

## Low repeatability of preferred body temperature in four species of Cordylid lizards: Temporal variation and implications for adaptive significance

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**Abstract** Preferred body temperatures ( $T_{\text{sel}}$ ) of ectotherms are important for ecological and evolutionary studies. In lizards, the measurement of  $T_{\text{sel}}$  is controversial for several reasons, generally related to hypotheses addressing how  $T_{\text{sel}}$  may evolve in the wild. Although seldom explicitly tested, evolutionary hypotheses of adaptation to local climate require that  $T_{\text{sel}}$  meets the conditions of natural selection, which include repeatability, heritability and a link to fitness. Here, we investigated repeatability ( $\tau$ , intra-class correlation coefficient) of  $T_{\text{sel}}$  at several time-scales using four Cordylid species from heterogeneous thermal habitats. Although there was significant inter-individual variation within days ( $P < 0.005$  in most cases), there was no significant inter-individual variation when calculated across several days ( $P > 0.05$ ). Repeatability was low in all species investigated (from 0 to 0.482) when compared against other estimates of repeatability of  $T_{\text{sel}}$  in the literature. Irrespective of how  $T_{\text{sel}}$  was calculated, it showed inconsistent and variable temporal effects across species. Furthermore, repeatability of  $T_{\text{sel}}$  did not change with acclimation to laboratory conditions. These data have implications for understanding the evolution of thermoregulation in these and other ectotherms.

**Keywords** Thermoregulation · Coadaptation · Inter-individual variation · Selection · Temporal effects · *Cordylus*

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## Introduction

Preferred or selected temperature ( $T_{\text{sel}}$ ) is defined as the range of body temperatures ( $T_{\text{b}}$ ) maintained by an organism in a laboratory thermal gradient that provides an array of thermal environments free of physical and biotic constraints (Licht et al. 1966). In reptiles,  $T_{\text{sel}}$  may represent the  $T_{\text{b}}$  at which numerous processes (e.g. locomotion, digestion) function at an optimal level (Stevenson et al. 1985; Angilletta et al. 2002a, b; but see Angilletta and Werner 1998). Attention is given to  $T_{\text{sel}}$  for two main reasons. First, there is the advantage of defining thermal behavior in a laboratory set-up where the researcher can control environmental conditions (e.g. photoperiod, temperature) and eliminate ecological constraints (e.g. predation, costs of thermoregulation). Second, since the work of Hertz et al. (1993),  $T_{\text{sel}}$  has become a key component in quantitative indices of temperature regulation (e.g.  $\bar{d}_{\text{b}}$ , the mean absolute value of deviations of field-active  $T_{\text{b}}$ 's from  $T_{\text{sel}}$ ). These indices include the assumption that  $T_{\text{sel}}$  of an organism reflects the temperature that the animal attempts to achieve during thermoregulation in nature (Licht et al. 1966; Huey 1982). However,  $T_{\text{sel}}$  is not a simple characteristic, and by contrast, reflects a complex interplay of physiological and behavioral mechanisms (Huey 1982; Angilletta et al. 2002b; Seebacher and Franklin 2005). Consequently, studies of  $T_{\text{sel}}$  are controversial for several reasons, of which three are perhaps most significant.

First,  $T_{\text{sel}}$  observed in a thermal gradient can reflect the temperature for optimum ( $T_{\text{opt}}$ ) physiological performance. For example,  $T_{\text{sel}}$  correlates with the  $T_{\text{opt}}$  of several tissue and cellular functions (Licht et al. 1966; Dawson, 1975; Beitingger and Fitzpatrick 1979), and also with locomotor performance (Bauwens et al. 1995) and digestion (Du et al. 2000; Angilletta 2001). The coadaptation hypothesis (Huey and Bennett 1987; Angilletta et al. 2006) predicts that  $T_{\text{sel}}$  corresponds to the  $T_{\text{opt}}$  for physiological performance. By selecting  $T_{\text{b}}$ 's that maximize performance, individuals could achieve greater fitness. However, results from studies reporting correlations between  $T_{\text{sel}}$  and  $T_{\text{opt}}$  have provided mixed support (reviewed in Angilletta et al. 2002b).

Second,  $T_{\text{sel}}$  may result from an adaptation to diverse thermal regimes (Huey and Slatkin 1976; Huey 1982; Huey and Kingsolver 1989; Angilletta et al. 2002b). For example, *Anolis* lizards show striking differentiation between  $T_{\text{b}}$  and  $T_{\text{sel}}$  along an altitudinal gradient (Huey and Webster 1976). If  $T_{\text{sel}}$  evolves by means of natural selection, it must meet three requirements: it should be heritable, have consistent inter-individual variation, and be linked to the fitness of the species in question (Endler 1986). Thermal preference may be heritable as significant variation in  $T_{\text{b}}$  is sometimes associated with family membership (Sinervo 1990), thereby suggesting a genetic basis (see also Huey and Kingsolver 1989; Hoffmann et al. 2003). Although  $T_{\text{sel}}$  varies among genera (e.g. Dawson 1975; Angilletta and Werner 1998),  $T_{\text{sel}}$  is often conserved within genera (Huey 1982; Bennett and John-Alder 1986), indicating that thermoregulatory behavior might be constrained and evolve slowly (Huey 1982). In general, however, there is little direct evidence (e.g. via selection experiments, genetic correlations) that  $T_{\text{sel}}$  responds to natural selection (but see Good 1993). Understanding the evolutionary factors determining  $T_{\text{sel}}$  is important for the establishment of a readily interpretable measure of thermal preference for physiological, ecological and evolutionary studies (see discussion in Angilletta et al. 2006).

Third, there is a discrepancy in the way that  $T_{\text{sel}}$  is measured and analyzed. According to several studies, lizards thermoregulate between lower and upper set-points (dual set-point model) rather than around a single  $T_b$  (Berk and Heath 1975; Barber and Crawford 1977). While some studies report the mean  $T_{\text{sel}}$  (e.g. Lailvaux et al. 2003), others determine the median of the frequency distribution of all individuals pooled together (Bauwens et al. 1995), or the mean of individual upper and lower set-point temperatures (e.g. Christian and Weavers 1996). Furthermore, there is variation in the determination of set-point temperatures (e.g. the central 50% (Hertz et al. 1993) or 80% (Bauwens et al. 1995) of the data) even though this difference will affect the values of temperature regulation indices (Hertz et al. 1993; Wills and Beaupre 2000). Therefore, in order to infer adaptive significance, a relevant measure of  $T_{\text{sel}}$  is required. Moreover,  $T_{\text{sel}}$  may result in significant diel (Firth and Belan 1998; Angilletta et al. 1999) and seasonal shifts (Sievert and Hutchison 1989; Christian and Bedford 1995), and may vary with sex (Patterson and Davies 1978), physiological state (e.g. pregnancy, feeding status; Rock et al. 2000; Brown and Griffin 2005) and temperature acclimation (Wilhoft and Anderson 1960). Therefore, the use of an appropriate time scale and physiological state may be important for some species and is often neglected in studies of lizard thermoregulation (Seebacher 2005). Finally, because the measurement of temperature indices is confounded by temporal dependence of data at several time scales (Gaines and Denny 1993), clarification of temporal variation in  $T_{\text{sel}}$  is especially important for predictions of ectotherm responses to climate change (Helmuth et al. 2005).

The determination of consistent inter-individual variation (or repeatability sensu Falconer and Mackay 1996) has played a central role in linking functional and evolutionary investigations of physiology and behavior (e.g. Boake 1989; Bertheaux et al. 1996). By partitioning phenotypic variance into within- and between-individual components, repeatability ( $\tau$ ) can give a first indication of whether  $T_{\text{sel}}$  is highly variable among individuals, consistent over time and, might have evolved by means of natural selection (see discussion in Bennett 1987, 1997; Dohm 2002). If repeatability of  $T_{\text{sel}}$  is high, this trait may be easily predicted within the chosen time-frame, and may have evolved as a response to selection. Alternatively, if repeatability is low (i.e. there is a greater within- than among-individual variation), variation in  $T_{\text{sel}}$  is unlikely to translate into variation in fitness. In the latter case, a number of genetic and environmental causes of variation among individuals would need to be considered before assuming that there are no genetically based differences among them (see Dohm 2002). A lack of repeatability may simply indicate that an ecologically relevant time-frame has not been chosen or that the methodology used is flawed (Hayes et al. 1998).

Despite many reptilian studies reporting  $T_{\text{sel}}$  (mostly with snakes and lizards), few have examined the extent of intra- and inter-individual variation of this trait (but see Christian et al. 1985; Peterson and Arnold 1986). In fact, repeatability of  $T_{\text{sel}}$  is largely unknown (Van Damme et al. 1990; Wills and Beaupre 2000). Repeatability determination can provide insight into innate variation of this trait and offers an indication of whether or not  $T_{\text{sel}}$  can be influenced by natural selection.

In this study, we addressed the following questions: (1) Is the  $T_{\text{sel}}$  of individual lizards repeatable at several time scales (within a day, among days, and before and after acclimation to controlled laboratory conditions)? (2) Is  $T_{\text{sel}}$  equally repeatable when expressed as a mean (central tendency) or as lower and upper set-point temperatures? If  $T_{\text{sel}}$  is adaptive in the strict sense, such that it confers differential

fitness, we should find high repeatability both within and among days (unless it was repeatable in the past but subsequently lost its function). This study was undertaken on lizards of the family Cordylidae, in which the majority of species are distributed in southern Africa, and for which little information is currently available regarding their thermal biology.

## Materials and methods

### Study organisms

We collected adults of four species of Cordylid lizards (*Cordylus cordylus*, *Cordylus niger*, *Cordylus polyzonus* and *Cordylus oelofseni*) in the Western Cape of South Africa during summer (January–March) of 2004 and 2005 (See Table 1 for species and site characteristics). This region is temperate and seasonal (average ambient temperature in summer  $\sim 21.7^\circ\text{C}$  and winter  $\sim 12.8^\circ\text{C}$ ). All four species are rock dwelling and shelter in rock crevices. They are characterized as sit-and-wait predators, viviparous and primarily insectivorous (SCT pers. obs.; Cooper et al. 1997). We transported all lizards within the same day of collection to the University of Stellenbosch where they were maintained at  $20 \pm 3^\circ\text{C}$  in glass terrariums ( $90 \times 50 \times 60$  cm) that contained rocks, crevices and water ad libitum. The environmental chamber housing the terrariums fluctuated by  $\sim 6^\circ\text{C}$  daily. Not more than five individuals were kept in the same terrarium. Fluorescent bulbs provided a similar photoperiod to natural conditions (14:10 h L:D). Work on a single species was completed before continuing with the next species (i.e. four experimental blocks of equal duration).

### Thermal gradient

We measured  $T_{\text{sel}}$  in a laboratory photothermal gradient established within 10 separated runways. Each runway had a  $130 \times 30$  cm floor covered with small ( $\sim 1$ – $3$  cm diameter) rocks. We produced the thermal gradient by maintaining the controlled-environment room at a temperature of  $15^\circ\text{C}$  and by placing a 250-W infrared bulb at one end of each runway. The temperature of the gradient increased gradually from  $17^\circ\text{C}$  to  $55^\circ\text{C}$ , was monitored daily and stayed consistent throughout experiments. The room was equipped with three 30-W fluorescent tubes for illumination.

We monitored body temperatures of each lizard by inserting a 36 SWG thermocouple 1–1.5 cm into the cloaca of *C. cordylus*, *C. niger* and *C. oelofseni* and

**Table 1** Characteristics of Cordylid lizards used for the estimation of preferred temperature ( $T_{\text{sel}}$ )

Species	$N$	SVL (mm)	$M_1$ (g)	$M_2$ (g)	Site location	GPS coordinates	Altitude (m asl)
<i>C. cordylus</i>	12	$74.5 \pm 4.4$	$13.9 \pm 2.8$	$14.3 \pm 3.1^*$	Inland	$33^\circ 46' \text{ S}, 18^\circ 47' \text{ E}$	289
<i>C. niger</i>	10	$74.9 \pm 2.7$	$14.0 \pm 1.5$	$14.0 \pm 1.5$	Coastal	$32^\circ 59' \text{ S}, 17^\circ 52' \text{ E}$	5
<i>C. polyzonus</i>	11	$96.3 \pm 7.0$	$35.8 \pm 7.4$	$35.2 \pm 7.8$	Coastal	$32^\circ 59' \text{ S}, 17^\circ 52' \text{ E}$	5
<i>C. oelofseni</i>	10	$60.8 \pm 3.1$	$8.2 \pm 0.7$	$7.9 \pm 0.6$	Montane	$34^\circ 02' \text{ S}, 19^\circ 00' \text{ E}$	1080

\* $P = 0.046$

Snout-vent length (SVL) was determined upon field collection. Body mass was measured upon field collection ( $M_1$ ) and after acclimation ( $M_2$ ).  $M_1$  and  $M_2$  are compared using paired  $t$ -tests and significant differences are indicated by (\*). Means  $\pm$  standard deviation

1.5–2 cm for *C. polyzonus*. The trailing thermocouple was taped with surgical tape around the tail and secured with a thin cable tie. The thermocouple was suspended 1 m above the gradient floor to enable free movement of a single lizard along the gradient and was removed every day after measurement periods. We connected all thermocouples to an AM32 Multiplexer linked to a CR10 datalogger (Campbell Scientific, Utah, USA). The datalogger was calibrated before use against a digital thermometer ( $\pm 0.1^\circ\text{C}$ ; BAT-12, Physitemp, USA) and recorded body temperature every 5 min for a minimum of 7 h for three consecutive days starting the day after capture (Day 1). After each  $T_{\text{sel}}$  trial, lizards were placed in original terrariums overnight and maintained at  $20^\circ\text{C}$ . Thus, all individuals were in a similar acclimation state throughout  $T_{\text{sel}}$  experiments. Measurements occurred between 1000 and 1800 h to correspond with observed activity periods for each species in the field. We repeated this schedule after the animals were acclimated at  $20(\pm 3)^\circ\text{C}$  for seven more days (14:10 h L:D). Water was accessible ad libitum during  $T_{\text{sel}}$  trials. Lizards were fed with mealworms ad libitum during the acclimation period, but were deprived of food 2 days before the acclimated trials. We assigned runways randomly every trial day. Lizards that became entangled with the thermocouple or did not move during trials were eliminated from the statistical analysis for that particular day. Data recorded during the first hour of  $T_{\text{sel}}$  measurements were considered to reflect the lizard's habituation to the new environment and were eliminated from the analysis. We did not observe lizards during  $T_{\text{sel}}$  trials to avoid disturbance.

## Analyses

Autocorrelation of data collected every 5 min in the thermal gradient was analyzed using Time Series analysis in STATISTICA (v. 6.0, Statsoft Inc., USA) for each individual. Typically,  $T_b$ 's recorded 10–15 min apart were independent of each other. Since, in most cases, data were normally distributed (Shapiro-Wilk's test), we determined  $T_{\text{sel}}$  for each individual as (1) the mean of  $T_b$ 's selected every 20 min and (2) the lower and upper set-points calculated as the bounds of the central 50% of the observed  $T_b$ 's recorded every 20 min in the thermal gradient. We analyzed repeated measurements with PROC MIXED using a compound symmetry (CS) covariance structure (Littell et al. 1996) in SAS (v. 9.0, SAS Institute Inc., USA). The CS covariance structure was selected as it produced the lowest Akaike Information Criterion as compared with several other covariance structures available in SAS. Individual, day of experiments (field fresh: days 1–3, acclimated: days 10–12) and gender were treated as categorical variables, with time of day as a continuous variable. We did not correct for body mass, as preliminary analysis in each species revealed that it was not related to  $T_{\text{sel}}$ .

We calculated repeatability ( $\tau$ ) as the intra-class correlation coefficient, using variance components derived from one-way ANOVA and following Lessells and Boag (1987). In case our parametric estimates of  $\tau$  were biased, we also calculated repeatability from Kendall's test of concordance (i.e. non-parametric test). Within-day repeatability values were determined using  $T_b$ 's selected by lizards every 20 min in the thermal gradient from 1100 to 1800 h and were calculated for the 2nd day after field collection and the 2nd day after acclimation. Among-day repeatability values were calculated separately for (1) daily individual means of preferred temperature, (2) daily individual lower set-points and (3) daily individual upper set-points

determined from  $T_{bs}$  selected by lizards every 20 min in the thermal gradient from 1500 to 1800 h. This time interval was selected to allow maximum habituation in the gradient. We calculated 95% confidence limits (CLs) for  $\tau$  according to Krebs (1999). All analyses were tested for statistical significance at the  $P < 0.05$  level. Table-wide step-up false discovery rate correction was used to avoid Type I errors (Garcia 2004). All means are presented with their standard deviations unless otherwise stated.

## Results

We obtained data for 43 Cordylid lizards representing four species. Within each species, there was no effect of gender on  $T_{sel}$  ( $P > 0.45$  in all cases), and thus, sex was ignored for subsequent analyses. Mean  $T_{sel}$  ranged from 30.8 to 34.0°C among species and was fairly consistent across days within species (Table 2). There was no significant difference in body mass between lizards in the field-fresh and after-acclimation groups for three of the four species studied (Table 1). The small but significant mass gain in *C. cordylus* is unlikely to affect  $T_{sel}$  since lizards were post-absorptive during the acclimated trials.

Within-day repeatabilities ( $\tau$ ) of  $T_{sel}$  varied from 0.075 to 0.361 across species, and there was significant variation in  $T_{sel}$  among individuals (Table 3). Repeated measures ANOVA of  $T_{sel}$  from the 2nd day after field collection showed that for all species, time of day and time  $\times$  individual interactions significantly affected  $T_{sel}$  ( $P < 0.001$  and  $P < 0.05$ , respectively), indicating that individuals exhibited different patterns of variation throughout the day. In contrast, analyses of the 2nd day after acclimation revealed a time effect as well as a time  $\times$  individual effect for only two of the four species (*C. cordylus*: time:  $F_{1, 237} = 32.0$ ,  $P < 0.0001$ ; time  $\times$  individual:  $F_{11, 237} = 6.72$ ,  $P < 0.0001$ ; *C. polyzonus*: time:  $F_{1, 220} = 7.01$ ,  $P = 0.009$ ; time  $\times$  individual:  $F_{10, 220} = 2.20$ ,  $P = 0.018$ ).

Among-day repeatabilities for mean  $T_{sel}$  ranged from 0 to 0.261 across all species (Table 4), while those for lower and upper boundaries ranged from 0 to 0.163 and from 0 to 0.280, respectively (Table 5 and 6). For all three measures, there was rarely significant variation in  $T_{sel}$  among individuals (Tables 4–6). Repeatability did not differ before and after acclimation as revealed by the consistent overlap of upper and lower confidence limits (Tables 3–6). Repeated measures ANOVA revealed no effect of day (which would include an acclimation effect) on  $T_{sel}$  within any species, although several effects such as time, time  $\times$  day, time  $\times$  individual and time  $\times$  individual  $\times$  day were significant but not consistent across species (Table 7). To determine the possibility that low repeatability was a consequence of small sample size ( $N$ ), we modeled this effect using a resampling-with-replacement procedure in Microsoft Excel. The results show that an increase in  $N$  would result in a decrease in repeatability of  $T_{sel}$  (Fig. 1). In addition, repeatabilities ( $r$ ) calculated using Kendall's test of concordance (nonparametric analysis) were equally low and fairly consistent with the intraclass correlation coefficient estimates (Tables 3–6).

## Discussion

Within- and among-day repeatability values of  $T_{sel}$  were low because inter-individual differences represented a small fraction of total variance relative to the intra-individual

**Table 2** Summary statistics of preferred temperatures ( $T_{\text{sel}}$  in °C) collected from four species of Cordylid lizards in a photothermal gradient (17–55°C) from 1500 to 1800 h

	Day 1	Day 2	Day 3	Day 10	Day 11	Day 12
<i>C. cordylus</i>						
<i>N</i>	9	7	9	10	12	11
Sex ratio F:M	6:3	4:3	6:3	7:3	8:4	7:4
Mean	30.8 ± 3.0	32.8 ± 1.8	32.3 ± 1.6	32.1 ± 1.5	32.5 ± 1.7	32.1 ± 1.8
Median	30.7 ± 2.7	32.7 ± 1.8	32.4 ± 1.6	32.3 ± 1.7	32.5 ± 1.5	32.1 ± 2.0
Mean max	34.4 ± 2.0	35.3 ± 1.9	34.4 ± 1.1	35.8 ± 0.9	35.4 ± 1.9	34.3 ± 1.8
Absolute max	36.6	37.4	36.0	36.6	37.2	37.0
Mean min	26.2 ± 5.3	29.9 ± 2.3	29.9 ± 3.5	27.6 ± 3.5	29.4 ± 3.3	29.7 ± 2.2
Absolute min	15.8	25.9	23.8	20.9	23.5	24.0
Variance	9.0 ± 8.4	4.4 ± 5.6	3.4 ± 4.6	4.8 ± 6.0	4.2 ± 2.8	2.9 ± 2.4
Mean 25 percentile	29.1 ± 4.0	31.3 ± 2.8	31.2 ± 2.2	30.3 ± 2.0	31.6 ± 1.6	30.9 ± 2.2
Mean 75 percentile	33.1 ± 2.3	34.1 ± 1.8	33.5 ± 1.4	34.1 ± 1.0	33.7 ± 1.9	33.1 ± 1.8
<i>C. niger</i>						
<i>N</i>	10	10	9	10	10	10
Sex ratio F:M	4:6	4:6	4:5	4:6	4:6	4:5
Mean	32.3 ± 1.3	32.4 ± 0.7	32.4 ± 1.0	32.9 ± 0.6	32.8 ± 0.6	33.0 ± 1.0
Median	32.3 ± 1.5	32.6 ± 1.0	32.4 ± 1.0	32.9 ± 0.6	32.7 ± 0.7	33.1 ± 0.9
Mean max	34.2 ± 0.9	34.2 ± 0.6	33.9 ± 1.1	34.3 ± 0.7	34.2 ± 0.4	34.4 ± 1.0
Absolute max	35.7	34.9	36.0	35.2	34.8	36.0
Mean min	30.1 ± 1.9	29.7 ± 1.0	30.6 ± 1.8	31.3 ± 1.1	31.2 ± 0.9	31.3 ± 1.4
Absolute min	26.3	28.2	26.9	28.9	29.7	28.9
Variance	1.9 ± 1.8	2.6 ± 1.8	1.2 ± 0.7	1.0 ± 0.6	0.9 ± 0.5	1.2 ± 1.2
Mean 25 percentile	31.7 ± 1.5	31.6 ± 1.3	31.8 ± 1.0	32.4 ± 0.6	32.3 ± 0.6	32.3 ± 1.3
Mean 75 percentile	33.2 ± 0.9	33.3 ± 1.0	33.0 ± 0.8	33.6 ± 0.7	33.4 ± 0.6	33.8 ± 1.0
<i>C. polyzonus</i>						
<i>N</i>	11	11	11	11	11	11
Sex ratios F:M	7:4	7:4	7:4	7:4	7:4	7:4
Mean	33.8 ± 0.9	33.5 ± 0.9	33.1 ± 0.7	34.0 ± 0.6	33.7 ± 0.5	33.3 ± 0.5
Median	33.9 ± 0.7	33.5 ± 0.9	33.0 ± 0.7	34.1 ± 0.6	33.6 ± 0.5	33.2 ± 0.7
Mean max	35.3 ± 1.2	34.9 ± 1.1	35.0 ± 0.8	35.3 ± 0.7	35.0 ± 0.6	35.2 ± 0.8
Absolute max	37.2	36.5	36.3	36.7	36.1	36.5
Mean min	31.9 ± 1.5	32.0 ± 1.1	31.9 ± 1.1	32.5 ± 1.4	32.0 ± 0.9	31.7 ± 0.8
Absolute min	29.8	29.3	30.1	28.7	30.8	30.3
Variance	1.4 ± 0.9	1.0 ± 0.5	1.2 ± 0.8	0.9 ± 0.7	0.9 ± 0.6	1.3 ± 0.8
Mean 25 percentile	33.1 ± 1.0	32.8 ± 0.9	32.5 ± 1.0	33.5 ± 0.6	33.1 ± 0.7	32.7 ± 0.6
Mean 75 percentile	34.5 ± 0.8	34.2 ± 0.9	33.7 ± 0.8	34.6 ± 0.6	34.3 ± 0.6	33.9 ± 0.7
<i>C. oelofseni</i>						
<i>N</i>	10	9	10	10	9	8
Sex ratios F:M	5:5	5:4	5:5	5:5	4:5	4:4
Mean	33.4 ± 0.7	33.1 ± 1.3	33.8 ± 0.9	33.7 ± 1.3	33.5 ± 0.8	34.0 ± 1.1
Median	33.6 ± 0.9	33.3 ± 1.4	34.1 ± 1.0	34.0 ± 1.0	33.8 ± 0.8	34.3 ± 1.0
Mean max	35.3 ± 0.5	34.4 ± 1.3	35.5 ± 0.7	35.7 ± 1.0	35.5 ± 1.0	36.5 ± 0.9
Absolute max	36.1	35.6	36.5	36.7	36.8	37.5
Mean min	30.7 ± 2.2	31.2 ± 2.3	30.2 ± 3.5	30.7 ± 3.2	30.5 ± 2.3	30.8 ± 2.9
Absolute min	26.4	26.1	20.9	22.3	25.9	25.9
Variance	2.8 ± 2.4	1.3 ± 1.4	3.5 ± 5.1	4.1 ± 7.0	2.7 ± 1.8	3.3 ± 3.0
Mean 25 percentile	32.5 ± 1.3	32.7 ± 1.4	33.2 ± 1.0	32.8 ± 1.6	32.4 ± 0.8	33.2 ± 1.4
Mean 75 percentile	34.5 ± 0.8	33.7 ± 1.4	34.9 ± 0.9	34.9 ± 1.0	34.4 ± 1.0	35.0 ± 0.6

Statistics were calculated from 20 min data. *N* = number of individuals. Median and variance are means of individual values. Means ± standard deviation. Min = minimum, max = maximum. Lizards were acclimated to 20(± 3)°C between experiments

**Table 3** Within-day repeatability of preferred temperature ( $T_{sel}$ ) calculated in four species of Cordylid lizards

Species	$N$	$\tau$	(LCL; UCL)	ANOVA	$P$	$r$	$\chi^2$	$P$
<i>C. cordylus</i>								
Field-fresh	8	0.361	(0.171; 0.717)	$F_{7, 143} = 11.53$	0.000*	0.286	30.00	0.092
Acclimated	12	0.325	(0.175; 0.598)	$F_{11, 249} = 11.52$	0.000*	0.162	37.40	0.015
<i>C. niger</i>								
Field-fresh	10	0.149	(0.051; 0.413)	$F_{9, 196} = 4.67$	0.000*	0.177	33.50	0.041
Acclimated	10	0.163	(0.061; 0.433)	$F_{9, 210} = 5.26$	0.000*	0.078	16.46	0.743
<i>C. polyzonus</i>								
Field-fresh	11	0.196	(0.083; 0.463)	$F_{10, 220} = 6.24$	0.000*	0.281	59.04	0.000*
Acclimated	11	0.123	(0.041; 0.347)	$F_{10, 231} = 4.29$	0.000*	0.076	17.46	0.683
<i>C. oelofseni</i>								
Field-fresh	10	0.209	(0.087; 0.501)	$F_{9, 198} = 6.60$	0.000*	0.102	19.17	0.574
Acclimated	9	0.075	(0.010; 0.298)	$F_{8, 189} = 2.82$	0.006*	0.160	30.26	0.087

Repeatability (intra-class correlation coefficient ( $\tau$ ) and Kendall's concordance test ( $r$ )) were determined using body temperatures selected every 20 min in a thermal gradient (17–55°C) from 1100 to 1800 h. Estimates of repeatability were calculated for the 2nd day after collection in the field ("Field-fresh") and the 2nd day after 7 days of acclimation at 20°C ("Acclimated"). Lower and upper 95% confidence limits (LCL and UCL, respectively) calculated according to Krebs (1999).  $N$  = number of individuals. (\*) denotes significant difference after applying a step-up false discovery rate correction

component. These results indicate that individual differences in  $T_{sel}$  were not consistent over time. Surprisingly, repeatabilities were similarly low whether they were calculated from daily means or upper and lower set-points (Tables 4–6). If the dual set-

**Table 4** Among-day repeatability ( $\tau$ , intra-class correlation coefficient;  $r$ , Kendall's concordance test) of mean preferred temperature ( $T_{sel}$ ) calculated in four species of Cordylid lizards

Species	$N$	$\tau$	(LCL; UCL)	ANOVA	$P$	$r$	$\chi^2$	$P$
<i>C. cordylus</i>								
Field-fresh	11	0.000	(0.000; 0.533)	$F_{10, 13} = 0.97$	0.508	0.750	3.00	0.223
Acclimated	12	0.478	(0.101; 0.789)	$F_{11, 21} = 3.52$	0.006	0.037	0.67	0.717
6 days	12	0.261	(0.037; 0.600)	$F_{11, 46} = 2.71$	0.009	0.743	7.43	0.191
<i>C. niger</i>								
Field-fresh	10	0.160	(0.000; 0.625)	$F_{9, 18} = 1.52$	0.215	0.109	1.75	0.417
Acclimated	10	0.118	(0.000; 0.589)	$F_{9, 20} = 1.44$	0.236	0.030	0.60	0.741
6 days	10	0.000	(0.000; 0.177)	$F_{9, 48} = 0.64$	0.755	0.139	5.57	0.350
<i>C. polyzonus</i>								
Field-fresh	11	0.108	(0.000; 0.547)	$F_{10, 22} = 1.36$	0.262	0.174	3.82	0.148
Acclimated	11	0.318	(0.000; 0.704)	$F_{10, 22} = 2.37$	0.044	0.529	11.64	0.003*
6 days	11	0.075	(0.000; 0.387)	$F_{10, 55} = 1.50$	0.168	0.272	14.95	0.011
<i>C. oelofseni</i>								
Field-fresh	10	0.327	(0.000; 0.730)	$F_{9, 19} = 2.42$	0.050	0.049	0.89	0.641
Acclimated	9	0.219	(0.000; 0.687)	$F_{8, 17} = 1.80$	0.147	0.063	1.00	0.607
6 days	10	0.055	(0.000; 0.393)	$F_{9, 46} = 1.32$	0.252	0.153	5.37	0.373

Repeatability was determined using daily means of individual  $T_{sel}$  obtained from 20 min data collected in the thermal gradient (17–55°C) from 1500 to 1800 h. After capture,  $T_{sel}$  was determined for 3 consecutive days ("Field-fresh"). After an acclimation at 20°C for 7 days,  $T_{sel}$  was recorded for 3 more days ("Acclimated"). Six days = 3 days at "Field-fresh" and 3 days at "Acclimated" state. (\*) denotes significant difference after applying a step-up false discovery rate correction



**Table 5** Among-day repeatability ( $\tau$ , intra-class correlation coefficient;  $r$ , Kendall's concordance test) of lower set-point temperature calculated for four species of Cordylid lizards

Species	<i>N</i>	$\tau$	(LCL; UCL)	ANOVA	<i>P</i>	<i>r</i>	$\chi^2$	<i>P</i>
<i>C. cordylus</i>								
Field-fresh	11	0.000	(0.000; 0.452)	$F_{10, 13} = 0.78$	0.648	0.250	1.00	0.607
Acclimated	12	0.301	(0.000; 0.687)	$F_{11, 21} = 2.18$	0.060	0.086	1.56	0.459
6 days	12	0.132	(0.000; 0.469)	$F_{11, 46} = 1.73$	0.096	0.629	6.29	0.279
<i>C. niger</i>								
Field-fresh	10	0.295	(0.000; 0.716)	$F_{9, 18} = 2.18$	0.076	0.141	2.25	0.325
Acclimated	10	0.054	(0.000; 0.523)	$F_{9, 20} = 1.16$	0.369	0.030	0.60	0.741
6 days	10	0.008	(0.000; 0.314)	$F_{9, 48} = 1.05$	0.414	0.114	4.57	0.470
<i>C. polyzonus</i>								
Field-fresh	11	0.146	(0.000; 0.579)	$F_{10, 22} = 1.51$	0.200	0.058	1.27	0.529
Acclimated	11	0.333	(0.000; 0.714)	$F_{10, 22} = 2.48$	0.036	0.554	12.18	0.002*
6 days	11	0.163	(0.000; 0.405)	$F_{10, 55} = 1.59$	0.133	0.242	13.29	0.021
<i>C. oelofseni</i>								
Field-fresh	10	0.442	(0.047; 0.793)	$F_{9, 19} = 3.30$	0.014	0.198	3.56	0.169
Acclimated	9	0.269	(0.000; 0.718)	$F_{8, 17} = 2.06$	0.100	0.187	3.00	0.223
6 days	10	0.065	(0.000; 0.407)	$F_{9, 46} = 1.38$	0.223	0.160	5.61	0.346

Daily lower boundary (25 percentile) of preferred temperature ( $T_{sel}$ ) was determined per individual by using 20 min data collected in the thermal gradient (17–55°C) from 1500 to 1800 h. (\*) denotes significant difference after applying a step-up false discovery rate correction

**Table 6** Among-day repeatability ( $\tau$ , intra-class correlation coefficient;  $r$ , Kendall's concordance test) of upper set-point temperature calculated for four species of Cordylid lizards

Species	<i>N</i>	$\tau$	(LCL; UCL)	ANOVA	<i>P</i>	<i>r</i>	$\chi^2$	<i>P</i>
<i>C. cordylus</i>								
Field-fresh	11	0.000	(0.000; 0.437)	$F_{10, 13} = 0.75$	0.670	0.750	3.00	0.223
Acclimated	12	0.482	(0.106; 0.791)	$F_{11, 21} = 3.56$	0.006*	0.049	0.89	0.641
6 days	12	0.280	(0.051; 0.616)	$F_{11, 46} = 2.88$	0.006*	0.688	6.88	0.229
<i>C. niger</i>								
Field-fresh	10	0.042	(0.000; 0.530)	$F_{9, 18} = 1.12$	0.396	0.141	2.25	0.325
Acclimated	10	0.167	(0.000; 0.619)	$F_{9, 20} = 1.62$	0.176	0.310	6.20	0.045
6 days	10	0.022	(0.000; 0.336)	$F_{9, 48} = 1.12$	0.368	0.180	7.21	0.205
<i>C. polyzonus</i>								
Field-fresh	11	0.000	(0.000; 0.308)	$F_{10, 22} = 0.69$	0.722	0.174	3.82	0.148
Acclimated	11	0.372	(0.009; 0.737)	$F_{10, 22} = 2.78$	0.022	0.430	9.45	0.009
6 days	11	0.070	(0.000; 0.378)	$F_{10, 55} = 1.45$	0.183	0.260	14.32	0.014
<i>C. oelofseni</i>								
Field-fresh	10	0.013	(0.000; 0.520)	$F_{9, 19} = 1.04$	0.440	0.037	0.67	0.717
Acclimated	9	0.257	(0.000; 0.711)	$F_{8, 17} = 1.99$	0.112	0.149	2.39	0.303
6 days	10	0.020	(0.000; 0.340)	$F_{9, 46} = 1.11$	0.374	0.217	7.58	0.181

\* $P < 0.05$

Daily upper boundary (75 percentile) of preferred temperature ( $T_{sel}$ ) was determined per individual by using 20 min data collected in the thermal gradient (17–55°C) from 1500 to 1800 h. (\*) denotes significant difference after applying a step-up false discovery rate correction

point model (Barber and Crawford, 1977) holds for Cordylid lizards, one may expect higher repeatability in upper and lower set-point temperatures than in mean  $T_{sel}$ , especially if these set-points are several degrees apart. Moreover, repeatability

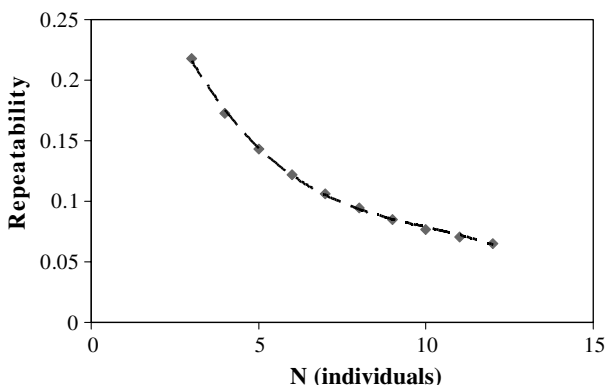
**Table 7** Results of repeated measures PROC MIXED model using 20 min data on preferred temperature ( $T_{sel}$ ) from 1500 to 1800 h

Source	df	<i>F</i>	<i>P</i>	df	<i>F</i>	<i>P</i>
	<i>C. cordylus</i>			<i>C. niger</i>		
Ind	11, 41	2.33	0.025	9, 43	0.69	0.715
Time	1, 464	3.89	0.049	1, 464	5.79	0.016*
Day	5, 41	1.24	0.309	5, 43	1.09	0.379
Time × Day	5, 464	1.48	0.195	5, 464	0.50	0.776
Time × Ind	11, 464	3.73	<0.001*	9, 464	1.40	0.184
Time × Ind × Day	41, 464	1.87	0.001*	43, 464	2.30	<0.001*
	<i>C. polyzonus</i>			<i>C. oelofseni</i>		
Ind	10, 50	1.13	0.357	9, 41	0.91	0.528
Time	1, 528	2.76	0.097	1, 448	6.95	0.009*
Day	5, 50	2.33	0.056	5, 41	1.42	0.238
Time × Day	5, 528	3.35	0.005*	5, 448	3.57	0.004*
Time × Ind	10, 528	1.77	0.063	9, 448	1.52	0.137
Time × Ind × Day	50, 528	1.81	0.001*	41, 448	0.98	0.512

\* $P < 0.05$ 

Ind = individuals; Time = time of day, Day = 6 days of  $T_{sel}$  (3 days field-fresh, 3 days in acclimated state after 1 week of acclimation at 20°C). A lack of a Day effect indicates that acclimation did not alter  $T_{sel}$ . (\*) denotes significant difference after applying a step-up false discovery rate correction

remained low irrespective of the time scale investigated (Table 3 versus 4–6), although at the within-day level significant among-individual variation occurred ( $P < 0.006$  for all cases and species, Table 3). Assuming that acclimation reduces the among-individual variation attributed to acclimatization, one would expect the repeatability of  $T_{sel}$  in acclimated lizards to exceed that of field-fresh lizards. By contrast, acclimation seldom improved repeatability of  $T_{sel}$  (CLs overlapped, Tables 3–6). Furthermore, it is unlikely that the low repeatability was a consequence of small sample sizes. The model (Fig. 1) suggests that even when sample sizes are low, total among-individual variation has already been accurately represented in the populations investigated. In addition,



**Fig. 1** A resampling-with-replacement model showing how mean population  $T_{sel}$  repeatability (intra-class correlation coefficient) is affected by an increase in sample size (individuals). Model training data were taken from recorded *C. cordylus*  $T_{sel}$  values (in °C). Note that standard deviations of the model were smaller than the symbols depicted. A similar pattern was observed when the model was run with increasing number of measurements per individual, i.e. repeatability decreased in a non-linear manner (data not shown). This model indicates that increasing the sample size would result in a decline in repeatability

our study provides four assessments of repeatability (i.e. four species) and it is highly unlikely that on all four occasions we failed to capture representative values of population variation.

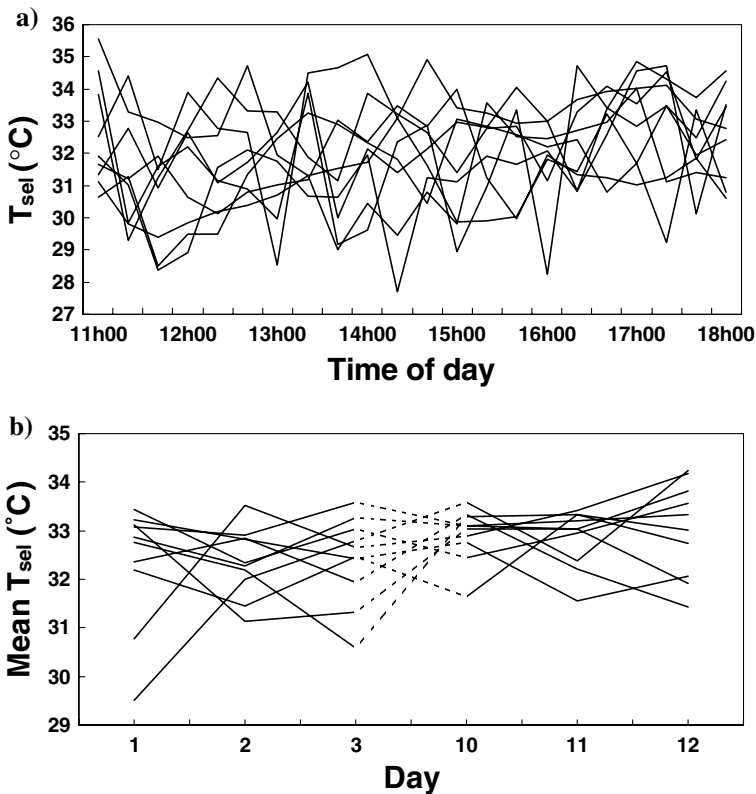
It is difficult to compare our values of repeatability for  $T_{sel}$  with those from other studies because, to our knowledge, only one study has calculated repeatability of  $T_{sel}$  in a lizard species previously. Le Galliard et al. (2003) found higher repeatability ( $\tau = 0.66$ ) for  $T_{sel}$  in female *Lacerta vivipara* between two successive days. Dohm et al. (2001) also found higher repeatability ( $\tau = 0.66$ ) of  $T_{sel}$  in marine toads (*Bufo marinus*) across five trial days, which included pre- and post-exposure to ozone. In juvenile garter snakes, Arnold et al. (1995) found a significant repeatability of 0.47 for  $T_{sel}$  recorded twice a day during five consecutive days. Although there is little information on repeatability of  $T_{sel}$  in lizards, more information is available on repeatability of other traits, such as locomotor performance. Repeatability of sprint speed and endurance varies considerably depending on the species and time scale investigated (i.e. days to years), and ranges from 0.02 to 0.97 and 0.25 to 0.80, respectively (Garland 1985; Huey and Dunham 1987; Tsuji et al. 1989; van Berkum et al. 1989; Garland et al. 1990; Huey et al. 1990; Angilletta et al. 2002b). These values show that the repeatability of  $T_{sel}$  determined in our study was low compared with that of other species for which repeatability was estimated or compared to other performance measures. However, these comparisons should be made with caution as several studies used the Pearson product-moment correlation or Spearman rank correlation (e.g. van Berkum et al. 1989; Angilletta et al. 2002b) instead of the intra-class correlation coefficient. It is important to note that the correlation measures generally indicate the degree of association of a trait between two time-points whereas the intra-class correlation coefficient describes the proportion of variance in a trait that occurs among- versus within-individuals over repeated measurements of the trait. Therefore, the intra-class correlation coefficient likely produces a more accurate representation of the variation that natural selection may operate on. However, intra-class coefficient analyses are only meaningful when there are no systematic trends in a particular direction for the trait of interest, but temporal effects found in this study were generally not systematic among individuals.

There are three main reasons that could explain the relatively low repeatability of  $T_{sel}$  that we report here. First, the protocol used to define the trait might be flawed (Hayes et al. 1998). However, there is little evidence that this may be the case. Photothermal gradients have been used extensively in other studies (e.g. Castilla and Bauwens 1991; Le Galliard et al. 2003; Zhang and Ji 2004), some showing that  $T_{sel}$  corresponded with  $T_{opt}$  for locomotor performance (e.g. Bauwens et al. 1995) and digestion (Angilletta 2001). Also, we habituated all lizards to the gradient prior to the estimation of  $T_{sel}$ , experimenters were not present in the climate chamber during trials to reduce possible stress effects and, multiple measurements were obtained to account and test for temporal effects. Regardless, the possibility that a thermal gradient may represent an artificial environment that lowers repeatability of  $T_{sel}$  cannot be discarded (Christian and Weavers 1996). Behavioral thermoregulation is an effective response to spatial and temporal heterogeneity of the thermal environment, but a thermal gradient only simulates the spatial component of this heterogeneity. Thus, if the cue for thermoregulation is largely daily temporal patterns, a thermal gradient may not provide the appropriate cue for active thermoregulation. Indeed, relatively little is known about temperature-sensing

mechanisms in these and other lizard species (Seebacher and Franklin 2005, but see Patapoutian et al. 2003).

Second, low repeatability over increasing time scales may indicate that a trait needs to be redefined, perhaps as a set of age- or environment-specific traits (Hayes et al. 1998). It is not likely that such changes influenced  $T_{sel}$  in our study, as all our experimental animals were adults. Moreover, since there was no tendency for repeatability to improve with acclimation to constant conditions (Day effect, Table 7), it is unlikely that acclimatization (seasonal effects) experienced before all trials (i.e. phenotypic plasticity) altered the results of our study significantly. A longer acclimation period would probably not result in further variation of  $T_{sel}$  since even after 5 weeks of acclimation, Wheeler (1986) observed no change in  $T_{sel}$  of *Cordylus jonesi*. However, extensive investigations of the time-course of thermal acclimation in  $T_{sel}$  would be required to discount this possibility completely. In addition, the lack of a consistent  $T_{sel}$  change between days decreases the possibility of stress effects.

Third, low repeatability might reflect real, random, biological variation (i.e. biological noise; Hayes et al. 1998). Our results tend to support this option. The high



**Fig. 2** Temporal variation of preferred temperature ( $T_{sel}$ ) obtained for 10 individuals of *C. niger* in a thermal gradient, (a) within day:  $T_{sel}$  selected every 20 min is plotted from 1100 to 1800 h on the 2nd day after field collection, (b) among days: daily individual means are plotted for 6 days (Day 1–3 at “Field fresh” and Day 10–12 at “Acclimated” state). Broken lines depict the acclimation period

number of significant interactions between time and individual or day, found in both the within- and among-day analyses (Fig. 2), indicates the random nature of temporal effects (i.e. lack of consistent patterns within or between species) and supports the unpredictability of  $T_{\text{sel}}$  at the time scales investigated. The causes underlying such temporal effects are not clear. Cordylid species have generally low movement patterns and stay close to crevices (Cooper et al. 1997), therefore,  $T_{\text{sel}}$  may not be reflective of locomotor performance but instead may be reflective of other physiological mechanisms that may only be essential at specific times of the year (e.g. winter thermoregulation, mating season). Also,  $T_{\text{sel}}$  in nature may reflect simultaneous physiological processes (i.e. reproduction, digestion, sprint speed) requiring a wide performance breadth or a complex trade-off among performances with different thermal optima, which may weaken selection on  $T_{\text{sel}}$  (Gilchrist, 1995; Angilletta et al. 2002b; 2006). Studies of locomotor performance and digestion (e.g. assimilation) may reveal which mechanisms underlie  $T_{\text{sel}}$  in these species. In Cordylidae, sprinting  $T_{\text{opt}}$  in *Platysaurus intermedius* averaged 31.9°C, which is remarkably similar to the mean  $T_{\text{sel}}$  of this species (31.2°C; Lailvaux et al. 2003) and to  $T_{\text{sel}}$  values found in our study.

Lizards exposed to thermal gradients in the laboratory are free to move through an array of temperature options without any immediate physiological challenge that may be related to selection. Under such circumstances, there may be less need for careful regulation, hence low repeatability. This argument is supported by the following points. First, repeatability of physiological measures increases (i.e. intra-individual variability decreases) when animals work near their maximum physiological capacity (Berteaux et al. 1996). Second, selection for thermoregulation may be low depending on the thermal heterogeneity of the habitat and the temporal scale investigated (Angilletta et al. 2006). Interestingly,  $T_{\text{sel}}$  values for lizards in this study did not differ markedly with those of other species of Cordylids (Table 8) for which  $T_{\text{sel}}$  are available (maximum difference among species: 2.6°C). This suggests that  $T_{\text{sel}}$  is likely conserved, and may not reflect an adaptation to current thermal environments of *Cordylus* spp. (Angilletta et al. 2002b). Instead,  $T_{\text{sel}}$  may reflect what Huey et al. (2003) recently named “the Bogert effect”, or behavioral inertia, such that lizard’s thermoregulatory behavior may inhibit selection for evolutionary shifts in thermal physiology. However, without further investigation of spatial and temporal variation in body and operative temperatures in the field, as well as the breadth of different physiological performances and their temperature sensitivities, we are currently unable to distinguish among these hypotheses.

If no repeatable measure of  $T_{\text{sel}}$  can be found, the evolution of lizard thermal preference might not be enlightened by studies of inter- and intra-individual

**Table 8** Preferred body temperatures ( $T_{\text{sel}}$ ) available for species belonging to the family Cordylidae

Species	Mean $T_{\text{sel}}$ (°C)	Source
<i>C. cordylus</i>	32.1 ± 0.7	This study
<i>C. niger</i>	32.6 ± 0.3	This study
<i>C. oelofseni</i>	33.6 ± 0.3	This study
<i>C. polyzonus</i>	33.6 ± 0.3	This study
<i>C. vittifer</i>	32.1 ± 1.8	Skinner (1991)
<i>C. jonesi</i>	33.5 ± 0.3	Wheeler (1986)
<i>Platysaurus intermedius</i> w.	31.5 ± 1.7	Lailvaux et al. (2003)

Values from this study are the grand means of daily mean  $T_{\text{sel}}$  from Table 1

variation. However, there are clear evolutionary benefits associated with consistent between-individual variation of  $T_{\text{sel}}$ . Low repeatabilities of this trait reduce, but do not eliminate, the possibility of a significant relationship between  $T_{\text{sel}}$  and other variables indicative of fitness (e.g. locomotor performance), and our results may simply reflect a lack of statistical power. However, we are of the opinion that this is not the case since similar patterns were documented in all four species under controlled conditions. The varied temporal effects reported in this study highlight the need for assessment of  $T_{\text{sel}}$  over different time scales, both in the field and in the laboratory, before attempting to link performance and fitness in Cordylids. By determining the magnitude of intra-individual variation of  $T_{\text{sel}}$ , this study highlights the importance of multiple measurements (see e.g. Hayes and Jenkins 1997; McNab 2003). The comparison of  $T_{\text{sel}}$  repeatabilities between species with different lifestyles (e.g. sit-and-wait versus active forager, diurnal versus nocturnal) as well as between diverse geographic and climatic conditions may help elucidate predominantly low repeatabilities of  $T_{\text{sel}}$  for Cordylids. Most importantly, unless repeatability of  $T_{\text{sel}}$  or the time scale at which selection may operate on this trait are determined, caution is required when interpreting the adaptive significance of this trait.

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## References

- Angilletta MJ (2001) Thermal and physiological constraints on energy assimilation in a widespread lizard (*Sceloporus undulatus*). *Ecology* 82:3044–3056
- Angilletta MJ, Werner YL (1998) Australian geckos do not display diel variation in thermoregulatory behavior. *Copeia* 1998:736–742
- Angilletta MJ, Montgomery LG, Werner YL (1999) Temperature preference in geckos: Diel variation in juveniles and adults. *Herpetologica* 55:212–222
- Angilletta MJ, Hill T, Robson MA (2002a) Is physiological performance optimized by behavioral thermoregulation? A case study of the eastern fence lizard, *Sceloporus undulatus*. *J Therm Biol* 27:199–204
- Angilletta MJ, Niewiarowski PH, Navas CA (2002b) The evolution of thermal physiology in ectotherms. *J Therm Biol* 27:249–268
- Angilletta MJ, Bennett AF, Guderley H, Navas CA, Seebacher F, Wilson RS (2006) Coadaptation: a unifying principle in evolutionary thermal biology. *Physiol Biochem Zool* 79:282–294
- Arnold SJ, Peterson CR, Gladstone J (1995) Behavioral variation in natural populations. VII Maternal body temperature does not affect juvenile thermoregulation in a garter snake. *Anim Behav* 50:623–633
- Barber BJ, Crawford EC Jr (1977) A stochastic dual-limit hypothesis for behavioral thermoregulation in lizards. *Physiol Zool* 50:53–60
- Bauwens D, Garland T Jr, Castilla A, Van Damme R (1995) Evolution of sprint speed in lacertid lizards: morphological, physiological and behavioral covariation. *Evolution* 49:848–863
- Beitinger TL, Fitzpatrick LC (1979) Physiological and ecological correlates of preferred temperature in fish. *Am Zool* 19:319–329

- Bennett AF (1987) Inter-individual variability: An underutilized resource. In: Feder ME, Bennett AF, Burggren WW, Huey RB (eds) New directions in ecological physiology. Cambridge University Press, Cambridge, pp 1–8
- Bennett AF (1997) Adaptation and the evolution of physiological characters. In: WH Danzler (eds) Handbook of physiology, Sect. 13: comparative physiology, Vol 1. Oxford University Press, New York, pp 3–16
- Bennett AF, John-Alder H (1986) Thermal relations of some Australian skinks (Sauria: Scincidae). *Copeia* 1986:57–64
- Berk ML, Heath JE (1975) An analysis of behavioral thermoregulation in the lizard *Dipsosaurus dorsalis*. *J Therm Biol* 1:15–22
- Berteaux D, Thomas DW, Bergeron J-M, Lapierre H (1996) Repeatability of daily field metabolic rate in female Meadow Voles (*Microtus pennsylvanicus*). *Funct Ecol* 10:751–759
- Boake CRB (1989) Repeatability: its role in evolutionary studies of mating behavior. *Evol Ecol* 3:173–182
- Brown RP, Griffin S (2005) Lower selected body temperatures after food deprivation in the lizard *Anolis carolinensis*. *J Therm Biol* 30:79–83
- Castilla AM, Bauwens D (1991) Thermal biology, microhabitat selection, and conservation of the insular lizard *Podarcis hispanica atrata*. *Oecologia* 85:366–374
- Christian KA, Bedford GS (1995) Seasonal changes in thermoregulation by the frillneck lizard, *Chlamydosaurus kingii*, in tropical Australia. *Ecology* 76:124–132
- Christian KA, Weavers BW (1996) Thermoregulation of monitor lizards in Australia: an evaluation of methods in thermal biology. *Ecol Mon* 66:139–157
- Christian KA, Tracy CR, Porter WP (1985) Inter- and intra-individual variation in body temperature of the Galapagos land iguana. *J Therm Biol* 10:47–50
- Cooper WE, Whiting MJ, Van Wyk JH (1997) Foraging modes of cordyliform lizards. *S Afr J Zool* 32:9–13
- Dawson WR (1975) On the physiological significance of the preferred body temperatures of reptiles. In: Gates DM, Schmerl RB (eds) Perspectives of biophysical ecology: ecological studies. Vol 12. Springer-Verlag, New York, pp 433–473
- Dohm MR (2002) Repeatability estimates do not always set an upper limit to heritability. *Funct Ecol* 16:273–280
- Dohm MR, Mautz WJ, Looby PG, Gellert KS, Andrade JA (2001) Effects of ozone on evaporative water loss and thermoregulatory behavior of marine toads (*Bufo marinus*). *Env Res* 86:274–286
- Du W-G, Yan S-J, Ji X (2000) Selected body temperature, thermal tolerance and thermal dependence of food assimilation and locomotor performance in adult blue-tailed skinks, *Eumeces elegans*. *J Therm Biol* 25:197–202
- Endler JA (1986) Natural selection in the wild. Princeton University Press, Princeton
- Falconer DS, Mackay TFC (1996) Introduction to quantitative genetics. Prentice Hall, Harlow
- Firth BT, Belan I (1998) Daily and seasonal rhythms in selected body temperatures in the Australian lizard *Tiliqua rugosa* (Scincidae): field and laboratory observations. *Physiol Zool* 71:303–311
- Gaines SD, Denny MW (1993) The largest, smallest, highest, lowest, longest, and shortest: extremes in ecology. *Ecology* 74:1677–1692
- Garcia LV (2004) Escaping the Bonferroni iron claw in ecological studies. *Oikos* 105:657–663
- Garland T Jr (1985) Ontogenetic and individual variation in size, shape, and speed in the Australian agamid lizard *Amphibolurus nuchalis*. *J Zool* 207:425–439
- Garland T Jr, Hankins E, Huey RB (1990) Locomotor performance and social dominance in male lizards. *Funct Ecol* 4:243–250
- Gilchrist GW (1995) Specialists and generalists in changing environments. I Fitness landscapes of thermal sensitivity. *Am Nat* 146:252–270
- Good DS (1993) Evolution of behaviours in *Drosophila melanogaster* in high temperatures: genetic and environmental effects. *J Insect Physiol* 39:537–544
- Hayes JP, Jenkins SH (1997) Individual variation in mammals. *J Mamm* 78:274–293
- Hayes JP, Bible CA, Boone JD (1998) Repeatability of mammalian physiology: evaporative water loss and oxygen consumption of *Dipodomys merriami*. *J Mamm* 79:475–485
- Helmuth B, Kingsolver JG, Carrington E (2005) Biophysics, physiological ecology and climate change: does mechanism matter?. *Ann Rev Physiol* 67:177–201
- Hertz PE, Huey RB, Stevenson RD (1993) Evaluating temperature regulation by field-active ectotherms: the fallacy of the inappropriate question. *Am Nat* 142:796–818
- Hoffmann AA, Sørensen JG, Loeschcke V (2003) Adaptation of *Drosophila* to temperature extremes: bringing together quantitative and molecular approaches. *J Therm Biol* 28:175–216

- Huey RB (1982) Temperature, physiology and the ecology of reptiles. In: Gans C, Pough FH (eds) *Biology of the Reptilia*. Vol 12. Academic Press, London, pp 25–91
- Huey RB, Bennett AF (1987) Phylogenetic studies of coadaptation: preferred temperature versus optimal performance temperatures of lizards. *Evolution* 41:1098–1115
- Huey RB, Dunham AT (1987). Repeatability of locomotor performance in natural populations of the lizard *Sceloporus merriami*. *Evolution* 41:1116–1120
- Huey RB, Kingsolver JG (1989) Evolution of thermal sensitivity of ectotherm performance. *Trends Ecol Evol* 4:131–135
- Huey RB, Slatkin M (1976) Cost and benefits of lizard thermoregulation. *Quart Rev Biol* 51:363–384
- Huey RB, Webster TP (1976) Thermal biology of *Anolis* lizards in a complex fauna: the *crustellus* group on Puerto Rico. *Ecology* 57:985–994
- Huey RB, Hertz PE, Sinervo B (2003) Behavioral drive versus behavioral inertia in evolution: a null model approach. *Am Nat* 161:357–366
- Huey RB, Dunham AE, Overall KL, Newman RA (1990) Variation in locomotor performance in demographically known populations of the lizard *Sceloporus merriami*. *Physiol Zool* 63:845–872
- Krebs CJ (1999) *Ecological methodology*. Addison-Wesley Educational Publishers, California
- Lailvaux SP, Alexander GJ, Whiting MJ (2003) Sex-based differences and similarities in locomotor performance, thermal preferences, and escape behavior in the lizard *Platysaurus intermedius wilhelmi*. *Physiol Biochem Zool* 76:511–521
- Le Galliard J-F, Le Bris M, Clobert J (2003) Timing of locomotor impairment and shift in thermal preferences during gravidity in a viviparous lizard. *Funct Ecol* 17:877–885
- Lessells CM, Boag PT (1987) Unrepeatable repeatabilities: a common mistake. *Auk* 104:116–121
- Licht P, Dawson WR, Shoemaker VH, Main AR (1966) Observations on the thermal relations of western Australian lizards. *Copeia* 1966:97–110
- Littell RC, Milliken GA, Stroup WW, Wolfinger RD (1996) *SAS systems for mixed models*. SAS Institute, Cary, North Carolina
- McNab BK (2003) Sample size and the estimation of physiological parameters in the field. *Funct Ecol* 17:82–86
- Patapoutian A, Peier AM, Story GM, Viswanath V (2003) ThermoTRP channels and beyond: mechanisms of temperature sensation. *Nature Rev Neurosci* 4:529–539
- Patterson JW, Davies PMC (1978) Preferred body temperature: seasonal and sexual differences in the lizard *Lacerta vivipara*. *J Therm Biol* 3:39–41
- Peterson CR, Arnold SJ (1986) Individual variation in the thermoregulatory behavior of free-ranging garter snakes, *Thamnophis elegans*. *Am Zool* 26:112
- Rock J, Andrews RM, Cree A (2000) Effects of reproductive condition, season and site on selected temperatures of a viviparous gecko. *Physiol Biochem Zool* 73:344–355
- Seebacher F (2005) A review of thermoregulation and physiological performance in reptiles: what is the role of phenotypic flexibility?. *J Comp Physiol B* 175:453–461
- Seebacher F, Franklin CE (2005) Physiological mechanisms of thermoregulation in reptiles: a review. *J Comp Physiol B* 175:533–541
- Skinner DC (1991) Effect of intraperitoneal melatonin injections on thermoregulation in the Transvaal girdled lizard, *Cordylus vittifer*. *J Therm Biol* 16:179–184
- Sievert LM, Hutchison VH (1989) Influences of season, time of day, light and sex on the thermoregulatory behavior of *Crotaphytus collaris*. *J Therm Biol* 14:159–165
- Sinervo B (1990) Evolution of thermal physiology and growth rate between populations of the western fence lizard (*Sceloporus occidentalis*). *Oecologia* 83:228–237
- Stevenson RD, Peterson CR, Tsuji JS (1985) The thermal dependence of locomotion, tongue flicking, digestion, and oxygen consumption in the wandering garter snake. *Physiol Zool* 58:46–57
- Tsuji JS, Huey RB, van Berkum FH, Garland T Jr, Shaw RG (1989) Locomotor performance of hatchling fence lizards (*Sceloporus occidentalis*): quantitative genetics and morphometric correlates. *Evol Ecol* 3:240–252
- van Berkum FH, Huey RB, Tsuji JS, Garland T Jr (1989) Repeatability of individual differences in locomotor performance and body size during early ontogeny of the lizard *Sceloporus occidentalis*. *Funct Ecol* 3:97–105
- Van Damme R, Bauwens D, Verheyen RF (1990) Evolutionary rigidity of thermal physiology: the case of the cool temperate lizard *Lacerta vivipara*. *Oikos* 57:61–67
- Wheeler PE (1986) Thermal acclimation of metabolism and preferred body temperature in lizards. *J Therm Biol* 11:161–166



- Wilhoft DC, Anderson JD (1960) Effect of acclimation on the preferred body temperature of the lizard, *Sceloporus occidentalis*. *Science* 131:610–611
- Wills CA, Beaupre SJ (2000) An application of randomization for detecting evidence of thermoregulation in timber rattlesnakes (*Crotalus horridus*) from Northwest Arkansas. *Physiol Biochem Zool* 73:325–334
- Zhang Y-P, Ji X (2004) The thermal dependence of food assimilation and locomotor performance in southern grass lizards, *Takydromus sexlineatus* (Lacertidae). *J Therm Biol* 29:45–53