Improving Water Use Efficiency and Yield of Maize (*Zea mays* L.) by Foliar Application of Glycinebetaine under Induced Water Stress Conditions

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T WO FIELD experiments were conducted in a sandy soil in the extension field in El-Kassasein, Ismailia Governorate, Egypt during 2007 and 2008 summer seasons. The work aimed to study the effect of five levels of glycinbetaine *i.e.*, 0, 5, 10, 15, 20mM/fad(1ha=2.381fad) on the response of SC 10 maize hybrid to three rates of drip irrigation water (1.00. 0.80 and 0.60 of the estimated crop evapotranspiration, which represented 2625, 2100 and 1575 m³water/fad, respectively). The most important findings could be summarized as follows:

Irrigation by 1575 m^3/fad instead of 2625 m^3/fad reduced significantly ear leaf blade area, total chlorophyll, relative water content and leaf water potential, except the content of GB in leaves which was significantly increased in both seasons. Meanwhile, increasing the level of glycinebetaine (GB) up to 15 mM/fad increased these traits and the content of GB in leaves compared with their untreated analogues.

Decreasing the amount of irrigation water from 2625 to 1575 m^3 /fad reduced significantly the grain yield, protein yield and water use efficiency (IWUE). While, the relative increase percentages due to application of 15mM GB/fad compared with zero GB were 28.47 and 25.30%, 54.53 and 47.25% and 27.61 and 25.10% for these traits in both seasons, respectively.

The interaction between both studied factors showed that under moderate water stress condition (2100 m³/fad) without GB addition the responses of these traits were only 11.59 and 10.77 ardab/fad, 135.29 and 119.69 kg/fad and 0.773 and 0.718 kg m⁻³ compared with 14.31 and 13.49 ardab/fad, 195.07 and 176.25 kg/fad and 0.954 and 0.899 kg m⁻³ when the concentration of GB was increased to 15mM GB/fad in both seasons, respectively.

Keywords: Maize, Glycinebetaine, IWUE, Drought, Evapotranspiration.

Maize (*Zea mays* L.) is one of the most important cereal crops, which plays a critical role in animals and human feeding not only in Egypt but also in, almost, all countries.

The total maize consumption has been increased drastically due to the overgrowing population. Improving maize productivity can be achieved by breeding high yielding varieties and by application of improved agro-techniques. Water stress-associated with high temperature is often considered to be a limiting factor in maize (*Zea mays* L.) grown under arid and semiarid regions. Drought has different effects on grain yield depending on the developmental stage at which it occurs. It has been reported that maize is relatively tolerant to water stress in the vegetative stage, very sensitive during the period of tasseling, silking, and pollination and moderately sensitive during the grain-filling stage (Shanahan & Nielsen, 1987 and Abo-El-Kheir & Mekki, 2007). Increasing water stress significantly decreased relative water content, chlorophyll content, leaf water potential (Shlemmer *et al.*, 2005 and Premachandra *et al.*, 2008), number of grains/ear, 1000-grain weight and grain yield (Muhammad *et al.*, 2001). Thus water stress is the most important limitation on corn productivity in arid and semiarid regions.

Accumulation of solutes, either actively or passively, is an important adaptation mechanism for plants in response to osmotic stress. The accumulation of stress metabolites like proline, sugars, amino acids and betaines to maintain structural and metabolic integrity, occurs in response to drought and other stresses. Glycinebetaine (N, N and N- trimethylglycine) is accumulated by many species of Gramineae, Amaranthaceae, Malvaceae and Poaceae families. Glycinebetaine (thereafter referred to as betaine) is a common compatible solute in many different organisms, including higher plants (Grote *et al.*, 1994 and Rhodes & Hanson, 1993).

Using foliar application of glycinebetaine (GB) protects the plant by acting as an osmolytic and hence maintaining the water balance between the plant cell and the environment and by stabilizing macromolecules during cellular dehydration and at high salt concentration is a major goal for improving drought tolerance of plants in arid zones as in Egypt. Moreover, exogenous application of betaine to leaves or roots has been shown to increase the tolerance to various stresses of several species of plants, including both natural accumulators and nonaccumulators (Mäkelä et al., 1996 and Allard et al., 1998). It has been shown that GB, when applied to foliage, is translocated from leaves to other plant parts within several hours (Mäkelä et al., 1996), where it acts as a non-toxic cytoplamic osmolyte and plays a central role in the protection of macrocomponents of plant cells, such as protein complexes and membranes, under stress conditions (Martin et al., 1997 and Jagendrof & Takab, 2001). It has also been reported that exogenous glycinebetaine led to increase photosynthetic activity, leaf area, leaf water potential, water use efficiency, total chlorophyll, relative water content and grain yield when it was applied to maize, sorghum, wheat and barley (Agboma et al., 1997; Nayyar & Walia, 2004; Abd Alla Kotb, 2005; Abd Alla Kotb & Gaballah, 2007 and Nawaz & Ashraf, 2007). When 12 mMGB/fad was applied to barley plant under water stress, the flag leaf blade area, total chlorophyll, relative water content and

grain yield were increased by 23%, 35%, 30% and 24%, respectively (Abd Alla Kotb & Gaballah, 2007).

The present investigation aimed to investigate the effect of foliar application of glycinebetaine to improve drought tolerance of maize grown under induced water limited conditions.

Materials and Methods

Two field experiments were conducted in a sandy soil in the extension field in El-Kassasein, Ismailia Governorate, Egypt $(30^{\circ} 58' \text{ N}, 32^{\circ} 23' \text{ E} \text{ and } 10\text{m}$ above mean sea level) during 2007 and 2008 summer seasons. The study aimed to find out the effect of five levels of GB(0, 5, 10, 15, 20mM/fad) on SC 10 maize hybrid under three amounts of irrigation water (1.00. 0.80 and 0.60 of the estimated crop evapotranspiration) using drip irrigation system. A split plot design with three replicates was used in each season. The irrigation treatments and the levels of GB were randomly allocated in the main and sub-plots, respectively.

Three amounts of irrigation water were calculated as 0.6 (IR1), 0.8 (IR2) and 1.0 (IR3) of the estimated crop evapotranspiration (Etc). Maize plants were given 22 irrigations at 4 days intervals starting after 24 days from sowing. In the two growing seasons, the amount of water needed for each irrigation was calculated according to the crop coefficient (Kc) and the daily reference potential evapotranspiration (ETo). The latter was determined according to the Penman-Monteith equation (Allen *et al.*, 1998) depending on the predicted climatic factors at each irrigation time and the growth stage of maize plant. As recommended by Allen *et al.* (1996) and Neale *et al.* (1996), the FAO Kc of maize plant were 0.40 for initial stage, 0.80 for crop development stage, 1.15 for mid-season stage and 0.70 for last-season stage. At the end of the last irrigation, the quantity of water applied for each of the three irrigation treatments was calculated according to the total amount of water added in the 22 irrigations for the two seasons. The average amounts of water during the two growing seasons were 1575, 2100 and 2625m³/fad for the irrigation treatments, respectively.

Levels (0 GB: spray with tap water, 5mM equal to 0.525kg/fad, 10mM equal to 1.050kg/fad, 15mM equal to 1.575kg/fad and 20mM equal to 2.1kg/fad) were foliar applied in 80 liter water/fad after 28, 48 and 68 days from sowing.

Some physical and chemical properties of the upper of 60 cm layer of the experimental field soil as well as the predicted monthly climatic data at Ismailia region during the growing seasons of corn are presented in Tables 1 and 2, respectively. Soil analysis was done at Institute of Efficient Productivity Laboratories. Soil bulk density was determined by a classical method, using cylinders 100 mm wide and 60 mm height according to Grossmann & Reinsch (2002), while both field capacity and wilting point were determined following the method of Cassel & Nielsen (1986).

Soil depth (cm)	Coarse sand (%)	Fine sand (%)	Silt (%)	Clay (%)	Texture	Soil bulk density (g cm ⁻³)
0-60cm	65.20	26.92	4.58	3.30	sandy	1.71
Soil	Field	Wilting		Organic		EC
depth	capacity	point	pН	matter		$(d\mathbf{S} \mathbf{m}^{-1})$
(cm)	(%)	(%)		(%)		(us m)
0-60cm	7.42	1.51	7.7	0.18		0.37

 TABLE 1. Soil physical and chemical properties of the experimental field soil over the two seasons (Source: Institute of Efficient Productivity Laboratories).

 TABLE 2. Monthly climatic data at Ismailia Governorate during the growing periods of corn.

Months		Avera	age tem	peratu	Avera	ge RH	Average			
	Minimum		Maximum		Average		(%)		wind speed (km/h)	
	2007	2008	2007	2008	2007	2008	2007	2008	2007	2008
May	15	15	30	27	22.5	21	45	50	15	14
June	20	22	31	34	25.5	28	52	52	12	13
July	24	24	33	35	28.5	29.5	55	64	13	13
August	25	23	35	36	30	29.5	59	68	11	12
September	24	23	36	32	30	27.5	64	61	11	11

Data collected from Agriculture Research Center Meteorological Station in Ismailia RH:Relative Humidity.

The sowing date was 25 May in both seasons in hills 20 cm apart. The subplot area was 16.8 m² included 6 rows of 4 m long and 70 cm apart. The preceding crop was lupine in the two growing seasons. To ensure full germination, 27mm of irrigation was applied to the all field area at planting. In addition, 37mm was applied at 20days for complete establishment of seedlings. Irrigation was scheduled every 4 days throughout the growth period. Twenty days after sowing, maize plants were thinned to one plant/hill. Nitrogen fertilizer was applied at a level of 120 kg N/fad as ammonium sulphate (20.5% N) in four equal doses, every 12 days from 20 days after sowing. Phosphorus fertilizer was applied at a level of 100 kg P_2O_5 /fad as calcium superphosphate (15.5 % P_2O_5). Potassium fertilizer was applied at a level of 50 kg K_2O /fad as potassium sulphate (48 % K_2O). Phosphorus and potassium fertilizers were applied before sowing in all treatments. The other agronomic practices were done as recommended.

At 85 days from sowing, five plants were randomly taken for estimating the vegetative growth characters as follows:

- 1- Ear leaf blade area (cm^2) .
- 2- Total chlorophyll (μ Mm⁻²), it was determined using the Minolta SPAD-502 chlorophyllmeter according to Markwell *et al.* (1995).

- 3- Leaf GB content (μ g/g fresh weight), it was determined according to Gricve & Grattan (1983).
- 4- Relative water content (RWC): The relative water content was determined according to Schonfeld *et al.* (1988), where the fresh weight of twenty discs, from the youngest fully expanded leaf, was determined within 2 hr after excision. Turgid weight was obtained after soaking the discs for 16 to 18 hr in distilled water. After soaking, discs were immediately and carefully blotted dried with tissue paper prior to the determination of turgid weight. Dry weight was obtained after drying the discs sample for 72 hr at 70°C. Relative water content was calculated using the following equation:

$$RWC = \underline{Fresh \ weight} - dry \ weight} \qquad x \ 100$$

Turgid weight- dry weight

5- Leaf water potential $(-\psi)$, it was determined according to Edward (1967).

At harvest (120 days from sowing), the plants of the fourth and fifth rows $(5.6m^2 \text{ areas})$ of each plot were used to determine:

- 1- Number of ears/plant.
- 2- Number of grains/ear.
- 3- 100-grain weight (g).
- 4- Grain yield (ardab/fad), it was adjusted to 15.5% moisture content.
- 5- Grain N content (%), it was measured using the modified micro-Kijeldahl apparatus as described by A.O.A.C (1980).
- 6- Protein yield (kg/fad), It was calculated from multiplying grain yield in Kg/fad with grain nitrogen content and with 6.25.
- 7- Irrigation water use efficiency (IWUE) in kg m^{-3} .

It was calculated as IWUE= GY /IR x 100, where GY is grain yield (kg/fad) and IR is the amount of applied irrigation water (m^3/fad) for each irrigation treatment.

The analysis of variance and least significant differences (LSD at 5% levels) were used according to Steel *et al.* (1997).

Results and Discussion

Growth

The results in Table 3 indicate that each decrease in the amount of irrigation water from 2625 to 1575 m³/fad decreased significantly and gradually the averages of ear leaf blade area, total chlorophyll, relative water content and leaf water potential but the content of GB in leaves was significantly and gradually increased in both seasons. The relative reduction percentages due to water stress were 40.41 and 43.93%, 39.90 and 41.80%, 46.54 and 46.63% and 17.78 and 20.70% for ear leaf blade area, total chlorophyll, relative water content and leaf water potential in both seasons, respectively. In the same trend, similar results were obtained by Shlemmer *et al.* (2005) and

Premachandra et al. (2008) where they indicated that water stress reduced relative water content, chlorophyll content, and leaf water potential than their unstressed maize plants. It seems evident that subjecting maize plants to water stress, through reducing the amount of irrigation water reduced all growth attributes; probably due to impairing photosynthetic process which could have been decreased by the drastic decrease of leaf relative water content. These results are in agreement with those obtained by Talukder (1987), Abd Alla Kotb (2005) and Abd Alla Kotb & Gaballah (2007). Concerning to the increment of GB content in leaves due to water stress, it was reported that GB is accumulated by many species of Gramineae in response to drought and other stresses (Rhodes & Hanson, 1993 and Grote et al., 1994). This increment in GB during water stress can not alleviate the negative effects of water lake on growth characters.

TABLE 3. Effect of water stress treatments (IR) and GB levels on growth attributes of maize at 85 days from planting.

Items	Ear leaf blade area (cm ²)		Total chlorophyll (µMm ⁻²)		Leaf GB content (µg/g fresh weight)		Relative water content (%)		Leaf water potential (-y)	
	2007	2008	2007	2008	2007	2008	2007	2008	2007	2008
Water treatments (IR)										
1575 m ³ /fad	400.17	349.73	34.27	31.39	638.95	602.71	44.52	42.02	-1.073	0.997
2100 m3/fad	534.76	484.75	45.19	41.72	612.95	578.30	62.46	58.66	-0.991	-0.916
2625 m3/fad	671.57	623.71	57.02	53.93	590.58	545.65	83.27	78.74	-0.911	-0.826
LSD 0.05	1.37	2.7	1.28	1.02	7.20	13.1	1.98	3.01	0.02	0.01
RD %	40.41	43.93	39.90	41.80	-	-	46.54	46.63	17.78	20.70
RI%	-	-	-	-	8.19	10.46				
-GB levels					1		1	1		
0mM/fad	478.60	426.53	40.21	37.14	536.17	514.85	56.63	51.91	-1.104	-1.019
5mM/fad	523.07	474.41	42.90	39.59	592.13	550.78	63.36	60.25	-1.001	-0.927
10mM/fad	558.57	507.21	47.83	44.59	625.86	577.84	65.25	61.90	-0.946	-0.878
15mM/fad	595.89	544.50	52.63	49.61	649.20	603.13	67.89	64.88	-0.897	-0.812
20mM/fad	521.37	477.66	43.91	40.80	667.46	631.17	63.96	60.08	-1.011	-0.931
LSD 0.05	2.026	3.29	1.58	1.57	6.84	14.41	1.30	1.71	0.02	0.02
RI* %	24.51	27.66	30.89	33.58	21.08	17.47	19.88	24.99	18.75	20.31
Interaction (IR x GB)	*	*	*	*	*	*	*	*	*	*

a=2.381fad

RD%: Relative decrease percentage due to decreasing irrigation water amount from 2625 to 1575m3/fad

RI%: Relative increase percentage due to decreasing irrigation water amount from 2625 to 1575m³/fad

RI*%: Relative increase percentage due to increasing GB levels from zero to 15 mM/fad.

The effect of foliar application of GB on plant growth was significant in both seasons (Table 3). The highest values for growth attributes were obtained from application of 15mM GB/fad in both seasons except the leaf GB content which was increased by increasing the level of GB up to 20mM/fad in the two seasons. Moreover, the results showed that the relative increase percentages due to application of 15mM GB/fad were 24.51 and 27.66% for ear leaf blade area, 30.89 and 33.58% for total chlorophyll, 21.08 and 17.47% for leaf GB content, 19.88 and 24.99% for relative water content and 18.75 and 20.31% for leaf water potential in the first and second seasons, respectively compared with their untreated analogues. These results are in harmony with those obtained by Agboma et al. (1997), Abd Alla Kotb (2005) and Nawaz & Ashraf (2007). It is clear from the data recorded that increasing GB concentration to 20mM decreased ear leaf blade area, total chlorophyll, relative water content and leaf water potential in both seasons. This reduction of theses traits is seemed to be affected by the high concentration of exogenous glycinebetaine application. Results from other studies showed, also, that high concentration of GB could stimulate necrotic blotches on the leaves of wheat and could reduce aboveground biomass as reported by Agboma et al. (1997) and Abd Alla Kotb (2005).

Concerning the interaction between irrigation treatments and GB levels, results in Table 4 and Fig. 1 to 5 showed that both of them interacted with each other significantly for all growth analysis. Ear leaf blade area, total chlorophyll, relative water content and leaf water potential were gradually and significantly increased by increasing both of irrigation water amount and GB levels up to 15mM/fad, except the content of GB in leaves which was significantly and gradually increased by decreasing the amount of water and increasing the exogenous application of GB up to 20mM/fad in both seasons. In the same direction, Rhodes & Hanson (1993) and Grote *et al.* (1994) reported that glycinebetaine is accumulated by many species of Gramineae in response to water stress.

Under the sever water stress treatment (0.6 of the estimated crop evapotranspiration) and spray with 15 mM GB/fad, the responses of ear leaf blade area, total chlorophyll, GB content, relative water content and leaf water potential were 461.30 and 410.10cm², 40.29 and 37.27 μ Mm⁻², 666.47 and 621.80 μ g/g fresh weight, 48.04 and 46.34% and 0.990 and 0.905 (- ψ) compared with 345.30 and 293.30 cm², 29.88 and 27.01_ μ Mm², 575.97 and 551.31 μ g/g fresh weight, 38.47 and 35.63% and, 1.199 and 1.114 (- ψ) when the concentration of GB was decreased to zero in both seasons, respectively. The highest values of ear leaf blade area, total chlorophyll, relative water content and leaf water potential were obtained from 15mM GB/fad under un-stress irrigation treatment (normal irrigation) in both seasons. These results indicate, also, that maize plants responded to GB addition in both seasons under normal irrigation as well as water stress conditions.

Interactions		Ear leaf blade area (cm²)		Total chlorophyll (µMm ⁻²)		Leaf GB content (µg/g fresh weight)		Relative water content (%)		Leaf water potential (-y)	
		2007	2008	2007	2008	2007	2008	2007	2008	2007	2008
IR	GB										
	0mM/fad	345.30	293.30	29.88	27.01	575.97	551.31	38.47	35.63	-1.199	-1.114
1575	5mM/fad	385.10	338.17	32.14	29.02	618.67	573.92	44.64	42.37	-1.085	-1.021
$m^{3/}$	10mM/fad	421.20	369.83	36.02	33.10	641.57	596.91	46.05	43.32	-1.027	-0.966
fad	15mM/fad	461.30	410.10	40.29	37.27	666.47	621.80	48.04	46.34	-0.990	-0.905
	20mM/fad	387.93	337.27	33.02	30.53	692.10	669.62	45.39	42.45	-1.066	-0.981
	0mM/fad	475.30	424.10	40.30	36.68	532.37	514.71	55.79	50.44	-1.104	-1.019
2100	5mM/fad	522.80	471.60	42.81	39.14	600.07	557.40	62.62	59.49	-1.001	-0.927
m ³ /	10mM/fad	560.30	508.90	47.50	44.15	622.97	580.30	64.25	60.92	-0.949	-0.889
fad	15mM/fad	595.60	544.40	52.82	49.15	647.87	605.31	66.69	63.33	-0.895	-0.810
	20mM/fad	519.80	474.73	42.52	39.48	661.50	633.80	62.96	59.10	-1.005	-0.935
	0mM/fad	615.20	562.20	50.46	47.74	500.17	478.53	75.62	69.66	-1.009	-0.924
2625	5mM/fad	661.30	613.47	53.73	50.63	557.67	521.02	82.83	78.88	-0.917	-0.832
$m^{3}/$	10mM/fad	694.20	642.90	59.97	56.52	613.03	556.30	85.44	81.47	-0.863	-0.778
fad	15mM/fad	730.77	679.00	64.77	62.41	633.27	582.28	88.93	84.98	-0.808	-0.723
	20mM/fad	656.37	620.97	56.18	52.38	648.77	590.11	83.52	78.70	-0.960	-0.875
LSD	0.05	3.51	5.70	1.00	0.98	12.97	15.94	3.25	1.45	0.03	0.04

TABLE 4. Interaction effect between water stress treatments (IR) and GB levels on growth attributes of maize at 85 days from planting.

1ha=2.381fad



Fig. 1. Interaction effect between water stress treatments and glycinebetaine levels (GB) on ear leaf blade area.

(IR1, IR2 and IR3=1575, 2100 and 2625m $^3{\rm fad},$ respectively) 1ha=2.381fad

111a=2.5011au



Fig. 2. Interaction effect between water stress treatments and glycinebetaine levels (GB) on total chlorophyll (μ Mm-²). (IR1, IR2 and IR3=1575, 2100 and 2625m³ /fad, respectively)



Fig. 3. Interaction effect between water stress treatments and glycinebetaine levels (GB) on Leaf GB content.

(IR1, IR2 and IR3=1575, 2100 and 2625m³/fad, respectively).

1ha=2.381fad



Fig. 4. Interaction effect between water stress treatments and glycinebetaine (GB) on leaf relative water content.

(IR1, IR2 and IR3=1575, 2100 and 2625m³/fad, respectively)







From these results, it could be concluded that exogenous GB application with a proper dose helped stressed maize plants to accumulate more chlorophyll and GB *Egypt. J. Agron*. **34**, No.1 (2012)

contents and hence had higher leaf area, relative water content and leaf water potential than their untreated analogues. This could be due to a possible increase in the stability of chloroplast membranes (Mamedove *et al.*, 1991), protection of photosystem II by GB (Papageorgiou *et al.*, 1991), improved water status and reduced transpiration via effects on stomatal regulation. These beneficial effects of GB might have had improved the growth of maize plants under water-stress conditions. These results are in harmony with those obtained by Abd Alla Kotb (2005), Abd Alla Kotb & Gaballah (2007) and Quanqil *et al.* (2008).

Yield and water use efficiency

Decreasing the amount of irrigation water from 2625 to 1575 m³/fad reduced significantly the yield and its attributes in both seasons (Tables 5 and 6). The relative decrease percentages were 44.98 and 47.30%, 44.87 and 44.67%, 69.96 and 70.80% and 8.29 and 12.24% for grain yield, grain N content, protein yield and water use efficiency in both seasons, respectively. In the same trend, the results obtained by Ni (1992) referred that drought during the vegetative growth stage indirectly affected yield potential by adversely affected leaf area and photosynthetic capacity. These results are in agreement with those of Muhammad *et al.* (2001) and Abd Alla Kotb (2005).

Items	Numl ears/	ber of plant	Number of grains/ear		100-grain weight (g)		Grain yield (ardab/fad)		Grain N content (%)	
	2007	2008	2007	2008	2007	2008	2007	2008	2007	2008
Water treatments (IR)										
1575 m3/fad	0.808	0.741	217.79	200.94	15.051	14.432	9.581	8.960	1.069	1.023
2100 m3/fad	1.053	0.977	302.44	284.31	20.549	19.647	12.865	12.031	1.459	1.396
2625 m3/fad	1.387	1.287	418.67	392.89	27.957	26.809	17.414	17.002	1.939	1.849
LSD 0.05	0.011	0.006	2.58	6.65	0.08	0.17	0.30	0.16	0.080	0.025
RD%	41.75	42.42	47.98	48.86	46.16	46.17	44.98	47.30	44.87	44.67
GB levels										
0mM/fad	1.050	0.972	299.96	278.99	19.350	18.553	11.681	11.300	1.362	1.297
5mM/fad	1.076	0.998	304.89	284.19	20.488	19.570	12.803	12.294	1.435	1.369
10mM/fad	1.078	1.000	319.07	297.95	21.756	20.787	14.051	13.169	1.528	1.468
15mM/fad	1.145	1.066	334.18	313.45	23.854	22.919	15.006	14.159	1.626	1.530
20mM/fad	1.064	0.971	306.73	289.00	20.480	19.651	12.892	12.399	1.494	1.449
LSD 0.05	0.009	0.005	3.13	6.78	0.11	0.11	0.31	0.23	0.054	0.025
RI %	9.05	9.67	11.41	12.35	23.28	23.53	28.47	25.30	19.38	17.96
Interaction (IRxGB)	*	*	ns	ns	*	*	*	*	ns	ns

 TABLE 5. Effect of water stress treatments and GB levels on yield and its attributes of maize.

1ha =2.381fad

1 ardab=140kg of shelled grains

RD%: Relative decrease percentage due to decreasing irrigation water amount from 2625 to $1575 \mathrm{m}^3/\mathrm{fad}$

RI%: Relative increase percentage due to increasing GB levels from zero to 15 mM/fad.

Main effects and	Protein	yield	Water use efficiency			
interactions	(kg/f	ad)		(kg m°)		
inter actions	2007	2008	2007	2008		
Water treatments (IR)						
1575 m ³ /fad	90.11	80.60	0.852	0.796		
2100 m ³ /fad	164.83	147.52	0.858	0.802		
2625 m ³ /fad	297.25	276.06	0.929	0.907		
LSD 0.05	3.41	3.39	0.021	0.012		
RD%	69.96	70.80	8.29	12.24		
Glycinebetaine (GB)						
0mM/fad	147.05	136.50	0.775	0.745		
5mM/fad	169.86	156.82	0.849	0.810		
10mM/fad	198.75	179.35	0.928	0.869		
15mM/fad	227.24	200.99	0.989	0.932		
20mM/fad	177.41	166.65	0.856	0.818		
LSD 0.05	6.23	3.79	0.021	0.018		
RI%	54.53	47.25	27.61	25.10		
Interaction (IRxGB)	*	*	*	*		

TABLE 6. Effect of water stress treatments and GB levels on protein yield and water use efficiency of maize.

1ha=2.381fad

RD%: Relative decrease percentage due to decreasing irrigation water amount from 2625 to 1575m³/fad

RI%: Relative increase percentage due to increasing GB levels from zero to 15 M/fad.

In both seasons, the results showed that numbers of ears/plant, number of grains/ear, 100-grain weight, grain yield, grain N content, protein yield and irrigation water use efficiency were significantly affected by foliar application of GB. The results in Tables 5 and 6 revealed that 15mM GB produced the highest values for yield and yield attributes compared with the other treatments in both seasons. Foliar application of 15mM increased grain yield, grain N content, protein yield and irrigation water use efficiency by 28.47 and 25.30%, 19.38 and 17.96%, 54.53 and 47.25% and 27.61 and 25.10% compared with untreated plants in the first and second seasons, respectively. Augmentation GB concentration up to 20mM/fad resulted in a significant decrease in values of vield and vield attributes. The increments in vield and vield attributes could be attributed to the increments in leaf blade area, total chlorophyll, relative water content and leaf water potential, which in turn resulted in higher values of dry matter accumulation per unit area and consequently higher yield and its attributes.

It is clear that exogenous glycinebetaine might have had increased photosynthetic activity, leaf area, leaf water potential, water use efficiency, total chlorophyll, relative water content and grain yield as reported in maize, sorghum, wheat and barley by Agboma et al. (1997), Naryyar & Walia (2004), Abd Alla Kotb (2005), Abd Alla Kotb & Gaballah (2007) and Nawaz & Ashraf (2007), in respective order.

In contrast, increasing application of GB to 20mM/fad decreased growth, yield and its components. This may be due to toxicity of GB when accumulated in higher concentration within cells and inhibition of metabolic process (Agboma *et al.*, 1997 and Abd Alla Kotb, 2005). The obvious results indicate, also, that the content of GB in leaves was significantly and gradually increased by increasing the exogenous application of GB up to zo mM/fad.

Concerning the interaction between both studied factors in the two seasons, the results showed that both of them interacted with each other significantly for number of ears/plant, 100-grain weight, grain yield, protein yield and water use efficiency, but number of grains/ear and N content of leaves were not significantly affected.

The highest values of yield and yield attributes were obtained from applying 2625 m^3 /fad and spraying with 15mM GB/fad compared with the same amount of irrigation water and without GB addition in both seasons (Table 7 and Fig. 6 to 10). These results show, also, that with increasing level of GB in both seasons, yield and yield attributes did not respond to more than 15mM under normal irrigation or water stress and followed by a significant decrease in these traits.

Interactions		Number of ears/plant		100-grain weight (g)		Grain yield (ardab/fad)		Protein yield (kg/fad)		Water use efficiency (kg m ⁻³)	
			2008	2007	2008	2007	2008	2007	2008	2007	2008
IR	GB										
	0mM/ fad	0.782	0.720	13.673	13.260	8.490	8.017	71.93	64.11	0.755	0.712
1575	5mM/fad	0.795	0.733	14.563	13.880	9.347	8.677	82.79	72.91	0.831	0.771
m ³ /	10mM/fad	0.808	0.746	15.497	14.750	10.070	9.397	97.77	86.97	0.895	0.835
tad	15mM/fad	0.840	0.778	17.007	16.300	10.523	9.850	108.76	97.32	0.936	0.875
	20mM/fad	0.815	0.726	14.513	13.970	9.477	8.860	89.30	81.71	0.842	0.788
	0mM/fad	1.010	0.934	19.233	18.413	11.593	10.770	135.29	119.65	0.773	0.718
2100	5mM/fad	1.039	0.963	19.973	19.053	12.473	11.650	155.73	138.65	0.832	0.777
$m^{3}/$	10mM/fad	1.069	0.993	20.667	19.657	13.393	12.570	176.29	158.41	0.893	0.838
rad	15mM/fad	1.110	1.034	22.913	21.997	14.313	13.487	195.07	176.25	0.954	0.899
	20mM/fad	1.035	0.962	19.957	19.113	12.550	11.677	161.78	144.64	0.837	0.778
	0mM/fad	1.358	1.262	25.143	23.987	14.960	15.113	233.95	225.76	0.798	0.806
2625	5mM/fad	1.394	1.298	26.927	25.777	16.590	16.557	271.05	258.90	0.885	0.883
m ^{-/} fad	10mM/fad	1.357	1.261	29.103	27.953	18.690	17.540	322.21	292.66	0.997	0.935
	15mM/fad	1.483	1.387	31.643	30.460	20.180	19.140	377.89	329.39	1.076	1.021
	20mM/fad	1.341	1.226	26.970	25.870	16.650	16.660	281.14	273.59	0.888	0.889
LSD	0.05	0.016	0.012	0.20	0.19	0.53	0.39	10.80	6.56	0.038	0.037

 TABLE 7. Interaction effect between water stress treatments (IR) and GB levels on yield and its attributes, and irrigation water use efficiency of maize.

1ha=2.381fad

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Fig. 6. Interaction effect between water stress treatments and glycinebetaine levels (GB) on number of ears/plant.

(IR1, IR2 and IR3=1575, 2100 and 2625m³/fad, respectively)



Fig. 7. Interaction effect between water stress treatments and glycinebetaine levels (GB) on 100-grain weight.

(IR1, IR2 and IR3=1575, 2100 and 2625m³/fad, respectively) 1ha=2.381fad

1ardab=140kg of shelled grains





(IR1, IR2 and IR3=1575, 2100 and 2625m³/fad, respectively)



Fig. 9. Interaction effect between water stress treatments and glycinebetaine levels (GB) on protein yield.

(IR1, IR2 and IR3=1575, 2100 and 2625m³/fad, respectively)

1ha=2.381fad

1ardab=140kg of shelled grains



Fig. 10. Interaction effect between water stress treatments and glycinebetaine levels (GB) on water use efficiency. (IR1, IR2 and IR3=1575, 2100 and 2625m³/fad, respectively)

The obtained results in Fig. 6 to 10 indicate that grain yield, protein yield and water use efficiency (kg grain/m³ water) were increased up to 20.18 and 19.14 ardab/fad, 377.89 and 329.39 kg/fad and 1.076 and 1.021 kg m⁻³ with interaction 2625 m³ water/fad and 15mM GB/fad in comparison with 14.96 and 15.11 ardab/fad, 233.95 and 225.76 kg/fad and 0.798 and 0.806 kg m⁻³ by using full irrigation and without GB in both seasons, respectively.

Under moderate and severe water stress conditions (2100 and 1575 m³/fad), yield and yield attributes responded significantly up to 15mMGB/fad compared with the interaction between water stress and without application GB in both seasons. Under moderate water stress condition (2100 m³) without GB addition the responses of grain yield, protein yield and irrigation water use efficiency were only 11.59 and 10.77 ardab/fad, 135.29 and 119.69 kg/fad and 0.773 and 0.718 kg m⁻³ compared with 14.31 and 13.49 ardab/fad, 195.07 and 176.25 kg/fad and 0.954 and 0.899 kg m⁻³ when the concentration of GB was increased to 15mM GB in both seasons, respectively. In the same observation, exogenous application of GB to lowaccumulating or non-accumulating plants may help to reduce the adverse effects of environmental stresses (Mäkela *et al.*, 1996 and Yang & Lu, 2005).

These results mean that maize plants responded to GB application in the both seasons under water stress conditions as well as under un-stressed conditions. It was clear that GB played a crucial role as osmoprotectants in improving the tolerance of plants to environmental stresses. The foliar application of glycinebetaine on maize plants increased nitrogen uptake, leaf area, leaf water potential, total chlorophyll,

¹ha=2.381fad

relative water content. The increase of these traits can improved water use efficiency and both of the maize grain and protein yields. These results may explain the reported response of maize plants to GB under water stress conditions (Agboma *et al.*, 1997; Nayyar & Walia, 2004, Abd Alla Kotb, 2005; Abd Alla Kotb & Gaballah, 2007 and Nawaz & Ashraf, 2007).

Conclusion

From these results, it could be concluded that subjecting maize plants to water stress decreased significantly growth, yield and its attributes. Meanwhile, exogenous application of GB by a proper level (15m M/fad) enhanced growth, yield and its attributes. Moreover, the interactions between water treatments and levels of GB were significant, indicating that GB played an important role for minimizing the adverse effect of water stress and hence improved water use efficiency, grain and protein yields. From these previous results it could be concluded that GB acted as osmoregulating substance and enhanced the tolerance of maize plants to water stress when was applied at a level of 15mM/fad. But foliar application of GB by a higher level (20mM/fad) decreased growth and yield, probably, due to a possible inhibition of photosynthesis.

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تحسين كفاءة استخدام الماء ومحصول الذرة بالرش بالجلايسين بيتايين تحت ظروف استحداث إجهاد مائي

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أجريت تجربتان حقليتان بارض رملية بمنطقة القصاصين بمحافظة الإسماعيلية خلال موسمي ٢٠٠٧و٢٠٨٠ بهدف دراسة تاثير ٥ مستويات من الرش بالجلايسين بيتايين١٥،١٠،٥٠ و٢٠ ملليمول/فدان) على استجابة الذرة (هجين فردى ١٠) إلى ٣ معاملات رى بالتتقيط (١، ، ، ، و٦، ، من قيمة النتح- بخر لمحصول الذرة والتى تعادل ٢٦٢٥ و ٢١٠٠ و ١٥٩٥ م٣ ماء/فدان على الترتيب) ويمكن تلخيص أهم النتائج المتحصل عليها كما يلى:

أدى الإجهاد المائي (١٥٧٥م ٣/فدان) مقارنة بالرى العادى ٢٦٢٥م ٣/فدان إلى نقص معنوي لكل صفات النمو المدروسة ماعدا محتوى الأوراق من الجلايسين بيتايين والذى زاد. وقد بلغ النقص كنسبة مؤية ٢٠،٧٦، ٤٠،٤ و ٢،،١٤ % و ٣٩،٩٠ و ٢،٥٢٤ ٢،٦٣، ٤٦،٣٤ و ٢،،٧١، ٢،٠٢ % لكل من مساحة نصل ورقة الكوز، محتوى الكلوروفيل الكلى ومحتوى الماء النسبي للأوراق والجهد المائى للورقة في الموسم الأول والثاني على الترتيب. بينما ادى زيادة مستويات الجلاسين بيتاين من صفر إلى ١٥ملليمول/فدان إلى زيادة تلك المقاييس ومحتوى الأوراق من الجلايسين بيتايين إلى ٢٠،٣١ % و ٢،٠٢٠ % و ١٥،٢٠ % و ٢،٥٢٨، ٢،٠٥٩ % و ١٥،٢٠٠ % و ٢٠،٣١، % ما ما ما النفسي ومحتوى الأوراق من الجلايسين بيتايين إلى ٢٠،٣١ % و ٢،٠٢٠ % في الموسم الأول والثاني على الترتيب مقارنة بالنباتات غير المعاملة.

أدى نقص كميات ماء الرى من ٢٦٢٥ إلى ١٥٧٥م ٣/فدان إلى نقص معنوي للمحصول ومكوناته وكفاءة استخدام الماء وكان معدل هذا النقص هو ٤٤،٩٨ ٢٠،٣٠ و والبروتين وكفاءة استخدام ماء الرى في الموسم الأول والثاني على الترتيب. بينما ادى زيادة مستويات الجلاسين بيتاين من صفر إلى ١٥ملليمول/فدان إلى زيادة تلك المقاييس بمعدل ٢٨،٢٧، ٢٥،٣٠ و ٥٤،٥٥، ٢٥،٢٥ و ٤٧،٢١، ٢٥،١٠

وأوضح التفاعل بين عاملى الدراسة أنه تحت ظروف الأجهاد المائى (٢١٠٠م٣/فدان) وبدون اضافة جلايسين بيتايين فان هذه الصفات اعطت فقط (١١،٥٩ ، ١٩،٧٧ إردب/فدان و١٣،٥٣، ١١٩،٦٩ كجم/فدان و ١٩،٠،، ، ١١،٥٩ كجم/م٣ مقارنة مع ١٣،٤٩ ، ١٤،٣١ إردب/فدان و ١٩٥،٠٧، ١٧٦،٢٥ كجم/فدان و ١٩٠،٠ ، ١٩٩، كجم/م٣ وذلك عند زيادة تركيز الجلايسين بيتايين إلى ١٥ ملليمول/فدان في الموسم الاول والثاني على الترتيب.