Effect of Seed Weight and Salinity on the Germination of Garden Orache (Atriplex hortensis L.)

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ABSTRACT

Garden orache (Atriplex hortensis L.), a halophytic forb in the family Chenopodiaceae, is well adapted to dry saline habitats. Here, we presented the results of the effect of salinity and seed weight on seed germination and radicle emergence of A. hortensis. Highest germination percentages were obtained under non-saline conditions and increases in salinity inhibited seed germination with less than 50% of the seeds germinating at 260 mM NaCl. Seeds of A. hortensis exhibited a very pronounced morphological and physiological seed polymorphism. Seed weight varied from 0.11 to 0.19 g. Large seeds had a mean dry weight of 0.19 mg and a mean length of 2.6 mm; medium seeds and mean dry weight of 0.15 mg and mean length of 1.9 mm; small seeds and mean dry weight of 0.11 mg and mean length of 1.1 mm. Seedling dry weight was related to initial seed size. The degree of salt tolerance increased progressively with increasing seed dry weight. Radicle elongation was increased by low salinity and it decreased with an increase in salinity. Based on the observations, it is inferred that the germination percentage of large seed size class of A. hortensis was highest as compared to small seed size class. The seed size is an important factor that influences the germination under normal as well as, salinity stress conditions.

Key words: Annual Atriplex, salinity, germination, seed weight.

INTRODUCTION

Halophytes are potentially useful for ecological applications, such as landscaping, or rehabilitation of damaged ecosystems. Halophyte species vary in their tolerance to salinity during seed germination (Khan et al., 2002; Debez et al., 2011). Seeds of halophytes can recover the capacity to germinate after exposure to salt stress that inhibits their germination (Woodell, 1985; Khan et al., 2002). Population level effects of salinity stress may be likely to arise at the seed germination stage, particularly for annual species, because salinity has strong impacts on germination in many plants including halophytes (Li et al., 2010).

Annual halophytes vary in their upper limit of salt tolerance and increases in salinity usually cause a delay in germination (Ungar, 1996). Atriplex species are frequent in many arid and semi-arid regions of the world, particularly in habitats that combine relatively high soil salinity with aridity (Redondo-Gómez et al., 2007) and therefore constitute a useful material for the identification of physiological mechanisms involved in salt stress resistance.

Seed polymorphism can enable halophytes to adapt to varying salt-marsh or dry salt desert environments. It also enhances chances for seedling establishment and survival in a saline environment (Imbert, 2002). The presence of different kinds of seed/fruit morphs appears to be related to the ecological strategies of Atriplex species to maximize survival under variable environmental stresses.
Although, many halophytes are salt tolerant as adults, seed germination is often impaired by salinity (Atia et al., 2010a). Salt impact on the germination of halophytes is influenced by several factors including the salt type, light and temperature, but also the provenance, that is, geographical origin of the species investigated (Sosa et al., 2005; Atia et al., 2010a). Baker (1974) found that *A. triangularis* plants arising from large seeds had faster root and shoot growth than seedlings from small seeds that germinated at the same time.

In this context, the objectives of this study are to evaluate the effect of seed size (weight) and salinity on seed germination recovery and radicle elongation using seeds of *Atriplex hortensis*.

**MATERIALS AND METHODS**

**Plant material**

*A. hortensis* (L.), a C₃ xero-halophyte of the family Chenopodiaceae, is an annual species that is well adapted to saline and drought conditions. Seeds of *A. hortensis* were obtained from CN Seeds Ltd. (Ely, UK). Effects of salinity on the germination of various size classes of *A. hortensis* seeds and subsequent seedling growth were studied in the light.

The effects of seed weight on seed germination, seedling survival and growth of *A. hortensis* were studied and seeds of each of the 2 weight classes were separately soaked for 24 h in distilled water under laboratory conditions.

The seeds were categorized into two size classes, small (1.1 to 1.9 mm) and large size class (2 to 2.6 mm). On the basis of diameter, the standard of the weight class is the medium sized seed class and 50 seeds were used for each class.

Germination tests were carried out in Petri dishes with 5 ml of test solution, and each dish was placed in a 10 cm diameter plastic Petri dish as an added precaution to avoid water loss from dishes by evaporation. Six replicates of 25 seeds each were used for each of the treatments and seeds to have germinated after radicle emergence were considered.

**Effects of salinity on germination**

Three levels of salinity stress were produced through three concentrations of NaCl (90, 180 and 260 mM NaCl/L) against distilled water (0 mM NaCl/L) being run as control. The experiment used a randomized complete block design (RCBD), and seeds were considered as germinated when their emerging radicles were visible. Germinated seeds were counted and removed daily for a period of 15 days, after which no more germination was observed.

**Radicle elongation percentage**

Seeds were incubated initially in deionized water at 25 °C in the light for 15 days. When the radicles had barely emerged (<1.0 mm), 20 of these young seedlings were transferred into petri dishes containing deionized water control. Seedling incubation was terminated after 15 days and mean radicle length recorded.

**Final germination percentage**

Final germination percentage (GP) was calculated as the cumulative number of germinated seeds with normal radicles (Larsen and Andreasen, 2004) given as:

\[
GP = \sum n,
\]

Where *n* is number of seeds that germinated at each counting.

**Germination rate**

The rate of germination was estimated using a modified Timson index germination velocity (TIGV) given as:

\[
TIGV = \frac{\sum G}{t}
\]

Where *G* is the percentage of seed germination at 2 day intervals and *t* is the total germination period (Khan and Ungar, 1984).

**Data analysis**

Seed germination percentage and radicle length were expressed as mean ± s. e. One-way analysis of variance (ANOVA; *P* < 0.05) was used to compare the treatment effects.

**RESULTS**

**Effects of seed weight on germination**

Germination was rapid with almost all seeds germinating in control conditions. The frequency distribution indicated a bimodal distribution (Figure 1). Large seeds had a mean dry weight of 0.19 mg and a mean length of 2.6 mm; small seeds and mean dry weight of 0.11 mg and mean length of 1.1 mm. Seeds of all sizes in all treatments recovered very quickly. Final germination percentages for all seed sizes in all treatments were different from each other. Pre-soaking increased the final germination percentage of large seeds as compared with small seeds (Figure 2). However, radicle elongation progressively
Figure 1. Variation in seed weight of *Atriplex hortensis*.

Figure 2. Effect of seed polymorphism of *Atriplex hortensis* on germination after 15 days of incubation in deionized water. Values are means ± s. e.
increased for all seed sizes (Figure 3). Radicle growth of large seeds was much higher than that of small seeds.

**Effects of salinity on germination**

Analysis of Figure 4 revealed that at NaCl concentrations of 90 to 260 mM, germination rate decreased. Seed germination percentage did not reach 50% at salinities exceeding 260 mM until after 15 days of incubation (Figure 4). As the pre-treatment concentration of salinity increased, the percentage and rate of germination decreased, and for all salinities germination recovery percentages were higher than that of the control (distilled water). Germination recovery did not differ among seeds transferred from 0 to 90 mM NaCl solutions to deionized water.

To evaluate the effects of salinity on germination of *Atriplex hortensis*, germination percentage (GP) and germination rate (GR) were examined (Figure 5). Germination was affected by salt stress. As salinity increased from 0 to 260 mM NaCl, the rate and percentage of germination decreased and they were inhibited significantly by increased severity of salinity (Figure 5).

**Effects of salinity on radicle growth**

Radicle growth recovered after seedlings were transferred to deionized water. Concentrations of 90 to 260 mM NaCl decreased radicle elongation. No radicle elongation occurred at NaCl concentrations of ≥260 mM (Figure 6). However, elongation recovery decreased with the increase in pre-treatment salinities. Seedlings from solutions ≥260 mM NaCl showed no capacity for recovery (Figure 6). Furthermore, the radicle elongation was severely influenced by NaCl, the radicles from seedling under non-stress conditions were the longest, whereas those under the highest NaCl concentration were the shortest. Increasing the NaCl concentration resulted in a decrease in the radicle length, but no difference between 90 mM NaCl and the control was observed. The radicle growth was noticeably inhibited by 260 mM NaCl but not by 90 mM.

**DISCUSSION**

Germination is a principal component of seedling
Figure 4. Effect of NaCl concentration on germination of *Atriplex hortensis* seeds incubated at 25°C in the light for 15 days. Values are means ± s.e.

Figure 5. Effects of salinity on final percentage germination (GP) and the rate of germination (TIGV). Each point represents the mean of six replications.
establishment and survival and is considered the most critical phase of the plant life cycle (Rajjou et al., 2012). Germination is affected by several environmental and plant conditions including salinity, temperature, genotype, seed size and soil conditions (Bewley and Black, 2012). The study presented here revealed how seed weight and salinity can significantly affect germination of annual *Atriplex*. Our study demonstrated that germination was significantly enhanced when salinities were increased from 90 to 260 mM. The research presented here indicated differences in the germination percentage and radicle lengths of the *Atriplex* specie studied. Seeds of most halophytes germinate better in distilled water than in saline solutions, but they differ from glycophytes in their ability to germinate at higher salinities (Khan and Ungar, 1999). Halophytic grasses have been reported to have different levels of salt tolerance, ranging from 300 to 430 mM respectively (Khan and Ungar, 1999). Seeds of some halophytes are reported to tolerate high salinity during the period when they are dormant in the soil and subsequently germinate when soil salinities are reduced (Khan and Ungar, 1997).

Other investigations here showed that *Atriplex* spp. produce polymorphic/dimorphic dispersal units or polymorphic/dimorphic seeds (Mandák and Pyšek, 2001a). Seed size also affected seed germination in salt or drought stress in such *Atriplex* spp. as *A. nummularia* Lindl, *A.prostrata* and *A. patula* (Katembe et al., 1998).

Recent studies showed that seed size has strong effects on *A. sagittata* entire life cycle and it has an important impact on population regeneration in successive years (Mandák and Pyšek, 2005). Nobs and Hagar (1974) showed that, during the first 5 h, the large brown seeds of *A. hortensis* had a very rapid rate of water imbibition and reached saturation at 48 h. The small black seeds showed a considerably slower rate of water uptake. Osmond et al. (1980) reported that, under similar conditions, uptake was 100 times faster in brown than in black seeds of *A. hortensis*. The absorption of water by black seeds was more limited by the conductance of the seed coat than by the addition of NaCl to the solution (Osmond et al., 1980).

A similar insensitivity to NaCl was observed with the black seeds of *A. holocarpa* (Courtic, 1971). Increase in salinity progressively inhibited seed germination and 50% seeds germinated in the 260 mM NaCl treatment (Figure 4). Dimorphic seeds of the halophytes, *Atriplex prostrata* and *Atriplex patula* were treated with various solutions of NaCl and it was suggested that the influence of NaCl is a combination of an osmotic effect and a specific ion effect (Katembe et al., 1998).

In the present study, the radicle length was reduced by NaCl and the shortest length measured was under 260 mM NaCl (Figure 6). Seedlings of halophytes can develop in solutions with low concentrations of salts; however, radicle growth may be greatly retarded by high salinity (Malcolm et al., 2003).

Figure 6. Effect of NaCl concentration on radicle elongation of *Atriplex hortensis* at 25°C in the light after 15 days of incubation. Values are means ± s.e.
The germination percentage and rate of germination decreased with salt stress in *Atriplex* seeds (Figure 5). Germination rate decreased by salinity in *Atriplex hortensis*. This agreed with the results of Gholamin and Khayatnezhad (2010) in wheat; Mostafavi (2011) in safflower and Yildirim et al. (2011) in Physalis. Due to this fact more studies with salt stressed seeds germination are necessary for the complete elucidation of its effect on Orache development.

Conclusion

The overall results of this experiment showed the inhibitory effects of salt stress on seed germination parameters of *Atriplex* seeds. In conclusion, salt stress inhibited the germination of *A. hortensis* seeds and decreased the germination percentage and radicle lengths. Thus, understanding of the mechanics of seedling regeneration and seed bank formation is crucial to the successful management and restoration in habitats with high soil salinity.

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REFERENCES


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