Chapter 2

Genetic Improvement of Forest Trees

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Introduction

This chapter is designed to provide an introduction to the fundamental concepts of forest genetics. It can be used to gain a basic understanding of why certain procedures are used to improve forest trees or it can be used as a source of more detailed information. A number of references are listed-both for historical background and for technical details.

Examples are frequently cited from operational tree improvement programs to focus the chapter on an applied level. Many of the examples used are taken from the southern US since the author has had over 35 years of experience working in that region.

Terminology

Many of the terms used in this chapter are listed in the Glossary ( ). A more detailed glossary may be found in Snyder 1972 and Wright 1976. Comprehensive references on Forest Genetics include Dorman (1976), Wright (1976) and Zobel and Talbert (1984). Key words or phrases are underlined the first time they are used in the text.Definitions of these key words and phrases are found in the glossary.

Forest Genetics is the general term often used for the study of inheritance in forest trees. Forest Tree Improvement usually refers to the applied use of Forest Genetics concepts to actually improve the quality of the trees. Tree breeding is often used as a synonym for tree improvement, but it also may be found referring to specific activities such as controlled pollination. Zobel and Talbert (1984) describe Forest Tree Breeding as:"activities geared to solve some specific problem or to produce a specifically desired product". Tree Improvement will be the term used most frequently in this chapter.

It is important to understand that tree improvement is an integral part of silviculture. Tree improvement provides the raw material for artificial regeneration which is one of the most important tools in the arsenal of the silviculturist. Tree improvement provides a direct avenue to inject genetically improved seedlings (or cuttings) into the reforestation system with no additional "handling fees". It costs no more to plant a genetically, improved seedling than a "woods-run" seedling. (N.B. Although the costs of producing genetically improved planting stock are not insignificant, they can be viewed as an investment in future increased productivity.) Dividends accrue in terms of increased growth, better form and wood quality, and improved insect and disease resistance.

Allocation of Resources

One of the key elements of land management is allocation of resources. An ever-expanding world population demands an ever-increasing supply of wood products. These must be produced on both private and public lands. The most productive sites should be devoted to maximum timber production. Maximum wood production on these acres relieves the pressure on other acres which can be devoted to native vegetation, wildlife production, aesthetic considerations, and other uses not compatible with maximum timber production.

Even those acres devoted to maximum wood production via artificial regeneration with genetically improved planting stock are not lost to most aspects of good forest management practices. These acres will support strong wildlife populations, preserve watersheds, and provide many recreational opportunities. All these are fully compatible with timber production.

Monoculture

Critics of plantation forestry programs often cite the dangers of monoculture as reasons to reject these programs. The reasons quoted range from disease outbreaks to site deterioration, but usually focus on lack of biodiversity. In point of fact, there are few documented cases of severe problems, even in clonal plantations. Where there have been losses from pathogens, the increased productivity of the plantations usually greatly overbalance the losses.

There can be interactions between intensive culture and diseases, as in the case of fusiform rust, which may be increased by site preparation procedures and fertilization (Miller 1972). Some clonal plantations such as cottonwood and eucalyptus have encountered disease problems but these are often due to off-site planting rather than the lack of genetic diversity. Even the fabled case of "Saxony spruce", where pure stands of Norway spruce were blamed for "site deterioration", were actually cases of off-site planting. This, in conjunction with poor management and poor seed source selection, led to drastically decreased productivity of the plantations (Lutz and Chandler 1946).

Gene Conservation

The fundamental concepts of gene conservation are an integral part of the tree improvement process. In the preservation of selected trees in seed orchards, clonal banks and progeny tests, valuable germplasm is not only preserved but also replicated on different sites where it may enrich local tree populations via wind pollination. The planting of genetically improved seedlings on new sites likewise enriches the local gene pool of that species. Future populations of these trees can be expected to be more heterogeneous than the local stands as well as more productive (Namkoong 199x).

During the selection and breeding of forest trees a tremendous volume of data is generated regarding species-site interactions, growth and tree quality information, and physiological relationships. These data serve to increase our understanding of the importance of high quality seedlings which are well-adapted to the planting site. Good forest stewardship requires vigorous, fast-growing trees as well as a diversity of flora and fauna.

The use of isozyme analysis has been adopted by many forest geneticists as a tool for estimating diversity in natural populations. Shimizu and Adams (1993) for example, using isozyme analysis in natural stands of Douglas-fir, found no evidence that planting of nursery-grown seedlings contributed any less genetic diversity than natural regeneration.

Tree Improvement vs Crop Improvement

The genetic improvement of forest trees has many similarities to the breeding of field crops. Most of the concepts are the same, namely the selection of above-average individuals from large populations, and subsequently breeding these individuals using a specified mating design. Following the breeding phase, the progeny must be tested on a variety of sites and climatic conditions. Progeny tests are specially designed genetic tests that expose hereditary differences among trees, by bringing different genotypes together under a common set of environmental conditions.

When the progeny have developed sufficiently for a reliable assessment of their value, improved individuals or groups can be released for operational use and/or the breeding cycle can be repeated.

There are two major differences between working with field crops and forest trees. The first is time. Field crops such as corn and wheat reach reproductive maturity in a few months-most trees require many years. Crop rotations with corn and wheat are also only a matter of a few months while trees may not produce a marketable crop for 25 to 100 years! Even in the tropics, it is rare to harvest a timber crop in less than 8 or 10 years. In practical terms this means that a corn or wheat breeder can complete a breeding cycle in 2 or 3 years compared to the tree breeder's 8 to 10 years, at the very least.

Table 1—The time factor:field crops vs trees.

	Field crops	Trees
Reproductive maturity	1-2 months	5-20 years
Rotation length	4-6 months	10-100 years
Breeding cycle	1-2 years	8-20 years

The second major difference is that most field crop breeding is done with domesticated varieties which have been manipulated by man for centuries and are often genetically homogeneous. Forest tree breeding, in contrast, usually starts with natural, wild stands of trees which have been little changed by man. An exception here is "highgrading," the common logging practice of cutting the best quality trees and leaving the worst to regenerate the next generation. Unfortunately, tree improvement foresters are often forced to work with the results of one or more cycles of highgrading, namely trees of poor form and marginal value for breeding material. On the other hand, working with wild, unselected stands of trees does provide an opportunity to produce large gains in quality in the first few generations of breeding.

Field crop breeding usually involves working with well-known varieties which are often pure lines (genetically pure). With corn, for example, pure lines are crossed to produce heterozygous (genetically different) progeny which exhibit hybrid vigor (improved performance due to the interaction of different genotypes). Site considerations are important here, as the corn will be planted on uniform, well prepared sites while the trees may be planted on rough,cut-over sites with little or no site preparation. Adaptation is also a consideration as the corn is bred for a very narrow group of soils, sites and climatic zones. The trees, on the other hand, may be planted over a much wider range of soils, sites and climatic zones.

The Biology of the Species

The genetic improvement of any crop will be effective only after a careful analysis of the biology of the species and how this influences the breeder's approach to the problem. For example, insect pollinated species require special considerations from tree breeders. Genera such as *Acer*, *Liriodendron*, *Magnolia*, *Salix*, *Tilia*, *Ulmus*, and many tropical species are all insect pollinated and therefore cannot be managed with the same techniques as wind-pollinated species. The majority of our commercial timber species are both wind pollinated and Monoecious (Latin: one house) producing both male and female flowers or stroboli on the same tree. he location of these flowers usually favors cross pollination:for example, most conifers bear female flowers in the upper areas of the crown with the males below, usually favoring cross- rather than self-pollination.

Cross pollination, in most plants, is an adaptation designed to increase heterozygosity (a mixture of genetic material) which is usually linked to vigorous growth, high fertility and strong resistance to attack by pathogens. Conversely, self pollination often leads to poor growth, weakness and reduced fertility. Most production seed orchards are designed to favor cross pollination for these reasons.

Some tree species are Dioecious (Latin: 2 houses) with the sexes separated on different trees. Examples are: *Fraxinus*, *Ilex*, *Juniperus*, *Populus*, *Salix*, and *Taxus*. Fortunately, many of these genera can be propagated vegetatively. Also, in the case of the poplars, cross pollination can be accomplished very quickly on a greenhouse bench by simply brushing pollen on the receptive female flowers

Precocious (early flowering) species are adaptable to seedling seed orchards since the flowers are produced at a young age and seed production is abundant. Examples are *Pinus virginiana*, *P. clausa*, *P. contorta*, and *Alnus* (E. black). Species which can be vegetatively propagated present unique opportunities since sexual reproduction is not necessary and therefore the recombination of parental characteristics can be avoided. Species such as Populus deltoides can be produced vegetatively with unrooted stem cuttings planted directly in the field. Other species require rooting under special conditions before they can tolerate field planting. Examples of these are Monterey pine, Norway spruce, Cryptomeria japonica, Alaska yellow cedar, sweet gum and sycamore.

Concepts of Genetic Improvement

Phenotype - Genotype:

When we look at a Sitka spruce, a cherrybark oak, a Rocky Mountain juniper, or even an Angus bull we see a phenotype. This is the living organism with its own unique genetic constitution, as modified by its environment. In contrast, the genotype of the organism is encoded in its DNA. Each tree, therefore, has its own individual set of genetic blueprints. These are the instructions that determine the genetic potential of its progeny.

The formula: phenotype is the product of the genotype as affected by its environment is often written: $P = G + E$. The phenotype is the organism which we see, measure, and with which we work. Life would be much simpler if the genotype was as obvious! Geneticists spend a great deal of their time and energy working to ascertain the actual genotype of their target organism. A major reason for progeny testing is to gain a better understanding of the genotypes of the selections which we are breeding. Recent advances in gene mapping with loblolly pine (Sewell and Neale 1995) indicate that real progress is being made with the description of the loblolly genome. Some day we will understand the genomes of Douglas-fir, ponderosa pine and loblolly pine as well as we understand E. coli and the common fruit fly.

The Genetic Code

The physical basis of genetic information is the DNA molecule, a long, helical, molecule with two strands connected by base pairs. The molecule is sufficiently stable to provide for the continuity of the species, yet flexible enough to allow for periodic changes. DNA therefore serves as both the blueprint for cell structure and metabolism and also the template for the replication of many exact duplicates. These unique properties enable evolution to proceed in a remarkably stable universe. The evolutionary forces of mutation, migration, hybridization, and natural selection are responsible for the great variety of life that exists today.

Many of our tree species which coexist on the same sites maintain their status as separate species primarily by a separation in flowering time. On transitional sites when one species is accelerated or retarded in flowering time hybrids often result as in the case of the coulter x jeffrey hybrids in California (Zobel 1951) and the pond x loblolly pine mixtures in North Carolina (Saylor and Kang 1973).

New genotypes which result from mutations may move about (migration) and interbreed with other genotypes (hybridization). The new gene combinations which result are then sorted out by the process of natural selection. If these new genotypes are able to survive, reproduce and leave more progeny than their competitors, they are well-adapted. Therefore the tree species, races an stands with which we are working are well-adapted to a specific site b virtue of their survival and reproduction in that environment.

Chromosome numbers:

Chromosome numbers can change as a result of mutations. Polyploidy has been an important evolutionary factor in the plant kingdom. In most of the commercially important conifers chromosome numbers range from n=11 to 13 (Saylor 1972). A notable exception is redwood, which is hexaploid (6n=66).

In contrast, chromosome numbers in the commercially important broadleaved trees vary widely, from n = 7 to 19, with a number of polyploids, including the alders, birches, several *Prunus* species, and magnolias. A comprehensive table of chromosome numbers is found in Wright (1976).

Selection

Almost every process of genetic improvement starts with selection. This is true regardless if we are working with dairy cattle, winter wheat or forest trees. The concept of selection involves the selection of a very small proportion of a population for one or more desirable characteristics. The difference between the proportion selected and the population mean (average) is called the selection differential. Graphically this can be depicted as in Figure 1.

Figure 1: The selection differential.

Genetic gain or progress is measured by the product of the selection differential and the heritability (degree of genetic control) of the trait in question (e.g.height,straightness, volume). Therefore by selecting individuals which are well above average in height,for example, and assuming that the heritability (h2) of height growth is sufficiently high to show progress, some gain in height should be expressed in the next generation. On the other hand, if the population in question is extremely uniform in height, and/or the heritability of height growth is low, selection may not be an effective approach. In some species (e.g. *Pinus resinosa*) the population is so uniform that selection for many traits is not cost-effective (Fowler and Morris 1977).

Hybridization

When populations are uniform and selection is not likely to be effective one possible technique of genetic improvement is hybridization (crossing individuals within a species or genus). Most of the successful hybrids in forestry have been interspecific (between species) hybrids. Examples are
hybrid larch (*Larix leptolepis* x *decidua*), hybrid poplars (*Populus* spp. widely hybridized with many cultivars), the *Pinus rigida* x *taeda* cross in Korea (Hyun (1976) and the eucalyptus hybrids (Campinos 1980).

Heterosis (hybrid vigor) is a controversial topic among tree breeders. Many interspecific hybrids grow better than their parental species when planted in transitional environments. The actual documentation of heterosis is seldom published however.

A great deal of effort has been expended with the goal of producing a hybrid chestnut which would have resistance to the chestnut blight (Parasitica endothica). Unfortunately, the American chestnut (*Castanea dentata*, which was devastated by the introduced disease at the turn of the century, has little resistance to the disease. It is possible to cross American chestnut with Chinese chestnut which is resistant to the blight. The hybrids produced are resistant to the blight, but unfortunately their form is so poor that they have little value as timber trees. Genetic engineering offers new hope for the American chestnut (see Genetic Engineering).

Natural hybridization occurs frequently on transitional sites or ecotones. Examples are the *Pinus coulteri* x *jeffreyi* hybrid in California (Zobel 19 ,and *Pinus sondereggeri*, the longleaf x loblolly cross (Namkoong (1963) in the South. Hybridization often occurs near the edge of the range where the species is losing its adaptive advantage. In eastern North

Carolina, for example, here is a transition zone where loblolly and pond pine (*P. serotina*) often hybridize (Saylor and Kang 1973). Likewise in southeastern Oklahoma and northeastern Texas shortleaf and loblolly occupy many sites together and hybrids are not uncommon (Abbott 1974).

Natural hybridization is a common phenomenon among the oaks (Muller 1952) some birches (Barnes and others 1974) and aspens (Pauley 1956).

Testing for Breeding Value

After the elite/select/superior individuals have been selected, some system of testing their genetic value must be used. We have identified these trees as good phenotypes but we do not know their genotypes and therefore we are uncertain as to their value as breeding stock. Sometimes the outstanding trees in a stand may be taller than their neighbors due to an environmental advantage such as better soil or more moisture. It is important to use only trees with better than average genetic characteristics, as the environmental differences will not be passed on to future generations. In natural stands it is important to determine the age of individual trees. Trees growing together may have a similar size, yet be quite different in age. Obviously we would prefer that our select trees not be outstanding merely based on the fact that they have been growing longer than their neighbors.

Trees which can be vegetatively propagated are usually tested by planting in blocks and comparing performance with a standard population. This may be a clone of known performance, or in some cases seedlings from a standard seed lot may be used. Tests of this type which are designed to evaluate the relative performance of a specific clone are called clonal tests .

Trees propagated from seed are usually progeny tested using one or more test designs modified from crop breeding. Early work with trees involved the use of open-pollinated tests where cones/seeds were collected from select trees and the half-sib progeny (female parent known-males unknown) were evaluated in plantations. As technology evolved, control-pollinated tests were developed which provided much better estimates of breeding values.

Most progeny tests are designed with row plots in field plantations although single-tree plots have some advantages over row plots. A relatively new technique has been developed by the Western Gulf Forest Tree Improvement Cooperative utilizing greenhouse testing (Lowe and van Buijtenen 1989). With this technique, culling of the poorest 17 to 20% of the progeny can be done at about 5 months, based on shoot dry weight..

Screening for Fusiform Rust Resistance:

Due to the economic importance of fusiform rust on the southern pines the USDA Forest Service established a rust testing center at Bent Creek, North Carolina in 1976. Forest Service pathologists developed a standardized innoculation system which could be used to screen loblolly and slash pine seedlings for susceptibility to fusiform rust (Knighten 1988). The Resistance Screening Center inoculates an average of 40,000 seedlings annually. The three southern tree improvement cooperatives routinely screen all new selections by sending seeds to the Rust Testing Center for evaluation. This is an essential part of the progeny testing procedure.

Screening for white pine blister rust resistance:

Cooperative programs designed to develop resistance to white pine blister rust have been operating for a number of years in California and Idaho. The USFS Region 5 (Pacific Southwest) program has identified 985 rust resistant sugar pines for future tree improvement use. Family selection has been used as a breeding strategy.

Advanced generation breeding:

Advanced generation breeding is usually designed with a combination of selections from progeny test plantations in conjunction with new selections from operational plantations or other sources. A major advantage of selection in plantations is that the environment is usually more uniform than in natural stands. Tree age, spacing and soils are often relatively uniform with the result that the genotype more closely approaches the phenotype. In this case selection is more efficient and gain can be increased. In most advanced generation breeding plans the best individuals are selected from the best families. It is important however to separate the production population from the breeding population to minimize the effects of inbreeding (Lowe and van Buijtenen 1986).

Starting a Tree Improvement Program

Establishing objectives:

Prior to starting a tree improvement program a comprehensive analysis of the situation should be made. Tree improvement is long-term work. A great deal of time and energy can be saved with some careful planning. The following questions should be considered:

The products to be produced.
The wood properties desired to produce these products.
The volume of wood required.
Possible species to be used- native or exotic? (What are the long-term consequences of using exotic species?)
Rotation length? (Reducing the rotation length results in major improvements in gain.)
Reforestation system to be employed: seed propagation or vegetative? bareroot or container nursery? storage and distribution? planting techniques? cultural procedures to be used?
Plantation survival system to be used?
Personnel required + level of knowledge.
Facilities and equipment needed?

Identifying the Raw Material to be Used

Native versus Exotic Species:

Are there native species available that are well-adapted to the planting sites to be used or would exotic species be more productive? The temptation to introduce an exotic species may be strong but there are a number of advantages of native species:

They have evolved in harmony with their environment and usually have developed a mutual tolerance with competitors and pathogens. Exotics, on the other hand, may not perform well in a new environment. They have not been exposed to the stresses of this new environment and they have often not had sufficient time to adapt to local conditions.

Native species have well-defined management regimes which have been tested over time. Reforestation personnel have learned how to grow, ship, store and plant the seedlings/cuttings.

An exotic species introduced into a new environment does not necessarily produce wood with the same characterisitics as in its place of origin. Excessive amounts of juvenile wood are common, as are wide bands of earlywood and narrow bands of latewood. These growth patterns lead to low-density wood and drying defects (Zobel 1981). There are notable exceptions (e.g. *Pinus radiata* in New Zealand), but in general the wood quality of native species is more desirable than that of exotics.

Public opinion is running strongly in favor of native species both in the US and overseas. Plantation forestry with exotic species has encountered strong public resistance in a number of locations.

Successful introductions of exotics

There have been a number of successful introductions of exotic species world- wide. A few examples of native US species introduced into other countries are Monterey pine (*Pinus radiata*), a minor species in coastal California has become the backbone of the forest products industry in New Zealand, a country with few conifers of economic importance. Monterey pine has also done well in Australia Chile, and South Africa. Douglas-fir (*Pseudotsuga menziesii*) and Sitka spruce (*Picea sitchensis*), native to the Pacific Northwest, have been widely planted in Great Britain and nothern Europe. Several of the southern pines have been widely planted in Australia, South America, and South Africa. See table___.

Table : Successful introductions of exotic species.

Land races.

When the decision has been made to utilize a given exotic species the question arises as to the source of material to be used. Often it is more efficient to select within a land race of the species rather than the original population in its native environment. A land race has become adapted to its new environment by virtue of its survival there for a number of years. For example, Monterey pine has been growing in New Zealand for over 100 years. During that period it has weathered many storms and fought off many pathogens. This process of natural selection has enriched the population by the gradual elimination of individuals not well adapted to their new environment. Selection of individual trees within this land race will be more efficient therefore than returning to the native populations in California.

Geographic Variation

One of the first definitive studies of geographic variation was established by Philip Wakeley of the USDA Forest Service in 1926-7. Wakeley collected loblolly pine seed from Arkansas, Georgia, Texas and locally (Louisiana), grew the seedlings and planted them on a site near Bogalusa, Louisiana.

Table 2: Bogalusa seed source (Wakeley 1944)

This study was the first solid evidence that the source of seed was important in the growth and rust resistance of loblolly pine. The local source produced almost twice the volume of the other sources after 22 years. In addition, this was the first evidence that loblolly pine from Livingston Parish, Louisiana, had special merit as a rust-resistant source.

The Southwide Pine Seed Source Study was designed by Wakeley as a cooperative project involving 17 different agencies, with field plots established from Texas to the Atlantic Coast. This study demonstrated that loblolly sources from west of the Mississippi River usually had better planting survival and greater rust resistance than eastern sources. On the other hand, many of the south Atlantic coastal sources had faster growth rates than the western sources (Wells 1969, Wells and Wakeley 1966, Wells 1983).

The results of this study have led to widespread planting of Livingston Parish loblolly throughout the southeastern coastal plain leading to major reductions in fusiform rust infection (Wells 1985). Likewise, forest industry has planted Atlantic coastal sources of loblolly in Arkansas and Oklahoma with impressive gains in volume growth on the better sites (Lambeth et al 1984).

(Figure   Livingston Parish Loblolly + Atlantic Coastal)

On the West Coast, the Eddy Tree Breeding Station was established in California in 1925. This later became the Western Institute of Forest Genetics which played a major role in the developement of Forest Genetics in the West. The 2 varieties of Douglas-fir (Coastal-Interior) have been studied extensively (Kung and Wright 1972). The coastal variety has been widely planted in Great Britain and northern Europe. Other western species with pronounced racial differentiation are ponderosa pine, and grand and white fir. In the northern United States, white spruce occupies an extensive east-west range with considerable racial variation (Nienstaedt 1968).

Utilizing the Raw Material

Seed Production Areas

Time is a critical factor in determining the route to follow in a tree improvement program. A useful expedient is the seed production area (seed stand). This is a high-quality stand which can be thinned to remove the lower quality individuals and then managed for seed production (Cole 1963; Rudolf 1959). Although the gain from these areas is not high, (Easley 1963) the time saved can be more important than the degree of improvement. These stands can be burned, fertilized and sprayed for insect control. Seed collection can be done by climbing, shaking, tarping or felling trees. An efficient system can be designed where the felling of trees is planned to coincide with good seed crops.

Although natural stands are preferred sources for seed production areas, plantations are often used where the seed source can be verified. In these cases the plantation is treated like a land race - good performance over a given time is evidence that this plantation has adaptive value on this site.

Figure______ Loblolly pine seed production area
Virginia Department of Forestry

Clonal Seed Orchards

Clonal seed orchards have been established for many outcrossing species. The procedures used involve selection of individual trees, progeny testing to determine their breeding value, and replication of the ortets in an orchard environment. In actual practice the orchard is usually established by grafting and the progeny testing is done by controlled pollinations within the orchard or clonal banks.

Seed production usually begins prior to the completion of progeny testing resulting in the production of improved seeds which cannot be certified (Seed Certification is covered in Chapter x) until progeny testing is completed and the orchard can be rogued (removal of trees with low breeding value).

(Figure Clonal seed orchard.)

Selection of plus trees

Selection of the plus trees from wild stands that are pure and even-aged, usually involves grading of the candidate tree in comparison with the best adjacent crop trees (of similar age) in the stand. Characteristics compared are straightness, height, DBH, volume, form class, crown size, branch diameter, branch angle, natural pruning and wood quality. Any evidence of insect or disease susceptibility usually calls for rejection of the candidate. Acceptance of the candidate tree depends on the numerical rating of the tree, its wood quality, age class, geographic location and any special attributes.

Selection of orchard sites

All seed orchards require good access, level topography, and well-drained soils Since vehicle traffic is essential in the managemenmt and harvesting of orchards, a coarse-textured soil is mandatory. Subsoiling is practiced in many seed orchards to fracture hard pans formed from compaction by vehicles. Even in sandy soils compaction can seriously reduce root growth of the trees. Establishment of a year-round ground cover is important to stabilize the soil and prevent/reduce erosion.

Establishment

Most clonal orchards are established by grafting although at least one slash pine orchard has been planted with cuttings (Bengston 1969). Rootstock can be planted in the field and grafted in-place (field grafting) or grafts can be done on potted stock grown in a greenhouse, lath-house, or in the field (pot grafting). Grafting on potted stock is more cost-effective but field grafting is preferred by some orchard managers due to often a shorter time to reach commercial cone production.

Graft incompatibility occurs in many species including most conifers. This is a problem in roughly 22% of southern pine clones (Lantz 1973) and it is particularly serious in Douglas-fir, where up to 67 % of the clones may be affected (Wheat 1967). There is some evidence that clonal root stocks from related material may reduce incompatibility rates but the data are not conclusive. Copes (1967) has developed a tissue sampling technique that can be used to predict incompatibility in Douglas-fir.

Management:

Most seed orchards are fertilized to promote flowering and some are irrigated to reduce the impact of moisture stress. A number of references on seed orchard management are listed at the end of this chapter. Insect control is essential for maximum seed production. In the absence of cone and seed insect control Belcher and DeBarr (1975) have estimated that 11% of the loblolly cones were attacked by cone worms (Dioryctria spp.). In this survey of 26 seed orchards surveyed over 3 years, an average of 9.9% of the collected seeds were damaged by insects. There was a large amount of clonal variation as the range of cone worm attack was from 0 to 67% depending on clonal susceptibility.

More recently, Jett and Hatcher (1987) reported that cone worms can cause losses exceeding 90% of loblolly cones when pesticides are not used.

Pollen Contamination

Most seed orchards are located with some consideration for pollen contamination. Unfortunately economics often dictates locations which cause a major problem with pollen contamination. Early seed orchard establishment in the South carried a recommendation of at least 400 foot isolation zones surrounding the orchard (Squillace 1967). Later studies indicated that vegetation formed a more effective barrier than either soil or sod.

Pollen contamination in seed orchards was estimated by Adams and Birkes (1989). using isozyme analysis. In seed orchards of Douglas-fir, loblolly pine, and Scots pine they estimated that as much as 50% of the pollination within the

orchards was due to contamination from outside pollen. The amount of self-fertilization within the orchards was estimated as less than 10%, with
considerable variation by clone.

An additional study by Smith and Adams (1983) also indicated that pollen contamination was in the 40-52% range for 2 Douglas-fir seed orchards in Oregon. Suggestions for reducing contamination included more complete geograhic isolation, water spraying to retard flower development, and supplemental mass pollination.

Supplemental mass pollination

Supplemental mass pollination(SMP) has been used effectively in southern pines, Douglas-fir, and Scots pine (Bridgwater et al 1993). The authors have summarized their recommendations for success with SMP:

1. Clearly define the goals of SMP.
2. Monitor the orchard phenology. Apply SMP prior to the predicted maximum pollen flight in the orchard.
3. Use pollen (fresh or stored) with high viability.
4. Use an effective delivery system.
5. Monitor success with isozymes or other procedures.

Harvesting

Cones/seeds/fruits may be harvested by climbing, with bucket trucks, aerial lifts, tree shakers, or seed collection nets. Seed collection nets are effective when bulk collections are harvested from the orchard. When individual tree (or clonal) collections are needed the other systems must be used. The USDA Forest Service Missoula Equipment Development Lab developed the Net Retrieval System concept (Mc Connell and Edwards 1984), which has been widely copied and modified. This system is an effective method of harvesting southern pine seeds on nets when orchard mix collections are needed.

Genetic Gains

Realized genetic gains in volume growth from first generation southern pine clonal seed orchards have ranged from 6% with unrogued loblolly and slash pine orchards to 17% for rogued orchards of these species (Squillace 1989).

Advanced Generation Breeding

Advanced generation breeding often is designed to combine the best individuals from the best families in the first generation with unrelated individuals from a separate breeding population. The Western Gulf Forest Tree Improvement Program has developed a subline system which separates the breeding population into separate breeding groups which are allowed to mate and produce seed only when a production orchard is established (Lowe and van Buijtenen 1986). With this system, inbreeding is restricted to the breeding populations and the production populations are not affected.

A similar system has been used with Northern red oak in Indiana. Coggeshall and Beineke (1986) have designed 6 sublines with 30 clones in each for a total of 180 clones. These sublines will be crossed only when the production seed orchard is established.

The gains from advanced generation loblolly orchards have been predicted at 25% greater volume than unimproved material for the second generation, 35% for the third generation, and 45% for the fourth generation (Zobel and Talbert 1984).

Seedling Seed Orchards

When working with precocious species considerable time can be saved by collecting open-pollinated seed from select trees, growing the half-sib progeny in a nursery, and establishing a progeny test/seed orchard with the seedlings. A major problem with this system is to design a plantation which will be effective for progeny testing and will also permit effective seed production after the poor performers are removed. Effective designs have been developed by Wright (1976) and Hodge et al.(1995).

A recent report by Hodge and others (1995) indicated a gain of 10.7% in volume for a seedling seed orchard of longleaf pine at 8 years. In this case the heritability of volume growth was calculated at .21 and there was a moderate genotype x environment interaction related to geographic regions.

Accelerated Breeding

Accelerated breeding techniques have been developed in the last few years which will substantially reduce the breeding cycle. One of the most direct methods is selection at younger ages. A pilot scale accelerated breeding study was developed by van Buijtenen et al (1986). This study used a three-phase procedure with one-half of the loblolly families eliminated in each test. The tests included dry weight, root growth potential, and resistance to heat stress. The survivors of these tests were then subjected to flower induction techniques.

Potted seed orchards growing in greenhouses can reduce length of the breeding cycle by at least 20% (Zobel and Talbert 1984). In this case a 20-year cycle can be reduced to 16 years by accelerating flowering in the greenhouse as compared to a conventional outdoor seed orchard. The trees can be maintained in 20- to 40-gallon tubs with a drip irrigation system. McKeand and Weir (1983) calculated that a reduction of 6 years in the breeding cycle with a 30,000 seedlings/year regeneration program would amount to a $2 million savings. A similar system has been reported for western hemlock (Bower and others 1986). In this case potted ramets produced about 10 times the seed as field-grown trees.

With a potted orchard several techniques can be utilized to increase both male and female flowers. Water stress and applications of GA 4/7 will promote early female flowers (Todhunter 1988), while out-of-phase dormancy (Greenwood 1981) will speed up the development of male flowers. Wire girdling is also an effective way to promote male flowering.

Topworking grafted loblolly ramets has also accelerated flower production (Bramlett and Burris 1995). In this case both male and female flowers were produced in the year following grafting.

Deployment of Genetically Improved Material

Seed Zones:

Seed zones have been established for most of the major commercial forest species. These are areas which are environmentally similar and within which a given source can be expected to perform uniformly. When reforestation is needed within the zone seeds should be collected from that zone. In some cases when seed is not available from that zone, seed from an adjacent zone may be substituted.

In the western US seed zones can be quite narrow, depending on the topography. For example, seed zones for Douglas-fir in Oregon are delineated on 500-foot elevation intervals (Ching 1978). Restricted zones have also been recommended in the northern Rockies for western white pine and ponderosa pine (Rehfeldt and Hoff 1976).

In the South, seed zones for most species are much broader, reflecting the larger geographic provinces and more uniform topography (Lantz and Kraus 1987). West of the Mississippi River the Western Gulf Forest Tree Improvement Program has defined specific seed deployment zones (Byram et al. 1988), while to the East, the NCSU-Industry Cooperative Tree Improvement Program has adopted a more flexible approach (McKeand et al 1992)

Genotype x Environment Interactions:

Progeny tests of the first and second generation select trees have highlighted some outstanding families in the southern pines. Some of these families perform well on dry sites-some on wet sites- and some do well across-the-board. The famous International Paper clone, 7-56 for example seems to be a top performer wherever it is planted.

In general the genotype x environment interaction (change in relative rank) of most improved material has been unimportant. The University of Florida Cooperative Forest Genetics Research Program has reported strong G x E interaction for growth with some recent loblolly tests however (Hodge et al.1995).

Single-Family block Plantations

Forest industry in the South routinely establishes single-family block plantations and often records a growth advantage compared to mixed family blocks (Williams et al. 1983). Using block plantings rather than progeny tests, Gladstone et al (1987) recorded 16% greater stand volumes for single-family plantings compared to woods-run material. Mixed family blocks had only 11%
more volume than the checks.

Although single-family blocks may perform well on company land for short rotations, few non-industrial private forest landowners (NIPF) understand the risks involved. When a single family is planted on private land where long roations may be used and where natural regeneration may be employed, genetic diversity can be reduced to a low level. In only 1 or 2 cycles of natural regeneration inbreeding could seriously reduce growth and productivity.

Molecular Biology

Isozymes:

Brewbaker (1967) was one of the first scientists to propose the use of isozymes in forestry. Since then, isozymes have been widely used for taxonomic work, pollen contamination estimates, heterozygosity estimates, and a number of other uses. The concept of isozyme analysis is that a single gene codes for a single protein which can be graphically represented on an electrophoretic gel. Comparisons of protein samples on the stained gels may be interpreted as a direct reflection of the genotype of the tree. Cotyledons, needles or embryos (all diploid tissue) may be used or pollen grains and female gametophytes (haploid tissue) may be used.

Isozymes have been used to compare the rates of of heterozygosity and outcrossing as done by El-Kassaby et al. (1986) with Douglas-fir. The authors found no significant differences between clonal and seedling seed orchards in outcrossing rates. There were significantly greater proportions of homozygous progeny from the seedling orchard however.

Although isozyme analysis has been an effective tool for many forest genetics studies, Libby and others (1996) summarizing a southern meeting on genetic diversity, found that isozyme data have a number of limitations when used to estimate the genetic variation within a single species.

Isozyme analysis has been widely used to estimate the amount of pollen contamination in seed orchards: for example, Adams and Birkes 1989 (see Pollen Contamination-Seed Orchards).

The USDA Forest Service has established a National Forest Genetics Electrophoresis Laboratory in Camino, California where genetic variation studies, taxonomic determinations, "fingerprinting", and the effect of silvicultural and management procedures can be evaluated. This lab served an important role after Hurricane Hugo demolished the Francis Marion longleaf seed orchard in coastal South Carolina. Isozyme and DNA analyses were used to identify the surviving ramets in the orchard and facilitate reconstruction of the orchard.

Gene Mapping:

New techniques such as RFLP's (restriction fragment length polymorphisms)(Nance and Nelson 1989) and RAPD's (random amplification of polymorphic DNA) (Sewell and Neale 1995), have paved the way for significant advances in gene mapping of QTL's (quantitative trait loci). Conkle (1981) produced linkage maps for several Pinaceae species and Sewell and Neale (1995) constructed a "consensus" map for loblolly pine. Another mapping technique called PCR (polymerase chain reaction) markers has recently been developed for use with pines by Harry and Neale (1993). Other mapping work has been done with eucalyptus and poplar species. These mapping techniques are resulting in a great deal of data on the genome of loblolly pine. Hopefully this information will allow more efficient selection procedures (marker assisted selection) to be employed in the future.

In addition to the work done on pollen contamination and heterozygosity using isozymes, RAPD markers have been used to assess genetic variation in aspen following the 1988 Yellowstone fires (Tuskan 1995).

Fingerprinting:

Electrophoresis has been used for a number of years to "fingerprint" clonal material in seed orchards. Now PCR techniques have been used to identify Douglas-fir seed lots produced in a seed orchard in British Columbia and RAPD markers have been used to identify Norway spruce clones in Austria (Neale 1995).

Genetic Engineering:

Genetic engineering has received a considerable amount of attention by the media but few examples of forest tree application are available. There has been a case of gene transfer in hybrid poplar which conferred resistance to glyphosate (herbicide). There is also interest in tranfer of DNA with resistance to chestnut blight (Carraway et al. 1993). In this case somatic embryogenesis would be used to establish ovules and zygotic embryos on culture media.Transfer of this material would be accomplished by bombardment with plasmid DNA containing the resistant gene/s.

Tree Improvement Cooperatives

Tree improvement cooperatives have been established in the major timber growing regions of the U.S., including including California, the Pacific Northwest, the Inland Empire, the Lake States and the South. Advantages of these cooperatives include a long-term breeding plan, often developed by Forest Geneticists at a land grant university, statistical support for progeny test design and analysis, laboratory facilities for wood quality determinations and soil tests, and pollen and seed processing facilities. Technology transfer of new developments in the field of tree improvement and training is also an important function of these cooperatives.

Often select trees, pollen, seed, and grafting material are shared among members of the cooperative. Some cooperatives share orchards and even nursery sites. Duplication of effort is minimized and cooperative members gain significant economies of scale as they share breeding and testing workloads. Separate staffs are not needed by the individual organizations as thescientists and support personnel employed by the cooperatives are shared by all member organizations.

The South

The first tree improvement cooperative in the U.S. was initiated in 1951 when Bruce Zobel accepted a faculty appointment at Texas A & M University. Zobel organized a cooperative with 14 forest industries in the states of Arkansas, Louisiana and Texas. A few years later, one of Zobel's former graduate students, Tom Perry, organized the University of Florida Cooperative Forest Genetics Research Program. Zobel later moved to N.C. State University and organized the N.C. State University-Industry Cooperative Tree Improvement Program which is the largest of the three southern cooperatives. The Western Gulf Forest Tree Improvement Cooperative was organized by J.P. van Buijtenen in 1969 at Texas A & M University. These 3 southern cooperatives currently include 28 forest industries, 12 state forestry agencies, and 3 seed companies. Collectively these organizations produce an average of from 70 to 100 tons of pine seed annually and plant more than 1.8 million acres of land each year (Table).

Table :Summary of Southern Tree Improvement Cooperatives.

The Pacific Northwest

Tree Improvement activities started in the 1950's when the Industrial Forestry Association coordinated the establishment of clonal seed orchards for coastal Douglas-fir. In the 1960's forest industry hired forest geneticists and individual programs were started by a number of companies. About this time the USDA Forest Service started a tree improvement program for Douglas-fir, Western hemlock, ponderosa pine, Western white pine, and sugar pine on national forest land in Oregon and Washington (Daniels 1994). The Forest Service was followed in the 1970's by the Bureau of Land Management with a tree improvement program for Douglas-fir in western Oregon.

From 1967 to 1985 Roy Silen (USFS Pacific Northwest Forest and Range Experiment Station) and Joe Wheat (Industrial Forestry Association) developed 20 Douglas-fir cooperatives and 2 with Western Hemlock. These "Progressive Tree Improvement Programs" featured low intensity selection of large numbers of roadside trees followed by open-pollinated progeny tests. (In contrast with the high intensity selection practiced in most of the Southern Pine programs). In 1986, the Industrial Forestry Association-Pacific Northwest Program was changed to the Northwest Tree Improvement Cooperative of the Western Forestry and Conservation Association. This organization currently has 37 members, with a land base of 6.9 million acres and more than 80 breeding zones (Daniels 1994).

The overall Pacific Northwest region had a total of 282 seed orchards with a total of 3,473 acres in western Washington, western Oregon, and northern California in 1994 (Daniels 1994). Federal agencies own 64% of this acreage, industry 30%, and states and other cooperative groups 6%.

The Inland Empire

The Inland Empire Tree Improvement Cooperative membership has 20 separate organizations, including forest industry, state forestry agencies, tribal councils, federal agencies, universities, and other private organizations. The cooperative was established in 1978 by Lauren Fins at the University of Idaho, and covers Idaho, western Montana, amd eastern Washington. The cooperative has established 42 acres of western white pine and ponderosa pine orchards which have produced an average of 710 pounds of seed annually. In addition to these cooperative orchards, many member organizations have established their own orchards.

California:

The California Tree Improvement Association was organized in 1978 with 26 members managing over 9 million acres of forest land. Ponderosa pine was the first species selected, followed by Douglas-fir and sugar pine. Members included forest industry, the State of California and the USDA Forest Service. Local tree improvement associations were formed to focus on one or more of the California tree seed zones.

The main objectives of the association are selection of superior trees, establishment of clone banks, establishment of progeny test sites, and the establishment of a ponderosa pine seed orchard.

The Lake States:

The Minnesota Tree Improvement Cooperative was established in 1980 and currently has 18 Full Members and 7 Supporting Members. The cooperative is working with black and white spruce and jack, red and white pine. There are 35 seed orchards occupying about 125 acres. In 1995 about 84 bushels of cones were collected from 3 of these orchards. Six orchards were approved for production of certified seed in 1995. Gains in height growth have ranged from 3 to 9%.