Jointly Modeling the Evolution of Discrete and Continuous Traits

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*Abstract*

Rates of evolution have changed throughout the history of life and produced the rich diversity of morphology, behaviour, and ecology that characterizes the biodiversity we see today. As such, models favoring variation in rates and states has fast become the rule, as opposed to the rare exception in empirical applications of comparative methods. Our model, which we call hOUwie, uses hidden Markov models as a statistically rigorous way to learn about variation in the evolutionary process. hOUwie is composed of two processes: one describes the evolution of a discrete character (such as whether a mammal is a carnivore, herbivore, or omnivore) and the other the evolution of a continuous character (such as mammalian body size).

Rates of evolution have changed throughout the history of life and produced the rich diversity of morphology, behaviour, and ecology that characterizes the biodiversity we see today. As such, models favoring variation in rates and states has fast become the rule, as opposed to the rare exception in empirical applications of comparative methods.

Rate variation b/c biology and methods. BMV still active. OU more realistic. OU extended multiple times. OU needs rate variation. OU variation methods and biology. Leads to data drive vs hypothesis driven.

Questions of how plant life habit influence genome size evolution (Beaulieu et al. 2012), how substrate use alters limb length evolution (Mahler et al. 2013), or how tooth morphology slowly changes in response to habitat and diet (Toljagić et al. 2018) are evidence of a recurring interest in testing whether evolutionary changes in one variable is linked to change in another. To study questions such as these, one very common phylogenetic comparative approach is to employ an Ornstein-Uhlenbeck (OU) model, which assumes distinct regimes, described by the evolution of a discrete character, are known completely *a priori* (e.g., Butler and King 2004; Hansen et al. 2008; Beaulieu et al. 2012), or assumes that “shifts” in regimes can be inferred directly from the distribution of the continuous trait (e.g. Ingram and Mahler 2013; Uyeda and Harmon 2014; Khabbazian et al. 2016). While these approaches are practical, they assume that the combination of independently estimated discrete and continuous models will produce a joint model, but with the discrete trait being the driving force underlying the evolution of the continuous character. However, evolution is rarely as simple as cause and effect, and we suspect that as often as a discrete character causes change in the continuous character, continuous characters also influence discrete character evolution, or at the very minimum, help provide information about this evolution.

Progress along these lines has mostly involved acknowledging uncertainty in the evolution of the discrete character by way of stochastic maps. That is, a large set of alternative reconstructions of the discrete character are obtained completely uninformed by the continuous trait’s evolution, then the likelihood of the continuous character becomes the average of the likelihoods across these maps (e.g., Revell 2012). The advantage of this approach is that there is an explicit model for how regimes change through time, but the evolution of these regimes remains entirely independent of the continuous trait, and the probability of these regimes is not explicitly considered. For example, it may be possible that the model which best fits the discrete data generates stochastic maps which do not fit the continuous data. A promising approach was recently described for detecting adaptive codon evolution (Jones et al 2020), where a set of maps obtained for a discrete phenotype under a standard Markov process is optimized along with parameters associated with genotype properties, thus forcing an emergent dependency between the two. Similarly, May and Moore (2020) developed a joint model for discrete and continuous characters under a state-dependent Brownian motion model. Their approach takes advantage of a Bayesian framework through the use of priors in order to accommodate variation in the “background” rate of evolution in the continuous trait (i.e., rate variation across lineages that is independent of the discrete character under consideration). The novel Bayesian pipeline recently developed by Tribble et al. (2021) is the first attempt that we are aware of for jointly modeling discrete and continuous traits under an OU framework. Their approach samples discrete stochastic mappings informed by the discrete trait along with regime mappings which were informed by the continuous trait while accounting for the potential of hidden variation. This method allows for a more effective test of correlation between the discrete and continuous characters. One drawback, however, is that they do not explicitly account for the joint probability of the discrete and continuous parameter estimates together. They assume that the combination of independently estimated discrete and continuous models produces a joint estimate.

Here we propose an explicitly joint estimate of the likelihood for a discrete and continuous character by combining the probability of the continuous character given a particular regime and the probability of that discrete regime painting, integrated over many regime paintings. Specifically, we combine hidden Markov models of discrete character evolution (Beaulieu et al. 2013) with generalized Ornstein-Uhlenbeck models (Hansen 1997; Butler and King 2004; Hansen et al. 2008; Beaulieu et al. 2012; Ho and Ané 2014a). We demonstrate how our framework, which we call “hOUwie”, can be used to test hypotheses of correlated evolution between discrete and continuous characters while also accounting for hidden character states and unobserved variation. Finally, we apply several hOUwie models to test the correlated dynamics of the mode of seed dispersal and climatic niche evolution and compare our results to those that did not account for the potential joint evolution of discrete and continuous variables.

**Materials and Methods**

*The hOUwie model*

Our model is composed of two processes: one that describes the evolution of a discrete character and the other describes the evolution of a continuous character. To model the evolution of a single continuous character we use an Ornstein-Uhlenbeck (OU) model (Hansen 1997; Butler and King 2004; Hansen et al. 2008; Beaulieu et al. 2012; Ho and Ané 2014a). Formally, the OU process is an Itô diffusion satisfying:

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This model combines the stochastic evolution of a trait through time with a deterministic component which models the tendency for a trait to evolve towards an optimum. In this model, the value of a trait, , is pulled towards an optimum, , at a rate scaled by the parameter . The optimum, , is a piecewise constant on intervals and takes values in a finite set. This can represent the set of “selective regimes”, “regimes”, or Simpson’s “adaptive zones” (Cressler et al. 2015), though it is consistent with a variety of true underlying microevolutionary models (Hansen 2014). Additionally, random deviations are introduced by Gaussian white noise , which is distributed as a normal random variable with mean zero and variance equal to . Thus, is a constant describing the rate of stochastic evolution away from the optimum. We use the set of extensions introduced by Beaulieu et al. (2012) and implemented in the R package OUwie. This allows for multiple primary optima in which both the pull strength () and the rate of stochastic evolution () can vary across the phylogeny. However, the algorithm used to calculate the likelihood described in Beaulieu et al. (2012) involves matrix inversion -- a computationally costly procedure. Therefore, we implement a linear-time computation of the likelihood of Gaussian trait models following (Ho and Ané 2014a). To do this, we first transform the phylogeny such that its variance covariance matrix, , is 3-point structured. We can write the variance covariance matrix of the untransformed phylogeny as , where following Beaulieu et al. (2012) and Ho and Ané (2014),

and,

where, is the distance from the root to the beginning of the selective regime () for the number of selective regimes along the path from the root to the last common ancestor of and , or from the root to the terminal tip. Our transformed phylogeny now has a variance covariance matrix and diagonal matrix . We can then calculate the quadratic quantities and determinant of (Ho and Ané 2014a). The probability of our continuous trait is given by

where is the number of tips in the phylogeny (, is the continuous trait value of each species, and is the expected value of each species given the continuous trait model – calculated following equation (11) of Beaulieu et al. (2012), *D* is the discrete character data, is a particular regime mapping, and are the parameters of the hOUwie model.

Next, we describe the calculation of the probability of the underlying regime structure, , which is the joint probability of discret­e characters () and stochastic mapping (). This calculation is analogous to the pathway likelihood of Steel and Penny (2000). Recently, May and Moore (2020) suggested that the joint probability of a regime structure and the discrete character is the product of the probabilities of exponentially distributed waiting times. By this definition, branch lengths are the sum of waiting times. However, under this definition, when we calculate the probability of starting and ending a branch in state *i*, the likelihood of a regime structure was unaffected by the number of transitions (Supplemental Materials), even though the maximum likelihood estimate should be zero transitions (O’Meara 2008). We suspect this is an issue of treating the probability density functions independently, when their convolution is required, because the time of the second transition depends on the timing of the first as well as the total branch length available. Ultimately, the effect of this inaccuracy is that the number of transitions has no influence on the probability of a branch’s state reconstruction and that the sum of the joint probabilities will be greater than the marginal probability. For further details, and R code necessary to demonstrate these issues, we refer readers to the Supplemental Materials.

To calculate the probability of discret­e characters () and stochastic mapping () we instead use an approximation. Our approximation relies on a finite number of degree-2 internodes and uses the standard Chapman-Kolmgorov equation to calculate the probabilities of beginning in a particular state and ending in state (Pagel 1994) and is identical to a marginal probability of a set of state reconstructions (Yang 2006). As the number of internodes increase, the amount of time between nodes decreases and the approximation improves (Rao and Teh 2013). The joint probability of a regime structure and the discrete character i

where the instantaneous rate matrix (, is the phylogeny, is the root state probability (Pagel 1994; Yang 2006; Maddison et al. 2007), is the number of external nodes (tips), internal nodes, and internodes (degree-2 nodes) summed, indicates a particular branch, , where is an indicator function which ensures that we only use the probability of states indicated by the specific the regime mapping instead of summing over all possible state combinations. The continuous character probability requires the discrete state(s) to be defined along the entire branch, thus we place transitions halfway between any two nodes.

For each set of parameters evaluated during the maximum likelihood search, a set of stochastic mappings are generated to evaluate the discrete and continuous likelihoods (Fig. 3). To do this, we first approximate of the conditional state probabilities at nodes. The conditional state probability, unlike the more common marginal reconstruction or joint state reconstruction (Pupko et al. 2000; Felsenstein 2004; Yang 2006), calculates the probability that a node has a particular state value conditioned only on the observations of its descendants. For a particular focal node, we calculate the probability of the observing all pairwise descendant values given the OU model parameters, integrated over all possible rootward node states, and observed tipward discrete states (Fig 3d, see Supplemental for more detail). Although this is only an approximation of the conditional state probabilities, it proves to be an essential improvement over the typical procedure of sampling many stochastic maps based solely on the discrete process (Fig. 4). Next, the conditional probabilities of states at nodes are sampled starting with the root. Once the root is sampled, descendent states are sampled based on both the conditional ancestral values and the sampled ancestral state. This is achieved by multiplying the conditional probability of the node states by the probability of starting in the sampled rootward ancestral value and ending in any of the tipward states (the latter is calculated using familiar matrix exponentiation methods (e.g., Pagel 1994). Finally, under usual stochastic mapping procedures we would use rejection sampling (Nielsen 2002; Rao and Teh 2013) to simulate a path between the sampled rootward and tip ward nodes. However, for increased computational efficiency, we opt to place transitions at pre-defined internodes. Thus, under our procedure, after nodes and internodes are sampled in step two, mappings are evaluated to ensure consistency with the discrete model (i.e., impossible transitions do not occur) and branches are painted based on the sampled nodes with transitions occurring half-way between nodes or internodes.

Our likelihood function for the joint probability of a continuous and a discrete character is therefore,

where summing over all generated maps (), is the probability of the continuous character () given the discrete character data (), mapping (), hOUwie parameters (), and phylogeny (). is the joint probability of the discrete character data () and stochastic mapping () given the hOUwie parameters () and phylogeny ().

*The hOUwie model space*

Our simulation studies examined 22 possible hOUwie model structures for a binary discrete character, although the possible number of models is significantly higher. For the discrete component of the model, we assumed that transitions between the observed characters were equal, although when hidden states were included, rates within the hidden rate classes differed. We constrained transitions between hidden states to be the same for observed states, but this constraint can be relaxed if desired. The continuous model structures allowable in hOUwie are a generalized form of those allowed in OUwie and now include models in which only alpha varies (OUA), only sigma varies (OUV), and combinations of an OU and BM process (OUBM1 and OUBMV). We note that the OUBM1 model within hOUwie, differs from The Ornstein–Uhlenbeck Brownian-motion (OUBM) model presented in Hansen et al. (2008) and Bartoszek et al. (2012) since latter models are of multiple continuous characters, rather than different processes describing the same continuous character.

The potential model structures range from completely character-dependent to character-independent. Character-dependent (CD) models are models in which any continuous OU parameter differs between observed discrete state, whereas character-independent models (CID) test whether observed discrete states can be described by the same OU parameters. There are two types of character-independent model (Fig. 1). First, character-independent models include structures where there are no differences between any OU parameters. Under this model the entire evolutionary history of the clade can be described by a single alpha, sigma, and optimum value (Fig. 1a). To combat this unrealistic assumption we introduce a character-independent model which allows for differences in the OU parameters to depend upon an unobserved hidden state (CID+) and has been shown to correct for the bias towards detecting correlation (Boyko and Beaulieu 2022). This addition allows for heterogeneity within the evolutionary process without the necessity of it being linked to a focal trait (Fig. 1c). In total we examine 22 unique model structures (2 CID, 10 CD, and 10CID+).

*Simulation study*

For each of the 22 hOUwie model structures, we simulated 50 datasets for phylogenies of 25, 100, and 250 taxa for a total of 3300 unique datasets. Phylogenies were pure birth phylogenetic tree with , and rescaled tree height to 1. The root state was fixed to state 1. The parameters used to generate a phenotypic dataset depend on the structure of the generating model. For example, an OUM model and OU1 model can have identical , but they must differ in or else OUM will collapse into OU1 (model structures associated with model name are shown in Table 1). The simulating parameters were chosen to match Beaulieu et al. (2012) with . Once a phylogeny and phenotypic dataset were simulated, we fit our models to assess parameter estimation accuracy and model selection power. Although this represents a small subset of the potentially vast parameter space available to OU models, the behavior of these models has been thoroughly characterized and thus we chose parameters within the range of typical identifiability (Beaulieu et al. 2012; Ho and Ané 2014*a*; Cressler et al. 2015). Additionally, because hOUwie uses a variable number of mappings, we evaluate changing the number of stochastic maps. We fit each model using 25, 100, and 250 stochastic mappings per likelihood evaluation. Each dataset was evaluated using the true generating model, a BM1, an OU1, and either the character-dependent or character-independent counterpart to the generating model. For example, if the data was simulated under a character-dependent OUM model where the value of depend on the observed character, a character-independent OUM model would also be fit as part of the model set. Under the CID+ OUM model, a variable is still allowed, but it is unlinked to the focal character and thus should provide a more reliable character independent null hypothesis than BM1 or OU1 (Beaulieu and O’Meara 2016; Boyko and Beaulieu 2022).

*The impact of climatic variables on seed dispersal*

An ideal way to address the complexity of a trait syndrome would be to enumerate all the phenotypes which comprise the seed dispersal syndrome and study the dynamics in-depth. However, on macroevolutionary scales, this approach is intractable due to the vast amount of species-specific data required and the increasing number of parameters introduced into models as the number of traits increases. Thus, we propose an alternative which will use a single feature of fruit morphology as a proxy for seed dispersal, but also acknowledges that not all lineages with the same fruit morphology are going to evolve in the same way. It is for this reason that we use hidden Markov models (HMM) as the basis for describing discrete character evolution. With HMMs we allow for the fact that dynamics within broadly defined categories are likely inadequate descriptions when treated homogeneously.

Here we examine three predictions outlined in Vasconcelos et al. (2021), but specifically measuring aridity. First, we expect that the climatic optima for abiotically dispersed seeds will be drier compared to biotically dispersed seeds (). Second, we expect that abiotically dispersed seeds will have faster rates of climatic niche evolution (). Finally, we expect that the climatic niches of biotically dispersed seeds will be more conserved through time (). We apply several hOUwie models to test these hypotheses and compare our results to those discussed in Vasconcelos et al. (2021). We expect that any differences found between this study and Vasconcelos et al. (2021) are because we can explicitly account for the joint probability of the discrete and continuous characters. We choose Ericaceae specifically because Vasconcelos et al. (2021) found two counter-intuitive results. First, they found that the phenotypic optima of abiotically dispersed seeds was more humid than biotically dispersed seeds. Second, they found that the rate of climatic evolution was greater in biotically dispersed seeds than abiotically dispersed seeds.

We include 25 hOUwie models within our model set: 2 CID, 10 CD, 10 CID+, and 3 HYB. *Gaultheria* is technically a dry-fruited genus within Ericaceae but has a persistent fleshy calyx that attracts frugivores (Stevens et al. 2004). For that reason, we code this as fleshy fruited within our dataset. Models are evaluated using the sample size corrected Akaike Information Criterion (AICc) and model averaging is conducted when discussing how our results relate to our hypotheses (Burnham and Anderson 2002). Finally, we evaluate the expected values, the optima, and compare our results to Vasconcelos et al. (2021).

**Results**

*Simulation study*

For character independent models, our heuristic adaptive sampling algorithm consistently produced more probable mappings than using purely discrete mappings for all models examined. On average, adaptive sampling produced mappings which were 38 log likelihood units better than purely discrete sampling when examining joint probabilities. This was driven primarily by the improved continuous probabilities which were on average 38.4 log likelihood units better. In contrast, the discrete probability of each mapping was similar with discrete-only simulations producing maps that were on average 0.39 log likelihood units better (Table 1; Figure 3). For character-dependent models, the difference was negligible (not shown). This is because when the discrete and continuous character are linked, discrete-only mappings will match the continuous character’s distribution well.

Most character dependent models showed low parameter estimation errors across all model types. Like previous studies, the largest RMSE was for alpha at 1.76 and 1.65 (if variable alpha) and errors were generally higher for more complex models. All other parameters had relatively similar RMSE, ranging from 0.1 for discrete the rate to 0.75 for sigma^2\_2. The BMV, OUV, OUA, and OUM models generally had the lowest errors, but there were some biases present. Most notably, alpha was biased upwards for OUM and OUV models and under variable alpha models, the difference between the estimates tended to be overestimated. The more complex models had larger error variances but showed similar biases as the simple models. Finally, OUBM models showed a significantly downward biased alpha, suggesting BM like processes (Figure 5; Table 2).

Character independent models with rate heterogeneity models generally performed well in terms of parameter estimates, but as expected, due to their inherit uncertainty, CID+ models had larger errors than CD models. The largest error was estimates of sigma^2\_2 which had an RMSE of 8.5, although the median error value was only 0.03, suggesting that the large RMSE is driven by a long rightward tail of the estimates. Alpha\_1 and alpha\_2 consistently showed large RMSE at 3.6 and 1.2. In general, alpha was underestimated with medians of -0.4 and -1.4 below the simulating values of 3 and 1.5. This means that models for CID+ models tended to be more BM like even under an OU generated data (Figure 5; Table 2).

Increasing the number of taxa examined improved both CD and CID+ performance. Alpha RMSE was nearly cut in half between when moving from 25 tips to 250 tips from 5.2 to 2.8 under CID+ models. Nonetheless, some parameters continued to be estimated poorly, such as sigma2\_2. Interestingly, increasing the number of stochastic maps improved CID+ performance, but did not substantially improve estimation under CD models (Fig. 5bc).

Generally, evidence of CD when it was the generating model was consistent across all model types. The lowest support for the OUA and OUBM1 models at an average AICwt of 0.31 and 0.13. For complex models, such as OUMVA, model support for was 0.81 and highest for OUMV at 0.97. CID+ models fared worse in terms of generating consistent support even when they were the generating model. Models which were difficult to estimate under character dependence were difficult to find consistent support for under character independence. The most extreme case was OUA model for which CID+ model was never chosen as the best supported model. However, models which performed well for CD tended to perform well under CID+. For example, OUM models garnered consistent support when with an average AICwt of 0.733 (Table 3; Figure 6).

For both CD and CID+ models, support improved when increasing the number of tips analyzed. Support for a CD model when CD was the generating model increased from 0 to to for 25, 100, 250 tips and support for a CID+ model when it was the generating model increased from tp to . Similarly, increasing the number of stochastic maps generally improved the fit, but not as much as increasing the number of tips. We found that false evidence of correlation was generally not an issue for variable theta models. Variable theta models had average AICwts for false character-dependence ranging from 0.03 to 0.23 and for none of our simulations models was a CD model best supported. Under a simple OUM model, CID+ models helped correct any potential bias with an average AICwt of 0.68. However, false evidence of correlation was an issue for variable sigma and alpha models. False support for CD ranged from 0.34 to 0.44 when theta was fixed and alpha and/or sigma varied. Although CID+ models did not garner much support when these models were fit, OU1 and BM1 models served as reasonable null hypotheses in these cases. In general, we found that when CID models were the generating model, evidence of CID was strongest and when CD models were the generating model, evidence of character dependence was strongest. This suggests that the effect of rate heterogeneity causing false correlations is not as pronounced as other comparative methods (Table 3; Figure 6).

*Seed dispersal and climatic evolution*

We found evidence of a character dependent model over either a simple or hidden state character independent model, suggesting a link between the climatic niche of Ericaceae lineages and their fruit type (Table 6). The best supported models were OUMVA and OUVA with AIC weights of 0.41 and 0.32 respectively. This suggests that there were character dependent differences in phenotypic optima, rates of evolution, and overall phylogenetic signal. To evaluate support for our hypotheses we examined the model averaged parameter estimates (Table 7). The estimated optimum 0.81 aridity index of fleshy fruits suggests dryland as their optimal habitat, and the 0.97 AI of dry fruits corresponds to a more humid environment (Middleton and Thomas 1997). Both and interact to create tip variance, so in addition to , we measured stationary variance . As predicted, we found that Ericaceae lineages with dry fruits were more variable in their climatic niche evolution (, ) compared to fleshy fruits (, ). Additionally, the phylogenetic signal of fleshy fruited lineages was greater than dry fruited lineages (). This corresponds to phylogenetic half-lives of and which are 38% and 25% of the total tree height respectively. Transitions to fleshy fruit occurred at 0.0015 transitions per million years which is more than 4.3 times faster than transitions to dry fruits (0.00035 transitions per million years). Given the total branch length in the tree is 10,120 MY, we would expect 15.6 transitions to fleshy fruit and 3.6 transitions to dry fruits to have occurred throughout the history of Ericaceae. Finally, on average lineages were in more arid environments than predicted by the model (average difference of 0.19 AI), with some species expected to be in much more humid environments (difference between current AI and optimal AI ranged from -4.4 to 0.85; Figure 7).

**Discussion**

Phylogenetic comparative methods have been widely applied to study discrete and continuous characters separately, but there are surprisingly few options which simultaneously use both classes of character (Felsenstein 2012). Here we describe the hOUwie model and demonstrate how it can be used to test hypotheses of correlated evolution between discrete and continuous characters while accounting for hidden character states and unobserved variation. Our model jointly models discrete and continuous characters by linking both via a common regime painting. However, unlike other similar methods, our likelihood formula explicitly calculates the probability of the underlying regimes. This has the advantage of describing the discrete character evolution probabilistically and allows information from the discrete and continuous characters to jointly contribute to the overall likelihood.

*The significance of jointly modeling characters*

Most implementations of an OU process within PCMs assume that the regimes are known or estimate them without considering discrete character evolution (Uyeda and Harmon 2014). These approaches leave little room for inference about the regimes themselves and how they change through time, nor the possibility that the evolution of the continuous character could influence change in the regimes or vice versa. The primary difference between hOUwie and other applications of the OU process is the explicit joint modeling of the underlying regime structure and continuous trait. There are three main advantages to jointly modeling discrete and continuous traits. First, it represents a more realistic view of trait evolution. The evolution of any discrete or continuous trait is not independent of all other traits a lineage possesses. Second, the mutual information of multiple variables makes our inferences more robust (Cover and Thomas 1991). If we are certain of one variable (seed size), but uncertain about another variable (climate), we can use the knowledge of the relationship between our variables to draw more accurate inferences about the uncertain variable. Finally, using hidden Markov models allows for the discovery of hidden variation. There is little reason to believe that any relationship between discrete and continuous variables will be homogenous throughout the history of a lineage. With hOUwie, it is theoretically possible to discover regions of the phylogeny where different types of relationships emerge.

A joint model also makes the parameter estimates related to continuous trait evolution independent of pre-defined regime paintings. The use of pre-defined regime mappings can be often be useful for hypothesis testing (Butler and King 2004; Beaulieu et al. 2012). For example, testing for evidence for character displacement by mapping either a sympatric or allopatric evolutionary history (Butler and King 2004). However, when using this approach, the mapping is taken as an absolute certainty. There is no room for inference of other potential patterns nor is there the acknowledgement that the *apriori* mapping is a probabilistic sample of many possibilities. Instead of contrasting mappings, hOUwie’s methodology integrates over the uncertainty of alternative regimes and relies on the interpretation of parameter estimates from contrasting model structures to find evidence for hypotheses. To that end, modeling the joint probabilities puts our models into a likelihood framework. This allows for model comparison, model averaging, and other tools to be utilized when comparing macroevolutionary hypotheses (Burnham and Anderson 2002). This framework may be preferable to testing whether a particular hypothesis is better than a trivial null model (Beaulieu and O’Meara 2016; Caetano et al. 2018). Detecting a significant correlation between two traits is an important first step in establishing an evolutionary relationship, however a richer understanding may be achieved through examining the models’ parameterization in relation to specific hypotheses. Model averaged parameters and tip rates are a way to both test preexisting hypotheses and begin developing new hypotheses based on interpretation (misse). Additionally, because the relative explanatory power of each model is related to parameter estimates, by using model averaging we account for model structure uncertainty in addition to parameter estimation uncertainty (Burnham and Anderson 2002).

*It might be tempting to critique our map sampling maps as inefficient in comparison to popular MCMC techniques*

*"One thing that surprised us was the inefficiency of sampling potential maps from the univariate simmap model. For our simulation conditions, many potential simmaps, even though fairly good for discrete characters (thus their sampling under this process), are absolutely terrible for the continuous regimes and so the joint model with these maps contributes little to the overall likelihood. For a typical run, 90% of the total likelihood for the best set of parameters came from just <small percent> of the attempted simmaps. In some ways this is good: it is clear that the continuous characters have information about the placement of regimes (see classic OU examples (bayou, ouwie, ouch, etc.) where the continuous data are all that provides info on regimes). But it also makes sampling good regimes to get an accurate estimate of the likelihood hard -- we developed our "cherry" algorithm to help with this, as sampling discrete only simmaps was not efficient enough. Approaches that pick simmaps at complete random might work even worse."*

*Seed dispersal and climatic niche evolution in Ericaceae*

For sedentary organisms, such as plants, dispersal is mainly limited to a brief stage of their life cycle and mediated mainly through the movement of seeds (Levin et al. 2003). Generally, the expectation is that seeds dispersed by frugivores are going to be dispersed to environments more like their parents’ environment, whereas abiotically dispersed seeds are likely to be more erratic in their dispersal patterns (Schupp 1993; Westoby et al. 1996). Furthermore, it has been proposed that adaptations for frugivorous dispersal is linked to tropical and subtropical biomes. This is because in these warmer and wetter habitats, large trees create shady environments where competition for light is more important. A shadier habitat then imposes a selective pressure for larger seeds because more nutrients are needed for germination (Foster and Janson 1985). However, the evolution of larger seeds comes with a tradeoff as they have a significantly lower dispersal potential (Howe and Smallwood 1982). Thus, we might expect that the climatic variables of a habitat influence the probability of transitioning between abiotic and biotic modes of dispersal, with transition rates from abiotic to biotic being greater in less arid environments. Here we revaluated three hypotheses related to climatic niche evolution and seed dispersal: (1) the climatic optima for abiotically dispersed seeds will be drier compared to biotically dispersed seeds (), (2) abiotically dispersed seeds will have faster rates of climatic niche evolution (), and (3) climatic niches of biotically dispersed seeds will be more conserved through time (). Consistent with previous findings we did not find evidence of these hypotheses in Ericaceae. This may have been the result of *Dracophyllum,* a large dry-fruited genus, being distributed in particularly wet environments (Wagstaff et al. 2010) or the possibility that arid environments tend to have a higher climatic niche evolution. The latter hypothesis is consistent with our finding that lineages in arid environments (which was predominantly associated with dry fruits) transitioned to fleshy fruits at a rate 6-times faster than the alternative transition.

*Caveats*

There are three important caveats of our model. First, our discrete mapping probability ( is merely an approximation. What we calculate here is the probability of starting in a particular state and ending a particular state , summed over all possible paths. However, the continuous model probability is based off a particular pathway history which is defined throughout the entire branch (Hansen 1997). Our justification for this approximation is that as the length between internodes decreases this approximation improves because the probability of more than a single transition (or no transitions) decrease. The second caveat is that we do not force hOUwie to sum over all possible mappings . This is because the number of mappings will grow exponentially as the number of nodes and internodes increases and the computation will quickly become infeasible. Instead, we simulate node, internodes, and tip states (tip states only in the case of hidden Markov models) using the stochastic mapping procedure described in Bollback (2006) and modified to include the possibility of internodes. Our final caveat is that because we generate node mappings using the stochastic mappings procedure, they would typically be based solely on the discrete character process. This can be particularly problematic for character independent models where the discrete character and continuous character are decoupled. An imperfect solution to this problem is to weight tip probabilities when first calculating the conditional likelihoods (Bollback 2006). Typically, tip values for hidden Markov models are given a weight of 1 for each state (Beaulieu et al. 2013). However, we can modify this value to be proportional to the probability of sampling a particular tip’s continuous trait value given the models theta and sigma value for each discrete state. This is the approach we take; we assume that the conditional probability of a tip being in a particular hidden state is not equal, but rather proportional to ; the probability of sampling the continuous trait value under a normal distribution for each possible state . In cases where there are no differences between parameters, conditional probabilities have equal weights.

*Future extensions*

*A common theme of all models is that it is only the parameters that shift, not the trait values themselves. Biological examples? There is a citation for this. In other words the value of the discrete or continuous value doesn’t exert the effect, but the parameter grouping does.*

We demonstrate a potentially serious problem when fitting OU models without allowing for heterogeneity in the evolution process. In cases where discrete and continuous characters are unlinked, but there is heterogeneity in the dynamics continuous character, we find high false-positive support for character dependent models. Although hOUwie is designed to mitigate these concerns, due to the caveats previously discussed it is far from a complete solution. The joint estimation procedure implemented in hOUwie has high parameter estimation error when focal discrete characters are unlinked to the continuous phenotype. There are several avenues to improve the hOUwie model that are actively being explored. Statistical work on Markov-Modulated Ornstein-Uhlenbeck models represent an intriguing possibility but have yet to be applied in phylogenetic comparative biology (Huang et al. 2016). However, if implemented in a phylogenetic context, it would remove the need for a simulation approach. Finally, it is possible to extend hOUwie to include state-dependent speciation and extinction dynamics. This extension would require a different calculation of the underlying regime mapping probability but would be relatively straightforward. A challenging aspect of this extension would be generating high joint probability mappings, although approaches for stochastically mapping SSE models exist (Freyman and Höhna 2019).

**Conclusion**

The use of pre-defined discrete character mappings can be useful for testing hypotheses which rely on distinct, well-defined differences in the evolutionary histories of lineages. However, this approach assumes that the underlying mapping is known with complete accuracy and ignores the probabilistic nature of discrete regimes. hOUwie’s methodology integrates over the uncertainty of high probability character mappings and relies on the interpretation of parameter estimates from contrasting model structures to find evidence for hypotheses. Our simulations have shown how hidden states modeling heterogeneity in the evolutionary process can lead to false character dependent signals. Although we have shown that hOUwie can theoretically resolves this issue, in practice, additional work to improve the computational costs of integrating over all possible stochastic mappings is needed. Nonetheless, the implementation of a joint discrete and continuous model in a likelihood framework improves our inferences about the parameters related to the underlying regimes. Rather than assuming an apriori mapping, hOUwie is able to utilize the mutual information about the discrete and continuous characters to learn something about the underlying regimes evolution.

**Tables**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| model\_type | sampling\_procedure | disc | cont | joint |
| CID+\_BMV | adaptive\_sampling | -15.17 | 22.5 | 7.33 |
| CID+\_BMV | discrete\_only | -17.52 | 13.38 | -4.13 |
| CID+\_OUA | adaptive\_sampling | -16.45 | 45.85 | 29.4 |
| CID+\_OUA | discrete\_only | -19.63 | 39.72 | 20.09 |
| CID+\_OUV | adaptive\_sampling | -26.6 | 45.44 | 18.84 |
| CID+\_OUV | discrete\_only | -29.85 | 37.16 | 7.31 |
| CID+\_OUVA | adaptive\_sampling | -13.22 | 51.52 | 38.3 |
| CID+\_OUVA | discrete\_only | -16.33 | 35.51 | 19.18 |
| CID+\_OUM | adaptive\_sampling | -42.09 | 45.97 | 3.88 |
| CID+\_OUM | discrete\_only | -37.57 | -61.17 | -98.74 |
| CID+\_OUMA | adaptive\_sampling | -18.59 | 44.11 | 25.52 |
| CID+\_OUMA | discrete\_only | -16.65 | 1.01 | -15.64 |
| CID+\_OUMV | adaptive\_sampling | -30.56 | 45.13 | 14.57 |
| CID+\_OUMV | discrete\_only | -21.11 | -9.42 | -30.53 |
| CID+\_OUMVA | adaptive\_sampling | -28.52 | 37.72 | 9.2 |
| CID+\_OUMVA | discrete\_only | -26.37 | 14.25 | -12.12 |
| CID+\_OUBM1 | adaptive\_sampling | -19.1 | 5.59 | -13.51 |
| CID+\_OUBM1 | discrete\_only | -19.8 | -59.5 | -79.31 |
| CID+\_OUBMV | adaptive\_sampling | -26.96 | 18.36 | -8.6 |
| CID+\_OUBMV | discrete\_only | -28.45 | -32.44 | -60.89 |

**Table 1:** Discrete, continuous, and joint log probabilities for 100 stochastic maps. Maps are generated based on parameters which were used to generate the dataset being fit.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| model\_type | model\_class | rate | alpha\_1 | alpha\_2 | sigma.sq\_1 | sigma.sq\_2 | theta\_1 | theta\_2 |
| BMV | CD | 0.11 | NA | NA | 0.09 | 0.27 | 0.21 | NA |
| OUV | CD | 0.1 | 1.17 | NA | 0.15 | 0.3 | 0.05 | NA |
| OUA | CD | 0.11 | 1.64 | 1.77 | 0.12 | NA | 0.06 | NA |
| OUM | CD | 0.11 | 1.33 | NA | 0.09 | NA | 0.07 | 0.12 |
| OUVA | CD | 0.09 | 1.41 | 1.1 | 0.12 | 0.96 | 0.06 | NA |
| OUMV | CD | 0.13 | 1.72 | NA | 0.14 | 0.31 | 0.06 | 0.17 |
| OUMA | CD | 0.12 | 2.28 | 2.66 | 0.32 | NA | 0.12 | 0.51 |
| OUMVA | CD | 0.12 | 1.58 | 1.04 | 0.1 | 1.17 | 0.86 | 1.13 |
| OUBM1 | CD | 0.09 | 2.71 | NA | 0.08 | NA | 0.07 | NA |
| OUBMV | CD | 0.08 | 2.26 | NA | 0.13 | 1.72 | 0.07 | NA |
| BMV | CID | 0.04 | NA | NA | 0.27 | 11.03 | 0.23 | NA |
| OUV | CID | 0.04 | 1.03 | NA | 0.32 | 2.01 | 0.05 | NA |
| OUA | CID | 0.04 | 3.26 | 1.29 | 0.39 | NA | 0.07 | NA |
| OUM | CID | 0.07 | 2.35 | NA | 0.14 | NA | 0.49 | 0.19 |
| OUVA | CID | 0.05 | 1.19 | 1.15 | 0.28 | 16.72 | 0.07 | NA |
| OUMV | CID | 0.08 | 2.46 | NA | 0.15 | 2.4 | 1.49 | 0.75 |
| OUMA | CID | 0.04 | 10.28 | 1.26 | 0.26 | NA | 7.11 | 0.82 |
| OUMVA | CID | 0.06 | 6.3 | 1.26 | 0.2 | 10.33 | 10.12 | 1.47 |
| OUBM1 | CID | 0.05 | 3.61 | NA | 0.37 | NA | 0.13 | NA |
| OUBMV | CID | 0.04 | 3.78 | NA | 0.29 | 10.63 | 0.14 | NA |

**Table 2:** Average RMSE per model type and class.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| model\_type | model\_class | AICwt.BM1 | AICwt.OU1 | AICwt.CD | AICwt.CID+ | best\_model |
| BMV | CD | 0.15 | 0.15 | 0.69 | 0.01 | 0.67 |
| OUV | CD | 0.02 | 0.2 | 0.77 | 0.01 | 0.76 |
| OUA | CD | 0.06 | 0.55 | 0.32 | 0.07 | 0.17 |
| OUM | CD | 0.03 | 0.02 | 0.91 | 0.04 | 0.92 |
| OUVA | CD | 0.03 | 0.14 | 0.76 | 0.07 | 0.77 |
| OUMV | CD | 0.01 | 0.01 | 0.96 | 0.02 | 0.97 |
| OUMA | CD | 0.1 | 0.08 | 0.71 | 0.11 | 0.73 |
| OUMVA | CD | 0.04 | 0.08 | 0.81 | 0.07 | 0.81 |
| OUBM1 | CD | 0.18 | 0.57 | 0.13 | 0.12 | 0.08 |
| OUBMV | CD | 0.05 | 0.15 | 0.78 | 0.02 | 0.8 |
| BMV | CID | 0.36 | 0.27 | 0.35 | 0.03 | 0.01 |
| OUV | CID | 0.03 | 0.49 | 0.44 | 0.04 | 0.01 |
| OUA | CID | 0.05 | 0.55 | 0.39 | 0.02 | 0 |
| OUM | CID | 0.19 | 0.08 | 0.03 | 0.69 | 0.73 |
| OUVA | CID | 0.06 | 0.51 | 0.39 | 0.04 | 0.03 |
| OUMV | CID | 0.23 | 0.18 | 0.14 | 0.45 | 0.46 |
| OUMA | CID | 0.41 | 0.39 | 0.15 | 0.05 | 0.05 |
| OUMVA | CID | 0.22 | 0.38 | 0.23 | 0.16 | 0.16 |
| OUBM1 | CID | 0.23 | 0.55 | 0.18 | 0.04 | 0.01 |
| OUBMV | CID | 0.22 | 0.33 | 0.34 | 0.11 | 0.1 |

**Table 3**: Average AIC weights summarized over 150 simulated datasets

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| nTaxa | model\_type | AICwt.BM1 | AICwt.OU1 | AICwt.CD | AICwt.CID+ |
| 25 | CD | 0.12 | 0.22 | *0.51* | 0.15 |
| 100 | CD | 0.06 | 0.22 | *0.7* | 0.02 |
| 250 | CD | 0.02 | 0.14 | *0.82* | 0.02 |
| 25 | CID | 0.28 | 0.35 | 0.24 | *0.14* |
| 100 | CID | 0.21 | 0.4 | 0.23 | *0.15* |
| 250 | CID | 0.11 | 0.34 | 0.32 | *0.22* |

**Table 4:** Average AIC weight depending on the number of taxa. Italics highlights the generating model support.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Model | np | lnLik | DiscLik | ContLik | AIC | dAIC | AICwt |
| FitSD=CD\_BMV | 5 | -243.78 | -32.62 | -207.08 | 497.56 | 40.85 | 0 |
| FitSD=CD\_OUA | 6 | -224.95 | -32.58 | -189.48 | 461.9 | 5.19 | 0.03 |
| FitSD=CD\_OUBM1 | 5 | -243.84 | -32.57 | -206.67 | 497.68 | 40.97 | 0 |
| FitSD=CD\_OUBMV | 6 | -243.79 | -32.61 | -206.99 | 499.57 | 42.87 | 0 |
| FitSD=CD\_OUM | 6 | -224.12 | -32.57 | -187.79 | 460.24 | 3.53 | 0.07 |
| FitSD=CD\_OUMA | 7 | -223.21 | -32.58 | -187.97 | 460.42 | 3.71 | 0.06 |
| FitSD=CD\_OUMV | 7 | -224.05 | -32.62 | -188.15 | 462.1 | 5.39 | 0.03 |
| *FitSD=CD\_OUMVA* | ***8*** | ***-220.35*** | ***-32.6*** | ***-183.27*** | ***456.71*** | ***0*** | ***0.41*** |
| FitSD=CD\_OUV | 6 | -225.49 | -32.62 | -188.47 | 462.98 | 6.27 | 0.02 |
| FitSD=CD\_OUVA | 7 | -221.62 | -32.58 | -184.44 | 457.24 | 0.53 | 0.32 |
| FitSD=CID\_BM1 | 4 | -243.89 | -32.62 | -206.67 | 495.78 | 39.07 | 0 |
| FitSD=CID\_OU1 | 5 | -225.5 | -32.62 | -188.28 | 461.01 | 4.3 | 0.05 |
| FitSD=CID+\_BMV | 7 | -244.8 | -33.11 | -205.78 | 503.59 | 46.89 | 0 |
| FitSD=CID+\_OUA | 8 | -226.42 | -33.17 | -188.53 | 468.84 | 12.13 | 0 |
| FitSD=CID+\_OUBM1 | 7 | -244.44 | -33.16 | -206.67 | 502.88 | 46.17 | 0 |
| FitSD=CID+\_OUBMV | 8 | -225.58 | -32.71 | -186.58 | 467.17 | 10.46 | 0 |
| FitSD=CID+\_OUM | 8 | -226.43 | -33.32 | -189.07 | 468.87 | 12.16 | 0 |
| FitSD=CID+\_OUMA | 9 | -225.57 | -32.68 | -189.92 | 469.14 | 12.43 | 0 |
| FitSD=CID+\_OUMV | 9 | -225.2 | -33.39 | -182.88 | 468.39 | 11.68 | 0 |
| FitSD=CID+\_OUMVA | 10 | -227.39 | -33.13 | -185.15 | 474.79 | 18.08 | 0 |
| FitSD=CID+\_OUV | 8 | -228.77 | -32.98 | -190.16 | 473.55 | 16.84 | 0 |
| FitSD=CID+\_OUVA | 9 | -244.38 | -33.43 | -202.12 | 506.76 | 50.05 | 0 |
| FitSD=HYB\_BMS | 9 | -244.46 | -33.08 | -204.83 | 506.93 | 50.22 | 0 |
| FitSD=HYB\_OUM | 10 | -224.12 | -32.67 | -188.99 | 468.23 | 11.52 | 0 |
| FitSD=HYB\_OUMVA | 16 | -226.56 | -33.03 | -179.11 | 485.13 | 28.42 | 0 |

**Table 5:** Empirical model fitting results.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Continuous** | **Alpha** | **Sigma.sq** | **Theta** | **Discrete** | **To dry** | **To fleshy** |
| **Dry** | **0.015** | **0.011** | **0.97** | **From dry** | **-** | **0.0015** |
| **Fleshy** | **0.023** | **0.007** | **0.81** | **From fleshy** | **00036** | **-** |

**Table 6:** Model averaged parameter estimates.

**Figures**

A picture containing diagram

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Figure 1: A state-transition diagram describing the examined hOUwie model structures.

Calendar

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Figure 2: Forward simulations of some of the different possible model types in hOUwie.

Diagram

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Figure 3: Adaptive sampling procedure.



Figure 4: For a single simulation, the performance of adaptive sampling (blue) compared to pure discrete stochastic mappings (red). Dashed line likelihood under generating map.

Chart, box and whisker chart

Description automatically generated

**Figure 5:** Parameter estimation performance. Raw differences from generating parameters.

Timeline, box and whisker chart

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**Figure 6:** Model AIC weights by simulating model.

A picture containing diagram

Description automatically generated

**Figure 7:** Ericaceae dataset and phylogeny.

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