Low Cost of Locomotion in the Banded Gecko:
A Test of the Nocturnality Hypothesis

Kellar Autumn[1,2,*]
Claire T. Farley[2]
Maya Emshwiller[2]
Robert J. Full[2]
[1]Museum of Vertebrate Zoology, University of California, Berkeley, California 94720; [2]Department of Integrative Biology, University of California, Berkeley, California 94720

ABSTRACT

This study tested the hypothesis that there has been an evolutionary increase in locomotor performance capacity at low temperature in nocturnal lizards. Nocturnal lizards are often active at low and suboptimal body temperatures. An evolutionary decrease in the minimum cost of locomotion could increase endurance capacity at low temperature, partially offsetting the thermal handicap of nocturnality. In support of the nocturnality hypothesis, we discovered that minimum cost of locomotion of a nocturnal gecko, *Coleonyx variegatus* (4.2 g), was only 58% of the minimum cost of locomotion of *Phrynosoma douglassii,* a diurnal lizard (4.5 g). As a result, maximum aerobic speed was 2.3 times as great in the nocturnal lizard compared to the diurnal lizard. By using the method of phylogenetically independent contrasts at the species level, we showed that the relationship between mass and minimum cost of locomotion in diurnal lizards was similar to that of the ahistorical standard allometry and that low minimum cost of locomotion in geckos represents a significant evolutionary change from the ancestral diurnal pattern. The decrease in the minimum cost of locomotion concordant with the evolution of nocturnality suggests that geckos evolved a greater capacity for sustained locomotion at low temperature.

Introduction

Nocturnal lizards provide an excellent model system with which to study the effects of an important evolutionary shift in environment. Lizards are ancestrally diurnal, and the majority of lizard species, genera, and families have remained diurnal. Because nocturnal lizards face unique environmental challenges, it is not surprising that nocturnality has evolved only several times over the past 200 million yr. Nocturnal lizards are usually active at body temperatures 10°–35°C below those of diurnal lizards (Cowles and Bogert 1944; Brattstrom 1965; Bustard 1967; Porter and Gates 1969; Thomas 1981; Pianka 1986; Huey et al. 1989; Valakos 1989; Henle 1990; Autumn et al. 1994). Diurnal lizards are generally active at body temperatures near their thermal optimum (30°–40°C; Brattstrom 1965; Dawson 1975; Huey 1982; Avery 1984; Pianka 1986; Huey and Bennett 1987; Huey et al. 1989) for a variety of functions including endurance, sprint speed, growth, digestive efficiency, and hearing. Thus, performance capacity approaches thermally maximal levels during activity in diurnal lizards. In contrast, nocturnal lizards experience much lower body temperatures during activity than is optimal. Even for lizards well adapted to nocturnality, these low temperatures can be substantially suboptimal for sprinting (Huey et al. 1989), growth (Autumn and DeNardo 1995), and sustained locomotion (Autumn et al. 1994). Thus, nocturnality represents a thermal handicap that constrains performance to submaximal levels (Autumn et al. 1994). Given typical rate-temperature effects ($Q_{10} = 2.5$; Bennett 1982), a 15°C drop in body temperature causes a 75% decrease in performance capacity.

The Nocturnality Hypothesis

Nocturnal lizards may have physiological adaptations that offset the thermal handicap. In this study we tested the hypothesis that there has been an evolutionary increase in locomotor performance capacity at low temperature in nocturnal lizards. An animal's maximum aerobic speed (MAS) is an important determinant of its locomotor capacity and is strongly affected by temperature (see Bennett [1982] for review). The MAS is the slowest speed at which the maximum rate of aerobic metabolism (maximum rate of oxygen consumption, $\dot{V}O_{2max}$) is attained. Locomotion at speeds below the MAS is fueled primarily by aerobic metabolism and is sustainable for long periods of time (minutes to hours). Locomotion at speeds above the MAS is fueled primarily by accelerated glycolysis and cannot be sustained for long periods of time (seconds to minutes; John-Alder and Bennett 1981; Gatten et al. 1992). The MAS is a function of both $\dot{V}O_{2max}$ and fuel economy (minimum cost of locomotion, C_{min}). C_{min} determines the speed at
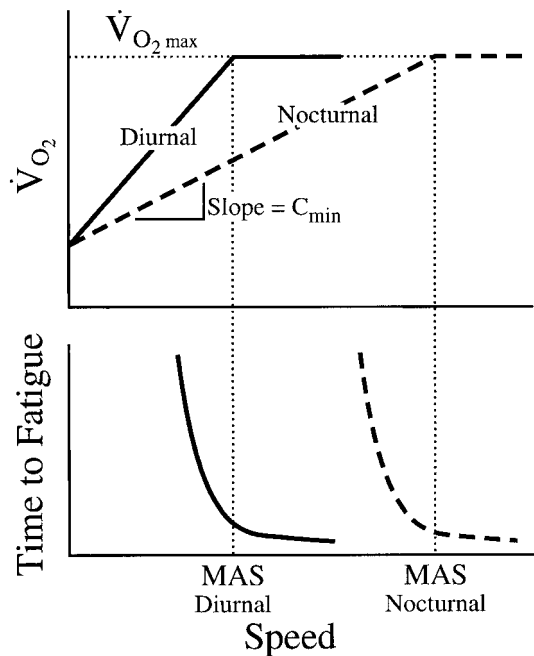
Figure 1. Illustration of the hypothesis that nocturnal lizards have a lower C_{min} than do diurnal lizards. For terrestrial locomotion, $\dot{V}_{O_2}$ increases linearly with running speed. The slope of the lines relating $\dot{V}_{O_2}$ and speed is C_{min} (the amount of energy required to move 1 g over 1 km). The MAS is the speed at which $\dot{V}_{O_{2max}}$ is reached. Time to fatigue (endurance capacity) is strongly dependent on the MAS. Only speeds below MAS are sustainable for long periods of time (minutes to hours). A lower C_{min} causes $\dot{V}_{O_{2max}}$ to be reached at a greater speed, yielding a greater MAS, which in turn increases endurance capacity at sustainable speeds.

which $\dot{V}_{O_{2max}}$ is attained; a lower C_{min} allows a higher MAS. Similarly, an increase in $\dot{V}_{O_{2max}}$ would allow a higher MAS. Thus, the MAS of a nocturnal lizard active at low body temperatures could be increased either by decreasing the C_{min} or by increasing the $\dot{V}_{O_{2max}}$. C_{min} in lizards is independent of body temperature (John-Alder and Bennett 1981; Rome 1982), but $\dot{V}_{O_{2max}}$ is strongly temperature-dependent, decreasing by a factor of two to three for each 10°C decrease in body temperature (Bennett 1982). As a result of the strong dependence of $\dot{V}_{O_{2max}}$ on body temperature, nocturnal lizards face a thermal handicap when they are active at low body temperatures.

We previously hypothesized that the thermal handicap resulting from the reduced $\dot{V}_{O_{2max}}$ at low body temperatures may be offset by a decrease in C_{min} in nocturnal lizards (Fig. 1). This hypothesis is supported by data on C_{min} and the thermal sensitivity of $\dot{V}_{O_{2max}}$ in nocturnal frog-eyed geckos, *Teratoscincus przewalskii* (Autumn et al. 1994). The thermal optimum for $\dot{V}_{O_{2max}}$ is approximately 35°C in *T. przewalskii* (Autumn and Full 1994). Activity at typical nocturnal temperatures (15°C) causes a 62% decrease (thermal handicap) in $\dot{V}_{O_{2max}}$, which does not differ significantly from predicted values for

diurnal lizards (Autumn and Full 1994). *T. przewalskii* has a remarkably low energetic cost of locomotion compared with literature values for diurnal lizards of similar body mass. In fact, these geckos use only one-third of the predicted metabolic energy required by diurnal lizards to travel 1 m. As a result of this low C_{min}, individuals of *T. przewalskii* are capable of sustaining aerobically speeds that are 2.5 times greater than the speeds that can be sustained by phylogenetically comparable diurnal lizards of similar body mass. This is due to a 2.5-fold relative increase in MAS in *T. przewalskii*, which partially offsets the thermal handicap of nocturnality. This observation may explain why it is possible for many nocturnal lizards to be active foragers (Semenov and Borkin 1992; Autumn et al. 1994; B. E. Dial, personal communication; Y. L. Werner, personal communication) in spite of aerobic limitations at low body temperatures.

Farley and Emshwiller (1996) went on to provide evidence for the mechanism resulting in low fuel economy. The nocturnal gecko *Coleonyx variegatus* was shown to be nearly twice as efficient in performing mechanical work during uphill locomotion as the diurnal lizard *Eumeces skiltonianus*. By conducting this study on animals of the same mass, Farley and Emshwiller (1996) made the first direct comparison of C_{min} in a nocturnal versus a diurnal lizard. Their results are consistent with the original hypothesis of nocturnal evolution proposed by Autumn et al. (1994), who compared a nocturnal gecko with literature values of diurnal lizards.

Testing the generality of the nocturnality hypothesis benefits from direct comparisons but ultimately requires a consideration of allometry and phylogeny. In the present study, we conduct a direct comparison of nocturnal and diurnal lizards but do so in a manner that controls for both body mass and evolutionary history.

A Phylogenetically Independent Allometry

Our first objective was to produce a phylogenetically independent allometry of C_{min}. Conclusions about variation in C_{min} are typically reached by standard regression analysis (Full 1989). We first compared data from the present study on nocturnal western banded geckos (*C. variegatus*) and diurnal dwarf short-horned lizards (*Phrynosoma douglassii*) to the standard relationship between C_{min} and body mass developed for lizards (John-Alder et al. 1986). We then applied a phylogenetic statistical method of analysis, because it has become apparent that phylogenetic patterns can affect (and potentially confound) the interpretation of allometric comparisons among species (Felsenstein 1985; Garland et al. 1992). Species are not independent samples because common ancestry tends to produce common features (Losos and Miles 1994). If a feature arises once and persists in future populations and species, each specific instance of the feature is not a statistically independent replicate. For example, numerous, small diurnal lizards could
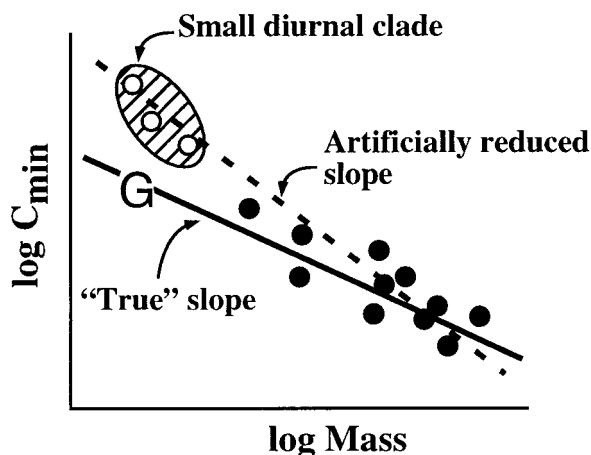
Figure 2. Hypothetical allometry of C_{min} and body mass in lizards. The data points are arbitrary but show the typical negative allometric slope described by John-Alder et al. (1986). The solid line represents the theoretical slope of the statistical population of all lizard species. If small diurnal lizards (*open circles*) belong to a clade with a marginally high C_{min}, this could significantly decrease the slope (elevate the negative slope; *dashed line*), making geckos (*G*) seem as if they have a low C_{min} when they actually have a C_{min} typical of lizards of the same body mass. This study rejected this possibility by using phylogenetic statistical analysis.

belong to a single clade with a marginally high C_{min} (Fig. 2). Inflating the sample size of standard allometry by phylogenetic pseudoreplication could significantly decrease the slope (elevate the negative slope), making geckos seem as if they have a low C_{min} when they actually have a typical C_{min}. Since prior analyses of species variation in C_{min} have not included phylogeny (John-Alder et al. 1986), only an explicitly phylogenetic statistical method of analysis can eliminate these possibilities.

  We selected the method of phylogenetically independent contrasts (Felsenstein 1985; Garland et al. 1992), which takes the nested dependency of species values into account and generates a "phylogenetically independent" allometric relationship. If phylogeny has little or no effect on the allometric pattern, the phylogenetically independent allometry will be similar or identical to a standard allometry. Autumn et al. (1994) used the independent contrasts method to analyze C_{min} in diurnal lizard families. This phylogenetic analysis was consistent with the standard species allometry of John-Alder et al. (1986). However, because it included only family means, it is still possible that the more detailed phylogenetic analysis at the species level used in this study would reveal a different pattern. In order to determine if a low C_{min} is a unique character of *T. przewalskii* or a general characteristic of geckos, a species-level analysis is necessary.

  The independent contrasts method is also capable of identifying patterns of variation in C_{min} that are explained primarily by history and not by body mass. Since we needed to analyze variation in C_{min} and mass at the phylogenetic levels of lizards

(Squamata) in general and geckos (Gekkonoidea) in particular, this method was especially appropriate. If a low C_{min} is associated with nocturnality, we expected the independent contrasts method to separate the ancestral lizard pattern from the derived gecko pattern because geckos share a history of nocturnality. This would indicate that geckos do not follow the same allometric relationship as diurnal species and that a standard allometric analysis of mass and C_{min} in lizards could be misleading.

Evolution of C_{min}

Our second objective was to test the hypothesis that nocturnal geckos evolved a lower C_{min} than their diurnal ancestors. Therefore, we needed to determine the ancestral relationship between C_{min} and mass in diurnal lizards and to test if geckos differ significantly from this pattern. To test this hypothesis, we must eliminate the possibility that low C_{min} evolved in a clade that includes geckos and diurnal lizards. If the independent contrasts allometry reveals that significant differences in C_{min} occurred at the level of geckos and not in a more inclusive, diurnal clade, this would suggest that C_{min} decreased concordantly with the evolution of nocturnality in geckos and that geckos evolved a greater capacity for sustained locomotion (greater MAS) at low temperature.

Material and Methods

Animals

We used five adult males of *Phrynosoma douglassii* (mean ± SD = 4.5 g ± 0.66 g) collected at the Shasta-Trinity National Forest, Siskiyou County, California (California collector's permit 7162 to K.A.) and five adult males of *Coleonyx variegatus* (4.2 g ± 0.58 g) purchased from a commercial collector in April 1992. Body temperatures of diurnally active *P. douglassii* (mean ± SE = 34.9° ± 0.64°C; Pianka and Parker 1975) are close to the temperatures they prefer in the laboratory (35°C; Prieto and Whitford 1971). In contrast, individuals of *C. variegatus* are active at night with body temperatures as low as 15°C (mean = 24.8°C; Brattstrom 1965), substantially lower than the temperatures they prefer in the laboratory (mean ± SE = 32.8° ± 1.26°C; Dial and Grismer 1992). During the study, the lizards lived individually in plastic terraria in an environmental room (24°C). Diurnal lizard terraria had heat lamps. Nocturnal lizard terraria had heat strips that provided a thermal gradient of 25°–40°C and direct ultraviolet lighting from a fluorescent tanning lamp. Lights and heat strips were on a 13-h on : 11-h off cycle. The terraria had a sandy gravel substrate. The lizards ate a diet of mealworms, crickets, and vitamin/mineral supplement, and drank water ad lib.

Oxygen Consumption Measurements

We exercised the lizards in a miniature treadmill-respirometer (Herreid et al. 1981). Air leaving the chamber was dried with Drierite, and CO_2 was removed with Ascarite. The O_2 concentration was monitored by an O_2 analyzer (Ametek S3A) interfaced with a personal computer (Macintosh II) and data-acquisition hardware (NBMIO-16 Board, National Instruments) and software (LabView version 2, National Instruments). The precision of the O_2 analyzer is $\pm0.001\%$, whereas the deflections used for measurements were greater than 0.01%. Over the entire run, we sampled O_2 concentration every 10 s and recorded the mean of 100 O_2-concentration samples taken at 1 kHz. Response time of a flow-through system depends on the effective volume of the system and on the flow rate, so we chose a flow rate (50 mL min^{-1}) that minimized response time (<10 s to full deflection) while yielding a deflection greater than 0.01% at steady state. We calculated mass-specific, steady state O_2 consumption ($\dot{V}O_2$) from the O_2 concentration values (Withers 1977). Lizards fasted before trials, and we placed them in the treadmill chamber for a 30-min equilibration period, which was sufficient for body temperatures to reach 25°C. We adjusted the length of the chamber to slightly greater than body length to ensure steady locomotion rather than intermittent sprinting. A plastic-bristled brush provided aversive stimulus at the rear of the chamber. We used a dark screen in the front to simulate a refuge, which seemed to be a positive stimulus. The brush and screen, combined with bright fiber-optic lighting from the sides of the chamber, seemed to contribute greatly in obtaining steady runs. We also used a lightweight metal prod to tap on the tail or hind leg if the brush was not sufficient. Treadmill speeds ranged from 0.03 to 0.3 km h^{-1}. Before measurement, we had each lizard do numerous practice trials of walking on the treadmill. The lizards' treadmill locomotion improved dramatically over the course of these practice trials as they learned to walk steadily at the front of the treadmill. After data collection began, we terminated a trial if an animal repeatedly bumped against the brush, struggled, or refused to walk for more than 10 s.

The calculation of aerobically submaximal $\dot{V}O_2$ for an individual at a single speed was made from the mean of steady state $\dot{V}O_2$ during the last 3 min of at least 10 min of continuous locomotion. We defined a steady state as a window of 3 min of $\dot{V}O_2$ data through which a regression ($n = 18$ mean values from each 10-s interval) on time had a slope of less than 0.05% O_2 min^{-1} and did not include an obvious peak in $\dot{V}O_2$. Most trials included more than 20 min of continuous locomotion. At the two highest speeds, some individuals were not able to sustain locomotion for more than 6 min; however, these speeds were not included in the calculation of C_{min} since they were above the MAS. We used three measurements for each individual at each speed, which represents 135 runs (over seven submaximal speeds) for *Coleonyx* and 90 runs (over three submaximal speeds) for *Phrynosoma*.

MAS and C_{min}

The typical relationship between aerobically submaximal $\dot{V}O_2$ and running speed is a linear equation (Fig. 1; Autumn et al. 1994; see Gatten et al. [1992] for review): $\dot{V}O_2 = y_0 + (C_{min} \times speed)$, where y_0 is the y-intercept of the $\dot{V}O_2$ versus speed curve (or idling cost) and C_{min} is the slope. We defined the maximum rate of O_2 consumption, or $\dot{V}O_{2max}$, for each individual of each species as the mean $\dot{V}O_2$ attained when an increase in speed resulted in no significant increase in $\dot{V}O_2$. The MAS is the speed at which $\dot{V}O_{2max}$ is attained.

Statistical Analysis

In order to identify the MAS for each individual, we first calculated linear regressions of $\dot{V}O_2$ on speed for the lowest three speeds, and then by sequentially including higher speeds, we compared the fit of the data to the regressions. We selected MAS as the speed above which the r^2 of the regression decreased. This method agreed closely with a visual analysis of the data. We used nonparametric tests (Mann-Whitney) to compare y-intercept, $\dot{V}O_{2max}$, C_{min}, and MAS between species. We used the program StatView version 4.5 (Abacus Concepts 1989) on PowerBook 5300 (Apple) and Power Center 150 (Power Computing) computers.

Phylogenetic Analysis

Regressions using standardized, phylogenetically independent contrasts of C_{min} on body mass followed the protocol of Felsenstein (1985) and Garland et al. (1992). We used the computer program CAIC version 2.0.0 (Purvis and Rambaut 1995) to calculate independent contrasts. Phylogenetic relationships for independent contrasts were taken from Kluge (1987), Estes et al. (1988), and Garland (1994). We used C_{min} and body mass values for adults of 18 diurnal lizard species reported by John-Alder et al. (1986). For some species, John-Alder et al. (1986) report C_{min} from both large and small individuals. It is not the focus of this study to address the issue of intraspecific allometry or to study the effects of ontogeny on allometry; therefore, we excluded the measurements with low mass if they differed more than 10-fold from the measurements with the higher mass(es), since these might not be adults. We used a model of equal branch lengths and regression through the origin (Garland et al. 1992) to calculate allometries of standardized independent contrasts of log C_{min} versus log body mass. To compare values of C_{min} in diurnal and nocturnal lizards, we plotted the independent contrasts allometry using only diurnal lizards (John-Alder et al. [1986] and our data for *P. douglassii*). We then recalculated the independent contrasts allometry by including the only existing data from nocturnal lizards (geckos): *Teratoscincus przewalskii* (Autumn et al. 1994) and *C. variegatus* (present study). We then plotted the contrasts

between the two geckos and between the gecko clade (Gekkonoidea) and its sister taxon (Autarchoglossa) on the independent contrasts allometry for diurnal lizards. To check the assumptions of independent contrasts and linear regression, we searched for correlations and patterns in plots of residuals versus independent contrasts in mass and independent contrasts in C_{min}. There was no significant correlation ($P > 0.05$) or pattern evident in any of these plots, which indicates that the assumptions were supported.

Results

MAS and C_{min}

The mean ($n = 5$) C_{min} in *Coleonyx variegatus* was 1.49 mL O_2 g^{-1} km^{-1} (± 0.08 SE), only 58% of that in *Phrynosoma douglassii* (2.55 mL O_2 g^{-1} km^{-1} [± 0.32 SE]; Fig. 3), which was a highly significant difference (Mann-Whitney $U = 0$; $n = 10$; $P = 0.009$). Regressions of $\dot{V}O_2$ versus speed for individuals of the two species had r^2s between 0.89 and 1.00, using 21 measurements over seven speeds per individual in *C. variegatus* and nine measurements over three speeds in *P. douglassii*. The mean y-intercept was 0.16 mL O_2 g^{-1} h^{-1} (± 0.01 SE) in *C. variegatus* and 0.17 mL O_2 g^{-1} h^{-1} (± 0.03 SE) in *P. douglassii*. The mean $\dot{V}O_{2max}$ was 0.50 mL O_2 g^{-1} h^{-1} (± 0.03 SE) in *C. variegatus* and 0.44 mL O_2 g^{-1} h^{-1} (± 0.04 SE) in *P. douglassii*. There were no significant differences in y-intercept ($U = 10$; $n = 10$; $P = 0.6$) or $\dot{V}O_{2max}$ ($U = 7$; $n = 10$; $P = 0.25$) between species. Therefore, as a result of its lower C_{min}, mean MAS in *C. variegatus* was 0.23 km h^{-1} (± 0.03), 2.3 times the mean MAS in *P. douglassii* (0.10 km h^{-1} [± 0.004 SE]; Fig. 3; $U = 0$; $n = 10$; $P = 0.009$).

Standard Allometric Analysis

C_{min} in *P. douglassii* fell within the 95% forecast confidence limits of the mass-specific allometry for diurnal lizards, and C_{min} in *C. variegatus* fell below the 95% forecast confidence limits (Fig. 4). The new allometry for diurnal lizards including *P. douglassii* (log C_{min} [J kg^{-1} m^{-1}] $= 1.08 - 0.28$ log mass [kg]; $r^2 = 0.85$; $P < 0.0001$) is identical to the allometry of John-Alder et al. (1986).

Phylogenetically Independent Allometric Analysis

The slopes of the standard (slope $= -0.28$) and phylogenetically independent (slope $= -0.32$) allometries for diurnal lizards (including *P. douglassii*) were very similar. The phylogenetically independent contrast between nocturnal geckos and Autarchoglossa (Fig. 5) fell far below the 95% confidence limits for the diurnal independent contrasts allometry, which supports the hypothesis that the nocturnal geckos have derived a low C_{min} (Fig. 6). The independent contrasts within the geckos
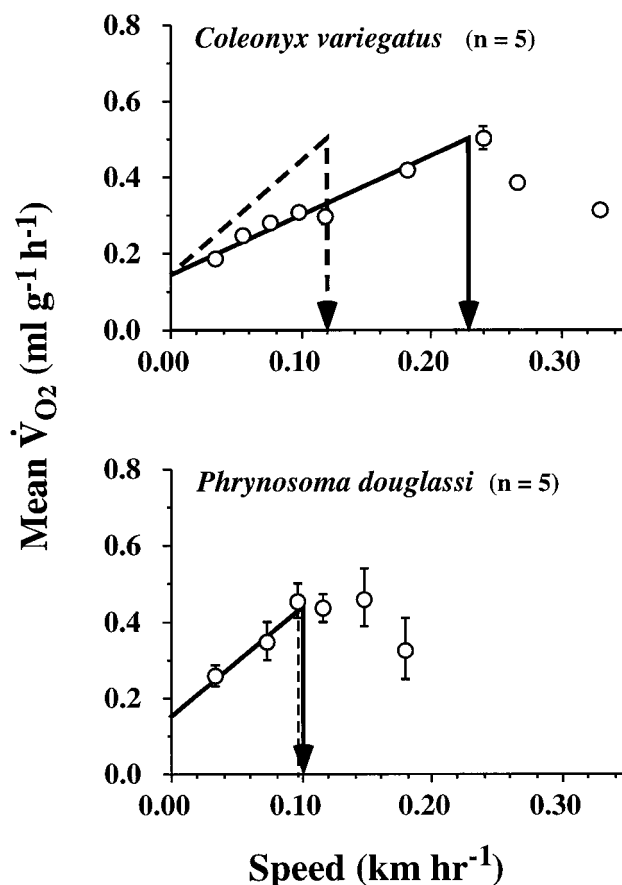

Figure 3. Mass-specific steady state $\dot{V}O_2$ during treadmill exercise in diurnal (*Phrynosoma douglassii*) and nocturnal (*Coleonyx variegatus*) lizards. Circles represent species means (± 1 SE) of $\dot{V}O_2$ at each speed. The slopes of the solid lines relating aerobically submaximal $\dot{V}O_2$ and speed represent the observed C_{min}. Dashed lines represent C_{min} and MAS predicted by allometry for a diurnal lizard of the same mass, y-intercept, and $\dot{V}O_{2max}$ (John-Alder et al. 1986). C_{min} in *C. variegatus* was only 58% of that predicted by allometry, yielding a MAS 230% of predicted values, while C_{min} and MAS values in *P. douglassii* were similar to predicted values. C_{min} was significantly lower in *C. variegatus* than in *P. douglassii*. The relatively low mean C_{min} in *C. variegatus* yielded a relatively high mean MAS (*arrows*) compared with *P. douglassii*, which had a comparable $\dot{V}O_{2max}$.

fell near the 95% confidence limits for the diurnal independent contrasts allometry.

Discussion

The original discovery that a low C_{min} in frog-eyed geckos (*Teratoscincus przewalskii*; Autumn et al. 1994) increases MAS at low, nocturnal temperatures raises the question of whether *T. przewalskii* is a uniquely adapted lizard or whether a low C_{min}, increased MAS, and nocturnality evolved concordantly in geckos as a group (the nocturnality hypothesis). Direct comparisons of diurnal and
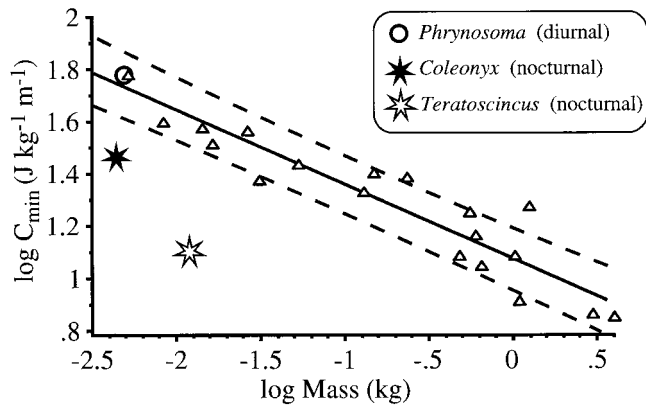
Figure 4. C_{min} as a function of body mass for lizards. Triangles represent literature values for diurnal lizards (John-Alder et al. 1986). C_{min} for *Phrynosoma douglassii* (*circle*) falls within the 95% forecast confidence limits (*dashed lines*) of the diurnal lizard allometry (*solid line*). C_{min} for the nocturnal lizards *Coleonyx variegatus* (this study; *solid star*) and *Teratoscincus przewalskii* (Autumn et al. 1994; *open star*) fall far below the 95% forecast confidence limits.

nocturnal lizards of the same body mass under the same experimental conditions by Farley and Emshwiller (1996) and in this study support the hypothesis of nocturnality. For example, in the present study there was a striking difference in C_{min} between the nocturnal and diurnal species: C_{min} in *Coleonyx variegatus* was only 58% of that of the *Phrynosoma douglassii*. As a result, MAS of *C. variegatus* was 2.3 times that of the diurnal *P. douglassii* (Fig. 3). Since C_{min} in *P. douglassii* and *Eumeces skiltonianus* (Farley and Emshwiller 1996) is within the range of values predicted by the standard allometry (Fig. 4; John-Alder et al. 1986; Autumn et al. 1994), there seems to be a similar functional relationship between body mass and C_{min} in small and large diurnal lizards. This is important in comparisons involving nocturnal and diurnal lizards because, while diurnal lizards come in all sizes (ca. 1 g to 150 kg), nocturnal lizards tend to be small (ca. 1–300 g). The allometric analysis also showed that C_{min} in *C. variegatus* was very low in comparison to that in diurnal lizards (Fig. 4). C_{min} in the gecko was comparable to that of diurnal lizards more than 10 times as heavy. This supports the nocturnality hypothesis. Without factoring out phylogenetic similarity among diurnal species, however, we cannot evaluate the statistical significance of the difference in C_{min} between *C. variegatus* and diurnal lizards. For example, if the smallest diurnal lizards in our analysis belong to a clade that has marginally high C_{min}, which decreases the allometric slope, then the seemingly low C_{min} in *C. variegatus* and *T. przewalskii* could be a historical artifact. To solve this problem, we factored out differences in C_{min} caused by both mass and historical noise to focus on the signal of interest—nocturnality.

Phylogenetically Independent Allometry

The slope of the phylogenetically independent allometry for diurnal lizards (−0.32; Fig. 6) was similar to that of the standard allometry for diurnal lizards (−0.28; Fig. 4). This suggests that at the phylogenetic level of lizards (Squamata) there is a functional dependence between mass and C_{min} that in most cases transcends phylogeny. In other words, body mass, not history, explains most of the variation in C_{min} in diurnal lizards. The independent contrasts analysis also demonstrates that the value of C_{min} in *P. douglassii* was not atypical of diurnal lizards and is valid for comparison with *C. variegatus*.

The relationship between body mass and C_{min} has remained remarkably constant over the approximately 200 million yr of squamate evolution. There are at least two interpretations of this conservative allometric pattern. One is the view of allometry as a line of adaptation (Harvey and Pagel 1991), where C_{min} is free to vary but, for a given mass, selection favors values near the line. In another view, allometry is interpreted as a line of constraint. In other words, there is something about body mass that mechanistically limits variation in C_{min}. The former view could be termed a functionalist explanation, while the latter could be termed a structuralist explanation (Lauder 1982; Reippel 1988). It is likely that both explanations have some degree of validity. The functionalist explanation requires ongoing convergent or parallel selection that results in evolution of all populations of each species toward an optimal mass-specific value of C_{min}. This explanation for allometry predicts that there should be ongoing stabilizing selection for individuals to have values of mass and C_{min} that lie close to the allometry (Fig. 4). Dramatic exceptions to the allometric rule such as the nocturnal geckos *T. przewalskii* (Autumn et al. 1994) and *C. variegatus* (present study), which have evolved a much lower C_{min} than predicted, challenge the functionalist explanation because evolution has not proceeded in the direction expected if the allometry is a line of optimality. In order to explain a deviation from the allometry, the functionalist interpretation becomes more complicated; patterns such as those in *C. variegatus* would require ongoing directional selection to balance stabilizing selection maintaining the optimal allometric pattern.

The structuralist explanation requires no selection to maintain the allometric pattern and is therefore a more parsimonious null model. This interpretation is supported by the striking allometric similarity of mass and C_{min} in lizards, birds, mammals, and invertebrates (Full 1989). The structuralist interpretation of allometry as a line of constraint will be tested by progress in the study of mechanisms underlying the relationship between body mass and the energetic cost of locomotion. From this perspective, points near the independent contrasts allometry (Fig. 6) represent taxa that retain the ancestral lizard pattern and require no adaptive explanation (Harvey and Pagel 1991). Deviations from the independent contrasts allometry that increase performance represent an adaptive evolutionary change in C_{min}.

Evolution of C_{min}

Our results suggest that there has been an adaptive evolutionary change in C_{min} within geckos. C_{min} is much lower in nocturnal
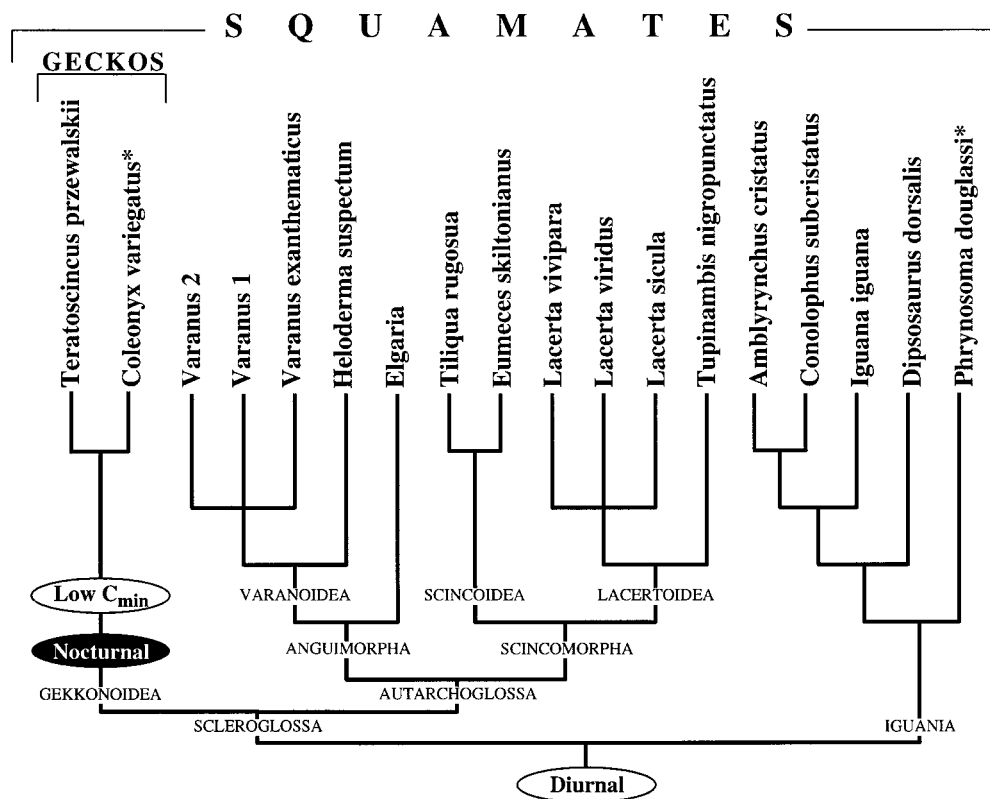
Figure 5. Cladogram of diurnal and nocturnal lizard species for which there are measurements of C_{min}. The topology follows Kluge (1987), Estes et al. (1988), and Garland (1994). Polytomies should be interpreted as soft (in other words, there is not enough information to resolve relationships among taxa, but we do not assume that the taxa split simultaneously). Branch lengths represent order of branching, not absolute time. *Asterisks* denote species from this study.

geckos than in diurnal lizards of similar mass under the same experimental conditions (Fig. 3) and in diurnal lizards of a wide range of body mass after the effects of mass and phylogeny have been considered (Fig. 6). The independent contrasts between the geckos (Gekkonoidea) and their sister taxon (Autarchoglossa) fell below the 95% confidence limit of the independent contrasts allometry. This suggests that C_{min} decreased in the geckos. However, it is necessary to consider the possibility that *T. przewalskii* is unique among geckos in having a very low C_{min}. If this were true, this extremely low value could be solely responsible for the significant difference between Gekkonoidea and Autarchoglossa. The data do not support this possibility. The independent contrasts between *T. przewalskii* and *C. variegatus* fell near the 95% confidence limit of the independent contrasts allometry, indicating that there was no substantial difference in C_{min} between *T. przewalskii* and *C. variegatus.* Moreover, the independent contrasts between *C. variegatus* (excluding *T. przewalskii*) and Autarchoglossa fell far below

the 95% confidence limit. This implies that a low C_{min} is a shared, derived character of Gekkonoidea, not a unique character of one species. Measurements of C_{min} in other gecko species will further test this hypothesis.

Increased Performance at Low Temperature

The low C_{min} in the geckos increases endurance capacity at low temperature and partially offsets the thermal handicap of nocturnality. This supports our hypothesis that there has been an evolutionary increase in locomotor performance capacity at low temperature in nocturnal lizards. Since temperature affects aerobic metabolism dramatically, it is reasonable to consider the effects of temperature on C_{min}. This is an important point because comparisons of C_{min} among species may involve measurements taken at different temperatures. There is strong evidence that C_{min} is temperature-independent in lizards (John-Alder and Bennett 1981; John-Alder et al. 1983; Bennett and John-Alder 1984; Autumn et al. 1994). However, $\dot{V}O_{2max}$ is strongly temperature-dependent. The thermal sensitivity of the MAS is due to the thermal sensitivity of $\dot{V}O_{2max}$, not to any change in C_{min}. A decrease in C_{min} is adaptive for activity at low temperature because it can offset the thermally induced decrease in $\dot{V}O_{2max}$. Since $\dot{V}O_{2max}$ in *P. douglassii* was nearly identical to that in *C. variegatus,* the dramatic difference in MAS was solely because of a low C_{min} in *C. variegatus.* This difference
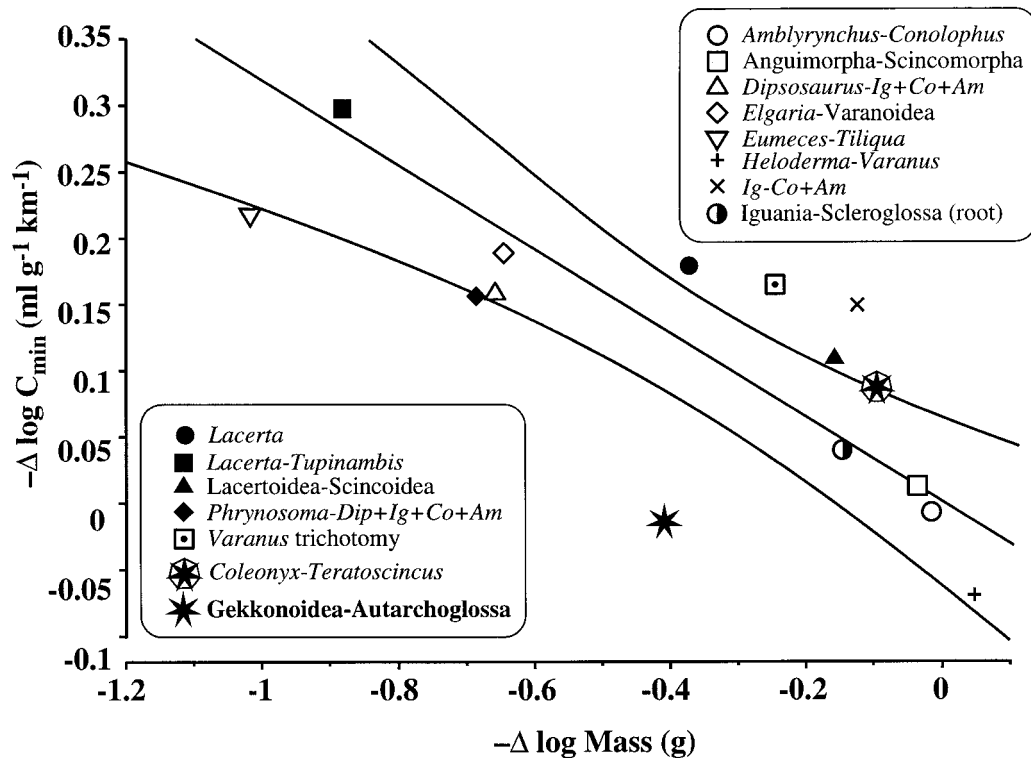
Figure 6. Phylogenetically independent contrasts in log C_{min} (Δ log C_{min}) versus independent contrasts in log body mass (Δ log mass). Phylogenetically independent contrasts are differences in character value (log mass and log C_{min}) between sister taxa. The solid line represents a regression through the origin (slope = -0.32; R^2 = 0.89) of $-\Delta$ log C_{min} on $-\Delta$ log mass in diurnal lizards only, which is a phylogenetically independent allometry showing a correlation between evolutionary change in log mass and log C_{min} in diurnal lizards. The dashed lines represent the 95% confidence limits of the diurnal lizard independent contrasts allometry. Contrasts (*geometric shapes*) that fall within the confidence limits represent sister taxa for which a difference in mass is accompanied by a typical difference in C_{min}. Abbreviated names of taxa are Am, *Amblyrynchus;* Dip, *Dipsosaurus;* Ig, *Iguana;* and Co, *Conolophus.* The independent contrasts between nocturnal geckos and Autarchoglossa (*star*) fell far below the 95% confidence limits for the diurnal independent contrasts allometry, which supports the hypothesis that the nocturnal geckos have derived a low C_{min}.

is very striking when observing the animals moving on a treadmill at their MAS at 25°C. The MAS of the gecko (0.24 km h^{-1}) is a speed that seems quite typical of small lizards moving in nature, while the MAS of *P. douglassii* (0.096 km h^{-1}) is so slow that we doubt it could forage effectively at 25°C.

Although the geckos have evolved an improved locomotor capacity at low temperature, they are still suboptimal at night because the thermal optimum for $\dot{V}O_{2max}$ does not seem to coincide with nocturnal temperatures (Autumn and Full 1994). $\dot{V}O_{2max}$ is reduced at low temperature in both diurnal and nocturnal lizards (Bennett 1982; Gatten et al. 1992; Autumn

and Full 1994; Autumn et al. 1994). It is interesting to note that a low C_{min} is potentially an advantage at all body temperatures. An animal with a reduced C_{min} will have an increased MAS at any given temperature. Measurements of C_{min} in secondarily diurnal geckos that are active at high temperature, such as species of *Rhoptropus* or *Phelsuma,* will test the advantages of low C_{min} under different environmental conditions. If secondarily diurnal geckos have C_{min} typical of ancestrally diurnal lizards, this would suggest a trade-off between fuel economy and another performance-related variable that is important for diurnal activity.

This study underscores the need to consider phylogeny in comparative physiology. Geckos form an extremely speciose clade (> 800 species) and most geckos are small (< 30 g). If C_{min} is unusually low in geckos as a group, a standard allometric analysis of a representative sample of lizard species would erroneously show that mass had little effect on C_{min} because of the large number of small lizards (geckos) with low C_{min} (Fig. 7A). A phylogenetically correct analysis can solve this problem by separating the ancestral lizard pattern from the derived gecko pattern (Fig. 7B), clearly showing that the geckos do not follow the same allometric relationship as diurnal lizards.

As a result of this study, our preliminary conclusion is that geckos evolved a low C_{min}, which increases locomotor performance at low temperature and is adaptive for nocturnal activity. The generality of this pattern will depend on future data on C_{min} in a representative sample of geckos. Also, there are other nocturnal squamates besides geckos. Two other
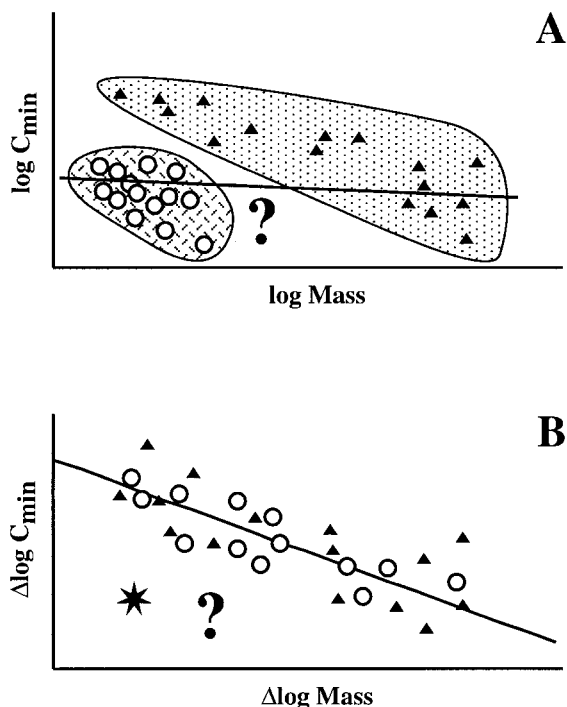
Figure 7. A, Hypothetical allometry using real data of C_{min} and body mass in diurnal lizards (*triangles*) from John-Alder et al. (1986) and hypothetical data for nocturnal lizards (*circles*). If nocturnal lizards have relatively low C_{min} for their body mass, a standard allometry for lizards as a group (*solid line*) might have an erroneously low slope. Since most nocturnal lizards form a clade (Gekkonoidea), grouping according to phylogeny (*shaded regions*) provides a way to separate the ancestral lizard pattern from the derived gecko pattern. B, Hypothetical phylogenetically independent allometry of C_{min} and body mass in diurnal lizards (*triangles*) and nocturnal geckos (*circles*). This hypothetical analysis shows how a phylogenetic approach could be used to separate historical patterns from the effects of body size. If the most recent common ancestor of living geckos evolved an unusually low C_{min} (*star*), evolution of C_{min} within geckos may still follow a typical allometric pattern (*solid line*). More data on nocturnal lizards are required before this hypothesis can be tested.

groups that contain many nocturnal forms are the skinks and the snakes. Our results have little direct bearing on the evolution of nocturnality in those groups; however, our results set the stage for future comparisons between diurnal and nocturnal species that differ greatly in mass and evolutionary history. The low C_{min} measured in the nocturnal snake *Crotalus cerastes* (Secor et al. 1992) may support the generality of the nocturnality hypothesis but is complicated by the sidewinding mode of locomotion in this species. There are two logical steps that follow from our results: further research on other gecko species to test the nocturnality hypothesis in the clade Gekkonoidea, and measurements of C_{min} in nocturnal skinks and snakes to test the generality of the nocturnality hypothesis in squamates.

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