# Individual variation in thermal plasticity and its impact on mass-scaling

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# Abstract

1. Physiological processes of individuals can be highly variable and there is accumulating evidence that individuals can differ in how they respond to environmental change. The ability for individuals to reversibly adjust their metabolic rate in response to temperature (i.e., metabolic thermal plasticity) may affect mass-scaling at the population level. This process has rarely been investigated before.
2. This study characterised the repeatability of metabolic thermal plasticity and tested how mass-scaling exponents change at different temperatures in the delicate skink (*Lampropholis delicata*). We repeatedly measured standard metabolic rate of forty-two individuals at six temperatures over the course of three months (*N*[observation] = 5040). We explicitly accounted for multi-level variation in our data in order to quantify more precise estimates of mass-scaling exponents at different environmental temperatures.
3. We found that individual differences in metabolic thermal plasticity were consistent over time. Repeatability of average standard metabolic rate increased as a function of temperature, which was associated with a decrease in residual variance, suggesting that individuals may be responding more predictably at higher temperatures. Overall, cross-temperature correlations were positive but were not uniform across the reaction norm.
4. After taking into account within- and among-individual level variation in our data, our estimates for mass-scaling did not change with temperature and were in line with published values for snakes and lizards. This implies that repeatable plastic responses may contribute to thermal stability of scaling exponents.
5. Our work contributes to our understanding of whether phenotypic plasticity has the capacity to respond to selection which is important for animals coping with rapid environmental change. Considering multi-level variation in body mass and metabolic rate is not only important for comparative studies interested in mass-scaling across the animal kingdom, but also to theoretical research interested using the predictive power of mass-scaling.

# Keywords

Phenotypic plasticity, reaction norm, thermal sensitivity, repeatability, thermal performance curves

# Introduction

All biological processes hinge on the availability of energy (Allen, Gillooly & Brown 2005). Metabolic rate (MR) governs how much energy is available to be allocated to competing processes such as growth, reproduction and maintenance (De Jong & Van Noordwijk 1992; Brown *et al.* 2004; Biro & Stamps 2008). MR is thought to be critical to fitness due to its functional link to morphology, behaviour and life-history which promotes among these traits (Réale *et al.* 2010; Friesen, Johansson & Olsson 2017; Malishev, Bull & Kearney 2017; Biro & Stamps 2010). For example, short-lived ecotypic garter snakes tend to have much higher mass-specific metabolic rates, larger body sizes, faster growth rates and invests more heavily into reproduction compared their long lived ecotypic counterparts {Bronikowski:2010ir}. The integration of these traits may be due to the close association between body mass and metabolic rate. Body mass and metabolic rate typically show a power relationship with an scaling exponent ranging from 0.64 to 0.88 {White:2006fw}. Scaling exponents less than one mean that energy expenditure scales disproportionately with mass, such that small organisms tend to have a much energy expenditure after controlling for body mass. Metabolic scaling exponents are incredibly heterogenous among {White:2006fw}(Uyeda *et al.* 2017) and within taxa (Burton *et al.* 2011; Norin & Gamperl 2018), yet the drivers of variation in mass-scaling relationships not well understood.

Metabolic theories use the predictive power of mass-scaling to explain ecological processes across levels of biological organisation. These studies assume energy consumption is the same across individuals despite metabolic rate varying several-fold among individuals of the same body mass (reviewed in {Biro:2010ee, Burton:2011fe}. For instance, individuals can differ in relative organ masses or body composition yielding different energetic demands {Steyermark:2005bxa, Scott:1996en}. Additionally, variation in mitochondrial efficiency results in different SMR in fish{Salin:2016fq} despite mass remaining the same. Ignoring individual physiological processes may be problematic for comparative studies because individual variation can be erroneously absorbed into higher hierarchical levels {vandePol:2009em}. This may bias estimates, increasing variation in mass-scaling exponents across studies. Moreover, mass-scaling exponents may be susceptible to sampling variability because metabolic rate and body mass tend to be measured once per individual and averaged across a population. Understanding the consistency of metabolism at the individual level may help explain why mass-scaling exponents are so heterogenous in the literature.

Temperature fluctuates extensively within the lifetime of organisms and has an overarching effect on metabolic rate. Numerous studies have found that scaling exponents show temperature dependence in a multitude of ways (reviewed in {Glazier:2005ei}). For example, mass-scaling exponents increased with temperature in teleost fish, (refs{Killen:2010cw}), but decreased with temperature in crustaceans {Ivleva:1980cf}. Individuals can vary in their metabolic thermal plasticity, that is, their capacity to adjust their metabolic rate in response to temperature {Nussey:2007bz}. This can be important for understanding temperature dependence of mass-scaling and understanding both how selection might shape plastic responses but this has rarely been considered {Piersma:2003dj, Barneche:2016ke}. Low consistency in individual reaction norms (i.e., low repeatability) can introduce variability in metabolic rate across temperatures which can give rise to spurious patterns of temperature dependence. If individuals respond to temperature consistently, mass-scaling is expected to be robust to temperature changes (Clarke 2004). Despite studies on a range of taxa recognising that individuals differ in their metabolic thermal plasticity, its repeatability has rarely been formally estimated (but see {Briga:2017dr}{Reveillon:2019jda}).

Here we examine how individuals differ in energy expenditure in relation to body size and acute temperature changes and how it may impact mass-scaling exponents in the delicate skink (*Lampropholis delicata*) . We repeatedly measured routine metabolic rate (RMR) over four months address three key questions. (1) Does metabolic thermal plasticity consistently differ among individuals? (2) How does repeatability of RMR change at a given temperature? (3) Do population mass-scaling exponents change with temperature when accounting for among- and within-individual variation in RMR? Unravelling the complexities of individual physiological processes will have important consequences for understanding how populations respond in challenging environments.

# Materials and Methods

## Lizard collection and husbandry

*Lampropholis delicata* is a small oviparous, skink found in throughout Eastern Australia {Chapple:2011kj}. They have a short lifespan (2 – 4 years in the wild) and their reproductive season is from September – February {Chapple:2014ey}. Between 28 August and 8 September 2015, forty-two male *L. delicata* were collected from two sites near Sydney, Australia. Lizards were caught by hand or by mealworm fishing and were transported individually in calico bags in an ice-cooler to Macquarie University. Lizards were housed in a temperature-controlled room set at 26ºC and were provided with a thermal gradient to allow for thermoregulation. Each lizard was kept individually in an opaque plastic enclosure measuring 35cm x 25cm x 15cm (L x W x H). Each enclosure was lined with newspaper and lizards were given access to a water bowl and tree bark as a refuge. Enclosures were placed under UV light (11L:13D). Lizards were fed three to four small crickets (*Acheta domestica*) dusted with calcium powder and multi-vitamin every two days when metabolism measurements were not taking place. Animal collection was approved by the New South Wales National Parks and Wildlife Service (SL101549) and procedures were approved by the Macquarie University Ethics committee (ARA 2015/015) and University of New South Wales Animal Care and Ethics committee (ACEC 15/51A).

## Measuring routine metabolic rate at different temperatures

Closed-system respirometry was used to measure RMR which is the rate of energy consumption of lizards at a given temperature in a resting, post-absorptive state but also includes energetic costs of random activity that we were not able to completely control for (Withers 1992; Mathot & Dingemanse 2015). Given the scale of our experiment, it was not possible to use intermittent-flow through respirometry. RMR was measured as CO2 production per unit time ( mL min-1) because CO2 production was more readily detectable for smaller organisms and were less susceptible to fluctuations in water vapour. Our data showed that CO2 production was strongly correlated with O2 consumption nonetheless (*r* =0.94, p = <0.05]). Measurements took place between 26 December 2016 - 19 March 2017. Due to logistical constraints, lizards were randomly assigned to one of two blocks for metabolism measurements (block 1: n = 23, block 2: n = 22). We used two incubators (LabWit, ZXSD-R1090) to precisely control the temperature at which measurements were taken (+/- 1ºC). Measurements were taken in a random order between 22ºC – 32ºC over three days (measurements at two temperatures per day). Each animal was repeatedly measured across these temperatures every 10 days (10 sampling sessions in total). We also statistically accounted for the order of temperatures in our analyses in case of carry over effects of extreme temperatures experienced by an individual on subsequent metabolic measurements (see below).

After a 24 hour fast, the body temperature of each individual inside their enclosure was taken using an infrared laser gun (Stanley stht0-77365) in the morning (~06:00). Each lizard was gently encouraged into their 146mL opaque chamber and then weighed using a digital scale to the nearest 0.01g (Ohaus SP-202). The chambers were placed inside the incubators in the dark at a randomised temperature for 30 minutes. The lids of the chambers were left ajar during this time to minimise CO2 build up. After 30 minutes, each chamber was flushed with fresh air and sealed. A 3 mL ‘control/baseline’ air sample was immediately taken via a two-way valve to account for any residual CO2 that was not flushed from the chambers. The chambers were left in the incubator at the set temperature for lizards to respire. After 90 minutes, two 3mL air samples were taken from each chamber. Chambers were then reopened and flushed with fresh air before placed back into the incubator for the next measurement temperature following the same procedure.

All air samples were injected into the inlet line of a Sables System FMS (Las Vegas NV, USA) with the flow rate set to 200 mL min-1 to measure and *.* Water vapour was scrubbed from the inlet air with Drierite. Output peaks were processed using the R package ‘metabR’ (<https://github.com/daniel1noble/metabR>). The rate of CO2 produced by an individual was calculated following equation 4.21 in Lighton 2008:

Equation: 1

where %CO2 is the cumulative percentage of CO2 in air sample above baseline, which was corrected by subtracting any ‘residual’ CO2 from the initial flush from the larger of the two air samples; Vchamber is the volume of the chamber (146 mL); Vlizard is the volume of the lizard, assuming that the mass of the lizard is the same as its volume, and *t* is the duration of time in minutes after where the chamber has been sealed and the first air sample was taken (90 minutes).

## Statistical analyses

All statistical analyses were conducted using the R environment, version 3.6.1 (Core Team 2013). For details on data cleaning see electronic supplementary materials (see ESM). Initial analyses showed that there were no differences in logbetween statistical blocks of lizards therefore ‘block ID’ was not included in our final models (see ESM). Although lizards were kept in a temperature-controlled room, there may still have been temperature differences between enclosures that had carry-over effects on metabolic rate. We tested whether the previous measurement temperature or the body temperature measured in the home enclosure before the first measurement influenced log at subsequent temperatures. We found that a model containing ‘previous temperature experience’ as a covariate was better supported compared to a model without it (WAIC (Full model – reduced model) = -8.39), we therefore included ‘previous temperature experience’ in all subsequent analyses (Table S1). Collinearity between our predictor variables was checked using a scatterplot matrix (Fig. S1) and Pearson correlation coefficients are presented in Table S2. All data and code with which to generate our results are openly available via the Open Science Framework (see Data Accessibility).

We used Bayesian generalised linear mixed models from either the package ‘brms’ (Bürkner 2017) or ‘MCMCglmm' (Hadfield 2010). For logistical and feasibility reasons, we fitted the random slope model using ‘MCMCglmm’, and a multivariate response model using ‘brms’. We verified that the overall results did not change depending on what package was used. Details on model priors and set up are presented in the ESM. For every model, we pooled the posterior estimates from multiple chains and presented posterior means and their 95% credible intervals.

Measurement error and repeatability of metabolic thermal plasticity

Repeatability is a ratio of among-individual, within-individual and residual variance components (R = σA / σA +σW + σR) and represents the proportion of phenotypic variance attributed to among-individual differences (Nakagawa & Schielzeth 2010). The relative contribution to each variance component can shed light on the processes that promote repeatable traits (Dingemanse & Dochtermann 2013). Within-individual variation, also referred to as ‘predictability’, constitutes as the deviation of an individual relative it its own mean (Westneat, Wright & Dingemanse 2014; Cleasby, Nakagawa & Schielzeth 2014). Measurement error can bias the estimation of within-individual variance which could affect repeatability estimates. {Ponzi:2018hb}. Consequently, we fitted a nested random effect of individual ID and sampling session and temperature (ID:sampling session :Temp) so that variance due to measurement error could be partitioned into the residuals. See {Ponzi:2018hb} for further explanation.

We also wanted to take into consideration that metabolic rate could change over time and this change could depend on temperature because our study spanned over four months. We fitted a nested random effect of individual ID and sampling session (ID:sampling session) to decompose the variance that is attributed to the timing of each sampling sessions for every individual. For further explanation, see Araya-Ajoy, Mathot & Dingemanse 2015.

We fitted the following random slope model in 'MCMCglmm’ with a long format dataset (nobs = 4952) in order to quantify the repeatability of metabolic thermal plasticity (i.e. slopes for each individual.

log ~ Temp + zlogBodyMass + PriorTemp + (1+ Temp| ID) + (1+ Temp| ID:session) + (1| ID:session :Temp)

where: log is log-transformed ; Temp is the temperature in degrees Celsius; zlogBodyMass is log-transformed body mass that is then subsequently z-transformed; PriorTemp is previous temperature experience. Individual ID, as well as sampling session nested within individual ID was included as a random intercept. Temperature was included as a random slope for both these intercepts . Finally, temperature nested within sampling session nested within individual ID was included as a random intercept to deal with measurement error. The repeatability of the slope is calculated following equation 1 in the ESM (see also Araya-Ajoy et al. 2015).

Repeatability of the average SMR at each temperature and cross-temperature correlations of metabolic rate

After assessing whether individuals differ in their metabolic thermal plasticity, we were interested in knowing whether consistent among-individual differences in average SMR change across temperatures. We fitted a multivariate response model to a wide format dataset (nobs = 802) where we treated log measured at each temperature as a 6 x 6 response matrix.

~ zlogBodyMass + PriorTemp + (1|ID) + (1| ID:session)

Where, is the metabolic rate for individual 1 in sampling session 1 at 22ºC and is the metabolic rate for individual 1 in sampling session 10 at 22ºC so forth. Similar to the random slope models, we included zlogBodMass and PriorTemp as fixed effects. Note that temperature is no longer a predictor or a random slope as temperature is now part of the response matrix. We included Individual ID and sampling session nested within ID were as random intercepts. In this model, sampling session nested within ID will partition out measurement error to the residuals. We calculated temperature specific repeatability following Equation 2 in the ESM.

We were interested in the extent to which SMR was correlated across all temperatures as this may illuminate trade-offs in physiological function at different temperatures. We obtained cross-temperature correlations at the among-individual and among-sampling session-within-individual level using the variance-covariance matrices from the multivariate response model.

Mass-scaling exponents at different temperatures

Population estimates of scaling exponents can be affected by the different contributions of within- and among-individual variation (van de Pol & Wright 2009). We therefore wanted to partition out within-individual effects in order to obtain more precise among-individual estimates of mass-scaling across temperatures. To achieve this, we calculated the mean mass across all sampling sessions for each individual (among-individual effect), and subtracted an individual’s mass from its own mean to account for within-individual effects (i.e. within-individual centering, see van de Pol & Wright 2009). These mass effects were log-transformed and included in two models fitted in ‘brms’ using a long complete dataset of nobs = 3933. The first model (interaction model) had the following structure,

log~ Temp \* logAmongIDMass + Temp \* logWithinIDMass + (1 + logWithinIDMass|ID) + (1| ID:sampling session :Temp)

where: Temp \* logAmongIDMass is the interaction term between temperature and the log transformed among individual mass effect; Temp \* logWithinIDMass is the interaction term between temperature and the log transformed within individual mass effect. Individual ID was fitted a random intercept with logWithinIDMass as a random slope as individuals masses changed at different rates through the study (see Fig S3). We also included (1| ID:sampling session :Temp) to account for measurement error. The second model (main effects model) only had the main effects of temperature, the among individual mass effect and the within-individual mass effect and the same random effects structure as the interaction model. We tested whether population mass-scaling exponents (i.e. the among individual mass effects) changed with temperature by comparing information criterions (wAIC and loo values) between model one and two. We also presented in the ESM (Fig. S4, Table S5) an analysis that compared the within- and among individual scaling exponents with exponents from a model that represents the typical analysis of a metabolic scaling study model that did not account for the multi-level variation in the data.

# Results

## Repeatability of metabolic thermal plasticity

Individual slopes were repeatable (Rslope = 0.25, Lower CI = 2.48 10-8, Upper CI = 0.69) indicating a significant individual by environment interaction (I x E) that was consistent over time (Fig. 1). This suggests that individuals’ metabolic rate show different capacities to adjust to temeperature.



**Figure 1** – Predicted individual reaction norms for forty-two individuals of log (mL) at six measurement temperatures at an average log mass at sampling session one (left panel), five (middle panel) and ten (right panel) reaction norms were estimated using Points represent predicted values for log . Each line represents a unique individual (n = 42).

## Repeatability of average standard metabolic rate at each temperature

We found that the repeatability of average (i.e. individual intercepts) showed a slight increase with temperature (Fig. 2). Temperature-specific repeatability was greatest at 30ºC and the lowest at 22ºC (Fig. 2 ,Table S3). Upon closer inspection of the variance components at each temperature, within individual variation decreased over the temperature gradient, whereas among individual variation remained relatively consistent with temperature (Fig 2B). In other words, individuals were responding more consistently as temperatures became hotter and while variation among individuals increased slightly (Fig 2).



**Figure 2** – A) Posterior mean of repeatability and variance components of log (mL) at six measurement temperatures estimated over four-month period across n = 42 individuals. Left axis is for adjusted repeatability. Blue axis is for variance. Circles represent adjusted repeatability. Blue upright triangles represent among-individual variance. Inverted blue triangles represent within-individual variance. Error bars represent 95% credible intervals. See Statistical analyses and ESM for more details.

## Cross-temperature correlations in metabolic rate

Metabolic rate across temperatures were positively correlated at both the among-individual and within-individual level (Fig. 3, Table S4). Although, the magnitude of correlations were weaker at the within-individual level (Fig.3). Some individuals maintained a consistently high metabolic rate relative to other individuals, while others had a relatively low metabolic rate across all temperatures. This created a positive relationship between metabolic rate at the among individual-level. Metabolic rate measured at neighbouring temperatures (e.g. 22ºC and 24ºC) were strongly correlated, but the strength of this correlation decreased with increasing differences between the two temperatures (Fig. 3). 

**Figure 3** – Cross-temperature correlations of log (mL) at the among-individual level (left) and at the within-individual level (right) estimated from n = 42 individuals. Diagonal values are each measurement temperatures. Lower triangle represents posterior mean estimates of correlations. Width and colour of the ellipse in the upper triangle represents the strength of the correlation. See Statistical analyses and ESM for more details.

## Temperature dependence of population mass-scaling exponents

The model containing only the main effects of temperature and the among- and within-individual mass effects was better supported than a model that included the interaction terms (Main effects model: WAIC = 2133.9, loo = 2322.2, Interaction model: WAIC = 2124.90, loo = 2358.5). This suggests a lack of temperature dependence in mass scaling Fig. 4. Across all temperatures, the average mass-scaling exponent is 0.96 (Lower CI = 0.39, Upper CI = 1.52) which is in line with values reported for Squamates (Uyeda *et al.* 2017). There was a trend for within-individual exponents to be larger than among-individual exponents (Fig. S4, Table S5). In an additional analysis presented in the ESM, estimates of mass-scaling exponents tended to be spurious and estimated with a larger degree of error when the within- and among-individual effects as well as measurement error are not statistically accounted for (Fig. S4, Table S5).

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**Figure 5** – (Top) Posterior mean estimates of population mass scaling exponents (i.e. among individuals) of (mL) across six measurement temperatures when within individual variation in mass and measurement error in metabolic rate has been statistically accounted for. The dashed line represents the mass-scaling exponent of 0.83 estimated for squamates from Uyeda (2017). Error bars represent 95% credible intervals. (Bottom) Raw log plotted against log body mass for a random subset of n = 20 individuals at six measurement temperatures. Each uniquely coloured point represents one individual. Thick bold line represents the change in log over log body mass across all individuals (among-individual mass-scaling slope). Thin lines represent the change in log over log body mass within an individual (within-individual mass-scaling slopes)

# Discussion

Our results show that metabolic thermal plasticity was somewhat repeatable over the four months of study. Moreover, the repeatability of average SMR increased with temperature which was largely due to a decrease in within-individual variance. Cross-temperature correlations of SMR were all positive at the among-individual and within-individual level. However, the strength of these correlations was not uniform across all temperatures. Population mass scaling exponents were not strongly affected by temperature and were also more precise and in line with values reported for squamates when within-individual and measurement error variation was partitioned out. Below we discuss the implications of our results for understanding how plasticity may evolve, and how SMR scales at different hierarchical levels.

## Consistent variation in metabolic thermal plasticity

Natural selection acts on phenotypic variation among individuals. Consistent among-individual variation is therefore a key prerequisite for any trait to evolve and sets the ‘upper limit of heritability’ (Falconer 1952, c.f. Dohm 2002). Our findings \individuals differ consistently in how their metabolic rate responds to acute temperature changes over an ecologically relevant time. As temperatures became hotter, an individual’s SMR showed a decrease in within-individual variance, which suggests that individuals were responding with greater predictability {Westneat:2014ki}. Macronutrient breakdown and the production of ATP may be at homeostatic balance at warmer temperatures promoting consistent metabolic rate within individuals

(Somero 1978). In support of this, 32ºC is well within the range of preferred temperatures of this species where biochemical activities are likely to be operating optimally (Merritt, Matthews & White 2013; Goulet, Thompson & Chapple 2016). The compounding effects of relatively high among-individual and low within-individual variation at warmer temperatures may mean that, not only is there a greater potential for selection in hot environments, but selection can operate more effectively (Cleasby *et al.* 2014; Nakagawa *et al.* 2015). Assuming that individual differences has a genetic basis and therefore heritable, our results suggests that metabolic thermal plasticity may undergo natural selection and may facilitate adaptive evolutionary change in population metabolic reaction norms (Ghalambor *et al.* 2007). Indeed, individuals were wild caught and their pedigree unknown, our repeatability estimates encompass permanent environment effects as well as parental effects {Dingemanse:2013kf}. Common-garden studies that set out to measure the consistency and heritability of metabolic thermal plasticity in animals with known relatedness would be a valuable next step.

## Cross-temperature correlations

Metabolic rate was positively correlated across all temperatures at among-individual level. This suggests that individuals differ in their plastic responses, but their rank order maintained the same across different thermal environments. Individuals may vary in their acquisition or allocation of resources to their physiological system which may enable certain individuals to maintain a consistently high SMR across all temperatures (De Jong & Van Noordwijk 1992; Angilletta *et al.* 2003). Moreover, consistent individual differences in SMR, irrespective of the thermal environment, may be functionally linked with other aspects of the phenotype. Our results give precedence to ‘pace-of-life’ theory where individual differences in energetic expenditure may promote consistent differences in behaviour and life-history within the same population (Biro & Stamps 2008; Careau *et al.* 2009).

Cross-temperature correlations may be useful way to detect trade-offs which has been hypothesised as an important mechanism in shaping reaction norms (Angilletta *et al.* 2003). Generalist-specialist trade-offs occur when enhanced physiological function in one environment diminishes function in another environment and can manifest as a negative cross temperature correlation. We show that at the within individual level cross-temperature correlations were weakly positive which implies no strong trade-offs between 22ºC and 32ºC. Since the majority of the estimated reaction norms do not intersect between these temperatures, we cannot rule out the possibility of trade-offs at may occur at extreme temperatures. Future efforts should attempt estimating thermal reaction norms between temperatures that range from the critical thermal minima and maxima of *L.delicata* to determine thermal specialisation and the proximate mechanisms that may promote it {Goulet:2017dr, Greer:2005vu}

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Assuming phenotypic cross-temperature correlations reflect the underlying genetic architecture of metabolic rate (Roff 1995), our results may also have important implications in understanding constraints on the evolution of metabolic thermal plasticity. The strength of among-individual cross-temperature correlation can dictate how strongly selection acting on one component of the reaction norm will result in indirect selection on another (Via *et al.* 1995). In this study, the strength of cross-temperature correlations between neighbouring temperatures (e.g., 28°C vs. 32°C) were stronger compared to correlations at more distant temperatures (e.g., 22°C vs. 32°C). More efficient physiological processes at the preferred temperature of *L. delicata* may result in stronger correlations at higher temperatures. This result is congruent with decreased residual variance at higher temperatures. Acclimation studies that shift the thermal optimal of individuals would be an insightful way to test this hypothesis.

## Population mass scaling across different temperatures

Our mass-scaling exponents did not change with temperature, which is in disagreement with the growing number of studies that show temperature dependence of mass scaling exponents (Glazier 2005; Killen *et al.* 2010; Price *et al.* 2012; Glazier 2015; Barneche *et al.* 2016). Disparity between our results and these studies may be due to the method with which we quantified mass scaling exponents. In our study, we sampled sexual mature adults repeatedly over four months in order to estimate a static mass scaling relationship, while other studies tend to measure ontogenetic allometry (i.e. measure body mass and metabolic rate throughout development, Glazier 2009). The energetic demands of growth during ontogeny may be more sensitive to changes to temperature and therefore result in temperature-dependence in ontogenetic mass scaling exponents (Hirst, Glazier & Atkinson 2014; Barneche & Allen 2018). In support of this, a recent comparative analysis has shown that development (passing through life stages) shows stronger temperature dependence than increase in mass (Forster, Hirst & Woodward 2011).

The magnitude and precision of mass scaling exponents may be affected by processes occurring at different hierarchical levels. Genetic and developmental differences that impact the physiological system can maintain variation among individuals (Dingemanse & Wolf 2013). While fluctuations in the internal environment, such as circulating hormones and body composition, can affect the predictability of an individual’s response (Scott, Mitchell & Evans 1996; McCue 2010; Dupoué, Brischoux, Lourdais, Angelier 2013). After accounting for within individual level and measurement effects, our exponent estimates were more in line with values reported from a phylogenetically informed analyses in squamates (Uyeda *et al.* 2017). This result has important implications for current designs of metabolic scaling studies as SMR and body mass tend to only be measured once, making them sensitive to sampling error and within-individual ‘noise’. Theoretical studies that make use of predictive relationship between body mass and metabolism and other traits should be more aware of the different sources of variation when trying to extrapolate individual level processes to higher levels of biological organisation. Future work is needed to investigate the degree to which intra-individual variance in SMR and body mass impact scaling exponents as this has largely been neglected and yet may help elucidate why mass scaling exponents are variable at higher levels of biological organisation (see Glazier, 2005, {McLean:2007tl, Maxwell:2003hh}).

# Conclusion

In this study, we aim to link individual plastic responses to temperatures changes to understand how energy expenditure may change across a population. We found evidence that metabolic thermal plasticity is indeed repeatable over ecologically relevant time scales and could be subjected to natural selection to shape population reaction norms to cope with climate change. This may be particularly important for ectotherms that inhabit variable environments. Our work highlights the role of within-individual variance and temperature may play in the context of facilitating the evolution of metabolic thermal plasticity. While the underlying mechanisms for cross-temperature correlations are not well understood, they suggest that thermal reaction norms have the capacity to evolve more malleable forms which may allow populations to better track warming temperatures. We emphasise the importance of considering among- and within-individual in the context of estimating mass-scaling at the population level. If multi-level variation is not corrected for, we run the risk of misinterpreting and adding more variation to population exponents which may be composite of among- and within individual effects.

# Author contributions

All authors conceived the ideas and designed the study; FK and CF collected the data; FK, DN, SN analysed the data; FK wrote the first draft and all authors contributed to revising the manuscript.

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# Data accessibility

Datasets and code used to generate results of this study is accessible via Open Science Framework (DOI 10.17605/OSF.IO/TZ2H5).

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