel in-depth knowledge of the additional processes acting in such pondscapes, further increasing our understanding of how these habitat networks function.

In conclusion, we found that both space and environment shape the metacommunity of a pondscape even when the spatial extent is relatively small. Differences in dispersal traits between organism groups are likely attributable to group-specific differences in spatial patterns, with communities of actively dispersing larger animals and small passive dispersers showing weak or no spatial signals. In contrast, communities of weak-disperser macroinvertebrates and especially large-bodied passive dispersers are structured by space to a greater extent. The fact that spatial patterns occur in metacommunity structure and central ponds in the network host higher richness highlights the importance of studying and protecting ponds as parts of a network. This needs to be taken into consideration during conservation planning to maximise the protection of overall biodiversity at both the local and landscape levels.

References

- Alzate, A. & Onstein, R.E. (2022) Understanding the relationship between dispersal and range size. *Ecol. Lett.* **25** , 2303–2323, doi: 10.1111/ELE.14089
- Apprill, A., McNally, S., Parsons, R. & Weber, L. (2015) Minor revision to V4 region SSU rRNA 806R gene primer greatly increases detection of SAR11 bacterioplankton. *Aquat. Microb. Ecol.* **75** , 129–137, doi: 10.3354/ame01753.
- Baguette, M., Blanchet, S., Legrand, D., Stevens, V.M. & Turlure, C. (2013) Individual dispersal, landscape connectivity and ecological networks. *Biol. Rev.* **88** , 310–326, doi: 10.1111/BRV.12000
- Bauman, D., Drouet, T., Fortin, M.-J. & Dray, S. (2018) Optimizing the choice of a spatial weighting matrix in eigenvector-based methods. *Ecology* **99** , 2159–2166, doi: 10.1002/ecy.2469.
- 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, doi: 10.1890/0012-9658(2006)87[2985:TROEAS]2.0.CO;2.
- Bie, T., Meester, L., Brendonck, L., Martens, K., Goddeeris, B., Ercken, D., Hampel, H., Denys, L., Vanhecke, L., Gucht, K., Wichelen, J., Vyverman, W. & Declerck, S.A.J. (2012) Body size and dispersal mode as key traits determining metacommunity structure of aquatic organisms. *Ecol. Lett.* **15** , 740–747, doi: 10.1111/j.1461-0248.2012.01794.x
- Biggs, J., Corfield, A., Walker, D., Whitfield, M. & Williams, P. (1994) New approaches to the management of ponds. *Br. Wildl.* **5** (5), 273– 287.

- Bilton, D.T., Freeland, J.R. & Okamura, B. (2001) Dispersal in freshwater invertebrates. *Annu. Rev. Ecol. Syst.* **32** , 159–181, doi: 10.1146/annurev.ecolsys.32.081501.114016
- Boothby, J. (1997) Pond conservation: towards a delineation of pondscape. *Aquat. Conserv. Mar. Freshw. Ecosyst* **7** , 127–132, doi: 10.1002/(SICI)1099-0755(199706)7:2<127::AID-AQC224>3.0.CO;2-6.
- Borcard, D., Gillet, F. & Legendre, P. (2018) Spatial Analysis of Ecological Data. In: *Numerical Ecology with R Use R!* (ed. by D. Borcard, F. Gillet, & P. Legendre), 299–367. Springer International Publishing, Cham.
- Borthagaray, A.I., Berazategui, M. & Arim, M. (2015) Disentangling the effects of local and regional processes on biodiversity patterns through taxon-contingent metacommunity network analysis. *Oikos* **124** , 1383–1390, doi: 10.1111/oik.01317
- Calabrese, J.M. & Fagan, W.F. (2004) Connectivity metrics. *Front. Ecol. Environ.* **2** , 529–536, doi: 10.1890/1540-9295(2004)002[0529:ACGTCM]2.0.CO;2.
- Choi, H. & Kim, J.G. (2009). Landscape analysis of the effects of artificial lighting around wetland habitats on the giant water bug *Lethocerus deyrollei* in Jeju Island. *J. Ecol. Field Biol.*, **32**, 83–86.
- Cote, J., Dahirel, M., Schtickzelle, N., Altermatt, F., Ansart, A., Blanchet, S., Chaine, A.S., De Laender, F., De Raedt, J., Haegeman, B., Jacob, S., Kaltz, O., Laurent, E., Little, C.J., Madec, L., Manzi, F., Masier, S., Pellerin, F., Pennekamp, F., *et al.* (2022) Dispersal syndromes in challenging environments: A cross-species experiment. *Ecol. Lett.* **25** , 2675–2687, doi: 10.1111/ele.14124.
- Cottenie, K. & De Meester, L. (2003) Connectivity and cladoceran species richness in a metacommunity of shallow lakes. *Freshw. Biol.* **48** , 823–832, doi: 10.1046/J.1365-2427.2003.01050.X
- Cottenie, K., Michels, E., Nuytten, N. & De Meester, L. (2003) Zooplankton metacommunity structure: Regional vs. Local processes in highly interconnected ponds. *Ecology* **84** , 991–1000, doi: 10.1890/0012-9658(2003)084[0991:ZMSRVL]2.0.CO;2.
- Csardi, G. & Nepusz, T. (2006) The igraph software package for complex network research. *InterJournal Complex Syst.* **1695** .
- Cunillera-Montcusi, D., Boix, D., Sala, J., Compte, J., Tornero, I., Quintana, X.D. & Gascon, S. (2020) Large- and small-regional-scale variables interact in the dispersal patterns of aquatic macroinvertebrates from temporary ponds. *Aquat. Ecol.* **54** , 1041–1058, doi: 10.1007/s10452-020-09792-8.
- Cunillera-Montcusi, D., Borthagaray, A.I., Boix, D., Gascon, S., Sala, J., Tornero, I., Quintana, X.D. & Arim, M. (2021) Metacommunity resilience against simulated gradients of wildfire: disturbance intensity and species dispersal ability determine landscape recover capacity. *Ecography*. **44** , 1022–1034, doi: 10.1111/ecog.05347.
- De Marco, P., Nogueira, D.S., Correa, C.C., Vieira, T.B., Silva, K.D., Pinto, N.S., Bichsel, D., Hirota, A.S.V., Vieira, R.R.S., Carneiro, F.M., de Oliveira, A.A.B., Carvalho, P., Bastos, R.P., Ilg, C. & Oertli, B. (2014) Patterns in the organization of Cerrado pond biodiversity in Brazilian pasture landscapes. *Hydrobiologia* **723** , 87–101, doi: 10.1007/s10750-013-1695-2.
- Dray, S., Bauman, D., Blanchet, G., Borcard, D., Clappe, S., Guenard, G., Jombart, T., Larocque, G., Legendre, P., Naima, M. & Wagner, H.H. (2022) adespatial: Multivariate Multiscale Spatial Analysis.
- Ficetola, G.F., Padoa-Schioppa, E., Monti, A., Massa, R., Bernardi, F. De & Bottoni, L. (2004) The importance of aquatic and terrestrial habitat for the European pond turtle (*Emys orbicularis*): implications for conservation planning and management. *Can. J. Zool.* **82** , 1704–1712, doi: 10.1139/z04-170.
- Fletcher, R.J., Burrell, N.S., Reichert, B.E., Vasudev, D. & Austin, J.D. (2016) Divergent Perspectives on Landscape Connectivity Reveal Consistent Effects from Genes to Communities. *Curr. Landsc. Ecol. Reports* **1** , 67–79, doi: 10.1007/s40823-016-0009-6.

- Florencio, M., Diaz-Paniagua, C., Gomez-Rodriguez, C. & Serrano, L. (2014) Biodiversity patterns in a macroinvertebrate community of a temporary pond network. *Insect Conserv. Divers.* **7** , 4–21, doi: 10.1111/icad.12029.
- Fox, J. & Weisberg, S. (2019) *An R Companion to Applied Regression* , Third Edit. Thousand Oaks CA: Sage.
- Gall, M. Le, Chaput-Bardy, A. & Huste, A. (2017) Context-dependent local movements of the blue-tailed damselfly, *Ischnura elegans* : effects of pond characteristics and the landscape matrix. *J Insect Conserv* **21** , 243–256, doi: 10.1007/s10841-017-9971-5.
- Galvez, A., Peres-Neto, P.R., Castillo-Escriva, A., Bonilla, F., Camacho, A., Garcia-Roger, E.M., Iepure, S., Miralles-Lorenzo, J., Monros, J.S., Olmo, C., Picazo, A., Rojo, C., Rueda, J., Sahuquillo, M., Sasa, M., Segura, M., Armengol, X. & Mesquita-Joanes, F. (2023) Inconsistent response of taxonomic groups to space and environment in mediterranean and tropical pond metacommunities. *Ecology***104** , e3835, doi: 10.1002/ecy.3835.
- Godet, C. & Clauzel, C. (2021) Comparison of landscape graph modelling methods for analysing pond network connectivity. *Landsc. Ecol.***36** , 735–748, doi: 10.1007/s10980-020-01164-9.
- Google Earth Pro V 7.3.4.8642 (9.6.2014). Apaj bomb crater ponds, Hungary. 47deg07'23.27"N, 19deg08'13.35"E, Eye alt 1.38 km. (Accessed 26.10. 2022.) Image 2022 Maxar Technologies
- Guillou, L., Bachar, D., Audic, S., Bass, D., Berney, C., Bittner, L., Boutte, C., Burgaud, G., de Vargas, C., Decelle, J., del Campo, J., Dolan, J.R., Dunthorn, M., Edvardsen, B., Holzmann, M., Kooistra, W.H.C.F., Lara, E., Le Bescot, N., Logares, R., *et al.* (2012) The Protist Ribosomal Reference database (PR2): a catalog of unicellular eukaryote Small Sub-Unit rRNA sequences with curated taxonomy. *Nucleic Acids Res.* **41** , D597–D604, doi: 10.1093/nar/gks1160.
- Guzman, L.M., Thompson, P.L., Viana, D.S., Vanschoenwinkel, B., Horvath, Z., Ptacnik, R., Jeliaskov, A., Gascon, S., Lemmens, P., Anton-Pardo, M., Langenheder, S., De Meester, L. & Chase, J.M. (2022) Accounting for temporal change in multiple biodiversity patterns improves the inference of metacommunity processes. *Ecology* **103** , e3683, doi: 10.1002/ecy.3683.
- Hanly, P.J. & Mittelbach, G.G. (2017) The influence of dispersal on the realized trajectory of a pond metacommunity. *Oikos* **126** , 1269–1280, doi: 10.1111/OIK.03864
- Heino, J. (2013) Does dispersal ability affect the relative importance of environmental control and spatial structuring of littoral macroinvertebrate communities? *Oecologia* **171** , 971–980, doi: 10.1007/s00442-012-2451-4.
- Holmes, C.J., Rapti, Z., Pantel, J.H., Schulz, K.L. & Caceres, C.E. (2020) Patch centrality affects metapopulation dynamics in small freshwater ponds. *Theor. Ecol.* **13** , 435–448, doi: 10.1007/s12080-020-00463-w.
- Holyoak, M., Caspi, T. & Redosh, L.W. (2020) Integrating Disturbance, Seasonality, Multi-Year Temporal Dynamics, and Dormancy Into the Dynamics and Conservation of Metacommunities. *Front. Ecol. Evol.***8** , doi: 10.3389/fevo.2020.571130.
- Horvath, Z., Ptacnik, R., Vad, C.F. & Chase, J.M. (2019) Habitat loss over six decades accelerates regional and local biodiversity loss via changing landscape connectance. *Ecol. Lett.* **22** , 1019–1027, doi: 10.1111/ele.13260.
- Horvath, Z., Vad, C.F. & Ptacnik, R. (2016) Wind dispersal results in a gradient of dispersal limitation and environmental match among discrete aquatic habitats. *Ecography (Cop.)*. **39** , 726–732, doi: 10.1111/ecog.01685.
- Horvath, Z., Vad, C.F., Toth, A., Zsuga, K., Boros, E., Voros, L. & Ptacnik, R. (2014) Opposing patterns of zooplankton diversity and functioning along a natural stress gradient: when the going gets tough, the tough

get going. *Oikos* **123** , 461–471, doi: 10.1111/j.1600-0706.2013.00575.x.

Hyseni, C., Heino, J., Bini, L.M., Bjelke, U. & Johansson, F. (2021) The importance of blue and green landscape connectivity for biodiversity in urban ponds. *Basic Appl. Ecol.* **57** , 129–145, doi: 10.1016/j.baae.2021.10.004.

Kassambara, A. (2020) ggpubr: “ggplot2” Based Publication Ready Plots.

Khan, H.R. (2012) Effects of dispersal on community structure of aquatic insects in Arctic lakes and streams. Umea University. <https://www.diva-portal.org/smash/get/diva2:580149/FULLTEXT02>

Kozich, J.J., Westcott, S.L., Baxter, N.T., Highlander, S.K. & Schloss, P.D. (2013) Development of a Dual-Index Sequencing Strategy and Curation Pipeline for Analyzing Amplicon Sequence Data on the MiSeq Illumina Sequencing Platform. *Appl. Environ. Microbiol.* **79** , 5112–5120, doi: 10.1128/AEM.01043-13.

Langenheder, S. & Lindstrom, E.S. (2019) Factors influencing aquatic and terrestrial bacterial community assembly. *Environ. Microbiol. Rep.* **11** , 306–315, doi: 10.1111/1758-2229.12731

Leibold, M.A. & Chase, J.M. (2018) *Metacommunity Ecology, Volume 59* . Princeton University Press.

Leibold, M.A., Holyoak, M., Mouquet, N., Amarasekare, P., Chase, J.M., Hoopes, M.F., Holt, R.D., Shurin, J.B., Law, R., Tilman, D., Loreau, M. & Gonzalez, A. (2004) The metacommunity concept: A framework for multi-scale community ecology. *Ecol. Lett.* **7** , 601–613, doi: 10.1111/j.1461-0248.2004.00608.x

Lindstrom, E.S. & Langenheder, S. (2012) Local and regional factors influencing bacterial community assembly. *Environ. Microbiol. Rep.* **4** , 1–9, doi: 10.1111/J.1758-2229.2011.00257.X

Martinez-Sanz, C., Cenzano, C.S.S., Fernandez-Alaez, M. & Garcia-Criado, F. (2012) Relative contribution of small mountain ponds to regional richness of littoral macroinvertebrates and the implications for conservation. *Aquat. Conserv. Mar. Freshw. Ecosyst.* **22** , 155–164, doi: 10.1002/aqc.2227.

Marton, Z., Szabo, B., Vad, C.F., Palfy, K. & Horvath, Z. (2023) Environmental changes associated with drying climate are expected to affect functional groups of pro- and microeukaryotes differently in temporary saline waters. *Sci. Rep.* **13** , 3243, doi: 10.1038/s41598-023-30385-6.

McCauley, S.J. (2010) Body size and social dominance influence breeding dispersal in male *Pachydiplax longipennis* (Odonata). *Ecol. Entomol.* **35** , 377–385, doi: 10.1111/j.1365-2311.2010.01191.x.

McMurdie, P.J. & Holmes, S. (2013) phyloseq: An R package for reproducible interactive analysis and graphics of microbiome census data. *PLoS One* **8** , e61217, doi: doi.org/10.1371/journal.pone.0061217.

Mony, C., Uroy, L., Khalfallah, F., Haddad, N. & Vandenkoornhuyse, P. (2022) Landscape connectivity for the invisibles. *Ecography*. **2022** , doi: 10.1111/ecog.06041.

Mony, C., Vandenkoornhuyse, P., Bohannan, B.J.M., Peay, K. & Leibold, M.A. (2020) A Landscape of Opportunities for Microbial Ecology Research. *Front. Microbiol.* **11** , 2964, doi: 10.3389/fmicb.2020.561427

Nychka, D., Furrer, R., Paige, J. & Sain, S. (2021) fields: Tools for spatial data.

Oertli, B., Joye, D.A., Castella, E., Juge, R., Cambin, D. & Lachavanne, J.-B. (2002) Does size matter? The relationship between pond area and biodiversity. *Biol. Conserv.* **104** , 59–70, doi: 10.1016/S0006-3207(01)00154-9.

Oksanen, J., Blanchet, F.G., Friendly, M., Kindt, R., Legendre, P., McGlinn, D., Minchin, P.R., O’Hara, R.B., Simpson, G.L., Solymos, P., Stevens, M.H.H., Szoecs, E. & Wagner, H. (2020) vegan: Community Ecology Package.

Padial, A.A., Ceschin, F., Declerck, S.A.J., Meester, D. & Bonecker, L. (2014) Dispersal Ability Determines the Role of Environmental, Spatial and Temporal Drivers of Metacommunity Structure. *PLoS One* **9** , 111227, doi: 10.1371/journal.pone.0111227.

- Parada, A.E., Needham, D.M. & Fuhrman, J.A. (2016) Every base matters: assessing small subunit rRNA primers for marine microbiomes with mock communities, time series and global field samples. *Environ. Microbiol.* **18** , 1403–1414, doi: 10.1111/1462-2920.13023.
- Pereira, M., Segurado, P. & Neves, N. (2011) Using spatial network structure in landscape management and planning: A case study with pond turtles. *Landsc. Urban Plan.* **100** , 67–76, doi: 10.1016/j.landurbplan.2010.11.009
- Quast, C., Pruesse, E., Yilmaz, P., Gerken, J., Schweer, T., Yarza, P., Peplies, J. & Glockner, F.O. (2012) The SILVA ribosomal RNA gene database project: improved data processing and web-based tools. *Nucleic Acids Res.* **41** , D590–D596, doi: 10.1093/nar/gks1219
- QGIS Development Team (2022). QGIS Geographic Information System (v.3.26.0). Open Source Geospatial Foundation Project. <http://qgis.osgeo.org>
- R Core Team (2021) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.
- Ray, J.L., Althammer, J., Skaar, K.S., Simonelli, P., Larsen, A., Stoecker, D., Sazhin, A., Ijaz, U.Z., Quince, C., Nejstgaard, J.C., Frischer, M., Pohnert, G. & Troedsson, C. (2016) Metabarcoding and metabolome analyses of copepod grazing reveal feeding preference and linkage to metabolite classes in dynamic microbial plankton communities. *Mol. Ecol.* **25** , 5585–5602, doi: 10.1111/mec.13844.
- Rohwer, R.R., Hamilton, J.J., Newton, R.J. & McMahon, K.D. (2018) TaxAss: Leveraging a Custom Freshwater Database Achieves Fine-Scale Taxonomic Resolution. *mSphere* **3** , e00327–18, doi: 10.1128/mSphere.00327-18.
- Schloss, P.D., Westcott, S.L., Ryabin, T., Hall, J.R., Hartmann, M., Hollister, E.B., Lesniewski, R.A., Oakley, B.B., Parks, D.H., Robinson, C.J., Sahl, J.W., Stres, B., Thallinger, G.G., Van Horn, D.J. & Weber, C.F. (2009) Introducing mothur: Open-Source, Platform-Independent, Community-Supported Software for Describing and Comparing Microbial Communities. *Appl. Environ. Microbiol.* **75** , 7537–7541, doi: 10.1128/AEM.01541-09.
- Smith, D.J., Timonen, H.J., Jaffe, D.A., Griffin, D.W., Birmele, M.N., Perry, K.D., Ward, P.D. & Roberts, M.S. (2013) Intercontinental dispersal of bacteria and archaea by transpacific winds. *Appl. Environ. Microbiol.* **79** , 1134–1139, doi: 10.1128/AEM.03029-12/SUPPL_FILE/ZAM999104100SO1.PDF
- Smith, M.A. & Green, M.D. (2005) Dispersal and the metapopulation paradigm in amphibian ecology and conservation: are all amphibian populations metapopulations? *Ecography*. **28** , 110–128, doi: 10.1111/j.0906-7590.2005.04042.x.
- Soininen, J., Kokocinski, M., Estlander, S., Kotanen, J. & Heino, J. (2007) Neutrality, niches, and determinants of plankton metacommunity structure across boreal wetland ponds. *Ecoscience* **14** , 146–154, doi: 10.2980/1195-6860(2007)14[146:NNADOP]2.0.CO;2
- Szabo, B., Szabo, A., Vad, C.F., Boros, E., Lukić, D., Ptacnik, R., Márton, Z. & Horváth, Z. (2022) Microbial stowaways: Waterbirds as dispersal vectors of aquatic pro- and microeukaryotic communities. *J. Biogeogr.* **49** , 1286–1298, doi: 10.1111/JBI.14381
- Taylor, P.D., Fahrig, L., Henein, K. & Merriam, G. (1999) Connectivity is a vital element of landscape structure. *NCASI Tech. Bull.* **2** , 534, doi: 10.2307/3544927 NCASI.
- Thompson, P.L., Guzman, L.M., De Meester, L., Horváth, Z., Ptacnik, R., Vanschoenwinkel, B., Viana, D.S. & Chase, J.M. (2020) A process-based metacommunity framework linking local and regional scale community ecology. *Ecol. Lett.* **23** , 1314–1329, doi: 10.1111/ele.13568
- Thornhill, I., Batty, L., Hewitt, M., Friberg, N.R. & Ledger, M.E. (2018) The application of graph theory and percolation analysis for assessing change in the spatial configuration of pond networks. *Urban Ecosyst*

21 , 213–225, doi: 10.1007/s11252-017-0724-8.

Urban, D. & Keitt, T. (2001) Landscape connectivity: A graph-theoretic perspective. *Ecology* **82** , 1205–1218, doi: 10.1890/0012-9658(2001)082[1205:LCAGTP]2.0.CO;2

Vad, C.F., Pentek, A.L., Cozma, N.J., Foldi, A., Toth, A., Toth, B., Bode, N.A., Mora, A., Ptacnik, R., Acs, E., Zsuga, K. & Horvath, Z. (2017) Wartime scars or reservoirs of biodiversity? The value of bomb crater ponds in aquatic conservation. *Biol. Conserv.***209** , 253–262, doi: 10.1016/j.biocon.2017.02.025

Van De Meutter, F., Stoks, R. & De Meester, L. (2008) Size-selective dispersal of *Daphnia* resting eggs by backswimmers (*Notonecta maculata*). *Biol. Lett.* **4** , 494–496, doi: 10.1098/RSBL.2008.0323 The Royal Society London.

Van Der Gucht, K., Cottenie, K., Muylaert, K., Vloemans, N., Cousin, S., Declerck, S., Jeppesen, E., Conde-Porcuna, J.M., Schwenk, K., Zwart, G., Degans, H., Vyverman, W. & De Meester, L. (2007) The power of species sorting: Local factors drive bacterial community composition over a wide range of spatial scales. *Proc. Natl. Acad. Sci. U. S. A.***104** , 20404–20409, doi: 10.1073/pnas.0707200104.

Vanormelingen, P., Cottenie, K., Michels, E., Muylaert, K., Vyverman, W. & De Meester, L. (2008) The relative importance of dispersal and local processes in structuring phytoplankton communities in a set of highly interconnected ponds. *Freshw. Biol.* **53** , 2170–2183, doi: 10.1111/J.1365-2427.2008.02040.X

Vanschoenwinkel, B., De Vries, C., Seaman, M. & Brendonck, L. (2007) The role of metacommunity processes in shaping invertebrate rock pool communities along a dispersal gradient. *Oikos* **116** , 1255–1266, doi: 10.1111/J.0030-1299.2007.15860.X

Verreydt, D., De Meester, L., Decaestecker, E., Villena, M.-J., Van Der Gucht, K., Vannormelingen, P., Vyverman, W. & Declerck, S.A.J. (2012) Dispersal-mediated trophic interactions can generate apparent patterns of dispersal limitation in aquatic metacommunities. *Ecol. Lett.***15** , 218–226, doi: 10.1111/j.1461-0248.2011.01728.x.

Vyverman, W., Verleyen, E., Sabbe, K., Vanhoutte, K., Sterken, M., Hodgson, D.A., Mann, D.G., Juggins, S., Vijver, B. Van de, Jones, V., Flower, R., Roberts, D., Chepurnov, V.A., Kilroy, C., Vanormelingen, P. & Wever, A. De (2007) Historical processes constrain patterns in global diatom diversity. *Ecology* **88** , 1924–1931, doi: 10.1890/06-1564.

Wickham, H. (2016) ggplot2: Elegant Graphics for Data Analysis. Springer-Verlag New York.

Wickham, H. (2007) Reshaping Data with the reshape Package. *J. Stat. Softw.* **21** (12), 1–20.

Wisnoski, N.I., Leibold, M.A. & Lennon, J.T. (2019) Dormancy in Metacommunities. *Am. Nat.* **194** , 135–151, doi: 10.1086/704168.

Wisnoski, N.I. & Shoemaker, L.G. (2022) Seed banks alter metacommunity diversity: The interactive effects of competition, dispersal and dormancy. *Ecol. Lett.* **25** , 740–753, doi: 10.1111/ele.13944.