

Ecological Opportunity from Innovation, not Islands, Drove the Anole Lizard

Adaptive Radiation

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Supplementary Figures and Tables

MORPHOLOGICAL DISPARITY BETWEEN ISLAND AND MAINLAND ANOLES

Morphological data were available for more species than were present in available phylogenies that included anoles, resulting in sampling scaling with different phylogenetic hypotheses. To ensure similar morphological relationships between island and mainland species, we compared their morphological diversity of across three trait datasets scaled to match the tips in three different phylogenetic trees: Mahler et al. (2010), Zheng and Wiens (2016), and Poe et al. (2017). For the Mahler et al. (2010) and Zheng and Wiens (2016), traits were ln-transformed and regressed against ln-svl using the `phyl.resid` function implemented in `phytools` (Revell 2012). For the Poe et al. (2017) phylogenetic tree in which some species were missing sequences and were placed using morphology, we corrected for size using non-phylogenetic residuals using a simple linear model. We then calculated morphological disparity using the disparity function implemented with the R package `geiger` (Harmon et al. 2008). Island anoles exhibit greater morphological disparity than mainland anoles, regardless of sampling, with 1.72-fold (Mahler et al. 2010), 1.55-fold (Zheng and Wiens 2016), and 1.61-fold (Poe et al. 2017) differences in disparity.

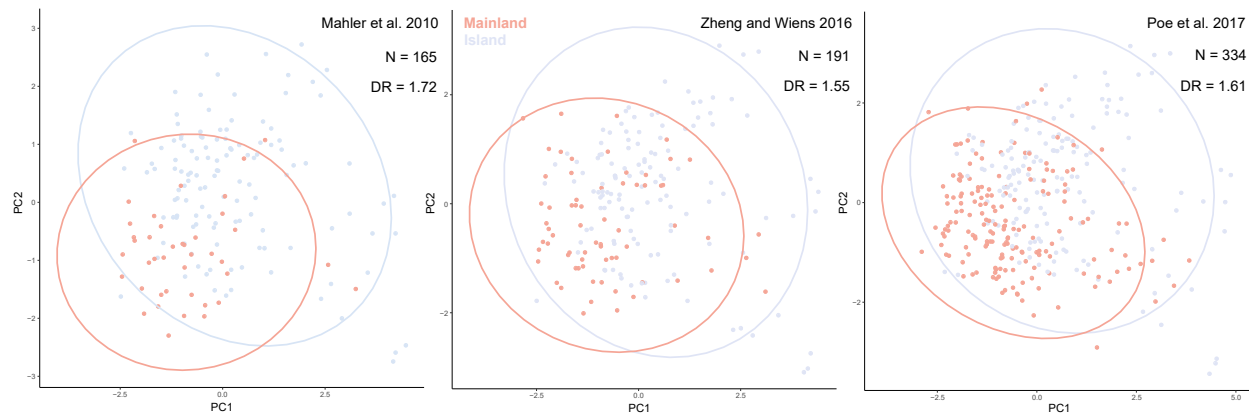


Figure S1. Morphological disparity between island and mainland anoles across three different sampling schemes that match different phylogenetic trees. PCA is only meant for visualization. Note that disparity was calculated using size-corrected trait values, not PC score values.

RELEVANCE OF MORPHOLOGICAL TRAITS

We wanted to verify that the morphological traits included in our multivariate analyses were relevant for the anole adaptive radiation, specifically for microhabitat specialization that characterizes the Caribbean island assemblages. Therefore, we compared each trait among ecomorphs using phylogenetic ANOVA with a residual randomization permutation procedure (Collyer and Adams 2018) implemented in the geomorph R package (Adams and Otárola-Castillo 2013). Statistical significance was determined using 10k permutations. These analyses revealed that snout-vent length, femur length, toe length, and tail length varied among the ecomorphs (Table S1; Fig. S2). In particular, crown-giants had large snout-vent lengths, whereas trunk and twig ecomorphs were small bodied. Twig species are characterized by short femora, toes, and tails. Species in the grass-bush ecomorph exhibit long tails. Collectively, the six traits separated the ecomorphs (based on PCA; Fig. S2), with crown-giants and twig ecomorphs being especially morphologically distinct.

Table S1. Comparison of morphological traits among the classic anole Caribbean island ecomorphs using phylogenetic ANOVA.

Trait	F	P
Snout-vent length	15.39	<0.0001
Femur length	12.53	<0.0001
Head length	1.38	0.230
Toe length	9.60	<0.0001
Tail length	12.12	<0.0001
Number of lamellae	1.24	0.293

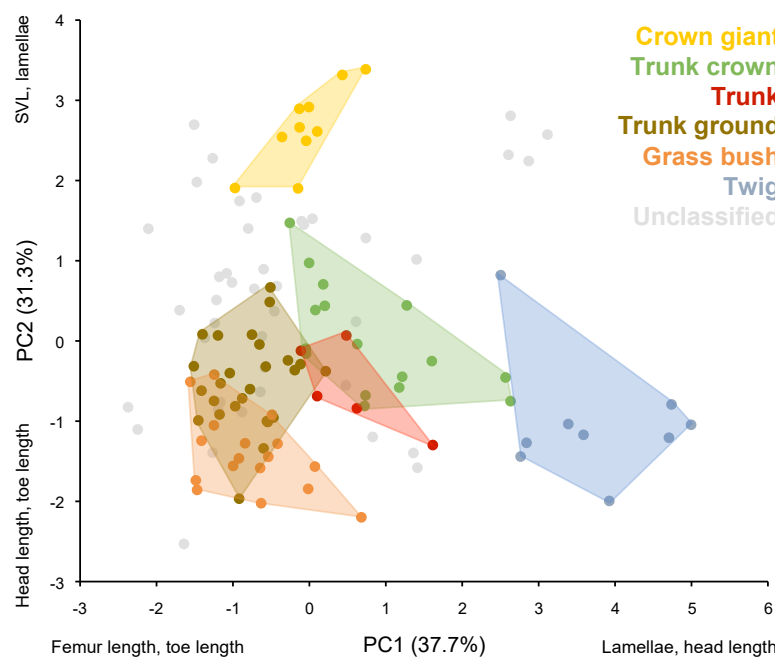


Figure S2. Morphological relationships among the classic Caribbean island ecomorphs. PCA meant for visualization, see Table S1 for quantitative comparisons of ecomorphs.

ROBUSTNESS OF USER SPECIFIED PRIORS

The Multiple State-Specific Rates of continuous character evolution model (MuSSCRat; May and Moore 2020) uses a prior on the number of expected rate shifts. We repeated the Markov chain Monte Carlo (MCMC) using different priors on the number of rate shifts to assess its impact on posterior estimates of key parameters. For the models comparing evolutionary rates between island and mainland anoles, the prior number of rate shifts (10, 20, 30, and 40 shifts), had no effect on the number of state changes (of the discrete character; Fig. S3) or the posterior probability that the evolutionary rates were state-dependent (PP = 0.101, 0.154, 0.133, and 0.145, respectively). The prior on the expected number of rate shifts strongly predicted the posterior estimate of the number of rate shifts (Fig. S3), as expected (Moore et al. 2016; May and Moore 2020). All MCMCs used in the main text of the study or in the Supplementary materials had effective sample sizes (ESS) above 400.

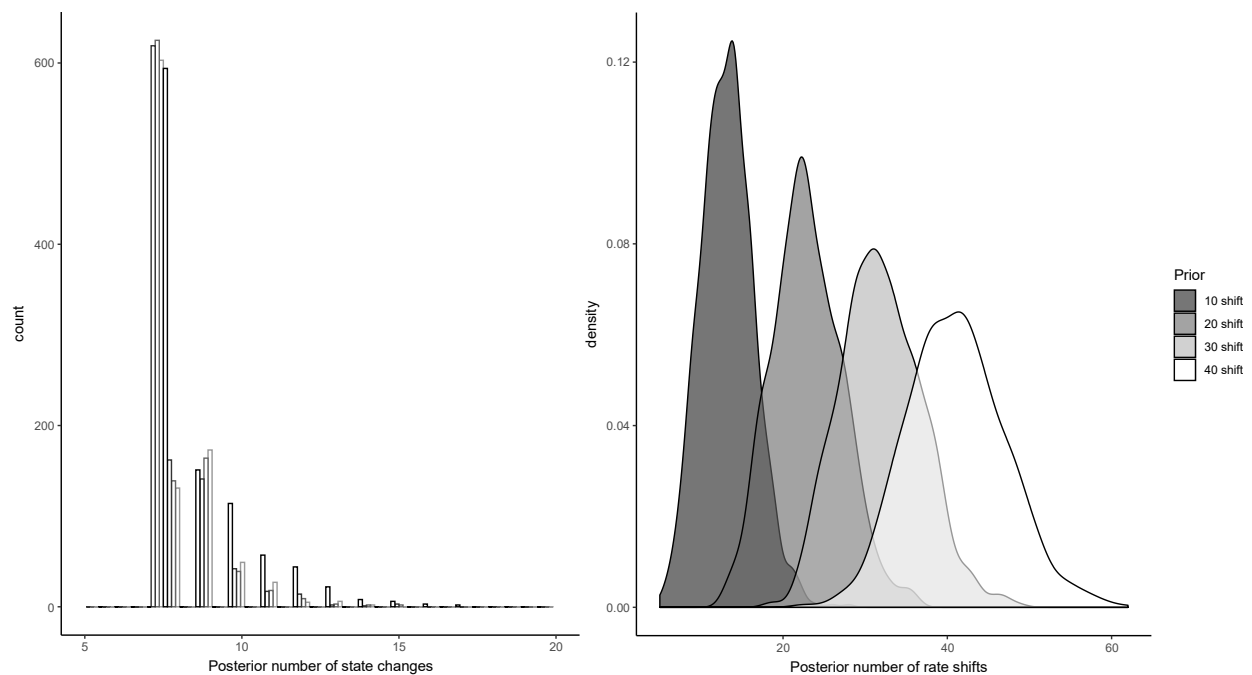


Figure S3. Assessment of the impact of the prior number of rate shifts on posterior estimates of transitions among discrete character states (left panel) and number of rate shifts (right panel) estimated by the MuSSCRat model comparing island and mainland anoles.

For the models comparing evolutionary rates between speciation modes, the prior number of rate shifts (10, 20, 30, and 40 shifts), had no effect on the number of state changes (of the discrete character; Fig. S4) or the posterior probability that the evolutionary rates were state-dependent (PP = 0.255, 0.261, 0.229, and 0.175, respectively). The prior on the expected number of rate shifts strongly predicted the posterior estimate of the number of rate shifts (Fig. S4), as expected (Moore et al. 2016; May and Moore 2020).

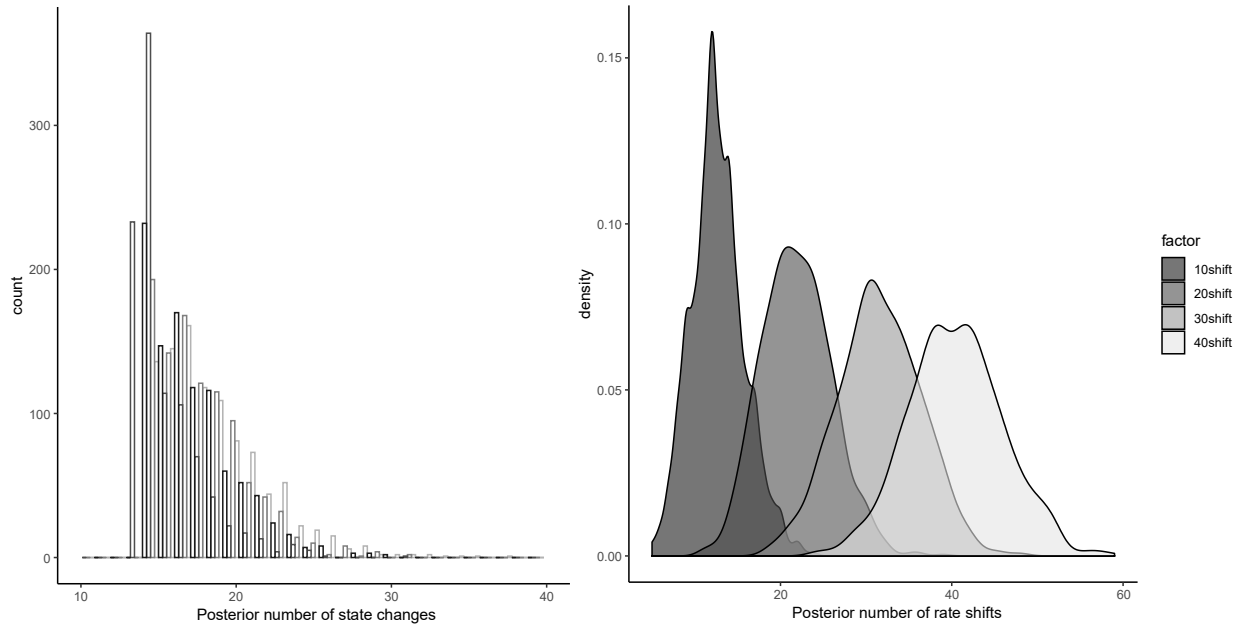


Figure S4. Assessment of the impact of the prior number of rate shifts on posterior estimates of transitions among discrete character states (left panel) and number of rate shifts (right panel) estimated by the MuSSCRat model comparing speciation modes.

For the models comparing evolutionary rates among Caribbean islands, the prior number of rate shifts (10, 20, 30, and 40 shifts), had no effect on the number of state changes (of the discrete character; Fig. S5) or the posterior probability that the evolutionary rates were state-dependent (PP = 0.179, 0.174, 0.344, and 0.140, respectively). The prior on the expected number of rate shifts strongly predicted the posterior estimate of the number of rate shifts (Fig. S5), as expected (Moore et al. 2016; May and Moore 2020).

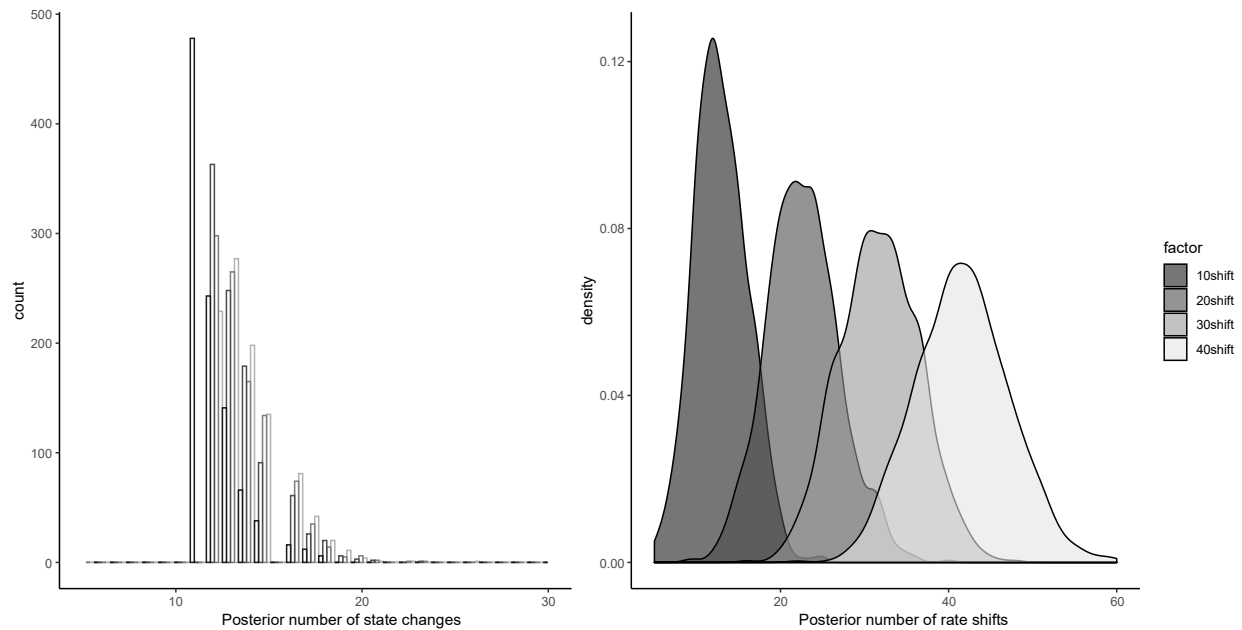


Figure S5. Assessment of the impact of the prior number of rate shifts on posterior estimates of transitions among discrete character states (left panel) and number of rate shifts (right panel) estimated by the MuSSCRat model comparing specific Caribbean islands.

ROBUSTNESS TO SAMPLING, PHYLOGENETIC TREE, AND MODEL

In the main text, we performed all phylogenetic comparative analyses across the Mahler et al. (2010) phylogenetic tree, which included 165 species (124 island, 41 mainland) that matched the morphological dataset. We wanted to evaluate the robustness of our main result to sampling and choice of phylogenetic tree. Therefore, we repeated the core analyses using the Zheng and Wiens (2016) phylogenetic tree, which included 191 (124 island, 67 mainland) species that matched the morphological dataset, differing from the Mahler et al. (2010) tree primarily in the number of mainland species. As in the main text, there was no difference in observed speciation rates between island and mainland species (Fig. S6), which we estimated using Hidden State-Dependent Speciation and Extinction models (HiSSE; Beaulieu and O'Meara 2016) implemented in RevBayes (Höhna et al. 2016) as described in the main text. Likewise, there was no “island effect” on rates of morphological evolution (Fig. S6), estimated with Multiple State-Specific Rates of continuous character evolution (MuSSCRat; May and Moore 2020) as described in the main text. These results were consistent across models using different priors on the expected number of rate shifts (Fig. S6).

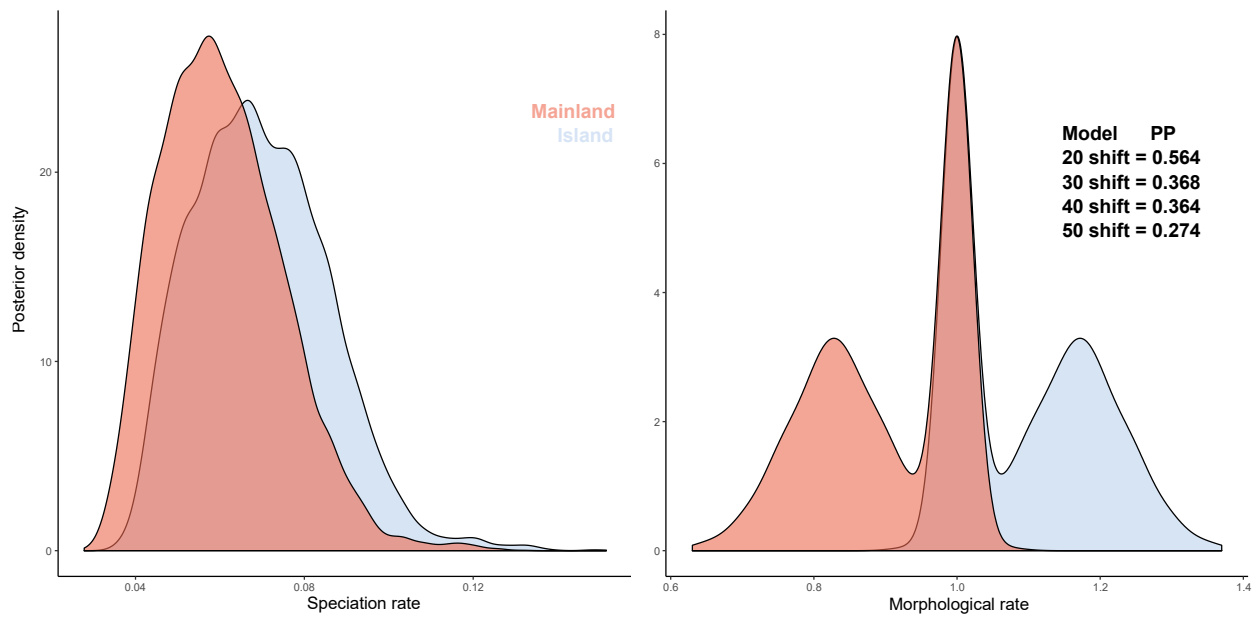


Figure S6. State dependent rates of speciation (left panel) and morphological evolution (right panel) if the analyses in the main text were repeated with a larger dataset that matches the Zheng and Wiens (2016) phylogenetic tree.

We also wanted to test the robustness of the correlation between rates of speciation and morphological evolution across anoles; therefore, we repeated these analyses using the Zheng and Wiens (2016) phylogenetic tree. In the main text, we found that these rates were correlated ($r=0.199$; $t=7.69$; $p<0.0001$; Fig. 2) and that there was no “island effect” on the slope of the line fit to this relationship, as a single slope model was preferred over a model that permitted island and mainland anoles to have their own slopes (based on AIC; Fig. 2). Using the Zheng and Wiens (2016) phylogenetic tree, rates of speciation and morphological evolution were also correlated ($r=0.266$; $t=2.38$; $p=0.018$) and there was no “island effect” on the slope of the line fit to this relationship (based on AIC; Fig. S7).

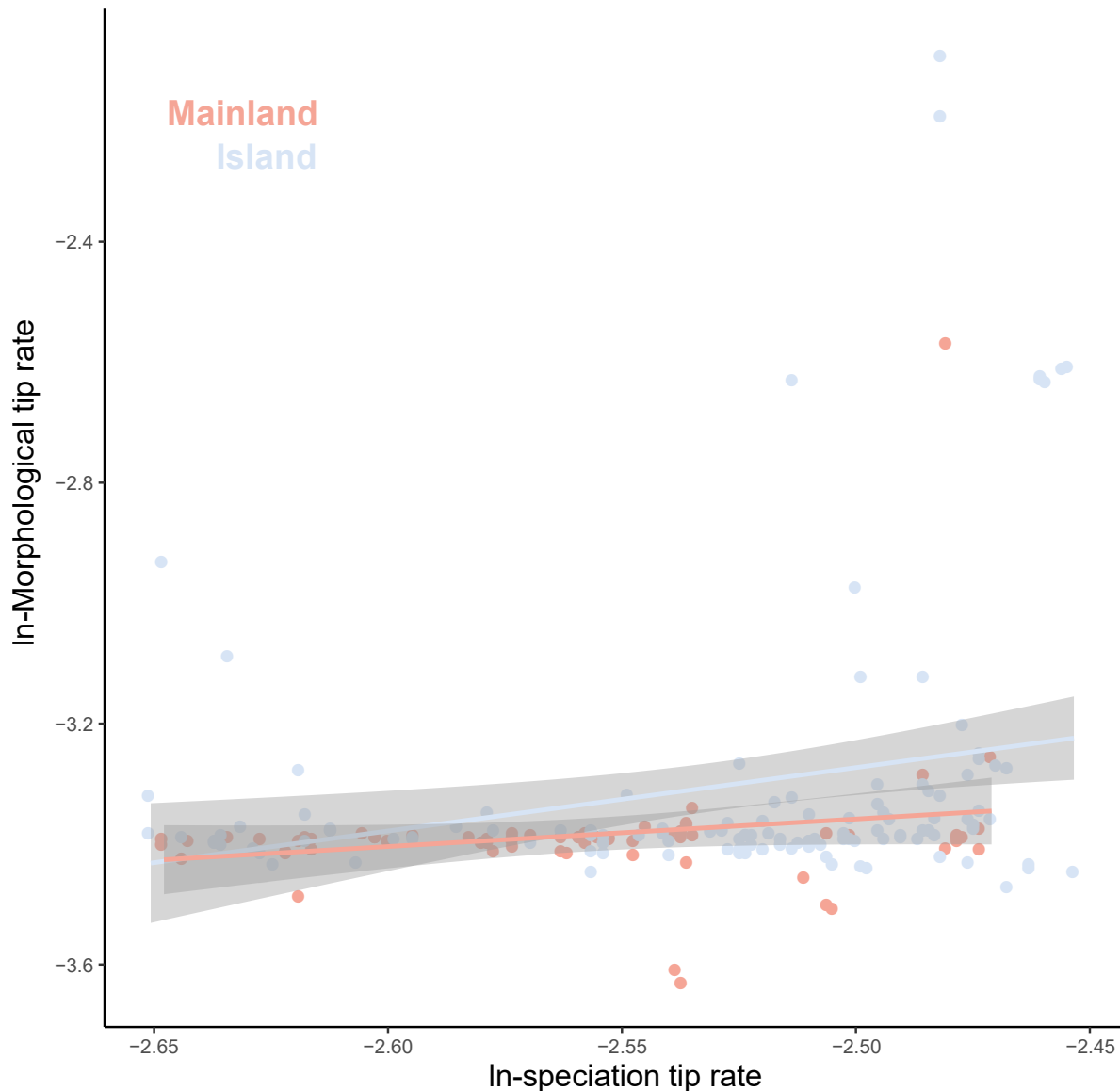


Figure S7. Relationship between rates of speciation and morphological evolution among anoles if the analyses in the main text were repeated using a larger dataset that matches the tips of the Zheng and Wiens (2016) phylogenetic tree. Best-fit lines correspond to island and mainland species to depict their similar slopes.

Additionally, we repeated the MuSSCRat analyses using an uncorrelated lognormal (UCLN) relaxed-clock model in which the rates have no phylogenetic structure, as opposed to a model in which rates are inherited with a probability of a rate shift (*i.e.*, random local clock; May and Moore 2020). Using this alternative model, we found the same results reported in the main text – rates of morphological evolution did not vary between mainland and island anoles (PP=0.200), according to speciation mode (PP=0.185), or among the four major Caribbean islands (PP=0.014; Figure S8).

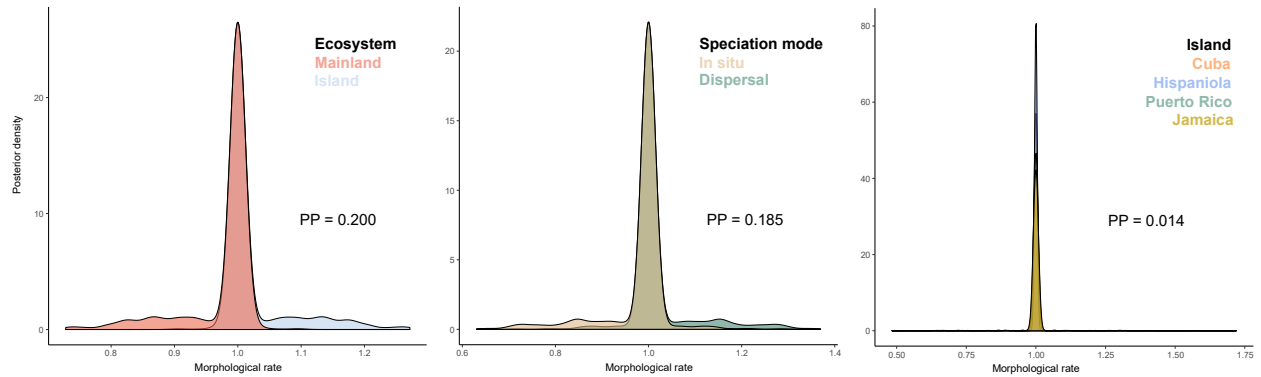


Figure S8. Rates of morphological evolution estimated with MuSSCRat plus an uncorrelated lognormal (UCLN) model.

SENSITIVITY TO BACKGROUND RATE VARIATION

Lastly, we evaluated the importance of accommodating background rate variation in avoiding type I error. To achieve this, we performed a reduced MuSSCRat analyses in which the Brownian motion model was not relaxed to permit rates to vary among branches (May and Moore 2020); thereby attributing all rate variation to the discrete character (and correspondingly using a single rate model as the null model). We found that such a model led to false positives, including 2.37-fold faster rates of morphological evolution on islands as on the mainland (PP=1.0; Figure S9a) and 1.85-fold faster rates of morphological evolution in anoles that dispersed to their host island as opposed to evolving *in situ* (PP=0.978; Figure S9b). There were still no differences in rates among the major Caribbean islands (PP=0.0289).

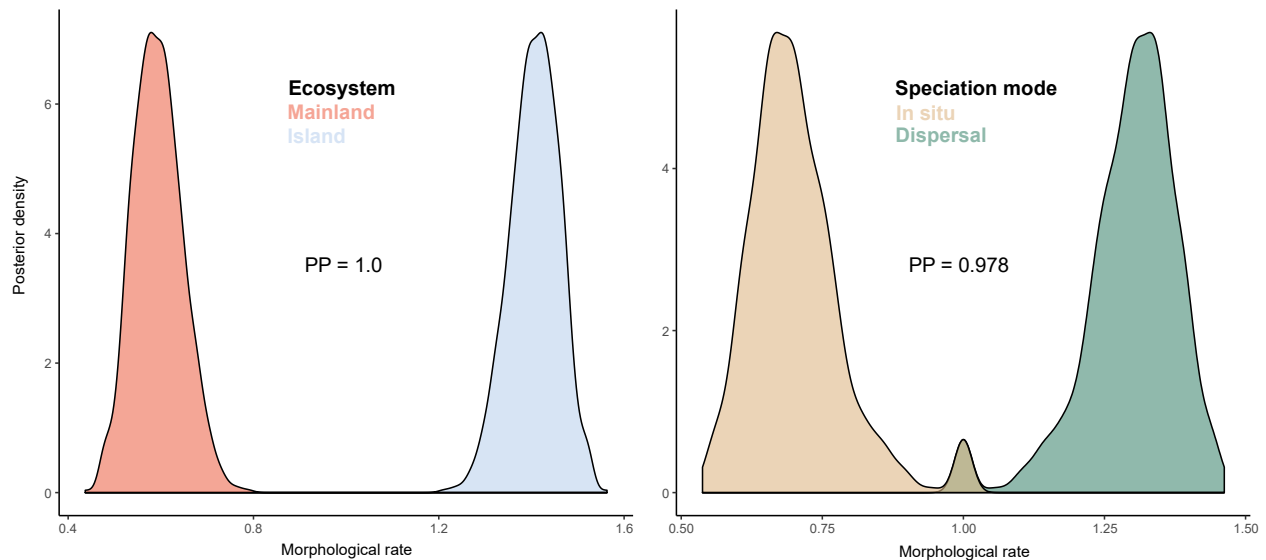


Figure S9. Rates of morphological evolution estimated with a reduced MuSSCRat model in which rates were not allowed to vary among branches (i.e., no background rate heterogeneity), which led to false positives for the effect of ecosystem and speciation mode.

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