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PRIMARY RESEARCH PAPER

Effects of ultraviolet radiation on productivity and nitrogen fixation in the Cyanobacterium, *Anabaena* sp. (Newton's strain)

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Abstract The biological effects of ultraviolet radiation (UVR; 290–400 nm), especially the UV-B (320–400 nm) component of the spectrum, include both direct and indirect effects on many cellular processes. In cyanobacteria both photosynthesis and nitrogen fixation can be affected directly by UVR, and indirectly by UVR through the production of reactive oxygen species (ROS). For the heterocystous cyanobacterium, *Anabaena* sp. (Newton's strain), exposure to UVR causes a significant decline in the quantum yields of photosystem II (PSII) fluorescence and maximum productivity despite an increase in UVR absorbing compounds, mycosporine-like amino acids (MAAs), in those cells exposed to UVR. Concurrent with these observations are significant increases in the activities of superoxide dismutase indicative of an increase in the level of oxidative stress in cells exposed to UVR. Additionally, measurements of nitrogenase activity (acetylene reduction) show a significant decrease in cyanobacteria exposed to UVR, which manifests itself as a decrease in cellular nitrogen and an increase in C:N ratios. These results show that these nitrogen-fixing cyanobacteria are particularly sensitive to UVR, both its direct and indirect effects. The effects of UVR reported here add to the increasing evidence that UVR effects on this important group of prokaryotes could affect the input of new nitrogen, and the biogeochemical cycling of this essential macronutrient in terrestrial, marine, and freshwater habitats.

Keywords Ultraviolet radiation · Cyanobacteria · Nitrogen fixation · Photosynthesis · Oxidative stress

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Introduction

The decrease of stratospheric ozone from anthropogenic inputs of chlorinated fluorocarbons has resulted in an increase in the amount of harmful ultraviolet-B radiation (UV-B, 290–320 nm) reaching the biosphere (Blumthaler & Ambach, 1990; Stolarski et al., 1992; Kerr & McElroy 1993; Madronich et al., 1995). Additionally, UVR can penetrate to significant depths in marine and aquatic ecosystems (Díaz et al., 2000; Tedetti & Sempéré, 2006) with significant biological effects. Ultraviolet radiation, both UV-A (320–400 nm) and UV-B, is known to have a detrimental effect on photosynthesis and growth in cyanobacteria (Vincent & Roy, 1993). The harmful effects of UVR also involve direct damage to DNA and proteins, oxidation of membrane lipids, and photooxidation of chlorophyll or damage to photosystem II (PSII) (Vincent & Neale, 2000; Lesser, 2006).

The prokaryotic contributors to primary productivity in marine and freshwater ecosystems, both heterotrophic bacteria (Herndl et al., 1993) and cyanobacteria are known to be sensitive to UVR (Vincent & Roy, 1993; Vincent & Quesada, 1994). In particular, UVR causes a reduction in growth rates (Vincent & Quesada, 1994), photosynthesis (Hirosawa & Miyachi, 1983a, b), function of PSII (Vincent & Quesada, 1994), damage to DNA (He et al., 2002), negative phototaxis (Donkor et al., 1993), and nitrogen fixation (Falcón et al., 2002) in many cyanobacteria. Studies on the causes for these effects have shown that oxidative stress, caused by the production of reactive oxygen species (ROS), is the primary mechanism of damage (Asada & Takahashi, 1987; Halliwell & Gutteridge, 1999; He & Häder, 2002a, b; Lesser, 2006). Cells produce ROS such as superoxide radicals (O_2^-) via the univalent pathway of reduction of oxygen, while the continued reduction of O_2^- produces hydrogen peroxide (H_2O_2) and hydroxyl radicals ($HO^{\cdot}$) as intermediate and final products (Asada & Takahashi, 1987; Halliwell & Gutteridge, 1999; Lesser, 2006). The enzymes superoxide dismutase (SOD), catalase, and various peroxidases normally function together to inactivate O_2^- and H_2O_2, thereby preventing the formation of $HO^{\cdot}$ the most reactive of the ROS, and subsequent cellular damage (Fridovich, 1986).

In addition to antioxidant protection another mechanism to prevent the direct and indirect effects of UVR exposure is to produce a variety of UVR-absorbing compounds. Cyanobacteria produce an extracellular sheath pigment, scytonemin, which is a lipid-soluble compound exhibiting a broad (280–450 nm) absorption band (Garcia-Pichel & Castenholz, 1993a, b). Although scytonemin absorbs wavelengths in the UV-B range, it is primarily considered to be a UV-A blocker, but cyanobacteria also produce a group of UVR absorbing compounds known as mycosporine-like amino acids (MAAs) (Garcia-Pichel & Castenholz, 1993a, b). MAAs are water-soluble and have a range of absorption maxima from 310 nm to 360 nm, spanning both the UV-B and UV-A wavelengths. MAAs are found in a wide variety of marine organisms including microalgae, macroalgae, invertebrates, fish, and cyanobacteria (Ehling-Schulz & Scherer, 1999; Sinha et al., 2001b; Shick & Dunlap, 2002). A protective role for these compounds has been inferred from their UVR-absorbing properties, increased biosynthesis subsequent to UVR exposure, and their antioxidant properties (Shick & Dunlap, 2002; Lesser, 2006). Studies by Garcia-Pichel & Castenholz (1993a, b) have demonstrated a protective function for these compounds in cyanobacteria. For MAAs in cyanobacteria, however, their effectiveness has been shown to be below 50%, suggesting that damaging photons of UVR are still reaching critical cellular targets.

As nitrogen is known to be a limiting nutrient in many environments the ability of certain prokaryotes to reduce dinitrogen to ammonia is of fundamental importance in producing organic and inorganic forms of nitrogen in the biosphere (Paerl, 1990). In marine and freshwater cyanobacteria, unicellular and filamentous heterocystous and non-heterocystous forms are the predominant and best-described group from an ecological perspective. Nitrogen fixation is catalyzed by the enzyme nitrogenase which consists of two functional proteins, a Mo-Fe protein (245 kd) and a smaller Fe protein (60.5 kd), both of which are susceptible to the direct effects of UVR (Newton et al., 1979; Kumar et al., 2003), and inactivation by molecular oxygen and ROS (Fay, 1992).

Amongst the taxa that do fix nitrogen several strategies to protect nitrogenase have evolved to avoid, or decrease, the effects of molecular and ROS, since both are produced in high quantities during photosynthesis in diazotrophs (Pienkos et al., 1983; Smith et al., 1987; Fay, 1992). These strategies include the sequestering of nitrogenase in thick-walled cells called heterocysts. *Anabaena* and *Nostoc* are two genera that produce these highly reduced, and spatially separated, environments to allow nitrogen fixation to occur simultaneously with photosynthesis, since there are no oxygen evolving components (PSII) within the heterocysts. In the absence of heterocysts the alternative strategy to protect nitrogenase from molecular oxygen, or ROS, is to temporally separate photosynthesis and nitrogen fixation (Fay, 1992).

In this study a common filamentous and heterocystous cyanobacterium is exposed to spectrally realistic irradiances of UVR in laboratory experiments to examine the effects of UVR on photosynthesis and nitrogen fixation, and to assess the induction of protective mechanisms to ameliorate those effects.

Materials and methods

Culture conditions

The freshwater heterocystous cyanobacterium, *Anabaena* sp. (Newton's strain, Newton & Herman, 1979), from the University of New Hampshire maintained microbial culture collection was grown in batch cultures on a 14 h:10 h light:dark cycle at 25°C and maintained in fabricated cylindrical vessels in a temperature-controlled incubator, bubbled with air, illuminated from above, and stirred magnetically. Culture volume was 3.0 l with the tops of the cylinders made of 0.25 inch UVR transparent (UVT; cutoff at 290 nm) and UVR opaque (UVO; cutoff at 385 nm) Plexiglas (Cadillac Plastics, Baltimore, MD). Ports were installed in the sides of the containers for adding and withdrawing samples. The cultures were started in these containers ($N = 3$ for each treatment) under visible irradiance only and transferred to their respective treatments 24 h later and maintained under a 14 h:10 h light:dark cycle at 25°C for 11 days. The media used for these cultures was DY IV growth medium (Culture Collection of Marine Phytoplankton, Bigelow Laboratory of Ocean Sciences) without an inorganic nitrogen source to stimulate nitrogen fixation. Irradiances of visible (photosynthetically active radiation (PAR), 400–700 nm, μmole quanta m^{-2} s^{-1}) and UVR (290–400 nm, W m^{-2}) for acclimation experiments were supplied by fluorescent lamps (visible: F40, General Electric; UVR: UV−340, Q-Panel Inc.) suspended ~50 cm above the cultures.

Measurement of visible and ultraviolet radiation

Ultraviolet radiation (UV-B and UV-A) and PAR were measured using a wavelength and radiometrically calibrated (NIST traceable standards) CCD spectrometer and fiber optics (Ocean Optics, Inc.). Three scans were taken and the mean reported in units of W m^{-2} nm^{-1}. Integrated values of unweighted total UVR, UV-A and UV-B (W m^{-2}) were calculated for each treatment. Weighted UVR irradiances were obtained by multiplying the spectral UVR data by the biological weighting function (BWF) for picocyanobacterial photosynthesis described in Callieri et al. (2001).

Biomass measurements

Cell-free extracts for enzyme analyses at the end of the experiment were obtained by collecting 50 ml samples of culture. These samples were sonicated in microcentrifuge tubes at 4.0°C for 1–2 min in 100 mM phosphate buffer pH 7.0, and the cell suspension centrifuged for 30 min at 10,000 RPM using a microcentrifuge. The supernatant was then used for the measurement of enzyme activities. The concentration of chlorophyll *a* (chl *a*) was measured on triplicate samples of 10 ml collected on Whatman GF/F filters and extracted in 10 ml of 90% acetone at 4.0°C in the dark for at least 24 h. The chlorophyll extracts were read against acetone blanks on a spectrophotometer (Hewlett Packard HP 8452A diode array) at 630 and 663 nm, and the equations of Jeffrey & Humphrey (1975) used to calculate chl *a* concentrations.

Triplicate specimens of 10 ml were taken at the end of the experiment for the determination of particulate carbon and nitrogen. These samples were filtered onto baked (450°C for 4 h) GF/F filters and stored in a desiccator. Samples were subsequently frozen at -50°C, and freeze-dried overnight prior to use. These samples were then combusted in a Control Equipment Corporation (Perkin–Elmer) 240 XA elemental analyzer with an automatic sampler in an air-tight box to keep the samples dry. Acetanilide was used as a standard and the results expressed as C:N (μg:μg) ratios to assess the nutrient status of the cells.

Active fluorescence

A pulse-amplitude modulated (PAM) fluorometer (Walz, Inc. PhytoPam) was used for discrete measurements of in vivo chlorophyll fluorescence during the culture experiments. Fluorescence was measured after at least 30 min acclimation in the dark. Due to the interaction between cyanobacterial respiratory and photosynthetic electron transport, which energizes the thylakoid membrane, and the fact that dark acclimation in cyanobacteria does not lead to state 1, but state 2, there is the potential for the underestimation of F_m (Büchel & Wilhelm, 1993). PAM measurements were, therefore, adjusted by changing the amplitude of the non-actinic measuring beam, using a greater biomass of cells, and testing different frequencies and duration's of saturating flashes to obtain good

measurements of PSII quantum yields. Under normal conditions, the in vivo fluorescence of chlorophyll varies between a minimum yield, F_o, and an enhanced yield, F_m. These values correspond to whether the stable PSII electron acceptor of the complex, Q_a, is oxidized or reduced. The difference between F_o and F_m fluorescent yields ($F_m - F_o$) is the variable fluorescence, F_v, a relative measure of the ability of the reaction center to accumulate reduced Q_a, and F_v/F_m is the quantum yield of PSII fluorescence or a measure of the number of functional PSII units.

Photosynthesis versus irradiance curves

The method of Lewis & Smith (1983) was used to make measurements of photosynthesis as a function of irradiance (*P-E*). Samples ($N = 23$) from each treatment were inoculated with ^{14}C-bicarbonate (final activity approximately 10 μCi ml^{-1}) and aliquots of 1 ml were dispensed into scintillation vials (7 ml capacity) in a temperature-controlled aluminum block. A range of irradiances was provided from below with 2 ENH projection lamps directed through a heat filter of circulating water, and attenuated with neutral density screens. Quantum scalar irradiance in each position was measured with a Biospherical Instruments QSL-100 4π sensor with a modified collector, small enough to fit in the bottom half of a scintillation vial for the measurements. Incubations began within 30 min of sampling and were terminated after 20 min. After incubation samples were immediately poisoned with 50 μl borate-buffered formalin, then acidified with 0.25 ml 6N HCl and shaken in a hood to expel inorganic ^{14}C. Subsequently, scintillation fluid was added (Ecolume, ICN) and dpm's were determined in a scintillation counter, using the H# method of quench correction. Subsamples (20 μl) from the original inoculation were placed in 4 ml fluor plus 0.2 ml phenethylamine to determine the amount of label added. Total CO_2 under these culture conditions has been determined to be 2.1 mM. All measurements were corrected for quench. No correction for isotope discrimination was made.

The *P-E* equation of Platt et al. (1980) was used to model the productivity results to calculate P_s^B (μg C μg chl a^{-1} h^{-1}), the instantaneous rate of photosynthesis at any specific irradiance; α^B (μg C μg chl a^{-1} h^{-1} (μmol quanta m^{-2} s^{-1})$^{-1}$) is the initial slope of the *P-E* curve, and β^B (μg C μg chl a^{-1} h^{-1} (μmol quanta m^{-2} s^{-1})$^{-1}$) is a parameter used to characterize photoinhibition at irradiances greater than the saturation irradiance (E_k) of the *P-E* curve. The maximum rate of photosynthesis, P_{max}^B (μg C μg chl a^{-1} h^{-1}), was calculated as described by Platt et al. (1980).

Nitrogen fixation

All measurements of nitrogen fixation were made on samples at the end of the experiments using the acetylene reduction assay (Hardy et al., 1968), as a measure of nitrogenase activity. Samples of cultures were incubated in serum bottles (100 ml bottles with 50 ml of culture) in triplicate for each culture tested and acetylene injected into the headspace. Acetylene is reduced to ethylene by the enzyme nitrogenase and quantified using gas-chromatography (GC) and flame ionization detection. The rates of acetylene reduction were normalized to chl *a* concentrations.

Superoxide dismutase activity

Total SOD activity was measured spectrophotometrically as described by Elstner & Heupel (1976) and Oyanagui (1984). Standard curves were produced using purified Fe SOD (Sigma). All enzyme assays were conducted at the acclimation temperature and expressed as units (U) of enzyme activity (1.0 U = μmole substrate converted per minute) per μg chl *a*.

Mycosporine-like amino acids

Three 25 ml aliquots were taken at the end of the experiment from each treatment, filtered as described above, and placed in 5 ml of 100% methanol. Mycosporine-like amino acids were separated and quantified in these samples using isocratic reverse-phase HPLC as described by Lesser & Stochaj (1990). MAAs were identified using co-chromatography with known primary and secondary standards, and concentrations were calculated using peak-area calibration factors from the analysis of primary and secondary standards, and normalized on a chl *a* basis.

Statistical analysis

All end point measurements are reported as means (±SE), and were statistically analyzed using an analysis of variance (ANOVA) while the fluorescence data were analyzed as a two-factor (treatment and time) ANOVA with Student-Neuman-Keuls (SNK) post-hoc testing as needed.

Results

UVR irradiances for the UVT cultures were 3.65 W m^{-2} compared to 0.04 W m^{-2} for the UVO cultures while UV-B irradiances for the UVT cultures were 0.36 W m^{-2} compared to 0.00 W m^{-2} for the UVO cultures. Visible irradiances for both treatments were $\sim$100 µmol quanta m^{-2} s^{-1} with ±3% variability over the field of downwelling exposure for the experimental set up. Weighted irradiances, using the cyanobacterial BWF for UVR effects on photosynthesis were 2.62×10^{-7} (dimensionless) for UVT cultures versus 5.33×10^{-11} for UVO cultures

At the end of the experimental period the mean concentration of chlorophyll *a* in the cultures exposed to UVR (2.1 ± 0.09 mg l^{-1}) was significantly (ANOVA: $P < 0.0001$) lower than UVO cultures (5.3 ± 0.06 mg l^{-1}). There was a significant effect of treatment (ANOVA: $P < 0.001$) and time (ANOVA: $P < 0.01$) with a significant interaction (ANOVA: $P < 0.001$) on the dark-acclimated quantum yields of PSII fluorescence. Quantum yields were significantly reduced by exposure to UVR on day 7 (Fig. 1, SNK: $P < 0.001$) and 11 (Fig. 1, SNK: $P < 0.01$) suggesting the accumulation of un-repaired damage over time in cultures exposed to UVR.

Photosynthesis-irradiance curves were measured on day 11. Of the photosynthetic parameters calculated chlorophyll specific maximum photosynthesis (P^B_{max}) was significantly reduced (ANOVA $P < 0.05$) by UVR (1.43 ± 0.09 µg C µg chl a^{-1} h^{-1}) exposure compared to UVO (1.90 ± 0.11 µg C µg chl a^{-1} h^{-1}) values (Fig. 2a, b). This represents a 25% decline in maximum productivity for cultures exposed to UVR. While measurements of β^B (µg C µg chl a^{-1} h^{-1} (µmol quanta m^{-2} s^{-1})$^{-1}$) were not significantly different between treatments (ANOVA: $P > 0.05$), treatment comparisons of α^B, a measure of the efficiency of light capture, were significantly different between UVR (0.013 ± 0.002 µg C µg chl a^{-1} h^{-1} (µmol quanta m^{-2} s^{-1})$^{-1}$) and UVO (0.055 ± 0.009 µg C µg chl a^{-1} h^{-1} (µmol quanta m^{-2} s^{-1})$^{-1}$ treatments (ANOVA: $P < 0.01$).

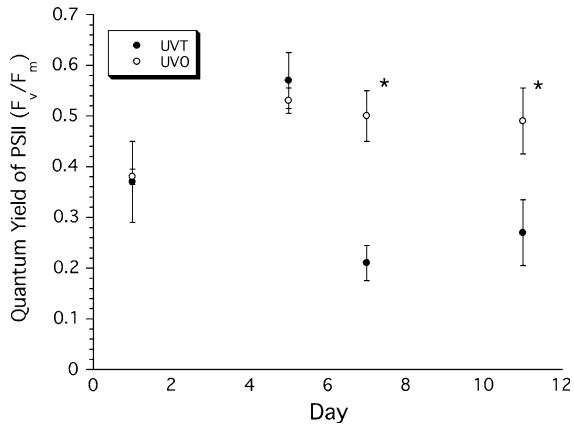

Fig. 1 Measurements of the quantum yield of PSII in cultures of *Anabaena* sp. exposed to UVR (UVT) and control cultures without exposure to UVR (UVO) over the course of the experiment. Results are reported as mean ± SE. Time periods with an * indicate statistically significant differences between UVT and UVO cultures

Rates of nitrogen fixation (Fig. 3a), measured as the activity of nitrogenase, were significantly reduced in UVR cultures (1.76 ± 0.52 nmole C_2H_4 µg chl a^{-1} h^{-1}) compared to UVO cultures (4.06 ± 0.39 nmole C_2H_4 µg chl a^{-1} h^{-1}) which is a 57% decline in the rate of nitrogen fixation. The decline in nitrogen fixation is manifested as a decline in particulate nitrogen and an increase in the C:N ratio of cells exposed to UVR (Fig. 3b). Cells exposed to UVR had significantly (ANOVA: $P < 0.05$) higher C:N ratios (10.4 ± 1.9) than cultures in the UVO treatment (6.2 ± 1.3).

In response to UVR exposure cells of *Anabaena* sp. respond by significantly (ANOVA: $P < 0.05$) increasing their activities of the antioxidant enzyme SOD (Fig. 4a, UVT; 15.2 ± 4.10 U µg chl a^{-1}, UVO; 7.5 ± 2.3 µg chl a^{-1}) and the concentration of total MAAs (Fig. 4b, UVT; 22.5 ± 8.6 nmole µg chl a^{-1}, UVO; 5.3 ± 3.3 nmole µg chl a^{-1}). The MAA shinorine made-up 100% of the MAA pool in the UVO cultures while both shinorine (93%) and mycosporine-glycine (7%), an MAA with known antioxidant properties (Shick & Dunlap, 2002), made-up the MAA pool in the UVR cultures.

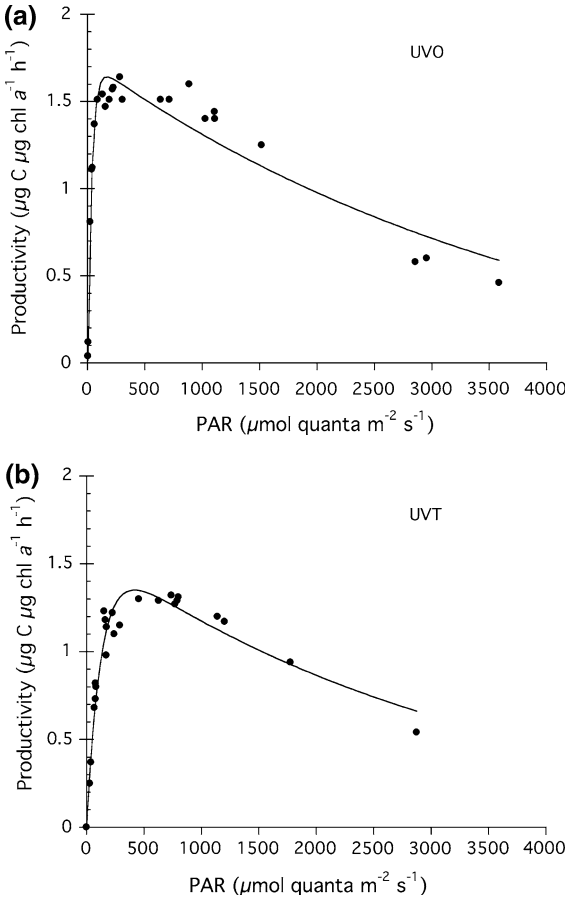

Fig. 2 Photosynthesis versus irradiance (*P-E*) curves for *Anabaena* sp. (**a**) cultures not exposed to UVR (UVO), (**b**) cultures exposed to UVR (UVT). See Results for analysis of fitted values

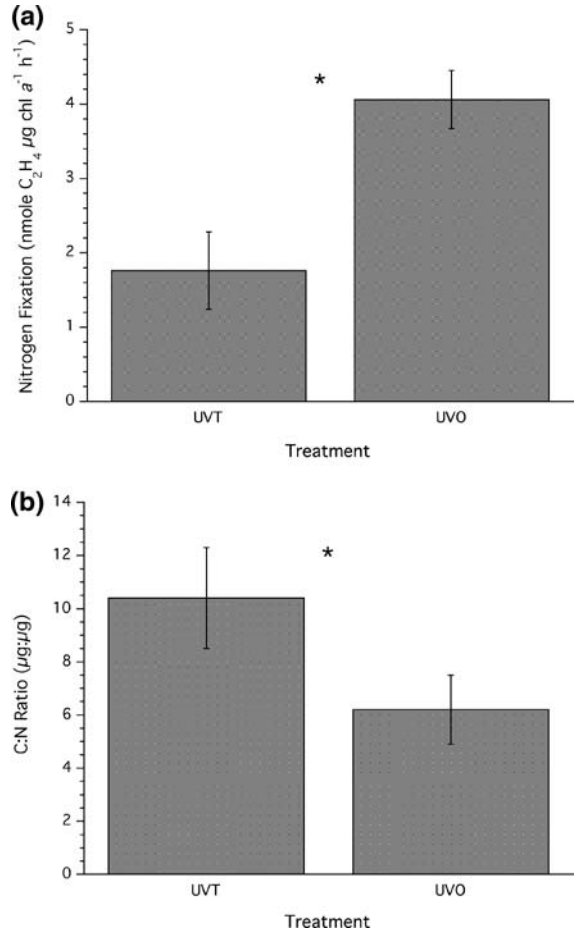

Fig. 3 (**a**) Measurements of nitrogen fixation in cultures of *Anabaena* sp. exposed to UVR (UVT) and control cultures without exposure to UVR (UVO) at the end of the experiment. (**b**) Measurements of carbon:nitrogen (C:N) ratios in cultures of *Anabaena* sp. exposed to UVR (UVT) and control cultures without exposure to UVR (UVO) at the end of the experiment. All results are reported as mean ± SE. * indicates statistically significant differences between UVT and UVO cultures

Discussion

The acclimation studies presented here show that both photosynthesis and nitrogen fixation in *Anabaena* sp. (Newton's isolate) are significantly affected by moderate irradiances of UVR far below what is commonly observed in the natural environment (Tedetti & Sempéré, 2006). Dark-acclimated quantum yields of PSII fluorescence showed a decline upon UVR exposure that is consistent with damage to PSII at critical PSII proteins such as D1 (Lesser et al., 1996; Hazzard et al., 1997; Bouchard et al., 2006). Turnover of the D1 protein is known to be enhanced during exposure to UVR (Greenberg et al., 1989; Bouchard et al., 2006) and since the quantum yield measurements were made on dark acclimated samples it is unlikely that dynamic photoinhibition and the accompanying non-photochemical quenching explains the long-term changes in quantum yields.

Consistent with the changes in the quantum yield of PSII fluorescence, both chlorophyll *a* and maximum photosynthetic capacity were significantly lower in UVR exposed cultures at the end of the experiment. Chlorophyll *a* decreased by ~60% and maximum photosynthetic capacity, normalized to chlorophyll, decreased by ~25% in cultures exposed to UVR. Decreases in cellular chlorophyll might not, a priori, result in a decrease in photosynthesis if the

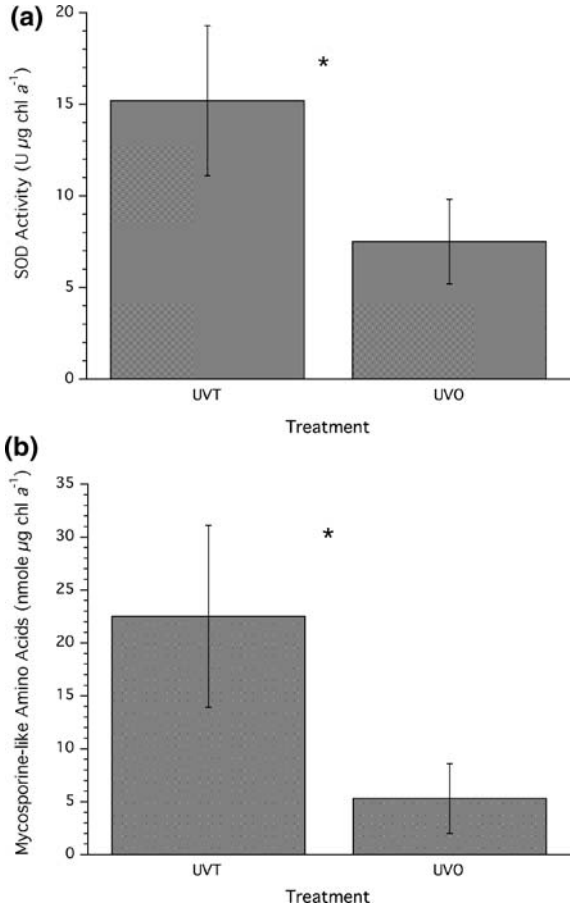

Fig. 4 (**a**) Measurements of superoxide dismutase (SOD) activities in cultures of *Anabaena* sp. exposed to UVR (UVT) and control cultures without exposure to UVR (UVO) at the end of the experiment. (**b**) Measurements of mycosporine-like amino acids (MAAs) in cultures of *Anabaena* sp. exposed to UVR (UVT) and control cultures without exposure to UVR (UVO) at the end of the experiment. All results are reported as mean ± SE. * indicates statistically significant differences between UVT and UVO cultures

efficiency of photon absorption increases as a result of a reduction in the "package effect" (Dubinsky et al., 1986). The observed decreases in maximum photosynthetic capacity could also be caused, in part, from UVR damage to ribulose bisphosphate 1, 5 carboxylase/oxygenase (Rubisco) since maximum productivity is controlled by the activity of Rubisco in a variety of microalgae, including cyanobacteria (Lesser, 1996; Lesser et al., 1996; Hazzard et al., 1997; Sinha et al., 2001a, MacDonald et al., 2003).

Exposure to UVR had a significant effect on the activity of nitrogenase, measured as the reduction of acetylene, in cultures of *Anabaena* sp. exposed to UVR. While this study does not discern between the direct and indirect effects of UVR on nitrogenase and subsequently nitrogen fixation, the 57% decline in nitrogen fixation and particulate nitrogen explains most of the increase in the C:N ratios for cultures exposed to UVR. These declines are actually modest compared to studies showing complete cessation of nitrogen fixation under short term exposures (i.e., hours) of UVR irradiances highly enriched in short-wavelength UV-B (Kumar et al., 2003). Biomass specific growth rates were not assessed during these studies but these cultures started out using the same biomass and the decreases in productivity and nitrogen fixation clearly contributed to the lower chlorophyll *a* biomass observed in cultures exposed to UVR which is similar to studies on both laboratory cultures and natural communities of cyanobacteria (Falcón et al., 2002; Han et al., 2003).

The response to exposure to UVR also included an increase in the activities of the antioxidant enzyme SOD. Oxidative stress is a well-known byproduct of exposure to UVR and can result in damage to PSII, DNA damage, and a decrease in the activities of both Rubisco and nitrogenase (Asada and Takahashi, 1987; Richter et al., 1990a, b; Fay, 1992; He et al., 2002), and an increase in the activities of antioxidant enzymes is a common response to sub-lethal oxidative stress (Lesser, 2006). Additionally, in the cultures of *Anabaena* sp. the MAA shinorine was present and increased in concentration when exposed to UVR as observed in previous investigations on cyanobacteria (Sinha et al., 2001b). The synthesis of MAAs that have strong broadband protective filter effects against UVR has been observed in many species of microalgae and are a component of the acclimation response to UVR (Lesser, 1996; Lesser et al., 1996; Neale et al., 1998; Shick & Dunlap, 2002), including cyanobacteria (Sinha et al., 2001b). The biosynthesis of the MAA mycosporine-glycine was also induced by exposure to UVR, and in addition to being one of the few MAAs whose absorption maxima occurs in the UV-B portion of the spectrum, it has also been shown to quench ROS (Shick and Dunlap, 2002).

Over the timescale of the experiments the effects of moderate UVR exposures caused significant declines in productivity and nitrogen fixation of *Anabaena* sp. (Newton's isolate). These changes

were not ameliorated by significant increases in the ability to block the direct effects of UVR, or by an increase in antioxidant protection. In those environments where heterocystous cyanobacteria are found exposed to natural irradiances of UVR the costs of defending against the effects UVR exposure, such as decreases in carbon and nitrogen fixation, could have significant ecological impacts.

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