Available online at www.sciencedirect.com
ScienceDirect
Environmental and Experimental Botany 63 (2008) 123–129
Environmental and Experimental Botany
www.elsevier.com/locate/envexpbot

Photosynthesis and growth of *Arthrospira* (*Spirulina*) *platensis* (Cyanophyta) in response to solar UV radiation, with special reference to its minor variant

Kunshan Gao [a,b,c,*], Zengling Ma [a]

[a] *Marine Biology Institute, Shantou University, Shantou, Guangdong 515063, China*
[b] *State Key Laboratory of Marine Environmental Science, Xiamen University, Xiamen 361005, China*
[c] *State Key Laboratory of Freshwater Ecology and Biotechnology, Institute of Hydrobiology, The Chinese Academy of Sciences, Wuhan, Hubei 430072, China*

Received 26 January 2007; received in revised form 3 October 2007; accepted 20 October 2007

Abstract

The minor variant of the economically important cyanobacterium, *Arthrospira platensis*, usually appears in commercial production ponds under solar radiation. However, how sensitive the minor variant to solar UVR and whether its occurrence relates to the solar exposures are not known. We investigated the photochemical efficiency of PSII and growth rate of D-0083 strain and its minor variant in semi-continuous cultures under PAR (400–700 nm) alone, PAR + UV-A (320–400 nm) and PAR + UV-A + UV-B (280–700 nm) of solar radiation. The effective quantum yield of D-0083 at 14:00 p.m. decreased by about 86% under PAR, 87% under PAR + UV-A and 92% under PAR + UV-A + UV-B (280–315 nm), respectively. That of the minor variant was reduced by 93% under PAR and to undetectable values in the presence of UV-A or UV-A + UV-B. Diurnal change of the yield showed constant pattern during long-term (10 days) exposures, high in the early morning and late afternoon but the lowest at noontime in both strains, with the UVR-related inhibition being always higher in the variant than D-0083. During the long-term exposures, cells of D-0083 acclimated faster to solar UV radiation and showed paralleled growth rates among the treatments with or without UVR at the end of the experiment; however, growth of the minor variant was significantly reduced by UV-A and UV-B throughout the period. Comparing to the major strain D-0083, the minor variant was more sensitive to UVR in terms of its growth, quantum yield and acclimation to solar radiation.

Keywords: *Arthrospira platensis*; Minor variant; Carotenoids; Effective quantum yield; Specific growth rate; UV-absorbing compounds; UVR

1. Introduction

Arthrospira spp. are photoautotrophic filamentous cyanobacteria which have been widely exploited as a source of human health food (Ciferri, 1983; Mosulishvili et al., 2002), animal feed (Lu et al., 2002) and cosmetic colorants (Vonshak, 1990). Optimizations of its growing conditions for massive production have been focused on the improvement of production efficiency and the reduction of costs (Jensen and Knutsen, 1993; Torzillo et al., 1998; Wu et al., 2005a; Ma et al., 2006). Its productivity in commercially operated ponds depends on a number of environmental parameters. Light is the most important factor that governs its photosynthesis and growth (Vonshak, 1997). Photosynthetic characteristics of *A. platensis* are usually dependent on growth conditions, with the cells grown under higher levels of PAR being less susceptible to photoinhibition (Vonshak et al., 1983); those grown at suboptimal temperature were more sensitive to photoinhibition than those grown at the optimal temperature (Jensen and Knutsen, 1993). A relatively low morning temperature with rapid increase in light intensity could induce photoinhibitory stress, and heating the cultures significantly reduced the inhibitory effect (Vonshak et al., 1994).

Solar UV radiation (UVR, 280–400 nm) is an important environmental factor that affects *Arthrospira* in natural environment as well as in outdoor commercial production ponds. It was found recently that the spiral structure of *A. platensis* filament was broken under solar radiation (PAR + UVR) on sunny days (Wu et al., 2005b), and its photosynthetic performance was negatively affected by UVR (Rajagopal et al., 2000; Wu et al., 2005b).

* Corresponding author at: Marine Biology Institute, Shantou University, Shantou, Guangdong 515063, China. Tel.: +86 754 2903280; fax: +86 754 2903977.
E-mail address: ksgao@stu.edu.cn (K. Gao).

0098-8472/$ – see front matter © 2007 Elsevier B.V. All rights reserved.
doi:10.1016/j.envexpbot.2007.10.031

Abnormal morphologies, such as *Arthrospira* minor (i.e. minor variant, with the trichome length and helix diameter are much less than the normal filaments) and even linear strains occurred in laboratory and commercial cultures (Jeeji Bai, 1985; Wang and Zhao, 2005). Minor variant of *A. platensis* D-0083 has been found in the production ponds, though only the normal length strain was inoculated at the initiation of production. Minor variants of *Arthrospira* spp. are notorious for reducing efficiency of harvest and production (Vonshak and Richmond, 1988; Belay, 1997). The spiral structure of *A. platensis* has been previously found to be broken and altered by solar UVR (Wu et al., 2005b). However, how sensitive the minor variant is to solar UVR and whether its occurrence relates to the solar exposures and the consequent spiral breakage are not known. In addition, little has been documented about the biological characteristics of the minor variant. The aim of the present work was to assess the sensitivity of *A. platensis* and its minor variant to solar UVR and to characterize their physiological behavior under outdoor conditions.

2. Materials and methods

2.1. Organisms

A. platensis D-0083, 280 μm (±30, $n = 100$) long with compressed spirals, was obtained from Hainan DIC microalgae Co. Ltd., Hainan, China. The minor variant (72 ± 7 μm, $n = 100$), about 1/4 length of D-0083 with shorter helix and width, was isolated from *A. platensis* D-0083 production ponds in April, 2003, and its clonal culture was maintained indoor since then. Both strains were cultured under indoor conditions with aerated Zarrouk's medium at 30 °C and 100 μmol photons m^{-2} s^{-1} of cool-white fluorescent light (14L:10D) and sustained stable morphological features before the experiments.

2.2. Culture conditions

In order to eliminate the effects of nutrition limitation and self-shading, semi-continuous cultures (Zarrouk's medium, optical cell density at 560 nm is 0.3) were adopted in our experiments. The culture media were partially renewed (30–70% according to the actual optical cell density) every other day to maintain a constant biomass density (0.33 g dry wt L^{-1}) after dilution. Quartz tubes (Φ 4 cm, 40 cm long) were used for outdoor cultures to allow full spectrum of solar radiation to penetrate. The cultures were aerated (0.4 L min^{-1}) with ambient air and maintained in a flow-through water bath for temperature control within a range of 24–28 °C during the experimental period from 29 October to 7 November 2004.

2.3. Solar radiation measurements and treatments

Incident solar UV-B (280–315 nm), UV-A (315–400 nm) and PAR (400–700 nm) radiations were continuously monitored using a broadband filter radiometer (Eldonet, Real Time Computer Inc., Germany) permanently installed on the roof at Shantou University (116.6°E, 23.3°N). The reliability of this instrument has been internationally recognized (Häder et al., 1999; Korbee-Peinado et al., 2004), and certificated with the correspondence error less than 0.5% in comparison with the most accurate instrument (certificate No. 2006/BB14/1). Three different radiation treatments were implemented: (1) cells receiving full solar radiation (PAB treatment; 295–700 nm)—quartz tubes covered with Ultraphan 295 (Ultraphan, Digefra, Munich, Germany); (2) cells receiving UV-A and PAR (PA treatment; 320–700 nm)—quartz tubes covered with Folex 320 (Montagefolie, No. 10155099, Folex, Dreieich, Germany); (3) cells receiving only PAR (P treatment; 400–700 nm)—quartz tubes covered with Ultraphan film 395 (UV Opak, Digefra, Munich, Germany). There was a 5 nm difference between the measured and exposed UV-A irradiance, which gives about 2% higher percent of UV-A than that the cells were actually exposed to. The cut-off foils reflected about 4% of the solar PAR under water (Gao et al., 2007a), however, such a reflection was identical for the foils used in the PAB, PA and P treatments, therefore, it cannot affect the comparison among them.

2.4. Determination of biomass density and specific growth rate

Biomass density of the culture was measured by filtering 30 mL of the cultures on a pre-dried Whatman GF/F glass fiber filter (diameter 25 mm), drying in an oven at 80 °C for 24 h, and cooling off in a desiccator, weighing on an electronic balance and subtracting the known weight of the dried filter. While the sample was being filtered it was washed with 20 mL acidified distilled water (pH 4) in order to remove residual salts. Specific growth rate (μ, day) was calculated as $\mu = (\ln x_2 - \ln x_1)/\Delta t$, where x_1 and x_2 represent the initial biomass density and that after Δt time, Δt is the time interval (2 days) between two dilutions.

2.5. Light-adapted fluorescence measurement

Effective quantum yield ($\Delta F/F'_m$), representative of the apparent photochemical efficiency of PSII (Schreiber et al., 1995; Aguirre-von-Wobeser et al., 2000), was monitored with a portable pulse amplitude modulated fluorometer (WATER-ED, Walz, Effeltrich, Germany) for the two strains while being exposed to different solar radiation treatments. ΔF (variable fluorescence for light-adapted state) was defined as $F'_m - F'_t$, F'_t and F'_m were the current steady-state and maximal fluorescence for the light-adapted samples. A decline in $\Delta F/F'_m$ values as a result of elevated irradiance or UV radiation were considered as a photoinhibition of PSII (Kolber and Falkowski, 1993).

2.6. UV-absorbance spectrum and photosynthetic pigments contents

UV-absorbance spectrum, Chl *a* and carotenoids contents were determined by filtering (Whatman GF/C, 25 mm) 10 mL culture, resuspending in 10 mL absolute methanol overnight, centrifuging at 5000 × *g* for 5 min and measuring the absorption spectrum with a scanning spectrophotometer (DU-530, Beck-

man, USA). Chl *a* concentration was calculated according to Porra (2002), and the concentration of carotenoids was determined using the equation of Parsons and Strickland (1963).

2.7. Assessment of inhibition on growth and effective quantum yield

Inhibition induced by UVR was estimated as follows: $(P_P - P_{PAB})/P_P \times 100\%$, where P_P and P_{PAB} represent the specific growth rates or the quantum yields of the samples treated with solar radiation screened off UVR (P_P) or with UVR (P_{PAB}), respectively.

2.8. Statistical analysis

All data were obtained from at least three replicates. The variance was analyzed and statistical significance was established with Tukey test at 0.05 level using SPSS for Windows (SPSS 12.0, USA).

3. Results

When *A. platensis* D-0083 and the minor variant were exposed to solar radiation, during the short-term (from 8:00 to 16:00, 29 October 2004) experiments, the effective quantum yield decreased straightly under PAR alone treatment from the morning to early afternoon (14:00) for both strains (Fig. 1). Solar PAR alone decreased the yield to the lowest level at 14:00 respectively by 86% in D-0083 and by 93% in the minor variant compared to their initial values. Presence of UV-A as well as UV-B caused further reduction of the yield by 1% (UV-A) and by 5% (UV-B) in D-0083 and to an undetectable level in the minor variant at 14:00. Existence of UV-B (PAR + UV-A + UV-B) resulted in significant reduction ($p < 0.01$) in the yield compared to PAR + UV-A or PAR alone at all the times of the measurement except at 16:00 in D-0083. In the late afternoon at 16:00, the yield recovered for both strain under all the treatments. At this point, UV-A still resulted in significantly ($p < 0.01$) lower yield, while presence of UV-B did not result in further reduction in D-0083 (Fig. 1A). In contrast, both UV-A and UV-B resulted in significant ($p < 0.01$) reduction of the yield in the minor variant even in the late afternoon (Fig. 1B).

We carried on the outdoor cultures for a long-term period of 10 days (29 October–7 November 2004) to see the strains' acclimation to solar radiation (Fig. 2). Over such a period, the effective quantum yield in both strains was higher in the morning and afternoon and the lowest at noontime, following the irradiation regime. Regardless of diurnal fluctuation, the effective quantum yield increased with extended exposures to solar radiation. Under full spectrum, 6 days later, the yield increased to 77% and 48% (8:00), 33% and 25% (12:00), and 68% and 59% (16:00) of the initial values for the major and the minor strain, respectively. Filtering out of the UV-B or UV-A + UV-B resulted in higher yields than those under the PAB treatments, which was more obvious during the initial phase, and much more pronounced in the minor strain (Fig. 2). UVR-induced inhibition was lower in the morning and late afternoon and higher during

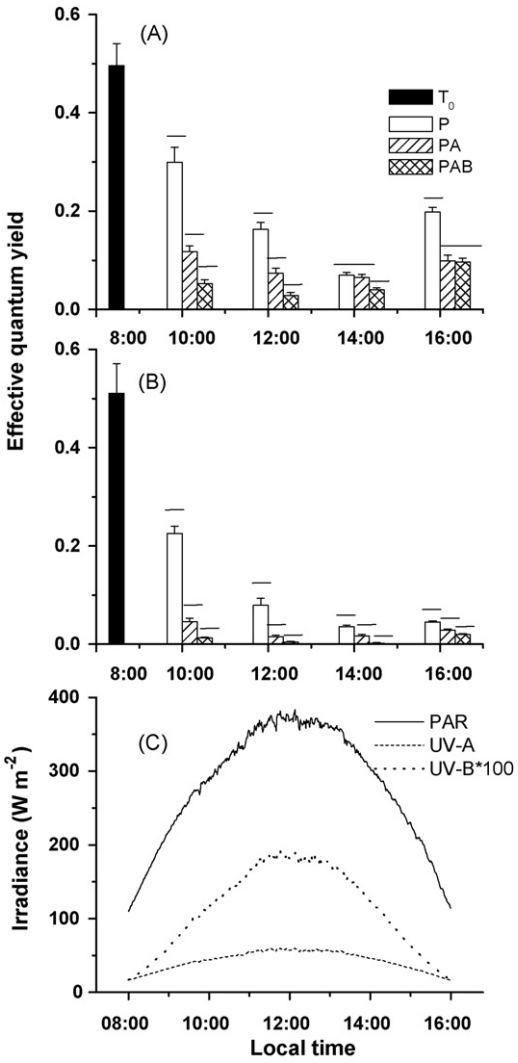

Fig. 1. Changes in the effective quantum yield ($\Delta F/F'_m$) of *Arthrospira platensis* D-0083 (A) and the minor variant (B) under PAB, PA and P treatments during the daytime on 29 October 2004. The black bar represents the value of cells before the exposure to solar radiation. Mean ± S.D. ($n = 3$). Horizontal lines at different heights indicate significant difference ($p < 0.05$).

the noontime throughout the period of exposure. It decreased in both strains, irrespective of the time of the day with prolonged exposure, reflecting an acclimation to the solar UVR (Fig. 3).

The specific growth rate increased while the cells acclimated from indoor light to outdoor solar radiation, while UVR-induced inhibition declined with time (Fig. 4). Specific growth rate of D-0083 decreased about 38%, 50% and 58% (compared to the maximal growth rate indoors) in 2 days since the cells were exposed to PAR, PAR + UV-A and PAR + UV-A + UV-B, respectively, and then increased with prolonged exposure. In D-0083, at the end of long-term cultures, no difference ($p > 0.05$) in the growth rate was observed under the three treatments (Fig. 4A); moreover, there was no significant difference ($p > 0.05$) between P and PA treatments throughout the experimental period. In the minor variant, exposures with UV-A and UV-B resulted in significant ($p < 0.01$) reduction of growth rate throughout the experimental period.

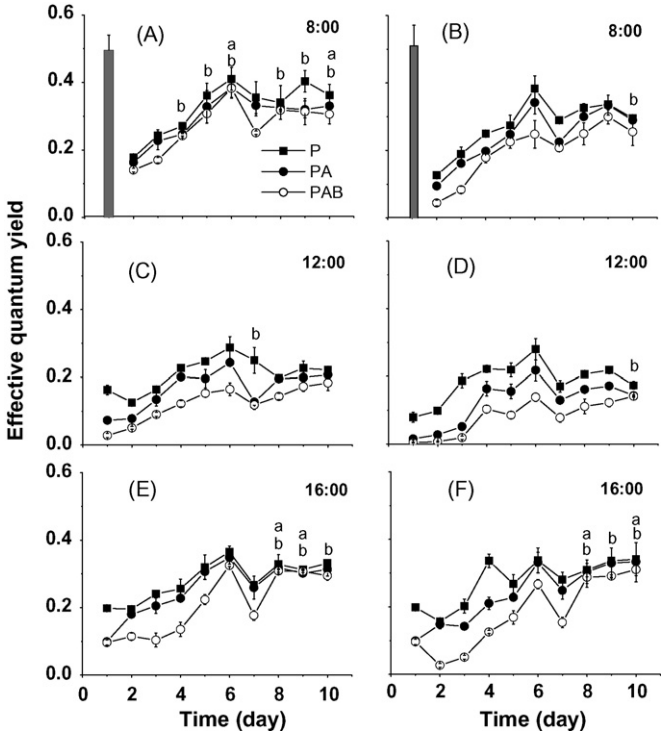

Fig. 2. Diurnal changes in the effective quantum yield ($\Delta F/F'_m$) of *A. platensis* D-0083 (A, C, E) and the minor variant (B, D, F) under PAB, PA and P treatments over a long-term period from 29 October to 7 November 2004. Bar represents the initial value before they were exposed to the different solar radiation treatments. Water temperature ranged from 24 to 28 °C during this period. Corresponding solar irradiance is shown in Fig. 4C. Mean ± S.D. ($n = 3$). Letter 'a' or 'b' denotes insignificant difference between P and PAB treatments or PA and PAB treatments at $p = 0.05$ level, respectively.

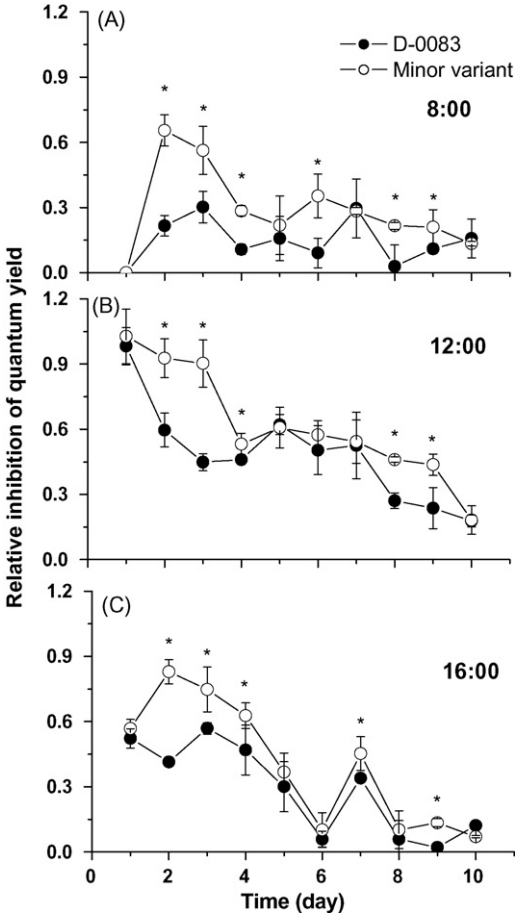

Fig. 3. UVR-induced inhibition of the effective quantum yield in *A. platensis* D-0083 and the minor variant. The inhibition was calculated as $(P_P - P_{PAB}) \times 100\%/P_P$, P_P and P_{PAB} present the photochemical activities of the samples treated with solar radiation screened off UVR (P_P) and with UVR (P_{PAB}). Mean ± S.D. ($n = 3$). The symbol (*) indicates that UVR-induced inhibition on the minor variant's yield are significantly different from that of D-0083 at $p = 0.05$ level.

Relative inhibition of growth due to UVR decreased (Fig. 5) as that of the effective quantum yield with prolonged exposure time, reflecting a consistency of PSII efficiency with growth during the acclimation. The highest inhibition of growth caused by UVR was 39% in D-0083 and 96% in the minor strain (Fig. 5), respectively. Such UVR-related inhibition of growth also decreased with time, to 5% in the major and 22% in the minor variant 10 days later, respectively, indicating more damage caused by UVR to the minor variant. The inhibition caused by UV-A was higher than by UV-B during the initial phase of the acclimation, but such difference decreased with time.

Few UV-absorbing compounds (absorbance between 310 and 360 nm) were found in the cells of the major and the minor strains even when exposed to full spectrum of solar irradiance (Fig. 6A and B). Chl *a* contents decreased with time during exposure to solar radiation (Fig. 6C and D), being much less under UVR + PAR than PAR alone. Contents of carotenoids in the major strain were persistent with relatively stable values except for the days 4th and 6th when they increased (Fig. 6E). In the minor variant, in contrast, the carotenoids increased under full spectrum of solar radiation but decreased under PAR alone (Fig. 6F). The ratio of carotenoids to Chl *a* increased with exposure time under both PAR and full spectrum of solar radiation (Fig. 6H and I) with a significant ($p < 0.01$) difference between P and PAB treatments in both strains.

4. Discussion

Previously, solar UVR was found to reduce photosynthetic O_2 evolution of *A. platensis* D-0083 (Wu et al., 2005b) under similar conditions as in the present study. Here, we demonstrated that presence of UV-A and UV-B resulted in significant inhibition of the effective quantum yield and growth of this species, though high levels of PAR caused most of the inhibition. The inhibition caused by high levels of PAR and UVR was more pronounced in the minor variant.

UVR inhibits photosynthesis of cyanobacteria (Sinha et al., 1995; Kumar et al., 1996; Han et al., 2003; Wu et al., 2005b) and other photosynthetic organisms (Häder et al., 2001; Gao et al., 2007b). High levels of UV-B may reduce phycobilisomes-related fluorescence and PSII activity in *Arthrospira* (Rajagopal et al., 2000, 2005). Damage to D1 protein is known to be responsible for the inhibited activity of PSII (Sass et al., 1997). In the present study, faster reduction of the photosynthetic quantum yield in the minor strain reflects more damages to the key proteins compared to the major one.

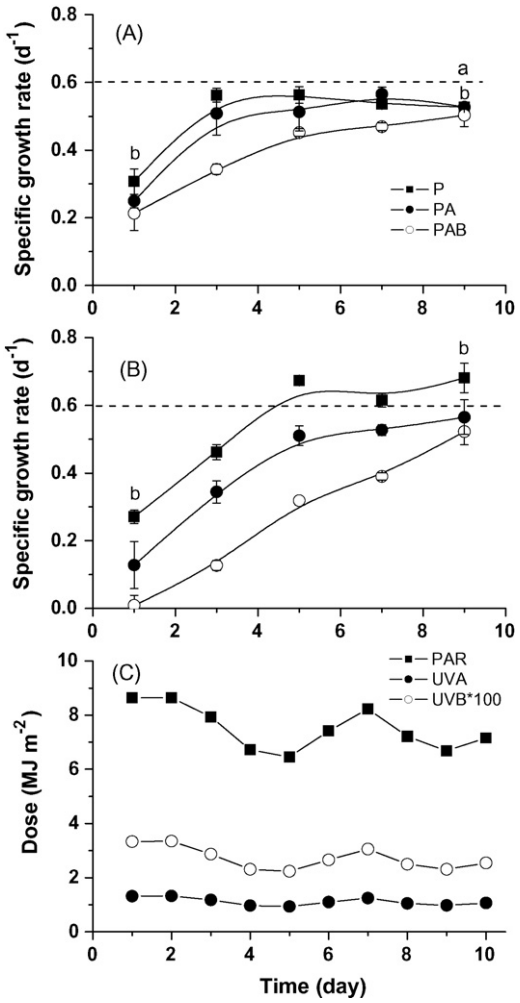

Fig. 4. Specific growth rate of *A. platensis* D-0083 (A) and the minor variant (B) under PAB, PA and P treatments during the long-term outdoor cultures, and the daily doses (C) during the exposure period of 29 October–7 November 2004. The dash lines represent the maximum specific growth rate in semi-continuous cultures under indoor conditions (PAR: 250 μmol m^{-2} s^{-1}, 25 °C). Mean ± S.D. ($n = 3$). Letter 'a' or 'b' denotes insignificant difference between P and PAB treatments or PA and PAB treatments at $p = 0.05$ level, respectively.

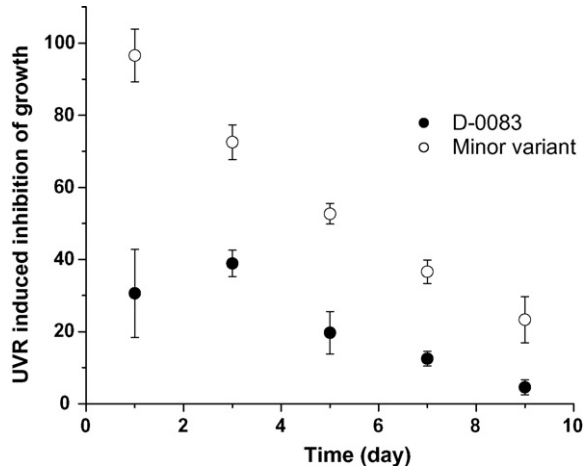

Fig. 5. UVR-induced inhibition of specific growth rate in *A. platensis* D-0083 and the minor variant. The inhibition was calculated as $(G_P - G_{PAB}) \times 100\%/G_P$, G_P and G_{PAB} present the specific growth rate of the samples treated with solar radiation screened off UVR (G_P) and with UVR (G_{PAB}). The difference between the two strains was significant for all the days when their growth rate was measured. Mean ± S.D. ($n = 3$).

The UV-absorbing compounds (UVAC), such as mycosporine-like amino acids (MAAs) and scytonemin, are known to play significant roles to screen off UVR (Garcia-Pichel and Castenholz, 1993; Brenowitz and Castenholz, 1997; Dillon et al., 2003). The amount of UVAC in D-0083 and its minor variant was negligible and not induced by exposures to UVR, suggesting that synthesis of UVAC was not a functional mechanism for *A. platensis* to protect cells from UV radiation damage. Gas vacuoles in the cells (Rajagopal et al., 2005) and tightened spiral structure (Wu et al., 2005b) were suggested to play protective roles against UVR in *Arthrospira* spp. via scattering or shading effects. The size of the minor variant was only about 1/4 of the major, therefore, cells of the minor variant were more prone to be harmed by high levels of PAR and UVR in view of the self-shading effects of each individual filament. Bigger cells tend to be more resistant to UVR than smaller ones in terms of DNA damage (Karent et al., 1991; Boelen et al., 2002). Bigger spirals of *Arthrospira* spp. could provide more shading for protection against UVR.

In the present study, contents of carotenoids increased with time while the cells acclimated to solar radiations. Carotenoids can function as a protective agents quenching highly reactive singlet oxygen and dissipating excess excitation energy as heat under high irradiance (Nonnengiesser et al., 1996). They were also found to be effective in protecting the photosynthetic apparatus against UV-B (Rakhimberdieva et al., 2004). Our results indicate a protective role by carotenoids in *A. platensis* D-0083 and its minor variant grown under high PAR and UVR. In the cultures under high PAR, the ratio of carotenoids to Chl *a* concentration increased due to the loss of Chl *a* in both D-0083 and the minor strain. While in the cultures exposed to UVR + PAR, there was an increase (days 4 and 6) in carotenoids concentration in D-0083, the observed increase in the ratio on other days was due to the loss of Chl *a*. In the minor variant, the increased ratio was due to both the increase of carotenoids and loss of Chl *a*. The cellular levels of carotenoids in the minor variant were inadequate to prevent the photo-oxidation of Chl *a* by the levels of UVR encountered during the period, which was reflected in the subsequent slow recovery at reduced levels of solar radiation.

Spiral structures of *Arthrospira* spp. can alter according to environmental changes. Mühling et al. (2003) found that the helix orientation could be reversed by temperature changes. Wang and Zhao (2005) proved that the morphogenesis of *Arthrospira* filaments from spiral to linear was due to DNA mutation. Wu et al. (2005b) found that UVR broke the spiral structure and suggested that, over long-time scales (i.e. decades), adaptation to solar UVR could bring about changes in the spiral structure from a rather loose to a very compressed helix. The morphology of the minor variant has been consistent with the same configuration under our laboratory conditions (PAR alone of 100 μmol m^{-2} s^{-1}) for more than 500 generations, reflecting a heritable feature. The broken spirals caused by solar UVR, as reported previously (Wu et al., 2005b), might be responsible

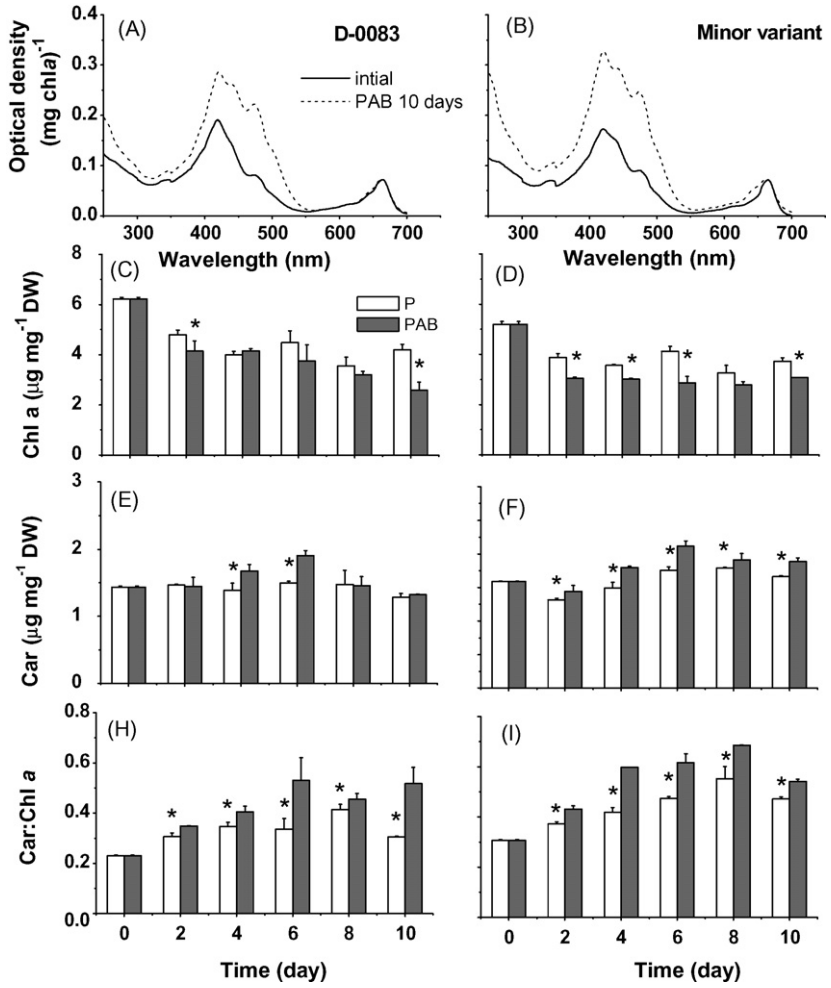

Fig. 6. Spectral characteristics of methanol extracted supernatant (A, B), concentrations of Chl *a* (C, D) and carotenoids (E, F) and the ratio of carotenoids to Chl *a* (H, I) from *A. platensis* D-0083 and the minor variant while grown under P and PAB treatments. The optical density was normalized to Chl *a* concentration. Mean ± S.D. ($n = 3$). The symbol (*) indicate that there was significant difference between P and PAB treatments at $p = 0.05$ level.

for the occurrence of the minor variant that gained the heritable (non-reversible under indoor low PAR) morphology over decades of outdoor production with mutated DNA. Therefore, solar UVR can be considered responsible for the minor variant's appearance in the commercial ponds due to its sensitivity to UVR damage. The minor variant of *Arthrospira* spp. has been considered as a challenge (Vonshak and Richmond, 1988) due to the difficulty for its harvest and negative effects on the quality control. On the other hand, the flotation activity of the minor variant was much lower than the major strain (data not shown), so the minor variant tends to be less capable of floating up to the culture surface with its own buoyancy, which decreases its feasibility to face strong solar radiation. Duration of exposures of *Arthrospira* cells to high levels of solar radiation would affect occurrence of the minor variant.

Acknowledgements

This study was funded by the National Natural Science Foundation of China (No. 90411018), Guangdong Natural Science Foundation and Hainan DIC Microalgae Co.

References

Aguirre-von-Wobeser, E., Figueroa, F.L., Cabello-Pasini, A., 2000. Effect of UV radiation on photoinhibition of marine macrophytes in culture systems. J. Appl. Phycol. 12, 159–168.

Belay, A., 1997. Mass culture of Spirulina outdoors—the Earthrise farms experience. In: Vonshak, A. (Ed.), *Spirulina platensis* (*Arthrospira*): Physiology, Cell-biology and Biotechnology. Taylor & Francis Publishers, London, pp. 131–158.

Boelen, P., Post, A.F., Veldhuis, M.J.W., Buma, A.G.J., 2002. Diel patterns of UVBR-induced DNA damage in picoplankton size fractions from the Gulf of Aqaba, Red Sea. Microbiol. Ecol. 44, 164–174.

Brenowitz, S., Castenholz, R.W., 1997. Long-term effects of UV and visible irradiance on natural populations of a scytonemin-containing cyanobacterium (*Calothrix* sp.). FEMS Microbiol. Ecol. 24, 343–352.

Ciferri, O., 1983. *Spirulina*, the edible microorganism. Microbiol. Rev. 47, 551–578.

Dillon, J.G., Miller, S.R., Castenholz, R.W., 2003. UV-acclimation responses in natural populations of cyanobacteria (*Calothrix* sp.). Environ. Microbiol. 5, 473–483.

Gao, K., Wu, Y., Li, G., Wu, H., Villafañe, V.E., Helbling, E.W., 2007a. Solar UV radiation drives CO_2 fixation in marine phytoplankton: a double-edged sword. Plant physiol. 144, 54–59.

Gao, K., Guan, W., Helbling, E.W., 2007b. Effects of solar ultraviolet radiation on photosynthesis of the marine red tide alga *Heterosigma akashiwo* (Raphidophyceae). J. Photochem. Photobiol. B: Biol. 86, 140–148.

Garcia-Pichel, F., Castenholz, R.W., 1993. Occurrence of UV-absorbing mycosporine-like compounds among cyanobacterial isolates and an estimation of their screening capacity. Appl. Environ. Microbiol. 59, 163–169.

Häder, D.P., Lebert, M., Marangoni, R., Colombetti, G., 1999. ELDONET: European Light Dosimeter Network hardware and software. J. Photochem. Photobiol. B 52, 51–58.

Häder, D.P., Lebert, M., Helbling, E.W., 2001. Photosynthetic performance of marine macroalgae measured in Patagonia on site. Trends Photochem. Photobiol. 8, 145–152.

Han, T., Sinha, R.P., Häder, D.P., 2003. Effects of intense PAR and UV radiation on photosynthesis, growth and pigmentation in the rice-field cyanobacterium *Anabaena* sp. Photochem. Photobiol. Sci. 2, 649–654.

Jeeji Bai, N., 1985. Competitive exclusion or morphological transformation? A case study with *Spirulina fusiformis*. Arch. Hydrobiol. 38 (Suppl. 71), 191–199 (Algol Stud.).

Jensen, S., Knutsen, G., 1993. Influence of light and temperature on photoinhibition of phytosynthesis in *Spirulina platensis*. J. Appl. Phycol. 5, 495–504.

Karent, D., Cleaver, J.E., Mitchell, D.L., 1991. Cell survival characteristics and molecular responses of Antarctic phytoplankton to ultraviolet-B radiation. J. Phycol. 27, 326–341.

Kolber, Z., Falkowski, P.G., 1993. Use of fluorescence to estimate phytoplankton photosynthesis *in situ*. Limnol. Oceanogr. 38, 1646–1665.

Korbee-Peinado, N., Abdala-Díaz, R.T., Figueroa, F.L., Helbling, E.W., 2004. Ammonium and UV radiation stimulate the accumulation of mycosporine-like amino acids in *Porphyra columbina* (Rhodophyta) from patagonia, argentina. J. Phycol. 40, 248–259.

Kumar, A., Sinha, R.P., Häder, D.P., 1996. Effect of UV-B on enzymes of nitrogen metabolism in the cyanobacterium *Nostoc calcicola*. J. Plant Physiol. 148, 86–91.

Lu, J., Yoshizaki, G., Sakai, K., Takeuchi, T., 2002. Acceptability of raw *Spirulina platensis* by larval tilapia *Oreochromis niloticus*. Fish. Sci. 68, 51–58.

Ma, Z., Gao, K., Watanabe, T., 2006. Effects of dissolved organic matter on the growth and pigments synthesis of *Spirulina platensis* (*Arthrospira*). Prog. Nat. Sci. 16, 50–54.

Mosulishvili, L.M., Kirkesali, E.I., Belokobylsky, A.I., Khizanishvili, A.I., Frontasyeva, M.V., Pavlov, S.S., Gundorina, S.F., 2002. Experimental substantiation of the possibility of developing selenium- and iodine-containing pharmaceuticals based on blue-green algae *Spirulina platensis*. J. Pharm. Biomed. Anal. 30, 87–97.

Mühling, M., Harris, N., Belay, A., Whitton, B.A., 2003. Reversal of helix orientation in the cyanobacterium *Arthrospira*. J. Phycol. 39, 360–367.

Nonnengiesser, K., Schuster, A., Koenig, F., 1996. Carotenoids and reaction center II-D1 protein in light regulation of the photosynthetic apparatus in *Aphanocapsa*. Bot. Acta 109, 115–124.

Parsons, T.R., Strickland, J.D.H., 1963. Discussion of spectrophotometric determination of marine plant pigments, with revised equation for ascertaining chlorophylls and carotenoids. J. Mar. Res. 21, 155–163.

Porra, R.J., 2002. The chequered history of the development and use of simultaneous equations for the determination of chlorophylls *a* and *b*. Photosynth. Res. 73, 149–156.

Rajagopal, S., Murthy, S.D.S., Mohanty, P., 2000. Effect of ultraviolet-B radiation on intact cells of the cyanobacterium *Spirulina platensis*: characterization of the alterations in the thylakoid membranes. J. Photochem. Photobiol. B: Biol. 54, 61–66.

Rajagopal, S., Sicora, C., Várkonyi, Z., Mustárdy, L., Mohanty, P., 2005. Protective effect of supplemental low intensity white light on ultraviolet-B exposure-induced impairment in cyanobacterium *Spirulina platensis*: formation of air vacuoles as a possible protective measure. Photosynth. Res. 85 (2), 181–189.

Rakhimberdieva, M.G., Stadnichuk, I.N., Elanskaya, I.V., Mustárdy, L., Mohanty, P., 2004. Carotenoid-induced quenching of phycobilisome fluorescence in photosystem II-deficient mutant of *Synechocystis* sp. FEBS Lett. 574, 85–88.

Sass, L., Spetea, C., Máté, Z., Nagy, F., Vass, I., 1997. Repair of UV-B induced damage of photosystem II via de novo synthesis of the D1 and D2 reaction centre subunits in *Synechocystis* sp. PCC 6803. Photosynth. Res. 54, 55–62.

Schreiber, U., Endo, T., Mi, H., Asada, K., 1995. Quenching analysis of chlorophyll fluorescence by the saturation pulse method: particular aspects relating to the study of eukaryotic algae and cyanobacteria. Plant Cell Physiol. 36, 873–882.

Sinha, R.P., Kumar, H.D., Kumar, A., Häder, D.P., 1995. Effects of UV-B irradiation on growth, survival, pigmentation and nitrogen metabolism enzymes in cyanobacteria. Acta Protozool. 34, 187–192.

Torzillo, G., Bernardini, P., Masojídek, J., 1998. On-line monitoring of chlorophyll fluorescence to assess the extent of photoinhibition of photosynthesis induced by high oxygen concentration and low temperature and its effect on the productivity of outdoor cultures of *Spirulina platensis* (cyanobacteria). J. Phycol. 34, 504–510.

Vonshak, A., 1990. Recent advances in microalgal biotechnology. Biotechnol. Adv. 8 (4), 709–727.

Vonshak, A., 1997. *Spirulina*: growth, physiology and biochemistry. In: Vonshak, A. (Ed.), *Spirulina platensis* (*Arthrospira*): Physiology, Cell-biology and Biotechnology. Taylor and Francis, London, pp. 43–66.

Vonshak, A., Richmond, A., 1988. Mass production of the blue-green algae *Spirulina*: an overview. Biomass 15 (4), 233–247.

Vonshak, A., Boussiba, S., Abeliovich, A., Richmond, A., 1983. Production of *Spirulina* biomass: Maintenance of monoalgal culture outdoors. Biotechnol. Bioeng. 25, 341–349.

Vonshak, A., Torzillo, G., Tomaseli, L., 1994. Use of chlorophyll fluorescence to estimate the effect of photoinhibition in outdoor cultures of *Spirulina platensis*. J. Appl. Phycol. 6, 31–34.

Wang, Z., Zhao, Y., 2005. Morphological reversion of *Spirulina* (*Arthrospira*) *platensis* (Cyanophyta): from linear to helical. J. phycol. 41, 622–628.

Wu, H., Gao, K., Ma, Z., Watanabe, T., 2005a. Effect of solar ultraviolet radiation on biomass production and pigment contents of *Spirulina platensis* in commercial operations under sunny and cloudy weather conditions. Fish. Sci. 71, 454–456.

Wu, H., Gao, K., Villafañe, V., Watanabe, T., Helbling, E.W., 2005b. Effects of solar UV radiation on morphology and photosynthesis of the filamentous cyanobacterium *Arthrospira platensis*. Appl. Environ. Microbiol. 71, 5004–5013.