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Studies on the photosynthesis of the terrestrial cyanobacterium *Nostoc flagelliforme* subjected to desiccation and subsequent rehydration

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The photosynthetic response of *Nostoc flagelliforme* to desiccation and rehydration was investigated. The initial, variable and maximal chlorophyll fluorescence increased from the beginning of desiccation (fully hydrated), reached maxima at a water loss of about 35% and then decreased with further water loss. The area over the fluorescence induction curve reached a maximum at a water loss of about 70%. Pigment contents of chlorophyll *a*, phycocyanin and allophycocyanin were constant during the photosynthetic recovery phase following rehydration. A fast initial phase of recovery was completed within half an hour after rehydration, and further changes in 77 K fluorescence emission spectra were mainly related to the fluorescence level at 695 nm. A decrease in fluorescence intensities from phycobiliproteins was not detected from 0.5 to 24 h. The initial fluorescence was high at the beginning of rehydration and decreased sharply during photosynthetic recovery. This decrease appeared to be mainly related to the activation or repair of photosystem II (PS II) rather than changes in the coupling and assembly of phycobilisomes. The change of variable fluorescence and the PS II photochemical efficiency value (F_v/F_m) correlated well with the recovery of PS II activity during rehydration. *Nostoc flagelliforme* required light and *de novo* protein synthesis to recover its photosynthetic activity fully during rehydration. In darkness, F_v/F_m recovered 3.4% of activity in dried field samples but 18.9% of activity in samples dry for 2 days. When light was provided in the presence of chloramphenicol, F_v/F_m recovered 10.1% or 39.5% of activity in dried field samples and in samples dry for 2 days, respectively.

INTRODUCTION

Desiccation affects the growth, development, metabolism and yield of plants. Plants growing under desiccating conditions may have adapted to survive in ways such as stress avoidance and tolerance. There has been considerable interest in the desiccation tolerance of terrestrial *Nostoc* Vaucher species over the past 20 years (Potts 1994, 2000). These organisms are subjected to repeated cycles of wetting and drying, interspersed with variable periods of desiccation in their habitats (Gao 1998). They can remain desiccated for months or years and recover metabolic activity fully within hours to days upon rehydration (Dodds *et al.* 1995). The time required for maximal photosynthetic and respiratory activities to recover after rehydration is dependent on the time of storage in dryness. Longer storage requires more time for recovery (Scherer *et al.* 1984).

The photosynthetic recovery of *Nostoc commune* Vaucher in relation to the amount of water absorbed or time after initiation of rehydration has been investigated recently (Satoh *et al.* 2002). *Nostoc commune* is cosmopolitan and occurs as parchment-like colonies, especially in limestone or karst regions (Potts 2000). *Nostoc flagelliforme* (Berkeley & Curtis) Bornet & Flahault is a highly desiccation-tolerant cyanobacterium with hair-like appearance (Gao 1998). It occurs in environments subject to high rates of water loss for a majority of the time, but becomes saturated due to temporary rain or night-time dew for shorter periods (Whitton *et al.* 1979; Gao 1998). *Nostoc flagelliforme* absorbs and loses water much faster than *N. commune* (Whitton *et al.* 1979; Scherer *et al.* 1984). Fast water uptake and rapid water loss are important ecologically and physiologically for growth of *N. flagelliforme* in the dry areas where other terrestrial *Nostoc* species hardly survive (Gao 1998). The photosynthetic recovery of *N. flagelliforme* has been correlated with the recovery of energy charge (Scherer *et al.* 1986) and requires addition of exogenous potassium (Qiu & Gao 1999; Qiu *et al.* 2004). However, the details of photosynthetic recovery in *N. flagelliforme* remain unknown.

The photosynthetic apparatus is very sensitive and liable to injury, and needs to be maintained or quickly repaired upon rehydration (Godde 1999). Its physical properties are of crucial importance in desiccation-tolerant plants (Alamillo & Bartels 2001). Upon reintroduction of water, the metabolic recovery of desiccated cells depends on the protective mechanisms set in place during drying and on the repair mechanisms taking place during and following rehydration (Oliver & Bewley 1997). The recovery of photosynthesis in bryophytes after desiccation can be remarkably rapid and appears to leave little time for repair processes requiring synthesis of proteins or other cell components on any substantial scale (Proctor & Smirnoff 2000). However, far less attention has been paid to the effect of protein synthesis on the photosynthetic recovery of *N. flagelliforme* from desiccation.

In the present paper, the photosynthetic recovery of *N. flagelliforme* during rehydration and the loss of photosynthetic activities by drying are characterized. Studying these process-

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es might provide useful information about the mechanisms behind the reasons why *N. flagelliforme* is tolerant to desiccation and about its ability to recover during rehydration.

MATERIAL AND METHODS

Material and experimental procedure

Nostoc flagelliforme was collected at Sunitezuoqi, Inner Mongolia, air-dried and stored dry for 2 years until used for experiments. Samples were rehydrated in BG_{11} medium at 25°C as previously described (Qiu & Gao 2001). Chloramphenicol (CMP; Roche Applied Science, Indianapolis, IN, USA) was used as an inhibitor of protein synthesis at 2.5 mM.

Chlorophyll fluorescence measurements

The polyphasic rise in fluorescence transients was measured by a Plant Efficiency Analyser (PEA; Hansatech Instruments, King's Lynn, UK) with an actinic light of 1500 μmol m^{-2} s^{-1} (Lu & Vonshak 1999). Illumination was obtained from an array of six high-intensity light-emitting diodes (with a peak wavelength of 650 nm) focused on the sample surface to provide homogeneous illumination over an area 4 mm in diameter. Samples occupied the whole of the illuminated area and were dark-adapted for 15 min before measurements. All fluorescence transients were recorded within a time span of 70 μs to 5 s with a data acquisition rate of 10^5 readings s^{-1} for the first 2 ms, 10^3 readings s^{-1} after the first 2 ms and 10 readings s^{-1} after the first second. The difference in the area over the fluorescence rise curve with and without 3-(3′,4′-dichlorophenyl)-1,1-dimethylurea (DCMU) can be used to measure the size of the photosystem II (PS II) electron acceptor pool, i.e. the plastoquinone (PQ) pool size. Because the fluorescence induction curve has been obtained in the absence of DCMU but with the sufficiently strong actinic light used here, the area over the induction curve is proportional to the number of electrons that have passed through the primary electron acceptor of PQ (Q_A) to the PQ pool before the latter becomes completely reduced (Falkowski & Raven 1997).

77 K fluorescence emission spectrophotometry

Dried field samples were rehydrated in BG_{11} medium at 40 μmol m^{-2} s^{-1} and 25°C. The 77 K fluorescence emission spectra under 580 nm excitation were measured with a Hitachi F-4500 fluorescence spectrophotometer (Hitachi, Tokyo, Japan).

Measurement of PS II activity

Dried field samples (0.1 g dry weight) were rehydrated in BG_{11} medium at 40 μmol m^{-2} s^{-1} and 25°C for a certain period, and then cut into filaments less than 0.5 cm in length. The electron transport activity of PS II was determined with H_2O as the electron donor and *p*-benzoquinone (*p*-BQ) as the electron acceptor in 1.5 ml reaction mixtures containing 25 mM Tricine–NaOH (pH 7.8), 0.2 mM sucrose, 5 mM NaCl, 5 mM $MgCl_2$ and 5 mM *p*-BQ. In this system, electrons are transported from water to *p*-BQ through the Mn cluster, P680, pheophytin, Q_A and the secondary electron acceptor of PQ (Q_B).

Light-induced evolution of oxygen was monitored with a Clark-type oxygen electrode (Chlorolab 2; Hansatech Instruments). Temperature was controlled at 25°C with a polystat refrigerated bath (Cole-Parmer Instrument Co., Vernon Hills, IL, USA). Illumination was provided by a high-intensity white light source (LS2; Hansatech Instruments) and passed through a fibre-optic cable (A8; Hansatech Instruments) to the surface of the reaction cuvette. Irradiance was measured with a quantum sensor (QRT1; Hansatech Instruments), and the intensity was adjusted to 950 μmol m^{-2} s^{-1} with neutral density filters.

Determination of pigment content

Photosynthetic pigments were assayed in samples rehydrated as above for certain periods. Chlorophyll *a* content was determined with 80% acetone (Inskeep & Bloom 1985). The biliproteins were extracted by repeated freezing and thawing in the presence of 0.05 M phosphate buffer (mixing equal volumes of 0.1 M NaH_2PO_4 with 0.1 M Na_2HPO_4 solutions). Samples were ground in freezing buffer with sand as an abrasive and the extraction process was repeated three times. The homogenized solutions were centrifuged at 4000 × *g* for 15 min, and then the absorbance of the supernatant was determined. The concentrations of phycocyanin and allophycocyanin were calculated according to Siegelman & Kycia (1978).

Determination of water loss

The initial wet weight of fully recovered samples was determined after blotting to remove excess water. They were air-dried at room temperature (23–28°C) and instantaneously weighed to assess water loss. Dry weight was determined after the samples had been dried at 80°C for 24 h and cooled down in a desiccator. Water loss (WL) was calculated as follows:

$$WL = (W_w - W_t)/(W_w - W_d) \times 100\%,$$

where W_d is the dry weight, W_t the instantaneous weight of samples measured at certain intervals and W_w the initial wet weight.

RESULTS

Responses of chlorophyll fluorescence to desiccation

The initial fluorescence (F_0), variable fluorescence (F_v) and maximal fluorescence (F_m) showed similar tendencies to water loss (Fig. 1). They increased from the beginning of desiccation (fully hydrated), reached a maximum at a water loss of about 35% and then decreased with further water loss. The PS II photochemical efficiency (F_v/F_m) began to decline when water loss was higher than 70%. The time required for the appearance of maximal fluorescence (T_{fm}) is a function of the rate of photochemical reaction and the pool size of electron acceptors. T_{fm} increased with water loss. The area over the induction curve, which is proportional to the number of electrons that have passed through Q_A to the PQ pool before the latter becomes completely reduced, reached a maximum at a water loss of about 70% and then decreased rapidly with further water loss.

Fast fluorescence induction curves during rehydration

The fluorescence transient can be used as a quick monitor of the electron acceptor side reactions, the pool heterogeneity

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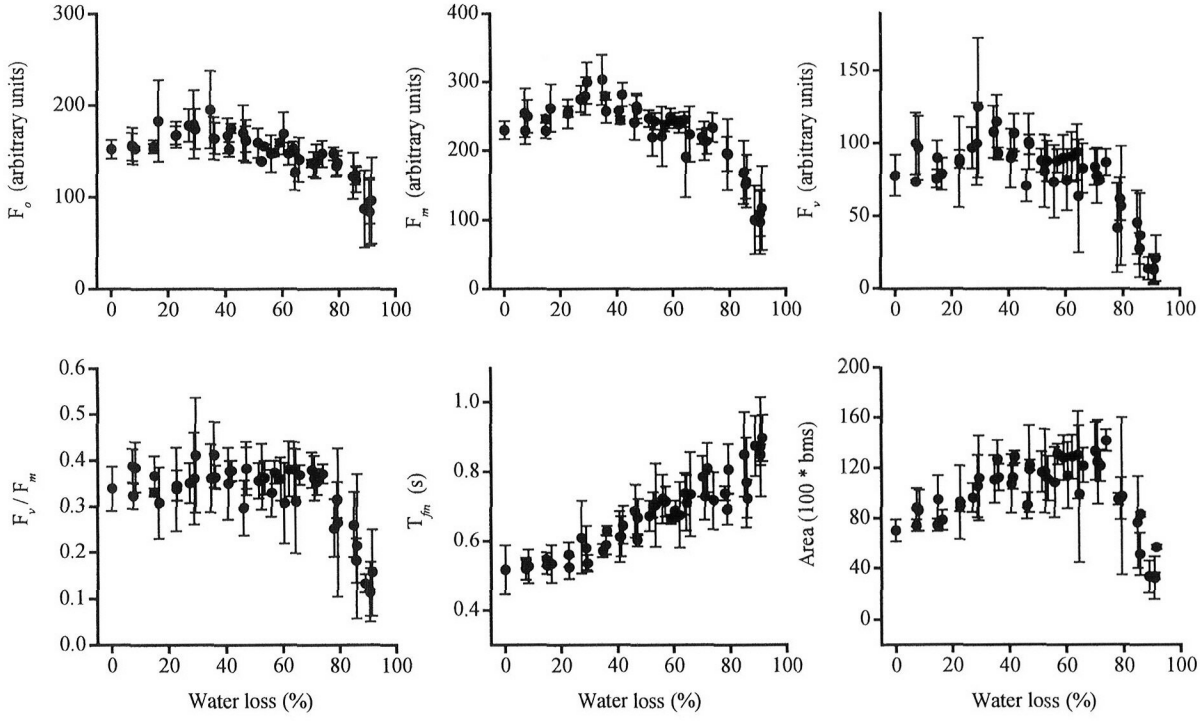

Fig. 1. Changes of fluorescence parameters with water loss from *Nostoc flagelliforme*. Dried field samples were rehydrated in BG_{11} medium at 40 μmol m^{-2} s^{-1} and 25°C for 26 h, and then air-dried at room temperature (23–28°C). Area, the area over the fluorescence rise curve. Data indicate $\bar{x} \pm s$ ($n = 3$).

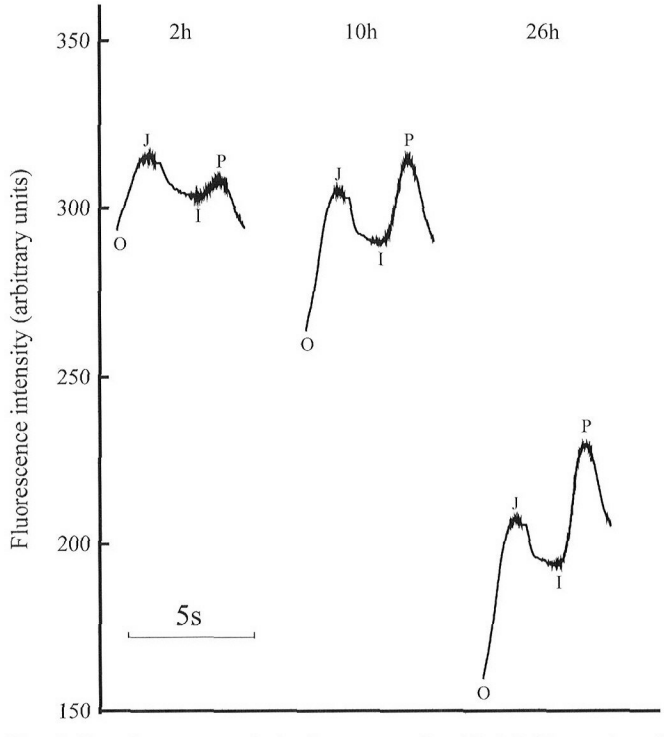

Fig. 2. Fast fluorescence induction curves for dried field samples of *Nostoc flagelliforme*. Samples were rehydrated in BG_{11} medium at 40 μmol m^{-2} s^{-1} and 25°C for 2, 10 or 26 h. Each curve indicates the mean of nine measurements.

and pool sizes, and the effects of inhibitors and mutations on these processes, as well as on the donor side (Govindjee 1995). During the rehydration of dried field samples, the fluorescence transients were followed and they exhibited a typical polyphasic rise including phases O, J, I and P (Fig. 2). The initial fluorescence at level O reflects the minimal fluorescence yield when all molecules of Q_A are in the oxidized state. Level P corresponds to the situation in which all molecules of Q_A are in the reduced state. The transition from phase O to J is controlled by photochemical charge separation leading to the reduction of Q_A, whereas the appearance of phase I and P is limited by dark reactions. The initial fluorescence was high at the beginning of rehydration. Its decrease was accompanied by an increase in the variable fluorescence during the photosynthetic recovery. The fluorescence signals rose faster at the initial phase of rehydration and the fluorescence yields remained higher compared with those for fully rehydrated samples. Thus, samples suffered from a block in the electron flow downstream of PS II during the initial phase of rehydration and the inhibitory site of desiccation could be at the acceptor side of PS II.

Characteristics of 77 K fluorescence emission spectra during rehydration

In dry samples, fluorescence intensities of *N. flagelliforme* were quite low. Within 0.5 h of rewetting, the emission peak at 645 nm associated with phycocyanin and the fluorescence level of allophycocyanin (the shoulder at around 658 nm) increased, respectively, to 95% and 86% of the value attained after 24 h. The intensity of fluorescence at 685 nm was emitted from the chlorophyll of PS II and the core-membrane link-

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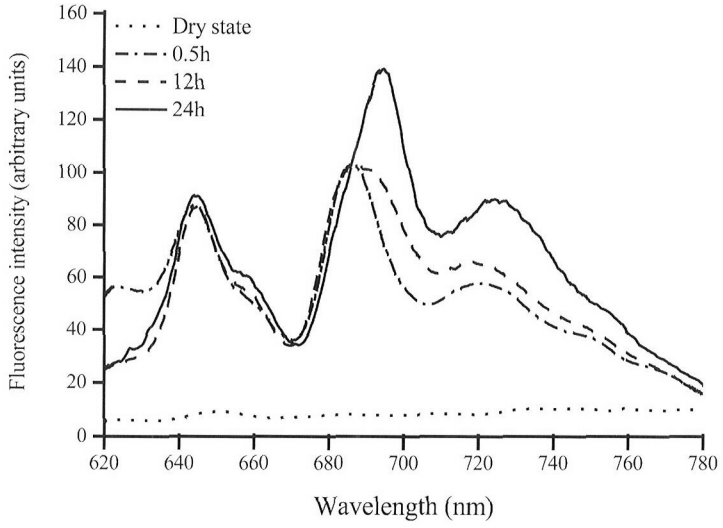

Fig. 3. The 77 K fluorescence emission spectra of *Nostoc flagelliforme* with 580 nm excitation. Dried field samples were rehydrated in BG_{11} medium at 40 μmol m^{-2} s^{-1} and 25°C for 0.5, 12 or 24 h. The 77 K fluorescence of dry samples was used as control.

er peptide (L_{CM}). It recovered rapidly and reached a maximum at 0.5 h. The fluorescence level at 695 nm was attributed to the chlorophyll of PS II and was very low at 0.5 h. Thus, the fluorescence at 685 nm can mainly be attributed to L_{CM} at 0.5 h. The rapid and large increase in fluorescence from phycocyanin (645 nm), allophycocyanin (658 nm), L_{CM} (685 nm) and photosystem I (PS I; 721 nm) at 0.5 h suggests the functional recovery of phycobiliproteins and PS I complexes and active energy transfer from allophycocyanin to L_{CM}.

A fast initial phase of recovery was completed within half an hour, and further change of 77 K fluorescence emission spectra was mainly related to the fluorescence level at 695 nm (Fig. 3), which increased by about 16% and 32% at 12 and 24 h, respectively. The decrease in fluorescence intensities of phycobiliproteins (645–685 nm), resulting from changes in their coupling and assembly (Inoue *et al.* 2000), was not detected from 0.5 to 24 h. The F685 : F695 ratio was 1.41 at 0.5 h and decreased to 0.69 at 24 h (Table 1). The F695 : F721 level increased from 1.23 to 1.54 during rehydration. These would suggest PS II recovery because the fluorescence emission at 695 nm originated from the PS II reaction centre complexes.

Recovery of PS II activity during rehydration

In cyanobacteria, phycobiliprotein fluorescence and PS I fluorescence contribute to the F_0 level, thus decreasing F_v/F_m; this makes F_v/F_m a less reliable indicator of potential PS II activity (Campbell *et al.* 1998). The oxygen-evolving activity of intact cells was examined in the presence of *p*-BQ as an artificial acceptor of electrons (Fig. 4). This would give a more direct indication of PS II activity and its possible change during rehydration. The change in F_v/F_m during rehydration correlated well with the recovery of PS II activity, with a linear correlation coefficient of 0.9895 ($P < 0.05$, *t* test) (Figs 4, 6). The variable fluorescence arises from PS II. Its change also correlated well with the recovery of PS II activity during rehydration, and the linear correlation coefficient was 0.9922 ($P < 0.05$, *t* test) (Figs 4, 6). Thus, we could use variable fluorescence or the F_v/F_m value as an index of PS II activity during rehydration. The PS II activity of dried field samples could reach 280 μmol O_2 h^{-1} g^{-1} dry weight after 24 h rehydration. It took about 4 h for them to recover 50% of activity.

Changes in pigment concentration during rehydration

The values of fluorescence parameters could also vary somewhat with pigment concentration (Ting & Owens 1992), so the pigment concentration of *N. flagelliforme* during rehydration was assayed. Pigment contents of chlorophyll *a*, phycocyanin and allophycocyanin were constant during the photosynthetic recovery (Fig. 5).

Effects of light and inhibitor of protein synthesis on photosynthetic recovery

The photosynthetic recovery of dried field samples upon rehydration is shown in Fig. 6. Only 3.4% of the PS II photochemical efficiency was recovered in darkness. Recovery could reach 10.1% when light was provided in the presence of CMP. Thus, it was shown that the photosynthetic recovery required light and the *de novo* synthesis of protein. The initial

Table 1. Changes in 77 K fluorescence emission ratios of *Nostoc flagelliforme* during rehydration. Data indicate $\bar{x} \pm s$ (n = 3–5). Those with different superscript letters are significantly different ($P < 0.05$, Tukey multiple comparison).

Rewetting time (h)	F645 : F658	F645 : F685	F685 : F695	F695 : F721
0.5	1.67 ± 0.13[a]	0.90 ± 0.32[a]	1.41 ± 0.05[a]	1.23 ± 0.10[a]
12	1.59 ± 0.16[a]	0.86 ± 0.02[a]	1.11 ± 0.28[a]	1.43 ± 0.01[a]
24	1.50 ± 0.08[a]	0.95 ± 0.01[a]	0.69 ± 0.06[b]	1.54 ± 0.04[b]

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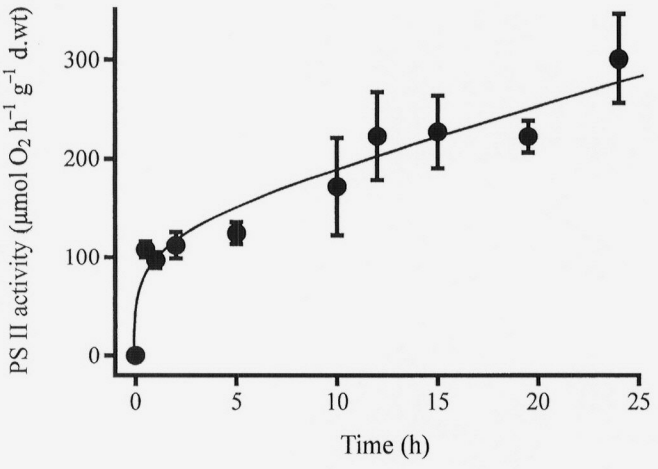

Fig. 4. The recovery of PS II activity in *Nostoc flagelliforme* as measured by the oxygen-evolving activity of intact cells in the presence of *p*-BQ as an artificial acceptor of electrons. Dried field samples were rehydrated in BG_{11} medium at 40 μmol m^{-2} s^{-1} and 25°C. Data indicate $\bar{x} \pm s$ ($n = 5$).

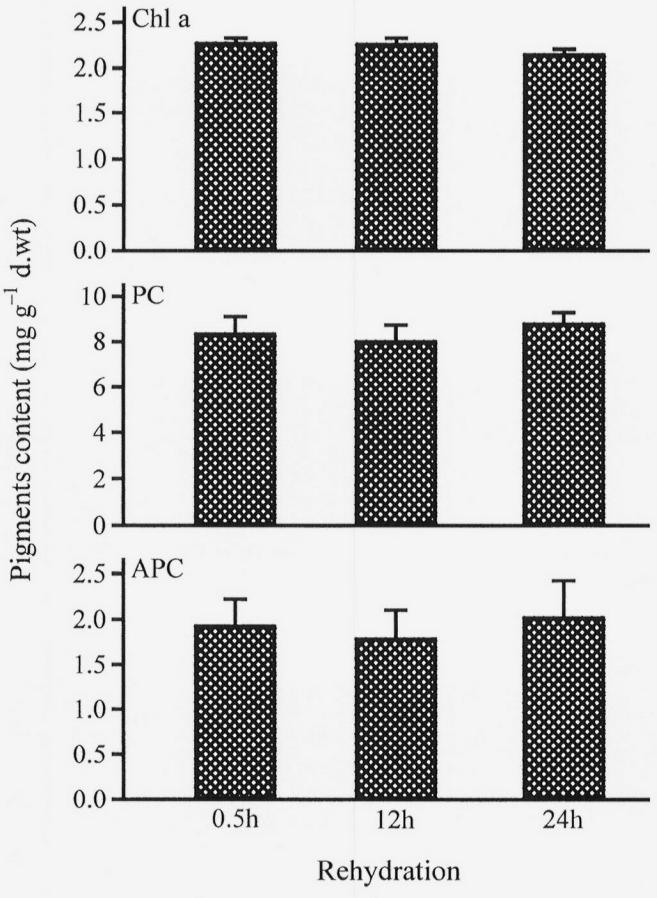

Fig. 5. Changes of pigment content in *Nostoc flagelliforme* during rehydration. Dried field samples were rehydrated in BG_{11} medium at 40 μmol m^{-2} s^{-1} and 25°C for 0.5, 12 or 24 h. Chl a, chlorophyll *a;* PC, phycocyanin; APC, allophycocyanin. Data indicate $\bar{x} \pm s$ (n = 6–15).

fluorescence for samples rehydrated in light was significantly lower and the variable fluorescence was significantly higher compared with those rehydrated in darkness or with the addition of CMP in light ($P < 0.05$, *t* test).

Dried field samples fully recovered their photosynthetic activity at 40 μmol m^{-2} s^{-1} and 25°C in BG_{11} medium after being wind-dried at room temperature for 24 h. These samples were used to investigate the effects of light and inhibition of protein synthesis on photosynthetic recovery (Fig. 7). The PS II photochemical efficiency for those samples rehydrated in darkness or in light with the addition of CMP could reach 18.9% and 39.5% of controls, respectively. These values were significantly higher than those for dried field samples ($P < 0.05$, *t* test). The recovery of PS II photochemical efficiency was less different between those with and without the addition of CMP during the first hour ($P > 0.05$, *t* test). The PS II photochemical efficiency for samples with the addition of CMP tended to decrease with time in darkness, so it was significantly lower than those without the addition of CMP in the next 3 h ($P < 0.05$, *t* test). The initial fluorescence for samples with the addition of CMP decreased rapidly when the light was on. It began to increase 2 h later and was close to the initial values 15 h later. The variable fluorescence for samples with the addition of CMP decreased in darkness. It increased rapidly when light was on and decreased 2 h later. The time required for the appearance of maximal fluorescence was about 880 μs with the addition of CMP. When the light was on, the time to maximal fluorescence increased to about 0.57 s for samples without the addition of CMP. The area over the induction curve for samples with the addition of CMP was less than 21.9% of those samples fully recovered in light.

DISCUSSION

The initial fluorescence reached a maximum at a water loss of about 35% and then decreased with further water loss (Fig. 1). However, the values of F_0 are high during the initial phase of rehydration (Figs 2, 6, 7). Its rise might occur in the first several minutes of rehydration. This has been demonstrated recently in *N. commune* (Satoh *et al.* 2002). On rehydration, the initial fluorescence decreased sharply and its decrease was accompanied by an increase in the F_v value (Figs 6, 7). The F_0 level of fluorescence is thought to represent emission by excited antenna chlorophyll molecules occurring before the excitons have migrated to the reaction centres (Mathis & Paillotin 1981). Several factors could contribute to changes in F_0. The coupling or assembly of phycobilisomes (Inoue *et al.* 2000), a state transition from the high fluorescent state (state I) to the low fluorescent state (state II) (Krause & Weis 1991) and an increase in energy dissipation such as the heat dissipation dependent on the xanthophyll cycle (Demmig *et al.* 1987) could all result in the decrease of initial fluorescence. On the other hand, both the build-up of a stable Q_A species (Yamane *et al.* 2000) and irreversible damage or reversible inactivation of PS II reaction centres (Schreiber & Armond 1978; Yamane *et al.* 1997; Hong *et al.* 1999; Inoue *et al.* 2000) could lead to a rise in F_0. Thus, we attempted to determine the cause of the F_0 decrease during rehydration.

First, we could rule out the possibility of changes in the coupling and assembly of phycobilisomes resulting in F_0 decrease. The principal light-harvesting complexes of cyanobacteria are phycobilisomes peripheral to the thylakoid membranes (Bhaya *et al.* 2000). The contribution of fluorescence

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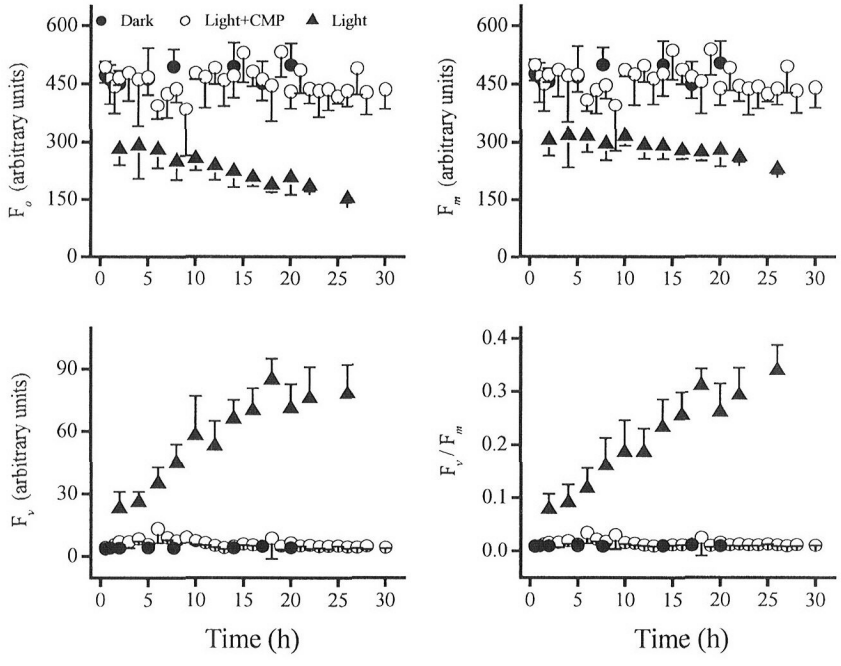

Fig. 6. Recovery of fluorescence parameters for dried field samples of *Nostoc flagelliforme* when rehydrated in dark, light (40 μmol m^{-2} s^{-1}) and light (40 μmol m^{-2} s^{-1}) plus protein-synthesis inhibitor (2.5 mM CMP). The CMP was added 0.5 h before the light was turned on at 0000 h. All samples were rehydrated in BG_{11} medium at 25°C. Data indicate $\bar{x} \pm s$ ($n = 6$). Other details as in Fig. 1.

from phycobilisomes to F_0 is usually quite large in cyanobacteria (Satoh *et al.* 2002). Its contribution to F_0 fluorescence could be a low-yield fluorescence emission from coupled phycobilisomes or a high-yield emission from a small population of uncoupled phycobilisomes or free phycobiliproteins (Campbell *et al.* 1998). One could easily imagine that desiccation affected the phycobilisome assembly and their coupling to the reaction centres. The 650 nm light supplied by the PEA is not selective for chlorophyll *a*, and there will also be very strong phycobilisome excitation at this wavelength. Therefore,

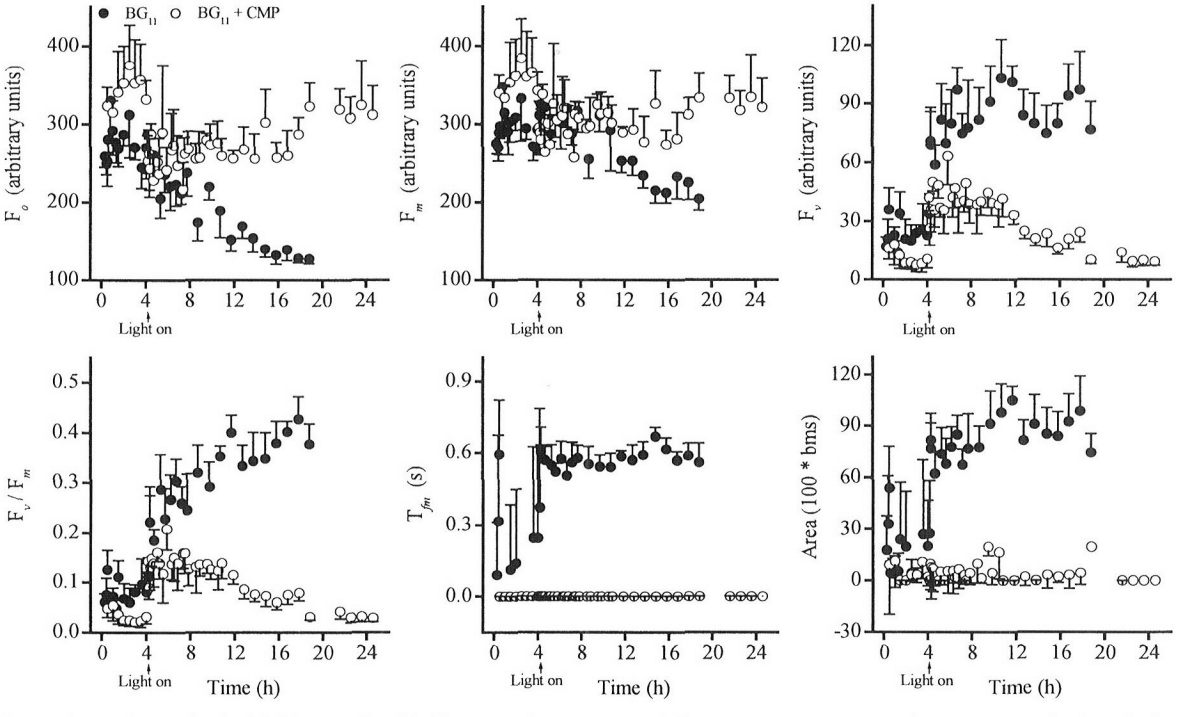

Fig. 7. Effects of protein-synthesis inhibitor (2.5 mM CMP) on the recovery of fluorescence parameters for secondly rehydrated *Nostoc flagelliforme*. Dried field samples were rehydrated in BG_{11} medium at 40 μmol m^{-2} s^{-1} and 25°C, wind-dried at room temperature (23–28°C) and then rehydrated again in BG_{11} medium at 25°C about 20–30 h later. Irradiance was provided with 40 μmol m^{-2} s^{-1} at 0400 h. Data indicate $\bar{x} \pm s$ ($n = 6$). Other details as in Fig. 1.

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it is possible that the decrease of F_0 during rehydration is due to the coupling or assembly of phycobilisomes. However, a fast initial phase of recovery was completed within half an hour, and further change of 77 K fluorescence emission spectra was mainly related to the fluorescence level at 695 nm (Fig. 3). The decrease in fluorescence intensities of phycobiliproteins (645–685 nm) resulting from changes in their coupling and assembly was not ever detected from 0.5 to 24 h (Fig. 3). Thus, the decrease in F_0 is not due to changes in the coupling and assembly of phycobilisomes.

Second, the decrease of F_0 during rehydration could not result from a state transition. Nonphotochemical quenching in cyanobacteria largely reflects changes in the PS II fluorescence yield as a result of the state transition mechanism, which regulates the distribution of excitation energy between PS II and PS I (Mullineaux & Allen 1986; Campbell *et al.* 1998). The F695:F721 value increased significantly from 1.23 to 1.54 during rehydration ($P < 0.05$, Tukey multiple comparison) (Table 1). This indicated that *N. flagelliforme* shifted towards state I if the state transition occurred in this process. However, state I was associated with high PS II fluorescence yield and low nonphotochemical quenching (Campbell *et al.* 1998). Thus, the state transition could not be used to explain the F_0 decrease during rehydration.

Furthermore, the increase in energy dissipation is not the cause of the F_0 decrease during rehydration. The xanthophyll cycle is implicated in regulating nonphotochemical dissipation of excess light energy. Cyanobacteria seem to lack this cycle but have a sustained content of the carotenoid zeaxanthin, and this might be associated with fluorescence quenching similar to nonphotochemical quenching (Ibelings *et al.* 1994). However, the half-time of dark relaxation for the xanthophyll cycle is less than 1 min (Quick & Stitt 1989; Krause & Weis 1991). Meanwhile, the irradiance used during the rehydration was just 40 μmol m^{-2} s^{-1}, and all samples had been dark-adapted for 15 min before measurements. Thus, the increase in energy dissipation is not important during rehydration and would not lead to a decrease in F_0. Based on all the above reasons, we could conclude that the decrease in F_0 appeared to be mainly related to the activation or repair of PS II.

Upon rehydration, dried populations of *N. flagelliforme* required *de novo* protein synthesis to recover their photosynthetic activity fully (Figs 6, 7). This mechanism is different from that which occurs in some other species, such as desiccation-tolerant bryophytes, in which photosynthetic recovery can be remarkably rapid and appears to leave little time for repair processes requiring synthesis of proteins or other cell components on any substantial scale (Proctor & Smirnoff 2000). The present study showed that PS II photochemical efficiency of dried field samples of *N. flagelliforme* recovered by only 3.4% in darkness (Fig. 6). Such a degree of recovery is so small that we could not detect it in an earlier study (Gao *et al.* 1998). For samples stored dry for 2 days, PS II photochemical efficiency recovered 18.9% when rehydrated in darkness (Fig. 7). However, Scherer *et al.* (1984) found that oxygen evolution of *N. flagelliforme* recovered by 46.5–54.2% when samples stored dry for 2 years were recovered from desiccation in the dark. The discrepancy between our data and those of Scherer *et al.* (1984) might come from the differences in methods and materials employed.

The photosynthetic recovery of *N. flagelliforme* during rehydration exhibits similarity to that of photoinhibited *Synechocystis* Sauvageau PCC 6714. The photosynthetic recovery of *Synechocystis* PCC 6714 from photoinhibition has also been shown to require illumination and *de novo* protein synthesis (Constant *et al.* 2000). The photosynthetic recovery of *N. flagelliforme* could be divided into three constituent parts: recovery of photosynthetic function that can occur in the absence of light, recovery that is light dependent but does not require protein resynthesis, and recovery dependent on both light and protein resynthesis. These three parts could be related to functional PS II centres, reversibly inactivated centres and nonfunctional centres in the cycle of inactivation, damage and repair of PS II in terms of their requirements for light and protein synthesis (Andersson & Aro 2001). Recently, we have reported data suggesting that reactive oxygen species might contribute to desiccation-induced damages to the photosynthetic apparatus of *N. flagelliforme* (Qiu *et al.* 2003). Based on the metabolism of reactive oxygen species, we could also associate desiccation-induced damages and subsequent photosynthetic recovery upon rehydration in *N. flagelliforme* with the cycle of inactivation, damage and repair of PS II.

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