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# Thresholds of hypoxia of two Red Sea coral species (*Porites* sp. and *Galaxea fascicularis*)

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Anthropogenic pressures have driven large-scale declines in coral cover on >50% of tropical coral reefs. Most research efforts have focused on ocean warming, ocean acidification, and overfishing impacts. Despite increasing instances of reef-associated hypoxic events, the role of reduced O<sub>2</sub> in affecting coral reef performance is largely unknown. Here, we assessed the hypoxic thresholds of two Red Sea coral species: *Porites* sp. and *Galaxea fascicularis*. We exposed coral fragments of both species to one control treatment (6.8 mg O<sub>2</sub> L<sup>-1</sup>) and three reduced dissolved oxygen treatments (5.25, 3.5, and 1.25 mg O<sub>2</sub> L<sup>-1</sup>) during a 10-day experiment. Across the two species, maximum (*F<sub>v</sub>/F<sub>m</sub>*) and effective (*F'<sub>v</sub>/F<sub>m</sub>'*) photochemical efficiency, chlorophyll *a*, and dark respiration declined under the lowest O<sub>2</sub> treatment (1.25 mg O<sub>2</sub> L<sup>-1</sup>). *Porites* sp. coral fragments, however, were remarkably resistant and showed no signs of sublethal bleaching after 10 days of exposure to reduced O<sub>2</sub>. Conversely, 17% of *G. fascicularis* fragments bleached after only three nights of exposure to the lowest O<sub>2</sub> treatment (1.25 mg O<sub>2</sub> L<sup>-1</sup>). Our data show that longer-term hypoxic events (i.e., days to weeks) can induce coral bleaching, but these effects depend on the extent of O<sub>2</sub> reduction and are likely species-specific. Importantly, the levels of O<sub>2</sub> usually defined as hypoxic (~2.0 to 2.8 mg O<sub>2</sub> L<sup>-1</sup>) do not adequately capture the thresholds reported here. Hence, further research is urgently needed to more accurately describe the vulnerability of coral taxa to hypoxic and anoxic events.

## KEYWORDS

coral reefs, dissolved oxygen, deoxygenation, O<sub>2</sub> depletion, exposure time

## 1 Introduction

Coral reefs are one of the most productive and biodiverse ecosystems on Earth (Connell, 1978; Moberg and Folke, 1999; Costanza et al., 2014). They play an essential role as ecological and economic resources, providing food, shelter, and habitat for many marine organisms (Hoegh-Guldberg et al., 2017). Coral reef ecosystems strongly depend on the complex interactions between Scleractinian corals and their photosynthetic dinoflagellates of the family Symbiodiniaceae spp. (LaJeunesse et al., 2018), as well as an array of microorganisms such as bacteria, archaea, fungi, and viruses (Knowlton and Rohwer, 2003; Marhaver et al., 2008; Bourne et al., 2016)—together this association is termed the coral holobiont. This beneficial relationship involves an exchange of photosynthetic end products (carbon and oxygen) from the Symbiodiniaceae to the coral host, which can provide the host with up to 90% of their daily energy requirement (Muscatine and Porter, 1977). Indeed, this association is essential for coral survival and growth but can rapidly deteriorate under environmental stress (Glynn, 1993; LaJeunesse et al., 2018). A common manifestation of this deterioration is coral bleaching, where the coral host exhibits a decrease in Symbiodiniaceae cells from their gastrodermal tissues and parallel reductions in photosynthetic pigments under prolonged environmental stress.

Global warming, ocean acidification, overfishing, and nutrient pollution are considered the main drivers of coral reef degradation (Hughes et al., 2003). However, recent research indicates that acute hypoxia events may be increasing in coral reef ecosystems (Altieri et al., 2017; Johnson et al., 2021a). Recently, reported hypoxia events (defined as  $O_2$  levels  $\leq 2$  mg  $O_2$   $L^{-1}$ ) on tropical reefs have caused mass coral bleaching and mortality (varying from 0% to 100% mortality, depending on the species; Altieri et al., 2017; Alderdice et al., 2021; Johnson et al., 2021a). Hypoxia events are driven by numerous, and sometimes interacting, drivers that include temperature increases (directly *via* decreases in the solubility of  $O_2$  and indirectly through heightened community respiration in response to elevated temperatures), eutrophication (*via* the breakdown of organic matter and resulting microbial  $O_2$  demand), and stratification of seawater surface (Keeling et al., 2010; Breitburg et al., 2018; Hughes et al., 2020; Klein et al., 2020), among others.

In coral reef ecosystems, extreme diel dissolved oxygen (DO) fluctuations have been reported to vary between 0 and 18 mg  $O_2$   $L^{-1}$  (DeCarlo et al., 2017). These changes in  $O_2$  depend on the location (and depth), time of the day, and water movement, owing to the differential influence of community metabolic production and consumption processes on  $O_2$  availability in the water medium (Al-Horani, 2005; Guadayol et al., 2014; Giomi et al., 2019; Nelson and Altieri, 2019). Within the coral holobiont, Symbiodiniaceae produce more  $O_2$  than the coral host can consume during the daytime, acting as a net  $O_2$  source. However, Symbiodiniaceae cease  $O_2$  production at nighttime

and rely on efficient  $O_2$  extraction from the surrounding seawater medium (Kühl et al., 1995). At nighttime, the holobiont actively consumes  $O_2$ , and this process presumably involves a competition between the holobiont partners for  $O_2$  when its supply is limited. Hence, corals may be particularly vulnerable to  $O_2$  deprivation at nighttime when productivity ceases, and holobiont respiration consumes oxygen. Indeed, reductions in DO availability can directly limit aerobic respiration and affect other coral holobiont processes such as photosynthesis and calcification (Yonge et al., 1928; Gardella and Edmunds, 1999; Al-Horani et al., 2007; Nelson and Altieri, 2019). Consequently, declines in DO can potentially modify the interactions between corals, Symbiodiniaceae, and their microbiota, leading to competition for  $O_2$  under hypoxic conditions between the otherwise partners.

Exposure time is another factor known to influence the biological responses of marine taxa to hypoxia. Steckbauer et al. (2020) found a positive trend between the effect of the exposure time and hypoxia impacts on the responses of marine taxa, suggesting that prolonged exposure to hypoxic conditions negatively impacts the survival of marine organisms. Despite the potential for, and observed, impacts of hypoxic events on corals, hypoxic thresholds of corals that consider exposure time remain poorly studied. Although scarce, the reported hypoxic thresholds differ substantially among coral species. For instance, coral bleaching events in Costa Rica and Panama caused widespread bleaching of *Pocillopora* spp., which suffered 90% to 100% mortality. On the other hand, *Porites lobata*, *Pavona clavus*, *Pavona gigantea*, and *Gardinoseris planulata* experienced no mortality after the same event (Guzmán et al., 1990; Nelson and Altieri, 2019). Haas et al. (2014) reported a lethal threshold of 2 to 4 mg  $O_2$   $L^{-1}$  in *Acropora yongei* by analyzing the effects of hypoxia at nighttime. Another experimental study done by Altieri et al. (2017) found that *Stephanocoenia intersepta* were highly resistant to hypoxia, even after being exposed to hypoxic seawater (0.5 mg  $O_2$   $L^{-1}$ ) for 1 week. In most instances, published experiments that test the hypoxic effects on corals typically apply lower temperature regimes and short-term exposure. However, thresholds of hypoxia are not static and increase with warming (Matthews and McMahon, 1999; Vaquer-Sunyer and Duarte, 2008). Discounting the potential role of thermal regimes may lead to underestimating coral tolerance. In addition to the lower temperature regimes, another research gap in the published studies lies in the manipulation of experimental  $O_2$  levels, where  $N_2$  gas is used to reduce  $O_2$ . However, this approach inadvertently displaces  $CO_2$  and increases seawater pH (Klein et al., 2017), thereby affecting the carbonate system and confounding any observed effects.

Here, we aimed to examine the response of two abundant Red Sea coral species—*Porites* sp. and *G. fascicularis* corals—to reduced DO concentrations and characterized their hypoxic thresholds. We hypothesized that the prolonged exposure of

corals to reduced DO concentrations could trigger coral bleaching and eventual mortality beyond an  $O_2$  threshold where  $O_2$  availability is insufficient to maintain the symbiotic relationship between corals and their symbionts. We hypothesized that corals would exhibit a decrease in both maximum ( $F_v/F_m$ ) and effective ( $F'/F_m'$ ) photochemical efficiency, lower respiration rates, symbiont counts, and chlorophyll *a* (Chl *a*) content under two decreased  $O_2$  treatments (3.5 and 1.25 mg  $O_2$  L<sup>-1</sup>). We further hypothesized that the corals would exhibit different phenotypes of coral health degradation, such as coral bleaching/paling, rapid tissue disintegration (also termed “melting phenomenon”), or even mortality at low DO (~1.25 mg  $O_2$  L<sup>-1</sup>) concentrations.

## 2 Materials and methods

### 2.1 Coral collection

We identified and collected three adult colonies of both *Porites* sp. and *G. fascicularis* off the coast of Thuwal in the Central Red Sea, Saudi Arabia (22°18'12.4"N, 38°57'38.3"E) at depths between 8 and 12 m in early June of 2021. To minimize the likelihood of collecting identical genotypes, we sampled colonies separated by at least 5 m (Rädecker et al., 2017). *Porites* sp. is a reef-building coral easily distinguished by its surface morphology (polyp diameter of ~1 mm) and corallite skeletal characteristics (Smith et al., 2007). *G. fascicularis* is also a scleractinian coral, characterized by larger polyp diameters of, on average, 1 cm (Veron, 1980).

After collection, the colonies were immediately transported to the experimental laboratory facilities of the Coastal and Marine Resources Lab (CMR) at King Abdullah University of Science and Technology (KAUST). We fragmented the colonies into 156 coral fragments per species (~2 cm width) and allowed them to recover for 2 days at 28°C under a 12:12 light–dark cycle [350  $\mu$ mol photons m<sup>2</sup> s<sup>-1</sup> of photosynthetic active radiation (PAR)]. Then, the temperature was increased to 32°C over 7 days, at a rate of 0.5°C per day, to mimic summer temperatures in the Central Red Sea (Chaidez et al., 2017; Osman et al., 2018; Genevier et al., 2019). The fragments were kept in a continuous flow-through tank during the recovery period, where fresh filtered seawater (20  $\mu$ m pore size) was supplied from the Red Sea.

### 2.2 Experimental design

For the two species, we conducted two separate but identical 10-day experiments. The experimental design for both species included a stable DO control treatment (6.8 mg  $O_2$  L<sup>-1</sup>) and three reduced  $O_2$  treatments (5.25, 3.5, and 1.25 mg  $O_2$  L<sup>-1</sup>), hereafter termed high, medium, and low  $O_2$  treatments,

respectively. We selected these DO concentrations according to normoxic and hypoxic levels reported by the meta-analysis of Steckbauer et al. (2020). For both coral species, pH ( $8.2 \pm 0.07$ SD) levels, temperature ( $32^\circ\text{C} \pm 0.22$ SD), salinity (~44 ppt), and PAR (350  $\mu$ mol photons m<sup>-2</sup> s<sup>-1</sup>) were held constant in a 12:12 light:dark diel cycle throughout all four treatments and represented conditions in the field at the time of coral collection. Each experiment evaluated 48 coral fragments, which we randomly distributed among the four DO treatments ( $n = 12$ ).

Prior to the commencement of each experiment, coral fragments were acclimated to the experimental system. We used four 600-L tanks as water baths to control the temperature inside each 2-L aquarium. The acclimation day started at 5:00 a.m. by placing one coral fragment per 2-L of glass aquarium; to minimize potential effects associated with individual water baths, we randomly distributed the 2-L aquaria across four water baths. Each aquarium was equipped with one standard aquarium bubbler and one transparent plastic lid to prevent seawater evaporation and minimize the influence of air/seawater gas exchange. Next, we started the experimental gas treatments by introducing each coral fragment into the control conditions (6.8 mg  $O_2$  L<sup>-1</sup>  $\pm$  0.38SD,  $32^\circ\text{C} \pm 0.22$ SD, and pH  $8.2 \pm 0.07$ SD). To transition the corals from acclimation conditions to our defined control and reduced DO treatments, we used an 8-h  $O_2$  drift based on hypoxic diel cycle duration reported by Tyler et al. (2009). The transition started at 10:00 p.m. on the acclimation day and ended at 6:00 a.m. on the next day (experiment day 1). Our control treatment remained stable throughout the  $O_2$  drift at 6.8 mg  $O_2$  L<sup>-1</sup>.

On the other hand, for the rest of the treatments, the  $O_2$  concentration decreased as follows: the high treatment decreased 0.19 mg  $O_2$  h<sup>-1</sup> until reaching a final concentration of 5.25 mg  $O_2$  L<sup>-1</sup>, the medium treatment decreased 0.41 mg  $O_2$  h<sup>-1</sup> until it reached 3.5 mg  $O_2$  L<sup>-1</sup>, and the low treatment decreased 0.70 mg  $O_2$  h<sup>-1</sup> until reaching a final concentration of 1.25 mg  $O_2$  L<sup>-1</sup> (Figure S4). Throughout the experiments, we exchanged 100% of the seawater in each aquarium at 6:00 a.m. each day.

### 2.3 Gas manipulation

To customize the gas treatments, we used a modified version of the LabView software (*sensu* Klein et al., 2017). We delivered the gas treatments *via* four sets of three mass flow controllers (MFCs) [ $\Omega$ ,  $N_2$  (0 to 2 L min<sup>-1</sup>),  $CO_2$  (0 to 20 ml min<sup>-1</sup>), and  $O_2$  (0 to 2 L min<sup>-1</sup>)]. We connected the MFCs to pure  $O_2$  and  $CO_2$  cylinders (Air Liquide, Jeddah) and liquid  $N_2$  dewars equipped with vaporizers, which we used to deliver pure  $N_2$  gas at high flow rates. The four unique gas mixtures were continuously supplied to the 2-L aquaria *via* stainless steel manifolds connected to tubing equipped with aquarium air stones. This approach permitted the equilibration of the

seawater to the desired treatment levels. We manually controlled the gas flow rates to the individual aquaria with standard five-way aquarium manifolds. We measured temperature, pH, and DO levels every day using a Mettler Toledo sensor (at 4 a.m. and 2:00 p.m.) (Table S1).

## 2.4 Analysis of carbonate chemistry

To characterize the seawater carbonate chemistry, we collected 120 ml of seawater samples for total alkalinity (TA) analysis on days 1, 3, and 10 of each experiment. We randomly selected three replicate aquaria from each of the four treatments for TA analysis. The samples were collected in borosilicate glass bottles through a drawing tube to prevent the formation of bubbles and immediately preserved the seawater with 50  $\mu$ l of mercuric chloride  $\text{HgCl}_2$  (5%). The samples were then stored at 23°C and analyzed within 30 days of the conclusion of the experiments. We measured TA *via* an open-cell titration method with 0.1 M of hydrochloric acid, using an Alkalinity Titrator (MODEL AS-ALK2, Apollo SciTech, U.S.A, Kloehe - U.S.A). The titrator was equipped with two pumps (VersaPump 6, Kloehe), a pH meter (Orion Star A2II, Thermo Fisher, U.S.A.), and a pH probe (SI Analytics, Germany), following the method retrieved from Dickson et al. (2007). We then calculated the partial pressure of  $\text{CO}_2$  ( $p\text{CO}_2$ ) from measured values of TA, pH, temperature, and salinity values using the “carb” and “errors” functions in the seacarb package 3.3.0v in R software, obtained from Lavigne and Gattuso (2011).

## 2.5 Mortality and coral bleaching

When the coral fragments exhibited any of the following signs—there was evident tissue degradation, polyps showed either discoloration or appeared withdrawn, or when substantial reductions in pigment were visible—they were recorded as dead (Anton et al., 2020). Visual observations were conducted immediately after simulated sunrise and sunset (i.e., twice per day).

## 2.6 Physiological response variables

### 2.6.1 Dark respiration ( $\text{O}_2$ flux)

Rates of dark respiration (DR) were measured at the end of each experiment *via*  $\text{O}_2$  flux incubations. For this, we used a FireSting- $\text{O}_2$  (PyroScience, Germany) connected to four fiber-optic sensors that measured DO concentrations every 20 s. We calibrated the sensors at 32°C using a two-point calibration (0% and 100%  $\text{O}_2$  saturation). On day 10 of each experiment, we conducted  $\text{O}_2$  flux incubations in two rounds. Each round consisted of six coral fragments and five seawater blanks. The

water used for each measurement was seawater taken directly from the same aquarium as the sampled coral fragment (i.e., seawater premanipulated to conditions of the appropriate treatment). Each round measured fragments (and seawater blanks) from all four treatments so as not to confound the measurement rounds with the treatments examined. We randomized the sample selection to ensure we analyzed the same number of fragments per treatment ( $n = 3$ ) per set. We used chambers with a volume of 50 ml, which we equipped with internal stir bars. We used waterproof paper ( $\sim 4 \text{ cm}^2$  in size) as an interior shelf to separate the coral fragment from the magnetic stirrer, which allowed the seawater to mix and maintain homogeneous  $\text{O}_2$  levels within the chamber. In total darkness, we used an IKA RT 15 magnetic stir plate to control the stir bars and keep the temperature at 32°C inside a thermally controlled growth chamber (Percival Scientific Inc.). For *Porites* sp., incubations lasted 2.5 h, while for *G. fascicularis*, incubations lasted 1 h. Finally, we calculated the DR rates for each coral fragment by subtracting the blank DR rate from the sample DR rate and dividing it by the superficial area of each fragment ( $\text{mg O}_2 \text{ cm}^{-2} \text{ h}^{-1}$ ).

### 2.6.2 Maximum ( $F_v/F_m$ ) and effective photochemical efficiency ( $F'/F_m'$ )

On each day of the experiments, we measured dark-adapted maximum photochemical efficiency ( $F_v/F_m$ ) and effective photochemical efficiency ( $F'/F_m'$ ) of photosystem II (PSII) in all aquaria. We did the measurements at 04:00 a.m. and 2:00 p.m. local time, respectively, using a MINI-PAM (Walz, Germany). To avoid potential artifacts associated with the timing of measurements, we changed the order in which the aquaria were measured. For *G. fascicularis*, we conducted three technical measures per fragment to reduce variability in the values taken. We used the average (mean) values of the technical measurements for statistical analysis, which we considered the statistical unit/biological replicate. However, due to the small size of *Porites* sp., we conducted only one technical replicate, which we used for data analysis.

### 2.6.3 Symbiodiniaceae cell density and chlorophyll *a*

After the  $\text{O}_2$  flux incubations, we froze and stored all coral fragments at  $-20^\circ\text{C}$ . Before analysis, the fragments were thawed at room temperature ( $\sim 23^\circ\text{C}$ ) and incubated in 3 ml of sodium dodecyl sulfate (SDS) 0.1% solution for 30 min. We then removed the coral tissue from each fragment by blasting them with air (using a 1,000  $\mu$ l pipette tip connected to tubing and benchtop air supply) in a fume hood. We transferred the resulting slurry to 15-ml Falcon tubes (Fisherbrand, U. S. A) and homogenized the sample with a tissue homogenizer (MicroDisTec homogenizer 125, Thermo Fisher Scientific) for 30 s. Next, we transferred two 500- $\mu$ l aliquots of the slurry (one for Symbiodiniaceae cell counting and one for chlorophyll *a* to

1.5 ml Eppendorf tubes). The aliquots allocated to Symbiodiniaceae cell counts were resuspended after centrifugation at 16,000g for 15 min. Then, we discarded the supernatant (500  $\mu$ l) and washed the remaining Symbiodiniaceae pellet using 1 $\times$  phosphate buffer solution (PBS, 500  $\mu$ l), and we centrifuged it for 5 min at 16,000g. After the centrifugation, we discarded the supernatant and repeated the process with Milli-Q water. Then, we resuspended the remaining pellet using 250  $\mu$ l of PBS and 250  $\mu$ l of SDS. Finally, the Symbiodiniaceae cells were counted in three replicate aliquots of 10  $\mu$ l, each using a hemocytometer (Hausser Scientific, U. S. A.) under an upright fluorescence microscope (Leica DM6000 B running LAS X).

The 500- $\mu$ l aliquots of the coral slurry allocated to measurements of Chl *a* were centrifuged (3,000g at 4°C) for 10 min. After centrifugation, the supernatant was discarded and the pellet was resuspended in 500  $\mu$ l of ethanol 100%. The samples were incubated overnight at 4°C in total darkness extraction. The samples were then centrifuged at 16,000g and 4°C for 5 min. Finally, we transferred an aliquot of 150  $\mu$ l of each sample (in triplicates) into a 96-well flat-bottom plate; ethanol 100% was used as a blank. We measured absorbance at 629 and 665 nm using a SpectraMax Paradigm microplate reader (Molecular Devices, U. S. A.). Finally, total Chl *a* concentrations were obtained using the following equation ( $-2.6094 \times A_{629} + 12.4380 \times A_{665}$ ) for dinoflagellates in ethanol (Ritchie, 2006).

Counts of Symbiodiniaceae cells and concentrations of Chl *a* were both normalized to coral fragment surface area (cm<sup>2</sup>).

## 2.7 Surface area

To calculate the superficial area of each fragment, we used the aluminum foil method (Marsh, 1970). First, we calculated a conversion factor of  $3.82 \times 10^{-3}$  g cm<sup>-2</sup> by weighing six 10-cm<sup>2</sup> aluminum foil squares. Then, each sample was wrapped in aluminum foil three times (i.e., three technical replicates), covering the entire surface of the fragment while carefully avoiding overlap. The foil was then removed, weighed (to the nearest milligrams), and then averaged. Finally, we calculated the superficial for each sample using the conversion factor previously obtained.

## 2.8 Statistical analysis

We analyzed the response variables of maximum ( $F_v/F_m$ ) and effective ( $F'/F_m'$ ) photochemical efficiency, O<sub>2</sub> flux, symbiont density, and Chl *a* using the linear mixed models (LMMs) in SPSS (IBM SPSS, version 28, U. S. A.). Before starting the analyses, we checked all data for the assumptions of normality and homoscedasticity by using Q-Q plots of the

residuals and standardized residual plots. For maximum ( $F_v/F_m$ ) and effective ( $F'/F_m'$ ), we considered all factors (species and O<sub>2</sub> treatment) except time as fixed factors; all factors were fixed for the rest of the dependent variables. In addition, we considered colony number as a random factor for all dependent variables. Preliminary analyses for all dependent variables revealed significant effects of colony number, so it was retained in further analyses. In all repeated-measure LMMs, we analyzed different models [e.g., CS, AR (1), and AR (1) heterogeneous] to assess the model that best fitted by comparing various goodness-of-fit statistics [e.g., -2 restricted log-likelihood, Akaike's information criteria (AIC), and Bayesian information criterion (BIC)]. To quantify the sublethal exposure time (SLT<sub>50</sub>), we used a two-parameter log-logistic dose-response model from the drc package in R (Ritz et al., 2015). We conducted all sublethal threshold analyses in R Studio (version 3.6.2). Data are reported as mean  $\pm$  one standard error (1SE) unless otherwise indicated (e.g., standard deviation as 1SD).

## 3 Results

### 3.1 Mortality and coral bleaching

We observed no mortality in either species during the experiments. However, after the third night of exposure to the low O<sub>2</sub> treatment, we observed color loss in 17% of the *G. fascicularis* fragments, and by the end of the experiment, 58% of the fragments had bleached. In contrast, no *Porites* sp. fragments bleached under the same O<sub>2</sub> treatment. Across both experiments/species, no signs of bleaching were observed for any other O<sub>2</sub> treatments (Figure 1).

Unfortunately, logistical constraints associated with the sacrificial nature of some of the measurements limited our ability to examine more replicates across a higher number of O<sub>2</sub> treatments; hence, the statistical power for our SLC<sub>50</sub> calculation was admittedly low ( $n = 4$ ). Therefore, we calculated the median sublethal exposure time (SLT<sub>50</sub>) under the low O<sub>2</sub> treatment at  $5.15 \pm 0.80$ SE days at 32°C (Figure 2), which is supported by greater statistical power ( $n = 10$ ).

### 3.2 Physiological response variables

#### 3.2.1 Dark respiration (O<sub>2</sub> flux)

DR rates differed among the two species ( $p = 0.009$ , Table 1) [i.e., *G. fascicularis* were 88.6% (control treatment), 216.0% (high treatment), 25.8% (medium treatment), and 32.4% (low treatment) higher than *Porites* sp.]. The rates of DR also differed significantly among O<sub>2</sub> treatments ( $p = 0.031$ ), regardless of the species examined. For example, DR rates of



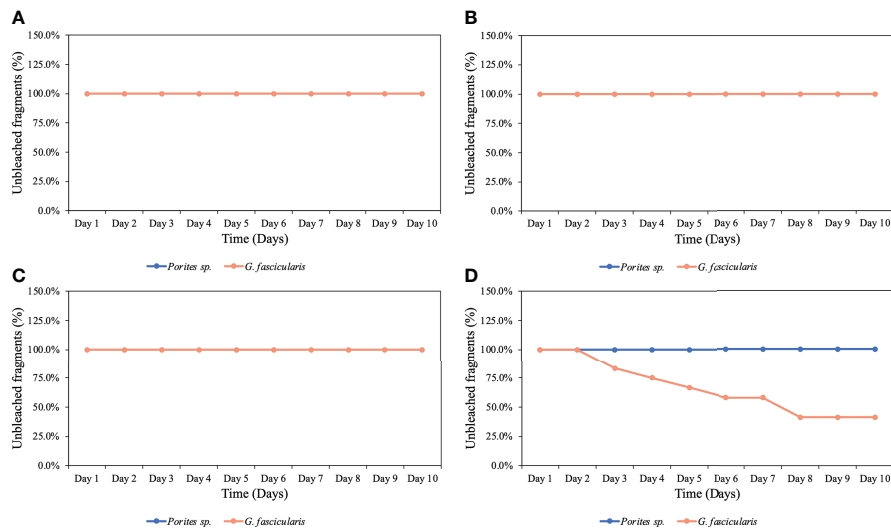


FIGURE 1

Percentage of unbleached coral fragments throughout the complete experiments for both species. (A) Control ( $6.8 \text{ mg O}_2 \text{ L}^{-1}$ ), (B) high  $\text{O}_2$  treatment ( $5.25 \text{ mg O}_2 \text{ L}^{-1}$ ), (C) medium  $\text{O}_2$  treatment ( $3.5 \text{ mg O}_2 \text{ L}^{-1}$ ), and (D) low  $\text{O}_2$  treatment ( $1.25 \text{ mg O}_2 \text{ L}^{-1}$ ). *Porites sp.* did not experience any bleaching at any of the four different treatments; on the other hand, *G. fascicularis* started experiencing bleaching after the third night of exposure to the low  $\text{O}_2$  treatment.

*G. fascicularis* were 9.3% higher in the control treatment than the high DO treatment and 170.2% higher in the control treatment relative to the low DO treatment. On the other hand, DR rates of *Porites sp.* were higher ( $0.12 \text{ mg O}_2 \text{ cm}^{-2} \text{ h}^{-1}$ ) in the control treatment, but the rates declined by 45.4%, 38.9%, and 47.3% in the high, medium, and low DO treatments (Figure 3). Taken together, these results show sharp reductions in DR rates in the lower DO treatments for both species relative to those in the control, indicating oxygen-conforming behavior.

### 3.2.2 Maximum ( $F_v/F_m$ ) and effective photochemical efficiency ( $F'/F_m'$ )

We found a significant interaction between species and time for both maximum and effective photochemical efficiency ( $p < 0.001$ , Tables 2, 3), indicating that the  $F_v/F_m$  and  $F'/F_m'$  values differed among species, but these differences depended on exposure time. For instance, *Porites sp.* coral fragments exhibited lower  $F_v/F_m$  values at the beginning of the experiment ( $0.492 \pm 0.010\text{SE}$ ) regardless of the treatment, which then

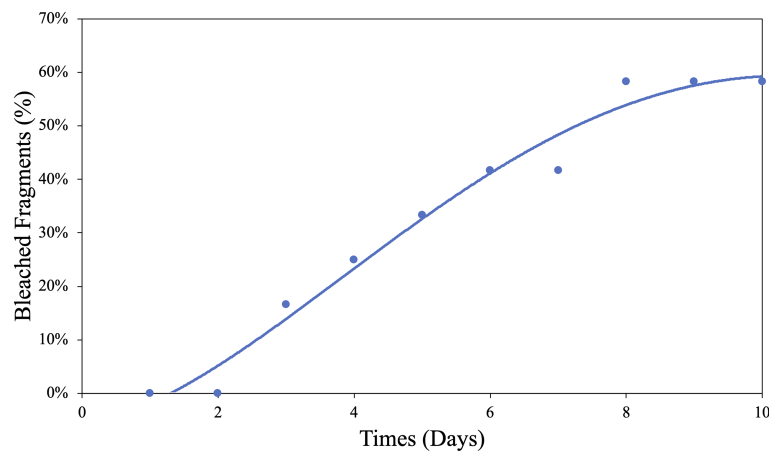


FIGURE 2

Hypoxic bleaching threshold determination for *G. fascicularis*. The median sublethal bleaching time for *G. fascicularis*, the  $\text{SLT}_{50}$ , was estimated at  $5.15 \pm 0.80\text{SE}$  days at  $32^\circ\text{C}$ .

TABLE 1 Summary of results for LMMs comparing dark respiration rates between treatments for both *Porites* sp. and *G. fascicularis*.

Source	Numerator <i>df</i>	Denominator <i>df</i>	<i>F</i>	<b>Sig.</b>
Species	1	15.019	9.112	<b>0.009</b>
O <sub>2</sub> treatment	3	15.026	3.878	<b>0.031</b>
Species * O <sub>2</sub> treatment	3	15.026	2.039	0.152

p-values in bold are statistically significant ( $p < 0.05$ ). BIC (Bayesian information criterion) = 188.043 and AIC (Akaike's information criterion) = 186.498. \*Interaction between species and O<sub>2</sub> treatment.

increased by 33.8% on day 10. On the other hand, *G. fascicularis*  $F_v/F_m$  values exhibited a decreasing trend throughout the experiment (regardless of the treatment) starting at  $0.719 \pm 0.008\text{SE}$  (day 1) and ending at  $0.644 \pm 0.007\text{SE}$  (day 10) (Figure 4). *Porites* sp. and *G. fascicularis*  $F'/F_m'$  showed inconsistent patterns throughout the experiment, regardless of the treatment (Figure 5).

We further detected a significant interaction between species and O<sub>2</sub> treatments for both  $F_v/F_m$  and  $F'/F_m'$  indicating that differences among species were inconsistent among the O<sub>2</sub> levels tested. For  $F_v/F_m$  ( $p < 0.001$ , Table 2) and  $F'/F_m'$  ( $p = 0.023$ , Table 3), values in the low O<sub>2</sub> treatment were reduced relative to all other treatments across both species (Figures 4, 5).

There was also a significant interaction between time and O<sub>2</sub> treatments for both maximum and effective photochemical efficiency suggesting that differences among O<sub>2</sub> treatments were influenced by exposure time regardless of the species analyzed. That is, for  $F_v/F_m$  ( $p = 0.009$ , Table 2) on day 1, values in all treatments were around  $0.605 \pm 0.004\text{SE}$ , increasing to  $0.680 \pm 0.009\text{SE}$  by day 10 in the control, high, and medium treatments. However, values in the low treatment decreased to  $0.561 \pm 0.017\text{SE}$  regardless of the species (Figure 4). On the other

hand, for  $F'/F_m'$  ( $p = 0.044$ , Table 3), all treatments had similar values throughout the experiment, except for the low O<sub>2</sub> treatment which exhibited 14.4% reduction by the end of the experiment (Figure 5). We calculated the SLT<sub>50</sub> for both species in the low O<sub>2</sub> treatment at 32°C. For *Porites* sp., we calculated the SLT<sub>50</sub> at  $5.23 \pm 3.81\text{SE}$  ( $F_v/F_m$ ) and  $6.54 \pm 3.55\text{SE}$  ( $F'/F_m'$ ) days; on the other hand, for *G. fascicularis*, we obtained SLT<sub>50</sub> at  $3.22 \pm 1.65\text{SE}$  ( $F_v/F_m$ ) and  $5.43 \pm 2.43\text{SE}$  ( $F'/F_m'$ ) days.

### 3.2.3 Symbiodiniaceae cell density

We found a significant difference in the symbiont density between species ( $p < 0.001$ , Table 4), which indicates that cell density differed between the two species regardless of the O<sub>2</sub> treatments. The cell density for *G. fascicularis* fragments was one order of magnitude higher than for *Porites* sp. fragments. In addition, the cell density also differed between O<sub>2</sub> treatments regardless of species ( $p = 0.035$ , Table 4). For example, the cell density of *G. fascicularis* in the control treatment was 34.5% and 71.4% higher when compared with the medium and low O<sub>2</sub> treatments; however, when compared with the high treatment, it was 18.5% lower. On the other hand, *Porites* sp. fragments had a greater cell density in the control treatment by 104.6% than in the low O<sub>2</sub> treatment; however, it was lower by 1.8% and 36.3% when compared with the high and medium treatments, respectively. Both species exhibited the lowest cell density in the low O<sub>2</sub> treatment, which suggests that symbionts were experiencing stress due to the reduced DO concentrations (Figure 6).

### 3.2.4 Chlorophyll *a*

We found a significant interaction between species and O<sub>2</sub> treatments ( $p = 0.028$ , Table 5), indicating that differences in Chl *a* content between the two species were inconsistent among the

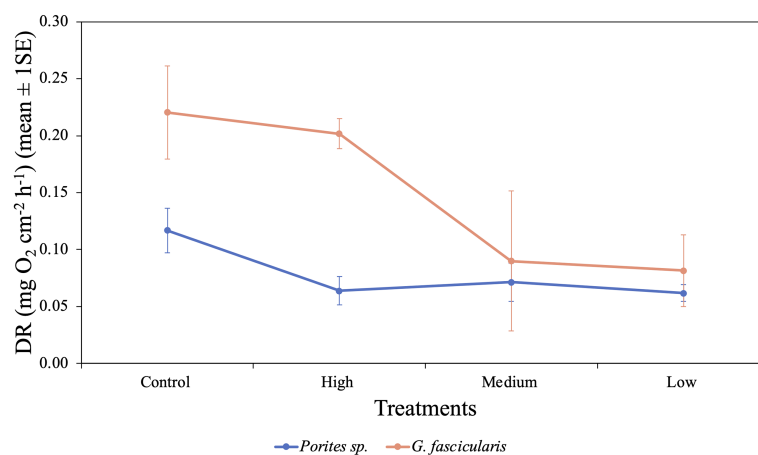


FIGURE 3

Mean ( $\pm$  1SE) rates of dark respiration ( $\text{mg O}_2 \text{ cm}^{-2} \text{ h}^{-1}$ ) for *Porites* sp. and *G. fascicularis* under four different O<sub>2</sub> treatments.

TABLE 2 Summary of results for LMMs comparing maximum  $F_v/F_m$  reading between treatments throughout the experiment.

Source	Numerator <i>df</i>	Denominator <i>df</i>	<i>F</i>	Sig.
Species	1	84.586	287.227	<0.001
Time	9	143.544	8.259	<0.001
O <sub>2</sub> treatment	3	84.584	26.714	<0.001
Species * time	9	143.544	20.812	<0.001
Species * O <sub>2</sub> treatment	3	84.584	4.144	<b>0.009</b>
Time * O <sub>2</sub> treatment	27	143.517	1.979	<b>0.006</b>
Species * time * O <sub>2</sub> treatment	27	143.517	1.074	0.378

The model of best fit was compound symmetry (CS), BIC (Bayesian information criterion) = -1,823.852, and AIC (Akaike's information criterion) = -1,838.126. *df* = degrees of freedom (numerator, denominator). *p*-values in bold are statistically significant ( $p < 0.05$ ). \*Interaction between species and O<sub>2</sub> treatment.

O<sub>2</sub> treatments. For instance, *Porites* sp. fragments under the low O<sub>2</sub> treatment exhibited Chl *a* contents 56.6%, 54.4%, and 58.7% lower than fragments in the control, high, and medium treatments, respectively. However, in *G. fascicularis* fragments, Chl *a* content was higher in the control and high O<sub>2</sub> treatments, relative to the medium and low O<sub>2</sub> treatments (Figure 7).

## 4 Discussion

Our findings support the hypothesis that prolonged exposure to hypoxic conditions negatively affects coral physiological performance in the Red Sea and may induce bleaching. Our experiment results showed differences in the physiological responses among the two species tested, with *Porites* sp. showing higher resistance to the reduced DO treatments tested. Overall, these findings are consistent with previous observations, where massive corals are considered to be more resistant to hypoxic events (Guzmán et al., 1990; Laboy-Nieves et al., 2001). Our results are consistent with the *in-situ* observations of Laboy-Nieves et al. (2001), where *Porites* spp. corals from the Morrocoy National Park in Venezuela survived a mass mortality event due to abnormal levels in several physicochemical parameters, including reduced concentrations of DO (<2.3 mg O<sub>2</sub> L<sup>-1</sup>). Similar results were observed by Fine

et al. (2013) and Furby et al. (2013), where *Porites* spp. experienced a lower percentage of bleaching compared with other branching species such as *Acropora*, *Pocillopora*, and *Stylophora*, in the 2010 Red Sea mass bleaching event. However, since DO was not measured in either of these two studies, and the temperature effects were not analyzed in the present study, we can only suggest that hypoxic effects may have influenced the results observed in the 2010 Red Sea bleaching event. Therefore, future experiments should analyze the interaction between increased temperature and decreased DO.

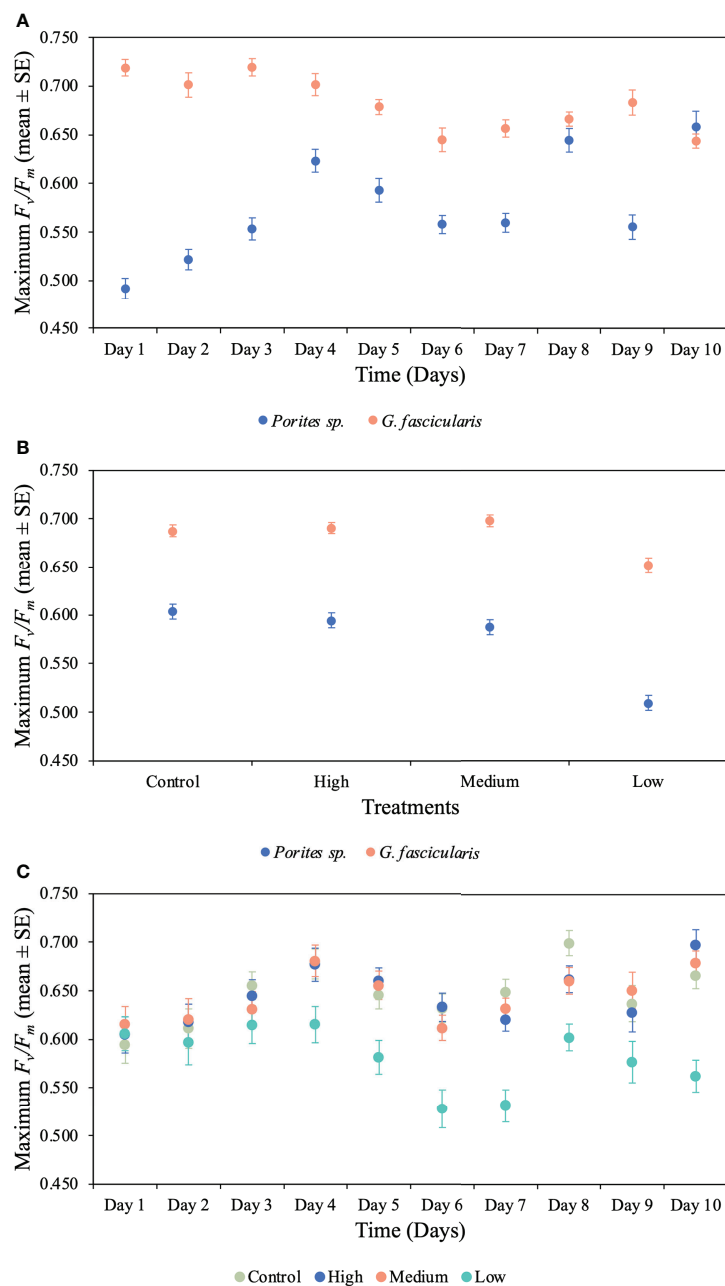
Contrary to the apparent resistance of *Porites* sp., our results showed hypoxia-induced bleaching for 58% of the *G. fascicularis* fragments under the low O<sub>2</sub> treatment by the end of the experiment. Similar results were observed by Furby et al. (2013) and Monroe et al. (2018), where the genus *Galaxea* was one of the most impacted by the 2010 Red Sea bleaching event, aligning with previous findings implicating a potential role of O<sub>2</sub> deprivation in coral bleaching tolerance (Alderdice et al., 2021), although future experiments are required to test this hypothesis. In the present study, the harmful effects for *Porites* sp. were not observed even after 10 days of exposure to the different reduced O<sub>2</sub> treatments. However, *G. fascicularis* started bleaching after the third night of the experiment. We further calculated the median bleaching time (SLT<sub>50</sub>) for *G. fascicularis* of 5.15 ± 0.80SE days under our low O<sub>2</sub> treatment at 32°C. To

TABLE 3 Summary of results for LMMs comparing effective  $F'/F_m'$  reading between treatments throughout the experiment.

Source	Numerator <i>df</i>	Denominator <i>df</i>	<i>F</i>	Sig.
Species	1	89.869	90.002	<0.001
Time	3	89.868	8.936	<0.001
O <sub>2</sub> treatment	8	126.161	8.462	<0.001
Species * time	8	126.161	9.765	<0.001
Species * O <sub>2</sub> treatment	3	89.868	3.334	<b>0.023</b>
Time * O <sub>2</sub> treatment	24	126.161	1.631	<b>0.044</b>
Species * time * O <sub>2</sub> treatment	24	126.161	0.944	0.544

The model of best fit was compound symmetry (CS), BIC (Bayesian information criterion) = -1,821.382, AIC (Akaike's information criterion) = -1,835.360. *df* = degrees of freedom (numerator, denominator). *p*-values in bold are statistically significant ( $p < 0.05$ ). \*Interaction between species and O<sub>2</sub> treatment.





**FIGURE 4**  
 Mean ( $\pm$  1SE) rates of maximum photochemical efficiency ( $F_v/F_m$ ) and its interaction between **(A)** species and time for *Porites sp.* and *G. fascicularis* throughout the 10 days of the experiment ( $p < 0.001$ ), **(B)** species and  $O_2$  treatment for *Porites sp.* and *G. fascicularis* under all four different  $O_2$  treatments ( $p = 0.009$ ), and **(C)** time and  $O_2$  treatment for *Porites sp.* and *G. fascicularis* throughout the 10 days of the experiment ( $p = 0.006$ ).

this day, and according to our knowledge, this is the first hypoxic bleaching threshold yet reported for *G. fascicularis* or Red Sea corals.

Based on historical bleaching events in the Red Sea, it has been suggested that, although *G. fascicularis* is considered to be relatively resilient, acute environmental stress can still disrupt its

symbiotic relationship with Symbiodiniaceae (Brown et al., 1995; Hou et al., 2018). Our results suggest that exposure time plays a critical role in disrupting this symbiosis, experiencing more severe repercussions when corals are exposed to extended periods of severe hypoxia. Given these results, we hypothesize that previous research testing short exposure times (hours to a

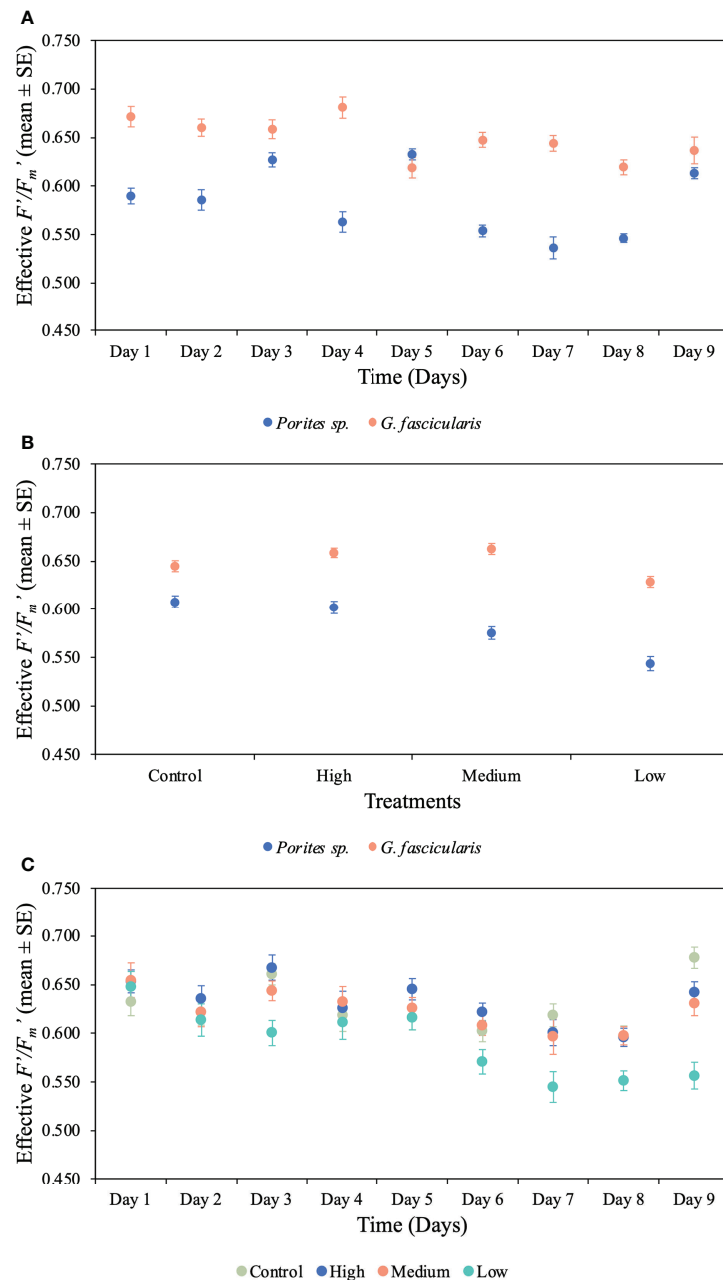


FIGURE 5

Mean ( $\pm$  1SE) rates of effective photochemical efficiency ( $F'/F_m'$ ) and its interaction between (A) species and time for *Porites sp.* and *G. fascicularis* throughout the 10 days of the experiment ( $p < 0.001$ ), (B) species and  $O_2$  treatment for *Porites sp.* and *G. fascicularis* under all four different  $O_2$  treatments ( $p = 0.023$ ), and (C) time and  $O_2$  treatment for *Porites sp.* and *G. fascicularis* throughout the 10 days of the experiment ( $p = 0.044$ ).

few days) may be underestimating the resistance of coral species to hypoxia (Table 6). Therefore, we urge the community to research not only short-term hypoxic events (diel cycles) but also long-term hypoxic events.

Corals from both species, *Porites sp.* and *G. fascicularis*, consistently exhibited reduced DR rates under the medium and

low  $O_2$  treatments, which indicates that the corals are capable of metabolic regulation as an adaptive mechanism to resist hypoxia. Given our results, we hypothesize that DO availability directly influences respiration by increasing respiration rates under high concentrations of  $O_2$  and decreasing when found in low  $O_2$  concentrations. Osinga et al.

TABLE 4 Summary of results for LMMs comparing Symbiodiniaceae cell densities between treatments for both species.

Source	Numerator <i>df</i>	Denominator <i>df</i>	<i>F</i>	Sig.
Species	1	15.305	97.051	<0.001
O <sub>2</sub> treatment	3	15.265	3.715	<b>0.035</b>
Species * O <sub>2</sub> treatment	3	15.265	3.189	0.054

*p*-values in bold are statistically significant ( $p < 0.05$ ). BIC (Bayesian information criterion) = 422.873 and AIC (Akaike's information criterion) = 421.328.

\* Interaction between species and O<sub>2</sub> treatment.

(2017) reported similar results for *G. fascicularis* DR rates, where under hypoxic conditions ( $\sim 1.62$  mg O<sub>2</sub> L<sup>-1</sup>) respiration was lower than in the normoxic ( $\sim 8.11$  mg O<sub>2</sub> L<sup>-1</sup>) and hyperoxic ( $\sim 12.17$  mg O<sub>2</sub> L<sup>-1</sup>) treatments. Similar results were reported by Gardella and Edmunds (1999), where respiration in symbionts isolated from *Dichocoenia stokesii* in their normoxic treatment (7.53 to 7.84 mg O<sub>2</sub> L<sup>-1</sup>) respired significantly more than those isolated in the hypoxic treatment (3.84 to 4.15 mg O<sub>2</sub> L<sup>-1</sup>). In addition, DR rates differed substantially between species, with *G. fascicularis* supporting greater respiration rates than *Porites* sp. in both control and high O<sub>2</sub> treatments. This divergence in respiration rates between species could be due to the differences in polyp size, also reflected in a higher symbiont load and Chl *a* content from *G. fascicularis*.

Our analysis showed that the interaction between time, O<sub>2</sub> treatment, and species significantly affected the maximum photochemical efficiency ( $F_v/F_m$ ), showing the lowest values of ( $F_v/F_m$ ) in the low O<sub>2</sub> treatment by day 10 for both species. A decline in photochemical efficiency is often used as an indicator of stress because it is considered to be a precursor of coral bleaching due to damage in the photosystems of the symbionts (Jones et al., 1998). Hence, our results suggest that corals in the low O<sub>2</sub> treatment experienced damage in their symbionts'

photosynthetic capacity due to prolonged exposure to severe hypoxic conditions. This finding is also supported by Haas et al. (2014), where the maximum ( $F_v/F_m$ ) measurements for the *A. yongei* coral fragments under the hypoxic treatment (2 to 4 mg O<sub>2</sub> L<sup>-1</sup>) declined until they reached  $\sim 70\%$  of their initial values after the third night of the experiment. Similar results were obtained by Ulstrup et al. (2005), where *Pocillopora damicornis* coral fragments experienced a reduction in their maximum ( $F_v/F_m$ ) under anoxic conditions (0 mg O<sub>2</sub> L<sup>-1</sup>). The SLT<sub>50</sub> for maximum  $F_v/F_m$  values for the low O<sub>2</sub> treatment was calculated for both *Porites* sp. ( $5.23 \pm 3.81$ SE days) and *G. fascicularis* ( $3.22 \pm 1.65$ SE days) at 32°C.

The interaction between time, oxygen treatment, and species also significantly affected our experiment's effective photochemical efficiency ( $F'/F_m'$ ), with the low O<sub>2</sub> treatment displaying the lowest ( $F'/F_m'$ ) values for both species by day 9. Haas et al. (2014) showed effective ( $F'/F_m'$ ) values that mirrored our results, where the effective ( $F'/F_m'$ ) measurements for the *A. yongei* coral fragments under the hypoxic treatment (2 to 4 mg O<sub>2</sub> L<sup>-1</sup>) declined until they reached  $\sim 80\%$  of their initial values. The SLT<sub>50</sub> values for effective  $F'/F_m'$  values for the low O<sub>2</sub> treatment at 32°C for *Porites* sp. and *G. fascicularis* were equal to  $6.54 \pm 3.55$ SE days and  $5.43 \pm 2.43$ SE days, respectively.

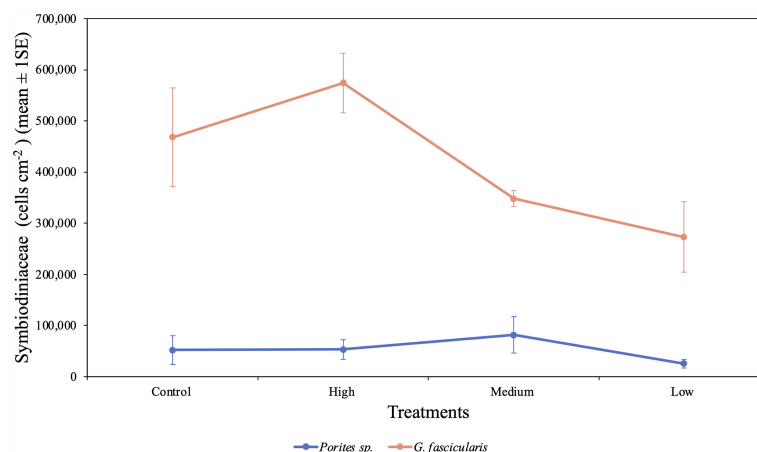


FIGURE 6

Mean ( $\pm$  1SE) rates of Symbiodiniaceae cell density (cells cm<sup>-2</sup>) for *Porites* sp. and *G. fascicularis* under all four O<sub>2</sub> treatments. Control and high O<sub>2</sub> treatment had a more significant cell density for *G. fascicularis*, while medium and low O<sub>2</sub> treatments presented a minor density. *Porites* sp. presented similar cell density between all four treatments.

TABLE 5 Summary of results for LMMs comparing Chl *a* content between treatments for both species.

Source	Numerator <i>df</i>	Denominator <i>df</i>	<i>F</i>	Sig.
Species	1	15.041	44.900	<0.001
O <sub>2</sub> treatment	3	15.841	5.964	<b>0.007</b>
Species * O <sub>2</sub> treatment	3	15.841	4.010	<b>0.028</b>

p-values in bold are statistically significant ( $p < 0.05$ ). BIC (Bayesian information criterion) = 10.739 and AIC (Akaike's information criterion) = 9.194. \*Interaction between species and O<sub>2</sub> treatment.

Both species also displayed the lowest cell count in the low O<sub>2</sub> treatment. Similar results were reported by Alderdice et al. (2021) in their short-term hypoxia assay, where the Symbiodiniaceae cell density (cells cm<sup>-2</sup>) of *Acropora selago* decreased after 12 h of nighttime low DO (~2 mg O<sub>2</sub> L<sup>-1</sup>) exposure. However, the Symbiodiniaceae cell density of *A. selago* fragments did not significantly decrease under their control treatment (~6 mg O<sub>2</sub> L<sup>-1</sup>). Altogether, these results indicate that the severity and duration of deoxygenation events should be considered when estimating coral thresholds to constrained O<sub>2</sub> availability.

In our experiment, the resilient *Porites* sp. fragments displayed a minor decrease in Chl *a* content under the low O<sub>2</sub> treatment; however, the susceptible *G. fascicularis* experienced a significant reduction in Chl *a* content for both medium and low O<sub>2</sub> treatments. Alderdice et al. (2021) reported similar results in their short-term deoxygenation assay, where the Chl *a* content of *Acropora tenuis* fragments remained almost the same in both control and hypoxic (~6 and ~2 mg O<sub>2</sub> L<sup>-1</sup>, respectively) treatment after 12 h of nighttime low O<sub>2</sub> exposure. However, *A. selago* displayed a lower Chl *a* content in the hypoxic treatment (~2 mg O<sub>2</sub> L<sup>-1</sup>) after 12 h of nighttime low O<sub>2</sub> exposure.

In the present study, the chosen experimental DO levels tried to mimic the deoxygenation events observed on coral reefs (Breitburg et al., 2018; Johnson et al., 2021a). Therefore, in the present study, we tested reduced O<sub>2</sub> levels down to between 1.0 and 1.5 mg O<sub>2</sub> L<sup>-1</sup>. However, some *in-situ* studies have recorded deoxygenated events on reefs that can reach anoxic conditions (<1 mg O<sub>2</sub> L<sup>-1</sup>) (Chan et al., 2008; Johnson et al., 2021a). The duration of our study likely aligns with the exposure time of events observed on reefs (Johnson et al., 2021a); nonetheless, it is possible that we could have observed bleaching effects on *Porites* sp. by further reducing DO levels to below 1 mg O<sub>2</sub> L<sup>-1</sup>. Given the differential susceptibility between the two coral species examined here, further research testing lower DO levels (<1 mg O<sub>2</sub> L<sup>-1</sup>) will better determine bleaching thresholds for the resilient species.

Our experimental results support the hypothesis that prolonged exposure to hypoxic conditions with increasing ocean warming may negatively impact coral reefs' survival in the Red Sea's warm water. The Red Sea is inherently prone to hypoxia due to the warm temperatures in summer (up to 35°C; Chaidez et al., 2017) and high salinity, which reduce the solubility of O<sub>2</sub>. In addition, high temperatures lead to high respiratory oxygen demand, so corals may be recurrently

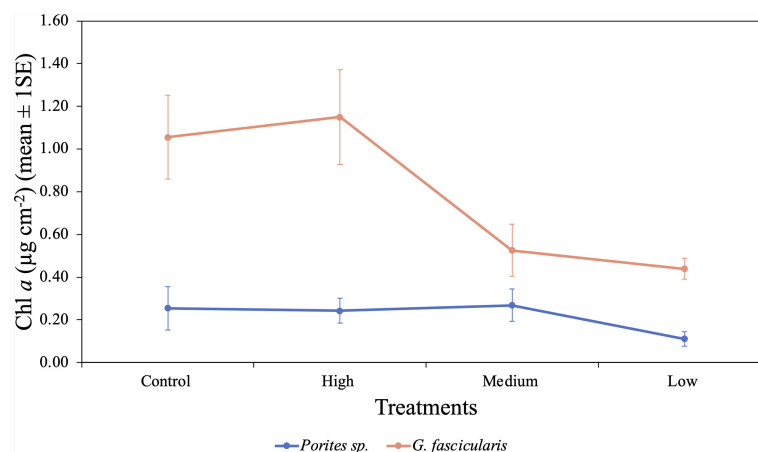


FIGURE 7

Mean ( $\pm$  1SE) Chl *a* ( $\mu\text{g cm}^{-2}$ ) rates for *Porites* sp. and *G. fascicularis* under four O<sub>2</sub> treatments. High and low O<sub>2</sub> treatments had greater Chl *a* content for *G. fascicularis*, while control and medium O<sub>2</sub> treatments presented a minor density. *Porites* sp. presented similar protein content between all four treatments.

**TABLE 6** To-date summary of hypoxic thresholds at which different coral species presented evident tissue degradation, polyps showed either discoloration or appeared withdrawn, or when substantial reductions in pigment were visible.

Species	Hypoxic threshold	Treatments	Exposure time	Source
<i>Acropora selago</i>	~2 mg O <sub>2</sub> L <sup>-1</sup>	~6.00 and 2.00 mg O <sub>2</sub> L <sup>-1</sup>	12 h	<a href="#">Alderdice et al. (2021)</a>
<i>Acropora tenuis</i>	Not observed			
<i>Acropora yongei</i>	2 to 4 mg O <sub>2</sub> L <sup>-1</sup>	6.00 to 8.00, 4.00 to 6.00, and 2.00 to 4.00 mg O <sub>2</sub> L <sup>-1</sup>	10 days	<a href="#">Hass et al. (2014)</a>
<i>Agaricia lamarcki</i>	0.5 mg O <sub>2</sub> L <sup>-1</sup>	>5.00 and 0.50 mg O <sub>2</sub> L <sup>-1</sup>	7 days	<a href="#">Altieri et al. (2017)</a>
<i>Stephanocoenia intersepta</i>	Not observed			
<i>Galaxea fascicularis</i>	~ 1.25 mg O <sub>2</sub> L <sup>-1</sup>	6.80, 5.25, 3.50, and 1.25 mg O <sub>2</sub> L <sup>-1</sup>	10 days	This study
<i>Lophelia pertusa</i>	1.57 mg O <sub>2</sub> L <sup>-1</sup>	~5.32, 2.92, and 1.57 mg O <sub>2</sub> L <sup>-1</sup>	7 days	<a href="#">Lunden et al. (2014)</a>
<i>Acropora cervicornis</i>	~ 1.0 mg O <sub>2</sub> L <sup>-1</sup>	6.25, 4.25, 2.25, and 1.00 mg O <sub>2</sub> L <sup>-1</sup>	5 days	<a href="#">Johnson et al. (2021b)</a>
<i>Orbicella faveolata</i>	Not observed		11 days	
<i>Acropora</i> spp.	<2 mg O <sub>2</sub> L <sup>-1</sup>	Not tested	Not reported	<a href="#">Raj et al. (2020)</a>
<i>Montipora</i> spp.				
<i>Pocillopora</i> spp.				

exposed to hypoxic conditions, often approaching their sublethal thresholds, during summer nights. However, the two species tested showed considerable differences in vulnerability to hypoxia, with *G. fascicularis* bearing large polyps with high respiratory O<sub>2</sub> demands, being highly vulnerable, and *Porites* sp. bearing much smaller polyps with much lower respiratory needs, being far more resistant. Specifically, *Porites* sp. showed high resistance to hypoxia, although all the measured response variables declined under hypoxia.

In contrast, we resolved an O<sub>2</sub> SLT<sub>50</sub> bleaching threshold for *G. fascicularis* of 5.15 ± 0.80SE days under the low O<sub>2</sub> treatment (1.25 mg O<sub>2</sub> L<sup>-1</sup>) at 32°C, the average maximum temperature in the central Red Sea. Hence, sustained heat waves are expected to lead to hypoxia-induced bleaching of vulnerable Red Sea corals, such as *G. fascicularis*. Overall, our findings support the hypothesis that oxygen limitation could be an aggravating factor in increasing coral bleaching in a warming ocean, although greater empirical evidence is required to confirm this.

## Data availability statement

The original contributions presented in the study are included in the article/[Supplementary Material](#). Further inquiries can be directed to the corresponding author.

## Author contributions

Conceptualization: CD and SK. Experimental design: JG, SK and CD. Data collection: JG, SK, TA, SA, AS, and AP. Data analysis/interpretation: JG, SK and CD. Supervision: CD. Writing—original draft: JG. Writing—review and editing: JG, SK and CD. All authors contributed to the article and approved the submitted version.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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## Supplementary material

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fmars.2022.945293/full#supplementary-material>



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