ORIGINAL ARTICLE

ELEVATED RATES OF MORPHOLOGICAL AND FUNCTIONAL DIVERSIFICATION IN REEF-DWELLING HAEMULID FISHES

Samantha A. Price,[1,2,3] Jose J. Tavera,[3,4] Thomas J. Near,[5] and Peter. C. Wainwright[1]

[1]Department of Evolution and Ecology, University of California Davis, Davis, California 95616

[2]E-mail: saprice@ucdavis.edu

[4]Centro de Investigaciones Biológicas del Noroeste, S.C., La Paz, B.C.S., México

[5]Department of Ecology & Evolutionary Biology, Yale University, New Haven, Connecticut 06520

The relationship between habitat complexity and species richness is well established but comparatively little is known about the evolution of morphological diversity in complex habitats. Reefs are structurally complex, highly productive shallow-water marine ecosystems found in tropical (coral reefs) and temperate zones (rocky reefs) that harbor exceptional levels of biodiversity. We investigated whether reef habitats promote the evolution of morphological diversity in the feeding and locomotion systems of grunts (Haemulidae), a group of predominantly nocturnal fishes that live on both temperate and tropical reefs. Using phylogenetic comparative methods and statistical analyses that take into account uncertainty in phylogeny and the evolutionary history of reef living, we demonstrate that rates of morphological evolution are faster in reef-dwelling haemulids. The magnitude of this effect depends on the type of trait; on average, traits involved in the functional systems for prey capture and processing evolve twice as fast on reefs as locomotor traits. This result, along with the observation that haemulids do not exploit unique feeding niches on reefs, suggests that fine-scale trophic niche partitioning and character displacement may be driving higher rates of morphological evolution. Whatever the cause, there is growing evidence that reef habitats stimulate morphological and functional diversification in teleost fishes.

KEY WORDS: Disparity, functional diversity, Haemulidae, phylogenetic comparative methods, reef.

The relationship between habitat complexity and species richness is well established; as habitat complexity increases so does the number of species that live in the habitat the area (e.g., MacArthur and MacArthur 1961; Rosenzweig 1995). Physically and biologically complex habitats create microenvironmental heterogeneity and resource patchiness thereby providing ideal conditions for niche partitioning and specialization, which in turn may raise the number of species that can stably coexist (MacArthur and Levins 1964; Schoener 1974). In the marine ecosystem hard-bottomed reefs, which include coral reefs in the tropics and rocky reefs

with kelp forests in temperate zones, are the most structurally complex, highly productive ecosystems (Fraser and Currie 1996; Steneck et al. 2002; Monismith 2006). Hard-substratum environments are topographically more complex (Gratwicke and Speight 2005) and are frequently dominated by active primary producers (corals and macroalgae) that provide additional structural complexity. Substrate complexity is positively associated with fish density in Alaskan kelp beds (Hamilton and Konar 2007) as well as fish abundance and diversity on Kenyan coral reefs (McClanahan 1994) and tropical habitats in the British Virgin Isles (Gratwicke and Speight 2005). Species diversity is particularly high in reef ecosystems (Renema et al. 2008). Coral reefs

[3]These two authors contributed equally to this manuscript.

are the most biologically diverse shallow water marine ecosystem on earth (Roberts et al. 2002) and harbor the highest species richness of fishes (Harmelin-Vivien 2002). Rocky reefs are also biologically diverse, supporting a wide variety of plant and animal species (Levinton 1995), especially in comparison to many other temperate marine ecosystems.

Biologically and structurally complex habitats are also likely to promote in situ diversification of both lineages and phenotypes. A recent paleontological study found that throughout the Phanerozoic, biogenic reefs—where the reef structure is created by the animals themselves—have acted as cradles of evolution exporting diversity to other marine habitats (Kiessling et al. 2010). Similarly, large-scale phylogenetic comparative studies have shown that coral reefs elevate the accumulation of lineage diversity in several major groups of coral reef fishes, including pufferfishes and their relatives, butterflyfishes, damselfishes, and cardinalfishes (Alfaro et al. 2007; Bellwood et al. 2010; Cowman and Bellwood 2011). Furthermore, the rate of ecomorphological diversification in wrasses and parrotfishes has been shown to be higher in lineages that live on reefs (Price et al. 2011). Speciation and diversification of functional morphology on reefs may be driven by the same factors that allow the stable coexistence of many species on reefs, as there are likely to be more ecological opportunities when the potential for resource partitioning and specialization is high. In particular, the variety and abundance of food items available on reefs is likely to provide many ecological opportunities for heterotrophs through prey specialization. Indeed, within parrotfishes and wrasses living on coral reefs the high rates of phenotypic diversification are at least partially driven by the occupation of novel regions of morphospace associated with the evolution of unique feeding strategies (Price et al. 2011).

In this study, we test whether complex reef habitats promote functional morphological diversification in trophic, body shape, and locomotor traits using a clade of teleost fishes known as grunts (Haemulidae). Grunts are a valuable counterpoint to labrid fishes, which have previously been shown to exhibit elevated rates of morphological diversification in trophic traits on coral reefs (Price et al. 2011) as they are phylogenetically and ecologically very different. Labrids exhibit extensive species, morphological and ecological diversity including a wide variety of different diets (Randall 1967; Wainwright et al. 2004). Haemulids are far less species rich on reefs (Bellwood and Wainwright 2002), and mainly move off-reefs at night to feed on infaunal invertebrates, hard-shelled prey, and zooplankton, showing less trophic diversity than labrids. It is therefore possible that higher rates of diversification in trophic morphology in reef-dwelling labrids may be related to their exceptional dietary diversity. However, if reef-dwelling haemulids also show higher rates of evolution in feeding and locomotion structures than haemulids living in soft-bottom habitats, this may imply that complex reef habitats are generally

more evolutionarily dynamic and an important source of functional diversity for adaptation in the future.

We use phylogenetic comparative methods to estimate the rate of morphological evolution in reef and nonreef species, which under a Brownian motion model can be viewed as an estimate of disparity that takes into account the effect of time and phylogeny (Hutcheon and Garland 2004; O'Meara et al. 2006). Our approach takes into account uncertainty in the phylogeny (topology and branch lengths) by sampling from the Bayesian posterior distribution of trees (from Tavera et al. 2012), in the history of reef-dwelling by using stochastic character mapping (Nielsen 2002; Huelsenbeck et al. 2003) and finally in model choice by using model averaging (Burnham and Anderson 2002). To assess the robustness and broad applicability of the conclusions, we include haemulid species from both tropical and temperate reefs together with nonreef species from a wider variety of habitats, including freshwater and brackish ecosystems. In addition, we incorporate functionally important postcranial shape and locomotor traits as well as trophic morphology.

Methods
MORPHOLOGICAL DATA

Using 127 specimens from 50 haemulid species, we measured 24 functional morphological traits related to feeding (11 traits) and locomotion (13 traits). We focused on trophic traits that have previously been found to underlie differences between fish species in components of feeding performance (Wainwright and Richard 1995; Schmitz and Wainwright 2011; Holzman et al. 2012). These feeding traits included the mass of the jaw-closing adductor mandibulae muscle (AM mass), the mechanical advantage of jaw closing (close ratio) and opening (open ratio), the length of the ascending process of the premaxilla bone which indicates the capacity for upper jaw protrusion, the length of longest gill raker on the ceratohyal of the first gill arch, and the diameter of the eye. In addition, we calculated suction index, a morphologically based estimate of the capacity to generate suction pressure during prey capture, by combining measures of buccal cavity length, buccal cavity width, head width, head height, and head length (see Carroll et al. 2004; Collar and Wainwright 2006 for further details).

The 13 traits related to the body shape and locomotion included body fineness ratio (Lighthill 1975), which we calculated as the ratio of body length to the square root of maximum body width times maximum body depth; low fineness ratios indicate short-fat shapes and high ratios, long thin shapes. Fineness ratio for fish bodies is thought to be negatively correlated with drag exerted on the body (Bainbridge 1960). The size and shape of propulsive surfaces was measured by caudal fin aspect ratio

(Webb 1984) and length of the base of the spiny and soft dorsal and anal fins, the average spine length of the spiny dorsal fin, and the perimeter and area of the caudal peduncle. In addition, we measured maximum body width, maximum body depth, the body position of maximum body depth expressed as a fraction of fish standard length, and the horizontal and vertical position of the anterior-dorsal pectoral fin base expressed as fractions of standard length and maximum body depth.

To ensure that the magnitude of character change was unrelated to the trait value (larger changes are less likely when trait values are small), we log transformed all linear measurements. Masses were cube root transformed prior to log transformation so that all nonratio traits were on a linear scale. We calculated size-corrected values for all traits across haemulids using the phylogenetic methods outlined by Revell (2009). To assess the relationship between the 24 characters, we calculated a phylogenetically informed correlation matrix. This was done by calculating the standardized independent contrasts (Felsenstein 1985) for each trait on each of the 500 phylogenies sampled from the posterior distribution of trees generated by BEAST and correlating the traits through the origin. The results were summarized as means, minimum, and maximum correlation coefficients (see Supporting Information I). We performed all dataset manipulations and statistics in the R software environment for statistical computing (R Development Core Team 2012) using the ape (Paradis et al. 2004), PHYLOGR (Diaz-Uriarte and Garland 2010), phytools (Revell 2012), and geiger (Harmon et al. 2008) phylogenetic packages.

PHYLOGENY

We used the nucleotide dataset from Tavera et al. (2012), this included 60 species of haemulid and nine out groups; about 85% of the New World haemulid diversity was included in the phylogeny. The nucleotide dataset was 2909 bp long and consisted of partial sequences of three mtDNA genes (*16S* rRNA, *COI*, and *cytb*) and two nuclear genes (*RAG2* and *S7* ribosomal protein intron 1). Further details concerning DNA extraction and sequence alignment can be found in Tavera et al. (2012).

Relative divergence times of the sampled haemulid species were estimated using an uncorrelated lognormal (UCLN) model of molecular evolutionary rate heterogeneity implemented in the computer program BEAST v. 1.6.1 (Drummond et al. 2006; Drummond and Rambaut 2007). Each gene was treated as a separate data partition and for the protein coding genes (*COI*, *cytb*, and *RAG2*), we applied three partitions that corresponded to three codon positions. The posterior density of relative divergence times was estimated using the UCLN model in BEAST along with a birth–death speciation prior for branching rates in the phylogeny. A normally distributed arbitrary age prior with a normal mean of 100.0 and a standard deviation equal to 3.0

was applied to the root node of the phylogeny. These analyses were run three times with each run consisting of 6.0×10^7 generations. The resulting trees and log files from each of the three runs were combined using the computer program Log-Combiner v. 1.5.3 (http://beast.bio.ed.ac.uk/LogCombiner). We assessed convergence of model parameter values and estimated node heights to their optimal posterior distributions by plotting the marginal posterior probabilities versus the generation state in the computer program Tracer v. 1.5 (http://beast.bio.ed.ac.uk/Tracer). The posterior probability density of the combined tree and log files was summarized as a maximum clade credibility tree using TreeAnnotator v. 1.5.3 (http://beast.bio.ed.ac.uk/TreeAnnotator). The mean and 95% highest posterior density estimates of divergence times and the posterior probabilities of inferred clades were visualized using the computer program FigTree v. 1.2.3 (http://beast.bio.ed.ac.uk/FigTree). From the Bayesian posterior distribution generated by BEAST, we randomly sampled 500 trees that we use throughout the article as a way of including uncertainty in tree topology and branch length into our phylogenetic comparative analyses. Ten haemulid species were pruned from these trees to match the morphological dataset.

RECONSTRUCTING REEF LIVING

To compare the rates of morphological evolution in reef and non-reef dwelling species, we first assigned each species to either a "reef" (R) or "nonreef" (NR) habitat. Species qualified as reef fishes only if they are intimately associated with coral or other rocky reefs (i.e., feeding and/or taking refuge on reefs, nonreef species are not found on reefs or the surrounding habitats). For these designations, we relied on studies of haemulid feeding ecology and our own observations (Randall 1967; Cervigón 1993; Allen and Roberts 1994; McKay and Schnider 1995; Thomson et al. 2000; Chirichigno and Cornejo 2001; Lindeman and Toxey 2003; McEachran and Fechelm 2005).

We used stochastic character mapping (Huelsenbeck et al. 2003 and references therein) to sample possible histories of reef living in proportion to their posterior probability, as implemented in the program SIMMAP V1.0 (Bollback 2006). We allowed the branch lengths to directly represent the evolutionary rate by not including a gamma prior on the tree lengths and used an uninformative symmetric Beta prior ($\alpha = 1$ and $\kappa = 19$) on the symmetry of the transition rate matrix (i.e., the extent to which transitions favor one state (0) over the other (1)). We then sampled 500 character histories in proportion to their posterior probability for each of the 500 trees generating 250,000 character maps from which we randomly sampled 10,000 representatives. We then integrated the parameter estimates over these sampled histories and calculated standard errors (SEs) (following Collar et al. 2009). Unlike parsimony or maximum likelihood methods of ancestral state reconstruction, these character maps allow us to incorporate the

uncertainty associated with the tree topology, branch lengths, and timing of the transitions between the reef and nonreef habitats into our rate parameter estimates and SEs.

EVOLUTIONARY RATES

In a phylogenetic context, morphological diversity or disparity is frequently measured as the rate parameter from a Brownian motion model of phenotypic evolution (see Hutcheon and Garland 2004; O'Meara et al. 2006; Thomas et al. 2006). The faster the Brownian rate, the more morphological diversity among lineages is generated per unit of time. We estimated the maximum likelihood Brownian rate parameter for each trait on the time-calibrated phylogeny across the 10,000 trees with stochastic maps using the phytools package (Revell 2012) in the statistical software R, based on O'Meara et al. (2006). In the first model, we fit a single Brownian rate of morphological evolution across the whole tree, representing the same rate of morphological evolution for reef and nonreef associated fishes. In the second model, we fit a two-rate model, allowing species living on reefs to evolve at different rates to those that do not live on reefs.

Our analyses resulted in distributions of model parameter estimates and fit scores for the one- and two-rate models for every trait, these represent variation in the results due to uncertainty in tree topology, branch length, and character history. To integrate over this uncertainty, we calculated the average fit and parameter estimates over the 10,000 trees. The SE of the rate estimate includes two sources of potential error. The first is the SE associated with uncertainty in tree topology and character mapping estimated from the variance in parameter estimate across the 10,000 trees. The second is the approximate SE of the rate estimate as estimated from the Hessian matrix. The curvature of the likelihood surface estimated from the Hessian should perhaps be viewed with caution as it is not robust to model misspecification and may sometimes underestimate the variance as the surface is approximated as quadratic. Finally, to incorporate uncertainty about model choice into the parameter estimate, we calculated model-averaged reef and nonreef rate using the Akaike weights from the mean AICc scores (Akaike Information Criterion with small sample size correction, Hurvich and Tsai 1989). AICc weights describe the proportion of support a model receives in relation to the support for all models (Burnham and Anderson 2002), they are calculated by dividing the relative likelihood of the model by the sum of the relative likelihoods across all models investigated. We calculated SEs on the model-averaged results using the variance, following the methods outlined in Burnham and Anderson (2002).

As many of the characters are best fit by a single-optimum Ornstein–Uhlenbeck (OU) model (see Supporting Information II), young clades analyzed under our two-rate Brownian motion model may appear to evolve more quickly than older clades. This is problematic as higher rates will be an artifact rather than evidence of increased disparity. To ensure that this behavior is not responsible for the rate differences we may see between reef and nonreef haemulids, we ran simulations under the maximum likelihood estimates of the OU parameters for each trait to create a "null" estimate of rate differences for each trait (see Supporting Information II for further details).

Results

Stochastic character mapping of reef and nonreef habitats on the 500 phylogenies sampled from the posterior distribution generated by BEAST revealed that on average there are three independent transitions to reef living in haemulids and 0.75 transitions away from reefs to other habitats (see Fig. 1 for an example).

The results of the model fitting are summarized as means and SEs for the one- and two-rate models across the 10,000 character-mapped phylogenies along with their Akaike weights (Table 1), and the model-averaged rates for reef and nonreef taxa with SEs (Table 2). There is strong support for the two-rate model that allows reef and nonreef species to evolve at different rates in the majority of trophic traits. The Akaike weights for the two-rate model are >0.75 for all traits except jaw closing ratio and eye width. Taking into account the weight of the different models, the model-averaged rates for each trait indicate that the rate of morphological evolution is higher on reefs than it is on nonreefs. The model-averaged rate of reef evolution ranges from 1.34 to 13.4 times faster than in nonreef species (see Table 2), with an average of 5.2 times faster on reefs.

In contrast, support for the two-rate model is much weaker in the locomotion and body shape traits, with only body depth, pectoral fin horizontal ratio, and caudal aspect ratio, having Akaike weights >0.75 for the two-rate model. However, the model-averaged rates for each trait do indicate that the rate of morphological evolution is higher on reefs than it is on nonreefs. On average, locomotor traits evolve 2.5 times faster in reef species. Although this result is not as pronounced as in the trophic traits, the model-averaged relative rate of reef evolution ranges from 1.02 to 5.77 times faster than in nonreef species. The one exception is the anal fin, which evolves slightly faster in nonreef species ($\times 1.02$ faster than reef species, Fig. 2, Table 2).

The impact of uncertainty in the phylogeny and character history on the rate estimates varies considerably between traits, as indicated by the 95% confidence intervals (95% CIs) around the rate estimates (see Fig. 2, Table 1). In general, the faster the rate estimate, the wider the confidence interval around the estimate. For example the confidence interval around suction index encompasses a 5–50 times faster rate on reef species relative to nonreef
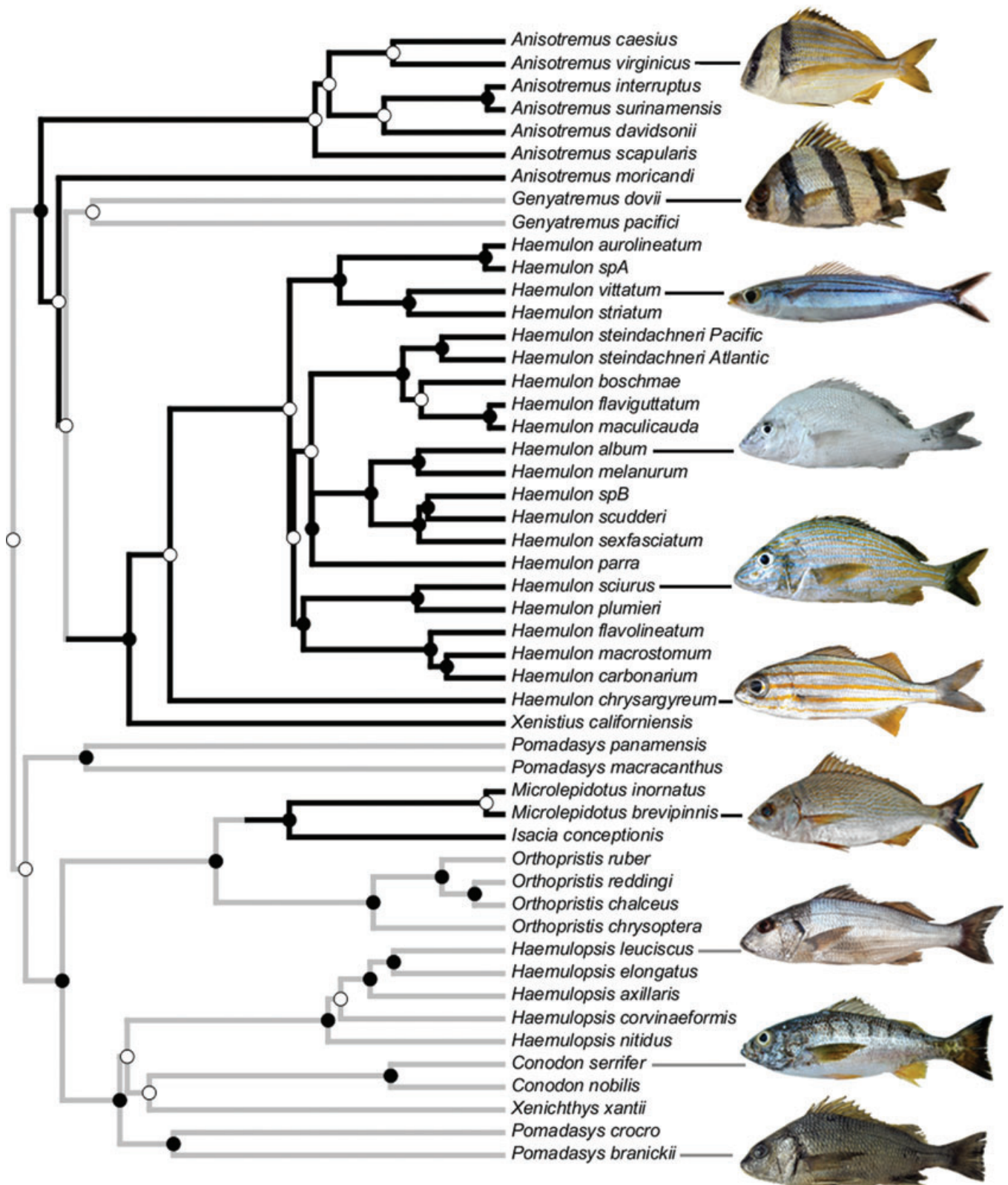
Figure 1. One of the 10,000 phylogenies used in the analyses, it depicts one of possible mapping reef living in gray and nonreef in black generated in SIMMAP v1.0 (Bollback 2006). Posterior probabilities of >0.95 are indicated by black circles on the nodes, these were calculated from the complete Bayesian posterior distribution of trees using the 50% majority rule tree. Nodes with white circles show that support was below 0.95 or that the branch did not appear in the 50% majority-rule tree.

Table 1. Model fit and estimated Brownian rate parameters for trophic and locomotion traits in haemulid fishes.

Type	Character	Single-rate model (two parameters)			Two-rate model (three parameters)			
		Likelihood	BM1rate ±SE	AICc weight	Likelihood	Rate nonreef ± SE	Rate reef + SE	AICc weight
Trophic morphology	Adductor mandibulae mass	−4.86	3.51±0.32	0.12	−1.78	1.57±1.08	4.68 ± 1.18	0.89
	Open ratio	60.48	0.26±0.07	0.07	64.15	0.10±0.78	0.35 ± 0.09	0.93
	Close ratio	65.52	0.21±0.06	0.49	66.62	0.14±0.06	0.25 ± 0.09	0.51
	Ascending process	−0.12	2.89±0.74	0.00	6.78	0.73±0.91	4.21 ± 0.97	1.00
	Raker length	−17.93	5.91±1.54	0.01	−12.44	1.82±1.81	8.36 ± 1.99	0.99
	Eye width	37.13	0.65±0.14	0.33	38.92	0.37±0.19	0.81 ± 0.17	0.67
	Suction index	−5.03	3.74±1.74	0.00	7.24	0.44±1.33	5.95 ± 2.70	1.00
	Buccal length	37.16	0.65±0.17	0.00	44.83	0.15±0.21	0.96 ± 0.22	1.00
	Buccal width	−7.72	3.92±0.97	0.24	−5.52	2.06±1.16	5.03 ± 1.25	0.76
	Head height	−26.51	8.62±3.41	0.00	−16.40	1.36±2.83	13.07 ± 5.05	1.00
	Head length	41.22	0.57±0.19	0.00	49.97	0.11±0.18	0.84 ± 0.27	1.00
Locomotion and body shape	Fineness ratio	−20.33	6.44±1.37	0.30	−18.43	3.50±1.97	8.26 ± 1.66	0.70
	Caudal aspect ratio	−55.06	26.05±6.79	0.04	−50.89	9.45±8.37	36.77 ± 9.32	0.96
	Spiny dorsal fin	42.85	0.51±0.11	0.72	42.96	0.49±0.15	0.53 ± 0.17	0.28
	Soft dorsal fin	21.60	1.20±0.26	0.26	23.69	0.64±0.35	1.55 ± 0.33	0.74
	Anal fin	32.15	0.79±0.16	0.73	32.22	0.82±0.23	0.77 ± 0.26	0.27
	Average spine length	22.05	1.19±0.28	0.17	24.11	0.57±0.36	1.56 ± 0.34	0.83
	Peduncle perimeter	46.78	0.44±0.10	0.50	47.86	0.29±0.13	0.53 ± 0.13	0.50
	Penduncle area	43.94	0.49±0.11	0.57	44.73	0.35±0.14	0.58 ± 0.15	0.43
	Body width	48.52	0.42±0.12	0.57	49.33	0.30±0.12	0.49 ± 0.17	0.44
	Body depth	19.98	1.31±0.41	0.00	26.56	0.33±0.43	1.94 ± 0.59	1.00
	Max. body depth position	51.28	0.37±0.08	0.58	52.01	0.27±0.11	0.43 ± 0.11	0.42
	Pectoral fin horizontal ratio	94.31	0.07±0.02	0.04	98.71	0.02±0.02	0.09 ± 0.02	0.97
	Pectoral fin vertical ratio	81.33	0.11±0.03	0.39	−159.45	0.07±0.03	0.14 ± 0.03	0.61

taxa, whereas that around eye width encompasses a 1.6–2.8 rate difference between reef and nonreef haemulids. Phylogenetic uncertainty also had a strong effect on the estimated correlation coefficients between certain traits (see Supporting Information I), for example, the correlation between suction index and body width was −0.52 on average but it ranged from weak (−0.25) to very strong (−0.83) correlations.

The simulations using the maximum likelihood estimates of the single-optimum OU parameters for each trait confirm that the younger clades do cause us to estimate an elevated rate of morphological evolution in reef fishes (see Supporting Information II). However, the magnitude of rate differences in the real data frequently exceed those estimated from the simulated data. Within the trophic traits, the median rates estimated from the data for ascending process, suction index, head height, and head length all exceed the 95% CI intervals around the rates estimated from the single-optimum OU simulations. In the locomotion and shape parameters only body depth exceeds the simulated OU rate. There are, however, only five traits (ascending process, head height, head length, buccal length, and body depth) where the CIs from the real- and single-peak OU rate estimates do not overlap. This means that the difference between reef and nonreef rates for these five traits exceed any rate that could be generated by a single-peak OU model and are therefore reef species clearly exhibit greater disparity. It should also be noted that the rate parameters for reef and nonreef taxa could not be estimated for eight of the 24 simulated OU datasets as the likelihood would not consistently converge in probability to the value being estimated and for many others it is only estimated for a small number of datasets that converged (see Supporting Information). This was most likely caused by a flat likelihood surface due to the lack of variance in these datasets, as the rate and selection parameter estimates were very

Table 2. Model averaged rate parameters for trophic and locomotion traits in haemulid fishes.

		Model-averaged rate	
Type	Character	Nonreef±SE	Reef±SE
Trophic morphology	Adductor mandibulae mass	1.79±1.19	4.54±1.19
	Open ratio	0.12±0.08	0.34±0.09
	Close ratio	0.17±0.06	0.23±0.07
	Ascending process	0.73±0.92	4.21±0.97
	Raker length	1.87±1.84	8.33±2.01
	Eye width	0.46±0.24	0.76±0.33
	Suction index	0.44±1.33	5.95±2.70
	Buccal length	0.15±0.21	0.96±0.22
	Buccal width	2.51±1.25	4.75±1.34
	Head height	1.36±2.83	13.05±5.05
	Head length	0.11±0.18	0.84±0.27
Locomotion and body shape	Fineness ratio	4.39±2.24	7.71±1.77
	Caudal aspect ratio	10.17±8.76	36.31±9.42
	Spiny dorsal fin	0.51±0.12	0.52±0.12
	Soft dorsal fin	0.79±0.41	1.46±0.34
	Anal fin	0.80±0.19	0.78±0.18
	Average spine length	0.68±0.41	1.50±0.36
	Peduncle perimeter	0.36±0.13	0.49±0.12
	Penduncle area	0.43±0.14	0.53±0.13
	Body width	0.36±0.12	0.45±0.13
	Body depth	0.34±0.43	1.93±0.59
	Max. body depth position	0.33±0.09	0.33±0.10
	Pectoral fin horizontal ratio	0.03±0.02	0.09±0.02
	Pectoral fin vertical ratio	0.08±0.04	0.13±0.03

low for these traits (see Supporting Information II). This lack of convergence indicates a poor fit of the two-rate model to these simulated datasets and the inability of the single-peak OU model to adequately represent these traits.

Discussion

Haemulids exhibit few trophic novelties and show comparable ecological diversity and species richness on and off reefs. Nevertheless, rates of ecomorphological evolution are faster in reef-dwelling species. Although we found evidence that some traits fit a single-optimum OU model better than the Brownian model, which could drive the faster rates on reefs for 11 of 24 traits, the overall pattern still supports the hypothesis that complex reef habitats promote the evolution of morphological diversity particularly within the trophic system of haemulids.

Both the strength of support for the conclusion of faster evolutionary rates in reef haemulids and the magnitude of the difference depend on the type of trait. The rate disparity between reef and nonreef species and the Akaike weight placed on the two-rate model is on average far greater in the trophic traits (average rate is five times faster on reefs and AICc weights

range from 0.51 to 1) compared to body shape or locomotor traits (average rate is 2.5 times faster on reefs and AICc weights range from 0.27 to 1). The stronger signal in trophic traits suggests a major role for trophic ecology in driving the rate disparity. Diet represents one of the key ways that organisms interface with their environment and is thus one of the primary axes of niche differentiation among species. Within surgeonfishes (Acanthuridae) and wrasses (Labridae) significant ecological diversification has been associated with dietary differences, as reflected in morphological differences in trophic and locomotion traits (Acanthuridae: Clements et al. 2003; Klanten et al. 2004; Labridae: Cowman et al. 2009; Price et al. 2011). The trophic traits we evaluated are part of the functional systems involved in prey capture and processing, and are known to underlie several components of feeding performance (Wainwright and Richard 1995; Holzman et al. 2012). These traits are thus linked to diet and are therefore likely to differentiate concomitantly with the trophic niche. Suction index, a complex index trait determined by five anatomical parameters and which is a morphological estimate of the capacity to generate pressure in the buccal cavity during suction feeding (Carroll et al. 2004; Collar and Wainwright 2006; Wainwright et al. 2007) shows the largest rate difference between reef and nonreef taxa
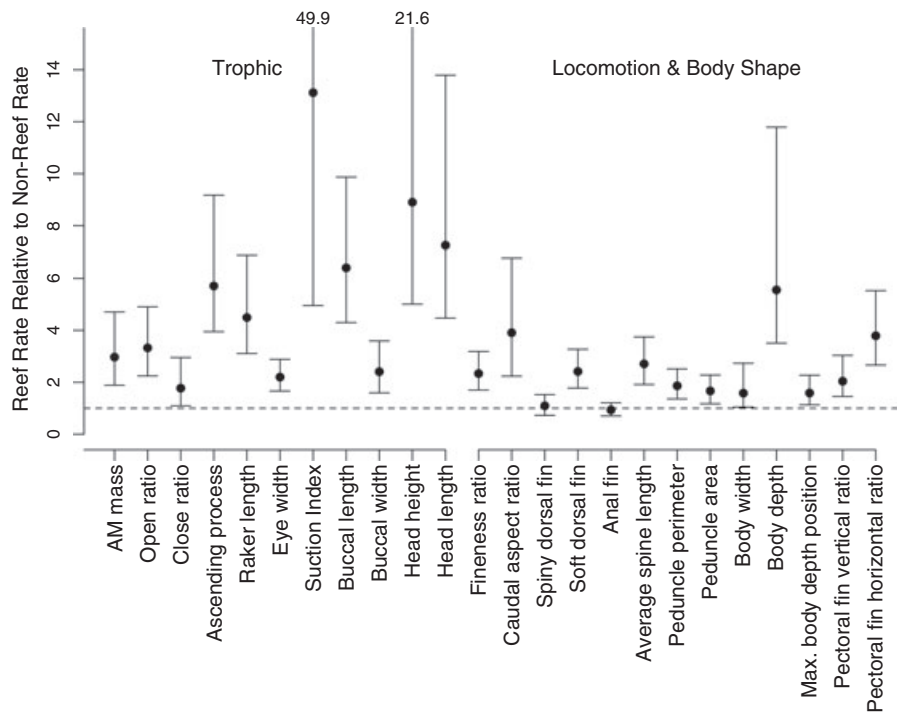
Figure 2. Estimated rate of Brownian motion evolution in reef-dwelling haemulids relative to the rate calculated in nonreef taxa from the two-rate model. Black error bars indicate 95% CIs of the rate estimates calculated across 10,000 stochastically mapped trees and the dashed horizontal line indicates rate equality when reef and nonreef taxa are evolving at the same rate.

although the CIs between the OU simulated and real data estimates do overlap. Of the five other trophic traits that exhibit strong rate differences, only head height is strongly correlated with suction index (see Supporting InformationI). This result indicates that diversification of suction feeding ability is a major component of trophic evolution in reef-dwelling haemulids but cannot explain all the differences observed.

Differences in the body shape and locomotor traits can reflect differences in diet. For example, high fineness ratios (a slender body shape) and high caudal fin aspect ratios are found in haemulid species that forage in midwater on zooplankton. However, locomotor traits are not as closely linked to diet as the trophic traits, which may explain why they exhibit slower rates of evolution. Alternatively, fin and shape traits may reflect habitat differences in reef fishes. In rabbitfishes (Siganidae) habitat variability appears to be an important driver of body-shape diversity as two distinct forms are found in different habitats; deep-bodied species are found on reef fronts and fusiform on reef flats (Borsa et al. 2007).

There are many reasons why some habitats may foster increased ecomorphological diversification compared to others (reviewed by Collar et al. 2010). We hypothesize that the biological and structural complexity of hard-bottomed reefs provide many opportunities for the organisms living upon them. High levels of resource biomass, species diversity, and abundance increase the potential for competition as well as resource partitioning, both of which may promote morphological diversification. In haemulids our interpretation is unfortunately hindered by the lack of detailed data concerning their diets. Reef and nonreef species appear to have similar dietary strategies; in contrast to labrids (Cowman et al. 2009; Price et al. 2010) there is no evidence of unique feeding niches evolving in reef dwellers. The majority of grunts feed on a variety of infaunal and mobile benthic invertebrates with a few species having more durophagous or zooplanktivorous niches (Randall 1967). However, some haemulids do show preferential feeding habits within these broadly defined dietary categories: bivalve molluscs in *Anisotremus scapularis* (Vargos et al. 1999; although see Medina et al. 2004), small shrimps in *Pomadasys tetradactylum and P. kaakan* (Hajisamae et al. 2003), gastropod/scaphopods in *Haemulopsis leuciscus* (Rodriguez-Romero et al. 2009). It is therefore possible that high diversity and abundance of prey types on and around reefs may facilitate niche partitioning and dietary specialization and thus elevate rates of morphological evolution in haemulids, especially as diversification rates are highest in trophic traits.

Moreover, competition may be stronger in biologically complex habitats where the diversity and density of competitors is likely to be greater. On reefs, the fish diversity and density is typically greater than in nonreef marine habitats. Increased competition may lead to elevated rates of character displacement either

during sympatric speciation or when species enter into secondary sympatry (see review by Dayan and Simberloff 2005) and thus raise rates of morphological evolution on reefs. Indeed, increased densities have been shown to favor extreme morphologies intraspecifically within three-spined stickleback (Bolnick 2005), which if it lead to speciation would likely elevate rates of character evolution among species.

Additionally, increased physical complexity may also contribute to elevated rates of morphological diversification as species adapt to different microhabitats. Reef environments are physically complex both in terms of rugosity (complexity of the seabed) and patchiness. Traditionally, high species diversity on coral reefs has been partly ascribed to habitat-type variation, because reef fishes tend to be sedentary and, to a limited extent, habitat specialists (Sale 1977). Indeed, a significant positive relationship between species diversity and habitat complexity has been recorded in both coral reef (e.g., Luckhurst and Luckhurst 1978) and rocky reef fishes (e.g., Garcia Charton and Perez Ruzafa 1998). It has also been suggested that in coral reef fishes, habitat shifts predate trophic diversification (Harmelin-Vivien 2002). Furthermore, both intra- and interspecific agonistic interactions for shelter sites have been shown to be higher than similar interactions for food in reef-dwelling fish (Shulman 1985), although this study did not include haemulids.

Are the morphological rate differences we observe linked to changes in lineage diversification? According to fishbase, approximately 50% of haemulid species are reef dwelling (www.fishbase.org, accessed 04/02/12), which suggests that ecomorphological and species diversity may not to be linked within haemulids. This inference is consistent with previous work on taxa as diverse as trilobites (Foote, 1993) and plethodontid salamanders (Adams et al., 2009). However, our study is primarily focused on species in the New World, which harbors more haemulid diversity on reefs (36 species) than nonreef habitats (27 species) (Tavera et al. 2012). Furthermore, New World haemulids are dominated by two speciose groups (*Haemulon and Anisotremus*) that appear to have radiated within reef habitats (Tavera et al. 2012). Therefore, we cannot rule out the possibility that the morphological diversity in reef species evolved as a result of ecological diversification during speciation. Unfortunately, we cannot quantitatively test for an association between reef living and lineage diversification using the Binary-State Speciation and Extinction models recently developed (BiSSE, Maddison et al. 2007; FitzJohn et al. 2009), as the Haemulidae phylogeny currently contains only 36% of the living species. Even if the phylogeny was complete with all 139 haemulid species, it may still be too small to provide enough power for accurate parameter estimates.

Patterns of elevated morphological diversity on reefs have been described qualitatively in other fish groups as well as sponges. For example, in marine angelfishes (Pomacanthidae), the pygmy angelfishes appear to exhibit notably higher morphological and ecological disparity than other angelfish clades along with greater fidelity to coral reefs (Bellwood et al. 2004). However, the stimulating impact of reefs on ecomorphological diversification in fishes may not be universal. It has been suggested that *Chaetodon*, a primarily reef-associated group of butterflyfishes (Chaetodontidae), exhibit reduced morphological diversity due to their obligate relationship with corals through corallivory (Bellwood et al. 2010). As an increasingly complete picture emerges concerning the impact of reefs on fish phenotypic diversification, a broader appreciation of the factors that cause this interaction will likely emerge. Finally, the greatest variety of morphologies in tropical intertidal sponges is found in coral reef environments compared to sand rubble, cave, boulders, mangrove swamp, and sea grass meadows habitats (Barnes 2001).

The invasion of structurally and biologically complex habitats is repeated across the tree of life. However, surprisingly few studies have investigated the impact that such invasions have upon the evolution of morphological disparity. In dragon lizards, a study recently found that ground-dwelling forms exhibit greater ecomorphological differentiation than rock or tree-living species, which may be due to the increased opportunities in the terrestrial habitat such as burrowing, living in leaf litter, in grass, or open areas or due to increases ecological interactions (Collar et al. 2010). In terms of community composition, freshwater fish assemblages in structurally more complex habitats have been shown to exhibit greater morphological diversity (Willis et al. 2005). But perhaps the most intriguing parallel result to our conclusion that highly productive and complex reef environments promote morphological diversification comes from an experimental evolution study of the bacterium *Pseudomonas fluorescens*. Morphological disparity within the *Pseudomonas* community increased with increases in the resource supply rate (analogous to productivity) in a complex environment with multiple food sources (Hall and Colegrave 2007).

Our results are consistent with the hypothesis that highly productive and complex habitats are centers of morphological diversification. Trophic morphology and to a lesser extent body shape and locomotory morphology evolves at higher rates in reef-dwelling haemulids. For 11 of 24 traits examined these rate differences between reef and nonreef taxa could potentially be an artifact of the trait evolving under a single-optima OU model. For a further eight traits, the best-fitting OU model parameters could not generate any difference in rates between reefs and nonreefs leading to a flat likelihood surface for the two-rate Brownian motion model, thus, the rate differences we observe in these traits are unlikely to be an artifact. Finally, there are four trophic traits (ascending process, head height, head length, and buccal length) and one locomotor trait (body depth) that unambiguously evolve faster in reef species regardless of whether they evolved under a

two-rate Brownian motion model or single-peak OU. Although the drivers of these high rates remain difficult to demonstrate, we suggest that greater diversity and abundance of prey and competitors on and around reefs may be driving fine-scale trophic niche partitioning and character displacement resulting in higher rates of morphological diversification on reefs. Our replication of the result that reef habitats lead to elevated diversification (Price et al. 2011) in a clade of phylogenetically and ecologically distant fishes provides further evidence that biologically and structurally reef complex habitats have stimulated morphological and functional diversification in marine fishes.

LITERATURE CITED

Alfaro, M., F. Santini, and C. D. Brock. 2007. Do reefs drive diversification in marine teleosts? Evidence from the pufferfishes and their allies (order Tetraodontiformes). Evolution 61:2104–2126.

Allen, G. R., and C. M. Roberts. 1994. Fishes of the tropical eastern Pacific. Univ. of Hawaii Press, Honolulu.

Bainbridge, R. 1960. Speed and stamina in three fish. J. Exp. Biol. 37:129–153.

Barnes, D. K. A. 2001. High diversity of tropical intertidal zone sponges in temperature, salinity and current extremes. Afr. J. Ecol. 37:424–434.

Bellwood, D. R., and P. C. Wainwright. 2002. The history and biogeography of fishes in coral reefs. Pp. 5–32 in P. F. Sale, ed. Coral reef fishes. Dynamics and diversity in a complex ecosystem. Academic Press, Boston, MA.

Bellwood, D. R., L. van Herwerden, and N. Konow. 2004. Evolution and biogeography of marine angelfishes (Pisces: Pomacanthidae). Mol. Phylogenet. Evol. 33:140–144.

Bellwood, D. R., S. O. Klanten, P. F. Cowman, M. S. Pratchett, N. Konow, and L. van Herwerden. 2010. Evolutionary history of the butterflyfishes (f: Chaetodontidae) and the rise of coral feeding fishes. J. Evol. Biol. 23:335–349.

Bollback, J. P. 2006. SIMMAP: Stochastic character mapping of discrete traits on phylogenies. BMC Bioinform. 7:88.

Bolnick, D. I. 2005. Can intraspecific competition drive disruptive selection? an experimental test in natural populations of sticklebacks. Evolution 58:608–618.

Borsa, P., S. Lemer, and D. Aurelle. 2007. Patterns of lineage diversification in rabbitfishes. Mol. Phylogenet. Evol. 44:427–435.

Burnham, K. P., and D. R. Anderson. 2002. Model selection and multimodel inference: a practical information theoretic approach. Springer-Verlag, New York.

Carroll, A. M., P. C. Wainwright, S. H. Huskey, D. C. Collar, and R. G. Turingan. 2004. Morphology predicts suction feeding performance in centrarchid fishes. J. Exp. Biol. 207:3873–3881.

Cervigón, F. 1993. Los peces marinos de Venezuela. Fundación Científica Los Roques, Caracas.

Chirichigno, N. F., and M. U. Cornejo. 2001. Catálogo comentado de los peces marinos del Perú. Instituto del Mar del Perú, Callao, Perú.

Clements, K. D., R. D. Gray, and J. H. Choat. 2003. Rapid evolutionary divergences in reef fishes of the family Acanthuridae (Perciformes: Teleostei). Mol. Phylogenet. Evol. 26:190–201.

Collar, D. C., and P. C. Wainwright. 2006. Discordance between morphological and mechanical diversification in the feeding mechanism of centrarchid fishes. Evolution 60:2575–2584.

Collar, D. C., B. C. O'Meara, P. C. Wainwright, and T. J. Near. 2009. Piscivory limits diversification of feeding morphology in Centrarchid fishes. Evolution 63:1557–1573.

Collar, D. C., J. A. I. Schulte, B. C. O'Meara, and J. B. Losos. 2010. Habitat use affects morphological diversification in dragon lizards. J. Evol. Biol. 23:1033–1049.

Cowman, P. F., and D. R. Bellwood. 2011. Coral reefs a drivers of cladogenesis: expanding coral reefs, cryptic extinction events, and the development of biodiversity hotspots. J. Evol. Biol. 24:2543–2562.

Cowman, P. F., D. R. Bellwood, and L. van Herwerden. 2009. Dating the evolutionary origins of wrasse lineages (Labridae) and the rise of trophic novelty on coral reefs. Mol. Phylogenet. Evol. 52:621–631.

Dayan, T., and D. Simberloff. 2005. Ecological and community-wide character displacement: the next generation. Ecol. Lett. 8:875–894.

Diaz-Uriarte, R., and T. Garland. 2010. PHYLOGR: functions for phylogenetically based statistical analyses.

Drummond, A. J., and A. Rambaut. 2007. BEAST: Bayesian evolutionary analysis by sampling trees. BMC Evol. Biol. 7:214.

Drummond, A. J., S. Y. W. Ho, M. J. Phillips, and A. Rambaut. 2006. Relaxed phylogenetics and dating with confidence. PLoS Biol. 4:e88.

Felsenstein, J. 1985. Phylogenies and the comparative method. Am. Nat. 125:1–15.

FitzJohn, R. G., W. P. Maddison, and S. P. Otto. 2009. Estimating trait-dependent speciation and extinction rates from incompletely resolved phylogenies. Syst. Biol. 58:595–611.

Fraser, R. H., and D. J. Currie. 1996. The species richness-energy hypothesis in a system where historical factors are thought to prevail – coral reefs. Am. Nat. 148:138–159.

Garcia Charton, J. A., and A. Perez Ruzafa. 1998. Correlation between habitat structure and a rocky reef fish assemblage in the southwest Mediterranean. Marine Ecol. 19:11–128.

Gratwicke, B., and M. R. Speight. 2005. The relationship between fish species richness, abundance and habitat complexity in a range of shallow tropical marine habitats. J. Fish Biol. 66:650–667.

Hajisamae, S., L. M. Chou, and S. Ibrahim. 2003. Feeding habits and trophic organization of the fish community in shallow waters of an impacted tropical habitat. Estuar. Coast. Shelf Sci. 58:89–98.

Hall, A. R., and N. Colegrave. 2007. How does resource supply affect evolutionary diversification. Proc. R. Soc., Lond. B 274:73–78.

Hamilton, J., and B. Konar. 2007. Implications of substrate complexity and kelp variability for south-central Alaskan nearshore fish communities. Fish. Bull. 105:189–196.

Harmelin-Vivien, M. L. 2002. Energetics and fish diversity on coral reefs. Pp. 265–274 in P. F. Sale, ed. Coral reef fishes. Dynamics and diversity in a complex ecosystem. Academic Press, Boston, MA.

Harmon, L. J., J. T. Weir, C. D. Brock, R. E. Glor, and W. Challenger. 2008. GEIGER: investigating evolutionary radiations. Bioinformatics 24:129–131.

Holzman, R. A., D. C. Collar, R. S. Mehta, and P. C. Wainwright. 2012. An integrative approach to elucidate suction feeding performance. J. Exp. Biol. 215:1–13.

Huelsenbeck, J. P., R. Nielsen, and J. P. Bollback. 2003. Stochastic mapping of morphological characters. Syst. Biol. 52:131–158.

Hurvich, C. M., and C.-L. Tsai. 1989. Regression and time series model selection in small samples. Biometrika 76:297–307.

Hutcheon, J. M., and T. Garland. 2004. Are megabats big? J. Mammal. Evol. 22:257–277.

Kiessling, W., C. Simpson, and M. Foote. 2010. Reefs as cradles of evolution and sources of biodiversity in the Phanerozoic. Science 327:196–198.

Klanten, S. O., L. van Herwerden, J. H. Choat, and D. Blair. 2004. Patterns of lineage diversification in the genus Naso (Acanthuridae). Mol. Phylogenet. Evol. 32:221–235.

Levinton, J. S. 1995. Marine biology. function, biodiversity, ecology. Oxford Univ. Press, New York.

Lighthill, J. 1975. Mathematical biofluiddynamics. CMBS-NSF regional conference series in applied mathematics. Vol. 17. Society for Industrial and Applied Mathematics. Philadelphia, PA. Pp. 1–283.

Lindeman, K. S., and C. S. Toxey. 2003. Haemulidae. Pp. 1522–1550 in K. E. Carpenter, ed. FAO species identification guide for fishery purposes. The living marine resources of the Western Central Atlantic. Volume 3 Bony fishes part 2 (Opistognathidae to Molidae), sea turtles and marine mammals. FAO Library, Rome.

Luckhurst, B. E., and K. Luckhurst. 1978. Analysis of the influence of substrate variables on coral reef fish communities. Marine Biol. 49:317–323.

MacArthur, R. H., and R. Levins. 1964. Competition, habitat selection, and character displacement in a patchy environment. Proc. Natl. Acad. Sci. 51:1207–1210.

MacArthur, R. H., and J. W. MacArthur. 1961. On bird species diversity. Ecology 42:594–593.

Maddison, W. P., P. E. Midford, and S. P. Otto. 2007. Estimating a binary character's effect on speciation and extinction. Syst. Biol. 56:701–710.

McClanahan, T. R. 1994. Kenyan coral reef lagon fish: effects of fishing, substrate complexity, and sea urchins. Coral Reefs 13:231–241.

McEachran, J. D., and J. D. Fechelm. 2005. Fishes of the Gulf of Mexico: scorpaeniformes to tetraodontiformes. Univ. of Texas Press, Austin, TX.

McKay, R. J., and M. Schnider. 1995. Haemulidae. In W. Fischer, F. Krupp, W. Schneider, C. Sommer, K. E. Carpenter, and V. H. Niem, eds. Guía FAO para la identificación de especies para los fines de la pesca: Pacífico centro-oriental. FAO, Roma.

Medina, M., M. Araya, and C. Vega. 2004. Alimentacion y relaciones troficas de peces costeros de la zona norte de Chile. Invest. Mar. 32:33–47.

Monismith, S. G. 2006. Hydrodynamics of Coral Reefs. Ann. Rev. Fluid Mech. 39:37–55.

Nielsen, R. 2002. Mapping mutations on phylogenies. Syst. Biol. 51:729–739.

O'Meara, B. C., C. Ane, M. J. Sanderson, and P. C. Wainwright. 2006. Testing for different rates of continuous trait evolution using likelihood. Evolution 60:922–933.

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

Price, S. A., P. C. Wainwright, D. R. Bellwood, E. Kazancioglu, D. C. Collar, and T. J. Near. 2010. Functional innovations and morphological diversification in parrotfishes. Evolution 64:3057–3068.

Price, S. A., R. A. Holzman, T. J. Near, and P. C. Wainwright. 2011. Coral reefs promote the evolution of morphological diversity and ecological novelty in labrid fishes. Ecol. Lett. 14:462–469.

R. Development Core Team. 2012. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.

Randall, J. E. 1967. Food habits of reef fishes of the West Indies. Stud. Trop. Oceanogr. 5:655–847.

Renema, W., D. R. Bellwood, J. C. Braga, K. Bromfield, R. Hall, K. G. Johnson, P. Lunt, C. P. Meyer, L. B. McMonagle, R. J. Morley, et al.

2008. Hopping hotspots: global shifts in marine biodiversity. Science 321:654–657.

Revell, L. 2009. Size-correction and principal components for interspecific comparative studies. Evolution 63:3258–3268.

Revell, L. J. 2012. Phytools: an R package for phylogenetic comparative biology (and other things). Methods Ecol. Evol. 3:217–223.

Roberts, C. M., C. J. McClean, J. E. N. Veron, J. P. Hawkins, G. R. Allen, D. E. McAllister, C. G. Mittermeier, F. W. Schueler, M. Spalding, F. Wells, et al. 2002. Marine biodiversity hotspots and conservation priorities for tropical reefs. Science 295:1280–1284.

Rodriguez-Romero, J., X. G. Moreno-Sanchez, L. A. Abitia-Cardenas, and D. S. Palacios-Salgado. 2009. General characteristics of diet of the raucous grunt, Haemulopsis leuciscus (Perciformes: Haemulidae), in Bahia Almejas, Baja California Sur, Mexico. Mar. Biodivers. Rec. 2:e141.

Rosenzweig, M. L. 1995. Species diversity in space and time. Cambridge University Press, Cambridge, UK.

Sale, P. F. 1977. Maintenance of high diversity in coral reef fish communities. The American Naturalist 11:337–359.

Schmitz, L., and P. C. Wainwright. 2011. Nocturnality limits morphological and functional diversity in the eyes of reef fishes. BMC Evol. Biol. 11:338.

Schoener, T. W. 1974. Resource partitioning in ecological communities. Science 185:27–39.

Shulman, M. J. 1985. Coral reef fish assemblages: intra-and interspecific competition for shelter sites. Environ. Biol. Fishes 13:81–92.

Steneck, R. S., M. H. Graham, B. J. Bourque, D. Corbett, J. M. Erlandson, J. A. Estes, and M. J. Tegner. 2002. Kelp forest ecosystems: biodiversity, stability, resilience and future. Environ. Conserv. 29:436–459.

Tavera, J. J., A. P. Acero, E. F. Balart, and G. Bernardi. 2012. Molecular phylogeny of grunts (Teleostei, Haemulidae), with an emphasis on the ecology, evolution, and speciation history of New World species. BMC Evol. Biol. 12:57.

Thomas, G. H., R. P. Freckleton, and T. Székely. 2006. Comparative analyses of the influence of developmental mode on phenotypic diversification rates in shorebirds. Proc. R. Soc.i, Lond. B 273:1619–1624.

Thomson, D. A., L. T. Findley, and A. N. Kerstitch. 2000. Reef fishes of the Sea of Cortez: the rocky-shore fishes of the Gulf of California. Univ. of Texas Press, Austin.

Vargos, M., P. Fuentes, P. Hernaez, A. Olivares, and P. Rojas. 1999. Relaciones troficas de cinco peces costeros comunes en el area submareal del norte de Chile. Rev. Biol. Trop. 47. 601–604.

Wainwright, P. C., and B. Richard. 1995. Predicting patterns of prey use from morphology with fishes. Environ. Biol. Fishes 44:97–113.

Wainwright, P. C., D. R. Bellwood, M. W. Westneat, J. R. Grubich, and A. S. Hoey. 2004. A functional morphospace for the skull of labrid fishes: patterns of diversity in a complex biomechanical system. Biol. J. Linn. Soc. 82:1–25.

Wainwright, P. C., A. M. Carroll, D. C. Collar, S. W. Day, T. E. Higham, and R. A. Holzman. 2007. Suction feeding mechanics, performance, and diversity in fishes. Integr. Comp. Biol. 47:96–106.

Webb, P. W. 1984. Body form, locomotion and foraging in aquatic vertebrates. Am. Nat. 24:107–120.

Willis, S. C., K. O. Winemiller, and H. Lopez-Fernandez. 2005. Habitat structural complexity and morphological diversity of fish assemblages in a Neotropical floodplain river. Oecologia 142:284–295.

Associate Editor: L. Harmon

Supporting Information

The following supporting information is available for this article:

Supporting Information I. Phylogenetically correct correlations between all morphological traits.
Supporting Information II. Description of analyses and results using the single-peak Ornstein-Uhlenbeck (OU) model.

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

Please note: Wiley-Blackwell is not responsible for the content or functionality of any supporting information supplied by the authors. Any queries (other than missing material) should be directed to the corresponding author for the article.