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Ecological Morphology of Locomotor Performance in Squamate Reptiles

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INTRODUCTION

Relationships between morphology, physiology, or biochemistry, on the one hand, and behavior and ecology, on the other, have been widely documented, as this volume attests. Such relationships provide evidence that most, if not all, organisms are to some extent "adapted" to their current environment. Quantifying how well adapted an organism is, or testing the biological and statistical significance of putative adaptations, may, however, be very difficult (Brooks and McLennan, 1991; Harvey and Pagel, 1991; Losos and Miles, chap. 4, this volume). As well, many studies in ecological morphology, and in the conceptually related fields of physiological ecology (Feder et al., 1987) and comparative biochemistry (Hochachka and Somero, 1984), have ignored the crucial intermediate step of organismal performance (Arnold, 1983; Huey and Stevenson, 1979; Losos, 1990b) when trying to correlate morphology with ecology. In this chapter, we review the literature pertaining to the ecological morphology of locomotor performance in reptiles and relate this knowledge to current paradigms and analytical techniques in organismal and evolutionary biology. We will argue that both maximal whole-animal performance abilities (what an animal can do when pushed to its limits; generally measured in the laboratory, and not to be confused with *efficiency*; see Gans, 1991; Lauder, 1991) and behavior (what an animal actually does when faced with behavioral options; best measured in the field) must be considered when attempting to understand the mechanistic bases of relationships between morphology and ecology.

Locomotion is in many ways ideally suited for studies of ecological morphology. Most behavior involves locomotion, and measures of both locomotor performance (e.g., speed, stamina) and its morphological bases (e.g., limb length, heart size) come easily to mind. Some reptiles are good subjects for measurement of locomotor performance in the laboratory (e.g., with race tracks or treadmills), for quantifying its morphological, physiological, and biochemical bases, and for

demographic study and behavioral observation in the field. Measurement of performance is crucial, and reptiles are certainly easier subjects than are some other vertebrate groups, such as birds or bats (Ricklefs and Miles, chap. 2, this volume; Norberg, chap. 9, this volume).

The Morphology → Performance → Fitness Paradigm

"Not infrequently, performance characteristics, measured as maximal speed or endurance, make the difference between eating and being eaten" (Tenney, 1967, p. 1–7). The foregoing quotation certainly contains some truth, but actual data indicating the frequency of "close encounters of the worst kind" between predators and prey are few and far between (cf. Castilla and Bauwens, 1991, p. 78; Christian and Tracy, 1981; Hertz et al., 1988; Jayne and Bennett, 1990b).

Studies of ecological morphology implicitly concern fitness and adaptation. Within populations, individual variation in morphology may be related to variation in Darwinian fitness; among populations and higher taxa, morphological variation may indicate adaptation to different lifestyles. Arnold (1983) proposed a conceptual and statistical—and hence operational—framework for using data on individual variation to study adaptation within populations (fig. 10.1). This paradigm addresses the question of whether natural selection is currently acting on morphology or performance within a single population. Arnold's (1983) discussion considered multiple morphological characters and multiple measures of performance, as well as correlations within these two levels. He pointed out that multiple regression and path analysis could be used to estimate and test the significance of performance gradients (quantifying the effects of morphology on performance), which can be studied in the laboratory (but see Wainwright, chap. 3, this volume), and fitness gradients (quantifying the effects of performance on fitness), which require field studies (see also Emerson and Arnold, 1989; Wainwright, 1991).

This perspective suggests that intrapopulational variation in morphology may have significant influences on fitness only to the extent that it affects performance. Measures of organismal performance capacities thus become central (cf. Bennett, 1989; Bennett and Huey, 1990; Emerson and Arnold, 1989; Pough,

Morphology → Performance → Fitness
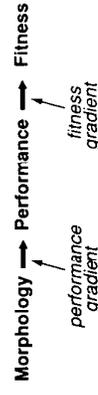
performance gradient   fitness gradient

FIGURE 10.1 Simplified version of Arnold's (1983) original paradigm.

1989), and measures of locomotor performance fall easily into the paradigm. The focus on organismal performance as pivotal is in sharp contrast to many previous studies of locomotor ecology, in which the starting point (and sometimes the ending point) has been measurement of limb proportions (see "Case Studies" below). Moreover, the realization that it is easier to study one or the other rather than both gradients simultaneously, and that both parts of the equation are of interest, has stimulated research.

Physiological and biochemical traits may be included within the category of "morphology," and we will subsequently use morphology as shorthand for all three types of traits. This is not to imply that morphology, physiology, and biochemistry are equivalent, nor are we trying to deny the distinction between "form" and "function." The point is simply that all three types of traits are (generally) at a level of biological organization below the whole-organism, and all may influence organismal performance. Calling all three types of traits "morphology" serves to emphasize that similar tools and approaches are useful for studying their effects on organismal performance (cf. Wainwright, 1991).

Arnold's (1983) paradigm was designed specifically to interface with multivariate quantitative genetics theory (Lande and Arnold, 1983). In the quantitative genetic framework (e.g., Boake, 1994; Brodie and Garland, 1993; Falconer, 1989), adaptive phenotypic evolution consists of two parts: natural selection, which is a purely phenotypic phenomenon, and genetic response, which involves inheritance. Some do not like this separation of selection and inheritance (e.g., Endler, 1986), but we agree with Lande and Arnold (1983) that it has considerable operational advantages in allowing the two elements to be studied independently. It also emphasizes that selection may be futile; if a trait is not heritable, then selection cannot lead to or improve adaptation.

How Does Behavior Fit into the Paradigm?

The place of behavior in the paradigm of figure 10.1 is ambiguous. Arnold (1983) did not mention behavior as a distinct level; subsequently, however, Emerson and Arnold (1989; also Schluter, 1989) have included behavior within the category of morphology. We offer an alternative categorization, as depicted in figure 10.2 (modified from Garland, 1994a).

Many biologists imagine that selection acts most directly on what an animal actually does in nature, that is, its behavior. Performance, on the other hand, as defined operationally by laboratory measurements, generally indexes an animal's ability to do something when pushed to its morphological, physiological, or biochemical limits. (Whether animals routinely behave at or near physiological limits under natural conditions is an important empirical issue for which precious few data exist: Daniels and Heatwole, 1990; Dial, 1987; Garland, 1993;

Physiology
Morphology → Performance → Behavior → Fitness
Biochemistry

FIGURE 10.2 Expansion of Arnold's (1983) paradigm to include behavior, as proposed by Garland (1994a).

Garland et al., 1990a; Gatten et al., 1992; Gleason, 1979a; Hertz et al., 1988; MacArthur, 1992; Morgan, 1988; Pough et al., 1992; Seymour, 1982, 1989; van Berkum et al., 1986, Wyneken and Salmon, 1992.) Thus, morphology limits organismal performance, which in turn constrains behavior, and natural and sexual selection act most directly on behavior—what an animal actually does (Garland, 1994a). This modification of Arnold's (1983) original paradigm adds one more level of analysis and places specific emphasis on behavior as the focus of selection. Behavior is seen as a potential "filter" between selection and performance (Garland et al., 1990b).

Further Extensions of the Paradigm

Inserting behavior between performance and fitness seems relatively straightforward (fig. 10.2). But this addition does not necessarily mean the paradigm is complete or general. Many more possible links can be imagined, and a relatively simple chain rapidly becomes a complicated web (e.g., fig. 10.3).

In particular, habitat, broadly defined, is another important factor which may influence behavior, performance capabilities, and even morphology (see also Dunson and Travis, 1991; Huey, 1991). For example, availability of perches or basking sites, their size, and their distribution may affect both what an animal does (e.g., Adolph, 1990b; Grant, 1990; Moermond, 1986; Pounds, 1988; Waldschmidt and Tracy, 1983; see discussion of the "habitat matrix" model below) and what it is capable of doing (e.g., sprint speed in lizards is affected by perch diameter and substrate: Carothers, 1986; Losos and Sinervo, 1989; Losos et al., 1993; Miles and Althoff, 1990; Sinervo and Losos, 1991). Temperature is a habitat characteristic that may affect performance indirectly through its effects on various physiological processes, and by having direct influences on behavior, such as the switches in defensive behavior at low body temperature that occur in some lizards and snakes (Arnold and Bennett, 1984; Crowley and Pietruszka, 1983; Hertz et al., 1982; Mautz et al., 1992, Van Damme, Bauwens et al., 1990; Schieffen and de Queiroz, 1991). Temperature affects locomotor performance both in absolute terms (Bauwens et al., in press; Bennett, 1990; Garland, 1994b) and, to a lesser extent, relative to other individuals or species. Individual differences in locomotor performance are consistent across temperatures (i.e., fast individuals tend to be fast at all temperatures); but not perfectly so (references in

Interspecific
Interactions

Morphology → Performance → Behavior → Fitness
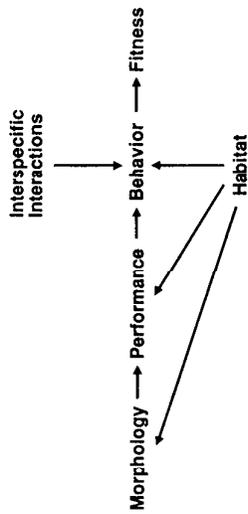
Habitat

FIGURE 10.3 Inclusion of some other factors that may affect elements of Arnold's (1983) paradigm. Habitat characteristics, such as temperature, may affect basic physiological and biochemical properties as well as behavior (see text). (Of course, behavior and physiology may affect an animal's body temperature; the present diagram is extremely simplified.)

Bennett, 1990; Bennett and Huey, 1990). Thus, the temperature at which an individual happens to be when it encounters a predator may affect its relative fitness (e.g., Christian and Tracy, 1981), and individual differences in thermoregulatory behavior may become crucial (cf. Christian et al., 1985; Waldschmidt and Tracy, 1983).

More subtle habitat effects are also possible. Food in the stomach (Ford and Shuttlesworth, 1986; Garland and Arnold, 1983; Huey et al., 1984; nutritional state (for experiments with mammals, see Brooks and Fahey, 1984, and Astrand and Rodahl, 1986), hydrational state (Moore and Gatten, 1989; Preest and Pough, 1987; Wilson and Havel, 1989, but see Crowley, 1985b; Gatten and Clark, 1989; Stefanski et al., 1989) as well as disease or parasite infection (Schall, 1986, 1990; Schall et al., 1982; but see Daniels, 1985b) all may affect performance ability. Hydrational (Crowley, 1987; Feder and Londos, 1984; Pough et al., 1983; Putnam and Hillman, 1977) or nutritional state may also affect activity levels, that is, behavior. Inter- and intraspecific interactions can also affect behavior in numerous ways (e.g., Fox et al., 1981; Garland et al., 1990a; Henrich and Bartholomew, 1979; Schall and Dearing, 1987; Stamps, 1984). Even hydrational or thermal conditions during incubation or pregnancy can affect locomotor performance of offspring (Miller et al., 1987; Van Damme et al., 1992).

Extending the Paradigm to Population and Species Variation

The paradigm in figure 10.2 can also be applied to understand or predict a relationship between morphology and habitat use among populations or species. The logic of this extension is as follows. First, to the extent that morphological differences among individuals within populations lead to differences in performance abilities that affect fitness, then, assuming the absence of constraints (Maynard Smith et al., 1985), the most "fit" morphology should evolve within

any population (Emerson and Arnold, 1989). Second, to the extent that different morphologies function best in different habitats, then natural selection will tend to favor their evolution in the appropriate habitats. If one has an understanding of which morphologies are best suited in given habitats (based on biomechanical or functional analyses, including optimality models, or on empirical studies of natural selection within populations), then one can test the prediction that taxa have adapted to different environments (Baum and Larson, 1991; Bock and von Wahlert, 1965; Losos and Miles, chap. 4, this volume). Caution must be exercised when taking this view, however, as we have little empirical evidence that any given trait(s) in any given population will have reached its selective optimum by the time we study it (Arnold, 1987; Ware, 1982). Moreover, multiple (sub)optimal solutions, which confer equivalent fitness, may exist (Denny, chap. 8, this volume; Feder et al., 1987; Ware, 1982), depending on the shape of the fitness surface, movement from one peak to another may be difficult.

Although Arnold (1983) suggested path analysis for studying the causes (performance gradients, e.g., fig. 10.5 below) and consequences (fitness gradients) of individual variation in performance (and behavior), path analysis might also be employed to study species-level selection processes (cf. Emerson and Arnold, 1989). For example, rather than values for individuals, data points could be population, species, or clade means for morphological, performance, behavioral, or ecological traits. As components of the "fitness" of a population, species or clade (cf. Futuyma, 1986; Vrba, 1989), one might consider geographic range (cf. Jablonski, 1986), evolutionary longevity, and/or number of descendant populations or species (the latter might require paleontological information; but see Nee et al., 1992). Alternatively, some measure of a population's or of a species' "fitness" or "adaptedness" (Michod, 1986) to its current environment might also be possible, such as physiological tolerances, breadth of the Grinnellian niche (James et al., 1984), or demographic traits (e.g., population density, intrinsic rate of natural increase: cf. Baker, 1978; Birch et al., 1963). To quote Stini (1979, p. 388): "A well-adapted population would be . . . one that enjoys a relatively high probability of survival under conditions highly likely to occur." Of course, a path analysis of comparative data would require proper allowance for phylogenetic non-independence (see below). As noted by Emerson and Arnold (1989, p. 302), "there are no strong theoretical grounds for expecting similar performance topographies at the intra- and interspecific levels and there has been virtually no empirical exploration."

Does Morphology Affect Fitness Directly?

Regardless of how complicated a paradigm one wishes to consider, an outstanding conceptual and empirical issue is whether direct paths exist from morphology to fitness (fig. 10.4). Returning to the original formulation, the most

Interspecific
interactions

Morphology → Performance → Behavior → Fitness
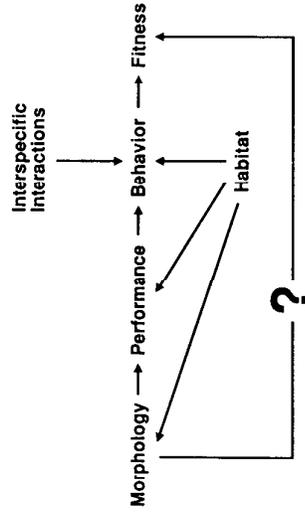
Habitat

?

FIGURE 10.4 An outstanding conceptual and empirical issue is whether any direct paths from morphology to fitness are significant (see text).

general path model is one in which all possible effects are depicted, including those directly from morphology to fitness (Arnold [1983, fig. 3] omitted these paths). Consider some hypothetical possibilities. Some individual garter snakes are born with a single or no eyes, an external heart, or a severely kinked tail or spine (Garland, 1988, pers. obs.; Arnold and Bennett, 1988, pers. comm.). These morphological deformities greatly impair locomotor capacities, which in turn limit behavioral options (as compared with "normal" individuals), and would certainly have fitness consequences in nature. In this case, a direct path from morphology to fitness seems unnecessary.

But contemplate two other examples. First, all else being equal (i.e., assuming behavior is unaffected), an albino snake will likely suffer a fitness decrement relative to a normally pigmented individual, because the former will more likely be discovered and eaten by a predator prior to its reproducing. Thus, a direct path appears to exists from morphology (pigmentation) to fitness. Alternatively, if one considers some measure of crypsis as a "performance" variable, then albinism acts through its effects on crypsis (cf. King, 1992), and performance, but not morphology, would seem to have a direct path to fitness, bypassing behavior. But, if an animal could somehow become "aware" that it was differently colored and so alter its behavior to compensate (cf. Morey, 1990), then the effect of albinism might be entirely through the performance → behavior link. Albinism would also affect thermoregulation, making it more difficult for the snake to warm by basking, hence causing it to bask for longer periods of time and increasing its exposure to predators (cf. Andren and Nilson, 1981).

Second, in many species, body size affects the outcome of intraspecific behavioral interactions (Tokarz, 1985; references in Garland et al., 1990a; Faber and Baylis, 1993). This effect may occur simply because size affects strength and stamina, and hence performance at fighting. But in some cases differences in size alone may influence decisions to fight or not, and hence may determine the out-

come of an agonistic interaction before any actual fighting occurs. This example suggests a direct effect of a morphological trait (body size) on behavior, and hence on a component of fitness (dominance rank).

Some might consider it logically impossible that morphology can affect fitness other than through its effects on organismal performance (and hence behavior). The idea is that form only matters if it affects function and hence performance; otherwise, morphological variation is selectively neutral. We would prefer to consider the absence of direct morphology → fitness paths as an hypothesis, subject to empirical test. Such tests might involve measurement of vertebral numbers (in snakes) or limb length (in lizards), as well as locomotor performance and survivorship (cf. Arnold and Bennett, 1988; Jayne and Bennett, 1990b; Tsuji et al., 1989). A significant path from morphology to (a component of) fitness (fig. 10.4) would indicate either a direct effect of morphology on fitness or the presence of some unmeasured (latent) performance variable.

LEVELS AND METHODS OF ANALYSIS

Interpopulation

Although analyses relating morphology, performance, behavior, and fitness (broadly defined) most commonly involve interspecific comparisons (to be discussed below) or, more recently, individual variation, studies of interpopulation differences are essential to evolutionary analyses (Garland and Adolph, 1991; James, 1991). Most previous studies of population (geographic) variation focus on morphometric or allozymic characters (e.g., Zink, 1986), although studies of variation in mitochondrial DNA are now common (e.g., Avise et al., 1987; Lamb et al., 1989). Consequently, phylogenetic analyses of population differentiation cannot be far off (cf. Schuter, 1989; Snell et al., 1984), and we encourage such studies of population differences in locomotor performance and its correlates. If possible, such studies should include a "common garden" approach, in which animals are raised in the laboratory for at least one generation to maximize the probability that observed phenotypic differences are actually genetically based (Garland and Adolph, 1991). Common garden controls are important for studies of different species as well, although most biologists seem less concerned at this level. Population differences in locomotor performance may be consistent across years (Huey and Dunham, 1987), but year-to-year variation in performance exists (Huey et al., 1990) and may confound attempts to correlate morphology with ecology (cf. Wiens and Rotenberry, 1980).

Intrapopulation: Individual, Ontogenetic, and Sexual Variation

Arnold (1983, fig. 10.1) considered studies of individual variation within populations, including effects of morphology on performance (e.g., mechanistic physiology) and the effects of performance on fitness (e.g., direct studies of natu-

ral selection in the wild). Quite a few such studies of reptilian locomotion have been completed since 1983. A major conclusion of these studies is that measures of locomotor performance show substantial and repeatable individual variation within single populations (Bennett, 1987; Bennett and Huey, 1990; Huey et al., 1990; Jayne and Bennett, 1990a; see also Djawdan, 1993; Friedman et al., 1992, on mammals). This variation and repeatability is, of course, a prerequisite for attempts to quantify relationships between morphology and performance (performance gradients) or between performance and behavior, fitness, or ecology (e.g., fitness gradients).

One advantage of studying individual variation is that phylogenetic effects are not a concern. So, for example, the effects of body size can be studied by examining an ontogenetic series (e.g., Garland, 1984, 1985; Jayne and Bennett, 1990a; Pough, 1977, 1978) rather than multiple species. Similarly, the mechanistic correlates of performance variation can be studied (e.g., Garland, 1984, 1985; Garland and Else, 1987; Losos et al., 1989; Tsuji et al., 1989) without concern that phylogenetic effects may confound the results (cf. Losos, 1990a, b, c).

Quantitative Genetic Analysis

Individuals within a population may not provide statistically independent data points, because they are related to varying extents. Quantitative genetics uses this fact to partition observed phenotypic variances and covariances into genetic (due to inheritance, which is analogous to phylogenetic descent; cf. Lynch, 1991) and environmental sources, each of which can be more finely partitioned (Boake, 1994; Brodie and Garland, 1993; Falconer, 1989; Garland, 1994a).

Quantitative genetic analyses are not a traditional part of ecological morphology. They must become an integral part, however, if we are to move towards an understanding of the mechanisms of microevolution. We will not consider quantitative genetic analyses of reptilian locomotor performance in detail. Only a few studies have been completed, all on garter snakes (Thamnophis) or lizards (Sceloporus, Lacerta; reviews in Bennett and Huey, 1990; Brodie and Garland, 1993; Garland, 1994a). All studies to date have relied on analyses of presumed full-sibling families to estimate heritabilities. For many reptiles, gravid females can be captured in relatively large numbers in the field. After offspring are born or hatched in the laboratory, measurements of locomotor performance are made on each. Unfortunately, heritability estimates from sets of full-siblings represent neither a "narrow-sense" nor a "broad-sense" heritability; in addition, multiple paternity will lead to an underestimation of additive genetic effects in full-sibling data sets (Brodie and Garland, 1993; Falconer, 1989; Garland, 1994a; Schwartz et al., 1989). Thus, significant among-family variance in studies of full-siblings suggests heritability, but does not prove it. With one exception (Bauwens et al.,

1987), all studies to date have found significant among-family variance for measures of locomotor performance in reptiles.

Experimental Approaches

Experimental approaches can be used in several ways, for example: (1) to examine the mechanistic bases of performance variation; (2) to mimic the effects of short- or long-term changes that may occur naturally within individuals; (3) to examine the effect of conditions during development on morphology and performance abilities; and (4) to increase the range of variation in organismal performance and so increase statistical power to detect its ecological and selective importance. Experimental approaches have the advantage that they can isolate and study the effects of variation in one variable independent of correlations with other variables (cf. Lande and Arnold, 1983; Mitchell-Olds and Shaw, 1987; Slinker and Glantz, 1985; Wade and Kalisz, 1990). Experimental approaches have been underutilized for analyzing links in the morphology → locomotor performance → behavior → fitness chain (or web) and in ecomorphology in general (but see Benkman and Lindholm, 1991; Carothers, 1986; Hanken and Wake, 1991; Hillman and Withers, 1979; Huey et al., 1991; James, 1991; Jayne and Bennett, 1989; Lauder and Reilly, 1988; Ruben et al., 1987; Webster and Webster, 1988).

Causal mechanistic relationships suggested by correlative studies of individual variation in locomotor abilities (fig. 10.5: e.g., Garland and Else, 1987; Gleeson and Harrison, 1988; John-Alder, 1984a, b, 1990) can be tested with such physiological techniques as blood doping (cf. Withers and Hillman, 1988), but this has scarcely been attempted in reptiles (Gleeson, 1991). Hormonal (John-Alder, 1990; Joos and John-Alder, 1990; Moore and Marler, 1987; Moore and Thompson, 1990) or pharmacological (e.g., John-Alder et al., 1986b) manipulation to change metabolism and performance is also possible. (Levels of some hormones fluctuate rapidly in reptiles, whereas some measures of locomotor performance are quite repeatable, which suggests that the former may have little effect on the latter.) With respect to morphology, the importance of tail length and loss, toe loss, toe fringes, and skin flaps for sprinting and gliding performance in lizards and snakes has also been assessed experimentally (e.g., Arnold, 1984a; Carothers, 1986; Daniels et al., 1986; Formanicwiz et al., 1990; Huey et al., 1990; Jayne and Bennett, 1989; Losos et al., 1989; Marcellini and Keefer, 1976; Pond, 1981).

Within-individual variation in reptilian locomotor performance has been examined as a consequence of several factors, such as physical conditioning (training), feeding, reproductive state, and hormonal state. Physical conditioning studies of the type so common in mammalian exercise physiology (e.g., Brooks

automated as easily as they can with mammals. An outstanding issue is the extent to which "natural training" occurs in the wild (Burghardt, 1984; Garland et al., 1987). Acclimation and acclimatization of reptilian locomotor performance has been studied only rarely (Gatten et al., 1988; Hailey and Davies, 1986; Kaufmann and Bennett, 1989; Payne and Gatten, 1988), as has seasonal variation, which is in some cases significant (Garland, 1985; Garland and Else, 1987; Gleeson, 1979b; Huey et al., 1990; John-Alder, 1984b). Infection with pathogens or parasites could also be used to lower performance (Schall, 1990; Schall and Dearing, 1987; but see Daniels, 1985b; Schall, 1986).

Body size, which often correlates with locomotor performance (see below), can be manipulated in a variety of ways. For example, variation in diet or in thermal regimen (Sinervo and Adolph, 1989) may affect growth rate and hence age-specific body size; such experimentally induced variation may be useful in studies of static allometry (i.e., within an age class). Sinervo and Huey (1990; Sinervo, 1990; Sinervo et al., 1992; see also Bernardo, 1991; Hahn and Tinkle, 1965; Janzen, 1993; Sinervo and Licht, 1991) have used experimental manipulation of egg size in an attempt to separate the effects of body size per se from other factors that may affect speed or stamina. Embryo manipulation studies are common in mammals (e.g., Atchley et al., 1993; Cowley et al., 1989; Hill and Mackay, 1989; Kirkpatrick and Rutledge, 1988) but apparently have not been attempted in reptiles.

Hydric and thermal conditions during incubation can affect locomotor performance of reptiles (Miller et al., 1987; Van Damme et al., 1992), and such effects may not be uncommon (references in Garland and Adolph, 1991). For example, thermal conditions during pregnancy can affect the number of body and tail vertebrae developed by garter snakes (Fox, 1948; Fox et al., 1961; Osgood, 1978; C. R. Peterson and S. J. Arnold, pers. comm.), which in turn may affect locomotor performance (Arnold and Bennett, 1988; Jayne and Bennett, 1989; M. R. Dohm and T. Garland, in preparation). Many other factors may affect maternal size and/or condition and in turn affect offspring size and/or performance; some of these effects can be controlled for statistically via regression analysis and computation of residuals (Brodie, 1989b; Brodie and Garland, 1993; Garland, 1988; Garland and Bennett, 1990; Tsuji et al., 1989).

Truly evolutionary experiments, involving organismal performance or components thereof, are possible using artificial selection (e.g., Bennett et al., 1990; Garland and Carter, 1994; Hill and Caballero, 1992; Huey et al., 1991; Rose et al., 1987; Schlager and Weibust, 1976), but such experiments have not yet been reported for locomotor performance in any organism. Relatively long generation times may preclude such possibilities for reptiles, although experiments with mice are now being conducted (T. Garland, unpubl.). Direct manipulation of the
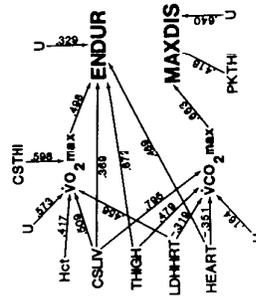

FIGURE 10.5 Path analysis of performance gradients for treadmill endurance at 1.0 km/h (ENDUR) and maximal distance running capacity (MAXDIS) around a circular track in the lizard *Ctenosaura similis* (data from Garland, 1984). For this analysis, only those variables that entered into multiple regression equations as significant predictor (independent) variables and/or that could be explained to a significant extent as dependent variables were considered (see Table 4 of Garland, 1984: SMR was also excluded). Path coefficients were estimated in two ways: first, from standardized partial regression coefficients as described in Nie et al. (1975); second, from the standardized solution output by LISREL Version 4 (Joreskog and Sorbom, 1978), an iterative, maximum likelihood fitting procedure. These two approaches yielded virtually identical results; the figure shows the LISREL results. A variety of path analytic models were fitted with LISREL in order to obtain the model which was judged to best fit the data based on an approximate chi-square-goodness-of-fit statistic and contained no nonsignificant (i.e., $P > .05$) path coefficients (approximate 2-tailed t-tests with 14 degrees of freedom). The path analytic model shown here had a chi-square of 22.4 (df = 20, $P = .3199$), indicating an acceptable fit to the data. Individual path coefficients had t-values of between 2.57 and 14.8, which, by comparison with $t_{(14, .05)} = 2.145$, suggests that all paths are significant. CSTHI = thigh citrate synthase activity (per gram of tissue), Hct = hematocrit, CSLIV = liver citrate synthase activity, THIGH = total mass of right thigh muscles, LDHHRT = lactate dehydrogenase activity in the heart, HEART = heart mass (including atria), PKTHI = thigh pyruvate kinase activity, U = unexplained variation. (See Arnold, 1983; also Sokal and Rohlf, 1981; Bulova, in press; and assumptions in Emerson and Arnold, 1989, p. 299.)

CSLIV is significantly related to ENDUR, $\dot{V}O_2$max, and $\dot{V}CO_2$max (see also multiple regressions in Table 4 of Garland, 1984). Deleting CSLIV from these predictive models resulted in lower coefficients of determination for the multiple regression equations or a higher chi square for the LISREL-fitted path analytic model. Garland (1984) interpreted these results (and data for mammals) as suggesting that liver oxidative capacity plays a significant role in the activity metabolism of ctenosaurs, perhaps via conversion of metabolites during or after activity. Recent studies, however, suggest that the liver is not an important site of lactate metabolism during recovery in amphibians or reptiles (Gleeson and Dalessio, 1989; Gleeson 1991).

and Fahey, 1984; Astrand and Rodahl, 1986) have been attempted only twice with reptiles. These two studies employed very different training regimens and species from different families, yet both failed to improve organismal performance (speed, stamina, maximal oxygen consumption: Garland et al., 1987; Gleeson, 1979b; but see Gleeson, 1991, p. 189). On the other hand, captivity and the accompanying relative inactivity may decrease maximal oxygen consumption ($\dot{V}O_2$max) (Bennett and John-Alder, 1984; Garland et al., 1987; but see John-Alder, 1984b). Training studies definitely deserve further attention; unfortunately, they can be quite labor-intensive because training regimens cannot be

germ line (e.g., genetic engineering to produce transgenic mice) is now routine in many animals (see Hill and Mackay, 1989) but has not been attempted with reptiles.

CONFOUNDING ISSUES IN THE STUDY OF PERFORMANCE AND ECOLOGICAL MORPHOLOGY

Measuring "Performance" as Opposed to "Behavior"

Arnold (1983, p. 352) defined performance as "the score in some ecologically relevant activity, such as running speed. . . ." Most estimates of maximal locomotor performance in reptiles are made in the laboratory, although some field estimates are available (e.g., Belkin, 1961; on mammals see Djawdan and Garland, 1988; Garland et al., 1988). In either laboratory or field, however, definition and measurement of "performance" as opposed to "behavior" is not always simple (cf. Friedman et al., 1992; Garland, 1994a, b). For example, if maximal sprint speed is measured by chasing an animal along a race track, how can one be sure that each individual actually runs at its morphological, physiological, or biochemical limits? Animals may vary in their response to stimuli (their "motivation"), such that some run at their physiological limits and others do not. Thus, behavioral variation, just like morphological or physiological variation, can affect laboratory measurements of performance (see also Wainwright, chap. 3, this volume).

In the laboratory, repeated testing of individuals and use of the fastest trial(s) as an index of maximal speed (e.g., Bennett, 1980; Formanowicz et al., 1990; Garland, 1984, 1985, 1988; Gleeson and Harrison, 1988; Huey 1982a; Losos et al., 1989; Marsh, 1988; Marsh and Bennett, 1985, 1986; Sinervo et al., 1991; but see Jayne and Bennett, 1990a, b) may help circumvent motivational problems. (It is well known in human, horse, and dog racing that performances of individuals vary significantly with the competition and setting.) For some performance measures, it may be possible to verify by supplementary tests that physiological limits have been reached. Thus, physiological exhaustion in endurance trials can be supported by testing for loss of righting response (e.g., Huey et al., 1984, 1990), or by measuring whole-body (Arnold and Bennett, 1984) or blood (Djawdan, 1993) lactic acid concentrations. Alternatively, measures of "race quality" can be used in statistical analyses (Tsuji et al., 1989). In any case, what some workers term "performance" others term "behavior" (e.g., Bennett, 1980).

Another possibility is to test for correlations between individual (or inter-specific) differences in performance and traits thought to affect performance. If underlying morphological, physiological, or biochemical traits explain (statistically) a large fraction (e.g., 47–89%; Garland, 1984, fig. 10.5; Garland and Else, 1987) of the variance in locomotor performance, then it is unlikely that

variation in performance is due solely to differences in motivation or willingness to run. To date, published studies of individual variation have been somewhat more successful in identifying physiological correlates of endurance than of sprint speed (see "Case Studies" below), which suggests that it may be easier to obtain measures of physiologically limited performance capacities in stamina- than in sprint-type activities.

Some studies of individual variation indicate that measures of "behavior" may show correlations with measures of "performance." For example, antipredator display (Arnold and Bennett, 1984), scored at the end of treadmill endurance trials, showed significantly positive correlations with both treadmill endurance and sprint speed in the garter snake *Thamnophis sirtalis* (Garland, 1988; see also Arnold and Bennett, 1988, on *T. radix* and Brodie, 1992, on *T. ordinoides* concerning the correlation between speed and distance crawled prior to assuming an antipredator display). Arnold and Bennett (1984) previously showed that whole-body lactic acid concentrations of *T. radix* exhibiting antipredator displays (at the end of stamina trials) were similar to those of snakes forced to exercise for thirty minutes. Thus, one might expect the antipredator display to be partly dependent on, and hence limited by, physiological capacities. However, Garland, et al. (1990b) found that, whereas speed and endurance showed significant (although weak) correlations with lower-level morphological, physiological, or biochemical traits, antipredator display did not (see also Arnold and Bennett, 1988). Thus, an alternative interpretation is that underlying variation in some axis of "motivation" (Bolles, 1975) has effects on measures of speed, endurance, and antipredator display (higher scores are more offensive and seem to require more physical exertion), leading to some positive correlation.

The foregoing examples emphasize that caution must be exercised when designing or interpreting measures of locomotor "performance." Our discussions of Arnold's (1983) paradigm and extensions thereof assume that true measures of morphologically or physiologically limited performance can be obtained.

Allometry and its Importance

Body size affects many traits, including locomotor performance (e.g., Dunham et al., 1988; Garland, 1984, 1985; Garland and Huey, 1987; Losos, 1990a, b, c). Variation in body size may therefore obscure or enhance relationships between other traits (Emerson et al., chap. 5, this volume). Unfortunately, much of the older ecomorphological literature has attempted to remove the effect of size by using ratios, which is generally ineffective and potentially misleading (cf. Packard and Boardman, 1988).

The importance of considering allometry can be illustrated with a hypothetical example. Suppose that sprint abilities determine habitat use in lizards. Many

studies of lizards have noted correlations between relative limb length (expressed as a proportion of snout-vent length) and various habitat variables (see "Limb Length and Habitat Use" below), and have implicated differences in locomotor ability as the underlying cause of the relationships. Both within and between species, limb length rarely scales isometrically with snout-vent length (i.e., as individuals or species increase in size, limb length either becomes relatively longer or shorter; see below, figs. 10.7c, 10.8c). Further, within and between species, sprint ability usually increases with body size. Consequently, a relationship may exist between habitat use and body size due to the effect of size on sprint speed. Because relative limb length is partly a function of body size (except when limb length scales isometrically with size), a spurious relationship would exist between relative leg length and habitat use. Our reanalysis of Pianka's (1969, 1986) data illustrates this problem (see discussion below and figs. 10.7, 10.8).

Confounding effects of body size can be controlled in a variety of ways. Perhaps the most common way is to regress each variable of interest (e.g., sprint speed, limb length) on some measure of body size (e.g., body mass, snout-vent length) and then compute residuals. These residuals can then be used in correlation or regression analyses or various multivariate techniques, such as principal components analysis (e.g., Garland, 1984, 1985, 1988; Jayne and Bennett, 1990a, b; Losos, 1990a, b, c). If the effects of additional covariates (e.g., temperature) or categorical variables (e.g., sex, season, population) need to be removed as well, then residuals can be computed from multiple regressions including dummy independent variables (e.g., Garland and Else, 1987; Gatten et al., 1991; Jackson, 1973b; Packard and Boardman, 1988; Sokal and Rohlf, 1981). Regressions to compute residuals need not be restricted to linear models (cf. table 3 of Garland and Else, 1987; Jackson, 1973b; Jayne and Bennett, 1990a; see also Chappell, 1989). With interspecific data, methods that allow for statistical complications due to phylogeny must be used when computing residuals (Garland et al., 1992; Harvey and Pagel, 1991; Losos and Miles, chap. 4, this volume; Martins and Garland, 1991).

The foregoing approach is not without problems, however. We will mention three here (cf. Huey and Bennett, 1987; Tracy and Sugar, 1989). First, least-squares regression analysis assumes that the independent variable contains no measurement error. This assumption is not true of measures of body size, resulting in underestimates of true structural relationships (Harvey and Pagel, 1991; LaBarbera, 1989; Pagel and Harvey, 1988; Riska, 1991; Sokal and Rohlf, 1981). Unfortunately, alternatives to least-squares regression slopes (e.g., reduced major axis, majoraxis) are not easily employed where multiple independent variables need to be considered. Second, using the same individual measurement of body size (e.g., each animal weighed or measured a single time) as the indepen-

dent variable for a series of dependent variables may result in correlated errors being introduced into all residuals. Such correlated errors can be avoided by taking several measurements of body size (e.g., when speed is measured, when stamina is measured, when limb length is measured; Garland, 1984; Garland and Else, 1987). Third, when correlating the residuals, one degree of freedom should perhaps be lost for each dependent variable for which residuals are computed.

One alternative to the residual approach outlined above is to simply use multiple regression of the dependent variable (e.g., sprint speed) on both a measure of body size and, say, limb length (e.g., Snell et al., 1988). The problem here is that body size and limb length will generally be highly correlated, and the results of multiple regression analyses are unreliable in the face of such multicollinearity (Slinker and Glantz, 1985). We believe that regression of both speed and limb length on body size, then testing for correlation between their residuals, is a more reliable procedure. Alternatively, experimental manipulations that change mass or limb length—but not both—could be helpful in reducing the correlations between independent variables (cf. Lande and Arnold, 1983; Mitchell-Olds and Shaw, 1987; Slinker and Glantz, 1985).

In some cases, the actual value of the allometric exponent is of interest, perhaps in relation to theoretical models of scaling (e.g., Emerson, 1985; Garland, 1985; Harvey and Pagel, 1991; LaBarbera, 1989; Marsh, 1988). Unfortunately, how best to estimate allometric relationships is unclear. As noted above, the independent variable in allometric studies always incorporates some "error variance," which means that slopes will tend to be underestimated. Moreover, for comparisons of population and/or species means, allometric slopes should be estimated phylogenetically, not merely by a regression involving values for tips of a phylogeny (Garland et al., 1992, 1993; Garland and Janis, 1993; Harvey and Pagel, 1991; Losos, 1990c; Lynch, 1991; Martins and Garland, 1991; Purvis and Pagel, 1993).

Phylogeny and its Importance

Inheritance of a phenotypic trait cannot be studied without knowledge of the relatedness of individuals. Analogously, the evolution of a phenotypic trait cannot properly be studied without knowledge of phylogenetic relationships. That all organisms are descended in a hierarchical fashion from common ancestors means that no set of taxa can be assumed to be biologically or statistically independent. Phylogenetic non-independence has implications for all aspects of statistical analyses, including hypothesis testing, power to detect significant relationships between traits, and estimation of the magnitude of such relationships (Felsenstein, 1985; Harvey and Pagel, 1991; Losos and Miles, chap. 4, this volume; Lynch, 1991; Martins and Garland, 1991; Pagel, 1993). Several

methods now exist for incorporating phylogenetic information into comparative analyses, and various examples exist in which phylogenetic analyses lead to qualitatively different conclusions (Garland et al., 1991, 1993; Harvey and Pagel, 1991; Nee et al., 1992). It should also be noted that phylogenetic methods for estimating and testing, for example, character correlations, can sometimes increase—not just decrease—statistical significance as compared with an inappropriate nonphylogenetic analysis.

As the vast majority of previous comparative studies have been analyzed with inadequate allowance for phylogenetic non-independence, conclusions drawn from them must be viewed with caution. For example, many of the allometric studies we discuss were done nonphylogenetically; practical constraints (e.g., lack of suitable phylogenies: see figs. 10.7, 10.8 below) and time limitations have precluded our trying to redo all of them! Nevertheless, future population- or species-level examination of the morphology → performance → behavior → fitness paradigm should be done with appropriate allowance for phylogenetic non-independence (e.g., Losos, 1990b). An interspecific path analysis, comparable to Arnold's (1983) paradigm for microevolutionary studies (cf. Emerson and Arnold, 1989), would be particularly desirable.

CASE STUDIES

Morphology → Performance

Interspecific differences in locomotor performance are well established in reptiles (e.g., Bennett, 1980; Garland, 1994b; Huey and Bennett, 1987; Losos, 1990b,; van Berkum, 1988; references therein). Population differences have been shown a number of times as well (e.g., Garland and Adolph, 1991; Huey et al., 1990; Sinervo et al., 1991; Snell et al. 1988; but see Bennett and Ruben, 1975). A somewhat surprising finding has been the substantial variation in performance among individuals within single populations (Bennett, 1987; Bennett and Huey, 1990; Huey et al., 1990; Pough, 1989). Sex differences in performance exist and are in some cases due to sex differences in body size; unfortunately, few studies have actually tested for sex differences with adequate sample sizes (e.g., Garland, 1985; Huey et al., 1990; Jayne and Bennett, 1989; Tsuji et al., 1989). Some individual variation in performance ability is due to differences in age and/or size (Garland and Else, 1987; Hailey and Davies, 1986; Marsh, 1988; Pough, 1977, 1978); their effects have been thoroughly separated in only two studies of reptiles (Huey et al., 1990; Sinervo and Adolph, 1989).

Variation in performance calls for both proximate and ultimate explanations. In this section, we consider the former—studies examining the mechanistic bases of performance variation. Note that studies of individual variation in performance and morphology constitute attempts to quantify performance gradients

(Arnold, 1983), although special assumptions are required when individuals of multiple ages are studied (Emerson and Arnold, 1989).

Endurance. Variation in endurance has been less studied than has variation in sprinting ability (see "Sprint Speed" below). Most commonly, endurance is measured as running time to exhaustion on a motorized treadmill. Interspecific comparisons of lizards indicate that treadmill endurance capacity has evolved in concert with both body mass and body temperature (Autumn et al., 1994; Garland, 1994b). Interspecific correlates of endurance have not been studied in detail, but appear to include the energetic cost of locomotion (lower cost leads to higher stamina at a given speed), $\dot{V}O_2$max, and indices of blood oxygen carrying capacity (Autumn et al., 1994; Bennett et al., 1984; Garland, 1993; Gleeson, 1991; Gleeson and Bennett, 1985; Gleeson and Dalessio, 1989; John-Alder et al., 1983; John-Alder et al., 1986a; Secor et al., 1992). Two populations of *Sceloporus merriami*, which differ in maximal sprint speed, apparently do not differ in treadmill endurance (Huey et al. 1990).

Within populations, treadmill endurance increases ontogenetically in most species of lizards (Garland, 1984, 1994b, unpubl.; Huey et al., 1990; see also Daniels and Heatwole, 1990) and in the two species of snakes that have been studied (Jayne and Bennett, 1990a; Secor et al., 1992), although not necessarily in a linearly allometric fashion (Garland and Else, 1987; Jayne and Bennett, 1990a). Positive static allometry occurs in garter snakes (Garland, 1988; Jayne and Bennett, 1990b).

Morphological, physiological, and biochemical correlates of individual differences in treadmill endurance have been studied in the lizards *Ctenosaura similis* (fig. 10.5) and *Ctenophorus nuchalis* (Garland, 1984; Garland and Else, 1987). Correlations of each variable with body mass were removed by computing residuals from regression equations. After this procedure, several underlying variables were shown to correlate significantly with endurance (e.g., $\dot{V}O_2$max, thigh muscle mass, enzyme activities). Correlations with $\dot{V}O_2$max and with thigh muscle mass occur in three of five species studied to date (Garland, 1984; unpublished data on *Callisaurus draconoides* and *Cnemidophorus tigris*; Garland and Else, 1987; John-Alder, 1984b). These studies were the first to document performance gradients for reptilian locomotion.

Treadmill endurance does not correlate with residual hindlimb length in *Sceloporus merriami* (Huey et al., 1990) or in hatchling *S. occidentalis* (Tsuji et al., 1989). However, a small (r = .218) but significant correlation exists between treadmill endurance and residual tail length in hatchling *Sceloporus occidentalis* (Tsuji et al., 1989). Treadmill endurance correlates positively with $\dot{V}O_2$max in *Thamnophis sirtalis* (Garland and Bennett, 1990; Garland et al., 1990b).