

RAPID CLADOGENESIS IN MARINE FISHES REVISITED

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Abstract.—Lineages that underwent rapid cladogenesis are attractive systems for the study of mechanisms underlying taxonomic, ecological, morphological, and behavioral diversification. Recently developed statistical methods provide insights into historical patterns of diversity and allow distinguishing bursts of cladogenesis from stochastic background rates in the presence of confounding factors such as extinction and incomplete taxon sampling. Here, we compare the dynamics of speciation in several marine fish lineages some of which were previously proposed to have undergone significant changes of cladogenesis through time. We tested for evidence of episodes of rapid cladogenesis using the constant rate and Monte Carlo constant rate tests that are robust to incomplete taxon sampling. These tests employ the statistic γ to measure the relative position of internal node in a chronogram. For the first time, we conducted a comparative analysis to address the behavior of the statistic under different chronogram-constructing methods (Langley-Fitch, nonparametric rate smoothing, and penalized likelihood). Although estimates of γ sometimes differ widely among methods, acceptance or rejection of the constant rate model within a particular clade appears to be robust to the choice of method. Bursts of cladogenesis were detected in 14 of 34 studied datasets. Some of these were previously proposed to represent marine fish “radiations,” whereas others are identified anew. Our results indicate that the wider application of tree shape methods that are able to detect significantly elevated rates of speciation is useful to more precisely define clades that underwent episodes of rapid cladogenesis in marine fish clades. Contrasting the patterns of phylogenetic diversification in marine fish lineages may facilitate the identification of common evolutionary trajectories versus idiosyncrasies, and ultimately help towards a better understanding of the factors and processes underlying speciation in the marine realm.

Key words.—Constant rate test, diversification rates, nonparametric rate smoothing, penalized likelihood, radiation.

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Molecular phylogenies have become increasingly important in understanding the tempo and mode of diversification that shaped the biodiversity we observe today. Tree topologies can be employed to identify asymmetries in the number of descendant species between contemporaneous sister clades, and hence variation in diversification rates among lineages (Slowinski and Guyer 1989; Sanderson and Donoghue 1994, 1996; Agapow and Purvis 2002; Chan and Moore 2002). In addition, branch length information can be used to detect changes in rates of cladogenesis through time within a single lineage, for example, in association with specific climatic or geological events (Nee et al. 1994a, 1996; Pybus and Harvey 2000; Magallón and Sanderson 2001; Barraclough and Vogler 2002; Pybus et al. 2002). Branch length information can also be used to identify early episodes of rapid cladogenesis on a phylogenetic tree, and thus help to determine the historical factors (e.g., evolution of key innovations or colonization of novel habitats) underlying the emergence of taxonomic, ecological, morphological, or behavioral disparity (Simpson 1953; Stanley 1979; Sanderson and Donoghue 1994; Schluter 2000; Magallón and Sanderson 2001; Harmon et al. 2003).

Although approximately 13,000 marine teleost species are known, which account for about 61% of the total teleost diversity (Nelson 1994), little is known about the evolutionary mechanisms underlying diversification in marine fish species. In contrast to the wealth of studies documenting bursts of cladogenesis in lacustrine environments related to, for example, colonization of novel habitats and niche exploitation (e.g., Fryer and Iles 1972; Echelle and Kornfield 1984; Schluter 1996), few studies have focused on marine fishes (e.g.,

Clark and Johnston 1996; Johns and Avise 1998; Streebman et al. 2002). Often, poorly resolved trees are interpreted as representing marine fish “radiations” or “rapid speciation” events without further statistical testing (e.g., Clements et al. 2003; but see Rüber et al. 2003).

A noteworthy exception is the study by Johns and Avise (1998) on the temporal patterns of cladogenesis in two marine fish lineages (rockfishes and notothenioids). Significant bursts of cladogenesis were detected in the two datasets studied, identifying the two marine lineages as ancient species flocks (see also Schluter 2000). Hence, their data showed that evolutionary radiations are not only restricted to well documented lacustrine fish groups, but may also occur in marine systems.

The comparative analyses of Johns and Avise (1998) were based on a “null model of stochastic diversification” proposed by Wollenberg et al. (1996). However, Paradis (1998) showed that this model is not appropriate to test changes of cladogenesis through time. Furthermore, Pybus and Harvey (2000) pointed out that several existing methods that address changes of cladogenesis through time, including the one by Wollenberg et al. (1996), are limited by the assumption of complete taxon sampling. Pybus and Harvey (2000) and Pybus et al. (2002) proposed new statistical methods, based upon the constant rate (CR) and Monte Carlo constant rate (MCCR) tests, to study changes of cladogenesis through time that are robust to incomplete taxon sampling, and are conservative with respect to extinction.

Pybus and Harvey’s (2000) test use the statistic γ to measure the relative position of internal nodes in a linearized phylogeny (chronogram). Under a constant speciation rate

model, γ values of complete reconstructed phylogenies follow a standard normal distribution. Positive or negative values of γ indicate that tree internal nodes are closer to the tips (apparent increase in diversification rates towards the present) or to the root (apparent deceleration in diversification rates towards the present) than expected under the null model, respectively. Because incomplete taxon sampling results in lower γ values compared to fully sampled phylogenies (Nee et al. 1994b), the MCCR test can be used to simulate the null distribution of γ when only a portion of the extant species are sampled (for details, see Pybus and Harvey 2000).

With the rapid accumulation of molecular sequence data there has been an increased interest in divergence time estimation. However, comparisons of relative rates of molecular evolution between lineages often show strong departure from a molecular clock, an observation that has prompted the development of several new approaches of divergence time estimation in the absence of rate homogeneity (Sanderson 1997, 2002; Thorne et al. 1998). Although several studies have focused on comparisons of divergence time estimates obtained under different methods and/or different fossil calibration points (Magallón and Sanderson 2001; Sanderson 2002), the behavior of the parameter γ under these different methods has not been studied thus far (but see Barraclough and Vogler 2002).

Here, we revisit evidences of episodes of rapid cladogenesis in several marine fish clades. To this end we reanalyzed the “ancient” marine species flocks studied by Johns and Avise (1998) based on extended datasets, as well as other representative marine fish clades. The specific aim of this study was (1) to test for deviations in the constant rate model, and (2) to compare the behavior of the parameter γ under different chronogram reconstruction methods.

MATERIALS AND METHODS

Testing Diversification Rate through Time

A total of 14 datasets representing 11 marine fish clades were obtained from GenBank. For several of the datasets, we also analyzed specific subclades separately (resulting in a total of 34 datasets; see Table 1 for a summary of the datasets). Protein coding nucleotide sequence datasets were aligned by eye, whereas 12S rRNA and 16S rRNA gene nucleotide sequence datasets were aligned with SOAP version 1.05a (Löytynoja and Milinkovitch 2001). For each of the latter datasets, we used SOAP to generate 45 alternative CLUSTAL W (Thompson et al. 1994) alignments (gap opening penalty ranging from 7–15; gap extension penalty ranging from 3–7; both in increments of one). Unstable positions that differed between the alternative alignments were excluded (strict alignment option in SOAP) and alignments were further inspected visually. The hierarchical likelihood ratio test (hLRT) implemented in MODELTEST version 3.06 (Posada and Crandall 1998) was used to determine the substitution model that best fit each of the datasets. Maximum likelihood (ML) analyses were conducted with PAUP* version 4.10 (Swofford 2002) using the model parameters obtained from MODELTEST. Likelihood ratio tests (Huelsenbeck and Crandall 1997) were performed with ML trees with and with-

out a molecular clock constraint to test constancy of evolutionary rates among taxa.

To perform tests of diversification rates, chronograms were constructed using three different methods, implemented in r8s version 1.60 (Sanderson 2003): Langley-Fitch (LF; Langley and Fitch 1974), nonparametric rate smoothing (NPRS; Sanderson 1997), and penalized likelihood (PL; Sanderson 2002). Langley-Fitch assumes a molecular clock whereas NPRS uses a least squares smoothing of local estimates of substitution rates under a relaxed molecular clock assumption. Finally, PL also uses a relaxed molecular clock assumption by combining a parametric model with different substitution rates on every branch with a nonparametric roughness penalty which costs the model more if rates change too quickly from branch to branch. The TN algorithm was used for both the LF and PL methods, whereas the Powell algorithm was used for the NPRS method (Sanderson 2003). Cross-validation was used for all datasets and methods. To find the optimal smoothing parameter (λ) for PL, cross-validation was performed over a range of values of λ ranging from 1 to 10^4 .

We investigated rates of cladogenesis through time in each of the datasets taking incomplete taxon sampling into account. To this end, we followed the procedures proposed by Pybus and Harvey (2000) using the CR and MCCR tests. Values of γ were calculated based upon the LF, NPRS, and PL trees using Genie version 3.0 (Pybus and Rambaut 2002). We then conducted the MCCR test by calculating the γ distribution with 10,000 replicates using MCCRTTest (Pybus 2000) and the numbers of total and sampled species as given in Table 1.

The CR and MCCR tests were conducted under the assumption that diversification occurs equally among lineages. In order to test this assumption we used two approaches to detect nonstochastic differences in species diversity among lineages. First, the B_1 index, (calculated with MESA version 1.5.3d; Agapow 2002) was used as a measure of tree imbalance (Kirkpatrick and Slatkin 1993). Second, the relative cladogenesis statistic (Nee et al. 1994a, 1996) as implemented in End-Epi version 1.0 (Rambaut et al. 1997) was used to identify lineages with unusually slow or rapid diversification rates.

RESULTS

The molecular clock was clearly rejected for all datasets analyzed (Table 1). The B_1 test indicated that rates of diversification in several groups were not equal among lineages (Table 1). In those cases, we used the relative cladogenesis statistic to identify lineages with unusually slow or rapid diversification rates. Following Pybus and Harvey (2000), we sequentially removed species from those lineages creating new phylogenies until they were not rejected under the B_1 test (see Table 1 for details). For each dataset we reconstructed chronograms under LF, NPRS, and PL, and the estimated γ values are shown in Figure 1. To test if γ significantly differs from the constant-rate pure birth model, we employed the CR/MCCR tests that take into account the effect of incomplete taxon sampling (Nee et al. 1994b; Pybus and Harvey 2000). The critical values of γ are shown in

TABLE 1. Description of the datasets analyzed in this study. Datasets with the same number and different letter refer to the same alignment with different number of species considered. The prefix “s” indicates subclades.

Dataset	Taxon	Genes	Alignment (bp)	Number of sampled species (x)	True number of species (y) ¹³	x/y ratio	Molecular clock (LRT) rejected	B _i
1A	Acanthuridae	12S rRNA + 16S rRNA	884	25	82	0.30	yes (P < 0.001)	12.48 (P > 0.05)
s1B	Acanthurini	12S rRNA + 16S rRNA	884	11	46	0.24	—	5.28 (P > 0.05)
2A	Acanthuridae	16S rRNA	521	40	82	0.49	yes (P < 0.001)	17.18 (P < 0.05)
2B	Acanthuridae	16S rRNA	521	39 ³	81	0.48	—	17.07 (P < 0.05) ¹⁴
s2C	Acanthurini	16S rRNA	521	11	46	0.24	—	3.45 (P > 0.05)
s2D	Naso	16S rRNA	521	20	20	1.00	—	9.40 (P > 0.05)
3A	Carangidae	cytochrome b	1140	56	140	0.40	yes (P < 0.001)	25.86 (P < 0.05)
3B	Carangidae	cytochrome b	1140	51 ⁴	140	0.36	—	24.46 (P > 0.05)
s3C	Carangini	cytochrome b	1140	39	96	0.41	—	18.40 (P > 0.05)
4A	Chaethodontidae	cytochrome b + 12rRNA	759	44	114	0.39	yes (P < 0.001)	22.95 (P > 0.05)
s4B	Chaetodon	cytochrome b + 12rRNA	759	34	89	0.38	—	17.35 (P > 0.05)
s4C	Chaetodon	cytochrome b + 12rRNA	759	32 ⁵	89	0.36	—	17.52 (P > 0.05)
5A	Cheilodactylidae ¹	cytochrome b + COI	806	25	31	0.81	yes (P < 0.001)	12.51 (P > 0.05)
s5B	Cheilodactylidae ²	cytochrome b + COI	806	22	27	0.81	—	11.01 (P > 0.05)
6A	Gobiosomatini	12S rRNA + tRNA Val + 16S rRNA	1646	54	131	0.41	yes (P < 0.001)	25.41 (P < 0.05)
6B	Gobiosomatini	12S rRNA + tRNA Val + 16S rRNA	1646	52 ⁶	131	0.40	—	25.04 (P > 0.05)
7A	Notothenioidei	12S rRNA + 16S rRNA	764	36	112	0.32	yes (P < 0.001)	15.21 (P < 0.05) ¹⁴
7B	Notothenioidei	12S rRNA + 16S rRNA	764	35 ⁷	111	0.32	—	15.10 (P < 0.05) ¹⁴
s7C	Trematominae	12S rRNA + 16S rRNA	764	11	14	0.79	—	4.08 (P < 0.05)
s7D	Trematominae	12S rRNA + 16S rRNA	764	10 ⁸	14	0.71	—	4.08 (P > 0.05)
8A	Notothenioidei	16S rRNA	1651	53	123	0.43	yes (P < 0.001)	26.97 (P > 0.05)
s8B	Notothenioidei	16S rRNA	1651	49 ⁹	111	0.44	—	25.78 (P > 0.05)
s8C	Channichthyidae	16S rRNA	1651	16	16	1.00	—	8.59 (P > 0.05)
9	Pomacanthidae	12S rRNA + 16S rRNA	841	24	88	0.27	yes (P < 0.001)	13.00 (P > 0.05)
10	Pomacentridae	cytochrome b + ATP8 + 6	1982	102	355	0.29	yes (P < 0.001)	51.82 (P > 0.05)
11A	Scaridae	12S rRNA + 16S rRNA	806	21	90	0.23	yes (P < 0.001)	9.02 (P < 0.05)
11B	Scaridae	12S rRNA + 16S rRNA	806	20 ¹⁰	90	0.22	—	8.88 (P > 0.05)
s11C	Scarinae	12S rRNA + 16S rRNA	806	9	72	0.13	—	4.50 (P > 0.05)
s11D	Spariosomatinae	12S rRNA + 16S rRNA	806	12	18	0.67	—	5.28 (P > 0.05)
12A	Sebastes	cytochrome b	782	77	110	0.70	yes (P < 0.001)	36.01 (P < 0.05)
12B	Sebastes	cytochrome b	782	75 ¹¹	110	0.68	—	35.48 (P < 0.05) ¹⁴
13A	Sparidae	16S rRNA	548	60	110	0.55	yes (P < 0.001)	24.94 (P < 0.05)
13B	Sparidae	16S rRNA	548	58 ¹²	110	0.53	—	24.79 (P < 0.05) ¹⁴
14	Sparidae	cytochrome b + 16S rRNA	1694	41	110	0.37	yes (P < 0.001)	21.78 (P > 0.05)

¹ Including the Latridae. ² Excluding the latrid genera *Larridopsis* and *Latris*. ³ *Luvanus imperialis* (Luvaridae, 1 sp) excluded. ⁴ *Uraspis helvola*, *Selene peruviana*, *Selene dorsalis*, *Caranx hippos*, and *Caranx melampygus* excluded. ⁵ *Chaetodon aculeatus* and *Chaetodon kleini* excluded. ⁶ *Elacatinus oceanops* and *Garmannia robustum* excluded. ⁷ Basal *Eleginops maclovius* (Eleginopidae, 1 sp) excluded. ⁸ *Trematomus loembergi* excluded. ⁹ Members of the basal families Bovichthidae (10 spp), Pseudaphritidae (1 sp), and Eleginopidae (1 sp) excluded. ¹⁰ *Sparisoma viridae* excluded. ¹¹ *Sebastes minor* and *Sebastes aleutianus* excluded. ¹² *Crenidens crenidens* and *Lithognathus mormyrus* excluded. ¹³ Species numbers are from Bellwood et al. (2004); Pomacanthidae, Burridge and Smolenski (2004); Cheilodactylidae, Eastman and Eakin (2000); Notothenioidei; see also Near et al. (2004). For dataset 7A members of the basal families Bovichthidae (10 spp) and Pseudaphritidae (1 sp) were not included in the phylogeny. Klanten et al. (2004); Acanthuridae, Luvaridae (1 sp) and Zanchidae (1 sp) were also included based on Tang et al. [1999] and Clements et al. (2003), Nelson (1994); Carangidae, Chaethodontidae, and *Sebastes*), Orrell and Carpenter (2004; Sparidae), Quenouille et al. (2004; Pomacentridae), Rüber et al. (2003; Gobiosomatini), and Strelman et al. (2002; Scaridae). ¹⁴ Sequential removal of more taxa did not result in significantly more balanced trees.

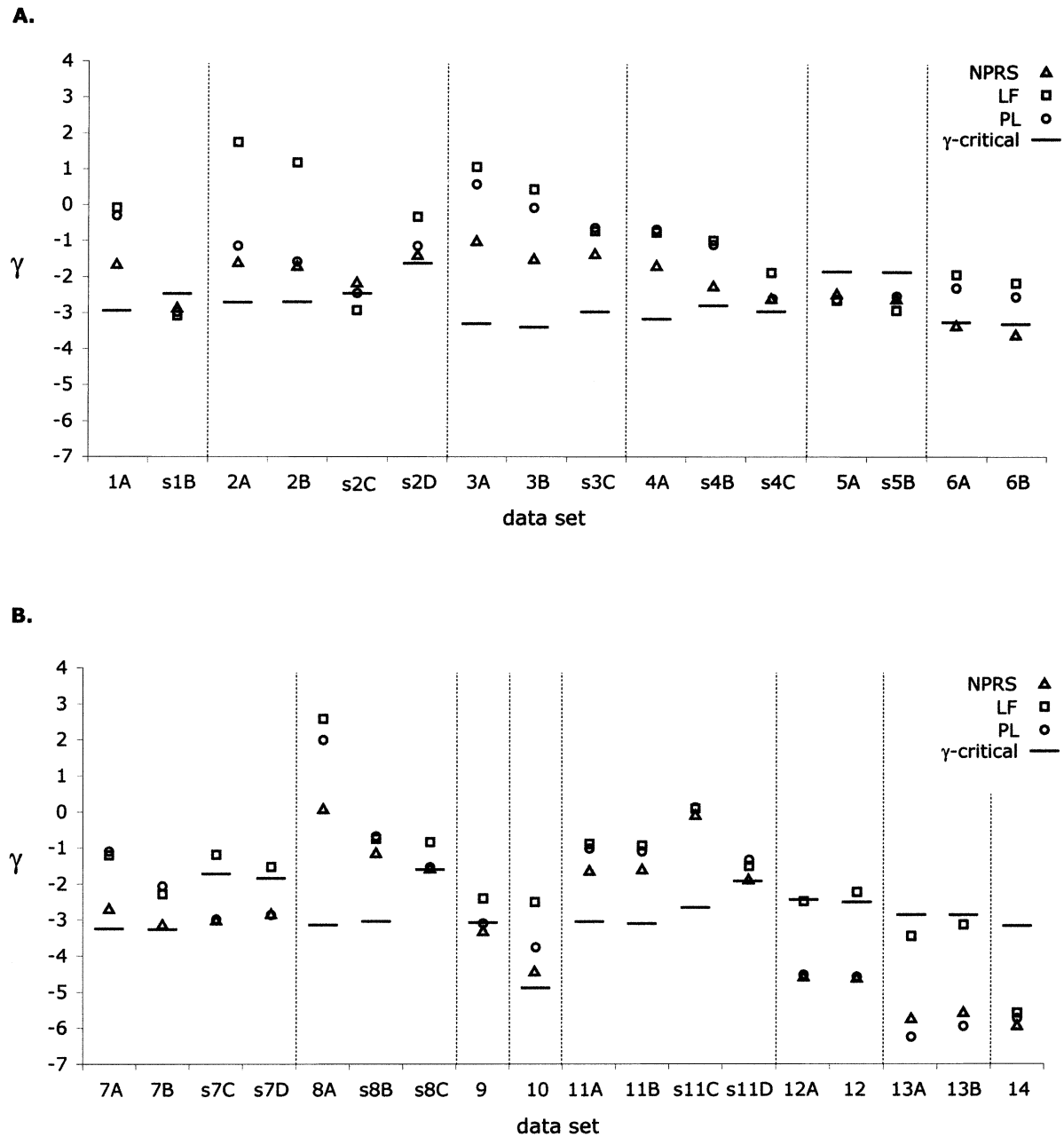


FIG. 1. Testing episodes of early rapid cladogenesis in representative marine fish clades. Significance of the statistic γ was evaluated with the CR/MCCR tests based on Langley-Fitch, nonparametric rate smoothing, and penalized-likelihood chronograms. The critical value of γ taking incomplete taxon sampling into account is given. Those γ values below the critical γ values reject the constant rate model and therefore support early rapid episodes of speciation (as highlighted in Table 2). Numbers on the x-axis refer to datasets given in Table 1.

Figure 1. Those γ values obtained with LF, NPRS, or PL below the critical value indicate a significant clustering of internal nodes at the base of the tree, indicative of an early episode of rapid cladogenesis followed by a slowdown in diversification rate. For some datasets, estimation of γ using the three chronogram-reconstruction methods rendered similar values. However in other cases, estimations of γ with the different methods were widely spread. Generally, the LF, PL, and NPRS methods obtained the highest, intermediate, and lowest γ values, respectively. It is interesting to note

that in several cases the γ values obtained with PL were very close to those obtained under a clock-enforced tree (LF), although the molecular clock was clearly rejected in all datasets. Significant rejection of the constant rate model with all methods (LF, NPRS, and PL) was found for seven (s1B, 5A, s5B, 12A, 13A, 13b, and 14) out of 34 analyzed datasets (Fig. 1 and Table 2). Another four datasets (s7C, s7D, 9, and 12B) were rejected with NPRS and PL, two only with NPRS (6A and 6B), and one only with LF (s2C). Therefore, our results indicate episodes of rapid cladogenesis for the

following taxa: Cheilodactylidae, Pomacanthidae, Sparidae, Trematominae, Acanthurini, Gobiosomatini, and *Sebastes* (Table 2).

DISCUSSION

Methodological Considerations and Areas of Future Research

Here, we used phylogenetic and statistical methods to detect changes in diversification rate through time in representative marine fish clades. To this end, we applied the CR/MCCR tests that take incomplete taxon sampling into account (as recommended by Pybus and Harvey 2000). The results allowed us to identify episodes of rapid cladogenesis in 14 of 34 studied datasets using different methods of chronogram reconstruction (LF, NPRS, and PL) (Fig. 1 and Table 2). To our knowledge, our study is the first to compare the behavior of the tree-shape parameter γ in relation to LF, NPRS, and PL reconstruction. Even though estimates of γ sometimes clearly differed among methods, results regarding acceptance or rejection of the constant rate model were generally congruent within datasets.

There are several methodological issues that need to be considered regarding the significance of our findings: (1) We found evidence for significant deviations of the constant rate model in 14 datasets. However, the CR/MCCR tests are quite conservative and have high type II errors (Pybus and Harvey 2000). Therefore, we cannot discard that some of the 20 datasets that failed to reject the constant rate model might in fact also represent true episodes of rapid cladogenesis. (2) Some of the nodes in the reconstructed phylogenies showed little statistical support. Phylogenetic error could represent a potential pitfall of the CR/MCCR tests. However, estimation of the parameter γ should not be substantially affected by minor changes in the topology since the CR/MCCR tests use internode-distance information (Pybus and Harvey 2000), and regions of topological uncertainty generally correspond to short nodes. (3) Our alignments varied between 521–1982 nucleotides. Estimates of branch length are more consistent with larger datasets. When more sequence information becomes available, it will be interesting to address how current estimates of γ will be affected by additional nucleotide sampling. (4) CR/MCCR tests may be affected by over- or underdispersed taxon sampling (Pybus and Harvey 2000). However, we are not aware of any summary statistic to detect the direction of nonrandom taxon sampling. Nevertheless, several of the analyzed datasets have a fair representation of the clade's species diversity (>50% of the species), and in these cases, it is reasonable to assume an unbiased taxon sampling. Another related issue is that some of the reconstructed molecular phylogenies are in clear conflict with current taxonomy, and this further complicates the evaluation of sampling biases. (5) In Table 1, we provide current total species numbers for each clade. In the future, new species might be described. A sensitivity analysis for the critical value of γ can be easily used to evaluate the robustness of the results in that event.

Rapid Cladogenesis in Marine Fishes

Broad-scale allopatry has traditionally been thought to be the main speciation mechanism in the ocean because of the apparent absence of geographic isolating barriers, and the potential for long-distance dispersal of pelagic fish larvae that counteract local adaptation (Palumbi 1994; Shulman and Bermingham 1995; Mora and Sale 2002). However, evidence of self-recruitment is accumulating, and speciation in the marine realm may therefore occur at more localized geographic scales than previously believed (Cowen et al. 2000; Taylor and Hellberg 2003). Lineages showing bursts of cladogenesis are particularly well-suited model systems to evaluate the relative contribution of ecological differentiation and geographical isolation in the speciation process (Schluter 2000).

Based upon molecular phylogenetic analyses, we identified episodes of significantly elevated rates of cladogenesis in rockfishes (genus *Sebastes*), Antarctic fishes (subfamily Trematominae), surgeonfishes (tribe Acanthurini), American seven-spined gobies (tribe Gobiosomatini), morwongs (family Cheilodactylidae), angelfishes (family Pomacanthidae), and porgies (family Sparidae). Thus, our analyses provide compelling evidence for episodes of cladogenetic bursts in the marine realm that can be compared with well-known examples from lacustrine and terrestrial environments (Schluter 2000). Although some of these marine fish clades were previously identified as "radiations" with either alternative (Johns and Avise 1998) or similar (Rüber et al. 2003) statistical tests, others were merely suggested without further statistical testing, and yet others are identified anew in this study (see Table 2 for details).

In contrast to the results of Johns and Avise (1998), we did not find a significant clustering of cladogenetic events in time for the entire notothenioids (excluding the Bovichtidae, the Pseudaphritidae, and the Eleginopidae). Hence, our results do not support the hypothesis that the evolution of antifreeze glycopeptides, in the ancestor of the notothenioids was a key innovation that prompted a notothenioid diversification burst (Bargelloni et al. 1994; Clark and Johnston 1996; Eastman 2000; Eastman and McCune 2000). In other previously suggested (but not statistically tested) "radiations," including Acanthuridae (Clements et al. 2003, and Scaridae (Streelman et al. 2002), we could not detect significant changes of cladogenesis through time.

Although it is beyond the scope of this work to discuss in detail the potential causes underlying each of the detected cladogenetic bursts, we provide in Table 2, a summary of main proposed factors in the source papers. Generally, colonization of novel habitats followed by ecological differentiation is invoked as a main factor promoting rapid diversification of lineages and ecomorphological traits (Schluter 2000; Rüber et al. 2003; Streelman and Danley 2003) (Table 2). It is thought that a decrease in the rate of cladogenesis following an episode of rapid speciation may be due to a decrease of speciation opportunities as ecological and/or geographical spaces are filled (Schluter 2000).

We hope that the detection of rapid cladogenetic events in marine fish clades using CR/MCCR tests as shown here, will prompt future research efforts towards more complete taxon

TABLE 2. Comparison of previous findings and results obtained in this study for representative marine fish clades.

Dataset	Taxon	Data source references	Summary of findings about episodes of rapid cladogenesis (source studies and references therein)	Evidence (this study)
1A	Acanthuridae	1, 2	Two periods of rapid radiation inferred. Not statistically tested. Potential causes mentioned include: eocene radiation characterized by ecomorphological divergence.	No
s1B	Acanthurini	1, 2	Rapid radiation inferred. Not statistically tested. Potential causes mentioned include: ecomorphological divergence.	Yes; NPRS, PL, LF
2A	Acanthuridae	1, 2	Two periods of rapid radiation inferred. Not statistically tested. Potential causes mentioned include: Eocene radiation characterized by ecomorphological divergence.	No
2B	Acanthuridae	1, 2	Two periods of rapid radiation inferred. Not statistically tested. Potential causes mentioned include: Eocene radiation characterized by ecomorphological divergence.	No
s2C	Acanthurini	1, 2	Rapid radiation inferred. Not statistically tested. Potential causes mentioned include: ecomorphological divergence.	Yes; LF
s2D	<i>Naso</i>	3	Rapid cladogenesis discarded. Not statistically tested, but lineage through time (LTT) plot on absolute timescale presented.	No
3A	Carangidae	4	Episodes of rapid cladogenesis not considered.	No
3B	Carangidae	4	Episodes of rapid cladogenesis not considered.	No
s3C	Carangini	4	Episodes of rapid cladogenesis not considered.	No
4A	Chaethodontidae	Not available	No published molecular phylogeny available.	No
s4B	<i>Chaetodon</i>	5	Episodes of rapid cladogenesis not considered.	No
s4C	<i>Chaetodon</i>	5	Episodes of rapid cladogenesis not considered.	No
5A	Cheilodactylidae	6	Episodes of rapid cladogenesis not considered.	Yes; NPRS, PL, LF
s5B	Cheilodactylidae	6	Episodes of rapid cladogenesis not considered.	Yes; NPRS, PL, LF
6A	Gobiosomatini	7	Episodes of early rapid cladogenesis inferred. Statistically tested and found significant. Potential causes mentioned include: adaptive radiation in stages (major habitat shifts followed by ecomorphological diversification).	Yes; NPRS
6B	Gobiosomatini	7	Episodes of early rapid cladogenesis inferred. Statistically tested and found significant. Potential causes mentioned include: adaptive radiation in stages (major habitat shifts followed by ecomorphological diversification).	Yes; NPRS
7A	Nothothenioidei	8–16	Episodes of early rapid cladogenesis inferred. Statistically tested and found significant. Potential causes mentioned include: adaptive radiation (colonization of Antarctic waters, facilitated by the key innovation of antifreeze glycoproteins (AFGP), pelagization).	No
7B	Nothothenioidei	8–16	Episodes of early rapid cladogenesis inferred. Statistically tested and found significant. Potential causes mentioned include: adaptive radiation (colonization of Antarctic waters, facilitated by the key innovation of AFGP, pelagization).	No
s7C	Trematominae	16, 17	Rapid radiation suggested. Statistically tested and found significant. Potential causes not further discussed.	Yes; NPRS, PL
s7D	Trematominae	16, 17	Rapid radiation suggested. Statistically tested and found significant. Potential causes not further discussed.	Yes; NPRS, PL
8A	Notothenioidei	8–16	Episodes of early rapid cladogenesis inferred. Statistically tested and found significant. Potential causes mentioned include: adaptive radiation (colonization of Antarctic waters, facilitated by the key innovation of AFGP, pelagization). Episodes of early rapid cladogenesis inferred.	No

TABLE 2. Continued.

Dataset	Taxon	Data source references	Summary of findings about episodes of rapid cladogenesis (source studies and references therein)	Evidence (this study)
s8B	Notothenioidei	8–16	Statistically tested and found significant. Potential causes mentioned include: adaptive radiation (colonization of Antarctic waters, facilitated by the key innovation of AFGP, pelagization).	No
s8C	Channichthyidae	13, 14	Episodes of rapid cladogenesis not considered.	No
9	Pomacanthidae	18	Episodes of rapid cladogenesis not considered.	Yes; NPRS, PL
10	Pomacentridae	19, 20	Episodes of rapid cladogenesis not considered. Evidence for a marine adaptive radiation presented but not for episodes of early rapid cladogenesis.	No
11A	Scaridae	21, 22	Potential causes mentioned include: adaptive radiation (major habitat shifts followed by ecomorphological diversification, sexual selection important in later stages of the radiation). Evidence for a marine adaptive radiation presented but not for episodes of early rapid cladogenesis.	No
11B	Scaridae	21, 22	Potential causes mentioned include: adaptive radiation (major habitat shifts followed by ecomorphological diversification, sexual selection important in later stages of the radiation). Evidence for a marine adaptive radiation presented but not for episodes of early rapid cladogenesis.	No
s11C	Scarinae	21, 22	Potential causes mentioned include: adaptive radiation (major habitat shifts followed by ecomorphological diversification, sexual selection important in later stages of the radiation). Evidence for a marine adaptive radiation presented but not for episodes of early rapid cladogenesis.	No
s11D	Spariosomatinae	21, 22	Potential causes mentioned include: adaptive radiation (major habitat shifts followed by ecomorphological diversification). Episode of early rapid cladogenesis inferred. Statistically tested.	No
12A	<i>Sebastes</i>	15, 16, 23, 24	Potential causes mentioned include: adaptive radiation characterized by rapid speciation, divergence in habitat use, ecomorphology, and reproductive traits. Episode of early rapid cladogenesis inferred. Statistically tested.	Yes; NPRS, PL, LF
12B	<i>Sebastes</i>	15, 16, 23, 24	Potential causes mentioned include: adaptive radiation characterized by rapid speciation, divergence in habitat use, ecomorphology, and reproductive traits. Episode of early rapid cladogenesis inferred. Statistically tested.	Yes; NPRS, PL
13A	Sparidae	25	Episodes of rapid cladogenesis not considered.	Yes; NPRS, PL, LF
13B	Sparidae	25	Episodes of rapid cladogenesis not considered.	Yes; NPRS, PL, LF
14	Sparidae	25	Episodes of rapid cladogenesis not considered.	Yes; NPRS, PL, LF

References: 1, Clements et al. (2003); 2, Tang et al. (1993); 3, Klanten et al. (2004); 4, Reed et al. (2002); 5, McMillan et al. (1999); 6, Burrige and Smolenski (2004); 7, Rüber et al. (2003); 8, Eastman and McCune (2000); 9, Klingenberg and Ekau (1996); 10, Montgomery and Clements (2000); 11, Clark and Johnston (1996); 12, Bargelloni et al. (1994); 13, Bargelloni et al. (2000); 14, Near et al. (2004); 15, Schluter (2000); 16, Johns and Avise (1998); 17, Ritchie et al. (1996); 18, Bellwood et al. (2004); 19, Jang-Liaw et al. (2002); 20, Quenouille et al. (2004); 21, Streelman et al. (2002); 22, Streelman and Danley (2003); 23, Kai et al. (2003); 24, Rocha-Olivares et al. (1999); 25, Orrell and Carpenter (2004).

and gene sampling in these groups. The wider application of CR/MCCR tests to detect episodes of rapid cladogenesis, together with analyses of morphological and ecological disparity among evolutionary lineages (e.g., Harmon et al. 2003), should assist in answering fundamental questions about tempo and mode of marine fish radiations. For instance, applying absolute time scales to the study of changes in diversification rates through time may allow the identification of common evolutionary trajectories (such as geological or climatic events) versus idiosyncrasies of the different marine fish lineages.

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