LETTER

Accelerated rates of climatic-niche evolution underlie rapid species diversification

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Abstract

A major goal of ecology is to explain differences in species richness between regions and among clades. The diversification rate of clades is a key parameter for understanding both patterns. Here, we combine phylogenetic and climatic data for 250 species of plethodontid salamanders and show for the first time that rapid species diversification is associated with accelerated climatic-niche evolution among species. Both rates are particularly rapid in tropical regions, and where few clades geographically overlap. These results offer a surprising ecological explanation for why diversification rates are often higher in the tropics: rapid shifts between climatic regimes, rather than specific environmental conditions (e.g. high productivity, energy) drive higher diversity. They also suggest that climatic-niche evolution may be particularly rapid in regions where climate is particularly stable. Finally, these results indicate that evolutionary conservatism in climatic niches may be influenced by interactions between species and clades, rather than physiological tolerances alone.

Keywords

Amphibians, climate change, diversification, diversity, niche conservatism, niche evolution, phylogeny, Plethodontidae, salamanders, speciation, species interactions, species richness.

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INTRODUCTION

A major goal of ecology is to explain differences in species richness between regions and groups of organisms. The diversification rate of a clade is a key parameter for understanding both patterns, and is the rate at which species accumulate over time, or the rate of speciation minus the rate of extinction. For example, some studies suggest that primarily tropical clades often have higher diversification rates, and that this may explain higher tropical richness (e.g. Cardillo *et al.* 2005; Ricklefs 2006; Mittelbach *et al.* 2007; Wiens 2007; Svenning *et al.* 2008). Many other factors have been proposed as correlates of higher diversification rates, including the geographic extent of clades (e.g. Rabosky 2009a), invasion of new adaptive zones (e.g. Simpson 1944; Mitter *et al.* 1988), morphological diversification (e.g. Adams *et al.* 2009) and sexually selected traits (e.g. Seddon *et al.* 2008).

Rapids shifts in climatic niches among species might also drive high diversification rates (e.g. Moritz *et al.* 2000; Kozak & Wiens 2007). For example, occurrence of incipient sister species in different climatic environments is a

potential cause of reproductive isolation between them (if individuals of one species cannot persist in the geographic range of the other; Sobel *et al.* 2010), and could be an important factor promoting parapatric and allopatric speciation. In addition, species and clades in which climatic tolerances can evolve rapidly may be less susceptible to extinction from climatic change (e.g. Holt 1990), and may diversify more rapidly by spreading into many different environments, thereby reducing competition and creating additional opportunities for speciation. In contrast, climatic-niche conservatism may promote allopatric speciation (e.g. when populations of mesic-adapted species become isolated on separate mountaintops during periods of global warming; Kozak & Wiens 2006), but may restrict elevational and geographic ranges of species and clades and leave them more susceptible to extinction. Many studies have used GIS-based climatic data to test whether speciation is related to niche divergence or conservatism (e.g. Peterson *et al.* 1999; Kozak & Wiens 2006, 2007; Raxworthy *et al.* 2007; Warren *et al.* 2008; Hua & Wiens 2010). However, no studies have directly tested for a relationship between the rate at which environmental differences evolve among species and the

rate at which the clade containing them diversifies, and it is unclear whether rates of climatic-niche evolution should be positively or negatively associated with diversification rates (or unrelated).

It is also unclear why rates of climatic-niche evolution might vary among clades. Several studies have found evidence for climatic-niche conservatism among species using phylogenetic methods (e.g. Crisp *et al.* 2009; Kozak & Wiens 2010), whereas others have shown evidence for niche divergence (e.g. Knouft *et al.* 2006; Evans *et al.* 2009). These contrasting results suggest that rates of climatic-niche evolution might be fast in some clades and slow in others, but variation among clades is poorly documented and its causes are poorly understood (but see Smith & Beaulieu 2009).

A study of speciation in salamanders (Kozak & Wiens 2007) implied that rates of climatic-niche evolution might be higher in tropical clades, based on lesser overlap in climatic distribution between 14 tropical sister species relative to 16 temperate pairs. These authors related these patterns to Janzen's (1967) hypothesis of 'why mountain passes are higher in the tropics'. Specifically, they suggested that tropical species may show greater divergence in climatic niches because of reduced overlap in climatic conditions at different elevations in the tropics, associated with limited seasonality. These authors speculated that this pattern of speciation and climatic divergence could lead to higher diversification rates in tropical regions. Many studies have found evidence for higher diversification rates in the tropics, but none tested whether this is related to faster climatic divergence among tropical species, and most did not address the underlying causes at all (e.g. Cardillo *et al.* 2005; Ricklefs 2006; Wiens 2007; Svenning *et al.* 2008).

In general, the rate of climatic-niche evolution may depend on the rate of evolution of physiological tolerances to these environmental conditions (e.g. tolerance to heat or drought). For example, limited tolerance to freezing temperatures may constrain lineages from dispersing from tropical to temperate regions, whereas tolerances that evolve rapidly might allow for rapid changes in the climatic distributions of species. Furthermore, clades with larger geographic areas may exhibit greater climatic-niche evolution, as their species may encounter a broader range of environments than highly localized clades. However, the rate of climatic-niche evolution may depend on biotic factors as well (Wiens *et al.* 2010). Perhaps most importantly, the presence of a clade in one climatic regime may limit other clades from dispersing into that regime or diversifying in it, such as when two broadly sympatric clades occupy different elevational ranges. Thus, geographic overlap between clades may limit rates of climatic-niche evolution and species diversification, and geographic isolation of clades may accelerate these rates. For example,

Hawaiian silverswords seem to have undergone rapid species diversification and rapid divergence in climatic niches, possibly because mainland clades that would otherwise occupy these climatic niches are absent (e.g. Baldwin & Sanderson 1998). Yet, the effects of geographic overlap of clades on rates of climatic-niche evolution have never been explicitly tested.

Here, we test the relationship between rates of climatic-niche evolution and rates of species diversification, using lungless salamanders (Plethodontidae) as a model system. Plethodontids include 394 of 585 salamander species (Amphibiaweb 2010) and are the only salamander family to have radiated extensively in the tropics (Wiens 2007). Recent studies have yielded a well-resolved time-calibrated phylogeny that includes the majority of plethodontid species (e.g. Adams *et al.* 2009), which is critical for estimating evolutionary rates. Earlier studies showed that tropical plethodontids have higher diversification rates (Wiens 2007; Adams *et al.* 2009) and that tropical sister species tend to occur in divergent climatic niches (Kozak & Wiens 2007; based on only 30 species). Previous studies also suggested that rates of diversification and morphological evolution are decoupled in plethodontids (Adams *et al.* 2009), leaving open the question of what explains the extensive variation in diversification rates among clades and why tropical clades have higher rates. Here, we show that variation in diversification rates among clades is strongly related to rates of climatic-niche evolution. Furthermore, we show that higher rates of species diversification and climatic-niche evolution are associated with geographic isolation of clades. This latter result suggests that pre-emptive occupancy of climatic niches within a region may be an important but underappreciated factor driving climatic-niche conservatism.

MATERIAL AND METHODS

Phylogeny and diversification rates

Rate estimates and phylogenetic comparative analyses were based on the chronogram of Adams *et al.* (2009). This chronogram includes 235 of 394 recognized species (AmphibiaWeb 2010, http://amphibiaweb.org/), and 15 distinct phylogeographic lineages not yet formally recognized as species (Kozak *et al.* 2009), totalling 250 putative species. This chronogram was estimated by combining that of Kozak *et al.* (2009) for plethodontids exclusive of most bolitoglossines (based on multiple nuclear and mitochondrial genes), with the phylogeny of Wiens *et al.* (2007) for bolitoglossines (from mitochondrial data). Each phylogeny is strongly supported overall. Chronograms were estimated using penalized likelihood in r8s (Sanderson 2003) assuming three possible root ages for Plethdontidae (48, 61 and 69 Myr) and four internal minimum-age constraints based

on fossils. Although it may be preferable to estimate phylogeny and divergence times simultaneously, this was difficult given the size of these datasets and incomplete overlap of taxa and characters. Our main results are robust to a range of possible ages for the root of Plethodontidae (see below).

To analyse rates of diversification and climatic-niche evolution, we selected 16 phylogenetically non-overlapping clades from the chronogram [Fig. 1; those used by Adams *et al.* (2009) plus *Batrachoseps*]. Given that these clades correspond to traditionally recognized taxonomic groups (e.g. genera), species that were not included in the phylogeny could still be unambiguously assigned to these

well-supported monophyletic groups. Clades were also selected to avoid estimating rates in clades with < 4 species. Different clade divisions that meet these two criteria are possible. Nevertheless, alternative divisions that lump or subdivide these 16 focal clades produced similar results (see Results).

We used the method-of-moments estimator for crown groups to estimate the net diversification rate of each clade (Magallón & Sanderson 2001), given its age (from the chronogram) and species richness [based on AmphibiaWeb (2010), plus the 15 phylogeographic lineages]. We used three possible root ages for plethodontids [48, 61 and 69 Myr; Wiens (2007)], which generate a range of relative clade ages
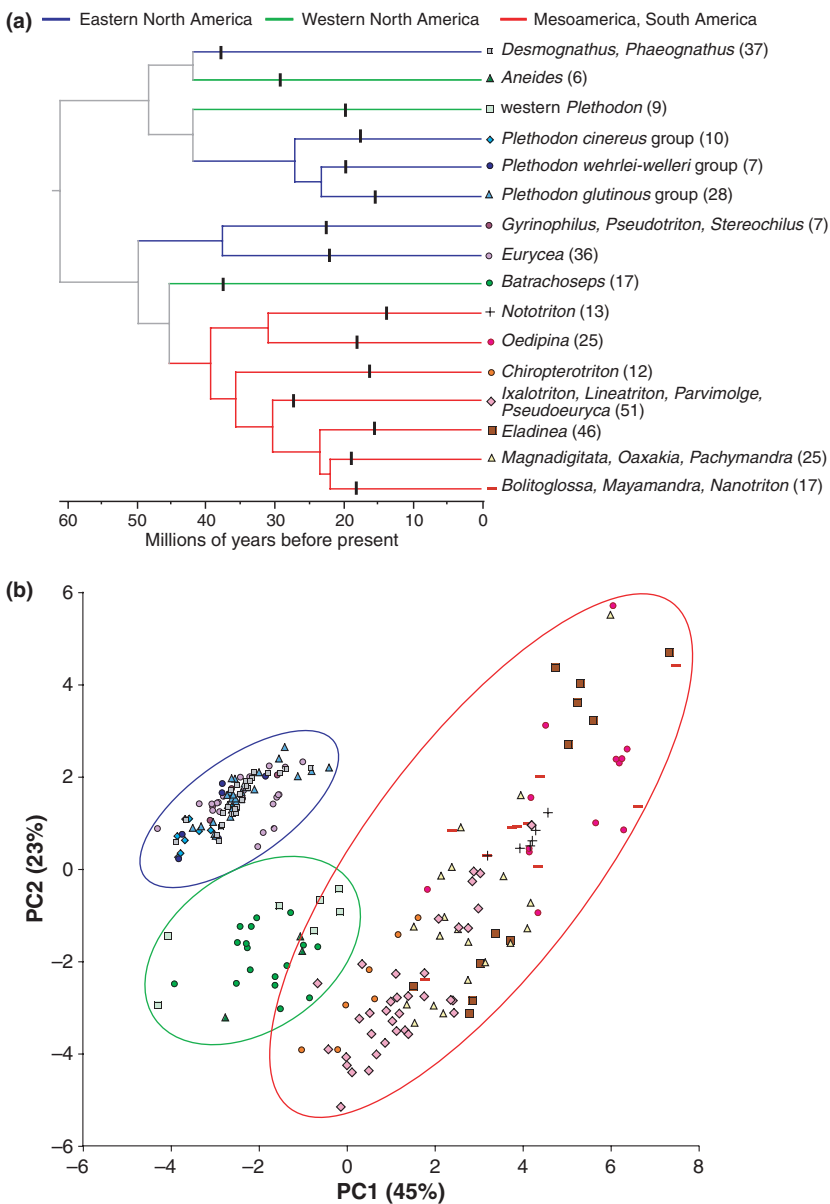

Figure 1 (a) Chronogram showing the divergence times, geographic distribution and phylogenetic relationships of 16 major clades of plethodontid salamanders; species richness of each clade is shown in parentheses. Grey branches indicate ancestors with uncertain biogeographic distributions. Vertical bars on terminal branches indicate the crown age of the corresponding clade. (b) Principal components analysis depicting the position of the 16 focal clades of plethodontid salamanders in climatic-niche space, with each data point representing the mean factor scores for a given species.

throughout the tree, given that we used several internal minimum-age constraints within Plethodontidae based on fossils. We also explored a broad range of relative extinction rates (0.00, 0.45 and 0.90), but different root ages and relative extinction rates gave similar results (see Table S1 for results from different root ages). For brevity, only results using an intermediate root age and relative extinction rate (61 Myr, 0.45) are presented.

Some authors (e.g. Rabosky 2009b) have speculated that if diversification rates vary over time within a group then they may be problematic for determining causes of species richness patterns. For example, high diversification rates in young clades may decouple diversification rates from patterns of species richness. However, we found that diversification rates and species richness of clades were strongly correlated (r^2 = 0.48–0.52, P = 0.001–0.003; depending on root age). We also tested for variation in diversification rates over time within each clade using the gamma statistic (Pybus & Harvey 2000), using 5000 simulation replicates per clade to account for incomplete taxon sampling (implemented in LASER; Rabosky 2006). Gamma values were negative in most clades (12–15 of 16 clades, depending on the overall root age; Tables S2–S4), suggesting declining diversification rates. However, there was generally no relationship between gamma values and species richness, clade age or net diversification rates (Table S5), suggesting that these apparent declines are not relevant to understanding the richness patterns studied here. Finally, our goal is to estimate and compare net diversification rates among clades (the number of species that have accumulated since the clade's origin) and we do not expect speciation and extinction to occur constantly and at the same rates at every point in time before the present. Even if diversification rates change over time, variation in net rates of species accumulation still appear to explain differences in species richness among these clades, whereas patterns of declining rates over time do not.

Climate data, climatic disparity and rates of climatic-niche evolution

We mapped 19 195 georeferenced collection locations (mean = 76 locations/species, range = 1–2446) obtained from the U.S. Museum of Natural History (Smithsonian), Museum of Vertebrate Zoology (University of California, Berkeley) and published systematic studies. These records summarize the known distributions of the 250 species included in the chronogram, and were verified against distribution maps and elevational ranges published by the Global Amphibian Assessment (GAA; http://www.iucn redlist.org). For each record, we extracted values for elevation and 19 climatic variables from the WorldClim dataset with 30-s spatial resolution (Hijmans et al. 2005).

We then calculated the mean value for each variable for each species.

To characterize the climatic-niche space occupied by each clade (i.e. the macroclimatic conditions under which its species are found), we performed a principal components analysis (PCA) on the correlation matrix of the 20 environmental variables (Table 1). PC1–PC4 accounted for > 90% of climatic variation among species, but only PC1–PC3 explained more variation than expected based on

Table 1 Factor loadings and eigenvalues from the principal components analysis of the 20 environmental variables. PC1–PC4 explained > 90% of the variation among species

Variable	PC1	PC2	PC3	PC4
Elevation	−0.21	0.78	−0.44	−0.14
BIO1	−0.76	−0.33	0.53	−0.12
BIO2	0.51	0.32	0.47	0.08
BIO3	−0.91	0.34	−0.03	−0.02
BIO4	0.90	−0.39	0.04	0.02
BIO5	0.09	−0.60	0.78	0.02
BIO6	−0.95	−0.04	0.26	−0.01
BIO7	0.93	−0.26	0.14	0.01
BIO8	−0.54	−0.27	0.34	−0.65
BIO9	−0.64	−0.13	0.40	0.52
BIO10	−0.05	−0.74	0.65	−0.09
BIO11	−0.93	0.00	0.32	−0.05
BIO12	−0.78	−0.42	−0.39	0.09
BIO13	−0.89	−0.09	−0.21	0.08
BIO14	0.11	−0.79	−0.50	−0.07
BIO15	−0.51	0.73	0.33	0.16
BIO16	−0.89	−0.11	−0.22	0.10
BIO17	0.06	−0.81	−0.50	−0.03
BIO18	−0.66	−0.30	−0.45	−0.32
BIO19	−0.32	−0.60	−0.25	0.58
Eigenvalue	8.90	4.63	3.30	1.24
% Variation	44.50	23.15	16.50	16.24

Bioclimatic variables are as follows: BIO1 = annual mean temperature; BIO2 = mean diurnal temperature range [mean of monthly (maximum temperature − minimum temperature)]; BIO3 = isothermality (BIO2/BIO7 × 100); BIO4 = temperature seasonality (standard deviation of monthly temperature); BIO5 = minimum temperature of the coldest month; BIO6 = maximum temperature of the warmest month; BIO7 = temperature range (BIO6 − BIO5); BIO8 = mean temperature of the wettest quarter; BIO9 = mean temperature of the driest quarter; BIO10 = mean temperature of the warmest quarter; BIO11 = mean temperature of the coldest quarter; BIO12 = annual precipitation; BIO13 = precipitation of the wettest month; BIO14 = precipitation of the driest month; BIO15 = precipitation seasonality (standard deviation of monthly precipitation); BIO16 = precipitation of the wettest quarter; BIO17 = precipitation of the driest quarter; BIO18 = precipitation of the warmest quarter; BIO19 = precipitation of the coldest quarter.

a broken-stick distribution (Jackson 1993), and were used in all analyses.

To quantify the breadth of climatic-niche space occupied by each clade, we extracted factor scores (for PC1–PC3) for each species and then quantified climatic disparity for each clade (mean Euclidean distance among species). To compare climatic divergence among clades in different regions, we conducted separate multivariate analyses of variance (MANOVA) for clades restricted to eastern North America, western North America and Mesoamerica + South America, where clade membership of species was the factor, and species' factor scores along PC1–PC3 were dependent variables.

Prior to estimating rates of climatic-niche evolution, we used the OUCH package (Butler and King 2004) in R to assess the likelihood of observing the pattern of climatic divergence in each clade under Brownian motion (BM) vs. Ornstein-Uhlenbeck models of evolution. We assessed model fit using the sample-size corrected Akaike Information Criterion. Given the strong overall fit of BM across most clades (Table S6), we used factor scores from PC1 to PC3 for each species and the topology and branch lengths from the chronogram to estimate the rate of climatic-niche evolution (σ^2) for each clade under the BM model (following O'Meara *et al.* 2006). We summed diagonal elements of the evolutionary variance–covariance matrix for PC1–PC3 to obtain a multivariate estimate of the rate of climatic-niche evolution (following Adams *et al.* 2009). We estimated the evolutionary variance–covariance matrix using GEIGER (Harmon *et al.* 2008). Analyses using all 20 PCs simultaneously gave similar results (not shown), as did those excluding four clades not fitting BM for two or more of PCs 1–3 (see Results).

In general, we assume some relationship exists between climatic distributions and climatic tolerances of species, and that the trait which evolves is the physiology underlying these climatic tolerances. In partial support of this hypothesis, we previously found a strong relationship between these large-scale climatic data and field body temperatures in temperate and tropical plethodontids (Kozak & Wiens 2007).

Given that current methods for estimating rates do not incorporate within-species variance, we based most analyses on mean climatic values for each species. Although climatic conditions can vary across geographic ranges of species, PCA of the climatic data (Fig. 1) shows that species from the same major climatic and geographic regions cluster in climatic-niche space (especially in temperate regions) strongly suggesting that within-species variation has not obscured the large-scale patterns in the climatic distribution of species and clades. Furthermore, tropical plethodontid species generally have narrow climatic distributions (Kozak & Wiens 2007), and so estimates for these species should not be misled by within-species variation. We also

conducted separate analyses based on maximum and minimum values for each species, to explore the sensitivity of our results to measures that may more closely approximate the limits of species' climatic niches.

Not all plethodontid species were included in the estimation of rates of climatic-niche evolution, because some are not included in the time-calibrated phylogeny. However, Spearman's rank correlation analysis shows no relationship between the rate of climatic-niche evolution in a clade and the proportion of species sampled ($P > 0.18$). Although the clade with the highest rates of climatic-niche evolution is also the most poorly sampled (*Eladinea*), more complete taxon sampling within this clade should actually lead to higher estimated rates (as simulations suggest that poorly sampled clades may have underestimated rates; O'Meara *et al.* 2006, p. 925). Furthermore, adding taxa that inhabit presently unsampled portions of climatic space should tend to increase estimates of the rate of climatic-niche evolution (all other things being equal).

This analysis addresses only the realized climatic-niche space occupied by a clade (based on species averages, maxima and minima), which may correspond to only a subset of the fundamental climatic-niche space (e.g. when geographic distributions of species are limited by other clades; Pearson & Dawson 2003). Under such circumstances, the rate of climatic-niche evolution for a clade may be overestimated (e.g. differences between species will be exaggerated). However, we find that clades with higher rates of climatic-niche evolution exhibit less geographic overlap with other clades, the opposite of the pattern expected if species interactions led to inflated estimates of climatic-niche evolution. Thus, discrepancies between realized and fundamental climatic niches do not appear to have determined our conclusions.

Clade area and overlap

We obtained species range maps from the GAA (IUCN 2010, http://www.iucnredlist.org), imported them into ArcGIS, and merged maps across all included species to obtain a map for each clade. We then converted these maps to a Lambert Azimuthal Equal-Area projection, and used Hawth's Tools in ArcGIS to calculate area (km^2) for each clade.

For each clade, we calculated the area that its geographic range overlapped with other clades as a proportion of the clade's total area. When a clade overlapped with more than one clade, we merged the distributions of all overlapping clades and calculated the proportion of the clade's geographic distribution that was contained within the merged clade ranges. Thus, our index of clade overlap potentially ranged from 0 (i.e. a clade that showed no overlap with any other clades) to 1 (i.e. a clade completely contained within

the distribution of one or more other clades). This is not a strict index of clade sympatry, because species might overlap at a macrogeographic scale, but still not co-occur locally (e.g. low vs. high elevation species). Nevertheless, it clearly captures large-scale patterns of clade isolation and overlap that may influence the diversification and climatic distributions of species (e.g. *Eladinea* is largely allopatric relative to all other plethodontids; *Desmognathus* is broadly sympatric with three other clades).

The geographic overlap of species from the same clade might also impact rates of diversification and niche evolution. We quantified this overlap in each clade in two ways (Table S7), one based on the minimum level of overlap

(proportion of clade's area in which two or more species overlapped), and the other based on the maximum level of overlap (maximum number of sympatric species within a clade).

Comparative phylogenetic analyses

For each clade, we calculated: (1) species richness, (2) age, (3) net diversification rate, (4) climatic disparity, (5) rate of climatic-niche evolution, (6) area and (7) extent of geographic overlap with other plethodontid clades (Table 2). First, we tested whether clade age and net diversification rate are positively correlated with species richness. We then

Table 2 Number of sampled species (*n*), species richness, area, age, diversification rates, clade overlap and rates of climate evolution for the 16 major clades designated for analysis

Clade	*n*	Species richness	Area (km²)	Age (Myr)	Clade overlap	Diversification rate	Climatic disparity	Climatic-niche rate
Subgenus *Eladinea** (*Bolitoglossa*)	12	46	2 218 964	16.3	0.03	0.18	37.31	0.371
Subgenera *Magnadigitata, Oaxakia, Pachymandra** (*Bolitoglossa*)	20	25	50 679	19.4	0.51	0.12	22.68	0.163
Subgenera *Bolitoglossa, Mayamandra, Nanotriton** (*Bolitoglossa*)	10	17	493 429	18.8	0.54	0.10	23.11	0.169
*Ixalotriton, Lineatriton, Parvimolge, Pseudoeurycea**	37	51	170 551	27.6	0.16	0.11	15.61	0.205
*Chiropterotriton**	7	12	18 611	16.6	0.69	0.10	8.08	0.039
*Oedipina**	13	25	284 355	18.0	0.83	0.13	21.10	0.178
*Nototriton**	6	13	2021	13.5	1.00	0.13	10.97	0.080
Batrachoseps	17	17	161 426	36.5	0.97	0.05	8.68	0.032
Gyrinophilus, Pseudotriton, Stereochilus	4	7	2 763 258	23.4	1.00	0.03	5.32	0.024
Eurycea	24	36	5 500 929	22.7	0.93	0.10	9.60	0.047
Western *Plethodon*	7	9	384 310	20.5	0.56	0.04	20.20	0.098
Plethodon cinereus group	9	10	2 298 441	18.1	0.85	0.08	4.20	0.025
Plethodon wehrlei-welleri group	7	7	522 013	19.9	1.00	0.06	9.63	0.029
Plethodon glutinosus group	28	28	2 486 206	15.7	0.83	0.16	10.07	0.066
Aneides	5	6	532 466	30.4	1.00	0.03	20.41	0.076
Desmognathus, Phaeognathus	28	37	3 065 655	36.9	0.96	0.07	10.03	0.079

Asterisks indicate tropical clades.

tested whether diversification rates are associated with rates of climatic-niche evolution. Finally, we explored whether rates of diversification and climatic-niche evolution are correlated with clade area and geographic overlap. All tests used phylogenetic least-squares regression (PGLS) in APE (Paradis *et al.* 2004), and we used sequential Bonferroni correction (Rice 1989) to assess statistical significance.

Given that variables such as clade area, clade overlap and rates of niche evolution may independently or jointly influence diversification rates, we used path analysis to identify the strongest predictors of variation in diversification rates. We used GEIGER to calculate phylogenetically independent contrasts for clade area, overlap, rate of climatic-niche evolution and diversification rate. We then used these independent contrasts to calculate the direct and indirect effects of clade area, clade overlap and rate of climatic-niche evolution on diversification rates in R version 2.10.

A correlation between diversification rate and rate of niche evolution might be explained as an artefact of scaling of climatic disparity and species richness. Similarly, a significant association between diversification rate and rate of climatic-niche evolution might arise if both variables scale with clade age. To explore these possibilities, we tested for a relationship between climatic disparity and clade species richness. We also tested for a relationship between clade age and rate of niche evolution. Climatic disparity and species richness are uncorrelated ($r^2 = 0.11$, $P = 0.321$). Clade age is marginally (negatively) associated with net diversification rate ($r^2 = 0.33$, $P = 0.051$), and is uncorrelated with rate of climatic-niche evolution ($r^2 = 0.05$, $P = 0.252$). Thus, any correlation between rates of diversification and rates of climatic-niche evolution cannot be attributed to the scaling of climatic disparity with species richness, or the scaling of both rates with clade age.

RESULTS

Principal components analysis reveals extensive variation in the breadth of the climatic-niche space occupied by each clade and their overlap in climatic space (Fig. 1). Clades from eastern North America, western North America and Mesoamerica + South America each cluster into distinct regions in climatic space (Fig. 1). Tropical clades exhibit greater disparity than temperate clades ($F = 5.345$, $P = 0.036$), and show greater separation in climatic space among their species (MANOVA: $F = 4.30$) than temperate clades from eastern (MANOVA: $F = 1.49$) and western North America (MANOVA: $F = 3.47$).

We found a strong positive correlation between net diversification rate and species richness ($r^2 = 0.48$, $P = 0.003$), but not between clade age and species richness ($r^2 = 0.006$, $P = 0.733$). Diversification rates are significantly

correlated with rates of climatic-niche evolution ($r^2 = 0.41$, $P = 0.008$); clades with high diversification rates exhibit greater rates of change in multivariate climatic-niche space (Table 3; Fig. 2). This relationship remains significant when four clades that do not conform to a BM model of climatic evolution (Table S6) are excluded ($r^2 = 0.52$, $P = 0.007$),

Table 3 Results of phylogenetic least-squares regressions for the 16 clades in Table 1

Analysis	β	SE	t	P-value
Clade age vs. clade richness	0.008	0.026	0.291	0.772
Diversification rate vs. clade richness	**0.044**	**0.011**	**3.604**	**0.003**
Diversification rate vs. clade area	0.004	0.005	0.695	0.493
Diversification rate vs. clade overlap	−0.075	0.040	−1.867	0.083
Diversification rate vs. climatic-niche rate	**1.307**	**0.426**	**3.071**	**0.008**
Climatic-niche rate vs. clade area	0.018	0.010	1.756	0.101
Climatic-niche rate vs. clade overlap	**−0.252**	**0.042**	**−5.996**	**0.0004**

Variables that exhibit statistically significant associations based on sequential Bonferroni adjustment (Rice 1989) are shown in bold.
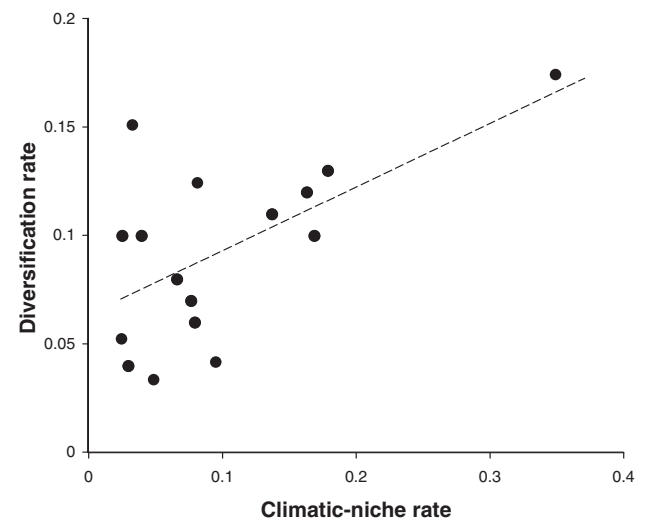

Figure 2 Relationships between rates of species diversification and rates of climatic-niche evolution among the 16 major clades of plethodontid salamanders. Significance of the relationship was analysed using phylogenetic least-squares regression (summary statistics presented in Table 2).
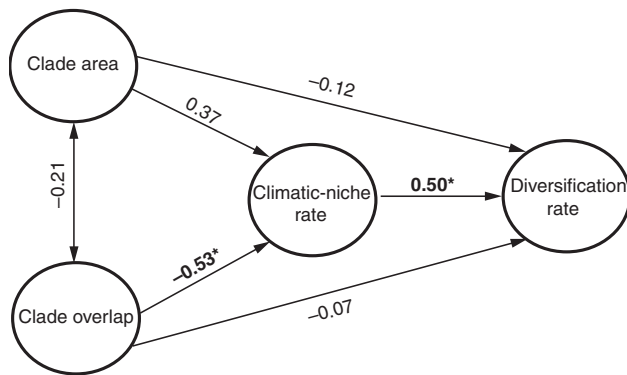
Figure 3 Path analysis depicting direct and indirect effects of clade area, clade overlap and rate of climatic-niche evolution on diversification rate. Standardized correlation coefficients are shown above the arrows connecting variables. Statistically significant correlation coefficients ($P < 0.05$) are depicted in bold and marked with an asterisk.

and when the 16 focal clades are lumped or further subdivided to produce 10 and 22 clades (Table S8) ($r^2 = 0.25$–0.49, $P = 0.01$–0.02). Although the geographically isolated South American clade *Eladinea* contributes strongly to this relationship, rates of diversification and climatic-niche evolution remain marginally correlated when this clade is removed ($r^2 = 0.34$, $P = 0.052$). Among-clade variation in rates of diversification and climatic-niche evolution are not significantly associated with clade area (Table 3), or the degree of sympatry among species within each clade (Table S9). However, rates of diversification and climatic-niche evolution show an inverse relationship with degree of clade overlap (Table 3). Analyses using maximum and minimum values of climatic variables within species (rather than means) gave similar results (Table S10).

Path analysis shows that neither clade area nor clade overlap has a significant, direct effect on rate of diversification (Fig. 3). Furthermore, clade area does not have significant, indirect effects on rate of diversification. Instead, variation in clade diversification rate seems to be directly influenced by the rate of climatic-niche evolution ($\beta = 0.50$). In turn, clade overlap has a smaller, indirect effect on diversification rate through its influence on rates of climatic-niche evolution ($\beta = -0.26$).

DISCUSSION

In this article, we show that higher rates of species diversification are associated with higher rates of climatic-niche evolution. We also show that these higher rates of niche evolution are associated with decreased geographic overlap among clades. Ours is the first study to show that diversification rates are linked to climatic-niche evolution,

and to show that geographic overlap among clades may be important in limiting climatic-niche evolution (i.e. niche conservatism).

We suggest that higher diversification rates are caused by higher rates of climatic-niche evolution, not *vice versa*. We previously showed that sister species of tropical salamanders tend to have more divergent climatic niches than temperate sister species (Kozak & Wiens 2007), suggesting that differences in climatic niches promote speciation in tropical clades, which have higher diversification rates (Table 2; Wiens 2007; Adams *et al.* 2009). This result suggests a mechanism by which climatic-niche evolution might drive speciation and rapid diversification. In contrast, we know of no plausible mechanism by which higher rates of diversification would lead to higher rates of climatic-niche evolution. An older clade with more species might be expected to spread into more climatic niches by chance alone. However, this possibility is accounted for by considering the *rate* of climatic-niche evolution within a clade, and not simply the variance or disparity in climatic values among species. Indeed, we found no relationship between climatic disparity and species richness. Furthermore, although clades that occupy larger areas might be expected to accumulate more species per unit of time and have broader climatic distributions, clade area was uncorrelated with rates of diversification and climatic-niche evolution. Instead, our path analysis shows that the presence of sympatric clades influences rates of diversification by limiting the rate of climatic-niche evolution.

There are two mechanisms by which a higher rate of climatic-niche evolution might increase diversification rate: by increasing speciation or decreasing extinction. The relative contribution of changes in speciation and extinction rates to changes in diversification rates is notoriously difficult to untangle from phylogenies of extant species (reviewed in Ricklefs 2007). Divergence in climatic niches seems to promote speciation in tropical plethodontids (Kozak & Wiens 2007), suggesting that higher speciation rates might drive this pattern. However, climatic-niche lability could also help buffer clades from extinction (e.g. species adapt when climate changes, rather than going extinct; Holt 1990).

Our results may have important implications for explaining the latitudinal diversity gradient, one of the oldest and most controversial patterns in ecology. Many previous analyses showed higher diversification rates in tropical clades (e.g. Cardillo *et al.* 2005; Ricklefs 2006; Wiens 2007; Svenning *et al.* 2008; but see Wiens *et al.* 2006, 2009). However, few studies tested the ecological mechanisms that drive this pattern. Tropical salamander clades also have higher diversification rates (Table 2), and higher rates of climatic-niche evolution ($t = 3.04$, $P = 0.009$). We find these results intriguing for several reasons.

First, many previous studies have suggested that a particular set of environmental conditions (e.g. high energy, productivity and precipitation) drives higher tropical diversity, presumably by allowing larger numbers of species to co-exist locally (reviews in Willig *et al.* 2003; Mittelbach *et al.* 2007). Our results show instead that rapid changes between different climatic regimes underlie this pattern in plethodontids. In tropical plethodontids, regional diversity peaks at higher elevations (*c.* 1000–2500 m), rather than in lowland tropical forests (Wiens *et al.* 2007). Furthermore, analyses of diversification rates among tropical plethodontids show no evidence for higher diversification rates in subclades occurring predominately at lower elevations, where energy and productivity should be highest (Wiens *et al.* 2007). In contrast to expectations, the highest local diversity of plethodontids is in temperate communities, not tropical communities (up to 19 species in temperate North America, vs. a maximum of 11 in lower Central America; García-París *et al.* 2000; Kozak *et al.* 2009). In addition, high local diversity of temperate plethodontids does not seem to be associated with reduced abundance, as they appear to have greater local abundance (García-París *et al.* 2000). Thus, high diversification rates in tropical plethodontids do not seem to be associated with greater environmental carrying capacities of tropical ecosystems, as predicted by some hypothesized explanations for the latitudinal diversity gradient (reviewed in Willig *et al.* 2003; Mittelbach *et al.* 2007).

Second, our results reinforce the importance of Janzen's (1967) hypothesis for explaining high tropical diversity. Janzen suggested that limited climatic seasonality in the tropics might lead to selection for narrow ranges of climatic tolerances in tropical species, leading to greater turnover of species along elevational gradients. Although Janzen's hypothesis is frequently mentioned in discussions of tropical richness (e.g. Mittelbach *et al.* 2007), the specific evolutionary pathway by which climatic zonation leads to greater richness has been uncertain. Our results here suggest that greater climatic zonation in the tropics leads to greater rates of climatic-niche evolution, which leads to higher diversification rates and higher regional species richness. Although some might expect climatic niches of species to evolve more rapidly in the temperate zone (given climatic fluctuations and glaciation), our study demonstrates that shifts between climatic regimes occur more rapidly in tropical regions, where climate is relatively stable (e.g. limited temperature seasonality, limited glaciation).

Third, previous authors have suggested that stronger interactions between species at low latitudes promote high tropical diversity (e.g. Schemske *et al.* 2009). However, we found marginally faster diversification in more spatially isolated clades, just the opposite expected if species interactions drive tropical diversification. Instead, our results suggest that shifts in climatic niches among closely related, non-sympatric species, rather than biotic interactions among coexisting species, promote diversification in tropical plethodontids.

Our results also have important implications for understanding the causes of climatic-niche conservatism. Recent studies suggest that conservatism in climatic tolerances may be important in explaining many different ecological and evolutionary patterns including speciation, species richness, community assembly, responses to global warming and spread of invasive species (reviewed in Wiens & Graham 2005). Many papers on niche conservatism assume (implicitly or explicitly) that range expansion of species and clades is limited by intrinsic tolerances to novel climatic conditions (e.g. physiology), and not species interactions. Indeed, several studies have shown evidence against the role of species interactions in setting range limits and driving patterns of niche conservatism (e.g. Wiens *et al.* 2006; Stephens & Wiens 2009), even in plethodontids (Kozak & Wiens 2006, 2010).

In contrast, our results here show lower rates of climatic-niche evolution in clades having greater geographic overlap with other clades. For example, within many plethodontid clades, species occur at a broadly similar range of elevations (e.g. some clades tend to occur at lower elevations, others at higher elevations; Wiens *et al.* 2007). In contrast, the rapidly evolving clade *Eladinea*, which has very little overlap with other plethodontid clades (Table 1), has evolved to occupy a broad range of elevations and climatic regimes, from near sea level to > 3600 m (IUCN 2010, http://www.iucnredlist.org). Our results suggest that ecological interactions between species of different clades may constrain the large-scale climatic distribution and diversification of these clades over millions of years, even though most plethodontid clades are sympatric with one or more other clades at the local scale (e.g. García-París *et al.* 2000; Kozak *et al.* 2009).

Finally, our study provides some evidence for the importance of species interactions in constraining rates of diversification. Recent studies have suggested that diversification rates will decrease within clades over time, as the 'density' of species increases (e.g. Rabosky & Lovette 2008; Phillimore & Price 2008). However, most have not included ecological information on the local diversity of species or the overlap of clades (but see Weir 2006). For example, Rabosky (2009a) used clade area alone as a surrogate for ecological limits on clade diversification. Here, we show that in plethodontids it is not area, but rather the extensive overlap among coexisting clades that seemingly limits invasion of new climatic niches, and restricts subsequent diversification. We suggest that attempts to investigate the role of species interactions and density in diversification should include relevant ecological information on the overlap between species and/or clades.

In summary, we show that for plethodontid salamanders, rapid species diversification is associated with rapid climatic-niche evolution, and that these rates are particularly rapid in tropical regions and where few clades overlap geographically. These patterns may be important for explaining large-scale patterns of richness among regions and clades, and patterns of climatic-niche evolution and conservatism. Of course, not every group will show identical patterns. For example, in contrast to salamanders, frogs do not show greater climatic divergence between tropical species pairs (Hua & Wiens 2010), nor does every group show faster tropical diversification. However, the generality of these findings can be readily tested in other groups using similar methods and by integrating time-calibrated phylogenies with data on species climatic distributions.

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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? *Proc. R. Soc. Lond. B*, 276, 2729–2738.

Amphibia Web: Information on amphibian biology and conservation. (2010). [Web application]. AmphibiaWeb, Berkeley. Available at: http://www.amphibiaweb.org. Last accessed 1 March 2010.

Baldwin, B.G. & Sanderson, M.J. (1998). Age and rate of diversification of the Hawaiian silversword alliance (Compositae). *Proc. Natl. Acad. Sci. USA*, 95, 9402–9406.

Butler, M.A. & King, A.A. (2004). Phylogenetic comparative analysis: a modeling approach for adaptive evolution. *Am. Nat.*, 164, 683–695.

Cardillo, M., Orme, C.D.L. & Owens, I.P. (2005). Testing for latitudinal bias in diversification rates: an example using New World birds. *Ecology*, 86, 2278–2287.

Crisp, M.D., Arroyo, M.T.K., Cook, L.G., Gandolfo, M.A., Jordan, G.J., McGlone, M.S. *et al.* (2009). Phylogenetic biome conservatism on a global scale. *Nature*, 458, 754–756.

Evans, M.E., Smith, S.A., Flynn, R.E. & Donoghue, M.J. (2009). Climate, niche evolution, and diversification of the 'bird-cage' evening primroses (*Oenothera*, sections *Anogra* and *Kleinia*). *Am. Nat.*, 173, 225–240.

García-París, M., Good, D.A., Parra-Olea, G. & Wake, D.B. (2000). Biodiversity of Costa Rican salamanders: implications of high levels of genetic differentiation and phylogeographic structure for species formation. *Proc. Natl. Acad. Sci. USA*, 97, 1640–1647.

Harmon, L.J., Weir, J., Brock, C., Glor, R.E. & Challenger, W. (2008). GEIGER: investigating evolutionary radiations. *Bioinformatics*, 24, 961–964.

Hijmans, R.J., Cameron, S.E., Parra, J.L., Jones, P.G. & Jarvis, A. (2005). *The WorldClim Interpolated Global Terrestrial Climate Surfaces, Version 1.3*. Available at: http://www.worldclim.org.

Holt, R.D. (1990). Microevolutionary consequences of climate change. *Trends Ecol. Evol.*, 9, 311–315.

Hua, X. & Wiens, J.J. (2010). Latitudinal variation in speciation mechanisms in frogs. *Evolution*, 64, 429–443.

IUCN, Conservation International, and NatureServe. (2010). Global Amphibian Assessment. Available: http://globalamphibians.org. Last accessed 1 March 2010.

Jackson, D.A. (1993). Stopping rules in principal components analysis: a comparison of heuristical and statistical approaches. *Ecology*, 74, 2204–2214.

Janzen, D.H. (1967). Why mountain passes are higher in the tropics. *Am. Nat.*, 101, 233–249.

Knouft, J.H., Losos, J.B., Glor, R.E. & Kolbe, J.J. (2006). Phylogenetic analysis of the evolution of the niche in lizards of the Anolis sagrei group. *Ecology*, 87, S29–38.

Kozak, K.H. & Wiens, J.J. (2006). Does niche conservatism promote speciation? A case study in North American salamanders. *Evolution*, 60, 2604–2621.

Kozak, K.H. & Wiens, J.J. (2007). Climatic zonation drives latitudinal variation in speciation mechanisms. *Proc. R. Soc. Lond. B*, 274, 2995–3003.

Kozak, K.H. & Wiens, J.J. (2010). Niche conservatism drives elevational diversity patterns in Appalachian salamanders. *Am. Nat.*, 176, 40–54.

Kozak, K.H., Mendyk, R.W. & Wiens, J.J. (2009). Can parallel diversification occur in sympatry? Repeated patterns of body-size evolution in co-existing clades of North American salamanders. *Evolution*, 63, 1769–1784.

Magallón, S. & Sanderson, M.J. (2001). Absolute diversification rates in angiosperm clades. *Evolution*, 55, 1762–1780.

Mittelbach, G.G., Schemske, D.W., Cornell, H.V., Allen, A.P., Brown, J.M., Bush, M.B. *et al.* (2007). Evolution and the latitudinal diversity gradient: speciation, extinction and biogeography. *Ecol. Lett.*, 10, 315–331.

Mitter, C., Farrell, B. & Wiegmann, B. (1988). The phylogenetic study of adaptive radiation: has phytophagy promoted insect diversification? *Am. Nat.*, 132, 107–128.

Moritz, C., Patton, J.L., Schneider, C.J. & Smith, T.B. (2000). Diversification of rainforest faunas: an integrated molecular approach. *Annu. Rev. Ecol. Syst.*, 31, 533–563.

O'Meara, B.C., Ané, C., Sanderson, M.J. & Wainwright, P.C. (2006). Testing for different rates of continuous trait evolution using likelihood. *Evolution*, 60, 922–933.

Paradis, E., Claude, J. & Strimmer, K. (2004). APE: analyses of phylogenetics and evolution in R language. *Bioinformatics*, 20, 289–290.

Pearson, R.G. & Dawson, T.P. (2003). Predicting the impact of climate change on the distribution of species: are bioclimate envelope models useful? *Glob. Ecol. Biogeogr.*, 12, 361–371.

Peterson, A.T., Soberón, J. & Sánchez-Cordero, V. (1999). Conservatism of ecological niches in evolutionary time. *Science*, 285, 1265–1267.

Phillmore, A.B. & Price, T.D. (2008). Density-dependent cladogenesis in birds. *PLoS Biol.*, 6, e71.

Pybus, O.G. & Harvey, P.H. (2000). Testing macro-evolutionary models using incomplete molecular phylogenies. *Proc. R. Soc. Lond. B*, 267, 2267–2271.

Rabosky, D.L. (2006). LASER: a maximum likelihood toolkit for detecting temporal shifts in diversification rates from molecular phylogenies. *Evol. Bioinform.*, 173, 662–674.

Rabosky, D.L. (2009a). Ecological limits on clade diversification in higher taxa. *Am. Nat.*, 173, 662–674.

Rabosky, D.L. (2009b). Ecological limits and diversification rate: alternative paradigms to explain the variation in species richness among clades and regions. *Ecol. Lett.*, 12, 735–743.

Rabosky, D.L. & Lovette, I.J. (2008). Density dependent diversification in North American wood-warblers. *Proc. R. Soc. Lond. B*, 275, 2363–2371.

Raxworthy, C.J., Ingram, C.M., Rabibisoa, N. & Pearson, R.G. (2007). Applications of ecological niche modeling for species delimitation: a review and empirical evaluation using day geckos (*Phelsuma*) from Madagascar. *Syst. Biol.*, 56, 907–923.

Rice, W.R. (1989). Analyzing tables of statistical tests. *Evolution*, 43, 222–225.

Ricklefs, R.E. (2006). Global variation in the diversification rate of passerine birds. *Ecology*, 87, 2468–2478.

Ricklefs, R.E. (2007). Estimating diversification rates from phylogenetic information. *Trends Ecol. Evol.*, 22, 601–610.

Sanderson, M.J. (2003). r8s: inferring absolute rates of molecular evolution and divergence times in the absence of a molecular clock. *Bioinformatics*, 19, 301–302.

Schemske, D.W., Mittelbach, G.G., Cornell, H.V., Sobel, J.M. & Roy, K. (2009). Is there a latitudinal gradient in the importance of biotic interactions? *Ann. Rev. Ecol. Evol. Syst.*, 40, 245–269.

Seddon, N., Merrill, R.M. & Tobias, J.A. (2008). Sexually-selected traits predict patterns of species richness in a diverse clade of suboscine birds. *Am. Nat.*, 171, 620–631.

Simpson, G.G. (1944). *Tempo and Mode in Evolution*. Columbia University Press, New York.

Smith, S.A. & Beaulieu, J.M. (2009). Life history influences rates of climatic niche evolution in flowering plants. *Proc. R. Soc. Lond. B*, 276, 4345–4352.

Sobel, J.M., Chen, G.F., Watt, L.R. & Schemske, D.W. (2010). The biology of speciation. *Evolution*, 64, 295–315.

Stephens, P.R. & Wiens, J.J. (2009). Bridging the gap between historical biogeography and community ecology: niche conservatism and community structure in emydid turtles. *Mol. Ecol.*, 18, 4664–4679.

Svenning, J.-C., Borchsenius, F., Bjorholm, S.W. & Balslev, H. (2008). High tropical net diversification drives the New World latitudinal gradient in palm (Arecaceae) species richness. *J. Biogeogr.*, 35, 394–406.

Warren, D.L., Glor, R.E. & Turelli, M. (2008). Environmental niche equivalency versus conservatism: quantitative approaches to niche evolution. *Evolution*, 62, 2868–2883.

Weir, J.T. (2006). Divergent timing and patterns of species accumulation in lowland and highland Neotropical birds. *Evolution*, 60, 842–855.

Wiens, J.J. (2007). Global patterns of species richness and diversification in amphibians. *Am. Nat.*, 170, S86–S106.

Wiens, J.J. & Graham, C.H. (2005). Niche conservatism: integrating evolution, ecology, and conservation biology. *Ann. Rev. Ecol. Evol. Syst.*, 36, 519–539.

Wiens, J.J., Graham, C.H., Moen, D.S., Smith, S.A. & Reeder, T.W. (2006). Evolutionary and ecological causes of the latitudinal diversity gradient in hylid frogs: treefrog trees unearth the roots of high tropical diversity. *Am. Nat.*, 168, 579–596.

Wiens, J.J., Parra-Olea, G., Garcia-Paris, M. & Wake, D.B. (2007). Phylogenetic history underlies elevational patterns of biodiversity in tropical salamanders. *Proc. R. Soc. Lond. B*, 274, 919–928.

Wiens, J.J., Sukumaran, J., Pyron, R.A. & Brown, R.M. (2009). Evolutionary and biogeographic origins of high tropical diversity in Old World frogs (Ranidae). *Evolution*, 63, 1217–1231.

Wiens, J.J., Ackerly, D.D., Allen, A.P., Anacker, B.L., Buckley, L.B., Cornell, H.V. *et al.* (2010). Niche conservatism as an emerging principle in ecology and conservation biology. *Ecol. Lett.*, 13, 1310–1324.

Willig, M.R., Kaufman, D.M. & Stevens, R.D. (2003). Latitudinal gradients of biodiversity: pattern, process, scale, and synthesis. *Ann. Rev. Ecol. Evol. Syst.*, 34, 273–309.

SUPPORTING INFORMATION

Additional Supporting Information may be found in the online version of this article:

Table S1 Results of phylogenetic least-squares regressions for the 16 clades in Table 1, showing the robustness of the main results to alternate root ages for Plethodontidae (48 and 69 Myr vs. 61 Myr).

Table S2 Constant rate tests and Monte Carlo constant rate tests for species diversification in each of the 16 focal clades (root age = 48 Myr).

Table S3 Constant rate tests and Monte Carlo constant rate tests for species diversification in each of the 16 focal clades (root age = 61 Myr).

Table S4 Constant rate tests and Monte Carlo constant rate tests for species diversification in each of the 16 focal clades (root age = 69 Myr).

Table S5 Relationship between estimated values of the gamma statistic (from constant rates test) for each clade and clade age, net diversification rate and species richness, using three different root ages for Plethodontidae.

Table S6 Comparison of the fit of Brownian motion (BM) and Ornstein-Uhlenbeck (OU) models of evolution to climatic variation in the 16 focal clades of plethodontid salamanders in Table 1.

Table S7 The proportion of each clade's total area in which two or more of its species are sympatric (prop. overlap) and the maximum number of species from each clade that are regionally sympatric (max. sympatric).

Table S8 Alternative clade divisions that lump (10 clades) or further subdivide (22 clades) many of the 16 focal clades in Table 1. Clade ages, net diversification rates (using $e = 0.45$), and rates of climatic-niche evolution are based on a root age of 61 Myr for Plethodontidae.

Table S9 Results of phylogenetic least-squares regressions of the association between diversification rate, rate of climatic niche-evolution, two indices for geographic overlap of species within clades: the proportion of the clade's range in which two or more species have overlapping ranges (prop. overlap) and the maximum number of sympatric species within the clade (max. sympatric).

Table S10 Results of phylogenetic least-squares regressions employing maximum and minimum values (within each species) for the 20 environmental variables, rather than mean values. Rates of climatic-niche evolution for each clade were calculated by summing the diagonal elements of the evolutionary variance-covariance matrix for PC1-PC3 (see text). Clade divisions follow Table 1.