Propagation of *Dryas octopetala* L. and *Alchemilla alpina* L. by direct seeding and planting of stem cuttings

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ABSTRACT

Experiments focused on propagation of *Dryas octopetala* and *Alchemilla alpina* were established on wind eroded sites in South Iceland. Seeds were sown in four microsite types and cuttings were planted in a sandy gravel soil with and without added fertilizer (N, P). D. *octopetala* seedlings emerged in 88% of seeding spots. Seedling survival was best in biological soil crust, lowest in gravel bed and intermediate in moss microsites or under dwarf shrubs. Very few *A. alpina* seedlings emerged during the first growing season but appeared in 96% of seeding spots in the second year. Seedling survival was greatest in biological soil crust and lowest in gravel or moss. Five percent of *D. octopetala*-cuttings and eighty percent of A. *alpina* base-stem cuttings survived for one year. After three years most of the fertilized A. *alpina* plants and some of the unfertilized ones survived, but the *D. octopetala* plants were all dead. The results show sowing as a promising method for propagation of both species, but planting of cuttings was also successful for *A. alpina*. Utilization of these species in restoration, e.g. to increase floral diversity, should be tested further.

Keywords: establishment, native species, restoration

YFIRLIT

Fjölgun holtasóleyjar og ljónslappa með sáningu og græðlingum

Markmið rannsóknarinnar var að kanna leiðir til að fjölga holtasóley, *Dryas octopetala,* og ljónslappa, *Alchemilla alpina,* á röskuðum svæðum. Fræi var sáð og græðlingum plantað á vindrofnum svæðum á Suðurlandi. Sáð var í bletti með fjórum svarðgerðum og tvígildur (N, P) áburður borinn á helming þeirra. Holtasóleyjarfræ spíraði í 88% sáningarbletta. Flestar kímplöntur lifðu af í lágplöntuskán, færri í mosa eða í krækilyngi en fæstar í malarsverði. Ljónslappi spíraði lítið fyrsta árið en kímplöntur fundust í 96% sáðbletta á öðru ári eftir sáningu. Kímplöntum vegnaði best í lágplöntuskán eða krækilyngi, verr í möl eða mosa. Græðlingum var plantað í örfoka sandmel og tvígildur áburður borinn á helming þeirra. Eftir ár voru fimm af 100 holtasóleyjargræðlingum lifandi en 80 af 100 ljónslappagræðlingum. Eftir þrjú ár voru flestar ábornu ljónslappaplönturnar lifandi og nokkrar af þeim óábornu en engin holtasóley lifði svo lengi. Niðurstöðurnar sýna að sáning er álitleg leið til að fjölga báðum tegundum og mögulegt er að fjölga ljónslappa með græðlingum. Áhugavert væri að prófa báðar tegundir í uppgræðslu, til dæmis til að auka fjölbreytni gróðurs.

INTRODUCTION

Revegetation of eroded landscapes in Iceland in order to restore their structure and function has been based on relatively few species, many of which are non-native. Revegetation methods are changing with more ecological understanding (Magnússon 1997) and there is an interest in increasing the diversity of native species that can be used for this purpose. This paper reports on propagation experiments with *Dryas octopetala* L. and *Alchemilla alpina* L., which are common in native heathland vegetation of Iceland. Both species are potential candidates for use in restoration. They can grow under difficult conditions on disturbed sites and *A. alpina* is often found on unstable land such as gravelly slopes, screes and erosion escarpment.

D. octopetala is widely distributed in circumpolar regions of the Northern Hemisphere (Hultén & Fries 1986) and has a record of early colonization following deglaciation (e.g. Elkington 1971, Bennike et al. 1999, Bergman et al. 2005). The general ecology of *D. octopetala* is fairly well known (Elkington 1971). Propagation is primarily by seed which is wind dispersed, although some vegetative recruitment may occur as creeping branches set adventitious roots. Quality of seed can vary considerably, probably due to the climatic factors during seed development (Welker et al. 1997, Wada 1999, Hagen 2002, Wada & Nakai 2004).

Propagation of *A. alpina* has not been studied very much. Dispersal is primarily by seed, but can also be vegetative by fragmentation of larger rhizome systems (Kershaw 1960). Germination of *Alchemilla* seeds in regulated experiments has been rather poor (e.g. Austrheim & Eriksson 2003). On the other hand, spontaneous emergence of *Alchemilla* species in disturbed soil is not uncommon as the seed is rather well conserved in soil (e.g. Milberg 1992, Smith et al. 2002).

Experiments with sowing of seed and planting of cuttings of *D. octopetala* and *A. alpina* started in 2002, as part of a larger study of their regeneration ecology. First results were published in 2004 (Karlsdóttir 2004). This paper contains a two year follow-up of the experiments.

MATERIALS AND METHODS

Study area

Sowing experiments were set out at Vakalág, South Iceland (63.850°N, 20.306°W, 60 m a.s.l.) and the cutting experiments were placed at nearby Geitasandur (63.812°N, 20.154°W, 65 m a.s.l.). Both experiments were on eroded sites, with shallow infertile soils on a sandstone bed (Bergsveinsson 2000). These sites had an unstable soil surface due to cryoturbation and wind erosion. The soil surface was characterized by sand and gravel and the vegetative cover was sparse (<10%) (Gretarsdottir et al. 2004, Aradóttir & Halldórsson 2004).

Large patches of the Vakalág area have been revegetated and the seeding experiment was established on a gradient from untreated, eroded surface to a patch receiving light fertilization in 1982. The fertilized patch was characterized by dwarf shrubs, biological soil crust, and small cushions of *Racomitrium* moss, 0-5 cm thick.

The cuttings experiment on Geitasandur was established on untreated, eroded soil.

Seed

Seed was hand-picked in the summer of 2002 and stored in paper bags at approximately 17°C. Seed was weighed in January 2004 after 506 (*A. alpina*) and 546 (*D. octopetala*) days of storage. Five times 0.50 g of seed was counted for each species. *D. octopetala* seeds with dispersal hairs weighed on the average 941±30 µg seed^{-1} or 1067±33 seeds g^{-1}. *A. alpina* seeds with sepals weighed on the average 539±8 µg or 1857±26 seeds g^{-1}.

A preliminary test of seed quality was set up in the autumn of 2001 by sowing 200 seeds from each species on cultivating soil in shallow plastic trays which were kept outdoors over the winter of 2001-2002. Seedlings were counted in May 2002. Seed germination was 24% of *A. alpina* and 7% of *D. octopetala*.

Seeding

The seeding experiment was established on three 20x2 m transects, extending from a fertilized area dominated by dwarf shrubs and biological soil crust to an untreated area with sand and gravel. Twenty seeding spots of four microsite types were selected on each transect, totaling 240 seeding spots. The microsite types were bare ground with gravel surface, biological soil crust, moss and under *Empetrum* dwarf shrubs. On each spot ca. 100 *D. octopetala* seeds or ca. 50 *A. alpina* seeds were sown. About 1 g of synthetic fertilizer (26% N, 6.1% P, 3% Ca) was applied to 50% of the seeding spots which corresponds to about 30 g m^{-2} or about 7.5 g N m^{-2}. Species and fertilization treatments were assigned randomly to the seeding spots within each microsite type. The seeding took place on 27 October 2002 and seedlings were counted on 21 April and 17 August 2003, 17 June and 5 Oct. 2004 and 11 June 2005. The fate of individual seedlings was not followed and thus the data show number of living seedlings on each occasion but do not show total germination or mortality.

Planting of cuttings

Cuttings were prepared from five adult plants of each species in June 2002. All cuttings had part of a stem with leaves and buds. *A. alpina* cuttings had the uppermost part of the root attached and *D. octopetala* cuttings had some adventitious roots forming from creeping stems. Flowers were removed. Cuttings were of two sizes, <10 cm or >10 cm long. Cuttings were kept in plastic bags overnight and planted the following day.

Five identical cuttings were planted in each 1 m^2 study plot. The plots were arranged side by side, eight in a row, and replicated in five rows. The eight possible combinations of plant species, cutting size and fertilization treatment were randomized within each replication. In the fertilized treatment 1 g of fertilizer (26% N, 6.1% P, 3% Ca) was applied to each cutting at the time of planting and again the following spring. Living plants were counted in July and September 2002, August 2003, August 2004 and July 2005. A plant was considered alive if it had one or more green leaves, regardless of its vigor. Flowering was recorded in August 2003, July 2004 and August 2005.

Data analysis

The effects of species, microsite type and fertilization on percentage of spots with living seedlings and number of seedlings were analyzed separately for each observation date using factorial ANOVA, with transects as replications. Tukey's HSD was used for mean separation of microsite types ($\alpha=0.05$). Factorial ANOVA was also used to estimate the effects of species, cutting size and fertilization on percentage of living plants in planting plots. After the first growing season, only *A. alpina* was included in the analysis as nearly all the *D. octopetala* plants had disappeared. The statistical analysis was done by SPSS, version 13.0 (SPSS 2004).

RESULTS

Sowing

Seeds were sown in the autumn of 2002 and in the spring of 2003 seedlings were found in 88% of the spots sown with *D. octopetala*, 518 seedlings in all, while only eight seedlings of *A. alpina* had emerged (Figure 1 and 2). One year later the *A. alpina* seeds had germinated abundantly and 2053 seedlings were observed, representing nearly 34% of the seeds sown. There was a significant difference in germination of seeds between species on all observation dates ($p<0.01$). *D. octopetala* was found in more seeding spots and in higher numbers per spot than *A. alpina* during the 2003 growing season. However *A. alpina* was more abundant in the subsequent years (Figures 1 and 2).

The effect of microsite type was significant for combined species ($p\leq0.01$) on all observation dates, for both percentages of seeding spots and for number of seedlings. Initial germination, observed in April 2003, was greatest in the gravel and lowest in the dwarf shrub microsite type. In August 2003 and on all subsequent observation dates, the biological crust microsite had the greatest number of seedlings

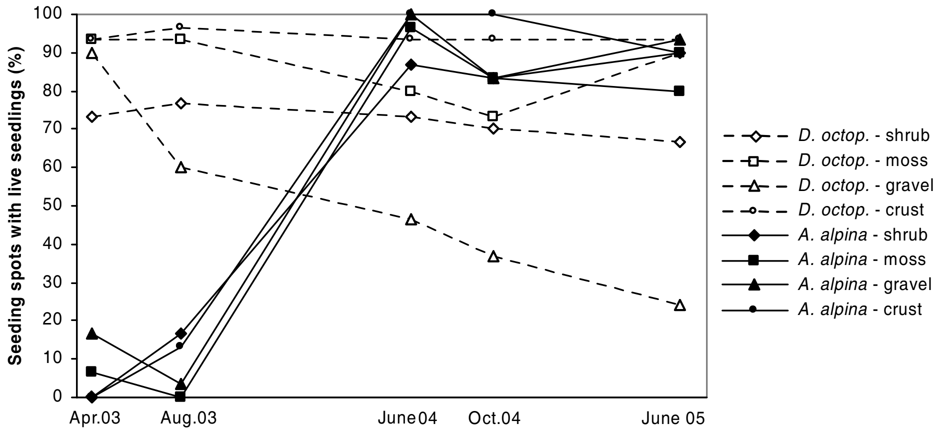

Figure 1. Proportion of seeding spots with living seedlings by species and microsite-type.

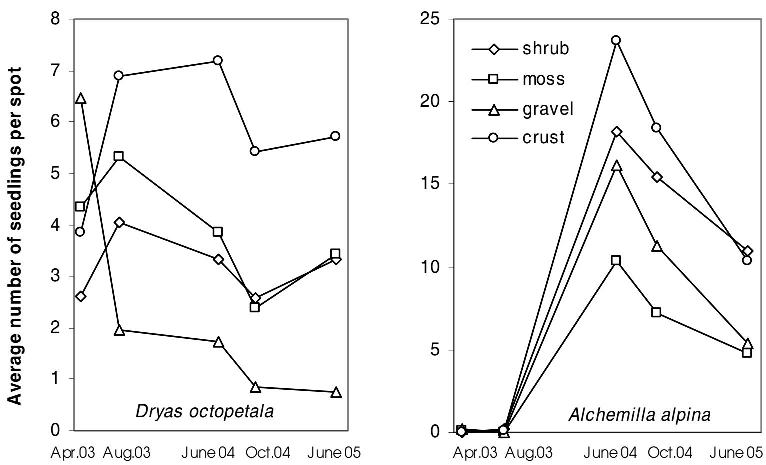

Figure 2. Average number of living seedlings by species and microsite-type.

and highest percentage of filled seeding spots. There was, however, a significant interaction ($p<0.05$) between species and microsite type on all but the first two dates. *D. octopetala* was more adversely affected in the gravel microsite type than *A. alpina*.

Fertilization did not significantly ($p>0.1$) affect the percentage of filled seeding spots on any observation date, whereas the number of seedlings was negatively affected by fertilization ($p<0.001$) on June 2004 and at all subsequent observations. This negative effect was especially strong in *A. alpina*, and there was significant interaction between species and fertilization ($p<0.01$) for both of the observations in 2004.

Cuttings

Most of the *D. octopetala* cuttings withered and died the first summer, but *A. alpina* cuttings had a greater survival rate (Figure 3). The difference between the species was significant ($p<0.001$). In the first growing season, there was neither a significant effect of cutting size nor an effect of fertilization. However, significant interaction between species and fertilization ($p<0.05$) was found, because survival of unfertilized *D. octopetala* cuttings was greater than survival of fertilized cuttings. The reverse was observed for *A. alpina*. Only a handful of *D. octopetala* cuttings survived after the first growing season and no plants of that species

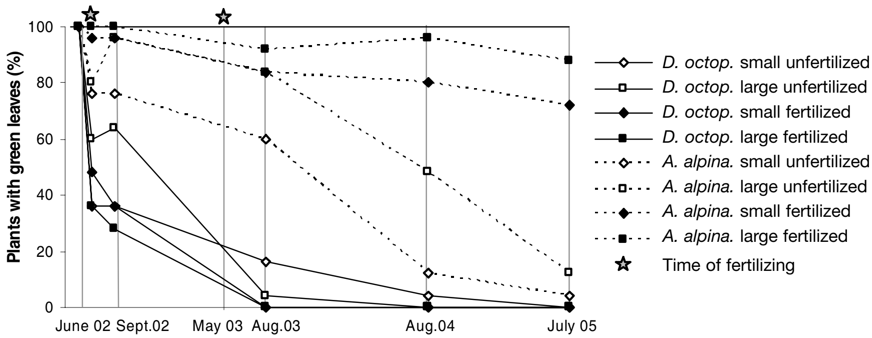

Figure 3. Proportion of plants with one or more green leaves by species, size of cuttings planted and fertilization.

were alive in 2005.

Eighty of the original 100 *A. alpina* cuttings were alive in August 2003, 59 in August 2004 and 44 in July 2005. The effect of fertilization on survival of *A. alpina* was positive (Figure 3, $p<0.001$) and in 2005 very few unfertilized plants were left. The effect of cutting size on survival was mixed (Figure 3). Large cuttings of *D. octopetala* died before smaller cuttings. On the other hand, survival of large *A. alpina* cuttings was better than that of small cuttings, but the effect of cutting size on survival was significant only in August 2004.

None of the *D. octopetala* cuttings flowered after planting. In August 2003, 30 of the 80 living *A. alpina* plants had flowered, but fewer plants flowered in the following years. Fertilization had a significant effect on flowering ($p<0.05$).

DISCUSSION

Both *A. alpina* and *D. octopetala* regenerated successfully from direct seeding on an eroded and partially reclaimed site (Figure 1), but planting of cuttings into a sparsely vegetated eroded site was only successful for *A. alpina* (Figure 3). The effectiveness of these two propagation methods could be attributed to differences in species characteristics, but environmental differences between the two study sites can not be ruled out.

Sowing experiment

In the first spring after seeding, seedlings were found in most of *D. octopetala* seeding spots. Most of the *A. alpina* seed took another year to germinate (Figure 1), indicating that there was some dormancy in the seed. A comparable pattern has been reported by Austrheim & Eriksson (2003), who found more seedlings of *A. alpina* the second year than in the first year after sowing.

Germination and mortality may have continued in both species throughout the study (Figure 2), but as the fate of individual seedlings was not followed it is not possible to assess the germination percentages. Based on the amount of seed and results from preliminary germination tests, about 7 seedlings of *D. octopetala* and 12 seedlings of *A. alpina* were expected to emerge in each seeding spot. The average number of *D. octopetala* seedlings was greatest in August 2003 or 4.6 per seeding spot (Figure 2), or 65% of the expected germination. The viability of the *A. alpina* seed was clearly underestimated by the preliminary germination tests. The average number of seedlings per spot reached 17.1 in June 2004, over 140% of the expected germination. The maximum number of seedlings in one seeding spot was 22 for *D. octopetala* and 48 for *A. alpina,* showing a high potential for germination in both species. Previous research has shown varying germination of *D. octopetala* seed, ranging from <10% (Hagen 2002) to >94% (Welker et al. 1997). Germination ability of *D. octopetala* seed is proportional to weight (Welker et al. 1997) which increases with temperature during flowering and seed development (Wookey et al. 1995, Wada 1999). A considerable difference in seed germination can therefore arise depending upon location and weather conditions. The germination of *A. alpina* seed has not been subject to much study, but Austrheim and Eriksson (2003) reported a seedling recruitment exceeding 15% of *A. alpina* seed in disturbed pasture plots.

Seeding success, measured as the proportion of seeding spots with at least one seedling and the average number of seedlings in the seeding spots, varied between microsite types (Figures 1 and 2). *D. octopetala* had a high initial emergence in the gravel microsite, but due to high mortality the seeding success was lowest in that microsite on subsequent observation dates (Figure 2). High emergence in open microsites such as gravel has been observed under similar conditions (Elmarsdottir et al. 2003, Magnússon 1994). Mortality in gravel microsites (Figures 1 and 2) can be attributed to drought in the summer and cryoturbation in winter (Magnússon & Magnússon 1990, Aradottir 1991). *A. alpina* did better than *D. octopetala* in the gravel microsite, especially with regard to filled seeding spots (Figure 1). This could be indicative of a tolerance to the adverse conditions of this microsite, which is supported by the common occurrence of *A. alpina* plants on unstable and sparsely vegetated sites. On the other hand, this difference could simply be the artifact of the time lag between seedling emergence of the two species.

Seedling establishment of both species was good in biological soil crust, which is favourable for seedling establishment of many species (e.g. Bliss & Gold 1999, Aradottir & Arnalds 2001, Elmarsdottir et al. 2003). Among reported effects of biological soil crust formation are surface stabilization, elevated surface temperature, and increased levels of available plant nutrients and water (Belnap 2003).

The number of *D. octopetala* seedlings was initially greater in thin moss cushions than under the shadow of dwarf shrub, but this difference had disappeared by October 2004 (Figure 2). On the other hand, *A. alpina* seedlings were more numerous under dwarf shrub than in the moss microsite on all observations from June 2004. *Empetrum* species may produce bibenzyl with allelopathic effects (Jarevång et al. 1998, Nilsson et al. 2000) but the results of the present study give no reason to suspect phytocide effects on seed or seedlings.

Fertilization had no or a negative effect on seedling establishment in this experiment. On the other hand, fertilization may indirectly facilitate seedling establishment because fertilization on sparsely vegetated ground can stimulate formation of biological soil crust (Elmarsdottir et al. 2003, Aradóttir & Halldórsson 2004), which was favourable for seedling establishment of both species.

Planted cuttings

Seedlings were planted in June 2002. In August 2003, 5% of *D. octopetala* plants and 80% of *A. alpina* plants were still alive (Figure 3). At that point, the survival of *D. octopetala* was not significantly affected by cutting size but fertilization had a negative effect on the survival rate. All *D. octopetala* plants were dead in 2005. On the other hand, fertilization facilitated *A. alpina* plant survival, growth and flowering.

Propagation by cuttings is a common practice in gardening for both species but the cuttings are usually sheltered until rooted. In comparison, the handling of plants in this experiment was rather harsh. Cuttings were prepared in June, when leaves were abundant and flowering in process, and planted directly at an unsheltered site with only a fraction of the normal root system intact. On the first observation date most of the plants had withered leaves and low vigor, indicating effects of shock. This handling, along with the harsh conditions of the site, can explain the high mortality of *D. octopetala* in the first growing season. Continuing mortality, which led to complete extinction of *D. octopetala* in the experiment, was probably due to instability of the surface, caused by cryoturbation and drifting sand. The latter may destroy the evergreen leaves and even damage bark and buds in winter as well as in the growing season. Forbs and grasses have a better chance to survive in such an environment. Research by Hagen (2002) also showed poor survival of *D. octopetala* cuttings in comparison to other Arctic plants including evergreen dwarf shrubs.

The negative effect of fertilization on survival of *D. octopetala* may simply be due to osmotic effects in the rather dry soil. Robinson et al. (1998) also reported decreasing *D. octopetala* cover in fertilized plots in a simulated climate change study at Svalbard, but attributed the damage to delayed hardening caused by the fertilization. Although fertilization did nothing to help establishment of *D. octopetala* cuttings, it greatly benefited *A. alpina* cuttings.

CONCLUSIONS

Our results indicate that sowing can be an effective method for introducing both *Dryas octopetala* and *Alchemilla alpina* into disturbed areas and both species have some characteristics which make them promising candidates for use in restoration. However, for restoration other factors also need to be taken into consideration, such as the possibility of collecting seed on a larger scale. In addition, the establishment of a self-supporting plant population may take some time, considering the slow growth of the *D. octopetala* and uncertain seed germination of *A. alpina*.

A. alpina is hardy when it comes to propagation with cuttings and the method may be effective under certain circumstances. On the other hand, the mortality of *D. octopetala* plants was far too high, in the harsh environment of our experiment, for the method to be feasible. This study is not conclusive, however, about the potential use of *D. octopetala* cuttings, which might still prove a functional choice under different conditions.

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