

ECOGRAPHY

Research

Biogeographic deconstruction of phylogenetic and functional diversity provides insights into the formation of regional assemblages

Rafael Molina-Venegas, Gianluigi Ottaviani, Giandiego Campetella, Roberto Canullo and Stefano Chelli

R. Molina-Venegas (<https://orcid.org/0000-0001-5801-0736>), Univ. de Alcalá, GLOCEE – Global Change Ecology and Evolution Group, Dept of Life Sciences, Alcalá de Henares, Spain and Dept of Ecology, Faculty of Science, Univ. Autónoma de Madrid, Madrid, Spain. – *G. Ottaviani* (<https://orcid.org/0000-0003-3027-4638>) ✉ (gianluigi.ottaviani@gmail.com), Inst. of Botany of the Czech Academy of Sciences, Třeboň, Czech Republic. – *G. Campetella* (<https://orcid.org/0000-0001-6126-522X>), *R. Canullo* (<https://orcid.org/0000-0002-9913-6981>) and *S. Chelli* (<https://orcid.org/0000-0001-7184-8242>), School of Biosciences and Veterinary Medicine, Camerino Univ., Camerino, Italy.

Ecography

2022: e06140

doi: 10.1111/ecog.06140

Subject Editor: Luis Mauricio Bini

Editor-in-Chief: Miguel Araújo

Accepted 2 March 2022



Evolutionary history and environmental filtering shape the phylogenetic and functional structure of regional assemblages. However, detecting the footprint of such eco-evolutionary drivers is challenging because these may often counter each other's signature. Here, we examined whether a biogeographic deconstruction approach of phylogenetic (PD) and functional diversity (FD) patterns may help in identifying eco-evolutionary signals in extant regional assemblages. As model system, we used forest understorey angiosperms found in three regions of Italy (Alpine, Mediterranean, Continental). We quantified PD and FD of all species inhabiting the three regions (regional assemblages). Then, we computed PD and FD for the subsets of species restricted to each region (biogeographic elements), also examining diversity patterns of species found across the three regions (widespread element). We used aboveground and belowground traits capturing major plant functions to calculate FD. Additionally, we assessed FD patterns decoupled from phylogeny. We found that species restricted to climatically harsh regions (Alpine and Mediterranean elements) were phylogenetically and functionally clustered, whereas widespread species were characterised by overdispersion. Species confined to the climatically intermediate (Continental) region were randomly sorted. By including all species occurring within a region, the patterns found for the region-restricted species blurred. Phylogenetically decoupled FD patterns were qualitatively similar to non-decoupled ones with the exception of the Alpine element, where we detected a clear signature of functional differentiation between closely related species. This suggests that recent speciation events contributed to shaping the Alpine flora. Compared to the belowground compartment, aboveground traits showed a more coherent pattern with that of all-trait FD – likely because most biomass is allocated aboveground in forest understoreys. This biogeographic deconstruction study illustrates which type of eco-evolutionary insights can be gained by implementing multifaceted and integrated approaches at the macroecological scale.

Keywords: biogeographic elements, forest understorey angiosperms, functional biogeography, macroecology, phylogeny, trait-based plant ecology



www.ecography.org

© 2022 The Authors. Ecography published by John Wiley & Sons Ltd on behalf of Nordic Society Oikos

This is an open access article under the terms of the Creative Commons Attribution License, which permits use, distribution and reproduction in any medium, provided the original work is properly cited.

Introduction

Biogeographic deconstruction examines the geographic components of species assemblages separately to better understand the variety of life found within and across regions (Kornas' 1972, Jiménez-Alfaro et al. 2014). For example, Arroyo and Marañón (1990) found that the biogeographic affinities of species largely explained soil type preferences in the flora of Southern Iberian Peninsula, and Jiménez-Alfaro et al. (2021) reported that environmental predictors of strictly alpine and non-alpine species richness in regional assemblages of Central European alpine grasslands differ between the two groups. However, biogeographic deconstruction has so far been applied on taxonomic diversity, and no studies have explored the phylogenetic and functional diversity of regional assemblages from this angle. Yet, this method has the potential to provide key information into eco-evolutionary mechanisms shaping these assemblages (Fig. 1).

Regional assemblages are primarily governed by within-region speciation, extinction events and dispersal from surrounding areas (Cornell and Harrison 2014), and they are ultimately filtered by prevailing macroenvironmental

conditions of each region (Jiménez-Alfaro et al. 2018). Speciation events produce groups of closely related species which tend to show similar macroenvironmental preferences within each group due to evolutionary conservatism of major ecological niches, and thus adaptive divergence between distinct macroenvironments occurs infrequently. Consequently, closely related species often show similar geographic distributions as predicted by the phylogenetic niche conservatism hypothesis (Wiens and Donoghue 2004, Donoghue 2008). This may be particularly true in harsh regions (e.g. cold or arid climates). There, few lineages include species – either evolved as a consequence of geographically restricted speciation or dispersed from surrounding areas – with fine-tuned functional traits to cope with the strong macroenvironmental filters imposed by prevailing regional climates (Qian et al. 2019). In contrast, regions experiencing milder environmental conditions may allow the establishment of multiple lineages with disparate functional trait values. Additionally, some species show widespread distributions across regions characterised by different macroclimates, indicating a greater ability to maintain viable populations within a wider variety of macroenvironmental conditions compared to species

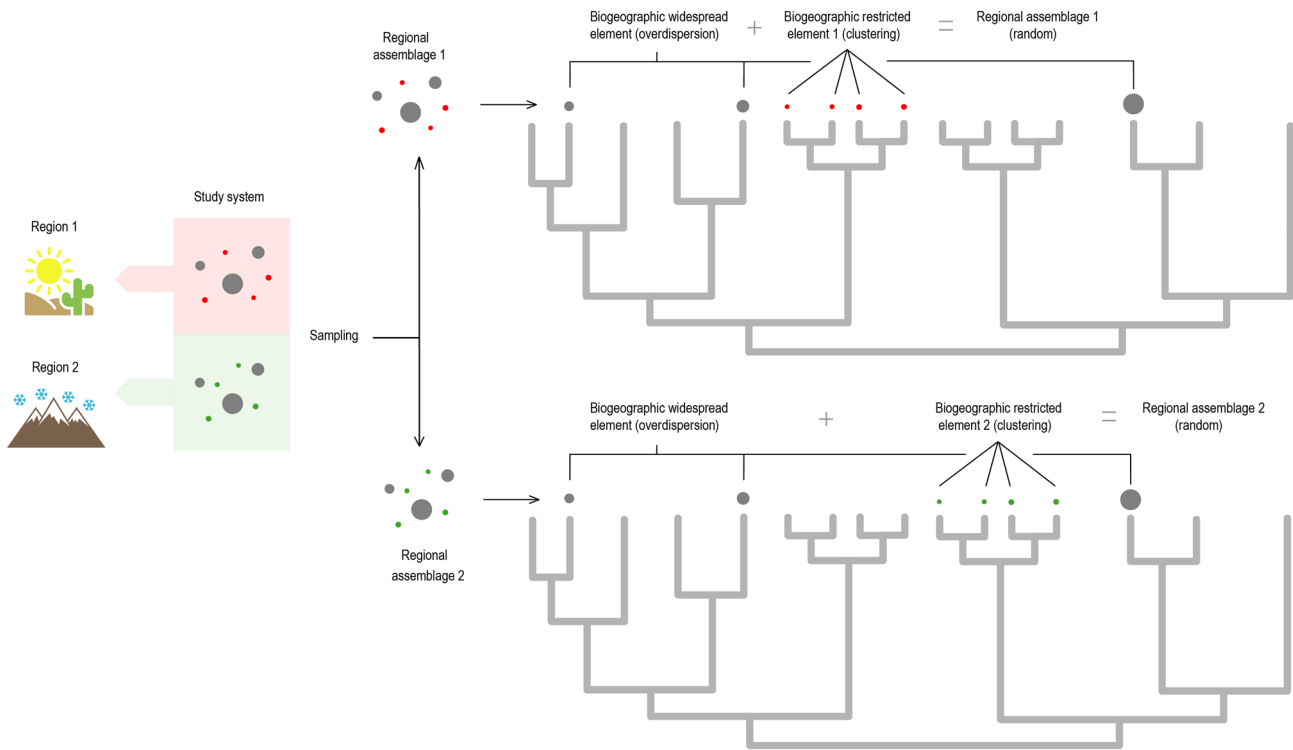


Figure 1. Biogeographic deconstruction of regional assemblages. Two hypothetical regional assemblages are shown, representing all the species (circles) occurring in an arid (assemblage 1) and a cold (assemblage 2) region. While some of the species are distributed across the two regions (grey circles), others are restricted to either region (circles in red and green for region 1 and 2). The subset of species that are restricted to either region constitute the biogeographic restricted elements 1 and 2, whereas the subset of species that are distributed across the two regions constitute the biogeographic widespread element. The size of the circles informs on the functional proximity between the species (the more similar they are in size, the closer they are in the functional space), and the phylogeny represents the evolutionary relationships among all the species (phylogenetic tips missing circles represent species that do not occur in either of the two regions). Note that the phylogenetic and functional patterns of the regional assemblages (in this case, random) are masking the phylogenetic and functional clusters exhibited by the biogeographic elements 1 and 2 and the overdispersion pattern depicted by the widespread element.

that are restricted to specific regions (Slatyer et al. 2013). Widespread species should therefore differ in their overall phenotypes as they also show different macroenvironmental preferences (Lavergne et al. 2004).

Importantly, the phylogenetic and functional signature left by species that are restricted to harsh regions (i.e. clustering) may be blurred with that of widespread species inhabiting there and elsewhere, thus hindering the identification of possible drivers responsible for the formation of regional assemblages (Conti et al. 2022). To tackle this, we implement here a biogeographic deconstruction approach to examine phylogenetic and functional diversity patterns within regions for entire regional assemblages and biogeographic elements (i.e. subsets of species restricted to each region), separately. We also assessed whether the fraction of functional diversity that can be decoupled from the phylogeny provides further insight into eco-evolutionary mechanisms shaping regional assemblages. As model system, we used a unique angiosperm plant dataset representative of all Italian forest understoreys across three regions, namely Alpine, Continental and Mediterranean.

At the regional scale, most functional diversity studies focused on a set of aboveground plant functional traits (Swenson and Weiser 2010). Here, we expand to traits which can capture the multifunctionality of plants, including both aboveground (i.e. leaf-height-seed scheme [LHS]; Westoby 1998) and belowground traits (i.e. related to clonal and resprouting abilities; Klimešová et al. 2018) that are associated with major functions played out by plants. While an increasing number of ecological and biogeographic studies included belowground traits, most of this research focused on acquisitive traits related to fine roots and mycorrhizal associations (Laliberté 2017, Weemstra et al. 2021). As a result, other non-acquisitive functions and traits linked to coarse organs – e.g. space occupancy captured by lateral spread of rhizomes – which can largely affect species persistence and distribution remain overlooked (Klimešová et al. 2021) especially at the macroecological scale considered in this study.

Here, we looked at the eco-evolutionary dynamics of regional assemblages through the lens of biogeographic deconstruction. Specifically, we asked: (Q1) Are species restricted to regions characterised by harsh climatic conditions, namely Alpine (winter cold) and Mediterranean (summer aridity) biogeographic elements, distinguished by phylogenetic and (multiple-trait) functional clustering patterns? This pattern should emerge because of 1) phylogenetic niche conservatism together with strong environmental filtering acting at the regional scale, or 2) recent geographically restricted speciation. (Q2) Do species occurring across all regions (i.e. widespread element) and those restricted to the climatically intermediate region (i.e. Continental element) show phylogenetic and functional overdispersion patterns? If phylogenetic niche conservatism prevails, widespread and Continental elements should be distinguished by several distantly related lineages showing disparate functional trait values. (Q3) Is the phylogenetic and functional signature of Alpine and Mediterranean biogeographic elements

confounded with that left by the set of all species occurring in each region (regional assemblages)?

Material and methods

Study area, sampling design and species categorisation

The study area is constituted by the entirety of Italian forests. We selected 201 forest sites by using a systematic approach, which can therefore be considered representative of all Italian forests (Chiarucci et al. 2019). The sampling design was based on a grid of 16×16 km superimposed onto the whole country; at each node of the grid, a sampling site was selected if a forest larger than 1 ha was found after field validation. This system belongs to the transnational network for monitoring the forest health status in Europe (ICP Forest 2016). At each site, we sampled understorey angiosperm plant species in plots of 400 m^2 – understorey being defined as the herb and shrub layer < 5 m in height. The sampling was done during summer of 2007 following standardised protocols (Chelli et al. 2019a, b). The 201 sites were distributed across the three biogeographic regions occurring in Italy, identified according to the EU policy for nature protection (ETC-BD 2006), namely Alpine (81 sites), Continental (32 sites) and Mediterranean (88 sites) (Fig. 2). A principal component analysis conducted on 19 WorldClim bioclimatic variables showed that most Alpine and Mediterranean sites are located toward the ends of a precipitation and temperature gradient (colder and wetter for Alpine sites and warmer and drier for Mediterranean sites), whereas Continental sites occupied an intermediate position in the gradient (see Supporting information).

The total number of species ($n=1072$) was split in two different ways. On the one hand, we defined regional assemblages as the set of all species occurring in each region – Alpine ($n=595$), Continental ($n=334$), Mediterranean ($n=687$). On the other hand, the species were sorted into exclusive biogeographic elements, namely species that were found only in one region – Alpine ($n=301$), Continental ($n=37$), Mediterranean ($n=355$). We also included species that were observed across the three regions as a fourth biogeographic element (i.e. hereafter widespread element; $n=165$).

Phylogeny

We obtained a time-calibrated phylogeny of DNA for all the understorey plant species in the dataset using the R package ‘V.PhyloMaker’ (Jin and Qian 2019). Briefly, this software serves from a global mega-tree (GBOTB.extended; Jin and Qian 2019) to which missing species are bound following a systematic procedure. Once missing species have been inserted, the mega-tree is pruned to the list of species provided by the user to produce a phylogeny. Here, the missing species ($n=350$, equalling 33% of the pool) were bound to a randomly selected position in the tree at and below the corresponding genus crown node (binding scenario 2; Jin and

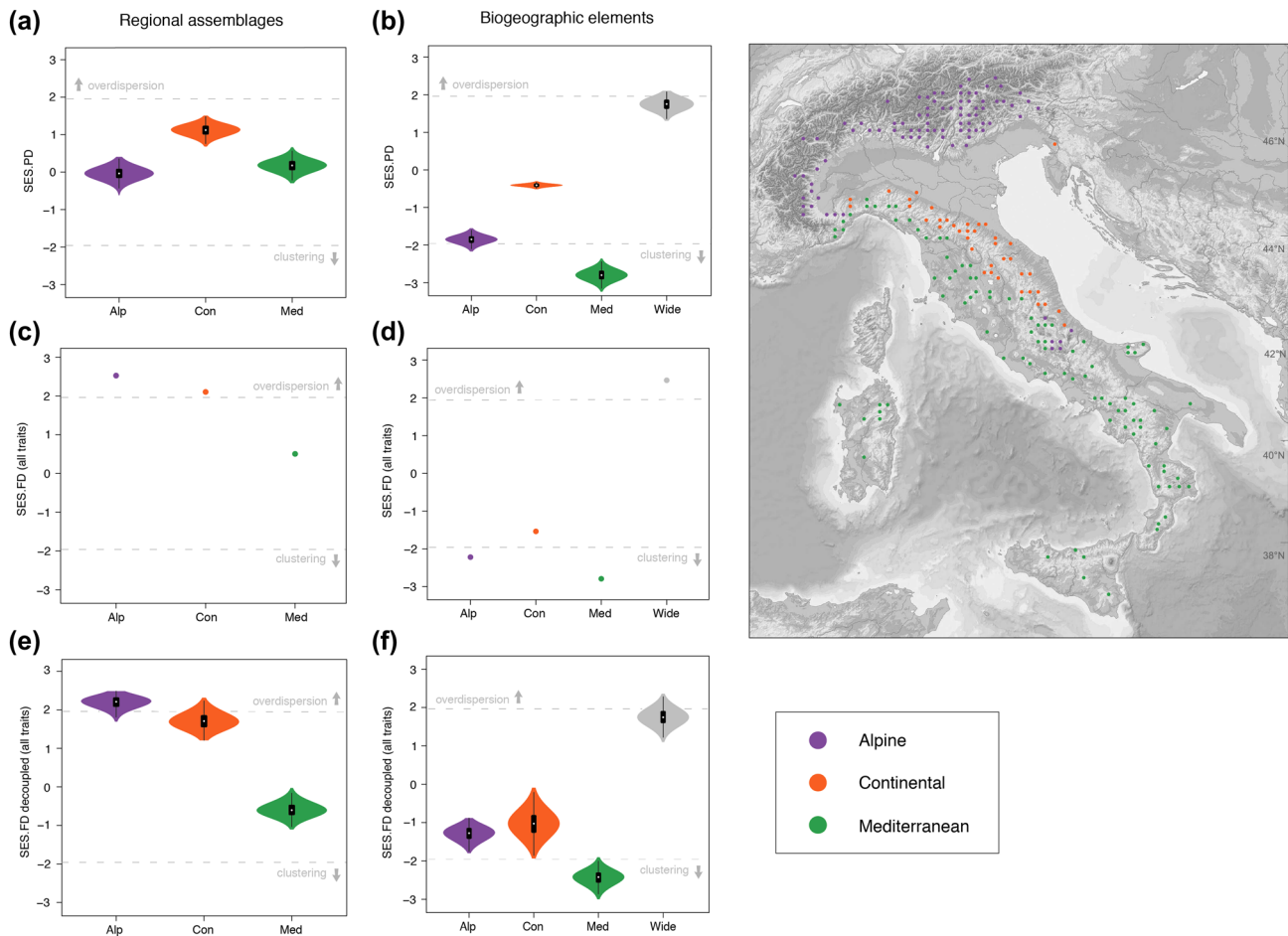


Figure 2. On the right, the map reporting the location and distribution of the 201 sites in Italy. The upper panels show violin plots with the distribution of standardised phylogenetic diversity values (SES.PD scores computed from 1000 possible phylogenies) for (a) regional assemblages (species occurring in the Alpine, Continental and Mediterranean region, respectively) and (b) biogeographic elements (Alpine-restricted, Continental-restricted, Mediterranean-restricted and widespread species). The panels in the centre show the standardised functional diversity values (SES.FD scores) derived from all traits combined for (c) the same regional assemblages and (d) biogeographic elements. The lower panels report phylogenetically decoupled FD patterns for (e) regional assemblages and (f) biogeographic elements. The horizontal grey dashed lines are visual references at $y = \pm 1.96$ (significance thresholds for a 5% nominal alpha).

Qian 2019), and this procedure was repeated to generate a distribution of 1000 possible phylogenies. All subsequent phylogenetic analyses were replicated and results averaged over 1000 trees to account for phylogenetic uncertainty (Rangel et al. 2015, Molina-Venegas et al. 2020a).

Functional traits

We included five aboveground and belowground functional traits. The selected traits are well-known to respond to changing environments, especially to macroclimate (Lavorel and Garnier 2002, Chelli et al. 2019a, b). Specifically, for the aboveground compartment, we selected LHS traits (Westoby 1998), namely: 1) specific leaf area (SLA; informing on resource economics, especially for light capture in forest understoreys), 2) plant height (linked to aboveground

competitive ability), 3) seed mass (associated with dispersal ability and establishment success). Concerning the belowground compartment, we chose two traits that can greatly affect local persistence through non-acquisitive functions (Ottaviani et al. 2017, Klimešová et al. 2018): 4) belowground bud bank size (related to resprouting potential), 5) lateral spread (capturing space occupancy and resource exploration). We could not gather trait data directly linked to belowground resource acquisition and conservation strategies (e.g. specific root length or root dry matter content) because trait coverage in the databases for the species list was very low. However, evidence showed that this functional axis can be proxied by SLA (Freschet et al. 2010, de la Riva et al. 2016). All trait data were gathered from existing databases and published literature at the species level, namely from Kleyer et al. (2008), Royal Botanical Gardens Kew (2008),

Campetella et al. (2011) and Klimešová et al. (2017) (see Supporting information for details on trait coverage).

Phylogenetic and functional diversity of regional assemblages and biogeographic elements

Before running the diversity analyses, we estimated phylogenetic signal in the traits using the Pagel's lambda model of evolution (Pagel 1999). The lambda statistic has a natural scale between zero (complete lack of correlated evolution between species) and one (correlated evolution equates Brownian motion expectation), and it has proven to be strongly resilient to branch length inaccuracies in the phylogenies (Molina-Venegas and Rodríguez 2017). We used likelihood ratio tests to assess whether there is significant deviance from a scenario of complete lack of phylogenetic signal in the traits. These tests were conducted using the *phylosig* function of 'phytools' R package (Revell 2012).

We used the phylogenetic diversity metric (i.e. minimum spanning path connecting a set of species in a dendrogram; Faith 1992) as the basis to compute both phylogenetic diversity (PD) and functional diversity (FD). These metrics were calculated for regional assemblages (i.e. all species occurring in the Alpine, Continental and Mediterranean region, respectively) and biogeographic elements (i.e. species restricted to the Alpine, Continental and Mediterranean region, respectively, and widespread ones). PD was computed for each set of species using the phylogeny described above (phylogenetic dendrogram), and FD was quantified using functional dendrograms derived from Euclidean distance matrices. We used the UPGMA agglomeration method to derive the functional dendrograms because it has been demonstrated to be more robust than alternative classifications (Podani and Schmera 2006). Traits were standardised (mean=0, SD=1) prior to computing distance matrices, and functional dendrograms were obtained with the *hclust* R function (<www.r-project.org>). FD was calculated for different grouping levels of traits, namely 1) 'all-traits' (aboveground and belowground traits combined), 2) 'aboveground' (LHS traits together) and 'belowground' (belowground bud bank size and lateral spread together) and 3) for single traits. We only considered species with known values across all traits in each grouping level for the analyses (see Supporting information). To assess the evolutionary imprint on all-trait FD patterns, we decoupled FD from the phylogeny using the phylogenetic eigenvector-based approach proposed by de Bello et al. (2017). Briefly, the phylogeny is decomposed in eigenvectors from a principal coordinate analysis which are in turn used as explanatory variables in a model where traits are set as response variables. The residuals of the model are taken as the variation in species' traits decoupled from phylogeny, and they are used to compute a decoupled functional distance matrix (Desdevises et al. 2003, Diniz-Filho et al. 2012). Provided that functional traits show some degree of phylogenetic signal, decoupled FD will be higher and lower than non-decoupled FD to the extent that closely related and distantly related species show more and less divergent trait

values than expected for the given phylogeny, respectively. We used the *decouple* R function with default settings (de Bello et al. 2017) to compute decoupled functional distance matrix for each regional assemblage and biogeographic element, and these matrices were processed as described above to obtain decoupled functional dendrograms.

PD and FD are not statistically independent from species richness. Therefore, we computed standardised effect size (SES) scores for PD and FD using a null model approach and the *ses.pd* function as implemented in the 'picante' R package (Kembel et al. 2010). Null distributions were generated for each raw PD and FD value by shuffling taxa labels 999 times across the tips of the phylogenetic (n=1072) and functional dendrograms – all traits (n=558), aboveground (n=758), belowground (n=635), SLA (n=817), plant height (n=867), seed mass (n=990), belowground bud bank size (n=638) and lateral spread (n=635) – and SES scores were computed as:

$$SES = \frac{M_{obs} - M_{null}}{SD_{null}} \quad (1)$$

where M_{obs} is the raw value of the corresponding metric (i.e. PD or FD) and M_{null} and SD_{null} are the mean and standard deviation of the null distribution, respectively (Kembel 2009). For a nominal alpha of 5%, PD or FD will be significantly higher (overdispersion) or lower (clustering) than expected for the given null model if the corresponding SES scores are > 1.96 and < -1.96 , respectively.

We used a 'hot node' approach to complement the phylogenetic diversity analyses described above. The hot node analysis serves to identify phylogenetic clades encompassing a significantly high number of species with a certain property of interest (e.g. being restricted to a given region) relative to the entire phylogeny (Saslis-Lagoudakis et al. 2012, Molina-Venegas et al. 2020a, b), and hence those clades mainly contributing to a phylogenetic structuring. Here, we used this approach to identify the phylogenetic clades that were significantly overrepresented in the biogeographic elements that showed a clear tendency towards phylogenetic clustering (see Molina-Venegas et al. 2020a for a detailed description of the method).

Table 1. Phylogenetic signal of functional traits. The averaged values of the lambda statistic across n=1000 phylogenetic trees are provided together with 95% confidence intervals. All likelihood ratio tests were significant for a nominal alpha of 0.1%. SLA=specific leaf area.

Trait	Compartment	Mean lambda	Lower 95% CI	Upper 95% CI
SLA	Aboveground	0.6822	0.6817	0.6827
Seed mass	Aboveground	0.9824	0.9808	0.9840
Plant height	Aboveground	0.9533	0.9531	0.9536
Bud bank size	Belowground	0.5225	0.5215	0.5235
Lateral spread	Belowground	0.5006	0.5000	0.5011

Results

Phylogenetic signal on functional traits

Phylogenetic signal was stronger in the aboveground compartment, with both seed mass and plant height showing significantly high values of the lambda statistic (close to Brownian motion expectation) and SLA distinguished by an intermediate-to-high signal (Table 1). In contrast, belowground traits showed intermediate signals.

Phylogenetic diversity patterns

The phylogenetic diversity (PD) of regional assemblages did not deviate from random expectation in any of the regions (Fig. 2). However, the Mediterranean biogeographic element was characterised by strong phylogenetic clustering, and a tendency towards clustering was observed also for the species restricted to the Alpine region. In contrast, the Continental biogeographic element was randomly structured, whereas the widespread element tended to be overdispersed (Fig. 2).

The hot node analysis suggested that the phylogenetic clustering observed for the Mediterranean biogeographic element and, to a lesser extent, the Alpine element were driven by different phylogenetic clades. On the one hand, the Mediterranean element was dominated by clades including Lamiaceae, Cistaceae and the Trifolieae tribe (Fabaceae) (Fig. 3). On the other hand, the Alpine element was largely constituted of species affiliated to the Saxifragaceae, Crassulaceae, Ranunculaceae, Ericales (Ericaceae, Primulaceae) and

Asterales clades. The latter included a high number of nested genus-level hot nodes (*Phyteuma*, *Campanula*, *Carduus* + *Cirsium*, *Centaurea*) – a pattern that was not observed in the Mediterranean element (Fig. 3).

Functional diversity patterns

For functional diversity (FD) including all traits together, we found overdispersion in the Alpine and Continental regional assemblages and random pattern in the Mediterranean one (Fig. 2). However, these patterns strongly differed from the FD observed in the corresponding biogeographic elements, which instead largely mirrored phylogenetic diversity patterns, namely significant clustering for the Mediterranean and Alpine elements, random pattern for Continental, and overdispersion for the widespread element (Fig. 2). Phylogenetically decoupled FD patterns were qualitatively similar to non-decoupled ones, with the notable exception of the Alpine element, which shifted from clustering to random regardless of phylogenetic uncertainty (Fig. 2). Phylogenetically decoupled FD in the widespread element became less overdispersed than the non-decoupled metric, and a similar trend was observed for the Continental regional assemblage.

The aboveground compartment FD (i.e. LHS traits together) of regional assemblages and biogeographic elements mirrored the patterns observed for all-trait FD, with the exceptions that the Alpine assemblage showed random pattern and the Continental element showed marginally



Figure 3. Results of the hot node analysis conducted on the biogeographic elements that showed significant levels of phylogenetic clustering, i.e. Alpine in purple (left phylogeny) and Mediterranean in green (right phylogeny). Hot nodes are highlighted on one randomly selected tree ($n = 1000$ possible topologies) that included all the hot clades detected in the analysis. We only considered as ‘hot’ those nodes that were observed and showed statistical significance (5% nominal alpha) in at least 95% of the trees analysed. The circles on the phylogenetic tips represent the constituent species of each biogeographic element.

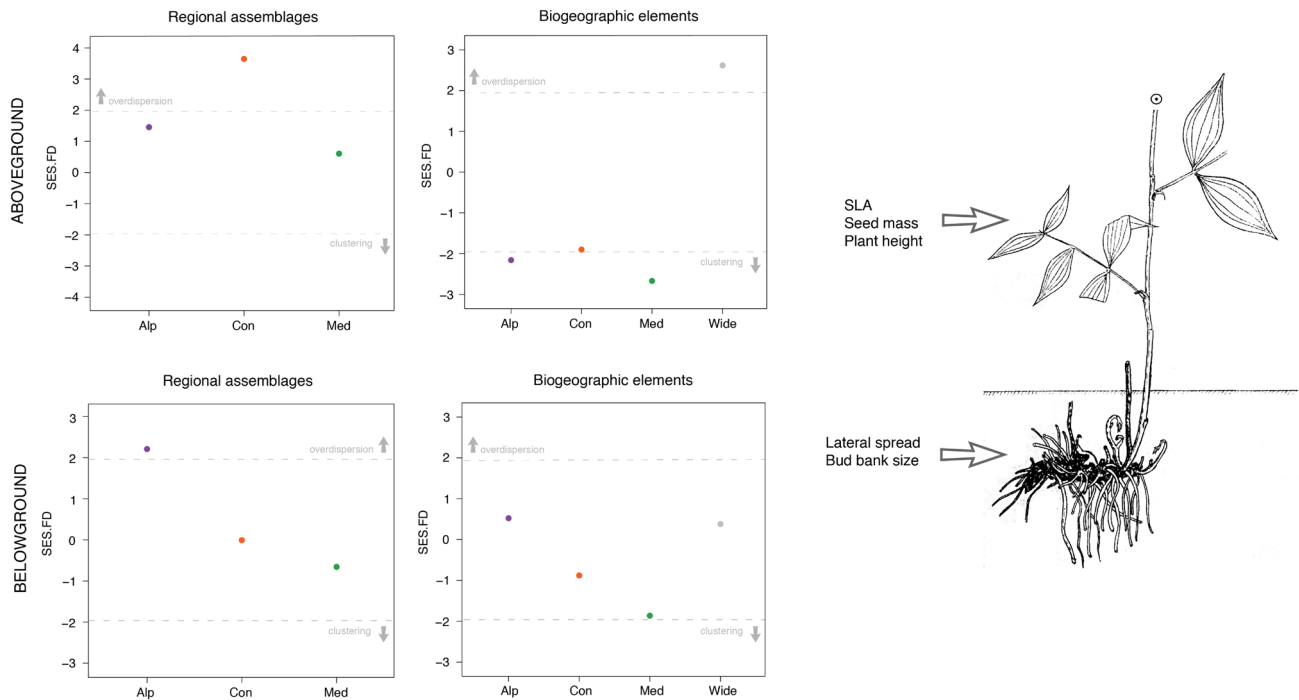


Figure 4. Standardised functional diversity values (SES.FD scores) associated with the aboveground (upper panels) and belowground (lower panels) compartments for regional assemblages (left panels) and biogeographic elements (right panels). The horizontal grey dashed lines are visual references at $y = \pm 1.96$ (significance thresholds for a 5% nominal alpha). The drawing illustrates an example of a forest understorey species (*Lathyrus vernus*) with traits indicated – image extracted and modified from CloPla database (Klimešová et al. 2017).

significant clustering instead (Fig. 4). In contrast, in the belowground compartment (i.e. bud bank size and lateral spread together) only the Mediterranean biogeographic element showed a marginally significant FD pattern towards clustering, whereas regional assemblages showed similar patterns as revealed for all-trait FD with the exception that the Continental assemblage showed random pattern instead of overdispersion.

For single-trait FD, we generally revealed random patterns both for regional assemblages and biogeographic elements (see Supporting information). Exceptions included SLA in the Alpine regional assemblage showing some degree of clustering, and seed mass in the Alpine element distinguished by strong clustering. Regarding belowground traits, only lateral spread showed a marginal tendency towards clustering in the Mediterranean element.

Discussion

We identified that species restricted to regions characterised by harsher climatic conditions (i.e. Alpine and Mediterranean elements) form phylogenetic and functional clusters, yet these patterns are likely driven by different mechanisms. This eco-evolutionary signal, however, was masked when all species found within each region were considered. This finding may have far-reaching implications: these non-random ‘hidden’ eco-evolutionary patterns would have gone undetected had we focused only on the regional assemblages, hence proving

the informative value of deconstructing regional assemblages’ phylogenetic and functional diversity patterns into biogeographic elements.

Different eco-evolutionary mechanisms behind phylogenetic and functional (all-trait) diversity patterns of biogeographic elements

We found support for our first expectation as we revealed phylogenetic and functional clusters formed by species restricted to the harsh Mediterranean and Alpine regions, likely related to a strong climate forcing. Yet, this forcing may be associated with different prevailing evolutionary mechanisms, which can operate with a variable strength in different regions. The hot node analysis revealed that the phylogenetic clustering observed in the Alpine element is largely due to a few closely related groups of congenics (all of them within the Asterales clade). Although the Alpine element showed functional clustering, its phylogenetically decoupled FD revealed that within each group of congenics, species show some degree of functional differentiation. This suggests that recent geographically restricted speciation events (possibly adaptive) may have contributed to shaping the Alpine flora. The aforementioned groups of Asterales congenics are distinguished by highly variable (aboveground) traits, with differences up to 3- or 4-fold in the raw values (e.g. across *Phyteuma*, *Campanula*, *Cirsium*, *Centaurea* species, see Supporting information). Indeed, some of these genera underwent profuse speciation across alpine habitats of Europe

(Smyčka et al. 2021) and North America (DeChaine et al. 2014) during the Pleistocene. Besides, other hot clades – such as Ranunculaceae, Ericaceae and Saxifragaceae – represent long-recognised montane lineages of cold-temperate affinity (Schwery et al. 2015). Despite the species in the aforementioned hot clades largely contributed to the observed trend towards phylogenetic clustering in the Alpine element, it is worth mentioning that these clades are distantly related, which may have countered the tendency.

Conversely, the strong functional clustering identified for the Mediterranean element persisted after accounting for phylogenetic relatedness. This may imply a prevalent role of phylogenetic niche conservatism, with certain lineages showing similar trait values. Indeed, Lamiaceae and Cistaceae hot-node clades largely underpinned the strong clustering pattern reported for the Mediterranean element (Fig. 3), and these tend to thrive under dry and seasonally warm climates. Previous studies have documented a diversification process occurring with the onset of the seasonal, dry and warm Mediterranean climate, so that lineages sharing a functional syndrome rapidly radiated (Verdú and Pausas 2013). However, the hot node analysis suggests that this syndrome-driven diversification may not be reflected in our data, likely because recently evolved Mediterranean species are found in open rather than closed, forested habitats as the understories considered in this study (cf. Molina-Venegas et al. 2017, Buirra et al. 2021). Therefore, the Mediterranean element seems composed of a diversity of closely related species sharing similar trait values as a result of phylogenetic niche conservatism together with strong macroenvironmental filters acting at the regional scale.

The Mediterranean Basin is a melting pot for biodiversity, including not only typically Mediterranean species that diversified after the onset of the Mediterranean climate (Verdú and Pausas 2013) but many putatively older lineages of tropical (e.g. *Hedera*, *Tamus*) and temperate (e.g. *Amelanchier*, *Frangula*) affinities representing species that evolved in pre-Mediterranean tropical-like scenarios (Herrera 1992). In the Mediterranean forest understories of Italy, these lineages are represented by widely distributed and evolutionarily distinct species (*Hedera* and *Tamus* are the only representative genera of the Araliaceae and Dioscoraceae families in the Mediterranean, respectively) that can considerably increase phylogenetic diversity at the regional level. As a result, the evolutionary footprint left by Mediterranean restricted species (clustering) could be masked. This example further illustrates the usefulness of biogeographic deconstruction methods to unravel the long-lasting effects of past ecological scenarios into the phylogenetic and functional structure of present assemblages (Barthelemy et al. 2021).

Species restricted to the Continental region revealed a general lack of structuring (random phylogenetic and functional pattern) while the expectation was overdispersion. Continental Italian forests have been subjected to historical intensive logging (e.g. Po plain, Romano and Zullo 2016) and are nowadays reduced to scattered small-sized forest

patches. Thus, non-climatic environmental stressors, such as habitat destruction and edge effects, may have also played a role in shaping the random pattern in the Continental element. Species belonging to distantly related lineages with disparate functional trait values may have been filtered out because unable to thrive in fragmented and/or disturbed forest patches, therefore contributing to diminish the expected overdispersion. Nonetheless, this result should be taken cautiously due to the low number of sites (32) and species (37) restricted to the Continental region which may prevent the identification of clear patterns.

Concerning widespread species (i.e. those occurring across all regions), we found support for our expectation as these species show a consistent tendency towards phylogenetic and functional overdispersion. Widespread species exhibited diverse functional strategies and proved their ability to arrive, colonise, establish and reproduce successfully under highly different climates (i.e. regions) while belonging to disparate lineages. Yet, decoupled FD suggests that some distantly related widespread species tend to show more similar trait values than expected from their phylogenetic relationships, which questions phylogenetic niche conservatism as the only explanation for their FD pattern. Nevertheless, statistical significance of decoupled FD was strongly dependent on phylogenetic uncertainty (Fig. 2f), and therefore any inference about possible mechanisms should be handled with caution.

Breaking down functional diversity patterns: aboveground and belowground traits

We revealed that the all-trait FD patterns across the four biogeographic elements were largely mirrored in the FD pattern of the aboveground compartment, whereas belowground traits were mainly characterised by random settings. This implies that across the four biogeographic elements, the major role was played by the aboveground dimension in shaping the functional syndrome, possibly due to most biomass being allocated aboveground in forests (Ottaviani et al. 2020a). Yet, the belowground compartment also contributed (although to a lesser extent) in modulating the all-trait FD clustering syndrome for the Mediterranean element (Fig. 4). When zooming into individual trait FD patterns, these either aligned or opposed that observed for all traits. For instance, seed mass was strongly clustered in the Alpine element (see Supporting information), confirming that the harsh climates (i.e. cold winters and short growing seasons) may select for species having similar sexual reproduction and dispersal strategies (DeMalach et al. 2019). Conversely, SLA in the Mediterranean element showed a random pattern (see Supporting information). Among the species responsible for this pattern, a striking example is represented by *Capparis spinosa*, a tropical relic characterised by acquisitive leaves (SLA value ca 5 times higher than the average of the Mediterranean element). This contrasts with most Mediterranean-restricted species typically distinguished by resource-conservative leaves (Cowling et al. 1996).

Our results on individual traits call for careful interpretation of functional diversity patterns. Inferences should be tightly

connected with the specific functions captured by the trait, and generalisations extended to overall plant strategies should be considered with caution. Only when the selected traits can capture major plant functions able to proxy the multidimensional nature of plants (as suggested for e.g. the island syndrome; Ottaviani et al. 2020b), interpretations may be generalised.

Concluding remarks

This study shows how macroecological research can benefit from biogeographic deconstruction exercises that aim at analysing phylogenetic and functional diversity patterns of co-distributed subsets of species separately (i.e. biogeographic elements). As such, this approach was able to reveal 'hidden' phylogenetic and functional structuring in regional plant assemblages. Our findings point towards a regional climate forcing that forms clusters of species sharing similar key functional strategies to cope with harsher macroenvironmental conditions – a pattern that would have gone otherwise undetected. However, the eco-evolutionary mechanisms generating the phylogenetic and functional diversity patterns are likely different among regions; phylogenetic niche conservatism appears more important in the Mediterranean region whereas recent geographically restricted speciation events seem also relevant in the Alpine region. Additionally, we used a multifunctional, whole-plant approach including traits that can largely affect species persistence strategies and distribution, which is rarely implemented at the macroecological scale. We encourage future studies to 1) further explore the potential of deconstruction methods for phylogenetic and functional macroecology and biogeography, and 2) expand the use of multifaceted, integrated and cross-regional approaches to other areas and biomes.

Acknowledgements – We thank the subject editor Luis Mauricio Bini, Leandro Duarte and two anonymous reviewers for providing insightful comments during the review process. We also thank the Scientific Computation Centre of Andalusia (CICA) for the computing services they provided.

Funding – This work was partially supported by the Thünen Inst. (Hamburg) and the ICP Forests' Italian Focal Centre (CUFA, Comando per la Tutela della Biodiversità e dei Parchi – Ufficio Studi e Progetti, Roma). RM-V was supported by the TALENTO program (2018-T2/AMB-10332) of the Regional Government of the Community of Madrid (Spain). GO was supported by the Czech Science Foundation (GACR project no. 19-14394Y), and the long-term research development project no. RVO 67985939 of the Czech Academy of Sciences.

Author contributions

Rafael Molina-Venegas: Conceptualization (lead); Data curation (lead); Formal analysis (lead); Investigation (lead); Methodology (lead); Visualization (lead); Writing – original draft (lead); Writing – review and editing (lead). **Gianluigi Ottaviani:** Conceptualization (equal); Investigation (equal);

Writing – original draft (lead); Writing – review and editing (lead). **Giandiego Campetella:** Funding acquisition (equal); Resources (equal); Writing – review and editing (supporting). **Roberto Canullo:** Funding acquisition (equal); Resources (equal); Writing – review and editing (supporting). **Stefano Chelli:** Data curation (supporting); Funding acquisition (equal); Project administration (lead); Writing – review and editing (supporting).

Transparent peer review

The peer review history for this article is available at <<https://publons.com/publon/10.1111/ecog.06140>>.

Data availability statement

Data used in this research is available from the Figshare Digital Repository: <<https://doi.org/10.6084/m9.figshare.19222473>> (Molina-Venegas et al. 2022).

Supporting information

The supporting information associated with this article is available from the online version.

References

- Arroyo, J. and Marañón, T. 1990. Community ecology and distributional spectra of Mediterranean shrublands and heathlands in Southern Spain. – *J. Biogeogr.* 17: 163–176.
- Barthelemy, E. et al. 2021. Imprints of past habitat area reduction on extant taxonomic, functional and phylogenetic composition. – *Front. Ecol. Evol.* 9: 332.
- Buira, A. et al. 2021. The contribution of the edaphic factor as a driver of recent plant diversification in a Mediterranean biodiversity hotspot. – *J. Ecol.* 109: 987–999.
- Campetella, G. et al. 2011. Patterns of plant trait–environment relationships along a forest succession chronosequence. – *Agric. Ecosyst. Environ.* 145: 38–48.
- Chelli, S. et al. 2019a. Climate is the main driver of clonal and bud bank traits in Italian forest understories. – *Perspect. Plant Ecol. Evol. Syst.* 40: 125478.
- Chelli, S. et al. 2019b. Effects of climate, soil, forest structure and land use on the functional composition of the understorey in Italian forests. – *J. Veg. Sci.* 30: 1110–1121.
- Chiarucci, A. et al. 2019. Exploring patterns of beta-diversity to test the consistency of biogeographical boundaries: a case study across forest plant communities of Italy. – *Ecol. Evol.* 9: 11716–11723.
- Conti, L. et al. 2022. Insularity promotes plant persistence strategies in edaphic island systems. – *Global Ecol. Biogeogr.* 31: 753–764.
- Cornell, H. V. and Harrison, S. P. 2014. What are species pools and when are they important? – *Annu. Rev. Ecol. Evol. Syst.* 45: 45–67.
- Cowling, R. M. et al. 1996. Plant diversity in Mediterranean-climate regions. – *Trends Ecol. Evol.* 11: 362–366.

- de Bello, F. et al. 2017. Decoupling phylogenetic and functional diversity to reveal hidden signals in community assembly. – *Methods Ecol. Evol.* 8: 1200–1211.
- de la Riva, E. G. et al. 2016. A plant economics spectrum in Mediterranean forests along environmental gradients: is there coordination among leaf, stem and root traits? – *J. Veg. Sci.* 27: 187–199.
- DeChaine, E. G. et al. 2014. Integrating environmental, molecular and morphological data to unravel an ice-age radiation of arctic-alpine *Campanula* in western North America. – *Ecol. Evol.* 4: 3940–3959.
- DeMalach, N. et al. 2019. Mechanisms of seed mass variation along resource gradients. – *Ecol. Lett.* 22: 181–189.
- Desdevises, Y. et al. 2003. Quantifying phylogenetically structured environmental variation. – *Evolution* 57: 2647–2652.
- Diniz-Filho, J. A. F. et al. 2012. On the selection of phylogenetic eigenvectors for ecological analyses. – *Ecography* 35: 239–249.
- Donoghue, M. J. 2008. A phylogenetic perspective on the distribution of plant diversity. – *Proc. Natl Acad. Sci. USA* 105: 11549–11555.
- ETC-BD (European Topic Centre on Biological Diversity) 2006. The indicative map of European biogeographical regions: methodology and development. – France ETC-BD.
- Faith, D. P. 1992. Conservation evaluation and phylogenetic diversity. – *Biol. Conserv.* 61: 1–10.
- Freschet, G. T. et al. 2010. Evidence of the ‘plant economics spectrum’ in a subarctic flora. – *J. Ecol.* 98: 362–373.
- Herrera, C. M. 1992. Historical effects and sorting processes as explanations for contemporary ecological patterns: character syndromes in Mediterranean woody plants. – *Am. Nat.* 140: 421–446.
- ICP Forest 2016. MANUALS. – <<http://icp-forests.net/page/icp-forests-manual>>.
- Jiménez-Alfaro, B. et al. 2014. Biogeographic deconstruction of alpine plant communities along altitudinal and topographic gradients. – *J. Veg. Sci.* 25: 160–171.
- Jiménez-Alfaro, B. et al. 2018. History and environment shape species pools and community diversity in European beech forests. – *Nat. Ecol. Evol.* 2: 483–490.
- Jiménez-Alfaro, B. et al. 2021. Post-glacial determinants of regional species pools in alpine grasslands. – *Global Ecol. Biogeogr.* 30: 1101–1115.
- Jin, Y. and Qian, H. 2019. VPhyloMaker: an R package that can generate very large phylogenies for vascular plants. – *Ecography* 42: 1353–1359.
- Kembel, S. W. 2009. Disentangling niche and neutral influences on community assembly: assessing the performance of community phylogenetic structure tests. – *Ecol. Lett.* 12: 949–960.
- Kembel, S. W. et al. 2010. Picante: R tools for integrating phylogenies and ecology. – *Bioinformatics* 26: 1463–1464.
- Kleyer, M. et al. 2008. The LEDA Traitbase: a database of life-history traits of the Northwest European flora. – *J. Ecol.* 96: 1266–1274.
- Klimešová, J. et al. 2017. CLO-PLA: a database of clonal and bud-bank traits of the Central European flora. – *Ecology* 98: 1179–1179.
- Klimešová, J. et al. 2018. Belowground plant functional ecology: towards an integrated perspective. – *Funct. Ecol.* 32: 2115–2126.
- Klimešová, J. et al. 2021. Incorporating clonality into the plant ecology research agenda. – *Trends Plant Sci.* 26: 1236–1247.
- Kornas, J. 1972. Corresponding taxa and their ecological background in the forests of temperate Eurasia and North America. – In: Valentine, D. H. (ed.), *Taxonomy, phytogeography and evolution*. Academic Press, pp. 37–59.
- Laliberté, E. 2017. Below-ground frontiers in trait-based plant ecology. – *New Phytol.* 213: 1597–1603.
- Lavergne, S. et al. 2004. The biology and ecology of narrow endemic and widespread plants: a comparative study of trait variation in 20 congeneric pairs. – *Oikos* 107: 505–518.
- Lavelle, S. and Garnier, E. 2002. Predicting changes in community composition and ecosystem functioning from plant traits: revisiting the Holy Grail. – *Funct. Ecol.* 16: 545–556.
- Molina-Venegas, R. and Rodríguez, M. Á. 2017. Revisiting phylogenetic signal; strong or negligible impacts of polytomies and branch length information? – *BMC Evol. Biol.* 17: 53.
- Molina-Venegas, R. et al. 2017. Climatic and topographical correlates of plant palaeo- and neoendemism in a Mediterranean biodiversity hotspot. – *Ann. Bot.* 119: 229–238.
- Molina-Venegas, R. et al. 2020a. Phylogenetic patterns of extinction risk in the endemic flora of a Mediterranean hotspot as a guiding tool for preemptive conservation actions. – *Front. Ecol. Evol.* 8: 373.
- Molina-Venegas, R. et al. 2020b. Connecting plant evolutionary history and human well-being at Mt. Kilimanjaro, Tanzania. – *Bot. J. Linn.* 194: 397–409.
- Molina-Venegas, R. et al. 2022. Data from: Biogeographic deconstruction of phylogenetic and functional diversity provides insights into the formation of regional assemblages. – Figshare Digital Repository, <<https://doi.org/10.6084/m9.figshare.19222473>>.
- Ottaviani, G. et al. 2017. On plant modularity traits: functions and challenges. – *Trends Plant Sci.* 22: 648–651.
- Ottaviani, G. et al. 2020a. Linking plant functional ecology to island biogeography. – *Trends Plant Sci.* 25: 329–339.
- Ottaviani, G. et al. 2020b. The neglected belowground dimension of plant dominance. – *Trends Ecol. Evol.* 35: 763–766.
- Pagel, M. 1999. Inferring the historical patterns of biological evolution. – *Nature* 401: 877–884.
- Podani, J. and Schmera, D. 2006. On dendrogram-based measures of functional diversity. – *Oikos* 115: 179–185.
- Qian, H. et al. 2019. Phylogenetic dispersion and diversity in regional assemblages of seed plants in China. – *Proc. Natl Acad. Sci. USA* 116: 23192–23201.
- Rangel, T. F. et al. 2015. Phylogenetic uncertainty revisited: implications for ecological analyses. – *Evolution* 69: 1301–1312.
- Revell, L. J. 2012. phytools: an R package for phylogenetic comparative biology (and other things). – *Methods Ecol. Evol.* 3: 217–223.
- Romano, B. and Zullo, F. 2016. Half a century of urbanization in southern European lowlands: a study on the Po Valley (Northern Italy). – *Urban Res. Pract.* 9: 109–130.
- Royal Botanical Gardens Kew 2008. Seed Information Database (SID), Ver. 7.1. – <<http://data.kew.org/sid/>>.
- Saslis-Lagoudakis, C. H. et al. 2012. Phylogenies reveal predictive power of traditional medicine in bioprospecting. – *Proc. Natl Acad. Sci. USA* 109: 15835–15840.
- Schwery, O. et al. 2015. As old as the mountains: the radiations of the Ericaceae. – *New Phytol.* 207: 355–367.
- Slatyer, R. A. et al. 2013. Niche breadth predicts geographical range size: a general ecological pattern. – *Ecol. Lett.* 16: 1104–1114.

- Smyčka J et al. 2021. Tempo and drivers of plant diversification in the European mountain system. – Preprint at: <www.research-square.com/article/rs-959411/v1>.
- Swenson, N. G. and Weiser, M. D. 2010. Plant geography upon the basis of functional traits: an example from eastern North American trees. – *Ecology* 91: 2234–2241.
- Verdú, M. and Pausas, J. G. 2013. Syndrome-driven diversification in a Mediterranean ecosystem. – *Evolution* 67: 1756–1766.
- Weemstra, M. et al. 2021. Patterns in intraspecific variation in root traits are species-specific along an elevation gradient. – *Funct. Ecol.* 35: 342–356.
- Westoby, M. 1998. A leaf-height-seed (LHS) plant ecology strategy scheme. – *Plant Soil* 199: 213–227.
- Wiens, J. J. and Donoghue, M. J. 2004. Historical biogeography, ecology and species richness. – *Trends Ecol. Evol.* 19: 639–644.