Photosynthetic performance and pigment content in the aquatic liverwort *Riella helicophylla* under natural solar irradiance and solar irradiance without ultraviolet light

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Abstract

The effects of solar radiation and solar radiation deprived of ultraviolet radiation (λ < 395 nm) on photosynthetic performance and pigment content of the aquatic liverwort *Riella helicophylla* (Bory et Mont.) Mont. was investigated throughout a natural daily-light cycle, from sunrise to sunset. Photosynthetic performance was estimated by means of pulse amplitude-modulated (PAM) fluorescence and oxygen production. Photosynthetic characteristics changed daily and between the two solar radiation conditions. The lowest values of optimal quantum yield (ΦPS II) took place at noon (70 and 55% lower than early in the morning under UV- and non UV-screened plants, respectively), in agreement with the highest amounts of solar radiation. However, at this time, the electron transport rate (ETR) achieved the highest values of the day (63 and 125 μmol m⁻² s⁻¹ under PAR + UVA + UVB and PAR treatments, respectively). Significant differences (P < 0.05) in Fv/Fm, ETR, initial slope of ETR versus irradiance curve, photosynthetic capacity, chlorophyll a and phenolic compounds were found between the two radiation conditions, especially at noon. Recovery of inhibited photosynthesis took place in the afternoon, so the current ultraviolet radiation...
does not seem to cause, irreversible damage to the photosynthetic apparatus of \textit{R. helicophylla} in the short term. © 2002 Elsevier Science B.V. All rights reserved.

**Keywords:** Bryophytes; Liverwort; PAM fluorescence; Photosynthesis; \textit{Riella helicophylla} (Bory et Mont.) Mont.; Solar radiation; Ultraviolet radiation

1. Introduction

The quality and intensity of solar irradiance reaching the earth’s surface changes throughout the day, depending on the angle of the sun and the presence of clouds. It has been shown that net positive photosynthesis of terrestrial (Schreiber et al., 1994) and aquatic (Hanelt, 1996) plants outdoors, is restricted to a specific range of irradiances. At low irradiances (as occurs early in the morning and evening), the rate of photosynthesis may fall below the light compensation point. At the other extreme, high irradiance can damage the photosynthetic apparatus (Henley, 1993). Photosynthetic performance (estimated as quantum yield of PS II) often decreases around noon, coinciding with the highest solar radiation and this may be considered a protective mechanism to avoid the damage caused by excessive irradiance (Hanelt, 1996; Flores-Moya et al., 1998). However, previous studies did not take into account changes in the electron transport rate (ETR), which are closely related to the photosynthetic rate estimated by O$_2$-production (Beer and Björk, 2000).

The effects of different solar wavelengths must be considered because the levels of ultraviolet-B radiation (UVB; $\lambda = 280–315$ nm) at the earth’s surface are increasing due to the depletion of stratospheric ozone (Seckmeyer and McKenzie, 1992), relative to ultraviolet-A (UVA; $\lambda = 315–400$ nm) and photosynthetic active radiation (PAR; $\lambda = 400–700$), since these portions of solar spectrum are not absorbed by the stratospheric ozone layer. Photosynthetic organisms may be affected by this shift towards UVB radiation (Cooihill, 1991; Allen et al., 1998). Since the principal biological target of UVB radiation is the photosynthetic apparatus (Greenberg et al., 1989), the possible role of UVB radiation on the inhibition of photosynthesis must be taken into account in the studies of photoinhibition under natural conditions. The synthesis and accumulation of phenolic compounds with appropriate UV absorptive properties could be a protection mechanism in plants to avoid the harmful effects of short wavelength radiation (Cadwell, 1981), as yet there is no role proposed for these compounds as UV screens (Waterman and Mole, 1994).

The studies on photoinhibition of photosynthesis and its role in protecting the photosynthetic apparatus have been focussed on terrestrial plants (Baker and Bowyer, 1994) and more recently, on micro- and macro-algae and seagrasses (see reviews by Franklin and Forster, 1997; Häder and Figueroa, 1997), but few studies have been carried out in terrestrial and aquatic bryophytes (Takács et al., 1999). Therefore, the aim of this work was to study the effects of PAR and ultraviolet radiation (UVR: $\lambda < 395$ nm) on photosynthetic performance, pigment content and total phenolic compounds of the aquatic liverwort \textit{Riella helicophylla} (Bory et Mont.) Mont. (Sphaerocarpales, Riellaceae), throughout a natural daily-light cycle, from sunrise to sunset.
2. Materials and Methods

2.1. Plant material and sampling site

*R. helicophylla* is a dioecious aquatic liverwort with a monostratified blade. It reaches 2–3 cm in length and grows during the spring season in temporally, deep lakes of brackish water, along the Mediterranean region, (Cirujano et al., 1988).

Samples were collected at 5–10 cm depth (its typical vertical distribution range) in Lake Laguna Cerero (UTM = 30SUG0138) the day before experiments were carried out (25th May 1998). The lake is located in an endoreic basin of southern Spain. At this time of the year, typical values of temperature and salinity of water were 22 °C and 4 ‰, respectively. After sampling, the samples were transported to the laboratory in darkness in an ice-chest with water from the sample site. In the laboratory, the samples were kept overnight in darkness and in filtered (Whatman GF/C) lake water, at 22 ± 1 °C.

150 randomly selected thalli (the female:male ratio was 3:2) were placed in two, 10 l white polyvinylchloride tanks (75 plants per tank) in filtered (Whatman GF/C) lake water. The tanks were placed on the roof of the Faculty of Sciences of the University of Málaga 1 h before sunrise. The water temperature was maintained at 22 ± 1 °C. To avoid nutrient depletion, the water was renewed every 2.5 h.

Two irradiance treatments were used, full solar radiation (PAR + UV A + UVB) and solar radiation deprived of UVR (just PAR). For this purpose, one of the tanks was covered with a Ultraphan-395® filter (Digefra GmbH, Munich, Germany) for the just PAR treatment, which show a 2 and 91% transmission for UVR + PAR and just PAR, respectively, while the other tank was covered with an Ultraphan-295® filter, with 82 and 92% transmission for UVR + PAR and just PAR, respectively, for the PAR + UVA + UVB treatment. The spectral transmission characteristics of the cut-off filters were shown in Figueroa et al. (1997). To avoid any lens effect because of water droplets, the filters were put into the tanks below the water level.

During the day, samples in the tanks were collected every 2.5 h, from 06:00 to 18:30 (GMT).

2.2. Measurements of natural solar radiation

Underwater measurements of solar radiation at 1, 50, 100, 150 and 200 cm depth were carried out in Laguna Cerero by means of a LI-1800 UW-spectroradiometer (Li-Cor Inc., Lincoln, NE). PAR, UVA and UVB irradiances were obtained by integrating between the limit wavelengths of every fraction of the spectrum. In this case, UVB values were underestimated because the device is unable to measure below 300 nm. The vertical attenuation coefficient for each radiation band ($K_{\text{PAR}}$, $K_{\text{UVA}}$ and $K_{\text{UVB}}$) was obtained using Lambert Beer’s law: $K_{\text{PAR, UVA, UVB}} = (\ln E_o - \ln E_z)z^{-1}$, where $E_o$ and $E_z$ are the irradiance of PAR, UVA or UVB, at the surface and at the depth $z$, respectively.

Measurements for daily irradiance were performed every 15 min from 6:00 to 18:30 h using different radiometers. Instantaneous PAR was measured using a quantum radiometer LI-189 equipped with a 2π quantum sensor (Li-Cor Inc., Lincoln, NE). To measure UVA and UVB radiation two different flat sensors (Dr. Gröbel, UV-Elektronik GmbH, Germany) were
used. The values obtained were transformed to those measured with a radiometer ELDONET (Real time Computer, Möhrendorf, Germany) which follows the international range of the C.I.E. (International Commission of Illumination, 1935) in three wavelength bands (PAR, UVA and UVB). The correction factors for the light devices were 0.90 (LI-189/ELDONET), 0.50 (Gröbel/ELDONET) and 0.17 (Gröbel/ELDONET) for PAR, UVA and UVB, respectively.

In order to calculate the weighted irradiance of the inhibition of photosynthesis, the solar radiation in wavelengths 300–800 nm was measured at 1 nm intervals, every hour using the LI 1800 UW-spectroradiometer. The biologically weighted irradiance (BWI) was calculated as the area under the curve which results from multiplying a weighting function, i.e. the action spectrum for the inhibition of photosynthesis in isolated chloroplasts (Jones and Kok, 1966), by the incident spectral irradiance (Rundel, 1983).

\[
BWI = \int_{\lambda_0}^{\lambda_1} I_\lambda e_\lambda d\lambda
\]

where \(I_\lambda\) is the irradiance at \(\lambda\) (nm) and \(e_\lambda\) the biological response at \(\lambda\) (nm) defined by the action spectrum.

2.3. In vivo measurements of chlorophyll a fluorescence

Chlorophyll a fluorescence of the PS II was measured with a pulse amplitude-modulated-2000 (PAM) fluorometer (Walz, Effeltrich, Germany) following Schreiber et al. (1986). The effective quantum yield (\(\Phi_{\text{PS II}}\) defined as \(\Delta F/F_m\)) in illuminated samples and the optimal quantum yield (\(F_v/F_m\)) in dark-acclimated samples were determined following Schreiber et al. (1986).

In order to discern how energy is dissipated during photosynthesis, the photochemical (\(q_P\)) and nonphotochemical (\(q_N\)) quenching of fluorescence were determined following Schreiber et al. (1986).

At different times of the day, the instantaneous ETR throughout the electron transport chain was calculated as follows:

\[
\text{ETR} = \Phi_{\text{PS II}} \times \text{PAR} \times AF \times 0.5
\]

where AF is the absorption factor of the thalli and 0.5 corresponds to the fraction of photons absorbed by PS II (assuming that half of the photons which are necessary to fix one carbon molecule, are absorbed by PS II). The AF values were calculated from absorbance (A) by using the equation:

\[
A = -\log(1 - AF)
\]

where AF is the absorption factor estimated with the opal glass technique (Mercado et al., 1996).

The relationship between ETR and incident PAR was determined at different sampling periods of the day. For this purpose, thalli were exposed to increasing irradiance between 0 and 300 \(\mu\)mol m\(^{-2}\)s\(^{-1}\) (0–61 Wm\(^{-2}\)), which is the highest value that could be achieved with the built-in red LEDs (\(\lambda_{\text{max}} = 650\) nm) from the PAM, at intervals of 30 s, after
which, a saturating pulse of white light was applied to determine $\Phi_{\text{PS II}}$. LEDs rather than halogen lamps were used because the spectrum changes with irradiance in the halogen lamp.

The initial slope of the ETR versus irradiance ($\alpha$ under light-limited conditions) relationships was determined by linear fitting of the three first points of the ETR versus irradiance curve.

2.4. Oxygen exchange measurements

Rates of dark respiration (DR) and light-saturated net photosynthesis ($P_{\max}$) were determined as oxygen exchange by means of a Clark-type electrode YSI 5331 (Yellow Springs Instruments Co., OH). About 3–4 thalli (approximately 10–12 mg fresh weight) were incubated in a chamber filled with 7 ml of natural water from the collection site at 22 ± 0.1 °C. To estimate DR, the chamber was covered with aluminium foil. Previous experience (data not shown) showed that the light-saturated rate was achieved at 400–500 $\mu$mol m$^{-2}$ s$^{-1}$ and no photoinhibition was obtained even at 1500 $\mu$mol m$^{-2}$ s$^{-1}$. Therefore, to estimate $P_{\max}$ the samples were incubated at 700 $\mu$mol m$^{-2}$ s$^{-1}$ of PAR. Both DR and $P_{\max}$ were obtained when the oxygen concentration changed linearly as a function of time (approximately 10 min).

2.5. Analysis of the photosynthetic pigment and phenolic compounds content

According to Inskeep and Bloom (1985), samples were maintained for 12 h in $N,N$-dimethylformamide at 4 ± 1 °C, in darkness, after which, the absorbance of the extracts was measured spectrophotometrically. The concentrations of chlorophyll a, chlorophyll b and total carotoids were determined according to the equations from Wellburn (1994).

To estimate the concentration of phenolic substances, samples were extracted overnight in micro-tubes with 1 ml 80% (v/v) aqueous methanol. The mixture was centrifuged for 15 min at 5000 g. Supernatant was collected and the Folin and Ciocalteu (1927) assay was used. Total phenolic compounds were expressed as equivalents of phloroglucinol concentration.

2.6. Statistical analysis

Data were compared by a two-way (time of day and solar radiation conditions) model I ANOVA. Tukey’s honestly significant difference method was applied when significance was found. The relationship between the optimal quantum yield and the logarithm of the BWI was tested by the Pearson’s correlation coefficient and linear regression. In all cases, normality of data was assessed by Kolmogorov–Smirnov test. Homogeneity of variances was verified by Barlett’s test when the number of replications was $n \geq 6$ or the $F_{\max}$-test when the number of replications was $n \leq 5$. In the case of quenching analysis, homocedasticity of data was not achieved even when different transformations were tested and, so the nonparametric Kruskal–Wallis test was used. All the statistical tests were carried out in accordance with Sokal and Rohlf (1995).
Table 1
Irradiance water properties of Laguna Cerero (south Spain) on 25th May 1998

<table>
<thead>
<tr>
<th></th>
<th>PAR</th>
<th>UVA</th>
<th>UVB</th>
</tr>
</thead>
<tbody>
<tr>
<td>$K_d$ (m$^{-1}$)</td>
<td>0.88</td>
<td>3.30</td>
<td>10.33</td>
</tr>
<tr>
<td>Percentage of $I_0$ on the bottom (2.0 m)</td>
<td>13.30</td>
<td>0.05</td>
<td>0.00</td>
</tr>
<tr>
<td>Percentage of $I_0$ to 0.1 m depth</td>
<td>91.50</td>
<td>72.00</td>
<td>35.50</td>
</tr>
</tbody>
</table>

Vertical attenuation coefficients ($K_d$) and the percentage of surface irradiance ($I_0$) at the different depths for PAR, UVA, and UVB.

3. Results

3.1. Measurements of natural solar radiation in water and air

The light climate at the bottom of the lake (5–10 cm depth) where R. helicophylla grows, was characterized by very high levels of PAR and UVA, due to the shallow depth and low extinction coefficients (Table 1). However, UVB levels at 10 cm depth were reduced to around one third of those at the surface because the extinction coefficient of UVB was very high, i.e. one magnitude order higher than that for UVA (Table 1). In fact, no detectable UVB were found at the deepest end of the lake (Table 1).

The outdoor experiments were performed on a sunny day, with only short cloud events at noon and during the evening (Fig. 1). The maximum irradiance at the different wavebands occurred around noon (394.6, 63.2 and 1.5 Wm$^{-2}$ of PAR, UVA and UVB, respectively). At this time of day, the solar radiation was more enriched in UVR than in PAR, in comparison to the ratios found in the early morning or in the evening (Fig. 2). Daily doses of solar radiation were 10344.5, 1509.8 and 32.2 kJ m$^{-2}$ of PAR, UVA and UVB, respectively.

Fig. 1. Daily changes in solar radiation during the study period (26th May 1998). UVB ($\times 10$) ($\lambda = 280–315$ nm), UVA ($\lambda = 315–400$ nm) and PAR ($\lambda = 400–700$ nm) under both filter: Ultraphan-295 and Ultraphan-395 used to obtain two irradiance treatments.
3.2. In vivo measurements of chlorophyll a fluorescence

A similar daily pattern was found both in $F_v/F_m$ and $\Phi_{PSII}$ (data not shown), with high values in the early morning and in the evening, and the lowest around noon. The decrease in $F_v/F_m$ at noon was more dramatic when UVR was present ($P < 0.001$) than under the just PAR (Fig. 3). The recovery of $F_v/F_m$ in the afternoon began earlier ($P < 0.05$; Kruskal–Wallis test) in the absence of UVR than under the full solar spectrum. However, at the end of the day the $F_v/F_m$ values were significantly lower ($P < 0.001$) than those measured at the beginning of the day. A significant negative correlation ($r_p = -0.89$, $P < 0.001$, $n = 30$) was found between the logarithm of UVR at each experimental time.
of the day and the values of $F_{v}/F_{m}$ from *R. helicophylla*. This coefficient was improved ($r_p = -0.93$, $P < 0.001$, $n = 30$) when the $F_{v}/F_{m}$ values were correlated with the logarithm of biological weighted irradiance for the inhibition of photosynthesis in isolated chloroplasts (Fig. 4).

Although decreases in $F_{v}/F_{m}$ occurred throughout the morning (Fig. 3), the electron flow in the electron transport chain increased significantly ($P < 0.001$) in agreement with increasing PAR (Fig. 1). However, a clear deleterious effect of UVR on ETR was detected because the ETR under just PAR was significantly higher ($P < 0.001$) than under the full solar spectrum treatment (Fig. 5). The AF, used to calculate the ETR, showed significant differences ($P < 0.05$) between both treatments only at noon (Table 2).
Table 2
Variation of chlorophyll a (mg g\(^{-1}\) FW), total phenolic compounds (mg g\(^{-1}\) FW) and AF of plants during the day, under both irradiance treatments (PAR + UVA + UVB and PAR)

<table>
<thead>
<tr>
<th>Time (GMT) (h:min)</th>
<th>Chlorophyll a</th>
<th>Total phenolic compounds</th>
<th>AF</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>PAR + UVA +</td>
<td>PAR + UVA +</td>
<td>PAR + UVA +</td>
</tr>
<tr>
<td></td>
<td>UVB</td>
<td>UVB</td>
<td>UVB</td>
</tr>
<tr>
<td>6:00</td>
<td>0.34 ± 0.09</td>
<td>0.34 ± 0.09</td>
<td>0.19 ± 0.04</td>
</tr>
<tr>
<td>8:30</td>
<td>0.10 ± 0.07</td>
<td>0.22 ± 0.06</td>
<td>0.14 ± 0.07</td>
</tr>
<tr>
<td>11:00</td>
<td>0.23 ± 0.03(\text{a,b})</td>
<td>0.32 ± 0.07(\text{a,b})</td>
<td>0.22 ± 0.03</td>
</tr>
<tr>
<td>13:30</td>
<td>0.10 ± 0.01</td>
<td>0.18 ± 0.04</td>
<td>0.27 ± 0.08(\text{a})</td>
</tr>
<tr>
<td>16:00</td>
<td>0.32 ± 0.07</td>
<td>0.39 ± 0.06</td>
<td>0.18 ± 0.01</td>
</tr>
<tr>
<td>18:30</td>
<td>0.27 ± 0.07</td>
<td>0.26 ± 0.06</td>
<td>0.28 ± 0.10</td>
</tr>
</tbody>
</table>

Data are the average ± S.D. of three samples.

\(\text{a}\) Indicate significant differences (\(P < 0.05\)) between irradiance treatments at this time of the day.

\(\text{b}\) Means significant differences (\(P < 0.05\)) with other time of the day under the same treatment.

Fig. 6. Daily changes in (A) ETR at 300 \(\mu\text{mol m}^{-2}\text{s}^{-1}\) of red light (\(\lambda_{\text{max}} = 650\text{ nm}\)) and (B) initial slope of the ETR versus irradiance curves of \(R.\) \textit{helicophylla} samples under filtered (PAR) and unfiltered (PAR + UVA + UVB) solar radiation. Data are means ± S.D. of three samples.
In order to compare the ETR versus irradiance relationship obtained at each time, the initial slope (estimator of the efficiency in the harvest of photons) of the curves and ETR at 300 \( \mu \text{mol m}^{-2} \text{s}^{-1} \) of red light was used. Although, the ETR was not saturated at this irradiance, it is the highest intensity that could be achieved with the built-in LEDs lamps from the PAM device. Under just PAR, the values of ETR at 300 \( \mu \text{mol m}^{-2} \text{s}^{-1} \) red light increased during the morning until 11:00, when it reached maximum. Then, it decreased, reaching similar values in the evening to those measured in the early morning. In contrast, when UVR was present, it decreased from 08:30 onwards, (Fig. 6A). The efficiency of electron flow at low irradiance (Fig. 6B) did not change during the day under just PAR, but decreased under full solar spectrum treatment during the morning giving lower values \( (P < 0.05) \) than under just PAR.

Photochemical quenching did not change under any treatment or throughout the day (Fig. 7A). However, nonphotochemical quenching increased early in the morning \( (P < 0.05; \text{Kruskal–Wallis test}) \). This increase took place earlier in the morning under full sunlight than under sunlight depleted in UVR (Fig. 7B).

![Fig. 7. Daily changes in (A) photochemical \( (q_P) \) and (B) nonphotochemical \( (q_N) \) quenching of fluorescence of \textit{R. helicophylla} samples under filtered (PAR) and unfiltered (PAR + UVA + UVB) solar radiation. Data are means ± S.D. of five samples.](image)
3.3. Light-saturated photosynthesis and DR rates

Light-saturated photosynthetic rate under the just PAR treatment increased slightly during the morning, being higher \( (P < 0.05) \) around noon than when the ultraviolet radiation was present. Starting from this time, the photosynthetic rate began to decrease, reaching similar values to those measured in the early morning, under both irradiance conditions (Fig. 8A).

DR rate increased \( (P < 0.001) \) during the day, irrespective to the conditions of solar radiation (Fig. 8B).

3.4. Photosynthetic pigments

The lowest \( (P < 0.05) \) concentrations of chlorophyll a (Table 2) were measured around 11:00 h, in agreement with maximum irradiance. Furthermore, at this time of day the concentrations were lower under the presence of ultraviolet radiation than under just PAR. In any case, at the end of the day the concentrations were similar under both solar radiation
treatments, and close to those obtained in the morning. The concentrations of accessory pigments did not show significant daily variation and without significant differences between the two irradiance treatments (just PAR, and full solar spectrum).

3.5. Phenolic compounds

No clear daily pattern of phenolic compound content was found (Table 2). Nevertheless, a higher phenolic concentration was detected \((P < 0.05)\) at 13:30, under full solar spectrum than under just PAR treatment, coinciding with the highest values of solar radiation. The highest levels of phenolic compounds were found at the end of the day under the just PAR treatment.

4. Discussion

*R. helicophylla* grows in temporally shallow lakes along the Mediterranean region during the spring season (Cirujano et al., 1988). The latitude of this region, at this season, is exposed to high levels of solar radiation. The underwater irradiance measurements showed that the bottoms where *R. helicophylla* grows (0.1 m as maximum) were exposed to high irradiance conditions, with high levels of harmful ultraviolet irradiance (Coohill, 1991). Therefore, it could be expected that this species is adapted to high light conditions, like typical sun-adapted plants.

The exposure to increasing solar radiation caused a decline in effective and optimal quantum yield with the lowest values at noon, coinciding with highest solar radiation. This fact was also found in higher plants (Schreiber et al., 1994) and seaweeds (Hanelt, 1996; Flores-Moya et al., 1998). The recovery of the optimal quantum yield throughout the afternoon, when the solar irradiance declined, suggested a dynamic photoinhibition process, which caused a harmless dissipation of excessively absorbed energy by thermal radiation (Krause and Weis, 1991; Hanelt, 1996). In this way, the decline in quantum yield was accompanied by a rapid increment of \(q_N\). This could be due to the removal of the excessive energy which cannot be absorbed by the PS II (Weis and Berry, 1987; Schreiber et al., 1994).

The results suggest that the PS II from the light-harvesting antenna in *R. helicophylla* is less efficient at higher rather at lower irradiances. Nevertheless, the efficiency of electron flow at low radiation (initial slope of ETR-irradiance curve) did not change throughout the day under just PAR treatment. Furthermore, the highest ETR occurred around noon coinciding with the highest level of solar irradiance of the day. The last is in agreement with the fact that \(q_P\) did not change during the day. Weis and Berry (1987) found a correlation between ETR (calculated from fluorescence measurements) and CO\(_2\) fixation rate. Moreover, Beer and Björk (2000) found a similar value as theoretical molar relationship between photosynthetic O\(_2\) evolution and electron transport (4 mol electron transported per mol O\(_2\) evolved or CO\(_2\) fixed) in the seagrass *Halodule wrightii* Archers.

On the other hand, under the just PAR treatment, the maximum \(P_{max}\) and ETR at 300 \(\mu\)mol m\(^{-2}\) s\(^{-1}\) happened around noon. Henley (1993) considers that although the time at which \(P_{max}\) is maximal varies, it is normally reached in the morning or at noon. Other authors have found lower in situ O\(_2\) production rate in correlation with maximum solar
radiation (Huppertz et al., 1990; Flores-Moya et al., 1998). The diminution of \( P_{\text{max}} \) and ETR at 300 \( \mu \text{mol m}^{-2} \text{s}^{-1} \) of red light during the afternoon, could be due to an endogenous rhythm (Henley et al., 1991). Hanelt and Nultsch (1990) proposed that the movement of the chloroplasts could be able to reduce the absorption of energy by selfshading and, therefore, reduce the photosynthetic activity. In this way, Aguilera et al. (1999) show the effect UVR in thallus absorption and package effect. Henley et al. (1991) emphasized that diurnal increase in DR can modify the endogenous pattern of \( P_{\text{max}} \).

When UVR was present, the decrease in effective and optimum quantum yield was greater than when the plants were exposed to just PAR. Similar results were obtained by Dring et al. (1996) and Hanelt et al. (1997) in macro-algae, both found that UVR can increase the light stress of photosynthesis. In agreement with this fact, under full solar irradiance \( q_N \) increased faster and was higher than when UVR was absent, indicating higher heat dissipation. The fact that the efficiency of electron flow under ultraviolet treatment was lower starting around noon (at this time, the highest ratio UV/PAR occurs) could indicate that some proportion of the photoinhibition could be due to UVR. On the other hand, because the loss of efficiency is also accompanied by a loss of the photosynthetic capacity, estimated by \( P_{\text{max}} \) (in this case, a lower \( P_{\text{max}} \) under ultraviolet treatment takes place only at noon), could suggest a chronic photoinhibition.

In the afternoon, a recovery of quantum yield took place under both irradiance conditions. Nevertheless, under full solar radiation the recovery was delayed in comparison to the recovery under just PAR. This delay could be in agreement with the suggestion that recovery process to high-light stress consists of both a fast and slow recovery process (Leittsch et al., 1994; Hanelt, 1998). When ultraviolet radiation was absent, only a dynamic phoinhibition of the PS II could happen as has just been commented, which is a rapidly reversible process (fast phase), while under UVR a dynamic photoinhibition could be followed by chronic photoinhibition, which is a slowly reversible process, due to the fact that the damaged D1 protein turnover is slow (slow phase). Although neither efficiency of electron flow at low radiation (initial slope of relative ETR-irradiance curve) nor the electron transport capacity under ultraviolet condition was recovered at the end of daylight, the recovery process could continue throughout the night, as has been reported in some seaweeds (Häder et al., 1996b; Hanelt et al., 1997).

A better significant linear relationship was found between \( F_v/F_m \) and the logarithm of BWI for the inhibition of photosynthesis in isolated chloroplasts (Jones and Kok, 1966) than between \( F_v/F_m \) and the logarithm of unweighted ultraviolet irradiance. In the weighting function from Jones and Kok (1966) the higher load of effectiveness is located in the UVB portion of the solar spectrum. It could be concluded that the photosynthetic performance in \( R. \) helicophylla could be more affected by UVB than by UVA.

Chlorophyll a presented a daily pattern with the lowest values around noon, especially when UVR was presented. This fact has also been observed in the red algae \( R. \) verruculosa (Flores-Moya et al., 1998). López-Figueroa and Niell (1990) pointed out that the acclimation to high irradiance and light quality can be produced in short-term periods (hours) in some seaweeds, and López-Figueroa (1992) suggested that daily variations in pigment content are regulated by photomorphogenic photoreceptors.

Phenolic compounds of \( R. \) helicophylla blades were accumulated at the end of the diurnal period under the just PAR treatment, but a good explanation for this pattern cannot
be addressed. However, significantly higher concentrations of phenolic compounds were found at noon when the blades were exposed to full solar radiation in comparison to those exposed to just PAR. This suggests that the ultraviolet fraction could stimulate the synthesis and/or accumulation of phenolic compounds because they peaked when the UVR/PAR ratio achieved the highest values.

From the results obtained in *R. helicophylla*, the possible role of the synthesis and accumulation of phenolic compounds as a protective mechanism against UVR remains an open question.

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