1982. In Ghillean T. Prance (ed.), Biological
Diversification in the Tropics.
Columbia University Press, New York. 714 pages.

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Distributions, Relationships, and Zoogeography of Lowland Frogs

The *Leptodactylus* Complex in South America, with Special Reference to Amazonia

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ABSTRACT An analysis of the distributions and evolutionary relationships of lowland frogs of the species-rich *Leptodactylus* complex is presented. Composite species distributions are derived and carefully examined for general patterns. Centers of species diversity are defined and compared with the present distribution of morphoclimatic domains. The high species diversity observed in the *Leptodactylus* complex is attributed to the occurrence of two major ecological groupings of frogs and three distinct adaptive patterns of these frogs.

Evolutionary relationships among representative *Leptodactylus* species based on comparative studies of albumin sequence differentiation are also described. This work reveals that speciation events are Tertiary, not Pleistocene events, and that there exists some intraspecific variation that appears to extend back into the Pliocene.

The refuge theory accounts for very few distributional events and no speciation events in the *Leptodactylus* complex.

The *Leptodactylus* complex of frogs has been the subject of extensive systematic analyses by the senior author for the past twelve years. More recently we have initiated biochemical analyses of these same species in hopes of providing new insights into the relationships among this interesting but enigmatic assemblage of frogs. This symposium has provided us the impetus to synthesize our current understanding of the distributions and evolutionary relationships of the members of the *Leptodactylus* complex. Because much systematic and biochemical work is still going on, what we present here must be construed as a progress report. However, we believe that at this time we are able to make some basic statements concerning the distribution, evolutionary relationships, and zoogeography of the group.

The focus of this symposium is the testing of what has come to be called the refuge theory (Simpson & Haffer 1978), based on studies of avifaunas primarily in

Our joint work has been supported by the Director's Office, National Museum of Natural History, Smithsonian Institution, and the Smithsonian Research Awards program.

Raymond F. Laurent and Roy W. McDiarmid provided muscle samples from Argentina and Peru, respectively.

Roy W. McDiarmid, Alan H. Savitzky, and P. E. Vanzolini have read over the manuscript and offered constructive comments.

WHR's work on systematics of the *Leptodactylus* complex has been supported by the Smithsonian Institution Research Foundation, the Smithsonian Research Award Program, and the Smithsonian Amazon Ecosystem Research program.

WHR expresses special thanks to P. E. Vanzolini for a personal introduction to the Amazon.

LMR's work on biochemical systematics of leptodactylid frogs has been generously supported by the Department of Genetics and Development, University of Illinois, Urbana.

We thank these institutions and individuals for their support.

Amazonia (Haffer 1969). Additional vertebrate evidence supporting this theory comes from studies of iguanid lizards (Vanzolini & Williams 1970). The group of frogs that we are studying possesses some unique features which help to provide a different perspective for an understanding of the zoogeography of the Amazon basin.

The leptodactylid frogs are quite different from those organisms used to develop the refuge theory. Previous zoogeographic analyses of Amazonian frogs that discussed the refuge theory used the theory as an uncritical explanation of the patterns identified in the studies (e.g., Duellman 1972; Heyer 1973). We attempt here to make some zoogeographic statements based on patterns of distributions and relationships. We then compare the results of our analysis with predictions drawn from the refuge theory. As will develop, the frogs we are studying differ from other groups of organisms in aspects of both zoogeography and speciation. Only through study of a variety of plant and animal groups will the total zoogeography of any region be understood. This is especially true for a region as old and complex as the Amazon basin.

The distributions and ecological affinities of the frogs we study suggest that they are ideal candidates for a zoogeographical analysis centering on the Amazon basin. Collectively, the species range throughout tropical and subtropical lowland South America. Some species occur only in the Amazon basin. Others occur only outside the basin. Still others occur broadly throughout the basin and adjacent areas. Some species occur only within the rainforests proper. Others occur only in open vegetation. Others occur in the forests, at forest edges, and in the open.

We do not believe that analysis of members of the *Leptodactylus* complex will allow a total understanding of the zoogeography of the Amazon basin. On the other hand, we think that any zoogeographic construct of the Amazon basin will be incomplete unless it includes the data derived from frogs, and that members of the *Leptodactylus* complex comprise an exemplary system for an analysis of frog zoogeography in Amazonia.

Methods and Materials

Two kinds of data are used in the analysis: distributional data and comparative molecular data.

The distributional data are based on the point locality maps published elsewhere (Heyer 1970, 1973, 1978, 1979). General distribution maps were produced by circumscribing the individual localities. The criterion used for determining where the species boundary lines should be drawn was that reasonable fidelity had to be maintained to the previously published point locality maps. Thus, someone who compares a range map produced for this study with a previously published point locality map can determine that the range map is indeed a general reflection of the point locality map and further, comparison will show where and how decisions were made on whether to consider any given point as a part of a large distribution pattern or as an isolated outlier population (for example, compare figure 20.6 in this paper with figure 27 in Heyer 1973). One other practical method was used in drawing boundary lines. Ab'Saber's (1977a) morphoclimatic domain map of South America was compared with the individual point location maps. If one of the morphoclimatic domain boundary lines (or any part thereof) would describe the boundary line for the point distributions as well as a smoothly curved boundary line, the former was used. Use of morphoclimatic domain distributions introduces other factors in the analysis. The disadvantage introduced is that of a certain amount of subjectivity.

This amount of subjectivity is not critical, as the morphoclimatic domain boundary lines were used only where there were point localities which supported using those boundaries. Where point distributions did not coincide with a morphoclimatic domain, a smooth curve was drawn around the point localities. The advantage introduced is generality. For those species represented in collections by many specimens from many localities, it is clear that Ab'Sáber's morphoclimatic domains correlate best with the actual data. We do not have enough data to perform intricate correlation analyses, but we are convinced by the correlations we see. The purpose of our analysis is to identify some general distribution patterns. To do this, we have gone from specific point distribution maps to general range maps, sacrificing precision, but gaining generality. The point distribution maps are not predictive in terms of what species can be found in an as yet unsampled locality—the general distribution maps are.

Until recently, the species comprising the genera *Adenomera* and *Vanzolinius* also were included in the genus *Leptodactylus*. As far as the distributions are known for these groups (Heyer 1970, 1973), they are included in this analysis.

Certain species are excluded from analysis. As the focus of this paper is on South America, particularly Amazonia, all species having primarily or entirely Middle American or West Indian distributions are omitted. These are *Leptodactylus albilabris*, *L. fallax*, *L. fragilis*, *L. melanonotus*, and *L. poecilochilus*.

Five species are known from too few localities to estimate a distribution range: *Adenomera lutzi*, *Leptodactylus dantasi*, *L. geminus*, *L. marambaiae*, *L. syphax*.

Two species that have been reviewed have since been found to be a composite of at least two species whose ranges are not individually known: *Adenomera bokermanni* and *Leptodactylus wagneri*.

Member of the *Leptodactylus ocellatus* group have not been reviewed recently at the group level so no detailed locality maps are available for the members of this group. Two members of this group occur in the Amazon basin. The names currently associated with this complex are: *Leptodactylus bolivianus*, *L. chaquenis*, *L. macrosternum*, *L. ocellatus*, and *L. viridis*.

With the above exceptions, the distributional analyses are based on the 30 remaining known members of the *Leptodactylus* complex: *Adenomera andreae*, *A. hylaedactyla*, *A. marmorata*, *A. martinezi*, *Leptodactylus amazonicus*, *L. bufonius*, *L. elenae*, *L. flavopictus**, *L. fuscus**, *L. gracilis**, *L. knudseni**, *L. labrosus*, *L. labyrinthicus**, *L. laticeps**, *L. latinasus**, *L. laurae*, *L. longirostris*, *L. mystaceus*, *L. mystacinus*, *L. notoaktites*, *L. pentadactylus**, *L. podocipinus*, *L. pustulatus*, *L. rhodomystax*, *L. rhodonotus*, *L. rugosus*, *L. stenodema**, *L. troglodytes*, *L. ventrimaculatus*, and *Vanzolinius discodactylus*.

The molecular data were obtained using the quantitative immunochemical technique of microcomplement fixation (MC′F) to compare sequence similarities of the serum albumin proteins of the frogs. Purified albumin from *Leptodactylus fuscus* (Brazil: São Paulo; Boracéia) and *L. pentadactylus* (Panama: Canal Zone; Frijoles) were used to prepare antiserum for comparisons with other species of *Leptodactylus*. Plasma and phenoxyethanol preserved muscle tissue from representatives of nine species of *Leptodactylus* were used as sources of albumin for comparisons with the two antisera. Those species in the preceeding list that are marked with asterisks were used in the MC′F studies: Voucher specimens of all species used in this study are now or will be deposited in the Smithsonian collections. The details of the MC′F procedure can be found in Champion et al. (1974), and Maxson, Highton, and Wake (1979). The MC′F derived data are reported in immunological distance units. For albumin one unit of immunological distance is roughly equivalent to one amino acid substitution (Maxson & Wilson 1974). The mean evolutionary rate of albumin approximates one immunological distance unit per 0.54 million years (Carlson, Wilson, & Maxson 1978) and this relationship was used in drawing the time scales in figures 20.11 and 20.12.

Certain terms as used in the text are defined as follows:

Domain. A morphoclimatic domain in the sense of Ab'Sáber (1977a). Of particular interest are the forested domains of Amazonia and the Atlantic forests of Brazil and open formation Chaco, Cerrado, and Caatinga domains.

Vegetation. This is the actual vegetation at a given site. Two major kinds of vegetation are discussed; forest vegetation and open formation vegetation. The forest vegetation has a closed canopy, the open formation vegetation an open canopy, including vegetations characteristic of the cerrados and caatingas as well as natural and man-made clearings.

Delimited taxa. These are taxa limited to certain vegetation types. Of importance are forest delimited taxa, which occur in and are limited to forest vegetations and open formation delimited taxa which occur in and are limited to open formation vegetations.

An example of how these terms are used is that the forested domain of Amazonia contains a broad expanse of forest vegetation and it also contains a network of open formation vegetation.

Species Distributions

We first examine composite distributions of species to look for general patterns and then examine some specific distributions to clarify the general patterns or to raise questions concerning these patterns. The patterns of overlapping species distributions are not viewed as analytic panaceas, but as an analytic approach that may give rise to interesting patterns and questions.

The composite map was made by tracing each species distribution on a single base map. Only contiguous ranges were used; individual outlying localities were omitted for the analyses leading to figures 20.1, 20.3, 20.4 and 20.5 only. The composite distribution map (fig. 20.1) is presented with a map (fig. 20.2) extracting some of the major morphoclimatic zones recognized by Ab'-Sáber (1977a), which was used as an aid to delimit individual species ranges. The patterns of lines resulting from the composite ranges is complex, but two general statements can be drawn from figure 20.1. First, the distributions of members of the *Leptodactylus* complex in the Amazon are rela-

Figure 20.1 Composite distributions of 30 species of the *Leptodactylus* complex. Heavy lines indicate where three or more species boundaries coincide.

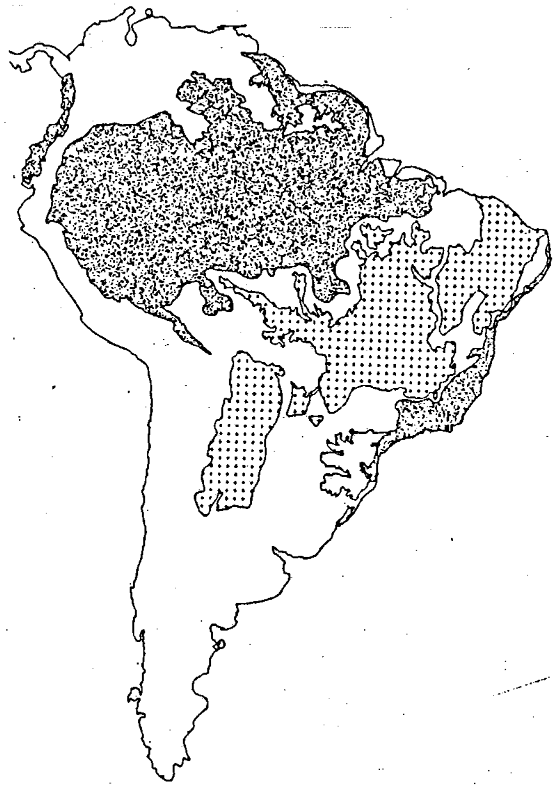

Figure 20.2 Selected morphoclimatic domains (Ab'-Sáber, 1977a) pertaining to distributions of species in the *Leptodactylus* complex. Heavy, uneven stippling indicates the forested domains of (left to right) the Pacific Equatorial Domain, the Amazon Equatorial Domain (Amazonia), and Atlantic Tropical Domain (Atlantic Forests). Crosses indicate the open vegetation formation domains of (lower left to upper right) Central Chaco Domain, Cerrados Domain, and Caatingas Domain. Normal stippling indicates Araucaria Domain.

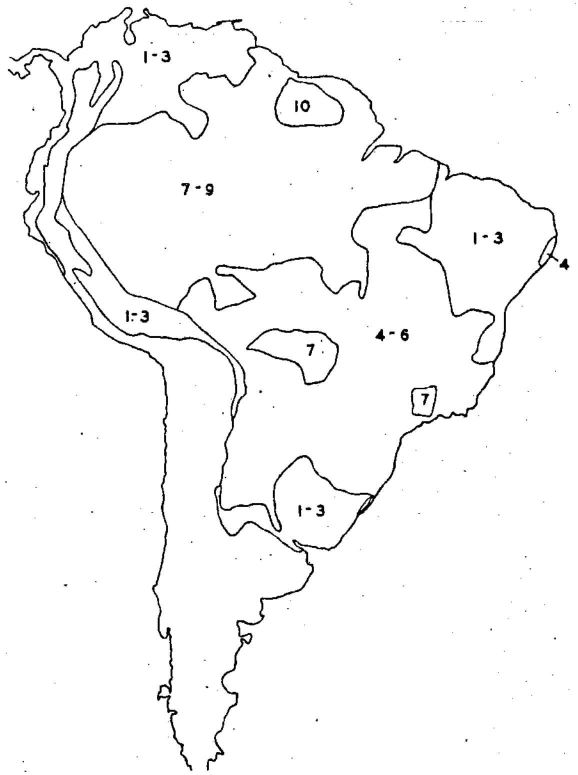

Figure 20.3 Species density map for 30 species of the *Leptodactylus* complex. Isophene contour lines of intervals of three species derived from figure 20.1. An example of how to read the figure is that Amazonia has 7–9 species; the Roraima area has 10 species.

tively homogeneous. Second, the domain where the greatest number of species ranges coincide is the Amazon Equatorial Domain, or Amazonia.

A map of isophenes of species densities is presented in figure 20.3. The pattern shows a diversity gradient, with the highest diversity generally occurring in the middle of the composite range and lower diversity around the periphery. If the Middle American species were included, the species diversity in northwestern South America would be higher and the number of species would decrease from Costa Rica to southernmost Texas. It is clear that Amazonia has a high species density and that the zoogeography of the *Leptodactylus* complex in Amazonia is a key step to understanding the zoogeography of the complex throughout its range. The four areas of highest diversity are rather different in terms of habitats and ranges. Those of Amazonia (broad region with 7–9 species in figure 20.3) and the Atlantic Forest of Brazil (area with 7 species in southeast Brasil in figure 20.3) represent major morphoclimatic domains and most of the species represented have their centers of distribution in those domains. In north-central South America 10 species ranges overlap in an area where two morphoclimatic domains interdigitate (see fig. 20.2); this area represents an overlapping of species ranges whose centers of distribution lie elsewhere. The area in south-central South America (area of 7 species where Brazil, Bolivia, and Argentina border each other) is a transition area between three major morphoclimatic domains. No species is restricted to that area; their centers of distribution lie elsewhere.

A basic tenet to understanding the zoogeography of the frogs of Amazonia is the recognition of forest and open formation delimited taxa (Heyer 1976, as tutored by Vanzolini, pers. comm.). We know of two species that occur both in open formation and forest vegetations: *Adenomera marmorata* and *Leptodactylus pentadactylus*. These, together with *L. knudseni*, *longirostris*, *mystaceus*, *rhodonotus*, and *ventrimaculatus*, for which we have no information, are omitted from this analysis. There remain 5 species which are forest delimited taxa (fig. 20.4) and 18 which are open formation delimited taxa (fig. 20.5).

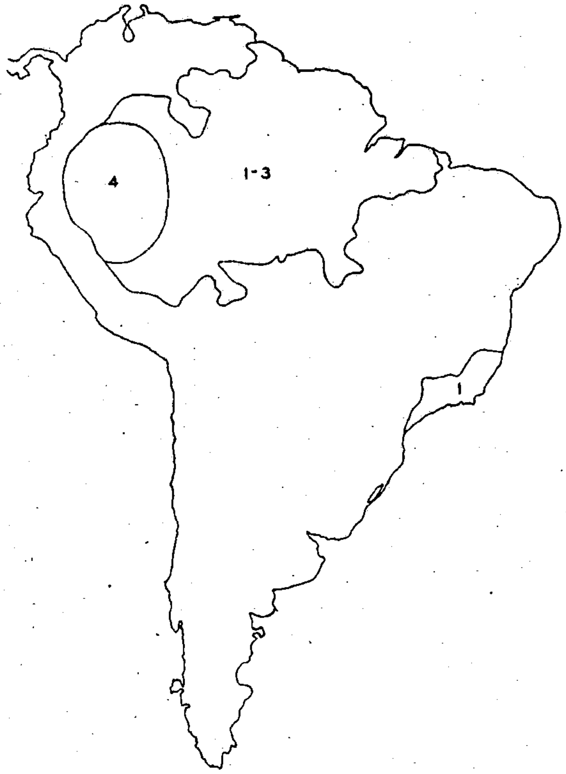

Figure 20.4 Species density map for 5 forest delimited species of the *Leptodactylus* complex.

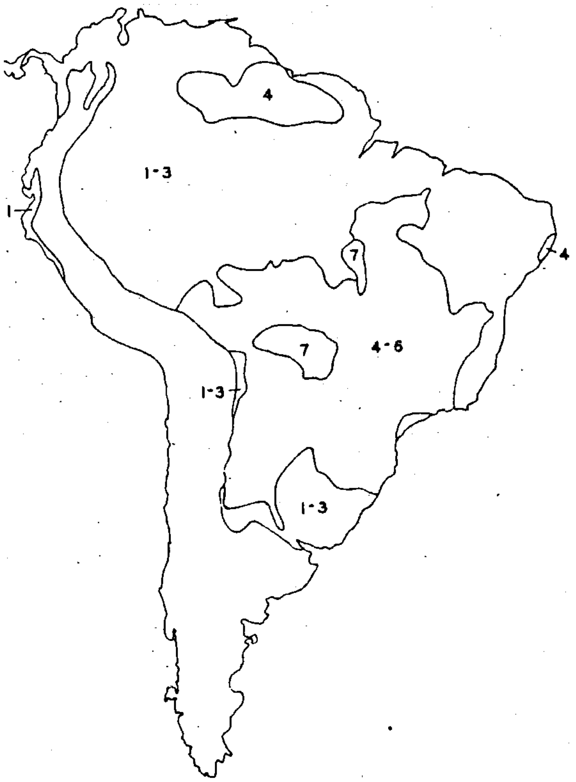

Figure 20.5 Species density map for 17 open formation delimited species of the *Leptodactylus* complex. Field observations for *Leptodactylus notoaktites* were made after the figure was prepared. Addition of this species does not appreciably change the pattern (distribution of *L. notoaktites* shown in fig. 20.8).

As expected, the forest delimited species occur in and are restricted to forest domains (fig. 20.4). The diversity pattern of forest delimited taxa (fig. 20.4) differs from the total pattern (fig. 20.3), in that the density pattern of the forest delimited species is a subset of the total pattern, but is not the same as any part of the total pattern. The pattern of open formation delimited species (fig. 20.5) is similar to the total pattern (fig. 20.3) in the open formation domains of South America. The differences lie in the forest domain regions. In contrast to the limitation of forest delimited frogs to forested domains, some open formation delimited species also occur in forest domains. The high species densities found in the forested domains of South America are

Figure 20.6 Distribution map of *Adenomera andreae*. Hatching indicates homogeneous, differentiated populations.

Figure 20.7 Distribution map of *Adenomera hylaedactyla*. Hatching indicates homogeneous, differentiated population.

due to the overlap of the two ecological groupings, forest and open formation delimited taxa.

In order to better understand the nature of the open formation delimited taxa occurring in forested domains, we need to examine some specific distribution patterns.

Of all the members of the *Leptodactylus* complex analyzed to date, intraspecific variation has been studied in detail only for members of the genus *Adenomera* (Heyer 1973). This is because several species of *Adenomera* have considerable color pattern polymorphism not matched in *Leptodactylus* or *Vanzolinius* species. Two examples show the kind of variation encountered, as well as the distributions of species delimited to forest and open formation vegetation.

Adenomera andreae occurs on the leaf litter of the forest floor. Individuals are active in the daytime and most specimens have been collected diurnally. The distribution coincides with the Amazonian forest domain (fig. 20.6). Two homogeneous and differentiated population systems were recognized previously (fig. 20.6 extrapolated from Heyer 1973). Notice that there are two isolated locality records outside the major area of distribution of this species. These records, as well as isolates of other species (certain of following maps) are somewhat troublesome. The distributions of the species involved would be easier to understand without them. Either they represent errors in locality data (which we have tried to confirm), misidentifications, or isolated, disjunct populations. The topic of disjunct isolates is discussed further in the zoogeography section. *Adenomera hylaedactyla* is a nocturnal species found in areas of open vegetations, such as river edges and agri-

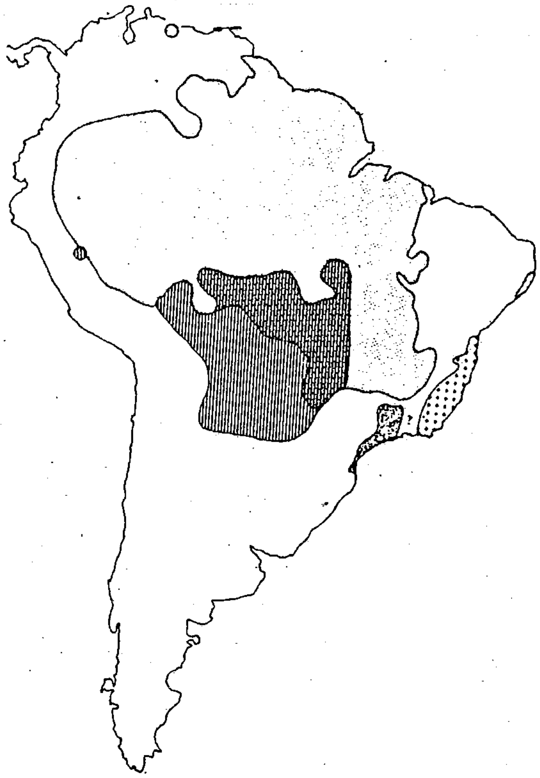

Figure 20.8 Distribution map of four species of *Leptodactylus*. Regular stippling indicates *L. amazonicus*, hatching indicates *L. elenae*, crosses indicate *L. mystaceus*, heavy, uneven stippling indicates *L. notoaktites*.

Figure 20.9 Distribution map of *Leptodactylus pentadactylus*.

cultural clearings. Although *A. hylaedactyla* is an open formation delimited species, it has a distribution generally associated with a forest domain (fig. 20.7) and has a pattern of intraspecific variation similar to that of *A. andreae*, the forest delimited species (fig. 20.7 extrapolated from Heyer, 1973). The ecological segregation of *A. andreae* and *hylaedactyla* is striking. The two species occur within a few steps of each other, but almost never occur together.

A cluster of four closely related species until recently were recognized as a single species, *L. mystaceus*. For the most part, these four species have allopatric distributions (fig. 20.8). One of the species (*L. elenae*) occurs in open formation domains; the other three (*amazonicus, mystaceus, notoaktites*) generally occur in forested domains (fig. 20.8). We have field experience with two of the last three taxa, *L. amazonicus* and *notoaktites;* both are open formation delimited species found along river and forest edges, and in natural and manmade clearings.

Leptodactylus pentadactylus occurs in forests, forest edges, and open formation vegetations; its distribution coincides with forested domains (fig. 20.9).

Leptodactylus fuscus is an open formation delimited species which has a very broad distribution, encompassing forest and open formation domains (fig. 20.10).

Analyses of the general and specific distribution patterns lead to the following conclusions. The high diversity of the *Leptodactylus* complex in forest domains is due to the occurrence of species with three kinds of adaptive patterns. The first pattern consists of species adapted to forests and which have undergone their evolution in the rainforest morphoclimatic domain with which

Figure 20.10 Distribution map of *Leptodactylus fuscus*. Numbers refer to immunological distance units (see text).

they are presently associated. The second pattern consists of species adapted to open formation vegetations, but which have undergone their evolution in forested morphoclimatic domain regions. Any rainforest region contains a network of open formations associated with river edges, natural clearings, or peculiar soil conditions. It is in this open formation network (which historically presumably was larger in extent in more arid times) that certain species have evolved. The distributions in figure 20.8, for example, suggest that the open areas within different forested morphoclimatic domains differ sufficiently such that adaptations to those differences have led to the evolution of distinct species within them. The third pattern consists of species adapted to open formation vegetations that underwent their evolution in open formation morphoclimatic domains and have subsequently invaded the open formation vegetation networks within the forest domains (e.g., *Leptodactylus fuscus*, fig. 20.10).

Relationships

The biochemical data on relationships that we present here represent the first results of albumin studies on members of the *Leptodactylus* complex. We are currently in the process of gathering and analyzing data on several more species to gain insights into the evolutionary history of the complex. The results we have, while preliminary to an understanding of the evolutionary history of the entire complex, do provide information relevant to an understanding of its zoogeography. The results presented here are consistent with previously hypothesized relationships based on morphological analyses. In a later paper we will deal with conflicting hypotheses of relationships based on morphological and biochemical data.

Two sets of relationships are presented here, one from the *Leptodactylus fuscus* group (fig. 20.11), the other from the *Leptodactylus pentadactylus* group (fig. 20.12). Of initial interest is the fact that two of the *L. fuscus* populations we sampled (São

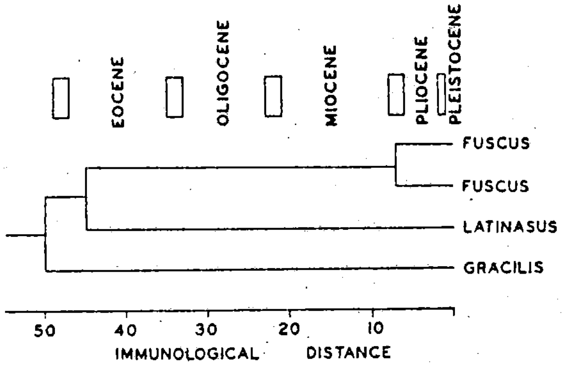

Figure 20.11 Proposed relationships for members of the *Leptodactylus fuscus* species group based on immunochemical data.

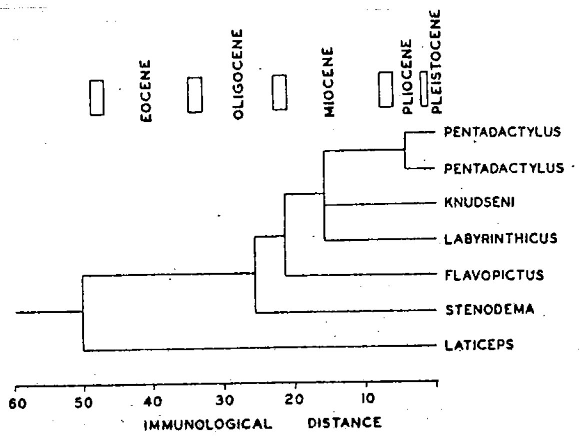

Figure 20.12 Proposed relationships for members of the *Leptodactylus pentadactylus* species group based on immunochemical data.

Paulo, Brazil and Tucumán, Argentina) are immunologically distinct—differing by 14 immunological distance units. Similarly, two populations of *pentadactylus* (from Panama and Amazonian Peru) differ immunologically (8–9 units). In most other MC'F studies of anurans we have found no distances greater than 0–2 units between members of the same species. A notable exception concerns eastern and western populatons of the North American treefrog *Hyla chrysoscelis*. These frogs differ by an average of 7 units and appear to have been isolated reproductively for several million years (Maxson, Pepper, & Maxson 1977). Additionally, several subspecies of European treefrog *Hyla arborea* differ by 8–11 units (Maxson 1978). However, many herpetologists propose elevating these subspecies to specific status. Thus the magnitude of albumin differentiation measured here is suggestive of long-term reproductive isolation.

One exciting feature of the MC'F technique of comparing albumins is that results can be interpreted along a time axis (see Wilson, Carlson, and White 1977 for a review). Challenges to assumptions behind the calibration of the albumin clock (Radinsky 1978) have recently been answered (Carlson, Wilson, & Maxson 1978). Albumin clock interpretations based on frog data correlate well with known geological events (Maxson & Wilson 1975). In this study we use the figure of 1 million years of time corresponding to an average immunological distance of 1.8 units (Wilson, Carlson, & White 1977). Even if the correlation of time and albumin evolution is not as precise as most evidence suggests it is, and recognizing there can be some variation in the rate of the albumin clock, two conclusions can be drawn from the results presented. First, some apparent intraspecific variation dates back beyond the Pleistocene into the Pliocene. Second, the speciation events so far analyzed (including data not reported here) are Tertiary events. The kinds of criticisms expressed about calibration of the albumin clock are concerned with relative precision; critics agree that there is a general correlation between immunological distance and time. The important conclusion strongly supported by our data is that speciation events in the *Leptodactylus* complex are in terms of millions, not thousands of years.

Zoogeography

While the available data will not permit a comprehensive understanding of the zoogeography of the *Leptodactylus* complex, certain conclusions can be drawn.

Leptodactylus fuscus is a widespread species characteristic of open formation vegetations. The MC'F data on albumins thus far gathered indicate that the species as currently understood is not panmictic. When comparing frogs from Manaus, Brazil to our antiserum made from frogs from São Paulo, Brazil we find no immunological differences. On the other hand, a sample of *fuscus* albumin from Tucumán, Argentina, differs by 14 units from the São Paulo *fuscus* albumin (fig. 20.10). Our interpretation of these data is that the species originated in open formation morphoclimatic domains and that in the Pliocene, a derivative stock became adapted to the network of open formation vegetations that exist in rainforest morphoclimatic domains. The derivative stock recently invaded and expanded within that network.

The presently understood range of *L. pentadactylus* is composed of two disjunct units (fig. 20.9). Albumin antiserum produced from the Middle American unit (Panama) differs by 9 immunological units from the albumin sample of Amazonian *pentadactylus* (Peru). Pliocene orogenic events likely separated these two population units of *pentadactylus*.

The distribution pattern of *Leptodactylus labyrinthicus* at first glance appears to have a pattern with a central core and several outlying isolated populations (fig. 20.13). The species occurs in open formation vegetations, never entering closed forest. Each of the known localities in Amazonia is in an open formation domain enclave. The pattern demonstrated by the coastal Venezuelan and Amazonian localities represents a relictual one, reflecting a broader distribution when open formation domains were more widespread in Amazonia. The coastal Venezuelan population is morphologically distinguishable from the Brazilian contiguous population (Heyer 1979). The level of morphological differentiation is slightly greater than that found in the disjunct populations of *pentadactylus*. We predict that the coastal Venezuelan population represents an early (probably Pliocene) geographic isolate of *labyrinthicus*. The Amazonian isolates likely reflect Pleistocene distributional events and thus are not well differentiated from the main *labyrinthicus* stock.

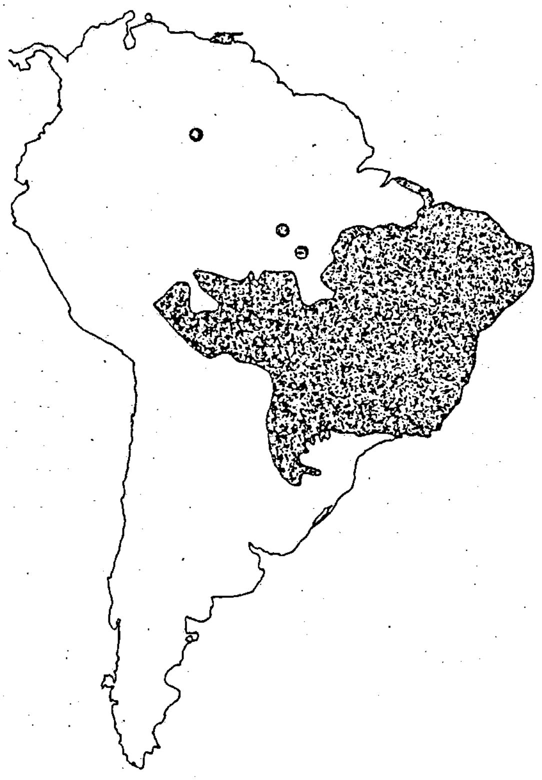

Figure 20.13 Distribution map of *Leptodactylus labyrinthicus.*

Most biochemical data we have analyzed are for members of the *Leptodactylus pentadactylus* group. These data suggest that certain speciation events within that group are almost as ancient as any speciation event yet dated in the entire *Leptodactylus* complex. Thus, events that occurred within the *pentadactylus* group span almost the entire history of the complex. For the scenario outlined below for the *pentadactylus* group, data and hypotheses are incorporated from previous analyses (Heyer 1975, 1979).

The ancestral *Leptodactylus pentadactylus* group stock was associated with the Neotropical Tertiary Geoflora by early Tertiary times. The foam nest (eggs laid in frothy foam) was an adaptation to the wet forest environments of the Neotropical Tertiary Geoflora. The foam nest served as a preadaptation for the successful penetration of open formation habitats by members of the *pentadactylus* group several times. The first recorded speciation event involved the isolation in and adaptation to the chaco environment of what is now *L. laticeps* (Fig. 20.12). This was perhaps an Eocene event. During the Oligocene an additional speciation event occurred involving what is now *L. stenodema*. Degree of morphological differentiation indicates Oligocene speciation events for *rhodomystax, rhodonotus, rugosus,* and *syphax.* The most recent speciation events have involved the evolution of a facultatively carnivorous tadpole. As the oldest taxon to have this adaptive type of larva is *L. flavopictus*, the ancestor to this species cluster may well have been a wet forest species, occurring in the Atlantic forests of Brazil, not very different, perhaps, from *flavopictus.* The essential allopatric distributions of 3 of the 4 species with the facultatively carnivorous tadpole in South America suggest the vicariant model of speciation. These speciation events apparently took place in the Miocene. Intraspecific differentiation continued through the Pliocene to the present.

The zoogeography of members of the *Leptodactylus pentadactylus* group is a good example of the palimpsest phenomenon. The same parchment has been written on and partially erased so many times that the biogeographic details will never be discernable. The present distribution patterns are a combination of recent associations with morphoclimatic domains, and the distribution of morphoclimatic domains and climatic events of at least the Pliocene and Pleistocene. The most recent record is the clearest, of course, and the best correlations of present distributions are with present morphoclimatic domains (compare figure 20.2 with individual species distributions herein). Some distributional details are accounted for by Pleistocene distribution patterns, such as the Amazonian distributions of *fuscus* and *labyrinthicus.* Other details of distribution patterns are accounted for by hypothesized Pliocene distributions, such as the coastal Venezuelan population of *labyrinthicus* and the Middle American population of *pentadactylus.*

The palimpsest phenomenon allows an explanation for the several disjunct, isolated population units found in several members of the *Leptodactylus* complex. If all distributions had to be accounted for by Pleistocene or Recent phenomena, many of the disjunct isolates could only be due to misidentifications or faulty interpretations of species limits. With the expanded time frame of speciation events occurring throughout the Cenozoic, the disjunct, isolated populations might be evidences of some of the previous writings that were not erased when the more recent events were recorded.

Speciation Models

Two basic modes of speciation have been proposed for Amazonian vertebrates (for a recent review, see Endler 1977).

The first is the allo-parapatric speciation mode. The essential features of this mode are: 1) an ancestral species spreads over a large area, 2) some groups of populations become separated, 3) differentiation proceeds in isolation, 4) secondary contact occurs after differentiation, 5) isolation mechanisms evolve, and 6) speciation is completed. This is the speciation model used by the proponents of the refuge theory. The refuge theory was proposed as an explanation of how populations could become separated in what today appears as the relatively uniform Amazon wet forest system. The explanation centers on the idea that in more arid times, the rain forest was distributed in small, disjunct patches—refuges. In such times populations of organisms associated with the forests would become spatially isolated and differentiation would occur. A body of corroborating evidence has effectively established not only the existence of these refuges, but their geographic limits during periods of maximum aridity (e.g., Ab'Sáber 1977b). The time period during which refuges are proposed to have existed is the Pleistocene. Isolation and expansion of these refuges was a cyclical phenomenon.

The second is the parapatric speciation mode. The essential features of this mode are: 1) an ancestral species spreads over a large area, 2) populations remain in contact, 3) differentiation proceeds in adjacent contacting areas, 4) shallow clines are formed, 5) steep clines are formed, 6) hybrid zones are formed, 7) isolation mechanisms evolve, and 8) speciation is completed.

The speciation events within the *Leptodactylus* complex appear to have occurred so long ago that it is fruitless to try to differentiate between the allo-parapatric and parapatric modes of speciation. Either or both could have been involved. Present distributional data regarding the *Leptodactylus* complex are best explained by the present distribution of morphoclimatic domains (Ab'Sáber 1977a). Only one of 30 species distributions suggests a strong correlation with Pleistocene forest refuges (*Vanzolinius discodactylus*). The intraspecific patterns of differentiation in *Adenomera andreae* (fig. 20.6) and *Adenomera hylaedactyla* (fig. 20.7) in a general fashion correlate with forest refuges, but not precisely enough to be convincing.

We are not arguing against the reality of forest refuges during more xeric times. The existence of these refuges was real. Ideally, the refuges should be defined on the basis of morphoclimatic techniques (Ab'Sáber 1977b) and not on species distributions. Perhaps the present task of biologists is to explain patterns of species distributions in light of Pleistocene refugial dynamics and not to propose refuges on the basis of organismal distribution patterns.

Several proponents of the refuge theory have argued for Pleistocene speciation correlating with the cyclical climatic events that caused expansion and contraction of the forest refuges (e.g. Haffer 1969; Vanzolini & Williams 1970). A priori estimates of speciation times can be misleading. Based on distribution patterns and ecological association of most taxa with open formations, the senior author previously considered the *Leptodactylus* complex to be a young complex, with Pleistocene speciation a distinct possibility. However, the biochemical data on the *Leptodactylus* complex demonstrate that at least for one vertebrate group, speciation events occurred well prior to the Pleistocene. Thus Pleistocene distributional events and speciation events are not one and the same for all organisms. We suggest that any further studies keep this separation in mind.

With respect to the *Leptodactylus* complex, the forest refuge theory does explain certain features, namely remnants of relatively recent distributional events. The data also indicate that the climatic cycles that caused expansions and contractions of species ranges during the Pleistocene are only distributional events for some groups of organisms. These Pleistocene cycles have not led to speciation in all groups of organisms in the Amazon basin.

Summary

The species rich *Leptodactylus* complex fauna of Amazonia is distributed rather homogeneously throughout the basin. The high diversity is attributed to the occurrence of two ecological groupings of frogs—forest delimited species and open formation delimited species. Three kinds of adaptive patterns are associated with these species. Pattern 1: The adaptive history of forest delimited frogs took place in the rain forest morphoclimatic domain with which they are presently associated. Pattern 2: The adaptive history of some open formation delimited species took place in the forested morphoclimatic domains within which they presently occur. Pattern 3: The adaptive history of other open formation delimited species took place in open formation morphoclimatic domains; these taxa subsequently invaded the open formation vegetation networks within the forested domains.

Results of microcomplement fixation studies of albumins indicate that: 1) some intraspecific variation dates back beyond the Pleistocene into the Pliocene, and 2) speciation events are Tertiary events.

Zoogeography of the *Leptodactylus* complex illustrates the palimpsest phenomenon. Present distributional patterns are a combination of recent associations with morphoclimatic domains, and the distribution of morphoclimatic domains and climatic events of at least the Pliocene and Pleistocene. The most recent record is the clearest; the best correlations of present distributions are with present morphoclimatic domains. Some distributional details are accounted for by Pleistocene distribution patterns, others by Pliocene distribution patterns.

The refuge theory accounts for very few distributional, and no speciation events in the *Leptodactylus* complex.

References

Ab'Sáber, A. N. 1977a. Os domínios morfoclimáticos na America do Sul. Primeira aproximação. *Geomorfologia* 53:1-23.

____ 1977b. Espacos ocupados pela expansão dos climas secos na América do Sul, por ocasião dos períodos glaciais quaternários. *Paleoclimas (São Paulo)* 3:1-19.

Carlson, S. S., A. C. Wilson, and R. D. Maxson. 1978. Do albumin clocks run on time? *Science* 200: 1183-85.

Champion, A. B., E. M. Prager, D. Wachter, and A. C. Wilson. 1974. Micro-complement fixation. In C. A. Wright, ed., *Biochemical and Immunological Taxonomy of Animals*, pp. 397-416. London: Academic.

Duellman, W. E. 1972. South American frogs of the *Hyla rostrata* group (Amphibia, Anura, Hylidae). *Zool. Mededelingen, Leiden* 47:177-92.

Endler, J. A. 1977. *Geographic Variation, Speciation, and Clines*. Monographs in Population Biology 10. Princeton, N.J.: Princeton Univ. Press.

Haffer, J. 1969. Speciation in Amazonian forest birds. *Science* 165:131-37.

Heyer, W. R. 1970. Studies on the frogs of the genus *Leptodactylus* (Amphibia: Leptodactylidae). 6: Biosystematics of the Melanonotus group. *Contributions in Science* 191:1-48.

____ 1973. Systematics of the Marmoratus group of the frog genus *Leptodactylus* (Amphibia, Leptodactylidae). *Contributions in Science* 251:1-50.

____ 1975. A preliminary analysis of the intergeneric relationships of the frog family Leptodactylidae. *Smithsonian Contrib. Zool.* 199:1-55.

____ 1976. Notes on the frog fauna of the Amazon basin. *Acta Amazônica* 6:369-78.

____ 1978. Systematics of the *fuscus* group of the frog genus *Leptodactylus* (Amphibia, Leptodactylidae). *Nat. His. Mus. Los Angeles County Sci. Bull.* 29:1-85.

____ 1979. Systematics of the *pentadactylus* species group of the frog genus *Leptodactylus* (Amphibia, Leptodactylidae). *Smithsonian Contrib. Zool.* 301: 1-43.

Maxson, L. R. 1978. Immunological evidence pertaining to relationships between Old World *Hyla arborea* (Amphibia, Anura, Hylidae) and North American *Hyla*. *J. Herpetology* 12:98-100.

Maxson, L. R., R. Highton, and D. B. Wake. 1979. Albumin evolution and its phylogenetic implications in the plethodontid salamander genera *Ensatina* and *Plethodon*. *Copeia*, pp. 502-8.

Maxson, L. R., E. Pepper, and R. D. Maxson. 1977. Immunological resolution of a diploid-tetraploid species complex of tree frogs. *Science* 197:1012-13.

Maxson, L. R. and A. C. Wilson. 1974. Convergent morphological evolution detected by studying proteins of tree frogs in the *Hyla eximia* species group. *Science* 185:66-68.

____ 1975. Albumin evolution and organismal evolution in tree frogs (Hylidae). *Syst. Zool.* 24:1-15.

Radinsky, L. 1978. Do albumin clocks run on time? *Science* 200:1182-83.

Sarich, V. M. and J. E. Cronin. 1976. Molecular systematics of the primates. In M. E. Goodman and R. E. Tashian, eds., *Molecular Anthropology*, pp. 141-70. New York: Planum Press.

Simpson, B. B. and J. Haffer. 1978. Speciation patterns in the Amazonian forest biota. *Ann. Rev. Ecol. Syst.* 9:497-518.

Vanzolini, P. E. and E. E. Williams. 1970. South American anoles: The geographic differentiation and evolution of the Anolis chrysolepsis species group (Sauria, Iguanidae). *Arq. Zool. S. Paulo* 19:1-298.

Wilson, A. C., S. S. Carlson, and T. J. White. 1977. Biochemical evolution. *Ann. Rev. Biochem.* 46: 573-639.