Action of plant growth regulators and salinity on seed germination of Ceratoides lanata

M. Ajmal Khan, Bilquees Gul, and Darrell J. Weber

Abstract: Ceratoides lanata (Pursh) J.T. Howell is a shrub with numerous annual branchlets from the family Chenopodiaceae and is widely distributed in brackish water playas of northern Utah. Seeds had no dormancy, and about 90% of the seeds germinated in nonsaline control. Seed germination decreased with the increase in salinity, and more than 10% of the seeds germinated at 900 mmol/L NaCl. Almost all seeds germinated in less than 24 h, and no additional seed germinated after this time. Gibberellic acid had no effect in alleviating salinity effects; however, kinetin and fuscinocin substantially alleviated the effect of salinity on germination, while ethyphen almost completely reverted the effect of salinity.

Key words: Ceratoides lanata, gibberellic acid, ethyphen, fuscinocin, halophytes, kinetin.

Introduction

Ceratoides lanata (Pursh) J.T. Howell (Diotis lanata Pursh; Eurotia lanata (Pursh) Moq.; Krasseninnikovia lanata (Pursh) Meeuse & Smith) (winterfat, white-sage) is a member of the goosefoot family, the Chenopodiaceae, some of the characteristics of which are described under green molly and shadscale. Winterfat typically is about 30 cm high, but it may on occasion reach a height of 1 m. Winterfat is generally regarded as one of the most desirable winter forage and despite 50 years of research, very few stands have ever been artificially established by seeding. Winterfat seed germination occurs in distilled water or under reduced salinity stress (Khan et al. 2001a, b), and ethylene concentration of delay of seed germination in halophytes (Baskin and Baskin 1998). Germination-regulating compounds such as gibberellic acid (GA₃) and kinetin (Ungar 1977, 1982, 1984; Proseus 1996; Khan and Rizvi 1994; Yaniv et al. 1995; Pyler and Proseus 1996; Khan et al. 1998; Khan and Ungar 2001a), fuscinocin (Ismael 1990; Gul and Weber 1998; Gul et al. 2000; Khan et al. 2000, 2001a, 2001b, 2002b), Maximum halophyte seed germination occurs in distilled water or under reduced salinity stress (Khan et al. 2001a). Great Basin desert halophyte species are very highly tolerant to NaCl (Rivers and Weber 1971; Ungar 1974; Khan and Weber 1986; Gul and Weber 1999; Khan and Ungar 1999; Khan et al. 2000, 2001a, 2001b, 2002b). Maximum halophyte seed germination occurs in distilled water or under reduced salinity stress (Khan et al. 2001a). Great Basin species with a very high salt tolerance at the germination stage include Kochia americana (1700 mmol/L NaCl, Clarke and West 1969), Salicornia pacifica (856 mmol/L NaCl, Khan and Weber 1986), Allenrolfea occidentalis (800 mmol/L NaCl, Gul and Weber 1999), Salicornia rubra (1000 mmol/L NaCl, Khan et al. 2000), Suaeda moquinii (1000 mmol/L NaCl, Khan et al. 2001a), Kochia scoparia (100 mmol/L NaCl, Khan et al. 2001b), and Sarcobatus vermiculatus (1000 mmol/L NaCl, Khan et al. 2002a).


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The present study is designed to study the effect of salinity and germination-regulating chemicals on seed germination of *C. lanata*.

### Materials and methods

Seeds of *C. lanata* were collected during August 1996 from a salt marsh situated 30 mi. (1 mi. = 1.609 km) south of the Great Salt Lake, at Faust, Utah. Seeds were separated from the inflorescence and were stored at 4 °C. Germination studies were started in January 1996. Seeds were surface sterilized using the fungicide Phygon (Hopkins Agricultural Chemical Co., Madison, Wis.). Germination was carried out in 50 mm × 9 mm (Gelman 7232) tight-fitting plastic Petri dishes with 5 mL of test solution. Each dish was placed in a 10 cm diameter plastic Petri dish as an added precaution against loss of water by evaporation. Four replicates of 25 seeds each were used for each treatment. Seeds were considered to be germinated at the emergence of the radicle.

Seeds were germinated in a growth chamber at an alternating temperature regime of 25–35 °C, where the higher temperature coincided with the 12-h light period (Sylvania cool white fluorescent lamps, 25 μmol·m⁻²·s⁻¹, 400–750 nm) and the lower temperature coincided with the 12-h dark period. Concentrations of 10 mmol/L ethephon, 5 μmol/L fusicoccin, 3 mmol/L GA₃, 0.05 mmol/L kinetin, and 0, 300, 600, and 900 mmol/L NaCl solutions were used. Percent germination was recorded every alternate day for 20 d. The rate of germination was estimated by using a modified Timson index of germination velocity: germination velocity = \( \Sigma Gt^{-1} \), where \( G \) is the percentage of seed germination at 2-d intervals and \( t \) is the total germination period (Khan and Ungar 1985). The maximum value possible using this index with our data was 50 (i.e., 1000/20). The higher the value, the more rapid the seed germination.

Germination data were transformed (arcsine) before a statistical analysis was performed. An ANOVA was used to determine whether significant differences were present among means. A Bonferroni test was carried out to determine whether significant \((P < 0.05)\) differences occurred between individual treatments (SPSS Inc. 1996).

### Table 1. F ratios and significance for the results of a two-way analysis of variance of germination responses by regulator and salinity treatments.

<table>
<thead>
<tr>
<th>Dependent variable</th>
<th>Salinity</th>
<th>Regulator</th>
<th>Salinity x regulator</th>
</tr>
</thead>
<tbody>
<tr>
<td>Germination</td>
<td>93.004</td>
<td>67.108</td>
<td>6.013</td>
</tr>
<tr>
<td>Rate of germination</td>
<td>91.678</td>
<td>65.540</td>
<td>5.732</td>
</tr>
</tbody>
</table>

*Note: All F ratios have a significance of \(P < 0.0001\).*

### Fig. 1. Final germination percentages of *Ceratoides lanata* seeds in 0, 300, 600, and 900 mmol/L NaCl. Values for salinity levels having the same letter are not significantly different \((P > 0.05)\) from the control (Bonferroni test).

### Fig. 2. Rate of germination of *Ceratoides lanata* seeds in 0, 300, 600, and 900 mmol/L NaCl.
Results

A two-way ANOVA showed a significant effect of salinity, germination-regulating chemicals, and their interaction on the percent germination and rate of germination (Table 1).

Seeds of *C. lanata* germinated (94%) in nonsaline control and germination was inhibited by an increase in salinity (Fig. 1). About 10% of seeds germinated with the 900 mmol/L NaCl treatment. Seed germination of *C. lanata* reached its peak in 24 h, and no further increase in germination was recorded in any salinity treatment (Fig. 2). Application of GA₃ had no effect on germination, under both nonsaline and saline conditions (Fig. 3). Kinetin significantly alleviated the effect of salinity, and at 90 mmol/L NaCl the germination was promoted from 10% to 50% (Fig. 3). Fusicoccin was more effective at the lower salinity concentrations used (300 and 600 mmol/L NaCl) by almost completely alleviating the effects of salinity (Fig. 3). Ethephon was the most effective of all the growth regulators in alleviating the effects of salinity; it completely alleviated the effects under low salinity, and at 900 mmol/L NaCl the germination increased from 10% in the nontreated control to 84% in the ethephon-treated seeds (Fig. 3).

Rate of germination was significantly increased by the treatment of kinetin, fusicoccin, and ethephon, while the effect of GA₃ was not significantly different from the effect of the control (Fig. 4).

Discussion

Seeds of *C. lanata* showed little dormancy when germinated under nonsaline control. Seed germination in *C. lanata* was inhibited with an increase in salinity, and 10% of the...
Fusicoccin, kinetin, and ethephon alleviated the inhibitory effects of salinity on germination, whereas GA3 showed no effect. Seed dormancy enforced by salinity was substantially alleviated by fusicoccin. Alleviation of salinity effect on seed germination by fusicoccin was also reported in seeds of other halophytes such as *Zygophyllum qatarensis* (Ismail 1990), *Allenrolfea occidentalis* (Gul and Weber 1998), *Atriplex stocksii* (Khan and Ungar 2000), *Sporobolus arabisicus* (Khan and Ungar 2001a), *Halopyrum mucronatum* (Khan and Ungar 2001b), and *Salicornia rubra* (Khan et al. 2002b). This alleviation may be due to the stimulation of ATPase production; ATPase rapidly increases during the early phases of germination to facilitate proton extrusion and K+ uptake (Marre 1979; Stout 1988). Fusicoccin has the ability to remove the inhibitory effect of abscisic acid on germination of normal seeds and on embryo growth of decoated seeds (Lado et al. 1975). It is more likely that abscisic acid production due to salinity stress could be counteracted by fusicoccin and alleviate the inhibitory effect of salinity. Fusicoccin, however, has little effect in alleviating the salinity effects on seed germination of *Triglochin maritima* (Khan and Ungar 2001c).

Effects of salinity on germination of *C. lanata* seeds were completely alleviated with the presence of ethephon in the medium. Application of ethephon relieves dormancy in seeds of several species (Ketring 1977; Bewley and Black 1994; Abeles and Lonski 1969; Adkins and Ross 1981; Corbino et al. 1989; Whitehead and Nelson 1992) and reverses the inhibitory effect of abscisic acid and osmotic stress (Karssen 1976; Schonbeck and Egley 1981). Ethylene may act by stimulating the germination of nondormant seeds or by breaking dormancy in seeds that exhibit an embryo dormancy (Ketring and Morgan 1969; Egley and Dale 1970;
Whitehead and Nelson 1992; Sutcliff and Whitehead 1995). However, seeds of many plants do not respond to ethylene (Ismael 1982), or some of the promotive effects are not substantial. Alleviation of salinity effects on the germination of halophytes by ethylene is quite variable. Ethephon completely alleviated the effect of salinity in *Allenrolfea occidentalis*, *Suada fruticosa*, *Kochia scoparia*, *Limonium stockissi*, and *Aeluropus lagopoides* (Gul and Weber 1998; Gulzar and Khan 2002; M.A. Khan, unpublished data), partially alleviated the effects of salinity on germination in *Arthrocnemum indicum*, *Salicornia rubra*, *Suada moquinii*, *Haloxylon glomeratus*, *Salicornia utahensis*, *Salsola iberica*, *Sporobolus ioclados*, *Zygophyllum simplex*, and *Atriplex rosea* (Khan and Ungar 2002; Khan et al. 2002b; M.A. Khan, unpublished data), but had no effect on *Triglochin maritima*, *Urophandra setulosa*, *Sarcobatus vermiculatus*, *Cressa cretica*, *Atriplex stockissi*, and *Atriplex prostrata* (Gulzar and Khan 2001; Khan and Ungar 1999, 2001a, 2001b; M.A. Khan, unpublished data).

GA3 failed to alleviate the effect of salinity on the germination of *C. lanata* seeds, under both saline and nonsaline conditions. GA3 was reported to cause a differential response to the germination of halophytes. Seed germination under saline conditions was almost completely alleviated in *Atriplex stockissi* and *Zygophyllum simplex* (Khan and Rizvi 1994; Khan and Ungar 1997), while some positive effects were reported for *Atriplex triangularis*, *Salicornia utahensis*, *Allenrollea occidentalis*, *Chrysothamnus nauseosus*, *Cressa cretica*, *Arthrocnemum macrostachyum*, *Polygonum aviculare*, and *Salicornia rubra* (Khan and Ungar 1995, 1998; Khan and Weber 1986; Khan et al. 1987; Gul and Weber 1998; Khan et al. 2002b; M.A. Khan, unpublished data), and species like *Triglochin maritima*, *Sporobolus ioclados*, *Urophandra setulosa*, *Suada fruticosa*, *Salsola imbricata*, and *Haloxylon stockissii* failed to respond to any GA3 treatment to alleviate the effects of salinity on germination (Khan and Ungar 2000; Gulzar and Khan 2002; M.A. Khan, unpublished data).

Kinetin substantially alleviated seed germination of *C. lanata* in salinities tested. Kinetin also caused a similar response in *Atriplex triangularis*, *Atriplex stockissi*, and *Zygophyllum simplex* (Khan and Ungar 1985, 1998; Khan and Rizvi 1994), and while it alleviated somewhat the effect of salinity on the seed germination of *Salicornia utahensis*, *Zygophyllum simplex*, *Arthrocnemum macrostachyum*, *Allenrollea occidentalis*, *Triglochin maritima*, and *Salicornia rubra* (Khan and Weber 1986; Khan and Ungar 1998, 2001; Khan et al. 1998, 2002b), it had no effect on the germination of *Cressa cretica*, *Sporobolus ioclados*, *Urophandra setulosa*, *Suada fruticosa*, *Salsola imbricata* and *Haloxylon stockissii* (Gulzar and Khan 2002; M.A. Khan, unpublished data).

*Ceratoides lanata* produces numerous seeds at the end of autumn and the beginning of winter. Seeds readily germinate if proper conditions are provided. Seeds in the natural environment were prevented from germination because of very cold temperatures. Seeds started germinating very early during spring, and germination decreased with the increase in salinity. This decrease in germination appears to be mediated through a reduction in germination-regulating chemicals like kinetin and germination inhibitors.


