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TOLERANCE OF PEA PLANTS TO DROUGHT STRESS IN RELATION TO ANTIOXIDANT APPLICATION

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ABSTRACT*: Pea plants cv. Master B was grown in pots Experment to investigate the effect of selenium (Se) and salicylic acid (SA) as antioxidants in relation to drought stress. The experimental treatments included three water regem levels; 100% of field capacity control (W_0); 60% of field capacity (W_1) and 40% of field capacity (W_2) and two antioxidants such as Se (0.6 mM) and SA (0.5 mM). The growth, physiological and chemical composition as well as yield of pea plants were determined. The drought had decreases in plant height, root length, leaf area, root / shoot ratio and produced less dry matter as compared with the control plants (W_0). Relative water content (RWC), water use efficiency (WUE), pressure potential, enzymes activity, total soluble sugars, total free amino acid and K were decreased in droughted plants. Supplementary Se and SA ameliorated the negative effect of drought on the previous parameter. Membrane permeability in leaves increased under water stress and this increase was adversed with Se and SA treatments. Total weight of green pods, average number of green pods and weight of seeds / plant were increased in the plants treated with Se and SA than those under drought stress(W_1 and W_2)*

Key Words: ***Drought, Antioxidant, selenium, salcilylic acid, , peroxidase, catalase, compatible osmolyte, yield.***

INTRODUCTION

Irrigation is the most important factor affecting the production and quality of plant. However, at present time , there is a crltical balance between water requirements and water consumption, thus water saving is becoming a decisive factor for agricultural expansion. Water requirements is the water needed for raising a crop in a given period of time field conditions. It includes consumptive use and other economically unavoidable losses.

Researches have shown that drought has many adverse impacts on plant growth parameters (Xu *et al.,* 2007). Drought stress also significantly inhibited the biomass accumulation of wheat seedlings (Xiaoqin *et al.,* 2009). Exposure plants to drought stress substantially decrease leaf water potential (ψ_w), relative water contents (RWC) and transpiration rate with associated increase in leaf temperature (Halder and Burrage, 2003). Drought stress produces changes in photosynthetic pigments (Loggini *et al.,* 1999), damages photosynthetic apparatus (Fu and Huang, 2001) and decreases the activities of calvin cycle enzymes (Monakhova and Chernyadev, 2004).

Selenium (Se) has been recognized as an essential trace element for plant based on its presence in antioxidative defence system (Djanaguiraman *et al.,* 2005) and hormone balance (Pallud *et al.,* 1997). Selenium exerts as a beneficial on growth and stress tolerance of plants by enhancing their oxidative capacity (Xue and Hartikainen, 2000 and Kong *et al.,* 2005). Selenium increases plant resistance against oxidative stress caused by free oxygen radicals. Recently it has been shown that selenium has ability to regulate the water status of plants under drought conditions (Kuznetsov *et al.,* 2003). Application of Se up to 50 ppm in soybean increased the yield by preventing chlorophyll degradation and maintaining longer leaf area duration (Djanaguiraman, *et al.,* 2005).

Salicylic acid (SA) plays an important role in a biotic tolerance, and considerable interest has been focused on SA because of increased antioxidant enzymes which induce a protective effect on plant under stress (Horvath *et al.,* 2007). Plants treated with SA before stress reduced the damage effect of water stress on the cell membrane in the leaves and increased proline levels (Senaratna *et al.,* 2000). SA also improved the protoplasmic drought tolerance (Abreu *et al.,* 2008). In this connection, Khodary (2004) reported increased chlorophyll and carotenoids contents in maize plants by SA application.

The objective of this study was to investigate the beneficial effect of Se and SA to increase the antioxidative capacity of plants grown under drought and to counteract senescence related oxidative system to improve the growth and yield of stressed plants.

MATERIALS AND METHODS

Two pots experiments were conducted in greenhouse at the Experimental Station Farm, Faculty of Agriculture, Minufiya University during the winter seasons of 2007/2008 and 2008/2009. Pea plants cv. Master-B were planted in October 21st in plastic pots (25 cm diameter). Seven kilogram loamy soil was weighed for each pot. Some physical and chemical properties of soil used were determined according to Jackson (1967). The particles size distribution of the studied soil were sand (8.1%), salt (42.20%) and clay (49.7%). Also this soil characterized by pH (7.76), EC (1.45 dsm^{-1}), contents (meq/100g soil) of soluble cations i.e. Ca^{2+} (4.40), Mg^{2+} (2.32), Na^{+} (6.32) and K^{+} (1.41) and the content of soluble anions as meq/100g soil was HCO_{-3} (2.53), Cl^{-} (6.41) and SO_4^{2-} (5.51). Six seeds / pot were sown onto the upper layer of soil and covered with thin layer of sand soil. Pots were arranged in six replicates randomized block design. After twenty one day seedlings were thinned to four seedlings per pot.

All pots were fertilized with calcium super-phosphate (15.5%P_2O_5) at the rate of 200 Kg/fed.(1.4 g/pot) before sowing. Nitrogen and potassium fertilizer in amount equivalent to 100 Kg/fed, ammonium nitrate (33.5 %N) at the rate of

0.7 g/pot and 100 g/pot potassium sulphate (48 % K_2O) at the rate of 0.7 g/pot at various growth stages represent 30 and 60 days after sowing.

The plants were subjected to one of nine treatments:
(1) 100 % of field capacity (100% of F.C (W_0)),
(2) 100% of F.C + Se (W_0 + Se),
(3) 100% of F.C + SA (W_0 + SA),
(4) 60% of F.C (W_1),
(5) 60% of F.C + Se (W_1 + Se),
(6) 60% of F.C+ SA (W_1 + SA),
(7) 40% of F.C (W_2),
(8) 40% of F.C. + Se (W_2 + Se)
(9) 40% of F.C+SA (W_2 + SA).

Se (0.6mM) was added as sodium selenite (Na_2SeO_3) (Djanaguiraman *et al.,* 2005) and SA (0.5 mM) (Senaratna *et al.,* 2000).Se and SA were sprayed twice at 30 and 45 days after sowing. Tween 20 was added to the spraying solution at 0.5 % as a surfactant.

Plant samples were successively taken randomly from every treatment at 75 days after sowing. Three plants were taken out carefully from each pot, and the following data were recorded.

1- **Growth characters: growth characters such as plant height, root length, roots and shoots dry matter, roots / shoots ratio and leaf area were determined. Water used efficiency (WUE) was calculated as described by Raeini-Sarjaz *et al.* (1998).**
2- **Water relations: leaf water potential (Ψw) using the modified dye method of Marathe (1989). Osmotic potential ($\Psi\pi$): values of total soluble solids of the cell sap were obtained from the pressed sap using an Abbe Reflectometer and osmotic potential values were calculated by using special tables according to the method described by Gosev (1960). Leaf pressure potential (Ψp) was calculated from the relationship: $\Psi w = \Psi\pi + \Psi p$, assuming leaf matrix potential was zero (Nobel, 1991). Relative water content (RWC) was calculated by the equation of Larcher (1995):**

$$RWC = [(Wt - Wd) / (Wf - Wd)] \times 100,$$

Where, Wt: turgid weight, Wf: fresh weight and Wd: dry weight of leaves.

Membrane integrity:To indicate the extent of membrane damage in tissues subjected to moisture measurements on the leakage of solutes, absorption at Ultraviolet Wavelength 273 nm was determined following the method of Leopold *et al.* (1981).

3- **Chemical composition: (A) Photosynthetic pigments was estimated in fresh leaves as described by Witham *et al.* (1971).**

(B):Antioxidant Enzymes Activity: Peroxidase activity in O.D./g fresh weight after 2 min. was measured in pea fresh leaves using the method described by Fehrman and Dimond (1967). Phenoloxidase activity in O.D. / g fresh weight after 45 min. was determined in fresh leaf samples was extracted by the method of Broesh (1954).

Catalase activity was estimated according to Samantary (2002) and expressed as µM H_2O_2 reduced mg^{-1} protein min^{-1}.Glutathione peroxidase activity was measured by a modification method of Flohe and Gunzler (1984) and expressed as µM mg^{-1} protein min^{-1}.

4- Compatible Osmolytes: Proline, total soluble sugars, total free amino acids concentration were estimated according to the methods described by Bates *et al.* (1973), Dubois *et al.* (1956) and Rosen (1957) respectively. K^+ was determined as mentioned by A.O.A.C (1990).

5- Yield and its components: At harvest time (about 110 days after sowing). The following parameters were recorded: Average number of green pods/plant, total weight of green pods/plant and weight of seeds/plant

All Data were statistically analyzed using a CO-STAT-C NOVA program. Statistically different groups were determined by Duncan's Test ($p < 0.05$) and calculated according to Duncan (1955) and Gomez and Gomez (1984).

RESULTS AND DISCUSSION

1. Growth characters:

Exposure pea plants to water stress led to decreases in plant height, root length, root and shoot dry matter. W_2 treatment was more effective in reducing previous parameters (Table 1). Se application significantly increased the shoot growth in term of previous parameters which increased by 32, 95, 39 and 64%, respectively in W_1+Se treatment compared with W_0. Moreover, SA treatments also recorded similar results as Se treatment.

Drought stress especially at W_2 significantly reduced leaf area (LA) by 25.1%, whereas W_1+Se treatment significantly increased it by 23% as compared with W_0 in the first season. Moreover, W_2+Se and/or W_2+SA treatments significantly increased LA by 37 and 27% compared with the droughted plants (W_2) in the first season. The growth stimulating effect by Se may be related to its antioxidanive function as demonstrated by diminished lipid peroxidation, H_2O_2 and superoxide radical production, and higher contents of chlorophyll a, b and total chlorophyll than the control (Djanaguiraman *et al.,* 2005). The combination between drought stress and Se supply significantly increased root activity. This indicated that Se supply further increased the resistance of wheat seedlings to drought stress (Xiaoqin *et al.,* 2009) addition of SA significantly alleviated the plant growth inhibition (Bai *et al., 2009)*

Table (1): Effect of selenium and salicylic acid on some growth characters of pea plants grown under drought stress during two seasons (2007/2008 and 2008/2009).

Characters / Treatments	Plant height (cm)	Root length (cm)	Root dry matter (g)/p	Shoot dry matter (g)/p	Root / shoot ratio	Leaf Area (cm^2/p)
	2007 / 2008 season					
W_0	31.67^{cd}	3.17^{c}	0.057^{c}	1.06^{c}	0.054	347.2^{d}
W_0+Se	36.00^{bc}	3.67^{b}	0.070^{b}	1.49^{b}	0.047	380.5^{bc}
W_0+SA	35.00^{bc}	3.83^{b}	0.067^{b}	1.52^{b}	0.044	397.2^{ab}
W_1	28.00^{d}	3.07^{c}	0.040^{d}	0.90^{c}	0.044	300.9^{e}
W_1 + Se	41.67^{a}	6.17^{a}	0.079^{a}	1.74^{a}	0.045	427.4^{a}
W_1 + SA	39.67^{ab}	6.00^{a}	0.097^{a}	1.59^{ab}	0.061	420.8^{a}
W_2	22.00^{e}	2.83^{c}	0.020^{d}	0.60^{d}	0.033	260.0^{f}
W_2 + Se	35.33^{bc}	3.67^{b}	0.050^{c}	1.06^{c}	0.047	357.2^{cd}
W_2 + SA	31.44^{cd}	2.83^{c}	0.050^{c}	1.04^{c}	0.048	330.3^{de}
	2008 / 2009 season					
W_0	26.33^{c}	3.33^{c}	0.047^{de}	1.07^{d}	0.044	396.8^{c}
W_0+Se	28.33^{bc}	4.27^{b}	0.060^{c}	1.56^{b}	0.038	460.0^{b}
W_0+SA	29.67^{ab}	4.00^{b}	0.060^{c}	1.37^{c}	0.044	446.9^{b}
W_1	22.00^{d}	2.00^{d}	0.040^{e}	0.95^{d}	0.042	350.7^{d}
W_1 + Se	32.33^{a}	5.77^{a}	0.123^{a}	1.75^{a}	0.070	487.0^{a}
W_1 + SA	31.00^{ab}	5.93^{a}	0.113^{b}	1.57^{b}	0.072	478.7^{a}
W_2	20.33^{d}	1.50^{d}	0.030^{f}	0.65^{e}	0.046	300.5^{e}
W_2 + Se	25.67^{c}	3.67^{c}	0.057^{cd}	1.00^{d}	0.057	392.5^{c}
W_2 + SA	23.00^{d}	3.90^{c}	0.050^{cde}	1.07^{d}	0.047	377.3^{c}

Means followed by the same letter within each column are not significantly different according to Duncan's Multiple Range Test (P≤0.05).
W_0 = 100% Field capacity (control) W_1= 60% Field capacity W_2= 40% Field capacity
Se= Selenium (0.6 mM) SA= Salicylic acid (0.5 mM)

Water stress decreased root / shoot ratio and the protective effect of Se and SA against water stress was shown by the greater root/shoot ration of treated plants under water stress. The highest significant values recorded with W_1 + SA followed by W_1 + Se in both seasons. Szepesi *et al.* (2005) show that, SA substantially improved tomato growth under salt stress conditions. Sakhabutdinova *et al.* (2003) showed that, SA treatment completely prevented stress induced declines in the concentration of IAA and cytokinens in seedlings and reduced accumulation of ABA, which might be a prerequisite for acceleration of growth.

Water use efficiency (WUE):

High drought level (W_2) showed a significant decline in WUE (23%) compared with W_0 (Table 2). Se and SA treatments caused a significant increase in WUE compared with droughted plants (W_1 and W_2) The greatest increase in WUE was observed with W_1+Se (25 and 39%) and W_1 +SA (16 and 24%) compared with W_0 in the first and second seasons, respectively. These data are confrmed by Rajasekaran and Blake (1999).

2- Water relations:

Pea plants treated with Se and SA showed an enhancing plant tolerance to drought stress as evidenced by the greater relative water content (RWC),osmotic potential (OP), leaf water potential(LWP) and lower membrane integrity, so its adverse drought effect is less (Table 2).

Table (2): Effect of selenium and salicylic acid on water use efficiency and water relations of pea plants grown under drought stress during two seasons (2007/2008 and 2008/2009).

Characters / Treatments	RWC (%)	Leaf water potential (Ψw)-Mpa	Osmotic potential (Ψπ)-Mpa	Pressure potential Ψp - Mpa	MI (%)	WUE g Dr.wt. kg^{-1} H_2o. day^{-1}
	2007 / 2008 season					
W_0	71.84^{d}	1.50^{e}	6.26^{b}	4.76^{a}	74.90^{b}	1.58^{cd}
W_0+Se	80.01^{c}	1.86^{e}	6.13^{b}	4.27^{a}	65.72^{c}	1.67^{b}
W_0+SA	75.90^{c}	2.30^{c}	6.10^{b}	3.80^{b}	66.93^{c}	1.67^{b}
W_1	65.55^{d}	4.82^{b}	7.16^{a}	2.34^{c}	79.66^{a}	1.69^{d}
W_1 + Se	88.63^{a}	2.90^{d}	6.54^{b}	3.64^{b}	55.06^{d}	1.97^{a}
W_1 + SA	85.30^{b}	2.50^{d}	6.10^{b}	3.60^{b}	58.20^{d}	1.83^{a}
W_2	57.49^{e}	5.95^{a}	7.72^{a}	1.77^{d}	82.86^{a}	1.22^{d}
W_2 + Se	75.18^{c}	3.50^{c}	6.95^{b}	3.45^{b}	67.83^{c}	1.69^{b}
W_2 + SA	73.90^{d}	3.00^{c}	6.90^{b}	3.90^{b}	69.99^{c}	1.64^{b}
	2008 / 2009 season					
W_0	76.85^{c}	1.62^{f}	5.79^{b}	4.17^{a}	70.08^{b}	1.59^{d}
W_0+Se	82.81^{b}	1.75^{f}	5.90^{b}	4.15^{a}	63.21^{b}	1.78^{c}
W_0+SA	81.32^{b}	2.20^{e}	5.85^{b}	3.65^{b}	65.42^{b}	1.75^{c}
W_1	70.28^{d}	4.91^{b}	6.50^{a}	1.59^{d}	80.14^{a}	1.66^{c}
W_1 + Se	88.19^{a}	2.85^{d}	6.10^{b}	3.25^{b}	49.25^{c}	2.21^{a}
W_1 + SA	86.40^{a}	2.10^{e}	5.95^{b}	2.85^{c}	54.51^{c}	1.97^{b}
W_2	60.45^{e}	5.91^{a}	6.90^{a}	0.99^{e}	83.03^{a}	1.33^{e}
W_2 + Se	75.99^{c}	3.60^{c}	6.50^{a}	2.90^{c}	55.01^{c}	1.65^{c}
W_2 + SA	76.34^{c}	2.80^{d}	6.20^{a}	3.40^{b}	68.14^{b}	1.66^{c}

Means followed by the same letter within each column are not significantly different according to Duncan's Multiple Range Test (P≤0.05).

W_0 = 100% Field capacity (control) W_1= 60% Field capacity W_2= 40% Field capacity
Se= Selenium (0.6 mM) SA= Salicylic acid (0.5 mM)

To regarding RWC, the highest data were recorded under W_1 with Se and SA. Whereas, RWC decreased with increasing drought but Se and SA increased it under drought conditions in both seasons.

Leaf water potential (Ψw) decreased with increasing water stress (more negative). Application Se and SA under drought recorded highest Ψw. The same trend was observed in osmotic potential (Ψπ), meanwhile the pressure potential (Ψp) decreased with increasing water stress and the maximum value was recorded with Se under the control and followed by SA. Application Se and SA under water stress recorded a good water status

compared with stressed plants alone (Germ, 2008). Se application improved water relations in plant. The strong correlation between plant water relation components had the accumulation of compatible solutes (GB and Free proline) under drought indicated the involvement of compatible solutes with the maintenance of Ψ_w, Ψ_p and improved leaf water status under drought (Farooq *et al.,* 2009).

The increased water potential values in SA treatment under ionic and non-ionic osmotic stress suggest that accumulation of inorganic or organic osmolytes makes the surplus of water uptake possible as it can also be seen from the increase in RWC (Szepesi *et al.,* 2005). Recently it has been shown that Se has the ability to regulate water status of plants under drought condition, Se causes enhance water relation in wheat tissues (Kuznetsov *et al.,* 2003 and Djanaguiraman *et al.,* 2005) they cited that Se caused a higher Ψ_w in leaves, that could enable higher transpiration rate.

The interesting data was membrane integrity which was highest and increased in membrane leakage under W_2 compared with W_0. A shift in phospholipid concentration could explain membrane damage because drought increased lipids peroxidation results from the formation of free radicals (O^-_2 , H_2O_2 and / or OH^-) which destabilize chloroplast, mitochondrial and / or microsomal membranes. Moreover, plants treated with Se and SA under drought stress recorded minimum membrane leakage which was the best results. Se applications helped to maintain membrane integrity under drought degrees, which were the most effective compounds in reducing leakage by 30.9(W_1+Se) and 18%(W_2 +Se) compared to droughted plants (W_1 and W_2), respectively in the first season and the same trend was observed in the second season. These results are in agreement with Farooq *et al.* (2009) and Rajasekaran and Blake (1999).

3- Chemical compostion:

(A):Photosynthetic pigments:

Data in Table (3) recorded a highly significant reduction in chl. a + b and carotenoids in pea leaves grown under drought stress. These reductions in the first season were 14.5 and 50.7% of chl. a+b, 12.2 and 20.1% of carotenoids under W_1 and W_2, respectively compared with W_0. A maximum values of chl. a + b were recorded with Se and/or SA treated plants especially with W_1 and the increases under W_2 were 60 and 56%, respectively compared with W_2 only. These results are in agreement with those obtained by Grem (2008) on potato. The increased chlorophyll content by SA and Se treated plants might be attributed to efficient scavenging of ROS by oxidase and glutathione peroxidase or otherwise, they would have destroyed the chlorophyll pigments (Thomas *et al.,* 2001). SA application in drought stressed wheat increased the photosynthetic pigments and carboxylase activity of Rubisco (Singh and Usha 2003)

Table (3): Effect of selenium and salicylic acid on photosynthetic pigments, proline, total soluble sugars, total free amino acid and potassium of pea leaves grown under drought stress during two seasons (2007/2008 and 2008/2009).

Characters / Treatments	Chl. a + b mg/g dwt	Carotenoids mg/g dwt	Proline µg g^{-1} dwt	Total soluble sugar mg/g dwt	Total free amino acid (mg/g dwt)	K (%)
	2007 / 2008 season					
W_0	2.21^{c}	1.39^{c}	318.9^{d}	16.44^{c}	25.72^{bc}	2.45^{c}
W_0+Se	2.31^{c}	1.50^{b}	330.8^{d}	20.00^{b}	28.63^{abc}	2.81^{b}
W_0+SA	2.65^{b}	1.28^{d}	325.8^{d}	22.00^{b}	29.8^{abc}	2.96^{b}
W_1	1.89^{d}	1.22^{d}	360.8^{c}	15.76^{e}	20.78^{c}	2.00^{e}
W_1 + Se	2.63^{b}	1.57^{b}	390.4^{b}	26.90^{a}	39.12^{a}	3.44^{a}
W_1 + SA	2.96^{a}	1.68^{a}	400.8^{b}	25.99^{a}	37.12^{ab}	3.22^{a}
W_2	1.09^{f}	1.11^{e}	440.5^{b}	12.88^{e}	19.68^{c}	1.32^{f}
W_2 + Se	1.74^{e}	1.18^{e}	460.8^{a}	16.44^{c}	25.64^{bc}	1.99^{d}
W_2 + SA	1.70^{e}	1.37^{c}	480.5^{a}	16.22^{c}	22.76^{abc}	1.84^{d}
	2008 / 2009 season					
W_0	2.91^{c}	1.60^{c}	340.6^{ef}	21.22^{c}	29.75^{bcd}	2.38^{c}
W_0+Se	3.04^{c}	1.71^{b}	350.4^{e}	24.58^{b}	34.88^{bc}	2.67^{b}
W_0+SA	3.65^{b}	1.71^{b}	355.9^{e}	26.38^{b}	34.73^{bc}	2.77^{ab}
W_1	2.29^{d}	1.30^{e}	380.9^{d}	20.18^{c}	28.25^{bcd}	2.25^{c}
W_1 + Se	3.49^{b}	2.23^{a}	410.9^{c}	30.00^{a}	47.90^{a}	2.99^{a}
W_1 + SA	3.96^{a}	2.54^{a}	420.4^{c}	30.40^{a}	43.08^{ab}	2.98^{a}
W_2	2.08^{e}	1.16^{d}	450.6^{b}	15.80^{e}	18.38^{e}	1.22^{e}
W_2 + Se	2.75^{bc}	1.64^{c}	490.3^{a}	23.52^{b}	25.13^{cde}	1.69^{d}
W_2 + SA	2.67^{bc}	1.64^{c}	500.4^{a}	22.60b^{b}	23.44^{de}	1.60^{d}

Means followed by the same letter within each column are not significantly different according to Duncan's Multiple Range Test (P≤0.05).
W_0 = 100% Field capacity (control) W_1= 60% Field capacity W_2= 40% Field capacity
Se= Selenium (0.6 mM) SA= Salicylic acid (0.5 mM)

(B):Antioxidant Enzymes activity:

Regarding to the activity of peroxidase, phenoloxidase, catalase and glutathione peroxidase (Figs.1,2,3 and 4), W_2 decreased the activity of previous enzymes.These values recorded 24, 23, 28 and 33% in the first season respoctivelly, compared with control. Significant increase in the activity of peroxidase, phenoloxidase catalase and glutathione peroxidase was seen by Se and SA treatments over control or drought stress. Generally, W_1 +Se followed by W_1+SA recorded the highest values for all studied enzymes. Exogenous Se and SA application led to increases in antioxidant capacity (Ananieva *et al.,* 2002), and stimulate enzymes activities in plant cells (Fazeli *et al.,* 2007) because of enhanced accumulation of hydrogen peroxide under such conditions (Rao *et al.,* 1997 and Janda *et al.,* 2003), it is known that water and salt stress induced the generation of reactive oxygen species (ROS) in plants (Polle, 1997 and Borsani *et al.,* 2001). It is possible that Se and SA promotes scavenging of produced H_2O_2 through increased glutathione peroxidase activity and consequently, reduces the need for their scavenger ROS (Djanaguiraman *et al.,* 2005 and Shakirova, 2007). It is possible that the presently observed induction of such enzymes by SA and Se can explain the improvement of pea plants growth under water stress

(Djanaguiraman *et al.,* 2005 ,Rios *et al.,* 2009 and Horvath *et al.,* 2007).

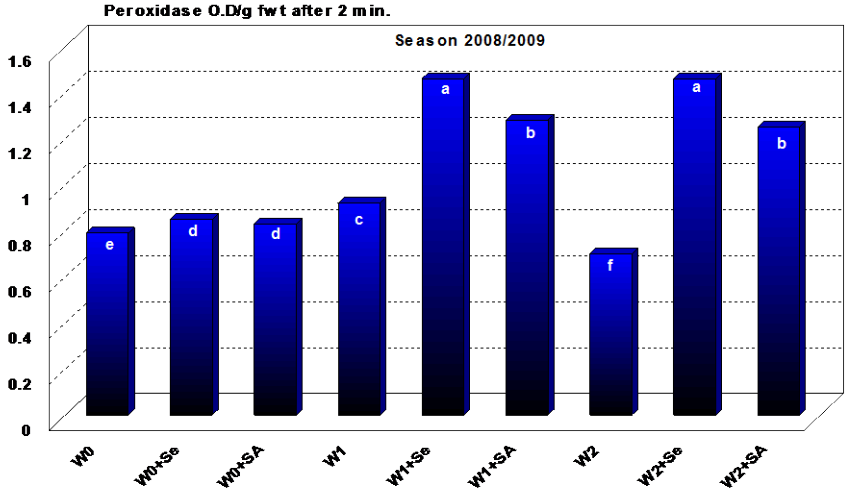

Figure (1): Effect of Selenium and Salicylic acid on Peroxidase of pea plants grown under drought stress during season 2008/2009.

Means followed by the same letter within each column are not significantly different according to Duncan's Multiple Range Test (P≤0.05).

W_0 = 100% Field capacity (control) W_1= 60% Field capacity W_2= 40% Field capacity

Se= Selenium (0.6 mM) SA= Salicylic acid (0.5 mM)

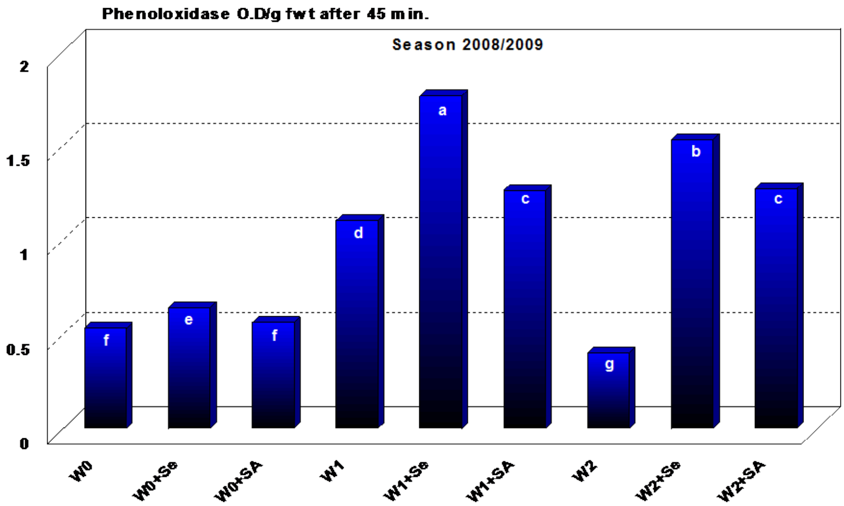

Figure (2): Effect of Selenium and Salicylic acid on Phenoloxidase of pea plants grown under drought stress during season 2008/2009.

Means followed by the same letter within each column are not significantly different according to Duncan's Multiple Range Test (P≤0.05).

W_0 = 100% Field capacity (control) W_1= 60% Field capacity W_2= 40% Field capacity
Se= Selenium (0.6 mM) SA= Salicylic acid (0.5 mM)

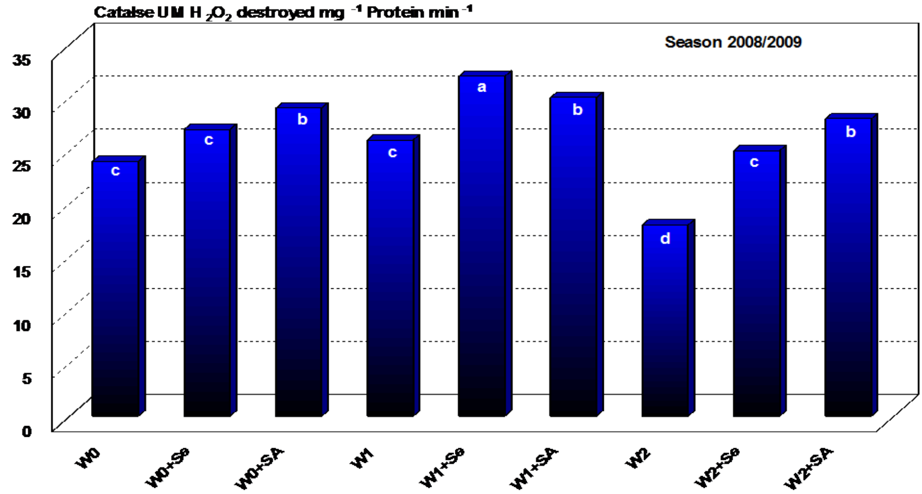

Figure (3): Effect of Selenium and Salicylic acid on Catalase of pea plants grown under drought stress during season 2008/2009.

Means followed by the same letter within each column are not significantly different according to Duncan's Multiple Range Test ($P \leq 0.05$).
W_0 = 100% Field capacity (control) W_1= 60% Field capacity W_2= 40% Field capacity
Se= Selenium (0.6 mM) SA= Salicylic acid (0.5 mM)

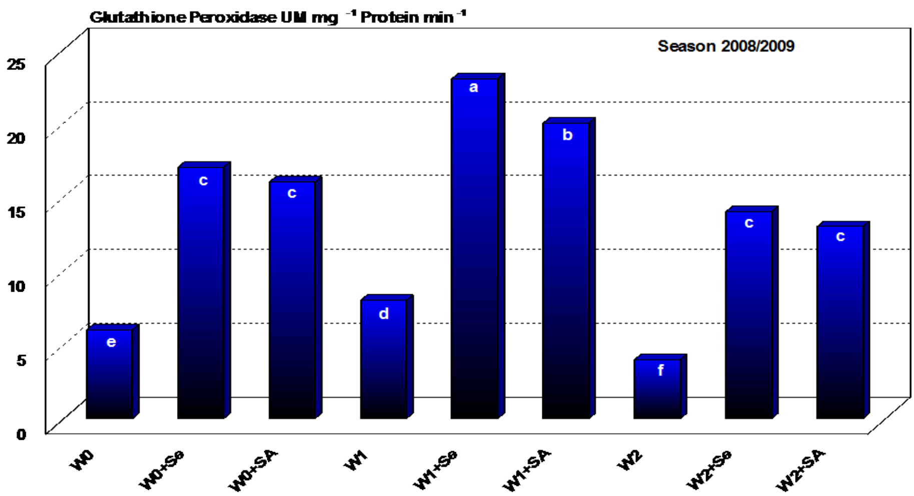

Figure (4) : Effect of Selenium and Salicylic acid on Glutathione Peroxidase of pea plants grown under drought stress during season 2008/2009.

Means followed by the same letter within each column are not significantly different according to Duncan's Multiple Range Test ($P \leq 0.05$).
W_0 = 100% Field capacity (control) W_1= 60% Field capacity W_2= 40% Field capacity
Se= Selenium (0.6 mM) SA= Salicylic acid (0.5 mM)

4- Compatible osmolytes (proline, total soluble sugars, total free amino acids and K^+):

Proline concentration was significantly increased under water stress. Meanwhile, SA and Se treated plants recorded a highly increases in proline concentration compared with untreated plants especially under W_1 and W_2 levels (Table 3). The protective role of proline against water stress in higher plants is widely recognized by Farooq *et al.,* (2009). Moreover, total soluble sugars, total free amino acid and K^+ content (Table 3) significantly decreased under W_2 treatment, meanwhile Se and SA treated plants under drought had significant induction compared the droughted plants. The accumulation of these previous osmolytes seems to correlate with greater tolerance against stress. Drought tolerance in rice was well associated with the accumulation of compatible solutes (leaf GB and free proline), maintenance of tissue water protential and enhanced potency of antioxidant system, which improved the integrity of cellular membranes and facilitated the rice plant to sustain photosynthesis and general metabolism (Farooq *et al.,* 2009 and Xiaoqin *et al.,* 2009)

5- Yield and its components:

Drought had a significant reduction on average number of green pods and weight of green pods as well as weight of seeds / plant (Table 4). The highest reduction recorded with W_2 treatment. The highest yield recorded with W_1 + Se followed by W_1 + SA. The same trend was observed in both seasons.

These results call for further studies of the role of Se and SA in protecting plants against drought stress. However, they clearly indicated that Se and SA may exert a beneficial role in plants under stress and thus, they support the finding of Germ (2008) that Se and SA defend plants subjected to drought. Even if the present study provided some support for the Se and SA induced antioxidatives enzymes activities which explain the enhanced antioxidatives capacity and stimulated growth with good water relations which reflected higher yield of pea droughted plants.

Finally, it can be concluded from this work that adding Se and SA as foliar application on pea plants grown under water stress can show a better performance with good yield and its quality by enhancing the antioxidant defense system in leaves of pea plants.

Table (4): Effect of selenium and salicylic acid on total weight of green pods, average number of green pods and weight of seeds of pea plants grown under drought stress during two seasons (2007/2008 and 2008/2009).

Treatments \ Characters	Total weight of green pods (g/plant)	Average number of green pods / plant	Weight of seeds/ plant
	2007/2008 season		
W_0	5.47c	3.53d	3.33cd
W_0+Se	6.27c	3.90c	4.01cd
W_0+SA	5.97c	3.90c	3.40cd
W_1	4.00d	3.00e	3.00d
W_1 + Se	8.75a	5.24a	7.44a
W_1 + SA	7.50b	4.83b	5.43b
W_2	3.74d	2.93e	2.20e
W_2 + Se	5.61c	3.97c	4.31c
W_2 + SA	5.64c	3.63d	3.37cd
	2008/2009 season		
W_0	6.29c	4.00d	4.31e
W_0+Se	6.98c	4.97c	5.04d
W_0+SA	7.21c	4.77c	5.45c
W_1	4.00e	3.40e	4.00f
W_1 + Se	9.39a	6.40a	7.58a
W_1 + SA	8.48b	5.17b	6.66b
W_2	3.25f	3.20f	2.60g
W_2 + Se	6.30d	3.83d	5.41c
W_2 + SA	7.10c	3.60e	4.60e

Means followed by the same letter within each column are not significantly different according to Duncan's Multiple Range Test ($P \leq 0.05$).
W_0 = 100% Field capacity (control) W_1= 60% Field capacity W_2= 40% Field capacity
Se= Selenium (0.6 mM)   SA= Salicylic acid (0.5 mM)

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تحمل نباتات البسلة لظروف الجفاف وعلاقة ذلك باستخدام مضادات الاكسدة

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الملخص العربي

تم زراعة بذور البسلة صنف ماستر بى تحت ظروف الصوبة خلال الموسم الشتوى لعامى ٢٠٠٧/٢٠٠٨ و ٢٠٠٨/٢٠٠٩ تحت ثلاث مستويات من الجفاف ١٠٠% (كمقارنة) و٦٠و ٤٠% من السعة الحقلية واجرى هذا البحث بمزرعة كلية الزراعة ـ جامعة المنوفية وذلك لدراسة تـأثير استخدام الـرش بالسلنيوم وحامض السلسيليك كمضادات للأكسدة على تقليل الاضرار الناتجة من الجفاف على النمو و العلاقات المائية و الصفات الكيماوية و المحصول وكانت اهم النتائج المتحصل عليها كالتالى :

- **ادى التعرض للجفاف الى نقص معنوى فى طول النبات وطول الجذور و الوزن الجاف للجذور والساق ومساحة الاوراق وايضا نقص معنوى فى تركيز الكلوروفيلات و الكاروتينيدات والسكريات الكلية الذائبة و الاحماض الامينية بينما حدث العكس فى محتوى الاوراق من البرولين**
- **أدى تعرض نباتات البسلة لمعاملات الجفاف الى نقص معنوى فى محتوى الماء النسبى والضغط الاسموزى وكذلك الضغط المائى للاوراق الخضراء بينما حدث العكس بالنسبة لنفاذية الجدر الخلوية حيث ادى الجفاف لارتفاع هذة النسبة ارتفاعاً معنوياً مقارنة بنباتات المقارنة**

من جهة اخرى ادت معاملات الجفاف الى نقص معنوى فى وزن القرون الخضراء وعددها ووزن البذور للنبات مقارنة بنباتات المقارنة.

اوضحت الدراسة انة بإستخدام كلاً من السلنيوم وحامض السلسيليك (تحت ظروف الجفاف) ادى الى تحسن واضح فى صفات النمو الخضرى والعلاقات المائية (محتوى الماء النسبى

والضغط المائى للاوراق) والصبغات و السكريات الذائبة و الاحماض الامينية و المحصول. كذلك ادت المعاملة بالسلنيوم وحامض السلسيليك الى زيادة فى نشاط الانزيمات (البيروكسيديز- والفينول كسيديز والكتاليز والجلوتاثيون بيروكسيديز) وعنصر البوتاسيم مقارنة بمعاملات الاجهاد المائى منفردة

وخلاصة هذه الدراسة وجد ان المعاملة بالسلنيوم او حمض السلسيليك تحت ظروف الجفاف أدى الى تحسن واضح فى الصفات الخضرية للنبات وكذلك المحصول وذلك بتحسن واضح فى العلاقات المائية وكفاءة استخدام الماء وايضاً زيادة فى نشاط انزيمات الاكسدة التى ربما تحمى النبات من الشقوق الحره التى تتكون تحت ظروف الجفاف

Peroxidase o.D/g fwt after 2 min^{-1}.

Phenoloxidase o. D/g fwt after 45 min^{-1}.

Catalase μM H_2O_2 destroyed mg^{-1} Protein min^{-1}.

Glutathione Peroxidase μM mg^{-1} Protein min^{-1}.

Peroxidase o.D/g fwt after 2 min^{-1}.

Phenoloxidase o. D/g fwt after 45 min^{-1}.

Catalase μM H_2O_2 destroyed mg^{-1} Protein min^{-1}.

Glutathione Peroxidase μM mg^{-1} Protein min^{-1}.

Peroxidase o.D/g fwt after 2 min^{-1}.

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Peroxidase o.D/g fwt after 2 min^{-1}.

Phenoloxidase o. D/g fwt after 45 min^{-1}.

Catalase μM H_2O_2 destroyed mg^{-1} Protein min^{-1}.

Glutathione Peroxidase μM mg^{-1} Protein min^{-1}.