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ADJUSTMENT OF INTERNAL WATER BALANCE IN TWO SOYBEAN CULTIVARS TO REDUCED SOIL WATER POTENTIAL

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ABSTRACT:

One challenge of the future will be to maintain or even increase crop production with less water that often may be of poor quality. Selection of plant species/crop cultivars with considerable resistance to drought stress has been considered an economic and efficient means of utilizing drought-prone areas. The primary objective of this study was to assess the response of two cultivars (Giza 111 and 21) of soybean and make a comparison between the two varieties of soybean mainly concerned with the means of adjustments under major ecological stresses (namely: drought, salinity at equipotential levels) and one of such means is water relations of plant which includes transpiration, stomatal resistance, transpiration efficiency, relative water content, dry matter content, osmotic pressure and changes in chlorophyll a and b content, ratio and their stability to heat (mainly as the simplest one of osmo-metabolic adjustments means). Soybean Giza 111 was the most tolerant variety exhibited the highest content of photosynthetic pigment at the highest level of both types of water stress. Thus, there may be a possibility of using this trait as a selection criterion for breeding for improved yield when subjected to equipotential levels of two different water stresses (matric and osmotic water potential stress).

INTRODUCTION:

Crop plants are usually exposed to different environmental stresses which limit their growth and productivity. Among these, soil salinity and drought are the most severe ones. It has been estimated that more than

20% of all cultivated lands around the world contain salt levels that are high enough to cause salt stress to crop plants (Moud & Maghsoudi, 2008). Reduced water potential is a common consequence of both salinity and drought (Legocka & Kluk, 2005). Abiotic stress leads to a series of morphological, physiological, biochemical and molecular changes that adversely affect plant growth and productivity (Wang et al., 2004). These stresses not only have an impact on current crop species, but they are also significant barriers to the introduction of crop plants into new areas that are not currently being used for agriculture.

Studies of crop response to water and salt stress vary either salinity with a high leaching fraction or irrigation in the absence of salinity isolate and quantify the effects of the two types of stress. Under deficit irrigation with saline water, a water conserving practice, the crop experiences simultaneous matric and osmotic stress, and it is not known if experiments designed to isolate stress effects may be used to predict crop response to simultaneous stresses (Shani and Dudly, 2001). Thus, a study was conducted wherein yields were determined under the same levels of salinity and irrigation (equipotential level) so that the different effect of the two different stresses will be detected individually.

Subjected to the stress caused by water deficit, plant not only change in morphology, structure (Danin, 1991) and metabolic process (Mehmet *et al.*, 2006), but also osmoticum that is considered to be the mechanism for maintaining turgor by osmotic adjustment (osmoregulation, Blum, 1988) in plant confronting increasing water stress (Turner and Jones, 1980).

Low water potential in soil makes difficulty for the plant to take up water, leading plant to avoid water loss, thus, modifying water uptake and loss. An overall picture of these responses must include changes in water fluxes and water relations at the whole plant and the cellular levels, such as a decreased cell water potential and stomatal closure, resulting in reduced CO_2 availability for the plant (Chaves 2009). The drought tolerance of plants can be characterized by growth response, changes in water

relations of tissues exposed to low water potential, accumulation of ions in tissues and stomatal conductance of leaves…….etc. (Blumwald et al., 2000). Plants resist low water potential and related stresses by modifying water uptake and loss to avoid soil low water potential.

Stomatal closure may assume important functions in this context since it will limit water loss and may also maintain water use efficiency at acceptable values. The ability of the plant to reach an optimal physiological compromise depends on its ability to perform photosynthesis and therefore provide carbon skeletons and energy for osmotic adjustment despite stomatal closure. A better understanding of the physiological strategy adopted by a drought resistant plant to cope with water deficit requires thorough study of the relationship between osmotic adjustment, water use efficiency, photosynthesis and transpiration (Martinez et al., 2003).

The aim of the present work was to run a comparative study among two varieties of soybean. Comparison is mainly concerned with the means of adjustments under major ecological stresses (namely: drought, salinity at equipotential levels). One of such means is changes in water relations of plant which includes:

transpiration, stomatal resistance, transpiration efficiency, relative water content, dry matter content and osmotic pressure and changes in chlorophyll a and b content, ratio and their stability to heat mainly as the simplest one of osmo-metabolic adjustments means.

MATERIALS AND ETHODS:

In this work seeds of two Soybean varieties were tested including c.v. Giza 111 and c.v. Giza 21. These varieties were obtained from Crop Science Department, Faculty of Agriculture, Assiut University.

Adjustments of drought levels (matric potential, Ψm):

Different levels of soil matric water potential, ψm: 0(control), -0.1,-0.3,-0.5, -0.7, -0.9 & -1.1MPa can be obtained by characteristic curve data of soil moisture desorption curve achieved by the pressure plate technique (Richards 1947).

Adjustments of salinity levels (osmotic potential, ψ_s) (at a fixed sodium adsorption ratio (SAR=1/20)):

Different osmotic water potential levels were prepared by using solutions of ($NaCl+CaCl_2$) in concentrations that yield different osmotic potentials (Ψ_s) at a definite SAR ratio (1/20): Ψ_s levels were chosen at the same ψ_m levels: 0 (control), -0.1, -0.3, -0.5, -0.7, -0.9 & -1.1 MPa. The concentrations of NaCl and $CaCl_2$ in solutions prepared are based on calculations explained by El-Sharkawi (1968).

Previously weighed one-liter volume plastic pots lined with double polyethylene bags were used in growing the experimental plants. Each pot was filled with 1000 gm air dry soil (sand/clay 1:2 v/v). Two plants were allowed to grow in each pot as an experimental unit. The soil water content was maintained near field capacity most of the time, during germination and growth, until the treatment was planned to start. This was achieved by a periodic (daily or twice daily, if necessary) adjustment of the weight of the whole System (the pot and its contents) by watering distilled water using four replicates for each treatment. The plants were kept in the Greenhouse during experimentation to secure mild climatic conditions.

The content of chlorophyll a and chlorophyll b in both fresh and heated samples was calculated by using the Mackinney equations given by Vishniac (1937).

The osmotic pressure (atm) was determined by the cryosopic method of Walter (1949).

Experimentation was carried out on the vegetative stage only. Statistical analysis of randomized complete block desigen according to Gomez and Gomez (1984) of experimental data took place. The significant means of any trait studied were compared using LSD at 5% probability level according to Waller and Duncan (1969).

RESULTS:

I) A-Transpiration rate, Stomatal resistance and Transpiration efficiency:

Data are shown in figures 1 and 2:

Transpiration generally corresponded to diurnal fluctuations in climatic factors.

In Giza 111 (fig. 1), diurnal pattern of transpiration rate showed a positive increase response to increasing of both temperature and VPD of air during the early part of the day, reaching a maximum at mid-day period (2 p.m.) and in the late afternoon, a decline in transpiration rate deviated from the effect of steadily higher (more or less constant) temperature, VPD. Otherwise, imposing water stresses appreciably affected the magnitude of transpiration. There is a decrease in transpiration rate at the moderate (-0.5 & -0.7 MPa) and sever (-0.9 & -1.1 MPa) water potential levels compared to control (untreated, well-watered plant) and high water potential levels (-0.1 & -0.3 MPa) in both matric and osmotic that exert higher transpiration rate. In Giza 21 (fig. 2), the previous manner differs completely. The highest transpiration rate was at 8-10 am and a gradual decrease in mid-day period (12-4 pm). This clearly indicates the role of stomatal control at this time of day. Moreover, under matric water potential stress this variety shows a different magnitude of transpiration rate. There is a wide gap between transpiration rate at the severe potential level (-0.9 & -1.1 MPa) and other levels. This gap is due to the steadily high transpiration allover day time. In Giza 111 the maximum stomatal resistance was at 6 pm under both type of water potential stresses, however, there is a relative sharp peaks at (10 am & 2 pm) but still lower than that at 6 pm and that explain the lowest transpiration rate at this time of the day. In Giza 21 the stomatal resistance was low in the early morning and increased gradually in response to increasing air temperature and VPD and this is parallel to that decrease in transpiration rate. The constant very low resistance was under low soil water matric potential levels (-0.9 & -1.1 MPa), while at the osmotic potential the lowest resistance was at the moderate levels (-0.5 & -0.7 MPa) and was the highest at severe stress (-0.9 & -1.1 MPa).

That shows a different response in these cultivars under the equipotential levels of two different type of water stress (matric and osmotic water stresses).

The transpiration efficiency, expressed as an increase in dry matter per gram water transpired in shown in table (1). This ratio is an expression of the plant ability to build up material in return to water consumed by transpiration. The positive value indicates accumulation of dry matter and the negative values are due to decrease in dry matter. In Giza 111 dry matter accumulation in response to transpired water was the maximum especially in unstressed plant and the highest potential levels (-0.1 and -0.3 MPa under Ψs and -0.1, -0.3 and -0.5 MPa under Ψm) but this ability to build up material slightly decrease at low water potential levels of the two different water stress.

In Giza 21, the only positive value of transpiration efficiency was belonging to (-0.1 & -0.7 that may be critical level) in both Ψm and Ψs, while the other levels have the negative one that may indicate a tendency to increase respiration over photosynthesis rate.

Otherwise the negativity (-22 & -24) increases in the lowest matric water potential stress (-0.9 & -1.1, respectively).

Statistically, as shown in Table 1, all single factors had significant effects on transpiration rate and stomatal resistance of the two cultivars.

variety	Giza 111				Giza 21			
levels	Transpiration rate		Stomatal resistance		Transpiration rate		Stomatal resistance	
(-MPa)	Ψm	Ψs	Ψm	Ψs	Ψm	Ψs	Ψm	Ψs
Cont	5.0a	5.0a	5.4a	5.4a	3.2a	3.2a	24.4a	24.4a
0.1	4.3a	8.0b	12.4b	4.8a	5.2a	7.0b	34.8b	16.0b
0.3	2.4ab	3.2c	11.8b	13.1b	4.7a	6.6b	35.0b	12.7b
0.5	0.9c	2.6d	9.6c	12.8b	7.3b	6.8b	17.1c	3.8c
0.7	1.7d	3.0c	9.6c	13.3b	6.9b	4.7a	21.7a	5.2c
0.9	1.9d	2.5cd	7.4d	25.0c	24.7c	9.6d	5.8d	25.0d
1.1	1.7d	2.1cd	3.1e	9.9d	25.0c	6.9b	4.3d	31.8e

Table 1: Mean values of the effect of water stress (decreased matric and osmotic water potential (Ψm & Ψs) and in the two cultivars investigated (values with the same symbols are not significantly different in the vertical order).

I) B- Relative water content and dry matter content:

Data are shown in Figures 3 and 4:

The pattern of diurnal fluctuation in RWC of Giza 111 (fig. 3) indicates suffering a progressive decrease in turgidity under reduced matric water potential levels especially at the severest ones (-0.9 & -1.1 MPa) while the control (unstressed) plant has a higher turgidity allover daytime, but under the same level of stress in case of osmotic water potential all plants exhibit the same trend of the control and allover daytime. In Giza 21 (fig. 4) under both water potential stresses, nevertheless sever potentials, plants tend to have relatively high turgor early in the morning after which there was a progressive decrease in turgidity until 12 Noon recovering turgidity in the afternoon and the evening reaching values even higher than that in the morning. This behavior is the same in unstressed ones. An obvious different manner appears in the high level of the matric potential stress, where the sharp decrease in turgidity and unchangeable trend allover daytime this may be apparently a reflection to changes in transpiration rate.

However, transpiration rate is not the sole factor affecting RWC as the rate of water absorption from the soil (related to both efficiency of plant root and soil water availability) is equally important. In unstressed plant of Giza 111, dry matter content has constant value during the early time of the day till 2 pm then tends to decline in the evening so the net productivity (yield) is not as high as that at the beginning. There is a different manner of level -0.1 MPa in the two different stresses (Ψm & Ψs). At Ψm = - 0.1 lowest dry matter content was recorded but under osmotic stress of the same level the highest dry matter rose over the other levels. Contrary with this levels -0.3 MPa that induce low dry matter content in case of Ψs and tend to slightly increase dry matter accumulation in case of Ψm. Generally, the other levels under the two different stresses (Ψm & Ψs) are relatively similar in their trend. Applying water stress, whatever its type, causes a great lowering in dry matter accumulation in Giza 21 this will be obvious when compared with the untreated plant that ever has the highest value allover daytime.

I) C- Osmotic pressure:

Data are shown in Figures 5:

Plants tend to vary internal osmotic pressure of the sap in response to different stresses in the present study. Generally, stressed plants at all levels of stress show a trend of highly significant increase in their total osmotic pressure.

In Giza 111, under the osmotic water potential stress a slight decrease in leaf total osmotic pressure exists at -0.1 MPa on the other hand any prorgressive increase in the osmotic stress induces increase in the osmotic pressure upto -0.3 MPa above which the osmotic pressure value remains constant and terminated to increase at -1.1 MPa. Matric potential stress (Ψm) exerts a different attitude. Initial increase in the osmotic pressure at both -0.7 & -0.9 MPa followed by a constant value at the next level (-1.1 MPa). In Giza 21, generally, the osmotic pressure under both water potential stresses was higher than those of Giza 111. There is a similarity in the response of the osmotic pressure under lower water potential stresses where a slight increase in osmotic pressure followed by high increase at -0.9

& -1.1 MPa and at -1.1 MPa in case of matric and osmotic stress, respectively.

II) A- Chlorophyll content (chl a and b ratio and chlorophyll stability to heat):

Data are shown in Figures 6:

In Giza 111 increase in both stresses induce increase in chlorophyll a content even over that of control plant upto -0.7 MPa after which chlorophyll a content start to decrease, While chlorophyll b content shows different pattern compared to chlorophyll a with increasing level of stress but under Ψm chlorophyll b content tend to decline up to -0.7 MPa then begins to rise again at -0.9 MPa .

In Giza 21, there is no significant change in chlorophyll a content with increasing matric stress except at the high level of stress (-0.9 & -1.1 MPa), there was an obvious decline in chlorophyll a content. - 0.1 MPa osmotic water potential level induces increasing in chlorophyll a content.

Increasing osmotic stress upto -0.5 MPa, the content of chlorophyll a decreased. Increasing in chlorophyll a content returns at -0.7 after that return to decreases again at severe level of stress. A similar trend was observed in both types of water stress regarding chlorophyll b content, there was a tendency to increase chlorophyll b content at the lowest (-0.1 & - 0.3 MPa) and moderate level (-0.5 & -0.7 MPa) and to decline at the highest one (-0.9 & -1.1 MPa). Generally, in both cultivars the probability of the distraction (breaking down) of photosynthetic apparatus under matric stress is higher than that under osmotic stress as indicated by the data of chlorophyll a and b ratios.

Chlorophyll a stability to heat was unchangeable in case of Giza 111 under the matric stresses but this trend show an obvious decline at the moderate levels (-0.5 and -0.7 MPa) in case of osmotic stress.

Chlorophyll b stability to heat shows a different response to both stresses. Low water potential induce a decrease in the stability standing at constant trend in the highest levels of the osmotic stress while the matric stress exert a drastic negative effect on the chlorophyll b stability upto -0.9 MPa after which recovery in the stability occur at -1.1 MPa. In Giza 21, the stability of both chlorophyll a and b was drastically affected by the matric potential stress. There was a marked decrease in the stability by increasing the stress. But under osmotic stress there was a slight decrease when compared with that under matric stress.

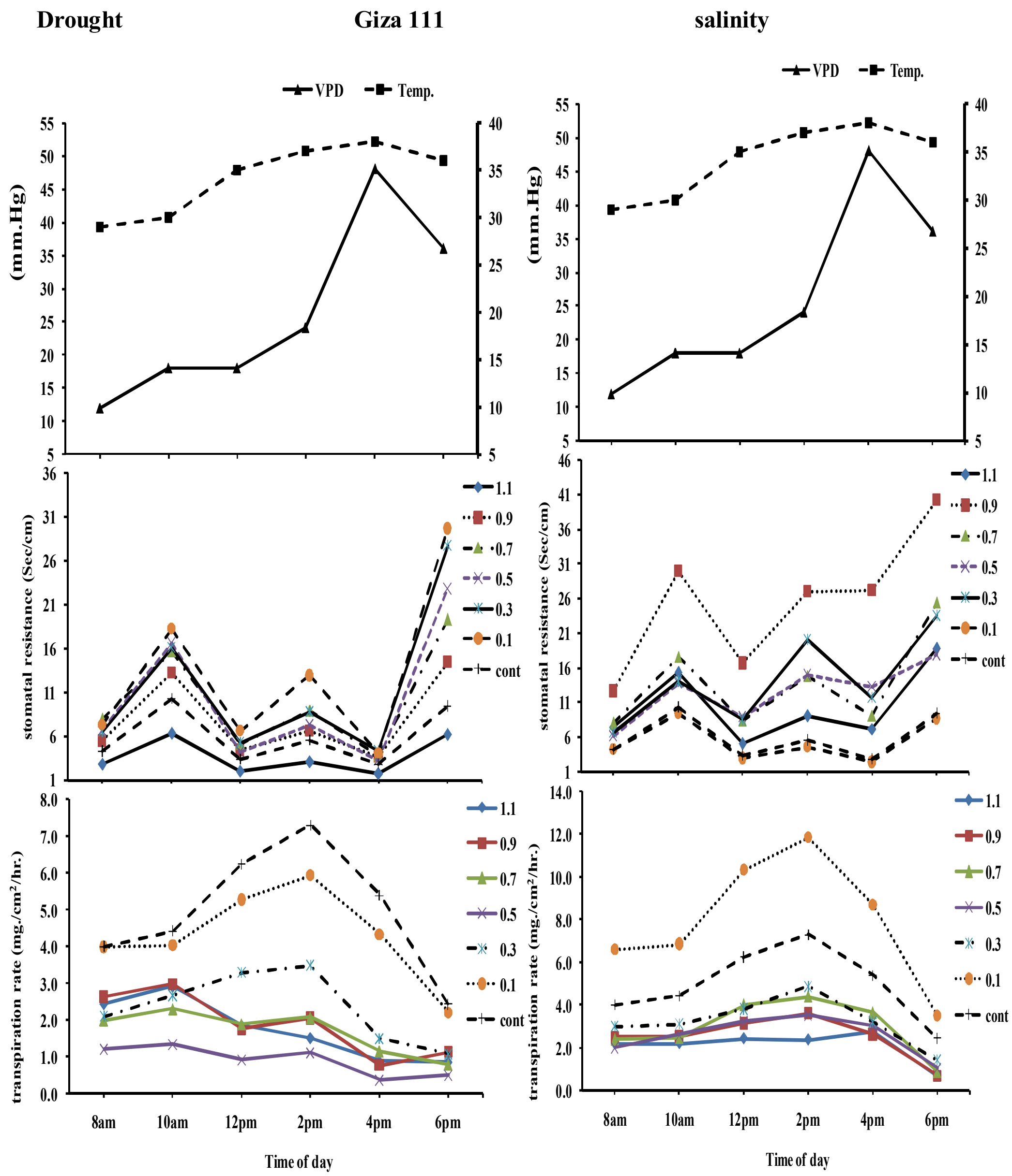


Figure (1): Diurnal fluctuations in transpiration rate and stomatal resistance in Giza 111 (soybean) with intrinsic climatic factors (vapour pressure deficit, VPD, and temperature) at different times of the day under equipotential levels of two different type of water stress (matric and osmotic water potential stresses).

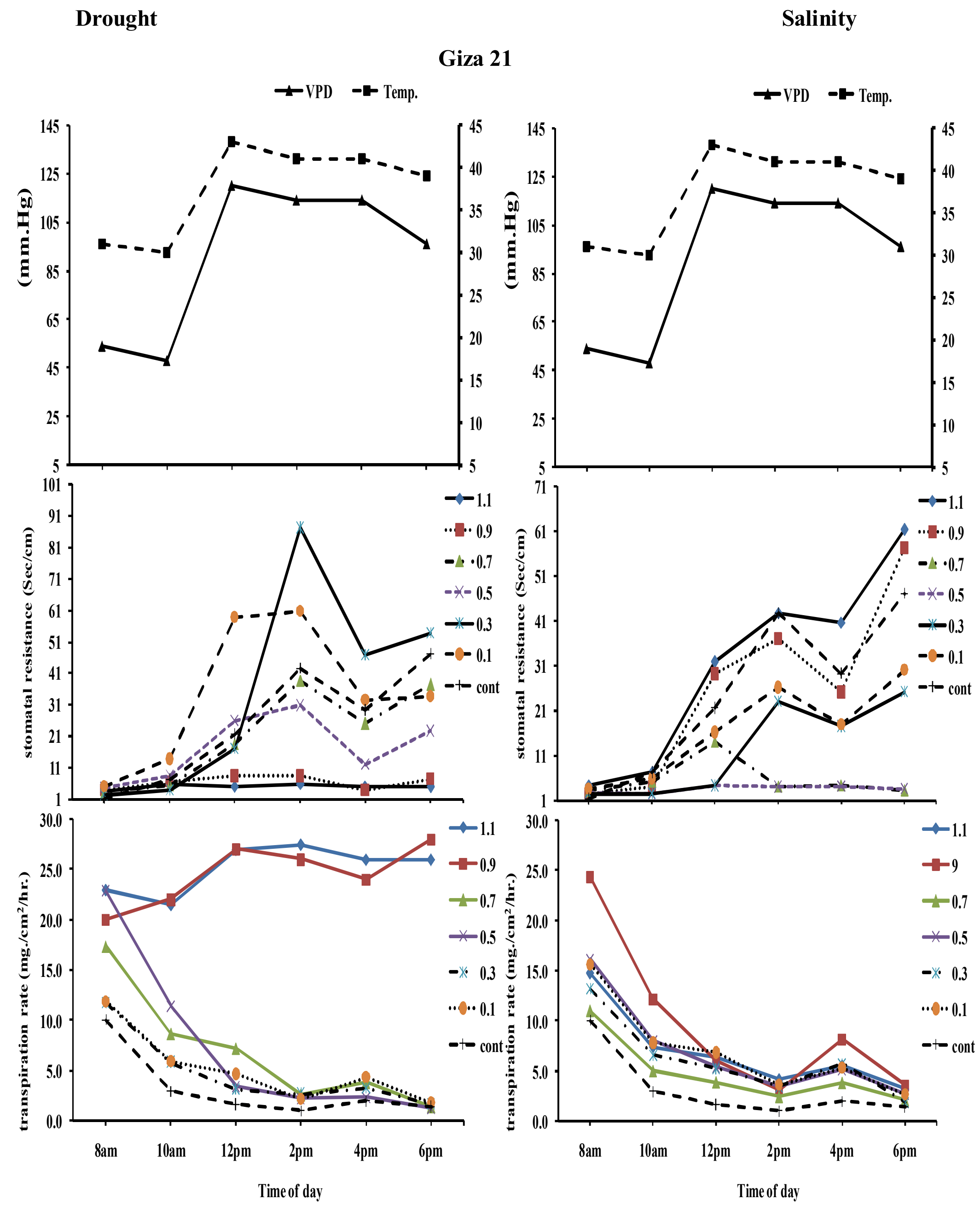


Figure (2): Diurnal fluctuations in transpiration rate and stomatal resistance in Giza 111 (soybean) with intrinsic climatic factors (vapour pressure deficit, VPD, and temperature) at different times of the day under equipotential levels of two different type of water stress (matric and osmotic water potential stresses).

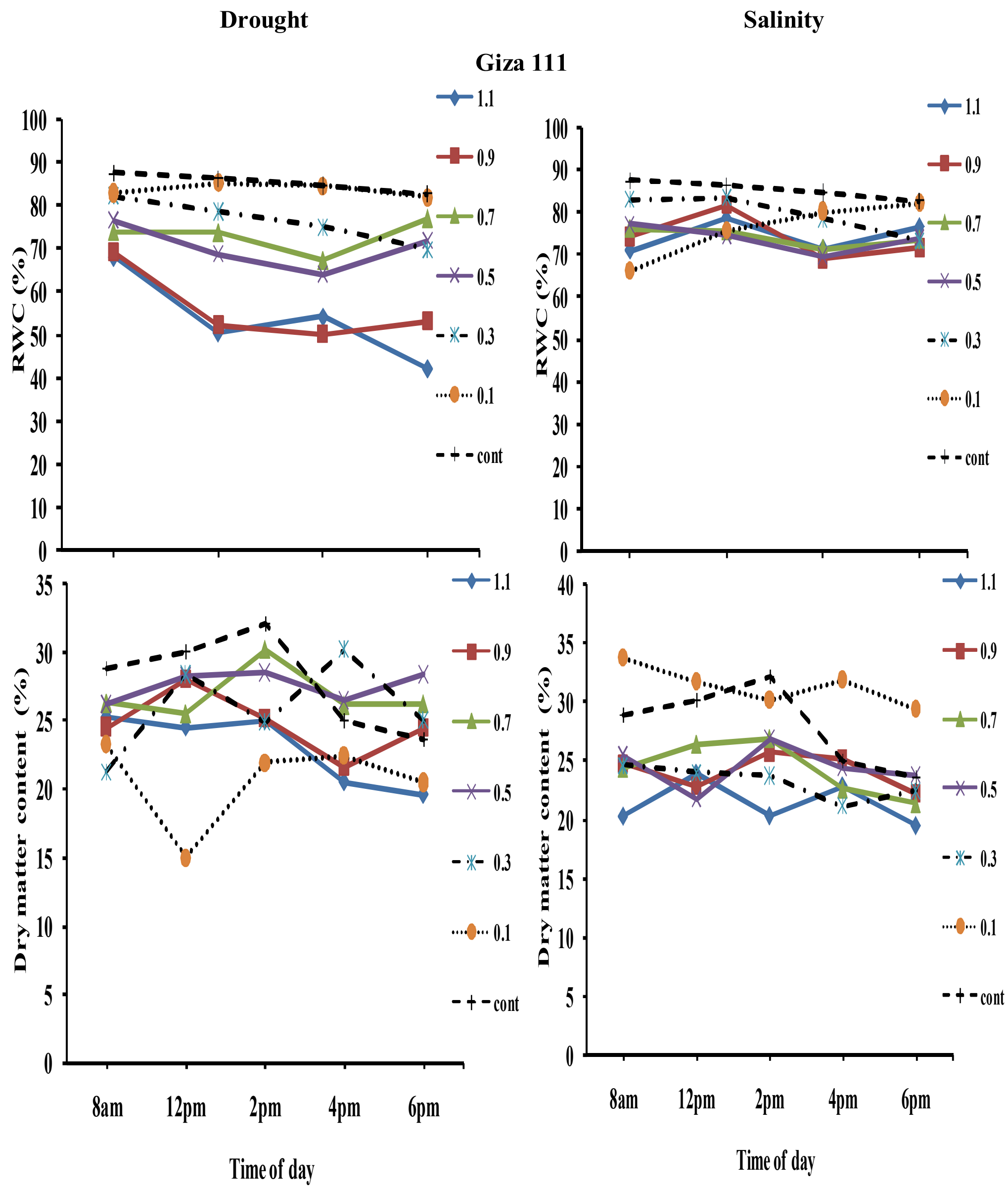


Figure (3): Diurnal fluctuation in relative water content (RWC %) and dry matter content (%) in Giza 111 at various times of the day under equipotential levels of two different types of water stress (matric and osmotic water potential stresses).

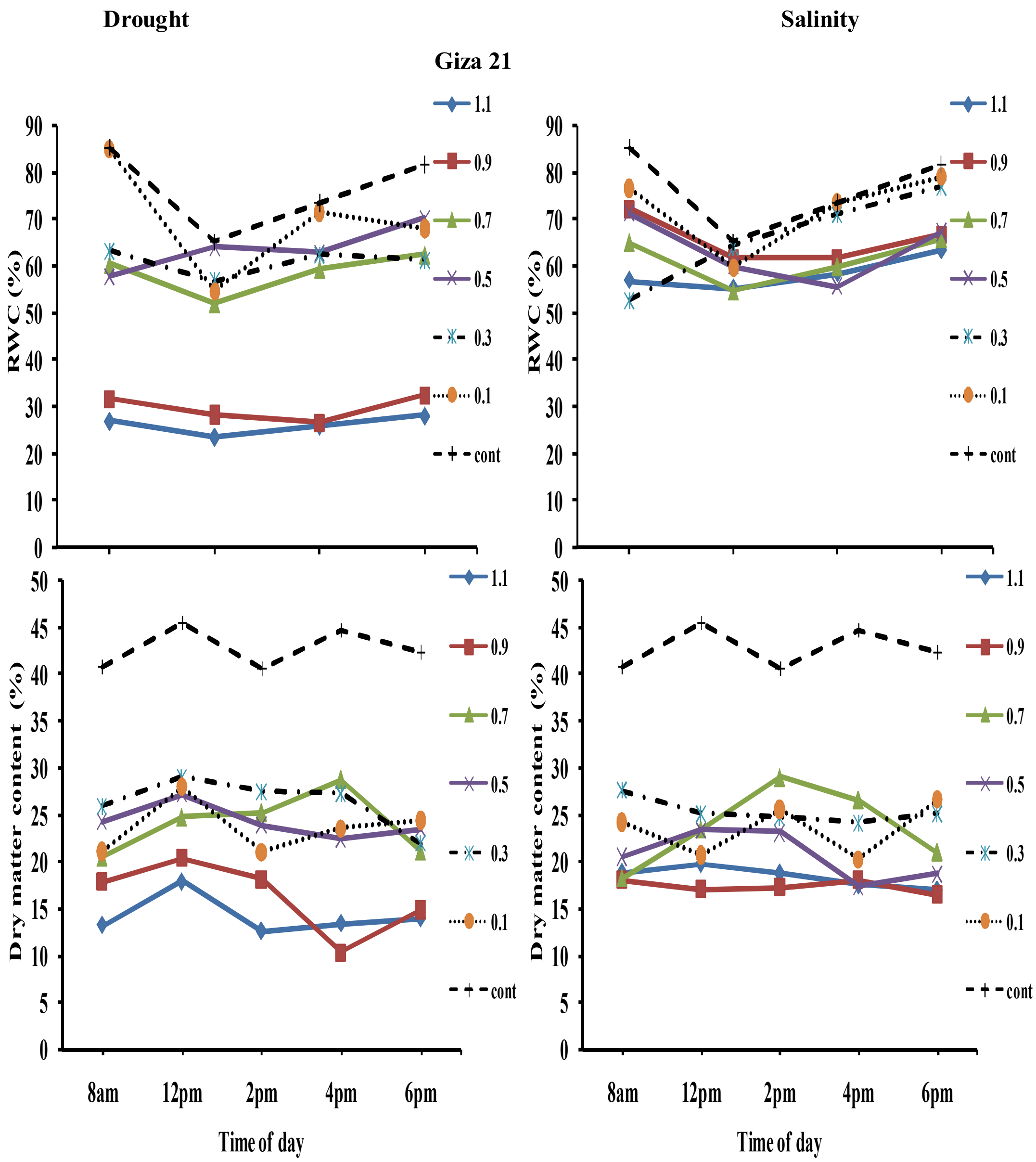
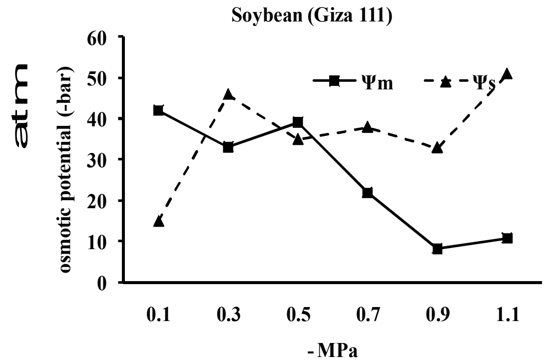
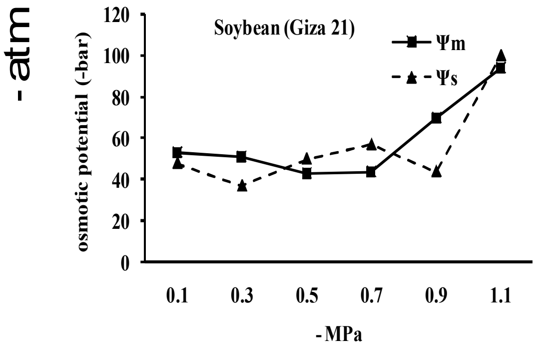


Figure (4): Diurnal fluctuation in relative water content (RWC %) and dry matter content (%) in Giza 21 at various times of the day under equipotential levels of two different types of water stress (matric and osmotic water potential stresses).



Soil water potential (-MPa)



Soil water potential (-MPa)

Figure (5): The total average of osmotic potential (atm) in investigated species under equipotential levels of two different types of water stress (matric and osmotic water potential stresses.

		Giza 111	Giza 21
	cont	14.79	1.58
Ψs	-0.1	10.68	2.36
	-0.3	12.83	-2.44
	-0.5	9.31	-1.90
	-0.7	7.03	2.76
	-0.9	7.43	-1.47
	-1.1	8.21	-10.86
Ψm	-0.1	13.23	3.32
	-0.3	12.75	-3.98
	-0.5	11.12	-0.80
	-0.7	8.98	0.69
	-0.9	8.89	-22.01
	-1.1	7.34	-24.27

Table (2): Values of daily transpiration efficiency (mg FW/g H_2O loss) in soybean cultivars at the equipotential levels of two different types of water stress (matric and osmotic water potential stresses, - MPa).

N.B.: **Negative values indicate a decrease in dry matter con**

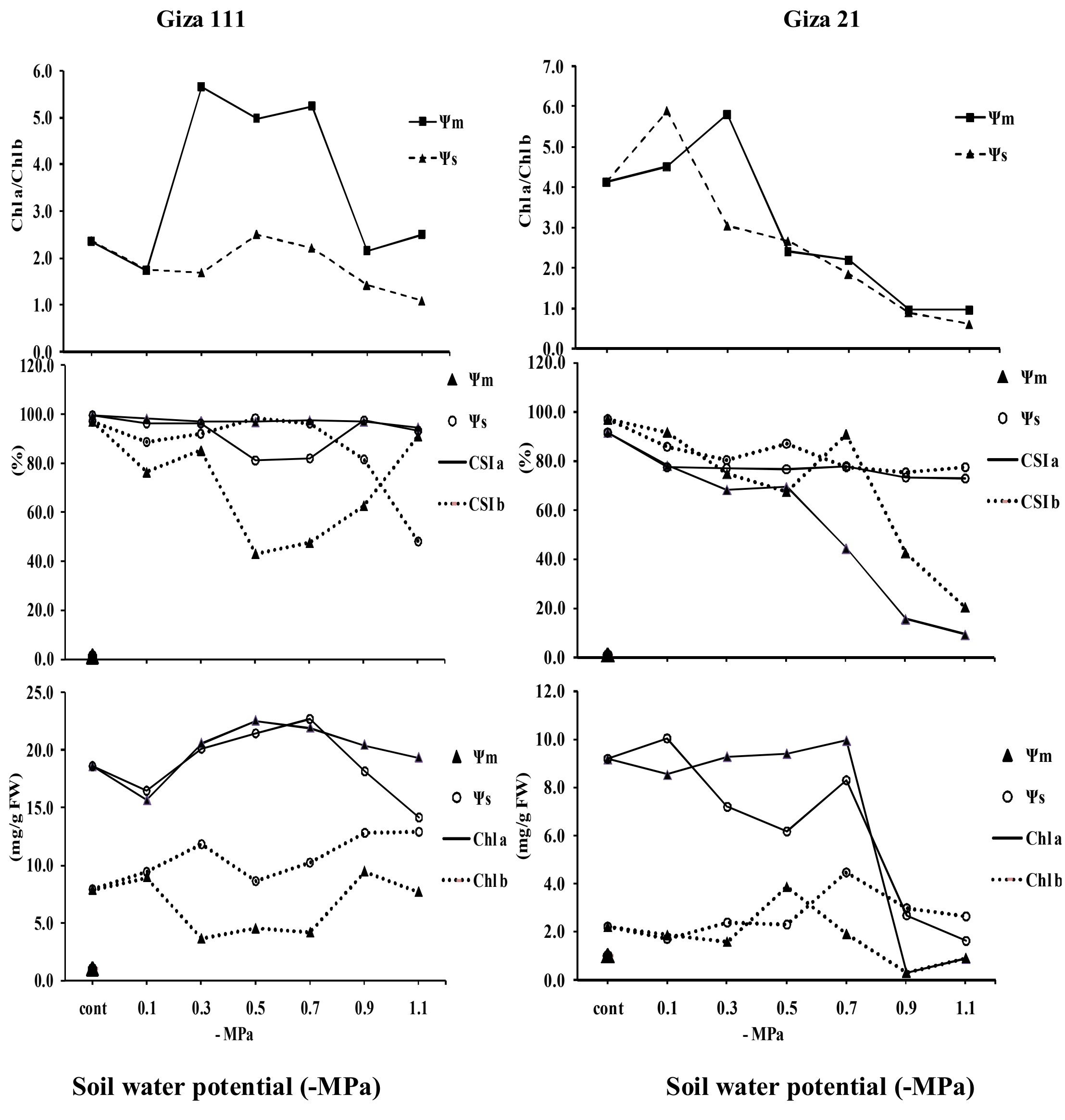


Figure (6): Changes in chlorophyll contents, chl.a/ chl.b ratio and chlorophyll stability index (CSI) to heat in soybean cultivars (Giza 111 and Giza 21) in response to, decreased soil water potentials, equipotential levels of two different types of water stress (matric and osmotic water potential streses).

DISCUSSION CONCLUSIONS:

The investigation presented aimed to understand the differences in adaptation mechanisms among cultivars of a crop plant, namely soybean, in adjusting to osmotic and matric stresses at equipotential levels and how to maintain its internal water balance and also to quantify the effects of the two different types of water stress on some physiological and metabolic characters affecting plant water relations.

Imposing osmotic and matric stress in the soil caused a reduction in transpiration rate compared to unstressed plants. There is one special case under matric stress in Giza 21 the lowest water potential induces high transpiration rate which may be attributed to the disturbance in stomatal control at these levels (Chaves and Oliveira, 2004).

Early biochemical effects of plant water deficits induce changes in leaf biochemistry that result in down-regulation of the photosynthetic metabolism may occur in response to stomatal closure under prolonged stresses. Stomatal closure is also affected by the root physiological characteristics (Flexas et al., 2008).

Tardieu et al. (1991) showed that the stomatal conductance was controlled by the root water potential through an increase of abscisic acid concentration and its role in the hormonal control of water balance (Tal and Imber 1971).

The data on changes in relative turgidity (RWC) of the two cultivars shows that the stressed plants maintained lower turgor than plants under optimal water potential. This may be due to insufficient root system to compensate for water lost by transpiration and or the unavailability of water in the soil (Donald and Harris 1973).

During water stress, some biochemical processes of plant cell change. One of such processes is the accumulation and biosynthesis of chlorophyll and consequently its stability to heat (Miyashita 2005) thus water stress induces the chlorophyll egradation and alters its stability in case of Giza 21 c.v. under matric stress of both chl a and b.

There is a great similarity between dry matter yield and photosynthetic pigments. The dry matter content of Giza 111 was enhanced by increasing of both matric and osmotic stresses especially at moderate levels. Thus this cultivar seems to be tolerant to matric and osmotic stresses at least up to -0.9 MPa.

On the other hand and interestingly, the cultivar Giza 21 seemed to be sensitive under matric and osmotic stresses. Therefore, the growth of this c.v., as shown in dry matter content fluctuation, dropped drastically at higher levels of both stresses.

Accordingly, the data of dry matter content and photosynthetic pigment revealed that c.v. Giza 111 was matric and osmotic tolerant one and this classification is based on: dry matter production and accumulation, the efficiency of photosynthetic pigment which affects positively photosynthetic assimilates that may have a role in the adjustment.

In Giza 111 all levels gained a positive value of transpiration efficiency that reflect the ability of this cultivars to keep photosynthesis near its normal rate and hence its associated metabolic sequences, leading finally to accumulation of the metabolic products as dry matter content with minor sacrifice of water loss by transpiration.

Trails of Giza 21 to survive by increasing its osmotic potential and exert adaptive response which serves to reduce its transpintional water loss as well as increasing the potential gradient between leaves and root medium (El-Sharkawi et al., 1988) especially at the low matric water potential, however, the transpiration rate at these levels was still very high even over the unstressed plant.

Under such mentioned levels, Giza 111 c.v. can tolerate and doesn't have to increase its osmotic potential as high as in Giza 21. Many literatures interest in this field illustrate that the osmotic potential of leaf is a criterion of (indication to) the omotic adjustment so that Giza 21 has a higher capacity for osmotic adjustment than Giza 111.

Several authors consider that the role of osmotic adjustment (OA) is to maintain turgor to sustain normal metabolic processes and stomatal conductance but this strategy does not appear to be efficient in Giza 21 since biomass production was lower in this cultivar exhibiting the highest capacity for OA. This may be related to findings of Turner and Jones (1980) who reported that the energetic cost of OA could impair growth, as well as to several experimental evidences indicating that turgor maintenance is not necessarily sufficient to allow cell elongation (Neumann, 1995). Thus, if biomass production is considered as a criterion for water stress resistance, OA is probably not a valuable process in Giza 21.

The data presented is, therefore, valuable in choosing cultivars suitable for desert condition.

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الانضباط في التوازن المائي الداخلي لسلالتين من نبات فول الصويا تحت النقص في الجهد المائي للتربة

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استهدف موضوع البحث دراسة تأثير نقص الجهد المائي (سواء بواسطة نقص الماء او زيادة الملوحه) على العلاقات المائية لسلالتين من نبات فول الصويا، والفرق بين تأثير كلاً من الجفاف والملوحة على الانضباط في التوازن المائي الداخلي ومدى تأقلم هذه السلالات للظروف الصحراوية. وقد شملت العوامل المختبرة في هذه الدراسة ما يلي:

تحليل منحنيات النتح والتغيرات النهارية له والاستجابة للعوامل المناخية المؤثرة.

دراسة التغيرات في المحتوي اليخضوري لكل من كلوروفيل (أ) وكلوروفيل (ب) ونسبة الاول للثاني ومقاومة نوعي اليحضوري للحرارة.

دراسة التغير في التركيز الاسموزي كمؤشر للانضباط الاسموزي.

تقييم إحصائي لمعنوية تأثير ودور العوامل المنفردة.

وقد أظهرت الدراسة ما يلي:

السلالات تستجيب بدرجات متفاوته لما يفرض عليها من نقص الجهد المائي.

نقص الجهد المائي في التربة يؤدي إلي انخفاض معدل النتح. كما أبدت النباتات المختبره استجابة واضحة للنقص في الجهد المائي من حيث محتواها من مادة اليخضور النباتي (كلوروفيل أ و ب) ، حيث كان هناك زيادة فالمحتوي اليخضوري في جيزا ١١١ ونقص في جيزا ٢١ وقد ترتب علي هذه الزياده في جيزا ١١١ زيادة في نشاط عملية البناء الضوئي مما ادي الي زيادة نواتج البناء الضوئي (كما تعكسها التغير في محتوي المادة الجافة للأوراق). وقد اتضح من حساب كفاءة استخدام النبات للماء (معدل الزيادة في الوزن الجاف مقابل كل جرام من الماء المفقود في عملية النتح) إن جيزا ١١١ له قابليه لبناء وتراكم المادة الجافة تحت مستويات من نقص الجهد المائي في التربة علي عكس جيزا ٢١ الذي تبين التأثير السلبي لنلك المستويات من الإجهاد المائي علي كفاءة استخدام النبات للماء. وهذا يجعلنا نفضل زراعة جيزا ١١١ في البيئة الصحراوية الجفافيه أو الأراضي المستصلحة المحتوية علي أملاح.