Limitation of photosynthetic processes in photosystem II in alpine mosses exposed to low temperatures: Response of chlorophyll fluorescence parameters

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Abstract

In this study, we evaluated the effects of low and sub-zero temperature on the fast chlorophyll fluorescence transient (OJIP) and OJIP-derived parameters in 4 different mosses: *Sphagnum girgensohnii*, *Polytrichum formosum*, *Hylocomium splendens* and *Pleurozium schreberi*. The low temperature stress was applied on the mosses for 90 min. at 3 different temperatures (5°C, -1°C and -10°C). To investigate the effects of this stress on the functioning of photosystem II (PS II), the chlorophyll fluorescence measurements were taken at control temperature (22°C) and, after a 90 min. acclimation period, at each experimental temperature. The shape of OJIP curves and chlorophyll fluorescence parameters were found temperature-dependent in all the species. The mosses differed in their sensitivity to the stress but general trends in response to low temperature were similar. The results support the idea that *S. girgensohnii* is less resistant to low temperature stress than the other species. We were also interested in the K and L steps in OJIPs, representing different disorders caused by low temperature. The K-step was seen in *P. formosum* and *P. schreberi* and the L-step in *H. splendens* and *S. girgensohnii*.

Key words: Kautsky effect, chlorophyll fluorescence transient, OJIP, low temperature stress, K-step, L-step

***List of symbols and abbreviations*:** ChlF – chlorophyll fluorescence, ET – electron transport, Q_A – primary quinone acceptor, LHC II – light-harvesting complex II, OEC – oxygen-evolving complex, OJIP – fast chlorophyll fluorescence transient, PQ – plastoquinone, PS II – photosystem II, RC – reaction centre

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Introduction

The fast chlorophyll fluorescence induction or Kautsky effect represents an increase of fluorescence signal during the first two seconds of light exposure of dark-adapted sample of a photosynthesizing organism. It has been used for the detection of stress effects in photosystem II (PS II) of higher plants and, recently, lichens (Marečková et al. 2018). The general trend of the fast fluorescence rise has been described as a polyphasic chlorophyll fluorescence growth reaching particular levels: O-J-I-P rise (Strasser et al. 1995). The O phase is the initial fluorescence when the reaction centres are fully oxidised after dark adaptation and chlorophyll fluorescence induced at the very begining of light exposure (typically 10 μs) is emitted exclusively from light harvesting complexes. With the time of light exposure the other chlorophyll fluorescence levels, J and I, appear. They are the points related to the reduction of plastoquinone pool by reduced Q_A. After 1.0-1.5 s of light exposure, the P peak appears. The P peak is the maximum chlorophyll fluorescence. The O-J phase, also called photochemical phase shows a gradual reduction of the primary quinone electron acceptor of PS II with a weak reduction of acceptors beyond Q_A. The following J-I phase, or thermal phase reflects the reduction of the plastoquinone (PQ) pool by reduced Q_A, the I-P phase has been explained by different mechanisms (Ilík et al. 2006). Fast chlorophyll fluorescence transients (here abbreviated as OJIPs) have been used in mosses to evaluate the response of PS II to a variety of stress conditions, such as *e.g.* thallus dehydration (Bartošková et al. 1999) as well as their ecophysiological specifity (Liepina et Ievinsh 2013, for epigeic vs epiphytic mosses, aquatic vs semi-aquatic mosses). Recently, OJIP have been used to quantify elevated nitrogen effects on moss physiological activity (Liu et al. 2016).

Under various stress conditions, two steps (the K-step and the L-step) are frequently analyzed. They are found between O and J levels of chlorophyll fluorescence. The K-step can be found at around 300 μs and it has been associated with a dissociation of the oxygen-evolving-complex (OEC) (Oukarroum et al. 2007). There is evidence that the K-step arises when the electron flow to the acceptor side exceeds the electron flow from the donor side, leading to an oxidation of the RC (Strasser 1997). The L-step usually appears at around 100-150 μs (Goltsev et al. 2016) and its presence indicates that the PS II units are less grouped or less energy was being exchanged between independent PS II units (Mathur et al. 2011a). The shapes of the OJIP transients are similar in a great variety of plant species in their optimal physiological state, however, some differences exist in cyanobacteria (*see* e.g. Tsimili-Michael et al. 2009, Bueno et al. 2004).

In higher plants and algae the OJIP shape differ from optimum according to the negative factors affecting physiological state of the sample, its photosynthetic apparatus in particular. This makes the fast chlorophyll fluorescence induction a useful tool for non-invasive detection of various stress effects (Stirbet et Govindjee 2011, Stirbet et al. 2018), both chemical or physical stresses (Kalaji et Guo 2008). The chlorophyll fluorescence parameters derived from OJIPs also change with different stresses. In a majority of parameters, the stress causes a negative effect. This is true for the maximal photochemical efficiency of PS II (F_V/F_M), the probability of an electron to be used for electron transport (ψ_0), the potential for energy conservation (PI_{ABS}), the efficiency of ET (φ_{Eo}) and the flux of ET per reaction centre (ET_0/RC). Decrease with stress; however, is apparent in the quantum yield of energy dissipation (φ_{Do}) and the flux of dissipated energy (DI_0/RC),

as the time to reach maximal chlorophyll fluorescence (φ_{PAV}), increase in stress situations (Strasser et al. 2004).

In plants exposed to low and subzero temperature, water moves out from the cell to form extracellular ice, imposing a considerable desiccation stress (Burke et al. 1976). The withdrawal of water molecules from cell compartments during freezing results in the changes in the conformation of biomolecules (Crowe et al. 1990). These changes alter the arrangement of proteins with thylakoid membranes, resulting in a reduction of the light absorption surface on the membranes and in an increase in self-absorption of emitted fluorescence. During frozen periods, proteins conformation inhibits the energy transfer between LHC II and PS II, leading to a reduction in the number of functional PS II centres (Lovelock et al. 1995).

In this study we evaluated the response of PS II to low and subzero temperature stress in 4 different moss species from alpine biome applying fast chlorophyll fluorescence induction. We hypothesized that the moss species show generally the same response in OJIP shape to temperature. On the other hand, we expected species-specific differences in the sensitivity to the stress caused by subzero temperature.

Material and Methods

Species characteristics

Four different moss species were used in this study: *Sphagnum girgensohnii*, *Polytrichum formosum*, *Hylocomium splendens* and *Pleurozium schreberi*. *P. schreberi* is reported for open habitats of alpine zone in the Jeseniky Mts. (Ziedler et al. 2010), specifically for the Praděd natural reserve (Klos et al. 2012) and the Tabulové skály. Both *P. schreberi* and *H. splendens* are considered typical moss species of species-poor grasslands with dominating *Nardus stricta* near or above the alpine timberline (Krahulec et al. 2007). *P. schreberi* is one of the components of the moss community forming patterened vegetation cover of the rocks (for the species forming the community, *see* Kučera et al. 2009). In the Jeseniky Mts., *H. splendens* is reported for subalpine springs but it is rare above timberline (Táborská 2013). *P. formosum* and *S. girgensohni* are typical species reported from the Jeseniky Mts.: the first one from mountainous spruce forests and the second one from wet localities such as lake margins, peat bogs, springs and streams.

Sample collection and handling

Samples of *S. girgensohnii* and *P. fomosum* were collected in Kapitánská stezka, meanwhile *H. splendens* and *P. schreberi* were collected at the Tabulové skály rocks in September 2018 (*see* Table 1).

The Tabulové skály rocks are located 300 – 450 m N of the top of the Praděd Mt. at the altitude ranging 1430 – 1450 m a.s.l. The locality belongs among unique ones in the Jeseníky Mts. with several species of bryophytes known only from this locality. After the collection, the samples were transferred to Brno, where they were used for the experiment. The samples were stored in the roof of the Faculty of Science during all the experiment.

Species	Coordinates		Altitude
	Locality: Tabulové skály rocks		
Pleurozium schreberi	50° 5´ 10.12´´ N	17° 13´ 50.29´´ E	1 456 m a.s.l.
Hylocomium splendens	50° 5´ 10.12´´ N	17° 13´ 50.29´´ E	1 456 m a.s.l.
	Locality: Kapitánská stezka		
Polytrichum formosum	50° 3´ 49.16´´ N	17° 17´ 38.00´´ E	940 m a.s.l.
Sphagnum girgensohnii	50° 3´ 54.07´´ N	17° 17´ 12.06´´ E	959 m a.s.l.

Table 1. Overview of the sampling localities in the Jeseníky Mts.

Chlorophyll fluorescence measurements

For the experiment, 10 samples were selected from each moss species. These samples were acclimated at room temperature (usually 22°C) for 30 min. before the first measurements. This was the temperature selected as reference or control temperature. Then, a cooling chamber was used to decrease the temperature of the mosses to target temperature and acclimate for 90 min. This was done at 3 different temperatures: 5°C, -1°C and -10°C. During the cooling period the samples were illuminated by a lamp, and the sample temperature was measured by the CuCo thermocouple linked to an EdgeBox datalogger (Environmental Measurement Systems, Brno, Czech Republic). After each new temperature the measurements were taken by the fluorometer FluorPen (Photon Systems Instrument, Drásov, Czech Republic). The fluorometer FluorPen enables a precise measurement of 27 chlorophyll fluorescence parameters. We selected 9 of them (Table 2) based on their importance and relation to this project.

In this study, we used 10 samples of each moss species. Therefore, 10 measurements were taken for each species at each different temperature, resulting in 40 different OJIP curves and chlorophyll fluorescence derived parameters values for every moss species. The means of this data were calculated for the further analysis, so finally, in the different species, each temperature consisted in one OJIP curve and one value for every parameter.

For visualisation of the K-step the data was double normalized between F_0 and F_J (2 ms), expressed as $W_{OJ} = (F_t - F_0)/(F_J - F_0)$, this enables visualisation at around 240-300 μs (Marečková et al. 2017). F_J values were directly taken from the OJIP curves because the exact time of the J-step usually varies with stress. On the other hand, for the L-step, the data was double normalized as $W_{OK} = (F_t - F_0)/(F_K - F_0)$, where FK represents the chlorophyll fluorescence value at 301 μs. For L- and K-step, the data used were the difference between the temperature-stressed samples and control (mosses measured at 20°C). The differences were plotted as a function of time, *i.e.* the time of light exposure.

Different chlorophyll fluorescence parameters were used in this investigation. They were directly derived from the OJIP data and they can be described as follows: F_V/F_M is the maximal quantum yield of PS II, ψ_0 is the probability that a trapped exciton is used for electron transport beyond Q_A, φ_{Eo} is the quantum yield for ET, φ_{Do} is the quantum yield (at t=0) of energy dissipation, φ_{PAV} is the time to reach maximal chlorophyll fluorescence, PI_{ABS} is the potential for energy conservation from exciton to the reduction of intersystem electron acceptors, ABS/RC is the absorption flux per RC, TR_0/RC is the trapped energy flux per RC, ET_0/RC is the ET flux per RC and DI_0/RC is the flux of dissipated excitation energy at time 0 (Strasser et al. 2004).

$\psi_0 = 1 - V_J = ET_0/TR_0$ *Eqn.* 1

$\varphi_{Eo} \equiv ET_0 / ABS = [1 - (F_0/F_M)] . \psi_0$ *Eqn.* 2

$\varphi_{Do} = 1 - \varphi_{Po} = F_0/F_M$ *Eqn.* 3

$\varphi_{PAV} = \varphi_{Po} . (S_M / t_{FM})$ *Eqn.* 4*

$PI_{ABS} = (RC/ABS) . [\varphi_{Po} / (1 - \varphi_{Po})] . [\psi_0 / (1 - \psi_0)]$ *Eqn.* 5

$ABS/RC = M_0 . (1 / V_J) . (1 / \varphi_{Po})$ *Eqn.* 6

$TR_0/RC = M_0 . (1 / V_J)$ *Eqn.* 7

$ET_0/RC = M_0 . (1 / V_J) . \psi_0$ *Eqn.* 8

$DI_0/RC = (ABS/RC) - (TR_0/RC)$ *Eqn.* 9

Table 2. Overview of the chlorophyll fluorescence parameters calculated from OJIPs. * - The equation 4 is taken from the FluorPen manual (PSI, Drásov, Czech Republic) [1].

Results and Discussion

The OJIP curves plotted for particular experimental temperature showed species-specific differences (Fig. 1). In *S. girgensohnii* and *H. splendens* the J-step increased with moss temperature decrease; however, from O to P, the more pronounced decrease in temperature, the lower values of ChlF. Low temperatures, contrastingly resulted in a general increase of ChlF values for *P. formosum*, while ChlF dropped in *P. schreberi* due to decreasing samples temperature.

Background chlorophyll fluorescence (F_0 at O point) increased with drop of temperature in majority of species (*see* Fig. 1), however, *P. schreberi* showed no change. This is contradictory to the finding of Deltoro et al. (1999) who reports a decrease in F_0 in *Leucodon sciuroides* during freezing. Similarly, Lovelock et al. (1995) found F_0 decrease with low-rate freezing of Antarctic moss *Grimmia antarctici*. The likely reason is that the authors used relatively high light during freezing which induced photoinhibitory changes to the chloroplastic photosynthetic apparatus resulting in F_0 decrease. The four experimental moss species used in our study, however, showed a high resistance to photoinhibition (Barták, experimental data from other experiment, not shown here). In the two studies (Deltoro et al. 1999, Lovelock et al. 1995), however, F_V/F_M decline with freezing is shown as well as in our study (*see* Fig. 1). As seen in Fig. 1, *S. girgensohnii*, followed by *P. formosum*, displays the biggest changes on the shapes; moreover, *H. splendens* and *P. schreberi* do not display as much differences as the other studied species.

Subsequently, K-step and L-step were calculated using the method explained in *Chlorophyll fluorescence measurements*. The K-step (Fig. 2), representing a dissociation of the OEC, was clearly found in *P. formosum* at 230 μs and in *H. splendens* at 200 μs. In both cases this phenomenon was just seen at the temperature of -10°C. The ratios W_{OK}, W_{OJ} were used to show the temperature-induced changes in the amplitude in the K step (*see* Fig. 2, Fig. 3). The higher the Wk values, the larger was the subzero temperature-induced damage to the PS II donor side. Similar donor-side damage has been earlier found for other stressors such as *e.g.* drought stress (Guha et al. 2013), salinity stress (Venkatesh et al. 2012), heat stress (Lazár et al. 1997, Mathur et al. 2011b), leaf senescence (Zhang et al. 2012).

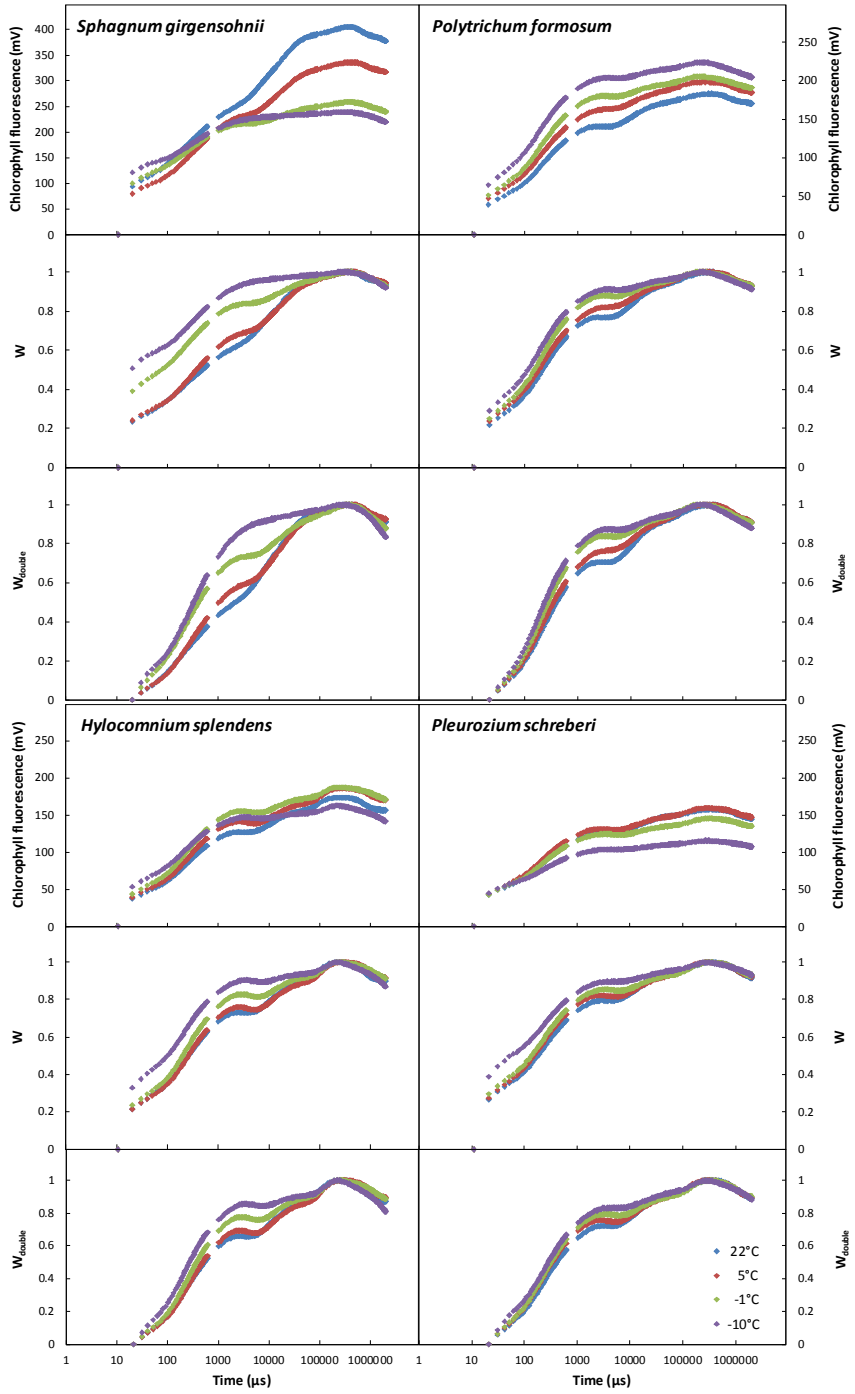

Fig. 1. OJIP curves, normalized curves (W) and double normalized curves (W_{double}) of the mosses species.

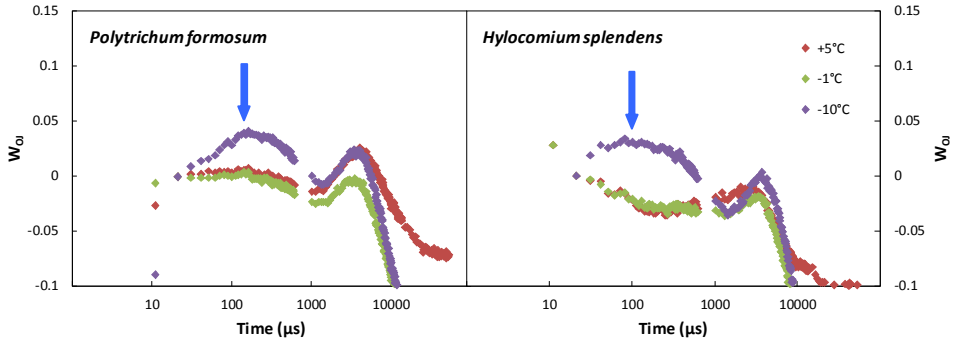

Fig. 2. Differential plot of the initial part of OJIP – W_{OJ} (relative variable fluorescence normalized to the amplitude of the J phase) in response to experimental temperature for K-step.
Equation: $W_{OJ} = (F_t - F_0)/(F_J - F_0)$. K-step found for -10°C at ~ 200 µs is indicated by an arrow.

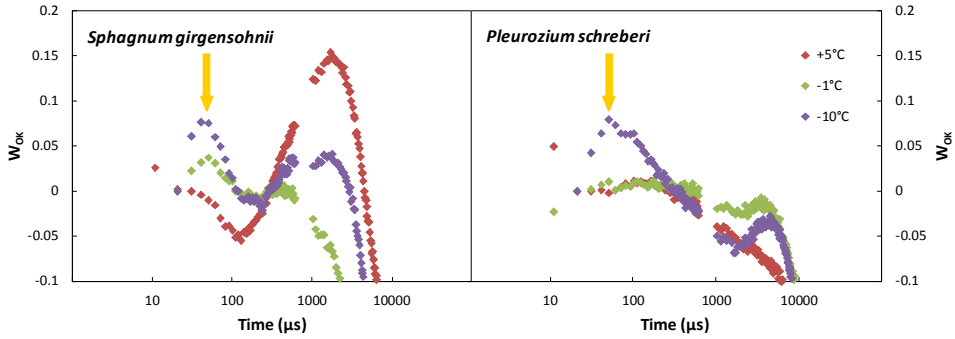

Fig. 3. Differential plot of the initial part of OJIP – W_{OK} (relative variable fluorescence normalized to the amplitude of the K phase) in response to experimental temperature for L-step.
Equation: $W_{OK} = (F_t - F_0)/(F_K - F_0)$. L-step found for -10°C at ~ 50 µs is indicated by an arrow.

On the other hand, the L-step (Fig. 3) was found in *S. girgensohnii* at the time of 50 µs and at 54 µs in *P. schreberi*. The L-step, that reflects changes in the connectivity between LHCs and RC, was visualized in both species at -10°C; however, *S. girgensohnii* also reflected it at the temperature of -1°C. These results are comparable to the K- and L-step found in Antarctic lichen treated by subzero temperature (Marečková et Barták 2017, Marečková et al. 2018, under review).

The parameters related with quantum yields, electron transport flux or performance index (F_V/F_M, ψ_0, φ_{Eo}, PI_{ABS} and ET_0/RC) decreased with low temperatures (Fig. 4). Most of fully developed plant leaves under no stress conditions show a maximum F_V/F_M value of 0.83 The decrease of this parameter reveals stress-induced changes in PS II functions and declines the yield of ET (Kalaji et al. 2011). At control temperature, 22°C, the mosses showed high F_V/F_M values: *S. girgensohnii* with 0.8 F_V/F_M value, *P. formosum* and *H. splendens* with 0.78, and *P. schreberi* with 0.75. The parameters related with absorption flux, energy dissipation and the time to reach maximal ChlF (φ_{Do}, φ_{PAV}, ABS/RC and DI_0/RC) increased, as expected, with the lowering of thallus temperature.

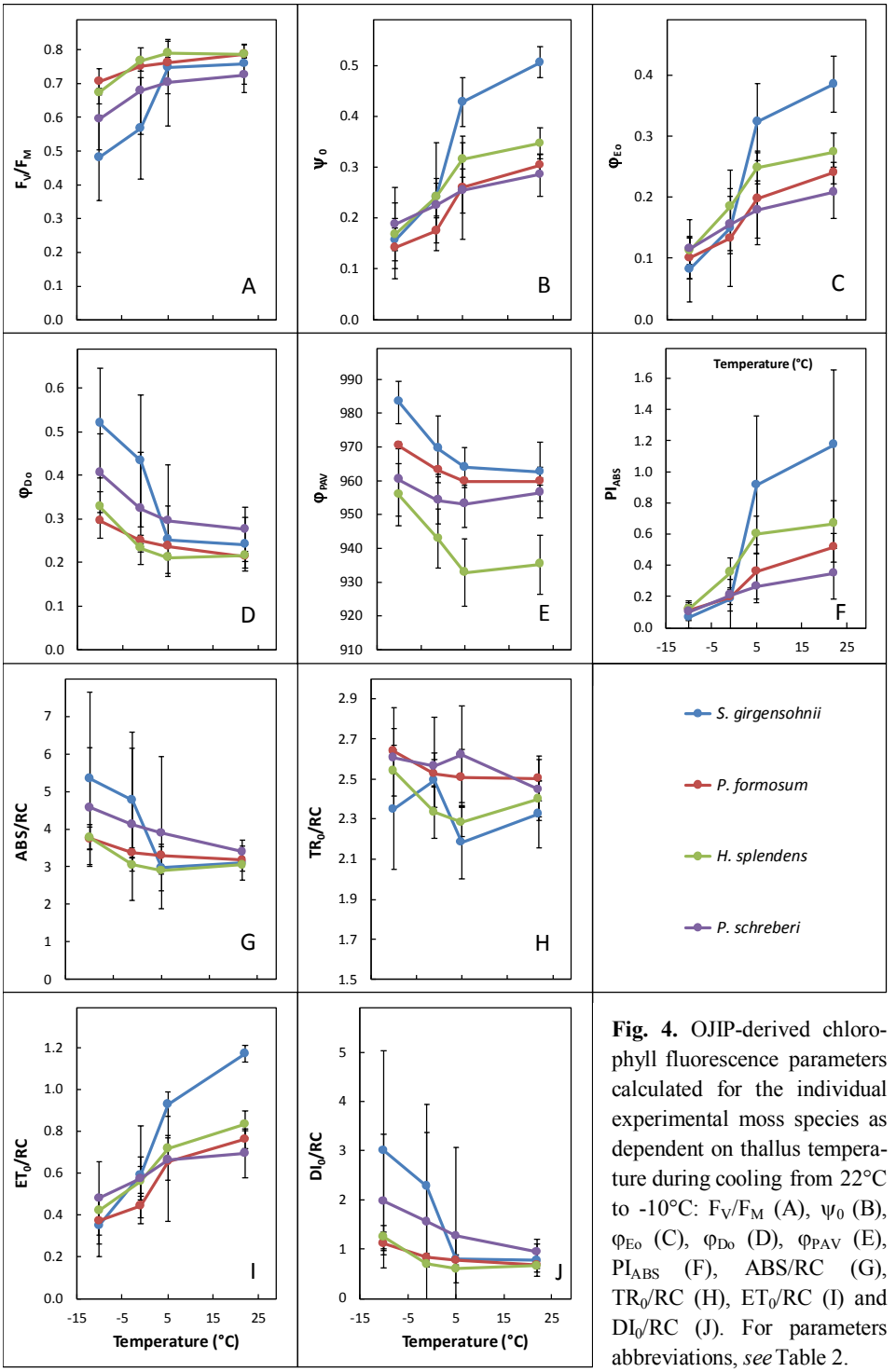

Fig. 4. OJIP-derived chlorophyll fluorescence parameters calculated for the individual experimental moss species as dependent on thallus temperature during cooling from 22°C to -10°C: F_V/F_M (A), ψ_0 (B), φ_{Eo} (C), φ_{Do} (D), φ_{PAV} (E), PI_{ABS} (F), ABS/RC (G), TR_0/RC (H), ET_0/RC (I) and DI_0/RC (J). For parameters abbreviations, *see* Table 2.

TR_0/RC, representing trapped energy flux (leading to Q_A reduction) per RC, was the only parameter that remained unaffected by low temperatures.

TR_0/RC in *S. girgensohnii* shows the most different parameter values compared to the other moss species. Also, it shows irregular trend with the temperature decrease, *i.e.* it fluctuates up and down at -1°C and +5°C, respectively. In general, *S. girgensohnii* exhibited slightly different temperature-response curves from the other species. There is one exception with φ_{PAV}, where *H. splendens* shows a similar effect as *S. girgensohnii*; but also, in TR_0/RC there is no correlation between the values and the stress.

In conclusion, the shape of OJIP curves was found temperature-dependent in all the species and, although the mosses differed in their sensitivity to the stress, general trends in response to low temperatures were similar. The results support that *S. girgensohnii* is less resistant to low temperature stress than the other species. The temperature-induced changes in the OJIP parameters recorded in the experimental species suggest the indicative value of the parameters. Numerical values of all the parameters were found sensitive to the lowering of thallus temperature. Therefore, they indicated altered PS II functioning before actual appearance of visible stress symptoms could be distinguished (Kalaji et al. 2011). This study supports the idea that fast chlorophyll fluorescence induction is a useful and non-invasive method for detecting stress in alpine mosses.

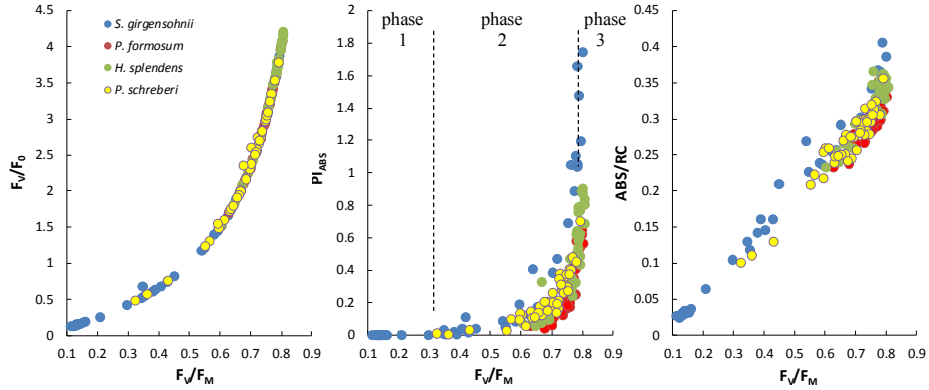

Fig. 5. Relationship between maximum quantum efficiency of PS II (F_V/F_M) and (A) photochemical efficiency of PSII (F_V/F_0), (B) Performance Index (PI_{ABS}) and (C) density of active reaction centres (ABS/RC) (C) in the experimental moss species. Blue symbols - *Sphagnum girgensohnii*, red symbols - *Polytrichum formosum*, green symbols - *Hylocomium splendens*, yellow symbols - *Pleurozium schreberi*.

The F_V/F_0 to F_V/F_M relation showed an exponential relationship for pooled data for the studied moss species (Fig. 5). The relationship was similar to that found by Lepina et Ievinsh (2013) and attributed to a strong relation between the minimal and the maximal fluorescence level of PS II in the studied moss species. Performance index (PI_{ABS}) in dependence to F_V/F_M showed a triphasic response. No photochemical performance of PS II was seen at the F_V/F_M values below 0.4 (phase 1). Then, an exponential increase of the PI values was apparent at the F_V/F_M values below 0.77

(phase 2). Finally, a steep linear increase of PI_{ABS} values was found at the F_V/F_M values above 0.77. The ABS/RC to F_V/F_M relation increased linearly with the increase of F_V/F_M up to 0.75, followed by much steeper rise in ABS/RC at the F_V/F_M values above 0.75. Similarly to PI_{ABS}, ABS/RC showed species-specific clusters. The relationships between the three chorophyll fluorescence parameters indicated, similarly to Liepina et Ievinsh (2013), three functionally different stages in the photochemical processes of photosynthesis. No efficient photochemical reactions of photosynthesis occurred at $F_V/F_M < 0.4$ in spite of the linearly increasing proportion of active RCs as indicated by the changes in ABS/RC (*see* Fig 5, C). Then, in the range of $0.30 < F_V/F_M < 0.77$, a slow restoration of photochemical efficiency was apparent. A further increase of photochemical performance at the F_V/F_M values above 0.77 could be associated with the further improvement of photochemical at optimal hydration.

References

BARTOŠKOVÁ, H., KOMENDA, J. and NAUŠ, J. (1999): Functional changes of photosystem II in the moss *Rhizomnium punctatum* (Hedw.) induced by different rates of dark desiccation. *Plant Physiology*, 154: 597-604.

BUENO, M., FILLAT, M. F., STRASSER, R. J., MALDONADO-RODRIGUEZ, R., MARINA, N., SMIENK, H., GOMEZ-MORENO, C. and BARJA, F. (2004): Effects of lindane on the photosynthetic apparatus of the cyanobacterium *Anabaena*: Fluorescence induction studies and immunolocalization of Ferredoxin-$NADP^+$ reductase. *Environmental Science and Pollution Research International*, 11: 98-106.

BURKE, M., GUSTA, L., QUAMME, H., WEISER, C. and LI, P. (1976): Freezing and injury in plants. *Annual Review Plant Physiology*, 27: 507-528.

CROWE, J., CARPENTER, J., CROWE, L. and ANCHORDOGUY, T. (1990): Are freezing and dehydration similar stress vectors? A comparison of modes of interaction of stabilizing solutes with biomolecules. *Cryobiology*, 27(3): 219-231.

DELTORO, V., CALATAYUD, Á., MORALES, F., ABADÍA, A. and BARRENO, E. (1999): Changes in net photosynthesis, chlorophyll fluorescence and xanthophyll cycle interconversions during freeze-thaw cycles in the Mediterranean moss *Leucodon sciuroides*. *Oecologia*, 120(4): 499-505.

GOLTSEV, V.N., KALAJI, H.M., PAUNOV, M., BĄBA, W., HORACZEK, T., MOJSKI, J., KOCIEL, H. and ALLAKHVERDIEV, S.I. (2016): Variable chlorophyll fluorescence and its use for assessing physiological condition of plant photosynthetic apparatus. *Russian Journal of Plant Physiology*, 63(6): 869-893.

GUHA, A., SENGUPTA, D. and REDDY, A.R. (2013): Polyphasic chlorophyll a fluorescence kinetics and leaf protein analyses to track dynamics of photosynthetic performance in mulberry during progressive drought. *Journal of Photochemistry and Photobiology B: Biology*, 119: 71-83.

ILÍK, P., SCHANSKER, G., KOTABOVÁ, E., VÁCZI, P., STRASSER, R. J. and BARTÁK, M. (2006): A dip in the chlorophyll fluorescence induction at 0.2–2 s in *Trebouxia*-possessing lichens reflects a fast reoxidation of photosystem I. A comparison with higher plants. *Biochimica et Biophysica Acta*, 1757: 12-20.

KALAJI, H., GUO, P. (2008): Chlorophyll fluorescence: A useful tool in barley plant breeding programs. *In*: A. Sánchez, S. Gutiérrez (eds.): Photochemistry Research Progress, 12: 447-471.

KALAJI, H., BOSA, K., KOSCIELNIAK, J. and HOSSAIN, Z. (2011): Chlorophyll *a* Fluorescence – A usefull tool for the early detection of temperature stress in spring barley (*Hordeum vulgare* L.). *A Journal of Integrative Biology*, 15(12): 925-934.

KLOS, A., RAJFUR, M., ŠRÁMEK, I. and VÁCLAVEK, M. (2012): Mercury concentration in lichen, moss and soil samples collected from the forest areas of Praděd and Glacensis Euroregions (Poland and Czech Republic). *Environmental Monitoring and Assessment,* 184: 6765-6774.

KRAHULEC, F., CHYTRÝ, M. and HÄRTEL, H. (2007): Smilkové trávníky a vřesoviště (Calluno-Ulicetea). Nardus grasslands and heathlands. – *In*: Chytrý M. (ed.): Vegetace České republiky. 1. Travinná a keříčková vegetace [Vegetation of the Czech Republic. 1. Grassland and Heathland Vegetation], pp. 281–319, Academia, Praha. (In Czech).

KUČERA, J., ZMRHALOVÁ, M., SHAW, B., KOŠNAR, J., PLÁŠEK, V. and VÁŇA, J. (2009): Bryoflora of the selected localities of the Hrubý Jeseník Mts summit regions. Časopis Slezského zemského muzea. Série A, Vědy přírodní, Opava (A), 58: 115-167. ISSN 1211-3026 (In Czech).

LAZÁR, D., ILÍK, P. and NAUŠ, J. (1997): An appearance of K-peak in fluorescence induction depends on the acclimation of barley leaves to higher temperatures. *Journal of Luminescence*, 72–74: 595-596.

LIEPINA, L., IEVINSH, G. (2013): Potential for fast chlorophyll a fluorescence measurement in bryophyte ecophysiology. *Estonian Journal of Ecology*, 62: 137-149.

LIU, B. Y,, LEI, C. Y., JIN, J. H., GUAN, Y. Y., LI, S., ZHANG, Y. S. and LIU, W. Q. (2016): Physiological responses of two moss species to the combined stress of water deficit and elevated N deposition (II): Carbon and nitrogen metabolism. *Ecology and Evolution*, 6: 7596-7609.

LOVELOCK, C. E, JACKSON, A. E., MELICK, R. D. and SEPPELT, R. (1995): Reversible photoinhibition in Antarctic moss during freezing and thawing. *Plant Physiology*, 109: 955-961.

MAREČKOVÁ, M., BARTÁK, M. (2017): Short-term responses of primary processes in PS II to low temperature are sensitively indicated by fast chlorophyll fluorescence kinetics in Antarctic lichen *Dermatocarpon polyphyllizum*. *Czech Polar Reports*, 7(1): 74-82.

MAREČKOVÁ, M., BARTÁK, M. and HÁJEK, J. (2018, under review): Temperature effects on photosynthetic performance of Antarctic lichen *Dermatocarpon polyphyllizum* - chlorophyll fluorescence study. *Polar Biology*, manuscript after revision.

MATHUR, S., ALLAKHVERDIEV, S. and JAJOO, A. (2011a): Analysis of high temperature stress on the dynamics of antenna size and reducing side heterogeneity of photosystem II in wheat leaves (*Triticum aestivum*). *Biochimica et Biophysica Acta*, 1807(1): 22-29.

MATHUR, S., JAJOO, A., MEHTA, P. and BHARTI, S. (2011b): Analysis of elevated temperature-induced inhibition of photosystem II using chlorophyll a fluorescence induction kinetics in wheat leaves (*Triticum aestivum*). *Plant Biology*, 13: 1-6.

OUKARROUM, A., MADIDI, S. E., SCHANSKER, G. and STRASSER, R. J. (2007): Probing the responses of barley cultivars (*Hordeum vulgare* L.) by chlorophyll *a* fluorescence OLKJIP under drought stress and re-watering. *Environmental and Experimental Botany*, 60: 438-446.

STIRBET, A., GOVINDJEE (2011): On the relation between the Kautsky effect (chlorophyll *a* fluorescence induction) and photosystem II: Basics and applications of the OJIP fluorescence transient. *Journal of Photochemistry and Photobiology. B, Biology*. 104: 236-57.

STIRBET, A., LAZÁR, D., KROMDIJK, J. and GOVINDJEE (2018): Chlorophyll *a* fluorescence induction: Can just a one-second measurement be used to quantify abiotic stress responses? *Photosynthetic*a, 56: 86-104.

STRASSER, R.J., SRIVASTAVA, A. and GOVINDJEE (1995): Polyphasic chlorophyll a fluorescence transient in plants and cyanobacteria. *Photochemistry and Photobiology*, 61(1): 32-42.

STRASSER, R. J. (1997): Donor side capacity of photosystem II probed by chlorophyll *a* fluorescence transients. *Photosynthesis Research*, 52: 147-155.

STRASSER, R.J., TSIMILLI, M. and SRIVASTAVA, A. (2004): Analysis of the chlorophyll *a* fluorescence transient. *In*: G. C.Papageorgiou, Govindjee (eds): Chlorophyll *a* fluorescence. Advances in Photosynthesis and Respiration, 19: 321-362.

TÁBORSKÁ, M. (2013): Mountain spring bryophyte communities in the Hrubý Jeseník Mts. and the main driving environmental factors. *Zprávy Vlastivědného muzea v Olomouci*, 305: 31-50 (In Czech).

TSIMILLI-MICHAEL, M., STAMATAKIS, K. and PAPAGEORGIOU, G. C. (2009): Dark-to-light transition in Synechococcus sp. PCC 7942 cells studied by fluorescence kinetics assesses

plastoquinone redox poise in the dark and photosystem II fluorescence component and dynamics during state 2 to state 1 transition. *Photosynthesis Research*, 99(3): 243-255.

VENKATESH, J., UPADHYAYA, CH.P., YU, J.-W., HEMAVATHI, A., KIM, D. H. and STRASSER, R. J. (2012): Chlorophyll *a* fluorescence transient analysis of transgenic potato overexpressing D-galacturonic acid reductase gene for salinity stress tolerance. *Horticulture Environment and Biotechnology*, 53: 320-328.

ZHANG, Z., LI, G., GAO, H., ZHANG, L., YANG, CH., LIU, P. and MENG, Q. (2012): Characterization of photosynthetic performance during senescence in stay-green and quick-leaf-senescence *Zea mays* L. Inbred Lines. *Plos One*, 7: e42936.

ZIEDLER, M., BANAŠ, M., DUCHOSLAV, M. and LEŠKOVÁ, M. (2010): The impact of dwarf pine plantation on alpine plant communities in the Hrubý Jeseník Mts. *Příroda*, Praha, 29: 37-50 (In Czech).

Web sources / Other sources

[1] FluorPen manual, Photon Systems Instruments (PSI), Drásov, Czech Republic
http://www.psi.cz/download/document/manuals/fluorpen/FluorPen_Monitoring_Manual_2018.pdf