Variation in Blister Rust Resistance Among 226 *Pinus monticola* and 217 *P. lambertiana* Seedling Families in the Pacific Northwest

A.J. Kegley
R.A. Sniezko

Abstract—*Pinus monticola* and *P. lambertiana* families sown in 1989 and 1994 were inoculated with *Cronartium ribicola* after two growing seasons. Development of rust symptoms and mortality were followed for five years. During this period, 96 to 99 percent of the seedlings became infected, and 91 to 99 percent of the seedlings developed stem symptoms in the four screening trials (family means varied from 30 to 100 percent). Survival of infected seedlings 5 years after inoculation varied from 1.6 to 13.1 percent for the four trials; family means varied from 0 to 54.8 percent. The frequency and level of resistance responses in half-sib progeny of phenotypic selections from natural stands were generally low, but some families had survival comparable to the full-sib checklots. In both species the families with the highest levels of survival generally had higher than average levels of several resistance responses, including percentage of seedlings without stem symptoms, delayed appearance of stem symptoms, bark reaction, and survival with stem symptoms. Levels of bark reaction were generally low in the four trials (3.5 to 12.3 percent of the trees), with half-sib families varying from 0 to 59.9 percent. Many trees with bark reaction also had normal cankers and died by the end of the assessment period. Selections from the top individuals within these trials have been grafted into seed orchards. Breeding efforts are focused on the development of seedlings with more durable rust resistance for reforestation and restoration.

Key words: blister rust, resistance responses, screening, pines

Introduction

The introduced disease white pine blister rust, caused by the pathogen *Cronartium ribicola* J.C. Fisch. in Raben., has devastated populations of five-needle pines in many parts of western North America. Development of genetic resistance to this pathogen will be a key to restoration of these species. Due to land management changes and limited opportunities for planting seedlings on federal lands, maintaining these species as viable components of the ecosystem will depend even more on the development of durable resistance.

Evaluation of white pine blister rust resistance and/or operational screening has occurred for a number of species in North America, Asia, and Europe (Kim and others 1982; Blada, this proceedings; Stephan, this proceedings; Daoust and Beaulieu, this proceedings; McDonald and others, this proceedings; King and Hunt, this proceedings; Bingham 1983; Zsuffa 1981). In North America, the identification of some genetically resistant individuals (Bingham 1983; Kinloch and others 1970; McDonald and others, this proceedings) led to the initiation of several resistance breeding programs for western white pine (*Pinus monticola* Dougl. ex D. Don.) and sugar pine (*P. lambertiana* Dougl.) (Bingham 1983; King and Hunt, this proceedings; Sniezko 1996; Kinloch and Davis 1996; Samman and Kitzmiller 1996). In Oregon and Washington, Region 6 of the USDA Forest Service, works began on operational screening and breeding of western white pine (WWP) and sugar pine (SP) for resistance to *C. ribicola* in the late 1950s, but screening of whitebark pine (*P. albicaulis* Engelmann) has only recently begun. The screening program for all three species is based at the Dorena Genetic Resource Center (Dorena); the Center staff work with geneticists and land managers throughout Oregon and Washington to develop breeding populations and seed orchards with resistant parent trees. Seedling common-garden studies (Campbell and Sugano 1987; Campbell and Sugano 1989) have provided the foundation for establishing breeding zones in Oregon and Washington for both SP and WWP. Rust resistance of progeny from parent trees in natural stands or plantations has been evaluated for most of these breeding zones; moreover, breeding among resistant progeny has begun for several of the zones.

Since the 1970s blister rust resistance has been evaluated using half-sib progeny of phenotypically selected trees from National Forest lands as well as lands of the USDI Bureau of Land Management (BLM), and other landowners in Oregon and Washington. The phenotypic selections were made on sites varying from low to high incidence of blister rust. In general the selected trees were vigorous and either free of rust cankers or showed fewer cankers than other WWP or SP in the local area. Progeny of approximately of 4,900 WWP and 4500 SP phenotypic selections have now been screened at Dorena. Based upon screening results, selections among the progeny of the selected parents have been made and placed into seed orchards.

The Region 6 program was based on protocols and resistance mechanism studies in WWP by Forest Service re-

In: Sniezko, Richard A.; Samman, Safiya; Schlarbaum, Scott E.; Kriebel, Howard B., eds. 2004. Breeding and genetic resources of five-needle pines: growth, adaptability and pest resistance; 2001 July 23–27; Medford, OR, USA. IUFRO Working Party 2.02.15. Proceedings RMRS-P-32. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station.

A.J. Kegley and R.A. Sniezko are with the USDA Forest Service, Dorena Genetic Resource Center, 34963 Shoreview Road, Cottage Grove, Oregon, 97424, USA. Phone (541) 767-5716; Fax (541) 767-5709; e-mmail: akegley@fs.fed.us; rsniezko@fs.fed.us.

searchers in Idaho (Bingham 1983; McDonald and others, this proceedings; Sniezko 1996). Recently, screening of seedlings has been modified to incorporate an assessment for a major gene conditioning a hypersensitive response (HR) in needles of sugar pine and western white pine (see Kinloch and others 1999; Kinloch and Dupper 2002 for details on screening for major gene resistance); screening for HR complements but does not replace standard operational screening. Artificial inoculation with blister rust allows seedlings to be categorized into those with complete resistance (no stem infection) and those with partial resistance (stem infection present).

There is little published information on the inherent levels of blister rust resistance in WWP or SP in Oregon and Washington (Sniezko 1996). However, a synthesis of results from the Region 6 program has begun. This paper reports on the levels of resistance for 226 seedling families of WWP and 217 families of SP, representing a large part of their geographic ranges in Oregon and Washington. The data reveal the different types of resistance responses and relative degrees of responses between trials, species, and families, and compares them to existing checklot families.

Some refinements and modifications to Dorena screening summaries are incorporated here for the first time. A summary of these four trials along with analyses of other Region 6 studies will improve the advanced-generation breeding program. Moreover, better comparisons with the resistance screening and breeding programs in British Columbia, California, and Idaho will be possible.

Materials and Methods

Study Design

Four blister rust screening trials were examined, one WWP and one SP, from each of two test years, 1989 and 1994. These trials are denoted as WWP1989, SP1989, WWP1994, and SP1994. The seedling families in these trials are progeny of parent trees that cover much of the range of WWP in Oregon and Washington and of SP in Oregon (fig. 1a,b).

In each of the four trials, seed from 120 families were sown in a randomized complete block design with six blocks. Within each block, 10 seedlings per family were planted in row plots. Each trial contained a maximum of 7,200 seedlings (6 blocks x 1,200 seedlings/block). Seedlings were grown outside in open boxes (0.91 m wide x 1.21 m long x 0.30 m high; 10 boxes per block) for two growing seasons before artificial inoculation with blister rust.

Families

The majority of families included in each of these trials are half-sib seedling progeny ("Wild OP" families) of phenotypic selections from natural forests or plantings (table 1). In the WWP1994 trial, 42 of 110 Wild OP families were from the Quinault Indian Nation in western Washington, 57 were from the Colville National Forest (NF) in eastern Washington, and 11 were from several other National Forests in

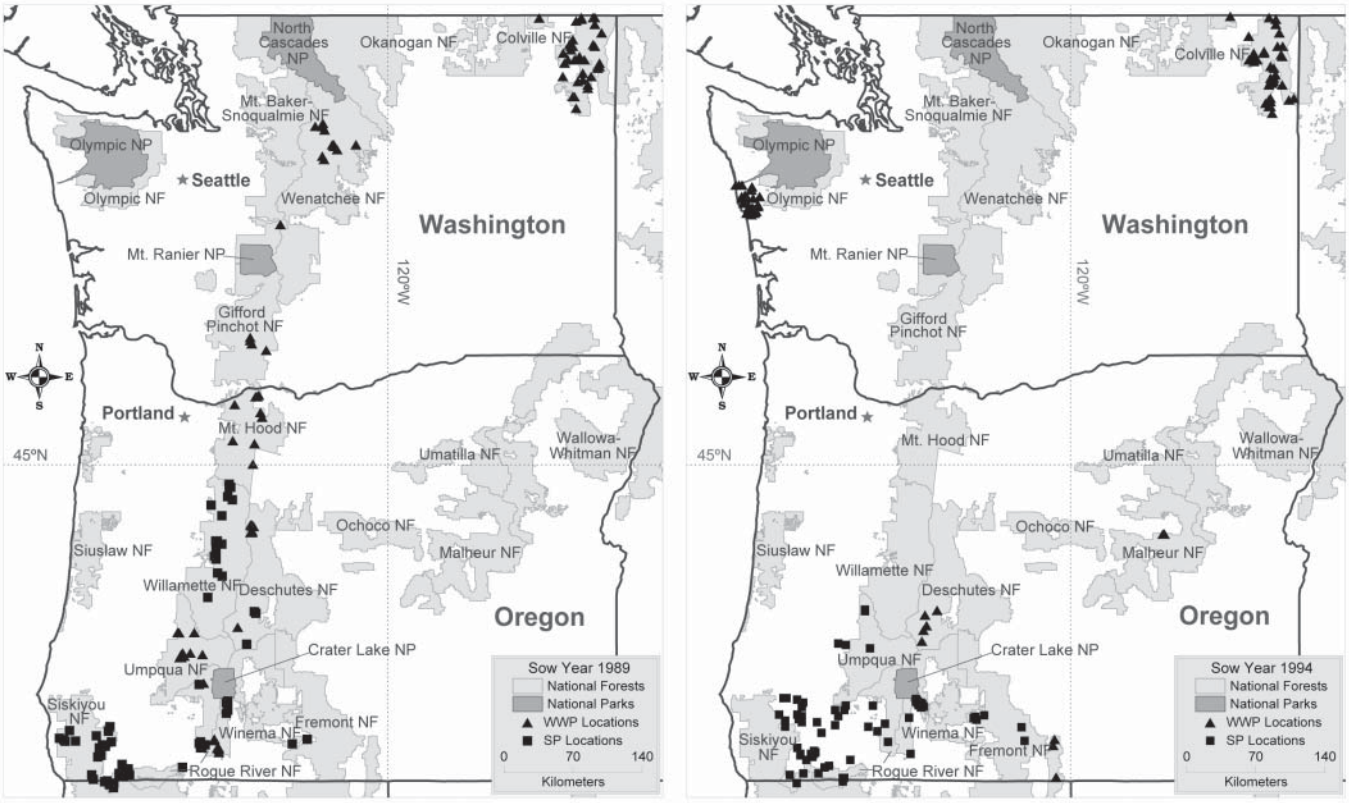

Figure 1—Geographic location of parent trees with half-sib progeny included in the four trials.

Table 1—Geographic location of parent trees of families included in four blister rust screening trials at Dorena Genetic Resource Center.

Species	Breeding Zone	National Forest	Elevation (ft)[c]	Sow Year 1989	Sow Year 1994
western	02180	Fremont	—		3
white	05010	Mt Baker-Snoqualmie	—	1	
pine	06020	Mt Hood	—	20	
	09070	Olympic[b]	—		42
	15040	Umpqua	—	28	
	16140	Wallowa-Whitman	—		3
	17110	Wenatchee	—	13	
	18030	Willamette	—	9	5
	21100	Colville	—	45	57
	Idaho[a]		—		4
	Dorena BR Check[a]	Colville	—		1
	Dorena HR Checks[a]	Umpqua	—	4	4
	Total			120	119
sugar	02176	Fremont	All	5	11
pine	10025	Rogue River	>4000	17	7
	10043	Rogue River	<2500		25
	10044	Rogue River	2500-4000	25	21
	10045	Rogue River	>4000	6	42
	11053	Siskiyou	<2500	3	
	11054	Siskiyou	2500-4000	27	7
	11055	Siskiyou	>4000	2	
	18033	Willamette	<3000	19	
	Dorena HR Checks[a]	Umpqua	—[d]	2	4
	Dorena LR Check	Umpqua	—[d]	1	
	Total			107	117

[a] The checklots are technically not listed by breeding zone but rather by source (Dorena or Idaho) or by resistance type HR (hypersensitive reaction), BR (bark reaction), or LR (low resistance).
[b] Although Breeding Zone 09070 corresponds with Olympic NF, the 42 families from breeding zone 09070 in Sow Year 1994 were from the neighboring Quinault Indian Nation.
[c] Unlike sugar pine, elevation is not a criterion for establishing western white pine breeding zones.
[d] Since these checklots are full-sib crosses between selected parents from different locations, or a mix (LR Check), elevations are not reported for these families.

Region 6. In the WWP1989 trial, 45 of 116 Wild OP families were from the Colville NF, 20 from Mt Hood NF, 28 from the Umpqua NF, and lesser numbers from other NFs (table 1 and fig. 1). The two SP trials sampled progeny of selected trees predominantly from the Rogue River NF and Siskiyou NF in southern Oregon (table 1 and fig.1). Because a few families had insufficient germination (less than 20 seedlings) for inclusion in analyses, the numbers of Wild OP families were as follows: 104 in SP1989; 116 in WWP1989; 110 in WWP1994; and 113 in SP1994.

In addition to the 226 WWP and 217 SP Wild OP families, two to four Dorena full-sib high resistance checklots per trial, one low resistance seedlot (bulk of three half-sib families, used only in SP1989), one half-sib bark reaction checklot (BR checklot) from the Colville NF (only in WWP1994), and four standard WWP full-sib checklot families from the Forest Service rust resistance program in Idaho (only in WWP1994; Aram Eramian, personal communication) were included. The Dorena full-sib high resistance checklots were progeny of crosses among parents that have been confirmed to have a hypersensitive reaction in the needles (HR, see Kinloch and Dupper 2002 for discussion of HR in WWP and SP). Three Dorena full-sib HR checklots were common between the two WWP trials; two full-sib HR checklots were common between the two SP trials.

Inoculation

Details of the inoculation procedure have been previously described by Sniezko (1996) and Samman (1982). Approximately 18 months after sowing, seedlings were moved to a large room (13.7 m x 10.1 m x 3.0 m) where they were inoculated with blister rust (table 2). Temperature within the inoculation chamber was maintained at around 16.7°C (62°F) and relative humidity at 100 percent.

Ribes leaves (the alternate host) infected with *C. ribicola* at the telial stage were collected from various forest sites in Oregon and Washington as well as the Dorena Ribes Garden. These leaves were placed on wire frames above the seedlings, telial side down. *Ribes* leaves were randomly distributed among the six blocks of each trial except in WWP1994; in that trial, leaves from the Dorena Ribes Garden were used on Block 5, and leaves from near Champion Mine on the Umpqua NF were used on Block 6. A virulent pathotype of the rust (with the vcr2 gene) that neutralizes HR in WWP (and the associated Cr2 gene) had been noted in both these locations (Kinloch and others 1999). Blocks 1 through 4 in WWP1994 used *Ribes* leaves from areas without known occurrence of vcr2.

Spore fall was monitored until the desired inoculum density was reached for each box (table 2); the *Ribes* leaves were

Table 2—Inoculation summary statistics for four blister rust screening trials at Dorena Genetic Resource Center.

	WWP1989[a]	SP1989	WWP1994	SP1994
Spore Density[b]				
Target Spore Density	3500	6000	3000	6000
Average Spore Density	3470	5233	3348	7216
Range in Spore Density	3095-3870	3355-6390	2925-3975	6215-8040
% Ribes leaves with vcr2[c]	85%	65%	33.3%	46.2%
Inoculation Temperature	58-63°F	60-62°F	61-64°F	62-63°F
Inoculation Humidity	100%	100%	100%	100%
Spore Germination %	—[d]	86.3%	94.7%	95.6%

[a] Where WWP=western white pine and SP=sugar pine and the year indicates year sown for testing.
[b] The number of basidiospores of *Cronartium ribicola* per square centimeter.
[c] Percentage of total leaves used that originated from areas where vcr2 pathotype virulent to HR in WWP have been confirmed. In WWP1989, SP1989, and SP1994, leaves with vcr2 (Ribes Garden and Champion Creek) were randomly distributed among the blocks. However in WWP1994, potential leaves with vcr2 were placed on blocks 5 and 6 only. Note: vcr2 does not appear to overcome HR in SP.
[d] Basidiospore germination was not assessed in the WWP1989 trial.

then removed. After the target inoculum density was reached, the seedlings were left in the inoculation chamber for approximately 48 hours to ensure spore germination; boxes were then returned to their previous outdoor location. In each of the 2 years, the sugar pine and western white pine trials were inoculated separately.

Assessments of Resistance Traits

Seedlings were assessed for blister rust symptoms six times over a period of 5 years. The first inspection occurred approximately 9 months after inoculation. Seedlings were evaluated for the presence and number of needle lesions ("spots"). The checklots in each trial were used to monitor the presence of needle lesions on all secondary needles until the number of spots reached a maximum. These checklot counts were then used to establish five needle lesion classes specific to each trial. The scale was set up to have approximately 25 percent of the seedlings in each needle lesion class from 1 to 4. Table 3 lists the number of spots in each needle lesion class for each trial.

The second inspection occurred approximately 3 months after the first inspection (1 year after inoculation). Seedlings were assessed for the presence of needle lesions, stem symptoms (cankers and bark reactions), and height. Subsequent inspections of stem symptoms and mortality occurred annually. Bark reaction data was only recorded through the third year after inoculation (inspection 4).

Based upon data collected from the six inspections, presence or absence of resistance responses (table 4) was determined for each seedling. In general, seedlings were characterized by the presence or absence of needle lesions and/or stem symptoms, the type of stem symptom, their survival after 3 and 5 years, and their height after three growing seasons (1 year after inoculation).

Analyses of variance (Proc GLM, SAS 1999) were performed separately for each of the four trials using family block means of each trait for two subsets of data: (1) all families, including checklots and (2) Wild OP families only. The model included Family and Block effects. Although the data for many of the traits were not normally distributed, F-tests are fairly robust against this violation (Cochran and Cox 1967; Zolman 1993).

Table 3—Number of needle lesions (spots) in each class for seedlings in four screening trials.

	# Spots/Needle Lesion Class				
Trial[a]	0	1	2	3	4
WWP1989	0	1-2	3-5	6-10	11+
SP1989	0	1	2	3-5	6+
WWP1994	0	1-10	11-25	26-48	49+
SP1994	0	1-2	3-5	6-15	16+

[a] Where WWP=western white pine and SP=sugar pine and the year indicates year sown for testing.

Table 4—Traits and derived variables used in assessing seedlings for blister rust resistance.

Category	Trait		Description of Trait
General	Infected		Seedling developed needle lesions and/or stem symptoms
	NOINFECT		Seedling had no needle lesions present at either first or second inspection and had no stem symptoms in subsequent inspections.
	RSURV3		Infected tree is alive 3 years after inoculation
	RSURV5		Infected tree is alive 5 years after inoculation
	TSURV5		Seedling (infected or uninfected) is alive 5 years after inoculation
	HT3		Total height (cm), including any lammas growth, of seedling after three growing seasons
Needle Lesions	NLC	Needle Lesion Class	A categorical classification of number of needle lesions ('spots') on all secondary needles on a seedling at first inspection
	SPO	Spots Only	Seedling had needle spots but did not develop stem symptoms through 5 years after inoculation
	SPOT%		Seedling had needle lesions present at either first (SPOT1%) or second inspection (SPOT2%)
Stem Symptoms	SSFREE	Stem symptom free	Seedling was free of stem symptoms (initial orange discoloration of the bark, normal canker or bark reaction) at any of the six inspections following inoculation
	SS	Stem Symptoms	Seedling exhibited normal canker, bark reaction, or was dead of rust
	ESS3	Early Stem Symptoms	Calculated as the ratio of seedlings with stem symptoms (SS) one year after inoculation relative to those with SS three years after inoculation. A lower value indicates families with relatively slower or delayed appearance of stem symptoms[a]
	SSAL3	Stem symptom alive	Seedling with stem symptoms (SS) and alive (AL) 3 years after inoculation[a]
	SSAL5		Seedling with stem symptoms (SS) and alive (AL) 5 years after inoculation[a]
	NCANK	Normal Canker	Seedling exhibits initial orange discoloration of the bark or fusiform swelling with an active orange margin (Kinloch and Davis 1996; Hunt 1997) at any inspection[a]
	BR	Bark Reaction	Seedling exhibits bark reaction, that is, an incompatible interaction with the fungus (Theisen 1988). BR manifests as a sunken necrotic lesion, often at the base of a needle fascicle, on stem tissue. When no fungal activity is observed, the BR is considered 'complete.' An 'incomplete' or 'partial' BR does not completely halt fungal growth (Kinloch and Davis 1996; Franc 1988)[ab]

[a] The denominators for the BR%, NCANK%, ESS3%, and SSAL% calculations included only trees with stem symptoms.
[b] A seedling could be scored as having both a bark reaction and a normal canker due to the presence of (1) multiple stem infections or (2) a transition in status from normal canker to bark reaction or vice-versa in different inspection years.

Results

Survival and Growth

Survival of Wild OP families 5 years after inoculation (TSURV5) was low in all trials, averaging about 2 percent to 14 percent (table 5). TSURV5 was higher in the 1989 trials than in the 1994 trials (table 5). TSURV5 was slightly higher for SP relative to WWP in 1989, but the reverse was true in the 1994 trials. Overall survival of rust-infected Wild OP seedlings (RSURV5) was generally low in all trials, but varied by trial and species (table 5, fig. 2). Like TSURV5, RSURV3 and RSURV5 were higher for the 1989 trials than the 1994 ones (table 5, fig. 2, 3). Fifth-year survival of rust-infected Wild OP seedlings was 9.9 percent and 13.1 percent for WWP and SP, respectively, in the 1989 trials and 3.2 and 1.6 percent in the 1994 trials (table 5, fig. 3). In the 1989 trials the decline in survival began later, between the third and fourth inspections (approximately 2 and 3 years after inoculation), than in the 1994 trials (fig. 2). Despite the very low overall survival, all four trials had at least one family with more than 20 percent survival of infected seedlings (see RSURV5 in table 5 and fig. 3).

RSURV5 of the Idaho full-sib families was moderate, in the 30 to 40 percent range, slightly lower than the Dorena BR checklot (56 percent, table 5). For those seedlings with bark reactions, survival was low but higher than those with normal stem cankers (BRSURV5 and NCSURV5,

Table 5—Trial means and means of the Dorena and Idaho checklots as well as several Wild OP families with relatively high levels of resistance in four blister rust screening trials.

			Survival and Growth[b]										Needle Symptoms[b]					Stem Symptoms[b]			
Trial	Family	Type[a]	infect	no infect	rsurv3	rsurv5	tsurv5	ssal3	ssal5	nc surv5	br surv5	ht	NLC	spo	spot1	spot2	spot	ss	ess3	br	ncank
			Percent									cm		Percent							
WWP	06023-513	Wild OP	97.6	2.4	76.9	52.1	40.0	74.0	36.7	33.9	68.0	31.7	1.6	25.1	88.4	35.3	88.4	72.6	13.9	32.7	61.3
1989	15045-443	Wild OP	98.3	1.7	83.5	54.8	51.7	74.1	32.4	27.1	66.7	29.5	2.7	30.6	98.3	56.8	98.3	67.8	71.8	20.5	64.1
	15045-642	Wild OP	100.0	0.0	78.0	42.2	40.0	76.3	37.6	26.5	58.3	29.8	2.6	8.9	100.0	48.7	100.0	91.1	41.3	34.0	80.4
	21104-418	Wild OP	100.0	0.0	75.0	35.0	35.0	71.4	28.3	16.1	47.8	37.3	2.2	10.0	100.0	68.3	100.0	90.0	43.2	50.4	78.3
	17114-631	Wild OP	100.0	0.0	86.7	54.8	53.3	84.7	44.6	35.0	71.3	32.0	2.4	22.6	100.0	40.4	100.0	77.4	55.3	27.2	67.2
	15045-861 x 15045-837	HR check	100.0	0.0	73.0	18.9	18.3	68.7	1.9	1.9	0.0	32.8	2.3	17.2	98.3	37.7	100.0	82.8	51.3	4.2	82.8
	15045-861 x 15045-862	HR check	100.0	0.0	88.0	30.6	30.0	83.7	10.0	10.0	0.0	33.5	1.9	22.0	98.3	49.1	98.3	78.0	56.4	2.1	78.0
	15045-862 x 18034-392	HR check	90.0	10.0	97.9	65.5	63.3	95.8	39.8	29.3	61.7	36.5	1.7	39.9	90.0	23.1	90.0	50.1	35.0	25.7	43.3
	15045-896 x 15045-862	HR check	100.0	0.0	91.7	30.7	30.0	87.2	7.0	2.1	0.0	36.1	2.0	25.7	98.3	27.4	98.3	74.3	56.6	2.4	69.3
		Wild OP avg	**99.4**	**0.6**	**66.6**	**9.9**	**9.5**	**65.3**	**6.0**	**4.8**	**10.8**	**30.2**	**2.4**	**4.6**	**98.1**	**59.0**	**98.4**	**94.9**	**65.9**	**12.3**	**93.1**
SP	18033-317	Wild OP	86.3	4.5	68.8	24.5	21.7	77.8	20.8	14.1	33.3	33.2	1.5	6.5	62.5	32.9	64.6	89.0	31.5	8.9	78.3
1989	18034-112	Wild OP	87.7	11.6	74.6	39.4	36.7	69.8	33.1	6.9	25.0	35.9	1.7	12.2	75.2	31.6	79.5	76.2	40.8	15.4	47.8
	10044-212	Wild OP	84.3	15.7	92.6	42.8	46.7	94.3	23.6	14.6	100.0	39.7	1.5	23.1	68.8	27.7	74.8	61.2	46.7	5.7	53.8
	10044-230	Wild OP	77.4	22.6	83.7	49.8	36.7	72.2	31.7	9.7	83.3	47.1	1.5	18.8	58.8	25.7	71.3	58.6	34.7	23.6	42.1
	11054-877	Wild OP	85.0	15.0	84.8	37.4	45.0	82.1	23.2	16.4	38.9	45.3	1.8	15.4	75.0	33.3	76.7	69.6	45.3	36.9	56.3
	B1054-004 x B1054-034	HR check	78.3	22.8	67.4	60.8	68.3	56.7	44.3	17.3	62.5	40.1	1.7	33.6	66.3	17.7	71.3	43.7	80.0	34.4	27.5
	B1054-034 x 10044-050	HR check	77.4	22.8	50.3	40.4	51.7	24.5	5.8	6.0	0.0	37.3	1.9	30.0	68.5	33.3	74.1	47.1	67.1	27.3	45.5
	Low Resistance Mix	LR Check	94.4	5.6	75.6	10.4	15.0	72.6	3.9	2.1	0.0	41.0	2.0	7.0	76.1	43.0	81.7	87.4	52.7	8.1	80.0
		Wild OP avg	**95.8**	**3.8**	**70.0**	**13.1**	**14.1**	**70.6**	**8.4**	**4.7**	**13.5**	**39.0**	**2.1**	**5.5**	**78.1**	**52.5**	**85.1**	**90.7**	**58.9**	**7.2**	**85.7**
WWP	02187-047	Wild OP	98.1	0.0	10.0	6.7	6.7	11.1	5.6	5.6	—	17.1	2.2	4.2	98.1	46.9	98.1	95.8	50.3	0.0	95.8
1994	21105-853	Wild OP	100.0	0.0	41.7	20.0	20.0	31.7	8.9	3.9	10.0	45.5	2.2	11.7	98.3	46.7	100.0	88.3	48.8	31.7	83.3
	09070-852	Wild OP	98.3	1.7	30.7	25.2	25.0	9.9	0.0	0.0	0.0	38.0	1.5	25.5	93.1	28.7	93.1	72.8	38.0	7.1	71.2
	09070-892	Wild OP	96.7	3.7	39.6	13.9	16.7	26.6	4.0	0.0	5.6	48.7	1.7	10.9	95.0	61.7	95.0	85.4	32.0	32.8	78.3
	09070-896	Wild OP	100.0	0.0	20.0	13.3	13.3	11.4	5.4	3.7	16.7	46.4	2.2	8.3	100.0	53.3	100.0	91.7	58.5	25.6	88.3
	21104-036	Wild OP	100.0	0.0	34.3	13.7	13.3	32.1	9.6	9.8	17.3	41.1	3.1	5.2	100.0	72.6	100.0	94.8	48.4	38.8	93.1
	21105-052	BR check	100.0	0.0	81.5	55.7	55.0	73.3	35.2	29.1	52.0	46.4	2.3	28.7	96.7	52.4	96.7	71.3	35.7	59.9	62.4
	17 x 293	Idaho	98.3	1.7	54.4	32.2	33.3	47.3	23.1	17.5	41.7	56.2	2.5	11.7	96.7	50.0	98.3	86.7	38.0	48.2	81.7
	221 x 220	Idaho	100.0	0.0	45.0	33.3	33.3	22.4	13.9	2.1	26.7	41.4	3.3	22.8	100.0	53.3	100.0	77.2	43.9	16.5	65.2
	208 x 314	Idaho	96.7	3.5	67.8	40.0	41.7	54.0	23.9	20.4	30.7	45.0	2.0	22.0	95.0	31.7	95.0	74.4	22.4	33.5	69.4
	222 x 225	Idaho	100.0	0.0	38.3	31.7	31.7	24.2	19.0	17.6	30.0	47.2	3.2	17.2	100.0	43.3	100.0	82.8	49.6	26.3	81.1
	15045-861 x 15045-862	HR check	100.0	0.0	40.0	38.3	38.3	2.9	0.0	0.0	0.0	39.8	3.3	43.9	100.0	51.7	100.0	56.1	47.7	2.9	56.1
	15045-862 x 15045-837	HR check	100.0	0.0	53.5	51.9	51.7	5.0	0.0	0.0	0.0	48.3	2.9	51.9	100.0	44.8	100.0	48.1	43.3	15.0	48.1
	15045-862 x 18034-392	HR check	100.0	0.0	51.7	36.7	36.7	26.5	2.1	2.1	0.0	45.8	2.7	35.6	100.0	50.0	100.0	64.4	54.8	20.9	64.4
	15045-896 x 15045-862	HR check	100.0	0.0	40.0	40.0	40.0	0.0	0.0	0.0	0.0	42.4	3.0	40.9	100.0	37.1	100.0	59.1	59.2	2.1	59.1
		Wild OP avg	**99.5**	**0.3**	**6.9**	**3.2**	**3.4**	**4.6**	**0.7**	**0.6**	**1.2**	**42.3**	**2.6**	**2.6**	**98.4**	**76.4**	**98.8**	**97.1**	**70.7**	**7.2**	**96.8**
SP	98-01-018	Wild OP	100.0	0.0	5.0	0.0	0.0	3.7	0.0	0.0	0.0	44.7	2.6	0.0	100.0	75.1	100.0	100.0	79.9	26.1	98.1
1994	B1053-1380	Wild OP	91.7	8.3	53.6	28.7	33.3	2.4	5.2	2.4	33.3	54.2	1.5	34.7	91.7	43.3	91.7	56.9	62.1	11.3	51.8
	15043-402	Wild OP	100.0	0.0	20.9	13.7	10.0	16.3	6.9	6.9	—	42.9	2.1	7.9	95.5	60.4	95.5	92.1	66.1	0.0	92.1
	B1052-1993	Wild OP	100.0	0.0	5.0	3.3	3.3	5.0	3.3	3.3	0.0	56.7	3.0	0.0	97.9	79.5	100.0	100.0	74.4	4.4	100.0
	B2-2400	Wild OP	98.3	1.7	21.1	10.2	11.7	15.1	3.5	3.5	—	53.4	1.7	6.7	91.7	50.7	96.7	91.7	61.4	0.0	91.7
	B1054-004 x B1054-034	HR check	79.8	20.2	62.6	60.7	63.3	15.7	15.7	10.0	50.0	49.1	1.9	45.3	79.8	24.3	79.8	34.5	69.3	10.7	31.2
	B1054-034 x 10044-050	HR check	94.8	5.4	54.2	50.8	51.7	5.6	5.6	0.0	33.3	51.6	2.3	51.3	91.1	46.3	94.8	43.2	87.8	15.3	40.5
	B1054-005 x B1054-034	HR check	92.6	7.4	67.4	46.4	41.7	23.6	18.1	0.0	100.0	60.9	2.0	43.3	90.7	27.0	90.7	49.3	54.2	18.1	42.8
	B1054-005 x 10044-049	HR check	93.4	0.0	43.3	37.0	26.7	9.7	9.7	5.6	16.7	58.1	1.6	34.8	85.4	38.5	93.4	65.2	65.3	20.8	62.4
		Wild OP avg	**99.5**	**0.2**	**3.3**	**1.6**	**1.6**	**2.0**	**0.5**	**0.4**	**3.4**	**47.6**	**2.9**	**1.2**	**97.8**	**82.9**	**98.6**	**98.6**	**84.5**	**3.5**	**98.4**

[a] Where type refers to the classification of the material: Wild OP=half-sib progeny of phenotypic selections, BR check=half-sib family previously screened and having a high level of bark reaction, Idaho=full-sib checklot from the Idaho blister rust program, HR check=Dorena full-sib checklot with hypersensitive reaction in the needles, LR check=mix of low resistant half-sib material, Wild OP avg=mean of all Wild OP families included in the trial.

[b] Traits are defined in Table 4 except ncsurv5 (survival with normal canker 5 years after inoculation) and brsurv5 (survival with bark reaction 5 years after inoculation).

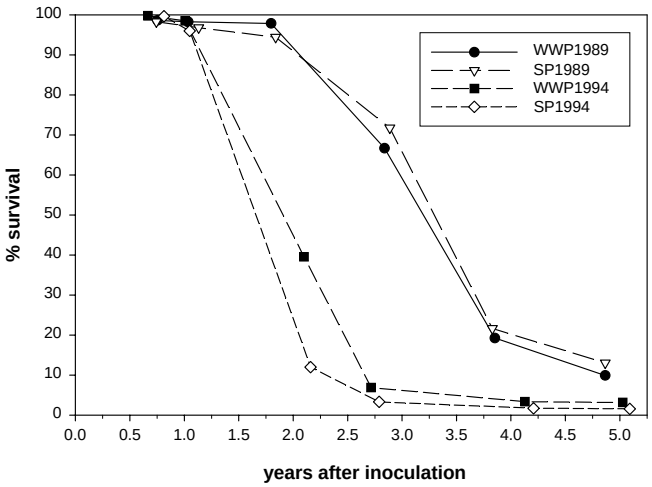

Figure 2—Survival of infected half-sib progeny of western white pine (WWP) and sugar pine (SP) after inoculation with blister rust. Each inflection point represents date of assessment.

respectively, table 5). Survival of the Dorena HR WWP checklots was about 42 percent in the 1994 trial compared to 36 percent in the 1989 trial. Survival of the two HR SP checklots common to both test years was slightly higher, ranging from 40 to 61 percent. RSURV5 of the Low Resistant SP checklot was lower than the average of the Wild OP families (10 percent versus 13 percent) (table 5).

Families varied significantly for height in all four trials (table 6). The checklots were generally intermediate for HT (fig. 4, table 5). Mean height was greater in the 1994 trials than the 1989 trials (table 5, fig. 4).

Rust Infection

Infected seedlings in the Wild OP families (infection percentage) averaged 95.8 to 99.5 percent for the four trials (table 5). Sugar pine seedlings had more variation in the frequency of infection than WWP seedlings. Wild OP family means for SP ranged from 77.4 to 100 percent in the 1989 trial and from 83.3 to 100 percent in the 1994 trial. For WWP

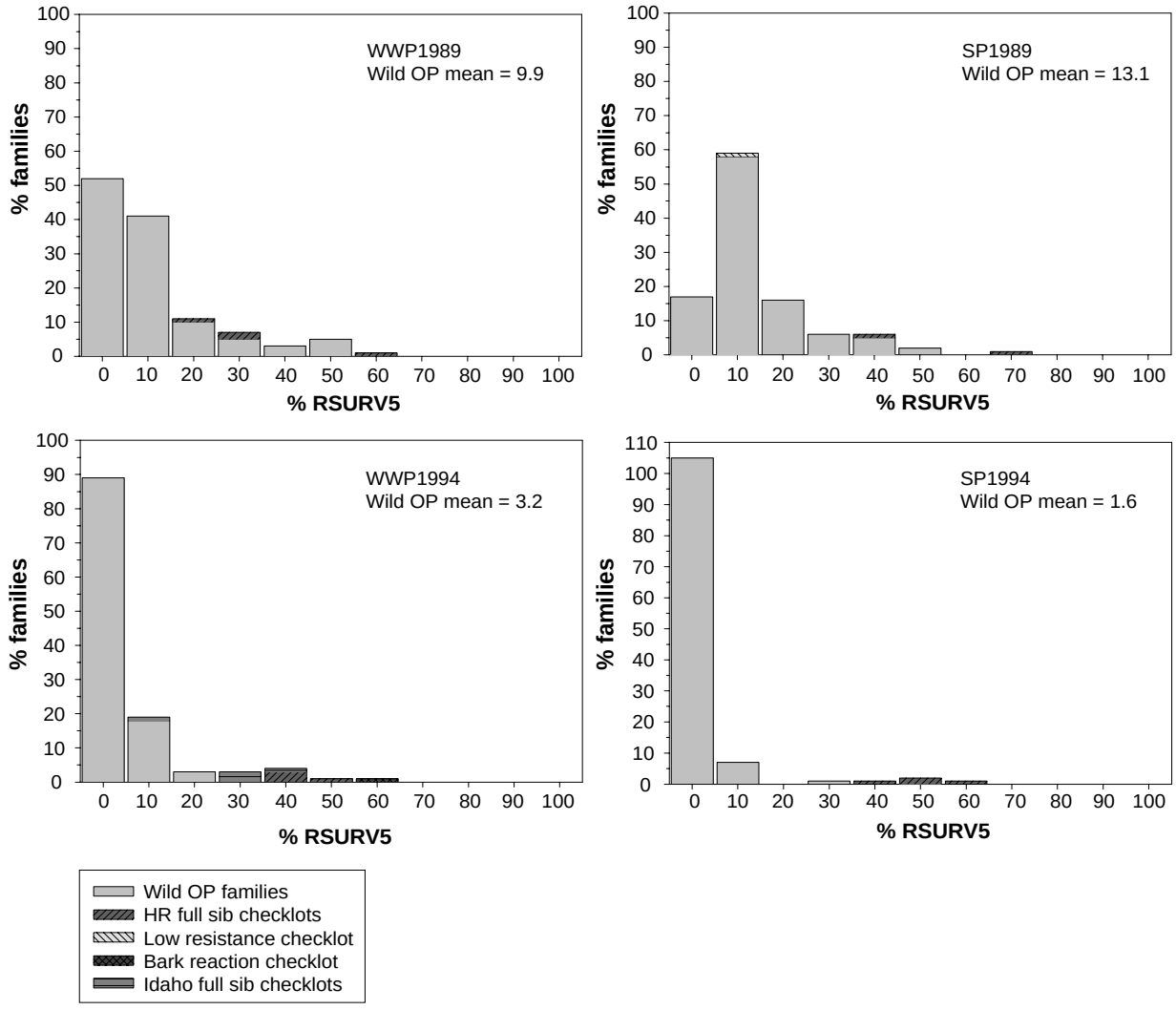

Figure 3—Distribution of family means for survival of infected seedlings 5 years after inoculation (RSURV5) for four blister rust screening trials.

Table 6—P-values associated with F-tests for significant differences among families from analyses of variance for each of four blister rust screening trials at Dorena.

		Wild OP families only				All families			
	Trait	WWP 1989	SP 1989	WWP 1994	SP 1994	WWP 1989	SP 1989	WWP 1994	SP 1994
General	Infect %	**0.2100**[a]	<0.0001	**0.2430**	**0.2635**	0.0113	<0.0001	**0.1656**	<0.0003
	NOINFECT %[b]	**0.2100**	<0.0001	0.0006	0.0183	0.0113	<0.0001	0.0003	<0.0001
	RSURV3 %	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
	RSURV5 %	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
	TSURV5 %	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
	HT	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Needle Lesions	NLC	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
	SPO %	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
	SPOT%	**0.1247**	<0.0001	0.0349	**0.0725**	**0.0836**	<0.0001	0.0152	0.0042
	SPOT1 %	**0.2309**	0.0007	0.0024	0.0197	**0.1929**	0.0009	0.0009	0.0012
	SPOT2 %	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Stem Symptoms	SSFREE %	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
	SS %	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
	ESS3 %	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
	SSAL3 %	<0.0001	<0.0001	<0.0001	0.0108	<0.0001	<0.0001	<0.0001	<0.0001
	SSAL5 %	<0.0001	<0.0001	<0.0001	**0.0747**	<0.0001	<0.0001	<0.0001	<0.0001
	BR %	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
	NCANK %	<0.0001	<0.0001	**0.1453**	**0.5214**	<0.0001	0.0045	0.0017	<0.0001

[a] P-values in bold type are not significant at the $\alpha = 0.05$ level.
[b] NOINFECT% is not necessarily equal to 100-infect%. To be classified as NOINFECT, a seedling must survive through the end of the assessment period (6th Inspection) without ever developing needle lesions or stem symptoms.

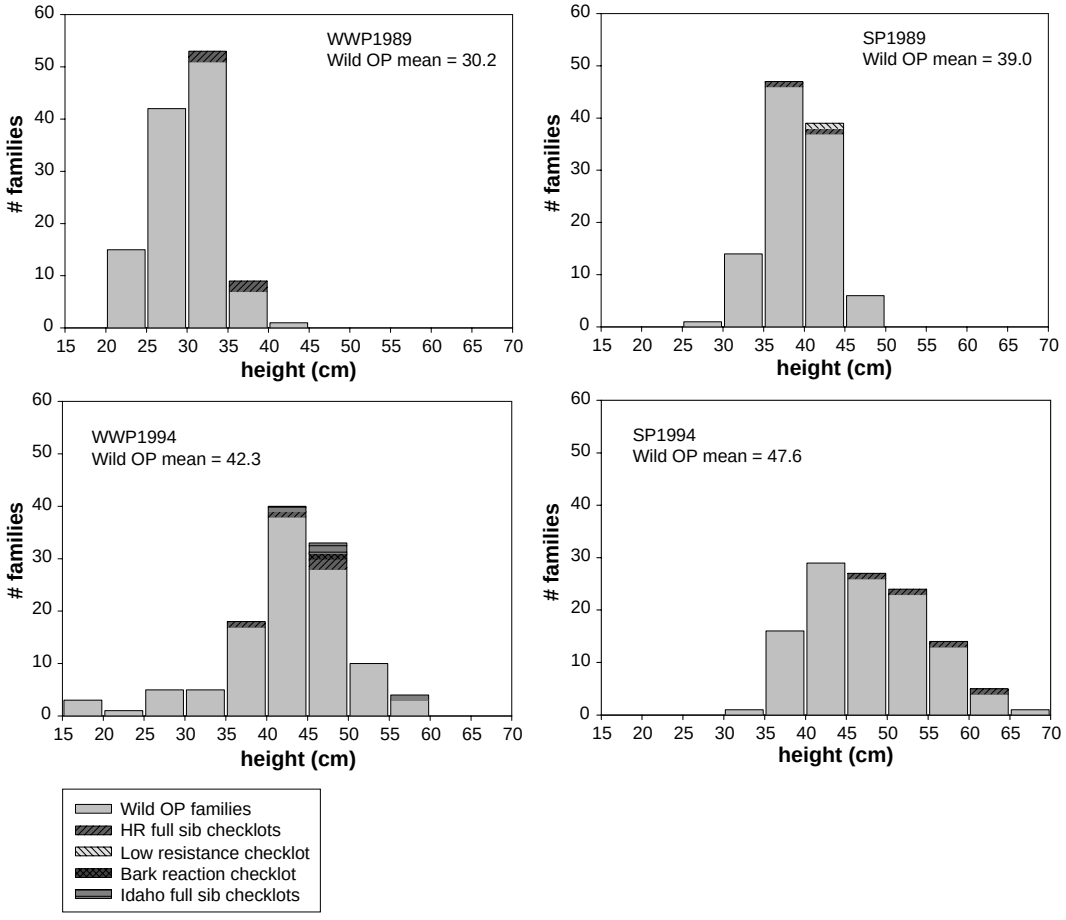

Figure 4—Distribution of family means for height after three growing seasons for the four trials.

family means ranged from 96.7 to 100 percent, including the full-sib families from Dorena and Idaho. Checklots of SP were not consistent in their infection levels between the two trial years. In SP1989, the two resistant Dorena checklots had 77 to 78 percent infection, and the low resistant control had 94 percent infection, whereas the checklots in SP1994 had 80 to 95 percent infection (table 5). Checklots for WWP had 90 to 100 percent infection in the two trials. In three of the four trials, infection percentage was nearly constant over the course of the evaluation, but in SP1989 infection levels increased from first to fourth inspection (fig. 5).

There were very few uninfected seedlings (noninfected seedlings were without any needle lesions and without any stem symptoms). The mean percentage of uninfected seedlings was less than 1 percent for the Wild OP families in the WWP1989 trial and the two 1994 trials; the percentage was about 4 percent for Wild OP families in the SP1989 trial (table 5).

With a few exceptions, families varied significantly ($p<0.0001$) in all traits within each trial, whether or not checklots were included in the analyses (table 6).

Needle Lesions

The percentage of seedlings with needle lesions (spot percent) was very high in all trials, averaging 85 to 99 percent among Wild OP families (table 5). At first inspection, approximately 98 percent of the Wild OP seedlings in WWP1989, WWP1994, and SP1994 had needle lesions, while only 78 percent of SP1989 had spots (spot1, table 5). By the second inspection (approximately 1 year after inoculation), the percentage of trees with needle spots dropped in all four trials; Wild OP run averages for spot2 ranged from 59 to 83 percent (table 5, fig. 6). The shedding of needles was most likely responsible for spot2 values being lower than spot1. The checklots were among the families with the lowest percentage of seedlings with needle lesions at second inspection (table 5, fig. 6).

The number of needle lesions per needle lesion class also varied by trial and species (table 3). In these trials the minimum number of spots required to enter the highest needle lesion class (Class 4) ranged from 6 to as high as 49 (table 3). More needle lesions were present in the 1994 trials relative to the 1989 trials and in western white pine relative to sugar pine (despite the lower inoculum density in the WWP trials). Family mean needle lesion class (NLC) ranged from 1.5 to 4.0, but the distribution of families across classes shifted depending upon the species and the trial (fig. 7). Within a trial, the SP HR checklots generally had fewer than average needle lesions, but the Dorena HR WWP checklots were not consistent between trials (fig. 7, table 5). There was a wide range in NLC among the four Idaho full-sib checklots in WWP1994 (table 5, fig. 7).

The mean percentage of seedlings with needle spots and no stem symptoms (SPO) was low for Wild OP families in all trials, ranging from 1.2 to 5.5 percent (table 5). Some Wild OP families in each trial had more than 10 percent of seedlings with only needle spots with some families showing 30 to 35 percent seedlings with SPO (table 5, fig. 8). All the checklot families, except the Low Resistant checklot in SP1989, showed much higher levels of SPO than the Wild OP trial mean (table 5, fig. 8).

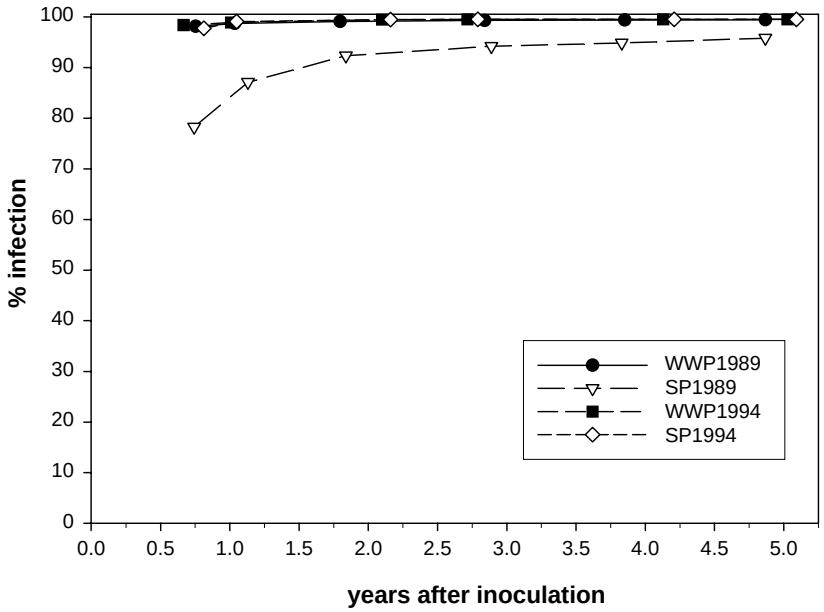

Figure 5—Percent infection of half-sib progeny of phenotypic selections of western white pine (WWP) and sugar pine (SP) in four blister rust screening trials. Each inflection point represents date of assessment.

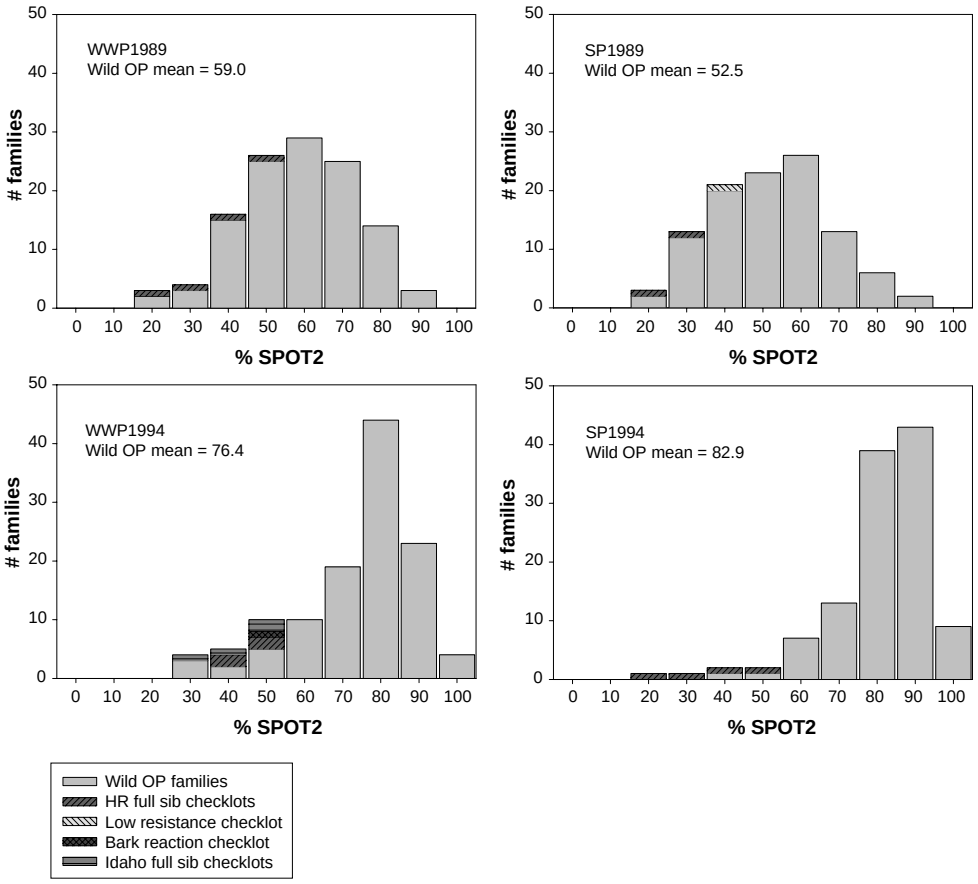

Figure 6—Distribution of family means for the percentage of seedlings with needle spots (SPOT2) 1 year after inoculation (second inspection) for the four trials.

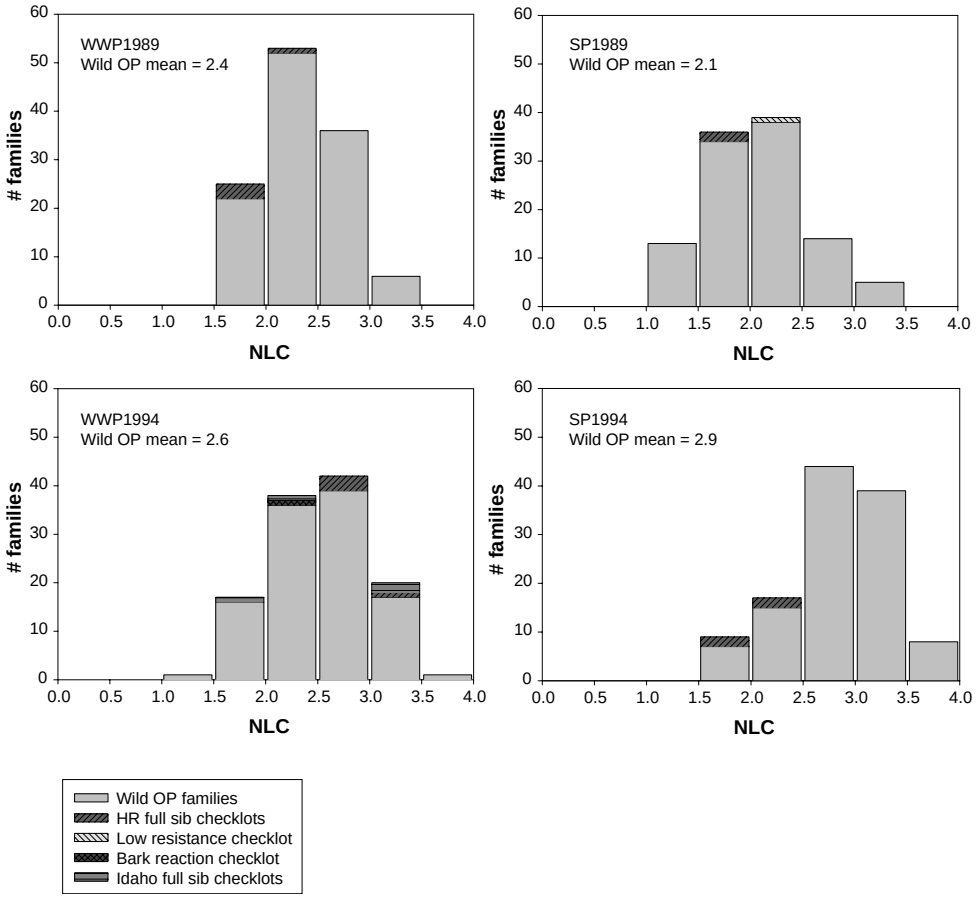

Figure 7—Distribution of family means for needle lesion class (NLC) for the four trials. NLC is a measure of the relative frequency of needle lesions within a test.

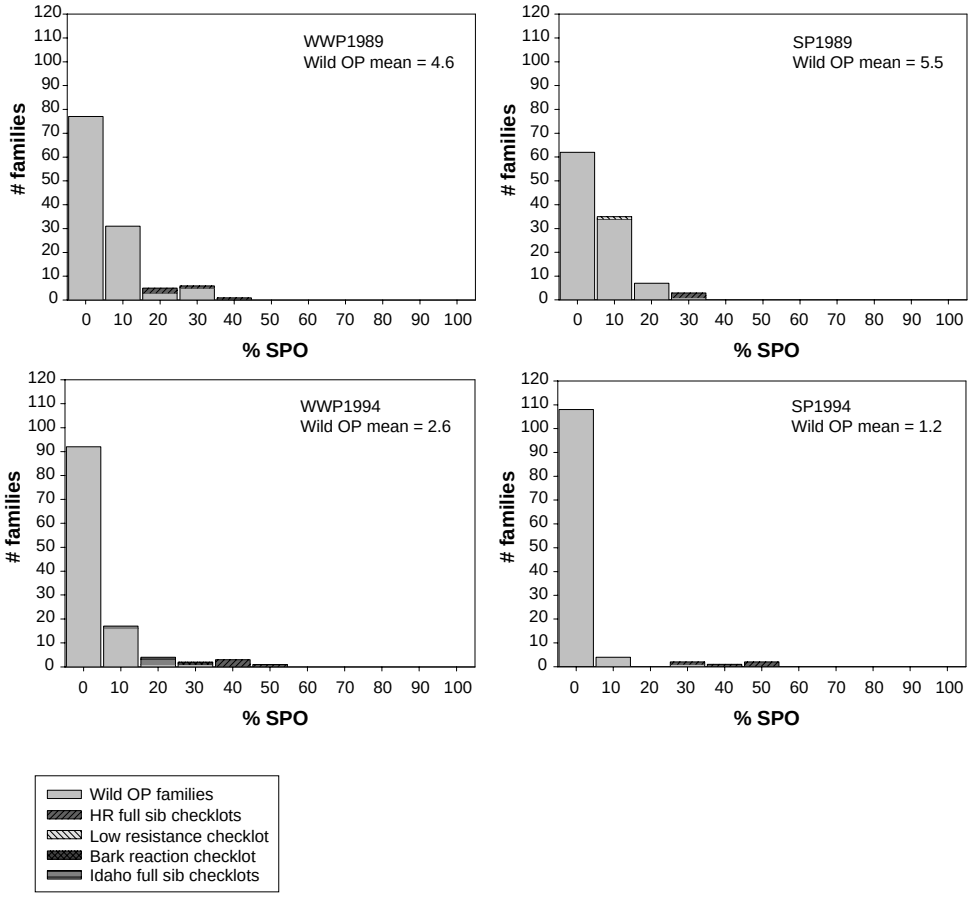

Figure 8—Distribution of family means for percentage of seedlings with needle spots but no stem symptoms (SPO) in the four trials.

Stem Symptoms

More than 90 percent of the seedlings in most Wild OP families developed stem symptoms (SS); the trial average percentages varied from 91 to 99 percent SS (table 5). Even so, there were significant differences among families in all four trials (table 6). The same general pattern of an increase over time in the percentage seedlings with stem symptoms developed in the four trials, although there were some differences in when the SS percent attained a maximum. For example, in SP1989, SS percent continued to increase slightly to the end of the evaluation period (sixth inspection, 5 years after inoculation) whereas SS percent stabilized by the fourth inspection (3 years after inoculation) in the other three trials (fig. 9).

Each of the four trials had at least one Wild OP family with less than 75 percent of the seedlings with stem symptoms (fig. 10 and table 5); SP1989 had nine Wild OP families, WWP1989 had five families, and the two 1994 trials each had one family with less than 75 percent of the seedlings having stem symptoms (table 5, fig. 10). The Dorena WWP and SP checklots with HR (hypersensitive reaction in the foliage) generally had among the lowest SS percent in all trials. The sugar pine checklots with HR averaged 45 percent and 48 percent SS in the two SP trials, respectively; the resistant western white pine checklots averaged 71 percent and 57 percent SS in the WWP trials. In the WWP1994 trial, the Idaho checklots and the Dorena BR checklot also showed low SS percent (71 to 87 percent) relative to the Wild OP families; however, the Dorena HR checklots were even lower with 48 to 64 percent SS in this trial (fig. 10 and table 5). The Dorena Low Resistant checklot in SP1989 had relatively high SS percent (87.4 percent) (table 5, fig. 10).

Normal Cankers—The percentage of Wild OP seedlings with normal cankers in at least one inspection (NCANK) varied from 86 to 98 percent for the four trials. The percentage was higher in 1994 trials relative to 1989, but the ranks between species were not consistent for the 2 years (table 5). The Wild OP sugar pine families with the lowest NCANK had values of 42 and 52 percent, whereas the western white pine families with the lowest NCANK had values of 61 and 71 percent (table 5). Survival of cankered seedlings from Wild OP families 5 years after inoculation (NCSURV5) was very low, ranging from 0.4 to 4.8 percent among the four trials (table 5). Survival of trees with a normal canker was higher for Wild OP families in the 1989 trials than for the

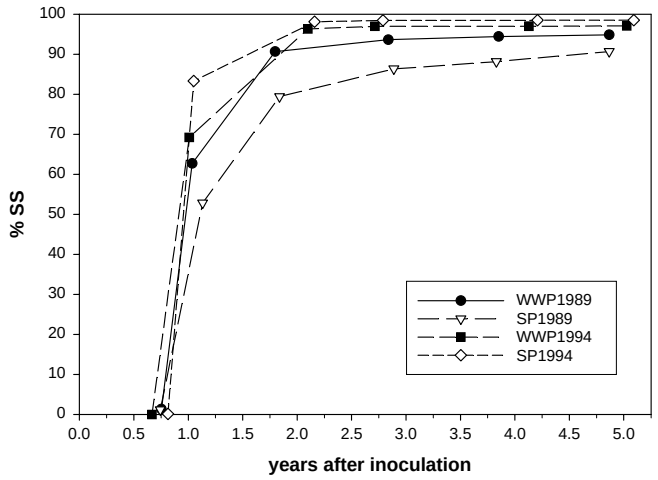

Figure 9—Percentage seedlings with stem symptoms (SS) of half-sib progeny of western white pine (WWP) and sugar pine (SP) in four screening trials. Each inflection point represents date of assessment.

1994 trials. Within a year, survival with a normal canker was slightly higher for WWP than for SP Wild OP families (table 5).

The BR checklot in WWP1994 was among the families with the lowest NCANK percent in that trial (fig. 11, table 5). The Dorena HR checklots had less NCANK percent than the Wild OP average, and the low resistance checklot approached the mean NCANK percent for SP1989 (table 5).

Bark Reaction (BR)—The incidence of trees with bark reactions was low in all four trials, ranging from 3.5 to 12.3 percent (table 5, fig. 12). Significant differences existed among Wild OP families for BR in all four trials (table 6). WWP showed slightly higher amounts of BR than SP (table 5, fig. 12). In all four trials, the majority of seedling families had at least one seedling with bark reaction (unpublished data). In all trials, there was at least one family with more than 20 percent of the seedlings with bark reaction (fig. 12). The Wild OP family with the highest BR (50.4 percent) was from the Colville NF (Family 21104-418) in the WWP1989 trial (table 5). The Dorena SP HR checklots had higher than average BR in both trials whereas the Dorena WWP HR

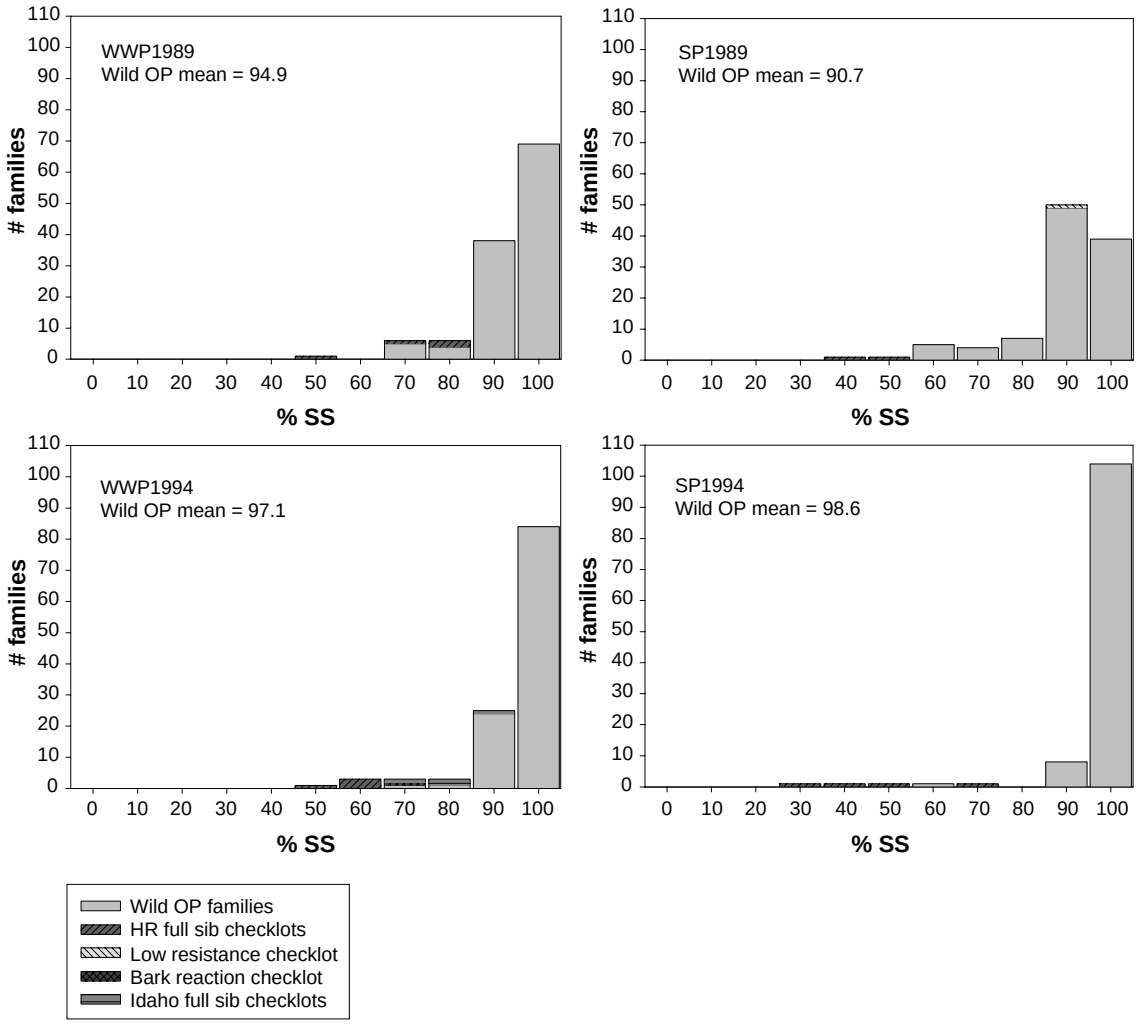

Figure 10—Distribution of family means for seedlings with stem symptoms (SS) in the four trials.

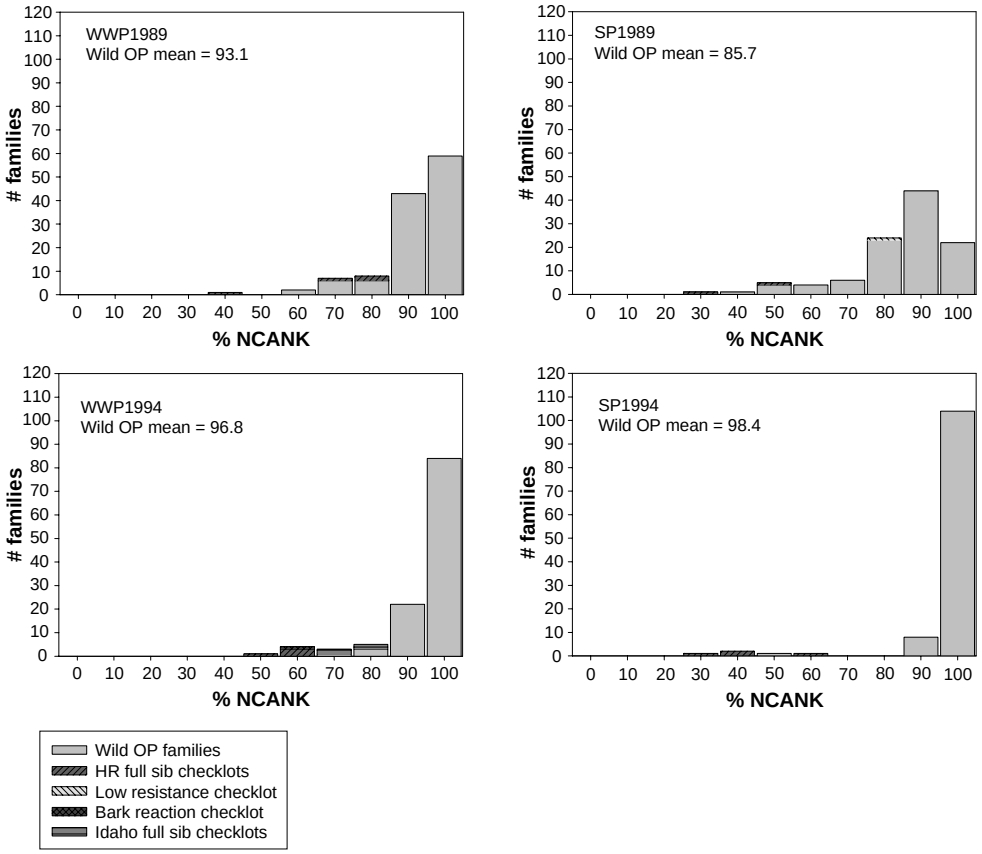

Figure 11—Distribution of family means for the percentage of seedlings with normal cankers (NCANK) in the four trials.

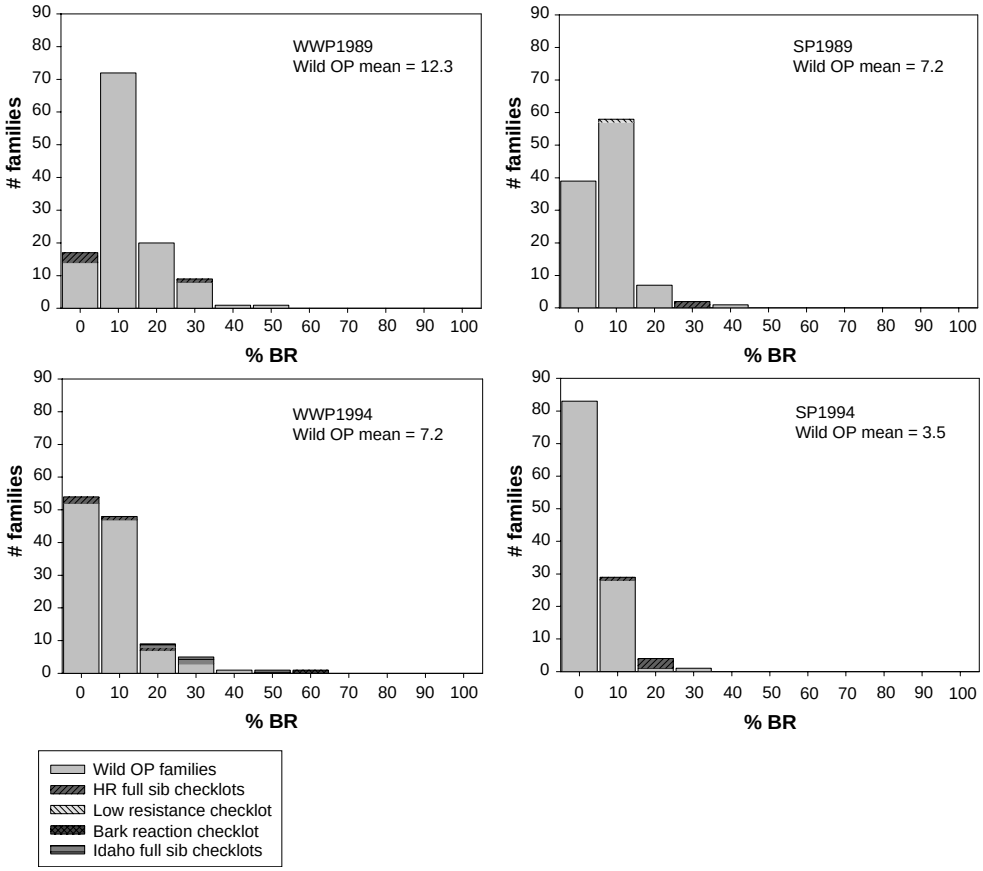

Figure 12—Distribution of family means for the percentage of seedlings with bark reaction (BR) in the four trials.

checklots showed relatively little BR, with a few exceptions (table 5, fig. 12). The Dorena BR checklot, 21105-052 in WWP1994, showed the highest BR percent of any family tested, including the Idaho full-sib checklots (fig. 12, table 5). This family had a high BR percent when first tested as a Wild OP in a 1988 sowing (Sniezko and Kegley 2003). All four of the Idaho checklots in WWP1994 had higher than average BR percent (table 5, fig. 12).

Many of the trees with bark reactions also had normal cankers and died; mean survival 5 years after inoculation of Wild OP seedlings with BR ranged from 1 to 14 percent in the four trials (table 5, fig. 13). Families varied greatly for survival with BR (fig. 13). More of the seedlings with BR in the 1989 trials survived relative to the 1994 trials (table 5), and within a year, slightly more SP survived with BR relative to WWP (table 5).

Early Stem Symptom Percentage (ESS3 Percent)—The average ESS3 for Wild OP families ranged from 58.9 percent (WWP1989) to 84.5 percent (SP1994) (table 5), indicating that over half the seedlings in all four trials showed stem infections approximately 12 months after inoculation. In general the 1989 trials had fewer seedlings with early stem symptoms compared with the 1994 trials. SP1994 showed the highest and least variable family mean ESS3 percent (fig. 14). The trends between species were not consistent in the 2 years (table 5). There were significant differences among Wild OP families in all four trials (table 6). The WWP checklots had families with more delayed onset of stem symptoms (lower ESS3 percent) and included some of the most outstanding families, while the sugar pine checklots were more variable (fig. 14). All four of the Idaho full-sib checklots had among the lowest ESS percent in WWP1994, and the Dorena BR checklot also had relatively few early stem symptoms (table 5, fig. 14).

Survival of Seedlings with Stem Symptoms—Survival of seedlings with stem symptoms (SSAL3) was relatively high (greater than 65 percent) 3 years after inoculation in the 1989 trials but very low in the 1994 trials (table 5). However, by the fifth year after inoculation, many of the seedlings with SS had died; there were few families with high SSAL5 in any trial (table 5). In the 1989 trials SSAL5 of the best families ranged from 21 to 38 percent, but SSAL5 was less than 10 percent in the best Wild OP families in 1994 (table 5). The Dorena HR full sib checklots were mixed in

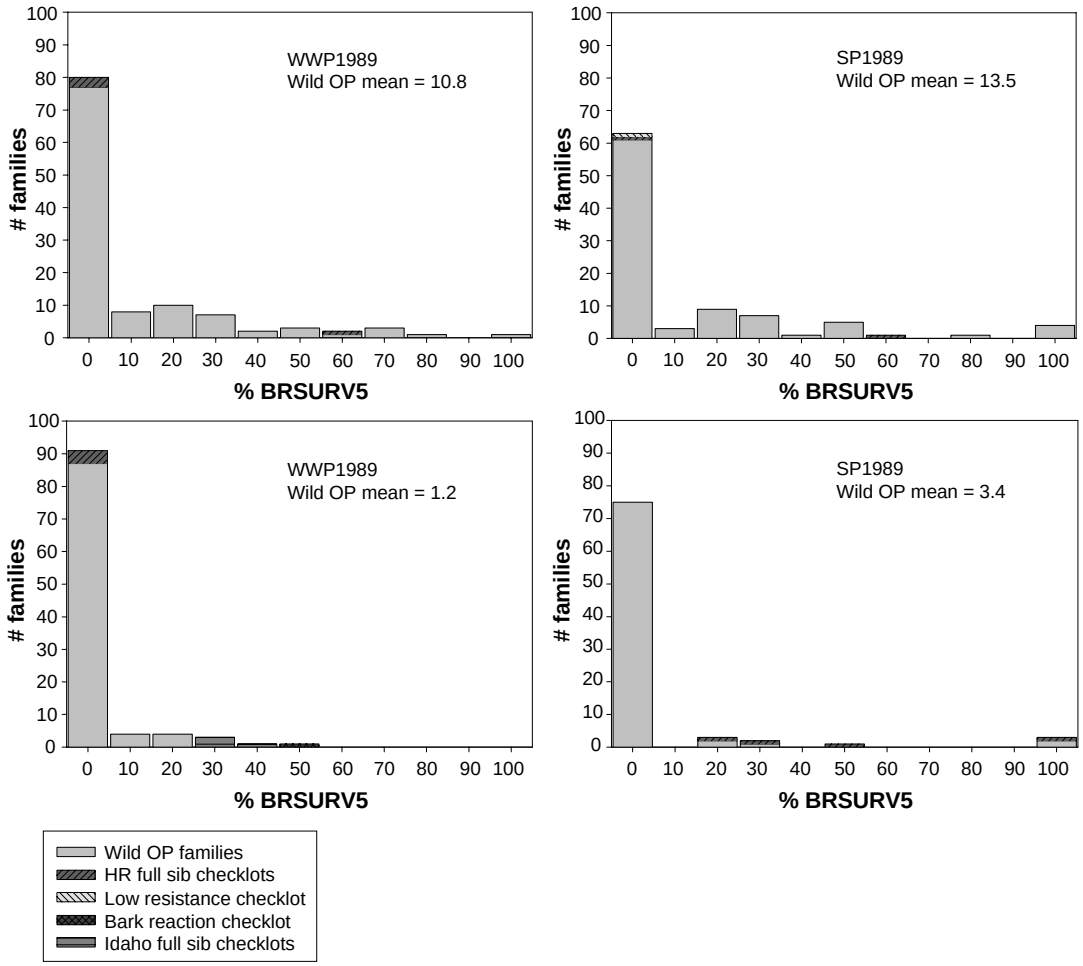

Figure 13—Distribution of family means for percentage of seedlings with bark reaction and surviving 5 years after inoculation (BRSURV5) in the four trials.

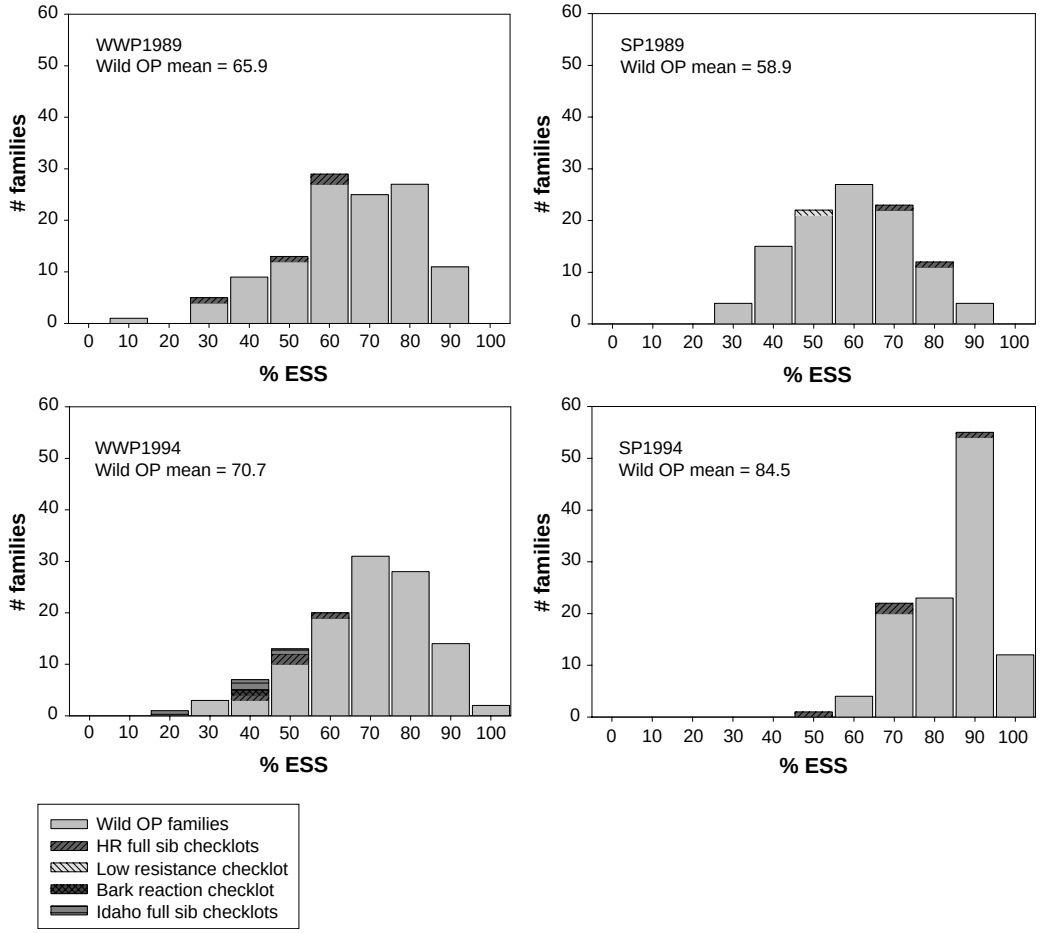

Figure 14—Distribution of family means for the percentage of seedlings with early stem symptoms (ESS3) in the four trials.

performance but generally had low fifth-year survival of seedlings with stem symptoms (SSAL5 percent) (table 5). Two of the full sib HR checklots, SP Family B1054-004 x B1054-034 and WWP Family 15045-862 x 18034-392 were outstanding for SSAL5 in the 1989 trials, and B1054-004 x B1054-034 was also well above the Wild OP mean for SSAL5 in 1994 (table 5). The Dorena BR checklot was well above the Wild OP mean with 35.2 percent SSAL5 in WWP1994 (table 5). Only 3.9 percent of the seedlings in the low resistant sugar pine checklot survived with SS in SP1989 (table 5).

Checklot Performance—More seedlings from Dorena WWP full-sib HR checklots developed stem symptoms (SS percent) in 1989 relative to 1994 (table 5), even though survival of infected seedlings was fairly comparable between the 2 years. In spite of a higher percentage of seedlings with stem symptoms in 1989, those with normal cankers had higher survival than those in 1994 (NCSURV5) (table 5). Mean needle lesion class was higher for the Dorena WWP full sib checklots in 1994 relative to 1989. On average, the Dorena WWP full sib checklots had fewer needle lesions than the Wild OP mean in 1989, while the reverse was true in 1994 (table 5).

For WWP1994, the five Idaho full-sib families generally had higher survival, bark reaction, and survival of seedlings with stem symptoms than the Wild OP families. The Idaho families also had fewer seedlings with stem symptoms and fewer with early stem symptoms than the Wild OP families. The Dorena BR checklot generally had higher levels of resistance responses than the Idaho checklots, and only one of the Idaho families had fewer early stem symptoms (table 5).

Of interest in the WWP1994 trial was whether or not the Idaho full-sib checklots and Wild OP families exhibited a differential reaction when inoculated with a pathotype of rust known to be virulent to HR in WWP. Survival of the infected seedlings from the Dorena and Idaho checklots in WWP1994 was relatively consistent across the first four blocks (in which the pathotype of rust virulent to HR in WWP was excluded), with the Dorena checklots showing higher survival (fig. 15). However, in block 5 and 6 where a pathotype of rust virulent to HR in WWP was present, survival of the four Dorena checkots was dramatically lower, approaching zero in block 6. The Dorena checklot with known bark reaction, however, had higher survival in blocks

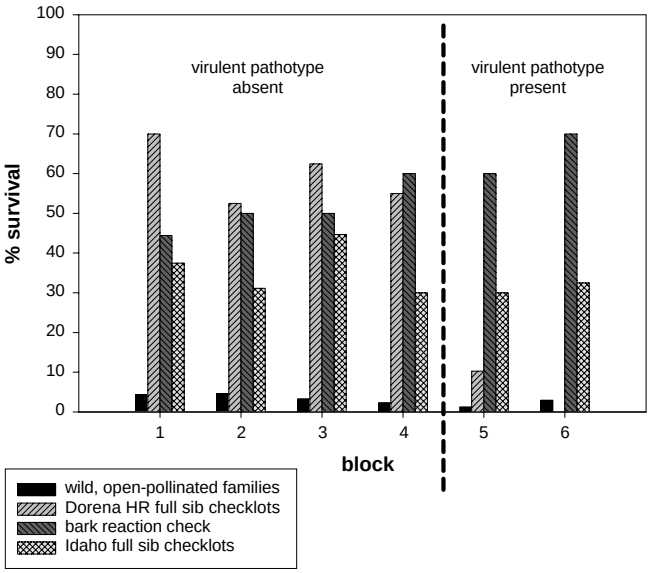

Figure 15—Survival of infected seedlings of several sources of western white pine, by block, in the 1994 trial, with and without a pathotype of rust virulent to HR in western white pine.

5 and 6. There was essentially little or no change in the average survival of the Wild OP families or the Idaho checklots (fig. 15).

SP Checklots—Survival of infected seedlings from the Dorena full-sib checklots was relatively high (37 percent to 68 percent) in both 1989 and 1994 (table 5). The two full-sib checklots common to 1989 and 1994 had similar levels of RSURV5 and SS percent in both trials (table 5). A relatively high proportion (20 percent) of seedlings in the full-sib checklots showed no infection in 1989, and Family B1054-004 x B1054-034 also had a high proportion of uninfected seedlings in 1994 (noinfect, table 5). This family also had a relatively high proportion of seedlings alive with stem symptoms five years after inoculation in both trials (table 5). Bark reaction was higher for the full-sib checklots in 1989 relative to 1994 (table 5, fig. 12). In 1989 mean NLC of the full-sib checklots and the low resistance checklot were close to the run mean of Wild OP families, whereas in 1994, the checklots had fewer than average needle lesions (table 5). In both years, all checklots were below the mean for presence of needle lesions 1 year after inoculation (table 5, fig. 6).

Discussion

Artificial inoculation was successful in infecting more than 95 percent of the 2-year-old seedlings in each trial. This level of infection is probably similar to a high hazard field site but reflects only a single inoculation event. It is unknown whether the use of inoculum in these tests from a wide geographic area increased the infection and mortality levels beyond that of using inoculum from a single geographic source.

SP1989 had 13 percent fewer trees with spots than the other three trials. This is likely attributable to the much lower and more variable inoculum density relative to SP1994 (5,200 spores/cm^2 versus 7,200 spores/cm^2, respectively, table 2). It has been suggested that prevention of needle infection (corresponds to 'no infect' in this paper) is a threshold trait, dependent upon inoculation intensity (Hoff and McDonald 1980a). Results from experiments using different inoculum densities of fusiform rust (*C. quercuum* f. sp. *fusiforme*) on loblolly (*P. taeda* L.) and slash pine (*P. elliottii* Engelmann) indicated that intermediate levels of resistance were more distinguishable at lower inoculum densities (Laird and others 1974). Even when the inoculum density is within the targeted range for WWP or SP, the number of needle lesions per seedling can vary remarkably by trial. Worth noting is that the two SP HR checklots common to these trials had slightly higher percentages of seedlings with stem symptoms in SP1989 even though the mean for the Wild OP families was nearly 8 percent less than in SP1994. Recent data for SP1989 show stem symptoms on trees previously stem symptom free, presumably from natural infections in 1995 and 1997. This had been noted in western white pine; Hunt (1990) reported the presence of stem symptoms on 84 percent of unspotted seedlings in a 1987 inoculation trial. Furthermore, many of those trees without spots had latent canker development (Hunt 1990).

In general, survival provides the ultimate guideline of utility of resistance for immediate use. Five years after inoculation, the mortality in all four trials was high. Only a few families examined here (including checklots) had moderate levels of survival. Both SP and WWP are very susceptible. However, results are dependent on the trial—families, inoculum source, and environmental conditions—and the traits examined. The low survival of progeny from phenotypic selection described here is similar to other reports; Zsuffa (1981) observed very low survival of progeny of resistant eastern white pine (*P. strobus* L.) selections in artificial screening trials (2 percent, family means ranging from 0 to 11.3 percent). Hoff (1984) reported that nearly 90 percent of the cankered western white pine seedlings are dead by the fourth year after inoculation.

One of the major differences between the 1994 and 1989 tests was the lower final survival (total and/or rust-infected survival) in the 1994 tests. The lower survival may have been influenced by the 1994 nursery regime that resulted in the presence of late season lammas growth on both species, and/or a higher number of needle lesions on the trees. Rust infection on lammas growth may circumvent putative resistance mechanisms that prevent stem infection (McDonald and Hoff 1971).

While sugar pine had higher percentages of seedlings without stem symptoms and higher survival relative to western white pine in the 1989 trials, the reverse was true in the 1994 trials. A previous summary of relative blister rust resistance of five-needle pines ranked sugar pine as slightly more susceptible than western white pine (Bingham 1972) as did a field study (Sniezko and others 2000).

The length of the evaluation period after inoculation can have dramatic results on the interpretation of the level of resistance in families (for example, SSAL3 versus SSAL5 for WWP1989 and SP1989). Much of the mortality at Dorena occurs after the third year following inoculations.

The operational program in Idaho terminates formal assessments following the third year after inoculation (Franc 1988). The availability of longer-term data gives the Region 6 program the option to make selections using either the third year or fifth year data.

Trees with normal cankers surviving 5 years after inoculation (a subgroup of SSAL5) are relatively rare for both species. On the surviving trees, some of these cankers are noted to be inactive during the final inspection. Further tracking of the surviving but cankered trees over time would be informative, as large trees with old basal cankers have been observed in the field for both WWP and SP (Hoff 1984; Sniezko and others, this proceedings; Dean Davis, personal comm.) as well as eastern white pine (Hirt 1948).

Bark reaction was present in low frequency in many families and moderate frequency in a few families. Many of the individuals with bark reaction died by the end of the 5-year test period. Survival of seedlings with bark reaction ranged from 1.2 to 13.5 percent in the four trials, but some families had more than 50 percent. Generally trees with complete bark reactions would be expected to live (unless the seedling was girdled or the seedling also had a normal canker). All of the SP resistant checklots had bark reaction levels higher than the Wild OP mean and fewer trees free of stem symptoms than expected. Incompatible or aborted bark reactions have been reported on SP seedlings with HR; these symptoms were associated with infected primary needles (Kinloch and Littlefield 1977; Kinloch and Comstock 1980). It is possible that some of the SP HR seedlings with these atypical symptoms were classified as having stem symptoms.

In examining the Wild OP families for a given trial, there were a few families that approached the levels of survival, bark reaction, or stem-symptom-free of the Dorena or Idaho full-sib checklots. In the 1989 tests, several families had survival levels as high or higher than some of the HR checklots, but in the two 1994 tests, the survival of the Wild OP families was generally much lower than the checklots. In the 1994 tests, SSAL5 levels were very low in the Wild OP families; not many infected trees survived 5 years after inoculation. There may be opportunities for within-family selection for families with a very low incidence for some of the resistant responses.

The Dorena full-sib checklots used in these trials are known to segregate for HR, and they provide linkages between tests. The presence of a pathotype virulent to the HR in WWP (vcr2) notably reduced the expected survival of the Dorena full-sib checklots in WWP1989, as well as in the two blocks of WWP1994 in which the *Ribes* sources with this pathotype were used. In comparison the Idaho full-sib families, the bark reaction checklot, and many Wild OP families in WWP1994 did not show increased levels of stem symptoms when challenged with the virulent pathotype.

Although there was wide variation in family mean needle lesion class (NLC) within each of the four trials, most trees developed stem infections. Individuals with reduced needle lesion frequency (fewer spots) were hypothesized to have fewer stem infections and higher survival (Hoff and McDonald 1980b). However, number of needle lesions in artificial screening has been found to be a poor predictor of cankering in the field (Hunt 2002). In a paired test of high and low spotting individuals within a family planted in the field, the low spotting individuals were as likely to develop stem infections as their highly spotted siblings (Hunt 1990). However, in our trials, there was a trend for the families with higher mean NLC to have a higher percentage early stem symptoms (unpublished data). This has been previously noted with WWP; seedlings with fewer spots generally had fewer cankers 16 to 18 months post-inoculation (Hunt 2002; Meagher and Hunt 1996). Similarly, Hunt (1990) observed that many spot-free WWP seedlings had latent stem symptom development. The implication is that selection for fewer needle lesions may be an indirect selection for delayed stem symptom development.

Several of the resistance responses evaluated here (bark reactions, needle lesion class, latent development of stem symptoms (low ESS3), and stem symptom alive) may be forms of partial resistance. These types of resistances may need several generations of breeding to increase utility for field use. Control crosses among parents with partial resistance traits and testing of their progeny are underway in the Region 6 program. Additionally, field validation of families with complete or partial resistances is in progress (Sniezko and others, this proceedings). Questions remain about whether additional breeding would increase the survival of trees with bark reactions and which of these resistance responses may prove to be more effective in the field as the trees get larger. Many of the families with relatively higher levels of survival expressed more than one resistance response. It is unknown whether these resistances are under the control of genes in tightly linked loci or whether they represent a continuum of response, delaying the expression of disease symptoms or reducing the severity of those symptoms.

At least four mechanisms that lead to complete resistance (lack of stem symptoms) in western white pine have been previously described (Hoff and McDonald 1980a, Hoff and McDonald 1971, McDonald and Hoff 1970, Kinloch and others 1999). There is tentative evidence of a fifth one (unpublished data). One mechanism for complete resistance has been described in sugar pine (Kinloch and others 1970). The hypersensitive reactions in SP and WWP are known to be under the control of separate, single major genes, and premature needle shed and fungicidal reaction in the short shoot are hypothesized to be under the control of separate single recessive genes. However, there may be more than one resistance mechanism or gene underlying a particular phenotypic expression. Differentiating similar phenotypes controlled by different genes will be difficult without virulent strains specific to each, or without the aid of molecular techniques.

The Region 6 program has evaluated blister rust resistance of progeny of thousands of sugar pine and western white pine selections. Results from the trials examined here indicate that progeny of most of these parents are very susceptible. Given the high susceptibility of very young seedlings, natural regeneration may be very unlikely on many sites, exacerbating the decline of these species. Stabilizing and reversing the decline of WWP and SP will be dependent on the development and deployment of resistant material coupled with use of silvicultural tools such as appropriate site selection and pruning. Selections of progeny showing one or more resistance responses have been made and grafts have been established in breeding orchards

and seed orchards. Resistant seed is available for some breeding zones for both western white pine and sugar pine. Advance-generation breeding has started for WWP. Major gene resistance in WWP and SP are likely to play an important role for immediate restoration and reforestation efforts, but partial resistance traits will likely be more important in the future development of durable resistance.

Acknowledgments

The Region 6 Area Geneticists and the many tree improvement workers who made all the phenotypic selections and organized the cone collections. Bob Danchok, the late Delbert Albin, and all members of the Dorena rust screening team for the extensive assessments. Jude Danielson for help in organizing the operational elements of the program. The USFS Region 6 Genetics and Forest Health programs for funding and logistical support, and the Bureau of Land Management and Quinault Indian Nation provided seed from their field selections. Joan Dunlap and Scott Kolpak for their reviews of an earlier draft of this paper.

References

Bingham, R.T. 1972. Taxonomy, crossability, and relative blister rust resistance of 5-needled white pines, p. 271-278. In: R.T. Bingham, R.J. Hoff, and G.I. McDonald (eds.) Biology of Rust Resistance in Forest Trees: Proceedings of a NATO-IUFRO Advanced Study Institute. USDA Forest Service Misc. Publ. 1221.

Bingham, R.T. 1983. Blister rust resistant western white pine for the Inland Empire: the story of the first 25 years of the research and development program. USDA Forest Service General Technical Report INT-146. 45 p.

Campbell, R.K. and Sugano, A.I. 1987. Seed zones and breeding zones for sugar pine in southwestern Oregon. USDA Forest Service Research Paper PNW-RP-383. 18 p.

Campbell, R.K. and Sugano, A.I. 1989. Seed zones and breeding zones for white pine in the Cascade Range of Washington and Oregon. USDA Forest Service Research Paper PNW-RP-407.

Cochran, W.G. and Cox, G.M. 1967. Experimental designs, 2nd edition. John Wiley and Sons, Inc. New York. 611 p.

Franc, G.C. 1988. The white pine program in the northern region, p. 21-26. In: R.S. Hunt, ed. Proc. of a western white pine management symposium. Nakusp, BC. Pacific Forestry Centre, Victoria, BC.

Hirt, R.R. 1948. Evidence of resistance to blister rust by eastern white pine growing in the northeast. *J. For.* 46:911-913.

Hoff, R.J. 1984. Resistance to *Cronartium ribicola* in *Pinus monticola*: Higher Survival of Infected Trees. USDA Forest Service Research Note INT-343, 6p.

Hoff, R.J. and McDonald, G.I. 1980a. Improving rust-resistant strains of inland western white pine. USDA Forest Service Research Paper INT-245. 13 p.

Hoff, R.J. and McDonald, G.I. 1980b. Resistance to *Cronartium ribicola* in *Pinus monticola*: reduced needle-spot frequency. *Can. J. Bot*. 58:574-577.

Hoff, R.J. and McDonald, G.I. 1971. Resistance to *Cronartium ribicola* in *Pinus monticol*a: short shoot fungicidal reaction. *Can. J. Bot*. 49:1235-1239.

Hunt, R.S. 1990. Blister rust in inoculated and plantation-tested western white pine in British Columbia. *Can. J. For. Res.* 12:279-282.

Hunt, R.S. 1997. Relative value of slow-canker growth and bark reactions as resistance responses to white pine blister rust. *Can. J. Plant Pathol*. 19:352-357.

Hunt, R.S. 2002. Relationship between early family-selection traits and natural blister rust cankering in western white pine. *Can. J. Plant Path.* 24:200-204.

Kim, H.J., Yi, C.K., and La, Y.J. 1982. Present status of Korean pine (*Pinus koraiensis*) blister rust and relative resistance of three species of five-needled pines to *Cronartium ribicola*. Research Report of Forest Research Institute, Seoul, Korea. 29:239-252.

Kinloch, B.B., Jr. and Comstock, M. 1980. Cotyledon test for major gene resistance to white pine blister rust in sugar pine. *Can. J. Bot.* 58: 1912-1914.

Kinloch, B.B. and Davis, D. 1996. Mechanisms and inheritance of resistance to blister rust in sugar pine, p. 125-132. In: B.B. Kinloch, M. Marosy, and M.E. Huddleston (eds.). Sugar pine: status, values, and roles in ecosystems: Proceedings of a symposium presented by the California Sugar Pine Management Committee. Univ. Calif. Div. Agr. Res. Publ. 3362.

Kinloch, B.B., Jr. and Dupper, G.E. 2002. Genetic specificity in the white pine-blister rust pathosystem. *Phytopathology* 92:278-280.

Kinloch, B.B., Jr. and Littlefield, J.L. 1977. Whtie pine blister rust: hypersensitive resistance in sugar pine. *Can. J. Bot.* 55:1148-1155.

Kinloch, B.B., Jr., Parks, G.K., and Fowler, C.W. 1970. White pine blister rust: simply inherited resistance in sugar pine. *Can. J. Bot*. 58:1912-1914.

Kinloch, B.B., Sniezko, R.A., Barnes, G.D., and Greathouse, T. E. 1999. A major gene for resistance to white pine blister rust in western white pine from the western Cascade Range. *Phytopathology* 89:861-867.

Laird, P.P., Knighten, J.L., and Wolfe, R.L. 1974. Reduced inoculum density enhances sensitivity of resistance detection in fusiform rust resistance testing. USDA Forest Service Southeastern Area, State and Private Forestry, Division of Forest Pest Management Report 74-1-10. 10 p.

McDonald, G.I. and Hoff, R.J. 1970. Resistance to *Cronartium ribicola* in *Pinus monticola*: early shedding of infected needles. USDA Forest Service, Res. Note INT 124. 8 p.

McDonald, G.I. and Hoff, R.J. 1971. Resistance to *Cronartium ribicola* in *Pinus monticola*: genetic control of needle-spots-only resistance factors. *Can. J. For. Res*. 1:197-202.

Meagher, M.D. and Hunt, R.S. 1996. Heritability and gain of reduced spotting vs. blister rust on western white pine in British Columbia, Canada. *Silv. Gen.* 45(2-3)75-81.

Samman, S.A. 1982. The white pine blister rust program of Region 6, p. 133-183. in Breeding insect and disease resistant forest trees: Proceedings, Servicewide genetics workshop, Eugene, Oregon. USDA Forest Service.

Samman, S. and Kitzmiller, J.H. 1996. The sugar pine program for development of resistance to blister rust in the Pacific Southwest region, p. 162-170. In: B.B. Kinloch, M. Marosy, and M.E. Huddleston (eds.). Sugar pine: Status, values, and roles in ecosystems: Proceedings of a symposium presented by the California Sugar Pine Management Committee. Univ. Calif. Davis. Agr. Natural Resources Publ. 3362.

SAS Institute Inc. 1999. SAS OnlineDoc, Version 8. Cary, NC: SAS Institute Inc.

Sniezko, R.S. 1996. Developing resistance to white pine blister rust in sugar pine in Oregon, p. 125-132. In: B.B. Kinloch, M. Marosy, and M.E. Huddleston (eds.). Sugar pine: Status, values, and roles in ecosystems: Proceedings of a symposium presented by the California Sugar Pine Management Committee. Univ. Calif. Davis. Agr. Natural Resources Publ. 3362.

Sniezko, R.A., Bower, A., and Danielson, J. 2000. A comparison of early field results of white pine blister rust resistance in sugar pine and western white pine. *HortTechnology* 10(3): 519-522.

Sniezko, R.A. and Kegley, A.J. 2003. Blister rust resistance of five-needle pines in Oregon and Washington. Proceedings of Second IUFRO Rust of Forest Trees Working Party Conference. 19-23 August 2002. Yangling, China. Forest Research 16 (Suppl.): 101-112.

Theisen, P.A. 1988. White pine blister rust resistance mechanisms of sugar and western white pines. USDA Forest Service Region 6 Tree Improvement paper 13. 24 p.

Zolman, J.F. 1993. Biostatistics: experimental design and statistical inference. New York, Oxford University Press. 343 p.

Zsuffa, L. 1981. Experiences in breeding *P. strobus* L. for resistance to blister rust. Proc. 18th IUFRO World Cong. Kyoto, Japan. P 181-183.