Parallel gigantism and complex colonization patterns in the Cape Verde scincid lizards *Mabuya* and *Macroscincus* (Reptilia: Scincidae) revealed by mitochondrial DNA sequences

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The scincid lizards of the Cape Verde islands comprise the extinct endemic giant *Macroscincus coctei* and at least five species of *Mabuya*, one of which, *Mabuya vaillanti*, also had populations with large body size. Phylogenetic analysis based on DNA sequences derived from the mitochondrial cytochrome *b*, cytochrome oxidase I and 12S rRNA genes (711, 498 and 378 base pairs (bp), respectively) corroborates morphological evidence that these species constitute a clade and that *Macroscincus* is unrelated to very large skinks in other areas. The relationships are ((*M. vaillanti* and *Mabuya delalandii*) (*Mabuya spinalis* and *Macroscincus coctei* (*Mabuya fogoensis nicolauensis* (*Mabuya fogoensis antaoensis* and *Mabuya stangeri*)))). The Cape Verde archipelago was colonized from West Africa, probably in the Late Miocene or Early Pliocene period. The north-eastern islands were probably occupied first, after which the ancestor of *M. vaillanti* and *M. delalandii* may have originated on Boavista, the ancestor of the latter species arriving on Santiago or Fogo later. The *M. fogoensis*–*M. stangeri* clade colonized the islands of Branco, Razo, Santa Luzia and São Vicente from São Nicolau and reached Santo Antão after this. Colonization of these northeastern islands was slow, perhaps because the recipient islands had not developed earlier or because colonization cut across the path of the Canary Current and the Northeast Trade Winds, the main dispersing agents in the region. Rapid extension of range into the southwestern islands occurred later in *M. spinalis* and then in *M. vaillanti* and *M. delalandii*. The long apparent delay between the origin of these species and their southwestern dispersal may have been because there were earlier colonizations of the southern islands which excluded later ones until the earlier inhabitants were exterminated by volcanic or climatic events. The evolution of large size in *Macroscincus* occurred in the northwestern islands and was paralleled in the eastern and southern islands by populations of *M. vaillanti*. Both cases of size increase in Cape Verde skinks were accompanied by the development of herbivory.

Keywords: mitochondrial DNA; phylogeny; conservation; colonization; radiation; ancient DNA

1. INTRODUCTION

The scincid lizards of the Cape Verde islands (figure 1) are assigned to two genera: *Mabuya* Fitzinger 1826, which is also found in southern Asia, Africa and the Neotropics, and the endemic *Macroscincus* Bocage 1873. *Macroscincus* comprises a single species, *Macroscincus coctei* (Duméril & Bibron 1839), which reached at least 320 mm from snout to vent (Greer 1976) and a total length of *ca.* 600 mm (Andreone & Gavetti 1998; Adreone 2000), making it one of the largest scincids. Since its description, *Macroscincus* has only been certainly known from the small northern Cape Verde islands of Razo and Branco (figure 1), but recently discovered subfossils show that it also once occurred on neighbouring São Vicente and Santa Luzia (López-Jurado *et al.* 2001). *Macroscincus* appears to have become extinct by *ca.* 1900, with the last wild-living animals formally reported being seen by Alexander (1898) and Fea (1898).

On present evidence *Mabuya* has five living species in the archipelago: *Mabuya delalandii* (Dúmeril & Bibron 1839), *Mabuya vaillanti* Boulenger 1887, *Mabuya spinalis*

Boulenger 1906, *Mabuya stangeri* (Gray 1845) and *Mabuya fogoensis* (O'Shaugnessy 1874). Subfossil *Mabuya* recently found on Maio and Boavista are apparently conspecific with *M. vaillanti* from more southwestern islands but much larger, with the Maio population growing to an estimated 240 mm from snout to vent (López-Jurado *et al.* 2001). The types of *M. fogoensis*, which were collected in 1865 or earlier, were believed to come from Fogo and São Vicente, but no further specimens appear to have been collected from these islands, the species only being known from populations on São Nicolau and Santo Antão, which are presently named *M. f. nicolauensis* and *M. f. antaoensis*. *Mabuya geisthardti* Joger 1993 is conspecific with the latter form (S. Carranza and E. N. Arnold, personal observation). The taxonomy of Cape Verde skinks is discussed further elsewhere (Carranza *et al.* 2001).

Macroscincus has been considered as being related to other very large skinks, for example *Tiliqua* of Australasia (Bocage 1873) and the extinct *Didosaurus mauritianus* Günther 1877 of Mauritius (Hoffstetter 1949) (*Didosaurus* is now referred to *Leiolopisma*, see Arnold 1980). An alternative, better-substantiated hypothesis is that *Macroscincus* is most closely related to the endemic *Mabuya* of the Cape Verde islands with which it is

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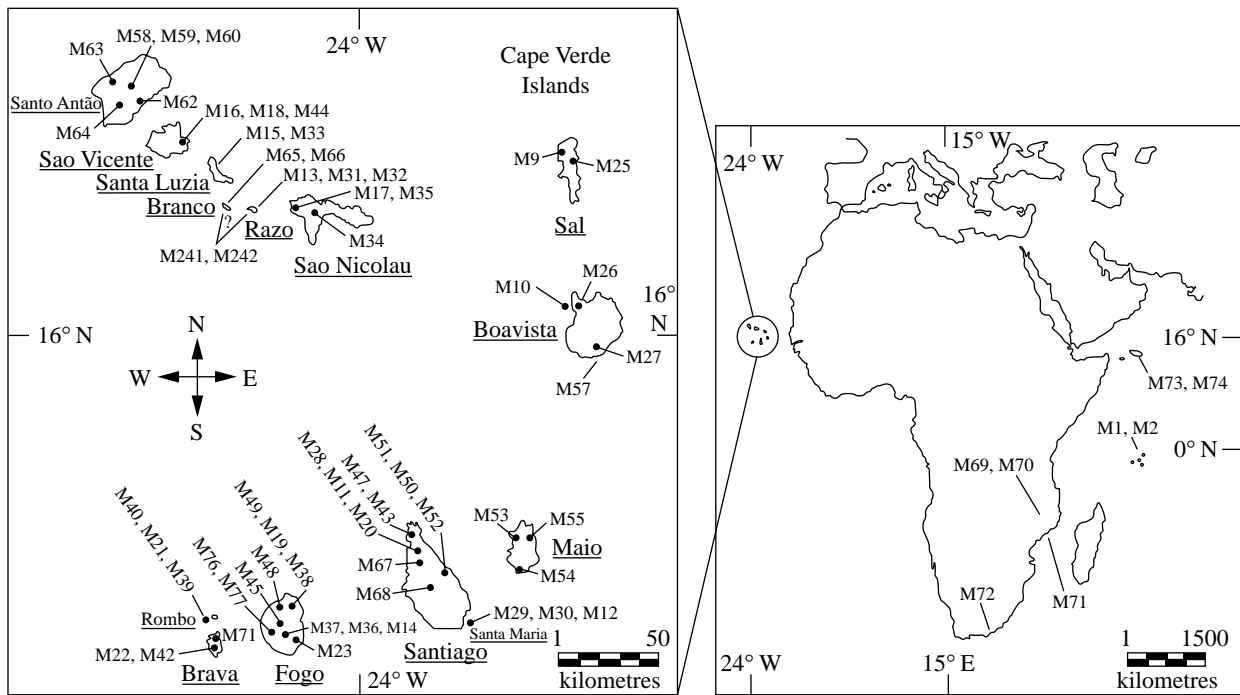
1595

Figure 1. Cape Verde islands and Africa showing the localities of *Mabuya* and *Macroscincus* (M) samples used in the present study. See electronic Appendix A and figure 3 for further details.

believed to constitute a monophyletic radiation (Greer 1976). The case for the Cape Verde skinks forming a clade is based on morphological features that are rare or absent in *Mabuya* outside the archipelago and are very probably derived rather than primitive. These include an anterior medial process on the ectopterygoid bone (which is absent in *M. delalandii* and paralleled in East African *Mabuya irregularis*) (Greer 1976), strong ossification of the palpebral bone (E. N. Arnold, personal observation), anterior autotomic caudal vertebrae with transverse processes that are clearly directed anteriorly (a feature not or only weakly developed in some other *Mabuya*) (E. N. Arnold, personal observation) and body scales, which are often numerous (45–112 around the mid-body except in *M. spinalis* and *M. stangeri*). The relationship between *M. delalandii* and *M. vaillanti* is supported by fusion of the paired frontoparietal scales and of the interparietal and paired parietal scales. The remaining Cape Verde species are characterized by their members nearly always possessing 27 pre-sacral vertebrae instead of the 26 that are usual in *Mabuya* (Greer *et al.* 2000).

Here we use mitochondrial DNA for testing the competing hypotheses about the affinities of *M. coctei* and exploring the relationships among the Cape Verde skinks. Three genes (cytochrome *b*, cytochrome oxidase I and 12S rRNA) were investigated from 16 outgroup taxa and all recognized Cape Verde taxa including individuals from all island populations.

2. MATERIAL AND METHODS

(a) *Samples and DNA extraction*

A total of 75 skinks were used in this study. Their data are given in electronic Appendix A (available on The Royal Society web site) and their localities are given in figure 1. The Genbank

accession numbers for the mitochondrial DNA sequences (which are also given in electronic Appendix A) are all consecutive and span from AF280114 to AF280333.

For most species, total genomic DNA was extracted from $2–3 \, mm^3$ of tail tissue following standard protocols described elsewhere (Harris *et al.* 1998; Carranza *et al.* 2000). The primers used in both amplification and sequencing were cytochrome b_1, cytochrome b_2 (Kocher *et al.* 1989), the forward primer of cytochrome b_2 and CB3-3′ (Palumbi 1996) for the cytochrome *b* gene, COIf-5′ and COIa-3′ for the cytochrome oxidase I gene (Palumbi 1996), and 12Sa and 12Sb for the 12S rRNA gene (Kocher *et al.* 1989). Thermocycling consisted of an initial 90 s at 94 °C followed by 35 cycles of 30 s at 94 °C, 45 s at 45 °C and 1 min at 72 °C and then a single cycle of 10 min at 72 °C. Amplified bands were cut out and purified using a silica-based method (Boyle & Lew 1995). Direct sequencing of the PCR (polymerase chain reaction) products was carried out using an ABI 377 automated sequencer (PE Biosystems, Warrington, UK).

Macroscincus coctei DNA was extracted from specimens in methylated alcohol that were collected before 1891. In order to avoid contamination, both DNA extraction and PCR amplification were carried out in isolation using specific techniques described elsewhere (Carranza *et al.* 1999). Because the *Macroscincus* DNA was degraded, additional internal primers were necessary for amplification of the first part of the cytochrome *b* gene (307 base pair (bp) segment delimited by the primers cytochrome b_1–cytochrome b_2); these were CB107F and CB144R (Carranza *et al.* 1999).

(b) *Phylogenetic analysis*

DNA sequences were aligned by hand using the alignment editor GDE (Smith *et al.* 1994) and with reference to the published secondary structure for 12S rRNA (Hickson *et al.* 1996). Alignment gaps were inserted in order to resolve length differences between sequences and positions that could not be
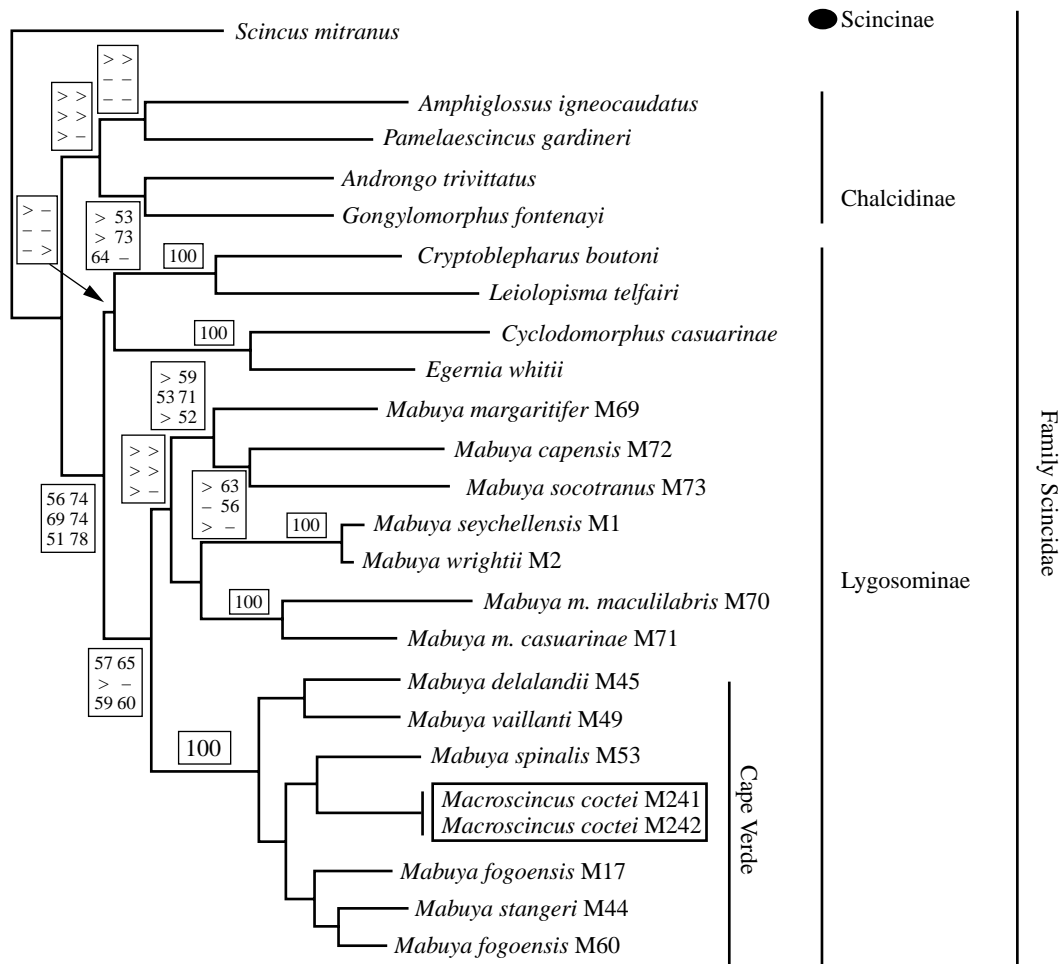
Figure 2. Maximum-likelihood tree (log likelihood −11 326.94, GTR model of sequence evolution and cytochrome *b* third codon positions included) for some representatives of the family Scincidae. Bootstrap support for particular nodes is shown in the boxes with the figures indicating the percentage support for different analyses. Upper left, bootstrap support derived by maximum likelihood (GTR) and including the cytochrome *b* third codon positions. Upper right, bootstrap support derived by maximum likelihood (GTR) and excluding the cytochrome *b* third codon positions. Middle left, bootstrap support derived by minimum evolution (GTR + I + G) and including the cytochrome *b* third codon positions. Middle right, bootstrap support derived by minimum evolution (GTR + I + G) and excluding the cytochrome *b* third codon positions. Lower left, bootstrap support derived by maximum parsimony (cytochrome *b* third codon Ts = 0 and all the rest Ts = Tv). Lower right, bootstrap support derived by maximum parsimony (cytochrome *b* third codon Ts = 0 and all the rest Ts = 1 and Tv = 4). When the difference between the four bootstrap support figures was < 5% only the average value is shown. The ‘ > ’ symbol is used to show that the bootstrap support for that node is lower than 50% and the ‘—’ symbol indicates that a particular node is never recovered when using this method. The codes (M1, M2, etc.) indicate the samples used, details of which can be found in electronic Appendix A and their localities in figure 1.

unambiguously aligned were excluded. No ambiguous alignments were found for cytochrome oxidase I and cytochrome *b*, and no gaps were postulated. All the cytochrome oxidase I and cytochrome *b* sequences were translated into amino acids using the vertebrate mitochondrial code and no stop codons were detected, suggesting that all the sequences analysed were functional.

Given that the various phylogenetic methods available often involve different assumptions about models of evolutionary change, the similarity of the phylogenies produced by different methods increases confidence that their topologies are representative of the evolutionary history of the genes involved. Therefore, three methods were used in our phylogenetic analysis and the results compared. These were maximum parsimony, distances (minimum evolution and neighbour joining) and maximum likelihood. All analyses were performed using PAUP* (Swofford 1998) except where stated. Modeltest v. 3.0 (Posada & Crandall 1998) was used to select the most appropriate model of sequence

evolution for the distance and maximum-likelihood analyses. The shape of the gamma distribution and the number of invariable sites could not be incorporated in the maximum-likelihood analyses of figure 2 due to the computational time required. The maximum-parsimony and minimum-evolution analyses from figure 2 were heuristic searches involving tree bisection and reconnection (TBR) branch swapping with 1000 random stepwise additions of taxa. Gaps were considered as a fifth state in the maximum-parsimony analyses.

The maximum-parsimony analyses for the tree in figure 3 used a strategy for avoiding unnecessary swapping involving replicates that do not locate one of the ‘islands’ containing optimal trees (Giribet & Wheeler 1999). This strategy involved maxtrees = 10 000, a heuristic search with TBR branch swapping, specifying not to store more than ten trees of length 1 and then inactivating this constraint and swapping on all the stored trees to completion.
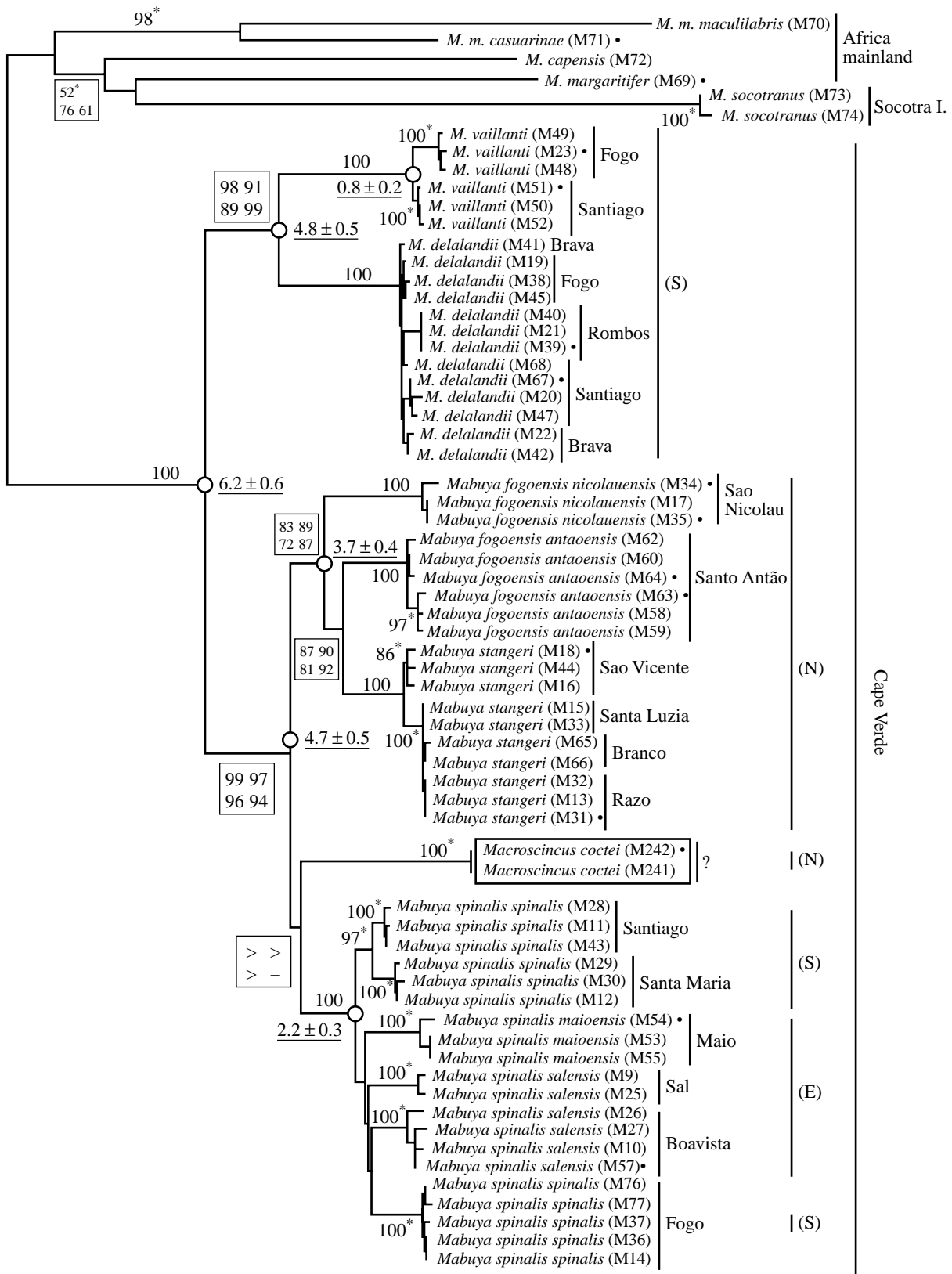
Figure 3. Neighbour-joining tree under the GTR + I + G model of sequence evolution for 58 *Mabuya* and two *Macroscincus* individuals from Cape Verde plus six other *Mabuya* used as outgroups. Cytochrome *b* and cytochrome oxidase I third codon positions were included in the analysis. Bootstrap support for particular nodes is shown in boxes with the figures indicating the percentage support for different analyses. Upper left, bootstrap support derived by neighbour joining (GTR + I + G). Upper right, bootstrap support derived by maximum likelihood (GTR + I + G). Lower left, bootstrap support derived by maximum parsimony (Ts = Tv). Lower right, bootstrap support derived by maximum parsimony (Ts = 1 and Tv = 6). When the difference between the four bootstrap support figures was less than 5% only the average value is shown. The ' > ' symbol is used to show that the bootstrap support for that node is lower than 50%, the '—' symbol indicates that a particular node is never recovered when using this method and '*' indicates that the maximum-likelihood analysis did not include enough individuals for testing

Table 1. Statistical support for alternative hypotheses on *Macroscincus* phylogenetic relationships.

(SH, Shimodaira–Hasegawa test; KH, Kishino–Hasegawa test. $p < 0.005$ suggests that the constrained and unconstrained solutions are significantly different.)

			p-value	
tree	$-\log$ likelihood	$\Delta - \log$ likelihood	SH test	KH test
unconstrained tree (tree from figure 2)	11 326.94	best	—	—
Macroscincus sister to *Egernia* plus *Cyclodomorphus*	11 430.80	103.86	< 0.005	< 0.005
Macroscincus sister to *Leiolopisma*	11 431.21	104.27	< 0.005	< 0.005

The robustness of the inferred maximum-parsimony, minimum-evolution, neighbour-joining and maximum-likelihood trees was tested by bootstrapping (Felsenstein 1985), which in all cases involved 1000 pseudo-replications. Molecular clock assumptions were incorporated in order to assess the age of the speciation events. The likelihood ratio test (Huelsenbeck & Crandall 1997) was used for testing the statistical significance of the difference between the log likelihood of the trees calculated with and without the clock assumptions. The incongruence length difference (ILD) test (Mickevich & Farris 1981; Farris *et al.* 1994) was used to check for incongruence between the genes used. In this test, 10 000 heuristic searches were made and invariable characters were always removed before starting the analysis (Cunningham 1997). Topological constraints were generated using MacClade (Maddison & Maddison 1992) and compared to our optimal topologies using Kishino–Hasegawa (Kishino & Hasegawa 1989) and Shimodaira–Hasegawa (Shimodaira & Hasegawa 1999) tests with full optimization and 1000 bootstrap replicates.

3. RESULTS

(a) *Is* Macroscincus *a giant Cape Verde* Mabuya*?*

Macroscincus was included in a phylogenetic analysis with 22 other skink taxa comprising members of all three main groupings of the Scincidae and including the following possible relatives: *Cyclodomorphus casuarinae* and *Egernia whitii* (which are closely related to *Tiliqua*), *Leiolopisma telfairi* (which is closely related to *Leiolopisma mauritiana*), African *Mabuya* (seven taxa) and Cape Verde *Mabuya* (six taxa). A total of 1089 bp (711 bp of the cytochrome *b* and 378 bp of the 12S rRNA genes) were used in the analysis. Out of these, 498 bp were variable and 425 parsimony informative. The ILD test showed that the two gene fragments were congruent (ILD $p > 0.73$) and could consequently be combined in a total evidence analysis. As evolutionary relationships may be obscured if sites have become saturated by multiple substitutions (Swofford *et al.* 1996), the 12S rRNA gene and each codon position of the cytochrome *b* gene were independently tested for saturation by plotting the observed proportions of transitions (Ts) and transversions (Tv) against the uncorrected sequence divergence (see electronic Appendix B) (available on The Royal Society Web site). Only the 237 cytochrome *b* third codon Ts showed some degree of

saturation and, therefore, subsequent distance (minimum evolution) and maximum-likelihood analyses were performed both with and without these. When they were eliminated, the numbers of variable and parsimony informative sites in the data set were reduced to 270 and 209, respectively. In the maximum-parsimony analyses, the saturated cytochrome *b* third codon Ts were given a weight of 0. An overall Ts:Tv ratio of 3.5 was estimated for this data set.

Maximum-likelihood, minimum-evolution and maximum-parsimony analysis all produced similar trees to the one shown in figure 2. This was rooted using *Scincus mitranus*, a morphologically primitive taxon (Greer 1970). The tree corroborates the clade status of the Cape Verde skinks (100% bootstrap support), thereby clearly indicating that they are monophyletic with respect to the other taxa in the analysis and that *Macroscincus* is part of the clade. In order to check whether there was a significant difference between the present tree, in which *Macroscincus* is part of the Cape Verde *Mabuya* clade and ones where it is sister to other postulated relatives, the maximum-likelihood tree from figure 2 was compared with trees constrained so that *Macroscincus* was sister to *Egernia* plus *Cyclodomorphus* and in a second analysis to *Leiolopisma*. The results of the Kishino–Hasegawa and Shimodaira–Hasegawa tests (table 1) clearly indicate that the two constrained trees are significantly different from the unconstrained solution, the version of relationships (*Macroscincus* plus all Cape Verde *Mabuya* monophyletic) being consequently preferred.

(b) *Phylogenetic relationships of the endemic skinks of the Cape Verde islands*

In order to investigate the relationships of the species and island populations of Cape Verde skinks further, another analysis was performed including 58 individuals of Cape Verde *Mabuya* and two of *Macroscincus*. Six African *Mabuya* taxa were used as outgroups since these appeared as the closest relatives of the Cape Verde species in the first analysis (figure 2). Seven hundred and eleven base pairs of the cytochrome *b*, 498 bp of the cytochrome oxidase I and 378 bp of the 12S rRNA genes were sequenced for all individuals, the new alignment consisting of 1587 positions, with 580 being variable and 520 parsimony informative. A saturation analysis of Ts

the monophyly of the group. Only the 15 species indicated with a dot were used in the maximum-likelihood analysis and the test of the molecular clock. Other figures at nodes are molecular clock estimates of divergence. Although gross approximations, they give some idea of when events may have occurred and the relative amounts of time between them. N indicates north, S indicates south and E indicates east of Cape Verde archipelago.

and Tv carried out for the 12S rRNA gene and each codon position of the cytochrome *b* and cytochrome oxidase I genes (see electronic Appendix B) gave no indication of saturation within the Cape Verde skinks and, consequently, the third codon positions of cytochrome *b* and cytochrome oxidase I were included in the analysis. An overall Ts:Tv ratio of 5.4 was estimated for this data set. An ILD test showed that all three mitochondrial genes were congruent with each other (ILD, $p > 0.89$) and could therefore be combined in a total evidence analysis.

The results of the phylogenetic analysis are shown in figure 3, with all neighbour-joining, maximum-parsimony and maximum-likelihood trees having the same general topology. Thirteen Cape Verde and two outgroup individuals were selected from the total of 66 for the maximum-likelihood analysis. The results indicate that, with the exception of *M. spinalis spinalis* and *M. spinalis salensis*, all the taxa included in the analysis are monophyletic, thereby corroborating their validity. The same is true of conspecifics from particular islands, except for *M. stangeri* on St Luzia in which the sequence is very similar to that on the neighbouring islets of Razo and Branco. The relationships between the species are congruent with those suggested by morphology (see § 1), the southern *M. delalandii* and *M. vaillanti* forming one unit, which is called here the *M. delalandii* clade and the remaining species forming another, the *M. coctei* clade. *Mabuya fogoensis* and *M. stangeri* constitute a subclade in which *M. stangeri* is more closely related to *M. fogoensis antaoensis* than to *M. f. nicolauensis*, making *M. fogoensis* paraphyletic. The detailed relationship between this unit, *M. spinalis* and *Macroscincus* is essentially unresolved. In order to explore the origin and relationships of the two giant forms of Cape Verdian skinks further (*Macroscincus* and *M. vaillanti*), the log likelihood of the maximum-likelihood tree presented in figure 3 (-6720.473) was compared with the log likelihood of a maximum-likelihood tree constrained so that *M. coctei* and *M. vaillanti* were sister taxa (-6771.325). The results of the Shimodaira–Hasegawa and Kishino–Hasegawa tests showed that the constrained trees are significantly different from the unconstrained solution ($\Delta-$log likelihood $= 50.852$) ($p < 0.05$), the version of relationships presented in figure 3 (where the two giant skinks have smaller forms as the closest relatives) being consequently preferred.

In order to obtain some idea of the times of cladogenic events in the phylogeny we first performed a maximum-likelihood ratio test to see whether the mitochondrial DNA sequences were evolving according to a molecular clock. The cytochrome oxidase I gene was not included in the molecular clock analysis in order to make the results comparable with similar studies in the region (Arnold 2000; Carranza *et al.* 2000). The topology of the resulting maximum-likelihood tree including the cytochrome *b* and 12S rRNA mitochondrial genes (1089 characters in total, with 333 being variable and 236 parsimony informative) is exactly the same as that of the maximum-likelihood tree including all three mitochondrial genes (see figure 3). The resulting log likelihood value (-4548.41939) was compared with the log likelihood of the same tree constructed under molecular clock assumptions (-4557.05382) and showed that there is no significant

difference between the likelihoods of the two trees (likelihood ratio test statistic ($-2\log\Delta$) $= 17.269$ which approximates to a χ^2_{13} distribution under the null hypothesis) ($p > 0.05$). Thus, the sequence can be used for estimating dates. In order to calibrate the clock, a 1.96% genetic divergence (corrected with the Kimura two-parameter model) (Kimura 1980) per million years (Myr) was used based on a previous analysis of lacertid lizards (*Gallotia*) from the Canary Islands in which the likely age of particular island populations was estimated on geophysical grounds (S. Carranza and E. N. Arnold, personal observation). This involved the cytochrome *b* and the 12S rRNA genes only. Dates based on this calibration are shown in figure 3. They are inevitably rough approximations and most useful for giving a conception of the relative amounts of time between different events indicated by branching points on the estimate of phylogeny. The first speciation event that separated the two main clades of Cape Verde skinks occurred *ca.* 6.2 Myr ago, providing evidence that the group has been in the archipelago for at least this long.

4. DISCUSSION

(a) *Historical biogeography*

The Cape Verde islands arose as volcanoes that were separated from the neighbouring African mainland and, in most cases, from each other by deep water (Mitchell-Thomé 1985). São Vicente, Santa Luzia, Razo and Branco are exceptional in being situated on a bank less than 50 m below the surface of the sea (Morris 1989) and so were very probably connected during Pleistocene falls in sea level that could be up to 120 m. This is corroborated by the low levels of mitochondrial DNA differentiation between the populations of *M. stangeri* on these islands and those of the geckos *Tarentola caboverdiana* and *Tarentola gigas* (Carranza *et al.* 2000). The colonization of the individual islands of the Cape Verde archipelago by skinks was consequently almost entirely through transmarine migration, presumably on natural rafts of vegetation. The monophyly of Cape Verde skinks makes it likely that they arose from a single colonizing ancestral species. This almost certainly originated in West Africa, 460 km to the East. *Mabuya* species also occur in the Neotropics, but there is no evidence that the Cape Verde taxa are more closely related to these than to at least some African ones. Colonization from America would also have involved a much longer journey against the Atlantic Ocean currents that run southwest and then westwards towards America (Guppy 1917; Carranza *et al.* 2000).

The prevailing agents of transmarine dispersal in the area, i.e. the Canary Current and Northeast Trade Winds, both run southwestwards from the African coast and through the Cape Verde archipelago, so colonizers are likely to have come from a northeasterly direction. When phylogeny, the number of interisland journeys and transmarine distances covered are taken into account, and journeys substantially against the prevailing winds and currents excluded, the presently known species distribution of Cape Verde skinks is most economically explained as follows. Initial arrival was on one of the northeastern islands of São Nicolau, Sal or Boavista, after which the ancestor of the *M. delalandii* clade originated on

the latter island perhaps around 6 Myr ago. Later, a propagule from here, representing the ancestor of *M. delalandii*, travelled south with the prevailing dispersal agents to Santiago or Fogo. The *M. coctei* clade divided into the progenitor of *M. spinalis* and the ancestor (or ancestors) of *M. coctei* and the *M. fogoensis–M. stangeri* subclade. In the latter, phylogenetic topology and the minimization of distances covered indicate that the colonization sequence was from São Nicolau to São Vicente and associated islands and then to Santo Antão. The ancestor of *M. coctei* moved in the same direction although this species has not yet been found on Santo Antão. *Mabuya spinalis* colonized the eastern and more southern islands at a later date and, since the phylogeny of the various island assemblages is essentially a polychotomy (see figure 3), spread is likely to have been much faster than that of the *M. fogoensis–M. stangeri* clade.

Further colonization events in the *M. delalandii* clade appear to be quite recent. This includes the separation of the populations of *M. vaillanti* on Santiago and Fogo and the spread of *M. delalandii*, which may have been even closer to the present (see figure 3). In fact, the low genetic divergence and the lack of morphological variation between its populations may even indicate that this species has been inadvertently spread over most of its present range by human introduction. This receives some support from the quite recent discoveries of isolated individuals on Maio (S. Carranza and J. Mateo, personal observation) and Boavista (Schleich 1987).

The intervals between the production of the five main units of Cape Verde *Mabuya* by speciation and their subsequent spread to the limits of their present ranges appear relatively long, and this requires explanation. The delays in colonization of the northern islands of São Vicente and associated islands and Santo Antão by the *M. fogoensis–M. stangeri* clade seem substantial, considering the journeys involved are very short (*ca.* 20 km each). As estimates of age are apparently not available for these islands, it is possible that colonization could not take place until they rose above the sea. Alternatively, northwestern spread may have been slow because it cut across the path of the main currents and winds in the area. In this case, colonization may have depended on rare meteorological events. This would parallel the situation in Canary Island lacertid lizards (*Gallotia*), which are estimated to have taken at least 8 Myr to get from the African mainland to the island of La Gomera, a modest dispersal by the standards of many lizards (Arnold 2000) involving four minimum sea crossings of just 100, 90, 70 and 30 km (S. Carranza and E. N. Arnold, personal observation).

Neither of these hypotheses can explain the apparently long delays between the origins of *M. spinalis*, *M vaillanti* and *M. delalandii* (2.5, 4 and > 4 Myr, respectively) and their spread, which was probably in the southwestern direction. The colonized islands have been in place for a long time (Maio from at least the Miocene period, Santiago from 10.3 Myr ago and Brava from 5.5 Myr ago) (Mitchell-Thomé 1985) and dispersal agents act in an appropriate direction for colonization. One possibility is that populations of the same or similar taxa previously colonized these islands, excluding later propagules, but have subsequently become extinct, after which rapid recolonization was possible. Exclusion of lizard species

from islands by similar forms that are already present is known to occur elsewhere (Case & Bolger 1991) and extinction is a real possibility in the southern Cape Verde islands, as there has been extensive volcanic action and the archipelago is a semi-desert area where rainfall may fail for many years at a time (Correia e Silva 1995). A catastrophe of either of these kinds could have exterminated all lizards, after which there could have been rapid reinvasion, accounting for the pattern seen in *M. spinalis*. It is notable that the gecko *Tarentola rudis* seems to have invaded the southern islands from the northwestern ones at about the same time (Carranza *et al.* 2000). Less extensive extinction at a later date may have removed competitors of present *M. vaillanti* and *M. delalandii*, thereby permitting their spread. This explanation based on extinction would be corroborated by the discovery of fossil skinks in the southern islands older than the estimated dates of invasion. Populations precluding colonization would not have to be different species. In none of the ones shown in figure 3 is there clear evidence of more than one invasion of an island from its source area, which suggests occupation prevents significant migration subsequently (as noted by Darwin 1859). Presumably, this is because the island concerned is likely to maintain a large population so that, even if a propagule consisting of one or a few individuals made a successful landing, it would be unlikely to contribute its mitochondrial DNA to the population, at least in periods of ecological stability.

Speciation and dispersal has resulted in many islands having more than a single species. While some, such as Santo Antão, São Nicolau and Brava, have just one, most possess two or, in the case of São Vicente, Razo, Branco, Boavista and Maio, did so in the quite recent past. Santiago and Fogo are exceptional in both having three. On all islands with more than a single species, one of the large partly vegetarian giants is (or was) present alongside one or more smaller mainly insectivorous species. There is no information about how these small forms are differentiated from each other ecologically on three-species islands.

The southern islands of Fogo and Santiago have the highest number of reptile species in the archipelago, even compared with other islands of similar size. This may be because they lie in or close to the path of currents and winds passing through the more northern islands so that they are likely to have received multiple colonizations from these.

(b) *Evolution of the Cape Verde skinks*

The two Cape Verde giant forms (up to 320 mm and an estimated 240 mm from snout to vent, respectively) are far larger than other skinks in the archipelago, as other populations of *M. vaillanti* only reach *ca.* 130 mm and the remaining forms no more than 90 mm. Taking into account the fact that no *Mabuya* outside the Cape Verde archipelago attains the size of its giant forms, the topology of the phylogeny presented in figure 3 and the result of the Kishino–Hasegawa and Shimodaira–Hasegawa tests presented in § 3(b) (a tree constrained so that the hypothesis that the two Cape Verdian giant skinks were sister taxa was rejected), it is most parsimonious to assume that very large size has developed twice independently in Cape Verde skinks, i.e. in the recently

extinct *Macroscincus* population and in some extinct populations of *M. vaillanti*.

Possibly, large size results from character displacement (Brown & Wilson 1956) in both the northern and southern islands, where the giants occurred sympatrically with smaller species. These very big skinks have both become partly herbivorous *in situ* (Greer 1976). In association with this, their dentition has been modified in a similar way to other pleurodont herbivorous lizards, the lateral teeth being linguo-labially compressed with additional cusps in contrast to the more primitive conformation found in non-herbivorous *Mabuya* that feed mainly on arthropods. However, the details of cusping are different, *M. vaillanti* usually having three cusps per tooth while *Macroscincus* may have many more.

Very large size has evolved in other oceanic island lizard groups, for instance twice in Canary Island lacertids (*Gallotia*) (Gonzalez *et al.* 1996) and once each in Mauritian *Leiolopisma* (Arnold 1980), Baja California iguanids (*Sauromalus*) (Petren & Case 1997) and Cape Verde geckos (*Tarentola*) (Carranza *et al.* 2000). Change in dimensions in insular lizards does not always involve gigantism and size can also decrease markedly as it has done in the Mauritian gecko *Nactus coindemerensis* (Arnold 2000; J. Austin and E. N. Arnold, personal observation) and in populations of the Cape Verde *Hemidactylus bouvieri*. Nor is gigantism always associated with the evolution of herbivory. The smaller ancestors of island *Sauromalus* were already plant feeders (Petren & Case 1997) and there is no evidence from tooth shape and tooth wear for herbivory in *L. mauritiana* (Arnold 2000).

Although evolutionary outcomes in Cape Verde skinks and sympatric *Tarentola* geckos are similar in that both groups show substantial radiation, species sympatry and marked size increase in some forms, their histories are significantly different. As in the skinks, large *Tarentola* increased their size in sympatry with smaller congenerics in the northwestern Cape Verde islands. However, in the more southern ones, where large *Mabuya* evolved independently from those in the northwestern islands, big *Tarentola* may well have arrived fully formed, invading from the north after their size increase there (Carranza *et al.* 2000).

5. CONCLUDING REMARKS

Mitochondrial DNA sequences corroborate the hypothesis of Greer (1976) that *Macroscincus* is closely related to the endemic *Mabuya* species of the Cape Verde islands. They also enable the phylogeny and history of the Cape Verde skinks to be reconstructed and their taxonomy revised (Carranza *et al.* 2001*a*). Together with the other studies of the reptile fauna of the archipelago (Carranza *et al.* 2000) it is now possible to assess the real extent of the reptile biodiversity there. This is an essential preliminary to designing a conservation strategy for unique radiations that have already lost some of their most spectacular components through extinction.

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