Molecular Phylogenetics and Evolution 61 (2011) 784–800

Contents lists available at SciVerse ScienceDirect

Molecular Phylogenetics and Evolution

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

# Phylogenetic relationships of the *Dactyloa* clade of *Anolis* lizards based on nuclear and mitochondrial DNA sequence data

María del Rosario Castañeda [a,b,*], Kevin de Queiroz [b]

[a] Department of Biological Sciences, The George Washington University, 2023 G Street NW, Washington, DC 20052, USA
[b] Department of Vertebrate Zoology, National Museum of Natural History, Smithsonian Institution, MRC 162, Washington, DC 20560, USA

ARTICLE INFO

*Article history:*
Received 27 January 2011
Revised 14 June 2011
Accepted 5 July 2011
Available online 21 July 2011

*Keywords:*
*Anolis*
*Dactyloa*
Phylogenetics
South America
Taxonomy

ABSTRACT

The *Dactyloa* clade, one of two major subgroups of mainland *Anolis* lizards, is distributed from Costa Rica to Peru, including the Amazon region and the southern Lesser Antilles. We estimated the phylogenetic relationships within *Dactyloa* based on mitochondrial (ND2, five transfer-RNAs, COI) and nuclear (RAG1) gene regions using likelihood and Bayesian methods under different partition strategies. In addition, we tested the monophyly of five previously recognized groups within *Dactyloa*. The data strongly support the monophyly of *Dactyloa* and five major clades: eastern, *latifrons*, *Phenacosaurus*, *roquet* and western, each of which exhibits a coherent geographic range. Relationships among the five major clades are less clear: support for basal nodes within *Dactyloa* is weak and some contradictory relationships are supported by different datasets and/or phylogenetic methods. Of the previously recognized subgroups within *Dactyloa*, only the *roquet* series consistently passed the topology tests applied. The monophyly of the *aequatorialis*, *latifrons* (as traditionally circumscribed) and *punctatus* series was strongly rejected, and the monophyly of *Phenacosaurus* (as traditionally circumscribed) yielded mixed results. The results of the phylogenetic analyses suggest the need for a revised taxonomy and have implications for the biogeography and tempo of the *Dactyloa* radiation.



## 1. Introduction

*Anolis* (Squamata: Iguanidae) is one of the most diverse groups of vertebrates traditionally ranked as genera, with 377 currently recognized species (Uetz and Etzold, 1996). Its members are distributed from southeastern North America to middle South America, including the West Indies (Etheridge, 1959; Peters and Donoso-Barros, 1970; Schwartz and Henderson, 1991). These lizards are characterized by the presence of adhesive toe pads and brightly colored dewlaps (Etheridge, 1959), and are typically of small size, arboreal habits and insectivorous diet, though there is significant interspecific variation in these traits (Schwartz and Henderson, 1991).

Early systematics studies (Etheridge, 1959; Williams, 1976a,b) divided *Anolis* into two sections (designated alpha and beta) based on the morphology of the caudal vertebrae. Each section was further subdivided into series and species groups based on several other osteological characters (e.g., post-xiphisternal rib formula, presence versus absence of a splenial, shape of the interclavicle). Subsequent phylogenetic studies have used a variety of data including morphology, allozymes, karyotypes, albumin immunology, and DNA sequences (e.g., Brandley and de Queiroz, 2004; Burnell and Hedges, 1990; Creer et al., 2001; Glor et al., 2003; Gorman and Atkins, 1967; Gorman and Kim, 1976; Gorman et al., 1968, 1980, 1983; Jackman et al., 2002; Poe, 1998; Schneider et al., 2001; Shochat and Dessauer, 1981). In these analyses, monophyly of the beta section and of several series and species groups has been supported, though others clearly are not monophyletic, and the phylogenetic relationships within and among some groups remain controversial (e.g., Creer et al., 2001; Giannasi et al., 2000; Glor et al., 2003; Jackman et al., 1999, 2002; Nicholson, 2002; Poe, 2004; Schneider et al., 2001).

Of the major groups within *Anolis*, the most poorly known regarding phylogenetic relationships is the clade designated as M1 (Mainland1) by Pinto et al. (2008) and recognized as the *latifrons* series by Etheridge (1959) and the genus *Dactyloa* by Guyer and Savage (1986; in the last two cases with the exclusion of *Phenacosaurus*; see below). However, the recognition of *Dactyloa* and other groups of anoles as genera (Guyer and Savage, 1986; Savage and Guyer, 1989) is controversial (Cannatella and de Queiroz, 1989; Williams, 1989). Following recent authors (e.g., Brandley and de Queiroz, 2004; de Queiroz and Reeder, 2008; Nicholson, 2002), who have applied the names of some of Guyer and Savage's genera to clades within *Anolis* regardless of rank and not necessarily identical in composition, we use the name *Dactyloa* for the clade

* Corresponding author. Address: P.O. Box 37012, MRC 162, Division of Amphibians and Reptiles, Smithsonian Institution, Washington, DC 20013-7012, USA. Fax: +1 202 633 0182.
*E-mail address:* castanedar@si.edu (María del Rosario Castañeda).

1055-7903/$ - see front matter © 2011 Elsevier Inc. All rights reserved.
doi:10.1016/j.ympev.2011.07.004

originating with the most recent common ancestor of the species included in the genus *Dactyloa* by Savage and Guyer (1989), which also includes the anoles formerly assigned to the genus *Phenacosaurus* according to the results of recent phylogenetic analyses (e.g. Jackman et al., 1999; Nicholson et al., 2005; Poe, 1998, 2004).

Only a handful of phylogenetic studies have included *Dactyloa* species (Jackman et al., 1999; Glor et al., 2001; Poe, 2004; Nicholson et al., 2005), and most of these have included relatively few, particularly mainland, species of *Dactyloa*. Moreover, most of what is known about the systematics of this group is based on morphological characters, which have been used to recognize six subgroups ranked as species groups by Williams (1976a) and as series by Savage and Guyer (1989): *aequatorialis*, *laevis*, *latifrons*, *punctatus*, *roquet*, and *tigrinus* (Williams, 1976b). The monophyly of these subgroups has never been tested, and there is no published hypothesis describing the relationships among them.

In this study, we present new molecular data for 40 of the 82 currently recognized species of *Dactyloa*, two potentially new *Dactyloa* species and 12 outgroup species (non-*Dactyloa Anolis* and non-*Anolis* Polychrotinae) to resolve the phylogenetic relationships within *Dactyloa*. In addition, we test hypotheses of monophyly of *Dactyloa* including *Phenacosaurus* species, *Dactyloa* excluding *Phenacosaurus* species and five of seven previously recognized species groups/series within *Dactyloa*.

## 2. Materials and methods

### 2.1. Taxa and character sampling

We collected new DNA sequence data from 40 species of *Dactyloa* (62 specimens), including representatives of the previously described *aequatorialis*, *latifrons*, *punctatus*, and *roquet* series, as well as *Phenacosaurus*, and two specimens suspected (based on morphological data and geographic distribution) to be new species. Only one representative of the *tigrinus* series was included, but we did not have any representatives of the *laevis* series. In addition, we included 12 species as outgroups: three non-*Anolis* species of Polychrotinae (*Polychrus marmoratus*, *Pristidactylus scapulatus*, *Urostrophus gallardoi*), and nine species representing nine series of non-*Dactyloa Anolis* (*Anolis bimaculatus*, *Anolis cupreus*, *Anolis cuvieri*, *Anolis equestris*, *Anolis lucius*, *Anolis marcanoi*, *Anolis occultus*, *Anolis sagrei*, *Anolis smaragdinus*). Sequenced fragments include two mitochondrial regions: a fragment including the entire NADH dehydrogenase subunit II (ND2), five transfer-RNA ($tRNA^{Trp}$, $tRNA^{Ala}$, $tRNA^{Asn}$, $tRNA^{Cys}$, $tRNA^{Tyr}$), and the origin for light-strand replication ($O_L$) (Macey et al., 1997; ~1500 b), and a fragment of the cytochrome oxidase subunit I (COI, ~650 b), as well as one nuclear region: the recombination activating gene (RAG-1, ~2800 b). This selection of genes contains both highly conserved areas that are informative for deeper divergences and rapidly evolving regions that are informative for more recent divergences (Groth and Barrowclough, 1999; Jackman et al., 1999; Miyata et al., 1982). Previously collected sequences for the ND2 region were obtained from GenBank for all three non-*Anolis* Polychrotinae, seven non-*Dactyloa Anolis* and ten *Dactyloa* species. A complete list of samples, with voucher/catalogue and GenBank numbers and collection localities is given in Appendix A.

### 2.2. Laboratory protocols

Genomic DNA was extracted from liver or muscle tissue using DNeasy Tissue Extraction Kits (QIAGEN Inc.). Polymerase chain reaction (PCR) was used for amplification of the particular genomic regions and performed in a DNA Engine (PTC-200) Peltier Thermal Cycler (MJ Research) and a DNA Engine Dyad® Peltier Thermal Cycler (Bio-Rad Laboratories). Two alternative PCR cycling protocols were used, depending on the combination of primers and their respective optimal annealing temperatures. When both primers had similar optimal annealing temperatures (less than 2° difference), the protocol used was: pre-denaturation at 94 °C for 120 s, followed by 30–35 cycles of denaturation at 95 °C for 30 s, annealing at the average optimal temperature between the two primers for 30 s, and primer extension at 72 °C for 60–120 s. When primers used had a large difference in optimal annealing temperatures (more than 2° difference), the protocol used was: pre-denaturation at 94 °C for 120 s, followed by 5 cycles of denaturation at 95 °C for 30 s, annealing at the higher optimal temperature between the two primers for 30 s, and primer extension at 72 °C for 60–120 s. Then, two sets of 5 cycles each were run with identical denaturation and primer extension conditions as before, but with decreased annealing temperatures (each set decreased by 2–3°). A final set with the annealing temperature set at the lowest optimal annealing temperature between the two primers was run for 20 cycles. Primer extension time was adjusted according to the length of the fragment being amplified (~1 min per 1000 b). PCR products were purified using ExoSAP-IT (USB Corporation) or magnetic beads (AMPure, Agencourt Bioscience Corporation). Cycle sequencing reactions were performed using BigDye Terminator chemistry (Applied Biosystems) directly on purified PCR products. The sequencing protocol used was denaturation at 96 °C for 10 s, annealing at 50 °C for 10 s, and primer extension 60 °C for 240 s for 35 cycles. Sequenced products were purified using Sephadex G-50 columns (SIGMA), and run on an automated sequencer (ABI Prism 3100 and 3730xl Genetic Analyzer, Applied Biosystems). The complete list of primers used in amplification and sequencing reactions is given in Appendix B. Assembly of sequences was performed with SeqMan II (DNASTAR, Inc.).

### 2.3. Alignment procedures and data matrices

Protein-coding regions were aligned using Clustal X (Thompson et al., 1997), under default gap costs, and subsequently translated into amino acids using MacClade v4.07 (Maddison and Maddison, 2001) to verify the correct translation frame. Genes coding for tRNAs were aligned manually to incorporate secondary structure information, following Kumazawa and Nishida's (1993) structural model for mitochondrial transfer RNAs. Sequences were strictly aligned following this model (i.e., no gaps were introduced in areas with conserved lengths: AA, AC, TΨC and D-stems, junctions between AA- and D-stems, D- and AC-stems, and AC- and TΨC-stems, and the anticodon loop). From the set of tRNA regions that according to this model can exhibit length variation (and potentially result in ambiguous alignments), those that showed length variation were excluded from the analyses.

Three different data matrices were analyzed, one including the nuclear gene region (RAG1), one including both mitochondrial regions (ND2–COI), and one combining all three gene regions (ND2–COI–RAG1).

### 2.4. Phylogenetic analyses and data partitions

Phylogenetic relationships were estimated using likelihood and Bayesian inference methods. Likelihood analyses were performed with GARLI-PART (Zwickl, 2006) v0.97 and GARLI (Zwickl, 2006) v1.0 using multiple partitioning strategies for each matrix (Table 1). Each analysis was run with 20 replicates using random starting trees (other settings were left as defaults). The models of evolution for the different partitions were selected based on the Akaike Information Criterion (AIC) as implemented in Modeltest (Posada and Crandall, 1998) v3.7. The trees inferred using GARLI v1.0 with an unpartitioned strategy were compared with those inferred using

**Table 1**
Partitioning strategies used in the phylogenetic analyses of the three datasets.

| Dataset | Number of partitions | Partitions |
|---|---|---|
| RAG1 | 1 | Unpartitioned |
| | 3 | 1st, 2nd and 3rd codon positions |
| ND2–COI | 1 | Unpartitioned |
| | 4 | 1st, 2nd and 3rd codon positions of all protein-coding genes (not partitioned by gene); tRNAs |
| | 7 | 1st, 2nd and 3rd codon positions of each protein-coding gene (parititioned by gene); tRNAs |
| ND2–COI–RAG1 | 1 | Unpartitioned |
| | 4 | 1st, 2nd and 3rd codon positions of all protein-coding genes (not partitioned by gene); tRNAs |
| | 7 | 1st, 2nd and 3rd codon positions of protein-coding mitochondrial genes; tRNAs; 1st, 2nd and 3rd codon positions of nuclear gene |
| | 10 | 1st, 2nd and 3rd codon positions of each protein-coding gene (partitioned by gene); tRNAs |

PAUP* v4.0 (Swofford, 2002) with a successive approximations approach (Sullivan et al., 2005; Swofford et al., 1996). To assess nodal support, non-parametric bootstrap values (BS; Felsenstein, 1985) were calculated in GARLI-PART v0.97 and GARLI v1.0, with 500 pseudoreplicates, using random starting trees and the same model of evolution (estimating parameter values for each pseudoreplicate) used in the tree searches. All other settings were left as defaults, except the number of multiple search replicates (searchreps), which was set to 1.

Bayesian analyses were performed with MrBayes v3.1.2 (Ronquist and Huelsenbeck, 2003) using the same partition strategies as in the likelihood analyses (Table 1). The RAG1 and ND2–COI matrices were analyzed using four independent runs of 10 million generations with a random starting tree, each with four Markov chains and default heating settings. The ND2–COI–RAG1 dataset under all partitions was analyzed using four independent runs, each with four Markov chains, with the temperature set at 0.15 to improve parameter mixing. The analyses of the unpartitioned and four partition strategies were run for 15 million generations, and the analyses of the 7 and 10 partition strategies were run for 25 million generations to ensure that convergence was achieved (see below). Trees were sampled with a frequency of one every 1000 generations. The first 25% of the sampled trees were discarded as the 'burn-in' phase. To confirm that stationarity was achieved within the first 25% of sampled trees, we examined plots of the $-\ln L$ versus generation number using TRACER v1.5 (Rambaut and Drummond, 2007) and compared the average standard deviation of split frequencies between chains and the potential scale reduction factor (PSRF) of all the estimated parameters for the four runs combined. The average standard deviation of split frequencies of two converging chains is expected to approach zero and the PSRF should approach 1 as the runs converge. In addition, an effective sample size (ESS) higher than 200—as calculated in TRACER—for all parameters in each analysis, was considered as an indication that an adequate number of independent samples was obtained in the post burn-in sample. Bayesian posterior clade probabilities (PP) were calculated based on the post burn-in trees for all four independent runs combined. Nodes with posterior probabilities higher than 95% were considered strongly supported, with the caution that PP might overestimate clade support, especially for short internodes (Alfaro et al., 2003; Lewis et al., 2005; Suzuki et al., 2002).

Akaike information criterion scores incorporating the correction for small sample size (AICc) were used to compare the results of alternative data partition strategies used in the likelihood analyses. AICc scores were calculated following the formula $\mathrm{AICc} = -2\ln L + 2K + 2K\,(K+1)/(n-K-1)$, where $K$ is the number of parameters in the model and $n$ is the number of bases in the dataset (Burnham and Anderson, 2004). A value of ΔAICc higher than 10 was considered an indication of strong support for the partition strategy with the smaller AICc. Bayes factors (BF) were used to evaluate the results from different partitioning strategies in the Bayesian analyses (Kass and Raftery, 1995; Pagel and Meade, 2005). Comparing model $a$ to model $b$, the Bayes factor is the ratio of the marginal likelihood of model $a$ to that of model $b$ (Pagel and Meade, 2005). Marginal likelihoods can be estimated by calculating the harmonic mean of the likelihoods of a sample from the posterior distribution, as obtained from a MCMC method (Newton and Raftery, 1994; Pagel and Meade, 2005). 2Ln-Bayes factors were calculated in TRACER v.1.5 (Rambaut and Drummond, 2007; Suchard et al., 2001) using harmonic means of the likelihoods of the trees in the post burn-in sample. Following the criteria outlined by Kass and Raftery (1995), BF values higher than 10 were considered an indication of strong support for partition strategy $a$ over partition strategy $b$.

### 2.5. Hypothesis testing

Wilcoxon signed-ranks tests (WSR; modified from Templeton, 1983, as described below), Approximately Unbiased tests (AU; Shimodaira, 2002) and a Bayesian approach based on the presence

**Table 2**
Comparison of alternative partitioning strategies used in the likelihood and Bayesian analyses. The number of partitions ($P$; Table 1), number of model parameters ($K$), likelihood values ($-\ln L$) and Akaike information criterion (AICc) scores of the maximum likelihood tree, harmonic means of likelihood scores (as calculated in TRACER from the post-burn set of Bayesian trees) and Bayes Factors are given. A value of ΔAICc > 10 was considered as an indication of strong support for the partition with smaller AICc (Burnham and Anderson, 2004). A value of 2Ln ($B_{ab}$) > 10 indicates very strong support for partitioning strategy $a$ (more partitions) over partitioning strategy $b$ (fewer partitions; Kass and Raftery, 1995).

| Dataset | $P$ | $K$ | $-\ln L$ | AICc | ΔAICc (larger AICc – smaller AICc) | Harmonic mean | 2Ln Bayes factors ($a$/$b$) |
|---|---|---|---|---|---|---|---|
| RAG1 | 3 | 21 | −13363.0 | 26768.2 | 455.9 (1–3) | −13625.7 | 184.8 (3/1) |
| | 1 | 10 | −13602.0 | 27224.1 | | −13718.1 | |
| ND2–COI | 7 | 60 | −45331.1 | 90783.7 | 833.5 (4–7), 2683.7 (1–7) | −45421.4 | 851.2 (7/4), 2719.7 (7/1) |
| | 4 | 38 | −45770.3 | 91617.2 | 1850.2 (1–4) | −45847.0 | 1868.5 (4/1) |
| | 1 | 10 | −46723.7 | 93467.4 | | −46781.3 | |
| ND2–COI–RAG1 | 10 | 81 | −59018.6 | 118202.0 | 859.7 (7–10), 4168.4 (4–10) 6232.0 (1–10) | −59185.4 | 847.7 (10/7), 4083.9 (10/4), 6164.9 (10/1) |
| | 7 | 59 | −59471.1 | 119061.8 | 3308.9 (4–7), 5372.3 (1–7) | −59609.2 | 3236.2 (7/4), 5317.2 (7/1) |
| | 4 | 38 | −61147.0 | 122370.6 | 2063.4 (1–4) | −61227.3 | 2081.0 (4/1) |
| | 1 | 10 | −62207.0 | 124434.0 | | −62267.8 | |

or absence of topologies containing particular hypotheses of relationships in the 95% credible set of trees (Huelsenbeck et al., 2001; Larget and Simon, 1999), were used to test the following hypotheses: (1) monophyly of *Dactyloa* excluding *Phenacosaurus*, (2) monophyly of the subgroups *aequatorialis*, *latifrons*, *Phenacosaurus*, and *punctatus* and (3) non-monophyly of *Dactyloa* (including *Phenacosaurus*) and of the *roquet* series. Hypotheses of non-monophyly were tested when the group in question was monophyletic in the optimal (unconstrained) trees. Given that we obtained data for only one species of the *tigrinus* series and none of the *laevis* series, no tests were performed regarding the monophyly of these taxa.

The tree resulting from the likelihood analysis of the ND2–COI–RAG1 matrix was compared with trees resulting from likelihood analyses of the same data set incorporating each alternative hypothesis as a constraint. Likelihood analyses were performed using the preferred partitioning strategy (selected based on AICc scores) in GARLI-PART v0.97 as described above. Alternative topologies were constructed using MacClade v4.07 (Maddison and Maddison, 2001) and incorporated into GARLI-PART v.097 as topological constraints. The undescribed species (*A.* sp1 and *A.* sp2) were excluded from these analyses, as their affinities to the different series are unclear. The WSR tests were performed in R (R Development Core Team, 2010) using site-likelihood scores (instead of number of parsimony steps, as originally proposed by Templeton (1983)). Site-likelihoods were calculated in GARLI-PART v0.97 using the same model and partition strategy as in the unconstrained analysis. The AU test was performed in CONSEL (Shimodaira and Hasegawa, 2001) v0.1i, with default settings. For the Bayesian approach, the results from the analyses of the ND2–COI–RAG1 dataset under the preferred partitioned strategy were used. Trees contained in the 95% credible set of trees from the post burn-in sample (from which taxa suspected to be new species were pruned) were loaded into PAUP* and filtered based on constraints corresponding to specific hypotheses. Topologies that were not present in the 95% credible set of trees (i.e., constraints for which no trees passed through the filter) were considered rejected by the data.

## 3. Results

A total of 4720 bases (b) were unambiguously aligned in the most inclusive matrix (ND2–COI–RAG1), consisting of 1312 b in the ND2 region, 654 b in the COI region and 2754 b in the RAG1 region. Two gaps were introduced in the ND2 alignment: (1) in positions 1033–1035 (in *P. scapulatus* and *U. gallardoi*) and (2) in position 958–960 (in *P. marmoratus*). No length variation was found in the COI fragment and therefore no gaps were introduced in the alignment. Six gaps were introduced in the alignment of the RAG1 gene region: (1) in positions 37–51 (in *A. equestris*), (2) in positions 76–87 (in both samples of *A. fitchi*), (3) in positions 121–123 (in *A. smaragdinus*), (4) in positions 217–219 (in all samples except both of *A. aequatorialis*), (5) in positions 280–294 (in all samples of *A. chloris*, *A. festae* and *A. peraccae*), and (6) in positions 634–636 (in all samples except *P. marmoratus*). Due to ambiguous alignment, 148 positions were excluded from the tRNA section of the ND2 region (see Section 2.3). No characters were excluded from the COI or RAG1 regions.

Akaike information criterion (AICc) scores and Bayes factors (BF) favored the most partitioned strategy for all three datasets (Table 2). The trees inferred from the 20 replicates performed in GARLI for each likelihood analysis (under the preferred partition strategies) had identical topologies for the RAG1 dataset, and non-conflicting topologies for the ND2–COI and ND2–COI–RAG1 datasets. Likelihood analyses in GARLI v1.0 with an unpartitioned strategy resulted in trees identical to those inferred with PAUP* using the successive approximations approach. In the Bayesian analyses of the ND2–COI–RAG1 dataset under 7 and 10 partitions, the four independent runs did not converge onto the same likelihood values, but instead reached stationarity in two different likelihood value regions (i.e., two runs reached stationarity at similar lower −Ln likelihood scores and the remaining two reached stationarity at similar higher scores). For this reason, two of the four runs were discarded and only the two with lower −Ln likelihood scores were used for tree and harmonic mean estimation. Relationships among species in the resulting topologies (from the runs with high versus low likelihood scores) differed only in one (7 partitions) or two (10 partitions) poorly supported nodes. Nodal

**Table 3**
Nodal support values (bootstrap proportions and posterior probabilities) for *Dactyloa* and the five major clades inferred in the likelihood (L) and Bayesian (B) analyses, by gene region and partition strategy (*P*; see Table 1). The strategy with seven partitions applies only to the combined data sets, and that with 10 partitions applies only to the ND2–COI–RAG1 combined data set. NA = not applicable.

| Clade | | *P* | Dataset | | |
|---|---|---|---|---|---|
| | | | RAG1 | ND2–COI | ND2–COI–RAG1 |
| *Dactyloa* | L | 1 | 100 | 67 | 100 |
| | | 3/4[a] | 100 | 61 | 100 |
| | | 7 | NA | 61 | 100 |
| | | 10 | NA | NA | 100 |
| | B | 1 | 1.00 | 0.95 | 1.00 |
| | | 3/4[a] | 1.00 | 0.99 | 1.00 |
| | | 7 | NA | 1.00 | 1.00 |
| | | 10 | NA | NA | 1.00 |
| *latifrons* | L | 1 | 100 | 100 | 100 |
| | | 3/4[a] | 100 | 98 | 100 |
| | | 7 | NA | 99 | 100 |
| | | 10 | NA | NA | 100 |
| | B | 1 | 1.00 | 1.00 | 1.00 |
| | | 3/4[a] | 1.00 | 1.00 | 1.00 |
| | | 7 | NA | 1.00 | 1.00 |
| | | 10 | NA | NA | 1.00 |
| *roquet* | L | 1 | 100 | 99 | 100 |
| | | 3/4[a] | 100 | 100 | 100 |
| | | 7 | NA | 100 | 100 |
| | | 10 | NA | NA | 100 |
| | B | 1 | 1.00 | 1.00 | 1.00 |
| | | 3/4[a] | 1.00 | 1.00 | 1.00 |
| | | 7 | NA | 1.00 | 1.00 |
| | | 10 | NA | NA | 1.00 |
| Eastern | L | 1 | 83 | 98 | 100 |
| | | 3/4[a] | 85 | 98 | 100 |
| | | 7 | NA | 98 | 100 |
| | | 10 | NA | NA | 99 |
| | B | 1 | 1.00 | 1.00 | 1.00 |
| | | 3/4[a] | 1.00 | 1.00 | 1.00 |
| | | 7 | NA | 1.00 | 1.00 |
| | | 10 | NA | NA | 1.00 |
| Western | L | 1 | 97 | 81 | 99 |
| | | 3/4[a] | 95 | 76 | 99 |
| | | 7 | NA | 81 | 99 |
| | | 10 | NA | NA | 100 |
| | B | 1 | 1.00 | 1.00 | 1.00 |
| | | 3/4[a] | 1.00 | 1.00 | 1.00 |
| | | 7 | NA | 1.00 | 1.00 |
| | | 10 | NA | NA | 1.00 |
| *Phenacosaurus* | L | 1 | 100 | 100 | 100 |
| | | 3/4[a] | 100 | 100 | 100 |
| | | 7 | NA | 100 | 100 |
| | | 10 | NA | NA | 100 |
| | B | 1 | 1.00 | 1.00 | 1.00 |
| | | 3/4[a] | 1.00 | 1.00 | 1.00 |
| | | 7 | NA | 1.00 | 1.00 |
| | | 10 | NA | NA | 1.00 |

[a] For ND2–COI and ND2–COI–RAG1 datasets there are four data partitions in the ND2 gene representing codon positions (1st, 2nd, 3rd) and tRNAs; for the two protein coding regions COI and RAG1 there are three data partitions (no tRNAs).

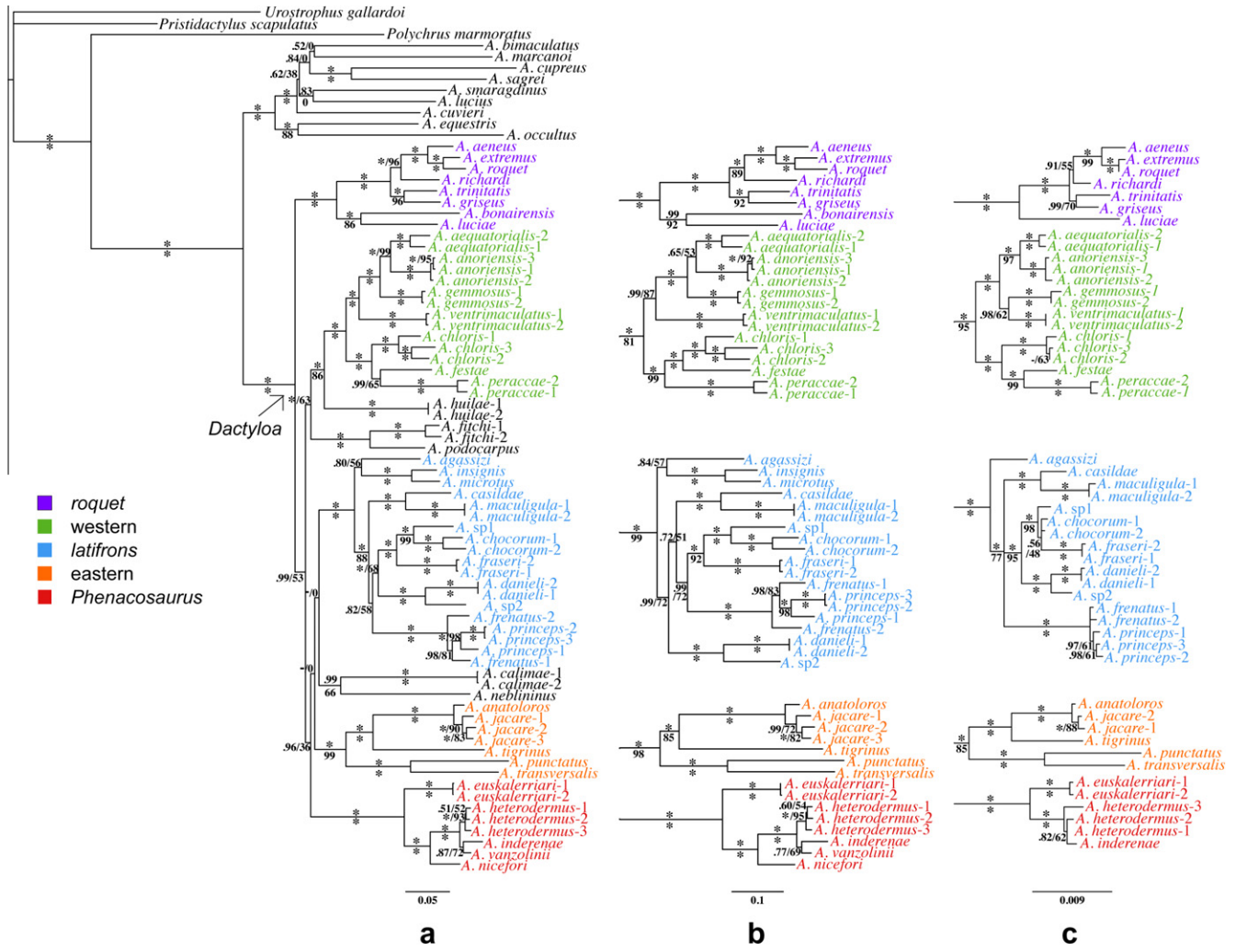


**Fig. 1.** (a) Maximum likelihood tree resulting from the analysis of the ND2–COI–RAG1 dataset partitioned by gene region, codon position and tRNAs (10 partitions). Bayesian posterior probabilities are shown above branches and bootstrap support values are shown below branches. Asterisks (*) indicate PP = 1.0 or BS = 100%; branches not inferred in the Bayesian analysis (of which there are three) are indicated with a dash (–). Major *Dactyloa* subclades (see text for details) are differentiated by color; the *Dactyloa* clade is indicated with an arrow pointing to the corresponding node. Maximum likelihood topologies for each major clade, inferred with the (b) ND2–COI dataset partitioned by gene region, codon position and tRNAs (seven partitions) and the (c) RAG1 dataset partitioned by codon position (three partitions) are also shown.

support and estimated parameter values were also very similar, except for the rate variation among sites (alpha), the proportion of invariant sites (pinvar) and the rate multiplier (m) for several partitions.

### 3.1. *Monophyly of* Dactyloa

Monophyly of the *Dactyloa* clade (including *Phenacosaurus*) was moderately to strongly supported in the likelihood and Bayesian analyses of all datasets under all data partitions (BS = 61–100%, PP = 0.95–1.00; Table 3; Figs. 1 and 2). Differences among datasets and phylogenetic methods are found in the most basal nodes within *Dactyloa* (Fig. 2); otherwise, the topologies are fairly consistent, including the inference of the same five major clades in all analyses (see following section). The inference of each major clade in the different analyses, with respective nodal support, is given in Table 3.

### 3.2. *Major* Dactyloa *subclades*

One major subclade is composed of twelve species: *Anolis agassizi*, *Anolis casildae*, *Anolis chocorum*, *Anolis danieli*, *Anolis fraseri*, *Anolis frenatus*, *Anolis insignis*, *Anolis maculigula*, *Anolis microtus*, *Anolis princeps*, *Anolis* sp1, and *Anolis* sp2. Most species in this clade were previously included in the *latifrons* species group (Williams, 1976b), or *latifrons* series (Savage and Guyer, 1989),[1] so hereafter this clade will be referred to as the *latifrons* clade (see also Section 4.2.1). This clade is strongly supported in all analyses (BS ⩾ 98%, PP = 1.00), and relationships within it are fairly consistent among gene regions, including the paraphyly of *A. frenatus* relative to *A. princeps* (Fig. 1). A second major clade inferred and strongly supported (BS ⩾ 99%, PP = 1.00) with a consistent topology throughout all analyses is composed of eight species: *Anolis aeneus*, *Anolis bonairensis*, *Anolis extremus*, *Anolis griseus*, *Anolis luciae*, *Anolis richardi*, *Anolis roquet*, and *Anolis trinitatis*. This group of species has previously been recognized as the *roquet* species group or series (see Section 4.2.2), and therefore will be hereafter called the *roquet* clade. The third major clade inferred with a consistent topology and strongly supported in all analyses (BS = 83%, PP = 1.00) is composed of five species: *Anolis*

[1] These two groups differ from the *latifrons* series of Etheridge (1959), which is a more inclusive group that contains all species in the *Dactyloa* clade except the species previously placed in the genus *Phenacosaurus*.

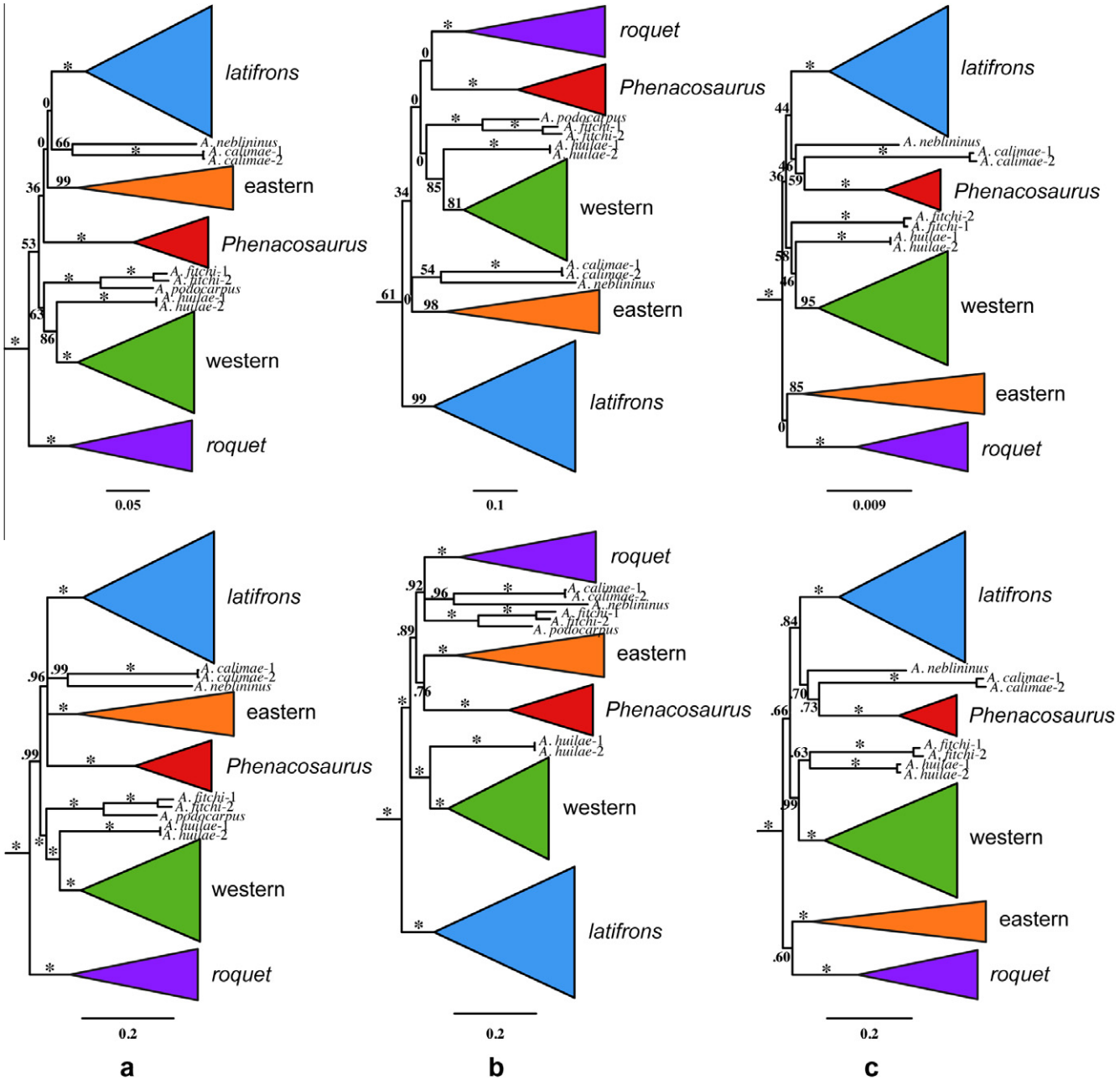


**Fig. 2.** Relationships among the five major clades within *Dactyloa* as inferred in the likelihood (top) and Bayesian (bottom) analyses of the (a) ND2–COI–RAG1, (b) ND2–COI, and (c) RAG1 datasets. The position of those species not included in any of the major clades is also indicated. The size of the triangles is proportional to the number of sampled specimens in each clade. Bootstrap support values and Bayesian posterior probabilities are shown above the branches. Asterisks (*) indicate PP = 1.0 or BS = 100%.

*anatoloros*, *Anolis jacare*, *Anolis punctatus*, *Anolis tigrinus*, and *Anolis transversalis*. All of these species are distributed in Amazonia, the northern portion of the eastern cordillera of the Colombian Andes and the Venezuelan Andes, so hereafter this clade will be referred to as the eastern clade (Fig. 3). A fourth clade inferred and strongly supported in all analyses (BS = 76%, PP = 1.00) is composed of seven species: *Anolis aequatorialis*, *Anolis anoriensis*, *Anolis chloris*, *Anolis festae*, *Anolis gemmosus*, *Anolis peraccae*, and *Anolis ventrimaculatus*. This clade is distributed in the western and central cordilleras of the Colombian Andes, the western slope of the Ecuadorian Andes and the Pacific lowlands of Panama, Colombia and Ecuador and hereafter will be referred to as the western clade (Fig. 3). The relationships within the western clade are fairly consistent across analyses (Fig. 1). The fifth major clade inferred and strongly supported in all analyses (BS = 100%, PP = 1.00) is composed of five species: *Anolis euskalerriari*, *Anolis heterodermus*, *Anolis inderenae*, *Anolis nicefori*, and *Anolis vanzolinii*. All these species were previously placed in the genus *Phenacosaurus* (see Section 4.2.5) and hereafter this clade will be referred to as the *Phenacosaurus* clade. However *Anolis neblininus*, also previously placed in the genus *Phenacosaurus*, was not inferred as part of this clade. Species relationships within the *Phenacosaurus* clade are consistent, except in the analyses of the RAG1 dataset (Fig. 1c) in which *A. heterodermus* is not inferred as monophyletic, as *A. inderenae* is nested within it.

Despite the consistent inference of these five major clades, the relationships among them vary significantly among gene regions, phylogenetic methods and even among different data partitioning strategies (Fig. 2). This variation reflects the low nodal support found in most of the basal nodes within *Dactyloa* in most of the

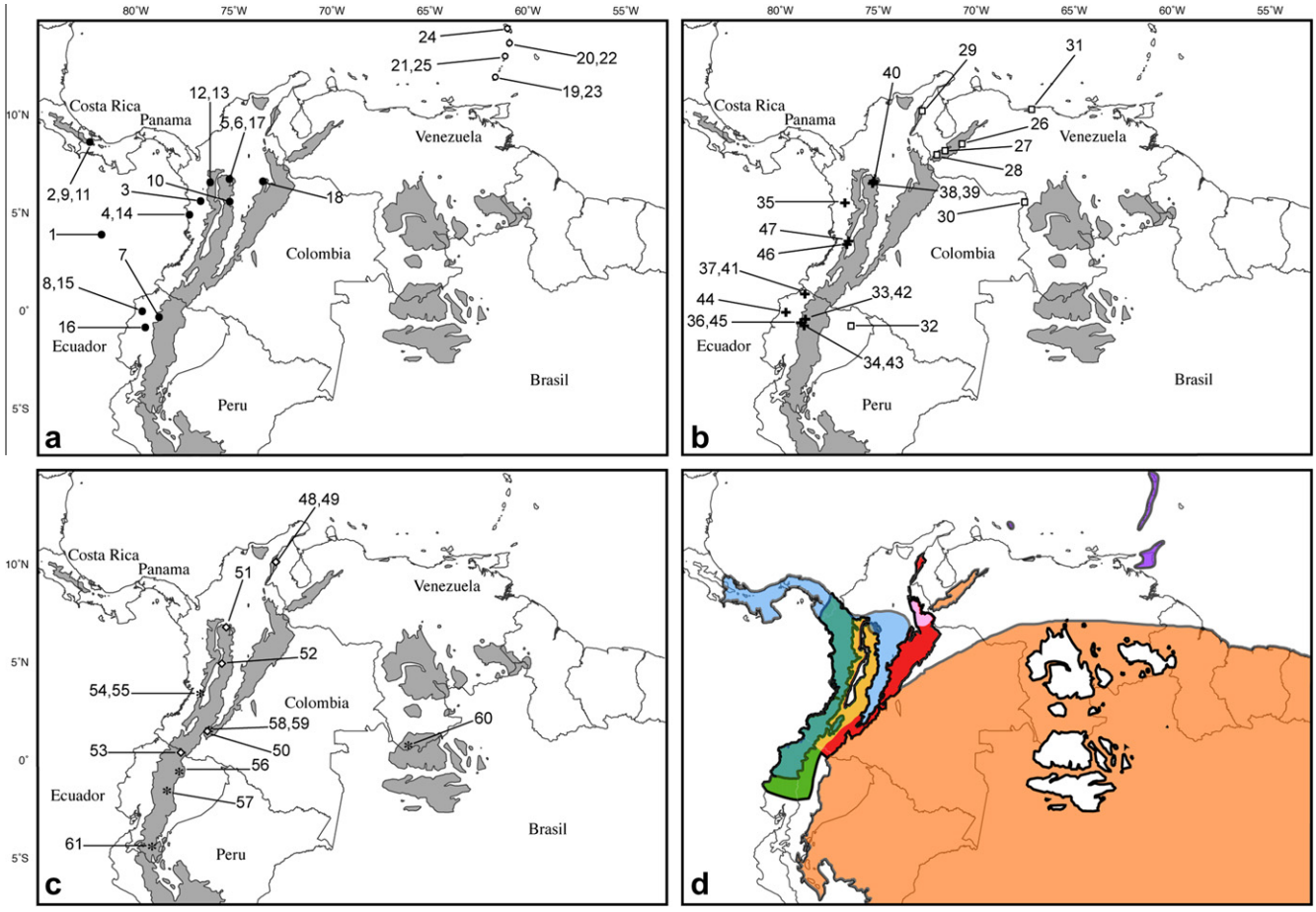


**Fig. 3.** Map indicating the localities of the samples included in this study grouped by major clade. Numbers in parenthesis correspond to those used in Appendix A to distinguish different samples referred to the same species. a. ***latifrons* clade (•)**: 1. *A. agassizi*, 2. *A. casildae*, 3. *A. chocorum* (1), 4. *A. chocorum* (2), 5. *A. danieli* (1), 6. *A. danieli* (2), 7. *A. fraseri* (1), 8. *A. fraseri* (2), 9. *A. frenatus* (1), 10. *A. frenatus* (2), 11. *A. insignis*, 12. *A. maculigula* (1), 13. *A. maculigula* (1), 14. *A. princeps* (1), 15. *A. princeps* (2), 16. *A. princeps* (3), 17. *A.* sp2, 18. *A.* sp1; ***roquet* clade (○)**: 19. *A. aeneus*, 20. *A. extremus*, 21. *A. griseus*, 22. *A. luciae*, 23. *A. richardi*, 24. *A. roquet*, 25. *A. trinitatis*; b. **eastern clade (□)**: 26. *A. anatoloros*, 27. *A. jacare* (1), 28. *A. jacare* (2), 29. *A. jacare* (3), 30. *A. punctatus*, 31. *A. tigrinus*, 32. *A. transversalis*; **western clade (+)**: 33. *A. aequatorialis* (1), 34. *A. aequatorialis* (2), 35. *A. chloris* (1), 36. *A. chloris* (2), 37. *A. chloris* (3), 38. *A. anoriensis* (1), 39. *A. anoriensis* (2), 40. *A. anoriensis* (3), 41. *A. festae*, 42. *A. gemmosus* (1), 43. *A. gemmosus* (2), 44. *A. peraccae* (1), 45. *A. peraccae* (2), 46. *A. ventrimaculatus* (1), 47. *A. ventrimaculatus* (2); c. ***Phenacosaurus* clade (◇)**: 48. *A. euskalerriari* (1), 49. *A. euskalerriari* (1), 50. *A. heterodermus* (1), 51. *A. heterodermus* (2), 52. *A. heterodermus* (3), 53. *A. vanzolinii*; **species not included in any of the major clades** (∗): 54. *A. calimae* (1), 55. *A. calimae* (2), 56. *A. fitchi* (1), 57. *A. fitchi* (2), 58. *A. huilae* (1), 59. *A. huilae* (2), 60. *A. neblininus*, 61. *A. podocarpus*; d. approximate geographic distributions of the *latifrons* (blue), *roquet* (purple), eastern (orange), western (green), and *Phenacosaurus* (red) clades. Yellow areas indicate regions where the western and *Phenacosaurus* clades overlap; teal areas indicate regions where the *latifrons* and western clades overlap; pink areas indicate regions where the eastern and *Phenacosaurus* clades overlap. Gray areas indicate elevations higher than ~1400 m (4500 ft).

analyses (BS = 0–53%, PP = 0.66–1.00; Fig. 2). In some cases, however, conflicting relationships were strongly supported by different data sets: e.g., Bayesian analyses of the ND2–COI–RAG1 dataset showed high support for the *roquet* clade as the sister clade to the rest of *Dactyloa* (PP = 0.99; Fig. 2a), whereas in the Bayesian ND2–COI analyses, the *latifrons* clade is strongly supported as the sister clade to the rest of *Dactyloa* (PP = 1.00; Fig. 2b). Interestingly, the Bayesian analyses of the RAG1 dataset do not strongly support any particular clade as the sister clade to all the other clades (Fig. 2c).

The positions of *Anolis calimae*, *A. fitchi*, *A. huilae*, *A. neblininus*, and *A. podocarpus* were inconsistent across gene regions and/or phylogenetic methods (Fig. 2).

### 3.3. Hypothesis testing

Results from the topological tests are summarized in Table 4. The non-monophyly of *Dactyloa* (including *Phenacosaurus*) and the non-monophyly of the *roquet* series were strongly rejected by WSR and AU tests ($P \leqslant 0.002$). Monophyly of the previously recognized subgroups *aequatorialis*, *latifrons* (as traditionally circumscribed; see Section 4.2.1), and *punctatus* was strongly rejected by the data in WSR and AU tests ($P < 0.001$). Monophyly of *Phenacosaurus* as traditionally circumscribed—which includes the sampled species *A. euskalerriari*, *A. heterodermus*, *A. inderenae*, *A. neblininus*, *A. nicefori*, and *A. vanzolinii*—was rejected by the WSR test ($P < 0.001$), but not by the AU test ($P = 0.194$). In addition, the exclusion of *Phenacosaurus* (as traditionally circumscribed) from *Dactyloa* was rejected by WSR and AU tests ($P < 0.001$).

The post burn-in sample of the Bayesian analysis consisted of 37,500 trees. The 95% set of credible trees from the post burn-in sample contained 2934 distinct topologies. Topologies containing monophyletic groups corresponding in composition to the *aequatorialis*, *latifrons* (as traditionally circumscribed), and *punctatus* series, *Phenacosaurus* (as traditionally circumscribed), as well as those excluding *Phenacosaurus* (as traditionally circumscribed) from *Dactyloa*, were not present in the 95% credible set of post burn-in topologies, thus rejecting those hypotheses. In agreement with WSR and AU tests, the Bayesian tests supported the monophyly of the *Dactyloa* (including *Phenacosaurus*) and *roquet* clades, as all trees from the 95% credible set (as well as the entire post burn-in sample) contained these groups.

**Table 4**
Results of explicit tests of various phylogenetic hypotheses. Differences between likelihood scores ($\Delta - \ln L$) of optimal trees from unconstrained analyses and those from analyses employing topological constraints corresponding to specified hypotheses as well as *P*-values for the likelihood-based Wilcoxon signed ranks (WSR) and approximately unbiased (AU) tests are given. For the Bayesian approach (B), the presence (+; indicating failure to reject the test hypothesis) or absence (–; indicating rejection of the test hypothesis) of the topologies conforming to the test hypothesis in the 95% credible set of trees is shown. Significant results are indicated with an asterisk (*).

| Test hypothesis | $\Delta - \ln L$ | *P*-value | | B |
|---|---|---|---|---|
| | | WSR | AU | |
| *Dactyloa* not monophyletic | 67.181 | <0.001* | <0.001* | –* |
| *roquet* series not monophyletic | 33.416 | <0.001* | 0.002* | –* |
| *aequatorialis* series | 317.658 | <0.001* | <0.001* | –* |
| *latifrons* series[a] | 141.711 | <0.001* | <0.001* | –* |
| *punctatus* series | 403.72 | <0.001* | <0.001* | –* |
| *Phenacosaurus*[b] | 8.441 | <0.001* | 0.194 | –* |
| *Dactyloa* excluding *Phenacosaurus*[b] | 20.055 | <0.001* | 0.001* | –* |

[a] As traditionally circumscribed, which includes (of the species sampled in this study) *A. casildae*, *A. danieli*, *A. fraseri*, *A. frenatus*, *A. insignis*, *A. microtus*, and *A. princeps*.
[b] As traditionally circumscribed, which includes (of the species sampled in this study) *A. euskalerriari*, *A. heterodermus*, *A. inderenae*, *A. neblininus*, *A. nicefori*, and *A. vanzolinii*.

## 4. Discussion

All datasets (RAG1, ND2–COI, ND2–COI–RAG1), analyzed under different phylogenetic methods (likelihood and Bayesian) and data partition strategies (Table 1), yielded with few exceptions strong nodal support for the *Dactyloa* clade and five major clades within it (see below), two of which have not been recognized previously. Relationships among these five subclades vary among datasets and/or phylogenetic methods, and in some cases, contradictory results were strongly supported. Given the absence of a consistent pattern in the basal relationships within *Dactyloa* (Fig. 2), we consider the relationships among these five clades as unresolved.

Of the insertion/deletion (indel) events inferred from the gaps inserted in the RAG1 alignment, all three that exhibit variation within *Dactyloa* corroborate results inferred from the DNA sequences. Two such indels further support the monophyly of two *Dactyloa* species: Both samples of *A. aequatorialis* have an inferred insertion in positions 217–219 (which corresponds to the protein residue 106 of the mouse RAG1 protein; Melville and Hale, 2009; Sadofsky et al., 1993), and all (and only the) samples of *A. fitchi* have an inferred deletion in positions 76–87 (corresponding to residues 59–62). In addition, an inferred deletion of 15 bases (between positions 280–294 in the RAG1 alignment; corresponding to residues 127–131) was present in all specimens of the species *A. chloris*, *A. festae* and *A. peraccae*, further supporting this clade, which was consistently inferred in all analyses (see Section 4.2). All gaps inserted in the RAG1 alignment are located in the N-terminal domain of the RAG1 gene, in agreement with the results of Melville and Hale (2009), but two of the indels in *Dactyloa* species were located outside the highly variable region within the N-terminal domain identified by them.

### 4.1. *Monophyly of* Dactyloa

The *Dactyloa* clade, defined in the present paper as the clade originating with the most recent common ancestor of the species included in the genus *Dactyloa* by Savage and Guyer (1989), which also includes the species previously placed in the genus *Phenacosaurus*, is consistently inferred and strongly supported in all analyses (Fig. 2). This result is further supported by topology tests, which strongly rejected the alternative hypothesis of non-monophyly of *Dactyloa* and the hypothesis of *Dactyloa* excluding *Phenacosaurus* species. These results are in agreement with previous studies that have supported the inclusion of *Phenacosaurus* species within *Anolis* (Jackman et al., 1999; Nicholson et al., 2005; Poe, 1998, 2004).

### 4.2. *Major* Dactyloa *subclades*

Each of the five major clades consistently inferred within *Dactyola* has a coherent geographic range and some have species compositions similar to that of previously recognized species groups (Williams, 1976b) or series (Savage and Guyer, 1989) based on morphological characters.

#### 4.2.1. *The* latifrons *clade*

The *latifrons* clade is distributed in the pacific middle elevations and lowlands of Colombia (including Malpelo Island), Costa Rica (where it also occurs in the Atlantic lowlands), Ecuador, and Panama, and the Colombian inter-Andean valleys, below 1500 m elevation (except *A. danieli*, which ranges from 1700 to 2200 m; Fig. 3). Males of all sampled species in the *latifrons* clade except *A. chocorum* reach a snout-to-vent length larger than 100 mm, a characteristic that gave the group its earlier name of the giant anoles group (Dunn, 1937). All the species of the *latifrons* series that were sampled in this study were inferred as part of the *latifrons* clade. Three additional species inferred in the *latifrons* clade were not previously placed in the *latifrons* species group (according to Williams, 1976b) or series (according to Savage and Guyer, 1989): *A. agassizi*, which was not assigned to any series or species group (based on the presence of caudal autotomy throughout ontogeny [a condition otherwise found among *Dactyloa* species only in the *roquet* series], a reduced dewlap in males, and minute dorsal scales [not found in any other *Dactyloa* species]; Etheridge, 1959); *A. chocorum*, which was previously assigned to the *punctatus* series or species group (based on its similarity in scalation and size with *A. punctatus* and *A. transversalis*; Williams and Duellman, 1967); and *A. maculigula*, previously assigned to the *aequatorialis* series or species group (based on its scalation similarities with *A. eulaemus*; Williams, 1984). The inclusion of these three species presumably explains why topology tests strongly rejected the monophyly of the previously recognized *latifrons* group.

Within the *latifrons* clade, the inference that *A. frenatus* is paraphyletic relative to *A. princeps* is not surprising, as it remains unclear whether *A. frenatus*, *A. princeps* and *A. latifrons* (not included in this study), are separate species (Ayala and Castro, unpublished manuscript; Savage and Talbot, 1978; Williams, 1988). The main morphological characters used to distinguish these species are the size and shape of the superciliary scales and the coloration in life—a trait known to vary physiologically and exhibit significant intraspecific geographic variation: *latifrons* is emerald green, *princeps* is olive green and *frenatus* is brownish green (Ayala and Castro, unpublished manuscript). All three species occur at low elevations and replace each other geographically: *A. latifrons* occurs on the Pacific coast of Panama and in northern and central Colombia, *A. princeps* occurs on the central and southern Pacific coast of Colombia and northern Ecuador, and *A. frenatus* occurs in the Atlantic lowlands of Costa Rica and Panama, in southwestern Costa Rica, and the in northern lowlands and inter-Andean valleys of Colombia (Ayala and Castro, unpublished manuscript; Savage and Talbot, 1978).

#### 4.2.2. *The* roquet *clade*

The *roquet* clade corresponds to the previously described *roquet* species group or series (e.g., Underwood, 1959; Gorman and Atkins, 1967; Lazell, 1972; Williams, 1976a; Savage and Guyer, 1989; Creer et al., 2001). This clade is distributed in the southern Lesser Antilles, from Martinique to Grenada, as well as the islands

of Bonaire, Tobago and Trinidad (where *A. aeneus* and *A. trinitatis* have been introduced; Gorman and Dessauer, 1965, 1966; Gorman and Atkins, 1967) and Guyana (where *A. aeneus* has been introduced; Gorman et al., 1971) (Fig. 3). Topology tests further support the monophyly of this group by rejecting the non-monophyly hypothesis (Table 4). The relationships within the *roquet* clade inferred in this study were consistent across all datasets and analyses, and identical to those inferred by Creer et al. (2001) based on DNA sequences (same mitochondrial ND2 region used in this study) and allozyme data; however, the positions of *A. griseus* and *A. richardi* differ from those inferred by Giannasi et al. (2000) based on mitochondrial cytochrome b sequences (600 b). If the discrepancy is the result of an error (e.g., a mislabeled sample or contamination) in one of the data sets, our results suggest that the data set of Creer et al. (2001) is not the one with the error, given that we inferred identical topologies from different samples of the same species with the same and different genes.

#### 4.2.3. The eastern clade

The eastern clade, distributed in Amazonia, the northern portion of the eastern cordillera of the Andes of Colombia and the Venezuelan Andes (Fig. 3), shows a consistent topology among datasets and analyses. All the species in this clade were previously placed in the *punctatus* species group (Williams, 1976b) or series (Savage and Guyer, 1989), except for *A. tigrinus*, which was classified in the *tigrinus* series or species group. However, six additional species included in this study (*A. calimae*, *A. chloris*, *A. chocorum*, *A. festae*, *A. huilae*, *A. peraccae*) were also assigned to the *punctatus* series based on morphological characters, but none of them was inferred as part of the eastern clade in any of our analyses. The eastern clade exhibits a distinct geographic separation between its two primary subclades: species with a wide distribution throughout Amazonia—*A. punctatus* and *A. transversalis*—are strongly supported as sister species (BS = 100%, PP = 1.00), and species distributed in the eastern cordillera of the Andes of Colombia and the Venezuelan Andes—*A. anatoloros*, *A. jacare*, and *A. tigrinus*—also form a strongly supported clade (BS = 85–100%, PP = 1.00).

#### 4.2.4. The western clade

The western clade is distributed in the western and central cordilleras of the Andes of Colombia, the western slope of the Andes of Ecuador and the Pacific lowlands of Panama, Colombia and Ecuador (Fig. 3). Within it two subclades are consistently inferred with strong support (BS = 87–100%, PP = 0.99–1.00): the first is composed of species of smaller size, including *A. chloris*, *A. festae*, and *A. peraccae* (*A. chloris*, the largest species, has a maximum snout-vent length [SVL] of 62 mm for adult males (Williams et al., 1995)), with a lowland (up to 1000 m of elevation), humid forest distribution. In addition to support from DNA sequences, this subclade is characterized by a deletion of 15 bases in the RAG1 gene region. All of these species were previously placed in the *punctatus* species group or series based on morphological characters, though other species previously included in that group (i.e., *A. anatoloros*, *A. calimae*, *A. chocorum*, *A. huilae*, *A. jacare*, *A. punctatus*, *A. transversalis*) were not inferred to be closely related. The second subclade is composed of species of larger size, including *A. aequatorialis*, *A. anoriensis*, *A. gemmosus*, and *A. ventrimaculatus* (*A. gemmosus*, the smallest species, has a maximum SVL of 66 mm for adult males (Williams et al., 1995)), with an elevational distribution from 1500 to 2000 m. All species in this second subclade were previously placed in the *aequatorialis* species group (Williams, 1976b) or series (Savage and Guyer, 1989) (see Williams and Duellman (1984) for the assignment of *A. gemmosus*). In most analyses, *Anolis huilae* was inferred as the sister group of the western clade, with nodal support ranging from weak to strong (Fig. 2), which suggests that this species is more closely related to the western clade than to any of the four other major clades within *Dactyloa*. However, based on the exclusively western geographic distribution of the species included in the western clade and the eastern distribution of *A. huilae*, we do not consider this species part of the western clade.

#### 4.2.5. The Phenacosaurus clade

The *Phenacosaurus* clade is found in the Andean regions of Colombia, Ecuador, and Venezuela, from 1300 m to 3750 m of elevation (Ayala and Castro, unpublished manuscript; Etheridge, 1959; Fig. 3), reaching the highest elevations of any *Anolis* species (MRC, personal observation). This clade includes all of the sampled species previously placed in the genus *Phenacosaurus* (Lazell, 1969; Myers and Donnelly, 1996; Poe and Yañez-Miranda, 2007; Williams et al., 1996), with the exception of *A. neblininus*, which in this study was inferred in different positions across analyses but was never inferred to be more closely related to the *Phenacosaurus* clade than was *A. calimae*, which was not previously placed in the genus *Phenacosaurus* (see below). In agreement with previous studies (Jackman et al., 1999; Nicholson et al., 2005; Poe, 1998, 2004), the *Phenacosaurus* species were inferred, with strong support, to be nested within *Dactyloa* and therefore also within *Anolis*. Results of the topology tests further support this inference by rejecting the alternative hypothesis of *Dactyloa* excluding *Phenacosaurus* (Table 4). Within the *Phenacosaurus* clade, the two earliest diverging species, *A. euskalerriari* and *A. nicefori*, have small body size (maximum male SVL = 53 mm and 63 mm, respectively (Williams et al., 1995)), while the more deeply nested *A. heterodermus*, *A. inderenae* and *A. vanzolinii* are larger (maximum male SVL = 76 mm, 98 mm, and 104 mm, respectively (Williams et al., 1995)), suggesting an increase in body size within this clade. This more deeply nested clade (*A. heterodermus*, *A. inderenae*, *A. vanzolinii*) corresponds to the *heterodermus* subgroup (characterized by strongly heterogeneous flank scalation and well-developed casquing) of the *heterodermus* group (characterized by flank scale heterogeneity and lamellar subdigital scales, i.e., those that are wider than long, with a distal free edge, extending under the most proximal phalanges of all digits, including pedal digit IV) (Williams et al., 1996).[2] The *heterodermus* group (including *A. nicefori*, *A. tetarii* and the *heterodermus* subgroup) was also supported by our data, though *A. tetarii* was not sampled.

*Anolis heterodermus* is the only species in the *Phenacosaurus* clade that occurs in all three cordilleras of the Andes of Colombia (>1600 m). The relationships among the multiple samples of this species do not correspond to the geographic distance between them. Samples 1 and 2, which are inferred as sister taxa in all analyses (with weak nodal support), are geographically more distant from each other than either is from sample No. 3 (Fig. 3; Appendix A). In the analysis of the RAG1 dataset, *A. heterodermus* is not inferred as monophyletic, as *A. inderenae* is nested within it (Fig. 1). Williams et al. (1996) suggested previously that *A. heterodermus* may represent more than one species, noting that "the present concept of *P.* [*A.*] *heterodermus* is an unresolved complex of sibling species" (p. 12) because of the large morphological variability observed within this species (e.g., in dorsal crest scalation, detailed in Lazell (1969)). However, previous attempts to recognize new species based solely on morphological characters (i.e., *P.* [*A.*] *richteri* Dunn (1944) and *P.* [*A.*] *paramoensis* Hellmich (1949)) have failed (Lazell, 1969).

When *A. neblininus* (found on Cerro de la Neblina, Venezuela) was described (Myers et al., 1993), it was tentatively placed in

[2] Within the genus *Phenacosaurus* Williams et al. (1996) recognized three groups, the *heterodermus* group including *A. heterodermus*, *A. inderenae*, *A. nicefori*, *A. tetarii* and *A. vanzolinii*, the *orcesi* group including *A. orcesi* and *A. euskalerriari*, and the *neblininus* group, including *A. neblininus* and *A. carlostoddi*.

the genus *Phenacosaurus* and considered closely related to then undescribed *Phenacosaurus* [*Anolis*] *carlostoddi* from the Chimantá Tepui, Venezuela, later described by one of the same authors (Williams et al., 1996). The authors also noted that *A. neblininus* showed an overall morphological resemblance to the non-*Phenacosaurus* species *A. jacare* and *A. nigropuntactus* (which were later regarded as conspecific (Ugueto et al., 2007)). In this study, *A. neblininus* was not consistently inferred as closely related to other *Phenacosaurus* species, which were consistently and strongly supported as a monophyletic group. Only in the analyses of the RAG1 dataset, is *A. neblininus* inferred close to other *Phenacosaurus* species, though with low nodal support (BS = 46%, PP = 0.70; Fig. 2c) and always with *A. calimae* as the sister group of the *Phenacosaurus* clade: (*A. neblininus* (*A. calimae* (*Phenacosaurus* clade))). *Anolis calimae* is distributed in the Valle del Cauca in the central part of the western cordillera of Colombia, between 1300–1800 m (Ayala et al., 1983) and interestingly, it shares some external morphological features with *Phenacosaurus* species (including *A. neblininus*): a robust body, short fore- and hindlimbs (proportional to body length) and short thick toes (characters from Lazell, 1969). In addition, it shares small body size (SVL < 65 mm; Williams et al., 1995) with the two earliest diverging species of the *Phenacosaurus* clade and *A. neblininus*. In other analyses where *A. neblininus* is not inferred as closely related to the *Phenacosaurus* clade, the clade (*A. calimae*, *A. neblininus*) is inferred with variable support (BS = 54–66%, PP = 0.96–0.99; Fig. 2), suggesting a close relationship between these two geographically distant species. However, despite the morphological similarity between these two species with the species in the *Phenacosaurus* clade, the molecular data presented here are ambiguous about whether *A. neblininus* is closely related to *Phenacosaurus* species.

### 4.3. Previously recognized subgroups

The non-monophyly of the *Dactyloa* clade and the *roquet* series were strongly rejected by all topology tests (Table 4). These clades were inferred in the optimal topologies (i.e., maximum likelihood tree and Bayesian consensus tree) with strong support, and the rejection of their non-monophyly in the topology tests provides further support for their monophyly. The hypotheses of monophyly of the *aequatorialis*, *latifrons* (as traditionally circumscribed) and *punctatus* series were strongly rejected by all topology tests applied (Table 4), a result that confirms the caveats mentioned by E.E. Williams while describing these subgroups of mainland *Anolis*: morphological characters used for series delimitation might show a high degree of convergence and parallelism among species (e.g., 1976b, p. 260), and on occasion, series were described only for convenience (e.g., 1979, p. 10). Another factor, not considered by Williams, is that series were sometimes based on shared ancestral characters, which are not indicative of close phylogenetic relationships.

### 4.4. Taxonomic implications

The results of our phylogenetic analyses and topology tests indicate that several previously recognized series/species groups are not monophyletic, suggesting the need for a revised taxonomy. However, because roughly half of the currently recognized species were not included in the present study, we will only make some preliminary taxonomic suggestions. The five well-supported clades described above (Section 4.2) are the most obvious candidates for formal taxonomic recognition. In some cases it seems appropriate, based on similarity in diagnostic characters and/or species composition, to apply previously used names to those clades. Previously used names have commonly combined the name of an included species with the rank of species group (e.g., Williams, 1976b) or series (e.g., Savage and Guyer, 1989); however, because the species group terminology was based on inclusion of the taxa in question with a larger series (the *latifrons* series of Etheridge, 1959), and because we have recognized that group as the *Dactyloa* clade, we will consider only the series terminology in our discussion.

The *latifrons* clade corresponds roughly to the *latifrons* series of previous authors (e.g., Savage and Guyer, 1989). All of the species of the *latifrons* series that were sampled in this study are in the *latifrons* clade, though the *latifrons* clade additionally includes *A. agassizi* (previously unassigned to a series), *A. chocorum* (previously included in the *punctatus* series) and *A. maculigula* (previously included in the *aequatorialis* series), and all but one (*A. chocorum*) exhibit large body size. Given that *A. agassizi* was not previously assigned to a series, its inclusion in the *latifrons* series would pose no taxonomic problems; inclusion of the other two species would necessitate only that their previous taxonomic assignments be considered incorrect. Whether the species *A. apollinaris*, *A. latifrons*, *A. propinquus*, *A. purpurescens* and *A. squamulatus* (previously included in the *latifrons* series but currently lacking molecular data) belong to this clade remains to be determined. Nevertheless, the geographic distributions, except for that of *A. squamulatus*, and large body sizes of these species, except possibly for *A. purpurescens* (known from relatively few specimens), are consistent with their inclusion in the *latifrons* clade. In addition, *Anolis ibanezi* (not explicitly included in the *latifrons* series and lacking molecular data) may be a member of this clade given its hypothesized close relationship with *A. chocorum* (Poe et al., 2009) and geographic distribution. Although the inclusion of *A. latifrons* in the *latifrons* clade seems likely based on its presumed close relationship to *A. frenatus* and *A. princeps* (see Section 4.2.1), we will not formally name the clade as the *latifrons* series here. Because the formal series name is based on the name of that species, it seems preferable to defer formal naming until an explicit phylogenetic analysis confirms inclusion of *A. latifrons* in this clade.

The *roquet* clade corresponds closely to the *roquet* series of previous authors (e.g., Savage and Guyer, 1989; Creer et al., 2001) in that all species included in the *roquet* clade were previously included in the *roquet* series. The only other species previously included in the *roquet* series, *A. blanquillanus*, was not sampled in this study, but can be assigned to the *roquet* clade (and series), as the sister group of *A. bonairensis*, based on the results of previous phylogenetic studies (e.g., Yang et al., 1974; Creer et al., 2001).

The *Phenacosaurus* clade corresponds closely to the genus *Phenacosaurus* of previous authors (e.g, Williams et al., 1996). All of the species included in the *Phenacosaurus* clade were previously included in the genus *Phenacosaurus*. *Anolis neblininus* was previously included in the genus *Phenacosaurus*; however, in our study evidence for the inclusion of this species in the *Phenacosaurus* clade is ambiguous. Evidence is also ambiguous concerning the inclusion of *A. calimae*, which was not previously included in *Phenacosaurus* but shares some morphological characters with *A. neblininus* and basal members of the *Phenacosaurus* clade (see Section 4.2.5). The inclusion of other species previously considered part of the genus *Phenacosaurus* (*A. bellipeniculus*, *A. carlostoddi*, *A. orcesi*, *A. tetarii*, and *A. williamsmittermeierorum*) remains to be determined. Nevertheless, the inclusion of *A. orcesi*, *A. williamsmittermeierorum*, and *A. tetarii* is consistent with the morphological similarity of the first two species with *A. euskalerriari* and the last one with *A. nicefori*, while the placement of *A. bellipeniculus* and *A. carlostoddi* is less certain based on their morphological similarity with *A. neblininus*.

The two other well-supported clades, the eastern and western clades, do not correspond closely to any previously recognized taxa. If those clades were to be given names using the series terminology, then the western clade would be the *aequatorialis* series and the eastern clade would be the *punctatus* series, based on the inclusion of the species *A. aequatorialis* and *A. punctatus* in the

respective clades, despite considerable differences in the species composition of these clades relative to the *aequatorialis* and *punctatus* series as previously recognized (e.g., Savage and Guyer, 1989). Alternatively, the eastern and western clades could be given new names not based on the series terminology.

The species that were not consistently inferred in one position across our various analyses, and therefore for which we consider relationships uncertain (i.e., *A. calimae*, *A. fitchi*, *A. huilae*, *A. neblininus*, and *A. podocarpus*), are currently left unassigned to any of the five subclades within *Dactyloa*. We plan to develop a more formal and comprehensive taxonomy based on combined phylogenetic analyses of morphological and molecular data that will include species for which molecular data are currently unavailable (Castañeda and de Queiroz, in prep).

### 4.5. Geographic distribution patterns

Each of the five major clades within *Dactyloa* has a coherent (i.e., largely continuous) geographic range, with those of the different clades overlapping to different degrees (Fig. 3). The *latifrons* and western clades overlap in the Chocó region (Pacific lowlands of Colombia and Ecuador, below 1000 m), where three species of the *latifrons* clade and three of the western clade (those of smaller size) are present. The remaining four species of the western clade—those of larger size—are distributed between 1500 and 2000 m and overlap the distribution of the *Phenacosaurus* clade, though exclusively with *A. heterodermus*, which is the only *Phenacosaurus* species distributed in the western cordillera of the Colombian Andes. The remaining four sampled species in the *Phenacosaurus* clade are distributed in the eastern Andes of Colombia, Ecuador and Venezuela, and two of them (*A. euskalerriari*, and *A. nicefori*) fully overlap with the Andean species of eastern clade (*A. anatoloros*, *A. jacare*, and *A. tigrinus*). In contrast, the ranges of several pairs of clades exhibit no overlap (e.g., *latifrons* versus eastern, western versus eastern); however, the only major clade within *Dactyloa* that shows no geographic overlap with any other subclade is the *roquet* clade, distributed in the southern Lesser Antilles.

Within the *latifrons* clade, the clade (*A. agassizi* (*A. insignis*, *A. microtus*)) is consistently inferred as sister to the remaining nine species in all analyses. *Anolis agassizi* is found on Malpelo, an island located 380 km from the Pacific coast of Colombia and 365 km from the coast of Panama. Members of its sister clade are distributed in Central America: *A. insignis* is found at low to middle elevations (<1500 m) on the Atlantic and Pacific slopes of Costa Rica and the Pacific slopes of Panama, and *A. microtus* is found at mid elevations (1000–1500 m) on the Pacific slopes of Costa Rica and Panama (Savage and Talbot, 1978). The remaining species of the *latifrons* clade (except *A. danieli* and *A.* sp2, which occur in the northern regions of central and western cordilleras of Colombia [above 1700 m] and the Magdalena river valley in Colombia, respectively) are present in the Pacific lowlands from Panama to Ecuador. Assuming an oceanic dispersal event, as Malpelo has never been connected to other islands or the mainland (Graham, 1975), *A. agassizi* could be the result of a colonization event from either Central or South America. The direction of surface currents (and surface winds; Fiedler, 2002) around Malpelo (3°59′N, 81°36′W) does not particularly support either alternative; the North Equatorial Countercurrent (NECC, which flows west-to-east) lies between 3°N and 10°N (Sverdrup et al., 1942), becoming the South Equatorial and North Equatorial currents off the continent as it turns south and west and north and west respectively. The NECC shifts further north from the equator during the northern hemisphere's summer (Fiedler, 2002; Sverdrup et al., 1942), so that currents flowing to Malpelo originate from Central or South America depending on the season.

### 4.6. Low nodal support and tempo of the Dactyloa radiation

Despite consistent inference and strong support for five major *Dactyloa* subclades (Section 4.2), there is no consensus between gene regions and/or analyses regarding the relationships among these major clades, as low nodal support values were obtained in most analyses and sometimes conflicting relationships were obtained in different analyses. Within *Anolis*, low support for basal nodes was also found by Jackman et al. (1999) and Poe (2004), and within the *Norops* clade by Nicholson (2002). Poor resolution/support could result from the use of molecular markers that do not provide resolution for the level of interest. In the present study, molecular markers with slower (RAG1 gene) and faster (ND2, COI regions) rates of evolution were sequenced to cover a wide range of node ages and avoid this problem, though the utilization of molecular markers with a broad range of evolutionary rates does not guarantee (even in the absence of conflict in phylogenetic signal) the resolution and strong support of all nodes. Therefore, it is possible that the lack of resolution in the basal nodes of *Dactyloa* results from a rapid radiation within the group—evidenced by very short internodes that are not strongly supported. Jackman et al. (1999) and Nicholson (2002) invoked a rapid radiation to explain the weak support in basal nodes, though Jackman et al. (1999) found statistical support for sequential branching, contradicting the most extreme case of the rapid radiation hypothesis (a star tree or hard polytomy). Without estimating the amount and quality of data necessary to resolve an internode of certain length, and therefore defining what should be considered sufficient/insufficient amounts of data, it would be premature to hypothesize a rapid radiation as the cause for the lack of resolution and/or poor support in the basal nodes of the *Dactyloa* clade.

## Acknowledgments

James Clark, James Schulte, and Omar Torres-Carvajal provided help with data collection, analyses and earlier versions of this paper. This research was partially funded by The George Washington University, Sigma-Xi (GWU Chapter), Cosmos Club Foundation (Young Scholars Award) and the Laboratories of Analytical Biology at the Smithsonian Institution. Many thanks to those who provided tissues: Tito Barros, Wesley Chun, Luis A. Coloma (Universidad Católica de Quito-QCAZ), Richard Glor, Jonathan Losos, Roy McDiarmid, Kirsten Nicholson, Vivian Páez (Universidad de Antioquia-MHUA) and James Schulte. For assistance in the field the first author would like to thank Fernando Ayala, Tito Barros, Eduardo Castañeda, Wesley Chun, Paul D.A. Gutiérrez, Juan Pablo Hurtado, Gustavo A. Londoño, Juan L. Parra, Gilson Rivas and Julian Velasco. Jonathan B. Losos helped track down locality data for some of the sequences obtained from GenBank. Derrick Zwickl provided help with site-likelihood estimations in GARLI. The Instituto Humboldt (thanks to Fernando Gast and Mauricio Alvarez) facilitated permits in Colombia. In Venezuela, access to tissue samples was granted under the access to genetic resources permit between the IVIC–MINAMB; Tito Barros (MBLUZ), associate researcher of IVIC, loaned the tissues. Cristiano Moreira helped in preparing the distribution map. Laboratory work was performed at the Laboratories of Analytical Biology-Smithsonian Institution (thanks to Lee Weight) and Universidad de Los Andes, Colombia.

## Appendix A

See Table A1

## Appendix B

See Table B1

**Table A1**
List of specimens used in this study, including voucher numbers, collection localities, including country, state, province/department (or equivalent unit) and specific locality when available, and GenBank accession numbers. Acronyms are as follows: LSUMZ = Louisiana State University Museum of Zoology, MBLUZ = Museo de Biología de La Universidad del Zulia, MHNLS = Museo de Historia Natural La Salle, Caracas–Venezuela, MHUA = Museo de Herpetología, Universidad de Antioquia, MHUA-T = Colección de Tejidos Museo de Herpetología, Universidad de Antioquia, MVUP = Museo de Vertebrados, Universidad de Panama, MVZ = Museum of Vertebrate Zoology, University of California at Berkeley, SNOMNH = Sam Noble Oklahoma Museum of Natural History, QCAZ = Museo de Zoología, Pontificia Universidad Católica del Ecuador, USNM = National Museum of Natural History, FBC and PT = field numbers of Félix B. Cruz, JBL = field numbers of Jonathan B. Losos, JMR and J. Renjifo = field numbers of Juan Manuel Renjifo, JMS = field numbers of Jay M. Savage, KEN = field numbers of Kirsten E. Nicholson, MRC = field numbers of María del Rosario Castañeda, REG = field numbers of Richard E. Glor. For species with multiple samples, numbers in parentheses correspond to those used in Fig. 3.

| Species | Voucher number | Collecting locality | GenBank no. |
|---|---|---|---|
| *Anolis aeneus* | JBL 442 | Grenada, St. George, Grand Anse Bay[a] | **ND2**: AF055950[b] |
| | USNM 319162 | Grenada, St. George, West end of Grand Anse Bay | **COI**: JN112719 |
| | | | **RAG1**: JN112592 |
| *A. aequatorialis* | QCAZ 6855 | (1) Ecuador, Pichincha, Mindo, on road from Mariposas de Mindo to Mindo Garden | **ND2**: JN112662 |
| | | | **COI**: JN112720 |
| | | | **RAG1**: JN112593 |
| | QCAZ 6883 | (2) Ecuador, Pichincha, El Placer, on road to Conchacato, Río Chisinche | **ND2**: JN112663 |
| | | | **COI**: JN112721 |
| | | | **RAG1**: JN112594 |
| *A. agassizi* | KEN 2004-2 | Colombia, Gorgona Island | **ND2**: JN112667 |
| | | | **COI**: JN112722 |
| | | | **RAG1**: JN112595 |
| *A. anatoloros* | MHNLS 17872 | Venezuela, Barinas, San Isidro | **ND2**: JN112668 |
| | | | **COI**: JN112723 |
| | | | **RAG1**: JN112596 |
| *A. anoriensis* | MHUA-T 516 | (1) Colombia, Antioquia, Anorí, El Retiro, Hacienda Chaquiral | **ND2**: JN112664 |
| | | | **COI**: JN112734 |
| | | | **RAG1**: JN112607 |
| | MHUA-T 517 | (2) Colombia, Antioquia, Anorí, El Retiro, Alto de La Forzosa | **ND2**: JN112665 |
| | | | **COI**: JN112735 |
| | | | **RAG1**: JN112608 |
| | MHUA 11568 (MHUA-T0715) | (3) Colombia, Antioquia, Anorí, Cañadahonda | **ND2**: JN112666 |
| | | | **COI**: JN112736 |
| | | | **RAG1**: JN112609 |
| *A. bonairensis* | LSUMZ 5464 | Netherlands Antilles, Bonaire | **ND2**: AF317070[c] |
| *A. calimae* | MRC 118 | (1) Colombia, Valle del Cauca, on road to San Antonio, Television tower | **ND2**: JN112669 |
| | | | **COI**: JN112724 |
| | | | **RAG1**: JN112597 |
| | MRC 119 | (2) Colombia, Valle del Cauca, on road to San Antonio, Television tower | **ND2**: JN112670 |
| | | | **COI**: JN112725 |
| | | | **RAG1**: JN112598 |
| *A. casildae* | JMS 214 | Panama, Chiriquí, near STRI-Fortuna Biological Station | **ND2**: AY909745[d] |
| | | | **COI**: JN112726 |
| | | | **RAG1**: JN112599 |
| *A. chloris* | MRC 126 | (1) Colombia, Chocó, Quibdó, Tutunendo | **ND2**: JN112673 |
| | | | **COI**: JN112729 |
| | | | **RAG1**: JN112602 |
| | QCAZ 6877 | (2) Ecuador, Pichincha, La Unión del Toachi, Centro de Interpretación Ambiental Otongachi, Otonga Foundation | **ND2**: JN112671 |
| | | | **COI**: JN112727 |
| | | | **RAG1**: JN112600 |
| | QCAZ 6920 | (3) Ecuador, Esmeraldas, San Lorenzo, grounds of Hosteria Tundaloma | **ND2**: JN112672 |
| | | | **COI**: JN112728 |
| | | | **RAG1**: JN112601 |
| *A. chocorum* | MRC 123 | (1) Colombia, Chocó, Quibdó, Tutunendo | **ND2**: JN112674 |
| | | | **COI**: JN112730 |
| | | | **RAG1**: JN112603 |
| | MRC 134 | (2) Colombia, Chocó, Bajo Baudó, Pilizá | **ND2**: JN112675 |
| | | | **COI**: JN112731 |
| | | | **RAG1**: JN112604 |
| *A. danieli* | MHUA 11564 (MHUA-T 0711) | (1) Colombia, Antioquia, Anorí, Cañadahonda | **ND2**: JN112676 |
| | | | **COI**: JN112732 |
| | | | **RAG1**: JN112605 |
| | MHUA 11567 (MHUA-T 0714) | (2) Colombia, Antioquia, Anorí, Cañadahonda | **ND2**: JN112677 |
| | | | **COI**: JN112733 |
| | | | **RAG1**: JN112606 |
| *A. euskalerriari* | MBLUZ 934 | (1) Venezuela, Zulia, Sierra de Perijá, Villa del Rosario | **ND2**: JN112678 |
| | | | **COI**: JN112737 |
| | | | **RAG1**: JN112610 |
| | MBLUZ 935 | (2) Venezuela, Zulia, Sierra de Perijá, Villa del Rosario | **ND2**: JN112679 |
| | | | **COI**: JN112738 |
| | | | **RAG1**: JN112611 |

(*continued on next page*)

**Table A1** (*continued*)

| Species | Voucher number | Collecting locality | GenBank no. |
|---|---|---|---|
| *A. extremus* | USNM 321940 | St. Lucia, Castries Quarter | **ND2**: AF317065[c] |
| | USNM 321945 | St. Lucia, Castries Quarter, grounds of Cunard La Toc Hotel | **COI**: JN112739 |
| | | | **RAG1**: JN112612 |
| *A. festae* | QCAZ 6930 | Ecuador, Esmeraldas, San Lorenzo, grounds of Hosteria Tundaloma | **ND2**: JN112680 |
| | | | **COI**: JN112740 |
| | | | **RAG1**: JN112613 |
| *A. fitchi* | QCAZ 6742 | (1) Ecuador, Napo, Pacto Sumaco | **ND2**: JN112681 |
| | | | **COI**: JN112741 |
| | | | **RAG1**: JN112614 |
| | QCAZ 6910 | (2) Ecuador, Tungurahua, Río Verde | **ND2**: JN112682 |
| | | | **COI**: JN112742 |
| | | | **RAG1**: JN112615 |
| *A. fraseri* | QCAZ 6862 | (1) Ecuador, Pichincha, Mindo, on road to Mindo Garden at Muyu Mindala Hostal | **ND2**: JN112683 |
| | | | **COI**: JN112743 |
| | | | **RAG1**: JN112616 |
| | QCAZ 6867 | (2) Ecuador, Esmeraldas, Mache Chindú Reserve, Bilsa Biological Station, Río Ducha | **ND2**: JN112684 |
| | | | **COI**: JN112744 |
| | | | **RAG1**: JN112617 |
| *A. frenatus* | JMS 192 | (1) Panama, Chiriquí, near STRI-Fortuna Biological Station | **ND2**: AY909752[d] |
| | | | **COI**: JN112745 |
| | | | **RAG1**: JN112618 |
| | MHUA 11519 (MHUA-T 676) | (2) Colombia, Antioquia, San Luís, Río Claro, El Refugio Natural Reserve | **ND2**: JN112685 |
| | | | **COI**: JN112746 |
| | | | **RAG1**: JN112619 |
| *A. gemmosus* | QCAZ 6851 | (1) Ecuador, Pichincha, Mindo, on road from Mariposas de Mindo to Mindo Garden | **ND2**: JN112686 |
| | | | **COI**: JN112747 |
| | | | **RAG1**: JN112620 |
| | QCAZ 6884 | (2) Ecuador, Pichincha, El Placer, on road to Conchacato, Río Chisinche | **ND2**: JN112687 |
| | | | **COI**: JN112748 |
| | | | **RAG1**: JN112621 |
| *A. griseus* | [Not given] | [Not given] | **ND2**: AY296176[e] |
| | USNM 321983 | St. Vincent, St. Andrew, Kingston Botanical Gardens | **COI**: JN112749 |
| | | | **RAG1**: JN112622 |
| *A. heterodermus* | MRC 145 | (1) Colombia, Huila, Palestina, La Guajira, La Riviera Private Reserve | **ND2**: JN112688 |
| | | | **COI**: JN112752 |
| | | | **RAG1**: JN112625 |
| | MHUA 11265 (MHUA-T 26) | (2) Colombia, Antioquia, Anorí, El Retiro, El Castillo stream | **ND2**: JN112690 |
| | | | **COI**: JN112751 |
| | | | **RAG1**: JN112624 |
| | MHUA 11396 (MHUA-T 232) | (3) Colombia, Caldas, Manizales, Río Blanco | **ND2**: JN112689 |
| | | | **COI**: JN112750 |
| | | | **RAG1**: JN112623 |
| *A. huilae* | MRC 146 | (1) Colombia, Huila, Palestina, Jericó, El Silencio coffee plantation | **ND2**: JN112691 |
| | | | **COI**: JN112753 |
| | | | **RAG1**: JN112626 |
| | MRC 149 | (2) Colombia, Huila, Palestina, Jericó, El Silencio coffee plantation | **ND2**: JN112692 |
| | | | **COI**: JN112754 |
| | | | **RAG1**: JN112627 |
| *A. inderenae* | JMR 3744[a] | Colombia, no further locality data[a] | **ND2**: AY296145[e] |
| | | | **COI**: JN112755 |
| | | | **RAG1**: JN112628 |
| *A. insignis* | MVUP 2021 | Panama, Chiriquí, Reserva Forestal Fortuna | **ND2**: JN112693 |
| | | | **COI**: JN112756 |
| *A. jacare* | MHNLS 17870 | (1) Venezuela, Merida, Antonio Pinto Salinas, Santa Cruz de Mora, La Macana | **ND2**: JN112694 |
| | | | **COI**: JN112757 |
| | | | **RAG1**: JN112629 |
| | MHNLS 17237 | (2) Venezuela, Táchira, Paramo la Negra, on road between Sabana Grande and La Grita, La Guacharita | **ND2**: JN112695 |
| | | | **COI**: JN112759 |
| | | | **RAG1**: JN112630 |
| | MBLUZ 1136 | (3) Venezuela, Zulia, Sierra de Perijá, Villa del Rosario | **ND2**: JN112696 |
| | | | **COI**: JN112758 |
| *A. luciae* | USNM 321960 | St. Lucia, Castries, grounds of Cunard La Toc Hotel | **ND2**: JN112697 |
| | | | **COI**: JN112760 |
| | | | **RAG1**: JN112631 |
| *A. maculigula* | MHUA 11558 (MHUA-T 705) | (1) Colombia, Antioquia, Frontino, Cuevas Peñitas, Don Luis property | **ND2**: JN112698 |
| | | | **COI**: JN112761 |
| | | | **RAG1**: JN112632 |

**Table A1** (*continued*)

| Species | Voucher number | Collecting locality | GenBank no. |
|---|---|---|---|
| | MHUA 11559 (MHUA-T 706) | (2) Colombia, Antioquia, Frontino, Cuevas Peñitas, Don Luis property | **ND2**: JN112699 **COI**: JN112762 **RAG1**: JN112633 |
| *A. microtus* | MVZ 204040 | Costa Rica, Cartago, Refugio Nacional Tapantí | **ND2**: AF055947[b] |
| *A. neblininus* | USNM 322912 | Venezuela, Amazonas, Río Negro, Cerro de la Neblina, 12.5 km NNW of pico Phelps (=pico Neblina) | **ND2**: JN112700 **COI**: JN112763 **RAG1**: JN112634 |
| *A. nicefori* | J. Renjifo 2537 | [not given] | **ND2**: AF055948[b] |
| *A. peraccae* | QCAZ 6869 | (1) Ecuador, Esmeraldas, Mache Chindú Reserve, Bilsa Biological Station | **ND2**: JN112701 **COI**: JN112764 **RAG1**: JN112635 |
| | QCAZ 6879 | (2) Ecuador, Pichincha, La Unión del Toachi, Centro de Interpretación Ambiental Otongachi, Otonga Foundation | **ND2**: JN112702 **COI**: JN112765 **RAG1**: JN112636 |
| *A. podocarpus* | QCAZ 6047 | Ecuador, Loja, Parque Nacional Podocarpus, Romerillos Alto | **ND2**: JN112703 **COI**: JN112780 |
| *A. princeps* | MRC 135 | (1) Colombia, Chocó, Bajo Baudó, Pilizá | **ND2**: JN112706 **COI**: JN112768 **RAG1**: JN112639 |
| | QCAZ 6868 | (2) Ecuador, Esmeraldas, Mache Chindú Reserve, Bilsa Biological Station | **ND2**: JN112704 **COI**: JN112766 **RAG1**: JN112637 |
| | QCAZ 6892 | (3) Ecuador, Los Ríos, Centro Científico Río Palenque | **ND2**: JN112705 **COI**: JN112767 **RAG1**: JN112638 |
| *A. punctatus* | MHNLS 17698 | Venezuela, Amazonas, Atures, 12 km S of Puerto Ayacucho | **ND2**: JN112707 **COI**: JN112769 **RAG1**: JN112640 |
| *A. richardi* | USNM 321792 | Grenada, St. George, SW coast of Grand Anse Bay | **ND2**: JN112708 **COI**: JN112770 **RAG1**: JN112641 |
| *A. roquet* | USNM 321824/5 | France, Martinique, Le Marin, Anse Mitan | **ND2**: JN112709 **COI**: JN112771 **RAG1**: JN112642 |
| *A. tigrinus* | MHNLS 17863 | Venezuela, Vargas, on road Junquito-Colonia Tovar | **ND2**: JN112710 **COI**: JN112772 **RAG1**: JN112643 |
| *A. transversalis* | QCAZ 5936 | Ecuador, Orellana, Yasuní Scientific Station | **ND2**: JN112711 **COI**: JN112773 **RAG1**: JN112644 |
| *A. trinitatis* | [Not given] | [Not given] | **ND2**: AY296204[e] |
| | USNM 321992 | St. Vincent, St. George, Villa town | **COI**: JN112774 **RAG1**: JN112645 |
| *A. vanzolinii* | QCAZ 6926 | Ecuador, Sucumbios, Santa Bárbara, La Bretaña sector, on road between El Playón and El Carmelo | **ND2**: JN112712 **COI**: JN112775 |
| *A. ventrimaculatus* | MRC 091 | (1) Colombia, Valle del Cauca, La Cumbre, Chicoral, La Minga property | **ND2**: JN112713 **COI**: JN112776 **RAG1**: JN112646 |
| | MRC 112 | (2) Colombia, Valle del Cauca, on road to San Antonio, Television tower | **ND2**: JN112714 **COI**: JN112777 **RAG1**: JN112647 |
| *A.* sp1 | MHUA 11455 | Colombia, Santander, San Vicente de Chucurí, Centro, La Cartagena stream, El Castillo property | **ND2**: JN112715 **COI**: JN112778 **RAG1**: JN112649 |
| *A.* sp2 | MHUA 11562 (MHUA-T 704) | Colombia, Antioquia, Anorí, El Roble, La Forzosa forest | **ND2**: JN112716 **COI**: JN112779 **RAG1**: JN112648 |
| Outgroups | | | |
| *A. bimaculatus* | USNM 321912 | St. Christopher, Trinity Palmetto Point, east of Boyd's | **ND2**: AF055930[b] **COI**: JN112781 **RAG1**: JN112650 |
| *A. cupreus* | JMS 71 | Costa Rica, Guanacaste, OTS-Palo Verde Biological Station | **ND2**: JN112717 **COI**: JN112782 **RAG1**: JN112651 |

(*continued on next page*)

**Table A1** (*continued*)

| Species | Voucher number | Collecting locality | GenBank no. |
|---|---|---|---|
| *A. cuvieri* | USNM 321864<br>REG 2104 | Puerto Rico, Arecibo, Reserva Foresta Cambalache<br>Puerto Rico, Arecibo, Reserva Foresta Cambalache | **ND2**: AF055973.2[b]<br>**COI**: JN112783<br>**RAG1**: JN112652 |
| *A. equestris* | JBL RB4<br>USNM 337647 | [Not given]<br>Cuba, La Habana, Playa Jibacoa | **ND2**: AF055978.2[b]<br>**COI**: JN112784<br>**RAG1**: JN112653 |
| *A. lucius* | USNM 498030 | Cuba, Cienfuegos, Cienfuegos Botanical Garden | **ND2**: AF055962[b]<br>**COI**: JN112785<br>**RAG1**: JN112654 |
| *A. marcanoi* | JBL 274<br>JBL 455 | [Not given]<br>Dominican Republic, Peravia, between Baní and El Recodo | **ND2**: AF055955[b]<br>**COI**: JN112786<br>**RAG1**: JN112655 |
| *A. occultus* | MVZ 215144<br>USNM 321891 | Puerto Rico, Caribbean National Forest (El Yunque), 13.3 km S of Palmer on road 191<br>Puerto Rico, Humacao, Caribbean National Forest,<br>Sierra de Luquillo, 13.3 km S of Palmer (=Mameyes) | **ND2**: AF055976[b]<br>**COI**: JN112787<br>**RAG1**: JN112656 |
| *A. sagrei* | USNM 498107<br>MVZ 217371 | Cuba, La Habana, La Habana (Ciudad)<br>United Kingdom, Cayman Islands, Little Cayman, McCoy's Lodge | **ND2**: AF337778[f]<br>**RAG1**: JN112657 |
| *A. smaragdinus* | USNM 549537 | The Bahamas, South Bimini Island, vicinity of airport | **ND2**: JN112718<br>**COI**: JN112788<br>**RAG1**: JN112658 |
| *Polychrus marmoratus* | SNOMNH 36693 | Brazil, Pará, approx. 101 km S and 18 km E Santarem, Agropecuaria Treviso LTDA | **ND2**: AF528738[g]<br>**COI**: JN112789<br>**RAG1**: JN112659 |
| *Pristidactylus scapulatus* | PT 4810 | Argentina, Río Negro, 2 km S Esperanza | **ND2**: AF528732[g]<br>**COI**: JN112790<br>**RAG1**: JN112660 |
| *Urostrophus gallardoi* | FBC 0036 | Argentina, Córdoba, aprox. 2 km S L. V. Marsilla | **ND2**: AF528735[g]<br>**COI**: JN112791<br>**RAG1**: JN112661 |

[a] J.B. Losos (personal communication).
[b] Jackman et al. (1999).
[c] Creer et al. (2001).
[d] Nicholson et al. (2005).
[e] Harmon et al. (2003).
[f] Glor et al. (2001) direct submission.
[g] Schulte et al. (2003).

**Table B1**
List of primers used in amplification and sequencing reactions. The 'f' and 'r' in the primer name indicate the read direction of the primer, forward (the primer aligns at the 5′ end of the gene) or reverse (the primer aligns at the 3′ end of the gene), respectively.

| Primer name | Gene | Primer sequence 5′–3′ | Reference |
|---|---|---|---|
| ILEf.6 | tRNAIle | AAGGGNTACTTTGATAGAGT | Schulte et al. (2003) |
| METf.6 | tRNAMet | AAGCTTTCGGGCCCATACC | Macey et al. (1997) |
| ND2f.5 | ND2 | AACCAAACCCAACTACGAAAAAT | Macey et al. (1997) |
| ND2r.6 | ND2 | ATTTTTCGTAGTTGGGTTTGRTT | Macey et al. (1997) |
| ND2f.14 | ND2 | TGACAAAAACTAGCCCC | Schulte et al. (1998) |
| ND2f.15 | ND2 | TGACAAAAACTAGCACC | Macey et al. (1997) |
| ND2r.26 | ND2 | GATGAGTATGCTATTARTTTTCG | Townsend and Larson (2002) |
| ND2f.48 | ND2 | CCTTGYATAACTTCTGGNAGTCA | J.A. Schulte (pers. comm.) |
| TRPf.12 | tRNATrp | AACCAAGRGCCTTCAAAG | Schulte et al. (2003) |
| ALAf.9 | tRNAAla | CATCAYCTGAATGCAACYCAG | J.A. Schulte (pers. comm.) |
| ASNr.2 | tRNAAsn | TTGGGTGTTTAGCTGTTAA | Macey et al. (1997) |
| ASNr.9 | tRNAAsn | TTGGGRAGTTAGCTGTTAA | This study |
| CO1r.1 | COI | AGRGTGCCAATGTCTTTGTGRTT | Macey et al. (1997) |
| CO1r.8 | COI | GCTATGTCTGGGGCTCCAATTAT | Weisrock et al., 2001 |
| COIr.10 | COI | TCTTYGGTGCCTGRGCYGGAATAGT | J.A. Schulte (pers. comm.) |
| REPTBCf | COI | TCAACAAACCAYAAAGAYATYGG | L. Weigt (pers. comm.) |
| REPTBCr | COI | TAAACTTCAGGGTGGCCRAARAATCA | L. Weigt (pers. comm.) |
| LCO1490 | COI | GGTCAACAAATCATAAAGATATTGG | Folmer et al. (1994) |
| HCO2198 | COI | TAAACTTCAGGGTGACCAAAAAATCA | Folmer et al. (1994) |
| R13f | RAG1 | TCTGAATGGAAATTCAAGCTGTT | Groth and Barrowclough (1999) |
| R27f | RAG1 | CTAYCCTTGYTTTCCTACNGA | Townsend et al. (2004) |
| R28r | RAG1 | GACTGCCTSGCATTCATTTTYC | Townsend et al. (2004) |
| R57r | RAG1 | CGTCTGAAAAGTTTGTTCCCA | J.A. Schulte (pers. comm.) |

**Table B1** (*continued*)

| Primer name | Gene | Primer sequence 5′–3′ | Reference |
|---|---|---|---|
| JRAG1f.2 | RAG1 | CAAAGTRAGATCACTTGAGAAGC | Schulte and Cartwright (2009) |
| JRAG1r.3 | RAG1 | ACTTGYAGCTTGAGTTCTCTTAGRCG | Schulte and Cartwright (2009) |
| JRAG1f.5 | RAG1 | TATGTATGCATAAACAAAGGTGG | J.A. Schulte (pers. comm.) |
| JRAG1r.6 | RAG1 | GCTTTAAGACATCTTCCATTTCATAG | Schulte and Cartwright (2009) |
| JRAG1r.8 | RAG1 | GACTCATTTCCCTCACTTGCCCAAG | Schulte and Cartwright (2009) |
| JRAG1f.9 | RAG1 | CAAAGTTTTGTCACCANTGTTGG | Schulte and Cartwright (2009) |
| JRAG1r.10 | RAG1 | GAATTGCTTAATTTCTTTTGAA | Schulte and Cartwright (2009) |
| JRAG1f.11 | RAG1 | TCAATCTCTTGCCAGATCTGTGAGC | Schulte and Cartwright (2009) |
| JRAG1f.12 | RAG1 | GAGTGGAAACCACCCTTGAAAAATG | Schulte and Cartwright (2009) |
| JRAG1r.13 | RAG1 | CATTTTTCAAGGGTGGTTTCCACTC | Schulte and Cartwright (2009) |
| JRAG1f.14 | RAG1 | CTTGATATGGCTGGAATCCCAAG | Schulte and Cartwright (2009) |
| JRAG1f.15 | RAG1 | ATGAATGGGAATTTTGCCAGAARGCT | Schulte and Cartwright (2009) |
| JRAG1r.16 | RAG1 | GGTTTCATCTTMAGGTAAAGGTCCATGAG | Schulte and Cartwright (2009) |
| JRAG1r.24 | RAG1 | TCTTTCTCTTCACTCCAYGG | Schulte and Cartwright (2009) |
| JRAG1f.25 | RAG1 | CARGAGGAGGTCTGTTTGGG | Schulte and Cartwright (2009) |
| JRAG1r.26 | RAG1 | GACTCCTTGTGATGGAATGAAG | Schulte and Cartwright (2009) |

## References


Alfaro, M.E., Zoller, S., Lutzoni, F., 2003. Bayes or bootstrap? A simulation study comparing the performance of Bayesian Markov Chain Monte Carlo sampling and bootstrapping in assessing phylogenetic confidence. Mol. Biol. Evol. 20, 255–266.

Ayala, S.C., Harris, D., Williams, E.E., 1983. New or problematic *Anolis* from Colombia. I. *Anolis calimae*, new species, from the cloud forest of western Colombia. Breviora 475, 1–11.

Brandley, M.C., de Queiroz, K., 2004. Phylogeny, ecomorphological evolution, and historical biogeography of the *Anolis cristatellus* group. Herpetol. Monogr. 18, 90–126.

Burnell, K.L., Hedges, S.B., 1990. Relationships of West Indian *Anolis* (Sauria: Iguanidae): An approach using slow-evolving protein loci. Caribb. J. Sci. 26, 7–30.

Burnham, K.P., Anderson, D.R., 2004. Multimodel inference: understanding AIC and BIC in model selection. Sociol. Methods Res. 33, 261–304.

Cannatella, D., de Queiroz, K., 1989. Phylogenetic systematics of the anoles: is a new taxonomy warranted? Syst. Zool. 38, 57–69.

Creer, D.A., de Queiroz, K., Jackman, T.R., Losos, J.B., Larson, A., 2001. Systematics of the *Anolis roquet* series of the southern Lesser Antilles. J. Herpetol. 35, 428–441.

de Queiroz, K., Reeder, T., 2008. Squamata—Lizards. In: Crother, B.I. (Ed.), Scientific and Standard English Names of Amphibians and Reptiles of North America North of Mexico, with Comments Regarding Confidence in Our Understanding. Society for the Study of Amphibians and Reptiles. Herp. Circular, vol. 37, pp. 24–45.

Dunn, E.R., 1937. The giant mainland anoles. Proc. New Engl. Zool. Cl. 16, 5–9.

Etheridge, R., 1959. The Relationships of the Anoles (Reptilia: Sauria: Iguanidae): An Interpretation Based on Skeletal Morphology. PhD Thesis, University of Michigan, Ann Harbor, Michigan.

Felsenstein, J., 1985. Confidence limits on phylogenies: an approach using the bootstrap. Evolution 39, 783–791.

Fiedler, P.C., 2002. The annual cycle and biological effects of the Costa Rica Dome. Deep Sea Res. Part I 49, 321–338.

Folmer, O., Black, M., Hoeh, W., Lutz, R., Vrijenhoek, R., 1994. DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. Mol. Mar. Biol. Biotechnol. 3, 294–299.

Giannasi, N., Thorpe, R.S., Malhotra, A., 2000. A phylogenetic analysis of body size evolution in the *Anolis roquet* group (Sauria: Iguanidae): character displacement or size assortment. Mol. Ecol. 9, 193–202.

Glor, R.E., Vitt, L.J., Larson, A., 2001. A molecular phylogenetic analysis of diversification in Amazonian *Anolis* lizards. Mol. Ecol. 10, 2661–2668.

Glor, R.E., Kolbe, J.J., Powell, R., Larson, A., Losos, J.B., 2003. Phylogenetic analysis of ecological and morphological diversification in Hispaniolan trunk-ground anoles (*Anolis cybotes* group). Evolution 57, 2383–2397.

Gorman, G.C., Atkins, L., 1967. The relationships of the *Anolis* of the *roquet* species group (Sauria: Iguanidae). II. Comparative chromosome cytology. Syst. Zool. 16, 137–143.

Gorman, G.C., Dessauer, H.C., 1965. Hemoglobin and transferring electrophoresis and relationships of island populations of *Anolis* lizards. Science 150, 1454–1455.

Gorman, G.C., Dessauer, H.C., 1966. The relationships of *Anolis* of the *roquet* species group (Sauria: Iguanidae). I. Electrophoretic comparison of blood proteins. Comp. Biochem. Physiol. 19, 845–853.

Gorman, G.C., Kim, Y.J., 1976. *Anolis* lizards of the eastern Caribbean: a case study in evolution. II. Genetic relationships and genetic variation of the *bimaculatus* group. Syst. Zool. 20, 167–185.

Gorman, G.C., Thomas, R., Atkins, L., 1968. Intra- and interspecific chromosome variation in *Anolis cristatellus* and its closest relatives. Breviora 293, 1–12.

Gorman, G.C., Licht, P., Dessauer, H.C., Boos, J.O., 1971. Reproductive failure among the hybridizing *Anolis* lizards of Trinidad. Syst. Zool. 20, 1–18.

Gorman, G.C., Buth, D.G., Soulé, M., Yang, S.Y., 1980. The relationships of the *Anolis cristatellus* group: electrophoretic analysis. J. Herpetol. 14, 269–278.

Gorman, G.C., Buth, D., Soulé, M., Yang, S.Y., 1983. The relationships of the Puerto Rican *Anolis*: electrophoretic and karyotypic studies. In: Rhodin, A.G.J., Miyata, K. (Eds.), Advances in Herpetology and Evolutionary Biology. Museum of Comparative Zoology, Cambridge, Massachusetts, pp. 626–642.

Graham, J.B., 1975. The Biological Investigation of Malpelo Island, Colombia. Smithsonian Institution Press, Washington, District of Columbia.

Groth, J.G., Barrowclough, G.F., 1999. Basal divergences in birds and the phylogenetic utility of the nuclear RAG-1 gene. Mol. Phylogenet. Evol. 12, 115–123.

Guyer, C., Savage, J.M., 1986. Cladistic relationships among anoles (Sauria: Iguanidae). Syst. Zool. 35, 509–531.

Harmon, L.J., Schulte II, J.A., Larson, A., Losos, J.B., 2003. Tempo and mode of evolutionary radiation in iguanian lizards. Science 301, 961–964.

Huelsenbeck, J.P., Ronquist, F., Nielsen, R., Bollback, J.P., 2001. Bayesian inference of phylogeny and its impact on evolutionary biology. Science 294, 2310–2314.

Jackman, T.R., Larson, A., de Queiroz, K., Losos, J.B., 1999. Phylogenetic relationships and tempo of early diversification in *Anolis* lizards. Syst. Biol. 48, 254–285.

Jackman, T.R., Irschick, D.J., de Queiroz, K., Losos, J.B., Larson, A., 2002. Molecular phylogenetic perspective on evolution of lizards of the *Anolis grahami* series. J. Exp. Zool. B Mol. Dev. Evol. 294, 1–16.

Kass, R.E., Raftery, A.E., 1995. Bayes factors. J. Am. Stat. Assoc. 90, 773–795.

Kumazawa, Y., Nishida, M., 1993. Sequence evolution of mitochondrial tRNA genes and deep-branch animal phylogenetics. J. Mol. Evol. 37, 380–398.

Larget, B., Simon, D.L., 1999. Markov chain Monte Carlo algorithms for the Bayesian analysis of phylogenetic trees. Mol. Biol. Evol. 16, 750–759.

Lazell Jr., J.D., 1969. The genus *Phenacosaurus* (Sauria: Iguanidae). Breviora 325, 1–24.

Lazell Jr., J.D., 1972. The anoles (Sauria, Iguanidae) of the Lesser Antilles. Bull. Mus. Comp. Zool. Harvard Univ. 143, 1–115.

Lewis, P.O., Holder, M.T., Holsinger, K.E., 2005. Polytomies and Bayesian phylogenetic inference. Syst. Biol. 54, 241–253.

Macey, J.R., Larson, A., Ananjeva, N.B., Fang, Z., Papenfuss, T.J., 1997. Two novel gene orders and the role of light-strand replication in rearrangement of the vertebrate mitochondrial genome. Mol. Biol. Evol. 14, 91–104.

Maddison, D.R., Maddison, W.P., 2001. MacClade: Analysis of Phylogeny and Character Evolution. Version 4.03. Sinauer Associates, Sunderland, Massachusetts.

Melville, J., Hale, J.M., 2009. Length variation in the N-terminal domain of the recombination-activating gene 1 (RAG1) across squamates. Mol. Phylogenet. Evol. 52, 898–903.

Miyata, T., Hayashida, H., Kikuno, R., Hasegawa, M., Kobayashi, M., Koike, K., 1982. Molecular clock of silent substitution: at least six-fold preponderance of silent changes in mitochondrial genes over those in nuclear genes. J. Mol. Evol. 19, 28–35.

Myers, C.W., Donnelly, M.A., 1996. A new herpetofauna from Cerro Yaví, Venezuela: first results of the Robert G. Goelet American Museum-TERRAMAR expedition to the northwestern Tepuis. Am. Mus. Novit. 3172, 1–56.

Myers, C.W., Williams, E.E., McDiarmid, R.W., 1993. A new anoline lizard (*Phenacosaurus*) from the highland of Cerro de la Neblina, southern Venezuela. Am. Mus. Novit. 3070, 1–15.

Newton, M.A., Raftery, A.E., 1994. Approximate Bayesian inference with the weighted likelihood bootstrap. J. Roy. Stat. Soc. Series B Stat. Methodol. 56, 3–48.

Nicholson, K.E., 2002. Phylogenetic analysis and a test of the current infrageneric classification of *Norops* (Beta *Anolis*). Herpetol. Monogr. 16, 93–120.

Nicholson, K.E., Glor, R.E., Kolbe, J.J., Larson, A., Hedges, S.B., Losos, J.B., 2005. Mainland colonization by island lizards. J. Biogeogr. 32, 929–938.

Pagel, M., Meade, A., 2005. Mixture models in phylogenetic inference. In: Gascuel, O. (Ed.), Mathematics of Evolution and Phylogeny. Oxford University Press, Oxford, pp. 121–139.

Peters, J.A., Donoso-Barros, R., 1970. Catalogue of the Neotropical Squamata: Part II. Lizards and amphisbaenians. US Nat. Mus. Bull. 297, 1–293.
Pinto, G., Mahler, D.L., Harmon, L.J., Losos, J.B., 2008. Testing the island effect in adaptive radiation: rates and patterns of morphological diversification in Caribbean and mainland *Anolis* lizards. Proc. Roy. Soc. Lond., B 275, 2749–2757.
Poe, S., 1998. Skull characters and the cladistic relationships of the Hispaniolan dwarf twig *Anolis*. Herpetol. Monogr. 12, 192–236.
Poe, S., 2004. Phylogeny of anoles. Herpetol. Monogr. 18, 37–89.
Poe, S., Yañez-Miranda, C., 2007. A new species of phenacosaur *Anolis* from Peru. Herpetologica 63, 219–223.
Poe, S., Latella, I.M., Ryan, M.J., Schaad, E.W., 2009. A new species of *Anolis* lizard (Squamata, Iguania) from Panama. Phyllomedusa 8, 81–87.
Posada, J.L., Crandall, K.A., 1998. Modeltest: testing the model of DNA substitution. Bioinformatics 14, 817–818. Version 3.7 (June 2005).
R Development Core Team, 2010. R: A Language and Environment for Statistical Computing. Version 2.11.1. <http://www.R-project.org>.
Rambaut, A., Drummond, A.J., 2007. Tracer. Version 1.5. <http://beast.bio.ed.ac.uk/Tracer/>.
Ronquist, F., Huelsenbeck, J.P., 2003. MrBayes 3: Bayesian phylogenetic inference under mixed models. Bioinformatics 19, 1572–1574.
Sadofsky, M.J., Hesse, J.E., McBlane, J.F., Gellert, M., 1993. Expression and V(D)J recombination activity of mutated RAG-1 proteins. Nucl. Acids Res. 21, 5644–5650.
Savage, J.M., Guyer, C., 1989. Infrageneric classification and species composition of the anole genera, *Anolis*, *Ctenonotus*, *Dactyloa*, *Norops*, and *Semiurus* (Sauria: Iguanidae). Amphibia-Reptilia 10, 105–116.
Savage, J.M., Talbot, J.J., 1978. The giant anoline lizards of Costa Rica and western Panama. Copeia 1978, 480–492.
Schneider, C.J., Losos, J.B., de Queiroz, K., 2001. Evolutionary relationships of the *Anolis bimaculatus* group from the northern Lesser Antilles. J. Herpetol. 35, 1–12.
Schulte II, J.A., Cartwright, E.M., 2009. Phylogenetic relationships among iguanian lizards using alternative partitioning methods and *TSHZ1*: a new phylogenetic marker for reptiles. Mol. Phylogenet. Evol. 50, 391–396.
Schulte II, J.A., Valladares, J.P., Larson, A., 2003. Phylogenetic relationships within Iguanidae inferred using molecular and morphological data and a phylogenetic taxonomy of iguanian lizards. Herpetologica 59, 399–419.
Schwartz, A., Henderson, R.W., 1991. Amphibians and Reptiles of the West Indies: Descriptions, Distributions and Natural History. University of Florida Press, Gainesville, Florida.
Shimodaira, H., 2002. An approximately unbiased test of phylogenetic tree selection. Syst. Biol. 51, 492–508.
Shimodaira, H., Hasegawa, M., 2001. CONSEL: for assessing the confidence of phylogenetic tree selection. Bioinformatics 17, 1246–1247.
Shochat, D., Dessauer, H.C., 1981. Comparative study of albumins of *Anolis* lizards of the Caribbean islands. Comp. Biochem. Physiol. A 68, 67–73.
Suchard, M.A., Weiss, R.E., Sinsheimer, J.S., 2001. Bayesian selection of continuous-time Markov chain evolutionary models. Mol. Biol. Evol. 18, 1001–1013.
Sullivan, J., Abdo, Z., Joyce, P., Swofford, D.L., 2005. Evaluating the performance of a successive-approximations approach to parameter optimization in maximum-likelihood phylogeny estimation. Mol. Biol. Evol. 22, 1386–1392.
Suzuki, Y., Glazko, G.V., Nei, M., 2002. Overcredibility of molecular phylogenies obtained by Bayesian phylogenetics. Proc. Natl. Acad. Sci. 99, 16138–16143.
Sverdrup, H.U., Johnson, M.W., Fleming, R.H., 1942. The Oceans: Their Physics, Chemistry, and General Biology. Prentice Hall, Englewood Cliffs, New Jersey.
Swofford, D.L., 2002. PAUP*. Phylogenetic Analysis Using Parsimony (* and Other Methods), Version 4. Sinauer Associates, Sunderland, Massachusetts.
Swofford, D.L., Olsen, G.J., Waddell, P.J., Hillis, D.M., 1996. Phylogenetic inference. In: Hillis, D.M., Moritz, C., Mable, B. (Eds.), Molecular Systematics, second ed. Sinauer Associates, Sunderland, Massachusetts, pp. 407–514.
Templeton, A.R., 1983. Phylogenetic inference from restriction endonuclease cleavage site maps with particular reference to the evolution of humans and apes. Evolution 37, 221–244.
Thompson, J.D., Gibson, T.J., Plewniak, F., Jeanmougin, F., Higgins, D., 1997. Clustal X windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. Nucl. Acids Res. 24, 4876–4882.
Townsend, T., Larson, A., 2002. Molecular phylogenetics and mitochondrial genomic evolution in the Chamaeleonidae (Reptilia, Squamata). Mol. Phylogenet. Evol. 23, 22–36.
Townsend, T.M., Larson, A., Louis, E., Macey, J.R., 2004. Molecular phylogenetics of Squamata: the position of snakes, amphisbaenians, and dibamids, and the root of the squamate tree. Syst. Biol. 53, 735–757.
Uetz, P., Etzold, T., 1996. The EMBL/EBI Reptile Database. Herpetol. Rev. 27, 174–175. <http://www.reptile-database.org/> (accessed 10.01.11).
Ugueto, G.N., Rivas-Fuenmayor, G., Barros, T., Sánchez-Pacheco, S.J., García-Pérez, J.E., 2007. A revision of the Venezuelan Anoles I: A new *Anolis* species from the Andes of Venezuela with the redescription of *Anolis jacare* Boulenger 1903 (Reptilia: Polychrotidae) and the clarification of the status of *Anolis nigropunctatus* Williams 1974. Zootaxa 1501, 1–30.
Underwood, G., 1959. The anoles of the eastern Caribbean (Sauria, Iguanidae): revisionary notes. Bull. Mus. Comp. Zool. Harvard Univ. 121 (part III), 191–226.
Weisrock, D.W., Macey, J.R., Ugurtas, I.H., Larson, A., Papenfuss, T.J., 2001. Molecular phylogenetics and historical biogeography among salamandrids of the "true" salamander clade: rapid branching of numerous highly divergent lineages in *Mertensiella luschani* associated with the rise of Anatolia. Mol. Phylogenet. Evol. 18, 434–448.
Williams, E.E., 1976a. West Indian anoles: a taxonomic and evolutionary summary. 1. Introduction and a species list. Breviora 440, 1–21.
Williams, E.E., 1976b. South American anoles: the species groups. Pap. Avulsos Zool. 29, 259–268.
Williams, E.E., 1979. South American anoles: the species groups. 2. The proboscis anoles (*Anolis laevis* group). Breviora 449, 1–19.
Williams, E.E., 1984. New or problematic *Anolis* from Colombia. III. Two new semiaquatic anoles from Antioquia and Chocó, Colombia. Breviora 478, 1–22.
Williams, E.E., 1988. New or problematic *Anolis* from Colombia. V. *Anolis danieli*, a new species of the *latifrons* species group and a reassessment of *Anolis apollinaris* Boulenger, 1919. Breviora 489, 1–25.
Williams, E.E., 1989. A critique of Guyer and Savage (1986): cladistic relationships among anoles (Sauria: Iguanidae): are the data available to reclassify anoles? In: Woods, C.A. (Ed.), Biogeography of the West Indies: Past, Present and Future. Sandhill Crane Press, Gainsville, Florida, pp. 434–478.
Williams, E.E., Duellman, W.E., 1967. *Anolis chocorum*, a new *punctatus*-like anole from Darién, Panamá (Sauria, Iguanidae). Breviora 256, 1–12.
Williams, E.E., Duellman, W.E., 1984. *Anolis fitchi*, a new species of the *Anolis aequatorialis* group from Ecuador and Colombia. In: Siegler, R.A., Hunt, L.E., Knight, J.L., Malaret, L., Zuschlag, N.L. (Eds.), Vertebrate Ecology and Systematics – A Tribute to Henry S. Fitch. Special Publications No. 10. Museum of Natural Museum of Kansas, The University of Kansas, Laurence, Kansas, USA, pp. 257–266.
Williams, E.E., Rand, H., Rand, A.S., O'Hara, R.J., 1995. A computer approach to the comparison and identification of species in difficult taxonomic groups. Breviora 502, 1–47.
Williams, E.E., Praderio, M.J., Gorzula, S., 1996. A phenacosaur from Chimantá Tepui, Venezuela. Breviora 506, 1–15.
Yang, S.Y., Soulé, M., Gorman, G.C., 1974. *Anolis* lizards of the eastern Caribbean: a case study in evolution. I. Genetic relationships, phylogeny, and colonization sequence of the *roquet* group. Syst. Zool. 23, 387–399.
Zwickl, D.J., 2006. Genetic Algorithm Approaches for the Phylogenetic Analysis of Large Biological Sequence Datasets under the Maximum Likelihood Criterion. Ph.D. Dissertation, The University of Texas at Austin, Texas.