

Supplementary Material

Title: Countergradient variation in reptiles: thermal sensitivity of developmental and metabolic rates across locally adapted populations

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Methods

Meta-analysis for thermal sensitivity of developmental and metabolic rates across locally adapted populations

Published articles presenting data on developmental and metabolic rates across populations of reptiles were collected from ISI *Web of Science* using search terms “local adaptation”, “latitude”, “altitude” OR “gradient” AND “development*”, AND “temperat*” OR “therm*” together with the terms “lizard” OR “snake” OR “reptile”. This search yielded 83 unique entries, and of these 62 were rejected based on irrelevance. Studies were only included if they included either a common garden (CG) or reciprocal transplant (RT) method, whereby reptile species from more than one population were collected and eggs incubated under either a single (CG) or multiple (RT) ecologically relevant, experimentally controlled, temperatures. Development time was measured as the number of hours from oviposition until hatching (h). Metabolic rate during development was measured as embryo heart rate in beats per minute (BPM). Studies were only included if they reported sample size or a test statistic from which sample size could be calculated (e.g., degrees of freedom) as well as a measure of error from which standard deviation could be determined in order to minimise sampling bias.

I calculated effect sizes for each study using Hedges’ g , which provides unbiased, standardised mean differences in D or MR for comparisons between cold- and warm-adapted populations, while adjusting effects for small sample sizes (Borenstein et al., 2009). To account for shared evolutionary history among species driving any observed patterns in D and MR reaction norms, I constructed a tree topology using the open tree of life and the ‘rotl’ package and compared the fit of models with or without phylogeny included using the ‘pgls’ function in the R package ‘caper’ and Rv3.6.1 (Orme et al., 2013; Hinchliff et al., 2015; Michonneau et al., 2016). When comparing models with (“phylogeny model”) and without (“null model”) phylogeny, I found that phylogeny did not explain sufficient variance in

Hedges' g , ($AIC_{\text{phylogeny}}: 94.42 > AIC_{\text{null}}: 83.52$), hence effect sizes were not corrected for phylogenetic signal.

Publication bias

To test for publication bias (arising from sampling a greater proportion of statistically significant, and therefore published results) I explored the relationship between the effect size, (Hedges' g) and precision of each estimate ($1/\text{SE}$) using funnel plots (Borenstein *et al.* 2011). In the case of publication bias, we would expect to see an absence of effect sizes from studies with small sample size, and therefore low precision. Despite a paucity of available studies meeting the meta-analysis criteria, effect sizes were distributed symmetrically around the mean, and there appeared to be little evidence of publication bias.

Table S1: Summary of reptile data used to calculate effect sizes (Hedges' g) for differences in the thermal sensitivity of developmental time (D) and metabolic (heart) rate (MR) across cold and warm-adapted populations. Data includes studies reporting development time (D; time from oviposition until hatching) and metabolic rate (HR; measured as heart rate) for embryos measured across either a single incubation temperature (common garden; CG) or multiple incubation temperatures (reciprocal transplant; RT). Hedges' g provides standardised mean differences in D and HR between cold- and warm-adapted populations that adjusts effects for small sample sizes. Positive Hedges' g values indicate cold-adapted populations have longer developmental times (D) or higher metabolic rates (HR) relative to warm-adapted populations (and vice versa), NS (non-significant). Species natural nest temperature ranges ($^{\circ}\text{C}$) are included where provided by each study, and incubation temperature variance ($^{\circ}\text{C}$) is provided where studies used fluctuating thermal regimes.

| Family | Species | Trait | Gradient | Method | Natural nest temperature range ($^{\circ}\text{C}$) | Incubation temperatures ($^{\circ}\text{C}$) and variance | Effect size (Hedges' g) | Evidence for CoGV or CnGV? | Reference |
|-----------------|-----------------------------------|-------|-------------------------|--------|---|---|----------------------------|----------------------------|--------------------------|
| Order: Squamata | | | | | | | | | |
| Agamidae | <i>Phrynocephalus przewalskii</i> | DT | Climate (cold/warm) | CG | N/A | 28.0 | -0.201 | CnGV | Zeng et al., (2013) |
| Agamidae | <i>Phrynocephalus przewalskii</i> | MR | Climate (cold/warm) | CG | N/A | 28.0 | -0.579 | CoGV | Zeng et al., (2013) |
| Dactyloidae | <i>Anolis cristatellus</i> | DT | Temperature (cold/warm) | CG | 25.5 – 33.6 | 26.7 | -0.276 | CnGV | Tiatragul et al., (2017) |
| Dactyloidae | <i>Anolis cristatellus</i> | DT | Climate (cold/warm) | RT | 25 – 43 | 26.5, 29.0, 39, 43 | 0.597 | CoGV | Hall and Warner (2018) |

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|-------------|------------------------------|----|--------------------------------|----|-------------|---|--------|------|-------------------------------|
| Dactyloidae | <i>Anolis cristatellus</i> | MR | Climate (cold/warm) | RT | 25 – 43 | 26.5, 29.0, 39, 43 | -0.302 | CoGV | Hall and Warner (2018) |
| Dactyloidae | <i>Anolis sagrei</i> | DT | Temperature (cold/warm) | CG | 25.5 – 33.6 | 26.7 | 0.218 | CoGV | Tiatragul et al., (2017) |
| Dactyloidae | <i>Anolis sagrei</i> | DT | Latitude (cold/warm) | CG | N/A | 28.0 | -0.100 | CnGV | Fetters and McGlothlin (2017) |
| Lacertidae | <i>Eremias multiocellata</i> | DT | Altitude (cold/warm) | RT | N/A | 25.0 (7), 29.0 (11), 31.0 (13), 35.0 (17) | -0.705 | CnGV | Tang et al., (2012) |
| Lacertidae | <i>Eremias argus</i> | DT | Altitude (cold/warm) | CG | 15.5 – 31.8 | 24.0 (6) | -1.810 | CnGV | Sun et al., (2013) |
| Lacertidae | <i>Eremias argus</i> | MR | Altitude (cold/warm) | RT | 15.5 – 31.8 | 24.0 (6), 28.0 (6) | -0.093 | CoGV | Sun et al., (2013) |
| Lacertidae | <i>Iberolacerta cyreni</i> | DT | Altitude (cold/warm) | RT | N/A | 22.0, 26.0, 30.0 | -0.045 | NS | Monasterio et al., (2016) |
| Lacertidae | <i>Podarcis muralis</i> | DT | Latitude (cold/warm) | RT | 9.5 – 33.5 | 20.0, 24.0 | -0.373 | CnGV | While et al., (2015) |
| Lacertidae | <i>Podarcis muralis</i> | MR | Latitude (cold/warm) | RT | 9.5 – 33.5 | 20.0, 24.0 | 0.023 | NS | While et al., (2015) |
| Lacertidae | <i>Psammodromus algirus</i> | DT | Altitude, Latitude (cold/warm) | CG | N/A | 28.0 (4) | -0.344 | CnGV | Díaz et al., (2012) |
| Lacertidae | <i>Psammodromus algirus</i> | DT | Altitude (cold/warm) | CG | N/A | 24.0 | -0.569 | CnGV | Verdú-Ricoy et al., (2014) |
| Lacertidae | <i>Psammodromus algirus</i> | DT | Altitude (cold/warm) | RT | N/A | 27.0, 30.0 | -0.391 | CnGV | Iraeta et al., (2006) |
| Lacertidae | <i>Takydromus wolteri</i> | DT | Altitude (cold/warm) | CG | 15.5 – 31.8 | 24.0 | -0.555 | CnGV | Sun et al., (2013) |
| Lacertidae | <i>Takydromus wolteri</i> | MR | Altitude (cold/warm) | RT | 15.5 – 31.8 | 24.0, 28.0 | -0.004 | NS | Sun et al., (2013) |

| | | | | | | | | | |
|--------------------------|-----------------------------|----|--------------------------------|----|-------------|----------------------|--------|------|---------------------------------------|
| Lacertidae | <i>Zootoca vivipara</i> | DT | Altitude (cold/warm) | RT | 1 – 35 | 21.0, 25.0, 29.0 | -1.241 | CnGV | Rodriquez-Diaz and Braña (2012) |
| Phrynosomatidae | <i>Sceloporus undulatus</i> | DT | Latitude (cold/warm) | RT | 20 – 34 | 24.0 (10), 27.0 (14) | -0.874 | CnGV | Oufiero and Angilletta Jr (2006) |
| Phrynosomatidae | <i>Sceloporus undulatus</i> | DT | Altitude, Latitude (cold/warm) | CG | 20 – 34 | 27.0 (14) | -0.084 | NS | Niewiarowski and Angilletta Jr (2008) |
| Phrynosomatidae | <i>Sceloporus undulatus</i> | DT | Latitude (cold/warm) | RT | 20 – 33.5 | 25.0, 28.0 | -0.336 | CnGV | Du et al., (2010) |
| Phrynosomatidae | <i>Sceloporus undulatus</i> | MR | Climate (cold/warm) | RT | 20 – 33.5 | 25.0, 28.0 | 0 | No | Du et al., (2010) |
| Scincidae | <i>Plestiodon chinensis</i> | DT | Altitude (cold/warm) | RT | 25 – 31 | 24.0, 28.0, 32.0 | -0.047 | NS | Lu et al., (2014) |
| <i>Order: Testudines</i> | | | | | | | | | |
| Emydidae | <i>Chrysemys picta</i> | DT | Latitude (cold/warm) | RT | N/A | 27.5, 28.5 | 0 | No | Bodensteiner et al., (2019) |
| Geoemydidae | <i>Mauremys mutica</i> | DT | Latitude (cold/warm) | RT | 21.5 – 29.2 | 26.0, 28.0, 30.0 | -0.097 | CnGV | Zhao et al., (2015) |
| Geoemydidae | <i>Mauremys mutica</i> | DT | Latitude (cold/warm) | RT | 23.2 – 29.5 | 26.0 (3), 28.0 (1.5) | 0.520 | CoGV | Li et al., (2018) |
| Trionychidae | <i>Pelodiscus sinensis</i> | DT | Latitude (cold/warm) | RT | 18.6 – 37.2 | 26.0 (3), 31.0 (3) | -0.059 | CnGV | Li et al., (2018) |
| Trionychidae | <i>Pelodiscus sinensis</i> | MR | Latitude (cold/warm) | RT | 18.6 – 37.2 | 26.0 (3), 31.0 (3) | 0.844 | CnGV | Li et al., (2018) |

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