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It's about time: Linkages between heat tolerance, thermal acclimation and metabolic rate at different temporal scales in the freshwater amphipod *Gammarus fossarum* Koch, 1836.

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Abstract:

Temperature has a profound impact on ectotherms. Warming increases the metabolic oxygen demand of ectotherms, which could result in a mismatch between their oxygen demand and their ability to extract and deliver sufficient oxygen to meet demand. This hypothesis has been mainly tested using short-term exposure to intense thermal stress. However, the thermal responses of organisms can be different on longer timescales, where physiological acclimation becomes increasingly important. Such thermal acclimation effects may reduce the vulnerability of ectotherms to warming on the long term. Thus, responses to intense, short-term thermal stress may be different from responses to moderate, prolonged thermal stress. Here, we examine the effect of thermal acclimation on heat tolerance and metabolism in the aquatic ectotherm *Gammarus fossarum* (Koch, 1836). Amphipods were acclimated to either $11.1 \pm 0.1^\circ\text{C}$ or $19.8 \pm 0.1^\circ\text{C}$ and after thermal acclimation we measured both their metabolism and their survival time at different temperatures. Our results show the metabolism strongly increased with increasing temperatures in the cold-acclimated group, but less so in the warm-acclimated group. Cold-acclimated amphipods were also more sensitive to thermal stress, especially during prolonged exposure. Thus, the differences between both thermal acclimation groups support the idea of oxygen-limited heat tolerance: cold-acclimated amphipods showed increased oxygen consumption and decreased thermal tolerance. However, across individuals, those that sharply increased oxygen consumption with increasing temperature did not differ in heat tolerance from individuals whose metabolism was much less sensitive to temperature. Thus, acclimation to different temperatures appeared to be beneficial, but a role for oxygen limitation could not be demonstrated unambiguously. Beneficial effect of acclimation were much larger during prolonged exposure, with the acclimation response ratio (ARR) ranging from 0.03 to over

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