Environmental and Experimental Biology (2021) 19: 89–96
http://doi.org/10.22364/eeb.19.09

Original Paper

Effect of UV-B radiation on physiological and biochemical changes in a freshwater cyanobacterium, *Scytonema hofmannii*

ISSN 2255-9582

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Abstract

The effect of ultraviolet-B (UV-B) radiation and UV-B supplemented with low photosynthetically active radiation was investigated on photosynthesis, photosynthetic pigments, phycobilisomes, peroxidation of membrane lipids, fatty acids, and UV-B absorbing pigments in the freshwater cyanobacterium, *Scytonema hofmannii*. The present work aimed to study the UV-B radiation effect on photosynthesis and relate it to oxidative damage and protection provided by the UV-B absorbing compounds in a freshwater cyanobacterium. The results showed that photosynthesis and photosynthetic pigments were primarily affected by UV-B radiation, which seems to be the consequence of the generation of reactive oxygen species (ROS), as both ROS generation and oxidative damage to membrane lipids were observed, resulting in decreased survival of the test organisms. There was no significant change in the saturation and unsaturation level of fatty acids due to the treatments. We also suggest the role of mycosporine-like amino acids (MAAs) and scytonemin in protection against UV-B damage. Our results suggest that changes in photosynthesis, pigments, and peroxidation of membrane lipids may be a consequence of damage. In contrast, changes in fatty acids essentially resulted from the adaptation process against UV-B, and changes in UV-B absorbing compounds such as MAAs and scytonemin provided protection against the harmful effect of UV-B radiation.

Key words: cyanobacteria, fatty acids, mycosporine like amino acids, oxidative damage, photosynthesis, pigments, scytonemin, UV-B.
Abbreviations: ROS, reactive oxygen species; MAAs, mycosporine like amino acids; PAR, photosynthetically active radiation; TBA, thiobarbituric acid; TCA, trichloroacetic acid; UV-B, ultraviolet-B.

Introduction

Cyanobacteria are an ancient and diverse group of microorganisms that can inhabit and thrive in an incredible variety of environments (Singh et al. 2016). In addition to being a key player in aquatic productivity, several cyanobacteria have drawn attention for their ability to produce a variety of bioactive secondary metabolites such as pigments, vitamins, and enzymes with varied applications (de Morais et al. 2015). The changes in climatic conditions such as increased atmospheric carbon dioxide, temperature, precipitation, ultraviolet (UV-B) radiation, etc., can affect various physiological and biochemical processes in aquatic and crop plants, thereby affecting the food chain of the primary producers (WHO 1989). In the aquatic ecosystem, UV rays are not reflected from the water surface, as they mainly penetrate it and may go deep into the water (Kuhn et al. 1999). This highly energetic UV-B radiation has great potential to affect the metabolism of aquatic organisms directly by affecting their critical cellular types of machinery such as lipids, proteins, and DNA, or to induce indirect effects by the generation of reactive oxygen species (ROS) and other free radicals (Vincent, Neale 2000; Rastogi et al. 2010; Rajneesh et al. 2019). These free oxygen radicals are formed as a normal part of the metabolism in photosynthetic organisms. However, stress conditions such as UV-B are reported to result in excessive production of ROS, resulting in damage to lipids, proteins, DNA and other biomolecules (Bhandari, Sharma 2007), affecting vital functions of the cells such as growth, enzyme activity, pigmentation, photosynthesis, nitrogen fixation and assimilation (Sinha, Häder 2000).

Cyanobacteria are responsible for a large part of global photosynthetic productivity and the carbon sink, which may be affected by changing environmental conditions in the atmosphere with impact on the aquatic species and ecosystems (Olsson-Francis et al. 2013). It is also essential that we understand the mechanism of damage and protection in aquatic systems under a higher UV-B radiation level on various damaging, protective and adaptational processes in freshwater cyanobacteria. One of the mechanisms of tolerance and adaptation against abiotic stress is the synthesis of UV-B absorbing compounds. Several UV-B absorbing compounds identified in cyanobacteria are induced by UV radiation, and their role as photoprotectants is recognized (Bultel-Ponce et al. 2004). Mycosporine-like amino acids (MAAs) and scytonemin are induced due to UV-B exposure and are capable of acting as

photoprotective substances (Gacesa et al. 2018; Lawrence et al. 2018; Pandey et al. 2020).

In this work, we investigated the effect of UV-B and UV-B radiation supplemented with low photosynthetically active radiation (PAR) on photosynthesis, photosynthetic pigments, lipid peroxidation, and fatty acids in a freshwater cyanobacterium, *Scytonema hofmannii*. In addition, the effects of both treatments on the defense mechanism, such as induction of UV-absorbing compounds like MAAs and scytonemin, was also studied. The work has relevance to agriculture and the environment and also may result in a commercial application. The work is important to understand the algal response to changing environmental conditions, mainly UV-B, and predict future consequences.

Materials and methods

Materials and culture conditions

The cyanobacterial culture *Scytonema hofmannii* Agardh ex Bornet et Flahault was collected from rice fields of a freshwater ecosystem. Cultures were isolated and routinely grown in autoclaved liquid culture medium BG 11 according to Rippka et al. (1979). Cultures were maintained in 100 mL conical flasks filled to 40% of their volume and kept on a shaker set to a temperature of 30 ± 2 °C under cool white fluorescent tubes providing approximately 80 μmol m^{-2} s^{-1} photosynthetic active radiation (PAR) at the culture level with a 12 h of photoperiod. *Scytonema* was allowed to grow for 25 days to obtain their respective balanced phase of growth.

Experimental conditions

The UV-B source (T-6M source with a λ max at 312 nm, Vilbour-Lourmat, France) was fitted in a biological oxygen demand (BOD) chamber. A cyanobacterial culture in an open petri plate (without lid) was directly exposed to UV-B radiation (0.8 ± 0.1 mW cm^{-2}) from the top in the BOD chamber at 30 °C up to 6 h while keeping the algal culture continuously stirred using a small magnetic flea to avoid the shading of the culture. UV-B radiation was measured using a UV-B radiometer procured from the same supplier (VLX-312 with a λ max at 312 nm). For the other treatment, white light of 80 μmol photons m^{-2} s^{-1} PAR was supplemented using a light source with fiber optics at a 60° angle to the culture during the UV-B treatment. In nature, UV-B radiation is always present along with visible radiation, thus in this study we supplemented UV-B radiation with low intensity PAR.

Chlorophyll fluorescence measurement

The chlorophyll fluorescence (Fv/Fm ratio) was measured according to Sharma et al. (1998a) using a chlorophyll fluorometer PAM 101 with actinic light PAM 102 (Walz, Germany). The dark adapted culture was exposed to modulated light with an intensity of 3 to 4 μmol photons m^{-2} s^{-1} to measure initial fluorescence (F0). This was followed by exposure to a saturating pulse of white light of 4000 μmol photons m^{-2} s^{-1} to provide the maximum fluorescence (Fm). Variable fluorescence (Fv) was determined as Fv = Fm – F0 and the Fv/Fm ratio was calculated. Each experiment was repeated thrice and the data presented are with means and standard deviations.

Photosynthetic pigment analysis

Cyanobacterial cells were collected after centrifugation of a culture at 8000×g for 15 min and 0.1 g of cells was extracted in 1 mL 80% (v/v) methanol in a homogenizer at 4 °C under dim light, followed by centrifugation at 6000×g for 10 min at 4 °C. The pigments were separated by HPLC according to Sharma and Hall (1996) using reverse phase column (Waters Spherisorb ODS 25 μm × 4.6 mm × 250 mm) and a photodiode-array detection detector (Waters 2996). The quantity of pigments was calculated from peak area value using β-carotene as an external standard. Identification of pigments was carried out using the retention time against standards and using the spectral profile of individual peaks using a PDA detector in the range of 400 to 700 nm. Each experiment was repeated thrice and the data presented are using means and standard deviations (SD).

Phycobilisome analysis

Cell samples were concentrated by centrifugation for 15 min at 6000×g and 0.1 g of the pellet was resuspended in 5 mL 20 mM sodium acetate buffer, pH 5.5. Cells were broken using a sonicator (Bandelin UW 2200, Germany) at 50% power with nine cycles for 1 min. Phycobilisomes were precipitated by incubating with 1% streptomycin sulfate (w/v) for 30 min at 4 °C and collected by centrifugation at 8000×g for 30 min at 4 °C. The amount of phycocyanin, allophycocyanin, and phycoerythrin were calculated according to Liotenberg et al. (1996). Each experiment was repeated thrice and the data presented are using means and standard deviations.

Measurement of reactive oxygen species

Production of reactive oxygen species (ROS) was determined using the epinephrine method (Boveris 1984). Epinephrine (4.5 mM) was added to the buffer containing 6 mM ethylenediaminetetraacetic acid in 10 mM sodium carbonate (pH 10.2) with or without algal tissue. This resulted in the production of ROS, which was determined by enzyme kinetics at 480 nm using a double beam spectrophotometer (Shimadzu, UV-2450). The experiment was replicated thrice and the data presented are using means and standard deviation.

Lipid peroxidation analysis

Lipid peroxidation was determined by the production of thiobarbutaric acid (TBA)-malonaldehyde adduct formation according to the method described by Sharma et

al. (1998b). The algal culture was harvested by centrifuging at 8000×g for 15 min. The algal pellet was homogenized in a tissue homogenizer and redissolved in a fresh culture medium with a ratio of 1:5 (w/v). The resuspended algal culture (5 mL) was again centrifuged and the algal pellet was homogenized in 0.5% trichloroacetic acid (TCA). The supernatant was collected and used for measuring the peroxidation of membrane lipids. One milliliter of the supernatant was added to a test tube containing 2.5 mL freshly prepared (0.5%) TBA in (20%) TCA and allowed to incubate for 30 min at 90 °C in a water bath. The optical density was taken at 532 nm (Schimadzu, UV-250). Peroxidation of lipids was measured using an extinction coefficient of 155 mM^{-1} cm^{-1}. This experiment was repeated thrice and the data presented are using means and standard deviations.

Extraction of lipids

Total lipids were extracted according to Turnham and Northcote (1984) with modifications (Bhandari, Sharma 2011). The freshly harvested algal pellet was homogenized in chloroform/methanol (1:2, v/v) containing 0.01% butylated hydroxytoluene as an antioxidant agent to make a final volume of 15 mL. The lipid extract was centrifuged for 5 min at 2000×g to get rid of cell debris, and to the supernatant, 0.8 mL distilled water was added followed by 5 mL chloroform and 5 mL of 0.88% potassium chloride in a separating funnel to make a ratio of chloroform/methanol/water 1:1:0.9. The mixture was shaken vigorously for 5 min and allowed to separate for 30 min. The solvent phase was collected and used for fatty acid analysis.

Esterification of fatty acids

The fatty acid methyl esters were prepared for gas chromatography analysis according to Christie (1982). Methyl esters of fatty acids were run on a Schimadzu gas chromatograph equipped with a flame ionization detector and chromatopack data processor. Fatty acid methyl esters peaks were identified by comparing their retention times with methyl esters of pure fatty acid standards and were quantified by using the peak areas to give the mole % of different fatty acids directly. Each experiment was repeated thrice and the data presented are using means and standard deviations.

Extraction and separation of MAAs

Extraction and purification of MAAs were carried out according to the method of Sinha and Häder (2000) using HPLC (Waters, 2996). Cyanobacterial cells (0.1 g) were homogenized using 20% (v/v) aqueous methanol (HPLC grade) and incubated at 45 °C for 2.5 h. The pellet was removed and the supernatant was evaporated to dryness. The dried supernatant was dissolved in 0.2% acetic acid and analyzed using the HPLC (Waters Spherisorb) detection program (Waters 2996 PDA detector) at 280 nm with a flow rate of 1.0 mL min^{-1} using an isocratic mobile phase of 0.2% acetic acid. Spectra of MAAs were measured using a PDA detector (Waters, 2996) at a wavelength range of 200 to 700 nm. This experiment was repeated thrice and the data are presented using means and standard deviations.

Extraction and separation of scytonemin

The cyanobacteria were homogenized in 100% acetone and kept for extraction overnight in a refrigerator at 4 °C. The resulting suspension was centrifuged and the supernatant was filtered through a 0.2 μm pore filter. Further analysis of the samples was done by HPLC (Waters) according to Sinha et al. (1999). The wavelength range for detection was 240 to 500 nm with a flow rate of 1.5 mL min^{-1}, and a mobile phase of solvent A (double distilled water) and solvent B (acetonitrile/methanol/tetrahydrofuran, 75:15:10, v/v/v) with a 0 to 3 min linear increase from 30% solvent B to 3 – 23 min at 100% solvent B. Each experiment was repeated thrice and the data are presented using means and standard deviations.

Results

Effect of UV-B and UV-B supplemented with PAR on chlorophyll fluorescence was studied in *Scytonema hofmannii* by measuring the Fv/Fm ratio (Fig. 1), There was a decrease in the Fv/Fm ratio in response to the increased duration of both UV-B and UV-B supplemented with PAR treatment. A 6 h of UV-B treatment resulted in a 70% decrease in the Fv/Fm ratio, while a 77% decrease in the Fv/Fm ratio was observed due to UV-B supplemented with PAR. F0 level was also decreased due to the treatment in the species studied.

HPLC data showed significant qualitative and quantitative changes in the pigments in *S. hofmannii* (Table 1). The photosynthetic pigments observed were chlorophyll *a*, β-carotene, alloxanthin, and myxoxanthophylls. UV-B alone and UV-B supplemented

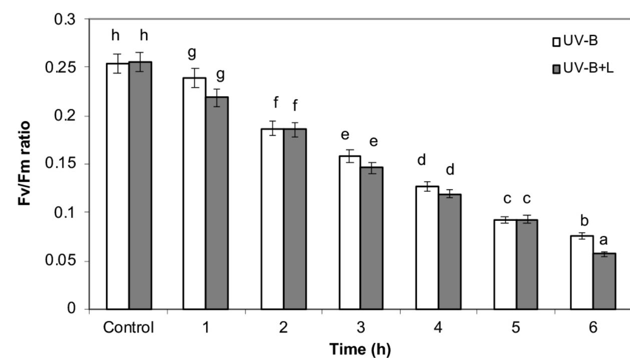

Fig. 1. Effect of UV-B and UV-B supplemented with low PAR up to 6 h on chlorophyll fluorescence (Fv/Fm ratio) in *Scytonema hofmannii*. Each bar represents the mean ± SD, n = 3. Means with identical letters are not statistically significantly different (p < 0.05).

Table 1. Effect of UV-B and UV-B supplemented with low PAR on concentration of photosynthetic pigments (μg g^{-1} FM) in *Scytonema hofmannii*. Quantitation of the pigments is based on the peak area. Means with identical letters within the columns are not statistically significantly different ($p < 0.05$)

Pigment	Control	UV-B	UV-B + L
Carotenoids	0.531 ± 0.004 c	0.320 ± 0.005 b	0.284 ± 0.004 a
Chlorophyll *a*	0.778 ± 0.003 c	0.200 ± 0.004 a	0.490 ± 0.006 b
Phycobilins	0.898 ± 0.006 c	0.636 ± 0.004 b	0.358 ± 0.005 a
β-carotene	0.551 ± 0.004 b	0.087 ± 0.005 a	0.465 ± 0.004 b

with low-intensity PAR treatments resulted in an overall decrease in pigment concentration. As a result of 6 h of UV-B treatment, chlorophyll a concentration decreased by 54%, myxoxanthophylls decreased by 55%, and β-carotene decreased by 88% compared to the control. UV-B radiation supplemented with low-intensity PAR showed a greater decline in all pigments than for UV-B radiation alone (Table 1).

After UV-B treatment there was a decline in phycocyanin, allophycocyanin, and phycoerythrin concentration, but the decrease of allophycocyanin concentration was comparatively less than for phycocyanin and phycoerythrin (Fig. 2). Phycocyanin concentration declined linearly to 64% after 6 h of the treatment as compared to the control. However, allophycocyanin declined to a relatively lesser extent of 53% after 6 h of UV-B treatment as compared to the control. Phycoerythrin declined even more to 55% after 6 h of treatment compared to the control (Fig. 2). UV-B radiation supplemented with a low level of PAR also showed a linear decline in content of all three pigments.

The effect of UV-B radiation and UV-B supplemented with low PAR in *S. hofmannii* showed an increase in ROS production. UV-B alone treatment increased the ROS production by 10%, whereas UV-B supplemented with low PAR increased the level of ROS generation to 34% compared to their control (Fig. 3).

The effect of UV-B radiation and UV-B supplemented with low visual radiation on MDA formation, which is indicative of the level of peroxidation of cell membrane lipids, was also studied (Fig. 4). In *S. hofmannii*, treatments to UV-B and UV-B supplemented with low PAR increased the level of lipid peroxidation. Six hours of UV-B treatment to *Scytonema* showed a 115 % increase in MDA formation as compared to the control. However, six-hour treatment with UV-B supplemented with low PAR treatment caused a higher level of peroxidation than seen in UV-B treatment alone (Fig. 4).

The fatty acid groups present in *S. hofmannii* were lauric acid C12, myristic acid C14, palmitic acid C16, stearic acid C18, oleic acid C18:1, linoleic acid C18:2 and linolenic acid C18:3 (Table 2). UV-B and UV-B supplemented with low PAR treatment did not show any appreciable changes in the saturated fatty acid concentration. However, concentration of unsaturated fatty acids such as oleic acid increased to 35% while linoleic acid and linolenic acid increased by 13 and 51%, respectively, as compared to control.

HPLC analysis in our study showed a significant presence of MAAs (shinorine) with an absorption maximum of 294 nm (Table 3). Exposure of UV-B radiation for 6 h resulted in a 130% increase in MAA compared to the control, while UV-B radiation supplemented with low-intensity PAR resulted in a further increase in the MAA concentration to 146% as compared to the control.

Scytonemin was determined with an absorption

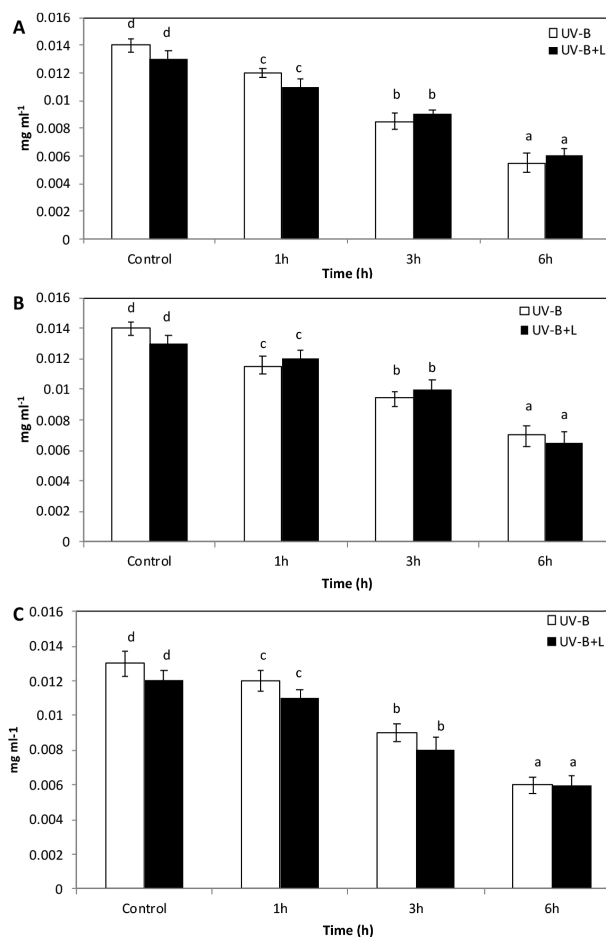

Fig. 2. Effect of UV-B and UV-B supplemented with low PAR up to 6 h on concentration of phycocyanin (A), allophycocyanin (B), phycoerythrin (C) in *Scytonema hofmannii*. Each bar represents the mean ± SD, $n = 3$. Means with identical letters are not statistically significantly different ($p < 0.05$).

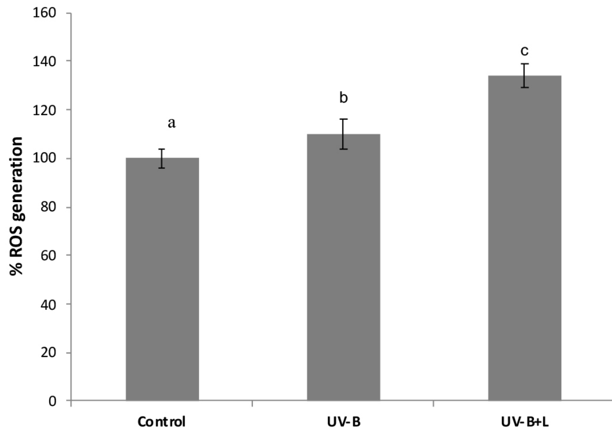

Fig. 3. Effect of UV-B and UV-B supplemented with low PAR on ROS generation in *Scytonema hofmannii*. Each bar represents the mean ± SD, $n = 3$. Means with identical letters are not statistically significantly different ($p < 0.05$).

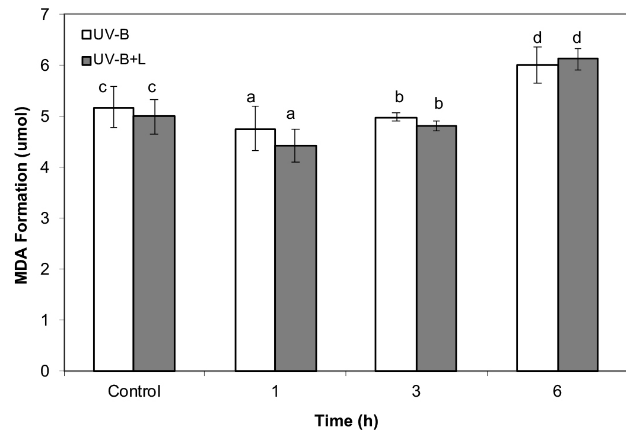

Fig. 4. Effect of UV-B and UV-B supplemented with low PAR up to 6 h on lipid peroxidation in *Scytonema hofmannii*. Each bar represents the mean ± SD, $n = 3$. Means with identical letters are not statistically significantly different ($p < 0.05$).

maximum of λ max at 237, 273, and 334 nm (Table 3). A six-hour exposure to UV-B radiation resulted in a 118% increase in scytonemin concentration as compared to the control. UV-B radiation supplemented with low-intensity PAR resulted in a further increase in the scytonemin concentration to 127% as compared to the control.

Discussion

The observed decrease in Fv/Fm ratio due to UV-B and UV-B supplemented with low PAR treatment (Fig. 1), was primarily due to the decrease in the F0, indicative of a decrease in the excitation energy reaching the photosynthetic reaction centre II due to loss of pigments in the light-harvesting complex of photosystem II, as well as due to the decrease in the Fm value, indicating damage to the photosystem II reaction centre itself under the UV-B treatment. The decrease in Fv/Fm ratio caused by irradiation was linearly related to the decrease in photosynthesis optimal quantum yield (Bhandari, Sharma 2007). A decrease in photosynthetic activity during UV-B exposure may be due to direct damage to critical components within the D1 protein of photosystem II (Xue et al. 2005). A drastic loss of photosynthetic thylakoid membranes and metabolic-related structures, such as accessory pigments and polyphosphate granules of *Cylindrospermopsis raciborskii* was observed to be induced by various UV radiation fractions (Noyma et al. 2015).

The decrease observed in photosynthetic pigments due to UV-B treatment in our study is suggested to be due to the generation of reactive oxygen species (Table 1), which may oxidize the pigment content, as pigments are highly sensitive to oxidation and peroxidation reactions (Sharma 2002). The chromopheric compounds involved in photosynthesis, such as chlorophyll, phycobiliproteins, and quinones, absorb UV-B radiation and photosensitize ROS generation. There are reports of a decrease in photosynthetic pigments due to the UV-B exposure to algae (Bischof et al. 2000; Xue et al. 2005; Kannaujiya, Sinha 2017). It has been also reported that bleaching of photosynthetic pigments and simultaneously inhibition of the photosynthetic processes can occur in cyanobacteria (Simioni et al. 2014; Ganapathy et al. 2015).

Quantitative decrease in the phycobilisome concentration due to UV-B and combination of UV-B and PAR treatment in the cyanobacterium in our study may also be due to bleaching of accessory light-harvesting pigments (Fig. 2). It has been also reported that exposure to UV-B radiation resulted in a decrease in phycobilins with a concurrent loss of energy transfer from phycobilisomes to the photosystem (Zhou et al. 2005). Dissociation of phycobilisomes from thylakoid membranes when *Synechococcus* sp. was exposed to UVA + UVB + PAR was reported (Six et al. 2007). Similar

Table 2. Effect of UV-B and UV-B supplemented with low PAR on fatty acid composition of total lipids in *Scytonema hofmannii* (mol %). Means with identical letters within the columns are not statistically significantly different ($p < 0.05$)

Treatment	Fatty acid composition (mol %)						
	Lauric acid 12:0	Myristic acid 14:0	Palmatic acid 16:0	Stearic acid 18:0	Oleic acid 18:1	Linoleic acid 18:2	Linolenic acid 18:3
Control	11.30 ± 0.06 a	17.60 ± 0.04 a	14.80 ± 0.06 a	9.40 ± 0.04 a	3.20 ± 0.06 a	19.90 ±0.06 a	3.30 ± 0.03 a
UV-B (6 h)	11.30 ± 0.04 a	17.60 ± 0.05 a	14.80 ± 0.07 a	9.10 ± 0.06 a	3.20 ± 0.05 a	19.90 ±0.04a	3.30 ± 0.05 a
UV-B + L (6 h)	11.40 ± 0.07 a	17.70 ± 0.07 a	14.90 ± 0.05 a	9.40 ± 0.05 a	3.60 ± 0.05 a	21.30 ±0.03 b	4.30 ± 0.02 b

Table 3. Effect of UV-B and UV-B supplemented with low PAR on concentration of MAAs and scytonemin (μg g^{-1} FM) in *Scytonema hofmannii*. Quantitation is based on the peak area. Means with identical letters within the columns are not statistically significantly different ($p < 0.05$)

Treatment	MAAs	Scytonemin
Control	0.400 ± 0.003 a	0.611 ± 0.005 a
UV-B	0.523 ± 0.005 b	0.725 ± 0.004 b
UV-B + L	0.585 ± 0.005 b	0.778 ± 0.004 b

effects about phycoerythrin and allophycocyanin were found (Dohler et al. 1995; Gerber, Häder 1995). The effects of UV-B on the red macroalgae *Gracilaria domingensis* included the destruction of chloroplasts, internal organization, the disappearance of phycobilisomes, and decrease in the concentration of photosynthetic pigments, including chlorophyll *a*, phycoerythrin, phycocyanin and allophycocyanin (Schmidt et al. 2010).

Our data on ROS production resulting from the treatments is also substantiated by our results on oxidative damage to lipids, observed as lipid peroxidation in *S. hofmannii*. Although ROS production was higher in the UV-B treatment, MDA production was less in UV-B supplemented with PAR treatment (Fig. 3 & 4), as low PAR and UV-B help in the activation of protective antioxidant enzymes (Bhandari, Sharma 2011). The rich presence of photosynthetic pigments and redox components such as chlorophylls, phycobiliproteins, and quinones, which exhibit absorption in the UV range and photosensitize the formation of ROS, including superoxide radical, hydroxyl radical, hydrogen peroxide, and singlet oxygen, by electron or energy transfer to oxygen, tend to exert oxidative damage, inducing lipid peroxidation, DNA damage, inhibition of photosynthesis and bleaching of photosynthetic pigments such as chlorophylls and phycobilins. Under UV-B stress, the inhibition of the electron transport chain due to the degradation of the D1 protein in photosystem II might promote energy transfer from triplet chlorophylls to oxygen to form singlet oxygen by UV irradiation and promote the formation of superoxide radicals at the level of ferredoxin in photosystem I (Bischof et al. 2000; Vega, Pizarro 2000). Shorter wavelengths of both the UV-A and the UV-B regions were more effective in generating ROS than their higher wavelengths and photosynthetic active radiation (Singh et al. 2014).

Our experiments showed that UV-B exposure did not result in any changes in the saturated fatty acid concentration of total lipids (Table 2), thus changing the ratio of unsaturation to the saturation level of fatty acid in favor of unsaturation, which is required for adaptation. The changes in the membrane lipid profile could be due to ROS (He et al. 2002). An increase in the unsaturation level of fatty acids due to UV-B supplemented with low PAR could be due to an increase in desaturase level activity in *Spirulina platensis* (Funtue et al. 1997). The fluidity of membrane lipids is directly correlated with the level of unsaturated fatty acids in biological membranes. The increase in the unsaturation of fatty acids in membrane lipids may help provide protection/adaptation to cyanobacteria against UV-B radiation. An increase in membrane lipid unsaturation enhanced the tolerance of cyanobacterium to abiotic stress (Gombos et al. 1997).

Organisms synthesize UV absorbing or screening compounds such as mycosporine-like amino acids, scytonemin, and phenolic compounds to protect themselves from the UV-B radiation (Karsten et al. 1998), the production of the enzymatic and non-enzymatic antioxidative mechanism responsible for scavenging of reactive oxygen species (Sharma et al. 1998b) and repair of UV-induced damage of DNA by photoreactivation and excision repair (Häder 2001).

The present study demonstrated that cyanobacteria can synthesize mycosporine-like amino acids (MAAs) and scytonemin to protect against UV-B radiation (Table 3). Compounds like MAAs and scytonemin are known to have UV-B absorbing properties, thereby protecting sensitive sites against UV-B radiation. In our study, the accumulation of scytonemin (an extracellular sheath pigment) prevents penetration of UV-B radiation in the cell, representing an adaptive benefit against the harmful effects of UV radiation. Scytonemin is a highly stable compound and prevents up to 90% of solar UV radiation from entering the cell (Singh et al. 2010). On the other hand, MAAs is produced in the cytoplasm and helps in UV absorption of whatever UV-B radiation penetrates the cell even after scytonemin. MAAs contain a central cyclohexanone or cyclohexenimine ring responsible for UV absorption within the cell (Llewellyn, Airs 2010; Wada et al. 2015). Thus, both extracellular scytonemin and intracellular MAAs have a significant effect in preventing UV radiation from reaching prospective targets like DNA and proteins in living cells (Fleming, Castenholz 2007). Several authors have reported the synthesis of UV-B absorbing MAAs and scytonemin due to UV-B exposure (Fleming, Castenholz 2008; Mushir, Fatma 2012; Rastogi, Incharoensakdi 2014; Pathak et al. 2017). The concentration of UV-absorbing substances such as MAAs can be correlated with various factors such as duration and irradiance quantity (Post, Larkum 1993). However, when UV-B was supplemented with low-level PAR, it caused a far greater increase in the MAAs (Bhandari, Sharma 2011).

Conclusions

UV-B and UV-B supplemented with low PAR exposure caused a damaging effect on various parameters such as photosynthesis, photosynthetic pigments, peroxidation of cell membrane lipids, and fatty acids, affecting photosynthesis, and loss of productivity in *Scytonema*

hofmannii. The observed effects may be consequences of the generation of reactive oxygen species due to the treatments. The results indicated the oxidative nature of damage to cyanobacteria under UV-B conditions indicated by the peroxidation of lipids. The resultant oxidative damage leads to the decreased survival of the test organisms. There was no significant change in the saturation to unsaturation level of fatty acids. We observed some of the protective and adaptive cyanobacteria processes due to UV-B and UV-B supplemented with low PAR, such as the synthesis of UV-B absorbing compounds like mycosporine-like amino acids (MAAs) and scytonemin. Changes in some of the parameters such as photosynthesis, pigments, peroxidation, etc., may be a consequence of the damage, while changes in fatty acids may be a result of the adaptation process. Still, further changes in some other parameters such as MAAs and scytonemin may be to protect the harmful effect of UV-B. The findings have significance in increasing the MAAs and scytonemin production manifold by UV-B treatment for commercial applications.

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Received 1 May 2021; received in revised form 26 May 2021; accepted 10 June 2021