**RESEARCH PAPER**

# Scale decisions can reverse conclusions on community assembly processes

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## ABSTRACT

**Aim** Phylogenetic diversity patterns are increasingly being used to better understand the role of ecological and evolutionary processes in community assembly. Here, we quantify how these patterns are influenced by scale choices in terms of spatial and environmental extent and organismic scales.

**Location** European Alps.

**Methods** We applied 42 sampling strategies differing in their combination of focal scales. For each resulting sub-dataset, we estimated the phylogenetic diversity of the species pools, phylogenetic α-diversities of local communities, and statistics commonly used together with null models in order to infer non-random diversity patterns (i.e. phylogenetic clustering versus over-dispersion). Finally, we studied the effects of scale choices on these measures using regression analyses.

**Results** Scale choices were decisive for revealing signals in diversity patterns. Notably, changes in focal scales sometimes reversed a pattern of over-dispersion into clustering. Organismic scale had a stronger effect than spatial and environmental extent. However, we did not find general rules for the direction of change from over-dispersion to clustering with changing scales. Importantly, these scale issues had only a weak influence when focusing on regional diversity patterns that change along abiotic gradients.

**Main conclusions** Our results call for caution when combining phylogenetic data with distributional data to study how and why communities differ from random expectations of phylogenetic relatedness. These analyses seem to be robust when the focus is on relating community diversity patterns to variation in habitat conditions, such as abiotic gradients. However, if the focus is on identifying relevant assembly rules for local communities, the uncertainty arising from a certain scale choice can be immense. In the latter case, it becomes necessary to test whether emerging patterns are robust to alternative scale choices.

**Keywords**

Apha diversity, assembly rules, community ecology, ecophylogenetics, null models, sampling design.

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## INTRODUCTION

A key objective of ecology is to better understand the structure and diversity of species communities and the ecological and evolutionary processes driving community assembly (Diamond, 1975; Leibold *et al.*, 2004). In the last decade there has been a trend towards studying these questions with increasingly large community datasets spanning large spatial extents, combined with measures of functional or phylogenetic diversity (Mouquet *et al.*, 2012). However, interpreting patterns of diversity is a

**Table 1** Criteria used to constrain the species pool according to different scale choices.

| | | Data description | Ecological reasoning | Considered levels |
|---|---|---|---|---|
| Organismic scale | Phylogenetic constraint | In total we observed 3081 species, belonging to 773 genera and 135 families | Niche overlap is often related to phylogenetic distance, so that closely related species may interact more strongly | Six levels: All species versus only lowest stratum versus only Asteraceae versus only Asteraceae in lowest stratum versus only herbaceous species versus only herbaceous in lowest stratum |
| | Growth form constraint | Species were classified as either herbaceous or non-herbaceous (shrubby or woody) | Growth form may influence resource use. Thus, species of the same growth form may interact more strongly | |
| | Vegetation stratum constraint | Organisms were classified as either belonging to the lowest stratum (grass and herb stratum) or to higher strata (bush and tree strata) | Life stage influences resource use. Tree seedlings may show stronger symmetric niche-based competition with herbaceous plants than fully grown trees | |
| Spatial and environmental scale | Environmental extent | Community plots were located in polygons of homogeneous land-cover type | Species composition in different land-cover types differs strongly. Merging these communities in one dataset strongly increases the diversity of the species pool | Seven levels (three levels per focal land-cover type polygon): All data versus grasslands versus bare-rock vegetation versus sparsely vegetated areas versus focal grassland polygon versus focal bare-rock polygon versus focal sparsely vegetated polygon |
| | Spatial extent | Varied from the size of one polygon to the entire study region (see Fig. 1) | Species distributions can be limited by dispersal. Merging distant communities increases the species pool even if environmental conditions are kept constant | |

For each of the three focal land cover-type polygons, a total of 18 different species pools resulted from combining the three different choices of spatial and environmental extents with the six different choices of organismic scales. For all three focal land-cover types together, we obtained 42 different species pools because the species pools with the largest spatial and environmental extent were the same for all three land-cover types.

challenging endeavour due to the multitude of driving forces involved, such as environmental filtering, biotic interactions, dispersal, historical legacies and niche evolution (Leibold *et al.*, 2010; Mayfield & Levine, 2010; Peres-Neto *et al.*, 2012). The same process may cause opposing patterns of diversity due to the interplay with other processes, and the same pattern may result from different processes. For example, high phylogenetic diversity (over-dispersion) can result from limiting similarity of competing species if niche evolution created a strong phylogenetic signal (Blomberg & Garland, 2002). However, it can also result from environmental filtering if niches evolved under convergent evolution (Webb *et al.*, 2002). Additionally, as the underlying processes of assembly can act at different scales, a strong debate arises about the scale dependence of community diversity patterns and the ecological assumptions that underlie different scale choices (Vamosi *et al.*, 2009; Thuiller *et al.*, 2010; see also Appendix S1 in Supporting Information).

Two main considerations of scale choices can influence the outcome of community assembly studies (Magurran, 2004). (1) At what spatial extent (size of the study area) and environmental extent (range of environmental conditions) should communities be compared? (2) At which organismic scales should communities be delimited (e.g. which taxonomic groups and growth forms should be included)? Both decisions affect the estimated set of species ('species pool' in the following) that are potentially able to assemble into local communities but may be filtered out by ecological processes such as environmental filtering or competition. The species pool influences the tests on whether the observed phylogenetic diversities in local communities (hereafter called α-diversity) are significantly higher or lower than expected by chance. This is because these tests rely on the comparison of observed α-diversity values with diversity values of randomly assembled communities from the estimated species pool. In practice the species pool is often estimated by the full set of species in a dataset, even though recent studies suggest that a refinement of the species pool can help to disentangle interlinked patterns of different assembly processes (Hardy, 2008; De Bello *et al.*, 2012; Lessard *et al.*, 2012). The different aspects of scale choice influence the analysis of diversity patterns by affecting both the observed values of α-diversity and the null models via the composition and size of the species pool. This is especially critical, as scale decisions are often not primarily driven by the specific research question but also by more arbitrary aspects such as data availability or an a priori focus on certain species groups or study regions. Different important scale aspects are discussed below in more detail (see also Table 1 and Appendix S1).

**1.** The spatial extent is a critical aspect when deciding which community plots should be compared with each other (Cavender-Bares *et al.*, 2006; Thuiller *et al.*, 2010). Increasing the spatial extent is likely to increase the size of the species pool and the relative influence of historical factors (e.g. dispersal limitation; Svenning *et al.*, 2010). However, if the spatial extent

is too small then local processes (such as biotic interactions) may remove species from the entire species pool which will hinder the detection of a signal of these local processes (Pärtel *et al.*, 2011). Another important aspect that is often, but not necessarily, related to the spatial extent is the regional heterogeneity of environmental conditions, namely the environmental extent (Willis *et al.*, 2010). The greater the number of different climatic and habitat conditions considered in the study, the more dissimilar are the species in the pool. This is likely to produce overwhelming signals of environmental filtering in local communities (low α-diversity) even if different assembly processes act in concert (Willis *et al.*, 2010).

2. The choice of a certain organismic scale, i.e. the set of considered organisms, also has an important influence on the detection of non-random diversity patterns. Several studies focused on a subset of organisms that was relatively small compared to the overall pool of potentially interacting organisms. Among other criteria, these subsets can be defined by phylogenetic relatedness (e.g. hummingbirds; Graham *et al.*, 2009), growth form (e.g. trees; Whitfeld *et al.*, 2012) or size classes (e.g. plants in the forest understorey; Gilbert & Lechowicz, 2004). A general argument in this context is that reduced organismic scales amplify the detection of high diversity patterns if both environmental filtering and limiting similarity processes act together (Vamosi *et al.*, 2009; Thuiller *et al.*, 2010). The underlying assumption of this argument is that the selected subset of organisms shares more similar niches than the larger pool.

Community assembly studies that have directly compared different spatial, environmental or organismic scales have focused on only one or two specific scaling criteria (e.g. phylogenetic extent and spatial resolution) and/or have compared only two distinct scales (Appendix S1). Overall, the results of these studies are either mixed or tend to be in agreement with the above arguments that over-dispersion should be more prevalent at finer organismic scales and clustering should be more prevalent at larger scales (Appendix S1). However, interpretations of these findings can be very different and range from a suggested shift in the niche evolution process (Cavender-Bares *et al.*, 2006) to scale-dependent demographic processes (Vamosi *et al.*, 2009). A recent simulation study gives additional evidence that the interplay between scaling and identified diversity patterns is not as simple as assumed (Chalmandrier *et al.*, accepted). However, neither the relative importance of different scaling aspects nor the strength and the direction of potential biases have been studied systematically. In addition, the biases produced by different scale choices may have varying importance for different types of analyses in community assembly studies, a point that has not been recognized so far but is highly important for applied work.

Here we systematically test the effect of varying spatial and environmental extents and organismic scales on the local diversity patterns. By using a large-scale phytosociological dataset (18,919 community plots) encompassing a large biogeographic region (the European Alps), a wide range of environmental conditions (from mediterranean type to the temperate alpine climates), and a high diversity of vascular species (3081 species

belonging to 773 genera and 135 families), we aim at answering the following questions:

1. How strong is the influence of scale choices when comparing observed α-diversity with null model expectations?
2. Which scale choices influence the emerging signal of community structure most? Do any general trends exist?
3. For which research questions is the influence of scale choices most relevant?

## METHODS

### Study area, phytosociological dataset and land-cover classification

The Alpine Convention delimits the study area, which covers about 258,000 km$^2$ and a wide range of climatic conditions and land-cover types (Fig. 1). We compiled a comprehensive dataset of homogeneous community plots by merging the Alps Vegetation Database (AVD; Lenoir *et al.*, 2012), consisting of several national and regional databases, and the French National Alpine Botanical Conservatory database (CBNA). To ensure consistency between the two databases and improve data quality we included only those community plots that passed a number of filters (see Appendix S2 for more details). To guarantee that the community plots covered homogeneous and representative vegetation, all plots were chosen and surveyed by professional botanists. The land-cover classification for each of the selected community plots was based on the CORINE Land Cover database and considered 11 land-cover types (Table 2; see Appendix S2 for more details).

### Different resampling strategies for different scale choices

To compare the effects of different scale choices on α-diversities and identified signals of clustering versus over-dispersion among certain sets of community plots, we selected a single homogeneous and continuous land-cover type polygon for each of the three most frequent land-cover types (apart from forest). In the following we call these three polygons 'focal polygons' and the community plots within the focal polygons 'focal community plots' (Fig. 2). For each land-cover type we chose the polygon with the largest sampling effort (Table 2, Fig. 1). The three focal polygons consisted of: 304 community plots within a 85,254 ha grassland polygon, 97 community plots within a 4748 ha sparsely vegetated polygon and 226 community plots within a bare rock polygon of 3770 ha. The size of the community plots within these three polygons varied with the land-cover type. They ranged from 40 to 314 m$^2$ within grasslands (mean of 203 m$^2$), from < 1 to 90 m$^2$ within bare rocks (mean of 46 m$^2$) and from 1 to 250 m$^2$ within the sparsely vegetated land-cover type (mean of 32 m$^2$). Note that we did not include a focal forest polygon to avoid mixing two different vegetation types and to facilitate the comparison of sampling design choices. Forests are also highly managed and planted in the Alps and information on management was not available for our community-plots.
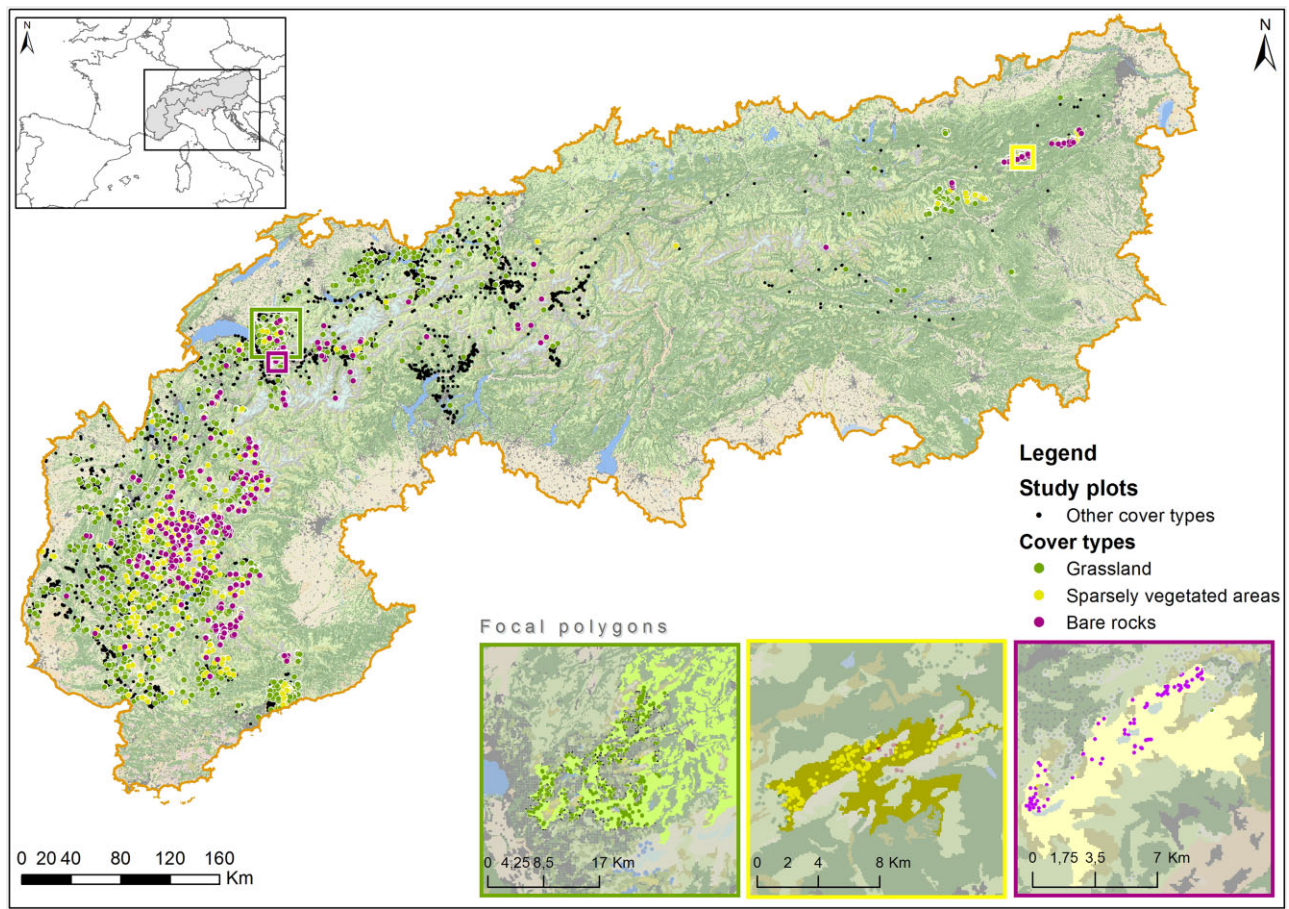
**Figure 1** Map of the study area with the three focal land-cover types, the three focal polygons and their focal community plots.

**Table 2** Description of the land-cover type classification: For each land-cover type the number of observed community plots, the number of polygons, their respective average area, their range of areas and their total area in the Alps (added over all polygons of the same type) are listed. Land-cover types are based on CORINE, which is a European map of the environmental landscape based on interpretation of satellite images. The CORINE code used here represents the original CORINE code used for the reclassification of land-cover types (see Appendix S2 for details).

| Land-cover type | No. of community plots | No. of polygons | Mean polygon area (km²) | Range of polygon area (km²) | Total land-cover area (km²) | CORINE code |
|---|---|---|---|---|---|---|
| Built-up | 446 | 19,055 | 0.66 | < 1–254 | 12,621 | 11–14 |
| Arable | 952 | 13,722 | 3.02 | < 1–7457 | 41,455 | 21 |
| Permanent crops | 75 | 3254 | 1.31 | < 1–241 | 4248 | 22 |
| Grassland | 5423 | 50,383 | 1.25 | < 1–3562 | 62,858 | 32 |
| Forest | 8530 | 33,915 | 3.05 | < 1–24,370 | 103,396 | 31 |
| Sclerophyllous vegetation | 409 | 872 | 1.74 | < 1–107 | 1518 | 32 |
| Sparsely vegetated areas | 1444 | 14,815 | 0.76 | < 1–83 | 11,216 | 33 |
| Bare rocks | 1462 | 4341 | 3.15 | < 1–605 | 13,687 | 33 |
| Glaciers and perpetual snow | 25 | 910 | 2.62 | < 1–367 | 2381 | 33 |
| Inland marshes | 93 | 777 | 0.72 | < 1–102 | 562 | 41 |
| Water bodies | 57 | 3639 | 0.99 | < 1–581 | 3593 | 51 |
| Unidentified | 4 | 29 | 5.47 | < 1–136 | 159 | 52 |

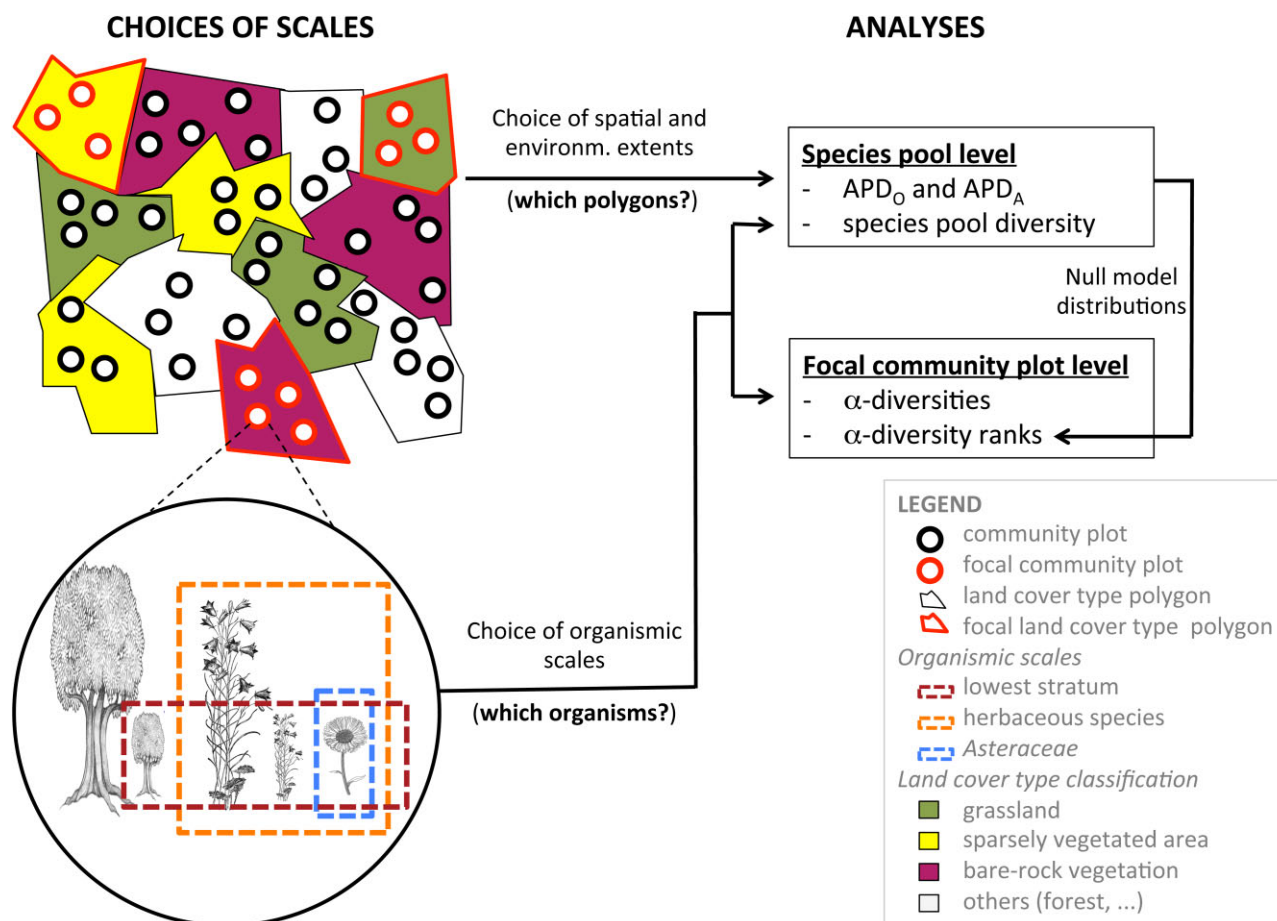**CHOICES OF SCALES**　　　　　　　　　　　**ANALYSES**



**Figure 2** Schematic overview of the scale choices and their influence on the composition of the species pools and the diversity of the focal community plots. For example, when the constraints include reducing the environmental extent to only grasslands and the organismic scale to only herbaceous species in the lowest stratum, then α-diversity is calculated by considering only herbaceous species from the lowest stratum in the focal grassland polygon (three community plots in the overview figure). In this case, α-diversity-percentiles are calculated based on a species pool that contains all community plots from the grassland polygons (12 community plots in three polygons in the overview figure) and only herbaceous species from the lowest stratum.

We used all community plots across the Alps to build the largest possible species pool (Fig. 1) that we further constrained with respect to the different scale choices (Table 1, Fig. 2). In a first step, we considered different choices of environmental and spatial extents. The constrained species pools could consist of community plots from (1) all land-cover type polygons, (2) only one type of land cover to reduce the environmental extent (e.g. grassland) or (3) the focal land-cover type polygon to reduce the spatial extent in addition to the environmental extent. In that way, choices of spatial and environmental extents, by influencing the species pool, did not influence observed α-diversities in the focal community plots but only the identified signals of clustering versus over-dispersion.

In a second step, we constrained all community plots by a choice of organismic scales (Fig. 2). The constrained communities consisted either of (1) all observed plant species, (2) only plant species in the lowest height stratum (herb and grass layer, called vegetation stratum constraint in the following), (3) all herbaceous species (called growth form constraint in the follow-

ing), (4) all herbaceous species in the lowest height stratum, (5) only Asteraceae (called phylogenetic constraint in the following), or (6) only Asteraceae in the lowest height stratum. We used Asteraceae for the phylogenetic constraint because it is one of the most ecologically labile families in the Alpine flora, and was the most commonly observed plant family in the three focal land-cover polygons. Species of this family were thus observed in most focal community plots. As a result we were able to estimate the phylogenetic diversity of most focal community plots under the phylogenetic constraint. Choices of organismic scales influenced both observed α-diversities and the identified signals of clustering versus over-dispersion. In total we obtained 18 different species pools for each of the three focal polygons by combining the three different choices of spatial and environmental extents with the six different choices of organismic scales, and thus 42 different species pools for all three focal land-cover types together (note that the species pool with the largest spatial and environmental extent was the same for all three land cover types; Table 1).

## Diversity indices and abundance phylogenetic divergence indices

We calculated taxonomic and phylogenetic $\alpha$-diversities with an equivalent number transformation of the quadratic entropy of Rao ($Q_c$). Rao's $\alpha$-diversity estimates the average dissimilarity between species pairs in a community and allows us to include species abundances and distances between species (for taxonomic diversity all distances are assumed to be equal):

$$Q_c = \frac{1}{1 - \sum_{i=1}^{s} \sum_{j=1}^{s} d_{ij} p_i p_j}$$

where $d_{ij}$ is the dissimilarity between each pair of species $i$ and $j$ and $p_i$, $p_j$ are the relative abundances of species $i$ and $j$. We used cophenetic distances based on branch length information from a dated genus-level phylogeny of the Alpine plants to calculate phylogenetic dissimilarities. Phylogenetic distances were rescaled to a range between zero and one. The phylogeny of the Alpine plants was constructed following the workflow proposed by Roquet *et al.* (2013) and thus contained absolute-age branch length information (see Appendix S2 for details on the phylogeny). The equivalent number transformation (Jost, 2007) was required to avoid violating the concept of equivalent numbers when decomposing diversity in $\alpha$, $\beta$ and $\gamma$ components. Rao's $\alpha$-diversities ($Q_{c\alpha}$) were calculated by considering species and relative abundances in the community plots. Species pool diversities ($\gamma$ component, $Q_{c\gamma}$) were calculated by considering species and their relative abundances in the species pools.

## Assessing statistical significance in community structure

To test for patterns of over-dispersion or clustering, we simulated 999 random communities for each community plot and each species pool, calculated the related random diversity values (null model distribution in the following) and reported the percentile of the observed $\alpha$-diversity in the null model distribution ($\alpha$-diversity-percentile in the following). Thus the $\alpha$-diversity-percentile is the test statistic which allows us to decide whether an observed pattern is different from random expectations: If the observed value was lower (respectively higher) than the 5th (respectively 95th) percentile of the distribution, the observed $\alpha$-diversity was significantly lower (respectively higher) than expected by chance. With this choice of a critical significance value we assume that significance tests are based on one-sided hypotheses. An example would be a null hypothesis of a random pattern or over-dispersion and an alternative hypothesis of a convergent pattern. Because non-random distributions of species frequencies of occurrence and/or abundances can cause a bias in test results, we tested for such non-random patterns with the abundance phylogenetic divergence index (Hardy, 2008) and, if necessary, accounted for this pattern in the randomization schemes (following the procedure suggested by Hardy, 2008; see Appendices S3 & S4 for more detail).

## Regression analyses

To investigate the influence of scale choices on the identification of assembly patterns, we applied a set of regression and partial-regression analyses:

### Accounting for between-community differences

As we were primarily interested in the changes of phylogenetic diversity patterns due to changes in scale choices we aimed at removing the effect of between-community variation. We first tested how much of the variation in $\alpha$-diversities and $\alpha$-diversity-percentiles depended on idiosyncrasies of the community plots, i.e. the physical sampling units. We regressed separately the observed $\alpha$-diversities and the $\alpha$-diversity-percentiles against community plot identities (i.e. a factor with as many values as there were communities in the focal polygon) and used the residuals of these regressions (called $\alpha$-diversity-residuals and percentile-residuals in the following) in the further analyses. In other words we calculated the deviance of the 18 different values (18 different species pools for each of the three focal polygons) for one diversity estimate (either $\alpha$-diversities or $\alpha$-diversity-percentiles) for one community from the mean of these values. These residuals only contain information on whether a scale change increases or decreases the focal diversity estimate; all information on whether the estimate was generally high or low in comparison to the other communities was removed.

### The influence of scale choices on species pool diversity and $\alpha$-diversity-residuals

To investigate which of the scale constraints exerted the highest influence on the species pool diversity (re-estimated for each applied scale choice and resulting species pool), we regressed the species pool diversity against organismic scale choices (i.e. against phylogenetic, growth form and vegetation stratum constraints, each represented by a factor with two levels; see Table 1 and Appendix S5) and against spatial and environmental extents (each represented by a factor with two levels, all polygons versus one polygon for spatial extent and all polygons versus polygons of only one land-cover type for environmental extent; see Table 1 and Appendix S5). We regressed $\alpha$-diversity-residuals against organismic scale choices, but not against spatial and environmental extents as these did not influence local observed diversity.

### The influence of scale choices on percentile-residuals

To identify which of the four components exerted the highest influence on the percentile-residuals, and thus on conclusions about community assembly, we first regressed in four separate univariate models and then in a common multivariate model the percentile-residuals against: (1) organismic scales, (2) spatial and environmental extents, (3) species pool diversity, and (4) $\alpha$-diversity-residuals.
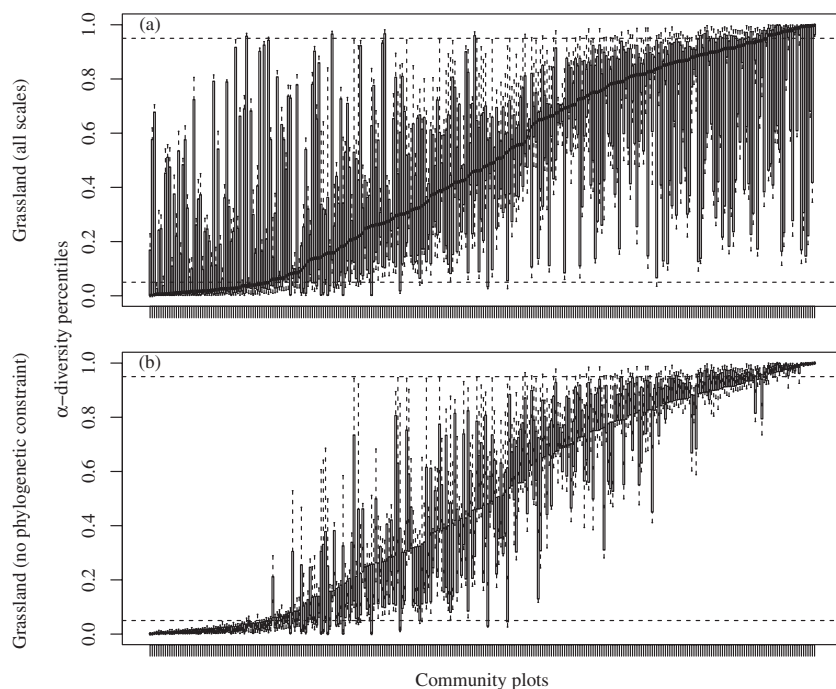
Figure 3 Distribution of α-diversity-percentiles within grassland community plots across (a) all scale choices (18 species pools) and (b) all scale choices except the organismic scale choices with phylogenetic constraints (i.e. no species pool with only Asteraceae species to show the remaining effects after removing the most influential scale choice; 12 species pools remain). Each boxplot shows the distribution of α-diversity-percentiles across the tested scale choices. Outliers are not plotted. Community plots are ranked according to the median position of their observed α-diversity-percentiles.

*The influence of scale choices on the position of communities along a low- to high-diversity continuum*

In order to investigate the ordering of communities along a low-diversity to high-diversity continuum, we performed Spearman rank correlations between the medians of all α-diversity-percentiles (excluding the focal sampling design) and the α-diversity-percentiles for a focal sampling design.

Note that the dataset for these analyses included for each community plot six values of observed α-diversities (due to the different organismic scales) and 18 values of α-diversity-percentiles (due to the 18 different scale choices; Table 1). For all regressions, we reported the adjusted $R^2$, a goodness of fit measure that adjusts for the number of explanatory terms, the degrees of freedom and the residual standard errors. All analyses were carried out using the software R 2.14.1 (R Development Core Team, 2011).

## RESULTS

### How strong is the influence of scale choices?

For grasslands, the medians of the α-diversity-percentiles for different community plots spanned the entire gradient from values significantly higher to significantly lower than expected by chance, indicating that assembly processes differed between community plots (Fig. 3). Considering all scale choices (18 species pools; Fig. 3a), for 7% (18%) of the community plots the median of α-diversity-percentiles was above 0.95 (below 0.05). In addition to this strong effect of community plot identity there was also a strong effect of scale choices (cf. boxplot width in Fig. 3). Indeed, 9% of grassland community plots showed an extremely wide variability in the α-diversity-percentiles

(ranging from below 0.1 to above 0.9). In 1% of the grassland community plots the observed α-diversity could be either significantly high or significantly low depending on the scale choice (Fig. 3a). Reducing the organismic scale with phylogenetic constraints (i.e. a focus on Asteraceae species) had the strongest influence on the variation of α-diversity-percentiles. As this very strong effect may have masked other effects, we also report a subset of results without the phylogenetic constraint (Fig. 3b). We found that without the phylogenetic constraint, ranges of α-diversity-percentiles were strongly reduced for highly clustered (low median of α-diversity-percentiles) or over-dispersed community plots (high median of α-diversity-percentiles), while ranges for community plots with random patterns (intermediate median of α-diversity-percentiles) were still large (Fig. 3b). Results for bare-rock vegetation (see Appendix S6) and sparsely vegetated areas (see Appendix S7) were similar, but ranges of α-diversity-percentiles were generally smaller.

### Which scale choices are most critical for the outcomes?

The distribution of α-diversity-percentiles across grassland community plots was concave with more low-ranked and high-ranked than intermediately ranked community plots for the majority of scale choices (Fig. 4). Only for the phylogenetic constraint was the distribution convex, with a majority of intermediately ranked community plots. The distributions under the growth form constraint (only herbaceous species), environmental extent constraint (only grasslands) and spatial extent constraint (only one land-cover type polygon) did not differ much from the distributions at the largest scales. Results for bare-rock vegetation (see Appendix S8) and sparsely vegetated areas (see
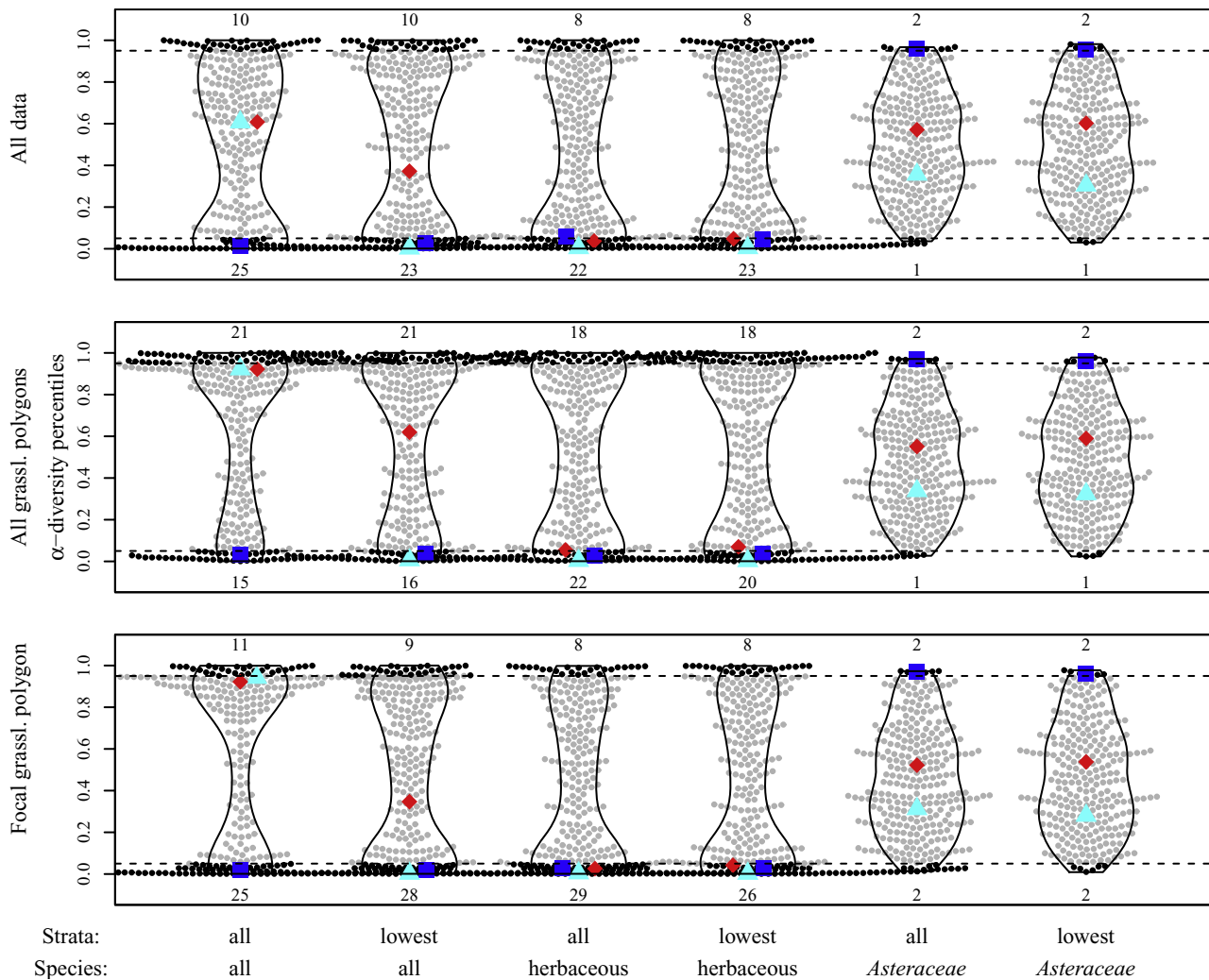
**Figure 4** Distribution of α-diversity-percentiles within scale choices across grassland community plots. Each dot in a violin plot represents one of the focal community plots; black dots identify community plots with significant high or low α-diversity-percentiles, small numbers below (and above) the violin plots give the percentage of community plots with significant patterns of clustering (or over-dispersion). The width of each violin plot refers to the density of data points. The enlarged triangle, diamond and square help to visualize the magnitude of change. Each of these identifies one community across different scale choices. Organismic scale constraints on the *x*-axis include phylogenetic constraints (all versus only Asteraceae family), growth form constraints (all versus herbaceous species only) and vegetation stratum constraints (all versus only lowest stratum). Environmental and spatial extent constraints on the *y*-axis include reduced environmental extent (all grassland polygons) and reduced spatial extent (focal grassland polygon).

Appendix S9) were similar, even though the community plots were in general less diverse.

In the regression and partial-regression analyses that included all scale choices for grassland community plots we found that neither organismic scales nor spatial and environmental extents explained much of the variation in percentile-residuals (adjusted $R^2 = 0.02$ for both; see Fig. 5a and Appendix S5). However, organismic scale explained the variation of species pool diversity very well (adjusted $R^2 = 0.98$; Fig. 5a) and the variation of α-diversity-residuals well (adjusted $R^2 = 0.63$; Fig. 5a). Spatial and environmental extents had no explanatory power for these two variables (Fig. 5a). Finally, α-diversity-residuals on their own could only explain a small part of the

variation in percentile-residuals (adjusted $R^2 = 0.15$), but explanatory power increased when we additionally accounted for the interaction of α-diversity-residuals with species pool diversity (adjusted $R^2 = 0.35$). This can be explained by the two-fold argument that the correlation between percentile-residuals and α-diversity residuals was much stronger under the phylogenetic constraint (e.g. focusing on Asteraceae only) than when considering all plant families (see Appendix S10) and that species pool diversity was strongly linked to the application of the phylogenetic constraint. Species pool diversity alone had no explanatory power for percentile residuals (Fig. 5a). To show remaining effects after removing the most influential scale choice, we performed the same analyses excluding phylogenetic
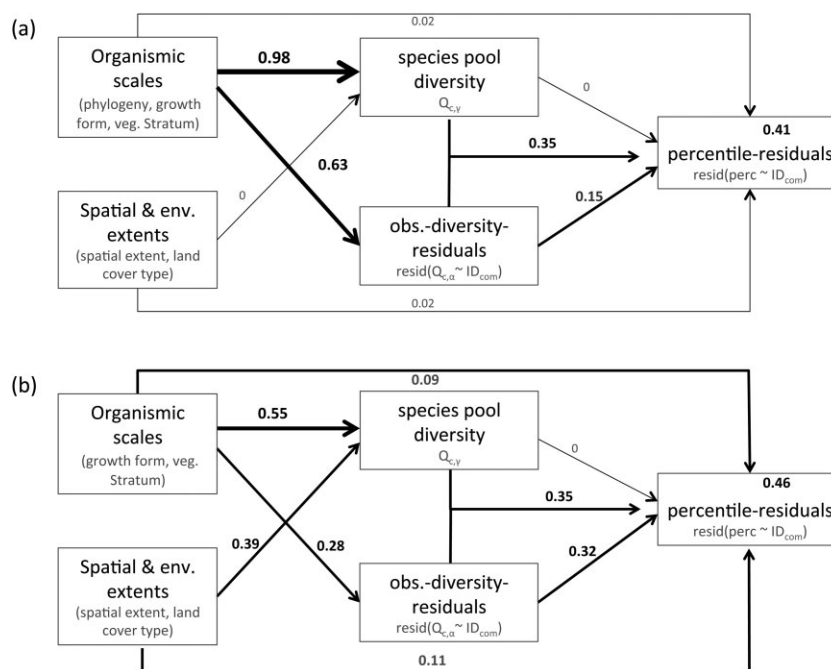
**Figure 5** Visual presentation of the regression and partial regression analyses for grassland community plots with arrows representing the effect of explanatory variables on response variables and numbers representing adjusted $R^2$ values of the respective models (see Appendix S9 for more details): (a) includes all scale choices (accordingly 18 species pools) and (b) includes all scale choices besides the choices that only include Asteraceae species (no family-only scale choices to show remaining effects after removing the most influential scale choice; 12 species pools remain). Organismic scale constraints include phylogenetic constraints (all versus only Asteraceae family), growth form constraints (all versus herbaceous species only) and vegetation stratum constraints (all versus only the lowest stratum). Environmental and spatial constraints include land-cover types and spatial extent (see Table 1 for more details). In a first step, we studied the influence of scale choices on the species pool diversity ($Q_{c\gamma}$) and on the $\alpha$-diversity-residuals (resid($Q_{c\alpha}\sim ID_{com}$), i.e. residuals of the regression of observed $\alpha$-diversity, $Q_{c\alpha}$, against community plot identity, $ID_{com}$). In a second step, we studied the influence of all variables on percentile-residuals [resid(perc$\sim ID_{com}$), i.e. residuals of the regression of $\alpha$-diversity-percentiles against $ID_{com}$].

constraints. For the grassland community plots we found an increased impact of spatial and environmental extents on both percentile-residuals (adjusted $R^2 = 0.11$, Fig. 5b) and species pool diversity (adjusted $R^2 = 0.39$). Results for bare-rock vegetation (see Appendix S11) and sparsely vegetated areas (see Appendix S12) were similar when including all scale choices, but the direct relationship between organismic scales and percentile-residuals was stronger (see Appendix S5).

In summary, organismic scale revealed the strongest overall effect. This was mainly due to phylogenetic constraints. Spatial and environmental extents also showed strong effects when phylogenetic constraints were excluded and when focusing on sparsely vegetated areas. Species pool diversity, $\alpha$-diversity-residuals and sometimes their interaction mediated these effects. All these effects differed in strength across land-cover types and stronger constraints could either lead to higher or lower $\alpha$-diversity-percentiles, indicating that there was no general trend towards more over-dispersion when reducing scales.

### For which questions is the influence of scale choices most relevant?

Signals of higher or lower than expected $\alpha$-diversities were frequent in grassland communities (Fig. 4). When choosing large

scales without constraining spatial and environmental extents ('all data') or species pools with both constraints ('focal grassland polygon'), the detection of significantly low diversity was two to three times more likely than the detection of significantly high diversity. Only scale choices with environmental, but without spatial, constraints ('all grassland polygons') increased the probability of high diversity patterns, such that phylogenetically over-dispersed community plots seemed to be about as frequent as phylogenetically clustered community plots. The organismic scale choices with phylogenetic constraints were an exception. In the latter, almost no community plot showed a significant pattern. For bare-rock vegetation (see Appendix S8) and sparsely vegetated areas (see Appendix S9) we found similar patterns with regard to low diversity but significantly high diversity was almost never detected.

However, when investigating the ordering of community plots along a low- to high-diversity continuum, i.e. a continuum from phylogenetic clustering to over-dispersion, changes were much less extreme between different scale choices. For all but the phylogenetic constraint, Spearman rank correlations between the median of $\alpha$-diversity-percentiles for a community plot and the $\alpha$-diversity-percentiles for a specific scale choice were high for all three vegetation types ($> 0.82$; Fig. 6, and see Appendices S13 & S14). Only under the phylogenetic
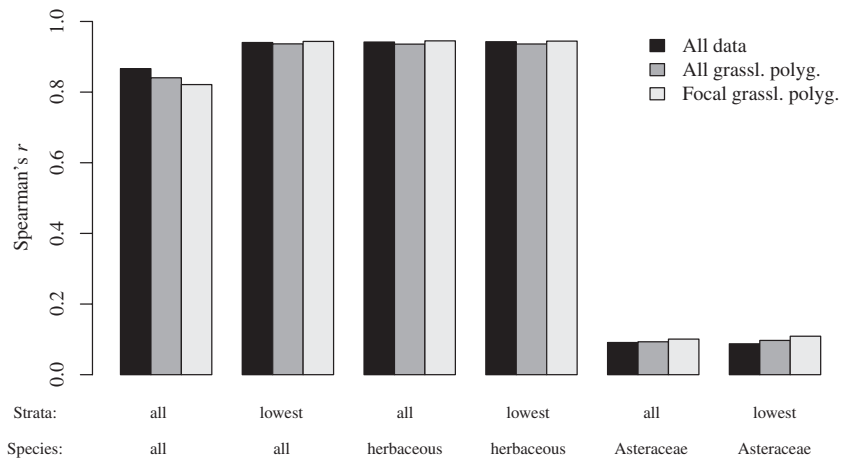
**Figure 6** Spearman rank correlations between the median of all α-diversity-percentiles for a community plot (without the focal scale choices) and the α-diversity-percentiles under focal scale choices for grassland communities.

constraints was the ordering of community plots along a low- to high-diversity continuum completely changed (Spearman rank correlations < 0.18; Fig. 6, and see Appendices S13 & S14).

## DISCUSSION

An increasing number of studies have investigated the role of ecological and evolutionary processes in community assembly by examining patterns of community structure and comparing them with null model expectations (Emerson & Gillespie, 2008; Mouquet *et al.*, 2012). While some studies have focused on a wide range of co-occurring organisms (e.g. Baraloto *et al.*, 2012), others have concentrated only on a specific clade (e.g. a family; Strauss *et al.*, 2006) or an ecological guild (e.g. Graham *et al.*, 2009). Studies have considered either just one type of habitat or a geographically restricted region (e.g. Chalmandrier *et al.*, 2013), or have examined diversity patterns at large continental scales (e.g. Hardy *et al.*, 2012). Here, we argue that scale choices in community ecology studies may be the primary drivers of their particular findings. Our results demonstrate, for a range of plant community plots across the Alps, the strong impact of different scale choices on local and regional diversities and thus ultimately on the resulting interpretation.

### How strong is the influence of the sampling design?

In the grassland polygon we found both phylogenetically over-dispersed and clustered community plots. In the two other land-cover polygons, diversity patterns were either random or clustered, but still covered a large range of diversity patterns. The overall high variation in diversity patterns among community plots may represent an indication of different processes controlling the assembly of different communities. For example, under the assumption that species niches show a high phylogenetic signal, we could hypothesize that over-dispersed communities reflect processes that limit similarity, such as competition driven by niche partitioning (Webb *et al.*, 2002) or habitat filtering in very heterogeneous habitats (Willis *et al.*, 2010). In contrast, clustered communities would reveal processes fostering the

co-occurrence of similar species, such as environmental filtering in homogeneous habitat (Webb *et al.*, 2002) or neutral dynamics of functionally equivalent species (Mayfield & Levine, 2010). Communities with random patterns can result from interacting processes, such as environmental filtering and niche-based competition (Chalmandrier *et al.*, accepted), or from strong source–sink dynamics creating strong propagule pressure from locally not well-adapted species (Münkemüller *et al.*, 2012).

In addition to the considerable between-community plot variation we found a strong effect of focal scales. For a few community plots a change of the focal scales even reversed the identified patterns from clustering to over-dispersion. This was either due to resulting changes of the locally observed set of species in the community plot or due to changes of the species pool only (i.e. for identical sets of locally observed species under changing spatial or environmental extents). It is important to note that these scale effects could neither be attributed to differences in species richness (see Appendix S15) nor to the spatial arrangement of plots (see Appendix S16).

### Which choices concerning sampling design are most critical?

We found that reducing organismic scales and especially phylogenetic scale had the strongest impact on the identification of diversity patterns. Low or completely lacking effects of growth form constraints or vegetation stratum constraints, respectively, may well be explained by the fact that we focused on non-forest systems dominated by herbaceous species. The main change when considering only one family was that the observation of significant patterns of both phylogenetic clustering and over-dispersion was much less likely. This can be explained by the reduced statistical power resulting from the reduced number of species in the local communities and in the species pool (Gotelli & Ulrich, 2012). In contrast to our results, most earlier studies found stronger patterns of over-dispersion at smaller phylogenetic scales (Appendix S1) and argued that species pools at smaller phylogenetic scales represent a functionally clustered subset from the overall species pool (Cavender-Bares *et al.*,

2006; Swenson *et al.*, 2006; Hardy & Senterre, 2007). A potential explanation for our contrasting results may be that Asteraceae form a large and ecologically highly diverse family, and thus species from this family may represent a functionally more or less random sample from the larger species pool. The lessons that can be learned from our results are, firstly, that reducing phylogenetic scales in order to improve the detection of potential patterns of over-dispersion can only be successful if the phylogenetically constrained species pool is functionally constrained as well (compared with the overall species pool) to outbalance the loss of statistical power. Secondly, the diversity patterns detected with the phylogenetically reduced scales can be very different from the diversity patterns found for the overall community of interacting species and thus cannot be used to infer general community assembly rules.

The effect of changes in spatial and environmental extent was stronger when focusing on bare-rock vegetation and sparsely vegetated areas than on grasslands. In bare-rock vegetation and sparsely vegetated areas, reducing spatial and/or environmental extents led to a decrease in phylogenetic clustering due to functionally more clustered species pools. This result is in concordance with earlier studies showing that functionally more clustered species pools allow for a better detection of limiting similarity, when this process is important for shaping community structure (Willis *et al.*, 2010; Chalmandrier *et al.*, 2013). In grassland communities, reducing the environmental extent of the species pool led to a decrease of phylogenetic clustering and to an increase of phylogenetic over-dispersion. However, reducing the spatial extent did not affect diversity patterns much. One possible explanation could be that the resulting species pool missed certain species that could potentially exist in the focal polygon, but that were either dispersal limited or outcompeted from the polygon by competitors. Another possibility is that the reduced number of species in the species pool resulted in low statistical power (Gotelli & Ulrich, 2012).

### For which questions is the influence of scale choices most relevant?

When investigating a number of plots that differed in their community assembly rules and when the focus was rather on the comparison of these communities then the impact of the chosen scales was much less severe. We found that the order of the plots along a low- to high-diversity continuum was not much affected by scale choices except for the phylogenetic constraint to only Asteraceae species.

### CONCLUSIONS

We show that scale choices are of minor importance for the ordering of community plots along a low- to high-diversity continuum. This is because changing absolute values of diversity measures has a negligible effect on the overall pattern (even if a few communities reverse their diversity patterns). Therefore, we do not expect a strong bias of scale choices for studies that aim at relating diversity patterns to abiotic variables such as climatic,

topographic, edaphic or historical variables (Graham *et al.*, 2009; Parra *et al.*, 2011; Root & Nelson, 2011) or to different levels of autocorrelation represented by spatial predictors (Peres-Neto *et al.*, 2012) in order to study, for example, diversification (Davies & Buckley, 2011), ecological theories such as the stress-gradient hypothesis (Bertness & Callaway, 1994), dispersal effects or conservation questions (Meynard *et al.*, 2011). However, our results also demonstrate that the scale choice can become a major issue when the focus of a study is on testing for significant diversity patterns (clustering versus over-dispersion) as it can reverse conclusions. This is, for instance, true for studies that aim at identifying community assembly processes for single community plots (e.g. Graham *et al.*, 2009) or when disentangling several assembly processes occurring together (e.g. Chalmandrier *et al.*, accepted). Even if scale choices can be of major importance for these studies, the answer to the question 'Which process should be studied at which scale?' is difficult to answer a priori. In cases where a priori definition of the appropriate scale is not possible it becomes necessary to consider multiple choices and support the conclusions with the comparison of results across scales.

### ACKNOWLEDGEMENTS

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Additional references can be found at the end of Appendices S1 & S2 in the online supporting information.

## SUPPORTING INFORMATION

Additional supporting information may be found in the online version of this article at the publisher's web-site.

**Appendix S1** Overview of community assembly studies that explicitly address scaling.

**Appendix S2** Details of the method.

**Appendix S3** Boxplots of distributions of α-diversity-percentiles within grassland community plots across 30 repetitions of the same scale choice.

**Appendix S4** Table with overview of species pool structures.

**Appendix S5** Table with results from regression and partial regression analyses to explain the influence of scale choices.

**Appendix S6** Boxplots of distributions of α-diversity percentiles within bare-rock community plots.

**Appendix S7** Boxplots of distributions of α-diversity-percentiles within sparsely vegetated community plots.

**Appendix S8** Violin plots of distributions of α-diversity-percentiles within scale choices across bare-rock community plots.

**Appendix S9** Violin plots of distributions of α-diversity-percentiles within scale choices across sparsely vegetated community plots.

**Appendix S10** Plot of interactive effect of α-diversity-residuals and phylogenetic constraints on percentile-residuals.

**Appendix S11** Visual presentation of the regression and partial regression analyses for bare-rock community plots.

**Appendix S12** Visual presentation of the regression and partial regression analyses for sparsely vegetated community plots.

**Appendix S13** Spearman rank correlations of α-diversity-percentiles for bare-rock communities.

**Appendix S14** Spearman rank correlations of α-diversity-percentiles for sparsely vegetated communities.

**Appendix S15** Scatterplots of the greatest range of α-diversity-percentiles across all scale choices against number of species.

**Appendix S16** Scatterplots of the greatest range in α-diversity-percentiles in space.

### BIOSKETCH

**Tamara Münkemüller**'s main interest is to better understand the ecological and evolutionary processes that drive community assembly.

T.M., L.G., S.L. and W.T. conceived the ideas and the structure of the paper and the analyses. J.L. built the AVD database with contributions from all co-authors, J.R. cleaned out the CBNA database made available by J.V.E., L.G. and S.A. C.R. built the phylogenetic tree. T.M. run all the analyses and led the writing.

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