Growing Endangered Plants to Save Them: Germinating, Propagating, and Restoring Pink Sandverbena

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Abstract

Pink sandverbena is an endangered plant of Pacific Coast beaches. Declining populations and continued threats have lead the Oregon Department of Agriculture to initiate a re-introduction program for the species on the Oregon coast. This program involves documenting the life-history of the species in natural populations to provide a context for evaluating re-introduced populations. Restoration efforts focus on developing seed germination and field propagation techniques. Germination was highest when seeds were removed from the fruit and placed at alternating temperatures and photo periods, but benefits from cold stratification differ from year to year. Direct seeding and transplanting greenhouse-grown stock both show potential for establishing plants in the field, and can produce offspring in experimental plots on dredged sand.

Keywords

rare plant, restoration, re-introduction, coast, Oregon, *Abronia umbellata* ssp. *breviflora*

Introduction

Re-introduction may be essential for endangered plant conservation, especially for species with few remaining wild populations (Falk et al. 1996). Although re-introduction is underway for some plants species, the field is in its infancy. In a bibliography of 600 published accounts and grey literature on plant re-introduction and restoration (Atkinson et al. 1995), only about 10% were detailed re-introduction case histories. Descriptions of effective procedures for rare plant propagation and establishment in the field are crucial for advancing the practice of species re-establishment.

Many sandverbenas (*Abronia* spp.) are rare and prone to extinction, a characteristic that lead Wilson (1972) to call them "disappearing species." Their conservation may rely on restoration of habitat and re-establishment of populations, which in turn, requires information on the behavior of natural populations (Pavlik 1996). Pink sandverbena (*A. umbellata* ssp. *breviflora*) is a rare member of the four-o'clock family (*Nyctaginaceae*) indigenous to the Pacific Coast of North America from British Columbia to northern California. Due to the invasion and subsequent stabilization of foredune systems by European beachgrass (*Ammophila arenaria*) (Wiedemann 1984, Rittenhouse 1994) and disturbance by off road vehicles, pink sandverbena is now restricted to three wild populations on the southern Oregon Coast and perhaps a dozen in California; it is thought to be extirpated from Washington and British Columbia (Gamon et al. 1986). Pink sandverbena is listed by the Oregon Department of Agriculture (ODA) as endangered because of its steady decline in geographic range. It is considered a Species of Concern by the U.S. Fish and Wildlife Service. ODA has initiated a re-introduction program for pink sandverbena in Oregon that involves observations of wild populations, propagation experiments, and field trials before extinction in the wild occurs (Kaye 1995). The primary goal of this report is to document propagation techniques and methods for re-introducing pink sandverbena on dredged sand and natural beaches at Port Orford and elsewhere on the Oregon Coast. A secondary goal is to document the life-history of the species and to identify aspects of population dynamics that are crucial to survival, such as mode of reproduction and life-span of individual plants.

Methods

Study Sites and Species

Pink sandverbena is a low-growing, herbaceous plant from a central taproot. It has deep pink to purple flowers in clusters of 12-20 in leaf axils. Each flower produces a single-seeded fruit (achene), which is broadly 3-5-winged, presumably to promote dispersal (Wilson 1976). It is not clear from the literature whether the plants are annual or perennial (e.g., Hitchcock et al. 1964, Hickman 1992). We observed natural populations on Oregon beaches at Port Orford and Otter Point State Park and performed field experiments at Port Orford and Greggs Creek, about 20 miles south of Port Orford.

Port Orford offered ideal conditions for experimenting with seeding and transplanting methods because the population there produced thousands of seeds. Also, a large, flat area of bare sand was prepared by Army Corps of Engineers contractors by spreading dredged sand 1-m thick over a foredune, thus smothering the European beachgrass and providing a competition-free habitat for pink sandverbena growth (Braun 1991). But because this site was vulnerable to vandalism and occurred on artificially deposited substrate, propagation experiments were repeated on a natural beach at Greggs Creek.

Life History Observations

All pink sandverbena plants greater than 5-cm diameter were mapped and tagged within a 700 m^2 area at Port Orford in June of 1992-1994 to document plant mortality and longevity in a wild population. From 1992 through 1998, all >5-cm plants in this area were measured for number of inflorescences and plant area. The population was situated on dredge material from 1989-91 that was piled high enough to be out of reach of storm waves. At Otter Point, we recorded the total number of plants present from 1994 through 1998 (except 1995) to document temporal variability in population size, and measured plants in August of 1996-98.

Germination Tests

We conducted a series of germination tests to develop an effective germination protocol for producing large numbers of seedlings. Prior to germination tests, seeds from 20 individuals collected in 1991 and 1992 from the Port Orford population were analyzed for viability using tetrazolium tetrachloride. Viability in these tests was nearly 100% for seeds collected in both years (Kaye, unpub. data). In a pilot study, germination of seeds in clipped and unclipped fruits in a factorial combination with wet-stratified (for two months at 4°C) and untreated seeds was attempted on sand in a heated greenhouse (18°C constant temperature). After one month, <5% germination occurred from these treatments (Kaye, unpub. data)

Alternating temperatures and photo periods (20°C, 16 hrs dark/30°C, 8 hrs flourescent light, recommended in Chirco and Turner [1986]) were used to compare germination of seeds with the following treatments: whole fruits on moist sand, clipped fruits on moist sand, dry-stored seeds on moist pads, dry-stored seeds on pads soaked with 0.2% potassium nitrate solution (KNO_3, see Copeland and McDonald [1995]), cold-stratified seeds on moist pads, stratified seeds on pads moistened with KNO_3, dry-stored seeds on moist sand, and dry-stored seeds on sand moistened with KNO_3 solution. Each treatment was replicated 6 times, with 40 seeds per replicate in the first six treatments and 25 seeds per replicate in the last two. Seeds were checked daily for two weeks to record the total number germinated. Germination was defined as 5 mm growth of the radical. We used ANOVA to test for a treatment effect and Tukey's multiple range test to compare means.

In 1997, a smaller test was performed with seeds harvested in 1996 to again test for a stratification effect on germination of seeds removed from fruits. Six replicates of 20 seeds each were stored dry while an identical set of replicates were cold-stratified on moist pads at 4 °C for eight weeks. We used a t-test to compare mean percentage germination.

Field Propagation

Several methods of growing plants in field plots were compared, including seeding and transplanting. Each treatment was replicated 16 times in a randomized block design, duplicated at two sites. Sixteen 2x4-m blocks were placed in a 8x32-m area of level sand on dredged material at Port Orford and a natural beach at Greggs Creek. Each block had room for eight 50x50-cm plots bordered by a 50-cm buffer of untreated area (to reduce edge effects). Both plots were kept clear of competing vegetation by hand-pulling periodically.

In December 1993, pink sandverbena seeds were sown on the surface, buried 3-cm, and buried 10-cm. In each of these treatments, 50 seeds were placed in each plot. Seedlings occurring in these plots were counted in June of 1993 to determine percentage seedling recruitment for each seeding treatment.

Four transplant methods compared the effect of planting date and density on plant growth and fecundity. Plants grown in a greenhouse at OSU from seed collected at Port Orford in 1992 and germinated in February 1993 served as transplants for the following treatments. In April of 1993, sets of one, three and six small plants (5-10 cm diameter, arranged singly or in triangles and hexagons, respectively) were transplanted. In June, 1993, single large (30-40-cm) plants were dug into the plots. The sizes and number of flowers produced by each of these plants were measured in August of 1993. In April of 1994, the number of seedlings established in each 50x50-cm plot and untreated controls were counted to measure the overall fecundity of each seeding and planting treatment.

Data are available from Greggs Creek for 1993 only, because of high tides and sand movement during the fall and winter. One-way ANOVA with a blocked design was used to test for a treatment effect on seedling emergence through June, 1993 from seeding plots (data were log-transformed prior to analysis). The same procedure was used to test for an effect of planting method on plant diameter, inflorescence production (log-transformed), and mortality in October 1993 and August 1994. Finally, we tested for an effect of all seeding and planting methods on second-generation seedling emergence in April 1994.

Results

Natural Population Dynamics and Plant Sizes

The size of natural populations at Port Orford and Otter Point varied substantially from 1992 through 1998 (Table 1). At Port Orford, the population declined from 1992 to 1994, but increased in 1995-1998. At Otter Point, the population grew substantially from around a dozen plants in 1995 and 1996 to over 100 plants in

1997 and 1998. A total of 3.9% of the plants tagged in 1992 at Port Orford survived to reproduce (flower) in 1993, and this percentage increased to 10.5% from 1993 to 1994. However, none of the plants tagged in 1992 survived to 1994. On a natural beach at Otter Point, in contrast, all individuals observed in 1996 and 1997 were killed by winter storms.

Average plant sizes also differed from year to year at both sites. For example, plant diameter ranged from an average of 53.0 cm in June of 1992 to only 9.4 cm in 1998; in general, plant size declined at Port Orford since monitoring began (Table 1). In addition, flowering varied from a high of 153 inflorescences per plant in 1992 to a low of 1.2 in 1994. Although average plant size was usually under a square meter, some individuals reached sizes in excess of 9 m^2 in 1992. At Otter Point, plant diameter and fecundity were typically higher than at Port Orford. Average plant diameter ranged from 36.3 cm to 80.4 cm, and inflorescence production was 19.9 to 113.4 (Table 1). Finally, the Otter point population tended to have a greater proportion of reproductive individuals (0.78-0.84) than Port Orford (0.06-0.77). Observations from these sites should be compared only with caution, however, since Port Orford measurements were made in June, and observations from Otter Point were from August of each year.

Germination Tests

Treating seeds prior to placing them in alternating temperatures had a significant effect on germination (df=7, F=34.73, P<0.0001). Specifically, removing the fruit from the seed increased germination several-fold: only 0.8% and 1.7% of seeds with fruit-husks left intact or clipped germinated, while those with the fruit material removed ranged in germination from 52% to 74% (Figure 1). Differences among the latter treatments, however, were not significant. In contrast, in 1997-tests with seed collected the year before, stratification significantly improved germination from an average of 32.5% to 80.8% (df=10, t=2.23, P<0.0001). In all cases, most seeds germinated within seven days after placement in moist conditions at alternating temperatures.

Field Propagation

Sowing seeds directly in plots in November was an effective method of producing seedlings the following June (Figure 2), but seedling establishment from seeds buried 10-cm was significantly lower than shallow buried or surface sown seeds at Port Orford (df=2, F=22.3, P≥0.0001) and Greggs Creek (df=2, F=8.02, P=0.002). Seed burial at 3-cm produced the best results, with an average of 37.5% emergence at Port Orford and 25.9% at Greggs Creek.

Plant diameter was strongly and significantly affected by planting method at Greggs Creek in 1993 (df=3, F=6.95, P=0.0006), but only marginally so at Port Orford in 1993 (df=3, F=2.66, P=0.059) or 1994 (df=3, F=2.73, P=0.055). At Greggs Creek, plants placed singly and late in the season (June) were larger (mean: 18 cm) by October than April transplants in any density (one, three, or six per plot,

Table 1. Mean (SE) pink sandverbena population size, diameter, fecundity, and proportion reproductive in natural populations at Port Orford in June and Otter Point in August (no Otter Point observations available for 1995; plant sizes were not recorded in 1993 and 1994).

year	population size	diameter (cm)	no. inflorescences	proportion reproductive
Port Orford				
1992	233	53.0 (4.0)	153 (15.5)	0.77
1993	77	32.9 (3.1)	13.5 (4.4)	0.57
1994	42	22.0 (2.0)	1.2 (0.5)	0.31
1995	140	12.6 (1.4)	3.0 (0.9)	0.15
1996	160	28.2 (2.1)	22.7 (3.3)	0.06
1997	145	16.9 (1.2)	7.6 (2.8)	0.41
1998	180	9.4 (0.7)	1.6 (0.5)	0.12
Otter Point				
1993	12	–	–	–
1994	15	–	–	–
1996	9	80.4 (18.0)	113.4 (40.3)	0.78
1997	177	64.5 (8.1)	111.6 (28.9)	0.84
1998	136	36.3 (2.3)	19.9 (2.4)	0.79

mean range: 8.6–9.3 cm). However, average plant diameter was much larger at Port Orford than Greggs Creek in either year (Figure 3, regardless of treatment; mean range: 75.6–115.1 cm). Planting method also had no effect on inflorescence production at Port Orford in 1994 (df=3, F=1.47, P=0.235), where average plants produced 13 to 27 flower clusters.

Plant mortality did not differ among transplant times or densities at either site, even after two growing seasons at Port Orford (df=3, F=0.59-2.03, P=0.123-0.627). Average mortality was less than 7% for all planting methods by October of the first season at Port Orford. By June of 1994, average mortality of 1993 transplants was below 40%. At Greggs Creek, mortality ranged from 12% to 37%, depending on the treatment, by October of 1993. By 1994, however, no plants survived at Greggs Creek.

Recruitment of seedlings into all sowing or transplanting plots in April of 1994 varied substantially among treatments (df=6, F=8.78, P≥0.0001). So many seeds were produced in 1993 that seedlings even emerged in the control plots, where no plants were grown previously (Figure 4). The fewest seedlings occurred in the control and 10-cm seed burial plots (<11 per plot on average), while the 3-cm seed burial plots and the April 1993 transplants (all densities) produced the most (mean range: 61.2–109 per plot). Other treatments produced intermediate numbers of seedlings.

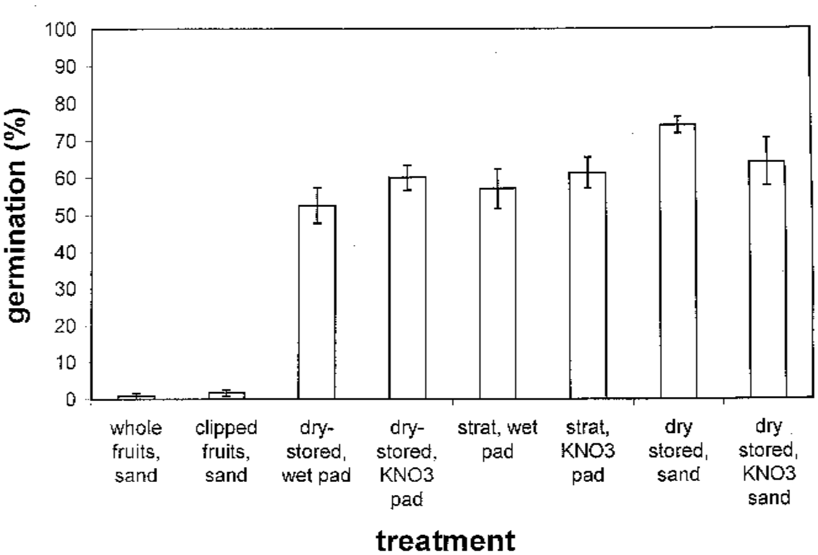

Figure 1. Percentage germination of pink sandverbena seeds in various treatments (error bars represent ±1 SE). See text for explanation of treatments.

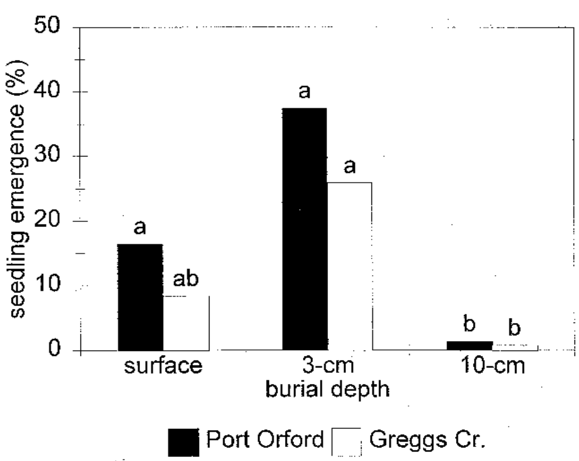

Figure 2. Mean seedling emergence from plots seeded at various depths at Port Orford and Greggs Creek. Bars with the same letters did not differ at the 0.05 level of probability.

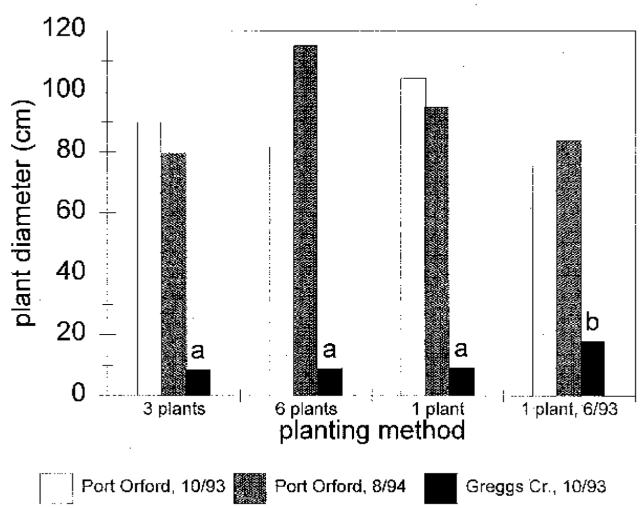

Figure 3. Average diameter of transplants from four planting treatments at Port Orford and Greggs Creek. Bars with the same letters did not differ at the 0.05 level of probability. There was no significant treatment effect for bars without letters.

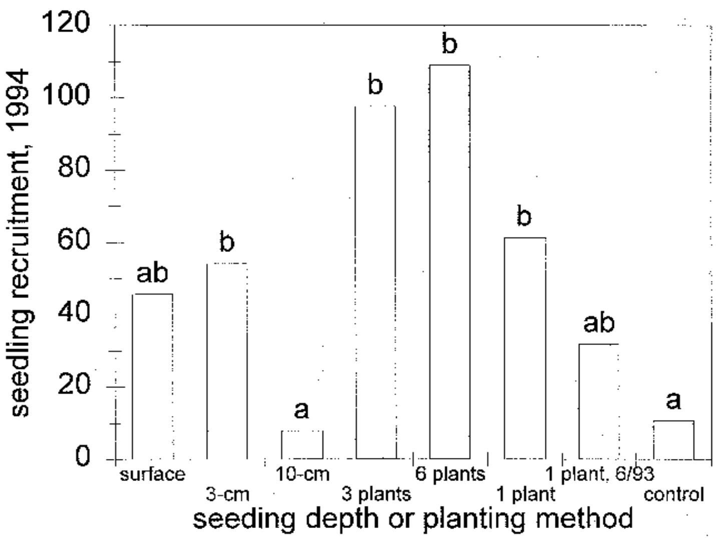

Figure 4. Number of seedlings produced in experimental plots during June 1994. These seedlings were the progeny of experimental plants established from seeds or transplants 10 to 18 months earlier.

Discussion

Pink sandverbena appears to be a facultative perennial that reproduces by seed only. Most individuals observed in this study behaved as annuals, flowering and dying in their first year of growth. However, a small percentage of individuals in the natural population at Port Orford grew and flowered for two years, and transplanted individuals on dredged sand had a high probability of living for two seasons. At Otter Point, however, where the population occurs on a beach, all individuals lived for only one season. Several *Abronia* species are described as being both annual or perennial, or biennial and perennial; perennation of *A. angustifolia* is possible only when individuals occur in highly favorable locations, such as protected micro-sites with high moisture availability (Royce and Cunningham 1982). Such flexibility can improve the ability of pink sandverbena to persist at a given site or disperse seeds to new locations, since individuals that live for more than one season may be very large, produce thousands of fruits, and contribute a large proportion of a population's annual seed production (Kaye, unpub. data). In addition, reintroduction efforts should expect high annual mortality of individual plants; low survival should be considered normal.

Seed germination was greatest when the seed was removed from the fruit. Once that was accomplished, germination of seeds collected in 1992 was not affected by stratification, exposure to KNO_3, or substrate (germination pad or sand), all of which yielded about 50-70% germination in alternating temperatures and photo periods. However, stratification substantially improved germination of seeds collected from Port Orford in 1996. Apparently, germination requirements for this species differ from year to year, possibly due to annual variation in climate during maturation and while fruits are on the ground, prior to collection. Propagation methods for this species may need to be revised periodically to germinate seeds from different years and possibly different locations.

Transplanting and sowing seeds directly into appropriate habitat resulted in short-term plant establishment on dredged material and a natural beach. Late plantings on dredged sand at Port Orford grew as well as early season transplants in 1993 and 1994, and most plants grew vigorously (compared to wild populations). At Greggs Creek, growth was generally poor, probably due to burial by wind-blown sand in late summer, and no plants survived the winter. Clustering plants (one, three, or six plants per plot) also had no effect on plant growth at either location, even though mixed-species clusters have higher growth and survival than plants placed singly for revegetation of mined lands (MacMahon 1987).

Which field-propagation method, transplants or seeding, is best for establishing pink sandverbena? In the long term, it is not clear how well either one will work, but both show potential. Theoretical models suggest that planting small transplants will produce more vigorous populations than sowing an equal number of seeds (Guerrant 1996). However, seeds are much easier to sow than plants are to transplant, so larger numbers of pink sandverbena seeds than plants can be used, possibly outweighing any transplant advantage. In fact, second-season seedling recruitment in this study was generally the same from transplant and seeded plots, with the exception of controls and plots with seeds buried 10-cm (Figure 4). Additional studies are underway at several sites on the Oregon coast to test our ability to establish plants from seed or greenhouse-grown transplants.

Pink sandverbena may be a poor competitor for resources. The Port Orford population declined in number of plants and plant size from 1992 through 1994. During this period, no dredging was conducted at that site, the habitat sustained no large-scale disturbance, and the abundance of competing vegetation, including European beachgrass, increased (Kaye, pers. observation). In 1995, disturbance from dredging was renewed and the population grew. Natural populations of pink sandverbena almost always occur on beaches, below the foredune, possibly because individuals do not compete well with established dune vegetation. In fact, many coastal Abronia species do not compete well in stable sandy areas, occurring instead in ecologically dynamic habitats (Purer 1936, Couch 1941), and acting as pioneer species (Johnson 1985). Dredged sand offered an excellent substrate for short-term plant growth and reproduction. But unless it is disturbed frequently to remove competing vegetation, pink sandverbena may ultimately decline. Our results to date support the hypothesis that pink sandverbena populations require frequent disturbance, either by man or nature, to be viable. Sowing seeds or transplanting greenhouse-grown individuals on beaches may be effective methods for re-introducing pink sandverbena. Sites with high summer winds should be avoided, however, because the plants appear intolerant of burial during the growing season.

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