

Research Note

Germination of *Dyckia encholirioides* (Gaudichaud) Mez var. *encholirioides* under saline conditions

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Summary

Dyckia encholirioides (Gaudichaud) Mez var. *encholirioides* is an endangered endemic bromeliad occurring in the Brazilian Atlantic Forest running along of the Atlantic Ocean. We investigated its seed germination at different salt levels (0, 100, 200, 300, 400 and 500 mmol L⁻¹ NaCl) under different temperature regimes (10-20, 15-25, 20-30 and 25-35°C). The highest seed germination rate was obtained at 0, 100 and 200 mmol L⁻¹ NaCl at 20-30°C and a further increase in salinity resulted in a gradual decrease in germination. Less than 5% of seeds germinated at 500 mmol L⁻¹ NaCl. Few seeds germinated at concentrations higher than 200 mmol L⁻¹ NaCl. Germination rate was fastest at 20-30°C and slowest at 10-20°C. Low seed germination rate was obtained in the dark in comparison to seeds germinated in a 12 hours photoperiod under saline conditions. Recovery experiments showed that exposure of seeds to various salinity and temperature regimes had little effect on viability of seeds *D. encholirioides*.

Experimental and discussion

Each plant species has particular requirements for seed germination as a result of adaptive radiation into patchy and changing environments. Soil salinity is an important constraint to plant fitness affecting about 95 million hectares worldwide. Although some plants are moderately tolerant to saline conditions, many others are adversely affected. Salinity impairs seed germination, reduces nodule formation, plant development and reduces crop yield (Szabolcs, 1994).

Tolerance to salinity during germination is critical for the establishment of plants growing in saline soil of arid regions (Schabes and Sigstad, 2005). In these regions, germination occurs during rainy seasons when soil salinity levels are usually reduced. Even in the halophytic species, best germination is obtained under non-saline conditions and their germination decreases with increases in salinity (El-Keblawy 2004, El-Keblawy and Al-Rawai, 2005).

The Atlantic Rain Forest is a Brazilian biome containing high biological diversity and endemism (Pompelli and Guerra, 2005). The large scale fragmentation of this biome and its associated ecosystems resulted in a genetic erosion of its components. *Dyckia encholirioides*, is a bromeliad native of this biome. This species is not halophyte, nevertheless it is distributed at long of seaboard under rocks where is vulnerable to water with low osmotic potential. Possible sources of moisture are the ocean water or summer rains. *D. encholirioides* flowers once a year, from September to November and a large number of seeds are produced from October to December. After dispersal seeds become part of the seed bank and rarely germinate. The few plantlets found are originated by stolons. The average temperature during the summer period ranges from 20°C at night to 35°C during the day or 40°C or more on the rocks.

The present study focuses on *in vitro* requirements for seed germination of *Dyckia encholirioides* by analyzing the response to low osmotic potential and the effects of light and temperature in the seed germination.

Seeds of *D. encholirioides* were collected in the Santinho beach (Florianópolis, SC, South Brazil –27°26' S, 48°22' W), transported and disinfected in agreement with Pompelli and Guerra (2005), except that the seed were inoculated in Petri dishes containing 10 mL of test solution (distilled water jelled with agar – 6 g L⁻¹ and supplemented with different NaCl levels). Three replicates with 40 seeds each were used in each treatment. Seeds were considered to be germinated at the emergence of the apical shoot, in agreement with Pompelli *et al.* (2005). Seeds were germinated in germinator at four alternating temperature regimes of 10-20°C, 15-25°C, 20-30°C and 25-35°C. A 24 hours cycle was employed where the temperatures of 20°C, 25°C, 30°C and 35°C were associated to 12 hours light period provided by Sylvania cool white fluorescent lamps resulting in a light intensity of 40 μmol photons m⁻²s⁻¹. The temperatures of 10°C, 15°C, 20°C and 25°C were coincident with a 12 dark period. Seeds were germinated at six salt levels (0, 100, 200, 300, 400 and 500 mmol L⁻¹ NaCl). Germination rate was recorded every two days during 20 days. Non-germinated seeds were transferred to distilled water after 20 days to study the recovery of germination, which was also recorded at two days intervals for 20 days.

The statistical analysis showed significant effects of salinity, temperature and their interaction on germination rate and recovery rate of *D. encholirioides* seeds. The highest germination rate in light was observed in the non-saline control under all temperature regimes. Different salinity levels resulted in a gradual decrease in the germination rate and this reduction ranged with the changes in the temperature regime. Highest germination rate under saline conditions was observed at 20-30°C treatment (figure 1). Germination rate was highest in non-saline controls except at 15-25°C and addition of NaCl reduced the germination rate. Temperature also influenced germination rate. At low and high temperatures, seeds showed a reduced germination rate from 100 to 300 mmol L⁻¹ NaCl than the values observed at 20-30°C and 15-25°C (figure 1).

Temperature also affected germination velocity under both saline and non-saline conditions. Highest germination in the distilled water control was obtained after two days under all temperature regimes except for 10-20°C. In saline solutions highest germination ranged from eight 8 to 20 days. Seed germination peaked at 10 days for all salt concentrations at 15-25°C, except in 300 mmol L⁻¹ (20 days) (figure 2).

GERMINATION OF *DYCKIA ENCHOLIRIROIDES* UNDER SALINE CONDITIONS

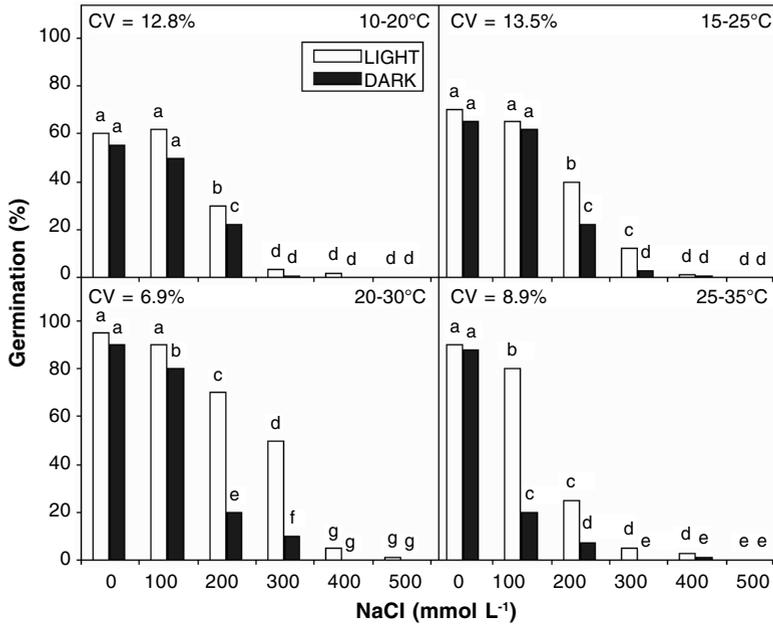


Figure 1. Germination rate of *Dyckia encholirioides* as affected by salinity, temperature and light or dark conditions. Bars with the same letter within each temperature treatment are not significantly different (Newman-Keuls $p \leq 0.01$).

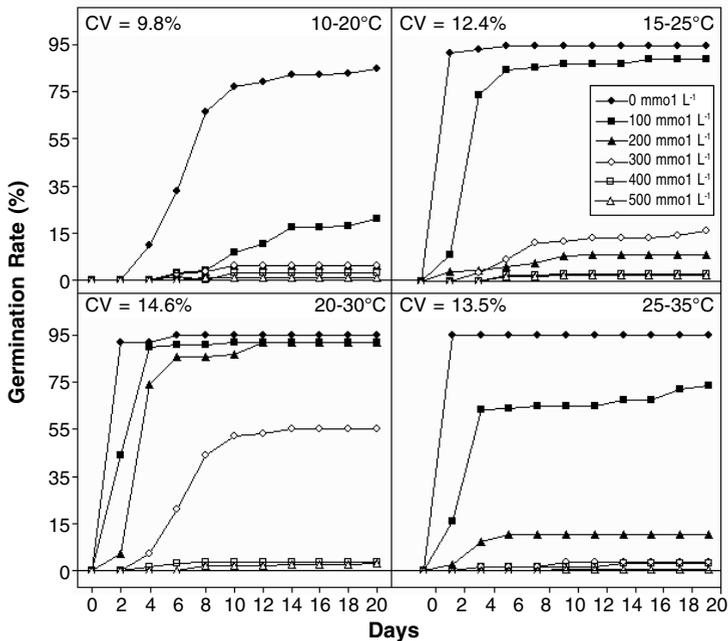


Figure 2. Cumulative mean percent germination of *Dyckia encholirioides* seeds over time in 0, 100, 200, 300, 400 and 500 mmol L⁻¹ NaCl in 12 hours light and 12 hours dark photoperiod.

In this study seed germination was also influenced by temperature. We found 20-30°C to be the optimal temperature for germination and any increase or decrease in temperature inhibited germination. This inhibition was progressively increased by salinity, suggesting that this is responsible for the absence of seed germination of this species in its natural sites. In agreement with Khan and Gul (2002) germination of halophyte seeds in subtropical coastal and inland salt marshes usually occurs after monsoon rains, which causes a reduction in temperature and reduces the soil salinity.

Several reports show that seed germination rate is more sensitive to salinity than is overall germination rate (Marcar, 1987). Very rapid germination was reported for *Haloxylon recurvum* Bunge ex. Boiss and *H. salicornicum* Bunge ex. Boiss (Sharma and Sen, 1989) and *Limonium axillare* (Forssk.) (Mahmoud *et al.*, 1983) and this was considered to be a strategy to utilize the brief period of water availability after rainfall. Rogers *et al.* (1995) suggested that fast germination ensures rapid seedling establishment which can minimize competition.

In this study, seeds of *D. encholirioides* germinated rapidly in the control treatment and in up to 200 mmol L⁻¹ NaCl at 20-30°C, a temperature regime similar to the average early summer period in south of Brazil. The pattern of germination of *D. encholirioides* is very similar to the pattern described by Rubio-Casal and coworkers (2003) for *Arthrocnemum macrostachyum* and *Salicornia ramosissima* J. Woods or *Triglochin bulbosa* L. and *T. striata* Ruiz and Pavón (Naidoo and Naicker, 1992).

Germination was also reduced by darkness in all temperature and salinity treatments and the same was observed in *Limonium stocksii* seed by Zia and Khan (2004). However, a contrary behaviour was described by Navarro and Guitián (2003) and other species (see Martin *et al.*, 1995).

Dyckia encholirioides seeds displayed a great tolerance to high-salinity and temperature stress before germination. Seeds germinated within two days when transferred to non-saline media from various salinity treatments and temperature regimes. Similar results were obtained by Mahmoud *et al.* (1983) for *Limonium axillare*, which showed 95% recovery for 60%-100% seawater treatments. Khan (1999) reported a quick recovery of seeds at all temperature and salinity regimes. The ability of halophyte seeds to survive in hypersaline conditions and germinate when salinity is reduced provides them with multiple opportunities for their establishment in unpredictable saline environments.

In *D. encholirioides* the dormancy reduces the risk of seedling mortality when moisture is limited and salinity is increased during summer. The inhibitory effect of salinity at higher temperatures has ecological significance since it prevents seeds from germinating in salt-affected habitats and consequently avoids seedling mortality during this period. This could be true if the damage caused by Na⁺ toxicity is not irreversible (El-Keblawy and Al-Rawai, 2005). To compensate the low rate and germination speed, an alternative strategy for *D. encholirioides* plants is the asexual reproduction through offshoot, as methodology described by Pompelli and Guerra (2005) to *D. distachya* Hassler. This strategy allows the continuous colonization of new areas and the perpetuation of the species. Even considering a low germination rate plants derived from seeds contain new allelic combinations that fit to this harsh environment. These reproductive strategies are responsible to the adequate fitness of this species in this environment.

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