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Germination Responses of the Mediterranean Saltbush (*Atriplex halimus* L.) to NaCl Treatment

[1]A. Abbad, [2]A. El Hadrami and [1]A. Benchaabane
[1]Laboratoire d'Ecologie Végétale, Département de Biologie, Faculté des Sciences, Semlalia B.P. 2390, Université Cadi Ayyad Marrakech, Maroc
[2]Department of Plant Science, 222 Agriculture Building, University of Manitoba Winnipeg, MB R3T 2N2, Canada

Abstract: The effect of salinity on the germination of the seeds before and after exposure to different NaCl concentrations were studied using seven *Atriplex halimus* provenances. The study showed a highly significant effect of salinity, provenance and their interaction on the percentage of germination. Seed germination was reduced in all provenances with an increase in salinity. Maximum germination levels were observed in tests using distilled water. SBO provenance showed a lesser reduction in germination in response to salt stress than other provenances. IDE provenance (Saharan provenance) showed low germination at 0% NaCl concentration and failed to germinate in 2% NaCl. Germination in distilled water indicated significantly different provenance responses. For TAO, IMS and SAL provenances, inhibition of germination was reversible, since the total germination after recovery of seeds exposed to three concentration of NaCl (1, 1.5 and 2%) did not differ significantly ($P<0.05$) from the distilled water control, whereas the inhibition of germination for NAD, OUD and IDE provenances are not reversible.

Key words: *Atriplex halimus*, provenances, germination, salt stress, recovery germination

INTRODUCTION

Germination is a critical stage in the development and life cycle of many desert plants. It ensures the reproduction and consequently control the dynamics of the population[1]. Several uncontrolled factors may influence germination percentages in arid natural environments, particularly the presence of salt in soil[2].

Atriplex halimus (Chenopodiaceae) is a xerohalophyte, perennial, native in arid and semi arid Mediterranean regions. Owing to its environmental (reducing erosion), energy (firewood) and fodder qualities, *Atriplex halimus* is a very desirable species in several rehabilitation programs in degraded lands where excessive salinity and low moisture level are the main factors limiting plant growth; and where there is also a need to provide animals with forage, especially during drought periods[3,4]. Many authors have suggested that seeds of most halophytes, especially the genus *Atriplex*, are very sensitive to elevated salinity during germination and early seedling establishment phases[5,6]. However, *A. halimus* is characterized by broad important polymorphism, mainly dependent on its very wide ecological amplitude[7,8]. There is little information on the germination behaviour of the species under salt stress in relation to this polymorphism. The selection of provenances (ecotypes) tolerant to salinity is therefore of interest in the reclamation and rehabilitation programs of arid regions using this species.

This study was carried out to obtain information on the effect of salinity (NaCl) on the germination before and after exposure to salt stress using seven provenances of *Atriplex halimus*.

MATERIALS AND METHODS

In November 2002 (period of mature fruits), fruits were collected from seven natural populations of *A. halimus* located in different environments (Table 1). Seeds were removed from the bracts by hand. They were then surface sterilized for 20 s in 3% (w/v) calcium hypochlorite and rinsed three times with deionised water. Seeds (4 replicates of 25 seeds) were placed to germinate in Petri dishes on filter paper soaked with saline water (5 ml) of different NaCl concentrations (0, 0.5, 1, 1.5 and 2%). Petri dishes were sealed with parafilm to avoid loss of water

Corresponding Author: Abbad Abdelaziz, Département de Biologie, Faculté des Sciences, Semlalia B.P. 2390, Laboratoire d'Ecologie Végétale, Université Cadi Ayyad Marrakech, Maroc Tel: +212 44 43 46 49 (poste 535) Fax: +212 44 43 74 12 E-mail: abbad@ucam.ac.ma

Table 1: Geographical situation and climatic characteristics of the stations of the studied provenances

	Abbreviation	Geographic situation	Longitude West	Latitude North	Altitude (m)	Rainfall (mm)
Oudaya	OUD	20 km W of Marrakech	8° 02'	31° 37'	470	242
Idelssen	IDE	27 km NE of Ouarzazate	6° 32'	31° 04'	1225	115
Sidi Bouzid	SBO	10 km N of Safi	9° 15'	32° 18'	s15	327
Nador	NAD	airport	2° 56'	35° 11'	3	370
Salouane	SAL	30 km S of Nador	2° 56'	35° 11'	17	370
Taourirt	TAO	120 km SW of Oujda	2° 54'	34° 25'	390	206
Imssouane	IMS	90 km N of Agadir	9° 40'	31° 00'	75	313

and incubated at 26±1°C in the dark[4]. Seed germination (radicle > 1 mm) was monitored at 2-d intervals for 14 d and germinated seeds were removed from dishes after each counting. To study the aptitude of seeds recovery germination after exposure to salinity for 14 d, ungerminated seeds from the NaCl treatment were removed from the Petri dishes, rinsed with deionised water and transferred into new Petri dishes on a filter paper imbibed with 5 ml of distilled water for further period for germination (14 days). The germination parameters evaluated for each provenances included : germination rate (GR) expressed as the percentage of seeds germinated after 14 days, Timson Index of germination velocity (IT) expressed as the sum of the germination values measured at 2-d intervals for 14 d divided by the number of measurement periods[9] and corrected germination rate (GC) expressed as the number of seeds germinated in a concentration of salt divided by the number of germinated seeds in distilled water (control) for 14-d[10].

The data were subject to analysis of variance (ANOVA). $P<0.05$ was used to define statistical significance[11]. If a significant difference was determined among means, a Student-Neuman-Keuls (SNK) test was used to determine significant difference between pairwise comparisons among individual treatments. Germination data was transformed (arcsine) before statistical analysis to ensure homogeneity of variance.

RESULTS

The analysis of variance showed a highly significant effect ($P<0.001$) of provenance, salinity and of their interaction (provenance x salinity) (Table 2). For all the studied provenances, germination of seeds was significantly reduced ($P<0.05$) with increasing salinity (Table 3). The highest values of germination were recorded on distilled water (control). However, the germination rates at a given salt concentration varied according to provenance. NAD, SAL and OUD provenances showed a highest germination rates at 0% NaCl (GR=89%, TI=39; GR=94%, TI=42.8; GR=89%, TI=41; Table 3). SBO provenance (littoral provenance), despite its germination rate been relatively weak in 0% NaCl (GR=76%, TI= 33.78) shows less reduction in

Table 2: Result of two-way analysis of variance (ANOVA) of provenances and salinity effect and their interaction for the germination parameters

		Mean Squares		
Source of variance	df	GR	TI	GC
Provenance	6	0.65***	970.18***	0.239***
Salt	4	8082***	10208.16***	20.39***
Provenance x Salt	24	0.185***	269.615***	0.169***

Df : degree of freedom; TI: Timson Index ; GR : Germination rate; GC : Corrected germination; (***) $P < 0.001$

Table 3: The germination parameters of different *Atriplex halimus* provenances subjected to different salinity levels. Data are means of four replicates

Provenances	Germination rates (GR%)	Timson Index (TI)	Corrected germinations (GC%)
Prov. NAD			
0 (%)	89a*	38.85a	100.00a
0.5 (%)	40b	15.28b	44.85b
1 (%)	17c	7.07c	19.42c
1.5 (%)	7cd	1.78d	7.82d
2 (%)	3d	1.07d	3.45d
Prov. IMS			
0 (%)	70a	23.85a	100.00a
0.5 (%)	37b	8.07b	32.45b
1 (%)	14c	4.57bc	21.1bc
1.5 (%)	9c	2.93c	14.25c
2 (%)	6c	1.93c	8.27c
Prov. TAO			
0 (%)	62a	30.00a	100.00a
0.5 (%)	26b	11.00b	41.34b
1 (%)	21b	8.00bc	33.27b
1.5 (%)	9c	3.21cd	14.77c
2 (%)	3c	0.64d	4.57c
Prov. SBO			
0 (%)	76a	33.78a	100.00a
0.5 (%)	59a	25.78b	71.25b
1 (%)	35b	14.07c	45.45c
1.5 (%)	23b	9.93c	30.15c
2 (%)	4c	1.5d	4.62d
Prov. SAL			
0 (%)	94a	42.8a	100.00a
0.5 (%)	74b	27.92b	79.65b
1 (%)	23c	9.00c	24.4c
1.5 (%)	11d	4.28cd	11.6d
2 (%)	5d	2.21d	5.19e
Prov. OUD			
0 (%)	89a	40.85a	100.00a
0.5 (%)	38b	15.28b	42.72b
1 (%)	25c	10.64c	27.87c
1.5 (%)	7d	2.64d	7.8d
2 (%)	5d	1.71d	5.57d
Prov. IDE			
0 (%)	35a	13.35a	100.00a
0.5 (%)	11b	3.64b	33.01b
1 (%)	13b	3.71b	43.4b
1.5 (%)	9b	2.71b	28.97b
2 (%)	0c	0.00b	0.00c

*Value followed by different letters in a column differ significantly at $P<0.05$. Values are means of 4 replicates

Table 4: Mean percents (SE) of total germination (under stress+recovery germination in distilled water) of different provenances of *Atriplex halimus* for seeds originally germinated in 1, 1.5 and 2% NaCl

Provenances	Control	Tot. 1%	Tot. 1.5%	Tot. 2%
OUD	89 (3.41)a*	68 (2.83)b	48 (2.82)c	49 (3.00)c
IDE	35 (5.97)a	21 (3.00)b	23 (1.91)b	20 (1.63)b
SBO	76 (6.32)a	53 (4.72)b	52 (2.83)b	64 (2.3)ab
NAD	89 (4.43)a	56 (5.65)b	60 (2.83)b	72 (1.63)b
SAL	94 (2.58)a	85 (7.72)a	87 (1.91)a	89 (1.00)a
TAO	62 (4.16)a	51 (5.00)a	49 (1.91)a	52 (3.65)a
IMS	70 (8.25)a	45 (2.52)a	55 (7.72)a	59 (1.91)a

*Values followed by different letters in a row differ significantly at P<0.05.

germination in response to salt stress than other provenances. In this provenance, the germination percentage was relatively high even at a concentration of 1.5% NaCl (GR=23%; TI=9.93 and GC=30.15%). IDE provenance (Saharan provenance), however, showed the lowest germination at 0% NaCl concentration. This provenance also failed to germinate in 2% NaCl. The total germination after recovery in distilled water for seeds pre-treated in 1, 1.5 and 2% NaCl, also varied between provenances. For TAO, IMS and SAL provenances, inhibition of germination was reversible, since the total germination after recovery of seeds exposed to three concentration of NaCl (1, 1.5 and 2%) did not differ significantly from the distilled water control, whereas the salt inhibition of germination in NAD, OUD and IDE provenances was not reversible (Table 4). For the SBO provenance, those seeds which were exposed to high NaCl concentration, displayed an ability to recover germination when subsequently germinated on substrate containing distilled water which did not differ significantly from control.

DISCUSSION

The *Atriplex halimus* provenances subjected to a saline treatment presented variable behaviour in their germination. This variability, which is likely to be related to the different geographical origins of the seeds and may confirm the existence of polymorphism of the species. Debez *et al.*[12] reported the same observation for two Tunisian *Atriplex halimus* provenances. The effect of geographical origin on seed germination has also been found in other species[13,14]. As with the majority of halophytes, *Atriplex halimus* shows a great sensitivity to salinity at the germination stage. The maximum germination was recorded in distilled water whereas high concentrations of salt inhibit this germination but may not kill the seed. Indeed, seeds of many halophytes are known for their tolerance to salinity during their time in the soil and may germinate when the salinity of the soil is reduced[15,16]. This property has been described by some authors as a means of resistance to salinity[5,17,18]. According to these authors, the inability of halophyte seeds to germinate under hypersaline conditions but then initiate germination when salinity decreases is a criterion of salt tolerance which distinguishes them from most glycophytes. In fact, recovery germination response has been demonstrated in several halophytes such as *Salicornia europea*[19], *Spergularia marina*[20], *Suaeda depressa*[21], *Arthrocnemum australis, Triglochin stricta, Suaeda australis* and *Juncus maritimus*[22], *Atriplex patula*[17], *Haloxylon recurvum, Suaeda fruticosa* and *Triglochin maritime*[9]. Despite this, in *Atriplex halimus*, this ability of germinate after exposure to salinity is variable according to provenance. In the case of OUD, NAD and IDE provenances for example, salinity is toxic to seed germination after exposure to salinity for 14 d, an effect also reported by Debez *et al.*[12] for scarified seeds of two Tunisian *A. halimus* provenances. The SBO provenance (littoral provenance), appear to tolerate high NaCl levels and germinate at acceptable levels when salinity is removed. This effect of high tolerance on seed germination has also been reported by Khan and Ungar[9] in *Haloxylon recurvum, Suaeda fruticosa* and *Triglochin maritime* and by Keiffer and Ungar[18] in *Salicornia europaea* and *S. calceoliformis.* The weak germination rates recorded in the IDE provenance (Saharan provenance) could be related to a reduced imbibition of seeds. In fact, the integuments of seeds of IDE provenance are thicker and harder than other provenances. Similar differences in germination have been found between two Tunisian *A. halimus* provenances originating from two different climatic environments[12]. In the present study, the germination rate of the provenance originating from a less arid climate was found to be higher than that of provenances originating from a more arid climate. Difference in seed coat impermeability have also been reported for other species such as *Atriplex repanda*[23] and *A. cordobensis*[24].

Light and temperature appear to have a stimulating effect on germination of some halophytes[9,25]. Despite this, *A. halimus* seeds seemed to germinate well in darkness and at constant temperature. This work has suggested the significant role that the geographical origin may play in the germination behaviour and of seeds before and after exposure to salinity.

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REFERENCES

1. Radosevich, S., J. Holt and C. Ghersa, 1997. Weed Ecology Implications for management, Wiley, New York.
2. Mayer, A.M. and A. Poljakoff-Mayber, 1982. The Germination of Seeds. Oxford, Pergamon Press. pp: 211.
3. Le Houerou, H.N., 1992. The role of saltbushes (*Atriplex* spp.) in arid grazing land rehabilitation in the Mediterranean Basin: a review. Agroforestry Systems, 18: 107-148.
4. Bajji, M., J.M. Kinet and S. Lutts, 2002. Osmotic and ionic effects of NaCl on germination, early seedling growth and ion content of *Atriplex halimus* (Chenopodiaceae). Canadian J. Bot., 80: 297-304.
5. Khan, M.A. and I.A. Ungar, 1984. The effect of salinity and temperature on the germination of polymorphic seeds and growth of *Atriplex triangularis* Willd. American J. Bot., 71: 481-489.
6. Khan, A.M. and I.A. Ungar, 1996. Influence of salinity and temperature on the germination of *Haloxylon recurvum*. Ann. Bot., 78: 547-551.
7. Kinet, J.M., E. Benrebiha, S. Bouzid, S. Laihacar and P. Dutuit, 1998. Biodiversity study on *Atriplex halimus* for *in vitro* and *in vivo* detection of plants resistant to harsh environmental conditions and for potential micropropagation. Cahiers agricultures, 7: 505-509.
8. Le Houerou, H.N., 2000. Utilisation of fodder trees and shrubs (trubs) in the arid and semi arid zones of western Asia and northern Africa (WANA): History and perspectives. a review. Arid Soil Research and Rehabilitation, 14: 1-37.
9. Khan, A.M. and I.A. Ungar, 1997. Effects of thermoperiod on recovery of seed germination of halophytes from saline conditions. American J. Bot., 84: 279-283.
10. Smith, S.E. and A.K. Dobrenz, 1987. Seed age and salt tolerance at germination in Alfalfa. Crop Sci., 27: 1053-1056.
11. SPSS., 1994. SPSS: SPSS 6.1 for Windows update. SPSS Inc., Chicago IL.
12. Debez, A., W. Chaibi and S. Bouzid, 2001. Effet du NaCl et de régulateurs de croissance sur la germination d'*Atriplex halimus L.* Cahiers Agricultures, 10: 135-138.
13. Vakshasya, R.K., O.P. Rajora and M.S. Rawat, 1992. Seed and seedling traits of *Dalbergia sissoo* Roxb.: Seed source variation studies among ten sources in India. Forest Ecol. Manag., 48: 265-275.
14. Gilfedder, L. and J.B. Kirkpatrick, 1994. Genecological variation in the germination, growth and morphology of four populations of a Tasmanian endangered perennial daisy *Leucochrysum albicans.* Australian J. Bot., 42: 431-440.
15. Khan, M.A. and I.A. Ungar, 1986: Life history and population dynamics of *Atriplex triangularis*. Vegetatio, 66: 17-25.
16. Ungar, I.A., 1995. Seed germination and seed-bank ecology in halophytes. In: Kigel, J. and Galili, G. (Eds.), Seed development and germination. Marcel Dekker Inc., New York., pp: 599-628
17. Ungar, I.A., 1996. Effect of salinity on seed germination, growth and ion accumulation of *Atriplex patula* (Chenopodiaceae). American J. Bot., 83: 604-607.
18. Keiffer, C.H. and I.A. Ungar, 1997. The effect of extended exposure to hypersaline conditions on the germination of five inland halophyte species. American J. Bot., 84: 104-111.
19. Ungar, I.A., 1962. Influence of salinity on seed germination in succulent halophytes. Ecology, 43: 763-764
20. Ungar, I.A., 1967. Influence of salinity and temperature on seed germination. Ohio J. Sci., 67: 120-123.
21. Ungar, I.A. and F. Capilupo, 1969. An ecological life history study of *Suaeda depressa* (Pursh) Wats. Advancing Frontiers of Plant Sci., 23: 137-158.
22. Clarke, L.D. and N.J. Hannon, 1970. The mangrove swamp and salt marsh communities of the Sydney district. III. Plant growth in relation to salinity and water logging. J. Ecol., 58: 351-369.
23. Lailhacar, D.S. and H.M. Laude, 1975. Improvement of seed germination in *Atriplex repanda* Phil. J. Range Manag., 26: 491-494.
24. Aiazza, M.T. and J.A. Arguëllo, 1992. Dormancy and germination studies on dispersal units of *Atriplex codobensis* (Gondoger and Stucker) (Chenopodiaceae). Seed Sci. Technol., 20: 401-407.
25. Khan, M.A. and D.J. Weber, 1986: Factors affecting seed germination in *Salicornia pacifica* var. *utahensis*. American J. Bot., 73: 1163-1167.