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Research article

RESPONSES OF SOYBEAN LEAVES AND GRAIN YIELD TO WATER STRESS AT REPRODUCTIVE STAGES

Kazem Ghassemi-Golezani*, Ramin Lotfi

Department of Plant Eco-physiology, Faculty of Agriculture, University of Tabriz, Tabriz, Iran

Email: golezani@gmail.com*

ABSTRACT : A field experiment was carried out as split plot based on RCB design with four replications in 2011 to assess the effects of different irrigation treatments (I_1, I_2, I_3 and I_4: well-watering on the bases of 70 mm evaporation from class A pan and irrigation disruptions during flowering, during grain filling and during flowering and grain filling stages, respectively) on some physiological traits and grain yield of three soybean cultivars (Clark, Williams and L17). Results showed that with increasing water stress, leaf temperature increased, while chlorophyll content index, quantum yield of the PSII (Fv/Fm) and grain yield decreased. Maximum leaf temperature and minimum chlorophyll content were observed under water stress. Maximum reduction in Fv/Fm and grain yield per unit area was observed when plants were subjected to water stress during flowering and grain filling stages (I_4). Williams produced the highest grain yield per unit area, which related with higher leaf chlorophyll content of this cultivar.

Keywords: Chlorophyll content, fluorescence, grain yield, soybean, water stress

INTRODUCTION

Plants are subjected to several harsh environmental stresses that adversely affect their growth, metabolism, and yield. To survive against the stress, plants have involved in a number of physiological changes [1, 2]. The susceptibility of plants to drought stress varies depending on stress level, different accompanying stress factors, plant species and their developmental stages [3].

Photosynthesis and cell growth are the primary processes which are affected by stress [4]. Chlorophyll is one of the major chloroplast components for photosynthesis, and relative chlorophyll content has a positive relationship with photosynthetic rate [5]. Leaf chlorophyll content is fundamental to understanding a plant response to the environment in which it resides. Stresses that involve deficiencies of N and water will adversely affect the amount of chlorophyll in plants [6].

Fluorescence of chlorophyll reflected the photochemical activities of PSII [7], with optimal values of around 0.832 measured from most plant species [8]. Values lower than this are measured when the plant is exposed to stress, indicating a particular phenomenon of photo-damage to PSII reaction centers, and the development of slowly relaxing quenching process which reduce the maximum efficiency of PSII photochemistry [9, 10]. Environmental stresses that affect PSII efficiency leads to a characteristic decrease in the Fv/Fm ratio [11, 12].

A plant stress measurement with hand-held infrared thermometers (IRT) is based on the fact that transpiration cools the leaf surface. As water becomes limiting, stomatal conductance and transpiration decrease and leaf temperature increases. A temperature measurement on individual leaves is a good indicator of water potential [13] and plant stress [14]. Thus, this research was carried out to evaluate changes in chlorophyll content, fluorescence and leaf temperature in leaves of soybean cultivars in response to water stress during reproductive stages and their consequences on crop yield.

MATERIAL AND METHODS

A split plot experiment (using RCB design) with four replications was conducted in 2011 at the Research Farm of the Faculty of Agriculture, University of Tabriz, Iran (latitude 38.05°N, longitude 46.17°E, altitude 1360 m sea level). The climate is characterized by mean annual precipitation of 245.75 mm per year and mean annual temperature of 10°C.

Irrigation treatments (I_1, I_2, I_3 and I_4: well-watering on the bases of 70 mm evaporation from class A pan and irrigation disruptions at flowering, seed filling and during flowering and seed filling stages, respectively) were located in main plots and cultivars (Clark, Williams and L_{17}) were allocated to sub plots.

Seeds of soybean cultivars were treated with 2 g kg^{-1}Benomyl and then were sown by hand on 11 May 2011 in 5 cm depth of a sandy loam soil. Seeding density was 60 seeds m^{-2}. Each plot consisted of 6 rows of 5 m length, spaced 25 cm apart. All plots were irrigated immediately after sowing and after seedling establishment, plants were thinned to 45 plants m^{-2}. Subsequent irrigations were carried out on the bases of 70 mm evaporation from class A pan up to flowering. Thereafter, irrigation disruptions were applied according to the treatments. Hand weeding of the experimental area was performed as required.

After seedling establishment, a plant was marked in each pot and Leaf temperature (ºC) and Leaf chlorophyll content index (CCI) of upper, middle and lower leaves were measured. Leaf CCI and was directly measured by a chlorophyll meter (CCM-200, Opti- Science, USA) and Leaf temperature was recorded by an infrared thermometer (TES-1327) every 10 days. Changes in mean CCI and Leaf temperature were shown by regression fits on mean data (Figures 1 and 2).

The chlorophyll fluorescence induction parameters were measured in leaves by a chlorophyll fluorometer (OS-30, OPTISCIENCES, USA) at three stages of plants development (vegetative, flowering and grain filling). Fluorescence emission was monitored from the upper surface of the leaves. Dark-adapted leaves (30 min.) were initially exposed to the weak modulate measuring beam, followed by exposure to saturated white light to estimate the initial (F_0) and maximum (Fm) fluorescence values, respectively. Variable fluorescence (Fv) was calculated by subtracting F_0 from Fm. The quantum yield (Fv/Fm) measures the efficiency of excitation energy capture by open PSII reaction centers, representing the maximum capacity of light-dependent charge separation in PSII [15, 16].

At maturity, the plants in 1 m^2 of each plot were harvested and grains were detached from the pods. Finally, grains were weighed and grain yield per unit area for each treatment at each replicate was determined.

All the data were analyzed on the basis of the experimental design, using MSTATC and SPSS softwares. The means of each trait were compared according to Duncan multiple range test at P≤0.05. Excel software was used to draw figures.

RESULTS AND DISCUSSION

Regression curves showed that chlorophyll content index (CCI) under all irrigation treatments increased with proceeding plant growth, up to about 75 days after sowing and then CCI decreased with further plant development. Water limitation, particularly water disruption during flowering and grain filling stages (I_4) led to severe reductions in maximum CCI of plants (Figure 1a). Leaf CCI of Clark and Williams during vegetative stages was similar. However, during reproductive stags CCI of Williams was higher than that of Clark. Leaf CCI of L_{17} at the early stages of growth was slightly higher than that of other cultivars, while at later stages CCI of Clark and Williams was higher than that of L_{17}. Maximum CCI for L_{17} observed at 68 days after sowing, but for Clark and Williams, it was obtained at 80 days after sowing (Figure 2b).

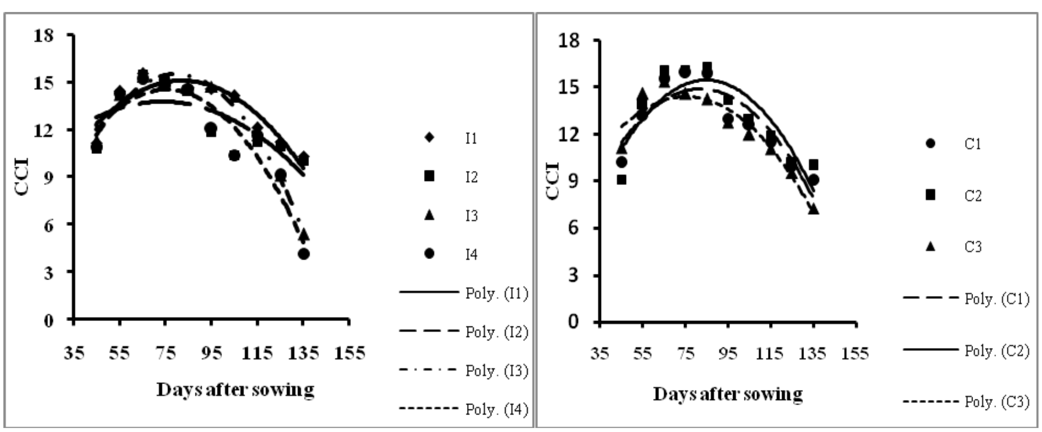

Figure 1. Changes in leaf chlorophyll content index (CCI) of soybean for different irrigation treatments and cultivars.

I_1, I_2, I_3 and I_4: well watering and irrigation disruption at flowering, grain filling and during flowering and grain filling stages, respectively (a). C_1, C_2 and C_3: Clark, Williams and L_{17}, respectively (b).

Reduction in chlorophyll content index (CCI) under water stress (Figure 1) could be related to increasing damage to chloroplasts by active oxygen species, pigment photo-oxidation and chlorophyll degradation [17, 18, 19, 5]. Also, this may be partially resulted from low nutrient uptake. Reduction in leaf chlorophyll content can limit light absorption, stomatal conductivity and photosynthesis rate [20]. Similar results were reported for pinto bean [21], soybean [19] and sedum [22]. Differences in CCI among cultivars (Figure 1b) indicate that this trait can also be influenced by genetic constitution.

At the most stages of vegetative growth, leaf temperature for all irrigation treatments and cultivars was almost similar. However, leaf temperature also increased with increasing water stress duration at reproductive stages. The highest and the lowest leaf temperatures were recorded for irrigation disruption during flowering and grain filling stages (I_4) and well watering (I_1), respectively (Figure 2a). Leaf temperatures of Williams and L_{17} were generally lower and higher than those of other cultivars, respectively (Figure 2b).

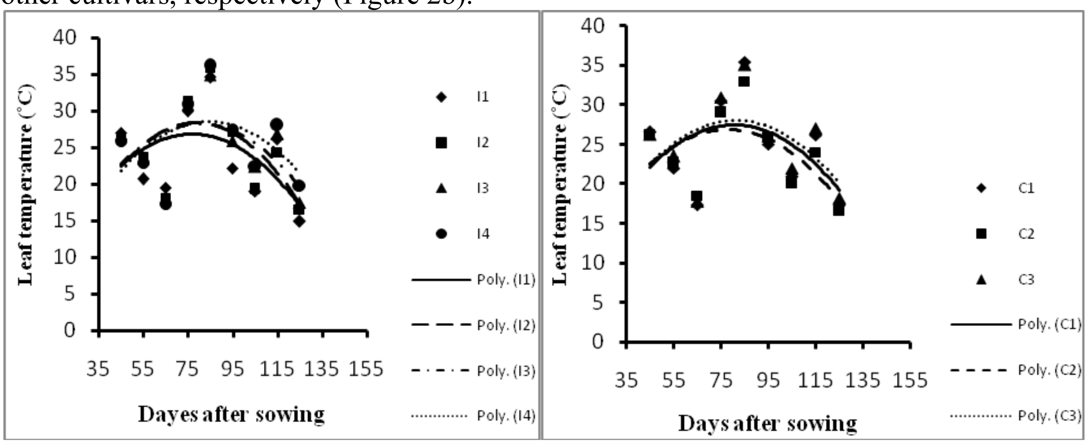

Figure 2. Changes in leaf temperature of soybean for different irrigation treatments and cultivars. I_1, I_2, I_3 and I_4: well watering and irrigation disruption at flowering, grain filling and during flowering and grain filling stages, respectively (a). C_1, C_2 and C_3: Clark, Williams and L_{17}, respectively (b).

Increasing leaf temperature due to water stress during reproductive stages (Figure 2) is possibly related to decreasing stomatal conductance and transpiration [14, 13, 23]. This may be inhibits photosynthesis by limiting the availability of CO_2 within the leaf [24, 25] and predispose leaves to photo-inhibition [26] and decrease photosynthetic efficiency by stimulating photorespiration [27].

Analysis of the data (Table 1) showed that water stress had significant effects on photochemical efficiency (quantum yield, Fv/Fm) and grain yield per unit area. Grain yield per unit area was also significantly affected by cultivars. The interaction of irrigation × cultivar was not significant for Fv/Fm and grain yield per unit area.

Table 1.Analysis of variance of the data for chlorophyll fluorescence (quantum yield, Fv/Fm) and grain yield per unit area of soybean cultivars under different irrigation treatments.

Source	df	Fv/Fm	Grain yield
Replication	3	0.010	662.607
Irrigation (I)	3	0.091*	41527.593**
E_a	9	0.013	1776.241
Cultivar (C)	2	0.011	2549.868*
I*C	6	0.020	604.187
E_b	24	0.010	649.909
Total	47		
CV %		6.59	27.32

*,** Significant at $p \leq 0.05$ and $p \leq 0.01$, respectively

The quantum yield (Fv/Fm) and grain yield were reduced under water stress at reproductive stages. The Fv/Fm ratio during well-watering and water stress during flowering stage (I_2) was statistically similar. Maximum loss in grain yield was observed under severe water stress (I_4). However, grain yield per unit area under I_2 and I_3 was not significant. The Fv/Fm was not different among cultivars. Grain yield per unit area of Williams was significantly higher than that of other cultivars, but grain yield for Clark and Williams was not statistically different (Table 2).

Table 2. Means of the chlorophyll fluorescence (quantum yield, Fv/Fm) and grain yield per unit area of soybean for irrigation treatments and cultivars.

Treatment	Fv/Fm	Grain yield (g m^{-2})
Irrigation		
I_1	0.7801 a	198.42 a
I_2	0.7372 a	127.93 b
I_3	0.6128 b	116.28 b
I_4	0.6087 b	54.87 c
Cultivar		
C_1	0.702 a	120.11 ab
C_2	0.698 a	138.77 a
C_3	0.654 a	114.71 b

Different letter in each column indicate significant difference at $p \leq 0.05$

I_1, I_2, I_3 and I_4: well-watering and irrigation disruption at flowering, grain filling and during flowering and grain filling, respectively.

Chlorophyll fluorescence analysis is a sensitive indicator of the tolerance of the photosynthetic apparatus to environmental stress [9]. Reduction in quantum yield of the PSII (Fv/Fm) under water stress at reproductive stages (Table 2), indicate that occurrence of chronic photo-inhibition due to photo-inactivation of PSII centers probably associated with the degradation of D_1 protein [28, 29]. Various studies reported the Fv/Fm ratio is an indicator of stress. Similarly, some researchers showed that Fv/Fm reduces during water stress [30, 31].

The superiority of Williams in grain yield per unit area could be attributed to higher CCI and lower leaf temperature of this cultivar, compared with other cultivars (Figures 1b, 2b). Therefore leaf temperature and chlorophyll content can be used as reliable criterion in selection of water stress tolerant soybean cultivars. Large reductions in grain yield per unit area clearly show that soybean is a sensitive plant to water stress at reproductive stages, but the extent of this sensitivity varies among cultivars (Table 2).

CONCLUSION

Water stress during reproductive stages can significantly influence leaf chlorophyll content, leaf temperature and quantum yield of the PSII (Fv/Fm). These parameters are sensitive indicators of water stress, which are closely related with grain yield. Williams is comparatively tolerant cultivar to water stress.

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