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Seed Germination of Halophytes Exposed to High Salinity and Temperature in the Seed Bank

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Abstract: The ability of hâlophytes seeds to tolerate high salinity and temperature stress were I discussed. The percentage of un-germinated seeds that recovered when they were transferred to distilled water varied significantly with variation in salinity and temperature regimes in different species. Seeds of some species failed to germinate when exposed to high salinity and temperature stress. While seeds of other halophytic species showed various levels of recovery ranging from 20% to complete recovery of germination. There are some species where recovery of germination is higher than untreated control. Higher temperature inhibited germination recovery for most of the species reported.

Key words: Seed germination, Halophytes, Salinity, Temperature, Seed bank

The rapidly growing demand for increased food, fiber, and fuel in the face of rapidly declining availability of agricultural land due to increased soil salinity makes it imperative that crop production under saline conditions be significantly increased (Lieth et at., 1999; Hamdy et at., 1999). Halophytic species are broadly distributed in salt marsh and salt desert environments throughout the world (Waisel, 1972; Ungar, 1974; Adam, 1990; Khan and Ungar , 1995) .These natural environments cover a significant fraction Of the land surface of the earth (Szabolcs, 1976). Salinization of agricultural lands is more widespread than it's commonly realized. Almost one billion acres of cropland are already too salty for I conventional crop production (Yensen, 1995), and a million acres each year are lost to production worldwide due to salt problems (Kelly et at., 1979). It has been estimated that in the western United States alone 200 000-300 000 acres annually become non-productive, because of poor irrigation practices, dumping of saline water as a waste for petroleum drilling, deicing of highways and other activities (Yensen, 1995). It has become very important to learn how to use halophytes as crops, £or bioremediation of salinized cropland, for re-vegetation of non-arable areas salinized by human activity, and for restoration of saline tropical desert areas where native vegetation has been altered or destroyed. Germination is an essential first step in the establishment of halophyte under natural conditions. It is

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Note: To whom correspondence should be addressed: Dr. Bilquees Gul, P. 0. Box 8452. University of Karachi. Karachi- 75270. Pakistan. Telephone and Fax S2-21-9243976; E-mail:gul@botany.ku.edu.pk important to understand the various seed germination strategies employed by halophytes facing a variety of environmental stresses.

Halophytes seeds are dispersed during fall in the temperate climate (Ungar, 1995) and after monsoon rains in the sub-tropical deserts (Khan and Gul, 1998). Seeds are exposed to various environmental stresses while present in the seed bank (Ungar, 1991; Baskin and Baskin, 1998). In the temperate climate seeds face a cold winter and are buried under the snow for few months. During this period salinity decreased considerably but cold temperature prevents seeds from germination. While in the subtropical environment seeds are usually exposed to high salinity and temperature stresses if present in salt marshes and only high temperature stress when present in the saline deserts (Khan and Ungar, 1999). Halophytes could only maintain the continuity of their lineage only if they could survive physicochemical stresses such as flooding, drought, salinity, extreme temperatures and their interaction while in the seed bank. Seeds of halophytes germinate better in distilled water like non-halophytes but differ in their ability to germinate at higher salinities and also remain viable for an extended period when immersed in saline water (Ungar, 1995). Species reported to have high salt tolerance at germination stage include *Salicornia bigelovii* (856 mM NaCl, Rivers and Weber, 1971), *Cressa cretica* (856 mM NaCl, Khan, 1991), *Tamarix* sp. (1000 mM NaCl, Waisel, 1958; Ungar, 1967), *Salicornia pacifica var. utahensis* (860 mM NaCl, Khan and Weber, 1986), *Suaeda torreyana*, *Salicornia rubra*, *Salsola iberica*, and *Halogeton glomeratus* (1000 mM NaCl, Khan *et al.*, 1999, 2000, 2001). Many halophyte seeds have the ability to maintain seed viability for extended period of exposure to hyper-saline conditions with other physicochemical factors and germinate when conditions are favorable (Woodell, 1985; Keiffer and Ungar, 1995, 1996; Khan and Ungar 1997, Pujol *et al.*, 2001). Seed germination of some halophytes when pretreated with salinity show the priming effect of salinity on germination, while others showed no effect of salinity and recover immediately after salinity stress is removed, and still other halophytes failed to germinate when exposed to high salinity (Ungar, 1995; Khan and Ungar, 1997; Keiffer and Ungar, 1995). Increase in temperature has negative effect on the recovery of germination of most halophytic species (Ungar, 1991; Khan and Ungar, 1996, 1997, 1998ab).

The enforced dormancy response for halophyte seeds to saline conditions is of selective advantage to plants growing in highly saline habitats because seeds could withstand high salinity stress and provide a viable seed bank for recruitment of new individuals, but seed germination would be limited to periods when the soil salinity levels were within the species tolerance limits (Ungar, 1982). However halophyte seeds differ in their ability to recover from salinity stress and germinate after being exposed to hypersaline conditions (Table 1 and 2). Halophytes from the Great Basin desert (a cool temperate area) are highly salt tolerant to salinity (Table 1). Halophytes like *Allenrolfea occidentalis* (Gul and Weber, 1999), *Kochia scoparia* (Khan *et al.*, 2001a), *Salicornia rubra* (Khan *et al.*, 2000) and *Salsola iberica* (Khan *et al.*, unpublished data) had 80% or higher recovery of germination when exposed to 1000 mM NaCl (Table 1). A substantial recovery from germination occurred at the NaCl concentrations up to 600 mM NaCl in *Halogeton glomeratus* (Khan *et al.*, 2001b), *Sarcobatus vermiculatus* (Khan *et al.*, 2002), *Suaeda moquinii* (Khan *et al.*, 2001a) and *Triglochin maritima* (Khan and Ungar, 1999). This data showed that seed Great Basin halophytes have the ability to tolerate high salinity when present in the seed bank. All the species reported here recovered substantially up to 600 mM NaCl but some could almost completely recover from the NaCl concentration of 1000 mM NaCl (Table 1).

Table 1 Percentage recovery of germination of temperate halophytes at various NaCl concentrations

Name of species	NaCl (mM)						References
	0	200	400	600	800	1000	
Kochia scoparia	0	85	88	100	100	100	Khan, Gul and Weber, 2001
Salsola iberica	1	2	22	37	60	82	Khan, Gul and Weber, 2002
Halogeton glomeratus	100	85	72	52	22	8	Khan, Gul and Weber, 2001
Allenrolfea occidentalis	0	82	83	98	98	98	Gul and Weber, 1999
Salicornia rubra	0	1	23	38	60	78	Khan, Gul and Weber, 2000
Sarcobatus vermiculatus	0	0	61	47	22	0	Khan, Gul and Weber, 2002
Suaeda moquinii	0	0	62	50	25	8	Khan, Gul and Weber, 2001
Triglochin maritima	15	36	80	65	—	—	Khan and Ungar, 1999

Recovery of germination of sub-tropical halophytes also showed some variabilities (Table 2) and they appeared to be less salt tolerant while in the seed bank when compared with temperate desert species (Table 1 and 2). *Arthrocnemum macrostachyum* showed a substantial recovery at 1000 mM NaCl (Khan and Gul, 1998) while all others recovered in up to 600 mM NaCl (Table 2). *Aeluropus lagopoides* (Gulzar and Khan, 2001), *Atriplex stocksii* (Khan, 1999), *Limonium stocksii* (Zia and Khan, unpublished data) and *Urochondra setulosa* (Gulzar *et al.*, 2001) showed about 75% recovery at 600 mM NaCl (Table 2). While *Cressa cretica* (Khan, 1999), *Haloxylon stocksii* (Khan and Ungar, 1996), *Salsola imbricata* (Khan, unpublished data), *Suaeda fruticosa* (Khan and Ungar, 1998) and *Sporobolus ioclados* (Gulzar and Khan, unpublished data) showed poor recovery responses.

Seed germination of halophytes under natural conditions is regulated by variation in soil salinity and ambient thermoperiod (Khan and Ungar, 1984; Badger and Ungar, 1989; Ungar 1995). The salt tolerance of seeds appears to be affected by thermoperiod (Morgan and Myers, 1989; Khan and Ungar, 1996). Seeds of halophytes are known to tolerate high salinity during their presence in the soil (Ungar, 1995). Halophyte seeds are known to germinate when soil salinities reduced (Khan and Ungar, 1996; Ungar, 1995). Recovery of germination responses has been demonstrated in *Salicornia europaea* (Ungar, 1962), *Spergularia marina* (Ungar, 1967), *Suaeda depressa* (Ungar and Capilupo, 1969), *Suaeda linearis* (Ungar, 1962), *Arthrocnemum australsicum*, *Triglochin striata*, *Suaeda australis*, *Juncus maritimus*, and *Casuarina glauca* (Clarke and Hannon, 1970). Boorman (1967, 1968) and Woodell (1985) also reported salt stimulation of seed germination following treatment with seawater for a number of salt marsh species. Woodell (1985) classified germination responses to salinity into three categories; Type 1 species, usually found in dunes or on the drift line, were all inhibited by half strength seawater. Recovery was relatively high, but no salt stimulation was observed in this group. Seeds of Type 2 species were strongly inhibited by half-strength seawater but had recovery germination (56% to 98%) from seawater in distilled water that was similar to the original germination percentages in the control. Type 3 species has less than 10% germination in seawater, were salt stimulated and had greater than 60% germination in distilled water recovery treatments. Keiffer and Ungar (1995) exposed seeds of 5 halophytes (*Atriplex prostrata*, *Hordeum jubatum*, *Salicornia europaea*, *Spergularia marina*, and *Suaeda calceoliformis*) to an extended period of salinity treatments and determined their recovery responses when transferred to distilled water. They used Woodell (1985) classification system and placed *Atriplex prostrata* seeds in the Type 1, *Hordeum jubatum* and *Spergularia marina* in the Type 2, and *Salicornia europaea* and *Suaeda calceoliformis* in the Type 3 category.

Table 2 Percentage recovery of germination of sub-tropical halophytes at various NaCl concentrations

Name of species	NaCl (mM)						References
	0	200	400	600	800	1000	
Aeluropus lagopoides	0	60	82	89	—	—	Gulzar and Khan, 2001
Arthrocnemum macrostachyum	0	19	83	96	98	97	Khan and Gul, 1998
Atriplex stocksii	23	38	71	75	—	—	Khan, 1999
Cressa cretica	4	76	72	28	2	0	Khan, 1999
Haloxylon stocksii	20	6	58	50	—	—	Khan and Ungar, 1996
Limonium stocksii	0	82	98	98	—	—	Zia and Khan, 2002
Salsola imbricata	0	1	17	19	14	—	Khan, 2002
Sporobolus ioclados	40	19	21	39	—	—	Gulzar and Khan, 2002
Suaeda fruticosa	70	40	0	0	—	—	Khan and Ungar, 1998
Urochondra setulosa	38	88	75	60	—	—	Gulzar, Khan and Ungar, 2001

Role of temperature in the recovery of germination was poorly reported and most studies only focused on the recovery of seed germination based on the variation in salinity (Ungar, 1962, 1967; Clarke and Hannon, 1970; Ungar and Capilupo, 1969; Boorman, 1967, 1968; Woodell 1985; Keiffer and Ungar, 1995). Khan and Ungar (1996) reported that variation in the recovery responses of *Haloxylon stocksii* seed germination with the change in thermoperiod under various NaCl salinity treatments. They reported better recovery of germination at warmer thermoperiod. A number of studies on the effect of temperature regimes on the recovery of germination have since been conducted on the various kinds of halophytes from many parts of the world (Table 3). Best seed germination of temperate desert halophytes occurred at 25—35℃ (Khan and Gul, 2002), however optimal recovery of germination of temperate desert halophytes occurred at various temperature regimes (Table 3). Optimal seed germination of *Allenrolfea occidentalis* (Gul and Weber, 1999), *Halogeton glomeratus* (Khan *et al.*, 2001b), *Sarcobatus vermiculatus* (Khan *et al.*, 2002), *Salsola iberica* (Khan *et al.*, 2002) were reported in 25—35℃ while at 10—20℃ and 15—25℃ temperature regimes seeds of *Suaeda moquinii* (Khan *et al.*, 2001a), *Salicornia rubra* (Khan *et al.*, 2000), *Kochia scoparia* (Khan *et al.*, 2001c) recovered better while *Triglochin maritima* showed a better recovery at 5—25℃ (Khan and Ungar, 1999). The *Polygonum aviculare*, a native of moist temperate region showed best recovery at colder temperature regimes (5—15℃) (Khan and Ungar, 1998).

Recovery of seed germination of subtropical halophytes does not show any pattern (Table 4). Few halophytes (*Aeluropus lagopoides* and *Limonium stocksii*) showed almost complete recovery at all temperature regimes studied (Gulzar and Khan, 2001; Zia and Khan, unpublished data). *Atriplex stocksii* and *Suaeda fruticosa* showed about 70% recovery at 20—30℃ and 15—25℃ respectively (Khan and Ungar, 1998; Khan, 1999), while most other halophytes showed a recovery response about 50% or less (*Arthrocnemum macrostachyum*, Gul and Weber, 1998; *Haloxylon stocksii*, Khan and Ungar, 1996; *Salsola imbricata*, Khan, unpublished data), while still other made little recovery at any temperature regime (*Cressa cretica*, Khan, 1999; *Sporobolus ioclados*, Gulzar and Khan, 2001; *Urochondra setulosa*, Gulzar *et al.*, 2001). It appears from the published data the recovery of germination of most subtropical halophytes are poor in comparison to temperate halophytes and they do not show any consistent pattern of recovery of germination responses with the change in temperature.

Table 3 Percent recovery of germination of temperate halophytes in 400 mM NaCl at various thermoperiods

Name of the species	5—15℃	5—25℃	10—20℃	15—25℃	20—30℃	25—35℃	References
Allenrolfea occidentalis	39	—	5	51	100	98	Gul and Weber, 1999
Suaeda moquinii	66	—	100	93	26	15	Khan, Gul and Weber, 2001a
Salicornia rubra	80	—	98	99	58	58	Khan, Gul and Weber, 2000
Kochia scoparia	79	—	93	94	57	57	Khan, Gul and Weber, 2001c
Sarcobatus vermiculatus	69	—	52	46	40	61	Khan, Gul and Weber, 2002a
Salsola iberica	46	—	36	38	30	81	Khan, Gul and Weber, 2002b
Triglochin maritime	—	79	10	30	—	—	Khan and Ungar, 1999
Polygonum aviculare	95	39	54	9	—	—	Khan and Ungar, 1998
Halogeton glomeratus	42	—	28	35	61	72	Khan, Gul and Weber, 2001b

Our data was unable to fit in the classification given by Woodell (1985), and could be categorized into four types and with further subdivisions. Type 1 are those where germination is completely inhibited at 300 mM NaCl and seeds showed a poor recovery response. Type 2 are the seeds which failed to germinate at 600 mM NaCl and most of them completely recover and about 30% showing less than 60% recovery. Type 3 are those seeds that failed to germinate at 900 mM NaCl and showed a complete recovery. Type 4 are those group that would include species with the complete inhibition of germination occurred beyond 900 mM NaCl and 70% completely recovered while seeds of 30% species failed to recover. Halophyte seeds from Great Basin desert are more tolerant to salinity during germination in comparison to subtropical halophytes (Khan and Gul, 2002) and germinated better at warmer temperature regimes. However, tolerance to salinity and temperature during seed bank did not show any clearly distinct pattern.

Table 4 Percentage recovery of germination of sub-tropical halophytes in 400 mM NaCl at various thermoperiods

Name of the species	10—20℃	10—30℃	15—25℃	20—30℃	25—35℃	References
Aeluropus lagopoides	42	—	65	89	88	Gulzar and Khan, 2001
Arthrocnemum macrostachyum	34	39	42	—	45	Khan and Gul, 1998
Atriplex stocksii	15	38	—	75	0	Khan, 1999
Cressa cretica	4	—	17	17	12	Khan, 1999
Haloxylon stocksii	55	30	40	—	5.5	Khan and Ungar, 1996
Limonium stocksii	95	—	98	92	98	Zia and Khan, 2001
Salsola stocksii	03	—	4	15	3.2	Khan, 2001
Sporobolus ioclados	11	—	4	25	8	Gulzar and Khan, 2001
Suaeda fruticosa	30	38	71	—	51	Khan and Ungar, 1998
Urochondra setulosa	11	—	27	57	29	Gulzar *et al.*, 2001
Zygophyllum simplex	03	4	15	—	3.2	Khan and Ungar, 1996

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