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Effect of NaCl Pretreatment on the Germination and Emergence of Seven Cultivars of Wheat Seeds

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Abstract

Seeds of seven different cultivars of wheat (*Triticum aestivum*) were pre-imbibed in water or in 5, 10, 20% NaCl solutions for 2 days at room temperature. The effects of these pre-treatments on the percentage seed germination, seedling emergence and spread of emergence were observed. All viable seeds germinated within 48 hours under laboratory conditions. However, in pot experiments, first seedling emergence was recorded after 150 hours (6.25 days). Seed germination tests carried out in the laboratory reflected profiles similar to that in pot experiments. Water pre-treatment had no effect on the percentage seed emergence in pot experiments but decreased the spread of seedling emergence compared with that of control. NaCl treatment at 5 percent concentration had no effect on seedling emergence in pot experiments, but less seeds germinated and emerged in 10 percent NaCl treatments compared with the 20 percent NaCl treatment or control seeds. Some cultivars were found to be less resistant to salt treatments than the others.

Introduction

When a seed is sown in the field, it is exposed to a number of environmental factors which directly or indirectly affect the seed germinability and hence the growth and ultimately the final yield and quality of the crop (Matthews and Wells, 1986). As a seed is allowed to come in contact with water, hydration of seeds is an initial step towards germination and seedling establishment (Bewley and Black, 1986). Nature and size of the seed, hydratability of seeds and temperature determine the amount of water absorbed which may not exceed 2-3 times the dry weight of the seeds. Seeds, however, may take up more water than they need to germinate which may result in loss of a wide variety of solutes through the membrane (Hegarty, 1978).

The effects of salts on the germination and growth of wheat seeds have invariably been studied. Wiebe and Jessen (1979) have shown that salt treated seeds are desiccation-sensitive which caused physiological injuries in seeds resulting in reductions in seed germinability. Dasabramanian and Sarin (1974) determined that NaCl (5mM) and CaCl₂ (75mM) solutions limited the growth of wheat plants with smaller number of ears and reduced grain weight. Similarly, Maas *et al.* (1994) found that salinity and drought reduced the development and viability of tillers in wheat. The salt effect on wheat has also been shown to change the relative nutrient contents (i.e., N, Mg, Zn, Cu, K, Mn, Fe ions) (Chachar *et al.*, 1990) and total nitrogen content (Khattak *et al.*, 1996) in plants. The effect of salts on cellular membrane integrity has also been found detrimental (Leopold and Willing, 1984) though in dry and mature seed membrane is also disorganized and incomplete and takes few minutes during imbibition to repair (Simon and Raja-Harun, 1972; Bewley and Black, 1986). Therefore, it is suggested that salt and seed desiccation treatments may have similar effects on membrane disruption (Senaratna and McKersie, 1983).

Materials and Methods

Seeds of seven different cultivars harvested in 1995 were obtained from Regional Institute of Agricultural Research, Bahawalpur. Following cultivars were selected for experiments, i.e., Inqalab, Pasban, v.7222, v.7398, v.88163, v.87189, v.7061.

Seed Pretreatments: Following seed pretreatments were employed at 16°C.

1. Water Imbibition: Seeds were allowed to imbibe for 48 hours, surface and air dried for a week and used.
2. NaCl treatment: Seeds were imbibed in 5, 10, 20 percent NaCl solutions for 2 days, surface and air dried for a week and used.

Seed Germination: Wheat seeds, treated or untreated, were allowed to germinate in glass petri dishes containing double filter papers soaked with distilled water. The petri dishes were placed at 16°C and germination was checked until all the viable seeds had germinated. Protrusion of radicle was considered as an indication of the completion of 'germination'.

Seed Emergence: Seeds were placed in garden pots (8 x 9 inches) filled with soil rich in organic manure (*Vattar* soil) at a depth of 1-2 cm. Seedling emergence was noticed daily until seedling emerged out of the soil. Experiments were carried out in duplicate and data was recorded and analyzed.

Measurement of Electrical Conductance: 15 Wheat seeds were allowed to imbibe in 10ml double distilled water. Electrical conductance (E.C.) was measured at 16°C with conductivity meter (Milwaukee-CON 1000) within one minute of start of imbibition and named the value, zero hour. Then after regular intervals E.C. was measured and expressed as $\mu\text{S}/15$ seeds.

Table 1: Percentage seed germination after 8 days or when all the viable seeds germinated at 16°C. Seeds were pretreated for two days. Results are mean of three independent experiments with S.E. < 10%. ; N.D. is not determined.

| Treatment | Inqalab | Pasban | v.7222 | v.7398 | v.88163 | v.87189 | v.7061 |
|-----------|---------|--------|--------|--------|---------|---------|--------|
| Control | 100 | 100 | 100 | 100 | 100 | 100 | 100 |
| Water | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. | N.D. |
| 5%NaCl | 25 | 35 | 55 | 35 | 40 | 25 | 50 |
| 10%NaCl | 35 | 30 | 40 | 30 | 25 | 40 | 80 |
| 20%NaCl | 35 | 60 | 75 | 60 | 30 | 75 | 80 |

Results and Discussion

Germination profiles of pretreated seeds: Wheat untreated control seeds were germinated at 16°C, the results show 100 percent germination for all the seed cultivars within 2 days (Table 1). Seeds pre-treated with varying concentrations of NaCl solutions for 2 days exhibited 20-75 percent reductions in the total number of seeds germinated. These results suggest that soaking in 5-20 percent NaCl solutions for 2 days has major effect on seed germination performance.

The effect of salt solutions on the germination performance of seedlings is well documented (Wiebe and Tiessen, 1979; Brocklehurst and Dearman, 1984; Haigh and Barlow, 1987a,b). The soaking of seeds for two days was probably the more period than should have been allocated to seeds (for example, 8 hours or 24 hours), since it is known that DNA replication starts after 12 hours of imbibition in wheat seeds which is very dependent on early protein synthesis during first few hours of imbibition (Osborne, 1983; Bewley and Black, 1986). It is therefore suggested that salt ions might have had damaging effect on the machinery involved in seed germination (Brocklehurst and Dearman, 1984; Bewley and Black, 1986). Further, our unpublished data and E.C. values given in Figure 2 show that imbibitional injury in NaCl treated seeds is higher when such seeds are re-soaked in water.

Percent seed emergence: When these pretreated seeds were placed in garden pots and seedling emergence recorded, 100 percent emergence was recorded in untreated controls (Table 2). The first seed emerged after 150 hours. However, water treated seeds exhibited 20-60 percent reductions in emergence for all the cultivars; minimum seedling emergence was observed in v.7061 (40% emergence). The spread of emergence was less in water treated seeds than the control ones and in the two seed lots (v.7061 and v.87189) all the viable seeds emerged (40 and 50%, respectively) on the same time (Table 2).

In NaCl treated seeds, further reductions in seedling emergence have been seen (Table 2). Seeds of all the cultivars soaked in 5 percent NaCl solution exhibited 100 percent emergence except Inqalab and Pasban wherein

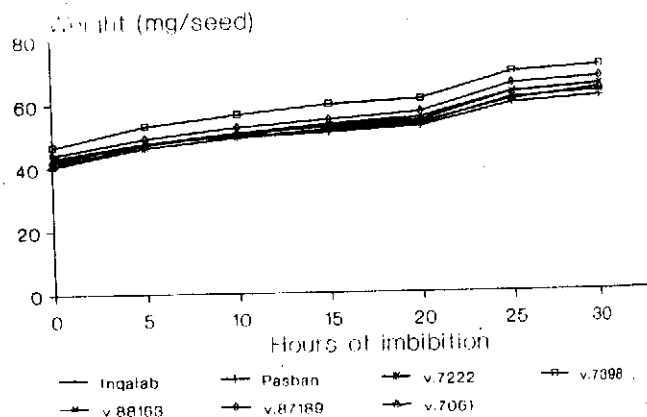


Fig. 1: Fresh weight changes during imbibition of different cultivars of wheat

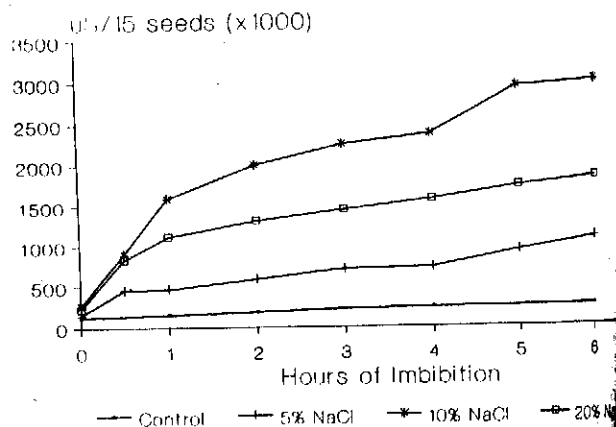


Fig. 2: E.C. measurements during imbibition of NaCl-treated 'V.87189' cultivar.

seedling emergence was 30 and 60 percent, respectively. Among various concentrations of NaCl, minimum emergence was noticed in 10 percent NaCl treated seeds though the spread of emergence was less in it than 5 or 20 percent NaCl treated seeds. It means, NaCl at 5 or 20 percent have similar effects whilst 10 percent concentration more effective in getting minimal seedling emergence.

Table 2: Percentage seed emergence and spread of emergence in pots.

| Cultivar | Treatment | Germination (%) | Spread of germination (Hours) |
|----------|-----------|-----------------|-------------------------------|
| Malab | Control | 100 | 175-293 (118) |
| | Water | 80 | 223-265 (42) |
| | 5%NaCl | 30 | 150-199 (49) |
| | 10%NaCl | 50 | 199-265 (66) |
| | 20%NaCl | 20 | 199-293 (94) |
| Saban | Control | 100 | 216-315 (99) |
| | Water | 70 | 199-293 (94) |
| | 5%NaCl | 60 | 150-216 (66) |
| | 10%NaCl | 60 | 199-293 (94) |
| | 20%NaCl | 40 | 265-293 (28) |
| 7222 | Control | 100 | 175-315 (140) |
| | Water | 80 | 199-293 (94) |
| | 5%NaCl | 100 | 150-265 (115) |
| | 10%NaCl | 60 | 150-223 (73) |
| | 20%NaCl | 60 | 199-265 (99) |
| 398 | Control | 100 | 175-265 (90) |
| | Water | 60 | 265-315 (50) |
| | 5%NaCl | 100 | 150-265 (115) |
| | 10%NaCl | 30 | 150 199 (49) |
| | 20%NaCl | 80 | 193-315 (122) |
| 8163 | Control | 100 | 168-216 (48) |
| | Water | 70 | 265-293 (28) |
| | 5%NaCl | 100 | 150-193 (43) |
| | 10%NaCl | 30 | 199-265 (66) |
| | 20%NaCl | 80 | 193-293 (100) |
| 7189 | Control | 100 | 150-293 (143) |
| | Water | 50 | 265 (1) |
| | 5%NaCl | 100 | 150-199 (49) |
| | 10%NaCl | 40 | 265 (1) |
| | 20%NaCl | 80 | 175-315 (140) |
| 061 | Control | 100 | 150-193 (43) |
| | Water | 40 | 265 (1) |
| | 5%NaCl | 100 | 150-265 (115) |
| | 10%NaCl | 40 | 223 (1) |
| | 20%NaCl | 80 | 175-315 (140) |

Results are mean of three independent experiments. S.E. 0%. Numbers in () indicate the total number of hours during which time all the viable seeds emerged.

These findings are explainable on the basis that complex interactions are seen when seeds are placed in soil (Bewley and Black, 1986; Chanway, 1997). The fate of each seed is different from its neighbour but the overall pattern shows that seeds during two days of pretreatment had lost a major portion of organic molecules in the leachate during which time mobilization of starch had already been started which was then diminished when seeds were dried after the pretreatment (Bewley and Black, 1986). However, on germinating seeds at the threshold programmed to germination

were emerged but those seeds committed suicide in which the processes of radicle emergence were not operational. Moreover, it is also documented that seed rhizosphere helps in seedling emergence in soil (emergence-promoting rhizobacteria, EPR; plant growth promoting rhizobacteria, PGPR) but since the majority of seeds have lost its inorganic and organic matter in the pretreatment, the leachate of it in the soil is not sufficient enough to support the growth of rhizobacteria (Chanway, 1997).

Changes in electrolytes conductance : Figure 1 shows the changes in electrolytes leakage after the seed pretreatment in selected representative cultivar. Higher E.C. values have been seen in salt treated seeds than in the control (E.C. of 2-days water imbibed seeds was not recorded). E.C. of 10 percent NaCl treated seeds is higher than 20 percent (or 5%) NaCl treatment which indicates that damage done to seeds is greater in 10 percent treated seeds than that of 20 percent NaCl treated seeds. This pattern has already been depicted in the germination tests wherein 10 percent salt treated seeds performed poorly than 20 percent (or 5%) salt treated seeds (Tables 1-2). It is suggested that the intake of Na⁺ may be greater in the seeds in 10 percent concentrations than in 5 percent (lower rates of Na⁺ intake) or 20 percent (inhibitory concentration of Na⁺) solutions which may have caused disturbances in the cellular osmotic balance (Brocklehurst and Dearman, 1984). It resulted more damage to plasma membrane and hence increased leakage. It was later found that increased conductance was not due to Na and Cl ions in the leachate, which one might think off, but actually the UV absorbable material was coming out of the seed and less than 5 ppm Na ions were found in the leachate, as determined by flame photometry (unpublished data). In summary, the low germinability and emergence of seeds is attributed to higher efflux of inorganic and organic ions through due to the salt damage occurred to membranes and conductivity measurements of such seeds look a good indicator of germination performance of seeds (Coolbear *et al.*, 1984).

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