Molecular Phylogenetics and Evolution 50 (2009) 268–281

Contents lists available at ScienceDirect

Molecular Phylogenetics and Evolution

journal homepage: www.elsevier.com/locate/ympev

Molecular phylogeny reveals extensive ancient and ongoing radiations in a snapping shrimp species complex (Crustacea, Alpheidae, *Alpheus armillatus*)

Lauren M. Mathews [a,*], Arthur Anker [b,1]

[a] Department of Biology, Worcester Polytechnic Institute, 100 Institute Road, Worcester, MA 01609, USA
[b] Smithsonian Tropical Research Institute, Naos Unit 0948, APO A.A. 34002, USA

ARTICLE INFO

Article history:
Received 7 May 2008
Revised 18 October 2008
Accepted 30 October 2008
Available online 14 November 2008

Keywords:
Species complex
Snapping shrimp
Alpheidae
Alpheus armillatus
Cryptic radiation
Western Atlantic
Eastern Pacific
Transisthmian

ABSTRACT

Tropical marine habitats often harbor high biodiversity, including many cryptic taxa. Though the prevalence of cryptic marine taxa is well known, the evolutionary histories of these groups remain poorly understood. The snapping shrimp genus *Alpheus* is a good model for such investigations, as cryptic species complexes are very common, indicating widespread genetic diversification with little or no morphological change. Here, we present an extensive phylogeographic investigation of the diversified amphi-American *Alpheus armillatus* species complex, with geographic sampling in the Caribbean Sea, Gulf of Mexico, Florida, Brazil, and the tropical Eastern Pacific. Sequence data from two mitochondrial genes (16SrRNA and cytochrome oxidase I) and one nuclear gene (myosin heavy chain) provide strong evidence for division of the species complex into six major clades, with extensive substructure within each clade. Our total data set suggests that the *A. armillatus* complex includes no less than 19 putative divergent lineages, 11 in the Western Atlantic and 8 in the Eastern Pacific. Estimates of divergence times from Bayesian analyses indicate that the radiation of the species complex began ~10 MYA with the most recent divergences among subclades dating to within the last 3 MY. Furthermore, individuals from the six major clades had broadly overlapping geographic distributions, which may reflect secondary contact among previously isolated lineages, and have apparently undergone several changes in superficial coloration, which is typically the most pronounced phenotypic character distinguishing lineages. In addition, the extensive substructure within clades indicates a great deal of molecular diversification following the rise of the Isthmus of Panama. In summary, this investigation reflects substantial biodiversity concealed by morphological similarity, and suggests that both ancient and ongoing divergences have contributed to the generation of this biodiversity. It also underlines the necessity to work with the most complete data set possible, which includes comprehensive and wide-ranging sampling of taxa.

1. Introduction

The study of biodiversity is one of the oldest empirical disciplines in biology, with roots extending as far back as the works of Aristotle (Tipton, 2006), but in recent decades this field has undergone drastic changes for at least two reasons. First, the introduction of molecular genetic technologies has changed the way we describe and catalogue biological diversity, and the use of these technologies has led to new and exciting inferences into the evolutionary dynamics of lineages and the interplay between genetic and morphological evolution. Second, declines in species numbers and in population sizes in many marine taxa (Powles et al., 2000) have placed a premium on our understanding of ecological systems and the origin and maintenance of biodiversity. Consequently, understanding the complex processes by which biodiversity is generated has become both a goal of basic science and a tool of importance to how we manage biological resources.

Many marine habitats, especially tropical shallow-water areas, are known to harbor high biodiversity (Gray, 1997). This presents an interesting conundrum, as most marine habitats were long thought to be highly interconnected by gene flow, with populations of all but the least dispersive taxa predicted to be genetically homogeneous (Palumbi, 1994). However, high biodiversity in marine systems suggests that genetic divergence and speciation may be common in marine systems, and the evolutionary processes (e.g., vicariance or divergent selection) that generate this biodiversity remain largely unknown. A number of investigations suggest that marine populations may not be as interconnected as they seem; some taxa show moderate to high levels of genetic differentiation between populations, in some cases even over small spatial scales (Hedgecock, 1986; Palumbi, 1994; Duffy, 1996a,b; Palumbi

* Corresponding author.
E-mail address: lmathews@wpi.edu (L.M. Mathews).
[1] Present address: 250A Dickinson Hall, Florida Museum of Natural History, University of Florida, Gainesville, FL 32611-7800, USA.

1055-7903/$ - see front matter © 2008 Elsevier Inc. All rights reserved.
doi:10.1016/j.ympev.2008.10.026

et al., 1997; Benzie, 1999; Barber et al., 2002; Gutiérrez-Rodríguez and Lasker, 2004; Perrin et al., 2004; Baratti et al., 2005; Bilodeau et al., 2005; Mathews, 2007). Other investigations have revealed evidence for strong genetic differentiation among evolutionary lineages previously thought to represent single species, a phenomenon that has been referred to widely in the recent literature as "cryptic speciation", because it is difficult or even impossible to detect these species using only traditional morphology-based methods. Where an evolutionary lineage has undergone frequent cryptic speciation, this process has been referred to as a "non-adaptive radiation" (Gittenberger, 1991).

Knowlton (1993) considered the occurrence of cryptic species to be common in marine habitats. For example, molecular evidence has contributed to the detection of cryptic taxa in a wide range of marine taxa, including mollusks (Lee and Ó Foighil, 2004; Ellingson and Krug, 2006), polychaetes (Bastrop et al., 1998), algae (Šlapeta et al., 2006), cnidarians (Dawson and Jacobs, 2001), sponges (Solé-Cava et al., 1991), echinoderms (Baric and Sturmbauer, 1999; Wilson et al., 2007), foraminiferans (de Vargas et al., 1999), ascidians (Tarjuelo et al., 2001; Caputi et al., 2007), crustaceans (Rocha-Olivares et al., 2001; Machordom and Macpherson, 2004; Mathews, 2006), and fishes (Colborn et al., 2001), as well as in some parasitic and "commensal" taxa (Miura et al., 2006; Baker et al., 2007). However, though the taxonomic importance of marine cryptic species is well established, the evolutionary dynamics of large-scale non-adaptive radiations in marine ecosystems are still understudied.

Snapping shrimps of the family Alpheidae are a ubiquitous component of benthic communities in tropical and subtropical marine habitats. Most of them, including the focal taxon, live under rocks, in crevices of coral rocks, or in self-excavated burrows in sandy, rocky, or muddy substrates. *Alpheus* and *Synalpheus* are by far the most speciose genera, with almost 300 recognized species of *Alpheus* (Williams et al., 2001; A. Anker, personal observation) and ~160 recognized species of *Synalpheus* (Chace, 1988; Macdonald et al., 2006; Ríos and Duffy, 2007). In addition, empirical evidence for the presence of many cryptic lineages (Knowlton and Keller, 1983; McClure and Greenbaum, 1994; Bruce, 1999; Williams et al., 2001; Anker, 2001a; Macdonald et al., 2006; Mathews, 2006; Anker et al., 2007a,b, 2008a,b) suggests that both genera conceal much cryptic biodiversity. For these reasons, snapping shrimp have been used as models for phylogeographic studies. For example, seminal papers by Knowlton et al. (1993) and Knowlton and Weigt (1998) investigated the role of the Isthmus of Panama in generating species diversity in *Alpheus*. More recently, Morrison et al. (2004) reported evidence for an extensive radiation in the *Synalpheus gambarelloides* species group in the Caribbean approximately 5–7 million years ago (MYA). Large numbers of cryptic alpheids are known worldwide (e.g., Anker, 2001a), and at least some diversification in the genera occurred around the time of the closure of the Isthmus of Panama, around 3 MYA (Coates and Obando, 1996).

In the present investigation, we have targeted the *Alpheus armillatus* species complex for elucidation of its evolutionary history. This complex currently includes four nominal species in the Western Atlantic: *A. armillatus*, very inadequately described by Milne-Edwards (1837); *A. lancirostris*, described by Rankin (1900); *A. verrilli*, described by Schmitt (1924); and *A. angulosus*, described by McClure (1995, 2002); and six species in the Eastern Pacific: *A. scopulus*, *A. hyeyoungae*, *A. tenuis*, *A. martini*, all described by Kim and Abele (1988); *A. wickstenae*, described by Christoffersen and Ramos (1987); and *A. agrogon*, described by Ramos (1997). Two of them, *A. lancirostris* and *A. verrilli*, are presently considered as junior synonyms of *A. armillatus* (Verrill, 1922; Armstrong, 1949; for complete synonymy see Anker, 2001b). The main morphological characters distinguishing the *A. armillatus* complex are the flattened, V- or U-shaped postrostral area, the absence of balaeniceps setae on the minor chela (in both sexes), and the walking legs ending in a simple conical dactylus. Chace (1972) reported *A. armillatus* from as far north as North Carolina, USA, through the Caribbean, and to as far south as São Paulo, Brazil; strangely, he did not comment on the status of *A. verrilli*. Hendrix (1971) first suggested that *A. armillatus* may include cryptic taxa, based on differences in morphology and coloration. Later, Christoffersen and Ramos (1988) reported two different "color types" of *A. armillatus* in Brazil. These observations strongly suggest that this complex may indeed include potentially a large number of cryptic lineages.

Here, we present a molecular phylogenetic investigation of a large collection of specimens from the entire Caribbean region (Honduras, Puerto Rico, Cuba, Jamaica, Panama, Costa Rica, Venezuela), the southeastern United States (Florida), northeastern Brazil (Atol das Rocas), and the Pacific coast of Central America (several localities in Panama, Costa Rica). This taxa set considerably extends previous phylogeographic studies of the *A. armillatus* complex, which focused on southeastern USA and northern Caribbean (Mathews, 2006), providing a much more comprehensive picture of the evolutionary history of this diverse species complex. In addition, we discuss phenotypic differentiation in this morphologically conserved complex, and test alternative hypotheses on the evolution of color patterns, which have been cited as characters of potential systematic importance in snapping shrimp (Knowlton and Mills, 1992).

2. Material and methods

2.1. Sample collection, identification, and processing

Shrimp were collected from a total of 27 sites (Fig. 1). From all sites, shrimps were collected by hand or with the aid of a small hand-net. From sites 1–5, 7–8, and 10–12, specimens were collected by L.M. and handled as described in Mathews (2006); additional sequences from individuals in those collections were obtained for the current investigation. From sites 6 and 13–25, all specimens were collected by A.A. as follows: site 6, collected by hand under rocks at low tide; sites 13–14, collected with a hand-net while snorkeling, or extracted from crevices in rocks and coral rubble by hand or suction pump; site 15, collected by flipping large rocks while scuba diving; sites 16–25, collected by hand from under rocks at extreme low tide; site 26, collected from subtidal rubble by a diver team. From site 27, shrimps were collected by Paulo S. Young (late of Museu Nacional, Rio de Janeiro, Brazil); field notes indicate they were collected by hand in the intertidal. All specimens were preserved in 75–98% ethanol. Most specimens were photographed alive prior to preservation; brief color notes were made for specimens collected from sites 6 and 13, and 27. All specimens were identified to currently recognized species or to undescribed morphospecies (e.g., as *A. cf. armillatus*) based on morphological criteria and color patterns.

We recorded information on color characteristics for several features that appear to show variation among evolutionary lineages (Table 1 and Fig. 2); these include the overall coloration and pigmentation pattern of the dorsal carapace and abdomen; the position and number of dark spots on the abdominal somites (when present); the coloration and pigmentation pattern of the major chela; and the coloration of the antennular and antennal flagella. We note that at this time we have no data on intraspecific variability in color patterns, and consequently, we have limited our phylogenetic consideration of color patterns to those characteristics that are likely to be consistent within a lineage, based on our previous experiences with the species complex; specifically, these are the presence or absence of abdominal bands and paired

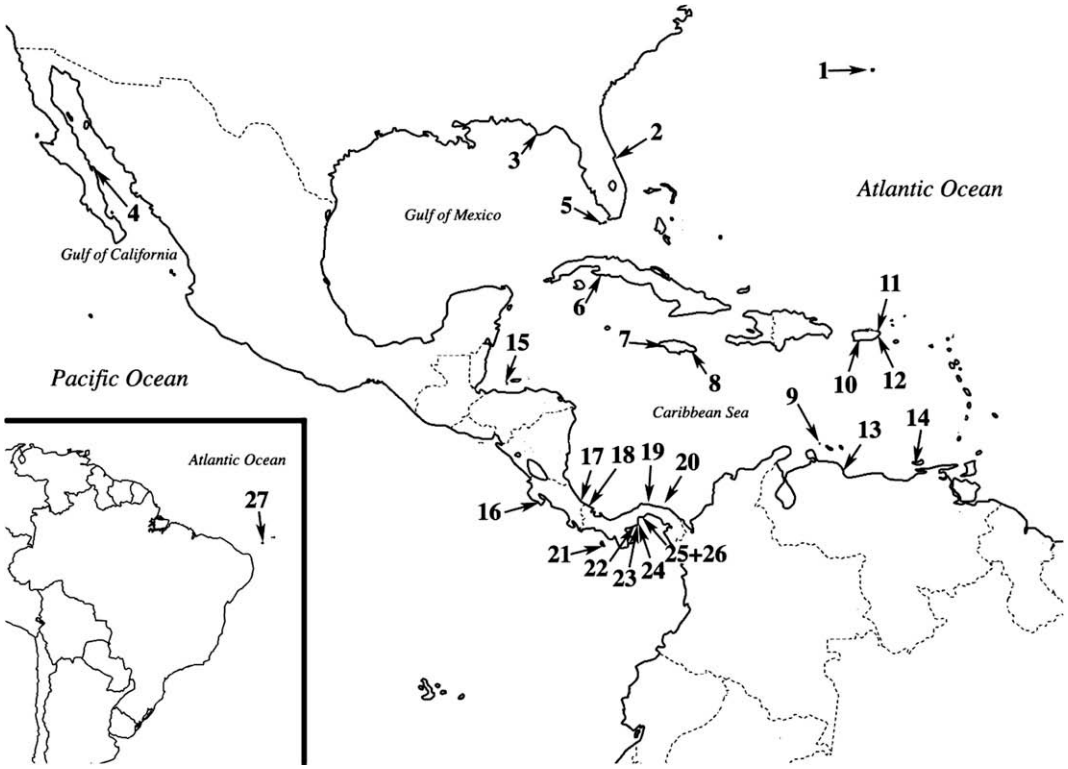

Fig. 1. Map showing collection sites of shrimp specimens used in this study; 1, Somerset Bridge, Bermuda; 2, Fort Pierce, Florida; 3, Carrabelle, Florida; 4, Playa Santispac, Baja California, Mexico; 5, Florida Keys (Key West, Pine Key); 6, Playa Girón, Cuba; 7, Sandy Bay, Jamaica; 8, Runaway Bay, Jamaica; 9, Baby Beach, Aruba; 10, Aguirre, Puerto Rico; 11, Las Croabas, Puerto Rico; 12, Ceiba, Puerto Rico; 13, Morrocoy, Venezuela; 14, Isla Margarita and Isla Cubagua, Venezuela; 15, Utila, Honduras; 16, Punta Morales, Costa Rica; 17, Cahuita, Costa Rica; 18, Bocas del Toro, Panama; 19, Isla Grande, Panama; 20, San Blas Islands, Panama; 21, Coiba, Panama; 22, Playa Venao, Panama; 23, Río Mar, Panama; 24, Veracruz, Panama; 25, Isla Taboga, Panama; 26, Casco Viejo and Amador Causeway, Panama; 27, Atol das Rocas, Brazil.

Table 1
Description of major color pattern types observed in lineages of the *A. armillatus* complex. Photo numbers refer to Fig. 2; phylogenetic associations of color types are illustrated in Fig. 4.

Color form	Overall coloration	Paired lateral abdominal spots	Major chela	Antennae	Photo
Abdomen non-banded (NB types)					
NB1	Yellowish and finely reticulated	None	Marbled, without spots, bright pink or purplish distally	Yellowish	2A
NB2	Grayish, speckled with small dark spots	None	Marbled with grey and green, without spots	Bluish	2B
NB3	Orange–brown, with minute spots	None	Red–brown, speckled with whitish spots	Orange proximally, bluish distally	2C
NB4	Brown to grey–green, sometimes yellowish, faintly reticulated	None	Marbled with grey and green, lower notch conspicuously whitish	Bluish	2E
NB5	Whitish, speckled with greenish spots	None	Uniform green–brown, not speckled or blotched	Pale greenish-blue	2L
NB6	Yellowish, reticulated with brown	None	Speckled with interconnected spots and blotches	Yellowish proximally, bluish distally	2O
Abdomen banded (BA types)					
BA1	Broad, brown to bluish-green abdominal bands	None	Brown–green, speckled with round bluish-whitish spots, not marbled	Yellow	2I
BA2	Broad, brown to bluish-green abdominal bands	None	Marbled and blotched with brown–green and white + whitish spots	Blue	2H
BA3	Broad, brown to bluish-green abdominal bands	None	Marbled and blotched with brown–green and white + whitish spots	Yellow	2G, J
BA4	Brown abdominal bands with strong speckling and reticulation	Somites 2 and 4: lateral spots; Somite 3: dorsolateral spots	Brown covered with blotches and minute spots	Pale brown to orange	2F
BA5	Brown-greenish abdominal bands with some spotting	Somites 2 and 4: lateral spots	Brown–green with whitish spots	Greenish-yellow	2N
BA6	Brown abdominal bands, narrower than in NB4, dark patch on 5th abdominal somite	Somite 3: dorsolateral spots	Uniform brown–green, without spots	Greenish-yellow	2K
BA7	Orange–brown with fine yellow reticulation, interrupted by narrow white bands	None	Pale brown to greenish-brown with white or bluish blotches and spots	Bluish-green	2M
BA8	Very broad brown bands (interrupted by very narrow white bands)	Somites 2 and 4: lateral spots	Uniform brown–green, without spots or blotches	Pale green–yellow	2D

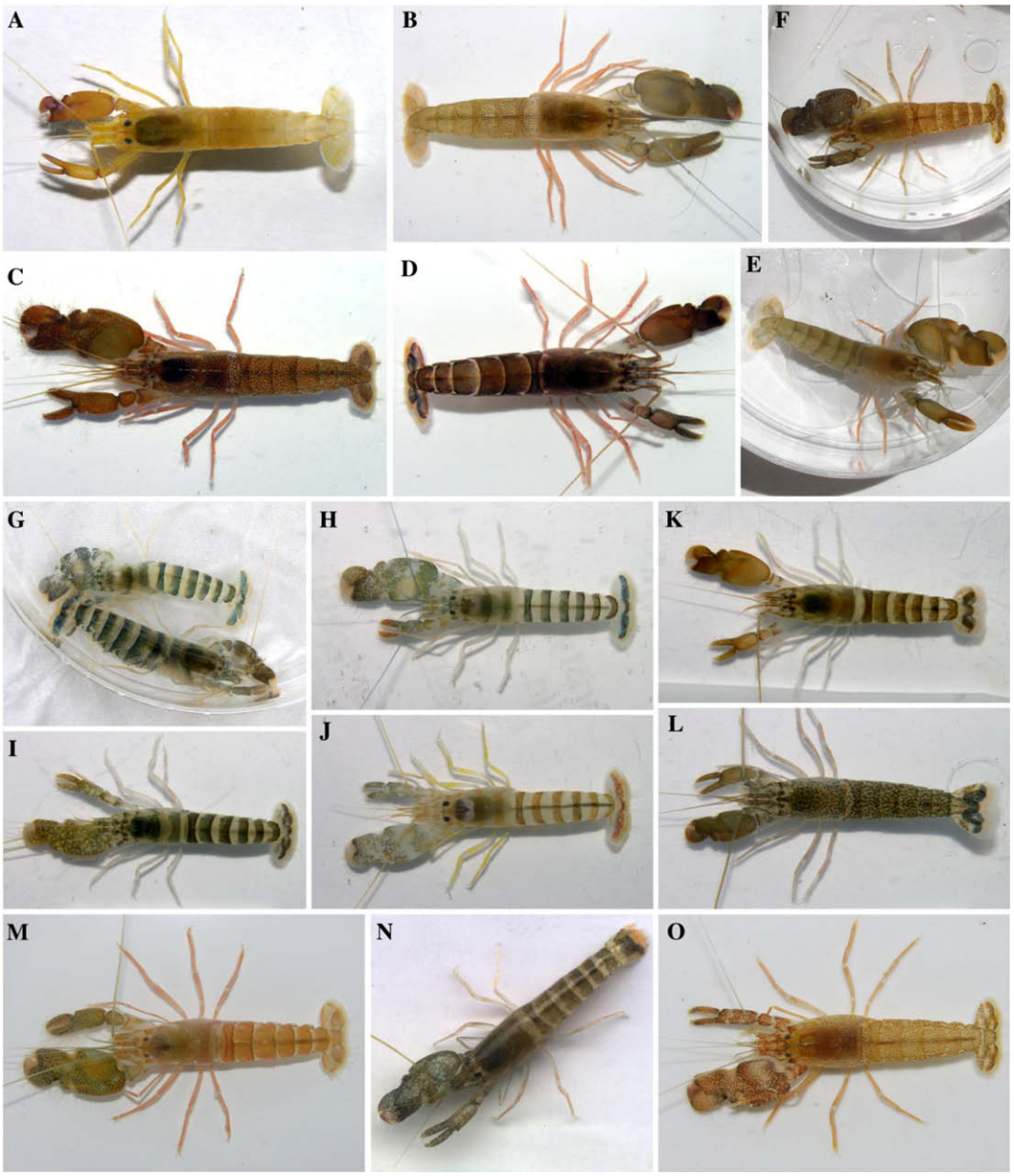

Fig. 2. Representatives of major color pattern types (defined in Table 1) in the *Alpheus armillatus* species complex and the sister clade *A. viridari*. (A) NB1; (B) NB2; (C) NB3; (D) BA8; (E) NB4; (F) BA4; (G) BA3; (H) BA2; (I) BA1; (J) BA3; (K) BA6; (L) NB5; (M) BA7; (N) BA5; (O) NB6.

spots on abdominal somites 2 and 4. For four specimens that were not collected by either A.A. or L.M., color information was not available (specimens were preserved in ethanol, which rapidly degrades color patterns).

2.2. DNA extraction, PCR, and sequencing

Genomic DNA was extracted from claw or abdominal muscle tissue dissected from fresh, ethanol-preserved or frozen shrimp using the Puregene DNA extraction kit (Gentra). From each individual, ~5–10 mg of tissue were removed (for abdominal muscle, the gut was removed to avoid contamination with its contents). Polymerase chain reaction (PCR) was performed on each DNA sample using the primers listed in Appendix A to amplify fragments of the mitochondrial gene cytochrome oxidase I (COI) and the nuclear gene myosin heavy chain (MyHC); to amplify the 16S gene, the primers 16S-1472 (5′-AGATAGAAACCAACCTGG-3′) (Schubart et al., 2000) and 16S-L2 (5′-TGCCTGTTTATCAAAAACAT-3′) (Mathews et al., 2002) were used for all samples. Reactions for each gene were carried out in 20 μL volumes with the following components: 10 ng of genomic DNA, 0.5 μM of each primer, 0.06 mM of each dNTP, 1× Thermopol buffer (New England Biolabs) and 0.5 U of Taq DNA polymerase (New England Biolabs). PCRs were performed with the following conditions: 95 °C for 2 min, followed by 40 cycles of 95 °C for 30 s, annealing temperature (COI: Appendix A; MyHC: 58 °C; 16S: 48 °C) for 30 s, and 72 °C for 60 s, followed by a final extension of 10 min at 72 °C. Reactions were treated with exonuclease I/shrimp alkaline phosphatase. Sequencing reactions were carried out with BigDye v. 3.1 (Applied Biosystems) and the forward or reverse primer. Sequencing reactions were purified by ethanol precipitation and were sequenced directly in both directions on an Applied Biosystems 3730 automated sequencer at the Cornell University Life Sciences Core Laboratories Center or at the DNA Analysis Facility on Science Hill (Yale University).

Additional sequences from a previous study (Mathews, 2006) were obtained from GenBank (Appendix A). These included

16SrRNA sequences used in the earlier study; for the current investigation, additional sequences were obtained from the same individuals, including COI and, for some individuals, MyHC.

For each sequence, both strands were assembled and edited with the SeqMan module of Lasergene v. 6.1 (DNASTAR). For MyHC, heterozygous sites were indicated by double peaks in both forward and reverse sequences. Sequences were individually trimmed to lengths covered by high quality sequence from both strands, and edited sequences were aligned (for each gene separately) with the Clustal W program (Thompson et al., 1994) implemented in BioEdit 7.0.4.1 (Hall, 1999) with default parameters. Alignment of the 16S sequences was refined by eye, and all alignments were trimmed further such that all sequences were of the same length.

2.3. Phylogenetic analyses

For both the mitochondrial and nuclear data sets, the program MrModeltest 2.2 (Nylander, 2004) was used to determine the best-fit nucleotide substitution model. Hierarchical likelihood ratio tests supported a general time reversible model with Γ-distributed rate variation and a proportion of invariant sites (GTR + I + Γ) for each of the mitochondrial genes (transition/transversion ratio: 16S = 5.1261, COI = 6.0623; shape parameter: 16S = 0.4337, COI = 0.7057; proportion of invariant sites: 16S = 0.3567, COI = 0.5042) and an HKY model with Γ-distributed rate variation and a proportion of invariant sites (HKY + I + Γ) for the nuclear gene MyHC (transition/transversion ratio = 4.4565, shape parameter = 0.2514, proportion of invariant sites = 0.6748). Tests of partition homogeneity carried out in PAUP* 4.0b10 (Swofford, 2003) indicated no evidence for partition incongruence for either the two mitochondrial genes ($p = 0.01$) or for all three genes ($p = 0.71$), according to the recommendations of Cunningham (1997), which indicate that combining datasets decreases phylogenetic accuracy at $p < 0.001$. Pairwise genetic distances between each of the major mtDNA clades were estimated in PAUP*.

Phylogenetic analyses were carried out using both Bayesian and Maximum Likelihood (ML) methods on three datasets: the combined mtDNA genes, the nuclear MyHC gene, and the combined set of all three genes. For all analyses of the mtDNA genes, sequence from *A. viridari*, a putative sister taxon to the *A. armillatus* complex, was used as an outgroup. For analyses of the MyHC gene, sequence from *A. lottini* (AJ493186) was used as an outgroup. Maximum Likelihood analyses were carried out in the program TREEFINDER (Jobb et al., 2004) under the best-fit model of nucleotide substitution for each gene, with confidence estimated with 1000 bootstrap repetitions.

Bayesian analyses were carried out in BEAST v1.4.7 (Drummond and Rambaut, 2007) under the best-fit model of nucleotide substitution for each gene and an uncorrelated lognormal relaxed clock, which allows independent rates of nucleotide substitution on different branches. All analyses in BEAST were carried out with a Yule tree prior, and were run for 10 million generations, with sampling every 1000 generations. The first one million runs were discarded as burn-in. The program TRACER v.1.4 (Rambaut and Drummond, 2007) was used to assess convergence and mixing of chains and to ensure that the effective sample sizes (ESS) were >100 for all parameters.

For the combined mtDNA dataset, divergence times were estimated by using the least divergent transisthmian species pair (specimens 7 and 8, based on genetic distance estimates of 0.1086 for COI and 0.04262 for 16S) as a calibration point. This analysis assumed that these two lineages diverged at the time of the closure of the Isthmus of Panama, ~3 MYA. In BEAST, we used a normal prior of 3 MYA for this node, yielding mitochondrial trees for which branch lengths are measured in estimated divergence time. A default mean substitution rate of 1.0 was applied to all analyses incorporating the nuclear sequence data from MyHC, yielding branch length measurements in units of substitutions/site.

In addition, the program BEAST was used to test hypotheses on the evolution of color patterns in this species complex. Specifically, we hypothesized that color patterns are consistent with phylogenetic relationships within the species complex, such that lineages with shared color patterns form monophyletic clades. For this analysis, we considered only two color characteristics: the absence or presence of (1) abdominal banding patterns and (2) paired lateral spots on abdominal somites 2 and 4. Though the intralineage variability in these two color characteristics has not been empirically investigated, our sampling of this taxon indicates that these two traits are likely to be consistent within lineages (see e.g., Mathews, 2006). According to these two color characteristics, our sampling of the species complex includes three pattern categories (banded/spotted; banded/unspotted, and unbanded/unspotted; Table 1). We tested two models that could account for these three color categories with the minimum of color change steps (Fig. 3). In model 1, lineages that share the abdominal banding pattern form a monophyletic clade, with a nested monophyletic clade characterized by abdominal spots in addition to the banding pattern. In model 2, lineages that share the unspotted phenotype are monophyletic, with a nested monophyletic clade characterized by the absence of abdominal banding. Both of these models require only 2 color change steps; however, we note that specimens of *A. viridari*, a sister taxon to the *A. armillatus* complex, have neither abdominal banding nor spotting patterns, and therefore this may represent the ancestral color pattern for *A. armillatus*. These two models were compared to a model with no constraints on color pattern evolution. For these three analyses, four specimens for which color information was lacking were excluded (Fig. 4). Models were compared using Bayes factors, estimated as the difference in the harmonic means of the posterior likelihoods (calculated in TRACER). The sig-

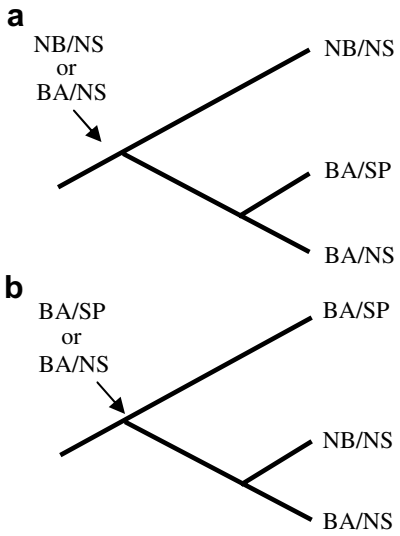

Fig. 3. Alternative models for color pattern evolution among members of the *A. armillatus* complex included in this investigation that were evaluated in BEAST. Three color types are considered based on the presence (BA) or absence (NB) of transverse bands and the presence (SP) or absence (NS) of paired lateral spots on abdominal somites 2 and 4. The NB/NS group included color patterns NB1-5; the BA/NS group included color patterns BA1-3, and BA6; the BA/SP group included color patterns BA4-5 and BA8. (a) In model 1, the banded color phenotype is monophyletic, and the spotted phenotype is nested in the banded clade; (b) in model 2, the unspotted color phenotype is monophyletic, and the unbanded phenotype is nested in the unspotted clade. Both of these two models require a minimum number of changes in these two color characteristics ($n = 2$) to account for the three phenotypes that occurred in our collection.

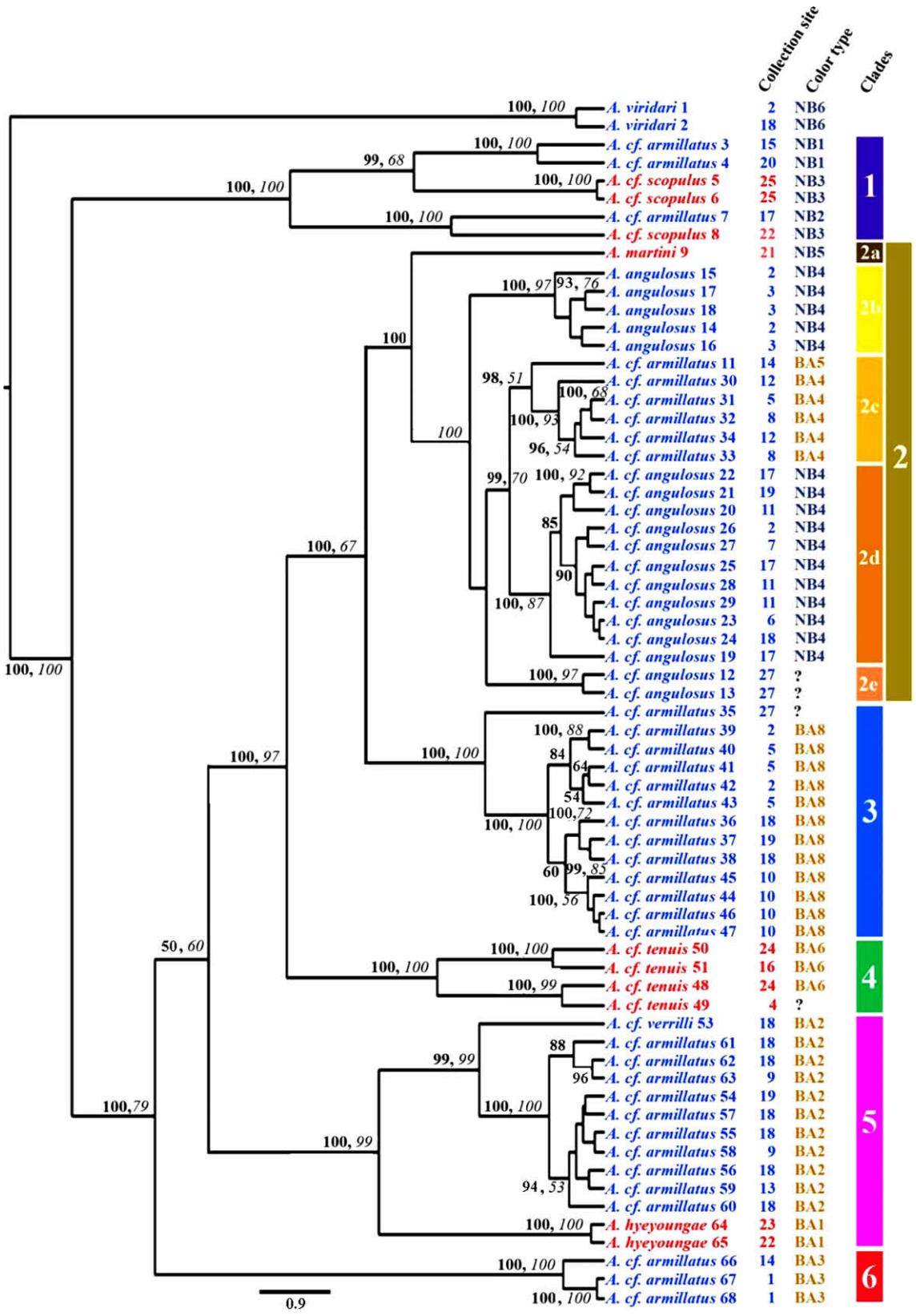

Fig. 4. Bayesian phylogenetic tree (maximum clade credibility) from combined 16S and COI sequence data for known and unidentified members of the *Alpheus armillatus* species complex. Taxa in red text are Eastern Pacific specimens; taxa in blue text are Western Atlantic specimens. Numbers immediately following each terminal taxon are specimen numbers listed in Appendix A. Numbers in the left column are collection sites (red: Eastern Pacific, blue: Western Atlantic. The letters (BA for banded or NB for non-banded) following by numbers in second column are codes of color patterns; specimens with no available color information are denoted with a question mark. Numbers next to nodes indicate Bayesian posterior probabilities (in bold) or bootstrap support (in italics) from the ML analysis. Only posterior probabilities or bootstrap values $\geqslant$50 are shown. Scale bar represents time before present in MY. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

nificance of Bayes factors was evaluated according to the guidelines of Kass and Raftery (1995).

3. Results

Sequences from both mitochondrial genes were obtained from a total of 67 individual shrimp specimens; this included 515 base pairs (bp) of the 16S gene, and 533 bp of the COI gene. For an additional 2 specimens, sequence data from the COI gene either could not be amplified with any primer combination (*A. cf. armillatus* 10, collected from site 20), or was considered a likely pseudogene (reportedly common for COI in *Alpheus*: Williams and Knowlton, 2001), because of multiple non-synonymous substitutions (*A. cf. armillatus* 52, collected from site 18). These two specimens were excluded from the overall mitochondrial phylogeny (Fig. 4) but were included in separate analyses of the 16S gene alone (carried out under the same conditions as for the combined data set; not shown), which were nearly identical in topology to the combined analyses.

Sequence from the nuclear gene MyHC (285 bp) was obtained from 26 individuals, including one individual from each clade identified in the mtDNA analyses (this gene was substantially less variable than the mitochondrial genes, and therefore was used for inferences into deeper divisions among major clades only). Examination of the COI sequences revealed multiple non-synonymous substitutions in one sequence (for *A. cf. armillatus* 52); because COI pseudogenes have been reported in *Alpheus* (Williams and Knowlton, 2001), this sequence was considered to be a likely pseudogene, and the individual was excluded from further phylogenetic analysis. This left a final sample of 66 individuals in the mtDNA analyses and 27 individuals in the MyHC and three-gene analyses.

The Bayesian analyses on the mtDNA data set revealed the presence of six major clades, hereafter Clades 1–6 (Fig. 4 and Table 2); in addition, the analysis showed substantial genetic substructure within these six clades, with >20 mitochondrial lineages represented among these individuals. Some of these lineages correspond to described species, based on morphology and collecting location; these species are listed with species names (e.g., *A. hyeyoungae*) and are discussed below. Other lineages apparently represent undescribed cryptic species, with very subtle or no obvious morphological differences. There was substantial overlap in the geographic distributions of clades (Fig. 4), therefore providing no evidence for allopatry in the current ranges of evolutionarily divergent lineages. The analysis supports the occurrence of at least five separate transisthmian divergences in the *A. armillatus* complex (Fig. 4); based on mitochondrial gene sequences, however, only one lineage (Clade 1) included two transisthmian sibling species pairs: *A. cf. armillatus* 3–4/*A. cf. scopulus* 5–6, and *A. cf. armillatus* 7/*A. cf. scopulus* 8 (Fig. 4). The ML analysis on the combined mtDNA dataset (log likelihood = −7017.28) provided the same topology as the Bayesian analysis with respect to the six major clades, except that the ML analysis placed *A. martini* 9 as a lineage basal to both Clades 2 and 3, while the Bayesian analysis placed this lineage as basal within the Clade 2.

Phylogenetic analysis of the nuclear sequences (Fig. 5) recapitulated some of the relationships supported in the mtDNA analysis. The Bayesian analysis provided moderate or strong support for all of the major clades, except that it failed to resolve the relationship between lineages falling into mtDNA Clades 2 and 3; the ML analysis (log likelihood = −658.58) supported Clades 3, 4, and 6 with moderate to strong support, but provided poor resolution for the other lineages. Bayesian and ML (log likelihood = −5714.25) analyses of the dataset including all three genes ($N = 26$ individuals) yielded topologies among the six major clades that were identical to that of the combined 16S/COI dataset, and are not shown.

Divergence time estimates from analysis of the mtDNA sequences in BEAST suggests that the species complex has been radiating for the last ~10 MY, with continued extensive diversification within the last 3 MY, following the final closure of the Isthmus of Panama. Though our collections are primarily from Atlantic and Caribbean locations, one Eastern Pacific clade (Clade 4) shows evidence for ongoing diversification in the Pacific basin; in this clade, two deeply divergent clades appear to share several morphological characteristics of *A. tenuis* and likely represent different species.

Comparisons of alternative models for color pattern evolution (Fig. 3) revealed strong support for the unconstrained model (harmonic mean = −6165.758) over either model 1 (harmonic mean = −6244.392, 2 $\log_e[B_{10}] = 157.27$) or model 2 (harmonic mean = −6244.129, 2 $\log_e[B_{10}] = 156.742$).

Table 2
Pairwise mean genetic distances between *A. armillatus* mtDNA clades under nucleotide substitution model GTR + I + Γ. COI is above the diagonal, 16S is below the diagonal.

Clade	1	2	3	4	5	6
1	—	0.2458	0.2484	0.2656	0.2802	0.2316
2	0.2065	—	0.1253	0.1992	0.2473	0.2361
3	0.2168	0.0618	—	0.2115	0.2487	0.2361
4	0.1848	0.0999	0.1151	—	0.2595	0.2434
5	0.1756	0.1077	0.1221	0.1127	—	0.2216
6	0.2022	0.1226	0.1498	0.1185	0.1447	—

4. Discussion

Many taxonomic groups, particularly among marine animals, are known to harbor cryptic biodiversity in the form of divergent lineages, often with undetected undescribed species, but these phenomena remain relatively unstudied. Our large-scale investigation of the evolutionary relationships in the *A. armillatus* species complex presents a clear evidence for extensive cryptic biodiversity, with ancient and ongoing radiations over a substantial part of the geographic range of this taxon.

The mitochondrial data set indicates that the morphological species complex is subdivided into at least six major clades, which have likely diverged around ~10 MYA. We caution, however, that divergence time estimates are based on the assumption that the least divergent transisthmian species pair in our data set diverged ~3 MYA, in association with the final closure of the Isthmus of Panama. However, Knowlton and Weigt (1998) reported that many transisthmian divergences in *Alpheus* predate the final closure of the Isthmus of Panama, and therefore our divergence time estimates may well be biased downwards.

Our data also show extensive substructure within each of the six major clades. Furthermore, the complex includes five transisthmian separations (Fig. 4), providing strong additional evidence that the diversification of this complex began well before the closure of the Isthmus of Panama. These observations are consistent with those of other investigations. For example, Knowlton and Weigt (1998) reported that divergences between most transisthmian sibling lineages of *Alpheus* began prior to the final closure of the Isthmus, rather than in association with this event. In addition, Anker et al. (2007a,b, 2008a,b) revised four transisthmian species complexes of *Alpheus*, in which the estimated divergence times ranged from 6 to 12 MYA, well before the closure of the Isthmus. This analysis also provides insight into the evolution of color patterns in this species complex. While distinctive color patterns are known to be useful in distinguishing nearly cryptic snapping shrimp lineages (Knowlton and Mills, 1992; Williams et al., 2001), our data suggest that changes in color patterns have

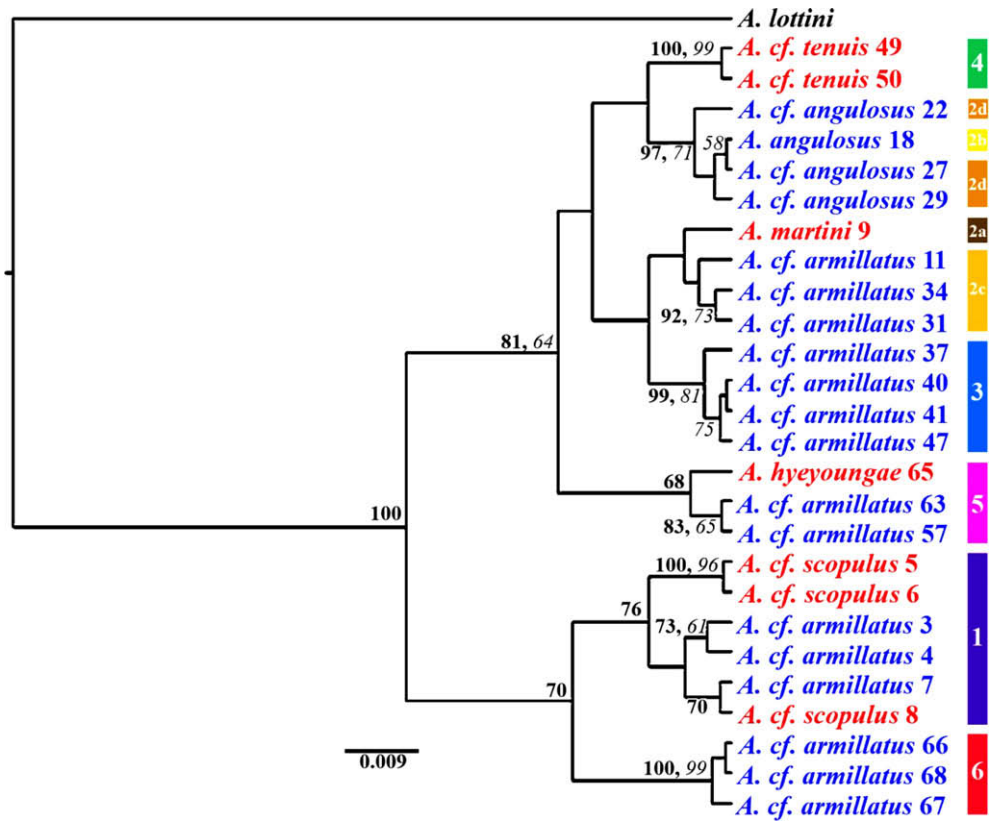

Fig. 5. Bayesian phylogenetic tree (maximum clade credibility) from MyHC sequence data for known and unidentified members of the *Alpheus armillatus* species complex. Taxa in red text are Eastern Pacific specimens; taxa in blue text are Western Atlantic specimens. Numbers following each terminal taxon are specimen numbers listed in Appendix A. Numbers next to nodes indicate Bayesian posterior probabilities (in bold) or bootstrap support (in italics) from the ML analysis. Only posterior probabilities or bootstrap values ⩾50 are shown. Clade numbers are from corresponding mtDNA clade (Fig. 3). Scale bar represents nucleotide substitutions. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

occurred multiple times in this species complex. Bayesian analyses in BEAST did not support either of two simple evolutionary models (Fig. 3) for the evolution of two color characteristics (abdominal banding and spotting), each of which minimizes the number of color change steps among the known members of the complex. Rather, the model with no constraints on the evolution of color patterns was strongly preferred (evaluated by Bayes factors). The mtDNA phylogeny supports at least two origins of the spotted phenotype (in Clades 2 and 3), while the abdominal banding phenotype may have been secondarily lost in the unbanded members of Clade 2. Shifts in these color characteristics, therefore, may be genetically simple and selectively neutral.

While these data indicate much ancient diversification in this species complex, they also provide evidence for substantial recent (<3 MY) diversification within ocean basins. In the *A. armillatus* complex, substructure within each of the major clades shows that evolutionary diversification continued within each basin following the closure of the Isthmus. On the whole, this species complex has undergone a remarkable radiation with comparative morphological stasis. Whether these patterns are a unique characteristic of one or a few taxa within *Alpheus*, or are representative of the genus as a whole, remains to be investigated. Knowlton and Weigt (1998) suggested that, to their knowledge, the *A. armillatus* complex appears to include a greater number of potential cryptic species than some other complexes; additional analyses of other species complexes in the genus, as well as in the closely related genus *Synalpheus*, will help shed more light on the patterns of cryptic radiations in snapping shrimps.

In general, the nuclear sequence data confirmed the separation of the complex into six clades, though the less variable nuclear dataset showed much poorer resolution, and provided strong support for only several of the six mtDNA clades. Consequently, our major inferences are based primarily on the mitochondrial genome. Below, we discuss each of the major six clades in more detail, including potentially diagnostic morphological characteristics (e.g., color patterns and the shape of the postrostral plate; Fig. 6).

Clade 1 is a very distinctive transisthmian clade, characterized by generally similar non-banded color patterns (types NB1-3, Fig. 2A–C), and by a sharply delimited, not elevated, U- or V-shaped postrostral plate (Fig. 6C; see also figures of *A. scopulus* in Kim and Abele, 1988). In addition, all species of Clade 1 live in lower intertidal and subtidal habitats. For instance, the Caribbean *A. cf. armillatus* 3 and 4 and the Eastern Pacific *A. cf. scopulus* 5 and 6 were collected at 10–20 m, while individuals of the Western Atlantic species represented by *A. cf. armillatus* 7 have been collected at 1–3 m on seagrass-rubble beds (A. Anker, personal observation). Morphological differences (e.g., shape of the postrostral plate and dorsal notch of the major claw) and color patterns support the genetic separation of *A. cf. armillatus* 3–4, *A. cf. scopulus* 5–6, and *A. cf. armillatus* 7. On the other hand, two Eastern Pacific lineages divergent in both mitochondrial and nuclear genes (represented by *A. cf. scopulus* 5–6 and *A. cf. scopulus* 8), appear to be similar in color pattern and morphology (A. Anker, personal observation). Investigation of additional specimens and populations should provide more insight into the relation between these two lineages.

Clade 2 is genetically the most diverse clade and is also the only of the six major clades with three sharply different color pattern types. This clade comprises primarily Western Atlantic taxa; its single Eastern Pacific representative and a possible sister group to the Western Atlantic lineages (Clade 2a in Fig. 4) is *A. martini*,

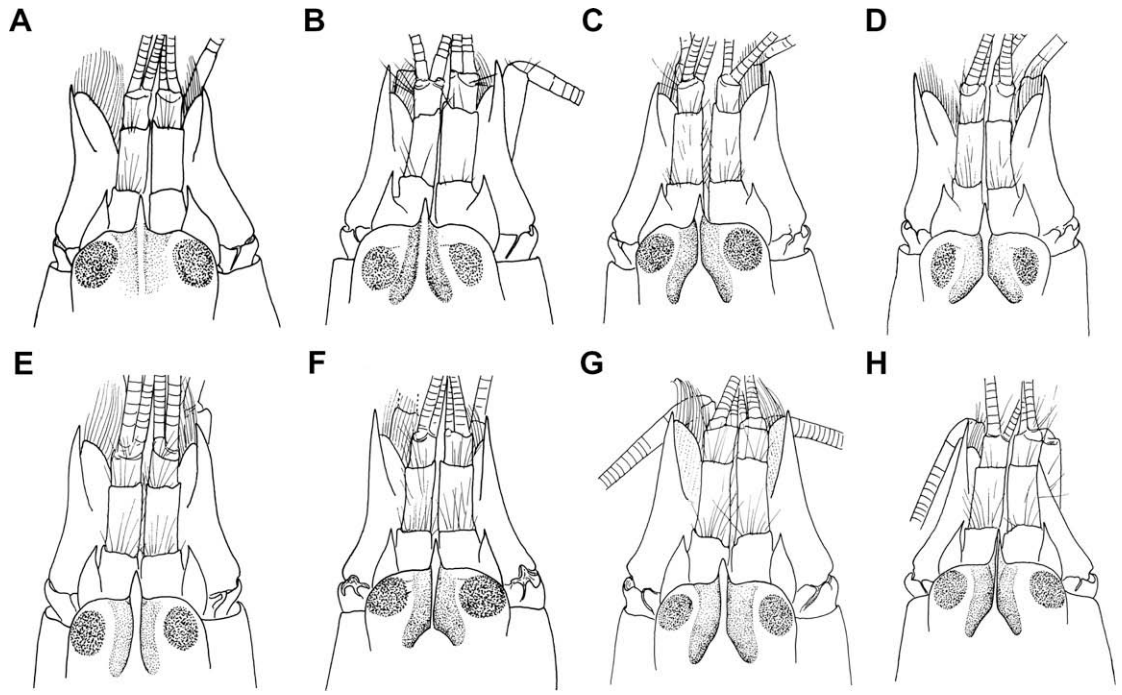

Fig. 6. Morphological variation in the shape of the flattened postrostral plate on the frontal region of the carapace. Species names refer to Table 3 (clade numbers to Fig. 3). (A) *A. viridari* from Guadeloupe; (B) *A. cf. angulosus* A2 from Atol das Rocas, Brazil (Clade 2a); (C) *A. cf. armillatus* D from Florida Keys (Clade 1); (D) *A. cf. armillatus* B from Cubagua, Venezuela (Clade 6); (E) *A. cf. angulosus* A3 from Playa Girón, Cuba (Clade 2a); (F) *A. cf. verrilli* from Florida Keys (Clade 5); (G) *A. cf. armillatus* F from Indian River Lagoon, Florida (Clade 2b); (H) *A. cf. armillatus* A from Morrocoy, Venezuela (Clade 5).

though this species has a unique color pattern (type NB5, Fig. 2L) different from the rest of Clade 2. At least some members of Clade 2 represent *A. angulosus* sensu stricto (sensu McClure, 1995). To our best knowledge, the clade represented by specimens *A. angulosus* 14–18 is the only lineage ever collected from the Gulf of Mexico (see also Mathews, 2006, 2007), which includes the type locality for *A. angulosus* (McClure, 1995); in addition the uniformly brown and unspotted color pattern of these individuals (type NB4, Fig. 2E) matches the description for that species. Furthermore, two previous investigations (Mathews, 2006, 2007) showed evidence for homogeneity in nuclear genomes between individuals falling into two separate subclades within Clade 2 (Clades 2b and d in Fig. 4) that are characterized by an unbanded, unspotted phenotype, and we presently consider that both of these subclades represent *A. angulosus*. On the other hand, Clades 2b and d are genetically differentiated and apparently reproductively isolated from the lineage represented by Clade 2c (see Fig. 4). Most members of the Clade 2c have a unique banded-spotted color patterns (type BA4, see Fig. 2F) that is markedly different from that of Clades 2b and d. In Mathews et al. (2002), the lineage 2c was treated as *A. armillatus*, but we currently consider it to be an undescribed species (see Table 3). The identity of *A. cf. armillatus* 11 from Venezuela, which is genetically somewhat divergent from the rest of Clade 2c and has a slightly different color pattern (type BA5, see Fig. 2N), is presently not clear (see Table 3).

Clade 2 also includes two specimens from Brazil (*A. cf. angulosus* 12 and 13), which share the morphology and (according to brief field notes on those specimens) the color pattern characteristics of Clades 2b and d. These two specimens formed a small subclade that is quite divergent from the Caribbean, Gulf of Mexico, and Atlantic members of the Clade 2. This may represent strong geographic structure within *A. angulosus*, since those two specimens were collected from a site separated by both a large geographic distance and by potentially inappropriate habitat (the Amazon River outflow, which may form a natural barrier to migration; see Rocha et al., 2002). However, thorough sampling in regions between the Caribbean sites and the Brazilian site may include genetic intermediates. In general, *A. angulosus* may be the most geographically widespread member of the complex (at least among its Western Atlantic representatives) and is an ecologically variable species, found in mudflats with rocks in Texas (McClure, 1995), seagrass beds with coral rubble in Florida and Panama (A. Anker, personal observation), and areas with a mixed rock and sand substrate (L. Mathews, personal observation).

Clade 3 is a purely Western Atlantic clade with a distinct color pattern (type BA8, Fig. 2D), ranging from Florida to Panama and Brazil, and is genetically strongly subdivided by geographic regions (Fig. 4). We detected three subclades representing different collecting regions; specimens 36–38 were collected from Panama, specimens 39–43 from Florida, and specimens 44–47 from Puerto Rico. In addition, this clade includes a more divergent lineage, represented by *A. cf. armillatus* 35, collected from Atol das Rocas in Brazil. This topology is reminiscent of that of *A. cf. angulosus* in Clades 2a and b, which showed an evolutionary division among Brazilian, Caribbean, and Gulf of Mexico populations. However, no specimens were available from any sites between the broadly separated Caribbean sites and the Brazilian site; therefore, it remains unknown whether genetically intermediate or admixed populations may exist.

Clade 4 is a purely Eastern Pacific clade distinguished by color pattern (type BA6, Fig. 2K) and by a feebly marked, V-shaped postrostral plate (see figures of *A. tenuis* in Kim and Abele, 1988). This group probably contains at least two species, as four specimens preliminarily identified as *A. cf. tenuis* fell into two deeply separated mtDNA subclades; however, the subclades were not divergent in the nuclear data. The identities of *A. cf. tenuis* 48–49 and *A. cf. tenuis* 50–51 will be clarified through a careful comparison with the type specimens of *A. tenuis*, as well as additional geographic and genetic sampling.

Clade 5 is a well supported transisthmian clade, distinguished by similar color patterns (types BA1–2, BA7, see Fig. 2H, I, and M) and by a broad, sharply delimited, elevated, U-shaped postrostral plate (Fig. 6F and H; see also figures of *A. hyeyoungae* in Kim and

Table 3
Taxonomic summary of known members of the *A. armillatus* species complex and *A. viridari*. Geographic distribution is based on presently available published and unpublished (A. Anker, personal observation) data. Specimen and clade numbers refer to Figs. 4 and 5; color types refer to Table 1 (see also Fig. 2). *Abbreviations used:* NA, specimens non-available; SC, sister clade to *A. armillatus* sensu lato.

Putative species	Taxonomic status	Specimen #	Clade	Color type	Distribution
I. Western Atlantic taxa					
A. cf. armillatus A	To be fixed as *A. armillatus* Milne Edwards (=*A. armillatus* sensu stricto)	54–63	5	BA2	Honduras, Panama, Costa Rica, Venezuela, French and Dutch Antilles, Florida
A. cf. armillatus B	Possibly *A. lancirostris* Rankin	66–68	6	BA3	Bermuda, Venezuela
A. cf. armillatus C	Undescribed species	3–4	1	NB1	Panama, Honduras, Florida Keys
A. cf. armillatus D	Undescribed species	7	1	NB2	Panama, Costa Rica, Honduras, Florida Keys
A. cf. armillatus E	To be investigated, either undescribed species or same as *A. cf. armillatus* F	11	2c	BA5	Venezuela, Brazil
A. cf. armillatus F	Undescribed species; same as clade 2 in Mathews (2006)	30–34	2c	BA4	Florida (Indian River, Florida Keys), Panama, Jamaica
A. cf. armillatus H1	Undescribed species; same as clade 4 in Mathews (2006)	36–47	3	BA8	Florida (Indian River, Florida Keys), Panama
A. cf. armillatus H2	To be investigated, possibly same as *A. cf. armillatus* H1	35	3	?	NE Brazil (Atol das Rocas)
A. cf. armillatus I	To be investigated, known only from 1 juvenile specimen	53	5	BA2	Panama (Bocas del Toro)
A. cf. armillatus J	To be investigated	10*	5?	BA3	Panama (San Blas)
A. cf. angulosus A1	*A. angulosus* McClure; same as clade 3 in Mathews (2006)	14–18	2b	NB4	Gulf coast of Florida probably to Texas
A. cf. angulosus A2	To be investigated, possibly same as *A. cf. angulosus* A1	12–13	2e	?	NE Brazil (Atol das Rocas)
A. cf. angulosus A3	To be investigated, perhaps same as *A. cf. angulosus* A1; same as clade 1 in Mathews (2006)	19–29	2d	NB4	Panama, Costa Rica, Cuba, Honduras, Puerto Rico, Jamaica, Florida
A. cf. verrilli	To be investigated; probably *A. verrilli* (Schmitt)	52*	3 or 5?	BA7	Florida Keys, Panama, Venezuela, Barbados
A. viridari	*A. viridari* (Armstrong)	1–2	SC	NB6	Panama, Costa Rica, Honduras, Dominican Republic, Venezuela, Guadeloupe, Florida
II. Eastern Pacific taxa					
A. cf. scopulus A	*A. scopulus* Kim and Abele	8	1	NB3	Panama, Galapagos
A. cf. scopulus B	Undescribed species	5–6	1	NB3	Panama (Taboga Island)
A. hyeyoungae	*A. hyeyoungae* Kim and Abele	64–65	5	BA1	Panama, Costa Rica, Gulf of California
A. martini	*A. martini* Kim and Abele	9	2a	NB5	Panama
A. cf. tenuis A	Possibly *A. tenuis* Kim and Abele	48–49	4	BA6	Panama, Gulf of California
A. cf. tenuis B	Possibly undescribed species	50–51	4	BA6	Costa Rica, Panama, Gulf of California
A. wickstenae	*A. wickstenae* Christoffersen and Ramos	NA	—	NB, no details	Colombia (Malaga Bay)
A. agrogon	*A. agrogon* Ramos, presently known only from holotype	NA	—	Unknown	Colombia (Gorgona Island)

* No COI data were available; phylogenetic affinities based on 16S sequence only (see text).

Abele, 1988). This clade includes *A. hyeyoungae* and a genetically substructured clade that appears to include two closely related species, represented by *A. cf. armillatus* 54–63 and *A. cf. armillatus* 53, which is unfortunately a juvenile specimen. Because of its wide distribution and conspicuously banded color pattern, we believe that the clade represented by *A. cf. armillatus* 54–63 may be the best taxon to be referred to as *A. armillatus* sensu stricto (A. Anker, personal observation).

For two specimens, COI data were unavailable. One specimen, *A. cf. verrilli* 52, was excluded from Fig. 4 because this specimen yielded a likely COI pseudogene. In analyses including only the 16S data set, this specimen was basal in clade 5 (posterior probability = 100, bootstrap = 87) and its divergent position from the other members of the clade, along with striking color differences between this specimen (Fig. 2M) and the rest of the clade (Fig. 2H), indicate that this specimen may represent a distinct lineage. For another taxon, *A. cf. armillatus* 10 from San Blas, Panama (color type BA3, Fig. 2J), sequence from the COI gene was unavailable because of failed amplifications, but in the 16S dataset, this taxon grouped together with Clade 3 (posterior probability = 100, bootstrap = 100), while in the MyHC dataset, this taxon grouped weakly with Clade 5 (posterior probability = 63, not shown), with which it shares a similar color pattern (Fig. 2H). The relationship of this taxon to the rest of the complex, therefore, remains unclear.

Clade 6 is a purely Western Atlantic clade, characterized by color pattern (type BA3, Fig. 2G) and a broadly V-shaped, well-delimited postrostral plate (Fig. 6D). This clade probably represents a single species ranging from Bermuda to the southern Caribbean, and may correspond to *A. lancirostris* of Rankin (1900), originally described from Bermuda.

Finally, the Western Atlantic *A. viridari* may be a sister clade of the *A. armillatus* complex, as suggested by the combined mitochondrial data set (Fig. 4). This is corroborated by its overall similarity in color pattern (Fig. 2O) and morphology, except for the absence of a flattened postrostral plate (Fig. 6A).

Thus, our data support the presence of at least 11 genetic lineages in the Western Atlantic (see Table 3), separated by subtle differences in morphology and color patterns. At least four of these lineages may represent undescribed species, whereas two species currently placed in synonymy of *A. armillatus* will need to be taxonomically resurrected. Similarly, in the Eastern Pacific, six distinct lineages were identified by the DNA analysis, and corroborated by color patterns and morphological characters, including two possibly undescribed species (Table 3). Two additional Eastern Pacific lineages, *A. wickstenae* and *A. agrogon*, were unavailable for inclusion in this investigation, but are likely members of the complex based on morphological similarity; these taxa bring the number of known complex members in the Eastern Pacific to a total of eight, and the total number of known putative species in the *A. armillatus* complex to at least 19. This biodiversity is surprising, considering that only a few decades ago all specimens with the most obvious morphological features of *A. armillatus*, and from both sides of the Americas, were lumped together under the same species (Wicksten, 1983). Additionally, the complex remains

poorly known over much of both coasts of South America (Colombia, Ecuador, Brazil), and the Pacific coast of Mexico, which may harbor more unidentified lineages.

Preliminary consideration of the geographic distribution of complex members indicates that biodiversity is unlikely to be randomly distributed over the known range of the complex. For example, in the Western Atlantic, two sites (Bocas del Toro, Panama and the Florida Keys) each harbor at least seven species (Table 3), while only one species is known from either the Gulf of Mexico (*A. angulosus*) or Bermuda (*A. lancirostrus*). However, the geographic distributions of most lineages in the complex are poorly known, with collections from only one or a few sites, and therefore broader sampling is required for robust inferences into the phylogeography of this complex.

Our data indicate that divergences in the species complex have been ongoing for the last ~10 MY, with deeper and older divergences separating the complex into putative species, and more recent divergences potentially reflecting genetic structure, with or without geographic association, within some species. Our age estimates are subject to at least two sources of error; first, the calibration point for the least divergent transisthmian species pair was based on the assumption that these taxa split in association with the closure of the Isthmus of Panama, and therefore our age estimates are likely to underestimate true node ages. Secondly, because genetic divergence precedes species or population divergence (Arbogast et al., 2002), and this discrepancy is greater for younger divergences than for older ones (because the overestimate represents a greater fraction of the total divergence time: Knowles and Maddison, 2002), our divergence time estimates for the youngest lineages may be biased upwards.

Interestingly, another comprehensive investigation of cryptic biodiversity in a Caribbean mussel, the *Brachidontes exustus* species complex (Lee and Ó Foighil, 2005), also revealed evidence for a number of cryptic divergences in the same geographic area, but showed primarily allopatric ranges for cryptic lineages. Our data for the *A. armillatus* complex indicate that even deeply divergent clades are relatively widespread over the sampled region, with substantial geographic overlap. This may reflect either divergences in sympatry or substantial range changes following allopatric divergences, resulting in coexisting sister species that are nearly identical morphologically and with a strong potential for ecological overlap. Taylor and Hellberg (2005) reported similar patterns in tropical cleaner gobies (*Elacatinus*), where some sister taxa had a substantial geographic overlap. In that study, the authors argued that in gobies, ecological specialization may have driven evolutionary divergences. In the *A. armillatus* complex, we have observed some ecological differences among the deeply divergent clades. For instance, species of Clade 1 tend to occur subtidally in reef habitats or rocky shores; species of Clades 2–4 mostly occur in intertidal habitat of mixed mud or sand and rocks, sometimes close to estuaries or mangroves; species of Clades 5 and 6 are usually found in coral reef habitats, rocky shores, or on seagrass-rubble fields that are close to reefs. However, the ecological relationships among the major clades and among subclades are not yet clearly delineated.

Among marine animals, particularly those with dispersive larvae, there is ongoing discussion over the relative importance of historical biogeography (e.g., transient allopatry driven by geographic changes followed by, in some cases, secondary contact) and current connectivity (e.g., the realized "openness" to gene flow in apparently open systems). Historical processes, such as the dynamics surrounding the closure of the Isthmus of Panama or sea level changes during the Pleistocene, have long been considered likely drivers of evolutionary diversification in marine taxa (Avise, 1992; Knowlton and Weigt, 1998; Williams and Benzie, 1998; Lessios et al., 2001; Wares and Cunningham, 2001). Others have pointed to evidence for surprisingly low connectivity among populations, even in taxa with apparently dispersing larvae (Swearer et al., 1999, 2002; Cowen et al., 2000; Warner and Cowen, 2002), and in some cases, ecological factors can shape the dispersal of organisms, leading to phylogeographic structure (Riginos and Nachman, 2001; Rocha et al., 2002, 2004. For example, interspecific differences in physiology and habitat preferences may have contributed to sharp differences in phylogeographic patterns in the penaeids *Farfantepenaeus aztecus and Litopenaeus setiferus* (McMillen-Jackson and Bert, 2003). For the *A. armillatus* complex, the mechanisms driving evolutionary divergence among lineages remain to be explored; additional studies with exhaustive geographic sampling from all major clades and over the entire range (including more specimens from both coasts of South America and the Gulf of California) would elucidate the evolutionary history of this highly diversified group of snapping shrimps.

Acknowledgments

We are grateful to the staffs of the Discovery Bay Marine Laboratory, Tropical Research Laboratory, and the Bermuda Biological Station for Research for logistic support. Additionally, we thank the Government of Bermuda (Permit No. SP050601) and to the National Environment and Planning Agency of Jamaica, and the Autoridad Nacional del Ambiente of Panama for granting collection and exportation permits. This research was funded by a WPI Faculty Advancement in Research grant to L.M., and by a postdoctoral fellowship from Nancy Knowlton (USNM/STRI) to A.A. Tim Page (Griffith University, Brisbane) and a second anonymous reviewer made a number of valuable comments considerably improving the overall quality of the manuscript. A.A. thanks Tim Coles and Sammy De Grave for inviting him to participate in the Operation Wallacea invertebrate survey in Utila, Honduras. We also thank Paulo Young (late of MNRJ) and Christoph Schubart (University of Regensburg) for collection of some specimens.

Appendix A

List of specimens with corresponding amplification information for the COI and MyHC gene fragments, and GenBank Accession Nos. All primers were designed for this study except where references are given.

Specimen	COI primers[a]	COI t_{an}	MyHC primers[b]	MyHC accession	16S accession	COI accession
1	1, 2	52	–	–	DQ682894	FJ528483
2	1, 2	52	–	–	FJ528440	FJ528484
3	1, 2	52	1, 4	FJ528568	FJ528441	FJ528485
4	4, 5	46	3, 4	FJ528569	FJ528442	FJ528486
5	1, 2	52	1, 4	FJ528566	FJ528443	FJ528487
6	1, 2	52	1, 4	FJ528567	FJ528444	FJ528488
7	4, 5	46	1, 5	FJ528570	FJ528445	FJ528489
8	4, 5	46	1, 4	FJ528571	FJ528446	FJ528490
9	1, 2	52	1, 5	FJ528555	FJ528447	FJ528491
10	–	–	2, 4	FJ528576	FJ528481	–
11	1, 2	52	1, 4	FJ528556	FJ528448	FJ528492
12	1, 2	52	–	–	DQ682849	FJ528493
13	4, 5	46	–	–	DQ682853	FJ528494
14	1, 2	52	–	–	DQ682858	FJ528495
15	1, 2	52	–	–	DQ682857	FJ528496
16	1, 2	52	–	–	DQ682860	FJ528497
17	1, 2	52	–	–	FJ528449	FJ528498
18	1, 2	52	3, 7	FJ528552	DQ682917	FJ528499
19	1, 2	52	–	–	FJ528450	FJ528500
20	1, 2	52	–	–	FJ528451	FJ528501

Appendix A (*continued*)

Specimen	COI primers[a]	COI t_{an}	MyHC primers[b]	MyHC accession	16S accession	COI accession
21	1, 2	52	–	–	FJ528452	FJ528502
22	1, 2	52	1, 5	FJ528551	FJ528453	FJ528503
23	1, 2	52	–	–	FJ528454	FJ528504
24	1, 2	52	–	–	DQ682908	FJ528505
25	1, 2	52	–	–	DQ682905	FJ528506
26	1, 2	52	–	–	DQ682910	FJ528507
27	1, 2	52	3, 7	FJ528553	DQ682903	FJ528508
28	1, 2	52		–	DQ682873	FJ528509
29	1, 2	52	3, 7	FJ528554	DQ682862	FJ528510
30	1, 2	52	–	–	FJ528455	FJ528511
31	1, 2	52	3, 7	FJ528557	DQ682869	FJ528512
32	1, 3	53	–	–	DQ682875	FJ528513
33	1, 2	52	–	–	FJ528456	FJ528514
34	1, 2	52	3, 7	FJ528558	FJ528457	FJ528515
35	4, 5	46	–	–	FJ528458	FJ528516
36	4, 5	46	–	–	FJ528459	FJ528517
37	4, 5	46	1, 6	FJ528559	FJ528460	FJ528518
38	4, 5	46	–	–	DQ682886	FJ528519
39	1, 2	52	–	–	DQ682884	FJ528520
40	1, 2	52	3, 7	FJ528560	FJ528461	FJ528521
41	1, 2	52	3, 7	FJ528561	DQ682885	FJ528522
42	1, 2	52	–	–	DQ682882	FJ528523
43	1, 2	52	–	–	DQ682885	FJ528524
44	1, 2	52	–	–	DQ682880	FJ528525
45	1, 2	52	–	–	DQ682881	FJ528526
46	1, 2	52	–	–	FJ528462	FJ528527
47	1, 2	52	3, 7	FJ528562	AF501648	FJ528528
48	1, 2	52	–	–	FJ528463	FJ528529
49	1, 2	52	3, 7	FJ528549	FJ528464	FJ528530
50	1, 2	52	1, 5	FJ528550	FJ528465	FJ528531
51	1, 2	52	–	–	FJ528466	FJ528532
52	–	–	1, 5	FJ528575	FJ528482	–
53	4, 5	46	–	–	FJ528467	FJ528533
54	1, 2	52	–	–	FJ528468	FJ528534
55	1, 2	52	–	–	FJ528469	FJ528535
56	1, 2	52	–	–	FJ528470	FJ528536
57	1, 2	52	1, 5	FJ528565	FJ528471	FJ528537
58	3, 5	46	–	–	FJ528472	FJ528538
59	1, 2	52	–	–	FJ528473	FJ528539
60	1, 2	52	–	–	FJ528474	FJ528540
61	1, 2	52	–	–	FJ528475	FJ528541
62	1, 2	52	–	–	FJ528476	FJ528542
63	1, 2	52	1, 4	FJ528564	FJ528477	FJ528543
64	4, 5	46	–	–	FJ528478	FJ528544
65	4, 5	46	3, 4	FJ528563	FJ528479	FJ528545
66	4, 5	46	3, 7	FJ528574	FJ528480	FJ528546
67	1, 2	52	3, 7	FJ528572	DQ682892	FJ528547
68	1, 2	52	3, 7	FJ528573	DQ682893	FJ528548

[a] 1 = COIf, 5′-CCTGCAGGAGGAGGAGAYCC-3′ (Palumbi et al., 1991); 2 = COIa, 5′-AGTATAAGCGTCTGGGTAGTC-3′ (Palumbi et al., 1991); 3 = armCOIF1, 5′-GAA.TATCGCCGCGGCATT-3′; 4 = armCOIF2, 5′-GGAGTAACGTCGTGGTAT-3′; 5 = armCOIR1, 5′-TATCAACATTTATTYTGATT-3′.

[b] 1 = MyHCF, 5’-GCTCTCGACCATTCCAATA-3′ (Williams et al., 2001); 2 = MyHCF2, 5′-CAGAAGCACATCAAGAAGA-3′; 3 = MyHC-1124, 5′-AAGCTCGAGTCTGACATCA-3′; 4 = MyHCR, 5′-AGGTTGGCATTCTGGGAGG-3′ (Williams et al., 2001); 5 = MyHCR2, 5′-GTTCTTGGCCTCATTCAAC-3′; 6 = MyHCR3, 5′-CCTTGCGCATCTTCTCC TG-3′;7 = MyHC-lottR, 5′-CTTTCTCTTGGTGACGGTG-3′.

References

Anker, A., 2001a. Two new species of snapping shrimps from the Indo-Pacific, with remarks on colour patterns and sibling species in Alpheidae (Crustacea: Caridea). Raffles Bull. Zool. 49, 57–72.

Anker, A., 2001b. Taxonomie et évolution des Alpheidae (Crustacea, Decapoda). Ph.D. thesis, Muséum National d'Histoire Naturelle, Paris. 2 volumes, 547 + 331 pp.

Anker, A., Hurt, C., Knowlton, N., 2007a. Revision of the *Alpheus nuttingi* (Schmitt) species complex, with description of a new species from the tropical eastern Pacific (Crustacea: Decapoda: Alpheidae). Zootaxa 1577, 41–60.

Anker, A., Hurt, C., Knowlton, N., 2007b. Three transisthmian snapping shrimps (Crustacea: Decapoda: Alpheidae: *Alpheus*) associated with innkeeper worms (Echiura: Thalassematidae). Zootaxa 1626, 1–23.

Anker, A., Hurt, C., Knowlton, N., 2008a. Revision of the *Alpheus websteri* Kingsley, 1880 species complex (Crustacea: Decapoda: Alpheidae), with revalidation of *A. arenensis* (Chace, 1937). Zootaxa 1694, 51–68.

Anker, A., Hurt, C., Knowlton, N., 2008b. Revision of the *Alpheus formosus* Gibbes 1850 species complex, with redescription of *A. formosus* and description of a new species from the tropical Western Atlantic (Crustacea: Decapoda: Alpheidae). Zootaxa 1707, 1–22.

Arbogast, B.S., Edwards, S.V., Wakeley, J., Beerli, P., Slowinski, J.B., 2002. Estimating divergence times from molecular data on phylogenetic and population genetic timescales. Ann. Rev. Ecol. Syst. 33, 707–740.

Armstrong, J.C., 1949. New Caridea from the Dominican Republic. Am. Mus. Novitates 1410, 1–27.

Avise, J.C., 1992. Molecular population structure and the biogeographic history of a regional fauna: a case history with lessons for conservation biology. Oikos 63, 62–76.

Baker, J.M., Funch, P., Giribet, G., 2007. Cryptic speciation in the recently discovered American cycliophoran *Symbion americanus*; genetic structure and population expansion. Mar. Biol. 151, 2183–2193.

Baratti, M., Goti, E., Messana, G., 2005. High level of genetic differentiation in the marine isopod *Sphaeroma terebrans* (Crustacea: Isopoda: Sphaeromatidae) as inferred by mitochondrial DNA analysis. J. Exp. Mar. Biol. Ecol. 315, 225–234.

Barber, P.H., Palumbi, S.R., Erdmann, M.V., Moosa, M.K., 2002. Sharp genetic breaks among populations of *Haptosquilla pulchella* (Stomatopoda) indicate limits to larval transport: patterns, causes, and consequences. Mol. Ecol. 11, 659–674.

Baric, S., Sturmbauer, C., 1999. Ecological parallelism and cryptic species in the genus *Ophiothrix* derived from mitochondrial DNA sequences. Mol. Phylogenet. Evol. 11, 157–162.

Bastrop, R., Jürss, K., Sturmbauer, C., 1998. Cryptic species in a marine polychaete and their independent introduction from North America to Europe. Mol. Biol. Evol. 152, 97–103.

Benzie, J.A.H., 1999. Major genetic differences between crown-of thorns starfish (*Acanthaster planci*) populations in the Indian and Pacific Oceans. Evolution 53, 1782–1795.

Bilodeau, A.L., Felder, D.L., Neigel, J.E., 2005. Population structure at two geographic scales in the burrowing crustacean *Callichirusislagrande* Decapoda, Thalassinidea: historical and contemporary barriers to planktonic dispersal. Evolution 59, 2125–2138.

Bruce, A.J., 1999. *Alpheus sorror*, a new snapping shrimp cryptospecies from Sri Lanka Crustacea: Decapoda: Alpheidae. Raffles Bull. Zool. 47, 453–463.

Caputi, L., Andreakis, N., Mastrototaro, F., Cirino, P., Vassillo, M., Sordino, P., 2007. Cryptic speciation in a model invertebrate chordate. Proc. Natl. Acad. Sci. USA 104, 9364–9369.

Chace, F.A., 1972. The shrimps of the Smithsonian-Bredin Caribbean expeditions with a summary of the West Indian shallow water species (Crustacea: Decapoda: Natantia). Smith. Contrib. Zool. 98, 1–179.

Chace, F.A., 1988. The caridean shrimps (Crustacea: Decapoda) of the *Albatross* Philippine expedition, 1907–10. Part 5: family Alpheidae. Smith. Contrib. Zool. 466, 1–99.

Christoffersen, M.L., Ramos, G.E., 1987. A new snapping shrimp (Caridea, Alpheidae) from the Pacific coast of Colombia. Rev. Biol. Trop. 35, 333–338.

Christoffersen, M.L., Ramos, G.E., 1988. A new species of *Alpheus* (Caridea, Alpheidae) from the Pacific coast of Colombia. Revista Nordestina de Biologia, 6, 61–65.

Coates, A.G., Obando, J.A., 1996. The geological evolution of the Central American Isthmus. In: Jackson, J.B.C., Budd, A.F., Coates, A.G. (Eds.), Evolution and Environment in Tropical America. University of Chicago Press, Chicago, Illinois, pp. 21–56.

Colborn, J., Crabtree, R.E., Shaklee, J.B., Pfeiler, E., Bowen, B.W., 2001. The evolutionary enigma of bonefishes (*Albula* spp.): cryptic species and ancient separations in a globally distributed shorefish. Evolution 55, 807–820.

Cowen, R.K., Lwiza, K.M.M., Sponaugle, S., Paris, C.B., Olson, D.B., 2000. Connectivity of marine populations: open or closed? Science 287, 857–859.

Cunningham, C.W., 1997. Can three incongruence tests predict when data should be combined? Mol. Biol. Evol. 14, 733–740.

Dawson, M.N., Jacobs, D.K., 2001. Molecular evidence for cryptic species of *Aurelia aurita* (Cnidaria, Scyphozoa). Biol. Bull. 200, 92–96.

de Vargas, C., Norris, R., Zaninetti, L., Gibb, S.W., Pawlowski, J., 1999. Molecular evidence of cryptic speciation in planktonic foraminifers and their relation to oceanic provinces. Proc. Natl. Acad. Sci. USA 96, 2864–2868.

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

Duffy, J.E., 1996a. Resource-associated population subdivision in a symbiotic coral-reef shrimp. Evolution 50, 360–373.
Duffy, J.E., 1996b. Species boundaries, specialization, and the radiation of sponge-dwelling alpheid shrimp. Biol. J. Linn. Soc. 58, 307–324.
Ellingson, R.A., Krug, P.J., 2006. Evolution of poecilogony from planktotrophy: cryptic speciation, phylogeography, and larval development in the gastropod genus *Alderia*. Evolution 6011, 2293–2310.
Gittenberger, E., 1991. What about non-adaptive radiation? Biol. J. Linn. Soc. 43, 263–272.
Gray, J.S., 1997. Marine biodiversity: patterns, threats and conservation needs. Biodiv. Conserv. 6, 153–175.
Gutiérrez-Rodríguez, C., Lasker, H.R., 2004. Microsatellite variation reveals high levels of genetic variability and population structure in the gorgonian coral *Pseudopterogorgia elisabethae* across the Bahamas. Mol. Ecol. 13, 2211–2221.
Hall, T.A., 1999. BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. Nucleic Acids Symp. Ser. 41, 95–98.
Hedgecock, D., 1986. Is gene flow from pelagic larval dispersal important in the adaptation and evolution of marine invertebrates? Bull. Mar. Sci. 39, 550–564.
Hendrix, G.Y., 1971. A systematic study of the genus *Alpheus* (Crustacea: Decapoda: Alpheidae) in South Florida. Ph.D. Dissertation, University of Miami, Coral Gables, 184 pp.
Jobb, G., von Haeseler, A., Strimmer, K., 2004. TREEFINDER: a powerful graphical analysis environment for molecular phylogenetics. BMC Evol. Biol. 4, 18.
Kass, R.E., Raftery, A.E., 1995. Bayes factors. J. Am. Stat. Assoc. 90, 773–795.
Kim, W., Abele, L.G., 1988. The snapping shrimp genus *Alpheus* from the eastern Pacific (Decapoda: Caridea: Alpheidae). Smith. Contrib. Zool. 454, 1–119.
Knowles, L.L., Maddison, W.P., 2002. Statistical phylogeography. Mol. Ecol. 11, 2623–2635.
Knowlton, N., 1993. Sibling species in the sea. Ann. Rev. Ecol. Syst. 24, 189–216.
Knowlton, N., Keller, B.D., 1983. A new, sibling species of snapping shrimp associated with the Caribbean sea anemone *Bartholomea annulata*. Bull. Mar. Sci. 33, 353–362.
Knowlton, N., Mills, D.E.K., 1992. The systematic importance of color and color pattern: evidence for complexes of sibling species of snapping shrimp (Caridea: Alpheidae: *Alpheus*) from the Caribbean and Pacific coasts of Panama. Proc. San Diego Soc. Natl. Hist. 18, 1–5.
Knowlton, N., Weigt, L.A., 1998. New dates and new rates for divergence across the Isthmus of Panama. Proc. R. Soc. Lond. B 265, 2257–2263.
Knowlton, N., Weigt, L.A., Solorzano, L.A., Mills, D.K., Bermingham, E., 1993. Divergence in proteins, mitochondrial DNA, and reproductive compatibility across the Isthmus of Panama. Science 260, 1629–1632.
Lee, T., Ó Foighil, D., 2004. Hidden Floridian biodiversity: mitochondrial and nuclear gene trees reveal four cryptic species within the scorched mussel, *Brachidontes exustus*, species complex. Mol. Ecol. 13, 3527–3542.
Lee, T., Ó Foighil, D., 2005. Placing the Floridian marine genetic disjunction into a regional evolutionary context using the scorched mussel, *Brachidontes exustus*, species complex. Evolution 59, 2139–2158.
Lessios, H.A., Kessing, B.D., Pearse, J.S., 2001. Population structure and speciation in tropical seas: global phylogeography of the sea urchin *Diadema*. Evolution 55, 955–975.
Macdonald, K.S., Rios, R., Duffy, J.E., 2006. Biodiversity, host specificity, and dominance by eusocial species among sponge-dwelling alpheid shrimp on the Belize barrier reef. Div. Distrib. 12, 165–178.
Machordom, A., Macpherson, E., 2004. Rapid radiation and cryptic speciation in squat lobsters of the genus *Munida* (Crustacea, Decapoda) and related genera in the South West Pacific: molecular and morphological evidence. Mol. Phylogenet. Evol. 33, 259–279.
Mathews, L.M., 2006. Cryptic biodiversity and phylogeographic patterns in a snapping shrimp species complex. Mol. Ecol. 15, 4049–4063.
Mathews, L.M., 2007. Evidence for restricted gene flow over small spatial scales in a marine snapping shrimp *Alpheus angulosus*. Mar. Biol. 152, 645–655.
Mathews, L.M., Schubart, C.D., Neigel, J.E., Felder, D.L., 2002. Genetic, ecological, and behavioural divergence between two sibling snapping shrimp species (Crustacea: Decapoda: *Alpheus*). Mol. Ecol. 11, 1427–1437.
McClure, M.R., 1995. *Alpheus angulatus*, a new species of snapping shrimp from the Gulf of Mexico and the northwestern Atlantic, with a redescription of *A. heterochaelis* Say 1818 (Decapoda: Caridea: Alpheidae). Proc. Biol. Soc. Wash. 108, 84–97.
McClure, M.R., 2002. Revised nomenclature of *Alpheus angulatus* McClure, 1995 (Decapoda: Caridea: Alpheidae). Proc. Biol. Soc. Wash. 115, 368–370.
McClure, M.R., Greenbaum, I.F., 1994. Biochemical variation in *Alpheus* Decapoda, Caridea, Alpheidae from the coast of Texas: evidence for cryptic species. Southwest. Nat. 39, 63–66.
McMillen-Jackson, A.L., Bert, T.M., 2003. Disparate patterns of population genetic structure and population history in two sympatric penaeid shrimp species (*Farfantepenaeus aztecus* and *Litopenaeus setiferus*) in the eastern United States. Mol. Ecol. 12, 2895–2905.
Milne-Edwards, H., 1837. Histoire naturelle des Crustacés, comprenant l'anatomie, la physiologie et la classification de ces animaux. Librairie encyclopédique de Roret, vol. 2. Paris, pp. 1–531 (atlas: 1–32, pls. 1–42).
Miura, O., Torchin, M.E., Kuris, A.M., Hechinger, R.F., Chiba, S., 2006. Introduced cryptic species of parasites exhibit different invasion pathways. Proc. Natl. Acad. Sci. USA 103, 19818–19823.
Morrison, C.L., Ríos, R., Duffy, J.E., 2004. Phylogenetic evidence for an ancient rapid radiation of Caribbean sponge-dwelling snapping shrimps (*Synalpheus*). Mol. Phylogenet. Evol. 30, 563–581.
Nylander, J.A.A., 2004. MRMODELTEST version 2.2. Program Distributed by the Author. Evolutionary Biology Centre, Uppsala University, Uppsala, Sweden.
Palumbi, S.R., 1994. Genetic divergence, reproductive isolation, and marine speciation. Ann. Rev. Ecol. Syst. 25, 547–572.
Palumbi, S.R., Martin, A., Romano, S., McMillan, W.O., Stice, L., 1991. The Simple Fool's Guide to PCR: A Collection of PCR Protocols, Version 2. University of Hawaii, Honolulu.
Palumbi, S.R., Grabowsky, G., Duda, T., Geyer, L., Tachino, N., 1997. Speciation and population genetic structure in tropical Pacific sea urchins. Evolution 51, 1506–1517.
Perrin, C., Wing, S.R., Roy, M.S., 2004. Effects of hydrographic barriers on population genetic structure of the sea star *Coscinasteriasmuricata* (Echinodermata, Asteroidea) in the New Zealand fiords. Mol. Ecol. 13, 2183–2195.
Powles, H., Bradford, M.J., Bradford, R.G., Doubleday, W.G., Innes, S., Levings, C.D., 2000. Assessing and protecting endangered marine species. ICES J. Mar. Sci. 57, 669–676.
Rambaut, A., Drummond, A.J., 2007. Tracer v1.4. Available from: <http://beast.bio.ed.ac.uk/Tracer>.
Ramos, G.E., 1997. *Alpheus agrogon*, a new species of alpheid shrimp (Decapoda: Alpheidae) from Gorgona Island, Pacific coast of Colombia. Rev. Biol. Trop. 44–45, B, 395–400.
Rankin, W.M., 1900. The Crustacea of the Bermuda Islands, with notes on the collections made by the New York University expeditions in 1897 and 1898. Ann. NY Acad. Sci. 12, 521–548.
Riginos, C., Nachman, M.W., 2001. Population subdivision in marine environments: the contributions of biogeography, geographical distance and discontinuous habitat to genetic differentiation in a blennioid fish, *Axoclinus nigricaudus*. Mol. Ecol. 10, 1439–1453.
Ríos, R., Duffy, J.E., 2007. A review of the sponge-dwelling snapping shrimp from Carrie Bow Cay, Belize, with description of *Zuzalpheus*, new genus, and six new species (Crustacea: Decapoda: Alpheidae). Zootaxa 1602, 1–82.
Rocha, L.A., Bass, A.L., Robertson, D.R., Bowen, B.W., 2002. Adult habitat preferences, larval dispersal, and the comparative phylogeography of three Atlantic surgeonfishes (Teleostei: Acanthuridae). Mol. Ecol. 11, 243–252.
Rocha, L.A., Robertson, D.R., Roman, J., Bowen, B.W., 2004. Ecological speciation in tropical reef fishes. Proc. R. Soc. Lond. B 272, 573–579.
Rocha-Olivares, A., Fleeger, J.W., Foltz, D.W., 2001. Decoupling of molecular and morphological evolution in deep lineages of a meiobenthic harpacticoid copepod. Mol. Biol. Evol. 18, 1088–1102.
Schmitt, W.L., 1924. Report on Macrura, Anomura and Stomatopoda collected by the Barbados-Antigua expedition from the University of Iowa in 1918. Stud. Nat. Hist. Iowa Univ. 10, 65–99.
Schubart, C.D., Neigel, J.E., Felder, D.L., 2000. The use of the mitochondrial 16S rRNA gene for phylogenetic and population studies of Crustacea. Crust. Issues 12, 817–830.
Šlapeta, J., López-García, P., Moreira, D., 2006. Global dispersal and ancient cryptic species in the smallest marine eukaryotes. Mol. Biol. Evol. 23, 23–29.
Solé-Cava, A.M., Klautau, M., Boury-Esnault, N., Borojecic, R., Thorpe, J.P., 1991. Genetic evidence for cryptic speciation in allopatric populations of two cosmopolitan species of the calcareous sponge genus *Clathrina*. Mar. Biol. 111, 381–386.
Swearer, S.E., Caselle, J.E., Lea, D.W., Warner, R.R., 1999. Larval retention and recruitment in an island population of a coral-reef fish. Nature 402, 799–802.
Swearer, S.E., Shima, J.S., Hellberg, M.E., Thorrold, S.R., Jones, G.P., Robertson, D.R., Morgan, S.G., Selkoe, K.A., Ruiz, G.M., Warner, R.R., 2002. Evidence of self-recruitment in demersal marine populations. Bull. Mar. Sci. S70, 251–271.
Swofford, D.L., 2003. PAUP*. Phylogenetic Analysis Using Parsimony (*and Other Methods), Version 4.0B10. Sinauer Associates, Sunderland, MA.
Tarjuelo, I., Posada, D., Crandall, K.A., Pascual, M., Turon, X., 2001. Cryptic species of *Clavelina* (Ascidiacea) in two different habitats: harbours and rocky littoral zones in the northwestern Mediterranean. Mar. Biol. 139, 455–462.
Taylor, M.S., Hellberg, M.E., 2005. Marine radiations at small geographic scales: speciation in neotropical reef gobies (*Elacatinus*). Evolution 59, 374–385.
Thompson, J.D., Higgins, D.G., Gibson, T.J., 1994. CLUSTAL W—improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. Nucleic Acids Res. 22, 4673–4680.
Tipton, J.A., 2006. Aristotle's study of the animal world. Perspec. Biol. Med. 49, 369–383.
Verrill, A.E., 1922. Decapod Crustacea of Bermuda. Part II. Macrura. Trans. Conn. Acad. Arts Sci. 26, 1–179. pls. 1–48.
Wares, J.P., Cunningham, C.W., 2001. Phylogeography and historical ecology of the North Atlantic intertidal. Evolution 55, 2455–2469.
Warner, R.R., Cowen, R.K., 2002. Local retention of production in marine populations: evidence, mechanisms, and consequences. Bull. Mar. Sci. 70, 245–249.
Wicksten, M.K., 1983. A monograph of the shallow-water caridean shrimps of the Gulf of California, Mexico. Allan Hancock Foundation Monographs 13, 1–59.
Williams, S.T., Benzie, J.A.H., 1998. Evidence of a biogeographic break between populations of a high dispersal starfish: congruent regions within the Indo-west Pacific defined by color morphs, mtDNA, and allozyme data. Evolution 52, 87–99.
Williams, S.T., Knowlton, N., 2001. Mitochondrial pseudogenes are pervasive and often insidious in the snapping shrimp genus *Alpheus*. Mol. Biol. Evol. 18, 1484–1493.

Williams, S.T., Knowlton, N., Weigt, L.A., Jara, J.A., 2001. Evidence for three major clades within the snapping shrimp genus *Alpheus* inferred from nuclear and mitochondrial gene sequence data. Mol. Phylogenet. Evol. 20, 375–389.

Wilson, N.G., Hunter, R.L., Lockhart, S.J., Halanych, K.M., 2007. Multiple lineages and absence of panmixia in the "circumpolar"crinoid *Promachocrinus kerguelensis* from the Atlantic sector of Antarctica. Mar. Biol. 152, 895–904.