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EFFECTS OF THERMOREGULATION ON FORAGING IN
ANOLIS CAROLINENSIS

LARA R. TROZZO
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Reviewed and approved* by the following:

Margaret Voss
Associate Professor of Biology
Thesis Supervisor

Michael Campbell
Associate Professor of Biology
Honors Adviser

Dr. Roger Knacke
Professor of Physics and Astronomy
Head of School of Science

* Signatures are on file in the Schreyer Honors College.

ABSTRACT

Lara R. Trozzo
Schreyer Honors College Senior Thesis

EFFECTS OF THERMOREGULATION ON FORAGING IN *ANOLIS CAROLINENSIS*

Abstract: Carolina anoles (*Anolis carolinensis*) use behavioral thermoregulation to maintain internal body temperature within an optimal range to support locomotion. They experience a trade-off when foraging for food because they must travel to an area with lower than optimal temperatures, but they cannot stay too long or their body temperature will drop too low to support bodily functions such as digestion. This trade-off was analyzed by observing the thermoregulatory behavior and monitoring internal body temperature of anoles as they traveled between foraging and basking sites. An optimality model was used to analyze the data and calculate optimal body temperature and the percent time the animals allocate to foraging. At warmer temperatures, anoles spend more time away from the basking site to forage; as the temperature decreases, the duration of foraging trips also decreases. Anoles exhibit hysteresis in that they heat at faster rates than they cool. This results in broad optimal temperature ranges to support active foraging behavior. As a result, anoles can allocate over 90% of their time to foraging in relation to basking and appear to have more flexibility in their activity patterns at lower temperatures than was previously thought.

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Introduction

The inspiration for my research came from the unique thermoregulatory behavior of the Galapagos marine iguana (*Amblyrhynchus cristatus*). This terrestrial ectotherm is unusual in that it forages in the Pacific Ocean, a very cool environment. All animals must maintain internal body temperatures within an adequate range to support metabolism; this range is referred to as the animal's operating temperature. The foraging environment of the marine iguana is well below its operating temperature, necessitating its periodic return to rocky basking sites to rewarm and maintain its normal metabolic processes. Understanding the trade-off between the competing functions of foraging and basking is central to understanding the larger problem of how ectotherms manage behavioral time budgets in cold environments. It is not, however, feasible to bring marine iguanas in the lab. It is also difficult to collect the required body temperature data in the field to test ideas about optimal foraging behavior. Thus, my research made use of the Carolina Anole (*Anolis carolinensis*) as a model system to examine ectothermic foraging behavior in cool environments.

Marine iguanas are herbivorous and must dive underwater in relatively cold ocean waters to eat algae. They maintain an appropriate body temperature by basking on lava rocks between diving for food (Badger 2002). Their unique feeding behavior has been the subject of several studies on marine iguana metabolism and energy expenditure. Marine iguanas' foraging behaviors are likely the result of the competing influences of natural and sexual selection. These pressures have caused the development of both physiological and morphological adaptations to meet with their feeding and

thermoregulatory needs. Larger body sizes were selected for to maintain body temperature when foraging in cold conditions. Additionally, male body size has been pushed larger due to intersexual selectiveness and size dimorphism (Vitousek et al. 2007). Consequently, the large size of this lizard, combined with its sparse foraging environment, have resulted in foraging behaviors that are comparable in some ways to herbivorous, grazing lizards and in others to more active, carnivorous lizards (Vitousek et al. 2007). The iguanas have a maximum metabolic rate at optimal body temperatures of 35-36°C, which can only be reached during periods of resting/basking (Bennett et al. 1975). In spite of its unusual diet and foraging behavior, the metabolic rate of the marine iguana is similar to other lizard species (Bennett and Dawson 1976).

Marine iguanas typically allocate 8% of their daily energy expenditure to foraging activities. Their total available energy expenditure is dependent on ambient temperature, which leads to differences in energy budgets between the warm and cool seasons; body temperature can vary as much as 4.7°C between the warmest and coolest parts of the year (Drent et al. 1999). Body size is limited by food intake in these animals, but also directly affects the efficiency of feeding. Bigger animals have a larger bite size and can take in more food in one bite than smaller animals; therefore, they can take in a larger quantity of food within the same duration of foraging. However, this provides less energy per body unit mass, and excessively large animals are constrained to a suboptimal energy balance (Wikelski et al. 1997).

Although I began my research by working with telemetry data for marine iguanas from the work of Maren Vitousek and Martin Wikelski, it became clear that to understand ectotherm foraging behavior in cold environments I would need to use a

comparable model system that I could manipulate experimentally in a laboratory setting. The Carolina anole (*Anolis carolinensis*) is a small lizard (females average approximately 45 mm and males average approximately 61 mm; Schoener and Schoener 1982) of the family Iguanidae commonly found across most of the southeastern United States. As the only anole species native to the US, these small ectotherms experience a wide range of ambient temperatures in their natural environment (Jenssen et al. 1996). Similar to the behavior of the larger marine iguana, anoles use behavioral thermoregulation (shuttling between warmer and cooler environments) to maintain adequate operating temperatures and suitable metabolic rates (Jenssen et al. 1996). To find food, anoles must forage in environments that are often well below their optimum; however, they must subsequently return to warmer basking sites to raise their internal body temperatures, support digestion, and maintain metabolic rate. Although the thermoregulatory and foraging requirements of small lizards may occasionally be matched, they are more often in conflict in highly variable thermal environments (Cabanac 1985). Thus anoles and marine iguanas both face potential tradeoffs; in order to obtain food, the lizards must venture into cooler areas, but the longer they hunt, the lower their body temperature drops. If they stay away from their basking site for an extended time period, their body temperature could drop below the operative temperature, at which point body systems and functions begin to shut down. Given the necessary tradeoff between foraging and basking, we can predict that there must be an optimal time period that a small anole or a larger marine iguana can devote to feeding before it must reheat itself.

Although there have been many studies designed to understand foraging behavior in marine iguanas, little research has been done on anole foraging behavior. Jenssen et al.

(1996) found that anoles are capable of raising their internal body temperature above external temperatures through the use of basking. A significant increase in metabolic rate was seen with increasing body temperatures. Anoles experience ambient temperatures ranging from 37°C to 62°C in their natural environment. In winter, anoles may spend as much as two thirds of their time in the sun. When total time budget of the average anole is considered, 92.2% of time was allocated to resting/basking. The next most commonly observed behavior was foraging (6.4% of total time budget), while other behaviors were less frequent [i.e., agonistic interactions (1.1%), courtship (0.2%), and predator avoidance (less than 0.1%)]. Their highest voluntary body temperature was 34°C and average body temperature was 23°C (Jenssen et al. 1996)

Carolina anoles do not hibernate in winter; they decrease their food intake to compensate for their slower metabolism caused by lower temperatures. They also limit their thermoregulatory behaviors to limit unnecessary energy expenditure (Jenssen et al. 1996). Another study in which anoles' food intake was limited found that after fasting, anoles thermoregulated at lower temperatures. This allowed them to maintain lower metabolic rates and expend less energy (Brown and Griffin 2005).

Modeling Thermoregulation

Since temperature and time are relatively easy to measure, the trade-off between foraging behavior and temperature regulation has been quantified in a number of organisms using several different optimality models. Driesig (1985) suggested that thermoregulating ectotherms should select an optimal temperature in the middle of their activity range. Conversely, several models have been constructed for lizards based on a

dual-threshold model in which temperature is regulated between two limits rather than cycling around one central temperature (Barber and Crawford 1977, 1979). Some lizards may increase the number of foraging trips while simultaneously decreasing their duration when forced to forage at lower temperatures; these animals strategically time basking stops between foraging trips to maintain adequate body temperatures. In this case, thermoregulation can take priority over feeding (Cabanac 1985). When performing activities with a higher metabolic requirement (e.g., digestion), animals may exhibit almost constant basking behaviors with little shuttling in order to maintain a high body temperature. These observations suggest that predictions of foraging time allocation should be modeled based not only on body temperature but also on activity levels (Dreisig 1985).

Before turning to activity levels and behavior observations, it will be important to consider what is known about how ectotherms cool. Several mathematical models of ectothermic cooling have been developed over the years (Barber and Crawford 1979, Bartholomew and Tucker 1963, Dreisig 1985, Turner and Schroter 1985, Voss and Hainsworth 2001). Most are based to some degree on Newton's law of cooling. The simplest first-order model of ectothermic cooling can be described by:

$$T_{Bt} = (T_{B0} - T_e)e^{-k_2t} + T_e \quad \text{Equation 1}$$

Where T_{Bt} is the body temperature at time t , t is time, T_{B0} is the initial body temperature, T_e is the environmental temperature, and k_2 is the cooling rate constant. The same equation can be used to model heating by replacing the cooling rate constant (k_2) with the appropriate heating rate constant (Voss and Hainsworth 2001).

First order models, however, are usually not adequate to describe the complexity of heating or cooling in a vertebrate body. The basic vertebrate body plan includes several layers (e.g., skin, fat, bone, muscle) and compartments (e.g., the peritoneum, the circulatory system, and the digestive tract). When ectothermic vertebrates heat or cool, there are often more than one heating or cooling rate constant in operation (Turner and Schroter 1985). The result is not a linear increase or decrease in temperature with respect to time (i.e., a linear function), but rather a change in temperature that approximates a logarithmic function. This pattern of heating or cooling is better described by a second-order model, such as:

$$T_{Bt} = (T_{B0} - T_{B\infty}) \left(\frac{-k_2}{k_1 - k_2} \right) e^{-k_2 t} + (T_{B0} - T_{B\infty}) \left(\frac{k_1}{k_1 - k_2} \right) e^{-k_1 t} + T_e \quad \text{Equation 2}$$

Where T_{Bt} is the body temperature at time t , t is time, T_{B0} is the initial body temperature, $T_{B\infty}$ is the asymptotic body temperature that would be obtained if the body was allowed to equilibrate with the environment, k_2 is the first-order cooling or heating rate constant, k_1 is the second-order rate constant, and T_e is environmental temperature (Voss and Hainsworth 2001).

These models (*equations 1 and 2*) can be parameterized for a given organism, such as an anole, by taking actual measurements for T_{B0} , $T_{B\infty}$, k_2 , and k_1 under controlled environmental conditions. Ectothermic animals undergo cycles of behavior that include both a time to cool while foraging (t_{cool}), and time to heat while basking (t_{bask}). When cooling and heating rates are known, the models can be used to predict the time an animal can cool and heat for a given set of environmental temperatures. Once these times are known, the percent time (P) an animal can allocate to foraging can be calculated by:

$$P = (100) \frac{t_{cool} - \tau}{t_{cool} + t_{bask}} \quad \text{Equation 3}$$

Where P is the percent of a complete behavioral cycle (i.e., $t_{cool} + t_{bask}$) devoted to foraging, t_{cool} is the time period of cooling (foraging), t_{bask} is the time period of heating (basking), and τ represents the average travel time needed to find food (Hainsworth et al. 1998). The quantity $(t_{cool} - \tau)$ represents the amount of time the animal can actively gain energy, while t_{bask} represents time during which food energy is expended, but heat can be gained.

Hypotheses

My research was designed to address the question of how temperature influences foraging behavior and energy balance in the Carolina anole. I first wanted to understand whether behavioral thermoregulation would limit foraging time. To address this question, I wanted to 1) quantify optimal basking and foraging temperatures and 2) discover what combinations of foraging and basking temperature would allow anoles to maximize the percent of time they allotted to foraging behavior. I predicted lizards would spend less time foraging at cooler temperatures because their body temperature would quickly drop below the required operating temperature and that they would need to return sooner and remain longer at the basking site to compensate. Conversely, I predicted that the percent time allotted to foraging would increase with increasing ambient temperature. Based on these predictions, I believed that anoles would have broad foraging optima at high ambient temperature and more narrow foraging optima at cooler ambient temperatures. Finally, I wanted to identify the point in time at which an extended period of cooling would become too costly for a small anole to balance by

reheating to its operating temperature. This point would represent a temporal trade-off; beyond this point the animal should no longer be able to support foraging trips in a cold environment because they could not compensate by reheating in a timely manner.

Methods

Animal Housing and Care

Adult anoles of known sex (12 males and 12 females) were obtained from Carolina Biological Supply in January 2008. Individuals were distinguished from each other by unique temporary markings placed on their backs using a black sharpie marker. Male and female anoles were housed in two separate long twenty gallon aquaria. The floor of each aquarium was covered with bark bedding; leafy and wooden perches were added to create appropriate structure for the animals. Adjustable basking lights (100W GE Reveal) were set to a 12:12 light:dark cycle. Ambient temperatures and humidity levels were maintained within appropriate ranges (20-30°C with warmer basking spot and 60-70% humidity). The anoles were fed half-inch sized live crickets three times a week, supplemented periodically with a calcium supplement treatment (crickets rolled in calcium powder). They ate about two or three crickets each at a feeding. The crickets were fed on Nature Zone Cricket Total Bites.

Behavioral Thermoregulation Experiment

Experimental Arena

The experimental arena was a twenty gallon aquarium (30 inches wide by 13 inches deep by 13 inches high) with minimal structure for clear observation of shuttling behavior. A basking lamp (150W Zoo Med Basking Spot Lamp) was placed at one end of the tank, with food available at the opposite, cooler end of the tank. This arrangement

created a thermal gradient that was monitored with temperature dataloggers (Onset Computer Corporation HOBO) placed every ten centimeters along the tank. The dataloggers recorded the tank temperatures every five seconds. A grid system was drawn on the tank for recording lizard placement during shuttling trials (Fig. 1).

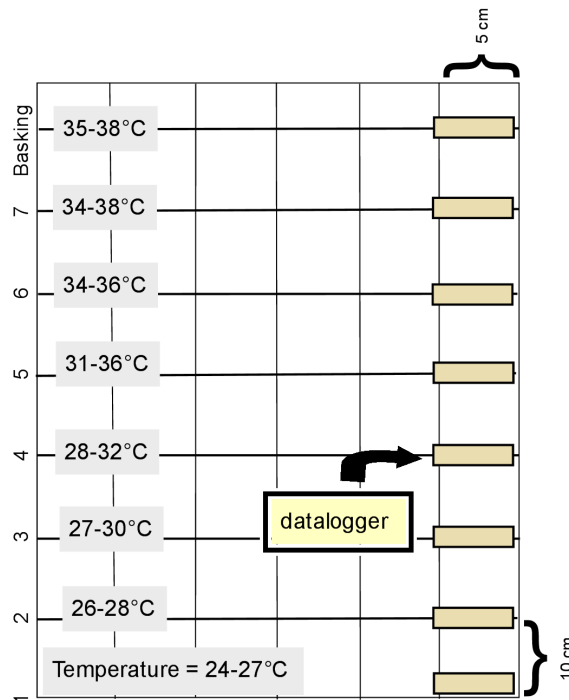


Figure 1: This diagram shows the tank used as the experimental arena. The dataloggers used to monitor the temperature gradient (labeled) were placed every 10 centimeters along the length of the tank.

Data Collection

While in the foraging arena, each anole's body temperature (T_B) was monitored with a fine-scale 24 gauge type T thermocouple inserted a few millimeters into the cloaca and anchored to the tail with surgical tape. The thermocouple was attached to a datalogger which recorded the temperature every five seconds.

During a typical trial, an anole would be weighed, connected to the thermocouple and placed in the experimental arena. After a short acclimation period (5-10 minutes), data collection would begin. In addition to body and environmental temperature measurements, the anole's placement within the arena was recorded every five minutes on a grid sheet corresponding to the tank grid. This mapped the anole's placement in relation to the heat and/or food as well as showing their movements within the tank over time. A trial was typically run for about two or three hours.

Rate Constant Analysis

Temperature data were retrieved from data loggers using BoxCar Pro Version 4.2.4 analysis package and exported to Microsoft Excel. The placement charts were consulted to determine time periods of cooling (moving away from the heated end) or heating (moving towards the heated end). Data were analyzed from periods of cooling behaviors in an excel worksheet using the solver tool and *equation 2* above to calculate k_1 and k_2 , the cooling rate constants. This analysis was performed on cooling episodes for each lizard and the data were compiled (Table 1).

Table 1: Cooling data collected from foraging anoles. For each anole, data were selected from periods where they were observed to shuttle away from the basking area and towards the cooler end of the tank. For each trial, the duration, body temperature, and tank temperature were recorded. The first and second rate cooling constants were calculated.

Date	Animal	Trial	Time cool (min)	Body Temp (°C)	Tank Temp (°C)	k_{c2}	k_{c1}
02/21/08	Female U	1	5	33.17	36.13	0.0000	
02/21/08	Female U	2	5	33.59	36.55	0.0000	
02/21/08	Female U	3	5	33.64	32.76	-0.0001	
02/21/08	Female U	4	5	34.42	37.00	-0.0081	
02/21/08	Female U	5	15	37.74	36.42	0.0000	55.1316
02/21/08	Female U	6	5	37.44	34.01	0.0000	55.1316
03/06/08	Male F	1	5	33.02	31.12	-0.0001	0.0000
03/06/08	Male F	2	10	33.62	34.22	0.0000	52.8790
03/06/08	Male F	3	5	34.92	33.56	-0.0062	54.7710
03/06/08	Male F	4	5	34.03	33.94	0.0002	55.0016
03/06/08	Male F	5	5	34.43	32.20	0.0000	36.5151
03/06/08	Male F	6	5	36.10	34.54	0.0000	55.1316
03/06/08	Male F	7	10	36.10	33.30	0.0000	55.1316
03/20/08	Female M	1	5	39.00	47.43	0.0000	
03/27/08	Male R	1	5	37.32	55.97	-0.0005	
03/27/08	Male R	2	5	38.00	48.95	0.0000	
03/27/08	Male R	3	15	37.57	45.13	0.0003	
03/27/08	Male R	4	5	37.23	47.43	0.0004	
04/03/08	Male J	1	10	35.35	30.10	-0.0162	54.9782
04/03/08	Male J	2	5	32.89	36.57	0.0028	
04/03/08	Male J	3	5	30.69	29.50	0.0001	54.7285
04/03/08	Male J	4	5	26.07	31.53	0.0000	
04/10/08	Female d.d.	1	5	34.10	35.70	0.0000	
04/10/08	Female d.d.	2	5	34.10	35.70	0.0000	
04/10/08	Female d.d.	3	5	37.77	34.63	-0.0002	55.4392
04/17/08	Female C	1	5	37.86	35.27	-0.0004	55.2246
04/17/08	Female C	2	5	38.77	35.27	0.0001	55.1316
04/24/08	Male O	1	5	39.77	34.01	0.0002	55.1316
04/24/08	Male O	2	5	38.63	35.27	0.0001	55.1316
04/24/08	Male O	3	5	40.00	35.27	0.0000	55.1316
04/24/08	Male O	4	5	37.00	34.80	0.0000	55.1316
04/24/08	Male O	5	5	38.67	35.70	-0.0003	55.1356
05/01/08	Male G	1	5	38.00	34.85	0.0000	55.1316
05/01/08	Male G	2	10	37.49	35.07	-0.0004	54.8102
05/01/08	Male G	3	5	39.00	29.50	0.0000	55.1316
05/01/08	Male G	4	15	39.41	32.77	0.0000	55.1316
05/01/08	Male G	5	5	39.00	33.55	0.0000	55.1316
05/01/08	Male G	6	5	39.00	35.27	0.0000	55.1316
05/01/08	Male G	7	5	39.00	35.06	0.0000	55.1316

Data for use in the foraging models were taken only from periods when the initial body temperature (T_{B0}) was higher than the tank temperature (T_e) at $t=0$ and a negative k_2 was present (Table 2). The cooling data were first analyzed in an excel spreadsheet to find the cooling rate constants k_1 and k_2 for use in *equations 1* and *2* above. To determine which model was most appropriate (a first order or a second order model of cooling), the isolated cooling rate constants were used in *equations 1* and *2* above to compare with the observed changes in body temperature (T_{Bt}). There was no statistical difference between data points from the first order and second order curves (paired t-test, $p=0.08$, $df=43$), thus the first order model and a single rate constant was used for simplicity.

Foraging Modeling for Different Environmental Temperatures

The rate constants from Table 2 were used in *equation 1* to predict how long a lizard could forage (t_{cool}) before experiencing a temporal trade-off at three environmental temperatures (15°C, 5°C, and 0°C). The point of trade-off was determined by examining a plot of t_{cool} paired with the time to reheat to the animal's original body temperature. In general, ectotherms experience cooling hysteresis; they reheat faster than they cool at a given environmental temperature. The hysteresis effect means that graphs of ectothermic cooling and heating tend to be asymmetrical; however, there is often a point at every environmental temperature at which heating may take as long, if not longer than the concomitant time to cool. This is the point of the trade-off; at this point, the animal is spending more time foraging (cooling) than it can easily compensate for during reheating. This trade-off can also be quantified by using the times required to cool and reheat at a

temperature in *equation 3* to predict the optimal time allocations of foraging and basking at each environmental temperature.

Table 2: Data used in the calculation and analysis of cooling rate constants. Data were selected in which T_{B0} was higher than T_e and k_{c2} was negative. The first and second order models were used to predict T_{B5} which was compared with the observed T_{B5} . The predicted values from the models were not statistically different (paired t-test, one-tailed, 9 df, $t_{crit}=1.83$, $p=0.08$).

Actual		Observed			Predicted	
T_e	T_{B0}	T_{B5}	k_{c2}	k_{c1}	T_{B5}	T_{B5}
36.42	37.44	37.88	0.0000	55.1316	37.44	37.44
34.01	37.44	37.44	0.0000	55.1316	37.44	37.44
31.12	32.20	34.10	-0.0001	53.8558	32.20	32.23
34.22	35.10	32.20	0.0000	52.8790	35.10	35.09
33.56	34.10	36.10	-0.0062	54.7710	34.09	36.98
33.94	35.10	31.20	0.0002	55.0016	35.10	35.05
32.20	32.20	32.20	0.0000	36.5151	32.20	32.20
34.54	36.10	36.10	0.0000	55.1316	36.10	36.10
33.30	36.10	36.10	0.0000	55.1316	36.10	36.10
29.50	33.10	27.30	0.0001	54.7285	33.10	32.99
34.63	37.00	37.00	-0.0002	55.4392	37.00	37.17
35.27	37.00	38.00	-0.0004	55.2246	37.00	37.19
35.27	39.00	39.00	0.0001	55.1316	39.00	38.91
34.01	40.00	40.00	0.0002	55.1316	39.99	39.72
35.27	39.00	39.00	0.0001	55.1316	39.00	38.88
35.27	40.00	39.00	0.0000	55.1316	40.00	39.98
34.80	37.00	37.00	0.0000	55.1316	37.00	37.00
35.70	38.00	39.00	-0.0003	55.1356	38.00	38.25
34.85	38.00	38.00	0.0000	55.1316	38.00	38.00
35.07	36.10	39.00	-0.0004	54.8102	36.10	36.22
29.50	39.00	39.00	0.0000	55.1316	39.00	39.00
32.77	39.00	40.00	0.0000	55.1316	39.00	39.05
33.55	39.00	39.00	0.0000	55.1316	39.00	39.00
35.27	39.00	39.00	0.0000	55.1316	39.00	39.00
35.06	39.00	39.00	0.0000	55.1316	39.00	39.00

Results

Rate Constants

The average first order cooling rate (k_{c2}) observed for 25 foraging trials was $3.2(\pm 5.7) \times 10^{-5}$, while the average warming rate (k_{w2}) was $1.3(\pm 2.2) \times 10^{-3}$. The predicted body temperatures from the two models were not statistically different (paired t-test, one-tailed, 9 df, $t_{crit}=1.83$, $p=0.08$).

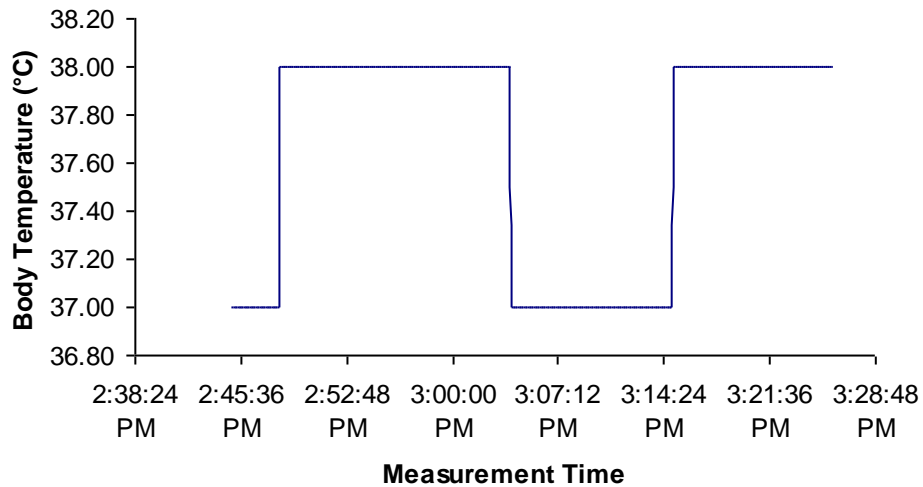


Figure 2: *This plot of T_B versus time shows the typical thermoregulatory shuttling cycle. This lizard starts out foraging then moves towards the warmer end of the tank and basks, forages, then basks again.*

Predicted Foraging Times

An environmental temperature of 15°C , which is within the thermal neutral zone (TNZ) for Carolina anoles, allowed the animals to forage for 3092 seconds before they experienced a temporal trade-off at a body temperature of 36.76°C (Fig. 3).

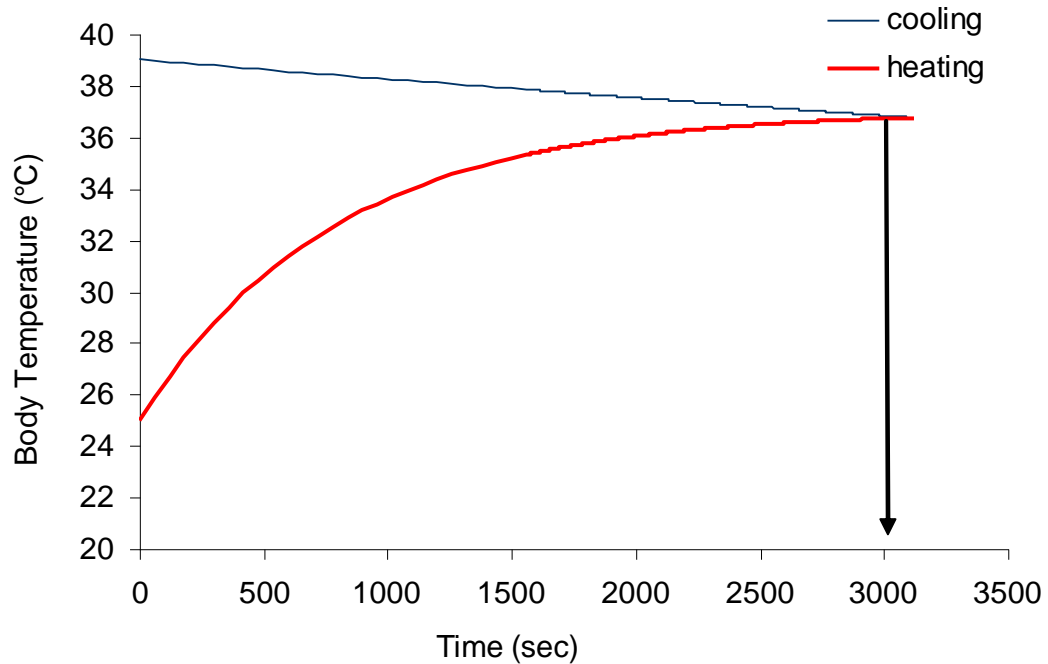


Figure 3: *Temperature trade-off at 15°C, within TNZ. Changes in body temperature conform to a negative exponential decay during cooling or increase exponentially during heating, depending on initial body temperature. At an environmental temperature of 15°C, the times required to cool and reheat trade-off after 3092 seconds. This is the point when anoles must stop foraging and return to the basking site in order to maintain their body temperature at 36.76°C; beyond this point, the time to reheat cannot compensate for the extended cooling time.*

At temperatures higher than 36.76°C, the relatively low rate of cooling allows the animals to forage for an extremely long period of time before moving out of their thermoneutral zone and into a range below their operating temperature (below 25°C). At these higher exit temperatures, the relatively rapid rate at which the animals reheat allows the animal's time budget to favor longer times to cool and therefore longer times to forage. If the animal left the basking site at 36.76°C, it could not cool for very long before having to return to the basking site to reheat to 36.76°C. The limitation occurs because at the lower temperature, the time to cool, and therefore forage, is so tightly

constrained. The animal is unlikely to have enough time away from the basking site to find food before it must return to an operative temperature that can support its metabolism.

At a lower environmental temperature of 5°C, which is below the anoles' thermal neutral zone, the animals could forage for a shorter amount of time, 2457 seconds. At this point, their body temperature would be 36.46°C (Fig. 4), when they experienced a trade-off between foraging time and the necessary time to reheat as described above.

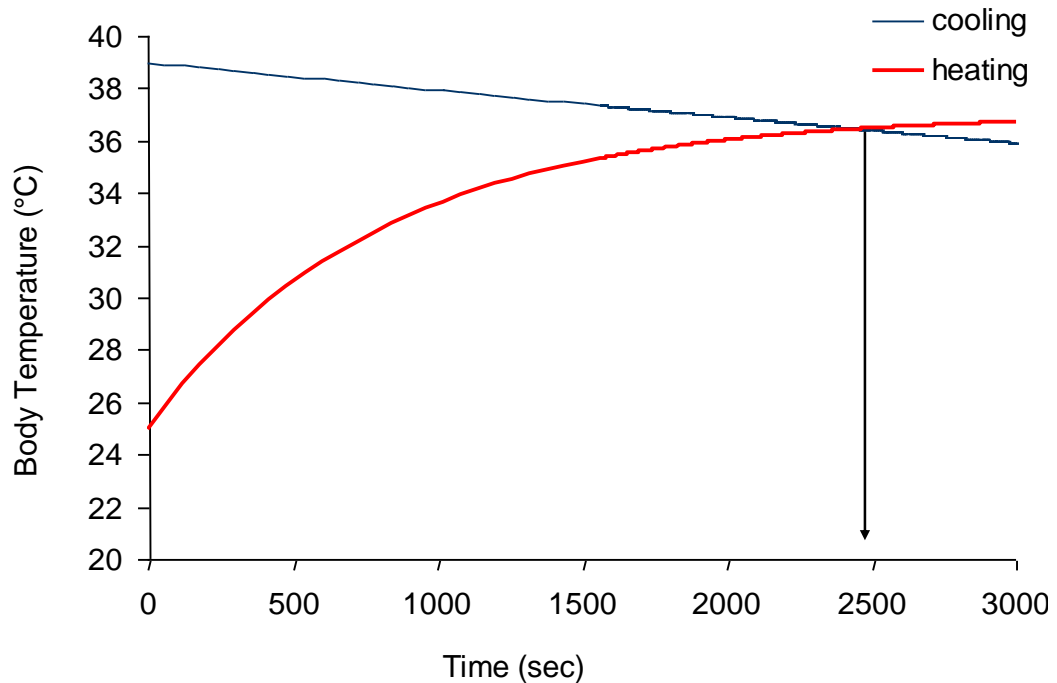


Figure 4: *Temperature trade-off at 5°C, below TNZ. Compared to environmental temperatures within their thermal neutral zone, at lower temperatures, cooling occurs more rapidly and heating more slowly. The animals can forage for a shorter duration of 2457 seconds before experiencing a trade off that causes them to switch from foraging to basking in order to regulate their body temperature at 36.46°C, which is slightly lower than they maintained when within their thermal neutral zone.*

Finally, an environmental temperature of 0°C was modeled. This is well below anoles' thermal neutral zone and lower than would be usually seen in their natural habitats. According to the first-order model, they can still forage for 2263 seconds before achieving the trade-off between foraging time and time required to reheat at a body temperature of 36.21°C (Fig. 5). These results suggest that anoles can still function as external temperatures approach freezing, which was somewhat unexpected. However, it is not completely surprising as these animals do not hibernate and temperatures of 0°C do sometimes occur in their natural environment.

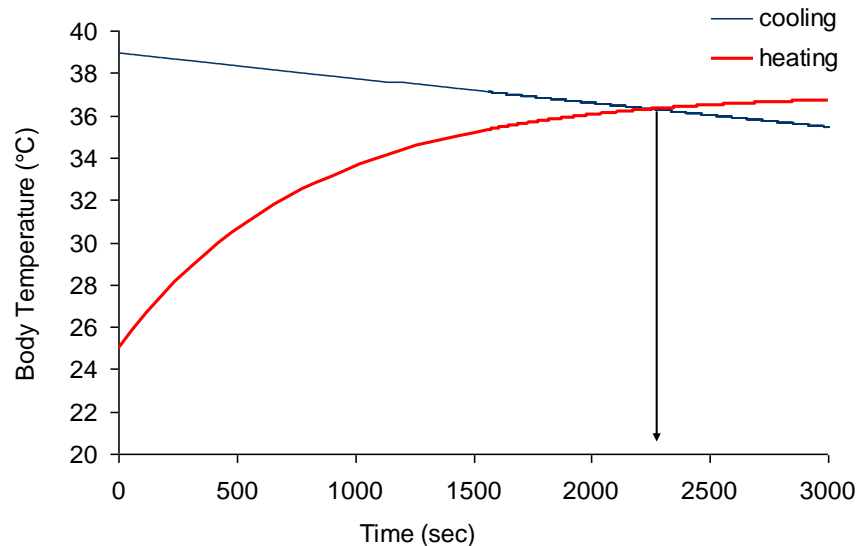


Figure 5: Trade-off between time to cool and time to reheat at 0°C, well below TNZ and normal environmental temperatures. The times an anole could cool (forage) and reheat (bask) became more similar at lower environmental temperatures; the animals would be at a breakeven point. Foraging is still possible, but would be costly in terms of the basking time required to offset cooling. At these temperatures, anole metabolism would be suboptimal; however, these conditions are not entirely outside of the anoles' normal temperature range. In order to maintain operative body temperatures, their foraging trips were more time-limited (2263 seconds) before they had to return to basking or suffer physiological damage. The animals would regulate their body temperature at about 36.21°C in these conditions.

Predicted Time Allocations

At environmental temperatures within their thermal neutral zone, anoles can forage over a variety of temperatures and for a significant amount of time (Fig. 6). They demonstrated a broad optimum, showing that temperatures between 25 and 37°C are adequate to support extensive foraging time. Within the optimal range, the animals can allocate about 92.75-97.83% of their time to foraging behaviors; while requiring only 2.17-7.25% of their time for basking.

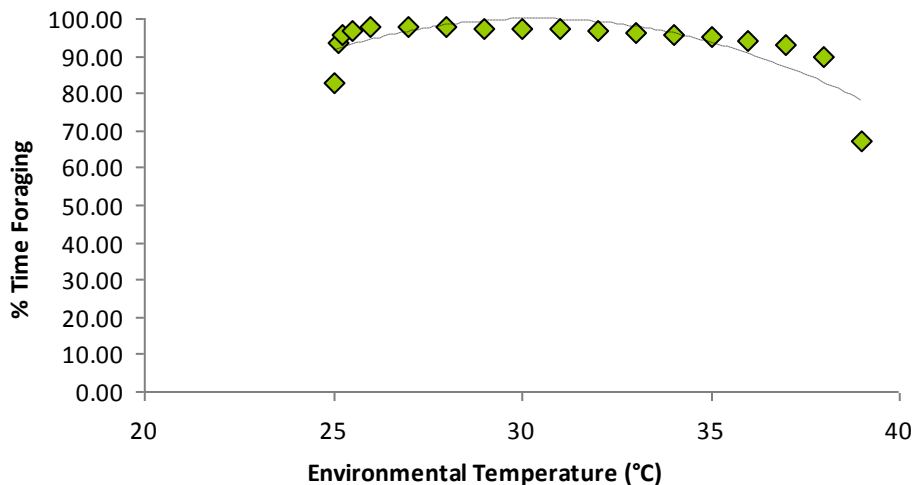


Figure 6: *Percent time allocated to foraging at Temperatures in TNZ. At environmental temperatures within their thermal neutral zone, the anoles could spend most of their daily time budget foraging over a wide range of temperatures (26-37°C). They could maximize their foraging and allocate about 92-98% of their time to foraging behaviors, only using the other 2-8% for basking.*

At lower environmental temperatures of 5°C, the optimum was equally broad (Fig. 7). Anoles could still theoretically allocate 92.75-97.83% of their time to foraging. The optimum occurred at temperatures ranging from 6-17°C.

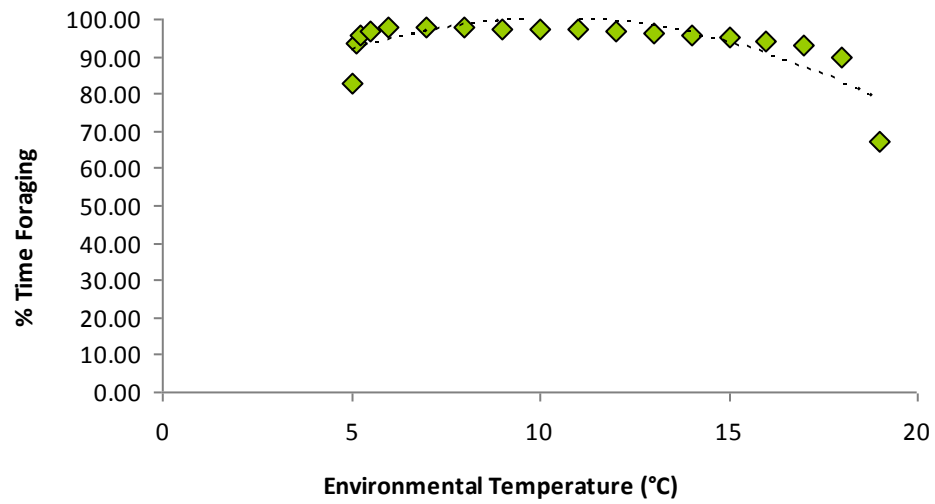


Figure 7: *Percent time allocated to foraging at Temperatures below TNZ. The anoles have the ability to allocate large percentages of time to foraging even at low external temperatures. This is likely due to physical adaptations that permit the animals to cool slowly, but reheat quickly, thus maximizing the time they can devote to foraging under a wide range of thermal conditions.*

Discussion

The Hysteresis Effect

The broad optima in percent time foraging predicted for Carolina anoles is likely due to an extreme thermal hysteresis effect. Hysteresis was apparent in the difference between the cooling and heating rate constants measured during the foraging trials in this experiment. The animals clearly heat faster than they cool which implies that they can spend a short amount of time heating to compensate for long periods of time spent away from their basking sites to forage. At the same time, the extremely low cooling rate constants suggest these animals have a large capacity to prolong the time they can devote to foraging.

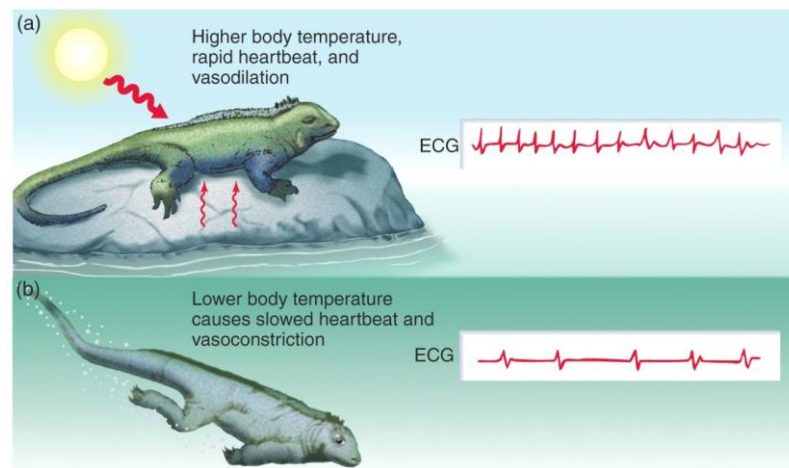


Figure 8: *The hysteresis effect. Most animals tend to heat faster than they cool. On land, the basking marine iguana absorbs heat from the sun's rays. Vasodilation of cutaneous blood vessels and rapid heart beat assure efficient circulation and heating of blood. This quickly dissipates heat throughout the body. The trend is reversed in cool water; rate of heat loss is higher in cool water than rate of heating. (Figure from Randall, et al. 2002; Eckert Animal Physiology).*

Optimal Temperatures

The trade-off analysis showed that as environmental temperature decreased, the amount of time anoles could forage and the temperature they could allow their body temperature to drop to decreased, but not nearly as much as expected. It would appear that the 10°C temperature range of the experimental arena was large enough to accurately capture the trade-off. The results suggest that anoles can withstand much cooler environmental temperatures than previously suspected. The models predict that even at very cool temperatures, anoles can forage for a long time before their body temperature drops far enough to place limits on foraging and requires them to bask or suffer physiological damages.

Time Allocations

The calculated time budgets showed that the anoles can allocate a very large percent of their time to foraging relative to basking. This, of course, is dependent on environmental temperature. Based on the data and the models, there are multiple equally optimal temperatures that support a high percent of time foraging within the anole thermoneutral range. At all of these temperatures, an extremely high percentage (over 90%) of the overall time budget time could be devoted to foraging, with only the remaining small portion of time required for basking. At these higher environmental temperatures, the anoles are less limited and their body temperature does not drop as quickly. This clearly illustrates the hysteresis effect in that they can afford to allocate such a small percentage of time to basking due to the higher warming rates.

It was expected that at temperatures below their thermal neutral zone, both the width of the optimal temperature range and the percent time spent foraging would decrease dramatically. However, this did not occur and it appears that the anoles can still forage for large percentages of their time. This is a function of both the hysteresis effect and the fact that the data used in the calculations was unexpectedly well within the anole thermoneutral range. It is important to realize that the percent time of foraging does not specify the duration or frequency of individual foraging/basking cycles, rather the overall sum of foraging activity relative to basking.

Although my result suggest a broad optimal range of temperatures that maximize foraging behavior, it is also possible that there are multiple discrete optima that this experiment could not differentiate. Although this has not been examined for anoles, some ectotherms are known to have physiological adaptations that allow them to cope with extreme changes in environmental temperature; these animals often exhibit optimal functioning peaks at multiple temperature ranges. For example, some animals have enzymes which function optimally in different thermal environments. The muscle lactate dehydrogenase (M₄-LDH) enzyme has two different homologues which occur in animals that live in different habitats (polar compared to desert). These enzymes are related but have become specialized to function at different temperatures; they denature or function suboptimally when exposed to the wrong thermal environment. Differential temperature LDH isozymes may be present in the same organism, causing it to have multiple temperature optima for physiological functioning (Willmer et al. 2000).

The plots for percent time allocated to foraging show clear optima, yet there is not a significant decrease in functioning at lower temperatures. This raises the possibility

that anoles may have isozyme adaptations similar to the LDH example described. This would be an advantage in their natural environment. The weather in southeastern US is typically well within optimal operating temperature range for anoles; occasionally however (e.g. the winter of 2009-2010), record cold spells would present ambient temperatures much lower than those within anoles' thermal neutral zone. At this point, alternate isozymes and enzyme systems may be activated, allowing anoles to survive a harsh winter even if they are functioning at slightly lower than normal capabilities.

Carolina Anoles compared with Galapagos Marine Iguanas

Similar to the anoles, marine iguanas are known to exhibit the hysteresis effect (Fig. 7). Originally, 10 years of data collected from marine iguanas in the Galapagos Islands by Maren Vitousek and Martin Wikelski were reanalyzed for part of this project. However, without fine resolution behavioral data to correlate with the temperature data, few of the data could be reliably interpreted. The iguana data also seemed to suggest broad temperature optima. Although no concrete conclusions could be made due to the small amount of data used, I suspect behavioral patterns similar to those shown in the anole data. It is likely that marine iguanas and anoles have similar thermal characteristics including: evidence of behavioral thermoregulation, broad temperature optima, and adaptations allowing for functioning at multiple temperatures.

Conclusion

This study was the first to monitor internal body and external environmental temperatures correlated with position to quantify behavioral thermoregulation in anoles. The first and second order heating and cooling models were used in data analysis and gave comparable results. The first-order models were used to calculate cooling and heating rates. Because heating occurs at a faster rate than cooling, these lizards exhibited the hysteresis effect, similar to what is seen in Galapagos marine iguanas and some other ectotherms.

Optimal foraging conditions were predicted; the anoles could forage for longer periods of time and maintain body temperatures closer to optimal when foraging and basking at environmental temperatures within their thermal neutral zone. At lower optimal temperatures, they should maintain a slightly lower body temperature by foraging for shorter durations with more frequent returns to their basking site.

An analysis of the rate of net energy gain showed that anoles can allocate over 90% of their time to foraging at optimal temperatures and only require less than 10% of their time for basking. The range of optimal temperatures was broad and expected to be narrower at decreased temperatures; however, these results were equally broad with associated high percentages of time to allocate to foraging. This suggests that anoles have adaptations allowing them to maintain successful functioning when faced with cold temperatures in the winter. Thermal behavior characteristics may be more flexible and less limited than previously thought.

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Curriculum Vita of Lara Trozzo

2219 Manordale Dr.
Export, PA 15632
724-552-7362

Penn State Behrend MB#1108
4701 College Dr.
Erie, PA 16563

Education:

Penn State Erie, The Behrend College
Biology - General Option, Bachelor of Science, May 2010
Chemistry, Minor

Research Experience:

Great Lakes WATER Institute REU Summer 2009
• Project: *Feeding Habits of Hemimysis anomala*
Schreyer Honor Scholar Research 2007-2010
• Thesis: *Effects of Thermoregulation on Foraging in Anolis carolinensis*

Relevant Course work:

Aquatic Ecology	Theoretical Populations Ecology
Comparative Vertebrate Anatomy	Animal Behavior
Physiological Ecology	Mammalian Physiology
Experimental Design	Evolution
Organic Structure	X-ray Crystallography

Skills:

Animal handling and care	Dissection
Chemistry lab techniques	Respirometry
Thermocouples/ Dataloggers	Spectroscopy
Microsoft Office: Word, Excel, PowerPoint, Access	SAS

Teaching Experience:

Teaching Assistant – Animal Behavior Spring 2010
Comparative Vertebrate Anatomy Fall 2009
• Assist professor with setting up and running labs, and grading
STEM Mentor 2008-2010
• Mentor incoming students in STEM Scholarship Program.

Honors and Activities:

Presidential Scholarship for Penn State Erie	2009-2010
Sigma Xi Undergraduate Research Conference	2009
PSU Schreyer's Honors College	2008- 2010
Behrend College Scholarship	2008-2009
Beta Beta Beta Biological Honor Society (Historian)	2007-2010
Dean's List	2006-2010
Biology Club	2006-2010
Newman Club	2006-2010