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Growth and Cold Hardiness of Intervarietal Hybrids of Douglas-fir

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Summary. Potentials for improvement of the interior variety of Douglas-fir (*Pseudotsuga menziesii* var. 'glauca') by hybridization with the coastal variety (*P.m.* var. 'menziesii') were explored. The primary objective was to assess possibilities for increasing growth potentials of the interior variety while maintaining adaptation to relatively cold inland environments. Seventy full-sib hybrid families and their half-sib parental lines were grown in two contrasting environments common to the interior variety. Nine traits related to growth, phenology, and freezing tolerance in 4-year-old trees were compared.

For traits related to growth (height and diameter) in the inland environment, hybrids equaled the growth of the coastal variety and exceeded the interior variety by 40 percent. For traits related to adaptation (bud burst, bud set, frost damage, tree form, and freezing tolerance), hybrids were intermediate but approached levels characteristic of the interior variety. Survival of hybrids equaled that of the interior variety and was superior to that of the coastal variety.

Hybrid characters could not be predicted reliably from those of parental lines. Yet, quantitative genetic analyses suggest that expression of characters related to growth depends on nonadditive genetic effects, but expression of those related to adaptation is somewhat dependent on additive effects.

Realization of the tremendous potential of hybridization for improvement of the interior variety will require at least one backcross generation or additional crosses utilizing introgressed populations.

Key words: *Pseudotsuga menziesii* - Douglas-fir - Hybridization - Cold Hardiness - Physiologic Adaption - Growth Pattern

Introduction

Intervarietal hybridization of Douglas-fir (*Pseudotsuga menziesii*) provides a means for incorporating the growth potential of the coastal variety (*P.m.* var. 'menziesii') into the germ plasm of the interior variety (*P.m.* var. 'glauca'). Adaptation to a maritime climate (Fig. 1) has endowed the coastal variety with growth rates superior to the interior variety. Yet, superior growth potentials are realized only in mild climates (Haddock et al. 1967; Sorensen 1967). Whereas the interior variety is adapted to the cold winters of the Rocky Mountain and Intermountain regions, the coastal variety sustains high mortality and frost damage in continental climates of the United States (Wright et al. 1971; Gerhold 1965; Baldwin and Murphy 1956) and Germany (Stern 1974). In relatively cold inland environments, hybridization may effectively increase the growth potential of the interior variety while maintaining cold hardiness.

Although the concept of hybridization for improvement of Douglas-fir was suggested long ago (Duffield 1950), current effort emphasizes improvement of the coastal variety. In the maritime environment of British Columbia, growth of intervarietal hybrids is inferior to that of coastal parental lines (Orr-Ewing 1966; Orr-Ewing et al. 1972). Yet, these same hybrids may be considerably more valuable to inland environments than to maritime environments (Orr-Ewing et al. 1972).

The objective of the present study is to explore the potential of intervarietal hybridization for improving Douglas-fir in the Northern Rocky Mountains. Primary emphasis is given to growth potential, physilogic and phenologic adaptation to the growing season, and cold hardiness. However, since hybrids must withstand the interrelated complex of environmental factors that characterize the interior, the numerous additional traits that differentiate the varieties cannot be discounted. In addition to chemical differences associated with monoterpenes (von Rudloff 1973), the varieites differ in ability to withstand drought (Ferrell and Woodard 1966; Pharis and Ferrell 1966), in patterns of growth (Irgens-Moller 1968), and in tolerance to *Rhabdocline pseudotsugae* (Stephan 1973). Therefore, the present report of the growth and the cold hardiness of 4-year-old hybrid families and their parental lines will be supplemented by field tests.

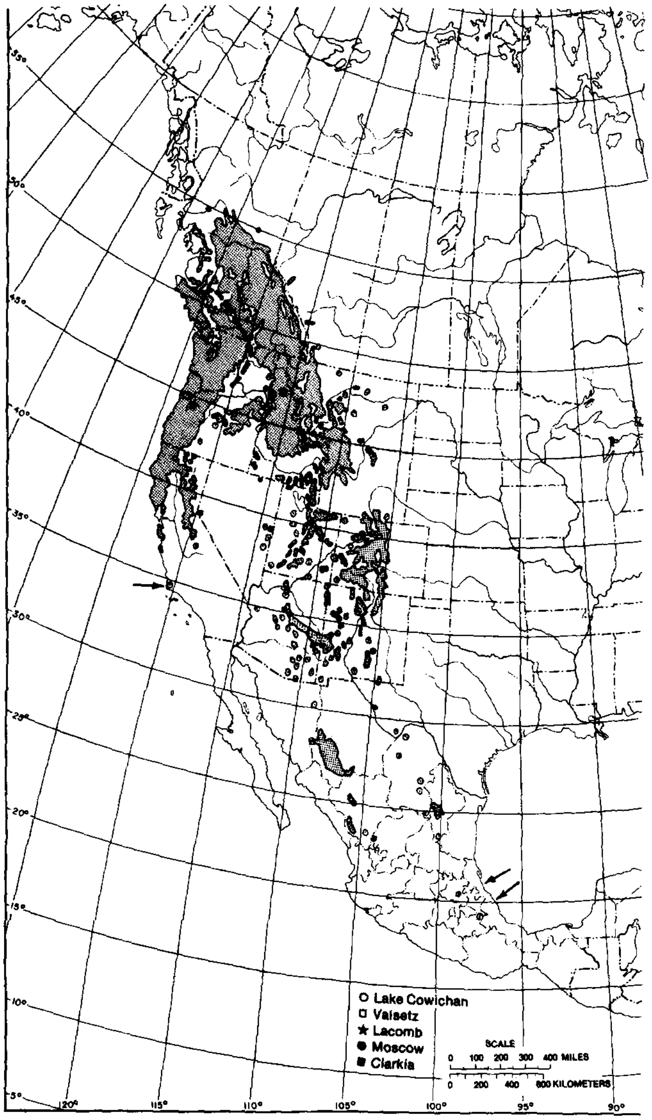

Fig.1. Geographic distribution of Douglas-fir *(Pseudotsuga menziesii)* and location of provenances from which hybrids were developed. The broken line approximates separation of the two varieties *P.m.* var. 'menziesii' and *P.m.* var. 'glauca' (from Little 1971)

Materials and Methods

In addition to 70 hybrid families produced from controlled pollinations (Table 1), wind-pollinated cones were obtained from the 15 trees used as paternal parents and the 18 trees used as maternal parents. Thus, tests included 70 full-sib hybrid families and 33 half-sib families that represented parental lines.

Seeds were sown in October 1971 in unreplicated nursery beds at the Priest River Experimental Forest Nursery. During the first year of growth, the following observations were made: (1) date on which 90 percent of the seedlings emerged, (2) date on which 75 percent of the trees set bud, (3) occurrence of chlorophyll-deficient and albino seedlings, (4) height of 50 seedlings of each family, (5) percent survival after the first winter.

Nursery trials were established with 1-year-old trees at the Coeur d'Alene (Cd'A) and Priest River Experimental Forest (PREF) Nurseries. For all families, five seedlings were transplanted at a spacing of 30 cm into row plots in each of two replications at both nurseries. When seedlings were 3 years old, height was measured and damage from an early fall frost was scored. During the fourth season of growth, rate of bud burst, date of bud set, and tree form were scored for each tree. Tree height, diameter at the ground, and leaf length also were measured. Frost damage to each tree was scored on 10 October by means of a scale from 1 to 3:#1 = no damage, 2 = injury to leaves, 3 = damage to leaves and wood. Bud burst was scored 19 May at Cd'A and 2 June at PREF according to a scale of 1 to 6:#1 = bud scales closed with no green leaves visible, 6 = elongation to such an extent that new shoots drooped pronouncedly. The date by which all buds had set on each tree was scored at weekly intervals. Form was scored on a scale of 1 to 11: 1 = pronounced dominance of a single leader, 7 = a major fork resulting in codominance of two leaders, and 11 = no dominant leader.

Where necessary, appropriate transformations were made for normalizing frequency distributions (Steel and Torrie 1960), and the following statistical analyses were made on data from nursery trials: (1) analyses of variance for assessing the magnitude of differences among and within parental varieties and their hybrids, (2) quantitative genetic analyses for hybrid families, and (3) regression analyses for predicting the performance of hybrid families from that of their parental lines.

After data had been collected from the nursery trials, artificial freezing tests were made for assessing differential tolerance to cold of hybrid families and their parental lines. For both replications at each nursery, lignified twigs from the five trees representing a single family were cut, moistened, packaged together in polyethylene bags, and stored in a cooler. This procedure was followed biweekly from late September to late November. Twigs were transported to the Moscow laboratory where they were stored at 3°C. Except for the samples collected in late November, all samples were frozen at a cooling rate of 5°C/h to predetermined temperatures within 36 hours from collection. As outlined by Sakai and Weiser (1973), an attempt was made to induce maximum hardiness into twigs collected in late November. Since the samples had been collected at temperatures below 0°C, twigs were stored at -7°C for 1 week and at -10°C for 3 days. Thereafter, they were cooled at a rate of 5°C/d to predetermined temperatures.

Twigs were removed from the freezer at the desired temperature, thawed at 5°C for at least 24 hours, and placed in water in a greenhouse. After 1 week, damage from freezing was scored on the basis of browning of leaves. The number of twigs damaged for each family was recorded according to each replication at both nurseries. Regression analyses were used to assess differential ability of families to tolerate freezing.

Results

General Observations

As anticipated from the results of Allen (1960, 1961), 90 percent of the seedlings representing the interior

Table 1. Mating design for production of 70 full-sib hybrid families. Family codes included the hybrids, 15 paternal half-sib families, and 18 maternal half-sib families. Hybrid codes are derived from the combination of codes that key their respective parental lines

Maternal provenances, interior variety	Tree number		Paternal provenance - coastal variety														
			Lacomb, Oregon					Valsetz, Oregon					Lake Cowichan, B.C.				
			8	10	11	35	100	100	101	102	103	104	98	99	100	106	107
		Family Code	A	B	C	D	E	F	G	H	I	J	U	V	W	X	Y
Moscow, Idaho	2	2	2A	2B	2C	2D	2E										
	3	3	3A	3B	3C	3D	3E										
	4	4	4A	4B	4C	4D	4E										
	5	5						5F	5G	5H	5I	5J					
	7	7						7F	7G	7H	7I	7J					
	8	8						8F	8G	8H	8I	8J					
	9	9	9A	9B	9C	9D	9E										
	10	10	10A	10B	10C	10D	10E										
Clarkia, Idaho	7	11	11A					11F					11U				
	9	12		12B					12G					12V			
	16	13			13C					13H					13W		
	17	14				14D					14I					14X	
	18	15					15E					15J					15Y
	19	16	16A					16F					16U				
	22	17		17B					17G					17V			
	25	18			18C					18H					18W		
	26	19				19D					19I					19X	
	27	20					20E					20J					20Y

variety had emerged by 17 May and seedlings representing the coastal variety emerged 2 weeks later. The rate of emergence of hybrid seedlings was the same as that of the interior variety. No mutant seedlings occurred in hybrid families. Chlorophyll-deficient or albino seedlings were present in three families of the interior variety and in two families of the coastal variety. Seventy-five percent of the seedlings from interior, hybrid, and coastal families had set buds by 3 August, 28 August, and 26 September, respectively. Whereas trees of the interior variety averaged 5 cm in height, hybrids and the coastal variety averaged 7.5 cm.

High rates of mortality occurred during the winter of 1972. Temperatures reached -26°C when there was little or no snow cover. Whereas 44 percent of the trees of hybrid and interior origin survived, only 9 percent of those of coastal origin survived. Of the last group, survival was only 5 percent for trees from Oregon, but was 20 percent for those from British Columbia.

Unfortunately, it is not possible to separate the effects of climate from those of transplanting on seedling survival. Many trees of coastal origin had set buds only 1 week before nursery tests were established. Little additional mortality occurred during the next 3 years, although it was not until the winter of 1974 that some trees were taller than the maximum snow depth. Yet, these general observations suggest that the capability of hybrid families to survive in the inland climate approaches that of the interior variety. As a group, hybrids were intermediate between parental varieties in some traits, similar to the interior variety in others, and approached the coastal variety in still other traits. A lack of chlorophyll-deficient and albino mutants in hybrid families implies unique allelic or genic contributions to the genetic load of parental varieties.

Nursery Studies

Except for families from Oregon, most seedlings in the nursery tests that died during the first winter were replaced in the spring of 1973. After transplanting, five seedlings represented most families in each replication at both nurseries, but the number of seedlings representing each family from Oregon averaged less than one in each replication. Because seedlings used for replacement survived the first winter, they may represent the most hardy individuals from each family. Thus, particularly for coastal origins, germ plasm represented in the nurseries may be skewed toward maximal hardiness. Possible effects of a skewed distribution on interpretation of statistical analyses will be assessed subsequently.

Analyses of variance revealed that the large mean differences for all variables except bud burst at Cd'A (Table 1) were associated with significant (1 % level) main effects for varieties and nurseries. Analyses also revealed substantial variation among families within parental varieties and among hybrid families for most variables. However, except for tree form and rate of bud burst, intraclass correlations for varieties were 2 to 5 times greater than those associated with families within varieties. Significant interactions between nurseries and varieties were detected for only two variables. Frost damage to hybrids at PREF was intermediate between that of the two varieties. At Cd'A, where damage was much less severe than at PREF, hybrids suffered the same level of damage as the interior variety. Bud burst at Cd'A occurred at approximately the same time for all varieties. At PREF bud burst for the coastal variety was delayed. High intraclass correlations were associated with variance within families. Depending on the variable, these effects accounted for 40 to 75 percent of the variance.

Mean differences for traits related to growth (height and diameter) indicate that traits of the hybrids and the coastal variety exceed the interior variety by about 40 percent (Table 2). In fact, over 70 percent of the hybrid families were of significantly (LSD 0.05) greater height and diameter than the family representing its maternal parent and about 25 percent were also larger than the family representing its paternal parent. For traits related to adaptation to the inland climate (bud burst, bud set, frost damage, and form), hybrids were generally intermediate between their parental representatives (Table 2), but comparisons of means (LSD 0.05) showed that a smaller proportion of hybrid families deviated significantly from their maternal line than from their paternal.

Similarities in results for several variables are elucidated by the correlation matrix (Table 3). First, trees that set buds late tended to suffer frost damage; high levels of frost damage were associated with a poor form (high score); trees of poor form had a low growth rate (negative deviation from the regression line); and low growth rate characterized short trees. Second, tall trees tended to have large diameters, long leaves, and a high growth rate. Yet, these coefficients fail to represent character association within the divergent varieties. Underlined coefficients (Table 3) reflect associations that differed significantly (5 % level)

Table 2. Mean values for eight variables according to nurseries and varieties

	4-year height (cm)	Diameter (mm)	Bud set[1] (weeks)	Form (classes)	Leaf length (mm)	Height deviation[2] (cm)	Frost damage (classes)			Bud burst (classes)	
							PREF[3]	Cd'A[4]	Both nurseries	PREF	Cd'A
Nurseries											
PREF	50.2	12.3	3.8	5.9	31.8	-1.11	1.62			4.00	
Cd'A	57.2	15.9	2.6	3.9	30.4	+1.11		1.04			3.78
Varieties											
Interior	33.9	10.0	2.1	4.2	30.3	-1.05	1.08	1.00	1.04	4.17	3.67
Hybrid	54.1	14.0	3.1	4.4	31.5	+1.02	1.63	1.04	1.33	4.02	3.69
Coastal	53.2	13.6	4.3	6.2	29.2	-1.04	2.23	1.33	1.72	3.56	3.90

[1] Weeks after August 1

[2] Deviation from regression of 4-year height on 3-year height

[3] Priest River Experimental Forest Nursery

[4] Coeur d'Alene Nursery

Table 3. Simple correlation coefficients among eight variables. Correlations were based on data from individual seedlings from all families, nurseries, and replications. Significance at the 1 percent level of probability is indicated by $r > 0.08$ and $r < -0.08$. Underlined coefficients reflect associations that differed significantly in parental varieties

Variable	4-year height	Frost damage	Bud burst	Bud set	Form	Diameter	Height deviation[1]
Frost damage	$\underline{-0.05}$						
Bud burst	$\underline{-0.04}$	$\underline{0.05}$					
Bud set	$\underline{0}$	$\underline{0.38}$	$\underline{0.03}$				
Form	-0.12	$\underline{0.29}$	$\underline{0.08}$	0.30			
Diameter	0.87	$\underline{-0.14}$	$\underline{-0.11}$	$\underline{-0.08}$	$\underline{-0.01}$		
Height Deviation[1]	0.51	$\underline{-0.37}$	$\underline{-0.01}$	$\underline{-0.08}$	-0.36	$\underline{0.39}$	
Leaf length	0.33	0.11	0.01	$\underline{0.10}$	$\underline{0.07}$	0.31	$\underline{0.06}$

[1] Deviation from regression of 4-year height on 3-year height

for parental varieties. All of the indicated associations pertain to either direct or indirect relationships with bud burst and bud set. Thus, different matrices primarily reflect the degree that phenological events of the coastal variety were out-of-phase with the inland climate.

Results of these analyses emphasize genetic divergence of the two varieties along with subordinate genetic variation within each variety. Yet, parental varieties are sufficiently related that inviabilities, incompatibilities, or negative heterosis were not evidenced by the performance of hybrid families. In the inland climate, hybrids expressed growth potentials similar to the coastal varietiy and adaptational features that approached those of the interior variety. A high degree of variation among and within hybrid families approbates quantitative analyses for elucidating genetic variances.

Even though theoretical concepts in quantitative genetics for hybrid populations are rather limited, quantitative methods were used for data from the 70 full-sib hybrid families. Results of these analyses should provide only a general assessment of gene effects, genetic variances, and selection procedures for the hybrid families.

Although results of quantitative analyses (Table 4) are presented as if the mating design had been hierarchial, the most appropriate design involved seven sets of factorial matings (Table 1). Statistical analyses and their inferences were made according to both designs, but the hierarchial model is presented for simplicity.

Contrasting genetic effects differentiated the various traits. Genetic effects accounted for little variance in tree form, height deviation, or frost damage at Cd'A. Genetic variances for height and diameter were associated primarily with interactions between males and females. Whereas small but significant main effects of females were indicated by leaf length, genetic variance in bud set was determined primarily by the main effects of males. That for frost damage at PREF and bud burst was divided between main effects of males and females.

Additional analyses were made according to the following model for assessing the relative importance of maternal line, paternal line, and nursery environment in determining hybrid performance:

$$Y_{i♀♂} = a + bX_{i♀} + bX_{i♂} + bX_n$$

where:

$Y_{i♀♂}$ = mean value of hybrid of maternal family ♀ and paternal family ♂ growing in nursery i.

$X_{i♀}$ = mean value of the half-sib family representing maternal tree ♀ in nursery i.

$X_{i♂}$ = mean value of the paternal half-sib family ♂ growing in nursery i.

X_n = dummy variable for nurseries.

According to this model, results of analyses are subject to bias introduced by the confounded mating design. To alleviate effects of confounding, the relative importance of independent variables in determining the dependent variable was assessed by means of standardized partial regression coefficients for

Table 4. Intraclass correlations derived from analyses of variance for examining perfor-

Source of variance	df	4-year height	Diameter	Bud set	Form
Nurseries	1	0.06**	0.20**	0.17**	0.11**
Replication	1	0.03**	0.01*	0.01**	0
Families	69	0.17**	0.16**	0.08**	0.04**
Females	17	0.01	0.01	0.01	0.0
Males/females	52	0.16**	0.15**	0.07**	0.04**
Males	14	0	0.01	0.03**	0.02**
Females/males	55	0.17**	0.15**	0.05**	0.02**
N × families	69	0	0	0.02	0.04*
N × females	17	0	0	0	0.03**
N × males/females	52	0	0	0.02	0.01
N × males	14	0	0	0	0
N × females/males	55	0	0	0.02	0.04*
Experimental error[2]	139	0.18**	0.09**	0.10**	0.05**
Within cells	1,042	0.56	0.54	0.62	0.75

[1] Deviation from regression of 4-year height on 3-year height
[2] Contains sources of variation from the three interactions involving replication
* Significance of F value at 0.05 level of probability
**Significance of F value at 0.01 level of probability

which effects of intercorrelation among independent variables are eliminated.

Results of regression analyses (Table 5) depict an expected predominance of nursery effects in determining hybrid performance for most traits. Also apparent for all traits, except frost damage and bud burst, is character expression in hybrids that is independent of that of their parental lines. Although these results are somewhat contradictory to the foregoing, little influence of parental lines could be expected in traits for which little genetic variance or nonadditive effects were pronounced in preceding analyses. Failure of regression analyses to corroborate paternal effects indicated by previous analyses may reflect either reduced penetrance of the coastal germ plasm in the inland environment or bias introduced from small samples of several paternal lines. Yet, both analyses support maternal effects for frost damage and bud burst. Regardless, except for bud burst, parental lines account for little of the variance determined by the regression models.

Quantitative analyses show substantial levels of genetic variation among hybrid families. Growth of hybrid families seems to be dependent on nonadditive genetic variances. Adaptational features seem to be determined by relatively weak additive effects. Except for bud burst, the performance of maternal and paternal half-sib lines poorly reflects the performance of hybrid families.

Table 5. Coefficients of determination and standardized partial regression coefficients derived from multiple regressions of eight variables for prediction of hybrid mean values from those of maternal parental lines, paternal parental line, and nursery environments

Variable	R^2	$b'_{\text{♀}}$	$b'_{\text{♂}}$	b'_N
4-year height	0.09*	0.08	-0.11	-0.33*
Diameter	0.28**	0.11	-0.05	-0.49**
Bud set	0.33**	0.18	0.18	0.33**
Form	0.31**	-0.15	-0.06	0.69*
Leaf length	0.14**	0.23	0.20	0.24**
Height deviation	0.63**	-0.01	-0.01	-0.81**
Frost damage	0.75**	0.25*	-0.12	0.80**
Bud burst	0.37**	0.54**	0.04	0.15

** Significance at the 1 percent level of probability
* Significance at the 5 percent level of probability

mance of hybrid families for eight variables. A model of random effects was assumed

		Frost damage			Bud burst	
Leaf length	Height deviation[1]	PREF	Cd'A	Both nurseries	PREF	Cd'A
0.04**	0.40**			0.46**		
0	0	0	0	0	0.13**	0.05**
0.12**	0.03**	0.29**	0.03	0.05**	0.21**	0.44**
0.04**	0.0	0.07*	0.0	0.02**	0.09**	0.28**
0.08**	0.03*	0.22**	0.03	0.03**	0.12**	0.16**
0.01	0	0.07*	0	0.01	0.06*	0.25**
0.11**	0.03*	0.22**	0.03	0.04**	0.15**	0.19**
0	0			0.04**		
0	0			0		
0	0			0.04**		
0	0			0.03**		
0	0			0.01		
0.08**	0.05**	0.05**	0.01	0.03*	0.10**	0.07*
0.75	0.52	0.67	0.95	0.42	0.57	0.44

Freezing Tests

Results of artificial freezing tests depict expected patterns for the development of cold hardiness (Table 6). The development of low levels of cold tolerance is triggered by frosts and develops at a rate responsive to minimum temperatures (Levitt 1972). Consistent with results of Timmis and Worrall (1975) for the coastal variety, lignified twigs collected prior to frost in late September tolerated temperatures of -10°C but were injured at -15°C. Even though minimum temperatures were consistently lower at PREF than at Cd'A, only

Table 6. Percentage of twigs damaged in freezing treatments according to nurseries and varieties

	24 & 25 September			15 & 16 October			4 & 5 November			20 & 26 November			
	°C			°C			°C			°C			
	-2	-10	-15	-15	-20	-25	-15	-22	-30	-20	-25	-30	-35
Nurseries													
PREF	0	0	87	25	90	98	1	28	93	0	3	21	48
Cd'A	0	0	91	74	96	99	1	45	96	0	4	35	70
Varieties at PREF													
Interior variety	0	0	56	8	72	90	0	14	82	0	1	5	25
Hybrids	0	0	72	25	94	99	1	28	95	0	2	23	51
Coastal variety	0	0	100	75	98	100	8	79	100	4	24	52	89
Varieties at Cd'A													
Interior variety	0	0	76	51	87	96	0	18	83	0	1	11	35
Hybrids	0	0	70	78	97	100	1	48	99	0	4	37	78
Coastal variety	0	0	98	89	100	100	4	89	98	4	16	75	96

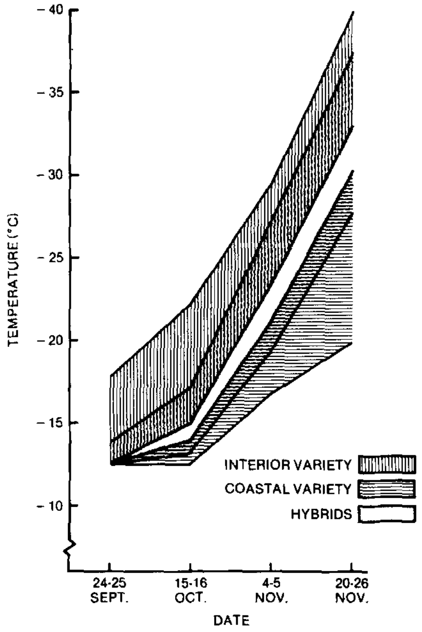

Fig.2. Range in average temperatures for which 50 percent of the twigs were damaged at four sampling dates for the coastal variety, interior variety, and hybrids

slight differences in seedling hardiness during late September could be associated with nursery environments. As autumn progressed, hardiness developed at a faster rate for trees at the colder PREF environment than at Cd'A.

Attempts to induce maximum levels of hardiness evidently failed. Consistent differences associated with nursery environments were found in samples collected in late November. Therefore, despite the cold and lengthy storage, data obtained from twigs collected on 26 November probably reflect hardiness for late November. Regardless, the correspondence between observed damage for this date (Table 6) and previous data (Sakai and Weiser 1973) suggests that trees of both varieties were near maximum levels of hardiness.

For each sampling date, seedlings of the coastal variety sustained more injury than those of the interior. The number of injuries to hybrids was intermediate between the two (Table 6). For assessment of the differential rates that hardiness developed in various families, the average temperature resulting in injury to 50 percent of the twigs for each family at each sampling date was estimated by interplation and, in some cases, by extrapolation. Uniform levels of hardiness characterized the coastal variety in late September (Fig.2). Although variation in hardiness characterized the interior variety at each date, variation for the coastal variety became pronounced as hardening advanced. Haridness developed at a faster rate and to lower levels in trees of the interior variety than in those of the coastal variety. Although intermediate, hardiness of hybrid families was more similar to that of the interior variety than the coastal. In fact, the rate that hardiness developed in 17 hybrid families exceeded that of the family of interior origin characterized by the lowest rate. Hardiness of only four hybrid families developed at a slower rate than that of the coastal family of fastest rate.

Genetic relationships between the hardiness of hybrid families at a particular date and that of their parental lines were examined according to the model:

$$Y_{♀♂} = a + bX_{♀} + bX_{♂}$$

where:

$Y_{♀♂}$ = temperature associated with injury to 50 percent of the twigs for the hybrid family developed from maternal tree ♀ and paternal tree ♂.

$X_{♀}$ = temperature value for the half-sib family representing maternal tree ♀.

$X_{♂}$ = temperature value for the half-sib family representing paternal tree ♂.

Nursery effects have been omitted from the model because excessive extrapolation was necessary for inclusion of their effects. Comparisons of the effects of independent variables on the dependent were made according to standardized partial regression coefficients.

As was expected from Fig.2, hardiness of a hybrid family in late September was related to that of neither parental line (Table 7). However, as was indicated by standardized partial regression coefficients, the hardiness of hybrids in mid-October was determined by equal contributions from the male and female lines. In November the effect of the maternal line was much more pronounced than that of the paternal line. Regardless, only one model accounted for as much as 30 percent of the variation. Consistent with results of nursery studies, development of hardiness in hybrid families also is predicted poorly by that of parental lines.

Table 7. Coefficients of determination and standardized partial regression coefficients derived from regressions of the temperature associated with 50 percent damage to hybrid twigs as predicted from that of their maternal and paternal lines

	R^2	$b'_♀$	$b'_♂$	$b'_♀ - b'_♂$
24 & 25 September	0.03	0.16	-0.03	ns
15 & 16 October	0.22**	0.23*	0.37**	ns
4 & 5 November	0.32**	0.56**	-0.01	*
20 & 26 November	0.08*	0.24	0.09	ns

** Significance at the 1 percent level of probability
* Significance at the 5 percent level of probability

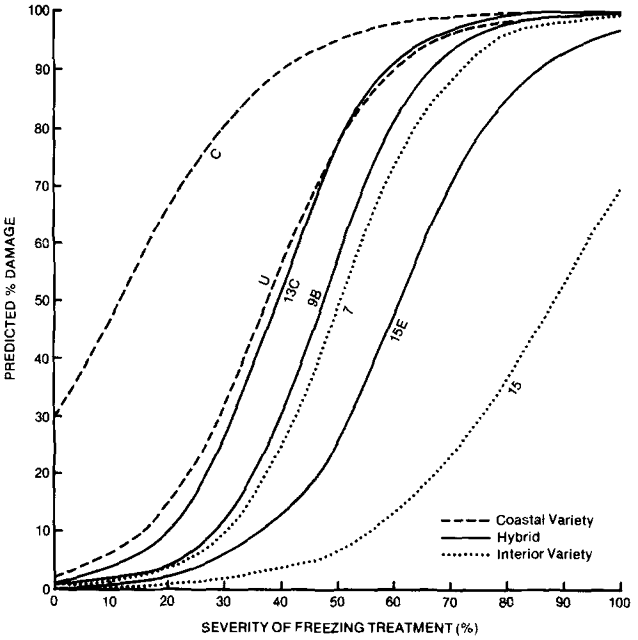

Fig.3. Models of freezing damage according to treatment severity for families representing the range in response for the coastal variety (C and U), interior variety (7 and 15), and hybrids (13C and 15E)

To provide a further assessment of differential abilities of families to tolerate freezing of various severities, a linear regression analysis was made for each family according to the transformed logistic model:

$$Y_{ijk} = \frac{1}{1 + be^{-rX_k}},$$

which is represented by the linear model

$$\ln\left(\frac{1}{Y_{ijk}} - 1\right) = -rX_k + \ln b$$

where:

Y_{ijk} = proportion of twigs injured in sample i from family j of treatment severity k.

X_k = index of freezing severity = percentage of twigs of all families injured by freezing treatment k.

$b = \frac{1}{y_0} - 1$ where y_0 is the predicted percent damage if no freezing treatment is applied ($X_k = 0$).

r = rate of increase in damage associated with an increase in severity index.

The percentage of twigs from a single nursery that were injured by freezing to a particular temperature on a given sampling date is the index of freezing severity. The array of indices corresponds to the percent damage associated with nursery environments (Table 6). Data for -2°C were omitted because no twigs were injured.

Models for all families were statistically significant (5 % level). Although coefficients of determination ranged from 0.33 to 0.93, they averaged about 0.75. Low coefficients occurred for several families of coastal origin for which as few as 12 observations were available.

Fig.3 depicts extreme models for families of parental varieties and hybrids. Fig.4 illustrates

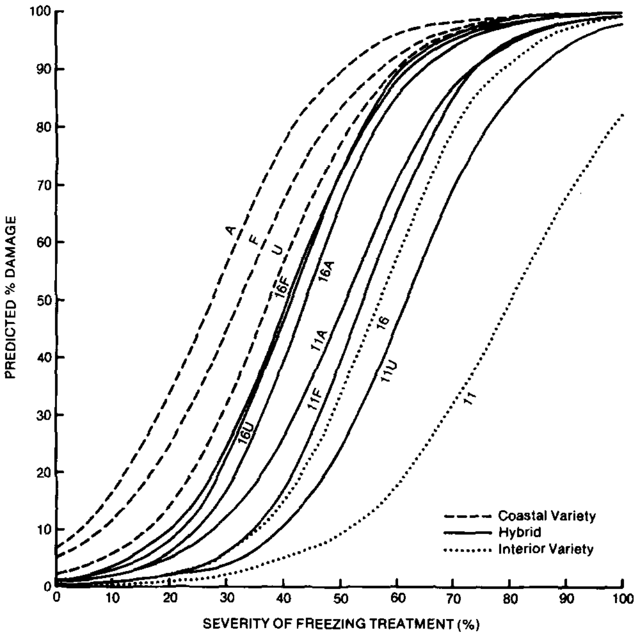

Fig.4. Models of freezing damage according to treatment severity for several parental lines and their hybrids (Families are keyed to Table 1)

genetic relationships between parental lines and their hybrids. For either value r or b in the regression equation, models for families 15 and C (Fig. 3) differed significantly (5 % level) from all other models presented. No differences were detected among models for families 15E, 7, and 9B; between models for families U and 13C; and between models for families 13C and 9B.

Results of freezing tests emphasize tremendous differences in tolerance to freezing between the varieties. More importantly, models for hybrid families were more similar to those of the interior variety than the coastal. Eighteen hybrid families showed greater tolerance to freezing than the interior family of lowest tolerance. In fact, for either value r or b, the model for family 7 (Fig. 3) differed significantly from those of only four hybrid families.

Models depicted in Figs. 3 and 4 accurately represent the pattern of all 103 models. Graphs of the various models rarely intersected. When intersection occurred, interaction was negligible, as illustrated for models of U and 13C (Fig. 3). Therefore, models that depict the relative ability of families to tolerate freezing can be represented by the predicted amount of damage at a severity of 50 percent. These values were used to assess the influence of parental lines on the frost tolerance of hybrids according to the model:

$$Y_{♀♂} = a + bX_{♀} + bX_{♂}$$

where:

$Y_{♀♂}$ = estimated damage at 50 percent freezing severity for the hybrid family developed from maternal tree ♀ and paternal tree ♂.

$X_{♀}$ = estimated damage for the half-sib family representing maternal tree ♀.

$X_{♂}$ = estimated damage for the half-sib family representing paternal tree ♂.

This regression was statistically significant (1 % level), but it accounted for only 28 percent of the variation of hybrid families. Yet standardized partial regression coefficients indicated that the interior line was of primary importance in determining tolerance in hybrids ($b'_{♀} = 0.52^{**}$, $b'_{♂} = 0.05$, $b'_{♀} - b'_{♂}{}^{**}$). Even so, freezing tolerance of parental lines poorly predicts freezing tolerance of hybrids.

Variation in the ability of families to tolerate freezing is related to variation in several characters examined in nursery trials. Significant (1 % level) correlation coefficients, based on family means, were found between estimated damage at a freezing severity of 50 percent and each of the following: 4-year height (r = 0.41); date of bud set (0.66); frost damage (0.64); and bud burst at PREF (-0.40). However, since the value of these coefficients may be determined primarily by the contrasting character associations of parental varieties, actual associations of parental characters in the hybrids may not be reflected. Both the strength and direction of these relationships were altered in correlation coefficients based only on hybrid data. Significant (5 % level) relationships were observed between estimated freezing damage and the following: 4-year height (r = 0.29); diameter (0.27); and frost damage (0.29).

Despite low values of correlation coefficients, hybrid families of greatest height and diameter also tend to have the greatest tolerance to freezing. The low value of the correlation between freezing tolerance and frost damage and lack of relationship between tolerance and date of bud set for hybrid data imply two relatively independent components to winter hardiness. As elucidated in provenance tests of the coastal variety (Campbell and Sorensen 1973), an ability to tolerate early fall frosts depends on the interval by which bud set precedes frosts. Even if bud set precedes the frost to such an extent that lignification is complete, genetic variation in tolerance to low temperatures exists among families. Frost damage in the nursery trial occurred on only partially lignified twigs. Since only fully lignified twigs were used in artificial freezing tests, the correlation between frost damage in the nursery and freezing tolerance is low. It is probable that the rate of dehardening in the spring represents an additional independent component of cold hardiness in hybrid families.

Discussion

Tests on numerous traits for 70 hybrid families and their parental lines have shown that growth potentials of hybrids were generally superior to those of the interior variety but similar to those of the coastal variety. For traits related to adaptation to the inland climate, hybrids expressed an intermediacy that was somewhat closer to the performance of the interior variety than the coastal. Inheritance of traits related to growth of hybrids seemed to depend on specific combining effects. Inheritance of traits controlling adaptation seemed to rely on relatively weak additive effects. Traits of hybrids could not be predicted from those of parental lines.

High levels of mortality during the first winter imply that genotypic distributions were truncated by selection for cold hardiness. The coastal variety would have been affected more than the hybrids or the interior variety. Although families of coastal origin would have been represented only by genotypes of maximal hardiness, families of hybrid and interior origin would have lost only genotypes of minimal hardiness. Since linkage equilibrium and independence of characters cannot be assumed, genotypic distributions truncated by selection would have had pronounced effects on interpretations of results. Provided that cold hardiness in the coastal variety is negatively associated with growth potential, growth of hybrids would have been intermediate between varieties. On the other hand, for traits related to adaptation (particularly cold hardiness), hybrids would have shown an even greater affinity to the interior variety than that expressed.

Hybrids may favor either parental line for some traits and are intermediate for others. Hybrid performance for single traits depends on specific combining effects or relatively weak additive effects. Hybrid performance is predicted poorly from performance of parental lines. These statements typify results of interracial hybridization programs for species of forest trees: *Pinus sylvestris* (Nilsson 1970, 1973); *Picea abies* (Nilsson and Andersson 1969); *Pseudotsuga menziesii* var. 'menziesii' (Orr-Ewing et al. 1972); *Pinus taeda* (Woessner 1972a, 1972b); and *Liquidambar styraciflua* (Stairs 1968). In addition, these results for hybridization of forest trees are strikingly similar to those involving interracial hybridization of herbaceous species (Hiesey 1964; Hiesey and Nobs 1970; Clausen and Hiesey 1960).

Genetic systems of wild species that are governed by complex genic interactions may be considered coherent, balanced, buffered, and resilient. Implicit in these features are the complex and unpredictable gene actions that control the characters described by Clausen (1958, 1959) for several herbaceous species. Interracial hybridization combines two differentially coadapted genetic systems. The performance of individual hybrid families depends not only on the degree to which specific parental genotypes integrate, but also on the degree to which specific gametes integrate. Thus, results of the present study show (1) a high proportion of variance within hybrid families; (2) an inability to predict hybrid performance from that of parental lines; and (3) a relatively independent expression of traits in hybrids that is not common to buffered parental lines.

Partially because of the complexities of wild genetic systems, the concept of heterosis (defined as hybrid performance superior to both parental lines) is difficult to apply to the performance of intervarietal hybrids of Douglas-fir. If the present results are tempered by possible effects of a skewed distribution of coastal genotypes, expression of single traits in hybrid families was not heterotic. Still, in a general sense, the performance of most hybrids tested in the present study was heterotic. Eighty percent of the hybrids showed growth superiority to their interior line and all hybrids expressed greater survival potential than their coastal line.

In this study the performance of the hybrid suggests tremendous potential for the improvement of the interior variety. Hybrids expressed a growth potential that was about 40 percent greater than the interior line and many hybrids showed adaptational features that approached those of the interior variety. However, suitable hybrids must equal the interior variety in adaptation to the inland climate. Of the 70 hybrid families (Table 1) tested in this study, only nine (3A, 3D, 8F, 8J, 9A, 10D, 11F, 14D, and 20Y) were (1) greater in height and diameter than their interior parental line, and (2) equal or superior to the families of interior origin that had the worst values for each of the five traits related to adaptation. No particular parental line consistently produced hybrids of adequate quality, although combinations 3, 8, A, and F accounted for six of these nine families. Finally, no family had mean values for all traits analyzed in nursery and freezing tests that were consistently among the upper 10 percent of all hybrid families. Thus, the most suitable hybrid families were those that were slightly above the average for all traits.

Tests mentioned here concerned only nine traits related to growth, phenology, and tolerance to freezing. Notably absent were tests of the ability of hybrids to survive the summer droughts that characterize the inland climate. Field tests have been established for further assessment of the adaptation and growth of these families, but it is extremely unlikely that a hybrid family or an individual tree contains all adaptational features necessary for realization of its high growth potential in the inland environment. Because of little success in predicting the performance of hybrids from that of parental lines and because of independence of adaptational traits in hybrid families, it is unlikely that suitable sources of coastal germ plasm can be found for mass production of hybrids that are of high growth potential and adequate adaptation.

The concept of coherence, developed from analyses of wild species (Clausen and Hiesey 1960), suggests that linkage groups of parental varieties will be largely maintained in gametes of F_1 hybrids; so practical application of hybridization for improvement of the interior variety may be only as distant as the first backcross generation. Yet, the time required for production of a backcross generation can be circumvented. The chemical composition of leaf oils of some populations in southern British Columbia is intermediate between the two varieties (von Rudloff 1973). Seed produced from crosses of these introgressed populations and the interior variety may represent a backcross. Regardless, the tremendous possibilities for improving the interior variety by means of intervarietal hybridization should not be dismissed.

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