

None of the previous conventional studies recognized the Mahe and Praslin populations as conspecific to one another and also specifically distinct from the Madagascan population. Some of these studies considered the Praslin and Madagascan populations as conspecific, whilst others considered the Aldabra and Mahe populations as conspecific. Additional permutations are indicated by various authors (reviewed briefly in Thorpe, 1985, and fully in Gardner, 1984) but none reflects the arrangement as indicated by the biochemical affinities (Thorpe, 1985). These conventional studies were based on morphological characteristics so the question arises as to whether it was differences in taxonomic methodology/philosophy or differences in character type that resulted in the novel conclusions of the previous biochemical study (Thorpe, 1985).

This study aims to answer the question by using the same philosophical approaches and procedures (i.e. ordination analysis for phenetics and Wagner trees for cladistics) to analyse morphological features as were used to analyse the biochemical data. Dentition and osteology provide few differences between the taxa, and the degree of autapomorphy in the colour pattern renders it difficult to detect homologous characters. Consequently, shape and scalation were used as the morphological character sets.

To be logically complete, one should have a phenetic and cladistic classification of the 'characters' used in the conventional studies and also subject to the biochemical and quantitative morphological data to 'conventional' analysis. In practice, however, neither is possible because there is no consistent, *a priori*, list of quantified characters in the conventional studies that could be readily used in phenetic and cladistic classification and any attempt of the current author to simulate the 'author-dependent' approach of a 'conventional taxonomist' would be meaningless.

As well as these taxonomic considerations, the relative rates of divergence of the lineages, i.e. the anagenesis, shown by different character systems is of evolutionary interest. Therefore, the phylogenies inferred from the Wagner trees are compared with respect to the relative anagenesis of the lineages.

Methods

Populations

Five populations were investigated using male specimens. The central granitic Seychelles were represented by samples from Praslin ($n = 8$) and Mahe ($n = 8$); the 2 geographically close coralline islands of Aldabra ($n = 10$) and Assumption ($n = 10$) were sampled as was *P. madagascariensis* ($n = 7$) from Madagascar. The conventional classification and geographical position of these island populations are given in Thorpe (1985).

Phylogenetics

Both the biochemical and morphological data were coded/recoded as binary variables (as described below). The unrooted minimum length Wagner trees were then derived directly from a character \times population matrix (Farris, 1970) and the character states at the nodes were hypothesized. Exhaustive searches for minimum length trees, using an updated version of Colless's (1980) Wagner program, yielded several equally parsimonious trees for each data set.

The extent of anagenesis in a lineage is taken as the length of the lineage from its most divergent population to the junction with the rest of the tree. Instead of presenting an unrooted tree, the mid-point is taken as the root but, apart from a few rare cases, the mid-point is generally near, or on, the point at which the major lineages branch. An outgroup root could not be used because single-taxa outgroups can be very unreliable and the biochemical procedures did not allow several taxa to be used as an outgroup (Thorpe, 1985).

Ordination

The relative similarity of the populations was assessed by principal component/coordinate analysis on the same character \times population matrices as were used for computing the Wagner trees. The position of a Wagner tree node along a principal component is found as the sum product of the hypothesized character state vector of the node and the eigenvector of the component. This allows a Wagner tree to be fitted directly on to an ordination diagram to illustrate convergence and divergence.

The recoding procedure that produced the binary variables on which the PCA's were based did not fully take into account the within-group covariance between the morphological characters. Therefore, the 'shape' and 'scalation' principal component analyses were checked by canonical analysis of untransformed characters as this technique does take into account the within-group covariances. The canonical analyses were based on multiple-group principal component scores (Thorpe *et al.*, 1982; Thorpe & Leamy, 1983; Thorpe, 1983*a*). This may allow heteroscedasticity in the within-group covariance matrices to be revealed (Thorpe, 1983*a*) and a growth vector to be defined and deleted if so desired (Thorpe *et al.*, 1982; Thorpe & Leamy, 1983).

Characters, coding and recoding

Biochemical

Details of the biochemical analysis and data coding are given in Thorpe (1985). The biochemical data represents presence or absence of isoelectric focused keratin monomers derived from the epidermis (Thorpe & Giddings, 1981, 1983). Band differences at 17 isoelectric points were recorded giving 17 binary variables (0 for absence, 1 for presence).

Shape

The following 16 linear dimensions were recorded: (1) snout-vent length, (2) jaw length, (3) snout length, (4) eye diameter, (5) ear to eye length, (6) ear length, (7) jaw to ear length, (8) snout width, (9) width between eyes, (10) skull width, (11) upper hind leg length, (12) lower hind leg length, (13) skull depth, (14) neck length, (15) hind leg fourth toe length, (16) hind leg fourth toe width.

Body size varies between populations and would be a valid character to include if it could be measured independently of the ontogenetic growth stage of these growing and unaged lizards. Egg size or hatching size would be a useful 'growth-independent' measured size but these measurements were not available for all the populations. Snout-vent length was taken as a measure of 'size' and, because of the problems associated with it being growth-influenced, the Wagner trees and principal component analyses were both with and without it.

Obviously, all the other linear dimensions are also influenced by growth. As explained in Thorpe (1983*a, b*), there are two facets to the problem when ontogenetic growth influences the character states used to assess inter-population affinity. First, the samples may be growth-biased so that one population contains a preponderance of young, small specimens, whilst another contains a preponderance of old, larger specimens. Secondly, and more important, the influence of a general factor, such as growth, results in high within-group (and therefore high between-group, Thorpe, 1976) character correlations because each character is repeatedly measuring a similar feature of the phenotype, e.g. size. If one can take into account the within-group covariance, then the second facet is not a problem. Whilst this is readily achieved by the selection of an appropriate ordination technique, i.e. canonical analysis, it is more of a problem when constructing Wagner trees.

Wagner trees can be constructed from the (within-group orthogonal) multiple-group principal component scores (Thorpe *et al.*, 1982; Thorpe, 1983*a*), but such data manipulation is unlikely to appeal to cladists. Consequently, for the Wagner trees and principal component analyses, linear dimensions 2 to 16 were regressed against a standard measure of size (i.e. snout-vent length) using the pooled within-group regression slope (Thorpe, 1976). This effectively removes the inter-correlating influence of growth size (Thorpe, 1983*b*).

The characters were then gap-coded (Mickevich & Johnson, 1976; Johnson & Mickevich, 1977; Thorpe, 1984) using a gap of one pooled within-group standard deviation between sample means. Failure to regress the linear dimensions against snout-vent length results in them all having a similar gap-coding pattern to snout-vent length. That is, similar coding (1) for Madagascar and Praslin populations because their lizards are large and similar coding (0) for Mahe, Aldabra and Assumption populations because their lizards are relatively small.

TABLE I(a)
Body proportions (characters 4 to 16 adjusted)

Character	Madagascar	Assumption	Aldabra	Mahe	Praslin
1	1	0	0	0	1
4	1	0	0	0	0
6	0	1	0	1	1
7	0	1	0	0	0
8	1	0	0	1	1
9	1	1	1	0	0
10	0	1	1	0	0
13	1	1	1	1	0
14	0	1	0	0	0
16	0	1	1	0	0

Adjusted characters, 4, 6, 7, 8, 9, 10, 13, 14 and 16 possessed one or more gaps. When more than one gap is present, the data were converted to a binary state by additive coding (Sokal & Sneath, 1963). The resultant 11 binary shape variables are listed in Table I(a) together with gap-coded snout-vent length (1).

The 'growth-free' canonical analysis of shape was based on multiple-group principal component scores derived from the original 16 linear dimensions. The first multiple-group principal component was taken as representing ontogenetic 'growth' and was excluded.

Scalation

The following scalation characters were recorded: (1) number of femoral pores, (2) number of scales in femoral scale row, (3) number of rows from first toe to femoral row, (4) number of lamellae on hind fourth toe, (5) number of posterior upper labials, (6) number of lower labials, (7) number of anterior upper labials in front of eye, (8) number of scales above anterior upper labials, (9) number of scales in contact with first 4 upper labials, (10) number of scales in contact with lower labials, (11) extent of rostral creasing, (12) number of upper labial scales in front of third lower labial, (13) number of scale rows between eyes, (14) number of scale rows between ear and eye, (15) number of scale rows between jaw angle and ear, (16) number of lamellae on hind first toe, (17) keeling on flanks, (18) number of scale rows on belly, (19) number of scale rows between the vent and the enlarged subcaudals, (20) number of longitudinal caudal rows on the second tail segment, (21) number of circum-tail rows on the second tail segment, (22) comparative size of the parasubcaudals, (23) comparative size of the small subcaudals, (24) number of dorsal rows on the first tail segment, (25) proportion of divided subcaudals on the first tail segment.

Fourteen of these characters (Nos. 1, 3, 4, 6, 7, 13, 14, 15, 16, 18, 19, 20, 21, 22) could be gap-coded using a gap of one pooled within-group standard deviation between group means. The gap-coding for characters 1, 4, 14 and 15 was carried out after they had been regressed against snout-vent length as they had a significant within-group correlation with snout-vent length. Where more than one gap was

TABLE I(b)
Scalation (*A = adjusted*)

Character	Madagascar	Assumption	Aldabra	Mahe	Praslin
1(A)	1	0	0	0	0
3	1	0	0	1	1
4(A)	1	0	0	1	1
6	1	0	1	1	0
7	1	0	0	1	1
13	1	0	0	0	0
	1	0	1	1	1
14(A)	1	0	0	1	1
	1	0	0	0	0
15(A)	1	0	0	1	1
	1	0	0	0	0
16	1	0	0	0	1
18	1	0	0	0	0
19	1	0	0	0	0
20	1	0	1	1	1
21	1	0	0	1	1
	0	0	0	1	1
22	1	0	1	1	1

present, the data were converted into binary variables by additive coding. The resultant 18 binary scalation variables are listed in Table I(b). The canonical analysis was run on all 25 original scalation characters.

Results

Biochemical

The adopted taxonomy, as explained in the introduction, is based on the biochemical data. The details of the biochemical results are given in Thorpe (1985), where a Wagner tree (tree (i), Fig. 1(b)) is fitted directly on to a principal component ordination. The principal component analysis and alternative trees are presented separately here. The principal component analysis (Fig. 1(a)) indicates that the five populations fall into three groups corresponding to the species *P. madagascariensis* (Madagascar), *P. abbotti* (Aldabra and Assumption) and *P. sundbergi* (Mahe and Praslin).

Two equally parsimonious Wagner trees were derived from the biochemical data (Fig. 1(b)). These trees are very similar in that they show three specific lineages corresponding to the three phenetic groups and they show the *abbotti* lineage with less anagenesis than the other two lineages. These two trees differ in the relative position of Aldabra and Assumption and in the relative position of the mid-point root to the node joining the *madagascariensis* lineage.

Shape

The growth-free canonical analysis clearly shows (Fig. 2(a)) that the five populations aggregate according to their specific relationships into the same three groups as indicated by the biochemical

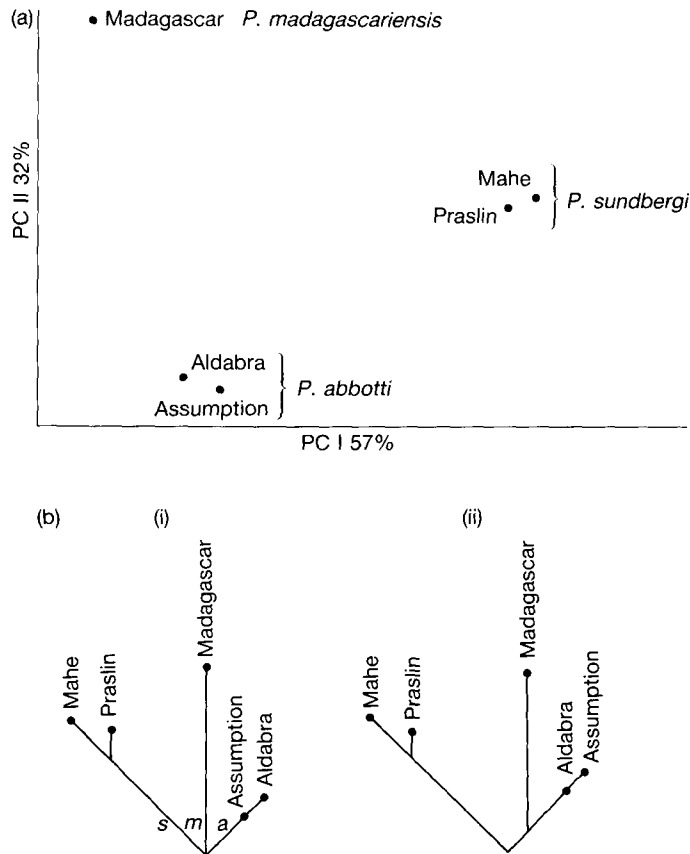


FIG. 1. Analysis of biochemical (keratin) data based on 17 binary variables (Thorpe, 1985) for (a) principal component analysis. (b) Wagner trees 18 units long. *Phelsuma madagascariensis* (*m*), *P. abbotti* (*a*) and *P. sundbergi* (*s*).

data, i.e. *madagascariensis*, *abbotti* and *sundbergi*. This is confirmed by the principal component analysis (Fig. 2(c)) based on the 11 binary shape variables (Table I(a)).

Three equally parsimonious Wagner trees were computed from the 11 binary shape variables (Fig. 2(b)). Trees 2b(i) and (iii) are similar to one another in that there is a cladistic level at which the same three lineages are apparent. These three lineages correspond to the three phenetic groups, i.e. *madagascariensis*, *abbotti* and *sundbergi*. Plotting tree 2(i) on the corresponding PCA (Fig. 2(c)) reveals no noticeable convergence in shape between the five populations.

When size (character 1) is added to shape to give 12 binary variables (Table I(a)) the PCA is basically the same (Fig. 2(d)) as that based on just shape variables (Fig. 2(c)). Four equally parsimonious Wagner trees were extracted from this size plus shape data and they are basically similar to the tree based on shape alone. Consequently, inclusion or exclusion of a single ontogenetically influenced size character makes little difference to either the phenetic or phylogenetic analyses. It is important to note that failure to regress the other linear dimensions against snout-vent length would mean that all of the linear variables would be a repeated measure

of 'size' and the resultant Wagner tree would be very different from the ones presented here as 'size plus shape' trees.

Two of the populations are based on specimens that are rather large for *Phelsuma*, i.e. Praslin and Madagascar. Is their similar size due to convergence? One of the Wagner trees (Fig. 2(e)(i)) hypothesizes large size convergence of the Praslin and Madagascan specimens but the other trees (i.e. trees 2(e)(ii) to (iv)) do not. If tree 2(e)(i) is fitted on the corresponding PCA (Fig. 2(d)), the hypothesized convergence is illustrated. Whilst tree 2(e)(i) hypothesizes large size convergence, the remaining trees, 2(e)(ii) to (iv), hypothesize 'small' size convergence of the Mahe population and the Assumption/Aldabra lineage. Consequently, the choice is between large size convergence in one tree versus small size convergence in three of the trees and the question is unresolved. The trees based on shape (and size plus shape) show less anagenesis in the *madagascariensis* lineage than in the other two lineages (i.e. *abbotti* and *sundbergi*).

Scalation

The canonical analysis (Fig. 3(a)) of the original scalation characters shows the same three phenetic groups as the keratin and shape analyses, i.e. *P. abbotti*, *P. madagascariensis* and *P. sundbergi*. This is confirmed by the PCA (Fig. 3(c)) on the 18 binary scalation variables listed in Table I(b).

Three equally parsimonious Wagner trees (Fig. 3(b)) were extracted from this scalation data (Table I(b)) but only one of these trees (3(b)(i)) is cladistically congruent with the Wagner trees based on the biochemical and shape data. The other two trees (3(b)(ii) and (iii)) do not show Mahe and Praslin as a monophyletic clade and are consequently incongruent with all other trees in this study. Note that the phenetic analysis of scalation is congruent with all the other phenetic analyses and would result in a classification consistent with the classification produced by the cladistic analysis except for the above-mentioned 3(b)(ii) and 3(b)(iii) trees.

When tree 3(c)(i) is fitted on to the corresponding PCA (Fig. 3(c)) there is no clear convergence of any of the taxa. In the scalation data set (tree 3(b)(i)), it is the *P. sundbergi* lineage which shows less anagenesis than the other two lineages.

Total data set

The total data set is based on binary variables from the keratin biochemistry (17), shape (11) and scalation (18), which gives 46 binary variables in all. Since the phenetic analyses for each separate character set generally indicated the same three groups, it is not surprising to find that the PCA of the total data set also shows these same three groups, i.e. *P. abbotti*, *P. sundbergi* and *P. madagascariensis* (Fig. 4(a)).

Four equally parsimonious Wagner trees were extracted from the total data set (Fig. 4(b)). They are all very similar and differ only slightly in the extent of anagenesis. The only obvious difference in the trees is in the extent of anagenesis of the Aldabran population. These trees all show three lineages corresponding to the three phenetic groups, i.e. *P. abbotti*, *P. sundbergi* and *P. aldabra*. The three lineages show approximately equal divergence for the total data set. When plotted on the corresponding PCA (Fig. 4(a)), no outstanding convergence is apparent. The appearance of relatively great divergence between Aldabra and Assumption compared to that between Mahe and Praslin is simply a function of the fact that the first two principal components do not represent the total variation between the latter two populations.

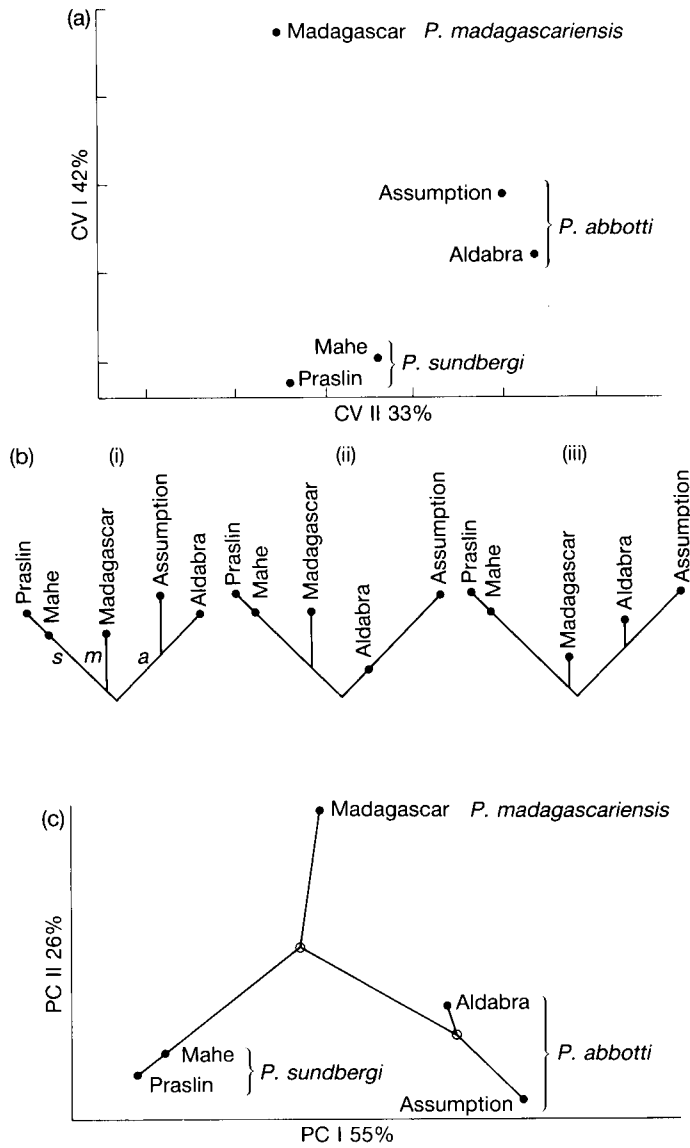
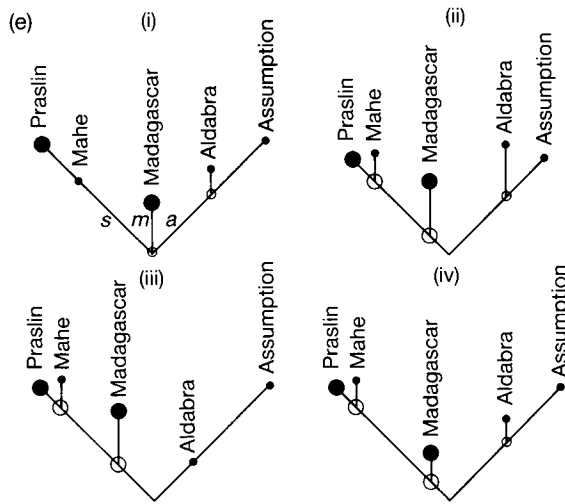
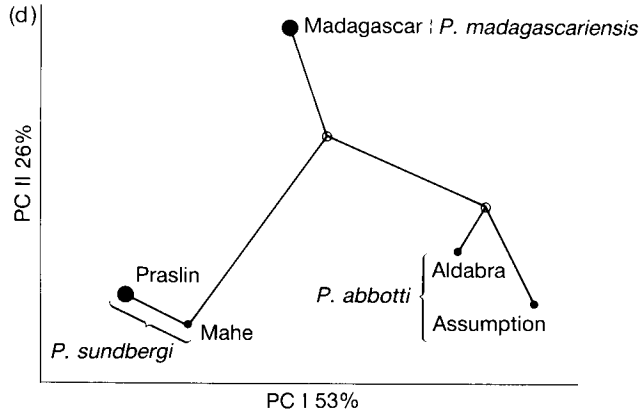


FIG. 2. Analysis of shape and size data. *Phelsuma madagascariensis* (m), *P. abbotti* (a) and *P. sundbergi* (s). Where appropriate, populations are solid circles (●) and tree nodes are empty circles (○) with large and small snout-vent length indicated by large or small circles. (a) Growth-free canonical analysis. Axis units are two within-group standard deviations. (b) Wagner trees, 13 units long, based on 11 binary shape variables. (c) Principal component analysis of 11 binary shape variables with Wagner tree 2(b)(i) fitted on. (d) Principal component analysis of 12 binary 'size plus shape' variables with Wagner tree 2(e)(i) fitted on. (e) Wagner trees, 14 units long, based on 12 binary 'size plus shape' variables.



Conclusions and discussion

One of the aims of this study was to test whether the difference between the previous numerical analysis of biochemical affinities (Thorpe, 1985) and the conventional taxonomy was due to information (i.e. morphology vs. biochemistry) or a difference in taxonomic philosophy and methodology. The results show clearly that when morphological data are numerically analysed in the same manner as the biochemical data, then the taxonomies are congruent irrespective of whether phenetic or cladistic procedures are employed. Consequently, the difference between the conventional taxonomy and the phenetic and cladistic biochemical affinities appears to be due to the type of methodology/philosophy rather than type of information. Out of the three types of taxonomic procedure, the phenetic and cladistic taxonomies agree, whilst the conventional taxonomy differs. The agreement between the numerical analysis of the biochemical and morphological data confirms the taxonomy suggested in Thorpe (1985), i.e. the five populations fall into three taxa at the species level, *P. sundbergi* (Mahe, Praslin), *P. abbotti* (Assumption, Aldabra) and *P. madagascariensis* (Madagascar).

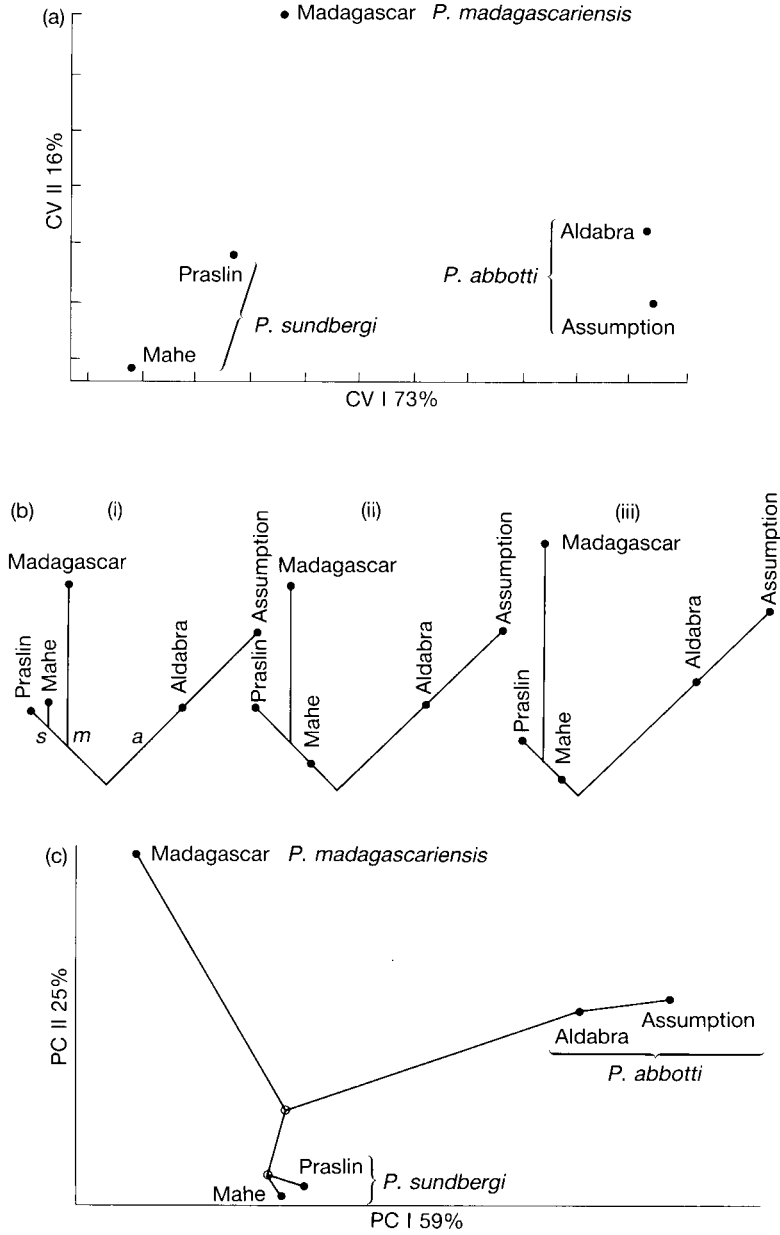


FIG. 3. Analysis of scalation data. *Phelsuma madagascariensis* (m), *P. abbotti* (a) and *P. sundbergi* (s). (a) Canonical analysis of scalation. Axis units are two within-group standard deviations. (b) Wagner trees 20 units long based on 18 binary scalation variables. (c) Principal component analysis based on 18 binary scalation variables with Wagner tree 3(b)(i) fitted on. Populations are solid circles (●) and nodes are empty circles (○).

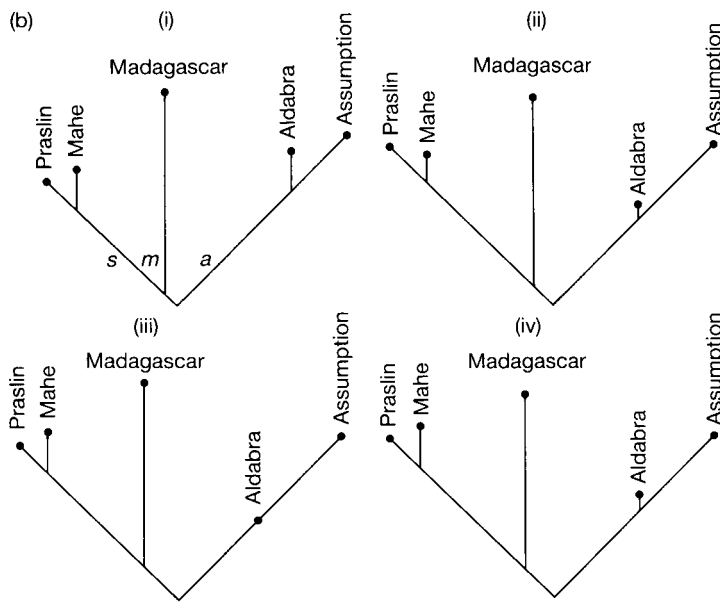
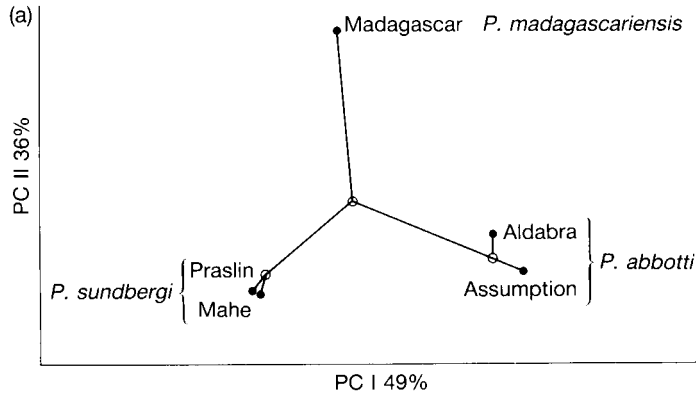


FIG. 4. Analysis of total data set. *Phelsuma madagascariensis* (*m*), *P. abbotti* (*a*) and *P. sundbergi* (*s*). (a) Principal component analysis based on 46 binary variables with Wagner tree 4(b)(i) fitted on (symbols as for Fig. 3(c)). (b) Wagner trees 51 units long based on 46 binary variables from biochemistry, shape and scalation.

The phenetic analyses appeared very robust between character types. All the character sets (biochemistry, shape, scalation and total) show the three compact, largely equidistant groupings. Moreover, the two types of ordination technique, based on two types of coding system and character selection, were also entirely congruent.

The phylogenetic analyses were also generally robust between character types, although two of the three scalation trees were incongruent. As is to be expected (Colless, 1983), all the data sets allowed several equally parsimonious trees to be constructed. With the exception of the

above-mentioned scalation trees, these alternative trees were very similar but differed in relative anagenesis.

It is notable that, whilst the 'total character' Wagner trees showed relatively equal amounts of anagenesis in each of the three lineages, the trees derived from the individual character sets each showed one lineage with less divergence than the other two. For the biochemical data set, the *P. abboti* lineage showed less anagenesis, for the shape data set, the *P. madagascariensis* lineage showed less anagenesis, and for the scalation data set, the *P. sundbergi* lineage showed less anagenesis.

This differential anagenesis could be an artefact of original character selection and/or selection during recoding. Whilst the number of original characters in each set is comparable to the number used in similar studies, the number used was not particularly large, i.e. biochemistry 17, shape 16, scalation 25. It is therefore possible that a larger sample of characters in each individual set would result in equal anagenesis in each lineage, as is shown by the large 'total' data set.

Selection due to recoding pertains to the morphological but not the biochemical sets. In the morphological sets, selection for gap-coding did operate as not all the characters could be gap-coded. Gap-coding was used for the morphological characters because a previous study (Thorpe, 1984) indicated that it gave better congruence between trees than other coding methods and because it can give binary data commensurate with the binary biochemical data. The previous study also indicated that gap recognition could be fortuitous and the anagenesis consequently unreliable (see also Riska, 1979).

However, in this current study most of the characters could be gap-coded, so the rejection rate for characters that could not be gap-coded was very low, i.e. 40%. This is very much lower than in the study that questioned the reliability of anagenic rates when based on a few gap-coded characters (Thorpe, 1984). In that study, the rejection rate was in excess of 90% for unscreened characters and 73% for screened characters (where screening refers to rejecting characters with no significant difference between groups or high within-group correlation with other characters). This rejection figure is the same as that in other studies (Mickevich & Johnson, 1976; Johnson & Mickevich, 1977), where only six out of 22 characters were gap-coded, giving a rejection rate of 73%.

Since few characters were rejected by gap-coding in this study, and since gap-coding does not in any case pertain to the biochemical set, I feel that the chance events of character rejection during recoding are unlikely to have contributed much to causing the unequal pattern of anagenesis between individual character sets.

Whatever the reason for different patterns of anagenesis between individual character sets, the results argue for a range of character systems or large numbers of characters being used when comparative anagenesis is studied. Whilst the Wagner trees were generally congruent between data sets, the incongruence of some scalation trees would also support the use of a range of character systems for cladistic studies. In general terms, this study did not indicate that biochemical data are superior to morphological data for studying relative similarity, anagenesis or cladogenesis.

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