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Effects of thallus temperature and hydration on photosynthetic parameters of *Cetraria islandica* from contrasting habitats

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Abstract

Two methods of induced *in vivo* chlorophyll (Chl) fluorescence were used to investigate the effects of varying thallus temperature and hydration on the performance of photosynthetic apparatus of a foliar lichen *Cetraria islandica*: slow Chl fluorescence induction kinetic with the analysis of quenching mechanisms, and rapid irradiance response curves of photosynthesis derived from quantum yield of photochemical reactions of photosystem 2 (Φ_2) recorded at increasing irradiances. We compared responses of photosynthetic apparatus in populations of *C. islandica* growing in lower altitude (LAP: 1 350 m a.s.l.) and in higher altitude (HAP: 2 000 m a.s.l.). At each altitude, the samples were collected both in fully irradiated sites (HI) and in shade (LI). Temperature optimum of photosynthetic processes was the same for LAP and HAP thalli of LI populations (18 °C), while it was significantly lower for HI HAP (14 °C). Gradual dehydration of fully hydrated thalli led to initial increase (up to 20 % of water saturation deficit, WSD) in F_V/F_M and Φ_2, no change at 20-50 % WSD, and a dramatic decrease of the parameters within 50-80 % of WSD. LI HAP of *C. islandica* was the best adapted population to low temperature having higher rates of photochemical processes of photosynthesis than HI HAP within temperature range of –5 to +5 °C. The differences between populations were apparent also in Chl content and thallus morphology.

Additional key words: chlorophyll fluorescence; *Cladonia rangiferina*; high and low altitude; high and low temperature; iceland moss; irradiance response curves; lichen; *Pseudevernia furfuracea*; shade acclimation.

Introduction

Changes in photosynthetic processes are frequently used as an extremely sensitive indicator of plant responses to environment. Traditionally used gasometric approach to the measurements of photosynthetic rates of higher plants is rather complicated when used in lichens due to CO_2 output from respiratory processes of fungal layers of lichen thalli. Nevertheless, some basic studies on photosynthetic performance in lichens have been done over last decades, *e.g.*, daily courses of photosynthesis (Lange 1988, Lange *et al.* 1998), irradiance-response curves (Sundberg *et al.* 1997). New development of measuring techniques promoted recently an application of fluorometric methods in photosynthetic studies of lichens. Using the methods of induced chlorophyll (Chl) fluorescence, lichen photosynthesis can be evaluated in terms of potential photochemical proceses (F_V/F_M), actual quantum yield of photosystem 2 (PS2) activity, and analysis of quenching mechanism of Chl fluorescence.

Among lichens, *Cetraria islandica* is a common experimental object. Thus, photosynthetic performance of the species is known to more details than in other species. The knowledge is, however, far from being satisfactory. Well documented are, *e.g.*, both *in situ* and laboratory-made daily courses of photosynthesis recorded continuously over several days in *C. isladnica* from polar regions (Lange *et al.* 1998). Also irradiance-response curves of photosynthesis have been measured in the species both under field and laboratory conditions (Reiter and Türk 2000). Only a very limited attention, however, has yet been devoted to inter-specific differences in photosynthesis between *Cetraria* populations growing at contrasting environmental conditions, air temperature in particular. To our best knowledge, the only attempt was made by Schipperges *et al.* (1995) who compared photosynthesis of *C. nivalis* along longitudal gradient. Lichen photosynthesis under low temperature has been studied almost exclusively in Arctic and Antarctic species (*e.g.*, Hovenden *et al.* 1994, Schroeter *et al.* 1994) while in European

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mountain species similar studies are lacking. Lichens of the genus *Cetraria* are an important component of tundra-like vegetation of higher mountains in temperate zone, which may be particularly vulnerable to expected climate changes. Better understanding of their physiological responses to environmental factors is thus highly desirable. The aim of our study was to evaluate responses of *C. islandica* thalli to dehydration and decreasing air temperature, with a special respect to subzero temperatures.

Materials and methods

Lichen collection and handling: Samples of *C. islandica* were collected in the ground of stony slopes of the Tatra Mountains in September 2000. The samples were collected at 1 350 m (lower altitude populations, LAP) or at 2 000 m (higher altitude populations, HAP). At each elevation, samples were taken both from the populations fully exposed to global radiation (HI) and from shaded sites (LI). Long-term annual mean of air temperature at the lower elevation was +2.6 °C, while in higher elevation only –0.8 °C (derived from climatological data presented by Kňazovický 1970). After collection, the field-dried lichen samples were transferred to a laboratory in Brno and stored in dark at 5 °C. Before measurements, the thalli were placed between two sheets of chromatographic paper supplied with water for 2 h in dark at room temperature (22 °C) to reach full hydration (sufficient duration – tested before). The over-saturation by water was avoided by placing the thalli between two sheets of dry paper and gentle pressing to remove surplus water immediately before measurements. Then, a set of Chl fluorescence measurements (specified below) was done on the lichen samples exposed to (*1*) changing irradiance (from 40 to 400 μmol m^{-2} s^{-1}, for details see the Chl fluorescence parameters, irradiance response curves), (*2*) thallus temperature changing within the ranges of –10 to 45 °C or –5 to 22 °C), and (*3*) slow dehydration (calculated as water saturation deficit – WSD) using the equation:

$$WSD = 100\,[1 - (FM_a - DM_t)/(FM_{max} - DM_t)] \quad (1),$$

where FM_{max} is water-saturated fresh mass of thallus, FM_a is actual fresh mass of thallus, DM_t is dry mass of thallus. Constant thallus temperature was controlled either in a small-volume (25 000 cm^3) cooled box (*URAS*, Germany, temperature range of –10 to 22 °C) or in a heated thermostat (*KBC G65*, Poland, temperature range of 20 to 45 °C). Thallus surface temperature was continuously monitored by a thermistor touching the upper surface of the thallus, and recorded by a data logger *HOBO H8* (*Onset Computer Corporation*, USA). At each thallus temperature (for single sample typically in the following order: 22, 17, 8, 6, 5, 0, –5, –10, +35, +45 °C), at least 12 h acclimation was provided to assure equilibrium of physiological processes. Full thalli hydration during acclimation and consequent measurements was assured by wet paper placed in contact with lower surface of a thallus. In the study of lichen photosynthetic response to hydration, lichen thalli were gradually dehydrated from water-saturated state at room temperature (in terms of hours) until they reached required WSD (checked gravimetrically). Subsequently, a darkening clip was attached to the thalli and the rest of thalli were covered by aluminium foil not to loose water. After dark adaptation, the thalli were measured fluorometrically as described below (see Chl fluorescence parameters). High dehydration of thalli (WSD above 60 %) was reached by the exposition of thalli to dry air at 30 °C until the required WSD was reached. In the experiment, two additional species (*Cladonia rangiferina*, *Pseudevernia furfuracea*) collected at the same sites, were used.

Structural characteristics of thalli and content of pigments: Interspecific differences in thallus anatomy and morphology were evaluated using several biometrical characteristics. Thickness of the thallus was measured in cross-section made in the half height of a single thallus. Specific thallus area (STA) was evaluated as one-side area of a flat thallus *per* dry mass unit of the thallus. The area was measured by a table scanner (*Hewlett Packard ScanJet II P*) and an area determining software, dry mass of thallus was evaluated using precise laboratory scale (*Ohaus TS 400 D*, USA). For anatomical study, 7 μm thick cross-sections were prepared using a freezing microtome (*Cryo-cut*, *AOC*, USA) and scanned by an optical microscope (*Olympus*, *BX60F-3*, Japan) equipped with an *Olympus C-3030* camera. The digitised images were evaluated by *LUCIA* software v. *6.0* (*Laboratory Imaging*, Czech Republic) for image analysis. Contents of Chl *a* and *b* and total carotenoids were determined spectrophotometrically (*Shimadzu UV-1601*, Japan) according to Lichtenthaler and Wellburn (1983) in 100 % acetone extract. The pigment content was expressed on thallus dry mass basis.

Chl fluorescence parameters (F_V/F_M, Φ_2, q_P, q_N – for terminology see van Kooten and Snell 1990, Roháček and Barták 1999) were determined from analysis of slow kinetics of Chl fluorescence supplemented with saturation pulses (recorded by a *PAM-2000* fluorometer, *Walz*, Germany). On dark-adapted (10 min, sufficient duration tested before by checking F_0) thalli of lichens, a weak irradiance of 0.5 μmol m^{-2} s^{-1} was applied in order to determine basic Chl fluorescence (F_0) accompanied with a saturation pulse (5 000 μmol m^{-2} s^{-1}) to calculate maximum capacity of PS2 (F_V/F_M). Then, low actinic irradiance (20 μmol m^{-2} s^{-1}) was applied for 5 min until steady-state Chl fluorescence (F_S) was reached, followed by

a saturation pulse. This enabled to determine the quantum yield of photochemical reactions of PS2 (Φ_2), the photochemical quenching (q_P), and the non-photochemical quenching (q_N).

$$\Phi_2 = (F'_M - F_S)/F'_M \quad (2),$$
$$q_P = (F'_M - F_S)/(F'_M - F_0) \quad (3),$$
$$q_N = (F_M - F'_M)/(F_M - F_0) \quad (4),$$

where F_M is maximum Chl fluorescence on dark-adapted thalli, and F'_M is maximum Chl fluorescence on light-adapted thalli (under actinic irradiance).

Rapid irradiance response curves of photosynthesis (RIC) were recorded using a *PAM-2000* (*H. Walz*, Germany) fluorometer and a method originally developed by White and Critchley (1999) in the modification of Barták (unpublished). The principal of the method is the exposure of sample to a stepwise ascending irradiance with saturation pulse given at each irradiance. We exposed dark-adapted thalli (10 min) to 8 ascending irradiances provided by a halogen lamp. At each irradiance (40, 51, 66, 100, 135, 195, 270, and 400 μmol m^{-2} s^{-1}), the saturation pulse (5 000 μmol m^{-2} s^{-1}) was applied in order to determine Φ_2 and consequently the electron transport rate (ETR):

$$ETR = \Phi_2 \times 0.5 \times a \times PPFD \quad (5),$$

where 0.5 reflects the fact that two photons are required per one electron transported, a is absorptance of thalli (estimated as 0.8), and PPFD is photosynthetic photon flux density of photosynthetically active radiation incident on the top surface of thalli. Apparent rate of gross CO_2 assimilation (P_G) was calculated using the equation:

$$P_G = ETR\ \Phi_{CO2} \quad (6),$$

where Φ_{CO2} is the quantum yield of CO_2 fixation (assumed as 0.125; Krall and Edwards 1992) considering that minimum 8 quanta is required per one molecule of CO_2 fixed. Rapid irradiance response curves were constructed by plotting P_G against PPFD used as actinic irradiance. Maximum P_G (P_{Gmax}) was calculated as an asymptotic value of exponential fit (Potvin *et al.* 1990).

Results

Thalli structural characteristics: The studied populations of *C. islandica* differed in majority of structural characteristics. Chl and carotenoid contents are summarised in Table 1. HAP compared to LAP showed higher (by about 25 %) Chl *a* and Chl *b* contents expressed on dry mass unit of thallus. The content of carotenoids in HAP increased only in HI thalli while it remained almost unchanged in LI thalli. Generally, HI compared to LI thalli showed significant decrease of about 20 % in Chl *a* and Chl *b* contents. Ratio of total carotenoids to chlorophylls was significantly higher in HI than LI thalli and in LAP than HAP populations. The latter difference was, however, more apparent in HI thalli.

Both the thickness of photobiont layer and its ratio to thallus thickness were affected by habitat conditions, irradiance in particular, since there were significant differences in the two characteristics between LI and HI thalli. Photobiont layer thickness showed either no difference (between the populations from higher altitude, Table 1) or increase (by the factor of 1.6) in populations from lower altitude with enhanced availability of radiant energy (HI LAP *vs.* LI HAP). The ratio of photobiont layer thickness to thallus thickness was higher in the populations with availability of radiant energy enhanced by the factor of 1.2 (populations from lower altitude) or 1.5 (populations from higher altitude).

Response of photosynthetic parameters to temperature: Gradual decrease of air temperature from +35 to −10 °C induced changes in Chl fluorescence parameters

Table 1. Basic description and biometrical parameters (STA = specific thallus area, Chl = chlorophyll, Car = carotenoids, TT = thallus thickness, TPL = thickness of photobiont layer) of sun (HI) or shade (LI), lower (LAP) or higher (HAP) altitude populations of *Cetraria islandica*.

	LI-LAP	LI-HAP	HI-LAP	HI-HAP
STA [cm^2 kg^{-1}]	110.16±13.80	112.69±18.50	99.81±9.90	118.79±6.10
Chl *a* [g kg^{-1}(DM)]	0.65±0.20	0.82±0.13	0.46±0.08	0.67±0.15
Chl *b* [g kg^{-1}(DM)]	0.18±0.06	0.20±0.03	0.08±0.01	0.17±0.04
Chl (*a*+*b*) [g kg^{-1}(DM)]	0.83±0.26	1.02±0.16	0.54±0.09	0.84±0.17
Car [g kg^{-1}(DM)]	0.16±0.06	0.18±0.03	0.16±0.04	0.20±0.03
Chl *a*/*b*	3.59±0.17	4.02±0.12	5.91±0.49	4.07±0.24
Car/Chl	0.19±0.01	0.17±0.02	0.29±0.03	0.24±0.02
TT [μm]	112.73±10.40	195.77±14.7	150.16±12.70	128.31±8.10
TPL [μm]	19.90±4.80	27.38±7.90	31.46±11.60	27.14±7.90
TPL/TT	0.18±0.01	0.14±0.01	0.21±0.01	0.21±0.02

(Fig. 1). Φ_2 showed typical temperature response curve with two minima at extremely low (–10 °C) and high temperature (about 45 °C), and temperature optimum characterised by maximal Φ_2. In LI population, temperature optimum of Φ_2 was the same for LAP and HAP thalli (22 °C), contrasting to HI population that showed distinguished optima for LAP (18 °C) and HAP (12 °C). F_V/F_M was more or less constant over a wide range of temperature (0 to 30 °C) showing only slight decrease at subzero temperatures. At high temperature, however, F_V/F_M showed dramatic decrease starting at temperature above 30 °C. Temperature response curve of photochemical quenching of Chl fluorescence (q_P) was similarly shaped as the Φ_2 curve. It showed low- and high-temperature minima, as well as q_P temperature optimum within a broad temperature interval from 15 to 25 °C. Non-photochemical quenching of Chl fluorescence (q_N) remained low within temperature interval of –5 to 10 °C. At the temperatures below –5 and/or above 10 °C, q_N increased which was more apparent at high temperature.

Rapid irradiance response curves (RIC): The shape of RIC differed when measured under 22, 10, and 5 °C (Fig. 2). Maximum gross photosynthesis (P_{Gmax}) was

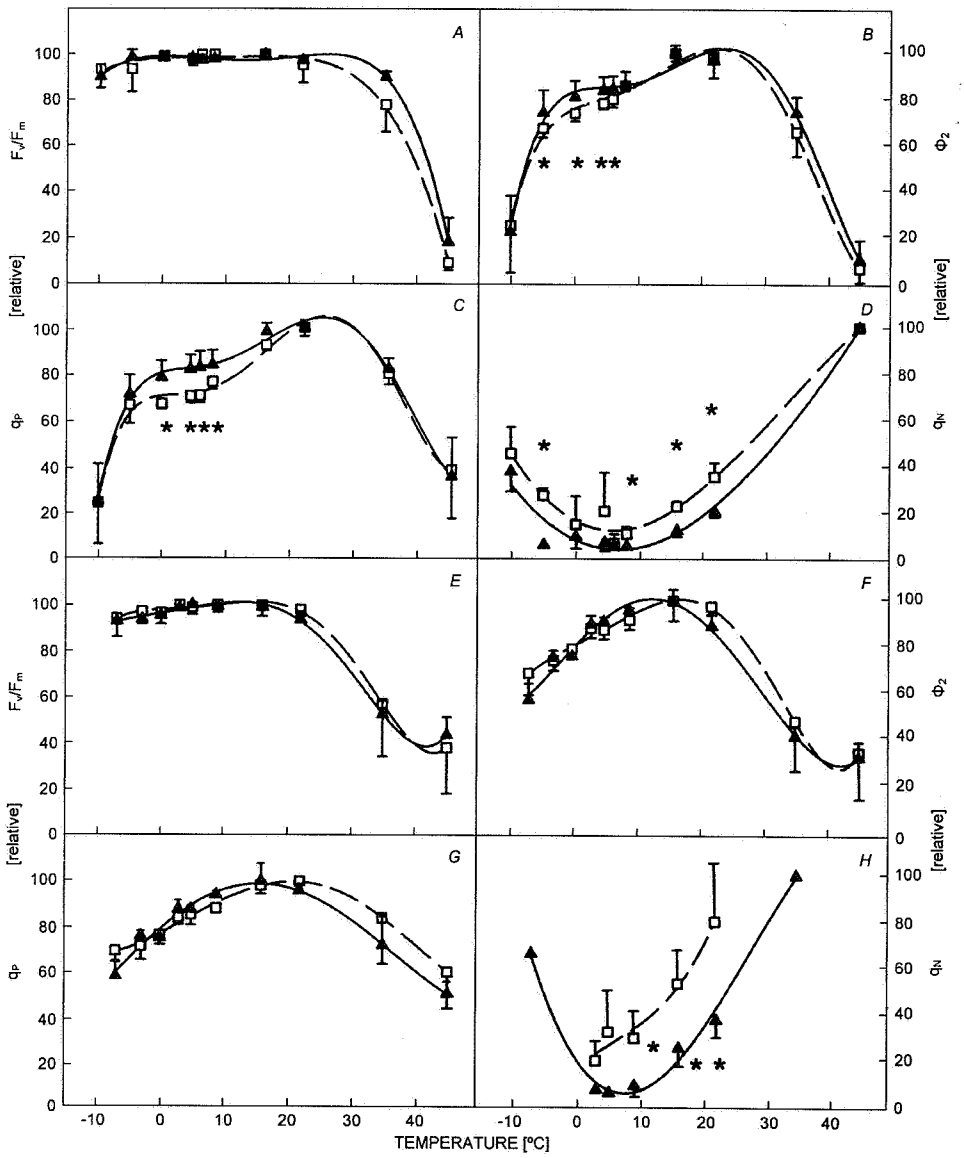

Fig. 1. Temperature dependence of basic chlorophyll fluorescence parameters (variable to maximum Chl fluorescence ratio, F_V/F_M; quantum yield of photochemical reactions of PS2, Φ_2; photochemical quenching, q_P; non-photochemical quenching, q_N) recorded on fully hydrated thalli of *Cetraria islandica* collected from shaded sites (LI population – *A* to *D*) and sun exposed sites (HI population – *E* to *H*). Thalli of lower altitude population (LAP) are indicated by □, thalli of higher altitude population (HAP) by ▲. Means of at least 5 replicates. Statistically significant differences ($p<0.05$, Student *t*-test) between populations are indicated by asteriscs*.

reached at PPFD of about 300 μmol m^{-2} s^{-1} at all temperatures tested. The values of P_{Gmax} varied also in dependence on origin of *C. islandica* populations. Maximum values of P_{Gmax} in LAP were found at 22 °C indicating

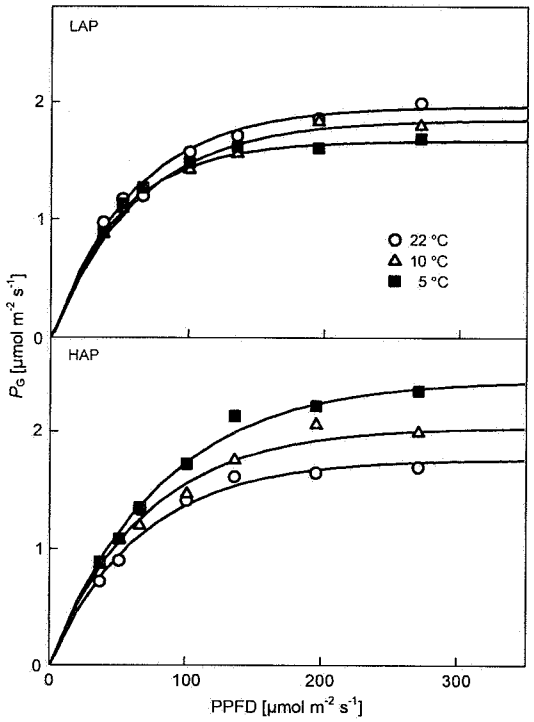

Fig. 2. Rapid irradiance response curves of gross photosynthetic rate (P_G) in optimally hydrated thalli of lower (LAP) and higher altitude (HAP) populations of *C. islandica* as measured at thalli temperature of 22 °C (○), 10 °C (Δ), and 5 °C (■).

that temperature optimum of photosynthesis in LAP lies close to that temperature. In contrast, HAP exhibited maximum P_{Gmax} when measured under 5 °C. Absolute maxima of P_{Gmax} values, however, were almost identical for LAP and HAP thalli: 1.9-2.2 μmol(CO_2) m^{-2} s^{-1}.

Response to dehydration: LAP of *C. islandica* exhibited maximum values of F_V/F_M and Φ_2 at low WSD (Fig. 3). At WSD above 50 %, both Chl fluorescence parameters declined. Dramatic decrease of F_V/F_M and Φ_2 was found within the range of 60-70 % of WSD. Zero values of the two parameters were found at about 70 % of WSD. HAP showed different response to thalli dehydration. In the initial phase, from fully to partially hydrated state (WSD 0-40 %), they exhibited either plateau or increase of F_V/F_M and Φ_2. Maximum values of Φ_2 were found within WSD range of 40-50 %. With following desiccation (WSD 50-65 %), *C. islandica* showed decline in both parameters. Similarly to LAP of *C. islandica*, gradually desiccated thalli of *Cladonia rangiferina* showed comparable shape of the F_V/F_M and Φ_2 course. Thalli of *Pseudeveria furfuracea* showed, on the contrary, courses comparable to those of HAP of *C. islandica*.

When fully hydrated, LAP and HAP thalli had P_{Gmax} (derived from RICs, see Fig. 4) of 1.0-1.3 or 0.9-1.3 μmol(CO_2) m^{-2} s^{-1}, respectively. With moderate dehydration within the WSD range of 10-50 %, the thalli showed either no change or slight increase in P_{Gmax}. Severe dehydration of *C. islandica* thalli to WSD of about 70 % led to a decrease of P_{Gmax}: LAP 0.3-0.5 μmol(CO_2) m^{-2} s^{-1}, HAP 0.4-0.9 μmol(CO_2) m^{-2} s^{-1}. Generally, P_{Gmax} was slightly higher in HI than LI thalli (by the factor of about 1.2).

Discussion

Temperature dependence of photosynthetic parameters: Differences among the four populations of *C. islandica* were most pronounced at low temperatures. There was an inter-population variation within the temperature range from –5 to +10 °C, where the differences in Φ_2 were rather small in absolute terms but represented 10-15 % higher effectivity of photochemical reactions of PS2 in HAP compared to LAP. Thus, under low temperatures HAP may utilise about 10-15 % more of incident radiation energy than LAP. This finding is in reasonable agreement with the findings of Schipperges *et al.* (1995) who reported a gradient of cold adaptation of photosynthesis in *C. nivalis* towards more northern populations.

At low temperature, limitation of photosynthesis in *C. islandica* was caused most probably by inhibition of biochemical processes of photosynthesis in chloroplasts of symbiotic alga (*Trebouxia* sp.). Photochemical processes are thought to be less limited at low temperature. In lower plants, F_V/F_M is usually almost temperature independent at above-zero temperatures, but it declines progressively at subzero temperatures (0 to –10 °C, *e.g.*, Deltoro *et al.* 1999). We found that F_V/F_M in *C. islandica* was almost unaffected by subzero temperatures suggesting a high degree of low temperature adaptation. At the temperature range of 0 to –5 °C, Φ_2 reached about 70 % of its maximum. Also a rapid decline of F_V/F_M at the temperatures above 30 °C supports the idea of low-temperature adaptation of the photosynthetic apparatus of the *C. islandica*.

The absolute minimum of temperature at which Φ_2 was above zero in our experiments (LI thalli, –12 °C) corresponds to the values of positive net photosynthesis reported in some lichen species but it is far from –20 °C determined by Kappen *et al.* (1966b). The temperature at which our samples had close-to-zero but still positive Φ_2 was lower than –12 °C, but this can not be extrapolated satisfactorily. Our values, however, confirm that *C. islandica* is photosynthetically active at least at temperatures around –10 °C. The extent of freezing temperature with positive photosynthesis is, however, dependent

on the degree of low-temperature adaptation of the species (Kappen *et al.* 1996a). The adaptation of mountain lichens to low temperature is connected with the presence of large quantities of cryoprotective compounds, *e.g.*, mannitol and ribitol (Ahmadjian 1993, Fontaniella *et al.* 2000) and other mechanisms.

Interpretation of the low-temperature response of photosynthetic parameters and its extrapolation to habitat conditions is disputable. The reason is that full thalli hydration is rather rare at temperature below 0 °C in the

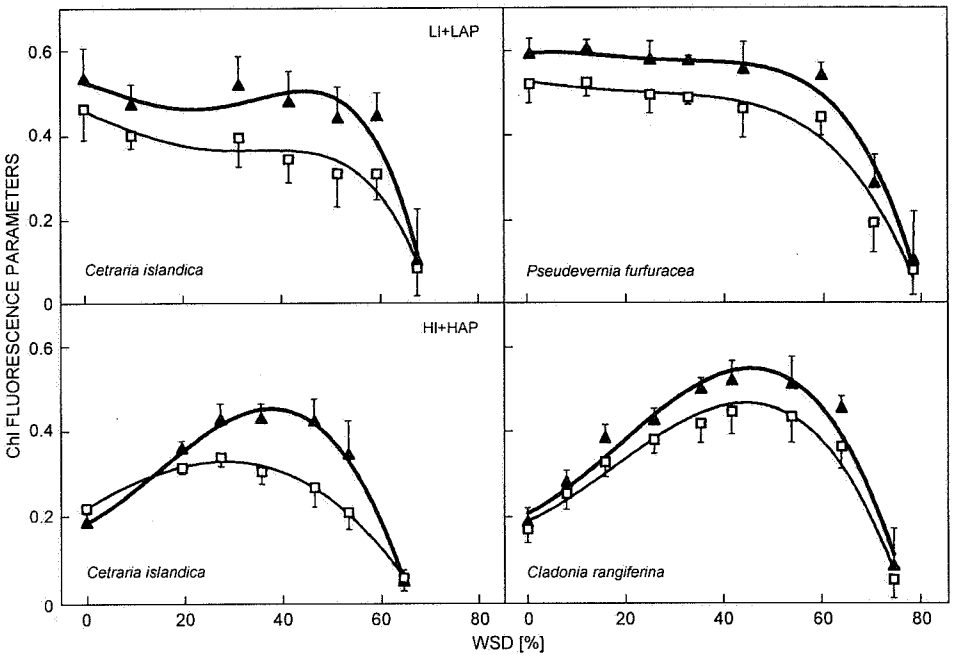

Fig. 3. Dependence of basic Chl fluorescence parameters of thalli of *Cetraria islandica* (*left panels*) and *Pseudoevernia furfuracea* and *Cladonia rangiferina* (*right panels*) on hydration (water saturation deficit, WSD). F_V/F_M is indicated by ▲, quantum yield of PS2 (Φ_2) by □. Means of 5 measurements ± SD. Measurements were made at thallus temperature of 22 °C and under constant CO_2 concentration [400 μmol(CO_2) mol^{-1}].

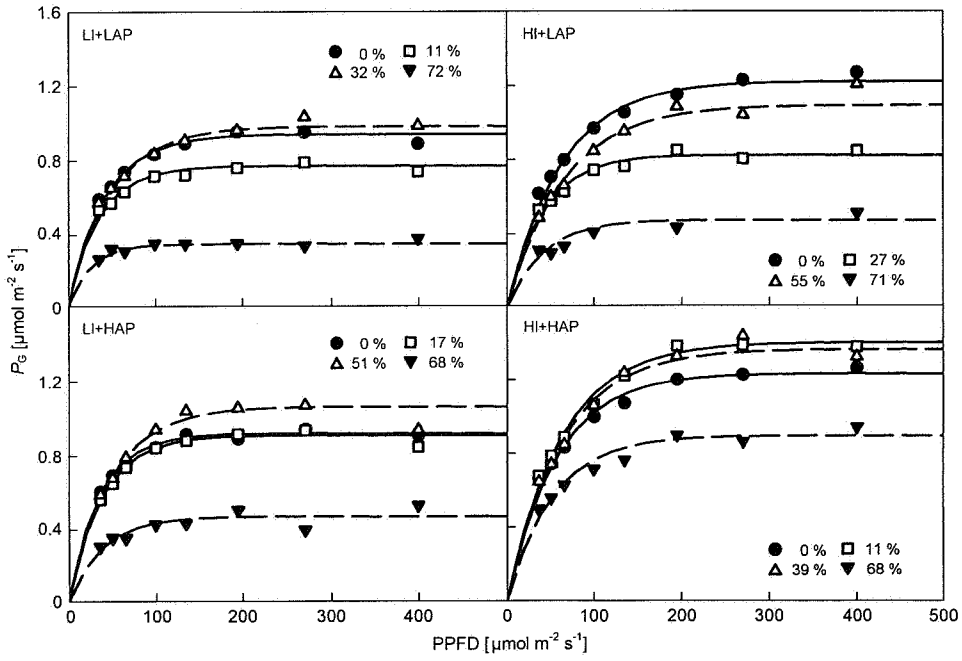

Fig. 4. Rapid irradiance response curves of gross photosynthetic rate (P_G) in thalli of lower (LAP) or higher (HAP) altitude populations of *Cetraria islandica* grown in sun (HI) or shade (LI) as measured at different water saturation deficit (WSD, 0 to 71 %) of thallus. Means of at least 5 measurements.

field. Therefore, decrease of *in situ* photosynthesis at below 0 °C might be caused by interactive effect of low temperature and low water content in thallus. Relative share of these two factors on decrease of lichen photosynthesis is hardly separable in field conditions.

At high temperature, no significant effect of differences in mean temperature in habitats (at different altitudes) on F_V/F_M of lichen samples was apparent. However, Meyer and Santarius (1998) reported effect of short-term (in terms of hours) exposure of mosses to high temperature leading to induced resistance (higher F_V/F_M) to high temperatures. The effect might be explained by an increased thermal stability induced by osmotic stress ongoing with dehydration at high temperatures above 45 °C (Meyer and Santarius 1998). In contrast, in our experiments no decrease of water potential took place due to full hydration of thalli. Moreover, under full hydration, the temperature at which $F_V/F_M = 0$ did not exceed 45 °C.

Responses of P_G to irradiance at different temperatures derived from Φ_2 data might be compared to irradiance response curves of P_N measured gasometrically by some other authors on several species of *Cetraria*. The comparison, however, is complicated due to the respiration of both photo- and mycobiont that is implicitly included in the P_N measured gasometrically, but not in the P_G derived from fluorometric data. Similarly to *C. nivalis* (Schipperges *et al.* 1995), we found saturation irradiance at about 300 μmol m^{-2} s^{-1} in *C. islandica*. In contrast to other authors (Schipperges 1992, Kappen *et al.* 1995) who studied *C. nivalis* and *C. delisei*, we found no significant decrease in saturation irradiance in the thalli from the higher (and thus colder) site. This discrepancy might be attributed to probably lower accuracy of fluorometric method in P_G estimation and high variability of fluorometric values. We found a strong low temperature adaptation of photosynthesis in *C. islandica* thalli from high altitudes since HAP thalli showed their maximum P_G when measured under 5 °C. Low-temperature adaptation of photosynthesis in HAP thalli is supported also by temperature optimum of Φ_2 which was found lower in HAP than in LAP (see Fig. 1). Interpretation of results presented by Schipperges *et al.* (1995) also supports the low temperature adaptation of photosynthesis in *Cetraria* from colder sites (higher latitudes). The authors reported lower P_{Nmax} for *C. nivalis* population form Svalbard (79°N) than in population from southern Sweden (59°N) when both measured under 15 °C. After a 5 year-lasting acclimation of the two populations to the same conditions, difference in P_{Nmax} diminished.

Responses to dehydration: In lichens, severe dehydration leads to gradual inactivation of photobiont cells that induces decrease in photosynthetic rate. We found that moderate dehydration (to about 50 % of WSC) did not lead to substantial loss of photosynthetic activity in *C. islandica*. However, positive P_{Gmax} does not necessarily mean positive P_N because respiration was not evaluated in this study. Respiration rates might be high in lichens depending strongly on thalli temperature and hydration (Sundberg *et al.* 1999). We may estimate respiratory loss to about 50 % of P_{Gmax} because it typically varies within the range of 40-70 % under the temperatures from 5 to 15 °C (Sundberg *et al.* 1997, Palmqvist *et* Sundberg 2000). Therefore, it can be concluded that P_N at 50 % of WSD might be still positive in *C islandica.*

Two typical courses of photosynthetic parameters in response to thallus dehydration are usually found in lichens: (*1*) plateau type with decrease at high WSD, (*2*) parabolic type with substantial decrease both at low and high values of WSD (see, *e.g.*, Smith and Griffiths 1998). In our study, *C. islandica* showed both types of the courses (Fig. 3). The reason why LAP exhibited plateau type and HAP parabolic type of curve is yet unclear. There are, however, some indications (not shown here) that, appart from temperature, irradiance and other microclimatic parameters in habitats may affect the curve shape. Moreover, the course of photosynthetic parameters as related to dehydration might be also species-specific as shown for *C. rangiferina* and *Pseudevernia furfuracea* (Fig. 3).

The parabolic course shape in HAP of *C. islandica* is typical for some lichen species and has been documented by measurements both under laboratory (Deltoro *et al.* 1998) and field conditions (Lange and Green 1997). The decreased values of photosynthetic parameters at low WSD are usually explained by the limitation of CO_2 diffusion from the outside to the photobiont due to increased resistance of hydrated hyphal layer (Lange and Green 1997). Under these conditions, less CO_2 than required is available for algal photobiont, which is reflected in a decrease of P_N. If equilibrium between biochemical and photochemical processes of photosynthesis is assumed, then a decrease in P_N must be accompanied with a decrease in Φ_2. The reason for a similar decrease found in F_V/F_M at low WSD is not clear. We may hypothesise that the decrease of F_V/F_M at low WDS in HAP of *C. islandica* could be attributed to the functional changes in cells of symbiotic alga *Trebouxia* after full hydration.

Plateau type of Φ_2 and F_V/F_M courses during dehydration was found for both cyanobacterial (*Dagelia plumbea,* Gauslaa and Solhaug 1998) and algal lichen species (*Parmelia quercina,* Calatayud *et al.* 1997; *Hypogymnia physodes, Peltigera aphthosa,* Jensen *et al.* 1999). The response may be common in most poikilohydric plants, as described also for bryophytes (Deltoro *et al.* 1998). In our study, plateau type courses were found only in LAP in contrast to HAP of *C. islandica* (Fig. 3). The reason why high water content did not inhibit Φ_2 and F_V/F_M in LAP is not clear. Lange *et al.* (1997) checked the possible role of some secondary metabolites in this response but no ultimate explanation has yet been published. Dehydration of a lichen thallus is accompanied by some structural and functional changes in photosynthetic apparatus.

Among them, activation of zeaxanthin formation from violaxanthin is of great importance: the newly formed zeaxanthin serves as a dissipative pathway for excitation energy in PS2 (Calatayud *et al.* 1997). Another response of lichen thalli to dehydration is an increased synthesis of antioxidative enzymes and substrates (*e.g.*, superoxidismutase, ascorbate, and reduced glutathion). They are important for resistance to dehydration-induced oxidative stress. Also structural re-arrangement of chloroplast in the photobiont cells might be considered as a consequence of thalli dehydration because it has been found in a desiccating moss (Bartošková *et al.* 1999, Matoušková *et al.* 1999).

References

Ahmadjian, V.: The Lichen Symbiosis. – John Wiley, New York 1993.

Bartošková, H., Nauš, J., Výkruta, M.: The arrangement of chloroplasts in cells influences the reabsorption of chlorophyll fluorescence emmission. The effect of desiccation on the chlorophyll fluorescence spectra of *Rhizomnium punctatum* leaves. – Photosynth. Res. **62**: 251-260, 1999.

Calatayud, A., Deltoro, V.I., Barreno, E., del Valle-Tascon, S.: Changes in *in vivo* chlorophyll fluorescence quenching in lichen thalli as a function of water content and suggestion of zeaxanthin-associated photoprotection. – Physiol. Plant. **101**: 93-102, 1997.

Deltoro, V.I., Calatayud, A., Gimeno, C., Abadía, A., Barreno, E.: Changes in chlorophyll *a* fluorescence, photosynthetic CO_2 assimilation and xanthophyll cycle interconversions during dehydration in desiccation-tolerant and intolerant liverworts. – Planta **207**: 224-228, 1998.

Deltoro, V.I., Calatayud, A., Morales, F., Abadía, A., Barreno, E.: Changes in net photosynthesis, chlorophyll fluorescence and xanthophyll cycle interconversions during freeze-thaw cycles in the Mediterranean moss *Leucodon sciuroides*. – Oecologia **120**: 499-505, 1999.

Fontaniella, B., Vicente, C., Legaz, M.E.: The cryoprotective role of polyols in lichens: Effects of redistribution of RNase in *Evernia prunastri* thallus during freezing. – Plant Physiol. Biochem. **38**: 621-627, 2000.

Gauslaa, Y., Solhaug, K.A.: The significance of thallus size for the water economy of the cyanobacterial old-forest lichen *Dagelia plumbea*. – Oecologia **116**: 76-84, 1998.

Hovenden, M.J., Jackson, A.E., Seppelt, R.D.: Field photosynthetic activity of lichens in the Windmill Island Oasis, Wiekies Land, Continental Antarctica. – Physiol. Plant. **90**: 567-576, 1994.

Jensen, M., Chakir, S., Feige, G.B.: Osmotic and atmospheric dehydration effects in the lichen *Hypogymnia physodes*, *Lobaria pulmonaria*, and *Peltigera aphthosa*: an *in vivo* study of the chlorophyll fluorescence induction. – Photosynthetica **37**: 393-404, 1999.

Kappen, L., Schroeter, B., Hestmark, G., Winkler, J.B.: Field measurements of photosynthetis of umbilicarious lichens in winter. – Bot. Acta **109**: 292-298, 1996a.

Kappen, L., Schroeter, B., Scheidegger, C., Sommerkorn, M., Hestmark, G.: Cold resistance and metabolic activity of lichens below 0 °C. – Adv. Space Res. **18**(12): 119-128, 1996b.

Kappen, L., Sommerkorn, M., Schroeter, B.: Carbon acquisition and water relations of lichens in polar regions – potentials and limitations. – Lichenologist **27**: 531-545, 1995.

Kňazovický, L.: Západné Tatry. [Western Tatra Mountains.] – VSAV, Bratislava 1970. [In Slovak.]

Krall, J.P, Edwards, G.E.: Relationship between photosystem II activity and CO_2 fixation in leaves. – Physiol. Plant. **86**: 180-187, 1992.

Lange, O.L.: Ecophysiology of photosynthesis performance of poikilohydric lichens and homoiohydric Mediterranean sclerophylls. – J. Ecol. **76**: 915-937, 1988.

Lange, O.L., Green, T.G.A.: High thallus water contents can limit photosynthetic productivity of crustose lichens in the field. – Bibliotheca lichenol. **68**: 81-99, 1997.

Lange, O.L., Green, T.G.A., Reichenberger, H., Hesbacher, S., Proksch, P.: Do secondary substances in the thallus of a lichen promote CO_2 diffusion and prevent depression of net photosynthesis at high water content? – Oecologia **112**: 1-3, 1997.

Lange, O.L., Hahn, S.C., Meyer, A., Tenhunen, J.D.: Upland tundra in the foothills of the Brooks Range, Alaska, USA: Lichen long-term photosynthetic CO_2 uptake and net carbon gain. – Arct. alp. Res. **30**: 252-261, 1998.

Lichtenthaler, H.K., Wellburn, A.R.: Determinations of total carotenoids and chlorophylls *a* and *b* of leaf extracts in different solvents. – Biochem. Soc. Trans. **11**: 591-592, 1983.

Matoušková, M., Bartošková, H., Nauš, J., Novotný, R.: Reaction of photosynthetic apparatus to dark desiccation sensitively detected by the induction of chlorophyll fluorescence quenching. – J. Plant Physiol. **155**: 399-406, 1999.

Meyer, H., Santarius, K.: Short-term thermal acclimation and heat tolerance of gametophytes of mosses. – Oecologia **115**: 1-8, 1998.

Palmqvist, K., Sundberg, B.: Light use efficiency of dry matter gain in five macro-lichens: relative impact of microclimate conditions and species-specific traits. – Plant Cell Environ. **23**: 1-14, 2000.

Potvin, C., Lechowicz, M.J., Tardif, S.: The statistical analysis of ecophysiological response curves obtained from experiments involving repeated measures. – Ecology **71**: 1389-1400, 1990.

Reiter, R., Türk, R.: Investigation on the CO_2 exchange of lichens in the Alpine belt. II. Comparative patterns of net CO_2 exchange in *Cetraria islandica* and *Flavocetraria nivalis*. – Phyton **40**: 161-177, 2000.

Roháček, K., Barták, M.: Technique of the modulated chlorophyll fluorescence: basic concepts, useful parameters, and some applications. – Photosynthetica **37**: 339-363, 1999.

Schipperges, B.: Patterns of CO_2 gas-exchange and thallus water-content in Arctic lichens along a ridge profile near NY-Alesund, Svalbard. – Polar Res. **11**: 47-68, 1992.

Schipperges, B., Kappen, L., Sonesson, M.: Intraspecific variations of morphology and physiology of temperature to arctic populations of *Cetraria nivalis*. – Lichenologist **27**: 517-529, 1995.

Schroeter, B., Green, T.G.A., Kappen, L., Seppelt, R.D.: Carbon dioxide exchange at subzero temperatures. Field measurements on *Umbilicaria aprina* in Antarctica. – Cryptogam. Bot. **4**: 233-241, 1994.

Smith, E.C., Griffiths, H.: Intraspecific variation in photosynthetic responses of trebouxoid lichens with reference to the activity of a carbon concentrating mechanism. – Oecologia **113**: 360-369, 1998.

Sundberg, B., Campbell, D., Palmqvist, K.: Predicting CO_2 gain and photosynthetic light acclimation from fluorescence yield and quenching in cyano-lichens. – Planta **201**: 138-145, 1997.

Sundberg, B., Ehblad, A., Näsholm, T., Palmqvist, K.: Lichen respiration in relation to active time, temperature, nitrogen and ergosterol concentrations. – Funct. Ecol. **13**: 119-125, 1999.

van Kooten, O., Snell, J.F.H.: The use of chlorophyll fluorescence nomenclature in plant stress physiology. – Photosynth. Res. **25**: 147-150, 1990.

White, A.J., Critchley, C.: Rapid light curves: A new fluorescence method to assess the state of the photosynthetic apparatus. – Photosynth. Res. **59**: 63-72, 1999.