

INVITED OPINION

A Framework for Modelling Thermal Load Sensitivity Across Life

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ABSTRACT

Forecasts of vulnerability to climate warming require an integrative understanding of how species are exposed to, are damaged by, and recover from thermal stress in natural environments. The sensitivity of species to temperature depends on the frequency, duration, and magnitude of thermal stress. Thus, there is a generally recognized need to move beyond physiological metrics based solely on critical thermal limits and integrate them with natural heat exposure regimes. Here we propose the thermal load sensitivity (TLS) framework, which integrates biophysical principles for quantifying exposure with physiological principles of the dynamics of damage and repair processes in driving sublethal impacts on organisms. Building upon the established thermal death time (TDT) model, which integrates both the magnitude and duration of stress, the TLS framework attempts to disentangle the accumulation of damage and subsequent repair processes that alter responses to thermal stress. With the aid of case studies and reproducible simulation examples, we discuss how the TLS framework can be applied to enhance our understanding of the ecology and evolution of heat stress responses. These include assessing thermal sensitivity across diverse taxonomic groups, throughout ontogeny, and for modular organisms, as well as integrating additional stressors in combination with temperature. We identify critical research opportunities, knowledge gaps, and novel ways of integrating physiological measures of thermal sensitivity to improve understanding and predictions of thermal vulnerability at various scales across life.

1 | Introduction

Climate change is exposing species not just to gradual warming but also to increase the frequency and severity of extreme heat events that impose physiological stress on organisms. Thermal vulnerability to heat stress depends on two key processes—exposure and sensitivity (Huey et al. 2012; Williams et al. 2008). Exposure reflects the extent to which organisms experience a potentially stressful environmental change. It is the outcome of the interaction between environmental factors and characteristics of the organism that determines body temperature. Exposure also incorporates the organism's ability to select microenvironments. New developments in the field of biophysical ecology have largely resolved the conceptual and technical barriers to predicting exposure to heat stress, though uptake of these methods has been gradual (Briscoe et al. 2023; Buckley and Kingsolver 2021). Sensitivity describes the thermal responsiveness of an organism to temperature stress that leads to physiological damage or death (Clusella-Trullas et al. 2021; Jørgensen et al. 2022); it depends on life history and physiology (Buckley and Kingsolver 2021). These factors are not independent: sensitivity can be moderated by the dynamics of exposure (intensity and duration) and by the capacity of species' physiology to recover from thermal stress. We therefore need a general, quantitative framework to capture the physiological mechanisms of both damage and recovery if we are to effectively predict how increasingly erratic and extreme thermal regimes will impact the function, survival, and reproduction of organisms.

Approaches to assessing thermal sensitivity vary across taxonomic groups and research fields (Bennett et al. 2018; Geange et al. 2021). Assessments of static endpoints, such as critical thermal limits and the quantification of the cumulative impact of prolonged exposure to different (potentially stressful) temperature regimes, are common procedures (Klockmann et al. 2017). The large variation in body size and lifespan among organisms affects the feasibility of measuring thermal tolerance consistently; it is necessarily assessed on vastly different life stages (e.g., fruits, seeds, eggs, larvae, adults) and on different scales, from components of an individual (e.g., leaves, flowers) to whole individuals and populations (e.g., bacterial colonies, *Drosophila* populations, soil seed banks) (Klockmann et al. 2017; Wahid et al. 2007).

Effective assessment of the thermal vulnerability of populations thus requires an integrated knowledge of the mechanisms by which temperature-induced damage leads to functional incapacitation, reproductive failure, or death in individuals. In many cases, assessing lethal limits is not possible for logistical or ethical reasons (e.g., in vertebrates or rare and long-lived species) and may not even be desired, given that we should be interested in detecting vulnerability at ecologically relevant thresholds prior to thermal death. To overcome this, researchers apply a range of proxies, such as thermal limits of biological processes, changes to activity budgets, and assessment of damage and mortality during extreme climatic events in nature (Marchin, Esperon-Rodriguez, et al. 2022; Sinervo et al. 2010; Welbergen et al. 2008). There is a need for developing integrative probabilistic and mechanistic models to characterize physiological responses to temperature with predictions that can be empirically tested and validated.

Here we demonstrate the potential to combine physiological models of thermal sensitivity with general models of exposure dynamics to enhance our ability to understand and predict the effects of temperature on organisms. We use example cases to illustrate why considering repair together with damage is essential and to highlight potential uses for the framework across disparate taxonomic groups and life stages to generate useful and testable predictions in the face of rapid global change. We identify key targets for focused research, whereby taking a unified approach with standardized terminology should improve predictive capacity.

2 | Thermal Death Time (TDT) Models Explicitly Incorporate Duration of Heat Exposure

Critical thermal limits (e.g., CT_{max}) have been used widely as static point thresholds or endpoints to represent the temperature at which physiological processes cease to function (Bennett et al. 2018). In some cases, critical temperatures are explicitly lethal (Lutterschmidt and Hutchison 1997), but they can also range from the temperature at which an insect can no longer right itself or is knocked down (van Heerwaarden et al. 2016) to onsets of spasms in lizards (Taylor et al. 2021), loss of equilibrium in fish (Ern et al. 2023), or dysfunction of photosynthetic machinery in plants (Arnold et al. 2021). For some comparative research questions, there are benefits to using point estimates as they are relatively easy to obtain, which permits large comparisons of thermal tolerances among taxa (Bennett et al. 2021; Camacho et al. 2024; Sunday et al. 2011) or sites (Dewenter et al. 2024; Sunday et al. 2019). The use of different indices and limitations of point estimates, and endpoints, like CT_{max} , have been comprehensively reviewed and critiqued since at least the 1990s (Clusella-Trullas et al. 2021; Jørgensen et al. 2021; Jørgensen et al. 2019; Lutterschmidt and Hutchison 1997; Ørsted et al. 2022; Rezende et al. 2020; Rezende et al. 2014; Santos et al. 2011; Terblanche et al. 2011). The consistent opinion from these works is that derived point estimates—which are often collapsed into a mean lethal temperature for a population—can be dependent on methodological differences (e.g., in heating rate; Arnold et al. 2021; Payne et al. 2025). Consequently, variance from non-biological sources can be high and calls into question the validity of broad comparative studies that use vastly different methods without adjusting for these (discussed in Perez et al. 2021).

Finding a singular temperature threshold to define thermal limits inherently overlooks the interplay between the intensity and duration of temperature exposure that leads to compounding physiological dysfunction (Hochachka and Somero 2002; Jørgensen et al. 2021; Michaelsen et al. 2021; Rezende et al. 2020; Rezende et al. 2014). The need to explicitly capture the intensity and duration of exposure (also referred to as thermal dosage, cumulative heat sum, heat load, or heat dose), along with integrating such information with dynamic, realistic thermal environments, has all led to the rise of the TDT model in ecology.

The TDT is not a new concept—it was first explicitly introduced in the 1920s to ensure that bacteria were killed during the canning process of food (Ball 1923). Subsequently, it has been applied to ectothermic animals to estimate survival times under various

acclimation and exposure temperatures (Maynard Smith 1957; Mellanby 1954). Relating thermal tolerance with exposure time re-emerged as a contemporary tool in thermal ecology in the past two decades (Armstrong et al. 2009; Rezende et al. 2014; Santos et al. 2011). Essentially, the TDT became an extension of the typical thermal performance curve—which provides insight into the optimal temperature, upper and lower limits, and temperature breadth for performance (Angilletta 2006, 2009)—with the added dimension of exposure time to thermal stress (Rezende et al. 2014). The TDT model has since been applied to several insects to understand thermal impacts on fertility and survival (e.g., Ørsted et al. 2024; Youngblood et al. 2025). It has also been applied to plants to optimize weed management in agriculture, where thermal treatments were applied to soil to eradicate weed seeds (Dahlquist et al. 2007) and to determine the effects of thermal load on the function of photosystems (Cook et al. 2024).

The TDT explicitly models how both exposure time and exposure temperature affect lethal limits (e.g., LT_{50} —the lethal temperature limit when 50% mortality occurs), which captures such relationships as:

$$T = CT_{\max 1h} - z \cdot \log_{10}(t) \quad (1)$$

where, T =temperature for, say, 50% mortality (LT_{50}), $CT_{\max 1h}$ is the critical thermal maximum ($^{\circ}\text{C}$), z =thermal sensitivity and t =time (in hours) before reaching the 50% damage threshold. Note that because $\log_{10}(1)=0$, the intercept of Equation 1, $CT_{\max 1h}$, corresponds to the lethal temperature for 1 h of exposure. While we standardise $CT_{\max}$ to 1 h, time can be scaled to other units (e.g., minutes) depending on what is biologically relevant to the organism's ecology. Given that survival follows a typical dose–response curve, logarithmic transformation makes the relationship between lethal temperature and time approximately linear (Rezende et al. 2014).

Alternatively, we can flip the axes to account for the fact that temperature is the main factor manipulated in experiments, allowing one to re-parametrise the TDT curve as follows:

$$\log_{10}(t) = \alpha + \beta \cdot T \quad (2)$$

In the above equation, time to reach 50% mortality, t , is on the y-axis and temperature, T , on the x-axis. We can recover $CT_{\max 1h}$ and z by back-transformation using the new slope (β) and intercept (α) from this relationship as follows: $CT_{\max 1h} = -\frac{\alpha}{\beta}$ and $z = -\frac{1}{\beta}$. The parameterization of the TDT curve as in Equation 2 is useful because it allows one to capture how damage accumulates over time as follows (see Jørgensen et al. 2021; Ørsted et al. 2024):

$$\text{Accumulated damage} = \sum_{i=1}^{T_e > T_c} \frac{100 \cdot (t_{i+1} - t_i)}{10^{(\beta \cdot \max(T_i, T_{i+1}) + \alpha)}} \quad (3)$$

where the equation calculates the accumulated damage (as a %) from time, t_i to time t_{i+1} , using the parameters from the TDT curve (Equation 2). The accumulated damage function assumes overheating risk and injury occurrence when T_e (the exposure temperature) exceeds T_c (the assumed critical temperature above

which heat injury accumulates) (Ørsted et al. 2024). When the accumulated damage reaches 100%, the lethal limit (i.e., the defined threshold; LT_{50} in this example) has been reached.

2.1 | Potential for Extending the TDT Model to Explore Sublethal Effects

Generally, TDT models are sensitive to the chosen endpoint, are phenomenological in nature, are usually quantified at the whole-organism level, and they assume that survival declines exponentially with exposure duration. However, mortality may not occur immediately under moderately stressful temperatures, and there can be both direct and immediate effects on other fitness components (Buckley and Huey 2016). It is also possible that organisms can cope with moderately stressful temperatures for a relatively long time, where survival remains at 100%, before they suddenly succumb to the stress (e.g., Gómez-Gras et al. 2022). The thermal conditions that organisms are exposed to during their development and at crucial life stages prior to—or in conjunction with—heat stress can substantially alter fitness outcomes beyond simple mortality. Generating predictions from dose–response curves could allow for a range of different limit thresholds to be used. For example, sublethal measurements (e.g., critical fertility limits and functional inhibition thresholds) can be used in conjunction with and can extend the value of TDT models (Cook et al. 2024; Faber et al. 2024; Ørsted et al. 2024).

While predictions for mortality thresholds align well with empirical data in ramping assays, they may not predict the survival probability curve if temperatures fluctuate (Rezende et al. 2020). This is partly due to the unknown capacity for repair processes to offset damage or injury accumulation during reprieves from damaging temperatures (Huey and Kearney 2020; Jørgensen et al. 2021; Ørsted et al. 2022). Dynamic, probabilistic modelling approaches attempt to circumvent this problem, and they seem to predict mortality under fluctuating conditions quite well when the empirical survival curves obtained at constant temperatures are adequately described (Rezende et al. 2020). These approaches offer exciting potential and will require additional empirical study to validate the net effect of damage–repair processes on physiological function that determines survival probability and what other impacts these—and other natural, interacting processes—have on the fitness of individuals and populations. TDT does not provide much insight into the amelioration of thermal stress (although recent studies are exploring acclimation, e.g., Baeza Icaza et al. 2025; Wehrli et al. 2024; Youngblood et al. 2025), which is a function of damage, repair, and acclimation. For this reason and for linguistic accuracy as the framework is used for broader applications, we propose that TDT is referred to as thermal load sensitivity (TLS) when used as a general framework that is inclusive of non-lethal measures and examines damage, repair, and/or acclimation processes.

3 | Damage and Repair: The Physiological Cost of Extreme Temperatures

The TLS framework allows for modelling approaches to be integrated with, or used to predict, both lethal and sublethal

limits (i.e., not necessitating *death* as in TDT). It places specific emphasis on *disentangling* the processes of damage and repair through time in dynamic environmental conditions. Specifically, we make the distinction that damage accumulates *during* stress and may be increasingly apparent *following* stress, while repair occurs *during* as well as *between* stresses, and the relative magnitude of these processes determines the extent to which the organism recovers at a given time point (Buckley et al. 2025; Williams et al. 2016). The shift to a TLS perspective is important as we progress our understanding of the effects of thermal stress accumulation, variability, and extremes on vital physiological processes that in turn affect demographic and ecological processes. There is growing empirical evidence of the important role of recovery from physiological damage following thermal stress (Bai et al. 2019; Curtis et al. 2014; Malmendal et al. 2006).

Ørsted et al. (2022) reviewed the nature of damage processes in ectotherms that occur beyond the ‘permissive’ temperature range in which normal function is possible (i.e., the ‘stressful’ range). As homeostasis is disrupted under thermal stress, there is a balance of two antagonistic processes: damage (injury accumulation) and repair. It is assumed that these processes may occur simultaneously; they both depend on the severity and duration of the thermal stress and legacy or carryover effects of environmental conditions prior to and following thermal stress (Buckley et al. 2025; Ørsted et al. 2022). If damage accumulates from a given heat load, it will need to be partially or completely repaired to re-establish homeostasis, both during and after cessation of stressful conditions. The buildup of heat load over longer time periods will not only result in damage accumulation but will also limit the extent of repair (Ørsted et al. 2022). Life processes are governed by complex chemical transformations within and between cells mediated by protein and membrane integrity. Mechanisms of heat damage generally involve increasingly misfolded or unfolded proteins (Feder and Hofmann 1999; Wahid et al. 2007) and oxidative damage to DNA, lipids, and proteins that ultimately compromises cellular function (Georgieva and Vassileva 2023; Hasanuzzaman et al. 2013; Ritchie and Friesen 2022; Tuteja et al. 2001). All these phenomena are influenced by temperature through the laws of thermodynamics (Michaletz and Garen 2024).

The TDT model captures repair and damage occurring simultaneously in the stressful range (where damage outweighs repair) through z , but repair will be far more important and impactful outside of the stressful range. Repair mechanisms can be diverse, but many are thought to be conserved between plants and animals (Tuteja et al. 2001). They include the regulation of (heat) shock proteins and other chaperone proteins to refold or to degrade misfolded proteins (Liu and Howell 2016; Wahid et al. 2007). Repair pathways are known for excising damaged DNA resulting from bursts of oxidative stress (e.g., base excision repair; Tuteja et al. 2001) and replacement of oxidized fatty acids (e.g., Wagner and Chitnis 2023), but the details of repair are less well understood compared to factors contributing to damage. Repair rates are known to be temperature-dependent in flies (Bowler and Kashmeery 1979; Dingley and Maynard Smith 1968; Ørsted et al. 2022), bacteria (Iandolo and Ordal 1966; McKellar et al. 1997), and plants (Curtis et al. 2014).

Theoretical advances allow for simulations of the dynamics of physiological damage and repair depending on temperature (Michaletz and Garen 2024), which are needed to predict sensitivity and vulnerability to stress in nature (Ørsted et al. 2024). For example, Klanjscek et al. (2016) developed a damage and repair model for oxidative stress that could potentially be applied to heat stress. Jørgensen et al. (2021) developed a mathematical model for estimating accumulated injury from thermal stress using static and dynamic knockdown data in TDT models. Rezende et al. (2020) also showed that dynamic TDT models, which assume that individuals that survived the thermal stress can repair damage between bouts of heat stress (e.g., overnight), could estimate survival probability of drosophilids in the laboratory and field. Such studies are foundational to test and validate that there is a dynamic interplay between damage and repair processes through exposure to thermal stress that varies in frequency, duration, and intensity.

3.1 | Modelling the Dynamics of Damage and Repair Using the TLS Framework

Modeling damage and repair in natural ecosystems requires us to connect physiological sensitivity with realistic thermal exposure at sufficiently fine resolution. Biophysical models can now approximate microclimates at hourly resolution globally, which can be coupled with models of thermoregulatory behaviors to predict operational temperatures of organisms (Kearney et al. 2020; Kearney and Leigh 2024; Klinges et al. 2022; Meyer et al. 2023). This relatively new capacity to predict the temperatures to which organisms are exposed can be combined in the TLS framework to make more nuanced predictions of risk to thermal stress at fine scales or under scenarios with dynamic and extreme environmental conditions. The inclusion of damage and repair enables the cumulative impacts of thermal stress to be modeled under natural, fluctuating conditions, including stress and reprieve. The rate of repair and the decay in the rate of repair, resulting from temperature stress or reduced physiological condition, can both be explicitly incorporated into simulations using the TLS framework. Such feedback processes are expected to alter organism function and homeostasis during exposure to heat stress and benign temperatures that facilitate repair (e.g., overnight or during periods of reprieve from heat). Thus, a key strength of the TLS framework is to explicitly include and evaluate these dynamic and interconnected processes that are often neglected in predictions of thermal tolerance or vulnerability.

To illustrate how the feedback processes of damage and repair could play out theoretically, we simulated the effects of temperature on physiological function while altering repair rates and their dependence on physiological function (additional details in [Supporting Information](#)). We estimated the thermal sensitivity of a hypothetical ectotherm (Figure 1a) and then simulated damage rate increasing rapidly with temperature (Figure 1b). We applied a Sharpe-Schoolfield Arrhenius model to simulate repair rates based on a repair rate coefficient (k) to set the rate of repair at 20°C (Figure 1c).

It is essential to recognize that damage and repair have non-linear relationships with temperature and that both processes

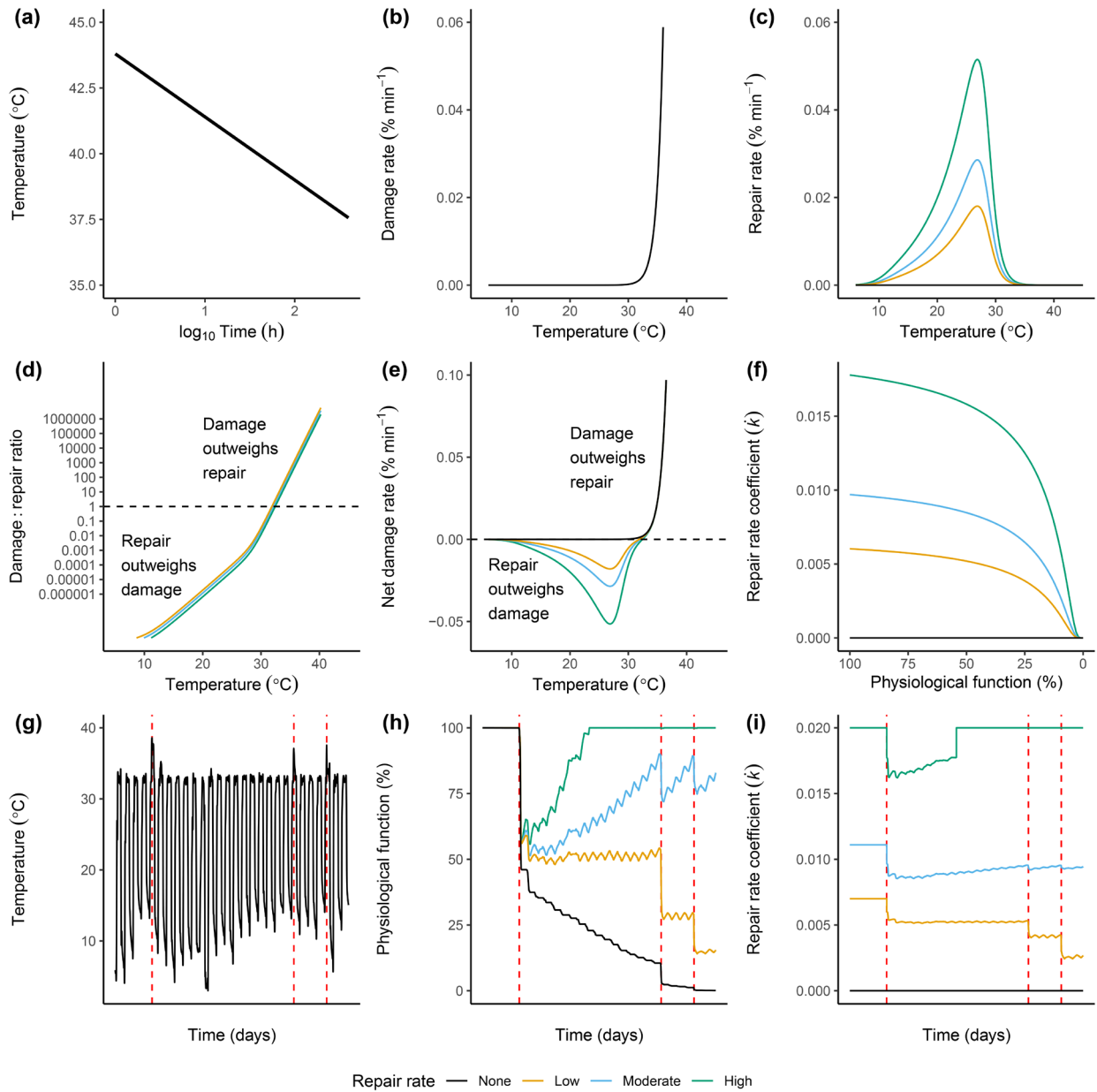


FIGURE 1 | Simulations of the counteracting processes of damage and repair during heat exposure of a hypothetical ectotherm. (a) The underlying thermal sensitivity curve for the ectotherm with intercept $CT_{max/1h}$ (critical thermal maximum of 1 h of exposure) and slope z (thermal sensitivity) parameters. (b) Damage rate simulated as a function of temperature using Equation 3. (c) Repair rates as a function of temperature, simulated for the hypothetical ectotherm with no (black), low (orange), moderate (blue), and high (green) repair capacity using Arrhenius functions. (d) The damage/repair ratio as a function of temperature, where the dashed black line represents a 1:1 damage/repair ratio. (e) The net damage rate as a function of temperature (the balance between damage and repair processes), where the dashed black line represents equal damage and ratio. (f) The repair rate coefficient ($\hat{k}$), which is the rate of repair at 20°C, as a function of the organism's physiological function. (g) The modeled body temperature during summer over a four-week time course. Dashed red lines in panels (g–i) represent extreme heat days during the time course. (h) Physiological function (%), the proportion of full performance possible following exposure to physiological stress that accumulates over the time course, simulated with different repair rates using the TLS framework, illustrating how this response may substantially impact the outcome of thermal stress events over time. (i) The dependence of the repair rate coefficient ($\hat{k}$) on physiological function over the time course.

will occur simultaneously. Outside the stressful range of temperatures, repair outstrips damage, whereas inside the stressful range, damage outstrips repair. TDT focuses mainly on the balance within the stressful zone but ignores repair outside the stressful range, within the permissive range. Although damage may be the net result of exposure to high temperature, repair processes, such as protein synthesis and chaperoning to limit

protein misfolding, are occurring whenever temperatures permit (Santra et al. 2019). Therefore, we calculated the damage/repair ratio (Figure 1d) and the net damage rate (Figure 1e), based on the balance between damage and repair at different temperatures, to predict the range of temperatures across which damage outweighs repair and *vice versa*. The processes that facilitate repair are likely also dependent on physiological condition, such

that the repair rate itself declines when an organism is in poor physiological condition from accumulating thermal damage (Figure 1f).

We applied this model to gridded hourly estimates of air temperature from the *microclimOZ* dataset (Kearney 2019) to predict body temperatures of our hypothetical ectotherm for 4 weeks, including 3 days that reach damaging extreme temperatures (Figure 1g). Predicted body temperatures were assumed to equal shaded air temperature, as in a small insect (note that heat budgets can be computed with the ectotherm model of *NicheMapR* (Kearney and Porter 2020) for more complex scenarios where this simplifying assumption would not hold).

Next, we integrated repair rate into probabilistic dynamic thermal ‘tolerance landscape’ models (Rezende et al. 2020). Note that the actual magnitude of the thermal stress is contingent on the temperature trajectories throughout the day. Thus, we simulate how the cumulative dosage of sublethal heat stress compromises physiological function, which is altered by (and further alters) the balance between damage and repair during the thermal regime (Figure 1h). Finally, we visualized the assumed dependence of repair rate on physiological condition as a feedback process that reduces the repair rate coefficient (k) when damage accumulates from exposure to heat (Figure 1i; details in [Supporting Information](#)). Box 1 provides an example application of the TLS framework incorporating damage and repair feedback for *Drosophila suzukii*, and an additional example for weed seeds is provided in the [Supporting Information](#).

While these process-based simulations of TLS are useful for generating plausible predictions about the balance between damage and repair of physiological function for a broad range of organisms, heat exposure scenarios, and different scales, they need further empirical characterization and validation. Both the general shape of the recovery curve as a function of temperature and the dependence of recovery on physiological function or temperature are, to our knowledge, still largely unknown (although the Arrhenius function in Box 1 appears to capture this well for *D. suzukii*). Various mathematical functions could be used to model the assumed temperature dependence of damage and repair, much like the suite of plausible functions that can be fit to thermal performance curves (Padfield et al. 2021); the most appropriate function will likely differ among life forms (Ørsted et al. 2022). It will therefore be necessary to design experiments to quantify damage and repair rates to parameterize and to validate these models, which remains challenging for real organisms (Bai et al. 2019; Huey and Kearney 2020; Kingsolver and Woods 2016; Klanjscek et al. 2016). Broad taxonomic groups might have similar sensitivity responses due to evolutionary conserved mechanisms of cellular damage and repair, but this is yet to be tested. We recognize that varying these damage and repair assumptions could significantly alter model outcomes (e.g., Youngblood et al. 2025), and this is an exciting area for investigation for which we advocate targeted investigations into damage-repair processes across diverse taxa.

4 | TLS Could Help Address Key Outstanding Questions in Global Change Biology and Thermal Ecology

Global change biology and thermal ecology inherently need to consider multiple stressors in combination and the impacts of the timing and magnitude of these stressors in an organisms’ life. Below we provide an exploratory, conceptual overview of some of the emerging areas of research for which the TLS framework could be used for both theoretical and empirical insight.

4.1 | Sublethal Measures of Thermal Sensitivity and Impacts on Modular Systems of an Organism

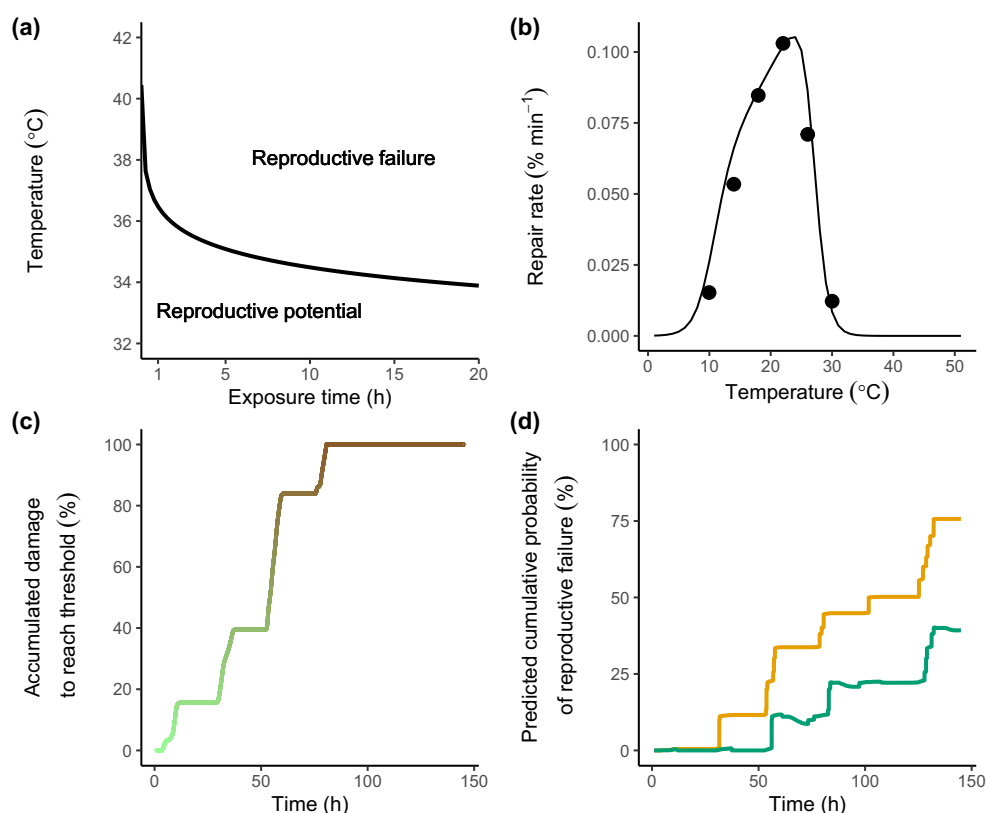
The role of heat exposure in causing sublethal detrimental effects on organism fertility has come into sharp focus as an important climate change impact on population growth, extinction risk, and species distributions (Bretman et al. 2024; van Heerwaarden and Sgrò 2021; Walsh et al. 2019). The TLS framework allows investigations into potential spatial distributions based on thermal effects on sublethal traits (Box 2). Thermal sensitivity to heat stress in animals usually focuses on whole-organism physiology and ignores more vulnerable modular organs and life stages (Bennett et al. 2018). The thermal sensitivity of essential organs and primary biological functions like reproduction is arguably more ecologically valuable to understanding the potential vulnerability of organisms to global change stressors than are their lethal endpoints (van Heerwaarden and Sgrò 2021).

In plants, most of the thermal vulnerability indices are calculated for leaves or cut leaf sections and thus describe thermal limits at the functional level at a very fine scale (e.g., photosynthetic machinery). The temperature ranges realized in most plant species’ geographic range are far narrower than measured thermal limits (Lancaster and Humphreys 2020), and there is little evidence that extreme temperatures alone kill adult plants, especially trees (Marchin, Backes, et al. 2022). Both the onset of functional impairment of photosystems and the damage to leaf tissue are clearly dependent on thermal exposure time (Cook et al. 2024; Faber et al. 2024; Neuner and Buchner 2023). However, we know little about how accumulated thermal damage to modular organs like leaves then affects the state of larger components such as a tree crown or the entire tree, and what the resource or energy costs are for repair or discarding dead tissue and regenerating. To illustrate these concepts, we used data from a heatwave during the dry summer of 2020 in Sydney, Australia. Daily maximum air temperature exceeded 45°C on multiple occasions during a period of no rainfall, within which it is too dry to repair the damage from heat stress (orange area of Figure 2a), resulting in crown dieback (Figure 2b). Although there were then small rainfall events, extreme temperatures were still occurring, and these conditions remain unfavorable for substantial repair (blue area of Figure 2a), but crown cover loss was less dramatic (Figure 2b). Larger rainfall events coupled with a reduction in maximum air temperature then provided conditions

BOX 1 | Application of the TLS framework to *Drosophila suzukii*.

Drosophila suzukii is a globally invasive pest that is a prime candidate species for studies of TLS. We used raw data for productivity of female flies from Ørsted et al. (2024) to explore damage accumulation and repair under combinations of temperature and exposure duration. Productivity of females is a crucial (sublethal) contributor to population viability that is more sensitive to temperature than thermal coma or death.

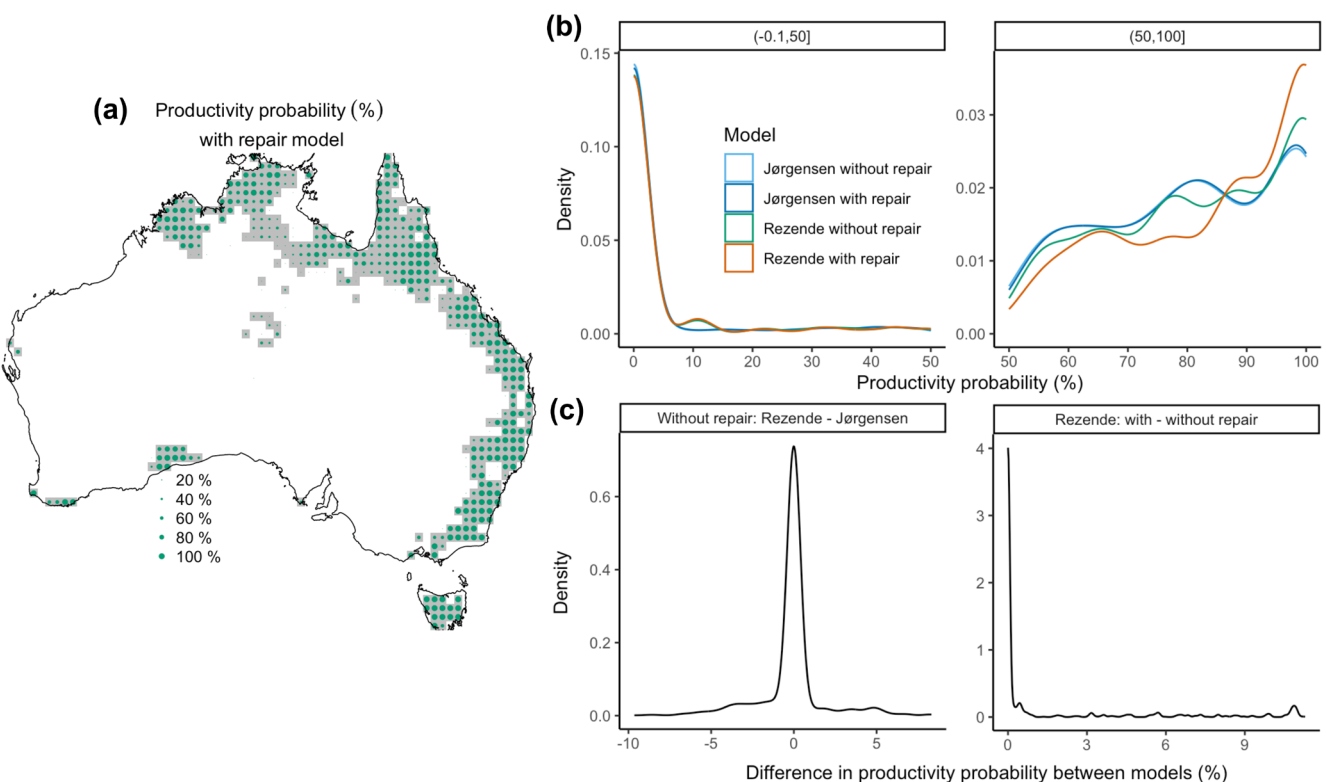
Using these data, we show how the relationship between temperature and exposure duration determines the conditions under which reproduction can potentially occur or fail (Box 1 Figure a). To illustrate the potential for repair to alter heat failure rates and outcomes, we used *metaDigitise* (Pick et al. 2019) in the R Environment for Statistical Computing v4.3.1 (R Core Team 2023) to digitize Figure 5c from Ørsted et al. (2022), extract preliminary repair values (%) at six 'repair temperatures' for *D. suzukii*, and convert them to repair rate per minute (% min⁻¹). These repair values correspond to the improvement of knockdown time relative to a first heat exposure after 6 h of recovery at different temperatures to allow for repair before another knockdown assay. We recognize that these data are preliminary and correspond to knockdown rather than reproductive viability (Ørsted et al. 2022), but there is little empirical data on temperature-dependent repair rates available. We developed a simple model to simulate repair rates, where repair is modeled using the Sharpe-Schoolfield Arrhenius model (Schoolfield et al. 1981) that uses a repair rate coefficient (*k*) to set the rate of repair at 20°C (*de facto* optimum), such that instantaneous repair rates are high at optimal temperatures but drop rapidly at thermal extremes (equation and fitted parameters in Supporting Information). The six reported repair rate data points derived from Ørsted et al. (2022) correspond closely with the Arrhenius model for repair rate (Box 1 Figure b). Using a six-day simulation of realistic body temperatures (that ranged 6°C–34°C; Figure S1) derived from *NicheMapR* (Kearney and Porter 2020), we applied the damage accumulation function (Equation 3) to demonstrate the accumulation of damage up to the *T*₅₀ threshold (50% reproductive viability), which is reached after around 81 h (Box 1 Figure c). With no repair, a dynamic 'tolerance landscape' function (Rezende et al. 2014) shows that a 50% probability of reproductive failure is reached around 100 h. Accounting for repair reduces the probability of reproductive failure to below 50% for the entire simulation (Box 1, Figure d). Thus, these models using data for heat failure with and without repair provide markedly different fitness outcomes.



BOX 1 FIGURE. Conceptual and practical application of the thermal load sensitivity (TLS) framework to female *Drosophila suzukii* reproduction. (a) Regression between temperature (y-axis) and time (h) to event (in this case *T*₅₀, x-axis) data is then used to estimate the *CT*_{max1h} (intercept of curve) and thermal sensitivity *z* (slope of the log₁₀-linear relationship). (b) Repair rates as a function of temperature. Points are estimates for *D. suzukii* repair rate from Ørsted et al. (2022), and the curve is modelled repair rates using an Arrhenius function. (c) Simulating temperature exposure across six days with cool nights and applying the accumulated damage model (Equation 3) to illustrate how damage accumulates up to reach the threshold *T*₅₀. (d) Predicted cumulative probability of reproductive failure using dynamic tolerance landscape models without repair (orange) and with repair (green) that is occurring both during stress and also outside of the stressful range of temperatures.

BOX 2 | Estimating the potential spatial distribution of the invasive pest *Drosophila suzukii* as a function of damage accumulation and repair capacity.

Drosophila suzukii is a globally invasive pest that would have devastating consequences for agricultural industries if it were to establish itself in Australia. Current pest risk analysis reports indicate it would have major impacts on berry, stone fruit, and viticulture, collectively worth at least \$5.4 billion AUD (DAFF 2013). To identify regions where *D. suzukii* could maintain productivity (positive population growth), we extend the example from Box 1 to estimate the spatial extent in which female *D. suzukii* could remain productive for seven days in January in Australia (summer) using gridded microclimate data from *microclimOZ* (Kearney 2019). First, we fitted a traditional static 50% threshold ($CT_{max1h} = 36.3^{\circ}\text{C}$) model to determine the spatial extent within which *D. suzukii* could remain productive (grey background area in Box 2 Figure a; Figure S2). Then, we fitted dynamic thermal landscape models from Rezende et al. (2020) and dynamic CT_{max} models from Jørgensen et al. (2021), each with and without implementing the damage-repair feedback (details in Supporting Information), applied to each grid cell. The size of the green circles in Box 2 Figure a indicates the probability of females producing offspring based on the dynamic tolerance landscapes model with repair (for maps of each model, see Figure S3). Box 2 Figure b shows the density (proportion of grid cells) of producing offspring according to the four models. This shows that the different models generally behave similarly, while including repair increases the proportion of locations with productivity above 85%. Box 2 Figure c left panel shows that there is relatively little difference between the Rezende and Jørgensen modeling approaches also shown by Youngblood et al. (2025), while the right panel shows that there is up to 12% difference in productivity probability when repair is included. The damage accumulated over the seven-day simulation was reduced when we included damage-repair dynamics. Thus, applying the TLS damage-repair model provides a more detailed perspective on the intensity of sublethal heat stress, highlighting geographic areas where the persistence of *D. suzukii* may depend on repair processes. Such insights could be used to more effectively identify growing regions that might be susceptible to incursion and population establishment. Our model examples suggest that even during a hot week in summer, female *D. suzukii* could still reproduce in large portions of Australia's most productive agricultural regions. For example, the predicted distribution of the area where the fly could reach high productivity includes significant areas for growing strawberry in southeast Queensland, and grape and stone fruit growing regions in eastern New South Wales, eastern Victoria, and much of Tasmania.



BOX 2 FIGURE. (a) Spatial map for potential extent for *Drosophila suzukii* to remain productive during a hot week in summer in Australia. (b) Density plots of productivity probability across the grid cells and (c) of pairwise comparisons between different models with and without repair.

that allowed repair of damage (green area of Figure 2a), and then at least two species of urban trees had the capacity to regenerate their crowns, while others were too damaged (Figure 2b).

4.2 | Demographic Scaling Across Life Stages

The need for more ecologically relevant measures of temperature stress has given rise to the adoption of other less extreme

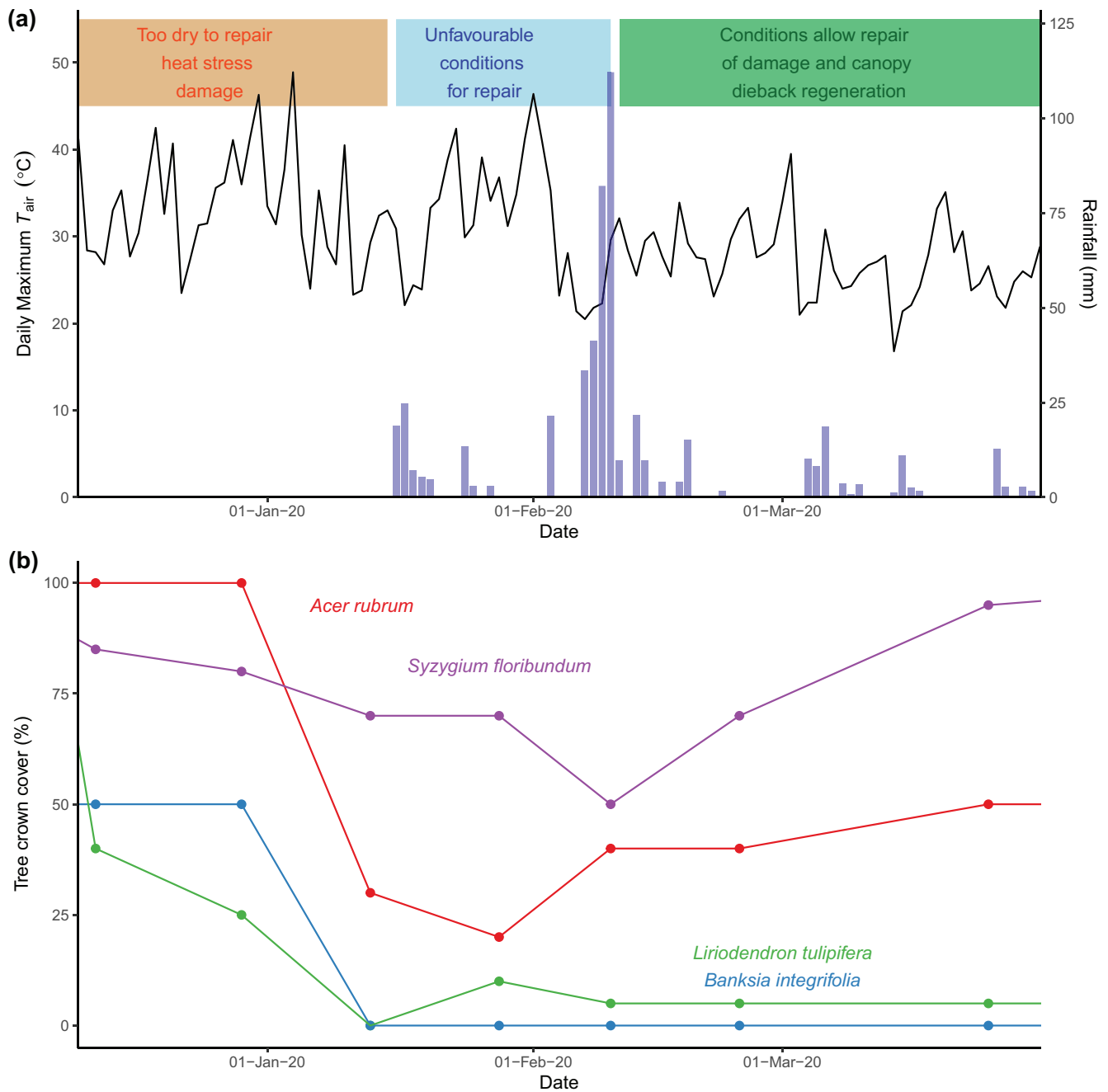


FIGURE 2 | Example of (mostly) sublethal effects of heat on modular components of organisms (e.g., leaves on trees). (a) Extreme heat during dry conditions in Sydney, Australia, during the 2019–2020 austral summer. The black line is the daily maximum air temperature, and the blue bars are rainfall events. (b) Recovery of tree crown foliage from heat stress in urban tree species during this time was conditional on heat tolerance and water availability. Responses were species-specific: Some trees died when maximum air temperature surpassed physiological thresholds (*Banksia integrifolia*, blue), while surviving trees began recovering by resprouting new leaves in the weeks after rainfall (*Acer rubrum*, red; *Syzygium floribundum*, purple). The young leaves of some species were vulnerable to further heat damage (*Liriodendron tulipifera*, green), and full recovery of lost foliage of trees that accumulated substantial heat damage took multiple years for many individuals (data adapted from Marchin, Esperon-Rodriguez, et al. (2022)).

(sublethal) indices of thermal vulnerability, like thermal fertility limits (Walsh et al. 2019). Different life stages clearly have different temperature stress thresholds, typically with pollen development and seedling stages being the most thermally sensitive in plants (Ladinig et al. 2015; Rosbakh et al. 2018; Tushabe and Rosbakh 2025) and sperm the most thermally sensitive in animals (Dahlke et al. 2020; van Heerwaarden and Sgrò 2021). Early life stages that are sessile can be more vulnerable to

overheating and may have lower heat tolerance (e.g., butterfly eggs (Klockmann et al. 2017), tadpoles (Ruthsatz et al. 2022), and intertidal gastropods (Truebano et al. 2018)). However, in other cases, less mobile instars and pupal stages of insects can be more tolerant than eggs or adults due to their reliance on inherent heat resistance rather than behavioral heat avoidance (Bowler and Terblanche 2008; Kingsolver et al. 2011). Small and large organisms (including the same species at different stages of

growth) can have size-dependent body temperature and thermal resistance due to thermal inertia and changes to boundary layer properties (Kearney et al. 2021). Organisms around the millimeter scale, including larval stages of invertebrates, may have very subtle and fine-scale microclimates available to them to avoid overheating (Pincebourde and Woods 2020). In plants, the life stage at which the plant is exposed to thermal stress is crucial in determining the impact of that stress on individual plant responses, their reproductive success, and subsequent population dynamics (Everingham et al. 2021; Notarnicola et al. 2021, 2023; Satyanti et al. 2021). However, most available thermal tolerance data are measured on adults, largely ignoring earlier life stages or actively reproducing individuals, both of which are crucial for assessing the true vulnerability of a population to environmental stress (Bennett et al. 2018).

Climate warming will expose different life stages to different intensities of heat events due to variation in microclimates, sessility, and thermoregulatory behavior (Levy et al. 2015). In reptiles with temperature-dependent sex determination, nesting habitats that are exposed to consistently warmer temperatures or fluctuating extreme heat events may no longer support balanced sex ratios necessary for population stability (Valenzuela

et al. 2019). Shifts in developmental rates and timing of reproduction could also dissociate species' trophic interactions or interspecific dependencies that make environments viable (Kronfeld-Schor et al. 2017). Ecologically relevant evaluations of thermal sensitivity and vulnerability across life stages are needed to effectively model impacts on population demographics. As an illustrative example, we simulated life-stage-specific sensitivity to thermal load in a hypothetical plant (Figure 3a,b) and applied a simple matrix population model (Figure 3c) to simulate demographic projections (Figure 3d,e). This approach (see also Salguero-Gómez et al. 2015) is a basis for allowing TLS to alter probabilities for transition within matrices (Figure 3b,c) if thermal stress occurs during a given life stage (see also Wiman et al. 2014). Further integrations of sublethal thermal effects on growth and reproduction informed by TLS could also be built into sophisticated trait-based demographic process models (e.g., Falster et al. 2016; Towers et al. 2024).

4.3 | Phenotypic Plasticity and Thermal Legacies

Prior exposure to stressors can result in plastic changes that make organisms (intragenerational) or their offspring (intergenerational)

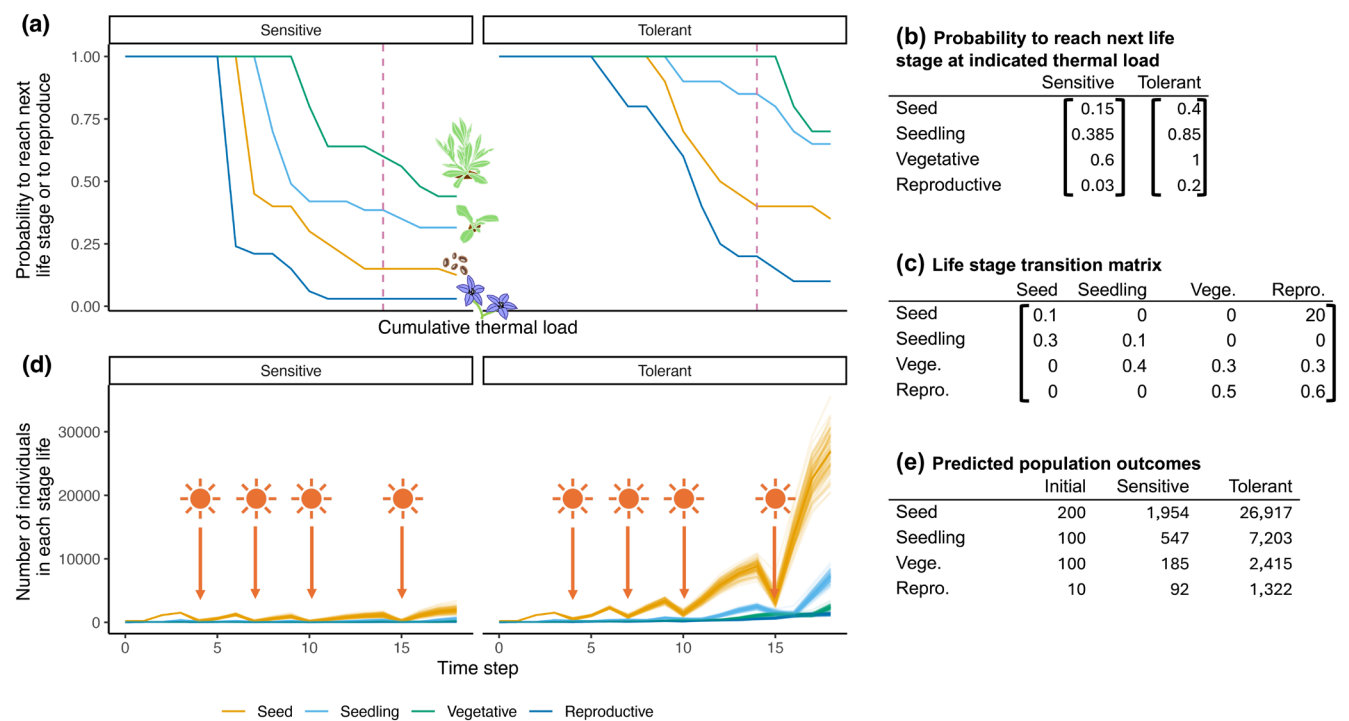


FIGURE 3 | Simulation of how thermal load sensitivity can differ across life stages in sensitive and tolerant populations of a hypothetical plant species with four distinct life stages: seed, seedling, vegetative (non-reproductive adult), and reproductive (actively flowering adult). (a) As cumulative thermal load increases toward prolonged high temperature, the probability of progression to later life stages and reproducing is reduced. The left panel shows probability declining with cumulative thermal load in a sensitive population, and the right panel shows the same for a tolerant population. (b) Vectors of probabilities for transition to the next life stage in the populations at the thermal load indicated by the dashed line. (c) Life stage transition matrix showing the proportion of each life stage transitioning to the next life stage or reproducing at each time step (e.g., 10% of seeds remain seeds, 30% become seedlings, which implies 60% fail to establish as seedlings, while 60% of reproductive plants remain in the reproductive stage, 30% stop flowering and return to the vegetative stage, 10% die, and each reproductive plant in the reproductive stage at the time step produces 20 viable seeds that return to the seedbank). (d) Predicted population dynamics through time as the number of individuals in each life stage from 100 simulations under a scenario where a heat event equivalent to the thermal load indicated in (a) occurs at four of the time steps (indicated by the sun symbol with arrows). (e) Initial population size at time step 0 and the final population of sensitive and tolerant populations at time step 20, showing the persistent effects of different sensitivity of life stage to cumulative thermal load that could have persistent or lag effects on population dynamics.

less sensitive to future stress events through acclimation or developmental or transgenerational plasticity. For example, acclimation through heat hardening is expected to mitigate damage through ‘resistance’ mechanisms that protect cells, such as upregulation of heat shock proteins (Moseley 1997). Early growth environments alter development of offspring (Monaghan 2007) through developmental plasticity—the ability for an organism to alter its phenotype in response to its environment during development (West-Eberhard 2003). Thus, exposure to heat stress early in life could lead to altered sensitivity to heat stress (i.e., thermal load) later in life via stress priming (e.g., Hoffman et al. 2018; Hossain et al. 2018). A comprehensive meta-analysis of ectotherms found that developmental temperatures often slightly increased heat tolerance but did not consistently result in persistent effects on later life stages (Pottier, Burke, et al. 2022). It is not always the case that developmental environments shift responses to temperature, and it is not yet clear if and how thermal sensitivity is altered by marginally stressful thermal histories. Thermal tolerance and plasticity can have complex patterns throughout ontogeny, further altered by the history of exposure to chronic or acute thermal stress. These ‘thermal legacy’ effects can alter threshold-based thermal tolerance and physiological plasticity (Geange et al. 2021; Lancaster and Humphreys 2020; Marasco et al. 2023; Payne et al. 2025) and will therefore likely also modify the rates and sensitivity of both damage and repair processes (Burton et al. 2022; Einum and Burton 2023).

4.4 | Multi-Stressor Integration

The TLS framework can be extended to understand the combined effects of multiple stressors, whether biotic (e.g.,

competition, disease) or abiotic (e.g., salinity, nutrients, water). Such an approach is feasible given that exposure to additional stressors may affect similar underlying physiological processes of damage and repair through cross-tolerance (Bryant et al. 2024; Hossain et al. 2018; Katam et al. 2020). In natural environments, a range of potential abiotic and biotic stressors frequently co-occur and interact with thermal stress, increasing the challenge of predicting cumulative effects of thermal stress. Thermoregulation in plants is complex and highly dynamic, with significant differences in realized temperatures and leaf-to-air offsets that depend on canopy structure and scale (Arnold et al. 2025; Dong et al. 2017; Guo et al. 2023); however, it is clear that water availability will moderate responses to high temperatures (Ruehr et al. 2016). For example, heatwaves often occur during droughts. Experiments have found that at moderate levels of water stress, plants may exhibit a priming response that increases heat tolerance but, at extreme levels, water stress greatly decreases the ability of plants to cool their leaves and so may exacerbate heat stress (Cook et al. 2021; Marchin, Backes, et al. 2022). Other biotic interactions, such as pathogen infection that occur simultaneously with heat stress, can not only suppress resilience to the pathogen but also reduce the heat tolerance of the host in invertebrates (Hector et al. 2021) and plants (Desaint et al. 2021).

Ultimately, when organisms are exposed to two or more stressors, the cumulative effect of all stressors can be additive, synergistic, or antagonistic (Orr et al. 2020) (Figure 4a,b), and disentangling multi-stressor effects on thermal tolerance should be a major focus of future work. Exposure to additional stressors will alter the TLS parameters, along with damage

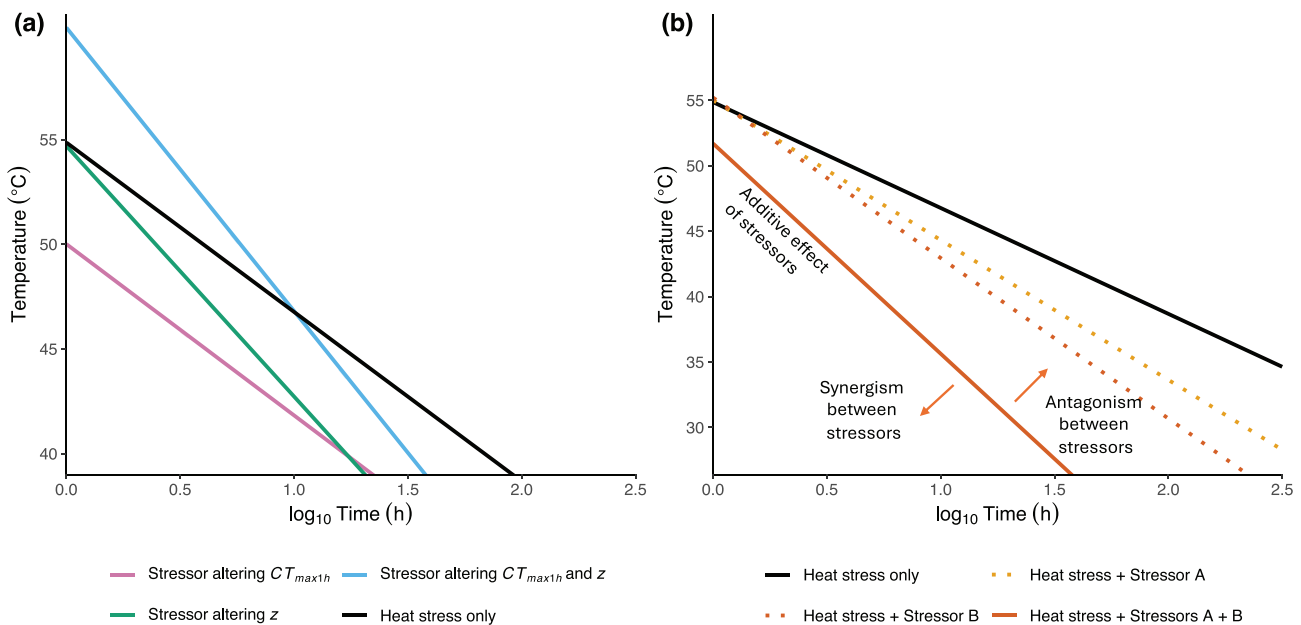


FIGURE 4 | Conceptual depiction of the effects of heat stress in combination with additional stressors within the TLS framework. (a) Different colored lines represented potential changes in the CT_{max1h} and/or z parameters of the TLS curves from heat stress only (black line) when subject to an additional stressor. (b) The difference between the TLS curves with heat stress alone (black solid line) and the TLS curves of heat stress with other stressors individually (A, yellow dotted line, and B, orange dotted line). From these lines we would predict that the effect of all three stressors (heat, A, and B) is additive by summing the difference between heat stress only and heat stress with one stressor (orange solid line). If the net effect of the three stressors is more extreme than the additive effect, then the stressors accumulate synergistically, but if the effect of all three is less than the additive effect, then the stressors are antagonistic, and the net effect is less than the sum of their individual effects.

and repair rates and thresholds for enzyme inactivity, potentially in complex or non-linear ways. As a simple (linear) example, a change in intercept (CT_{max1h}) with no change in slope (z) with the addition of non-thermal stressors implies an additive effect of the stressors (Figure 4b). Changes in slope, with or without changes in intercept, imply an interactive effect, either synergistic or antagonistic (Figure 4b), as the extra effect of the non-thermal stress can also be temperature-dependent (Duncan and Kefford 2021). For these simplified examples, multi-stressor effects on CT_{max1h} and z can be evaluated by including interaction terms in statistical models. The limited empirical data available with multiple abiotic stressors (e.g., Enriquez and Colinet 2017; Maynard Smith 1957; Verberk et al. 2023; Youngblood et al. 2025) suggest the slope can change, implying an interactive effect.

5 | Conclusions and Agenda

The TLS framework provides a step toward reconciling the ways in which organisms deal with natural dynamics of heat stress, whether that be temperature alone or in combination with other stressors. The TLS framework has strong foundations in biophysical and ecophysiological principles, while substantial flexibility for empirical study and theoretical modeling is achieved by integrating the dynamics of thermal stress exposure with the dynamics of physiological damage and repair. The following five key areas, as discussed above, stand out as being important foci for investigation, extension, and application of the TLS framework to better understand and predict thermally mediated impacts on organisms at various scales.

1. *Adopt TLS terminology because it is inclusive of sublethal effects and applies across developmental states and the tree of life.* As discussed above, we advocate a shift in language and inherent focus from lethal effects to sublethal effects that are more ecologically relevant, which includes taxonomic groups for which it is difficult or undesirable to estimate whole-organism death. Large datasets for diverse thermal tolerance limits are emerging, mostly for ectothermic animals (e.g., Bennett et al. 2018; Lancaster and Humphreys 2020; Pottier, Lin, et al. 2022). While these provide a foundation, there is a need to expand them to cover a more representative sample of life. Improving understanding of the biological processes that underpin a given sublethal effect and testing assumptions to better parameterize models will improve the efficacy of thermal vulnerability predictions for a given species.
2. *Apply emerging tools to identify universal damage and repair mechanisms that impact recovery from thermal stress.* Disentangling damage and repair mechanisms is crucial (Ørsted et al. 2022). Integrative computational models for genome-scale protein folding and stress responses are emerging for microbes (Chen et al. 2017; Zhao et al. 2024); however, empirical data and understanding of these dynamic biological processes remain very limited for complex life forms. Developing effective methods for measuring rates of damage and repair in plants and animals could be tackled with multifaceted flow cytometry approaches using consensus panel markers of stress, damage, and repair

(Buerger et al. 2023). By determining the conditions under which proteins unfold and inactivate and oxidative stress responses are expressed and by mapping programmed cell death pathways during and after thermal stress (Chen et al. 2020; Roychowdhury et al. 2023), we can begin to understand mechanisms of damage and repair reciprocity. Repair will be particularly important when damage is not excessive, and we therefore need to better understand the trade-offs between repairing or replacing damaged cells and tissues and how these depend on metabolic repair costs (Rennolds and Bely 2023). Linking bioenergetics at the cellular level to physiological and ecological functions and fitness is a crucial research frontier (Sokolova 2021). We need, however, empirical data to build a deeper understanding of the complex cellular processes underlying damage and repair to construct and evaluate mechanistic models.

3. *Ascertain principles determining how multiple stressors, both abiotic and biotic, affect TLS.* Different stressors and biological interactions are expected to impact the damage and repair processes by acting through common mechanisms across plants and animals (Wek et al. 2023). However, combinations of stressors and/or biotic interactions and their timing may have complex effects on damage accumulation that must be factored into assessments of vulnerability (Georgieva and Vassileva 2023; Prash and Sonnewald 2015; Taborsky et al. 2022). Few studies have evaluated how additional stressors modify TLS, and given that stresses co-occur in nature, this is an essential avenue for future investigations.
4. *Integrate plasticity in response to past stress to determine mechanisms and scale of stress priming.* A clearer understanding is required of the biological mechanisms and environmental cues that contribute to priming and the plasticity of responses to stress. Plasticity in damage and repair processes and the time course or rates of these plastic responses can alter sensitivity and lead to differences in vulnerability of populations (Burton et al. 2022; Dupont et al. 2024; Einum and Burton 2023). Thus, exploring the timing of stresses and rates of plastic responses will be pivotal to being able to model and predict how environmental exposure affects individuals throughout ontogeny and then scales up to affect the vulnerability of populations.
5. *Improving understanding of the plastic and evolutionary potential of thermal tolerance will inform conservation and management decision-making and breeding for food security.* Finally, we need a better understanding of genetic variation in stress tolerance across diverse taxa. The genetic variation underlying thermal sensitivity likely depends on multiple complex mechanisms acting over different time scales (González-Tokman et al. 2020; Logan and Cox 2020). Quantitative genetics can reveal evolutionary constraints, selection, and heritability of TLS parameters (Leiva et al. 2024). Understanding phenotypic and genetic variation in thermal sensitivity among populations is essential for predicting how they could adapt to future environmental conditions, which facilitates strategic conservation planning and adaptive management (Bennett et al. 2019; Rilov et al. 2019). Breeding crops that are resilient to

thermal extremes from climate change while maintaining yield to meet food security demands will rely on building a deep understanding of the adaptive signatures and genetic mechanisms underlying thermal sensitivity before making use of synthetic biology tools and quantitative genomics (Lohani et al. 2020; Razzaq et al. 2021).

Researchers need to recognize the cumulative effects of thermal load on damage and repair processes and how they will interact to affect biological responses to global change. The TLS framework builds on the established principles of the TDT model used in ecophysiology (Ørsted et al. 2022; Rezende et al. 2014). This framework forms a strong basis for further research into additional dimensions (e.g., sublethal effects, tissue types, life stages, spatial models, multiple stressors) that impact sensitivity and the underlying molecular and genetic architecture of organisms. We hope that a broader focus through the TLS framework will provide opportunities to better predict organism vulnerability in a time of profound global change. Death is just one, albeit severe, consequence of thermal stress; predicting loss of individual reproduction and ecological function while realistically incorporating dynamic environmental and biological processes is much more challenging but arguably more important for population persistence and ecological stability. Integrating these essential components into our theoretical and modeling frameworks is a step toward better understanding organism vulnerability to significant environmental stressors.

Author Contributions

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Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

The data and modelling code that support the findings of this study are openly available in the Zenodo repository at <http://doi.org/10.5281/zenodo.14271443>.

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Supporting Information

Additional supporting information can be found online in the Supporting Information section.