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ORIGINAL ARTICLE

The effect of Water limitation on Water Relations, growth and seed yield of four soybean (*Glycine max* merri.) genotypes

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ABSTRACT

Drought is one of the major constraints for soybean in Iraq. In order to investigate the effects of different soil moisture levels on the water relations, mineral nutrients (K, Ca) content, growth and seed yield of four Glycine max genotypes, a split-plot experiment (using CRB design) with four replications was conducted .Irrigation regimes :50, 60, 70, 80, and 100% of field capacity , respectively and soybean genotypes : S, Pi, G and Lee were allocated to main and sub plots, respectively. The parameters that were measured are the leaf relative water content, K and Ca content , dry weight of shoot and root, root length, Yield component and plant seed yield. There was decrease in the leaf relative water content in plants under water deficit, treatment under water deficiency 60 and 50% of F.c. had fall to 74.6 and 60.6%, respectively. when compared with the control plants (100% field capacity) 85.33 %. While osmotic potential increase to 0.716 and 0.647. Genotype interacted significantly (P<0.05) with water level when studying osmotic potentials but not significantly with relative water content. Drought stress not affect significantly Ca and K uptake in plants. The genotypes considerably differed with respect to K uptake ,genotype Pi uptake more K 466.2 m mole Kq-1 than other genotypes. The results showed that dry matter accumulation were reduced due to water deficit. Water limitation also reduced pods per plant and 1000 seeds weight. Consequently, seed yield per plant under limited irrigation was considerably lower than that under well-watering. The numbers of pods per plant were the most affected yield components under drought stress. Lee genotype had the highest dry matter 8.9 g, pods per plant 81.84, 1000 seeds weight 15.2 g. The superiority of Lee genotype in plant seed yield 2.613 g resulted from higher pods per plant and 1000 seeds weight. S and Lee genotypes consumed large amount of water 1199.9 and 1158.5 mm, while Pi and G genotypes consumed low amount of water 876.5 and 892.7 mm therefore the results show the possibility of the use of Pi and G genotypes in the less moisture. May be due to the osmotic adjustment which is one of the major adaptive mechanisms to survive drought.

Key words: Glycine max, water relations, mineral nutrients content, growth, seeds yield ,water stress.

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INTRODUCTION

Soybean [*Glycine max* (L) Merrill] is one of the most important oil seed crops in the world. The crop has gained popularity because of its potential to supply high quality protein [1]. In spite of its great potential, soybean production is still inadequate in Iraq owing to various limitations which result in low yield per unit area. Seed yields of soybean cultivars are generally low in Iraq compared to other places in the world. Yield on growers' farms is often lower than 1500kg ha-1, compared to yields > 2500 kg ha-1 in the USA, 3000 kg ha-1 in Brazil and >3500 kg ha-1in Turkey [2]. There is therefore a wide gap between what is currently being produced and what is needed. Increasing soybean production to meet the required quantities can best be achieved through an increase in yield per unit area, which can partly be achieved by the cultivation of high-yielding improved varieties.

Environment inadequate conditions due to water [3], heat [4], salt [5] and mineral stress [6] can cause reductions in morphological and agronomical parameters, as well as disorders at physiological, biochemical and molecular levels [7].Drought is a common component in agricultural regions due the

irregular rainfalls and/or inadequate irrigation supply to the crop [8]. In addition, water deficit during vegetative, reproductive and maturation periods result in lower growth and development rates [9], moreover it promotes flower abortion during the reproductive period [10]. The water deficit is characterized by water losses that exceed the absorption rate and of this way it acts directly in the plant water relations [11], in which the plant damages depend on the intensity and the exposure period, besides promoting changes in the cell and the molecular pathways [12], and decreased grain yield [13].

The root system is considered to be a primary sensor of drought stress and may play an important role in drought avoidance [14] by making deep penetration into soil in search of water. However drought is always unpredictable, regular selection of cultivars at a particular site under natural conditions is extremely difficult [15]. Drought stress may involve the uptake of mineral elements in plant tissues by affecting root growth and nutrient mobility in soil and nutrient uptake [16]. Decreasing water availability under drought generally results in reduced total nutrient uptake and frequently causes reduced concentrations of mineral nutrients in crop plants. The most important effect of water deficits is observed on the transport of nutrients to the root and on the root growth and extension. Reduced absorption of the nutrient elements results from an interference of nutrient uptake and unloading mechanisms and reduced transpiration flow [17]. However, plant species and genotypes within species differ in their response to nutrient element uptake under water stress [18]. Drought significantly reduced nutrient use efficiency in plants and selection of improved genotypes adaptable to drought conditions has been a major contribution to the overall gain in crop productivity [19]. Drought is undoubtedly deleterious for plant growth [16, 18]. The drastic effect of drought stress observed is on the transport of nutrients to the root and on the root growth and extension [20]. Inorganic nutrients such as N, P, K+, Ca2+ and Mg2+ ion play multiple essential roles in plant metabolism. Potassium plays an important role in osmoregulation, activates enzymes of respiration and photosynthesis and has a role in stomatal regulation. Calcium is the component of cell membrane and thus plays a significant role in plant growth [21]. Nonetheless, each of these nutrients must be maintained at an optimum concentration range for proper growth.

It is well known that water deficit in plants can induce a lowering of osmotic potential in some species and cultivars which contributes to cell turgor maintenance at low leaf water potentials [22]. Maintenance of turgor pressure helps the plant in maintaining stomatal opening, photosynthesis, and more water uptake from the soil [22]. Genotypic differences in turgor maintenance have been reported on several grasses such as wheat [23] and sorghum [24]. Water potential and its components are generally considered a reliable measurement of the water status of plant tissue [22]. By the mid eighty's, relative water content was proposed as a better indicator of water status than water potential [25]. This is because relative water content, through its relation to cell volume, may more closely reflect the balance between water supply to the leaf and transpiration rate. Measurements of total leaf water potential, leaf osmotic potential and relative water content relate directly or indirectly to plant response to water stress [22]. All of these are, in theory, potential water stress resistance screening criteria [26]. The importance of plant water for the maintenance of turgidity required for plant growth is widely recognized. Many workers [27, 22] have shown that water stress-resistant plants have smaller water deficit per unit decrease in leaf water potential than more water-stress susceptible plants. Several reports suggest that plant metabolic processes are in fact more sensitive to turgor and cell volume than to absolute water potential [28]. Among the physiological mechanisms that act to maintain leaf turgor pressure, decreased osmotic potential resulting either from a decrease in osmotic water fraction or from an osmotic adjustment (net accumulation of solutes in the symplast) was pointed out [29]). Changes in tissue elasticity in response to drought, which modify the relationship between turgor pressure and cell volume. might contribute to drought tolerance, as observed in sunflower [30]. Leaf water relations data may provide a useful indication of the capacity of species to maintain functional activity under drought [31] The aim of this study was to evaluate the impact of water stress on relative water content, osmotic potential, potassium and calcium content, root growth and yield of four soybean genotypes.

MATERIAL AND METHODS

The experiment was designed to evaluate the response of four soybean genotypes to soil moisture levels . The experiment was designed as a 4 x 5 factorial in RCBD, laid out in split-plots arrangement, with four soybean genotypes (S, Pi ,G and Lee) as the main plots, and five soil moisture levels (50 , 60 ,70 , 80 and 100 % of F.C.) as the sub-plots, with four replications.

Growth conditions

The experiment was carried out in pots. Equal weight plastic pots of same size (45cm diameter) were filled with equal weights (10kg) of clay loam soil. Analysis of the soil used in the experiment was carried out in Soil department, college of Agriculture, Baghdad university. The soil used was clay loam. The other contents of the soil were as follows: Field

capacity 34%, texture clay loam, CaCO3 15.2 mg.kg-1, pH 7.55, EC 4.3 dS/m, organic matter (OM) 0.56%, NO3 1.6 mg.kg-1. exchange K 0.6 m mole , Ca 1.22 m mole and available P was 15.20 mg/kg; . Then the soil in each pot was completely saturated with normal irrigation water. When the moisture contents were at field capacity, three seeds were hand sown At 15 /5 . Thinning of plants was done 15 days after germination to maintain one plant per pot. The genotypes were grown in a glasshouse under natural light conditions. A basal dose of N (Urea) and P (Calcium super phosphate) were applied to all the pots at 1.35 and 5.4 P2O5 g , respectively, and mixed thoroughly with soil. The pots were moved and rearranged daily to give a random distribution of growth conditions in the glasshouse during the experimental period.

Soil moisture levels and water consumption

Five water stress treatments [field capacity, (control) and 50,60,70 and 80% field capacity] were started after seed germination. The moisture contents of droughted pots were

maintained and regularly monitored by keeping the weight of each pot equal to that

calculated for 50,60.70 and 80% field capacity through addition of normal irrigation water if required on daily basis till the maturation of the crop. and water consumption was calculated.

Harvest

Plants were harvested at the end of the experiment 15/10, washed and oven dried at 70°C for 96 h until reaching a constant mass after which time dry weight of shoot and root were recorded. Root length , weight of shoot and root ,weight of seeds per plant , number of pods per plant and seed yield per plant were recorded at harvest.

Plant water relations

leaf relative water content (LRWC) was carried out with 10 mm disks of diameter, it was calculated as: LRWC = $[(FW-DW)/(TW-DW)] \times 100$, in which FW is fresh weight, TW is the turgid weight measured after 24h of saturation on demonized water at 4°C in the dark, and DW is the dry weight determined after 48 h in oven at 80°C [32]. The plants were placed in dried oven under 65 °C for 72 h and after of the dehydration, it was measured the plant dry matter. Leaf osmotic potentials were measured after leaves freezing in liquid nitrogen and measuring osmotic potential in the expressed sap using a vapor pressure osmometer.

Determination of inorganic elements in plant

The dried ground material (0.1 g) of leaf and root was digested with 2 mL Sulphuric acid-hydrogen peroxide mixture according to the method of Wolf [33]. Potassium and calcium in the digests were determined using a flame photometer.

Statistical analysis

The data for all variables were subjected to analysis of variance following the split- plots model using Genstat 5.32 statistical package and significant means were separated by the Least Significant Difference (LSD0.05) at 5% probability level.

RESULTS AND DISCUSSION

Plant water relations and plant analysis

Water deficiency promoted significant decrease in leaf relative water content in stress treatment (Table 1), treatment under water deficiency 60 and 50% of F.c had fall to 74.6 and 60.6 %, respectively. when compared with the control plants (100% field capacity) 85.33 %.

The reduction in leaf relative water content was provoked by the water deficiency in soil, because during the photosynthesis occur water loss through of the stomatal mechanism and the water assimilation rate is negatively affecting during water stress [34]. Lobato *et al.* [35] reported gas exchanges during photosynthesis and transpiration processes, those it promotes progressive water losses coming from leaf tissue to air/environment. Lobato et al. [8] studying *Glycine max* plants induced to water deficit described that this species less tolerant than other legumes. Water deficiency promoted increase in osmotic potential to 0.647 and 0.716 in treatments 60 and 50 % of field capacity, respectively . Drought induced osmotic stress triggers a wide range of adverse effects in plants ranging from poor growth, imbalanced ion transport [36], considerable decrease in transpiration rate and impaired membrane permeability [36] resulting in a reduced absorbing capacity of crop plants [20]. Genotype interacted significantly (P<0.05) with soil water level when studying osmotic potentials but not significantly with relative water content (Table 1). Values of relative water content of the four genotypes S 85.3 %, Pi 83.3 %, G 84.8 % and Lee 78.6 %. Schonfeld *et al.* [37], believed that cultivars to be more water stress resistant usually maintained higher leaf relative water contents under stress.

The data in Table 1 show that the leaf osmotic potentials of Pi and G genotypes were significantly higher than S by 42.91,40.08 % and Lee by 42.91, 58.00 %, respectively.

Plants accumulate different types of organic and inorganic solutes in the cytosol to lower osmotic potential thereby maintaining cell turgor [38].

Iannucci *et al.* [39] who reported that proline levels were more closely related to the decrease in RWC. Cuin and Shabalah [40] reported that solutes like glycinebetaine, proline, mannitol, trehalose or inositol significantly reduced K+ efflux from the cell and maintains cytosolic K+ homeostasis possibly through the enhanced activity of H+- ATPase. This in turn controls voltage-dependent outward-rectifying K+ channels and created the electrochemical gradient necessary for secondary ion transport processes [41].

Drought stress not affect significantly Ca and K uptake in plants (Table 1).. The genotypes considerably differed with respect to K uptake "genotype Pi uptake more K 466.2" m mole Kg-1than other genotypes , and genotype S uptake less K 228.5 m mole Kg-1.Ca uptake not differ significantly in all genotypes. K uptake greatly differed among the genotypes (Table 1). Drought is deleterious for plant growth, yield and mineral nutrition [19, 16] .Cultivars differ in their response to environmental stress. Better understanding of genotypic control of nutrient uptake mechanisms is required for soybean to maintain the production under rain fed or drought conditions. Bruckner and Frohberg [42] suggest that cultivars with low growth reduction values are presumed to be drought resistant because they exhibit a smaller reduction in dry weight than the average dry weight under stress compared with favorable conditions. Similarly, cultivars exhibiting smaller reduction in nutrient uptake and efficiency should be presumed to be drought resistant, and nutrient uptake efficiency involves drought tolerance mechanisms [43]. Water and different nutrients exist together in plant tissues in close association, because nutrient ions are dissolved in the soil solution and nutrient uptake by plants depends on water flow through the soil-rootshoot continuum). Therefore leaf transpiration creates the tension necessary for the roots to absorb the soil solution containing essential nutrients [44]. Decreasing water availability under drought generally results in reduced total nutrient uptake and frequently reduces the concentrations of mineral nutrients in crop plants [17]. Plant species and genotypes within species vary in their response to mineral uptake under water stress [18]. In the present study 4 soybean genotypes greatly varied with respect to K nutrient uptake. Great differences were reported previously in N, P and K uptake among 20 genotypes of chickpea [45]. According to Rengel [46], on soils under micronutrient deficiency, micronutrient- efficient genotypes have a greater yield in comparison to inefficient ones. In a study carried out with faba bean under water deficit, N, P and K uptake significantly decreased [47]. As suggested by Samarah et al. [6] this also indicates that nutrient efficiency may involve drought tolerance mechanisms and selection of nutrient-efficient cultivars contributes to environmentally- benign agriculture by lowering the input of chemicals and energy. However, there are some studies explaining the relationship between nutrient uptake and drought tolerance in sovbean [16] in faba bean [47]. As suggested by Garg [18] the interaction between soil moisture deficits and nutrient uptake is of principal importance, . Samarah et al. [16] recently suggested that the uptake of minerals in soybean seed grown under drought stress may have an important role in drought tolerance.

Plant dry matter

Significant difference was showed in the shoot and root dry matter, in which the treatment kept under 100% of field capacity (control) had increase to 10.25 and 13.08 g ,respectively (Table 2). The smaller dry matter 3.1 and 5.5 g were shows in plants submitted to water restriction 50% of F.c , water stress had no effect on root length ,and genotypes roots not differ in their length .The root growth is a strategy used by the plants to water capture in substrate under water deficit conditions [9], in which the growth and development plant is dependents of the cell turgor, as well as the water fills the cell space and practice a positive pressure that promotes through this mechanism the tissue extension [48].Moisture levels x genotype interaction effects were significant for shoot weight. Lee genotype irrigated with 100% of F.c gave highest weight reached to 13.1g, while G genotype irrigated with 50% Of F.c gave the lowest shoot weight 1.80 g (Table 4).

Recent studies evaluating *Glycine max* under water deficiency describes an increase in root dry matter with objective of re-establish plant water necessity and consequently increase the absorption range into substrate [9].

Yield component and plant seed yield

Number of seeds per pod was not significantly affected by irrigation regimes (Table 2)and genotypes (P>0.05) (Table 3). The lowest pods per plant 12.53, weight of 1000 seeds 8.10 g and plant yield 0.381 g were obtained under severe water deficit 50% of F.c as compared with 100% of F.c which gave the highest pod per plant 83.85, weight of 1000 seeds per plant 15.5 g and plant yield 2.78 g (Table 2). The number of pods per plant, followed by weight of 1000 seeds was the most affected plat yield of seeds under drought stress. Legume plants with relatively high water requirements are very sensitive to periodical soil drought Since, there is a certain correlation between dry matter production and grain yield in faba bean crop [49]. In our research, water stress exerted a large adverse influence on number of pods

per plant , 1000 seeds weight and plant yield of seeds of soybean genotypes. These results are in accordance with the data obtained by Abayomi [50]. Water deficit through the reduction in the leaf area expansion and photosynthetic capacity reduces total dry matter and eventually CGR [51] because the earliest response to the leaf water deficit is stomata closure, which limits CO2 diffusion to chloroplasts and limits photosynthesis [52]. Water stress generally accelerates leaf senescence and shortens grain filling duration soybean [53].

Table 1. Effect of soil water levels and genotypes on relative water content (RWC), osmotic potential (0.P), potassium (K) and calcium (Ca) content of soybean.

Treatments	RWC %	OP	K m mole Kg-1	Ca m mole Kg-1
Irrigation (F.C)*				
!00	85.33	0.411	280.83	360.33
80	85.15	0.455	378.05	298.75
70	84.35	0.522	328.53	561.85
60	74.60	0.647	374.73	5 61.85
50	60.60	0.716	357.05	447.38
L.S.D.0.05	8.14	0.120	N.S	N. S
Genotypes				
S	85.30	0.529	228.50	519.00
Pi	83.30	0.756	466.20	401.70
G	84.80	0.741	386.20	322.70
Lee	78.60	0.469	294.50	369.00
L.S.D.0.05	N.S	0.161	50.136	N.S
L.S.D.0.05	N.S	0.161	50.136	N.S

* F.C = Field capacity

Table 2. Effect of soil water levels on seed yield and yield components of soybean.

Treatment (F.C)*	Shoot dry weight (g)	Root dry weight (g)	Root length) (Cm)	Number of seeds /pod	Number of pods /plant	1000seed weight (g	Plant) yield (g)
100	10.25	13.08	51.75	2.2	83.85	15.5	2.783
80	6.13	9.45	49.00	2.2	55.67	12.4	1.815
70	7.20	8.70	49.03	2.1	38.53	11.0	1.203
60	4.38	7.68	48.73	2.1	22.67	9.00	0.696
50	3.10	5.50	48.83	2.0	12.53	8.10	0.381
L.S.D.0.05	2.848	2.12	N.S	N.S	6.14	1.44	0.11

* F.C = Field capacity

Table 3. Comparison of yield and yield components of four soybean genotypes.

Genotypes	Shoot dry weight (g)	Root dry weight (g)	Root length (Cm)	Number of seeds /pod	Number of pods /plant	1000seed weight (g)	Plant yield (g)
S	7.52	8.10	41.54	2.2	77.21	14.88	2.470
Pi	4.40	6.60	40.70	2.1	60.01	14.1	1.834
G	4.00	9.60	56.26	2.1	57.11	13.91	1.699
Lee	8.90	11.20	59.37	2.2	81.84	15.20	2.613
L.S.D.0.05	2.353	N.S	N.S	N.S	4.11	1.02	0.17

Table 4. Soil water levels x genotype interaction effects on shoot dry weight of soybean.

Genotypes			Soil water levels of F.C*			
	100	80	70	60	50	
S	7.60	8.60	10.00	7.20	4.20	
Pi	8.80	3.20	4.40	3.00	2.50	
G	11.50	1.50	3.40	2.60	1.20	
Lee	13.10	11.20	11.00	4.70	4.50	

L.S.D.o.o5 5.70

* F.c =Field capacity



Figure 1. Effect of soil water levels on water consumption of soybean



L.S.D.0.05=14.861

Figure 2. Comparison of water consumption of four soybean genotypes.

The soybean genotypes varied significantly in number of pods per plant, weight of 1000 seeds and Plant yield, while showed no significant differences in their number of seeds per pod (Table 3) However,. Number of pods was lowest with G and PI and the value was significantly lower than in Lee genotype by 41.21 and 36.38 %, respectively may be due to the genetic variations between genotypes in number of flowers per plant and abortion of flowers. Lee genotype had significantly higher weight of 1000 seeds than G genotype by 9.27%, while other genotypes showed no significant differences. Plant yields of seeds among the evaluated genotypes varied significantly, seed yield was significantly higher for Lee and S genotypes 2.613 an 2.470 g /plant, respectively. G genotype had the lowest seeds yield 1.699 g / plant which was significantly lower than that of other genotypes. The superiority of Lee and S genotypes in plant seed yield resulted from higher pods per plant and 1000 seeds weight (Table 3), Other workers have reported significant yield differences among soybean genotypes [54]. Soybean yields have been related to moisture availability in many earlier reports [55]. Oya et al. [56] reported that drought stress is one of the main constraints for soybean production. Aduloju et al.[57] reported that results of the simple linear regression of grain yield on growth and yield components showed that plant height and number of pods per plant were the most important factors affecting grain yield. Regression of yield on individual vield components recently has been used to show that the most important vield components in vield formation were number of pods and number of seeds per unit area [58]. Other workers have also shown the importance of number of pods/seeds per area in grain yield improvement in soybean [59]. Modali [58] also showed by correlation analysis that number of pods per area was more important in determining number seeds per unit area than was seeds per pod. Lee genotype produced the highest number of pods per plant, and hence the highest seed yield. Seed yield was lowest in G genotype due to

lowest number of pods per plant. The reduced number of pods per area has been shown to play a prominent role in decreasing yield [60]. The reduction in the number of seeds per plant was due mainly to less number of productive pods retained per plant. This is consistent with reports on other legumes including common bean [61]. Drought-induced abortion of pods in line with the suggestions of Daie [62], the higher rate of pod abortion may be due to limited assimilate supply under drought conditions.

Plant water consumption

That to maintain soil moisture close to field capacity, increase the amount of evapotranspiration as given moisture level 100% of F.c higher evapotranspiration was 1718.7 mm, while decrease with the decrease in soil moisture levels to 680.7 mm at the level of moisture 50% of F.c. Bidinger [66] reported the radiation incident on soil surface increase the evapotranspiration components because elevate the soil surface temperature, and more than 70% of this energy used to change the status water from liquid to vapor. Lee and S genotypes used high quantities of water in evapotranspiration 1158.5 and 1199.9 mm compared with Pi and G genotypes which consumed 876.5 and 892.7 mm, respectively. The results show the possibility of the use of Pi and G genotypes in the less moisture. May be due to the osmotic adjustment which is one of the major adaptive mechanisms to survive drought.

CONCLUSION

The results highlight the fact that water stress influenced leaf water relations in soy bean plants. Relative water content at 50% of F.c was reduced in greater extent, but osmotic potential was highest. Drought stress significantly but differentially affected the growth and yield of the four genotypes. The numbers of pods per plant were the most affected yield components under drought stress. Lee genotype had larger number of reproductive sinks (pods and seeds) under drought stress. Water limitation considerably reduced plant yield of soybean genotypes , due to large reductions in growth, number of pods per plant and weight of 1000 seeds Superiority of well-watered plants in growth ,pods per plant and 1000 seeds weight resulted in production of comparatively more and larger seed and consequently higher seed yield per plant. Therefore, sufficient water supply during plant growth and development is necessary to ensure a satisfactory supply of assimilate to the seeds via an extensive and long-lived foliage. Lee showed better performance than other genotypes . These results show the possibility of the use of Pi and G genotypes in the less moisture.

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