



RESEARCH PAPER

OPEN ACCESS

Some physiological responses of mung-bean at different plant densities to water deficit

Kazem Ghassemi-Golezani*, Salar Farhanghi Abriz, Sirous Hassannejad, Saeid Hassanpour-Bourkheili

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

Key words: Fluorescence, Grain yield, Leaf area index, Mung-bean, Water stress

<http://dx.doi.org/10.12692/ijb/4.12.19-26>

Article published on June 22, 2014

Abstract

A large part of the surface of the world is arid. Water deficit is the major abiotic factor limiting plant growth and crop productivity around the world. A field experiment was carried out as split plot based on RCB design with three replications in 2013 to assess the effects of different irrigation treatments (I₁, I₂, I₃, I₄ for irrigation after 70, 100, 130 and 160 mm evaporation from class A pan, respectively) and different plant densities (D₁, D₂ and D₃ with 30, 50 and 70 plants/m²) on some physiological traits and grain yield of mung-bean. Leaf area index, chlorophyll fluorescence of the PSII (Fv/Fm) and grain yield significantly decreased as water deficit increased. LAI was statistically similar under I₃ and I₄. LAI increased with increasing plant density up to 50 plants/m², with no significant change thereafter. Leaf temperature significantly increased with increasing plant density under severe water stress (I₄), however no significant differences among densities were observed under I₁, I₂ and I₃. Fv/Fm slightly, but not significantly, decreased as plant density increased. Increasing leaf temperature and decreasing Leaf area index (LAI) and photosystem efficiency under water stress led to significant reduction in grain yield per unit area. The highest grain yield was obtained at 50 plants/m².

* **Corresponding Author:** Kazem Ghassemi-Golezani ✉ golezani@gmail.com

Introduction

In a large part of the agricultural areas in the world, water deficit is an important factor limiting growth and productivity of the crops (Borsani *et al.*, 2001; Micheletto *et al.*, 2007). Water deficit occurs when water potentials in the rhizosphere are sufficiently negative to reduce water availability to sub-optimal levels for plant growth and development (Boyer 1982). To survive against the stress, plants have involved a number of morphological, physiological and biochemical responses (Demirevska *et al.*, 2009; Munns *et al.*, 2006). Photosynthesis and cell growth are the primary processes which are affected by stress (Munns *et al.*, 2006). Under drought stress, plant leaves are dehydrated, and photosynthesis is decreased. The decrease in photosynthesis of dehydrated leaves is usually caused by decrease in stomatal conductance and transpiration (Cornic, 1994; Limousin *et al.*, 2010). As a consequence of the reduction in transpiration rate, leaf temperature increases and the effects of drought and heat stresses frequently combine to scorch leaves (Mohammadian *et al.*, 2005).

All green plants can contribute to primary production by photosynthetic activity in leaves that play a major role in this process. Therefore, adequate leaf area should be critical to plant regeneration for a constant primary production. Ideally, leaf area index (LAI) among crops varies between 2-3 and 11, depending on the morphological and anatomical structure of the plants (Nelson and Sommers, 1995). Reduced canopy photosynthetic rates under conditions of water stress is attributed to decreased leaf expansion, leaf area, leaf age of the canopy and increased senescence rates (Marani *et al.*, 1986).

Stresses that involve deficiencies of N and water will adversely affect the amount of under drought stress, disturbances of photosynthesis at the molecular level are connected with the restricted electron transport through PSII and/or with structural injuries to PSII (Flexas *et al.*, 2004; Hura *et al.*, 2007). Fluorescence of chlorophyll reflected the photochemical activities of PSII (Ganivea *et al.*, 1998), with optimal values of

around 0.832 measured from most plant species (Johnson *et al.*, 1993). Values lower than this are measured when the plant is exposed to stress, indicating a particular phenomenon of photo-damage to PSII reaction centres (Maxwell and Johnson, 2000; Baker and Rosenqvist, 2004). Environmental stresses that affect PSII efficiency leads to a decrease in the Fv/Fm ratio (Krause and Weis 1991; Mamnouie *et al.*, 2006).

The effect of drought stress on growth and yield depends on function of genotype, duration of stress, weather conditions, growth, and developmental stages of crops (Robertson and Holland, 2004). Moderate to high drought stress can reduce plant biomass, number of pods and seeds, days to maturity, harvest index, seed yield and seed weight in common bean (Ghassemi-Golezani and Mardfar, 2008). Plant density is not stable for a variety at different climatic conditions. In a plant community, after a saturation threshold, adding more plants will not increase yield per unit area. On an individual plant basis, as density increases, individual plant yield decreases non-linearly (Roush and Radosevich, 1985). This research was carried out to evaluate changes in leaf area index, fluorescence and leaf temperature in leaves of mung-bean in response to water stress and their consequences to crop yield.

Material and methods

Experimental design

The experiment was conducted at the Research Farm of Tabriz University, Tabriz, Iran (latitude 38.05°N, longitude 46.17°E, Altitude 1360 m above sea level) in 2013. The climate is characterized by mean annual precipitation of 245.75 mm per year and mean annual temperature of 10°C. The experiment was arranged as split plot on the basis of randomized complete block in three replicates, with irrigation treatments (I₁, I₂, I₃, I₄ for irrigation after 70, 100, 130 and 160 mm evaporation from class A pan, respectively) in main plots and plant densities (D₁=30, D₂=50 and D₃=70 plants/ m²) in sub plots.

Sowing

Seeds of Mung-bean were treated with 2 g kg⁻¹ Benomyl and then were sown by hand in 4 cm depth of a sandy loam soil. Each plot consisted of 6 rows of 3.5 m length, spaced 25 cm apart. All plots were irrigated immediately after sowing. Subsequent Irrigation treatments were applied after seedling establishment. Weeds were controlled by hand during crop growth and development as required.

Measurements

A plant was marked in each pot and Leaf temperature (°C) of upper, middle and lower leaves were measured by an infrared thermometer (TES-1327) at flowering. At this stage, three plants from each treatment were harvested and leaf area of the mung-bean were measured by a portable Leaf area- meter (ADCAM300). The chlorophyll fluorescence induction parameters were measured in leaves by a chlorophyll fluorometer (OS-30, OPTISCIENCES, USA) at flowering just before irrigation of each plot. 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 (F₀) and maximum (F_m) fluorescence values, respectively. Variable

fluorescence (F_v) was calculated by subtracting F₀ from F_m. The quantum yield (F_v/F_m) measures the efficiency of excitation energy capture by open PSII reaction centres, representing the maximum capacity of light-dependent charge separation in PSII (Rizza *et al.*, 2001; Basu, 2004).

At maturity, the plants in 1 m² 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 analysed 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

Analyses of variance

Analysis of the data (Table 1) showed that water stress had significant effects on leaf temperature, Leaf area index (LAI), chlorophyll fluorescence and grain yield. Grain yield was also significantly affected by plant density. The interaction of irrigation × density was only significant for leaf temperature.

Table 1. Analysis of variance of the effects of water stress on some physiological characters and grain yield of mung-bean at different plant densities.

Source	df	LAI	Temperature	Fv/Fm	Grain yield
Replication	2	0.014	1.243	0.025*	487.659
Irrigation(I)	3	0.455**	21.392**	0.133**	2623.419*
Ea	6	0.011	0.244	0.002	299.463
Density (D)	2	0.122**	0.738**	0.009*	15434.591**
I × D	6	0.012	0.235*	0.006	133.346
Eb	16	0.009	0.079	0.017	50.802
CV%		4.9	1.16	4.51	4.18

*, **: Significant at p≤0.05 and p≤0.01, respectively.

Leaf area index (LAI)

LAI decreased with increasing irrigation intervals, but no significant difference was observed between I₃ and I₄ (Table 2). Leaf area is a critical parameter controlling many biological and physical processes associated with vegetation (Running, 1990; Bonan, 2003). Water stress reduces photosynthesis by decreasing leaf area (McCree, 1986). At the crop level

the demand for water as affected by plant size is controlled by leaf area index (LAI), which is the total area of live leaves per unit ground surface. Photosynthesis by crops is severely inhibited and may cease altogether as water deficits increase. The decrease in leaf growth, or increasing senescence of leaves under drought conditions, may also inhibit photosynthesis in existing leaves (David *et al.*, 1998).

LAI increased with increasing plant density up to 50 plants/m², with no significant change up to 70 plants/m² (Table 2). Leaf area index (LAI) is one of the most important crop parameters that determine radiation intercepted by the crop canopy, and therefore has strong impacts on crop canopy

photosynthesis and transpiration (Serdar and Demirsoy, 2006). Increase in leaf area more than usual, causing competition for light. Any change in canopy leaf area index is accompanied by modifications in crop productivity.

Table 2. Means LAI, leaf temperature, chlorophyll fluorescence and grain yield of mung-bean for different irrigation treatments and plant densities.

Treatments	LAI	Temperature (°C)	Fv/Fm	Grain yield (g/m ²)
Irrigation				
I ₁	2.301 a	23.25 c	0.7791 a	188.83 a
I ₂	1.983 b	23.57 b	0.7545 a	178.88 b
I ₃	1.872 c	23.72 b	0.7015 ab	163.59 c
I ₄	1.787 c	26.57 a	0.620 b	149.99 d
Density				
D ₁	1.870 b	24.06 b	0.7285 a	129.19 c
D ₂	2.034 a	24.23 b	0.7215 a	195.07 a
D ₃	2.054 a	24.55 a	0.6918 a	186.71 b

Different letters in each column indicate significant difference at $P \leq 0.05$.

I₁, I₂, I₃, I₄: Irrigation after 70, 100, 130 and 160 mm evaporation from class A pan, respectively

D₁, D₂ and D₃: 30, 50 and 70 plant/m², respectively.

Leaf temperature

Mean leaf temperature increased with increasing water stress. However, leaf area index, chlorophyll fluorescence of the PSII (Fv/Fm) and grain yield decreased as water deficit increased (Table 2). Increasing leaf temperature due to water stress is possibly related to decreasing stomatal conductance and transpiration (Ehrlér *et al.*, 1978; Reginato, 1983; Siddiaue *et al.*, 2000).

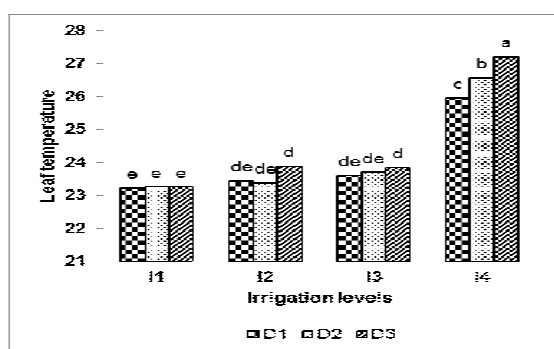


Fig. 1. Changes in mean leaf temperature of mung-bean at different plant densities under different irrigation treatments. Different letters at each column for each treatment indicate significant difference at $p \leq 0.05$. I₁, I₂, I₃, I₄: Irrigation after 70, 100, 130 and 160 mm evaporation from class A pan, respectively. D₁, D₂ and D₃: 30, 50 and 70 plant/m², respectively.

Leaf temperature significantly increased with increasing plant density under severe water stress (I₄), but no significant differences among densities were observed under I₁, I₂ and I₃ (Figure 1). With water availability limitation, stomatal conductance and transpiration decrease and leaf temperature increases (Reginato, 1983). This may inhibit photosynthesis by limiting the availability of CO₂ within the leaf (Boyer, 1976; Chaves, 1991) and predispose leaves to photo-inhibition (Bjorkman and Powles, 1996) and decrease photosynthetic efficiency by stimulating photorespiration (Brooks and Farquhar, 1985).

Chlorophyll fluorescence

Reduction in quantum yield of the PSII (Fv/Fm) under water stress (Table 2) indicate that occurrence of chronic photo-inhibition due to photo-inactivation of PSII centers probably associated with the degradation of D1 protein (He *et al.*, 1995; Giardi *et al.*, 1996). Despite of the fact that photosystem II is highly drought resistant, photosynthetic electron transport through PSII is inhibited by water stress. The Fv/Fm ratio characterizes the maximal quantum

yield of the primary photochemical reaction in dark adapted leaves (Debaeke and Aboudrare, 2004).

Fv/Fm slightly, but not significantly, decreased as plant density increased (Table 2). It has been well documented that the photosynthetic system is very sensitive to many environmental stresses and that chlorophyll fluorescence analysis is a good index for measuring rapidly the change in photosynthetic metabolism of plants to such environmental stresses as drought (Conroy *et al.*, 1986; Genty *et al.*, 1987). PSII has been shown to be the most thermally labile component of the electron transport chain and the critical site of damage by high temperatures (Allakhverdiev *et al.*, 2008). An important implication of PSII photo inactivation is that non-functional PSII centers, still embracing pigment molecules, can exacerbate photo oxidative damage to the thylakoid membranes unless light energy absorbed by the pigments is dissipated safely. Thus, it has been hypothesized that photo inactivated PSII complexes are able to efficiently dissipate excitation energy harmlessly (Krause, 1988).

Grain yield

Grain yield of mung-bean decreased with increasing water stress, possibly due to low Leaf area index (LAI) and low photosystem efficiency (Table 2). These may result in pod and flower abortion and reduction in mean grain number and weight (Ghassemi-Golezani *et al.*, 2012; Ghassemi-Golezani and Lotfi, 2012). These results clearly show that supplementary irrigation can considerably improve physiological performance and grain yield of mung-bean.

The highest grain yield was obtained at D₂ (50 plants/m²), followed by D₃ (70 plants/m²) and D₁ (30 plants/m²), respectively. The lowest grain yield per unit area at 30 plants/m² mainly associated with the lowest LAI at this density (Table 2). Decreasing grain yield at the highest density (D₃) strongly related with increasing competition for water and other environmental resources available to each plant, particularly under severe water stress. This is also reflected in significantly higher leaf temperature of D₃

plants under the most limited water supply (I₄) (Figure 1). Hence, to reach higher yields, proper plant density is critical in arid and semi-arid regions.

References

- Allakhverdiev SI, Kreslavski VD, Klimov VV, Los DA, Carpentier R, Mohanty P.** 2008. Heat stress: an overview of molecular responses in photosynthesis. *Photosynth Research* **98**, 541–50.
- Baker NR, Rosenqvist E.** 2004. Application of chlorophyll fluorescence can improve crop production strategies: an examination of future possibilities. *Journal of Experimental Botany* **55**, 1607-1621. <http://dx.doi.org/10.1093/jxb/erh196>
- Basu PS, Masood A, Chaturvedi SK.** 2004. Adaptation of photosynthetic component of chickpea to water stress. *International Crop Science Congress*. Brisbane, Australia.
- Bjorkman O, Powles SB.** 1994. Inhibition of photosynthetic reactions under water stress interaction with light level. *Planta*, **161**, 490-504. <http://dx.doi.org/10.1007/BF00407081>
- Bonan GB, Levis S, Sitch S, Vertenstein M, Oleson KW.** 2003. A dynamic global vegetation model for use with climate models: Concepts and description of simulated vegetation dynamics, *Global Change Biology* **9**, 1543-1566.
- Borsani O, Valpuesta V, Botella MA.** 2001. Evidence for a role of salicylic acid in the oxidative damage generate by NaCl and osmotic stress in Arabidopsis seedling. *Plant Physiology* **126**, 1024-1030.
- Boyer JS.** 1976. Water deficits and photosynthesis. In T. T. Kozlowski (ed.), *Water Deficits and Plant Growth*, Academic press, New York. 153- 190 p.
- Boyer JS.** 1982. *Plant Productivity and Environment Science* **218**, 443–448.

- Brooks A, Farquhar GD.** 1985. Effects of temperature on the O₂/CO₂ specificity of ribulose-1, 5-bisphosphate carboxylase/oxygenase and the rate of respiration in the light. Estimates from gas exchange measurements on spinach. *Planta* **165**, 397-406.
<http://dx.doi.org/10.1007/BF00392238>
- Chaves MM.** 1991. Effects of water deficits on carbon assimilation. *Journal of Experimental Botany*, **42**, 1-16.
<http://dx.doi.org/10.1093/jxb/42.1.1>
- Conroy JP, Smillie RM, Kupperts M, Bevege DI, Barlow EW.** 1986. Chlorophyll fluorescence and photosynthetic and growth responses of *Pinus radiata* to phosphorus deficiency, drought stress and high CO₂. *Plant Physiology* **81**, 423-429.
- Cornic G.** 1994. Drought stress and high light effects on leaf photosynthesis, in: Baker NR, (ed.). *Photo inhibition of Photosynthesis: From Molecular Mechanisms to the Field*. BIOS, Oxford, 297-313.
- David MM, Coelho D, Barrote I, Correia MJ.** 1998. Leaf age effects on photosynthetic activity and sugar accumulation in droughted and re watered *Lupinus alba* plants. *Australian Journal of Plant Physiology* **25**, 299-306.
- Debaeke P, Aboudrare A.** 2004. Adaptation of crop management to water limited Environments. *European Journal of Agronomy* **21**, 433-466.
- Demirevska K, Zasheva D, Dimitrov R, Simova-Stoilova L, Stamenova M, Feller U.** 2009. Drought stress effects on Rubisco in wheat: changes in the Rubisco large subunit. *Acta Physiologiae Plantarum* **31**, 1129- 1138.
- Ehrler WL, Idso SB, Jacleson RD, Reginato RJ.** 1978. Wheat canopy temperature: Relation to plant water potential. *Agronomy Journal* **70**, 251-256.
- Flexas J, Bota J, Loreto F, Cornic G, Sharkey TD.** 2004. Diffusive and metabolic limitations to photosynthesis under drought and salinity in C₃ plants. *Plant Biology* **6**, 269-279.
- Ganivea RA, Allahverdiyev SR, Guseinova NB, Kavakli HI, Nafisi S.** 1998. Effect of salt stress and synthetic hormone polystimuline K on the photosynthetic activity of cotton (*Gossypium hirsutum*). *Turkish Journal of Botany* **22**, 217-221.
- Genty B, Briantais JM, Da Silva JBV.** 1987. Effects of drought on primary photosynthetic processes of cotton leaves. *Plant Physiology* **83**, 360-364.
<http://dx.doi.org/10.1104/pp.83.2.360>
- Ghassemi-Golezani K, Mardfar RA.** 2008. Effects of limited irrigation on growth and grain yield of common bean. *Journal of Plant Science* **3**, 230-235.
<http://dx.doi.org/10.3923/jps.2008.230.235>
- Ghassemi-Golezani K, Lotfi R.** 2012. Responses of soybean leaves and grain yield to stress at reproductive stages cultivars. *International Journal of plant, animal and environmental sciences* **3**, 63-68.
- Ghassemi-Golezani K, Mustafavi SH, Shafagh-Kalvanagh J.** 2012. Field performance of chickpea cultivars in response to irrigation disruption at reproductive stages. *Research on Crops* **13**, 107-112.
- Giardi MT, Cona A, Gieken B, Kucera T, Masojidek J, Matto AK.** 1996. Long-term drought stress induces structural and functional reorganization of photosystem II. *Planta*, **199**: 118-125.
<http://dx.doi.org/1007/BF00196888>
- He JX, Wang J, Liang HG.** 1995. Effects of water stress on photochemical function and protein metabolism of photosystem II in wheat leaves. *Physiologia Plantarum* **90**, 771-777.
<http://dx.doi.org/10.1111/j.13993054.1995.tb05130.x>
- Flexas J, Bota J, Loreto F, Cornic G, Sharkey**

- Hura T, Hura K, Grzesiak M, Rzepka A.** 2007. Effect of long-term drought stress on leaf gas exchange and fluorescence parameters in C₃ and C₄ plants. *Acta Physiologiae Plantarum* **29**, 103-113.
<http://dx.doi.org/10.1007/s11738-006-0013-2>
- Johnson GN, Young AJ, Scholes JD, Horton P.** 1993. The dissipation of excess excitation energy in British plant species. *Plant, Cell and Environment*, **16**, 673-679.
<http://dx.doi.org/10.1111/j.13653040.1993.tb00485.x>
- Krause GH, Weis E.** 1991. Chlorophyll fluorescence and photosynthesis: the basis. *Annual Review of Plant Physiology and Plant Molecular Biology* **42**, 313-349.
<http://dx.doi.org/10.1146/annurev.pp.42.060191.001525>
- Krause GH.** 1988. Photo inhibition of photosynthesis. An evaluation of damaging and protective mechanisms. *Physiological Plant*, **74**, 566-574.
- Limousin J, Misson L, Lavoit A, Martin NK, Rambal S.** 2010. Do photosynthetic limitations of evergreen *Quercus ilex* leaves change with long-term increased drought severity. *Plant, Cell and Environment* **33**, 863-875.
<http://dx.doi.org/10.1111/j.1365-3040.2009.02112.x>
- Mamnouie E, Fotouhi-Ghazvini R, Esfahany M, Nakhoda B.** 2006. The effects of water deficit on crop yield and the physiological characteristics of barley (*Hordeum vulgare* L.) varieties. *Journal of Agricultural Science and Technology* **8**, 211-219.
- Marani A, Amirav A.** 1971. Effects of soil moisture stress on two varieties of upland cotton. The Coastal Plain Region. *Experimental Agriculture* **7**, 213-224.
- Maxwell K, Johnson GN.** 2000. Chlorophyll fluorescence-a practical guide. *Journal of Experimental Botany* **51**, 659-668.
- McCree KJ.** 1986. Whole-plant carbon balance during osmotic adjustment to drought and salinity stress. *Australian Journal of Plant Physiology* **13**, 33-43.
- Micheletto S, Rodriguez-Urbe L, Hernandez R, Richins RD, Curry V, Connell MA.** 2007. Comparative transcript profiling in roots of (*Phaseolus acutifolius*) and (*Phaseolus vulgaris*) under water deficit stress. *Plant Science* **173**, 510-520.
- Mohammadian R, Moghaddam M, Rahimian H, Sadeghian SY.** 2005. Effect of early season drought stress on growth characteristics of sugar beet genotypes. *Turkish Journal of Agriculture and Forestry* **29**, 357-68.
- Munns R, James AJ, Lauchli A.** 2006. Approaches to increasing the salt tolerance of wheat and other cereals. *Journal of Experimental Botany*, **57**, 1025-1043.
<http://dx.doi.org/10.1093/jxb/erj100>
- Nelson DW, Sommers LE.** 1995. Morphology and Systematics. In: R.F. Barnes, D.A. Miller and C.J. Nelson (eds.). *Forages*. Iowa State University Press. Ames, Iowa, USA. 15-30 p.
- Reginato RJ.** 1983. Field quantification of crop water stress. *Transaction of ASABE* **26**, 772-775.
- Rizza F, Pagani D, Stanca AM, aCattivelli L.** 2001. Use of chlorophyll fluorescence to evaluate the cold acclimation and freezing tolerance of winter and spring oats. *Plant Breeding* **120**, 389-396.
<http://dx.doi.org/10.1046/j.1439-0523.2001.00635.x>
- Robertson MJ, Holl, JF.** 2004. Production risk of canola in the semi-arid subtropics of Australia. *Australian Journal of Agriculture Research* **55**, 525-538.
- Roush LM, Radosevich SR.** 1985. Relationship between growth and competitiveness of four annual weeds. *Journal of Applied Ecology* **22**, 895-905.

Running SW. 1990. A bottom-up evolution of terrestrial ecosystem modeling theory, and ideas toward global vegetation modeling. In *Modeling the Earth System*. UCAR/Office for Interdisciplinary Earth Studies, Boulder (1990), 263-280 p.

Serdar U, Demirsoy H. 2006. Non-destructive leaf area estimation in chestnut. *Science of Horticulture*, **108**, 227-230.

Siddiaue MRB, Hamid A, Islam MS. 2000. Drought stress effects on water relations of wheat. *Botanical Bulletin of Academia Sinica*, **41**, 35-39.