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EVALUATION OF DROUGHT-INDUCED MODULATION IN PHOTOSYNTHETIC PERFORMANCE OF *TRITICUM AESTIVUM* VARIETY LOK-1

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ABSTRACT

Drought stress is a global issue that influences growth and productivity of plants. The main objective of this study was to identify indicators related to drought tolerance through fast polyphasic chlorophyll fluorescence kinetics in wheat var. LOK-1. Various parameters such as initial fluorescence (F_0), the maximum primary yield of photosystem II (F_V/F_M), maximum quantum yield of photosystem II (F_V/F_M), performance index, energy pipeline models were measured in response to drought stress. The results indicate that the plants have capability to maintain photosynthetic photosynthetic performance under mild drought stress.

Keywords: Triticum aestivum L. variety LOK-1, Photosystem II, Chlorophyll fluorescence kinetics, , Drought tolerance.

INTRODUCTION

Drought is one of the major abiotic stresses decreasing crop development and production (Jiang and Zhang, 2004). Usually, drought stress happens when the available water in the soil is decreased and atmospheric conditions cause continuous loss of water by transpiration or evaporation (Khajeh *et al.*, 2003). Drought also plays a serious role in affecting cereal grain production and quality. In the present time, with a growing population and global climate change; the situation is becoming more dangerous (Hongbo *et al.*, 2005).

Various adaptations and fast actions are essential to cope with drought stress. Plants under dry conditions modulate their metabolism process, resulted in many physiological and biochemical modifications (Ort, 2001). Usually, plants respond to early drought stress by a rapid closure of stomata which decreases the CO_2 transmission rate. This leads to a decreased CO_2 concentration that finally restricts photosynthetic machinery by direct inhibition of the photosynthetic enzyme RuBisCO (Haup-Herting and Fock, 2000) or ATP synthase (Tezara et al., 1999). Among drought responses, photosynthesis is the crucial processes to be affected by drought stress (Chaves, 1991). Evaluating changes in photosynthetic performance can give information on the drought tolerance of a plant. For this purpose, chlorophyll fluorescence has become a broad, non-invasive technique to study changes in the photosynthetic machinery (Nunes et al., 2008; Silvestre et al., 2014). Chlorophyll fluorescence analysis has been commonly applied as a non-destructive technique to investigate the photosynthetic efficiency of plants, under stress conditions. Definitely, the information of fluorescence signals gives complete information on the status and function of Photosystem II (PSII) reaction centers, both the donor and acceptor sides and light harvesting antenna complexes. Over the last few years, numerous techniques have been suggested to improve plant performance in drought conditions. In recent times, the chlorophyll *a* fluorescence OJIP transient has been used to measure the damage of the photosynthetic machine under drought stress. The OJIP transient is definite by O, J, I, and P steps, parallel to the redox states of PS II and PS I (Strasser et al., 2000, 2004).

The "JIP test" developed by Strasser *et al.*, (1995) based on the theory of "energy flow" through thylakoid membranes (Strasser *et al.*, 2000), converts the fluorescence measurements of the transients (O–J–I–P) into many phenomenological and biophysical parameters that measures PS II functions. Each letter explains a specific variation in the curve and this permits the analysis of the structural and functional parameters that finally shows the activity of the whole photosynthetic machinery (Strasser *et al.*, 2004).

Research Article

The JIP-test analyze the polyphasic rise of the chl fluorescence OJIP transient and has been established to examine *in vivo* the "vitality" of plants and the adaptive behavior of the photosynthetic apparatus to drought stress (Tsimilli-Michael *et al.*, 1996; Strasser *et al.*, 2004).

The chlorophyll fluorescence method is potentially useful for developing varieties for drought tolerance. To enhance crop production, it is crucial to find out the machinery of plant responses to drought conditions with the vital aim of developing drought tolerant crops in the water limited regions. On the basis of various parameters of fluorescence induction curves, F_V/F_M ratio, performance index (PI), and pipeline models, efforts were carried out to understand the effects of drought stress on photosynthesis in wheat var. LOK-1.

MATERIALS AND METHODS

Plant Materials and Growing Conditions: T. aestivum L. var. LOK-1 was evaluated concerning their ability to endure drought stress. Seeds were obtained from Maharana Pratap Agriculture University and Technology (MPUAT), Udaipur (Rajasthan, India) and were sown *in vivo* in germination trays containing 50% clay, 25% sand, and 25% humus under controlled conditions at 15°C under a 12 h photoperiod (Fig. 1a). Prior to sowing, surface sterilization of seeds was done with 0.1% HgCl₂ followed successive washings with distilled water. Seedlings were watered twice a day.

In Vivo Drought Treatment: The germinated plant of wheat variety was equally well watered for 3 weeks prior to exposure to drought stress treatment. After 3 weeks, at the stage of 2 fully developed leaves, the plants were divided into two sets (each of 100 plants), out of which one set was subjected to drought stress by withholding of water supply, while the second set was watered regularly and served as a control. **Analysis of polyphasic chlorophyll fluorescence kinetics:** Chlorophyll *a* fluorescence O-J-I-P transients were recorded till 6 days of drought stress treatment in the growth chamber at 20[°]C under dim green light with a Plant Efficiency Analyzer, (PEA, Hansatech Instruments, Kings Lynn, Norfolk, U.K.). Chlorophyll *a* fluorescence transient was induced by a red light (peak at 650 nm) of 3200 mol m⁻² s⁻¹ given by the PEA over an array of six light-emitting diodes. Fluorescence data were analyzed as per the equations of JIP test proposed by Strasser and Strasser, (1995) (Table: 1).

Jumpies	
Extracted and technical fluorescence parameter	
$Fo = F_{50 \ \mu s}$	fluorescence intensity at 50 µs
F _J	fluorescence intensity at at 2 ms
F _I	fluorescence intensity at at 30 ms
F _M	maximal fluorescence intensity
Specific fluxes or specific activities	
ABS/RC	Absorption flux per reaction center (RC)
$TR_0/RC = M_0 \cdot (1/V_J)$	Trapped energy flux per RC
$ET_0/RC = M_0 \cdot (1/V_J) \cdot \psi_0$	Electron transport flux per RC
Phenomenological fluxes or phenomenological	ogical activities
ABS/CS	Absorption flux per cross section
TR ₀ /CS	Trapped energy flux per cross section
ET ₀ /CS	Electron transport flux/ cross section
$DI_0/CS = (ABS/CS) - (TR_0/CS)$	Dissipated energy flux per cross section
Quantum efficiencies or flux ratios	
$\Phi_{\rm P0} = [1 - (F_0/F_m)] = F_v/F_m$	Maximum quantum yield for primary
	photochemistry
Density of RCs	
RC/CS	Density of reaction centers

Table 1: Formulae and glossary of terms used by the JIP-test for the analysis of chlorophyll *a* fluorescence transient OJIP emitted by dark-adapted photosynthetic samples

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RESULTS AND DISCUSSION

The analysis of the kinetics of chlorophyll fluorescence has been a non-invasive technique used broadly for the investigation of plant responses under various stress conditions such as salinity, chilling, high temperatures, nutrient deficiency and drought (Greaves and Wilson, 1987; Brennan and Jefferies, 1990; Hakam *et al.*, 2000). In the present study, chlorophyll fluorescence analysis was undertaken to understand the physiological basis of drought tolerance in wheat var. LOK-1. The chlorophyll fluorescence intensity started from the initial (F_0) intensity and increased to the highest intensity (F_M) in both control and drought-stressed plants (Fig. 1b). The shape of the O-J-I-P transient is very sensitive to drought stress (Strasser and Tsimilli-Michael, 2001; Sayed, 2003; Van Heerden *et al.*, 2003, Govindachary *et al.*, 2004). In the present study, a wide variation between chlorophyll fluorescence OJIP kinetics of control and drought stressed plants was observed. The fluorescence intensity decreased in response to only in moderate or severe drought conditions. Under severe drought stress condition, the shape of the O-J-I-P transient changed in wheat leaves to decrease in F_M , resulting in lowering of F_V .



Figure 1. (a) Three years old plants of *T. aestivum* var. Lok-1, (b) chlorophyll a fluorescence induction curves of control and drought-stressed plants, Thylakoid membrane and leaf models of control (c,d) and drought stressed plants (c,d) of *T. aestivum* var. Lok-1.

The O-J phase of the chlorophyll fluorescence transient is related to a photochemical reduction of the primary Quinone acceptor Q_A in PS II reaction center and is sensitive to severe stress. Schansker *et al.*, (2005) studied that the I-P phase depends on both, electron flow through PS I and a transient block of electron flow on the acceptor side of PS I which depends on PS I content and suggests a change in the

Research Article

ratio between PSI and PSII. F_V/F_M is considered to be a sensitive indicator of plant photosynthetic performance under drought stress condition. The maximum quantum yield for primary photochemistry (F_V/F_M) was declined from 0.737 (control) to 0.025 (after 6 days of drought stress treatment) in LOK-1.

In present study, it was noted that all component, except light harvest complexes (LHC), of photosynthetic apparatus of Lok-1 variety are highly resistant to drought stress. An increase in ABS/RC, TR/RC and ET/RC was observed with drought treatment (Fig. 1c,e). On the other hand, a slight reduction in ABS/CS, TR/CS and ET/CS was observed in plants subjected to water deficit condition (Fig. 1d,f). 6 days drought treatment caused a drastic reduction in the density of active reaction centers (RC/CS) in LOK-1 variety of wheat. Despite decline in RC/CS, the remaining active reaction centers enhanced their efficiency to maintain photosynthesis process in LOk-1 variety.

Leaf models suggest that the light harvesting complexes of plants are highly sensitive to drought stress. Degradation of LHCs might be the root cause of decline of photosynthesis in LOK-1 variety of wheat under water deficit condition. In future, through breeding programmes, the drought tolerance efficiency of Lok-1 variety of wheat can be enhanced by increasing drought resistance of LHCs in *T. aestivum* var. Lok-1.

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Research Article

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