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RESPONSE OF PHOTOSYNTHETIC APPARATUS OF TWO *DESCHAMPSIA* SPECIES WITH DIFFERENT DISTRIBUTION AREAS ON ABIOTIC STRESS

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Deschampsia antarctica (endemic of Antarctic region) and *Deschampsia caespitosa* (inhabitant of moderate climate regions) are two plant species of *Poaceae.* The influences of UV-B radiation and H_2O_2 on photosynthetic apparatus of these plants were studied. UV-B radiation induced degradation of chlorophyll *a* and *β*-carotene in leaves of plants of both *Deschampsia* species. The content of galactolipids in leaves of both species under conditions of UV-B radiation varied significantly, but comparatively stable sulfoquinovosyldiacylglycerol (SQDG) content was observed. UV-B radiation caused slight decrease of Q_A pool oxidation level in *D. antarctica* leaves and increase of this index in leaves of *D. caespitosa* plants. Also UV-B action induced slight decrease of non-photochemical quenching in *D. caespitosa* leaves, but PS II quantum efficiency of charge separation φ_p was unchanged. The ratio between the monomeric and oligomeric forms of LHC II (LHCP1/LHCP3) in photosynthetic apparatus of leaves of irradiated plants increased, especially significantly in leaves of *D. caespitosa* plants. H_2O_2 treatment cause insignificant decrease of SOD activity of both species. Pigment composition was characterized by increase of carotenoids content in leaves of *D.antarctica* plants and chlorophyll *a* content in both species. Glycolipid content was stable and SQDG content slightly increased in leaves of *D.antarctica* plants after H_2O_2 treatment.
Key words: *Deschampsia*, UV-B radiation, carotenoids, glycolipids.

Відповідь фотосинтетичного апарату двох видів *Deschampsia* з різним ареалом розповсюдження на абіотичний стрес.
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Реферат. *Deschampsia antarctica* (ендемік Антарктичного регіону) і *Deschampsia caespitosa* (представник регіонів помірного клімату) – два види рослин родини *Poaceae.* Досліджували вплив УФ-В випромінювання та H_2O_2 на фотосинтетичний апарат цих рослин. УФ-В випромінювання викликало деградацію хлорофілу *a* та *β*-каротину в листках рослин двох видів *Deschampsia.* Вміст галактоліпідів в листках обох видів в умовах УФ-В випромінювання суттєво варіював, але спостерігався відносно стабільний вміст сульфохіновозилдіацилгліцеролу. УФ-В випромінювання викликало неістотне зниження рівня окиснення пулу Q_A в листках *D. antarctica* та підвищення цього показника в листках *D. caespitosa.* Хоча дія УФ-В викликала невелике зниження нефотохімічного гасіння в листках *D. caespitosa,* квантова ефективність *ФС II* залишалася незмінною. Співвідношення між мономерними та олігомерними формами LHC II (LHCP1/LHCP3) у фотосинтетичному апараті опромінених рослин обох видів *Deschampsia* підвищувалося особливо суттєво для *D. caespitosa.* Обробка рослин H_2O_2 викликала несуттєве зниження активності СОД в обох видів. Пігментний склад характеризувався підвищенням вмісту каротиноїдів у листках рослин *D. antarctica* та вмісту хлорофілу *a* у обох видів. Вміст гліколіпідів у листках був стабільним, а вміст СХДГ дещо підвищувався після обробки H_2O_2 рослин *D.antarctica*.

Ответ фотосинтетического аппарата двух видов *Deschampsia* с различным ареалом распространения на абиотический стресс.
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Реферат. *Deschampsia antarctica* (эндемик Антарктического региона) и *Deschampsia caespitosa* (представитель регионов умеренного климата) – два вида растений семейства *Poaceae.* Исследовали

влияние УФ-В излучения и H_2O_2 на фотосинтетический аппарат этих растений. УФ-В излучение вызывало деградацию хлорофилла *a* и *β*-каротина в листьях растений двух видов *Deschampsia.* Содержание галактолипидов в листьях обоих видов в условиях УФ-В излучения существенно варьировало, но наблюдалось относительно стабильное содержание сульфохиновозилдиацилглицерола (СХДГ). УФ-В излучение вызывало несущественное снижение уровня окисления пула Q_A в листьях *D. antarctica* и повышение этого показателя в листьях *D. caespitosa.* Хотя действие УФ-В вызывало несущественное снижение нефотохимического тушения в листьях *D. caespitosa,* квантовая эффективность ФС II оставалась неизменной. Соотношение между мономерными и олигомерными формами LHC II (LHCP1/LHCP3) в фотосинтетическом аппарате облученных растений обоих видов *Deschampsia* повышалось особенно существенно для *D. caespitosa.* Обработка растений H_2O_2 вызывала несущественное снижение активности СОД у обоих видов. Пигментный состав характеризовался повышением содержания каротиноидов в листьях растений *D. antarctica* и содержания хлорофилла *a* у обоих видов. Содержание гликолипидов в листьях было нестабильным, а содержание СХДГ немного повышалось после обработки растений *D.antarctica* H_2O_2.

Introduction

The Antarctic geobotanical zone is a hostile environment for plant growth. Low temperatures, high light and stratospheric ozone depletion (causing increases in solar UV-B radiation) cause the formation of active oxygen species. Declines in global concentrations of stratospheric ozone over the past 15 years led to increase in levels of ultraviolet-B radiation (UV-B; 280–315 nm) reaching the earth's surface (Madronich et al., 1998) which is most pronounced in Antarctica (UNEP, 1998; Xiong, Day, 2001). High-intensity light and low temperatures in their turn can damage the photosynthetic apparatus of plants. Thus, increased solar UV-B radiation together with high light and low temperatures are the main abiotic factors which cause the formation of reactive oxygen species (ROS) inducing oxidative stress and disturbance of photosynthetic process. It is known, that dominant ROS in UV-irradiated plant leaves was $O_2^{-\bullet}$, while $^1O_2$ was minor **(**Hideg *et al.*, 2002**).** These species react with lipids, proteins, pigments, and nucleic acids and cause lipid peroxidation, membrane damage, inactivation of enzymes, thus affecting cell viability. Therefore, an efficient mechanism of ROS scavenging would contribute to support of photosynthetic activity and plant survival in Antarctic geobotanical zone. There are some metabolic pathways to defend photosynthesis mechanism from ROS action. The most important of them are the scavenging of photosynthetic apparatus from ROS with help of antioxidative enzymes and quenching of ROS by carotenoids. The antioxidative system of plants contain high- (antioxidative enzymes) and low-molecular (ascorbate, glutathione) compounds, that are principally constitutive and vary in plants on cellular and subcellular levels. Superoxide radicals generated in plant cells are converted to H_2O_2 under the influence of superoxide dismutase (SOD). The accumulation of H_2O_2 is prevented in cell by catalase (CAT) or by ascorbate-glutathione cycle. In this cycle ascorbate peroxidase (APX) reduces it to H_2O. Thus, these compounds interrupt the cascades of uncontrolled processes of oxidation in plant organism (Noctor, Foyer, 1998).

Carotenoids react with free radicals directly, forming a carotenoid radical (Palozza, Krinsky, 1992). It could be regenerated by interaction with tocopherols and ascorbate in the lipid phase of the membrane (Edge, McGarvey, Truscott, 1997). Carotenoids of the xanthophyll cycle (violaxanthin and zeaxanthin) are closely involved in control of ROS production by chlorophylls when the photosynthetic electron chain is saturated (Foyer *et al*., 1994). It is shown that zeaxanthin is a very efficient ROS scavenger (Lim *et al*., 1992; Sielewiesiuk, Matula, Gruszecki, 1997).

It is well-known, that lipids are integral components of thylakoid membranes and are substantial for their photosynthetic activity. The plant thylakoid membranes contain mainly nonphosphorous glycolipids such as the nonbilayer lipid monogalactosyldiacylglycerol (MGDG) and the bilayer lipid digalactosyl-diacylglycerol (DGDG) (Webb, Green, 1991; Lee, 2000), which contribute to thylakoid aggregation and stacking (Menikh, Fragata, 1993; Hincha, 2003). Besides, there is an anionic sulpholipid sulfoquinovosyldiacylglycerol (SQDG) with a sulfonic acid and

derivative of glucose. About 50–60% of polar lipids in photosynthetic tissues are represented by MGDG and 20–25% by DGDG. The third glycolipid (SQDG) comprises between 8 and 24% of the four major chloroplast lipids and contains a substantial quantity of high melting point fatty acids (Kenrick, Bishop, 1986; Murata, Siegenthaler, 1998; Joyard *et al*., 1998). The glycosyl moiety of it is characterized by carbon being directly bonded to sulfur as $C\text{-}SO_3^-$. Sulfonic acid of this type is chemically stable and strong acid in wide pH range (Barber, Gounaris, 1986).

The study of the core peptide D1 showed that MGDG, PG and SQDG molecules are bound in it in the molar ratio 1:3:17. The isolated LHCP-complex contained in bound form three MGDG molecules, one molecule of DGDG, one molecule of PG and one molecule of lutein. Less than one molecule of SQDG, β-carotene, neoxanthin and violaxanthin are found in LHCP-complex. In contrast to the lipids of the thylakoid membrane, the lipids bounded with proteins/peptides are characterized by a strongly saturated character (Gasser *et al*., 1999). Besides, the major thylakoid lipid MGDG required for activity of violoxanthine de-epoxidase (VDE) located in the thylakoid lumen (Hager, Holocher, 1994; Siefermann, Yamamoto, 1975; Yamamoto, Higashi, 1978). It is four times more efficient in precipitating VDE compared to the DGDG, and up to 38 times more efficient than other thylakoid lipids (Rockholm, Yamamoto, 1996).

VDE is a 43-kD nucleus-encoded protein that is localized in the thylakoid lumen (Bugos, Yamamoto, 1996). Information exists that the isolated violaxanthin de-epoxidase (VDE) revealed that for optimal activity the enzyme requires the presence of the major thylakoid lipid, MGDG, as a cofactor (Webb, Green, 1991). It was found that purified violaxanthin was no a suitable substrate for VDE unless suspended with MGDG (Yamamoto et al., 1974), moreover, VDE itself requires a small amount of absorbed MGDG for activity (Yamamoto, Higashi, 1978). VDE and zeaxanthin epoxidase are considered to be the members of lipocalin family. They are the first lipocalins identified from plants and are unique in that they also have catalytic activity (Bugos, Hieber, Yamamoto, 1998).

From the other hand, the highest level of VDE-independent violaxanthin conversion was observed when the xanthophyll was incorporated into liposomes made only of phosphatidylglycerol or SQDG, and it amounted to about 30 and 17% of the initial concentration of violaxanthin, respectively. MGDG seems to be necessary for VDE activity not as galactolipid but as non-bilayer-prone lipid forming reversed hexagonal structures. Such structures, created by both MGDG and PE are effective in sustaining of VDE enzymatic activity (Latowski, Åkerlund,| Strzałka, 2004).

Deschampsia antarctica Desv. (*Poaceae*) is the only native *Gramineae* found in the Antarctic, where it is restricted to the Antarctic Peninsula and its offshore islands. It was found that *D. antarctica* have higher levels of SOD and APX activity compared with other plants. It was found also that the xanthophyll cycle is more operative in this plant. Also it was proposed that photochemical quenching and particularly the high level of antioxidants help *D. antarctica* to resist photoinhibitory conditions. The relatively high antioxidant capacity of *D. antarctica* may be a determinant for its survival in the severe Antarctic environment (Perez-Torres et al., 2004).

Taking into account that oxidative stress is a main effect of UV-B radiation we consider it to be expedient to study antioxidant indexes and glycolipid composition of *Deschampsia* plants caused by H_2O_2 action as well. Also we investigated the influence of exogenous H_2O_2 on physiological reactions of *Deschampsia antarctica*. In order to compare physiological reactions of *D. antarctica* plants we used also *Deschampsia caespitosa* plants, that are the typical habitants of Ukrainian Carpathian ecosystems.

Materials and Methods

D. antarctica plants were collected from some offshore Antarctical islands. Physiological and biochemical characteristics of samples of *Deschampsia antarctica* plants selected from different Antarctic coastal islands are investigated. With the purpose of studying of adaptive reactions of

Deschampsia antarctica plants they were introduced in climatic conditions of the European temperate region and their reactions on UV-B radiation and oxidative stress actions comparing with *Deschampsia caespitosa* plants was investigated.

The control plants were grown under the laboratory conditions for 10 days under conditions of daily lamps illumination, air temperature 8-10 ºC, 16-hour photoperiod and then the half of this plants were irradiated by UV-B for 20 hours in 5-times exposition (4h on light period). The UV-B lamp with absorption filter (TL 20Вт/12RS (Philips) 280-300 nm) was used for illumination of plants. The biologically effective UV-B radiation (UV-BBE) was 6.17 kJ m^{-2} d^{-1}. Distance to the source of illumination was 10 cm. Oxidative stress was induced by spraying plants with H_2O_2 (500 μM for 4 hours).

The pigment content of leaves was determined with generally accepted method (Arnon, 1949). The carotenoid composition was revealed using TLC method (Merzlyak, 1978). Polar lipids were isolated according to L. Zill and E.Harmon (Zill, Harmon, 1962) in modification of G.Yakovenko and A.Mihno (Yakovenko, Mihno, 1971). Glycolipids were separated with the help of TLC and then MGDG and DGDG were determined by densitometring of TLC plates against standards (Yamamoto, 1980). SQDG was determined according to E.Kean (Kean, 1968).

The functional state of photosynthetic apparatus (PSA) was evaluated with the help of the chlorophyll fluorescence induction method. Chlorophyll fluorescence in the plant leaves was measured by XE-PAM fluorometer ("Walz", Germany) at 20 ^{o}C. The data record in the Excel file format was performed by UT-60E multimeter ("Unitrend International Ltd", China) connected with a computer. The modulated light stream of impulsive xenon lamp was passed through the blue-green filter BG-39 (5 MM, "Schott", Germany) in order to excite chlorophyll fluorescence. Fluorescence was registered at wave-lengths ≥ 695 nm, using the filters RG645/R65 (2 mm, 1 mm) and RG9 (1 mm). Leaves adapted to darkness during 30 min.

Minimal fluorescence (F_0) of dark-adapted leaves was determined at low photosynthetic photon flux density (PPFD) in dose of 0,2 μmol (quantum) $m^{-2}s^{-1}$. Maximal fluorescence of dark- (F_m) and light-adapted (F_m') leaves was detected under conditions of saturating irradiance (1 s) by halogen lamps [5 000 μmol (quantum) $m^{-2}s^{-1}$]. The potential photochemical efficiency (F_v/F_m) was determined after 30 min of dark adaptation of plants.

Fluorescence induction parameters F_v'/F_m', qP, qN and ϕ_{PSII} (PSII were measured under conditions of 200 μmol (quantum) $m^{-2}s^{-1}$ of actinic light and calculated as described by van Kooten (van Kooten 1990). The dark adaptation of plants was continued during 30 min.

Separations of thylakoid membranes carried out in non-denaturing polyacrylamide gel with the aim of its disintegration and with subsequent determination of content of pigment-protein complexes (Anderson, 1980). Pigment-protein complex content was evaluated as related to chlorophyll defined according to D.Arnon [Arnon, 1949]. Electrophoregramms were scanned by Shimadzu densitometer and analyzed.

The replications of experiments were fourfold; authenticity (validity) of differences between the mean arithmetic values of indexes was set after the Student's criterion. Differences were considered as reliable at the value of $p \leq 0,05$.

Results and Discussion

Our investigations performed with *D. antarctica* plant samples delivered from Antarctic showed that UV-B radiation action caused decrease of chlorophylls *a*, *b* content and Chl a/b ratio (Tabl. 1). Carotenoid composition was characterized by *β*-carotene decrease accompanied by zeaxanthin+violaxanthin accumulation. Chlorophyll content changes in *D.caespitosa* plants were similar with *D. antarctica* plants. Carotenoid content was characterized by all carotenoids studied decrease (μmol per 1g DM expression), but fraction composition expressed in mol % was characterized by *β*-carotene decrease and neoxanthin accumulation (Tabl. 2). But information presented in literature showed that chlorophyll *a* and carotenoids content increased in soybean

(*Glycine max*) plants irradiated by UV-B at one of two cultivars studied (Middleton, Teramura, 1993). The total carotenoid content of mature vine leaves was found also to be less in vines grown under a UV screen (Steel, Keller, 2000).

Table 1

Lipid and pigment composition in leaves of *D. antarctica* and *D.caespitosa* plants under conditions UV-B radiation

Variant	Lipids, mol %			Pigments, mg per 1 g DM			
	D.antarctica						
	MGDG	DGDG	SQDG	Chl *a*	Chl *b*	Chl *a+b*	Chl *a/b*
control	69,71 ± 1,38	17,78 ± 0,88	12,51 ± 0,95	9,87 ± 0,91	2,91 ± 0,05	12,78 ±1,40	3,39
UV-B	61,02 ± 1,62	21,44 ± 0,85	17,54 ± 0,76	6,35 ± 0,43	2,02 ± 0,10	8,37 ± 0,14	3,14
	D.caespitosa						
control	64,13 ± 0,92	29,88 ± 1,76	5,99 ± 0,67	10,54 ± 0,06	3,37 ± 0,10	13,91 ± 0,16	3,13
UV-B	69,05 ± 2,73	23,71 ± 1,31	7,24 ± 0,33	7,6 ± 0,50	2,65 ± 0,25	10,26 ± 0,75	2,87

Table 2

Carotenoid composition in leaves of *D. antarctica* plants under conditions of UV-B radiation

Variant	*β*-carotene		lutein		zeaxanthin+ violaxanthin		neoxanthin	
	D. antarctica							
	µmol per 1g DM	mol %	µmol per 1g DM	mol %	µmol per 1g DM	mol %	µmol per 1g DM	mol %
control	1,5 ± 0,1	25,3 ± 0,5	2,0 ± 0,1	33,1 ± 0,7	1,4 ± 0,2	23,6 ± 0,6	1,1 ± 0,2	18,0 ± 0,4
UV-B	1,2 ± 0,1	21,7 ± 0,6	1,9 ± 0,1	33,7 ± 0,7	1,6 ± 0,2	27,1 ± 0,4	1,0 ± 0,1	17,5 ± 0,2
	D. caespitosa							
control	1,9 ± 0,1	20,5 ± 0,3	3,2 ± 0,2	34,3 ± 0,4	2,2 ± 0,1	23,7 ± 0,6	2,0 ± 0,1	21,5 ± 0,3
UV-B	1,0 ± 0,1	16,1 ± 0,2	2,2 ± 0,1	34,4 ± 0,6	1,5 ± 0,1	23,8 ± 0,3	1,7 ± 0,2	25,8 ± 0,6

The *β*-carotene is present in antennae (LHCs) and reaction center (RC) of PSII and plays a major role in photoprotection by quenching the triplet state primary donor ($3P_{680}$) or reducing the oxidized form (P_{680}^{+}) (Telfer, De Las Rivas, Barber, 1991). Molecules of *β*-carotene protect LHCs and RC against photooxidative damage through quenching of singlet oxygen ($^1O_2$) and/or triplet excited of chlorophyll (3Chl*) (Telfer et al., 1994). The mechanism of $^1O_2$-protection leads to destruction of *β*-carotene molecules (Barber, 1994) and to some extent, is obstacle in the chain the oxidative reactions in plant cells (Latowski, Kostecka-Gugala, Strzalka, 2003).

Lipid composition was characterized by accumulation of DGDG and SQDG in leaves of *D.antarctica* plants and accumulation of MGDG and SQDG in leaves of *D.caespitosa* plants under conditions of UV-B radiation (Tabl. 3).

H_2O_2 treatment cause unreliable decrease of SOD activity of both species. Pigment composition was characterized by increase of carotenoids content in leaves of *D.antarctica* plants and chlorophyll *a* content in both species (Table 3).

Glycolipid content was stable and SQDG content slightly increased in leaves of *D.antarctica* plants after H_2O_2 treatment. In the same conditions only MGDG content decreased in leaves of other *Deschampsia* species (Table 3).

Thus, both UV-B radiation and H_2O_2 treatment caused different changes in *D.antarctica* pigment content. Galactolipid changes were insignificant in leaves of *D.antarctica* plants, but slight increase of SQDG content in leaves of this species took place. MGDG content decrease was more meaningful in leaves of *D. caespitosa* plants (Tabl. 1 and 3).

Table 3.

The influence of H_2O_2 on lipid and pigment composition in leaves of *D.antarctica* and *D.caespitosa* plants

Variant	Lipids mol %			Pigments, mg per 1 g DM					SOD activity
	D.antarctica								
	MGDG	DGDG	SQDG	Chl a	Chl b	Chl a+b	Chl a/b	Car	cond. units
control	55,1 ± 0,3	37,1±0,2	7,8±0,1	9,1±0,3	4,0 ± 0,1	13,1 ± 0,3	2,27	2,37 ± 0,1	31,5 ± 0,6
H_2O_2	54,3 ± 2,2	35,7±0,3	9,9±0,2	11,7±0,4	4,3 ± 0,1	16,0 ± 0,4	2,71	3,25 ± 0,2	28,0 ± 0,9
	D.caespitosa								
control	54,0 ± 1,1	34,7±0,2	11,3±0,4	8,9±0,3	3,2 ± 0,1	12,1 ± 0,4	2,77	2,4 ± 0,1	25,5 ± 0,7
H_2O_2	48,5 ± 0,6	40,5±0,4	11,0±0,5	11,3±0,2	4,5 ± 0,3	15,4 ± 0,3	2,79	2,1 ± 0,2	22,3 ± 0,5

Our data agree partly with results of Musil C.F. with colleagues (Musil, Chimphango, Dakora, 2007). According to their results content of chlorophyll a, b and carotenoids was variable in conditions of UV-B radiation depending on species of plants. The main trend has been the decrease of chlorophyll content in leaves under conditions of UV-B radiation, but some plants accumulate it, e.g. *Leucadendron laureolum* (chlorophyll ***a*** – by 17,8%, chlorophyll ***b*** – by 101,9%) and *Phylica pubescens* (chlorophyll ***a*** – by 27,2%, chlorophyll ***b*** – by 30,8%**,** carotenoids – by 18,9%).

There are few known data, which prove that oxidative processes was induced by high concentration of ozone. In addition, was not proved, that these processes induce the decrease of pigments and polar lipids content (mainly MGDG with some DGDG). It was accompanied by an increase of TAG and DAG (Sakaki, 1998) with stable anionic lipid (SQDG and phosphatidylinositol) content for the period of ozone exposure (in spinach leaves, at least). Similar lipid changes were also observed in several plant species, and in broad bean leaves, a relative increase in SQDG took place. Because both galactolipids were significantly destroyed during ozone exposure, the SQDG content expressed as mol % of the total glycolipids increased up to 45 mol % (depending upon species) (Sakaki *et al*., 1985, 1994). Considering these changes it is worth to mention that SQDG molecules in photosynthetic tissues stabilize CF_0-CF_1 ATPase, protect and stabilize D1/D2 dimers and LHCII (Livn, Racker, 1969; Pick *et al*., 1985). SQDG and the Rieske protein interaction in the cyt b_6f structures is also very important (De Vitry *et al*., 2004). Thus, SQDG seems to be involved in the turnover of cyt *f* in a similar manner like D_1 and raise the question of whether a similar mechanism underlies the role of SQDG in the assembly of both subunits (De Vitry *et al*., 2004).

A significant increasing of total lipid concentration and decreasing of free sterols (campesterol, cholesterol, sitosterol and stigmasterol) were registered in tobacco plants exposed to ozone. (Trevathan, Moore, Orcutt, 1979). Ozone induced the decrease of free sterols (FS) and increase of sterol glycoside (SG) and acylsterol glycoside (ASG) in bean leaves (Tomlinson, Rich 1971; 1973; Spotts, Lukezic, Lacasse, 1975; Trevathan, Moore, Orcutt, 1979; Whitaker, Lee, Rowland, 1990). Therefore it seems that ozone stimulates glycosylation and further acylation of sterols under severe stress. Thus, ozone enhances production of free fatty acids (FFA) from galactolipids; acylation of SG might play a scavenging role of FFA in leaf cells together with the synthesis of TG. In support, main fatty acid (FA) species increased in ASG include 18:3, a predominant FA in galactolipids (Tomlinson, Rich, 1973). Results obtained in experiments with spinach (*Spinacia oleracea* L., cv New Asia) plants treated with ozone also showed a large reduction of galactolipids accompanied by TG increase without a corresponding effect on leaf FFAs (Sakaki et al., 1985). Constituent FAs of galactolipids, especially MGDG, were largely converted to those of TG. Authors proposed that 1,2-DG liberated from MGDG is the direct precursor of TG synthesized in ozone-fumigated spinach leaves, based on the fact that 16:3, the

fatty acid specific to MGDG, was recovered in 1,2-DG as well as in TG. Molecular species and FA distribution of TG accumulated in spinach (*Spinacia oleracea* L.) leaves treated with ozone were compared with those of MGDG. Analysis of positional distribution of the fatty acids in MGDG and the accumulated TG by the enzymatic digestion method showed that hexadecatrienoate (16:3) was restricted to *sn*-2 position of the glycerol backbone in both MGDG and TG, whereas a-linolenate (18:3) was preferentially located at *sn*-1 position in MGDG, and *sn*-1 and/or *sn*-3 positions in TG, suggesting that 1,2-diacylglycerol moieties of MGDG are the direct precursor of TG in ozone fumigated leaves. Further analysis showed that TG increased with ozone fumigation consisted of approximately an equal molar ratio of *sn*-1,3-18:3-2-16:3 and *sn*-1,2,3-18:3. Because the molecular species of MGDG in spinach leaves is composed of a similar molar ratio of *sn*-1-18:3-2-16:3 and *sn*-1,2-18:3, it was concluded that MGDG was converted to 1,2-diacylglycerol and acylated with 18:3 to TG in ozone-fumigated spinach leaves (Sakaki *et al*.,1990). The similar results were represented in later work (Sakaki, Tanaka, Yamada, 1994). Analysis of eight species of leaf lipids after treatment with ozone revealed MGDG content decrease and TG accumulation in all plants, but the extent of the changes varied among the plant species. The FAs esterified to TG were mainly α-linolenic acid (18:3) in 18:3 plants and hexadecatrienoic acid (16:3) and 18:3 in 16:3 plants normally esterified to MGDG in the respective plant groups. Therefore MGDG seems to have been metabolized to TG via FFA and DG in all tested plants in response to ozone.

Chlorophyll fluorescence indexes in leaves of plants of two *Deschampsia species* were measured at actinic light of 200 jmol (quantum) $m^{-2}s^{-1}$.

The maximum energy conversion efficiency or quantum efficiency of PSII charge separation (F_v/F_m) is one of the main characteristics of PS II complexes. This parameter is used for estimation of PS II state in adapted leaves, when quinone acceptors Q_A are fully oxidized. UV-B radiation induced the decrease of maximum (F_v/F_m) and actual (F_v'/F_m') quantum efficiency of PSII photochemistry on 50% and 60% (Table 1), herewith these indexes were unchanged for *D. caespitosa* plants under conditions of UV-B radiation. The level of maximum fluorescence (F_m) was significantly decreased in UV-B irradiated plants of *D.antarctica*, but slightly increased in leaves of *D.caespitosa* plants. These data can indicate on quenching state of the light harvesting complex of PSII.

The important parameter of the functional state of photosynthetic apparatus is index *qP*, that characterizes the level of Q_A pool oxidation, the ability of this pool to accept electrons from previous components of electron transport chain.

Photochemical quenching (*qP*) in relation to a proportion of open PSII centers were decreased by 12% in leaves of *D.antarctica* plants and increased on 5 % in leaves of *D.caespitosa* plants under conditions of UV-B radiation (Table 4).

The coefficient of non-photochemical quenching *qN* characterizes the heat energy dissipation of plants in the reaction centers.

Non-photochemical quenching (*qN*) associated with thermal energy dissipation in the antenna system was unchanged in UV-B irradiated plants of *D.antarctica*, but decreased on 14 % in leaves of *D. caespitosa* plants. Through the mechanism of non-photochemical quenching, PSII is protected from photodestruction. The process of non-photochemical quenching (thermal energy dissipation in the antenna system) is a main mechanism of regulation of functional size of PSII antenna. This process protects photosynthetic apparatus from photooxidative damage (Štroch et al., 2004). In leaves of *D.antarctica* plants, grown under conditions of UV-B radiation, *qN* index was unchanged at actinic light of 200 μmol(quantum) m^{-2} s^{-1}. There were noted the decrease of electron transport and stable level of photon energy dissipation in leaves of UV-B irradiated plants of *D. antarctica*. Slightly decrease of *qN* observed in this conditions for *D. caespitosa* plants (Table 4).

To determine the real quantum yield of linear electron transport through PSII the parameter ϕ_{PSII} was used (Genty et al., 1989). Effective PS II quantum efficiency of charge separation (φ_p) is used for estimation of maximal efficiency of light photochemical reactions, when the part of quinone acceptors Q_A is reduced. The heat energy dissipation increase with decreasing of (φ_p), because these processes are parallel, competitive and interdependent. It was found, that *D. antarctica* plants, grown under conditions of UV-B radiation were characterized by decrease of ϕ_{PSII} (on 63%) in dose of 200 μmol (quantum) $m^{-2}s^{-1}$. However, this index was not changed under the same conditions for *D. caespitosa* plants.

The intensive decrease of potential (F_v/F_m) and actual (F_v'/F_m') quantum efficiency of PSII photochemistry of leaves of *D. antarctica* plants under conditions of UV-B radiation induces the decline of photosynthetic function. At the same time, both these indexes were unchanged under conditions of UV-B affection that can be regarded as adaptive mechanism on PS II level (Table 4).

Photochemical quenching (*qP*) depends on both factors: the inflow of electrons to Q_A and their outflow to PQ pool. Lower *qP*, observed in leaves of UV-B irradiated plants of D. *antarctica*, can be associated with more slow PQ pool oxidation, which is a result of decrease of electron transport to PSI. Increasing of *qP* in leaves of *D.caespitosa* plant can be the evidence of increasing of electron transport rate to PS II in radiation conditions (Table 4).

Table 4.

Chlorophyll fluorescence parameters of two *Deschampsia* species under UV-B radiation

Variant	F_v/F_m	F'_v/F'_m	*qP*	*qN*	φ_p
	D. antarctica				
Control	0,772±0,038	0,682±0,044	0,895±0,013	0,356±0,055	0,612 ± 0,046
UV-B	0,385±0,063	0,281±0,056	0,789±0,087	0,385±0,027	0,226 ± 0,061
	D. caespitosa				
Control	0,758±0,019	0,583±0,028	0,849±0,007	0,556±0,026	0,495 ± 0,027
UV-B	0,732±0,045	0,586±0,052	0,896±0,010	0,479±0,026	0,525 ± 0,049

The data prove that the part of UV-B in the general light flow induced the insignificant changes in photosinthetic apparatus in leaves of *D. caespitosa*.

Under conditions of UV-B radiation the quotum of light energy, absorbed by photosynthetic pigments of *D. antarctica* leaves, is increased, but this energy, probably, cannot be used for CO_2 assimilation. The essential quota of this energy is consumed on heat dissipation. The efficiency of using of quantum energy was higher for *D. caespitosa* plants.

Simultaneously, lower electron transport to Q_A is associated with high *qN* (high dissipation in LHCII) in leaves of UV-B irradiated plants of *D.antarctica*. The stability of *qN* indexes for leaves of *D.caespitosa* plants also can be considered as adaptive mechanisms. In addition, the real quantum yield of linear electron transport through PSII, ϕ_{PSII}, was lower in leaves of UV-B irradiated plants of *D. antarctica*, that was associated with lower *qP* of photosynthetic apparatus of these leaves (Table 4). The stability of ϕ_{PSII} and *qP* indexes of *D.caespitosa* plants under conditions of UV-B radiation can indicate resistance to UV-B affection. It could confirm in a large measure that photosynthetic apparatus of *D. caespitosa* is more adapted to UV-B radiation. It is worth to pay attention, because D. *caespitosa* is a natural inhabitant (dweller) of the moderate latitude of Europe, Siberia and Caucasus while *D. antarctica* is the endemic type of Antarctic Continent.

Close relationship between *qN* index and Zea content in various plants and under different conditions of cultivation was shown nowadays (Verhoeven et al., 1999; Havaux, Kloppstech, 2001; Behera, Choudhury, 2003; Müller-Moule et al., 2003). In previous works was noted, that reaction of high-energy-state quenching was not accompanied by Zea synthesis, but was associated with a reversible inactivation of PSII RC fraction (Finazzi et al., 2004)

One of the main ways of excess energy dissipation is conformational rearrangement of the main light-harvesting complex - LHCII, which forms the molecular basis for non-photochemical quenching of chlorophyll fluorescence. The transition into a state of dissipation is possible due to LHCII-bound carotenoid neoxanthin, by transferred the energy from chlorophyll *a* to a low-lying carotenoid excited state, identified as one of the two luteins (lutein 1) in LHCII. (Ruban et al., 2007). Neoxanthin accumulation in irradiated plants of *D. caespitosa* could testify to its involvement in the process of *qE* and LHCII transition in the state of dissipation, in contrast to *D. antarctica* plants, where the xanthophyll contents was stable (Table 3).

The function of carotenoids and fluorescence parameters can be understood within the framework of their binding to light-harvesting complex (Lhc) proteins. The analisys of Lhc of two plant species of *Deschampsia* identified of six green electrophoretic bands, which belong of pigment-protein complexes, according to the Anderson nomenclature:

CP1a – complex of reaction centre (RC) of PSI, which partially retains its own LHCI;
CP1 – complex of RC PSI, without LHCI;
LHCP1 – monomeric form LHCII;
CPa – LHC nearest to RC PSII;
LHCP3 – oligomeric form LHCII;
FP – free chlorophyll

The tendency to accumulate of pigment-protein complexes CP1a + CP1 in irradiated *D. antarctica* plants was observed. In contrast, destruction of pigment-protein complexes Cpa+CP1 was noticed in treated *D. caespitosa* plants compared with control variant (Table 5).

Total LHCII (LHCP1 + LHCP3) did not changed in control and irradiated *D. antarctica* plants whereas the increase in treated *D. caespitosa* plants took place thanking to accumulation of LHCP1. As result we observed the ratio increase almost twice between the monomeric and oligomeric forms LHCII (LHCP1/LHCP3) in control and irradiated plants of *D. caespitosa* (Table 5). Concerning carotenoid composition we could notice *β*-carotene destruction in both species and slight violaxanthin enlargement in *D.antarctica* plants under conditions of UV-B radiation.

Table 5

The thylakoid membrane pigment-protein complex content and ratio (% of the total chlorophyll quantity)

Pigment-protein complexes and their relationship	*Deschampsia antarctica*		*Deschampsia caespitosa*	
	Control	UV	control	UV
CP1a + CP1	15,10±0,41	19,30±0,23	19,16±0,36	16,98±1,47
LHCP1	11,10±0,69	11,89±0,34	15,67±0,43	22,66±0,99
CPa	8,53±0,31	5,67±0,17	8,96±0,18	7,25±0,16
LHCP3	28,11±0,21	27,43±0,22	24,59±0,39	22,23±0,26
FP	37,16±1,18	35,71±0,78	31,64±0,51	30,89±0,06
LHCP1/LHCP3	0,40±0,03	0,43±0,06	0,64±0,03	1,02±0,03
LHCP1 + LHCP3	39,22±0,48	39,32±0,25	40,25±0,03	44,89±1,25

Thus, our results correspond mainly to data available in literature and we could conclude, that UV-B and oxidative stress induced similar changes. Besides, *D.caespitosa* plants revealed MGDG destruction more hard, than *D. antarctica* .

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