Title

The contribution of climate and phylogeny to the richness pattern of freshwater fish of North America

Author

Rothstein-Kightly, Kevin Michael

Publication Date

2016

UNIVERSITY OF CALIFORNIA,
IRVINE

The contribution of climate and phylogeny to the richness pattern of freshwater fish of North America

THESIS

submitted in partial satisfaction of the requirements
for the degree of

MASTER OF SCIENCE

in Ecology and Evolutionary Biology

by

Kevin Michael Rothstein-Kightly

Thesis Committee:
Professor Bradford A. Hawkins, Chair
Professor Jennifer B.H. Martiny
Associate Professor Matthew E.S. Bracken
Assistant Professor Cascade J.B. Sorte

2016

DEDICATION

To

David Moss Junior, whose love and support has made
the toughest of obstacles easier to overcome

Friends, whose diversity of interests and perspectives
continues to shape my philosophy of life

Mentors, whose enthusiasm and love of knowledge
have inspired me to pursue my passions

Family, who ignited the fire that drives me to
move forward in the face of adversity

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

Page

ACKNOWLEDGMENTS

I would like to express the deepest appreciation to my committee chair, Professor Bradford A. Hawkins, for his mentorship throughout my graduate school career. Without his guidance and persistent help, this thesis would not have been possible.

I would like to thank my committee members, Professor Jennifer B.H. Martiny, Associate Professor Matthew E.S. Bracken, and Assistant Professor Cascade J.B. Sorte, whose guidance and feedback challenged me to evaluate the quality of my research and improve the clarity of this thesis.

In addition, I would like to thank fellow graduate student LuAnna L. Dobson, Ph.D. candidate, for her input on some of the technical aspects of my experiments.

I would like to thank the University of California, Irvine—specifically the Department of Ecology and Evolutionary Biology—for its financial support. This includes Francisco J. Ayala, whose graduate fellowship made it possible for me to attend UC Irvine. Without this assistance, I would not have had this incredible opportunity to undertake research and write this thesis.

ABSTRACT OF THE THESIS

The contribution of climate and phylogeny to the richness pattern of freshwater fish of North America

By

Kevin Michael Rothstein-Kightly

Master of Science in Ecology and Evolutionary Biology

University of California, Irvine, 2016

Professor Bradford A. Hawkins, Chair

Over large spatial scales, debate remains concerning the relative importance of ecological and evolutionary processes as drivers of species richness gradients. Ecological hypotheses propose that contemporary environmental variables are the primary drivers of species diversity, whereas evolutionary hypotheses, such as the metabolic theory and the time-for-speciation hypothesis, propose diversification rates or time. Previous work has compared ecological and evolutionary variables to quantify relationships with richness of three North American freshwater fish families and concluded that their richness gradients are driven by contemporary climate. We compiled the first time-calibrated phylogenetic tree consisting of 836 native North American freshwater fish species to evaluate both ecological and evolutionary hypotheses as contributors to species richness. Our results showed that climate, measured as actual evapotranspiration, is the strongest predictor of richness. In contrast, family ages and diversification rates are poor predictors of species richness, providing no support for the time-for-speciation hypothesis or metabolic theory. However, the contribution of species mean root distance, a phylogenetic metric, is greater than previously documented. We also found that climatic and phylogenetic variables together explain most of the variation in the broad-scale richness pattern. We further demonstrate that mean annual temperature indirectly shapes richness through species mean root distance and actual evapotranspiration, which we interpret as the result of local extirpations of fish species in response to Pleistocene glacial cycles. We argue that including historical and evolutionary processes alongside climate provides a stronger explanation for richness gradients than either considered separately.

INTRODUCTION

The many hypotheses proposed in the past 200 plus years to explain geographic patterns of species richness broadly encompass ecological processes, evolutionary rates, or history/time (Currie et al., 2004; Mittelbach et al., 2007). In the past thirty years in particular, there has been a large accumulation of literature that examines these explanations (see e.g., Field et al., 2009). Although most agree that species distributions are influenced by the past and present, teasing apart ecological and evolutionary influences is challenging, and the relative importance of historical and contemporary processes remains unresolved. More recent work highlights the importance of traits for understanding distributions and links processes operating over ecological and evolutionary scales. Phylogenetic niche conservatism is a widespread occurrence, manifested across evolutionary time scales and influencing how species respond to current and future environmental conditions, and thereby can directly and indirectly influence diversity gradients (Wiens et al., 2010). Even so, quantifying relationships between species richness patterns and contemporary components of environment, particularly climate, and incorporating legacies of evolutionary rates and history is an important step in understanding the relative contributions of ecology and evolution in driving species richness.

Kerr and Currie (1999) designed an early analysis that quantitatively evaluated the relationships of patterns of species richness with both the contemporary environment and an evolutionary process. They gridded species richness patterns of a family of terrestrial beetles and three families of freshwater fish across North America against two climatic variables (potential and actual evapotranspiration) and assemblage mean root distance, a measure of evolutionary development based on the topology of the phylogenetic tree of each clade. Their goal was to determine which set of metrics generated the strongest correlation with richness to evaluate whether it was necessary to invoke an evolution-based process or if contemporary climate was a sufficient explanation for the observed diversity gradients. Based on four sets of three simple correlations, they concluded that there is little reason to invoke evolutionary processes for understanding the richness patterns within any of the families.

The analysis performed by Kerr and Currie (1999) was necessarily simple and narrowly focused, particularly from the evolutionary perspective, due to the paucity of detailed phylogenies for large clades available at the time. The state of phylogenetic trees required the use of only the simplest evolutionary metric. However, much more phylogenetic information is now available for many large groups, and it is possible to estimate branch lengths as well as simple tree topology in most cases, expanding the range of potential evolutionary processes that can be explored. Finally, Kerr and Currie considered climatic and evolutionary influences on diversity patterns as alternatives, a point of view that has been argued elsewhere in the literature (see e.g., Algar et al., 2008; Fraser & Currie, 1996), whereas it has become apparent that climate and evolution can be simultaneously linked to species richness gradients via at least two mechanisms, metabolic theory (Brown et al., 2004) and phylogenetic niche conservatism (Wiens & Donoghue, 2004).

Metabolic theory represents a specific explanation for the more general diversification rate and evolutionary speed hypotheses (Rohde, 1992) by proposing that diversification rates are accelerated at higher temperatures via enzyme kinetics at the organismal level (Allen et al., 2002; Brown et al., 2004). Although there is evidence of increased molecular evolution in plants at higher temperatures (Wright et al., 2003; Davies et al., 2004; Wright et al., 2006), Schluter (2016) provides counter-evidence that speciation rates are currently highest at high latitudes and argues that speciation rates in general are not linked to climate in the simple way envisaged by metabolic theory. Even so, this does not mean that speciation rates play no role in explaining diversity at some scales, or that they are independent of climate.

The relationship between niche conservatism and diversity gradients derives from the ‘tropical’ origin of most major clades, the climatic history of Earth (Jablonski et al., 2006; Wiens & Donoghue, 2004; Schluter, 2016) and the fact that many modern species carry ancestral traits can be traced back to when and where clades originated. Thus, the tropical conservatism hypothesis, based on the general process of phylogenetic niche conservatism, proposes that diversity for most groups is lower in cold, dry climates, both because mid-Cenozoic global cooling and drying selectively extirpated cold-intolerant species from the north latitudes and because there has been more time for warm-adapted groups to diversify in the globally widespread warm climates that date back to at least the early Cretaceous (Behrensmeyer et al., 1992). Under this mechanism, climate has always driven diversity patterns, including the “snapshot” pattern we see now. This can also form the basis for projected effects of climate change on diversity gradients (Wiens et al., 2010).

We undertake the most complete analysis of the relative importance of ecological and evolutionary drivers in determining diversity patterns of North American freshwater fish. We take advantage of the improved phylogenetic information now available to compile the first time-calibrated, species-level phylogenetic tree for native North American freshwater fish and to generate metrics associated with a broader range of evolutionary mechanisms. We also approach this question from a somewhat different angle than Kerr and Currie (1999); whereas they saw contemporary climate and evolution as alternative explanations, we consider the possibility that they are two manifestations of a single mechanism and thus both contribute to an understanding of diversity gradients. We focus on 836 native freshwater fish species currently found in North America north of Mexico, since these species represent a diverse vertebrate group that is understudied relative to the intensively studied birds and mammals (but see Smith 2010; Griffiths 2010, 2014, 2015). A focus on freshwater fish also allows us to make a direct comparison with the conclusions of Kerr & Currie (1999).

METHODS

Watershed Data and Species Richness

Species distributions delimited at the watershed grain were downloaded for the conterminous United States from NatureServe (2010). For Alaska and Canada, species distributions from *The Freshwater Fishes of Alaska* (Scott & Crossman, 1985) and *Freshwater Fishes of Canada* (Morrow, 1980) were digitized. Watershed data for Alaska and Canada were downloaded from Watermolen (1999) and the Canada Centre for Mapping and Earth Conservation (2003), respectively. One species, *Lethenteron alakesnse*, was absent from *The Freshwater Fishes of Alaska*, so its distribution was digitized from an online map (NatureServe, 2013). Exotic species and the records of North American species that were introduced by human activity into watersheds where they did not naturally occur were excluded from the data set. A presence-absence matrix of 2,892 watersheds by 836 species was generated to quantify the species richness pattern (Figure 1) and calculate community-level phylogenetic metrics.

Phylogeny and Evolutionary Metrics

A family-level phylogeny was extracted from the DeepFin molecular phylogeny (Betancur-R *et al.*, 2013) to form the backbone of a species-level phylogeny generated using clade-specific publications (Figure 2). Two families, Goodiedae and Petromyzontidae, were absent from the DeepFin phylogeny, and their positions in the family-level tree were determined by referencing other publications. If a species' position in the tree was lacking in the literature, its location was designated as a polytomy within its genus. Phylogenetic metrics were extracted from the phylogeny to measure the contribution of evolutionary history to species richness. Diversification rates were calculated using several steps. First, family ages were estimated from the DeepFin phylogeny. Second, family ages were mapped onto nodes in the species-level phylogeny so that branch lengths could be calculated in Phylocom (Webb *et al.*, 2008). If only one species was present in a family, so that its family age was not accurately represented, a placeholder node was created in order to assign the correct family age. Last, these branch lengths were used to calculate diversification as the inverse of the Equal Splits measure for taxa at terminal nodes (Belmaker & Jetz, 2015).

Two alternative metrics were also calculated: 1) species root distance—the number of nodes separating a species from the root of the phylogeny—which is considered a measure of how derived a species is (Kerr & Currie, 1999), and 2) family root distance, assigned to species as the number of nodes from the base of the phylogeny the basal node of the family to which each species belongs. Family root distance measures if species in an assemblage are from more basal or more derived clades (Hawkins *et al*., 2006). When polytomies were present in part of the phylogeny, an average node value, based on the possible number of configurations, was applied to all species in that clade. Mean species and family root distances were then calculated for each watershed.

Environmental Data

Environmental data were downloaded from multiple online sources to evaluate climatic and primary productivity relationships with fish species richness. Temperature and precipitation data were obtained from BioClim (Hijmans *et al.*, 2005) at a resolution of 10-arc minutes, comprising annual mean temperature, maximum temperature of the warmest month, minimum temperature of the coldest month, temperature seasonality, annual precipitation, and precipitation seasonality. Net primary productivity (NPP) for the years 2000-2014 was obtained from the MOD17 project (Zhao *et al.,* 2005), and actual evapotranspiration (AET) and potential evapotranspiration (PET) for the years 2000-2013 were obtained from the MOD16 project (Mu *et al.*, 2011).

Analyses

Linear Regression

For each watershed, the means of all environmental and phylogenetic variables were calculated and used as predictors in regression models of species richness. Tiny coastal watersheds with missing phylogenetic, environmental, or richness information were excluded from the analysis. We regressed environmental and phylogenetic variables against richness to compare their relative predictive strength. For potential explanatory variables that were collinear (Table S1) with a variance inflation factor of 2.5 or greater (Table S2), the choice of which variable to remove from the analysis was based on the strength of its relationships with richness. The excluded variables were maximum temperature of the warmest month, minimum temperature of the coldest month, temperature seasonality, PET, and NPP.

It has recently been discovered that the repeated species co-occurrences almost always found in broad-scale multiple species distribution data can potentially generate artefactual structure in species-derived traits and attributes aggregated at the community/assemblage level (Zelený & Schaffers, 2012; Peres-Neto et al., 2016). The watershed phylogenetic metrics we used to evaluate influences of evolutionary processes on the continental fish richness gradient could suffer from this problem. To determine if spatial patterns in mean diversification rate or species/family mean root distances could be generated simply by high levels of repeated species co-occurrences, we randomized the values for each species for each phylogenetic variable 100 times and recalculated the mean values for each watershed with each set of randomized values. We then regressed species richness onto each set of means and compared the observed model fits with the randomized fits. Observed r^2 values less than two standard deviations from the means of the randomized models would indicate that ecological interpretations of model fits for these predictors could be "too good to the true" (Zelený & Schaffers, 2012).

Partial Regression and Spatial Autocorrelogram

To evaluate the independent and combined predictive power of ecological and evolutionary hypotheses on richness, a partial regression analysis was run in Spatial Analysis in Macroecology (SAM) (Rangel *et al.*, 2010). One climatic and one phylogenetic variable were introduced to the model at a time, starting with the variables most strongly correlated with richness. Variables were not included if their addition to the model did not increase the predictive power by more than 1%. To evaluate the adequacy of the models to explain the spatial pattern in richness, a spatial autocorrelogram of the residuals of the richness pattern was generated after including climatic, phylogenetic and spatial predictors.

Path Model

In the final step of the analysis, we generated a path model to further explore the potential causal relationships among climate, phylogeny, and richness. The models were created in the laavan package (Rosseel, 2012) in R (R Core Team, 2008), and the best-fit model was selected by ranking models by their AIC values. Since unexplained spatial autocorrelation can represent potentially important unmeasured effects on richness, the residuals of richness from the best-fit model were analyzed in SAM to generate eigenvectors that were added to the model to allow us to estimate a standardized path coefficient for these unknown variables. The eigenvector with the least amount of spatial overlap with the climatic and phylogenetic predictors was used as a spatial filter that was introduced into the path model.

RESULTS

Linear Regressions

Single-predictor regressions identified climatic variables as generally stronger predictors of species richness than phylogeny, although species mean root distance (MRD_S) ranked second only to actual evapotranspiration (Figure 3). AET was the only variable of the eight evaluated that explained over half of the variance in species richness (Figure 3a), whereas the best phylogenetic predictor explained approximately a third of the variance (Figure 3e). The remaining climatic variables individually explained between 18% - 27% of the variance, whereas the remaining phylogenetic variables individually explained < 10% of the variance in richness.

The observed r^2 for the relationship between MRD_S and richness was 3.22 SD units stronger than the mean randomized model fit ($r^2 = 0.060$), providing confidence that the phylogenetic structure of the fish communities in each watershed is not simply a product of the patterns of species co-occurrences across watersheds. For the remaining three phylogenetic variables, the observed r^2 values were < 0.367 SD from the null means, making ecological inference of these variables highly uncertain.

Partial Regression

The best-fit model for the partial regression analysis included climatic variables of mean actual evapotranspiration and mean annual temperature and phylogenetic variables of mean species root distance and mean family root distance. Notably, diversification rate was a poor correlate ($r < 0.001$) with richness and was not selected to be in the model. As suggested by the simple regressions, climate was a better predictor of richness than phylogeny. Further, climate independently explained almost three time more variation in richness than that explained independently by phylogeny. Even so, phylogeny and climate together explained almost three-quarters of the variation in species richness (Figure 4).

Residual Spatial Structure of Richness

The spatial autocorrelogram revealed that AET and MRD_s, followed by annual mean temperature and mean family root distance, were the predictors that best explained the spatial pattern in richness (Figure 5). Most of the unexplained residual richness was in the smallest distance classes, with low autocorrelation at moderate scales and none at the largest scale. Thus, the complete partial regression model captured the continental species richness gradient extremely well but had incomplete explanatory power at scales smaller than 2000 km.

Path Model

The best-fit model included annual mean temperature, precipitation seasonality, AET, MRD_s, and the spatial filter (Figure 6). The model was not significantly different from the structure of the data, indicating an adequate fit ($p = 0.138$). Similar to the results of the partial regression analysis, the variables selected by this model explained 69% of the variance in richness. The strongest relationships with richness were those of AET and MRD_s. There was an indirect effect of annual mean temperature on richness through AET, but AET was still a strong predictor of richness. However, the direct path between annual mean temperature and MRD_s was one of the two strongest paths in the model. Thus, the collinearly of climate and phylogeny that was present in the partial regression analysis was partially due to climate indirectly acting through phylogenetic variables, with AET having the strongest direct path to richness (0.66) whereas mean annual mean temperature had a weak direct path to richness but an equivalent total effect (0.67) due in part to an indirect path via phylogenetic structure.

DISCUSSION

We undertook the most complete analysis of the relative importance of ecological and evolutionary drivers on contemporary fish diversity gradients across North America by exploring how phylogenetic and contemporary climatic variables contribute to the broad-scale richness pattern. Similar to Kerr and Currie (1999), we found that both climatic and phylogenetic variables contribute to the variation in richness, although our interpretation differs from theirs.

Our finding that AET is the strongest determinant of species richness is consistent with many studies (summarized in Hawkins et al., 2003), showing that spatial species richness usually correlates well with water-energy variables in both terrestrial and freshwater environments. Although the underlying mechanism(s) for this correlation remain(s) unresolved, it has been shown that climate can affect ecological processes such as carrying capacities, which in turn affect evolutionary processes (Pontarp & Wiens, 2016). Indeed, the emergence of phylogenetic niche conservation as a potential evolutionary mechanism underlying broad-scale diversity gradients provides a powerful way to link past and current climate with diversity (see Wiens et al., 2010 for a recent review). And, the overlap of climate and phylogeny observed in our partial regression analysis is consistent with this explanation, which argues that climate has always influenced diversity, and so the co-varying patterns of climate and diversity that we see now are just a “snapshot” of a continual process driven by patterns of climate change over evolutionary and ecological time frames with limited abilities of organisms to adapt. But interestingly, based on the results of our path model analysis, the components of climate that drive evolutionary responses of fish to climate may differ from those driving the ecological response. The strongest link to the simple topological metric of the level of evolutionary development of fish assemblages, MRD_S, was annual mean temperature, whereas the strongest direct link to richness itself was AET, which is also strongly correlated with primary productivity. This provides indirect evidence that current patterns of fish richness may be in part food limited and in part influenced by the pattern of global cooling across the North Hemisphere that began in the mid-Eocene and became especially strong during the Pleistocene glacial cycles. Cenozoic climate change has been implicated in North American diversity gradients for terrestrial groups (Latham & Ricklefs, 1993; Hawkins et al., 2006; Hawkins et al., 2011). However, the case for niche conservatism as a partial driver of diversity is currently circumstantial, since we have no evidence of what, if any, relevant traits have been conserved. One candidate is dispersal ability, which is associated with the recolonization of fish into areas exposed by the retreat of the ice sheets after the Latest Glacial Maximum (Smith et al., 2010; Griffiths, 2010, 2015). The pattern of evolution of migratory behavior in fish would be a fruitful starting point for understanding to what extent niche conservation, climate change, and contemporary climates interact to influence the diversity gradient.

The niche conservatism explanation for diversity gradients is often linked to time for speciation (Weins & Donoghue 2004), arguing that older clades have had longer to diversify, but these clades are constrained to the older, warmer climates globally widespread in the Cretaceous and early Cenozoic. Under this scenario, we would expect to see older families in areas of high species richness and younger families in areas of low species richness (see e.g., Wiens & Donoghue, 2004). However, we found little if any association between family ages and species richness. This suggests that the standard explanation of niche conservatism determining time for speciation for clades in high diversity areas does not apply to freshwater fish.

The alternative evolutionary hypothesis for diversity patterns, differential diversification rates across space, also received no support, since average diversification rate had no relationship with richness. This is similar to the results of previous studies (Smith et al., 2010; Griffiths, 2015) that used general measures of diversification rates to examine broad-scale patterns in freshwater fish. We cannot rule out the possibility that the metric we used is not the strongest test of this hypothesis, but based on the available evidence, we conclude that rate-based hypotheses such as metabolic theory are probably insufficient to explain the current diversity gradient.

Although we reject both speciation rates and time, we were able to explain some of the variation in richness through the non-time-calibrated phylogenetic metric of root distance. The observation that areas of high richness tend to have higher values of root distance suggests that there is a greater amount of diversification of lineages in these areas, despite the fact that the diversification rate metric has no explanatory power. There are two possible reasons for this. First, average watershed-level diversification rates may be confounded by dispersal of fish species among watersheds following their diversification, so that current local rates are not indicators of the spatial distribution of rates in the past. Second, if this dispersal occurs but is limited across isolated watersheds over relatively short time periods, the observed phylogenetic structure could be the consequence of highly variable extinction rates during the Pleistocene. In this case, the southeastern U.S.A. is high in diversity not only because of a benign climate but because most or all clades in areas north and west were extirpated by glaciations and periods of aridity, leaving a legacy of low diversity and low root distances due to climatic instability (see Latham & Ricklefs, 1993 for the application of this explanation to trees). Under this scenario the primary historical drivers of diversity lie in the Pleistocene rather than the early Cenozoic, but quantifying the timing of key events is not currently possible given the current incomplete state of the fish phylogeny.

The observation that richness for freshwater fish is highest in the southeast United States, compared to the global pattern of richness decreasing with latitude, demonstrates how diversity patterns can differ at different spatial scales in both focus and extent (Brown & Maurer, 1989; Rahbek, 2004; Field et al., 2009). Oberdorff *et al.* (1995) concluded that much of the variation in global-scale freshwater fish richness can be predicted by river size (species-area hypothesis; MacArthur & Wilson, 1963) followed by primary productivity, regardless of glacial history, topography, or climate; the influence of surface area is further supported for riverine freshwater fish globally (Oberdorff et al., 2011). Our partial regression analysis also revealed scale dependence in the diversity gradient; it accounted for virtually all of the autocorrelation at the largest spatial scales, indicating that climatic and phylogenetic variables are the adequate explanations for species richness at the continental scale. However, there was still unexplained spatial autocorrelation at scales below ca. 500 km. This residual spatial autocorrelation is likely due to biotic and abiotic factors at local and regional scales that we did not account for in our analysis. Depending on species characteristics, such as life history traits, reproductive strategies, and feeding strategies, along with the effects of species interactions, such as predation and intra- and interspecific competition, a stream or lake can support a variable number of niches. In river systems, headwater streams surrounded by riparian vegetation are relatively nutrient and species poor, whereas mid-size rivers have much higher species richness and nutrient-rich waters as a result of the shredding of upstream allochthonous matter by aquatic insects (Ross, 2013). At the local and regional scales, lakes with larger surface area are associated with higher species richness (Griffiths, 1997). In addition, anthropogenic activities, such as agriculture and urbanization, negatively affect the survival and health of ecosystems by polluting waters with runoff and leading to the removal and fragmentation of habitats (McDonald, 2011). By considering species traits, species interactions, habitat characteristics, and anthropogenic impacts, the "unexplained" variation in species richness at local and regional level can be further explored.

In summary, with the accumulation of new ideas and better phylogenetic data over the last two decades, we were able to take a more nuanced approach than Kerr and Currie (1999) to address drivers of fish species richness. Similar to them we find that current climate remains the strongest correlate of the contemporary richness pattern, but we also found that phylogeny further explains variation in species richness than climate alone. WE also do not take an either/or point of view, arguing that that evolutionary and climatic history play a larger role than recognized by Kerr and Currie (1999). Our results further suggest that climate and phylogeny are not independent; at least some of climate is influencing phylogeny in shaping species richness patterns, most likely via climate change in the late Cenozoic. It is becoming evident that when attempting to understand drivers of contemporary richness patterns, historical hypotheses combined with ecological hypotheses provide stronger explanations than either alone.

REFERENCES

Algar AC, Kerr JT, Currie DJ (2008) Evolutionary constraints on regional faunas: whom, but not how many. *Ecology Letters*, **12**, 57-65.

Allen AP, Brown JH, Gillooly JF (2002) Global biodiversity, biochemical kinetics, and the energetic-equivalence rule. *Science*, **297**, 1545-1548.

Behrensmeyer AK, Damuth JD, DiMichele WD, Potts R, Sues H-D, Wing SL (1992) *Terrestrial ecosystems through time: evolutionary paleoecology of terrestrial plants and animals*. University of Chicago Press, Chicago.

Belmaker J, Jetz W (2015) Relative roles of ecological and energetic constraints, diversification rates and region history on global species richness gradients (ed Arita H). *Ecology Letters*, **18**, 563–571.

Betancur-R R, Broughton RE, Wiley EO et al. (2013) The Tree of Life and a New Classification of Bony Fishes. *PLoS Currents*.

Brown JH, Gillooly JF, Allen AP, Savage VM & West GB (2004) Toward a metabolic theory of ecology. *Ecology*, **85**, 1771–1789.

Brown JH, Maurer BA (1989) Macroecology: The Division of Food and Space Among Species on Continents. *Science (New York, N.Y.)*, **243**, 1145–1150.

Canada Centre for Mapping and Earth Conservation (2003) Atlas of Canada 1,000,000 national frameworks data, hydrology - drainage areas (wshed). Natural Resources Canada. http://geogratis.gc.ca/api/en/nrcan-rncan/ess-sst/ 33831182-51d3-59e9-8ebf-553da5a09388.

Currie DJ (1991) Energy and Large-Scale Patterns of Animal-Species and Plant-Species Richness. *The American Naturalist*, **137**, 27–49.

Currie DJ, Mittelbach GG, Cornell HV et al. (2004) Predictions and tests of climate-based hypotheses of broad-scale variation in taxonomic richness. *Ecology Letters*, **7**, 1121-1134.

Davies TJ, Savolainen V, Chase MW, Moat J, Barraclough TG (2004) Environmental energy and evolutionary rates in flowering plants. *Proceedings of the Royal Society B: Biological Sciences*, **271**, 2195–2200.

Field R, Hawkins BA, Cornell HV et al. (2009) Spatial species-richness gradients across scales: a meta-analysis. *Journal of Biogeography*, **36**, 132–147.

Fraser RH & Currie DJ (1996) The species-energy hypothesis in a system where historical factors are thought to prevail: coral reefs. *The American Naturalist*, **148**, 138-159.

Griffiths D (1997) Local and regional species richness in north American lacustrine fish. *Journal of Animal Ecology*, **66**, 49–56.

Griffiths D (2010) Pattern and process in the distribution of North American freshwater fish. *Biological Journal of the Linnean Society*, **100**, 46–61.

Griffiths D (2015) Connectivity and vagility determine spatial richness gradients and diversification of freshwater fish in North America and Europe. *Biological Journal of the Linnean Society*.

Griffiths D, McGonigle C, Quinn R (2014) Climate and species richness patterns of freshwater fish in North America and Europe (ed Dawson M). *Journal of Biogeography*, **41**, 452–463.

Hawkins BA, Diniz-Filho JAF, Jaramillo CA, Soeller SA (2006) Post-Eocene climate change, niche conservatism, and the latitudinal diversity gradient of New World birds. *Journal of Biogeography,* **33**, 770-780.

Hawkins BA, Field R, Cornell HV et al. (2003) Energy, Water, and Broad-Scale Geographic Patterns of Species Richness. *Ecology*, **84**, 3105–3117.

Hawkins BA, Rodríguez, Well SG (2011) Global angiosperm family richness revisited: linking ecology and evolution to climate. Jou*rnal of Biogeography,* **38**, 1253-1266.

Hijmans RJ, Cameron SE, Parra JL, Jones PG, Jarvis A (2005) Very high resolution interpolated climate surfaces for global land areas. *International Journal of Climatology*, **25**, 1965–1978.

Jablonski D, Roy K, Valentine JW (2006) Out of the tropics: evolutionary dynamics of the latitudinal diversity gradient. *Science*, **314**, 102-106.

Kerr JT, Currie DJ (1999) The relative importance of evolutionary and environmental controls on broad-scale patterns of species richness in North America. *Ecoscience*. **6**, 329-337.

Latham EL., Ricklefs RE (1993) Continental comparisons of temperate-zone tree species diversity. *Species diversity in ecological communities: historical and geographical perspectives* (ed. by R.E. Ricklefs and D. Schluter), pp. 294–314. University of Chicago Press, Chicago.

MacArthur RH, Wilson EO (1963) An equilibrium theory of insular zoogeography. *Evolution*, **17**, 373.

McDonald RI, Green P, Balk D, Fekete BM, Revenga C, Todd M, Montgomery M (2011) Urban growth, climate change, and freshwater availability. *Proceedings of the National Academy of Sciences of the United States of America*, **108**, 6312–6317.

Mittelbach, GG, Schemske, DW, Cornell, HV et al. (2007) Evolution and the latitudinal diversity gradient: speciation, extinction and biogeography. *Ecology Letters*, **10**, 315-331.

Morrow JE (1980) *The Freshwater Fishes of Alaska*. Alaska Northwest Publishing Company, Anchorage.

Mu Q, Zhao M, Running SW (2011) Improvements to a MODIS global terrestrial evapotranspiration algorithm. *Remote Sensing of Environment*, **115**, 1781-1800.

NatureServe (2010) Digital distribution maps of the freshwater fishes in the conterminous United States. Arlington, VA. http://www.natureserve.org/conservation-tools/data-maps-tools/digital-distribution-native-us-fishes-watershed.

NatureServe (2013) Lethenteron alaskense. The IUCN Red List of Threatened Species: e.T202623A18234298. http://dx.doi.org/10.2305/IUCN.UK.2013-1.RLTS.T202623A18234298.en.

Oberdorff T, Guegan JF, Hugueny B (1995) Global scale patterns of fish species richness in rivers. *Ecography*, **18**, 345–352.

Oberdorff T, Tedesco PA, Hugueny B, Leprieur F, Beauchard O, Brosse S, Dürr HH (2011) Global and Regional Patterns in Riverine Fish Species Richness: A Review. *International Journal of Ecology*, **2011**, 1–12.

Peres-Neto PR, Dray S, Braak CJFT (2016) Linking trait variation to the environment: critical issues with community-weighted mean correlation resolved by the fourth-corner approach. *Ecography*, 1–29. http://doi.org/10.1111/ecog.02302

Pontarp M, Wiens JJ (2016) The origin of species richness patterns along environmental gradients: uniting explanations based on time, diversification rate and carrying capacity. *Journal of Biogeography*, 1–14. http://doi.org/10.1111/jbi.12896R.

R Core Team (2008) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, http://www.R-project.org.

Rahbek C (2004) The role of spatial scale and the perception of large-scale species-richness patterns. *Ecology Letters*, **8**, 224–239.

Rangel TFLVB, Diniz-Filho JAF, Bini LM (2010) SAM: a comprehensive application for Spatial Analysis in Macroecology. *Ecography*, **33**, 46–50.

Rohde K (1992) Latitudinal Gradients in Species Diversity: The Search for the Primary Cause. *Oikos*, **65**, 514–527.

Ross, ST (2013). *Ecology of North American Freshwater Fishes*. University of California Press, Berkeley.

Rosseel Y (2012) lavaan: An R Package for Structural Equation Modeling. *Journal of Statistical Software*, **48**, 1–36.

Schluter D (2016) Speciation, Ecological Opportunity, and Latitude. *The American Naturalist*, **187**, 1-18.

Scott W, Crossman E (1985) *Freshwater fishes of Canada.* Minister of Supply and Services Canada, Ottawa.

Smith GR, Badgley C, Eiting TP, Larson PS (2010) Species diversity gradients in relation to geological history in North American freshwater fishes. *Evolutionary Ecology Research*, **12**, 693–726.

Watermolen J (1999) 1:2,000,000-scale Hydrologic Units of the United States. U.S. Geological Service, Reston, VA.

Webb CO, Ackerly DD, Kembel SW (2008) Phylocom: software for the analysis of phylogenetic community structure and trait evolution. *Bioinformatics*, **24**, 2098–2100.

Wiens JJ, Ackerly DD, Allen AP et al. (2010) Niche conservatism as an emerging principle in ecology and conservation biology. *Ecology Letters*, **13**, 1310–1324.

Wiens JJ, Donoghue MJ (2004) Historical biogeography, ecology and species richness. *Trends in Ecology and Evolution*, **19**, 639-644.

Wright SD, Gray RD, Gardner RC (2003) Energy and the rate of evolution: inferences from plant rDNA substitution rates in the western Pacific. *Evolution*, **57**, 2893–7.

Wright SD, Keeling J, Gillman L (2006) The road from Santa Rosalia: a faster tempo of evolution in tropical climates. *Proceedings of the National Academy of Sciences of the United States of America*, **103**, 7718–7722.

Zelený D, Schaffers AP (2012) Too good to be true; pitfalls of using mean Ellenberg indicator values in vegetation analyses. *Journal of Vegetation Science*, **23**, 419–431.

Zhao M, Heinsch FA, Nemani RR et al. (2005) Improvements of the MODIS terrestrial gross and net primary production global data set. *Remote Sensing of Environment,* **95**, 164–176.

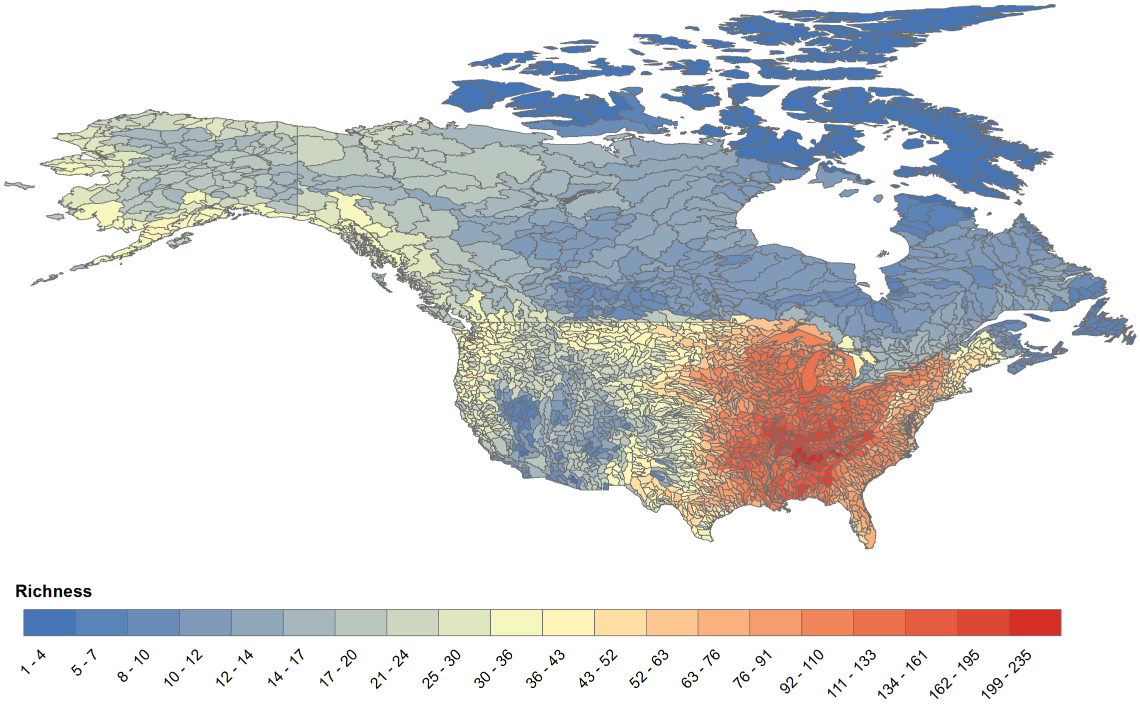

Figure 1. Richness of native North American freshwater fish by watershed.

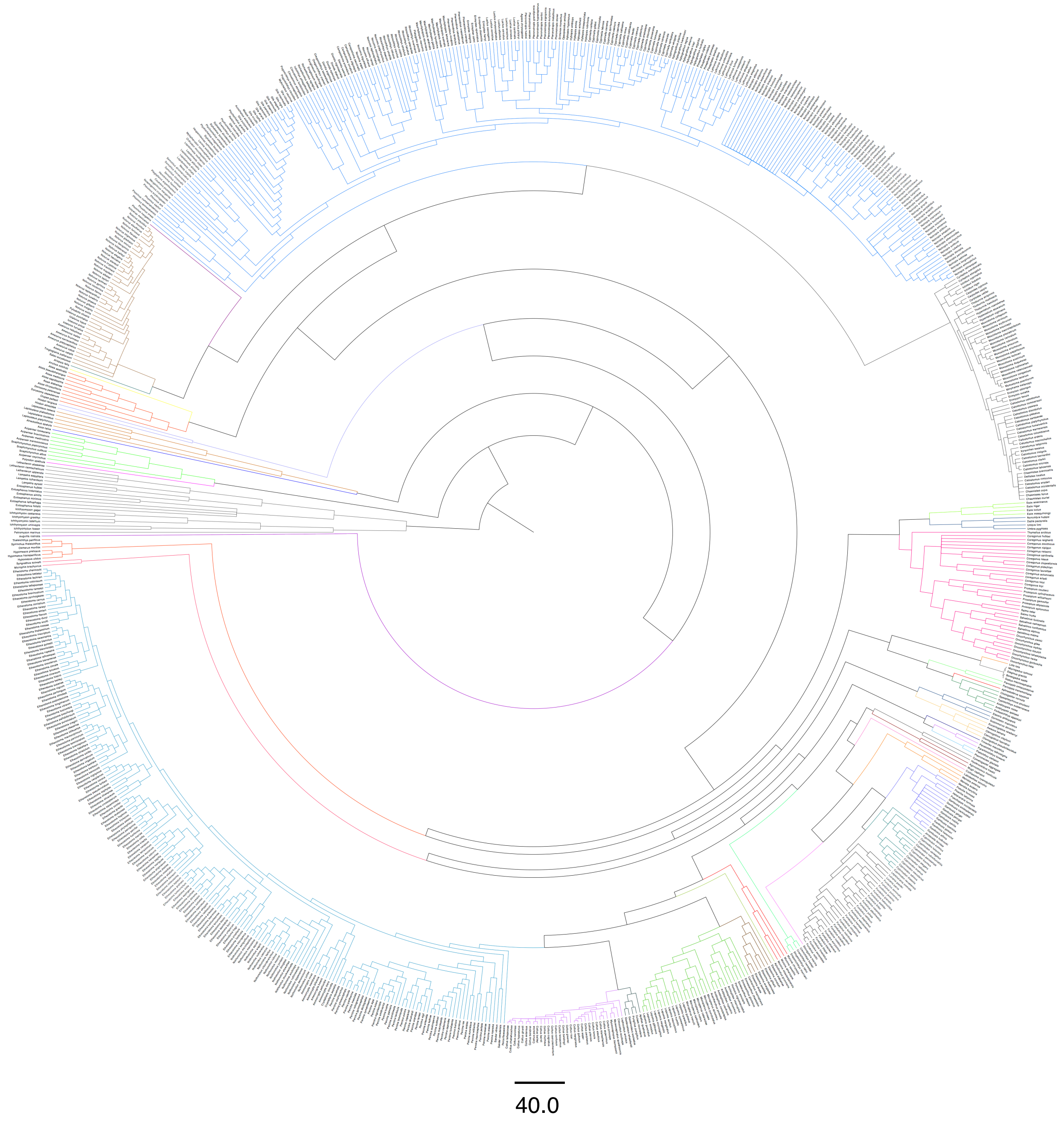

Figure 2. Phylogeny of native North American freshwater fish. Branch colors represent families. Branch lengths represent time in millions of years.

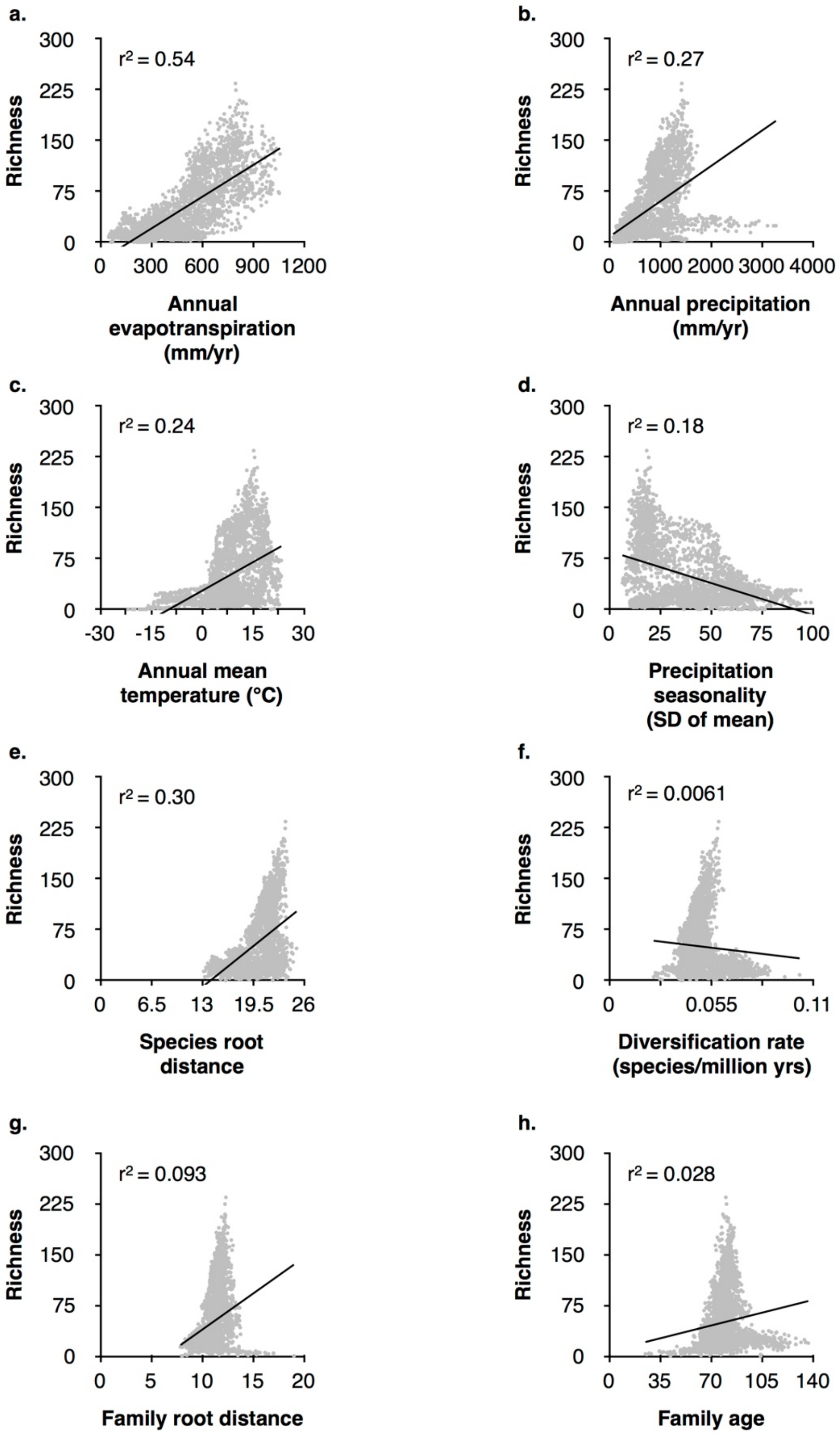

Figure 3. Richness as a function of climatic (a – d) or phylogenetic (e – h) variables across watersheds.

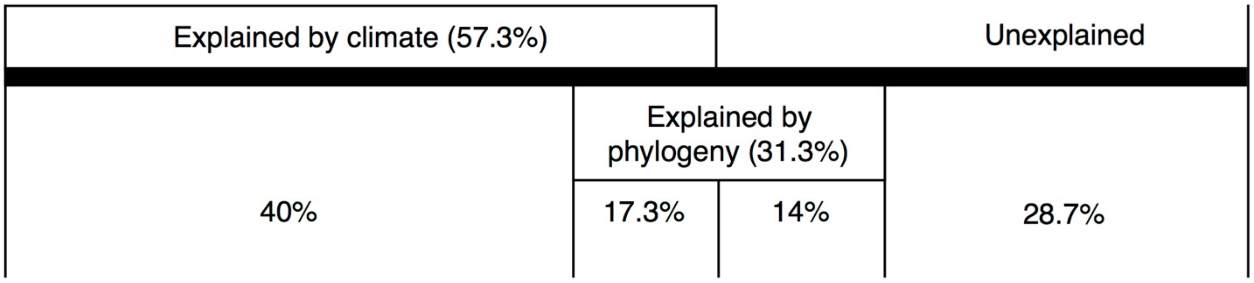

Figure 4. Partial regression representing how climate and phylogeny explain variation in species richness (thick horizontal line). The independent contribution of climate is greater (40%) than that of the independent contribution of phylogeny (14%). There is some co-linearity between climate and phylogeny (17.3%). There is unexplained variance (28.7%) that is not accounted for by the climatic and phylogenetic variables.

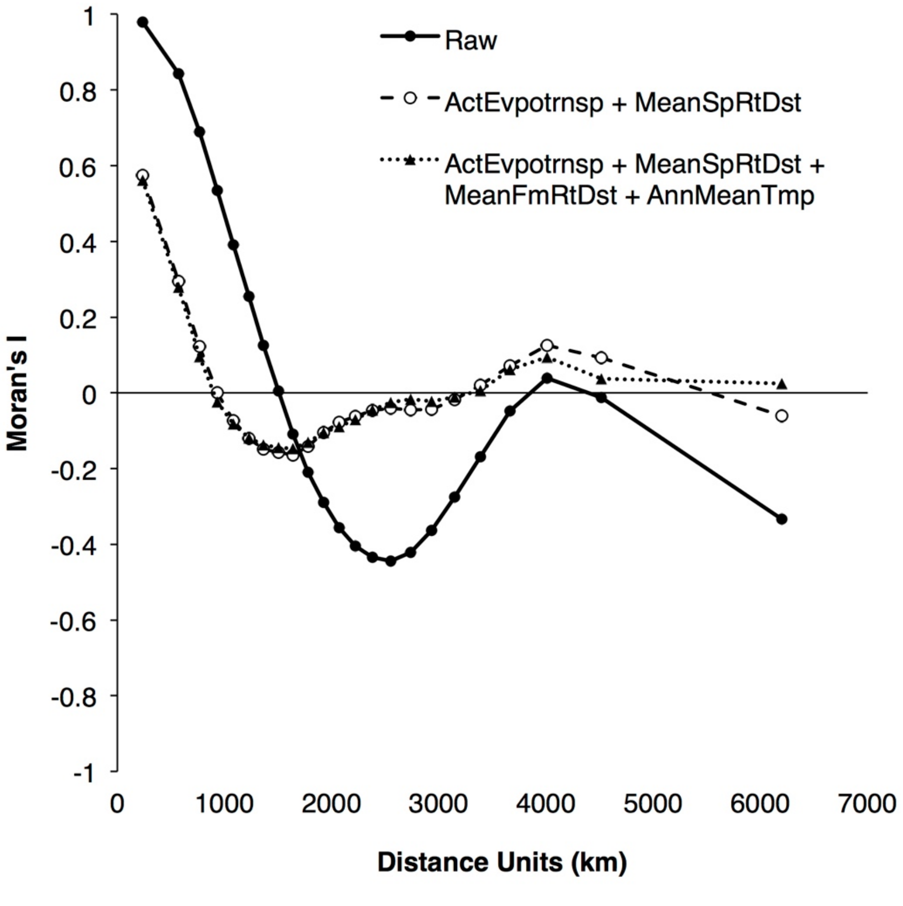

Figure 5. Spatial autocorrelogram representing spatial autocorrelation (Moran's I) of the residuals of the richness pattern (black line). After inputting mean species root distance and actual evapotranspiration, the spatial autocorrelation in the residuals was corrected (dashed line). The best fit model included the addition of mean family root distance and annual mean temperature (dotted line).

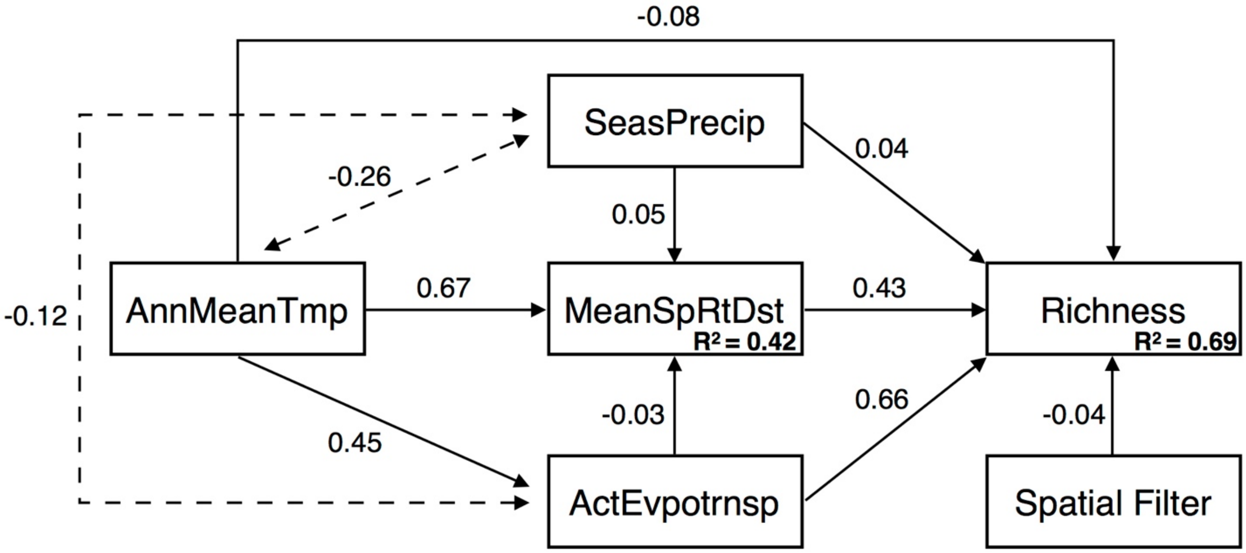

Figure 6. Path model best fitting the structure of the richness data. The numbers above the paths represent standardized coefficients, and the dashed path denotes a link hypothesized to lack directed causality.

Table S1. Correlations of all variables with each other.

	Richness	MRDS	MRDF	MFA	MDR	AET	PET	BIO1	BIO4	BIO5	BIO6	BIO12	BIO15	NPP
Richness	1													
MRDS	0.55	1												
MRDF	0.30	0.40	1											
MFA	0.17	-0.21	-0.61	1										
MDR	-0.08	0.36	0.01	-0.10	1									
AET	0.74	0.26	0.45	0.08	-0.17	1								
PET	0.15	0.66	0.09	-0.08	0.39	-0.09	1							
BIO1	0.49	0.64	0.37	0.00	0.20	0.45	0.79	1						
BIO4	-0.17	-0.27	-0.13	-0.20	-0.39	-0.30	-0.51	-0.71	1					
BIO5	0.42	0.71	0.26	-0.07	0.20	0.24	0.87	0.90	-0.42	1				
BIO6	0.40	0.51	0.29	0.13	0.29	0.44	0.69	0.94	-0.89	0.73	1			
BIO12	0.52	0.11	0.34	0.11	-0.02	0.76	-0.13	0.38	-0.46	0.07	0.47	1		
BIO15	-0.42	-0.25	-0.33	0.27	-0.14	-0.53	0.07	-0.18	0.13	-0.09	-0.18	-0.45	1	
NPP	0.46	0.07	0.35	0.14	-0.11	0.82	-0.06	0.40	-0.40	0.17	0.45	0.72	-0.35	1

Table S2. Collinearity, measured as variance inflation factors (VIF), between variables. Values in bold designate VIFs greater than 2.5.

	Richness	MRDS	MRDF	MFA	MDR	AET	PET	BIO1	BIO4	BIO5	BIO6	BIO12	BIO15	NPP
Richness	1													
MRDS	1.4	1												
MRDF	1.1	1.2	1											
MFA	1	1	1.6	1										
MDR	1	1.2	1	1	1									
AET	2.2	1.1	1.3	1	1	1								
PET	1	1.8	1	1	1.2	1	1							
BIO1	1.3	1.7	1.2	1	1	1.2	**2.6**	1						
BIO4	1	1.1	1	1	1.2	1.1	1.3	2	1					
BIO5	1.2	2	1.1	1	1	1.1	**4.2**	**5.2**	1.2	1				
BIO6	1.2	1.3	1.1	1	1.1	1.2	1.9	**8.6**	**4.8**	2.2	1			
BIO12	1.4	1	1.1	1	1	2.4	1	1.2	1.3	1	1.3	1		
BIO15	1.2	1.1	1.1	1.1	1	1.4	1	1	1	1	1	1.2	1	
NPP	1.3	1	1.1	1	1	**3**	1	1.2	1.2	1	1.3	2.1	1.1	1