Chapter 17
Genetic Implications of Nursery Practices

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Abstract

The genetic adaptation of forest trees to plantation sites can be impaired by nursery practices that favor the survival of some seedlings over others, thus producing a seedlot with a genetic makeup different from that of the original seedlot. Seed grading has considerable potential for directly altering the genetic mixture in the seedlot. Stratification period, sowing date, watering regime, lifting date, and other scheduling may have important but less direct influences on adaptation. For most seedlots, the risk of poor adaptation caused by nursery practices is probably no greater than risks caused by several current seed-collection practices. But for seedlots in which only a small percentage of seeds become seedlings that can survive outplanting, the risk may be as large as that in moving seeds between seed zones.

17.1 Introduction

The productivity of a forest plantation depends partly on the adaptation of seedlings to environmental conditions at the plantation site. If some seedlings, for example, break bud before the last spring frost, regenerate roots slowly when competition for moisture is high, or cease elongating when soil moisture is relatively high, their productivity will be low. Adaptation of seedlings to the planting site—or lack of it—reflects choices made by the forester, such as seed source, tree-breeding strategy, and planting-site preparation. But it also reflects seed- and seedling-related nursery practices, which can cause some plants to be culled or lost from the seedlot. In this chapter, we examine genetic implications of such practices in bareroot nurseries.

17.2 Genetic Principles

Although the genetic constitution of an individual plant (its **genotype**) cannot be measured directly, observable characteristics such as size and form (its **phenotype**) can. Phenotype is the result of the genotype's response to a particular environment. The average performance that would result if a seedling could be grown in a variety of environments is called its **genotypic value.** In most species of forest trees, each seedling has its own individual genotype and genotypic value.

Two assumptions are necessary to this discussion. First, the original seedlot provides the optimum genetic makeup for adaptation to the future planting site. The forest population has evolved through centuries of **natural selection** to contain a mixture of kinds of trees that match the climates of a seed zone and probably even microclimates of specific locations within that zone. This mixture is apparently balanced so that individuals can survive not only the stresses of the first few years but also competition from other plants and the rare climatic disasters that can occur during a tree's long life. The least risky procedure in the nursery is to maintain the original genetic mix in the seedlot.
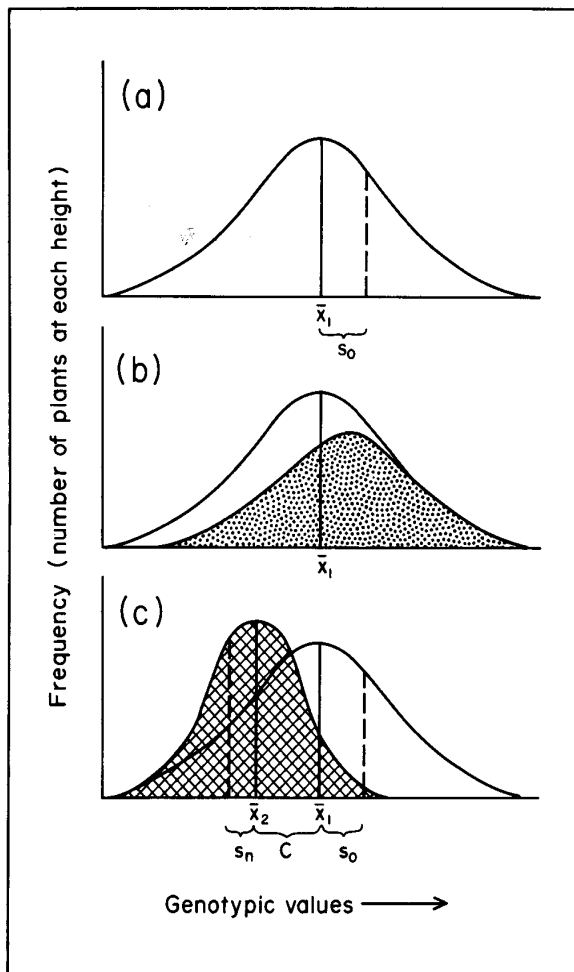
Second, culling and inadvertent favoring of certain plant types in the nursery result in **directional selection.** Selection is directional when one extreme type of seedling is saved, and the opposite extreme dies or is discarded. For example, culling all trees below a certain diameter limit selects for the larger diameter seedlings. Because phenotype depends partly on genotype, directional selection of phenotypes changes the genotypic mixture in a seedlot.

The power of directional selection (whether caused naturally or by humans) to change populations is well illustrated in a study of beech (*Fagus* spp.) in Germany [23]. Seeds from two locations (provenances), Bavaria and Rumania, were sown in a greenhouse and under a natural beech stand in Lower Saxony. After germination in the two habitats, the seedling populations were compared on the basis of genes expressed as enzymes (isozymes), and the two original seedlots were compared. Differences were measured in terms of genic distance, the frequency with which specific genes appear, and of genotypic distance, the frequency with which combinations of genes appear. The seed populations that germinated in the woods were genetically quite different from those that germinated in the greenhouse. The genetic distance, between populations caused by natural selection in the two environments was from 1/4 to 2/3 the distance that originally existed between seedlots of the two provenances.

Because phenotype depends partly on genotype, directional selection of phenotypes changes the genotypic mixture

in a seedlot. The genotypic values for the population of genotypes in a seedlot received at a nursery can be illustrated graphically (Fig. 1a). In this idealized figure, mean height ($\bar{x}_1$) of the original population occurs exactly in the center; the standard deviation, which measures variation of genotypic values around the mean, is represented by the distance s_0 on the x-axis. Though variation differs from trait to trait, remember that, for our discussion, all traits will be treated as having equal variation in the original seedlots.

Because phenotype describes response of a genotype to a specific environment, phenotypes are not precisely correlated with genotypes. Some seedlings are small because they have genotypes that produce small seedlings in an average environment. If, however, seedlings with "small" genotypes are blessed with more than average space, nutrients, or water, they may grow taller than seedlings with "tall" genotypes growing in an average environment. Culling of phenotypes, therefore, does not result in an exact culling of genotypes. In our example, culling the taller half of the seedlings in a seedlot does not remove all genotypes for tallest seedlings; it does cull most genotypic values from the taller half of the distribution as well

as a substantial proportion from the lower half, as illustrated by the stippled area in Figure 1b.

When natural or artificial selection removes seedlings at either the lower or upper end of the distribution of genotypic values, the distribution is changed in two ways. First, the remaining population has a different mean ($\bar{x}_2$); the change in mean value is represented by C in Figure 1c. Second, the variation around the new mean (s_n) is smaller than that around the original mean (s_0); note the narrower curve in Figure 1c. The cross-hatched area represents the shift in genotypic mixture: in this example, relatively more genotypes for short seedlings remain after culling than in the original mixture.

The method for calculating genetic changes is presented later in this chapter (17.5).

17.3 How Nursery Practices Alter Population Structure

Numerous separate practices—seed storage, stratification, sowing, fertilization, watering, weeding, lifting, culling, and packing—can alter nursery populations. Any alteration in population structure associated with these practices can be evaluated by assessing four factors:

- What proportion of the seedlings is lost
- What proportion of the loss is directional
- What correlation exists between phenotypes and genotypic values of the seedlings lost
- How large a correlation exists between the culled trait and some other trait that may cause growth loss or mortality at a later date

Other indirect effects may not appear until several years after outplanting; these include changes in growth-rhythm traits such as budset, which can cause changes in wood quality and disease resistance. Effects may not become evident until the planted stand is 1/3 or 1/2 rotation age. Types and sizes of such effects depend on correlations of phenotypes with genotypes (Table 1), which vary with traits, on correlations among genotypes (Table 2), which vary with combinations of traits, and on species; those given here are for Douglas-fir *[Pseudotsuga menziesii* (Mirb.) Franco].

Ultimately, seedling survival—and mortality—must be assessed. The most common term used to report nursery mortality is tree percent—the percentage of seeds that germinate and develop into usable seedlings ([5, p. 149]; see also chapter 5, this volume). Tree percent represents seedling survival to planting age and thus reflects both germinability of seed and the results of all nursery practices. But tree percent differs

Figure 1. Frequency distributions illustrating changes in the genotypic mixture for height of 2-year-old seedlings caused by culling the taller 50% of seedlings in a seedlot: (a) shows frequency of genotypic values (mean is $\bar{x}_1$ and standard deviation s_0) in the original seedlot; (b) indicates genotypic values removed by culling tallest seedlings (stippled area); (c) shows frequency of genotypic values after culling (cross-hatched area) —$\bar{x}_2$ is the new mean, s_n the new standard deviation, and C the change in mean genotypic value.

Table 1. Correlations between genotypic values and phenotypes for selected representative traits in Douglas-fir, based on nursery measurements of individual seed or seedlings.

Trait	Correlation
Seed weight[1]	0.35
Germination rate[2]	
4-week stratification	0.43
10-week stratification	0.50
Budset date, 1st year[3]	0.57
Budbreak date, 2nd year	0.65
Budset date, 2nd year	0.53
Height, 2nd year	0.44
Diameter, 2nd year	0.48

[1]Upper limit of correlation estimated by intraclass correlation coefficient calculated from data in Silen and Osterhaus [35].

[2]Estimates based on seeds from 200 open-pollinated families, west slope of the Cascade Mountains [unpubl. data, 8].

[3]Estimates of budset, budbreak, height, and diameter from 135 open-pollinated families from southern Oregon [unpubl. data, 8].

greatly among species, among seedlots within species (Table 3), and even among nurseries. Data from the Duncan Seed Centre, British Columbia Ministry of Forests [6], indicates that about 27% of the variation in average tree percent for several species was due to variation among nurseries.

Notwithstanding the voluminous literature on many aspects of nursery technology, we cannot now quantitatively estimate the genetic implications of individual nursery practices and may never be able to separate the effects of various practices. Seedling growth patterns reflect genotypes and their responses to the environment and competitive status with respect to neighboring seedlings. Because nursery conditions and practices—and chance—interact to create a seedling's environment, the effect of various practices on growth and survival is probably impossible to trace. Nevertheless, the genetic principles (see 17.2) are valid, and by keeping them in mind we can make reasonable guesses concerning which practices have the greatest potential for changing genetic structure. Seed-size grading,

Table 2. Genetic correlations between selected, representative traits of Douglas-fir observed in the nursery.[1]

	2nd year				
	Budbreak	Budset	Height	Diameter	Lammas shoot[2]
Budset, 1st year	0.59	0.56	0.96	0.46	0.63
Budbreak, 2nd year	. . .	0.52	0.65	0.10	-0.07
Budset, 2nd year	. . .	. . .	0.67	0.38	0.46
Height, 2nd year	. . .	. . .	. . .	0.79	0.95
Diameter, 2nd year	. . .	. . .	. . .	. . .	0.56

[1]Based on analysis of 135 open-pollinated families from southern Oregon [unpubl. data, 8].

[2]These correlations occurred only in a warm, moist environment that encouraged lammas-shoot growth in about half the seedlings.

stratification period, sowing date, and culling are important factors that may foster directional selection. Lifting date also may be important..

17.3.1 Seed-related practices

Within the nursery, mortality is most likely at or preceding germination and during culling. In species with poor germination capacity, most losses occur before germination, but in species with good germination capacity, losses are associated with culling [pers. commun., 31]. When seeds germinate poorly, seedbed density is low, and most seedlings exceed the culling standard; when seeds germinate well, seedbed density is high, and many seedlings are substandard.

Nursery managers attempt to adjust for differences in germination capacity between seedlots by calculating sowing density (see chapter 5, this volume). This undoubtedly helps but is not completely satisfactory. For some species, results of laboratory germination almost invariably overestimate field germination; the discrepancy is smallest for higher quality seeds that germinate rapidly [7]. Unfortunately, a sowing factor to adjust for such a discrepancy cannot easily be devised, particularly in view of the variability in germination time and percentage from year to year in most nurseries. Therefore, nursery practices which can potentially influence germination success should receive attention.

In Douglas-fir, at least two other pulses of selection come into play. Damping-off in the first few weeks after germination and factors associated with heat stress in late summer may contribute 20 to 30% of the mortality in the first growing season [pers. commun., 41]. Germination timing may be important because temperature in the first month after sowing influences disease incidence [4]. Environment and genetics determine the amount of selection, which varies by seedlot, year, nursery, seedbed, and fungus strain [3].

Table 3. Tree percents and corresponding Indexes of selection intensity for seedlots of representative western conifers after mortality and culling in the nursery.

Species	Tree percent[2]	Selection indexes[1] for change in	
		Mean genotypic value (i_1)	Variation (i_2)
White fir	6-60	1.98-0.64	0.85-0.58
[*Abies concolor* (Gord. & Glend.) Lindl. ex Hildebr.]			
Grand fir	2 5-70	1.27-0.50	0.76-0.51
[*A. grandis* (Doug]. ex D. Don) Lindl.]			
California red fir	4-75	2.15-0.42	0.87-0.47
(*A. magnifica* A. Murr.)			
Noble fir	30-70	1.16-0.50	0.74-0.51
[*A. procera* (Rehd.)]			
Sitka spruce	11-36	1.71-1.04	0.82-0.70
[*Picea sitchensis* (Bong.) Carr.]			
Engelmann spruce	30	1.16	0.74
(*P. engelmannii* Parry ex Engelm.)			
Lodgepole pine	48-7 5	0.83-0.42	0.64-0.47
(*Pinus contorta* Dougl. ex Loud.)			
Sugar pine	21-80	1.37-0.83	0.77-0.42
(*P. lambertiana* Doug].)			
Western white pine	32-90	1.14-0.19	0.72-0.29
(*P. monticola* Dougl. ex D. Don)			
Ponderosa pine	48-80	0.83-0.35	0.64-0.42
(*P. ponderosa* Doug]. ex Laws.)			
Douglas-fir	25-77	1.27-0.39	0.76-0.43
[*Pseudotsuga menziesii* (Mirb.) Franco]			
Western redcedar	25-43	1.27-0.91	0.76-0.67
(*Thuja plicata* Donn ex D. Don)			
Western hemlock	2-75	2.42-0.42	0.89-0.47
[*Tsuga heterophylla* (Raf.) Sarg.]			

[1]Selection index values from Shelbourne [34].

[2]Tree percents from U.S.D.A. Forest Service [42].

Quantitative estimates of seedling losses associated with individual nursery practices have not been reported. The few data published indicate tremendous variation among seedlots. For example, for Pinus species, Krugman and Jenkinson [25] reported that average nursery germination has ranged from 20 to 85% of the germination capacities found in laboratory tests; of seed germinated, as little as 19% and as much as 90% (average 55%) produced usable seedlings. For western hemlock [*Tsuga heterophylla* (Raf.) Sarg.], nursery germination ranged from 4 to 79% of viable seeds [7]; of seed germinated, as little as 12% and as much as 96% produced usable seedlings. Similar variation is found from nursery to nursery and seedlot to seedlot. At two Northwest nurseries [pers. commun., 31, 46], the consensus seems to be that patterns of mortality are distinct within seedlots and highly variable among seedlots, even within a single nursery. Whether the pattern for any given seedlot is consistent from year to year is unknown.

17.3.1.1 Seed-size grading

Seed-size grading provides an example of the complexity of predicting the effects of nursery practices. Retaining only part of the seedlot for sowing—usually the heavier seeds—has been used as a way to increase uniformity in seedling size [18, 28] and has received considerable attention in the literature. The practice provides a good example of directional selection. Seed size, however, is also strongly influenced by environment [18, 35] and year of collection [38]. Consequently, the correlation between genotypic value and phenotype is relatively small (0.35) (Table 1). In addition, seed size is affected by seed maturity—and the effects of maturity, in turn, are influenced by other nursery practices, such as duration of seed storage, stratification period, and germination temperature (Fig. 2).

Size grading that removes light seed, for example, also partly selects for genotypes encoding early seed maturity because mature seeds are usually heavier. Size grading thereby selects for other growth or developmental traits genetically correlated with early maturity. As Douglas-fir seeds mature, they gain in weight and germination capacity, right up to seed fall [32]. In some species, however, germination capacity may decrease at full maturity [15]. Maturation also varies from cone to cone within the same tree [10, 17, 30] and from tree to tree

and stand to stand [17, 35]. Consequently, variation in weight of seed in bulked lots undoubtedly reflects differences in seed maturity among trees and stands as well as time of collection. Furthermore, grading within a bulked seedlot may eliminate almost all seeds from some trees within seed zones and lots, reducing genetic variation within the lot [35]. Absence of grading, however, may have similar effects. Plants grown from smaller seeds in close mixture with plants grown from larger seeds often have smaller leaf areas in relation to their growing space and suffer disproportionate mortality [1].

17.3.1.2 Stratification period

Seed stratification and sowing date have considerable potential for changing genetic structure because they are correlated with so many aspects of growth and survival (see chapters 4 and 5, this volume).

Length of stratification affects germination energy (Fig. 2) and helps determine when an individual seed will germinate. Early germination usually increases risk of injury by spring frost, but individuals that survive are in a more favorable position to capture environmental resources [20]. These seedlings probably become more vigorous in the seedbed and are less likely to be culled or die by damping-off or heat stress. Therefore, any factor that increases variability of germination time in a seedlot favors early germinating seeds. A short stratification period, particularly when followed by cool germination temperatures, tends to promote such variability.

Germination time varies among seedlots from different geographic sources but also among seeds from trees within a single source and among seeds from a single tree. For example, consider the results from an experiment using seedlots from 185 seed trees from 100 sources in Oregon and Washington [unpubl. data, 8]. Seedlots from individual trees were stratified for 28 and 70 days and germinated at a constant 17°C. In seed stratified 28 days, variation among sources was 4 times greater than that in seed stratified 70 days. To a lesser degree, short stratification also increased variation among seeds from individual trees within sources. For seed stratified 28 days, 50% of the seed from the earliest germinating lots had germinated by 6.2 days, and 50% of the seed from the latest germinating lots by 11.9 days; comparable figures for seed stratified 70 days were 5.5 and 10.2 days. The greatest difference, however, was

Figure 2. The many paths by which conditions partly under human control may either directly (solid lines) or indirectly (broken lines) influence the number of seedlings remaining after culling or outplanting. For example, seed maturity and storage partly determine germination capacity, which influences the number of seedlings remaining after germination; some of those seedlings may then die, the number depending on several nursery practices. Total postemergent mortality affects seedling size and bed density, which together largely determine culling percentage. in a related path, sowing date affects emergence date. Disease and Its control, and water regime, correlate with emergence date and affect mortality before and after germination. Once again, mortality influences seedling size, bed density, and culling percentage. Shaded boxes are nodes at which data are sometimes available for calculating selection intensity.

among individual seeds from single trees. After 28 days of stratification, 99% of the seed had germinated by 34.12 days; after 70 days of stratification, 99% had germinated by 19.74 days.

The practical implication of length of stratification period is that mortality for both preemergent and postemergent seedlings is usually greater in seeds that are sown late or germinate late [2, 27]. Furthermore, seeds that germinate early have an advantage in competition and therefore tend to produce larger seedlings. In Campbell's [8] experiment, if only the seeds germinated in the first 7 days had survived, survival would have been 30% for seed stratified 28 days and 53% for seed stratified 70 days. But the generalization that early sowing produces more and better seedlings can be carried too far. There seems to be an optimum germination time in the nursery—sowing too early or too late results in suboptimal emergence [pers. commun., 40].

Differences in germination date probably would be even greater in the nursery because temperature during the germination period modifies the effects of stratification. Most nurseries sow in the spring throughout a period of rising temperatures; therefore, sowing date partly determines germination temperature. Variations in germination time among seeds, trees, and sources are exaggerated in cool temperatures [9]. Consequently, the shorter the stratification time and the cooler the temperature, the greater the variability in emergence date. We would expect the average temperature during nursery germination to be considerably lower than the 17°C used in Campbell's [8] experiment and the variability in days to germination to be correspondingly greater. Note, however, that stratification is likely to influence nursery performance significantly only in seedlots stratified for less than 60 to 90 days.

17.3.1.3 Sowing date

Sowing date modifies seedling growth in other ways. It is not surprising that seedlings from early sowings are larger [44], but other, more complex responses have been reported [37]. Sorensen [37] planted newly germinated seeds in seedbeds at intervals from April to June and found that the earlier the planting date, the longer the elongation period and the earlier the date of budset. At the end of the second season, this effect on budset persisted; seeds planted early set buds 17 days earlier than seeds planted later. But the early planting also shortened the elongation period in the second year. For germinants planted early, fertilizing the seedbed delayed first-year budset. Consequently, size of first-year seedlings was increased more by early sowing in fertilized plots than in unfertilized ones. The combined effect of fertilizers and early sowing persisted into the second year, increasing diameter disproportionately and thereby decreasing the height:diameter ratio of plants from seeds sown early.

Sorensen concluded that date of emergence influences not only a seedling's size and shape but also its growth rhythm. Such alterations of growth rhythm are not uncommon [21] and carry through at least 2 years and perhaps even beyond outplanting. Furthermore, the effect of sowing date can be modified by fertilization and other practices. But perhaps Sorensen's most significant finding is that genotypes reacted differently to sowing date. For some provenances, the amount and rhythm of seedling growth varied greatly according to sowing date; for others, variation was negligible. Sowing date apparently is an important factor affecting growth because it determines whether the climatic requirements of the seedling are well or poorly met by the nursery environment.

Because fertilizer affects seedling response to sowing date, the question arises as to whether moisture might also. In most nurseries, water is applied on a predetermined schedule. The average date of emergence of seedlings may or may not influence the start of the schedule, but, for the individual seedling, there will be some correlation between its emergence and its exposure to moisture saturation and depletion caused by the schedule. Therefore, emergence date partly determines the pattern of seedling moisture supply. To our knowledge, no reports describe interactions between seedling emergence and soil moisture.

Moisture stress in even the low to moderate range, however, affects budset date in 1-year-old Douglas-fir [19]. The practice of withholding water to induce early budset in nurseries is based on this reaction. Stresses are reflected in loss of potential height growth and thus photosynthetic capital (Fig. 2; also see chapters 12 and 15, this volume). As discussed earlier, treatments that change budset date may affect other aspects of growth rhythm and influence seedling development after outplanting (see 17.3.3). Furthermore, seed sources and, presumably, individual genotypes vary in their response to moisture stress [19, 45].

Length of stratification period and temperature during germination affect the variation in germination rate within seedlots. Consequently, practices or decisions that affect stratification and germination temperature tend to control variability among seedlings and opportunities for directional selection among genotypes. According to the OSU Nursery Survey (see chapter 1, this volume), Northwest nurseries prescribe stratification periods that vary greatly—for example, from 21 to 90 days for Douglas-fir. Shorter stratification periods, in particular, may induce considerable variability in seedbeds sown early or during unusually cool springs. Some seedlots are likely to react more strongly than others. The genetic implications, of course, are greater with poorer germination capacity and lower tree percent.

17.3.2 Seedling-related practices

17.3.2.1 Culling

Almost all Northwest nurseries surveyed (see chapter 1, this volume) reported culling for at least five traits: physical damage, multiple tops, height, stem caliper (diameter), and root mass. But not all losses due to culling cause directional selection.

Culling for physical damage probably results in selection without direction because most damage occurs randomly. Multiple tops may result from damage by natural agents such as frost and insects or from lammas-shoot growth (second flushing). If frost or lammas growth is the cause, selection is probably strongly directional. Frost damage is associated with phenological traits, such as budbreak and budset, which are strongly controlled genetically. Lammas growth tends to occur on the same individuals in successive years [12] and probably correlates well with genotype.

Culling for height and diameter will partly remove inbred plants and others that are weak or aberrant because of chance combining of bad genes from both parents [16, 36, 39]. This culling, although genetic, is not directional as we are using the term. Inbred seedlings tend to be small and more susceptible to disease and produce trees that cannot compete under forest conditions. Culling inbreds, therefore, probably improves the adaptedness and growth capability of the seedlot. Other culling for size will be directional but only partly effective in changing the mean genotypic value of the seedlot; the correlation values (Table 1) indicate only moderate genetic control of height (0.44) and diameter (0.48). Culling for root mass is probably similar to culling for top dimensions unless root diseases are involved, in which case culling may be more directional [11].

Even the mortality caused by natural selection in the nursery before and after seedlings emerge probably is not completely directional and certainly cannot be considered to be

selection for any single trait. Because researchers and managers lack knowledge about the complicated relationships among traits and acts of selection, however, the best procedure is to assume that directional selection applies to a single trait. On this assumption, we present indexes (explained in 17.5) of selection intensity (the proportion of seedlings remaining after culling and natural selection) that affect changes in genotypic value and variation among seedlots and species (Table 3). These indexes correspond with tree percents; for example, a tree percent of 6 indicates a high selection intensity of 1.98 for change in mean genotypic value and of 0.85 for change in variability. Because of the above qualifications, however, these intensities must be viewed as indicating the upper limit of directional selection resulting from all natural losses and directed culling from seed sowing to seedling packaging.

Some of the directional selection facilitated by variability among seedlings occurs at culling. Although the amount of selection contributed by culling is significant, 70 to 90% of seedlings normally are saved. Sometimes, however, the seedlings culled as being too small are sold as substandard lots or are transplanted (double-grading). Depending on the degree of culling, these seedlings represent 10 to 30% of the original seedlot. Foresters who want larger seedlings occasionally request heavier than normal culling, saving only 20 to 40% of the original lot. In these special cases, selection intensities are almost double normal intensities, shifting mean genotypic values of a seedlot twice as far as normal culling.

17.3.2.2 Lifting and storage

The final operation in which nursery practices contribute to directional selection is lifting (see chapter 21, this volume). Lifting date, modified by storage length [29] and storage conditions [28], accounts for a substantial part of first-year field mortality. Field survival of western conifers strongly correlates with lifting date [22]. Survival is affected by lifting date through its apparent control of root-growth capacity, which, in turn, depends largely on the chilling seedlings receive before lifting [26] and the effect of photoperiod on response to chilling [29]. Length of storage, independent of lifting date, can affect seedling height growth [33] and date of budbreak [24] for 1 to 3 years after outplanting.

Root-growth capacity seems highly correlated with budbreak timing [26] and with capacity for top growth after outplanting [22]. If root-growth capacity varies genetically within a seedlot, field mortality due to poor root-growth capacity undoubtedly favors some genotypes over others. The seedlings at greatest risk are those whose root-growth capacity has not been sufficiently enhanced by prelift chilling. Although natural selection only indirectly selects the genotypes that survive, phenotypes and genotypic values for developmental traits such as budbreak and budset are usually closely correlated (Table 1). Root-growth capacity probably is also strongly controlled genetically, and selection against incompletely chilled seedlings will change the genotypic mix of a seedlot.

Lifting and storage practices strongly affect natural selection after outplanting. Large changes in genetic structure are not expected from this selection, however, because poorly stocked plantations are often replanted. Failure rates in plantations in the Northwest may approach 30% [13], but survival through the first 2 years usually exceeds 50%, partly because most failed plantations (survival less than 20 to 25%) of public agencies are replanted. Thus, the influence of selection cannot be large unless established seedlings are the survivors of several successive regeneration attempts, each providing only a few seedlings from the much larger number planted. In lots made up of seedlings remaining after heavy nursery mortality, however, the intensity of accumulated selections from nursery and outplanting could be quite large.

17.3.3 Post-outplanting effects

Selection can also be fostered indirectly by any nursery practice that tends to produce seedlings that cannot survive after outplanting. The natural selection that occurs after outplanting can be attributed to nursery practices to the extent that the nursery has engendered a growth rhythm or physiological balance incompatible with that required by the plantation environment. For example, the proportion of nutrients allocated to roots, shoots, and needles may not be appropriate for the season or conditions at outplanting. Or the plant's dormancy cycle may be slightly out of phase with existing environmental conditions (see chapter 14, this volume). Therefore, any nursery practice or environment should be examined for potential causes of growth-rhythm incompatibilities if it produces seedlings with phenotypes characteristic of either very short or very long growing seasons, or if it favors (during culling, for example) seedlings with extra long or extra short vegetative cycles—which may, in turn, favor seedlings with extreme dormancy cycles after outplanting.

The proportion of plantation losses caused by nursery practices cannot be estimated on the basis of available information. Survival after 1 year in the field generally ranges from about 50 to 100% but can be lower. An additional 20 to 30% of surviving seedlings are sometimes lost during the next 2 years. By then, survival in some plantations may be less than 20%. Depending on factors such as site class, cause of loss, and economic constraints, forest managers usually decide to replant plantations with less than 20% survival [pers. commun., 14, 43]. Thus, survival of about 25% after natural selection—which can be considered the lower limit in young plantations—establishes the theoretical upper limit of selection intensity for post-outplanting effects of nursery practices. This limit corresponds to an index of 1.27 for change in mean genotypic value and 0.76 for change in variation (see 17.5). Thus, natural selection, combined with selection due to nursery practices, can cause significant changes in the genotypic mixture—although the specific impact of each remains indefinable.

17.4 Nursery Location

Because nursery environments influence the rhythm of seedling growth, any nursery with an environment greatly different from that of seedlot origin may induce directional selection within the nursery or after outplanting. Nurseries cannot, of course, be sited so as to optimally satisfy the environmental requirement of every seedlot. The choice then becomes one of selecting the nursery most appropriate for a seedlot.

With respect to genetic implications, the choice should depend on the amount of selection that occurs in the nursery and after outplanting. If, for example, a choice is made to sow eastern Oregon seed in nurseries in both eastern and western Oregon, both nursery tree percent and field survival should be closely followed for the first 5 years or so after outplanting. If seedlots planted in western Oregon nurseries have lower than average tree percents, are subject to heavier than average culling at the time of lifting, or routinely suffer heavier than average mortality after outplanting, there is genetic reason for raising the seedlings in "eastside" nurseries only. Observational evidence indicates that seedlots may react specifically to nurseries. Some lots have higher tree percents in some nurseries than would be expected from their performance at other nurseries [7]. This "nursery effect" may account for some of the large variation in tree percent among seedlots within species.

The genetic implications of nursery location may be greater for some species than others. Sitka spruce, western hemlock, and western redcedar survive poorly in all bareroot nurseries (Table 3). Whether this is due to nursery environment or to

cultural practices unsuitable for these species is not clear, but poor survival might be considered a genetic basis for restrict - ing a species to certain nurseries.

17.5 Calculating Genetic Changes

Recall Figure 1. The changes in mean genotypic value (C) and in genetic variation (so vs. so) produced by culling or other types of selection depend. on: (1) the correlation between geno- typic value and phenotype, h (see Table 1); the indexes of selection intensity, i_1 and i_2 (see Table 3); and (3) the genetic variation in the original seedlot, so.

The value of h ranges from 0 to 1. If, for example, h = 1 for seedling height, the nursery manager who discards the taller half of seedlings, as in Figure 1, would also be discarding exactly the upper half of genotypic values for seedling height— that is, everything above $\overline{x}_1$. In Figure (b, most of the stippled area representing culled genotypic values is above the mean, but some is below; therefore, h is greater than 0 but less than 1. The size of h can be modified by nursery practice; it can be increased somewhat by reducing the environmental variation within the seedbed. For instance, some seedlings may lack mycorrhizae, depending on chance distribution of fungal spores, and this may cause variation in seedling height. Use of proper inoculum could minimize this source of variation and improve the correlation between phenotype and genotype for height. Generally, however, the relative size of environmental varia- tion seems to be characteristic for each trait. For example, the correlation between genotype and phenotype is usually higher for budbreak or budset date than for height or diameter (Table 1); in other words, the average genotypic value would be changed more if seedlings were selected for budbreak or budset date than if they` were selected for height or diameter.

Values for selection-intensity indexes i_1 and i_2, derived from the normal curve, are found in prepared tables [34, p. 42 and 43]. The index i1 helps predict the average change in genotypic value, which depends on the proportion of plants saved. If, for example, all plants are saved, i_1 = 0; if 1% are saved, i_1 = 2.66. The index i_2 helps predict the average change in geno- typic variability in the seedlings left after selection. If all seed- lings are saved, i_2 = 0; if 1% are saved, i_2 = 0.90.

Yet another value, the genetic correlation between traits, r (see Table 2), is necessary because directional selection for one trait may affect the genetic structure for other traits. Values for correlation between genetic traits range from -1 to 1. A negative correlation implies, for example, that smallness in one trait is associated with largeness in another. Whether the correlation is positive or negative is not important here; we will use only positive values (0 to 1). The degree of correlation seems to be characteristic of traits and is not likely to be modified by nursery practice.

Four simple equations using combinations of h, i_1, i_2, s, and r provide approximations useful for indicating the genetic implications of a nursery practice. Two equations are needed to show the direct effect of culling on a trait; two others are needed to show the indirect effect on traits other than the one or several selected for.

The change in mean genotypic value of the primary trait may be represented:

$$C = i_1 \text{ x h x so}$$

Remember that all traits are treated as having equal genetic variation in the original seedlot. If. for example, s_o = 1 for all traits, it can be eliminated from the equation:

$$C = i_1 \text{ x h}$$

If 50% of seedlings are saved (i_1 = 0.80), as in Figure 1b, and if h for seedling height is, say, 0.4 5, then

$$C = 0.80 \text{ x } 0.45$$
$$= 0.36 \text{ standard units}$$

Thus, in this example, the mean genotypic value for height of seedlings left would be shifted from s_o by about 1/3 of a standard unit (Fig. 1c).

The variation left in the new population after seedlings have been culled either artificially or naturally may be represented:

$$s_n = s_o \text{ x} \sqrt{1 - (h^2 \text{ x } i_2)}$$

Then, on the basis of the above illustration:

$$s_n = 1 \text{ x } \sqrt{1 - (0.45^2 \text{ x } 0.64)}$$
$$= 0.93$$

Therefore, s_n is expected to be about 7% (1.00 - 0.93 = 0.07) smaller than s_o.

To indicate the indirect effects of culling on a related trait, two equations very similar to those just noted can be used. The change in mean genotypic value of the correlated trait (C_2)—in this case, diameter—may be represented:

$$C_2 = i_1 \text{ x h x r x } s_o$$

where so is the genotypic standard deviation for diameter. But if so = 1 for all traits,

$$C_2 = i_1 \text{ x h x r}$$

Assume r = 0.81 between seedling height and diameter. Then, by selecting 50% of seedlings for height, we change the mean genotypic value for diameter as follows:

$$C_2 = 0.80 \text{ x } 0.45 \text{ x } 0.81$$
$$= 0.29 \text{ standard units}$$

At the same time, s,, for diameter would be reduced by about 5%:

$$s_n = s_o \text{ x} \sqrt{1 - (h^2 \text{ x r x } i_2)}$$
$$= 1 \text{ x } \sqrt{1 - (0.45^2 \text{ x } 0.81 \text{ x } 0.64)}$$
$$= 0.95$$

In this section, we removed certain genotypes from the population present in the original seedlot to show how nursery practices can influence genetic structure. Results are the same whether removal is caused by natural or artificial selection. If culling is to change the average genotypic value of a population, the selection must be directional. If selection is directional, the effects of culling on mean genotypic values and variability may be quantified by indexes i_1 and i_2.

We have used equations including i, h, and r primarily to illustrate that the effects of nursery practices operate through all three factors. Because the assumptions are only partly met by these equations, the predictions are only approximations. The calculations encompass only the major, additive compo- nents of genetic variation. Nevertheless, the equations point out the type of information needed to identify nursery prac- tices that have potential for altering genetic structure.

If a practice directly or indirectly causes directional loss of a large proportion of seedlings, then i_1 or i_2 is large. If the trait affected by the practice (budset date, for example) also has a large h, then culling (or causing natural culling of) phenotypes for early budset accurately culls genotypes for early budset. We would expect such a practice to cause significant changes in the genotypic mixture—mean budset date would be delayed and variability of budset reduced. If early budset is closely correlated with another trait (frost resistance, for example), we would also expect similar changes in genotypic values for the secondary trait -the mean frost resistance would be decreased and would be less variable than in the original seedlot.

17.6 Genetic Risks of Nursery and Other Forestry Practices

Clearly, many nursery practices can change the genotypic mixture represented in a seedlot. We have hypothesized that

this will increase the risk that outplanted seedlings will be poorly adapted to the planting site. But how much does nursery-caused selection increase the risk? Because risk cannot be measured directly without long-term and expensive tests, we will attempt to evaluate it indirectly by assuming that selection is partly adaptational. We will assume that the original seedlot is best adapted to the field planting site, that directional selection in the nursery decreases adaptation to the field site, and that adaptation decreases as nursery losses increase. The loss can be equated to an adaptational risk and compared with adaptational risks associated with other forestry practices. Here, we compare adaptational changes associated with nursery practice with those arising from seed transfers during reforestation.

Let's look at two alternative nursery effects: an average loss which is partially directional and an extreme loss which is completely directional. Average loss might occur in a seedlot with relatively poor germination, lower than average seedbed density (and, consequently, light culling), but good lifting and storage conditions. Extreme loss might occur in a seedlot resulting from double-grading, planted on a severe site. For the average-loss situation, nursery tree percent is 50% and field survival 70%; half of the loss in both nursery and field is directional—correlation between phenotype and genotypic value is moderate (h is 0.45). For the extreme-loss situation, nursery tree percent is 25% and field survival 20%; all loss is directional—correlation between phenotype and genotypic value is fairly good (h is 0.65). By sequentially solving the equations given in 17.5—first for nursery selection, then for field selection—we find that the mean genotypic value of surviving plantings has been changed by about 0.31 standard units in the average-loss situation and 1.58 standard units in the extreme-loss situation.

A difference of approximately 0.3 standard units, as for the average-loss case, is equivalent to the difference in mean genotypic value expected between Douglas-fir stands in southern Oregon separated east to west by about 10 km, or north to south by about 44 km, or in elevation by about 125 m [unpubl. data, 8]. Equivalent distances for the extreme-loss case (difference of 1.58 standard units) are about 45 km east to west, or 170 km north to south, or 500 m in elevation. The increased risk in the average-loss case is probably no greater than would be expected from transferring seeds from collection location to plantation site within some of the standard seed zones in southern Oregon. In the extreme-loss case, however, risks might be as great as those encountered in moving seeds between adjacent or even more distantly separated seed zones. Unfortunately, we lack the long-term studies needed to judge whether the risk caused by moving seeds between zones is either negligible or important. In addition, standard deviations of genotypic values would be reduced by about 8% in the average-loss case and 33% in the extreme-loss case. This change might have additional and unknown effects on adaptation over the length of a rotation, such as providing less opportunity for the chance fitting of genotypes to suitable environments.

Other forestry practices may have genetic implications at least as important as those of nursery practices. One is collecting seed for reforestation in marginal seed years. Seedlots obtained in such years may include seeds from only a small, nonrepresentative sample of parent trees. Unfortunately, it is not possible to make valid comparisons between forest and nursery practices because the effects of forest practices on genotypic mixtures are even less understood than those of nursery practices. However, the nursery phase, which includes many intensive, interacting cultural operations that can alter the genotypic mix in seedlots, is critical to preserving the adaptedness of planted trees. Nursery managers should recognize the importance of their decisions and encourage the accumulation of knowledge about the genetic implications of nursery practices.

17.7 Conclusions and Recommendations

Nursery procedures that maximize tree percent and seedling survival after outplanting will minimize changes in the genotypic mixture of a seedlot. Because most nursery practices are designed to maximize the proportion of seeds that become healthy seedlings, the better the nursery management, the smaller the genetic change in the genotypic mixture.

Predicting seed recovery is a basic problem which, if solved, would answer many questions about the genetic implications of nursery practices. The problem has two parts. First, and most important, germination tests and field germination are poorly correlated. Because managers cannot predict accurately, they usually compensate by oversowing, which often produces too many small seedlings with poor root systems which are then culled after lifting. Not only is seed used inefficiently, but inadvertent genetic selection also occurs. Second, germination in the nursery is usually lower than that in the laboratory. This implies a loss of potentially healthy seedlings before germination and, perhaps, selection against genotypes adapted to the field situation.

Therefore, the first information needed is whether seedling loss caused by our inability to predict nursery germination changes the genotypic mix in seedlots. Isozyme analysis, which can monitor the fate of individual genes from newly germinated seed to outplanted seedling, may be a good approach for generating this information [23]. If overly dense beds are found to cause changes in the genotypic mix, then better prediction of germination will be important. The relationships among stratification period, germination temperature, and germination rate of seeds from many seedlots from known individual trees must be clarified. In particular, answers are needed for species or seedlots commonly producing low tree percents.

Another fruitful research strategy may be to study the seed recovery of specific seedlots planted in several nurseries over a number of years. We may find that some seed sources perform better or more predictably in some nurseries than in others. As long as any species or seedlot survives poorly in the field or nursery, any increase in survival can ameliorate potentially dysgenic effects caused by nursery practices.

References

1. Black, J. N. 1958. Competition between plants of different initial seed sizes in swards of subterranean clover (*Trifolium subterraneum* L.) with particular reference to leaf area and the light microclimate. Australian J. Agric. Res. 9:299-318.
2. Black, J. N., and G. N. Wilkinson. 1963. The role of time of emergence in determining the growth of individual plants in swards of subterranean clover *(Trifolium subterraneum* L.). Australian J. Agric. Res. 14:628-638.
3. Bloomberg, W. J. 1971. Diseases of Douglas-fir seedlings caused by *Fusarium oxysporum*. Phytopathology 61:467-470.
4. Bloomberg, W. J. 1973. Fusarium root-rot of Douglas-fir seedlings. Phytopathology 63:337-341.
5. Bonner, F. T. 1974. Seed testing. Pages 136-152 *in* Seeds of woody plants in the United States. U.S. Dep. Agric., Washington, D.C. Agric. Handb. 450.
6. Bowden-Green, R., and H. Rooke. 1980. Projected recovery of plantable trees per bed foot. Unpubl. rep. Mimeo. 1 p.
7. Buszewicz, G., and G. D. Holmes. 1960. A summary of ten years seed testing experience with western hemlock, *Tsuga heterophylla.* Rep. on Forestry Res., Great Britain J. Forestry Commun.: 110-119.
8. Campbell, R. K. 1982. Unpublished data, U.S.D.A. Forest Serv., Forestry Sciences Lab., Corvallis, Oregon.
9. Campbell, R. K., and S. M. Ritland. 1982. Regulation of seed germination timing by moist chilling in western hemlock. New Phytologist 92:173-182.
10. Ching, T. M. 1960. Seed production from individual cones of grand fir. J. Forestry 58:959-961.

11. Copes, D. L. 1982. Varied resistance to Phytophthora infection shown by cuttings of fourteen Douglas-fir clones. Annual meeting, Western Forest Genetics Assoc., Corvallis, Oregon, Aug. 3-5. Abstract.

12. de Champs, J. 1971. Pousses d'aout du Douglas. Pages 173-217 *in* Rapport Annuel L. Association Foretcellulose. Nangis, France.

13. Edgren, J. W. 1980. The reforestation system—a team effort. Pages 12-20 in Proc., North American forest tree nursery soils workshop (L. P. Abrahamson and D. H. Bickelhaupt, eds.). State Univ. New York, Coll. Environ. Sci. and Forestry, Syracuse.

14. Edgren, J. 1982. Personal communication, Region 6, U.S.D.A. Forest Serv., Portland, Oregon.

15. Edwards, D. G. W. 1978. Maturity and seed quality: a state of the art review. Pages 233-263 *in* Proc., A symposium on flowering and seed development in trees (F. Bonner, ed.). Mississippi State Univ., Starkville.

16. Eriksson, G., and D. Lindgren. 1975. Några genetiska reflexioner kring plantsortering. (Some genetic aspects on the grading of plants.) Sveriges Skogsvårdsförbunds Tidskrift 73:615-621.

17. Fowells, H. A. 1949. An index of ripeness for sugar pine seed. U.S.D.A. Forest Serv., Pacific SW Forest and Range Exp. Sta., Berkeley, California. Res. Note 64. 5 p.

18. Griffin, A. R. 1972. The effects of seed size, germination time and sowing density on seedling development in radiata pine. Australian Forest Res. 5:25-28.

19. Griffin, A. R., and K. K. Ching. 1977. Geographic variation in Douglas-fir from the coastal ranges of California. I. Seed, seedling growth and hardiness characteristics. Silvae Genetica 26:149-157.

20. Harper, J. L. 1965. Establishment, aggression and cohabitation in weedy species. Pages 243-268 in The genetics of colonizing species (H. G. Baker and G. L. Stebbins, eds.). Academic Press. New York.

21. Heide, O. M. 1974. Growth and dormancy in Norway spruce ecotypes. II. After-effects of photoperiod and temperature on growth and development in subsequent years. Physiologia Plantarum 31:131-139.

22. Jenkinson, J. L. 1980. Improving plantation establishment by optimizing growth capacity and planting time of western yellow pines. U.S.D.A. Forest Serv., Pacific SW Forest and Range Exp. Sta., Berkeley, California. Res. Pap. PSW-154. 21 p.

23. Kim, Z.-S. 1980. Veranderung der genetischen Struktur von Buchenpopulationen durch Viabilitatsselektion im Keimlingsstadium. Gottingen Res. Notes in For. Genetics, Lehrstuhl für Forstgenetik und Forstpflanzenzüchtung der Universität Göttingen. 88 p.

24. Kleinschmit, J., and J. Schmidt. 1974. Ergebnisse von Versuchen zur Kuhlhauslagerung von Forstpflanzen. Forstarchiv 45:165-170.

25. Krugman, S. L., and J. L. Jenkinson. 1974. *Pinus* L. Pine. Pages 598-638 in Seeds of woody plants in the United States. U.S. Dep. Agric.. Washington, D.C. Agric. Handb. 450.

26. Krugman, S. L., and E. C. Stone. 1966. The effect of cold nights on the root-regenerating potential of ponderosa pine seedlings. Forest Sci. 12:451-459.

27. Kummel, J. T., C. A. Rindt, and J. T. Munger. 1944. Forest planting in the Douglas-fir region. U.S.D.A. Forest Serv., Portland, Oregon. 154 p.

28. Lavender, D. P. 1978. Seed collection, handling, and damage. Pages 53-62 in Regenerating Oregon's forests (B. D. Cleary, R. D. Greaves, and R. K. Hermann, eds.). Oregon State Univ. Ext. Serv., Corvallis.

29. Lavender, D. P., and P. F. Wareing. 1972. Effects of daylength and chilling on the responses of Douglas-fir [*Pseudotsuga menziesii* (Mirb.) Franco] seedlings to root damage and storage. New Phytologist 71:1055-1067.

30. Maki, T. E. 1940. Significance and applicability of seed maturity indexes for ponderosa pine. 1. Forestry 33:55-60.

31. Morgan, P. 1982. Personal communication, D. L. Phipps Oregon State Forest Nursery, Elkton.

32. Olson, D. L., and R. R. Silen. 1975. Influence of date of cone collection on Douglas-fir seed processing and germination: a case history. U.S.D.A. Forest Serv., Pacific NW Forest and Range Exp. Sta., Portland, Oregon. Res. Pap. PNW-190. 10 p.

33. Rasanen, P. K. 1970. The effect of lifting date, packing, storing and watering on the field survival and growth of Scots pine seedlings. Acta Forestalia Fennica 112:1-27.

34. Shelbourne, C. J. A. 1969. Tree breeding methods. Forest Res. Institute, New Zealand Forest Serv., Rotorua. Tech. Pap. 55. 43 p.

35. Silen, R., and C. Osterhaus. 1979. Reduction of genetic base by sizing of bulked Douglas-fir seed lots. Tree Planters' Notes 30:24-30.

36. Snyder, E. B. 1972. Five-year performance of self-pollinated slash pines. Forest Sci. 18:246.

37. Sorensen, F. C. 1978. Date of sowing and nursery growth of provenances of *Pseudotsuga menziesii given* two fertilizer regimes. J. Applied Ecology 15:273-279.

38. Sorensen, F. C., and J. F. Franklin. 1977. Influence of year of cone collection on seed weight and cotyledon number in *Abies procera*. Silvae Genetica 26:41-43.

39. Sorensen, F. C., and R. S. Miles. 1974. Self-pollination effects on Douglas-fir and ponderosa pine seeds and seedlings. Silvae Genetica 23:135-138.

40. Tanaka, Y. 1982. Personal communication, Western Forestry Res. Center, Weyerhaeuser Co., Centralia, Washington.

41. Thompson, B. 1982. Personal communication, Western Forestry Res. Center, International Paper Co., Lebanon, Oregon.

42. U.S.D.A. Forest Service. 1974. Seeds of woody plants in the United States. Part 2. Dep. Agric., Washington, D.C. Agric. Handb. 450. 889 p.

43. Van Den Berg, D., and I. Holen. 1982. Personal communication, Medford District, Bureau of Land Management, Medford, Oregon.

44. van den Driessche, R. 1969. Forestry nursery handbook. B. C. Forest Serv., Victoria. Res. Note 48. 44 p.

45. White, T. L. 1981. Genecology of Douglas-fir from southwestern Oregon. Ph.D. thesis, Oregon State Univ., Corvallis. 103 p.

46. Wilson, B. 1982. Personal communication, Webster Nursery, Olympia, Washington.