

The seasonal response of metabolic rate to projected climate change scenarios in aquatic amphipods

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ABSTRACT

The responses of organisms to climate change are mediated primarily by its impact on their metabolic rates, which, in turn, drive various biological and ecological processes. Although there have been numerous seminal studies on the sensitivity of metabolic rate to temperature, little is empirically known about how this rate responds to seasonal temperature ranges and beyond under conservative IPCC climate change scenarios. Here, we measured the SMR of the aquatic amphipod, *Gammarus insensibilis*, which served as our subject species, with body masses ranging from 0.20 to 7.74 mg ash free weight. We assessed the response of the SMR across nine temperature levels ranging from 12 to 30.2 °C. These temperatures match seasonal temperature norms, with an incremental increase of 0.6–1.2 °C above each seasonal baseline, as projected for the years 2040 and 2100 under the modest climate change scenarios. Overall, our findings showed that the effect of temperature on SMR varies with body mass, as indicated by a negative size-temperature interaction, with larger conspecifics exhibiting less sensitivity to temperature changes than smaller ones. From the cold to warm season, the SMR increased by an average of 14% °C⁻¹, with increases of 18.4% °C⁻¹ in smaller individuals and 11.4% °C⁻¹ in larger ones. The SMR of smaller individuals peaked at a 0.6 °C increase from the current summer baseline (15.08% °C⁻¹, Q10 = 4.2), while in larger ones it peaked with a 1.2 °C increase beyond autumn temperatures (14.9% °C⁻¹, Q10 = 3.9). However, at temperatures reflecting global warming that exceed summer temperatures, the SMR of larger individuals levelled off, while that of smaller ones continued to increase. Overall, our findings suggest that smaller-sized individuals have a broader thermal window for SMR performance, while the SMR of larger-sized ones will become increasingly constrained at summer temperatures as those summer temperatures become hotter.

1. Introduction

Climate change is causing substantial alterations from individual organism to ecosystem, posing a threat to biodiversity (Bruno et al., 2015). Warming has already left a global footprint on animal populations, with its effects being more pronounced on aquatic ones, among others, evidenced by alterations in life cycle events (Poloczanska et al., 2013), body size (Forster et al., 2012) and geographical distribution (Angilletta, 2009). These shifts are expected to continue in response to ongoing climate warming which are largely mediated by fundamental ecological and metabolic rules (Sheridan and Bickford, 2011). Climate change, with a mean temperature increase of ~1 °C over the last four decades, has already resulted in increases in metabolic rate of up to 20%

in ectotherms (Dillon et al., 2010; Seebacher et al., 2015).

Metabolic rate, that is, the rate at which energy is expended, stands as one of the primary and central traits of organisms that respond to climate change, given its temperature dependency (Brown et al., 2004). It is considered a fundamental variable in ecology and physiology, connecting individual to ecosystem-level processes via the currency of energy (Brandl et al., 2022). In this context, the Metabolic Theory of Ecology offers an approach to mechanistically estimate this central trait of individual energy requirements, underscoring body mass and temperature as the chief components of metabolic rate (Brown et al., 2004). Accordingly, whole-organism metabolic rate is predicted to scale with body mass according to a power law, specifically raised to the three-quarter power, and exponentially with temperature from 0 to

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40 °C, due to its influence on the kinetic energy of cellular components (Arrhenius, 1889; Gillooly et al., 2001). However, the interaction between metabolic rates and temperature is more complex than a uniform exponential increase, as it can be influenced by thermal acclimation, vary across seasonal temperature ranges, and be constrained by an upper thermal limit (Norin and Metcalfe, 2019; Schulte, 2015; Sinclair et al., 2016).

Thermal acclimation often leads organisms to adjust their metabolic rates, in response to sustained increases in ambient temperature. This compensates for direct thermodynamic effects and thus deviates from a simple exponential relationship (Sandblom et al., 2014). Additionally, the response of metabolic rate to temperature often depends on the thermal physiology of an organism, subject to influences from factors like body size, life stage, latitude and local climate (Leiva et al., 2018; Padilla et al., 2024; Shokri et al., 2022; Terblanche and Chown, 2006). The optimal physiological responses of ectotherms are seen within the temperature range they experience locally (Pörtner and Knust, 2007). Beyond this range organism performance and fitness likely decline (Angilletta, 2009; Vasseur et al., 2014), which can potentially alter the long-term prospect of the species regarding distributional range (Usui et al., 2023), body size (Audzijonyte et al., 2020), and even survival (Hochachka and Somero, 2002). Furthermore, in aquatic ecosystems, the rise in metabolic rate due to temperature increases will be accompanied by a decrease in oxygen availability, potentially constraining the aerobic capacities (Deutsch et al., 2015; Jermacz et al., 2020). These combined effects could influence the relationship between temperature and aerobic metabolism, as well as how metabolic rates scale with size (Rubalcaba et al., 2020). With warming, the oxygen supply may no longer meet the increase in oxygen requirement, leading the metabolic rate to reach its ceiling or boundary (Schulte, 2015). This could be particularly pronounced in larger conspecifics, which have higher absolute oxygen demands and a lower surface-area-to-volume ratio, hindering their ability to uptake oxygen from the water and transport it to metabolically active tissues (Rubalcaba et al., 2020; Shokri et al., 2022).

In light of this, it is important not only to comprehend the adaptive capacities of individual populations in the face of warming, but also to empirically quantify the extent of their metabolic responses. Such insights are essential for laying the groundwork for a more realistic estimation of the broader ecological impacts of climate change, as metabolic rate is intimately linked to both an organism's biology and behaviour, profoundly shaping its ecological interactions (Auer et al., 2020; Metcalfe et al., 2016; Shokri et al., 2024a, 2024b). While foundational research has offered substantial understanding of how ectotherm populations respond ecologically to variations in temperature e.g. (Angilletta, 2009; Clarke and Fraser, 2004; Schulte, 2015), few studies have examined the impacts of realistic IPCC climate change projections across seasonal temperature ranges on individual metabolic rates (but see (Bestion et al., 2015)).

This study aimed to expand this body of knowledge by empirically evaluating the responses of standard metabolic rate (SMR) i.e. the maintenance metabolism, in aquatic amphipods to current seasonal temperature variations and to temperatures above these as projected under the most conservative climate change scenario, RCP2.6 (IPCC: Pachauri et al., 2014). As global warming is expected to result in warmer temperatures across all seasons, we sought to understand how these projected increases would affect SMR on a seasonal basis. We measured the SMR of the subject species, *Gammarus insensibilis*, over a temperature range that matches the seasonal variations locally experienced by this species, from winter to summer. Additionally, for each seasonal baseline temperature, we evaluated the response of SMR to narrow incremental increases of 0.6 °C and 1.2 °C to determine the impact of temperature increases projected for 2040 and 2100 under the RCP2.6 IPCC scenario (IPCC, 2014).

2. Materials and methods

2.1. The study organism

Gammarus insensibilis (Stock, 1966), an ectothermic crustacean amphipod, inhabits the coastal waters of the Atlantic-Mediterranean region (Costello et al., 2001). They play a crucial role in the trophic webs of aquatic ecosystems, feeding mainly on leaf litter and detritus, and providing resources for secondary consumers (Nelson, 2011). After hatching, juveniles of *Gammarus* sp. are released from the marsupium with a body length of approximately 1 mm, mature at ~4 mm, can grow to a maximum length of about 20 mm (Longo and Mancinelli, 2014), and have a lifespan of up to one year (Gerhardt et al., 2011; Węśławski et al., 2020). Their distribution reaches as far north as 53.9° N and extends southward to 37.4° N, being their southernmost latitudinal limit (Tillin and White, 2017).

2.2. Experimental design

The experiment was designed to investigate the response of the standard metabolic rate (SMR) in specimens of the tested species to both current annual climate variations and projected temperature increases. Temperatures were selected to match both the seasonal temperature range currently experienced by the population in its local climate and the predicted increase in temperature due to climate warming. To match the current climate experienced by the tested population, the assessment temperature levels were the winter temperature, which was 12 °C, determined by calculating the average of the coldest temperatures recorded from 2015 to 2019; the autumn temperature set at 18 °C, also closely approximated the average annual temperature in this water body, and the maximum summer temperature, which was 29 °C, determined as the mean warmest temperature from 2015 to 2019. The water temperature data for the collection site were obtained from the Copernicus Marine Environment Monitoring Service (CMEMS: Buongiorno Nardelli et al., 2013). The projected temperature levels were selected based on the most conservative climate change emission scenarios i.e. RCP 2.6 (IPCC, 2014), which forecasts an average global increase in water temperature of 0.6 °C by 2040 and 1.2 °C by 2100 (IPCC, 2014; Genner et al., 2017). Accordingly, we extended the experimental temperature gradient by introducing two increments of 0.6 °C and 1.2 °C above the mean seasonal temperatures, to estimate metabolic responses to a relatively minor expected increase in temperature beyond what the animals typically encounter during each respective season.

2.3. Collection and acclimation of specimens

The specimens of varying body sizes were collected in autumn from a transitional water ecosystem along the southwestern coast of the Adriatic Sea (40.444 N, 18.238 E). After collection, the specimens were moved to the laboratory in thermally insulated containers that were filled with water from the collection sites and aerated throughout transport. The specimens were kept in laboratory aquaria with a salinity level that matched that of their natural habitats i.e. 21 g L⁻¹. Thermal acclimation to both above and below the collection temperature was gradually achieved, ±0.6 °C per day, in aquaria located within climate-controlled environments (KW apparecchi scientifici). The specimens were acclimated to the assessment temperatures for two weeks, a period considered sufficient to mitigate any risk of thermal stress that could potentially alter the metabolic rates of the specimens (Semsar-kazerouni and Verberk, 2018). The temperature treatments were evenly spaced throughout the experimental period.

Throughout the acclimation phase in the aquaria, specimens were provided with conditioned and decomposing leaves of *Phragmites australis*, ad libitum. Conditioned *P. australis* provides their primary resource, which is the colonizing microfungi (Able and Hagan, 2000). The experiment was conducted exclusively on males, as egg

development in females might lead to non-size-related variations in energy demands (Shokri et al., 2021).

2.4. SMR measurements

Each temperature treatment involved 24 individuals, totaling 212 males measured after excluding four due to moulting during the experimental trial. On each experimental day (09:00–17:00), we measured 24 individuals sequentially, completing all measurements for one temperature treatment whose acclimation period had been completed before moving on to the next. The Standard Metabolic Rate (SMR, J day⁻¹) was measured individually as oxygen consumption (VO₂) using open-flow system respirometry (Glazier and Sparks, 1997; Shokri et al., 2019, 2022; Vignes et al., 2012).

The animals were individually placed in plastic beakers and were not fed for 24 h prior to the SMR experimental trial at the assessment temperatures. To measure SMR, animals were individually placed in chambers, organized into two racks, each consisting of 12 chambers (Fig. S1). Each rack was associated with a 1-litre glass water tank filled with water having characteristics similar to those set in the acclimation aquaria. The water in the glass tanks was stirred magnetically at 200 rpm and supplemented with air bubbling, which together maintained the water oxygen-saturated throughout the experimental trial. A constant flow of water, at a rate of 6 ml h⁻¹, was maintained in the respirometer chambers (6 ml, 90 mm) by a peristaltic pump (Watson-Marlow 205U) for each rack, which was regulated to ensure that the oxygen levels in the chambers always remained above 80% saturation. Typically, like most benthic invertebrates, *Gammarus* sp. settled calmly at the bottom of the vertically positioned chamber. In each respirometer chamber, a 0.3 mm nylon mesh was installed to further restrict the spontaneous movement of the individuals. The nylon mesh was positioned a few centimetres down from the open end of the chamber, after it was filled with water, before introducing the animal. A 3-h equilibration period was set as the time needed to achieve a steady concentration of dissolved oxygen and to allow the specimen to settle in the chamber before readings began. As the water flowed out from the chambers, it was channelled to the Clark-type microelectrodes (SI1302 Strathkelvin oxygen electrodes), where an oximeter continuously read the oxygen concentration, with the data being recorded by the Strathkelvin software. After the 3-h equilibration period, the partial pressure of dissolved oxygen (Δ torr) was measured for each individual through two phases: first, a 15-min recording of the oxygen concentration curve with a specimen present (ppin), followed by a 15-min period for the blank measurement without any specimens (ppout). The blank ppout was obtained by disconnecting the input tube from the chamber containing the specimen and connecting it directly to the microelectrode. This procedure (measuring the ppin and ppout) was conducted uniquely for each specimen, ensuring that both ppin and ppout measurements were specific to each specimen. In our experimental setup, a total of six microelectrodes were available. Measurements were conducted in rounds; each round involved connecting six microelectrodes to six chambers. After completing measurements in one set of six chambers, the microelectrodes were connected to the next set for subsequent readings. This cycling was repeated to systematically assess all chambers. The Oxygen microelectrodes were calibrated using aerated water before each experimental trial. After each experimental trial, parts of the respirometer system were cleaned using bleach and sterilised using an autoclave (Hiclave HV) to prevent microbial growth. Furthermore, to ensure minimal background respiration and consistency of the measured blanks (ppout), we intermittently measured the dissolved oxygen partial pressure in a chamber that did not contain any specimens, using one of the six electrodes and found no detectable background respiration.

The oxygen consumption for each individual, expressed in (μ mol O₂ h⁻¹), was calculated based on the difference in the partial pressure of dissolved oxygen between the outflow water of the blank and the partial pressure of dissolved oxygen in the respirometer chamber containing a

specimen (torr). This difference was then multiplied by flow rate (l h⁻¹) and adjusted by multiplying it with the dissolved oxygen solubility coefficient (μ mol l⁻¹ torr⁻¹) for each temperature. The oxygen consumption rate was transformed into metabolic rate (J day⁻¹) by applying an oxyjoule conversion factor (Gnaiger, 1983), and multiplying the result by 24 h.

The body length of the tested specimens, defined as the distance from the base of the first antenna to the base of the telson (Asochakov, 1994), was measured using image-based analysis with a Nikon SMZ1270 stereomicroscope. The specimens were then individually dried at 60 °C for three days and then weighed using a microbalance with an accuracy of ± 0.001 mg (Sartorius MC5). After weighing, the specimens were ashed at 450 °C for 6 h to determine their ash weight. The obtained ash weight was then subtracted from the dry weight to calculate the ash-free dry weight of each individual (M, mg AFDW). This allowed us to remove the inorganic tissue from the body of the specimen.

2.5. Statistical analyses

One-way ANOVA was used to analyse the differences in body mass (M, mg AFDW) across temperature levels. The scaling of individual standard metabolic rate (SMR, J day⁻¹) with body mass (M, mg AFDW) and temperature (T) was assessed via multiple linear regression. The response variable individual SMR and the explanatory variable M were log-transformed to fit the size-scaling relationship as a power law, and the temperature was inverse transformed (Brown et al., 2004):

$$\log(\text{SMR}) \sim \log(M) \times (-T_A) \quad (\text{Eq. 1})$$

$$T_A = \left(\frac{1}{k_B T_e} - \frac{1}{k_B T_0} \right) \quad (\text{Eq. 2})$$

T_A is a standardised inverse temperature, k_B is the Boltzman constant (8.618×10^{-5} eV/k), T_e is the assessment temperature, and T_0 sets the intercept at 286.15 K, corresponding to the lowest temperature level (i. e., 12 °C in this study). The R² partitioned of the explanatory variables, M and T, was then determined by the LMG metric (Lindeman et al., 1980).

Furthermore, we determined the temperature sensitivity (Q₁₀) of the SMR for each temperature against the baseline seasonal temperatures. The temperature coefficient (Q₁₀) quantifies the fractional change in the SMR in response to temperature, commonly used to estimate variations in an organism's metabolic rate due to temperature changes (Arrhenius, 1889). The temperature sensitivity coefficient is calculated as:

$$\left(\frac{\text{SMR}_2}{\text{SMR}_1} \right)^{\frac{10}{T_2 - T_1}} \quad (\text{Eq. 3})$$

Where SMR₁ is the standard metabolic rate at T₁ (°C), SMR₂ is the standard metabolic rate at a higher temperature T₂ (°C). To account for size dependence, we quantified the temperature sensitivity of the SMR following Eq. (3) across individuals of varying sizes. We categorised the individuals into two classes based on their body mass, using distribution quantiles of 0.5 and 1 for each temperature. These classes comprise small-sized individuals (mean \pm s.d. = 1.03 \pm 0.66 mg AFDW) and large-sized ones (mean \pm s.d. = 4.25 \pm 1.44 mg AFDW). The analyses were performed in R free software environment (R Core Team, 2024), using dplyr (Wickham et al., 2023) packages.

3. Results

Overall, we analysed 212 male individuals of *G. insensibilis* ranging between 4.12 and 20.01 mm in body length (10.77 [± 3.17 SD] on average) and from 0.20 to 7.74 mg AFDW in body mass (2.59 [± 1.95 SD] on average). The distribution of body mass (M, mg AFDW) did not significantly differ across the nine-temperature levels tested (ANOVA;

$F_{8, 203} = 0.51, p > 0.05$).

Individual SMR ranged from 0.2 to 7.72 J day⁻¹, averaging 2.71 [± 1.82 s.d.]. The individual SMR was significantly influenced by both body mass and temperature (Table 1). It scaled with body mass with a scaling exponent of 0.71 [0.64–0.76 95% CI], and with temperature with a scaling of 0.62 [0.59–0.67 95% CI] (Table 1, Fig. 1). Overall, 79.6% of the variance in individual SMR was explained by the considered sources of variation and their interactions (Table 1, Fig. 1). 39.1% of the variance was related to individual body mass ($p < 0.001$), and 38.5% was due to the influence of temperature ($p < 0.001$) (Table 1, Fig. 1). The remaining 1.9% of the variance was explained by the negative body mass-temperature interaction ($p < 0.001$) (Table 1, Fig. 1). This negative body mass-temperature interaction indicates that the relationship between temperature and SMR is dependent on body mass: the SMR of small-sized individuals continued to increase with temperature across the full temperature range, whereas the SMR of large individuals did not at high temperatures (Table 1, Fig. 1).

3.1. Thermal sensitivity of the SMR across sizes in current seasonal and forecast climates

From winter to autumn, we observed an increase in SMR, with a Q10 value of 2.39 (an 11.51% °C⁻¹ increase in SMR) in small-sized individuals and a Q10 of 2.34 (an 11.11% °C⁻¹ increase in SMR) in large-sized individuals (Fig. 2). When transitioning from autumn to summer temperatures, the SMR of small-sized individuals continued to increase, with a Q10 of 2.25 (a 13.1% °C⁻¹ increase in SMR) (Fig. 2). In larger individuals, SMR continued to increase from autumn to summer with a Q10 value of 1.67, corresponding to a 6.9% °C⁻¹ increase in SMR, although this was at a slower rate compared to the winter to autumn period (Fig. 2).

With a temperature increase of 0.6 °C from the current winter baseline of 12 °C, the Q10 value was 1.9 for small-sized individuals (a 3.95% increase in SMR) and 2.5 for large-sized individuals (a 5.64% increase in SMR) (Fig. 2). An increase of 1.2 °C relative to the average winter baseline resulted in a Q10 of 2.1 (a 9.51% increase in SMR) for small-sized and a Q10 of 3.3 (a 15.45% increase in SMR) for large-sized individuals (Fig. 2).

When the autumn temperature of 18 °C was increased by 0.6 °C, the observed Q10 was 2.75 (a 6.26% increase in SMR) for small-sized individuals and 3.83 (a 8.39% increase in SMR) for large-sized individuals (Fig. 2). An increase of 1.2 °C from the autumn baseline led to a Q10 of 3.31 (a 15.48% increase in SMR) for small-sized individuals and 3.95 (a 17.92% increase in SMR) for large-sized ones (Fig. 2).

Above the summer temperature baseline of 29 °C, the Q10 for small-sized individuals reached 4.23, corresponding to a 9.04% increase in SMR (Fig. 2). With an additional increase of 1.2 °C, the Q10 for these small-sized individuals continued to rise, with a Q10 of 3.25 (a 15.22% increase in SMR). In contrast, for large-sized individuals, the Q10 values decreased with temperatures above the summer baseline, falling to less than 1 (Fig. 2).

Table 1

Output of linear mixed model with standard metabolic rate (SMR) as the response variable and body mass (M) and temperature (T) as the predictors.

Predictors	log (SMR)		
	Estimates	CI	P
(Intercept)	-0.29	-0.36–0.23	<0.001
log (M)	0.71	0.64–0.76	<0.001
T	0.62	0.59–0.67	<0.001
log (M) × T	-0.12	-0.18–0.08	<0.001
Observations	212		
R ² /R ² adjusted	0.796/0.791		

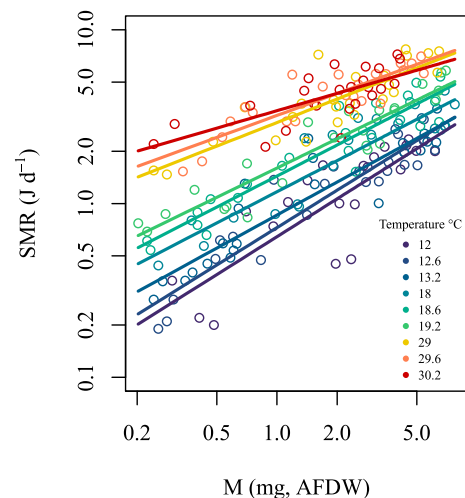


Fig. 1. Standard Metabolic Rate (SMR) in relation to body mass (M) across different temperature levels, log-log plotted.

4. Discussions

Metabolic rate, being one of the first organismal traits affected by global warming, triggers far-reaching consequences for higher order ecological processes (Bruno et al., 2015). In this era of climate change, despite its importance in forecasting ecosystem functioning, most predictions about metabolic-related processes rely on the expected thermal sensitivity of MTE. Here, we have empirically tested the metabolic rate in response to IPCC-RCP2.6 scenarios within seasonal temperatures.

We observed that individual SMR increased with body mass and temperature, which accords with general expectations of the metabolic theory of ecology (Brown et al., 2004). However, our results further showed that the scaling of SMR with body mass tends to decrease across the full range of temperatures, as indicated by the negative size-temperature interaction. This implies that larger individuals have a lesser sensitivity of their SMR to temperature than smaller ones. The observed decrease in the mass scaling exponents of SMR with temperature aligns with the Metabolic-Level Boundaries hypothesis (sensu Glazier (2020; 2005)) and with several recent experimental research e.g. Hoefnagel and Verberk (2015) and Shokri et al. (2022). Accordingly, volume-related tissue demand is expected to predominantly affect SMR at low metabolic levels, whereas surface-related resource supply and waste removal are likely to be the main influences on SMR at higher metabolic levels (Glazier, 2020). Cold temperatures in winter have also been suggested to increase the viscosity of the water and the boundary layer thickness surrounding the respiratory surfaces of ectotherms, potentially leading to lower metabolic rates (Verberk and Atkinson, 2013). This effect is expected to be more pronounced in smaller individuals, who are more susceptible to increased viscosity and face greater challenges in ventilating at low temperatures, compared to their larger counterparts (Verberk and Atkinson, 2013).

Furthermore, our results showed that the thermal sensitivity of SMR is season-dependent and varies with body mass. The highest thermal sensitivity in large-sized individuals was observed at temperatures reflecting global warming conditions, when autumn norms increase by 1.2 °C. However, at forecasted temperature increases above summer levels, the SMR of larger-sized individuals levelled off. In contrast, the thermal sensitivity of small-sized individuals continued to increase throughout the full temperature range, with the most notable increase observed at 0.6 °C above summer temperatures.

Larger individuals are likely to be already close to their upper thermal tolerance barrier at summer temperatures, where the capacity to further increase their metabolic rate is constrained (Carter et al., 2023; Schulte, 2015; Thyrring et al., 2020). Several empirical studies have

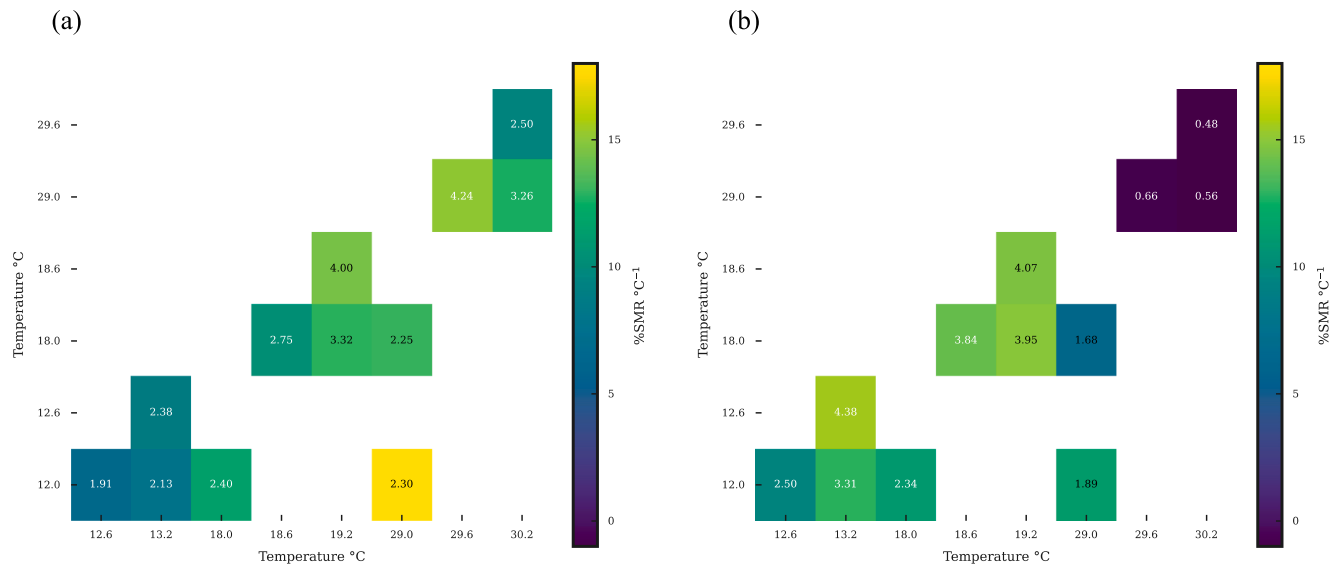


Fig. 2. Heatmap of temperature sensitivity coefficients (Q10) across various temperature ranges for (a) small-sized individuals and (b) large-sized ones. Each cell represents the Q10 value for each pair of temperatures. The color gradient indicates the percentage changes in standard metabolic rate (SMR) per degree Celsius. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

shown that aquatic ectotherms use compensation mechanisms in their metabolic rates when exposed to temperature variations that exceed their usual environmental conditions (Coggins et al., 2021; Precht et al., 1973; Shokri et al., 2022). This leads to a degree of cellular-level homeostasis in organisms, allowing them to sustain their performance despite temperature change (Precht et al., 1973). The compensation mechanism is likely to emerge when crucial metabolic substances could become limited if metabolic rates were to increase with temperature (Precht et al., 1973; Young, 1979). It is known that in aquatic ectotherms, oxygen availability is the constraining limiting factor for both metabolic rate and upper thermal limits (Precht et al., 1973; Verberk et al., 2016). This is because respiratory demands increase with temperature, but the availability of oxygen and the efficiency of its transport in water fall (Boardman and Terblanche, 2015; Verberk et al., 2016). As temperature rise, the oxygen supply may become insufficient to meet the increasing oxygen requirements of aquatic ectotherms, leading a point where the metabolic rate reaches a ceiling, subsequently reducing its thermal sensitivity (Rubalcaba et al., 2020). Among these, larger-sized conspecifics are expected to face more significant oxygen limitations with warming due to their higher absolute oxygen demands and their lower surface-area-to-volume ratio, which jointly narrow their aerobic scope (Atkinson et al., 2006; Lindmark et al., 2018; Verberk et al., 2022). This suggests that scenarios of global warming more extreme than those projected by conservative climate change forecasts, or the occurrence of heatwaves, could pose challenges to the survival of large-sized specimens, particularly during the summer.

To our knowledge, this is one of the first studies to empirically assess the effects of a modest predicted temperature increase of climate change on SMR within seasonal temperature ranges (see also Bestion et al. (2015)). Although empirical estimates of metabolic rates in response to projected global warming are crucial, further experimental studies should be extended over longer periods to reflect the adaptation of organisms in the face of climate change. As populations are exposed to increasing temperatures across generations, they may also genetically adapt, potentially leading to reduced metabolic rates (see Pettersen et al. (2024); Pilakouta et al. (2020)). It should also be noted that the findings of this study likely underestimate changes in SMR in the context of upcoming climate change, as the latest conservative scenario of the IPCC

has already projected a greater temperature increase of 1.5 °C by 2100 (IPCC: Lee et al., 2023).

In summary, our findings showed that the influence of temperature on SMR is size dependent, with larger conspecifics having a lesser sensitivity to temperature than smaller ones. We further demonstrated that smaller individuals possess a broader thermal window for SMR performance, while the SMR of larger individuals will become increasingly constrained as summer temperatures become warmer. Therefore, a small increment in temperature above seasonal norms, as predicted under climate change scenarios, impacts individuals' SMR. Consequently, even the least severe IPCC scenarios appear likely to affect higher-order ecological processes.

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CRediT authorship contribution statement

Milad Shokri: Writing – review & editing, Writing – original draft, Methodology, Investigation, Formal analysis, Conceptualization. **Ludovico Lezzi:** Writing – review & editing, Investigation. **Alberto Basset:** Writing – review & editing, Validation, Funding acquisition, Conceptualization.

Declaration of competing interest

Milad Shokri was a member of the editorial board of the J Thermal Biol at the time of submitting the manuscript but did not participate in the peer review or decision-making processes for this research article. The authors declare that they have no financial interests.

Data availability

Data are available at DOI 10.17605/OSF.IO/P66YA.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.jtherbio.2024.103941>.

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