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CHLOROPHYLL FLUORESCENCE OF UV-B IRRADIATED BEAN LEAVES SUBJECTED TO CHILLING IN LIGHT

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Introduction

The effect of UV-B radiation on plants can be modified by such environmental factors as non-optimal temperature (low or high) and light. Bean belongs to chilling-sensitive plants, i.e. susceptible to chilling temperature (below 10°C, but above the freezing point), as well as to UV-B-susceptible species [BORNMAN, TERAMURA 1993; TERAMURA, SULLIVAN 1994]. The photosynthetic apparatus is the most sensitive target for both stress factors [CEN, BORNMAN 1990]. The aim of this research was to study the effect of chilling in the presence of light on the primary photosynthesis reaction using chlorophyll *a* fluorescence detection of UV-B irradiated bean plants. This method is used in the studies on plant reaction to both stresses studied in this paper [LICHTENTHALER et al. 1986; ÖQUIST et al. 1987; KRAUSE, WEIS 1991; SCHREIBER et al. 1994; SKÓRSKA 1996, 1999; MURKOWSKI, SKÓRSKA 1997].

Material and methods

Seeds of bean (*Phaseolus vulgaris* L. cv. Aura) were watered in Petri dishes with distilled water, and after germination were transferred into pots with sand. The pots were placed on the rotary stage in two special chambers [SKÓRSKA 1999; 2000] in the following controlled conditions: temperature 22°C/18°C (day/night), air humidity 40%, LRF 400 lamps (PAR[1] 150 $\mu mol \cdot m^{-2} \cdot s^{-1}$), and photoperiod 12 h. The plants were watered daily with 50% Hoagland nutrient. One part of two-week old plants was subjected to UV-B irradiation ($UV\text{-}B_{BE}$ = 1.86 $kJ \cdot m^{-2} \cdot d^{-1}$, 3 days) with the lamp type VL-115 M [SKÓRSKA 2000], whilst the second part – the control plants – was grown for the same duration of time without UV-B. The applied UV-B dose was higher by 50% than that from the natural solar radiation. Thereafter, from the first leaves, sample discs (15 mm of diameter) were cut out and treated for two hours with chill (2°C) in increased PAR intensity (600 $\mu mol \cdot m^{-2} \cdot s^{-1}$). After the chilling, the samples were dark-adapted for 15 min and the measurements were done for the parameters of chlorophyll fluorescence F_v/F_m, F_v/F_o and Rfd, using Plant Efficiency Analyser (PEA Hansatech, England),

[1] PAR – Photosynthetically active radiation

according to LICHTENTHALER et al. [1986]. Parameters F_v/F_m and F_v/F_o are proportional to maximal photochemical efficiency, and to the activity of the donor side, respectively Rfd - vitality index; F_o, F_m, F_v denote respectively intensity of initial, maximal and variable fluorescence ($F_v=F_m-F_o$). After two weeks of further growth, in the stage of the third leaf couples, the plants were subjected to UV-B radiation ($UV\text{-}B_{BE}$ = 1.86 $kJ \cdot m^{-2} \cdot d^{-1}$, 6 days). Then, the discs were cut out from the third leaves, treated as previously, and similar measurements were done. The results are presented as means from 6 independent biological replications. Two-way analysis of variance was used, and the Duncan test – for separation of the groups, marked with the same letters, at the significance level of $p < 0.05$.

Results and discussion

The measured parameters of chlorophyll fluorescence were lower in UV-B irradiated bean leaves in comparison with the control (Table 1), especially in the first leaves. The third leaves seemed more tolerant to the applied dose of UV-B

Table 1; Tabela 1

Chlorophyll fluorescence parameters of the control and UV-B irradiated bean leaves after 2 hours at 20°C or at chilling temperature (2°C) in light

Parametry fluorescencji chlorofilu kontrolnych i napromieniowanych UV-B liści fasoli po 2 godzinach w temperaturze 20°C lub w 2°C na świetle

Variant Wariant	F_v/F_m		F_v/F_o		Rfd	
	control kontrola	UV-B	control kontrola	UV-B	control kontrola	UV-B
1st leaves ($UV\text{-}B_{BE}$=1.86 $kJ \cdot m^{-2} \cdot d^{-1}$, 3 days); pierwsze liście ($UV\text{-}B_{BE}$=1,86 $kJ \cdot m^{-2} \cdot d^{-1}$, 3 dni)						
20°C	0.80[a]	0.76[b]	4.1[a]	3.2[b]	2.0[a]	1.6[b]
2°C	0.60[c]	0.52[d]	1.6[c]	1.0[d]	0.8[c]	0.5[d]
Effect significance: Istotność efektu:						
Chilling; Chłód	***		***		***	
UV-B	***		***		**	
Chilling × UV-B Chłód × UV-B	n.s.		n.s.		n.s.	
3rd leaves ($UV\text{-}B_{BE}$=1.86 $kJ \cdot m^{-2} \cdot d^{-1}$, 6 days); trzecie liście ($UV\text{-}B_{BE}$=1,86 $kJ \cdot m^{-2} \cdot d^{-1}$, 6 dni)						
20°C	0.81[a]	0.80[a]	4.4[a]	3.9[b]	2.1[a]	2.2[a]
2°C	0.59[b]	0.53[c]	1.4[c]	1.1[d]	0.8[b]	0.3[c]
Effect significance: Istotność efektu:						
Chilling; Chłód	***		***		***	
UV-B	***		***		*	
Chilling × UV-B Chłód × UV-B	n.s.		n.s.		***	

abcd – the means denoted by the same letter (for one parameter) do not differ significantly ($p<0.05$); średnie oznaczone taką samą literą (dla jednego parametru) nie różnią się istotnie ($p<0.05$)

effect significant at the level *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$; istotność efektu na poziomie *$p < 0.05$, **$p<0.01$, ***$p<0.001$

n.s. – not significant; nieistotna

radiation, particularly as it was twice higher than previously – in the earlier phase of the plant growth. This may have resulted from the plant hardening to UV-B radiation, similarly as in the case of the effect of the other environmental stresses [STARCK et al. 1995].

As a result of chilling, further decrease in the measured parameters was observed in a similar degree for both groups, which indicates an inhibition of the photosynthesis light reactions in PS II. The value of the Rfd parameter decreased the most, showing disturbance in co-operation between the light and dark enzymatic reactions of photosynthesis [LICHTENTHALER et al. 1986].

Taking into consideration the results of similar experiments conducted on cucumber plants [SKÓRSKA 1999], one can assert that the studied bean plants showed greater tolerance both to UV-B radiation and the chilling stress.

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Key words: bean, chill, chlorophyll fluorescence, *Phaseolus vulgaris* L., ultraviolet

Summary

The plants of bean (*Phaseolus vulgaris* L. cv. Aura) grown in controlled conditions (PAR 150 μmol·m^{-2}·s^{-1}, air humidity 50%, temperature 22°C/18°C, day/night, photoperiod 12 h) were subjected to UV-B irradiation (UV-B_{BE} = 1.86 kJ·m^{-2}·d^{-1}). Then, sample discs were cut out from the leaves and treated for two hours with chill (2°C) in high light (PAR 600 μmol·m^{-2}·s^{-1}). The chilling induced a decrease in the measured chlorophyll fluorescence parameters, particularly in the UV-B irradiated plants, indicating disturbances of the primary photochemical reactions of photosynthesis. The greatest changes were observed in the Rfd parameter, determining co-operation between the light and dark enzymatic photosynthetic reactions.

FLUORESCENCJA CHLOROFILU LIŚCI FASOLI NAPROMIENIOWANYCH UV-B I PODDANYCH DZIAŁANIU CHŁODU NA ŚWIETLE

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Słowa kluczowe: chłód, *Phaseolus vulgaris* L., fasola, fluorescencja chlorofilu, ultrafiolet, UV-B

Streszczenie

Rośliny fasoli (*Phaseolus vulgaris* L. odmiany Aura) rosnące w kontrolowanych warunkach światła i temperatury (PAR 150 μmol·m^{-2}·s^{-1}, fotoperiod 12 h, 22°C/18°C, dzień/noc, wilgotność 50%) poddano napromieniowaniu UV-B_{BE} = 1,86 kJ·m^{-2}·d^{-1}). Następnie z liści wycięto krążki, które przez 2 godziny poddano działaniu obniżonej temperatury (2°C) przy zwiększonym poziomie PAR 600 μmol·m^{-2}·s^{-1}. Parametry fluorescencji chlorofilu F_v/F_m, F_v/F_o i Rfd, mierzone przy użyciu fluorymetru PEA Hansatech, uległy zmniejszeniu zarówno pod wpływem UV-B, jak i po ochłodzeniu, wskazując na inhibicję reakcji świetlnych fotosyntezy. Największym zmianom uległ parametr Rfd, określający współdziałanie między reakcjami świetlnymi i ciemniowymi reakcjami enzymatycznymi fotosyntezy.

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