BIOLOGIA PLANTARUM 51 (1): 189-192, 2007

BRIEF COMMUNICATION

# Influence of UV-B radiation on young triticale plants with different wax cover

E. SKÓRSKA* and W. SZWARC

*Department of Physics, Agricultural University of Szczecin, Papieża Pawła VI 3, PL-71459 Szczecin, Poland*

## Abstract

Ultraviolet-B radiation (biologically effective dose 2.6 kJ $m^{-2}$ $d^{-1}$) had negative influence on morphology and physiology of the young triticale plants. Plants exposed to UV-B were of lower height than control plants, their leaves were narrow, and the rate of net photosynthesis was decreased. The line RAH 336, which wax cover is lesser than that of traditional cultivar Magnat, was more susceptible to UV-B radiation, considering primary photosynthesis reactions, recorded by chlorophyll *a* fluorescence. An activation of protective mechanisms was observed: plants responded to UV-B by an increase of the content of UV-B absorbing compounds, and changes of antioxidant enzyme activities.

*Additional key words*: antioxidant enzymes, catalase, chlorophyll fluorescence, net photosynthetic rate, peroxidase, *Triticosecale*.

In natural environment plants are subjected to stress factors, among which UV-B radiation (280 - 320 nm) plays a considerable role, because more and more UV-B reaches the Earth's surface due to a depletion of stratospheric ozone (Deckmyn *et al.* 1994, Caldwell *et al.* 1998). UV-B radiation might have harmful impact on growth and metabolism of plants, causing a decrease of leaf area, plant biomass, chlorophyll content, inhibition of photosynthesis (Jordan 1996, Rajendiran and Ramanujam 2004). However, a lower area of leaf surface exposed to UV-B radiation can also mean one of protection measures. Diversity of mechanisms protecting against UV-B radiation depends on plant species and cultivars (Caldwell *et al.* 1998). One of them is an increased synthesis of flavonoids and phenylpropanoids in the leaf epidermis, which absorbs radiation of this range and can act as antioxidant (Caldwell *et al.* 1994). Among the anatomical changes of UV-B irradiated plants is an increased production of wax layer on leaves (Steinmüller and Tevini 1986, Gonzales *et al.* 1996, Pilon *et al.* 1999, Holmes and Keiller 2002). According to Holmes and Keiller (2002) the damaging radiation is reflected by the wax surface, hence it reaches deeper epidermis layer to a lower extent. It was shown that alpine species of *Poa* plants which grew under higher dose of UV-B radiation had inherently higher wax concentration than other *Poa* species (Pilon *et al.* 1999). Kinnunen *et al.* (2001) showed a negative correlation between the amount of waxes and UV-absorbing compounds in Scots pine needles. They found higher amount of UV-absorbing compounds at early and late stages of plant growth, when the amount of waxes was low or if epicuticular waxes were undeveloped. Cereals belong to species relatively tolerant to UV-B radiation in comparison with other crops (Caldwell *et al.* 1998). Steinmüller and Tevini (1986) showed that amount of cuticular waxes in barley leaves was five times greater than in bean or cucumber leaves. The purpose of this work was comparison of the influence of UV-B radiation on a traditional triticale cultivar and a line genetically devoid of wax. Different responses to UV-B, *e.g.*, in the content of flavonoids, and anatomical, biochemical or physiological characteristics were expected to be found.

As test plants the triticale (× *Triticosecale* Wittmack) traditional cv. Magnat and line RAH 336 which was almost lacking a wax layer (Maciorowski and Salak-

---

*Received* 9 September 2005, *accepted* 11 January 2006.

*Abbreviations*: Chl - chlorophyll; E - transpiration rate; $F_0$, $F_m$, $F_v$ - initial, maximum, and variable fluorescence ($F_v = F_m - F_0$); PAR - photosynthetically active radiation (400 - 700 nm); $P_N$ - net photosynthesis rate; PPFD - photosynthetic photon flux density; PS 2 - photosystem 2; Rfd - vitality index; UV-B - ultraviolet B radiation in the range of 280-320 nm; UV-$B_{BE}$ - biologically effective dose of UV-B radiation; $\Phi_{PS2}$ - quantum yield of PS 2.

*Acknowledgements*: The authors wish to thank Dr. R. Warzecha from the Institute of Plant Breeding and Acclimatisation in Radzików for seeds of the RAH 336 line.

* Corresponding author; fax: (+48) 91 4250391, e-mail: skorska@agro.ar.szczecin.pl

Warzecha 2002) were used. The plants were cultivated on 50 % Hoagland's solution in growth chambers, as described by Skórska (2000a). The photosynthetic photon flux density (PPFD) was 100 µmol m$^{-2}$ s$^{-1}$, 12-h photoperiod, and day/night temperature 17/15 °C. One chamber was equipped with *VL-115M* lamps (*Vilber Lourmat*, Marne-la-Vallée, France) as a source of UV-B radiation. The plants were exposed to UV-B radiation (0.65 W m$^{-2}$, 3 h daily) to give biologically effective radiation UV-B$_{BE}$ of 2.6 kJ m$^{-2}$ d$^{-1}$ calculated according to Caldwell (1971). The plants grown in the other chamber (without UV-B radiation) were the control ones. Measurements were done after 14-d treatment. The growth stage of the tested plants was at the first tiller detectable, one leader and one offshoot. Chlorophyll fluorescence was recorded by a portable fluorometer *PAM-200* (*Heinz Walz*, Effeltrich, Germany); the following parameters were determined: $F_v/F_0$, $\Phi_{PS2}$ (Schreiber *et al.* 1994) and Rfd (Lichtenthaler *et al.* 1986). Total chlorophyll (Chl) content was measured by the portable chlorophyll meter *SPAD 502* (*Minolta*, Osaka, Japan). The net photosynthetic ($P_N$) and transpiration (E) rates were measured by the gas-analyser *LCA-4* (*ADC BioScientific*, Hoddesdon, UK). The content of UV-absorbing pigments, mainly flavonoids, was analysed according to Caldwell *et al.* (1994) as the absorbance of leaf extracts at 305 nm. The catalase and peroxidase activity was determined spectrophotometrically (*Specord UV/VIS M42, Zeiss*, Jena, Germany) as described earlier (Skórska 2000a). The height of the plants and width of the first leaves were measured and then leaves of 20 plants were dried at 105 ºC and weighed. The two-way *ANOVA* and Tukey test were applied to data analysis.

One of the primary targets of UV-B radiation is an oxygen evolving complex in photosystem 2 (Renger *et al.* 1989, Jordan 1996), which is related to chlorophyll *a* fluorescence parameter $F_v/F_0$ (Schreiber *et al.* 1994). In our experiment this parameter was the same in the control plants of both lines, but it decreased in leaves of the RAH 336 line exposed to UV-B radiation (Table 1), which indicated a damage of this complex. Similar decrease was observed in sensitive species such as rapeseed, cucumber, pea and oat (Skórska 2000a,b, Skórska and Lewandowski 2003), but was not changed in triticale cv. Moreno (Skórska 2000b). The quantum efficiency of photosystem 2, $\Phi_{PS2}$, also was lower only in the UV-B treated RAH 336 plant. Decrease in this parameter was observed also in irradiated oat plants (Skórska 1999, Skórska and Lewandowski 2003). The plants of RAH 366 line exposed to UV-B radiation had a decreased value of Rfd parameter, named by Lichtenthaler *et al.* (1986) as the vitality index informing about an interaction between primary photosynthetic reactions and dark reactions leading to $CO_2$ assimilation. Decrease of this parameter is often related to photoinhibition induced by high irradiance or other stresses (Murkowski and Skórska 1997).

Chl content was almost constant in the UV-B irradiated plants. No difference of chlorophyll content was also observed in two grass species despite higher doses of UV-B radiation of 4.0, 5.3 or 6.5 kJ m$^{-2}$ d$^{-1}$ (Gwynn-Jones *et al.* 1996) as compared to our experiments. In our experiment the level of PAR was low compared to field conditions. This could significantly influence the $P_N$, because the high PPFD attenuates the negative consequences of UV-B and many other stress factors (Caldwell *et al.* 1998). The decrease of $P_N$ due to UV-B was more visible in the RAH 336 plants, though the increase of E was almost the same. These effects were also observed in irradiated plants of pea and triticale (Skórska 2000b,c). Deckmyn *et al.* (1994) observed a decrease of $P_N$ in UV-B irradiated plants of bean, and the most distinct effect was observed in plants, which were

Table 1. Characteristics of triticale plants grown in absence (control) or presence (UV-B) of UV-B radiation (UV-B$_{BE}$ = 2.6 kJ m$^{-2}$ d$^{-1}$, 14 d). Means of 20 (height, width, Chl content, dry mass), 9 ($P_N$, E), 6 (fluorescence), 5 (flavonoids, catalase) and 4 (peroxidase) replications ± SD. Means (in the row) marked by the same letters do not differ significantly at $P < 0.05$ (Tukey test).

| Parameters | cv. Magnat | | line RAH 336 | |
|---|---|---|---|---|
| | control | UV-B | control | UV-B |
| $F_v/F_o$ – oxygen evolving efficiency | 3.03 ± 0.29a | 2.97 ± 0.33a | 2.95 ± 0.31a | 2.47 ± 0.14b |
| $\Phi_{PS2}$ – quantum yield of PS 2 | 0.63 ± 0.02a | 0.63 ± 0.02a | 0.65 ± 0.02a | 0.54 ± 0.07b |
| Rfd – vitality index | 2.51 ± 0.34a | 2.32 ± 0.41a | 2.21 ± 0.25a | 1.58 ± 0.24b |
| Chlorophyll content [SPAD values] | 34.60 ± 5.20a | 32.80 ± 3.40ab | 30.70 ± 4.80b | 30.50 ± 2.80b |
| Net photosynthetic rate [µmol m$^{-2}$ s$^{-1}$] | 2.33 ± 0.50a | 1.77 ± 0.19bc | 2.26 ± 0.32ab | 1.45 ± 0.55c |
| Transpiration rate [mmol m$^{-2}$ s$^{-1}$] | 0.40 ± 0.17b | 0.98 ± 0.24a | 0.44 ± 0.16b | 1.03 ± 0.40a |
| Flavonoids content [A$_{305}$ g$^{-1}$] | 106.0 ± 45.0b | 315.0 ± 121a | 130.0 ± 53.0b | 346.0 ± 114a |
| Peroxidase activity [U g$^{-1}$] | 182.0 ± 25.0c | 197.0 ± 19.0c | 316.0 ± 36.0b | 431.0 ± 87.0a |
| Catalase activity [kU g$^{-1}$] | 8.6 ± 1.20a | 7.50 ± 1.50ab | 6.80 ± 0.70bc | 5.50 ± 0.40c |
| Plant height [mm] | 200.0 ± 17.0c | 183.0 ± 18.0d | 250.0 ± 32.0a | 217.0 ± 22.0b |
| Leaf width [mm] | 4.31 ± 0.47a | 4.00 ± 0.41b | 4.17 ± 0.48ab | 3.99 ± 0.49b |
| Leaf dry mass [mg]* | 29.30 | 35.50 | 26.20 | 28.90 |

grown under the lowest level of PAR. There are also reports which show that UV-B has no impact on $P_N$. There was no difference of $P_N$ between the irradiated and control plants of tomato, moreover increased $P_N$ under the influence of UV-B occurred in rape (Hao *et al.* 2000).

Triticale responded to UV-B by increasing production of flavonoids, which is one of the protective reactions in plants exposed to radiation. The effect was more distinct in cv. Magnat, which produced almost three times more of the mentioned pigments than the non-irradiated plants. Tevini *et al.* (1991) showed that accumulation of these pigments in rice caused a reduction of damage of photosynthetic activity of mesophyll chloroplasts. Gonzales *et al.* (1996) observed that UV-B irradiated pea plants of waxless lines JI1389 had the same content of flavonoids as the control plants, while the irradiated plants of the cultivars Scout and Vedette had a significant higher content.

Changes of the enzymatic activity were found in the irradiated plants of both triticale forms. The peroxidase activity increased in the irradiated plants, while catalase activity decreased. More marked changes in the activity of these enzymes were observed in RAH 336 plants than in cv. Magnat. The increase in peroxidase activity was observed by Panagopoulos *et al.* (1990) in irradiated sugar beet plants. Dai *et al.* (1997) found that ascorbate peroxidase activity in irradiated rice plants decreased with time, while in the control plants it stayed constant. The catalase activity was enhanced during the initial stage of treatment but after three weeks no difference was observed between the treated and control plants. Simultaneous changes of other enzymes activity: ascorbic acid, glutation and superoxide dismutases were observed, which shows that the extent of activity of each enzyme depends on plant species, time of UV-B treatment and also on experimental conditions (Dai *et al.* 1997).

We observed a decrease of plant height and width of leaves after exposure of plants to UV-B radiation, as compared to the control. The irradiated RAH 336 plants were characterised by relatively stronger height decrease (to 86 % of the control plants) than the cv. Magnat. The decrease of leaf width and increase of dry mass was similar in both triticale genotypes. Lower growth of the irradiated plants point to a negative effect of UV-B radiation as well as an activation of protective mechanisms such as a reduction of leaf surface, which allows an absorbance of a lesser radiation dose. This indicate that protection lies also in an increase of leaves thickness, which leads to effective protection of deeper cell layers, which corresponds with data obtained by Day *et al.* (1995) in pea leaves.

This study confirmed the harmful impact of UV-B radiation on triticale plants. Both the traditional cv. Magnat and RAH 336 line responded to applied radiation in a similar way. However, differences were observed as far as the extent of particular reactions, was concerned. Perhaps greater differences would be manifested later, in more advanced growth stages of the plants. Our intention is to do further studies on that topic.

## References

Caldwell, M.M.: Solar Ultraviolet Radiation and the Growth and Development of Higher Plants. - Academic Press, New York 1971.

Caldwell, M.M., Björn, L.O., Bornmann, J.F., Flint, S.D., Kulandaivelu, G., Teramura, A.H., Tevini, M.: Effects of increased solar ultraviolet radiation on terrestrial ecosystems. - J. Photochem. Photobiol. B **46**: 40-52, 1998.

Caldwell, M.M., Flint, S.D., Searles, P.S.: Spectral balance and UV-B sensitivity of soybean: a field experiment. - Plant Cell Environ. **17**: 267-276, 1994.

Dai, Q., Yan, B., Huang S., Liu, X., Peng S., Lourdes, M., Chavez, A., Vergara, B., Olszyk, D.: Response of oxidative stress defence systems in rice (*Oryza sativa*) leaves with supplemental UV-B radiation. - Physiol. Plant. **101**: 301-308, 1997.

Day, T.A., Vogelmann, T.C.: Alterations in photosynthesis and pigment distributions in pea leaves following UV-B exposure. - Physiol. Plant. **94**: 433-440, 1995.

Deckmyn, G., Impens, L.: The ratio UV-B/PAR determines the sensitivity of rye to increased UV-B radiation. - Environ. exp. Bot. **37**: 3-12, 1997.

Deckmyn, G., Martens, C., Impens, L.: The importance of the ratio UV-B/PAR during leaf development as determining factor of plant sensitivity to increased UV-B irradiance: effects on growth, gas exchange and pigmentation of bean plants. - Plant Cell Environ. **17**: 295-301, 1994.

Gonzalez, R., Paul, N.D., Percy, K., Ambrose, M., McLaughlin, C.K., Barnes, J.D., Areses, M., Wellburn, A.R.: Responses to ultraviolet-B radiation (280-315 nm) of pea (*Pisum sativum*) lines differing in leaf surface wax. - Physiol. Plant. **98**: 852-860, 1996.

Gwynn-Jones, D., Johanson, U.: Growth and pigment production in two subarctic grass species under four different UV-B irradiation levels. - Physiol. Plant. **97**: 701-707, 1996.

Hao, X., Hale, B.A., Ormrod, D.P., Papadopoulos, A.P.: Effects of pre-exposure to ultraviolet-B radiation on responses of tomato (*Lycopersicon esculentum* cv. New Yorker) to ozone in ambient and elevated carbon dioxide. - Environ. Pollut. **110**: 217-224, 2000.

Holmes, M.G., Keiller, D.R.: Effects of pubescence and waxes on the reflectance of leaves in the ultraviolet and photosynthetic wavebands: a comparison of a range of species. - Plant Cell Environ. **25**: 85-93, 2002.

Jordan, B.R.: The effects of ultraviolet-B radiation on plants: a molecular perspective. - Adv. bot. Res. **22**: 97-162, 1996.

Kinnunen, H., Huttunen, S., Laakso, K.: UV-absorbing compounds and waxes of Scots pine needles during a third growing season of supplemental UV-B. - Environ. Pollut. **112**: 215-220, 2001.

Lichtenthaler, H., Buschmann, C., Rinderle, U., Schmuck, G.: Application of chlorophyll fluorescence in ecophysiology. - Radiat. Environ. Biophys. **25**: 297-308, 1986.

Maciorowski, R., Salak-Warzecha, K.: [Photosynthesis and plant growth of waxless and reduced-own triticale]. - Folia Univ. Agr. Stetin A **228** (91): 57-62, 2002. [In Polish.]

Murkowski, A., Skórska, E.: Chlorophyll *a* luminescence – an index of photoinhibition damages. - Curr. Topics Biophys. **21**: 72-78, 1997.

Panagopoulus, I., Bornman, J.F., Björn, L.O.: Effects of ultraviolet radiation and visible light on growth, fluorescence induction, ultraweak luminescence and peroxidase activity in sugar beet plants. - J. Photochem. Photobiol. B **8**: 73-87, 1990.

Pilon, J.J., Lambers, H., Baas, W., Tosserams, M., Rozema, J., Atkin, O.K.: Leaf waxes of slow-growing alpine and fast-growing lowland *Poa* species: inherent differences and responses to UV-B radiation. - Phytochemistry **50**: 571-580, 1999.

Rajendiran, K., Ramanujam, M.P.: Improvement of biomass partitioning, flowering and yield by triadimefon in UV-B stressed *Vigna radiata* (L.) Wilczek. - Biol. Plant. **48**: 145-148, 2004.

Renger, G., Volker, M., Eckert, H.J., Fromme, R.: On the mechanism of photosystem II deterioration by UV-B irradiation. - Photochem. Photobiol. **49**: 97-105, 1989.

Schreiber, U., Bilger, W., Neubauer, C.: Chlorophyll fluorescence as a nonintrusive indicator for rapid assessment of *in vivo* photosynthesis. - In: Schulze, E.D., Caldwell, M.M. (ed.): Ecophysiology of Photosynthesis. Pp. 49-70. Springer-Verlag, Berlin 1994.

Skórska, E: The effect of UV-B radiation on the chlorophyll fluorescence parameters of the husked and the naked oat. - Acta agrobot. **52** (1-2): 149-152, 1999.

Skórska, E.: [Reaction of some crop plants on UV-B radiation.] - Rozprawy 192. Agricultural University of Szczecin, 2000a. [In Polish.]

Skórska, E.: Responses of pea and triticale photosynthesis and growth to long-wave UV-B radiation. - Biol. Plant. **43**: 129-131, 2000b.

Skórska, E.: The effect of ultraviolet-B radiation on triticale plants. - Folia Univ. Agr. Stetin A **82**: 249-254, 2000c.

Skórska, E., Lewandowski, R.: [Comparison of reactions of three oat varieties to UV-B radiation.] - Biul. IHAR **229**: 199-204, 2003. [In Polish.]

Steinmüller, D., Tevini, M.: UV-B-induced effects upon cuticular waxes of cucumber, bean, and barley leaves. In: Caldwell, M.M. (ed.): Stratospheric Ozone Reduction, Solar Ultraviolet Radiation and Plant Life. Pp. 261-269. Springer-Verlag, Berlin 1986.

Tevini, M., Braun, J., Fieser, G.: The protective function of the epidermal layer of rye seedlings against ultraviolet-B radiation. - Photochem. Photobiol. **53**: 329-333, 1991.

## Erratum

SERRANO, L., PEÑUELAS, J.: Contribution of physiological and morphological adjustments to drought resistance in two Mediterranean tree species. - Biol. Plant. **49**: 551-559, 2005.

We appologize to our authors and readers for misprints in Table 1 published on page 555. Correct table is the following:

Table 1. Descriptive statistics for several ecophysiological variables. Pressure-volume derived parameters [modulus of elasticity (ε), osmotic water potential at full ($\pi_{100}$) and zero pressure potential ($\pi_0$), relative water content at zero turgor ($RWC_0$), apoplastic water fraction (AWF), and saturated to dry mass ratio (TM/DM)] were measured at least on 8 twigs at each season and year. Net photosynthetic rate ($P_N$), stomatal conductance ($g_s$) and transpiration rate (E) were measured on 4 leaves of each species at each season and year. Hydraulic efficiency (HE) is derived from the ratio between stomatal conductance and plant water potential amplitude for a given day. SD and CV indicate standard deviation and coefficient of variation, respectively.

| Parameters | *Phillyrea latifolia* | | | | | *Quercus ilex* | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | minumum | maximum | mean | SD | CV [%] | minumum | maximum | mean | SD | CV [%] |
| ε [MPa] | 2.92 | 16.38 | 7.27 | 5.163 | 71.1 | 0.81 | 5.01 | 2.57 | 1.472 | 57.28 |
| $\pi_0$ [MPa] | -3.11 | -1.70 | -2.23 | 0.430 | 19.3 | -2.81 | -1.50 | -2.14 | 0.459 | 21.94 |
| $\pi_{100}$ [MPa] | -2.40 | -0.61 | -1.26 | 0.524 | 41.6 | -1.48 | -0.50 | -0.85 | 0.319 | 37.33 |
| $RWC_0$ [%] | 77.7 | 90.8 | 84.0 | 3.54 | 4.2 | 62.6 | 78.0 | 72.0 | 4.50 | 6.26 |
| TM/DM | 2.00 | 3.45 | 2.28 | 0.449 | 19.7 | 1.96 | 3.72 | 2.36 | 0.628 | 26.65 |
| AWF | 0.201 | 0.754 | 0.583 | 0.196 | 33.6 | 0.11 | 0.63 | 0.50 | 0.147 | 29.38 |
| $P_N$ [$\mu$mol $m^{-2}$ $s^{-1}$] | -2.386 | 5.129 | 2.694 | 2.311 | 85.8 | -2.06 | 7.32 | 3.81 | 2.772 | 72.74 |
| $g_s$ [mol $m^{-2}$ $s^{-1}$] | 0.038 | 0.320 | 0.125 | 0.109 | 87.4 | 0.02 | 0.53 | 0.14 | 0.154 | 106.6 |
| E [mmol $m^{-2}$ $s^{-1}$] | 0.381 | 3.687 | 1.82 | 0.982 | 53.9 | 0.352 | 3.72 | 1.593 | 0.949 | 59.6 |
| HE [mol $m^{-2}$ $s^{-1}$ $MPa^{-1}$] | 0.023 | 0.376 | 0.130 | 0.114 | 87.9 | 0.03 | 0.40 | 0.15 | 0.130 | 87.8 |