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Miocene Radiation of Deep-Sea Hydrothermal Vent Shrimp (Caridea: Bresiliidae): Evidence from Mitochondrial Cytochrome Oxidase Subunit I

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The evolutionary history of deep-sea shrimp (Caridea: Bresiliidae) inhabiting deep-sea hydrothermal vent and hydrocarbon seep environments was assessed using the mitochondrial Cytochrome *c* Oxidase subunit I (*COI*) gene (600 bp). Phylogenetic analyses (parsimony, likelihood, and neighbor-joining) recovered three distinct clades (A, *Rimicaris/Chorocaris/Opaepele;* B, *Alvinocaris;* and C, *Mirocaris*) consistent with higher level taxonomy based on morphology. However, robust phylogenetic results suggested that *Chorocaris* is paraphyletic and that *Mirocaris fortunata* and *M. keldyshi* may not be genetically distinct. A Kishino–Hasegawa likelihood approach was used to test alternative phylogenetic hypotheses based on biogeography and morphology. Evolutionary relationships of vent-endemic shrimp species did not appear to be correlated either with their extant biogeographic distribution or with the history of sea floor spreading. Additionally, *COI* data suggested that these vent-endemic organisms are not remnants of a Mesozoic vent assemblage; instead, they radiated in the Miocene.

INTRODUCTION

Since the discovery of hydrothermal vents and other reducing habitats in the deep sea (e.g., cold-water sulfide/hydrocarbon seeps), 11 species of vent- or seep-endemic shrimp (Infraorder: Caridea) have been described from the Atlantic and Pacific Oceans (Fig. 1). Although various species occur at depths ranging from 530 to 3660 m, any given species tends to have a more limited depth range (Table 1). Additionally, individual species appear to be restricted to a particular ridge axis, seep system, or seamount. The greatest diversity of these shrimp (7 species) occurs along the Mid-Atlantic Ridge (MAR), where individual vent fields (e.g., Broken Spur and Logatchev) may support as many as 4 caridean species. Typical MAR sites are dominated by dense clusters of shrimp (up to 25,000 individuals/m^2) that live near actively venting sulfide chimneys (Rona, 1986; Van Dover, 1995). In contrast to the MAR, all other hydrothermal vent and hydrocarbon seep sites in the Gulf of Mexico and Pacific Ocean are inhabited by only a single species (Table 1). Two species are known from the western Pacific, 2 from Pacific seamounts, and 2 from hydrocarbon seeps in the Gulf of Mexico.

The disjunct distributions of these caridean species raises fundamental biogeographical and evolutionary questions. What are the relationships between MAR shrimp species and Pacific species? Do evolutionary relationships among the vent shrimp reflect the history of seafloor spreading? Are the extant shrimp species relicts of a Mesozoic radiation as hypothesized for limpets and barnacles (McLean, 1985; Newman, 1985)? Are the vent-endemic shrimp derived from seep ancestors, as in bivalves and vestimentiferan tube worms (Craddock *et al.,* 1995; Black *et al.,* 1997; Peek *et al.,* 1997)? A solid phylogenetic framework is required before these questions can be addressed.

Current bresiliid taxonomy is in a state of flux. The 11 known caridean shrimp species from vent and seep environments have variously been placed in three families and six genera. A recent stream of proposals for new family- and genus-level reassignments, based primarily on morphological traits, has fueled considerable debate concerning evolutionary relationships (Williams and Rona, 1986; Williams, 1988; Williams and Dobbs, 1995; Martin and Hessler, 1990; Martin and Christiansen, 1995; Vereshchaka, 1996a, 1997; Shank *et al.,* 1998a). These taxonomic debates will undoubtedly continue as new species are discovered. For the purposes of this study, we adopted a conservative approach and recognized the family Bresiliidae (*sensu*

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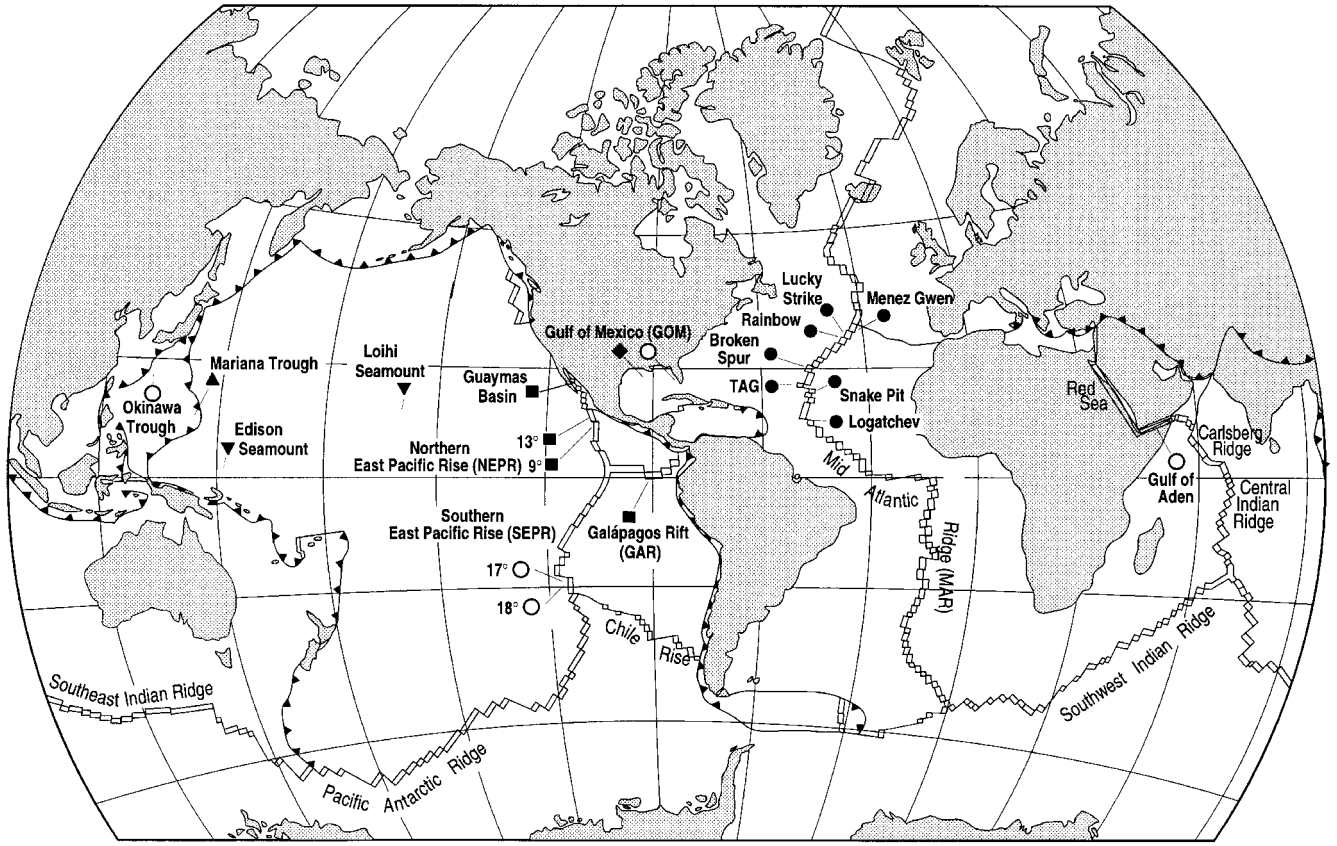

FIG. 1. Geographic location of deep-sea hydrothermal vent and hydrocarbon seep sites inhabited by vent- and seep-endemic shrimp. Filled symbols indicate collection sites used in this study, whereas open circles indicate additional sites where vent-associated shrimp have been described or reported (Okinawa Trough: Kikuchi and Ohta, 1995; Florida Escarpment: Williams, 1988; 17°–18° SEPR: Geistdoerfer *et al.*, 1995; Gulf of Aden: Juniper *et al.*, 1990). Filled circles, Atlantic vent species; filled squares, eastern Pacific vent species; filled triangles, western Pacific vent species; inverted triangles, Pacific seamount vent species; filled diamond = hydrocarbon seep species.

Williams and Dobbs, 1995), although we understand that the familial designation Alvinocarididae has been accepted by many for vent and seep-endemic shrimp and may be soon formally amended (A. Williams, pers. comm.).

Because the goal of this study was to develop a robust phylogenetic framework for these taxa, we developed a molecular phylogeny based on a 600-bp region of the mitochondrial Cytochrome *c* Oxidase I gene (*COI*). This particular gene segment has proved useful for addressing similar questions regarding vestimentiferan tube worms (Black *et al.*, 1997) and vesicomyid clams (Peek *et al.*, 1997) from deep-sea vents and seeps. Specifically, we address the phylogenetic congruence with taxonomy and biogeographic hypotheses. Finally, we examine the age of vent- and seep-endemic shrimp diversification.

MATERIALS AND METHODS

Specimens

The shrimp used in this study were collected from hydrothermal vent and hydrocarbon seep sites (Fig. 1) using manned submersibles (Alvin, Nautile, Shinkai 6500, and Johnson Sea Link) during several oceanic cruises. Collection locality and depth and number of individuals examined are given in Table 2. Voucher specimens, fixed in 10% buffered formalin (24 h) and preserved in 70% ethanol, were deposited with the U.S. National Museum, Washington, DC. For each species or operational taxonomic unit (OTU), between two and nine specimens were examined, with the exception of the single individual designated as "Lucky Strike." The vent-endemic shrimp *Alvinocaris longirostris* (Okinawa Trough; Kikuchi and Ohta, 1995) and the seep-endemic shrimp *Alvinocaris muricola* (Florida Escarpment: Williams, 1988) were not available due to their rarity. A previous study (Shank *et al.*, 1998a) of allozyme and mtDNA in *Rimicaris exoculata* (Williams and Rona, 1986) and *Iorania concordia* (Vereshchaka, 1996a), also described as *Rimicaris aurantiaca* by Martin *et al.* (1997), revealed that these morphotypes comprise a single randomly mating conspecific population at localities where they have been reported. Consequently, we treat these morphotypes as *R. exoculata*.

TABLE 1

Global Distribution of Deep-Sea Hydrothermal Vent- and Hydrocarbon Seep-Endemic Bresiliid Shrimp

	Mid-Atlantic Ridge vents							Gulf of Mexico seeps		NEPR vents			SEPR vents		MPAC vents		WPAC vents	
Species* / Depth (m)	Menez Gwen 850	Lucky Strike 1710	Rainbow ~2250	Broken Spur 3300	TAG 3650	Snake Pit 3480	Logatchev 3010	Florida Escarp. 3300	Louisiana Slope 530	13°N 2600	9°N 2520	GAR 2400	17°24-25′S β ~2700	18°22-37′S β ~2700	Loihi Seamount 980	Edison Seamount 1483	Mariana Trough 3660	Okinawa Trough 1360
Rimicaris exoculata	?	X	X	X	X	X	X											
Chorocaris chacei		X	Xv	X	X	X	X											
Chorocaris vandoverae																	X	
Opaepele loihi															X			
Alvinocaris markensis				Xv	X	X	X											
Alvinocaris lusca										X	X	X	X?	X?				
Alvinocaris n. sp. 1																X		
Alvinocaris n. sp. 2	X																	
Alvinocaris stactophila									X									
Alvinocaris longirostris							X?											X
Alvinocaris muricola							X?	X										
Mirocaris keldyshi				X	X	X	X											
Mirocaris fortunata	X	X	X	X														
Lucky Strike n. sp.		X																

Note. NEPR, Northern East Pacific Rise; SEPR, Southern East Pacific Rise; MPAC, Middle Pacific Seamouts; WPAC, Western Pacific Vents; β, "white shrimp" resembling *A. lusca* have been reported within vestimentiferan communities and around high-temperature smoker chimneys; v, observed on video only (T.M.S., pers. observ.); ?, unconfirmed reports (A. Gebruk and A. Vereshchaka, pers. comm.); *, *Iorania concordia* described from the Mid-Atlantic Ridge is considered synonymous with *Rimicaris exoculata* based on a previous genetic study (see Results and Shank *et al.* 1998a).

The outgroup taxa, *Stenopus hispidus* and *Crangon septimspinosa*, were purchased as live specimens from Pacific Bio-Marine Supply Co. (CA).

Immediately upon collection, whole shrimp were identified to species and stored frozen at −80°C for subsequent DNA extraction at Rutgers University or DNA was extracted aboard the research support vessel. Morphological identifications based on published morphological descriptions were confirmed by T.M.S. prior to DNA analyses. Distinctions between *Mirocaris fortunata* and *M. keldyshi* were based on the number of telson spines, the presence of movable spines on ischium of pereopod II, and the presence of spines on the propodus of pereopod I (Vereshchaka, 1997).

DNA Extraction, PCR, and Sequencing

Approximately 0.1 g of frozen abdominal muscle between the second and fourth pleura was removed, and total genomic DNA was extracted using a modified hexadecyl–trimethyl–ammonium bromide (CTAB) protocol (Doyle and Dickson, 1987) followed by a phenol extraction and ethanol precipitation. In the case of *Alvinocaris* n. sp. (A. Williams, pers. comm.) from Edison Seamount, 0.3 g of formalin-fixed/70% ethanol-preserved abdominal tissue was excised and genomic DNA was extracted using a modified Shiozawa *et al.* (1992) protocol consisting of an additional proteinase K treatment (France and Kocher, 1996). Purified DNA was hydrated in 100 to 150 μl deionized water and stored at −20°C.

A 710-bp region of the mt*COI* gene was amplified using universal primers LCO1490 and HCO2198 designed by Folmer *et al.* (1994). PCR amplification, cycle sequencing, and electrophoretic methods have been previously described in Shank *et al.* (1998a). Briefly, each 50-μl amplification reaction consisted of 35–70 ng of template DNA, 5 μl 10× buffer (supplied by manufacturer, 5 μl $MgCl_2$ at 2.5 μM), 5 μl of each primer (1 μM final conc.), 1.5 units of Taq polymerase (Promega Inc., WI), and 24 μl sterile dH_2O. After denaturation for 1 min at 95°C, 5 μl of a 2 mM stock solution of dNTPs was added to the reaction mix. Amplification occurred over 35 cycles at 95°C, 1 min, 52°C, 1 min, and 72°C, 1.5 min, followed by a final extension step of 7 min at 72°C. Negative controls were included and sterile procedures were consistently followed for all PCR experiments. PCR amplifications were confirmed with agarose gel electrophoresis, and the PCR product was purified via precipitation with 8 M ammonium acetate (pH 5.8) and ethanol. Sequencing reactions employed the oligonucleotides used in amplifications. The automated sequence reactions followed the manufacturer's (FS Dye Termination Mix, Applied Biosystems Inc., CA) recommendations using ~60 ng of purified PCR product as template. The reaction profile was 25 repetitions of denaturation at 96°C for 30 s, annealing at 50°C for 15 s, and extension at 64°C for 4 min. Fragments were visualized on a Perkin–Elmer ABI 373 DNA sequencer using dye-terminated dideoxy labeling (ABI, Foster

TABLE 2

Specimen Collection Sites, Identification of Shrimp Species, and Number of Individuals of Each Taxon from Which Mitochondrial Cytochrome *c* Oxidase (*COI*) Sequence Was Obtained

Infraorder	Family	Species	No. of indiv. sampled by site	Collection sites	Depth (m)	Dive no.	Collection date
Caridea	Bresiliidae	*Rimicaris exoculata*	2 TAG	(26°08.3′N; 44°49.6′W)	3650	A 3126	19 Jul. 1997
			2 Lucky Strike	(37°17.3′N; 32°15.5′W)	1705	J 180	27 Jul. 1996
			3 Broken Spur	(29°10.0′N; 43°10.0′W)	3044	A 3124	16 Jul. 1997
			2 Rainbow	(36°14.0′N; 33°53.0′W)	2251	A 3121	11 Jul. 1997
		Chorocaris chacei	3 Snake Pit	(23°22.1′N; 44°57.2′W)	3486	A 2618	19 Jun. 1993
			2 TAG	(26°08.3′N; 44°49.6′W)	3650	A 3126	19 Jul. 1997
		Chorocaris vandoverae	2 Alice Springs	(18°12.8′N; 144°42.4′E)	3589	S140	14 Sep 1992
		Opaepele loihi	2 Loihi Seamount	(18°55.0′N; 155°16.0′W)	980	T242	8 Jul. 1993
		Alvinocaris markensis	2 Snake Pit	(23°22.1′N; 44°57.1′W)	3398	A 2621	22 Jun. 1993
		Alvinocaris lusca	2 Galápagos	(00°48.2′N; 86°13.4′W)	2461	A 2224	29 May 1990
			2 9°North	(09°50.3′N; 104°17.4′W)	2520	A 2692	26 Dec. 1993
		Alvinocaris sp.	2 Edison Seamount	(03°18.8′N; 152°39.9′E)	1483	TVG29	20 Mar. 1994
		Alvinocaris stactophila	2 Louisiana Slope	(27°46.9′N; 91°30.3′W)	534	JSL	Jun. 1995
	Mirocarididae	*Mirocaris fortunata*	3 Lucky Strike	(37°17.3′N; 32°15.5′W)	1710	N P21	Jun. 1994
			3 Broken Spur	(29°10.0′N; 43°10.0′W)	3036	A 3124	16 Jul. 1997
			1 Menez Gwen	(37°50.4′N; 31°31.3′W)	850	A 3117	7 Jul. 1997
		Mirocaris keldyshi	2 Broken Spur	(29°10.0′N; 43°10.0′W)	3056	A 3124	16 Jul. 1997
			1 TAG	(26°08.3′N; 44°49.6′W)	3655	A 3126	19 Jul. 1997
			1 Logatchev	(14°45.0′N; 44°58.8′W)	3011	A 3132	26 Jul. 1997
		Undescribed species	1 Lucky Strike	(37°17.3′N; 32°16.1′W)	1685	N P21	10 Jun. 1994
	Crangonidae	*Crangon septemspinosa*	2 Purchased		—	—	Aug. 1994
	Nematocarcinidae	*Nematocarcinus(ensifer?)*	2 9°NEPR	(09°50.3′N; 104°17.4′W)	2505	A 2688	22 Dec. 1993
	Hippolytidae	*Lebbeus carinatus*	2 13° NEPR	(12°48.6′N; 103°56.4′W)	2613	N P02	21 Feb. 1996
	Pasiphaeidae	*Pasiphaea tarda*	2 Oregon Slope	(45°43.0′N; 124°92.0′W)	1152	Tr 22	24 Oct. 1994
	Pandalidae	*Pandalus tridens*	2 Guaymas Basin	(27°09.0′N; 111°42.0′W)	1568	Tr 2	Oct. 1994
Stenpodidea	Stenopodidae	*Stenopus hispidus*	2 Purchased		—	—	Aug. 1994

Note. Identical *COI* sequences were obtained from all species regardless of collection site with exception of individuals within *Mirocaris fortunata* and *Mirocaris keldyshi*. Subsequent phylogenetic analysis included only those sequences that contained nucleotide differences (Dive submersibles and sampling gear: A "Alvin"; J "Jason"; S "Shinkai 6500"; JSL "Johnson Sea Link"; N "Nautile"; Tr "Trawl"; TVG "Television Grab"; T "Turtle").

City, CA) under standard cycle-sequencing conditions. Both strands of the PCR product were completely sequenced.

Phylogenetic Analyses

Sequences were initially aligned with the program Clustal W (Thompson *et al.*, 1994). The alignment was then optimized by eye using GDE 2.2 editor and subsequently translated into amino acids using the published *Drosophila yakuba* translation code. The aligned data sets have been submitted to the public database TREEBASE (http://phylogeny.harvard.edu/treebase).

For the present phylogenetic analyses, we adopted a conservative approach and decided to draw conclusions only for results that were consistently supported under a variety of evolutionary assumptions. Therefore, phylogenetic analyses were performed using neighbor joining (NJ), maximum parsimony (MP), and maximum likelihood (ML) for the nucleotide data. Reconstructions were not conducted for the protein sequence data due to the lack of phylogenetically informative characters. For NJ analysis, the DNAdist and Neighbor programs of PHYLIP ver 3.57c (Felsenstein, 1993) were employed with an HKY model of nucleotide evolution (Hasegawa *et al.*, 1985) and empirical nucleotide frequencies. All heuristic parsimony searches using PAUP ver 4.0d61&62 (Swofford, 1993) employed TBR branch swapping and 50 random sequence-addition replications. For ML analyses, we used the fastDNAml ver 1.0.6 program (Olsen *et al.*, 1994) or the DNAml program from PHYLIP ver 3.57c with empirical nucleotide frequencies (i.e., an HKY model). For both parsimony and likelihood searches, a wide range of transition: transversion (Ti:Tv) ratios (1:1, 1.5:1, 2:1, 5:1, 10:1) were used. This range encompassed the empirically derived estimates (1.1:1–1.4:1) calculated using 1000 randomly constructed trees in MacClade ver 3.06 (Maddison and Maddison, 1992). Similar estimates of the empirical Ti:Tv were obtained with a likelihood approach (see Halanych and Robinson, 1997). MP and NJ bootstrap estimates (Felsenstein, 1985) of tree topologies used 500 iterations of resampling. Due to computation time, only 200 iterations were performed for ML trees. Nucleotide composition, pairwise comparisons,

and statistical information were gathered using MEGA ver 1.01 (Kumar *et al.*, 1993) and MacClade.

To root the resultant trees and determine character polarity, representatives of 5 of the 15 caridean superfamilies (Chace, 1992) were included as outgroups. These 5 taxa were chosen based on their affinity with the family Bresiliidae (e.g., Hippolytidae and Nematocarcinidae; Holthuis, 1955; Thompson, 1966; Zarenkov, 1976; de Saint Laurent, 1984; Crosnier and Forest, 1973) and because of their close proximity to vent environments (e.g., Shank *et al.*, 1998b). For illustrative purposes, the most taxonomically distant shrimp examined herein (*Stenopus hispidus*) was used to root the topologies.

Hypothesis Testing

In order to test alternative phylogenetic and biogeographic hypotheses, we constrained heuristic parsimony searches to find the best tree (or set of trees) consistent with a given hypothesis. Next, the branch lengths for the constrained topologies, and the overall best topology, were optimized with the likelihood parameters (described above) using the DNAml program of PHYLIP (user-defined tree option). Using a Kishino–Hasegawa likelihood approach, we compared the likelihood scores of the alternative (constrained) topologies to the score of the best topology. Significance was judged using a standard deviation measure (as in Kishino and Hasegawa, 1989).

RESULTS

The final sequenced *COI* product consisted of 600 bp when aligned for the 22 shrimp OTUs. Translation of the DNA sequences into protein sequences revealed no amino acid insertions or deletions. Of 199 amino acid residues, 41 were variable and only 8 were phylogenetically informative with regard to the 16 ingroup taxa (see Fig. 2).

Due to the small number of informative amino acid changes, phylogenetic analyses were performed only with aligned DNA sequences. Two hundred sixty-one variable positions revealed 217 phylogenetically informative substitutions. Aside from the genus *Mirocaris*, intraspecific comparisons identified no sequence variation among conspecific individuals (e.g., the nine *R. exoculata* individuals from four MAR vent sites, see Table 2). Typical of arthropod mtDNA, the nucleotide sequences were A-T rich and showed exceptionally low numbers of guanine residues (4.7%) at third positions. About half (33) of the 57 first-position variable sites resulted in an amino acid change. Most (181) informative substitutions were synonymous third-position changes.

Genetic distances within the Bresiliidae (as estimated under the HKY model) ranged from 0.11, between *R. exoculata* and *Chorocaris chacei,* to 0.219, between *M. keldyshi* (from TAG) and *Alvinocaris lusca* (Table 3). The maximum *d* value (0.294) was found between *A. lusca* and the outgroup species, *Pandalus tridens.* These higher levels of divergence between ingroup and outgroup taxa may approach the point of saturation for nucleotide substitutions in this mitochondrial coding gene. To assess whether nucleotide substitutions were saturated, the number of transitions and transversions, by codon position, were plotted against HKY sequence distances for all possible pairwise comparisons of OTUs (data not shown). The number of third-position substitutions for the ingroup increased linearly with genetic distances; distances for ingroup pairwise comparisons were <0.219, and the ingroup/ingroup comparisons were clearly separated from ingroup/outgroup and outgroup/outgroup comparisons. All three of these factors suggest that nucleotide substitution has not degraded the phylogenetic signal in this data set (see Halanych and Robinson, 1997).

All phylogenetic analyses (neighbor joining, maximum parsimony, maximum likelihood, employing various nucleotide substitution models) produced the same topology. This topology with branch lengths estimated via maximum likelihood is shown in Fig. 2. The topology was also robust under a wide range of Ti/Tv ratios (e.g., 1.5, 2.0, 5.0, 10). For the parsimony search in which Ti and Tv changes where weighted equally (i.e., 1:1), a tree length of 803 steps was found (CI = 0.5230; excluding uninformative characters = 0.4987). Bootstrap values for maximum parsimony, maximum likelihood, and neighbor joining (assuming the empirical Ti:Tv ratio) are shown in Fig. 2.

All reconstruction methods divided the bresiliids into three well-supported clades (A, B, C; Fig. 2). Clade A included *R. exoculata* and *C. chacei* from the Mid-Atlantic Ridge, *C. vandoverae* from Western Pacific, and *Opaepele loihi* from Loihi Seamount near Hawaii. This clade was supported in 100, 96, and 99% bootstrap iterations for ML, MP, and NJ, respectively. Clade B (ML 99%, MP 71%, NJ 78%) was composed of all the *Alvinocaris* species, including representatives from both Atlantic and Pacific localities. Clade C (ML, MP, NJ, all 100%) included all *Mirocaris* samples and the undescribed Lucky Strike (MAR) shrimp. Although the Lucky Strike shrimp differed only slightly ($\sim$0.025) from some of the *Mirocaris* OTUs, it had a highly divergent morphology (e.g., a single large, preorbital, pigmented eye and completely lacked a rostrum; A. Williams, pers. comm.).

Hypothesis Testing

We tested alternative hypotheses of bresiliid evolution using a Kishino–Hasegawa likelihood approach. Because *Opaepele* shares morphological features with both *Chorocaris* (including *Rimicaris*) and *Alvinocaris* (Williams and Dobbs, 1995), we tested whether there was significant support for *Opaepele* being more closely

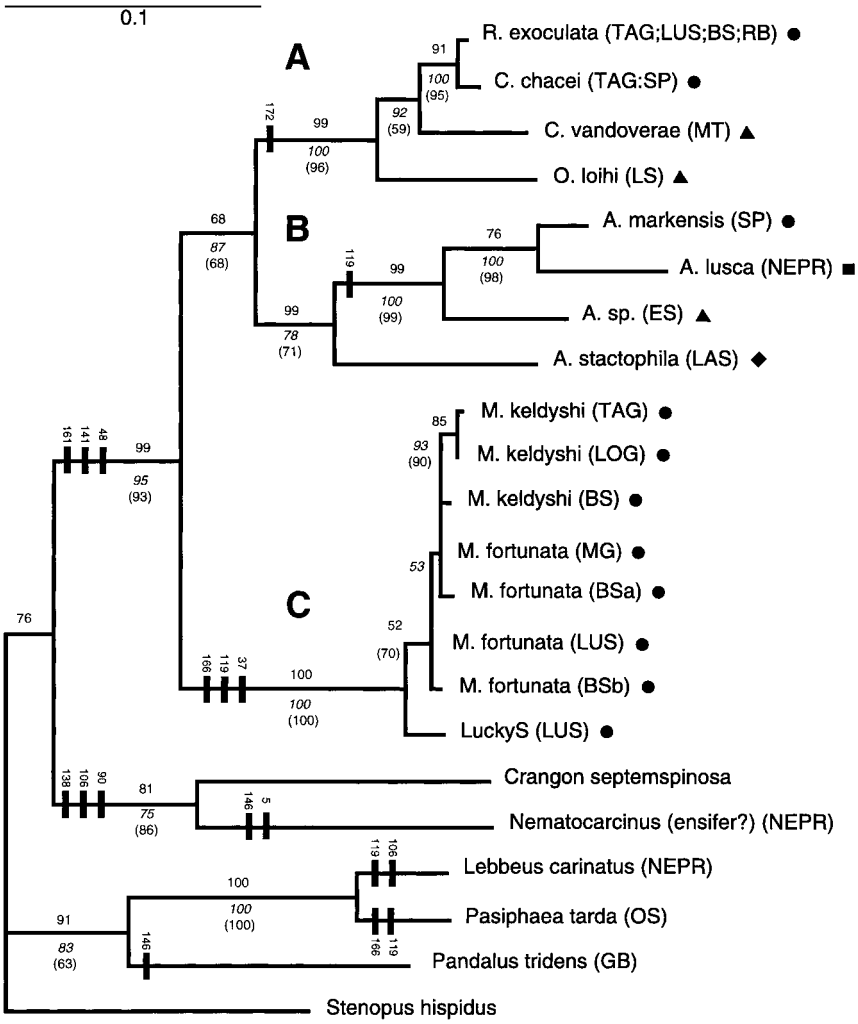

FIG. 2. Maximum likelihood tree (-LnLi = 4569.89; see text for details) based on *COI* nucleotide sequences and rooted with *Stenopus hispidus*. Bootstrap support (>50%) is shown by the numbers along the branches (ML, plain text; MP, italic text, NJ, in parentheses). Amino acid changes on the tree are shown by the bars on the relevant branch. The numbers adjacent to the bar refer to the amino acid position in the translated data set. Morphological species designations and collection sites (in parentheses and in symbol notation) are at ends of tree branches with the following abbreviations (alphabetical order): from the Mid-Atlantic Ridge: *BS*, Broken Spur (a and b represent different individuals); *LOG*, Logatchev; *LUS*, Lucky Strike; *MG*, Menez Gwen; *RB*, Rainbow; *SP*, Snake Pit; *TAG*, TAG hydrothermal mound; from the Pacific: *ES*, Edison Seamount; *GB*, Guaymas Basin; *LS*, Loihi Seamount; *MT*, Mariana Trough; *NEPR*, 9° and 13° Northern East Pacific Rise; *OS*, Oregon Slope; and from the Gulf of Mexico seeps: *LAS*, Louisiana Slope. See Fig. 1 for symbol definitions; scale bar is proportional to inferred nucleotide divergence.

related to the *Rimicaris/Chorocaris* clade (best tree) than to the *Alvinocaris* clade (constrained tree). The K-H test showed significant ($P < 0.05$) support for *Opaepele/Rimicaris/Chorocaris* hypotheses (Negative Natural Log Likelihood or -LnLi score = 4575.76) over the *Opaepele/Alvinocaris* alternative (-LnLi = 4631.16, SD 16.21).

Another alternative hypothesis based on a suggestion of Gebruk (1997) and others is that vent shrimp are derived from shallow-water seep ancestors. We asked whether the seep-endemic *Alvinocaris stactophila* was basal to any lineage other than *Alvinocaris*. *A. stactophila* is one of only two known seep-endemic shrimp. Therefore, we constrained *A. stactophila* to be basal to all vent shrimp. The best topology (-LnLi = 4575.76) was again significantly ($P < 0.5$) supported over the constrained topology (-LnLi = 4647.13, SD 17.04). Various topological permutations of this specific alternative hypothesis yielded similar results.

The last K-H test that we performed addressed Tunnicliffe and Fowler's (1996) hypothesis that the relationships of vent fauna reflect historical processes of ridge axes and tectonic plate movements. To fit the prediction of this hypothesis, we hierarchically grouped Atlantic species from similar sites together, grouped Pacific species together, and considered all possible placements of the seep species relative to these two clades. We found no support ($P < 0.5$) for this geographi-

TABLE 3

Nucleotide Substitutions and Composition by Codon Position for Cytochrome Oxidase Subunit I

Characteristic	Position in codon 1st	2nd	3rd	All sites
Percent nucleotide composition (598 bp)				
A	29.4	12.2	41.3	27.6
T	17.5	43.5	32.8	31.2
C	22.6	25.7	21.3	23.2
G	30.4	18.6	4.7	17.9
No. of variable sites	57	13	189	259
Nonsysnonymous	33	12	12	57
Informative sites	31	4	181	216

cally constrained tree (-LnLi = 4791.77, SD 33.41) over the best tree.

Because Tunnicliffe and Fowler (1996) more specifically hypothesized that geographic distance along ridge axes should be correlated to taxonomic (and hence genetic) divergence, we also used a Mantel test of matrix comparison (Douglas and Endler, 1982) to determine whether genetic distance increased with geographical distance. We used the empirical genetic distances (Table 4) against the shortest oceanic distances between vent sites and against distances along ridges and faults (data from Tunnicliffe *et al.*, 1996), with distances for Loihi Seamount, Louisiana Slope, and Edison Seamount added). Neither shortest distance (one-tailed, $P = 0.22$) nor ridge distance (one-tailed, $P = 0.22$) was significantly correlated with genetic distance.

Age of Vent Shrimp

To assess when the extant hydrothermal vent shrimp radiated, we employed a molecular clock estimate based on *COI* nucleotide data. Before such an estimate can be made, however, the assumption that nucleotide evolution proceeded in a clock-like fashion had to be tested. Therefore, we employed relative rate tests to determine if synonymous and nonsynonymous nucleotide substitutions (Muse and Gaut, 1994; modified to implement the *Drosophila* translation code) were accumulating at significantly different rates across the lineages examined. We found no evidence for significant rate heterogeneity among the present lineages.

In the absence of a reliable caridean fossil record (see Schram, 1986), we calibrated a molecular clock using *COI* divergence rates estimated for other caridean shrimp. Reproductive isolation and *COI* sequence divergence in seven Caribbean and Pacific shrimp species pairs separated by the well-dated rise of the Isthmus of Panama (3.0 to 3.5 MYA) were extensively examined by Knowlton *et al.* (1993). From these transisthmian species pairs, they estimated rates of *COI* sequence divergence to be between 2.2 and 2.6% per million years. Applying these rates of *COI* divergence to the present data, we estimated that *R. exoculata* and *C. chacei* shared a last common ancestor 0.42 to 0.5 MYA, the *Rimicaris/Chorocaris* clade and *Alvinocaris* shared a last common ancestor 6.2 to 9.9 MYA, and *Mirocaris* shared a last common ancestor with the *Rimicaris/Chorocaris/Alvinocaris* clade 6.7 to 11.7 MYA. We have conservatively interpreted these dates to be Miocene but it is possible that the rates of nucleotide substitution have significantly slowed in these shrimps. Nonetheless, the present rate estimates would have to be over fivefold greater to push the radiation of this group into the late Mesozoic.

DISCUSSION

The present *COI* nucleotide divergence estimates revealed that extant species of vent- and seep-endemic bresiliid shrimp constitute a natural (i.e., monophyletic) group that most likely radiated in the Miocene. The resultant tree topology (Fig. 2) suggests that diversification events within bresiliids do not correspond to the geographic location of hydrothermal vent areas. Also, the molecular data reveal that further consideration of the current taxonomy and its morphological criteria are warranted.

Although currently recognized intergeneric relationships tended to be well supported, the molecular topology, in some cases, disagreed with the morphologically based taxonomy. For example, *C. chacei* was originally placed in the genus *Rimicaris* as *R. chacei* (Williams and Rona, 1986) but with the discovery of *C. vandoverae, R. chacei* was assigned to the genus *Chorocaris* (Martin and Hessler, 1990). This novel placement was motivated by, among other considerations, the similarity of the reduced triangular rostrum in these two species. However, the *COI* results suggest that rostral size may have been independently reduced in the *Chorocaris/Rimicaris* lineage and the *Mirocaris* lineage, making this feature homoplastic. Clearly, evaluating the evolutionary diminution of the rostrum (and eyes; e.g., Segonzac *et al.*, 1994; Van Dover, 1995; Gebruk *et al.*, 1997) requires a comprehensive global representation of bresiliid species and not just representatives of a single ridge axis. A direct comparison of morphology and molecular data would be extremely informative; however, constructing morphological matrixes from the present literature would be extremely difficult and premature, given the wide-ranging expression of numerous morphological characters possessed by these shrimp species and the current lack of agreement on the relative importance of these individual characters. This notwithstanding, the present genetic data question the phylogenetic utility of these morphological characters and strongly support *Chorocaris* and *Opaepele* as likely synonyms of *Rimicaris* (Fig. 2).

TABLE 4

Pairwise Distance Comparisons for Nucleotide Data (above Diagonal Genetic Distances Corrected with the Kimura Two-Parameter Model; below Diagonal Absolute Pairwise Differences)

	Re	Cc	Cv	Ol	Mf (MG)	Mf (BSa)	Mf (BSb)	Mf (LS)	Mk (BS)	Mk (TAG)	Mk (LOG)	LS	As	Am	Al	A. sp.	Pt	N(e?)	Lc	Ptr	Cs	Sh
R. exoculata	—	0.011	0.058	0.098	0.181	0.174	0.176	0.181	0.179	0.181	0.183	0.177	0.165	0.174	0.186	0.163	0.254	0.265	0.253	0.267	0.277	0.235
C. chacei	7	—	0.067	0.1	0.183	0.176	0.178	0.183	0.181	0.187	0.189	0.179	0.169	0.174	0.183	0.163	0.256	0.263	0.255	0.272	0.272	0.23
C. vandoverae	33	38	—	0.112	0.19	0.187	0.19	0.19	0.187	0.19	0.192	0.183	0.169	0.182	0.201	0.178	0.263	0.27	0.261	0.261	0.277	0.232
O. loihi	54	55	61	—	0.205	0.203	0.2	0.205	0.203	0.205	0.207	0.212	0.193	0.18	0.196	0.184	0.279	0.273	0.267	0.276	0.275	0.269
M. fort. (MG)	96	97	100	107	—	0.005	0.007	0.003	0.003	0.008	0.007	0.029	0.203	0.205	0.219	0.215	0.252	0.246	0.249	0.261	0.25	0.241
M. fort. (BSa)	93	94	99	106	3	—	0.005	0.008	0.008	0.013	0.012	0.034	0.203	0.198	0.216	0.212	0.254	0.238	0.252	0.259	0.247	0.236
M. fort. (BSb)	95	95	100	105	4	3	—	0.003	0.01	0.012	0.013	0.247	0.201	0.196	0.21	0.206	0.25	0.241	0.247	0.259	0.247	0.236
M. fort. (LS)	96	97	100	107	2	5	2	—	0.007	0.008	0.01	0.025	0.201	0.201	0.214	0.211	0.247	0.246	0.244	0.261	0.25	0.241
M. keld. (BS)	95	96	99	106	2	5	6	4	—	0.012	0.01	0.028	0.201	0.203	0.216	0.213	0.254	0.243	0.251	0.263	0.247	0.238
M. keld. (TAG)	96	99	100	107	5	8	6	5	7	—	0.002	0.03	0.201	0.208	0.219	0.217	0.252	0.25	0.254	0.261	0.255	0.243
M. keld. (LOG)	97	100	101	108	4	7	8	6	6	1	—	0.032	0.203	0.21	0.221	0.219	0.254	0.253	0.256	0.263	0.257	0.246
L. Strike (n. sp.)	94	95	97	110	17	20	17	15	17	18	19	—	0.202	0.193	0.206	0.211	0.254	0.257	0.257	0.274	0.254	0.241
A. stactophila	88	90	90	101	106	106	105	105	105	105	106	105	—	0.161	0.189	0.158	0.264	0.236	0.262	0.237	0.274	0.232
A. markensis	92	92	96	95	107	104	103	105	106	108	109	101	86	—	0.07	0.102	0.263	0.256	0.258	0.263	0.273	0.246
A. lusca	97	96	104	40	113	112	109	111	112	113	114	107	99	40	—	0.124	0.276	0.263	0.272	0.294	0.292	0.255
A. sp.	87	87	94	57	111	110	107	109	110	112	113	109	85	57	68	—	0.244	0.238	0.244	0.261	0.259	0.254
P. tarda	128	129	132	138	128	129	127	126	129	128	129	129	133	132	137	124	—	0.267	0.072	0.238	0.254	0.259
N. (*ensifer*?)	133	132	135	136	125	122	123	125	124	127	128	130	121	130	133	122	134	—	0.27	0.261	0.22	0.272
L. carinatus	128	129	131	134	127	128	126	125	128	129	130	135	132	130	136	124	41	135	—	0.215	0.266	0.273
P. tridens	133	135	131	137	131	130	130	131	132	131	132	136	121	132	144	131	110	131	110	—	0.27	0.24
C. septemsp.	138	136	138	137	127	126	126	127	126	129	130	129	137	137	145	131	129	114	134	135	—	0.235
S. hispidus	120	118	119	134	123	121	121	123	122	124	125	123	119	125	129	128	122	136	136	122	121	—

Note. OTU labels presented in Fig. 2.

Thus, the taxonomic scheme used to separate *Chorocaris, Rimicaris,* and *Opaepele* needs to be critically assessed.

In contrast, both morphological evidence (Vereshchaka, 1997) and molecular data support the separation of *Mirocaris* OTUs from *Alvinocaris* and *Chorocaris.* It is noteworthy that genetic diversity within the proposed family Mirocarididae (Vereshchaka, 1997) is much less than genetic diversity in the *Rimicaris/Chorocaris/Opaepele/Alvinocaris* clades (putative Alvinocarididae, *sensu* Vereshchaka, 1997). Vereshchaka (1997) assigned *C. fortunata* as congeneric with the new species *Mirocaris keldyshi,* based in part on character comparisons shown in diagnostic drawings by Martin and Christiansen (1995). Although morphological differences are recognized between *M. fortunata* and *M. keldyshi* (Vereshchaka, 1997), these OTUs could not be distinguished with the present *COI* data. Lack of sufficient time for mitochondrial lineage sorting among the *Mirocaris* lineages may account for the observed levels of molecular polymorphism in *Mirocaris* compared to other bresiliids. Alternatively, *M. fortunata* and *M. keldyshi* may not warrant species-level separation. Population genetic studies of the two *Mirocaris* morphotypes are warranted.

Perhaps the most striking feature of the resultant tree topology was that the taxa did not cluster according to biogeographic regions (i.e., Atlantic and Pacific taxa do not form reciprocally monophyletic clades). The results of the K-H likelihood test rejected the expectation that taxa within a given ocean basin were most closely related to shrimp in the same basin. This result appears to contradict hypotheses suggesting that genetic similarity should be correlated to distance along ridge crests (e.g., Tunnicliffe and Fowler, 1996; Tunnicliffe *et al.,* 1996). Due to the dynamic nature of ridge crest systems (e.g., episodic tectonic movements and changes in historical connections), it is impossible to determine the role that ancestral, but now extinct, taxa may have played in structuring the phylogeny. To further test this premise, we employed a Mantel test and found no correlation ($r^2 = 0.0249$) between present ridge axes distances and genetic divergence (see Tunnicliffe and Fowler, 1996).

Because vents and seeps are both chemosynthetically based ecosystems, Hecker (1985) proposed that organisms inhabiting vents and seeps may be evolutionarily related (for shrimp, see Gebruk *et al.,* 1997). Several taxa are common to both environments (Tunnicliffe and Fowler, 1996). Thus, seeps may have played a significant role in the historical biogeography of vent-associated species, as inferred for vesicomyid clams (Peek *et al.,* 1997) and mussels (Craddock *et al.,* 1995). Whereas vestimentiferan phylogeny (Black *et al.,* 1997) shows directionality (i.e., a seep-like ancestor giving rise to a vent clade), the most parsimonious explanation of the current bresiliid phylogeny is that a vent-like ancestor gave rise to the seep lineage *A. stactophila* (supported by a K-H test and bootstrap values). However, before firm conclusions can be drawn about the role of seeps in shrimp biogeography and evolution, additional seep-associated species (e.g., *Alvinocaris muricola,* Florida Escarpment) must be examined.

Beyond the present biogeographic issues, understanding the origin and date of organismal radiations is of great interest because certain hydrothermal vent taxa have been proposed to be relicts of Paleozoic and Mesozoic faunas. These hypotheses are based mainly on the assessment of slit limpets and stalked barnacles found at vents (McLean, 1985, and Newman, 1985, respectively). While Silurian vent-associated metazoans are thought to have existed (Little *et al.,* 1997), there is no evidence to date linking present-day vent-endemic taxa to these pre-Cenozoic communities. Molecular estimates of divergence suggest that the vent-endemic bresiliid shrimp radiated not more than 20 million years ago, roughly 45 million years after the Cenozoic began. Compared to age estimates for other vent groups based on *COI* (vestimentiferan tubeworms $<$100 million years, Black *et al.,* 1997; vesicomyid clams $<$50 million years, Peek *et al.,* 1997), these vent shrimp comprise the youngest vent- and seep-associated diversification observed to date.

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REFERENCES

Black, M. B., *et al.* (1997). Molecular systematics of vestimentiferan tubeworms from hydrothermal vents and cold-water seeps. *Mar. Biol.* **130:** 141–149.

Chace, F. A. (1992). On the classification of the caridea (Decapoda). *Crustaceana* **63:** 70–80.

Craddock, C. *et al.* (1995). Evolutionary relationships among deep-sea mytilids (Bivalvia: Mytilidae) from hydrothermal vents and cold-water methane/sulfide seeps. *Mar. Biol.* **121:** 477–485.

Crosnier, A., and Forest, J. (1973). Les Crevettes Profondes de L'Atlantique Oriental Tropical. *In* "Faune Tropicale," pp. 409. O.R.S.T.O.M., Paris.

de Saint Laurent, M. (1984). Crustacea decapoda from the hydrothermal vents of the East Pacific Rise at 13°N, collected during the French Biocyatherm Expedition. *C.R. Acad. Sci. Paris* **299:** Série III, No. 9: 355.

Douglas, M. E., and Endler, J. A. (1982). Quantitative matrix

comparisons in ecological and evolutionary investigations. *J. Theoret. Biol.* **99:** 777–795.

Doyle, J. J., and Dickson, E. (1987). Preservation of plant samples for DNA restriction endonuclease analysis. *Taxon* **36:** 715–722.

Felsenstein, J. (1985). Confidence limits on phylogenies: An approach using the bootstrap. *Evolution* **39:** 783–791.

Felsenstein, J. (1993). PHYLIP (phylogeny inference package). Version 3.5c Department of Genetics, University of Washington, Seattle, WA.

Folmer, O., Hoeh, W. R., Black, M. B., and Vrijenhoek, R. C. (1994). Conserved primers for PCR amplification of mitochondrial DNA from different invertebrate phyla. *Mol. Mar. Biol. Biotech.* **3:** 294–299.

France, S. C., and Kocher, T. D. (1996). DNA sequencing of formalin-fixed crustaceans from archival research collections. *Mol. Mar. Biol. Biotech.* **5:** 304–313.

Gebruk, A. V., Galkin, S. V., Vereshchaka, A. L., Moskalev, L. I., and Southward, A. J. (1997). Ecology and biogeography of the hydrothermal vent fauna of theMid-Atlantic Ridge. *Adv. Mar. Biol.* **32:** 94–143.

Geistdoerfer, P., Auzende, J. M., Batiza, R., Bideau, D., Cormier, M., Fouquet, Y., *et al.* (1995). Hydrothermalisme et communautés animales associées sur la dorsale du Pacifique oriental entre 17°S et 19°S (campagne Naudur, décembre 1993). *C. R. Acad. Sci. Paris,* **Série IIa:** 47–54.

Halanych, K. M., and Robinson, T. J. (1997). Phlogenetic relationships of cottontails (*Sylvilagus,* Lagomorpha): Congruence of 12S rDNA and cytogenetic data. *Mol. Phylogenet. Evol.* **7:** 294–302.

Hasegawa, M., Kishino, H., and Yano, T. (1985). Dating of the human-ape splitting by a molecular clock of mitochondrial DNA. *J. Mol. Evol.* **22:** 160–174.

Hecker, B. (1985). Fauna from a cold sulfur seep in the Gulf of Mexico: Comparison with hydrothermal vent communities and evolutionary implications. *Biol. Soc. Wash. Bull.* **6:** 465–474.

Holthuis, L. B. (1955). The recent genera of the caridean and stenopodean shrimps (Class Crustacea, Order Decapoda, Supersection Natantia) with keys to their determination. *Zool. Verhand. Leiden* **26:** 1–57.

Juniper, S. K., Tunnicliffe, V., and Desbruyères, D. (1990). Regional-scale features of northeast Pacific, East Pacific Rise, and Gulf of Aden vent communities. *In* "Gorda Ridge: A Seafloor Spreading Center in the United States Exclusive Economic Zone" (G. R. McMurray, Ed.), pp. 265–278. Springer-Verlag, New York.

Kikuchi, T., and Ohta, S. (1995). Two caridean shrimps of the families Bresiliidae and Hippolytidae from a hydrothermal field on the Iheya Ridge, off the Ryukyu Islands, Japan. *J. Crust. Biol.* **15:** 771–785.

Kishino, H., and Hasegawa, M. (1989). Evaluation of the maximum likelihood estimate of the evolutionary tree topologies from DNA sequence data, and the branching order in Hominoidea. *J. Mol. Evol.* **29:** 170–179.

Knowlton, N., Weigt, L. A., Solorzano, L. A., Mills, D. K., and Bermingham, E. (1993). Divergence in proteins, mitochondrial DNA, and reproductive compatibility across the isthmus of Panama. *Science* **260:** 1629–1631.

Kumar, S., Tamura, K., and Nei, M. (1993). Mega: Molecular Evolutionary Genetics Analysis, version 1.01, The Pennsylvania State University, University Park, PA.

Little, C., Herrington, R. J., Maslennikov, V. V., Morris, N. J., and Zayko, V. V. (1997). Silurian hydrothermal-vent community from the southern Urals, Russia. *Nature* **385:** 146–148.

Maddison, W. P., and Maddison, D. R. (1992). MacClade: analysis of phylogeny and charcter evolution, Sinauer Associates, Sunderland, MA.

Martin, J., Signorovitch, J., and Patel, H. (1997). A new species of *Rimicaris* (Crustacea: Decapoda: Bresiliidae) from the Snake Pit hydrothermal vent field on the Mid-Atlantic Ridge. *Proc. Biol. Soc. Wash.* **110:** 399–411.

Martin, J. W., and Christiansen, J. C. (1995). A new species of the shrimp genus *Chorocaris* Martin and Hessler, 1990 (Crustacea: Decapoda: Bresiliidae) from hydrothermal vent fields along the Mid-Atlantic Ridge. *Proc. Biol. Soc. Wash.* **108:** 220–227.

Martin, J. W., and Hessler, R. R. (1990). *Chorocaris vandoverae,* a new genus and species of hydrothermal vent shrimp (Crustacea: Decapoda; Bresillidae) from the western Pacific. *Contrib. Sci. Nat. Hist. Mus. L.A. County* **417:** 1–11.

McLean, J. H. (1985). Preliminary report on the limpets at hydrothermal vents. *Biol. Soc. Wash. Bull.* **6:** 159–166.

Muse, S. V., and Gaut, B. S. (1994). A likelihood approach for comparing synonymous and nonsynonymous nucleotide substitution rates, with application to the chloroplast genome. *Mol. Biol. Evol.* **11:** 715–724.

Newman, W. A. (1985). The abyssal hydrothermal vent invertebrate fauna, a glimpse of antiquity? *Biol. Soc. Wash. Bull.* **6:** 231–242.

Olsen, G. J., Matsuda, H., and Hagstrom, R. (1994). FastDNAml: A tool for construction of phylogenetic trees from DNA sequences using maximum likelihood. *Cabio* **10:** 41–48.

Peek, A. S., Gustafson, R. G., Lutz, R. A., and Vrijenhoek, R. C. (1997). Evolutionary relationships of deep-sea hydrothermal vent and cold-water seep clams (Bivalvia: Vesicomyidae): Results from mitochondrial cytochrome oxidase subunit I. *Mar. Biol.* **130:** 151–161.

Rona, P. A. (1986). Direct observations of Atlantic black smokers. *EOS* **67:** 1327.

Schram, F. R. (1986). Caridea. *In* "Crustacea," pp. 606. Oxford Univ. Press, New York.

Segonzac, M., de Saint Laurent, M., and Casanova, B. (1994). The enigma of the trophic behaviour of alvinocaridid shrimps from hydrothermal vent sites on the Mid-Atlantic Ridge. *Cash. Biol. Mar.* **34:** 535–571.

Shank, T. M., Lutz, R. A., and Vrijenhoek, R. C. (1998a). Molecular systematics of shrimp (Decapoda; Bresiliidae) from deep-sea hydrothermal vents: I. Enigmatic "small orange" shrimp from the Mid-Atlantic Ridge are juvenile *Rimicaris exoculata. Mol. Mar. Biol. Biotech.* **7:** 88–96.

Shank, T. M., Fornari, D. J., Von Damm, K. V. D., Lilley, M. D., Haymon, R. H., and Lutz, R. A. (1998b). Temporal and spatial development of nascent deep-sea hydrothermal vent communities at 9° 50'N (EPR). *Deep-Sea Res.* **45:** 465–515.

Shiozawa, D. K., Kudo, J., Evans, R. P., Woodward, S. R., and Williams, R. N. (1992). DNA extraction from preserved trout tissues. *Great Britain Natur.* **52:** 29–34.

Swofford, D. L. (1993). PAUP: Phylogenetic Analysis Using Parsimony, Version 3.11, Computer program distributed by the Illinois Natural History Survey, Champaign, IL.

Thompson, J. R. (1996). The caridean superfamily Bresilioidea (Decapoda:Natantia). A revision and a discussion of its validity and affinities. *Crustaceana* **11:** 129–140.

Thompson, J. D., Higgins, D. G., and 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.

Tunnicliffe, V., and Fowler, M. R. (1996). Influence of sea-floor spreading on the global hydrothermal vent fauna. *Nature* **379:** 531–533.

Tunnicliffe, V. C., Fowler, M. R., and McArthur, A. (1996). Plate tectonic history and hot vent biogeography. *In* "Tectonic, Magmatic, Hydrothermal and Biological Segmentation of Mid-Ocean Ridges" (C. J. MacLeod, P. Tyler, and C. L. Walker, Eds.), pp. 225–238. Geol. Soc. Spec. Publ., London.

Van Dover, C. L. (1995). Ecology of Mid-Atlantic Ridge hydrothermal

vents. *In* "Hydrothermal Vents and Processes" (C. J. MacLeod, P. Tyler, and C. L. Walker, Eds.), pp. 257–294. Geol. Soc. Spec. Publ., London.

Vereshchaka, A. L. (1996a). A new genus and species of caridean shrimp (Crustacea:Decapoda:Alvinocarididae) from north atlantic hydrothermal vents. *J. Mar. Biol. Assn. U.K.* **76:** 951–961.

Vereshchaka, A. L. (1996b). Comparative analysis of taxonomic composition of shrimps as edificators of hydrothermal communities in the Mid-Atlantic Ridge. *Doklady Biol. Sci.* **351:** 576–578.

Vereshchaka, A. L. (1997). A new family for a deep-sea caridean shrimp from North Atlantic hydrothermal vents. *J. Mar. Biol. Assn. U.K.* **77:** 425–438.

Williams, A. B. (1988). New marine decapod crustaceans from waters influenced by hydrothermal discharge, brine, and hydrocarbon seepage. *Fish. Bull.* **86:** 263–287.

Williams, A. B., and Dobbs, F. C. (1995). A new genus and species of caridean shrimp (Crustacea:Decapoda:Bresiliidae) from hydrothermal vents on Loihi Seamount. *Proc. Biol. Soc. Wash.* **108:** 228–237.

Williams, A. B., and Rona, P. A. (1986). Two new caridean shrimps (Bresiliidae) from a hydrothermal vent field on the Mid-Atlantic Ridge. *J. Crust. Biol.* **6:** 446–462.

Zarenkov, N. A. (1976). Fauna of decapod crustaceans of American waters in the southern hemisphere. *Sov. J. Mar. Biol.* **2:** 277–285.