Photophysiology of a mesophotic coral 3 years after transplantation to a shallow environment

Or Ben-Zvi, Raz Tamir, Nir Keren, Dan Tchernov, Ilana Berman-Frank, Yuval Kolodny, Tal Benaltabet, Harel Bavli, Mor Friedman, Noga Glanz-Idan, Hadar Traugott, Yossi Loya & Gal Eyal

Abstract

With shallow coral reefs suffering from an ongoing rapid decline in many regions of the world, the interest in studies on mesophotic coral ecosystems (30–150 m) is growing rapidly. While most photoacclimation responses in corals were documented within the upper 30 m of reefs, in the present study we transplanted fragments of a strictly mesophotic species from the Red Sea, *Euphyllia paradivisa*, from 50 m to 5 m for a period of 3 years. Following the retrieval of the corals, their physiological and photosynthetic properties of the corals were tested. The transplanted corals presented evidence of photosynthetic acclimation to the shallow habitat, lower sensitivity to photoinhibition, and a high survival percentage, while also demonstrating a reduced ability to utilize low light compared to their mesophotic counterparts. This long-term successful transplantation from a mesophotic depth to a shallow habitat has provided us with insights regarding the ability of mesophotic corals and their symbionts to survive and withstand shallow environments, dominated by a completely different light regime. The extensive characterization of the photobiology of *E. paradivisa*, and its photoacclimation response to a high-light environment also demonstrates the plasticity of corals and point out to mechanisms different than those reported previously in shallower corals.

Introduction

Scleractinian corals, the building blocks of coral reefs, are characterized by their symbiosis with endocellular dinoflagellates, mostly from the family Symbiodiniaceae, commonly referred to as zooxanthellae (Trench and Harley [1971](#); Muscatine and Porter [1977](#); LaJeunesse et al. [2018](#)). Mesophotic coral ecosystems (MCEs) are light-dependent coral reefs habitats found at depths between 30 m and approximately 150 m. MCEs are characterized by a strong solar irradiance gradient, resulting in light fluxes that vary between 5 and 100 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ (Hinderstein et al. [2010](#); Einbinder et al. [2016](#); Eyal et al. [2016](#)). Consequently, mesophotic corals might experience more than 100-fold lower irradiance levels than those experienced by shallow corals. MCEs are also characterized by narrower light spectrum, primarily blue light spectrum centered around 467 nm, rather than the wider spectral range of shallow waters (Lesser et al. [2009](#); reviewed by Kahng et al. [2019](#)). Although corals respond to a variety of environmental factors, such as temperature (Fitt et al. [2009](#); Cantin et al. [2010](#)) and water motion (Jokiel [1978a](#)), light quality and irradiance levels have been suggested to play a key role in coral ecology (Veron [1995](#)). Light affects coral settlement (Maida et al. [1994](#); Mundy and Babcock [1998](#)), movement (Yamashiro and Nishira [1995](#)), and competition with other organisms (Benayahu and Loya [1985](#)). Primarily, light is an essential requirement because it enhances the calcification and enables photosynthesis of the coral and its algal symbionts (Chalker et al. [1983](#); Falkowski et al. [1990](#); Lesser [2000](#); Beer et al. [2014](#); Cohen et al. [2016](#)). Photoacclimation refers to acclimation to a particular light environment (Falkowski and Dubinsky [1981](#)). Corals may shift from autotrophic nutrition to heterotrophic or mixotrophic feeding in deeper habitats;

therefore, low-light environments (Mass et al. [2007](#); Alamaru et al. [2009](#); Lesser et al. [2009](#)) display depth-related changes in morphology to enable maximal light capture (Muscatine [1990](#); Anthony et al. [2005](#); Nir et al. [2011](#)) or prevent self-shading (Enríquez et al. [2017](#)) and reorganize their photosystems (Lesser [2000](#)). As the severe deterioration of shallow coral reefs continues globally (Hughes et al. [2018](#)), the interest and investigative studies on MCEs are growing rapidly (Loya et al. [2016](#)), including the reproduction patterns of coral species (Holstein et al. [2015](#); Eyal-Shaham et al. [2016](#); Feldman et al. [2018](#); Shlesinger et al. [2018](#)), community composition (Kahng et al. [2010](#), Kramer et al. [2019](#); Tamir et al. [2019](#)), and the mechanisms of acclimation to this unique environment (Nir et al. [2011](#); Eyal et al. [2016](#); Groves et al. [2018](#); Eyal et al. [2019](#)). The “Deep Reef Refugia Hypothesis,” coined by Bongaerts et al. ([2010](#)), discusses the potential role of mesophotic reefs to provide a source of propagules that may replenish the shallow reefs in cases of extreme loss. The dominance of the coral *Euphyllia paradivisa* in the mesophotic reefs of the Gulf of Eilat/Aqaba (GoE/A), and its complete absence from the shallow reef, suggest its robust adaptation to deep habitats (Eyal et al. [2016](#)). In this study, we thus examined the physiological and photosynthetic characteristics of *E. paradivisa* and its potential photoacclimation to different depths and light environments 3 years after transplantation from 50 m to 5 m depth.

Materials and methods

Experimental design and coral collection

Six colonies of the scleractinian coral *Euphyllia paradivisa* Veron ([1990](#)) were collected at 50 m depth in front of the Dekel Beach, Eilat (29° 32′ 17″ N, 34° 56′ 56″ E). The colonies were tagged and fragmented, with 4–8 polyps from each colony left at their native environment of 50 m depth (i.e., the “deep” fragments). The remaining 4–6 polyps from each colony were transplanted to a coral nursery at 5 m depth (i.e., the “shallow” fragments) in front of the Interuniversity Institute for Marine Science (IUI), Eilat (29° 30′ 16″ N, 34° 55′ 7″ E). All fragments were left at the sites for 3 years to allow the corals to fully acclimate throughout seasonal and yearly variation and were simultaneously retrieved for analysis at the end of this period. Following the coral retrieval and prior to analysis (24 h), the deep fragments were kept under a lighting filter “Lagoon blue” (Lee Filters, Hampshire, UK) that creates a light environment similar to that found at 40–50 m; and the shallow fragments were kept under ambient sunlight in running seawater aquaria systems.

Zooxanthellae density and chlorophyll a concentration

Tentacles from each fragment were sampled and weighed for normalization. The tissue was mechanically broken, centrifuged and separated into host and symbiont fractions. Zooxanthellae density (10^5 cells mg host tissue⁻¹) was determined with a hemocytometer under a light microscope. Following the isolation of the algal fraction from the host tissue, chlorophyll was extracted with 100% cold acetone for 15 h and cellular chlorophyll a concentrations (pg chlorophyll a cell⁻¹) were calculated following Jeffrey and Humphrey ([1975](#)).

Chlorophyll fluorescence measurements

Following retrieval, all fragments were immediately dark-incubated for 1 h and photosystem II (PSII) maximal quantum yield (F_v/F_m) was measured for each coral fragment, using a Diving-Pulse Amplitude Modulation fluorometer (Diving-PAM; Walz GmbH, Effeltrich, Germany) with white saturating pulse (4000 $\mu\text{mol photons m}^{-2}$, white light, 500 ms).

Fluorescence Induction and Relaxation (FIRe) fluorometer (Satlantic, Nova Scotia, Canada) was used to measure F_v/F_m , effective quantum yield ($\Delta F/F_m'$), rate of PSII acceptor side reduction (τ_{QA}), PSII absorption cross section (σ_{PSII}), and non-photochemical quenching (NPQ) values. Each fragment was measured using the FIRe blue light saturation pulse (10,000 $\mu\text{mol photons m}^{-2}$, 80 μs) after 1 h incubation at increasing light intensities (0, 30, 150, and 290 $\mu\text{mol photons m}^{-2} \text{ s}^{-1}$).

Imaging-PAM (Maxi-version, Walz GmbH, Effeltrich, Germany) was used to measure and calculate F_v/F_m , $\Delta F/F_m'$, NPQ, and the approximate relative electron transport rate (rETR). Each fragment was measured with an actinic light saturation pulse (2700 $\mu\text{mol photons m}^{-2} \text{ s}^{-1}$, 800 ms) after 15 min incubation at increasing light intensities (0, 21, 56, 111, 186, 281, 396, 531, and 701 $\mu\text{mol photons m}^{-2} \text{ s}^{-1}$).

Respiration metabolic chambers

Shallow and deep fragments ($n = 9$ from each depth) of similar size were incubated in 250-ml custom-made acrylic metabolic chambers containing 0.22 μ filtered seawater at 23 °C (ambient sea temperature). Prior to incubation, fragment volume was measured by buoyant weight for normalization (Jokiel [1978b](#)). Fragments were incubated under increasing light intensities for 20 min at each intensity (0, 10, 40, 140, 250, 400, 600 $\mu\text{mol photons m}^{-2} \text{ s}^{-1}$) using two full-spectrum metal halide lamp (400 W, 5000 K, 50 Hz, golden light, Israel). Light intensity (E) was recorded using a LI-COR LI-250A light meter (LI-COR, NE, USA). Oxygen evolution was monitored using ProODO Optical Dissolved Oxygen meter (YSI Inc., OH, USA) placed at the top of each chamber. Photosynthetic rates (P) were calculated according to the difference between the final and initial O_2 measurements of each incubation. Photosynthetic efficiency (i.e., slope at the light-limited region, α), irradiance compensation point (E_c), saturation irradiance (E_k), and maximal photosynthesis (P_{max}) were calculated through a hyperbolic fit function (Chalker [1981](#)).

Genetic identification of algal symbionts

DNA was extracted from the algal pellet isolated from each fragment using DNeasy blood and tissue kit (Qiagen, MD, USA) according to the manufacturer's protocol for tissue samples. A ~ 1000 bp fragment of Symbiodiniaceae *COXI* mtDNA was amplified using the primers COXI_FOR2 and COXI_REV1 following Pochon et al. ([2012](#)). Samples were bi-directionally sequenced using ABI 3730XL sequencers (MCLAB, CA, USA). Symbiodiniaceae genera were determined based on the *COXI* sequences available on GeneBank.

Statistical analyses

All statistical analyses were performed with R software (Team [2013](#)). Data were tested with a linear mixed effects model (LMEM) using the package {lme4} (Bates et al. [2015](#)) and {lmerTest} (Kuznetsova et al. [2017](#)) considering "depth" (i.e., the origin depth of the fragment) and "light" (i.e., light intensity when light curves were performed) as fixed effects and "colony" as a random

effect. Post hoc analyses were performed using {emmeans} package (Lenth [2018](#)). Models' residuals were checked visually for normality and with Levene's test for homogeneity of variance.

Results

Physiological responses

Fragments from both depths were retrieved from only four of the six colonies. Deep fragments from the remaining two colonies did not survive; therefore, for those colonies we retrieved only shallow fragments. The mean $\pm$ SD survival rate of the transplanted fragments (i.e., shallow fragments) was $76 \pm 0.29\%$ ($n = 25$), while that of the control group (i.e., deep fragments) was $32 \pm 0.37\%$ ($n = 10$). Zooxanthellae density measurements revealed both significantly higher algal density (Fig. 1a; LMEM, $F = 9.603$, $p = 0.0045$) and chlorophyll a concentration (Fig. 1b; LMEM, $F = 18.527$, $p = 0.0002$) in the deep fragments compared to the shallow fragments.

Fig. 1

Zooxanthellae density and chlorophyll a concentration of *Euphyllia paradivisa* from depth of 50 m and following a deep-to-shallow transplantation. **a** Zooxanthellae density (10^5 cells mg host tissue $^{-1}$) and **b** cellular chlorophyll a concentration (pg chlorophyll cell $^{-1}$) in shallow (5 m; represented in gray, $n = 16$) and deep (50 m; represented in black, $n = 11$) fragments of *E. paradivisa*. Boxes represent the upper and lower quartile, center lines represent medians, and whiskers extend to data measurements that are less than $1.5 \times \text{IQR}$ away from first/third quartile

Photosynthetic characteristics

The maximal PSII quantum yield (F_v/F_m) of the deep fragments measured by the Diving-PAM (Fig. 2) was found to be significantly higher (LMEM, $F = 20.814$, $p = 7.8e-5$) than that of the

shallow fragments. This measurement was performed close to the retrieval time of the corals (and after the 1 h dark-incubation), and therefore best reflects the maximal potential quantum yield of the corals after the transplantation. Deep fragments had a mean $\pm$ SE F_v/F_m value of 0.66 ± 0.02 , while the mean shallow fragments value was 0.49 ± 0.02 .

Fig. 2

Maximal quantum yield (F_v/F_m) of *Euphyllia paradivisa* from depth of 50 m and following a deep-to-shallow transplantation. F_v/F_m measurements obtained with a Diving-PAM of shallow (5 m; represented in gray, $n = 25$) and deep (50 m; represented in black, $n = 11$) fragments of *E.*

paradivisa following a 1 h dark-incubation. Boxes represent the upper and lower quartile, center lines represent medians, and whiskers extend to data measurements that are less than 1.5*IQR away from first/third quartile

Parameters obtained from the FRe measurements (Fig. 3) also indicate that the deep fragments had significantly higher F_v/F_m after the dark-incubation (Fig. 3a; pairwise comparison, $p = 0.009$) and higher $\Delta F/F_m'$ (LMEM, $F = 0.6816$, $p = 0.066$). This was accompanied by a lower σ_{PSII} (Fig. 3b; LMEM, $F = 0.414$, $p = 0.743$), and lower τ_{Qa} values (Fig. 3c; LMEM, $F = 4.13$, $p = 0.051$). NPQ in the deep fragments was significantly lower (Fig. 3d, EMS 1; LMEM, $F = 5.588$, $p = 0.024$) than in the shallow fragments. The difference between the two depths (shallow vs. deep) was more notable under low-light incubation (0 and 30 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$) compared to high light intensity (150 and 290 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$) in all FRe measurements.

Fig. 3

Photosynthetic parameters of *Euphyllia paradivisa* from depth of 50 m and following a deep-to-shallow transplantation obtained using the FRe system. Mean (dots) and SE (error bars) values of **a** effective quantum yield ($\Delta F/F_m'$), **b** functional absorption cross section of PSII (σ_{PSII}), **c** time constant of electron transport on the acceptor side of PSII (τ_{Qa}), and **d** the residuals of non-photochemical quenching (NPQ) of shallow (5 m; represented in gray, $n = 9$) and deep (50 m;

represented in black, $n = 9$) fragments of *E. paradviva*, following 1 h incubation under increasing actinic light intensities (0, 30, 150, and 290 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$)

A comparison of the effective quantum yield in deep and shallow fragments from the same colony using the Imaging-PAM (Fig. 4a, c) revealed that under low-light conditions (up to 281 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$) the shallow fragments demonstrated lower $\Delta F/F_m'$ while under light intensities higher than 281 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$, they became more efficient in utilizing the photons for photosynthesis. The Imaging-PAM also allowed us to notice that the skeleton area is photosynthetically active as well. However, photosynthesis performed by photosynthetically active organisms, on or inside the skeleton, was less efficient than the photosynthesis performed by the algal symbionts within the coral tissue.

Fig. 4

Photosynthetic parameters of *Euphyllia paradviva* from depth of 50 m and following a deep-to-shallow transplantation obtained from Imaging-PAM measurements. Mean (dots) and SE (error bars) values of **a** the effective quantum yield ($\Delta F/F_m'$) and **b** relative electron transfer rate (rETR)

of the shallow (5 m; represented in gray, $n = 3$) and deep (50 m; represented in black, $n = 3$) fragments of *E. paradivisa*, following 15 min incubation under increasing actinic light intensities (0, 21, 56, 111, 186, 281, 396, 531, and 701 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$). c Pseudo-colored images of the quantum yield of shallow polyps (fragment on the right) and deep polyps (fragment on the left) of the same colony measured after incubations under increasing light intensities of 0, 281, and 701 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$. Circles represent the areas of interest (AOI) measured in each fragment

Accordingly, calculation of the relative electron transfer rates (rETR) from the light curves performed with the Imaging-PAM (Fig. 4b) shows that the shallow fragments had lower rETR (LMEM, $F = 0.31$, $p = 0.57$) compared to the deep fragments following low-light incubations (0–280 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$). Under 300–400 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ the rETR was equal, and under higher light ($> 400 \mu\text{mol photons m}^{-2} \text{s}^{-1}$) the shallow fragments demonstrated higher rETR than the deep fragments.

Photosynthesis versus Irradiance/energy (P–E) Curves

Gross photosynthesis (Fig. 5a) was not significantly different between depths under 140 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$, but was higher in the shallow fragments beyond this light intensity (LMEM, $F = 2.749$, $p = 0.02$). Comparing net photosynthesis (Fig. 5b) with the gross photosynthesis (Fig. 5a) demonstrates that shallow fragments had higher respiration rates than the deep fragments but also produced more oxygen. Parameters derived from P–E curves are summarized in Table 1. Shallow fragments reached the compensation point (E_c) and saturation (E_k) at higher light intensities (LMEM, $F = 1.552$, $p = 0.233$ and $F = 1.092$, $p = 0.293$, respectively) and displayed higher P_{max} (LMEM, $F = 1.093$, $p = 0.289$), but lower α (LMEM, $F = 0.0349$, $p = 0.853$) when normalized to the whole polyp volume (tissue and skeleton).

Fig. 5

O₂ evolution as a function of light intensity in *Euphyllia paradivisa* measured using respiration metabolic chambers. Mean (dots) and SE (error bars) values of the **a** gross and **b** net photosynthesis (mg O₂ h⁻¹) of shallow (5 m; represented in gray, *n* = 6) and deep (50 m; represented in black, *n* = 3) fragments of *E. paradivisa*, following 20 min incubation under increasing light intensities (0, 10, 40, 140, 250, 400, and 600 μmol photons m⁻² s⁻¹). The red line represents the values in which respiration and O₂ synthesis are equal

Table 1 Photosynthetic parameters obtained from the photosynthesis versus irradiance (P-E) curves measured by respiration metabolic chambers

	Normalized to	α	E_c	P_{max}	E_k
Shallow	Polyp volume	0.009 ± 0.002	40 ± 7.3	1.007 ± 0.2	122 ± 20
Deep	Polyp volume	0.178 ± 0.009	29 ± 13.4	0.87 ± 0.2	100 ± 30
Shallow	Living tissue volume	5.8e-5 ± 1.4e-5	42 ± 8.3	0.007 ± 0.001	130 ± 23
Deep	Living tissue volume	1.3e-4 ± 8.1e-5	29 ± 13.4	0.006 ± 0.002	100 ± 31

The slope (α) represents the photosynthetic efficiency, E_c is the irradiance compensation point and E_k is the saturation irradiance of *Euphyllia paradivisa*. Data represent the mean ± SE of each parameter

Symbiodiniaceae genetic identification

In all our samples, we found Symbiodiniaceae from the genus *Cladocopium* (formerly known as clade C). Although we cannot eliminate the possibility that the corals had changed their algal symbionts during the 3-year period of the experiment, we can conclude that the different photosynthetic performances of the shallow and deep fragments presented in this study are not a results of different algal symbionts genus at the time of analyses.

Discussion

In light of the severe deterioration of shallow reefs, it was suggested that MCEs may serve as a source of replenishment for their affected shallow counterparts. In order to assess this suggested scenario, we successfully transplanted the mesophotic coral, *Euphyllia paradivisa*, from 50 m to 5 m. This study sought to provide an analysis of the photosynthetic characteristics of a mesophotic coral and evaluate its photoacclimation potential following long-term transplantation to the shallow environment. Moreover, this study provides a comparison between transplanted (shallow) and native (deep) fragments from the same colony, and therefore of the same genotype after 3 years.

Since the distribution of *E. paradivisa* is strictly mesophotic in the GoE/A (Eyal et al. [2016](#); Tamir et al. [2019](#)), we were surprised to find higher survival rates among the fragments that were transplanted from depth to the shallow site compared to the deep fragments that were left as a control group in their natural habitat at 50 m. The corals transplanted to the shallow site were placed on an elevated platform, as describes in previous deep-to-shallow coral transplantations (Cohen and Dubinsky [2015](#)), while the deep fragments were left in their natural position. This difference between shallow and deep fragments may explain the higher survival of shallow fragments following the transplantation, despite the higher light conditions. As MCEs are mainly characterized by a light gradient (Lesser et al. [2009](#); Eyal et al. [2016](#)), and reef corals are considered mostly autotrophic due to their symbiotic relationship with zooxanthellae (Muscatine et al. [1981](#)), corals must balance the reduced light availability to maintain their energetic requirements. In our genetic analysis of the algal symbiont, we found that all our corals harbored the same Symbiodiniaceae genus, *Cladocopium*. Therefore, we consider all of the differences in photosynthetic performances to result from photoacclimation of the photosynthetic apparatus rather than from a genetic shift in the algal community. The transplanted fragments (i.e., shallow fragments) of *E. paradivisa* presented evidence of photoacclimation to a high light regime, while their native, mesophotic, counterparts were more efficient at utilizing low light (Figs. 2, 3, 4, 5). Previous studies have investigated the changes in zooxanthellae densities and chlorophyll a concentration along a depth gradient or following transplantation (reviewed by Kahng et al. [2014](#)). Most reports indicated a decrease in the symbiotic algae density along with an increase in chlorophyll a concentration per cell in deeper corals or low-light conditions (Titlyanov et al. [2001](#); Mass et al. [2007](#)), while others showed that both parameters will increase with depth (Falkowski and Dubinsky [1981](#); Cohen and Dubinsky [2015](#); Polinski and Voss [2018](#)). Results of the current study revealed both higher zooxanthellae densities and higher chlorophyll a concentration in the deep corals that remained in their native habitat (Fig. 1). The higher zooxanthellae density and chlorophyll a concentration may assist mesophotic corals to capture more light and produce more photosynthates (Cohen and Dubinsky [2015](#)). The lower values of those parameters in shallow fragments may result from ultraviolet radiation, high PAR, and the formation of reactive oxygen species that are all known to negatively affect zooxanthellae (Gleason and Wellington [1995](#); Lesser [1996](#)). An additional factor that may have affected the zooxanthellae and chlorophyll a is a difference in temperature. Since our study sites differ at most in 1 °C, we find this factor as less probable to cause such significant variation between the deep and shallow fragments. Natively deeper corals were previously shown to present higher quantum yield compared to shallower corals (Lesser et al. [2010](#)). The deep fragments in the current study demonstrated a better photosynthetic efficiency presenting higher PSII quantum yield (F_v/F_m or $\Delta F/F_m'$) at lower light intensities, as measured with the Diving-PAM (Fig. 2), FIRE (Fig. 3a), and Imaging-PAM (Fig. 4a). The photosynthetic efficiency (α ; Table 1) and net photosynthesis (Fig. 5b) calculated from the respiration metabolic chambers were also greater in the deep fragments. The shallow fragments, in contrast, presented higher σ_{PSII} (Fig. 3b), τ_{Qa} (Fig. 3c), and NPQ (Fig. 3d) values. Those three parameters, which were higher under higher light intensities in the shallow fragments, may indicate a greater dynamic range for light utilization or point to a faster rate of relaxation of the occupied reaction centers of PSII. The higher σ_{PSII} value in shallow fragments may seem counterintuitive when accompanied by lower zooxanthellae density and lower

chlorophyll a concentration, but as we did not analyze all photosynthetic pigments, we can suggest that there are other accessory pigments that may be found at higher concentration in the shallow fragments. Both shallow and deep fragments presented E_k values previously reported for corals found under overhangs, therefore, corals that are experiencing intermediate light intensities (Anthony and Hoegh-Guldberg 2003). Yet, the shallow fragments reached the compensation point (E_c) at higher light intensities (Table 1), indicating that their rate of photosynthesis is lower than their respiration requirements under lower light conditions compared to the deep fragments (Fig. 5b). Furthermore, the transplanted fragments photosynthetic system remained efficient under higher light conditions. Still, the relative electron transfer rate (rETR) was lower in the transplanted shallow corals under low light, and equal to that of the deep corals at 300–400 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ (Fig. 4b). However, under high light conditions, the shallow corals revealed a slightly higher rETR (Fig. 4b), which may suggest higher resistance to photoinhibition, i.e., that they gained some advantage over the mesophotic corals in performing photosynthesis under high light conditions and thereby avoided damage to their photosynthetic apparatus. rETR calculation is based on the assumption that photons are evenly distributed between the two photosystems, and on an estimation that 84% of incident light is absorbed. Although these assumptions may not necessarily hold true, and hence the absolute value of ETR is uncertain, the changes in rETR with environmental parameters are informative nonetheless.

Excluding the parameters derived from the respiration metabolic chambers (Table 1), our measurements demonstrated higher heterogeneity within shallow fragments compared to deep fragments. Similarly, in Edwards and Kim (2010), higher diurnal variability in several photosynthetic parameters was found in shallower parts of the kelp *Macrocystis pyrifera* compared to deeper samples. “Flickering light effect,” caused by surface waves, is more prominent at the upper water column and causes dramatic fluctuations in light intensity (Schubert et al. 2001; Iluz et al. 2012). Hence, shallow environments will experience a less stable light intensity compared to mesophotic environments, which in turn can cause the shallow fragments to display a higher variability in their photosynthetic parameters.

It should be noted that PAM fluorometry is an indirect method for measuring photosynthetic efficiency derived from the fluorescence properties of chlorophyll (Schreiber et al. 1993) and has limitations, especially in corals depending on tissue composition, the presence of the skeleton, and the algal densities (Wangpraseurt et al. 2019). Also, changes in the optical properties resulting from changes in algal symbiont density or photosynthetic pigments concentration may also affect the measurement of photosynthetic rates (Scheufen et al. 2017). Hence, in this study we used different methods for photosynthesis assessment which complement one another and strengthen the results such as the respiration metabolic chambers that supply more direct information regarding photosynthetic efficiency and are influenced by the gas exchange of the deeper layers of coral tissue. Moreover, in all our experimental simulations, while we increased or decreased the light intensities, we did not modify the spectrum of the actinic light. Mass et al. (2007) has noted the issue of using non-ambient light-spectra for mesophotic coral measurements, and we should take into consideration that deeper corals might be more adjusted to a “bluer” light (Kinzie et al. 1984; Kinzie and Hunter 1987), whether through the possession of blue absorbing pigments (Smith et al. 2017) fluorescent proteins (Eyal et al. 2015),

different composition of the photosynthetic apparatus (i.e., different pigmentation ratios; Kaiser et al. [1993](#), Einbinder et al. [2016](#)), or other unknown pathways. Moreover, coral photophysiology may be actually impaired by other, longer, wavelengths as demonstrated by Wijgerde et al. ([2014](#)).

Research in the field of mesophotic coral physiology and photobiology is crucial, now more than ever, as the interest in mesophotic coral ecosystems increases, while shallow coral reefs are undergoing mass mortality events (Hoegh-Guldberg et al. [2007](#); Ainsworth et al. [2016](#); Hughes et al. [2018](#)). Our results only partially answer the question regarding the potential of mesophotic corals to successfully inhabit shallower reefs. However, we provide a novel example of the changes in photosynthetic performances after a successful long-term transplantation of a mesophotic coral to the shallow environment. Unlike previous mesophotic-to-shallow transplantation attempts (Einbinder et al. [2016](#); Laverick and Rogers [2018](#)), the majority of our *E. paradivisa* transplanted fragments survived after 3 years despite the significant change in the light conditions upon transplantation as described by Eyal et al. ([2016](#)). Additionally, as the photophysiological response of the corals in this study did not follow the accepted paradigms regarding photoacclimation, it is possible that they have other photoacclimation mechanisms that are yet to be discovered.

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Author information

Author notes

1. Or Ben-Zvi and Raz Tamir have contributed equally to this work.

Affiliations

1. *School of Zoology, Faculty of Life Sciences, Tel-Aviv University, Tel Aviv, Israel*
 - Or Ben-Zvi
 - , Raz Tamir
 - & Yossi Loya
2. *The Interuniversity Institute for Marine Sciences in Eilat, Eilat, Israel*
 - Or Ben-Zvi
 - , Raz Tamir
 - & Tal Benaltabet
3. *The Institute of Life Sciences, The Hebrew University of Jerusalem, Jerusalem, Israel*
 - Nir Keren
4. *The Leon H. Charney School of Marine Sciences, University of Haifa, Haifa, Israel*
 - Dan Tchernov
 - & Ilana Berman-Frank
5. *Applied Physics Department and The Center for Nanoscience and Nanotechnology, The Hebrew University of Jerusalem, Jerusalem, Israel*
 - Yuval Kolodny
6. *Institute of Earth Sciences, The Hebrew University of Jerusalem, Jerusalem, Israel*
 - Tal Benaltabet
7. *Department of Soil and Water Sciences, The Robert H. Smith Faculty of Agriculture, Food and Environment, The Hebrew University of Jerusalem, Rehovot, Israel*
 - Harel Bavli
8. *Department of Biotechnology Engineering, Ben-Gurion University of the Negev, Beer-Sheva, Israel*
 - Mor Friedman
9. *Plant Sciences and Genetics in Agriculture, The Robert H. Smith Faculty of Agriculture, Food and Environment, The Hebrew University of Jerusalem, Rehovot, Israel*
 - Noga Glanz-Idan

10. *School of Mechanical Engineering, Tel-Aviv University, Tel Aviv, Israel*

- Hadar Traugott

11. *ARC Centre of Excellence for Coral Reef Studies, School of Biological Sciences, The University of Queensland, St Lucia, Australia*

- Gal Eyal

12. *The Mina and Everard Goodman Faculty of Life Sciences, Bar Ilan University, Ramat Gan, Israel*

- Gal Eyal

Corresponding authors

Correspondence to Or Ben-Zvi or Raz Tamir or Gal Eyal.