Handling

I. Drying and Extracting

A. Introduction

Like agricultural seeds, many tree fruits dry as they mature, and the seeds are extracted best at low moisture contents. Other tree seeds are still very moist at maturity, and special considerations are needed for extraction. No matter what type of fruit is involved, however, the objective of extraction is to obtain the maximum amount of seeds in the best physiological condition in an economically efficient operation. During extraction, seed quality can be greatly reduced by excessively heating the fruit to force opening or by extracting by hand or machine.

B. Objectives

1. Recognize potential problems of seed extraction related to the type of fruit.
2. Identify the basic techniques of tree seed drying and extraction.

C. Key Points

The following points are essential to seed drying and extracting:

1. For species that require drying, excessive heat in the presence of high moisture content can be deadly.
2. Seed damage can occur during mechanical separation.
3. Good training of workers is essential.
4. Extraction strategy depends on the type of fruit involved.

D. Multiseed Fruits

Multiseed fruits include pods; moist, fleshy fruits; cones and capsules; and other multiple fruits. The steps in drying and extracting seeds are:

1. **For pods, Leguminosae**
   a. Dry the pods. Solar drying is fine, but supplemental heating can be used also.
   b. Thresh manually by:
      (1) Flailing with poles
      (2) Crushing by trampling
      (3) Hitting with heavy mallets
   c. Thresh mechanically with:
      (1) Slow, rotating drums (cement mixer)
      (2) CSIRO flailing thresher (Willan 1985)
      (3) Dybvig macerator
      (4) Hammer mills
   d. Use a series of steps for difficult species (e.g., *Prosopis):* break the pods open; soak in 0.1 N hydrochloric acid for 24 hours, wash and dry, and pound the dried material with a hammer. For species that have gummy material in the pods (e.g., *P cineraria):* break the pods and run them through a coarse meat grinder to extract the seeds; or break up the pods, redry, and run them through the thresher again.
2. **For moist, fleshy fruits—In** general, pulp removal will help germination. But, if fleshy coverings are thin, removal may not be required (e.g., *Vitex paruiflora).*
   a. Start quickly to avoid fermentation. (Fermentation may help some species; e.g., *Maclura pomifera* in the United States.) If this is not possible, spread in thin layers and stir occasionally.
   b. Soak the fruits in water until the pulp is soft if the fruits cannot be squeezed away easily with the fingers. The pulp must be soft for complete removal. Change the soak water to avoid fermentation.
   c. Extract with macerators, mixers, coffee depulpers (e.g., *Gmelina arborea),* feed grinders, hammer mills, etc.; use anything that can tear up the fruits without hurting the seeds.
   d. Run small fruits (e.g., *Rubus* and *Morus)* in blenders at slow speeds with lots of water to extract the seeds.
   e. Use a high-pressure stream of water against mesh bags of fruit (e.g., *Prunus* and *Vitis).*
   f. Float pulp fragments and small underdeveloped seeds away with running water.
3. **For cones, capsules, and other multiple fruits—Drying** for extraction assumes successful predrying.
   a. Air-dry on flat surfaces.
      (1) Canvas is best for large quantities. Use a tarpaulin (5 by 7 m or 5 by 10 m) to handle up to 12 to 16 hectoliters of *Pinus caribaea* cones in a layer one cone thick. In good drying weather, the cones will open in 3 days. *Pinus caribaea* should yield 4 to 6 kg of pine seeds from 16 hectoliters of cones. Agitate the cones every 2 to 3 hours to speed drying. Double the tarpaulin over to cover the cones at night. Remove seeds that have dropped out each morning. There will be some problems if it rains during drying (common in Central America).
      (2) Use screen trays for smaller lots, such as single-tree collections. Use larger trays to handle nearly 1 hec-

toliter at a time. Spread one cone deep, stir, and protect from rain. Elevate the trays to speed drying; catch extracted seeds on a sheet or cloth.

(3) Plastic sheets are not strong enough for *Pinus* cones (1 hL of cones = 35 kg). Plastic also encourages condensation when the cones are covered. Plastic sheets can be used indoors for lighter fruits, (e.g., *Liquidambar, Alnus, Casuarina,* or *Populus).*

(4) Some common principles: protect fruits from rain and predators, spread them thin and stir frequently, and collect seeds on the drying surface as they fall. Protection from rain is not needed if the fruits will be artificially dried later.

(5) Dry some species under shade (e.g., *Hopea* spp., *Triplochiton schelroxylon,* and *Pinus oocarpa);* dry others in direct sunlight (e.g., *P. caribaea* and *P. elliottii).*

b. Use solar kilns.

(1) One simple type of solar kiln is clear polyethylene stretched over the top of screen trays; this method is common in east Africa.

(2) Solar heat storage units are more sophisticated. Small buildings can contain solar panels and heat storage facilities, such as water barrels. Plans for one that can handle 15 to 18 hectoliters of cones at a time are available. Such units are good for easy-to-open cones of *Pinus,* such as *P strobus,* which open in 1 week with good sun.

c. Use heated kilns. These kilns are most efficient for large quantities of cones, but capital investment and fuel costs may be too high. There are several types of heated kilns:

(1) Progressive kilns—Cone containers move along a gradient of increasing temperature inside the kiln. Because they are slow, they are not used much. They can be vertical or horizontal. One company in the United States has a tunnel progressive kiln with multitrayed, wheeled carts and computerized environmental controls. A few have been installed in Canada and the Northwest United States.

(2) Large-batch kilns — Large, heated chambers in which trays hold cones and rotate positions. Typical drying capacity is 300 to 350 hectoliters per day. There is no provision for small lots, and fuel costs are high. Natural gas is used in the United States, but almost all these kilns have been phased out.

(3) Small-batch kilns—Wire-bottom drawers hold about one-third hectoliter of cones each. They are inserted into a chamber in which a fan and gas-fired heater push heated air through the drawers. The standard kiln holds about 16 hectoliters at a time, but the design is modular for possible expansion. One big advantage is that small lots can be kept separate.

(4) Stack-tray system — Developed in the Southern United States for *Pinus,* it is a flexible and portable system. Wooden trays with perforated sheet-metal bottoms (not wire screens) fit together in stacks of six; eight stacks are heated by one heating system. Heated air is blown up through the trays from a bottom plenum and recirculated to the heater. One such unit can hold 100 hectoliters at a time and should process 7,000 hectoliters in a season. Fuel costs are high, but flexibility is an advantage.

(5) Tumbler driers — New cylindrical batch kilns that rotate while drying to remove seeds as soon as possible. Humidity sensors and humidifiers allow moisture and temperature control. Many sizes are available to fit individual needs.

(6) Other batch kilns —Many local designs are available. The Robbins kiln, built in Honduras, seems to be a good, low-cost, forced-draft kiln. It has a capacity of 32 hectoliters, a drying period of 12 to 18 hours under good conditions, and uses old cones or wood as furnace fuel.

d. Set temperature and humidity parameters. The object is to remove moisture; high temperatures do this by creating a greater vapor pressure gradient. For conifers, 29 to 50 °C may be used, but initial temperatures should always be on the lower end of the scale. Pine cones of the Southern United States are predried to well below 50-percent cone moisture before going into the kiln. Good predrying decreases cone moisture to 25 percent. Cones of *P. contorta* should be predried to 20-percent moisture. Kiln temperatures should be 43 to 49 °C, for tough seeds, such as *P taeda* and *P elliottii,* but to 35.°C for "sensitive" seeds, suchN.s *P.'itrobus* and *P. palustris.* Cones should be dried to 10-percent moisture content in the kiln for best opening.

(1) Serotinous cones (e.g., *P banksiana, P. patula,* and *R clausa)* need 63 to 65 °C or a 15-second dip in boiling water to break the resin bonds before going into the kiln. Live steam can also be used to open these cones. *Pinus contorta* cones can be "scorched" by heating 2 minutes at 220 °C to melt resin, then opened by heating 24 hours at 60 °C.

(2) *Eucalyptus saligna* seeds, in Brazil, need 8 hours at 60 °C to dry capsules to below 20-percent moisture for best extraction.

(3) *Populus tremuloides* seeds, in Canada, release at catkin moisture content below 70 percent; they are picked 1 week early and air-dried for 3 to 6 days.

e. Extraction after drying— Once the cones are open, a simple tumbling action will extract the seeds.

(1) Tumbler driers were described previously.

(2) Cement mixers are versatile machines that can also crush fruits and extract and scarify seeds. Extraction can be aided by placing a wire cage inside the drum.

(3) Homemade tumblers are easy to construct. Wire-screen boxes that turn on a shaft can be tilted so that seeds fall out.

E. Single-Seed Fruits

Single-seed fruits include drupes and fleshy fruits and nuts with husks.

1. **Drupes and fleshy fruits *(Prunus* and *Vitis)* — *Use*** macerators, mixers, etc.; use water to soften the contact. (See "Moist, fleshy fruits.")
2. **Nuts with husks *(Juglans)*—** Use macerators or hand rubbing.

F. Sources

For additional information, see Willan 1985, p. 87-111.

II. Cleaning and Upgrading

A. Introduction

Cleaning seedlots is a basic step in proper seed utilization. Cleaning should remove wings or other seed appendages, empty seeds, damaged seeds, and nonseed trash. This cleaning should also provide dramatic decreases in insect and disease problems. Many seedlots can be upgraded by removing immature, damaged, and dead seeds after the initial cleaning. Many people view large mechanical operations as the only way to clean and upgrade seedlots, but seedlot quality can be improved with simple equipment and techniques.

B. Objectives

1. Learn the advantages of cleaned and upgraded seedlots.
2. Become familiar with the principles of seed-cleaning equipment and techniques and expected results.
3. Apply these principles when seed cleaning and upgrading are planned.

C. Key Points

The following points are essential for seed cleaning and upgrading:

1. Liquid flotation can be an essential aid for many species, especially recalcitrant ones.
2. Screen cleaning is the basic seed-cleaning method.
3. Air separation, including winnowing, is a valuable technique.
4. Cleaning small lots for testing or research may be very different from cleaning large lots.
5. Upgrading seedlots offers potential improvements in eight areas.
6. Seed sizing can be useful for some species or sources but not for others.

D. Cleaning

1. **Flotation—** The simplest technique of all. Good seeds sink and bad ones float.

a. Initial moisture content is crucial because it determines whether good seeds sink or float. Long soaks (up to 24 hours) may be needed to make good seeds sink if they are extremely dry when collected.

b. Orthodox seeds are redried after flotation, unless they are sown immediately, but recalcitrant seeds are not.

c. Flotation

(1) Removes light trash.

(2) Removes many empty, broken, diseased, or insect-damaged seeds.

(3) Is very good for large seeds with high moisture contents (e.g.,

Quercus, Carya, and *Juglans);* and for some small seeds (e.g., *Liquidambar, Pinus, Juniperus, Robinia, Gleditsia,* and other legumes).

2. **Aspirators—Any** machine that uses air to clean and separate.
   a. Large machines are found only in large-scale, seed-cleaning plants.
   b. Small-lot models are available for testing laboratories, research, or valuable small collections. Devices include:
      (1) General ER—a model with good air control, but low capacity (75 to 100 mL).
      (2) South Dakota—an old standby in testing laboratories, with enough capacity for small lots.
      (3) Stults— a new blower with less capacity than the South Dakota, but much better air control. It has a vacuum gage to help standardization. It has worked well with pines and small legumes.
      (4) Barnes — a laboratory blower that is seldom seen anymore. It works well with small conifers (e.g., spruce and Douglas-fir) but lacks power to separate Southern United States pines.
      (5) Other models — (see Willan 1985)
      (6) Homemade fan devices —" winnowing" type cleaning is fine, but weight separations usually are not good.
      (7) Carter Day Duo Aspirator — a machine that can handle a wide range of seedlot sizes in continuous flow.

3. **Screens and sieves**
   a. Hand screens —a good set of hand screens is extremely helpful, especially for testing and research laboratories and for small seedlots.
   b. Mechanical screens— some small "flat-screen" cleaners use screen agitation only; they are essential for large quantities.

4. **Air-screen cleaners—use** both aspiration and screening. These are the basic seed-cleaning machines in most seed plants.
   a. Air screens perform the following three functions:
      (1) Scalping— removes large materials (twigs, leaves, etc.) with the top screen.
      (2) Sizing—drops small particles through the second screen and allows the seeds to remain.
      (3) Aspiration—removes very light material, including some empty seeds.
   b. These machines are more efficient for cleaning, not sizing. Large machines (for tree-seed plants) have five screens and two air systems. They can clean 650 to 700 kg of *Pinus* seeds per hour. Most plants use one machine to scalp (two to three screens and one air system) and another for final cleaning and sizing (four to five screens and two air systems).
   c. Small tabletop cleaners are great for small lots; they can clean 30 to 40 kg of small seeds per hour.

5. **Electrostatic cleaners —** The Helmuth machine is good for small seeds (e.g., *Eucalyptus).* It is expensive, however, for such a limited function.

6. **Dewinging—** A special type of cleaning that reduces storage volume, makes upgrading possible for winged seeds (e.g., *Pinus* and *Liriodendron),* makes sowing easier in nurseries, and removes sources of pathogens. There are two basic methods of dewinging— wet and dry.
   a. The dry method is recommended for tough seeds because of the damage potential to thin seed coats. *Pinus palustris* is an exception.
      (1) The popcorn polisher (Crippen EP-26) is an old method that is rarely used now because of damage to seeds.
      (2) USDA Forest Service dewinger from Missoula Equipment Development Center flails seeds with rubber fingers in a cylinder with soft rubber on the walls to provide gentle action and little damage.
      (3) Dybvig macerators are good for *Liriodendron* and *Tilia* bracts.
      (4) Electric drum scarifiers can clean *Populus, Salix,* or other tiny seeds with hairy appendages.
      (5) New conifer dry dewingers have soft rubber walls and 90 kg per hour capacity in continuous flow.
   b. The wet method is usually preferred for pine and spruce seeds. It is quick enough to avoid much moisture uptake, but some redrying is usually needed for most species.

(1) Cement mixers are good for medium lots; seeds are sprayed with a mist while mixing.
(2) Commercial dewingers are available with capacities up to 90 kg per hour.
(3) Kitchen blenders can dewing small lots; water is added to the seeds at low speed for 10 seconds.
(4) Any cylinder with gentle agitation will dewing; even rapid stirring by hand will work for some species.

E. Upgrading

1. **Upgrading** is improving the potential performance of a seedlot by removing empty, damaged, weak, immature, or odd-sized seeds. Many people clean their seeds but never upgrade. Upgrading does not make good seeds from bad but can certainly improve the seedlot.
2. **Upgrading will:**
   a. Remove weak seeds
   b. Remove empty seeds
   c. Reduce chances of insect and disease damage by removing damaged seeds
   d. Improve control of density in nursery or germination beds
   e. Reduce planting time in the nursery
   f. Facilitate nursery operations with uniform seedings
   g. Reduce costs and improve uniformity in container operations
   h. Reduce storage space requirements
3. **Methods and equipment**
   a. Specific gravity by flotation removes empty and some damaged seeds.
      (1) Water is used for some *Pinus, Quercus,* and other large seeds.
      (2) Organic solvents (usually alcohols) of different densities work on some small seeds but may damage the seeds, especially if they are stored for long periods. Seeds floated in organic solvents should be sown soon. The solution must always be stirred vigorously to give every seed a chance to float.
   b. Air-screen cleaners, the basic cleaning machines, can also be used to upgrade.
      (1) They can separate by three physical properties: size, shape, and density.
      (2) They can upgrade some seedlots (e.g., *Platanus, Liquidambar,* and some *Pinus* spp.) by sizing or by removing empties with the air system.
      (3) Screen pattern, feed rate, airflow, screen oscillation (pulleys), and screen pitch (in some models) can be regulated.
   c. Air separators include large air-column separators, fractionating aspirators, and small laboratory blowers.
      (1) Large air-column separators are not common in tree-seed plants.
      (2) Fractionating aspirators are now widely used for *Pinus* in the United States.
      (3) Small laboratory blowers are suitable for small research or testing lots.
   d. Gravity separators were originally built to remove ore from clay. Heavy particles stay in contact with the shaking deck and "walk up" the high side. Light particles "float" on the air cushion and come off the low side.
      (1) Separators can separate seeds of the same size and different densities, or different sizes and the same density; they cannot separate a mixture of densities and sizes.
      (2) They are widely used to upgrade conifer seeds in North America.
      (3) The operator can regulate feed rate, air stream through deck, deck pitch (side and end), and eccentric thrust.
   e. Electrostatic separators create a charge that adheres to the seed surfaces.
      (1) The Helmuth cleaner can handle *Eucalyptus, Platanus,* and small conifer seeds. Falling seeds receive negative charges and are deflected toward a positively charged plate; deflection varies according to the weight of the particle.
      (2) Static electricity is used for very small seeds. The sides of a plastic beaker are wiped with a nylon cloth, and the seeds and chaff are poured into the beaker. The seeds are then poured into a noncharged glass beaker, and the chaff clings to the sides of the first beaker.
   f. X-ray radiography is typically used for valuable research lots only. With Kodak paper or Polaroid film, production is good. One United States company has studied mechanization of x-ray sorting.
   g. Color separators remove light-colored seeds. There is an application for high-priced lots, and one European company has a color sorter in its production line.
   h. The incubation, drying, and separation (IDS) method is used on some *Pinus* and

Picea species in Sweden (Simak 1984). The IDS method:

(1) Incubates seeds for 3 days in 100 percent relative humidity, 15 °C, and light.

(2) Dries them for 12 hours at 15 °C, 35 percent relative humidity, and light.

(3) Separates dead from live seeds by floating in water; in 5 minutes, viable seeds sink. Aspirators or gravity tables can also be used to separate the seeds.

(4) Redries the live seeds to 6- to 7-percent seed moisture.

(5) Can substitute stratification for 30 days at 4 °C for the incubation step in (1) above for some species.

4. **Sizing** helps with some species or seedlots, but not with others (table 8) [no equivalent table in Student Outline]; e.g., clonal mixes vs. single family seedlots.

Table 8.—*Correlation of seed size with germination rate and seedling size (adapted from Bonner 1987b) [no equivalent table in Student Outline]*

Species	Germination rate	Seedling size
	--- *Correlation*	----
Acacia albida	No	Yes
A. nilotica	No	Yes
Acer saccharum	Yes	
Araucaria angustifolia	Yes	Yes
Azadirachta indica	No	Yes
Carya illinoensis		Yes
Picea abies[t]	No	No
P abies[t]	No	Yes
P glauca		Yes
Pinus elliottii	No	Yes
P. koraiensis		Yes
P. roxburghii		Yes
P sylvestris[t]	Yes	No
P. sylvestris[t]	No	Yes
P. taeda[t]	Yes	Yes
P. taeda[t]		No
Quercus alba		Yes
Q. ilex	No	
Q. petraea		Yes
Q. prinus		Yes
Q. robur		Yes
Q. rubra		Yes
Q. velutina		Yes
Shorea contorta	Yes	Yes
Tsuga heterophylla		No
Theobroma cacao	No	Yes

Data not available.

[t]More than one experiment reported for these species.

F. Summary—See figure 29 [no equivalent figure in Student Outline] for a flow chart for extracting and conditioning *Pinus* seeds.

G. Sources

For additional information, see Bonner 1987b; Doran and others 1983, chap. 5; Willan 1985, p. 87-128.

III. Storage Principles

A. Introduction

The primary purpose of storing seeds is to have a viable seed supply when it is needed for regeneration. Successful storage of woody plant seeds must be carefully planned, and good planning depends on an understanding of the purposes of storage, seed deterioration, and the effects of the storage environment on the deterioration processes.

B. Objectives

1. Learn the objectives and rationale of seed storage.
2. Identify factors that affect seed longevity in storage.
3. Review the general process of seed deterioration.

C. Key Points

The following points are essential to understanding seed storage principles:

1. Longevity of seeds is a species characteristic.
2. Prestorage factors affect longevity in storage.
3. The most important factors in storage are seed moisture content and temperature.
4. Seed deterioration begins at abscission and involves complex physiological changes.

D. Objective of Storage

Once mature seeds are collected, deterioration starts; deterioration is first manifested as slower germination and then as death of seeds and sometimes an increase in abnormal germination. The objective of seed storage is to delay deterioration or decrease its rate until seeds are used.

E. Rationale for Storage

Seed storage includes short- and long-term storage; it may be extended for long periods for germplasm conservation.

1. **Short-term storage:**
   a. Is used for immediate operations
   b. Typically lasts less than 5 years
   c. Allows carry-over of surplus production in good seed years
   d. Allows minimum storage environment for most species

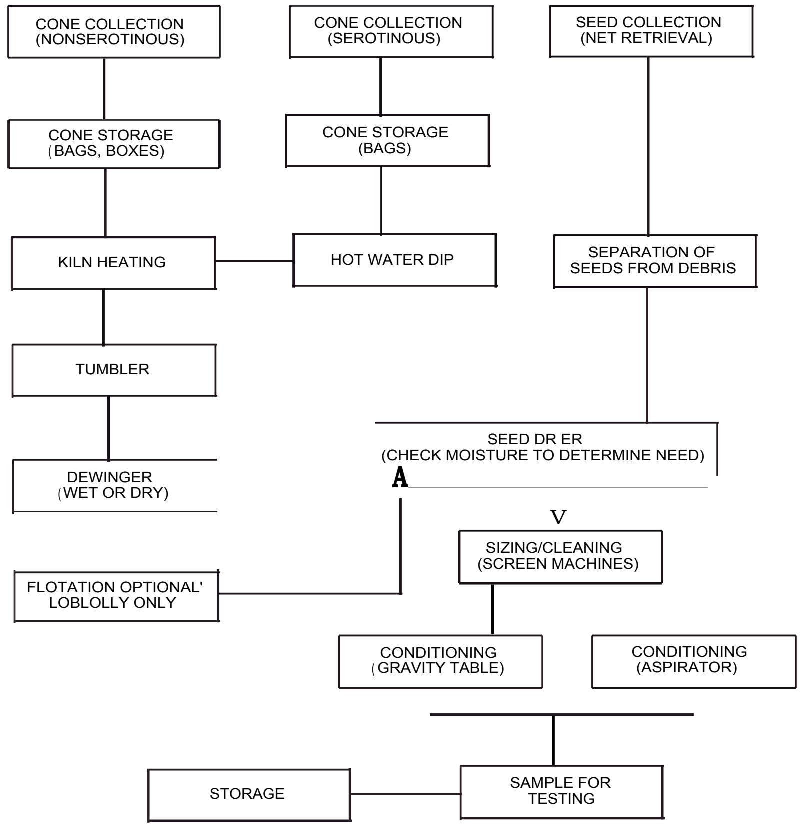

Figure 29. —*Flow chart for extracting and conditioning seeds of* Pinus *in a typical extractory in the Southern United States (Bonner 1991b) [no equivalent figure in Student Outline].*

2. **Long-term storage:**
   a. Typically lasts from 5 to 10 years
   b. Ensures constant seed supply for species that are irregular producers
   c. Is used to save special lots that will not be collected annually; e.g., in geographically remote areas
   d. Requires very good storage environments
3. **Germplasm conservation requires:**
   a. Very long-term storage with a goal of 50 years or more
   b. The best storage environment or perhaps special technology

F. Longevity in Storage

Seed characteristics, seed handling before storage, genetics, and the storage environment affect longevity in storage.

1. **Seed characteristics**
   a. Basic physiology
      (1) Orthodox seeds are tolerant of desiccation to low moisture contents (usually less than 10 percent); dried to this level, they may be stored at subfreezing temperatures.
      (2) Recalcitrant seeds are intolerant of desiccation, usually dying if dried below moisture levels of 25 to 30 percent or even 50 percent for some tropical species; at these levels, subfreezing temperatures are lethal.
      (3) These definitions were originated by Roberts (1973). They are not "biologically logical," but they are now well established.
   b. Seed structure — Thick or hard seedcoats restrict moisture uptake and gas exchange; thin seedcoats allow too much of both.
   c. Seed chemistry — Oily seeds tend to be harder to store than starchy seeds.
   d. Stage of maturity—Immature seeds usually will not store as well as seeds that were fully mature when collected. Therefore, maturity indices and time of collection become very important.
   e. Environmental stress during maturation — Drought, high temperatures, and perhaps nutrient deficiencies may decrease seed quality on the mother tree and thus decrease storage potential.

2. **Seed handling before storage**
   a. Physiological mistreatment, such as allowing overheating at high moisture levels, will damage storage potential.
   b. Processing can cause seedcoat cracks and bruised tissue that can lower seed quality, primarily by allowing invasion of pathogens (table 9) [no equivalent table in Student Outline].
3. **Genetics—Good** seed quality, and thus greater storage potential, seems to be inherited to some degree. There are also genetic differences among species; some are longer lived than others.
4. **Storage environment**
   a. Moisture content
      (1) Moisture content is the most important factor.
      (2) Potential damage thresholds are outlined in table 10 [table 5 in Student Outline].
      (3) The best range for orthodox seeds is 5 to 10 percent.
      (4) The best range for recalcitrant seeds is full imbibition.
      (5) The equilibrium moisture content is defined as the seed moisture content when seed moisture is in equilibrium with the moisture in the storage atmosphere (table **11)** [table 6 in Student Outline].
         (a) Equilibrium moisture content is influenced by seed chemistry; proteins are most hydroscopic, then carbohydrates, then lipids (figs. 30, 31) no equivalent figure in Student Outline for figure 30; figure 31 is figure 9 in Student Outline].
         (b) Equilibrium is rarely reached with recalcitrant seeds because metabolism rapidly changes seed weight and chemical condition.
         (c) Sorption and desorption difference must be recognized.
   b. Temperature
      (1) Generally, the cooler the seeds, the slower the deterioration rate for both orthodox and recalcitrant seeds.
      (2) The safe temperature range for orthodox seeds is related to the moisture content of the seeds; 20 percent may be the critical upper limit for 0 °C or lower (free water and ice crystals), 15 percent for

Table 9.— *Germination of unscarified and scarified seeds stored under different conditions for 2 months (Lauridsen and Stubsgaard 1987) [no equivalent table in Student Outline]*

Species	Seed storage condition	Scarification method: Seed gun	Seed burner
		Percent	
Acacia farnesiana	Unscarified, 0 to 4 °C	51 (±11)	98 (±2)
	Scarified, 30 °C, and 80 percent RH*	47 (±11)	97 (±3)
Prosopis cineraria	Unscarified, 0 to 4 °C	75 (±3)	94 (±3)
	Scarified, 30 °C, and 80 percent RH*	78 (±5)	

*RH = relative humidity.
Process not used for this species.

Table 10. —*Moisture content thresholds and potential effects on stored seeds ltable 5 in Student Outline]*

Moisture content	Effects
Percent	
>30	Germination begins
18 to 20	Overheating from respiration
10 to 18	Seed fungi become active
>9	Insect activity
5 to 8	Best range for sealed storage
<5	Desiccation damage possible in some species

Table 11. —*Equilibrium moisture contents at 4 to 5 °C and three relative humidities (Bonner 1981b, Justice and Bass 1978) [table 6 in Student Outline!*

Species	Relative humidity (*Percent*): 20	45	95
	Moisture content (*Percent*)		
Orthodox trees			
Carya ovata		10	15
Juglans nigra		11	20
Liquidambar styraciflua		8	20
Liriodendron tulipifera		10	19
Picea abies	6	8	
Pinus sylvestris	6	8	
P. taeda		10	17
Prunus serotina		9	17
Orthodox crops			
Glycine max	6	8	19
Zea mays	8	12	20
Recalcitrant trees			
Quercus alba		37	50
Q. nigra		13	29
Shorea robusta			35

Data not available.

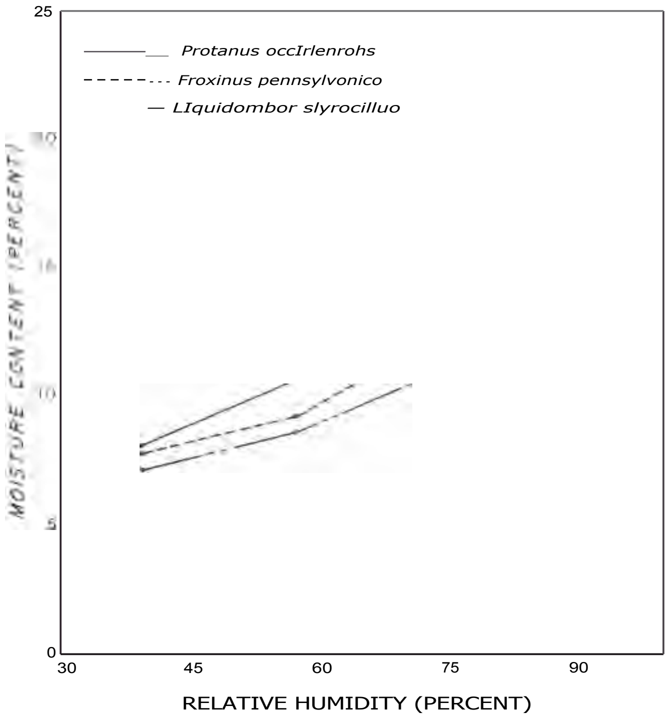

Figure 30. —*Equilibrium moisture content at 25 °C for three orthodox species (adapted from Bonner 1972b) [no equivalent figure in Student Outline].*

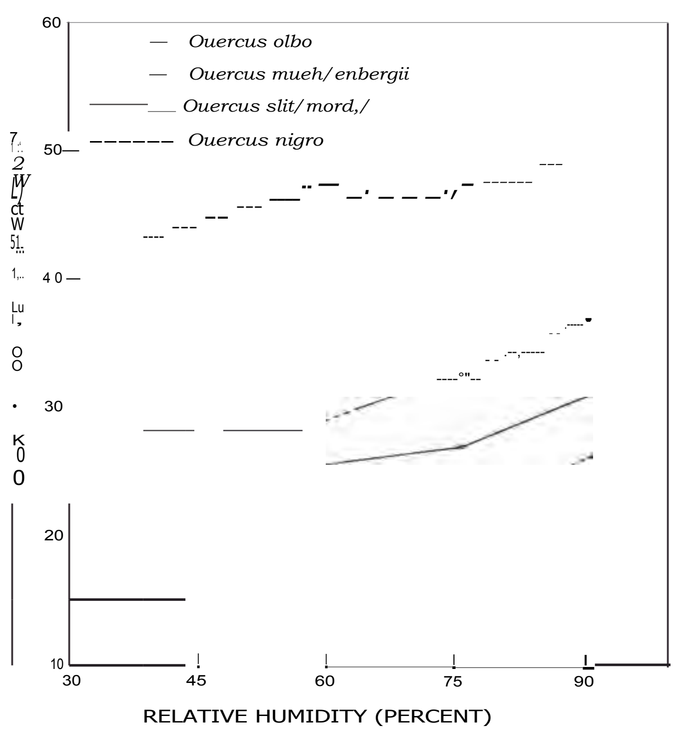

Figure 31. —*Equilibrium moisture content at 25 °C for four recalcitrant* Quercus *species (adapted from Willan 1985) [figure 9 in Student Outline].*

— 15 °C, and 13 percent for — 196 °C.

(a) Orthodox seeds at 5- to 10-percent moisture can be stored at most temperatures.

(b) Between 50 and 0 °C, every 5 °C lowering of storage temperature doubles the life of the seeds (Harrington 1972).

(3) The safe temperature ranges for recalcitrant seeds are:

(a) Temperate Zone species: —1 to 3 °C.

(b) Tropical species, usually above 12 to 15 °C because of chilling injury.

c. Storage atmosphere

(1) Reduced oxygen slows metabolism and increases longevity, but it is not usually practical to regulate oxygen level.

(2) Inert gases have been tested for storage. They show no advantage in long-term storage, but they may help in short-term storage:

(3) In sealed containers, the $CO_2/0_2$ ratio changes because seeds absorb oxygen in metabolism.

(4) Recent work shows that *Pinus radiata* stored in nitrogen or carbon dioxide retained viability long enough for transport in the Tropics.

(5) Research in Indonesia showed that coating *Shorea pinanga* seeds with wax retained close to 50-percent viability for 4 weeks; uncoated seeds died during this period. The coating reduced gas exchange.

G. Cells and Tissues During Seed Aging

The following changes occur in cells and tissues during aging:

1. Loss of food reserves by respiration.
2. Accumulation of metabolic byproducts from respiration. Some are toxic.
3. Irreversible enzyme deactivation because dried protein molecules lose their ability to form active protoplasms on rehydration.
4. Deterioration of cell membranes: endoplasmic reticulum and mitochondria.
5. Lipid peroxidation forming damaging free radicals. (There is some question as to whether peroxidation is the cause or an effect.)
6. Alterations of DNA, causing genetic mutations and physiological damage.

H. Sources

For additional information, see Bonner and Vozzo 1990; Harrington 1972; Justice and Bass

Table 13. — *Storage test results for true orthodox species (adapted from Bonner 1990) [table 8 in Student Outline]*

Species	Storage conditions: Temperature	Storage conditions: Seed moisture	Storage results: Time stored	Storage results: Viability loss
	°C	*Percent*	*Years*	*Percent*
Abies procera	0	9	7.0	11
Acacia leptopetala	20-25		18.0	1
A. mangium	4-8		1.2	6
A. pruinocarpa	20-25		16.0	20
Acer saccharum	— 10	10	5.5	5
Albizia falcataria	4-8		1.5	10
Araucaria cunninghamii	—15	16-23	8.0	few'
A. cunninghamii	19	7	0.1	0
Casuarina equisetifolia	—3	6-16	2.0	0-5
C. torulosa	20-25	8-12	18.0	6
Liquidambar styraciflua	3	5-10	9.0	3
Pinus caribaea var. *hondurensis*	8		2.7	± 16
P elliottii	4	10	50.0	30
P merkusii	4-5	<8	4.0	0
P ponderosa	0	8	7.0	0
Tectona grandis	0-4	=12	7.0	0
Tsuga heterophylla	5	8	2.0	0
T. heterophylla	— 18	8	2.0	0

*Data not available.
'Exact value not available from original source.

Table 14. — *Storage test results for suborthodox species (adapted from Bonner 1990) [table 9 in Student Outline]*

Species	Storage conditions: Temperature	Storage conditions: Seed moisture	Storage results: Time stored	Storage results: Viability loss
	°C	*Percent*	*Years*	*Percent*
Citrus limon	—20	5	0.9	+ 5
Fagus sylvatica	— 10	10	5.0	34
Gmelina arborea	—5	6-10	2.0	10
Populus deltoides	—20	6-10	6.0	21
Salix glauca	— 10	6-10	1.2	0

Table 15. — *Storage test results for temperate recalcitrant species (adapted from Bonner 1990) [table 10 in Student Outline]*

Species	Storage conditions: Temperature	Storage conditions: Seed moisture	Storage results: Time stored	Storage results: Viability loss
	°C	*Percent*	*Months*	*Percent*
Acer saccharinum	—3	50	18	8
Quercus falcata var. *pagodaefolia*	3	35	30	6
Q. robur	—1	40-45	29	31-61
Q. rubra	— 1 to —3	38-45	17	18-46
Q. virginiana	2		12	35

Data not available

3. **Temperate recalcitrant**
   a. Temperate recalcitrant seeds are intolerant of desiccation (table 15) [table 10 in Student Outline].
      (1) They cannot be dried below 20- to 30-percent moisture; thus, storage must generally be above freezing, although some *Quercus* species have been reported to survive storage at —2 °C.
      (2) Metabolism and pregermination are common in storage.
      (3) They cannot be stored in airtight containers; some gas exchange is necessary (table 12) [table 7 in Student Outline].
   b. Examples include *Quercus* (high lipid) and *Aesculus* (high carbohydrate).
4. **Tropical recalcitrant**
   a. Tropical recalcitrant seeds have the same moisture and gas exchange requirements as temperate recalcitrant seeds, but they are sensitive to low temperature. Chilling damage and death will occur below 12 to 20 °C, depending on the species (table 16) [table 11 in Student Outline].
   b. They are the most difficult group to store, even for short periods.
   c. Examples include species of *Shorea*,

Tabl e 16. — *Storage test results for tropical recalcitrant species (adapted from Bonner 1990) [table 11 in Student Outline]*

Species	Storage conditions		Storage results	
	Temperature	Seed moisture	Time stored	Viability loss
	°C	*Percent*	*Days*	*Percent*
Araucaria hunsteinii	19.0	25-30	54	-± 30
A. hunsteinii	2.0	30	365	82
Azadirachta indica	26.0	10-18	56	65
Hopea helferi	15.0	47	37	2
Shorea robusta	13.5	40-50	30	60
S. roxburghii	16.0	40	270	±30

Table 17. —*Storage test results for cryogenic trials of forest tree seeds (adapted from Bonner 1990) [Table 12 in Student Outline]*

Species	Seed moisture	Time stored	Viability loss
	Percent	*Days*	*Percent*
Abies alba		6	5
A. concolor	<13	180	0
Fagus sylvatica		6	100
Larix decidua		6	5
Picea abies		6	1
Pinus echinata		112	0
P ponderosa	<13	180	0
P sylvestris		6	0
Populus tremula x tremuloides		6	1
Ulmus pumila		112	0

*Data not available.

Hopea, Dipterocarpus, and even some legumes; e.g., in Costa Rica, *Pithocellobium.*

E. Cryogenic Storage

Cryogenic storage is a method for very long-term storage for germplasm conservation (table 17) [table 12 in Student Outline I.

1. **Techniques**
   a. Packages of seeds are immersed in liquid nitrogen (— 196 °C) or suspended above it in the vapor.
   b. The potential for germplasm conservation is good for small quantities but not for bulk lots.
   c. The upper (maximum) time limits of viability retention are not known. Only a few tests have been made on tree seeds.
2. **Costs—Comparable** with conventional storage in some cases of small seeds: $0.30 to $1.41 per year per sample of 1,000 seeds.

F. Physical Facilities

The optimum storage environment requires cold storage units, proper containers, and moisture management.

1. **Cold storage units**
   a. Cold storage units require a reliable power source, should not be used where floods or earthquakes may occur, should be located near other seed activities, should be rodent proof, and should be on high elevations when possible because ambient temperatures will be cooler.
   b. They should be built to hold a 5-year supply (or whatever the operational time).
   c. For germplasm conservation, **1** liter is needed of each sample (3,000 to 12,000 seeds) for medium-sized seeds (wheat and rice); e.g., 85 m^3 should hold 22,800 samples.
   d. Humidity control is not recommended in the Tropics; the seeds are dried and sealed in containers. If the power fails, seeds stay dry; this method is less expensive than humidity control.
   e. Direct or indirect vapor-compression refrigeration is best for the Tropics; it is usually available and reliable.
   f. Standby generators and safety alarms are recommended.
   g. When the power goes off and cooling stops, thermal time constants of 4 to 5 days should apply in large coolers, but if orthodox seeds are dry, serious damage may not occur for 2 weeks.
   h. Modular panel units are effective.
   i. Insulation depends on ambient conditions. A reliable local refrigeration specialist should be employed; an R value of 35 (heat transfer coefficient of 0.029 or less) is recommended.
2. **Containers**
   a. Fiber drums are very effective. Common capacities are 0.45 and 0.90 hL (25 and 50 kg for *Pinus taeda).*
   b. Generally, plastic is better than glass; plastic bags can be inserted inside glass containers.
   c. Rectangular containers utilize space better than round ones, but air spaces are still needed between containers. Round containers assure air spaces.
   d. Plastic bags should be 0.1 to 0.2 mm thick in humid atmospheres.
3. **Moisture management**
   a. Seeds will reach an equilibrium moisture content when exposed to the storage atmosphere.

b. If the storage unit has humidity control (50 to 60 percent relative humidity), orthodox seeds need not be sealed. Recalcitrant seeds cannot be sealed, so they cannot be stored in such a unit; the low humidity would desiccate the seeds.

c. Without humidity control, relative humidity will be 95 percent or more, which is fine for recalcitrant seeds. Orthodox seeds must be dried and stored in sealed containers in such a unit.

d. Humidity control is not recommended for the Tropics because both orthodox and recalcitrant seeds will be stored in the same facility.

e. Frost-free refrigerators are an alternative for humidity control because these systems remove the moisture from the unit.

G. Genetic Damage in Long-term Storage

1. Long-term storage could be devastating to germplasm conservation, but there is no strong evidence to date of lasting damage. Point aberrations occur on chromosomes, but they are not passed along to the next generation (fig. 32) [no equivalent figure in Student Outline].

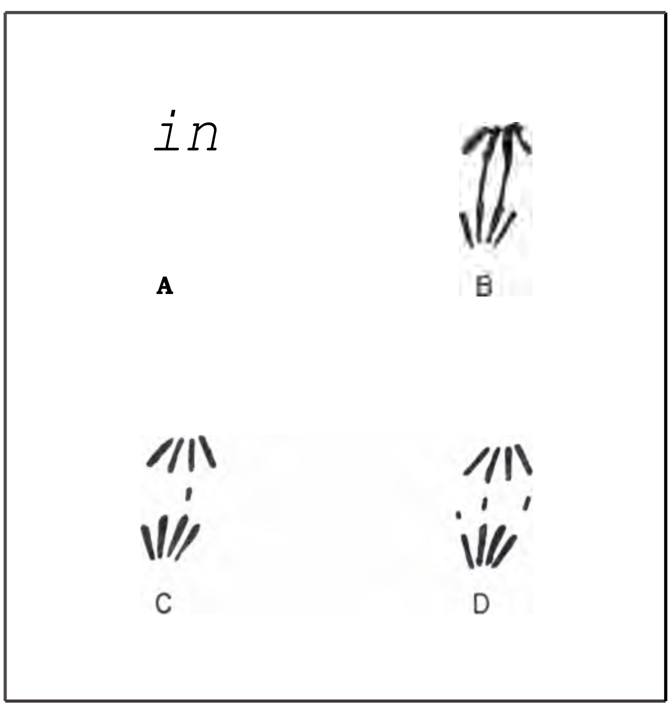

Figure 32. —*Chromosome aberrations found in germinated fresh and 50-year-old* Pinus echinata: (A) *single bridge, (B) double bridge, (C) single fragment, (D) multiple fragments (Barnett and Vozzo 1985) [no equivalent figure in Student Outline].*

2. Population changes are almost certain in heterogenous seedlots; some parts of the population die sooner in storage. The impact of this effect is still unknown.

H. Retesting in Long-term Storage

1. **Test procedures—** ISTA (1985) recommends 4 replications of 100. Subsequent tests can be 2 replications of 100; if viability has fallen 5 percent, the test is repeated with another 2 replications of 100.

2. **Test interval for orthodox seeds—** Should be initial year, third year, and every fifth year thereafter.

3. **Nondestructive testing—** Leachate conductivity could be used.

4. **Regeneration—In** annual plants, regeneration is done when viability falls to 50 percent. There may be a better plan for trees.

I. Viability Constants in Storage

The use of viability constants in storage is another technique introduced by Roberts (1973).

1. **Theory—Viability** retention will,be the same for a given species under a given set of storage conditions. This property can be expressed as a "viability constant" for each species.

2. **Practice**

a. Good results have been obtained with agricultural seeds.

b. Critics say varieties of a single species will differ.

c. One must start with very good seeds.

d. There are few data for tree seeds. Tompsett (1986) has developed constants for several tree species, and other data are available for two pines, *Liquidambar styraciflua,* and *Platanus occidentalis.*[2] The two hardwoods fit the expected pattern, but the pines have shown damage repair at high moisture levels after 8 years.

e. The above species are all orthodox. A different technique is needed to determine viability constants for short-lived recalcitrant species.

3. **Viability constants** for forest species could be useful in long-term storage for germplasm conservation or when good storage conditions are not available.

[2]Bonner, Franklin T. [n.d.] Unpublished research notes. On file with: U.S. Department of Agriculture, Forest Service, Southern Forest Experiment Station, Starkville, MS 39759.

4. **Procedures**
   a. Store samples in many combinations of temperatures and moisture contents, and test frequently.
   b. Plot seed death over time for each condition, using probit transformations.
   c. Calculate coefficients for time, temperature, and seed moisture content.
   d. See table 18 [no equivalent table in Student Outline].

J. Summary

Review table 12 [table 7 in Student Outline].

K. Sources

For additional information, see Bonner 1990, Bonner and Vozzo 1990, Chin and Roberts 1980, Harrington 1972, International Board for Plant Genetic Resources 1976, Justice and Bass 1978, Roberts 1973, Tang and Tamari 1973, Willan 1985.

Table 18. — *Viability equation* coefficients for seeds of* Ulmus carpinifolia *and* Terminalia brassii *(Tompsett 1986) and four United States species [no equivalent table in Student Outline]*

Species	*KE*	C,,,	CF,	*Cq*
Liquidambar styraciflua	5.3435	1.7616	0.0307	0.000869
Pinus elliottii	5.2463	0.9832	0.0508	0.000571
R taeda	3.2783	0.7300	0.0348	0.000328
Platanus occidentalis	5.1013	1.6742	0.0354	0.000838
Terminalia brassii	4.9990	2.1490	0.0350	0.000410
Ulmus carpinifolia	5.7150	2.9660	0.0340	0.000408

*$V = Ki - PI\ loKE - Cw\log_{io} m - C_H t - C_q t^2$,

where

V = probit percent viability,

Ki = initial viability,

p = storage time (days),

KE = species constant,

m = percent moisture,

t = temperature (°C), and

C_w, CH, C_q are coefficients.