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Response of Growth and Photosynthesis of *Emiliania huxleyi* to Visible and UV Irradiances under Different Light Regimes

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ABSTRACT

Microalgae are capable of acclimating to changes in light and ultraviolet radiation (UVR, 280–400 nm). However, little is known about how the ecologically important coccolithophore *Emiliania huxleyi* responds to UVR when acclimated to different light regimes. Here, we grew *E. huxleyi* under indoor constant light or fluctuating sunlight with or without UVR, and investigated its growth, photosynthetic performance and pigmentation. Under the indoor constant light regime, the specific growth rate (μ) was highest, while fluctuating outdoor solar radiation significantly decreased the growth rate. Addition of UVR further decreased the growth rate. The repair rate of photosystem II (PSII), as reflected in changes in PSII quantum yield, showed an inverse correlation with growth rate. Cells grown under the indoor constant light regime exhibited the lowest repair rate, while cells from the outdoor fluctuating light regimes significantly increased their repair rate. Addition of UVR increased both the repair rate and intracellular UV-absorbing compounds. This increased repair capability, at the cost of decreased growth rate, persisted after the cells were transferred back to the indoor again, suggesting an enhanced allocation of energy and resources for repair of photosynthetic machinery damage by solar UVR which persisted for a period after transfer from solar UVR.

INTRODUCTION

Emiliania huxleyi is a widespread coccolithophore species and an important component of the marine phytoplankton, frequently forming algal blooms in the world oceans (1), with cell concentrations up to 10^7 cells L^{-1} (2). In contrast to noncalcifying phytoplankton species, *E. huxleyi* uniquely performs calcification in addition to photosynthesis, influencing the balance of source and sink for atmospheric CO_2 (3). At the same time, the accumulation of intracellular dimethyl sulfoniopropionate (DMSP) and subsequent emission of dimethyl sulfide (DMS) by *E. huxleyi* and other algae play a crucial role in formation of cloud condensation nuclei and the growth of aerosol particles (4,5).

Despite the fact that *E. huxleyi* blooms occur in numerous coastal and pelagic waters, this species is mostly distributed within the upper mixed layer (UML) at a depth of approximately 10–30 m (6), being able to tolerate high levels of solar radiation. Nevertheless, transitions between relative darkness to near-surface high light intensity may occur within 1 h or even within minutes in turbulent oceans or estuarine areas (7). In surface waters, maximal sunlight can reach up to 2000–2500 μmol photons m^{-2} s^{-1} around noon, therefore *E. huxleyi* and other phytoplankton cells within the UML can be exposed to solar radiation levels of different orders of intensity, depending on their spatial and temporal distributions.

While *E. huxleyi* is known to be able to tolerate high light stress compared to other phytoplankton species, it performs less well under fluctuating light conditions. Previous work has demonstrated that *E. huxleyi* was more sensitive, in terms of viability loss, to excessive light exposure than the diatom *Thalassiosira weissflogii* when grown over a range of fluctuating irradiance regimes (8). *E. huxleyi* grows best under moderate irradiance (9), with its maximum growth observed at about 150–200 μmol photons m^{-2} s^{-1}, and maximal photosynthetic carbon fixation at about 300 μmol photons m^{-2} s^{-1} (10–14). Under highly fluctuating light regimes, *E. huxleyi* may invest additional energy into photoacclimation and regulation of the photosynthetic machinery (15,16).

Solar UV radiation (UVR, 280–400 nm) is known to damage protein (17–19) and DNA (20–22) and reduce photosynthesis and growth (23) of many algal species, though its impact differs at different depths (24), between coastal and offshore waters (25) or among different taxa (26). In contrast to other phytoplankton species, coccolithophores such as *E. huxleyi* produce coccoliths around the cells, which can shield off a significant part of UVR (27), thus play a photoprotective role.

Nevertheless, effects of UVR on photosynthetic organisms observed from laboratory studies may not reflect those of natural solar UV radiation (28). Photophysiological performances were found to be significantly different between indoor (low and constant light) and outdoor (fluctuating and usually high) acclimated cells (12,29). However, our current understanding of how *E. huxleyi* responds to light changes or to UVR is, to a large extent, based on indoor studies. We have limited understanding of the acclimation processes that might occur when cells are moved from constant irradiances typical of laboratory cultures to fluctuating solar PAR regimes with or without UVR. Both variations of light intensity and frequency might significantly influence the cells′ physiological performance. However, little has been documented on this alga about its photosynthetic performance during acclimation and de-acclimation (from solar radiation with UVR

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to constant laboratory conditions). In this study, we investigated and compared the growth and photosynthetic performance of *E. huxleyi* during constant indoor and fluctuating outdoor solar radiation with or without UVR, and then examined the cells′ performance after they were transferred back to the constant light condition during de-acclimation. At the same time, responses of the cells to acute UVR were examined under a solar simulator in order to determine rate constants for damage and repair processes and how these vary with growth conditions.

MATERIALS AND METHODS

Species and culture conditions. *Emiliania huxleyi* (CS-369) was obtained from the Australian National Algae Culture Collection (CSIRO, Hobart) and maintained in Aquil artificial seawater medium. During the indoor growth period, the cultures were exposed to cool-white fluorescent light at about 450 μmol photons m^{-2} s^{-1} (12L:12D) and 20°C in a plant growth chamber (GXZ-300D, Ningbo Southeast, China). During the outdoor growth period, the cultures were incubated in a water bath under full spectrum solar radiation with or without UVR at 20°C, with temperature controlled by a cooling circulator (CAP-3000, Rikakikai, Japan). Cultures were grown semi-continuously and diluted every 48 h to maintain the cells in their exponential growth phase (the cell numbers ranged from 5×10^3 to 5×10^4 cells mL^{-1}) and cultures were maintained for at least eight generations under indoor or outdoor light regimes before the physiological parameters were measured. The dilutions were carried out with freshly prepared sterilized medium equilibrated with the ambient CO_2 level. The carbonate system was thus maintained relatively stable with variations in pH of less than 0.07 and 0.05 units for indoor and outdoor cultures, respectively. The pH_{NBS} was measured every 24 h, before cell counts were carried out. The enclosed cultures were shaken three times a day to ensure equal exposure of the cells to light.

Experimental design. The experiments were carried out at the State Key Laboratory of Marine Environmental Science, Xiamen University. Four distinct light regimes, namely (1) PAR-I, (2) Solar PAR, (3) solar PAR + UV and (4) PAR-II. These represent constant (indoor) PAR, Outdoor sunlight without UVR, Outdoor full spectrum solar radiation with UVR and return to indoor previous conditions, respectively, and were set up in sequence undergoing these four light regimes successively. Each light treatment lasted for at least 7 days (about eight generations) to ensure sufficient acclimation time before the data were obtained.

Radiation treatment and UVR. The light intensity of PAR-I was set at 450 μmol photons m^{-2} s^{-1}. Solar PAR was incident sunlight with the cells in quartz tubes (500 mL) covered with a cut-off foil which transmitted solar radiation above 395 nm. Although UV radiation A (UVA, 315–400 nm) and UV radiation B (UVB, 280–315 nm) were present in the incident light for the Solar PAR treatment as shown in Fig. 1A, the cut-off foils ensured cells received PAR only. The Solar PAR + UV treatment involved exposure of the cells, in quartz tubes, to the full spectrum of solar radiation. The indoor constant light intensity was measured by a three-channel irradiation apparatus (PAM2100, Solar Light), while the outdoor solar radiation was continuously monitored using a broadband filter radiometer (ELDONET) which records PAR (400–700 nm), UVA and UVB irradiances every minute. The difference in PAR levels measured by the two devices was about 6%, which was calibrated for reasonable comparisons.

Measurements of physiological parameters. During each light regime, the cell numbers were counted with a Z2 Coulter Particle Count and Size Analyzer (Beckman, Instruments, Florida, US) at 7 pm every day (the photoperiod was from 6:30 am to 6:30 pm). At the end of each light treatment, pigment content was determined and the effect of acute UVR exposure on the quantum yield of *E. huxleyi* was then examined during a 60 min exposure under a solar simulator (Sol 1200 W, A.G., Hönle, Martinstried, German). The average irradiances of PAR, UVA and UVB during the 60 min exposure under the solar simulator were 123.05 (590 μmol photons m^{-2} s^{-1}), 30.76 and 1.95W m^{-2}, respectively. At the beginning of each experimental exposure, three quartz tubes were wrapped in aluminum foil to dark-adapt (15 min) the cells for the measurement of maximal fluorescence. The cultures were then exposed to three acute radiation treatments: (1) P (PAR), tubes covered with a 395 nm cut-off foil, transmitting irradiances above 395 nm; (2) PA (PAR + UVA), tubes covered with 320 nm cut-off foil, transmitting irradiances above 320 nm; (3) PAB (PAR + UVA + UVB), tubes covered with a 295 nm cut-off foil, transmitting irradiances above 295 nm. Triplicate cultures were run under each treatment.

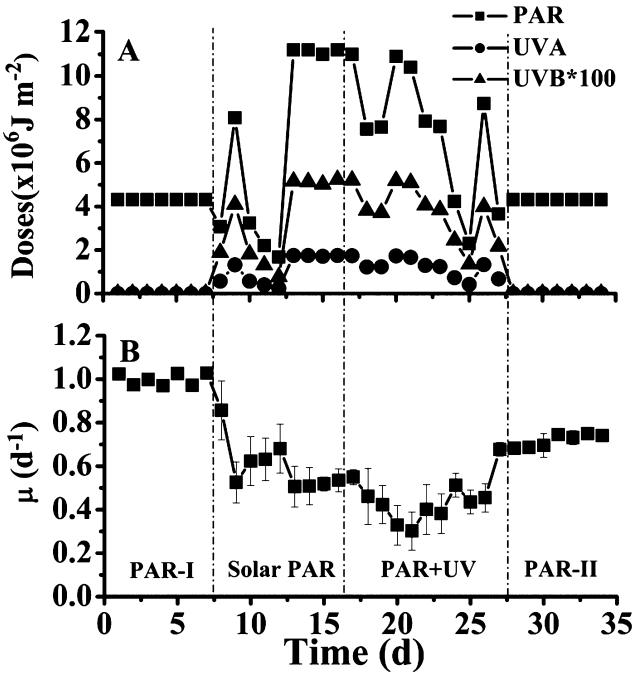

Figure 1. The daily doses (A) of PAR, UVA and UVB under four different light regimes during which *Emiliania huxleyi* (CS-369) cells were grown in sequence, PAR-I and PAR-II represent indoor constant light level at 450 μmol photons m^{-2} s^{-1}; Solar PAR and PAR + UV represent fluctuating solar radiation without or with UVR, respectively. Under the Solar PAR condition, cells only received PAR instead of the full spectrum solar radiation. (B) The corresponding specific growth rate (μ) of the *E. huxleyi* cells under four different light regimes. Triplicate cultures were run under each treatment. The vertical lines indicate SD (n = 3). The experiment was conducted on May 10 to June 13, 2012.

Determination of growth rate and pigments. The specific growth rate (μ) was determined as follows: $\mu = (\ln C_2 - \ln C_1)/(T_2 - T_1)$, where C_2 and C_1 represent the cell concentrations at T_2 (before dilution) and T_1 (after previous dilution) time, respectively.

Chlorophyll*a*, chlorophyll*c*, carotenoids and UV-absorbing compounds (UVACs) of about 4×10^4 cells were measured by filtering 100 mL culture on a Whatman GF/F filter and extraction with 5 mL 90% methanol overnight followed by centrifugation for 10 min at 4°C (5000 g). The absorption of the supernatant was measured by a scanning spectrophotometer (DU800, Beckman Coulter Inc.). Contents of Chl*a*, *c* and carotenoids were determined according to Porra (30), Ritchie (31), and Strickland and Parsons (32). The main absorption values for UV-absorbing compounds ranged between wavelengths of 310 and 360 nm, and the peak absorption value at 340 nm was used to estimate total absorptivity of UVACs according to Dunlap *et al.* (33). The absorptivity of UVACs was finally normalized to the Chl*a* content (μg (μg Chl*a*)$^{-1}$).

Measurements of photochemical fluorescence parameters. Chlorophyll fluorescence parameters were measured with a pulse-amplitude-modulated fluorometer (Xe-PAM, Walz, Germany) every 5–10 min during 60 min exposure under a solar simulator lamp. The effective quantum yield of PSII ($\Delta F/F_m'$) was measured with actinic light levels similar to that of the growth light with a saturating light pulse of 4000 μmol photons m^{-2} s^{-1} (pulse duration: 0.8s). $\Delta F = F_m' - F_t$, where F_m' and F_t are the maximal fluorescence and steady state fluorescence values under the relevant light condition. Nonphotochemical quenching (NPQ) was determined according to the Stern-Volmer equation: $NPQ = (F_m - F_m')/F_m'$, where F_m represents the maximum fluorescence in dark-adapted (15 min) cells. The rate of UVR-induced damage (k) of PSII and the

corresponding repair rate (r) were estimated according to the Kok equation (34):

$$P_n/P_0 = r/(r+k) + k/(r+k) * \exp(-(k+r) * t);$$

where P_n and P_0 represent effective quantum yield values at time T_n and T_0, respectively.

Data analysis. Prior to the statistical analyses, data were tested for normality and homoscedasticity. One-way ANOVA and Tukey tests were then used to determine the significance of differences in the physiological and biochemical parameters. A Two-way ANOVA was also performed to examine the significant difference of the interaction effects between light history and UVR levels on damage and repair rate. The significance level was set at 0.05.

RESULTS

Daily dose and specific growth rate

The specific growth rates of *E. huxleyi* were highest under the constant irradiance PAR regime (PAR-I) at 4.23 × 10^6 J m^{-2}, and decreased when cultures were transferred to Solar PAR alone (*i.e.* with UVR screened off) during outdoor exposure, even at an average daytime dose of 3.07 × 10^6 J m^{-2} in the first day of outdoor acclimation (Fig. 1A), with the highest μ values observed at the lowest daytime dose (1.68 × 10^6 J m^{-2}) on day 12. When the cells were sequentially transferred to exposures to PAR + UV, μ decreased further, with the lowest values found under the relatively higher solar radiation level at day 20 (Fig. 1B). After the cells were de-acclimated (transferred back to an indoor constant light regime at 4.23 × 10^6 J m^{-2}), μ increased, but did not attain the level reached during the previous indoor growth period before transfer of cultures to the solar exposures. An inverse linear relationship was established between the μ values and doses of PAR, both for the solar PAR alone treatment and for that with UVR (Fig. 2). Under the indoor constant light regimes, although the PAR levels were similar, the growth rate under PAR-I was significantly higher than that in PAR-II ($P < 0.01$). When compared at the same daily doses equivalent to those in the PAR-I regime (4.23 × 10^6 J m^{-2}), according to the linear fit equation, the growth rates were 0.66 and 0.51 d^{-1} for Solar PAR and PAR + UV, respectively. Fluctuating solar radiation thus reduced μ by 51.3% compared to the PAR-I light regime, while addition of UVR further decreased μ by about 29.4% (Fig. 2).

Pigment content

During the acclimation stages, the cell contents of chl*a*, chl*c* and carotenoids (Fig. 3A–C) under all light regimes changed significantly. Initially, in the indoor cultures, the three kinds of pigment showed the highest values, then significantly decreased ($P < 0.01$) when the cells were transferred to solar fluctuating PAR for 9 days (10 generations) (Fig. 3A–C). Higher values of these pigments were observed when the cells were acclimated (11 days) to solar radiation with UVR. When transferred back to the indoor light regime (PAR-II) without UVR (7 days), chl*a* ($P = 0.02$) and carotenoid ($P < 0.01$) contents decreased, while chl*c* showed an insignificant change ($P = 0.927$). The cell content of UVACs increased significantly ($P < 0.01$) under solar radiation with UVR, by 4.9, 12.9 and 8.5 fold compared to the other three light regimes, respectively. UVAC content was also significantly higher in the PAR-I grown cells than in those under Solar PAR ($P = 0.037$) and PAR-II ($P = 0.041$).

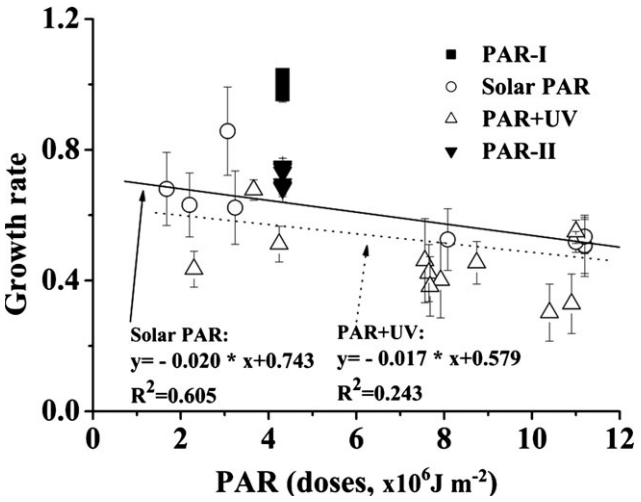

Figure 2. Relationship of the specific growth rates with daily PAR doses in *E. huxleyi* cells grown under the four different light regimes (Fig. 1A). The solid and dotted lines are representative for the linear relationship of μ with PAR doses in Solar PAR and PAR + UV outdoor light regimes, respectively. The vertical bars indicate the SD ($n = 3$), representing independent three cultures. Data derived from the Fig. 1.

Effective quantum yield and Nonphotochemical quenching

After acclimation to each of the light regimes, the initial effective quantum yield was the highest under PAR-I and the lowest under solar PAR + UV (Fig. 4A,C). While the dynamic changes of effective quantum yield during 60 min solar simulator exposure declined quickly within 10–15 min and then leveled off, for PAR-I and Solar PAR acclimated cells, the asymptotes of the effective quantum yield were always lowest under the PAB treatment, indicating a significant influence of UVR on the photochemical performance. The inhibition of the final asymptote yield value caused by UVA and UVB was 16.2% and 58.1% in PAR-I, while in the Solar PAR light regime the UVA and UVB caused inhibition of 13.7% and 49.2%, respectively. For PAR + UV and PAR-II grown cells, the eventual asymptote yield value was nearly the same for cells grown under the PAB, PA and P treatments (Fig. 4; $P > 0.5$). Similarly, in PAR-II, the asymptote yields were similar in all treatments ($P > 0.5$).

The change in NPQ during the exposures varied under the four different light regimes (Fig. 5). The NPQ values were the lowest under PAR + UV regimes compared to other treatments ($P < 0.01$) (Fig. 5C). Moreover, NPQ values were significantly higher in cells grown under PAR-II than in PAR-I ($P < 0.01$) (Fig. 5D). However, differences in NPQ among PAB, PA and P treatments were not significant in the cells grown under the PAR + UV and PAR-II light regimes (with P values ranging from 0.2–0.5) (Fig. 5C,D).

Repair and damage rates

The effects of acute UVR exposure on cells grown under the various irradiance regimes are shown in Fig. 6. The damage and repair rates of the PSII reaction center were estimated from the exponential decay in the effective quantum yield ($R^2 \geq 0.9$), as described by the Kok equation (35), when cells were exposed to short-term, acute exposures to the light from a solar simulator. In the PAR-I grown cells, there was a significant difference in

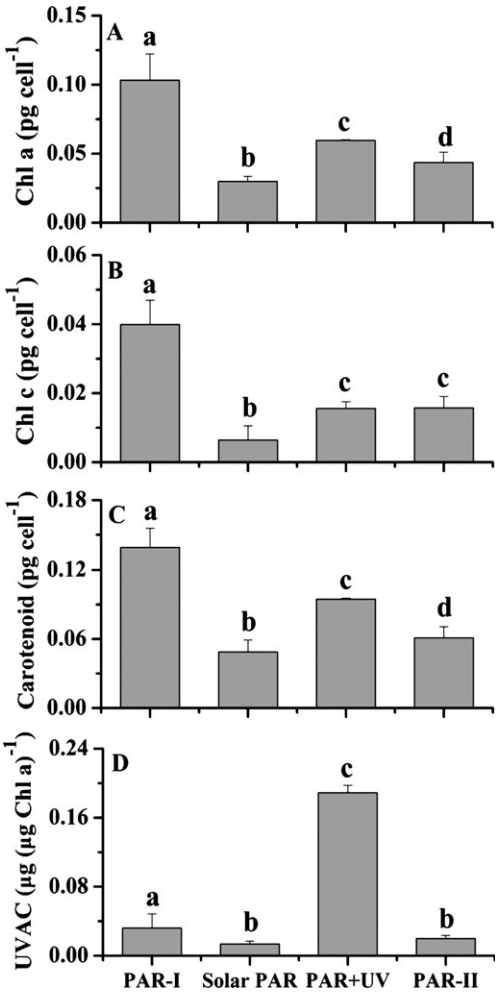

Figure 3. Contents of Chl*a* (A), Chl*c* (B), Carotenoids (C) and UV-absorbing compounds (UVACs) (D) in *E. huxleyi* (CS-369) under different light regimes (as detailed in Fig. 1). Sampling was conducted at day 7, 16, 27 and 34 for each light regime. Different superscripted letters represent significant differences among different light regimes. The vertical lines indicate the SD (n = 3), representing independent three cultures.

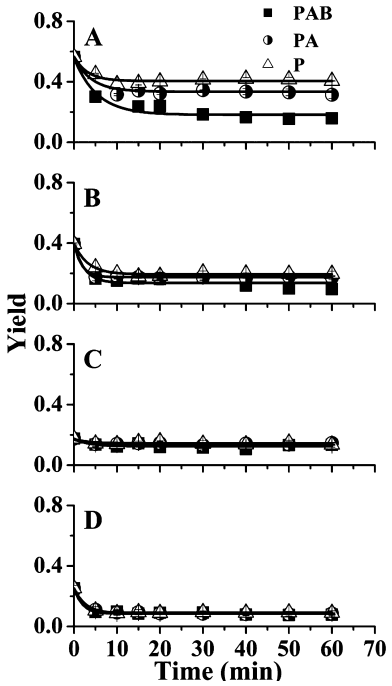

Figure 4. Dynamic changes in PSII effective quantum yield of *E. huxleyi* (CS-369) cells after PAR-I (A), Solar PAR (B), PAR + UV (C) and PAR-II (D) acclimation while exposed to solar radiation simulator for 60 min under PAB, PA and P treatments. The average light intensity for PAR, UVA and UVB was 123.05 (590 μmol photons m^{-2} s^{-1}), 30.76 and 1.95 W m^{-2}, respectively. Triplicate cultures were run under each treatment, the vertical bars indicate SD (n = 3).

repair rates among the PAB and PA (P = 0.035), PAB and P (P < 0.01) exposures. The P treatment resulted in the highest r while the presence of UVA decreased r by about 33%. Addition of UVB resulted in a further decrement of r by 60%. In the Solar PAR grown cells, P exposure resulted in a relatively higher repair rate while the presence of UVB resulted in the lowest and the presence of UVA lead to the highest, repair rate. Overall, the repair rate of Solar PAR grown cells was significantly higher than for cells grown under the other three light treatments. For the Solar PAR + UV treatment, the mean value of repair rate was much higher than that in PAR-I grown cells but lower compared to the Solar PAR grown cells. After transfer back to indoor light (PAR-II), the repair rate did not show a significant difference between P and PAB exposures (P = 0.178), while the presence of UVA significantly lowered the repair rate (Fig. 6A). However, compared to PAR-I grown cells, the repair rate of PAR-II cells was significantly higher (P < 0.01).

During the PAR-I acclimation, PAB exposure resulted in the highest damage rate k, which was 20% and 38.4% higher than those in PA and P exposures, respectively (Fig. 6B). For the Solar PAR treatment, the k under all three exposures were the highest compared to the other three light treatments, with values of k of 0.135, 0.102 and 0.079 min^{-1} for the PAB, PA and P exposures respectively. During the solar PAR + UV treatment, there were no significant differences in k among PAB, PA and P exposures ($p_{(PAB/PA)}$ = 0.836, $p_{(PAB/P)}$=0.617, $p_{(PA/P)}$=0.769), and the equivalent damage rates were the lowest of all, compared to the other three light treatments. Following the return of cultures indoors (PAR-II), the k did not show significant differences except for the PA exposure ($p_{(PAB/PA)}$<0.01, $p_{(PAB/P)}$ = 0.476, $p_{(PA/P)}$ = 0.038).

The r:k ratio gives a measure of the net impact of UVR exposure on effective quantum yield. Ratios<1 imply net damage while if r>k, repair capacity is greater than damage, so the exposure has no net effect on yield. Ratios of r:k were lowest in the PAR-I grown cells (Fig. 6C). In the Solar PAR treatment, the r:k ratios under all three exposures were higher than those in PAR-I grown cells. The solar PAR + UV treatment resulted in the highest r:k ratio, with the ratio under all three short-term exposures approaching 1. In the PAR-II incubation, there was no significant difference in r:k among the P, PA, PAB exposures ($p_{(PAB/PA)}$ = 0.903, $p_{(PAB/P)}$ = 0.334, $p_{(PA/P)}$ = 0.398), though overall they were much higher in PAR-II than those in PAR-I cultures.

DISCUSSION

Emiliania huxleyi (CS-369) showed highest growth rates, content of photosynthetic pigments and effective quantum yield under

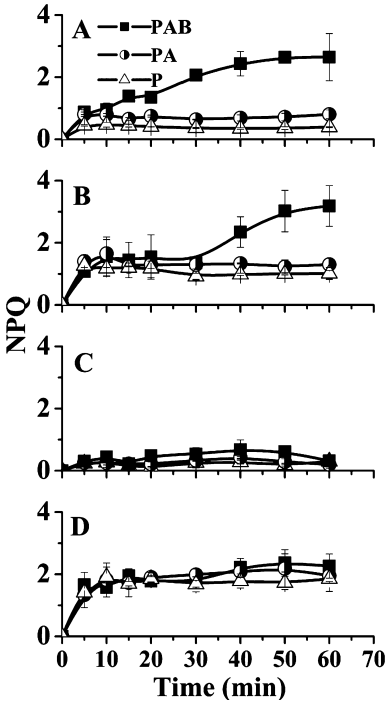

Figure 5. The dynamic changes of Nonphotochemical quenching (NPQ) of *E. huxleyi* (CS-369) cells during a 60 min acute exposure under PAB, PA and P treatments after acclimation to four different light histories: (A) PAR-I, (B) Solar PAR, (C) PAR + UV, (D) PAR-II. Triplicate cultures were run under each treatment, the vertical bars indicate SD (n = 3).

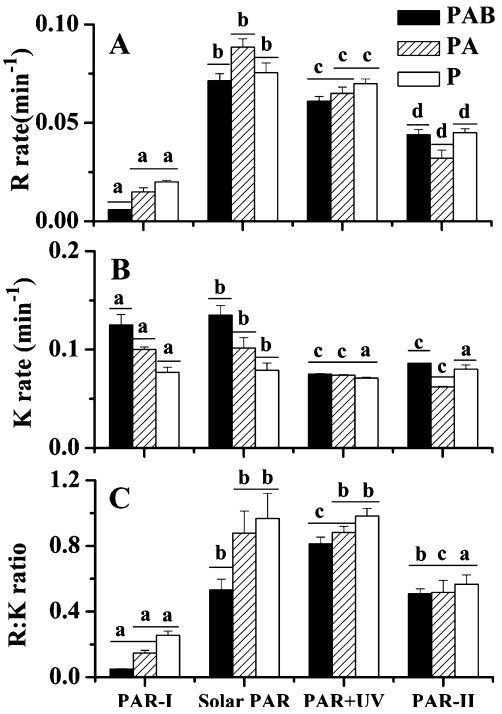

Figure 6. Rate of Repair r (A), damage k (B) and the ratio of repair: damage (C) after acclimation to four different light regimes during a 60 min acute exposure experiment under a solar radiation simulator. Different superscript letters represent significant differences ($P < 0.05$) among different light regimes. Triplicate cultures were run under each treatment, the vertical bars indicate SD (n = 3).

the constant indoor light regimes compared to those grown under fluctuating solar PAR with or without UVR. The presence of UVR decreased the growth rate, but increased the content of UV-absorbing compounds, so that the cells showed less UVR-related damage to PSII and had a higher value of *r:k*. De-acclimation, after transfer of cells back to the indoor light regime, did not lead to full recovery of the quantum yield, pigment content or growth rates after 7 days. Extrapolating from the data in Fig. 1, complete recovery would need approximately 32 days, based on the linear relationship between time and growth rate (data not shown). During outdoor acclimation, the decreased growth rate can be attributed to the high and fluctuating solar radiation that would increase the burden on photo-acclimation; addition of UVR resulted in a further decline in growth rate presumably because of the DNA damage, inhibition of PSII, and increased energy budget needed for PSII repair. The partial recovery in terms of growth rate and the quantum yield suggest that other cellular photo-protective mechanisms, up-regulated under the outdoor solar light stress, were still functioning after several generations of indoor constant light acclimation. In other studies, a decreased growth rate under UVR exposure has been interpreted as a sign of initiation of UVR-defence mechanisms (36), thus we postulate that the additional investment of resources and energy into those mechanisms markedly decreased the cells′ growth rate during the PAR-II acclimation.

Part of the acclimation strategy to fluctuating light stress includes changes in pigment composition, especially the well-known role of carotenoids in light-harvesting and photoprotection. Although the daily doses were almost the same, chlorophyll *a*, *c* and carotenoid contents were lower in PAR-II than in PAR-I, indicating that outdoor fluctuating solar radiation with UVR markedly damaged the intracellular photosynthetic apparatus and significantly decreased cells' photosynthetic ability, which had not recovered even after 7 days of acclimation to constant indoor irradiance (PAR-II). Thus, cells had down-regulated their pigment content in order to prevent further photodamage.

Furthermore, it is especially worth noting that the level of UVACs we found in *E. huxleyi* was significant higher under the PAR + UV acclimation. The maximum absorption peak for the UVAC compounds in our study ranged between 310–360 nm, which is consistent with the spectral characteristics of mycosporine-like amino acids (MAAs) (37). The production of MAAs is a very flexible and species-specific mechanism (38). Cell size influences the effectiveness of MAAs in terms of the investment and return on MAA formation (39). For the picoplankters (cell radius<1 μm), even a small increase in MAA content would require heavy investments. For a cell radius between 1 μm and 10 μm, MAA accumulation is performed at the cost of relatively heavy investments and restricted efficiencies compared to microplankters in which the cell radii are above 10 μm (36). In this study, the average cell radius ranged between 2 and 3 μm. After a period of outdoor acclimation, especially in the PAR + UV treatment, cells invested a good deal of resources and energy into UVAC synthesis. In PAR-I, Solar PAR and PAR-II acclimations, although the UVR was negligible, *E. huxleyi* synthesized a small quantity of UVAC. This may reflect their acclimation strategies in natural ecosystems. In a relatively low light level with weak UVR, *E. huxleyi* synthesizes minimal amounts of UVAC to reduce metabolic investment, while when cells are exposed, through vertical mix-

ing, to natural high light stress (both PAR and UVR), the already existing small amount of UVACs can provide limited protection to the cell from the initial onset of UVR stress, but then *E. huxleyi* gradually up-regulates synthesis of UVACs to cope with the ever-increasing levels of UVR stress. However, the increase of UVACs is performed at the cost of decreased growth rate with a relatively high investment and low internal self-shading efficiency. Previous study (40) has demonstrated that even *Thalassiosira weissflogii*, a larger cell, did not maintain elevated levels of MAAs after long term acclimation. When *E. huxleyi* was transferred to low UVR conditions, cells would down-regulate the synthesis of UVACs in order to save resources and energy for other cellular metabolic processes.

Although *E. huxleyi* can tolerate high light in the ocean (6), there is still the potential for damage to the photosynthetic machinery, primarily photosystem II. A daily occurrence in phytoplankton cells is the PSII damage and repair cycle, involving mainly the rapid degradation and resynthesis (turnover) of the D1 32 kDa reaction-center protein (41,42). Under natural excessive light stress and the fluctuating UVR conditions, the degradation rate of D1 protein exceeds the rate of its repair, resulting in increased net photoinhibition. Under such circumstances, the attempt to rebalance the damage and repair rate by adjusting intracellular energy and resources allocation is of great significance for cells′ survival. Despite the fact that there is evidence for direct impact on tyrosine residues in the D1 protein in PSII (43), previous studies have also demonstrated that the excess light energy absorbed by photosynthetic pigments can lead to accelerated photoinhibition by inhibition of the repair of photodamaged PSII (44). This is consistent with the observation that the chlorophyte alga *Dunaliella tertiolecta* (45) showed both increased repair and increased damage rate with increasing PAR. Under such a scenario, the outdoor acclimated cells in our experiment showed enhanced repair rates to maintain photosystem functioning, especially under the Solar PAR light regime, which had the average highest repair rate of all. Moreover, although the last total PAR dose (when sampling was conducted) under the Solar PAR + UV light regime was lowest compared to other three light regimes, the fluctuating solar light stress and the presence of UVR still induced an accelerated repair rate in spite of the fact that the cells had the highest concentration of UVACs. This accelerated repair rate even persisted when cells were under the de-acclimation (PAR-II) conditions. Compared to the PAR-I regime, the repair rate of PAR-II was higher, suggesting that fluctuating light condition indeed increased the cellular repair rate and that this persisted, albeit at a lower level than in the PAR + UV treatment, though we infer that the accelerated repair rate would finally disappear after prolonged stable indoor exposure.

In the cells that had acclimated to incident solar radiation must have obtained or up-regulated their photoprotective strategies, since they had the lowest NPQ when exposed to UVR. Such photoprotective strategies may include enhancement of repair or reduction of damage of photosynthetic machinery and stimulated heat dissipation as well as elevated content of UVACs.

By comparing specific growth and photosynthetic parameters under different light regimes, our results emphasize that the physiological performance of *E. huxleyi* cells can differ signally when grown under different light regimes or solar radiation fluctuations, considering the acclimation-persistent influence on the cells′ physiology. For *E. huxleyi,* physiological performance observed on a given day may be the net result of that day′s and/or previous days′ comprehensive influences. Therefore, light histories are essential when assessing its physiological, especially its photosynthetic performances. While full sunlight is only experienced in the upper meters of water columns, our results showed consistency with another study (46) in that fluctuating (solar) PAR decreased growth rates of some phytoplankton species in contrast to constant PAR exposure. In nature, phytoplankton cells are mixed up and down within the UML, they may need to spend extra energy to cope with the fast light changes and light stress, so that less energy is used for particulate carbon production. With enhanced stratification of the UML associated with global warming, phytoplankton cells within this layer may suffer more light stress due to increased integrated solar exposures with thinned mixing path.

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