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Short communication

Germination of *Denmoza rhodacantha* (Salm-Dyck) Britton & Rose (Cactaceae)

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Abstract

The germination response of *Denmoza rhodacantha* (Salm-Dyck) Britton & Rose to seed scarification and different calcium concentrations was analysed. Both scarified and unscarified seeds were treated with two different calcium concentrations (1.74 or 6.36 meq l^{-1} calcium sulfate in distilled water). All treatments were conducted under a constant temperature (30 °C), and a photoperiodic regime of 12 h light and 12 h dark. Germination was initiated significantly sooner (3.6 days), and the rate of final germination was significantly higher (90.8%), for scarified seeds treated with a solution of 6.36 meq l^{-1} Ca compared with all other treatments (9.0–11.4 days and 3.6–6.8%, respectively). There were no significant differences among the treatments for the time in which 50% of final germination occurred (6.9–13.6 days). The results suggest that both factors, scarification and calcium concentration, favor germination of *Denmoza* seeds.

Keywords: Cactus seeds; Scarification; Calcium solution; Cardonal; Argentina

Scarification is an important factor that favors the germination of cactus species in arid and semi-arid habitats (Alcorn and Kurtz, 1959; Pilcher, 1970; Bregman and Bouman, 1983; Potter et al., 1984; Cheema and Mehra, 1985; Rabenda, 1990; Romero-Schmidt et al., 1992; Rojas-Aréchiga and Vázquez-Yanes, 2000; Olvera-Carrillo et al., 2003). The presence of a hard seed coat could act as a mechanism that favors seed dormancy, which may be associated with physical protection (impermeability) or with the presence of

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inhibitors in the seed coat, as has been reported in cactus species (Williams and Arias, 1978; Rabenda, 1990). During personal observations of the *Larrea cuneifolia* community (“jarillal”) accompanied by *Denmoza rhodacantha* (“cardonal”), an increase in the density of juvenile populations of *D. rhodacantha* was found on eroded sites. This observation led to the theory that the mechanical abrasion of seed coats by rock particles moving down slope could favor an increase in seed germination on these eroded sites in years of higher precipitation.

In the present study, germination tests were used to determine if seed scarification by soil particles during high precipitation events could enhance the germination of *D. rhodacantha* (Salm-Dyck) Britton & Rose (Britton and Rose, 1922). *D. rhodacantha* seeds were mechanically scarified with sand as similarly as possible to what occurs in nature. The germination tests with scarified seeds were expected to yield higher rates of germination than tests with unscarified seeds. In addition, scarified and unscarified seeds were treated with two different calcium concentrations equal to those obtained from the soil chemical analysis from the eroded and uneroded seed collection sites. It was expected that treatment with higher calcium concentrations would favor germination by breaking seed dormancy. It is known that plants need essential elements, such as calcium, to grow and develop (Maximov, 1946; Bonner and Galston, 1967; Devlin, 1970). Furthermore, it has been suggested that the calcium ion plays an important role in initiating physiological processes (Plaxton, 1990; Lenormand et al., 1993) and as a second messenger that is important in the modulation of hormones (Hepler and Wayne, 1985; Owen, 1988). It has been found that many plants need at least small amounts of Ca to germinate. Could just that treatments of scarification and calcium concentration improve the germination? The objectives of the study were to: (1) examine the effects of scarification and calcium concentration on *D. rhodacantha* germination; and (2) determine the most appropriate presowing treatments for enhancing germination. Germination tests with *D. rhodacantha* could contribute to information that supports the conservation of cactus populations of this species.

The fruits of *D. rhodacantha* from populations of this species growing on northern slopes of the Médanos Mount between 1600 and 1800 m a.s.l. in the mountain area of the Mendoza River Valley (32°49′S, 69°10′W) were used in this study. *Denmoza* populations are practically intact on this site, because of the absence of roads, livestock grazing, and nearby human settlements. To perform germination tests with *D. rhodacantha* seeds, 10 ripe fruits were collected from 10 individuals during November and December, 2004. The fruits were cut from the plant, and the seeds were manually removed from the pulp. The ripe and black seeds from different fruits were mixed together and dried at room temperature (20 ± 2 °C) in the shade. The seeds were stored in paper bags. Half the seeds were immediately scarified with sand inside a plastic tube. Scarification time by mechanical agitation was 45 min, which was sufficient time to remove the external tegument and strophiole. After scarification the edges of the seeds were smooth and rounded when observed through a microscope. Unscarified seeds retained their tegument, strophiole, and rough edges. After this, six treatments were prepared, three using scarified seeds and another three using unscarified seeds. All treatments were performed with five replications of 50 seeds each. The seeds were placed in 9-cm-diameter Petri dishes, with filter paper on a cotton bed moistened with distilled water, and treated with solutions of calcium sulfate containing calcium concentrations of 1.74 or 6.36 meq l^{-1}. The Petri dishes were placed in a growth chamber (Precision, model 818, with dual timers for programming both light and temperature conditions, 220 V, 50 Hz, 4.9 A) at a constant temperature (30 °C), under a

Table 1
Mean values of percentages and times (expressed in days) of germination of *Denmoza rhodacantha* seeds under different treatments

Data	Treatments					
	Unscarified			Scarified		
	Water	Solution 1.74 meq l^{-1} Ca	Solution 6.36 meq l^{-1} Ca	Water	Solution 1.74 meq l^{-1} Ca	Solution 6.36 meq l^{-1} Ca
SG (days)	9.0 b	11.4 b	10.6 b	10.2 b	10.2 b	3.6 a
T_{50} (days)	12.3 a	13.6 a	10.2 a	10.9 a	11.1 a	6.9 a
PG (%)	3.6 b	4.8 b	1.6 b	5.2 b	6.8 b	90.8 a

Mean values in the same row followed by the same letter do not significantly differ at the 5% probability level. SG: start of germination; T_{50}: time in which 50% of the final germination occurs; and PG: percentage of final germination.

photoperiodic regime of 12 h light, with white light (20 W fluorescent lamp) and 12 h dark. Petri dishes were checked daily to record the number of germinated seeds (emerged radicles). The following data were determined: number of days to the start of germination (SG), number of days to obtain 50% of final germination (T_{50}), and percent germinated seed (PG). Germination was assumed to be finished 18 days after it had started and an additional 5 days during which no further germination was observed.

Data obtained were statistically analysed using ANOVA. The means were separated with DGC' test ($p<0.05$) (Di Rienzo et al., 2002). This test allows for multiple comparisons and was selected because it does not produce the typical overlap as other methods.

The results of the seed germination treatments are shown in Table 1. Germination was initiated significantly sooner (3.6 days), and the rate of final germination was significantly higher (90.8%), for scarified seeds with the addition of 6.36 meq l^{-1} Ca, compared with those for all the other treatments. In all the other treatments, germination was initiated much later (9.0–11.4 days), and the rate of final germination was very low (1.6–6.8%). There were no significant differences among all these other treatments. The number of days in which 50% final germination occurred was not significantly different among all treatments.

Even though scarification appears to favor germination of *D. rhodacantha* seeds, the calcium concentration also played a major role in seed germination in this experiment. While the scarified *D. rhodacantha* seeds in the 6.36 meq l^{-1} Ca ($=127.2$ mg l^{-1} Ca) solution resulted in very high germination rates, scarified seeds in the 1.74 meq l^{-1} Ca ($=34.8$ mg l^{-1} Ca) solution resulted in relatively low germination rates. Other studies have identified the important role of calcium in seed germination. For example, the seeds of *Hypericum perforatum* will not germinate in the presence of more than a trace amount of Ca, whereas tomato seeds germinate only when well supplied with this element (Daubenmire, 1947). Although, calcium chloride solutions are known to stimulate tissue growth (George and Sherrington, 1984; Clayton et al., 1990), opposite results have been found in species other than Cactaceae. Ongaro and Prado (1996), using solutions of 225 mM calcium chloride, reported a 14% decrease in the germination rates of *Chenopodium quinoa* Willd. In addition, germination was reduced by 50% using

300 mM calcium chloride, which was attributed to osmotic reasons. A specific response to calcium concentrations in the germination of gypsophytes and gypsovags sometimes cannot be identified (Escudero et al., 1997), and it has even been pointed out that gypsum could be selectively active, at times, in seedling establishment (Meyer, 1986).

It is suspected that the low germination rates obtained in the treatments with unscarified seeds in the different concentrations of calcium are a result of the presence of the seed tegument acting as a barrier to normal germination. Moreover, the presence of high calcium concentration on eroded sites may favor, in years of higher precipitation, water retention in the soil for a longer period of time, as has been demonstrated in gypsum soils in research regarding the germination of *Hordeum squamatum* (Meyer, 1986; Escudero et al., 1997). Water retention, as a result of higher calcium concentrations in the soil, may favor the germination of seeds of *Denmoza*.

The rapid initiation of *D. rhodacantha* seed germination could be the result of the joint action of scarification and treatment with the 6.36 meq l^{-1} Ca solution, which could break the inhibition mechanism produced by the seed tegument or unblock a possible unknown inhibitor. The low germination rates in the other treatments could be due to a presence of inhibitors or simply because the hard seed coat did not become permeable with these treatments. This research did not include the identification of why the seeds react to particular treatments or substances, or the optimal calcium concentrations to achieve maximum germination. However the results of these germination tests indicate that *D. rhodacantha* seeds do require presowing treatments with solutions of high concentrations of calcium and mechanical scarification to obtain the best germination rates. In conclusion, the effect of scarification is less than the effect of calcium on *Denmoza* seed germination. The higher concentration of calcium helped to reinforce the effect of scarification through a mechanism that is still not fully understood, but that improved the germination. This suggests that dormancy in these seeds might be due not only to the hardness and impermeability of the seed coat, but also to other inhibitors, still unknown, that could to be unblocked by higher calcium concentrations.

The large variation in the quantity of germinated seeds and juvenile plants observed on eroded relative to uneroded sites could be related to ecological differences between the sites. In agreement with the postulated hypothesis, eroded sites that contain soil calcium concentrations at mean values of 6.36 meq l^{-1} were found to contain the highest number of juvenile *Denmoza* plants.

The use of seed scarification, combined with treatment with a calcium solution, may be valuable information for further projects directed toward the restoration of *D. rhodacantha* populations on the mountain slopes in arid western Argentina.

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