

EFFECT OF BIOSTIMULANTS REMEDIATION SUBSTANCES (BRS) ON SEED GERMINATION AND SEEDLING GROWTH OF SOME SUGAR BEET CULTIVARS UNDER STRESS CONDITIONS

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ABSTRACT

Two newly cultivars from sugar beet (*Beta vulgaris*, L.; Chenopodiaceae Pleno(C1) and Plever(C2)) were used to study the effects of certain antioxidant application on elevating the harmful effects of drought stress condition on seed germination as well as seedling growth and its constituents Mannitol levels and/or Ascorbic; AsA, Salicylic; SA, Humic; HA and seaweed; SWE were examined as biostimulants remediation substances (BRS).

Analysis of variance showed a significant effects of Mannitol level and/or the BRS used ($P < 0.001$) on the rate of germination as well as fresh and dry weights of the seedling. The reduction in germination % and the increase in time required for seeds to germinate due to stress was observed in both sugar beet cv(s) studied. However, cv(2) was more susceptible than cv(1) in this respect.

Germination was progressively inhibited by an increase in mannitol level in both cv(s). The strongest inhibition occurred at the third mannitol level (0.3 M) in cv(2). However, cv(1) did not exert any significant effect on ultimate germination % under the 2nd mannitol level. Increasing mannitol level was associated with a marked reduction in AsA, glutathione; GSH, catalase; CAT, guaiacol peroxidase; GPOD and superoxide dismutase; SOD as well as total carbohydrates and total N concentrations, whereas, increased that of H₂O₂ and proline as well as T.S.S. and osmotic pressure of the leaves and roots in both seedling cv(s).

BRS decreased concentrations of H₂O₂ and proline whereas, increased that of AsA and GSH as well as CAT, GPOD and SOD in the development seedling of both cv (s). Treatment with SWE showed an additive effects to that of stress treatments on increasing non-enzymatic and enzymatic antioxidants concentrations in both cv (s). Cv (1) showed, in general higher concentrations of AsA and GSH as well as CAT, GPOD and SOD than that of cv (2).

The interactions treatments showed that any of the BRS used elevating the harmful effect drought stress caused by increasing mannitol level up to 3rd one (3.0 M). Again the SWE followed with HA treatments were the best in this respect.

Keywords: Germination, BRS, stress, mannitol, SWE, AsA, Ha, SA, GSH, CAT, GPOD, SOD, T.S.S, sugar beet.

INTRODUCTION

Water deficient is one of the main limiting factors of sugar beet production in arid and semi arid regions. It have serious impacts as germination and normal development of roots and shoot extension during germination. Moreover, it delaying seedling emergence prolonging critical growth period, increasing changes of seedling damage by pathogenetical and environmental factors. Seed germination and early seedling growth are the

stages most sensitive in sugar beet and it is related with genetical and environmental factors as well as seed pretreatments effects. It is most important for determining seed quality.

On the other hand, storage reserves within the seed are slowly depleted causing a decrease in seedling survival and growth vigour. Studies on abiotic stress tolerance in sugar beet have been undertaken for the identification of physiological and environmental factors to decide the ultimate crop yield (Tugnoli and Bettini, 2001).

It was suggested that, nutrient solution containing osmotic agent such as mannitol, and polyethylene glycol, PEG could be used in screening for drought tolerance in growing seedlings (Ghoulam and Fares, 2001). Osmotic adjustment in tolerant plant helps maintain leaf metabolism and root growth at relatively low leaf water potential by controlling turgor pressure in the cells.

Improve stress capacities of some existing varieties using biochemical's was reported by many investigators (Clapp, *et al.*, 2002; Panda and Khan, 2003; Zhao and Qin, 2004 ; Tang and Newton, 2005 ; Nabati, *et al.*, 2005 ; Faust, 2006 and Ozdoba, 2006).

The effects of BRS on controlling free radicals levels are considered to be the way of plant to tolerate stress (Chattopadhyay *et al.*, 2002) . Polyamines (PAs) greatly potentiate the effects of stress by enhancing reactive oxygen species; ROS generation during photosynthesis (Borsani *et al.*, 2001) used as markers of physiological stress (Tang and Newton , 2005) and play a role in antioxidative system and protect membrane from peroxidation in *in vitro* cultures and induced adaptation to stress (Mishra *et al.*, 2003)

The effects of two newly sugar beet cv (s) seed pretreatments with some BRS on the germination and seedling growth under stress agent (mannitol levels) was studied with an aim to choose an evaluation procedure for the identification of sugar beet tolerant to water stress at germination and early growth stages.

MATERIALS AND METHODS

Germination experiments were carried out at the laboratory of the sugar crops research Institute, Agric. Res. Center (ARC) , Egypt. During the seasons of 2007/2008 and 2008/2009 in an incubated condition at 25-27 °C for 16/18 hours day/night condition using white fluorescent tubes .

Seeds of multigerm sugar beet (*Beta vulgaris*,L; Chenopodiaceae cvs; Pleno and Plever) were obtained from Sugar Crop Institute, Agric. Res. Center (ARC), Ministry of Agric., Egypt.

Seeds (fruits) were graded , standardized, washed, sterilized and dried at room temperature prior to the experiments of germination tests. Seeds were soaked for six h in the specific antioxidants used. Germination was took place in boxes (37x55x13 cm) at the rate of 100 seeds/box which containing perlite. Stress treatments were performed at 0.0 (distilled water ; control) , 0.2 and 0.3 Ml^{-1} concentrations of mannitol approximately corresponding to -1, -5 and -7 bar osmotic pressure. The effects of certain antioxidants were

examined in the presence or absence of mannitol : Ascorbic , Salslic, Humic and SWE were obtained from Sigma Comp. and used at the level of 250 ppm, 250ppm, 1m/L and 1m/L respectively. Boxes were watered with 1/2 diluted Hoagland solution (Hoagland, and Arnon, 1950) for each different treatment.

The amount of these solutions was adjusted daily to keep a 2- cm level at the bottom . A factorial design with six replications was used for each treatment.

Germination % was recorded after 21 days . The relative germination was determined daily by the following calculation: No. of germinated seeds in the stress medium/ No. of germinated seeds in control medium x 100 (Smith and Dobrenz, 1987).

After germination , seedling were allowed to grow for 35 days from sowing . At the end of experiments, seedling characteristics including cotyledon fresh weight and its dry weight at 70 °C , root fresh and dry weight as well as root length were recorded.

Similarly, fresh and dry weights of roots and shoots (cotyledon+leaves) were calculated (Sadeghian and Yavari, 2004). Total carbohydrate content was estimated by the official methods (A.O.A.C. ,2000). Total nitrogen concentration (Nour, 1971), total soluble solide and osmotic pressure (O.P.) of the leaf sap (Slatyer and Mcllory, 1961) were estimated. Moreover, H₂O₂ (Velikova *et al.*, 2000), ascorbic acid (Cakmak and Marschner , 1992), glutathione (El-Hoseiny Hanan,2008),and proline (Bates *et al.*, 1973), also the activity of antioxidant enzymes; catalase (EC 1.11.1.6) according to Velikova *et al.*, 2000, guaiacol peroxidase (EC.1.11.1.7) (Urbaremek *et al.*, 1991) and superoxide dismutase EC 1.15.1.1. (Van Rossum *et al.*, 1997) were determined.

RESULTS AND DISCUSSION

The data representing the germination % and seedling characters of the two sugar beet cv(s) as affected by certain biostimulants remediation substances under drought stress condition using mannitol levels are recorded in Tables 3 and 4.

Decreased germination may be measured as delaying of emergence, reduction of the ultimate germination % and /or both. Analysis of variance showed a significant effects of mannitol level and/or the BRS used ($P<0.001$) on the rate of germination . The reduction in germination % and the increase in time required for seeds to germinate due to water stress was observed in both sugar beet cv(s) studied . However, cv(2) was more susibtable than cv(1) in this respect. In addition, germination was progressively inhibited by an increase in mannitol level in both cv(s). The strongest inhibition occurred at the second mannitol level (0.3 M) in cv(2). However, cv(1) did not exert any significant effect on ultimate germination % under the 2nd mannitol level. The 3rd level of mannitol significantly inhibited germination records of both cv(s).

The harmful effects of drought stress on germination may be due, mainly, to either the inhibition of colloidal inhibition of water occurred by seeds and/or unbalanced of osmotic water uptake occurred by germinated

seeds (Helaly, 1972) . Moreover, the effects of drought on altering the hormonal balances and decreasing endogenous cytokinins biosynthesis and auxin production (Schmidt,2005), decreasing water content , some nutrients uptake and root pull strength (Demir *et al.*, 2004) , decreasing antioxidants , α - tocopherol and carotenoids which necessary for PSII (Hatung, 2004) , increasing of lipid peroxidation and inactivation of enzymes (Smirnoff, 1995) , disturbances in cell membrane components (Salisbury and Ross, 1992) were detected. Increasing free-radical groups activity which are major elements for chlorophyll degradations (Fletcher *et al.*, 1988) and decreased the accumulation of reducing sugars within the plant tissues which decreased wilting resistance were also reported. Moreover, it was found that water stress did not induce an increase of ascorbic acid which not only quenches reactive oxygen but also, regenerates α - tocopherol (Schmidt,2005).

On the other hand, germinating the pretreated seeds may overcome all or part of the drought stress influence through the uptake of some of the surrounding solutes. Such seeds make the osmotic adjustment at the expense of time and risk of one or more from the physiological behavior reported above. Thus, the period elapsed for seed to germinate under stress condition was longer than that for seed germinated under normal condition (Table 3) . It is well known that, of the germinating seeds did not absorb enough water and solutes, they could not adjust to imposed stress influence.

Bio stimulant Remediation Substances; BRS used showed a contributing influences on germination % and delaying their radical emergence (Tables 3 and 4). Seeds of the two cv(s) were responded to these substances differently depending on the level of mannitol, cv and the BRS used. Pretreated seeds with any of the BRS used ,germination was took place over that of the non- pretreated seeds. The almost effective treatment on increasing germination% was found with SWE in both cv(s) with the superiority of cv(1); more resistance than cv(2). It is interesting to note that, the pretreatment with either of BRS used alleviated the harmful effects of the high mannitol level on germination of cv (1) as well as cv (2). In addition, pretreated seed with SWE and Humic acids; HA germinated faster than did those pretreated with Ascorbic, Salciic and distilled water in a descending order. Final germination% was also greater with SWE and Humic acid pretreatants . These results might be as an indicative of the influence of these biostimulants on stability , ion selectivity and orienting permeability membrane (Salisbury and Ross, 1992). The beneficial effects of BRS used on germination in the present investigation may be due, actually, to one or more of their effects on hormonal balance changes to favour cytokinins and auxins production so that, antioxidants production can continue when stress occurs (Schmidt, 2005) . The role of BRS used on promoting vitamin biosynthesis such as thiamine and biotein as well as their effects on activating nitrate reductance enzyme were also detected (Hatung, 2004). The hormone containing products treatments significantly improved to water status regulate cell membrane components under drought stress condition (Salisbury and Ross, 1992).

Table (1): Mean squares from analysis of variance of sugar beet lines tested for the germination rate in filter paper and perlite medium to gather with seedling growth characters of *in vitro* seedling at three levels of mannitol and five BRS levels.

Source of variance	d.f.	Germination rate		Fresh weight		Dry weight		Root length
		In paper	In perlite	Cot.+ leaves	Roots	Cot.+ leaves	Roots	
Season 2007/2008								
Replication	2	68.267	86.033	0.000	0.000	0.000	0.000	16.544
Genotype	1	96.100	144.40	0.120	0.040	0.000	0.000	134.444
Mannitol	2	2511.7	2827.90	90.958	13.648	0.005	0.001	2327.64
BRS	4	1295.65	1226.10	2.236	0.771	0.000	0.000	272.928
Variety x Mannitol	2	24.700	1.900	0.019	0.046	0.000	0.000	25.844
Variety x BRS	4	3.350	7.900	0.042	0.009	0.000	0.000	0.528
Mannitol x BRS	8	15.325	16.275	0.047	0.205	0.000	0.000	11.875
Vriety x Mannitol x BRS	8	1.325	1.525	0.010	0.004	0.000	0.000	978.25
Season 2008/2009								
Replication	2	106.93	46.13	0.000	0.000	0.000	0.000	16.578
Genotype	1	280.90	46.13	0.236	0.001	0.000	0.000	0.011
Mannitol	2	4059.30	3573.70	47.509	10.993	0.005	0.001	2607.51
BRS	4	1080.10	1269.10	2.099	0.757	0.000	0.000	266.428
Variety x Mannitol	2	250.90	13.30	0.019	0.046	0.000	0.000	4.311
Variety x BRS	4	13.40	10.90	0.049	0.003	0.000	0.000	9.594
Mannitol x BRS	8	20.05	10.90	0.060	0.119	0.000	0.000	20.428
Vriety x Mannitol x BRS	8	11.90	10.90	0.007	0.024	0.000	0.000	7.144

Table (2): Coefficient correlation estimated between germination rate and seedling characteristics of sugar beet .

	CDW.	CFW	Ger.In perlite	Ger.In paper	RDW	RFW	RL
CDW.	1						
CFW.	0.929	1					
Ger.In perlite	0.554	0.839	1				
Ger.In paper	0.839	0.929	0.982	1			
RDW	0.982	0.839	0.643	0.554	1		
RFW	0.982	0.982	0.714	0.839	0.929	1	
RL	0.839	0.982	0.929	0.982	0.714	0.929	1

Cotyledon+leaves dry weight; CDW, Cotyledon+leaves fresh weight; CFW, Ger. Root dry weight; RDW, root fresh weight; RFW, Root length; RL.

It has been reported that either of the BRS used increased the major elements uptake especially phosphorus and boron in addition to their effects on regulation of cell membrane components under drought stress (Hatung, 2004 and Yan, 1993). Phosphorus have been found to be mediator of most metabolic reaction within the cell and of hormonal response in plants and accelerated germination (El-Hadidi *et al.*, 1981).

Data in the same tables show that there are no significant differences between presowing drought hardening seeds of the two sugar beet cv(s) treated with Ascorbic acid and those treated with either of Salsilic and Humic acids. However, the seeds of the three treatments alleviated the retarding effects of mannitol on germination as compared with pretreated seeds with water (wet control). The most effective treatments was found with SWE. The effect of SWE was expected since SWE contain not only most of the major

and minor nutrients, amino acid and vitamins (B1, B2, C, E) but also cytokinins, auxins, GA₃ and ABA like substances (Hatung, 2004). Francki (1960) reported that increased trace elements; TE especially boron, B supply could explain only some of the beneficial effects of SWE. The effect of boron and TE on germination was more pronounced at drought stress than non-droughted media (Tables 3-6). The improvement of germination capacity due to boron and TE presented in SWE, especially under high level of mannitol, may be partially explained on the ground that boron and other TE may stimulate the activity of the enzyme synthesis responsible for transformation and /or translocation of carbohydrates. This may cause an increase in the osmotic pressure of the cell sap in the germinated seeds, and in the turn, could offset the osmotic unbalance between germinated seeds and the ambient solution (El-Hadidi *et al.*, 1981). They added that boron and other TE had positive effects on sugar translocation in plant tissues. The enhancement of hydrolytic enzyme activities in the germinated seeds treated with SWE and B was previously mentioned (Salisbury and Ross, 1992). Such changes in enzyme activities due to TE were accompanied with an intensification in respiration rate (Helaly, 1972). Seaweed extract and HA may enhance hydrophobic and hydrophilic antioxidant activity and thus promote germination and water status (Schmidt, 2005). The later author added that, antioxidant status could be manipulated with exogenous application of plant growth biostimulants SWE. The increase of this antioxidants may be triggered by excess production of reactive oxygen species in the photosynthetic apparatus under stress, increased α -tocopherol levels which may serve as an acclimation strategy of plants to tolerate water deficits.

Data in Table 3 show also that, drought stress caused by increasing mannitol level decreased significantly fresh and dry weight of the shoots (cotyledons + leaves), roots as well as whole seedling and shoot/root ratio whereas increased root length of the two cv(s) studied. The effects were more pronounced due to an increase in the osmotic pressure of mannitol level compared with that obtained at the low one. Similarly, cv(2) was more affected by stress than cv(1). The only exception was found with cv(1) at the low mannitol level which should an increase in all seedling characteristics studied under the investigation. Moreover, it was found that shoot growth was reduced to a much greater extent than that of the roots especially with cv(1); more resistance. This in turn, might account for the decrease in the shoot/root ratio. These results might be explained on the basis that, leaves seemed to be more sensitive to drought than roots. Furthermore, it might be assumed that, such phenomenon may be a kind of plant adaptation to stress which is responsible for water and essential nutritive elements uptake.

The harmful effect of stress on germination and seedling growth represented with the dry matter accumulation seemed to be due to the suppression of plant metabolism under such condition (Demir *et al.*, 2004). Salisbury and Ross, 1992) reported that, drought stress adversely affect the physico-chemical properties of the protoplasm and cell membranes.

Table (3): Germination rate of sugar beet seeds cv(s) as affected by water stress in the presence or absence BRS under stress (mannitol) levels

Stress (Mannitol)	BRS	Germination rate														
		Season 2007/2008														
		C1							C2							
		7		14		21			Mean	7		14		21		
Paper	Perlite	Paper	Perlite	Paper	Perlite	Paper	Perlite	Paper		Perlite	Paper	Perlite	Paper	Perlite	Mean	
Control	Control	30	35	60	62	63	65	52	28	32	58	60	62	65	51	
	Ascorbic acid	39	42	77	84	82	84	68	35	39	72	80	79	82	64	
	Salicylic acid	40	41	78	84	81	83	68	38	39	74	82	77	80	65	
	Humic acid	40	42	79	85	82	85	69	39	40	74	82	80	83	66	
	SWE	46	48	80	88	86	90	73	44	44	78	84	82	87	70	
Mean	39	42	75	81	79	81	66	37	39	71	78	76	79	63		
0.2 M	Control	25	28	48	50	52	54	43	25	27	44	50	52	52	42	
	Ascorbic acid	33	37	60	61	79	71	57	30	35	54	58	70	69	53	
	Salicylic acid	34	36	58	62	70	72	55	31	36	52	60	68	70	53	
	Humic acid	35	37	62	63	73	75	57	30	36	58	62	72	75	55	
	SWE	38	40	67	72	76	79	62	32	37	70	74	75	76	61	
Mean	33	36	59	62	68	70	56	30	34	62	61	68	68	54		
0.3 M	Control	20	22	42	44	48	50	38	20	23	42	44	45	50	37	
	Ascorbic acid	28	30	59	60	60	63	50	26	28	51	56	56	60	46	
	Salicylic acid	29	31	57	58	63	64	50	26	28	50	55	60	62	47	
	Humic acid	32	34	55	57	65	66	51	30	31	51	58	62	62	49	
	SWE	34	36	67	69	72	75	59	32	34	54	56	64	64	51	
Mean	29	30	54	56	61	63	49	27	28	52	50	57	60	46		
Mean	Control	25	28	50	52	54	59	44	24	27	48	51	53	56	43	
	Ascorbic acid	33	36	65	68	71	73	58	30	34	64	63	68	70	55	
	Salicylic acid	34	36	64	68	71	73	58	32	34	64	64	68	71	55	
	Humic acid	36	38	65	68	73	75	59	33	36	65	65	72	72	57	
	SWE	39	41	68	73	77	80	63	36	38	68	71	74	76	60	
LSD at 5% for Stress	0.8	1.0	1.3	1.4	1.6	1.6	1.0	1.2	1.2	1.4	1.4	1.4	1.5	1.5		
BRS	1.0	1.4	1.6	1.6	2.0	2.0	1.2	1.3	1.3	1.5	1.5	1.5	1.6	1.6		
StressxBRS	1.6	1.7	1.7	2.0	2.0	2.0	1.6	1.6	1.8	1.8	1.8	1.8	1.9	1.9		
Season 2008/2009																
Control	Control	33	34	59	60	65	66	53	30	31	60	62	64	67	52	
	Ascorbic acid	40	43	74	81	80	80	66	36	37	74	81	79	81	65	
	Salicylic acid	42	44	76	83	82	83	68	40	39	74	82	79	83	66	
	Humic acid	41	44	77	85	84	83	69	42	42	78	84	82	83	68	
	SWE	46	50	82	87	85	91	73	46	48	82	86	84	88	72	
Mean	40	43	74	78	79	81	66	39	39	74	79	78	80	66		
0.2 M	Control	27	30	46	48	50	52	42	26	26	48	52	55	55	44	
	Ascorbic acid	34	37	58	58	57	70	52	33	32	58	58	72	67	53	
	Salicylic acid	36	38	59	60	60	69	54	33	34	62	59	67	70	54	
	Humic acid	36	40	61	63	65	72	56	35	34	60	61	78	71	56	
	SWE	38	40	67	70	70	78	60	35	38	70	72	79	74	61	
Mean	34	37	58	58	79	68	56	32	33	60	60	70	67	54		
0.3 M	Control	20	22	35	33	44	43	33	22	23	40	42	44	47	36	
	Ascorbic acid	27	30	47	46	54	57	43	26	25	52	51	54	58	44	
	Salicylic acid	28	30	50	49	56	60	45	28	27	54	53	58	63	47	
	Humic acid	30	32	50	50	56	62	47	29	30	50	53	60	63	47	
	SWE	31	34	52	59	60	67	50	30	32	55	60	66	68	52	
Mean	27	30	47	47	54	58	44	27	27	50	52	60	60	46		
Mean	Control	27	28	47	47	53	54	43	26	27	49	52	54	78	48	
	Ascorbic acid	34	37	60	61	64	69	54	32	31	61	63	68	69	54	
	Salicylic acid	35	37	62	64	66	71	56	34	33	63	65	68	72	56	
	Humic acid	36	39	63	62	68	72	57	35	35	63	66	73	72	57	
	SWE	38	41	67	71	72	78	61	37	35	69	72	73	77	60	
LSD at 5% for Stress	1.1	1.2	2.5	1.2	1.4	1.4	0.9	1.1	1.8	1.4	1.4	1.4	1.6	1.6		
BRS	1.2	2.7	2.7	1.4	1.4	NS	1.1	2.4	2.4	1.6	1.6	1.6	1.6	1.6		
StressxBRS	1.5	4.0	4.0	NS	NS	NS	1.4	4.4	4.4	2.1	2.1	2.1	2.1	2.1		

Table (4): Fresh weight (F.Wt), dry weight (D.Wt) and root length (RL) of cotyledons + leaves and roots of sugar beet cv(s) as affected by manitol level and BRS levels.

Stress (Mannitol)	BRS	Seedling characters									
		Season 2007/2008									
		C1				C2					
		F.Wt g		D.Wt g		RL cm	F.Wt g		D.Wt g		RL cm
dons + Leave	Roots	dons + Leave	Roots	dons + Leave	Roots		dons + Leave	Roots			
Control	Control	4.266	1.980	0.043	0.020	55	4.350	2.005	0.044	0.020	56
	Ascorbic acid	4.601	2.308	0.046	0.023	58	4.702	2.335	0.048	0.022	58
	Salicylic acid	4.514	2.316	0.045	0.023	63	4.713	2.410	0.048	0.024	64
	Humic acid	4.882	2.708	0.049	0.027	67	4.840	2.650	0.050	0.025	65
	SWE	5.302	3.082	0.053	0.031	72	5.297	3.104	0.053	0.030	74
	Mean	4.713	2.475	0.047	0.025	63.0	4.780	2.601	0.049	0.024	63.4
0.2 M	Control	3.000	1.442	0.030	0.014	50	3.080	1.508	0.031	0.015	53
	Ascorbic acid	3.413	1.508	0.034	0.015	52	3.410	1.714	0.034	0.017	56
	Salicylic acid	3.384	1.565	0.034	0.016	53	3.545	1.690	0.036	0.017	56
	Humic acid	3.602	1.675	0.035	0.017	55	3.600	1.712	0.036	0.017	58
	SWE	4.211	1.800	0.043	0.018	58	4.180	2.002	0.042	0.020	60
	Mean	3.522	1.698	0.035	0.016	53.6	3.563	1.725	0.036	0.017	56.5
0.3 M	Control	1.810	1.118	0.018	0.011	41	2.005	1.007	0.020	0.010	44
	Ascorbic acid	2.008	1.140	0.020	0.012	44	2.312	1.156	0.022	0.012	46
	Salicylic acid	2.214	1.155	0.022	0.012	43	2.368	1.150	0.024	0.012	48
	Humic acid	2.405	1.170	0.024	0.012	44	2.414	1.178	0.024	0.012	50
	SWE	2.708	1.240	0.027	0.013	46	2.600	1.198	0.026	0.012	50
	Mean	2.229	1.165	0.022	0.012	43.6	2.340	1.138	0.023	0.012	47.6
Mean	Control	3.025	1.513	0.030	0.015	48.7	3.145	1.507	0.032	0.014	51.0
	Ascorbic acid	3.341	1.652	0.033	0.017	51.3	3.475	1.735	0.035	0.017	53.3
	Salicylic acid	3.371	1.679	0.034	0.017	53.0	3.542	1.750	0.036	0.018	55.9
	Humic acid	3.360	1.851	0.036	0.019	55.3	3.618	1.847	0.037	0.019	57.7
	SWE	4.074	2.034	0.041	0.021	58.7	4.026	2.101	0.040	0.020	61.3
LSD at 5% for Stress		0.001	0.002	0.0003	0.0001	0.14	0.001	0.003	0.0003	0.000	0.11
BRS		0.001	0.002	0.0003	0.0002	0.19	0.002	0.004	0.0003	0.000	0.14
StressxBRS		0.001	0.003	0.0005	0.0003	0.35	0.002	0.006	0.0006	0.0003	0.24
Season 2008/2009											
Control	Control	4.260	1.974	0.043	0.021	57	4.344	2.000	0.044	0.020	56
	Ascorbic acid	4.680	2.319	0.047	0.023	63	4.780	2.345	0.048	0.024	58
	Salicylic acid	4.574	2.322	0.046	0.023	65	4.745	2.400	0.048	0.024	64
	Humic acid	4.845	2.544	0.049	0.027	65	4.860	2.453	0.049	0.025	65
	SWE	5.333	3.100	0.054	0.031	70	5.245	2.824	0.053	0.027	74
	Mean	4.738	2.450	0.048	0.025	64.0	4.795	2.404	0.048	0.023	63.4
0.2 M	Control	3.041	1.398	0.031	0.014	53	3.112	1.418	0.031	0.013	53
	Ascorbic acid	3.225	1.700	0.033	0.017	58	3.440	1.722	0.035	0.015	56
	Salicylic acid	3.372	1.700	0.034	0.017	55	3.509	1.745	0.035	0.016	56
	Humic acid	3.514	1.662	0.035	0.016	56	3.585	1.787	0.036	0.015	58
	SWE	4.198	1.745	0.042	0.018	62	4.171	2.018	0.042	0.016	60
	Mean	3.470	1.641	0.035	0.016	56.8	3.563	1.738	0.036	0.015	56.6
0.3 M	Control	1.714	1.110	0.017	0.011	40	2.011	1.002	0.020	0.010	42
	Ascorbic acid	2.000	1.224	0.020	0.012	45	2.319	1.244	0.023	0.011	45
	Salicylic acid	2.208	1.250	0.022	0.013	45	2.350	1.256	0.024	0.011	45
	Humic acid	2.312	1.274	0.023	0.013	44	2.400	1.255	0.024	0.012	47
	SWE	2.622	1.354	0.027	0.014	50	2.565	1.300	0.026	0.013	49
	Mean	2.171	1.242	0.022	0.013	44.8	2.328	1.211	0.023	0.012	45.7
Mean	Control	3.005	1.491	0.030	0.015	50.0	3.155	1.473	0.032	0.014	50.3
	Ascorbic acid	3.302	1.748	0.033	0.017	55.3	3.513	1.770	0.035	0.016	53.0
	Salicylic acid	3.385	1.757	0.034	0.018	55.0	3.535	1.800	0.036	0.016	55.0
	Humic acid	3.557	1.827	0.036	0.019	55.0	3.615	1.832	0.036	0.017	56.6
	SWE	4.051	2.066	0.041	0.021	60.6	3.994	2.047	0.040	0.020	61.1
LSD at 5% for Stress		0.001	0.001	0.000	0.0001	0.18	0.001	0.001	0.000	0.000	0.24
BRS		0.001	0.001	0.000	0.0002	0.22	0.001	0.002	0.000	0.000	0.33
StressxBRS		0.002	0.002	0.000	0.0003	0.46	0.001	0.002	0.000	0.0002	0.47

Inhibition of cytokinin biosynthesis and hormonal unbalances, water content, some plant nutrient uptake, antioxidant enzymes (SOD, GR, ASP), biosynthesis of α -tocopherol, ascorbic acid and carotenoids as well as net photosynthetic rate accompanied with high respiration rate were also reported under stress condition (Schmidt, 2005).

Soaking sugar beet seeds with each of the BSR used can alleviate the harmful effects of drought stress on all seedling characters studied (Tables 3 and 4). It can be noticed that, soaking seeds in SWE, HA, Ascorbic and Salicylic acids in a descending order counteracted the depressing effects of drought stress on seedling growth to a different extent. It may be suggested that sugar beet seedling especially with cv(2) subjected to these treatments acquired a reasonable drought tolerance capacity. The more effective treatment restoring most of the plant growth capacity under high mannitol level was the biostimulants SWE and HA. This might be attributed to its effects on activate root cells and stimulate biosynthesis of endogenous cytokinins from the roots (Schmidt, 2005). The results in the present investigation indicate that soaking seeds in antioxidants used should be considered on soaking the factors which may improve growth in mannitol – affected seedling. It has been show that provision for adequate uptake of water and nutrients or adjusting the hormonal balance within the plant tissues were essential for improving development of sugar beet under stress condition (Demir *et al.*, 2004). Results in the same tables show also that the effect of decreasing water potential in 0.2 and 0.3 M mannitol levels resulted in seedling with less fresh weight during the germination processes and early growth compared with the control.

Analysis of variance for fresh weight of cotyledons and leaves as well as roots indicated that stress condition and BRS had significant differences for these parameters. Stress decreased water content and assimilate accumulation in the seedling of the two cv(s) as a consequence of osmotic pressure induced by mannitol.

The interaction effects between BRS and mannitol stress were significant for the germination rate pointing to the fact that differences in BRS type affecting the germination response of sugar beet are expressed at early stages under specific stress condition. The differences in germination % of seeds subjected to stress levels were more detectable in SWE and HA. Germination of seeds with ascorbic and salicylic acids proved to be most sensitive to water restriction at the higher level of osmotic potential but with HA revealed a relatively stable tolerance in both stress levels.

Evaluation of seedling characters presented as germination % of the control, data show a decrease for all growth parameters as stress levels intensified. The most tolerant seedling showed a better biological efficiency (increase in size and weight) under increased water deficiency, while a different distribution of biomass in leaves and roots were noted. Here again, SWE and HA responded best for relative germination and relative growth of root length. The other various BRS used responded differently for the relative growth measured as fresh weight. A positive correlation was found between germination rates and seedling characters in most cases except for the % of abnormal seedling recorded under high stress level without BRS addition.

addition. Moreover, Seed germination was closely related to the root length and in severe stress condition the highest value of root length was allocated to the drought – resistance . Absolute increases in root elongation rate are strongly related to a high water status in the plant organs. The exponents of Bewley and Black (1985) performed on *Brassica oleracea var stailica* seeds in water stress medium demonstrated that, the sensitivity of radical expansion and radical growth to water stress is markedly different . There are a distinguish between seed germination which is completed when the radical expand and penetrates the medium , consisting of only cell elongation and the cell division and radical (root length) which starts later on. On sugar beet, Sadeghian and Yavari (2004) indicated that the stress levels exceeding -8 bar did not permit seed imbibitions and distinction among the seed lines. Similarly, germination % of *Brassica oleracea* seeds was decline as water stress increase from 0 to -8 bar and finally stopped at -14 bar , whereas radical growth was started at -8 bar water potential, declined at -16 bar and slopped at -22 bar (Bewley and Black,1985) .

Distinction of significant differences in sugar beet seedling growth and physiological performance in water restriction stress lead to the conclusion that these parameters , especially germination rate and seedling root length, could be used as a criteria in screening the most tolerant progeny lines against abiotic stresses. *In vitro* controlled conditions seems to be more amenable for evaluating of genetic materials at early growth phase. Progeny lines having stable germination and seedling growth properties against a range of induced osmotic pressure may then be included in breeding programmes for yield potential and stability under water- restricted condition in experimental plot.

Effect of BRS with or without stress on H₂O₂, AsA , Glutathione, Proline and the activity of antioxidant enzyme:

Data in Table 5, show that, Increasing mannitol level was associated with a marked reduction in AsA, glutathione; GSH, catalase;CAT, guaiacol peroxidase;GPOD and superoxide dismutase;SOD whereas, increased that of concentrations of H₂O₂ and proline of the shoots in both seedling cv(s). The concentrations of H₂O₂ and proline were decreased due to BRS application. The decrease was more pronounced in response to SWE treatment in both cv(s) of sugar beet. The interaction treatments show that, the antioxidants counteracted the depressing effects of mannitol on all parameters studied in both cv (s) and cv(2) showed, in general, high concentration of H₂O₂ and proline values compared with cv (1).

A specific roles of BRS on elevating stress were reported by Metwally, Reda (2009) who reported that, the alleviation effect of ascorbic acid may be due to its enhancing effect on cell division and synthesis of hydroxyl-proline-rich protein. Wingate *et al.*, (1988) found that, antioxidants regulated the gene expression and being the precursor of phytochelatins. Increasing H₂O₂ under drought stress as well as other reductive oxygen species; antioxidants leading to oxidative stress is a principal component of their damaging effect on plant tissues (Schutzendubel and Polle, 2002).The protective role of antioxidants against H₂O₂ accumulation was previously reported (Cheng, 2003 and Munne- Bosch, 2005).

Table (5): Effects of BRS with or without mannitol level on H₂O₂ (µg/ g⁻¹ F.wt), AsA (µg/ g⁻¹ F.wt), glutathione;GSH (µg/ g⁻¹ F.wt), proline (mg/g⁻¹ F.wt.)and the activity of antioxidant enzyme (µg/ g⁻¹ F.wt)of shoots of sugar beet cv(s) during the growing seasons of 2008/2009.

Stress (Mannitol)	BRS	C1						
		H ₂ O ₂	AsA	GSH	Proline	SOD	CAT	GPOD
Control	Control	1.03	236	45.2	1.42	139	63	8
	Ascorbic acid	0.32	301	64.4	0.41	95	90	8
	Salicylic acid	0.30	312	52.6	0.55	99	87	7
	Humic acid	0.25	330	70.1	0.32	105	99	10
	SWE	0.18	342	75.4	0.11	132	105	11
	Mean	0.42	304	61.5	0.56	114	89	8
0.2 M	Control	1.22	168	32.6	1.62	108	44	5
	Ascorbic acid	0.72	205	45.2	1.22	122	56	7
	Salicylic acid	0.84	198	40.6	1.30	128	48	6
	Humic acid	0.56	232	44.3	0.96	130	60	8
	SWE	0.48	238	50.1	0.85	136	64	9
	Mean	0.76	208	42.6	1.19	125	54	6
0.3 M	Control	1.35	132	18.3	2.04	75	28	3
	Ascorbic acid	0.96	166	30.2	1.66	96	39	5
	Salicylic acid	0.99	152	27.6	1.72	86	36	4
	Humic acid	0.74	184	40.5	1.35	104	42	6
	SWE	0.57	192	44.3	1.22	110	48	7
	Mean	0.92	165	32.2	1.61	94	39	5
Mean	Control	1.20	179	32.0	1.69	107	45	6
	Ascorbic acid	0.67	224	46.6	1.11	104	62	7
	Salicylic acid	0.71	221	40.3	1.19	104	57	6
	Humic acid	0.52	249	51.6	0.88	113	67	8
	SWE	0.41	257	56.6	0.73	126	72	9
LSD at 5% for Stress		0.05	2.2	1.4	0.2	1.1	0.6	0.03
BRS		0.06	3.6	2.3	0.3	1.7	0.8	0.05
StressxBRS		0.11	5.1	3.4	0.5	2.3	1.1	0.06
Control	C2							
	Control	1.08	222	40.4	1.56	132	58	5
	Ascorbic acid	0.40	266	58.4	0.44	90	86	7
	Salicylic acid	0.36	300	46.3	0.62	93	81	6
	Humic acid	0.32	315	66.6	0.54	100	93	8
	SWE	0.27	320	70.2	0.26	118	99	10
Mean	0.49	285	56.4	0.68	107	83	7	
0.2 M	Control	1.18	154	27.3	1.70	100	40	4
	Ascorbic acid	0.78	200	33.7	1.33	112	51	6
	Salicylic acid	0.92	198	38.5	1.38	120	44	5
	Humic acid	0.64	227	40.6	1.05	124	55	7
	SWE	0.52	230	44.4	0.96	128	58	9
	Mean	0.81	202	36.9	1.28	117	50	6
0.3 M	Control	1.46	119	16.2	2.16	68	24	2
	Ascorbic acid	1.06	154	25.7	1.74	90	33	4
	Salicylic acid	1.12	140	23.3	1.80	81	30	4
	Humic acid	0.99	172	34.2	1.44	99	37	5
	SWE	0.75	178	38.9	1.30	104	43	6
	Mean	1.07	153	27.7	1.69	88	33	4
Mean	Control	1.24	165	28.1	1.81	100	41	4
	Ascorbic acid	0.62	207	39.3	1.17	97	57	6
	Salicylic acid	0.66	213	36.0	1.27	98	52	5
	Humic acid	0.57	238	47.1	1.01	107	62	7
	SWE	0.51	243	51.2	0.84	117	67	8
LSD at 5% for Stress		0.03	2.3	1.2	0.02	1.6	0.7	0.02
BRS		0.04	3.7	1.8	0.03	2.1	0.9	0.05
StressxBRS		0.10	4.8	2.7	0.05	3.0	1.2	0.07

According to Abdel-Salam, Heba (2006) , AsA reduced glutathione; GSH and α- tocopherol; T have each been shown to act as antioxidants in the detoxification of reactive oxygen species; ROS in ascorbic cells. They

have central and interrelated roles acting both chemically and as substrates in enzyme-catalyzed detoxification reactions. AsA is an important compound of the plant antioxidant defence system and serves as a reductant for the peroxidative removal of H_2O_2 . reduced glutathione; GSH directly reduces most of reactive oxygen species; ROS and maintain the ascorbate pool in plant cell. The reduction effects of AOs on proline concentration may be due to their stress alleviation effects (Abdel-Salam, Heba 2006) in addition to their enhancing effects on cell division (Sanchez-Fernandez, *et al.*, 1997) These effects lead to an acceleration of proline consumption in the synthesis of hydroxyl-proline-rich proteins which are necessary for progression through the cell cycle (Arrigoni *et al.*, 1992).

Data tabulated in Table 5 show also that, BRS especially with SWE increased the concentrations of AsA and GSH in sugar beet seedlings of both cv (s) grown under water stress used. Moreover, SWE showed higher effects than that of HA or AsA or SIA on increasing enzymatic and non-enzymatic antioxidants concentrations in both cv (s) since treatment SWE showed highest values in this respect.

The interaction effects between the cv (s), stress and the BRS treatments on antioxidants enzymes activities were significant. These results indicated that, BRS induced oxidative stress. Smeets *et al.*, (2005) reported that, plant cell respond to elevated levels of oxidative stress by activating their antioxidative defence system and the first group of enzymes involved in this defence are the ROS- quenching enzymes such as; CAT, PODS and SOD. Cheng (2003) reported that, CAT, PODS and SOD are important enzymes for plant adaptation to environmental stresses as the harmonious of the three enzymes make the balance between ROS production and elimination, thus keeping the level of ROS in plant tissues low, to prevent the injury of cells. In peroxisomes, H_2O_2 can be destroyed by CAT. CAT produces molecular oxygen and water from two molecules of H_2O_2 . Since these two molecules must impinge simultaneously at the active site, CAT has a very high maximum velocity. PODS is an important role in scavenging H_2O_2 and organic peroxidase (Smeets *et al.*, 2005). Moreover, Abdel-Salam, Heba (2006) reported that, SOD causes the catalytic dismutation of potentially toxic superoxide anion radical ($O_2^{\cdot-}$) to H_2O_2 whereas, CAT decomposes H_2O_2 to water and oxygen molecule, both enzymes provide an efficient mechanism for the removal of free radicals from the cells.

Total carbohydrates, total nitrogen, total soluble solids and osmotic pressure:

The data representing total carbohydrates, total nitrogen, total soluble solids and osmotic pressure of the two sugar beet cv (s) as affected by certain Biostimulant Remediation Substances; BRS under mannitol stress condition are presented in table 6.

Results in Table (6) revealed that increasing mannitol level was associated with a marked reduction in total carbohydrates and total N concentrations, whereas, increased that of T.S.S. and osmotic pressure of the shoots in both cv(s) tended to increase. The limitation of carbohydrates production under stress condition may be due to one or another of the two ways: first; an adjustment in the internal osmotic pressure of the cell sap as a

trial to go along with the external drought media (Salisbury and Ross, 1992) or increasing the intermediate substances of organic products (Hatung, 2004). These organic substances may serve as acceptor for the inorganic one, particularly those of N, P, K, Mg, Ca, Zn, Fe and Cu, thus causing an increase in the inorganic substances at the expense of total N (Crouch and Van Staden, 1993). Second: a production of higher energy, by means of respiration in order to overcome the relatively low availability condition of water and nutritional elements in stressed media (Demir *et al.*, 2004). Since carbohydrates are the principal substances used in respiration a depression in carbohydrates content of plants growth under stress condition could be expected. Other possibility could be the suffering of plants growth on stress media from K deficiency (Salisbury and Ross, 1992), which in turn may result in carbohydrates diminish. El-Hadidi, *et al.*, (1981) suggested that N compounds under stress may not be fully utilized and consequently the accumulation of these substances is more rapid than their utilization for the formation of new cells and tissues. The increase in total N of sugar beet under stress condition noticed in the present investigation in connection with the highly depressed in leaves growth more than in roots could be explained on the basis that N uptake was not affected as leaves growth by mannitol increase and consequently N accumulation was affected.

Regarding the effects of pretreatment with BRS, it was found that soaking the two cv(s) of sugar beet in SWE, HA, AsA and SA increased in a descending order the % of total carbohydrates in the two plant cv(s). Moreover, BRS treatments showed an additive

effects to that of stress on protein-N and total nitrogen concentrations as well as total soluble solids and osmotic pressure. However, soluble N showed a decrease as a result of pretreatment seeds with each of the BRS used. The increase in protein and total N may be attributed to a corresponding increase in amino acids and nucleic acids intimately connected with and incorporated into nitrogenous compounds (O'Donnell, 1973).

Data in the present investigation indicated that, the effects of these substances were more pronounced under high mannitol level. In this context, it was found that hardening treatments with biostimulants could partially overcome the disturbances occurred in the physiological statuses due to stress condition. The pretreated seeds restored growth, increased chlorophylls biosynthesis (Gardaye and Churin, 1996), inhibited the activity of free radical groups which are major elements for chlorophyll degradation (Fletcher *et al.*, 1988), stimulated the biosynthesis of α -tocopherol, ascorbic acid and carotenoids in the chloroplast which protect photosynthetic apparatus of PSII (Hatung, 2004), promoted the accumulation of reducing sugars which increased wilting resistance through enhancing osmotic pressure inside the plant tissues (Salisbury and Ross, 1992) and stimulated the chloroplast development (Demir *et al.*, 2004). Moreover, it was reported that, BRS regulate the hormonal balance, water status and nutrient availability within the plant tissues (Demir *et al.*, 2004). Likewise, Schmidt, (2005) found that SWE increased endogenous cytokinins biosynthesis under drought stress which regulate nutrient absorption cell membrane components as well as, redistribution and determination of the overall ions uptake selectivities

Table (6): Total carbohydrates, total nitrogen, total soluble solids and osmotic pressure(O.P) of the two sugar beet cv (s) as affected by certain Biostimulant Remediation Substances; BRS under mannitol stress condition during the two growing seasons of 2007/2008 and 2008/2009.

Stress (Mannitol)	BRS	Season 2007/2008							
		Total carbohydrates %		Total N %		Total soluble solids %		O.P (atm)	
		C1	C2	C1	C2	C1	C2	C1	C2
Control	Control	83.00	85.68	7.45	7.14	2.43	2.11	7.95	5.11
	Ascorbic acid	100.09	106.16	8.02	7.62	3.67	2.85	6.14	4.22
	Salicylic acid	105.12	105.72	7.85	7.55	3.50	2.83	6.60	4.35
	Humic acid	105.00	108.57	8.15	7.84	3.71	3.12	6.32	4.58
	SWE	110.08	116.47	8.45	8.06	4.08	3.92	5.00	4.20
	Mean	100.66	104.52	7.98	7.64	3.48	2.97	6.39	4.49
0.2 M	Control	70.58	72.83	6.30	6.25	3.49	3.02	7.44	5.21
	Ascorbic acid	92.58	92.34	6.65	6.54	3.91	3.50	7.66	5.00
	Salicylic acid	94.66	92.66	6.52	6.42	3.94	3.49	7.69	5.06
	Humic acid	92.80	93.05	6.71	6.61	4.11	3.95	7.20	5.00
	SWE	100.70	100.12	6.82	6.70	4.18	3.99	6.86	4.56
	Mean	90.26	90.20	6.60	6.50	3.93	3.59	7.37	4.97
0.3 M	Control	49.22	48.92	4.96	4.82	5.49	4.03	11.22	9.41
	Ascorbic acid	52.30	50.44	5.12	5.07	5.66	4.84	10.56	9.20
	Salicylic acid	50.42	50.66	5.07	5.02	5.92	4.82	10.30	9.25
	Humic acid	53.60	52.00	5.24	5.22	6.33	4.96	10.58	8.55
	SWE	58.65	56.87	5.35	5.27	6.48	5.22	10.12	8.55
	Mean	52.94	51.78	5.15	5.08	5.98	4.77	10.58	8.99
Mean	Control	67.80	69.14	6.24	6.07	3.80	3.05	8.87	6.58
	Ascorbic acid	81.66	82.98	6.61	6.41	4.41	3.73	8.12	6.14
	Salicylic acid	83.40	83.01	6.48	6.33	4.45	3.71	8.21	6.22
	Humic acid	83.47	84.84	6.70	6.56	4.72	4.01	8.03	6.04
	SWE	89.81	91.15	6.87	6.68	4.91	4.38	7.33	5.77
LSD at 5% for Stress BRS		0.06		0.09		0.033		0.35	
StressxBRS		0.11		0.11		0.039		0.38	
		0.17		0.18		-		0.42	
Season 2008/2009									
Control	Control	84.02	82.33	5.74	5.62	2.33	2.18	7.13	5.08
	Ascorbic acid	106.00	100.55	6.12	6.05	2.83	2.66	6.09	4.80
	Salicylic acid	105.56	100.96	6.06	6.00	3.11	2.68	6.11	4.80
	Humic acid	105.40	104.58	6.25	6.14	3.17	3.22	6.00	4.82
	SWE	116.11	112.88	6.34	6.22	3.69	3.86	5.78	4.50
	Mean	103.42	100.26	6.11	6.01	3.03	2.92	6.22	4.80
0.2M	Control	67.31	60.96	4.93	4.86	4.08	3.30	9.80	7.58
	Ascorbic acid	76.86	70.77	5.32	5.23	4.44	3.96	9.65	7.30
	Salicylic acid	76.56	72.23	5.16	5.12	4.83	4.00	9.50	7.23
	Humic acid	76.58	72.00	5.44	5.30	5.02	4.08	9.52	7.25
	SWE	80.22	77.18	5.52	5.41	5.40	4.14	9.36	7.00
	Mean	75.51	70.63	5.27	5.16	4.75	3.91	9.57	7.27
0.3M	Control	48.58	44.55	4.15	4.07	5.38	4.40	11.00	9.22
	Ascorbic acid	56.52	50.99	4.40	4.42	5.50	4.66	10.80	9.08
	Salicylic acid	53.23	54.58	4.35	4.33	5.84	4.70	10.80	9.00
	Humic acid	51.65	52.30	4.52	4.63	6.34	4.72	10.86	9.05
	SWE	60.56	58.66	4.60	4.71	6.60	5.07	10.78	8.86
	Mean	54.11	52.22	4.40	4.43	5.93	4.71	10.65	9.04
Mean	Control	66.64	62.61	4.94	4.86	3.93	3.29	9.31	7.29
	Ascorbic acid	79.79	74.10	5.28	5.23	4.26	3.76	8.85	7.06
	Salicylic acid	78.48	75.92	5.19	5.15	4.59	3.79	8.80	7.01
	Humic acid	77.88	76.29	5.40	5.36	4.84	4.01	8.79	7.04
	SWE	85.63	82.91	5.49	5.45	5.23	4.36	8.64	6.79
LSD at 5% for Stress BRS		0.05		0.11		0.030		0.38	
StressxBRS		0.08		0.13		0.034		0.40	
		0.12		0.22		0.041		0.47	

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تأثير بعض المواد المعالجة الحيوية على إنبات ونمو البادرة لبعض أصناف بنجر السكر تحت ظروف الآجهاد.

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تم دراسة تأثير بعض المواد المعالجة الحيوية تحت ظروف الإجهاد المائي الناتج عن إستخدام المانيتول على إنبات ونمو البادرة لصفينجديدين من بنجر السكر الذى يتبع العائلة المرمامية وهما Plover , Pleno وإستخدم لهذا الغرض كمواد معالجة حيوية حمض الأسكوربيك، حمض السليسليك، حمض الهيوميك ومستخلص الأعشاب البحرية.

وقد بينت نتائج تحليل التباين وجود تأثيرات معنوية لأى من مستوى المانيتول والمواد المعالجة الحيوية المستخدمة والتفاعل بينهما على كلا من الإنبات ونمو البادرة. وقد لوحظ إتخفاض نسبة الإنبات ومعدله بزيادة الإجهاد المائي لكلا الصنفين حتى تركيز ٠,٣ مول، وقد تأثر الصنف الثانى بدرجة أكبر عن الصنف الأول عند المستوى الثانى من المانيتول ٠,٢ مول.

سبب وجود الإجهاد إتخفاض كلا من حمض الأسكوربيك والجلوتازيون والنشاط الإنزيمى للكاتاليز والبيرو اكسيدز والجويكول والأكسيدز بالإضافة الى إتخفاض تركيز الكربوهيدرات الكلية والنتروجين الكلى، بينما أدى الإجهاد الى زيادة تركيز كلا من فوق أكسيد الهيدروجين والبرولين والمواد الصلبة الذاتية الكلية والضغط الإسموزى فى بادرات كلا الصنفين، وعلى العكس من ذلك، فقد أدى إستخدام المواد المعالجة الحيوية الى نقص تركيز كلا من فوق أكسيد الهيدروجين والبرولين مع زيادة نسبة حمض الأسكوربيك والجلوتاتيون والنشاط الإنزيمى للكاتاليز والبيرو اكسيدز والجويكول والأكسيدز فى البادرات النامية لكلا الصنفين. وكانت المعاملة بمستخلص الأعشاب البحرية أفضل المعاملات للمواد المعالجة الحيوية، كما تميز الصنف الأول بإرتفاع نسبة حمض الأسكوربيك والجلوتاتيون والنشاط الإنزيمى للكاتاليز والبيرو اكسيدز والجويكول والأكسيدز عن الصنف الثانى.

ولقد أوضحت نتائج معاملات التداخل أن للمواد المعالجة الحيوية تأثيراً تعويضياً للنقص المتسبب عن وجود الإجهاد المائي، وكانت أفضل المعاملات عند إستخدام مستخلص الأعشاب البحرية، وحمض الهيوميك حتى تركيز ٠,٣ مول مانيتول.

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