SEED PROPAGATION



SEED BANKING FOR THE FUTURE

Andrea N. Raven (503) 636-4112 ravena@rdrop.com The Berry Botanic Garden 11505 SW Summerville Avenue Portland, OR 97219

Abstract

The Berry Botanic Garden acts as a regional resource within the plant conservation community by maintaining a seed bank for rare and endangered native plants. Banked seeds represent an "insurance policy:" should wild populations be damaged or disappear, we hope to be able to provide the genetic material to enhance or re-establish those populations. The process of banking involves seed collection, cleaning, drying, packaging, freezing and documentation. Seeds can be withdrawn from the bank for reintroductions or scientific research. To effectively perform our seed banking, we work in cooperation with many individuals and organizations.

Keywords

endangered species, reintroduction, ex situ research, native plants

Introduction

One of the primary missions of The Berry Botanic Garden, located in Portland, Oregon, is to conserve rare and endangered plants native to this region. We serve as a resource for rare plant work in this region by specializing in *ex situ*, or off-site, conservation research. Our *ex situ* work includes rare plant seed storage, germination testing, propagation research, including tissue culture, the maintenance of living collections and common garden experiments (Guerrant 1997). All of these are intended to complement on-site efforts. In addition, we perform *in situ* or on-site research. Our on-site research will not be addressed in this article.

We work in cooperation with many individuals and organizations to carry out our mission. We are a charter member of the Center for Plant Conservation (CPC), which is a national network of 28 botanic gardens and arboreta interested in conserving sensitive species. The Berry Botanic Garden represents the Pacific Northwest region within the CPC network. As such, we work with rare taxa from Oregon, Washington, Idaho and portions of California and Nevada. We also belong to the American Association of Botanic Gardens and Arboreta and the American Association of Museums. We regularly work in partnerships with city, state and federal agencies, and with private conservation groups such as The Nature Conservancy. In addition, we depend heavily

on volunteers and interns.

The heart of our conservation program is the Seed Bank for Rare and Endangered Plants of the Pacific Northwest. The bank was the brainchild of Molly Grothaus, a former board member, and was established in 1983 with a grant from a local foundation (Fred Meyer Charitable Trust). Julie Kierstead Nelson was the first Conservation Director and Seed Bank Curator at Berry.

We consider our seed bank to be an "insurance policy" within an integrated conservation community. With the help of many individuals and organizations, we are working to bring genetically representative samples of this region's rarest and most vulnerable plants into long-term storage and to maintain those seeds. Should a wild population become extirpated, we hope to be able to provide the necessary genetic material and propagation knowledge to re-establish the population. Stored seeds can also be used to enhance damaged populations.

Seed banking by itself does not constitute conservation of a population or taxon. We believe that seed banking is a tool that is part of a coordinated community effort to conserve our native plants. We would caution that the science of storing seeds is still being developed and, even when the proper technology is used, stored seed samples can still be inadequate for reintroductions or other desired uses (for instance, samples sizes can be too small or the genetic mix can be inappropriate). Optimal storage conditions vary for each taxon. It is likely that our procedures are less effective or inappropriate for some taxa.

Seed Banking Procedures

An overview of the process of seed banking is as follows: we collect seeds, document the collection, dry, clean and count seeds, weigh and package seeds, and if resources allow we germinate a portion. Our procedures follow standards developed by the International Board for Plant Genetic Resources (Cromarty et al., 1982; Ellis et al., 1985a and b) and the Center for Plant Conservation (CPC, 1991; Wieland, 1995).

The first issue we address when banking is determining which species to collect. We work in cooperation with the Natural Heritage Programs, an essential resource. The Heritage Programs gather and store population-specific information on rare and sensitive plants for each state. They have generously provided us with detailed summaries on the region's rare plants and we use their data to prioritize plants by level of endangerment. In addition, we work in cooperation with regional land managers (such as the Forest Service, Bureau of Land Management and U.S. Fish and Wildlife Service) and other knowledgeable people to determine which plants they are most interested in banking.

The next issue we address is a determination of appropriate sample sizes. Key questions that must be answered when sampling rare plant populations include the following: how many populations should be sampled per species?, how many individuals should be sampled per population?), how many propagules should be collected per individual? and under what circumstances is a multiyear collection plan indicated? We attempt to balance the need for genetically and geographically representative samples with the possibility of damaging a given population by overcollecting or causing physical damage through trampling. We use collection guidelines established by the Center for Plant Conservation to assess whether we have a genetically representative sample (CPC, 1991). In most cases, the CPC recommends sampling a range of one to five populations per taxon, ten to 50 individuals per population and one to 20 propagules per individual. We choose the appropriate sample sizes for each taxon based on biological and management-related factors (such as the intended use of seeds for reintroduction).

We obtain seeds from a variety of sources. Federal botanists (Bureau of Land Management, Forest Service, U.S. Fish and Wildlife Service) collect over half of our seeds. Berry Garden staff members, interns and volunteers collect a great number of seeds. Other professionals, including university professors and students, Nature Conservancy staff and specially trained Native Plant Society members collect for us as well. All collection is done using the appropriate permits and with landowner permission.

Seeds are rapidly transported to the Garden and immediately placed in the Seed Vault, a walk-in, drying facility

that we had constructed specifically for seed banking. Seed moisture level and storage temperature have been identified as being the main factors influencing long-term seed viability. Currently, the experts are debating over optimal relative humidity (RH) levels and temperatures (Ellis et al., 1991; Smith, 1992; Vertucci and Roos, 1990, 1991, 1993). The internationally accepted procedure has been to dry seeds to equilibrium at 15% RH and 15°C. In recent years, researchers from the USDA National Seed Storage Laboratory in Fort Collins, CO have challenged this, arguing that those conditions dry seeds too much and can reduce seed survivorship and seedling vigor. We have attempted to take a conservative approach and our vault is maintained at a temperature of 15°C with relative humidity at approximately 22%.

The vault is surrounded by 8" of concrete to reduce the threat of fire damage. Air inside the vault is constantly circulated and we use silica gel as a desiccant. We generally allow seeds to dry for a minimum of one month, although the length of time varies depending on seed size, seed oil content and the total amount collected. During the drying period, seeds are removed from the vault and we clean and count them. After drying is completed, we weigh and heat-seal the seeds inside metal foil pouches. The pouches are then placed into a modified chest freezer at -18°C for long-term storage.

Seeds placed in the bank will not live forever. Seeds are periodically checked for viability, and those with low germination must be replaced. Seeds are also removed for other purposes, such as scientific research and reintroduction, and those must be replaced as well. We attempt to use small numbers of seeds for our germination tests, since they are rare. We also use seeds for tissue culture research. Through our tests, we hope to gain information on germination requirements for rare plants, since that information is usually not available. The results of our ongoing tests are available to interested professionals.

Seed Bank: Past, Present and Future

In 1983, we created the bank with 73 accessions of 60 different plants. Currently, the bank contains over 4,000 accessions representing over 280 taxa. Each accession contains seeds from a separate population or individual. Figure 1 shows the increase in taxa (diamonds) and accessions (bars) through time (1998 data not shown). As time passes, it becomes more difficult to add new taxa to the bank and hence, the number of taxa represented has leveled off in recent years. On the other hand, there has been a recent dramatic increase in the number of banked accessions due to a shift in collecting methods. Currently, we collect a greater number of accessions for a given population, each accession representing a single individual. Older accessions were often bulk collections, with one accession representing seeds from many plants collected at one time.

One example of our work with reintroduction involves the Malheur wire lettuce, *Stephanomeria malheurensis* (Asteraceae), one of Oregon's rarest plants. It is an annual and is known from only a single location in eastern Oregon. In 1985 and 1986, wild plants did not appear and the Fish and Wildlife Service contacted Berry.



Figure 1. Cumulative numbers of taxa (diamonds, left scale) and accessions (bars, right scale) since the establishment of the seed bank in 1983.

We grew out hundreds of plants using seeds we received from Dr. Les Gottlieb of Davis, CA, who had performed extensive research on S. malheurensis evolution (Gottlieb 1977, 1978, 1983, 1991). Garden-grown plants were transplanted into the original wirelettuce site in 1987 using an experimental approach (Parenti and Guerrant 1990; Marshall 1995; Guerrant 1996). The transplants were very successful and produced thousands of seeds the first year. Half of the seeds were collected and banked, and half were left at the site in an effort to reestablish the wild population. We were able to bulk up our bank collection of Stephanomeria seeds through this work, which provides a little extra "insurance" for the wild population. We continue today to work with the federal agencies to monitor and care for this population.

After 15 years of collecting and storing seeds, we have acquired millions of seeds representing many taxa. But how do we know when we have "enough" seeds? In the last few years, we have been involved in a focused, intensive effort to evaluate the geographic and genetic representation of each species in our collection. We are in the process of creating maps of wild population locations, using data supplied by the Oregon, Washington, Idaho and California Heritage Programs. We overlay maps of the populations represented in the bank with all known wild population locations. This allows us to easily view the geographic representation for each rare taxon. We then examine the number of seeds we have per population, the number of individual plants represented per population, the age of our accessions and the results of our germination trials. For each rare taxon, we can then evaluate whether additional seeds need to be collected and determine which populations should be sampled or re-sampled. We are currently working to fill in the gaps in the bank that we have identified through our evaluation. We have been able to perform these analyses and significantly enhance the seed bank due to a series of grants from the Institute of Museum and Library Services.

Our collection currently includes all federally endangered and threatened species for Oregon and Washington. We have seeds from over 90% of Oregon's and Washington's federal candidate plants and 68% of federal Species of Concern. We have 90% of Oregon's state endangered and threatened plants and 57% of state candidate taxa. Over half of Washington's endangered taxa are represented in the bank. Currently, we have seeds from only 10 taxa from Idaho, although plans are underway to acquire additional species.

Conclusion

Seed banking and other off-site conservation research are increasingly being used to help conserve rare taxa. The Berry Botanic Garden's *ex situ* work includes rare plant seed storage, germination testing, propagation through tissue culture and traditional horticultural techniques, maintenance of living collections and greenhouse, laboratory and garden experiments. We are able to run our bank and perform effective plant conservation work because we function within a coordinated community that includes federal, state and local agencies, private conservation organizations such as The Nature Conservancy, universities, other botanic gardens and arboreta, native plant societies and our volunteers and interns. We are excited to continue to work toward ensuring the survival of the rare and endangered plants that are our regional natural heritage.

Acknowledgments

Special thanks to all who have contributed to the seed bank since its establishment. Ed Guerrant thoughtfully reviewed this manuscript.

Literature Cited

- Center for Plant Conservation. 1991. Genetic sampling guidelines for conservation collections of rare plants. In Genetics and Conservation of Rare Plants, pp. 225-238. DA Falk and KE Holsinger, editors. Oxford University Press, New York.
- Cromarty, AS, RH Ellis and EH Roberts. 1982. The design of seed storage facilities for genetic conservation. Handbook for Genebanks, No. 1. International Board for Plant Genetic Resources, Rome, Italy.
- Ellis, RH, TD Hong and EH Roberts. 1985a. Handbook of seed technology for genebanks. Volume I. Principles and methodology. Handbooks for Genebanks, No.

2. International Board for Plant Genetic Resources, Rome, Italy.

- Ellis, RH, TD Hong and EH Roberts. 1985b. Handbook of seed technology for genebanks. Volume II. Principles and methodology. Handbooks for Genebanks, No. 2. International Board for Plant Genetic Resources, Rome, Italy.
- Ellis, RH, TD Hong and EH Roberts. 1991. Seed moisture content, storage, viability and vigour (Correspondence). Seed Science Research 1:275-277.
- Guerrant, EO. 1996. Experimental reintroduction of *Stephanomeria malheurensis*. In Restoring Diversity: Strategies for Reintroduction of Endangered Plants, pp. 399-402. DA Falk, CI Millar and M Olwell, editors. Island Press, New York, NY.
- Guerrant, EO. 1997. *Ex situ* conservation and The Berry Botanic Garden. In Conservation and Management of Native Plants and Fungi, pp. 105-115. TN Kaye, A Liston, RM Love, KL Luoma, RJ Meinke and MV Wilson, editors. Native Plant Society of Oregon, Corvallis, OR.
- Gottlieb, LD. 1977. Phenotypic variation in *Stephanomeria exigua* ssp. *coronaria* (Compositae) and its recent derivative species "*malheurensis*." American Journal of Botany 64:873-880.
- Gottlieb, LD. 1978. Allocation, growth rates and gas exchange in seedlings of *Stephanomeria exigua* ssp. *coronaria* and its recent derivative *S. malheurensis*. American Journal of

Botany 65:970-977.

- Gottlieb, LD. 1983. Interference between individuals in pure and mixed cultures of *Stephanomeria malheurensis* and its progenitor. American Journal of Botany 70:276-284.
- Gottlieb, LD. 1991. The malheur wirelettuce: a rare, recently evolved Oregon species. Kalmiopsis (Journal of the Native Plant Society of Oregon) 1:9-13.
- Marshall, KA. 1995. *Stephanomeria malheurensis* Progress Report 1993-1994. United States Department of the Interior, Bureau of Land Management, Burns District Office, Hines, Oregon.
- Parenti, RL and EO Guerrant. 1990. Down but not out: reintroduction of the extirpated Malheur wirelettuce, *Stephanomeria malheurensis*. Endangered Species Update 8:62-63.
- Smith, RD. 1992. Seed storage, temperature and relative humidity. Seed Science Research 2:113-116.
- Vertucci, CW and EE Roos. 1990. Theoretical basis of protocols for seed storage. Plant Physiology 94:1019-1023.
- Vertucci, CW and EE Roos. 1991. Seed moisture content, storage, viability and vigour (Correspondence). Seed Science Research 1:277-279.
- Vertucci, CW and EE Roos. 1993. Theoretical basis of protocols for seed storage. II. The influence of temperature on optimal moisture levels. Seed Science Research 3:201-213.

Weiland, GD. 1995. Guidelines for the management of orthodox seeds. Center for Plant Conservation, St. Louis, MO.