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# Spectroradiometrical evaluation of photosynthetic efficiency in durum wheat subjected to drought

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**SUMMARY –** We aimed to study the change in the spectral signature associated with both the development of photoinhibition and protective mechanisms during water stress in durum wheat. Seedlings were cultivated in pots in a green-house during summer time. In order to provide water stress, water was not supplied for 8 days. Photosynthetic and transpirative gas exchange, chlorophyll-modulated fluorescence and spectral changes in transmitted light were measured in fully and recently expanded leaves every 2-3 days. As the stress due to drought arose, the photosynthetic efficiency of the plants progressively decreased, whereas their non-photochemical quenching (Qnp) increased. Thus indicating a higher amount of incoming energy dissipating through processes other than photosynthesis. Such changes in photosynthetic efficiency and Qnp were associated with a decrease in the spectroradiometrical index PRI, whose ability to monitor the photosynthetical efficiency of leaves under drought was tested.

**Key words:** Chlorophyll fluorescence, durum wheat, photochemical reflectance index, photoinhibition, non photochemical quenching, water stress.

***RESUME –*** *"Evaluation spectroradiométrique de l'efficacité photosynthétique du blé dur soumis à la sécheresse". Le but du travail suivant est l´évaluation dans le cas du blé dur des changements dans la signature spectrale associés au développement de la photoinhibition et des mécanismes de photoprotection durant le stress hydrique. Des semis ont été cultivés durant l´été dans des pots placés dans une serre. Le stress hydrique s´est développé chez les plantes croissant sans eau durant une période de 8 jours. La photosynthèse, la transpiration, la fluorescence modulée des chlorophylles et les changements spectraux en lumière transmise étaient mesurés au sein des feuilles largement étendues chaque 2-3 jours. Lorsque le stress hydrique avance, les plantes montrent une diminution progressive dans l´efficience photosynthétique tandis que le "non-photochemical quenching" (Qnp) augmente indiquant ainsi la dissipation de l´excès d´énergie par des processus autres que la photosynthèse. Ces changements de l´efficience photosynthétique et du Qnp sont associés à une réduction des valeurs de l´indice spectroradiométrique. On discute l´utilité de l´indice PRI pour la télédétection de l´efficience photosynthétique des feuilles sous sécheresse.*

***Mots-clés :*** *Blé dur, fluorescence de la chlorophylle, indice photochimique de réflectance, photoinhibition, Qnp, stress hydrique.*

## Introduction

Exposure to radiation is often higher than required for carbon assimilation, which results in the photoinhibitory impairment of photosynthesis. Several mechanisms prevent the plants from damage associated with excessive light and photochemical reactions others than photosynthesis take place (Owens, 1996). Through these mechanisms, the excess of energy is dissipated as heat (Owens, 1996) leading to "non photochemical quenching" (Qnp) of the energy absorbed. In order to evaluate Qnp, chlorophyll fluorescence emitted by the photosystem II is measured (Bolhár -Nordenkampf and Oquist, 1993). The reversible conversion of violaxanthine into anteraxanthine and zeaxanthine (Gilmore, 1997) is regarded as the main process involved in Qnp. The dynamics of such interconversion, usually referred to as "xanthophylls cycle", is essential to adaptation of the photosynthetic apparatus to high irradiances and associated stresses, such as water stress. The pool of zeaxanthine in the chloroplasts can be monitored by indirect but non-aggressive measurements of the changes in the light reflected or absorbed at wavelengths around 530 nm ( Gamon *et al.,* 1990). Whereas the radiation absorbed by the leaf and not used in photosynthesis can be dissipated through the xanthophylls cycle, the photosynthesis efficiency (i.e. the net photosynthesis: radiation absorbed ratio) will decrease as a result of this excess of radiation. Thus, the changes in either reflectance or transmittance in this region of the spectra have been associated with the pool of zeaxanthine and, more indirectly, with the photosynthetic efficiency of leaves using the Photochemical Reflectance Index (PRI) (Gamon *et al.,* 1990; Filella *et al.,* 1996). Drought, defined as water deficit associated with high irradiance and temperature is the main abiotic stress in Mediterranean conditions, in which it greatly decreases the photosynthetic activity and leaf area duration of durum wheat during grain filling (Muller and Whitsitt, 1996; Araus *et al.,* 1998). In this context, a fast and non-destructive assessment of the photosynthetic efficiency under drought can be of major importance for breeding and crop management. Here we study the change in the spectral signature associated with the development of photoinhibition and the protective mechanisms during water stress in durum wheat.

## Matérials and methods

Durum wheat seeds (*Triticum turgidum* L. var. *durum*) cv Mexa were grown in 3 L plastic pots (3 plants per pot) filled with peat and perlite 2:1 (v/v). The seedlings were cultivated in a green-house with cooling system during summer. In 4-week-old plants, no water supply was provided in order to ensure water stress. The control plants were maintained at field capacity throughout the experiment. The water content of the substrate was determined by gravimetry. The relative water content (RWC) was measured in the basal half of the leaves used in the gas exchange and fluorescence measurements. Net $CO_2$ assimilation and stomatal conductance were measured with a portable gas-exchange system (LI- COR 6200, LI-COR Inc., Lincoln, NE, USA) at *in situ* PPFD conditions. Chlorophyll fluorescence parameters were measured with a modulated fluorometer (Mini PAM Photosynthesis Yield Analyzer, Waltz, Effeltrich, Germany) in the same leaves used for IRGA measurements. The maximum ($F_m$) and the initial fluorescence ($F_0$) were measured after 30 minutes of to darkness. $F_s$ and $F'_m$ were measured in the specific PPFD conditions of the leaves. $F'_0$ was estimated following Oxborough and Baker (1997), while the non-photochemical quenching (Qnp) and the quantum yield of PSII ($\Phi_{PSII}$ , $\Delta F/F'_m$) were calculated following Bolhár- Nordenkampf and Öquist (1993). Leaf transmittance was detected with a narrow-bandwidth visible/near infrared spectroradiometer (Field Spec UV/VNIR, Analytical Spectral Devices Inc., Boulder, Colorado, USA) fitted with an adapter (i.e. “transmittance chamber”) developed by the “Servei de Camps Experimentals” of the University of Barcelona. The transmittance spectrum was calculated as the ratio transmitted : incident spectra. The photochemical index (PI) was calculated as $(T_{531\,nm} - T_{570\,nm}) / (T_{531\,nm} + T_{570\,nm})$ (Gamon *et al.,* 1997). All measurements (IRGA, fluorescence and spectroradiometry) were performed at noon, every 2-3 days, in fully, recently expanded leaves.

## Results and discussion

The relative water content of control plants remained around 95% throughout the experiment. In the plants submitted to progressive water stress, the RWC strongly decreased after 4 days, reaching the final value of about 55% (Fig. 1a). Net $CO_2$ assimilation and stomatal conductance of stressed plants progressively and strongly decreased throughout the treatment, with final values close to zero (Fig. 1b). The plants undergoing drought stress showed a decrease in $\Phi_{PSII}$, related to the decrease in the photosynthetic rate, an increase in the Qnp (data not-shown), and a decrease in the photochemical index (PI). In contrast, none of these parameters showed significant ($p < 0.05$) changes in control plants . When the individual leaves used for measurements in the control and stressed plants during one day were combined, PI was markedly negatively related with both $\Phi_{PSII}$ (data not-shown) and Qnp (Fig. 2). The change in PI, caused by stress, has been associated with changes in the spectra (either reflected or transmitted) at wavelengths near 530 nm, which result from the interconversions between violaxanthin and zeaxanthin related with the cycle of the xanthophylls (Filella *et al.,* 1996). Thus, as well as Qnp (Ruban *et al.,* 1993), PI can be considered an indirect indicator of the photochemical efficiency of the photosystem II (Gamon *et al.,* 1997). Here, the high correlation between PI and Qnp would agree, at first sight, with the above-mentioned studies. However, for the set of individual leaves measured in both treatments during a given day, PI was surprisingly not correlated with $\Delta F/F'_m$ (data not-shown). A concomitant increase in the total pool of carotenoids, rather than changes in the relative content of zeaxanthin and violaxanthin, may account for the decrease in PI. The increase in the overall content of carotenoids has been reported as a photoprotective response of plants to different kinds of stress (Demmig-Adams *et al.,* 1992). Regarding barley, the correlation between PRI and the total content of zeaxanthin is very high, although PRI was much less correlated with the epoxidation state of the of the pool of xantophylls (Filella *et al.,* 1996). The progressive increase in Qnp throughout the drought stress treatment would support such possibility. However the lack of association between PI and $\Delta F/F'_m$ can be due to other reasons. Thus the changes in PI with drought stress could be due, at least partially, from mechanisms which are not related no related with energy quenching, e.g. the selective scattering due to changes in the water content of leaves. Further studies will be carried out to clarify these aspects.

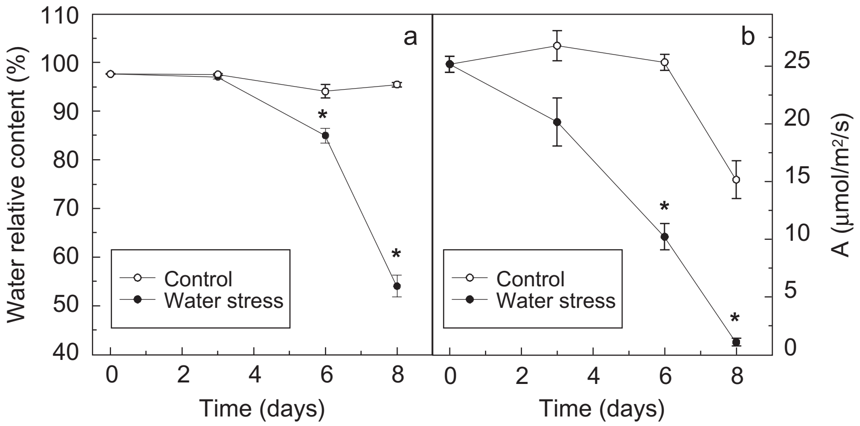


Fig. 1. Changes in (a) relative water content of leaves and (b) photosynthetic rate in control and water stressed plants of durum wheat. Each value represents the mean ± SE of 6 measurements performed at midday. Significant differences ($p \leq 0.05$) between treatments were found to according the LSD test (asterisk). For further details, see Material and methods.

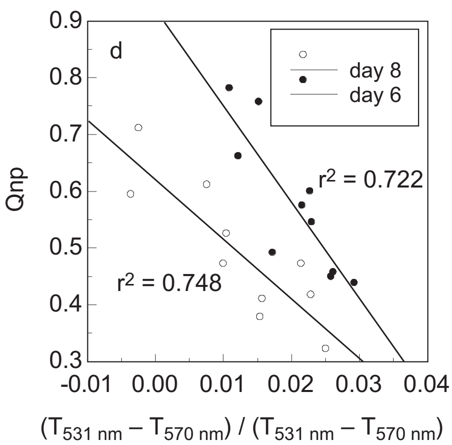


Fig. 2. Relationship between the non-photochemical quenching (Qnp) and the Photochemical Index (PI). Each point represents the mean of the measurements performed at midday on an individual leaf of either the stress or the control treatments. Measurements were performed 6 (filled circle) and 8 (empty circles) days after imposing water withholding. For further details, see Material and methods.

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