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Effects of Sib Mating on Cone and Seed Traits in Coastal Douglas-Fir

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Summary

Outcross, half-sib and full-sib, and self matings were made in an Oregon seedling seed orchard in 1987 and 1989. Cone and seed traits other than fertility, but including germination, were almost exclusively under maternal influence. The relations between 3 measures of fertility and inbreeding coefficient (F) were linear or predominantly linear. Our results were very similar to those previously reported by WOODS and HEAMAN (1989), and combining their results with ours gave 40.2, 30.9, 23.2, and 1.15 filled seeds per cone after cross, half-sib, full-sib, and self pollination, respectively. Selfing appeared to reduce seed set more in the orchards than in natural stands. Assuming linear relations between F and fertility and between F and vigor, it was shown that partial selfing between zero and 50% has a greater impact than partial sibbing on seed set; but that partial sibbing, particularly full-sibbing, has the greater impact on population vigor and wood production.

Key words: *Pseudotsuga menziesii*, seed orchard, inbreeding, inbreeding depression, fertility, germination, mixed mating.

ADC: 165.41; 181.5; 232.311.3; 174.7 *Pseudotsuga menziesii*.

Introduction

Many conifer seed orchards in the Pacific Northwest have been established with full- and half-sib seedling progenies, sometimes in conjunction with clonal material from parents of the progenies (WHEAT and BORDELON, no date). Having related individuals in the same orchard provides opportunity for both selfing and several types of lower intensity inbreeding. In this paper, we report on the effects of self, full-sib, half-sib, unrelated, and wind pollination on cone size and seed traits, including germination, using trees in a Douglas-fir seedling orchard in Oregon. Results from 2 years of pollination are compared with those reported by WOODS and HEAMAN (1989), who contrasted several levels of inbreeding with outcrossing in their effect on seed production in a Douglas-fir (*Pseudotsuga menziesii* (MIRB.) FRANCO) full-sibling factorial progeny test and in clone banks for the factorial on southern Vancouver Island.

Materials and Methods

All pollinations were made in the Vernonia block of the J. E. Schroeder Cooperative Seed Orchard, managed

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by the Oregon State Department of Forestry and located in the Willamette Valley about 25 km north of Salem, Oregon, at an elevation of about 50 m. Parent trees of the Vernonia block progenies are native at low elevation (250 m to 500 m) in the northwest corner of the Oregon Coast Range. The block contains full-sib families from biparental matings made in the natural stands. In a few cases, more than 1 family has a common parent.

Trial 1 pollinations were made in 1987 in the oldest portion of the Vernonia block. Trees were 15 years old and had a mean height of about 12 m. Pollinations were made on a total of 20 seed trees representing 12 full-sib families; i.e., trees were replicated within 8 of the families. Self (inbreeding coefficient, $F = 0.5$), full-sib ($F = 0.25$), and outcross polymix ($F = 0$) pollens were applied to each seed tree, and half-sib ($F = 0.125$) pollens were applied to 3 trees. The outcross polymix included equal volumes of pollen from 3 to 5 families, unrelated to the seed trees, but in the same orchard block. Several mixes were used in the course of the pollinations.

Isolation bags were applied to branch ends in the upper parts of crowns shortly before floral bud burst. Pollen was obtained from microsporangiate strobili on detached twigs after drying at about 25 °C in a low-humidity room. For pollination, bags were removed, pollen was poured from a small vial onto receptive female strobili (OWENS et al., 1981), and isolation bags were immediately reinstalled on branches. If any female strobili were not receptive at the time of first pollination, the bag was so marked and the flowers within it repollinated 3 to 4 days later. Only fresh pollen was applied. All work was done from a hydraulic lift. To control insect damage to cones and seeds, isolation bags were left in place until cone harvest.

Cones were collected in late summer shortly before scales started to flare. Five undamaged cones from each mating, if available, were bagged separately and set aside for dissection and estimation of seed production. (Seeds were extracted from other cones but were not used in determining seed production traits.). Sample cones were individually sealed in small kraft bags, initially dried in an open air shed, and given final drying at 20 °C to 25 °C in a heated room with dehumidifier. Cones were then taken apart by hand and, within the presumed fertile part of each cone, undeveloped flat and normal-appearing round seeds were counted. Round seeds were x-rayed and filled and empty seeds counted. Total filled seeds from each cone were weighed to the nearest mg.

Four traits were analyzed:

- 1) total seeds (flat + round) per cone;
- 2) percent of total seeds that were flat;
- 3) percent of round seeds that were filled; and
- 4) weight of filled seeds.

Regression and least-squares analyses of variance were applied to the 4 variables. A spring frost damaged the cones on some trees; 12 seed trees in 6 families had cones in all planned matings. Cones and seeds from these crosses were used in the analyses of variance. Arc sine transformations were used on percents (NETER and WASSERMAN, 1974) to make variances independent of means.

The mating structure was analyzed as a modified split plot with hierarchical classification (PETERSEN, 1985; ROWE, K., Department of Statistics, Oregon State University, Corvallis, personal communication) with seed trees nested within full-sib families and inbreeding treatments the same across seed trees. Seed-orchard families, seed trees within families, and cones within pollen treatments were random effects, inbreeding levels were fixed. Variance components were estimated following STEEL and TORRIE (1960). Additional details are given in CRESS (1989).

Because of frost damage to many of the conelets after the 1987 pollinations, trial 2 was installed in 1989. Floral production in both the older Vernonia orchard block and in a sub-block 3 years younger was stimulated the preceding year by partial girdling and fertilization with calcium nitrate (EBELL, 1971, 1972). The mating design was modified by using only one seed tree per orchard progeny, omitting self pollination which, as observed by WOODS and HEAMAN (1989), has extremely low seed yields, and by making more half-sib matings. The latter were made possible by including families in the younger subsection of the Vernonia block.

Twenty-four seed trees were pollinated with full-sib and polymix outcross and wind pollens as described under trial 1 except that pollen was applied with an atomizer. Fourteen of these trees also received half-sib pollen. Four of the half-sib matings had higher filled-seed percents than did their corresponding outcrosses. There was also a break between their filled-seed values and those of the other 10 matings. These half sibs were deleted from the analyses, because of the possibility of misidentification either at the time of initial field crossing or subsequently. Misidentification is not uncommon (ADAMS, 1981), but alternative explanations are discussed later. Six undamaged cones from each cross were hand-dissected, round seeds extracted, and the following cone and seed traits analyzed:

- 1) cone length (log transformation);
- 2) round seeds per cone;
- 3) filled seed percent (arc sine $\sqrt{\%}$);
- 4) filled seed weight (mg³⁵);
- 5) mean germination rate of 60 seeds per petri dish (days⁻¹); and
- 6) standard deviation of germination rate of 60 seeds (days⁻¹).
- 7) Germination percent also was recorded but was not analyzed because germination of filled seeds was essentially complete for all crosses.

Transformations (NETER and WASSERMAN, 1974; HINZ and EAGLES, 1976; CAMPBELL and SORENSEN, 1979) removed or greatly reduced heterogeneity of variances.

Seed tree and pollen type (inbreeding level + wind) effects were analyzed in 2 sets, 1 (set A) with 24 seed trees crossed with full-sib, polymix outcross, and wind pollens, and a second (set B) with 10 seed trees crossed with the

same 3 classes of males plus half-sib pollen. Analysis of variance followed that described in trial 1, except that there was not an hierarchy of seed trees within orchard families. In set-B analyses, linear and nonlinear effects associated with levels of inbreeding and their interactions with seed trees were tested within the analysis of variance. Results from set B are reported only for traits in which inbreeding effect is significant, and then to identify whether the change in the trait is linear or nonlinear with F.

The germination test was conducted on 3 replications of 60 seeds per family, soaked 24 hours in aerated, distilled water at room temperature (22 °C), stratified 71 days at 2 °C to 3 °C, and incubated at constant 15 °C with day length of 12 hours. Seeds were placed on filter paper in covered petri dishes and moistened with distilled water. Each replication was in a separate incubator. Counting and estimation of mean rate of germination and its standard deviation for seeds within a petri dish followed CAMPBELL and SORENSEN (1979). Abnormal germinant seedlings (twin, reverse, swollen megagametophytes) were not included in counts. The test ran 20 days, at the end of which ungerminated seeds were cut and classified as firm, empty, insected (seed chalcid, *Megastigmus spermotrophus*), or soft.

Results

"Seed potential" was estimated in trial 1 from counts of all seeds in the assumed fertile portion of the cone. It averaged 73.1 per cone with a range of 48 to 116. Round seeds per cone, which are perhaps the more true potential for producing filled seeds, averaged 61.7 and 56.7 in trials 1 and 2, respectively.

Cone Size and Seed Production other than Fertility

These traits were largely under maternal influence; that is, seed parent effect had much larger component of variance than did level of inbreeding (Tables 1 and 2). The only consistent effect due to type of male parent was observed in filled-seed weight in trial 2 (Table 2) in which weights were significantly lighter after wind pollination (12.9 mg/seed) than after control pollination (13.6 mg/seed). This was undoubtedly an isolation bag effect and has been reported previously for Douglas-fir (SOESEN and CAMPBELL, 1985) and *Picea abies* (SKRØPPA and THO, 1990).

We did not observe a significant effect of inbreeding on seed weight (Tables 1 and 2), although WOODS and HEAMAN (1989) did report a small effect, as did ORR-EWING (1954) when comparing seeds from selfing and outcrossing. Two factors may be involved in giving the contrasting results. Because megagametophyte and seed coat are maternal, presumably only the weight of the embryo is affected by inbreeding, and the embryo contributes little to total seed weight (STOCKWELL, 1938; BUCHHOLZ, 1946; SURLS et al., 1993). Second, seed size has been observed to increase with decreasing numbers of filled seeds per cone (SIMAK and GUSTAFSSON, 1954). There were many fewer filled inbred than outcross seeds per cone, and perhaps in the St. Paul Orchard environment this compensated for any inbreeding depression in seed weight. SKRØPPA and THO (1990) did not find a selfing effect on seed weight in *Picea abies*.

Interaction between orchard family and inbreeding level was significant for flat seed percent in trial I (Table 1). Because female gametophytes are fully developed before fertilization (ALLEN and OWENS, 1972), the interaction indicates that male pollen impacts megagametophyte development in some seed trees prior to fertilization.

Table 1. — Variance components^{a)} for total seed number per cone, flat seed proportion, and weight of filled seeds in trial 1 (1987 pollinations).

Sources of variation	Degrees of freedom	Seed number ^{b)}	Flat seeds ^{c)}	Seed weight ^{d)}
Orchard progeny (P)	5	34.3**	2.8	38.6*
Seed tree in progeny [T(P)]	6	27.0***	22.7***	20.5***
Inbreeding level (I)	2	0	0	0
P x I	10	0	17.2*	1.2
T(P) x I	12	0	5.9	0
Cones in T(P) x I	48	38.7	51.4	39.7

^{a)} Variance components given as percents; that is, as the proportion of variation assigned to each of the sources of variation.

^{b)} Seed number = number flat + number round seeds.

^{c)} Flat seed proportion = number flat seeds ÷ seed number.

^{d)} Mean weight of filled seeds.

^{e)} Levels of significance are * = $p < 0.05$; *** = $p < 0.001$.

Seed parent by male interactions in trial 2 were very highly significant (Table 2). For all traits, the contribution to interaction was greater when wind pollination (m4 in Table 2) was included in the interaction (Table 2, interaction term, T x [(m1 + m3) vs. m4]) than when it was not (interaction term, T x [m1 + m3]).

Fertility

Fertility was recorded in 3 ways: 1) filled seeds per cone, 2) filled seed percent (filled seed number ÷ round seed number), and 3) relative fertility [filled seed percent after

inbreeding) ÷ (filled seed percent after outcrossing)]. In contrast to seed and cone traits described previously, fertility was predominantly associated with level of inbreeding, and the effect was strongly linear with inbreeding coefficient, F (results presented for Filled Seed Percent in Tables 3 and 4). Interaction terms between seed tree and inbreeding level are mostly significant (Tables 3 and 4). This could have been due partly to technical problems during pollination, but also large variation among seed trees has been reported previously for inbreeding depression in seed set after self pollination (ORR-EWING, 1954 and 1957; PIESCH and STETTLER, 1971; SORENSEN, 1971), and after sib pollination (WOODS and HEAMAN, 1989).

Assuming linearity of response between fertility and inbreeding coefficient, a change of 0.1 in F was associated with mean changes of 6.8 filled seeds per cone, 12.6 in filled seed percent, and 0.18 in relative fertility. The last measure, which is independent of seed potential, means that seed set approaches zero at F = 0.5 (selfing), which seems to be true for Douglas-fir. This relation estimates relative self fertility as 0.10. Seed orchard values were lower (0.03 and 0.038, Table 5), but 0.10 is very close to the mean value of 0.113 estimated from selfing and outcrossing 35 naturally occurring Douglas-fir trees (SORENSEN, 1971).

Germination

The germination test was run only on seeds from the 1989 pollinations. Seeds still ungerminated at the end of the test were cut to determine if they were filled. Forty-eight (0.3% of initial seed number) were filled, 40 from wind-pollination families (22 from 1 family), and 4, 2, and

Table 2. — Variance components^{a)} for cone length, round seed number per cone, weight of filled seeds, mean germination rate, and standard deviation of rate for seeds within a petri dish for trial 2 (1989 pollinations). Sources of variation include 24 seed trees and outcross (m1), full-sib (m3), and wind(m4) pollinations.

Sources of variation	Degrees of freedom	Cone length ^{b)}	Round seeds	Seed weight ^{c)}	Germination rate	
					Mean ^{d)}	S.D. ^{e)}
Seed parent (T)	23	78.4*** ^{f)}	44.8***	78.6***	73.4***	36.9***
Male type (M)	2	0	0	1.9**	0.5	3.2*
(m1 + m3) vs. m4	1	n.s.	n.s.	***	n.s.	*
m1 vs m3	1	n.s.	n.s.	n.s.	n.s. ^{g)}	n.s.
T x M	46	5.3***	19.3***	5.3***	15.3***	21.0***
T x [(m1 + m3) vs. m4]	23	***	***	***	***	***
T x (m1 vs. m3)	23	***	*	***	***	n.s.
Cones in T x M ^{h)}	348	16.3	36.3	14.2	10.8	38.9

^{a)} Variance components given as percents; that is, as the proportion of variation assigned to each of the sources of variation.

^{b)} Cone length in \log_{10} cm.

^{c)} Seed weight in mg^{10} .

^{d)} Mean germination rate (days^{-1}) for 60 seeds in a petri dish.

^{e)} Standard deviation of (^{d)}.

^{f)} Levels of significance are n.s. = $p > 0.05$; * = $p < 0.05$; ** = $p < 0.01$; *** = $p < 0.001$.

^{g)} $p = 0.076$ for contrast, outcross vs. full sib.

^{h)} For germination traits, the interaction term involving blocks replaces cones in T x M as the error.

2 from outcross, half-sib, and full-sib populations, respectively.

Germination rate and standard deviation of rate were again predominantly under maternal influence (Table 2). A small inbreeding effect on mean germination rate pos-

sibly was indicated in the contrast, outcross vs. full sib, which approached significance for mean germination rate (Table 2, footnote g). In set B crosses, which included half sibs, the linear response of mean germination rate to inbreeding coefficient also approached significance [$p =$

Table 3. — Variance components^{a)} for filled seed percents (arc sine $\sqrt{\%}$) for trial 1 (1987 pollinations). Treatments included full-sib orchard progenies, 2 seed trees per progeny, 3 levels of inbreeding, outcross polymix (F = 0), full sib (F = 0.25) and self (F = 0.5), and 5 sample cones per mating.

Sources of variation	Degrees of freedom	Variance component
Seed orchard progeny (P)	5	0.1
Seed tree in progeny	6	5.7*** ^b
Inbreeding level (I)	2	69.3***
I _{linear}	1	***
I _{nonlinear}	1	n. s.
P x I	10	1.4
P x I _{linear}	5	n. s.
F x I _{nonlinear}	5	n. s.
T(P) x I	12	5.5**
T(P) x I _{linear}	6	n. s.
T(P) x I _{nonlinear}	6	**
Cones in T(P) x I	144	18.0

^{a)} Variance components given as percents of variation assigned to each of the sources of variation.

^{b)} Levels of significance are n. s. = $p > 0.05$; ** = $p < 0.01$; *** = $p < 0.001$.

Table 4. — Variance components^{a)} for filled seed percents (arc sine $\sqrt{\%}$) for trial 2 (1989 pollinations). Treatments included seed trees, 4 pollen types, polymix outcross (m1, F = 0), half sib (m2, F = 0.125), full sib (m3, F = 0.25) and wind (m4), and 6 sample cones per cross.

Sources of variation	Degrees of freedom	Variance component
Seed trees (T)	9	8.4*** ^b
Male types (M)	3	50.9***
(m1 + m2 + m3) vs m4	1	***
m1 vs m2 vs m3 _{linear}	1	***
m1 vs m2 vs m3 _{nonlinear}	1	n. s.
T x M	27	21.5***
T x [(m1 + m2 + m3) vs m4]	9	***
T x (m1 vs m2 vs m3 _{linear})	9	***
T x (m1 vs m2 vs m3 _{nonlinear})	9	***
Cones in T x M	185	19.2

^{a)} Variance components given as percents; that is, as the proportion of variation assigned to each of the sources of variation.

^{b)} Levels of significance are n. s. = $p > 0.05$; *** = $p < 0.001$.

Table 5. — Effect of outcrossing and levels of inbreeding on filled seeds per cone, filled seed percents, and fertilities relative to outcross. First column gives source of values, T1 and T2 = 1987 and 1989 trials, respectively, from this paper; WH = Woods and HEAMAN (1989). Last 2 columns are the coefficient of determination for the linear correlation between the trait and the inbreeding coefficient, F, and the change in the seed trait associated with a change of 0.1 in F.

Data source	Mating type					R ²	Δ0.1F
	Outcross	Half sib	Full sib	Parent-offspring	Self		
Filled seeds per cone							
T1	32.6	---a	18.5	---	1.1	0.996	6.3
T2a ^b	47.3	---	29.1	---	---	---	---
T2b	49.2	40.5	28.4	---	---	0.991	8.3
WH	31.6	21.3	16.7	15.5	1.2	0.989	5.9
Filled seed percents							
T1	55.0	---	28.6	---	1.8	0.999	10.6
T2a	82.4	---	51.8	---	---	---	---
T2b	88.2	79.7	51.5	---	---	0.912	14.7
Relative fertility							
T1	1.000	---	0.520	---	0.033	0.999	0.193
T2a	1.000	---	0.629	---	---	---	---
T2b	1.000	0.904	0.584	---	---	0.912	0.166
WH	1.000	0.674	0.528	0.491	0.038	0.990	0.190

^a) --- = no mating was made.

^b) T2a included 24 seed trees, but only cross and full-sib matings; T2b is based on 10 seed trees each with cross, half-sib, and full-sib progenies.

0.070; mean rates, outcross, 0.1757 (5.69 days); half sib, 0.1730 (5.78 days); full sib, 0.1706 (5.86 days)]. In other words, it took about 3% longer for full sibs to reach 50% germination than it did for outcrosses. The difference is small, but it is compatible with inbred embryos being slightly smaller or less vigorous on the average than outcross embryos. FRANKLIN (1969) likewise reported a small decrease in speed of germination of self-pollinated compared with cross-pollinated seeds, and SQUILLACE and KRAUS (1962) reported that both rate and percent of seedbed emergence were decreased by inbreeding in *Pinus elliotii*. SAITÔ et al. (1973) did not note any consistent difference in germination percent of filled *Pinus thunbergii* seeds between $F = 0$ and $F = 0.75$.

Male effects were also significant for standard deviation of the germination rate (Table 2), but the only significant contrast was between controlled crosses pooled and wind pollination: wind-pollination seeds had larger standard deviation of germination rates (0.0198 vs 0.0178 days⁻¹).

Discussion

Linearity of Response to Inbreeding

The relation between mean performance and decrease in heterozygosity should be linear regardless of degree of dominance, unless linkage or epistasis is involved (WRIGHT, 1922; WILLIS, 1993). A sample of the literature indicates variable results when this is tested. Linearity between F and growth or fertility traits is reported by SQUILLACE and KRAUS (1962), SING et al. (1967), HALLAUER and SEARS (1973), SAITÔ et al. (1973, 1986), CORNELIUS and DUDLEY (1974), BURTON et al. (1978), SPRAGUE (1983), WOODS and HEAMAN (1989), WILLIS (1993), and WOODS and STOEHR (1993). In contrast, ANDERSSON et al. (1974) and GRIFFIN and LINDGREN (1985) indicate little or no inbreeding depression at $F = 0.125$ or $F = 0.25$, while LAYTON and GODDARD (1983) found more inbreeding depression between $F = 0$ and 0.125 than between $F = 0.125$ and $F = 0.25$. GRIFFIN and LINDGREN (1985) develop an epistatic model to explain the curvilinear response.

Our fertility results with Douglas-fir also are potentially ambiguous. Four of the 14 families in trial 2 showed no inbreeding depression in seed set at $F = 0.125$. If these are excluded, all other data indicated no deviation from linearity (Table 4, m_1 vs. m_2 vs. $m_{3, \text{nonlinear}}$, nonsignificant), and the trial 1 data, which didn't include $F = 0.125$ matings, was linear (Table 3, $I_{\text{nonlinear}}$, nonsignificant). Other results with Douglas-fir also indicate a linear response between $F = 0$ and $F = 0.5$ (WOODS and HEAMAN, 1989; WOODS and STOEHR, 1993). As pointed out by ANDERSON et al. (1974), BURTON et al. (1978), and WOODS and HEAMAN (1989), different genomes and different parents and grandparents may influence the relation between F and inbreeding depression. Results in our test also showed variation among seed parents, even parents in the same full-sib orchard families, in response to inbreeding (Tables 3 and 4, interaction terms).

Potential Effect of Sib Mating on Plant Vigor

In general, the response in Douglas-fir seemed to be predominantly linear with possibly some deviation from linearity at $F = 0.125$. In the following discussion, we use a mixed-mating model and project the consequences of a linear relation between fertility or growth depression and F . The effects will be less than indicated for half-sib mating if the relation is nonlinear. Estimates of inbreeding depression in height growth were obtained from the literature.

For relative self fertility, there is an inbreeding depression of 0.18 for each increase in F of 0.1 between $F = 0$ and $F = 0.5$. For vigor, we assume an inbreeding depression in height of 6% for each 0.1 increase in F . This value comes from comparisons of outcross- and self-pollination Douglas-fir progenies, where the inbreeding depression in 10-year height was 29% at $F = 0.5$ (SORENSEN and MILES, 1982), and from 5-year data of WOODS and STOEHR (1993), who report a linear 5.8% decrease in height per 0.1 increase in F in 2 field plantings on Vancouver Island.

Figures 1 and 2 illustrate depressions in relative fertility and height for populations produced from varying mixtures of outcross and inbred pollen with 0% to 50% inbred pollen illustrated in the figures. The comparisons show that partial selfing is more detrimental to seed production than is partial sibbing (Figure 1). Partial sibbing, particularly full sibbing, on the other hand, is more detrimental to height growth than selfing with up to 50% inbred pollen (Figure 2), because inbreeding depression in growth of the progeny is a product both of survival of the inbred plants and of their depression in vigour. Even though the selfs have by far the greatest depression in vigour, very few of the self-fertilized embryos survive in Douglas-fir. Sibbing is still more detrimental if wood volume is considered. Inbreeding probably depresses diameter increment more than height increment (SAITÔ et al., 1986; BUSH and SMOUSE, 1991), but even if both are depressed to the same extent, inbreeding depression in volume will be about threefold that illustrated for heights (Figure 2). Additionally, individual plants will be less depressed in vigor if derived from sibbing than if derived from selfing and, consequently, will be more difficult to cull in the nursery and will be more persistent in occupying space in the plantation.

Further field testing of inbreds may change absolute values for inbreeding depression. For example, LAYTON and GODDARD (1983) reported a 7% decrease in 3-year height per 0.1 increase in F with depression greater between $F =$

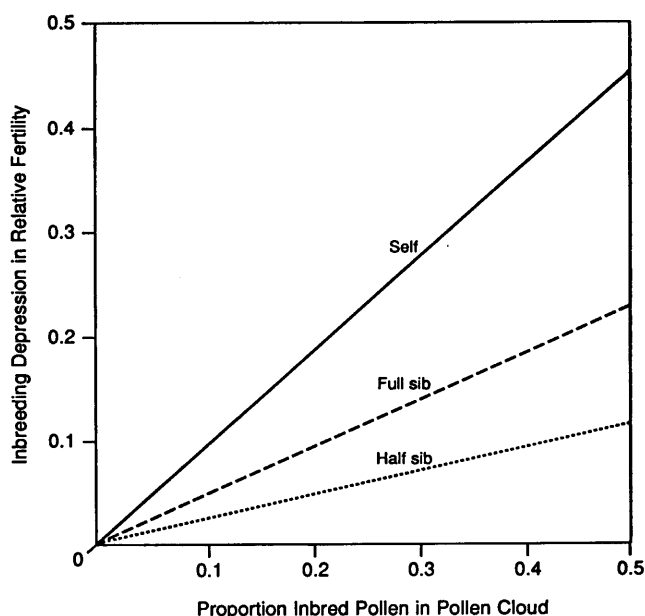


Figure 1. — Relation between proportion of inbred pollen (self, full sib, and half sib) in the pollen mix (abscissa) and depression in filled-seed set relative to crossing with unrelated pollen assuming 1 embryo per ovule. Lines are based on a decrease in relative seed set of 0.18 for each increase in F of 0.1 (Table 5, bottom, relative fertility). Example: if the pollen cloud contains 0.10 or 10% self pollen, seed set is reduced by 0.09 or 9% relative to outcross seed set.

0 and 0.125 than between $F = 0.125$ and 0.25. SQUILLACE and KRAUS (1962) reported a decrease in relative 1-year height of *Pinus elliotti* seedlings of 4% for an increase in F of 0.1, which BURROWS and ASKEW (1982) thought might be too severe. Results reported by LANGNER (1966) for 2-

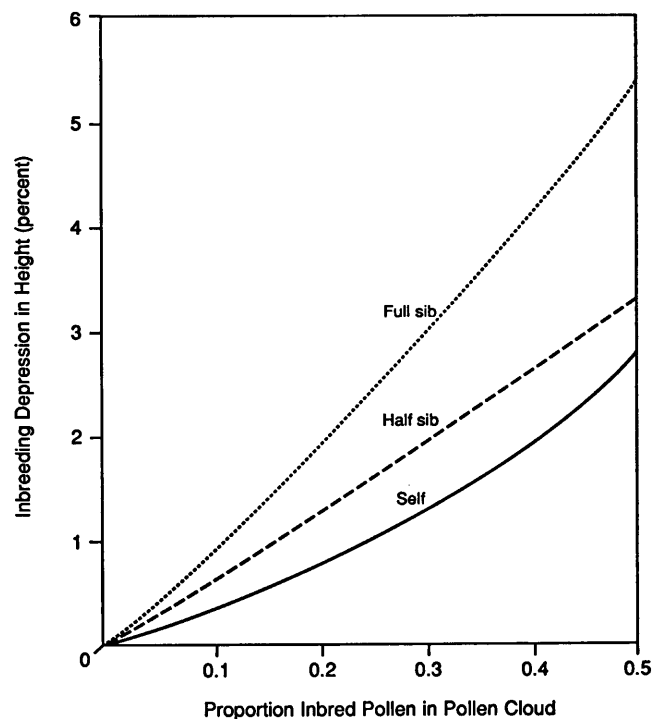


Figure 2. — Relation between proportion of inbred pollen (self, full sib, and half sib) in the pollen mix (abscissa) and depression in mean height of the progeny compared with outcross progeny. Lines represent the population mean height and, for the inbred portion of the population, are based on a decrease in relative height of 6% for each increase in F of 0.1.

year height in *Larix leptolepis* were curvilinear and indicated an 8% decrease in relative height per 0.1 increase in F between $F = 0$ and $F = 0.25$, but a 5% decrease between $F = 0.50$ and $F = 0.625$. Nevertheless, relative positions of the lines in figures 1 and 2 probably will not change greatly unless epistasis or linkage effects become larger than they now appear to be for Douglas-fir.

Polyembryony (SORENSEN, 1982) may change the position of the lines somewhat. With open or mixed mating and more than one fertilization per ovule, outcross and related embryos will occur in the same ovule. In Douglas-fir, most self embryos abort, which favors the frequency of outcrossing (see Figures 1 and 2 in SORENSEN, 1982). Half- and full-sib embryos are less likely to abort, and outcrossing will be less favored when the polyembryony includes outcross and sib embryos than when it includes outcross and self embryos. Consequently, to the extent polyembryony is a factor, the lines in figure 1 (this paper) should be closer together and the lines in figure 2 more fanned out than they are portrayed.

Environmental Influence on Self Fertility

Self pollination of seed orchard trees gave a very low proportion of filled seeds per cone (mean filled seed percent, or FSP = 1.8, Table 5). This is considerably lower than filled seed percents that have resulted from self pollination of naturally occurring trees in the woods (FSP = 9.5, ORR-EWING, 1954, 1957; 8.5, PIESCH and STETTLER, 1971; 7.9, SORENSEN, 1971). WOODS and HEAMAN (1989) gave their results in filled seeds per cone (1.2), which would probably equate to FSP < 3.0. In other words, self fertility seems to be lower in our test and in that of WOODS and HEAMAN (1989) than in tests using older trees growing in their native stands. The difference may be associated with juvenility or with the growth of the orchard trees on sites where they are not native. Other reports (PARSONS, 1971; KOMAKI, 1982) have indicated that harsh environments may increase the expression of genetic load, and there is evidence that in the legume *Cassia fasciculata* resource availability can influence the abortion of self embryos (MARTIN and LEE, 1993). The St. Paul Orchard is located in the Willamette Valley, a xeric location compared with the stands of the parent trees. WOODS and HEAMAN (1989) did their crossing at the Cowichan Lake Research Station on southern Vancouver Island, British Columbia, Canada. Although the Station is well within the forest zone, it is dry compared to the natural stands of the founder clones. Also root pruning and stem injection of gibberellic acid 4/7 were used to induce development of reproductive buds on many trees. Given the weakness of self embryos, these altered growing conditions may lead to increased environmental (PARSONS, 1971; MARTIN and LEE, 1993) or maternal lethal effects, perhaps related to those mentioned by BISHIR and NAMKOONG (1987).

Mixed-Mating Model

Recessive embryonic lethal alleles and embryo abortion, because they eliminate a large proportion of selfed progeny, influence the estimate of selfing rates in mixed-mating models (KÄRKKÄINEN and SAVOLAINEN, 1993; SORENSEN and ADAMS, 1993). If production of self seeds in some orchards or clone banks is indeed low due to these environmental causes of self-embryo abortion, it would mean that isozyme estimates of "selfing" in orchards reflect higher proportions of self pollination than would the same estimates of selfing in natural stands. For example, assuming one

embryo per ovule and relative self fertility (RSF) of 0.113 (SORENSEN, 1971), the published values of natural-stand selfing of 0.07 (SORENSEN, 1973) to 0.10 (SHAW and ALLARD, 1982) would result from 40% to 50% self pollination (SORENSEN and ADAMS, 1993). On the other hand, based on estimated orchard values of RSF (Table 5, this paper; RSF = 0.033 and 0.038, mean 0.0355), 40% to 50% self pollination would give natural selfing estimates of 0.02 to 0.04. Therefore, lower selfing or higher outcrossing estimates in the orchard compared with natural stands would not necessarily reflect differences in pollen dispersal. Likewise, similar estimates of outcrossing would not necessarily mean similarity with regard to pollen dispersal.

Practical Consideration

The important observation in this test and in that of WOODS and HEAMAN (1989) with regard to seed orchard management is that sib mating in Douglas-fir gives rather high seed yields. When that is combined with the fact that seedlings from sib mating will be difficult to cull before outplanting, the potential for vigor reduction becomes important (Figure 2). Sib mating within an orchard also may be enhanced by within-family similarity in floral phenology (ERICKSON and ADAMS, 1989). The main caution for management is to keep related individuals maximally spaced within the orchard (ERICKSON and ADAMS, 1989).

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Genetics of Second Flushing in a French Plantation of Coastal Douglas-Fir¹⁾

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Summary

The genetic control of second flushing and its relationships with stem height and forking defects were investigated in a coastal Douglas-fir (*Pseudotsuga menziesii*

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