

University of Nevada, Reno

**Foraging, Feeding, Energetics, and Environment: Interactions Between Physiology
and Ecology of Gila Monsters**

A dissertation submitted in partial fulfillment of the requirements for the degree of
Doctor of Philosophy in Ecology, Evolution and Conservation Biology

By

Christopher M. Gienger

Dr. C. Richard Tracy, Dissertation Advisor

August, 2009

THE GRADUATE SCHOOL

We recommend that the dissertation
prepared under our supervision by

Christopher M. Gienger

entitled

**Foraging, Feeding, Energetics, And Environment: Interactions Between Physiology
And Ecology Of Gila Monsters**

be accepted in partial fulfillment of the
requirements for the degree of

DOCTOR OF PHILOSOPHY

C. Richard Tracy, Advisor

Daniel D. Beck, Committee Member

Jack P. Hayes, Committee Member

Jill S. Heaton, Committee Member

Linda C. Zimmerman, Committee Member

Scott A. Mensing, Graduate School Representative

Marsha H. Read, Ph. D., Associate Dean, Graduate School

August, 2009

Dissertation Abstract

The ecology of Gila monsters is largely related to the endurance of unpredictable resource availability, and to the tolerance of predictably harsh environmental conditions. Gila monsters use a novel suite of behavioral, physiological and morphological adaptations to overcome the vagaries of their desert habitat, and as a result, they survive by interacting with their environment in unique ways that would seem impossible for other species.

This dissertation investigates the physiological and ecological consequences of the unique ways in which Gila monsters interact with their environment. Each chapter analyzes different aspects of how individuals acquire, process, store, and use essential food and water resources, while simultaneously managing the constraints and opportunities provided by the environment.

Chapter one is a laboratory study that investigates the thermal preferences of Gila monsters, the effects of feeding on body temperature selection, and the ability of individuals to physiologically control heat exchange. These investigations set the stage for the ecological importance of body temperature regulation, and also provide a baseline by which to appropriately measure behavioral thermoregulation.

Chapter two explores the importance of temperature regulation by Gila monsters in the field. The physical limitations imposed by the thermal environment are used as a framework to analyze trade-offs between foraging activity and thermoregulation. In order to forage, lizards must nearly always tolerate thermal conditions that are outside of their

preferred range, but in doing so, they sacrifice their ability to thermoregulate and risk reduced performance.

Chapter three examines the influences of body temperature regulation and activity on the use of energy and water. The magnitude of energy use is measured directly, and specific components of energy partitioning are modeled mathematically to yield seasonal and annual energy budgets for Gila monsters in southern Nevada. These analyses are then extended to a larger geographic scale, and the effects of both environmental variation (temperature) and individual variation (body mass) are used to simulate differences in resting energy use between four ecologically distinct populations of Gila monsters.

Each of these chapters takes an eco-physiological approach to investigating the interactions between Gila monsters and their environment. While in some ways Gila monsters may seem maladapted to desert life, I show how they use specific behavioral and physiological strategies to overcome environmental limitations.

Foraging, Feeding, Energetics, and Environment: Interactions Between Physiology and Ecology of Gila Monsters

C.M. Gienger

Table of Contents

Chapter 1

Thermal Biology of Gila Monsters (*Heloderma suspectum*): Behavioral, Physiological, and Ecological Patterns..... 1

Chapter 2

Environmental Constraints on Gila Monsters: Tradeoffs Between Activity and Thermoregulation..... 40

Chapter 3

Seasonal Use of Energy and Water By Gila Monsters 77

List of Tables and Figures

Chapter 1

Table 1. Activity body temperatures experienced by free-ranging Gila monsters	32
Table 2. Repeatability of body temperature selection	33
Figure 1. Body temperature and thermoregulatory set-point responses to feeding	34
Figure 2. Differences in precision of thermoregulation due to feeding	35
Figure 3. Time-course of body temperature selection after feeding	36
Figure 4. Heating and cooling rates between 0 and 35 °C.....	37
Figure 5. Geographic variation in the body temperature patterns of Gila monsters resting in under-ground shelters	38
Figure 6. Comparison of above vs. below-ground body temperatures in Nevada Gila monsters	39

Chapter 2

Table 1. Seasonal indices for above-ground activity, body temperatures, and thermoregulation.....	66
Table 2. Seasonal indices for below-ground body temperatures, and thermoregulation..	67
Figure 1. Annual distribution of achievable body temperatures, thermal quality of habitat, observations of activity, and opportunities for individuals to achieve their preferred body temperature range while above-ground.	68
Figure 2. Annual distribution of thermal constraints on activity	69
Figure 3. Operative temperatures and body temperature while Gila monsters are resting in below-ground refugia on a typical summer day	70
Figure 4. Monthly estimates of the accuracy of thermoregulation during above-ground activity.....	71
Figure 5. Monthly estimates of the accuracy of thermoregulation while resting below- ground	72

Figure 6. Seasonal comparisons of body temperature and accuracy of thermoregulation between above and below-ground locations	73
Figure 7. Annual distribution and timing of above-ground activity (2002-2004)	74
Figure 8. Monthly opportunities for above-ground activity and observations of actual activity for Nevada Gila monsters (2003).....	75
Figure 9. Graphical summary of seasonal trade-offs between activity and thermoregulation of Gila monsters	76

Chapter 3

Table 1. Body mass, CO ₂ production, and field metabolic rates by season	111
Table 2. Measured and predicted water flux rates, total body water, and water economy index by season.....	112
Figure 1. Seasonal field metabolic rates and partitioning of the annual energy budget to activity and rest.....	113
Figure 2. Seasonal rates of water influx.....	114
Figure 3. Influences of body mass, resting body temperature, and amount of above-ground activity on field metabolic rate and water flux in Gila monsters.....	115
Figure 4. Predicted monthly and annual total resting metabolism for Gila monsters from Arizona, Nevada, New Mexico, and Utah	116

Thermal Biology of Gila monsters (*Heloderma suspectum*): Behavioral, Physiological, and Ecological Patterns

Abstract

We investigated geographic patterns of body temperature (T_b) selection, behavioral thermoregulation, physiological control of heating and cooling, and geographic variation in preferred T_b of Gila monsters. The lizards behaviorally selected T_b s that were warmer after feeding, and the magnitude of post-prandial T_b increase was related to size of the meal, but not to time of day. Gila monsters maintained elevated T_b for three to six days after feeding (depending on meal size), and T_b selection was repeatable both within and among days.

Gila monsters heated and cooled at statistically indistinguishable rates, and individuals appear to lack physiological mechanisms to control T_b .

Populations of Gila monsters differed in resting T_b during the active season, but were remarkably similar in preferred T_b during above-ground activities. Differences in resting T_b s among populations are likely related to differences in below-ground thermal environments, while the nearly identical population T_b s preferred during activity may reflect lizards thermoregulating to optimize physiological performance.

Introduction

The foundational research of Raymond Cowles and Charles Bogert (Cowles 1939, 1940, Cowles and Bogert 1944, Bogert 1949a) on body temperature selection in lizards revealed, for the first time, the relationships between ectothermic vertebrates and their environments. Those early papers revealed important regulatory phenomena for which

we now have a relatively thorough understanding; ectotherms use a variety of behavioral, physiological, and morphological mechanisms to control body temperature (T_b) in heterogeneous thermal environments. Previous to this discovery, it was thought that ectotherms were largely passive participants in their thermal environment, rather than active regulators of T_b . Cowles and Bogert also showed that thermoregulation, or “*the maintenance of temperatures within the normal activity range*” (Bogert 1949a), could be influenced by many different aspects of the organism (body size, sex, color, and blood flow) as well as by features of the environment (season, microhabitat, solar radiation, etc.).

Among the many species that Bogert went on to study, it is perhaps the helodermatid lizards (Gila monsters and beaded lizards, genus *Heloderma*) with which he is most commonly associated. In fact, the only Guatemalan helodermatid, *Heloderma horridum charlesbogerti*, is named in his honor (Campbell and Vannini 1988). Bogert, along with his colleague Rafael Martín del Campo (1956), synthesized much regarding *Heloderma*, and not surprisingly, they provide the first (yet cursory) report on the thermal biology of the group. They report that three captive Gila monsters (*H. suspectum*) maintained in a heated cage selected a mean body temperature during “*normal activity*” of 27.7 C (range 27.3 to 33.7).

Since the earliest reports on the thermal biology of Gila monsters, there have been several studies on patterns of body temperatures for Gila monsters in the field (Table 1; see also Table 9 of Beck [2005]). All such studies describe body temperatures during activity, and all similarly arrive at seemingly ubiquitous aspect of Gila monster ecology;

they are a reclusive species that generally prefer relatively low body temperatures during activity, especially when compared to other diurnal, desert, and large bodied, lizards.

Despite the seemingly capacious literature, previous investigations lack several essential elements that make an integrative and comparative analysis of the thermal biology of Gila monster possible. First, while we generally know the T_b patterns for Gila monsters in their natural environment, the range of T_b s that lizards select in the absence of environmental pressures is unknown. This range of temperatures is commonly referred to as the set-point range (T_{set}) and is quantified by testing individuals in a laboratory thermal gradient (Licht et al. 1966). Thermal gradients offer a range of possible T_b s (including those outside of the preferred range), and eliminate possible thermal constraints of the environment on the T_b selected by lizards. Gradients also provide an environment free from predators, conspecifics, and other influences that may otherwise alter T_b selection.

Second, it is not known how temperature selection in Gila monsters covaries with any individual attribute or physiological state. There is a large literature describing the ecological and evolutionary influences on T_b regulation in reptiles in general, (see reviews of Avery 1982, Huey 1982, Zimmerman and Tracy 1989, Shine 2005), yet no experimental evidence exists for factors influencing T_b selection in Gila monsters. To date, explanations for variation in Gila monster T_b have been entirely attributed to geographic variation and the resulting differences in annual and season thermal regimes among study sites (Beck 2005; Table 1).

Third, because we lack even basic knowledge of the factors that influence T_b selection in Gila monsters, the importance of different regulatory mechanisms that work

to produce observed T_b patterns is unknown. Different mechanisms of temperature control (typically behavioral or physiological) can produce varied strategies for manipulating and interacting with the thermal environment (Bartholomew 1982, Tracy et al. 1992, Bauwens et al. 1996, Lutterschmidt et al. 2003, Seebacher and Franklin 2004, 2005). However, the strategies employed by Gila monsters to maintain their characteristically low T_b have remained unexplored.

Lastly, if thermoregulation is to be a factor in evolution and influence the ultimate success of a lineage (Bogert 1949b), then natural selection must be able to act on it through fitness effects on individuals. Differences among individuals are, therefore, important (Bennett 1987), and inter-individual variation may, in part, explain observed differences in T_b (Christian et al. 1985). Determining the repeatability of T_b selection provides a way to separate inter- and intra-individual variation, and can offer insight into whether T_b selection can be influenced by natural selection (Clusella-Trullas et al. 2007).

Without the missing data elements described above, we only have the ability to compare patterns of T_b between populations of Gila monsters in field. Although those observations can provide a quantitative measure of differences among populations, they do not *per se* allow inference into the processes producing observed differences.

The purpose of this paper is first to investigate aspects of the basic thermal biology of Gila monsters so that we may understand factors influencing T_b regulation in this species. We used laboratory studies to analyze the effects of environmental and individual influences on T_b selection, and investigated avenues of both behavioral and physiological regulation of T_b . First, we measured the preferred T_b range (T_{set}) in the

absence of environmental limitations. Then, we tested the hypotheses that behavioral thermoregulation and T_{set} can be altered by specific environmental (photo-period), and physiological influences (digestion). We then tested the hypothesis that Gila monsters have the ability to physiologically control T_b , by measuring heating and cooling rates under fixed temperature regimes. If individuals cool and heat at different rates, it would suggest that they have the ability use internal mechanisms (e.g. peripheral blood shunts, vaso-constriction/dilation, cardiac output, etc.) to control T_b , in addition to using behavioral thermoregulation.

Finally, we summarized the known information regarding T_b patterns of Gila monsters in the wild and highlight the importance of T_b variation in explaining ecological and physiological differences among populations.

Methods

Thermal Gradient

Experiments to assess T_b selection were conducted in a thigmothermal gradient. The gradient was 6.1 X 1.2 X 0.6 m and constructed of 1.5 cm thick plywood. A 1.0 cm thick aluminum base was fastened to the vertical plywood frame from below. The gradient was divided into four lanes (each 0.28 X 0.61 m) by three plywood pieces (61 cm high by 1.5 cm thick); each lane ran the entire length of the gradient. All plywood was coated with marine resin and sealed at joints with silicon sealant. A 1 cm layer of sand provided a substratum which could be changed among trials.

The aluminum floor of the gradient was cooled at one end from below by circulating an ethylene glycol solution maintained at 5 °C through copper tubing (0.75 cm internal diameter) wrapped back-and-forth along approximately one-third of the

length of the gradient. The copper tubing was secured to the gradient bottom with aluminum tape. The warm end of the gradient was heated from below using heating strips (Omega Flexible Heaters SRFG-148/5) taped to the underside of the aluminum base. Heat strips spanned the entire width of the gradient and were placed perpendicular across the lanes every ~30cm along two-thirds of the length of the gradient.

Each heating strip was wired to an electric dimmer switch, then to a solid-state relay, and finally to a relay multiplexer (Campbell Scientific AM416) connected to a computer-controlled datalogger (Campbell Scientific CR10X). To monitor and control the output of each heating strip, thermocouples were taped to the top surface of the aluminum base (beneath the sand substrate) and wired to the relay multiplexers. We adjusted each heating strip independently using a feedback loop program (LoggerNet 3.1, Campbell Scientific) that controlled the relays. Each strip was controlled to +/- 0.1 °C to provide an overall thermal gradient that ranged from 10 to 50 °C and changed approximately 6 °C per meter along the length of the gradient.

Lighting was provided from overhead fixtures suspended 1.2 m above the gradient surface. We used ten 125 W (6400 K) full spectrum light bulbs (Hydrofarm Inc., Petaluma, CA.) evenly placed across the length of the gradient in two parallel rows of five bulbs. This provided an average illumination of 1894 candella (SD ± 248) on the floor surface. Lighting was controlled to provide a 12:12 photo-phase:scoto-phase cycle, which matched the photoperiods provided to individuals in their cages between trials.

T_b sampling

Body temperatures (T_b) of 10 adult Gila monsters (mean mass = 479 ± 115 g) were measured using ThermoChron ibutton dataloggers (Dallas Semiconductor). Each

ibutton was calibrated against a standard thermometer, and they were attached to the chest of the lizard directly over the heart using a 4 X 3 cm strip of 0.5 cm thick foam insulation (Frost King, Thermwell Products, Sparks, NV). The insulated datalogger package was further secured using medical tape (Nexcare Absolute Waterproof tape, 3M, St. Paul, MN) wrapped around the circumference of the chest. The entire datalogger package was less than 5 g, which is less than 2% of the mass of each lizard.

To ensure that temperatures measured by ibuttons matched internal T_b , we compared ibutton recordings against cloacal temperatures. We placed one Gila monster (with attached ibutton) in each of the four lanes of the thermal gradient in the cool end where substratum temperature was 15 °C. A clear plastic shoebox (inverted) was placed over each lizard and T_b was allowed to equilibrate. We then recorded internal T_b using a Schultheis rapid-reading thermometer inserted 1.5 cm into the cloaca. This procedure was repeated at different positions in the gradient with substratum temperatures of 20, 25, 30, and 35 °C. The correlation coefficient between cloacal and ibutton temperature was greater than 0.995 for each lizard, indicating that ibuttons attached to the chest of Gila monsters can give very close estimates of T_b . During all experiments, body temperatures were recorded every 15 minutes for the entire time a lizard was in the gradient.

Behavioral Thermoregulation and Feeding Effects on T_b

Food was withheld from lizards for a minimum of two weeks prior to initiating each experiment. After putting individuals onto the thermal gradient they typically paced up and down the entire length of the gradient for one to four hours. After this period of exploration, lizards usually settled and adopted a thermoregulatory posture in which the ventral body surface was pressed against the substrate. We considered that an individual

was acclimated to the gradient after it was observed either sleeping or resting for six consecutive hours without the exploratory pacing of newly introduced individuals. We continued to log T_{bs} for a minimum of 72 h before feeding.

In the first set of trials, we tested the effect of feeding (large meals) on T_b regulation. We randomly assigned individuals to either a treatment or control group in the experiment. Each lizard was removed from the gradient, placed in a clear plastic shoebox, and either hand fed a meal of rat pups equaling 20% of the body mass of the lizard (treatment group), or fed nothing (sham control group). Lizards were returned to the gradient and T_{bs} were logged for seven consecutive days, and then removed from the gradient and returned to their cages. Each lizard was then switched from treatment to control or vice versa and again randomly assigned to lanes in the gradient for another assessment of selected T_b .

In a follow-up set of trials, we tested the effect of small meals on T_b regulation. We again randomly assigned individuals to the gradient, and gave them a rat meal that was equivalent to either 5 or 10% of body mass using the same protocol as above. Ultimately, each lizard was assigned as a sham, and treated with a meal of 5, 10, and 20% of body mass, and T_{bs} were recorded both before and after each meal or sham treatment. The gradient was cleaned between trials by replacing the sand substrate and wiping down gradient surfaces with a dilute bleach solution.

Means and precision of T_{bs} selected were calculated for each individual in each treatment; feeding period (pre and post-feeding), meal size (sham, 5, 10, or 20% of body mass), and time of day (photo- or scoto-phase). We followed the general procedures of Hertz et al. (1993), and defined the upper and lower set-points (T_{set} lower and T_{set} upper)

as the first and third quartiles of T_b distributions in each treatment. Thermoregulatory precision was evaluated using the index \bar{d}_b , which is the mean of deviations from the T_b set-point range. A \bar{d}_b of zero is taken to indicate that an individual was always within its set-point range.

Physiological Control of Heating and Cooling

To examine the degree to which Gila monsters use physiology to regulate T_b , we assessed the rates at which each lizard changed temperature while heating and cooling. We attached calibrated iButtons to Gila monsters, and placed them in cylindrical cages (14 X 14 X 56 cm) made from #8 mesh (27 ga.) wire hardware cloth. Individuals were placed in one of two adjacent constant temperature cabinets maintained at either 13.5 or 36 °C; N = 5 in each cabinet.

Lizards were allowed to equilibrate to the cabinet environmental temperature (T_e) for three hours and were then they were moved to the opposite cabinet, where T_b was again allowed to come to equilibrium with T_e . After three hours, lizards were switched again, and allowed to come to T_e . T_b was recorded every minute for the duration of the thermal transients.

We report the time individuals needed to change between 15 and 35 °C, as this is approximately the range of T_b s that individuals experience in the field (Beck 2005). We also calculated thermal time constants (TTC; the time for T_b to change 62.3% between starting and ending values in the thermal transient). Plotting $\ln(T_b - T_e)$ versus time for individuals yielded curvilinear relationships, suggesting that second order thermal dynamics were present in the initial part of the thermal transient (Turner 1987, Dzialowski and O'Connor 2001). Because we were interested only in the first-order

dynamics, we calculated TTC from the portion of the log-linear curves that were linear (when first-order dynamics were in effect). This usually meant disregarding the first 14 to 20 minutes of the curves, which was the time it took for the gradient between skin and body core (the second-order dynamics) to come to an equilibrium. TTC was calculated as the inverse of the slope of the $\ln(T_b - T_e)$ versus time curve, after the time point when second-order dynamics were absent (Bakken 1976).

Geographic Variation in T_b

We collected data on T_b from the literature to compare T_b variation among study sites (Table 1). These populations are distinct from one another and vary in many aspects of their ecology including size, coloration, diet, spatial habitat use, and reproduction (Beck 2005). Because the data among these studies were collected with differing sampling frequency (hourly to weekly) and duration (single season to multiple seasons), we generalized analyses to the finest level of resolution possible among studies; monthly mean T_b while resting under-ground, and annual T_b for above-ground activity.

Statistical Analyses

Because T_b data are often skewed (Dewitt and Friedman 1979), and fail to meet the assumptions of parametric tests, we transformed data prior to analyses. We attempted several data transformations (log, square-root, inverse), but none yielded normal distributions. We, therefore, conducted analyses on untransformed data, and because the experiments were perfectly balanced (identical sample sizes among treatments), violation of the normality and homogeneity assumptions should have a small effect on the probability of Type 1 error (Refinetti 1996).

Repeated measures analysis of variance (RM ANOVA) was used to determine differences in T_b selection, thermoregulatory set-points, and thermoregulatory precision (Huey 1982) as a function of feeding state (pre- or post-feeding), meal size (sham, 5, 10 or 20%), and time of day (photo- or scoto-phase). We also use RM ANOVA to test for differences in mean T_b between populations.

The magnitudes of significant treatment effects in ANOVAs are reported using partial ω^2 (Keren and Lewis 1979, Graham and Edwards 2001) presented as a percent. This estimate describes the contribution of a treatment factor to variability in the response variable. After testing main effects of treatment factors, post-hoc comparisons were conducted comparing levels of factors using Fisher's LSD; α was set to 0.05 for all tests and data are reported as means \pm 1 standard deviation unless otherwise noted.

We assessed the repeatability of T_b selection on the laboratory thermal gradient at different temporal scales by calculating the intra-class correlation coefficient, τ , (McGraw and Wong 1996) and Kendall's coefficient of concordance, W , (Zar 1999). Tau (τ) describes the amount of variation in a series of repeated measurements that can be attributed to within subjects (individual lizards in this case) versus among subjects (Lessells and Boag 1987, Hayes and Jenkins 1997). A τ of 1 indicates that individuals are entirely consistent in their T_b selection. Individual T_b values would be the same at every sample point, and all the variation would be among individuals. A τ of 0 indicates no among-individual variation, and differences in T_b would be all within individuals, and therefore not repeatable.

Because τ is sensitive to changes in the mean of repeated measures, we report repeatability for pre-feeding trials only, when T_b selection would not be influenced by digestion. The non-parametric alternative to τ is Kendall's coefficient of concordance, W , and we report it here for comparison.

We analyze heating and cooling rates, and thermal time constants of heating and cooling (Dzialowski and O'Connor 2001) using paired t-tests, and analyze these metrics using Pearson's product moment correlations.

Results

Effect of Time of Day on Thermoregulation

Time of day (day or night) did not have an effect on T_b regulation. There was neither an overall effect of time as factor on mean T_b in the full model (meal size * feeding period * time of day; $F_{1,9} = 0.22$, $P = 0.65$), nor was time of day significant in the interactions with meal size ($F_{3,27} = 0.29$, $P = 0.83$), with feeding state ($F_{1,9} = 1.16$, $P = 0.31$), or with both ($F_{3,27} = 0.60$, $P = 0.44$). Time of day was also not significant as a main or interaction effect for any other T_b set-point calculation (median T_b , T_{set} lower, T_{set} upper), so it was excluded from further analyses.

Effect of Feeding and Meal Size on Thermoregulation

While in a fasting (post-absorptive) state, Gila monsters on the thermal gradient had a mean T_b of 25.2 ± 0.6 °C and set-point range of 23.6 ± 0.92 to 27.1 ± 0.42 °C (mean of individual means for all lizards). There was a strong effect of both feeding and meal size on every T_b metric used to compare 72 h pre-feeding and 72 h post-feeding periods. Mean T_b , was significantly higher after feeding than before ($F_{1,9} = 63.9$, $P < 0.0001$; Figure 1) and it was also significantly effected by meal size ($F_{3,27} = 9.39$, $P <$

0.0001). These results are nearly identical for T_{set} lower ($F_{1,9} = 30.3$, $P = 0.0004$; $F_{3,27} = 12.9$, $P < 0.0001$; Figure 1), median T_b ($F_{1,9} = 31.0$, $P = 0.0003$; $F_{3,27} = 8.0$, $P = 0.0006$), and T_{set} upper ($F_{1,9} = 32.2$, $P = 0.0003$; $F_{3,27} = 4.2$, $P = 0.01$), for tests of feeding state and meal size respectively.

Precision of thermoregulation (\bar{d}_b) was different between pre-feeding and post-feeding periods overall ($F_{1,9} = 12.4$, $P = 0.007$), but was not effected by meal size ($F_{3,27} = 0.46$, $P > 0.5$). There was only a significant difference between pre-feeding and post-feeding periods of the 20% meal treatment (Figure 2; Fisher's LSD = 2.0, $P = 0.04$), suggesting that it takes a large meal to alter thermoregulatory precision (\bar{d}_b) of Gila monsters.

Interaction terms for feeding state * meal size were only significant for mean T_b ($F_{3,27} = 3.3$, $P = 0.04$) and T_{set} lower ($F_{3,27} = 4.3$, $P = 0.01$), but the magnitude of the interaction effect was low for both. Partial ω^2 (Graham and Edwards 2001) was 0.068 for mean T_b and 0.079 for T_{set} lower, indicating that both interactions explained less than 8% of the variance in their respective models.

After feeding, the amount of time that Gila monsters maintained T_b above the pre-feeding level varied with meal size (Figure 3). The sham treatment (0% meal) did not elicit an elevated T_b response, ($F_{7,63} = 1.7$, $P = 0.13$). The 5% meal treatment lead to a significant change in T_b overall ($F_{7,63} = 3.12$, $P = 0.007$), and T_b was significantly elevated for three days post-feeding. The larger size meal treatments lead to longer periods of elevated T_b post-feeding; both the 10% and 20% treatments gave significant overall responses ($F_{7,63} = 4.7$, $P < 0.001$; $F_{7,63} = 11.8$, $P < 0.001$ respectively) and T_b s were significantly elevated for five and six days, respectively (Figure 3).

Repeatability of T_b

Selection of T_b in Gila monsters was repeatable, both within a single day (24 h before feeding) and among several days (3 d before feeding), regardless of the assigned meal size treatment (Table 2). Within-day repeatabilities of T_b (τ) varied from 0.426 to 0.685, and there were significant among-individual differences ($P < 0.0001$ for all; Table 2). Among-day repeatabilities (mean T_b) showed a stronger pattern, ranging from 0.723 to 0.893 between treatments; again there were significant differences among-individuals for both the intra-class correlation (τ) and Kendall's coefficient of concordance (W).

Heating and Cooling Rates

Gila monsters heat and cool at statistically indistinguishable rates. Thermal time constants were 41 ± 4.4 min. for heating and 43 ± 8.7 min. for cooling ($t_9 = 0.84$, $P = 0.42$). Mass influenced time constants when lizards were cooling ($r = 0.725$, $P = 0.018$), but not while they were heating ($r = 0.632$, $P = 0.11$). Similarly, the times to heat or cool between 15 and 35 °C, was 116 ± 14.9 min. for heating and 118 ± 26.9 min. for cooling ($t_9 = 0.19$, $P > 0.5$; Figure 5). Body mass was important to cooling time ($r = 0.718$, $P = 0.019$) but not to heating time ($r = 0.438$, $P = 0.205$).

Geographic Variation in T_b

While above-ground, Gila monsters had mean T_b s that were noticeably similar across seven different populations (Table 1); mean = 29.7 ± 0.6 °C, coefficient of variation = 2.1%. The maximum difference between population means was 2.0 °C, and activity T_b s ranged from 17.4 to 37.7 °C across all populations.

For four populations from Arizona, Utah, Nevada, and New Mexico (Porzer 1981, Beck 1990, Gienger 2003, Beck 2005), we were able to collate data to the level of monthly mean T_b for individuals resting in under-ground refugia. During the activity season (April through August), there is a significant difference in resting T_b s among populations ($F_{3,12} = 17.3$, $P = 0.0001$, partial $\omega^2 = 0.71$), but this difference is due almost entirely to higher resting T_b s experienced by the Arizona population (Porzer 1981). Every individual population significantly differed from the Arizona population (Fisher's LSD $P \leq 0.002$ for all comparisons), but Utah, Nevada, and New Mexico did not differ from each other (Fisher's LSD $P \geq 0.337$ for all). Populations were also consistent in resting T_b across months of the activity season ($\tau = 0.94$; $F_{4,12} = 19.5$; $P < 0.0001$; $W = 0.87$; $\chi^2 = 13.9$; $P = 0.008$).

Discussion

Behavioral Thermoregulation and Influences on T_b Selection

Our analyses show that T_b s selected by Gila monsters in the laboratory are both lower than would be expected from field observations, and they can be influenced by feeding.

After feeding, Gila monsters selected higher T_b s and they thermoregulated with greater precision than during a fasted state. Presumably, by selecting higher T_b s lizards can improve digestion by reducing passage time of the meal through the gut (Waldschmidt et al. 1985) or by increasing digestive efficiency (Harlow et al. 1976, Beaupre et al. 1993). Digestive efficiency of Gila monsters is reported as 76.5% at constant T_b of 27 °C (Beck 1986) and 90.6% at 29 °C (Wegscheider 1998), indicating that

even a modest increase in T_b , such as those observed in this study, could yield improvement in digestive performance.

Although Gila monsters likely elevate T_b following feeding to increase digestive performance, they can also regulate digestive function without changing T_b . Gila monsters are reported to increase metabolism, nutrient uptake rate, and nutrient uptake capacity after feeding while being constantly maintained at 30 °C (Christel et al. 2007). The behavioral decision to select elevated T_b s, therefore, could work interactively with the physiological capacity to optimize digestive function (Tracy et al. 2005).

However, the benefit of using elevated T_b to up-regulate digestive machinery after feeding is likely governed by the size (quality) of the meal. A large meal (either 10 or 20% of body mass) was generally required to evoke a significant increase in thermoregulatory set points (Figure 1). This suggests that increasing T_b after eating small meals may not be necessary, because digestion can take place effectively in the absence of a post-prandial T_b increase, or that the metabolic costs of elevating T_b (Q_{10} effects), may be more than the value of the meal.

Digestive costs, calculated as specific dynamic action (SDA; Secor et al. 1994), for Gila monsters are roughly 18% of the energetic value of the meal (Christel et al. 2007), and research from other carnivorous reptiles has shown that digestive costs can be considerable even for small meals (Secor and Diamond 2000). To process a meal, the digestive organs and cellular machinery must be up-regulated from a state in which the gut is empty to a functioning state when the gut is full. Thus, the digestive process carries an energetic cost, regardless of meal size (Secor 2009).

Larger meals have larger digestive costs, but the net energetic gain on large meals may outweigh the expense to acquire and process the meal. If the metabolic costs of a post-prandial T_b increase are added to the SDA costs, along with the pursuit and handling costs of prey acquisition (Emlen 1966), it could mean that consuming small meals could actually lead to a net energy loss. This means that Gila monsters should only alter thermoregulation (increase T_b) if the costs of doing so are low compared to the energetic return (Huey and Slatkin 1976).

Depending on meal size, Gila monsters maintained elevated T_b for up to six days following feeding (Figure 3), and this is the same the amount of time that digestion is physiologically elevated in the absence of elevated T_b (Christel et al. 2007). This suggests that thermal performance of digestion and post-prandial T_b preference may be functionally linked processes, or that they have evolved as co-adapted traits (Huey and Bennett 1987, Angilletta et al. 2002, Angilletta et al. 2006), although a relation of these factors to fitness would need to be established to support the latter.

T_b selection did not appear to be influenced by time of day. Photoperiod is an important factor influencing temperature selection in other lizard species (Ballinger et al. 1969, Sievert and Hutchison 1991, Tracy et al. 2005), and this is likely due, in part, to voluntary hypothermia (Regal 1967). In an attempt to save energy, lizards sometimes seek cooler T_b s at night when predation risk is putatively lower, and there is less need to maintain high and constant T_b (Dawson 1975). Digestion probably poses a limit to any voluntary reduction in T_b (Tracy et al. 2005) and with large meals, the benefit of increasing digestive function by selecting warmer T_b may outweigh the potential energy savings of reducing T_b .

An alternative explanation might be that photoperiod is simply not a strong environmental cue for temperature regulation in nocturnal and crepuscular species, such as Gila monsters. Nocturnal geckos sometimes have no diel variation in preferred T_b (Angilletta and Werner 1998), while diurnal species routinely show strong day and night differences (Firth et al. 1989, Firth and Belan 1998, Tracy et al. 2005). While diurnal species may transduce photoperiodic cues using the pineal complex and its regulatory effects on melatonin production and T_b selection (Ralph et al. 1979, Lutterschmidt et al. 2003), nocturnal species may not be as sensitive to cues from photoperiod in regulating circadian processes (Hyde and Underwood 2000, Ellis et al. 2006). This idea is supported by the observations that Gila monsters are “*poor time-keepers*” (referencing their circadian patterns) and that they show no differences in activity under constant light or constant dark conditions (Lowe et al. 1967).

T_b Repeatability and Natural Selection

For natural selection to act on a trait, such as preferred T_b , it must have a heritable basis, have sufficient among-individual variation, and must be linked to fitness (Endler 1986). Our data appear to meet these criteria and provide initial, although anecdotal, support for the possibility that natural selection could act on T_b selection in Gila monsters.

In the field, resting T_b s selected by Gila monsters were significantly different among populations, but the differences were consistent within populations. This suggests that populations may be adapted to local environmental conditions (Sears and Angilletta 2003), and that the adaptation could, in part, have a population-specific genetic basis and be important in determining differences in T_b .

Additionally, selected T_b s on the thermal gradient were significantly different

among individuals, and they were repeatable both within and among days (Table 2). This repeatable variation provides the material on which selection could act, and further suggests that variation in performance (sprinting, endurance, digestion, etc.) could be linked to T_b selection (Huey and Bennett 1987, Bauwens et al. 1995). Because higher-performing individuals often have higher reproductive success (Lappin and Husak 2005, Husak et al. 2006), the preferred T_b effects on performance will likely be under control of natural selection.

Although repeatability analyses have been used extensively in performance traits such as sprint speed and oxygen consumption (Bennett 1987, 1990, Hayes et al. 1998, Nespolo and Franco 2007), it has only recently gained attention as a tool for evaluating the potential for natural selection to act on preferred T_b (Clusella-Trullas et al. 2007). Thus, it is difficult to compare T_b repeatabilities observed in this study to those for other ectotherms. Within-day T_b repeatabilities for Gila monsters ($\tau = 0.426$ to 0.685) were much higher than those for four species of cordylid lizards (0.075 to 0.361 ; Clusella-Trullas et al. 2007). Among-day repeatabilities (0.723 to 0.893) were also much higher than those for cordylid lizards ($0-0.261$), as well as other vertebrate ectotherms such as *Lacerta vivipara* (0.66 ; Le Galliard et al. 2003), *Bufo marinus*, (0.66 ; Dohm et al. 2001) and *Thamnophis elegans* (0.47 ; Arnold et al. 1995).

Physiological Control of T_b

Thermal gradient experiments showed that Gila monsters have the ability to attenuate both the magnitude and precision of selected T_b using behavioral thermoregulation. However, they do not appear to possess physiological mechanisms to manipulate rates of heating and cooling. Many reptiles warm more quickly than they cool

(Bartholomew 1982, O'Connor 1999), and this is usually described as way by which animals can exploit thermally sub-optimal conditions by continuing activity (foraging, mate searching, etc.) in habitats with environmental temperatures below preferred T_b .

Some species even rely entirely on this thermal hysteresis to exploit food resources in cool habitats. Galapagos marine iguanas grazing on benthic intertidal algae are forced to forage in ocean temperatures (22-27 °C) far lower than their preferred T_b (37 °C), and they physiologically slow cooling rate to increase the amount of time they can spend underwater foraging (Bartholomew and Lasiewski 1965).

We suspect that the reason Gila monsters do not show differences in heating and cooling is because these lizards have preference for low T_b during activity, and do not maintain high T_b s for physiological performance, or need to extend foraging activity as do diurnal (more heliothermic) lizards (Vitt et al. 2005). Gila monsters are potentially physiologically adapted to lower temperatures than are most lizards, and they have the highest $VO_{2\max}$ of any lizard measured at 25 °C, but perform near expected values at 35 °C (John-Alder et al. 1983). Because Gila monsters are almost always under-ground in refugia with nearly constant T_b , and largely crepuscular or nocturnal when they are active, these lizards may simply avoid wide variation in T_b and do not need to control body temperature physiologically.

Geographic Variation in T_b

There appears to be significant geographic variation among Gila monster populations in T_b while lizards are inactive in under-ground refugia (Figure 5). Individual populations had consistently different mean resting T_b s, suggesting that variation in above-ground temperature regimes could be important in determining below-ground T_b s

(Christian et al. 1983, Christian and Tracy 1984). However, mean activity T_b s were remarkably similar across study sites (29.7 ± 0.6 °C, coefficient of variation = 2.1%), even with many sources of potential error to bias the estimates (different techniques, different sample intervals, and different observers; Table 1), and the inherent climatic and phylogeographic differences among geographically separated populations. This suggests that mean T_b could be a conserved trait among these populations (Youssef et al. 2008), and that Gila monsters attempt to keep T_b near 30 °C, regardless of population, because it is within thermal optima for locomotion or other important processes (Huey and Stevenson 1979, Hertz et al. 1988, Bennett 1990).

The thermal sensitivity of locomotion is unknown for Gila monsters, but individuals appear to sprint maximally at rate of 1.56 ± 0.17 km h⁻¹ when T_b is ~ 31 °C (Beck et al. 1995), which is more than six times faster than the mean rate of travel in the field (0.25 km h⁻¹; Beck 1990). However, Gila monsters are unable to engage in even short periods of burst activity (John-Alder et al. 1983), and are not adept sprinters, so it seems unlikely that optimizing sprint performance is the driving force in their T_b selection. Additionally, these lizards use their venom apparatus for self-defense, so perhaps it is strike performance that is being optimized in regulating activity T_b (Webb and Shine 1998, Vincent and Mori 2008).

Integrating Lab and Field T_b : The Role of Thermal Constraints

Selected T_b s on the thermal gradient can be considered those body temperatures preferred in the absence of environmental constraints (Hertz et al. 1993), and on the gradient, Gila monsters selected much lower T_b s (mean $T_b = 25.2$, $T_{set} = 23.6$ to 27.1 °C) than those observed for individuals active above-ground in the field (Table 1). Active

individuals in the field always had mean T_b s greater than 29 °C, but mean T_b selected on the thermal gradient never reached 29 °C for any treatment combination in this study (Figure 1, top panel). Mean monthly T_b s for animals while resting under-ground were also often outside T_{set} (Figure 5), even during the April to August activity season.

So, do these mismatches suggest that there is little concordance between lab and field data? The answer, in part, relies in recalling how temperature preference and thermoregulation are appropriately estimated. Hertz et al. (1993) proposed formal criteria for evaluating body temperature selection, and remind us that many species thermoregulate between upper and lower set-points, not just around a single mean value (Berk and Heath 1975, Barber and Crawford 1977). When comparing T_{set} of animals on the thermal gradient against those from individuals in the field ($T_{set \text{ field}}$), ranges were markedly similar for resting, but did not overlap at all for active animals (Figure 6).

The differences between lab T_{set} and activity $T_{set \text{ field}}$ are most likely a function of thermal constraints of the environment forcing Gila monsters to be active at T_b s outside of the preferred range. Gila monsters are nest predators of rodents, ground nesting birds, and tortoises, and these animals concentrate reproductive effort in spring and summer. Thus, Gila monsters must forage for their food during the hottest part of the year.

Like Cowles and Bogert, our goal was to investigate how an organism's thermal biology can be determined by interactions between environmental, behavioral, and physiological influences. Like most species, Gila monsters use behavioral thermoregulation (but apparently not physiology) to regulate T_b , and different aspects of thermoregulation change with physiological state and performance needs. On a geographic scale, populations differ in the T_b s they experience, and those differences are

likely dictated by both differences in abiotic conditions, and by local adaptation acting by natural selection on repeatable individual variation.

Acknowledgements

This research was supported by the Biological Resources Research Center, Department of Biology, and Program in Ecology, Evolution and Conservation Biology at the University of Nevada, Reno. We thank the Zoological Society of San Diego for providing experimental animals. Experimental methods were approved by the UNR Institutional Animal Care and Use Committee (protocol A06/07-34). We thank J. Gray and K. Nussear for logistic and technical support. D. Beck, J. Davis, and D. DeNardo provided helpful advice and discussion.

Literature Cited

- Angilletta, M. J., Jr., A. F. Bennett, H. Guderley, C. A. Navas, F. Seebacher, and R. S. Wilson. 2006. Coadaptation: A unifying principle in evolutionary thermal biology. *Physiological and Biochemical Zoology* **79**:282-294.
- Angilletta, M. J., Jr., P. H. Niewiarowski, and C. A. Navas. 2002. The evolution of thermal physiology in ectotherms. *Journal of Thermal Biology* **27**:249-268.
- Angilletta, M. J., Jr. and Y. L. Werner. 1998. Australian geckos do not display diel variation in thermoregulatory behavior. *Copeia*:736-742.
- Arnold, S. J., C. R. Peterson, and J. Gladstone. 1995. Behavioural variation in natural populations. VII. Maternal body temperature does not affect juvenile thermoregulation in a garter snake. *Animal Behaviour* **50**:623-633.
- Avery, R. A. 1982. Field studies of body temperatures and thermoregulation. Pages 93-166 *in* C. Gans and F. H. Pough, editors. *Biology of the Reptilia*. Volume 12 Physiology C. Academic Press, New York.
- Bakken, G. S. 1976. An improved method for determining thermal conductance and equilibrium body temperature with cooling curve experiments. *Journal of Thermal Biology* **1**:169-175.
- Ballinger, R. E., J. Hawker, and O. J. Sexton. 1969. The effect of photoperiod acclimation on the thermoregulation of the lizard, *Sceloporus undulatus*. *Journal of Experimental Zoology* **171**:43-47.
- Barber, B. J. and E. C. Crawford. 1977. A stochastic dual-limit hypothesis for behavioral thermoregulation in lizards. *Physiological Zoology* **50**:53-60.
- Bartholomew, G. A. 1982. Physiological Control of Body Temperature. Pages 167-211 *in* C. Gans and F. H. Pough, editors. *Biology of the Reptilia*, Volume 12. Academic Press, New York.
- Bartholomew, G. A. and R. C. Lasiewski. 1965. Heating and cooling rates, heart rate and simulated diving in the Galapagos marine iguana. *Comparative Biochemistry and Physiology* **16**:573-582.
- Bauwens, D., T. Garland, Jr., A. M. Castilla, and R. Van Damme. 1995. Evolution of sprint speed in lacertid lizards: Morphological, physiological and behavioral covariation. *Evolution* **49**:848-863.

- Bauwens, D., P. E. Hertz, and A. M. Castilla. 1996. Thermoregulation in a lacertid lizard: The relative contributions of distinct behavioral mechanisms. *Ecology* **77**:1818-1830.
- Beaupre, S. J., A. E. Dunham, and K. L. Overall. 1993. The effects of consumption rate and temperature on apparent digestibility coefficient, urate production, metabolizable energy coefficient and passage time in canyon lizards (*Sceloporus merriami*) from two populations. *Functional Ecology* **7**:273-280.
- Beck, D. D. 1986. The Gila Monster in Utah: Bioenergetic and Natural History Considerations. Unpub. M.S. Thesis. Utah State University, Logan, Utah.
- Beck, D. D. 1990. Ecology and behavior of the Gila monster in southwestern Utah. *Journal of Herpetology* **24**:54-68.
- Beck, D. D. 2005. Biology of Gila Monsters and Beaded Lizards. University of California Press, Berkeley.
- Beck, D. D., M. R. Dohm, T. Garland, Jr., A. Ramírez-Bautista, and C. H. Lowe. 1995. Locomotor performance and activity energetics of helodermatid lizards. *Copeia* **1995**:577-585.
- Bennett, A. F. 1987. Inter-individual Variability: An Underutilized Resource. Pages 147-169 in M. E. Feder, A. F. Bennett, W. W. Burggren, and R. B. Huey, editors. *New Directions in Ecological Physiology*. Cambridge University Press, Cambridge.
- Bennett, A. F. 1990. Thermal dependence of locomotor capacity. *American Journal of Physiology - Regulatory, Integrative and Comparative Physiology* **259**:R253-R258.
- Berk, M. L. and J. E. Heath. 1975. Analysis of behavioral thermoregulation in the lizard, *Dipsosaurus dorsalis*. *Journal of Thermal Biology* **1**:15-22.
- Bogert, C. M. 1949a. Thermoregulation and ecritic body temperatures in Mexican lizards of the genus *Sceloporus*. *Anales del Instituto de Biología de México* **20**:415-426.
- Bogert, C. M. 1949b. Thermoregulation in reptiles, a factor in evolution. *Evolution* **3**:195-211.
- Bogert, C. M. and R. Martín del Campo. 1956. The Gila Monster and Its Allies. *Bulletin of the American Museum of Natural History* **109**:1-238.

- Campbell, J. A. and J. P. Vannini. 1988. A new subspecies of beaded lizard, *Heloderma horridum*, from the Motagua Valley of Guatemala. *Journal of Herpetology* **22**:457-468.
- Christel, C. M., D. F. DeNardo, and S. M. Secor. 2007. Metabolic and digestive response to food ingestion in a binge-feeding lizard, the Gila monster (*Heloderma suspectum*). *Journal of Experimental Biology* **210**:3430-3439.
- Christian, K. A. and C. R. Tracy. 1984. Physiological and ecological consequences of sleeping-site selection by the Galapagos land iguana (*Conolophus pallidus*). *Ecology* **65**:752-758.
- Christian, K. A., C. R. Tracy, and W. P. Porter. 1983. Seasonal shifts in body temperature and use of microhabitats by Galapagos land iguanas (*Conolophus pallidus*). *Ecology* **64**:463-468.
- Christian, K. A., C. R. Tracy, and W. P. Porter. 1985. Inter- and intra-individual variation in body temperatures of the Galapagos land iguana (*Conolophus pallidus*). *Journal of Thermal Biology* **10**:47-50.
- Clusella-Trullas, S., J. S. Terblanche, J. H. van Wyk, and J. R. Spotila. 2007. Low repeatability of preferred body temperature in four species of Cordylid lizards: Temporal variation and implications for adaptive significance. *Evolutionary Ecology* **21**:63-79.
- Cowles, R. B. 1939. Possible implications of reptilian thermal tolerance. *Science* **90**:465-466.
- Cowles, R. B. 1940. Additional implications of reptilian sensitivity to high temperatures. *American Naturalist* **74**:542-561.
- Cowles, R. B. and C. M. Bogert. 1944. A preliminary study of the thermal requirements of desert reptiles. *Bulletin of the American Museum of Natural History* **83**:265-296.
- Dawson, W. R. 1975. On the Physiological Significance of the Preferred Body Temperatures of Reptiles. Pages 443-473 *in* D. M. Gates and R. B. Schmerl, editors. *Perspectives of Biophysical Ecology*, New York.
- Dewitt, C. B. and R. M. Friedman. 1979. Significance of skewness in ectotherm thermoregulation. *American Zoologist* **19**:195-209.
- Dohm, M. R., W. J. Mautz, P. G. Looby, K. S. Gellert, and J. A. Andrade. 2001. Effects of ozone on evaporative water loss and thermoregulatory behavior of marine toads (*Bufo marinus*). *Environmental Research* **86**:274-286.

- Dzialowski, E. M. and M. P. O'Connor. 2001. Thermal time constant estimation in warming and cooling ectotherms. *Journal of Thermal Biology* **26**:231-245.
- Ellis, D. J., B. T. Firth, and I. Belan. 2006. Circadian rhythm of behavioral thermoregulation in the sleepy lizard (*Tiliqua rugosa*). *Herpetologica* **62**:259-265.
- Emlen, J. M. 1966. The role of time and energy in food preference. *American Naturalist* **100**:611-617.
- Endler, J. A. 1986. *Natural Selection in the Wild*. Princeton University Press, Princeton, N.J.
- Firth, B. T. and I. Belan. 1998. Daily and seasonal rhythms in selected body temperatures in the Australian lizard *Tiliqua rugosa* (Scincidae): Field and laboratory observations. *Physiological Zoology* **71**:303-311.
- Firth, B. T., J. S. Turner, and C. L. Ralph. 1989. Thermoregulatory behaviour in two species of iguanid lizards (*Crotaphytus collaris* and *Sauromalus obesus*): diel variation and the effect of pinealectomy. *Journal of Comparative Physiology B: Biochemical, Systemic, and Environmental Physiology* **159**:13-20.
- Gienger, C. M. 2003. Natural history of the Gila monster in Nevada. Unpub. M.S. Thesis. University of Nevada, Reno, Reno.
- Graham, M. H. and M. S. Edwards. 2001. Statistical significance versus fit: estimating the importance of individual factors in ecological analysis of variance. *Oikos* **93**:505-513.
- Harlow, H. J., S. S. Hillman, and M. Hoffman. 1976. Effect of temperature on digestive efficiency in the herbivorous lizard, *Dipsosaurus dorsalis*. *Journal of Comparative Physiology* **111**:1-6.
- Hayes, J. P., C. A. Bible, and J. D. Boone. 1998. Repeatability of mammalian physiology: Evaporative water loss and oxygen consumption of *Dipodomys merriami*. *Journal of Mammalogy* **79**:475-485.
- Hayes, J. P. and S. H. Jenkins. 1997. Individual variation in mammals. *Journal of Mammalogy* **78**:274-293.
- Hertz, P. E., R. B. Huey, and T. Garland, Jr. 1988. Time Budget, thermoregulation, and maximal locomotor performance: Are reptiles olympians or boy scouts? *American Zoologist* **1988**:927-938.

- Hertz, P. E., R. B. Huey, and R. D. Stevenson. 1993. Evaluating temperature regulation by field-active ectotherms: The fallacy of the inappropriate question. *American Naturalist* **142**:796-818.
- Huey, R. B. 1982. Temperature, Physiology, and the Ecology of Reptiles. Pages 25-91 in C. Gans and F. H. Pough, editors. *Biology of the Reptilia 12C*. Academic Press, New York.
- Huey, R. B. and A. F. Bennett. 1987. Phylogenetic studies of coadaptation: Preferred temperatures versus optimal performance temperatures of lizards. *Evolution* **41**:1098-1115.
- Huey, R. B. and M. Slatkin. 1976. Costs and benefits of lizard thermoregulation. *Quarterly Review of Biology* **51**:363-384.
- Huey, R. B. and R. D. Stevenson. 1979. Integrating thermal physiology and ecology of ectotherms: A discussion of approaches. *American Zoologist* **19**:357-366.
- Husak, J. F., S. F. Fox, M. B. Lovern, and R. A. Van Den Bussche. 2006. Faster lizards sire more offspring: Sexual selection on whole-animal performance. *Evolution* **60**:2122-2130.
- Hyde, L. L. and H. Underwood. 2000. Effects of melatonin administration on the circadian activity rhythm of the lizard *Anolis carolinensis*. *Physiology & Behavior* **71**:183-192.
- John-Alder, H. B., C. H. Lowe, and A. F. Bennett. 1983. Thermal dependence of locomotory energetics and aerobic capacity of the Gila monster (*Heloderma suspectum*). *Journal of Comparative Physiology B* **151**:119-126.
- Keren, G. and C. Lewis. 1979. Partial omega squared for ANOVA designs. *Educational and Psychological Measurement* **39**:119-128.
- Lappin, A. K. and J. F. Husak. 2005. Weapon performance, not size, determines mating success and potential reproductive output in the collared lizard (*Crotaphytus collaris*). *American Naturalist* **166**:426-436.
- Le Galliard, J. F., M. Le Bris, and J. Clobert. 2003. Timing of locomotor impairment and shift in thermal preferences during gravidity in a viviparous lizard. *Functional Ecology* **17**:877-885.
- Lessells, C. M. and P. T. Boag. 1987. Unrepeatable repeatabilities: a common mistake. *the Auk* **104**:116-121.

- Licht, P. E., W. R. Dawson, V. H. Shoemaker, and A. R. Main. 1966. Observations on the thermal relations of western Australian lizards. *Copeia* **1966**:97-110.
- Lutterschmidt, D. I., W. I. Lutterschmidt, and V. H. Hutchison. 2003. Melatonin and thermoregulation in ectothermic vertebrates: a review. *Canadian Journal of Zoology* **81**:1-13.
- McGraw, K. O. and S. P. Wong. 1996. Forming inferences about some intraclass correlation coefficients. *Psychological Methods* **1**:30-46.
- Nespolo, R. F. and M. Franco. 2007. Whole-animal metabolic rate is a repeatable trait: a meta-analysis. *Journal of Experimental Biology* **210**:2000-2005.
- O'Connor, M. P. 1999. Physiological and ecological implications of a simple model of heating and cooling in reptiles. *Journal of Thermal Biology* **24**:113-136.
- Porzer, L. M. 1981. Movement, behavior, and body temperature of the Gila monster (*Heloderma suspectum*) in Queen Creek, Pinal County, Arizona. Unpub. M.S. Thesis. Arizona State University, Tempe.
- Ralph, C. L., B. T. Firth, and J. S. Turner. 1979. The role of the pineal body in ectotherm thermoregulation. *American Zoologist* **19**:273-293.
- Refinetti, R. 1996. Demonstrating the consequences of violations of assumptions in between-subjects analysis of variance. *Teaching of Psychology* **23**:51-54.
- Regal, P. J. 1967. Voluntary hypothermy in reptiles. *Science* **155**:1551-1553.
- Sears, M. W. and M. J. Angilletta, Jr. 2003. Life-history variation in the sagebrush lizard: Phenotypic plasticity or local adaptation? *Ecology* **84**:1624-1634.
- Secor, S. M. 2009. Specific dynamic action: a review of the postprandial metabolic response. *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology* **179**:1-56.
- Secor, S. M. and J. M. Diamond. 2000. Evolution of regulatory responses to feeding in snakes. *Physiological and Biochemical Zoology* **73**:123-141.
- Secor, S. M., E. D. Stein, and J. Diamond. 1994. Rapid upregulation of snake intestine in response to feeding: a new model of intestinal adaptation. *American Journal of Physiology - Gastrointestinal and Liver Physiology* **266** G695-G705.
- Seebacher, F. and C. E. Franklin. 2004. Cardiovascular mechanisms during thermoregulation in reptiles. *International Congress Series* **1275**:242-249.

- Seebacher, F. and C. E. Franklin. 2005. Physiological mechanisms of thermoregulation in reptiles: a review. *Journal of Comparative Physiology B: Biochemical, Systemic, and Environmental Physiology* **175**:533-541.
- Shine, R. 2005. Life-history evolution in reptiles. *Annual Review of Ecology and Systematics* **36**:23-46.
- Sievert, L. M. and V. H. Hutchison. 1991. The influence of photoperiod and position of a light source on behavioral thermoregulation in *Crotaphytus collaris* (Squamata: Iguanidae). *Copeia* **1991**:105-110.
- Tracy, C. R., K. M. Flack, L. C. Zimmerman, R. E. Espinoza, and C. R. Tracy. 2005. Herbivory imposes constraints on voluntary hypothermia in lizards. *Copeia*:12-19.
- Tracy, C. R., C. R. Tracy, and J. S. Turner. 1992. Contrasting physiological abilities for heating and cooling in an amphibian (*Rana pipiens*) and a reptile (*Sauromalus obesus*). *Herpetologica* **48**:57-60.
- Turner, J. S. 1987. On the transient temperatures of ectotherms. *Journal of Thermal Biology* **12**:207-214.
- Vincent, S. E. and A. Mori. 2008. Determinants of feeding performance in free-ranging pit-vipers (Viperidae: *Ovophis okinavensis*): key roles for head size and body temperature. *Biological Journal of the Linnean Society* **93**:53-62.
- Vitt, L. J., J. P. Caldwell, S. S. Sartorius, W. E. Cooper, T. A. Baird, T. D. Baird, and V. Perez-Mellado. 2005. Pushing the edge: extended activity as an alternative to risky body temperatures in a herbivorous teiid lizard (*Cnemidophorus murinus* : Squamata). *Functional Ecology* **19**:152-158.
- Waldschmidt, S. R., S. M. Jones, and W. P. Porter. 1985. The effect of body temperature and feeding regime on activity, passage time, and digestive coefficient in the lizard *Uta stansburiana*. *Physiological Zoology* **59**:376-383.
- Webb, J. K. and R. Shine. 1998. Using thermal ecology to predict retreat-site selection by an endangered snake species. *Biological Conservation* **86**:233-242.
- Wegscheider, F. J. 1998. Assimilation efficiency of the Gila monster *Heloderma suspectum*. M.S. Thesis. California State University, Fullerton, Fullerton, CA.
- Youssef, M. K., S. C. Adolph, and J. Q. Richmond. 2008. Evolutionarily conserved thermal biology across continents: The North American lizard *Plestiodon gilberti* (Scincidae) compared to Asian *Plestiodon*. *Journal of Thermal Biology* **33**:308-312.

Zar, J. H. 1999. *Biostatistical Analysis*. Prentice Hall, Upper Saddle River, N.J.

Zimmerman, L. C. and C. R. Tracy. 1989. Interactions between the environment and ectothermy and herbivory in reptiles. *Physiological Zoology* **62**:374-409.

Table 1. Mean and range of T_b s experienced by free-ranging Gila monsters during activity.

Mean T_b	T_b Range	Reference	Location	Desert	Sample Method
29.0	22.0 - 34.0	Lowe et al. 1986	Arizona	Sonoran	Cloacal Thermometer
29.3	25.2 - 32.4	Coombs 1977	SW Utah	Mojave	Cloacal Thermometer
29.3	25.0 - 35.6	Porzer 1981	Arizona	Sonoran	Radio Telemetry
29.4	17.4 - 36.8	Beck 1990	SW Utah	Mojave	Radio Telemetry
30.0	-	Beck 1994	SW New Mexico	Chihuahuan	Radio Telemetry
30.1	-	Martin and Lowe (unpub.)	Arizona	Sonoran	Cloacal Thermometer
31.0	22.0 - 37.7	Gienger 2003	S Nevada	Mojave	Radio Telemetry
TBA	TBA	Davis and DeNardo 2009	Arizona	Sonoran	Internal Data Logger

* table adapted from Beck (2005) and Gallardo (2003)

Table 2. Within-day and among-day repeatability of T_b selection in *Heloderma suspectum*.

Meal Size	τ	95% CI	F	P	W	χ^2	P
<u>Within Day</u>							
0%	0.502	0.319 - 0.772	98	< 0.0001	0.590	510	< 0.0001
5%	0.426	0.255 - 0.714	72	< 0.0001	0.419	362	< 0.0001
10%	0.685	0.504 - 0.879	210	< 0.0001	0.607	524	< 0.0001
20%	0.452	0.277 - 0.736	80	< 0.0001	0.502	433	< 0.0001
<u>Among Days</u>							
0%	0.735	0.247 - 0.928	3.77	0.013	0.612	16.5	0.056
5%	0.723	0.214 - 0.924	3.61	0.016	0.723	19.5	0.021
10%	0.864	0.615 - 0.963	7.37	0.0002	0.709	19.1	0.024
20%	0.893	0.696 - 0.971	9.32	<0.0001	0.795	21.5	0.010

Note:

Repeatability measured using intra-class correlation coefficient (τ) and Kendall's coefficient of concordance (W). Data were collected at 15 min intervals in a thermal gradient (10-55°C) for 3 days before feeding in each of four feeding treatments (meal equal to 0 [sham], 5, 10, or 20% of lizard body mass; N = 10 lizards). Within day coefficients were calculated using T_b s recorded for 24 h before feeding (df = 9,950) and among day coefficients from means of individual daily T_b s (df = 9,20).

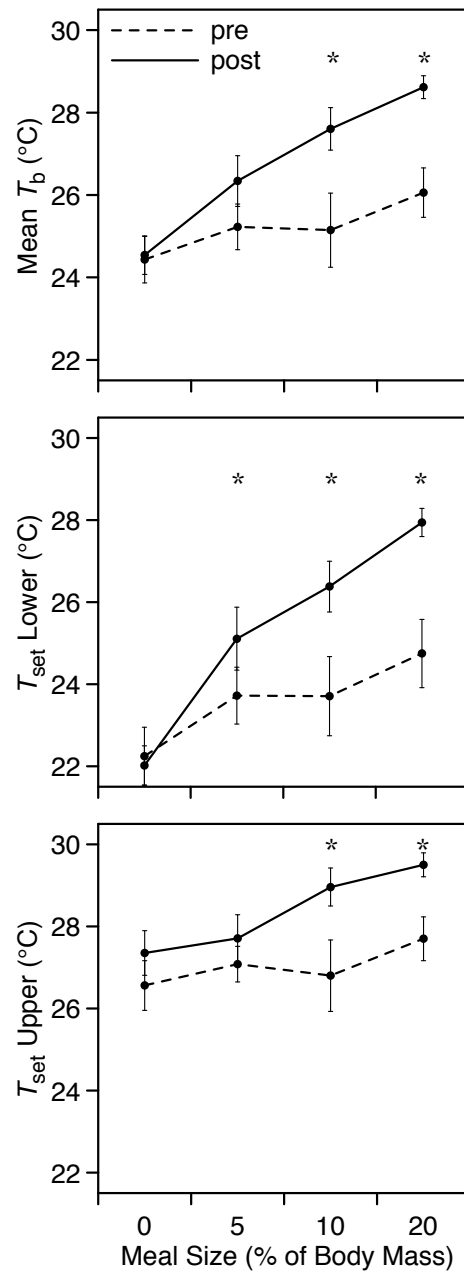


Figure 1. Body temperature (T_b) and thermoregulatory set-point responses to feeding in *Heloderma suspectum* over four feeding levels (meal size as % of lizard body mass) for 72 h pre-feeding and 72 h post-feeding. There is a significant effect of both feeding and meal size on elevating mean T_b , as well as thermoregulatory set-points (T_{set} lower, and T_{set} upper). Stars above pairs indicate significant differences between pre and post-feeding (Fisher's LSD). Values are mean of individual means \pm SE; $N = 10$ individuals for all.

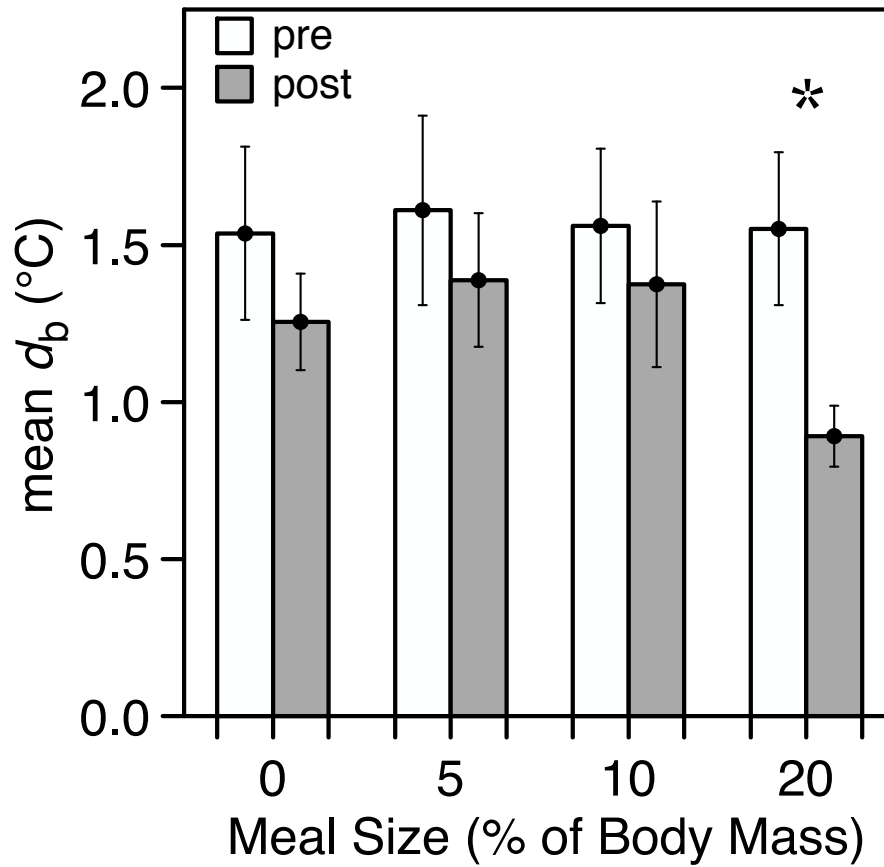


Figure 2. Differences in precision of thermoregulation by *Heloderma suspectum* as a function of feeding period (pre or post-feeding) and meal size. The index \bar{d}_b is the mean distance (in degrees) an individual is away from its preferred thermoregulatory set-point range; a value of zero would mean an animal is always within its preferred range. Bars are mean of individual means (± 1 SE); stars indicate significantly different pairs (Fisher's LSD).

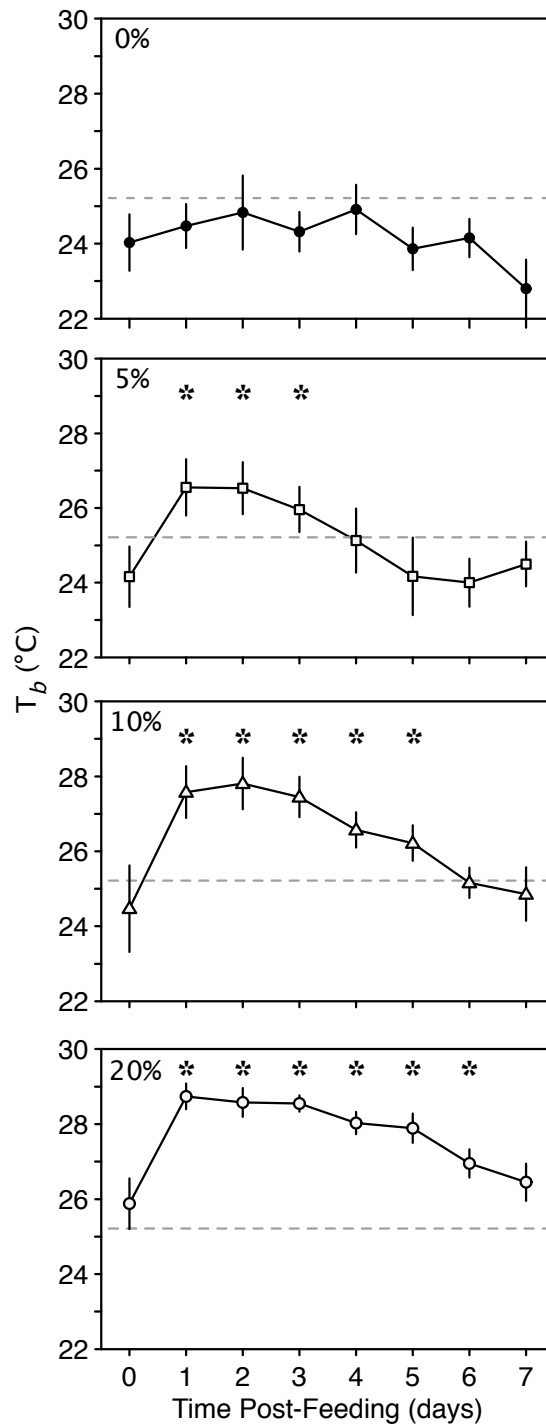


Figure 3. Mean body temperatures (T_b) of *Heloderma suspectum* prior to (day 0) and following feeding for each of four meal size treatments; meals equal to 0 (sham treatment), 5, 10, or 20% of lizard body mass. Values are mean of individual means (± 1 SE) for each combination of meal size and day post-feeding. $N = 10$ individuals for all. Dashed line is grand mean of pre-feeding T_b s.

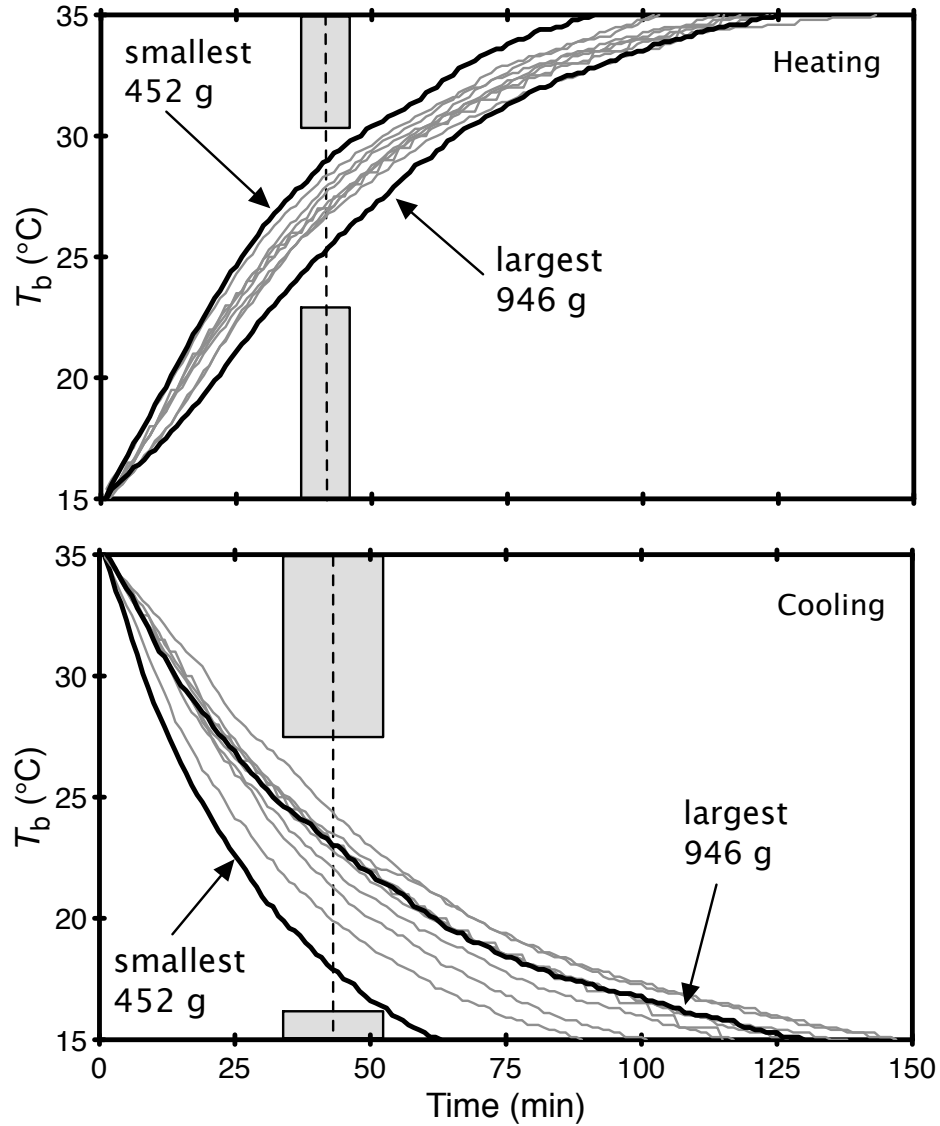


Figure 4. Heating and cooling rates of *Heloderma suspectum* between 0 and 35 °C (curved lines). Thermal time constants and standard deviations for heating and cooling are represented by the dashed vertical lines and vertical boxes (respectively). Mass was significant in determining both rates and time constants of cooling, but not for heating. Arrows point to smallest and largest individuals.

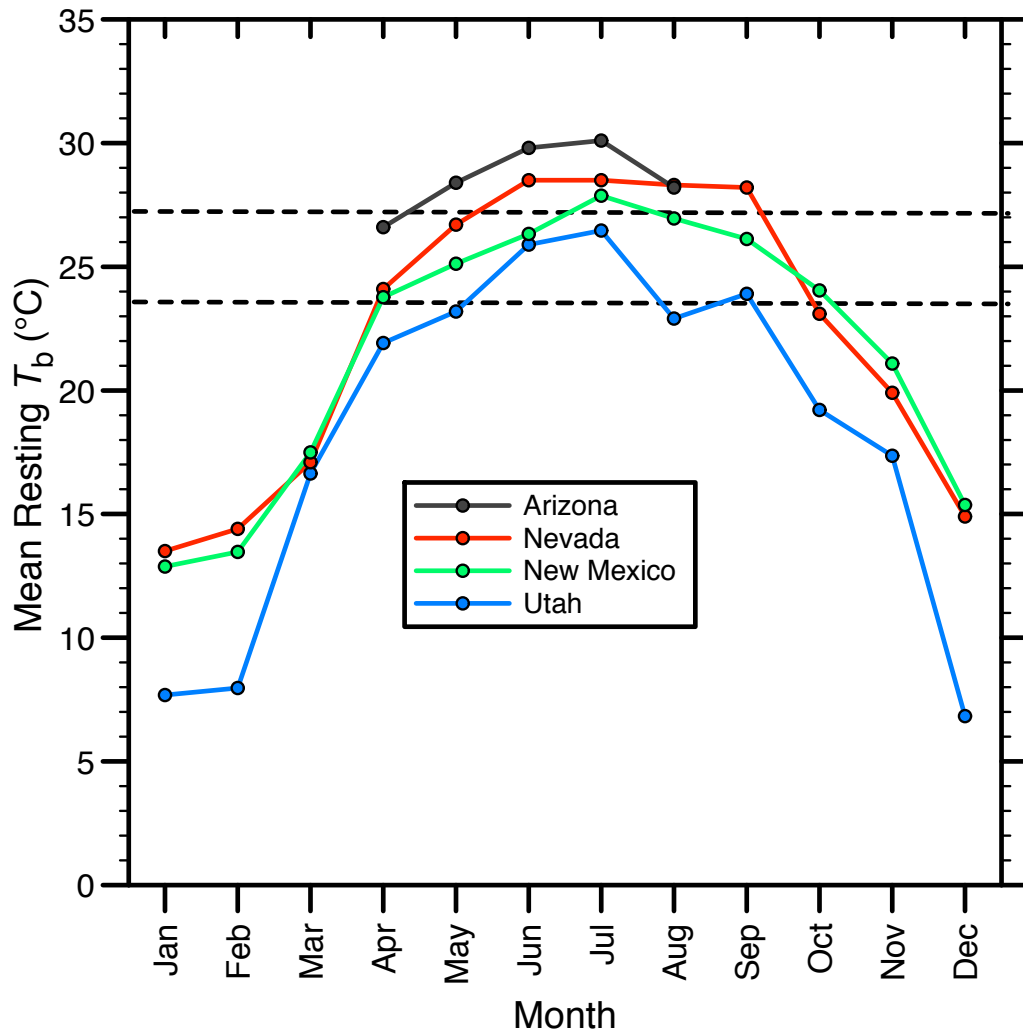


Figure 5. Geographic variation in T_b patterns of *Heloderma suspectum* while resting in under-ground shelters. Data are from Porzer 1981 (Arizona), Gienger 2003 (Nevada), Beck 2005 (New Mexico) and Beck 1990 (Utah). Dashed horizontal lines are the preferred range of individuals in the laboratory thermal gradient (T_{set}).

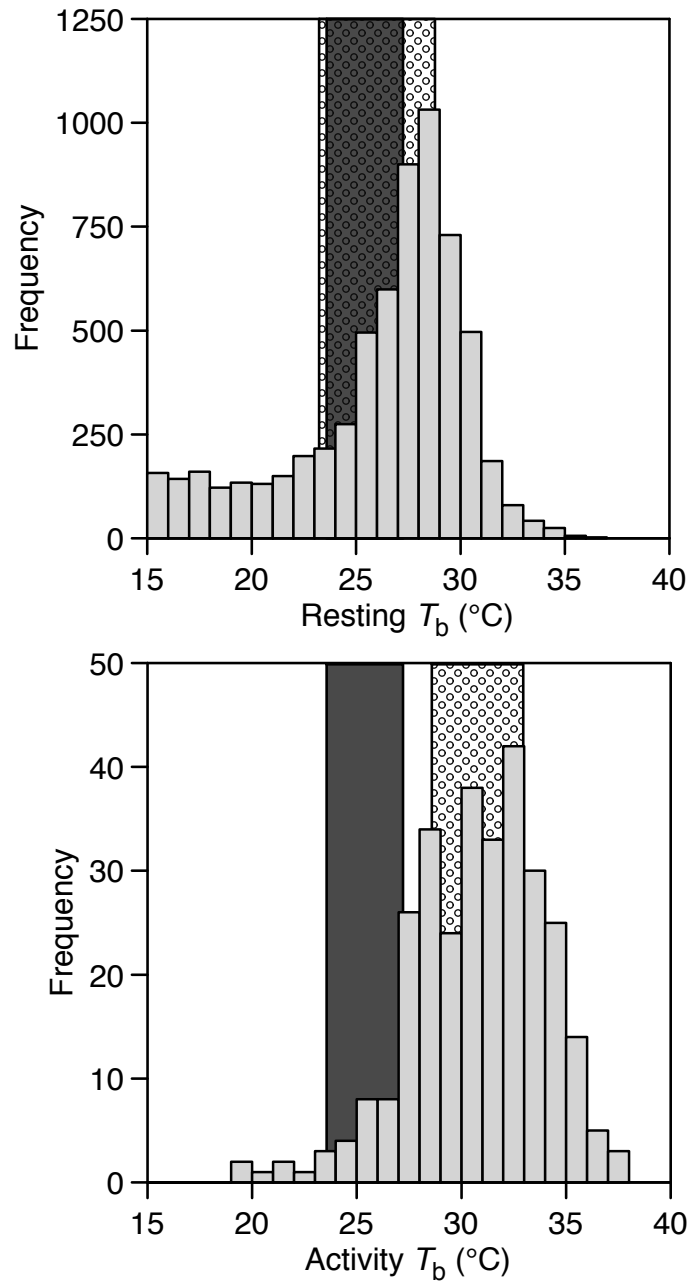


Figure 6. Distribution of T_b for Gila monsters while resting in below-ground shelters (upper) and while active above-ground (lower). Differences between T_{set} ranges measured on laboratory thermal gradient (vertical black bars) and T_{set} calculated from field observations (vertical spotted bars; calculated independently for each T_b category). Data are from Gienger 2003 and unpublished data (2000-2004 southern Nevada). Data below 15 °C are not shown for sake of clarity.

Environmental Constraints on Gila Monsters: Tradeoffs Between Activity and Thermoregulation

Abstract

Gila monsters are a unique lizard species in that many aspects of their ecology differ from what would be expected based on their size, habitat, or evolutionary history. They are reclusive, but unlike other secretive lizards, which tend to be sedentary and nocturnal, Gila monster activity is primarily diurnal, and they search over large areas for food and mates. They are a desert lizard, but appear to have physiologies that are not tolerant of high body temperatures. High above-ground temperatures limit their opportunities for activity, but yet they are most active during the hottest seasons of the year.

We propose that the success of this species is related to the activity patterns of individuals, rather than their ability to precisely thermoregulate by behavioral or physiological means. In order to engage in foraging activity during late summer, Gila monsters are forced to tolerate body temperatures well outside of their preferred range. They seem to nearly abandon thermoregulation late in the activity season, but avoid lethal temperatures by temporally shifting activity and becoming nocturnal. Prey availability poses an additional constraint late in the activity season. Above-ground activity nearly ceases by early fall when prey become unavailable, even though moderate above-ground temperatures would allow them to be active for 22h d⁻¹.

Introduction

For many lizards, thermal environments impose palpable constraints as well as opportunities for activity behavior (Grant and Dunham 1988, Niewiarowski 2001). These

constraints dictate to what extent individuals can use their environment for essential activities such as foraging, social interactions, and reproduction. Species with broad thermal tolerance (eurythermic) should be less constrained by environmental temperatures, and would likely exploit their environment to a larger degree than species with narrow temperature tolerances (Huey and Hertz 1984). On the other hand, a stenothermic lizard capable of regulating body temperature to a particular narrow range of temperatures is likely to be physiologically and behaviorally more efficient while operating at their preferred body temperatures (Huey 1982). Thus, eurythermy has both benefits and limits for the lizard.

Thermal constraints may be either temporal when only certain parts of the day, season, or year are suitable for activity (Adolph and Porter 1993, Sears 2005), or spatial, in which animals are limited to using selected microhabitats within their home range (Christian and Tracy 1985, Grant and Dunham 1988, Adolph 1990). Frequently, both types of limitation occur and they have an interactive effect that results in spatio-temporal windows in which only selected parts of an animal's habitat are suitable for specific times (Scott et al. 1982, Porter and Tracy 1983, Tracy and Christian 1986, Bashey and Dunham 1997).

When environmental temperatures are completely unsuitable for activity (either too hot or too cold), organisms often take refuge in underground burrows, rock crevices, or caves. And even though some reclusive animals choose voluntarily to spend the majority of time underground, it is often unknown to what degree individuals are forced into inactivity as a result of unsuitable thermal environments rather than being inactive voluntarily.

Here, we investigate the possible thermal constraints on activity and thermoregulation in Gila monsters (*Heloderma suspectum*). Thermal constraints are especially important to the ecology of Gila monsters for two reasons. First, Gila monsters have specialized diets requiring individuals to be active above-ground, searching widely for food. They are entirely nest predators, and feed on the nestlings and eggs of reptiles (especially tortoises), rodents, lagomorphs, and ground-nesting birds (Woodson 1949, Stahnke 1952). These prey items are high in quality, but far from ubiquitous in space and time. Gila monsters can spend weeks searching before successfully finding a meal. The breeding seasons of many of their prey species are limited to spring and summer, and so Gila monsters must forage during the hottest times of year.

Secondly, Gila monsters are relatively thermophobic compared to diurnal desert lizards, having both low preferred body temperatures (T_b) during activity (~ 30 °C; Gienger 2003, Beck 2005), and a low set-point range for thermoregulation ($T_{set} = 23.6$ to 27.1 °C; Chapter one). Maintaining T_b within the preferred range may be often difficult as these lizards are largely restricted geographically to the hottest deserts of the American southwest and northern México (Mojave, Sonoran, Chihuahuan). Consequently, it would seem that the thermal biology of these lizards is somewhat unexpected given their distribution and specific prey requirements, and that individuals could be severely limited in foraging opportunities and in ability to thermoregulate.

When not foraging, Gila monsters are very reclusive and spend the majority of time hidden in underground refugia (Lowe et al. 1986, Beck 1990), even when environmental temperatures are presumably favorable for activity above-ground. It is not known whether this inactivity is related to the constraints imposed by above-ground

temperatures, or whether patterns of inactivity can be attributed to other limiting factors of the environment (i.e., predation), or due simply to a penchant for inactivity.

We investigated aspects of both the magnitude and timing of possible thermal constraints on activity and opportunities for thermoregulation in Gila monsters. We asked (1) how does thermal quality of Gila monster environment change both daily and seasonally, and to what extent can Gila monster activity patterns be explained by limitations imposed by unsuitable above-ground thermal conditions. Secondly, how effective are Gila monsters at thermoregulating, and exploiting opportunities for thermoregulation, given their surprising preference for low T_b s and their geographic restriction to thermally challenging habitats.

Methods

Field Site

The field site, radio telemetry, and T_b methods are described in detail elsewhere (Gienger 2003), but will be briefly described here as well. We implanted 12 Gila monsters with temperature-sensitive radio transmitters (Telonics Inc., Mesa, AZ); lizards were collected from a single study site near Lake Mead, in the Mojave Desert of southern Nevada. Each transmitter was calibrated in a water bath over the range of 5 to 45 °C against a secondary-standard mercury-in-glass thermometer. Each transmitter comprised less than 5% of lizard body mass. Surgeries were conducted in the lab while lizards were under anesthesia (Isoflurane) using methods approved by the UNR-IACUC. Post-surgical lizards were offered water and chicken egg *ad libitum*, and individuals were returned to the field 24-48 h after surgery.

Characterizing the Thermal Environment

We used operative temperature models (Bakken 1992, Dzialowski 2005) to quantify the thermal environments at all hours of the day so that we could infer the amount of time available for Gila monsters to engage in above-ground activity (e.g. foraging, mate-searching, etc.). Operative models allow the quantification of the thermal environment at the same scale at which the animal experiences it (Tracy 1982, Bakken 1992). The thermal environment itself is a biophysical aggregate of air temperature, wind speed, relative humidity, and radiation (Tracy and Christian 1986, Walsberg and Wolf 1996, Campbell and Norman 1998), and operative models experience all these factors simultaneously, just as does an animal.

We constructed eight hollow copper models of Gila monsters by electroplating the surface of nickel coated wax castings made from preserved museum specimens (Porter et al. 1973). We painted the models (Krylon #8141 Khaki Ultra-Flat) to approximate the integrated solar absorptance of live individuals ($70.3\% \pm 4.1\%$; unpub. data), fitted models with thermistors, and connected each to temperature-recording dataloggers (Hobo H08; Onset Computer Corp.). Model temperatures were compared to a live restrained Gila monster in the field, and each was highly correlated with the live individual (r^2 of 0.98 or better for each; Pearson's product moment correlation).

Models were distributed in variety of microhabitats on the ground surface within known home ranges of radio-tracked individuals. For each model, we recorded placement as (1) full sun, where model would receive direct and unobstructed sunlight for the majority of the daytime, (2) full shade, where the model would receive no direct or filtered sunlight throughout the day, or (3) partial shade, typically under shrub cover, where a model would receive only filtered or scattered sunlight for the majority of

daytime.

Model temperatures were recorded every fifteen minutes throughout the day and models were moved to a new location every 24-48 h. Any potential bias in model placement would be mitigated by the frequent re-positioning of models to new sample locations. Temperatures were recorded continuously over time blocks of four to six consecutive days before being removed from the field to download data loggers. We sampled up to four time blocks per month, yielding 14 to 18 complete days of sampling for each month of 2003.

Quantifying Activity, Possible Activity, and Thermoregulation

We quantified activity of Gila monsters using direct observations of radio-implanted individuals tracked from May 2001 through July 2004. Each lizard was located during daylight hours, two-to-four times daily from March to September, and two-to-four times weekly from October to February (the non-activity and hibernation seasons). Each time a lizard was located, we recorded T_b and categorized the individual as either active if it was foraging or moving about above-ground, or inactive when either basking at burrows or while resting below-ground. Gila monsters do not often rest above-ground, and when they are active, they maintain nearly constant movement (either walking or digging for prey) and usually only pause during their locomotor activities for a few minutes at a time. A lizard was considered to have been active, if it was directly observed moving about above-ground, or if it had changed shelter locations in between observation periods.

Time available for Gila monsters to be active above-ground was estimated by collating data from the thermal models. Maximum and minimum operative temperatures

(T_e) were calculated from all models at 15-minute intervals over each sampled day (96 daily T_e samples). If the minimum operative temperature in each 15-minute block was greater than the voluntary thermal minimum (VT_{\min} ; 19.9 °C) and the maximum operative temperature less than the voluntary thermal maximum (VT_{\max} ; 37.6 °C), then it was considered possible for a Gila monster to engage in above-ground activity. We summed possible activity times to yield daily and semi-monthly activity estimates of possible activity. Semi-monthly estimates were calculated by averaging the daily means over the first and second two-week periods of each month (e.g. 1st through 15th and 16th through 31st).

Using the framework of Hertz et al. (1993), we calculated a number of indices that describe different aspects of thermoregulation and the relationships among the T_b s preferred by Gila monsters, those T_b s that are available in the environment, and those T_b s that are actually achieved in the environment. First, we calculate the accuracy of thermoregulation, \bar{d}_b , as the mean deviation of T_b s from the preferred range, T_{set} (23.6 – 27.1 °C; chapter one). The larger the value of \bar{d}_b , the further an individual is from the preferred range. Then, we determined the thermal quality of the environment (\bar{d}_e) as the mean deviation of T_e from T_{set} . As the thermal quality of the environment decreases, it should be increasingly difficult for individuals to keep T_b within T_{set} .

We use a modified calculation of \bar{d}_e , following the recommendations of Christian et al. (2006) to adjust for the potential influences that thermal inertia, due to the relatively large size of Gila monsters, has on T_b s achievable by lizards in the environment. We modeled the mean achievable T_b for each hour of the day using a combination of mean

hourly maximum and minimum T_e values from the operative models, empirically determined time constants for the lizards of 41 minutes for heating, 43 minutes for cooling (chapter one), and a lizard mass of 350 g. Details of the procedure are given in Christian et al. (2006), and the specific model parameters are reported here for reference. Then, we substituted mean achievable T_b for \bar{d}_e in subsequent calculations and averaged hourly values over semi-monthly and seasonal periods.

Effectiveness of thermoregulation was calculated first as $E_{all} = 1 - \bar{d}_b / \bar{d}_e$ for all observations, and secondly using only those observations where \bar{d}_e (hourly \bar{d}_e) was within T_{set} (E_{set}). The E index evaluates how closely an animal is able to maintain T_b s within the preferred range given the specific environmental conditions; a value of zero indicates random body temperatures or no thermoregulation (i.e. thermoconformity) and a value of one indicates perfect thermoregulation (always within the set-point). But because this index is calculated from a ratio, and has the potential to be misleading, we also used the $(\bar{d}_e - \bar{d}_b)$ index as suggested by Blouin-Demers and Weatherhead (2001b). This allows an unscaled comparison of lizard temperatures relative to environmental temperatures. For $(\bar{d}_e - \bar{d}_b)$, negative values would describe individuals avoiding thermally suitable habitats, positive values describe the magnitude of thermoregulation, and zero would indicate perfect thermoconformity.

Lastly, we examined Gila monster's exploitation of possible opportunities for thermoregulation while they are active. We calculate the proportion of the total T_b observations where both T_b and hourly \bar{d}_e were within the T_{set} range (Brown and Weatherhead 2000). This allows a comparison of the thermal opportunities provided by

the environment against those actually used by individuals. This index is conceptually identical to the metric E_x (Christian and Weavers 1996), which describes the amount of time T_{bs} are within T_{set} when the environmental conditions permit ($\bar{d}_e = 0$).

We conducted separated calculations for locations above-ground (activity) and below-ground (resting). Because Gila monsters spend the vast majority of their time below-ground, any composite estimates of thermoregulatory indices using both locations would be dominated by the below-ground analyses. Additionally, because our focus was to examine the possible trade-offs in thermoregulation as a function of activity (which necessitates being above-ground), splitting the analyses into above and below-ground locations allows a more meaningful and focused investigation.

Statistical Analyses

We calculated operative temperatures, and thermal constraints, on possible above-ground activity for the entire population, but we used individual estimates of actual activity and thermoregulation (indices described above). General Linear Mixed-Effects Models (StatSoft 2001) were used to test for differences between above and below-ground locations and for differences among sample periods. Individual lizards were considered a random effect to account for repeated measures. Because there were double the amount of semi-monthly T_e sample periods ($N = 24$) as there were individual lizards ($N = 12$), it was necessary to conduct statistical analyses at a coarser temporal scale that would allow sufficient power to detect differences among periods. Sample periods were subsequently grouped into ecologically relevant seasons of equal length; Spring (March 16 to April 30), Early Summer (May 01 to June 15), and Late Summer (June 16 to July

31). These three seasons cover 80% of the annual above-ground activity observed in this Nevada population.

Results

We recorded more than 3,200 T_b s from 12 lizards (7 male, 5 female) over 285 sampling days during 2003. Of these observations, 293 (9.2%) were from spring, 870 (27.2%) were from early summer, and 768 (24.0%) were from late summer. A total of 638 observations of above-ground activity were recorded in 2003 with 507 (79.5%) from the activity season; 72 (14.2%) observations were from spring, 206 (40.6%) from early summer, and 229 (45.2%) from late summer. Lizards were located on 129 of 136 possible sample days during the spring through summer activity season (16 March through 31 July). A comparison of descriptive and quantitative patterns of T_b , thermal quality of the habitat, thermoregulatory indices, and activity are presented in Tables 1 (above-ground) and 2 (below-ground).

Thermal Quality of Above and Below-Ground Habitats

Daily and seasonal variation in above-ground temperatures led to strong differences in the thermal quality of environments and T_b s available to Gila monsters. Thermal quality (\bar{d}_e) of the above-ground environment changed daily, monthly, and seasonally (Figure 1). As would be expected, available T_b s were lowest and hourly \bar{d}_e values high during the winter (Figure 1), when Gila monsters were hibernating (mean available $T_b \sim 5\text{-}15\text{ }^\circ\text{C}$; $\bar{d}_e \sim 17$). T_e s that would allow either activity or accurate thermoregulation (hourly $\bar{d}_e = 0$) were not available until mid-march (Figure 1), and this corresponded to the first observations of Gila monsters basking at the openings of winter hibernacula and first observations of above-ground activity (Figure 2).

Environmental $T_{\text{e}}\text{s}$ were highest during mid-day in late summer ($>40\text{ }^{\circ}\text{C}$; all individuals resting below-ground) and hourly \bar{d}_{e} values were often more than $25\text{ }^{\circ}\text{C}$ above T_{set} (Figure 1). Thermal quality of the above-ground environment was ubiquitously low at any time of the day during late summer ($\bar{d}_{\text{e}} = 11.6$ for all hours of the day) and even when considering only those times of day when lizards were actually observed active, $T_{\text{e}}\text{s}$ were still far from T_{set} ($\bar{d}_{\text{e}} = 8.6$). $T_{\text{e}}\text{s}$ were also potentially lethal during parts of the summer, and predicted $T_{\text{b}}\text{s}$ were above the critical thermal maximum body temperature (CT_{max}) of 42.5°C (Bogert and Martín del Campo 1956) for ~ 7.5 , 9.5 , and 6.5 h d^{-1} for June, July, and August, respectively.

Available times to maintain T_{b} within T_{set} shifted as a function of the changing thermal environment, producing narrow daily windows of time in which T_{b} could be within the preferred range while animals are above-ground (Figures 1 and 3). In March, T_{set} was only possible for a couple hours in mid-day, and as daily $T_{\text{e}}\text{s}$ progressively increased through the summer, T_{set} became possible only in early morning, or late at night. By the end of the activity season (July and August) it was almost never possible for *Gila* monsters to have T_{b} within T_{set} while active above-ground (Figures 1 and 4).

Body Temperatures (T_{b}) and Thermoregulation

Individuals were active above-ground when their $T_{\text{b}}\text{s}$ were between $19.9\text{ }^{\circ}\text{C}$ and $37.6\text{ }^{\circ}\text{C}$ (grand mean = $30.8\text{ }^{\circ}\text{C}$) and rested below-ground when their $T_{\text{b}}\text{s}$ were between $9.9\text{ }^{\circ}\text{C}$ and $35.1\text{ }^{\circ}\text{C}$ (grand mean = $25.5\text{ }^{\circ}\text{C}$). Mean monthly T_{b} peaked in July for both above- and below-ground environments (Figures 4 and 5), coinciding with the annual peak in $T_{\text{e}}\text{s}$ (Figure 1). Mean T_{b} was significantly different among seasons ($F_{2,14} = 180.1$,

$P < 0.0001$) as well as between above- and below-ground environments ($F_{1,9} = 15.6$, $P < 0.001$; Figure 6). T_b s increased over the activity season in both locations, and were always higher (on average) above-ground than below (Figure 6).

Overall, accuracy of thermoregulation (\bar{d}_b) was different among seasons ($F_{2,14} = 32.6$, $P < 0.001$) as well as between above- and below-ground environments ($F_{1,9} = 28.4$, $P = 0.0005$; Figure 6). Additionally, there was a significant interaction between season and location, and during the spring only, individuals were able to thermoregulate with higher accuracy by being active above-ground rather than by remaining inactive below-ground (Figure 6). Monthly estimates of \bar{d}_b for individuals in below-ground environments were lowest (highest accuracy) during May and October when T_b s were most often within T_{set} (Figures 1 and 5), and highest (lowest accuracy) during July in above-ground environments when T_b s were almost never within T_{set} (Figure 4) or able to be within T_{set} (Figure 1).

The proportion of T_b observations within T_{set} was different among seasons ($F_{2,14} = 6.2$, $P < 0.012$) as well as between above- and below-ground locations ($F_{1,9} = 5.3$, $P = 0.046$; Figure 6). Across seasons, individuals had 24% of T_b s within T_{set} while below-ground, but only 8% when above-ground. However, at certain times of the year, it wasn't possible to have T_b within T_{set} at all. During winter (November through mid-March) when lizards are inactive and underground hibernating, T_b s in above-ground environments were below T_{set} >95% of the time, and during the late summer months (June through August) T_b s were above T_{set} 85% of the time while below-ground, and 90% of the time while above-ground.

Effectiveness of Thermoregulation and Exploitation of T_{cs}

Regardless of season, Gila monsters were able to thermoregulate during above-ground activity. When $d_b < d_e$, individuals maintain T_b closer to T_{set} than would be expected if they were not actively regulating T_b , and were simply thermoconforming to the environment. Gila monsters had \bar{d}_b significantly lower than \bar{d}_e in each season (Table 1). Additionally, the difference between \bar{d}_e and \bar{d}_b can be considered the magnitude of thermoregulation (Blouin-Demers and Weatherhead 2001b), and Gila monsters became less effective at thermoregulating as the activity season progressed ($\bar{d}_e - \bar{d}_b$ became larger), and as environmental temperatures warmed \bar{d}_e became increasingly further from T_{set} .

Effectiveness of individual thermoregulation, E , was not significantly different among seasons (Table 1), and did not differ when it was calculated from all observations (E_{all} ; $F_{2,15} = 0.98$, $P = 0.38$), or just from those where the thermal environment was within T_{set} ($\bar{d}_e = 0$) and would permit the maintenance of T_b within T_{set} (E_{set} ; $F_{2,14} = 1.0$, $P = 0.37$).

Exploitation of the above-ground thermal environment was significantly different among seasons ($F_{2,11} = 11.0$, $P = 0.002$). During times of activity in which the thermal environment would permit T_b s to be within T_{set} (hourly $\bar{d}_e = 0$), active individuals actually had T_b s within T_{set} more than twice as much in the spring (63%) as during the summer (32 and 27% for early and late summer seasons).

Activity

Timing of activity shifted from diurnal in the spring to crepuscular in the early summer to almost completely nocturnal in the late summer (Figure 7). Gila monsters

were also most active in late summer (June and July) when T_{es} allowed activity for up to 17 h d⁻¹ (Figure 8). On a monthly basis, the amount activity (number of activity bouts) closely tracked opportunities for activity over the entire activity season. There was a significant effect of the amount of possible activity (hours per day) on the amount of actual activity ($F_{1,9} = 60.8$, $P < 0.00001$; Figure 8).

After July, however, above-ground activity decreased sharply, and individuals were about half as active in August as they had been in July, even though opportunities for activity increased by ~ 22% (13.9 h d⁻¹ possible in July and 16.9 h d⁻¹ in August). By early fall, Gila monster activity had nearly ceased (mean of 2.4 and 2.0 bouts per month for September and October respectively), even though opportunities for activity were at the annual peak (21.2 h d⁻¹ possible in September).

Discussion

Above-Ground Activity and Thermoregulation

In Gila monsters, the interplay between thermoregulation and activity is complex, but seems to be shaped largely by seasonal changes in temperature of the above-ground environment. Shifting T_{es} force activity and thermoregulation to covary negatively, and over the entire activity season, the ability to thermoregulate becomes increasingly difficult above-ground, even though opportunities for activity increase.

Individuals responded strongly to the increased opportunity for activity and though the relative magnitude of actual activity closely tracks that of possible activity (Figure 8), individuals still only use a fraction of the possible activity times in any given month or season. Beck (1990) reported that Gila monster activity bouts average 51 min in duration, and using that mean value, along with the observed seasonal rates of surface

activity (Table 1), we can estimate that Gila monsters use only 2.1 % of available activity times in Spring, 2.7% in early summer, and 3.0% in late summer. Gila monsters are, thus, extremely reclusive by choice during the active season, and not solely constrained by the thermal environment.

During the late summer and fall, activity decreased sharply even though possible activity increased to the annual peak of nearly 22 h d⁻¹ (September; Figure 8). This divergence is likely explained by the additional environmental constraints of prey availability. Nearly all prey species of Gila monsters nest in the spring and early summer (Beck 2005), and by late summer all nestlings have most likely fledged and food resources may be nearly entirely unavailable. Rather than use energy to search for prey above-ground, Gila monsters remain voluntarily inactive (to an even greater degree than they were in the activity season), and individuals are able conserve resources for use throughout the fall and winter (hibernation) seasons.

This voluntary reduction in activity has been shown to be an effective strategy in conserving energy and water resources when prey are unavailable or largely reduced in abundance. Some species of Australian varanid lizards become nearly completely inactive during the dry season when prey are scarce, and seasonal rates of energy and water use can drop to one-third that of the wet season (Christian and Weavers 1994, Christian et al. 1995).

Like activity, both T_b and thermoregulatory patterns of Gila monsters shifted with seasonal changes in the thermal environment. But unlike activity, which increased as T_e became higher, both the accuracy (\bar{d}_b) and magnitude ($\bar{d}_e - \bar{d}_b$) of thermoregulation decreased with increasing T_e (Table 1). However, effectiveness of thermoregulation (E ;

where 0 indicates random use of thermal environments and 1 indicating perfect thermoregulation) does not significantly vary among seasons, and is only marginally better when individuals are active at \bar{d}_e s within their preferred range (E_{set}) as compared to all times in which lizards were active (E_{all} ; Table 1). This may be related to the way in which Gila monsters behaviorally interact with their physical environment.

Gila monsters travel over somewhat large distances (up to 3 km travel distance per bout) and seldom stop to rest in shade or bask while above-ground. T_b s while active can vary by as much as 10 °C during a given bout (pers. obs.), and this variation is often the result of lizards emerging from cool underground shelters in early morning and continually warming until T_e forces them to seek refuge below-ground (Beck 1990). This further suggests that, for Gila monsters, behaviorally maintaining T_b within T_{set} is not of critical importance over short temporal windows, such as the duration of a single activity bout.

Gila monsters seem to be only modestly effective at thermoregulation (E) and effectiveness is similar among seasons (Table 1; mean $E_{all} = 0.52 \pm 0.07$). This may, in part, help explain the observed patterns of shifting seasonal activity, as observations of lizard activity closely track times in which the thermal environment is favorable for accurate thermoregulation (low \bar{d}_e ; Figure 1). By being active primarily when thermal conditions are benign (low \bar{d}_e), individuals would experience T_b s that were close to T_{set} , creating the opportunity for activity while simultaneously lessening the need for accurate thermoregulation.

This strategy, however, likely has effective limits, and as T_e s approach annual highs, maintaining T_b within T_{set} while active, might be largely impossible, at any time of the day. In late summer, our model predicts that above-ground activity would be possible for up to 15.3 h d^{-1} , however most of those opportunities for activity would be of rather poor quality ($\bar{d}_e = 11.6$ among all hours of day, 8.6 during hours of observed activity). In fact, predicted T_b s for July indicate that T_{set} would not be possible above-ground over virtually the entire month (Figure 1).

This difficulty in achieving T_{set} while active also reflects the degree to which Gila monsters actually use available thermal opportunities. While Gila monsters benefited by shifting activity times, they were increasingly unable to take advantage of the thermal environment late in the activity season. Individuals can exploit the thermal environment (E_x) by being active when both \bar{d}_e and $\bar{d}_b = 0$, thus negating the need to adjust T_b to achieve T_{set} . E_x decreased by approximately half in each successive six-week period of the activity season, despite temporal adjustments in activity times. Lizards were able to exploit the most opportunities during spring ($E_x = 0.63$) when activity was diurnal, but less during early and late summer (0.32 and 0.17) when activity shifted to crepuscular, and then shifted to nocturnal.

Below-Ground Thermoregulation

While we did not directly measure the thermal quality (\bar{d}_e) of below-ground habitats (burrows and caves), at least two lines of evidence suggest that conditions below-ground permit T_b to be within T_{set} more often than would be the case above-ground, and are therefore, environments of higher thermal quality (at least during the active season). First, while resting below-ground Gila monsters could have, under certain conditions,

constant T_b within T_{set} over entire days. Figure 3 shows that on a typical spring day, individuals under ground were never outside of T_{set} , except when choosing to bask, even though above-ground temperatures were outside T_{set} for ~20 h. In above-ground habitats, there was never an observed time period in which individuals could be within T_{set} for all, or even a majority of the day. Second, basking itself suggests that under-ground conditions permit lizards an adequate range of thermal opportunities, and individuals could select places within the burrow that would yield body temperatures within T_{set} , or individuals could simply shuttle between warmer and cooler places at the burrow opening to deep within the burrow.

The seasonal aspects of thermoregulation, while lizards are in below-ground refugia, are qualitatively similar to those during times of above-ground activity; lizards experience increasing T_b s and decreasing time within T_{set} across the activity season. In addition to providing physical refuge, underground burrows and caves also provide thermal refuge, and when *Gila* monsters are underground, monthly T_b s are almost always closer to T_{set} than when they are above-ground. An important exception occurs during early spring months (March and April), when above-ground T_e s first come into the range of T_{set} after winter hibernation. *Gila* monsters respond to these favorable conditions by basking in the openings of the hibernacula. They typically bask for two-to-three weeks before fully emerging from hibernation and commencing seasonal foraging activities.

Subterranean thermal regimes fluctuate much less both daily and annually underground than above-ground (Porter et al. 1973, Campbell and Norman 1998). This constancy of under-ground refuge sites is likely an important aspect of habitat selection of *Gila* monsters. Beck and Jennings (2003), showed that *Gila* monsters selected shelters,

in part, based on the quality of micro-environmental conditions within the refuge. Shelters selected by Gila monsters in New Mexico during the activity season allow lizards to achieve T_b s closer to T_{set} than those used during other seasons. When above-ground T_e s are at annual maxima and minima, Gila monsters choose burrows deep under-ground that allow T_b s to remain relatively constant and avoid extreme fluctuations of the above-ground environment.

While under-ground, Gila monsters can have relatively constant T_b for long periods (Porzer 1981, Beck 1990, Davis et al. 2008), and often T_b is within T_{set} (Figure 3). By being inactive under-ground, lizards are able to exploit available thermal regimes to a greater extent (during most months) than when active above-ground (Figures 4 and 5).

Attempting to maintain a relatively constant T_b is a feature of thermal biology of many species (McGinnis 1966, Christian et al. 1983, Zimmerman and Tracy 1989), and individuals use both microhabitat selection and behavioral thermoregulation to attenuate T_b variation (Avery 1982, Huey 1982). The putative advantage of regulating T_b at a constant level is that individuals may be able to optimize rates of physiological processes and aspects of performance necessary for routine, but important, activities, such as predator escape, prey detection, and digestion (Avery et al. 1982, Blouin-Demers and Weatherhead 2001a, Shine et al. 2002). Rather than overtly indulging in behavioral mechanisms such as shuttling between thermal suitable habitat micro-sites (Cowles and Bogert 1944), Gila monsters largely select refugia that would allow them passively to exploit preferred temperatures within the under-ground environment.

Undoubtedly, environmental constraints play an important role in the seasonal T_b and thermoregulatory shifts observed here. However, the potential contribution of

shifting seasonal preference for different T_b s on thermoregulation remains unknown. Christian and Bedford (1995) found seasonal differences in T_b selection in the tropical frillneck lizard *Chlamydosaurus kingii*, and by reducing T_{set} during the dry-hot part of the year, individuals were able reduce rates energy and water use.

While we did not test for seasonal differences in preferred T_b , shifting to a lower T_{set} during the hot and dry summer when food resources are low (as observed by Christian and Bedford 1995) would largely not help Gila monsters save energy and water. During the summer, Gila monsters likely inhabit the coolest microclimates available to them when resting in under-ground burrows, and they spend nearly all the time inactive. Lowering T_{set} would not allow them to achieve lower T_b s to save energy and water, except during the limited bouts of above-ground activity. However, lowering T_{set} while active might actually make thermoregulation more difficult, as activity T_b s would be even farther from T_{set} for the majority of the activity season.

Often, the assumption built into analyses that predict opportunities for activity and thermoregulation from operative temperatures and behavior, is that animals will consistently attempt to achieve their preferred range (Porter et al. 1973, Peterson 1987). In selecting a range of T_b s that allows optimal physiological performance (coadaptation hypothesis; Huey and Bennett 1987, Angilletta et al. 2006), individuals can benefit from precise thermoregulation.

While this assumption is likely true for small, highly active, or heliothermic ectotherms, other more reclusive and eurythermic species may not interact so closely with their thermal environment and may not behaviorally maintain T_b inside narrow limits. Other nocturnal and reclusive lizards (e.g. *Xantusia*, *Coleonyx*, and *Elgaria*) are

active at T_b s well outside T_{set} (Brattstrom 1965, Vance 1973, Kingsbury 1994) and some even abandon thermoregulation altogether when the costs of regulating T_b are high (Huey and Slatkin 1976, Blouin-Demers and Nadeau 2005).

Thus, for Gila monsters, behaviorally shifting to nocturnal activity late in summer allows individuals to take advantage of the last remaining opportunities for foraging before prey become unavailable in fall. And even though shifting the timing of activity allows the lizards to avoid lethal temperatures, their ability to thermoregulate is still dramatically reduced. Individuals seem to nearly, although not completely, abandon thermoregulation while active in late summer and they simply tolerate T_b far outside the preferred range (T_{set}). This eurythermic strategy therefore allows Gila monsters to trade-off a seasonal decrease in thermoregulatory performance for the potential benefit of increased prey acquisition.

Acknowledgements

We thank S. Cohen and S. Sheldon for help in recording T_b of Gila monsters and in placing, moving and downloading T_e models. K. Nussear, M. Sears, and E. Simandle provided technical assistance with electroplating models. Financial support was provided by the Clark County (Nevada) MSHCP, Biological Resources Research Center (UNR), and UNR Department of Biology and Program in Ecology, Evolution and Conservation Biology.

Literature Cited

- Adolph, S. C. 1990. Influence of behavioral thermoregulation on microhabitat use by two *Sceloporus* lizards. *Ecology* **71**:315-327.
- Adolph, S. C. and W. P. Porter. 1993. Temperature, activity, and lizard life histories. *The American Naturalist* **142**:273-295.
- Angilletta, M. J., A. F. Bennett, H. Guderley, C. A. Navas, F. Seebacher, and R. S. Wilson. 2006. Coadaptation: A unifying principle in evolutionary thermal biology. *Physiological and Biochemical Zoology* **79**:282-294.
- Avery, R. A. 1982. Field studies of body temperatures and thermoregulation. Pages 93-166 *in* C. Gans and F. H. Pough, editors. *Biology of the Reptilia*. Volume 12 Physiology C. Academic Press, New York.
- Avery, R. A., J. D. Bedford, and C. P. Newcombe. 1982. The role of thermoregulation in lizard biology: Predatory efficiency in a temperate diurnal basker. *Behavioral Ecology and Sociobiology* **11**:261-267.
- Bakken, G. S. 1992. Measurement and application of operative and standard operative temperatures in ecology. *American Zoologist* **32**:194-216.
- Bashey, F. and A. E. Dunham. 1997. Elevational variation in the thermal constraints on and microhabitat preferences of the greater earless lizard *Cophosaurus texanus*. *Copeia*:725-737.
- Beck, D. D. 1990. Ecology and behavior of the Gila monster in southwestern Utah. *Journal of Herpetology* **24**:54-68.
- Beck, D. D. 2005. *Biology of Gila Monsters and Beaded Lizards*. University of California Press, Berkeley.
- Beck, D. D. and R. D. Jennings. 2003. Habitat use by Gila monsters: the importance of shelters. *Herpetological Monographs* **17**:111-129.
- Blouin-Demers, G. and P. Nadeau. 2005. The cost-benefit model of thermoregulation does not predict lizard thermoregulatory behavior. *Ecology* **86**:560-566.
- Blouin-Demers, G. and P. J. Weatherhead. 2001a. An experimental test of the link between foraging, habitat selection and thermoregulation in black rat snakes *Elaphe obsoleta obsoleta*. *Journal of Animal Ecology* **70**:1006-1013.

- Blouin-Demers, G. and P. J. Weatherhead. 2001b. Thermal ecology of black rat snakes (*Elaphe obsoleta*) in a thermally challenging environment. *Ecology* **82**:3025-3043.
- Bogert, C. M. and R. Martín del Campo. 1956. The Gila Monster and Its Allies. *Bulletin of the American Museum of Natural History* **109**:1-238.
- Brattstrom, B. H. 1965. Body temperatures of reptiles. *American Midland Naturalist* **73**:376-422.
- Brown, G. P. and P. J. Weatherhead. 2000. Thermal ecology and sexual size dimorphism in northern water snakes, *Nerodia sipedon*. *Ecological Monographs* **70**:311-330.
- Campbell, G. S. and J. M. Norman. 1998. *An Introduction to Environmental Biophysics*. Springer-Verlag, New York.
- Christian, K. and B. Weavers. 1994. Analysis of the activity and energetics of the lizard *Varanus rosenbergi*. *Copeia* **1994**:289-295.
- Christian, K. A. and G. S. Bedford. 1995. Seasonal changes in thermoregulation by the frillneck lizard, *Chlamydosaurus kingii*, in tropical Australia. *Ecology* **76**:124-132.
- Christian, K. A., L. K. Corbett, B. Green, and B. W. Weavers. 1995. Seasonal activity and energetics of two species of varanid lizards in tropical Australia. *Oecologia* **103**:349-357.
- Christian, K. A. and C. R. Tracy. 1985. Physical and biotic determinants of space utilization by the Galapagos land iguana (*Conolophus pallidus*). *Oecologia* **66**:132-140.
- Christian, K. A., C. R. Tracy, and W. P. Porter. 1983. Seasonal shifts in body temperature and use of microhabitats by Galapagos land iguanas (*Conolophus pallidus*). *Ecology* **64**:463-468.
- Christian, K. A., C. R. Tracy, and C. R. Tracy. 2006. Evaluating thermoregulation in reptiles: An appropriate null model. *American Naturalist* **168**:421-430.
- Christian, K. A. and B. W. Weavers. 1996. Thermoregulation of monitor lizards in Australia: An evaluation of methods in thermal biology. *Ecological Monographs* **66**:139-157.
- Cowles, R. B. and C. M. Bogert. 1944. A preliminary study of the thermal requirements of desert reptiles. *Bulletin of the American Museum of Natural History* **83**:265-296.

- Davis, J. R., E. N. Taylor, and D. F. DeNardo. 2008. An automated temperature-based option for estimating surface activity and refuge use patterns in free-ranging animals. *Journal of Arid Environments* **72**:1414-1422.
- Dzialowski, E. M. 2005. Use of operative temperature and standard operative temperature models in thermal biology. *Journal of Thermal Biology* **30**:317-334.
- Gienger, C. M. 2003. Natural history of the Gila monster in Nevada. Unpub. M.S. Thesis. University of Nevada, Reno, Reno.
- Grant, B. W. and A. E. Dunham. 1988. Thermally imposed time constraints on the activity of the desert lizard *Sceloporus merriami*. *Ecology* **69**:167-176.
- Hertz, P. E., R. B. Huey, and R. D. Stevenson. 1993. Evaluating temperature regulation by field-active ectotherms: The fallacy of the inappropriate question. *American Naturalist* **142**:796-818.
- Huey, R. B. 1982. Temperature, Physiology, and the Ecology of Reptiles. Pages 25-91 in C. Gans and F. H. Pough, editors. *Biology of the Reptilia* 12C. Academic Press, New York.
- Huey, R. B. and A. F. Bennett. 1987. Phylogenetic studies of coadaptation: Preferred temperatures versus optimal performance temperatures of lizards. *Evolution* **41**:1098-1115.
- Huey, R. B. and P. E. Hertz. 1984. Is a jack-of-all-temperatures a master of none? *Evolution* **38**:441-444.
- Huey, R. B. and M. Slatkin. 1976. Costs and benefits of lizard thermoregulation. *Quarterly Review of Biology* **51**:363-384.
- Kingsbury, B. A. 1994. Thermal constraints and eurythermy in the lizard *Elgaria multicarinata*. *Herpetologica* **50**:266-273.
- Lowe, C. H., C. R. Schwalbe, and T. B. Johnson. 1986. *The Venomous Reptiles of Arizona*. Arizona Game and Fish Department, Phoenix.
- McGinnis, S. M. 1966. *Sceloporus occidentalis*: Preferred body temperature of the western fence lizard. *Science* **152**:1090-1091.
- Niewiarowski, P. H. 2001. Energy budgets, growth rates, and thermal constraints: Toward an integrative approach to the study of life history variation. *The American Naturalist* **157**:421-433.

- Peterson, C. R. 1987. Daily variation in the body temperatures of free-ranging garter snakes. *Ecology* **68**:160-169.
- Porter, W. P., J. W. Mitchell, W. A. Beckman, and C. B. DeWitt. 1973. Behavioral implications of mechanistic ecology: Thermal and behavioral modeling of desert ectotherms and their microenvironment. *Oecologia* **13**:1-54.
- Porter, W. P. and C. R. Tracy. 1983. Biophysical Analyses of Energetics, Time-space Utilization, and Distributional Limits. Pages 55-83 in R. B. Huey, E. R. Pianka, and T. W. Schoener, editors. *Lizard Ecology: Studies of a Model Organism*. Harvard University Press, Cambridge, Massachusetts, USA.
- Porzer, L. M. 1981. Movement, behavior, and body temperature of the Gila monster (*Heloderma suspectum*) in Queen Creek, Pinal County, Arizona. Unpub. M.S. Thesis. Arizona State University, Tempe.
- Scott, J. R., C. R. Tracy, and D. Pettus. 1982. A biophysical analysis of daily and seasonal utilization of climate space by a montane snake. *Ecology* **63**:482-493.
- Sears, M. W. 2005. Geographic variation in the life history of the sagebrush lizard: the role of thermal constraints on activity. *Oecologia* **143**:25-36.
- Shine, R., L. X. Sun, M. Kearney, and M. Fitzgerald. 2002. Thermal correlates of foraging-site selection by Chinese pit-vipers (*Gloydius shedaoensis*, Viperidae). *Journal of Thermal Biology* **27**:405-412.
- Stahnke, H. L. 1952. A note on the food habits of the Gila monster, *Heloderma suspectum* Cope. *Herpetologica* **8**:64.
- StatSoft. 2001. Statistica (data analysis software system), version 6. www.statsoft.com.
- Tracy, C. R. 1982. Biophysical Modeling of Reptilian Physiology and Ecology. Pages 275-321 in C. Gans and F. H. Pough, editors. *Biology of the Reptilia*. Academic New York.
- Tracy, C. R. and K. A. Christian. 1986. Ecological relations among space, time, and thermal niche axes. *Ecology* **67**:609-615.
- Vance, V. J. 1973. Temperature preference and tolerance in the gecko, *Coleonyx variegatus*. *Copeia* **1973**:615-617.
- Walsberg, G. E. and B. O. Wolf. 1996. An appraisal of operative temperature mounts as tools for studies of ecological energetics. *Physiological Zoology* **69**:658-681.
- Woodson, W. W. 1949. Mammal diet of *Heloderma*. *Herpetologica* **5**:152.

Zimmerman, L. C. and C. R. Tracy. 1989. Interactions between the environment and ectothermy and herbivory in reptiles. *Physiological Zoology* **62**:374-409.

Table 1. Seasonal indices for above-ground activity, body temperatures, and thermoregulation.

	Spring	Early Summer	Late Summer	Statistics	Significant Post-Hoc Differences
Timing of Activity	Mid-Day	Crepuscular	Nocturnal		
Activity Possible (h d ⁻¹)	7.4	14.9	15.3		
Activity (bouts week ⁻¹)	1.3	3.3	3.9	F _{2,16} = 24.1, P < 0.001	S<ES, S<LS
Activity T _b (°C)	28.0	30.5	33.1	F _{2,14} = 15.6, P < 0.001	S<LS
T _{set} possible (h d ⁻¹)	3.3	3.3	1.6		
% T _b obs. within T _{set}	19.6	3.8	0.7	F _{2,14} = 6.2, P < 0.012*	S>ES, S>LS
\bar{d}_e (all hours)	4.5	6.9	11.6		
\bar{d}_e (hours of activity)	1.5	4.3	8.6		
\bar{d}_b	1.8	3.6	6.0	F _{2,14} = 32.6, P < 0.001	S<ES, S<LS, ES<LS
$\bar{d}_b < \text{mean } \bar{d}_e$ **	Yes	Yes	Yes	t ₇ = 6.7, P < 0.001 (S) t ₉ = 12.2, P < 0.0001 (ES) t ₈ = 14.3, P < 0.00001 (LS)	
$\bar{d}_e - \bar{d}_b$	2.7	3.2	5.8	F _{2,15} = 23.9, P < 0.00001	S<LS, ES<LS
<i>E</i> _{all}	0.60	0.47	0.50	F _{2,15} = 0.98, P = 0.38	
<i>E</i> _{set}	0.47	0.41	0.48	F _{2,14} = 1.0, P = 0.37	
<i>E</i> _x (obs. T _b within T _{set} , $\bar{d}_e = 0$)	0.63	0.32	0.17	F _{2,11} = 11.0, P = 0.002	S>ES, S>LS, ES>LS

Notes:

Descriptive data and values of environmental parameters are reported for comparison between seasons (described in text).

Values are mean of individual means. Tests and Tukey HSD Post-hoc differences at $\alpha = 0.05$; * Data were arcsine transformed prior to analysis; ** one-tailed paired t-test.

Table 2. Below-ground body temperatures, and indices of thermoregulation.

	Spring	Early Summer	Late Summer	Statistics	Post-Hoc Differences
Resting T_b (°C)	21.7	27.1	29.0	$F_{2,16} = 84.3, P < 0.0001$	S<ES, S<LS, ES<LS
% T_b obs. within T_{set}	23.9	31.6	15.9	$F_{2,16} = 6.2, P < 0.01^*$	S<ES
\bar{d}_b	3.1	1.2	2.1	$F_{2,16} = 24.0, P < 0.0001^*$	S<ES, S<LS, ES<LS

* Data were arcsine transformed prior to analysis.

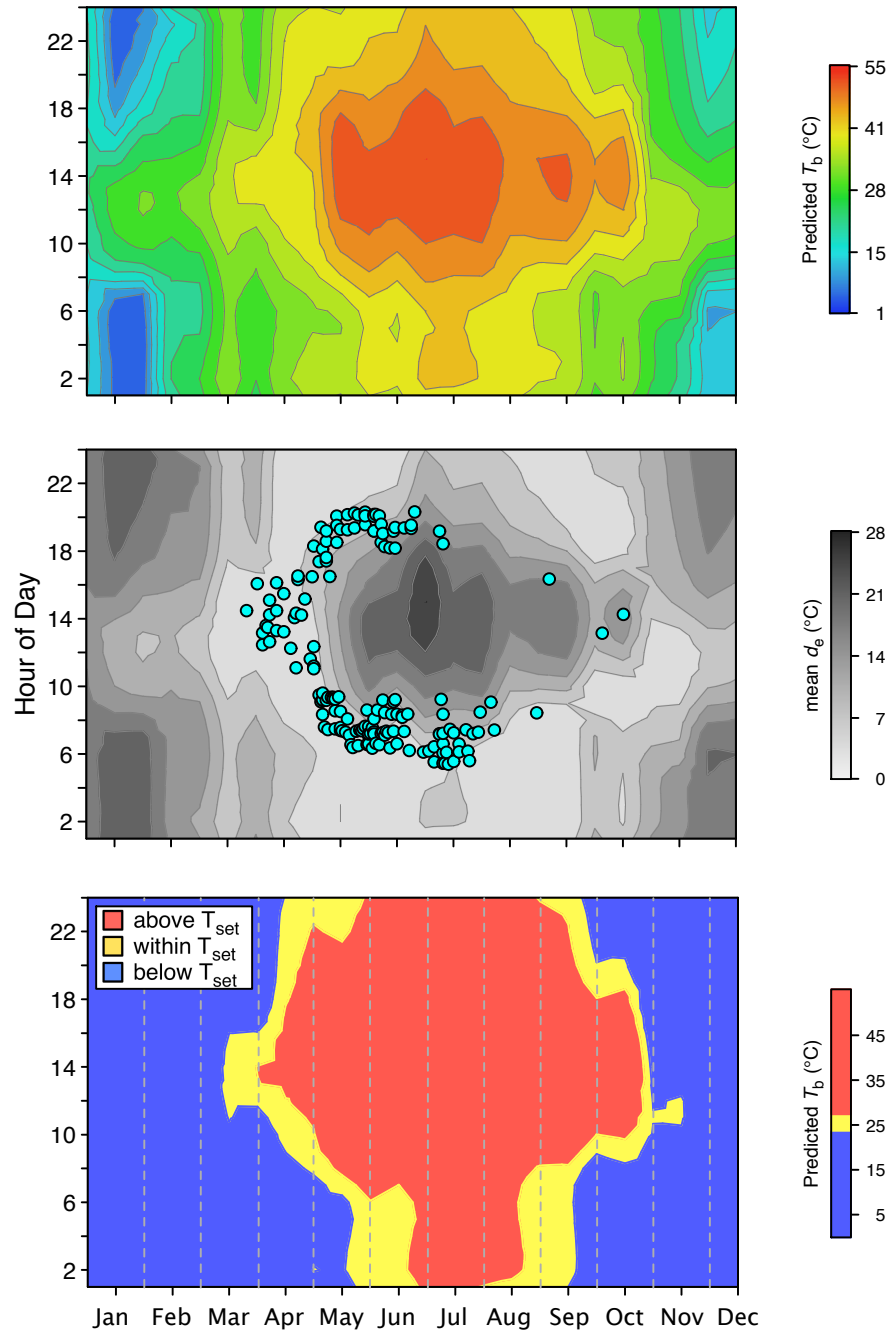


Figure 1. Contour plots showing (A) mean achievable T_b , (B) thermal quality of habitat (\bar{d}_e) and observations of activity (circles), and (C) possibilities for Gila monsters to achieve T_{set} while above-ground.

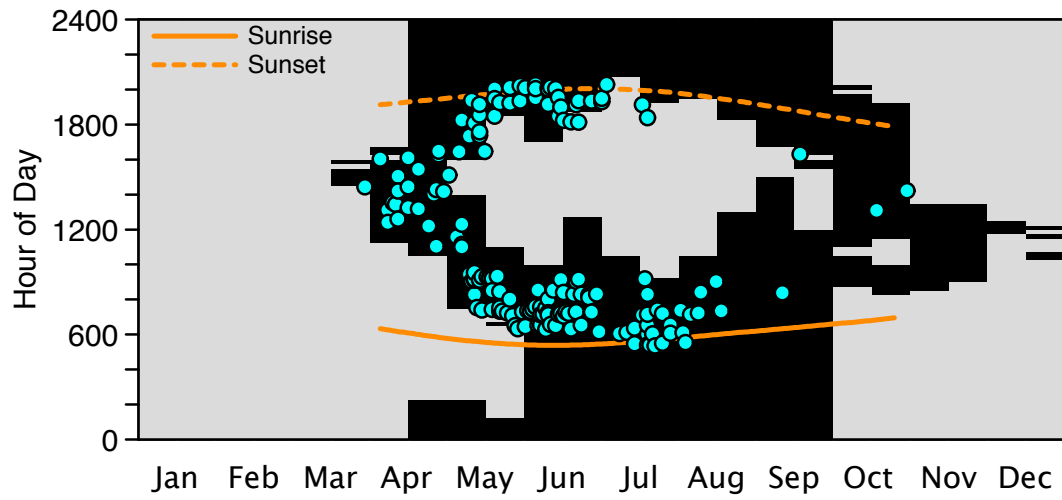


Figure 2. Thermal constraints on activity in Gila monsters. Black areas indicate times of day and month that would be predicted to be suitable for activity (T_{bs} within $V T_{max}$ and $V T_{min}$), gray indicates those times that would not be suitable.

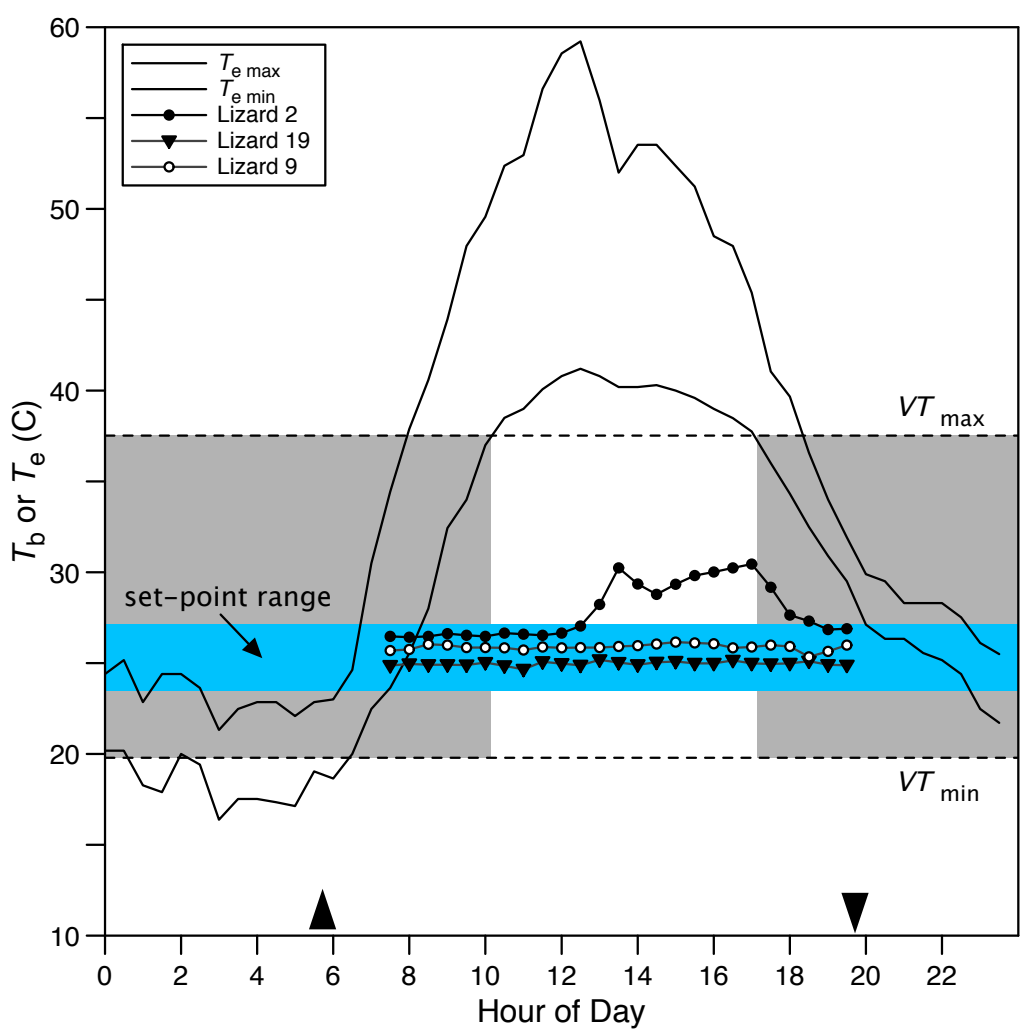


Figure 3. Operative temperatures (T_e) and body temperature (T_b) while Gila monsters are resting in below-ground refugia on a typical summer day (6/15/2003). Activity on this day would be possible for ~17 h (gray boxes). Individuals stay within the set-point range (T_{set}) for nearly the entire day while underground, but if they were above-ground, they would be outside T_{set} for ~20 hours. Lizard 2 was observed basking at the opening of the refuge site in the afternoon yielding T_b s outside T_{set} until returning deep underground. Triangles indicate time of sunrise-sunset.

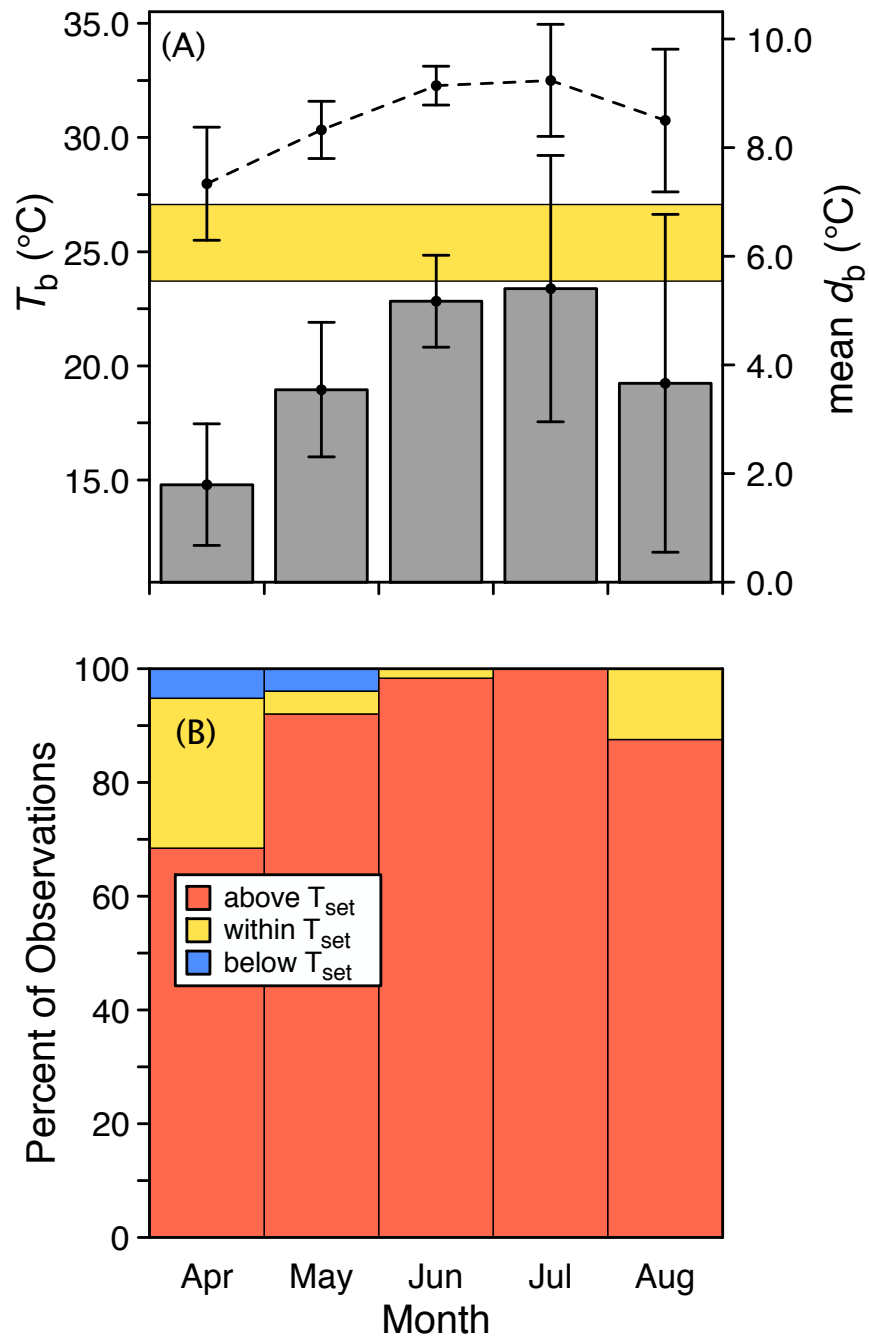


Figure 4. (A) Mean of individual mean T_b (dashed line) and \bar{d}_b (bars) while active above-ground ($\pm 1SD$). (B) Observations of activity T_b s relative to the preferred range, T_{set} .

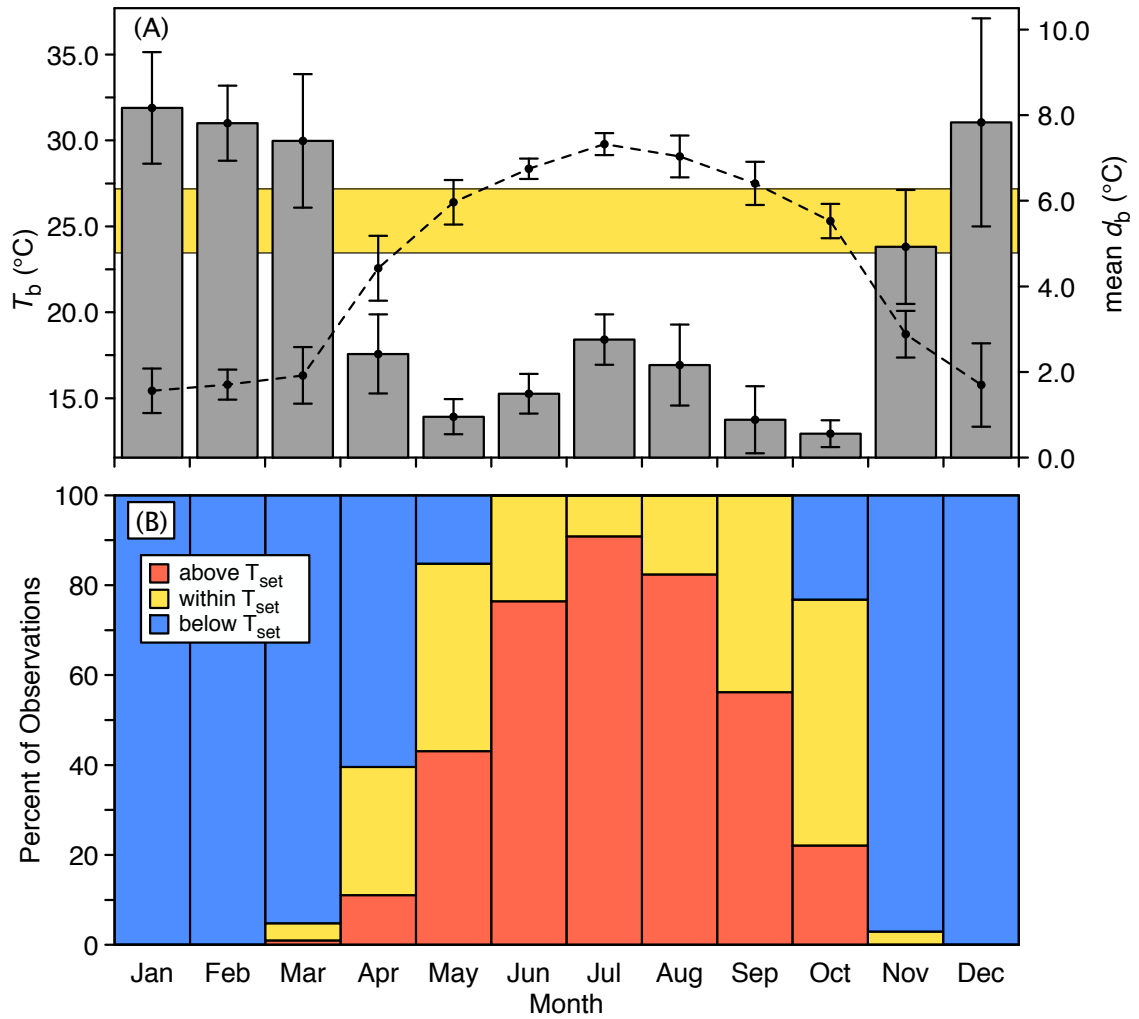


Figure 5. (A) Mean of individual mean T_b (dashed line) and \bar{d}_b (bars) while resting underground (± 1 SD). (B) Observations of T_b relative to the preferred range, T_{set} .

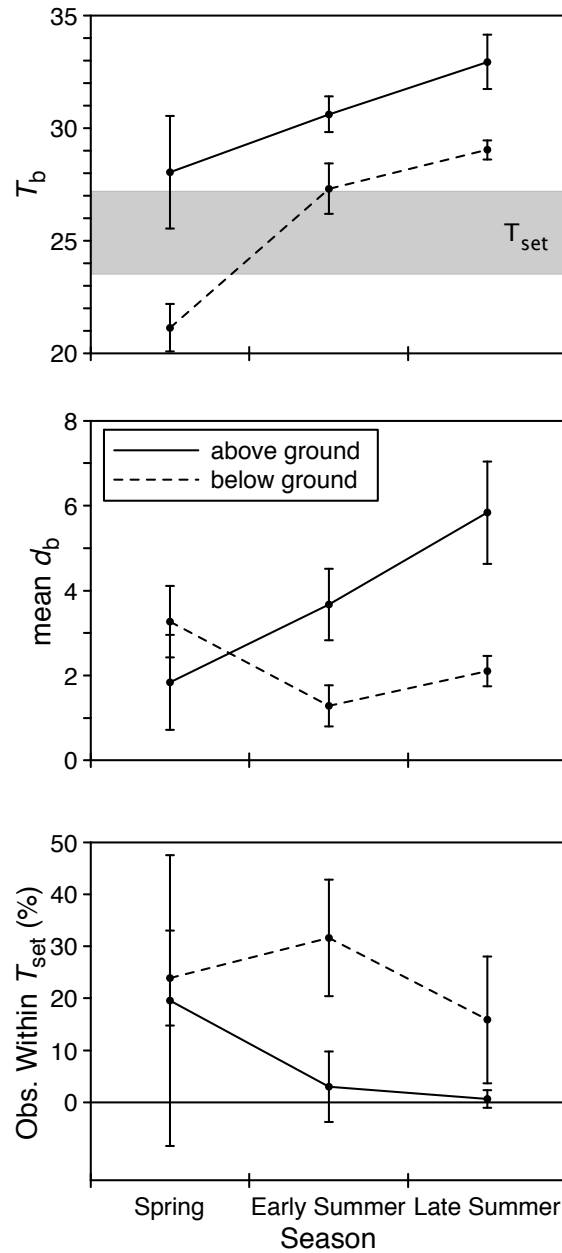


Figure 6. T_b , \bar{d}_b , and % observations within T_{set} between above and below-ground locations, and among seasons. Data are mean of individual means (± 1 SD).

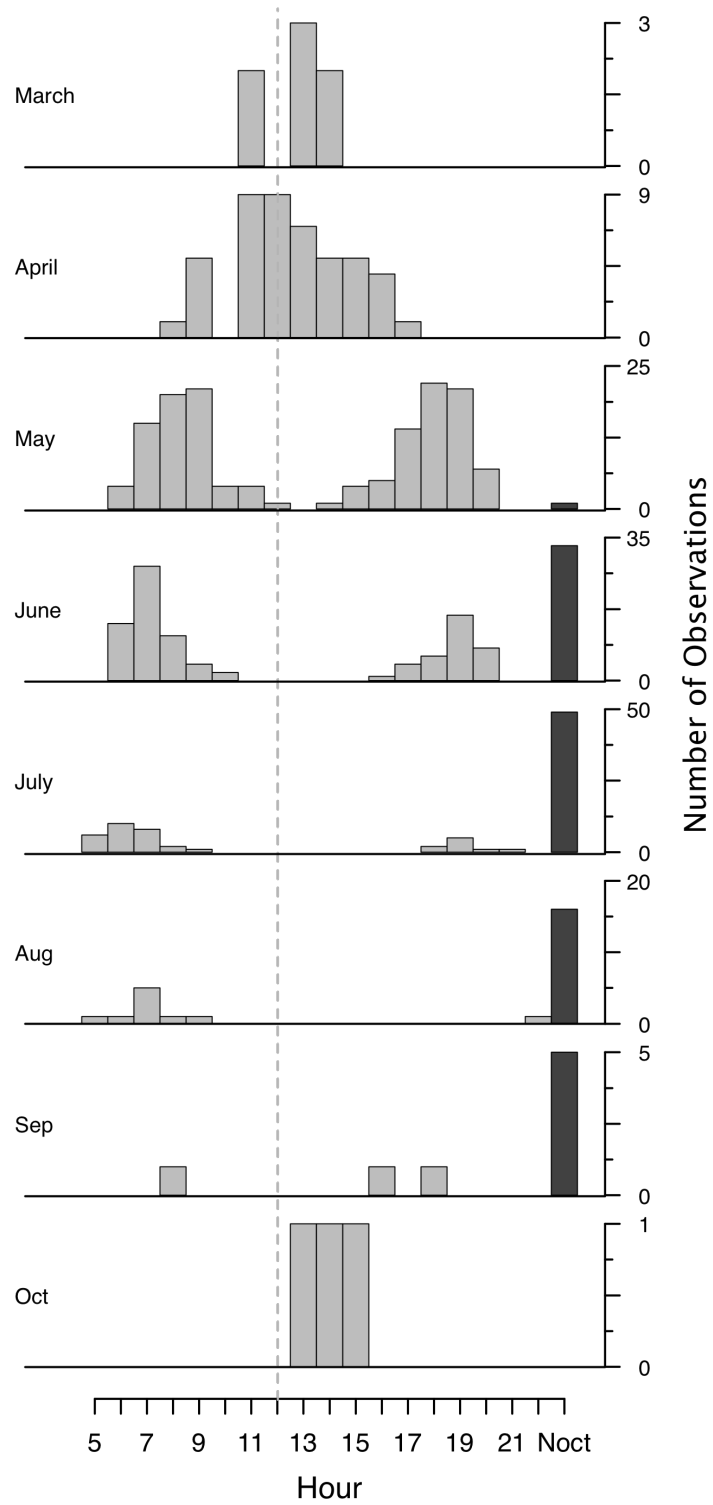


Figure 7. Annual distribution of above-ground activity of Gila monsters (2002-2004).

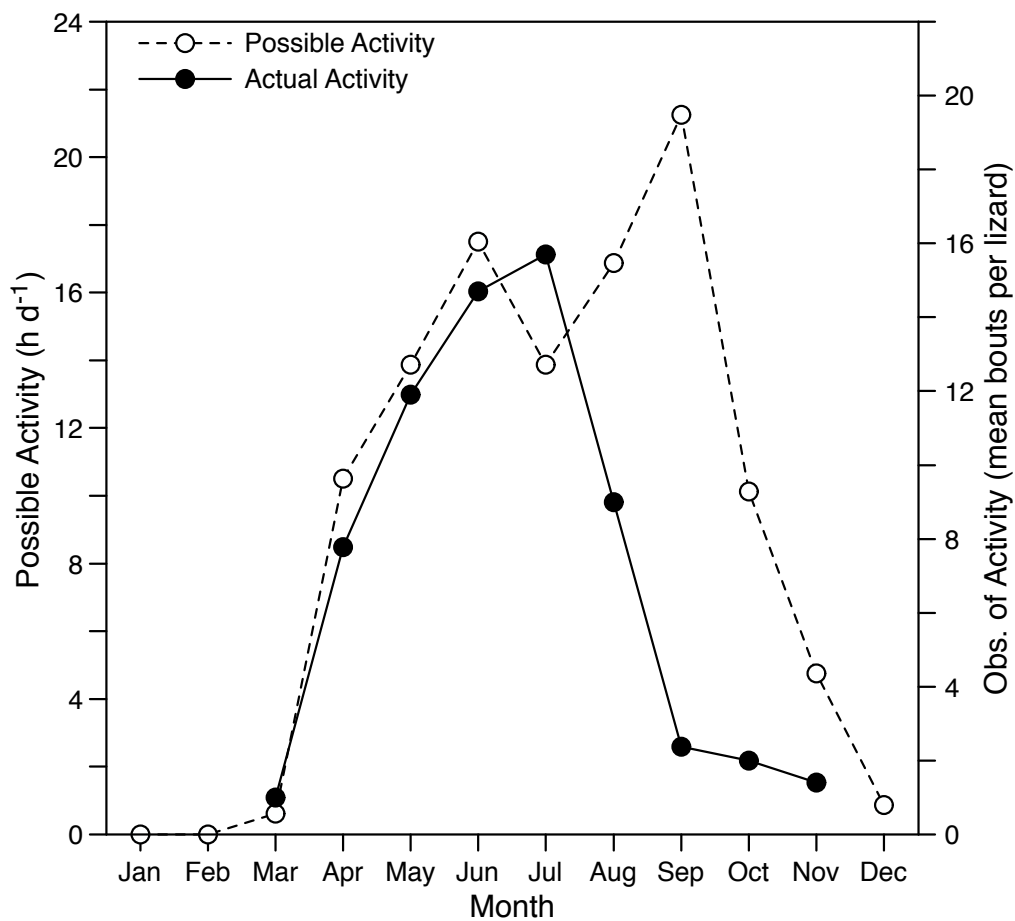


Figure 8. Monthly opportunities for above-ground activity and observations of actual activity for Nevada Gila monsters (2003).

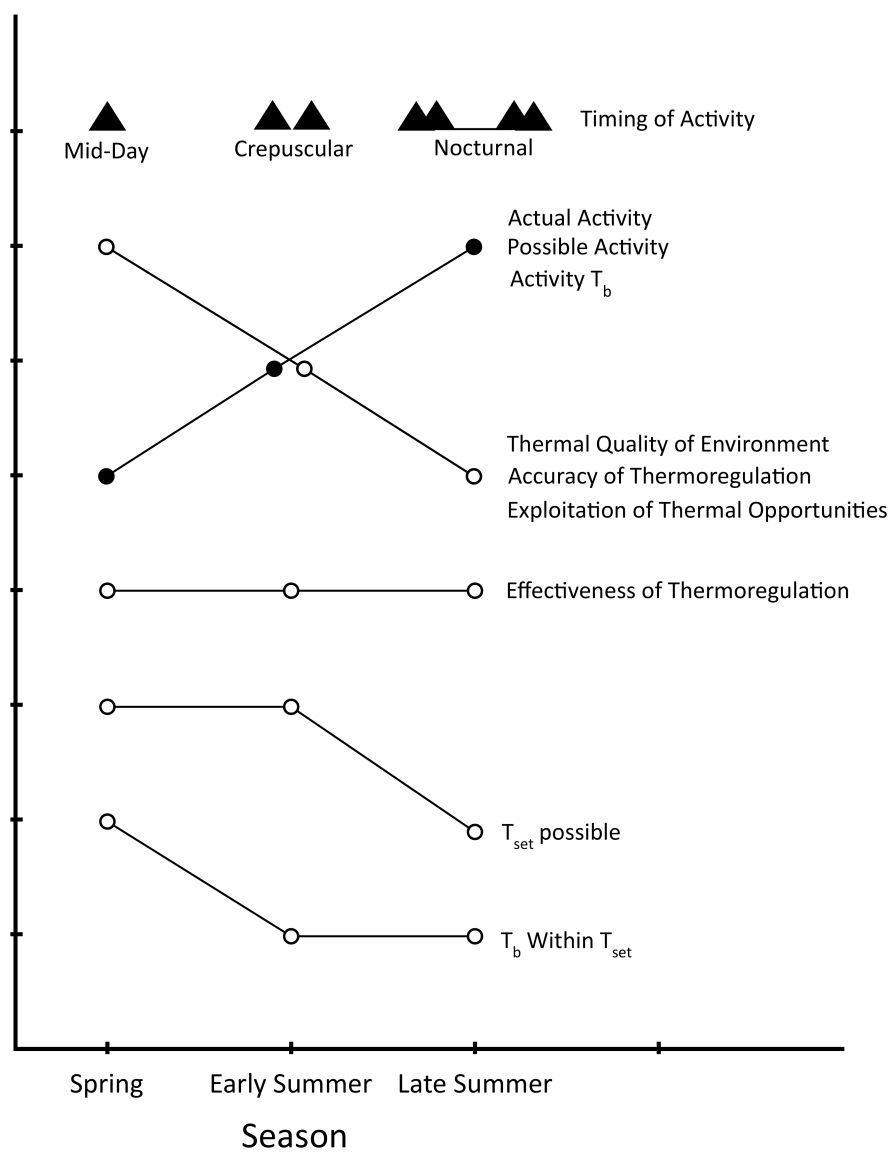


Figure 9. Graphical summary of seasonal trade-offs between activity and thermoregulation of Gila monsters. Analyses are given in Table 1.

Seasonal Use of Energy and Water By Gila Monsters

Abstract

Field metabolic rates (FMR) and water influx rates (WIR) were measured in Gila monsters (*Heloderma suspectum*) in all four seasons in southern Nevada. Gila monsters are extremely reclusive, and because they are inactive underground for most of year, they have rates of metabolism and water loss that are less than half of that expected generally for lizards their size. Seasonal rates were highest in early summer, coinciding with the peak of feeding and breeding activities. The amount of above-ground foraging and locomotor activity influenced both FMR and WIR, and approximately two-thirds of the annual energy budget is partitioned to activity and one-third to resting metabolism. Body mass of individuals influenced FMR and underground T_b was significantly related to WIR. Rates of resting metabolism vary geographically as a function of mass and T_b , and the differences in energetic requirements likely can explain observed ecological and life history differences among populations.

Introduction

Evaluating how organisms use and partition internal resources is one of the most basic goals of physiological ecology (McNab 2002, Karasov and Martínez del Río 2007). Resource availability, particularly the abundance and distribution of food and water, can impose strong limitations and opportunities on organisms. Those limitations, in turn, determine how animals can interact with their environment, and the limitations can shape the varied strategies animals use to acquire and exploit resources (Glasser 1984). In harsh and unpredictable environments it is often unusually difficult for organisms to persist,

and they may face intense selective pressure for development of novel adaptations and strategies for survival (Boyce 1979, Waterman 1999, 2001).

Gila monsters (*Heloderma suspectum*), are an interesting species in which to investigate organismal capacity for survival in unpredictable environments. They have a unique natural history, which is largely a function of these lizards' ability to tolerate large fluctuations in availability of food and water (Beck 2005), and several aspects of their behavior, physiology, and morphology provide interesting examples of adaptation to life under variable and unpredictable environmental conditions.

Gila monsters inhabit dry hot-desert and thornscrub areas of the American southwest and northwestern México, habitats that have extreme seasonal and annual variation in temperature, precipitation, and food availability. This species specializes as a nest predator that consumes the altricial nestlings of rodents and lagomorphs along with unhatched eggs of ground-nesting birds, snakes, lizards, and tortoises. Because such food items are relatively uncommon in both space and time, *Gila monsters* must forage over large areas to find food, and capitalize when these food items are available. The lizards travel as much as three kilometers during a single foraging bout (pers. obs.) and typically consume the entire contents of a nest once it is discovered. A single meal can be nearly one-third the body mass of the lizard itself (Stahnke 1950, 1952, Beck 1990), and three or four such large meals could potentially meet the entire yearly energetic demands of an individual (Beck 1986).

Gila monsters have low basal metabolic rates, needing only about half as much energy at rest as would be expected for generalized lizard species of similar size (Beck and Lowe 1994). There are no detectable differences between the sexes in resting

metabolism (Beck and Lowe 1994), or evaporative water loss (DeNardo, pers. comm.), although males seem to have a higher aerobic capacity (VO_2 max) and physical endurance (Beck et al. 1995). Male Gila monsters intensely fight one another for mating opportunities with females, and single combat events can last more than 12 hours (pers. obs.), pushing the combatants to the point of physical exhaustion.

Because of their frugal use of energy and their high digestive efficiency (~77 to 92%; Beck 1986, Wegscheider 1998), when food is available these lizards can quickly add body mass, and they often develop conspicuous fat stores that give them their distinct rotund body and sausage-shaped tail. They are also adept at storing water, and unlike many reptiles (including birds, crocodilians, and snakes), Gila monsters possess a urinary bladder (Beuchat 1986) that can be used as a hydric reservoir. During periods of water scarcity and dehydration, Gila monsters are thought to draw water from bladder urine to help regulate plasma osmolality (Davis and DeNardo 2007).

For a desert lizard, Gila monsters prefer unusually cool body temperatures. Their mean activity body temperature (T_b) is around 30°C, and they rarely voluntarily tolerate body temperatures greater than ~37°C, as high temperatures (>35°C) are physiologically stressful and may impair performance. Captive individuals have been observed to increase evaporative water loss (measured as cloacal loss; the primary avenue of water loss at high temperatures) by more than three orders of magnitude at body temperatures between 35 and 40°C (DeNardo et al. 2004).

Interestingly, Gila monsters are most active in summer, when environmental temperatures are near annual maxima (Gienger 2003, Beck 2005). But, in general, they prefer to spend the majority of their time underground in burrows, where temperatures

are often cooler. As summer temperatures become higher, Gila monsters seek successively deeper shelters in order to maintain resting T_{bs} at $\sim 27\text{-}30^\circ\text{C}$ (Beck and Jennings 2003, Gienger 2003). On an annual basis, Gila monsters spend more than 95% of the year inactive underground (Porzer 1981, Beck 1990) and more than half the year at T_{bs} less than 25°C , a strategy that allows considerable savings in water and energy.

These unique attributes of Gila monster natural history provide somewhat contradictory expectations as to how energy and water should be allocated as physiological resources. Gila monsters have low resting metabolism and high capacity for fat and water storage, both of which are conservation strategies. This suggests that the lizards should be frugal in their use of water and energy, and they should have strategies to minimize loss of those resources. But, this may not always be the case, as individuals likely squander both energy and water to travel long distances during the hottest parts of the year as they search for food that is both difficult to find and only sporadically available.

Additionally, Gila monsters are geographically distributed to some of the hottest and driest areas of North America, yet their preference for low activity T_{bs} , intolerance of high environmental temperatures, and potentially high rates of water loss relegates foraging opportunities to limited times of both day and year (Chapter 2). It seems, then, as though the physiology of Gila monsters is an unexpected strategy for the desert environments in which they occur.

Our goals here are to report on assessments of how these unique and seemingly conflicting aspects of Gila monster natural history determine the species use of energy and water as physiological resources. We first test the hypothesis that Gila monsters have

seasonal differences in the use of energy and water. Because these lizards live in seasonal environments, where the availability of food and water is inherently variable, Gila monsters should temporally adjust their use of these resources and conserve resources for times when they are not available (e.g. hibernation, seasonal drought).

Second, we test the hypothesis that energy and water expenditure between the sexes, possibly due to direct and indirect costs of reproduction. Although females have obvious and direct physiological expenditures as a result of producing offspring, males may have much more subtle reproductive costs. Male-male combat activities of Gila monsters are often direct physical competitions for mates (pers. obs.), and they should result in high, but temporary, levels of energy and water expenditure. Those expenditures may be limited to the reproductive season, which is the only time of year that combat has been observed. Lastly, we examine the roles of mass, T_b , and activity in determining expenditures in water and energy and extend those analyses to determine how Gila monsters budget energy and allocate it to the mutually exclusive components of activity and rest.

Methods

Study Site

Gila monsters were studied from March 2000 to August 2004 in the Mojave Desert, approximately 10 km west of Lake Mead, Clark County, Nevada (36°N and 114°W), with an elevation range of 550-650 m (Gienger 2003). Vegetation at the site is typical Mojave Desert scrub (Brown 1994), and the dominant plants include creosote bush (*Larrea tridentata*), bursage (*Ambrosia dumosa*), and burro brush (*Hymenoclea salsola*).

The Mojave is very seasonal (Rundel and Gibson 1996, Hereford et al. 2006) with respect to both temperature and precipitation. Climatic conditions at the study site are typical of the Mojave; site temperatures are highest in late summer (July and August; mean air temperature = 33.3°C) and lowest in winter (December and January mean air temperature = 8.8°C). Precipitation falls primarily in the form of winter rain and, to a much lesser degree, from small storms of monsoon rain in mid-summer (Esler and Rundel 1999).

Telemetry, Activity, and Spatial Habitat Use

Lizards were captured in the field, transported to a nearby field laboratory, anesthetized, and surgically implanted with a calibrated temperature-sensing radio transmitter (Telonics CHP-100L). After surgery, lizards were held for 24-48 h, offered raw chicken egg and water (*ad libitum*), and then released at the site of capture. Individuals were relocated and T_b s were recorded two to four times a day during the activity season (March through August) and then two to four times a week during the fall and winter months (September through February). Locations of shelter sites and active lizards were recorded using a handheld global positioning system (GPS).

Field Metabolic Rate (FMR) and Water Flux

Field metabolic rates (CO₂ production) and water flux of Gila monsters were measured using the doubly labeled water (DLW) method (Lifson and McClintock 1966, Speakman 1997). Individuals were weighed and injected intra-peritoneally with 1.2 mL of sterile water containing 95.5 atom percent ¹⁸O and 400 microcuries of tritiated water. After allowing 2-4 hours for isotopes to equilibrate with body water, a blood sample (~0.1 mL) was taken from either the orbital sinus, caudal vein, or a toe prick to estimate

total body water volume via ^{18}O dilution (Speakman 1997), and to initiate the measurement period for both FMR and water flux. Blood samples were taken from three individuals prior to enrichment to quantify natural background concentrations of both isotope tracers.

Individuals were re-captured for blood sampling and measurement of body mass at approximately two to three-week intervals during the activity season. Samples were collected opportunistically from animals active above ground, and sample intervals became much longer late in the activity season (hot summer and fall) when lizards were only infrequently active and available for sampling. We grouped samples by season as follows; spring (March through 15 May), early summer (16 May through June), late summer (July through September), and fall-winter (October through emergence from hibernation).

After one continuous year of sampling, which typically yielded four to six sampling periods for each lizard, levels of isotopic enrichment had become too low to be accurately measured, and it was necessary to re-enrich individuals. We then injected 0.5 mL of the same injection solution into each captured individual, and again took a blood sample after the equilibration period. This re-enrichment allowed us to extend measurements of FMR, and water flux through a second activity season, along with the single intervening hibernation period, and provided continuous data from nearly 18 months in the field.

Blood samples were stored in flame sealed heparinized capillary tubes and refrigerated until being transported to UCLA for analysis. Samples were vacuum distilled and isotope concentrations were determined by liquid scintillation counting (^3H) and

proton activation (^{18}O ; Wood et al. 1975, Nagy 1983). Rates of CO_2 production were calculated using Eq. 2 in Nagy (1980). Field CO_2 estimates were converted to energy units using a thermal equivalent of $25.7 \text{ kJ L}^{-1} \text{ CO}_2$, with an assumed respiratory quotient of 0.75 (Nagy 1982a). We also assumed that any changes in body mass over a sampling period were linear, and we used mean body mass of each sampling period in calculations of physiological rates.

Measured rates of energy and water use for Gila monsters during their active seasons were compared to predicted values derived from allometric equations. We compared FMR to the predictive equations for reptiles, lizards (generally), and desert lizards of Nagy et al. (1999) and water flux to the equations for arid and semi-arid reptiles and lizards of Nagy (1982b).

Energy Budget and Water Economy Calculations

We partitioned FMR over each sampling period into the energetic costs of resting in below-ground shelters, (total resting metabolism; TRM), and the energetic costs of being active above-ground (activity respiration; AR; Benabib and Congdon 1992). TRM under field conditions was estimated using a multiple regression equation calculated from Table 5 of Beck (1986) relating T_b and body mass (BM) to resting metabolic rate (RMR) of Gila monsters ($\text{RMR} = \log_{10} \text{SMR} = -1.340 + 0.042 * T_b + 0.460 * \log_{10} \text{mass}$; $r^2 = 0.92$).

For each sample period, we used mean below-ground T_b and mean body mass of the individual to calculate TRM. Thermal variation is low for individuals resting underground and T_b s usually change less than 2°C over the course of a day while lizards are in burrows or caves (Beck 1986, Davis et al. 2008).

AR was calculated by subtracting TRM from FMR (Benabib and Congdon 1992, Christian et al. 1997); AR can be considered the combined energetic costs associated with all activities, including locomotion, foraging, digestion, territory defense, mate acquisition, and with any growth and reproduction (Bennett 1978, van Marken Lichtenbelt et al. 1993). We also calculate percent of field costs associated with activity (%AR) as $AR/FMR*100$.

Because water loss and metabolism are linked as physiological processes, for each sampling period, we calculated Water Economy Index (WEI; Nagy 2004) as a measure of the efficiency of water conservation relative to the rate of energy use. WEI is reported in units of mL water used per kJ of energy metabolized (Nagy 2004). Natural selection may act on these rates independently, but this index provides a method to examine water use in a way that accounts for different levels of metabolism.

Statistical Analysis

Data were checked for compliance with assumptions for parametric statistics, and were either log or arcsin (for proportional data) transformed for analysis. However, to aid in interpretation, we present means and standard deviations of untransformed data. Rates of whole-animal metabolism, water flux, and components of energy budgets during seasons in which Gila monsters are active (Spring, Early Summer, and Late-Summer) were compared using repeated-measures ANOVA, using individual as the repeated factor. When seasons were significantly different overall, differences between pairs of seasonal means were tested using post-hoc Tukey-Kramer procedures.

We then used mixed effect GLM procedures (Bennington and Thayne 1994, Zuur et al. 2009) to analyze effects of mass, resting T_b , and amount of activity as covariates

influencing observed rates of energy and water use. For this, we used each covariate as a fixed effect and allowed individuals to vary randomly (random intercepts). We used the nlme package (Pinheiro and Bates 2000) adapted in the R statistical computing language. This mixed model approach has the advantage of accounting for both variation among individuals, and repeated measures within individuals. Additionally, because parameters in nlme are estimated using maximum likelihood, we should be able better to detect significant differences, even though the data set contains both small sample sizes ($N = 9$ lizards) and unbalanced distribution of samples (every individual not observed in every sampling period).

Statistics are presented as F , df , and P values in RM ANOVA, and as t and P for coefficient values in mixed models. There has been some debate on proper calculation of degrees of freedom in mixed-model maximum-likelihood estimates (Elston 1998), so we omit reporting them here to avoid confusion with models calculated from ordinary least squares analyses. Type I error rate (α) was set at 0.05 for all analyses.

We report mean values for each season, but we did not include Fall-Winter estimates in analyses because individuals are inactive below ground or hibernating. The resulting estimates of energy and water flux calculated over the four to five month inactive period inform us as to the relative costs of inactivity, but do not allow inference as to how environmental influences (temperature) or individual differences (T_b and activity) contribute to the use and budgeting of energy and water resources.

Results

We collected 62 total samples from nine lizards in 2002 and 2003. We obtained samples in each of the four seasons, and sample durations averaged 26 (± 7.4) days in the

spring, 19 (± 5.6) days in the early summer, 64 (± 27.1) days in late summer, and 150 (± 21.1) days for the fall-winter period.

Samples were excluded from analysis if they met any of a number of criteria that would make calculated estimates unreliable. Samples were excluded if, a) sample ^3H or ^{18}O concentration was less than 10% above background, b) if there was less than a difference of 10% between isotope levels of successive samples, or c) if rates of water flux were unusually high relative to rates of CO_2 production. Although the cutoff levels we chose are somewhat arbitrary, including samples that met these exclusion criteria could lead to unreliable and artificial estimates for energy and water flux (Nagy 1980, Nagy and Costa 1980, Congdon et al. 1982, Peterson et al. 1998).

Seasonal Differences in Energy and Water Use

Mean mass of Gila monsters was not statistically different between the spring, early Summer, and late Summer seasons (Table 1; RM ANOVA $F_{2,8} = 1.97$, $P = 0.20$), and we therefore did not further consider mass as a covariate in analyses among seasons, although we explicitly address its influence in analyses below.

Field metabolic rate (FMR) differed among activity seasons ($F_{2,8} = 4.49$, $P = 0.04$) and rates were highest in early summer (Figure 1 and Table 1). Post-hoc tests indicated that late summer rates were significantly different from both spring (Tukey-Kramer $q = 4.80$, $P = 0.02$) and early summer ($q = 6.31$, $P = 0.005$). Total Resting Metabolism (TRM) was different among seasons ($F_{2,8} = 5.08$, $P = 0.03$; Figure 1), but only spring and early summer were different on a pairwise basis ($q = 4.24$, $P = 0.04$). Energy used in activity (AR) differed seasonally (Figure 1; $F_{2,8} = 5.24$, $P = 0.03$), with early summer differing from late summer ($q = 4.14$, $P = 0.04$). Also, considering AR as a proportion of the

seasonal FMR (%AR), seasons differed ($F_{2,8} = 14.4$, $P = 0.002$), and again late summer was significantly lower than both spring and early summer ($q = 6.9$ and 6.1 , $P = 0.003$ and 0.006 respectively).

Total Body Water (TBW) did not differ significantly among seasons ($F_{2,8} = 0.22$, $P > 0.05$). Water Influx Rate (WIR) peaked in early summer and was significantly higher than both spring ($q = 6.9$, $P = 0.002$) and late summer ($q = 4.41$, $P = 0.03$), and was significantly different among all activity seasons ($F_{2,10} = 11.27$, $P = 0.003$; Table 2). After adjusting water flux to metabolic rate (WEI), water economy was different among seasons ($F_{2,8} = 4.41$, $P = 0.049$), highest in spring, and significantly different from late summer ($q = 4.06$, $P = 0.049$).

Influences on FMR and WIR

FMR and WIR did not differ between the sexes ($F_{1,7} = 0.47$, $P = 0.51$; $F_{1,7} = 0.06$, $P = 0.82$, respectively). FMR was significantly influenced by mass ($t = 2.08$, $P = 0.04$) and amount of activity ($t = 2.88$, $P = 0.008$) but not by resting T_b ($t = 0.44$, $P = 0.66$; Figure 3). WIR was significantly influenced by resting T_b ($t = 3.24$, $P = 0.002$) and activity ($t = 4.70$, $P < 0.0001$), but not by body mass ($t = 0.79$, $P = 0.43$; Figure 3).

Discussion

Gila monsters are extremely conservative in their use of both energy and water. Across the active season, they use energy at approximately half the rate expected in comparison to other (largely diurnal) desert lizards (Figure 1), even after accounting for body size. Spring and early summer rates (kJ d^{-1}) are 45 and 54% (respectively) of what might be expected for a desert lizard of their size, and during late summer energy use drops to 28% of predicted. Water intake (mL d^{-1}) follows a similar pattern; spring and

early summer with 33 and 65% of predicted rates, and a drop in late summer to 41% of predicted. Across the entire activity season, WIR is 46% of expected, and FMR is 42% of expected.

Of course, this remarkable difference in water and energy expenditures may be due largely to the combination of lower body temperatures normally preferred and achieved by Gila monsters, along with low levels of activity. Gila monsters minimize the influences of both T_b and activity during the active season by simply remaining inactive below-ground. Many lizards, especially those that are diurnally active, spend much of the time active because their diets require that they either actively forage for prey or that they maintain vigilant ambush behaviors to acquire prey (Huey and Pianka 1981, Pietruska 1986, Reilly et al. 2007). While active, diurnal lizards often maintain high and constant T_b in order to optimize aspects of prey acquisition such as sprinting and prey handling (Avery et al. 1982, Verwaijen and Van Damme 2007), but Gila monsters do not require such precise thermoregulation in order to forage for rodent pups, nestlings, and eggs.

The observed seasonal rates of energy and water consumption can be used to predict the amount of prey that Gila monsters would need to consume to meet maintenance energetic demands (Nagy 2001). Assuming that Gila monsters have a digestive efficiency of 76% (Beck 1986) and that neo-natal and juvenile rodents and lagomorphs have a caloric value of 7.5 kJ g^{-1} (Brisbin 1970, Cox and Secor 2007), then a typical individual with a annual energy demand of 3,766 kJ (Figure 1) would need to consume 660 g wet-mass of prey per year. This translates to roughly 5.9 liters of cottontail rabbits (*Sylvilagus audubonii* having mean litter size 3.4 and mean neonate size of 33

g; Swihart 1984) or 39 litters of kangaroo rats (*Dipodomys deserti* with mean litter mass ~17g; Jones 1985).

If we assume a value of digestive efficiency (91%, Wegscheider 1998) that is typical of reptilian carnivores (Smith 1976, Bedford and Christian 2000) then estimated intake needs would be slightly less. Using the predicted values of Nagy (2001) for feeding rates of wild reptiles, we again see that Gila monsters, on average, would need to consume less than half the amount of prey as would be predicted for reptiles in general, for all lizards, or for lizard carnivores (Table 1).

Partitioning of Energy Use

Although Gila monsters are inactive underground for the majority of day and year, nearly two-thirds of the annual energy budget is used in above-ground activity. This can be partially attributed to locomotor costs, as Gila monsters have activity energetics at approximately the predicted rate for their size (John-Alder et al. 1983, Beck et al. 1995), but this high proportion of energy allocated to activity more likely reflects low rates of resting metabolism (Beck and Lowe 1994). Because resting energetic rates are very low, even modest expenditures of time in activity causes activity to become a proportionally large component of the seasonal or annual energy budget. This is supported by examining %AR (Table 1), the percent of individual seasonal budgets used in activity. Spring and early summer rates are approximately three-fourths of the energy budget for those seasons (77 and 74% respectively), which is far above what might be predicted (on a time-specific basis) for an animal that spends nearly all its time inactive.

Many reptiles have shown pronounced seasonal differences in rates of energy and water expenditure (Nagy 1988, van Marken Lichtenbelt et al. 1993, Christian et al. 1995,

Henen 1997, de Souza et al. 2004). Often times seasonal differences are attributed to a voluntary reduction of energy and water that is achieved by individuals lowering the set-point of body temperature selection or by reducing the precision of thermoregulation (Christian et al. 1983, Christian and Bedford 1995, Christian et al. 1999, Ellis et al. 2008). Additionally, these physiological adjustments can be accompanied by behavioral adjustments that reduce levels of activity (Lister and Garcia-Aguayo 1992, Christian et al. 1995), thereby interactively yielding savings in energy and water.

We don't know whether Gila monsters lower their thermoregulatory set-point range (T_{set}) seasonally, but thermoregulation does become more difficult late in the activity season (Chapter 2). Environmental temperatures are highest in late summer, both above and below ground, and individuals are nearly always above their preferred range. This suggests that rates of energy and water turnover should be high due to the thermal dependence of these processes, and resting T_b does appear to influence WIR, but not FMR (Figure 3).

Seasonal activity is highest in mid and late summer (Chapter 2), and activity was significant in influencing both FMR and WIR. During August, activity is about half of what it was in June and July, and during September it is less than one-fifth of the seasonal peak. This reduction in activity can explain late-summer rates of energy use that are the lowest of the active season (Figure 1). Late in the active season, prey abundance is low and annual reproductive efforts are likely complete (Goldberg and Lowe 1997), and as a result lizards may choose to stay underground in estivation as a way to save energy (Storey and Storey 1990, Christian et al. 1999, Roe et al. 2008). But during the late-summer, the onset of monsoonal rains may influence Gila monsters to break

quiescence, and emerge above-ground to drink. Although the monsoons in southern Nevada are highly variable in intensity and duration (Hereford et al. 2006), available free-standing water may in part explain why observed rates of water intake during late summer are higher than during the spring when lizards are actively foraging.

Mass Effects

In nearly every taxon examined, body mass, (as an indicator of organismal size) has been shown to influence rates of physiological processes, including metabolism and water loss (Andrews and Pough 1985, Nagy and Peterson 1988, McNab 2002, Nagy 2005). In fact, the entire area of allometric analysis extends from the physical phenomenon of size, and its scaling relationship with functional and physiological processes (Calder 1984, Schmidt-Nielsen 1984). Our data follow the multitudes of studies reporting significant effects of body mass on physiological rates, and FMR of *Gila* monsters is significantly related to individual mass (Figure 3). However, WIR was not significantly related to body mass across the range of body sizes we observed. Our samples did not include data for juveniles or for individuals approaching the upper size limit of the species (mass range in this study = 272 to 379 g; maximum mass of free-range individuals ~880 g).

It is unlikely that body mass does not influence physiological rates of *Gila* monsters, but rather body mass itself may not always appropriately reflect body size. The actual mass of metabolizing tissue, which is likely the true parameter of interest, may vary differently from lizard mass due to lizard's ability to acquire and store large quantities of food and water. In the extreme, a lizard may consume a nest of rabbit or rodent pups that could be as much as one-third of its own body mass (Beck 1990). Time

to digest such a meal would be 7 to 12 days (Wegscheider 1998), and during the time food is in transit through the gut, lizard mass would be recorded as mass of the individual plus the mass of the undigested meal. As a consequence, the amount of actively metabolizing lizard tissue would be overestimated, and that error incorporated into analyses.

Additionally, Gila monsters may also store considerable volumes of dilute urine in the bladder (Davis and DeNardo 2007). While the urine may be important in hydroregulation, it could also overestimate actual lizard mass in the same way that prey consumption does and obscure significant influences of actual lizard mass on rates of energy use and water flux.

Geographic Variation

Here we have examined the seasonal differences in use and allocation of energy and water by Gila monsters, as well as the individual influences of mass, T_b , and above-ground activity. And although we have shown how these factors influence whole-animal physiology at a single site in southern Nevada, the magnitude of influences and individual responses likely vary among populations as well. Gila monster populations vary considerably in body size, T_b , and activity (Beck 2005), and one should, therefore, expect population differences in energy and water economy as well.

Thus, Gila monsters could be a species containing physiologically distinct, or locally adapted, populations (Garland and Adolph 1991), each varying as a function of environmental and individual influences. Preliminary analyses from a collaboration investigating geographic variation in resting metabolism of Gila monsters supports this (Gienger et al. 2009; Figure 4. See also Chapter 1, Table 1.), and results indicate that populations differ in both seasonal and annual resting energetic requirements. These

populations also differ in the amount of activity, body size, and potentially even reproductive output. The patterns of energy and water use observed here can partially explain the population differences, but a direct comparison of the use and energy and water resources among sites would be an interesting direction for future research.

Acknowledgements

We thank S. Cohen and S. Sheldon for field help with radio tracking and blood sampling of lizards. Financial support was provided by the Clark County (Nevada) MSHCP, Biological Resources Research Center (UNR), and UNR Department of Biology.

List of Abbreviations

AR	Activity Respiration
BM	Body Mass
DLW	Doubly Labeled Water
FMI	Fresh Matter Intake
FMR	Field Metabolic Rate
RMR	Resting Metabolic Rate
T_b	Body Temperature
TRM	Total Resting Metabolism
T_{set}	Thermoregulatory Set-Point Range
VO_2	Oxygen Consumption Rate
WEI	Water Economy Index

Literature Cited

- Adolph, S. C. 1990. Influence of behavioral thermoregulation on microhabitat use by two *Sceloporus* lizards. *Ecology* **71**:315-327.
- Adolph, S. C. and W. P. Porter. 1993. Temperature, activity, and lizard life histories. *The American Naturalist* **142**:273-295.
- Andrews, R. M. and F. H. Pough. 1985. Metabolism of squamate reptiles: allometric and ecological relationships. *Physiological Zoology* **58**:214-231.
- Angilletta, M. J., Jr., A. F. Bennett, H. Guderley, C. A. Navas, F. Seebacher, and R. S. Wilson. 2006. Coadaptation: A unifying principle in evolutionary thermal biology. *Physiological and Biochemical Zoology* **79**:282-294.
- Angilletta, M. J., Jr., P. H. Niewiarowski, and C. A. Navas. 2002. The evolution of thermal physiology in ectotherms. *Journal of Thermal Biology* **27**:249-268.
- Angilletta, M. J., Jr. and Y. L. Werner. 1998. Australian geckos do not display diel variation in thermoregulatory behavior. *Copeia*:736-742.
- Arnold, S. J., C. R. Peterson, and J. Gladstone. 1995. Behavioural variation in natural populations. VII. Maternal body temperature does not affect juvenile thermoregulation in a garter snake. *Animal Behaviour* **50**:623-633.
- Avery, R. A. 1982. Field studies of body temperatures and thermoregulation. Pages 93-166 *in* C. Gans and F. H. Pough, editors. *Biology of the Reptilia*. Volume 12 Physiology C. Academic Press, New York.
- Avery, R. A., J. D. Bedford, and C. P. Newcombe. 1982. The role of thermoregulation in lizard biology: Predatory efficiency in a temperate diurnal basker. *Behavioral Ecology and Sociobiology* **11**:261-267.
- Bakken, G. S. 1976. An improved method for determining thermal conductance and equilibrium body temperature with cooling curve experiments. *Journal of Thermal Biology* **1**:169-175.
- Bakken, G. S. 1992. Measurement and application of operative and standard operative temperatures in ecology. *American Zoologist* **32**:194-216.
- Ballinger, R. E., J. Hawker, and O. J. Sexton. 1969. The effect of photoperiod acclimation on the thermoregulation of the lizard, *Sceloporus undulatus*. *Journal of Experimental Zoology* **171**:43-47.

- Barber, B. J. and E. C. Crawford. 1977. A stochastic dual-limit hypothesis for behavioral thermoregulation in lizards. *Physiological Zoology* **50**:53-60.
- Bartholomew, G. A. 1982. Physiological Control of Body Temperature. Pages 167-211 in C. Gans and F. H. Pough, editors. *Biology of the Reptilia*, Volume 12. Academic Press, New York.
- Bartholomew, G. A. and R. C. Lasiewski. 1965. Heating and cooling rates, heart rate and simulated diving in the Galapagos marine iguana. *Comparative Biochemistry and Physiology* **16**:573-582.
- Bashey, F. and A. E. Dunham. 1997. Elevational variation in the thermal constraints on and microhabitat preferences of the greater earless lizard *Cophosaurus texanus*. *Copeia*:725-737.
- Bauwens, D., T. Garland, Jr., A. M. Castilla, and R. Van Damme. 1995. Evolution of sprint speed in lacertid lizards: Morphological, physiological and behavioral covariation. *Evolution* **49**:848-863.
- Bauwens, D., P. E. Hertz, and A. M. Castilla. 1996. Thermoregulation in a lacertid lizard: The relative contributions of distinct behavioral mechanisms. *Ecology* **77**:1818-1830.
- Beaupre, S. J., A. E. Dunham, and K. L. Overall. 1993. The effects of consumption rate and temperature on apparent digestibility coefficient, urate production, metabolizable energy coefficient and passage time in canyon lizards (*Sceloporus merriami*) from two populations. *Functional Ecology* **7**:273-280.
- Beck, D. D. 1986. The Gila Monster in Utah: Bioenergetic and Natural History Considerations. Unpub. M.S. Thesis. Utah State University, Logan, Utah.
- Beck, D. D. 1990. Ecology and behavior of the Gila monster in southwestern Utah. *Journal of Herpetology* **24**:54-68.
- Beck, D. D. 2005. *Biology of Gila Monsters and Beaded Lizards*. University of California Press, Berkeley.
- Beck, D. D., M. R. Dohm, T. Garland, Jr., A. Ramírez-Bautista, and C. H. Lowe. 1995. Locomotor performance and activity energetics of helodermatid lizards. *Copeia* **1995**:577-585.
- Beck, D. D. and R. D. Jennings. 2003. Habitat use by Gila monsters: the importance of shelters. *Herpetological Monographs* **17**:111-129.

- Beck, D. D. and C. H. Lowe. 1994. Resting metabolism of helodermatid lizards: Allometric and ecological relationships. *Journal of Comparative Physiology B* **164**:124-129.
- Bedford, G. S. and K. A. Christian. 2000. Digestive efficiency in some Australian pythons. *Copeia* **2000**:829-834.
- Benabib, M. and J. D. Congdon. 1992. Metabolic and water-flux rates of free-ranging tropical lizards *Sceloporus variabilis*. *Physiological Zoology* **65**:788-802.
- Bennett, A. F. 1978. Activity metabolism of the lower vertebrates. *Annual Review of Physiology* **40**:447-469.
- Bennett, A. F. 1987. Inter-individual Variability: An Underutilized Resource. Pages 147-169 in M. E. Feder, A. F. Bennett, W. W. Burggren, and R. B. Huey, editors. *New Directions in Ecological Physiology*. Cambridge University Press, Cambridge.
- Bennett, A. F. 1990. Thermal dependence of locomotor capacity. *American Journal of Physiology - Regulatory, Integrative and Comparative Physiology* **259**:R253-R258.
- Bennington, C. C. and W. V. Thayne. 1994. Use and misuse of mixed model analysis of variance in ecological studies. *Ecology* **75**:717-722.
- Berk, M. L. and J. E. Heath. 1975. Analysis of behavioral thermoregulation in the lizard, *Dipsosaurus dorsalis*. *Journal of Thermal Biology* **1**:15-22.
- Beuchat, C. A. 1986. Phylogenetic distribution of the urinary bladder in lizards. *Copeia*:512-517.
- Blouin-Demers, G. and P. Nadeau. 2005. The cost-benefit model of thermoregulation does not predict lizard thermoregulatory behavior. *Ecology* **86**:560-566.
- Blouin-Demers, G. and P. J. Weatherhead. 2001a. An experimental test of the link between foraging, habitat selection and thermoregulation in black rat snakes *Elaphe obsoleta obsoleta*. *Journal of Animal Ecology* **70**:1006-1013.
- Blouin-Demers, G. and P. J. Weatherhead. 2001b. Thermal ecology of black rat snakes (*Elaphe obsoleta*) in a thermally challenging environment. *Ecology* **82**:3025-3043.
- Bogert, C. M. 1949a. Thermoregulation and ecdyctic body temperatures in Mexican lizards of the genus *Sceloporus*. *Anales del Instituto de Biología de México* **20**:415-426.

- Bogert, C. M. 1949b. Thermoregulation in reptiles, a factor in evolution. *Evolution* **3**:195-211.
- Bogert, C. M. and R. Martín del Campo. 1956. The Gila Monster and Its Allies. *Bulletin of the American Museum of Natural History* **109**:1-238.
- Boyce, M. S. 1979. Seasonality and patterns of natural-selection for life histories. *American Naturalist* **114**:569-583.
- Brattstrom, B. H. 1965. Body temperatures of reptiles. *American Midland Naturalist* **73**:376-422.
- Brisbin, I. L., Jr. 1970. A Determination of live-weight caloric conversion factors for laboratory mice. *Ecology* **51**:541-544.
- Brown, D. E. 1994. *Biotic Communities: Southwestern United States and Northwestern Mexico*. University of Utah Press, Salt Lake City, UT.
- Brown, G. P. and P. J. Weatherhead. 2000. Thermal ecology and sexual size dimorphism in northern water snakes, *Nerodia sipedon*. *Ecological Monographs* **70**:311-330.
- Calder, W. A. 1984. *Size, Function, and Life History*. Dover Publications Inc., Mineola, New York.
- Campbell, G. S. and J. M. Norman. 1998. *An Introduction to Environmental Biophysics*. Springer-Verlag, New York.
- Campbell, J. A. and J. P. Vannini. 1988. A new subspecies of beaded lizard, *Heloderma horridum*, from the Motagua Valley of Guatemala. *Journal of Herpetology* **22**:457-468.
- Christel, C. M., D. F. DeNardo, and S. M. Secor. 2007. Metabolic and digestive response to food ingestion in a binge-feeding lizard, the Gila monster (*Heloderma suspectum*). *Journal of Experimental Biology* **210**:3430-3439.
- Christian, K. and B. Weavers. 1994. Analysis of the activity and energetics of the lizard *Varanus rosenbergi*. *Copeia* **1994**:289-295.
- Christian, K. A., R. V. Baudinette, and Y. Pamula. 1997. Energetic costs of activity by lizards in the field. *Functional Ecology* **11**:392-397.
- Christian, K. A. and G. S. Bedford. 1995. Seasonal changes in thermoregulation by the frillneck lizard, *Chlamydosaurus kingii*, in tropical Australia. *Ecology* **76**:124-132.

- Christian, K. A., G. S. Bedford, and T. J. Schultz. 1999. Energetic consequences of metabolic depression in tropical and temperate-zone lizards. *Australian Journal of Zoology* **47**:133-141.
- Christian, K. A., L. K. Corbett, B. Green, and B. W. Weavers. 1995. Seasonal activity and energetics of two species of varanid lizards in tropical Australia. *Oecologia* **103**:349-357.
- Christian, K. A. and C. R. Tracy. 1984. Physiological and ecological consequences of sleeping-site selection by the Galapagos land iguana (*Conolophus pallidus*). *Ecology* **65**:752-758.
- Christian, K. A. and C. R. Tracy. 1985. Physical and biotic determinants of space utilization by the Galapagos land iguana (*Conolophus pallidus*). *Oecologia* **66**:132-140.
- Christian, K. A., C. R. Tracy, and W. P. Porter. 1983. Seasonal shifts in body temperature and use of microhabitats by Galapagos land iguanas (*Conolophus pallidus*). *Ecology* **64**:463-468.
- Christian, K. A., C. R. Tracy, and W. P. Porter. 1985. Inter- and intra-individual variation in body temperatures of the Galapagos land iguana (*Conolophus pallidus*). *Journal of Thermal Biology* **10**:47-50.
- Christian, K. A., C. R. Tracy, and C. R. Tracy. 2006. Evaluating thermoregulation in reptiles: An appropriate null model. *American Naturalist* **168**:421-430.
- Christian, K. A. and B. W. Weavers. 1996. Thermoregulation of monitor lizards in Australia: An evaluation of methods in thermal biology. *Ecological Monographs* **66**:139-157.
- Clusella-Trullas, S., J. S. Terblanche, J. H. van Wyk, and J. R. Spotila. 2007. Low repeatability of preferred body temperature in four species of Cordylid lizards: Temporal variation and implications for adaptive significance. *Evolutionary Ecology* **21**:63-79.
- Congdon, J. D., A. E. Dunham, and D. W. Tinkle. 1982. Energy Budgets and Life Histories of Reptiles. Pages 233-271 in C. Gans and F. H. Pough, editors. *Biology of the Reptilia*, Vol. 13. Academic Press, New York.
- Cowles, R. B. 1939. Possible implications of reptilian thermal tolerance. *Science* **90**:465-466.
- Cowles, R. B. 1940. Additional implications of reptilian sensitivity to high temperatures. *American Naturalist* **74**:542-561.

- Cowles, R. B. and C. M. Bogert. 1944. A preliminary study of the thermal requirements of desert reptiles. *Bulletin of the American Museum of Natural History* **83**:265-296.
- Cox, C. L. and S. M. Secor. 2007. Effects of meal size, clutch, and metabolism on the energy efficiencies of juvenile Burmese pythons, *Python molurus*. *Comparative Biochemistry and Physiology - Part A: Molecular & Integrative Physiology* **148**:861-868.
- Davis, J. R. and D. F. DeNardo. 2007. The urinary bladder as a physiological reservoir that moderates dehydration in a large desert lizard, the Gila monster *Heloderma suspectum*. *Journal of Experimental Biology* **210**:1472-1480.
- Davis, J. R., E. N. Taylor, and D. F. DeNardo. 2008. An automated temperature-based option for estimating surface activity and refuge use patterns in free-ranging animals. *Journal of Arid Environments* **72**:1414-1422.
- Dawson, W. R. 1975. On the Physiological Significance of the Preferred Body Temperatures of Reptiles. Pages 443-473 in D. M. Gates and R. B. Schmerl, editors. *Perspectives of Biophysical Ecology*, New York.
- DeNardo, D. F., T. E. Zubal, and T. C. M. Hoffman. 2004. Cloacal evaporative cooling: a previously undescribed means of increasing evaporative water loss at higher temperatures in a desert ectotherm, the Gila monster *Heloderma suspectum*. *Journal of Experimental Biology* **207**:945-953.
- Dewitt, C. B. and R. M. Friedman. 1979. Significance of skewness in ectotherm thermoregulation. *American Zoologist* **19**:195-209.
- Dohm, M. R., W. J. Mautz, P. G. Looby, K. S. Gellert, and J. A. Andrade. 2001. Effects of ozone on evaporative water loss and thermoregulatory behavior of marine toads (*Bufo marinus*). *Environmental Research* **86**:274-286.
- Dzialowski, E. M. 2005. Use of operative temperature and standard operative temperature models in thermal biology. *Journal of Thermal Biology* **30**:317-334.
- Dzialowski, E. M. and M. P. O'Connor. 2001. Thermal time constant estimation in warming and cooling ectotherms. *Journal of Thermal Biology* **26**:231-245.
- Ellis, D. J., B. T. Firth, and I. Belan. 2006. Circadian rhythm of behavioral thermoregulation in the sleepy lizard (*Tiliqua rugosa*). *Herpetologica* **62**:259-265.
- Ellis, D. J., B. T. Firth, and I. Belan. 2008. Interseasonal variation in the circadian rhythms of locomotor activity and temperature selection in sleepy lizards, *Tiliqua*

- rugosa*. Journal of Comparative Physiology a-Neuroethology Sensory Neural and Behavioral Physiology **194**:701-712.
- Elston, D. A. 1998. Estimation of denominator degrees of freedom of F-distributions for assessing Wald statistics for fixed-effect factors in unbalanced mixed models. Biometrics **54**:1085-1096.
- Emlen, J. M. 1966. The role of time and energy in food preference. American Naturalist **100**:611-617.
- Endler, J. A. 1986. Natural Selection in the Wild. Princeton University Press, Princeton, N.J.
- Esler, K. J. and P. W. Rundel. 1999. Comparative patterns of phenology and growth form diversity in two winter rainfall deserts: the Succulent Karoo and Mojave Desert ecosystems. Plant Ecology **142**:97-104.
- Firth, B. T. and I. Belan. 1998. Daily and seasonal rhythms in selected body temperatures in the Australian lizard *Tiliqua rugosa* (Scincidae): Field and laboratory observations. Physiological Zoology **71**:303-311.
- Firth, B. T., J. S. Turner, and C. L. Ralph. 1989. Thermoregulatory behaviour in two species of iguanid lizards (*Crotaphytus collaris* and *Sauromalus obesus*): diel variation and the effect of pinealectomy. Journal of Comparative Physiology B: Biochemical, Systemic, and Environmental Physiology **159**:13-20.
- Garland, T., Jr. and S. C. Adolph. 1991. Physiological differentiation of vertebrate populations. Annual Review of Ecology and Systematics **22**:193-228.
- Gienger, C. M. 2003. Natural history of the Gila monster in Nevada. Unpub. M.S. Thesis. University of Nevada, Reno, Reno.
- Gienger, C. M., J. R. Davis, D. F. Denardo, D. D. Beck, and C. R. Tracy. 2009. Geographic variation in the energetics of inactivity: Individual and environmental influences on the resting metabolism of Gila monsters. Dynamic Deserts: Resource Uncertainty in Arid Environments, Tempe, AZ. February 25-28, 2009.
- Glasser, J. W. 1984. Evolution of efficiencies and strategies of resource exploitation. Ecology **65**:1570-1578.
- Goldberg, S. R. and C. H. Lowe. 1997. Reproductive cycle of the Gila monster, *Heloderma suspectum*, in southern Arizona. Journal of Herpetology **31**:161-166.

- Graham, M. H. and M. S. Edwards. 2001. Statistical significance versus fit: estimating the importance of individual factors in ecological analysis of variance. *Oikos* **93**:505-513.
- Grant, B. W. and A. E. Dunham. 1988. Thermally imposed time constraints on the activity of the desert lizard *Sceloporus merriami*. *Ecology* **69**:167-176.
- Harlow, H. J., S. S. Hillman, and M. Hoffman. 1976. Effect of temperature on digestive efficiency in the herbivorous lizard, *Dipsosaurus dorsalis*. *Journal of Comparative Physiology* **111**:1-6.
- Hayes, J. P., C. A. Bible, and J. D. Boone. 1998. Repeatability of mammalian physiology: Evaporative water loss and oxygen consumption of *Dipodomys merriami*. *Journal of Mammalogy* **79**:475-485.
- Hayes, J. P. and S. H. Jenkins. 1997. Individual variation in mammals. *Journal of Mammalogy* **78**:274-293.
- Henen, B. T. 1997. Seasonal and annual energy budgets of female desert tortoises (*Gopherus agassizii*). *Ecology* **78**:283-296.
- Hereford, R., R. H. Webb, and C. I. Longpre. 2006. Precipitation history and ecosystem response to multidecadal precipitation variability in the Mojave Desert region, 1893-2001. *Journal of Arid Environments* **67**:13-34.
- Hertz, P. E., R. B. Huey, and T. Garland, Jr. 1988. Time Budget, thermoregulation, and maximal locomotor performance: Are reptiles olympians or boy scouts? *American Zoologist* **1988**:927-938.
- Hertz, P. E., R. B. Huey, and R. D. Stevenson. 1993. Evaluating temperature regulation by field-active ectotherms: The fallacy of the inappropriate question. *American Naturalist* **142**:796-818.
- Huey, R. B. 1982. Temperature, Physiology, and the Ecology of Reptiles. Pages 25-91 in C. Gans and F. H. Pough, editors. *Biology of the Reptilia* 12C. Academic Press, New York.
- Huey, R. B. and A. F. Bennett. 1987. Phylogenetic studies of coadaptation: Preferred temperatures versus optimal performance temperatures of lizards. *Evolution* **41**:1098-1115.
- Huey, R. B. and P. E. Hertz. 1984. Is a jack-of-all-temperatures a master of none? *Evolution* **38**:441-444.

- Huey, R. B. and E. R. Pianka. 1981. Ecological consequences of foraging mode. *Ecology* **62**:991-999.
- Huey, R. B. and M. Slatkin. 1976. Costs and benefits of lizard thermoregulation. *Quarterly Review of Biology* **51**:363-384.
- Huey, R. B. and R. D. Stevenson. 1979. Integrating thermal physiology and ecology of ectotherms: A discussion of approaches. *American Zoologist* **19**:357-366.
- Husak, J. F., S. F. Fox, M. B. Lovern, and R. A. Van Den Bussche. 2006. Faster lizards sire more offspring: Sexual selection on whole-animal performance. *Evolution* **60**:2122-2130.
- Hyde, L. L. and H. Underwood. 2000. Effects of melatonin administration on the circadian activity rhythm of the lizard *Anolis carolinensis*. *Physiology & Behavior* **71**:183-192.
- John-Alder, H. B., C. H. Lowe, and A. F. Bennett. 1983. Thermal dependence of locomotory energetics and aerobic capacity of the Gila monster (*Heloderma suspectum*). *Journal of Comparative Physiology B* **151**:119-126.
- Karasov, W. H. and C. Martínez del Río. 2007. *Physiological Ecology: How Animals Process Energy, Nutrients, and Toxins*. Princeton University Press, Princeton, NJ.
- Keren, G. and C. Lewis. 1979. Partial omega squared for ANOVA designs. *Educational and Psychological Measurement* **39**:119-128.
- Kingsbury, B. A. 1994. Thermal constraints and eurythermy in the lizard *Elgaria multicarinata*. *Herpetologica* **50**:266-273.
- Lappin, A. K. and J. F. Husak. 2005. Weapon performance, not size, determines mating success and potential reproductive output in the collared lizard (*Crotaphytus collaris*). *American Naturalist* **166**:426-436.
- Le Galliard, J. F., M. Le Bris, and J. Clobert. 2003. Timing of locomotor impairment and shift in thermal preferences during gravidity in a viviparous lizard. *Functional Ecology* **17**:877-885.
- Lessells, C. M. and P. T. Boag. 1987. Unrepeatable repeatabilities: a common mistake. *the Auk* **104**:116-121.
- Licht, P. E., W. R. Dawson, V. H. Shoemaker, and A. R. Main. 1966. Observations on the thermal relations of western Australian lizards. *Copeia* **1966**:97-110.

- Lifson, N. and R. McClintock. 1966. Theory of use of the turnover rates of body water for measuring energy and material balance. *Journal of Theoretical Biology* **12**:46-74.
- Lister, B. C. and A. Garcia-Aguayo. 1992. Seasonality, predation, and the behaviour of a tropical mainland anole. *Journal of Animal Ecology* **61**:717-733.
- Lowe, C. H., C. R. Schwalbe, and T. B. Johnson. 1986. *The Venomous Reptiles of Arizona*. Arizona Game and Fish Department, Phoenix.
- Lutterschmidt, D. I., W. I. Lutterschmidt, and V. H. Hutchison. 2003. Melatonin and thermoregulation in ectothermic vertebrates: a review. *Canadian Journal of Zoology* **81**:1-13.
- McGinnis, S. M. 1966. *Sceloporus occidentalis*: Preferred body temperature of the western fence lizard. *Science* **152**:1090-1091.
- McGraw, K. O. and S. P. Wong. 1996. Forming inferences about some intraclass correlation coefficients. *Psychological Methods* **1**:30-46.
- McNab, B. K. 2002. *The Physiological Ecology of Vertebrates: A View From Energetics*. Cornell University Press, Ithaca.
- Nagy, K. A. 1980. CO₂ production in animals: analysis of potential errors in the doubly labeled water method. *American Journal of Physiology* **238**:R466-R473.
- Nagy, K. A. 1982a. Energy Requirements of Free-living Iguanid Lizards. Pages 47-59 *in* G. M. Burghardt and A. S. Rand, editors. *Iguanas of the World: Their Behavior, Ecology, and Conservation*. Noyes, Park Ridge, NJ.
- Nagy, K. A. 1982b. Field Studies of Water Relations. Pages 483-501 *in* C. Gans and F. H. Pough, editors. *Biology of the Reptilia*. Volume 12. Academic Press, London.
- Nagy, K. A. 1983. *The Doubly Labeled Water (3HH¹⁸O) Method: A Guide to its Use*. Publication 12-1417, University of California, Los Angeles.
- Nagy, K. A. 1988. Seasonal patterns of water and energy-balance in desert vertebrates. *Journal of Arid Environments* **14**:201-210.
- Nagy, K. A. 2001. Food requirements of wild animals: Predictive equations for free-living mammals, reptiles, and birds. *Nutrition Abstracts and Reviews, Series B* **71**:2R-12R.
- Nagy, K. A. 2004. Water economy of free-living desert animals. *International Congress Series* **1275**:291-297.

- Nagy, K. A. 2005. Field metabolic rate and body size. *Journal of Experimental Biology* **208**:1621-1625.
- Nagy, K. A. and D. P. Costa. 1980. Water flux in animals: analysis of potential errors in the tritiated water method. *American Journal of Physiology* **238**:R454-R465.
- Nagy, K. A., I. A. Girard, and T. K. Brown. 1999. Energetics of free-ranging mammals, reptiles, and birds. *Annual Review of Nutrition* **19**:247-277.
- Nagy, K. A. and C. C. Peterson. 1988. Scaling of water flux rate in animals. *University of California Publications in Zoology* **120**:1-172.
- Nespolo, R. F. and M. Franco. 2007. Whole-animal metabolic rate is a repeatable trait: a meta-analysis. *Journal of Experimental Biology* **210**:2000-2005.
- Niewiarowski, P. H. 2001. Energy budgets, growth rates, and thermal constraints: Toward an integrative approach to the study of life history variation. *The American Naturalist* **157**:421-433.
- O'Connor, M. P. 1999. Physiological and ecological implications of a simple model of heating and cooling in reptiles. *Journal of Thermal Biology* **24**:113-136.
- Peterson, C. C., B. M. Walton, and A. F. Bennett. 1998. Intrapopulation variation in ecological energetics of the garter snake *Thamnophis sirtalis*, with analysis of the precision of doubly labeled water measurements. *Physiological Zoology* **71**:333-349.
- Peterson, C. R. 1987. Daily variation in the body temperatures of free-ranging garter snakes. *Ecology* **68**:160-169.
- Pietruska, R. D. 1986. Search tactics of desert lizards: how polarized are they? *Animal Behavior* **34**:1742-1758.
- Pinheiro, J. C. and D. M. Bates. 2000. *Mixed-Effects Models in S and S-Plus*. Springer-Verlag, New York.
- Porter, W. P., J. W. Mitchell, W. A. Beckman, and C. B. DeWitt. 1973. Behavioral implications of mechanistic ecology: Thermal and behavioral modeling of desert ectotherms and their microenvironment. *Oecologia* **13**:1-54.
- Porter, W. P. and C. R. Tracy. 1983. Biophysical Analyses of Energetics, Time-space Utilization, and Distributional Limits. Pages 55-83 *in* R. B. Huey, E. R. Pianka, and T. W. Schoener, editors. *Lizard Ecology: Studies of a Model Organism*. Harvard University Press, Cambridge, Massachusetts, USA.

- Porzer, L. M. 1981. Movement, behavior, and body temperature of the Gila monster (*Heloderma suspectum*) in Queen Creek, Pinal County, Arizona. Unpub. M.S. Thesis. Arizona State University, Tempe.
- Ralph, C. L., B. T. Firth, and J. S. Turner. 1979. The role of the pineal body in ectotherm thermoregulation. *American Zoologist* **19**:273-293.
- Refinetti, R. 1996. Demonstrating the consequences of violations of assumptions in between-subjects analysis of variance. *Teaching of Psychology* **23**:51-54.
- Regal, P. J. 1967. Voluntary hypothermy in reptiles. *Science* **155**:1551-1553.
- Reilly, S. M., L. D. McBrayer, and D. B. Miles. 2007. *Lizard Ecology: The Evolutionary Consequences of Foraging Mode*. Cambridge University Press, Cambridge, UK.
- Roe, J. H., A. Georges, and B. Green. 2008. Energy and water flux during terrestrial estivation and overland movement in a freshwater turtle. *Physiological and Biochemical Zoology* **81**:570-583.
- Rundel, P. W. and A. C. Gibson. 1996. *Ecological Communities and Processes in a Mojave Desert Ecosystem: Rock Valley, Nevada*. Cambridge University Press.
- Schmidt-Nielsen, K. 1984. *Scaling: Why is Animal Size So Important?* Cambridge University Press, Cambridge.
- Scott, J. R., C. R. Tracy, and D. Pettus. 1982. A biophysical analysis of daily and seasonal utilization of climate space by a montane snake. *Ecology* **63**:482-493.
- Sears, M. W. 2005. Geographic variation in the life history of the sagebrush lizard: the role of thermal constraints on activity. *Oecologia* **143**:25-36.
- Sears, M. W. and M. J. Angilletta, Jr. 2003. Life-history variation in the sagebrush lizard: Phenotypic plasticity or local adaptation? *Ecology* **84**:1624-1634.
- Secor, S. M. 2009. Specific dynamic action: a review of the postprandial metabolic response. *Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology* **179**:1-56.
- Secor, S. M. and J. M. Diamond. 2000. Evolution of regulatory responses to feeding in snakes. *Physiological and Biochemical Zoology* **73**:123-141.
- Secor, S. M., E. D. Stein, and J. Diamond. 1994. Rapid upregulation of snake intestine in response to feeding: a new model of intestinal adaptation. *American Journal of Physiology - Gastrointestinal and Liver Physiology* **266** G695-G705.

- Seebacher, F. and C. E. Franklin. 2004. Cardiovascular mechanisms during thermoregulation in reptiles. *International Congress Series* **1275**:242-249.
- Seebacher, F. and C. E. Franklin. 2005. Physiological mechanisms of thermoregulation in reptiles: a review. *Journal of Comparative Physiology B: Biochemical, Systemic, and Environmental Physiology* **175**:533-541.
- Shine, R. 2005. Life-history evolution in reptiles. *Annual Review of Ecology and Systematics* **36**:23-46.
- Shine, R., L. X. Sun, M. Kearney, and M. Fitzgerald. 2002. Thermal correlates of foraging-site selection by Chinese pit-vipers (*Gloydius shedaoensis*, Viperidae). *Journal of Thermal Biology* **27**:405-412.
- Sievert, L. M. and V. H. Hutchison. 1991. The influence of photoperiod and position of a light source on behavioral thermoregulation in *Crotaphytus collaris* (Squamata: Iguanidae). *Copeia* **1991**:105-110.
- Smith, G. C. 1976. Ecological energetics of three species of ectothermic vertebrates. *Ecology* **57**:252-264.
- de Souza, S. C. R., J. E. de Carvalho, A. S. Abe, J. E. P. W. Bicudo, and M. S. C. Bianconcini. 2004. Seasonal metabolic depression, substrate utilisation and changes in scaling patterns during the first year cycle of tegu lizards (*Tupinambis merianae*). *Journal of Experimental Biology* **207**:307-318.
- Speakman, J. R. 1997. *Doubly Labeled Water Theory and Practice*. Chapman and Hall, London.
- Stahnke, H. L. 1950. The food of the Gila monster. *Herpetologica* **6**:103-106.
- Stahnke, H. L. 1952. A note on the food habits of the Gila monster, *Heloderma suspectum* Cope. *Herpetologica* **8**:64.
- StatSoft. 2001. Statistica (data analysis software system), version 6. www.statsoft.com.
- Storey, K. B. and J. M. Storey. 1990. Metabolic rate depression and biochemical adaptation in anaerobiosis, hibernation and estivation. *The Quarterly Review of Biology* **65**:145.
- Swihart, R. K. 1984. Body size, breeding season length, and life history tactics of lagomorphs. *Oikos* **43**:282-290.

- Tracy, C. R. 1982. Biophysical Modeling of Reptilian Physiology and Ecology. Pages 275-321 in C. Gans and F. H. Pough, editors. *Biology of the Reptilia* Vol. 12. Academic New York.
- Tracy, C. R. and K. A. Christian. 1986. Ecological relations among space, time, and thermal niche axes. *Ecology* **67**:609-615.
- Tracy, C. R., K. M. Flack, L. C. Zimmerman, R. E. Espinoza, and C. R. Tracy. 2005. Herbivory imposes constraints on voluntary hypothermia in lizards. *Copeia*:12-19.
- Tracy, C. R., C. R. Tracy, and J. S. Turner. 1992. Contrasting physiological abilities for heating and cooling in an amphibian (*Rana pipiens*) and a reptile (*Sauromalus obesus*). *Herpetologica* **48**:57-60.
- Turner, J. S. 1987. On the transient temperatures of ectotherms. *Journal of Thermal Biology* **12**:207-214.
- van Marken Lichtenbelt, W. D., R. A. Wesselingh, J. T. Vogel, and K. B. M. Albers. 1993. Energy budgets in free-living green iguanas in a seasonal environment. *Ecology* **74**:1157-1172.
- Vance, V. J. 1973. Temperature preference and tolerance in the gecko, *Coleonyx variegatus*. *Copeia* **1973**:615-617.
- Verwajen, D. and R. Van Damme. 2007. Correlated evolution of thermal characteristics and foraging strategy in lacertid lizards. *Journal of Thermal Biology* **32**:388-395.
- Vincent, S. E. and A. Mori. 2008. Determinants of feeding performance in free-ranging pit-vipers (Viperidae: *Ovophis okinavensis*): key roles for head size and body temperature. *Biological Journal of the Linnean Society* **93**:53-62.
- Vitt, L. J., J. P. Caldwell, S. S. Sartorius, W. E. Cooper, T. A. Baird, T. D. Baird, and V. Perez-Mellado. 2005. Pushing the edge: extended activity as an alternative to risky body temperatures in a herbivorous teiid lizard (*Cnemidophorus murinus* : Squamata). *Functional Ecology* **19**:152-158.
- Waldschmidt, S. R., S. M. Jones, and W. P. Porter. 1985. The effect of body temperature and feeding regime on activity, passage time, and digestive coefficient in the lizard *Uta stansburiana*. *Physiological Zoology* **59**:376-383.
- Walsberg, G. E. and B. O. Wolf. 1996. An appraisal of operative temperature mounts as tools for studies of ecological energetics. *Physiological Zoology* **69**:658-681.
- Waterman, T. H. 1999. Evolutionary challenges of extreme environments (Part 1). *Journal of Experimental Zoology* **285**:326-359.

- Waterman, T. H. 2001. Evolutionary challenges of extreme environments (Part 2). *Journal of Experimental Zoology* **291**:130-168.
- Webb, J. K. and R. Shine. 1998. Using thermal ecology to predict retreat-site selection by an endangered snake species. *Biological Conservation* **86**:233-242.
- Wegscheider, F. J. 1998. Assimilation efficiency of the Gila monster *Heloderma suspectum*. M.S. Thesis. California State University, Fullerton, Fullerton, CA.
- Wood, R. A., K. A. Nagy, N. S. MacDonald, S. T. Wakakuwa, R. J. Beckman, and H. Kaaz. 1975. Determination of oxygen-18 in water contained in biological samples by charged particle activation. *Analytical Chemistry* **47**:646-650.
- Woodson, W. W. 1949. Mammal diet of *Heloderma*. *Herpetologica* **5**:152.
- Youssef, M. K., S. C. Adolph, and J. Q. Richmond. 2008. Evolutionarily conserved thermal biology across continents: The North American lizard *Plestiodon gilberti* (Scincidae) compared to Asian *Plestiodon*. *Journal of Thermal Biology* **33**:308-312.
- Zar, J. H. 1999. *Biostatistical Analysis*. Prentice Hall, Upper Saddle River, N.J.
- Zimmerman, L. C. and C. R. Tracy. 1989. Interactions between the environment and ectothermy and herbivory in reptiles. *Physiological Zoology* **62**:374-409.
- Zuur, A. F., E. N. Ieno, N. J. Walker, A. A. Saveliev, and G. M. Smith. 2009. *Mixed Effects Models and Extensions in Ecology with R*. Springer, New York.

Table 1. Body mass, CO₂ production, and field metabolic rates (FMR) of *Heloderma suspectum* by season.

	Spring (n = 8)	Early Summer (n =9)	Late Summer (n = 6)	Fall-Winter (n=3)
Mass (g)	349 (25)	339 (36)	332 (37)	328 (40)
CO ₂ (mL g ⁻¹ h ⁻¹)	0.087 (0.020)	0.105 (0.021)	0.057 (0.042)	0.019 (0.018)
FMR (kJ g ⁻¹ d ⁻¹)	0.054 (0.012)	0.065 (0.013)	0.035 (0.026)	0.011 (0.011)
FMR (kJ d ⁻¹)	18.96 (4.81)	22.17 (4.59)	11.34 (7.69)	3.36 (3.08)
Predicted FMR:				
Reptile (kJ d ⁻¹)	35.7	34.8	34.2	
Lizard (kJ d ⁻¹)	40.5	39.5	38.7	
Desert Lizard (kJ d ⁻¹)	42.2	41.1	40.3	
Feeding Rate (g FMI d ⁻¹)	3.3	3.9	1.9	
Predicted Feeding Rate:				
Reptile (g FMI d ⁻¹)	7.8	7.6	7.4	
Lizard (g FMI d ⁻¹)	8.7	8.5	8.3	
Carnivorous Lizard (g FMI d ⁻¹)	8.2	7.9	7.8	
TRM (kJ d ⁻¹)	3.9 (0.6)	5.1 (0.5)	4.7 (0.7)	1.9*
AR (kJ d ⁻¹)	15.1 (5.0)	17.1 (4.6)	6.6 (7.5)	1.4*
%AR	77.4 (8.6)	73.6 (7.1)	33.1 (23.1)	
SusMS	5.1 (1.9)	4.4 (1.0)	2.3 (1.4)	

Data are mean of individual means ($\pm 1SD$). Predicted values for field metabolic rate (FMR) are calculated from Nagy et al. (1999) for active season metabolism and Nagy (2001) for feeding rates. Total resting metabolism (TRM) is modeled using body mass and mean Tb to predict resting metabolic rate over each sample period. *Fall-winter TRM was calculated by averaging calculations for two week intervals from onset of hibernation through spring emergence.

Table 2. Measured and predicted water flux rates, total body water, and water economy index for *Heloderma suspectum* by season.

	Spring (n = 8)	Early Summer (n = 9)	Late Summer (n = 6)	Fall-Winter (n=6)
Mass (g)	349 (25)	339 (36)	332 (37)	328 (40)
Water influx (mL d ⁻¹)	2.6 (1.5)	5.0 (1.1)	3.1 (1.7)	0.60 (0.1)
Water influx (mL kg ⁻¹ d ⁻¹)	7.4 (4.4)	14.7 (3.3)	9.3 (5.2)	1.8 (0.5)
Predicted:				
Arid/Semi-Arid Lizard (mL d ⁻¹)	7.9	7.7	7.5	
Tropical Lizard (mL d ⁻¹)	22.5	22.0	21.7	
TBW (%)	66.3 (3.5)	65.7 (6.4)	64.2 (9.3)	67.3 (3.3)
WEI (mL kJ ⁻¹)	0.15 (0.10)	0.28 (0.10)	0.36 (0.15)	0.23 (0.11)

Data are mean of individual means ($\pm 1SD$). Predicted values for water flux are calculated from Nagy (1982) for active seasons.

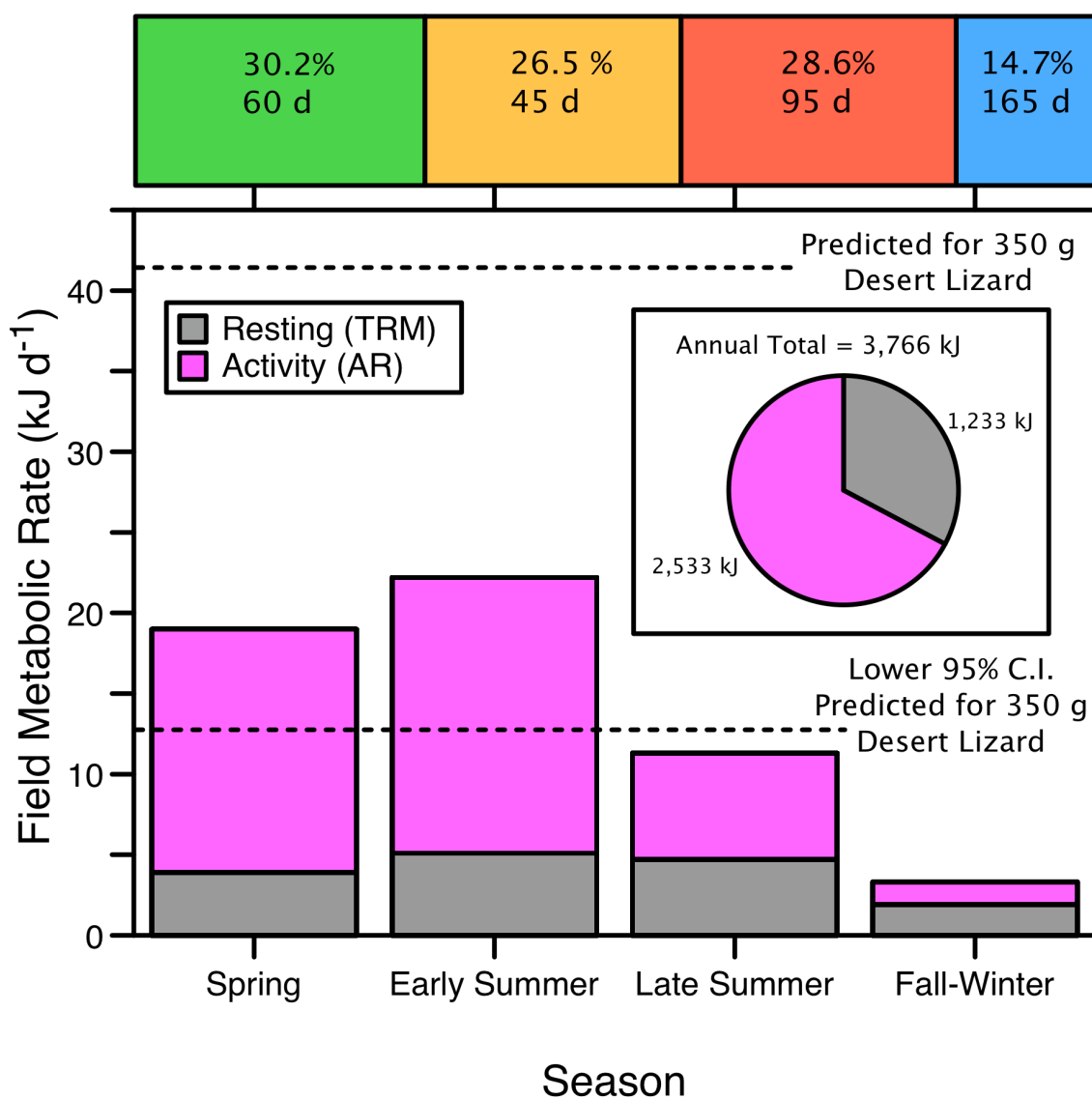


Figure 1. Seasonal field metabolic rates (FMR) of Gila monsters, and partitioning of the annual energy budget to activity (66% of annual) and rest (33% of annual). The average energy budget totals 3,766 kJ, and rates during the activity season are less than half what is predicted for a 350 g desert lizard (upper dashed line).

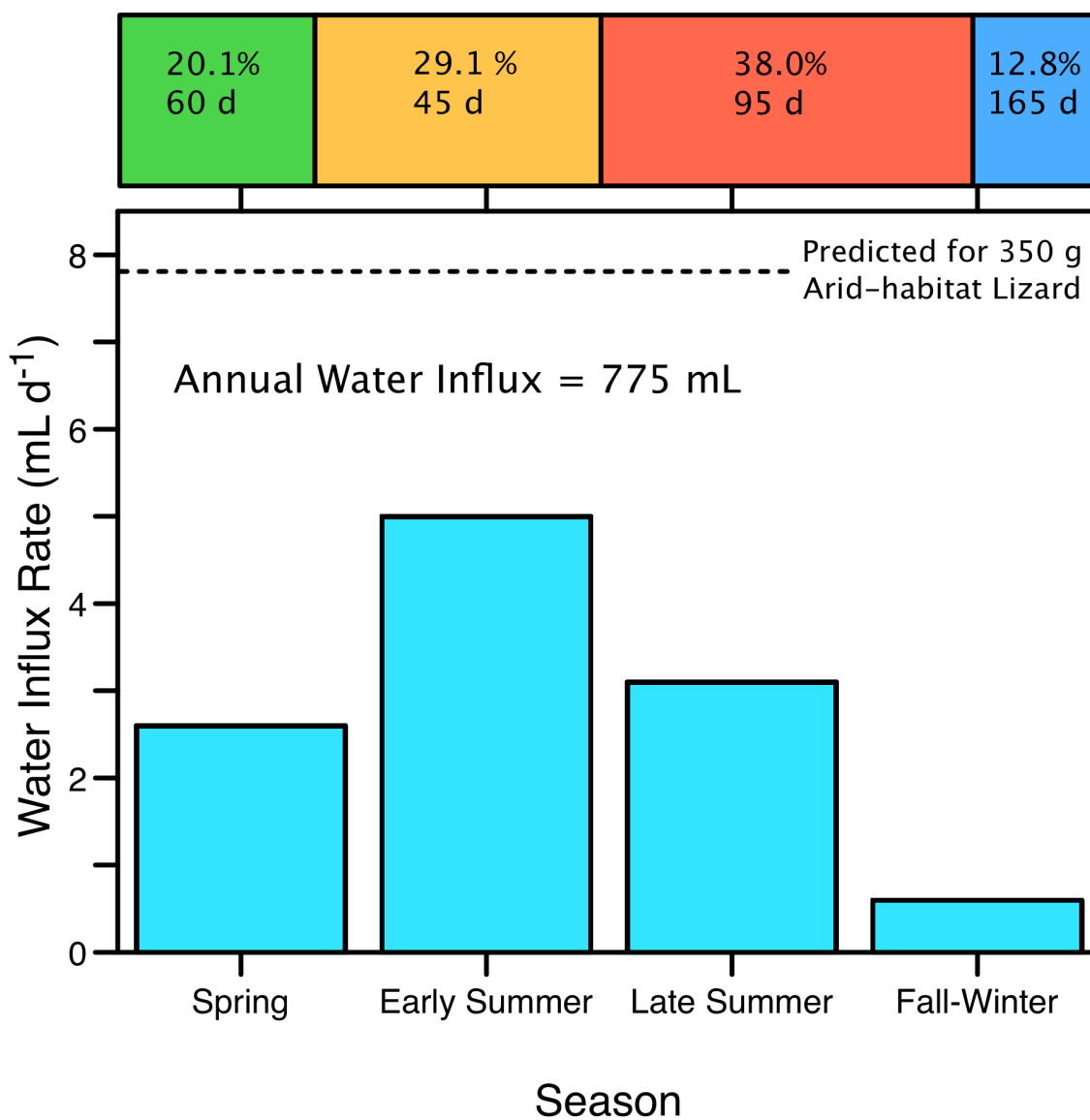


Figure 2. Seasonal rates of water influx in Gila monsters; individuals in southern Nevada have rates that are less than half of predicted for a 350 g desert lizard. Annual total intake averages 775 mL.

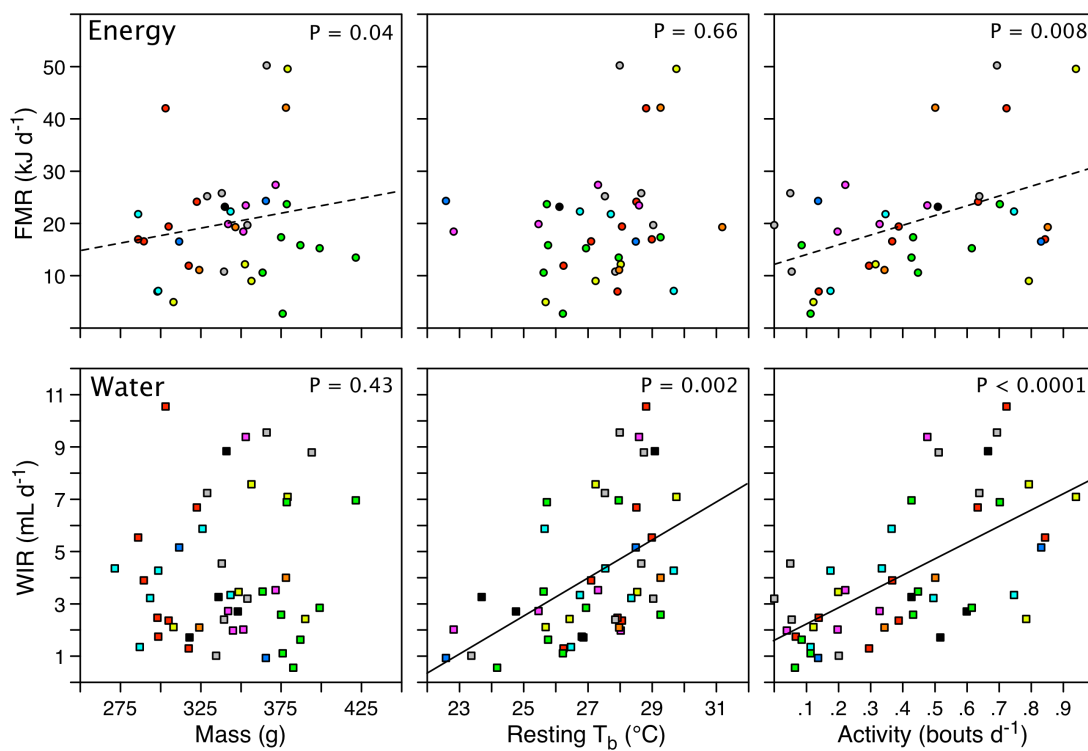


Figure 3. Influences of body mass, resting T_b , and amount of above-ground activity on FMR and WIR in Gila monsters. Activity significantly influences both FMR and WIR, whereas T_b influences only WIR and mass influences only FMR.

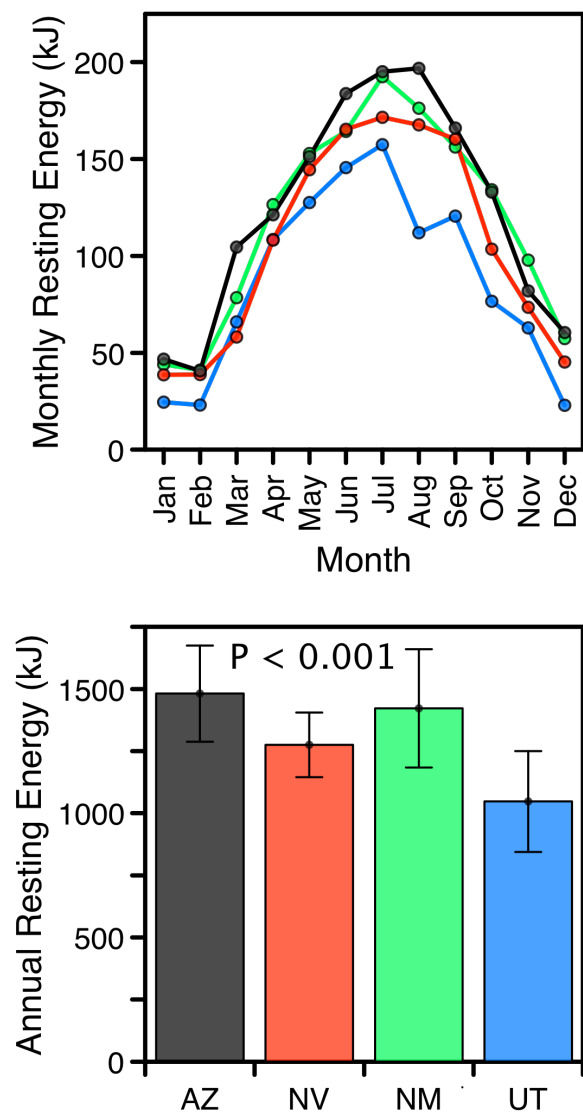


Figure 4. Predicted monthly and annual total resting metabolism (TRM) for Gila monsters from four populations. TRM was calculated for mean population mass and mean monthly resting T_b .