BRIEF COMMUNICATION

Amelioration of NaCl stress by triadimefon in soybean seedlings

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Abstract

NaCl stress decreased root growth, shoot length and dry matter production of *Glycine max* seedlings. It has also caused accumulation of proline and amino acids and decreased protein and nucleic acid contents of the seedlings. Addition of triadimefon to NaCl stressed seedlings partially restored the growth and increased the protein, amino acid, proline and nucleic acid contents of the seedlings. The root biomass production under combination of triadimefon and NaCl was even larger than control. Thus triadimefon can ameliorate the effect of NaCl stress in soybean.

Additional key words: amino acids, *Glycine max*, growth, nucleic acids, proline, proteins.

Salt stress is harmful during early stages of germination and seedling growth and its effect depends upon the concentration of the salts and plant species. Triadimefon is a triazole compound which have both fungitoxic and plant growth regulating properties (Fletcher 1985). Triazoles protect plants against various stresses including drought, low and high temperatures, salinity and air pollution and their protective effects are mediated by shifting the balance of important plant hormones including gibberellins, abscisic acid and cytokinins (Fletcher and Hofstra 1988). Triadimefon stimulated the growth of the water stressed seedlings of apple (Kulbe 1981) and barley (Buchenauer and Rohner 1981). In the present study an attempt has been made to determine the effect of NaCl and triadimefon on growth, protein, amino acid, proline, RNA and DNA contents in *Glycine max* seedlings.

Seeds of *Glycine max* L. Merrill. cv. CO-1 were obtained from Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu, India. Triadimefon (*Bayleton*) 25 % WP was obtained from *Bayer Ltd.*, Bangalore, India. Seeds were surface

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sterilized with 0.1 % $HgCl_2$ for 5 min followed by thorough rinsing with distilled water. Then the seeds were soaked for 12 h in distilled water (control), 60 mM NaCl, 60 mM NaCl + 5 mg dm^{-3} triadimefon and triadimefon alone (5 mg dm^{-3}). Later they were germinated in Petri plates containing *Whatman No.1* filter paper and irrigated with fixed volume of deionized water (control) and respective treatment solutions and maintained at temperature 28 ± 1 °C and irradiance of 250 μmol m^{-2} s^{-1} for 14 h per day till the end of experiment. The seedlings were harvested and washed with deionized water and separated into root, shoot and cotyledons on day 13 after sowing and used for the determination of dry mass and biochemical parameters.

The soluble protein content was determined using the method of Bradford (1976). Bovine serum albumin was used as standard. Total free amino acid content was estimated with the alcoholic ninhydrin reagent (Moore and Stein 1948), using leucine as standard. Free proline content was estimated using the method of Bates *et al.* (1973). The contents of DNA and RNA were estimated by the method of Neiman and Poulson (1963) using diphenylamine reagent for DNA (Burton 1956) and orcinol reagent for RNA (Peach and Tracey 1955).

NaCl salinity caused a reduction in root and shoot length. Addition of triadimefon to the NaCl stressed or unstressed seedlings increased the root length even above the level of control but reduced the shoot length below the level of control (Table 1). NaCl stress decreased the growth rate and biomass production in *Erythrina variegata* and damaged the polypeptides of the thylakoid membranes of the chloroplast by oxidation (Muthuchelian *et al.* 1996a,b). Pretreatment of seeds with a triazole compound LAB 150978 counteracted the inhibitory effect of salinity on root growth, but it inhibited the hypocotyl growth of sunflower and mungbean seedlings (Saha and Gupta 1993).

Table 1. Influence of triadimefon (5 mg dm^{-3}) on growth of NaCl (60 mM) stressed seedlings of *Glycine max* [means of 7 replicates significantly different at $P = 0.05$ (*) and 0.01 (**), respectively].

Parameters	Control	NaCl	NaCl + triadimefon	Triadimefon
Root length [cm]	1.21	0.57**	1.82**	1.46**
Shoot length [cm]	1.52	0.96**	0.76**	0.54**
Root dry mass [mg $seedling^{-1}$]	1.07	0.53**	1.36*	2.04**
Shoot dry mass [mg $seedling^{-1}$]	1.14	0.59*	0.80*	0.95**
Root/shoot ratio	0.94	0.90	1.70	2.14
Cotyledon dry mass [mg $seedling^{-1}$]	8.10	9.53*	9.09*	8.33*

NaCl stress caused a decrease in dry mass of the roots and shoots (Table 1). Treatment with triadimefon alone or in combination with NaCl increased the root dry mass while the shoot dry mass was lower than in control seedlings (Table 1). In cucumber seedlings the fresh and dry masses of the hypocotyl were lowered while those of cotyledons and roots were increased by the triadimefon treatment (Fletcher and Arnold 1986). Similar results were obtained with radish (Panneerselvam *et al.* 1997).

The NaCl stress decreased the protein content of root and shoot, while the protein content of cotyledons were higher than in the control (Table 2). The higher protein content in the cotyledons of NaCl stressed seedlings may indicate the slower hydrolysis of the storage proteins in the cotyledons and restricted mobilization of storage proteins to the embryonic axis as was found in the germinating mung bean seeds under water stress (Ranu and Kar 1994). The protein content of root and shoot increased and that of cotyledons decreased in the tiadimefon treated NaCl stressed and unstressed seedlings (Table 2). The protein content has been shown to decline in the cotyledons of soybean with uniconazole (a triazole compound) treatment (Kraus *et al.* 1993). Paclobutrazol (a triazole compound) treatment increased the protein content in the seedlings of *Brassica carinata* (Setia *et al.* 1995).

Table 2. Effect of NaCl (60 mM) and triadimefon (5 mg dm^{-3}) on protein, amino acid, DNA and RNA contents [mg g^{-1}(f.m.)] in soybean seedlings [means of 7 determinations significantly different at $P = 0.05$ (*) and 0.01 (**), respectively].

Treatment	Parameter	Roots	Shoots	Cotyledons
Control	protein	0.79	1.64	7.92
	amino acids	6.05	5.56	5.07
	proline	0.36	0.32	0.29
	DNA	0.55	0.43	0.42
	RNA	2.76	1.67	1.82
NaCl	protein	0.49*	1.24*	8.33**
	amino acids	7.56*	6.12*	5.97
	proline	0.73**	0.52*	0.49**
	DNA	0.27**	0.20**	0.48*
	RNA	1.20**	0.96**	1.88**
NaCl + triadimefon	protein	0.96*	1.52**	5.65**
	amino acids	8.84**	7.33**	6.12
	proline	1.92**	1.07**	0.79**
	DNA	0.58**	0.37**	0.42
	RNA	2.86**	1.38**	1.66**
Triadimefon	protein	1.29**	1.90*	4.96**
	amino acids	7.92*	6.87*	6.05
	proline	0.97**	0.86**	0.68**
	DNA	0.61**	0.42	0.44
	RNA	2.98**	1.56**	1.71**

Total free amino acids and proline increased in the seedlings under NaCl and triadimefon treatments, and the increase was higher in the triadimefon treated NaCl stressed seedlings. Highest amino acid content was recorded in roots followed by shoots and cotyledons (Table 2). In bean and black gram the amino acid content was increased with increasing concentration of salinity (Huber *et al.* 1977, Anilkumar *et al.* 1996). Proline accumulated significantly more in the roots than the shoots of salt stressed green gram seedlings (Sudhakar *et al.* 1993) and soybean seedlings (Durgaprasad *et al.* 1996). In sunflower and mungbean, LAB 150978 increased the free amino acid and proline content, which might act as an osmoticum to counteract salinity stress (Saha and Gupta 1993). Also mulberry plants treated with triadimefon showed a appreciable increase in free proline content (Sreedhar 1991).

Sodium chloride stress caused decrease in the DNA and RNA content in the roots and shoots but maintained a higher DNA and RNA content in the cotyledons (Table 2). The higher DNA and RNA content in the cotyledons may be due to the inhibition of hydrolysis and translocation of nucleotides from the cotyledon to the growing parts. Similar observations were made in *Lens esculenta* (Tewari and Singh 1991). and *Vicia faba* (Abdel-Samad 1993). The decreased content of nucleic acid in the salt affected roots and shoots may also be due to the increased activities of DNase and RNase as reported by Tewari and Singh (1991). Addition of triadimefon to the NaCl stressed and unstressed plants increased the DNA and RNA content in roots and shoots to a level higher than in NaCl stressed seedlings. Triadimefon and triadimenol (a triazole) treatments increased the nucleic acid content in the primary leaves of barley (Forster 1977).

In conclusion, NaCl stress inhibited the growth and decreased protein and nucleic acid contents as a consequence of decreased translocation of reserves from the cotyledons to the growing parts of the soybean seedlings. Triadimefon treatment of the NaCl stressed and unstressed seedlings increased dry mass of roots, protein, amino acid and nucleic acid content. However, the shoot growth was inhibited by the triadimefon treatment.

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