

## Changes of chlorophyll *a* fluorescence parameters in water limited maize IBM population

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### ABSTRACT

Drought is the second most important cause of yield loss after low soil fertility. Aim of this study was to investigate changes in chlorophyll *a* fluorescence parameters of maize subjected to drought stress during tasseling. Results have shown that all selected parameters in drought stressed maize differ significantly from control. Negative effects can be placed in three groups: inactivation of reaction centers turning them into heat sinks (decrease in RC/ABS, increase in DIO/RC), impairment of OEC (oxygen evolving center) (increase in F300), decrease in electron transport efficiency and related problems in  $Q_A$  (primary quinone acceptor) and plastoquinone oxidation.

**Keywords:** maize, drought, chlorophyll *a* fluorescence, JIP-test

### Introduction

The flexibility of normal metabolism allows the development of responses to environmental changes which fluctuate regularly and predictably over daily and seasonal cycles. Stress begins with a constraint or with highly unpredictable fluctuations imposed on regular metabolic patterns that cause bodily injury, disease, or aberrant physiology. After soil fertility, drought is the second most important cause of maize yield loss. Drought is a meteorological term usually defined as a period without significant rainfall. Drought stress in plants is characterized by reduction of water content, decreased water leaf potential, closure of stomata, loss of turgor and more severe drought stress causes inhibition of photosynthesis and disturbances in plant metabolism (Jaleel et al. 2008, Smirnov 1993). In maize, severe drought stress at tasseling stage reduces the yield by affecting the number of kernels per row, number of kernel rows, harvest index, number of kernels per cob and grain yield per plant (Anjum et al. 2011). Chlorophyll *a* fluorescence has been used as a probe to assay the state of photosystem II (PSII) in various types of stresses including drought (Araus et al. 1998, Oukarroum et al. 2007).

### Materials and methods

Trial was set in Altinova (Turkey) under well-watered conditions regulated by optimum irrigation and water limited regulated by suboptimum irrigation (70% of optimum irrigation) during July 2015. In Altinova mean monthly rainfall is less than 5 mm in June and July and plants completely depend on irrigation. Seeds of 216 entries including 212 near isogenic lines of intermated B73×Mo17 maize population (IBM population) were planted in 18 blocks for watered and water limited treatment, 20 plants per plot. Standard agrotechnical practices for maize were used in all pre-planting procedures.

*Abbreviations:* Fo – minimal fluorescence intensity, Fm – maximal fluorescence intensity, F300 – fluorescence intensity at 300 μs, AREA – area over the OJIP transient, Vj – relative variable fluorescence at J-step, Vi – relative variable fluorescence at I-step, N – turnover number, Tro/ABS – maximum quantum yield of primary photochemistry, TRo/ABS – maximum yield of electron transport, ETo/TRo – efficiency of a trapped exciton to move an electron into the electron transport chain further than  $Q_A$ , DIO/RC – specific flux for dissipation, RC/ABS – ratio of reaction centers and absorption

Control (Normal) was watered every 14 days and for water limited treatment water was withheld 14 days before tasseling.

Chlorophyll a fluorescence measurements were done during tasseling in the field (in July). Measurements were conducted in the morning before 10 a.m. due to midday depression of photosynthesis. Leaves were dark adapted before measurements for 30 minutes using dark adaption leaf clips. After dark adaptation chlorophyll a fluorescence was measured using Handy-PEA fluorimeter (Plant Efficiency Analyser, Hansatech Instruments Ltd, Great Britain). Fluorescence was measured on ear leaves in two replications per entry. Handy-PEA fluorimeter measures changes in chlorophyll fluorescence for 1 s, starting 50  $\mu$ s after the light pulse. Obtained data is analyzed using JIP test which outputs biophysical changes that quantify the flow of energy through PSII (Strasser et al. 2004).

Non-parametric Kruskal-Wallis one way analysis of variance by ranks was applied using R statistical software (R core team, 2013). Dunett's method was used to determine significance values of difference as compared to control samples. Number of observations was the same in normal and water limited environment and equaled 432.

## Results and discussion

Kruskall-Wallis one way analysis of variance showed that selected chlorophyll a fluorescence parameters differed significantly in water limited treatment compared to control (Table 1). Differences were significant at the  $P < 0.001$  significance level, except for turnover number (N) which was significant at the  $P < 0.01$  significance level.

Table 1. Effect of water limited treatment on maize plants analyzed by selected JIP-test parameters (n = 216, mean  $\pm$  SE)

Parameter	Treatment	
	Normal	Water limited
Fo	221,919 $\pm$ 0.732	251,734 $\pm$ 1.685***
F300	342,712 $\pm$ 1.902	396,430 $\pm$ 3.238***
Fm	1166,216 $\pm$ 5.763	1134,373 $\pm$ 5.275***
AREA	34384,882 $\pm$ 248.430	31651,146 $\pm$ 240.463***
Vj	0,301 $\pm$ 0.002	0,350 $\pm$ 0.003***
Vi	0,602 $\pm$ 0.004	0,648 $\pm$ 0.003***
N	63,726 $\pm$ 1.093	68,118 $\pm$ 0.818**
Tro/ABS	0,808 $\pm$ 0.001	0,776 $\pm$ 0.002***
Eto/ABS	0,565 $\pm$ 0.002	0,506 $\pm$ 0.003***
Eto/Tro	0,699 $\pm$ 0.002	0,650 $\pm$ 0.003***
DIo/RC	0,410 $\pm$ 0.006	0,562 $\pm$ 0.00***
RC/ABS	0,482 $\pm$ 0.003	0,423 $\pm$ 0.003***

\*\* Significantly different from control,  $\alpha = 0.01$

\*\*\* Significantly different from control,  $\alpha = 0.001$

Spiderplot (Figure 1) shows that values of quantum yields decreased (Tro/ABS, Eto/ABS, Eto/Tro) in water limited treatment. Decreases in yields that describe the efficiency of electron transport (Tro/ABS, Eto/ABS) suggest photoinhibitory damage to PSII caused by water deficit, likewise decrease in maximum quantum yield of PSII photochemistry suggests impaired PSII photochemical efficiency. Increased fluorescence at J and I step (Vj and Vi, respectively) suggest accumulation of reduced primary quinone acceptor ( $Q_A^-$ ) and plastoquinone or their inability to transfer electrons to dark reactions (Kalaji et al 2014) supporting the observed decrease in quantum yields. This is also backed by decrease in AREA parameter which is proportional to the pool size of reduced plastoquinone and a reduction in this parameter suggests that electron transfer from reaction centers to quinone pool is blocked. Decrease in the number of active reaction centers (RC/ABS) in water limited treatment suggests susceptibility to photoinhibition and inactivation of reaction

centers to form heat sinks to dissipate the excess of absorbed light which is backed by increase in dissipation energy (DIO/RC) in water limited treatment shown on Figure 1. Rise of initial fluorescence ( $F_o$ ), decrease of maximum fluorescence ( $F_m$ ) and the resulting decrease in  $F_v/F_m$  has been previously shown in drought and temperature stressed plants (Havaux 1995, Paknejad et al. 2007). F300 or fluorescence at 300  $\mu$ s, where the so called K peak usually appears, was also found to significantly increase under drought stress, implying that oxygen evolving center (OEC) of PSII suffered inactivation or inhibition of electron transport (Strasser and Srivastava 1995).

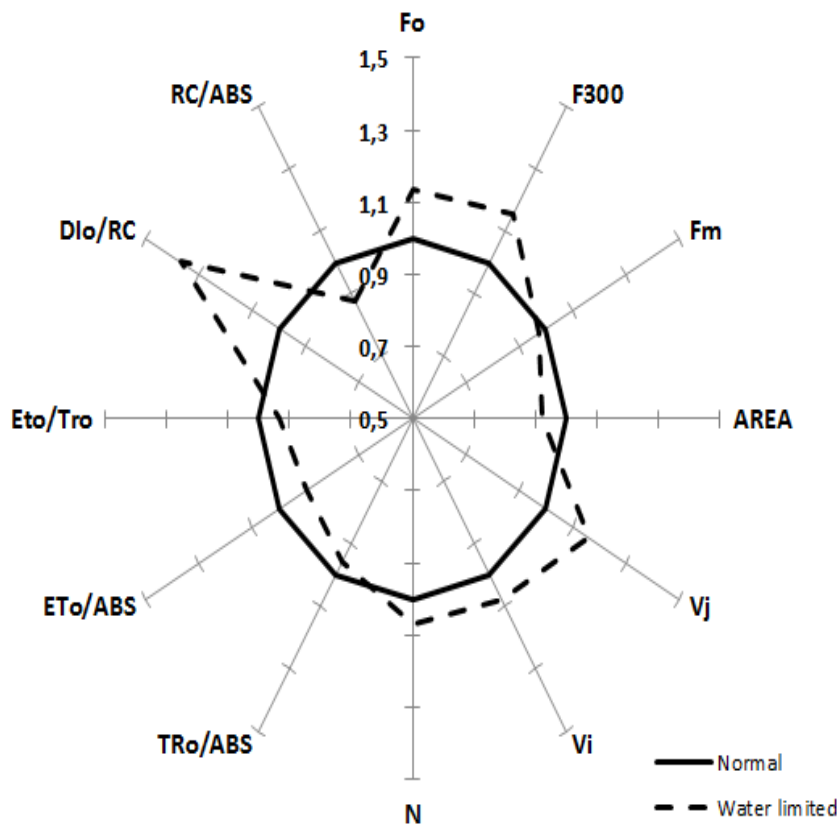


Figure 1. Effects of water limited treatment on maize plants analyzed by selected JIP-test parameters plotted relative to their respective controls (set as reference black circle = 1.0). Values represent averages (n = 432).

## Conclusions

Based on the results of this study we can conclude that drought during tasseling in the duration of 14 days resulted in significant changes of chlorophyll *a* fluorescence parameters. Negative effects of drought stress can be grouped in three groups: inactivation of reaction centers turning them into heat sinks (decrease in RC/ABS, increase in DIO/RC), impairment of OEC (increase in F300), decrease in electron transport efficiency and related problems in  $Q_A$  and plastoquinone oxidation.

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