Chapter 5: Seeds

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Introduction

Successful reforestation is highly influenced by seed origin and seed condition. This chapter summarizes the best practices for planning, collection, processing and storage of seed collected from trees in natural stands as well as trees in tree improvement programs. It is essential to use genetically diverse, high quality, local seed and seedling stock that is best adapted to the planting site to ensure long-term success. Conifers are long-lived and must contend with all of the various physical and biological factors and hardships contained within the ecosystems in which they have evolved. Natural selection results in the accumulation of an enormous range of adaptive genes over time. An individual organism's genetic make-up is its genotype. Each tree species responds in its own way to local environmental conditions such as extreme heat or cold, moisture stress, aspect, soil type, and competitive pressures. An organism's phenotype is the combination of observable physical characteristics as determined by its genes from both parents and interaction with the environment. However, it is the maternal parent's phenotype one observes when choosing seed trees for cone harvesting in open-pollinated stand collections.

Superior quality, locally adapted seeds produce vigorous seedlings that become better established; over time they will out-compete surrounding vegetation for moisture, nutrients and space and occupy the site more rapidly. As noted in the 'Tree Improvement' section of this chapter, improved seed produced from seed orchards supported by tree improvement associations and cooperatives in California combine the benefits of genetic improvements, strict quality control, and well-tracked genetic and locational information. When improved seeds of the desired species are not available from seed orchards, wild seeds collected and stored in seed banks will be the primary source of seed for reforestation projects. A successful and timely response for reforestation requires meticulous planning, collection, processing, and storage considerations to ensure that appropriate quantities of seeds of documented origin are available when they are needed. Conifer seed production and quality are highly variable from year to year and should be sufficiently evaluated before collections are initiated. It is the irregular nature of tree seed crops that necessitates storing large quantities of viable seed when it is available, for use during the potentially long stretches of poor seed production. Low quality seed crops do not generally warrant the costs and time that will be invested in seed procurement and seedling production, however special consideration may be necessary for species or areas of high conservation concern. This chapter describes the many factors that must be considered to ensure that collections are successful and that seed quality is maximized.

A key objective of the text is to review the relationship between seed maturity and seed quality. Many of the concepts presented here have been accumulated from generations of forest resource scientists, geneticists, and conifer seed specialists and are considered common knowledge for the genre. The writers add their collective personal experience with cone and seed planning and collections, seed processing (cleaning and upgrade), and testing and storage to illustrate specific topics in the discussion.

Rapid climate change caused by human activities is changing forested environments. Reliance on long-standing seed transfer guidelines is necessarily being re-examined. Climate predictions in this century strongly suggest that reforestation practices moving forward should favor tracking both current and modeled future climates in making seed transfer decisions. Broadly adapted seed sources are key to mitigating some of the challenges posed by climate change. Tree improvement programs offer forest resource professionals management options that promote adaptation and resilience in rapidly changing environments. (See Tree Improvement section at end of this chapter.)

Importance of Seed Origin

The success of any reforestation effort is dependent on the origin of the seed and its quality and condition. Seed origin is important because trees are locally adapted to the numerous characteristics of the environment they are naturally growing in. However, ongoing changes in forest climate conditions require explicit decisions on matching seedlings to the climatic conditions that the trees will face over coming decades. A key insight coming out of Douglas-fir seed source research across California, Oregon, and Washington has been the need to focus on "climate space" rather than "geographic space" as some of the key variables have been minimum winter temperature and maximum summer aridity, rather than simply latitude and longitude (Harrington and St. Clair 2017). In practical terms in California, this means a greater focus on the aspect as well as elevation where the seeds (or scions used in seed orchards) were collected rather than the seed zone number that often covers large elevation ranges in California. While California does not have as sophisticated a seed transfer system as the one recently developed for British Columbia (O'Neill et al. 2017), there is ongoing research across the Pacific States to build on forest genetics research to positively respond to new challenges (St. Clair and Howe 2017).

Development of the California Seed Zones

Foresters have long recognized that California's climate, topography and geology are very diverse and that these differences in the environment are significant to the productivity of a forest site and survival of the many tree species that are found there. This complexity resulted in the establishment of the California Tree Seed Zones (Fowells 1946) with revisions by Roy (1963) and Schubert (1966). In 1970 the Forest Tree Seed Committee (Buck et al. 1970) revised the original zones and restructured the recording system. It is this version of the California Tree Seed Zone map that is still in widespread use today. In addition to recognizing the vast topographic, climatic, and edaphic differences of the landscape, seed zones are meant to inform seed transfer decisions from the seed collection source to a specific planting location with minimal risk of maladaptation.

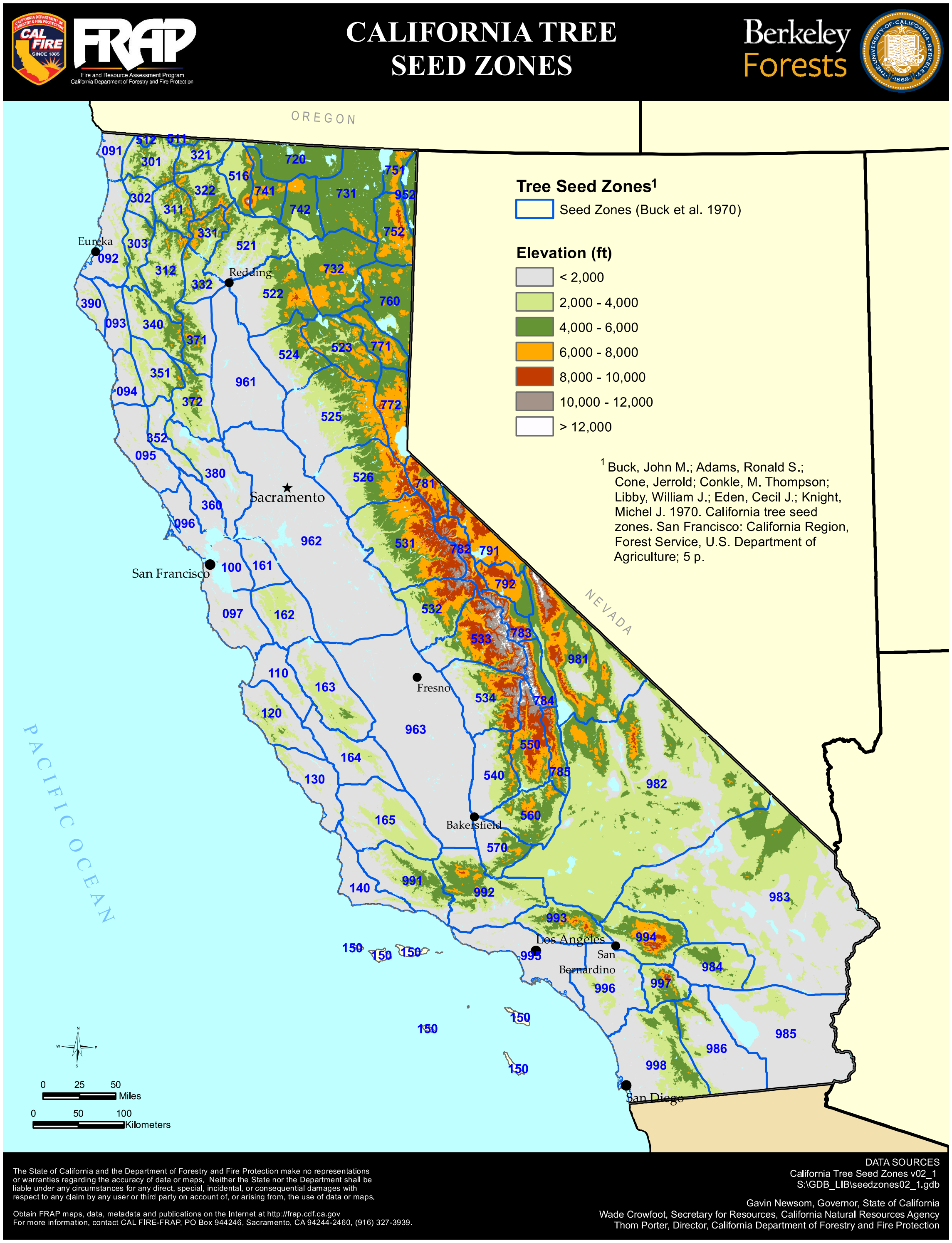


Figure 5.1 California Tree Seed Zone Map.

The state is divided into six major physiographic and climatic regions, thirty-two sub regions within these, and 85 Tree Seed Zones that are separate and unique (fig. 5.1). Plant associations within each region change as one moves from north to south, west to east and from low elevation to high elevation. In zones where climate and topography are more variable "local" generally implies the same microenvironment i.e., areas within the same drainage or those having the same aspect or elevation. Within the individual zones, conifer seed collections are catalogued by separate elevation bands with every 500 foot rise in elevation. The system of using seed zones and elevation bands to delineate areas of plant adaptation is used in California as well as many other states.

Main Features of the Physiographic and Climatic Regions

090 series – North Coast Redwood belt – extends along the coastal fog belt, approximately 5 to 30 miles in width, from California's northern border south to Monterey Bay. The belt lies slightly inland avoiding direct oceanic influences. Precipitation is plentiful in winter and summers are moderated by frequent fog. North of Cape Mendocino forest productivity is generally higher and associate conifers include Douglas-fir, grand fir, Sitka spruce, western hemlock, Port-Orford cedar and western redcedar. West of the redwoods, beach pine forest dominates on shallow, sandy soils. South of Cape Mendocino is predominantly lower site and redwood mixes with Douglas-fir and remnant patches of Bishop pine and grand fir. The isolated Cape Mendocino area (sz 390) is not in the 090 series because it has unique topo/climatic elements that preclude oceanic effects and cooling fog and therefore, does not support redwood.

100 Series – Central Coast – encompasses the coastal ranges from the Sacramento-San Joaquin Rivers south to Santa Barbara and includes the Channel Islands. The Pacific Ocean is the chief influence on temperature and moisture the area receives. While the southern extent of the range for coast redwood and Douglas-fir is found near Big Sur on north facing slopes and in drainages fed by summer fog, the central coast is predominately a mix of disjunct stands of closed-cone pine and cypress groves. The region also contains a series of valleys with elongated ridges that are parallel to but inland from the coast; the flat San Joaquin Valley borders on the east. The inland region is decidedly warmer and drier than areas west of the coast range. However, where the mountains intercept Pacific moisture in steep drainages, pockets of ponderosa pine, sugar pine and Santa Lucia fir can be found. Coulter pine is present at higher elevations with foothill pine and oak woodlands dominating the east-facing foothills and valley floors. The inland southern mountains and valleys are considered semi-desert and support an arid plant species mix.

300 Series – North Coast interior – extends from the summit of the Siskiyous south to San Francisco Bay and lies between the coastal fog belt and the Sacramento Valley. The western edge of the region lies in the rain shadow of the coast range and overall the climate is Mediterranean with winter rains and summer drought. The area supports a wide variety of conifer forest, woodland and prairie that differs considerably with topography, parent material, precipitation type and amount, and elevation. Indeed, the region has the largest number of sub-regions of any region. It also includes an isolated zone (390), located on the coast at Cape Mendocino which is predominately Douglas-fir.

500 Series – Western Slope Cascades and Sierra – an extension of the west slope of the Cascades in Oregon south through the Sierra Nevada to the Greenhorn Mountains in Kern County. It is bounded by the foothill belt of the Central Valley to the west and the crest of the Cascades and Sierra Nevada to the east. The region supports a rich diversity of conifers, though species occurrence on the landscape is highly dependent on latitude, elevation and aspect. These gradients define the temperature and precipitation type, the depth and duration of snowpack, and the length of the growing season. The lowest elevation conifer is foothill pine where it intermixes with the oak woodland type. The lower montane mixed conifer forest is dominated by a fluctuating mix of Douglas-fir, ponderosa and sugar pines, white fir, incense cedar and associated hardwoods. It is worth noting that, moving south of the Mokelumne River (526/531 border), Douglas-fir occurrence begins to decline. The upper montane contains the red fir forest type as pure stands or mixed variously with white fir, Jeffrey and western white pine, mountain juniper woodland, and well-defined groves of giant sequoia. Wetter habitats tend to support lodgepole pine. The subalpine type above 8,500 feet consists of a mix of slow-growing and scattered white pine and mountain hemlock associations.

700 Series – Modoc Plateau, Eastern Slope Cascades and Sierra - an extension of the east slope of the Cascades in Oregon south through the Sierra Nevada to Walker Pass in Kern County. The Warner Mountains mark the northeastern boundary of the region. The environment is character-istically warm and dry as it lies within the rain shadow of the southern Cascades and northern Sierra Nevada. The open plains and southern Cascades sport volcanic basalt and porous soils, in comparison to the granitic substrate of the Sierra Nevada. The northern reaches of the southern Cascades consist largely of white fir, Douglas-fir, western juniper, and disjunct groups of ponderosa and Jeffrey pine with a minor incense cedar component. Shasta red fir and Jeffrey pine forest join the mix on the western edge above 5,500 feet elevation. East of the Sierra Nevada (north and south of and including the Lake Tahoe basin) red and white firs, Jeffrey, lodgepole and white pines dominate in the higher elevations with western hemlock, while the singleleaf pinyon-juniper woodland is found below 5,250 feet. The most common forest vegetation in the Southeastern Great Basin is the limber pine forest with a lodgepole pine component and the whitebark pine subalpine type in the higher elevations. Trees growing here are adapted to a severe, cold-dry climate.

900 Series – Four Separate Areas – a 'Catchall' series:

950 – Great Basin: a high elevation desert valley in the far northeastern corner of California east of the Warner Mountain range, extending south from the Oregon border to Lower Alkali Lake.

960 – The Central Valley: bounded by the northern edge of the Sacramento Valley in the north to the base of the Tehachapi Mountains in the south. Natural vegetation includes grasslands, marsh, prairie, chaparral, and riparian and oak woodland habitats.

980 – Southern California Desert region: in far southeastern California. It encompasses the Mohave and Sonoran-Colorado deserts. Vegetation in the region is highly diverse but mostly devoid of conifers.

990 – Southern California Mountains: an area which comprises most of the natural conifer stands found in Southern California – a high elevation mix of mostly Jeffrey, sugar, ponderosa, coulter, and pinyon pines, white fir, incense cedar and a variable bigcone Douglas-fir component. A particularly challenging environment inland, winters are cold but relatively dry. The long droughty season is especially hot with frequent harsh, drying winds. Shallow soils and low precipitation are limiting factors for growth and survival of montane forests in the region. Three species of cypress: Sargent, Cuyamaca and Tecate, occur in the region in very small and widely scattered populations. They are naturally fire adapted but are increasingly threatened with post fire establishment failure in the wake of recurring human caused and climate driven conflagrations. The sub-alpine forest above 7,800 feet supports lodgepole, pinyon and limber pines and western juniper where temperatures remain low and snow persists well into summer.

Labeling Seed Zones and Elevation Bands

The numbering system for the tree seed zones uses a 3-digit designation, XYZ. Two digits follow the decimal and identify the 500 foot elevation band, XYZ.00.

Example: 500 feet = .05; 4000 feet = .40

X = Denotes major geographic areas having similar topographic, climatic and edaphic environments. **Physiographic and climatic regions**.

Y = Divisions within the major regions representing more localized topographic, climatic and edaphic environments. **Physiographic and climatic sub-regions**.

Z = Zone number –If Z is a number 1 through 9, then it is an arbitrary division of a sub-region to keep seed zones about 50 miles in latitude. If Z is 0, this is a unique zone and the zone is considered a sub-region.

Example: 340.15 – 3YZ represents the 300 series, or North Coast Interior physiographic and climatic region. 34Z represents one of the 17 sub-regions in the 300 series. 340 is a unique zone and therefore, a sub-region itself because there are no other 34Z zones. The .15 indicates an elevation between 1,001 and 1,500 feet.

Example: 526.45 – 5YZ represents the 500 series, or West Slope Cascades/Sierra physiographic and climatic region. 52Z represents one of the seven sub-regions in the 500 series. 526 is a zone, an arbitrary unit of the 52Z sub-region, in this case, the most southern zone in the 52Z sub-region. The .45 signifies an elevation between 4,001 and 4,500 feet.

The seed transfer guidelines, in use since 1970, served forestry professionals well for nearly 40 years but they assumed long-term environmental and climatic stability. Historically climate cycles have changed over millennia, century and decadal time frames and species have adapted by shifting ranges and adjusting, over considerable time, through genetic change (Rosenthal, Millar 2003). Science indicates that global climate change is currently accelerating more rapidly than in the past due to human influences. Such rapid changes demanded a review of the existing paradigms. New research-based seed movement guidelines were implemented in 2010 by the USDA Forest Service that may help to promote resiliency in a changing environment and are presented here:

Seed Transfer Guidelines

These guidelines modify the 1970 guidelines (Buck et al. 1970) based on ongoing research by the USDA Forest Service.

- Use source identified seed only – accurate and detailed seed collection records are key.
- Deploy forest reproduction materials, within a seed zone, upslope from source origin a minimum of one elevation band (500 feet) to 1000 feet upslope.
- Deployment of forest reproductive materials downslope from source origin is disallowed.
- Seed from an immediately adjacent seed zone (from south to north only) within the same physiographic and climatic sub-region (same XY) is considered compatible if the environmental conditions are similar. Use the closest available seed.

- Seed movement great distances is an uncertain solution, but if source origin is within 100 miles, not unreasonable (photoperiod changes more quickly at higher latitudes). (See inset for information about the Seed Lot Selection Tool, SST).
- Distance moved from source origin should consider whether the species is a generalist or a specialist in adaptive traits.
- Forest managers are encouraged to increase use of improved genotypes - improved genotypes from tree improvement programs are generally more broadly adapted and pest resistant and can be moved farther in both distance and elevation.
- Forest managers are encouraged to add a more diverse species mix and broader genetic base of a given species to account for climate uncertainties over the life of a stand.
- Forest managers are encouraged to use a <u>mixture</u> of seed sources that may be better adapted to the temperature and moisture extremes that are predicted in the next few decades.

Aitken and Bemmels (2015) suggested that the potential maladaption of trees to climate change could be addressed by choosing seeds from other regions with climates that match predicted future conditions. They further suggest that provenancing multiple seed sources to increase diversity and buffer against future climate uncertainty should be included in such as strategy.

The discussion about climate change and forest adaptation is in flux. Climate-driven changes necessitate that trees and forests adapt to new climatic conditions and environments. Forest resource professionals need to remain flexible when making deliberate seed transfer decisions. Genetic diversity is key – broad mixtures of species and sources that may be better adapted to future conditions may offer resilience in shifting environments.

Climate change mapping tools that support decisions on moving plant material across environmental gradients are becoming more readily available. These mapping applications are designed to assist with matching alternative seed sources with planting sites using selected climate variables. The main objectives are 1) given a planting site, what seed lot(s) will work or 2) given a seed lot, where on the landscape is it best suited for survival.

The Seedlot Selection Tool* (SST)** *is one web-based mapping tool that was designed, through a collaborative effort, by the USDA Forest Service, Oregon State University, and the Conservation Biology Institute. The goal of the SST is to help resource managers match seed lots (seed collections from a known origin) with appropriate planting sites based on ensemble models of current as well as predicted climate change projected across the landscape. This approach assumes that plants are adapted to the climate where they are currently found, however other important factors such as physiographic, edaphic, aspect/slope and competitive environment are not considered. Inputs allow users to select transfer limits, climate scenarios, and a set of climate variables. While the results from this tool are subject to the uncertainties and limits of such predictions, users can explore and compare the output derived from different selected variables to gain an understanding of how climate variation is distributed across a geographic area.* ***https://seedlotselectiontool.org/sst/

Future Climate Projections

Future climate scenarios are inexact but largely forecast a rise in temperature and changes in the amount and distribution of precipitation across the state (Fire and Resource Assessment Program 2018). Mean annual temperature is expected to increase in all ecological regions but the magnitude of temperature rise will vary by region; less near coastal environments and greater increase inland. While climate models and forecasts differ, a moderate range of increased warming from +2.5 to 4.6°C in Northern California is projected between 2000 and 2100 (Barr et al 2010). These projected increases are creating uncertainty about long-term forest tree adaptations and response to novel climates. There is widespread agreement that climate threats in California will include accelerated heat and drought (though some regions are projected to be warmer and wetter), increased fire frequency and intensity, and an increase in ancillary stressors such as insects, disease, and inter-species competition. As temperatures rise, more precipitation is expected to fall as rain rather than snow resulting in diminished water reserves in the Sierra Nevada. Less snow pack and earlier snow melt will result in increased flood events during winter and spring causing more soil erosion and less water availability for forest plants during the lengthy dry period (Fire and Resource Assessment Program 2018).

Greater climatic variability and uncertainty is expected to create serious implications for future tree health and cone production. It may be necessary to collect from a greater number of parent trees over a wider area than current standards require in order to capture as much variability as possible. The climate gradients that define an environment as compatible for a given population of trees are changing. Climate warming will impact winter minimum temperatures, summer maximum temperatures and the number of frost-free days. The impacts on bud set and bud flush are anticipated but will likely vary based on the biology of the species. As the rate of climate impacts increases, many trees may not adapt to the changed conditions fast enough and may be vulnerable to failure (Fire and Resource Assessment Program 2018). Tree communities located at the lower elevations or the southern, drier margins of a species range will likely show signs of decline first. In general, range expansion is expected at the leading edge (northern latitudes and higher elevations), with species' range contraction occurring at the trailing edge (southern margins and lower elevational limits).

In California, one current challenge is to prioritize collections of existing foothill and other low elevation seed sources as a hedge against loss of habitat type. In addition to potential genetic maladaptation to future conditions, foothill plant communities with limited moisture and high fire risk are especially vulnerable to loss of species abundance and distribution due to a variety of human caused factors. These include urban encroachment, fragmentation, overgrazing, past logging and mining activities, conversion to agriculture and increased fire and insect activity. Furthermore, these low elevation conifers are currently found in relatively hot and dry environments - they may have adaptations that will be valuable for planting out in a range of landscapes with future similar environments.

Biology

Cone and Seed Development

In conifers, cone and seed production involves four principal stages: the formation of reproductive buds, the development of male and female strobili (pollen and seed cones), pollination and fertilization, and seed maturation.

Bud initiation occurs each spring, but development into vegetative or reproductive structures depends on a variety of factors including environmental conditions, biological processes, and position on a branch. Bud differentiation takes place throughout the first growing season and is generally apparent by fall of the first year. The general appearance of the bud and position on the branch or part of the crown are indicative of bud type. Female (seed) cones are generally wider at the base, longer than male and vegetative buds, and are terminal or sub-terminal in position. Male (pollen) cones are generally laterally positioned and often found on undersides of lower branches in the crown. Under favorable weather conditions (dry, sunny weather in spring) and high nutrient status, a higher percentage of buds will become reproductive and fewer are likely to abort or become latent. The difference between an abundant cone crop and a poor one is more likely the result of the number of buds aborted rather than the number of buds produced in a given year.

Impediments to Formation and Development of Reproductive Buds

Impediments in this time period are thought to be unfavorable weather, timing of frost, and poor tree nutrition. While flowering can increase in trees under moisture stress, persistent drought conditions lead to decreased shoot growth, reduced crown development, and increased bud failure. Good nutrient status and greater light intensity favor good seed crops; therefore irrigation and fertilizer applications in seed orchards or seed production areas (SPAs) can promote bud development success. Additionally, thinning treatments in orchards and SPAs increase light intensity and reduce competition for moisture and nutrients for desirable leave (seed) trees.

In late fall reproductive buds slow their metabolic activity, becoming nearly dormant, but still undergo internal changes through winter. They resume active growth and burst the following spring when pollination (and for some species, fertilization) takes place – this is year two, the "cone development" year. The male pollen cones elongate rapidly and swell, then split open as they dry and release pollen grains into the wind (fig. 5.2). Nearly simultaneously the female seed cones elongate, causing the immature bracts to spread apart slightly and allowing the airborne pollen grains to enter. Upon capture by a receptive female cone, the pollen grains drift down the scales toward the ovules at the base of each scale and the female cone scales and bracts close again (fig. 5.3). In most species, with the exception of true firs which remain upright in the uppermost branches of the crown, the seed cones become pendant and pollination is complete (fig. 5.4).



Figure 5.2 (left) Male pollen cones *P. ponderosa*. **Figure 5.3** (center) Female cone *P. ponderosa*. **Figure 5.4** (right) Spent cone and pendant first year cone *P. menziesii*. *Photo:* Mary Ellen Harte.

Because conifers are wind pollinated, factors affecting success in this time period are mostly weather driven. Therefore, conditions of frost, drought, excessive wind, lack of wind, and excessive rainfall all have a strong influence on pollination success or failure. Losses can occur after pollination as well if temperatures become extremely low causing cones to abort. In instances when there is low pollen flight or mixing, self-pollination becomes a factor that can contribute to embryo degeneration or low vigor. Additionally, pollen viability, which can vary from one day to two to three weeks depending on species, and female cone receptivity may differ in timing, duration, or both in some years and further effect pollination success. With all of the variables involved at this early stage of cone development, it is difficult to predict in advance whether the end result will be a collectable crop of a given species in any given year.

Within the female cone, the pollen grains germinate and each will produce a long tube; one pollen tube penetrating a single ovule. After a series of cell divisions, fertilization occurs when a single sperm cell fuses with an egg cell. The time period for this process varies greatly by species and conifers generally fall into one of two categories: 2-year or 3-year species. For 2-year species fertilization closely follows pollination, usually within weeks, and cones develop through summer and mature in late summer or fall. For 3-year species the female ovule continues to grow slowly through the next winter while development of the pollen tube stops in mid to late summer; fertilization occurs the following spring and cones mature in the fall of the second year after pollination (approximately 15-16 months later). The cypresses comprise a third category and are mature after the embryo emerges from a dormant phase and resumes maturation in the year after fertilization for a total reproductive cycle of approximately three and one-half years.

Post-fertilization growth includes enlargement of the female cone and differentiation and maturation of the embryo within the female gametophyte (megagametophyte) or food reserve. The embryo has all of the necessary structures and genetic information to grow into a miniature plant; a radicle (primary root), hypocotyl (stem), and cotyledons (first leaves). The seedcoat protects the inner structures and may be thin, soft and papery as in true firs and incense cedar, or thin to thick and woody as in pines, Douglas-fir and juniper. In some species with especially hard seed coats, such as several species of pine and cypress, the seed coat may also restrict water uptake, gas exchange and emergence of the radicle and require treatment before germination can occur.

True firs, incense cedar, western red cedar and hemlocks have resin pockets in their seedcoats. It is thought that these resin vesicles protect the embryo from excessive drying and play a role in seed dormancy. Seeds with resin vesicles must be handled very carefully as these structures are easily damaged which may result in poor quality or death of the seed. The megametophyte surrounds the embryo and provides the nourishment necessary for initial growth of the embryo. Most conifer seed is winged which facilitates movement from the tree during dispersal; some wings are affixed lightly and are easily removed as with most pines while others are an integral part of the seed coat, as in true fir, Doulas-fir and incense cedar. Moisture reduction is the final stage of maturation before natural seed shed. This drying allows the seed to enter a quiescent period necessary for the synthesis of many enzyme systems including those required for seed dormancy and desiccation tolerance.

Impediments to Cone and Seed Development

Insects

Insects that feed on cones and seeds of forest trees can have a significant impact on reforestation planning. Included are various species of cone beetles, cone worms, moths, maggots, chalcids, and true bugs. For most conifers, cone and seed production is highly variable from year to year, producing moderate to heavy crops only periodically, interspersed by several years when seed production is low to scarce. This periodicity varies among conifer species so the size of insect populations can fluctuate wildly based on food supply from year to year. If there are back to back good crops, insect populations may increase and the second crop can be severely damaged. Likewise if a poor crop follows an abundant one insect populations will decrease or disappear and the subsequent crop may be insect-free.


Figure 5.5 (left) *Dioryctria spp*. feeding on buds of *P. ponderosa*. **Figure 5.6** (center) Pitch response on *P. menziesii* from *Barbara spp*. **Figure 5.7** (right) *Leptoglossus occidentalis* on white pine. *Photo*: Sandy Kegley, USDA Forest Service.

Insect activity may be minor or the damage to developing cones and seed can be severe. Early damage may be caused by defoliators that eat buds and foliage at the earliest stages of development (fig. 5.5). The spruce budworm, *Choristoneura spp.*, can decrease the quantity of pollen buds available and girdle female buds at the base causing them to abort. Cone beetles, *Conopthorus spp.*, kill developing cones by boring into the stalk – the conelets become brown and casehardened and cease development. Later damage occurs in developing cones by cone worm and cone moth larvae that hatch from eggs deposited onto the scale during pollen reception or from larva that bore through the cone scale or axis and consume the seed contents (fig. 5.6). The Western conifer seed bug, *Leptoglossus occidentalis*, feeds on conelets as nymphs and causes later damage to developing cones by the adult when it pierces the cone scale and extracts the seed tissues with its sucking mouth parts, causing the contents to collapse (fig. 5.7). Cone scale and cone gall midge activity deforms cones; this damage fuses the developing seed to the scale leading to extraction difficulties. Seed worms and chalcids may cause extensive damage to cone interiors but often go undetected because the cone may continue to develop normally. These losses are usually discovered only after samples are collected for quality and maturity assessments later in the summer.

Management strategies to control insect populations in native forest ecosystems are generally uneconomical but are feasible in seed orchards or seed production areas. Maintaining healthy trees and employing sound sanitation practices is key. Dead and injured branches should be pruned and destroyed. Stressors, such as soil compaction from equipment and competition from weedy vegetation, must be reduced as much as possible. It is very important to keep the orchard free of over-wintering pests by removing and destroying unharvested and spent cones on the ground that could usher damaging insect and disease pathogens into the following season.

Pathogens

There are several important pathogens that can adversely affect cones and seeds of conifers in California. One type is Sirococcus blight, caused by fungal spores that are ever-present and can spread to new shoots and cones in the spring. Infected seeds often have shrunken and discolored contents and the seed-borne inoculum results in a diseased seedling when exposed to favorable conditions for growth and spread during germination. *Sirococcus spp.* fungi become established in seed lots when older cones are included in the harvest and can contribute to seed deterioration and subsequent losses in nursery operations. Improved cultural practices over the years such as strict avoidance of squirrel caches, spent cones, and ground collections all have contributed to a significant reduction in the collection of *Sirococcus* diseased cones and seed.

Western gall rust, *Endocronartium harknessii*, is a disease that causes branch and stem galls on California pines. These galls can cause severe damage to cone bearing branches and result in cone and scale distortions in the following; knobcone, lodgepole, coulter, bishop, and Monterey pines, where the cone base and peduncle come into contact with the stem.

Pitch canker, *Fusarium circinatum*, is an introduced disease and is present in the coastal areas of California less than 75 miles inland from San Diego County in the south to southern Mendocino County in the north. Pitch canker is a highly virulent pathogen common to knobcone, bishop, and Monterey pines but is also found on other pine species. Because infections can include reproductive stages as well as vegetative, collection of cones and seeds of these species must remain local to avoid introductions of the disease into other areas of the state.

Some seed borne pathogens such as *Aspergillus, Fusarium, Mucor* and *Penicillium* may simply be indicators of poor seed quality rather than being the cause of deterioration. These agents may colonize seeds as a result of adverse environmental conditions such as high temperatures or moisture conditions, or physical damage due to improper handling during collection, transport, storage or during the extraction and cleaning processes. These fungi can infect seeds internally destroying the embryo and megagametophyte, or externally on the seed coat affecting seed germination when they cause damping off or root rot in the nursery. Disease development is also strongly affected by the degree of cone and seed maturation attained before cones are collected. Research has shown that seed viability and seed storage capacity decline when cones are not allowed to properly mature on the tree.

Sudden Oak Death, *Phytophthora ramorum*, is a disease that has killed countless oaks and other forest species since the mid-1990s. It also causes Ramorum blight, a twig and foliar disease in many other plant species including coast redwood and Douglas-fir. *P.ramorum* thrives in cool, moist environments and in California is currently limited to coastal evergreen and tanoak/redwood forests. It is a federally quarantined pest resulting in regulated or restricted movement of host material out of the zone of infestation (ZOI) to uninfected areas of the state or out of the country. Researchers have found that inoculum is not present on the cones or seeds of the conifer host species. Consequently, collections of Coast redwood were successful in 2008 in Santa Cruz County, and in 2009 in Mendocino County, albeit with strict sanitation precautions in place. The redwood cones were removed from the top half of the crown, “clipped in place” on site at the base of each cone to remove all green material, needles and twigs and transported in sterile cone sacks to the processing facility.

Mammals and Birds

Activities by mammals and birds may also cause loss of cones and seeds. By far the greatest losses are attributed to squirrels and chipmunks which collect and cache large quantities of ripe and unripe cones of pine, true fir and Douglas-fir for winter retrieval. A squirrel cache is a damp and decayed environment with a high level of diseased seed and debris. In addition, the cones and seeds that comprise them are from unknown parent trees, making these caches totally undesirable for collection purposes. Squirrel feeding can destroy 50 to 90 percent of a cone crop in some years. For high value trees, removing the lower branches and installing metal banding around the stem and increasing tree spacing to avoid crown to crown migration are known to significantly reduce damage by these agents. Mammals may also cause minor damage by feeding on cambial tissues, destroying buds and flowers – ponderosa pine is a favorite. Many bird species also feed on and cache developing and mature conifer seeds. While caching plays an essential role in seed dispersal and tree establishment, damage can be serious for collections when birds feed in flocks. Larger birds such as the Lewis's woodpecker, Steller's jay, and Clark's nutcracker mine through cones which may leave large damaged areas that cause significant seed loss and difficulties with extraction.

Humans

If cones are harvested too early, in most cases the maturation processes are interrupted and seeds cannot develop properly. Green cones and seeds are very high in moisture content and they will dry too rapidly if picked too soon. Research has shown that "drying on the tree" is necessary for the seed to enter the stage required for desiccation tolerance and germination when rehydration occurs (Bewley, Black 1994). The internal tissues have not completed the normal accumulation of storage food reserves and these reserves are insufficient when the connection to the parent tree is broken. Other impediments caused by human activity may be direct or indirect but all can create an unhealthy environment for growing trees. These include increased emissions and air pollution from urban areas, compaction of soils from overuse, and the myriad impacts realized as a result of climate change.

Cone Bearing Age

Conifers are slow growing, long-lived trees and the length of the juvenile period is extremely varied in the temperate zone. The majority of species may begin flower production at 8 to 20 years of age but likely produce only one sex or exhibit only intermittent flowering. It is generally many more years before conifers will reach cone bearing age (the age at which significant quantities of viable cones and good quality seed are produced) and become prolific producers later still. A dominant Douglas-fir, for example, may produce cones at 20 to 25 years of age but trees 80 to 100 years+ generally have many more viable cones per tree. There appears to be a tendency for earlier cone production on high sites. In most conifers there is strong correlation between tree size, crown position, vigor and cone production. Good cone production is generally best among dominant, young-to-mature vigorous trees with wide crown width and pointed top. These trees receive more sunlight from all sides. Cone production then tends to generally drop off as trees become over mature; this is true of ponderosa pine, sugar pine and true fir. However, in one study of the differences in cone production among different age classes of *Sequoiadendron giganteum*, it was found that the oldest age cohort (individuals 1,000 to 3,000+ years of age) consistently produced greater quantities of viable cones when compared to adjacent individuals comprising a second growth cohort (approximately 170 years old). The old growth trees are clearly still prolific even in advanced age.

Comprehensive Cone Collecting and Seed Banking Program

The "Seed Banking" System

Most native trees used for reforestation are grown from seed. Sufficient quantities of high quality, site-specific seed are therefore needed to restore and sustain plant communities that are increasingly threatened by catastrophic fire, insect and disease outbreaks, human impacts, fragmentation, invasive species, and the effects of climate change. In support of sustainable healthy forests the California Public Resources Code sec. 4681-4695 and Board of Forestry policy direct the Department of Forestry and Fire Protection, CAL FIRE, to provide an adequate, reliable, and continuous supply of site adapted seed of the widest possible diversity and highest quality to promote responsible reforestation and protect the genetic integrity of California's forested landscapes. To that end a comprehensive Forest Tree Seed Bank is maintained to provide the facilities for processing and storage of seed from a maximum number of species and adaptive seed zones. The Department of Forestry and Fire Protection maintains a full collection from all lands as well as provided quality seed storage for private owners who store seed collected from their lands for future use. In addition, private seed banks also store seed for some of their clients according to the same categorization protocols. Finally, the USDA Forest Service maintains its own seed bank.

Cone and seed production are highly variable from year to year. For that reason, developing cone crops must be sufficiently evaluated before collection can be considered. Good seed crops are the exception, so a high degree of advanced planning and cooperation among all stakeholders (USDA Forest Service, state and local agencies, forest companies and private landowners) is necessary to have the requisite reserves of each species from different seed zones and elevation bands in a seed bank for the years in between the bountiful, high quality seed crops.

Vigorous, site adapted seedlings are produced from good quality local seed. Successful seed production requires extensive knowledge of seed development, handling, cleaning, upgrade, and testing and storage procedures. The individual responsible for Seed Bank operations must have highly specialized knowledge of numerous species including: their ranges, adaptations, genetic variability, age to maturity, cone crop periodicity, seed structure and physiology, a variety of maturity indices, dormancy mechanisms, expected seed yield, storage capacity and more. This information is used to determine the seed needs list, the quantity of cones needed to satisfy long-term seed requirements, seed quality standards, crop monitoring systems, and identification of seed collection areas (Table 5.1). Planning in any given year will be based on this knowledge and budget constraints, collection procedures, the need for source certification, and the timing and pace of cone and seed ripening in that collection year. Understanding and monitoring these processes are essential to planning a successful cone collection.

STEP 1 - Determine Long-term Seed Needs

Planning

The first step in the development of any cone and seed collection program is to determine, for each species, the amount of seed required to meet the reforestation and emergency needs over a given planning period. The number of years in a planning period will vary with each species' periodicity and seed storage life expectancy, but is usually a minimum of ten years production plus an emergency reserve for unforeseen events. An adequate emergency reserve is another 50 percent or more above the 10-year production estimate. A simplified calculation is:

Planning period need minus number of trees potentially available from seeds in storage equals seed collection need.

The estimate of seed needed for each species is based on:

- The expected yield (in pounds of clean seed per bushel cones).
- The number of seeds per pound.
- The germination percent.
- The ratio of seedlings to viable seeds.

Table 5.1 Seed Lot Data used to Determine Seed Needs

Species	Avg. No. Cones/Bu	Avg. Lb. Clean Seed /Bu Cone	Avg. Germ %	Avg. Seed/Lb.	Nursery Corr. Factor [a] LAMRC Container Ops	Avg. # Plug Seedlings per Lb. of Seed
Jeffrey pine	35-50	1.2	85	3700	0.72	2664
Ponderosa pine	90-100	1.0	85	9200	0.76	6992
Sugar pine	12-18	1.4	80	1900	0.72	1368
Coulter pine	10-12	0.9	85	1360	0.72	979
Incense cedar	1000s	0.7	65	13600	0.54	7344
White fir	170-200	1.1	70	10200	0.54	5508
Red fir	50-65	0.9	70	4700	0.54	2538
Douglas-fir coastal	900-1000	0.5	85	33400	0.70	23380
Douglas-fir sierra	700-900	0.75	85	27400	0.70	19180
BC Douglas-fir	300-400	0.75	80	4300	0.70	3010
Coast redwood (no)	1000s	0.75	60	92700	0.42	38934
Coast redwood (so)	1000s	0.75	60	86900	0.42	36498

Estimates based on seed lot data from seed processed and grown at the CAL FIRE L. A. Moran Reforestation Center, 1980-2014.

[a] *Nursery correction factors are usually lower for bare root production due to its greater environmental variabilities - Container production is generally a more efficient use of good quality seed.*

Cone Crop Periodicity

Periodicity is the number of years between collectable cone crops. Nature produces a medium to heavy cone crop over a wide area only intermittently, usually once in 3-10 years, or even much longer (Table 5.2). Cone production differs by species, by regional location within a species, from stand to stand, and by individuals in a stand. Good cone crops may occur more frequently on a local basis while some locations regularly fail to produce viable cones. Indeed, failures may occur sporadically across a landscape even in abundant crop years. Furthermore, seed yield, quality, and storability vary considerably by species making procurement of an "adequate supply" an important and complicated consideration. The goal is to collect the maximum amount of high quality seed in good years and store them for the years in between. For example, coastal Douglas-fir trees may have conspicuous cones every three to five years but these are often a combination of insect-riddled and low filled seed cones. An abundant Douglas-fir cone crop with good quality seed in these areas has occurred only three times in the past 50+ years: 1968, 1982 and 1997. In another example, ponderosa pine generally produces a plentiful cone crop at 3 to 10 year intervals in much of its range, however, in the inland coastal area (300 series of zones) ponderosa pine has produced only very light to light crops approximately one in every 10 to 12 years and a good crop has failed to occur in more than 30 years. Cone survey records dating from 1958 to present document generally poor overall crop ratings statewide but show considerable local variation.

Table 5.2 Conifer Reproduction (and Periodicity) Data Chart

Important Reforestation Conifer Species In California	Elev. Range (m)	Max Ht. (m)	Onset of Cone Bearing Age (yrs.) [a]	Reproduction Range (yrs.)	# Years bet. Significant Crops [b]
White fir* *Abies concolor*	1000-3000	55	40	40-400	3-9
Red fir* *A. magnifica*	1400-2700	60	35-45	35-600	3-6
Incense cedar* *Calocedrus decurrens*	50-2960	50	20	20-500	3-6
Coulter pine *Pinus coulteri*	300-2000	25	10-20	10-100	Serotinous
Jeffrey pine* *P. jeffreyi*	500-2900	60	10-20	10-500	2-8
Sugar pine*	150-3000	65	40-80	40-600	3-5

P. lambertiana					
Ponderosa pine* *P. ponderosa*	150-2700	50+	15-20	15-600	4-10
Douglas-fir* *Pseudotsuga menziesii*	700-1800	75-90	20-25	20-500	5-15
Coast redwood* *Sequoia sempervirens*	10-975	84-120	15	15-2200	12-20
Giant sequoia *Sequoiadendron giganteum*	1400-2600	76	20	20-3000	Serotinous
More Narrowly Distributed Conifers in CA					
Pacific silver fir *A. amabilis*	1700-2100	70	20-30	20-500	2-6
Grand fir *A. grandis*	0-50	60	30	30-300	3-5
Sub-alpine fir *A. lasiocarpa*	1700-2200	30	50	50-200	3-5
Port-Orford cedar *Chamaecyparis lawsoniana*	800-1400	60	5-10	5-600	4-5
Whitebark pine *P. albicaulis*	2220-3660	20	20-30	20-700	3-8 [c]
Knobcone pine *P. attenuata*	180-2000	15	10	10-90	Serotinous
Foxtail pine *P. balfouriana*	2100-3700	22	20	20-1500	5-6
Lodgepole pine *P. contorta var. murrayana*	1500-3400	30	15-20	15-600	2-4
Limber pine *P. flexilis*	2200-3350	18	20-40	20-1000	3-6 [c]

Bristlecone pine *P. longaeva*	2600-3600	15	50	50-4000	2-4 [c]
Western white pine *P. monticola*	1370-3170	60	20	20-400	3-7
Bishop pine *P. muricata*	0-400	25	10	10-200	2-3
Monterey pine *P. radiata*	0-300	35	10	10-100	Serotinous
Foothill pine *P. sabiniana*	300-2100	25	25	25-200	2-4
Torrey pine *P. torreyana*	0-175	18	12-18	12-150	Serotinous
Singleleaf piñyon pine *P. monophylla*	1000-2900	12	35	35-800	5-7
Brewer spruce *Picea breweriana*	1150-2700	30-50	20-30	20-900	2-3
Sitka spruce *P. sitchensis*	0-20	70	25-40	25-800	3-4
Big cone Douglas-fir *Pseudotsuga macrocarpa*	275-2400	30	20	20-600	7-10
Western hemlock *Tsuga heterophylla*	10-400	60	20-30	20-500	5-8
Mountain hemlock *Tsuga mertensiana*	1900-2700	30	20-30	20-250	3-5

[a] *In the temperate zone the length of the juvenile period is extremely varied. Reliable and abundant production of viable seed tends to occur at a later age, often after 30 to 80 years.*

[b] *Periodicity - The authors' 60+ years collective experience has been that significant "collectable (economical and operational-size) crops" occur at irregular intervals that often exceed the parameters listed. The periodicity of collectable crops can often be longer at the lower, drier end of the elevation range.*

[c] *P.E. Maloney, personal communication.*

STEP 2 – The Annual Cone Crop Survey

Objectives of the Cone Crop Survey

The annual cone crop survey assesses the potential for collections in each of the specific physiographic and climatic regions of California. The purpose of the cone crop survey is threefold: to forecast the likelihood of successful cone production in the current year; to compare one stand or potential collection area with another; and to collect cone production information for long-term periodicity records. The decision to collect cones in a given year will be based on a systematic crop monitoring process that begins with visual observations of tree crowns in early summer and continues with assessments of the cones and seeds as they begin to mature. The cone crop rating is a numeric method for expressing the size of the potential crop and is based on two factors: 1) the number of well-distributed trees bearing cones in a mature stand, and 2) the relative number of cones in the acceptable portion of the crown on those individual trees. CAL FIRE uses the rating system described on the Cone Crop Survey Form (fig. 5.8). A rating of (4) medium and (5) heavy are generally considered collectable given that seed quality is determined to be acceptable as well.

> CAL FIRE plans to launch a beta version of the Cone Survey Mobile APP in lieu of the standard form below. Statewide rollout is expected in 2019 - 2020.

CALIFORNIA DEPT OF FORESTRY & FIRE PROTECTION
L.A. MORAN REFORESTATION CENTER
CONE CROP SURVEY FORM

Date: *July 10, 2001* **Rating By:** *L Lippitt*

Seed Zone: 523 **Elevation:** 3655'

T: 26N **R:** 10E **Sec:** 30 **Lat:** N40.04.803' **Long:** W120.53.795'

Stand Observed From: within stand _____ road __xx__

Motor Log: *Crescent Mills- from Hwy 89 take Arlington Rd. approx. 1.75 mi. to a seasonal road on the right accessing Beaty & Assoc. property. Survey is at the landing.*

Comments on Stand Condition: (form, density, access, safety for collection)
Mature mixed conifer stand – some hardwood component, medium density, easy access and safe for climbing. Permission from Beaty for collection access has been arranged.

RATING
(Check number that applies)

Species	PP	JP	SP	DF	CRW	WF	RF	IC	Blk. Oak
1. None					n/a				
2. Very Light								x	
3. Light				X					X
4. Medium	X		X			X			
5. Heavy									

The cone crop rating is based on ocular estimates of cone quantities on dominant and co-dominant trees, 12 inches d.b.h. or greater. Five classes are used as follows:

Rating	Criteria
1. None	No cones on any seed trees.
2. Very Light	Few cones on less than ¼ of the seed trees.
3. Light	Few cones on more than ¼ of the seed trees.
4. Medium	Many cones on ¼ to ½ of the seed trees.
5. Heavy	Many cones on more than ½ of the seed trees.

Figure 5.8 Example Cone Crop Survey Form

Judging "Few" and "Many"

Of particular importance is understanding the different cone production characteristics among the species being assessed, including the variance in number of cones considered abundant and their distribution throughout the crown. A cone crop rating of "few" or "many" is highly subjective and largely dependent on the experience of the surveyor. For example, 80 to 100 cones on a sugar pine or red fir tree would be classed as "many" while the same number on a Douglas-fir would scarcely be noticed. Likewise, several thousand cones may constitute "many" on a Douglas-fir but the same quantity would be rated as "few" on a mature incense cedar. In developing judgment of "few" and "many", attention is confined to the portion of the tree crown expected to bear acceptable cones for that species. In the white pines (sugar and western white) and true firs, cone production is often limited to the top four to five branch whorls. In abundant crop years, cones of ponderosa pine will occur in clusters of three to five or more on the previous year's growth in the upper half of the crown. Abundant small cones on coast redwood or incense cedar will alter the appearance of the tree as the branches literally droop under the weight of a heavy crop (fig. 5.9 to 5.21). For those species with cones found throughout the crown, the highest quality seed cones will be in the top half of the crown on vigorous branches exposed to the sun; seed cones in the lower crown are more likely to be self-pollinated. Additionally, isolated trees (individuals located more than 300 feet from any other of the same species) should be avoided in rating assessments and collections; they are likely selfed, or self-pollinated, and therefore not good genetic material.



Figure 5.9 (left) Coast redwood – few. **Figure 5.10** (center) Coast redwood – many. **Figure 5.11** (right) Coast redwood – many. *Photo:* Brian Barrett, CAL FIRE



Figure 5.12 (left) Douglas-fir – many. **Figure 5.13** (center) Douglas-fir – many. **Figure** 5.14 (right) Douglas-fir – few.




Figure 5.15 (left) White fir –few. **Figure 5.16** (center-left) WF – many. *Photo*: Brook Darley, CAL FIRE. **Figure 5.17** (center-right) Sugar pine – many. *Photo*: Craig Ostergaard, Sierra Pacific Industries. **Figure 5.18** (right) Sugar pine – few (right in photo).


Figure 5.19 (left) Ponderosa pine – few. **Figure 5.20** (center) Ponderosa pine – many. *Photo*: Dave Powell, USDA Forest Service. **Figure 5.21** (right) Sub-alpine fir - few-left, many-right. *Photo*: Canadian Forestry Service, 1989.

The cone crop surveys are completed in early summer when cones are readily visible, usually in June through July depending on geographic region, elevation, and cone year climate conditions. The surveys are done on a 'stand' basis within each designated seed zone and across elevation bands within a seed zone. Surveyors observe current year cones on well-distributed, dominant (and rarely co-dominant) seed trees. A seed tree is defined as one of cone bearing age, minimum diameter at breast height greater than 12 inches - often larger depending upon species, with good phenotype, fast growth rate, high vigor, disease-free, straight stem, and small branch size. Well-distributed means seed trees are 100 to 300 feet apart and stands are separated by approximately 600 feet – this large separation is required for wind-pollinated plants.

Surveyors must avoid two common errors when conducting surveys:

- Do not include spent cones (those that have already shed seed).
- Do not sample only roadside trees as they receive more light from all directions and will frequently have a heavier crop (generally not representative of the stand).

Visibility is improved if surveyors keep the sun behind them when viewing cone crops, especially species with small cone size such as Douglas-fir or coast redwood. The survey objective is to collect data from as many sites as possible to ensure a representative cross-section of cone production over a wide area. Surveyors are encouraged to evaluate 20 to 30 or more well-distributed inspection sites from low to high elevation per seed zone. Positive species identification is crucial. Sometimes identification is simple and straightforward because of the uniqueness of a species; Douglas-fir is one example. These cones are highly recognizable due to the characteristic three pronged bracts that overlap each scale. Other species, such as ponderosa and Jeffrey pine, are similar in appearance and have overlapping ranges. Evaluators must look at all species' characteristics to eliminate misidentification of similar-looking trees. In such cases, it is helpful to dissect a few cones and inspect seed structures as well.

Crop Rating

A crop rating is designated for each species present in the sample area and a separate survey form is used at each site evaluated. Based on the results of the cone crop estimates, potential collection areas can be selected for further evaluation. Each stand ultimately considered for collection must have sufficient area and number of acceptable cone bearing trees to meet collection standards and be cost effective. Additional information required on the Cone Crop Survey Form are details of the stand location and condition. It is important to be specific about location – a good motor log (in addition to GPS coordinates) is key to relocating a stand for further evaluation if necessary. The surveyor should also consider and catalog the features of the site when surveying for cone crops and choosing potential collection areas. Examples of useful site characteristics include the local topography and degree of slope, species composition and density, stand age and structure elements, overall stand health, and property ownership, if known.

If an area is selected for collection, then the microclimate features (aspect, slope, soil type and moisture holding capacity, drainage elements, etc.) will be recorded as well. Microclimates exist near bodies of water that moderate the local area. South facing slopes will be warmer and drier than those with a northern aspect. Low lying and shady areas will experience lower overnight temperatures and allow snow and frost to linger. Soils may be rocky or compacted, both of which will affect drainage. All elements of the site are necessary for consideration when making seedling deployment decisions and are included in a report of cone collection.

STEP 3 - Selection of Cone Harvest Trees

Desirable Characteristics

Characteristics such as growth rate and form, branchiness, and pest resistance are highly heritable and vary widely between individuals. Trees with a desirable phenotype may be genetically superior to neighboring trees with less desirable traits and often produce more viable seed. Forest tree selection for cone collection involves choosing parent trees that possess the following desirable characteristics:

- fast growth rate
- full, compact crown indicative of high vigor
- straight stem and minimal taper
- small branch size with horizontal or slightly upward angle
- high needle retention
- strong resistance to attack by insects and disease pathogens
- free of obvious defect such as forking or spiral grain

These genetic differences are important because they are passed on to progeny. Stands containing poorly formed and otherwise inferior trees should be avoided.

Genetic variation is necessary for populations to evolve and undergo adaptive changes in response to changing environmental conditions. The most likely place to collect seed with the genetic combinations that are best suited for long-term survival on a given site is on or very near the site itself. Decades of research has demonstrated that the use of non-local genotypes can result in immediate or delayed mortality, poor vigor and reduced fertility (Guinon 1992). In addition, non-local seed can introduce inferior or maladapted genotypes into the ecosystem. Failures from improper seed source may not become apparent for many years because of the slow growing nature of trees.

Broad or Narrow Genetic Base

The ability to adapt to different environmental conditions varies considerably among California tree species. Some species have more adaptive traits and are referred to as "generalists". Genetic generalists are varieties that perform well in a broad range of environments. Incense cedar, *Calocedrus decurrens* and western white pine, *P. monticola* are examples of species that can tolerate a wide range of elevational and climatic gradients and may be better able to adapt to a variety of environmental stresses. Ponderosa pine, *P. ponderosa* is one of the most widely distributed conifers in California and is characteristically present in the mixed conifer type from approximately 500 feet near coastal and foothill environments to more than 9,000 feet on the west slope of the Sierra Nevada. Ponderosa pine succeeds in a variety of soils, exposures and environments and is considered "intermediate" in adaptive traits. In broad terms, generalists and intermediates have more genetic diversity and can move farther in both seed zone and elevation than other species.

Douglas-fir, *Pseudotsuga menziesii* is known as a genetic or climatic "specialist" in adaptive traits. Specialists show strong genetic variances over small geographic and climate ranges. Douglas-fir has an extensive latitudinal range in the Pacific Northwest and is broadly distributed in California, but movement out of its local range typically causes decline in productivity or even death. The coastal and Sierran populations within California are morphologically and ecologically dissimilar and are not recommended as interchangeable. Measurable genetic differences can be found in Douglas-fir populations separated by just 200 m (656 feet) of elevational change and in response to temperature extremes, number of frost days and drought stress (Rehfeldt et al. 2014). Another specialist is bigcone Douglas-fir, *P. macrocarpa*; restricted to fragments of the Southern California forested region in canyons, canyon bottoms, and mostly northern and eastern slopes. Its endemic status in California and limited range of favorable environmental conditions of temperature, soil, and precipitation mark bigcone Douglas-fir as potentially maladapted to future projected environmental conditions. Adverse effects due to climate change are expected to have a greater impact on specialists and other marginal populations such as regional endemics, those at the trailing edge of range and high elevation alpine species, making them more vulnerable to extirpation if conditions change too rapidly.

Inbreeding

Within a stand, it is highly likely that many individuals are related. Conifers are wind-pollinated and therefore require a larger separation between seed trees than other plants to promote cross pollination; 200 to 300 feet apart is recommended. Selfing and close relative mating can have significant negative impacts on seed crops and tree populations produced from open-pollinated seed. Inbreeding results in loss of seed yield, lower germination rates, poor storability, poor seedling vigor, and higher mortality rates (Table 5.3).

Table 5.3 Inbreeding in Pinaceae

Trait	Loss Due to Inbreeding
Yield	-50%
Seed Germ %	-12%
Height Growth	-12%
Mortality	+1.3 times

Source: Franklin, 1970

It is essential to choose collection candidates with care. The best approach to avoid inbreeding and decreased vigor of progeny is to collect from a number of widely spaced trees in a stand, collect an equal number of cones from each, and from a number of different stands within the same elevation band of a seed zone. A stand is considered different when separated by a minimum of 600 feet from an adjacent collection area. With seed from many stands collections are more likely to include individuals adapted to local variations such as cold or drought hardiness, vegetative bud phenology, disease resistance, soil conditions, etc. In addition, collection priority for operational size collections should be given to stands and individuals at the center of the range. These are generally more broadly adapted and can be moved farther than material at the margins of a species range.

Location to location variation still exists, even among individuals and stands in the same elevation band. This is especially important on harsh sites or in regions that experience severe climate events. In this way the environmental, or climatic, distance between seed source and planting site is more important than geographic distance when considering a good match for deployment. Notably, collections from fragmented or threatened populations, or individuals at the trailing edge of the range where frequency is lower (areas typically excluded for bulk collections) should be considered a priority in future for gene conservation. These types of very specific collections should remain separate from operational-sized collections.

Selection of trees for collection in natural stands represents an opportunity for improving the overall health of the forest. While collection candidates are compared with adjacent trees for phenotypic characteristics, tree health or lack thereof is also critical to the success of collection efforts. Parent trees must show resistance to disease, a strongly inherited trait. Trees with mistletoe or other disease may have a heavy crop, but it is important to avoid collecting from these individuals or from stands containing diseased trees. Removal of diseased trees and general thinning is recommended to promote healthier and more resilient stands while also stimulating larger cone crops by reducing competition for light, nutrients and growing space.

Summary - Cone Harvest Tree Selection Standards (General Collections)

- Collect only from trees with desirable phenotype in stands with a high proportion of desirable trees – pollen from trees with poor form can affect the quality of progeny.
- Do not harvest cones in areas that had a poor pollen crop (as evidenced by observing the number of trees in the vicinity with abundant spent male cones present or on the ground).
- Collect only in years of moderate to heavy cone and seed production throughout most of the seed zone. Moderate means the majority of dominants and approximately 50 percent of the co-dominants are producing cones.

- Maximize diversity by maximizing the number of unrelated individuals represented.
- Avoid seed cones from trees with low filled seed counts (below 50% filled) that are not otherwise attributable to damaging agents such as frost or insects.
- Avoid collecting cones in the lower third of the live crown – there is an increased probability of self-pollination below mid-crown.
- Avoid isolated trees and small isolated stands – these are usually inadequately pollinated.
- Avoid stands or areas that have been selectively logged.

The goal is to collect seed lots that represent the maximum number of unrelated female parents – A minimum of 33-50 individuals is required for a 100 bushel size lot, ideally two bushels maximum per tree. Each stand collected from shall be separated by at least 600 feet.

Note - It is typical for some individuals in a population to produce more seed than others. Do not skew bulk collections by collecting more cones from "loaded" trees. An equal amount from each is necessary to maintain equal representation of donor genes.

STEP 4 – Cone Collection

The objective of any cone harvest is to collect the assigned quantity of high quality cones during the appropriate window of opportunity in a safe and cost efficient manner. Important factors to consider include:

- target species availability
- crop size
- collection timing
- collection site certification
- cone handling and transportation
- cost

Collections are inherently more efficient and successful when crops are bountiful and filled seed numbers are high. Yearly variation in cone production, collection timing, and seed quality can be very high so early determination of quality or lack thereof can prevent expensive failures.

Seed Maturity

Even with an abundant cone crop it is important to examine cones and seeds for quality and progress toward maturity. Seed maturity is vitally important! The relationship between seed maturity and germination capacity and yield is well established. Seed maturity has been defined as the stage at which seeds are capable of germination and successful storage (Edwards 1981). Seeds that are collected too early may germinate but they will be of low vigor and may perish in storage.

Mature seeds have greater vigor and establishment potential, higher yield, and increased storage capability. Seed maturity is generally associated with dispersal. The best time to collect cones is when they begin to open on the tree, but that is rarely practical with operational size collections. Instead, collectors must know how far in advance cones may be harvested without compromising seed quality. This may be a matter of days or up to three weeks in advance of natural seed shed depending on the species being collected and anticipated weather conditions. Cutting a sample of cones longitudinally and slicing the extracted seed is necessary for an accurate assessment.

Note: Specific gravity tests in the field had long been used to estimate collection start dates, but cone and seed cutting tests have become the preferred method because of increased reliability and ease of use in the field.

Evaluating the cone crop involves closely observing common indicators of ripeness including cone color and condition, seat coat and wing color and condition, evidence of insect or fungal damage, and internal tissue color, texture and condition. These physical and physiological indices are reasonably dependable but can be subjective and differences vary widely among species, between cones on the same tree, between individuals in a stand, and populations across the landscape. The reliability of these assessments is dependent on the experience of the observer.

Major Ripeness Indicators

As cones ripen, there is a gradual but obvious reduction in moisture content – they will “feel” lighter. This moisture loss is accompanied by changes in cone color and texture. Cone color varies by species but, in general, cone exteriors change from a vegetative green to some shade of yellow-green or golden to brown. There will be a slight flexing of the scale margins. Every effort should be made to collect when cones and seeds are fully mature. However “after-ripening”, a method infrequently used to improve cone and seed maturity when collections are premature, may be possible for particularly valuable collections (of DF, SP, or true fir) under strictly controlled conditions of cool temperature (10^{o}C), relative humidity, and sufficiently rapid air movement.

A representative sample is a must in order to accurately estimate the quantity of sound seed available and the degree of maturity attained – 2 to 3 cones from the top half of the crown from several unrelated trees in a stand is ideal. The sample cones are cut lengthwise through the center core and the exposed seeds are counted on one cut face. The seed minimum (average) per cut face varies by species but, in general, if 50 percent or more of the cut seed is empty or damaged then a collection is not considered worthwhile (fig.

5.22). The percentage of desirable seeds required increases to 75 percent for pines (fig. 5.23, 5.24). The seeds at the tip and base end of the cone are not considered in this count because, for most species, these areas likely contain a high percentage of undeveloped seeds (Table 5.4).

Figure 5.22 (left) Coast redwood - >75% filled. *Photo*: Bill Morrison, Soper-Wheeler Company. **Figure 5.23** (center) Sugar pine - poor filled seed count. **Figure 5.24** (right) Sugar pine – >75% filled.

Conifer seeds have three main components: the seed coat, embryo, and megagametophyte. Seed coat morphology among trees of different species is quite variable in color, shape, size, texture and the presence or absence of seed wings and resin vesicles. Even seeds from different trees of the same species will show great variability in seed color, shape, and size. As they mature:

- The seeds become easier to separate from the scale.
- The seed coat will begin to harden.
- The seed coat color changes from pale green, orange, magenta or tan-gold to golden, dark and/or mottled brown.
- The seed wing will darken, begin to dry and separate more easily from the scale, approach a golden to medium brown color, and feel somewhat brittle, similar to "parchment paper", when exposed to air.

Table 5.4 Major Ripeness Indicators

Selected California Conifers	Min. sound seeds per cut face [a]	Cone color and condition (not the best indicator of ripeness) [b]	Seed coat and wing color and condition	Embryo and Megagametophyte Color, texture and condition	Favorable collection period (wide geo-elevational variability) [c]
White fir *Abies concolor*	50%	Greenish-yellow to golden, may have a greyish or purplish tinge	Cream or tan color and soft, contains more resin vesicles than RF, loosely	Embryo occupies 90-100% of the cavity, yellow in color, cotyledons	True fir cones disintegrate fairly quickly

		at ends; Cone scales will become slightly pliable	attached to scale; wing is golden brown with magenta or golden margins and slightly translucent (if seed coats and wing all purplish still, it's too early), wings feel papery	well-developed; mega will appear whitish, fleshy and firm though may be somewhat oily in appearance	upon maturation, so must be collected slightly prematurely. Late Aug-October
Red fir *A. magnifica*	50%	Greenish-brown to russet brown with reddish or greyish tinge; cone scales will change from rigid to pliable before scales dehisce	Coat reddish-brown and thicker than WF, loosely attached to scale; Shasta RF have visible bracts on scale exterior; wing reddish-brown with magenta margin and papery	Embryo occupies 90-100% of the cavity, yellow in color (the radicle end is encased by a root cap); mega is fleshy and firm, somewhat oily in appearance	September through October
Incense cedar *Calocedrus decurrens*	2	Yellowish-green to golden tinged with shades of brown. Cone tip will flex slightly	Thin papery seed coat ripens to light tan color; has 2 persistent wings that must remain intact (to avert damage to embryo)	Embryo is bright yellow (may have pinkish radicle end) and 90% extended; mega is firm and oily in appearance	Mid-August to October Very perishable – be alert to overheating
Jeffrey pine *P. jeffreyi*	10	Greenish-purple to yellowish-brown or dk. purple to lighter purplish-brown, scales begin to flex	Coat pale brown color, hardened, smooth evenly brown on top, coarser on scale side; wing light tan to brown & brittle, adheres to seed coat	Embryo is creamy white and 90% length of cavity; mega is whitish color, opaque, firm and nutlike	Early September through October
Sugar pine *P. lambertiana*	12-15	Yellow-green to light brown; cone scales become less rigid and begin to flex	Seed coat is dark brown (orange & mottled orange-brown is pre-ripe color); wing is dark brown, thin and papery	Embryo is 90% length of cavity, yellow color; mega is creamy white, opaque and firm, not milky	Mid-August through October
Ponderosa pine *P. ponderosa, var. ponderosa*	8-10	Pale yellow-green to light brown-green to lustrous yellow-brown, scales begin to flex	Pale brown to grey-brown and usually mottled on scale side, hardened; wing golden to tan, brittle, adheres to seed coat	Embryo is 90% length, pale yellow color; mega is whitish color, opaque, firm & nutlike	Early August through September
Douglas-fir *Pseudotsuga menziesii var. menziesii*	5-6	Pale yellowish-green to golden brown color, 3-lobed bracts will	Golden brown to dk. brown, shiny and darker on one side, hardened; wing lt.	Embryo occupies 90-100% of the cavity, pale yellow color; mega is	Earliest ripening cone at lowest

		turn golden brown first (but may brown prematurely due to insect activity as well)	brown to tan, easily detaches from scale. Coastal seeds are longer with pointed tips; interior generally rounder tips and more triangular-shaped	whitish color, full, opaque and firm	elevations late July-August. Most seeds released by October
Coast redwood *Sequoia sempervirens*	3-4	Greenish-yellow to golden brown; cone scales separate slightly	Reddish-golden to red-brown (should not be green tinged). Wing is slightly lighter color and is part of the seed coat	Embryo and megagametophyte are barely indistinguishable without magnification, but whitish color and firm	Early October to November

[a] *For pine, DF, and RW – place cone on a hard surface and cut lengthwise from stem to tip through the center axis exposing the cut seeds, count the number of filled seeds on one half only. For true fir – cut cone lengthwise from stem to tip and parallel to the core but offset by ~¼ to ½ inch. The seed count is measured as the percent of exposed seeds that are sound. For incense cedar – cut cone through the bottom third widthwise – observe from one to four seeds per cone (rarely up to six).*

[b] *Determining maturity by exterior cone color can be very subjective; color varies by individual, population, site and weather.*

[c] *Physical characteristics vary with location and elevation. Maturity may differ by more than one month between low and high elevation sites. Seed dispersal is greatly influenced by local weather patterns. If unusually warm or drying winds, seeds of most conifers will disperse in a matter of days.*

Embryo and Megagametophyte

The seed cutting test is a seed anatomy test. It reveals the proportion of filled (presumably viable) seed and degree of maturity. A sample of seeds are bisected on the longest axis and thinnest dimension to visually examine the embryo condition and length; the cut must be precisely through the center because an improper cut will not show true embryo elongation. The number of seeds sampled varies by the precision required and whether it is a field test (10 to 20 seeds) or a post-harvest examination in the Seed Lab (100 seeds per lot). As it matures, the embryo loses moisture and differentiates from the megagametophyte, turns creamy to pale yellow in color and elongates to fill the embryonic cavity; embryos nearing acceptable maturity will fill approximately 90% of this cavity (fig. 5.25). The cotyledons develop fully and become visible at the rounded (chalazal) end and the hypocotyl and radicle extend toward the pointed (micropylar) end of the seed when fully elongated. A thin suspensor connecting the radicle to the micropylar end may still be visible if the embryo is underdeveloped (fig. 5.26). The color, texture and general appearance of the megagametophyte varies with the moisture content of the tissue as well. When immature, the megagametophyte appears gelatinous or milky (fig. 5.27). As it matures the color and texture changes; moisture is lost and the megagametophyte becomes opaque, firm and nutlike as it differentiates from the embryo. If cut while still immature, the megagametophyte will shrink away from the seed coat if it is left exposed to sunlight or air. In true fir and incense cedar the megametophyte normally appears somewhat oily because of the many resin vesicles within the seed coat. Damaged megagametophyte tissue will appear grey or yellowish in color and may have a rubbery or chalky texture.

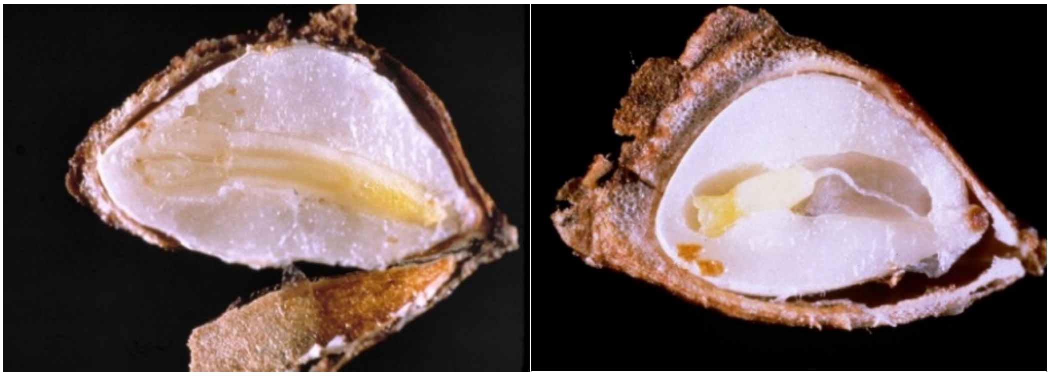


Figure 5.25 (left) *P. menziesii* seed **-** mature appearance with embryo fully extended (left).
Figure 5.26 (right) *P. menziesii* seed with embryo <50% and suspensor still clearly visible.
Photos: B. C. Ministry of Forests, 1989.

Figure 5.27 Immature *P. macrocarpa* seed: seed coat color pale, megagametophyte quite moist. *Photo:* Arnaldo Ferreira, USDA Forest Service, 2014.

Evidence of Insects

The presence of insects in cones is often externally apparent by one or all of the following: premature browning or flexing as a whole or in patches, small holes caused by boring, excessive pitch exudation, presence of frass, and areas of disfigurement (fig. 5.28, 5.29). Insect damage will result in lower seed counts and a decrease in the number of seeds that can be extracted from the damaged cones.


Figure 5.28 (left) Localized scale damage (darkened areas) on *P. menziesii* by *Contarinia oregonensis*. **Figure 5.29** (right) *P. ponderosa* damaged by *Dioryctria spp*. Cone scales patchy in color and frass grains at stem end. *Photo*: USDA Forest Service – Ogden, Bugwood.org.

Problems That Result from Collecting Immature or Insect Damaged Cones:

Many problems can be encountered with immature and insect damaged cones and seeds. Immature seeds are harder to extract and process because the cones tend to dry too rapidly once they are separated from the tree and the scales will case harden in a partially flexed position. This traps the seeds inside the cone and reduces yield. Immature seeds have reduced germinative capacity and they are slower to germinate. Germinants of immature seeds are less robust and produce seedlings with decreased vigor. Immature seeds are more susceptible to fungal attack and diseases, and they tend to have more abnormalities which leads to eventual death. When seeds are collected too soon they are less viable and will decline in storage at a much faster rate than seeds that are fully mature when collected.


Figure 5.30 (left) Excessive debris and pitch in this seed lot of *P. lambertiana* makes processing and seed upgrade difficult. **Figure 5.31** (right) *P. menziesii* cone heavily burrowed by *Choristonuera occidentalis*, western spruce budworm. Photo: Canadian Forestry Service, July 1980.

Insect damaged cones are harder to extract and process because they often are contaminated with frass and pitch exudation which fuses the seeds and scales, preventing the seeds from falling freely during extraction (fig. 5.30). In addition, insect activity often causes deformation of cones; the affected areas do not flex and the seeds are trapped (fig. 5.31). In the above situations there will be a significant decline in seed yield. During upgrade, seeds with an insect larvae inside may be more difficult to separate from filled seeds because they often have the same specific gravity – the result is a seed lot with fewer germinable seeds. Damaged cone fragments, seeds, frass and insects contaminate the seed lot and the equipment being used and make seed cleaning far more challenging. These seed lots must often be consigned to the end of the processing cycle, after the healthier lots are completed, because of the extra time and effort that they require.

Collection Timing is Critical

As noted, cones and seeds must be physiologically mature at the time of collection or seed viability and vigor will be unacceptable. In California, the majority of forest tree seeds reach physiological maturity in late summer and early fall about one to two weeks before natural seed fall. The actual timing of the collection and the length of the harvest period in a given season will depend on the species, area of the state, elevation of the site, and local weather conditions. Moisture, temperature and altitude play an important role in local ripening. For example, a warm, dry summer or sudden drying wind can cause early cone opening and seed release. Conversely, a late spring, cool summer or wet fall can delay cone ripening and lengthen the collection period. For the most part, cones ripen first at lower elevations and on west and south facing slopes and later at higher elevations and on eastern and northern exposures (Schubert, Adams 1971). Not all cones in a stand, or on a single tree, ripen at exactly the same time so it is best to wait until the majority are ripe than to begin too soon. However, collecting too late may be detrimental because seeds may be lost to seed shed, predation by animals or insects, or to seed deterioration (Karrfalt 2008).

Species with serotinous cones are an exception to the fall maturity and one to two-plus week collection window. The collection period for Coulter, Torrey, foothill and knobcone pines may instead be several months after fall maturity because resins prevent the scales from opening immediately after ripening. Collection of these species may be conveniently scheduled after other time-sensitive collections are complete.

> Serotiny is an ecological adaptation in conifers that prevents immediate seed dispersal upon maturation. The cones remain closed on the parent tree for a year or more after the seeds have matured. In some species of pine and cypress, serotiny is more severe and seed release usually occurs in response to an environmental trigger such as fire or after treatment artificially in a cone kiln at a processing facility. Cones that exhibit this type of serotiny include knobcone pine, Monterey pine, Bishop pine, Coulter pine from the central coast (seed zones in the 100 series) and the cypresses.

Giant sequoia, though not truly serotinous, is another interesting exception to the fall-collect pattern. Giant sequoia cones are persistent; they remain in the crown for many years past peak maturity and intermittent seed fall. Mature older cones (>6 years old – yellowish-green with brown stem/peduncle) may be collected at any time of the year but may have approximately ten percent fewer viable seeds depending on the age and condition of the cone. The very old cones (grey, usually lichen-covered) commonly contain very few viable seeds, having intermittently flexed their scales and released seeds during warm, dry periods over many years.

Fresh (three-year) cones are physiologically and morphologically mature in the fall thirteen months after fertilization and these cones open readily upon detachment and drying. However, first year cones (immature, just-fertilized females) may also attain nearly full size in this same time period (Sept to Oct) and have often been mistaken for mature cones (fig. 5.32). In one study on the University of California Whitaker's Forest adjacent to Kings Canyon National Park, researchers are looking at differences in seed yield and germination capacity between cones of different ages, where cone age is related to cone color and morphology (fig. 5.33, 5.34). The goal is to identify, for collection purposes, the ideal "collectable" cone. (Ken Somers, Center for Forestry, University of California, Berkeley, ongoing research, 2017).

Giant Sequoia Production Cycle

Year 1 Jan -------------------------------Jul-Aug--Dec
(male/female buds form)

Production
Year 1 --------------Apr-Early Jun---------------------Aug----------------------------Dec
Pollination (females small)
Fertilization (females ~50% size)

Production
Year 2 -----------------------Jun------------------Aug--------------------Oct--------------
embryo development slow (females ~75% size)
embryo dev. rapid (females ~full size)
Morphologically mature

Figure 5.32 Giant Sequoia Production Cycle.


Figure 5.33 (left) GS Tree #274 - June 2015, just pollinated female conelets (top), mature cone (bottom center). **Figure 5.34** (right) GS Tree #274 – June 2016, now-fertilized females (top). They must continue development before attaining full embryo maturity in Sept - Oct 2016. Same mature cone (mid-center). Photos: Ken Somers, University of California, Berkeley.

Collectors must be alert to sometimes subtle color differences between first year cones and those that are fully mature when fall collecting giant sequoia to avoid collecting unripe cones. Making this distinction is easiest in a late spring or early summer collection (late May to June) of giant sequoia cones that fully ripened the preceding fall when color and size differences between mature and immature cones are more conspicuous. Early findings show that seeds from delay-collected fall ripened cones consistently show higher germinative capacity and increased seed yield when compared to fall collected giant sequoia which may include unripe cones.

Cone and Seed Losses

Cones and seeds may fail, post development, for a variety of reasons. In a cutting test, the two most easily recognized causes for the failure of a seed to germinate are lack of a properly mature embryo and complete deterioration of all seed contents (Kolotelo 1997). Empty seeds are common among many conifers because they may still develop a seed coat and megagametophyte without fertilization. One example is *Abies* seed. Without a fertilized embryo the megagametophyte of an *Abies* seed will eventually deteriorate, leaving a seed coat that is woody and difficult to cut during sampling. Conversely, an unfertilized pine seed does not develop megagametophyte, therefore it will have a lower specific gravity and be easily detected by crushing during assessment, or later removed by air separation during upgrade.

Conifer seeds with partially or completely deteriorated contents (dead-filled seed) may have been attacked by pathogenic fungi or may be the result of insect damage. Several species of fungi are known to cause damage to conifer seed and seedlings. Particularly harmful are those belonging to the genus *Fusarium*, which causes pre- and post-emergence damping-off. The affected embryo and megagametophyte may still be visible but with obvious discoloration, may appear as a solid mass, or may be missing entirely. These dead-filled seeds often have the same specific gravity as healthy, filled seeds and are harder to separate during processing.

Seeds with resin vesicles (true fir, incense cedar) may be damaged by overheating and rough-handling at any stage post-harvest. Damage to resin vesicles will result in decreased germination or death of the seed. Seeds with damaged resin vesicles may emit a foul odor, have a tacky feel, and the seed coat may be discolored. It is important to keep freshly harvested cones out of direct sunlight and limit movement and potential damage to sensitive seeds during collection handling and transport. Do not stack the cone sacks in piles and do not throw them onto or off the truck while loading or unloading.

Cone and seed insects are the most significant cause of seed losses, post cone development stage, especially in years of light seed production. Coneworms, midges, seed chalcids and maggots may easily destroy 50 percent or more to nearly all of a seed crop in some years. An especially damaging insect affecting a majority of high value tree species is *Dioryctria*, a coneworm that bores through cones and seeds, feeding indiscriminately throughout the season. Coneworm caterpillars continue to cause damage in cones awaiting processing until they pupate later in the fall. If populations are especially high, it may be necessary to expedite seed extraction from affected seed lots and get the seed into coolers to slow further damage. Another harmful insect that attacks Douglas-fir, true fir, and pine species is *Megastigmus*, a seed chalcid. A single larva consumes the entire contents within each affected seed and remains undetected inside until a cutting test or x-ray radiograph reveals its presence. Some insect species' feeding or tunneling activities cause extensive seed yield losses as well by causing distortion to the cones or by fusing seeds to the scales, making seed extraction efforts difficult to impossible (See Table 5.5). The tendency of natural stands to produce large crops in some years and none for several years (periodicity) has led to biological strategies in which insect populations survive low cone production years by greatly reducing their numbers and then re-emerging in large numbers when cone production resumes, often destroying the entire cone crop (Kolotelo 2001).

Table 5.5 Significant Cone and Seed Insects

Significant Cone & Seed Insects								
	Host Conifer							
Insect	DF	WF/RF	PP	JP	SP	IC	Larva Color	Type & Extent of Damage
Cone Beetle *Conopthorus*			X	X	X		White, C-shape, lt. brown head	Bores into cone at stalk or base, killing cone-may see pitch globule here. Cones will be underdeveloped or brown & shriveled. Moderately common in some years – cones are not collectable.
Cone Moth *Barbara, Eucosma*	X	X	X	X	X		White w/ dark head	Burrows w/in cone. Cones are pitchy, brown & deformed. May see pitch & frass on cone exterior. Can see damage by mid-summer – extensive destruction of seed and scales. One larva can destroy 60% of seed in a cone, damage may be 50%+ of the crop.
Cone Worm *Dioryctria*	X	X	X	X			Amber brown to dark red-brown	Bores thru cone scales and seeds – feeds indiscriminately. May see round holes & coarse frass. Seeds may not flex in area of damage; cones badly distorted. May destroy most of a poor crop and >50% of a good crop.
Cone Scale Midge *Contarinia, Asynapta*	X	X					Small, bright orange to red	Deforms cone and fuses seed to scale, forming swollen galls. Greatly impedes to red extraction of seed from cones.
Seed Chalcids *Megastigmus*	X	X	X	X			Small, white, curved and footless	No external evidence of damage. One larva develops per seed and consumes all seed contents. The larva remains in seed or may see round exit hole where adult emerged. Damage is complete by harvest. A major pest on DF and PP.
Seed Moth *Cydia (Laspeyresia)*		X	X	X			White w/ black head	Consumes seed moving from one seed to another leaving each filled with frass, then bores into axis. Usually no external evidence of damage. Once in axis, no further seed damage is expected. May destroy 30%+ of crop.
Seed Cone Maggot *Earomyia, Hylemya*		X					White w/ black mouth hooks	Consumes seed & moves into axis. No external evidence of activity but will see holes on seed coat. Larva exit in late summer – damage is over by harvest. Abundant in true firs.
IC Tip Moth *Argyresthia*						X	Small, green w/red dorsal bands	Mines cones and seed. May destroy almost entire crop.
West. Spruce Budworm *Choristoneura*	X	X					Brownish head, body, blackish head capsule	Feeds on male and female flowers and bores into developing cones. May cause significant decline in seed production.
West. Conifer Seed Bug *Leptoglossus*	X		X	X	X	X	Adult insect w/orange & black bands on abdomen	Pierces seed scale & sucks out immature seed content, collapsing the megagametophyte and rendering it gray-brown; or may leave hard seed coat intact but the tissue inside becomes withered, off-color and spongy.

DF – Douglas-fir; WF – white fir; RF – red fir; PP – ponderosa pine; JP – Jeffrey pine; SP – sugar pine; IC – incense cedar.

Methods of Collection

Figure 5.35 Climber ascending a giant sequoia.

There are several different methods for accessing tree crowns for ripened cones on selected trees. There are advantages and disadvantages for each method listed in this section but the safety of the climber and the tree must be considered first and foremost. In California, collections in wild stands are usually done by trained individuals who free climb selected standing trees; gaining access to sturdy branches in the lower crown via rigging ropes or occasionally using special ladders. Entry into the lower crown of taller trees may be accomplished by firing rope up to 120 feet with a (Big Shot) sling or up to 200 feet with a crossbow to the first supporting branch and, using appropriate gear, rigging the ascent to the top of the crown (fig. 5.35). The climber then ties onto the bole for support and accesses cones at the ends of branches with pole pruners or special hook devices designed to pull the ends of the branch within easy reach for clipping.

Climbing spurs are infrequently used but should not be used on especially valuable trees or species with stems that could be easily damaged such as five-needle pines and white fir. Free climbing is practical in open grown stands of trees up to 100 feet tall. Branches must be well spaced and large enough to support the climber. Tools are hoisted up after the climber is safely secured in the tree crown. It is best to start at the top of the crown where the crop is normally heaviest and work down to the lowest acceptable collecting portion of the tree, usually mid-crown. Cones are bagged in the tree and lowered to the ground, or branch ends with cones attached may be clipped and dropped onto tarps when vegetation or other means of providing a cushioned landing is available. Sugar pine and true fir cones are known to suffer serious impact damage (shattering) when dropped to the ground, therefore should be bagged in the tree and lowered to the ground without exception (Lippitt, Griffis, unpublished data, L.A. Moran Reforestation Center 1986).

In orchard and plantation applications where tree spacing and terrain allows, truck-mounted ladders or hydraulic lifts are suitable to access tree crowns up to the designated height determined by the machinery in use. Impediments to this method are the high cost, availability and efficiency of the equipment being used, tree crown height limitations, and the enhanced organization of crew required on the ground.

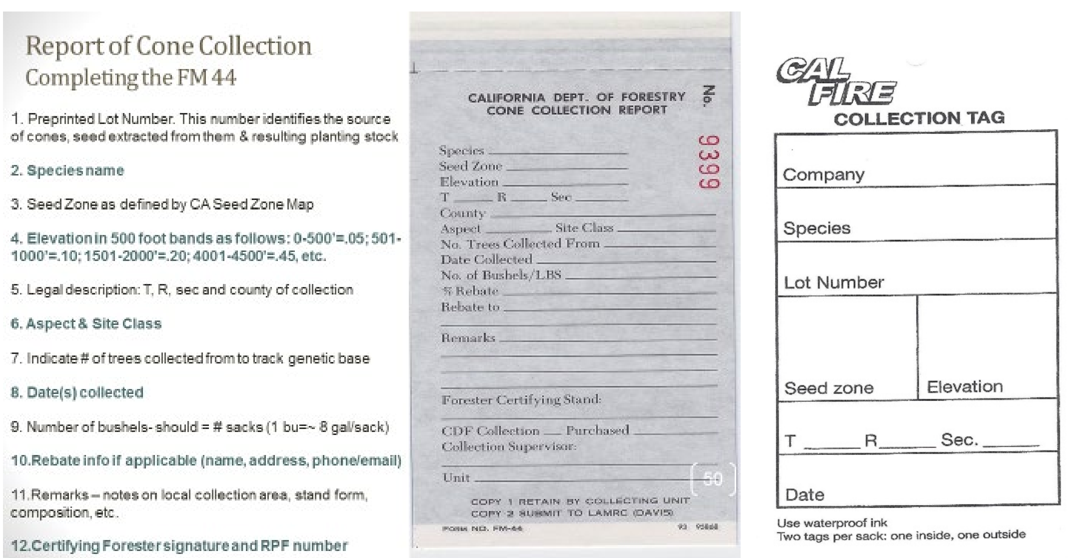
Helicopter collections using an aerial cone rake may be practical in coastal areas, on especially steep terrain, and other limited access areas where large cone crops are concentrated in the top whorls of narrow, conical crowns. This method has been very successful for coast redwood collections in California. Helicopter collections can be more economical when a large quantity of high quality cones are available but they are generally high cost, require highly trained pilots, are often hampered by unfavorable weather conditions, and pose a greater risk of damage to the trees.

Collecting from felled trees during active harvesting operations can make access to cones easier but this method offers many disadvantages. Timing is most critical: if falling occurs before the cones are properly mature, ripening will cease and the cones and seeds will dry out; if collected too late, the cones may open and seed may be lost. There is also a high risk of fungal contamination to cones through ground contact, a greater percentage of cone and seed losses due to impact damage and incomplete recovery, and high potential for injury to workers.

Seed Lot Identification

The importance of maintaining accurate seed lot identification cannot be overstated. The goal of conscientious labeling and record-keeping is to provide the most comprehensive seed collection and site specifications to forest managers so they have the tools they need to make sound deployment decisions. The collection supervisor (Registered Professional Forester or Certified Silviculturist) approving the cone collection shall record the species, location information including latitude, longitude and county of origin, date of collection, number of bushels collected, genetic base, and a complete profile of the collection site on a Report of Cone Collection. One example of a standard form for tracking seed lot archives is the FM-44 used by CAL FIRE. The seed lot data is recorded on this form during a mandatory site visit during the collection to verify the details of the collection. The FM-44 is a permanent record that includes a progressive numbering system identifying a given seed lot from collection through to nursery production (fig. 5.36). During collections, the seed lot number is written on each collection tag along with the species, seed zone and elevation, *latitude/longitude and/or township, range and section(s), and the date(s) of collection. Two seed lot tags are mandatory per sack; one inside, and one attached on the outside with a tie at the top – the inside tag is insurance against seed lot identity loss should the outside tag become detached. It is important to specify all information requested on the tags legibly with a permanent marker or waterproof ink to prevent fading or damage during inclement weather and to avoid seed lot confusion.

*Note: For decades, Township, Range, Section(s) was universally required to record location information but in recent years GPS coordinates have become standard. Computer programs easily convert one format to the other.



Report of Cone Collection
Completing the FM 44

1. Preprinted Lot Number. This number identifies the source of cones, seed extracted from them & resulting planting stock
2. Species name
3. Seed Zone as defined by CA Seed Zone Map
4. Elevation in 500 foot bands as follows: 0-500'=.05; 501-1000'=.10; 1501-2000'=.20; 4001-4500'=.45, etc.
5. Legal description: T, R, sec and county of collection
6. Aspect & Site Class
7. Indicate # of trees collected from to track genetic base
8. Date(s) collected
9. Number of bushels- should = # sacks (1 bu=~ 8 gal/sack)
10. Rebate info if applicable (name, address, phone/email)
11. Remarks – notes on local collection area, stand form, composition, etc.
12. Certifying Forester signature and RPF number

CALIFORNIA DEPT. OF FORESTRY
CONE COLLECTION REPORT
No. 9399

Species ____
Seed Zone ____
Elevation ____
T ____ R ____ Sec ____
County ____
Aspect ____ Site Class ____
No. Trees Collected From ____
Date Collected ____
No. of Bushels/LBS ____
% Rebate ____
Rebate to ____
Remarks ____
Forester Certifying Stand:
CDF Collection ____ Purchased ____
Collection Supervisor:
Unit ____
COPY 1 RETAIN BY COLLECTING UNIT
COPY 2 SUBMIT TO LAMRC (DAVIS)
FORM NO. FM-44

CAL FIRE
COLLECTION TAG

Company
Species
Lot Number
Seed zone | Elevation
T ____ R ____ Sec. ____
Date

Use waterproof ink
Two tags per sack: one inside, one outside

Figure 5.36 Samples of Agency collection documentation forms, FM-44 (left), Collection Tag (right).

Interim Handling and Storage

Cones and seeds are highly perishable. Correct field handling at this stage has long term effects on seed viability, seed longevity and seedling vigor. <u>Adequate ventilation and free air flow are the most important</u>

criteria. Even mature cones contain water – they expand as they dry and generate heat. Heat encourages mold and the webbing formed by mold results in direct damage to the seeds and hinders seed extraction efforts. Mold on the seed surface can compromise germination and subsequent storage life. Collections of true fir and incense cedar are especially prone to such damage because of their characteristically high moisture content at maturity.

Collection sacks should be constructed with strong mesh fabric, or burlap sacks may be used if not too heavy or woven too loosely – they must allow free air flow but disallow seed loss through the open weave. One bushel of cones per two-bushel sack is ideal, filling only to the half-way mark and fastened at the top, to allow ample room for cone expansion. Overfilled sacks damage seed; cones are more prone to mold or compression-hardened scales when they remain wet longer due to over-crowding. Debris in cone sacks such as needles, twigs, bark, and unacceptable cones should be avoided. Such debris are known to cause abrasion, bruising and tissue damage to seeds and result in decreased seed viability and reduced yield. Inclusion of spent cones may cause contamination of the seed lot with fungal spores.

In the field, interim cone storage racks and portable fans allow for unrestricted air movement. If racks are unavailable, a building with open sides may provide good cross ventilation. The sacks must lie free from each other in the shade, turned at regular intervals (daily if not on racks, every other day otherwise) to facilitate even curing and avoid case-hardening, and never left in piles on the ground. Avoid heating by direct sunlight and additional moisture from rain. Wet cones should be spread out to surface dry and re-sacked as soon as possible into dry sacks. Cone sacks should not be covered with plastic while interim stored or during transport. To minimize damage to cones and seeds from overheating, the cones should be transported to the processing facility as soon as possible and must not be left in a truck overnight. For best results, cone delivery within twenty four hours of collecting is highly recommended. Open-sided trucks or trailers with netting to stabilize the load and refrigerated vans are the best options. Closed vans without refrigeration should never be used to transport cones and seeds! The field supervisor should verify that each cone sack is properly tagged, provide an inventory by seed lot, and notify the extractory of the estimated time of arrival to expedite unloading of cones.

STEP 5 – Cone and Seed Processing

Cone and seed processing begins with the arrival of cones at the extractory and ends when the upgraded seed lots are entered into long-term storage. When cones arrive at the processing facility the cone/seed lot inventory and identification tags are verified and corrected, if required. Any cone sacks that may have been damaged in the field or during transport are identified and replaced to prevent loss of seed. A random and representative sample is withdrawn for a pre-conditioning assessment using the cutting test procedure described earlier. Adequate sampling is a must – a sampling of cones from ten percent of the cone sacks is standard. For example, a 100 bushel cone lot necessitates a minimum of several cones from each of ten individual sacks, randomly selected. This assessment will address the current condition of the cone lot: maturity level, insect and disease activity, potential yield, and need for proper cone curing. It will also provide the basis for prioritizing processing timing and define the subsequent cone and seed handling activities necessary for a successful outcome.

For most species, cone and seed processing involves several standard steps: drying (air dry, then kiln), extraction (cone tumbling), pre-cleaning, de-winging, seed processing and upgrade (with screens, pneumatic separator, or gravity table), final cleaning, and seed drying, if necessary. A schematic is presented in Fig. 5.37.

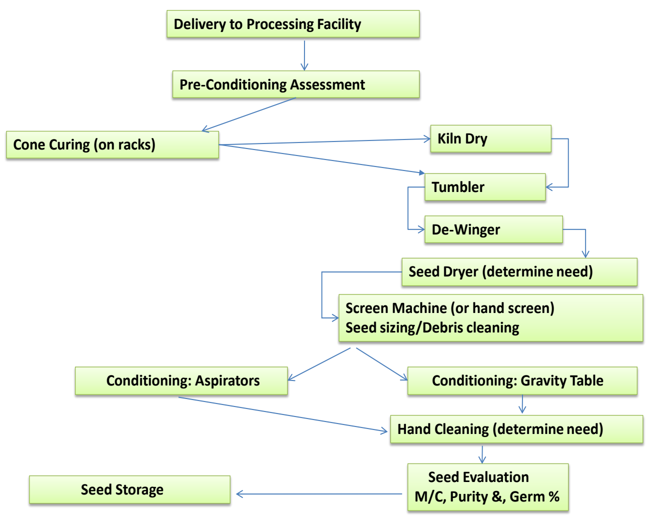


Figure 5.37 Typical cone and seed processing flowchart, sequence may vary.

Figure 5.38 Cones air drying on racks in a cone shed.

Cone Curing

Once delivered to the extractory, the cone sacks are spread out on steel mesh racks and stored under an open sided structure out of the direct sun and rain but exposed to good airflow from all directions (fig. 5.39). The sacks are turned every other day to promote gradual moisture loss and uniform curing and to prevent overheating and fungal growth. The mesh sacks permit visual monitoring of cone curing progress. As cone drying continues and the cone scales open and release their seeds, the frequency of sack turning may be reduced over time to one or two times per week. The length of cone storage (curing) varies by species and initial cone condition, but is typically two to four weeks.

Kiln Drying

Kiln drying refers to the cone drying process in which temperature, air flow, and in some facilities, relative humidity are carefully controlled. Kilning causes the cone scales to flex and open, allowing the seeds to be extracted. The temperature and duration of kilning may vary by species and cone condition but is generally effective between 32-37°C and 14-18 hours duration. It is important that cones be properly air dried before kilning to avoid a condition called case- hardening; the result of the outer layers of the cone drying more quickly than the moist interiors. In case-hardened cones, the scales freeze in a partially open position and trap the seeds inside causing significant to complete loss of seed yield.

There are two main types of kilns: batch kilns and rotary kilns. The batch type involves placing a thin layer of cones into trays fitted with a screened bottom, stacking the trays four to six high and capping the top box. The trays are placed onto plenums in the kiln which allow for heated air to course through the stacks of trays evenly at moderate velocity. The operational size kiln at the L.A. Moran Reforestation Center is capable of drying up to 300 bushels of cones from multiple seed lots at one time (fig. 5.39). A rotary type kiln combines drying and tumbling in the same phase and releases the extracted seeds from the drum to exit the heated and dry environment (fig. 5.40). The rotation speed and duration can be adjusted but this kiln type is restricted to processing a single seed lot at a time.

Figure 5.39 (left) Batch Kiln - kiln boxes loaded onto plenums for cone drying (LAMRC, Davis).
Figure 5.40 (right) Rotary Kiln, conveyor and cone tumbler (USDA Forest Service, Placerville).

There are variations to the above, of course. Species with serotinous cones, for example, generally require a hot water treatment and high temperature kilning to break a resin seal that prevents the scales from opening readily. One effective treatment is a 60-second boiling water dip followed by 18 to 20 hours of kiln drying at 50°C. Alternatively, serotinous cone collections may be delayed until early spring when the cones still retain significant moisture from the recent wet season. These cones will generally open with kilning alone, without the cumbersome and time consuming hot water treatment.

Another processing variation involves species that have soft and fragile seed coats. These types of seeds are prone to serious damage, especially to the resin vesicles in their seed coats. True fir and incense cedar are especially sensitive to heat and are easily damaged in a kiln. They may also sustain mechanical injury during de-winging and screening; all of which makes them particularly challenging in the extractory. When true fir cones have been allowed to dry naturally on racks, little or no additional drying may be needed because the cones disintegrate rather than flex to open and the seeds generally separate from the axis and scales readily. If kilning is necessary, the cones are spread out sparingly in the drying trays, the temperature should be ambient (may not exceed 30-32^{o}C), and kilning should be of short duration. Subsequent tumbling is usually cursory to separate the scales from the winged seeds or to help break apart any cones that did not dehisce during drying.

Incense cedar is best dried naturally in mesh sacks arranged flat on racks in a shady, well-aerated area. The sacks may be turned periodically for even curing but in a lifting and rolling motion, very carefully, to avoid piercing the seed coat and resin vesicles with needles or other sharp debris that may be present. Extraction is handled in small batches in a screened drum which separates the trident-shaped cones from the seeds. Alternatively, incense cedar cones may be separated from the seeds using hand screens.

Tumbling

Following batch-type kilning, the cones are transferred to a tumbler for extraction. The tumbler drum is fitted with a mesh screen sized to allow the seeds to fall through to a container below while the drum rotates and spent cones are disposed of through an opening in the drum at the end of the rotation. The length of the tumbling phase is vital to seed lot yield and quality; removal of all viable seeds is essential but the process must stop short of damaging seeds through excessive tumbling or introducing excessive debris from broken cones and scales into the seed lot. After a seed lot is passed through the kiln and tumbler, the seeds may be temporarily collected in rigid plastic boxes or fiber drums and interim-stored in a cooled environment (3-5^{o}C) to facilitate easy handling of seeds during subsequent processing steps. The cool temperatures also are effective in slowing down insect activity in a seed lot when it exists.

Screening

After seeds are separated from the cones, seed lots that contain considerable debris may be passed through a screen machine with air flow over a series of slotted, round-holed or mesh screens to pre-clean the lot before further processing. Screen size and shape is varied to suit the species being cleaned and the debris that is being removed. This process is called scalping and it is effective at removing cone scales, needles, pitch, small rocks, and other inert matter that could cause injury to the seed (fig. 5.41). Incense cedar is not screened using this process to avoid unnecessary damage.

Screening may also be done with a variety of hand screens similarly fitted with different sized openings that allow the target material to pass through. Note that a custom determination of screen size and shape is needed for each seed lot, even if the species is the same, since size of seed and debris may differ. Pre-cleaning the seed lot increases the efficiency of the processing steps to follow. Incense cedar is commonly screened using hand screens.

De-winging

De-winging is the process that removes the seed wing from its attachment to the seed coat. Seeds of most species may be de-winged by one of several methods. By far the majority of species are de-winged using a rotary drum specially designed to control rotation speed and angle and apply moisture if needed (fig. 5.42). Species that are "wet" de-winged include some pines and spruces which have seed wings that are weakly attached to the seed coat via the integument. To remove these wings, a short spray of water mist is intermittently applied while the seeds are rotating in the drum, causing the seed wing to expand at this connection and cleanly detach from the seed coat. In a slightly different scenario the seed wings of true fir and Douglas-fir, which are an integral part of the seed coat, must be broken off by mechanical friction. De-winging of these species is done with no added moisture – the paddles within the rotating drum allow the seeds to gently roll over one another, breaking the wings, though somewhat imperfectly and frequently incompletely. If the seed moisture content is high, then drying the seed lot to below 15 percent of fresh weight may result in a more brittle seed wing and more complete de-winging. For both wet and dry de-winging methods the seed wings then may be gently blown off in the de-winger with an air hose set on low or separated later with a pneumatic separator. It is important to note that an oil-free compressor should be used for blowing. Alternatively, removing the seed wings of small size seed lots (hard-coated seeds only) may be done by gently rubbing the seeds between one's hands. For each method listed it is important to avoid prolonged de-winging to minimize seed injury through bruising or abrasion. Seeds with resin vesicles such as *Abies spp.* are particularly susceptible to damage. It is recommended that de-winging be carefully monitored and be as brief as practical.

Figure 5.41 (left) Screen machine (or Scalper). **Figure 5.42** (right) Custom seed de-winger.

Figure 5.43 Incense cedar seed with radicle emerging from wing end. *Photo*: Dorus Van Goidsenhoven, CAL FIRE

Several species in California are not routinely de-winged. Incense cedar seed is one; the embryo is reversed in incense cedar with the radicle emerging from the seed wing end (fig. 5.43). This anatomical anomaly makes significant damage to the seed unavoidable with attempts to remove the wing. Lastly, there are several genera including *Sequoia*, *Sequoiadendron*, and *Cupressus*, which have very small seed wings that are tegumentary extensions of the seed coat and no attempt is made to remove them. The damage to these seeds would be severe.

Seed Upgrade

Seed processing deals with upgrading the seed lot. The reasons for upgrading seed lots are many and include the following: reduce bulk and weight, increase seed lot purity, increase germinative capacity by removing empty and damaged seeds, improve seed storage life, and make seedling production easier and more economical. Seed upgrade removes empty, immature and non-viable seeds, plus any fine debris that may still contaminate the seed lot. There are two main types of mechanical separators commonly used for this purpose: the pneumatic separator (also known as an aspirator or air separator), and the gravity table.

The pneumatic separator utilizes an adjustable air column fitted with a vacuum, or blower. A vibratory feeder is used to deliver the seed into the air column (fig. 5.44). Seeds are separated based on differences in weight. The air velocity can be manipulated to capitalize on the differences between the target seeds and the debris being sorted. There are several outlets for seed discharge in descending order of material weight (product-heaviest — lighter — lightest). An x-ray radiograph or cut test is used to calibrate the air flow settings and determine if the separation is acceptable; each seed lot requires its own unique settings. The goal is to separate the heavier seeds which are presumed to be filled and viable from lighter fractions which may contain partially filled, empty or otherwise undesirable seed and debris. While it may be tempting to use a stronger airflow setting to remove a greater quantity of empty or partially filled seed or debris, care must be taken not to use too strong an airflow if it also removes a significant number of small, filled seed with similar weight to larger but empty seed. Inadvertent removal of the smaller filled seed component would effectively reduce the genetic base one has worked so hard to achieve. If necessary, it is possible to re-run the seed in a discharge bin with an adjusted airflow setting when it contains a combination of filled and undesirable seed. Seed lots are often run through an air separator more than once with progressively finer-tuned settings to achieve the desired end product.

The vibratory feeder speed can influence upgrade results as well. Excessive speed may result in too much seed in the air column at one time, preventing the light seed from being drawn up into a discharge fraction. A slower vibratory feeder speed may permit a better separation but may take much longer and tie up the separator. A proper balance between processing efficiency and processing accuracy is desired. Pneumatic separators are available in various designs and sizes (fig. 5.45).

Figure 5.44 Wall-mounted pneumatic separator (vacuum version).

Figure 5.45 Four-chamber forced-air separator (free-standing).

Figure 5.46 Air gravity table.

The gravity table utilizes an inclined deck that moves in two directions – up and down, and backwards and forwards. The deck is covered with a mesh cloth that allows an air current below the deck to move the seeds based on specific gravity (fig. 5.47). When seeds are lighter they are lifted off the deck slightly and move toward the lower end of the deck. If the seeds are heavier and in contact with the deck, they move toward the upper end of the deck and can be collected in different fractions based on placement of dividers on the deck that are manipulated by the operator and based on seed weight. The seed lot is separated into heavy, light, and lighter fractions and collected in separate bins. An x-ray radiograph or cut test is used to calibrate the air flow and the settings related to deck tilt and the position of the dividers. Each seed lot requires its own custom setting. It is common to run a seed lot more than once with adjustments to these settings. A gravity separation can be an effective option for species that are more vulnerable to damage such as true fir, but the equipment requires a highly experienced operator to manage the many variables of the different settings and for a successful outcome.

Incense cedar seed poses a challenge to seed processors at every stage. Mechanical separation tends to be harsh on the delicate seed coats. A series of hand screens with slotted openings may be used to dispose of debris and the flat (empty) seeds, but care must be taken to avoid rubbing the seeds against the screen and causing damage to the seed coat and resin vesicles. The intact seed wing makes air separation challenging as well because of the natural lift of the wing by the air column, but it can be done. Incense cedar seed is easier to work with when chilled at 2 to 4^{o}C prior to separation. Using a slow speed on the vibratory feeder and a low vacuum setting for a light separation, the air column will pull light and empty seed up and out of the column with minimal effect on the wings of heavier (filled) seeds. The process is purposefully slow but suitable for smaller seed lot sizes (20 bushels or less) with minimal damage.

Hand cleaning a seed lot is the final stage in seed processing and cleaning. It is important to maintain a strict purity standard of 98 percent or higher of pure seed per unit weight for a variety of reasons: purity percent is one of the essential parameters when determining quantity of seed needed for nursery sowing; removal of impurities reduces bulk and the amount of storage space needed; and fewer impurities reduces the potential for injury to seeds in storage. After final cleaning, the seed lot is mixed thoroughly for homogeneity before testing and extended cold storage.

In each stage of cone and seed processing and upgrade, one must be careful not to over-process; seeds that do not possess a hard protective seedcoat are particularly susceptible to damage from heat, bruising, breakage, and abrasion. In practice, seeds should not be handled when conditions are too warm or if the seeds feel "sticky".

Avoid cross-lot contamination. The most important consideration during each of the cone and seed processing steps is to avoid contamination between seed lots. A thorough cleaning of all equipment and tools used is essential and may involve any or all of the following: vacuuming, brushing, sweeping, blowing, electrostatic cleaner, de-pitching, steam cleaning, and sterilization.

STEP 6 – Seed Lot Assessment

Analytical tests are performed on seed lots to determine:

- moisture content
- seed purity percent
- seeds per pound
- percent filled (by x-ray)
- germination percent

The most significant aspects of good seed testing are proper sampling and uniform testing procedures. Guidelines for these tests and sampling procedures are found in the Association of Official Seed Analysts (AOSA) and the International Seed Testing Association (ISTA) rules. These tests provide the basis for seed lot evaluation and for determining the number of seeds needed for sowing in the nursery. Additionally, knowledge of the quality of the seed lot(s) and the number of potential seedlings available in storage for reforestation is crucial to future collection planning efforts.

Sampling

Sampling, first and foremost, must be random and truly representative of the entire seed lot to be of value. Drawing the sample may be done with a seed probe (or trier) or by inserting an open hand into the seed lot to a sampling point and withdrawing the closed hand. If there are one to five containers, a primary sample must be taken from each and mixed thoroughly; if there are more than five containers, five of them plus ten percent of the remaining containers are required. This blend of primary samples constitutes the composite sample from which a smaller "working sample" is taken (approximately 2500 seeds). Sampling and seed testing are routinely conducted on-site at seed processing facilities in California rather than shipped to a separate seed lab for testing; therefore the sampling procedure described above to obtain the working sample follows the seed moisture test. After seed moisture content has been determined to be in the "safe" storage range of 5 to 9 percent, the working sample may be drawn.

Moisture Content

Measurement of seed moisture content is vital in preparing seeds for long term storage. Moisture content is the most important factor in viability retention. Correct low moisture content and low storage temperature assures metabolic rates are minimal and fungal and insect activity ceases. If seed moisture content exceeds the standard 5 to 9 percent threshold for storage, the seed lot must be dried down by one of the following methods: kiln drying for larger lots, small lot seed dryer, or solar drying. Seed moisture content may be measured by one of several methods including the oven-dry method or a rapid method using an electronic moisture meter. The oven-dry method is a destructive test but is acknowledged to be the most accurate. In the oven-dry method, duplicate samples of 5 g each per seed lot (3 g each for RW and GS) are dried in a forced-draft oven (for even, accurate drying) at 100^{o}C for 17-18 hours. It is important to use an accurate balance to three decimal places. At the end of drying, the samples are covered with a close fitting lid and cooled to room temperature (30 to 45 minutes), then re-weighed. The moisture content is expressed as the difference between the fresh weight and dry weight of the seeds, as a percentage of the fresh weight:

$$\text{Moisture content} = \frac{\text{fresh weight} - \text{dry weight}}{\text{fresh weight}} \times 100.$$

If the difference between the two samples exceeds .03%, ISTA rules require that the procedure be repeated.

An electronic moisture meter (Dole Meter Grain Tester) is one type of quick test and is generally accurate to within +/-1 percent of the oven dry value. It is non-destructive and commonly used for small seed lots or for a quick moisture check during processing. Logical timing during seed processing when a moisture check may be appropriate includes after "wet" de-winging or during an air separation run in which an effective separation falls short of expectation. Moisture values using a Dole Meter are generally more accurate when the seed is at room temperature. A disadvantage of the Dole Meter is that it requires calibrated values for each species that must be determined in advance. Calibrations have been done for the following: bishop, coulter, Jeffrey, Monterey, ponderosa and sugar pines, coast redwood, giant sequoia, grand, white, and red fir, Douglas-fir, and incense cedar. Moisture content also directly influences the relationship of weight to the number of seeds of a specified quantity, so the seed lot should be in the 5 to 9 percent moisture range before further tests are done.

Seed Purity Percent

The seed purity test defines the composition of a seed lot based on the weight of pure seed and the weight of any impurities contained within it. Using the working sample, one thousand seeds are counted out in groups of 100 and maintained in separate piles. Other crop seed or debris that is associated with the sample as it is counted are maintained as separate components. If cracked, broken or otherwise damaged seed is encountered, it is counted as crop seed if it comprises more than one-half the size of the seed. Damaged seeds in the bulk lot will reduce the germination percent and must be included in the purity and seed count determinations. The fractions are weighed separately and the purity is expressed as a percentage of the total weight:

$$\text{Purity percent} = \frac{\text{weight of pure seed (g)}}{\text{weight of pure seed + other crop seed + debris (g)}} \times 100.$$

The purity percent standard for tree seed at the L.A. Moran Reforestation Center in Davis is ≥98 percent. If impurities or overall debris comprise two percent or more of the seed lot, further cleaning is required.

Seeds per Pound

The number of clean seeds per unit weight of the seed lot (in pounds) is calculated using the pure seed and companion components from the final purity test:

$$\text{Seeds per pound} = \frac{\text{\# of seed/Lb.}}{\text{g/Lb (453.6)}} = \frac{\text{1000 seeds}}{\text{weight of pure seed + debris (g)}}$$

$$(\text{\# of seed/Lb.}) \times (\text{weight of pure seed + debris in g}) = 1000 \times 453.6$$

$$= \frac{453{,}600}{\text{weight of pure seed + debris (g)}}$$

The purity percent and seed per pound calculations are important factors for determining nursery sowing requirements. However, they do not provide any information relating to the tree-producing capability of the seed lot.

X-ray Technology

Since the mid-1980s, the use of high resolution x-ray images has become an increasingly important tool for initial evaluations and in the processing, upgrade and analysis of conifer tree seed. Radiographic images are non-destructive, as opposed to cutting tests, and provide valuable information on the interior morphology of the seed and the presence of insect larvae, abnormalities or disease. The x-ray cabinet component utilizes low-energy (soft) x-rays only and is deemed safe when personnel are properly trained and using appropriate precautions. The risk of damage to seeds by x-ray is considered minimal to none. Digital systems are available that can enlarge the image of the seed to allow more definition and apply coloration to tissues based on density. Long term storage and transfer of digital images has become convenient and routine. Preliminary and periodic x-ray images are used to determine appropriate settings for seed processing equipment and also to confirm separation progress during upgrade (fig. 5.47). Small seed lots, or especially valuable or delicate seeds that may be damaged using mechanical separation methods, may be sorted using celled acrylic trays that enable hand selection of higher quality seed. Blister rust resistant sugar pine seed lots and seed lots of endangered species have been processed using trays and hand selection (fig. 5.48).

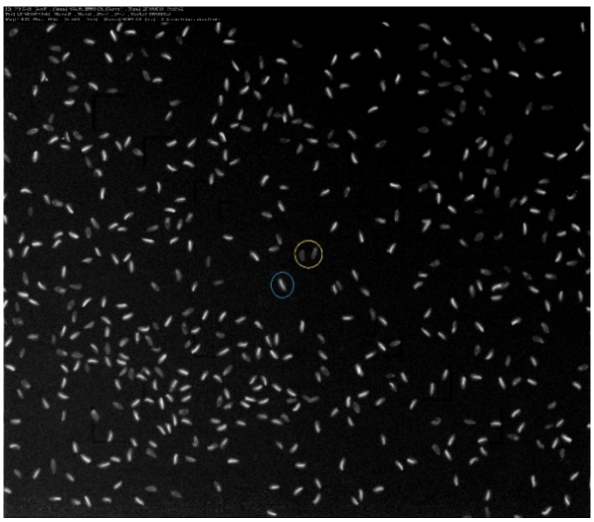


Figure 5.47 Giant sequoia seed (filled seed-blue circle; empties – yellow circle).

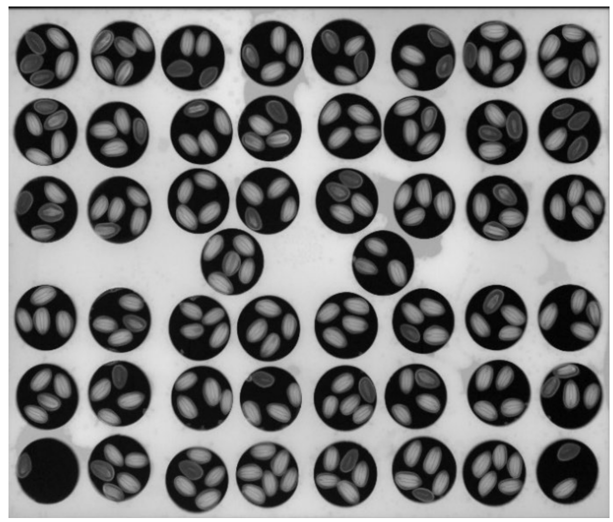


Figure 5.48 Sugar pine seeds trayed for upgrade, by x-ray. The empty and damaged seeds are removed using forceps.

The pure seed component from the purity and seed per pound analysis, above, allows the seed technician a quantifiable means to estimate germinability of the final seed lot, via x-ray image, and compare it to the laboratory germination result. Initially, interpretation of x-rays may be subjective, but over time the seed technologist acquires the aptitude to reliably correlate an x-ray image versus a germination test result. Admittedly, such correlations are better for some species such as redwood and less reliable for others, such as white or red fir.

Germination Percent

The laboratory germination test determines the quantity of seedlings that may be produced from a seed lot under optimal, controlled conditions. The germination capacity (GC) is the chief standard used to define seed lot quality – this is the percent of seeds that germinate <u>normally</u> during a specified period, usually 28 days. The germination value (GV) is a measure of vigor, or speed of germination. An official germination test consists of 400 seeds (four replicates of 100 seeds each) selected at random from the pure seed component of the purity and seed per pound analyses (fig. 5.49 and 5.50). A second 400-seed test is reserved for a germination retest after one year in long term storage and is enclosed within the storage container that holds the bulk seed lot until needed. Subsequent germination tests are conducted at three to five year intervals to insure that recent and reliable test data is available for nursery growing.

Figure 5.49 (left) Coulter pine seed germinants (day 7) on moist substrate (perlite). **Figure 5.50** (right) Ponderosa pine seed germinants (day 14) on moist substrate (cellulose paper)

Figure 5.51 White fir seed germinants (day 14). Germinants on left (3) show normal development of hypocotyl and radicle. Germinants on right (3) show abnormalities (stunted tissues) and fungal growth (middle-right.).

Not every seed that germinates is included in the germination count; only those that are defined as normal according to the standard guidelines (AOSA). A normal seedling is described as one that possesses the essential structures that indicate its ability to produce a normal plant under favorable conditions. The most common abnormalities identified in a germination test include a breached embryo, in which the cotyledons emerge before the radicle, stunting of tissues such as the hypocotyl or the radicle, or rapid internal fungal growth (fig. 5.51).

Germination percent is calculated by the following:

$$\frac{\text{Number of normal seedlings}}{\text{Number of seeds sown}} \times 100 \text{ (nearest whole number; do not round up)}$$

The four replicates are compared - if they are within the accepted range of tolerance established by the rules, the average of the replicate values is the germination result for the seed lot. If the replicate values are not within the established tolerance, the germination test must be repeated.

Standardized germination tests are designed to provide ideal conditions for maximum values with minimum variation and achieve results that are reproducible. The pretreatment regimes that have been adopted as the recommended procedure for each species are the result of research and comparative tests over time that produced the highest germination results with the least variation.

Standard testing protocols for several conifers in California are listed in Table 5.5.

Seed Dormancy

Seed dormancy is a condition of viable seed that prevents it from germinating even when exposed to suitable environmental conditions. Dormancy is the mechanism in conifer seeds that eliminates the risk of immediate germination after natural seed fall in autumn when conditions are generally unfavorable for growth and survival. Most California conifer seeds have the type of dormancy known as morphological (or physiological) dormancy. This type of dormancy is thought to be a consequence of immaturity within the internal seed tissues. In nature, the cool moist conditions of winter allow the seed to absorb moisture and advance the after-ripening processes, allowing germination the following spring. These conditions are mimicked in the seed lab and forest nurseries by soaking the seeds in water followed by moist chilling in a refrigerated chamber at the appropriate temperature and duration determined by experience for each species (Table 5.5). This process is called stratification or moist pre-chilling - the terms are used interchangeably here. In some pines, such as Coulter pine and foothill pine, the seed coat thickness may act as a barrier to moisture and oxygen exchange and can delay swelling and imbibition of the embryo. This type of seed coat dormancy is overcome simply by a longer soak in room temperature water (48+ hours). The longer water treatment tends to soften the seed coat and allows for imbibition which is then followed by an extended period of stratification.

Table 5.6 Germination protocols recommended for the listed species

Species	Imbibition # hours	Stratification [a] # weeks	Special Protocols	Germination Light-Dark [b] Temp °C	Test Duration # days
White fir, grand fir	24	6		25-15	28
Douglas-fir, Coast redwood, Monterey cypress	24	6		30-20	28

Red fir, Shasta red fir	24	10		25-15	28
Incense cedar, Bigcone DF, Sitka spruce, Tecate cypress	24	8	—	30-20	28
Giant sequoia	48*	6	*Running water rinse	30-20	28
Lodgepole pine	24	4		30-20	28
Ponderosa pine (low-high el.)	24	(4-8)		30-20	28
Ponderosa pine – orchard	24	8		30-20	28
Pinyon pine, Santa Cruz cypress	24	12		30-20	28
Jeffrey pine, Knobcone pine	48	6		30-20	28
Coulter pine	48	10		30-20	28
Torrey pine, Western white Pine	48	12		30-20	28
Sugar pine	48*	13-16	*Running water rinse	30-20	28-35
Foothill pine	48	16		30-20	28

[a] *Cold stratification is most effective between +2 and +4 degrees C.*

[b] *8 hours at higher temperature with illumination and 16 hours at lower temperature in dark.*

Methods to Prepare Seeds for Germination in the Laboratory

The degree of dormancy and the optimal length of stratification varies greatly between species and also among families of the same species, though differences among the latter can generally be attributed to geographic origin and latitude or elevation of the seed source. Moisture availability is the primary factor controlling germination. The optimal moisture content for seeds to overcome dormancy during stratification is between 30 and 35 percent. This moisture level is achieved by soaking or rinsing the seeds for the specified period, changing the water daily if more than 24 hours. For species requiring a running water rinse or bath, this is effectively done with mesh sacks in non-recirculating water and aerated with a bubbler from a small pump. The running water rinse is an effective strategy to remove fungal inoculum from seed coats, namely *Fusarium spp*. from sugar pine seeds and *Botrytis spp*. from giant sequoia seeds.

After soaking, the seeds are drained thoroughly and allowed to surface dry on blotter paper then placed into a dry zip lock-type bag for the duration of the moist prechill period at 1 to 2°C. There should be no fruit in the refrigerator used for the prechill, since ripening fruit gives off ethylene gas that is detrimental to germination. For proper aeration, the bags should have sufficient airspace above the seeds and should be turned at least weekly for the duration. After stratification an appropriate number of seeds are counted out and placed in closed, transparent boxes on a sterile, moist substrate where the roots may grow and be examined without damage. The closed boxes help to maintain the high seed moisture content, but the substrate should not be so wet that it restricts aeration, nor be allowed to dry out and cause seed desiccation. The seed analyst, when placing seeds onto the substrate, must leave space between each to reduce the potential for transfer of seedborne pathogens.

The temperature and light conditions within the laboratory germination testing cabinet or chamber are carefully controlled. The temperature in the germination cabinet primarily affects the rate of germination and is set to alternate with lows at night (16-hr dark cycle) and highs during daylight (8-hr light cycle) hours. The alternating light effects are thought to benefit germination in conifers seeds but are strongly related to and difficult to separate from the temperature effects.

For germination capacity (GC) the germinants are counted every seventh day for the duration of the test. Any abnormalities or decayed seeds are recorded as well. If there are a number of firm, un-germinated seeds at the end of the test, it may be extended an additional week to allow for more complete germination. The delay may indicate that the seed lot requires more prechilling and it should be noted in the record. For germination vigor testing, or germination value (GV), the counts are more frequent, usually three times per week.

Note: Rapid tests i.e., tetrazolium (TZ), excised embryo, etc., are not discussed here. See selected references for details on the different types, methods, and limitations of rapid tests. (Bonner 1994) (Bonner, Karrfalt, 2008).

Variations among Species

There are variations in germination rate expectations among conifer species in California. The variations appear, in part, to be related to seed morphology. The woody seeds of pines and Douglas-fir tend to germinate more readily and at higher rates, once impediments to dormancy are overcome, than seeds with softer seed coats. However some species of pine with deeper dormancy requiring a longer stratification period are known to have lower and sometimes erratic germination, i.e., western white pine and foothill pine. The soft seed coats of true fir and incense cedar, and the resin vesicles within them, are easily damaged by heat, poor handling, or by equipment used during processing. These species tend to have more non-viable seeds included in a seed lot not only because of this delicate nature, but also because of the tendency of seed processing staff to avert undue damage from excessive processing which results in more unfilled seeds in the lot. True fir seeds also tend to have a higher rate of defects such as reversed embryos and radicle irregularities.

Coast redwood and giant sequoia naturally tend to produce a high percentage of cones with empty or tannin-filled seed, often exceeding 70 percent non-filled. The seeds are extremely small and light-weight, offering little difference in seed weight (or specific gravity) between the viable and tannin-filled seeds. This tendency poses a significant challenge for a successful separation outcome between the filled and unfilled seed fractions. Continued attempts to upgrade will at first increase filled seed percent but will eventually cause a loss of viable seed as well.

The overarching goals of the CAL FIRE forest tree seed bank in California are to promote resiliency in a changing environment and to maintain a sizeable inventory of site-adapted seed of the broadest diversity and highest quality for reforestation, emergency restoration, gene conservation and climate change mitigation. To that end, minimum germination standards have been established for general population seed lots below which the seed may be culled and scheduled for replacement (Table 5.7). Exceptions are made for improved genotypes, other special accessions, and seed from zones with infrequent production (low periodicity). High quality seeds are essential for producing vigorous seedlings for outplanting and must be readily available for nursery production when they are needed.

Note: With a changing climate and the resulting uncertainties, there may be less frequent and more sporadic cone crops and perhaps even fewer filled seed and lower quality seed. In future, this may mitigate against culling seed that has decreased to minimum germination standards until sufficient replacement seed of better quality has been obtained. However, it must be understood that if lower quality seed is retained, it will have decreased storability, decreased germination capacity and vigor, and decreased seedling production value.

Table 5.7 Minimum germination standards for the listed species

Species	**Germ % (minimum)**	**Goal % Filled [a]**
Coulter pine, Jeffrey pine, ponderosa pine, knobcone pine, bishop pine, Douglas-fir, Bigcone Douglas-fir	80-85%	95%
Sugar pine, Torrey pine, foothill pine, western white pine	75-80%	95%
Incense cedar, white fir, red fir	60-75%	80%
Coast redwood, giant sequoia	40-50%	70%

[a] *Post-upgrade seed lot goal (x-ray percent)*

The Relation of the Seed Laboratory Germination Result to the Nursery

The nursery seed user must relate the seed lab results they are provided with to seed lot performance in their operation under local conditions. Over-sowing results in waste of valuable seed, disease problems brought on by high seedling densities, and in labor intensive activities such as thinning. Under-sowing wastes time, growing components, growing space and water, and results in fewer seedlings than contracted for. Both situations are unacceptable. Standardized seed testing procedures and dependable germination results from the seed lab have led to more reliable methods of calculating sowing rates and schedules in nursery operations.

Germination capacity - the germination result most often supplied by the seed lab - doesn't always provide all of the necessary details for success in the nursery. The lab germination test is designed to be a maximum under ideal, controlled conditions and may need to be adjusted downward based on nursery conditions. Nursery sowing, where germination environments are almost always more variable and less favorable than in the laboratory, may require a longer (or shorter) prechill. In addition, there are different criteria for gauging germination between the two. In the lab, a seed is considered germinated when the radicle is four times the length of the seed and all seed components appear healthy. This stage generally occurs between day seven and day 14 in a 28-day germination test. In contrast, the nursery judges germination at a point farther along in the seedlings' development when the cotyledons have emerged from the seed coat. Abnormalities or failures occurring at this later stage often are not considered in the laboratory evaluation. Another factor to consider is seed age and length of time in long-term storage. As seeds age, they begin to show signs of deterioration. Seed vigor declines more rapidly than viability. Therefore, a vigor test (germination value) may provide valuable insight in how best to care for the seed lot in the field. Seeds of low vigor often germinate better with a shorter prechill; normal prechill of such lots may decrease germination (Bonner 1994).

Other nursery cultural factors such as priming and sanitation practices may affect survival and cull as well, and must be accounted and prepared for. The nursery seed user must establish a survival factor unique to his/her nursery operation through a series of trials, experiments, and history plots which, over time, establishes the survival statistics that become the basis for calibrating the differences between the laboratory and nursery practices.

Testing and Trials – Lessons Learned

Seed germination test and nursery operation outcomes provide numerous opportunities to conduct in-house trials that examine the efficacy of different treatments on seeds of many species. For example, past imbibition trials largely determined the amount of water uptake that is necessary for metabolic processes to begin for a variety of species without raising moisture levels too high and contributing to anaerobic conditions. The result was a significant reduction in soak times for a variety of pines including Coulter, foothill and knobcone, and was accompanied by higher germination values and minimal losses to fungal disease. In another trial that looked at improving germination of true fir through longer stratification periods resulted in refined germination outcomes for white fir based on seed condition and elevation of source. Similar improvements in germination values for white fir seed were realized by exploring different temperature regimes in the germination chamber (alternate and constant) for a variety of species - this trial resulted in the adoption of lower temperature parameters (25-15^{o}C) for *Abies* seed.

One pre-treatment method developed in British Columbia, known as "stratification-redry", was used to determine the optimal moisture conditions and pre-chill period necessary to break dormancy in California red fir seed (including *var. shastensis*). The result was a measured moisture threshold between 30 and 35 percent, post-imbibition, and an extension of stratification from six to ten weeks that consistently achieved more rapid and uniform germination while also decreasing fungal issues and early germination in stratification.

Another noteworthy study looked at sterilization treatments to reduce seed surface pathogens on a variety of species when seed germination was low. Sterilants frequently used on hard-coated seeds have included varying solutions of sodium hypochlorite (2.5% household bleach) and hydrogen peroxide (H_2O_2) given that seeds are undamaged and rinsed thoroughly after treatment. The effect of surface sterilization with bleach on select conifers was mostly mixed but found to be harmful in many instances. However, a solution of 3% H_2O_2 was found to reduce levels of fungal inoculum in ponderosa pine when seed quality is low and the procedure has been adopted when seed source is of high value or scarce. A running water rinse in a non-recirculating water bath to remove fungal spores is considered safest for soft-coated seeds and has effectively improved germination in species susceptible to damping off fungi such as *Botrytis* and *Fusarium spp*. In all, the most successful outcomes of these treatments and trials and others have led to adoption as the standard practices and they have contributed to better results in the seed lab and more successful outcomes in nursery operations.

Note: Nurseries have also found that extending stratification periods beyond those in the testing standards may result in improved germination vigor for high quality seed lots with no reduction in germinative capacity (personal communication, Jopson 2017)

Step 7 – Long-term Storage

The Objectives of Long-term Storage

The objectives of long-term seed storage are:

1. To have a viable seed supply when it is needed for regeneration and,

2. To delay deterioration, or at least decrease its rate, until the seeds are needed or can be replaced by the next good seed crop.

Tree seed storage has been an important topic of research for many years and effective storage methods have been established for certain classes of seeds. For seeds of species that can be stored, proper storage conditions are critical to maintaining seed viability over an extended period. The two most important factors affecting success in long term storage are seed moisture content and storage temperature.

Orthodox or Recalcitrant

Conifers and other woody plants are generally classified as “orthodox” or “sub-orthodox” (intermediate) in their seed storage behavior. Orthodox species are tolerant of desiccation (below 10 percent of fresh weight) and may be stored for long periods (usually 20 to 25 years or more) without loss of viability. This type of seed tends to have a hard or woody seed coat that provides good protection from damaging agents. A hard seed coat also restricts moisture uptake and gas exchange that could contribute to seed deterioration. The best seed moisture range for successful storage is between 5 and 9 percent. *Pinus*, *Picea*, *Pseudotsuga*, and *Tsuga* are among species in California described as orthodox in storage behavior.

Sub-orthodox species also tolerate desiccation and sub-freezing temperatures but storage is limited to shorter periods (usually less than 15 years). This type of seed tends to have a thin, permeable seed coat, a higher lipid content rather than starch, and resin vesicles. These characteristics make them more susceptible to damage and therefore harder to store. Seeds of *Abies, Calocedrus, Sequoia and Sequoiadendron* may be described as sub-orthodox in storage behavior though some still consider them orthodox. Incense cedar seed is particularly sensitive to damaging agents and must be handled with extra care. Successful storage of incense cedar seeds is typically less than ten years.

Seeds of woody species that are classified as “recalcitrant” in storage behavior tend to be fleshy and will not tolerate drying below rather high moisture levels without losing viability. The seed moisture content must remain at 30 percent or higher and requires storage temperatures above freezing. These conditions allow for continuation of metabolic activity but also promote rapid fungal growth. This type of seed is best collected fresh in years when it is available and sown directly. Species known to be temperate recalcitrant include *Quercus*, *Umbellularia*, *Arbutus* and *Aesculus*.

Note: Cryogenic storage in liquid nitrogen may extend storage life, but it is not a practical method for bulk seed lots or in these applications and is not detailed here.

Seed Longevity in Storage

Seed longevity is a measure of:

- seed maturity at the time of collection
- seed quality
- seed morphology
- genetics
- pre-storage handling and treatment
- proper conditions of storage

Seed maturity indices and timing of collections are very important when considering long term storage potential. Fully ripened seeds retain viability longer, are less susceptible to handling damage, and have longer storage capability. Immature seeds tend to dry out rapidly, damaging vital tissues. They are also more likely to have low vigor and be more susceptible to disease.

Seed life expectancy varies greatly among different species of conifers. These differences are, in large part, related to seed morphology. A hard seed coat protects the embryo from mechanical injury and minimizes metabolic rates in storage by excluding moisture and oxygen when the seed is properly dried. For example, there are hundreds of pine and Douglas-fir seed lots that have been stored in the CAL FIRE forest tree seed bank in Davis, CA at sub-freezing temperatures for more than 35 years with minimal reduction in viability over this period. In contrast, seeds with a soft, permeable seed coat are more likely to suffer bruising of sensitive tissues or be subject to moisture uptake, both of which contribute to seed damage and storage difficulties. In germination trials over several years, it was found that many *Abies* seed lots from a variety of seed zone and elevation sources retained high viability status for a minimum of 15 years (in-house records, LAMRC unpublished. 1998). However, *Abies* seed with lower quality status generally presented greater germination decline over a shorter period of time.

Environmental stress during cone and seed development and maturation such as drought or nutrient deficiencies in the maternal parent may limit shoot growth, thus adversely affecting seed quality and storage potential.

<u>Genetics</u> also plays a role in seed quality - some trees are reliably better cone producers than others. It is believed that individual tree fecundity is an inherited trait, likewise influenced by the maternal parent.

<u>Pre-storage handling</u> that causes damage to seeds leads to reduced seed viability in general and particularly after any length of storage. Probable handling practices to watch out for include:

- impact damage during collection

- physiological overheating of cones or seeds at high moisture content during transport or kilning
- excessive tumbling, screening or de-winging that may cause bruising or breakage
- failure of coolers during interim storage

An undetected crack or other opening in a seed coat may allow invasion of harmful pathogens and cause certain loss of viability in the short and long term.

Proper Conditions of Storage

Seed moisture content is the most significant factor in the storage environment. Conifer seeds are dried to below 10 percent moisture content (fresh weight basis) for long term storage. Optimal moisture content is between five and nine percent; moisture levels four percent or lower may lead to over-drying during storage and deterioration, especially for seeds of sub-orthodox species (Table 5.8).

Table 5.8 Moisture content thresholds & potential effects on stored seeds

Moisture %	Effects
>30%	Germination begins
18-20%	Overheating from respiration
10-18%	Seed fungi become active
>9%	Insect activity occurs
5-9%	Best range for sealed storage
<5%	Desiccation damage (possible in some species)

Source: USFS General Technical Report SO-106 September 1994

To avoid moisture uptake, proper storage in airtight and moisture-proof containers is essential. Properly sealed containers not only deter fluctuations in seed moisture content but also circumvent the need for costly humidity controls in the freezer unit. Fiberboard drums with plastic liners are effective for storage, as are heavy-weight polyethylene bags (six to eight mil is recommended) positioned inside a lidded standard size cardboard or plastic storage box. While a wall thickness of six mil or greater is an effective moisture barrier, a single bag may be pierced or torn whereas doubling two four mil bags is less likely to be compromised. This extra precaution provides better maintenance of seed moisture content over long periods and minimizes the effect of a freezer malfunction. If stored seeds are exposed to ambient conditions, either through distribution or sampling activities, they may gain or lose moisture to the atmosphere depending on the humidity level. Seeds stored for long periods should be retested for moisture content at infrequent intervals to be certain seed moisture levels are maintained in the acceptable range.

Conifer seeds retain viability for longer periods when storage temperatures are very low. Seed metabolic rates are minimized at low temperatures well below freezing; temperatures above minus 10^{o}C may allow resumption of respiration. The standard temperature recommendation for successful storage of orthodox seeds is minus 18^{o}C, assuming seed moisture content is in the acceptable range of five to nine percent.

<u>The reliability of the storage unit and the equipment used is of paramount importance</u>. The storage unit must have a reliable and dedicated power source plus safety alarms and a back-up generator in the event of malfunction or power failure. Placement of the storage unit inside a building will increase security and help to deter vandalism (fig. 5.53). Any controls on the outside of the building should be appropriately enclosed to prevent manipulation by unauthorized persons (fig. 5.55). Temperature controls and freezer management should be handled by knowledgeable and authorized personnel only (fig. 5.54).

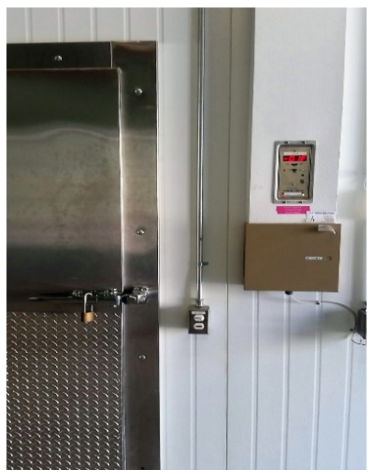




Figure 5.52 (left) Enclosed walk-in freezer at the L. A. Moran Reforestation Center, Davis. **Figure 5.53** (center) Freezer temperature control panel. **Figure 5.54** (right) Secured condenser units for walk-in freezer are protected from the elements.

The sealed storage boxes (above left) are light-weight, unbreakable, and maximize use of shelf space. Each box has a capacity of 15 to 25 pounds of seed depending on the species being stored; fiber drums can store 35 to 50 pounds per drum. Shelf systems must be sturdy and designed with seismic safety in mind.

All labeling must be clear and detailed; legible tags are located on the inside and outside of each bag with duplicate labeling affixed to the outside of the container. It is recommended that stacking of containers vertically be avoided to minimize the risk of crushing seed in the lowermost layers. Also, repeated opening and re-sealing of containers must be avoided to minimize moisture fluctuations. The storage location of an individual seed lot is recorded on its corresponding seed lot history form and in an electronic database.

Step 8 – Inventory Management

Records

It is essential to maintain meticulous records and compile a seed lot database to ensure the integrity of stored seed and associated source information. This is the data on which future collection planning and seed deployment decisions will be based. The preceding text has addressed the pertinent cone collection specifics recorded on the official Report of Collection (FM 44) and the importance of conscientious labeling throughout the collection, processing and storage phases. Critical seed quality information about individual seed lots may be assembled from the initial cone and seed assessment notes, extractory notes, preliminary and final x-rays and observations made during the germination testing process, all of which are contained in the permanent record.

A Seed Lot History (tracking) form is initiated upon delivery to the seed lab. This permanent record contains the source information from the Report of Collection and follows the seed lot through analysis, germination testing, repeat testing when appropriate, storage, inventory and distribution (fig. 5.55). If two or more seed lots are combined for any reason, all due diligence is necessary to make appropriate notations on records for each lot for tracking purposes. It is important to mix the combined seed lots thoroughly to maintain homogeneity.

SEED LOT HISTORY

SPECIES CODE	ZONE.ELEVATION	LOT NUMBER	SOURCE LAT: LONG:	CONE YEAR
PIPO	524.40	6076	T. 23N R. 4E SEC. 25, 26 CO. BUTTE	1994

NO. BU CONES	LBS. CLEAN SEED	LBS. C.S/B.C	INITIAL ASSESSMENT	PURITY%	SEED/LB.	MOISTURE %
50	52.7	1.05	Good X Fair___ Poor___ %Filled 84%	99.9%	9800	7.8%

X-RAY % FILLED 96%
GERMINATION RECORD

REBATE % ______
REBATE TO: none

STORAGE LOCATION 2 –B-3-2, ___ ___ ___ ___

DATE	PRE-TREATMENT	1st week	2nd week	3rd week	4th week	Final %	INVENTORY DISTRIBUTION	DATE	AMT USED	BALANCE
3/95	24 hr., no chill	0	3	12	146	40%	m/c, purity, s/lb., x-ray, 2 germs	1/17/95		52.76
3/95	24 hr., 6 wk. chill	10	318	63	0	97%	Magalia sow spring '95	1/17/95	4.2	48.5
8/96	24 hr., 6 wk. chill	9	326	54	0	97%	Davis sow '96	1/2/96	1.2	47.3
3/01	same	15	302	71	1	97%	Magalia sow '97	1/15/97	4.5	42.8
6/06	same	3	315	64	3	96%	Davis sow '98	12/30/97	1.2	41.6
8/11	24 hr., 6wk. chill	5	293	61	19	94%	Magalia sow '01, germ sample	1/15/01	4.0	37.6
4/14	24 hr., 6 wk. chill	5	289	62	24	95%	Davis sow '02	1/3/02	1.1	36.5
							Seed order # 02-17s	12/15/02	2.5	34.0
							Magalia sow '04	1/15/04	3.0	31.0
							Germ sample	5/3/06	0.1	30.9
							Magalia sow '08	1/12/08	2.6	28.3
							Seed order # 08-03s	11/29/08	1.4	26.9
							Germ sample	6/29/11	0.1	26.8
							Seed order # 14-32s, germ sample	2/28/14	2.1	24.7

Figure 5.55 Sample Seed Lot History form

Germination Retest Schedule

Ideally, operational seed lots are retested every three to five years to confirm germination percent or to identify potential loss in seed quality or vigor over time. Seed quality must be monitored at regular intervals because there is so much variation in quality between species and among individual seed lots and the particulars in which they were collected, processed and stored. An effective retest schedule is easily maintained by consulting the germination record in the master database. If germination loss has occurred, even under proper freezer storage conditions, the cause can generally be attributed to immature seed, poor seed quality, or deterioration due to harmful pathogens or advanced age.

When initial seed quality is high, seeds tend to store for extended periods and retain high quality status for much longer than seeds with low initial quality. This is especially true for incense cedar seeds. Incense cedar falls into the sub-orthodox category for seed storage behavior and typically declines more rapidly than hard-coated seeds. However if initial germination is high (more than 75 percent), loss is held to 10 to 20 percent or less over five to seven years and is still acceptable for the species. Alternatively, if initial germination percent is below 55 percent, then germination decline is generally much greater (20 to 40 percent) over a shorter period, usually three years or less.

Replacement Considerations

A germination retest may eventually indicate a drop in vigor whether germination capacity remains constant or has similarly declined. The speed of peak germination in a test may slow over time when compared to a previous result and is often the first expression of a decline in vigor. Vigor will decline in advance of viability. When both vigor and viability have waned and a subsequent test confirms the loss at 15 to 20 percent below the initial germination result for that seed lot, it is common practice to consider replacing the seed lot. As previously discussed, minimum germination standards vary by species and by collection region. If a seed lot declines to the minimum germination standard or lower for that species and its source is the northern or central Sierra where good cone crops are generally more frequent, the expectation is high that an opportunity to replace the failing seed lot will likely occur within the next local periodicity cycle. If the seed source is coastal, inland coastal, or in Southern California, where cone crops tend to be more erratic or lower quality, replacement may not be possible even if the germination is below standard. Without firm guidelines, the seed manager must thoroughly familiarize him/herself with the intricacies involved in sound seed replacement factors and strategies for many different species. Species such as pine and Douglas-fir may store for decades but other species including redwood, the true firs and incense cedar may require more frequent monitoring and plans to update the supply more often.

One must plan for sufficient inventory of viable seed to meet the projected need for a given planning period and for the myriad environmental challenges facing forest restoration professionals today including adapting to a changing climate, unforeseen events of uncontrolled fire, and massive die-off from elevated insect populations. The principal goals are to plan for the sometimes long intervals between good seed crops and to increase the odds for successful reforestation by obtaining the best seeds and storing them under optimal conditions intended to retain highest viability.

There is good reason to be as efficient as possible in this endeavor:

- Short supply of seeds
- Higher costs associated with collections (especially improved seed)
- Perceived increase in periodicity (number of years between bountiful cone crops)
- Increase in climate unknowns and unforeseen catastrophic events

Tree Improvement

The science and practice of forest genetics in California involves the selection of superior tree candidates, the establishment of clone banks, progeny test sites and seed production orchards, and the limited use of vegetative propagules. Tree improvement programs are designed to not only achieve sustained levels of improved volume growth and yield but also enhance the quality and broad adaptability of tree populations (Kitzmiller 1976). After a robust expansion in tree improvement activities through the 1980s, there was a rapid decline in state and federally funded tree improvement programs beginning in the early 1990s. This decline was primarily due to severe budget cuts plus the advent of shifting public views about harvest on public lands and greater emphasis on protecting old growth forests and habitat for endangered species. Opportunities for deploying improved stock were greatly reduced (Wheeler, et al. 2015). At this writing, state and federal participation still exists through tree improvement cooperatives but funding support remains low. For these agencies, the majority of forest tree seed collections are focused largely on collecting and utilizing local seed from the full range of commercially managed conifer species.

In the private sector, however, interest in faster growing, more productive forests continued and steered the desire to create forests of improved stock. This was accomplished mainly through the establishment of cooperatives which allow for sharing the considerable costs and benefits of such programs. Tree improvement associations in Northern California have active programs in the most productive sub-regions for four species: ponderosa pine and white fir in the Sierra, Douglas-fir on northern forests and sugar pine throughout the state for resistance to white pine blister rust, *Cronartium ribicola*. It is estimated that greater than sixty percent of reforestation plantings on private industrial forest lands in California are derived from such seed collections.

Genetic Quality of Seeds and Propagules

Tom Blush, USFS Geneticist

Genetic considerations should play a part in all forest regeneration efforts, whether they be natural or artificial, low or high management intensity. Stock selection for reforestation begins with a careful evaluation of the intended genetic constitution of the new forest. With the exception of vegetation propagules used for some coastal redwoods, the genetic considerations for most reforestation in California is addressed by collecting and utilizing quality seed from the region where the new seedlings will be planted. The high level of environmental site variability combined with the numerous commercial species that coexist on most sites has led to a much greater dependence on collecting seeds from local trees rather than region wide tree improvement programs. Most organizations managing forests are also involved in tree improvement programs for their key species of interest. These tree improvement programs follow a recurrent selection strategy where promising individuals are selected, bred, and tested over multiple generations (Wright 1976; Zobel and Talbert 1984; White, Adams, and Neale 2007). Offspring of the best individuals are deployed to the forest. Reforestation strategies can range from natural regeneration relying on sprouting or seed fall from residual stands to clonal propagation and deployment of elite varietals.

Natural Stands

Regeneration from natural stands and plantations represents the least-intensive management intensity of the genetic composition of the new forest. Foresters relying on regeneration from natural stands by seed collection, seed fall or by sprouting from residual trees would not rely on a tree improvement program for the species of interest. The genetic composition of the new forest would be determined by the genetic legacy available on the land. When relying on seed collection or regeneration from natural stands, the forester has considerable latitude and the obligation to control the genetic composition of the new forest to accomplish the long term reforestation goals. Traditional seed tree and shelterwood harvesting systems are intended to provide the manager with the opportunity to leave phenotypically desirable trees on the site as parents of the next generation. In California, appropriate seed collection and management depends to a large degree on collection of viable seeds by CAL FIRE and the USDA Forest Service. Restoration of disturbed ecosystems, a consequence of wildfire, insect infestation or storm damage, for instance, can also be achieved by regeneration from the residual stand. In this situation, foresters should carefully evaluate the potential of the residual forest to provide the necessary levels of genetic quality and diversity to achieve a desirable outcome. Restoration of disturbed ecosystems may require supplementing the genetic legacy of the site with reforestation stock from other sources deemed to be adapted to the site.

Seed Production Areas

Seed Production Areas (SPAs) are established by converting high-quality natural stands, plantations, or genetic evaluation tests to stands intensively managed for seed production (White, Adams, and Neale 2007). For species or seed zones without a tree improvement program, natural stands or plantations can be converted to SPAs with a well-planned and meticulous thinning operation. Prior to thinning, a careful marking operation should be done, attempting to leave only the fastest-growing, well-formed and disease-free trees. Plantations should not be converted to SPAs without verifying that the seed source of the planted trees is documented and preferably of local or near-local origin.

Converting genetic evaluation tests to SPAs has the potential to generate higher levels of genetic gain and diversity than conversion of natural stands or plantations to SPAs. Genetic evaluation plantations, such as progeny tests, are a component of a tree improvement program. The test plantation being converted to an SPA, as well as replicate test plantations, have usually been intensively measured for the desirable traits intended to be selected for in the tree improvement program. Analyses of these data are used to guide thinning and rogueing of the test plantation *to convert it to an SPA. In most cases, leave trees should have performed at least above the mean of the test population or some other quantitative benchmark.*

Open-pollinated Seed Orchards

Most tree improvement programs rely on open-pollinated seed orchards as their seed production populations (Zobel and McElwee 1964; Faulkner 1975; Simpson and Smith 1988). Seed orchards have several advantages as seed production populations. They can be located and managed specifically for seed production. Location is one of the most important factors in their success. Location should be carefully considered prior to establishing a new seed orchard. Many organizations have established seed orchards in administratively convenient locations, often near job sites on land that happens to be available, and have regretted this decision in the long run. Seed orchards should be located:

- *on land that is fertile and amenable to operability by farm machinery and aerial lifts,*
- *in climatically favorable zones for seed production, preferably isolated from wild stands or plantations of the same species,*
- *where management practices such as fertilization, irrigation, pest control, flower stimulation and crown management can be implemented to enhance seed production.*

The genetic quality of seed orchard seed is often neglected by managers in their drive to achieve high productivity. Seed orchards containing parents with high breeding values have the potential to produce high quality seed. This potential is realized when several biological assumptions essential for achieving genetic efficiency in seed orchards are met:

- *isolation from non-orchard pollen sources,*
- *balanced production of female and male flowers among the parents,*
- *flowering synchronization,*
- *random mating with equal compatibility for all crosses,*
- *minimal self-fertilization (Woessner and Franklin 1973).*

A seed orchard meeting all of these assumptions is said to be panmictic. But panmixia is rarely achieved. A manager striving to produce seed crops that reflect the potential genetic gain of the seed orchard population must identify deviations from these assumptions and correct or compensate for them (Hodge and White 1993).

Family Forestry

Erosion of genetic gain from open-pollinated seed orchards can be offset or overcome by managing the seed orchard to produce propagules with a defined family structure. Open-pollinated seed orchards are seldom panmictic, often deviating considerably from the seed orchard assumptions listed above. Although panmixia may promote a higher level of genetic diversity from the seed orchard crop, managers may decide to sacrifice some diversity for increased genetic gain by favoring families known to perform best for the traits of interest (Lindgren and Matheson 1986; Lindgren and El-Kassaby 1989; Kang, Lindgren, and Mullin 2001).

Open-pollinated families*. Parents in the seed orchard known to be especially desirable for a trait or traits of interest are identified. Seed is collected and processed from these parents. Depending on the variation among seed orchard parents and the selection intensity of the parents chosen for seed collection, considerable genetic gain can be realized in the planted forest using this simple and inexpensive management practice. Many variations of this strategy are possible. For instance, seed can be collected from a subset of top-quality "blue ribbon" seed orchard parents and mixed. The principle disadvantage of open-pollinated family deployment is that the pollen parent is uncontrolled (Adams and Tosh 1998). Many unknown pollen parents are represented in each open-pollinated family, often originating from lower quality parents located within the seed orchard or from "contaminants" from outside wild stands or plantations.*

Full-sib families*.* *Both the maternal and paternal parent is known in a full-sib family. Producing and planting full-sib families has the potential to dramatically increase realized genetic gain in the new forest (Adams and Tosh 1998). In most forest tree species that are wind pollinated, full-sib seedlots are produced by controlled pollination. Unreceptive female flowers are isolated with pollination bags and pollen is injected into the bag when the isolated flowers become receptive. The pollination bags are removed when the flowers are past receptivity and the developing seeds allowed to mature. Controlled pollination, done in the breeding population, is a laborious and expensive process because the purity of crosses generated for genetic testing is of paramount concern. In the seed orchard, large quantities of full-sib seed can be produced by controlled mass pollination. Controlled mass pollination (CMP) employs the techniques of traditional controlled pollination scaled up to a production process (Carson 1986; Bramlett et al. 1993; Bridgwater et al. 1998). Large quantities of pollen are collected, processed, and stored for immediate use or for use in subsequent pollination seasons. Inexpensive paper pollination bags are rapidly installed and pollination is accomplished using automated systems. CMP can be done cost effectively on a large scale because it is not necessary to achieve absolute purity in the full-sib seedlots destined for outplanting. Full-sib seedlots of, say, 80% purity are acceptable for outplanting. Full-sib families generated by CMP are usually deployed to the field as single-family blocks. This gives the silviculturist greater control over the stand. Family block plantings can be observed and managed as an entity throughout the rotation (Adams and Tosh 1998). Organizations committed to establishing a fine-grained pattern of genetic diversity across the landscape can mix a number of full-sib families for outplanting. An intimate mixture of individuals from comparable families can probably achieve levels of realized genetic gain on a par with single-family block plantings.*

An organization practicing family forestry must commit itself to a durable system of stand record keeping that documents the genetic composition of the stand, and to complete harvesting of single- or mixed-family blocks at the end of the rotation. Seed collection from, or regeneration cuts (i.e. seed tree and shelterwood systems) applied to family blocks should not be attempted. The seed parents resulting from this would be related, and the subsequent stand established from this residual population would suffer from the deleterious effects resulting from inbreeding depression in the progeny that establish the new forest.

Varietal Forestry

Cloning of highly desirable individual genotypes is the ultimate means of capturing all of the gains generated in a tree improvement program (Ahuja and Libby 1993a, 1993b). The principal weakness of relying on seed as the delivery vehicle for genetic gain is the genetic recombination process that occurs after pollination and fertilization. Recombination tends to break up or disassociate combinations of genes that work together in an additive or synergistic fashion to produce a desirable trait or combination of traits. Cloning, theoretically, offers the forester the ability to capture that one-in-a-million individual identified during the testing phase of the breeding program and bulk it up ad infinitum. In most forest tree species, however, cloning is technically very challenging. Deployment of varietals across the landscape also presents many challenging social and biological considerations related to genetic diversity that the responsible forest manager must address. In forest trees, varietals are typically generated by vegetative propagation via rooting of cuttings, and the tissue culture techniques of organogenesis and somatic embryogenesis.

Rooted cuttings*.* *Cloning by rooting of cuttings is a well-established process that has been practiced by horticulturists for centuries. For some species, such as those in the genus Populus, it is relatively easy to accomplish. In many conifer species, however, rooting of cuttings is technically demanding, inefficient, and costly. Few forest tree species lend themselves well to mass production of varietals from rooted cuttings. An important consideration in vegetative propagation in general and rooted cuttings in particular is maintaining juvenility in the stock plants. Rootability falls off rapidly as the stock plant matures. Rooted cuttings derived from more-mature stock plants may also grow slower and exhibit form traits more typical of mature plants. The most common means of maintaining juvenility in the stock plants is to cut them back or hedge them yearly and serially repropagate the stock plants as they become decadent. Maintaining juvenility, or at least slowing maturation is essential.*

Restoring mature individuals to a juvenile state does not appear to be feasible with current technology. Organizations contemplating mass production of varietals by rooted cuttings should employ an experienced horticulturist and must invest in the greenhouse and nursery technology required for success.

***Organogenesis**. Organogenesis is the tissue culture process of differentiating plant organs such as stems, leaves and roots from undifferentiated cells and tissues (Ahuja and Libby 1993a, 1993b). This tissue culture technique, applied to forest trees, is technically very demanding. It requires laboratory personnel and facilities to execute the tissue culture phases of the process. Organogenesis has been accomplished in numerous tree species, but only a few organizations have devoted the resources necessary for establishing a mass-production operation. The process requires, first, inducing shoot production and proliferation from callus tissue. These shoots are then harvested and induced to form roots. Timing and application of specific media formulations in a highly controlled, sterile environment are critical to success. Somaclonal variation, genetic changes induced by the tissue culture process, are a problem with organogenesis and must be carefully screened for. Mass production of varietals via organogenesis has limited potential in forest trees. It may be most useful in some hardwood species and as an avenue to facilitate genetic transformation approaches used by molecular geneticists.*

***Somatic embryogenesis**. Somatic embryogenesis (SE) is a promising tissue culture technology for cloning conifers by which somatic cells, usually immature just-fertilized embryonic tissue, is induced to proliferate and then to differentiate somatic embryos in a tissue culture system (Ahuja and Libby 1993a, 1993b). Like organogenesis, it is technically demanding and requires considerable expertise and facilities to accomplish. Unlike organogenesis, SE produces intact embryonic plantlets ready to germinate and grow. This makes it amenable to artificial seed technology. The starting material for SE is usually generated by controlled pollination, generating elite full-sib crosses. SE has, so far, proven to be highly genotype-specific in that the efficiency of the process varies widely depending on the female and male parents comprising the cross, the individuals within a cross and, often, the direction (female ↔ male) that the cross was made. Because SE is a tissue culture process, it also has the potential to integrate with genetic transformation technologies.*

Varietals can be mass produced by combining rooted cutting and tissue culture approaches. Tissue culture techniques can be used to establish lines, or clones, and the moderate number of propagules generated from the tissue culture process are then used to establish cutting orchards from which rooted cuttings can be mass produced. This capitalizes on the ability to cryopreserve tissue culture lines, thus indefinitely preserving and halting maturation in the donor line. And it takes advantage of rooted cutting technology to bulk up and mass produce cuttings in a less technically demanding greenhouse or nursery environment.

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Glossary of Cone & Seed Handling Terms

- adaptation – characteristics that have evolved by natural selection over time that enable an organism to be successful (survive and reproduce) in a given environment
- bract - a modified leaf which extends underneath a seed bearing scale (DF, Shasta RF)
- case-hardened - inability of cone scales to flex caused by too rapid drying, insects or disease
- corrosion cavity – the cavity in the central portion of the megagametophyte that forms through cell breakdown. The embryo will grow into this cavity
- cotyledon - primary leaf of the embryo
- damping-off – the killing of a seedling by micro-organisms before emergence from the soil or the collapse of the hypocotyl and/or radicle immediately after emergence
- dead-filled seed – the complete deterioration of all seed contents
- dormancy - a physiological state in which a seed is capable of germination but does not
- embryo dormancy - conditions within the embryo

- embryo - rudimentary plant within a seed
- empty seed - a seed that is hollow, corky, pitchy or filled with larvae
- endosperm - female gametophyte. Correctly termed Megagametophyte in gymnosperms
- fertilization - fusion of the sperm nucleus w/ egg cell nucleus and doubling of chromosomes (for 1-yr species - this occurs in early summer; for 2-yr species - this occurs in spring of the 2nd year)
- filled seed - a seed containing all tissues essential for germination and considered potentially viable
- frass – insect excrement found on or within cones; usually indicative of feeding activity/damage
- genetic "generalist" - species that show low genetic differentiation across a wide range of environmental gradients
- genetic "specialist" – species that exhibit strong genetic differentiation over small geographic and climate scales
- hypocotyl - embryonic stem below the cotyledons and directly above the radicle (primary root)
- imbibed – seeds that have become swollen and physiologically active due to uptake of water – this moisture must reach the embryo
- integument – the coat of an ovule which develops into the seed coat
- megagametophyte – storage tissue that provides nutrients to the developing embryo; also sometimes referred to as endosperm
- microclimate – The climate of small areas especially in regard to significant differences from the general climate of the region
- moisture content – for seed, is based on the proportion of moisture relative to the fresh weight of the seed and is usually presented as a percentage. M/C = fresh wt.- oven-dry wt. / fresh wt. x 100
- morphology – study of form and structure of an organism, especially their external form (size, color)
- orthodox – seeds that can be dried to low moisture content and stored for extended periods without losing viability

- peduncle – the stalk or stem of a cone
- phenotype - visual appearance of having desirable characteristics (genotype + environment)
- phenotypic plasticity – the ability of a genotype or a population to maintain high fitness across a range of environments by altering its phenotype
- periodicity - number of years between bountiful crops
- pollen cone - male reproductive structure - produces pollen grains
- pollination - transfer of pollen from male cone to female cone (in conifers-wind pollinated)
- poly-embryony – the formation of more than one embryo in a seed
- pre-conditioning - facilitating after-ripening in preparation for kilning +/or tumbling (racking & turning sacks)
- primordia - rudimentary structure at the earliest stages of development
- progeny - offspring of plants
- purity – seed lot characteristic describing the weight of pure seed in relation to the weight of seed + debris. A purity standard above 98% is necessary for nursery calculations
- radicle - primary root of the embryo
- recalcitrant – seeds that are resistant to drying and storing (must be fresh collected when available)
- scarification – degradation of the seed coat by mechanical abrasion, chemical or hot water treatment to increase water uptake and gas exchange
- seed - a matured ovule containing an embryo and megagametophyte, enclosed by a protective seed coat which is capable of developing into a plant under favorable conditions
- seed coat dormancy - impermeable to gas and moisture exchange
- seed cone - female reproductive structure - usually two seeds borne on each scale spirally arranged around a central axis
- seed lot - a quantity of cones or seed having uniformity of species, source, quality and year of collection
- seed source - location where the seeds were collected; seed zone and elevation (T, R, Sec and/or Lat/Long)
- seed zone - in Calif. arbitrary area designated on the basis of biogeographic and climate regions & latitude

- self-pollination – the transfer of pollen from the stamen to the stigma on the same plant (tree); aka "selfing"
- serotiny – pertaining to cones that remain closed on the tree for several months or years after maturity making them late in dispersing seeds
- stratification - pre-germination treatment of seeds to overcome dormancy and to promote rapid and uniform germination - synonymous with moist chilling
- superior tree – a phenotypically outstanding tree that has no visible undesirable characteristics and produces more cu ft. volume/yr. when compared to immediate even-aged neighbors. JK
- tolerance – the permitted deviation from a standard beyond which a germination test must be repeated
- viable - capable of germinating - viable seeds are filled but not all filled seeds are viable
- vigor – robustness of a seed lot; declines with age