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PHYLOGENY AND COADAPTATION OF THERMAL PHYSIOLOGY IN LIZARDS: A REANALYSIS

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Traditional comparative studies examine evolutionary associations of traits by comparing different species, regardless of their phylogenetic relationships. A problem with such an approach is that trait values for each species are not statistically independent. This is so because species partially share evolutionary histories (Felsenstein, 1985). Another problem is that "equilibrium" patterns seen among extant (or tip) species may not be reflective of preceding evolutionary "transformations" (Lauder, 1981). To help circumvent these difficulties, two of us (Huey and Bennett, 1987) developed a method to analyze the correlated evolution of continuous traits within an explicitly phylogenetic context. This method was developed specifically to examine "coadaptation" of physiological and behavioral traits relating to the thermal biology of a group of Australian scincid lizards. Subsequently, Martins and Garland (1991) have used computer simulation to compare the statistical properties of the Huey/Bennett approach, and some improvements thereof, with those of traditional nonphylogenetic analyses, and of the independent contrasts method of Felsenstein (1985). In addition, new information on the relationships and divergence times of the Australian lizards in our original study has become available. We have therefore reanalyzed our data according to this new information. Most of the previously significant correlations between traits have been altered, with implications not only for our own study but also for the design and interpretation of comparative studies in general. Our reanalysis is timely, because interest in phylogenetically based methods for analyzing comparative data has expanded rapidly since 1987 (e.g., Felsenstein, 1988; Bell, 1989; Burt, 1989; Donoghue, 1989; Grafen, 1989; Lauder and Liem, 1989; Gittleman and Kot, 1990; Maddison, 1990, 1991; Harvey and Pagel, 1991; Lynch, 1991).

Huey and Bennett (1987) examined the hypothesis that evolutionary changes in preferred body temperature (T_p) (determined in a laboratory thermal gradient) should be positively associated ("coadapted") with evolutionary changes in functional thermal limits and the thermal sensitivity of sprint running speed. To index the latter, we estimated the optimal temperature for sprinting (T_o) (the body temperature at which animals can run fastest) for 12 species of Australian skinks. These values were compared to previously published determinations for lower (CTMin) and upper (CTMax) critical temperatures for the species (Bennett and John-Alder, 1986). We applied two different analytical methods. First, following suggestions by Clutton-Brock and Harvey (1984), we tested for correlations using generic averages of traits ($N = 6$), because a nested ANOVA by taxonomic level indicated that 91% of the variance in T_p occurred among genera. Second, to examine coadaptation in an historical framework, we developed a phylogenetic approach, overlaying the thermal data on a phylogeny of the six genera and applying a "minimum evolution" algorithm to estimate ancestral values at nodes. Coadaptation was again suggested by significant regressions involving inferred changes (most recent nodes to generic tips, $N = 6$) in T_o vs. T_p and in CTMax vs. T_p.

Because both tests indicated significant positive correlations between thermal preferences and measures of thermal sensitivity, we concluded that thermoregulatory behavior and physiology were at least partially coadapted. This conclusion appeared robust because both tests appeared conservative: although 12 species were measured, only six generic averages or inferred changes (hence 4 degrees of freedom instead of 10) were used for significance tests. However, new phylogenetic information (Baverstock and Donnellan, 1990; Greer, 1990, pers. comm.; Hutchinson et al., 1991; S. C. Donnellan, pers. comm.) suggests that there are closer to three statistically independent taxa, not six (Fig. 1). Moreover, new phylogenetic methods enable us to calculate more accurate significance tests and better estimates of evolutionary correlations (Felsenstein, 1985, 1988; Grafen, 1989; Martins and Garland, 1991).

We apply three new analytical techniques: (1) simple correlation analysis of species values, using empirical null distributions derived from computer simulation for significance testing (Martins and Garland, 1991), rather than probabilities derived from conventional tests of correlation coefficients (e.g., Zar, 1984); (2) revised minimum evolution analyses that estimate

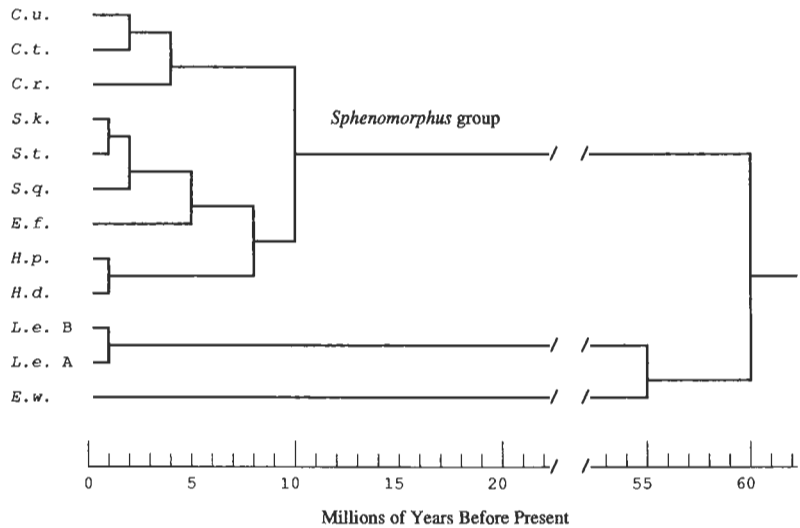

FIG. 1. Hypothesized phylogenetic relationships and estimated divergence times for 12 species of Australian scincid lizards (based on Baverstock and Donnellan, 1990; Greer, 1990, pers. comm.; Hutchinson et al., 1991; S. C. Donnellan, pers. comm.). For purposes of phylogenetic analyses of thermal data, species trichotomies within *Ctenotus* and *Sphenomorphus* were resolved by assuming that species closer in T_o (conceptually, the independent variable) were also closer phylogenetically. This assumption has a loose parsimony justification (Pagel, manuscript : cf Felsenstein, 1983). Absolute timing of the divergence of the *Egernia-Leiolopisma* group from the Sphenomorphus group (60 million years) and the diversification within the *Sphenomorphus* group (10 million years) is based on immunological distance and a molecular clock assumption (Baverstock and Donnellan, 1990; S. C. Donnellan, pers. comm.). Timing of the split between *Egernia* and *Leiolopisma* groups is assumed, based on the fact that morphological analyses (Greer, 1990, pers. comm.) indicate *Egernia* and *Leiolopisma* as sister taxa, whereas immunological data do not resolve the *Egernia-Leiolopisma-Sphenomorphus* group trichotomy. Species within a genus (*Sphenomorphus, Hemiergis, Leiolopisma*) were arbitrarily assumed to have diverged one million years ago, except for the three *Ctenotus,* which are in different species groups (Greer, 1990) and so were assumed to have split at two million year intervals. Timing of other splits is arbitrary.

whether inferred changes in traits are correlated along branches (Martins and Garland, 1991), and (3) independent contrasts methods (Felsenstein, 1985, 1988; Grafen, 1989). These three approaches generally yield the conclusion that the original analyses—though intended to be conservative—were not conservative enough, such that evidence for coadaptation of some traits is weaker than previously thought.

MATERIALS AND METHODS

Simple Correlation Analysis

Thermal data and hypothesized phylogenetic relationships of the 12 species studied by Huey and Bennett (1987) are given in Table 1 and Figure 1, respectively. [Huey and Bennett (1987) also used a generic mean value for *Mabuya* for T_p, CTMax, and CTMin. As no data are available for T_o, we have excluded this genus in the present reanalysis.] Pearson product-moment correlations for species (not generic) values of the four thermal variables are presented in Table 2 (first row in each comparison). These correlations constitute a traditional, nonphylogenetic approach, termed "TIPS" by Martins and Garland (1991). The correlations make no assumption as to whether the mode of evolution is punctuational or gradual (see below), but they do assume (inappropriately) that the species values are statistically independent. Conventional significance tests with $N - 2 = 10$ *df* indicate that correlations greater than +0.497 (1-tailed) are significant at $\alpha < 0.05$ (see Zar's, 1984, Table B.16). (One-tailed tests are appropriate for the stated hypothesis of positive coadaptation of thermal variables.) Judged in this way, three of six intercorrelations are significant [T_p with T_o (r = 0.585) and CTMax (r = 0.885), and T_o with CTMax (r = 0.619)], suggesting coadaptation between behavior and thermal sensitivity. Huey and Bennett (1987), working with generic averages, found similar patterns of significance.

The foregoing significance tests make no attempt to correct for phylogenetic nonindependence and therefore tend to overestimate the significance of correlations (Felsenstein, 1985; Grafen, 1989; Martins and Garland, 1991). Valid significance tests are possible, however, by reference to an empirical null distribution of correlation coefficients created via computer simulation. Using programs available from Martins and Garland (1991), we created such a null distribution (below). These simulations require specification of both branch lengths and model of evolutionary change (e.g., gradual or punctuational; Martins and Garland, 1991 : cf Friday, 1987) appropriate for the characters being analyzed. The former, based on immunological distance, have recently become available for Australian skinks (see Fig. 1). However, whether evolution in this

TABLE 1. Thermal data (°C) for species means for 12 scincid lizards [from Bennett and John-Alder (1986) and Huey and Bennett (1987)]. See text for abbreviations.

Species	T_p	T_o	CTMin	CTMax
Ctenotus uber	35.3	36.5	9.1	45.5
Ctenotus taeniolatus	35.3	35.7	11.4	44.7
Ctenotus regius	35.6	33.0	8.7	45.1
Sphenomorphus kosciuskoi	29.8	33.6	2.5	40.2
Sphenomorphus tympanum	29.5	33.1	2.9	39.8
Sphenomorphus quoyi	28.8	29.5	6.0	40.8
Eremiascincus fasciolatus	24.4	34.0	9.0	41.2
Hemiergis peronii	23.5	30.8	9.6	38.8
Hemiergis decresiensis	24.8	33.2	6.8	38.6
Leiolopisma entrecasteauxii B	33.9	34.5	2.5	42.8
Leiolopisma entrecasteauxii A	32.5	33.4	2.5	42.8
Egernia whitii	34.1	35.3	4.0	42.8

group was gradual or punctuational (or a combination of both) is unknown. As a result we analyze both extreme cases. First, we assume that evolutionary change was gradual (Brownian motion) and that the branch lengths of Figure 1 are correct. (Actually, only the proportionality of these divergence times need be assumed; that is, all could be off by a constant factor without invalidating the analysis.) Second, we assume all evolutionary change was punctuational, occurring only at speciation events, equally in both daughter species, and that all speciation events are represented in Figure 1 (Martins and Garland, 1991). Not all speciation events are depicted: many other extant species are in these genera, and the number of extinct species is unknown. Nevertheless, an analysis assuming punctuational change is at least useful for heuristic purposes. Moreover, our original analysis (Huey and Bennett, 1987) used a type of minimum evolution analysis that assumed punctuational change (see below). (Our use of a punctuational model for analytical completeness should not be taken to imply that we think such a model is realistic; we do not.)

The simulation procedures follow Martins and Garland (1991). Beginning with an initial value (we used the observed mean for the 12 species) at the bottom of the phylogeny of Figure 1, changes for two traits are drawn randomly from a bivariate normal distribution with correlation zero (because the null hypothesis is no correlation). For simulations assuming gradual evolution, these changes are added at intervals equivalent to the lowest common denominator of branch lengths (every million years in the present case) until values for two traits at the tips of the phylogeny are obtained. For simulations assuming punctuational evolution, a single change is added at each speciation event (that is, for each branch segment). These procedures are repeated 1,000 times resulting in 1,000 sets of simulated species data. A correlation coefficient is then computed for each of these 1,000 data sets. The resulting distribution of correlation coefficients exhibits a mean not significantly different from zero [all of the methods compared by Martins and Garland (1991) proved to be unbiased], but shows considerable spread, with individual values ranging between +1 and −1. The upper and/or lower 2.5% or 5.0% tails of this distribution can be used to establish empirical critical values for hypothesis testing.

Minimum Evolution Analyses

Nonphylogenetic approaches (above) identify correlations between variables in extant species, but provide no information on the direction or sequences of evolutionary change (Huey and Bennett, 1987; Huey, 1987). Thus, correlations based simply on tips of a phylogeny may not be good estimators of what actually happened during evolution (Martins and Garland, 1991). Accordingly, Huey and Bennett (1987) developed a "minimum evolution" approach in an attempt to reconstruct historical patterns of evolutionary change. These procedures have been generalized by Martins and Garland (1991) in a method termed "ME1." This general minimum evolution approach uses an iterative averaging algorithm to compute nodes as the weighted average of surrounding nodes or tips on a phylogeny (see also Maddison, 1991). This computation is done independently for each character. Then, changes between nodes and between nodes and tips are computed by simple subtraction. Finally, a Pearson product-moment correlation between these inferred changes is computed (the number of changes is $2N - 2 = 22$ for a phylogeny of 12 species). Correlations from the ME1 method cannot be tested for significance against standard critical values, but can be tested against empirical null distributions, created as described in the previous section. Two minimum evolution methods are applied: ME1G, which assumes gradual evolution, and ME1P, which assumes punctuational change. Based on the simulations for 12 species, correlations $>+0.789$ (gradual model) or $>+0.519$ (punctuational model) are significant at $\alpha < 0.05$ (1-tailed).

Independent Contrasts

Because new data on branch lengths for the Australian skinks are now available (Fig. 1), valid significance tests can also be obtained using Felsenstein's (1985, 1988) method of standardized independent contrasts. This method requires branch lengths in units of expected variance of change for each character, which is fully equivalent to the requirement of specifying branch lengths for the simulation of empirical null distributions, as described above. Whether immunological distance (or its conversion to time) provides a reasonable estimate of expected variance of change for the thermal variables of interest is unclear (cf. Sessions and Larson,

1987). We applied Felsenstein's method for the cases of both gradual (FL1G) and punctuational (FL1P) change. If either of these evolutionary models is valid, then resulting correlations can correctly be judged against conventional critical values (+0.497).

Finally, we applied Grafen's (1989) "standard regression," a modification and generalization of Felsenstein's approach. As noted above, the estimates of (relative) branch lengths must accurately indicate expected variance of change in order to apply Felsenstein's (1985) method for hypothesis testing validly. Rather than using these branch lengths directly, Grafen's (1989) programs for the "standard regression" use maximum likelihood techniques to estimate simultaneously correlations (or regressions) and "rho," a parameter that indicates, in essence, the power to which all branch lengths should be raised, as dictated by the data. Thus, a rho of unity would not change relative branch lengths. One degree of freedom is lost in estimating rho.

Results

Testing Simple Correlations

By applying the simulation procedure described above to the phylogeny in Figure 1, we computed appropriate significance levels for "TIPS" correlations (i.e., those involving raw species values, Table 2, first rows). For a model of gradual evolution, the distribution of simple Pearson product-moment correlation coefficients ranged from −0.991 to 0.987. Only correlations greater than +0.828 (1-tailed) can be considered statistically significant at $\alpha < 0.05$. T_p is positively correlated with both T_o and CTMax, but only the latter is significant (Table 2). For a model of punctuational change, only correlations greater than +0.643 (1-tailed) can be considered statistically significant at $\alpha < 0.05$. Again, only the correlation between T_p and CTMax is significant, although that between T_o and CTMax just falls short. Note that the use of conventional statistical tests ($r >$ +0.497, 1-tailed, is significant at $\alpha < 0.05$) incorrectly assigns significance to the correlations between T_p and T_o and between T_o and CTMax.

Minimum Evolution Methods

The only significant correlation found with the minimum evolution methods involves T_p and CTMax, assuming punctuational change (ME1P; third row of Table 2). Although the gradual model (ME1G) actually yielded a slightly higher correlation coefficient for T_p and CTMax than did the punctuational model (ME1P), only the latter was significant. This result is probably attributable to the former method having slightly lower power (i.e., ability to detect significant correlations; see Table 3 in Martins and Garland's [1991]). Correlations between T_p and T_o are nonsignificant under either model, contrary to our original analysis (Huey and Bennett, 1987).

Independent Contrasts Methods

Correlations from independent contrasts methods are presented in the fourth and fifth rows of Table 2. As compared with conventional critical values, only the correlation (assuming either gradual or punctuational change) between T_p and CTMax is significant.

Significance tests for this method using conventional critical values are valid only if branch lengths effectively standardize the independent contrasts. This assumption can be checked by testing whether the absolute value of each standardized contrast shows any correlation with its standard deviation. None of the plots for the four thermal variables exhibits a significant linear relationship (although for preferred temperature the correlation was −0.545, $P = 0.083$). Thus, the branch lengths of Figure 1 seem to standardize the independent contrasts adequately, and conclusions derived from comparisons with conventional critical values are valid. As a check on our simulation procedures, we also computed critical values for Felsenstein's method from the empirical null distributions. These empirical critical values (1-tailed) were +0.503 (gradual model) and +0.519 (punctuational model), versus +0.497 (Zar, 1984).

Grafen's (1989) modification of Felsenstein's (1985) method, applied to the present data and phylogeny, indicates that *none* of the correlations is statistically significant (A. Grafen, pers. comm.; results not shown). This discrepancy exists because the F statistics for sig-

Table 2. Pairwise correlations between thermal variables for species means for 12 scincid lizards (data from Table 1). Values are standard Pearson product-moment correlation ["TIPS" of Martins and Garland (1991)], minimum evolution method assuming gradual (Brownian motion) evolution and branch lengths of Figure 1 ["ME1G" of Martins and Garland (1991)], minimum evolution method assuming punctuational change (ME1P), Felsenstein's (1985) independent contrasts method assuming gradual evolution and branch lengths of Figure 1 (FL1G), and Felsenstein's (1985) method assuming punctuational change (FL1P). Significance tests for TIPS, ME1G, and ME1P are based on empirical null distributions created through computer simulations; significance tests for independent contrasts methods are based on comparisons with conventional critical values (from Zar, 1984). Critical values for 1-tailed tests are as follows: TIPS, +0.828 (gradual), +0.643 (punctuational); ME1G, +0.789; ME1P, +0.519; FL1G and FL1P, +0.497.

	T_o	CTMin	CTMax	Method
T_p	0.585	−0.089	0.885*	TIPS
	0.274	−0.372	0.752	ME1G
	0.193	−0.307	0.723*	ME1P
	0.340	−0.334	0.593*	FL1G
	0.193	−0.303	0.722*	FL1P
T_o		0.076	0.619	TIPS
		−0.155	0.296	ME1G
		−0.141	0.230	ME1P
		−0.461	0.080	FL1G
		−0.139	0.230	FL1P
CTMin			0.270	TIPS
			0.090	ME1G
			0.162	ME1P
			0.269	FL1G
			0.169	FL1P

* 1-tailed $P < 0.05$.

nificance testing are very sensitive to rho, and the data suggest that rho is not equal to unity. Also, because rho must be estimated, resulting in the loss of 1 *df*, Grafen's standard regression should have somewhat lower power than Felsenstein's (1985) original method—*if* the branch lengths of Figure 1 adequately represent expected variances of change.

Discussion

Using both nonphylogenetic and phylogenetic approaches, our earlier study (Huey and Bennett, 1987) found evidence of significant coadaptation between thermal preference (T_p) and measures of thermal sensitivity in Australian scincid lizards: genera with high thermal preferences (T_p) typically ran fastest at high body temperatures (T_o) and had high critical thermal maxima (CTMax). As discussed below, however, our approaches had several statistical problems. We now use three rather different approaches that correct for those problems. Our reanalyses suggest robustness of the positive correlation between T_p and CTMax. However, we find no significance for the relationships between T_p and T_o or between T_o and CTMax (Table 2). Thus, coadaptation between thermal preference and thermal sensitivity of these lizards is less pervasive, or at least more difficult to detect statistically, than suggested by our earlier study.

Our earlier analysis had at least three problems. First, as we previously emphasized (Huey and Bennett, 1987 p. 1103), inferred changes along branches are not independent, due to the nature of the minimum evolution averaging algorithm, resulting in overestimation of degrees of freedom. Second, the use of generic means rather than species values loses substantial information (Grafen, 1989; Harvey and Pagel, 1991), even if most of the variance in a particular trait is found among genera (see Huey and Bennett, 1987 p. 1102). Third, evolutionary correlations based only on inferred changes from most recent nodes to tips are considerably less accurate than those based on inferred changes occurring along all branches (Martins and Garland, 1991).

The first problem in the original analysis—over-estimation of degrees of freedom—is a common one in comparative studies (Felsenstein, 1985; Grafen, 1989). This is ironic, because we intentionally used generic averages (hence 4 rather than 10 *df*) and also correlated only inferred changes from most recent nodes to tips to reduce the nonindependence of data. However, new phylogenetic information (Fig. 1) suggests that these 12 Australian skinks belong to essentially three groups, which separated 55–60 million years ago. Indeed, 9 of 12 species belong to a single lineage that has had relatively little time for independent divergence (circa 10 million years). So even four degrees of freedom were too many.

One final caveat concerning our analyses is worth mentioning. Both Felsenstein's (1985) method and the minimum evolution methods (Martins and Garland, 1991) test for correlations of contemporary evolutionary changes. Huey and Bennett (1987), on the other hand, suggested that changes in preferred body temperature (a behavioral trait) should precede changes in optimal body temperature for sprinting (a physiological trait). If this hypothesis is true, then correlations between changes in T_p and subsequent evolutionary changes in T_o might be stronger and easier to detect statistically. Donoghue (1989, pers. comm.) and Maddison (1990) have discussed this distinction with regard to binary characters, but no statistical methodology for detecting sequentially correlated changes in continuous traits has yet been proposed.

Implications for Comparative Studies

As we (Huey and Bennett, 1986, 1987; Huey, 1987; Martins and Garland, 1991) and many others (review in Harvey and Pagel, 1991) have indicated, comparative studies can be most productive when undertaken within a phylogenetic framework. However, our reanalysis indicates that several important obstacles must be considered when designing such a study. First, and obviously, a phylogeny of the group of interest must be available. [Methods do exist for use with only partially resolved phylogenies (Grafen, 1989; Harvey and Pagel, 1991), but are necessarily less powerful.] Second, to be maximally useful, the phylogeny should include branch lengths, which are important for suggesting the statistical independence of various species and for developing gradual models of evolutionary change. {It should be noted that time alone, even if it were known without error, does not necessarily provide a good estimator of expected variance of change (Felsenstein, 1985, 1988; Grafen, 1989; Harvey and Pagel, 1991; Martins and Garland, 1991). For this reason, empirical checks of how effectively branch lengths standardize independent contrasts (see Results) are important [see also Grafen (1989) and Harvey and Pagel (1991) on ways to modify branch lengths].} Third, one must be willing to accept the available phylogeny as a working hypothesis. If the estimated phylogeny is revised, then reanalysis of the comparative data is necessary, and conclusions may change (as in the present case).

How does one choose a comparative method? The TIPS method exhibits both inaccurate estimates of evolutionary correlations and low power to detect nonzero relationships (Grafen, 1989; Martins and Garland, 1991). Thus, even though valid significance tests can be obtained by reference to simulated null distributions (assuming the phylogeny and branch lengths are known without error), the TIPS method has little to recommend it. Both minimum evolution and independent contrasts methods exhibit higher power and yield better estimates, although the actual estimates may differ substantially between these two approaches [Table 2; see also Appendix in Martins and Garland (1991)]. Discrepancies occur because different phylogenetically based comparative methods actually estimate different types of evolutionary correlation (see Martins and Garland, 1991). Limited simulation results suggest that FL1G may generally have greater power than does ME1G (Martins and Garland, 1991). Thus, one might prefer an independent contrasts method for hypothesis testing, but a minimum evolution approach for estimation, and there is nothing wrong with applying both for the two separate purposes (cf. Grafen, 1989 p. 141).

Breadth and detail of phylogenetic coverage is another concern for comparative studies. In the skinks discussed here, most of the interesting thermal evolution (e.g., the low preferred temperatures of *Eremiascincus* and *Hemiergis*; Table 1, Fig. 2) appears to have occurred rather recently and within a restricted subclade (the suprageneric *Sphenomorphus* group). Perhaps it would have been better to sample three more

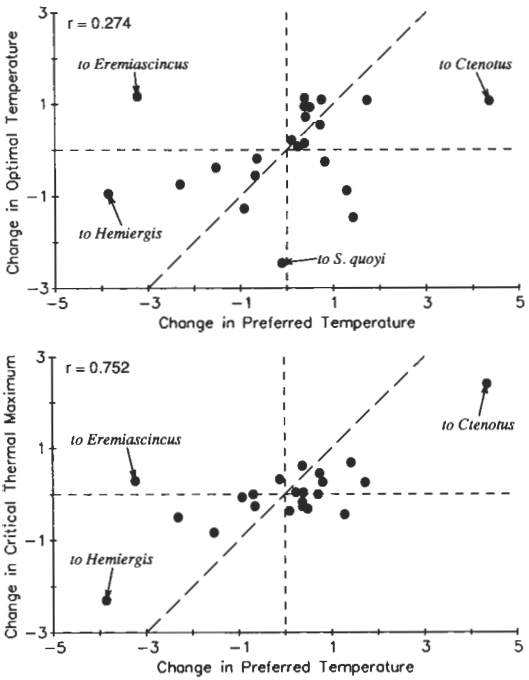

FIG. 2. Bivariate scatterplots of inferred changes in thermal variables, based on minimum evolution reconstructions of nodal values and assuming gradual change. Each point represents changes occurring along one of the 22 branch segments of Figure 1. Dashed line at 45° angle represents perfect coadaptation. Correlations are for ME1G method (see Table 2). The labels indicate changes occurring along branches leading to various taxa.

species within the *Sphenomorphus* group, rather than any *Leiolopisma* or *Egernia* (cf. Fig. 1), which are effectively "outgroups" to the former. However, "outgroups" are important for establishing directions of change in phylogenetic studies (e.g., Maddison et al., 1984; Huey and Bennett, 1987), which would argue for a broader phylogenetic scope. This consideration might suggest more even sampling of the three generic groups, rather than concentrating on one (9 of 12 species sampled belonged to the *Sphenomorphus* group). On the other hand, as we (Huey and Bennett, 1986) and others have noted before, "comparative biologists tend to suspect comparisons of distantly related species" (Felsenstein, 1988 p. 465).

We do not at present know how best to design a comparative study, but it does seem likely that power to detect significant evolutionary correlation will depend on the form of the phylogeny in relation to species sampled. This relationship points again to the need for information on branch lengths in addition to cladistic topologies. Information on branch lengths was not available at the time of our original study; thus, the relative nonindependence of some species and genera was not apparent [compare our Fig. 1 to Fig. 2 of Huey and Bennett (1987)].

Lest the preceding make phylogenetically based comparative studies seem too difficult, we note the following. First, phylogenies, often with some information on branch lengths, are becoming available for many groups. Branch lengths need not be in units of absolute or even relative time, nor do they need to come from molecular or immunological distances. Other possibilities exist. For example, if a cladistic analysis (of characters other than those being mapped onto the phylogeny in the comparative study) was the source of the available phylogeny, then the number of steps (character changes) along each branch might index relative branch lengths. Cheverud et al. (1985), Grafen (1989), and Harvey and Pagel (1991) discuss ways to obtain a set of starting branch lengths when only a taxonomy or when only a cladogram is available. As suggested by Grafen (1989 p. 147), the pragmatic approach is to begin with a "reasonable" set of branch lengths; if two different sets of branch lengths lead to different conclusions, then extreme caution should be used in inferring anything from the data. Second, programs to implement various comparative methods are now readily available (e.g., Grafen, 1989; Harvey and Pagel, 1991; Martins and Garland, 1991). Finally, the recognition by comparative biologists that they need phylogenies should do much to stimulate the work of systematists. In turn, systematists should encourage comparative biologists to study groups for which phylogenetic information is already available or currently being gathered. Collaboration between comparative biologists and systematists may thus become more common [and see comments by J. L. Edwards in Feder et al. (1987 p. 137)].

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