

COMMUNITY PHYLOGENETICS AND ECOSYSTEM FUNCTIONING

Phylogenetic niche conservatism – common pitfalls and ways forward

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Summary

1. The prevalence of phylogenetic niche conservatism (PNC) in nature is still a conflicting issue. Disagreement arises from confusion over its precise definition and the variety of approaches to measure its prevalence. Recent work highlighted that common measures of PNC strongly depend on the assumptions of the underlying model of niche evolution. However, this warning has not been well recognized in the applied literature and questionable approaches are still frequently applied.

2. The aim of this study is to draw attention to the assumptions underlying commonly applied simple measures of PNC. We used a series of simulations to illustrate how misleading results can be if assumptions of niche evolution are violated, that the violation of assumptions is a common phenomenon and that testing assumptions requires in-depth pretest.

3. We conclude that the seemingly simple measures of PNC, such as phylogenetic signal (PS) and evolutionary rate, are not so easy to apply if one accounts for the necessity to test model assumptions. In addition, these measures can be difficult to interpret. The common assumption that strong PS indicates PNC will be often invalid. In addition, the interpretation of some measures, for example the conclusion that evolutionary rate is slow enough to indicate PNC, requires a comparison with another clade, another trait or well-developed null model assumptions and thus additional data.

4. We suggest that studies investigating PNC should always compare alternative evolutionary models and that model comparisons should in particular include flexible niche evolution models such as multiple-optima OU models, although these are computational intensive. These models are directly inherited from the concept of macroevolutionary adaptive landscape and can indicate PNC either by relative few peak shifts or by narrow peaks in the adaptive landscape. A test of PNC thus requires comparing these parameters of the macroevolutionary landscape between clades or time periods.

5. The general prevalence of PNC in nature should be evaluated only based on studies keeping up to the high standards of communicating the used definition of PNC, testing the assumptions made in the modelling approaches and including newly developed models in a model comparison approach.

Key-words: Brownian motion, macroevolutionary model, neutral drift, niche lability, Ornstein–Uhlenbeck, rates of evolution

Introduction

The interplay between ecological and evolutionary processes drives the distribution and dynamics of biodiversity (Ricklefs 1987; Lavergne *et al.* 2010). A signature of these

processes is contained in phylogenetic trees and in the distribution of species' ecological characteristics along these trees (Webb *et al.* 2002). One of the most striking and long recognized signatures is the similarity of ecological niches among closely related species (Wiens and Graham 2005; Losos 2008a; Pearman *et al.* 2008). The degree to which observable species characteristics contain information

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about phylogenetic relationships can be broadly defined as 'phylogenetic signal' (PS, e.g. Hillis & Huelsenbeck 1992), but this definition may encompass several different metrics. A commonly used and operational perspective, which we will use in the following, is to define PS as the tendency for related species to resemble each other more than they resemble species drawn at random from the phylogenetic tree (Blomberg & Garland 2002).

Already Darwin (1859) identified the relatively recent common ancestry of related species as the primary reason for their ecological similarity (the pattern that much later has been termed PS). Grafen (1989) was one of the first to suggest that niche similarity of related species is not only due to shared ancestry but also due to shared environmental constraints and biogeographic history. He hypothesized that closely related species tend to have similar niches and to live more or less closely together. They should thus have high chances to enter the same new niches when they emerge. Over time, they should then be exposed to similar adaptive forces in these new niches, which should lead them to be more similar ecologically than expected under neutral drift (Grafen 1989). Grafen's explanation was then revisited by Harvey & Pagel (1991), who defined the ecological and evolutionary processes resulting in the tendency of species or lineages to retain their ancestral niches over time as phylogenetic niche conservatism (PNC). Recent theoretical work has shown that PS and PNC are not the same thing. Even under strong PNC, species niche differences can be unrelated to phylogenetic distances (no PS) and labile niches can sometimes lead to a strong relation between niche differences and phylogenetic distance (strong PS, Revell, Harmon & Collar 2008).

The recent rise of large and dated molecular phylogenies (Bininda-Emonds *et al.* 2008; Thuiller *et al.* 2011; Jetz *et al.* 2012; Pyron, Burbrink & Wiens 2013; Zanne *et al.* 2014) allows for testing multiple hypotheses related to PNC and past biodiversity dynamics and has led to an increased interest in PNC (Pearman *et al.* 2014). However, the concept was expanded and conflicting ideas emerged around two key questions: Firstly, is PNC a process or a pattern? Harvey & Pagel (1991, p. 38) introduced PNC as an adaptive ecological and evolutionary process. This initial interpretation may have been based on a population dynamic perspective where different factors, such as strong stabilizing selection, lack of appropriate genetic variation, genetic or functional constraints due to epistatic or pleiotropic effects, and gene flow impeding the emergence of novel adaptations, can lead to PNC (Hansen & Houle 2004; Walsh & Blows 2009; Lavergne *et al.* 2010). From this viewpoint, PNC always emerges from an impossibility of niche novelties to emerge due to conditions that restrict population growth (Holt 1996; Gomulkiewicz & Houle 2009). In contrast to this process perspective, other authors have suggested that PNC can be either a process or a pattern depending on the perspective from which the research question is asked (Wiens 2008; Wiens *et al.* 2010). Finally, it has been claimed that a pure pattern perspective would

be adequate as it allows for directly measuring PNC instead of just assuming its presence (Losos 2008). For Losos, PNC in itself cannot explain anything but can result from several processes (genetic constraints, stabilizing selection). Thus, a hypothesis about one given underlying process can never be confirmed with a pure phylogenetic approach. However, comparative approaches may be useful to reject certain classes of processes (Losos 2011).

Secondly, which constraints are needed to produce PNC? Some authors have suggested that kinship between species relations and resulting niche similarity between related species are not only essential but also sufficient, that is, that neutral drift is enough to drive PNC (Wiens 2008; Cooper, Jetz & Freckleton 2010; Wiens *et al.* 2010). Other authors have postulated that PNC requires additional constraints such as environmental filtering which induces pervasive stabilizing selection (Harvey & Pagel 1991; Desdés *et al.* 2003; Losos 2008) or limits to adaptive evolution (Holt 1996; Gomulkiewicz & Houle 2009). This is what motivated Losos (2008) to propose that the concept of PNC should be restricted to situations where conservative forces (among the ones listed above) are exerted on phylogenetically related taxa, ultimately resulting in niches diversifying so slowly that closely related species should resemble each other more than expected under neutral drift.

These conflicting ideas about what is and what is not PNC have resulted in a strong debate on its prevalence in nature. While some studies have reported support for PNC (Ackerly 2004; Gomez, Verdu & Perfectti 2010; Burns & Strauss 2011), others have provided counterexamples (Evans *et al.* 2009; Boucher *et al.* 2012; Pearman *et al.* 2014) and some studies have demonstrated its dependency on studied systems, time-scales and niche-related traits (Peterson, Soberon & Sanchez-Cordero 1999; Freckleton & Jetz 2009; Cooper, Freckleton & Jetz 2011). However, as the different studies define PNC differently and accordingly utilize different measures, they are not directly comparable (Revell, Harmon & Collar 2008; Ackerly 2009) and inevitably lead to inconsistent conclusions about the prevalence of niche conservatism (Losos 2008). Recent conceptual reviews have aimed at disentangling the different viewpoints on PNC and have highlighted that the performance of different measures strongly depends on the assumptions of a certain model of niche evolution (e.g. Cooper, Jetz & Freckleton 2010; Crisp & Cook 2012). However, in applied studies, PNC is still measured with one or the other approach without adequately testing for the underlying evolutionary process (e.g., Buckley *et al.* 2010). Also, questionable approaches are still frequently used (e.g. methods relying only on taxonomic topologies, Peterson, Soberon & Sanchez-Cordero 1999; Prinzing *et al.* 2001) and general conclusions on the prevalence of PNC in nature rely on these biased studies (Wiens *et al.* 2010). In fact, despite the fact that the term 'PNC' was almost unknown at the turn of last century, its use has expanded in recent literature and

nobody seems to really agree anymore on what it really is (Crisp & Cook 2012).

In the following, our study revisits earlier work on the different approaches for measuring PNC and on the assumptions underlying these approaches (Revell, Harmon & Collar 2008; Ackerly 2009; Cooper, Jetz & Freckleton 2010; Wiens *et al.* 2010; Crisp & Cook 2012). We go beyond this earlier work by incorporating recent advances in modelling PNC with flexible evolutionary models with no *a priori* about where niche shifts occur in the phylogeny. We further illustrate with simulated data that violations of basic assumptions of niche evolution models frequently lead to erroneous conclusions and highlight the challenges to test these assumptions. Finally, we suggest ways forward in the study of PNC hopefully contributing to a better common practice and thus to better comparable studies on the prevalence of PNC in nature.

Limitations of commonly applied measures of PNC

The niche of a species is usually defined as the set of abiotic and biotic conditions in which species are able to persist and maintain stable populations (Hutchinson 1957; Wiens and Graham 2005). For the sake of simplicity, we will assume in the following that species' niches can be described by a single continuous trait capturing the adaptation to those environmental conditions in which a species can thrive. We acknowledge that this is an oversimplification of reality, but this choice is driven by the availability of sophisticated models for the evolution of continuous traits on phylogenies (see below). Furthermore, extending this framework to include intraspecific variability and measurement error or to consider simultaneously multiple traits describing the niche is theoretically possible (although not implemented in all modelling approaches).

It is well-established that the performance of different tests of PNC depends on assumptions of underlying niche evolution models (Cooper, Jetz & Freckleton 2010; Crisp & Cook 2012). Nevertheless, PNC approaches are often applied without knowing which model of niche evolution best describes the data. Two arguments could support this practice. First, PNC approaches could be robust to moderate violations of model assumptions. Secondly, the often-assumed simple models, such as Brownian motion (see below), could well represent the vast majority of studied cases of niche evolution. In the following, we complete recent conceptual reviews with a series of simulations to illustrate that both of these arguments fail in practice. In our simulations, we model the evolution of a single niche trait. In a first step, phylogenetic trees are simulated under a Yule process. Then, niche evolution along these phylogenies is simulated through a discretized continuous time process following various niche evolution models (see below and Appendix S1 in the Supporting information for more details.). Finally, different measures of PNC are applied to the simulated data and results are compared.

MODELS OF NICHE EVOLUTION: THE BASICS

The models of niche evolution introduced here describe the evolution of quantitative characters on phylogenies. Stochasticity, branch lengths and internal or external constraints can influence niche evolution depending on the underlying model. Different niche evolution models can result in very different patterns of niche trait values among the tips of the phylogenies (i.e. species, see Fig. 1).

The Brownian motion

Under Brownian motion (BM, Felsenstein 1985), a continuous trait evolves according to a suite of random steps (i.e. random walk process), with no preferred direction. BM is thus typically used to represent neutral drift over macroevolutionary time-scales, but BM-like evolution can arise by a number of other processes such as selection in a quickly fluctuating environment (Hansen & Martins 1996). The size of the random steps depends only on evolutionary rate, that is, on the rate of phenotypic change in a given lineage.

The Ornstein–Uhlenbeck model

For the purpose of modelling evolution towards a selective optimum, the Ornstein–Uhlenbeck model was introduced to comparative analyses (OU, Hansen 1997; Butler & King 2004). This model is an extension of BM where an additional term describes attraction towards an optimal niche value. Even though its formulation is identical to Lande's model of stabilizing selection within a population (Lande 1976), the macroevolutionary OU model is only phenomenological, as it mimics (but does not demonstrate) stabilizing selection, constrained evolution in bounded phenotypic space (Boucher *et al.* 2014) or evolution towards shifting niche optima (Hansen 2012). Under an OU model, the evolution of the niche value (x) is governed by the following differential equation:

$$x(t + dt) = x(t) + \alpha(\theta(t) - x(t))dt + \sigma dte.$$

In this equation, $\varepsilon \sim N(0,1)$, σ is the Brownian motion rate, θ the optimal value (that can depend on time), α the selection strength, and dt is an elementary unit of time. BM is a special case of the OU model with no selection ($\alpha = 0$). When niche evolution has been so fast that all traces of shared ancestry are lost, niches are said to follow a white noise model (WN), which is equivalent to random draw of niche values independently of the phylogeny. WN is a special case of the OU model where the selection strength tends to infinity (see Appendix S1 for more information).

Further extensions

Multiple variations of the BM and the OU models have been proposed: BM with rates that vary over time like

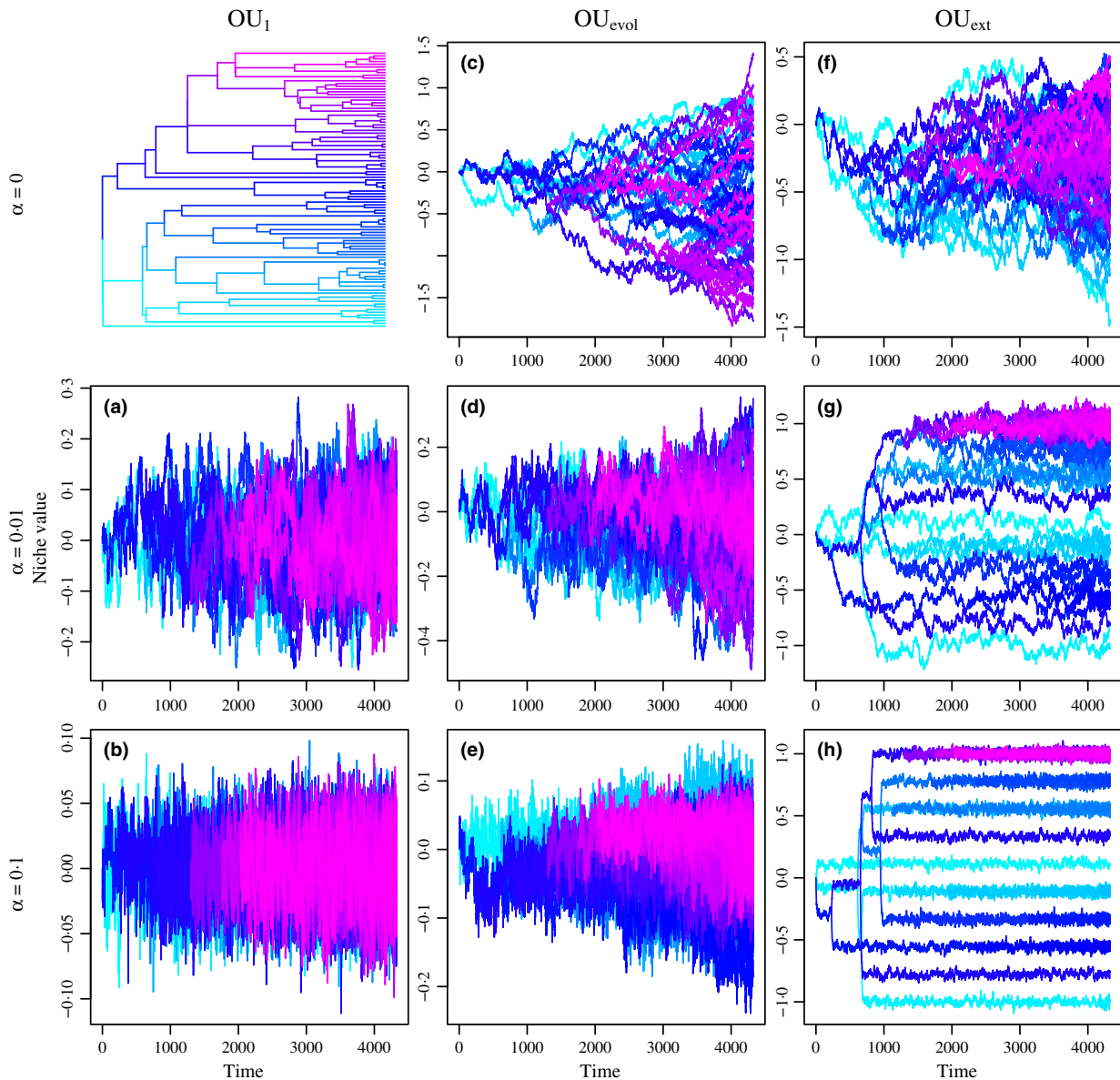


Fig. 1. Niche evolution over time for an OU_1 (left column), OU_{evol} (middle column) and OU_{ext} (right column) model with different strength of selection (α). The upper left plot shows the phylogeny; the middle and the upper right plot show two realizations of Brownian motion (BM). Colours indicate the position of branches in the phylogeny. When considering only the final distribution of niche values in the tips, then a OU_1 model with strong α can be easily misidentified as a white noise model and an OU_{ext} model with moderate α can be easily misidentified as a BM model.

the ACDC model (Blomberg, Garland & Ives 2003) that becomes the Early Burst model when there is slowdown over time (Harmon *et al.* 2010) or vary in different parts of the phylogeny (O’Meara *et al.* 2006; Thomas, Freckleton & Szekely 2006; Eastman *et al.* 2011), OU with different selective optima applied to different parts of the phylogeny (Butler & King 2004), and even OU models with different selective optima, where rate of drift and selection strength can also vary around different niche optima (Beaulieu *et al.* 2012). Taking into account heterogeneity in the process of niche evolution is important since simple homogeneous models frequently happen to be very poor descriptors of the data (Pennell *et al.* 2014).

Furthermore, this allows for testing for more complex scenarios and hypotheses of niche evolution. It is, however, important to note that different niche evolution models can cause either completely different or the same niche distribution patterns. For example, neutral drift and adaptation to quickly changing environments can lead to the same BM-like patterns even though underlying processes are very different. Therefore, sometimes, in the analysis of phylogenetic patterns of species niches, several models seem equally plausible in terms of statistical explanatory power, but they may not all be as biologically plausible. Choosing from the seemingly plausible models one that describes a biologically unrealistic

process obviously bears the strong risk of drawing wrong conclusions about PNC.

ARE COMMON MEASURES INFORMATIVE?

Phylogenetic signal, evolutionary rate, the selection strength in an OU model and model comparison analyses are commonly used to study PNC (Cooper, Jetz & Freckleton 2010; Wiens *et al.* 2010; Crisp & Cook 2012). The conflicting ideas about PNC (Is it a process or a pattern? Which constraints are needed to produce PNC?) resulted in a debate on which of these approaches are most appropriate. The different measures and the underlying philosophy are shortly outlined in the following together with their limitations (e.g. Revell, Harmon & Collar 2008; Ackerly 2009; Hunt 2012; Ho & Ane 2013).

Phylogenetic signal

Phylogenetic signal takes a pure pattern perspective. Different authors suggested different values of PS to be sufficient for PNC: while Wiens (2008) and Wiens *et al.* (2010) state that a pattern expected under neutral drift alone is sufficient and thus state that PNC can be produced by a random walk without additional constraints, others demand that there should be additional constraints (e.g. a strong stabilizing selection favouring the ancestral niche over time) leading to a PS that is stronger than expected under random walk (Losos 2008).

A strong limitation of this approach is that PS can be used to measure PNC unambiguously only under the assumption of a BM-like evolutionary process (Revell, Harmon & Collar 2008; Cooper, Jetz & Freckleton 2010; Fig. 1c,f). The analysis of our simulated data demonstrates how violations of the BM assumption can produce strongly misleading patterns of PS (Fig. 2). For instance, when niches evolve according to an OU process with a single optimum (Fig. 1a,b), strong constraints (leading to niches with only very little variation over time and thus to strong PNC in our simulations) result in low PS. Conversely, when niches evolve under an OU model with multiple optima in different parts of the phylogeny (Fig. 1g,h, which could be interpreted as labile niches, at least in the early history of the clade, see Appendix S1) PS is higher than expected under BM. In addition to the consequences of violating models' assumptions, there is also the problem that several indices are available to measure PS and since they capture different aspects of the concept, they can give significantly different answers (e.g. Blomberg's K , Pagel's λ , Moran's I and Abouheif's C_{mean} , see Münkemüller *et al.* 2012 for a comparison).

In summary, PS is only a useful measure of PNC if the underlying process of evolution is BM-like. Otherwise, it can be completely unrelated to PNC and even give spurious answers with respect to the underlying process. Showing evidence of PNC based on PS thus requires to test whether BM is the underlying model of evolution and (if

this is fulfilled) to accept a definition of PNC that basically equals BM. To us, this is problematic since the expression PNC itself suggests that some conservative forces should slow down niche evolution, whereas BM alone is not a conservative force at all.

Evolutionary rate and related measures

Evolutionary rate measures the rate of phenotypic changes over evolutionary time in a given lineage. Since PNC implies a certain resistance to change, it should be characterized by relatively slow rates of niche evolution (Ackerly 2009). However, using this measure requires a comparison, for example, between clades or between different periods of time, in order to identify which rates can be termed relatively 'high' vs. 'low'. To allow for such a comparison, Pagel's δ (Pagel 1999) has been suggested to measure niche retention, that is, whether most niche variance has accumulated early rather than late in the history of a clade (see also O' Meara *et al.* 2006; Thomas, Freckleton & Szekely 2006 for a method to test for different rates of evolution between clades). Pagel's δ is a scaling parameter that affects branch lengths according to their depth in the phylogenetic tree (Pagel 1999). Under the assumption that niche evolution along a δ -transformed phylogeny follows BM, values below unity indicate PNC in the more recent history in comparison with more labile niches in the past.

We would like to note that, in order to standardize the measure of evolutionary rate for comparison of different traits and/or different clades, trait values should be ln-transformed (Ackerly 2009). For example, if all species in the focal clade are small and all species in a related clade are large, and we are interested in relative changes in body size, then a log-transformation of body sizes would be appropriate to calculate evolutionary rates (Harmon *et al.* 2010).

Comparably to PS, the measure of evolutionary rate depends on the assumption of an underlying niche evolution model and can only meaningfully be interpreted as a parameter of this model (Hunt 2012). Often these models assume that evolutionary rate is constant across the entire phylogeny, which is a strong and (in most cases) unrealistic assumption and can lead to highly misleading results. For example, when evolutionary rate is measured via the variance of phylogenetic independent contrasts, then it is assumed that the underlying process of niche evolution can be meaningfully approximated by a BM-like process (Felsenstein 1985; Hansen & Martins 1996). The analysis of our simulated data illustrates how misleading estimates of evolutionary rates can be if these assumptions are violated. Indeed, an evolutionary rate inferred as the variance of phylogenetic independent contrasts is wrongly estimated if data are simulated with an Ornstein–Uhlenbeck (OU) model (Fig. 3a). This is because the OU model has a finite stationary variance ($\sigma^2/2\alpha$) that depends on α (see Appendix S1 and Fig. 1 for more information). The variance of niches under an OU process thus remains constant when

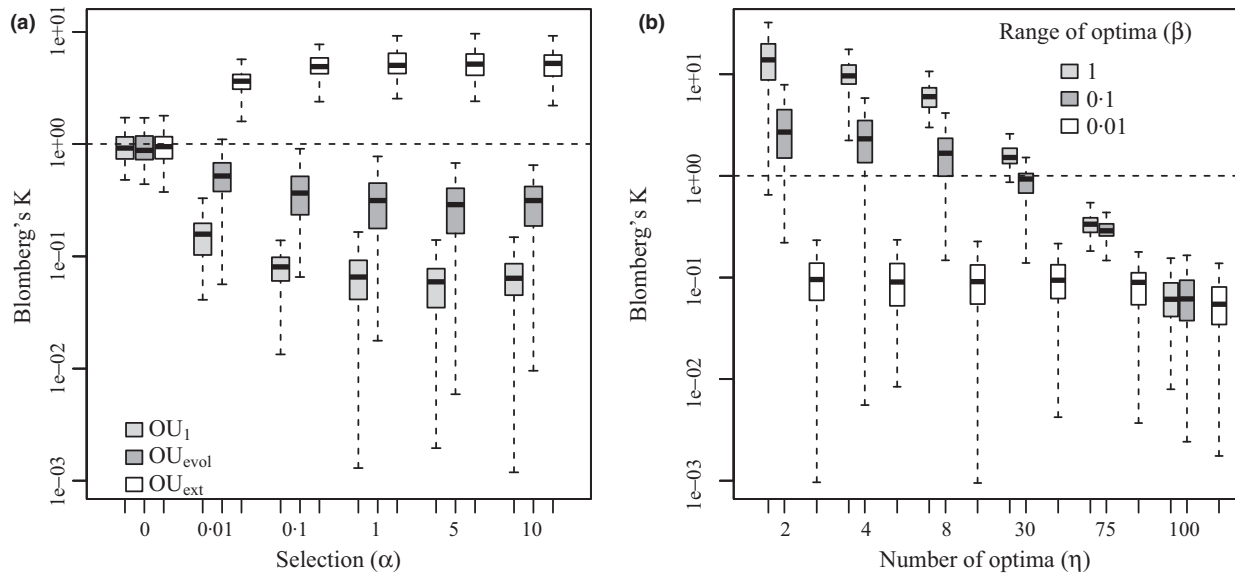


Fig. 2. We measured phylogenetic signal (PS based on Blomberg's K) in virtual data that we simulated with different niche evolution models. In the simulations, niche values evolved along phylogenetic trees under three different niche evolution models: OU_1 is a single-optimum OU model where we set the optimum value equal to the root value; OU_{evol} is a OU model where we set the optima of nodes to the niche values of the respective ancestor nodes; OU_{ext} is a OU model where η different clades differ strongly in their external selective optima (β indicates the range of different optima) and these optima also differ strongly from the root value (see Appendix S1 and S2 for more details). We found that PS decreased with parameter changes assumed to increase phylogenetic niche conservatism, such as high values of selection strength (α) in the OU_1 and OU_{evol} model. Phylogenetic signal, however, increased with parameter changes assumed to increase niche lability in the OU_{ext} model, such as moderate to high values of selection strength (α , panel a), high numbers of distinct selective optima (η , panel b) and increased ranges of selective optima niche values (high β , panel b). In all simulations, the variance of Brownian motion was set to $\sigma = 0.01$. In plot a, the three models are parameterized as follows: $\eta = 1$ for OU_1 , $\eta = 10$ and $\beta = 1$ for OU_{ext} and for OU_{evol} ; the optima are equal to the ancestor nodes; in plot b, $\alpha = 1$. Each boxplot represents 100 repetitions. Note that y-axes are logarithmic and that outliers were not plotted.

the age of the clade increases (see fig. 3 in Harmon *et al.* 2010), leading to a decrease in the overall evolutionary rate estimated on the phylogeny (Fig. 3a). Accordingly, Pagel's δ indicates that most variance accumulated close to the tips of the phylogeny (Fig. 3b). This is statistically true but should not be interpreted as evidence that niche evolution has accelerated recently, because in the simulations, the OU process has remained constant through time.

In summary, evolutionary rates are a useful means of comparing the relative speed of niche evolution (and thus the strength of PNC) for different traits or different clades but cannot provide an absolute answer on the presence vs. absence of PNC. They provide an accurate description of the process only if the right evolutionary model (BM if estimated via the variance of phylogenetic independent contrasts) is assumed. Models with variable BM rates could be useful to compare rates of niche evolution in different clades (O' Meara *et al.* 2006).

Parameters of a single-optimum OU model

The single-optimum OU model mimics constrained evolution around an optimal value (cf. Fig 1a,b) and its parameterization can be used to estimate PNC (Hansen 2012). Interpreting the estimated value of the α parameter in an OU model is appealing because it should in the-

ory be related to the strength of constraints on niche evolution.

However, the use of the α estimate (the selection strength) to inform on PNC is limited because it tends to have a bias towards high values. For instance, the analysis of data simulated with a single-optimum OU model demonstrates that it is difficult to accurately estimate the simulated α parameter (Fig. 4a, cf. in addition appendix from Butler & King 2004; Ho & Ane 2013), while the stationary variance of the OU process ($\sigma^2/2\alpha$) can be much more reliably estimated (Fig. 4b). It seems that this happens only when α is rather large compared to σ (as is the case in our simulations). However, α can be reliably estimated when it is small, and in this case, the stationary variance is wrongly estimated (C. Ané, pers. comm.). Since the stationary variance of the OU process quantifies the relative strength of drift vs. selection, we believe it is the most important quantity to report and interpret in empirical studies. In addition, rather than directly reporting values of α , one should report the phylogenetic half-life [i.e. $\ln(2)/\alpha$], which measures the time necessary for half of the information contained in phylogenetic relationships to be erased. This quantity is expected to decrease with the strength of PNC, as phylogenetic information contained in the covariance structure of the tree will be erased when selective constraints are too large.

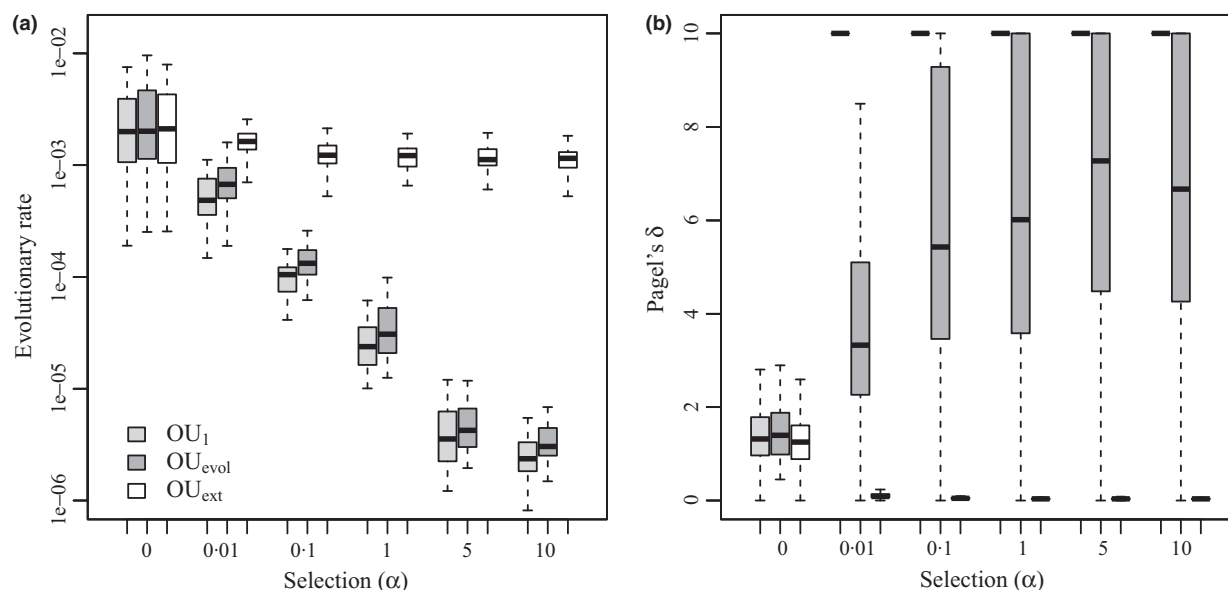


Fig. 3. We measured evolutionary rates (panel a) and Pagel's δ (panel b) in virtual data that we simulated with different niche evolution models (see the legend of Fig 2 and Appendix S1 and S2 for more details). Evolutionary rates decreased for parameter changes commonly assumed to increase phylogenetic niche conservatism (PNC; high values of selection strength, α , in the OU_1 and OU_{evol} model) and remained constant for parameter changes that increase the lability of niches (moderate to high values of selection strength in the OU_{ext} model; panel a). The retention parameter Pagel's δ , in contrast, increased for parameter changes commonly assumed to increase PNC and thus suggested more labile niche evolution towards the tips of the phylogeny. For parameter changes that increase the lability of niches, Pagel's δ decreased, which should indicate PNC according to common expectation (b). In addition, Pagel's δ had strong convergence problems. Note that outliers were not printed and that the y-axis in plot (a) is logarithmic.

We should mention that there is another limitation that relates equally to α and the stationary variance. Before interpreting the estimated parameters of an OU model, a model comparison test should be performed to decide whether a WN model, a BM model or an OU model best fits the data. Unfortunately, simulations showed that this test has very low power to detect a single-optimum OU model, at least if one follows common practice and includes a WN model in the model comparison (see Fig. 5 and next subsection). In sum, if the parameters of a single-optimum OU model are used to infer on PNC, then the stationary variance and eventually the phylogenetic half-life should be used rather than the selection strength. The underlying assumption is a single-optimum OU model, which in fact might be rarely a good expectation for an entire clade.

Model comparison

It has been suggested that a model comparison of WN, BM and OU models can be used to inform on PNC (Cooper, Jetz & Freckleton 2010). The expectation is that an OU model with a single optimum and high selection strength should be chosen when niches have been conserved throughout the evolutionary history of the clade under study. Very recent methodological developments allow to extend this model comparison approach towards multiple-optima OU models without any *a priori* on the

nodes where shifts between selective regimes would occur (see the recent SURFACE algorithm by Ingram & Mahler 2013; see Ingram & Kai 2014 for a recent application), to compare different models using Akaike Information Criterion (Akaike 1974) and to test whether more complex models are chosen solely because they have more free parameters or because they better describe the process based on a power analysis (phylogenetic Monte Carlo approach, Boettiger, Ralph & Coop 2011). These developments build on earlier work testing for *a priori* defined selective regimes on a phylogeny (Hansen 1997; see the R package ouch, Butler & King 2004; Beaulieu *et al.* 2012), constraining the adaptation of a trait to an optimum influenced by continuous, randomly changing predictor variables (see slouch algorithm, Hansen, Pienaar & Orzack 2008) and locating evolutionary shifts on a phylogeny using data-driven algorithms (Thomas & Freckleton 2012).

A major limitation of the frequently applied model comparison between WN, BM and OU models is that, first, a single-optimum OU process is often misidentified as WN and, secondly, a multiple-optima OU process is often misidentified as BM (see Fig. 5 for a demonstration with simulated data). The first misidentification is problematic since WN is often interpreted as a model of lability, while OU with one optimum is supposed to identify PNC (see Fig. 1 for the cause of this misidentification). The second limitation is problematic as BM is an assumption of many simple measures of PNC that can give biased

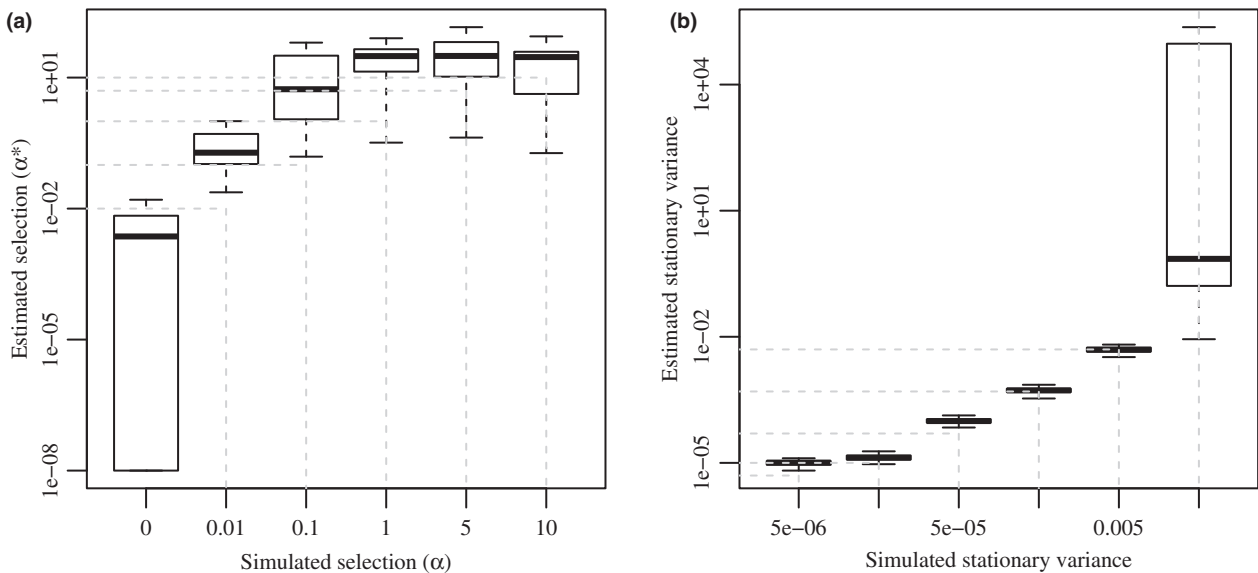


Fig. 4. We compared estimated parameters (backwards estimation from virtual data) against simulated parameters in a single-optimum OU model [OU₁ model, see Appendix S1 and S2 for more details]. Results show strong overestimations of the strength of stabilizing selection (α^*), especially if simulated selection strengths were low (a). In contrast, the estimates of the stationary variance were more accurate, at least if selection strength was not too high in comparison with the variance of the Brownian motion subprocess in the simulations (b). Each boxplot represents 100 repetitions. Note that outliers were not printed and that the y-axes are logarithmic.

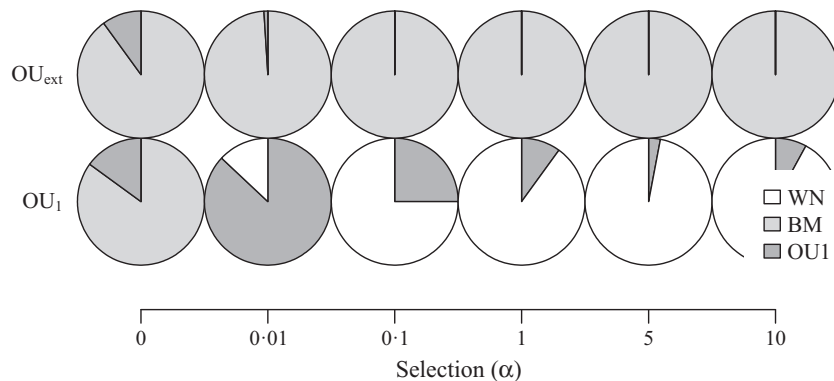


Fig. 5. Results of a model comparison analysis as is frequently done in the literature including white noise (WN), Brownian motion (BM) and single-optimum OU models (OU₁) applied to virtual data simulated with two different niche evolution models (see Appendix S1 and S2 for more details). The data for this analysis were simulated with varying strength of stabilizing selection (α) and under two different OU models (OU_{ext}, which simulates multiple external constraints, and OU₁, which simulates a single external constraint). A perfectly performing model comparison should identify BM for all simulations where $\alpha = 0$ and OU₁ for data simulated under OU₁ with $\alpha > 0$. Note that we did not include more complex OU model fitting in the model comparison, as we wanted to show results of common practice. Pie charts indicate how often which model was chosen in the 100 repetitions. The most striking results are that a single-optimum OU process is in the great majority of cases misinterpreted as white noise and that a multiple-optima OU process (OU_{ext}) is even more often misinterpreted as BM. However, note that if a more complex model with several optima was fitted, it would almost always outperform BM (not shown results).

results under a multiple-optima OU process. It is caused by the fact that a scenario where optima are too different from each other can reasonably resemble BM (Uyeda *et al.* 2011). More generally, it can as well be that simple models of flat or single-peak adaptive landscapes may simply be oversimplistic (Hansen 2012). This problem can be avoided by including multiple-optima OU models in the model comparison analysis. If they are included, then more complex models with several optima almost always

outperform more simple model choices (see following section: *Are common assumptions frequently fulfilled?*). To avoid that the choice of the more complex models is due to overfitting, a Monte Carlo-based power analyses should be applied to verify the model choice (Boettiger, Ralph & Coop 2011). Besides the risk of overfitting, the inclusion of more complex models in the model comparison makes the analysis more data demanding. Sometimes, it may also simply not be possible to fit the parameters of a multi-peak

OU model due to correlations between parameters and resulting ridges in the likelihood landscape (Beaulieu *et al.* 2012). In addition, interpretation is more difficult as it is not anymore based on testing a single value against null expectations but on the distribution of peaks and steepness in the estimated macroevolutionary adaptive landscape (Ingram & Mahler 2013; Mahler *et al.* 2013, see the following for more detail).

In summary, model comparison approaches should always consider the inclusion of more complex OU models since otherwise more complex niche evolution processes are easily confounded with BM. However, there may be reasons for discarding these models based on expert knowledge, for example, when evolution is known to have taken place more gradually than assumed in the multi-peak OU model (Pennell, Harmon & Uyeda 2014). We also suggest changing the common practice and excluding the WN model from model comparison. The reason is that the WN model is misleading, as it has no clear biological interpretation while it produces the same pattern as an OU model with strong attraction strength. Instead, although we stress out again that the OU model is only phenomenological, finding support for an OU model with strong attraction strength at least provides process-based hypotheses for why phylogenetic relationships do not correlate with niche differences in a given group of organisms: the niche might be evolving under strong stabilizing selection (Hansen 2012) or under diversifying selection within certain bounds (Boucher *et al.* 2014). Finally, we would like to highlight the potentially substantial impact of biological variation and measurement error on model inference and model comparisons. Whenever possible, appropriate methods for incorporating measurement errors should be applied (Hansen & Bartoszek 2012).

Are common assumptions frequently fulfilled?

The most important take-home message from revisiting the measures of PNC is that their performance crucially depends on whether or not the underlying assumptions of a particular model of niche evolution are fulfilled. Commonly, the critical assumption is a BM model of niche evolution. However, evidence from the literature suggests that BM is rarely the best model to describe the niche evolution process (e.g. Estes & Arnold 2007; Harmon *et al.* 2010; Uyeda *et al.* 2011). We support this evidence based on the analysis of body mass evolution in 17 clades of mammals (see Appendix S3.). For these mammal data, simple BM is never the best description of the underlying niche evolution process (see Table S1, Supporting information). This result is supported by a Monte Carlo power analysis (Table S1; with one exception: for the *Tamias* clade, the null hypothesis of an underlying BM model could not be rejected, P -value = 0.06). Furthermore, our results suggest that an OU model with a single optimum might often not be appropriate either, since in all clades except *Tamias*, at least two different optima

were detected. Finally, naively applying the different approaches of PNC to the mammal data once again demonstrates that results are often conflicting when assumptions of the approaches are not fulfilled (see Fig. S1, Supporting information).

Cautionary notes for future work on PNC

The debate on whether PNC should be defined more strictly or more loosely in concert with the limitations of approaches towards measuring PNC has diluted the concept of PNC and has rendered impossible any conclusion about how frequently PNC can be observed in nature. It seems a considerable challenge to find a single convincing answer to the question ‘how can I test whether there is PNC in my clade of interest?’ Here, we provide two answers to this question. The first one is simple but in our eyes almost useless – or little informative. The second one is informative but may be perceived as unsatisfying as it is more complex and requires rethinking the question.

SIMPLE IS NOT THAT SIMPLE AND PARTLY UNINFORMATIVE

The first answer aims at disentangling the conditions under which simple measures can be applied. Simple measures, such as PS, evolutionary rate, the tempo of niche evolution (Pagel’s δ parameter) and the selection strength in an OU model, remain frequently used for measuring PNC in empirical studies. They seem appealing methods to study PNC as they are often thought to allow for a straightforward conclusion on PNC and can be easily applied. However, to acknowledge the limitations outlined in the previous section, one would need to follow a much less simple workflow: 1. test assumptions, 2. apply appropriate measure and 3. interpret results. At least steps 1 and 3 of this workflow are not simple.

In step 1, the evolutionary model that best fits the data should be identified to allow judging whether assumptions of the measures of PNC are fulfilled (e.g. PS requires BM). Such a model comparisons should not be based on the BM/WN/OU₁ trilogy (Cooper, Jetz & Freckleton 2010; Wiens *et al.* 2010) because in doing so, one may misidentify complex scenarios of niche evolution as BM and single-optimum OU processes as WN (see Fig. 5). The necessity to carefully test model assumptions and to account for more complex OU models with different selection regimes in different parts of the phylogeny puts the advantage of simplicity already in a whole different light. In addition, there is good evidence from the literature and from our data that in most cases, simple assumptions will not be fulfilled (Estes & Arnold 2007; Harmon *et al.* 2010; Uyeda *et al.* 2011).

In step 3, results of the simple measures need to be interpreted, or in other words, the question of what we learn from the applied simple measures needs to be answered. This interpretation depends on our ideas about PNC (Is it

a process or a pattern? Which constraints are needed to produce PNC?). Therefore, calculating PS or the niche retention parameter δ in step 2 makes only sense if we accept that PNC is a pattern and that a random pattern suffices to indicate PNC (given that we can only unambiguously interpret PS when we assume a BM-like process), a view with which the authors of this study disagree. We would like to highlight that PS of course remains useful when the interest is not in PNC but in measuring how strongly closely related species resemble each other in their niches compared to distantly related species, a question that has a very common application in community assembly studies (Webb *et al.* 2002).

If a single-optimum OU process is identified as the underlying process of niche evolution, then we could interpret the stationary variance as an indicator of PNC. Finally, evolutionary rate is a practical and simple informative approach. However, both evolutionary rates and stationary variance only inform us on the *relative* strength of PNC. So basically, we cannot answer the question 'is there PNC' but need to reformulate to 'which clades show more or less PNC than others'. If we want an answer for a specific clade, we thus need to extend our analysis to related clades to be able to compare them with our focal clade. Again, this necessity to compare renders these approaches less simple than it seems on the first glance.

Overall, we conclude that a correct application of the simple measures is not simple at all in practice due to the required steps of assumption testing and interpretation. However, we would also like to mention that some simple measures, such as PS and evolutionary rate, could be used as simple preliminary tests of PNC because in many (but not all!) cases, they may indicate PNC even if assumptions are not fulfilled. However, when applied without checking underlying assumptions, they should not be interpreted on their own but should only be used to motivate a more in-depth study of PNC.

INTERESTING BUT COMPLEX

The second answer comes down to the postulate that conclusions on PNC should be based on an improved understanding of the niche evolution process. This requires refining the question and analyses by allowing selective regimes to change over time and to differ between clades. Recent methodological developments provide the toolkit for such refined studies. They make it possible to fit highly flexible models where different processes are allowed to act in different parts of the trees (e.g. OU models with multiple optima, Ingram & Mahler 2013; Mahler *et al.* 2013). Our analysis of the evolution of body mass in 17 groups of mammals demonstrates that such flexible models with high numbers of parameters are often the ones best fitting the data. As larger and larger phylogenies are published, these models will most probably take an increasing importance in the study of PNC (Cooper, Jetz & Freckleton 2010; Wiens *et al.* 2010).

These more complex OU models can be intuitively interpreted as representations of the macroevolutionary adaptive landscape, the analogue of Wright's adaptive landscape over long periods of time (Simpson 1944; Ingram & Mahler 2013; Mahler *et al.* 2013). In this landscape, each different optimum of the OU model is an adaptive peak, which width is determined by the stationary variance of the OU process. Strict PNC in this landscape might be evidenced when only one peak is occupied by a clade. However, when comparing the number of different peaks, the number of transitions between them and the width of peaks in different clades, the *relative* importance of PNC can be assessed. Recent work based on compilations of evolutionary rates and divergence times demonstrated that models of bounded evolution in short time-scales, with peak shifts occurring over longer time-scales, may constitute a good approximation to these processes (Uyeda *et al.* 2011). If disentangling a multi-peak OU model in its two components, that is, number of peak shifts and selection strength towards a peak, then PNC is indicated either by relative few peak shifts or by relative strong selection (i.e. narrow peaks in the adaptive landscape). However, there is no null expectation for the number of peak shifts and the strength of adaptation, and thus, the analyses of a multi-peak OU model are foremost descriptive.

One may wonder how to go beyond this more descriptive interpretation of the fitted OU models and give guidelines to decide whether or not there is PNC in a clade of interest. Such a decision must be based on a comparison. Such a comparison could either be (1.) between the clade of interest and a related clade (more PNC is identified in the clade of interest if it has less and/or narrower adaptive peaks than related clades) or (2.) between the estimated model parameters in the clade of interest and expected values under a certain null hypothesis. (1.) The first comparison requires the availability of data from related clades and ideally some hypothesis of more or less PNC in one of the clades. This comparison between clades can benefit from additional information both on the selective environmental regimes and on species. For example, a very stable niche in one clade in comparison with a quickly changing niche in another clade may be driven by a stable adaptive zone (e.g. riparian habitats for *Ginkgo biloba*, Royer, Hickey & Wing 2003) in comparison with a more quickly changing environment promoting frequent shifts between adaptive peaks. If the past spatial distributions of the clades and the respective histories of environmental regimes are known, this hypothesis can be tested and an environmental constraint as a driver of PNC can be identified. On the other hand, if not the environmental regimes but multiple trait information are available, one may use these to compare clades. For example, if some traits evolve similarly in different clades, while other traits show more PNC in one clade than in the other, this may indicate a very stable environmental regime for this clade that only selects the traits that show increased PNC (see Blomberg, Garland & Ives 2003 for such a comparison of traits with

measures of PS). (2.) One may also compare estimated model parameters in the clade of interest and expected values under a certain null hypothesis. This second type of comparison requires a consistent definition of PNC that allows deriving expected values under null models. For example, if one accepts that pure BM can be termed 'PNC' and that no further constraints besides kinship are required (i.e. the position of people advocating that PNC can be measured by PS), then either a model comparison test can be applied (PNC is identified if BM is the best model) or a flexible OU process can be fitted (PNC cannot be rejected if fitted selection strengths are not significantly different from zero and evolutionary rate is constant). If one defines PNC more strictly, null expectations have to come from additional information. For example, PNC driven by population dynamics or genetic constraints may be identified by significantly fewer niche shifts in a clade than expected given information on the changes of driving selective environmental regimes. Formulating adequate null models and deriving expected values in specific case studies may be challenging and may fuel further debates. However, one of the strong advantages of this approach is that defining a null model requires openly presenting underlying assumptions. We hope that this will strengthen the discussions on conflicting assumptions in the debate on conflicting evidence for PNC in nature.

Conclusions

Revisiting the published work on PNC and analysing additional simulated and real data sets, we come to the conclusion that the simple question 'is there PNC in my clade of interest' cannot be studied by simply applying a simple measure. The reasons are that

1. Simple measures are not that simple as they require in-depth pretest of the underlying assumption of a specific niche evolution models.
2. Some simple measures are uninformative as they require BM but are predefined under BM. The common assumption that strong PS indicates PNC will be invalid most of the time.
3. Simple measures become less simple if they require comparing values in the clade of interest with values in related clades (e.g. evolutionary rate).

We strongly advocate against being oversimplistic with both the question and the analysis when studying PNC. PNC needs to be well-defined such that the hypothesis of its presence is rejectable and the assumptions of approaches which are then applied need to be fulfilled. To date, the most promising approaches that we see for investigating PNC are as follows:

1. Model comparisons excluding WN model and instead including highly flexible OU models combined with a power test to avoid overfitting.
2. Fitting flexible OU models that can be descriptively interpreted as macroevolutionary adaptive landscapes,

where PNC is indicated by relative few peak shifts or by relative strong selection strength. Tests of PNC in a focal clade that are based on a comparison either with related clades (e.g. by comparing evolutionary rates when one is only interested in the pattern perspective) or with well thought through null model expectations.

Although parts of the problems that we highlight in this study have already been published, they have not been considered in many applied studies. We hope that our study helps to highlight and illustrate some of the most common problems and will pave the way to a better common practice.

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References

- Ackerly, D.D. (2004) Adaptation, niche conservatism, and convergence: comparative studies of leaf evolution in the California chaparral. *The American Naturalist*, **163**, 654–671.
- Ackerly, D. (2009) Conservatism and diversification of plant functional traits: evolutionary rates versus phylogenetic signal. *Proceedings of the National Academy of Sciences of the United States of America*, **106**, 19699–19706.
- Akaike, H. (1974) A new look at statistical model identification. *IEEE Transactions on Automatic Control*, **AU-19**, 716–722.
- Beaulieu, J.M., Jhwueng, D.C., Boettiger, C. & O'Meara, B.C. (2012) Modeling stabilizing selection: expanding the Ornstein-Uhlenbeck Model of adaptive evolution. *Evolution*, **66**, 2369–2383.
- Bininda-Emonds, O.R.P., Cardillo, M., Jones, K.E., MacPhee, R.D.E., Beck, R.M.D., Grenyer, R. *et al.* (2008) The delayed rise of present-day mammals (vol 446, pg 507, 2007). *Nature*, **456**, 274.
- Blomberg, S.P. & Garland, T. (2002) Tempo and mode in evolution: phylogenetic inertia, adaptation and comparative methods. *Journal of Evolutionary Biology*, **15**, 899–910.
- Blomberg, S.P., Garland, T. & Ives, A.R. (2003) Testing for phylogenetic signal in comparative data: behavioral traits are more labile. *Evolution*, **57**, 717–745.
- Boettiger, C.D., Ralph, P.L. & Coop, G. (2011) Is your phylogeny informative? Measuring the power of comparative methods. *Integrative and Comparative Biology*, **51**, E13.
- Boucher, F.C., Thuiller, W., Roquet, C., Douzet, R., Aubert, S., Alvarez, N. *et al.* (2012) Reconstructing the origins of high-alpine niches and cushion life form in the genus *Androsace* S.L. (Primulaceae). *Evolution*, **66**, 1255–1268.
- Boucher, F.C., Thuiller, W., Davies, T.J. & Lavergne, S. (2014) Neutral biogeography and the evolution of climatic niches. *The American Naturalist*, **183**, 573–584.
- Buckley, L.B., Davies, T.J., Ackerly, D.D., Kraft, N.J.B., Harrison, S.P., Anacker, B.L. *et al.* (2010) Phylogeny, niche conservatism and the latitudinal diversity gradient in mammals. *Proceedings of the Royal Society of London. Series B, Biological Sciences*, **277**, 2131–2138.

- Burns, J.H. & Strauss, S.Y. (2011) More closely related species are more ecologically similar in an experimental test. *Proceedings of the National Academy of Sciences of the United States of America*, **108**, 5302–5307.
- Butler, M.A. & King, A.A. (2004) Phylogenetic comparative analysis: a modeling approach for adaptive evolution. *The American Naturalist*, **164**, 683–695.
- Cooper, N., Freckleton, R.P. & Jetz, W. (2011) Phylogenetic conservatism of environmental niches in mammals. *Proceedings of the Royal Society of London. Series B, Biological Sciences*, **278**, 2384–2391.
- Cooper, N., Jetz, W. & Freckleton, R.P. (2010) Phylogenetic comparative approaches for studying niche conservatism. *Journal of Evolutionary Biology*, **23**, 2529–2539.
- Crisp, M.D. & Cook, L.G. (2012) Phylogenetic niche conservatism: what are the underlying evolutionary and ecological causes? *New Phytologist*, **196**, 681–694.
- Darwin, C. (1859) *On the Origin of Species by Means of Natural Selection, or the Preservation of Favoured Races in the Struggle for Life*. John Murray, London.
- Desdevises, Y., Legendre, P., Azouzi, L. & Morand, S. (2003) Quantifying phylogenetically structured environmental variation. *Evolution*, **57**, 2647–2652.
- Eastman, J.M., Alfaro, M.E., Joyce, P., Hipp, A.L. & Harmon, L.J. (2011) A novel comparative method for identifying shifts in the rate of character evolution on trees. *Evolution*, **65**, 3578–3589.
- Estes, S. & Arnold, S.J. (2007) Resolving the paradox of stasis: models with stabilizing selection explain evolutionary divergence on all timescales. *The American Naturalist*, **169**, 227–244.
- Evans, M.E.K., Smith, S.A., Flynn, R.S. & Donoghue, M.J. (2009) Climate, niche evolution, and diversification of the “bird-cage” evening primroses (*Oenothera*, sections *Anogra* and *Kleinia*). *The American Naturalist*, **173**, 225–240.
- Felsenstein, J. (1985) Phylogenies and the comparative method. *The American Naturalist*, **125**, 1–15.
- Freckleton, R.P. & Jetz, W. (2009) Space versus phylogeny: disentangling phylogenetic and spatial signals in comparative data. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, **276**, 21–30.
- Gomez, J.M., Verdu, M. & Perfectti, F. (2010) Ecological interactions are evolutionarily conserved across the entire tree of life. *Nature*, **465**, 918–919.
- Gomulkiewicz, R. & Houle, D. (2009) Demographic and genetic constraints on evolution. *The American Naturalist*, **174**, E218–E229.
- Grafen, A. (1989) The phylogenetic regression. *Philosophical Transactions of the Royal Society of London. Series B: Biological Sciences*, **326**, 119–157.
- Hansen, T.F. (1997) Stabilizing selection and the comparative analysis of adaptation. *Evolution*, **51**, 1341–1351.
- Hansen, T.F. (2012) Adaptive Landscapes and Macroevolutionary Dynamics. *The Adaptive Landscape in Evolutionary Biology* (eds E. Svensson & R. Calsbeek), pp. 205–226. Oxford University Press, Oxford.
- Hansen, T.F. & Bartoszek, K. (2012) Interpreting the evolutionary regression: the interplay between observational and biological errors in phylogenetic comparative studies. *Systematic Biology*, **61**, 413–425.
- Hansen, T.F. & Houle, D. (2004) Evolvability, stabilizing selection, and the problem of stasis. *Phenotypic Integration: Studying the Ecology and Evolution of Complex Phenotypes* (eds M. Pigliucci & K. Preston), pp. 130–150. Oxford University Press, Oxford.
- Hansen, T.F. & Martins, E.P. (1996) Translating between microevolutionary process and macroevolutionary patterns: the correlation structure of interspecific data. *Evolution*, **50**, 1404–1417.
- Hansen, T.F., Pienaar, J. & Orzack, S.H. (2008) A comparative method for studying adaptation to a randomly evolving environment. *Evolution*, **62**, 1965–1977.
- Harmon, L.J., Losos, J.B., Davies, T.J., Gillespie, R.G., Gittleman, J.L., Jennings, W.B. et al. (2010) Early bursts of body size and shape evolution are rare in comparative data. *Evolution*, **64**, 2385–2396.
- Harvey, P.H. & Pagel, M. (1991) *The Comparative Method in Evolutionary Biology*. Oxford University Press, Oxford.
- Hillis, D.M. & Huelsenbeck, J.P. (1992) Signal, noise, and reliability in molecular phylogenetic analyses. *Journal of Heredity*, **83**, 189–195.
- Ho, L.S.T. & Ane, C. (2013) Asymptotic theory with hierarchical autocorrelation: Ornstein-Uhlenbeck tree models. *Annals of Statistics*, **41**, 957–981.
- Holt, R.D. (1996) Demographic constraints in evolution: towards unifying the evolutionary theories of senescence and niche conservatism. *Evolutionary Ecology*, **10**, 1–11.
- Hunt, G. (2012) Measuring rates of phenotypic evolution and the inseparability of tempo and mode. *Paleobiology*, **38**, 351–373.
- Hutchinson, G.E. (1957) Concluding remarks. *Cold Spring Harbor Symposia Quantitative Biology*, **41**, 5–427.
- Ingram, T. & Kai, Y. (2014) The geography of morphological convergence in the radiations of Pacific Sebastes rockfishes. *The American Naturalist*, **184**, E115–E131.
- Ingram, T. & Mahler, D.L. (2013) SURFACE: detecting convergent evolution from comparative data by fitting Ornstein-Uhlenbeck models with stepwise Akaike Information Criterion. *Methods in Ecology and Evolution*, **4**, 416–425.
- Jetz, W., Thomas, G.H., Joy, J.B., Hartmann, K. & Mooers, A.O. (2012) The global diversity of birds in space and time. *Nature*, **491**, 444–448.
- Lande, R. (1976) Natural selection and random genetic drift in phenotypic evolution. *Evolution*, **30**, 314–334.
- Lavergne, S., Mouquet, N., Thuiller, W. & Ronce, O. (2010) Biodiversity and climate change: integrating evolutionary and ecological responses of species and communities. *Annual Review of Ecology, Evolution and Systematics*, **41**, 321–350.
- Losos, J.B. (2008) Phylogenetic niche conservatism, phylogenetic signal and the relationship between phylogenetic relatedness and ecological similarity among species. *Ecology Letters*, **11**, 995–1003.
- Losos, J.B. (2011) Seeing the forest for the trees: the limitations of phylogenies in comparative biology. *The American Naturalist*, **177**, 709–727.
- Mahler, D.L., Ingram, T., Revell, L.J. & Losos, J.B. (2013) Exceptional convergence on the macroevolutionary landscape in island lizard radiations. *Science*, **341**, 292–295.
- Münkemüller, T., Lavergne, S., Bzeznik, B., Dray, S., Jombart, T., Schiffrers, K. et al. (2012) How to measure and test phylogenetic signal. *Methods in Ecology and Evolution*, **3**, 743–756.
- O’Meara, B.C., Ane, C., Sanderson, M.J. & Wainwright, P.C. (2006) Testing for different rates of continuous trait evolution using likelihood. *Evolution*, **60**, 922–933.
- Pagel, M. (1999) Inferring the historical patterns of biological evolution. *Nature*, **401**, 877–884.
- Pearman, P.B., Guisan, A., Broennimann, O. & Randin, C.F. (2008) Niche dynamics in space and time. *Trends in Ecology & Evolution*, **23**, 149–158.
- Pearman, P.B., Lavergne, S., Roquet, C., Wüest, R., Zimmermann, N.E. & Thuiller, W. (2014) Phylogenetic patterns of climatic, habitat and trophic niches in a European avian assemblage. *Global Ecology and Biogeography*, **23**, 414–424.
- Pennell, M.W., Harmon, L.J. & Uyeda, J.C. (2014) Speciation is unlikely to drive divergence rates. *Trends in Ecology & Evolution*, **29**, 72–73.
- Pennell, M.W., FitzJohn, R.G., Cornwell, W.K. & Harmon, L.J. (2014) Model adequacy and the macroevolution of angiosperm functional traits. *bioRxiv*, Available at: <http://biorxiv.org/content/early/2014/04/07/004002>.
- Peterson, A.T., Soberon, J. & Sanchez-Cordero, V. (1999) Conservatism of ecological niches in evolutionary time. *Science*, **285**, 1265–1267.
- Prinzing, A., Durka, W., Klotz, S. & Brandl, R. (2001) The niche of higher plants: evidence for phylogenetic conservatism. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, **268**, 2383–2389.
- Pyron, R.A., Burbrink, F.T. & Wiens, J.J. (2013) A phylogeny and revised classification of Squamata, including 4161 species of lizards and snakes. *BMC Evolutionary Biology*, **13**, 93.
- Prugh, L. & Golden, C. (2013) Data from: Does moonlight increase predation risk? Meta-analysis reveals divergent responses of nocturnal mammals to lunar cycles. *Dryad Digital Repository*, <http://dx.doi.org/10.5061/dryad.tn723>.
- Revell, L.J., Harmon, L.J. & Collar, D.C. (2008) Phylogenetic signal, evolutionary process, and rate. *Systematic Biology*, **57**, 591–601.
- Ricklefs, R.E. (1987) Community diversity: relative roles of local and regional processes. *Science*, **235**, 167–171.
- Royer, D.L., Hickey, L.J. & Wing, S.L. (2003) Ecological conservatism in the “living fossil” Ginkgo. *Paleobiology*, **29**, 84–104.
- Simpson, G.G. (1944) *Tempo and Mode in Evolution*. Columbia University Press, New York, NY.
- Thomas, G.H. & Freckleton, R.P. (2012) MOTMOT: models of trait macroevolution on trees. *Methods in Ecology and Evolution*, **3**, 145–151.
- Thomas, G.H., Freckleton, R.P. & Szekely, T. (2006) Comparative analyses of the influence of developmental mode on phenotypic diversification

- rates in shorebirds. *Proceedings of the Royal Society of London. Series B, Biological Sciences*, **273**, 1619–1624.
- Thuiller, W., Lavergne, S., Roquet, C., Boulangeat, I. & Araujo, M.B. (2011) Consequences of climate change to the Tree of Life in Europe. *Nature*, **470**, 531–534.
- Uyeda, J.C., Hansen, T.F., Arnold, S.J. & Pienaar, J. (2011) The million-year wait for macroevolutionary bursts. *Proceedings of the National Academy of Sciences of the United States of America*, **108**, 15908–15913.
- Walsh, B. & Blows, M.W. (2009) Abundant genetic variation plus strong selection = multivariate genetic constraints: a geometric view of adaptation. *Annual Review of Ecology Evolution and Systematics*, **40**, 41–59.
- Webb, C.O., Ackerly, D.D., McPeck, M.A. & Donoghue, M.J. (2002) Phylogenies and community ecology. *Annual Review of Ecology and Systematics*, **33**, 475–505.
- Wiens, J.J. (2008) Commentary on Losos (2008): niche conservatism *deja vu*. *Ecology Letters*, **11**, 1004–1005.
- Wiens, J.J. & Graham, C.H. (2005) Niche conservatism: integrating evolution, ecology, and conservation biology. *Annual Review of Ecology Evolution and Systematics*, **36**, 519–539.
- 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. *Ecology Letters*, **13**, 1310–1324.
- Zanne, A.E., Tank, D.C., Cornwell, W.K., Eastman, J.M., Smith, S.A., FitzJohn, R.G. *et al.* (2014) Three keys to the radiation of angiosperms into freezing environments. *Nature*, **506**, 89–92.

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Supporting Information

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

Appendix S1. Simulated data.

Appendix S2. Approaches to estimate PNC.

Appendix S3. Mammal data.

Table S1. Results for the analyses of body mass evolution along the phylogeny.

Fig. S1. Application of PNC approaches to mammal data.