E-Note

# Direct and Indirect Effects of Environmental Temperature on the Evolution of Reproductive Strategies: An Information-Theoretic Approach

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ABSTRACT: For ectotherms, environmental temperature affects the optimal size and number of offspring via multiple mechanisms. First, temperature influences the performance of offspring, which directly affects the optimal size of offspring. Second, temperature influences maternal body size, which indirectly affects the optimal size and/or number of offspring when larger females acquire more energetic resources or provide better parental care. Although traditional statistical approaches might distinguish the relative importance of these effects, an information-theoretic approach enables one to estimate effects more accurately by identifying the best evolutionary model in a set of candidate models. Here, we use the Akaike Information Criterion to calculate the likelihoods of seven path models, each derived from one or more optimality models of reproduction. Variation in reproductive traits among populations of lizards (Sceloporus undulatus) was used to quantify support for the models. Our results overwhelmingly supported a model based on an indirect effect of temperature that is mediated by maternal size. Path coefficients of this model were consistent with the hypotheses that, first, larger females can acquire more energy for reproduction and, second, the survival of offspring depends on both their size and their density. Our analyses exemplify how information theory can identify evolutionary hypotheses that merit experimental testing.

Keywords: clutch size, egg size, life history, lizards, Sceloporus, temperature.

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The theory of reproductive allocation describes the factors that contribute to variation in the size and number of eggs produced by organisms (reviewed by Bernardo [1996] and Roff [2002]). Most of this theory relies on the concept of an optimal egg size, which results from opposing selective pressures acting at different stages of the life cycle. A greater allocation of resources per egg enhances the performance of the offspring, but a lesser allocation enhances the fecundity of the parent (Smith and Fretwell 1974; Sinervo 1999). In general, environments that retard the growth of offspring favor the production of fewer and larger eggs because of the resulting increase in the performance of offspring (Sibly and Calow 1983; Taylor and Williams 1984). But the state of the mother can matter as much as the state of the environment. Larger mothers often have greater lipid stores, higher feeding rates, larger body cavities, wider body openings, and more parental experience; all of these traits tend to increase the optimal size or number of eggs (Parker and Begon 1986; Congdon and Gibbons 1987; Sargent et al. 1987). Environmental and maternal characteristics combine to determine the optimal reproductive strategy. Because maternal characteristics are partly shaped by the environment, environmental factors can have direct and indirect effects on the optimal reproductive strategy.

Because both direct and indirect effects can occur, our goal should be to assess their relative importance in natural systems. Yet theorists often base evolutionary models on a single hypothetical mechanism. Similarly, empiricists usually weigh support for a single evolutionary model against support for an unrealistic null model (Yoccoz 1991; Johnson 1999; Anderson et al. 2000). Failures to consider multiple working hypotheses (sensu Chamberlin [1890] 1965) stem not only from the prevailing paradigm of null hypothesis testing but also from an ignorance of nontraditional statistical paradigms (see reviews by Dennis [1996]; Johnson [1999]; Anderson et al. [2000]; Ellison [2004]; and Clark [2005]). Adopting statistical paradigms

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derived from information theory (Burnham and Anderson 2002, 2004), some evolutionary biologists have shifted from traditional tests of null hypotheses to more valuable tests of multiple working hypotheses (Johnson 2002; Ergon et al. 2004; reviewed in Johnson and Omland 2004). These paradigms enable one to construct a set of candidate models and then compare the likelihoods of these models, given some observations. One can use this approach to evaluate theories about adaptation as long as each model predicts one or more distinct relationships between environmental and phenotypic variables.

Here, we illustrate the use of information theory to evaluate competing hypotheses about the causes of variation in reproductive traits. We constructed path models to capture various combinations of direct and indirect effects of environmental temperature on the optimal size and number of eggs. We evaluated the relative strength of support for these hypotheses using the Akaike Information Criterion. This estimate of the expected Kullback-Leibler information (i.e., the information lost when a particular model is used to approximate truth) enables one to rank competing models by their explanatory power when the true relationships between independent and dependent variables are unknown (Johnson 1999; Anderson et al. 2000; Burnham and Anderson 2002). We used path analysis to fit each model and information theory to rank models based on their likelihoods. Our combination of path analysis and information theory exemplifies a powerful way to evaluate the theory of reproductive allocation.

## Predictions of Optimal Allocation Theory

In ectothermic organisms, temperature directly and indirectly affects the optimal size and number of eggs. Direct effects of temperature are mediated by the thermal sensitivity of offspring performance. The optimal egg size generally increases as the capacity for growth by juveniles decreases (Sibly and Calow 1983; Taylor and Williams 1984). Because lower temperatures cause ectotherms to grow more slowly, females should produce fewer, larger eggs in colder environments (Perrin 1988; Yampolsky and Scheiner 1996). This prediction is based on the implicit assumptions that the thermal sensitivity of growth rate does not acclimate and the relationships among size, survival, and fecundity are unaffected by temperature.

Indirect effects are mediated by the thermal sensitivities of maternal phenotypes, such as body size. Temperature has major effects on adult size (Atkinson 1994; Ashton 2004; Blanckenhorn and Demont 2004), and several models describe mechanisms by which maternal size affects the optimal size and/or number of eggs. First, larger mothers generally have a greater capacity to acquire energy before reproduction. When the survival of offspring depends only on their size, larger mothers should use their additional energy to produce more eggs because the benefit of greater fecundity exceeds the benefit of producing eggs beyond a certain size (Parker and Begon 1986). When the survival of offspring depends only on their density, larger mothers should produce larger eggs because the benefit of more offspring would be offset by the reduction in their survivorship (Parker and Begon 1986). When the survival of offspring depends on both their size and their density, larger mothers should produce larger clutches and larger eggs.

Energy acquisition is not the only mechanism by which maternal size can affect the optimal reproductive strategy. Larger mothers may care for their offspring better than smaller mothers. Conceivably, larger mothers construct better nests, deter more predators, or provide more food for their offspring. When the survival of offspring depends on maternal size, larger mothers should produce fewer, larger eggs (Sargent et al. 1987; Hendry et al. 2001). Although both energy acquisition and parental care can increase the optimal egg size, they affect the optimal clutch size differentially; therefore, the relative importance of each mechanism determines the optimal clutch size.

The predicted relationships among environmental temperature, egg size, and clutch size (table 1) can be represented by statistical models. Each direct or indirect effect of temperature predicted by an optimality model corresponds to a particular statistical model (fig. 1, top row). Furthermore, the hypothetical mechanisms of optimality models can operate simultaneously; for example, both the thermal sensitivity of growth by offspring and the greater acquisition of energy by mothers could influence the optimal reproductive strategy. Hence, we can combine the predictions of multiple optimality models to create more complex statistical models (fig. 1, middle and bottom rows). We can evaluate these statistical models with phylogenetic comparative data, which describe the correlated evolution of two or more traits (e.g., independent contrasts). Support for a particular statistical model implies support for its corresponding optimality model(s). Using an information-theoretic approach, we can identify the model that best fits the available data and thus infer the relative importance of mechanisms described by optimality models.

#### Methods

#### Sources of Data

Our analyses focus on variation among populations of a monophyletic group of lizards—the *Sceloporus undulatus* 

	Thermal effect	Thermal effect	
Hypothetical conditions	on egg size	on clutch size	Source
1. Offspring grow faster in warmer environments	Decrease	Increase	Perrin 1988; Yampolsky and Scheiner 1996
2. Mothers are smaller in warmer environments; smaller mothers acquire less energy; offspring sur- vival increases as egg size increases	None	Decrease	Parker and Begon 1986
<ol> <li>Mothers are smaller in warmer environments; smaller mothers acquire less energy; offspring sur-</li> </ol>	Tione	Decrease	Funder und Degen 1700
vival decreases as density increases 4. Mothers are smaller in warmer environments; off-	Decrease	None	Parker and Begon 1986
spring survival increases as maternal size increases	Decrease	Increase	Sargent et al. 1987; Hendry et al. 2001

Table 1: Models of optimal reproductive allocation predict specific relationships between temperature and reproductive traits under sets of hypothetical conditions

species group (sensu Leaché and Reeder 2002). Several factors make this group an excellent focus for our study. First, phylogeographic evidence indicates *S. undulatus* expanded its range latitudinally or altitudinally on at least five occasions (Leaché and Reeder 2002). Second, the species exhibits twofold variation among populations in the size and number of eggs (Niewiarowski 1994; Niewiarowski et al. 2004); some of this variation could be caused by thermal clines because the growth of lizards is very sensitive to temperature (Niewiarowski and Roosenburg 1993; Sinervo and Adolph 1994; Angilletta 2001). Finally, females in colder environments delay reproduction until reaching a larger body size (Angilletta et al. 2004), suggesting temperature exerts direct and indirect effects on the optimal reproductive strategy.

We used published data for 17 populations of S. undulatus, one population of Sceloporus woodi, and one population of Sceloporus virgatus-the extant sister taxon of S. undulatus (see table A1). We used wet egg mass as a measure of reproductive allocation because this variable has been measured more often than dry egg mass. Mean air temperatures experienced by the populations were obtained from the National Climatic Data Center; although operative temperatures would be more appropriate than air temperatures, air temperature and operative temperature should be correlated for a group of organisms with similar physical properties (i.e., S. undulatus). Generally, we used data recorded from 1975 to 2002 by the weather station nearest to each population; however, we used data collected from 1954 to 1959 for a population of S. undulatus in Hidalgo County, New Mexico, because data for other years were unavailable. For populations in Arizona, New Mexico, and Utah, we corrected air temperatures for adiabatic cooling (lapse rate =  $0.65^{\circ}$ C/100 m) because the altitudes of the weather stations differed substantially from those of the study sites. Reproductive and climatic data were used for the evolutionary analyses described below.

We analyzed two complementary types of data: raw data and independent contrasts. Each analysis yields meaningful results under an extreme scenario. If phenotypic variation resulted entirely from plasticity, relationships estimated from raw data describe reaction norms that could be adaptations to environmental heterogeneity. In this scenario, phenotypic data would be independent in the sense that each population inhabited an independent thermal environment. If phenotypic variation resulted entirely from genetic divergence among populations, relationships between independent contrasts estimate correlated evolution throughout the clade. In this scenario, independent contrasts would be statistically independent because each contrast reflects evolutionary divergence between taxa since the most recent common ancestor (Felsenstein 1985). Because we do not know whether the phenotypic variation of interest was caused primarily by environmental or genetic effects, we present analyses of raw data and analyses of independent contrasts.

### Independent Contrasts

The use of independent contrasts is justified when phylogenetic relationships and timings of divergence among taxa are known (Garland et al. 1992). Although gene flow among populations can reduce the accuracy of this knowledge, genetic and phenotypic sources of evidence indicate limited gene flow among populations of *S. undulatus* (Niewiarowski et al. 2004). By computing contrasts for environmental temperature, we assumed environmental states were passed from generation to generation, as were phenotypic states (Garland et al.1992); this assumption is probably valid because the limited dispersal of lizards ensures that individuals in a relatively cold environment pro-



Figure 1: Path models depicting relationships among environmental temperature, maternal size, and reproductive traits under different sets of conditions. These path models are derived from the optimality models summarized in table 1. In each path model, predicted relationships (positive or negative) are linked to specific conditions by color coding.

duce offspring that experience a relatively cold environment. For these reasons, our use of independent contrasts seems justified.

Our phylogenetic hypothesis was based on the Bayesian analysis of mtDNA by Leaché and Reeder (2002), with additional data for populations in Indiana, Arizona, and Colorado (fig. 2). A likelihood ratio test using the general time-reversible model with parameters for invariable sites and variation among sites revealed heterogeneous rates of evolution among lineages (df = 17, P = .05); therefore, we estimated the relative durations of each branch in the phylogeny using the penalized likelihood procedure with the truncated Newton algorithm (r8s, ver. 1.71; Sanderson 2003). Because we were interested only in estimating relative divergence times, we fixed the root (S. virgatus) at an arbitrary age of 1.0 and scaled the remaining branches accordingly. We used the cross-validation procedure to estimate the optimal smoothing parameter. Because the penalized likelihood procedure collapsed some branches, we resolved these nodes with an extremely small internodal value (0.00001) when computing independent contrasts. The results of our phylogenetic comparative analyses were robust to our assumptions about evolutionary rates; in fact, we obtained quantitatively similar results using branch lengths estimated by a maximum likelihood procedure (i.e., the general time-reversible model) or dictated by a punctuational evolutionary model (i.e., all branches lengths = 1).

We used the program COMPARE (ver. 4.6b; Martins 2004) to compute standardized independent contrasts of environmental temperature, maternal snout-vent length, egg mass, and clutch size. To obtain adequately standardized independent contrasts, we had to multiply the branch lengths by a constant and log transform the products (Garland et al. 1992). Independent contrasts that were computed using the transformed branch lengths were adequately standardized, as judged by the absence of correlations between the absolute values of the independent contrasts and their standard deviations ( $r^2 \leq 0.08$  for all variables).



Figure 2: Phylogenetic relationships among populations of the *Sceloporus undulatus* species group. These relationships were based on the analyses of Leaché and Reeder (2002), with additional mtDNA sequence from the ND1 protein-coding gene for populations in Indiana (Angilletta et al. 2004), Arizona (GenBank accession no. DQ395142), and Colorado (GenBank accession no. DQ395143). Branch lengths were estimated by the penalized likelihood procedure (Sanderson 2002).

## Statistical Analyses

## To evaluate our competing models (fig. 1), we performed

path analyses of raw data and independent contrasts with AMOS 5.0 (SPSS, Chicago). This computer program uses maximum likelihood to obtain path coefficients and a  $\chi^2$  estimate of the lack of fit for each model. For analyses of independent contrasts, correlations were computed by assuming all relationships passed through the origin (Garland et al. 1992). For each model, we calculated the second-order Akaike Information Criterion (AIC<sub>c</sub>):

AIC<sub>c</sub> = 
$$\chi^2 + 2K + \frac{2K(K+1)}{N-K-1}$$
, (1)

where  $\chi^2$  is the  $\chi^2$  goodness of fit, *K* is the number of estimated parameters, and *N* is the sample size (i.e., the number of populations). Burnham and Anderson (2002) recommend this version of AIC for relatively small samples (N < 40K). Models were ranked based on their values of AIC<sub>C</sub>, after which these values were rescaled as simple differences between the value of AIC<sub>C</sub> for each model and that of the model with the lowest value (differential AIC<sub>C</sub>). Akaike weights were used to assess which model was most likely to be the best (the Akaike weight is the normalized likelihood that a model fits the data better than any other model in the set). Models were substantially supported if their differential AIC<sub>C</sub> was less than 2.0 (Burnham and Anderson 2002).

## Path analyses of raw data and path analyses of independent contrasts yielded concordant results. For all paths, the signs of the path coefficients were the same when analyses of raw data and analyses of independent contrasts were compared. Nevertheless, not all path coefficients were consistent with the predictions of theory. We expected a negative relationship between egg size and clutch size in all path models (fig. 1), but this relationship was positive for mod-

Results

els A, C, and F (figs. 3, 4). Therefore, the available data do not support the optimality models represented by these path models. Path coefficients of the other models (B, D, E, and G; figs. 3, 4) were entirely consistent with the predictions of theory. We relied on information theory to judge the relative support for these path models.

Based on information theory, temperature was far more likely to affect reproductive traits indirectly than directly (tables 2, 3). For both raw data (fig. 3) and independent contrasts (fig. 4), a model describing an indirect effect of temperature mediated by maternal size (fig. 1*B*) was strongly supported over all others. Path coefficients of this model (figs. 3*B*, 4*B*) were consistent with the hypotheses that, first, larger females can acquire more energy for reproduction and, second, the survival of offspring depends on both their size and their density (table 1, conditions 2 and 3). The best model was at least 20 times more likely than any other model in our set, meaning other models were essentially unsupported by the data. Thus, we conclude a decrease in environmental temperature increases



Figure 3: Path models describing relationships among environmental temperature, maternal size, and reproductive traits. Standardized coefficients are provided for each path (one asterisk = P < .05, two asterisks = P < .01, three asterisks = P < .001), along with standardized errors for endogenous variables. Model *B* is more than 20 times more likely to be the best model in the set than any other model (see table 2).



## D. Temperature and acquisition effects



#### F. Temperature, acquisition and/or parental care effects



## E. Acquisition and parental care effects



## G. Temperature, acquisition and parental care effects



Figure 4: Path models describing relationships among independent contrasts of environmental temperature, maternal size, and reproductive traits. Standardized coefficients are provided for each path (one asterisk = P < .05, two asterisks = P < .01, three asterisks = P < .001), along with standardized errors for endogenous variables. Model *B* is more than 50 times more likely than any other model to be the best model in the set (see table 3).

Table 2: Comparison of path models describing the relationships among environmental temperature, maternal size, and reproductive traits

Model	$\chi^{2}$	df	Κ	AIC <sub>C</sub>	$\Delta_i$	$W_i$
Acquisition effect (fig. 1B)	2.82	3	11	62.54	0	.9558
Parental care effect (fig. 1C)	9.01	3	11	68.73	6.19	.0433
Temperature effect (fig. 1A)	18.04	3	11	77.75	15.21	.0005
Acquisition and parental care effects (fig. 1 <i>E</i> )	2.24	2	12	78.24	15.70	.0004
Temperature, acquisition, and/or parental						
care effects (fig. 1F)	8.84	2	12	84.84	22.30	.0000
Temperature and acquisition effects (fig. 1D)	11.26	2	12	87.26	24.72	.0000
Temperature, acquisition, and parental care						
effects (fig. 1G)	2.06	1	13	100.86	38.32	.0000

Note: For each model, we list the  $\chi^2$  goodness of fit ( $\chi^2$ ), the degrees of freedom (df), the number of parameters (*K*), the Akaike Information Criterion (AIC<sub>c</sub>), the differential AIC<sub>c</sub> ( $\Delta_i$ ), and the Akaike weight ( $w_i$ ).

egg size and clutch size of *Sceloporus undulatus* via an increase in maternal size.

#### Discussion

When interpreting our results, we considered how correlations between independent variables (or collinearity) might have influenced our estimates of the path coefficients. Extreme collinearity tends to artificially inflate path coefficients (Petraitis et al. 1996). Fortunately, such bias was likely to be small in our analyses because environmental temperature and maternal size were weakly correlated. Furthermore, condition indexes derived from eigenvalues of the correlation matrix were well below the cutoff presumed to indicate a severe bias from collinearity; all indexes were less than 10, and the cutoff equaled 1,000 (see Petraitis et al. 1996). Most importantly, the structure of the best model (fig. 1*B*) was invulnerable to collinearity because partial correlations were not included.

Sample size was an equally important consideration. When using the  $\chi^2$  value to test for a lack of fit, an insufficient sample prevents one from rejecting virtually any model (Mitchell 1992). In our application of path analysis, the problem of a small sample was alleviated by our use of information theory instead of significance testing. The calculation and ranking of AIC does not rely on an inference about statistical significance. Before our analyses, we identified candidate models based on the theory of optimal reproductive allocation. Our goal, then, was simply to determine which models were well supported by the available data. When ranking models, we considered sample size explicitly because the second order term of AIC<sub>c</sub> heavily weights the selection of models by the number of observations. This term places a greater emphasis on parsimony (i.e., minimizing parameters) when observations are few. As in other cases where samples were small relative to the number of potential parameters (Johnson 2002; Ergon et al. 2004), the best model (B) was one of the most parsimonious models in the set. Nevertheless, two equally parsimonious models (A and C) were deemed less likely than more complex models based on multiple mechanisms (see table 3). Thus, parsimony alone was not the driving factor in our analysis. As more data become available, we can reevaluate the level of support for these models. Furthermore, the development of theory should lead to new models that will alter the level of support for current models. In this way, information theory enables one to draw the best inference given current knowledge.

After the fact, one might reason that a traditional analysis of a single path model would have led to the same conclusion that we drew from our information-theoretic approach. In our analyses of raw data, the full model (fig. 3G) had only three significant paths, which were the same paths included in the model with the lowest  $AIC_{C}$  (fig. 3*B*). However, nothing guaranteed this particular outcome. Also, despite this outcome, the information-theoretic approach was more informative than a traditional analysis would have been. First, we could not have used a single path model to distinguish among competing hypotheses because they share a subset of their predicted relationships (e.g., see fig. 1B, 1C). Using information theory, however, we were able to gauge the support for a suite of relationships specific to each hypothesis. Second, information theory enabled us to better estimate the magnitude of interesting effects because path coefficients depend on the inclusion or exclusion of certain paths (e.g., see relationships between egg size and clutch size in models F and G). Although we could have removed statistically insignificant links a posteriori, this practice can lead to erroneous inferences (Petraitis et al. 1996). Instead, information theory enabled us to identify the most likely model without testing a null hypothesis, emphasizing the estimation of effect sizes rather than their statistical significance (Anderson et al. 2000). A benefit of this approach can be gleaned from our analyses of independent contrasts (fig. 4). The full model (G) indicated a weak effect of

Model	$\chi^2$	df	K	AIC <sub>c</sub>	$\Delta_i$	<i>W</i> <sub>i</sub>
Acquisition effect (fig. 1B)	3.84	6	8	35.84	0	.9592
Acquisition and parental care effects (fig. 1E)	3.20	5	9	43.70	7.87	.0188
Temperature and acquisition effects (fig. 1D)	4.27	5	9	44.77	8.93	.0110
Parental care effect (fig. 1 <i>C</i> )	13.76	6	8	45.76	9.92	.0067
Temperature effect (fig. 1A)	14.83	6	8	46.83	10.99	.0039
Temperature, acquisition, and/or parental						
care effects (fig. 1F)	12.74	5	9	53.24	17.40	.0002
Temperature, acquisition, and parental care						
effects (fig. 1G)	2.19	4	10	53.61	17.77	.0001

Table 3: Comparison of path models describing the relationships among independent contrasts of environmental temperature, maternal size, and reproductive traits

Note: For each model, we list the  $\chi^2$  goodness of fit ( $\chi^2$ ), the degrees of freedom (df), the number of parameters (*K*), the Akaike Information Criterion (AIC<sub>c</sub>), the differential AIC<sub>c</sub> ( $\Delta_i$ ), and the Akaike weight ( $w_i$ ).

maternal size on egg mass, but the model with the lowest  $AIC_{\rm C}$  (B) indicated a much stronger effect. By accepting the model with the lowest  $AIC_{\rm C}$ , we estimated the indirect effect of temperature on egg mass to be 40% larger than it would have been had we accepted the full model. Finally, two or more models could have been well supported, leading us to use multiple models to generate weighted estimates of effect sizes (referred to as multimodel inference by Burnham and Anderson [2002]). For all of these reasons, information theory enables more accurate estimates of effects and thus emphasizes the determination of biological significance over statistical significance (Anderson et al. 2000).

Two assumptions limit the power of our path analytic approach: the relationships between variables were linear, and the effects of variables were additive (Sokal and Rohlf 1995; Petraitis et al. 1996). In other words, we assumed environmental temperature and maternal size independently influence the optimal egg size. Because most optimality models assume a nonlinear relationship between egg size and offspring survivorship, we can easily envision a model in which direct and indirect effects of environmental temperature on the optimal reproductive strategy are interdependent. Unfortunately, theorists have not considered the combined effect of these two variables on the optimal reproductive strategy. By formulating such models, we could discover whether our path analyses provided a suitable test of the theory of reproductive allocation.

Despite this limitation, the patterns we detected accord with those detected in previous studies of reproductive allocation. Laboratory studies have shown that many species produce larger eggs when reared at lower temperatures (reviewed by Atkinson et al. 2001). Furthermore, populations of *Drosophila melanogaster* maintained at a low temperature evolved a larger egg size than did populations maintained at a high temperature (Azevedo et al. 1996). Yet body size also exhibits plastic and evolutionary responses to temperature (Atkinson 1994; Partridge and French 1996). Moreover, selection for larger body size at a single temperature resulted in a larger egg size (Azevedo et al. 1996). Consequently, these comparative and experimental studies could not identify whether direct or indirect effects of temperature were primarily responsible for increases in egg size. Some comparisons of egg size along latitudinal gradients produced mixed results; for example, copepods (Poulin 1995) tend to produce smaller eggs at higher latitudes, but some other crustaceans do not (Poulin and Hamilton 1997). The biological basis for this discord might become more apparent if one were to apply an information-theoretic approach to these data.

Although our analysis clearly indicates that environmental temperature affects reproductive allocation indirectly, can we really conclude something about the mechanisms that drive this effect? Information theory supported the path model that corresponds to the optimality model of Parker and Begon (1986), in which the survival of offspring was hypothesized to depend on both their size and their density. As assumed by the model, lizards of the genus Sceloporus generally survive better at lower densities (e.g., see Tinkle and Dunham 1986; Tinkle et al. 1993). Furthermore, the survival of lizards often depends on their size at hatching (Ferguson and Fox 1984; Sinervo 1999; M. W. Sears, unpublished manuscript), which in turn depends strongly on egg size. In contrast, we have little reason to believe parental care-the mechanism implied by the second-ranked model-is related to maternal size in lizards. Lizards provide no care beyond the construction of a nest. Perhaps nest-site selection improves with age (and hence size), but lab and field studies of nesting behavior in Sceloporus undulatus do not support this hypothesis (Warner and Andrews 2002; M. J. Angilletta, M. W. Sears, and R. M. Pringle, unpublished data).

Possibly, the effect of maternal size on the egg size of *S. undulatus* was caused partly by physical constraints. Because eggs must pass through the pelvic aperture, their size is limited by the size of the mother (Congdon et al.

1983; Congdon and Gibbons 1987). Large eggs produced by small females could bind or rupture during oviposition (Sinervo and Licht 1991; Sinervo 1999). A model based on a pelvic constraint would predict a positive relationship between maternal size and egg size and a negative relationship between maternal size and clutch size, which are identical to the predictions of models based on parental care (Sargent et al. 1987; Hendry et al. 2001). However, the path model that stems from these models (fig. 1*C*) was poorly supported by data for *S. undulatus* (tables 2, 3). Moreover, the signs of the path coefficients were not entirely consistent with the hypothesis of parental care or pelvic constraints (see figs. 3*C*, 4*C*). Specifically, clutch size was positively related to egg size, but theory predicts a negative relationship.

Ultimately, experiments are needed to confirm adaptation of reproductive traits. Because experimental studies generally consume more time, energy, and money than comparative studies, information theory enables researchers to identify the most plausible hypotheses on which to focus their experimental efforts. The path model supported by information theory corresponded to the optimality model of Parker and Begon (1986) in which the survival of offspring was hypothesized to depend on both their size and their density. These assumptions could be validated experimentally in *S. undulatus*. We could use a variety of methods to manipulate egg size and quantify the survival of hatchlings in natural environments (Sinervo 1990, 1999). We could also manipulate the density of offspring in natural populations to confirm that their survival depends on density (Tinkle et al. 1993). Given the results of our comparative analyses, such experiments should be given higher priority than those designed to evaluate alternative models of reproductive allocation.

We have illustrated how evolutionary biologists can use information theory to identify the most likely model, estimate effect sizes more accurately, and define promising directions for experimentation. This approach seems timely, given the major role of phylogenetic comparative analyses in evolutionary biology. Because comparative studies involve observational rather than experimental data, poor randomization and confounding factors generate uncertainty about the distributions of test statistics (Anderson et al. 2000). The information-theoretic approach avoids the known pitfalls of null hypothesis testing, making it a superior choice for evolutionary biologists using comparative data (Johnson 2002).

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## APPENDIX

Table A1: Summary of the data used in analyses of the relationship between environmental temperature and egg mass in the Sceloporus undulatus species group

	Mean	Female			
	temperature	snout-vent	Egg mass	Clutch	
Species	(°C)	length (mm)	(g)	size	Source
Sceloporus undulatus:					
Alabama	15.6	72	.28	8.3	McKinney 1982, as cited in Smith et al. 1996
Arizona	13.7	65	.29	8.3	Tinkle and Dunham 1986
Colorado, Huerfano County	11.3	72	.32	10.8	Gillis and Ballinger 1992
Colorado, Mesa County	11.9	70	.42	7.9	Tinkle and Ballinger 1972
Florida	19.2	61	.32	5.0	Oufiero and Angilletta 2006; Oufiero et al., forthcoming
Indiana	11.9	77	.41	10.6	Oufiero and Angilletta 2006; Oufiero et al., forthcoming
Missouri	13.6	67	.38	11.0	Marion 1970, as cited in Ballinger et al. 1981
Nebraska	9.8	60	.24	5.7	Jones and Ballinger 1987
New Jersey	12.4	73	.38	8.9	Niewiarowski 1994; Angilletta et al. 2001; Oufiero and Angilletta 2006; Oufiero et al., forthcoming
New Mexico, Grant County	11.8	63	.29	7.2	Vinegar 1975 <i>b</i>

	Mean	Female			
	temperature	snout-vent	Egg mass	Clutch	
Species	(°C)	length (mm)	(g)	size	Source
New Mexico, Hidalgo					
County	18.2	68	.24	9.9	Vinegar 1975b
Ohio	10.9	75	.35	11.8	Tinkle and Ballinger 1972
Pennsylvania	10.2	72	.42	11.0	J. M. Matter, unpublished data
					Tinkle and Ballinger 1972; Oufiero and
South Carolina	18.0	66	.34	7.8	Angilletta 2006; Oufiero et al., forthcoming
Texas	17.9	57	.22	9.5	Tinkle and Ballinger 1972
Utah	14.0	69	.36	6.3	Tinkle 1972
Virginia	11.0	72	.44	9.1	Oufiero and Angilletta 2006; Oufiero et al., forthcoming
Sceloporus virgatus	11.6	59	.26	9.8	Vinegar 1975 <i>a</i> , Smith et al. 1995
Sceloporus woodi	21.5	54	.25	4.1	Jackson and Telford 1974

Table A1 (Continued)

Sources: Mean air temperatures were obtained from the National Climatic Data Center. Weather stations corresponding to each population are identified by their Cooperative Station identification number: Alabama (016121); Arizona (027661); Colorado, Mesa County (053488); Colorado, Huerfano County (058781); Florida (085793); Indiana (120784); Missouri (237455); Nebraska (256200); New Jersey (284229); New Mexico, Grant County (295754); New Mexico, Hidalgo County (293530); Ohio (334672); Pennsylvania (367312); South Carolina (380074); Texas (412792); Utah (427516); Virginia (440766); Sceloporus virgatus (026716); Sceloporus woodi (086414).

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