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An assessment of the relationship between chlorophyll *a* fluorescence and CO_2 gas exchange from field measurements on a moss and lichen

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Abstract. The relationship between CO_2 exchange and relative electron-transport rate through photosystem II (ETR, measured using chlorophyll *a* fluorescence) was determined for a moss and a green algal lichen, photobiont probably *Trebouxia* sp., in the field in Antarctica. Net photosynthesis (NP) and dark respiration (DR) were measured over temperatures from zero to 25 °C and gross photosynthesis (GP) calculated (GP = NP + DR). The strong response of DR to temperature in these organisms resulted in substantial changes in CO_2 exchange rates. The moss *Bryum argenteum* Hedw. showed a strong, linear relationship between GP and ETR. This was an unexpected result since mosses are C_3 plants and, in higher plants, this group normally has a curvilinear GP versus ETR relationship. It is suggested that suppression of DR in the light might be involved. The lichen, *Umbilicaria aprina* Nyl., had nonlinear relationships between ETR and GP that were different at each measurement temperature. In some cases the lowest ETR was at the higher CO_2 exchange rates. It is suggested that these relationships are the result of strong quenching mechanisms that are inversely proportional to GP. The results support a growing impression that the relationships between ETR and CO_2 exchange are complex in these organisms and different from those found for higher plants.

Key words: *Bryum* (photosynthesis) – Chlorophyll *a* fluorescence – Electron transport – Lichen (photosynthesis) – Photosynthesis – Respiration

Introduction

Chlorophyll *a* (Chl *a*) fluorescence measurements have, predominantly, been made on higher, vascular plants. Genty et al. (1989) found an excellent linear correlation between Φ_{PSII} (the effective quantum yield of photosystem II; $\Phi_{PSII} = \Delta F/Fm'$; variable fluorescence, ΔF, divided by maximal fluorescence, Fm′ measured during or immediately after illumination) and Φ_{CO_2} (the quantum yield of CO_2 fixation under specific conditions), and this relationship has since been confirmed for many other plants (Seaton and Walker 1990; Krall and Edwards 1991; Krall et al. 1991; Edwards and Baker 1993). However, it was quickly noted that, although Φ_{PSII} and Φ_{CO_2} were often linearly related in C_4 plants and C_3 plants under low (2%) oxygen, the relationship was strongly curvilinear for C_3 plants under normal ambient (21%) oxygen. This nonlinearity was present whether Φ_{CO_2} was altered by changing the ambient CO_2 concentration (Krall and Edwards 1992) or the photon flux density (PFD; Harbinson et al. 1990). The nonlinearity has been explained as being a result of photorespiratory metabolism.

The relationship between CO_2 fixation and Φ_{PSII} or other fluorescence parameters has been little studied for either lichens or bryophytes. Both groups are not only taxonomically distant from higher plants but they possess substantially different physiologies. Bryophytes and lichens, for instance, are poikilohydric, in contrast to the homoiohydric vascular plants, and can tolerate desiccation. Although bryophytes are true plants they are primitive members, the gametophyte, rather than the sporophyte, being the dominant generation. Bryophyte tissues are also close to 100% photosynthetic, somewhat similar to vascular plant leaves, whereas lichens are dual

Abbreviations: Chl = chlorophyll; DR = dark respiration rate; ETR = relative electron-transport rate through photosystem II (= $\Phi_{PSII} \times$ PFD); Fo′ = minimal fluorescence in the light using far-red light to induce PSI activity; Fm′ = maximal fluorescence in the light; Ft = fluorescence in the light; GP = gross photosynthetic rate (NP + DR); NP = net photosynthetic rate; PFD = photon flux density; qN = non-photochemical chlorophyll fluorescence quenching; qP = photochemical chlorophyll fluorescence quenching; Φ_{CO_2} = quantum yield of CO_2-fixation; Φ_{PSII} = the effective quantum yield of photosystem II ($\Phi_{PSII} = \Delta F/Fm'$; variable fluorescence, ΔF [Fm′-Ft], divided by maximal fluorescence, Fm′ measured during, or immediately after, illumination)

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organisms, a heterotrophic mycobiont in symbiosis with an autotrophic photobiont that can be cyanobacterial, algal or both. The mycobiont is estimated to form around 90% of the biomass of the lichen but no estimates seem to exist as to the relative respiratory rates of the two components. Dark respiration (DR) in these groups tends to be high with respect to net photosynthesis (NP) in contrast to proportionately lower rates for vascular plant leaves (Green and Lange 1994). Lichens with cyanobacterial and some with algal photobionts have also been shown to possess CO_2-concentrating mechanisms (CCMs) that can reduce photorespiration rates and lower CO_2 compensation values (Badger et al. 1993; Palmqvist et al. 1997). Amongst terrestrial plants only C_4 species behave similarly, albeit through different physiological processes. Cyanobacteria are known to have Chl *a* fluorescence characteristics different from those of higher plants (Sundberg et al. 1997) and the presence of a CCM has been shown to affect the photosynthetic performance of lichens (Palmqvist et al. 1994, Smith and Griffiths 1996, Palmqvist et al. 1997, Sundberg et al. 1997). It would not be unexpected, therefore, if these groups behaved very differently from higher, vascular plants in both their CO_2 exchange and Chl *a* fluorescence performance. Sundberg et al. (1997) investigated the relationship between CO_2 fixation and Φ_{PSII} in cynobacterial lichens and found a strongly nonlinear relationship. This is somewhat surprising since all cyanobacteria can concentrate CO_2. Leisner et al. (1997) showed in field measurements on a green algal crustose lichen that ETR did not reflect CO_2 exchange when the latter was reduced due to increased thallus diffusion resistances. This contrasts with the results of Lange et al. (1996) who found that ETR tracked CO_2 exchange when thallus water content changed in both cyanobacterial and green algal species.

In this study we investigated the relationship between CO_2 fixation and Φ_{PSII} or ETR (relative electron transport rate, $\Phi_{PSII} \times$ PFD) for a green algal lichen, *Umbilicaria aprina*, and a moss, *Bryum argenteum*. Temperature strongly influences DR of bryophytes and lichens and NP can become negative at even moderate temperatures. This effect is particularly marked in cold-adapted lichens and bryophytes (Longton 1988) so the opportunity was taken to carry out the work during a field expedition to the Granite Harbour area, Southern Victoria Land, Antarctica. The measurements were made over a range of temperatures and PFDs making it possible to clarify whether ETR and Φ_{PSII} track NP, GP (= NP + DR) or neither.

Materials and methods

Plant material. Thalli of the lichen *Umbilicaria aprina* Nyl. were collected as required from rocks forming part of the scree slope behind the beach at Botany Bay (77°01′S, 162°32′E). Individual thalli were then stored under the elevated floor of the research tent erected on the boulder beach nearby. Ambient air temperatures were always between −6 °C and −15 °C so that stored thalli remained frozen and received low light levels but never direct sunshine. Small sections of turf (about 20 cm^2) of the moss *Bryum argenteum* Hedw. were collected from sites that had just become exposed through the melting of snow banks. During the day these moss banks were moist and were not frozen. The particular samples used, which were dark green indicating a shade, rather than full sun, adaptation, were stored moist at low ambient light within the research tent (0–10 °C) before use within 1 or 2d. Before storage the samples were cut to a thickness of around 6 mm.

Gas exchange. The methods used are essentially identical to those of Schroeter et al. (1994). Gas-exchange measurements were carried out using two 'minicuvette' systems (CMS4P; Walz Company, Effeltrich, Germany), operated in the differential mode under controlled conditions of temperature and PFD, and at external CO_2 (normally fairly constant at around 350 μl l^{-1}). Illumination was provided by a fibre-optic lamp (Kaltlicht-Fiberleuchte FL-400 with Spezial-Fiberoptik 400-F; Walz) with which homogeneous light distribution is achieved by using a source made of 200 1-mm diameter plastic fibres arranged in a regular pattern within a circle of 95 mm diameter. Changes in PFD were made with no, or only minor, alteration to spectral energy distribution by using neutral density filters and adjustment of lamp power. Carbon dioxide exchange (NP and DR) was related to projected surface area (μmol CO_2 m^{-2} s^{-1}). Total Chl content (determined as in Schroeter et al. 1995 after specimens returned to Germany) and oven dry weight (24 h at 105 °C) were also measured and relevant data are given in the figure captions. Water content of the lichens was related to thallus dry weight.

Fluorescence. A pulse amplitude modulated fluorometer (PAM-2000; Walz) was used with the fibre-optic probes permanently fixed in the cuvettes about 10 mm from the sample. Standard routines programmed within the machine were used to determine effective PSII quantum yield (Φ_{PSII}), potential maximal PSII quantum yield [Fv/Fm = variable fluorescence/maximal fluorescence in dark-adapted state, where Fv = Fm − Fo (maximal fluorescence − minimal fluorescence in dark-adapted state)], photochemical (qP) and nonphotochemical quenching (qN), as in Schreiber and Bilger (1993). Relative electron transport rate (ETR) can be obtained as PFD × Φ_{PSII} (Bilger et al. 1995).

Experimental routine. The lichen samples were hydrated by first submerging them in water for several minutes, followed by shaking to remove adhering water drops, and light blotting. The samples were then enclosed in the gas-exchange cuvette and allowed to equilibrate in the dark to the chosen temperature. Temperatures from 0 °C to 25 °C were used with 5 °C steps. Once the selected temperature had been reached and maintained for 15 min a zero reading was taken and then the CO_2 differential after a 5 min settling time. Then Fv / Fm was measured. Further measurements of CO_2 exchange were made at 400, 1000 and 2000 μmol m^{-2} s^{-1}. At each PFD, 15 min was allowed to reach the steady state and then a zero reading was made; after a further 5 min the CO_2 differential and Φ_{PSII} were measured; the light was then turned off, the PAM-2000 far-red light turned on and Fo′ immediately determined in order to calculate qP and qN. The lichen samples were rehydrated between each temperature step. Moss samples were measured at a single, nearly saturating PFD of 400 μmol m^{-2} s^{-1}. The temperature steps were then made sequentially without removing the sample. Care was taken with both lichen and moss to ensure that the sample temperature was held at the chosen value at all times. The response of CO_2 exchange to PFD was also measured at selected temperatures.

Results

Moss – Bryum argenteum. The CO_2 exchange showed a response to increasing temperature that is typical for antarctic mosses (Longton 1988). At a constant PFD of 400 μmol m^{-2} s^{-1}, NP was maximal between 5 and

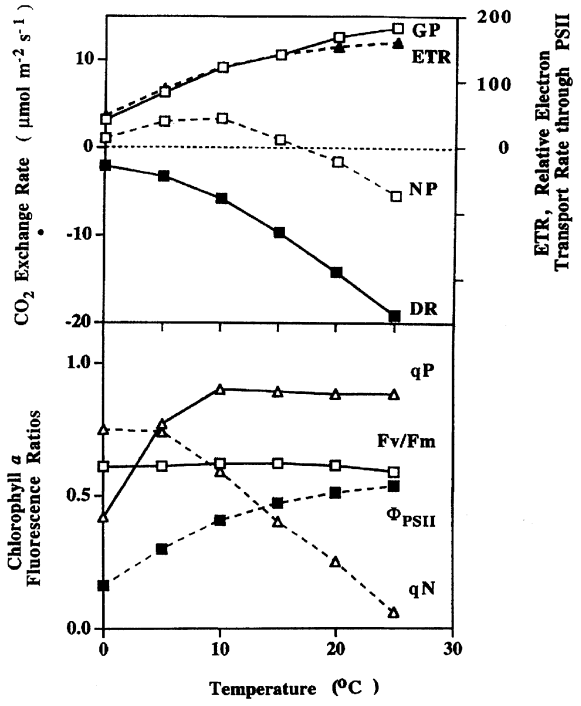

Fig. 1. CO_2 exchange (µmol CO_2 m^{-2} s^{-1}) and Chl *a* fluorescence measurements for *Bryum argenteum* measured at a constant PFD of 400 µmol m^{-2} s^{-1} at temperatures from 0 to 25 °C. Measurements were made as described in the text. ETR, GP, NP and DR upper panel: Chl *a* fluorescence parameters, qP, lower panel: Fv / Fm, Φ_{PSII} and qN. Thallus parameters: chlorophyll content, 306 mg Chl m^{-2}; chlorophyll *a*/*b* ratio, 1.90; dry weight, 1164 g (DW) m^{-2}

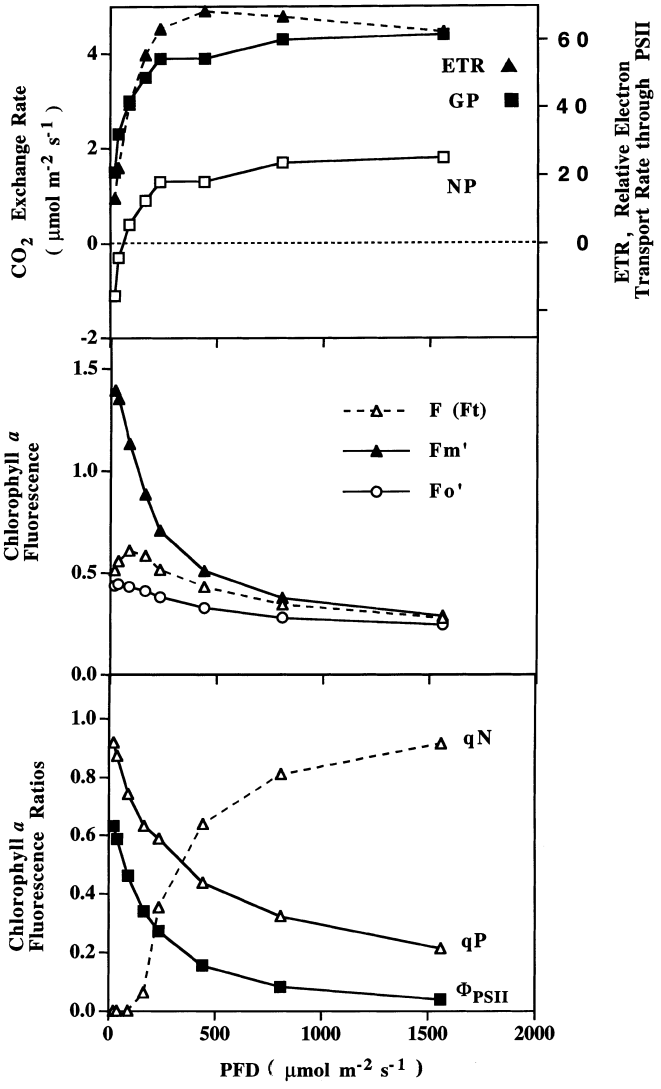

Fig. 2. CO_2 exchange (µmol CO_2 m^{-2} s^{-1}) and Chl *a* fluorescence measurements for *Bryum argenteum* measured at a constant temperature of 15 °C and a PFD of 0–1600 µmol m^{-2} s^{-1}. *Upper panel*: ETR and CO_2 exchange measurements; *middle panel*: Chl *a* fluorescence values; *lower panel*: Chl *a* fluorescence ratios. Labels for the lines are given in each panel. Thallus parameters: Chl content, 397 mg Chl m^{-2}; Chl *a*/*b* ratio, 1.99; dry weight, 1284 g (DW) m^{-2}

10 °C then declined with further increase in temperature to be negative above about 16 °C (Fig. 1). Changes in NP were apparently caused by a 10-fold increase in DR between 0 °C and 25 °C. Gross photosynthesis (GP) increased steadily with temperature and was maximal at 20–25 °C while ETR almost exactly paralleled the changes in GP. Fluorescence parameters also varied markedly with temperature (Fig. 1); Φ_{PSII} was lowest at 0 °C, around 0.15, then rose steadily with temperature to about 0.5 at 20–25 °C. In contrast, Fv/Fm remained almost constant at around 0.6 (Fig. 1). Minimal fluorescence in the light (Fo′) was also constant but maximal fluorescence in the light (Fm′) increased at temperatures above 5 °C. Consequently, qP rose from about 0.4 at 0 °C to a relatively constant 0.85–0.9 at 10 °C and above; qN was highest at low temperatures and fell steadily to near zero at 25 °C (Fig. 1).

The response of CO_2 exchange to PFD was a typical saturation curve with maximal values being reached at about 400 µmol m^{-2} s^{-1} (Fig. 2). Again, ETR almost exactly paralleled the changes in GP. Both qP and Φ_{PSII} showed an expected decline with the increase in PFD, and qN showed a typical increase as PFD saturation was exceeded. The values for Fm′ decreased with increase in PFD, as expected.

Changes in ETR with temperature and PFD were unrelated to NP but had a linear relationship (R^2 = 0.98) with GP (Figs. 3B,D). There was also a strong linear relationship (R^2 = 0.88 – 0.98) between Φ_{PSII} and Φ_{CO_2} whether the values were generated by changes in PFD or temperature (Figs. 3A,C). Deviation from linearity only occurred at high values of yield i.e. at low PFD below about 35 µmol m^{-2} s^{-1} (Fig. 3A).

Lichen – Umbilicaria aprina. The response of CO_2 exchange to temperature was measured on several samples from 0 to 25 °C at several different PFDs. The values for NP, DR and GP followed a similar pattern to that found for *B. argenteum* (Fig. 4). The GP was maximal at 15 °C to 20 °C (depending on sample and PFD) and DR increased markedly with temperature. The NP had a much broader response being near-optimal over the range 5 °C to 20 °C and, at a PFD of 2000 µmol m^{-2} s^{-1}, was still positive at 25 °C, the highest temperature tested (Fig. 4 and other data not

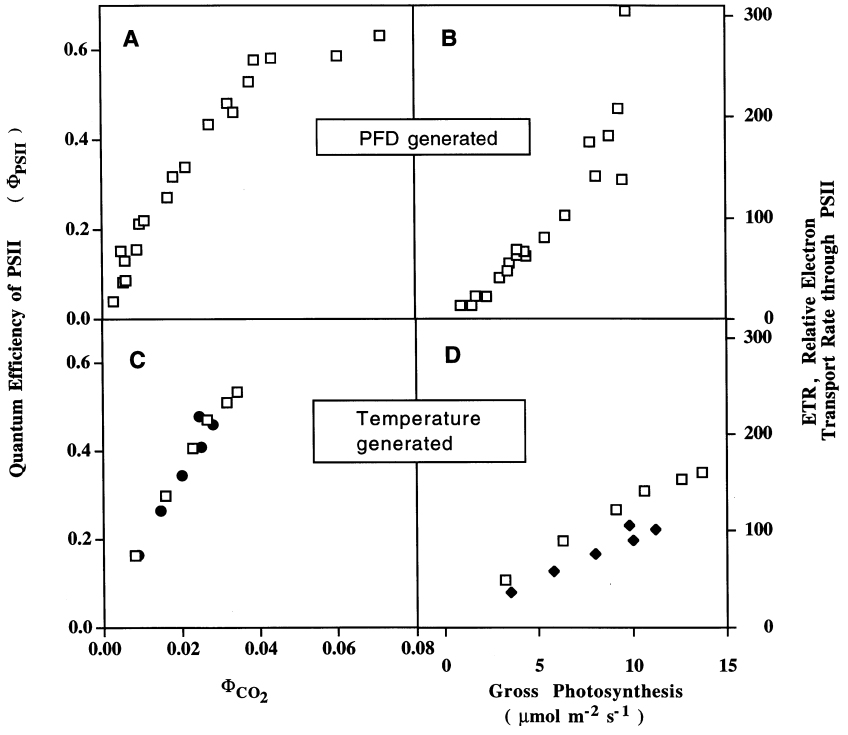

Fig. 3. Relationship between CO_2 exchange (μmol CO_2 m^{-2} s^{-1}) and Chl *a* fluorescence measurements for *Bryum argenteum*. *Upper panels*: PFD-generated relationship (at a constant 15 °C); *lower panels*: temperature-generated relationship (constant PFD of 400 μmol m^{-2} s^{-1}). **A, C** Φ_{PSII} (quantum efficiency of PSII) versus Φ_{CO_2} (quantum efficiency for CO_2 exchange, CO_2 exchange rate divided by PFD at selected values for PFD). **B, D** ETR versus GP. Data obtained from measurements in Figs. 1 and 2. Thallus parameters given in Figs. 1 and 2

shown). Fluorescence parameters, with the exception of Fv / Fm which was nearly constant at all temperatures, were very different from those of the moss. Despite the large changes in GP as temperature increased, qP remained almost stable and high up to 1600 μmol m^{-2} s^{-1} (Fig. 4). The values for qN were variable between experiments but were always higher at low temperatures (Fig. 4) and, where CO_2 exchange had been measured at more than one PFD, then qN was larger at higher PFDs (Figs. 4,5). Because Φ_{PSII} declined slightly with increase in temperature at both 400 and 1600 μmol m^{-2} s^{-1} PFD (Fig. 4), ETR showed little apparent correlation with CO_2 exchange at either PFD. A lower ETR actually occurred at the higher temperatures when GP values were close to maximal (Fig. 4).

Carbon dioxide exchange, measured at both 5 °C and 15 °C with PFDs from 0 to 2000 μmol m^{-2} s^{-1}, was not saturated at the highest tested PFD (Fig. 5); both GP and the PFD compensation point were higher at 15 °C. The values for qP declined with increase in PFD at 5 °C

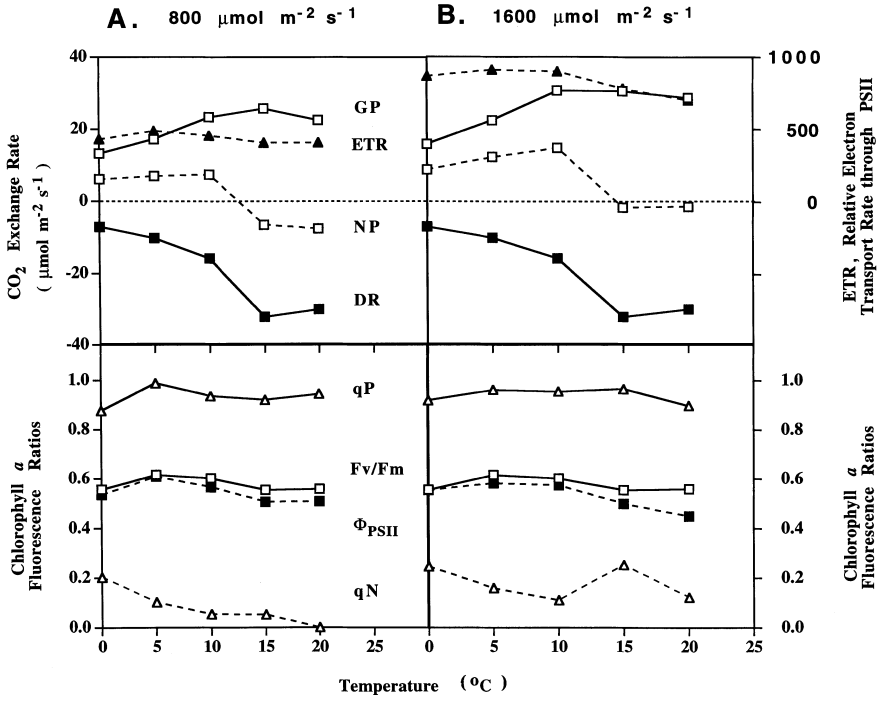

Fig. 4A,B. CO_2 exchange (μmol CO_2 m^{-2} s^{-1}) and Chl *a* fluorescence ratios for *Umbilicaria aprina* determined at a constant PFD of 800 μmol m^{-2} s^{-1} and 0–25 °C (**A**) and 1600 μmol m^{-2} s^{-1} and 0–20 °C (**B**). *Upper panels*: ETR, GP, NP and DR; *lower panels*: qP, Fv/Fm, Φ_{PSII} and qN. Thallus parameters: Chl content, 495 mg Chl m^{-2}, Chl *a*/*b* ratio, 3.84; dry weight, 524 g (DW) m^{-2}

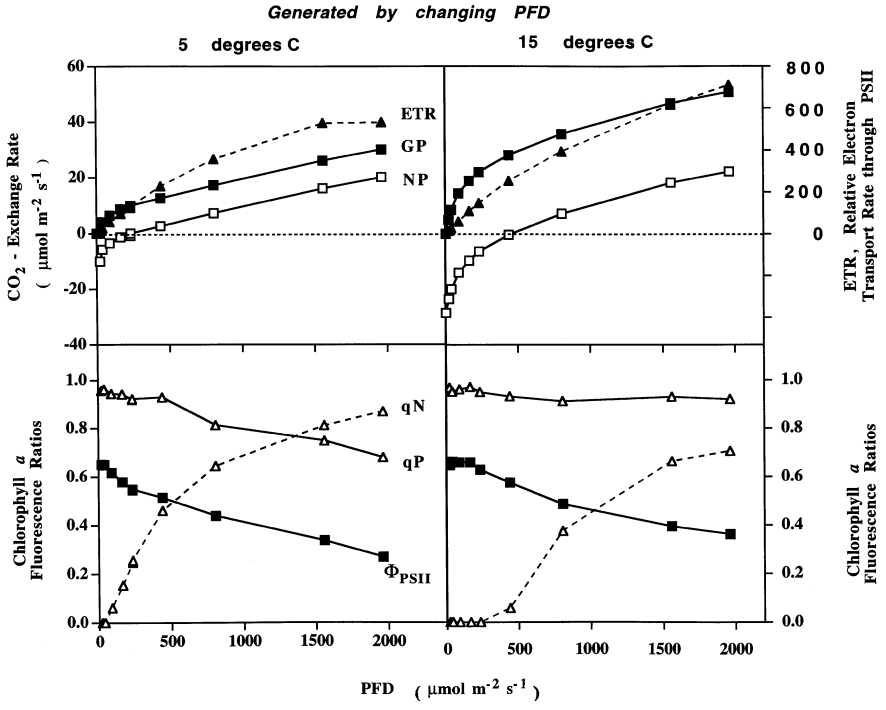

Fig. 5. CO_2 exchange (µmol CO_2 m^{-2} s^{-1}) and Chl *a* fluorescence measurements for *Umbilicaria aprina* measured at two constant temperatures, 5 and 15 °C, and PFDs from 0 to 2000 µmol m^{-2} s^{-1}. *Upper panel*: ETR, GP and NP; *lower panel*: Chl *a* fluorescence ratios (qP, Φ_{PSII} and qN). Labels for lines are given in the left-hand panels. Thallus parameters: chlorophyll content, 313 mg Chl m^{-2}; Chl *a*/*b* ratio, 2.91; dry weight, 276 g (DW) m^{-2}

but remained constant at a high value, more than 0.9, at 15 °C; qN was zero at low PFD and, as expected, rose with increase in PFD (Fig. 5). At both temperatures, Φ_{PSII} declined with increase in PFD.

The ETR: CO_2 exchange relationship was much more complex than found for *B. argenteum* and was strongly influenced by temperature (Fig. 6). The ETR did not give any indication of GP unless temperature was held constant. Even then the relationship ranged from changing ETR, but almost constant GP, at 0 °C, through a positive, linear relationship at intermediate temperatures to declining ETR with increasing GP at 20 °C (Fig. 6, left panel). At constant PFD (Fig. 6, right panel), ETR declined only slightly with increase in GP at 400 µmol m^{-2} s^{-1} and declined strongly with increasing GP at 1000 and 2000 µmol m^{-2} s^{-1} PFD. It should be noted (Fig. 6) that 20 °C represents some form of optimal value which coincides with the optimal temperature for GP. In a more detailed analysis at 5 °C and 15 °C with PFD from zero to 2000 µmol m^{-2} s^{-1} a nonlinear response was found for both the ETR response to GP and the Φ_{PSII} response to Φ_{CO_2} (Fig. 7, full data in Fig. 5). In both cases the nonlinearity was predominantly at low PFD, under about 400 µmol m^{-2} s^{-1}. Below about 200 µmol m^{-2} s^{-1} there was no change in Φ_{PSII} with increase in Φ_{CO_2} (Fig. 7, right panel).

Discussion

The objective of this work was to clarify, using a moss and lichen, the relationship between photosynthetic measurements made with two different systems: the standard gas-exchange methodology and the more rapid Chl *a* fluorescence obtained with a pulse amplitude modulated fluorometer. If ETR, as measured by fluorescence proved to be a good indicator of CO_2 exchange

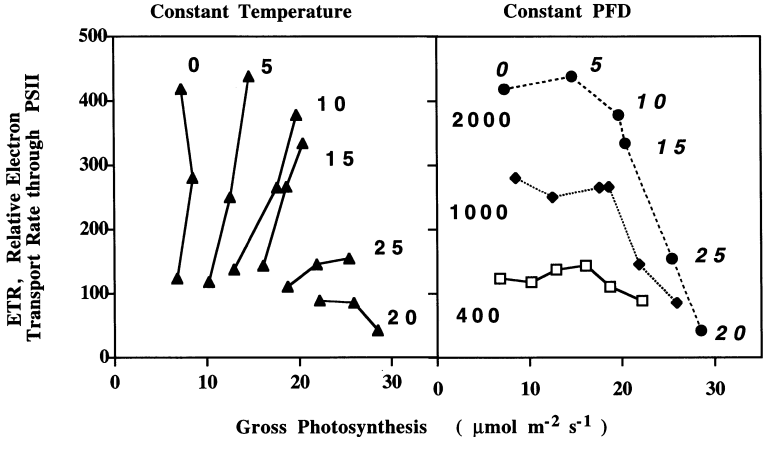

Fig. 6. Relationship between ETR and GP for *Umbilicaria aprina*. *Left panel*: the lines link data points at constant temperatures (individual lines labelled with temperature 0–20 °C) and three PFDs (400, 1000 and 2000 µmol m^{-2} s^{-1}). *Right panel*: the lines link data points at constant PFD (individual lines labelled with PFD) and temperatures from 0 to 25 °C. In both panels the temperatures are marked next to the lines. Thallus parameters as for Fig. 4.

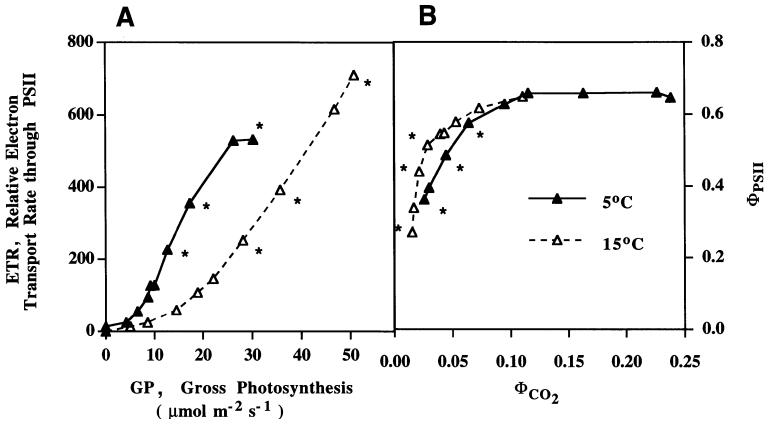

Fig. 7A. Relationship between GP and ETR ($\Phi_{PSII} \times$ PFD) for *Umbilicaria aprina*. Data generated at two constant temperatures of 5 (▲) and 15 °C (△) and at PFDs from 0 to 2000 μmol m^{-2} s^{-1}. Other data from this experiment are given in Fig. 5. The *stars* mark the position of data at 400, 1000 and 2000 μmol m^{-2} s^{-1} PFD, from left to right respectively. **B** Relationship between effective quantum efficiency of PSII (Φ_{PSII}) and the quantum efficiency for CO_2 exchange (Φ_{CO_2}) for *Umbilicaria aprina*. Data generated at two constant temperatures of 5 (▲) and 15 °C (△) and at PFDs from 0 to 2000 μmol m^{-2} s^{-1}. Other data from this experiment are given in Fig. 5. The *stars* mark the position of data at 2000, 1000 and 400 μmol m^{-2} s^{-1} PFD, from left to right respectively (reverse of order in **A**). Thallus parameters as for Fig. 5

then ecophysiologists would have a rapid and extremely useful method with which to monitor photosynthetic activity. The system would be particularly beneficial when studying cryptogams, like lichens and mosses, which are often closely associated with their substrates (see Schroeter et al. 1992; Schroeter 1994).

The results for the moss suggested that the fluorometer could, indeed, provide an excellent indication of photosynthetic activity. The values for GP and ETR were linearly related whether data were generated by changes in temperature or PFD. Similarly, Φ_{PSII} versus Φ_{CO_2} showed an almost perfect linear relationship (Fig. 3A,C) with only slight deviation at very low PFD (19 μmol m^{-2} s^{-1}). The excellent relationship between ETR and GP and the lack of relationship between ETR and NP suggest that electron flow through PSII equals (NP + DR) and it is generally accepted, in bryophytes and lichens, that GP can be obtained as (NP + DR). However, it is a little surprising that the relationship between GP and ETR was so good for *B. argenteum*. Mosses are typically regarded as normal C_3 plants with the presence of photorespiration being indicated by the high CO_2 compensation values and strong inhibition of photosynthesis at 21% oxygen, both of which increase at higher temperatures (Green and Lange 1994). From higher-plant studies we would expect a curvilinear relationship for *B. argenteum* both between ETR and GP, and between Φ_{PSII} and Φ_{CO_2}. The excellent relationship actually found is more similar to those for C_4 plants and for C_3 plants at low oxygen concentration (Genty et al. 1989; Seaton and Walker 1990; Krall and Edwards 1991; Krall et al. 1991; Krall and Edwards 1992).

We suggest that the excellent relationship is merely coincidental and, because moss tissue is nearly 100% photosynthetic, that DR is not only suppressed but is almost exactly compensated for by photorespiratory CO_2 release. Whether DR does continue in the light in photosynthetic tissue is still uncertain (Raven 1984, Amthor 1994) but the balance of opinion is that it does not. One indicator of DR suppression is the Kok effect (Kok 1948) where there is very high quantum efficiency for CO_2 fixation at PFDs below the compensation point. It is interesting, therefore, that the only values not on the linear fit for Φ_{PSII} versus Φ_{CO_2} (Fig. 3A) are the two points obtained below the PFD compensation point. This would support suppression of DR in the moss. Although the respiratory energy demand of the tissues will rise with increase in temperature this would be met by the photosystems, in particular be cyclic photophosphorylation by PSI, and this would not be detected as electron flow by the fluorometer which measures PSII fluorescence. However, photorespiration, which is expected to be present, will rise rapidly with temperature (Green and Lange 1994) and this will cause increased ETR since photorespiration occurs after CO_2 fixation. The present evidence does not allow us to decide whether this interpretation is correct; possibly, a combination of partial DR suppression and photorespiration is involved.

Although ETR might be an indicator of GP (albeit NP plus photorespiratory release) it gives no guide to the net carbon status of the moss. While ETR was greatest at the highest temperatures measured, at the same time, NP was at its most negative (Fig. 1). It would still be necessary to employ a gas-exchange system together with Chl *a* fluorescence in order to obtain a full understanding of the CO_2 balance of a moss.

The results from the lichen were much more complex and much more difficult to interpret. The values for ETR were nonlinearly related to GP and of different slope at each temperature (Figs. 4,6,7). At any PFD, ETR was actually lowest at the higher temperatures when GP was near maximal (Figs. 4,6). The relationships were again nonlinear when Φ_{PSII} and Φ_{CO_2} were compared (Fig. 7B). These results are surprising since, bearing in mind the results from higher plants (Harbinson et al. 1990; Krall and Edwards 1991; Krall et al. 1991), a linear relationship might well be expected for lichens because they often show little evidence of photorespiration, can have very low CO_2 compensation values and can have CO_2 concentrating mechanisms (Snelgar and Green 1980; Green and Snelgar 1981; Badger et al. 1993; Palmqvist et al. 1994).

We suggest that the patterns could result in part, as in the moss, from a suppression of DR in the light. Lichens are a double organism and only the DR of the photobiont would be suppressed in the light; mycobiont DR would continue although, possibly, at a slower rate

(Kappen and Lange 1972). There was some evidence of a Kok effect for *U. aprina*; in particular, the nonlinear portions of the ETR versus GP and the Φ_{PSII} and Φ_{CO_2} responses corresponded to the data points obtained below NP compensation. Moreover, the nonlinear portion of the ETR versus GP curve was more extended at 15 °C than 5 °C in about the same proportion as the increase in DR (Fig. 7A). The decline in ETR at the higher temperatures, when GP is near maximal, is a very unusual and unexpected feature of the lichen results. It is, perhaps, of relevance that when GP declined at the highest temperature (25 °C) then ETR started to recover (Fig. 6). There seems to be some other system accepting electrons with a rate that is inversely proportional to GP; possibly, this could be electron transfer to oxygen. The response of CO_2 exchange to PFD showed that *U. aprina* was not saturated at even 2000 μmol m^{-2} s^{-1} (Fig. 6). At any chosen PFD, Φ_{PSII} values were also much higher for *U. aprina* than for *B. argenteum*, indicating unsaturated photosystems. This tendency was also shown for *U. aprina* with measurements of net photosynthesis in the field by Kappen et al. (1998). Green and Lange (1994) concluded that lichens are predominantly shade plants and that there is decreased light at the photobiont by use of filters. Measurements are rare, but Buedel and Lange (1994) found a PFD of only a few percent of ambient at the photobiont level.

In both species the photosystems seemed to be protected when photosynthesis was limited by low temperatures. At the lower temperatures, *B. argenteum* had a high qN which declined as the temperature increased. The lichen had a much broader range for near-optimal NP; however, qN was lower in the range 10–15 °C, suggesting again some protection at low temperatures. In both lichen and moss, qP was high and stable across most of the temperature range, again suggesting reaction centres are kept open by means other than CO_2 fixation such as, perhaps, electron transfer to oxygen. Leisner et al. (1997) found similar high qP and ETR values when NP was low, due to high water content, in field studies on the green algal lichen *Lecanora muralis*.

It has been suggested that Fv/Fm is affected by temperature and is therefore an unsatisfactory parameter for measuring if thallus temperature changes (Hovenden and Seppelt 1995). We found that Fv/Fm was relatively stable across the entire temperature range for both the moss and the lichen (Figs. 1,6). It would seem, therefore, that this problem is not serious and that the ratio can be successfully measured in the field even when temperature changes might be expected during sample darkening. On some occasions, for *Umbilicaria aprina* it was noted that the dark-adapted Fv/Fm was actually lower than Φ_{PSII} measured at a PFD up to 230 μmol m^{-2} s^{-1}. A similar result has been reported from cyanobacterial lichens and seems to indicate a quenching system that operates in the dark and reduces the Fv/Fm ratio (Sundberg et al. 1997). As in cyanobacteria, it appears that the green algal *U. aprina* does not show optimal quantum efficiency at zero PFD but rather over a range of low PFD.

Overall, the comparison of CO_2 exchange and Chl *a* fluorescence parameters has revealed important information about the lichen and moss. Neither appear to behave in a manner entirely similar to higher plants. The moss showed an excellent relationship between ETR and GP but this seems to be coincidental. No indication of NP (i.e. CO_2 balance) could be obtained from fluorescence. For both the lichen and the moss we think that DR could be suppressed in the light and that ETR is affected by other processes, such as photorespiration and electron transfer to oxygen. Both lichen and moss appeared to be well adapted to the cold and both retained in relatively high qP across the entire temperature range studied. The moss seemed to handle excess light energy at low temperatures by a nonphotochemical quenching system. It is not entirely clear how the lichen photosystem responded to low temperatures but qN was again higher. The results have revealed the dangers of extrapolating from higher-plant studies to other plants such as bryophytes and lichens. This is in agreement with growing evidence that lichens and bryophytes have photosynthetic mechanisms different from those of higher plants (Palmqvist et al. 1994; Smith and Griffiths 1996). At present it would seem safer to limit fluorescence measurements to a predominantly monitoring role in ecophysiological studies (Schroeter et al. 1991) except when they are carefully linked to gas-exchange studies or used to assess photosystem status. Considerable work clearly remains to be done in order to obtain a better understanding of the photosynthetic behaviour of these poikilohydric plants with their exceptional stress resistance.

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