RESEARCH ARTICLE

Allopatric speciation, niche conservatism and gradual phenotypic change in the evolution of European green lizards

Urtzi Enriquez-Urzelai[1,2] | Fernando Martínez-Freiría[1,3] | Inês Freitas[1,3] | Ana Perera[1] | Íñigo Martínez-Solano[4] | Daniele Salvi[1,5] | Guillermo Velo-Antón[1,3,6] | Antigoni Kaliontzopoulou[1,7]

[1]InBIO Laboratório Associado, Campus de Vairão, CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, Universidade do Porto, Vairão, Portugal

[2]Czech Academy of Sciences, Institute of Vertebrate Biology, Brno, Czech Republic

[3]BIOPOLIS Program in Genomics, Biodiversity and Land Planning, Campus de Vairão, CIBIO, Vairão, Portugal

[4]Departamento de Biodiversidad y Biología Evolutiva, Museo Nacional de Ciencias Naturales, CSIC, Madrid, Spain

[5]Department of Health, Life and Environmental Sciences, University of L'Aquila, L'Aquila, Italy

[6]Grupo de Ecoloxía Animal (Lab 97), Torre Cacti, Departamento de Ecoloxía e Bioloxía Animal, Universidade de Vigo, Vigo, Spain

[7]Department of Evolutionary Biology, Ecology and Environmental Sciences, and Biodiversity Research Institute (IRBio), Universitat de Barcelona, Barcelona, Catalonia, Spain

Correspondence
Urtzi Enriquez-Urzelai, InBIO Laboratório Associado, Campus de Vairão, CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, Universidade do Porto, 4485-661 Vairão, Portugal.
Email: urtzi.enriquez@gmail.com

Funding information
Fundação para a Ciência e a Tecnologia, Grant/Award Number: DL57/2016/CP1440/CT0010 and CEECIND/00937/2018; Ministerio de Economía y Competitividad; European Social Fund, Grant/Award Number: RYC2019-026688-I/AEI; Spanish State Research Agency; Ramón y Cajal, Grant/Award Number: RYC-2019-026959-I/AEI; European Union, Grant/Award Number: ES-TAF-5876, GB-TAF-5330, CZ-TAF-6587 and FR-TAF-6534; European Regional Development Fund

Handling Editor: Krystal Tolley.

Abstract

Aim: The contributions of historical biogeography, morphology and climatic niche evolution in shaping species diversification have been typically examined separately. To fill this gap, we assessed the relative role of geologic history, environment and phenotypic trait evolution in lineage diversification of green lizards in the Mediterranean biodiversity hotspot.

Location: Eurasia and North Africa.

Taxon: Green lizards (genera *Timon* and *Lacerta*).

Methods: For all green lizard lineages, we characterized distributional ranges and external morphological traits across discrete biogeographical areas, occupied macro-habitats and climatic niches using environmental variables that represent average and extreme climatic conditions. To assess the contribution of geographical factors in shaping diversity patterns, we evaluated the fit of 24 biogeographical models. We used BAMM and estimated phylogenetic signal to assess the rates of lineage diversification and of phenotypic and climatic niche evolution, and to determine whether these processes occurred steadily or at specific time periods as a response to palaeogeological or palaeoclimatic events. Finally, we tested for associations between phenotypic traits and lineage diversification using trait-dependent diversification analyses (QuaSSE, ES-sim and STRAPP).

Results: Biogeographical analyses favoured a dispersal–vicariance model explaining speciation patterns in green lizards, including jump dispersal and constrained dispersal by geographical distance. Lineages accumulated gradually towards the present, with minor divergence in morphological traits and conservatism of climatic niches. In contrast, in the *Lacerta agilis* lineage, niche evolution may have allowed expansion towards colder environments. Morphological and climatic niche evolution were uncoupled from diversification rates.

Main Conclusions: Biogeographical processes largely explain the constant lineage diversification of green lizards in the Mediterranean Basin since the Miocene, followed by gradual phenotypic divergence unrelated to cladogenesis. Climatic niche conservatism promoted the accumulation of lineages within the Mediterranean, except for *L. agilis*, where climatic niche evolution might underpin its range spread towards higher latitudes.

KEYWORDS
climatic niche evolution, dispersal, diversity hotspot, integrative biogeography, *Lacerta*, phylogenetic comparative methods, *Timon*

1

1 | INTRODUCTION

Understanding the mechanisms that generate and maintain biodiversity is an ambitious task for biogeographers and evolutionary ecologists (Keil & Chase, 2019; Owens et al., 2017; Pennington et al., 2010). A plethora of factors including different combinations of geologic, ecological, evolutionary, climatic and demographic processes simultaneously contribute to shape phenotypic and species diversification (Bars-Closel et al., 2017; Moen & Wiens, 2017). Geologic events determine the timing and span of connections and disconnections between landmasses that ultimately promote broad-scale patterns of migration and admixture, or lead to isolation and divergence of populations (Gorscak & O'Connor, 2016; Leprieur et al., 2016; Zhao et al., 2020). In addition, changes in landmass configuration, like the emergence of islands and the uplift of mountain ranges, result in drastic climatic, ecological and habitat-structure changes that can lead to the emergence of new environmental and structural niches (Hoorn et al., 2010; Price et al., 2014). Concurrently, climatic oscillations often lead to the isolation of populations, promoting allopatric divergence, especially in topographically heterogeneous regions (e.g. Martínez-Freiría, Freitas, et al., 2020; Velo-Antón et al., 2013). Likewise, ecological opportunity (e.g. generated by the creation of new niches) has been shown to boost diversification in a wide range of taxa (Badgley et al., 2017; Esquerré et al., 2019; Pincheira-Donoso et al., 2015).

These non-mutually exclusive processes often, but not always, also trigger phenotypic divergence of the diversifying taxa. While in some systems (islands or crater lakes), extensive morphological changes can result from rapid niche shifts associated with species radiations (Garcia-Porta et al., 2016; Kozak & Wiens, 2016), in continental systems different processes dictating the accumulation of new lineages and their phenotypic evolution are probably at play. Similar to instances of morphological divergence accompanying or not cladogenesis, the degree of divergence in climatic niche during lineage diversification varies widely across groups of organisms. As such, climatic niche divergence, frequently mediated by local adaptation to contrasting environmental conditions, has been proposed as an important mechanism of allopatric or parapatric speciation in some systems (Ahmadi et al., 2021; Graham et al., 2004). Conversely, climatic niche conservatism, together with regional climatic stability, has been frequently invoked to explain the accumulation of diversity in prominent biodiversity hotspots such as the Mediterranean Basin, by preventing climatic niche evolution and geographical range expansion (Martínez-Freiría, Freitas, et al., 2020; Skeels & Cardillo, 2017, 2019). Because the aforementioned mechanisms and processes have been typically examined independently (i.e. focusing on historical biogeography, functional morphology or climatic niche evolution separately), the interplay among them and how they conjointly shape species diversification and evolution remains elusive.

To fill this gap, here we assess the relative role of geologic and environmental factors and phenotypic trait evolution in the diversification of green lizards (genera *Lacerta* and *Timon*) around the Mediterranean Basin (Figure 1), one of the global biodiversity hotspots (Myers et al., 2000). The diversification of green lizards is framed in a scenario of profound paleogeographic events that affected the Mediterranean basin since the Neogene (~23–2.5 million years ago [Mya]; Appendix S3). These include a gradual retraction of the Tethys sea, the closing of the Arabian Seaway (connecting the African-Arabian plate to Asia Minor), the closing (and later reopening) of the Gibraltar Strait connecting North Africa to the Iberian Peninsula during the Messinian Salinity Crisis and the uplift of Irano-Anatolian mountains (Ahmadi et al., 2021; Gómez & Lunt, 2015; Popov et al., 2006; Prista et al., 2015; Rögl, 1998; Steininger & Rögl, 1984). During this period, the Mediterranean underwent drastic and repeated global climate changes with associated habitat modifications, until the Pleistocene glacial cycles (Cavazza & Wezel, 2003; Hewitt, 2000; Jiménez-Moreno et al., 2010). Green lizards exhibit relatively high diversity of lineages in the Mediterranean and extensive phenotypic variation (particularly in body size: Arnold et al., 2007). As such—and given the complex geologic and climatic history of the Mediterranean Basin—they provide an excellent study system to assess the relative role of geology, climate and phenotypic evolution in determining lineage diversification and biogeographical patterns.

To this end, we first reconstructed the biogeographical history of green lizards and estimated rates of lineage diversification along the evolutionary history of the clade to test whether lineage diversification proceeded at a relatively constant rate through time or if it peaked at specific time periods (e.g. as a response to palaeogeological or palaeoclimatic events). Although the pattern of mostly parapatric distributions shown by green lizards suggests a scenario of climatic niche conservatism with little or no ecological shifts (i.e. in relation to microhabitat use; Reaney et al., 2018), previous studies pointed to a combination of climatic niche conservatism and divergence (Ahmadzadeh et al., 2013, 2016) underlying their diversification. Moreover, the morphological variation within green lizards is suggestive of variation in ecological niche. Therefore, to test for conservatism or divergence in morphology (associated to trophic/structural niche) and climatic niche traits, we evaluated their degree of phylogenetic signal. Finally, to test whether speciation and phenotypic evolution are coupled, we estimated rates of phenotypic macroevolution and tested for associations between phenotypic traits and rates of lineage diversification.

2 | MATERIALS AND METHODS

2.1 | Species data

2.1.1 | Phylogenetic data

We used the multilocus time-calibrated phylogeny of green lizards inferred by Adams et al. (2020). We collapsed all branches that diverged <2.5 Mya in the time-calibrated phylogeny (following Martínez-Freiría, Toyama, et al., 2020), obtaining a final tree with 26 tips. The decision to collapse nodes younger than 2.5 Mya was made to avoid including in the phylogeny most of the intraspecific
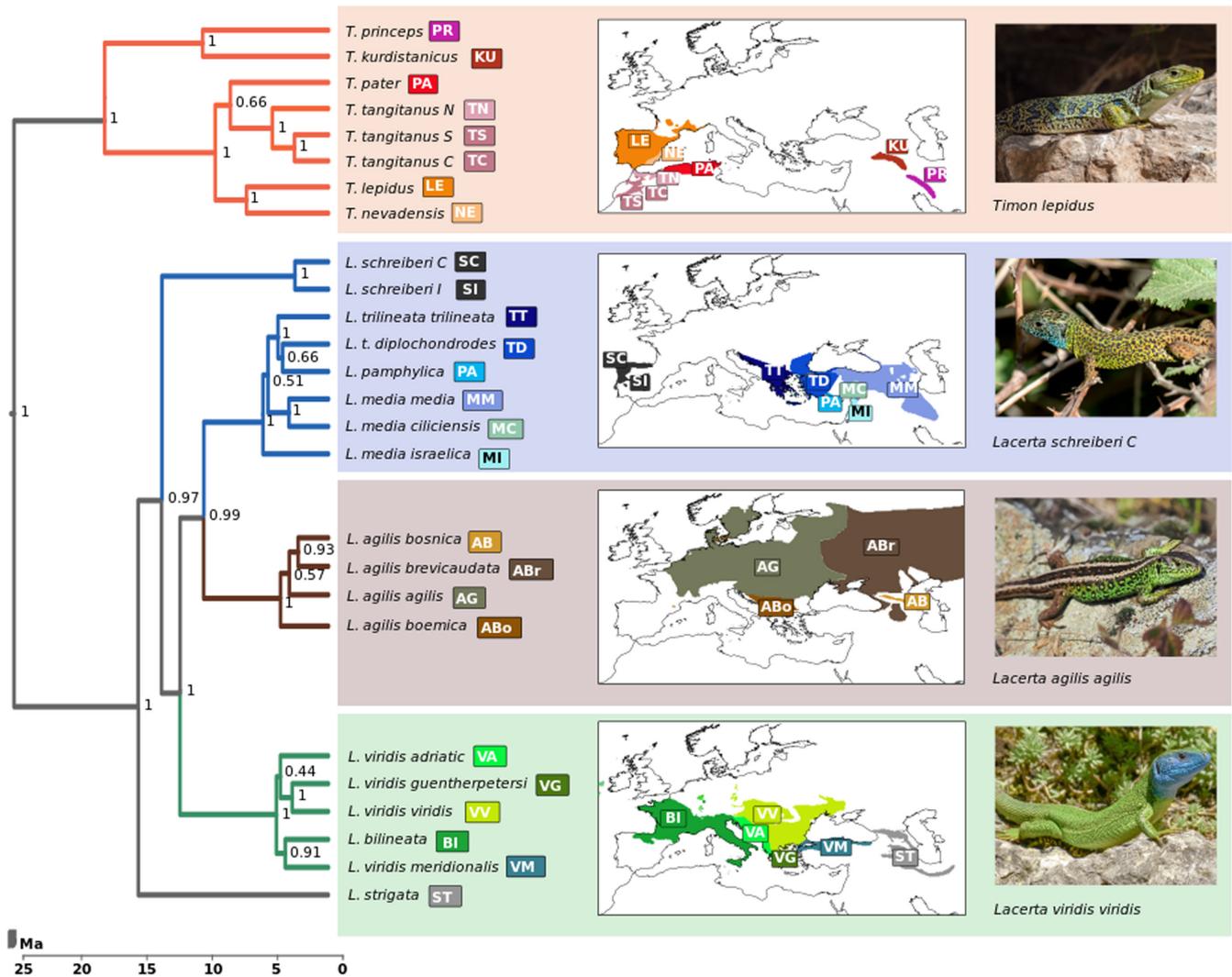
FIGURE 1 Phylogenetic relationships among *Lacerta* and *Timon* species considered in this study, and corresponding distribution ranges in Eurasia and North Africa. The phylogenetic tree corresponds to the tree obtained after collapsing recent branches (2.5 Mya).

phylogenetic variation, and restrict our analyses to evolutionary events that occurred before the Pleistocene. This allowed us to exclude recent lineages that appeared after the Mediterranean Basin reached its present configuration, and that represent population—rather than species-level evolutionary divergence (Martínez-Gil et al., 2021). Details on all procedures applied for phylogenetic inference can be found in Adams et al. (2020). We used the maximum clade credibility tree to reconstruct historical biogeography, and to investigate the processes involved in lineage diversification and phenotypic divergence (see below). However, whenever computationally feasible, we repeated analyses over 1000 phylogenetic trees randomly drawn from the posterior distribution of BEAST runs from Adams et al. (2020), to account for phylogenetic uncertainty.

2.1.2 | Morphological data

To describe morphological variation among green lizards, we measured museum vouchers of most species/lineages retrieved in the phylogenetic tree (see exceptions below). Specifically, we measured snout–vent length (SVL; a proxy of size), trunk length (TRL), forelimb length (FLL), hindlimb length (HLL), head length (HL), head width (HW) and head height (HH). With the three head measurements (HL, HW and HH), we computed head size (HS) as the cubic root of their product (Kaliontzopoulou et al., 2012; Mosimann, 1970). We only considered morphological measurements of male specimens, to circumvent any confounding factor arising from sexual dimorphism. Furthermore, to account for sampling bias, we retained the 10 largest individuals from each lineage (or mixture of lineages for collapsed branches, see Figures S1.1, S1.2) for data analyses (Table S2.1). We size-corrected all morphological measurements (except for SVL) by extracting the residuals from reduced major axis regressions between each variable and SVL, using the *sma* function of the 'smatr' R-package v3.4-8 (Warton et al., 2012). We log-transformed all morphological traits before analyses. Because morphological data were not available for *Lacerta agilis boemica*, *L. media ciliensis* and *L. viridis guentherpetersi*, we excluded these three taxa from all downstream analyses involving morphology.

2.1.3 | Distribution, climatic niche and macrohabitat data

To characterize the climatic niche of each lineage, we obtained raw distribution ranges for each nominal species from IUCN (2018). Subsequently, we divided species ranges with GIS software (QGIS, 2020) to match intraspecific lineages as defined by published phylogeographical studies (see references; Figure 1). To describe the climatic niche of each lineage, we used seven environmental variables that represent average and extreme climatic conditions (Martínez-Freiría, Toyama, et al., 2020) at a spatial resolution of 20km^2 downloaded from WorldClim v2 (Fick & Hijmans, 2017; http://worldclim.org/version2): average temperature (T_{avg}), maximum temperature (T_{max}), minimum temperature (T_{min}), water vapour pressure (VAPR), precipitation (PREC), solar radiation (SRAD) and elevation (ELEV). Following Martínez-Freiría, Toyama, et al. (2020), we extracted the central 95% values within each distribution range for each variable to minimize the risk of including extreme environmental values from areas in which species might not be present. With those values, we calculated average (T_{avg}, VAPR$_{avg}$, PREC$_{avg}$ and SRAD$_{avg}$), maximum (T_{max}, VAPR$_{max}$, PREC$_{max}$ and SRAD$_{max}$) and minimum monthly environmental conditions (T_{min}, VAPR$_{min}$, PREC$_{min}$ and SRAD$_{min}$), as well as average, maximum and minimum elevations for each lineage (ELEV$_{avg}$, ELEV$_{max}$ and ELEV$_{min}$). Environmental variables were obtained using the 'raster' R-package v3.5-2 (Hijmans, 2020). See Appendix S1 for additional details and Table S2.2 for a summary of values of these variables per lineage.

To characterize the general (macro)habitat occupied by each lineage, we overlaid estimated distribution ranges with biome information from the Ecoregions2017©Resolve initiative (ecoregions2017.appspot.com; Olson et al., 2001). Then, we calculated the proportion of each lineage distribution in each biome and identified the predominant one (i.e. the biome accounting for the highest proportion of the distribution of each lineage). All lineages predominantly (>70% in most cases) occupied one of the three following biomes: temperate Grasslands–Savannas–Shrublands (2 lineages), temperate broadleaf mixed forests (11 lineages) or Mediterranean Forests–Woodlands–Scrub (13 lineages), although in some cases the distribution across biomes was more even (Table S2.2).

2.2 | Historical biogeography

We reconstructed the biogeographical history of the group using the 'BioGeoBEARS' R-package v1.1.2 (Matzke, 2013; https://github.com/nmatzke/BioGeoBEARS), based on the time-calibrated phylogeny and the geographical range of each lineage across discrete geographical areas. We defined seven biogeographical regions representing landmasses that either connected/disconnected from each other during the geologic history of the Mediterranean Basin or acted as major refugia during Pleistocene climatic oscillations: Iberian Peninsula (I), Italian Peninsula (T), Balkan Peninsula (B), Asia Minor (A), Arabian Peninsula (R), North Africa (N) and central and northern Europe (C). We considered a lineage to occur in a given area when at least 10% of

its distribution included that area. Given that this threshold is somewhat subjective, we also used 20% and 30% of species' distributions as alternative thresholds to assess the congruence in results.

We considered a total of 24 models, based on the three main biogeographical models implemented in BioGeoBEARS: (i) Dispersal–Extinction–Cladogenesis (DEC), (ii) a likelihood-based DIspersal–VicAriance model (DIVALIKE) and (iii) a likelihood implementation of BAYAREA (BAYAREALIKE). These models differ in the biogeographical processes considered (see Matzke, 2013 for a synopsis of the processes implemented in each model). Biogeographical reconstructions under DEC are modelled considering dispersal, extinction, (narrow—if the ancestor lives in a single area, daughter lineages inherit the ancestral range and subset—if the ancestor is widespread, one daughter lineage will inherit a subset of this area; Matzke, 2013), sympatric speciation and (narrow) vicariance. Under DIVALIKE, dispersal, extinction, (narrow) sympatric speciation and (both narrow and widespread) vicariance are considered. BAYAREALIKE considers dispersal, extinction and exclusively (narrow and widespread) sympatric speciation. It is worth noting that dispersal, in this case, is not related to speciation but to a process of range evolution. In addition, we fitted all three models including jump-dispersal, linked to speciation (i.e. speciation by long distance colonization, +J models; Matzke, 2014). Then we included in the resulting six models a matrix of geographical distances between areas to constrain dispersal (distance-based dispersal, +x, models; van Dam & Matzke, 2016). Finally, we repeated analyses with all 12 models (with and without geographical distances between areas) accounting for the palaeogeographical history of the region (time-stratified analyses; +palaeo models). We considered eight different time periods from the present to 30Mya that match with the important paleogeographical and paleoclimatic periods outlined in the introduction: for each we coded which areas were connected or disconnected (Appendix S3). We compared the fit of all models based on AICc and Akaike weights (Akaike, 1978, 1979; Burnham & Anderson, 2004).

2.3 | Diversification analyses

2.3.1 | Lineage and phenotypic diversification

We used the time tree of green lizards generated by Adams et al. (2020) to explore whether rates of lineage diversification varied along time (from the origin of the group until 2.5 Mya). We applied three widely used methods (Appendix S4), although we only present results from BAMM v2.5 (Rabosky, 2014) in the main text. BAMM aims at identifying heterogeneity in diversification rates through time by simultaneously exploring a multitude of candidate models, using reversible-jump Markov-chain Monte Carlo (MCMC). To test for rate shifts in lineage diversification, as expected if the major geological and climatic events that occurred in the Mediterranean have shaped cladogenesis in green lizards, we ran 20 million generations in four MCMC chains with δT set to 0.01 and a swap period of 1000 generations. We sampled event data every 10,000 generations and discarded the first 10% of the samples as 'burn-in'.

To obtain a descriptive overview of whether phenotypic traits are conserved (i.e. more similar between closely related species than expected by chance), we quantified phylogenetic signal for each trait. We used Blomberg's *K* (Blomberg et al., 2003) and a randomization procedure to evaluate the strength and significance of phylogenetic signal. Significance values (i.e. *p*-values) were corrected using the false discovery rate procedure for multiple testing (Benjamini & Hochberg, 1995). A significant Blomberg's *K* statistic with a value of 1 implies that species are as similar as expected given a Brownian motion model of phenotypic evolution. Values of *K* lower or higher than 1 indicate that closely related species are more different or more similar than the null expectation, respectively (Blomberg et al., 2003). To test whether *K* values significantly differed from 1, we compared empirical values against a null distribution for *K* = 1.0 via simulation on our phylogeny. Specifically, we counted the number of simulated *K* values more extreme than observed values (based on absolute, logarithmic *K* values) following code available at http://blog.phytools.org/2011/12/testing-for-phylogenetic-signal-k.html. We also performed phylogenetic signal tests with 1000 trees to account for phylogenetic uncertainty. To identify changes in rates of phenotypic diversification along time, we used BAMM (Rabosky, 2014). For each phenotypic trait, we ran 200 million generations in four MCMC chains with δT set as 0.01 and the swap period as 1000 generations. We sampled every 5000 generations and discarded the first 50% of the samples as 'burn-in' (following Folk et al., 2019).

2.3.2 | Linking phenotypic and environmental traits with lineage diversification

To test for associations between phenotypic traits (morphology and climatic niche) and rates of lineage diversification, we employed three methods (see Appendix S1 for details). QuaSSE (quantitative state speciation and extinction; Fitzjohn, 2010) models diversification as a birth–death process in which speciation (λ) and extinction rates (μ) can be constant, linear, sigmoidal or hump-shaped functions of a quantitative trait. For each phenotypic trait, we fitted models representing all the different potential relationships between traits and speciation using the 'diversitree' R-package v0.9-16 (Fitzjohn, 2012) and we compared models using likelihood ratio tests. ES-sim (inverse of equal-splits with simulated null model) tests for trait-dependent diversification by measuring tip-specific speciation rates and simulating a null distribution under a given model of trait evolution (Harvey & Rabosky, 2018). We ran 10,000 simulations assuming Brownian motion and used Pearson's correlation to determine significant relationships. Finally, STRAPP (structured rate permutations on phylogenies; Rabosky & Huang, 2016) tests for association between phenotypic traits and diversification rates as estimated by BAMM. This method compares empirical values to a null distribution generated by permuting the diversification rates across the phylogeny. We correlated BAMM diversification rates and phenotypic traits by performing two-tailed STRAPP tests with 10,000

replicates and using Spearman's rank correlation coefficients with the 'BAMMtools' R-package v2.1.8 (Rabosky et al., 2014).

To test for an association between habitat and macroevolutionary rates, we used MuSSE (multivariate state speciation and extinction; Fitzjohn, 2012) and QuaSSE (Fitzjohn, 2010). MuSSE allows speciation (λ), extinction (μ) and transition (q) between states (the three predominant habitat types, in our case) to be either independent of (equal rates across habitats) or dependent on the habitat (separate rates for each habitat). We fitted several MuSSE models, recovered the maximum likelihood estimate of associated parameters and then compared models using likelihood ratio tests. We also analysed the effect of habitat type on diversification rates employing QuaSSE. Contrary to MuSSE, to fit QuaSSE models, we used data for each habitat type separately (i.e. considering the percentage of each lineage's distribution on each of the three habitat types). We fitted models representing all the potential relationships between traits and speciation (either constant, linear, sigmoidal or hump-shaped functions) and compared them using likelihood ratio tests with the 'diversitree' R-package (Fitzjohn, 2012). See Appendix S1 for further details.

3 | RESULTS

3.1 | Historical biogeography

BioGeoBEARS analyses favoured the dispersal–vicariance (DIVALIKE) model including jump dispersal (+J) and distance-based dispersal (constrained dispersal due to geographical distance between areas; +x). The next most supported models were the DEC+J+x model, and DIVALIKE and DEC models including +J +x and palaeogeographical information (+palaeo; Table 1; Table S4.1). While the four best models were not statistically distinguishable from one another in terms of AICc, the best model (i.e. DIVALIKE +J +x) clearly outperformed the other models in terms of AICc weights. Increasing thresholds to consider a species present in a given area (i.e. 10%, 20% or 30%) decreased the support difference between models (Table 1; Table S4.2) and slightly changed biogeographical reconstructions (Figures S4.1–S4.4). However, DIVALIKE +J +x always received the highest support regardless of the cut-off value used to

TABLE 1 Summary statistics for historical biogeography models with strongest support implemented in BioGeoBEARS

Model	LnL	k	AICc	AICc weight
DIVALIKE +J +x	−61.220	4	132.345	0.645
DEC +J +x	−62.648	4	135.201	0.155
DIVALIKE +J +x +palaeo	−63.143	4	136.191	0.094
DEC +J +x +palaeo	−63.330	4	136.566	0.078

Note: AICc: Akaike's Information Criterion for small sample sizes, AICc weight: relative model probability; Ln*L*: data likelihood, *k*: number of parameters. See Table S4.1 for summary statistics of all implemented models.

assign species to areas. This result indicates that the most important biogeographical processes during the diversification of green lizards were vicariance and dispersal (including jump dispersal) limited by geographical distance.

3.2 | Lineage and phenotypic diversification

BAMM did not support changes in diversification rates along time (Figure 2), with posterior probabilities and Bayes Factors (BFs)

supporting equal diversification rates among clades (0 shifts: posterior probability = 0.885; BF <0.207 in all cases; Table S4.5).

Considering morphological variables, we only found significant phylogenetic signal for body size (SVL) and relative head size (rHS). By contrast, we detected significant phylogenetic signal for most niche-related variables. Among these, we did not find significant differences between observed K values and the expected value of $K = 1$, suggesting that closely related lineages are as similar as expected under a Brownian Motion process and given the phylogeny (Table 2). These results are robust to phylogenetic uncertainty (Table S4.7).
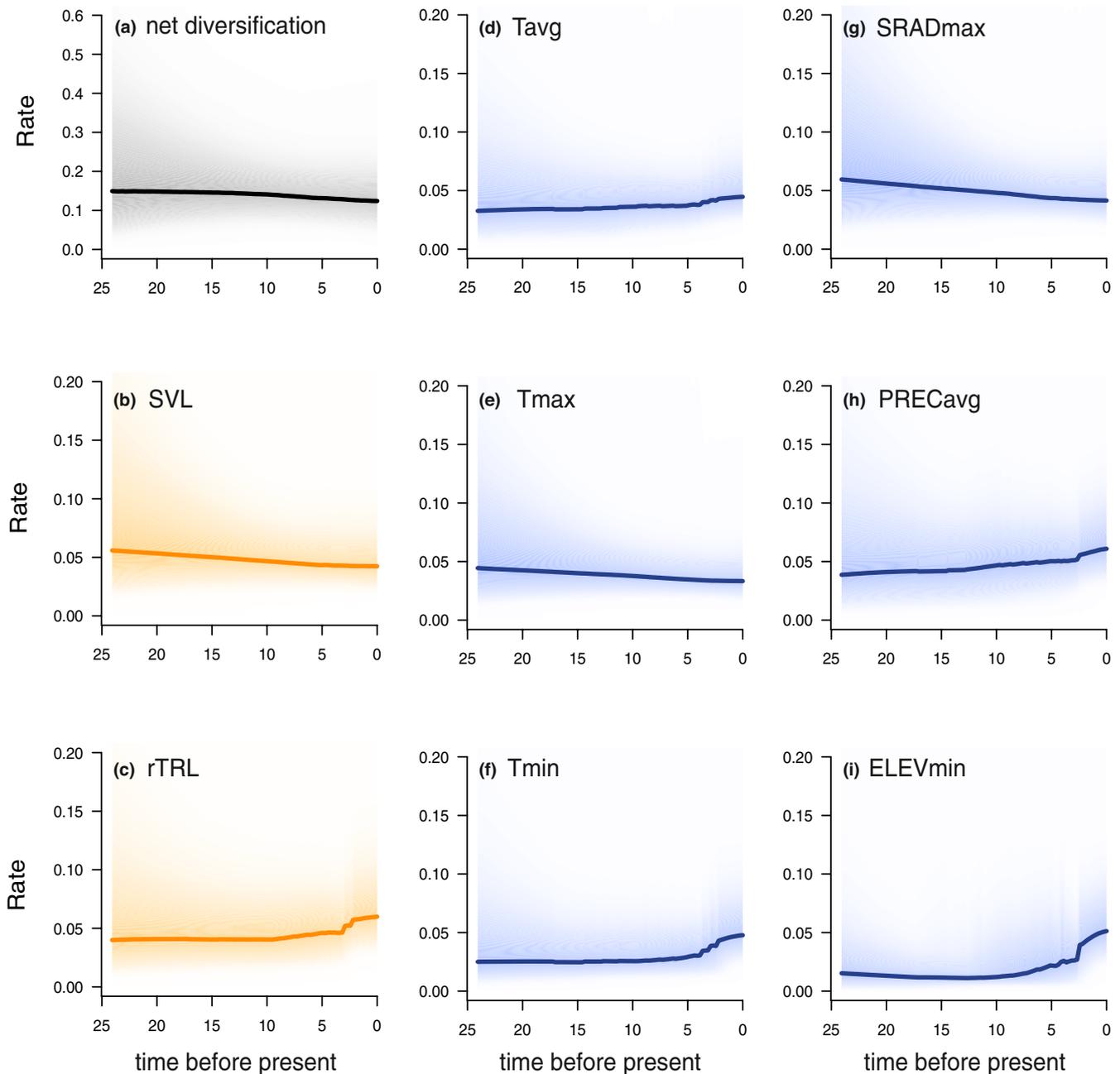

FIGURE 2 (a) Net diversification rate through time and (b–i) phenotypic macroevolutionary rates for all green lizard (*Lacerta* and *Timon*) lineages. (b, c) represent morphological diversification and (d–i) climatic niche trait diversification. Note that phenotypic macroevolutionary rates for each phenotypic trait have been scaled to range from 0 to 1.

TABLE 2 Phylogenetic signal as measured by Blomberg's K, their associated p-values before and after false discovery rate (FDR) correction, and the p-values of Blomberg's K differing from one for each phenotypic (morphological and climatic niche) trait, based on the maximum credibility tree

Phenotypic trait	Blomberg's K	p-value	Corrected p-value	$K \neq 1$ p-value
Morphology				
SVL	**1.093**	**0.001**	**0.005**	0.855
rTRL	0.719	0.975	0.976	0.508
rHS	**0.797**	**0.015**	**0.043**	0.622
rFLL	0.755	0.970	0.976	0.577
rHLL	0.575	0.253	0.422	0.293
Climatic niche				
T_{avg}	**1.173**	**0.002**	**0.005**	0.753
T_{max}	**1.216**	**0.001**	**0.002**	0.693
T_{min}	**1.082**	**0.002**	**0.003**	0.880
$SRAD_{avg}$	**1.149**	**0.001**	**0.002**	0.797
$SRAD_{max}$	**1.282**	**0.001**	**0.002**	0.652
$SRAD_{min}$	**0.678**	**0.003**	**0.003**	0.495
$PREC_{avg}$	**1.358**	**0.001**	**0.002**	0.564
$PREC_{max}$	**0.655**	**0.011**	**0.013**	0.415
$PREC_{min}$	**0.958**	**0.001**	**0.002**	0.943
$VAPR_{avg}$	0.810	0.362	0.395	0.700
$VAPR_{max}$	**0.918**	**0.004**	**0.006**	0.882
$VAPR_{min}$	0.807	0.527	0.527	0.710
$ELEV_{avg}$	**0.991**	**0.004**	**0.006**	0.992
$ELEV_{max}$	**1.232**	**0.002**	**0.002**	0.713
$ELEV_{min}$	**0.702**	**0.001**	**0.002**	0.502

Note: Significant values are highlighted in bold font. See Table S4.7 for analyses incorporating phylogenetic uncertainty.

The diversification of most morphological traits seemed steady, but diversification rates of SVL and trunk length (rTRL) changed over time, according to BAMM analyses (Figure 2; Figure S4.7). While diversification slowed down towards the present in SVL, rTRL showed a sharp increase in diversification rates ~3 Mya (Figure 2). Furthermore, a scenario encompassing an increase in diversification rates of rTRL in the clade of *L. agilis*, as compared to the background, received support of 53.4% of the posterior distribution (44.0% + 9.4%; Figure S4.7). BFs, however, did not favour models with higher rates in the *L. agilis* clade over models with equal rates among clades (BF <4 in all cases; Table S4.5). We also found rate heterogeneity in niche traits over time (Figure 2; Figure S4.8). T_{max} and $SRAD_{max}$ diversification rates decreased homogeneously towards the present, and rates for T_{avg}, T_{min}, $PREC_{avg}$ and $ELEV_{min}$ increased 3–5 Mya (Figure 2). For T_{avg}, diversification rates increased in the branch leading to the *L. agilis* clade (32.6% of the posterior) or, less likely, in the branch leading to the *L. agilis–media–trilineata* clade (7.2% of the posterior); however, BFs did not support different rates between clades (BF <4 in all cases). Different evolutionary rates between clades for $PREC_{avg}$

were also rejected (BF <8 in all cases). Finally, Tmin diversification rates were significantly faster in the *L. agilis* clade, as supported by both the posterior distribution (77.3%) and BFs; models with one shift leading to faster evolutionary rates within *L. agilis* lineages received moderate support (BF = 10.015), and two or more shifts stronger support (BF = 14.710). We also found support for a shift in diversification rates for $ELEV_{min}$ (BF = 18.815) leading to faster rates in the *L. schreiberi* and *T. tangitanus* clades (50.8% of the posterior).

3.3 | Linking phenotypic and environmental traits with lineage diversification

All three implemented approaches provided negligible support for any association between phenotypic/environmental traits and lineage diversification (Tables S4.8–S4.10). Although QuaSSE analyses identified associations of speciation rates with one morphological (rFLL) and several climatic niche variables (T_{max}, $PREC_{max}$, $ELEV_{avg}$), ES-Sim and STRAPP analyses yielded non-significant results for all morphological (ES-Sim: $p > 0.583$ and STRAPP: $p > 0.994$ in all cases) and climatic niche traits (ES-Sim: $p > 0.140$ and STRAPP: $p > 0.991$ in all cases). Similarly, the habitat occupied by lineages did not affect lineage diversification rates, according to MuSSE (LRT against the model with all rates equal across habitats: $p > 0.233$) and QuaSSE analyses (LRT: $p > 0.340$ in all cases).

4 | DISCUSSION

Integrative studies combining geographical, morphological, climatic niche and phylogenetic data can provide key insights on the major drivers of species diversification patterns and the underlying processes (Cavender-Bares et al., 2011; Gajdzik et al., 2019; Pato et al., 2019). The scarcity of such studies rests upon the difficulty in gathering all independent sources of information for a given taxonomic group. By integrating these data at the macro-evolutionary level in a phylogenetic comparative framework, we show that the diversification of green lizards was dominated by the biogeographical processes of dispersal and vicariance, and that morphological and climatic niche divergence did not impact on lineage diversification rates. These inferences are essential to understand the processes involved in shaping biodiversity patterns in the Mediterranean biodiversity hotspot.

4.1 | Lineage diversification across space and time

Our results indicate that dispersal into unoccupied areas and allopatric speciation have largely driven the diversification of green lizards (as inferred by the DIVALIKE +J +x model; Table 1). Jump-dispersal (+J) and notably distance-based dispersal (+x models) also stood out as important biogeographical processes, outlining the importance of physical geography (e.g. landmass configuration) as a major driver of

biodiversity patterns (Qian & Ricklefs, 2000; Sandel et al., 2020; van Dam & Matzke, 2016). It is worth noting that 'dispersal into unoccupied areas' refers to the colonization of areas without any green-lizard species represented in our phylogeny. Of course, our analyses are blind to any other (e.g. from other genera, or now extinct) species that may have been present in those areas. However, that is impossible to know without fossil data, which is quite scarce and scattered for the group.

This is exemplified by some events associated with the biogeographic history of the genus *Timon*. According to the favoured model (DIVALIKE +J +x), the most plausible biogeographical scenario suggests that the most recent common ancestor (MRCA) of *Timon* was widespread across North Africa and the Arabian plate, and later the MRCA of Iberian *Timon* species may have dispersed from North Africa to the Iberian Peninsula (Figure S4.1). Our biogeographical reconstruction, together with the time-calibrated phylogeny, dates dispersal through the Strait of Gibraltar in the Tortonian (9 Mya; 95 highest posterior density intervals: 10.5–7 Mya), some million years before the Messinian Salinity Crisis, MSC (Figures S1.1 and S1.2; see also Garcia-Porta et al., 2019; Paulo et al., 2008). Therefore, the colonization of Europe by *Timon* might have occurred through over-seas dispersal across one or multiple narrow sea corridors that were in place during the Tortonian (Betic Corridor, Guadalhorce Corridor, Rif Corridor; see Paulo et al., 2008; Rosenbaum et al., 2002). We acknowledge that divergence of *Timon* in north Africa prior to dispersal towards Europe might account for a split older than the colonization of Europe. But in this case, we would need to assume the occurrence of two old (>7 Mya) lineages in Africa, one of which colonized Europe and subsequently became extinct on the African continent. This scenario is less parsimonious than a scenario where the divergence between the two lineages (European and African) followed the colonization of Europe from Africa. Furthermore, a scenario of over-seas dispersal before the MSC has been also inferred for *Psammodromus* lizards (Mendes et al., 2017) and other organisms with low dispersal ability such as beetles (Mas-Peinado et al., 2021), worm lizards and salamanders (see Hewitt, 2011; Mendes et al., 2017 for a review).

Lineage diversification analyses did not support shifts in diversification rates at any time period along the evolutionary history of green lizards, or in association with any particular geologic, climatic or dispersal event (Figure 2a; Figures S4.5 and S4.6). The lack of association between diversification rates and dispersal events is quite surprising given that our biogeographical reconstructions show that the diversification of green lizards was dominated by dispersal into previously unoccupied areas. Often, the colonization of novel areas leads to bursts of diversification, associated with access to empty niches (Miller et al., 2018; Schenk et al., 2013; Schenk & Steppan, 2018). However, phenotypic constraints (e.g. low evolvabilities; Losos, 2010) or the presence of distantly related but ecologically similar competitors could have impeded green lizards to expand into new niches (Martínez-Freiría et al., 2008; Martínez-Freiría, Freitas, et al., 2020).

4.2 | Phenotypic macroevolution and links to lineage diversification

Our results show that morphological evolution is not coupled with lineage diversification in green lizards. Although several evolutionary theories support the idea that phenotypic evolution occurs either as a burst in the early history of a clade (e.g. ecological theory of adaptive radiations: Gillespie et al., 2020; Yoder et al., 2010) or in association with speciation events (punctuated equilibria: Gould & Eldredge, 1977), green lizards do not conform to these models. Similarly, we show that climatic niche evolution and macro-habitat use did not impact significantly on diversification rates in green lizards (Tables S4.9 and S4.10). However, the methods used to test for associations between phenotypic traits and diversification rates often suffer from low statistical power when implemented on small phylogenies. Thus, given the weak support for the role of several climatic niche variables in diversification (i.e. T_{max}, $PREC_{max}$, $ELEV_{avg}$) and the results of recent studies in subsets of the group (Ghane-Ameleh et al., 2021), we cannot completely exclude the possibility that climatic niche evolution may have contributed to speciation dynamics of this group during climatic oscillations.

Regarding morphological macroevolution, we found that the evolutionary rate of body size (SVL) decreased gradually through time (Figure 2). This possibly led to body sizes as similar as expected given shared evolutionary history (Table 2). The fact that all extant green lizard species, despite occupying a variety of habitats, are ground-dwellers, could partly explain this result (Arnold et al., 2007; Collar et al., 2011; Velasco et al., 2020). Likewise, among-lineage differences in head sizes were as expected given the phylogeny (Table 2). Since the head is involved in a variety of highly relevant ecological tasks such as feeding, refuge use, mating and male aggressive interactions (Gomes et al., 2016; Kaliontzopoulou et al., 2012), the gradual divergence in head dimensions could reflect minor trophic or structural niche divergence. The evolutionary rTRL, a trait related to manoeuvrability and climbing ability, among other functions (Žagar et al., 2017), peaked ~3 Mya for the whole group. However, and although rTRL did not show significant phylogenetic signal, its value did not result significantly different to that expected given the phylogeny. Thus, it also points out to minor structural niche divergence. As a cautionary note, we would like to add that although for neutrally evolving traits the link between evolutionary rates and phylogenetic signal might be direct, the relationship might not be as straightforward for traits under selection. Empirical data are a mix of traits evolving under selection (i.e. where trait value depends on the trait optimum and the speed of evolution towards it) and neutrally (i.e. where trait value depends on ancestral state and evolutionary rate). Since we have no means of evaluating which traits are evolving under selection or neutrally, we admit that our argument assumes neutrally evolving traits. Altogether, and although morphological divergence in concert with geographical expansion have been hypothesized to drive continued diversification (Kennedy et al., 2018; Ramírez-Barahona et al., 2016), our results suggest that the steady

diversification of green lizards occurred mainly due to geographical expansion, but unrelated to morphological evolution (Tables S4.8 and S4.10; see also Lee et al., 2016).

We also show that a combination of climatic niche conservatism and divergence played a central role in shaping their diversity around the Mediterranean hotspot. Previous studies on specific green lizard clades produced mixed results: while in the *Lacerta trilineata–pamphylica–media* group and the western *Timon* clade (*lepidus–nevadensis–tangitanus–pater*) climatic niches have diverged (Ahmadzadeh et al., 2013, 2016), they are conserved in the eastern *Timon princeps–kurdistanicus* clade (Ahmadzadeh et al., 2016). According to our phylogenetic signal estimates (Table 2), green lizards as a whole show conserved climatic niches (i.e. niche traits as similar as expected given a Brownian model of phenotypic evolution). According to BAMM, rates of maximum temperature and solar radiation diversification of green lizards decreased towards the present, again supporting conserved thermal niches, and plausibly linked to the observed decrease in body size evolutionary rates during a period of general global cooling (Burke et al., 2018). Furthermore, rates for T_{avg}, T_{min}, $PREC_{avg}$ and $ELEV_{min}$ increased 3–5 Mya ago and, remarkably, rates of T_{min} evolution seemed faster in the *L. agilis* group, which likely facilitated the spread of these lineages into colder environments (e.g. higher latitudes in Europe and Asia; Figure 1; Figure S4.1). These results agree with our hypothesis that, while climatic niche conservatism promotes the build-up of biodiversity in Mediterranean type ecosystems, rapid niche evolution allows transitions away from hotspots (Dinis et al., 2019; Martínez-Freiría, Freitas, et al., 2020; Rato et al., 2015; Skeels & Cardillo, 2017, 2019).

4.3 | Conclusion

Integrative studies combining phylogenetic, geologic, phenotypic, climatic and ecologic data are key to explaining how biodiversity hotspots such as Mediterranean type ecosystems are formed. Using a series of integrative analyses to link different sources of variation we show that biogeographical processes (i.e. dispersal events followed by allopatric divergence) largely governed the diversification of green lizards around the Mediterranean Basin. This was followed by minor morphological divergence not correlated with cladogenesis, which emerges as a recurrent pattern in continental systems (Adams et al., 2009; Ashman et al., 2018; Garcia-Porta et al., 2017; Lee et al., 2016). Furthermore, this study corroborates that a combination of climatic niche conservatism and divergence contributes to explain patterns of diversity in Mediterranean type ecosystems (Dinis et al., 2019; Martínez-Freiría, Freitas, et al., 2020; Rato et al., 2015; Skeels & Cardillo, 2017, 2019). In agreement with our expectations, our results suggest that the interplay between climatic niche conservatism/divergence and minor changes in functional morphology (and thus trophic and structural niche) led to the pattern of mostly parapatric distributions, centred around the Mediterranean hotspot of diversity. We stress the need for additional integrative studies to

better characterize and understand global biodiversity patterns and their underlying processes.

ORCID

Urtzi Enriquez-Urzelai https://orcid.org/0000-0001-5958-2250
Fernando Martínez-Freiría https://orcid.org/0000-0003-2311-8960
Inês Freitas https://orcid.org/0000-0001-9984-9378
Íñigo Martínez-Solano https://orcid.org/0000-0002-2260-226X
Daniele Salvi https://orcid.org/0000-0002-3804-2690
Guillermo Velo-Antón https://orcid.org/0000-0002-9483-5695
Antigoni Kaliontzopoulou https://orcid.org/0000-0002-7897-7204

REFERENCES

Adams, D. C., Berns, C. M., Kozak, K. H., & Wiens, J. J. (2009). Are rates of species diversification correlated with rates of morphological evolution? *Proceedings of the Royal Society B: Biological Sciences*, 276(1668), 2729–2738. https://doi.org/10.1098/rspb.2009.0543

Adams, D. C., Glynne, E., & Kaliontzopoulou, A. (2020). Interspecific allometry for sexual shape dimorphism: Macroevolution of multivariate sexual phenotypes with application to Rensch's rule. *Evolution*, 74(9), 1908–1922. https://doi.org/10.1111/evo.14049

Ahmadi, M., Hemami, M. R., Kaboli, M., Nazarizadeh, M., Malekian, M., Behrooz, R., Geniez, P., Alroy, J., & Zimmermann, N. E. (2021). The legacy of eastern Mediterranean mountain uplifts: Rapid disparity of phylogenetic niche conservatism and divergence in

mountain vipers. *BMC Ecology and Evolution*, 21(1), 1–13. https://doi.org/10.1186/s12862-021-01863-0

Ahmadzadeh, F., Flecks, M., Carretero, M. A., Böhme, W., Ihlow, F., Kapli, P., Miraldo, A., & Rödder, D. (2016). Separate histories in both sides of the Mediterranean: Phylogeny and niche evolution of ocellated lizards. *Journal of Biogeography*, 43(6), 1242–1253. https://doi.org/10.1111/jbi.12703

Ahmadzadeh, F., Flecks, M., Carretero, M. A., Böhme, W., Ilgaz, C., Engler, J. O., James Harris, D., Üzüm, N., & Rödder, D. (2013). Rapid lizard radiation lacking niche conservatism: Ecological diversification within a complex landscape. *Journal of Biogeography*, 40(9), 1807–1818. https://doi.org/10.1111/jbi.12121

Akaike, H. (1978). On the likelihood of a time series model. *The Statistician*, 27(3/4), 217. https://doi.org/10.2307/2988185

Akaike, H. (1979). A Bayesian extension of the minimum AIC procedure of autoregressive model fitting. *Biometrika*, 66(2), 237. https://doi.org/10.2307/2335654

Arnold, E. N., Arribas, O., & Carranza, S. (2007). Systematics of the Palaearctic and Oriental lizard tribe Lacertini (Squamata: Lacertidae: Lacertinae), with descriptions of eight new genera. *Zootaxa*, 1430(1), 1–86.

Ashman, L. G., Bragg, J. G., Doughty, P., Hutchinson, M. N., Bank, S., Matzke, N. J., Oliver, P., & Moritz, C. (2018). Diversification across biomes in a continental lizard radiation. *Evolution*, 72(8), 1553–1569. https://doi.org/10.1111/evo.13541

Badgley, C., Smiley, T. M., Terry, R., Davis, E. B., DeSantis, L. R. G., Fox, D. L., Hopkins, S. S. B., Jezkova, T., Matocq, M. D., Matzke, N., McGuire, J. L., Mulch, A., Riddle, B. R., Roth, V. L., Samuels, J. X., Strömberg, C. A. E., & Yanites, B. J. (2017). Biodiversity and topographic complexity: Modern and geohistorical perspectives. *Trends in Ecology and Evolution*, 32(3), 211–226. https://doi.org/10.1016/j.tree.2016.12.010

Bars-Closel, M., Kohlsdorf, T., Moen, D. S., & Wiens, J. J. (2017). Diversification rates are more strongly related to microhabitat than climate in squamate reptiles (lizards and snakes). *Evolution*, 71(9), 2243–2261. https://doi.org/10.1111/evo.13305

Benjamini, Y., & Hochberg, Y. (1995). Controlling the false discovery rate: A practical and powerful approach to multiple testing. *Journal of the Royal Statistical Society: Series B (Methodological)*, 57(1), 289–300. https://doi.org/10.1111/j.2517-6161.1995.tb02031.x

Blomberg, S. P., Garland, T., & Ives, A. R. (2003). Testing for phylogenetic signal in comparative data: Behavioral traits are more labile. *Evolution*, 57(4), 717. https://doi.org/10.1554/0014-3820(2003)057[0717:tfpsic]2.0.co;2

Burke, K. D., Williams, J. W., Chandler, M. A., Haywood, A. M., Lunt, D. J., & Otto-Bliesner, B. L. (2018). Pliocene and Eocene provide best analogs for near-future climates. *Proceedings of the National Academy of Sciences*, 115(52), 13288–13293. https://doi.org/10.5061/dryad.0j18k00

Burnham, K. P., & Anderson, D. R. (Eds.). (2004). *Model selection and multimodel inference*. Springer. https://doi.org/10.1007/b97636

Cavazza, W., & Wezel, F. C. (2003). The Mediterranean region—A geological primer. *Episodes*, 26(3), 160–168. https://doi.org/10.18814/epiiugs/2003/v26i3/002

Cavender-Bares, J., Gonzalez-Rodriguez, A., Pahlich, A., Koehler, K., & Deacon, N. (2011). Phylogeography and climatic niche evolution in live oaks (*Quercus* series Virentes) from the tropics to the temperate zone. *Journal of Biogeography*, 38(5), 962–981. https://doi.org/10.1111/j.1365-2699.2010.02451.x

Collar, D. C., Schulte, J. A., & Losos, J. B. (2011). Evolution of extreme body size disparity in monitor lizards (*Varanus*). *Evolution*, 65(9), 2664–2680. https://doi.org/10.1111/j.1558-5646.2011.01335.x

Dinis, M., Merabet, K., Martínez-Freiría, F., Steinfartz, S., Vences, M., Burgon, J. D., Elmer, K. R., Donaire, D., Hinckley, A., Fahd, S., Joger, U., Fawzi, A., Slimani, T., & Velo-Antón, G. (2019). Allopatric diversification and evolutionary melting pot in a north African palearctic relict: The biogeographic history of *Salamandra algira*. *Molecular Phylogenetics and Evolution*, 130, 81–91. https://doi.org/10.1016/j.ympev.2018.10.018

Esquerré, D., Brennan, I. G., Catullo, R. A., Torres-Pérez, F., & Keogh, J. S. (2019). How mountains shape biodiversity: The role of the Andes in biogeography, diversification, and reproductive biology in South America's most species-rich lizard radiation (Squamata: Liolaemidae). *Evolution*, 73(2), 214–230. https://doi.org/10.1111/evo.13657

Fick, S. E., & Hijmans, R. J. (2017). WorldClim 2: New 1-km spatial resolution climate surfaces for global land areas. *International Journal of Climatology*, 37(12), 4302–4315. https://doi.org/10.1002/joc.5086

Fitzjohn, R. G. (2010). Quantitative traits and diversification. *Systematic Biology*, 59(6), 619–633. https://doi.org/10.1093/sysbio/syq053

Fitzjohn, R. G. (2012). Diversitree: Comparative phylogenetic analyses of diversification in R. *Methods in Ecology and Evolution*, 3(6), 1084–1092. https://doi.org/10.1111/j.2041-210X.2012.00234.x

Folk, R. A., Stubbs, R. L., Mort, M. E., Cellinese, N., Allen, J. M., Soltis, P. S., Soltis, D. E., & Guralnick, R. P. (2019). Rates of niche and phenotype evolution lag behind diversification in a temperate radiation. *Proceedings of the National Academy of Sciences of the United States of America*, 166(22), 10874–10882. https://doi.org/10.1073/pnas.1817999116

Gajdzik, L., Aguilar-Medrano, R., & Frédérich, B. (2019). Diversification and functional evolution of reef fish feeding guilds. *Ecology Letters*, 22(4), 572–582. https://doi.org/10.1111/ele.13219

Garcia-Porta, J., Irisarri, I., Kirchner, M., Rodríguez, A., Kirchhof, S., Brown, J. L., MacLeod, A., Turner, A. P., Ahmadzadeh, F., Albaladejo, G., Crnobrnja-Isailovic, J., de la Riva, I., Fawzi, A., Galán, P., Göçmen, B., Harris, D. J., Jiménez-Robles, O., Joger, U., Jovanović Glavaš, O., … Wollenberg Valero, K. C. (2019). Environmental temperatures shape thermal physiology as well as diversification and genome-wide substitution rates in lizards. *Nature Communications*, 10(1), 1–12. https://doi.org/10.1038/s41467-019-11943-x

Garcia-Porta, J., Morales, H. E., Gómez-Díaz, E., Sindaco, R., & Carranza, S. (2016). Patterns of diversification in islands: A comparative study across three *Gecko* genera in the Socotra archipelago. *Molecular Phylogenetics and Evolution*, 98, 288–299. https://doi.org/10.1016/j.ympev.2016.02.007

Garcia-Porta, J., Simó-Riudalbas, M., Robinson, M., & Carranza, S. (2017). Diversification in arid mountains: Biogeography and cryptic diversity of *Pristurus rupestris rupestris* in Arabia. *Journal of Biogeography*, 44(8), 1694–1704. https://doi.org/10.1111/jbi.12929

Ghane-Ameleh, S., Khosravi, M., Saberi-Pirooz, R., Ebrahimi, E., Aghbolaghi, M. A., & Ahmadzadeh, F. (2021). Mid-Pleistocene transition as a trigger for diversification in the Irano-Anatolian region: Evidence revealed by phylogeography and distribution pattern of the eastern three-lined lizard. *Global Ecology and Conservation*, 31, e01839. https://doi.org/10.1016/j.gecco.2021.e01839

Gillespie, R. G., Bennett, G. M., de Meester, L., Feder, J. L., Fleischer, R. C., Harmon, L. J., Hendry, A. P., Knope, M. L., Mallet, J., Martin, C., Parent, C. E., Patton, A. H., Pfennig, K. S., Rubinoff, D., Schluter, D., Seehausen, O., Shaw, K. L., Stacy, E., Stervander, M., … Wogan, G. O. U. (2020). Comparing adaptive radiations across space, time, and taxa. *Journal of Heredity*, 111(1), 1–20. https://doi.org/10.1093/jhered/esz064

Gomes, V., Carretero, M. A., & Kaliontzopoulou, A. (2016). The relevance of morphology for habitat use and locomotion in two species of wall lizards. *Acta Oecologica*, 70, 87–95. https://doi.org/10.1016/j.actao.2015.12.005

Gómez, A., & Lunt, D. H. (2015). Refugia within refugia: Patterns of phylogeographic concordance in the Iberian Peninsula. In S. Weiss & N. Ferrand (Eds.), *Phylogeography of southern European refugia* (Vol. 36, pp. 155–188). Springer Netherlands. https://doi.org/10.1007/1-4020-4904-8_5

Gorscak, E., & O'Connor, P. M. (2016). Time-calibrated models support congruency between cretaceous continental rifting and titanosaurian evolutionary history. *Biology Letters*, 12(4), 20151047. https://doi.org/10.1098/rsbl.2015.1047

Gould, S. J., & Eldredge, N. (1977). Punctuated equilibria: The tempo and mode of evolution reconsidered. *Paleobiology*, 3, 115–151.

Graham, C. H., Ron, S. R., Santos, J. C., Schneider, C. J., & Moritz, C. (2004). Integrating phylogenetics and environmental niche models to explore speciation mechanisms in dendrobatid frogs. *Evolution*, 58(8), 1781–1793. https://doi.org/10.1111/j.0014-3820.2004.tb00461.x

Harvey, M. G., & Rabosky, D. L. (2018). Continuous traits and speciation rates: Alternatives to state-dependent diversification models. *Methods in Ecology and Evolution*, 9(4), 984–993. https://doi.org/10.1111/2041-210X.12949

Hewitt, G. (2000). The genetic legacy of the quaternary ice ages. *Nature*, 405(6789), 907–913. https://doi.org/10.1038/35016000

Hewitt, G. M. (2011). Mediterranean Peninsulas: The evolution of hotspots. In F. E. Zachos & J. C. Habel (Eds.), *Biodiversity hotspots*. Springer. https://doi.org/10.1007/978-3-642-20992-5_7

Hijmans, R. J. (2020). *raster: Geographic analysis and modeling with raster data*. R package version 3.1-5, 3.1(5).

Hoorn, C., Wesselingh, F. P., ter Steege, H., Bermudez, M. A., Mora, A., Sevink, J., Sanmartín, I., Sanchez-Meseguer, A., Anderson, C. L., Figueiredo, J. P., Jaramillo, C., Riff, D., Negri, F. R., Hooghiemstra, H., Lundberg, J., Stadler, T., Särkinen, T., & Antonelli, A. (2010). Amazonia through time: Andean. *Science*, 330, 927–931. https://doi.org/10.1126/science.1194585

IUCN. (2018). *IUCN 2018. The IUCN red list of threatened species*. Version 2018-1. http://www.iucnredlist.org

Jiménez-Moreno, G., Fauquette, S., & Suc, J.-P. (2010). Miocene to Pliocene vegetation reconstruction and climate estimates in the Iberian Peninsula from pollen data. *Review of Palaeobotany and Palynology*, 162(3), 403–415. https://doi.org/10.1016/j.revpalbo.2009.08.001

Kaliontzopoulou, A., Adams, D. C., van der Meijden, A., Perera, A., & Carretero, M. A. (2012). Relationships between head morphology, bite performance and ecology in two species of *Podarcis* wall lizards. *Evolutionary Ecology*, 26(4), 825–845. https://doi.org/10.1007/s10682-011-9538-y

Keil, P., & Chase, J. M. (2019). Global patterns and drivers of tree diversity integrated across a continuum of spatial grains. *Nature Ecology and Evolution*, 3(3), 390–399. https://doi.org/10.1038/s41559-019-0799-0

Kennedy, J. D., Borregaard, M. K., Marki, P. Z., Machac, A., Fjeldså, J., & Rahbek, C. (2018). Expansion in geographical and morphological space drives continued lineage diversification in a global passerine radiation. *Proceedings of the Royal Society B: Biological Sciences*, 285(1893), 20182181. https://doi.org/10.1098/rspb.2018.2181

Kozak, K. H., & Wiens, J. J. (2016). What explains patterns of species richness? The relative importance of climatic-niche evolution, morphological evolution, and ecological limits in salamanders. *Ecology and Evolution*, 6(16), 5940–5949. https://doi.org/10.1002/ece3.2301

Lee, M. S. Y., Sanders, K. L., King, B., & Palci, A. (2016). Diversification rates and phenotypic evolution in venomous snakes (Elapidae). *Royal Society Open Science*, 3(1), 150277. https://doi.org/10.1098/rsos.150277

Leprieur, F., Descombes, P., Gaboriau, T., Cowman, P. F., Parravicini, V., Kulbicki, M., Melian, C. J., de Santana, C. N., Heine, C., Mouillot, D., Bellwood, D. R., & Pellissier, L. (2016). Plate tectonics drive tropical reef biodiversity dynamics. *Nature Communications*, 7, 1–8. https://doi.org/10.1038/ncomms11461

Losos, J. B. (2010). Adaptive radiation, ecological opportunity, and evolutionary determinism: American society of naturalists E.O. Wilson award address. *American Naturalist*, 175(6), 623–639. https://doi.org/10.1086/652433

Martínez-Freiría, F., Freitas, I., Zuffi, M. A. L., Golay, P., Ursenbacher, S., & Velo-Antón, G. (2020). Climatic refugia boosted allopatric diversification in Western Mediterranean vipers. *Journal of Biogeography*, 47(8), 1698–1713. https://doi.org/10.1111/jbi.13861

Martínez-Freiría, F., Sillero, N., Lizana, M., & Brito, J. C. (2008). GIS-based niche models identify environmental correlates sustaining a contact zone between three species of European vipers. *Diversity and Distributions*, 14(3), 452–461. https://doi.org/10.1111/j.1472-4642.2007.00446.x

Martínez-Freiría, F., Toyama, K. S., Freitas, I., & Kaliontzopoulou, A. (2020). Thermal melanism explains macroevolutionary variation of dorsal pigmentation in *Eurasian vipers*. *Scientific Reports*, 10(1), 16122. https://doi.org/10.1038/s41598-020-72871-1

Martínez-Gil, H., Martínez-Freiría, F., Perera, A., Enriquez-Urzelai, U., Martínez-Solano, Í., Velo-Antón, G., & Kaliontzopoulou, A. (2021). Morphological diversification of Mediterranean anurans: The roles of evolutionary history and climate. *Biological Journal of the Linnean Society*, 135, 462–477. https://doi.org/10.1093/biolinnean/blab156/6486822

Mas-Peinado, P., García-París, M., Ruiz, J. L., & Buckley, D. (2021). The strait of Gibraltar is an ineffective palaeogeographic barrier for some flightless darkling beetles (Coleoptera: Tenebrionidae: Pimelia). *Zoological Journal of the Linnean Society*, 1, 1–34. https://doi.org/10.1093/zoolinnean/zlab088/6486828

Matzke, N. J. (2013). Probabilistic historical biogeography: New models for founder-event speciation, imperfect detection, and fossils allow improved accuracy and model-testing. *Frontiers of Biogeography*, 5(4), 242–248. https://doi.org/10.21425/f5fbg19694

Matzke, N. J. (2014). Model selection in historical biogeography reveals that founder-event speciation is a crucial process in Island clades. *Systematic Biology*, 63(6), 951–970. https://doi.org/10.1093/sysbio/syu056

Mendes, J., Harris, D. J., Carranza, S., & Salvi, D. (2017). Biogeographical crossroad across the pillars of Hercules: Evolutionary history of *Psammodromus* lizards in space and time. *Journal of Biogeography*, 44(12), 2877–2890. https://doi.org/10.1111/jbi.13100

Miller, E. C., Hayashi, K. T., Song, D., & Wiens, J. J. (2018). Explaining the ocean's richest biodiversity hotspot and global patterns of fish diversity. *Proceedings of the Royal Society B: Biological Sciences*, 285(1888), 20181314. https://doi.org/10.1098/rspb.2018.1314

Moen, D. S., & Wiens, J. J. (2017). Microhabitat and climatic niche change explain patterns of diversification among frog families. *American Naturalist*, 190(1), 29–44. https://doi.org/10.1086/692065

Mosimann, J. E. (1970). Size allometry: Size and shape variables with characterizations of the lognormal and generalized gamma distributions. *Journal of the American Statistical Association*, 65(330), 930–945. https://doi.org/10.1080/01621459.1970.10481136

Myers, N., Mittermeler, R. A., Mittermeler, C. G., da Fonseca, G. A. B., & Kent, J. (2000). Biodiversity hotspots for conservation priorities. *Nature*, 403(6772), 853–858. https://doi.org/10.1038/35002501

Olson, D. M., Dinerstein, E., Wikramanayake, E. D., Burgess, N. D., Powell, G. V. N., Underwood, E. C., D'amico, J. A., Itoua, I., Strand, H. E., Morrison, J. C., Loucks, C. J., Allnutt, T. F., Ricketts, T. H., Kura, Y., Lamoreux, J. F., Wettengel, W. W., Hedao, P., & Kassem, K. R. (2001). Terrestrial ecoregions of the world: A new map of life on earth: A new global map of terrestrial ecoregions provides an innovative tool for conserving biodiversity. *BioScience*, 51(11), 933–938.

Owens, H. L., Lewis, D. S., Dupuis, J. R., Clamens, A. L., Sperling, F. A. H., Kawahara, A. Y., Guralnick, R. P., & Condamine, F. L. (2017). The latitudinal diversity gradient in New World swallowtail butterflies is caused by contrasting patterns of out-of- and into-the-tropics dispersal. *Global Ecology and Biogeography*, 26(12), 1447–1458. https://doi.org/10.1111/geb.12672

Pato, J., Illera, J. C., Obeso, J. R., & Laiolo, P. (2019). The roles of geography, climate and sexual selection in driving divergence among insect populations on mountaintops. *Journal of Biogeography*, 46(4), 784–795. https://doi.org/10.1111/jbi.13540

Paulo, O. S., Pinheiro, J., Miraldo, A., Bruford, M. W., Jordan, W. C., & Nichols, R. A. (2008). The role of vicariance vs. dispersal in shaping genetic patterns in ocellated lizard species in the western Mediterranean. *Molecular Ecology*, 17(6), 1535–1551. https://doi.org/10.1111/j.1365-294X.2008.03706.x

Pennington, R. T., Lavin, M., Särkinen, T., Lewis, G. P., Klitgaard, B. B., & Hughes, C. E. (2010). Contrasting plant diversification histories within the Andean biodiversity hotspot. *Proceedings of the National Academy of Sciences of the United States of America*, 107(31), 13783–13787. https://doi.org/10.1073/pnas.1001317107

Pincheira-Donoso, D., Harvey, L. P., & Ruta, M. (2015). What defines an adaptive radiation? Macroevolutionary diversification dynamics of an exceptionally species-rich continental lizard radiation. *BMC Evolutionary Biology*, 15(1), 1–13. https://doi.org/10.1186/s12862-015-0435-9

Popov, S. V., Shcherba, I. G., Ilyina, L. B., Nevesskaya, L. A., Paramonova, N. P., Khondkarian, S. O., & Magyar, I. (2006). Late Miocene to Pliocene palaeogeography of the Paratethys and its relation to the Mediterranean. *Palaeogeography, Palaeoclimatology, Palaeoecology*, 238(1–4), 91–106. https://doi.org/10.1016/j.palaeo.2006.03.020

Price, T. D., Hooper, D. M., Buchanan, C. D., Johansson, U. S., Tietze, D. T., Alström, P., Olsson, U., Ghosh-Harihar, M., Ishtiaq, F., Gupta, S. K., Martens, J., Harr, B., Singh, P., & Mohan, D. (2014). Niche filling slows the diversification of Himalayan songbirds. *Nature*, 509(7499), 222–225. https://doi.org/10.1038/nature13272

Prista, G. A., Agostinho, R. J., & Cachão, M. A. (2015). Observing the past to better understand the future: A synthesis of the Neogene climate in Europe and its perspectives on present climate change. *Open Geosciences*, 7(1), 65–83. https://doi.org/10.1515/geo-2015-0007

QGIS. (2020). *Quantum GIS geographic information system.* Open Source Geospatial Foundation Project. http://qgis.osgeo.org

Qian, H., & Ricklefs, R. E. (2000). Large-scale processes and the Asian bias in species diversity of temperate plants. *Nature*, 407(6801), 180–182. https://doi.org/10.1038/35025052

Rabosky, D. L. (2014). Automatic detection of key innovations, rate shifts, and diversity-dependence on phylogenetic trees. *PLoS One*, 9(2), e89543. https://doi.org/10.1371/journal.pone.0089543

Rabosky, D. L., Grundler, M., Anderson, C., Title, P., Shi, J. J., Brown, J. W., Huang, H., & Larson, J. G. (2014). BAMMtools: An R package for the analysis of evolutionary dynamics on phylogenetic trees. *Methods in Ecology and Evolution*, 5(7), 701–707. https://doi.org/10.1111/2041-210X.12199

Rabosky, D. L., & Huang, H. (2016). A robust semi-parametric test for detecting trait-dependent diversification. *Systematic Biology*, 65(2), 181–193. https://doi.org/10.1093/sysbio/syv066

Ramírez-Barahona, S., Barrera-Redondo, J., & Eguiarte, L. E. (2016). Rates of ecological divergence and body size evolution are correlated with species diversification in scaly tree ferns. *Proceedings of the Royal Society B: Biological Sciences*, 283(1834), 20161098. https://doi.org/10.1098/rspb.2016.1098

Rato, C., Harris, D. J., Perera, A., Carvalho, S. B., Carretero, M. A., & Rödder, D. (2015). A combination of divergence and conservatism in the niche evolution of the Moorish gecko, *Tarentola mauritanica* (Gekkota: Phyllodactylidae). *PLoS One*, 10(5), 1–16. https://doi.org/10.1371/journal.pone.0127980

Reaney, A. M., Saldarriaga-Córdoba, M., & Pincheira-Donoso, D. (2018). Macroevolutionary diversification with limited niche disparity in a species-rich lineage of cold-climate lizards. *BMC Evolutionary Biology*, 18(1), 1–12. https://doi.org/10.1186/s12862-018-1133-1

Rögl, F. (1998). Palaeogeographic considerations for Mediterranean and Paratethys seaways (Oligocene to Miocene). *Annalen des Naturhistorischen Museums in Wien*, 99, 279–310. https://doi.org/10.2307/41702129

Rosenbaum, G., Lister, G. S., & Duboz, C. (2002). Relative motions of Africa, Iberia and Europe during alpine orogeny. *Tectonophysics*, 359(1–2), 117–129. https://doi.org/10.1016/S0040-1951(02)00442-0

Sandel, B., Weigelt, P., Kreft, H., Keppel, G., van der Sande, M. T., Levin, S., Smith, S., Craven, D., & Knight, T. M. (2020). Current climate, isolation and history drive global patterns of tree phylogenetic endemism. *Global Ecology and Biogeography*, 29(1), 4–15. https://doi.org/10.1111/geb.13001

Schenk, J. J., Rowe, K. C., & Steppan, S. J. (2013). Ecological opportunity and incumbency in the diversification of repeated continental colonizations by muroid rodents. *Systematic Biology*, 62(6), 837–864. https://doi.org/10.1093/sysbio/syt050

Schenk, J. J., & Steppan, S. J. (2018). The role of geography in adaptive radiation. *The American Naturalist*, 192(4), 415–431. https://doi.org/10.1086/699221

Skeels, A., & Cardillo, M. (2017). Environmental niche conservatism explains the accumulation of species richness in Mediterranean-hotspot plant genera. *Evolution*, 71(3), 582–594. https://doi.org/10.1111/evo.13179

Skeels, A., & Cardillo, M. (2019). Equilibrium and non-equilibrium phases in the radiation of hakea and the drivers of diversity in Mediterranean-type ecosystems. *Evolution*, 73(7), 1392–1410. https://doi.org/10.1111/evo.13769

Steininger, F. F., & Rögl, F. (1984). Paleogeography and palinspastic reconstruction of the Neogene of the Mediterranean and Paratethys. *Geological Society Special Publication*, 17, 659–668. https://doi.org/10.1144/GSL.SP.1984.017.01.52

van Dam, M. H., & Matzke, N. J. (2016). Evaluating the influence of connectivity and distance on biogeographical patterns in the southwestern deserts of North America. *Journal of Biogeography*, 43(8), 1514–1532. https://doi.org/10.1111/jbi.12727

Velasco, J. A., Villalobos, F., Diniz-Filho, J. A. F., Poe, S., & Flores-Villela, O. (2020). Macroecology and macroevolution of body size in *Anolis* lizards. *Ecography*, 43(6), 812–822. https://doi.org/10.1111/ecog.04583

Velo-Antón, G., Parra, J. L., Parra-Olea, G., & Zamudio, K. R. (2013). Tracking climate change in a dispersal-limited species: Reduced spatial and genetic connectivity in a montane salamander. *Molecular Ecology*, 22(12), 3261–3278. https://doi.org/10.1111/mec.12310

Warton, D. I., Duursma, R. A., Falster, D. S., & Taskinen, S. (2012). Smatr 3- an R package for estimation and inference about allometric lines. *Methods in Ecology and Evolution*, 3(2), 257–259. https://doi.org/10.1111/j.2041-210X.2011.00153.x

Yoder, J. B., Clancey, E., des Roches, S., Eastman, J. M., Gentry, L., Godsoe, W., Hagey, T. J., Jochimsen, D., Oswald, B. P., Robertson, J., Sarver, B. A. J., Schenk, J. J., Spear, S. F., & Harmon, L. J. (2010). Ecological opportunity and the origin of adaptive radiations. *Journal of Evolutionary Biology*, 23(8), 1581–1596. https://doi.org/10.1111/j.1420-9101.2010.02029.x

Žagar, A., Carretero, M. A., Vrezec, A., Drašler, K., & Kaliontzopoulou, A. (2017). Towards a functional understanding of species coexistence: Ecomorphological variation in relation to whole-organism performance in two sympatric lizards. *Functional Ecology*, 31(9), 1780–1791. https://doi.org/10.1111/1365-2435.12878

Zhao, Z., Shao, L., Li, F., Zhang, X., & Li, S. (2020). Tectonic evolution of the Tethyan region created the Eurasian extratropical biodiversity hotspots: Tracing *Pireneitega* spiders' diversification history. *Ecography*, 43, 1400–1411. https://doi.org/10.1111/ecog.05044

BIOSKETCH

Urtzi Enriquez-Urzelai is broadly interested in understanding the factors that determine species distributions and diversity patterns, from phenotypic adaptations (e.g. physiological, morphological and behavioural) to biogeographical aspects. This work represents part of his postdoctoral work within the MEDBIODIV project (https://cibio.up.pt/projects-1/page/591) of members of the PhenEvol research group (see https://sites.google.com/view/phenevol/about-us).

Author Contributions: FM-F and AK collected data; UEU, FM-F, IF, AP, ÍMS, DS, GVA and AK conceived the ideas and designed the methodology; UEU, IF, AP and AK analysed the data; UEU led the writing of the manuscript. All authors contributed to the drafts and gave final approval for publication.

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.