

Ecography

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Supplementary material

Appendix 1

1. Sample collection and analysis

Each pond was sampled once. The duration of the sampling campaign was 43 days (i.e., from May to July of 2013). To avoid interference from an effect of sampling time and any directional change in environmental condition associated with it, the order of sampling along the urbanization gradient was randomized over the different plot levels. Each day three ponds of an individual regional plot, and thus representing three different urbanization classes within a given plot, were sampled. Across days we randomized the plots to be sampled so that there was no bias with respect to region or urbanization level. This randomized sampling design proved effective, as we performed MANOVA analysis using sampling time as explanatory variable and detected no temporal effect on species composition ($p = 0.84$) or on the selected urbanization and environmental variables ($p > 0.5$ for all variables). At each sampling occasion, we measured pH and conductivity with standard electrodes. Water transparency was determined with a Snellers tube due to the characteristically shallow depths of the ponds. Depth-integrated water samples were collected in an open water zone of the pond using a tube sampler (length 1.5 m; diameter 75 mm) and kept in the cold (4 °C) and dark until further analysis. To ensure correct sampling of the water layer, contact with vegetation or the sediment was carefully avoided during sampling. We also estimated the percentage of cover of four different vegetation variables, defined as: (1) submerse, (2) floating, (3) emergent vegetation, and (4) total vegetation cover. Water depth was measured with a graduated stick in the central part of the pond, as well as at 10cm of the edge of the pond. The depth-integrated water samples were used for the assessment of the concentration of chlorophyll a, nutrients (total phosphorus and nitrates, and available phosphorus and nitrates), alkalinity and some major ions (calcium, chloride and sulphate ions, water

hardness). Sulphates, chlorides, calcium, alkalinity and hardness were measured following standard methods according to the Hach Water Analysis Handbook (HACH, 1992). Nitrate concentration was determined after filtration on a GF/F filter with a Technicon autoanalyser III. Total phosphorus concentration was measured with the ascorbic acid method after perchlorate digestion (Murphy and Riley, 1962). In situ Chlorophyll a concentrations were spectrophotometrically determined with a portable fluorometer.

2. Trait information

We extracted information on body size and plant association from literature. Body size values are according to Alonso (1996). Values of the trait plant association were determined mainly based on Barnett et al. (2007) and complemented with information provided by Declerck et al. (2007) and Alonso (1996). Body size values were complemented with our own measurements, when available. The trait filtration type was mainly based on Barnett et al. (2007). However, we further differentiated *Scapholeberis spp.* and *Simocephalus spp.* from the rest because those species do not belong to any of the categories presented in the literature. Trait values are given in Table A1.

Table A1: List of species recorded in our study and their respective trait values. Acronyms supplement Fig. 4 in the main text.

Species	Body size (mm)	Plant association	Filtration type	Acronym
<i>Alonella excisa</i>	0.35	1 (littoral)	1 (C-Filtration)	A.exc
<i>Alonella exigua</i>	0.4	1 (littoral)	1 (C-Filtration)	A.exi
<i>Alona guttata</i>	0.4	1 (littoral)	1 (C-Filtration)	A.gu
<i>Alona rectangula</i>	0.5	1 (littoral)	1 (C-Filtration)	A.re

<i>Bosmina</i>	0.6	3 (pelagic)	2 (B-Filtration)	B.lo
<i>longirostris</i>				
<i>Ceriodaphnia</i>	0.9	2 (intermediate)	3 (D-Filtration)	C.qu
<i>quadrangula</i>				
<i>Ceriodaphnia</i>	0.9	2 (intermediate)	3 (D-Filtration)	C.la
<i>laticaudata</i>				
<i>Ceriodaphnia</i>	0.8	2 (intermediate)	3 (D-Filtration)	C.pu
<i>pulchella</i>				
<i>Ceriodaphnia</i>	1.3	2 (intermediate)	3 (D-Filtration)	C.re
<i>reticulata</i>				
<i>Chydorus</i>	0.55	1 (littoral)	1 (C-Filtration)	C.sp
<i>sphaericus</i>				
<i>Daphnia</i>	2.5	3 (pelagic)	3 (D-Filtration)	D.cu
<i>curvirostris</i>				
<i>Daphnia magna</i>	4.0	3 (pelagic)	3 (D-Filtration)	D.ma
<i>Daphnia obtusa</i>	2.5	3 (pelagic)	3 (D-Filtration)	D.ob
<i>Daphnia pulex</i>	2.5	3 (pelagic)	3 (D-Filtration)	D.pu
<i>Graptoleberis</i>	0.6	1 (littoral)	1 (C-Filtration)	G.te
<i>testudinaria</i>				
<i>Leydigia</i>	0.8	1 (littoral)	1 (C-Filtration)	L.ac
<i>acanthocercoides</i>				
<i>Pleuroxus aduncus</i>	0.65	1 (littoral)	1 (C-Filtration)	P.ad
<i>Pleuroxus</i>	0.6	1 (littoral)	1 (C-Filtration)	P.de
<i>denticulatus</i>				
<i>Pleuroxus</i>	0.65	1 (littoral)	1 (C-Filtration)	P.tr

<i>truncatus</i>				
<i>Scapholeberis</i>	1.2	3 (pelagic)	4 (Scaphol. type)	S.mu
<i>mucronata</i>				
<i>Scapholeberis</i>	1.6	3 (pelagic)	4 (Scaphol. type)	S.ki
<i>kingi</i>				
<i>Simocephalus</i>	2.8	1 (littoral)	5 (Simoc. type)	S.ex
<i>exspinosus</i>				
<i>Simocephalus</i>	2.7	1 (littoral)	5 (Simoc. type)	S.ve
<i>vetulus</i>				

3. Selection of orthogonal eigenvectors describing β -diversity patterns

For β -diversity, the pair-wise output values obtained from Bray-Curtis and COMDIST (see Material & Methods) were synthesized into Principal Coordinate Analysis (PCoA) eigenvectors after Lingoes correction (Borcard et al. 2011; Swenson 2014). In principle, all eigenvectors generated by PCoA could be used as descriptors of β -diversity patterns in subsequent analysis. However, using all of them can introduce confounding effects in the analyses (Anderson & Willis 2003). Since each eigenvector is an orthogonal synthetic variable representing gradients in β -diversity patterns, it is possible that some of these gradients are unexplained by the measured factors, which might introduce confounding effects in posterior analyses. A solution is to select a subset of orthogonal eigenvectors that maximizes the association between patterns of β -diversity (taxonomic or functional-phylogenetic) and a set of explanatory variables (Anderson & Willis 2003). To select how many orthogonal eigenvectors should be retained for subsequent analysis, we applied an approach that is suitable for direct multiple regression analyses (Anderson & Willis 2003) [see also (Duarte et al. 2012)]. The selection procedure consisted of first including a single

eigenvector (i.e., the first eigenvector, which captures most of the variation in the original distance matrix) that describes β -diversity patterns as our response variable into the variation partitioning approach. Then, we computed the total $\text{adj}R^2_{(Y|X)}$ obtained for this combination of this single eigenvector (the first) as response variable and the selected environmental and spatial descriptors as explanatory variables. Next, we included the first two orthogonal eigenvectors as response variables and repeated the procedure, computing again the $\text{adj}R^2_{(Y|X)}$ for this combination of the first two eigenvectors and the predictor variables. This incremental approach was applied consecutively by including an increasing number of orthogonal eigenvectors (i.e., 1,2,3,4 and so forth), until we obtained a complete distribution of $\text{adj}R^2_{(Y|X)}$ values for each number of eigenvectors (response variables) included. Finally, we retained as many eigenvectors as required to maximize $\text{adj}R^2_{(Y|X)}$, which is the exact number that represents the best fit between the response and the explanatory matrices. In other words, when including less than that specific number of eigenvectors this results in a lower $\text{adj}R^2_{(Y|X)}$ because it captures too low variation in the original response matrix. In contrast, including more than that specific number decreases the $\text{adj}R^2_{(Y|X)}$ by adding residuals associated to redundant, meaningless variables [for further details on this selection procedure see also Anderson and Willis (2003) and Duarte et al. (2012)].

4. Results of a taxonomic approach based on Bray-Curtis

Using a taxonomic index of beta-diversity (i.e., Bray-Curtis) followed by PCoA analysis, resulted in a moderate increase of explanatory power provided by environmental and spatial factors compared to a taxonomic approach based only on species abundances after Hellinger transformation. However, the conclusions that can be drawn from both taxonomic-based approaches are largely the same. Specifically, the amount of explanatory power provided by environmental, urbanization and spatial factors are much higher for trait and phylogenetic-

based approaches than for any of the taxonomic-based approaches used (Fig. A1). Variation partitioning revealed that the relative importance of local environmental factors was much more important than that of spatial factors (Fig. A1). Finally, using the taxonomic approach based on Bray-Curtis resulted in a lack of explanatory power provided by urbanization (i.e., no urbanization variable was selected through the forward selection procedure approach).

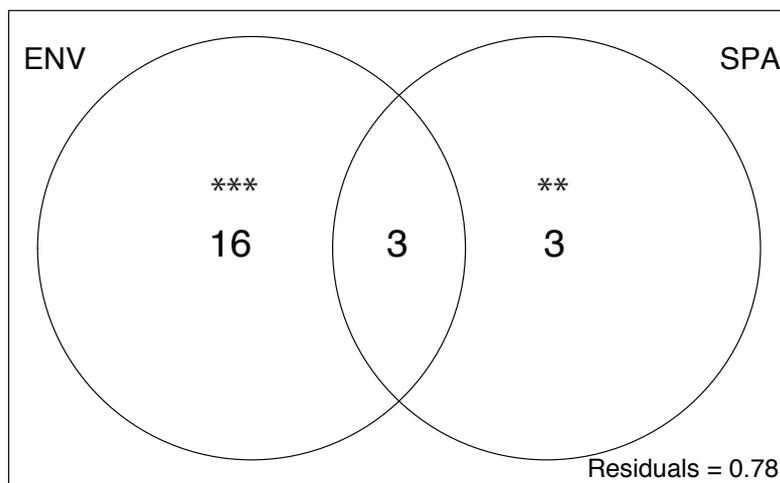


Figure A1: Venn diagram depicting the results of variation partitioning (i.e., explained variation; % $\text{adj}R^2$) for metacommunity analysis based on Bray-Curtis approach. Env = environmental model; Spa = spatial model. Three asterisks represent significant results $p < 0.001$; two asterisks $p < 0.01$.

5. Correlation between body size, phylogeny and the urbanization gradient

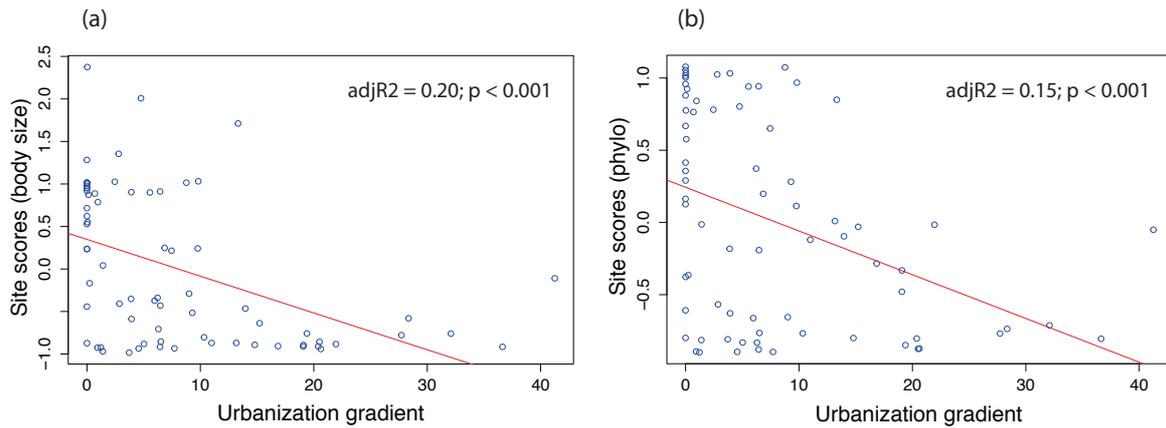


Figure A2: Correlation between urbanization (percentage built-up area at 50 meters) and body size composition within ponds (a) and between urbanization and phylogenetic composition within ponds (b). On the Y-axis site scores obtained from the first axis of PCoA based on COMDIST for body size (a) and phylogenetic distances (b) are given. (a) More positive scores are associated with communities dominated by larger species whereas negative scores are associated with communities dominated by smaller species. (b) More positive scores are associated with communities dominated by Daphniidae species whereas more negative scores are associated with communities dominated by Chydoriidae species.

6. Analysis of residuals

We observed a large amount of unexplained variance when using a taxonomic-based approach (i.e., traditional metacommunity analysis based on species abundances after hellinger transformation). This large fraction of unexplained variation could be due to idiosyncratic responses of species with similar traits. This can happen for example, when different species with similar traits respond similarly to the same type of environmental, urbanization or spatial factors. To test this idea, we first extracted the residuals of the best performing RDA model using environmental, spatial and urban-related variables on species composition. Then, we ran another variation partitioning analysis as described in the main text, but this time using the residuals as response variables and trait (both body size alone or

several traits) and phylogenetic information as explanatory variables. We found that trait and phylogenetic information significantly explained variation in the residuals of the taxonomic analysis (Fig. A3). This indicates that using trait and phylogenetic information helps to explain the large amount of unexplained variation that resulted from taxonomic-based approaches. This suggests that closely related species, or species that share similar traits, respond in the same way to environmental, spatial and urbanization gradients, but such species responses cannot be easily predicted by species (names) identities alone.

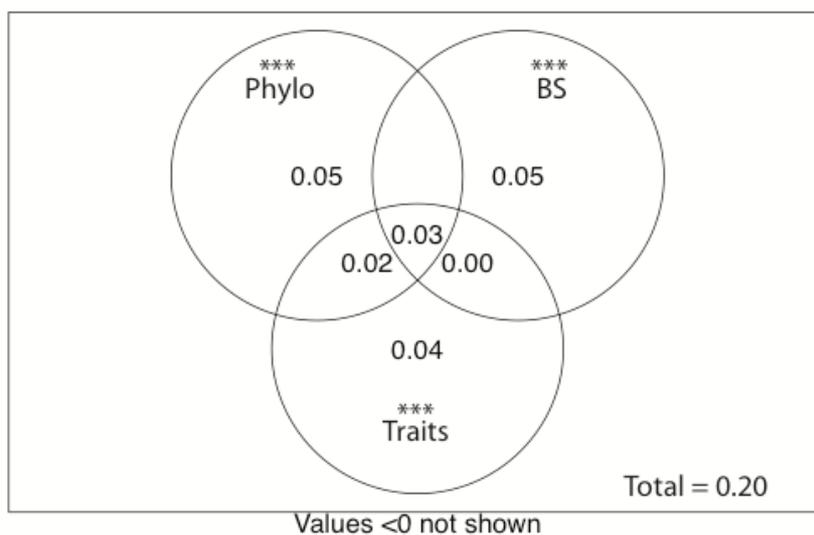


Figure A3. Venn diagrams showing results of variation partitioning using as response variable the residuals of RDA based on selected environmental, spatial and urban-related variables on species composition and as predictors phylogeny (Phylo), body size (BS) and multi-trait metrics (Traits). Three asterisks indicate significant results ($p < 0.001$).

6. References in the Appendix.

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