

Asian Resonance

Effect of Benzyladenine on Biochemical and Physiological Changes Deficit Stress in Maize Seedlings



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Abstract

Investigation was carried out at Department of Biochemistry, B. A. College of Agriculture, Anand Agricultural University, Anand to study the biochemical and physiological characterizations of two maize genotypes (resistance and susceptible) procured treatment was given to seed followed by artificial drought (PEG) stress at 15 DAS. For total chlorophyll, true protein and moisture content in biochemical analysis were decreased while total carbohydrates, total soluble sugars, reducing sugars, proline and free amino acids content were increased in both the genotypes (resistance and susceptible) due to drought stress. Application of BA (25 ppm) could be increased in all biochemical and physiological parameters for improved biochemical content in both genotypes. The moisture content was found minimum in resistant over susceptible genotype. However, resistant genotype showed better performance in higher root-shoot length and weight than susceptible genotype under drought stress conditions, through treatment BA was higher root-shoot length and weight in resistant and susceptible genotypes.

Keywords: Biochemical, Physiological, Maize, *Zea Mays*, Poly Ethylene Glycol, Benzyladenin.

Introduction

Zea mays L. (Maize) is the third most important cereal crop after wheat and rice all over the world. Taxonomically, *Zea mays* L. belongs to tribe Maydeae, family Poaceae and genus *Zea*. It is believed that *Zea* might be derived from old Greek name *Zola* named for food grass. The other *Zea* species, referred to as teosintes are largely wild grasses native to Mexico and Central America (Doebly, 1990). The center of origin for *Zea mays* ($2n=20$) has been established as the Mesoamerican region, now Mexico and Central America (Watson & Dallwitz, 1992). Benzyladenine (BA) is the synthetic form of Cytokinins (CKs) known to regulate several aspects of plant height growth and development, including the response of plants to abiotic stress (Haberer and Keiber, 2002).

Benzyladenine with seed treatment increases the growth rate and pod yield under moisture stress compared to water soaked seeds (Reddy et al., 2003). Chlorophyll content and relative water content increase by application of benzyl adenine.

Aim of the Study

1. To study biochemical changes in maize seedlings during induced drought stress in relation to their resistant and susceptible behavior.
2. To study the effect of drought stress and benzyladenine at physiological level in maize seedlings.

Materials and Methods

Two maize Cultivars CM-500 (Resistance) and GYC-9327 (Susceptible) cultivars were raised in pot with three replications. Fifteen DAS seedlings were treated with 10, 15 and 20 % PEG-6000 induced water deficit stress. Prior to this induction of stress seeds were treated with 25 ppm benzyladenine for one hour. There were eight treatments (T1 – Control, T2 - PEG- 6000 (10%) treatment, T3 - PEG- 6000 (15%) treatment, T4 – PEG- 6000 (20%) treatment, T5 - Seeds treatment with Benzyladenine (25 ppm) for 1hr., T6 - Seeds treatment with Benzyladenine (25ppm) + PEG (10%), i.e. T2+T5, T7 - Seeds treatment with Benzyladenine (25 ppm) + PEG (15%), i.e. T3 + T5, T8 -Seeds treatment with Benzyladenine (25 ppm) + PEG (20%), i.e. T4 + T5. (Shown in fig. 2). This eight treatments were divided into two groups, first group T1 to T5 to study the effect of PEG stimulated water stress and second group treatments T5 to T8 observed the

effect of benzyladenine under PEG induced water deficit stress. The seedlings were analysed for biochemical and physiological parameters.

Preliminary experiment was carried out to find out suitable concentration of BA bases on the germination percent of 25 ppm concentration found best which was selected for the further study. (Fig. 1) Before sowing maize seeds of both genotypes were treated with 25 ppm Benzyladenine (BA) for 3 hour and dried for 30 min. Then the seeds were germinated in underlaboratory condition, artificial drought stress was induced by using polyethylene glycol (PEG-6000). The 15 days after sowing (DAS) seedlings were taken into conical flask containing PEG-6000 (10 %, 15% and 20%) and artificial drought induced for 24 hrs. (Fig. 2) After 24 hrs seedlings were collected for their biochemical and physiological analysis

Sr.no.	Treatments	Sr.no.	Treatments
T1	Control	T5	25 ppm BA
T2	10 % PEG	T6	25 ppm BA + 10 % PEG
T3	15 % PEG	T7	25 ppm BA + 15 % PEG
T4	20 % PEG	T8	25 ppm BA + 20 % PEG

Review of Literature

Biochemical Parameters

Moisture

Abo-El-Kheir and mekki (2007) studied the effect of water deficits at different reproductive stages on growth. The water content in maize leaves was significantly decreased by decreasing soil moisture content.

Total Carbohydrates

Keyvan (2010) studied the effect of drought stress on bread wheat cultivars, viz., Chamran (C₁), Marvdasht (C₂) and Shahriar (C₃). He observed that leaf soluble carbohydrates was variable between 1.1 to 10.4 percent on 14 to 20 days after flowering according to cultivar, drought stress increased the concentration of these parameters in the leaf.

Ahemad *et al.*, (1989) studied the effect of phytohormone on water stressed maize plant. It was observed that drought adversely affect the synthesis and translocation of carbohydrate which affect an accumulation of amino acid proline. Application of phytohormone increased carbohydrate content in maize plant.

Total Soluble Sugars

Soluble sugars content plays a very important role in carbohydrate metabolism and has a close relationship with photosynthesis and production (Wilcox, 2001).

Various authors point out to the role of soluble sugars in the protection against stresses. Mobilisation of storage reserves in the endosperm of cereal seeds is tightly regulated and has a primary pivotal role in the interactions among sugar, ABA and gibberellin pathways responsible for the response to drought.

Yin *et al.*, (2012) studied effect of drought stress on physio-biochemical parameters of maize varieties and reported that water stress caused significantly increased soluble sugar content in maize.

Mohammadkhani and Heidari (2008) studied drought stress was applied with various concentration of *i.e.* 10, 20, 30 and 40 % polyethylene glycol (PEG) and without PEG (control). Soluble sugar accumulations of two maize (*zea mays* L.) genotypes were determined after drought stress. They recorded that, soluble sugar concentration increased from 1.18 to 1.90 % in root and shoot of both varieties under drought stress condition.

Reducing Sugars

Deshmukh *et al.*, (2001) reported that water stress induced by polyethylene glycol (PEG-6000) at seedling stage in three different varieties of *rabi* sorghum caused significantly increased in level of reducing sugars due to water stress. While reducing sugar was higher in RSLG-262 and lower in swati variety.

Sarwat and El-Sherif (2007) studied effect of benzyladenine on some hulled and hulled less genotype grown under two salinity levels of irrigation water. Barley plant irrigated with underground water of two salinity levels (4413 and 8761 ppm). The genotype Giza 123 and line-1 (salt resistant cultivar) had the highest reducing sugar as compare to with line-2 (Salt sensitive cultivar) genotype.

True protein

Ali *et al.*, (2011) observed the effect of growth promoter in drought stressed maize line. They found that the application of benzyladenine (50 mg/L) increased the total soluble protein.

Zeid and El-Semary (2008), studied the response of two differentially drought tolerant variety of maize, Giza 2 and single cross hybrid 155 were planted under different levels of drought stress induced by polyethylene glycol (PEG 4000). The maize genotype Giza 2 was more tolerant than SCH 155. The growth regulator treated plants had higher protein as compare to untreated plants. Accumulation of protein may account for high osmo-regulation in growth regulators treated plant under drought stress condition.

Deshmukh *et al.*, (2001) reported that water stress induced by polyethylene glycol (PEG-6000) at seedling stage in three different varieties of *rabi* sorghum caused significantly decreased in protein.

Total Chlorophylls

Chlorophyll concentration has been known as an index for evaluation of source therefore decrease of this can be consideration as a non stomata limiting factor in the drought stress conditions. There are reports about decrease of chlorophyll in the drought stress conditions (Majumdar *et al.*, 1991).

Ali *et al.*, (2011) observed the effect of growth promoter in drought stressed maize line. They found that the application of benzyladenine (50 mg L⁻¹) increased the chlorophyll.

Yin *et al.*, (2012) studied effect of drought stress on physio-biochemical parameters of several varieties of maize. Water stress caused significant decreased in chlorophyll content.

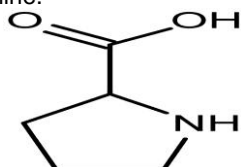
Efeoglu *et al.*, (2009) studied the response of the maize (*zea mays* L.). Cultivar Doge, Vero and

Luce to drought condition. Drought stress was imposed on the plant at 12 DAS by withholding irrigation for 12 days and then rewatering for 6 days. They observed that Chlorophyll a and b as well as total chlorophyll (a+b) of all maize cultivars significantly reduced under drought stress.

Tas and Tas (2007) studied the chlorophyll content of two *T. aestivum* and two *T. durum* cultivars under control and stress condition. They observed significant reductions in chlorophyll content in all cultivars under water stress.

Proline

Proline is an α -amino acid, one of the twenty DNA-encoded amino acids. It is not an essential amino acid, which means that the human body can synthesize it. It is unique among the 20 protein-forming amino acids in that the amine nitrogen is bound to not one but two alkyl groups, thus making it a secondary amine.



The protective role of proline in osmotic stress is not well understood, it is thought to function as a compatible osmolyte that stabilizes membranes and sub-cellular components. In addition, proline is proposed to scavenge free radicals (Fabro *et al.*, 2004). A positive correlation between proline accumulation and osmotic stress tolerance has been reported in several experimental systems (Gilmour *et al.*, 2000; Nanjo *et al.*, 1999).

Drought stress is a decrease of soil water potential, so plants reduce their osmotic potential for water absorption by congestion of soluble carbohydrates and proline and in other words osmotic regulation is performed (Martin *et al.*, 1993). Osmotic regulation helped in cell development and plant growth in water stress (Pessarkli, 1999). Functions of proline accumulation have also been proposed, including stabilization of macromolecules, a sink of carbon and nitrogen for use after relief from water deficit, radical detoxification and regulation of cellular redox status by proline metabolism.

Zlatev and Stoyanov (2005) suggested that proline accumulation of plants could be only useful as a possible drought injury sensor instead of its role in stress tolerance mechanism.

Moussa and Abdel-Aziz (2008) studied comparative response of drought tolerant and sensitive maize genotypes to water stress. Maize genotypes Giza 2 (drought tolerant) and Trihybrid 321 (drought sensitive) were sown in small pots. Water stress condition was created by irrigating the pots with polyethylene glycol (PEG) solution of 0.0, -5, -10 and -20 bars and observed on 21 days old seedlings. While increased accumulation of proline in Giza 2 (5.82 mM/gm) was more than trihybrid 321 (3.96 mM/gm) under drought stress.

Rai *et al.*, (2004) studied effect of waterlogging of some biochemical parameters during

early growth stages of maize. The 10 lines of maize (*zea mays* L.) after 20 days of growth were studied subject to waterlogging and changes. Biochemical characters were studied in 2 cm root apices after 48 and 96 hours of waterlogging. Proline content was increased but after 96 hour of waterlogging it decreased.

Mohammadkhani and Heidari (2008) studied drought-induced accumulation of proline in two maize varieties. The solute accumulation of two maize (*Zea mays* L.) cultivars (704 and 301) were determined after PEG-6000 induced drought stress. The free proline level increased (from 1.56 to 3.13 times) in response to drought stress and the increase in 704 variety was higher than 301 variety. It seems that proline may play a role in minimizing the damage caused by dehydration. Increase of proline content in shoots was higher than roots.

Akhkha *et al.*, (2011) conducted a pot experiment to evaluate the effects of water stress on wheat crop. Four cultivars of wheat (*Triticum durum*) viz., Al-gaimi, Sindy-1, Sindy-2 and Hab-ahmar were studied. The plants were subjected to three water regimes well-watered plants at 80% of the field capacity of soil (FC) and two levels of water stress, moderate stress at 50% FC and severe stress at 30% FC. The results of this study showed that water deficit led to generally high free proline levels in Hab- Ahmar cultivar, Sindy-1 and Sindy-2 with the latter being not significant. In contrast, water stress caused a decrease in the proline content in Al-gaimi cultivar. Al-gaimi was the most drought tolerant wheat cultivar followed by Sindy-1, while the most drought-sensitive cultivar was Hab-Ahmar followed by Sindy-2.

Free Amino Acids

The amino acids are basic building blocks of proteins. Apart from being bound as proteins, amino acids also exist in the free form in many tissues and are known as free amino acids. Very often in plants during water stress conditions, the free amino acid composition exhibits a change and hence, the measurements of total free amino acids give the physiological and health status of the plants.

Zeid and El-Semary (2008) studied response of two differentially drought tolerant variety of maize viz., Giza 2 and single cross hybrid 155. Both the varieties were planted under different levels of drought stress induced by polyethylene glycol (PEG 4000). The maize variety Giza 2 was more tolerant than SCH 155. Accumulation of amino acids may account for high osmo-regulation in growth regulators treated plant under drought stress condition.

Rai *et al.*, (2004) studied effect of waterlogging on some biochemical parameter during early growth stages of maize. Plants were subjected to waterlogging and changes in biochemical characters were studied in 2 cm root apices after 48 and 96 hours of waterlogging. Free amino acids content increased under waterlogging and amount further increased with waterlogging duration.

Sarwat El-Sherif (2007) studied effect of benzyladenine on some hulled and hulled less

genotypes of barley grown under two salinity levels of irrigation water. Barley plant irrigated with underground level of two salinity levels (4413 and 8761 ppm). Biochemical parameters observed that Giza 123 and line-1 genotype as compared with line-2 (Salt sensitive cultivar) genotypes result showed that total amino acids in shoots of all genotype increased significantly with increasing salinity at both growth stages.

Physiological studies

The impacts of drought condition on grain development and yield of crops depend on their severity and the stage of plant growth during which they occur. Seedling emergence is a stage of growth which is sensitive to water deficit. Seed germination, vigor and coleoptiles length are prerequisites for the success of stand establishment of crop plants. Under semiarid regions, low moisture is limiting factor during germination. The rate and degree of seedling establishment are extremely important factors in determining the both yield and time of maturity (Rauf, *et al.*, 2007).

The ability to withstand dehydration under water stress is an important characteristic contributing to drought tolerance in plants and can be assessed by measuring root length, shoot length, root weight and shoot weight under normal and stressed situations.

Lagerwerff *et al.*, (1961) indicated that PEG can be used to modify the osmotic potential of nutrient solution culture and thus induce plant water deficit in a relatively controlled manner. Polyethylene glycol molecules with a 6000 (PEG-6000) are inert, non-ionic and virtually impermeable chains that have frequently been used to induce water stress without causing physiological damage and maintain uniform water potential throughout experimental periods (Lu and Neumann, 1998). Molecule of PEG-6000 are small enough to influence the osmotic potential but large enough to not be absorbed by plant and not expected to penetrate intact plant tissues rapidly (Carpita, *et al.*, 1979) and because PEG does not enter the apoplast therefore water is withdrawn from the cell.

Root-Shoot Length

Alaei *et al.*, (2010) studied the root-shoot length of *durum* wheat under osmotic stress (PEG-6000). The results showed that the highest root-shoot length of wheat seedling noticed under control than the lowest stress conditions.

Datta *et al.*, (2011) determined the shoot and root length of five wheat varieties under drought stress (PEG-6000, -1.0 bar) and found that the highest shoot length was 10.36 cm observed in NW 2036 and the lowest was 6.15 cm in HUW 234 in case of controlled. On the other hand in stressed condition the highest shoot length was 18.06 cm

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observed in HD 2954 and the lowest was 3.647cm observed in HUW 234.

Root-Shoot Weight

Jabeen *et al.*, (2008) conducted greenhouse experiment on maize (*Zea mays* L.) cultivars for drought tolerance. Seven maize cultivars viz., Sahiwal-2002, Sadaf, EV-5098, Pak-Afgoyee, Agaiti-2002, Agaiti-85 and EV-1098 were grown under well watered or water deficit condition (60% of field capacity). They observed imposition of water deficit condition decreased the shoot and root weight in all maize cultivars.

Result and Discussion

Biochemical Parameters

The decrease in moisture, true protein and total chlorophylls content was observed in resistance (CM-500) and susceptible (GYC-9327) genotypes under drought stress conditions compared with the control condition. (Table 1.1). Both resistance and susceptible genotypes showed higher moisture, true protein and total chlorophylls content in treatment T5 (73.85, 2.64, 4.25 % and 76.07, 2.13, 3.96% respectively). The lowest moisture, true protein and total chlorophylls content was observed in treatment T4 (66.53, 1.88, 2.44 % and 71.03, 1.05, 2.18% respectively),

Fig.1

Suitable concentration of BA Base on the Germination Percentage of Maize Seeds

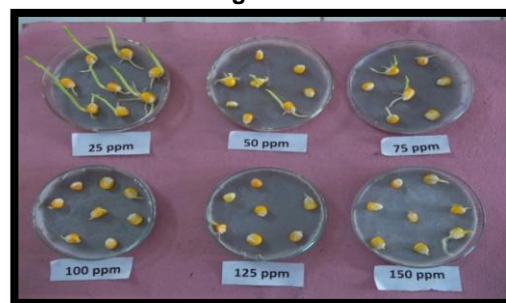


Fig. 2

Suitable Concentration of BA and PEG Bases on the 15 DAS of Maize Seedlings



Table 1.1
Biochemical Attributes of Maize Seedling on 15 DAS

Treatment	Total carbohydrate (%)		Total soluble Sugar (%)		Reducing Sugar (%)		True protein (%)		Proline ($\mu\text{g}/100\text{ mg}$)	
	CM- 500	GYC- 9327	CM- 500	GYC- 9327	CM- 500	GYC- 9327	CM- 500	GYC- 9327	CM- 500	GYC- 9327
T ₁ (Control)	2.63	1.85	1.51	0.96	1.47	1.21	2.39	1.75	1.68	1.29
T ₂ (PEG 10%)	2.76	2.04	1.64	1.19	1.58	1.33	2.10	1.32	1.83	1.41
T ₃ (PEG 15%)	2.88	2.27	1.79	1.30	1.69	1.46	2.00	1.17	1.96	1.57
T ₄ (PEG 20%)	3.00	2.45	1.90	1.46	1.80	1.59	1.88	1.05	2.08	1.69
T ₅ (BA)	3.09	2.59	2.05	1.58	1.92	1.71	2.64	2.13	2.19	1.88
T ₆ (BA + 10 % PEG)	3.15	2.65	2.19	1.64	2.09	1.83	2.25	1.39	2.28	2.00
T ₇ (BA + 15 % PEG)	3.22	2.76	2.28	1.79	2.23	2.00	2.22	1.25	2.37	2.11
T ₈ (BA + 20 % PEG)	3.39	2.89	2.40	1.93	2.37	2.16	2.19	1.19	2.49	2.29
Sem	0.08	0.05	0.03	0.04	0.03	0.03	0.018	0.023	0.04	0.02
C. D.	0.23	0.14	0.08	0.13	0.09	0.10	0.06	0.07	0.12	0.06
CV %	4.34	3.27	2.40	5.04	2.62	3.40	1.45	2.78	2.81	1.86

Table 1.1
Biochemical Attributes of Maize Seedling On 15 DAS

Treatments	Moisture (%)		FAA (%)		Total chlorophyll II (mg/g)		Chlorophyll a (mg/g)		Chlorophyll b (mg/g)	
	CM- 500	GYC- 9327	CM- 500	GYC- 9327	CM- 500	GYC- 9327	CM- 500	GYC- 9327	CM- 500	GYC- 9327
T ₁ (Control)	71.10	74.97	1.31	0.99	3.90	3.53	2.07	1.85	1.83	1.68
T ₂ (PEG 10%)	70.08	74.00	1.53	1.19	3.46	3.05	1.88	1.66	1.58	1.39
T ₃ (PEG 15%)	68.70	73.07	1.69	1.44	3.08	2.65	1.65	1.46	1.43	1.19
T ₄ (PEG 20%)	66.53	71.03	1.83	1.51	2.44	2.18	1.23	1.17	1.21	1.01
T ₅ (BA)	73.85	76.07	2.01	1.72	4.25	3.96	2.38	2.04	1.87	1.92
T ₆ (BA + 10 % PEG)	71.00	75.03	2.13	1.89	3.84	3.33	2.00	1.74	1.84	1.59
T ₇ (BA + 15 % PEG)	70.20	74.00	2.29	2.00	3.57	3.02	1.92	1.59	1.65	1.43
T ₈ (BA + 20 % PEG)	67.67	71.73	2.41	2.11	2.98	2.91	1.58	1.51	1.40	1.40
Sem	0.64	0.56	0.04	0.02	0.086	0.071	0.067	0.08	0.06	0.06
C. D.	1.91	1.68	0.11	0.07	0.26	0.21	0.21	0.25	0.17	0.19
CV %	1.58	1.31	3.44	2.59	4.33	3.97	6.31	8.72	6.05	7.56

Protein and total chlorophylls content as compared to control and drought stress. However, combined effect of BA and PEG treatment was increased moisture, true protein and total chlorophylls content of both genotypes in treatments T₆, T₇ and T₈ respectively as compared to drought stress. This finding is an accord with moisture content obtained by Abo-El-Kheir and Mekki (2007). These results were agreement with Dhruve and Vakharia (2008) who reported that chemical treatment BA soaked seed was significantly highest chlorophyll content than control.

Deshmukh et al., (2001) showed protein content decreasing trend with increase in water stress.

The total carbohydrates, reducing sugar, proline total soluble sugar and free amino acid content in susceptible resistant maize genotypes seedling at 15 DAS was found to increased during PEG induced drought stress as compare to control conditions (Table 1.1). Both resistant and susceptible genotypes had found higher in total carbohydrates, reducing sugar, proline, total soluble sugar and Free amino acid content in treatment T₈ (3.39, 2.37, 2.49, 2.40, 2.41 % and 2.89, 2.16, 2.29, 1.93, 2.11 %, respectively). The lowest total carbohydrates, reducing sugar, proline and Free amino acid content were

found in control T₁ (2.63, 1.47, 1.68, 1.51, 1.31 and 1.85, 1.21, 1.29, 0.96, 0.99, respectively).

Under drought stress condition, total carbohydrates, reducing sugar, proline and free amino acid content was increased in both genotypes (Treatments T₂, T₃ and T₄) as compared to control.

However, 25 ppm BA + 20 % PEG treatment (T₈) showed maximum total carbohydrates, reducing sugar, proline and free amino acid content in resistant and susceptible genotypes. However, due to combine effect of BA and PEG induced drought stress carbohydrates content was increased in resistant and susceptible genotypes in treatments T₆, T₇ and T₈ as compare to drought stress conditions. Resistant genotype had highest carbohydrate, reducing sugar, proline and free amino acid content than susceptible genotype. These results were agreement with Ahemad et al., (1989). The reducing sugar like glucose and fructose may be serving as osmolyte under water stress condition Vyas et al., (1985). The increase in free amino acid content could possibly due to protein hydrolysis and inhibition of their oxidation Ranieri et al., (1989).

Physiological Parameter

Root-shoot length and weight reduction was observed under stress condition in both genotypes.

The data was tabulated in (Table 1.2).

Table 1.2
Physiological Attributes of Maize Seedling on 15 DAS

Treatments	Root length (cm)		Shoot length (cm)		Root weight (gm)		Shoot weight (gm)	
	CM 500	GYC 9327	CM 500	GYC 9327	CM 500	GYC 9327	CM 500	GYC 9327
T ₁ (Control)	20.73	17.27	15.03	13.93	1.83	1.17	1.13	1.01
T ₂ (PEG 10%)	20.57	17.10	14.67	13.87	1.17	0.97	1.00	0.89
T ₃ (PEG 15%)	20.30	16.90	14.50	13.70	0.93	0.83	0.94	0.77
T ₄ (PEG 20%)	20.03	16.73	14.00	13.33	0.74	0.50	0.80	0.70
T ₅ (BA)	20.90	17.33	15.83	14.23	2.00	1.59	1.29	1.16
T ₆ (BA + 10 %PEG)	20.73	17.23	15.00	13.97	1.38	1.03	1.11	0.98
T ₇ (BA + 15 %PEG)	20.50	17.03	14.78	13.83	1.10	0.95	1.09	0.87
T ₈ (BA + 20 %PEG)	20.37	16.83	14.67	13.80	0.97	0.83	1.01	0.84
Sem	0.59	0.32	0.32	0.35	0.08	0.06	0.05	0.04
C. D.	NS	NS	NS	NS	0.23	0.18	0.16	0.13
CV %	4.71	3.27	3.69	4.34	10.54	10.73	9.05	8.14

Polyethylene glycol (PEG) treatment decreased available water required for plant growth. Root length of both susceptible and resistant maize genotypes were decreased under drought stress conditions. Both resistance and susceptible genotypes found longer root length in treatment T₅ (20.90 cm and 17.33 cm) and shoot length in treatment T₅ (15.83 cm and 14.23 cm) whereas lower root length was observed in treatment T₅ (20.03 and 16.73 cm, respectively) and shoot length (14.00 cm and 13.33 cm), respectively. Polyethylene glycol (PEG) treatment decreased available water required for plant growth.

Root length of both susceptible and resistant maize genotypes were decreased under drought stress conditions.

Root-shoot weight was decreased with increasing drought stress in both resistance and susceptible genotype. (Table 1.2). Both resistance and susceptible genotypes recorded higher root weight in treatment T₅ (2.00 gm and 1.59 gm) and shoot weight (1.29 gm and 1.16 gm), whereas lower root weight was observed in treatment T₄ (0.74 gm and 0.50 gm, respectively) and shoot weight (0.80 gm and 0.70 gm), respectively.

Under drought stress condition, root-shoot length and weight of both genotypes was decreased in treatment T₂, T₃ and T₄ respectively as compared to control. The BA soaked seed treatment T₅ showed highest root length and weight in both genotypes as compare to control and stress conditions. However, in combine treatment of BA and PEG (T₆, T₇ and T₈), root-shoot length and weight was increased in both genotypes as compare to control and drought stress condition. The resistant genotype exhibited the long root and shoot length than susceptible genotypes.

Results showed that the resistant genotype had higher root length than susceptible genotypes. Amin et al., (2007) reported that application with benzyladenine (BA) treatment caused increased in growth character.

Conclusion

Biochemical Parameters

Proximate analysis of maize seedling was analyzed from 15 DAS stage. Application of PEG for

artificial drought stress decreased the moisture content, total carbohydrates, total soluble sugar, reducing sugar, true protein, total chlorophyll, proline and free amino acid content. The chemical treatment Benzyladenine (T₅) had increased all biochemical content during drought stress. However, combine treatment of drought stress and benzyladenine had also increased all biochemical content as over control and drought stress in resistant and susceptible genotypes. Only moisture content was found high in susceptible genotype, rest of all biochemical parameters were higher in resistant genotype than susceptible genotype. Application of benzyladenine had better performance in all biochemical parameters during drought stress condition. These data could be useful for identification of biochemical changes in resistant and susceptible genotypes during drought stress.

Physiological Parameters

The root-shoot length and weight decreased during drought stress in both resistance and susceptible genotypes. Application of BA showed longer root-shoot length and weight in resistance and susceptible genotypes. However, resistant genotype had longer root-shoot length and weight than susceptible genotypes.

References

1. Abo-El-Kheir, M.S.A and Mekki, B.B. (2007). Response of maize single cross – 10 to water deficit during silking and grain filling stages. *World journal of agricultural science*, 3(3): 269-272.
2. Ahemad, A.M., Radi, A.F., Shaddad, M.A. and El-Tayeb, M.A. (1989). Effect of phytohormones on carbohydrates and nitrogen metabolism of some drought stressed crop plant. *Journal of Islamic Academy of science*, 2(2): 93-99.
3. Akhkha, A., Boutraa T. and Alhejely, A. (2011). The rates of photosynthesis, chlorophyll content, dark respiration, proline and abscisic acid (ABA) in wheat (*Triticum durum*) under water deficit conditions. *International Journal of Agricultural Biology*, 13: 215–221.

4. Alaei, M., Zaefizadeh, M., Khayatnezhad, M., Alaei, Z. and Alaei, Y. (2010). Evaluation of germination properties of different durum wheat genotypes under osmotic stress. *Middle-East Journal of Scientific Research*, 6(6): 642-646.
5. Ali, Z., Ahemad, S. M., Munir, H., Mahmood, A. and Yousaf, S. (2011). Mitigation of drought stress in maize by natural and synthetic growth promoter. *Journal of Agriculture and Social Science*, 7(2):56-62.
6. Amin, A. A., Rashad, El. sh. M., Hassanein, M. S. and Zaki, M. N. (2007). Response of some white maize hybrid to foliar spray with Benzyadenine. *Research Journal of Agriculture and Biological science*, 3(6): 648-656.
7. Carpita, N., Sabularse, D., Mofezinos, D. and Delmer, D. (1979). Determination of the pore size of cell walls of living plant cells. *Science*, 205: 114-1147.
8. Datta, J. K., Mondal, T., Banerjee, A. and Mondal, N. K. (2011). Assessment of drought tolerance of selected wheat cultivars under laboratory condition. *Journal of Agricultural Technology*, 7: 383-393.
9. Deshmukh, R. N., Laware, S. L. and Dhumal, K. N. (2001). Metabolic alteration in sorghum bicolor under water stress. *Journal of Maharashtra Agricultural University*, 26(1):50-53.
10. Dhruve, J.J. and Vakharia, D.N. (2008). Groundnut response to benzyl adenine under water stress at different phenophases. *Indian J. Agric. Biochem.*, 21(1 & 2): 21-26.
11. Doebley, J. (1990). Molecular evidence for gene flow among *Zea* species. *BioScience*, 40(6): 443-448.
12. Efeoglu, B., Ekmekci, Y. and Cichel, N. (2009). Physiological response of 3 maize cultivars to drought stress and recovery. *South African Journal of Botany*, 75: 34-42.
13. Fabro, G., Kovacs, I., Pavet, V., Szabados, L. and Alvarez, M. E. (2004). Proline accumulation and At P5 CS2 gene activation are induced by plant pathogen incompatible interactions in *Arabidopsis*. *Molecular Plant-Microbe Interactions*, 17: 343-350.
14. Gilmour, S. J., Audrey, M. S., Salazar, M. P. Everard, J. D. and Thomashow, M. F. (2000). Over expression of the *Arabidopsis* CBF3 transcriptional activator mimics multiple biochemical changes associated with cold acclimation. *Plant Physiology*, 124: 1854-1865
15. Haberer, G. and Kieber, J. J. (2002). Cytokinins new insights into a classic phytohormone. *Plant Physiology*, 128: 354-362.
16. Jabeen, F., Shahbaz, M. and Ashraf, M. (2008). Discriminative some prospective cultivar of maize for drought tolerant using gas exchange characteristics and proline contents as physiological marker. *Pakistan Journal of Botany*, 40(6): 2329-2343.
17. Keyvan, shami (2010). The effects of drought stress on yield, relative water content, proline, soluble carbohydrates and chlorophyll of bread wheat cultivar. *Journal of Animal & Plant Sciences*, 8(3): 1050-1060.
18. Lagerwerff, J. V., Ogata, G. and Eagle, H. E. (1961). Control of osmotic pressure of Culture solutions with polyethylene glycol. *Science*, 133: 1486-1487.
19. Lu, Z. and Neumann, P. (1998). Water-stress maize, barley and rice seedlings show species diversity in mechanisms of leaf growth inhibition. *Journal of Experimental Botany*, 49: 1945-1952.
20. Majumdar, S., Ghosh, S., Glick, B. R. and Dumbroff, E. B. (1991). Activities of chlorophyllase, phosphoenolpyruvatecarboxylase and ribulose-1, 5- bisphosphate carboxylase in the primary leaves of soybean drying senescence and drought. *Physiologia Plantarum*, 81:473-480.
21. Martin, M., Micell, F., Morgan, J. A., Scalet, M. and Zerbi. G. (1993). Synthesis of osmotically active substances in winter wheat leaves as related to drought resistance of different genotypes. *Journal of Agronomy and Crop Science*, 171: 176-184.
22. Mohammadkhani, N. and Heidari, R. (2008). Drought-induced accumulation of soluble sugars and proline in two maize varieties. *World Applied Sciences Journal*, 3: 448-453.
23. Moussa, H. R. and Abdel-Aziz, S. M. (2008). Comparative response of drought tolerant and drought sensitive maize genotypes to water stress. *Australian Journal of Crop Science*, 1(1): 31-36.
24. Nanjo, T., Kobayashi, M., Yoshiba, Y., Kakubari, Y., Yamaguchi, S. K. and Shinozaki, K. (1999). Antisense suppression of proline degradation improves tolerance to freezing and salinity in *Arabidopsis thaliana*. *FEBS Letter*, 461: 205-210.
25. Pessarkli, M. (1999). Hand book of plant and crop stress. Marcel Dekker Inc. 697 pages.
26. Rai, R. K., Srivastava, J. P. and Shahi, J. P. (2004). Effect of waterlogging of some biochemical parameter during early growth stages of maize. *Indian Journal of Plant Physiology*, 9(1): 65-68.
27. Ranieri, A. R., Lanese, B. R. and Soldatini, G. R. (1989). Changes in free amino acid content and protein pattern of maize seedling under water stress. *Envirmental Experimental Botany*, 29: 351-357.
28. Rauf, M., Munir, M., Ul-Hassan, M., Ahmed, M. and Afzai, M. (2007). Performance of wheat genotypes under osmotic stress at germination and early seedling growth stage. *African Journal of Biotechnology*, 8: 971-975.
29. Reddy, T. V. B., Prakasarao, J. S. and Vijayalaxmi, K. (2003). Physiological and biochemical evaluation of groundnut cultivars differing in drought tolerance. *Indian Journal of Plant Physiology*, 8(4): 359-363.
30. Sarwat, M. I. and El-sherif, M. H. (2007). Increasing salt tolerance in some barley genotypes (*Hordeum vulgure*) by using kinetin

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- and Benzyladenine. *World Journal of Agriculture*, 3(5): 617-629.
31. Tas, S. and Tas, B. (2007). Some physiological responses of drought stress in wheat genotypes with different ploidity in Turkiye. *World Journal of Agricultural Science*, 3: 178-183.
 32. Vyas, S. P., Kathju, S., Garge, B. K and Lahiri, A. N. (1985). Performance and metabolic alterations in *Sesamum indicum* L. under different intensities of water stress. *Annals of Botany*, 56(3): 323-331.
 33. Watson, L. and Dallwitz, M. J. (1992). *Grass Genera of the World: Descriptions, Illustrations, Identification, and Information Retrieval*. Version: 18thAugust.<<http://biodiversity.uno.edu/delta>>We bsource:<http://www.duchefa.com/msds/bog04.pdf>. Pp:-1-6.
 34. Watson, L. and Dallwitz, M.J. (1992). *Grass Genera of the World: Descriptions, Illustrations, Identification, and Information Retrieval*. Version: 18thAugust.<<http://biodiversity.uno.edu/delta>>We bsource:<http://www.duchefa.com/msds/bog04.pdf>. Pp:-1-6.
 35. Wilcox, J. R. (2001). Sixty years of improvement in publicly developed elite soybean lines. *Crop Science*, 41: 1711-1716.
 36. Yin, G., Shen, Y., Tong, N., Gu, J., Hao, L. and Liu, Z. (2012). Drought induced changes of Physio-biochemical parameter in maize. *Journal of Food Agriculture and Environment*, 10(1): 853-858.
 37. Zeid, M. and El-Semary, N. A. (2008). Response of two differentially drought tolerant varieties of maize to drought stress. *Pakistan Journal of Biological Science*, 4(7): 779-784.
 38. Zlatev, Z. and Stoyanov, Z. (2005). Effect of water stress on leaf water relations of young bean plans. *Journal of Central European Agriculture*, 6(1): 5-14.