

9

# Seed Germination and Sowing Options

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Seeds of many native species are challenging to germinate. One important thing a grower can do is to learn as much as possible about the life history, ecology, and habitat of the species he or she wishes to grow to understand the processes seeds from each target species go through in nature. Any observations will be valuable when trying to germinate and grow species that have little or no published information available. How seeds are handled, treated, and sown can affect the genetic diversity and the quality of the crop produced. Growers need to balance the desire for uniform crops and schedules with the need to protect the diverse characteristics within species. In this chapter, we discuss seed characteristics, treatments to improve or stimulate germination, and different types of sowing options for seeds.

# **Seed Characteristics**

As discussed in Chapter 8, Collecting, Processing, and Storing Seeds (and shown in figure 8.8), tropical seeds can be divided into four categories related to their longevity and ability to be stored (Hong and Ellis 2002, Kettle and others 2011).

**Viviparous**: seeds that germinate before they are dispersed from the mother plant. The most common examples are some species of mangroves such as *Avicennia* and *Rhizophora* and some tropical legumes.

**Recalcitrant**: seeds that germinate soon after maturation and dispersal from the mother plant, and cannot be dried without losing viability. Most species from the wet, humid tropics have recalcitrant seeds because conditions in these environments are consistently favorable for germination and seedling establishment. Examples of common species with recalcitrant seeds include cacao, mango, longon, and jackfruit.

**Intermediate**: seeds that can germinate immediately but may also survive partial drying without losing viability. For example, papaya seeds that have been dried to 10-percent moisture content have been stored successfully under conditions of 50-percent relative humidity for 6 years without affecting viability (Vozzo 2002). Species that have shown intermediate storage behavior include neem, cinnamon, citrus, mahogany, and coffee.

**Orthodox**: seeds that can be dried without losing viability. These seeds are considered "dormant" and often require specific treatments to encourage germination. Dormant seeds will not germinate immediately upon maturation and dispersal from the mother plant even when ideal environmental conditions exist.

Before attempting to grow a plant, it is important to know the seed germination type because that helps determine the best seed treatments and sowing options for that seed. For orthodox seeds, knowing about the species helps you to provide the best conditions to dissipate, or "break," seed dormancy and achieve good rates of germination.

## **Dormancy in Orthodox Seeds**

Dormancy is an adaptation that ensures seeds will germinate only when environmental conditions are favorable for survival. The conditions necessary to allow seeds to break dormancy and germinate can be highly variable among species, within a species, or among seed sources of the same species. This degree of variability is advantageous because seeds will germinate at different times over a period of days, weeks, months, or even years, ensuring that some offspring will be exposed to favorable environmental conditions for survival. Tropical species inhabiting areas with a strong wet-dry seasonal cycle, arid or semiarid climates, or at high elevations subjected to cold temperatures often have dormant seeds. The degree of dormancy in these species can vary among and within seed lots, between seed crop years, and individuals. Examples of species with dormant seeds include acacias such as Hawai'i's native *Acacia koa*, and pines including the Caribbean's *Pinus caribaea*.

Dormancy may be caused by factors outside (external) or inside (internal) to the seeds. Some species have a combination of external and internal dormancy, a condition known as double dormancy. Knowing the type of seed dormancy is essential for successful propagation.

#### External Seed Dormancy

External seed dormancy may be physical, physicalphysiological, chemical, or mechanical (Baskin and Baskin 1998, 2004). Seeds that have hard, thick seedcoats that physically prevent water or oxygen movement into seeds have physical dormancy. Physical dormancy is the most common seed dormancy type seen in the tropics. Species with external dormancy include many of the legumes (Fabaceae), mallows (Malvaceae), and other tropical species that are adapted to fire, or inhabit arid to semiarid island habitats or areas with pronounced wet-dry seasonal cycles. These seeds normally germinate over a period of several years. Depending on species and habitat, various environmental factors cause these seeds to become permeable over time or during a certain time of year. Seeds that require additional exposure to particular temperatures after they become permeable have physical-physiological dormancy.

The fruits that enclose the seeds cause other forms of external dormancy. Chemical dormancy describes fruits that contain high concentrations of germination inhibitors that prevent spontaneous germination of seeds. Mechanical dormancy describes tough, woody fruit walls that restrict seed germination and is best exemplified by the husks that surround coconuts (*Cocos nucifera*).

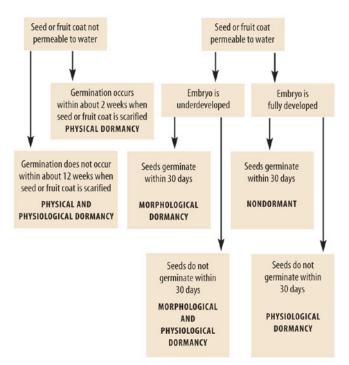
#### Internal Seed Dormancy

Internal dormancy may be morphological, physiological, or both (Baskin and Baskin 1998). Seeds with morphological dormancy have an underdeveloped embryo when dispersed from the mother plant. A period of after-ripening (usually warm and moist conditions) is needed for the embryo to fully mature before the seed is capable of germination. Tropical species that exhibit morphological dormancy are found in the Annonaceae, Dilleniaceae, Magnoliaceae, and Myristicaceae families and in many palm species. Seeds of this type may not germinate for several months to 1 year after sowing. Physiological dormancy is found in some species in arid and semi-arid tropical environments. Seeds are permeable to water, but certain environmental conditions are necessary to modify the internal chemistry of the seed and thus enable germination. Usually a period of cold, moist conditions or holding seeds in dry storage overcomes physiological dormancy.

Seeds with morphological-physiological dormancy usually require a combination of warm and cold conditions, often over an extended period of time, before they are capable of germination.

# Treatments To Overcome Seed Dormancy and Enhance Germination

A variety of seed treatments have been developed in response to the diversity of seed types grown in nurseries. Before treating seeds, be sure to consult available references to see what treatments have been used on that species; see the literature cited at the end of this chapter and the Native Plant Network (http://www.native plantnetwork.org). If no information is available, check references for closely related species. Any personal observations made on the species in the habitat may also provide some clues on how to germinate the seeds. In general, however, the process of treating seeds follows a fairly standard progression outlined in the following



**Figure 9.1**—Key to dormancy types. Knowing the type of seed dormancy is essential to successful seed propagation. Illustration by Jim Marin.



**Figure 9.2**—Seeds that are not cleaned before treatment or sowing can easily mold or be susceptible to serious pathogens such as damping-off disease. Photo by Thomas D. Landis.

sections. Nondormant seeds are planted immediately after collection and cleaning. Intermediate seeds may be stored for several weeks or months in suitable conditions, and then cleaned, fully rehydrated, and sown. For dormant (orthodox) seeds, dormancy must be overcome using one or more of the methods described in this chapter before seeds can be rehydrated, enabling germination. It is essential to determine which type(s) of dormancy the seed has so you can do what is needed to overcome dormancy (figure 9.1). The nursery must determine whether seeds will need to be cleaned, scarified, soaked, stimulated, stratified, and treated in other ways before sowing on a species-by-species basis. The following sections describe the seed treatment options available.

#### Cleaning

Seed cleaning helps prevent diseases in the nursery. Cleaning seeds of bacterial and fungal infestation is especially necessary for species that easily mold (figure 9.2). Often, molding can be related to the most common disease seen in nurseries, damping-off. Seed cleaning is especially important in humid climates and for species that take a long time to germinate. Often, without cleaning, seeds can be lost to pathogens before they are planted in the nursery.

One of the best cleaning methods is to simply soak seeds in a stream of running water for 24 to 48 hours. The running water flushes bacterial and fungal spores from the seeds (James and Genz 1981). This treatment can also be used to satisfy the soaking requirement described in the next section.

Seeds can also be cleaned with several chemicals, some of which also act to stimulate germination. Bleach (5.25 percent sodium hypochlorite) is the most common chemical used. Depending on the species, bleach cleaning solutions range between one part bleach in eight parts water to two parts bleach in three parts water. With most species, treatment duration is 10 minutes or less. Species with very thin seed coats should not be cleaned with bleach. Hydrogen peroxide can be an effective cleanser and can sometimes enhance germination (Narimanov 2000). The usual treatment is one part peroxide in three parts water. Tropical species that benefit from hydrogen peroxide rinses include *Albizia* species and camphor tree seeds (Vozzo 2003).

# Scarification

Seeds with external dormancy require scarification. Scarification is any method of disrupting an impermeable seed coat so that water and oxygen can enter the seeds. In nature, hard seed coats are cracked or softened by fire, extreme temperatures, digestive acids in the stomachs of animals, or by the abrasion of blowing sand. After the seed coat has been disrupted, oxygen and water pass into the seeds and germination can proceed.

Seeds can be scarified many ways. How well the method works depends on the species and the thickness of the seed coats. Whichever method is chosen, it is very important not to damage the endosperm, cotyledons, or embryo during the treatment. Taking time to learn seed anatomy of the



**Figure 9.3**—Mechanical scarification works well with large, easy-to-handle seeds. Great care is needed, however, so that the embryo or cotyledons are not damaged. Koa seed hand scarification (A), scarified lotus seeds (B). Photo A by Craig R. Elevitch, and photo B by Cardno JFNew Native Plant Nursery.



**Figure 9.4**—Hobby-size rock tumblers can be used to scarify seeds and avoid seed destruction that can occur with sulfuric acid or heat scarification. Photo by Tara Luna.

species is helpful. Trying several methods and recording the results will help determine the best method for that species and seed source.

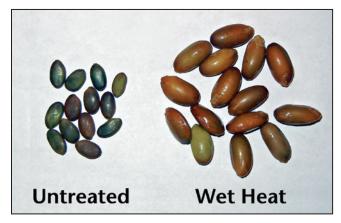
# **Mechanical Scarification**

Mechanical scarification includes filing or nicking seeds by hand and is most often used on large-seeded species such as *Acacia*, *Cassia*, and *Sesbania* (figure 9.3). Be sure to scarify on the side of the seed opposite the embryo. It is often done one seed at a time with a nail clipper. This method is time consuming and requires precision to adequately scarify the seed coat without damaging the internal portions of the seed. Sandpaper can be used on smaller seeded species such as sedges; placing seeds into a shallow wooden box and then rubbing them under a block of wood covered in sandpaper is the simplest technique. Often, however, the degree of scarification achieved with sandpaper can be variable.

Hobby-size rock tumblers can be used to process large batches of seed more quickly than manual mechanical scarification (figure 9.4). Dry tumbling involves placing seeds, a coarse carborundum grit (sold by rock tumbler dealers), and pea gravel in the tumbler and tumbling for several hours or several days. Wet tumbling includes the addition of water to the grit and pea gravel. A benefit of wet tumbling is that seeds are soaked in well-aerated water and chemical inhibitors may be leached from the seed.

## Heat Scarification

Many species, especially those from fire-adapted ecosystems, respond to germination cues from heat. Using either wet or dry heat to scarify the seeds can simulate this response. Using wet heat is an effective method for many small-seeded species because it provides a rapid, uniform treatment that can be assessed within a few hours. Wet-



**Figure 9.5**—Seeds that have been scarified by hot water are visibly larger than untreated seeds because the seed coat has been breached and seeds can then absorb water increasing their size. Photo by Greg Morgenson.

heat treatments are effective for many tropical species including *Acacia*, *Cassia*, *Senna*, *Sesbania*, and *Tamarindus* (Vozzo 2002). Because the thickness of the seed coat may vary among sources, it is wise to dissect a few seeds and examine the thickness of their seed coats to help determine treatment duration. Seeds are added to boiling water for 5 to 10 seconds and then immediately transferred to a vat of cold water so that they cool quickly to prevent embryo damage. Seeds imbibe the cool water for 1 day and are then ready for sowing or for stratification (figure 9.5). Some species cannot tolerate excessively high temperatures, so you may want to heat the water to only 158 °F (70 °C) and monitor your results.

Dry heat is most commonly used on fire-adapted species. Seeds are placed in an oven at temperatures ranging from 175 to 250 °F (80 to 120 °C) from a few minutes to 1 hour, depending on the species. The seed coat cracks open in response to the heat. To avoid damaging seeds, this treatment needs to be monitored closely.

#### **Chemical Scarification**

Sulfuric acid is most commonly used on species with very thick seedcoats and with stony endocarps that surround the embryo (figure 9.6). It has been used on some species of *Acacia, Albizia, Cassia, Leucaena, Parkinsonia,* and *Terminalia* (Vozzo 2002). Treatment length varies with the species and often among seed sources, and it must be carefully monitored because seeds can be destroyed if the treatment is too long. A simple way to monitor the process is by removing seeds at regular intervals and cutting them with a sharp knife. When the seeds are still firm but can be cut fairly easily, the treatment is probably sufficient. Another way is to run a pilot test on a subsample of seeds. Again, remove some seeds periodically and evaluate how well they germinate. After the best

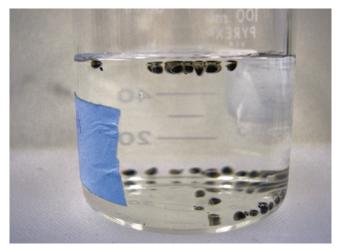
duration is known, the entire seedlot can be treated. Sulfuric acid is very dangerous to handle and requires special equipment, personal protective gear, and proper disposal after use. It should never be poured down sink drains. If acid is being diluted in water, the acid must be added to the water, never add water to acid—when water is added to acid, heat will be released, risking an explosion and other dangers. Some species have thick seed coats but can easily be damaged by sulfuric acid. Instead, citric acid or sodium or calcium hypochlorite baths with longer treatment durations may be used.

The safe use of sulfuric acid requires the following procedures:

- Treat seeds that are dry and at room temperature.
- Require workers to wear safety equipment, including face shield, goggles, thick rubber gloves, and full protective clothing.
- If diluting, add acid to water, never water to acid.
- Immerse seeds in an acid-resistant container, such as a glass, for the duration required.
- Stir seeds carefully in the acid bath; a glass rod works well.
- Immerse the container with seeds and acid in an ice bath to keep temperatures at a safe level for the embryos (this temperature depends on the species; many do not need this step).
- Remove seeds from the acid by slowly pouring the seed-acid solution into a larger volume of cool water, ideally one in which new, fresh water is continually being added.
- Stir seeds during water rinsing to ensure all surfaces are thoroughly rinsed clean.



*Figure 9.6*—Seeds that have been treated with sulfuric acid. Photo by Nancy Shaw.



*Figure 9.7*—Many species of dormant seeds benefit from 1 to several days of water soaking before sowing to fully imbibe seeds and remove any chemical inhibitors within the seeds or on the seed coats. Photo by Brian F. Daley.

# Soaking

After cleaning and scarification, seeds must have exposure to water and oxygen before germination can occur. The standard procedure is to soak seeds in water for 1 to several days until they are fully hydrated (figure 9.7). Hydration can be checked by taking a sample, allowing it to dry until the seed coat is still wet but dull, not glossy, and weighing it. When the weight no longer increases substantially with additional soaking time, the seeds have absorbed sufficient water. Scarified seeds will be more obvious; the seeds will enlarge drastically during the soak. Seeds that only had physical dormancy can be immediately planted. As mentioned previously, running water rinses are effective seed cleaning treatments that reduce the need for fungicides in nurseries (Dumroese and others 1990). Running water soaks also help to remove any chemical inhibitors present on or within the seeds. An aquarium pump can be used to agitate the seeds to improve the cleaning effect and keep the water well aerated. If seeds are not soaked with running water, change the water often (at least a couple of times each day).

# **Germination Stimulators**

Several chemicals are known to increase seed germination. These chemicals are usually applied after seeds are fully hydrated. In general, only seeds with internal dormancy receive this treatment. Germination stimulators include gibberellic acid, ethylene, smoke, and potassium hydroxide.

## **Gibberellic Acid**

Gibberellic acid is the most important plant hormone for the regulation of internal seed dormancy and is often used on seeds with complex internal dormancy and with those species having underdeveloped embryos. In some cases, it has been used to substitute for a warm, moist treatment and to hasten embryo after-ripening. Sandalwood is a species that has been successfully germinated using gibberellic acid. Gibberellic acid can be purchased from horticultural suppliers. Preferred concentrations vary, but most nurseries use 500 to 1,000 parts per million (ppm). High concentrations can cause seeds to germinate, but the resulting seedlings may be of poor quality. Therefore, it is best to experiment with low concentrations first. The following are some guidelines for treating seeds with gibberellic acid:

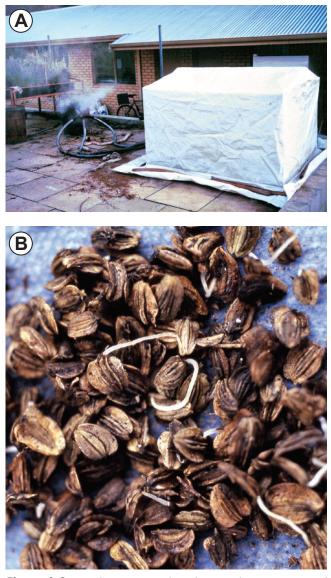
- Gibberellic acid takes a long time to dissolve. It may need constant stirring or you may want to prepare it the day before use.
- Store unused solution away from direct sunlight.
- Cut unbleached coffee filters into squares and fold them diagonally.
- Place gibberellic acid solution evenly into an ice cube tray.
- Place each folded coffee filter containing the seeds into the wells of the tray so that it wicks up the solution.
- After 24 hours, remove and either sow directly or place seeds into fresh coffee filters moistened with distilled water for stratification.

## Ethylene

This gas occurs naturally in plants and is known to stimulate the germination of some species. Ethylene gas is released from ethephon, a commercially available product. Ethephon, used either alone or in combination with gibberellic acid, has enhanced the germination in doum palm (Mousa and others 1998) and may be used for other species inhabiting arid to semi-arid tropical and saline environments. It may inhibit germination in other species, so consult the literature and experiment before using operationally.

## Smoke Treatments

Smoke stimulates germination in many fire-adapted species; for example, species from the California chaparral, longleaf pine communities in Florida, or species from firedependent ecosystems in Australia, South Africa, parts of South America, and the Mediterranean. Smoke especially stimulates seeds that have thin, permeable seed coats that allow entry of smoke into the seeds (Keeley and Fotheringham 1998). Seeds can be treated with smoke fumigation, a method in which smoke is piped into a specially constructed smoke tent containing seeds sown in trays (figure 9.8A),



**Figure 9.8**—Smoke treatments have been used to overcome seed dormancy and enhance germination rates for many native species inhabiting fire-dependent ecosystems. A smoke tent for treating seeds (A). Smoke water-treated seeds of angelica (B). Photo A by Kingsley Dixon, and photo B by Tara Luna.

or with smoke water. Smoke water is an aqueous solution of smoke extract made by burning vegetation and piping the smoke through distilled water or allowing the smoke to infuse into a container of water. Seeds are then soaked in the treated water (figure 9.8B). Conversely, growers can experiment with commercially available smoke products such as liquid smoke or smoke-infused paper discs, or by adding ash to growing media.

Many variables, such as the material used for combustion, the combustion temperature, and the duration of exposure, will need to be determined on a species-by-species basis. Experiments performed by Keeley and Fotheringham (1998) found that the length of exposure to smoke was very important in some species; a 3-minute difference in exposure resulted in seed mortality. Some fire species did not germinate under heat or smoke treatments alone. With some species, seed burial for 1 year or stratification was required in addition to smoke exposure. All these factors can have an effect on germination and should be considered when determining whether to use smoke treatments. Success with this novel treatment will require trials, so good record keeping is critical.

#### Potassium Hydroxide Rinses

Potassium hydroxide has been used to stimulate germination in several native plant species. Optimum concentration varies from 5.3 to 7.6 Molar for 1 to 10 minutes depending on the species; longer soaks at higher concentrations were found to be detrimental (Gao and others 1998).

#### **Other Stimulants**

Potassium nitrate, thiourea, and kinetin have been used to stimulate germination in seeds, although the use of these compounds with tropical native plants is lacking. You may choose to experiment with these compounds on a limited basis with difficult-to-germinate seeds.

#### Temperature and Moisture Treatments

Many seeds with internal dormancy require a moist period at certain temperatures similar to what occurs in the natural habitat before they germinate and grow. Historically, stratification was a temperate zone practice of alternating layers of moist soil and seeds in barrels and allowing these "strata" to be exposed to winter temperatures. Nowadays, stratification is often used more generically to describe the combined use of moisture and any temperature to overcome seed dormancy. We use the term "stratification" to refer to only cold, moist treatments (rare to use in the tropics, but we cover it anyway for nurseries growing highland species). We use the term "warm, moist treatment" instead of "warm, moist stratification."

Some native species with double internal seed dormancy require a combination of a warm, moist treatment for a period of time followed by stratification. Some species or seedlots may require only a few days or weeks of stratification, while others may require several months. As a general rule, it is best to use the maximum recommended treatment. Also keep in mind that what works well at one nursery may not necessarily work well at another nursery because of differences in seed source, handling, processing, cleaning, and storage. An advantage to stratifying seeds of some species is that it can speed up germination and make it more uniform, which is desirable in a container nursery.

#### Warm, Moist Treatments

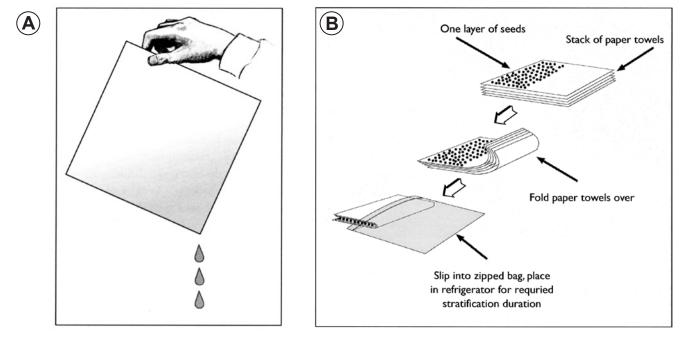
Warm, moist treatment enhances after-ripening of seeds with underdeveloped embryos. Warm, moist treated seeds are kept at temperatures of 72 to 86 °F (22 to 30 °C) for a period of time, usually in moist peat moss, sawdust, or other substrate. Although warm, moist treatments are not commonly used on tropical species, it can be considered for seeds with morphological or physiological seed dormancy.

#### Stratification

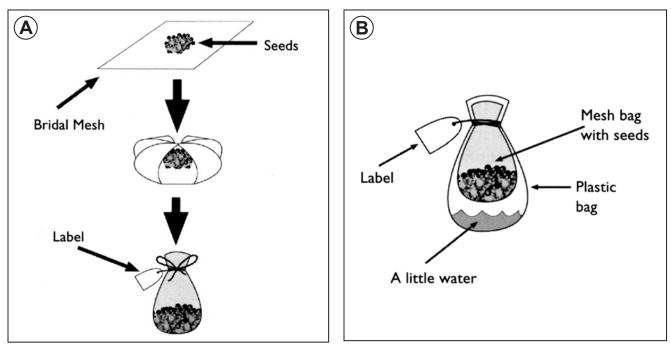
Stratification (cold, moist treatment) is used on seeds with internal dormancy from temperate areas, or high-elevation habitats in tropical regions. Some subtropical species may also benefit from a period of cool, moist stratification. In climates with four seasons, seeds sown in flats or containers in late summer or autumn and left outdoors during winter undergo "natural" stratification. This technique may be preferred if the species has double dormancy (requires both a warm, moist treatment or stratification), requires a very long stratification or requires low temperatures or fluctuating temperatures for a long period of time. Conversely, "artificial" stratification involves placing seeds under refrigeration at 34 to 38 °F (1 to 3 °C) for a period of time. Artificial stratification has several advantages: (1) it allows for a routine check of seeds to ensure they are moist and not moldy, (2) a large number of seeds can be stratified in a small space, and (3) seeds or seedlots that begin to germinate can be removed from the treatment and planted in the nursery as they become available. Artificial stratification is preferred over natural stratification unless the natural treatment provides higher rates of germination.

For artificial stratification of small seedlots and small seeds, seeds can be placed between sheets of moistened paper towels and inserted in an opened plastic bag or sown on a medium in flats with drainage holes. Paper towels need to be moist but not waterlogged, and seeds need to be evenly spread across the moist paper towel to help prevent molding (figure 9.9).

Another technique is "naked" stratification. Most conifer seeds, for example, are stratified in this manner (figure 9.10). Seeds are placed in mesh bags and then soaked in running water as described previously. After the seeds are hydrated, the bag is pulled from the soak, allowed to drip dry for 30 to 90 seconds, and then suspended in a plastic bag. Make sure the seeds are not in contact with standing water in the bag and hang the bags in the refrigerator. If naked seeds need a warm, moist treatment before stratification, it is easiest to first spread the seeds onto moistened paper towels enclosed in large plastic bags. After the warm treatment, the seeds can be returned to the mesh bags for stratification. One other hint: if a particular species or seedlot has a tendency to begin germinating during stratification, surface-dry the seed coats (seeds should be moist and dull, not shiny), and then put the seeds into the bag for refrigeration. The seeds need to still have enough moisture for chemical processes that dissipate dormancy to occur but not enough moisture to allow for germination.



*Figure 9.9*—Small seeds requiring only a few weeks of stratification can be stratified by moistening paper towels and holding by corner to let excess water drain away (A) or placing seeds onto moistened towels inserted into an unopened plastic zippered bag (B). Illustrations from Dumroese and others (1998).



*Figure 9.10*—Naked stratification: After soaking seeds in a mesh bag, allow the bag to drip dry for 1 minute and then suspend the mesh bag in a plastic bag (A). Hang the bag in the refrigerator. Make sure the seeds are not in constant contact with standing water in the plastic bag (B). Illustrations from Dumroese and others (1998).

Many wetland and aquatic species can be treated with naked stratification in water. In general, these species can be easily stratified in Ziploc<sup>®</sup>-type bags filled with water. Insert a soda straw into the bag, ensuring that the end is sticking out of the bag, to allow some oxygen to reach the seeds. Then, seal the rest of the bag securely. Place under refrigeration if in need of a cold, moist stratification period.

# Environmental Factors Influencing Germination

Four environmental factors affect germination: light, water, oxygen, and temperature. All plants have specific germination requirements based on ecological adaptations and the environmental cues that trigger germination for that species.

#### Light

Light quality and duration can influence germination. In nature, seeds of tropical pioneer species require high light levels associated with a canopy gap for germination and establishment, whereas shade tolerant species generally can germinate in very poor light or deep shade. Many small-seeded, tropical native species fall into this category. Thus, pioneer species, such as 'ōhi'a in Hawai'i, with very small dust-like seeds (figure 9.11), require light for germination and fail to germinate even if they are buried only 2 mm deep (Drake 1993). Therefore, these seeds need to be sown on the surface of the medium so they are exposed to light during germination. Other species are conditioned to germinate only if they are buried in the soil. Species requiring darkness to germinate are those that germinate readily under the deep shade of a closed forest canopy. Tropical trees and shrubs with medium to larger sized seeds often require darkness for maximum germination, but shade tolerant vines and herbaceous plants may have smaller seeds. Other species requiring darkness to germinate include some of the species that colonize sand dunes along coastlines.



**Figure 9.11**—In general, small-seeded pioneer species such as 'ōhi'a require light to germinate and must not be buried. Photo by Tara Luna.

#### Water and Oxygen

Water is also important for germination. Overwatering seeds during germination results in reduced levels of oxygen in the medium and promotes tissue breakdown and disease whereas underwatering delays or prevents germination. Therefore, seeds need to be kept evenly moist during germination. Although oxygen is needed for respiratory processes in germinating seeds, some aquatic species may require low oxygen levels for germination. For example, tropical floodplain forest or wetland species naturally germinate during periods of high water inundation and respond positively to low water oxygen levels (Kurbitzky and Ziburski 1994, Vozzo 2002).

#### Temperature

Temperature influences seed germination rate and percentage. Some germination patterns in response to temperature include seeds that require cool temperatures, tolerate cool temperatures, require warm temperatures, and (or) require alternating temperatures (Hartman and others 1997, Vozzo 2002). Species requiring cool temperatures generally germinate below 77 °F (25 °C), which coincides with high elevations in tropical or subtropical regions. Species that tolerate cool temperatures will germinate over a wide range of temperatures from 41 to 86 °F (5 to 30 °C). Many species will not germinate under excessively high temperatures. Most tropical species require warm temperatures and will only germinate if temperatures are above 70 °F (21 °C). In addition, some species germinate better when exposed to alternating temperatures. Alternating temperatures are particularly important with dormant, freshly harvested

seeds. Many tropical tree species germinate to their highest percentages at alternating temperatures of 86/68 °F (30/20 °C) or are provided with at least 10 °F (5.6 °C) difference between the day and night time temperature. Some difficult-to-germinate tropical species may require even greater temperature fluctuations.

## Seed Sowing Methods

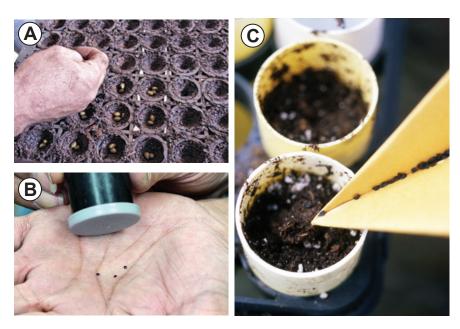
Several sowing techniques have been used for native plants (table 9.1) and are described in the following sections. The process of sowing seeds for nursery production will vary with the species, type of seed, seed quality, and nursery environment.

#### Direct Sowing

Direct sowing is fast, easy, and economical because it minimizes seed handling and labor. It can be mechanized when done on a large scale. For direct sowing to be efficient, the seeds need to be easy to handle, abundant in supply, have simple dormancy treatments, and have a known high-germination rate (figure 9.12). If the direct sowing will be mechanized, seeds must also be uniform in size and shape.

The success of direct seeding depends on the accuracy of seed germination information. Growers must realize that actual seedling emergence may be different from the results of laboratory germination tests that are conducted under ideal environmental conditions. Nursery managers must adjust for this discrepancy based on their own operational experience. Growers should conduct a small germination test of each seedlot to determine the

**Figure 9.12**—Direct sowing works well for seeds that have little or no dormancy (or have been treated to overcome dormancy), are easy to handle and are in abundant supply (A). Simple tools like a film canister (B) or a folded envelope (C) can be used to accurately sow small seeds. Photo A by Douglass F. Jacobs, and photos B and C by Dawn Thomas.



Propagation method	Good method for seeds with the following characteristics	Advantages	Disadvantages
Direct Sowing: Seeds are sown into containers	<ul> <li>Have a known high- percentage germination</li> <li>Are inexpensive</li> <li>Are in abundant supply</li> <li>Have uniform, smooth shapes</li> </ul>	<ul> <li>Fast and easy</li> <li>Economical</li> <li>Minimizes seed handling</li> <li>Seeds are all sown at once</li> </ul>	<ul> <li>Less efficient use of space, seeds, and/or growing medium</li> <li>Causes of poor germination are difficult to track</li> <li>May require thinning and/or consolidation and associated labor costs</li> <li>Not good for large or irregularly shaped seeds</li> </ul>
Planting Germinants: Seeds sprouting or germinating in trays or bags are sown into containers while roots are just beginning to emerge	<ul> <li>Are of unknown viability</li> <li>Are valuable or rare</li> <li>Have unknown germination requirements</li> <li>Germinate during an extended period of time or during strati- fication</li> </ul>	<ul> <li>Efficient use of seeds</li> <li>Efficient use of nursery space</li> <li>Can adjust for unknown seed quality or performance</li> </ul>	<ul> <li>Labor intensive</li> <li>May result in nonuniform crop development</li> <li>Root deformation possible</li> <li>Requires frequent, skilled monitoring</li> </ul>
Transplanting emergents: Seeds are sown into flats or seedbeds for germination; once germinated and leaves appear, seedlings are transplanted to containers	<ul> <li>Are being tested but will not be transplanted to produce a crop</li> <li>Do not respond well to other sowing methods</li> <li>Have long or unknown dormancy</li> <li>Good for trials to observe seed performance</li> </ul>	<ul> <li>Useful with fibrous rooted species</li> <li>Efficient use of seeds</li> <li>Efficient use of nursery space</li> <li>Can adjust for unknown seed quality or performance</li> </ul>	<ul> <li>Not recommended for woody and/or taprooted species because of problems with transplant shock and or root deformation</li> <li>Requires skilled labor</li> </ul>
Miniplug transplants: Seeds are sown directly into small containers. After germination, they are transplanted into larger containers	<ul> <li>Are of unknown quality</li> <li>Are valuable or rare</li> <li>Have unknown germination requirements</li> <li>Have very tiny seeds</li> <li>Will be transplanted into large containers</li> </ul>	<ul> <li>Efficient use of space</li> <li>Uniform crop development</li> <li>Low risk of transplant injury</li> </ul>	<ul> <li>Requires two sets of containers</li> <li>Timing is critical</li> <li>Transplanting by hand is labor intensive</li> </ul>

percentage of germination for each seedlot. Those percentages can then be used to determine the number of seeds to direct sow (see table 9.2). Follow these steps for successful direct sowing:

- Determine how many seeds must germinate to obtain the production target.
- Determine if seeds can be single-sown or will require multiple seeds to reach the production target (see following sections).
- Cleanse and treat seeds as necessary to break dormancy.
- Sow seeds, ideally centering the seeds in each container. Some seeds require a specific orientation for

optimal growth and development; if so, make sure seeds are sown in the correct orientation.

- Depending on the light requirements of the species, cover seeds with the correct amount of mulch.
- Gently water the seeds with a fine watering head to press them into the growing media.

## Multiple-Seed Sowing and Thinning

Sowing more than one seed into each container with the expectation that at least one will germinate is the most common direct-sowing practice. The number of seeds to sow can be calculated based on the seeds'



**Figure 9.13**—Calculating and testing germination rates help reduce costs and problems associated with thinning. Photo by Thomas D. Landis.

expected germination percentage. Two to five seeds are typically sown per container. As a general rule, seeds with less than a 50-percent germination are not recommended for direct sowing because the high density of nonviable seeds in the container may cause disease problems, more containers will need to be thinned, and many plants will be wasted (figure 9.13). Table 9.2 provides general recommendations for the number of seeds to sow per container based on the germination percentage. At some point, adding more seeds per container does not really increase the number of containers with plants (table 9.3) but does drastically increase the number of containers with too many plants and the amount of seed wasted. Sometimes it may be better to single-sow a few containers than thin extra seedlings from many containers. For example, sowing a single seed per container of a seedlot with 85-percent germination yields 15 percent empty containers whereas sowing two seeds per container yields only 2 percent empty containers, but sowing the extra seed requires

**Table 9.2**—For a given seed germination, increasing the number of seeds sown per container increases the number of filled containers. In general, a target of 90- to 95-percent filled containers is reasonable. Adapted from Dumroese and others (1998).

Seed germination percentage	Seeds to sow per container	Percentage of containers with at least one seedling
90 +	1 to 2	90 to 100
80 to 89	2	96 to 99
70 to 79	2	91 to 96
60 to 69	3	94 to 97
50 to 59	4	94 to 97
40 to 49	5	92 to 97

<b>Table 9.3</b> —A sowing example for a seedlot of Acacia koa having a 65-percent germination rate. Assuming 1,000 seedlings are
desired, notice that adding more than three seeds per container really does not improve the number of containers with seedlings and
wastes many seeds. Adapted from Dumroese and others (1998).

Seeds sown per container	Empty containers (%)	Containers with at least one seedling (%)	Seed sown	Seedlings produced
1	35	65	1,000	650
2	12	88	2,000	880
3	4	96	3,000	960
4	1	99	4,000	990
5	0	100	5,000	1,000

thinning 72 percent of the containers. The nursery manager may have been better off, in terms of seed use efficiency and labor, to have simply oversown 10 percent more containers rather than pay for the labor to thin. Therefore, the amount of seeds to sow per container is a function of germination, seed availability, nursery space, thinning costs, and so on.

When more than one seedling germinates in the same container, seedlings compete for light, water, and nutrients. This competition results in lower initial growth rates and requires that seedlings be thinned (clipped, culled, or removed from the container). For this reason, thinning should be done as soon as possible after seedlings emerge. Thinning is a labor-intensive practice and it can damage remaining seedlings if done improperly. Train workers to thin plants carefully and to follow these guidelines:

- Thin germinants as soon as possible; the more developed the root system becomes, the more difficult it is to thin.
- Retain the strongest seedling closest to the center of the container. Thinning is an opportunity for selecting the healthiest seedling while removing inferior plants.
- Pull or cut extra plants. For species with a long, slender taproot at germination (such as pine seedlings), extra seedlings can be easily pulled before they develop secondary roots. For species with vigorous, fibrous root systems, cutting extra plants at the stem with sharp scissors or nipping them with fingernails may be better.
- Discard culled plants into compost or waste.
- Check the remaining seedling and correct any disruptions caused by the thinning process. (For example, if thinning disrupted the mulch, adjust it so the seedling has the best environment possible.)

#### Single-Seed Sowing

Sometimes, particularly when seeds are scarce or costly or are expected to have close to 100-percent germination, single seeds can be directly sown into containers. This practice ensures that every seed has the potential to become a plant and no thinning will be necessary. If a particular number of plants are required, then extra containers are planted, often referred to as "oversowing," to make up for any empty cells. The number of extra containers to sow can be calculated based on the percentage of germination. If a seedlot has only a 78-percent germination, for 100 plants, you must sow at least 28 extra containers (100 desired seedlings/0.78 success rate = 128 containers required). The number of oversown containers may need to be increased to account for seedling losses during the growing cycle. Oversowing works best if the nursery has extra space and is using containers with individual, exchangeable cells because containers with live plants can be consolidated and the extra containers can be removed (see Chapter 7, Containers). Single sowing is efficient because no seeds are wasted and plants that do emerge are not subjected to competition or the stresses of thinning as they are with the multiple-sowing technique. Oversowing wastes potting materials and bench space, however, and consolidating the empty containers is labor-intensive.

# **Sowing Germinants**

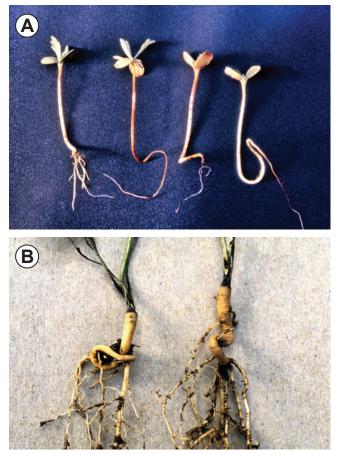
Germinant sowing ("sowing sprouts") is the practice of sowing seeds that are germinating (or sprouting) into the container when their young root emerges (figure 9.14). When done properly, germinant sowing ensures that one viable seed is placed in each container, thereby making efficient use of space and seeds. The resulting seedlings are often larger because they can begin to grow immediately without competition. This technique can be labor-intensive but results in minimal waste of materials and space. Sowing germinants works best for seeds that:

- Are from a rare or valuable seedlot.
- Have a low or unknown germination percentage.
- Are large or irregularly shaped.
- Germinate in stratification.
- Have deep dormancy and germinate over a long period of time.
- Rapidly produce a long root after germination (such as many desert and semidesert species).

Germinant sowing is a relatively simple process. Seeds are treated as necessary, then germinated in trays or bags.



*Figure 9.14*—Germinants must be sown as soon as the radicle emerges from the seed coat. Photo by Tara Luna.



**Figure 9.15**—When planting germinants, seeds must be sown as soon as the radicle is visible and must be oriented correctly when planted. Incorrect orientation leads to severe root deformation in woody species (A, B). Photo A by Thomas D. Landis, and photo B by R. Kasten Dumroese.

Seeds may be spread out between layers of moist paper towels or moist cardboard. Larger seeds are sometimes placed in plastic bags filled with a moist medium such as *Sphagnum* peat moss. Seeds are closely spaced, but far enough apart so that mold does not spread if it forms. Seeds are checked every few days. After seeds begin to germinate, they must be checked daily. Germinated seeds are removed daily and planted directly into growing medium in their containers. Larger seeds can be planted by hand; smaller seeds are often sown using tweezers.

Timing and root orientation are critical when sowing germinants. Seeds need to be sown into containers as soon as the root emerges. The embryonic root, often called a "radicle," needs to be short, ideally no longer than 0.4 in (1 cm). If the radicle becomes too long, it may be difficult to plant without causing root deformation (figure 9.15). Some growers like to prune the radicle of taprooted species before planting to ensure a more fibrous root system. No more than the very tip (up to 0.1 in [3 mm]) is trimmed with



**Figure 9.16**—For transplanting emergents, seeds are handsown in trays that are usually filled with about 2 in (5 cm) of peat moss-vermiculite growing medium. To prevent root deformities, it is important the trays be deep enough that the roots will not touch the bottom before the emergents can be transplanted. Photo by Thomas D. Landis.

clean scissors. The germinating seed is carefully placed in the container with the radicle extending downward. After the seeds are properly planted, the medium needs to be firmed around the root and the seed covered with mulch.

An advantage of planting germinants is that the germination process is more visible to the growers than when seeds are direct sown. Germination timing can be better monitored and the causes of germination problems are easier to track, but because seeds in trays or bags are very close together, a mold or pathogen can contaminate all the seeds if not properly monitored. Labor is required to routinely check for germination, skill is required to achieve proper planting orientation of the seeds, and planting must be done in a timely fashion. Because germinants may emerge over several weeks or longer for some species, crop development will be more variable and require special cultural treatments.

# **Transplanting Emergents**

Transplanting emergents ("pricking out") is a practice for germinating seeds in a small area. Seeds are hand-sown in shallow trays that are usually filled with about 2 in (5 cm) of peat moss-vermiculite (or similar) growing medium (figure 9.16). Soon after the seeds germinate, they are "pricked out" of the tray and transplanted into a container. This technique is not recommended for woody plants and other taprooted species, because root problems often result.

Transplanting emergents works best when-

• Species have a fibrous root system that recovers well from transplanting (herbaceous forbs without a taproot, grasses, sedges, and rushes).

- Tests or trials are being used to observe seed treatments, germination timing or percentage, early growth rate or other early developmental issues.
- Seeds are too small or fragile to be sown by any other method.
- Seeds have very complex dormancy or germinate over an extended period of time.
- Limited nursery growing space makes direct seeding uneconomical.
- Timing to transplant emergents is scheduled promptly.

Some key disadvantages include the following-

- Disease potential is high in densely planted trays.
- Root orientation and timing is critical; root malformations and other problems can result if neglected.
- Transplanting is skill and labor intensive.

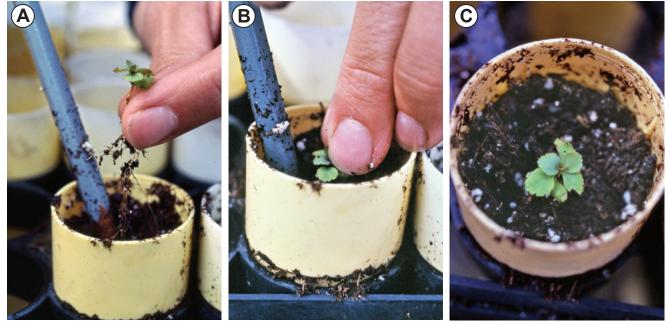
Training, care, good timing, and proper technique is required to prepare seedling trays, sow the seeds, and to transplant emergents properly. As with many other nursery functions, some trial and error occurs in finding the medium mixture and tray depth that works best for each species. Larger seeds are scattered by hand over the surface of the moistened medium, or place in indentations in the medium. Smaller seeds can be sown with a salt shaker with enlarged holes. Sown seeds are then covered with a light application of fine-textured mulch or medium, irrigated, and placed in a



**Figure 9.17**—Root and shoot development at various stages after germination of the Caribbean coastal shrub, Chrysobalanus icaco. The large-seeded species is easily transplanted at these stages. Photo by Brian F. Daley.

favorable environment for germination. Although the exact size or age to transplant the germinating seedlings varies by species, it is usually done at the primary leaf stage (after cotyledons emerge) and well before root systems reach the bottom of the seed tray (figure 9.17).

Emergents are carefully removed from the tray, usually by gently loosening the medium around them (figure 9.18A). A small hole is made in the medium of the container and the germinant is carefully transplanted, ensuring proper root orientation (figure 9.18B). Some species benefit from root pruning before transplanting. The potting medium is then firmed around the root and stem (figure 9.18C).



**Figure 9.18**—Transplanting emergents works well for fibrous-rooted shrubs, forbs, and grasses. Great care must be taken to lift the emergent from the pricking out tray without damaging the roots (A) and to carefully and properly transplant it into the new container filled with moistened growing media (B, C). Photos by Tara Luna.

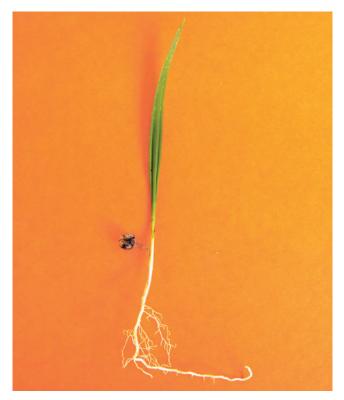
#### Seed Germination and Sowing Options

When timed incorrectly or done improperly, especially on taprooted woody species, transplanting emergents can produce a "J-root" or kink in the seedling stem or root (figure 9.19). These malformations can cause mechanical weakness, poor growth in the nursery and later in the field, and mortality after outplanting. Therefore, unless no other sowing method works, transplanting emergents of woody plants is discouraged.

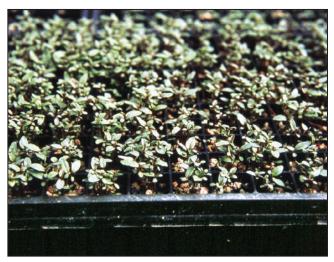
# **Transplanting Plugs**

Small-volume containers, such as miniplugs (figure 9.20) or expanded peat pellets, in which seeds are direct sown (see Chapter 7, Containers) can be transplanted into a larger container after the seedlings are well established. Transplanting small plugs has a number of benefits. The small plug container preserves healthy root form because damage to roots during transplanting is eliminated. Planting small plugs also makes efficient use of growing space. Large numbers of small plugs can be started in a very small area and managed intensively during germination and early growth.

Plants in miniplug containers must have a firm enough root plug to hold the plug together and withstand the



**Figure 9.19**—Transplant emergents early to avoid root malformation. This palm seedling was permitted to get too large in the tray, causing the root to grow at a 90° angle. This root malformation will cause poor growth and performance later in the plant's life. Photo by Brian F. Daley.



**Figure 9.20**—Miniplugs are a viable option for growing seedlings that will later be transplanted to a larger container. Miniplugs work very well with species with very tiny seeds. Photo by Tara Luna.

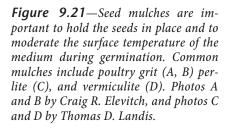
transplanting process, but they must not have so many roots that they are rootbound or the roots may become deformed after transplanting. If peat pellets are used, too few roots are not a problem because the entire pellet can be transplanted. A hole large enough to accept the plug is made in the medium of the larger container, and the small plug-grown seedling is carefully inserted. Planters need to ensure that the roots go straight down and are not deformed during transplanting. The medium is gently firmed around the root system, mulch is applied, and the plant is watered.

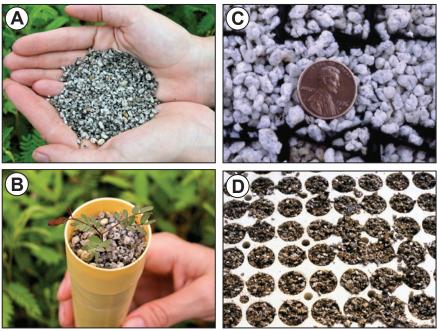
Transplanting small plugs is labor intensive and requires skill. In some arid, windy areas, small plugs are not practical because they dry out too quickly between waterings. Before investing in small plugs on a large scale, a small trial is advised.

# Seed Coverings (Mulch)

Regardless of the seed sowing method, a seed cover or "mulch" is recommended to create an optimal environment for germinating seeds. The only exception is for species that require light to germinate. Mulch is usually a light-colored, nonorganic material spread thinly over the seeds. Examples of mulches include granite grit (such as poultry grit) (figures 9.21A, 9.21B), pumice, perlite (figure 9.21C), coarse sand, or vermiculite (figure 9.21D). When properly applied, mulches—

• Create an ideal "moist but not saturated" environment around germinating seeds by making a break in the texture of the potting medium (water will not move from the medium into the mulch).





- Keep seeds in place. This practice improves contact with the medium and minimizes the number of seeds washed out of the containers by irrigation or rainfall.
- Reflect heat when mulches are light colored, so seeds do not get too hot on bright, sunny days.
- Reduce the development of moss, algae, and liverworts (figure 9.22).

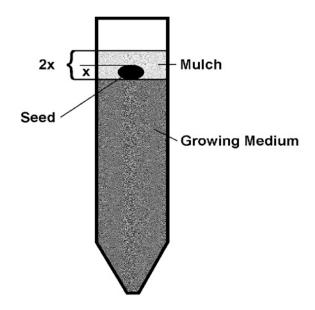
The recommended depth of the seed covering varies by species; a general rule is to cover the seed twice as deep as the seed is wide. If mulch is too shallow, seeds may float away in the irrigation water. If the mulch is too deep, small plants may not be able to emerge above it (figure 9.23).

Seeds requiring light need to be left uncovered. Very small seeds need to be left uncovered or barely covered with



**Figure 9.22**—Mulches help to prevent the development of mosses and liverworts, which can compete with the seedling. Photo by Thomas D. Landis.

a fine-textured material such as fine-grade perlite or milled *Sphagnum* peat moss. Uncovered and barely covered seeds must be misted frequently to prevent them from drying out. After light-requiring and light-sensitive species have emerged and are well established, mulch can be applied to prevent moss and liverwort growth and to help keep the medium moist.



**Figure 9.23**—A general rule of thumb for covering seeds with mulch is to cover the seed twice as deep as the seed is wide. Species requiring light for germination should never be covered with mulch, although mulch can be added after germination to reduce the growth of moss, liverworts, and weeds. Illustration by Jim Marin.

#### Germinating Fern Spores

Ferns (figure 9.24) and fern allies (moonworts, mosses, and horsetails) differ from seed plants in that they produce spores instead of seeds. An understanding of the life cycle of ferns is essential for successful fern propagation in nurseries.

Ferns have two life stages—the gametophyte and the sporophyte—the latter being the spore-producing fern plant with which we are all familiar. The sporangia (sporebearing structures) are variously placed on the lower surface of the leaves and occur in clusters known as sori. In many species, the sori are covered by specialized outgrowths of the leaf, known as the indusium, which lifts and shrivels when spores are ripe. A specialized layer of cells on the stalks of the spores, known as the annuli, contract and expand and the mature spores are disseminated with a catapult-like discharge.

After the spores disseminate, they germinate upon contact with a suitably moist substrate. Spore germination results in the gametophyte, which begins development as a small, pale-green, algae-like chain of cells known as the germ filament. Develop-



*Figure 9.24*—*Native Hawaiian fern, a* Sadleria *species. Photo by J.B. Friday.* 

ment continues into a flat, heart-shaped structure called the prothallus. Slender holdfasts, known as rhizoids, develop on the lower surface of the prothallus. The reproductive structures, the antheridia (male) and the archegonia (female), develop on the lower surface of the prothallus. Antheridia usually appear before the archegonia, mostly near the rhizoids. Archegonia appear near the notch of the prothallus.

Water must be present for the sperm to swim from the antheridia to the eggs in the archegonia. After fertilization, the young sporophyte receives its nutrients from the gametophyte via a foot-like structure. Further development is rapid and, after the sporophyte achieves a level of photosynthesis sufficient to maintain itself, the gametophyte disintegrates. The sporophyte completes the life cycle when it grows into a mature fern plant and produces spores.

#### Fern Propagation

When propagating ferns, maintaining a high level of sanitation during all phases of fern development is of the utmost importance. To collect fern spores, find fern fronds with ripe (dark brown, black, or gold) sori. Cut a piece off the frond. Lay each frond piece in a paper envelope, paper bag, or between two sheets of paper. Wait 24 to 48 hours for the spores to drop onto the paper, then collect (Lilleeng-Rosenberger 2005). Removal of nonspore material reduces the chances of contamination by fungi, algae, or bacteria during spore germination. Spores can be sterilized by adding them to a 2- to 5-percent bleach solution with a small drop of wetting agent, which prevents the spores from sticking together. Soak for a few seconds to 1 minute. Collect spores on filter paper and rinse with distilled water thoroughly for 2 minutes (Hoshizaki and Moran 2001).

Germinate spores using sterilized propagation flats that are at least 2 in (5.2 cm) deep with drainage holes. You can use peat cubes, peat pellets, or a sterilized clay brick placed within an airtight, clear plastic shoebox container. Any sterilized commercial growing mix without added fertilizers can also be used. Moisten the medium thoroughly with distilled water before sowing.

Spores can be sown by spraying them on the medium surface with an atomizer, or by delivering small amounts of spores through a syringe with distilled water. Another option is dip a cotton swab into an envelope filled with spores, and then apply the spores to the growing media using the cotton swab.

After sowing, irrigate with distilled water and cover immediately with a clear plastic lid to seal in moisture and to prevent fungal contamination. Place flats under 150- to 500-foot candles of light. If you are using artificial light (cool, white fluorescent), leave lights on for 8 to 24 hours per day. Optimum spore germination temperature is 68 to 86 °F (20 to 28 °C). Try to maintain a constant germination temperature to avoid excessive condensation in the sealed flats. Distilled water needs to be applied when the medium begins to dry slightly on the surface. Flats also need to be closely monitored for any fungal contamination. Spores germinate 10 to 20 days after sowing. If additional irrigation is needed, water only with distilled water delivered through a sterile mist bottle or by placing flats in trays filled with distilled water.

The presence of mold may require special treatments. Stop overhead watering. Make sure that water is not dripping excessively onto plants from condensation. Remove mold and at least 12 mm (0.5 in) of plant tissue and medium beyond the infected area. Apply a mild fungicide labeled for ferns if infection continues.

Shortly after spore germination, the thread-like germ filament can be seen with the aid of a microscope. In general, prothalli become visible 20 days after sowing. The prothalli continue to grow for up to 10 weeks before the reproductive structures, the antheridia and archegonia, become evident on the under surface of the prothallus. These structures can be seen with a microscope when sampling a few prothalli from a tray. After these structures appear, it is important to maintain a thin film of distilled water over the surface of the prothalli. It is very important to keep the germination surfaces evenly moist at all times.

The clear plastic lid is removed from the container when the antheridia have withered and disappeared, usually 4 weeks after their initial appearance. Flats are then transferred from under indoor lights to a shaded greenhouse. The young fern plants (sporophytes) with true leaves and a developing root system will appear sometime after fertilization; from a few weeks to a few months. They can then be transplanted into individual containers.

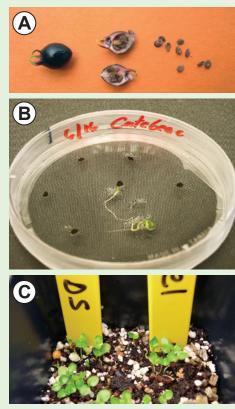
# Try Different Sowing Techniques and Keep Detailed Records

Native plant growers often work with seeds of species that have not yet been propagated in nurseries. Little literature or experience is available to answer questions about seed dormancy-breaking requirements, environmental requirements, germination percentages, and other factors. Understanding the biology and ecology of tropical plants will provide important clues on how to overcome seed dormancy (if any) and provide the correct environmental conditions needed for germination.

It is important to develop a good recordkeeping system to refine and improve results over time and prevent the loss of valuable information. Keep details on the general information of the species, seedlot, seed treatments, and resulting germination. To improve propagation results, use these details to develop and refine propagation protocols, as described in Chapter 4, Crop Planning: Propagation Protocols, Schedules, and Records.



*Figure 9.25*—Small trials of seed germination and sowing options will help you discover the most effective approach for different species. Photo by Brian F. Daley.



**Figure 9.26**—Seeds of Catesbaea melanocarpa shown here with a fruit, a fruit in cross section, and with cleansed seeds (A) were evaluated in sterile dishes (B) to develop a successful germination protocol for this endangered species (C). Photos by Brian F. Daley.

# Trials To Develop a Successful Propagation Protocol for an Endangered Species

*Catesbaea melanocarpa* of Puerto Rico and the U.S. Virgin Islands is considered federally endangered throughout its range. The seeds (figure 9.26A) are small and challenging to work with.

A few individuals of Catesbaea melanocarpa were discovered in a pasture after a wildfire burned grass and underbrush that was obscuring them. With so few plants remaining and in danger of future fires, propagation by seed became a priority. After months of monitoring, a dozen or so ripe fruit were collected, each containing an average of eight small, flat seeds. The seeds were too small to be sown in traditional seed trays and monitored effectively. Due to their small size and scarcity, researchers at the University of the Virgin Islands decided to treat the seeds similar to the way they handle orchid seeds. Sterile dishes of sucrose rich agar (gel) were prepared and 6-8 seeds were placed in each of 10 dishes (figure 9.26B). This intensive germination method allowed for daily observations on every seed collected. Less than one-half of the seeds germinated, and later dissection revealed the nongerminated seeds were smaller than the germinated seed and did not have embryos. When the germinants produced adult leaves, the entire dish was transplanted into plant pots with a mix of ProMix (peat, perlite, and vermiculite blend) and coarse sand. More than 25 percent of the plants desiccated and died shortly after transplanting. The rest of the plants established, but grew slowly. Even with the addition of sand, the media seemed too coarse for the tiny plants.

Later in the season, more fruit were collected. The researchers now knew that one-half of the seeds were likely viable, but that the smaller, malformed seeds could be discarded because they do not have embryos. Researchers added vermiculite to the growing medium mix and sifted it through a screen. Seeds were sown directly into the new, fine medium and germinated in trays in the greenhouse with 80 percent success (figure 9.26C). The second set of seedlings grew more vigorously and did not suffer the transplant shock that killed plants in the first trial.

The first trial led to discoveries about the seeds' viability and identified problems with the growing medium and transplant shock. The second trial used this information and resulted in high germination rates and healthy seedlings of a federally endangered plant in the greenhouse. Because growers have a number of options for sowing seeds, it is a good idea to do small trials of several of the methods described in this chapter (figure 9.25). See Chapter 20, Discovering Ways to Improve Nursery Practices and Plant Quality, for proper ways of conducting trials. Although several methods may "work"—that is, result in a viable plant produced—the question during the trials should be: Which method is optimal? Trials will help you decide how to answer this question yourself.

# Larger Role of Nurseries in Conservation of Species

Each seed is a link between the evolutionary processes of the past and the potential for future adaptation (Flores 2002). While you clean, treat, and germinate seeds to grow your crops, be mindful how your actions affect that species genetic diversity and ability to adapt to the future. Do your best to maintain as much genetic diversity as possible within the species you propagate.

Nurseries are often involved in genetic selection beyond seed collection practices, intentionally or not. For example, production schedules may cause growers to favor faster germinating over slower germinating individuals of the same species, although the quality of the resulting plant would be similar. For some species, the earliest sprouters may be the healthiest. But in other cases, sprouting later may be an adaptive trait; one that could be selected out accidentally by nursery practices. Following up with plant performance in the field can reveal if, in fact, the slower germinating individuals grow well. If so, no reason exists to select these individuals out with nursery practices, and every reason to keep their traits in the gene pool. In this example, simply planting the slower sprouting individuals, as well as the fast ones, could protect diversity. This example is just one that shows how different steps in collecting, storing, germinating, and sowing seeds can affect subsequent plant genetics. The desire for uniform crop size and standardized schedule must be balanced with the need to protect and perpetuate species and genetic diversity.

Nurseries may also have an increasing role in the conservation of tropical species by helping protect and restore genetic diversity of recalcitrant species (species whose seeds do not store well). You are probably familiar with the concept of "seed banks"—seed storage facilities used as reserves to protect and restore species in case their habitats are threatened. Seed banks are also used for some traditional food crops that have become rare with conventional agriculture. Seed banks work well for orthodox species, whose seeds can store for many years and remain viable. In tropical ecosystems, however, recalcitrant species are as numerous and as important as orthodox species. Because of this distinction, many tropical species cannot be "banked" in conventional seed banks. Instead, nurseries and seedling propagation efforts will play key roles in any efforts to conserve and restore tropical recalcitrant species (Kettle and others 2011).

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