



# Resilience to extreme temperature events: acclimation capacity and body condition of a polymorphic fish in response to thermal stress

ZACHARY W. CULUMBER<sup>1,2\*</sup> and SCOTT MONKS<sup>1</sup>

<sup>1</sup>*Centro de Investigaciones Biologicas, Universidad Autónoma del Estado de Hidalgo, Pachuca, Hidalgo 42001 Mexico*

<sup>2</sup>*Centro de Investigaciones de las Huastecas Aguazarca, 16 de Septiembre, Aguazarca, Calnali, Hidalgo 392, Mexico*

Received 20 September 2013; revised 30 October 2013; accepted for publication 1 November 2013

Considerable attention has been given to the potential impacts of global climate change on biodiversity. In the present study, we combine understudied themes by examining the ability of a freshwater fish (polymorphic for heat-sensitivity) to respond to short-term thermal stress mimicking an extreme temperature event. We simultaneously measured the effect of thermal stress on the body condition of heat-sensitive and heat-tolerant forms to evaluate an existing hypothesis regarding the underlying mechanism by which temperature affects the maintenance of genetic variation in this species. Surprisingly, the heat-sensitive allelic variant increased in body condition equally as much as a heat-tolerant variant under acute heat stress. More importantly, the heat-sensitive variant exhibited a significant response to thermal stress, with an upward shift of greater than 2 °C in critical thermal maximum. Our findings suggest a complexity to the relationship between thermal stress and male body condition that may depend on an interaction with other factors such as resource level. Although the evolutionary fate of species with respect to climate change is typically evaluated in terms long-term adaptive response, short-term selection events could drastically reduce fitness and reduce evolutionary potential. Our results suggest that heat-sensitive species may have considerably greater resilience to the short-term, extreme perturbations to the environment that are expected under climate change. © 2014 The Linnean Society of London, *Biological Journal of the Linnean Society*, 2014, **111**, 504–510.

ADDITIONAL KEYWORDS: climate change – pleiotropy – Poeciliidae – thermal biology – *Xiphophorus*.

## INTRODUCTION

Limits of physiological response to stress induced by the abiotic environment shape species distributions (Lexer *et al.*, 2003; Hampe & Petit, 2005; Sexton *et al.*, 2009). Understanding these limits and their mechanistic bases are central to predicting the response of species to climate change and consequent shifts in their geographical distributions (Somero 2005; Calosi, Bilton & Spicer, 2008; Portner & Farrell, 2008). In particular, thermal tolerance is a key factor limiting species distributions (Addo-bediako, Chown & Gaston, 2000; Culumber *et al.*, 2012). Thermal tolerance is of

particular importance given the changes in temperature that are already observed and predicted to continue under global climate change (Parmesan, 2006; Meehl *et al.*, 2007). Because evolutionary potential depends on the maintenance of genetic variation, selection as a result of climate change that would reduce fitness and consequently genetic variation would reduce the evolutionary potential of natural populations.

Investigation of thermal acclimation capacity and adaptive potential of upper thermal limits has flourished in recent years. However, this work has been almost entirely focused on invertebrates (Deutsch *et al.*, 2008; Jumbam *et al.*, 2008; Burgi & Mills, 2012; Ma & Ma, 2012) and, disproportionately, on drosophilids (Krebs & Feder, 1997; Hoffmann, 2010;

\*Corresponding author. E-mail: zach@cichaz.org

Mitchell, Sgro & Hoffmann, 2011; Kellermann *et al.*, 2012; Santos, Castaneda & Rezende, 2012). The study of freshwater biodiversity in these same contexts has lagged by comparison (Heino, Virkkala & Toivonen, 2009). It has become clear that inferences based on one genus cannot necessarily be generalized to an unrelated group, as demonstrated by the conflicting pattern between thermal tolerance and acclimation capacity in diving beetles and marine animals (Stillman, 2003; Calosi *et al.*, 2008). Most studies focus on functional response to long-term, upward trends in global temperatures (Mitchell & Hoffmann, 2010; Santos *et al.*, 2012; for a review, see Hofmann & Todgham, 2010) or have used long-term temperature trends to predict future distributions of diverse taxa (Peterson *et al.*, 2002; Thomas *et al.*, 2004; Jump & Penuelas, 2005; Perry *et al.*, 2005). The effects and importance of extreme temperature events remain comparatively understudied (McKechnie & Wolf, 2010; Burgi & Mills, 2012; Van Dooremalen, Berg & Eilers, 2013).

Multiple studies concur that the frequency and severity of extreme weather events such as short-term temperature spikes (e.g. so-called 'heat waves') will increase under climate change (Meehl & Tebaldi, 2004; Smith *et al.*, 2009; Hansen, Sato & Ruedy, 2012). These acute events rapidly push species towards their physiological limits causing acute stress and may be particularly efficient opportunities for selection to act. Studies of vertebrates, invertebrates, and plants have all demonstrated the importance of temperature extremes in structuring species distributions and in determining response to climate change (Helmuth & Hofmann, 2001; Ackerly, 2003; Culumber *et al.*, 2012). Indeed, some recent extreme temperature events have exceeded the physiological limits of some species including intertidal invertebrates and coral reefs (Donner, Knutson & Oppenheimer, 2007; Petes, Menge & Murphy, 2007; Harley, 2008). Although such events are rare at present, this emphasizes the potential importance of these events to pressure the acclimatory and adaptive potential of species, particularly those that are heat-sensitive or already near their upper thermal limits (Deutsch *et al.*, 2008). Because the survival and fitness of species in response to short-term events ultimately determines effects on genetic variation and evolutionary potential in response to long-term selection, resilience to extreme temperature events is necessary for evolutionary response to climate change.

The handful of studies that have investigated temperature extremes have focused on invertebrates and have not tested links to functional mechanisms by which extreme temperature events might reduce the fitness of the focal organisms (Allen, Clusella-Trullas & Chown, 2012; Burgi & Mills, 2012; Van Dooremalen *et al.*, 2013). Because direct selection on physiological

traits such as thermal limits represents only one way by which climate change will affect fitness (Parmesan, 2006), studies are needed on a wider range of taxa and that incorporate tests of potential mechanisms underlying organismal performance under stress (Heino *et al.*, 2009; Somero, 2012).

*Xiphophorus variatus* is a livebearing fish inhabiting freshwater streams of the subtropics in eastern Mexico. This species exhibits a genetic polymorphism, as controlled by alleles at a single autosomal locus, that produces pigmented tailspots (Culumber, Bautista-Hernández & Monks, 2014). Pattern alleles are dominant over the wild-type but co-dominant to one another (Borowsky, 1981). Fish homozygous for the wild-type allele (i.e. wild-type fish) have no pattern. Most interesting is that the tailspot types differ in their tolerance to physiological stress as a result of temperature (Culumber *et al.*, 2014). Wild-type fish have a significantly lower critical thermal maximum (CT<sub>max</sub>) than patterned fish collected from the same population (Culumber *et al.*, 2014), although there was no difference among pattern types. This demonstrates the importance of spatial and temporal variation in the abiotic environment (e.g. temperature) in the maintenance of adaptive polymorphism at the tailspot locus. Thus, within single populations, there is genetic variation for heat tolerance that can be assigned to discrete allelic variants. Because wild-type fish have a lower physiological tolerance to high temperatures, we consider them as being relatively 'heat-sensitive' compared to the more 'heat-tolerant' patterned fish.

This difference in thermal limits is consistent with observations of lower wild-type allele frequencies and lower wild-type female fecundity in natural populations during warm conditions (Borowsky, 1984). However, the relative body condition of wild-type males correlated positively with temperature (Borowsky, 1984) in the opposite direction to that expected based on the relatively lower CT<sub>max</sub> of wild-type fish. The effects on body condition could have important fitness consequences that might affect the maintenance of genetic variation, and therefore the evolutionary potential of thermal tolerance. Female livebearers prefer males that are in better condition (Plath *et al.*, 2005; Fisher & Rosenthal, 2006) and females invest more in offspring when mating with males in good condition (Kindsvater *et al.*, 2013). Thus, thermal stress could be an important mechanism of differential reproductive fitness of adult males of different tailspot genotypes.

In the present study, we exposed wild caught *X. variatus* to 5 days of thermal stress. We first aimed to determine the short-term acclimatory capacity of heat-sensitive fish to evaluate resilience to extreme temperature events. Stillman (2003) first reported

that porcelain crabs with lower  $CT_{max}$  have a greater acclimation capacity than those with higher  $CT_{max}$ . However, Calosi *et al.* (2008) showed that the opposite is true in *Deronectes* diving beetles in Europe, where species with a low  $CT_{max}$  have reduced acclimation capacity. Such a reduced acclimation capacity of heat-sensitive species could be particularly detrimental in the context of extreme temperature events that can approach or exceed physiological tolerances. Second, we tested the effect of heat stress on body condition of wild-type and patterned males to test whether previous correlations were driven by temperature itself or potentially by a correlated variable.

## MATERIAL AND METHODS

### COLLECTING AND EXPERIMENTAL SET-UP

Adult male *X. variatus* of two tailspot types (i.e. wild-type and upper cut crescent) were collected with baited minnow traps from two populations in separate drainages: Atlapexco (146 m; 21.017,098.339; Water temperature: 25.2 °C) on the Rio Atlapexco in the Panuco drainage and Cacahuatengo (170 m; 20.781,098.032; 23.6 °C) on the Arroyo Grande in the Tuxpan drainage. In both populations, the two tailspot types were observed and collected within the same pools and in the same minnow traps. Fish were transported in thermally insulated coolers to a nearby field station where they were gradually acclimated to municipal tap water at 24 °C. Fish were maintained without food for 24 h to eliminate any bias in body mass as a result of a difference in the amount of bait consumed during collection and were then digitally photographed. Wet mass was measured to the nearest 0.001 g using a digital scale. Individuals of each tailspot pattern were randomly assigned to 40-L aquaria representing the control (wild-type,  $N = 18$ ; upper cut crescent,  $N = 18$ ) or treatment (wild-type,  $N = 16$ ; upper cut crescent = 18) in groups of eight to ten. Males were housed within populations to limit potential aggression as a result of unfamiliarity.

Submersible aquarium heaters were used to maintain control tanks at 24 °C and to raise treatment tanks to 32 °C over 18 h. Water temperature in natural populations at low elevations can fluctuate at least as much as 6.5 °C in a 24-h period (Z. W. Culumber, pers. observ.). A change of 8 °C is therefore likely ecologically-relevant in terms of climate change. As in a previous study examining thermal tolerance of tailspot types Culumber *et al.*, 2014), individuals in the thermal tolerance assays in the present study began to show signs of physiological distress, including increased operculum beat rate, hyperactivity and attempts to jump out of the assay chamber, at 32 °C, suggesting that the temperature

used for our treatment tanks is likely a stressful environment for experimental fish.

Once treatment tanks reached 32 °C, the experiment began. Feeding rate was held constant among all tanks in the control and treatment groups. We modified the methods of Fisher & Rosenthal (2006) to feed a lower rate of 0.01 g of tetramin flake food per fish, four times per day at 09.00, 11.00, 13.00, and 15.00 h. On days 2–5, we reduced the rate to 0.005 g fish<sup>-1</sup> four times per day in all tanks to prevent observed accumulation of food. Twenty-five percent water changes were conducted on days 1 and 3 on all tanks. Clean water was preheated to the proper temperatures to ensure that water changes did not alter water temperature in the tanks.

### ACCLIMATION CAPACITY OF THERMAL TOLERANCE

At the end of the experiment, following a final measure of body mass, we tested the  $CT_{max}$  of wild-type fish to assess acclimation capacity in thermal tolerance after acute heat stress. Thermal tolerance assays were conducted in accordance with previously published methods (Culumber *et al.*, 2014). Because populations can vary in thermal tolerance because of their environmental background (Culumber *et al.*, 2012), we tested for evidence of acclimation in thermal tolerance for the population with the largest sample size of wild-type males, Cacahuatengo (Control,  $N = 12$ ; Treatment,  $N = 11$ ). Because patterned fish have a naturally greater ability to withstand temperature increases (i.e. higher  $CT_{max}$ ; Culumber *et al.*, 2014), we only tested the acclimation capacity of wild-type fish.

### THERMAL STRESS AND BODY CONDITION

At the end of the 5-day experiment, the mass of each male was measured a final time. Morphology in male *X. variatus* is highly variable, making it easy to distinguish among males. Each fish was compared to digital photographs taken before the start of the experiment to confirm identity and re-weighed to calculate the percentage change in body mass during the course of the experiment. Rather than calculating relative condition factor (Borowsky, 1984), which is a measure of mass controlling for body size, we used change in body mass because *Xiphophorus* males do not grow after reaching maturity. Therefore, change in body mass is a direct metric of change in body condition within a group of mature males. In the present study, we used only males because female body condition cannot be directly compared with male condition as a result of variation in reproductive state of females. For the same reason, measures of body mass are not necessarily comparable within females.

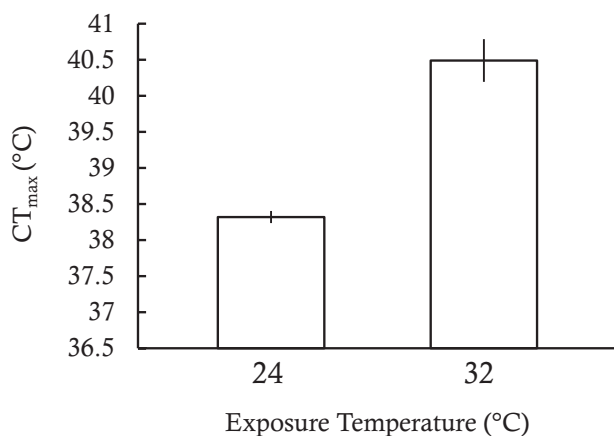
## STATISTICAL ANALYSIS

We first tested for a significant change in body mass over the course of the experiment using a paired Student's *t*-test. Preliminary Student's *t*-tests were conducted to test for differences between tanks within treatments and between the two populations. We assume that the tailspot types should have similar physiologies and physiological responses to stressors, regardless of population. Finding that there was no difference in change in body mass between tanks within a treatment, nor between populations, the data were combined. There was no difference between the two populations in initial mass, final mass or change in mass. We then tested for effects of temperature, tailspot genotype, and the interaction term genotype  $\times$  temperature on change in body mass using analysis of variance (ANOVA). A Shapiro–Wilk test was used to confirm that the data was normally distributed prior to running the ANOVA. A final Student's *t*-test was used to assess differences in  $CT_{max}$  of wild-type males between the control and treatment. All tests were two-tailed.

## RESULTS

## ACCLIMATION CAPACITY OF THERMAL TOLERANCE

There was a significant acclimation response of heat-sensitive wild-type fish exposed to short-term thermal stress ( $t = 6.68$ , d.f. = 22,  $P < 0.0001$ ) (Fig. 1). Wild-type fish from the 32 °C treatment had an upward shift in  $CT_{max}$ , which was 2.17 °C higher than fish that had been maintained at 24 °C (Fig. 1).



**Figure 1.** Heat-sensitive (wild-type) fish exhibited a significant acclimation capacity with a 2.17 °C increase in critical thermal maximum ( $CT_{max}$ ) after a 5-day exposure to 32 °C compared to males of the same tailspot type that were maintained at 24 °C ( $P < 0.0001$ ).

## THERMAL STRESS AND BODY CONDITION

Mortality occurred only in the 32 °C treatment tanks ( $N = 4$  individuals). Mortality was greatest in patterned fish ( $N = 3$ ) compared to wild-types ( $N = 1$ ). The remainder of fish remained healthy, exhibiting behaviour typical of *Xiphophorus* in aquaria (free swimming, eating, aggressive displays) and showing no symptoms of illness. Across all fish and treatments, there was a significant increase in body mass ( $t = 11.439$ , d.f. = 66,  $P < 0.0001$ ).

However, in contrast to expectations, there was no effect of temperature ( $F_{1,62} = 0.749$ ,  $P = 0.390$ ) (Figure 2A), nor tailspot genotype ( $F_{1,62} = 0.302$ ,  $P = 0.584$ ) on change in body mass. Consequently, there was also no effect of the interaction term tailspot  $\times$  temperature on body mass ( $F_{1,62} = 0.020$ ,  $P = 0.882$ ) (Figure 2B,C), indicating that neither of the temperatures affected the condition of the two pattern types.

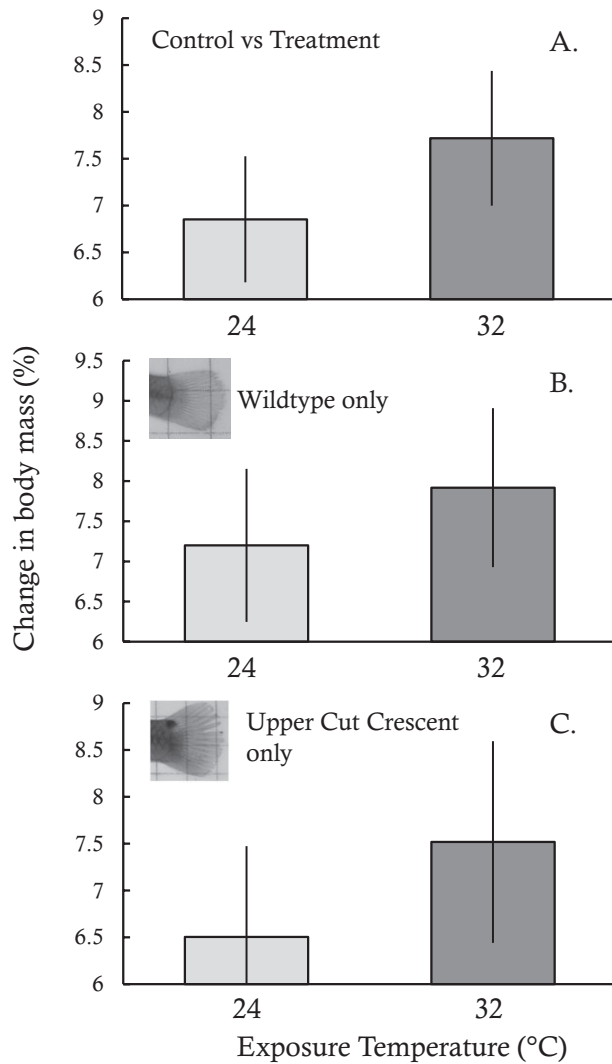
## DISCUSSION

Considerable uncertainty remains regarding the ability of species to respond to global climate change and the acclimation capacity of heat-sensitive species. This ability to buffer against and acclimate to thermal stress may be particularly important for tropical species that typically live near upper thermal limits (Deutsch *et al.*, 2008). Because short-term survivability and performance are critical to fitness and the long-term evolutionary potential of populations and species, it is important to know whether species are resilient to extreme temperature events. We observed considerable acclimation capacity to ecologically-relevant thermal stress in a heat-sensitive variant of a polymorphic fish. Wild-type fish exhibited almost no mortality and a significant acclimation capacity in upper thermal limit in response to a simulated extreme temperature event. By contrast to predictions based on heat tolerance and correlations between temperature and wild-type male body condition, wild-type fish exhibited no effects of thermal stress on body mass, nor did wild-type and patterned fish differ from one another in change in body mass. These findings suggest that the influence of temperature on male fitness and associated maintenance of the tailspot polymorphism may be more complex than a simple correlation between temperature.

EXTREME TEMPERATURE EVENTS AND ACCLIMATION CAPACITY OF  $CT_{max}$ 

The physiological response to stress associated with extreme temperatures is likely to be at least as important as longer-term adaptation to gradually increasing temperatures projected under global climate





**Figure 2.** Overall, fish gained body mass during the experimental period, although tailspot types did not differ. A, there was no overall effect of temperature on mean  $\pm$  SE percentage change in body mass across all fish, regardless of tailspot pattern. B, there was no difference in the change in body mass of wild-type males exposed to different temperatures. C, there was also no difference in change in body mass of upper cut crescent males between temperatures.

change. Outlier temperature events have been shown to impose greater selection than gradual changes in mean temperatures with respect to determining fitness and establishing geographical distributions in species closely related to *X. variatus* (Culumber *et al.*, 2012). Estimating thermal acclimation capacity to extreme events is therefore an equally important aspect for predicting the consequences of climate change. Species or individuals with relatively lower  $CT_{max}$  will be especially vulnerable to sudden tem-

perature increases, if they lack acclimation capacity to cope with acute events. We therefore tested the acclimation capacity of the comparatively heat-sensitive wild-type variant and found significant acclimation capacity. Wild-types exposed to acute temperature change followed by short-term thermal stress exhibited an almost 6% increase in  $CT_{max}$ , which is greater than the magnitude of change reported in other aquatic species (Calosi *et al.*, 2008). Heat-sensitive fish exhibited no difference in change in body condition compared to heat-tolerant fish and actually increased their body mass, indicating that there is potentially no or little cost to acclimation capacity in terms of energy reserves (see below).

#### THERMAL STRESS AND BODY CONDITION

Considerable study is still needed to understand the mechanistic basis of the reduced fitness expected to accompany climate change (Somero, 2010; Portner & Farrell, 2008). In male *X. variatus*, effects of temperature on body condition could be one such mechanism, and could potentially be driven by temperature-dependent effects on metabolism. The metabolism of fishes is typically directly affected by temperature (Johnston & Dunn, 1987). For example, in *Limia melanonotata*, a species of livebearer related to *X. variatus*, heat stress leads to a direct increase in metabolic rate (Haney & Walsh, 2003). However, the findings of the present study do not find support a positive correlation between wild-type male body condition and temperature. Fish at the higher temperature gained mass equally as well as at the cooler temperature, and the change in body mass of wild-type fish did not differ from that of patterned fish. Because an increased temperature leads to higher metabolic rates, the effect of temperature on body condition may result from a mechanism other than metabolic rates; for example, through effects of thermal stress on behaviour (*sensu* Du Plessis *et al.*, 2012).

Alternatively, whether through effects on metabolism, behaviour or other mechanism, it is plausible that reduced activity in captivity and ample food supply could have outweighed effects from thermal stress. If this is true, it would suggest that the effects of thermal stress are more pronounced under certain environmental conditions (e.g. higher flow velocity and resource limitation). Such factors will have to be addressed in future studies. Additionally, the original correlation (Borowsky, 1984) could have been driven by a variable correlated with temperature rather than by temperature itself. For example, oxygen limitation (Pörtner, 2001) could exacerbate thermal stress in natural populations because the dissolved oxygen concentration is temperature-dependent. In

our experiment, filters provided aeration and would have alleviated any potential hypoxia-related stress. Ultimately, our findings suggest that the relationship between body condition and temperature may not be as straightforward as a simple correlation, and may rely on the influence of other factors.

Although physiological response to thermal stress is known to differ among allelic variants of the polymorphic tailspot locus, thermal stress was not reflected in the body condition of adult males in the present study. The functional consequences of thermal stress alone on adult fish may therefore not be sufficient to generate significant selection on adult males. Our findings suggest that heat-sensitive tropical species living near their upper thermal limits may have greater resilience to extreme temperature fluctuations than might be expected. This capacity to acclimate to extreme temperature fluctuations represents an important mechanism for alleviating the short-term effects of climate change, and for maintaining the long-term evolutionary potential required for the response to sustained, gradual warming.

#### ACKNOWLEDGEMENTS

We would like to thank the Mexican Government for collecting permits and C. E. Bautista-Hernández for help with fish collection. This work was supported by a Consejo Nacional de Ciencia y Tecnología (CONACyT) grant (Clave 0127310) in basic science to SM, which supported ZWC as a postdoctoral researcher.

#### REFERENCES

- Ackerly DD. 2003.** Community assembly, niche conservatism, and adaptive evolution in changing environments. *International Journal of Plant Sciences* **164**: S165–S184.
- Addo-Bediako A, Chown SL, Gaston KJ. 2000.** Thermal tolerance, climatic variability and latitude. *Proceedings of the Royal Society of London Series B, Biological Sciences* **267**: 739–745.
- Allen JL, Clusella-Trullas S, Chown SL. 2012.** The effects of acclimation and rates of temperature change on critical thermal limits in *Tenebrio molitor* (Tenebrionidae) and *Cyrtobagous salviniae* (Curculionidae). *Journal of Insect Physiology* **58**: 669–678.
- Borowsky R. 1981.** Tailspots of *Xiphophorus* and the evolution of conspicuous polymorphism. *Evolution* **35**: 345–358.
- Borowsky R. 1984.** The evolutionary genetics of *Xiphophorus*. In: Turner BJ, ed. *Evolutionary genetics of fishes*. New York & London: Springer, 235–310.
- Burgi LP, Mills NJ. 2012.** Ecologically relevant measures of the physiological tolerance of light brown apple moth, *Epiphyas postvittana*, to high temperature extremes. *Journal of Insect Physiology* **58**: 1184–1191.
- Calosi P, Bilton DT, Spicer JI. 2008.** Thermal tolerance, acclimatory capacity and vulnerability to global climate change. *Biology Letters* **4**: 99–102.
- Culumber ZW, Bautista-Hernández CE, Monks S. 2014.** Physiological stress and the maintenance of adaptive genetic variation in a livebearing fish. *Evolutionary Ecology* **28**: 117–129.
- Culumber ZW, Shepard DB, Coleman SW, Rosenthal GG, Tobler M. 2012.** Physiological adaptation along environmental gradients and replicated hybrid zone structure in swordtails (Teleostei: *Xiphophorus*). *Journal of Evolutionary Biology* **25**: 1800–1814.
- Deutsch CA, Tewksbury JJ, Huey RB, Sheldon KS, Ghalambor CK, Haak DC, Martin PR. 2008.** Impacts of climate warming on terrestrial ectotherms across latitude. *Proceedings of the National Academy of Sciences of the United States of America* **105**: 6668–6672.
- Donner SD, Knutson TR, Oppenheimer M. 2007.** Model-based assessment of the role of human-induced climate change in the 2005 Caribbean coral bleaching event. *Proceedings of the National Academy of Sciences of the United States of America* **104**: 5483–5488.
- Du Plessis KL, Martin RO, Hockey PAR, Cunningham SJ, Ridley AR. 2012.** The costs of keeping cool in a warming world: implications of high temperatures for foraging, thermoregulation and body condition of an arid-zone bird. *Global Change Biology* **18**: 3063–3070.
- Fisher HS, Rosenthal GG. 2006.** Female swordtail fish use chemical cues to select well-fed mates. *Animal Behaviour* **72**: 721–725.
- Hampe A, Petit RJ. 2005.** Conserving biodiversity under climate change: the rear edge matters. *Ecology Letters* **8**: 461–467.
- Haney DC, Walsh SJ. 2003.** Influence of salinity and temperature on the physiology of *Limia melanonotata* (Cyprinodontiforme: Poeciliidae): a search for abiotic factors limiting insular distribution in Hispaniola. *Caribbean Journal of Science* **39**: 327–337.
- Hansen J, Sato M, Ruedy R. 2012.** Perception of climate change. *Proceedings of the National Academy of Sciences of the United States of America* **109**: E2415–E2423.
- Harley CDG. 2008.** Tidal dynamics, topographic orientation, and temperature-mediated mass mortalities on rocky shores. *Marine Ecology Progress Series* **371**: 37–46.
- Heino J, Virkkala R, Toivonen H. 2009.** Climate change and freshwater biodiversity: detected patterns, future trends and adaptations in northern regions. *Biological Reviews* **84**: 39–54.
- Helmuth BST, Hofmann GE. 2001.** Microhabitats, thermal heterogeneity, and patterns of physiological stress in the rocky intertidal zone. *Biological Bulletin* **201**: 374–384.
- Hoffmann AA. 2010.** Physiological climatic limits in *Drosophila*: patterns and implications. *Journal of Experimental Biology* **213**: 870–880.
- Hofmann GE, Todgham AE. 2010.** Living in the now: physiological mechanisms to tolerate a rapidly changing environment. *Annual Review of Physiology* **72**: 127–145.

- Johnston IA, Dunn J. 1987.** Temperature acclimation and metabolism in ectotherms with particular reference to teleost fish. *Symposia of the Society for Experimental Biology* **41**: 67–93.
- Jumbam KR, Jackson S, Terblanche JS, McGeoch MA, Chown SL. 2008.** Acclimation effects on critical and lethal thermal limits of workers of the Argentine ant, *Linepithema humile*. *Journal of Insect Physiology* **54**: 1008–1014.
- Jump AS, Penuelas J. 2005.** Running to stand still: adaptation and the response of plants to rapid climate change. *Ecology Letters* **8**: 1010–1020.
- Kellermann V, Overgaard J, Hoffmann AA, Fløjgaard C, Svenning J-C, Loeschke V. 2012.** Upper thermal limits of *Drosophila* are linked to species distributions and strongly constrained phylogenetically. *Proceedings of the National Academy of Sciences of the United States of America* **109**: 16228–16233.
- Kindsvater HK, Simpson SE, Rosenthal GG, Alonzo SH. 2013.** Male diet, female experience, and female size influence maternal investment in swordtails. *Behavioral Ecology* **24**: 691–697.
- Krebs RA, Feder ME. 1997.** Natural variation in the expression of the heat-shock protein Hsp70 in a population of *Drosophila melanogaster* and its correlation with tolerance of ecologically relevant thermal stress. *Evolution* **51**: 173–179.
- Lexer C, Welch ME, Durphy JL, Rieseberg LH. 2003.** Natural selection for salt tolerance quantitative trait loci (QTLs) in wild sunflower hybrids: implications for the origin of *Helianthus paradoxus*, a diploid hybrid species. *Molecular Ecology* **12**: 1225–1235.
- Ma G, Ma C-S. 2012.** Effect of acclimation on heat-escape temperatures of two aphid species: implications for estimating behavioral response of insects to climate warming. *Journal of Insect Physiology* **58**: 303–309.
- McKechnie AE, Wolf BO. 2010.** Climate change increases the likelihood of catastrophic avian mortality events during extreme heat waves. *Biology Letters* **6**: 253–256.
- Meehl GA, Stocker TF, Collins WD, Friedlingstein P, Gaye AT, Gregory JM, Kitoh A, Knutti R, Murphy JM, Noda A. 2007.** Global climate projections. *Climate Change* **3495**: 747–845.
- Meehl GA, Tebaldi C. 2004.** More intense, more frequent, and longer lasting heat waves in the 21st century. *Science* **305**: 994–997.
- Mitchell KA, Hoffmann AA. 2010.** Thermal ramping rate influences evolutionary potential and species differences for upper thermal limits in *Drosophila*. *Functional Ecology* **24**: 694–700.
- Mitchell KA, Sgro CM, Hoffmann AA. 2011.** Phenotypic plasticity in upper thermal limits is weakly related to *Drosophila* species distributions. *Functional Ecology* **25**: 661–670.
- Parmesan C. 2006.** Ecological and evolutionary responses to recent climate change. *Annual Review of Ecology, Evolution, and Systematics* **37**: 637–669.
- Perry AL, Low PJ, Ellis JR, Reynolds JD. 2005.** Climate change and distribution shifts in marine fishes. *Science* **308**: 1912–1915.
- Peterson AT, Ortega-Huerta MA, Bartley J, Sanchez-Cordero V, Soberon J, Buddemeier RH, Stockwell DRB. 2002.** Future projections for Mexican faunas under global climate change scenarios. *Nature* **416**: 626–629.
- Petes LE, Menge BA, Murphy GD. 2007.** Environmental stress decreases survival, growth, and reproduction in New Zealand mussels. *Journal of Experimental Marine Biology and Ecology* **351**: 83–91.
- Plath M, Heubel KU, de León FJG, Schlupp I. 2005.** Cave molly females (*Poecilia mexicana*, Poeciliidae, Teleostei) like well-fed males. *Behavioral Ecology and Sociobiology* **58**: 144–151.
- Portner HO, Farrell AP. 2008.** Physiology and climate change. *Science* **322**: 690–692.
- Pörtner H. 2001.** Climate change and temperature-dependent biogeography: oxygen limitation of thermal tolerance in animals. *Die Naturwissenschaften* **88**: 137–146.
- Santos M, Castaneda LE, Rezende EL. 2012.** Keeping pace with climate change: what is wrong with the evolutionary potential of upper thermal limits. *Ecology and Evolution* **2**: 2866–2880.
- Sexton JP, McIntyre PJ, Angert AL, Rice KJ. 2009.** Evolution and ecology of species range limits. *Annual Review of Ecology Evolution and Systematics* **40**: 415–436.
- Smith JB, Schneider SH, Oppenheimer M, Yohe GW, Hare W, Mastrandrea MD, Patwardhan A, Burton I, Corfee-Morlot J, Magadza CHD. 2009.** Assessing dangerous climate change through an update of the Intergovernmental Panel on Climate Change (IPCC): reasons for concern. *Proceedings of the National Academy of Sciences of the United States of America* **106**: 4133–4137.
- Somero GN. 2005.** Linking biogeography to physiology: evolutionary and acclimatory adjustments of thermal limits. *Frontiers in Zoology* **2**: 1–9.
- Somero GN. 2010.** The physiology of climate change: how potentials for acclimatization and genetic adaptation will determine ‘winners’ and ‘losers’. *Journal of Experimental Biology* **213**: 912–920.
- Somero GN. 2012.** The physiology of global change: linking patterns to mechanisms. *Annual Review of Marine Science* **4**: 39–61.
- Stillman JH. 2003.** Acclimation capacity underlies susceptibility to climate change. *Science* **301**: 65–65.
- Thomas CD, Cameron A, Green RE, Bakkenes M, Beaumont LJ, Collingham YC, Erasmus BFN, De Siqueira MF, Grainger A, Hannah L. 2004.** Extinction risk from climate change. *Nature* **427**: 145–148.
- Van Dooremalen C, Berg MP, Ellers J. 2013.** Acclimation responses to temperature vary with vertical stratification: implications for vulnerability of soil-dwelling species to extreme temperature events. *Global Change Biology* **19**: 975–984.