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Impact of NaCl salinity on the germination of *Acacia mellifera*, *Aloe vera*, *Amaranthus graecizans* and *Lawsonia inermis* plant seeds growing in Saudi Arabia

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ABSTRACT

The present investigation determined the salinity effect on seed germination of Saudi Arabian plant species namely *Acacia mellifera*, *Aloe vera*, *Amaranthus graecizans* and *Lawsonia inermis*. The response of seeds to different NaCl concentrations varied and the delay in radicle growth was proportional to salt concentration. *Acacia mellifera* seeds gave their maximum radicle growth after 10 days and the reduction in radicle growth was 92.7% compared to control. The growth of radicle continued for 13 days to record the maximum value in the case of *Aloe vera*. The delay in radicle growth was lower than that of *Acacia mellifera.* The radicle growth indicated that the *Aloe vera* was slightly more tolerant to salinity stress, since the radicle growth recorded at 200% NaCl was reduced by 87.2%. On the other hand, *Amaranthus graecizans* showed a reduction of 91% in the radicle length at 100% NaCl compared to the control. *Lawsonia inermis* showed a reduction of 76% in radicle length compared to the control and about 10% lower than that of *Aloe*. *Lawsonia inermis* showed the highest germination percentage followed by *Aloe vera*, *Amaranthus graecizans* then *Acacia mellifera* in a descending order. The correlation coefficient calculation between NaCl concentration and percentage germination showed that the strongest relation was found in *Lawsonia inermis* followed by *Aloe vera*, *Acacia mellifera* the *Amaranthus graecizans*.

Key words: *Acacia mellifera***,** *Aloe vera***,** *Amaranthus graecizans* and *Lawsonia inermis,* Salinity**,** seed germination, salt tolerance.

INTRODUCTION

A plant species regeneration niche is shaped by factors influencing seed production, dispersal, germination, seedling establishment, and growth (Anderson and Winterton, 1996). One of the most critical stages in the life cycles of plants is the period of germination (Al-Shoaibi and Al-Sobhi, 2004). A reduction in the surface soil salinity may be a pre-requisite for successful germination and establishment. An important attribute of the surviving plant is the ability of their seeds to withstand long periods at high salinities and then germinate when conditions ameliorate. Increased salinity leads to a reduction and/or delay in germination of halophyte seeds (Katembe *et al.,* 1998).

Saline soils are very often low in available nitrogen with lower rates of nitrogen mineralization (McClung and Frankenberger, 1987). Satisfactory growth on saline soils will require adequate salt tolerance and an adequate supply of nitrogen as well as other nutrients and/or the ability to fix sufficient atmospheric nitrogen for metabolism and growth.

Worldwide soil salinity is posing a major problem, especially in the arid and semiarid regions which have low rainfall and high evaporation rates (Larcher, 1995; Al-Zahrani, 2002; Al-zahrani, *et al.,* 2002). In the arid environments, such as that of Saudi Arabia, the salt poses an extreme stress on plant growth, which necessitates the emergence of plants salt-tolerance in order to be compromised with the existing environmental conditions (Al-Shoaibi and Al-Sobhi, 2004).

The present work investigated the effect of salinity on the germination of the seeds and radicle growth of four Saudi Arabian plants (*Acacia mellifera*, *Aloe vera*, *Amaranthus graecizans* and *lawsonia inermis*) collected from Al-Taef region.

MATERIAL AND METHODS

Plant Materials

Seeds of *Acacia mellifera*, *Aloe vera*, *Amaranthus graecizans* and *Lawsonia inermis* were collected from Al-Taef region. Seeds were initially stored at 5°C until they were used.

Germination conditions

Seeds were germinated on two layers of filter paper in seedling trays and irrigated with distilled water for 3 days until they were large enough to handle. After germination, the seedlings were then transplanted into 20 cm diameter free-draining pots containing approximately 2kg of steam-sterilized, washed, coarse sand. Pots were watered manually (for 5 days) with a basal nutrient solution containing (in mole/m^3): $CaCl_2$, 0.25; KCl, 0.15; K_2HPO_4, 0.06; KH_2PO_4, 0.06; $MgSO_4$, 0.25; FeEDTA, 0.12; and (in mol/m^6): H_3BO_3, 11.5; $MnSO_4$, 0.9; $ZnSO_4$, 0.2; $CuSO_4$, 0.07; and H_2MoO_4, 0.3. The experiments were carried out in a glasshouse with 12 hr photoperiod and a relative humidity of 70% (7). Pots were maintained at day/night temperatures of approximately 20/25°C. Plants were flushed manually with nutrient solution every day.

Salinity effect

Application of salt solutions started 5 days after seedling transplantation. Six NaCl concentrations (%) 25, 50, 75, 100, 150, 200 were made up in the nutrient solution described above. Control plants were given only the nutrient solution. All pots were watered to excess with approximately 250ml treatment solution per day. Seedlings were gradually exposed to NaCl at the rate of 50mM per day in order to minimize any shock (Al-Shoaibi and Al-Sobhi, 2004). To avoid stressing of seedlings by the high salinity levels, the solutions were aerated continuously. Pots were flushed weekly with non-saline nutrient solution to counteract any increase in salt concentrations, then immediately watered with the designed treatment solution. A seed was considered to have germinated when the radicle emerged. Germinated seeds were discarded

Table 1: Effect of different NaCl concentrations on the average daily germination of *Acacia mellifera* seeds assessed as root length (cm).

Time	The average germination value (±SD) at different NaCl concentrations (%)						
(days)	Control	25	50	75	100	150	200
1	0.48±0.0	0.3± 0.0	0	0	0	0	0
2	2.3 ±0.1	1.0±0.0	0	0	0	0	0
3	2.7 ±0.2	1.3±0.0	2.7±0.3	0	0	0	0
4	3.7 ±0.1	1.7±0.2	2.7±0.1	0	0	0	0
5	4.2 ±0.2	2.3±0.0	2.7±0.2	2.7±0.1	0	0	0
6	6.7 ±0.3	3.5±0.2	3.0±0.1	3.0±0.2	0	0	0
7	8.5 ±0.2	4.6±0.3	3.0±0.1	3.0±0.3	1.7±0.2	0	0
8	10.7 ±0.4	5.7±1.1	3.0±0.1	3.0±0.2	1.7±0.3	0.7±0.3	0
9	12.3 ±0.5	6.0±1.5	3.3±0.2	3.0±0.2	1.7±0.4	1.3±0.4	0
10	13.7 ±1.7	7.7±1.6	3.4±0.2	3.0±0.3	1.7±0.0	1.7±0.5	1.33±0.1
11	13.7 ±1.8	7.7±2.3	3.5±0.3	3.0±0.4	1.7±0.3	1.7±0.3	1.33±0.1
12	13.7 ±1.9	7.7±3.4	3.7±0.5	3.0±0.1	1.7±0.2	1.7±0.3	1.33±0.2
13	13.7 ±2.0	7.7±1.3	3.7±0.5	3.0±0.0	1.7±0.3	1.7±0.2	1.33±0.4
14	13.7 ±2.3	7.7±2.2	3.7±0.6	3.0±0.1	1.7±0.4	1.7±0.3	1.33±0.3
15	13.7 ±2.3	7.7±0.0	3.7±0.3	3.0±0.3	1.7±0.5	1.7±0.4	1.33±0.3

Table 2: Effect of different NaCl concentrations on the average daily germination of *Aloe vera* seeds assessed as root length (cm)

Time (days)	The average germination value (±SD) at different NaCl concentrations (%) Control	25	50	75	100	150	200
1	1.4±0.2	1.00± 0.0	0	0	0	0	0
2	3.3 ±0.2	1.5±0.0	1.0±0.2	0	0	0	0
3	3.7 ±0.1	1.663±0.0	1.1±0.3	0.8±0.1	0	0	0
4	4.2 ±0.2	2.7±0.2	1.7±0.1	1.0±0.2	0	0	0
5	4.5 ±0.2	3.2±0.0	2.7±0.2	2.2±0.1	0.7±0.1	0	0
6	6.1 ±0.1	3.8±0.2	3.1±0.1	2.8±0.2	1.0±0.4	0.5±0.1	0
7	8.0 ±0.3	5.6±0.3	4.0±0.1	3.0±0.3	1.7±0.2	0.9±0.4	0
8	10.8 ±0.4	5.9±1.1	4.2±0.1	3.3±0.2	2.3±0.3	1.7±0.3	0.7±0.1
9	12.0 ±0.6	6.0±1.5	5.30±0.2	5.0±0.2	3.8±0.2	2.9±0.4	0.9±0.4
10	14.7 ±1.7	7.7±1.6	5.34±0.2	5.4±0.3	3.8±0.0	2.9±0.5	1.6±0.1
11	16.7 ±1.8	8.7±2.3	6.00±0.3	5.4±0.4	3.8±0.3	2.9±0.3	1.8±0.1
12	17.7 ±1.9	10.7±3.4	8.00±0.5	5.4±0.1	3.8±0.2	2.9±0.3	2.3±0.2
13	18.1 ±2.0	11.4±1.3	8.00±0.4	5.4±0.0	3.8±0.3	2.9±0.2	2.3±0.4
14	18.1 ±2.3	11.4±2.2	8.00±0.3	5.4±0.1	3.8±0.4	2.9±0.3	2.3±0.3
15	18.1 ±2.3	11.4±0.0	8.00±0.4	5.4±0.3	3.8±0.5	2.9±0.4	2.3±0.3

Table 3: Effect of different NaCl concentrations on the average daily germination of *Amaranthus graecizans* seeds assessed as root length (cm)

Time(days) (days)	The average germination value (±SD) at different NaCl concentrations (%) Control	25	50	75	100	150	200
1	0	0	0	0	0	0	0
2	7.7±0.7	0	0	0	0	0	0
3	9.7 ±0.0	0	0	0	0	0	0
4	10.0 ±0.4	0.7±0.2	0.7±0.2	0	0	0	0
5	10.5 ±0.2	2.2±0.1	2.6±0.3	1.3±0.1	0	0	0
6	10.7 ±0.3	4.3±0.8	2.8±0.4	1.3±0.3	1.0±0.3	0	0
7	11.7 ±2.1	5.0±1.1	3.2±0.3	1.3±0.3	1.0±0.1	0	0
8	11.7 ±0.3	6.0±1.8	4.2±0.0	1.3±0.4	1.0±0.1	0	0
9	11.7 ±0.3	6.0±1.5	4.7±0.3	1.3±0.2	1.0±0.1	0	0
10	11.7 ±1.3	7.0±3.2	5.0±0.3	1.3±0.4	1.0±0.1	0	0
11	11.7 ±1.8	7.8±2.1	5.0±0.7	1.3±0.3	1.0±0.1	0	0
12	11.7 ±1.9	8.4±3.0	5.0±0.9	1.3±0.2	1.0±0.0	0	0
13	11.1 ±2.9	8.4±2.0	5.0±0.4	1.3±0.2	1.0±0.2	0	0
14	11.1 ±2.0	8.4±2.1	5.0±0.1	1.3±0.1	1.0±0.1	0	0
15	11.1 ±2.1	8.4±1.3	5.0±1.4	1.3±0.3	1.0±0.1	0	0

immediately and counts were made daily until no seeds had germinated over seven successive days. After the harvest, the radicle measurement was taken daily (in cm) and the germinated seeds were discarded and the final counts were scored and the germination percentage was then calculated.

Statistical analyses

The data obtained was the average of three readings. Statistical analyses were performed using Microsoft Excel program for the calculation of standard error and correlation coefficient (r).

RESULTS AND DISCUSSION

Salinity has a considerable effect on world agriculture, significantly reducing productivity of agricultural plants. Soil salinity is an increasing threat for agriculture and is a major factor in reducing plant productivity; therefore, it is necessary to obtain salinity-tolerant varieties (Hasegawa et al., 2000; Rocak and Linder, 2004). In saline environments, adaptation of plants to salinity during germination and early seedling stages is crucial for the establishment of species (Ungar, 1995; Tobe, and Omasa, 2000). Therefore, appropriate germination responses of halophytic species to environmental parameters determine their distribution in saline environment. Thus for the successful establishment of plants in saline environments, seeds must remain viable at high salinity and germinate when salinity decreases Tuteja, (2005). Recently, the implementation of biotechnology strategies to improve salinity tolerance identified salt tolerance effectors and the regulatory components that control them during the stress. Tuteja, (2005) has provided a new insight into plant tolerance to salinity by identifying a DEAD-box helicase, PDH45, as a novel genetic determinant of salt tolerance and yield stability. The *PDH45* transcript was also upregulated by other abiotic stresses, which suggests that the increase could be due to water stress resulting from salinity- and mannitol-induced desiccation. Induction of transcript was stimulated by the phytohormone, ABA, which modulates gene activation and repression under multiple environmental stress conditions, such as drought, cold, and salinity.

The seeds' germination of the selected plant species *(Acacia mellifera*, *Aloe vera*, *Amaranthus graecizans* and *Lawsonia inermis)* was assessed under the NaCl salinity stress using a concentration range of NaCl (25-200%). The variations in tolerance to different NaCl concentrations among the four plants were recorded.

The profile of salt tolerance of *Acacia mellifera* (Table 1) revealed that seeds gave their maximum radicle growth after 10 days and the

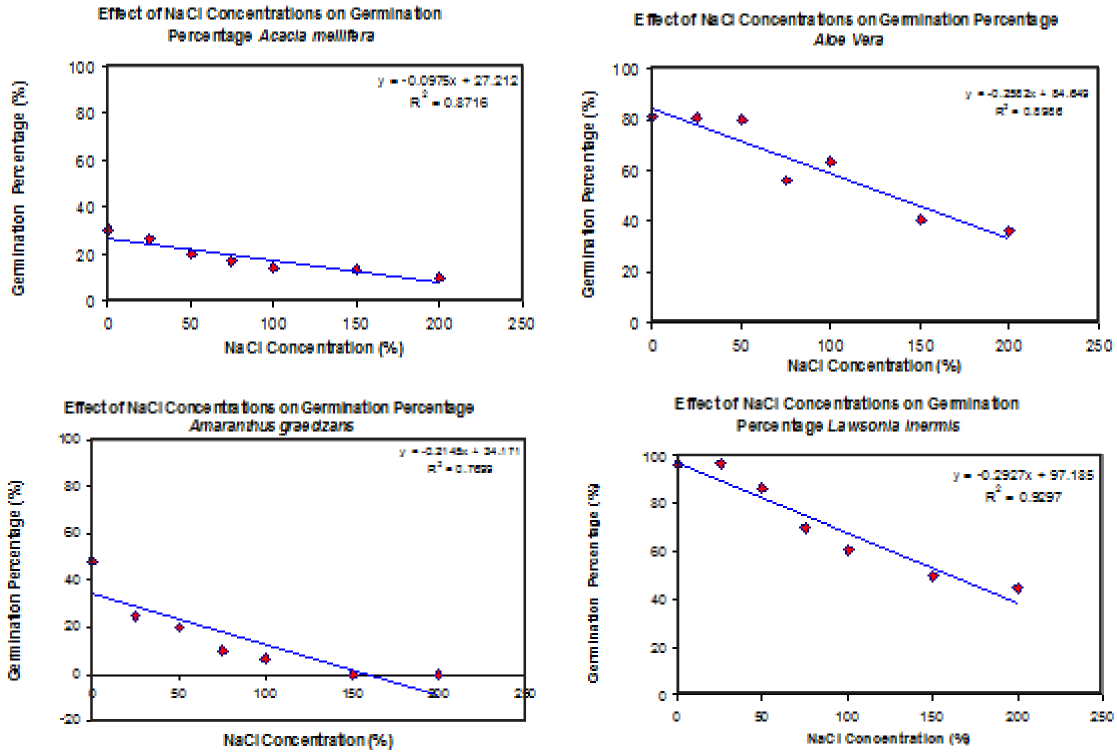

Fig. 1: Relationship between NaCl concentrations and germination percentage.

response to different NaCl concentrations varied. Fifty percent radicle growth was achieved at 25% NaCl concentration in comparison to control group. The delay in radicle growth was proportional with salt concentration. The reduction in radicle growth was 92.7% compared to that of the control.

For *Aloe vera* (Table 2), the growth of radicle continued for 13 days to reach the maximum value. The delay in radicle growth was lower than that of *Acacia mellifera*. The radicle growth indicated that the *Aloe vera* is slightly more tolerant to salinity stress, since the radicle growth recorded at 200% NaCl was reduced by 87.2%.

On the other hand, *Amaranthus graecizans* (Table 3) showed 100% reduction in the radicle length at 150 and 200% NaCl over the 20 days. The reduction in the radicle length at 100% NaCl was amounted to 91.4 compared to the control.

Although *Lawsonia inermis* (Table 4) continued to show radicle length increase up to 13 days which was similar to that of *Aloe*, the reduction in radicle length for *Lawsonia inermis* was 76.5% compared to that of the control and about 10% lower than that of *Aloe*. The delay in radicle growth at 200% NaCl was for 5 and 7 days for *Lawsonia* and *Aloe,* respectively.

Table 4: Effect of different NaCl concentrations on the average daily germination of *Lawsonia inermis* seeds assessed as root length (cm)

Time	The average germination value (±SD) at different NaCl concentrations (%)						
(days)	Control	25	50	75	100	150	200
1	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0
3	0.3 ±0.0	0	0	0	0	0	0
4	1.0 ±0.1	0.7±0.2	0.7±0.2	0	0	0	0
5	1.5 ±0.1	2.2±0.1	2.3±0.5	1.2±0.2	0	0	0
6	5.7 ±0.3	4.8±0.8	3.8±0.7	2.0±0.3	1.7±0.3	0.5±0.1	0.3±0.0
7	6.7 ±2.1	5.6±1.3	4.2±0.4	3.3±0.4	2.1±0.9	0.9±0.3	0.5±0.1
8	6.7 ±0.3	6.2±1.7	5.2±0.0	4.3±0.1	3.2±0.7	1.7±0.5	0.7±0.1
9	7.0 ±0.3	6.0±1.9	5.7±0.3	5.1±0.3	3.8±0.2	2.7±0.6	0.9±0.4
10	7.7 ±1.3	7.0±3.6	6.0±0.4	5.4±0.6	4.8±0.0	2.9±0.0	1.2±0.1
11	10.7 ±1.8	8.0±2.0	6.4±0.7	5.9±0.4	4.8±0.3	3.9±0.4	1.8±0.1
12	10.7 ±1.9	9.7±3.7	7.0±0.9	6.4±0.1	4.8±0.2	3.9±0.2	2.6±0.2
13	11.1 ±2.9	9.4±2.3	7.0±0.4	6.4±0.0	4.8±0.3	3.9±0.1	2.6±0.4
14	11.1 ±2.0	9.4±2.0	7.0±0.1	6.4±1.1	4.8±0.4	3.9±0.2	2.6±0.3
15	11.1 ±2.1	9.4±0.3	7.0±1.4	6.4±1.3	4.8±0.5	3.9±0.1	2.6±0.3

Table 5: Germination percentage for some selected plant seeds

NaCl conc. (mM)	Seeds germination (%) for the following plants			
	Acacia mellifera	***Aloe vera***	***Amaranthus gracizans***	***Lanwsonia inermis***
Control	30.3	81.2	48.5	96.3
25	26.7	80.6	25.0	96.5
50	20.1	80.0	20.0	86.4
75	17.2	56.1	10.0	70.0
100	14.3	63.2	7.0	60.5
150	13.3	40.7	0	50.0
200	10.1	35.8	0	45.0

Regarding the germination percentage (Table 5), *Lawsonia inermi*s showed the highest germination percentage followed by *Aloe vera*, *Amaranthus graecizans* and *Acacia mellifera* in a descending order. The correlation coefficient calculation showed that the strongest relation was found between *Lawsonia inermi*s germination percentage and NaCl concentration, followed by *Aloe vera*, *Acacia mellifera* and *Amaranthus graecizans* with the strength shown in Table 6.

Table 6: Calculation of correlation coefficient (r) and the strength (%) of the relationship (r^2) between NaCl concentrations and seed germination.

Plant	Correlation coefficient (r)	Strength (%) of the relation -ship (r^2)
Acacia mellifera	-0.933	87.04
Aloe vera	-0.947	89.68
Amaranthus graecizans	0.877	76.91
Lawsonia inermis	0.964	92.92

REFERENCES

1. Anderson, L. J. and Winterton, A. J. Germination as a determinant of seedling distributions among natural substrates in *Picea enelmannii* (Pinaceae) and *Abies lasiocarpa* (Pinaceae). *Ammerican J. Botany.* **83**: 112-117 (1996).
2. Al-Shoaibi, A. and Al-Sobhi, O. A. The effect of salinity on growth of Elephant grass (*Pennisetum purpureum*). *Proc. 2nd Saudi Sci. Conf.* Part I, 141-147 (2004).
3. Katembe, W. J., Ungar, I. A. and Mitchell, J. P. Effect of salinity on germination and seedling growth of two Atriplex species (Chenopodiaceae). *Annals of Botany*. **82**: 167-175 (1998).
4. McClung, G. and Frankenberger Jr., W.T. Nitrogen mineralization rates in saline vs. salt-amended soils. *Plant and soil* **104**: 13-23 (1987).
5. Larcher, W. Physiological plant ecology. 3rd Edn. Springer Verlag, New York (1995).
6. Al-Zahrani, H. S., Effects of salinity stress on growth of *Calotropis procera* seedlings. *Bulletin of Pure and Applied Sciences.* **21**B: 109-122 (2002).
7. Al-zahrani, H. S., Al-Sobhi, O.A. and Al-Ahmadi, S. B., Effects of salinity on some physiological activities of *Calotropis procera*. *Bio-Sience Research Bulletin.* **18**: 107-120 (2002).
8. Hasegawa P.M., Bressan R.A., Zhu J-K, and Bohnert, H.J., Plant cellular and molecular responses to high salinity. *Annu. Rev. Plant Physiol. Plant Mol. Biol.* **51**, 463-99 (2000).
9. Rocak S. and Linder, P., DEAD-box proteins: the driving forces behind RNA metabolism. *Nat. Rev. Mol. Cell Biol.* **5**: 232-241 (2004).
10. Ungar, I. A., Seed germination and seed-bank ecology in halophytes. In: Kigel, J., Galili, G. (eds.). Seed development and germination. New York: Marcel Dekker. 599-628 (1995).
11. Tobe, K., Li, X. and Omasa, K., Seed germination and radicle growth of a halophyte, *Kalidium capsicum* (Chenopodiaceae). *Annals of Botany*. **85**: 391-396 (2000)
12. Tuteja, A., Unwinding after high salinity stress: Development of salinity tolerant plant without affecting yield. ISB News Report. March 2005: 1-3 (2005).