Oikos 126: 447–457, 2017
doi: 10.1111/oik.03712

Subject Editor: Isabel M. Smallegange. Editor-in-Chief: Dries Bonte. Accepted 19 August 2016

Fine with heat, problems with water: microclimate alters water loss in a thermally adapted insular lizard

Anat Belasen, Kinsey Brock, Binbin Li, Dimitra Chremou, Efstratios Valakos, Panayiotis Pafilis, Barry Sinervo and Johannes Foufopoulos

A. Belasen (abelasen@umich.edu), Dept of Ecology and Evolutionary Biology, Univ. of Michigan, Ann Arbor, MI, USA. – AB, K. Brock, B. Li and J. Foufopoulos, School of Natural Resources and Environment, Univ. of Michigan, Ann Arbor, MI, USA. – BL also at: Nicholas School of Environment, Duke Univ., Durham, NC, USA. – D. Chremou, Dept of Biology, Univ. of Edinburgh, Edinburgh, UK. – E. Valakos, Dept of Animal and Human Physiology, National and Kapodistrian Univ. of Athens, Athens, Greece. – P. Pafilis, Dept of Zoology and Marine Biology, National and Kapodistrian Univ. of Athens, Athens, Greece. – B. Sinervo, Dept of Ecology and Evolutionary Biology, Univ. of California, Santa Cruz, CA, USA.

Global change, including habitat isolation and climate change, has both short- and long-term impacts on wildlife populations. For example, genetic drift and inbreeding result in genetic impoverishment in small, isolated populations, while species undergo range shifts or adaptive phenotypic change in response to shifts in environmental temperatures. In this study, we utilize a model system in which Holocene landscape changes have occurred to examine long-term effects of population isolation. To examine how isolation may constrain responses to climate change, we characterized ecophysiology across land-bridge island populations of Erhard's wall lizard *Podarcis erhardii*. We hypothesized that 1) small, isolated populations that are likely genetically depauperate would exhibit lower phenotypic variability; and 2) populations would be adapted to local microhabitat conditions. We compared a population at a low elevation site on the large island of Naxos with two small populations on nearby islets to determine the effects of population fragmentation. We further compared the low elevation Naxos population with two high elevation sites characterized by disparate microclimates to examine the effects of microclimate. To assess the thermal biology and ecophysiological limits of the study species we measured operative body temperatures (T_e), field body temperatures (T_b), preferred temperatures (T_p), thermal tolerances (CT_{max} and CT_{min}), and evaporative water loss (EWL). Our results indicate that small, isolated populations did not exhibit variation in thermal biology or evaporative water loss, while EWL and thermoregulatory effort varied according to microhabitat characteristics. This study integrates fine-scale measurements with environmental data to provide a holistic view of the relationships between ecophysiology, fragmentation, and microclimate. Our methods can be applied to other ectotherms to gain a better understanding of potential impacts of global change on natural populations.

Global biodiversity is threatened by a number of widespread processes, and yet much remains to be understood about the interactions of multiple stressors in natural populations. Two of the most widespread and severe wildlife stressors are global climate change and habitat separation and isolation (Pereira et al. 2010). While the impacts of each phenomenon on species and ecosystems have been widely studied and quantified, very little is known regarding synergistic effects (Stork et al. 2009).

Climate change can affect species both directly and indirectly. During extreme weather events, ambient temperatures may exceed a species' critical thermal maximum (CT_{max}), the temperature beyond which death by overheating ensues. CT_{max} tends to be more rigid than other aspects of thermal biology (Sinervo et al. 2010, Leal and Gunderson 2012, Araújo et al. 2013), placing constraints on adaptive potential in the face of a shifting climate. Indirect effects of climate change may be mediated through changes in habitat structure or composition, which in some cases decrease thermal niche quality (Parmesan 2006). Mismatches between the fundamental niche and altered conditions can constrict a species' potential activity period, which limits foraging or reproductive activities (Ceia-Hasse et al. 2014, Lara-Resendiz et al. 2015). This is especially relevant for ectotherms, which thermoregulate behaviorally and retreat to thermal refugia to avoid experiencing CT_{max} (Sinervo et al. 2010). Climate change can further impact wildlife through shifts in water availability (Lorenzon et al. 1999, Gunderson et al. 2011, Carneiro et al. 2015). The effects of altered water regimes are particularly important for ectotherms – especially in hot, arid ecosystems.

Isolation of populations as a result of habitat fragmentation can also have numerous effects, as it results in genetic impoverishment due to the combined effects of genetic drift and inbreeding. This can lead to inbreeding depression, which further decreases fitness and resilience (Eldridge et al. 1999, Frankham et al. 2002). Isolated populations may therefore be vulnerable not only due to their small size, but also because the resulting genetic impoverishment undermines their capacity for evolutionary responses to environmental change (Willi et al. 2006, Remon et al. 2011). Island species represent typical examples of such isolated populations. Wildlife adapts to insularity and deviates in numerous ways from mainland populations (Losos and Ricklefs 2009). These adaptations are likely linked to genetic diversity (Frankham et al. 2002, England et al. 2003), as island size has been reported to correspond with genetic variability (Frankham 1997, Hinten et al. 2003). Genetic erosion in populations on very small islands likely corresponds with reduced evolutionary potential (Frankham 1997), which may be a significant disadvantage in times of environmental change. However, this has been rarely tested in natural populations (Gunderson and Leal 2012).

Despite knowledge of the respective impacts of climate change and habitat isolation, little is known about how these two processes may interact. One direct interaction is the inability of populations in isolated landscapes to migrate in response to climate change (Davis and Shaw 2001, Opdam and Wascher 2003), but more nuanced synergisms are likely, and remain to be addressed. For example, non-fragmented populations that are genetically more diverse have been shown to exhibit greater resilience to climate change (Ehlers et al. 2008), which could be due to greater population-level variability in ecophysiological traits. Could genetic impoverishment resulting from population reduction and isolation also reduce population-level phenotypic heterogeneity, and thus impair population resilience against abrupt changes in environmental conditions? Might losses of genetic diversity also be tied to attenuation of plasticity (as seen in canalized island populations, Aubret and Shine 2010), thus removing an important buffer against climatic shifts? These types of interactions are particularly relevant for ectotherms such as reptiles, which are sensitive to environmental conditions and also exhibit low dispersal relative to other terrestrial vertebrates (Kubisch et al. 2016a, b).

In this study, we take advantage of a naturally fragmented system: the land-bridge islands that comprise the Cyclades archipelago (Aegean Sea, Greece). The present-day Cyclades formed over the last ~18 000 years when rising sea levels due to glacial melt fragmented the large ancestral landmass (Perissoratis and Conispoliatis 2003, Kougioumoutzis et al. 2014). These islands are inhabited by relict populations of once-contiguous reptile populations, which are now isolated on the islands due to poor overwater dispersal. Thus, these island reptiles constitute an excellent model system to investigate the potential long-term effects of population isolation and reduction (Newmark 1995, Foufopoulos and Ives 1999, Foufopoulos et al. 2011). Indeed, molecular analyses by Hurston et al. (2009) have demonstrated that across the Cyclades, widespread Erhard's wall lizard *Podarcis erhardii* populations are relicts from a single ancestral Holocene population, and vary in their genetic diversity such that those on small islets retain only a fraction of the diversity found on nearby large islands.

We use the Cyclades island lizards as a model system to examine the long-term impacts of isolation on thermal biology, a characteristic critical to the survival of ectotherms in areas affected by climate change (Sinervo et al. 2010, Kubisch et al. 2016b). Since ectotherms are known to modulate activity in response to local microclimates, we designed our study to test for interactions between population isolation and potential climatic effects. We integrate behavioral observations and fine-scale measurements of thermal biology to examine how population fragmentation or local adaptation to microclimate might affect susceptibility to environmental change. *P. erhardii* communities were selected that either varied in the size of their population (a proxy for genetic diversity), or in the type of microclimate inhabited. This study was designed to answer the following questions: 1) what is the effect of small population size on the variability displayed in the thermal biology of a population? More specifically, do populations that are large and genetically diverse show increased variance in their thermal biology, and thus greater potential resilience against altered environmental conditions? 2) Do the environmental differences caused by elevation and aspect determine the thermal and hydric physiology in lizards with comparable levels of genetic diversity?

Material and methods

Study species and study sites

Podarcis erhardii is a small (snout–vent length, SVL, ~75 mm) Lacertid lizard that occurs across a broad range of habitats on the south Balkans (Lymberakis et al. 2009) and on the central Aegean islands (Fig. 1; Valakos et al. 2008). These diurnal lizards have a bimodal activity pattern during summer, exhibiting a mid-day period of inactivity, which increases in duration with warmer conditions (Valakos 1990). Mating occurs in late spring through early summer and females lay up to two clutches (clutch size 1–5 eggs; Valakos 1990). *Podarcis erhardii* feeds on a wide variety of arthropods (Adamopoulou et al. 1999) though more exotic food sources may enhance its diet (Brock et al. 2014, Deem and Hedman 2014).

We conducted this study on the large island of Naxos and two satellite islets, all located in the Cyclades island cluster (Aegean Sea, Greece). Three study sites are located on Naxos proper and one each on two nearby islets (Fig. 2). The three

Figure 1. A male *Podarcis erhardii* photographed at a low elevation site on the Cycladic island of Naxos (photograph by J. Foufopoulos).

Naxos sites were chosen to span a range of microclimates that vary in terms of elevation and aspect. Two high elevation sites, Naxos south-facing (NSF, 37°6.5′N, 25°32.0′E, 642 m a.s.l.) and Naxos north-facing (NNF, 37°6.2′N, 25°31.6′E; 631 m a.s.l.), are located in proximity (~0.8 km apart) and at similar elevation, but situated on opposing slopes of the same valley. The NSF has few trees and is characterized by relatively dry 'phrygana' (Mediterranean dwarf bush), while the NNF is more heavily vegetated and contains patches of oak forest. Due to the northern aspect, the relative steepness of the slope (~35%), and the shade provided by the trees, this site is noticeably more humid than the neighboring NSF. A third Naxos site (Naxos low elevation, NLE; 37°3′N, 25°23.0′E; 25 m a.s.l.), located on the west coast of the island, was selected to evaluate altitudinal effects on thermal biology. This site is characterized by dry thermo-Mediterranean vegetation and borders small agricultural fields. NLE exhibits very similar climatic and ecological conditions to the two nearby offshore islet sites, allowing us to investigate the effects of small population size on populations subject to a similar microclimate. Aspronissi islet (ASP, 0.0102 km^2, 8 m a.s.l.) is located $<$ 1 km off the western coast of Naxos (37°2.8′N, 25°21.1′E) and is partially covered with halophytic shrubs. The second islet, Ovriokastro (OVR, 0.22 km^2, 23 m a.s.l.), is located ca 8 km W – NW of Naxos (37°9.07′N, 25°17.75′E) and is covered with open *phrygana* scrubs.

All study sites are inhabited by the same wall lizard subspecies (*P. erhardii naxensis*). Islet populations were separated from Naxos by Holocene sea level rise (Ovriokastro: 5600 years bp; Aspronissi: 6100 years bp). While Naxos harbors an estimated population of 10^6 *P. erhardii*, Ovriokastro harbors 2000–3000 and Aspronissi about 200 individuals (J. Foufopoulos unpubl.). Recent genetic analyses by Hurston et al. (2009) indicate that the Naxos population has much higher genetic diversity relative to the smaller satellite islands. For example, the population on Ovriokastro harbors 51% lower allelic richness and 38% lower heterozygosity than the Naxos population (Hurston et al. 2009).

The five study sites were sampled in spring/summer 2011 (0515 – 0715). Individual lizards were captured using either nooses or live mealworm *Tenebrio molitor* baits attached to a hook-less telescopic fishing pole. Individuals were marked with unique toe-clips and transferred to plastic terraria where they were provided with water and mealworms ad libitum. Sex, reproductive status for females (gravid or non-gravid), weight, SVL, tail length and tail condition (autotomized/regenerated or intact) were recorded for each lizard. Eggs that were laid by captive females were removed from enclosures, and each clutch was labeled with the mother's unique ID. Eggs were incubated and hatched in vermiculite-filled cups in a styrofoam incubator, and offspring were retained for an evaporative water loss heritability study. Prior to all laboratory experiments, lizards were acclimated to captive conditions for at least 24 h.

Thermal biology

To measure operative temperatures (T_e) we followed established methodology (Dzialowski 2005, Sinervo et al. 2010, Paranjpe et al. 2012) and used dual-probe HOBO loggers inserted into hollow 'ectotherm models' composed of a 0.25 × 1.5 × 8 cm (wall thickness × diameter × length) PVC pipe (roughly corresponding to the size of an adult *P. erhardii*). The models were painted with a mottled green and taupe pattern to match the likely reflectance of an adult *P. erhardii* (Peterson et al. 1993, Font et al. 2009). Models constructed according to this protocol were validated against

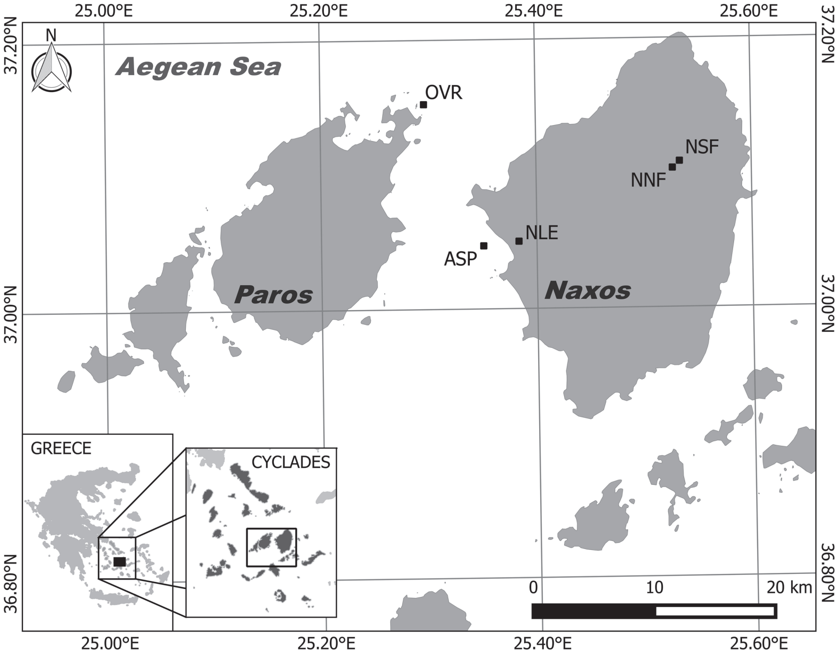

Figure 2. Map depicting the study region on and around Naxos Island, Greece in the northeast Aegean Sea. Study sites are indicated with dark triangles. Codes correspond to sites as follows: ASP – Aspronissi; OVR – Ovriokastro; NLE – Naxos low elevation; NSF – Naxos high elevation, south-facing; NNF – Naxos high elevation, north-facing.

a similar Mediterranean Lacertid lizard *Iberolacerta monticola* with no significant differences between model and lizard temperature readings. Goodness of fit was evaluated with a simple linear regression of T_b against T_e ($a = 1.04 \pm 1.14SE$, $b = 0.95 \pm 0.04$, with a not statistically different from 0 and b not statistically different from 1 and, $R^2 = 0.84$) as well as an analysis of individual deviations of T_e from *I. monticola* T_b ($\Delta = T_b - T_e = -0.458 \pm 0.070SE$). We note that these loggers are designed to represent a hypothetical lizard, since they were not validated specifically against our focal species *P. erhardii*. Calibrations with an African Lacertid revealed similar high goodness of fit between T_b and T_e (Kirchhof and Sinervo pers. comm.) as does the fit for a Phrynosomatid species in Mexico (Lara-Resendiz et al. 2015); therefore our models represent typical arid environment lizards relatively well.

Six data loggers, each with two probes for a total of twelve models, were deployed at each site for at least 24 h for each T_e sample. Sampling locations were spatially stratified within sites, using the parallel terraces lined by man-made dry stone walls as guides. Orientation of strata varied according to the spatial layout of the site. For example, at NSF, the first T_e sample was taken from a location at the northernmost end of the site where lizards were observed, the second from the northern-center of the site, the third from the center of the site, the fourth from the southern-center of the site, and the fifth from the southern end of the site. In each sampling location, two of the loggers were placed in continuous shade (in spaces within stone walls, i.e. primary shelters used by *P. erhardii* lizards), two in mixed sun and shade (e.g. at the margins of dry stone walls or vegetation, where active lizards are generally found) and two in the open sun (on typical basking surfaces). Temperature was recorded at 5-min intervals over a period of five days for each site on Naxos. Temperature was recorded over two sampling days for each of the islet sites, once in the early summer and once in mid-late summer corresponding to the highest period of lizard activity. Before analyzing T_e measurements, data files were manually trimmed to only include daylight hours (sunrise to sunset), and data from all twelve loggers were concatenated and treated as a single dataset from each sampling day to avoid issues associated with non-independence and spatial autocorrelation of this type of sampling (Ackley et al. 2015). Summary statistics for each sampling day at each site were calculated from these larger concatenated datasets. Air temperature data extracted from the Naxos weather station records were used as covariates in statistical analyses to account for daily weather fluctuations and to isolate individual site patterns in temperature.

Field body temperatures (T_b) were measured with a cloacal thermometer in the field within 20 s of capture. Substrate temperature and air temperature (5 cm above the substrate) were also measured contemporaneously. Each lizard's behavior at capture was recorded, and categorized as basking, hiding, or foraging, based on the individual's exposure and activity.

Preferred temperature (T_p) was determined in the laboratory for adult lizards from each of the five populations following the methods of Paranjpe et al. (2012) and Paranjpe et al. (2013). For females that were gravid upon capture, the measurements were repeated after oviposition; this paired design allowed us to evaluate how reproductive status might affect temperature preferences. For approximately eight hours prior to the beginning of the experiment, lizards were not fed to avoid possible confounding effects (Tosini et al. 1994). To measure T_p, a 0.8 mm thermocouple was inserted 5 mm into the cloaca and secured with a small piece of tape. The thermocouple was plugged into a digital thermometer that displayed continuous temperature readings. Lizards were placed in a fiberboard corridor (1 m L × 5 cm W × 30 cm H). We used a 100 W incandescent light bulb at one end, and ice packs at the other end of the corridor to create a thermal gradient from 45°C to 15°C (Van Damme et al. 1986). Lizards were allowed to acclimate in the corridor for 30 min. Body temperature was then recorded every 2 min for 1 h, and these measurements were averaged to obtain an estimate of T_p. For the later calculation of thermal indices, T_{set} was estimated as the 25th and 75th quartiles (Hertz et al. 1993, Lara-Resendiz et al. 2015).

Critical thermal tolerance (CT_{min} and CT_{max}) experiments were conducted on 10–15 post-partum females from each of the five study populations. These measurements provide an estimate of the range of conditions physiologically tolerated by the animals (Kour and Hutchison 1970, Huey and Stevenson 1979). Lizards were not fed for at least eight hours prior to the experiment to avoid any effects of digestion on performance (Tosini et al. 1994). Experimental methods for measuring CT_{min} and CT_{max} were adapted from Leal and Gunderson (2012). Each lizard was heated or cooled slowly (using a 100 W light or a –15°C freezer respectively) from resting temperature (generally 30–35°C), and flipped onto its dorsum at regular temperature intervals. After the lizard righted itself onto its abdomen, heat or cold were further applied, and the process repeated until the lizard could no longer right itself within 10 s. The maximum or minimum temperature at which the righting response was achieved was recorded as the CT_{max} or CT_{min}. At this point, the CT_{max} lizards were rapidly transferred to a container of cool water for several seconds to lower their body temperature while CT_{min} lizards were allowed to warm slowly at room temperature. During the CT_{max} experiment, lizards' body temperatures were continuously monitored using a thermocouple and digital thermometer as in the T_p experiment, whereas in the CT_{min} experiment, a cloacal thermometer was used to take instantaneous readings.

Thermoregulatory effectiveness

To examine the extent of thermoregulation across the five study sites, we used *E*, a metric of thermoregulatory effectiveness, calculated as

$$E = 1 - \left(\overline{d_b} / \overline{d_e} \right)$$

where $\overline{d_b} = T_p - T_b$, and provides an index of thermoregulatory accuracy (smaller d_b indicates greater accuracy); and $\overline{d_e} = T_p - T_e$, which represents a measure of the thermal quality of a habitat (small d_e indicates high thermal quality; Hertz et al. 1993). These calculations assimilate the thermal niche parameters T_e, T_b and T_p, so that the index *E* can be compared across populations (Hertz et al. 1993). *E* can vary from 0 to 1, with thermoconforming populations and/or species exhibiting lower values and thermoregulating ones exhibiting higher values.

Some inherent biases have been identified in the aforementioned model. A variety of different $\overline{d_e}$ and $\overline{d_b}$ combinations may result in the same values of E. For that reason, Hertz et al. (1993) proposed to directly examine $\overline{d_b}$ and $\overline{d_e}$ together with E to assess the real pattern of thermoregulation in a given biotope. In light of this, we also used the complimentary method proposed by Blouin-Demers and Weatherhead (2001) according to which the difference between $\overline{d_e}$ and $\overline{d_b}$ provides an extra index to evaluate thermoregulation. Positive values that arise from the latter difference indicate active thermoregulators, negative values denote lizards that avoid thermally benign habitats, and a value of zero indicates thermoconformers. In other words, a larger difference ($\overline{d_e} - \overline{d_b}$) indicates a higher effectiveness of thermoregulation.

Water loss rates

Evaporative water loss (EWL) data were obtained from both sexes but for different purposes: male EWL data were used for among site comparisons, while female-offspring data were used for a broad-sense heritability study. We quantified the ability of male lizards from four populations (NLE, NSF, NNF and ASP) to conserve water in a standard environment. The OVR site was omitted from the experiment because weather conditions at the end of the study season precluded travel to this islet. Post-reproductive females and their offspring were subject to EWL trials to test for heritability of this trait. Lizards were not fed for eight hours prior to beginning the experiment to avoid confounding effects of digestion or defecation. Lizards were initially weighed with a digital balance (1 mg accuracy), then placed individually in mesh-lid terraria lined with a single sheet of newspaper (Dmi'el et al. 1997).

Trials were run for approximately seven hours, and box positions were rotated twice during each trial to eliminate location effects. Temperature and humidity were recorded at the beginning, middle, and end of each trial using a wall-mounted thermometer/hygrometer to account for any covariant effects. At the end of the trial, each lizard was reweighed, and the difference in weight was taken as the EWL (Carneiro et al. 2015, 2016). EWL/hour was then calculated based on the duration of each trial. If a lizard defecated during the trial, it was reweighed and the experiment was restarted.

Body surface area was estimated for each lizard by making the simplifying assumption that the trunk was cylinder-shaped and the tail was cone-shaped. Cylinder surface area was calculated using SVL as the length and mid-body girth as the circumference. Similarly, cone surface area was estimated using tail length as the height of the cone and pelvic girth as the base circumference. The sum of these calculations was used to estimate total surface area. Water loss per unit time was then divided by surface area to obtain size-independent EWL. This methodology however does not allow for a distinction between cutaneous and respiratory water loss; thus we also make the implicit assumption that both aspects of water loss scale with lizard body surface area.

To investigate the relationship between EWL and moisture availability, we measured water content of soil samples collected in a standardized fashion from the three Naxos sites. At each site, three 10 × 10 cm samples were randomly selected within a three m radius of each other. Soil was collected until bedrock was encountered, generally at ~5 cm below the soil surface. Samples were weighed, air dried for 12–24 h, and then weighed again. Samples were repeatedly weighed for several days to verify complete drying (i.e. no additional weight change). Because all samples belonged to the same soil type (sandy loam), were collected to the same depth, and dried under identical conditions, moisture data were directly compared across sites.

Data analysis

Unless otherwise noted, data were analyzed and test assumptions verified using IBM SPSS Statistics ver. 20. Non-parametric tests were used when data were non-normal or did not meet parametric test assumptions.

We compared environmental conditions across sites by creating general additive mixed models (GAMMs) using the mgcv package in R (ver. 3.0.3, <www.r-project.org>) with daytime mean, maximum and minimum T_e (environmental temperatures) measured by HOBO data loggers as dependent variables (modified from Ackley et al. 2015). Using an additive model building procedure with GCV and as model choice determinants, the best models for all three temperature statistics included site as a fixed effect, and a zero-centered date as well as the corresponding Naxos weather station statistic (daily mean, maximum, or minimum temperature respectively) as covariates. The GAMMs also included an autoregressive structure (corAR1) to account for the interactive effects of site, model (HOBO data logger probe ID), and date. Spatiotemporal autocorrelation, reported as φ, varied between models: for mean T_e, $\varphi = 0.58$, for max T_e, $\varphi = 0.63$, and for min T_e, $\varphi = 0.57$. Including the autoregressive corAR1 structure allowed for a reduction in autocorrelation as evidenced by a reduction in significant linearity in autocorrelative function (acf) plots. Diagnostic graphs (Q-Q and residual plots) were used to verify that there were no significant departures from normality and homogeneity of variance in the models.

Data deposition

Data available from the Dryad Digital Repository: <http://dx.doi.org/10.5061/dryad.kp140> (Belasen et al. 2016).

Results

Variation in microclimate

Site significantly predicted mean T_e, with the study areas clustered into two distinct groups (GAMM, $R^2 = 0.302$, site effect: $F_4 = 17.01$, $p = 1.92 \times 10^{-12}$; Fig. 3): low elevation sites (NLE, ASP and OVR) were significantly warmer than high elevation sites (NSF and NNF) (Tukey's HSD $p < 0.05$ between sites from different clusters). Minimum temperature was similarly different among sites with two clusters, but the low elevation islet OVR clustered with high elevation sites in this case (GAMM, $R^2 = 0.814$, site effect: $F_4 = 29.98$, $p < 2 \times 10^{-16}$; Tukey's HSD, $p < 0.05$ between sites from different clusters; Fig. 3). No significant differences were detected across sites for maximum temperatures (GAMM, $R^2 = 0.144$, site effect: $F_4 = 6.028$, $p < 0.001$; Tukey's HSD > 0.05 for all pairwise comparisons).

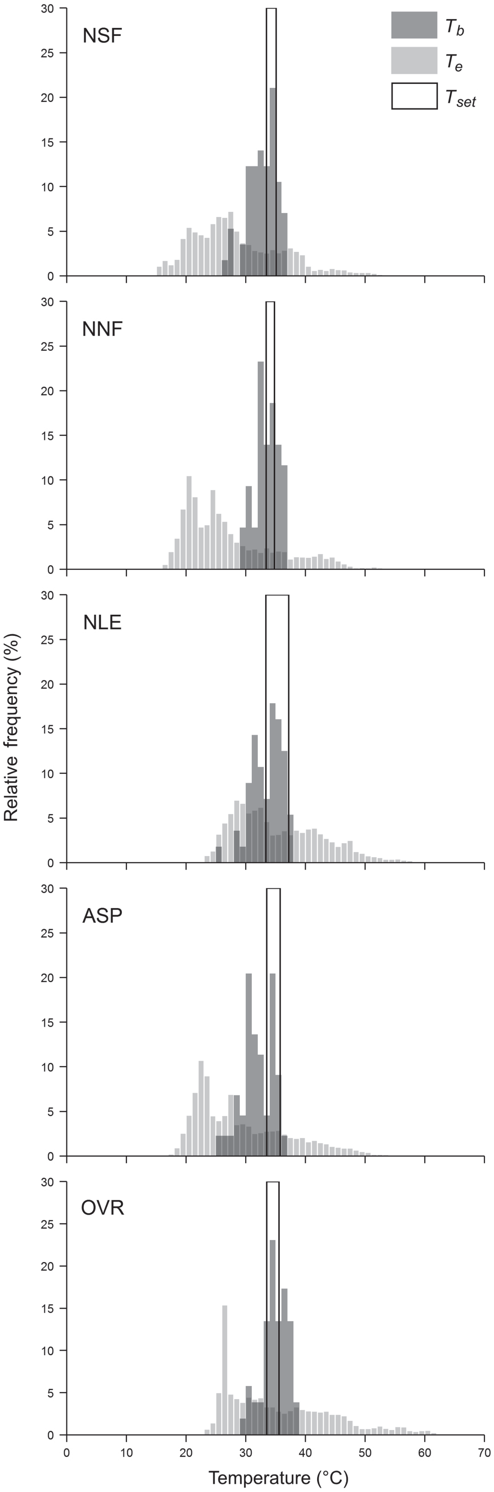

Figure 3. Thermal biology parameters measured at each of the five study sites. Field body temperatures (T_b) of lizards were measured within 20 s of field capture. Preferred temperatures (T_p) were estimated by taking the average of temperatures measured every two minutes for one hour while lizards were allowed to thermoregulate in a corridor containing a thermal gradient, and the set-point range (T_{set}) was calculated as the 25th – 75th quartile of T_p. Operative temperatures (T_e) were measured by data loggers connected to lizard models placed in thermoregulatory sites used by lizards at each study site. We note that T_e values were not collected over the same time period as T_b, and only daytime T_es were included in analyses.

Field and preferred body temperatures

We assessed mean T_b across sites using a GLM approach that included study site, sex of lizard (M/F), and activity (foraging, basking or hiding) as fixed effects, and lizard SVL (cm) and $T_{substrate}$ (°C) as covariates (Fig. 3). Variance of T_b across populations was assessed using Levene's test. Both site and activity were highly significant predictors of mean T_b (GLM, Site: $F_{4,154} = 6.209$, $p < 0.001$; Activity: $F_{2,154} = 5.686$, $p = 0.004$, Table 1). Post hoc pairwise comparisons revealed significantly higher marginal mean T_b in OVR lizards than ASP or NLE (Bonferroni-corrected post hoc mean comparisons: $p = 0.004$ and $p < 0.001$, respectively). Post hoc tests also revealed that foraging is associated with significantly increased T_b relative to basking or hiding (Bonferroni-corrected post hoc mean comparisons: $p < 0.001$ for both pairwise comparisons). Variance of T_b did not differ across populations (Levene's test, $p > 0.05$).

Since no substantial association was found between preferred temperature and body size in simple linear regressions (T_p vs SVL: $\beta = -0.074$, $p > 0.1$, $R^2 = 0.006$; T_p versus mass: $\beta = 0.012$, $p > 0.5$, $R^2 = 0.0001$), size was not included as a covariate in statistical analyses of T_p. Across populations, T_p did not vary in mean (ANOVA, $p = 0.705$) or variance (Levene's test, $p > 0.05$; Fig. 3). Given these results, data were then pooled across the study sites to compare males against independently sampled non-gravid and gravid females. We found no significant difference between non-gravid females and males (Kruskal−Wallis, $p > 0.05$) and consequently pooled these together to increase the sample size of our groups and to examine whether gravid females exhibit different preferred temperatures relative to the remaining population. Gravid females preferred significantly lower temperatures (− 1.18°C) on average than males and non-gravid females (independent samples t-test, $t_{174} = 2.795$, $p = 0.006$). T_p in females that were gravid upon capture were re-measured following oviposition. A comparison indicated that females preferred significantly lower temperatures (− 1.19°C) while gravid (paired samples t-test, $t_{38} = -3.346$, $p = 0.002$).

Effectiveness of thermoregulation

No differences were detected in thermoregulatory accuracy ($\overline{d_b}$) between the sexes (independent samples t-test, $t_{249} = 1.51$, $p > 0.05$) or across sites (ANOVA, $F_{4,163} = 2.430$, $p = 0.05$). However, thermal quality varied significantly across sites (ANOVA, $F_{4,\ 79.38} = 20.987$, $p < 0.0001$), with Naxos high elevation site (NNF) exhibiting significantly lower quality (higher $\overline{d_e}$) than all other sites, followed by NSF and ASP that had similar mean de, while NLE showed the higher thermal quality (Table 2). The effort among all sites was constrained to a narrow range near one (all sites ranged from 0.73–0.88), suggesting that lizards from all populations are effective thermoregulators.

Table 1. Values for the thermal parameters and water loss rates. Operative (T_e), body (T_b) and preferred temperatures (T_p), critical thermal maximum (CT_{max}), critical thermal minimum (CT_{min}) and evaporative water loss (EWL): means ± standard deviation. Sample size in parenthesis. For T_b, a mean for both non-gravid and gravid females is reported due to the difficulty in distinguishing reproductive status in the field. EWL is the only trait that varies among sites, while the others are markedly consistent among populations.

	T_e	T_b	T_p	CT_{max}	CT_{min}	EWL
Site						
NLE	25.27 ± 1.55 (5)	33.47 ± 0.83 (55)	34.35 ± 0.23 (29)	43.97 ± 0.37 (12)	11.51 ± 0.54 (12)	0.32 ± 0.14 (10)
NSF	21.37 ± 1.76 (5)	33.87 ± 0.77 (57)	34.17 ± 0.31 (28)	44.39 ± 0.37 (14)	12.61 ± 0.61 (11)	0.14 ± 0.17 (9)
NNF	21.74 ± 1.62 (4)	34.09 ± 1.23 (43)	34.23 ± 0.26 (22)	44.09 ± 0.45 (8)	12.62 ± 0.59 (10)	0.69 ± 0.21 (8)
OVR	26.52 ± 1.79 (4)	34.46 ± 0.64 (52)	34.37 ± 0.28 (23)	43.75 ± 0.41 (10)	12.21 ± 0.79 (6)	–
ASP	26.43 ± 1.35 (7)	32.74 ± 0.88 (44)	34.64 ± 0.22 (35)	43.76 ± 0.41 (10)	12.57 ± 0.56 (11)	0.31 ± 0.14 (9)
Sex						
M	–					
F_{ng}	–	33.79 ± 0.20 (138)	34.52 ± 0.31 (51)			
F_g	–	33.19 ± 0.24 (112)	34.65 ± 0.35 (26)			
Activity			33.92 ± 0.49 (60)			
Basking	–	33.33 ± 0.41 (124)				
Foraging	–	34.67 ± 0.55 (65)				
Hiding	–	32.47 ± 0.85 (47)				

The low thermal qualities (high mean d_e) at both Naxos high elevation sites were not equally reflected in thermoregulations effectiveness: NNF received the highest value (E = 0.88) while NSF had a lower one (E = 0.81) (Table 2). The alternative index (Blouin-Demers and Weatherhead 2001) highlighted the more effective thermoregulation in the two high elevation Naxos sites due to higher $\overline{d_e} - \overline{d_b}$ values (Table 2).

Critical thermal tolerance

GLMs were used to compare CT_{max} and CT_{min} across populations. For these analyses, SVL was included as the covariate, and site as a fixed effect. There were no significant differences in CT_{max} mean (GLM, $F_{5,48} = 2.922$, $p > 0.05$) or variance (Levene's test, $p > 0.05$) across sites. A significant association was detected between CT_{max} and SVL (GLM, $F_{1,48} = 2.059$, $p < 0.05$), with smaller lizards exhibiting higher CT_{max} values. For CT_{min} there were no significant associations with SVL (GLM, $F_{1,44} = 2.161$, $p > 0.1$) nor any significant differences between sites (GLM, $F_{5,44} = 2.338$, $p > 0.05$; Levene's test, $p > 0.05$). Marginally non-significant differences in CT_{min} between populations contributed to a significant site effect in the GLM ($F_{4,44} = 2.730$, $p = 0.041$; Tukey's HSD, $p = 0.09$ for NLE versus NNF and $p = 0.08$ for NLE versus ASP).

Table 2. Thermoregulatory indices (Hertz et al. 1993, Blouin-Demers and Weatherhead 2001). Means ± standard deviation are reported for three indices: deviation of operative temperatures from the preferred temperatures ($\overline{d_e}$), deviation of body temperatures from preferred temperatures, ($\overline{d_b}$) and effectiveness of thermoregulation (E). Both indices indicate differences in thermoregulatory effort among populations.

Site	$\overline{d_e}$	$\overline{d_b}$	E	$\overline{d_e} - \overline{d_b}$
NLE	5.30 ± 0.14	1.03 ± 0.21	0.80	4.27
NSF	7.39 ± 0.17	1.39 ± 0.22	0.81	6
NNF	8.52 ± 0.16	1.05 ± 0.16	0.88	7.47
OVR	6.76 ± 0.17	0.87 ± 0.14	0.87	5.89
ASP	7.48 ± 0.18	1.97 ± 0.31	0.73	5.51

Water loss rates: variation and heritability

Male EWL data were analyzed using a GLM, with site as a fixed effect and mean trial temperature as a covariate. We detected a significant effect of site on mean EWL (Fig. 3, GLM, $F_{3.31} = 7.188$, $p < 0.001$), while no difference was detected in variance of EWL across sites (Levene's test, $p > 0.05$). Post hoc tests showed that the NNF population had the highest EWL rates across the four sites, while the NSF population had the lowest (Tukey's HSD, $p < 0.05$). The NLE and ASP populations were not significantly different. NLE lizards also exhibited significantly higher EWL rates than those of NSF lizards. Soil moisture data from the three Naxos sites (NNF, NSF, NLE) revealed a parallel pattern of differences (Kruskal−Wallis, $H_2 = 11.431$ $p = 0.003$, Fig. 4), with NSF having the lowest average soil moisture (0.06 g ± 0.05 SE), NLE having slightly higher soil moisture (0.14 g ± 0.05 SE), though the difference was not

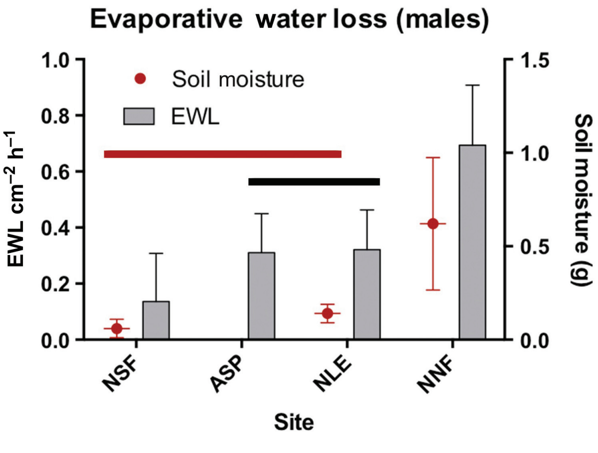

Figure 4. Evaporative water loss (black, left y-axis) and soil moisture (red, right y-axis) across sites. Bars represent marginal means (± 2 SE). Bars connected by the black line are not significantly different at α = 0.05 in the GLM analysis. The OVR population was omitted from the EWL experiment because weather conditions precluded additional travel to this islet, and ASP was omitted from soil collections because travel was no longer permitted to this site following archeological excavation.

significant, and NNF having the highest (0.62 g ± 0.39 SE; Mann−Whitney U, $U = 25.00$, $p < 0.01$ for both significant pairwise comparisons).

EWL measurements for females showed no significant differences between sampling sites (Kruskal–Wallis, $H_2 = 1.895$, $p > 0.05$). Consequently they were pooled for a parent–offspring analysis testing for broad-sense heritability of EWL. Juvenile EWLs were averaged within each clutch and a linear regression weighted according to clutch size (ranging from 1–3 offspring) showed a marginally non-significant relationship between mother and offspring EWL ($h^2 = 0.36$, $p = 0.059$, $R^2 = 0.248$, $n = 29$ juveniles across 15 clutches).

Discussion

In this study, we compared isolated insular lizard populations to investigate how environmental factors and population fragmentation may shape certain ecophysiological features. First, by comparing the thermal parameters of populations from a common genetic background living in different microclimates, we investigated to what extent prevailing conditions shape an organism's thermal biology. Furthermore, building on recent research (Hurston et al. 2009) that has shown that the small islet populations of *Podarcis erhardii* have greatly reduced genetic diversity relative to the large Naxos population, we tested whether population size and isolation affect the mean values and variability of ecophysiological traits, including thermoregulation and water loss.

In this study, we use variance in thermal biology as a proxy for population variability. Contrary to our expectations, variance in the measured traits did not decline in smaller populations. However, we found that microclimate differs predictably among study sites based on site elevation, aspect and insularity. Although T_e differed according to elevation, these microclimatic differences did not appear to correlate with shifts in T_b across sites (Table 1, Fig. 3). This result departs from previous studies demonstrating that the range of T_e influences the temperatures that animals achieve in the field (Díaz 1997, Adamopoulou and Valakos 2005).

Previous studies have indicated that T_{set} or T_p is affected by season, sex, age, reproduction, and body size (Andrews et al. 1999, Carretero et al. 2005, Carretero 2008, Veríssimo and Carretero 2009, Sagonas et al. 2013b). In this study, though sites differed considerably among the study populations in terms of elevation, thermal quality, and moisture, no impact was observed on T_p. This result contrasts with other studies that reported habitat-induced differences in thermal preferences (Sinervo and Adolph 1989, Van Damme et al. 1989, Sagonas et al. 2013a). Although sex did not contribute to differences in T_p, reproductive status did. Gravid females selected significantly lower temperatures than either males or non-gravid females (− 1.19°C; Table 1). A proposed explanation for this phenomenon is temperature sensitivity during embryonic development, as improper thermoregulation during gestation can have lasting negative impacts on offspring fitness (Burger 1989, Sinervo and Adolph 1989, Mathies and Andrews 1997, Wapstra 2000, Paranjpe et al. 2013). Preference for lower temperatures in gravid females therefore may reduce recruitment and population stability in the face of climate warming.

The overall conservativeness of T_b and T_p among the study populations contributed to relatively high thermoregulatory accuracy in all Naxos sites, though a striking deviation occurred in the two islet populations. Aspronissi lizards displayed the lowest accuracy ($\overline{d_b} = 1.97$) among all examined populations, whereas the nearby Ovriokastro population exhibited the highest ($\overline{d_b} = 0.87$). This discrepancy can be attributed to lower T_bs on ASP relative to other populations (Table 1). The very poor thermal quality on ASP apparently prevents the lizards from achieving T_bs within their set-point range.

Theory predicts that mean d_e describes relative thermal quality of microhabitats (Hertz et al. 1993). The most thermally challenging sites (the ones with the highest mean d_e) were NNF, NSF and ASP (Table 2). The low thermal quality of the high elevation sites (NSF and NNF) is unsurprising. Though high elevation sites are exposed to strong winds and higher precipitation levels (Ortega et al. 2016), driving T_e below T_p and thus inflating $\overline{d_e}$. However the high $\overline{d_e}$ of ASP, coupled with the relatively high value of OVR, suggests that altitude is not the only parameter affecting the thermal quality of a habitat. ASP is a very small islet that is very exposed to the typical strong winds of the NE Aegean during the summer season. The highest thermal quality was detected in the Naxos low elevation site (NLE), a site that combines protection from extreme climatic effects and high availability of thermal shelters.

All populations demonstrated high thermoregulatory effectiveness with E-values varying within a narrow range (0.73–0.88, Table 2). Lizards from all sites appear to be effective thermoregulators and thus are able to behaviorally buffer physiological performance (Angilletta 2001). Ectotherms that live in challenging environments are expected to put more effort into thermoregulation (Hertz et al. 1993). According to our results, NNF and OVR exceed all other sites with almost identical values (0.88 and 0.87 respectively), despite their quite different $\overline{d_e}$ values. Though NSF and ASP had lower thermal quality than OVR, they both achieved lower E-values (Table 2). The lizards on the small islet ASP appear to perform the least effective thermoregulation among all populations ($E = 0.73$).

As disparate $\overline{d_e}$ and $\overline{d_b}$ combinations may result in the same E-values, this can mask the true thermoregulatory profile (Hertz et al. 1993). A complementary approach (sensu Blouin-Demers and Weatherhead 2001) determining the thermoregulatory extent based on the difference between $\overline{d_e}$ and $\overline{d_b}$, provides a different view of thermoregulatory effectiveness in our study populations: NNF and NSF achieved the highest values (and thus demonstrated more effective thermoregulation) while OVR and ASP follow with quite similar values (Table 2). The lowest thermoregulatory effectiveness (smaller $\overline{d_e} - \overline{d_b}$) was observed in NLE, the site with the highest thermal quality. In other words, all populations obey the general rule predicting that lizards living in demanding habitats exert more thermoregulatory effort to achieve T_b-values close to their preferred temperatures (Hertz et al. 1993, Blouin-Demers and Nadeau 2005, Sagonas et al. 2013b).

CT_{max} and CT_{min} did not differ across populations, despite differences in the thermal environment as indicated by T_e (Table 1). While this finding departs from other studies (Leal and Gunderson 2012, Brusch et al. 2016), it may be the result of the overall conservativeness of these traits. Alternatively, the lack of differences among populations may be attributable to low selection pressure resulting from the buffering effects of active behavioral thermoregulation in *P. erhardii*. Our results indicate a negative relationship between CT_{max} and lizard size, suggesting that heat tolerance declines in larger, likely older lizards, perhaps relating to age-related senescence.

We detected significant differences in mean EWL across populations, with increases in EWL rates paralleling rising soil moisture across sites (Table 1, Fig. 4). This suggests that differences in microclimate can shape physiological water flux, which is a critical component of ectotherm biology (Lorenzon et al. 1999). We detected the highest EWL rates in the population at the mesic NNF site. While this pattern mirrors results from other studies (Dmi'el et al. 1997, Gunderson et al. 2011, Carneiro et al. 2015, 2016), it is not clear whether these intraspecific differences in EWL are genetically determined or the result of labile adaptation to local conditions. Our mother–offspring data suggest heritability of these differences. However, because this second EWL experiment was performed on dam–progeny pairs, the results remain confounded by both maternal effects and dominance variation and as such reflect broad-sense heritability (Lynch and Walsh 1998). These limitations notwithstanding, our results are indicative of at least a partial genetically inherited component to this trait. Deeper genetic studies of *P. erhardii* population structure on Naxos coupled with similar ecophysiological experiments would lend insight into genetic contributions to these phenotypic differences.

Climate in the Aegean Sea is expected to become both warmer and drier (Giorgi and Lionello 2008). Given the differences exhibited in EWL across our study sites, susceptibility to future aridification likely varies across populations. Regardless of the mechanisms involved, it appears that site characteristics that relate to moisture availability have an important effect on reptile physiology. This dovetails with paleoecological reconstructions of reptile extinctions on these islands, which appear to have been driven by increasing Holocene desiccation (Foufopoulos et al. 2011). Future studies aiming to assess reptile vulnerability to climate change therefore should consider the interactions between temperature and habitat moisture, and their effects on reptile water physiology and thermal biology.

Our results did not indicate a direct effect of population size reduction (a proxy for genetic impoverishment) on the thermal ecology of *P. erhardii*, as the islet populations did not exhibit reduced variability. This suggests that genetic impoverishment may not be as important in shaping thermal ecology of a population as other mechanisms. This result may be explained by a conservative genetic infrastructure and resistance to directional selection (Hertz and Nevo 1981, but see Scheers and Van Damme 2002), underlying physiological constraints on thermal biology, or strong mediating effects of behavioral plasticity that effectively eliminate selection for physiological adaptation (Gunderson and Stillman 2015). This is related to the 'Bogert effect,' whereby ectotherms with limited behavioral thermoregulation may compensate with higher plasticity in thermal tolerances (Bogert 1949). Since our results indicate relatively high thermoregulatory effort among populations in different microclimates while thermal preferences remain consistent, we posit that the mechanism underlying this common thermal profile may involve more than physiological plasticity.

Regardless of the consistency in thermal preferences among populations, genetic impoverishment in very small populations is still likely to impact evolutionary potential, as it ultimately reduces the variability on which selection can act (Willi et al. 2006). This would be particularly relevant for the Aspronissi (ASP) population, which consists only of a few hundred individuals. Genetic impoverishment combined with a lack of historic selection pressure may also result in canalization, or a decline in the propensity for plasticity (Via et al. 1995, Aubret and Shine 2010), which can have important implications for future adaptation to climate shifts.

Overall, most aspects of *P. erhardii* thermal ecology appear to be relatively rigid among populations, despite environmental differences among habitats and genetic differences among populations. The close reliance on ambient temperatures exhibited by ectotherms (Meiri et al. 2013) renders these animals particularly susceptible to changes in climate (Kearney et al. 2009, Buckley et al. 2015). The impacts are likely to be especially severe for ectotherms living in hot, arid systems, as the margin or 'buffer zone' between environmental temperature and the CT_{max} reaches a critical minimum (Sinervo et al. 2010, Gunderson and Leal 2012). Our data suggest that aridification due to climate change may be another critical factor to be considered in the fate of ectotherm populations. In summary, our findings indicate that changes in habitat suitability driven by climate have been and will continue to be significant in the survival of native species in the area (Foufopoulos et al. 2011).

Acknowledgements – The authors would like to acknowledge R. Lara for assistance in calculating the thermoregulation index *E*; G. Kapsalas and M. McCormick for assistance with figure design; and T. Y. James for valuable comments on the manuscript.
Funding – The authors would like to acknowledge funding support from the School of Natural Resources and Environment and Rackham Graduate School at the University of Michigan.
Permits – All work was carried out in accordance with the Hellenic National Law (Presidential Decree 67/81) on the humane use of animals, and the University of Michigan Committee on Care and Use of Animals (UCUCA, Protocol no. 10551-1).

References

Ackley, J.W. et al. 2015. Urban heat island mitigation strategies and lizard thermal ecology: landscaping can quadruple potential activity time in an arid city. – Urban Ecosyst. 18: 1447–1459.

Adamopoulou, C. and Valakos, E. D. 2005. Thermal ecology and activity cycle of *Podarcis milensis* in a sandy coastal area. – Isr. J. Zool. 51: 39–52.

Adamopoulou C. et al. 1999. Diet composition of *Podarcis milensis*, *Podarcis gaigeae* and *Podarcis erhardii* (*Sauria: Lacertidae*) during summer. – Bonn Zool. Beitr. 48: 275–282.

Andrews, R. M. et al. 1999. Field and selected body temperatures of the lizards *Sceloporus aeneus* and *Sceloporus bicanthalis*. – J. Herpetol. 33: 93–100.

Angilletta, M. J. J. 2001. Thermal and physiological constraints on energy assimilation in a widespread lizard (*Sceloporus undulatus*) . – Ecology 82: 3044–3056.

Araújo, M. B. et al. 2013. Heat freezes niche evolution. – Ecol. Lett. 16: 1206–1219.

Aubret, F. and Shine, R. 2010. Fitness costs may explain the post-colonisation erosion of phenotypic plasticity. – J. Exp. Biol. 213: 735–739.

Belasen, A. et al. 2016. Data from: Fine with heat, problems with water: microclimate alters water loss in a thermally adapted insular lizards. – Dryad Digital Repository, <http://dx.doi.org/10.5061/dryad.kp140>.

Blouin-Demers, G. and Weatherhead, P. J. 2001. Thermal ecology of black rat snakes (*Elaphe obsoleta*) in a thermally challenging environment. – Ecology 82: 3025–3043.

Blouin-Demers, G. and Nadeau, P. 2005. The cost–benefit model of thermoregulation does not predict lizard thermoregulatory behavior. – Ecology 86: 560–566.

Bogert, C. M. 1949. Thermoregulation in reptiles, a factor in evolution. – Evolution 3: 195–211.

Brock, K. M. et al. 2014. Novel records of frugivory and ovophagy in *Podarcis* lizards from east Mediterranean islands. – N.-W. J. Zool. 10: 223–225.

Brusch, G. A. et al. 2016. Turn up the heat: thermal tolerance of lizards at La Selva, Costa Rica. – Oecologia 180: 325–334.

Buckley L. B. et al. 2015. Thermoregulatory behaviour limits local adaptation of thermal niches and confers sensitivity to climate change. – Funct. Ecol. 29: 1038–1047.

Burger, J. 1989. Incubation temperature has long-term effects on behavior of young pine snakes (*Pituophis melanoleucus*). – Behav. Ecol. Sociobiol. 24: 201–207.

Carneiro, D. et al. 2015. Comparing ecophysiology traits in two *Podarcis* wall lizards with overlapping ranges. – Salamandra 51: 335–344.

Carneiro, D. et al. 2016. More than temperature. Is ecophysiology congruent with the present-day relictual distribution of a lizard group? – Herpetol. J. in press.

Carretero, M. A. 2008. Preferred temperatures of *Tarentola mauritanica* in spring. – Acta Herpetol. 3: 57–64

Carretero, M. A. et al. 2005. Variation in preferred body temperature in an oviparous population of *Lacerta* (*Zootoca*) *vivipara*. – Herpetol. J. 15: 51–55.

Ceia-Hasse, A. et al. 2014. Integrating ecophysiological models into species distribution projections of European reptile range shifts in response to climate change. – Ecography 37: 679–688.

Davis, M. B. and Shaw, R. G. 2001. Range shifts and adaptive responses to quaternary climate change. – Science 292: 673–679.

Deem, V. and Hedman, H. 2014. Potential cannibalism and intraspecific tail autotomization in the Aegean wall lizard, *Podarcis erhardii*. – Hyla 2014: 33–34.

Díaz, J. A. 1997. Ecological correlates of the thermal quality of an ectotherm's habitat: a comparison between two temperate lizard populations. – Funct. Ecol. 11: 79–89.

Dmi'el, R. et al. 1997. Evaporative water loss in nine insular populations of the lizard *Anolis cristatellus* group in the British Virgin Islands. – Biotropica 29: 111–116.

Dzialowski, E. M. 2005 Use of operative and standard operative temperature models in thermal biology. – J. Therm. Biol. 30: 317–334.

Ehlers, A. et al. 2008. Importance of genetic diversity in eelgrass *Zostera marina* for its resilience to global warming. – Mar. Ecol. Progr. Ser. 355: 1–7.

Eldridge, M. D. B. et al. 1999. Unprecedented low levels of genetic variation and inbreeding depression in an island population of the black-footed rock-wallaby. – Conserv. Biol. 13: 1–54.

England, P. R. et al. 2003. Effect of intense versus diffuse population bottlenecks on microsatellite genetic diversity and evolutionary potential. – Conserv. Genet. 4: 595–604.

Font, E. et al. 2009. Ultraviolet reflectance and cryptic sexual dichromatism in the ocellated lizard, *Lacerta* (Timon) *lepida* (Squamata: Lacertidae). – Biol. J. Linn. Soc. 97: 766–780.

Foufopoulos, J. and Ives, A. R. 1999. Reptile extinctions on land-bridge islands: life-history attributes and vulnerability to extinction. – Am. Nat. 153: 1–25.

Foufopoulos, J. et al. 2011. Climate change and elevated extinction rates of reptiles from Mediterranean islands. – Am. Nat. 177: 119–129.

Frankham, R. 1997. Do island populations have less genetic variation than mainland populations? – Heredity 78: 311–318.

Frankham, R. et al. 2002. Introduction to conservation genetics. – Cambridge Univ. Press.

Giorgi, F. and Lionello, P. 2008. Climate change projections for the Mediterranean region. – Global Planet. Change 63: 90–104.

Gunderson, A. R. and Leal, M. 2012. Geographic variation in vulnerability to climate warming in a tropical Caribbean lizard. – Funct. Ecol. 26: 783–793.

Gunderson, A. R. and Stillman, J. H. 2015. Plasticity in thermal tolerance has limited potential to buffer ectotherms from global warming. – Proc. R. Soc. B 282: 20150401.

Gunderson, A. R. et al. 2011. Tests of the contribution of acclimation to geographic variation in water loss rates of the West Indian lizard *Anolis cristatellus*. – J. Comp. Physiol. B 181: 965–972.

Hertz, P. E. and Nevo, E. 1981. Thermal biology of four Israeli agamid lizards in early summer. – Isr. J. Zool. 30: 190–210.

Hertz, P. E. et al. 1993. Evaluation temperature regulation by field-active ectotherms: the fallacy of the inappropriate question. – Am. Nat. 142: 796–818.

Hinten, G. et al. 2003. Genetic variation and island biogeography: microsatellite and mitochondrial DNA variation in island populations of the Australian bush rat, *Rattus fuscipes greyii*. – Conserv. Genet. 4: 759–778.

Huey, R. B. and Stevenson, R. D. 1979. Integrating thermal physiology and ecology of ectotherms: a discussion of approaches. – Am. Zool. 19: 357–366.

Hurston, H. et al. 2009. Effects of fragmentation on genetic diversity in island populations of the Aegean wall lizard *Podarcis erhardii* (Lacertidae, Reptilia). – Mol. Phylogenet. Evol. 52: 395–405.

Kearney, M. et al. 2009. The potential for behavioral thermoregulation to buffer "cold-blooded" animals against climate warming. – Proc. Natl Acad. Sci. USA 106: 3835–3840.

Kougioumoutzis, K. et al. 2014. Network biogeographical analysis of the central Aegean archipelago. – J. Biogeogr. 41: 1848–1858.

Kour, E. L. and Hutchison, V. H. 1970. Critical thermal tolerances and heating and cooling rates of lizards from diverse habitats. – Copeia 1970: 219–229.

Kubisch, E. L. et al. 2016a. Local extinction risk of three species of lizard from Patagonia as a result of global warming. – Can. J. Zool. 94: 49–59.

Kubisch, E. L. et al. 2016b. Vulnerability to climate warming of *Liolaemus pictus* (Squamata, Liolaemidae), a lizard from the cold temperate climate in Patagonia, Argentina. – J. Comp. Physiol. B 186: 243–253.

Lara-Resendiz, R. A. et al. 2015. Thermoregulation of two sympatric species of horned lizards in the Chihuahuan Desert and their local extinction risk. – J. Therm. Biol. 48: 1–10.

Leal, M. and Gunderson, A. R. 2012. Rapid change in the thermal tolerance of a tropical lizard. – Am. Nat. 180: 815–822.

Lorenzon, P. et al. 1999. Effect of water constraint on growth rate, activity and body temperature of yearling common lizard (*Lacerta vivipara*). – Oecologia 118: 423–430.

Losos, J. B. and Ricklefs, R. E. 2009. Adaptation and diversification on islands. – Nature 457: 830–836.

Lymberakis, P. et al. 2009. *Podarcis erhardii*. – The IUCN red list of threatened species 2009: e.T61546A12512784.

Lynch, M. and Walsh, B. 1998. Genetics and analysis of quantitative traits. – Sinauer.

Mathies, T. and Andrews, R. M. 1997. Influence of pregnancy on the thermal biology of the lizard, *Sceloporus jarrovi*: why do pregnant females exhibit low body temperatures? – Funct. Ecol. 11: 498–507.

Meiri, S. et al. 2013. Are lizards feeling the heat? A tale of ecology and evolution under two temperatures. – Global Ecol. Biogeogr. 22: 834–845.

Newmark, W. D. 1995. Extinction of mammal populations in western North American national parks. – Conserv. Biol. 9: 512–526.

Opdam, P. and Wascher, D. 2003. Climate change meets habitat fragmentation: linking landscape and biogeographical scale levels in research and conservation. – Biol. Conserv. 117: 285–297.

Ortega, Z. et al. 2016. The peak of thermoregulation effectiveness: thermal biology of the Pyrenean rock lizard, *Iberolacerta bonnali* (Squamata, Lacertidae). – J. Therm. Biol. 56: 77–83.

Paranjpe, D. A. et al. 2012. Measuring thermal profile of reptiles in laboratory and field. – In: Spink, A. J. et al. (eds), Proceedings of measuring behavior, Utrecht, the Netherlands, August 28–31, 2012, pp. 460–462.

Paranjpe, D. A. et al. 2013. Evidence of maternal effects on temperature preference in side-blotched lizards: implication for evolutionary response to climate change. – Ecol. Evol. 3: 1977–1991.

Parmesan, C. 2006. Ecological and evolutionary responses to recent climate change. – Annu. Rev. Ecol. Evol. Syst. 37: 637–669.

Peterson, C. R. et al. 1993. Snake thermal ecology: the causes and consequences of body temperature variation. – In: Seigel, R. A. and Collins, J. T. (eds), Snakes: ecology and behavior. McGraw-Hill, pp. 241–314.

Pereira, H. M. et al. 2010. Scenarios for global biodiversity in the 21st Century. – Science 330: 1496–1501.

Perissoratis, C. and Conispoliatis, N. 2003. The impacts of sea-level changes during latest Pleistocene and Holocene times on the morphology of the Ionian and Aegean seas (SE Alpine Europe). – Mar. Geol. 196: 146–156.

Remon, N. et al. 2011. Chronicle of an extinction foretold: properties of an extremely small population of *Iberolacerta monticola*. – Conserv. Genet. 13: 131–142.

Sagonas, K. et al. 2013a. The impact of insularity on the thermoregulation of a Mediterranean lizard. – J. Therm. Biol. 38: 480–486.

Sagonas, K. et al. 2013b. The effect of body size on the thermoregulation of lizards on hot, dry Mediterranean islands. – J. Therm. Biol. 38: 92–97.

Scheers, H. and Van Damme, R. 2002. Micro-scale differences in thermal habitat quality and a possible case of evolutionary flexibility in the thermal physiology of lacertid lizards. – Oecolohia 132: 323–331.

Sinervo, B. and Adolph, S. C. 1989. Thermal sensitivity of growth rate in hatchling *Sceloporus* lizards: environmental, behavioral and genetic aspects. – Oecologia 78: 411–419.

Sinervo, B. et al. 2010. Erosion of lizard diversity by climate change and altered thermal niches. – Science 328: 894–899.

Stork, N. E. at al. 2009. Vulnerability and resilience of tropical forest species to land-use change. – Conserv. Biol. 23: 1438–1447.

Tosini, G. et al. 1994. Effects of feeding on set point temperatures and thermoregulatory behaviour in the lizards. – Amphibia-Reptilia 15: 257–265.

Valakos, E. D. 1990. The ecology of the lizard *Podarcis erhardii* (Bedriaga, 1882) (Sauria: Lacertidae) in a typical insular ecosystem on Naxos Isl. – PhD thesis, Univ. of Athens.

Valakos, E. D. et al. 2008. The amphibians and reptiles of Greece. – Chimaira.

Van Damme, R. et al. 1986. Selected body temperatures in the lizards *Lacerta vivipara*: variation within and between populations. – J. Therm. Biol. 11: 219–222.

Van Damme, R. et al. 1989. Altitudinal variation of the thermal biology and running performance in the lizard *Podarcis tiliguerta*. – Oecologia 80: 516–524.

Verissimo, C. V. and Carretero, M. A. 2009. Preferred temperatures of *Podarcis vaucheri* from Morocco: intraspecific variation and interspecific comparisons. – Amphibia-Reptilia 30: 17–23.

Via, S. et al. 1995. Adaptive phenotypic plasticity: consensus and controversy. – Trends Ecol. Evol. 10: 212–217.

Wapstra, E. 2000. Maternal basking opportunity affects juvenile phenotype in a viviparous lizard. – Funct. Ecol. 14: 345–352.

Willi, Y. et al. 2006. Limits to the adaptive potential of small populations. – Annu. Rev. Ecol. Evol. Syst. 37: 433–458.