Pak. J. Sci. Ind. Res. 2008 **51**(2) 98-102

Effect of Halopriming on Sunflower Seed Germination and Seedling Establishment under Saline Environment

A. Shahzad, I. A. Mahmood*, Madeeha Khan and A. Ali
Land Resources Research Program, National Agricultural Research Centre, Park Road, Islamabad-45500, Pakistan

(received December 24, 2007; revised April 13, 2008; accepted April 15, 2008)

Abstract. The effect of halopriming of sunflower seeds with inorganic salts (KNO_3 and K_2SO_4) on seed germination and seedling establishment under salt stress (200 mM NaCl) was studied. Priming of sunflower seeds using low concentration of salt improved germination. Hundred percent germination and better seedling establishment were recorded with 2% K_2SO_4 at 200 mM NaCl salt stress. In plant tissues, higher concentration of Na^+ (0.61%) was observed in case of 2% KNO_3 and minimum with 2% K_2SO_4 (0.26%) and higher concentration of K^+ was observed with 2% K_2SO_4 followed by 1% K_2SO_4 and 2% KNO_3. Maximum concentrations of Ca and Mg in plant tissues were found with 2% K_2SO_4.

Keywords: halopriming, sunflower seeds, inorganic salts, salt stress, saline environment

Introduction

Sunflower (*Helianthus annus* L.) is the second important non-conventional source of vegetable oil in the world after soybean (Sackston, 1981). National average yield has increased from 750 to 1500 kg/ha but it is still quite low compared with other sunflower growing countries of the world (GOP, 2005). Sunflower is moderately salt tolerant crop therefore it has a wide margin to be grown successfully on most of the soils. It has 47.5% oil content and is ranked next to peanut (48.5) in terms of oil content among flax, soybean, safflower, cottonseed, mustard and rape (Teasar, 1984). The threshold for the sunflower, soybean and safflower is nearly identical; the rate of yield decline above the threshold is lesser for sunflower as compared to soybean and safflower (Francois, 1996).

Salinity has been recognized as a limiting factor for crop productivity. In the past two decades, intensive research has been conducted to improve seed germination and seedling emergence under saline conditions by pretreating seeds with solutions of different inorganic salts. Various studies indicate considerable improvement in seed germination, emergence, seedling establishment and final crop yield in salt-affected soils as a result of seed halopriming (Ashraf *et al.*, 1987; Ayers, 1952). Although salinity effect involved various aspects of plant life, it was not easy to establish a chain of priorities (Flowers *et al.*, 1977). It may not be simple to get a scale of salt tolerance for plants because some species are sensitive to salt conditions in certain periods of their growth and development while they become resistant in others (Sayed, 1985). In saline soils, salt concentration in soil solution creates high osmotic pressure reducing the availability of water to plant and specific ion effects such as toxicity of sodium and chloride. In order to meet the ever increasing vegetable oil demand, expansion of the area under sunflower cultivation is need of the day. The present laboratory investigation was planned to facilitate and enhance germination of sunflower seeds by priming with inorganic salts under NaCl salt stress.

*Author for correspondence

Materials and Methods

The experiment was conducted to study the effect of halopriming on sunflower seed germination and establishment under saline conditions in Soil Salinity Laboratory at National Agricultural Research Centre (NARC), Park Road, Islamabad during the year 2007. Seeds of sunflower were obtained from Oilseed Programme, NARC. Before the start of experiment, the seeds were washed, air dried and haloprimed for 6 h with inorganic salts (1%, 2%, 3%, 4%, 5% KNO_3 and K_2SO_4 + H_2O each) separately, whereas the control was treated with distilled water.

After halopriming, the seeds were taken out and air dried. Ten seeds per petri dish of each treatment were put for germination. Experiment was organized in two factor completely randomized design (CRD) using three replications of each treatment. Germination of controlled and treated seeds was evaluated between two layers of moist filter paper in covered petri dishes. NaCl solution was applied to develop 200 mM salinity except control. For control treatment, distilled water was used. The petri plates were kept covered for 20 h a day and were removed each day for four hours during germina period. After complete germination, the lids of petri plates were removed for seedling development. The germination percentage was recorded 2, 4, 6 and 9 days after the beginning of the experiment. After two weeks of germination, when the seedlings were fully established, root and shoot lengths were measured. Dried root and shoot samples were placed in oven at 60 °C to a constant weight and the dry matter yield was recorded. Ground plant samples were digested in perchloric-nitric acid (2:1 1 N) mixture (Rhoades, 1982) to estimate Na, K, Ca and Mg by atomic absorption spectrophotometer. The data thus obtained was subjected to statistical analysis according to GEM Stat.

Results and Discussion

Effects of different holopriming treatments on sunflower seed germination, shoot, root growth and yield and on ionic concentration in sunflower tissues are depicted in Table 1 and 2, respectively.

Seed germination. Comparison of treatment means elaborates that 100% seed germination was recorded when sunflower seeds were primed with 2% K_2SO_4 (Table 1). Haloprimed seeds with different concentrations of K_2SO_4 and KNO_3 showed more germination as compared to control (non primed-stressed) treatment. Among all treatments, priming with 4%, 5% KNO_3 and 5% K_2SO_4 gave relatively poor performance showing 76%, 73% and 70% germination, respectively. Priming with lower concentrations of KNO_3 and K_2SO_4 and even with distilled water gave better results as compared to the priming with higher concentrations of KNO_3 and K_2SO_4. It is obvious from the data that priming with higher concentrations of salts caused poor germination probably due to osmotic effect and insufficient moisture uptake by seeds. This effect was further aggravated when seeds were subjected to 200 mM NaCl stress. Similar findings have also been documented by Chang-Zheng *et al.* (2002), Ashraf and Rauf (2001) and Singh *et al.* (1999),

Table 1. Effect of different halopriming treatments on sunflower seed germination, shoot, root growth and yield (average of four repeats)

Treatment	Germination (%)	Shoot length (cm)	Root length (cm)	Shoot dry matter (g/plant)	Root dry matter (g/plant)
Unprimed control (grown in distilled water)	76.67 (±15.28)	13.77 (±2.87)	11.60 (±1.74)	0.79 (±0.13)	0.19 (±0.05)
Unprimed control (grown in 200 mM NaCl)	73.33 (±20.82)	4.88 (±0.77)	2.29 (±0.06)	0.32 (±0.06)	0.08 (±0.10)
1% KNO_3 primed (grown in 200 mM NaCl)	73.33 (±20.82)	7.80 (±1.38)	3.17 (±0.62)	0.32 (±0.02)	0.007 (±0.002)
2% KNO_3 primed (grown in 200 mM NaCl)	90.00 (±10.00)	6.97 (±2.15)	3.10 (±0.52)	0.38 (±0.08)	0.03 (±0.01)
3% KNO_3 primed (grown in 200 mM NaCl)	93.33 (±5.77)	8.38 (±0.78)	3.55 (±0.85)	0.44 (±0.04)	0.08 (±0.002)
4% KNO_3 primed (grown in 200 mM NaCl)	76.67 (±15.28)	3.35 (±1.20)	1.66 (±0.09)	0.28 (±0.02)	0.02 (±0.01)
5% KNO_3 primed (grown in 200 mM NaCl)	73.33 (±20.82)	3.83 (±0.43)	1.83 (±0.33)	0.31 (±0.03)	0.007 (±0.003)
1% K_2SO_4 primed (grown in 200 mM NaCl)	96.67 (±5.77)	8.46 (±1.97)	3.35 (±0.04)	0.37 (±0.02)	0.02 (±0.00)
2% K_2SO_4 primed (grown in 200 mM NaCl)	100.00 (±0.00)	8.80 (±3.20)	3.13 (±0.22)	0.45 (±0.04)	0.06 (±0.003)
3% K_2SO_4 primed (grown in 200 mM NaCl)	90.00 (±17.32)	7.95 (±0.25)	2.97 (±0.10)	0.34 (±0.07)	0.01 (±0.004)
4% K_2SO_4 primed (grown in 200 mM NaCl)	93.33 (±5.77)	6.41 (±0.84)	2.66 (±0.17)	0.30 (±0.02)	0.04 (±0.05)
5% K_2SO_4 primed (grown in 200 mM NaCl)	80.00 (±13.00)	6.63 (±1.20)	2.59 (±0.36)	0.39 (±0.07)	0.01 (±0.0009)
Distilled water primed (grown in 200 mM NaCl)	90.00 (±10.00)	6.07 (±0.95)	2.93 (±0.41)	0.25 (±0.02)	0.03 (±0.01)

who reported that sunflower seeds treated with dilute solutions of mixed salts germinated significantly more rapidly than that of unprimed seeds under salt stress condition.

Shoot and root growth. Maximum root and shoot growth (13.77 and 11.60 cm, respectively) was attained by the control without NaCl stress (Table 1). Nevertheless, reduction in root and shoot growth was noted due to salinity but priming with KNO_3 and K_2SO_4 caused a significant improvement in the growth of sunflower seedlings. Among all the treatments, 2% K_2SO_4 showed comparatively more shoot growth (i.e., 8.80 cm) followed by 1% K_2SO_4 and 3% KNO_3. It is quite true that successful seedling establishment generally depends on the emergence force, acquired through nitrogenous compounds, which facilitates embryo development. Further, plants, subjected to saline environment, absorb higher amounts of Na^+, whereas the uptake of K^+ and Ca^{2+} is significantly reduced. Reasonable amounts of both K^+ and Ca^{2+} are required to maintain the integrity and full functioning of cell membranes in plants. High K^+/Na^+ ratio is also an important criterion for seedling growth under saline conditions (Wenxue *et al.*, 2003). On the other hand, the cause of less shoot growth in case of priming with higher salt concentrations might be the osmotic effect (Chang-Zheng *et al.*, 2002; Ashraf and Rauf, 2001).

Shoot and root dry matter. Results in Table 1 indicate that maximum shoot dry matter (0.79 g/plant) was recorded when NaCl stress was not applied. Seeds primed with 2% K_2SO_4 showed maximum shoot dry matter yield (0.45 g/plant). Among all the treatments, priming with water under NaCl salt stress produced the lowest dry matter, i.e. 0.25 g/plant. This was presumably due to salt stress which inhibited seedling growth. Similar results have also been reported by Ashraf *et al.* (1987) and Ayres (1952) who observed that a single salt solution of NaCl caused significant reduction in plant growth.

Table 2. Effect of different halopriming treatments on ionic concentration (%) in sunflower tissues (average of four repeats)

Treatments	Na^+ (%)	K^+ (%)	Ca^{+2} (%)	Mg^{+2} (%)
Unprimed control (grown in distilled water)	0.28 (±0.003)	0.79 (±0.02)	0.23 (±0.001)	0.18 (±0.006)
Unprimed control (grown in 200 mM NaCl)	0.35 (±0.13)	1.05 (±0.14)	0.18 (±0.006)	0.28 (±0.07)
1% KNO_3 primed (grown in 200 mM NaCl)	0.30 (±0.004)	1.11 (±0.07)	0.30 (±0.06)	0.24 (±0.01)
2% KNO_3 primed (grown in 200 mM NaCl)	0.61 (±0.52)	1.32 (±0.28)	0.29 (±0.05)	0.28 (±0.04)
3% KNO_3 primed (grown in 200 mM NaCl)	0.27 (±0.01)	1.08 (±0.04)	0.35 (±0.11)	0.22 (±0.007)
4% KNO_3 primed (grown in 200 mM NaCl)	0.29 (±0.10)	1.24 (±0.05)	0.21 (±0.01)	0.23 (±0.07)
5% KNO_3 primed (grown in 200 mM NaCl)	0.28 (±0.03)	1.29 (±0.06)	0.22 (±0.001)	0.21 (±0.009)
1% K_2SO_4 primed (grown in 200 mM NaCl)	0.32 (±0.01)	1.25 (±0.09)	0.36 (±0.13)	0.22 (±0.09)
2% K_2SO_4 primed (grown in 200 mM NaCl)	0.26 (±0.007)	1.34 (±0.28)	0.79 (±0.57)	0.38 (±0.22)
3% K_2SO_4 primed (grown in 200 mM NaCl)	0.28 (±0.03)	1.05 (±0.04)	0.54 (±0.40)	0.23 (±0.04)
4% K_2SO_4 primed (grown in 200 mM NaCl)	0.38 (±0.02)	1.64 (±0.16)	0.52 (±0.28)	0.29 (±0.01)
5% K_2SO_4 primed (grown in 200 mM NaCl)	0.28 (±0.003)	1.30 (±0.07)	0.29 (±0.09)	0.20 (±0.008)
Distilled water primed (grown in 200 mM NaCl)	0.33 (±0.06)	0.92 (±0.10)	0.33 (±0.12)	0.25 (±0.04)

Comparison of treatment means indicated that maximum root dry matter (0.19 g/plant) was recorded when NaCl stress was not given. Although, a significant reduction in root dry matter was observed due to salt stress, but priming with K_2SO_4 and KNO_3 significantly improved both shoot and root biomass.

Ionic concentration. Data in Table 2 indicate that concentration of Na^+ in tissues varied from 0.26% to 0.61% and was found to be non-significant. However maximum Na^+ concentration (0.61%) in plant tissues was observed with 2% KNO_3 which was reduced by about 57% due to priming with 2% solution of K_2SO_4. Comparison of the means of seeds treated with diffe-rent inorganic salts and controlled seed treatment elaborates that seeds primed with 4% K_2SO_4 showed more K^+ concentration (1.64%) in the tissues as compared to the rest of the treatments followed by 2% K_2SO_4 (1.34%) and 2% KNO_3 (1.32%). The uptake and accumulation of ions in plants is considered an important indicator of plant establishment in saline medium. However, different patterns of ion accumulation with different halopriming treatments clearly indicate that more K^+ accumulation in tissues promoted the plant to absorb other ions and hence better seedling growth.

Salinity stress significantly affected the calcium concentration in plant tissues. NaCl salinity lead to deficiency of Ca^{+2}, and resulting poor growth performance (Table 2). The highest concentration (0.79%) was observed in seeds primed with 2% K_2SO_4 followed by 3% and 4% solution of K_2SO_4. Comparatively better seedling establishment with these treatments was presumably due to more K^+ and Ca^{+2} accumulation which promoted physiological metabolism by means of launching “ion pump” and “proton pump” (Table 2), while less amount of Ca^{+2} concentration (0.18%) was found in control. This indicated that without priming, high Na^+ concentration induced Ca deficiency in plant tissues which caused significant reduction in plant growth. These results are in accordance with those of Lynch and Lauchli (1988) who reported that salinity stress disturbed intracellular Ca in root protoplast. It is a well established fact that under saline conditions, role of calcium becomes even more important wherein it controls the uptake of Na by improving selectivity as well as integrity of the cell membrane (Rehman *et al.*, 1998; Maas and Grieve, 1987; Staples and Toennisessen, 1984). Magnesium concentration in tissues varied from 0.18% to 0.38%; maximum concentration (0.38%) in shoots was recorded with 2% K_2SO_4 treatment, while minimum concentration was recorded in control without priming.

Conclusion

In the study on halopriming of sunflower seeds, hundred percent seed germination was recorded with 2% K_2SO_4 priming under 200 mM NaCl salt stress. Among all the treatments, 2% solution of K_2SO_4 showed better performance in terms of shoot and root growth as well as dry matter yield production.

References

Ayres, A.D. 1952. Seed germination as affected by soil moisture and salinity. *Agron. J.* **44:** 82-84.

Ashraf, M., McNeilly, T., Bradshaw, A.D. 1987. Selection and heritability of tolerance to sodium chloride in four forage species. *Crop Sci.* **27:** 232-234.

Ashraf, M., Rauf, H. 2001. Inducing salt tolerance in maize (*Zea mays* L.) through seed priming with chloride salts: growth and ion transport at early growth stages. *Acta Physiol. Plant* **23:** 407-414.

Chang-Zheng, H., Jin, H., Zhi-Yu, Z., Song-Lin, R., Wen-Jian, S. 2002. Effect of seed priming with mixed salt solution on germination and physiological characteristics of seedlings in sunflower under stress condition. *J. Zhejiang Univ. (Agric. Life Sci.)* **28:** 175-178.

Flower, T.J., Troke, P.F., Yeo, A.R. 1977. The mechanism of salt tolerance in halophytes. *Annu. Rev. Plant Physiol.* **28:** 89-121.

Francois, L.E. 1996. Salinity effects on four sunflower hybrids. *Agron. J.* **88:** 215-219.

GOP. 2005. *Agricultural Statistics of Pakistan*, Ministry of Food, Agriculture and Livestock, Economic Wing, Government of Pakistan, Islamabad, Pakistan.

Lynch, J., Lauchli, A. 1988. Salinity affects intracellular Ca in corn root protoplasm. *Plant Physiol.* **87:** 351-356.

Maas, E.V., Grieve, C.M. 1987. Sodium induced calcium deficiency in salt stressed corn. *Plant Cell Environ.* **10:** 559-564.

Rehman, S., Harris, P.J.C., Bourne, W.F. 1998. The effect of NaCl on the Ca, K, Na concentration of the seed coat and embryo of *Acacia tortilis* and *A. coriacea*. *Ann. Appl. Biol.* **133:** 269-279.

Rhoades, J.D. 1982. Cation exchange capacity. In: *Methods of Soil Analysis. Part 2. Chemical and Microbiological Properties,* A.L. Page, R. H. Miller and D. R. Keeney (eds.), pp. 149-159, 2nd edition, American Society of Agronomy, Madison, Wisconsin, USA.

Sackston, W.E. 1981. The sunflower crop and diseases, progress, problems and prospects. *Plant Dis.* **65:** 643-647.

Sayed, H.I. 1985. Diversity of salt tolerance in a germplasm collection of wheat (*Triticum* sp.). *Theor. Appl. Genet*. **69:** 651-657.

Singh, G., Gill, S.S., Sandhu, K.K. 1999. Improved performance of muskmelon (*Cucmis melo*) seeds with osmo conditioning. *Acta Acrobat*. **52:** 121-126.

Staples, R.C., Toennisessen, G.H. 1984. *Salinity Tolerance in Plants*: *Strategies for Crop Improvement*, John Wiley and Sons, New York, USA.

Tesar, M.B. 1984. *Physiological Basis of Crop Growth and Development*, American Society of Agronomy: Crop. Science Society of America, USA.

Wenxue, W., Bilsborrow, P.E., Hooley, P., Fincham, D.A., Lombi, E., Foster, B.P. 2003. Salinity induced differences in growth, ion distribution and partitioning in barley between the cultivar Maythorpe and its derived mutant Golden Promise. *Plant Soil* **250:** 183-191.