



Research Article

Effect of Drought stress on Protein Contents, Respiration and Heat Shock Proteins in Crop Plants

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ABSTRACT

Drought stress is one of the most important abiotic stress factors which are generally accompanied by heat stress in dry season. Water deficit stress due to drought, salinity or extremes in temperature is the main limiting factors for plant growth and productivity resulting in large economic losses in many regions of the world. Plants can partly protect themselves against mild drought stress by accumulating osmolytes. Proline is one of the most common compatible osmolytes in drought stressed plants. For example, the proline content increased under drought stress in pea. Drought tolerance is a cost-intensive phenomenon, as a considerable quantity of energy is spent to cope with it. The fraction of carbohydrate that is lost through respiration determines the overall metabolic efficiency of the plant. Decreasing water availability under drought generally results in limited total nutrient uptake and their diminished tissue concentrations in crop plants. An important effect of water deficit is on the acquisition of nutrients by the root and their transport to shoots.

Key words: Antioxidation strategies, Glycine betaine, Nutrient relations, Stomatal conductance

INTRODUCTION

Water is essential at every stage of plant growth and agricultural productivity is solely dependent upon water and it is essential at every stage of plant growth, from seed germination to plant maturation (Turner, 1991). Drought stress is one of the most important abiotic stress factors which are generally accompanied by heat stress in dry season (Dash and Mohanty, 2001). Water deficit stress due to drought, salinity or extremes in temperature is the main limiting factors for plant growth and productivity resulting in large economic losses in many regions of the world (Borsani *et al.*, 2001). Plants respond to water stress through a number of biochemical, physiological and developmental changes (Pattanagul, 1999; Shinozaki, 1997). Drought is perceived as the most significant environmental stress in agriculture worldwide, and improving yield under drought is therefore a major goal of plant breeding (Cattivelli *et al.*, 2008). With a projected increase in drought with climate change, the breeding for drought-tolerant crops is even more emphasised (Witcombe *et al.*, 2008).

Protein contents

Plants can partly protect themselves against mild drought stress by accumulating osmolytes. Proline is one of the most common compatible osmolytes in drought

stressed plants. For example, the proline content increased under drought stress in pea (Sanchez *et al.*, 1998; Alexieva *et al.*, 2001). Proline accumulation can also be observed with other stresses.

Respiration

Drought tolerance is a cost-intensive phenomenon, as a considerable quantity of energy is spent to cope with it. The fraction of carbohydrate that is lost through respiration determines the overall metabolic efficiency of the plant (Davidson, 2000). The root is a major consumer of carbon fixed in photosynthesis and uses it for growth and maintenance, as well as dry matter production (Lambers, 1996). Plant growth and developmental processes as well as environmental conditions affect the size of this fraction (i.e. utilized in respiration). However, the rate of photosynthesis often limits plant growth when soil water availability is reduced (Huang and Fu, 2000). A negative carbon balance can occur as a result of diminished photosynthetic capacity during drought, unless simultaneous and proportionate reductions in growth and carbon consumption take place. In wheat, depending on the growth stage, cultivar and nutritional status, more than 50% of the daily accumulated photosynthates were transported to the root, and around 60% of this fraction was respired (Lambers, 1996).

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Nutrient relations

Decreasing water availability under drought generally results in limited total nutrient uptake and their diminished tissue concentrations in crop plants. An important effect of water deficit is on the acquisition of nutrients by the root and their transport to shoots. Lowered absorption of the inorganic nutrients can result from interference in nutrient uptake and the unloading mechanism, and reduced transpirational flow (Garg, 2003; McWilliams, 2003). However, plant species and genotypes of a species may vary in their response to mineral uptake under water stress. In general, moisture stress induces an increase in N, a definitive decline in P and no definitive effects on K (Garg, 2003). Transpiration is inhibited by drought, as shown for beech (Peuke, 2002), but this may not necessarily affect nutrient uptake in a similar manner. Influence of drought on plant nutrition may also be related to limited availability of energy for assimilation of $\text{NO}_3^-/\text{NH}_4^+$, PO_4^{3-} and SO_4^{2-} : they must be converted in energy-dependent processes before these ions can be used for growth and development of plants (Grossman and Takahashi, 2001). As nutrient and water requirements are closely related, fertilizer application is likely to increase the efficiency of crops in utilizing available water. This indicates a significant interaction between soil moisture deficits and nutrient acquisition. Studies show a positive response of crops to improved soil fertility under arid and semi-arid conditions. Currently, it is evident that crop yields can be substantially improved by enhancing the plant nutrient efficiency under limited moisture supply (Garg, 2003). It was shown that N and K uptake was hampered under drought stress in cotton (McWilliams, 2003).

Stomatal conductance

Plants grown under drought condition have a lower stomatal conductance in order to conserve water. Consequently, CO_2 fixation is reduced and photosynthetic rate decreases, resulting in less assimilate production for growth and yield of plants. Diffusive resistance of the stomata to CO_2 entry probably is the main factor limiting photosynthesis under drought (Boyer, 1970). Certainly under mild or moderate drought stress stomatal closure (causing reduced leaf internal CO_2 concentration (Ci)) is the major reason for reduced rates of leaf photosynthetic (Chaves, 1991; Cornic, 2000; Flexas *et al.*, 2004). Varieties significantly differed in photosynthetic activities, but these differences could only be expressed under the control conditions. In many experiments it has been shown that A decreases when gs decreases (e.g., Tenhunen *et al.*, 1987; Nilsen and Orcutt, 1996). Chaves and Oliveira (2004) concluded that gs only affect A at severe drought stress. The decrease in photosynthesis in drought stressed plants can be attributed both to stomatal (stomatal closure) and non-stomatal (impairments of metabolic processes) factors. Under control treatment, the yield of cultivars followed the same trend of A, under this condition 'Bivaniej' showed highest A and seed yield. At present most researchers agree that the stomatal closure and the resulting CO_2 deficit in the chloroplasts is the main cause of decreased photosynthesis under mild and moderate stresses (Flexas and Medrano, 2002).

Glycine betaine content

Drought stressed shallot plants showed an increase in glycine betaine content when compared to control. The glycine betaine content increased under drought stress in *Radix astragali* (Tan, 2006), in barley (Nakamura, 2001) and in higher plants (Jun, 2000). Glycine betaine is considered to be one of the most abundant quaternary ammonium compounds produced in higher plants under stressful environment (Yang, 2003). Glycine betaine has been shown to protect the enzymes and membranes and also to stabilize PSII protein pigment complexes under stressful conditions (Papageorgiou and Morata, 1995).

Heat shock proteins (Hsps)

Chen and Wang, 2003; Zhu and Zhang, 2003; Xie *et al.*, 2005 founded that the synthesis of some original proteins (namely stress-induced proteins) may be induced or up regulated to adjust osmotic potential of cells in order to keep a certain turgor and thus to ensure the normal proceeding of physiological processes such as cell growth, stomatal opening and photosynthesis it can concluded that to cope with environmental stress, plants activate a large set of genes leading to the accumulation of specific stress-associated proteins (Vierling, 1991; Ingram and Bartels, 1996; Bohnert and Sheveleva, 1998; Thomashow, 1999; Hoekstra *et al.*, 2001). Heat-shock proteins (Hsps) and late embryogenesis abundant (LEA)-type proteins are two major types of stress-induced proteins that accumulate upon water, salinity, and extreme temperature stress. They have been shown to play a role in cellular protection during the stress (Bakalova *et al.* 2008; Thomashow, 1998).

Antioxidation strategies

Drought stress is accompanied by the formation of ROS such as O_2 , H_2O_2 , and OH (Moran *et al.* 1994 ; Mittler 2002), which damage membranes and macromolecules. Plants have developed several antioxidation strategies to scavenge these toxic compounds. Enhancement of antioxidant defense in plants can thus increase tolerance to different stress factors. Antioxidants (ROS scavengers) include enzymes such as catalase, superoxide dismutase (SOD), ascorbate peroxidase (APX) and glutathione reductase, as well as non-enzyme molecules such as ascorbate, glutathione, carotenoids, and anthocyanins. Additional compounds, such as osmolytes, proteins can also function as ROS scavengers (Bowler *et al.* 1992; Noctor and Foyer 1998). The antioxidant defenses appear to provide crucial protection against oxidative damage in cellular membranes and organelles in plants grown under unfavorable conditions (Al- Ghamdi, 2009; Kocsy *et al.*, 1996). Plant cells synthesize a variety of antioxidants to cope with ROS produced under normal and stress conditions (Noctor and Foyer, 1998).

Chlorophylls

Drought stress produced changes in the ratio of chlorophyll 'a' and 'b' and carotenoids (Anjum *et al.*, 2003b; Farooq *et al.*, 2009). A reduction in chlorophyll content was reported in drought stressed cotton (Massacci *et al.*, 2008) and *Catharanthus roseus* (Jaleel *et al.*, 2008a-d). The chlorophyll content decreased to a

significant level at higher water deficits in sunflower plants (Kiani *et al.*, 2008) and in *Vaccinium myrtillus* (Tahkokorpi *et al.*, 2007). The foliar photosynthetic rate of higher plants is known to decrease as the relative water content and leaf water potential decreases (Lawlor & Cornic, 2002). However, the debate continues as, whether drought mainly limits photosynthesis through stomatal closure or through metabolic impairment (Lawson *et al.*, 2003; Anjum *et al.*, 2003b). Both stomatal and non-stomatal limitation was generally accepted to be the main determinant of reduced photosynthesis under drought stress (Farooq *et al.*, 2009). The limitation of photosynthesis under drought through metabolic impairment is more complex phenomenon than stomatal limitation and mainly it is through reduced photosynthetic pigment contents in sunflower (Reddy *et al.*, 2004). Chlorophyll b content increased in two lines of okra, whereas chlorophyll a remained unaffected resulting in a significant reduction in Chl a: b ratio in both cultivars under water limiting regimes (Estill *et al.*, 1991; Ashraf *et al.*, 1994).

MATERIALS AND METHODS

This article is review and the aims of effect of drought stress on protein contents, respiration and heat shock proteins in crop plants. The experiment 1 was conducted by forouzandeh *et al.* (2014). In this experiment the field experiment was conducted in 2013, 2014 growing seasons at the Agricultural Research Institute of University of zabol, Iran. The experiment was arranged complete randomized block in factorial design with three replications (forouzandeh *et al.*, 2014). The soil texture was sandy-loam, having 1.1% organic matter. Soil chemical analysis was as follows: pH = 7.7; ECdS/m) = 2.4; cations (meq/L): Ca+2 = 2.43, Mg+2 = 2.5, Na+ = 6.46, K+ = 2.74; anions (meq/L): CO₃-2 = zero, HCO₃- = 3.6, Cl- = 2.4, SO₄ 2- = 5.6 (Jackson, 1973). The experimental plot size was 2 meters long and 2 meters width, occupying an area of 4 m² and Seeds were planted on 14 December, 2014 in 40 cm row distance, 1.5 cm sowing depth. (I1: two times irrigation, I2: three times irrigation and I3: four times irrigation that are irrigation in germination, seedling, flowering and seed filing stages) and fertilizers treatment (T1: without fertilizer application (Control), T2: 10 t/ha vermicompost, T3: 15 t/ha compost and T4: 30 t/ha animal manure. The characteristics such as Biological yield (kg/ha), Total yield (kg/ha), Harvest Index, Essential oil yield (kg/ha) and Essential oil percentage by Clevenger were evaluated (forouzandeh *et al.*, 2014). The studied traits were measured on the 10 randomly selected. Weeds were controlled by hand weeding during crop growth and development. At maturity, plants of 2 m² in the middle part of each plot were harvested and calculated. All data were averaged and statistically analyzed using analysis of variance (ANOVA) by MSTATC and SAS analytical software. The Duncan's multiple range test level was used to compare means (forouzandeh *et al.*, 2014).

The experiment 2 was conducted by Nazariyan *et al.* (2009). In this experiment the study was carried out as a split-plot experiment based on a Randomized Complete Block Design with three replications at Agriculture and Natural Resources Research Center, Zanjan, Iran in 2009.

The main plot factor (stress treatment) included four levels (no-water stress, stress at head formation, stress at flowering stage and stress at grain filling stage) and the sub-plot factor (cultivars) included Master, Lakomka, Euroflour and Azargol (Nazariyan *et al.*, 2009). Before planting, the field was fertilized as recommended according soil test. In this study, each sub-plot had 4 rows with in-row spacing of 60 cm, length of 5 m and an area of 12 m². After planting, the field was irrigated once every 4-7 days up to plants establishment and then, it was carried out up to the end of season on the basis of 80 mm evaporation from the class A evaporation pan. In the stress treatment, the irrigation was stopped after 80-120 mm evaporation from the evaporation pan. The sampling was carried out at different stages. At harvest time, the traits measured included head size/plant, grain number/head, plant height, growth period duration, oil content, 1000-grain weight, biological yield, harvest index and grain yield (Nazariyan *et al.*, 2009).

The experiment 3 was conducted by Habibi (2013). In this experiment Seeds of barley (*Hordeum vulgare* L. cv. Rihane-03) were grown in a field trial in sandy loam soil near Malekan, NW Iran. Seeds planted on 5 rows in each plots, the rows distance was 20 cm and the plant distance on each row was 5 cm, beginning and end of each plots closed, with regarding area of each plots. For the basal fertilization, 100 kg ha⁻¹ nitrogen as NH₄NO₃ and 50 kg ha⁻¹ phosphorus and potassium as KH₂PO₄ were applied before sowing. Experiments were performed in complete randomized block design with 4 replications. The replicates were separated at random into two groups; well watered group and water-stressed group. For normal irrigation (well watered group), soil was kept at approximately 70% of field capacity by watering with tap water every 7 days and water holding at the beginning of stem elongation stage in water-stressed group of plants. After 35 days of drought exposure, selenium was sprayed at 30 g ha⁻¹ as sodium selenate. After 10 days of selenium exposure, the plants were harvested and parameters were determined. Thousand seed weight and seed yield were measured at the end of the experiment. *Plant harvest and analysis of water relations*: Leaves were washed with distilled water, blotted dry on filter paper and after determination of fresh weight (FW) were dried for 48 h at 70 °C for determination of dry weight (DW). Before harvest gas exchange parameters were measured. Net CO₂ fixation (A, $\mu\text{mol m}^{-2} \text{s}^{-1}$), transpiration rate (E, $\text{mmol m}^{-2} \text{s}^{-1}$) and stomatal conductance to water vapor (gs, $\text{mol m}^{-2} \text{s}^{-1}$) were measured with a calibrated portable gas exchange system (LCA-4, ADC Bioscientific Ltd., UK) either after 5 h into the light period and sealed in the leaf chamber under a photon flux density of $2000 \pm 100 \mu\text{mol m}^{-2} \text{s}^{-1}$ in field conditions. Chlorophyll fluorescence parameters were recorded using a portable fluorometer (OSF1, ADC Bioscientific Ltd., UK) for both dark adapted and light adapted leaves.

The experiment 4 was conducted by Mobasser and Tavassoli (2013). In this experiment the experimental design was split plot using randomized complete block design with three replications. Treatment was consisted of irrigation in 4 levels, S0: complete irrigation, S1: halted irrigation at squaring, S2: halted irrigation at 50% flowering, S3: halted irrigation at grain filling as main

plot; and cultivars in 3 levels, V1: Zaria, V2: Alstar, V3: Azargol as sub factor. Before planting, 200 kg/ha P (as triple super phosphate) and 150 kg/ha K (as potassium sulfate) was added to the respective treatments, while 150 kg/ha N as urea was applied in two doses; half at planting and the remaining half at 55 days after planting. Sunflower was planted manually in March 2004. Experiment plots were designed with 50 cm row to row distance and 20 cm between plants. Seeds were sown 5 cm deep. Weeds were removed by hand. After planting, irrigation was applied as required during the growing season. Data collected (obtained by combining the four center rows at each experiment unit) included: plant height, seed number of head, 1000 grain weight, yield grain, oil yield and oil percent. Seed oil content was determined according to A.O.A.C. (1990) using soxhlet apparatus and diethyl ether as a solvent.

RESULTS AND DISCUSSION

In the experiment 1 was conducted by forouzandeh et al (2014), the results showed that Drought stress had significant effect on Biological yield. Also the effect of fertilizer treatment and its interaction with irrigation on Biological yield was not significant (Table 1). The lowest Biological yield (309.08 kg/ha) was obtained from I1: two times irrigation (Table 2). The highest Biological yield resulted from 15 t/ha compost application treatment and animal manure (forouzandeh *et al.*, 2014).

The reduction of cumin Biological yield in water stress condition also has reported by Ahmadian *et al.* (2011a). In that experiment, the highest Biological yield resulted from 3 applications of irrigation with an application of 30 t/ha animal manure. Tatari (2004) reported that increasing irrigation times enhanced biological yield significantly. According to results of analysis of variance, total yield was significantly affected by Irrigation times at 1% probability level and Fertilizers at 5% probability level. Also interaction between irrigation times and fertilizers types were not significant differences (Table 1) (forouzandeh *et al.*, 2014). The highest (458.29 kg/ha) and lowest (146.08 kg/ha) total yield was produced under the treatments 4 and 2 irrigations times respectively (forouzandeh *et al.*, 2014). Also the application 30 t/ha animal manure treatment was obtained 316.39 kg /ha total yield (Table 2) (forouzandeh *et al.*, 2014). Manure application improves the soil structure and soil moisture content, provides plant with essential elements, increases growth, number of umbrella per plant and biological yield and finally led to increase seed yield (Ahmadian *et al.*, 2011b). Bilandi (2004) on cumin and Seghatoleslami (2013) on cumin also reported that manure application increases cumin yield. According to results of analysis of variance, essential oil yield was significantly affected by Irrigation times and Fertilizers at 1% probability level According to results of analysis of variance, all traits in different levels of drought stress were significant so the Essential oil yield decreased under drought stress (Table 2). Highest 7.08) and lowest (1.09) levels yield were obtained in 4 irrigations (I3) and 2 irrigations (I1) treatments, respectively (forouzandeh *et al.*, 2014).. In Fertilizers application, higher yield was related to compost treatment (4.84) and vermicompost

(4.77) and Control treatments (2.36) had lower Essential oil yield respectively. In conclusion, results showed that manure could be used effectively to modify the impact of water shortage and to stimulate an increase in cumin seed and essential oil yields probably through improving the water holding capacity of the soil (Seghatoleslami, 2013).

Analysis of variance showed that the Essential oil yield was significantly affected by Irrigation times at 5% probability level and Fertilizers and interaction between irrigation \times fertilizers at 1% probability level (Table 1). The highest (1.61) and lowest (1.41) Essential oil Percentage was produced under the treatments 4 and 2 irrigations times respectively. Also in the application to compost and vermicompost treatments was obtained highest Essential oil Percentage (Table 2) (forouzandeh *et al.*, 2014). These results are in agreement with those obtained by Forouzandeh *et al.* (2012). In the experiment 2 was conducted by Nazariyan *et al.* (2009), the results showed that there were significant differences in grain yield at 1% level among different stress levels, cultivars and their interactions (Table 3), so that no-stress and stress at head formation treatments had the highest and lowest grain yields (5104 and 2026 kg/ha), respectively. Among the cultivars, Azargol had the highest (4063 kg/ha) and Master had the lowest grain yield (3112 kg/ha) (Table 4) (Nazariyan *et al.*, 2009).

The means comparison of the interactions between cultivars and stress levels showed that Azargol under no-stress conditions had the highest grain yield (6220 kg/ha) and Master under stress conditions at head formation had the lowest one (Nazariyan *et al.*, 2009).. The results indicated that Azargol had higher grain yield than the other cultivars under both stress and no-stress conditions and that the stress from head formation until the end of growing season had the highest effect on yield components, especially 1000-grain weight and head diameter because the plants were exposed to drought stress for a longer time. There were significant differences in head diameter among different stress levels at 1% level and among cultivars at 5% level, but their interactions were not significant (Table 3), so that no-stress treatment had the greatest and stress at head formation treatment had the lowest head diameter (Table 4) (Nazariyan *et al.*, 2009).

The results of analysis of variance indicated that biological yield was significant at 1% level among stress levels, cultivars and their interactions (Table 1). Azargol and no-stress treatment had the highest biological yield (Tables 3 and 4). Since these treatments had the highest grain yield too, it could be concluded that grain yield had a direct relation with biological yield and that growth period could be extended under no-stress conditions which could lead to higher biological yield and consequently, higher grain yield. Also, the results showed that harvest index was affected by stress and cultivar (Table 4), so that stress at grain filling stage had the highest and no-stress treatment had the lowest harvest index (Table 4). The grain number/head among different moisture levels and the interactions between stress and cultivars was not significantly different, but the cultivars significantly affected this trait at 1% level (Table 3). Their means comparison showed no considerable difference among different stress levels, too (Table 4) (Nazariyan *et al.*, 2009).

Table 1: Square means of yield components affected by fertilizers organic and drought stress (forouzandeh et al., 2014).

Sources of variation	df	Biological yield	Total yield	Harvest Index	Essential oil percentage	Essential oil yield
Replication	2	106502.63*	29770.34*	382.21ns	0.020ns	0.060ns
Irrigation	2	993524.42**	316927.42**	1885.62**	0.116*	90.40**
Fertilizers	3	87177.91ns	26838.34*	150.24ns	1.52**	12.03**
Interaction F*I	6	39822.63ns	11212.20ns	10.44ns	2.40**	5.40**
Error	22	22504.19	5933.43	103.81	0.022	0.018
CV %	-	27.6	27.8	9.4	9.7	3.4

**, * statistical significant on 0.01 and 0.05 ns: not significant.

Table 2: Mean of yield components affected by fertilizers organic and drought stress (forouzandeh et al., 2014).

Sources of variation	Biological yield	Total yield	Harvest Index	Essential oil percentage	Essential oil yield
I1	309.08C	146.08 C	96.55B	1.90C	1.41B
I2	454.21B	223.92B	104.78B	2.91B	1.54 AB
I3	863.92A	458.29A	121.17A	7.08A	1.61 A
Control	407.67B	200.33B	102.06A	2.36C	1.08 C
vermicompost	530.44AB	272.00AB	109.89 A	4.77A	1.82A
compost	614.56A	315.67A	111.27A	4.84A	1.92 A
animal manure	616.94A	316.39A	106.78A	3.88B	1.26 B

There were no statistical differences among the means shown by the same letters at 5 % probability level.

Table 3: Analysis of variance for some traits affected by drought stress in different sunflower cultivars (Nazariyan et al., 2009)

Source of variation	df	Mean square					
		Economical yield	Biological yield	Harvest index	1000-grain weight	Capitule weight	Grain no./capitule
Replication	2	0.6902ns	67.56ns	62.59*	1.53ns	0.813ns	9162.25ns
Stress level	3	19.231**	463.32**	44.28*	453.1**	156.08**	122592.7ns
Error A	6	1.923	40.105	10.122	4.597	11.951	112602.9
Cultivar	3	1.906**	74.607**	42.71**	597.95**	4.46*	76399.2**
Stress × cultivar	9	0.924**	6.676**	18.29*	8.86ns	2.65ns	10648.2ns
Error B	24	0.295	1.987	6.322	3.922	1.167	4791.93
CV (%)		14.97	9.22	10.28	2.79	5.98	9.72

* and ** show significance at 5 and 1%, respectively, and ns shows non-significance.

Table 4: Means comparison of some traits as affected by drought stress (main plot) and different cultivars (sub-plots) (Nazariyan et al., 2009).

Factor	Economical yield	Biological yield	Harvest index	1000-grain weight	Head diameter	Grain no./head
Stress levels	LSD = 1200	LSD = 5480	LSD = 2.72	LSD = 1.85	LSD = 3	LSD = 290
No-stress	5104 a	23466 a	22.0 b	78.85 a	23.0 a	814.5 a
Stress at head formation	2026 c	8360 c	24.3 b	64.03 d	14.3 c	586.7 a
Stress at flowering	3533 b	14379 b	25.0 a	69.25 c	17.5 b	769.0 a
Stress at grain filling period	3858 b	14927 b	26.5 a	71.35 b	17.4 b	677.6 a
Cultivars	LSD = 905	LSD = 5478	LSD = 2.75	LSD = 1.9	LSD = 2.94	LSD = 83.9
Master	3112 b	12396 b	25.5 ab	66.7 c	17.7 a	671.9 b
Lakomka	3763 a	14723 b	26.5 a	80.24 a	17.6 a	620.7 b
Azargol	4063 a	18432 a	22.4 c	72.20 b	18.9 a	760.6 a
Euroflor	3582 a	15581 b	23.4 bc	64.4 d	18.1 a	794.5 a

Means with the same letter(s) in each column are not significantly different.

Table 5: Shoot dry weight (mg plant⁻¹), thousand seed weight (g), seed yield (kg ha⁻¹) and leaf relative water content (RWC, %) under different treatments. Each value is the mean ± SD of 20 replicates. Data of each column indicated by the same letters are not significantly different (P < 0.05) (Habibi, 2013).

Treatments	Shoot dry weight	Thousand seed weight	Seed yield	Relative water content
control	1845 ± 131 b	47.7 ± 4.19 a	2887 ± 141 a	72.5 ± 2.38 b
Drought	1120 ± 120 c	36.5 ± 4.43 b	1455 ± 161 b	55.2 ± 3.11 c
Selenium	2100 ± 212 a	51.2 ± 2.38 a	2995 ± 174 a	83.5 ± 3.69 a
Drought+Selenium	1210 ± 143 c	2 ± 2.50 b	37. 1565 ± 83 b	57.6 ± 2.64 c

In the experiment 3 was conducted by Habibi (2013), the results showed that both relative water content (RWC) and dry weight decreased dramatically in water-stressed plants. In contrast to drought, selenium spraying treatment increased relative water content and dry matter accumulation in well-watered plants, as compared with control plants (Table 5) (Habibi, 2013). Thus, the treatment with the highest dry matter accumulation (supplemented well-watered treatment) showed the highest relative water content (83.5%). At the end of the

experiment, Thousand seed weight and seed yield decreased by 23.4 and 49.6% under water stress, respectively, in comparison to their respective plants under well-watered conditions. Seed yield was not affected by selenium spraying treatment. The study of PSII photochemistry in the dark adapted leaves showed that there was no significant difference in the maximal quantum yield of PSII (F_v/F_m) between control and Supplemented plants under well-watered conditions (Table 6) (Habibi, 2013).

Table 6: Leaf physiological traits of barley plants under different treatments. A net photosynthetic rate, Etranspiration rate, gs stomatal conductance, WUE (A/E) water use efficiency, Fv/Fm maximum quantum yield of PSII, qP photochemical quenching, qN non-photochemical quenching, Φ PSII effective quantum yield of PSII. Each value is the mean \pm SD of 4 replicates. Data of each row indicated by the same letters are not significantly different ($P < 0.05$) (Habibi, 2013).

Photochemistry	Control	Drought	Selenium	Drought Selenium
Fv/Fm	0.84 \pm 0.01a	0.81 \pm 0.01 b	0.84 \pm 0.02 a	0.82 \pm 0.01ab
qP	0.96 \pm 0.02 a	0.96 \pm 0.02 a	0.95 \pm 0.02 a	0.95 \pm 0.01 a
qN	0.17 \pm 0.05 a	0.15 \pm 0.02 a	0.14 \pm 0.09 a	0.16 \pm 0.08 a
Φ PSII	0.79 \pm 0.01a	0.76 \pm 0.01 b	0.79 \pm 0.01a	0.76 \pm 0.01b
Gas exchange				
A (μ mol m ⁻² s ⁻¹)	14.2 \pm 3.52 a	5.17 \pm 1.63 b	16.3 \pm 1.46 a	1 6.93 \pm 2.40 b
E (mmol m ⁻² s ⁻¹)	5.95 \pm 0.67 a	3.61 \pm 1.34 a	5.54 \pm 0.14 a	4.65 \pm 1.40 a
gs (mol m ⁻² s ⁻¹)	0.41 \pm 0.05ab	0.27 \pm 0.13 b	0.52 \pm 0.13 a	0.36 \pm 0.10 ab
WUE (μ mol mmol ⁻¹)	2.39 \pm 0.59 a	1.44 \pm 0.07 b	2.94 \pm 0.16 a	1.49 \pm 0.12 b

Table 7: Effect of irrigation and sulphur levels on factor measured (Mobasser and Tavassoli, 2013).

Treatments	Plant height (cm)	Seed number of head	1000 grain weight (g)	Yield grain (ton/ha)	Oil yield (kg/ha)	Oil percent (%)
Irrigation						
S0	125.083 a	847.500 a	56.853 a	4.700 a	1.980 a	41.980 a
S1	104.750 c	708.833 c	45.633 bc	3.245 c	1.256 c	38.650 b
S2	116.750 b	796.917 b	41.242 c	3.267 c	1.207 c	36.970 b
S3	117.833 b	856.833 a	48.567 b	4.127 b	1.547 b	38.020 b
Cultivar						
V1	124.813 a	856.688 a	40.525 c	3.456 b	1.350 b	38.950 a
V2	100.938 a	750.625 b	49.700 b	3.754 b	1.450 b	38.440 a
V3	122.563 a	800.250 ab	53.794 a	4.293 a	1.712 a	39.310 a

Means followed by similar letters in each column are not significantly different at the 5% level of probability

Table 8: Mean comparison of interaction effects of factors measured (Mobasser and Tavassoli, 2013).

Treatments	Plant height (cm)	1000 grain weight (g)	Yield grain (ton/ha)	Oil yield (kg/ha)
S0V1	136.00 abc	42.00 d	3.87 bcd	6.60 bcd
S0V2	100.75 de	61.57 a	4.47 b	1.84 b
S0V3	126.75 abc	66.17 a	5.57 a	2.49 a
S1V1	114.75 abcde	42.27 d	2.84 f	1.10 d
S1V2	95.50 e	45.47 cd	3.46 cdef	1.33 cd
S1V3	104.00 cde	49.15 bc	3.42 cdef	1.33 cd
S2V1	126.50 a	33.57 e	3.04 ef	1.09 d
S2V2	96.25 bcde	43.92 cd	3.08 def	1.51 cd
S2V3	127.50 ab	46.22 cd	3.67 bcdef	1.37 bcd
S3V1	132.00 abcd	44.25 cd	4.05 bc	1.59 bcd
S3V2	100.25 e	47.82 cd	3.99 bc	1.47 bcd
S3V3	126.00 ab	53.62 b	4.32 b	1.65 bc

Means followed by similar letters in each column are not significantly different at the 5% level of probability

However, reduction of maximal efficiency of PSII in dark-adapted leaves (F_v/F_m) and effective quantum yield of PSII (Φ PSII) were detectable in leaves of water-stressed plants. In addition, stomatal conductance to water vapor (g_s) was positively correlated with F_v/F_m ($r = 0.70$, $P < 0.05$) in water-stressed plants. Photochemical quenching (qP) and non-photochemical quenching (qN) were not influenced under selenium spraying and drought conditions (Habibi, 2013). Net assimilation rate (A) was not influenced by selenium spraying, but was reduced by drought (Table 2). Transpiration rate (E) was not affected significantly by water stress, while g_s was reduced strongly under drought conditions but increased by selenium. In this study, a remarkable reduction in shoot dry weight in drought stressed plants was associated with a significant reduction of net CO_2 assimilation rate. Water use efficiency was significantly lower in drought-stressed plants. Thus, compared with the transpiration rate, the water use efficiency showed a greater decrease during the water deficit (Habibi, 2013).

In the experiment 4 was conducted by Mobasser and Tavassoli (2013). The results showed that plant height

affected by irrigation treatments ($P < 1\%$), so that the maximum and minimum plant height respectively was achieved from complete irrigation treatment with mean 117.833 cm and irrigation treatments in stage of halted irrigation at squaring with 104.750 cm (Table 7). Water stress causes deceleration of cell enlargement and thus reduces stem length by inhibiting inter nodal elongation and also checks the tillering capacity of plants. Interaction effect of irrigation treatments and cultivars in plant height was significant ($P < 5\%$). Maximum and minimum plant height was achieved from treatments of S0V1 and S1V2 with mean 136 cm and 95 cm respectively (Table 8).

According to results of variance analysis effect of treatments of irrigation and cultivars was significant on 1000 grain weight ($P < 1\%$). The highest 1000 grain weight (56.583 g) obtained from treatment of complete irrigation and the lowest amount of oil yield (41.242 g) was seen from treatment of stress in flowering stage (Table 3). The decrease of vegetative growth in condition of water shortage leads to decrease of photosynthesis materials production in plant and finally decrease of 1000 grain weight. Among cultivars treatments the most 1000

grain weight obtained from Azargol cultivar (53.794 g) and the lowest amount of it was achieved from Zarya cultivar (40.525 g) (Table 7). The interaction effect between irrigation and cultivar treatments was significant ($P < 5\%$). Maximum and minimum 1000 grain weight was achieved from treatments of S0V3 and S2V1 with average 66.175 g and 42.275 g respectively (Table 8). The results of variance analysis showed that effect of treatments of irrigation and cultivars was significant on grain yield ($P < 1\%$), so that the highest grain yield from treatment of complete irrigation with an average 4700 kg and the lowest grain yield was seen in treatment of halted irrigation at squaring with yield average 3245 kg. Among cultivars treatments the highest grain yield obtained from cultivar of Azargol with average 4300 kg and the lowest yield about 3450 kg was achieved from cultivar of Zarya (Table 7).

This matter can be by reason of higher resistance of Azargol cultivar to water deficit and having a high leaf surface duration reproductive stage. The interaction of irrigation and cultivars on grain yield was significant ($P < 5\%$), so that the highest and lowest grain yield was achieved from treatments of S0V3 (5750 kg) and S1V1 (2846 kg) respectively (Table 8). The results of variance analysis showed that effect of treatments of irrigation and cultivars was significant on grain yield ($P < 1\%$). The highest oil yield (1980 kg/ha) obtained from treatment of complete irrigation and the lowest amount of oil yield (1207 kg/ha) was seen from treatment of stress in flowering stage (Table 7). Probably adequate irrigation during the vegetative stage, leaf development and grain filling stage can increase grain weight and oil storage. Among cultivars treatments the most oil yield obtained from Azargol cultivar (1712 kg/ha) which can be due to the high yield of grain this cultivar compared to other cultivars and the lowest amount of it was achieved from Zarya cultivar (1350 kg/ha) (Table 7) (Mobasser and Tavassoli, 2013).

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