

Thermal specialization across large geographical scales predicts the resilience of mangrove crab populations to global warming

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| Abstract: | The broad prediction that ectotherms will be more vulnerable to climate change in the tropics than in temperate regions includes assumptions about centre/edge population effects that can only be tested by within-species comparisons across wide latitudinal gradients. Here, we investigated the thermal vulnerability of two mangrove crab species, comparing populations at the centre (Kenya) and edge (South Africa) of their distributions. At the same time, we investigated the role of respiratory mode (water- vs air-breathing) in determining the thermal tolerance in amphibious organisms. To do this, we compared the vulnerability to acute temperature fluctuations of two sympatric species with two different lifestyle adaptations: the free living <i>Perisesarma guttatum</i> and the burrowing <i>Uca urvillei</i> , both pivotal to the ecosystem functioning of mangroves. The results revealed the air-breathing <i>U. urvillei</i> to be a thermal generalist with much higher thermal tolerances than <i>P. guttatum</i> . Importantly, however, we found that, while <i>U. urvillei</i> showed little difference between edge and centre populations, <i>P. guttatum</i> showed adaptation to local conditions. Equatorial populations had elevated tolerances to acute heat stress and mechanisms of partial thermoregulation, which make them less vulnerable to global warming than temperate co-specifics. The results reveal both the importance of respiratory mode on thermal tolerance and the unexpected potential for low latitude populations/species to endure a warming climate and furthermore contribute to build a conceptual model on the latitudinal |

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1 **Title Page**

2 **Title:** Thermal specialization across large geographical scales predicts the resilience of
3 mangrove crab populations to global warming.

4 **Running head:** Latitudinal mangrove crab thermal specialization

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23

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1 Abstract

2 The broad prediction that ectotherms will be more vulnerable to climate change in the tropics
3 than in temperate regions includes assumptions about centre/edge population effects that can
4 only be tested by within-species comparisons across wide latitudinal gradients. Here, we
5 investigated the thermal vulnerability of two mangrove crab species, comparing populations at
6 the centre (Kenya) and edge (South Africa) of their distributions. At the same time, we
7 investigated the role of respiratory mode (water- vs air-breathing) in determining the thermal
8 tolerance in amphibious organisms. To do this, we compared the vulnerability to acute
9 temperature fluctuations of two sympatric species with two different lifestyle adaptations: the
10 free living *Perisesarma guttatum* and the burrowing *Uca urvillei*, both pivotal to the
11 ecosystem functioning of mangroves. The results revealed the air-breathing *U. urvillei* to be a
12 thermal generalist with much higher thermal tolerances than *P. guttatum*. Importantly,
13 however, we found that, while *U. urvillei* showed little difference between edge and centre
14 populations, *P. guttatum* showed adaptation to local conditions. Equatorial populations had
15 elevated tolerances to acute heat stress and mechanisms of partial thermoregulation, which
16 make them less vulnerable to global warming than temperate co-specifics. The results reveal
17 both the importance of respiratory mode on thermal tolerance and the unexpected potential for
18 low latitude populations/species to endure a warming climate and furthermore contribute to
19 build a conceptual model on the latitudinal thermal tolerance of these key species. This
20 highlights the need for an integrated population-level approach to predict the consequences of
21 climate change.

22

23

- 1 **Keywords:** East Africa; Global warming; Latitudinal effect; Mangrove macrobenthos,
- 2 Respiratory mode; Thermal specialization; Thermoregulation.
- 3

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1 **Introduction**

2 The current trend of global warming has severe implications for the structure and function of
3 ecosystems (Walther et al. 2002; Rockström et al. 2009; Byrne 2011, Borja, 2014). At large
4 geographical scales, increasing temperatures cause shifts in species distributions and
5 community composition (Sagarin et al. 1999; Deutsch et al. 2008; Beaugrand 2009;
6 Bozinovic, Calosi & Spicer 2011; Sunday, Bates & Dulvy 2011, 2012; Rius et al., 2014),
7 while at smaller scales temperature anomalies may cause local extinction of vulnerable
8 species often resulting in the contraction of a species range (Helmuth et al. 2002, 2006;
9 Pörtner & Farrell 2008). Global and local warming have direct effects on the functions of
10 organisms, population size and growth rate and consequently drive shifts in distribution
11 ranges which can destabilise community structure and dynamics (Helmuth et al. 2002;
12 Somero 2012; Sunday, Bates & Dulvy 2012). Ecologically, such shifts mediate and most
13 likely alter intra- and inter-specific dynamics such as competition for resources and prey-
14 predator interactions, often leading to the disruption of ecosystem functioning (Helmuth et al.
15 2006; Hoegh-Guldberg & Bruno 2010; Doney et al. 2012). However, the need to produce
16 reliable predictions of species response to global warming calls for accurate mechanistic
17 analyses able to integrate changes in several environmental variables and overcome the
18 intrinsic limitation of correlative approaches (Kearney et al. 2009). In this view, the
19 temperature-dependence of biological performance (e.g. fecundity, growth, foraging and
20 immune responses) in ectotherms achieved great relevance as a physiological cornerstone to
21 interpret vulnerabilities and resiliencies to the changing climate.

22 In particular, the capacity to sustain an efficient aerobic metabolism during acute
23 environmental temperature fluctuation has been proposed as a unifying functional principle to
24 understand species sensitivities to environmental change (Pörtner & Knust 2007; Pörtner &
25 Farrell, 2008; Pörtner & Giomi, 2013). Indeed, acute or episodic rises of temperature beyond

1 the thermal tolerance (defined as the upper and lower critical temperature) range of
2 individuals or species may determine the impairment of organismal energy status with a
3 consequent rebound on individual fitness, on the size and structure of populations and on the
4 overall community structure (Pörtner & Knust 2007; Farrell 2009; Bartolini et al. 2013). The
5 study of organisms' thermal tolerance at a large geographical scale is scarce and generally the
6 evaluation of animal resilience to global warming and the assessment of species vulnerability
7 have focused on temperate latitudes, while warm adapted stenotherms within tropical latitudes
8 have received only marginal interest (for a review see Parmesan 2006). This information gap
9 is partially addressed by meta-analyses and global observations that have predicted that the
10 major impact of climate change on biodiversity and animal redistribution is likely to occur at
11 low latitudes (Deutsch et al. 2008; Dillon, Wang & Huey 2010; Sunday, Bates & Dulvy 2011,
12 2012; Huey et al. 2012; Sunday et al., 2014).

13 By definition, however, global analyses merge single cases to project general rules and risk
14 losing informative elements at the organismal level (Williams et al. 2008; Marshall, McQuaid
15 & Williams 2010; Huey et al. 2012). In particular, to date, projections from meta-analytic
16 elaborations have not focused on possible selective differences between eurytherm and
17 stenotherm species and on their specific potential to cope with anomalous climatic events.
18 With these premises, the present work aims to address the current projection of a greater
19 vulnerability to warming of low latitude populations, incorporating an accurate comparison
20 between stenotherm and eurytherm model species with special reference to their respiratory
21 strategies. Intertidal organisms are ideal candidates for studies of thermal adaptation as they
22 live at the interface of marine and terrestrial systems, may rely on different sources of oxygen
23 (i.e. aerial or dissolved) and are susceptible to larger climate anomalies, since they have to
24 cope daily with abrupt and extreme heat stress (Helmuth et al. 2002, 2006; Kelly et al. 2012).
25 Intertidal organisms often live close to the upper limits of their thermal niches, i.e. the range

1 of operative temperatures that allows species survival and growth (Somero 2002; Helmuth et
2 al. 2010). We therefore examined the thermal vulnerability of two intertidal bimodal breathing
3 mangrove species, both pivotal for mangrove ecosystem functioning (Kristensen 2008),
4 *Perisesarma guttatum* (A. Milne Edwards, 1869) (Sesarmidae) and *Uca urvillei* (H. Milne-
5 Edwards, 1852) (Ocypodidae) at the centre of their distributions (Kenya) as well as their sub-
6 tropical southern limits (South Africa). Simultaneously, we tested the role of respiratory
7 physiology in buffering environmental stress by investigating the link between environmental
8 temperatures, respiratory mode and their overall thermal tolerance (Pörtner 2010; Verberk &
9 Bilton, 2013; Giomi et al. 2014).

1 **Materials and methods**

2 *Model species and study area*

3 *Perisesarma guttatum* and *Uca urvillei* are sympatric key-stone species that occupy the
4 seaward fringe of mangroves where they experience inundation twice daily (Macnae 1968;
5 Tomlinson 1995). *P. guttatum* lives among *Rhizophora mucronata* trees that provide shelter
6 from predators and resting spots during high tide (Slim et al. 1997; Flores, Saraiva & Paula
7 2002). *U. urvillei* prefers microhabitats provided by *Avicennia marina* trees, where there are
8 muddy open spaces to graze, filter sediment and build burrows (Hartnoll 1975; Litulo 2005).
9 Both species are important ecosystem engineers that contribute to ecosystem functioning
10 through the regulation of carbon and nutrient fluxes in the mangrove, changing the physico-
11 chemical properties of the litter, sediment and rhizosphere (Holguin, Vazquez & Bashan
12 2001; Kristensen & Alongi 2006; Cannicci et al. 2008; Lee 2008; Sousa and Dagremond
13 2011).

14 Gazi Bay, in Kenya (4° 22' S, 39° 30' E) is a semi-enclosed, shallow bay, 40 km south of
15 Mombasa (Fig.1). The climate there is influenced by the moist southeast monsoon from
16 March to September and the dry northeast monsoon from October to March (Kitheka 1996).

17 The mangrove forest in the Mngazana Estuary (31° 42' S, 29° 25' E) on the southeast coast of
18 South Africa (Fig.1) is one of the southernmost in the world and extends to the level of the
19 average high neap tide (Macnae 1963).

20

21 *Tidal regimes and climatic time series*

22 Tidal time series data were retrieved using Wtide software version 3.1.7 (www.wtide.com), in
23 order to link the tidal level in each site with the time-temperature data described below.

24 Time-series of temperature data were recorded in the field during October - November 2011:
25 49 days in Kenya and 43 days in South Africa. At each site, 20 temperature i-button loggers

1 (Maxim integrated product, ColdChain Thermodynamics) embedded in sealed silicon covers
2 that do not affect measurement quality (Roznik & Alford 2012) were placed about 3 cm above
3 the sediment surface and an additional 20, 20 cm below ground in areas inhabited by the focal
4 crabs. At each site, four additional i-button loggers were used to record air temperature and
5 humidity. These were attached to branches under the canopy about 4 m above ground to avoid
6 submersion and protected by an umbrella-like transparent plastic cover. All loggers were set
7 to record at 5 min intervals for 15 days, after which data were retrieved and the i-buttons re-
8 deployed. Data were downloaded with Cold Chain ThermoDymanimcs software (version 4.9 -
9 Fairbridge Technologies) and temperature and humidity were averaged at an hourly scale.

10

11 *Crab sampling for laboratory experiments*

12 Adult males of each species were collected at each site during the warmer season 2011
13 (average air temperature 27°C at both sites). Animals were maintained in tanks for two days
14 with fresh mud and aerated seawater at $27 \pm 0.5^\circ\text{C}$, 35‰ salinity before experimentation.
15 Crabs were fed with commercial cat food pellets and exposed to a natural 12 h light cycle.

16

17 *Oxygen consumption*

18 Routine rates of oxygen consumption (MO_2 hereafter) were measured separately in air and in
19 water for both species across a temperature ramp 17 - 37°C increasing at the rate of $1^\circ\text{C}\cdot\text{h}^{-1}$.
20 MO_2 was measured in an intermittent flow respirometer equipped with eight parallel darkened
21 Perspex chambers placed in a temperature-controlled water bath. An oxygen sensor (Sensor
22 Type PSt3 PreSens, Regensburg, Germany), glued to the inside wall of the chamber and
23 connected to a single channel oxygen transmitter Fibox 3 (PreSens, Regensburg, Germany)
24 through an optical SMA fiber, was used to measure the partial pressure of oxygen in air and
25 water. Data were recorded using FibSoft v.1.0 software (Loligo Systems ApS). Prior to

1 measurements, sensors were calibrated in air-equilibrated seawater (100% saturation) and in
2 sodium dithionite saturated solution in seawater (0%). During trials, oxygen concentration
3 was not allowed to fall below 60% in order to avoid exposing the animals to severe hypoxic
4 conditions (Schurmann & Steffensen 1992). The limited movement of individuals was
5 adequate to ensure mixing of the water and MO_2 was determined by measuring the linear
6 decline in oxygen saturation. An empty chamber was run as a control with each trial to
7 account for background oxygen depletion, which was less than 2% of crab consumption in
8 water and negligible in air. Before applying the temperature ramp, individuals were placed in
9 the chambers and allowed to recover from handling stress 8 hours at $27 \pm 0.5^\circ C$. MO_2 was
10 determined every two degrees of temperature. Following each experiment, animals were
11 weighed and their volume calculated by water displacement in a graduated cylinder. All
12 experiments lasted less than 24 h to avoid interference of metabolic rate by other factors such
13 as starvation (Terblanche et al. 2011).

14

15 *Aerobic performance*

16 Oxygen partial pressure in arterial and venous haemolymph (PO_2) was measured in two
17 separately experimental set, one during water respiration and the other one during the air
18 respiration. PO_2 was measured with fiber-optic oxygen microsensors (PreSens GmbH)
19 connected to an oxygen meter (Microx-TX, PreSens GmbH) with integrated signal processing
20 software. Sensors were calibrated before each experiment using a two-point calibration in
21 oxygen-free (addition of sodium dithionite) and air-saturated seawater. Animals were
22 acclimated overnight at $27^\circ C$ and PO_2 was estimated between 27 and $38^\circ C$ (accuracy $\pm 1^\circ C$),
23 using the protocol described for the MO_2 experiment.

24 To measure arterial PO_2 , haemolymph small haemolymph samples (less than 20 μl) were
25 collected through capillary action, using a manually sharpened Pasteur pipette in which the

1 oxygen sensor was previously inserted and positioned close to the tip. Arterial haemolymph
2 was withdrawn with such pipette from a hole (0.2 mm width) previously drilled through the
3 carapace, over the pericardial sinus following Frederich and Pörtner (2000). Similarly venous
4 blood was withdrawn from the sinus below the arthroal membrane, at the base of the fourth
5 or fifth pereopod (Giomi & Pörtner 2013). At each measurement the pipette was changed and
6 the sensor properly washed with sterile water. PO₂ data are recorded for only for Kenyan
7 population.

8

9 *Body temperature*

10 To measure the body temperature of animals, ten similar-sized adult male *U. urvillei* and *P.*
11 *guttatum* were selected (carapace width: 20 mm and 15 mm respectively). We used a two-
12 input digital thermometer (Omega©, HH504) with two micro- K-type thermocouples
13 (Omega©) to record body and environmental temperature simultaneously. The day before
14 experimentation, a hole was drilled through the carapace to the gill chamber. Crabs were then
15 kept in aerated sea water to recover overnight at 27°C (Frederich & Pörtner 2000). The
16 following day, the micro-thermocouple registering the body temperature was carefully
17 introduced into the gill chamber and fixed to the carapace using cyanoacrylate glue, while the
18 other one was fixed to the wire of the other sensor close by the crab to detect the exact
19 external temperature experienced by the animal. Specimens were released in the field during
20 low tide, allowed 30 min of field/handling acclimation and then followed for one hour, during
21 which body and environmental temperatures were recorded at 5 min intervals. Since the wire
22 of the thermocouple was really thin and light the animal behaved normally, not disturbed or
23 was not impeded. The position of the animal (e.g. in a hole or a crevice, in sun or shade, in
24 water) was also recorded.

25

1 *Statistical analysis*

2 To test the statistical significance among MO_2 , an ANCOVA performed with generalized
3 linear model technique was used, considering MO_2 as the response variable, temperature as
4 continuous explanatory variable, while Species (*P. guttatum*, *U. urvillei*), Region (Kenya,
5 South Africa) and Medium (water, air) the categorical explanatory variables.

6 The same test was applied to detect statistical significance of PO_2 , considering it as the
7 response variable and the temperature as explanatory variable, while Species (*P. guttatum*, *U.*
8 *urvillei*), Medium (water, air) and Haemolymph (Arterial, Venous) the categorical explanatory
9 variables. Due to the nature of our response variable following Poisson distribution, a log link
10 function has been used to obtain the best fitted model for both MO_2 and PO_2 . These statistical
11 analyses were carried out in R (R Development Core Team, 2012).

12 Full factorial Permutational Analyses of Variance (PERMANOVA, Anderson 2001) were
13 used to test the null hypothesis of no differences in, environmental temperature and humidity,
14 and body temperature. Post-hoc pairwise tests (p-hpt) were performed to examine significant
15 differences relevant to our hypotheses.

16 For the environmental data, the null hypothesis of no differences for temperature and
17 humidity across Region (KY, ZA), Species (*P. guttatum*, *U. urvillei*), kind of temperature
18 (Only for temperatures - Above ground, Below ground and Air) defined as fixed and
19 orthogonal was tested. Following the same experimental design, a PERMIDSP has been
20 performed to test the variability among temperatures and humidity.

21 For the experiments on body temperature, the null hypothesis of no difference across Region
22 (KY, ZA), Position (Hole/Crevise, Sun, Shadow, Water) and Temperature Type (Body and
23 Environmental), defined as fixed and orthogonal was tested. To test for statistical differences
24 in Position between Species and Type of temperature, a paired-PERMANOVA design was

1 used, as the same specimens were used for each experiment, with Species and Type as factors
2 in the model.

3 All data in the figures are expressed as means \pm SE and the analyses were performed using the

4 PERMANOVA+ routines for PRIMER 6 (Anderson, Gorley & Clarke 2008).

5

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1 Results

2 *Tidal regimes and climatic time series*

3 In Kenya, the tidal range is 0 to 4 m and 1 to 2.5 m during spring and neap tides, respectively.

4 In South Africa, the tidal range is 0 and 2 m and 0.5 to 1.5 during spring and neap tides,
5 respectively. Tidal levels during the observation periods differed between the two regions
6 (Fig. 2).

7 Thermal regimes were also markedly different between regions (PERMANOVA, $F= 1346.8$;
8 $df= 1,550$; $p<0.001$), air temperature being on average higher and less variable in Kenya
9 (PERMDISP, $t=2,43$; $p<0.0001$). In Kenya, temperatures in the area occupied by *P. guttatum*
10 were 23-37°C above ground, 26-33°C below ground and 22-35°C in the air. Temperatures
11 experienced by *U. urvillei* were similar: 23-38°C above ground (PERMANOVA, $t=1,32$;
12 $p=0.11$, p-pht), 28-31°C below ground and 22-35°C (PERMANOVA, $t=0.49$; $p=0.64$, p-pht),
13 in air (PERMANOVA, $t=1,45$; $p=0.08$, p-pht). South African temperatures were more
14 variable than the Kenyan ones (PERMDISP, $t=2,83$; $p<0.0001$). For *P. guttatum* micro-
15 habitat, temperatures were 16-37°C above ground, 22-30°C below ground and 15.4-32°C in
16 the air, with similar values for the *U. urvillei* area: 20-36°C above ground (PERMANOVA,
17 $t=0.88$; $p=0.93$, p-pht), 19-33°C below ground (PERMANOVA, $t=0.37$; $p=0.73$, p-pht), and
18 15-32°C in air (PERMANOVA, $t=0.16$; $p=0.89$, p-pht).

19 Humidity measurements ranged between 60 and almost 100% without any significant
20 differences between regions (PERMANOVA, $F= 0.55$ $df= 1,152$; $p=0.79$) both sites
21 depending on the state of the tide, with values again being more variable in South Africa than
22 Kenya.

23

24 *Oxygen consumption (MO_2)*

1 Thermal responses in MO_2 were clearly different between the two species across two
2 geographic regions and medium (Fig. 3, Tab. S1). In both regions, *P. guttatum* showed a
3 pronounced increase in MO_2 during the thermal ramp that was steeper than for *U. urvillei*. The
4 Kenyan population of *P. guttatum* had a higher metabolic rate in air than in water (ANCOVA;
5 $F= 605.6$, $df=1,1162$; $p < 0.0001$), while in South Africa the opposite was true, with lower
6 MO_2 in air. Surprisingly, the South African population showed significant indications of a
7 critical thermal threshold above $33^\circ C$ in both air and water (Fig.3), when the temperature
8 induced rise in MO_2 levelled off in air and inverted in water. In contrast, the temperature
9 dependent increase of MO_2 in the Kenyan population was monotonic, with an increasing trend
10 even at $37^\circ C$. *U. urvillei* showed a similar trend in rates of MO_2 in water and in air in both
11 regions, with no significant differences between regions or medium (ANCOVA; $F= 0.5741$,
12 $df=1,1162$; $p>0.05$).

13

14 *Aerobic performance*

15 For both species, haemolymph PO_2 (Tab. S2) showed a significant interaction between
16 haemolymph (venous or arterial), temperature and medium (ANCOVA; $F= 4.17$, $df=1,268$;
17 $p<0.05$; Fig. 4 a-f). *P. guttatum* showed lower efficiency of oxygen uptake in air than in water
18 (Fig. 4 a, c and e). Increasing temperature reduced the difference in PO_2 between arterial and
19 venous haemolymph, until they became equal at $37^\circ C$ suggesting oxygen delivery to be
20 impaired or $33^\circ C$ in air.

21 For *U. urvillei*, haemolymph PO_2 was significantly different between air and water, with a
22 significant dependence on temperature and source in air (ANCOVA; $F= 29.30$; $df, 1,268$;
23 $p<0.0001$). In water, haemolymph PO_2 was constantly low with no statistical differences
24 between venous and arterial blood (Fig. 4 b, d and f).

25

1 *Body temperature*

2 Body temperatures, and the relationships between body and environmental temperatures,
3 differed between Kenyan and South Africa populations (PERMANOVA, Tables S3a, b; Fig.
4 5). The body temperature of *U. urvillei* was never lower than environmental temperature, with
5 warmer body temperatures recorded in the South African population. *P. guttatum* in Kenya
6 showed body temperatures as much as 2°C lower than environmental temperatures, especially
7 during sun exposure ($t = 4.7873$, $df = 56$, $p = 0.0001$); though no down-regulation of body
8 temperature was observed for the South African population. We also found greater variability
9 in the data from the South African population than those from the Kenyan population for both
10 species (Fig. 5).

11

12

1 Discussion

2 The temperature and humidity data revealed expected, but distinct, thermal regimes for Kenya
3 and South Africa, in agreement with previous surveys (Smit et al. 2013; McClanahan 2014;
4 Hoepffner et al. 2014). The equatorial mangrove site was characterised by limited thermal
5 variation, while the sub-tropical system showed a broader temperature range on both daily and
6 seasonal scales. Besides the obvious latitudinal influence, a further source of variation in the
7 thermal regime was provided by the tidal cycle and tidal range. In South Africa, during neap
8 tides, the sediment can remain uncovered by water for up to five consecutive days, while in
9 Kenya the sediment is always covered by water at high tide.

10 Overall, *Uca urvillei* consistently increased its oxygen consumption, suggesting that its
11 respiration was not impaired by heat during aerial respiration. In addition, this species,
12 displays a large difference between arterial and venous PO₂ when relies on aerial oxygen. This
13 demonstrates a highly efficient gas exchange in air thanks to vascularised tissue development
14 in the gill chamber acting as true lungs (Farrelly & Greenaway 1994; Morris 2002) which
15 reveal a successful terrestrial adaptation. During acute warming, air - breathing arthropods can
16 sustain the increased energy demands of activity by exploiting the elevated oxygen content
17 and diffusiveness in air that would be constraining in water (Pörtner 2001, 2010; Stevens et al.
18 2010; Verberk et al. 2011; Giomi et al. 2014), considering also its more elevated density and
19 viscosity that add further challenge in underwater respiration (Verberk & Atkinson, 2013).
20 Consequently, the fact that *U. urvillei* is essentially an air breather can explain its pronounced
21 general heat tolerance, lack of thermoregulation and ability to sustain activity at high
22 temperatures (Fusi, unpublished data). This interpretation is supported by the dramatic
23 hypoxemia that this species develops when submerged, which is a sign of extremely
24 inefficient oxygen extraction from water. These findings indicate that *U. urvillei* is essentially
25 an air-breathing species, poorly adapted to underwater respiration, even though it inhabits

1 environments that are totally flooded twice a day. The ability to sustain prolonged periods
2 underwater is accomplished by burrow plugging behaviour. *U. urvillei* spends a considerable
3 amount of time looking after its burrow in order to prepare the surface of the sediment for
4 plugging of the hole by carving a mud disc adjacent to the entrance of the burrow, as
5 described for other fiddler crab species (de la Iglesia, 1994). Once plugged, crabs carefully
6 seal the disc from inside the hole to avoid water leakage and to trap air inside the burrow.
7 Plugging behaviour is well described in the genus *Uca* (Hartnoll 1988, Lee 1998; Skov &
8 Hartnoll 2001; Yamaguchi 2001; Litulo 2004; Shih, Mok & Chang 2005), though little is
9 known about such behaviour in this species. Plugging, in fact, guarantees a reservoir of
10 oxygenated air throughout the high tide (Marco Fusi, unpublished data). Similar mechanisms
11 to endure high tide have been described for other animals living in mangroves, such as the
12 mudskippers *Periophthalmus sobrinus* (Stebbins & Kalk 1961) and *Scartelaos histophorus*
13 (Lee et al. 2005) and the Australian mud-dwelling mangrove ant *Polyrhachis sokolova*
14 (Nielsen, Christian & Birkmose 2003; Nielsen 2011).

15
16 *P. guttatum* revealed a different physiological adaptation. Analysis of haemolymph PO_2
17 indicates that this species is a bimodal breather, with limited efficiency of oxygen extraction
18 in air. *P. guttatum* is a non-burrowing sesarmid, living freely on the floor of the forest,
19 sheltering within crevices and holes around prop roots where they feed on leaves and other
20 organic material (Fratini et al. 2005; Silva, Mesquita & Paula 2010b). *P. guttatum* tended to
21 avoid long periods of exposure to higher temperatures (Fusi, unpublished data), which would
22 require excessive energy expenditure to sustain metabolic demands (Fig. 4).

23

24 The results of this study reveal unexpected climate induced response of tropical ectotherms to
25 intertidal environments, supporting the need to adopt a multilevel integrated approach when

1 evaluating latitudinal vulnerability and plasticity of species to climate change (Williams et al.
2 2008). In particular, the two studied species showed different mechanisms of adaptation to
3 temperature regimes and a diverse degree of resilience to regional warming. *U. urvillei*,
4 through a high temperature tolerance and a consistent metabolic response across a wide range
5 of temperatures, exhibits a single level of thermal response in different environments and can
6 be described as a thermal generalist, highly tolerant to a broad range of temperatures across its
7 entire geographical distribution. In contrast, the specialist, *P. guttatum*, showed distinct
8 tolerance ranges across the geographical gradient and appeared to be adapted to local climatic
9 regimes at different latitudes (*sensu* Angilletta 2009; Huey et al. 2012). *P. guttatum* in Kenya
10 is highly specialised to a narrow range of environmental temperatures and shows an ability to
11 endure anomalous heats. In contrast, the South African population of *P. guttatum* is evolved
12 to cope with a wider range of environmental temperatures that vary on both daily and seasonal
13 scales and display the physiological mechanisms to tolerate larger climate fluctuations that
14 occur in its natural environment. The same population, however, manifests a critical thermal
15 threshold at 33°C, above which the temperature-MO₂ relationship levels off in both air and
16 water. Indeed, the body temperatures of *U. urvillei* matched those of the environment and
17 were occasionally even higher, while Kenyan individuals of *P. guttatum* were able to lower
18 their body temperature by up to two degrees during air exposure, indicating that the low
19 latitude population is capable of thermoregulation.

20 These findings accord with those on the temperature dependence of metabolism. The species
21 specific differences in temperature dependency of metabolism are a key element in
22 understanding the vulnerability to climate change across a wide latitudinal range and
23 forecasting changes in its distribution. Both *U. urvillei* and the Kenyan population of *P.*
24 *guttatum* are able to endure acute heat events and can efficiently meet the temperature induced
25 increase of metabolic oxygen demand. For *P. guttatum*, however, this efficient response is

1 likely sustained at a high cost in terms of metabolic expenditure, as suggested by the generally
2 higher MO_2 and the body temperature down-regulation in the Kenyan population. The tropical
3 mangrove ecosystem is probably an elective habitat for such metabolic adaptation, most likely
4 facilitated by relevant bio-energetic and environmental features such as elevated primary
5 production, food availability and climate stability (Saenger & Snedaker 1993; Dillon, Wang &
6 Huey 2010). The different vulnerability of the two species and particularly the increased
7 metabolism in *U. urvillei* confirm the role of air-breathing mode in enhancing the thermal
8 tolerance in amphibious animals (Verberk & Bilton 2013; Giomi et al. 2014).

9 The species-specific climate induced response seems trace the species genetic connectivity
10 (Rius et al. 2014). *Uca* species seem to have strong gene flow along the latitudinal
11 geographical gradient, demonstrating no genetic structure along the east African coast (Silva
12 et al. 2010a; Levinton & Mackie 2013) in contrast with *P. guttatum* exhibiting a significant
13 spatial genetic structuring between low and high latitude populations (Silva et al. 2010b), that
14 could have influenced the thermal specialization among the populations through local
15 adaptation to regional climate.

16 Based on the present results, we would propose a conceptual model to draft the differences in
17 thermal vulnerability between thermal specialist and thermal generalist species (Fig. 6). Since
18 such model is based on the comparisons between two populations at two latitudes future
19 studies are needed to validate and extend the proposed rationales.

20 Across its entire geographical distribution, a thermal generalist will maintain a wide window
21 of thermal tolerance and the capability of enduring acute warming. Populations at higher
22 latitudes, however, are closer to the geographic limits of distribution of the species, and
23 encounter thermal extremes less frequently than equatorial populations. Thus, in accordance
24 with previous predictions based on meta-analyses, if climate warming becomes permanent,
25 subtropical populations of the thermal generalist may cope better than low latitude

1 populations that are already closer to the edges of the thermal tolerances of the species
2 (Deutsch et al. 2008, 2010; Sunday, Bates & Dulvy 2012). In contrast, the thermal specialist is
3 more sensitive to temperature variations and locally adapted to regional climate. Thus, while
4 stenothermal populations at low latitudes are able to withstand increased heat stress, as stated
5 for high-latitude species by Sunday et al. (2012), those specialised to a subtropical climate
6 appear vulnerable to a long term warming. Further, we should also take into account that low
7 latitudes in this study were also the centre of distribution for these species, implying that other
8 factors must contribute to the well-being of the tropical populations. Perhaps populations at
9 the edge of their distribution have to cope with increased habitat degradation and therefore
10 genetic isolation compared to their tropical counterparts (Pearson et al. 2009).

11 In conclusion, a detailed appreciation of thermal induced response, extended at population
12 level, appears to be crucial to predicting the resilience to climate anomalies in species with
13 large geographical distributions. The predictions of higher vulnerability for low latitude
14 populations based on meta-analyses are over simplistic and our findings highlight the need to
15 integrate population level studies to compare local thermal response at different latitudes and
16 allow accurate forecasts of animal persistence and resilience in the face of climate change.

17

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13

1 **References**

- 2 Anderson, M.J. 2001. A new method for non-parametric multivariate analysis of variance. -
3 Austral Ecol. 26: 32-46.
- 4 Anderson, M.J. et al. 2008. PERMANOVA+ for PRIMER: guide to software and statistical
5 methods. PRIMER-E Ltd, Plymouth, UK.
- 6 Angilletta, M.J. 2009. Thermal Adaptation: a theoretical and empirical synthesis. Oxford
7 Univ. Press, Oxford.
- 8 Bartolini, F. et al. 2013. Climate change reduces offspring fitness in littoral spawners: a study
9 integrating organismic response and long-term time-series. - Glob. Chang. Biol. 19: 373-386.
- 10 Beaugrand, G. 2009. Decadal changes in climate and ecosystems in the North Atlantic Ocean
11 and adjacent seas. – Deep-Sea Res. II, 56(8-10): 656-673.
- 12 Borja, A. 2014. Grand challenges in marine ecosystems ecology. - Mar. Eco. Ecol 1:1.
- 13 Bozinovic, F. et al. 2011. Physiological Correlates of Geographical Range in Animals. - Annu.
14 Rev. Ecol. Evol. Syst. 42: 155-179.
- 15 Byrne, M. 2011. Impact of ocean warming and ocean acidification on marine invertebrate life
16 history stages: Vulnerabilities and potential for persistence in a changing ocean. - Ocean. Mar.
17 Bio. 49: 1-42.
- 18 Cannicci, S., et al. 2008. Faunal impact on vegetation structure and ecosystem function in
19 mangrove forests: a review. - Aquat. Bot. 89(2): 186-200.

- 1 De la Iglesia, H. O. et al. 1994. Burrow plugging in the crab *Uca uruguayensis* and its
2 synchronization with photoperiod and tides. – *Physiol. Behav.* 55(5): 913-919.
- 3 Deutsch, C.A. et al. 2008. Impacts of climate warming on terrestrial ectotherms across
4 latitude. - *PNAS* 105(18): 6668-6672.
- 5 Dillon, M.E. et al. 2010. Global metabolic impacts of recent climate warming. - *Nature*,
6 467:704-707.
- 7 Doney, S.C. et al. 2012. Climate change impacts on marine ecosystems. - *Annu Rev Mar Sci.*,
8 4: 11-37.
- 9 Farrell, A. 2009. Environment, antecedents and climate change: lessons from the study of
10 temperature physiology and river migration of salmonids. - *J. Exp. Biol.* 212: 3771-3780.
- 11 Farrelly, C.A. and Greenaway, P. 1994. Gas exchange through the lungs and gills in air-
12 breathing crabs. - *J. Exp. Biol.* 187: 113-130.
- 13 Flores, A.A.V. et al. 2002. Sexual maturity, reproductive cycles, and juvenile recruitment of
14 *Perisesarma guttatum* (Brachyura, Sesarmidae) at Ponta Rasa mangrove swamp, Inhaca
15 Island, Mozambique. - *J. Crust. Biol.* 22(1): 143-156.
- 16 Fratini, S. et al. 2005. Tree-climbing mangrove crabs: a case of convergent evolution. - *Evol.*
17 *Ecol. Res.* 7: 219-233.
- 18 Frederich, M. & Pörtner, H.O. 2000. Oxygen limitation of thermal tolerance defined by
19 cardiac and ventilatory performance in spider crab, *Maja squinado*. – *Am. J. Physiol.* 279:
20 1531-1538.

- 1 Giomi, F. and Pörtner, H.O. 2013. A role for haemolymph oxygen capacity in heat tolerance
2 of eurythermal crabs. - *Frontiers Physiol.* 4: 110.
- 3 Giomi, F. et al. 2014. Improved heat tolerance in air drives the recurrent evolution of air
4 breathing. - *P. Roy. Soc B-Biol.* 281: 1782. doi: 10.1098/rspb.2013.2927.
- 5 Hartnoll, R.G. 1975. The Grapsidae and Ocypodidae (Decapoda, Brachyura) of Tanzania. - *J.*
6 *Zoo.* 177: 305-328.
- 7 Hartnoll, R.G. 1988. The eco-ethology of mangroves. In *Behavioural Adaptations to Intertidal*
8 *Life.* - NATO ASI Series, Series A, Vol. 151 (eds G. Chelazzi & M. Vannini), pp. 477-489.
9 Plenum Press, New York.
- 10 Helmuth, B. et al. 2006. Mosaic patterns of thermal stress in the rocky intertidal zone:
11 implications for climate change. - *Ecol. Monogr.* 76: 461-479.
- 12 Helmuth, B., et al. 2010. Organismal climatology: analyzing environmental variability at
13 scales relevant to physiological stress. - *J. Exp. Biol.* 213: 995-1003.
- 14 Helmuth, B., et al. 2002. Climate change and latitudinal patterns of intertidal thermal stress.
15 *Science* 298: 1015-1017.
- 16 Helmuth, B. et al. 2006. Living on the edge of two changing worlds: forecasting the responses
17 of rocky intertidal ecosystems to climate change. - *Annu. Rev. Ecol., Evol. S.* 37: 373-404.
- 18 Hoegh-Guldberg, O. and Bruno, J.F. 2010. The impact of climate change on the world's
19 marine ecosystems. - *Science* 328: 1523-1528.

- 1 Hoepffner, N. et al. 2014. Thermal Infrared Remote Sensing and Sea Surface Temperature of
2 Marine and Coastal Waters Around Africa. *Remote Sensing of the African Seas* 55-73. Doi:
3 10.1007/978-94-017-8008-7_3.
- 4 Holguin, G. et al. 2001. The role of sediment microorganisms in the productivity,
5 conservation, and rehabilitation of mangrove ecosystems: an overview. – *Biol. Fert. Soils*
6 33(4): 265-278.
- 7 Huey, R.B., et al. 2012. Predicting organismal vulnerability to climate warming: roles of
8 behaviour, physiology and adaptation. - *P. Roy. Soc B-Biol.* 367: 1665-1679.
- 9 Kearney, M. & Porter, W.P. 2009. Mechanistic niche modelling: combining physiological and
10 spatial data to predict species' ranges. *Ecol. Lett.* - 12: 334–350.
- 11 Kelly, M. W. et al. 2012. Limited potential for adaptation to climate change in a broadly
12 distributed marine crustacean. - *P. Roy. Soc B-Biol.* 279: 349-356.
- 13 Kitheka, J.U. 1996. Water circulation and coastal trapping of brackish water in a tropical
14 mangrove-dominated bay in Kenya. - *Limnol. Oceanogr.* 41(1): 169-176.
- 15 Kristensen, E. 2008. Mangrove crabs as ecosystem engineers; with emphasis on sediment
16 processes. *J. Sea Res.* 59(1): 30-43.
- 17 Kristensen, E. & Alongi, D.M. 2006. Control by fiddler crabs (*Uca vocans*) and plant roots
18 (*Avicennia marina*) on carbon, iron, and sulfur biogeochemistry in mangrove sediment. -
19 *Limnol. Oceanogr.* 51(4): 1557-1571.
- 20 Lee, H.J. et al. 2005. Burrow air phase maintenance and respiration by the mudskipper
21 *Scartelaos histophorus* (Gobiidae: Oxudercinae). - *J. Exp. Biol.*, 208(1): 169-177.

- 1 Lee, S.Y. 1998. Ecological role of grapsid crabs in mangrove ecosystems: a review. - Mar.
2 Freshwater Res. 49(4): 335-343.
- 3 Lee, S.Y. 2008. Mangrove macrobenthos: assemblages, services, and linkages. - J. Sea Res.
4 59(1): 16-29.
- 5 Levinton, J. and Mackie, J. 2013. Latitudinal diversity relationships of fiddler crabs:
6 biogeographic differences united by temperature. - Global. Ecol. Biogeogr. 22(9): 1050-1059.
7 Doi: 10.1111/geb.12064.
- 8 Litulo, C. 2004. Fecundity of the Pantropical Fiddler Crab *Uca annulipes* (H.Milne Edwards,
9 1837) (Brachyura: Ocypodidae) at Costa do Sol Mangrove, Maputo Bay, Southern
10 Mozambique. - Hydrobiologia 525: 167-173.
- 11 Litulo, C. 2005. Population structure and reproductive biology of the fiddler crab *Uca urvillei*
12 (Brachyura: Ocypodidae) in Maputo Bay (south Mozambique). - J. Nat. Hyst. 39(25): 2307-
13 2318.
- 14 Macnae, W. 1963. Mangrove swamps in South Africa. - J. Ecol. 51: 1-25.
- 15 Macnae, W. 1968. A general account of the flora and fauna of the mangrove swamps and
16 forests in the Indo-Pacific Region. - Adv. Mar. Bio. 6: 73-270.
- 17 Marshall, D.J. et al. 2010. Non-climatic thermal adaptation: implications for species'
18 responses to climate warming. - Biol. Lett. 6(5): 669-673.
- 19 McClanahan, T. R. 2014. Decadal coral community reassembly on an African fringing reef. -
20 Coral Reefs. doi: 10.1007/s00338-014-1178-6.

- 1 Morley, S.A. et al. 2009. Geographic variation in thermal tolerance within Southern Ocean
2 marine ectotherms. - *Comp. Biochem. Physiol. A* 153: 154-161.
- 3 Morris, S. 2002. The ecophysiology of air-breathing in crabs with special reference to
4 *Gecarcoidea natalis*. - *Comp. Biochem. Physiol. B*. 131: 559-570.
- 5 Nielsen, M.G. 2011. Ants (Hymenoptera: Formicidae) of mangrove and other regularly
6 inundated habitats: life in physiological extreme. - *Myrmecological News*. 14: 113-121.
- 7 Nielsen, M.G. et al. 2003. Carbon dioxide concentrations in the nests of the mud-dwelling
8 mangrove ant *Polyrhachis sokolova* Forel (Hymenoptera: Formicidae). - *Aust. J. Entomol.*
9 42(4): 357-362.
- 10 Pearson, G.A. et al. 2009. Frayed at the edges: selective pressure and adaptive response to
11 abiotic stressors are mismatched in low diversity edge populations. - *J. Ecol.*, 97: 450-462.
- 12 Pörtner, H.O. and Farrell, A.P. 2008. Ecology: physiology and climate change. - *Science* 322:
13 690-691.
- 14 Pörtner, H.O. and Knust, R. 2007. Climate change affects marine fishes through the oxygen
15 limitation of thermal tolerance. - *Science* 315: 95-97.
- 16 Pörtner, H.O. 2001. Climate change and temperature-dependent biogeography: oxygen
17 limitation of thermal tolerance in animals. - *Naturwissenschaften* 88(4): 137-146.
- 18 Pörtner, H.O. 2002. Climate variations and the physiological basis of temperature dependent
19 biogeography: systemic to molecular hierarchy of thermal tolerance in animals. - *Comp.*
20 *Biochem. Physiol. A* 132(4): 739-761.

- 1 Pörtner, H.O. 2006 Climate-dependent evolution of Antarctic ectotherms: an integrative
2 analysis. - *Deep Sea Res. II*: 53(8-10): 1071-1104.
- 3 Pörtner, H.O. 2010. Oxygen-and capacity-limitation of thermal tolerance: a matrix for
4 integrating climate-related stressor effects in marine ecosystems. – *J. Exp. Biol.* 213: 881-893.
- 5 Pörtner, H.O. & Giomi, F. 2013. Nothing in experimental biology makes sense except in the
6 light of ecology and evolution. – *J. Exp. Biol.* 216: 4494-4495.
- 7 R Development Core Team 2012. R: A Language and Environment for Statistical Computing.
8 R Foundation for Statistical Computing, Vienna, Austria. Available at: URL [http://www.R-](http://www.R-project.org)
9 [project.org](http://www.R-project.org).
- 10 Reusch, T. B. 2014. Climate change in the oceans: evolutionary versus phenotypically plastic
11 responses of marine animals and plants. - *Evol. Appl.* 7(1): 104-122.
- 12 Rius M. et al. 2014. Range expansions across ecoregions: interactions of climate change,
13 physiology and genetic diversity. - *Global Ecol Biogeogr.* 23: 76-88.
- 14 Rockström, J. et al. 2009. A safe operating space for humanity. - *Nature* 461(7263): 472-475.
- 15 Roznik, E.A. and Alford, R.A. 2012. Does waterproofing Thermochron iButton dataloggers
16 influence temperature readings? - *J. Therm. Biol.* 37(4): 260-264.
- 17 Saenger, P. and Snedaker, S.C. 1993. Pantropical trends in mangrove above-ground biomass
18 and annual litterfall. - *Oecologia*, 96(3): 293-299.
- 19 Sagarin, R.D. et al. 1999. Climate-related change in an intertidal community over short and
20 long time scales. - *Ecol. Monogr.* 69(4): 465-490.

- 1 Schurmann, H. and Steffensen, J. 1992. Lethal oxygen levels at different temperatures and the
2 preferred temperature during hypoxia of the Atlantic cod, *Gadus morhua* L. – J. Fish Biol.
3 41(6): 927-934.
- 4 Shih, T. et al. 2005. Chimney Building by Male *Uca formosensis* Rathbun, 1921 (Crustacea:
5 Decapoda: Ocypodidae) after Pairing: A New Hypothesis for Chimney Function. – Zool.
6 Stud. 44(2): 242-251.
- 7 Silva, I.C. et al. 2010a. Lack of population structure in the fiddler crab *Uca annulipes* along
8 an East African latitudinal gradient: Genetic and morphometric evidence. - Mar. Biol. 157(5):
9 1113-1126.
- 10 Silva, I.C. et al. 2010b. Genetic and morphological differentiation of the mangrove crab
11 *Perisesarma guttatum* (Brachyura: Sesarmidae) along an East African latitudinal gradient. -
12 Biol. J. Linn. Soc. 99: 28-46.
- 13 Skov, M.W. & Hartnoll, R.G. 2001. Comparative suitability of binocular observation, burrow
14 counting and excavation for the quantification of the mangrove fiddler crab *Uca annulipes* (H.
15 Milne Edwards). - Hydrobiologia, 449: 201-212.
- 16 Slim, F. et al. 1997. Leaf litter removal by the snail *Terebralia palustris* (Linnaeus) and
17 sesarmid crabs in an East African mangrove forest (Gazi Bay, Kenya). – J. Exp. Mar. Biol.
18 Ecol. 215(1): 35-48.
- 19 Smit, A. J. et al. 2013. A Coastal Seawater Temperature Dataset for Biogeographical Studies:
20 Large Biases between In Situ and Remotely-Sensed Data Sets around the Coast of South
21 Africa.- PloS one, 8(12), e81944.

- 1 Somero, G.N. 2002. Thermal physiology and vertical zonation of intertidal animals: optima,
2 limits, and costs of living. - *Integr. Comp. Biol.* 42(4): 780-789.
- 3 Somero, G.N. 2012. The physiology of global change: linking patterns to mechanisms. *Annu.*
4 *Rev. Mar. Sci.* 4: 39-61.
- 5 Sousa, W.P. and Dangremond, E.M. (2011) Trophic Interactions in Coastal and Estuarine
6 Mangrove Forest Ecosystems. *Treatise on Estuarine and Coastal Science* 6: 43-93 (eds E.
7 Wolanski & D.S.). Waltham Academic.
- 8 Stebbins, R.C. and Kalk, M. 1961. Observations on the natural history of the mud-skipper,
9 *Periophthalmus sobrinus*. - *Copeia* 1: 18-27.
- 10 Stevens, M.M. et al. 2010. Oxygen limitation and thermal tolerance in two terrestrial arthropod
11 species. *J Exp. Biol.*, 213: 2209-2218.
- 12 Sunday, J. M. et al. 2014. Thermal-safety margins and the necessity of thermoregulatory
13 behavior across latitude and elevation. - *PNAS* 111(15): 5610-5615.
- 14 Sunday, J.M. et al. 2011. Global analysis of thermal tolerance and latitude in ectotherms. - *P.*
15 *Roy. Soc B-Biol.* 278(1713): 1823-1830.
- 16 Sunday, J.M. et al. 2012. Thermal tolerance and the global redistribution of animals. - *Nature*
17 *Climate Change* 2(9): 686-690.
- 18 Terblanche, J.S. et al. 2011. Ecologically relevant measures of tolerance to potentially lethal
19 temperatures. - *J. Exp. Biol.* 214: 3713-3725.
- 20 Tomlinson, P.B. 1995. *The Botany of Mangroves*. Cambridge Univ. Press, Cambridge.

1 Verberk, W.C. and Bilton D.T. 2013. Respiratory control in aquatic insects dictates their
2 vulnerability to global warming. *Biol. Lett.* 9(5): 20130473.
3 <http://dx.doi.org/10.1098/rsbl.2013.0473>.

4 Verberk, W. C., & Atkinson, D. 2013. Why polar gigantism and Palaeozoic gigantism are not
5 equivalent: effects of oxygen and temperature on the body size of ectotherms. - *Functional*
6 *Ecology* 27(6):1275-1285.

7 Verberk, W.C. et al. 2011. Oxygen supply in aquatic ectotherms: partial pressure and
8 solubility together explain biodiversity and size patterns. – *Ecology* 92(8): 1565-1572.

9 Walther, G.R. et al. 2002. Ecological responses to recent climate change. - *Nature* 416: 389-
10 395.

11 Webb, E.L. et al. 2013. A global standard for monitoring coastal wetland vulnerability to
12 accelerated sea-level rise. - *Nature Climate Change* 3(5): 458-465.

13 Williams, S.E. et al. 2008. Towards an integrated framework for assessing the vulnerability of
14 species to climate change. - *PLoS Biol.* 6: e325.

15 Yamaguchi, T. (2001) The mating system of the fiddler crab, *Uca lactea* (Decapoda,
16 Brachyura, Ocypodidae). - *Crustaceana* 74: 389-399.

17

1 **Figure Legends**

2 Figure. 1. Study sites along East African coast. The equatorial site was Gazi bay (Kenya) and
3 corresponds to the centre of distribution of the studied crabs. The sub-tropical site was
4 Mgazana River (South Africa) and corresponds to the southward limit of the mangrove biome.

5

6 Figure 2. Kenyan (31st October 2011 – 19th December 2011) and South African (16th January
7 2012 – 5th March 2012) tidal regimes and climatic time series of eulittoral belts occupied by
8 *Perisesarma guttatum* (a,c respectively) and *Uca urvillei* (b,d respectively). Black lines are
9 the daily averages of data recorded by i-buttons.

10

11 Figure 3. Oxygen consumption rate of and during air-breathing and water-breathing of
12 *Perisesarma guttatum* (a-b) and *Uca urvillei* (c-d); open circles South African population,
13 solid circles Kenyan population. Significant 3rd order polynomial regressions and the 95%
14 confidence intervals are represented with continuous lines, grey for South African and black
15 Kenyan population.

16

17 Figure 4. Arterial (grey circle) and venous (black circle) haemolymph PO₂ of Kenyan
18 population of *U. urvillei* and *P. guttatum* during air-breathing (a, b) and water-breathing (c,
19 d); e and f report the arterio-venous differences in PO₂ and provide indication on the relative
20 efficiency of the different respiratory strategies. Each point represents the mean \pm SE (n = 15
21 for PO₂).

22

23 Figure 5. Body temperature of Kenyan (KY) and South African (ZA) *P. guttatum* and *U.*
24 *urvillei* populations during activity at low tide, in the burrow (a, b, g, h), under direct exposure

1 to the sun (c, d, i, j), in shade but always on the ground (e, f, k, l). Grey area represents 95%
2 confidence interval, the bisect represents the isotherm line.

3

4 Figure. 6. Conceptual model illustrating different thermal adaptive strategies along a
5 geographical gradient. Temperature-dependent performances (curves depict responses to acute
6 temperature fluctuation) are maintained within the entire climatic range (horizontal bars) and
7 maximised around the optimum which generally represents the preferred temperature for
8 activity. Acute heat events (vertical bars) beyond the range of environmental temperatures
9 affect organismal fitness limiting the capability to sustain performance. Thermal generalists
10 undergo pronounced temperature fluctuations both on a daily and seasonal basis and manifest
11 a pronounced eurythermy throughout their distribution. This category shows a low thermal
12 vulnerability, even to anomalous warming, and a single pattern of responses to temperature
13 fluctuation along the geographic gradient (further supported by wide ranging gene flow
14 among populations). Thermal specialists manifest distinctive tolerance ranges among different
15 regions as a result of climate induced response to local climatic signatures and the genetic
16 differentiation of phylogeographic clades. Tropical stenotherms are particularly adapted to a
17 constantly hot climate and may endure irregular increases of environmental temperatures. In
18 contrast, temperate-adapted populations have a broader thermal tolerance, likely due to the
19 larger fluctuation of environmental temperature, but are more vulnerable to the effects of
20 global warming.

21

22

Supplementary material

Table S1

MO₂. ANCOVA on Species, Medium and Region as covariate to test for differences in slopes among Temperatures. The degrees of freedom, df, the value of the F statistic value and its probability level, p, are reported.

| Source | Df | F value | Pr(>F) |
|----------------------------------|----------|------------------|---------------|
| Temp | 1 | 1498.3011 | 0.0001 |
| Medium | 1 | 0.4386 | 0.5079 |
| Region | 1 | 14.7494 | 0.0001 |
| Species | 1 | 605.6 | 0.0001 |
| Temp × Medium | 1 | 0.7418 | 0.3893 |
| Temp × Region | 1 | 31.1718 | 0.0001 |
| Medium × Region | 1 | 155.5887 | 0.0001 |
| Temp × Species | 1 | 0.0304 | 0.8616 |
| Medium × Species | 1 | 4.2552 | 0.0394 |
| Region × Species | 1 | 4.097 | 0.0432 |
| Temp × Medium × Region | 1 | 2.2439 | 0.1344 |
| Temp × Medium × Species | 1 | 0.5741 | 0.4488 |
| Temp × Region × Species | 1 | 19.1195 | 0.0001 |
| Medium × Region × Species | 1 | 29.5014 | 0.0001 |
| Residuals | 1162 | | |

Table S2

PO₂. ANCOVA on Haemolymph, Species and Medium as covariate to test for differences in slopes among Temperatures. The degrees of freedom, df, the value of the F statistic value and its probability level, p, are reported.

| Source | Df | F value | Pr(>F) |
|--------------------------|----------|-----------------|---------------|
| Temp | 1 | 202.0208 | 0.0001 |
| Medium | 1 | 0.4623 | 0.4972 |
| Haemolymph | 1 | 204.6215 | 0.0001 |
| Species | 1 | 60.5157 | 0.0001 |
| Temp×Medium | 1 | 2.682 | 0.1027 |
| Temp×Haemolymph | 1 | 1.5349 | 0.2165 |
| Medium×Haemolymph | 1 | 34.7572 | 0.0001 |
| Temp×Species | 1 | 1.0087 | 0.3161 |
| Medium×Species | 1 | 252.3468 | 0.0001 |
| Haemolymph×Species | 1 | 2.5825 | 0.1092 |

| | | | |
|---------------------------------------|----------|----------------|---------------|
| Temp×Medium×Haemolymph | 1 | 0.2618 | 0.6093 |
| Temp×Medium×Species | 1 | 0.0039 | 0.9504 |
| Temp×Haemolymph×Species | 1 | 0.3402 | 0.5602 |
| Medium×Haemolymph×Species | 1 | 29.3037 | 0.0001 |
| Temp×Medium×Haemolymph×Species | 1 | 4.1758 | 0.0420 |
| Residuals | 268 | | |

Table S3a

P. guttatum Body Temperature. PERMANOVA test on differences in body and environmental Temperature across Species, Place and Temp, factors treated as fixed and orthogonal. The degrees of freedom, df, Variance, MS, the value of the pseudo-F statistic value and its probability level, P, are reported.

| Source | df | MS | Pseudo-F | P(MC) |
|-----------------------|----------|---------------|---------------|---------------|
| Region | 1 | 24.104 | 8.9749 | 0.003 |
| Temp | 1 | 0.87993 | 0.32764 | 0.5644 |
| Place | 3 | 123.26 | 45.897 | 0.0001 |
| Region Temp | 1 | 7.9532 | 2.9613 | 0.0815 |
| Region × Place | 3 | 8.4405 | 3.1428 | 0.0238 |
| Temp × Place | 3 | 4.0008 | 1.4897 | 0.2182 |
| Region × Temp × Place | 3 | 0.83232 | 0.30991 | 0.8221 |
| Res | 528 | 2.6857 | | |
| Total | 543 | | | |

Table S3b

U. urvillei Body Temperature. PERMANOVA test on differences in body and environmental Temperature across Species, Place and Temp, factors treated as fixed and orthogonal. The degrees of freedom, df, Variance, MS, the value of the pseudo-F statistic value and its probability level, P, are reported.

| Source | df | MS | Pseudo-F | p |
|---------------|----------|---------------|---------------|---------------|
| Region | 1 | 1010.7 | 278.5 | 0.0001 |
| Temp | 1 | 10.891 | 3.001 | 0.0835 |
| Place | 3 | 635.41 | 175.08 | 0.0001 |
| Region × Temp | 1 | 10.075 | 2.7762 | 0.0952 |

| | | | | |
|-----------------------|----------|--------------|---------------|---------------|
| Region × Place | 3 | 33.46 | 9.2197 | 0.0002 |
| Temp × Place | 3 | 3.4288 | 0.9448 | 0.421 |
| Region × Temp × Place | 3 | 7.6617 | 2.1111 | 0.1006 |
| Res | 504 | 3.6292 | | |
| Total | 519 | | | |

For Review Only

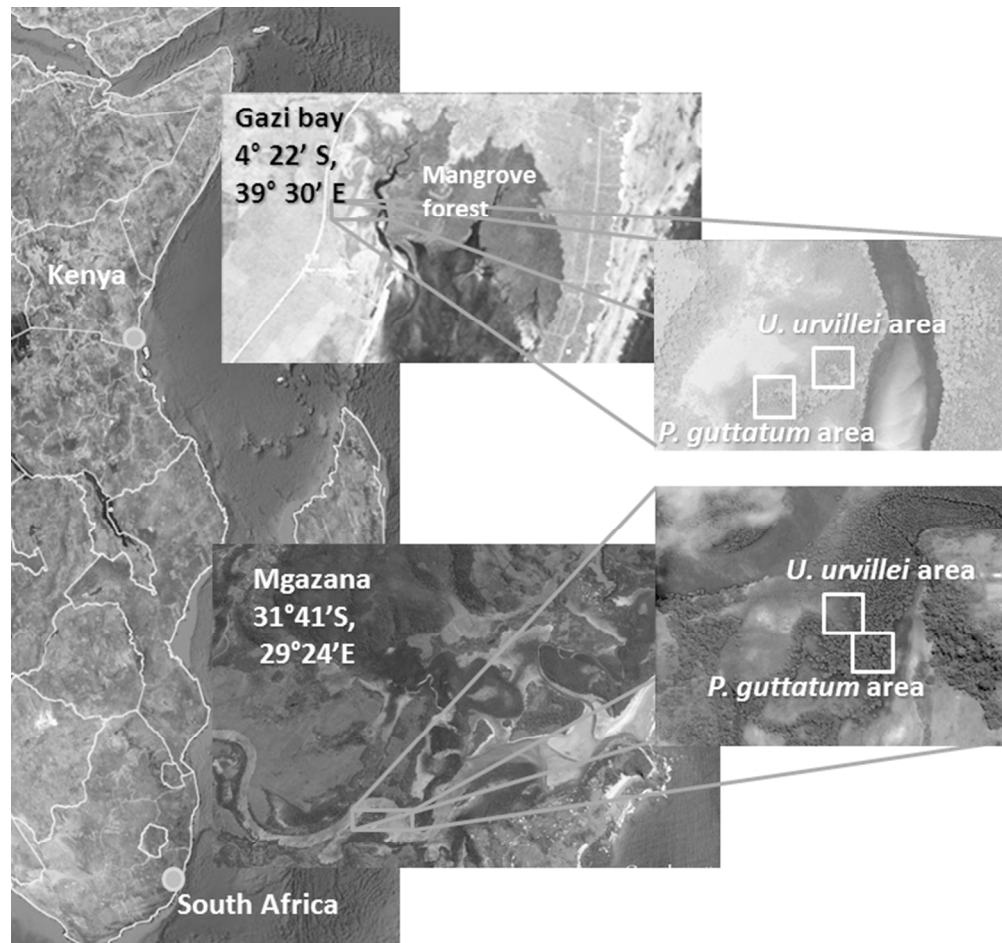


Figure. 1. Study sites along East African coast. The equatorial site was Gazi bay (Kenya) and corresponds to the centre of distribution of the studied crabs. The sub-tropical site was Mgazana River (South Africa) and corresponds to the southward limit of the mangrove biome.
271x254mm (72 x 72 DPI)



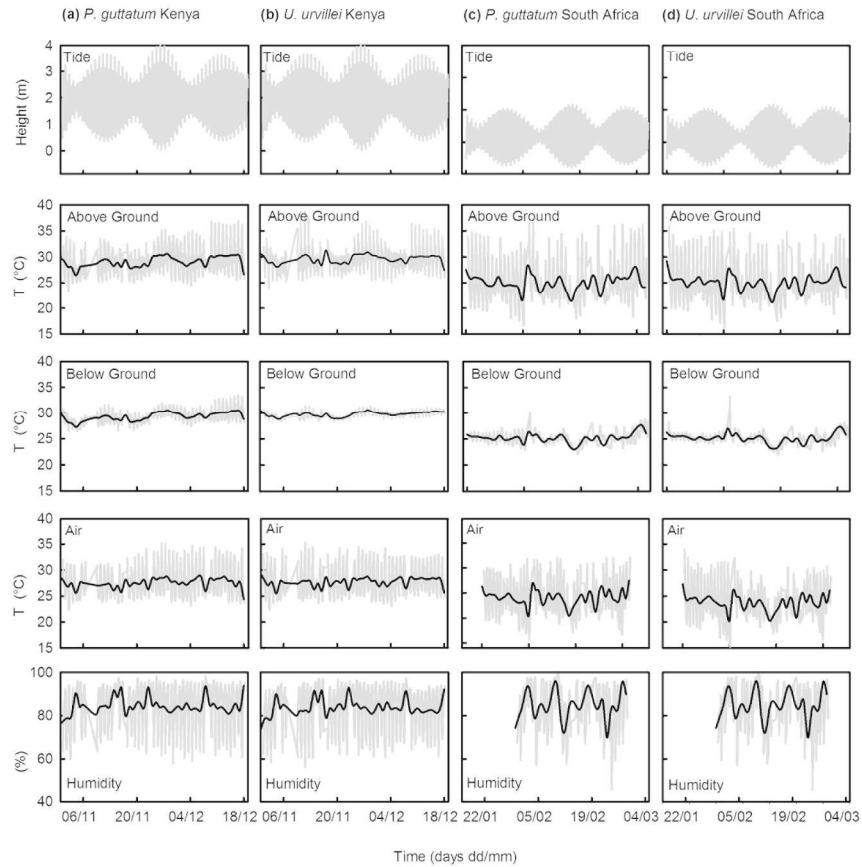


Figure 2. Kenyan (31st October 2011 – 19th December 2011) and South African (16th January 2012 – 5th March 2012) tidal regimes and climatic time series of eulittoral belts occupied by *Perisesarma guttatum* (a,c respectively) and *Uca urvillei* (b,d respectively). Black lines are the daily averages of data recorded by i-buttons.

212x296mm (300 x 300 DPI)

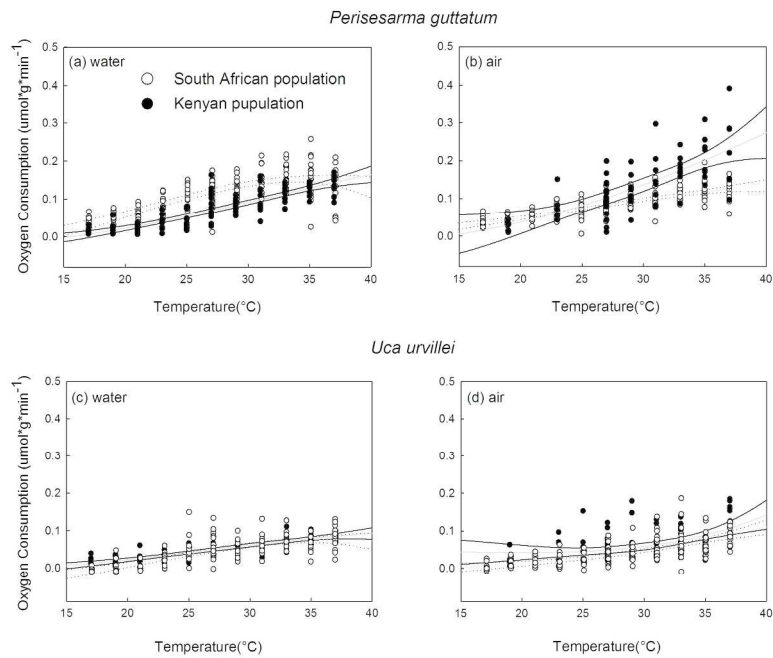


Figure 3. Oxygen consumption rate of and during air-breathing and water-breathing of *Perisesarma guttatum* (a-b) and *Uca urvillei* (c-d); open circles South African population, solid circles Kenyan population. Significant 3rd order polynomial regressions and the 95% confidence intervals are represented with continuous lines, grey for South African and black Kenyan population.

210x296mm (300 x 300 DPI)

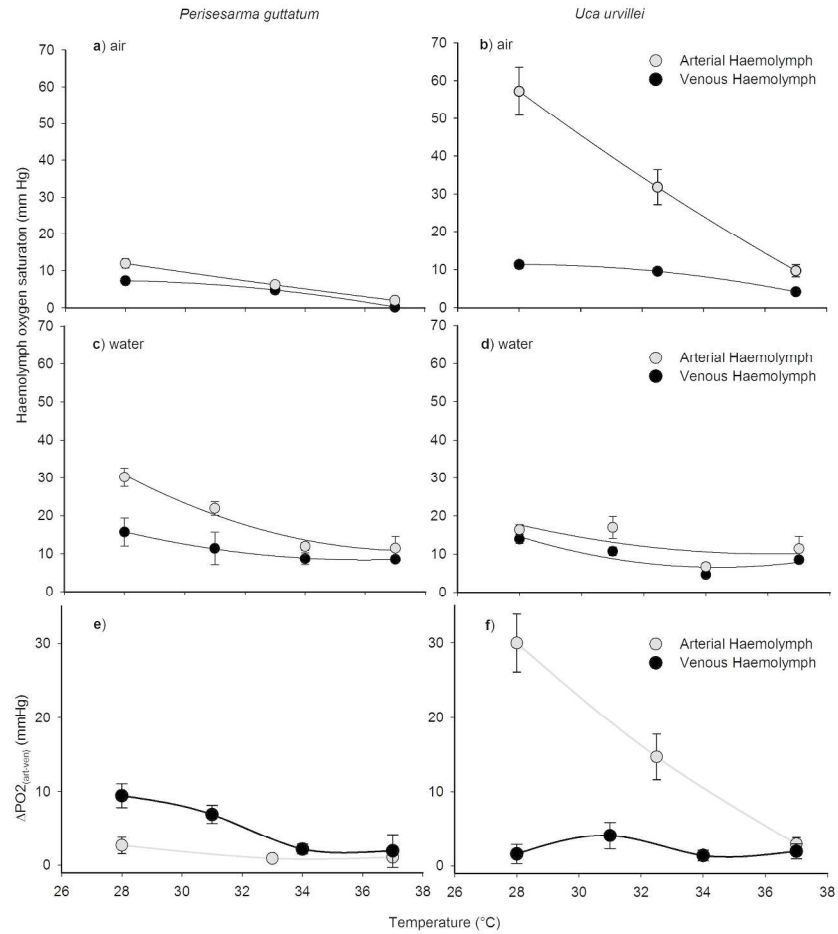


Figure 4. Arterial (grey circle) and venous (black circle) haemolymph PO₂ of Kenyan population of *U. urvillei* and *P. guttatum* during air-breathing (a, b) and water-breathing (c, d); e and f report the arterio-venous differences in PO₂ and provide indication on the relative efficiency of the different respiratory strategies. Each point represents the mean \pm SE (n = 15 for PO₂).

210x296mm (300 x 300 DPI)

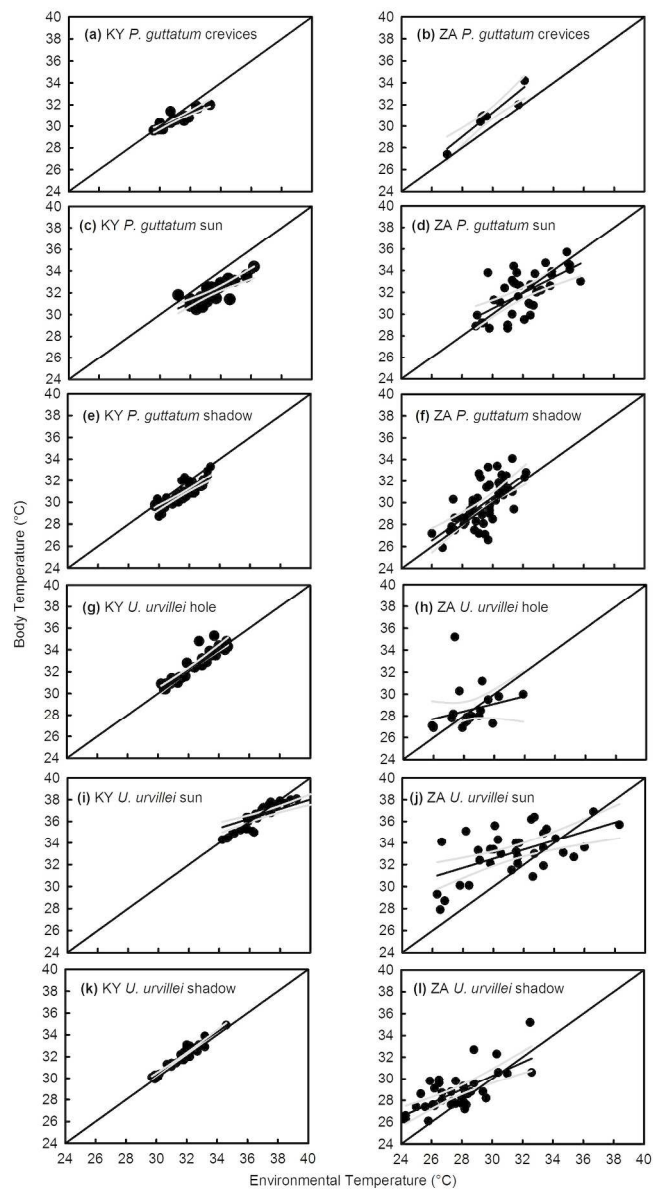


Figure 5. Body temperature of Kenyan (KY) and South African (ZA) *P. guttatum* and *U. urvillei* populations during activity at low tide, in the burrow (a, b, g, h), under direct exposure to the sun (c, d, i, j), in shade but always on the ground (e, f, k, l). Grey area represents 95% confidence interval, the bisect represents the isotherm line.

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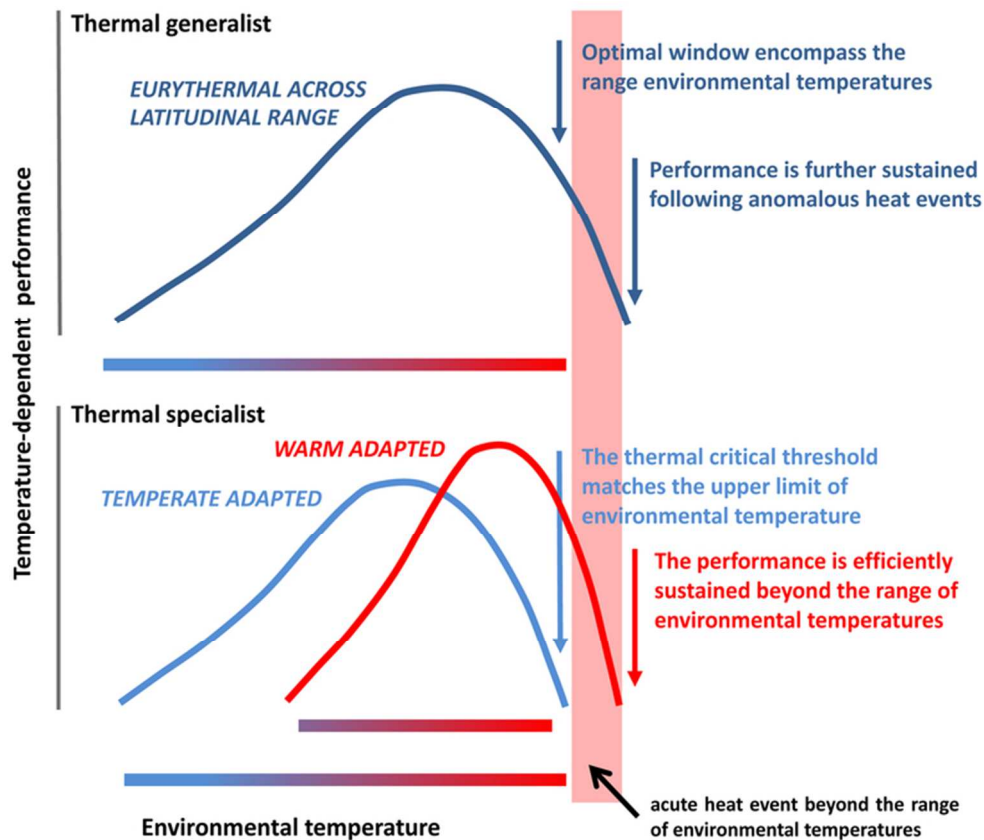


Figure. 6. Conceptual model illustrating different thermal adaptive strategies along a geographical gradient. Temperature-dependent performances (curves depict responses to acute temperature fluctuation) are maintained within the entire climatic range (horizontal bars) and maximised around the optimum which generally represents the preferred temperature for activity. Acute heat events (vertical bars) beyond the range of environmental temperatures affect organisal fitness limiting the capability to sustain performance. Thermal generalists undergo pronounced temperature fluctuations both on a daily and seasonal basis and manifest a pronounced eurythermy throughout their distribution. This category shows a low thermal vulnerability, even to anomalous warming, and a single pattern of responses to temperature fluctuation along the geographic gradient (further supported by wide ranging gene flow among populations). Thermal specialists manifest distinctive tolerance ranges among different regions as a result of climate induced response to local climatic signatures and the genetic differentiation of phylogeographic clades. Tropical stenotherms are particularly adapted to a constantly hot climate and may endure irregular increases of environmental temperatures. In contrast, temperate-adapted populations have a broader thermal tolerance, likely due to the larger fluctuation of environmental temperature, but are more vulnerable to the effects of global warming.

68x58mm (300 x 300 DPI)

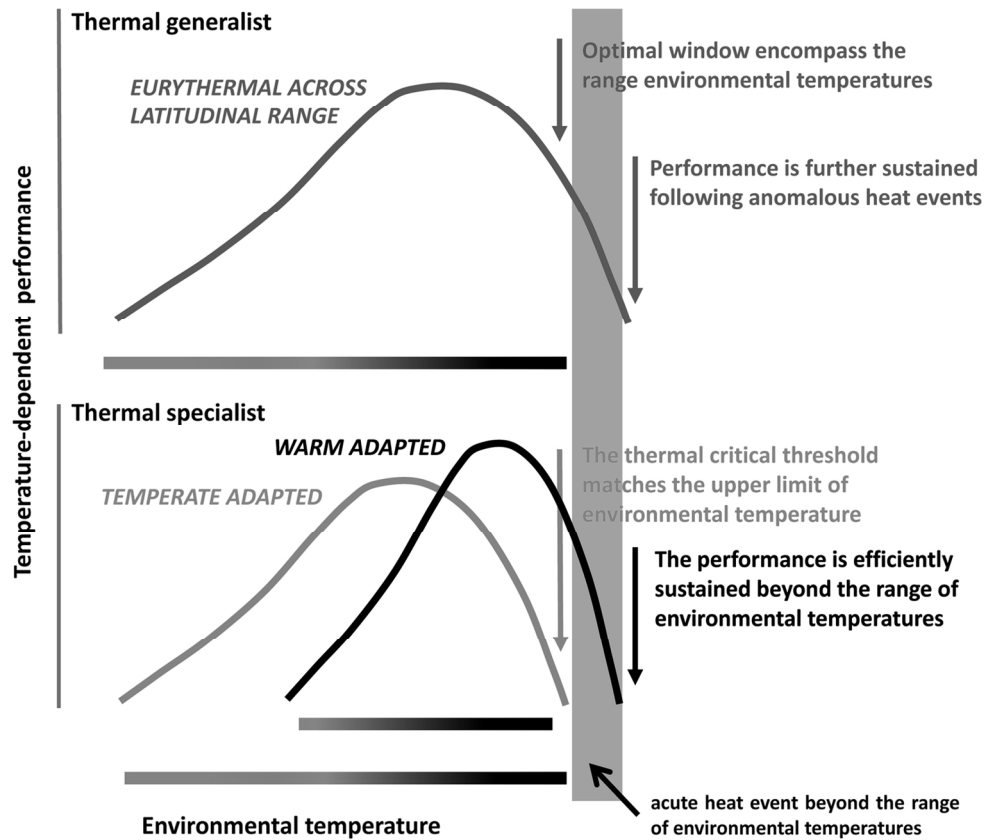


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144x122mm (300 x 300 DPI)