



Integrative Approaches to Understanding Organismal Responses to Aquatic Deoxygenation

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Abbreviations: OMZ, oxygen minimum zone; ROS, reactive oxygen species; TSR, temperature-size rule.

Abstract

Oxygen bioavailability is declining in aquatic systems worldwide as a result of climate change and other anthropogenic stressors. For aquatic organisms, the consequences are poorly known but are likely to reflect both direct effects of declining oxygen bioavailability and interactions between oxygen and other stressors, including two—warming and acidification—that have received substantial attention in recent decades and that typically accompany oxygen changes. Drawing on the collected papers in this symposium volume (“An Oxygen Perspective on Climate Change”), we outline the causes and consequences of declining oxygen bioavailability. First, we discuss the scope of natural and predicted anthropogenic changes in aquatic oxygen levels. Although modern organisms are the result of long evolutionary histories during which they were exposed to natural oxygen regimes, anthropogenic change is now exposing them to more extreme conditions and novel combinations of low oxygen with other stressors. Second, we identify behavioral and physiological mechanisms that underlie the interactive effects of oxygen with other stressors, and we assess the range of potential organismal responses to oxygen limitation that occur across levels of biological organization and over multiple timescales. We argue that metabolism and energetics provide a powerful and unifying framework for understanding organism-oxygen interactions. Third, we conclude by outlining a set of approaches for maximizing the effectiveness of future work, including focusing on long-term experiments using biologically realistic variation in experimental factors and taking truly cross-disciplinary and integrative approaches to understanding and predicting future effects.

Introduction

The detrimental effects of the lack of molecular dioxygen (hereafter, oxygen) are obvious, as encapsulated by the American Lung Association’s adage: “When you can’t breathe—nothing else matters” (American Lung Association, 2013). In terrestrial habitats, oxygen typically occurs at high concentrations in a medium (air) that is easily renewed or mixed and from which oxygen can be obtained relatively easily. In aquatic habitats, by contrast, oxygen is dissolved at low concentrations in a dense, viscous medium (water) from which it is more difficult to extract. In these freshwater and marine systems, oxygen bioavailability can be low and variable, requiring sophisticated morphologies, physiologies, and behaviors for extracting oxygen at rates high enough to support aerobic metabolism.

It is concerning, therefore, that oxygen concentrations in lakes and oceans have declined on average by about 2% in the past 50 years (Schmidtke *et al.*, 2017; Jane *et al.*, 2021), with declines of up to 7% predicted by 2100 (Keeling *et al.*, 2010). Declines are even greater in some parts of the ocean (Schmidtke *et al.*, 2017; Levin, 2018), and extreme hypoxic events are occurring more frequently (Hughes *et al.*, 2020). Here, we use the term “hypoxia” to describe partial pressures of oxygen (PO_2) that are lower than 21 kPa, which is the PO_2 of air-saturated water at sea level. Other, more specific meanings of hypoxia may be appropriate in other contexts (Vaquer-Sunyer and Duarte, 2011).

In a recent meta-analysis, Sampaio *et al.* (2021, p. 314) wrote, “Current global change-related research efforts should pay far more attention to the role of oxygen concentration as a stressor.” Indeed, although two other climate-related stressors—warming and acidification—have received substantial attention in recent years, the biological effects of deoxygenation are rarely addressed yet may have significant impacts on aquatic life. For example, Sampaio *et al.* (2021) suggest that realistic changes in oxygen availability often

have larger effects on biological performance traits, including growth, development, survival, metabolic rates, and reproduction, than do realistic changes in temperature and pH. Likewise, oxygen has more power to explain patterns of species diversity than other conditions (*e.g.*, temperature or CO_2 /carbonate chemistry) in coastal ecosystems (Sperling *et al.*, 2016) and high-altitude streams (Jacobsen *et al.*, 2003). Moreover, deoxygenation interacts with warming, acidification, and other abiotic and biotic stressors (*e.g.*, Verberk *et al.*, 2016; Reddin *et al.*, 2020; Frakes *et al.*, 2021). Unfortunately, potential interactions have been understudied, despite widespread recognition that they are important in aquatic ecosystems (Ormerod *et al.*, 2010; Hewitt *et al.*, 2016; Rillig *et al.*, 2019). Perhaps in part because the effects of insufficient oxygen are so obvious, scientific studies and the agencies that fund them have largely overlooked including oxygen as an important factor.

This article serves three purposes. First, we discuss the scope of natural and predicted anthropogenic changes in aquatic oxygen levels. The physical and biological causes are complex but relatively well understood. A key conclusion is that although modern organisms have long evolutionary histories with (diverse) natural oxygen regimes, anthropogenic change is now exposing them to more extreme conditions and novel combinations of low oxygen with other stressors.

Second, this article examines mechanistic interactions between deoxygenation and other stressors, emphasizing animal-centric perspectives. In doing so, we advocate for studying links between temporal and spatial scales of environmental change across a large suite of critical organismal responses, which range from movement to avoid hypoxic zones to the modification of phenotypes *via* plasticity (Piersma and van Gils, 2010). Ultimately, these multiple organism-level effects of deoxygenation scale up to drive higher-level population and ecosystem effects.

Third, we identify priorities for future research. Like other recent studies (Reddin *et al.*, 2020; Gruber *et al.*, 2021), we call for explicitly integrative approaches to studying interactions between deoxygenation (both external and internal) and other stressors. In addition to tests of acute environmental hypoxia, we especially call for longer-term manipulations of oxygen and other interacting factors that more closely approximate the magnitudes, variability, and durations of exposure that individuals and communities are experiencing in nature. Such efforts will be time and resource intensive—but essential to understanding the spectrum of potential responses at time-scales relevant to organisms and populations.

The points outlined above, and developed more fully below, draw on the breadth of papers in this symposium volume (“An Oxygen Perspective on Climate Change”). Collectively, these articles examine responses of a diverse set of aquatic systems to declining levels of oxygen. The studies focus on different levels of biological organization and use multiple approaches, including experimental manipulation of oxygen availability in the lab, longer-term observations of correlations between oxygen levels with the distribution and abundance of populations in nature, and historical studies of deep-time correlations between fossil communities and ocean conditions reconstructed from biogeochemical records. Rather than summarizing individual efforts here, we attempt to integrate them into a cohesive perspective that illuminates links between symposium papers and to other recent research papers.

Temporal and Spatial Scales of Environmental Change

In both lakes and oceans, oxygen levels can be highly variable in space and time, driven primarily by patterns of circulation, solubility, ventilation, photosynthesis, and respiration (Jane *et al.*, 2021; Pitcher *et al.*, 2021; Stramma and Schmidtke, 2021). Here, we distinguish natural patterns of variation—with which populations may have long ecological or evolutionary histories—*versus* anthropogenic change, which is superimposed onto these natural patterns.

Natural patterns of oxygen variability

Natural processes drive important patterns of oxygen variability in time and space in ways that are often at least somewhat predictable, both temporally and spatially. In many geographical regions, seasonal changes in surface wind velocity and direction cause upwelling events during which nutrient-rich, low-oxygen water is transported from depths to the surface, sometimes causing surface waters to become hypoxic in a matter of hours (Roegner *et al.*, 2011) or stimulating primary production of organic matter whose decay consumes oxygen over days to weeks. Another type of predictable variation is light-driven fluctuations of photosynthetic oxygen production and consumption, which occur in almost all shallow aquatic systems (Pacella *et al.*, 2018;

Nelson and Altieri, 2019; Saderne *et al.*, 2019; Lucey *et al.*, 2022). Oxygen levels can become hyperoxic during the day and hypoxic at night, and this fluctuation is amplified in warm, vegetated, or nutrient-rich conditions. Light-driven photosynthetic oxygen production in aquatic systems can, therefore, be broadly predicted from a combination of factors such as latitude, depth, and turbidity. Spatial variation is also important; for example, reef structures can restrict flow dynamics in nearshore environments, hindering oxygen replenishment and creating steep oxygen gradients on the scale of meters (Hughes *et al.*, 2020). Low-oxygen areas can also persist for long durations over both small spatial scales (*e.g.*, low-oxygen conditions encountered by aquatic animals living in organic sediments) and very large spatial scales (*e.g.*, profundal zones of stratified lakes and oxygen minimum zones in oceans).

Anthropogenic changes in oxygen availability

Superimposed on the variation described above are additional effects stemming from human activity, which can exacerbate exposure to low oxygen. Agricultural runoff, for example, increases nutrient levels in streams, lakes, and coastal areas. These increased nutrients cause algal blooms, which in turn can cause oxygen minima when the algae die off (Stramma *et al.*, 2012). The extent of “dead zones” stemming from anthropogenic nutrient inputs are increasing over time and are distributed worldwide (Breitburg *et al.*, 2018).

On longer timescales, total oxygen content is declining, and its distributions are shifting in oceans and lakes. As we continue to produce greater amounts of carbon dioxide and warm the planet, lakes and oceans hold less oxygen because of the lower solubility of oxygen in warmer water. Additionally, bodies of water become more thermally stratified, and therefore oxygen replenishment *via* convection of surface water is reduced (Reinhard and Planavsky, 2022). Rising temperatures also stimulate metabolic demand by biotic communities throughout the water column, which further reduces oxygen levels. Together, increased stratification and greater metabolic demand due to rising temperature can have greater effects on oxygen availability than does temperature-driven outgassing of oxygen *via* reductions in solubility (Fig. 1). This positive feedback loop makes acute and localized deoxygenation events even more intense (Breitburg *et al.*, 2018). At the same time, anthropogenic change is predicted to exacerbate oxygen limitation in other ways, for example, by lengthening the upwelling season and increasing upwelling intensity (Wang *et al.*, 2015). Organisms that are adapted to variable oxygen environments generally have physiological mechanisms to maintain oxygen uptake or reduce demand in the short term. These mechanisms often require greater allocations of energy, which may negatively impact fitness, especially during long-term or repeated exposure to hypoxia (Woods and Moran, 2020). The ability of aquatic organisms to cope with natural fluctuations in oxygen, in both the short

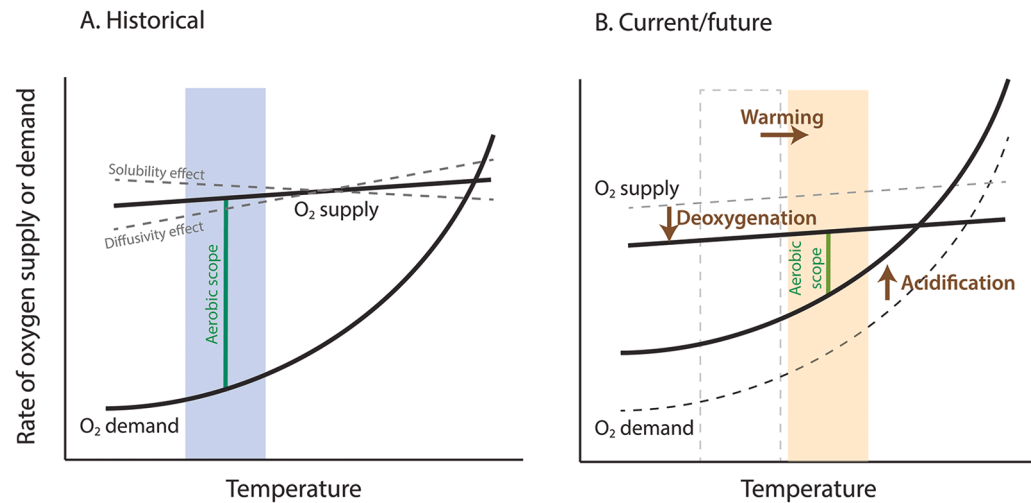


Figure 1. Schematic showing how components of the deadly trio—deoxygenation, warming, and acidification—combine to affect oxygen supply: demand relationships. (A) Aerobic organisms use oxygen to support their metabolisms, and demand for oxygen typically rises rapidly with warming over the short term, at least prior to acclimation or evolutionary adaptation. Simultaneously, the physical processes affecting rates of oxygen supply (solubility and diffusion coefficient of oxygen, shown as dotted lines; additional effects of temperature on viscosity of water are not shown) combine to cause rates of supply to increase approximately linearly with temperature but often much less steeply than demand. The aerobic scope (difference between resting metabolic demand and potential rate of oxygen supply; green line) defines the amount of metabolic power that can be devoted to tasks other than basal maintenance. Here, the historical range of temperatures is marked in blue, corresponding to a large aerobic scope. (B) Climate change is causing oxygen levels in oceans to decline, both from temperature-related declines in solubility but also from shifts in respiration by community members and changes in patterns of stratification and oxygen transport. Combined, these effects are depressing functional rates of oxygen supply to organisms. In addition, warmer temperatures (range indicated in orange) are stimulating short-term demand for oxygen more than they are increasing supplies. Finally, acidification has a wide range of direct effects but also likely is raising metabolic demand for oxygen by increasing costs of pH homeostasis and calcification. Organisms experiencing one or more conditions in the deadly trio may engage in greater overall levels of activity in attempts to find better conditions locally. Together, these effects may strongly narrow the aerobic scope.

and long term, will strongly influence their resilience to climate change-induced deoxygenation during the Anthropocene. Organisms that have evolved in environments with natural oxygen fluctuation will likely be more capable of responding adaptively to deoxygenation (Fig. 2).

Mechanisms Underlying the Organismal Consequences of Interactions between Low Oxygen and Other Stressors

Hypoxia, acidification (and associated hypercapnia, which refers to elevated levels of CO₂), and warmer temperatures

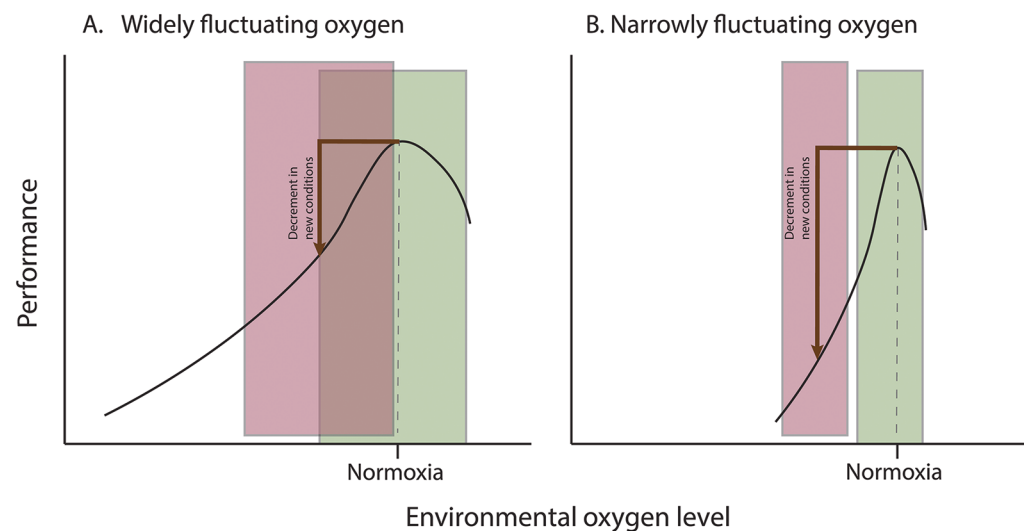


Figure 2. Natural oxygen regime is hypothesized to shape oxygen performance curves and vulnerability to deoxygenation. Reductions in performance likely differ for species that have evolved under variable oxygen conditions (a) and those from more stable conditions (b). In both panels, historic variability in oxygen conditions is indicated in green, while red indicates the new oxygen regime, which has the same fluctuation but a lower mean. Owing to differences in the breadth of the oxygen performance curve, loss of performance is less pronounced in species from variable oxygen conditions. Performance in hyperoxia (oxygen levels higher than normoxic) has been explored less than in hypoxia and can be highly variable among organisms and situations, with some organisms performing well and others showing performance decrements.

often co-occur, constituting “the deadly trio” (Fig. 1). In combination, these and other stressors can synergistically impact organismal performance and survival (Verberk *et al.*, 2016; Howard *et al.*, 2020; Lucey *et al.*, 2020; Tomasetti *et al.*, 2021). One approach to understanding the consequences of stressor combinations, and to predicting their impacts during future climate change, is to better understand the physiological mechanisms by which interactions among stressors are manifest within organisms. Here, as a start, we identify a number of two-way interactions involving oxygen, with the goal of highlighting relevant physiological processes that underlie interaction effects.

Oxygen-temperature interactions

With increasing temperature, metabolic demand for oxygen generally rises. Increases in demand reflect in part the acute effects of temperature on the thermodynamics of chemical reactions, but even after thermal acclimation, metabolic rates tend to be 2- to 3-fold higher in conditions that are 10 °C warmer (Q_{10} of 2–3; Seebacher *et al.*, 2015). This reflects the pervasive effects of warming on multiple life processes, such as feeding, activity, growth, and reproduction, all of which increase with warming up to a point and stimulate energy demands (Clarke and Fraser, 2004).

With increasing temperature, the solubility of oxygen declines (Dejours, 1981), and this is often cited as a source of hypoxic stress for organisms. Often overlooked, however, is that as oxygen solubility decreases with temperature, rates of oxygen diffusion increase (Woods, 1999; Verberk *et al.*, 2011; Boag *et al.*, 2018). Warmer water is also less viscous, which thins the boundary layers that act as diffusion barriers and lowers the cost of ventilating respiratory surfaces (Couturier *et al.*, 2007; Verberk and Atkinson, 2013). The net effect is therefore a slight rise in oxygen bioavailability with temperature (see Fig. 1; Atkinson *et al.*, 2022). With increasing temperature, however, rising metabolic demand for oxygen due to increased physiological rates ($Q_{10} \sim 2\text{--}3$ even after acclimation) typically outpaces increases in oxygen bioavailability ($Q_{10} \sim 1.1\text{--}1.4$). Depending on organism-specific gas exchange and delivery mechanisms, some aquatic organisms in warmer waters may therefore become oxygen limited. Such oxygen limitation arises not because of decreasing bioavailability but because the increase in supply cannot keep up with the increase in demand, or the costs of supply are exceedingly high (Verberk and Atkinson, 2013; Audzijonyte *et al.*, 2019; Rubalcaba *et al.*, 2020).

Thermally driven mismatches between oxygen supply and oxygen demand have been proposed as a unifying concept to explain thermal tolerance and geographic distribution patterns (Pörtner, 2012; Deutsch *et al.*, 2015). Although detrimental effects of hypoxia are widely acknowledged to be exacerbated by warming, whether warming causes oxygen to become limiting under normoxia has been debated (Verberk *et al.*, 2016; Jutfelt *et al.*, 2018; Lefevre *et al.*, 2018; Pauly and Cheung, 2018; Seibel and Deutsch, 2020; Wootton

et al., 2022). A nuance here is that not all organisms are equally at risk from oxygen-temperature interactions (Verberk *et al.*, 2016). Differences between organisms in their efficiency and regulation of oxygen uptake suggest that although thermal effects can sometimes be understood from an oxygen perspective, this is not a one-size-fits-all theory (Verberk *et al.*, 2018; Ern, 2019).

A slightly different take on oxygen-temperature interactions is seen in intertidal systems, in which marine organisms are exposed to air for extended periods. During these periods of aerial exposure, oxygen uptake can be inhibited as a result of a need for water conservation or because animals have aquatic-adapted gas exchange mechanisms that function poorly in air (Barnes *et al.*, 1963; Newell, 1973; Davenport and Irwin, 2003). Many intertidal organisms have evolved behavioral, physiological, and biochemical mechanisms that confer high tolerance to internal hypoxia during these times (Falfushynska *et al.*, 2020), but warming temperatures are likely to increase the cost of deploying these mechanisms and to exacerbate associated oxygen deficits through increased metabolic rates (Horn *et al.*, 2021).

Interactions between oxygen and hypercapnia/acidification

Increasing levels of atmospheric CO₂ (hypercapnia) alter the carbonate chemistry of seawater and reduce ocean pH (ocean acidification). Locally, acidification and hypoxia often co-occur, since faster rates of biological respiration cause both O₂ consumption and CO₂ production to increase, which can accelerate declines in pH in hypoxic areas (Feely *et al.*, 2010). Moreover, oxygen and pH covary naturally because both are affected by light and stratification of the water column (see *Natural patterns of oxygen variability*, above).

The combined and interactive effects of hypoxia and acidification remain poorly understood, although they share common links to respiration (Gobler and Baumann, 2016). Low environmental oxygen can directly depress oxygen content of blood and tissues, while high CO₂/low pH in the environment, if directly translated into the interior of the organism, can modulate the oxygen-binding affinity and capacity of oxygen transport proteins (when present) (Coates *et al.*, 2022). Thus, both stressors can elicit compensatory changes in ventilation, circulation, and acid-base status with associated changes in metabolism (Lehtonen and Burnett, 2016). Models based on bioenergetics and aerobic scope may thus prove useful for understanding and predicting organismal responses to the interactive effects of low oxygen/high CO₂.

Reduced oxygen bioavailability and acidification generally have negative additive or synergistic effects on fitness-related parameters such as survival, growth, activity, and metabolism (Gobler and Baumann, 2016; Sui *et al.*, 2016; Gu *et al.*, 2019), with responses varying between species, life stages, and exposure histories. In contrast, some studies have shown that elevated CO₂ can enhance the oxygen affinity of respiratory pigments (Mangum and Burnett, 1986), improve

aerobic performance in hypoxia (Stover *et al.*, 2013; Lehtonen and Burnett, 2016), and increase hypoxia tolerance (Montgomery *et al.*, 2019). Alone or in combination, low oxygen and acidification can also perturb the complex mechanisms employed by multicellular aquatic organisms to defend themselves against microbial infection by altering key components of the immune system (*e.g.*, the generation of reactive oxygen species [ROS], unstable molecules containing oxygen that are implicated in cell signaling and cell damage; see Box 1) as well as changing the abundance, distribution, and pathogenicity of microbial populations (Burnett and Burnett, 2022).

Mounting an immune response has been shown to impair metabolism in some crustaceans and to reduce levels of circulating oxygen (Burnett and Burnett, 2015). Fundamental questions remain about how aquatic organisms sense and respond to hypoxia (Sokolova, 2018; de Lima *et al.*, 2021) as well as how and to what extent they balance the competing energetic demands of maintaining immunity and fitness while responding to changes in oxygen, acidification, temperature, and salinity (Lucey *et al.*, 2020).

Oxygen-osmoregulation interactions

Gas-exchange surfaces generally are highly permeable. In fishes and many other water-breathing ectotherms, gills are the sites of both oxygen uptake and of water and ion movement. To maintain oxygen uptake, fishes exposed to aquatic

hypoxia often compensate by mechanically hyperventilating their gills (Farrell and Richards, 2009) or, over longer periods of time, remodeling gill structures in ways that enhance oxygen uptake, a form of plasticity (Nilsson *et al.*, 2012). Because gills are also a major site of ion and water exchange, hyperventilation also increases the cost of osmoregulation by increasing the diffusive movement of ions across the gills and thus the rate of compensatory active transport required for maintaining blood osmolality (Perry *et al.*, 2003; Evans *et al.*, 2005; Grosell *et al.*, 2010) (increased gill area may also raise the risk of infection; Nilsson *et al.*, 2012). This trade-off between gill structures optimized for oxygen uptake (*i.e.*, sufficiently high surface area and low diffusion resistance) and for ion regulation (*i.e.*, low surface area and high diffusion resistance) is called the osmorepiratory compromise (Randall *et al.*, 1972; Nilsson *et al.*, 2012). Likewise in aquatic insects, differences in breathing mode (aquatic gills *vs.* aerial gas exchange) are associated with sensitivity to salt (Verberk *et al.*, 2020). In euryhaline fishes, the cost of osmoregulation appears to be species specific, with estimates ranging from virtually zero to one-third of standard metabolism (reviewed by Ern *et al.*, 2014; see also Nilsson *et al.*, 2012). Furthermore, increasing salinity has been associated with reduced hypoxia tolerance in fish (Rogers *et al.*, 2016; Verberk *et al.*, 2022). In coastal and estuarine marine invertebrates, similarly high costs of osmoregulation may be incurred, especially when organisms hyperregulate in low-salinity water (Rivera-Ingraham

Box 1: Causes and consequences of reactive oxygen species

Reactive oxygen species (ROS) are small molecules and free radicals derived from molecular oxygen, for example, the superoxide anion ($\cdot\text{O}_2^-$) and hydrogen peroxide (H_2O_2). Reactive oxygen species are highly reactive, reacting with the first molecule with which they come into contact and thus causing damage to proteins, lipids, and DNA, a situation referred to as oxidative stress. Although low levels of ROS may be necessary for normal functioning as regulatory mediators in signaling processes (Dröge, 2002; Boardman *et al.*, 2012), oxidative stress contributes to senescence and ultimately death (Fenn *et al.*, 1967; Fridovich, 1998; Lane, 2002).

Reactive oxygen species can come from exogenous or endogenous sources. A primary exogenous source is high light intensities, especially in the ultraviolet part of the spectrum. Primary endogenous sources are mitochondria and chloroplasts (Murphy, 2009; Foyer, 2018). During the normal process of oxidative phosphorylation by mitochondria, for example, electrons leak and interact with molecular oxygen to form ROS. Rates of ROS production increase with mitochondrial membrane potential and are therefore minimized when rates of oxygen delivery to mitochondria (O_2 supply) match rates of oxygen utilization (O_2 demand). Consequently, oxygen delivery and utilization have to be tightly regulated to balance the generation of energy with the production of toxic oxidants. This is seen as a primary reason that oxygen delivery is regulated at many levels (*e.g.*, reversible oxygen binding with hemoglobin, dynamic regulation of both blood flow and ventilation).

Because of the cytotoxic nature of ROS, many organisms have evolved mechanisms to prevent or neutralize them and to repair the damage they cause. For example, many organisms produce pigments that absorb and then dissipate excess light energy to prevent exogenous ROS production (*i.e.*, melanin, xanthophyll; Goss and Latowski, 2020). Similarly, uncoupling proteins reduce mitochondrial membrane potential and ROS production. Once ROS are formed, cells can utilize antioxidant enzymes such as superoxide dismutase, peroxidase, and catalase, transforming them into less reactive molecules (Zelko *et al.*, 2002). Cells also have mechanisms to limit or repair the damage that occurs as a result of oxidative stress, including the mobilization of molecular chaperones, hydrolyzed nucleotides, and DNA repair enzymes (Nakabeppu *et al.*, 2004).

Although a direct link between the deoxygenation of aquatic ecosystems and oxidative stress in aquatic organisms has not been established, both deoxygenation and oxidative stress are increasing because of global warming (Paital, 2016), meaning that aquatic organisms will increasingly face each of these stressors and their compound effects as global temperatures rise.

and Lignot, 2017). Aquatic deoxygenation may thus have a broad array of effects on osmoregulatory capacities and energetics.

Oxygen-light interactions

The intrinsic, irradiance-driven covariance of light and photo-synthetically produced oxygen affects organisms in both positive and negative ways. As one example, coral reefs may be among the most vulnerable ecosystems to light conditions, especially in combination with the deadly trio of deoxygenation, warming, and acidification. Hermatypic, or calcifying, corals (Scleractinia) can tolerate considerable variation in light and oxygen, which naturally covary because of the diel cycle in oxygen availability *via* photosynthesis during the day and respiration at night. However, excess light (among other factors) can cause corals to expel their symbionts (bleaching), especially when they are thermally stressed (Fitt *et al.*, 2001; Bhagooli and Hidaka, 2004). The reasons for symbiont expulsion remain unresolved, but one prominent hypothesis implicates increased production of ROS by symbionts when they are stressed by too much heat or light, which causes oxidative stress in both coral and symbiont tissues (Downs *et al.*, 2002; Curran and Barnard, 2021).

Via photosynthesis, light can also improve the availability of oxygen to developing embryos in egg masses (Phillips and Moran, 2015) and to the tissues of heat-stressed animals that harbor stolen chloroplasts.

On a broader scale, hyperoxia in surface waters (due to light-driven photosynthesis) may offset some of the increased metabolic costs that come with warmer temperatures in coastal waters (Giomi *et al.*, 2019). Finally, in the water column, light is required for highly visual marine animals, and oxygen stress can disrupt their visual physiology, behavior, and ecology (McCormick and Levin, 2017; McCormick *et al.*, 2022). Hypoxia-impaired vision may impact essential behaviors such as feeding, reproduction, vertical distribution, and predator avoidance in visual organisms.

Metabolism and energetics as central integrators

Although the interactions discussed above emerge from diverse physiological mechanisms, they are linked into a common framework *via* their effects on energy. Much of the energy that aquatic organisms use to combat stressors comes from aerobic metabolism, which of course depends on adequate supplies of oxygen. The effects of deoxygenation may therefore manifest not just as direct physiological impacts on organisms (*e.g.*, anaerobiosis, reduced performance) but also in how metabolic energy is allocated among other competing functions like growth, reproduction, vision, food capture, anti-predator defense, and so on, when organisms are under stress (*e.g.*, Roman *et al.*, 2019). To be adaptive, organismal responses must not only help the organism tolerate or avoid the stress but also do so in ways that minimize overall reductions in fitness. For example, if an organism behaviorally responds to oxygen limitation by moving from hypoxic

to better-oxygenated areas, oxygen gains may be traded off against other ecological costs of changing location (*e.g.*, increased risks of predator encounter; Roman *et al.*, 2019).

How often does oxygen rather than energy limit performance or fitness? The answer will depend on the temporal and spatial distribution of metabolic demand relative to the size of safety margins protecting the organism from immediate impacts of resource budget deficits. In the short term, organisms are expected to die much sooner from suffocation than from starvation, because stores of oxygen within an organism typically are negligible compared with stores of energy. On somewhat longer timescales, an important safety margin is the aerobic scope, which is defined as the difference between maximum and standard metabolic rates (the latter is the minimal metabolic rate needed to sustain bodily processes). *Via* effects on aerobic scope, each leg of the deadly trio can be linked explicitly to energetics (Fig. 1; Jacobsen *et al.*, 2003; Pörtner and Knust, 2007; Verberk *et al.*, 2016; Deutsch *et al.*, 2020; Rubalcaba *et al.*, 2020; Seibel and Deutsch, 2020). Each stressor narrows the aerobic scope, either by depressing oxygen supplies (*e.g.*, deoxygenation effect beyond the temperature effect on oxygen solubility) or by increasing metabolic demands (*e.g.*, rising temperature, increases in acidification or pollution, changes in salinity). The narrowing of aerobic scope limits the energy available to organisms for fitness-enhancing activities such as growth and reproduction.

Short-term experiments will likely be biased toward finding that oxygen rather than food limits performance, although the intensity of the challenge will also play a role. However, this bias does not mean that oxygen is in general more limiting than food. Indeed, low-intensity but chronic increases in metabolic demand (*e.g.*, from a prolonged infection, rising temperatures, or increased ventilation of gas exchange structures) may cause longer-term limitation of food than of oxygen.

Timescales and Levels of Biological Response

Potential responses to deoxygenation and associated environmental stressors occur at all levels of biological organization and at all temporal scales, ranging from rapid molecular and behavioral responses to long-term evolutionary responses. Understanding and predicting future responses will thus likely emerge from integrative studies that work across scales, investigating not only how interacting effects of oxygen affect the performance of organisms but also the physiological mechanisms that give rise to these responses. The effectiveness of different responses is a function of the temporal scale and intensity of hypoxia (Fig. 3) and will vary across species that have experienced different patterns of oxygen variation during their evolutionary histories (Fig. 2). In addition, organisms generally employ a suite of responses (each with their own timescale), because no single response is likely to completely alleviate the negative effects of hypoxia. Moreover, even relatively small impacts of mild hypoxia (and, correspondingly, small benefits gained from deploying physiological

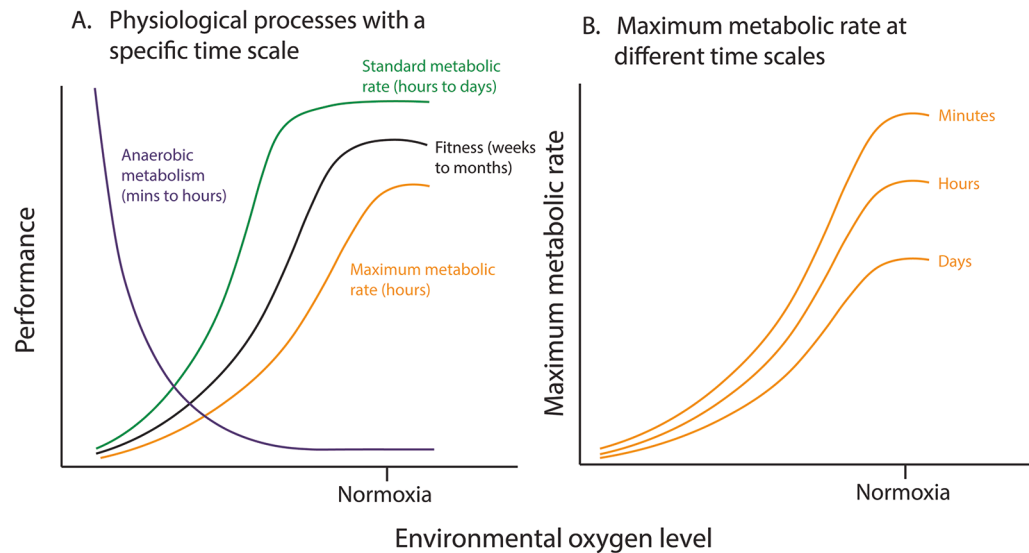


Figure 3. Physiological performance and temporal scale. Effects of oxygen on performance are modulated by temporal scale, with specific physiological processes changing on specific timescales. (A) For example, anaerobic metabolism will be used most at low partial pressure of oxygen (PO_2) and can supplement energy budgets on short temporal scales only. (B) In addition, a given physiological process (e.g., maximum metabolic rate) may be affected by oxygen availability differently when it has to be sustained for minutes, hours, or weeks, and long-term exposure to hyperoxia could even reduce performance as a result of toxicity effects.

mechanisms to alleviate it) can add up to large cumulative effects on fitness over time. Evaluating the chronic, sublethal effects of mild hypoxia and the organismal responses to avoid these effects is therefore essential to predicting long-term effects on organisms and ecosystems. From another perspective, organisms that are part of communities associated with habitats consistently low in oxygen, such as oxygen minimum zones, often have a broad range of mechanisms and phenotypes for coping with low oxygen levels, and these can act on a wide variety of timescales (see Box 2). Organisms adapted to consistently low oxygen may experience expanded habitat availability as hypoxic zones in the oceans grow.

Behavioral responses

Behavioral responses to hypoxia can be employed nearly instantaneously and are often a first line of defense. One ex-

ample is movement between oxygen microhabitats; different habitats offer gradients in oxygen availability across distinct spatial and temporal scales, which organisms can exploit in different ways depending on their stage, life history, body size, and mobility, in ways that are analogous to how organisms can exploit mosaics of locally available temperatures (Woods *et al.*, 2015). When faced with hypoxia, for example, mobile aquatic organisms may move to areas of higher oxygen, but this can expose them to other stressors such as increased temperature near the surface, higher irradiance, or predation (Koslow *et al.*, 2011; Larsson and Lampert, 2011). Hypoxia may also drive changes in visually based behaviors, including photobehavior (McCormick *et al.*, 2022), and alter how readily predators detect or capture prey as well as the ability of prey to detect and avoid predators (Breitburg *et al.*, 1994). By contrast, some aquatic organisms move into

Box 2: An integrative example: multiple responses by pelagic organisms in oxygen minimum zones

Organisms that inhabit oxygen minimum zones (OMZs) for much or all of their lives typically have morphological and physiological adaptations allowing them to maximize O_2 extraction, including enhanced capacity for ventilation and circulation, high gill surface area, high O_2 binding capacity and affinity of oxygen transport proteins, and a large Bohr effect (Childress and Seibel, 1998). Moreover, organisms that move transiently into OMZs typically are oxyregulators that maintain O_2 consumption rates when ambient levels of oxygen remain above some critical partial pressure (P_{crit}). A well-documented example is the jumbo squid, *Dosidicus gigas*, which migrates vertically on a daily basis into the OMZ in the Eastern Tropical Pacific (Gilly *et al.*, 2006). It copes with hypoxia by suppressing its metabolism and changing its swimming behavior (Rosa and Seibel, 2008). At environmental partial pressure of oxygen (PO_2) (below 1.6 kPa; at depths below 150 m), *D. gigas* switches from aerobic to anaerobic pathways (Rosa and Seibel, 2008). Similar responses have been described in other vertically migrating organisms. An additional challenge for diel migrators is the generation of reactive oxygen species (see Box 1), promoted by reoxygenation as they move into surface waters at night. Jumbo squid minimize damage from ROS by enhancing the heat shock protein response and the activity of key antioxidant enzymes (Trübenbach *et al.*, 2013).

hypoxic areas to escape from predators that cannot tolerate low oxygen (e.g., *Daphnia pulicaria*; Larsson and Lampert, 2011).

Another potential behavioral response to hypoxia is reduced feeding activity. Many organisms, including fish (Thetmeyer *et al.*, 1999; Magnoni *et al.*, 2018), crabs and molluscs (e.g., Das and Stickle, 1993), and annelids (Llanos and Diaz, 1994) feed at lower rates in hypoxia. Physiological effects of low oxygen on reproductive behaviors and investment are also possible (Wu and Or, 2005), although these effects are understudied.

Molecular responses and oxygen transport protein

Although many smaller organisms (<1 mm) can fulfill their metabolic needs by accessing oxygen through diffusion alone, larger organisms require circulatory systems and often utilize oxygen transport proteins (Decker and van Holde, 2010), which can enhance oxygen carrying capacity of blood and hemolymph by more than an order of magnitude. The active sites of oxygen transport proteins contain pro-oxidative metals—either iron (hemoglobins, hemerythrins) or copper (hemocyanins in arthropods and molluscs)—that not only enable the binding and transfer of O₂ but also restrict the production of harmful ROS (see Box 1) such as hydroxyl radicals (OH; Decker and van Holde, 2010; Coates and Talbot, 2018).

How will oxygen transport proteins interact with and respond to deoxygenation, warming, and acidification? First, oxygen transport proteins demonstrate impressive resistance to thermal denaturation: hemoglobins and hemocyanins remain undissociated at temperatures >50 °C and >90 °C, respectively (e.g., Georgieva *et al.*, 1998; Carvalho *et al.*, 2013). Thus, oxygen transport proteins are likely to remain structurally intact under substantial changes in ocean conditions (see Coates *et al.*, 2022), although their functional robustness *in vivo* will also depend on temperature effects on other processes. Second, organisms can adaptively increase oxygen transport capacity by increasing the expression of oxygen transport proteins or express isoforms with a different oxygen affinity (Decker and Föll, 2000; Brown-Peterson *et al.*, 2005). These changes typically occur on the timescale of hours to days; for example, the shrimp *Penaeus vannamei*, when exposed to low oxygen levels alone or in combination with elevated carbon dioxide for 24 hours, altered its mRNA profiles to include a novel hemocyanin isoform among other oxygen-associated regulatory pathways (Johnson *et al.*, 2015). Third, temperature-driven increases in metabolic demand by tissues can lead to rising levels of CO₂ and lactic acid, and the accompanying decreases in pH lowers the oxygen affinity of oxygen transport proteins, thereby promoting release of oxygen (the Bohr effect; Weber *et al.*, 2008). Such changes are virtually instantaneous (within minutes), facilitated by the built-in sensitivity of oxygen transport proteins to pH and the heterogeneity of oxygen transport protein subunits that underlie functional adaptations of organisms to their environments.

Long-term increases in water temperature, however, may lead to metabolic fatigue or even derangement (uncontrolled release of damaging free radicals), enhancing the susceptibility of organisms to disease (reviewed by Coates and Söderhäll, 2021). Oxygen transport proteins clearly have the potential to assist organisms in coping with some level of increased temperature and deoxygenation. What is unclear (and unstudied), however, is whether these proteins will evolve adaptively in response to climate change, especially in long-lived organisms (e.g., decapod crustaceans and shelled molluscs).

Physiological acclimation

On slightly longer timescales, organisms can recruit a variety of physiological mechanisms to help cope with the effects of hypoxia (Fry, 1971; Harrison *et al.*, 2018). In general, these mechanisms fall into three categories: those that increase oxygen uptake and transport, supporting aerobic metabolism by helping to maintain cellular oxygen supply; those that support metabolism by increasing reliance on anaerobic processes; and those that reduce cellular oxygen demand by depressing metabolism (Farrell and Richards, 2009; Richards, 2009). These mechanisms are not mutually exclusive, and a given species will employ one or more at different levels of oxygen stress or over different time courses of exposure (Fig. 3). Which strategies are employed appears to depend on the duration and intensity of the hypoxic stress that species experience in their natural habitats (Mandic and Regan, 2018). This suggests some degree of evolutionary matching between environmental conditions and the ability of species to mount effective responses. However, mismatches between oxygen supply and demand are likely to increase with climate change because organisms are likely to encounter environmental stressors at higher levels, for longer durations, and in novel combinations. At some point, an organism's acclimatory coping responses may become inadequate.

The capacity for acclimation to hypoxia varies substantially between species, with some species showing little capacity and others demonstrating much greater plasticity (Borowiec *et al.*, 2020). The process of acclimation to higher temperatures may also be relevant to an organism's ability to cope with subsequent exposure to hypoxia. Across a range of taxa, acclimation to moderately increased temperature generally tends either to have no effect or to improve performance in moderate hypoxia (Collins *et al.*, 2021). However, relatively few studies have used designs suitable for rigorously assessing these patterns (Collins *et al.*, 2021). There are even fewer studies that examine the capacity for acclimation to combinations of increased temperature and reduced oxygen. This is an area that is ripe for further investigation.

In addition, much of what we know about the ability of organisms to adjust to hypoxia comes from laboratory studies that expose organisms to constant rather than fluctuating oxygen conditions. In nature, fluctuating oxygen conditions

are common, yet much less is known about the capacity for physiological plasticity in this type of exposure (Remen *et al.*, 2012; Williams *et al.*, 2019). The relatively limited data that exist suggest that different mechanisms may be recruited depending on the pattern of exposure and, thus, that the strategies for coping with constant and intermittent hypoxia may differ. For example, in Atlantic killifish (*Fundulus heteroclitus*), acclimation to constant, but not intermittent, hypoxia was associated with physiological changes that may reduce the costs of ionoregulation, suggesting an overall strategy of metabolic suppression. In contrast, acclimation to intermittent hypoxia was associated with physiological changes that improved the capacity to recover from hypoxia during intervening normoxic periods (Borowiec *et al.*, 2015, 2018).

Plastic and evolutionary responses of body size

Higher developmental temperatures generally cause ectotherms to grow faster but to mature earlier at smaller sizes (Ray, 1960; Atkinson, 1994; Ohlberger, 2013), a phenomenon known as the temperature-size rule, or TSR (Atkinson, 1996). Support for a role for oxygen comes from the observations that (1) the TSR is more common and pronounced in aquatic environments (Forster and Hirst, 2012; Horne *et al.*, 2015), where oxygen bioavailability is substantially lower than in aerial systems (Verberk and Atkinson, 2013); and (2) experimental reduction in oxygen availability can lead to a more pronounced TSR (Frazier *et al.*, 2001; Hoefnagel and Verberk, 2015). However, evidence from experimental studies is still limited, and connections to oxygen are debated (Audzijonyte *et al.*, 2019; Verberk *et al.*, 2021). One interesting experimental example is *Lecane inermis*, a rotifer for which, like most animals, fecundity increases with body size. However, under a combination of high-temperature and low-oxygen conditions, smaller individuals were more fecund than larger ones, indicating that size reductions with warming constituted an adaptive response to temperature-dependent oxygen conditions (Walczyńska *et al.*, 2015).

Traditional explanations for the TSR invoke proximate mechanisms such as physiological constraints on the uptake of oxygen or other resources, which can be modeled with supply and demand-type growth models (Pütter, 1920). The TSR, however, can also occur in the absence of changes in baseline metabolic rate or in experiments with ample oxygen and food, suggesting that explanations need not invoke temperature-driven oxygen or food deficits (Wootton *et al.*, 2022). Moreover, cause and effect can be difficult to disentangle. For example, debate around the role of oxygen in the TSR in fish revolves around the question of whether the proportionally smaller gill surface areas of larger fish are a physiological constraint on oxygen uptake (*i.e.*, a cause) or whether they reflect proportionally lower oxygen requirements in larger fish (*i.e.*, a consequence) (Marshall and White, 2019; Pauly, 2019). Organisms generally have multiple mechanisms to en-

hance oxygen uptake, and these can confound experimental tests of oxygen-limited performance at high temperatures and large body sizes (Shishido *et al.*, 2019).

Ultimate explanations rooted in life-history theory suggest instead that changes in body size should be viewed from the perspective of optimally allocating resources to competing functions such as growth, maturation, and reproduction (Hoefnagel *et al.*, 2018; Audzijonyte *et al.*, 2019; Marshall and White, 2019; Verberk *et al.*, 2021). This could be related to temperature-dependent rates of mortality, relationships between size and fecundity, or costs of reproduction (Kozłowski *et al.*, 2004; Arendt, 2011). Indeed, temperature-driven changes in oxygen demand and supply could themselves select for optimal reaction norms as encapsulated by the “Ghost of Oxygen Limitation Past” hypothesis, which postulates that organisms have evolved to maintain oxygen provisioning under a range of conditions by different means (reducing activity, reducing growth, reducing cell size, early maturation at a smaller size) and that past phenotypes that did not accurately predict and prepare for oxygen limitation would be selected against (Verberk *et al.*, 2021). The life-history optimization model presented in this issue (Audzijonyte *et al.*, 2022) shows that neither life-history optimization nor physiological changes alone are likely to explain TSR responses, but that both are operating simultaneously. These results suggest that although oxygen constraints may shape adult body size in warmer conditions, they operate within a set of life-history trade-offs.

Similar questions arise on longer timescales. The role of oxygen in the evolution of body size was first suggested by comparative analyses of arthropods achieving larger (even giant) size when available levels of oxygen are high. One example is paleogigantism of dragonflies and other insect species during the Carboniferous, when oxygen levels in the atmosphere may have exceeded 31% (Berner *et al.*, 2007). Another (contemporary) example is polar gigantism, which aligns with generally greater oxygen availability in water toward the poles (Chapelle and Peck, 1999; Moran and Woods, 2012; Lane *et al.*, 2017). Although the physiological mechanisms underlying the two patterns are not completely analogous (Verberk and Atkinson, 2013), together they have led to broader analyses of correlations between body size and oxygen availability.

Evolution may be tracked as either a process or a product; the former can reveal mechanisms, while the latter shows their context-dependent outputs. From the process perspective, evolution of smaller body size under hypoxia has been experimentally demonstrated in some taxa (Zhou *et al.*, 2007; Klok and Harrison, 2009; Walczyńska and Sobczyk, 2022). Natural experiments (product) have also shown correlations between environmental oxygen levels and body size across species (Chapelle and Peck, 2004; Walczyńska *et al.*, 2021), and a recent paper reports fishes with a larger body size and cell size to be more susceptible to low oxygen in warm water (Verberk *et al.*, 2022). Furthermore, in intense oxygen minimum zones (see Box 2), benthic metazoan meiofauna and

small macrofauna dominate, whereas larger invertebrate taxa are rare (Levin, 2003; Gooday *et al.*, 2010). These experiments and patterns suggest that, at least from the perspective of small ectotherms, global declines in body size, which increasingly are thought to be driven by climate warming (Dau-fresne *et al.*, 2009), reflect functional oxygen deficiencies or functional hypoxia (Harrison *et al.*, 2018). So far, no data for evolutionary patterns are available for larger ectotherms, which prevents developing more universal conclusions.

Population ecology

Organismal-level effects that stem from rising temperatures and deoxygenation readily scale up to influence the population ecology and ranges of both pelagic and benthic species (Levin *et al.*, 2009; Cheung *et al.*, 2013; Roman *et al.*, 2019; Pinsky *et al.*, 2020; McGinty *et al.*, 2021). Shifts in population structure and ecology may result from multiple factors, including altered patterns of migration and dispersal, changes to the timing of reproduction and to the allocation of resources (e.g., trade-offs between offspring size and number), as well as biotic interactions among community members. As one example, warmer, hypoxic waters typically depress the body sizes of zooplankton and fish (see *Plastic and evolutionary responses of body size*, above), driving corresponding decreases in fecundity (Blueweiss *et al.*, 1978; Barneche *et al.*, 2018). Likewise, warmer, hypoxic waters can depress the efficiency of prey capture, growth, and reproduction, which together impact the productivity of populations. Recent evidence and models suggest that ocean warming and deoxygenation will progressively move and compress suitable habitats for marine organisms, with strong impacts on future species distributions (Deutsch *et al.*, 2015; Pinsky *et al.*, 2020).

Recommendations for Future Studies

Construct longer, multifactorial experiments using realistic environmental conditions

Our summary above leads to two conclusions about the kinds of studies needed going forward. Echoing other authors working on multifactorial approaches (Boyd *et al.*, 2018; Wootton *et al.*, 2022), we recommend that studies consistently build in interactions between oxygen and other relevant stressors. Such an approach rapidly expands the necessary number of treatment groups, although there are ways to avoid full-factorial experiments using, for example, experimental versions of a Morris analysis (implementing just subsets of all possible factorial treatments (Morris, 1991) or other ways of rationalizing and implementing fewer treatments (Boyd *et al.*, 2018).

Second, although short-term experiments are easier and less expensive to carry out, we need more long-term experiments, especially those that address intergenerational acclimation across at least three or four generations. Longer-term experiments will start to mimic the timescales of change occurring in nature (Brown *et al.*, 2001; Kuebbing *et al.*, 2018). Such experiments will allow us to determine not just whether

organisms can tolerate or compensate for short-term environmental change but also for the effects of modest but persistent changes. For example, emergency physiological responses to hypoxia (e.g., increased ventilation) may allow organisms to withstand short-term exposures but may become energetically unsustainable over longer periods of time (Woods and Moran, 2020). Also important is understanding the effects of increasing frequency of moderate or extreme stressors such as marine heat waves and hypoxic events. One potential approach would be to adopt for oxygen (and its interactions) the tolerance landscapes approach used for temperature (Rezende *et al.*, 2014), in which one analyzes responses to progressively longer exposures and then estimates responses to even longer exposures by projection (Fig. 3). Note also that longer-term experiments can more easily accommodate ranges of experimental hypoxia that are relevant to the system in question. Clearly, such efforts will require more time and money, and we urge funding agencies to prioritize this sort of work.

Report measured conditions more comprehensively

One problem impeding progress is lack of standardized approaches to reporting units and experimental approaches (Hofmann *et al.*, 2011). Some fields, such as physics, biogeochemistry, and ecology, tend to work in terms of oxygen concentrations, whereas others, such as physiology and organismal biology, use percent air saturation or partial pressures. As has been pointed out previously (e.g., Verberk *et al.*, 2011; Ultsch and Nordlie, 2019), oxygen concentration alone is not a sufficient predictor of organismal performance in aquatic systems. Oxygen moves into organisms' down-gradients of partial pressure, and the bioavailability of oxygen is therefore affected not just by concentration but also by the individual and interacting effects of factors such as temperature, solubility, diffusivity, viscosity, pressure, and water flow (Denny, 1993; Woods, 1999; Verberk *et al.*, 2011; Rubalcaba *et al.*, 2020; Frakes *et al.*, 2021; Atkinson *et al.*, 2022). As a result, data on environmental oxygen should be reported with associated parameters (temperature, atmospheric and hydrostatic pressure/depth, salinity, and flow fields if available) so that values can be compared across locations, units, and organisms. More broadly, to provide better ecological and physiological context, we need more comprehensive reporting of environmental conditions in space and time. Such approaches will better facilitate meaningful meta-analyses of biological data.

Focus on integrative work across levels of biological organization

"Integrative biology is both an approach to and an attitude about the practice of science" (Wake, 2003, p. 240). The importance of integrative biology is widely recognized, yet the term has diverse interpretations. Integrative biology can mean a multidisciplinary approach that incorporates diverse fields of science or subdisciplines of biology or a multilevel approach that bridges different levels of biological organization (Wake, 2003).

Similarly, research can integrate across timescales or life-history stages (Wake, 2008; Lailvaux and Husak, 2017). The basic, essential need of organisms for oxygen means that to fully understand its effects on biological systems, oxygen effects must be approached integratively from multiple (sub)disciplines. This is particularly important since other major environmental drivers, such as temperature and CO₂, influence oxygen dynamics at all levels of organization (e.g., molecular, cell, organism, ecosystem).

Today, society seeks to understand the effects of climate change on emergent ecosystem properties such as fish biomass or production, biodiversity, or phenology. Several integrative indices and concepts have been proposed to address the effects that deoxygenation and warming have on these emergent properties. These include the metabolic index (Deutsch *et al.*, 2015), the aerobic growth index (Clarke *et al.*, 2021), and the oxygen and capacity-limited thermal tolerance model (Pörtner *et al.*, 2017) as well as varied ecosystem models. Application of most of these approaches requires process measurements on individual organisms, sometimes at different life stages or temperatures; thus, integration across levels of biological organization is inherent to addressing many scientific questions and societal needs (Pörtner, 2021). We suggest that there remains significant scope for developing even broader indices derived from more diverse perspectives.

The papers in this issue highlight, from diverse perspectives, the importance of oxygen interactions, including their effects on the function of oxygen transport proteins (Coates *et al.*, 2022), immune defense (Burnett and Burnett, 2022), organismal performance and survival (Lucey *et al.*, 2022), life-history optimization (Atkinson *et al.*, 2022; Audzijonyte *et al.*, 2022), and plastic and evolutionary responses over multiple timescales (Borges *et al.*, 2022; Earhart *et al.*, 2022; Roman and Pierson, 2022; Sperling *et al.*, 2022; Walczyńska and Sobczyk, 2022). Other factors less often considered, such as light, can also interact with oxygen availability to affect organismal performance in surprising ways (McCormick *et al.*, 2022).

Integrative biology comes with substantial logistical hurdles. Experiments that measure the physiological and energetic effects of oxygen interactions, particularly among the “deadly trio,” are essential to making general predictions about climate change effects at higher levels; yet, these types of integrative projects require substantially more time, effort, and resources than single-stressor science. Effectively working across levels of organization (e.g., molecular, cell, organism, ecosystem) requires both broad and deep expertise in different fields of science. In addressing ocean ecosystems, the Intergovernmental Panel on Climate Change (IPCC) is increasingly recognizing the importance of understanding combined effects of multiple stressors and integration of those effects across life stages, different levels of biological organization, and trophic levels (Cooley *et al.*, 2022). Our hope is that, as happened with carbon dioxide and ocean acidification in the beginning of the twenty-first century, the fundamental importance of

oxygen in driving biological responses to climate change will eventually inform multiple levels of management and policy.

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