RELATIONSHIPS AND BIOGEOGRAPHY OF ANTILLEAN CICHLIDS

Abstract

Geological reconstructions proposed for the Greater Antilles fit into two major categories, each with different biogeographic consequences. One category of reconstructions suggests that Early Oligocene landbridge connected the Greater Antillean islands to northern South America. The other suggests a period of coalescence in the Mesozoic of some of the Antillean islands with portions of Central America. A molecular phylogenetic analysis that includes the endemic cichlid faunas of the Greater Antilles, as well as from South and Central America, results in a phylogeny that is congruent with the later reconstruction. Also, this analysis finds that a dispersal scenario is not the simplest or most robust explanation for the presence of these cichlids on the Antilles.

Introduction

Cichlidae is a monophyletic group of perciform fishes with a species diversity approaching 2,000 described species (Kullander 1998, Sparks 2001). The group is widely distributed on former Gondwanan fragments: South and Central America (300 spp.), Africa (1,000 spp.), Madagascar (>18spp.), India (3spp.), and the Middle East (3 spp.) (Figure 1). There has been resurgence of discussion of the historical biogeography of this group in recent years with the discovery of the oldest known cichlid fossils and new molecular evidence (Murray 2001, 2001b; Sparks 2001; Vences 2001; Kumazawa *et al.* 2000; see review in Chakrabarty 2003).

Recent phylogenetic work finds that each continental assemblage of cichlids forms its own monophyletic group with the exception of Madagascar (Sparks 2001; Farias *et al.* 1999, 2000; Streelman *et al.* 1998; Zardoya *et al.*1996, see Chakrabarty 2003

for alternatives and explanation). This has led some to propose a Cretaceous origin of Cichlidae before the break-up of Gondwana (Sparks 2001; Stiassny 1991). Dispersalists point out the minimum age of the Cichlidae based on the oldest known fossils is Eocene (45 million years ago), and use the lack of fossil cichlids before this time as evidence for recent dispersal events to explain current cichlid distributions (Murray 2001b; Lundberg 1993).

Malagasy cichlids are shown to be sister to cichlids on India in every analysis to include them (Stiassny *et al.* 2001; Sparks 2001; Farias *et al.* 1999, 2000; Streelman *et al.* 1998; Zardoya 1996; Cichocki 1976; Streelman & Karl 1997; Lippitsch 1995; Oliver 1984; Kullander 1998). This Malagasy-Indian relationship is the most well corroborated sister grouping of cichlids and in the dispersalist framework this relationship would mean that cichlids were capable of dispersing across the entire Indian Ocean but not across the Mozambique Channel (430 km wide at its narrowest point) to Africa.

Cichlids are primarily a strictly freshwater water group with some species able to tolerate brackish waters and a few that are considered salt tolerant (Murray 2001b). With no known cases of cichlids dispersing from one landmass to another across a salt-water divide, the marine environment should still be considered a barrier to cichlid dispersal.

There are landmasses that are not Gondwanan in origin that have cichlids, these are the islands of Cuba and Hispaniola (Figure 2). These islands have a complex geological history and there are a number of alternative hypotheses of how the fauna and flora of these islands evolved. The mechanism that allowed cichlids to invade these islands will be explored in this study.

The Greater Antilles

The relationships of the Greater Antillean cichlid fauna are important because the Antilles are not geologically Gondawanan in origin, although they may have in various times of their history been connected to Gondwanan fragments. Leon Croizat's (1962)

2

metaphor of vicariance biogeography being like reconstructing a pane of glass that has been repeatedly shattered seems particularly relevant to the Greater Antilles.

Geologically speaking, the Greater Antilles rest upon a small plate located between the much larger North America, South American and Cocos and Nazca plates. The Caribbean plate itself can be divided into a series of minor plates that have separated and merged at various times in their history (Perfit & Williams 1989). Despite their history and position on a tectonic plate, these islands are usually labeled as "oceanic." This nomenclature, like "secondary freshwater," assumes *a priori* that overwater dispersal is the only mechanism for organisms to populate these islands. Paulay (1994) defined oceanic islands as islands that have never been connected to a mainland continent and therefore are populated solely by dispersal. Given the aforementioned tectonic reconstructions, this definition does not fit the Greater Antilles.

Alternative Plate Tectonic Reconstructions for Middle America

Two alternative tectonic reconstructions involving a land connection between the Greater Antilles and either mainland South America or Central America will be discussed. Rosen (1975, 1985) developed a vicariance model based on Malfait and Dinkelman (1972) and Tedford's (1974) geological models of the Antilles. This view gives a Mesozoic age to the fauna of the Greater Antillean islands. This reconstruction of the Antilles, supported by many others (Pindell *et al.* 1988; Ross & Scotese 1988; Pindell & Barrett 1990; Pitman *et al.* 1993; Schuchert 1935; Dickinson & Coney 1980; but see alternatives in Hedges 1982), proposes that the Greater Antilles originated in the Pacific Ocean as a series of islands that connected North and South America as a temporary closely linked island chain circa 80 million years ago, in a position occupied by present day Central America. This arc then broke in the early Cenozoic and drifted to its current position. In Rosen's model, Jamaica, the Caymans, southwest Hispaniola, and western Cuba were connected to the southern and eastern parts of Yucatan (northern Central America) until they separated at the end of the Cretaceous, 65 million years ago.

In the alternative view proposed by Iturralde-Vinent and MacPhee (1999) based on geological evidence, a short-lived connection between the Greater Antillean Islands (Cuba, Hispaniola, Puerto Rico and possibly Jamaica) and northwest South America existed circa 32 million years ago. The authors name this Early Oligocene landbridge GAARlandia (from Greater Antilles + Aves Ridge). The consequence of this alternative is that the Greater Antillean Island chain would have had a more recent connection with South America than with Central America. The authors do not believe that there was a land connection between these islands and Central America in a way that would explain the current faunal distributions. This sets up two testable alternative hypotheses, between the relationships of Antillean cichlids with either Central or South American lineages. Non-congruence of phylogenetic pattern and sequence of divergence would favor dispersal.

Cichlids on the Antilles

There are five known cichlids from the Antilles, *Cichlasoma tetracanthus*, *C. ramsdeni*, *C. haitiensis*, *C. vombergi* and *C. woodringi* (see Myers 1928 and Darlington 1957 for discussion on other possible species, and distributions).   The first two are restricted to Cuba, and the others to Hispaniola. The fossil *C. woodringi* is either Upper or Middle Miocene (23 to 5 mya) in age (Rivas 1986; Myers 1928) or Pliocene (Casciotta & Arratia 1993; Murray 2001b).   It has the notoriety of being the only known freshwater fossil from the Antilles (Burgess & Franz 1989). Bussing (1985) and Rivas (pers. comm. in Burgess & Franz 1989) comment that this fossil is indistinguishable from *C. haitiensis,* an extant Hispaniolan species. Bussing (1985) also likens it to the *Parapetenia* of Central America. The generic name of these species (*Cichlasoma*) is sometimes referred to as *Nandopsis*, these names are homonyms and will be used interchangeable in this paper.

The genus *Cichlasoma* (*sensu lato*)*,* to which all the Antillean cichlids belong, dominates the Central American cichlid fauna (75 of about 100 or more species) (Roe *et*

al. 1997; Miller 1966; Martin & Bermingham 1998; Kullander 1983). This genus is also found throughout South America and north to Texas, but its diversity in Central America is unmatched (Bussing 1985).

To date no formal phylogenetic analysis has included the Cuban and Hispaniolan species with Central American and South American species. Rosen (1975) presented a cladogram that had a sister relationship between Central America and the Antillean cichlid fauna, however it included only the Central American and Antillean fauna. This four taxon cladogram was cited using a 'personal communication' from Cichocki who did not include this analysis in any published material. Loiselle (1985) also lists Central American sister species for the Antillean cichlids, but does not discuss his phylogenetic method; it appears as if his notion is based solely on his own similarity judgment. Without a phylogenetic diagnosis, we lack a measure for selecting between alternative mechanisms for explaining this disjunct distribution. Before discussing the results of the analyses done here, it is important to discuss some important contributions on prior hypotheses of vicariance and dispersal mechanisms for these cichlids and freshwater fauna of this area; they are reviewed below.

Overwater dispersal

Myers (1938, 1966; see also Darlington 1957) hypothesized that the freshwater fishes of the West Indies dispersed from Central America, mainly due to the salt tolerance of these 'secondary freshwater fishes.' Myers believed the islands formed *in situ* without connection to other landmasses. Rivas (1986) noted that the native cichlids on Cuba and Hispaniola are known only from landlocked freshwater habitats, never brackish or marine habitats. Bussing (1985) and Martin and Bermingham (1998)

hypothesize that South American and Central American cichlids may have dispersed around the continental landmasses by migrating along coastlines, pointing again to salt tolerance in these cichlids. Kullander (1983) gives the only mention of Neotropical cichlids being caught in brackish water. Endemism of cichlid species on Cuba and Hispaniola implies that either (1) they speciated there, presumably long ago, (2) there have been no successful dispersal events from mainland to island or from island to island, or (3) extinctions have left this pattern of endemism (so called "reciprocal monophyly").

A number of authors have stated that Cuba, particularly its eastern half, was once united with Hispaniola in the early history of the Caribbean (Williams 1989; Perfit & Williams 1989). According to Pitman *et al.* (1993), Cuba and Hispaniola did not separate until a shearing in the late Middle Eocene. Nearly 90 % of the 71 species of Antillean freshwater fishes occur on Cuba and Hispaniola (Burgess & Franz 1989). Sixty-five of these are endemic to an island or island group (Burgess & Franz 1989). Surprisingly, Puerto Rico, the fourth largest Antillean island, separated from Hispaniola by only the narrow Mona Passage (130 km), is completely lacking in native freshwater fishes. Puerto Rico does have available habitats, as an introduced African cichlid and many other introduced species maintain populations there (Burgess & Franz 1989). Fishes dispersing from Central or South America would also probably reach Jamaica or the lower Antilles first because of their location (Fig.8). There are no cichlids on Jamaica (it does have six other native freshwater species), and there are only two native freshwater fishes on the entire Lesser Antilles.   The absence of cichlids from Jamaica and particularly Puerto Rico does not bode well for the Iturralde-Vinent and MacPhee (1999) hypothesis, which posits a connection between Cuba, Hispaniola and South America, with Jamaica and

Puerto Rico in-between. A complete extinction of the freshwater fish fauna would be required on Puerto Rico, after the suggested landbridge, for their hypothesis to be plausible.

Congruent cladograms for the Greater Antilles

Based on his vicariance model, Rosen (1975) gave a Mesozoic minimum age to the freshwater fish fauna of the Antilles including cichlids, atherinids (silversides), poeciliids and other Cyprinodontiformes, synbranchid eels and gars. Rauchenberger (1988) attempted to create a composite area cladogram from 12 other cladograms using these taxa to support Rosen's vicariance model. Most of the trees she used in her analysis are poorly resolved (the cichlid area cladogram she used is an uninformative polytomy), as she notes herself. Only the *Gambusia* tree (a poeciliid) provided much resolution to her composite. Unfortunately the *Gambusia* tree she cites (but does not show) from Fink (1971, 1971b) ignores some key elements of the original cladogram (Fink pers. comm.). Her analysis also included only one South American species, which, due to its placement at the base of the cladogram, did not affect the composite tree. It is not surprising then that she found a close relationship between the Antillean and Central American taxa (the only possibility, given her sampling).

Iturralde-Vinent and MacPhee (1999) give an age no older than the Middle Eocene for the Greater Antillean islands (including Cuba and Hispaniola), therefore claiming that that fauna must be younger than this age. On this basis they reject Rosen's (1975) Mesozoic vicariance hypothesis. Although they agree that these island masses may be as old as the Jurassic, they believe that subduction into tectonic nappes, subsidence into the water, thrusting, folding, volcanic activity and K/T bolide impact-

related activity make comparisons of present day (or at least post-Eocene) Antillean islands and their Mesozoic precursors meaningless.

Materials & Methods

Selecting among biogeographic hypotheses

There are many methods that have been proposed for selecting between biogeographic hypotheses (see Crisci 2001). I adopt a cladistic biogeographic approach *sensu* Rosen (1978) and Nelson & Platnick (1981). This method was selected over others because it uses area cladograms that can be created from the published phylogenies reviewed here. This method was also selected over others because it is the only one that utilizes the principle of parsimony, which by minimizing assumptions finds the most efficient explanation of the evidence (Sober 1988).

The cladistic biogeographic approach assumes a shared correspondence between phylogenetic history and geological history. The relationship between these histories can be seen in congruent patterns of different taxonomic and area cladograms (cladograms with taxon names replaced by distributions) fitting a given pattern of geological history. In this method, dispersal is assumed not to explain a disjunct distribution until vicariance can be falsified (Kluge 1989; Croizat, Nelson & Rosen 1975). Vicariance is a more parsimonious interpretation than dispersal for congruent area cladograms of different taxa, because the congruence can be explained by a single event *(i.e.*, the rifting of a continent or orogeny). The same interpretation of distributions by dispersal would require concordant dispersal in the same sequence for many diverse taxa (Fig. 2, Fig. 3).

The essence of vicariance biogeography is that barriers arise secondarily to divide up species. Vicariance events, because they are tied to earth history, can only be

8

supported by a very limited range of phylogenetic patterns. Dispersal scenarios, because they can occur without any underlying congruent process, can be claimed to support an unlimited range of phylogenetic patterns.

Area cladograms that fit a hypothesized geographical fragmentation sequence may support vicariance, or at least do not falsify it. All distribution patterns can be explained by dispersal. Dispersal scenarios therefore should not be employed unless vicariance scenarios have been falsified.

Vicariance scenarios for freshwater fishes have the following potential falsifiers: (1) the phylogenetic pattern (sequence of lineage divergence) does not follow the timing of known geological processes (i.e. the sequence of fragmentation) (Figure 3), (2) members of particular lineages are younger than hypothesized related vicariance events (3) a species of the group under study is found on either side of a supposed barrier to dispersal (4) molecular clocks or sequence divergence times reliably show that lineages have diverged after the particular vicariant events under study.

Dispersal will be the favored mechanism to explain a disjunct distribution when falsifiers of vicariance - by adding assumptions to a vicariance hypothesis - make dispersal a more parsimonious alternative. The possibility also exists that the current evidence is insufficient to select between alternative explanations of disjunct distributions.

Molecular techniques

Specimens were purchased from an aquarist company, *Tangled Up In Cichlids* or from specimens that were bought from Michigan pet stores by the author. Specimens have yet to be vouchered and officially placed in the UMMZ (Univeristy of Michigan Museum of Zoology) collections. Photographic records of the left side of the included fishes were taken before tissue samples were removed. Molecular sequences were obtained from the University of Michigan Sequencing Core after DNA purification and amplification were completed by the author. Additional sequences were obtained from GenBank. Table 1 lists the species used in this study and tissue voucher numbers and GenBank accession numbers. Outgroups included members of Labridae (wrasses), the possible sister group to Cichlidae (Streelman & Karl 1997).   Closely related Indian cichlids in the genus *Etroplus* were included to verify monophyly of the Neotropical species.

Tissue was preserved in 70% ethanol and frozen at -20°C prior to DNA extraction. DNA was extracted from fin clips, or from muscle tissue from the right side of the specimen using Proteinase K dissolution. Tissue was then purified using a Qiagen Tissue Extraction Kit following the manufactures protocol. Polymerase Chain Reaction (PCR) was performed using Platnium Taq polymerase following the manufactures protocol to amplify about 600 base pairs (bp) of 16S ribosomal subunit and a 650 bp segment of cytochrome *c* oxidase subunit I (COI). The primers 16S ar-L 5'-CGCCTGTTTATCAAAACAT-3' and 16S br-H 5'-CCGGTCTGAACTCAGATCAGT-3' (Koucher et al.1989; Palumbi 1996) were used to amplify 16S. The primers LCO1490 5'-GGTCAACAAATCATAAAGATATTGG-3' and HCO2198 5'-TAAACTTCAGGGTGACCAAAAAATCA-3' from Folmer *et al.* (1994) were used to

amplify CO1. Amplification for 16S were carried out in 30 cycles according to the following protocol denaturing for 2 minutes at 95°C, annealing at 55°C for 1 minute and an extension for 2 minutes at 72°C, with an additional terminal extension for 72°C for 10 minutes. Amplification for COI was carried out in 35 cycles to the following temperature profile: denaturing for 20 seconds at 95°C, annealing for 1 minute at 45°C and an extension for 2 minutes at 72°C, with an additional terminal extension for 72°C for 10 minutes.

Sequences were aligned (using complete alignment option) and compiled in CLUSTAL X (Thompson *et al.* 1994). Indels were treated as missing characters in all analyses.

Parsimony and Maximum likelihood analyses were carried out in PAUP* 4.0b3 (Swafford, 1998). Heuristic searches were carried out with 1000 replicates (random addition of taxa, ACCTRAN optimization). For maximum likelihood 100 replicate heuristic was done and the assumption set is as follows: the transition/transversions rate was set as 2:1; base frequencies were set to "empirical frequencies"; for the variable sites option, "equal rates for all sites option" was selected; starting values for all parameters was "parsimony based approximations", "Rogers-Swafford method" was selected for starting branch lengths.

Results

Twenty 16S rDNA sequences were obtained by the author and 22 from GenBank (Table 1). Unfortunately no cichlid COI sequences for the taxa used in this study are available on GenBank.

Figure 3 shows the strict consensus parsimony tree of the 16S data alone. Total tree length was of 856 steps. Although the Neotropical taxa are shown to be monophyletic with a bootstrap support of 80, and cichlids are monophyletic with a support of 68, there is very little resolution within the Neotropical clade. There are two possible explanations for the resolution, that either this molecule is unable to resolve this level of divergence or that the species tree is a hard polytomy. Given that the combined data set of both 16S and COI genes are better resolved, it cannot be concluded that these species relationships is an irresolvable hard polytomy.

Figure 4, shows the resulting consensus tree of a maximum parsimony heuristic analysis of 1000 bootstraps. Central and North American species form a monophyletic group that includes the Greater Antillean species. This entire clade is nested within the South American clade. Cichlidae and the Neotropical cichlids are found to be monophyletic. The Antillean cichlids do not form a clade. The Cuban *Nandopsis ramsedeni* is found to be sister to a subclade of Mexican and Central American species and the Hispaniolan species *Cichlasoma haitiensis* is in the poorly resolved grouping outside of the clade that includes *N. ramsedeni*. The consistency index is 0.471963 and the retention index 0.567878.

Figure 5 shows the maximum likelihood tree, which is largely congruent with the parsimony tree. The Mexican and Central American species still form a clade nested with the South American clade. A major distinction is the position of the Greater Antillean species. *Cichlasoma haitiensis* is recovered as sister to *C. oblongus* a Guatemalan species; *Nandopsis ramsedeni* is recovered in a similar position to where it was in the parsimony analysis as sister to a larger clade of Central and Mexican cichlids.

Conclusion

Unfortunately due to the disparity of where the Antillean cichlids were recovered in the likelihood and parsimony analyses, little conclusively can be said in regards to their biogeography. Both analyses (likelihood and parsimony) found that the Antillean cichlids do not form a clade, so it can be stated with certainty that these cichlids are derived from different lineages. The Antillean fauna therefore did not arise from a single ancestral species dispersing across a marine environment or landbridge, or from one species dispersing during a vicariant event. At least two separate dispersal events or ancestors are necessary to explain these data.

Since both analyses (Figure 4 and 5) find the Antillean cichlids nested within the Central American clade, these analyses find evidence to falsify the GAARlandia hypothesis of Itturalde and MacPhee (1999). Evidence to support this hypothesis would have been to recover the Antillean cichlids as sister to some South American clades instead of nested within the Mexican-Central American clade.

With the Central American clade nested within the South American clade and the Neotropical clade recovered as monophyletic in this and other analyses (Sparks 2001; Farias 2001; Streelman *et al.* 1998; Zardoya 1996) it is apparent that South American taxa dispersed to Central America to colonize habitats there – fitting the hypotheses of Myers (1938, 1966) and Bussing (1985). Although there is evidence against dispersal across a South American landbridge of cichlids to give rise to the Antillean cichlids, there is no evidence supporting or falsifying the Rosen coalescence hypothesis or overwater dispersal being the mechanism for cichlids getting from Central America to the Greater

13

Antilles. Both scenarios are still plausible.   However, the cladistic biogeographic approach uses a simplicity criterion to favor one of these scenarios over the other.

Dispersal from Central America across the marine environment given the topologies of these analyses would require that the species that dispersed either speciated on these islands from at least two separate dispersal events, or that the species that currently are on the islands had their source mainland populations subsequently extirpated. There is no fossil record of any Cuban or Hispaniolan species on Central America or vice versa. Interestingly the relationship of the Antillean cichlids rules out successful dispersal events from one island to the other. Not only are there not shared populations of species between the islands, but also the species on Cuba and Hispaniola are not even closely related.

That the Antillean cichlids are not monophyletic still fits the Rosen hypothesis of drift vicariance. According to Rosen (1975, 1985) Jamaica, the Caymans, southwest Hispaniola, and western Cuba were connected to different parts of southern and eastern parts of the Yucatan and northern parts of Central America. This scenario would entail different cichlid species giving rise to the separated lineages in Cuba and Hispaniola.

The simplest scenario is that of Rosen (1975, 1985) rather than of a dispersalist scenario of overwater dispersal. Given the dispersalist view point cichlids on the Antilles would have had to disperse the hundreds of kilometers from Central America to Hispaniola and Cuba in two separate dispersal events, but not the 77 kilometers between these islands. The vicariance approach requires a unique event, to explain the current Antillean distributions of cichlids, atherinids (silversides), poeciliids and other Cyprinodontiformes, synbranchid eels and gars, since they all have largely congruent

cladograms (Rosen 1975; Rauchenberger 1988). The dispersalist scenario assumes each of these lineages independently dispersed from Central America to these islands.

There are several interesting non-biogeographic outcomes recovered in the tree topologies that will interest aquarists. *Cichlasoma* (= *Archocentrus*) *citrenellum* is perhaps the most popular aquarium trade cichlid. Three individuals of this species were samples in the analyses. *C. citrenellum*10 (as labeled in the trees and Table 1) is from a pet store, and has presumable been in the aquarium trade for many generations leaving it susceptible to hybridization with other captive bred species. *C. citrenellum*1 is an F1 generation from wild caught individuals, and *Archocentrus citrenellum*AF0490 is a GenBank sequence from this same species reported with its synonomized generic name. The later two individuals are identical but *C. citrenellum*10 is recovered as forming a sister relationship with *Amphillophus lyonsi*. Supporting that at least some aquarium populations of this species, sold as the "Midas cichlid" for its golden color, are hybrids of other species. Another popular aquarium population known as the "blue dempsey" *Nandopsis octofasciatus* has been reported in the aquarium literature as a hybrid of two distantly related taxa and not related to the common "dempsey" *Cichlasoma octofasciatus*. In these analyses the "blue dempsey" is found to be identical to individuals reported in GenBank and other populations of "dempsey" supporting the idea that this population is simply a color morph of the common wild population.

Clearly the other Antillean species need to be sampled, as should the COI data from those species from which only 16S data is sampled.   Finding that the additional two species from the islands are not related to the two species sampled here would favor a dispersalist scenario over a vicariant one.   A vicariance scenario would predict that the

ancestor of the species on each island arrived there as part of a single vicariant event and speciated on that islands. A morphological analysis would be useful because the fossil species *Cichlasoma woodringi* should be included in these analyses, and a morphological study would act as a congruent and independent test of the congruent molecular topologies found here.

References Cited

Burgess, G.H. and Franz, R. (1989) Zoogeography of the Antillean freshwater fish fauna In: *Biogeography of the West Indies: Past, Present, and Future*. (ed C.A.Woods). Sandhill Crane Press, Gainesville, pp. 263-304.

Bussing, W.A. (1985) Patterns of distribution of the Central American ichthyofauna. In: *The Great American Biotic Interchange*. (eds F.G. Stehli and S.D. Webb) Plenum, New York, pp. 453-473.

Casciotta, J. and Arratia, G. (1993) Tertiary cichlid fishes from Argentina and reassessment of the phylogeny of New World cichlids (Perciformes: Labroidei). *Kaupia - Darmstädter Beiträge zur Naturgeschichte* **2**, 195-240.

Chakrabarty, P. (2003) Cichlid biogeography: comment and review. *Fish and Fisheries*: in review.

Cichocki, F.P. (1976) *Cladistic history of cichlid fishes and reproductive strategies of the American genera* Acarichthys, Biotodoma, *and* Geophagus. Vol 1. Unpublished PhD Dissertation, The University of Michigan, Ann Arbor.

Crisci, J.V. (2001) The voice of historical biogeography. *Journal of Biogeography* **28**,157-168.

Croizat, L. (1962) *Space, Time, Form: The Biological Synthesis*. Published by the author, Caracus.

Croizat, L., Nelson, G. and Rosen, D.E. (1974) Centers of origin and related concepts. *Systematic Zoology* **23**, 265-287.

Darlington, P.J. Jr. (1957) *Zoogeography: The Geographical Distribution of Animals*. Chapman & Hall, London.

Dickenson, W.R. and Coney, P.J. (1980) Plate tectonic constraints on the origin of the Gulf of Mexico. In: *The origin of the Gulf of Mexico and the early opening of the central North Atlantic Ocean* (ed R.H. Pilger Jr.) Proceedings of a symposium at LSU, Baton Rouge, Louisiana. pp. 27-36.

Farias, I.P., Ortí, G., Sampaio, I., Schneider, H. and Meyer, A. (1999) Mitochondrial DNA phylogeny of the family Cichlidae: monophyly and fast molecular evolution of the Neotropical assemblage. *Journal of Molecular Evolution* **48**, 703-711.

Farias, I.P., Ortí, G. and Meyer, A. (2000) Total evidence: molecules, morphology, and the phylogenetics of cichlid fishes. *Journal of Experimental Zoology (Mol. Dev. Evol.)* **288**, 76-92.

Fink, W.L. (1971) A revision of the *Gambusia puncticulata* complex (Pisces: Poeciliidae) *Publications of the Gulf Coast Research Laboratory Museum*, **2**, 11-46.

Fink, W.L. (1971b) A revision of the *Gambusia nicaraguensis* species group (Pisces: Poeciliidae). *Publications of the Gulf Coast Research Laboratory Museum* **2**, 47- 77.

Folmer, O., Black, W., Hoeh, W., Lutz, R., Vrijenhoek, R. (1994). Dna primers for amplification of mitochondrial cytochrome *c* oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology* **3**, 294- 299.

Hedges, S.B. (1982) Caribbean biogeography: Implications of recent plate tectonic studies. *Systematic Zoology* **31**, 518-522.

Iturralde-Vinent, M.A. and MacPhee, R.D.E. (1999) Paleogeography of the Caribbean region: implications for Cenozoic biogeography. *Bulletin of the American Museum of Natural History* **238**, 1- 95.

Kluge, A.G. (1989) A concern for evidence and a phylogenetic hypothesis of relationships among Epicrates (Boidae, Serpentes) *Systematic Zoology* **38**,7-25.

Koucher, T.D., Thomas, W.K., Meyer, A., Edwards, S.V., Paabo, S., Villablanca, F.X. and Wilson, A.C. (1989). Dynamics of mitochondrial DNA evolution in animals: amplification and sequencing with conserved primers. *Proceedings of the National Academy of Sciences USA.* **86**, 6196-6200.

Kullander, S.O.(1983) *A revision of the South American cichlid genus Cichlasoma (Teleostei: Cichlidae).* The Swedish Museum of Natural History, Stockholm.

Kullander, S.O. (1998) A phylogeny and classification of the South American Cichlidae (Teleostei:Perciformes) In: *Phylogeny and Classification of Neotropical Fishes* (eds L.R. Malabarba, R.E. Reis, R.P. Vari, Z.M. Lucena, C.A.S. Lucena) Edipucrs., Porto Allegre, pp. 461- 498.

Kumazawa, Y., Yamaguchi, M. and Nishida, M. (2000) Mitochondrial molecular clocks and the origin of euteleostean biodiversity: familial radiation of perciforms may have predated the Cretaceous/Tertiary boundary. In: *The Biology of Biodiversity* (ed M.Kato) Springer-Verlag, Tokyo, pp. 35 – 52.

Lippitch, E. (1995) Scale and squamation character polarity and phyletic assessment in the family Cichlidae. *Journal of Fish Biology* **47**, 91-106.

Louiselle, P.V. (1985) *The Cichlid Aquarium.* Tetra-Press, Melle.

Lundberg, J.G. (1993) Africa-South American Freshwater Fish Clades and Continental Drift: Problems with a Paradigm In: Biological relationships between Africa and South America (ed P. Goldblatt), Yale University Press, New Haven. pp 157-199.

Malfait, B.T. and Dinkelman, M.G. (1972) Circum-Caribbean tectonic and igneous activity and the evolution of the Caribbean plate. *Geological Society of America, Bulletin* **83**, 251-272.

Martin, A.P., Bermingham, E. (1998) Systematics and evolution of lower Central American cichlids inferred from analysis of cytochrome b gene sequences. *Molecular Phylogenetics and Evolution* **9**,192-203.

Miller, R.R. (1966) Geographic distributions of Central American freshwater fishes. *Copeia* **1966,** 773-802.

Murray, A.M. (2000b) The Palaeozoic, Mesozoic and Early Cenozoic fishes of Africa. *Fish and Fisheries* **1,**111-145.

Murray, A.M. (2001) The oldest fossil cichlids (Teleostei: Perciformes): indication of a 45 million-year-old species flock. *Protocol of the Royal Society of London B.* **268**, 679 – 684.

Murray, A.M. (2001b) The fossil record and biogeography of the Cichlidae (Actinopterygii: Labroidei). *Biological Journal of the Linnean Society* **74**,517-532.

Myers, G.S. (1928) The existence of cichlid fishes in Santo Domingo. *Copeia*, **1928**, 33-36.

Myers, G.S. (1938) Fresh-water fishes and West Indian zoogeography. *Annual Report of the Smithsonian Institute.* **1937**, 339-364.

Myers, G.S. (1966) Derivation of the freshwater fish fauna of Central America. *Copeia* **1966,** 766-773.

Nelson, G. and Platnick, N.I. (1981) *Systematics and Biogeography: Cladistics and Vicariance*. Columbia University Press, New York.

Oliver, M.K. (1984) *Systematics of African cichlid fishes: determination of the most primitive taxon, and studies on the haplochromines of Lake Malawi.* Unpublished PhD dissertation, Yale University.

Palumbi, S.R. (1996) Nucleic acids II: the polymerase chain reaction. In: *Molecular Systematics* (D.M. Hillis, C. Moritz and B.K. Mable, eds.) 2nd ed., pp. 205-247. Sinaur, Sunderland.

Paulay, G. (1994) Biodiversity on oceanic islands - its origin and extinction. *American Zoologist* **34**, 134-144.

Perfit, M.R. and Williams, E.E. (1989) Geological constraints and biological retrodictions in the evolution of the Caribbean sea and its islands In: *Biogeography of the West Indies: Past, Present, and Future.* (ed C.A. Woods) Sandhill Crane Press, Gainesville, pp. 47-102.

Pindell, J.L., Cande S.C., Pitman III, W.C., Rowley, D.B, Dewey, J.F., LaBrecque, J., and Haxby, W. (1988) A plate-kinematic framework for models of Carribean evolution. *Tectonophysics* **155**, 121-138.

Pindell, J.L. and Barrett, S.F. (1990) Geological evolution of the Carribean: A plate tectonic perspective. In: *The Geology of North America. Vol.H., The Carribean Region.* (J.E. Case and G.Dengo eds.) A decade of North American Geology Series. Boulder, Colorado: Geological Society of America.

Pitman, W.C., III, Cande, S.C., LaBrecque, J., Pindell, J.L. 1993. Fragmentation of Gondwana: the separation of Africa from South America. In: Biological relationships between Africa and South America (ed P. Goldblatt), Yale University Press, New Haven. pp 15-34.

Rauchenberger, M. (1988) Historical biogeography of poeciliid fishes in the Caribbean. *Systematic Zoology* **37**, 356-365

Roe, K.R., Conkel, D. and Lydeard, C. (1997) Molecular systematics of Middle American cichlid fishes and the evolution of trophic-types in 'Cichlasoma (Amphilophus)' and 'C.(Thorichthys)'. *Molecular Phylogenetics and Evolution* **7**, 366-376.

Rivas, L.R. (1986) Comments on Briggs (1984): Freshwater Fishes and Biogeography of Central America and the Antilles. *Systematic Zoology* **35**, 633-639.

Rosen, D.E. (1975) A vicariance model of Caribbean biogeography. *Systematic Zoology* **24,** 431-464.

Rosen, D.E. (1985) Geological hierarchies and biogeographic congruence in the Caribbean. *Annals of the Missouri Botanical Garden* **72**, 636-659.

Sober, E. R. (1988) *Reconstructing the Past: Parsimony, Evolution and Inference*. Bradford, MIT Press, Cambridge.

Sparks, J.S. (2001) *Phylogeny and biogeography of the Malagasy and South Asian Cichlid Fishes (Teleostei: Perciformes:Cichlidae), including a survey of the freshwater fishes of Madagascar.* Unpublished doctoral dissertation, University of Michigan.

Stiassny, M.L.J. (1991) Phylogenetic interrelationship of the family Cichlidae: An overview. In: *Cichlid Fishes. Behavior, Ecology and Evolution.* (ed M.H.A. Keenlyside), Chapman and Hall, London. pp. 1-31.

Stiassny, M.L.J., Chakrabarty, P. and Louiselle, P.V. (2001) Relationship of the Madagascan cichlid genus *Paretroplus* with description of a new species from the Betsiboka River drainage of Northwest Madagascar. *Ichthyological Exploration of Freshwaters* **12**, 29-40.

Streelman, J.T. and Karl, S.A. (1997) Reconstructing labroid evolution with single-copy nuclear DNA. *Proceedings of the Royal Society of London B.* **264***, 1011-1020.

Streelman, J.T, Zardoya, R., Meyer, A. and Karl, S.A. (1998) Multilocus phylogeny of cichlid fishes (Pisces: Perciformes): Evolutionary comparison of microsatellite and single copy nuclear loci. *Molecular Biology and Evolution* **15**, 798-808.

Swafford, D.L. (1998) PAUP*4O.b3: Phylogenetic Analyses Using Parsimony, version 4. Sinaur Associates, Inc, Sunderland.

Tedford, R.H. (1974) Marsupials and the new paleogeography. In: *Paleogeographic Provinces and Provinciality* (ed C.A. Ross), *Society of Economic Paleontologists and Mineralogists, Special Publication* **21**,109-126.

Thompson, J.D., Gibson, T.J., Plewniak, F., Jeanmougin, F., and Higgins, D.G. (1997). The Clustal_X windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. *Nucleic Acids Research* 25: 4876-4882. Available from http://ncbi.nlm.nih.gov.

Vences, M., Freyhof, J., Sonnenberg, R., Kosuch, J. and Veith, M. (2001) Reconciling fossils and molecules: Cenozoic divergence of cichlid fishes and the biogeography of Madagascar. *Journal of Biogeography* **28**, 1091-1099.

Williams, E.E. (1989) Old problems and new opportunities in West Indian Biogeography, In: *Biogeography of the West Indies: Past, Present, and Future* (ed C.A.Woods), Sandhill Crane Press, Gainesville. pp. 1- 46.

Zardoya, R., Vollmer, D.M., Craddock, C., Streelman, J.T., Karl, S., and Meyer, A. (1996) Evolutionary conservation of microsatellite flanking region and their use in resolving the phylogeny of cichlid fishes (Pisces: Perciformes). *Proceedings: Biological Sciences* **263** ,1589-1598.