

SURVIVAL AND WHITE PINE BLISTER RUST INFECTION IN
A MINNESOTA WHITE PINE SCREENING STUDY¹

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Abstract. Survival and rate of non-infection by the white pine blister rust fungus was measured on trees representing 456 and 418 mother tree collections from northeastern Minnesota. These materials had completed 13 and 11 growing seasons respectively in the field at the time measurements were taken. Significant differences among families were found for survival but not for rate of non-infection. Families originating from the north shore of Lake Superior had significantly greater survival than families originating away from the lake.

Additional keywords: *Cronartium ribicola*, *Pinus strobus*, Disease Resistance, Disease Tolerance.

INTRODUCTION

In 1966 a cooperative project was initiated to screen eastern white pine families for resistance to white pine blister rust by the Quetico-Superior Wilderness Research Center, The U.S. Forest Service and the College of Forestry, University of Minnesota, under the direction of Scott Pauley and Clifford E. Ahlgren. The principle objectives of this project were to screen a large number of selections from the high rust infection areas of northeastern Minnesota and to provide basic information regarding genetic variation in the white pine of the region. Materials identified as resistant to the blister rust fungus were to be incorporated into applied breeding projects involving white pine. This report reviews the establishment of the project and presents results from the first systematic evaluation of the blister rust screening test.

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MATERIAL AND METHODS

Mother Tree Selection

The blister rust fungus was introduced to northeastern Minnesota in approximately 1916. Mother trees for this study were identified in this region beginning in 1966. White pine trees in age class 45 or less, with an adequate seed crop were selected. Trees this age and younger were assumed to have survived exposure to the fungus as a seedling. Additionally, older trees showing evidence of having survived infection were also selected. Seed collections were conducted in 1966, 1967, and 1969, primarily by C.E. Ahlgren. By the end of the 1969 season, 1180 separate mother tree collections had been made.

Plantation Establishment

The test is located on a 22 acre site in a high infection area on the Tofte District in the Superior National Forest. The site was control-burned in 1969 to eliminate a heavy sod cover, and disked in 1971 to expose a mineral soil planting site. Plants from the mother tree seedlots were propagated by the U.S. Forest Service at the Eveleth, Minnesota forest tree nursery. The seedlings were grown to age 2-1 (two years in the nursery and one year in a transplant bed) before being transplanted to the test site. Due to the size of the project, the seedlots were divided into two blocks of approximately 500 open-pollinated families each. The first block was field planted in the spring of 1972 and the second block was planted into the field in the spring of 1974.

The seedlings were established in two separate contiguous plantings. Within each planting, a randomized complete block design with 5 replications of 10 tree row-plots was used. An initial spacing of 5 X 5 feet was used in anticipation of early mortality. To insure adequate levels of inoculum, infected plants from native and introduced *Ribes* species were inter-planted among the rows of white pine when the plantation was established. The area around each white pine seedling was treated with Simazine immediately after planting. No other weed control was undertaken during the establishment period. To protect the test from deer and moose damage, an eight foot high fence was erected surrounding the study.

Measurements

In 1984, a systematic evaluation of the screening test was undertaken. The number of surviving stems and the number of non-infected stems were recorded for each plot. A non-infected stem was defined as a stem displaying no evidence of previous infection by the white pine blister rust fungus. Field measurements were begun in the fall of 1984 and completed in 1985.

Analysis

Age and family differences between the 1972 and 1974 materials necessitated separate analysis for the two plantings. Errors made during planting resulted in the exclusion of data from 21 families from the analyses.

Mother trees were grouped into regions of origin based on seed collection records (Table 1, Figure 1). The close proximity of some regions renders a detailed analysis of regional effect meaningless. However, most selections were made in two broad geographic areas; The Boundary Waters Canoe Area (Regions 3,4,5,6,7,8,9,10,21,22,23) and the North Shore of Lake Superior (Regions 1,2,11,13,14,15,16,17,18,19). A single degree of freedom contrast was used to compare the Boundary Waters Canoe Area and the North Shore. Although materials from five regions are present in both the 1972 and 1974 plantings, these regions are represented by different families in each planting. No family was grown in both years.

Survival and non-infection were tested for lack of independence with region and family using the likelihood ratio Chi-square procedure (Skoal and Rohlf, 1969). This procedure has been recommended in instances when the expected counts per cells are less than 5. A value of 0.1 was substituted for each plot with no surviving or non-infected stems to adjust cells with an actual count of zero.

Regional differences were analyzed for traits indicating lack of independence with family using standard analysis of Variance techniques.

RESULTS

The summary of survival and non-infection for both plantings is given in Table 2. The overall survival for the material planted in 1972 was 38 percent. Ninety seven percent of the surviving stems were infected. Overall, one percent of the stems planted survived and were non-infected. The survival for the 1974 planting was 41 percent. Ninety-nine percent of the surviving stems were infected. Of the 20,829 stems planted, only 118 (0.5 percent) survived and were non-infected.

Table 2. Summary Averages of Survival And Non-Infection For Both Plantings

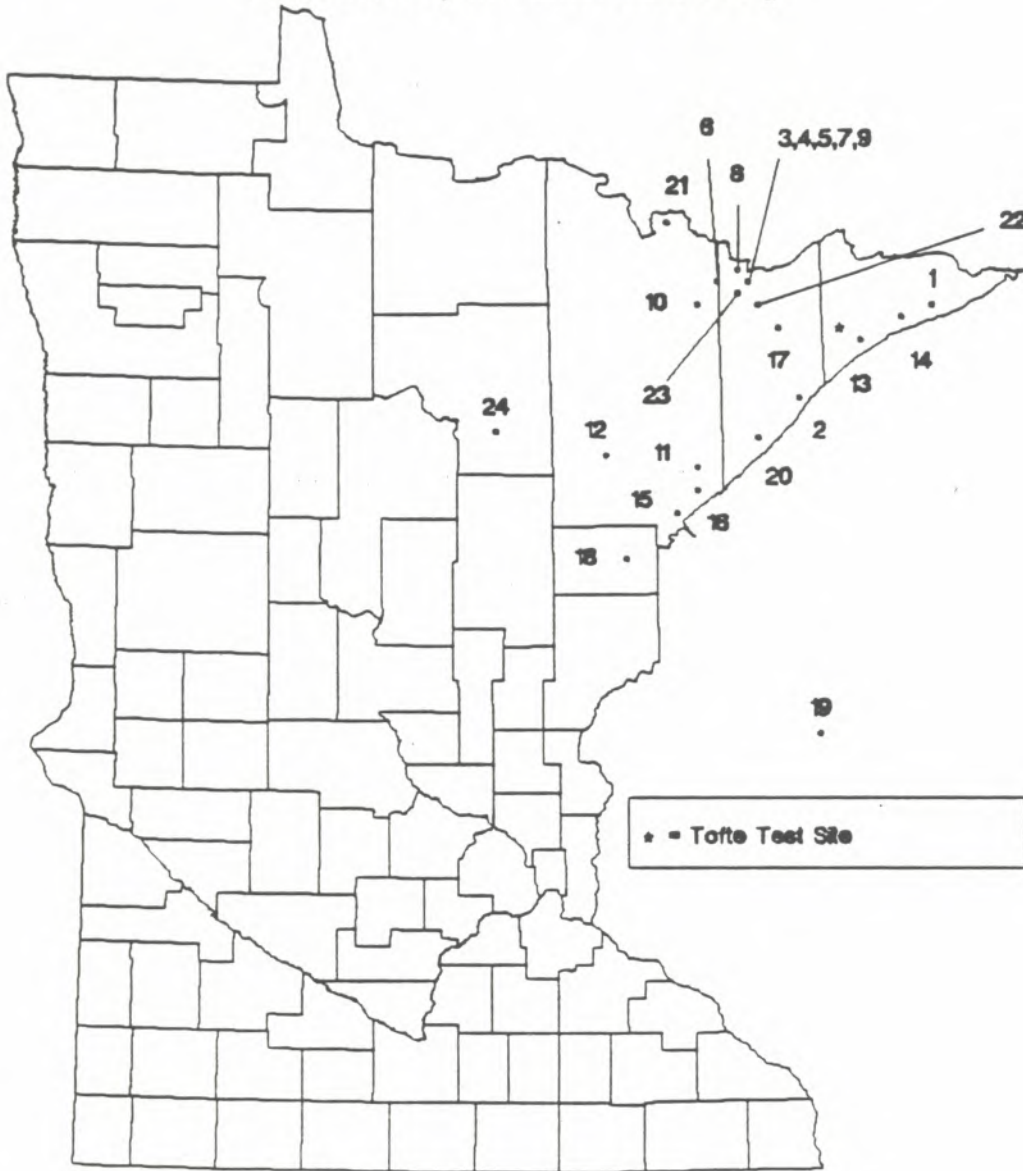
<u>Trait</u>	<u>1972</u> <u>(%)</u>	<u>1974</u> <u>(%)</u>
Overall		
% Survival	38	41
% Non-Infection	1	0.5
Family Range		
% Survival	2 - 78	10 - 74
% Non-Infection	0 - 14	0 - 6

Table 1.

Number of Families, Average Survival, And Non-Infection For The Different Geographic

Region	Location	<u>Regions Present In The Two Plantings</u>					
		1972			1974		
		Number Families	Survival (%)	Non-Infection (%)	Number Families	Survival (%)	Non-Infection (%)
1	Northern Lights Hill	14	47.0	1.00			
2	Beaver Bay	26	46.6	2.01			
3	Basswood Lake	7	31.0	0.88	18	35.1	0.11
4	Q-S Plantation	23	32.9	0.87			
5	Frog Bay	25	30.1	0.40			
6	Back Bay	11	38.5	0.18			
7	Boskos Point	41	30.7	0.54			
8	Kings Point, Canada	17	30.1	0.36			
9	Grave Yard Island	10	26.7	0.20			
10	Burntside State Forest	28	24.3	0.29			
11	Thompson Lake	85	43.7	1.09			
12	Meadowlands	28	43.5	1.01			
13	Tofte District	37	37.4	0.94	16	40.7	0.13
14	Grand Marais District	40	40.2	1.87	52	42.9	0.91
15	Duluth, Minnesota				1	50.0	2.00
16	Wilson Farm	25	41.2	1.87			
17	Isabella District	38	36.9	0.58	86	44.6	0.65
18	Scanlon Carlton Thompson Area	1	41.0	0.00	42	38.0	0.33
19	Sawyer County, Wisconsin				13	39.2	0.15
20	Two Harbors District				81	44.5	0.77
21	Echo Trail Laclacroix Dist.				37	33.2	0.33
22	Halfway District				40	42.6	0.65
23	Winton, Minnesota				3	44.0	0.00
24	Grand Rapids, Minnesota				28	35.5	0.36
Total		456	37.6	0.97	418	41.2	0.5

Figure 1. Location of mother tree regions
Tofte white pine blister rust study



Family mean survival in the 1972 planting ranged from 2-78 percent (Figure 2) whereas in the 1974 planting, survival ranged from 10-74 percent (Figure 3). Family non-infection rate ranged from 0-14 percent in the 1972 planting (Figure 4) , and 326 (71%) of the families had no non-infected individuals. In the 1974 planting (Figure 5) , the range in non-infected stems was from 0-6 percent with 320 (77%) families having no non-infected individuals.

Likelihood ratio Chi-squared analysis indicated survival was not independent of either family or regional effects in 1972 and 1974 (Table 3). Non-infection rate was independent of both family and regional effects in the 1974 planting. The 1972 non-infection rates were shown to be independent of family effects, but not independent of regional effects.

Table 3. Likelihood Ratio Chi-Square Values and Associated Probability Values From Test Of Independence for Survival and Non-Infection

1972	Likelihood Chi-Square	df	P-Value
Survival			
Region	413.2	16	<0.001
Family	2131.8	455	<0.001
Rep	301.7	4	<0.001
Non-Infection			
Region	30.2	16	0.017
Family	281.1	455	1.000
Rep	6.0	4	0.196
1974			
Survival			
Region	136.7	11	<0.001
Family	1254.8	417	<0.001
Rep	202.4	4	<0.001
Non-Infection			
Region	7.8	11	0.727
Family	111.8	417	1.000
Rep	6.2	4	0.185

Figure 2. Distribution of 1972 family survival

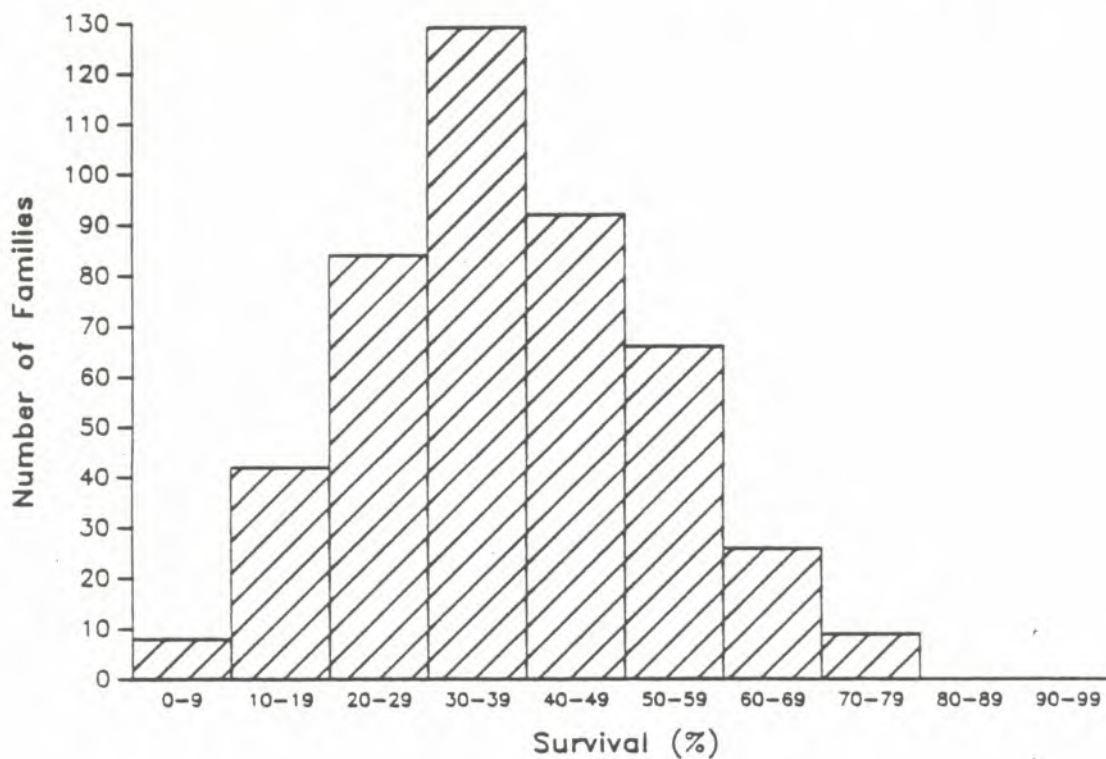


Figure 3. Distribution of 1974 family survival

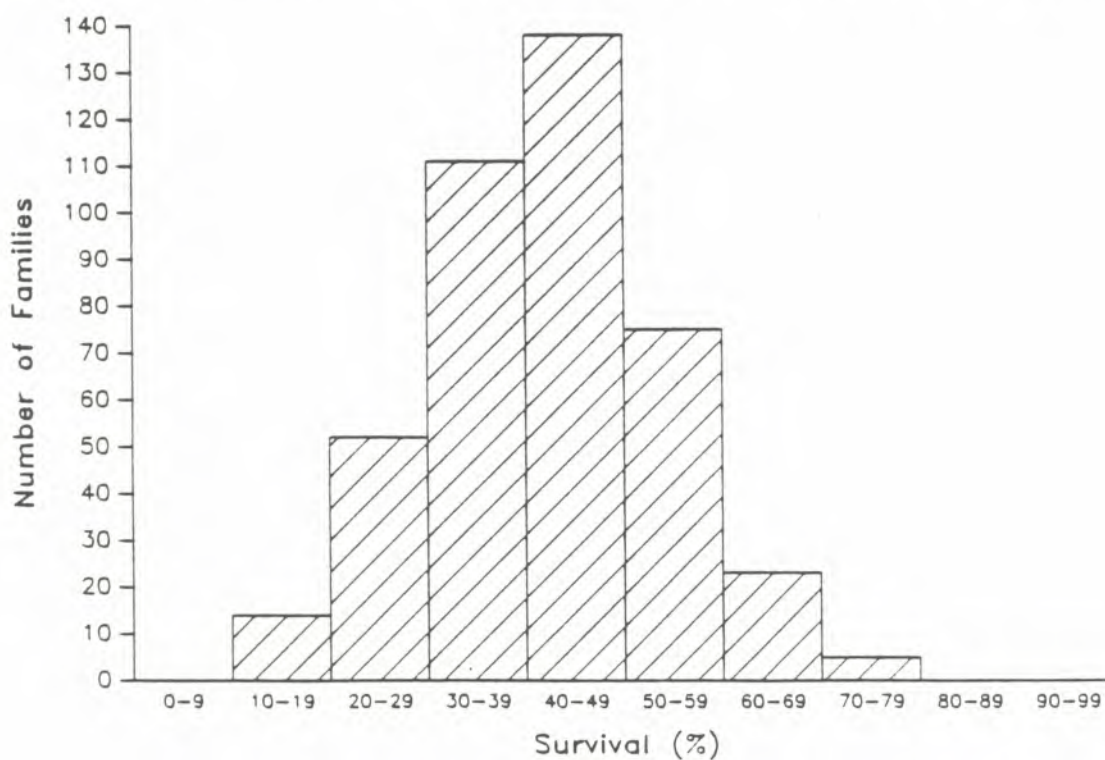


Figure 4. Distribution of 1972 family noninfection rate

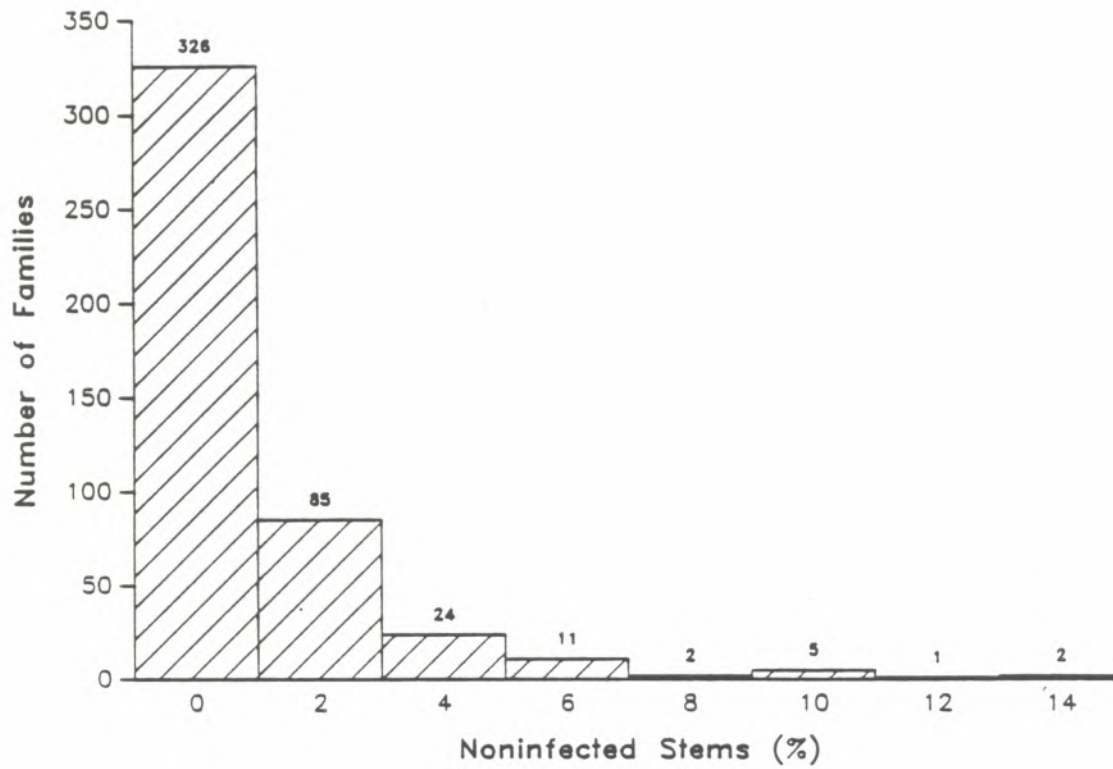
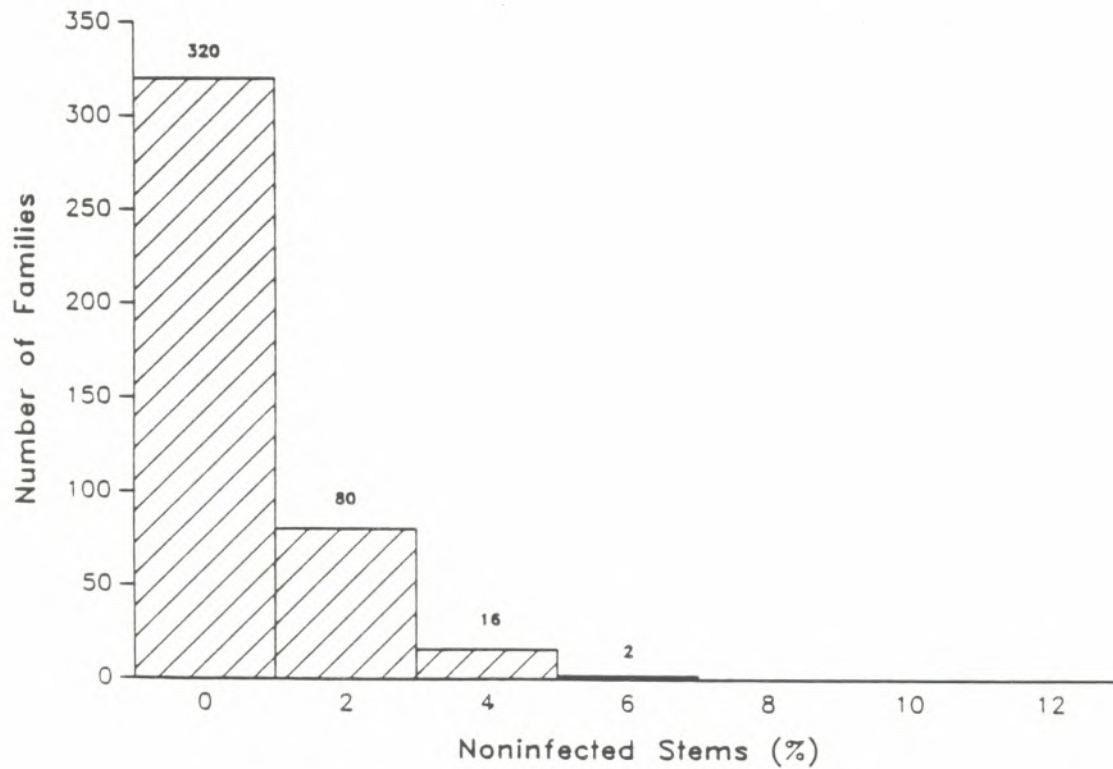


Figure 5. Distribution of 1974 family noninfection rate



The ANOVA table for survival (Tables 4 & 5) indicates that there was a significant difference among families originating from the North Shore, and those originating from the Boundary Waters in the 1972 planting. The mean survival from the North Shore was 41.5% and the mean survival of families from the Boundary Waters was 30.0% (Figure 6). Significant differences among families from these areas were not found in the 1974 materials. In 1974, the mean survival from the North Shore was 43.1% and the mean survival of families from the Boundary Waters was 37.7% (Figure 7).

DISCUSSION

Overall, survival was quite low for both the 1972 and 1974 plantings. Initial survival was in the range of 97-100 percent (based on estimates made by C.E. Ahlgren). It is reasonable to assume a majority of the subsequent mortality was caused by the blister rust fungus. This and the high infection rate among surviving stems indicates this test was very effective at exposing the progenies to high levels of inoculum.

Statistically significant differences were found for survival among families and regions for both plantings. Because the materials in this test originated from the same geographic area, survival differences attributable to poor adaptation seem unlikely. The geographic effect associated with survival in the 1972 planting suggests materials from the North Shore are more tolerant of infection by the blister rust fungus.

The causes of these geographic differences is open to speculation. There is a pronounced lake effect along the North Shore which results in cooler summers and milder winters. The region warms more slowly in the spring and cools later in the fall. There may be some phenologic differences between trees originating from the North Shore and other areas in northeastern Minnesota that lead to differences in survival.

The use of a single test site located on the North Shore may be another possible cause of the geographic differences in survival. There may be different races of the fungus that predominate in different regions of the state, and the families originating on the North Shore may be better adapted to the local race. Whatever the cause of these differences, it does appear that concentrating white pine selection on the North Shore will increase the tolerance to white pine blister rust in the selected population.

No differences were found among families for rate of non-infection. There appears to be some resistance to the fungus in the population but only at very low levels. The observed pattern of variation for non-infection is similar to what might be expected for a qualitative trait governed by one or two recessive genes occurring at low frequency in the population. Currently the

Table 4. Analysis of Variance For Survival - 1972 Planting

Source	df	SS	MS	F	P
Region	16	96637.1504	6039.8219	6.81	<0.001
Family (Region)	439	389505.9300	887.2572	2.10	<0.001
Rep	4	71717.6177	17929.4044		
Rep x Region	64	43484.8146	679.4502		
Error	1756	743225.3928	423.2491		
Total	2279	1344570.9047			
Contrast					
BWCA vs NS	1	22418.5043	22418.5043	25.27	<0.001

Table 5. Analysis of Variance For Survival - 1974 Planting

Source	df	SS	MS	F	P
Region	11	33249.9819	3022.7256	4.69	<0.001
Family (Region)	406	261707.3560	644.5994	1.66	<0.001
Rep	4	49359.4270	12339.8568		
Rep X Region	44	24337.3982	555.1227		
Error	1624	630488.2427	388.2317		
Total	2089	999142.0057			
Contrast					
BWCA vs. NS	1	1899.3815	1899.3815	2.95	0.087

Figure 6. Distribution of 1972 Regional Mean Survival

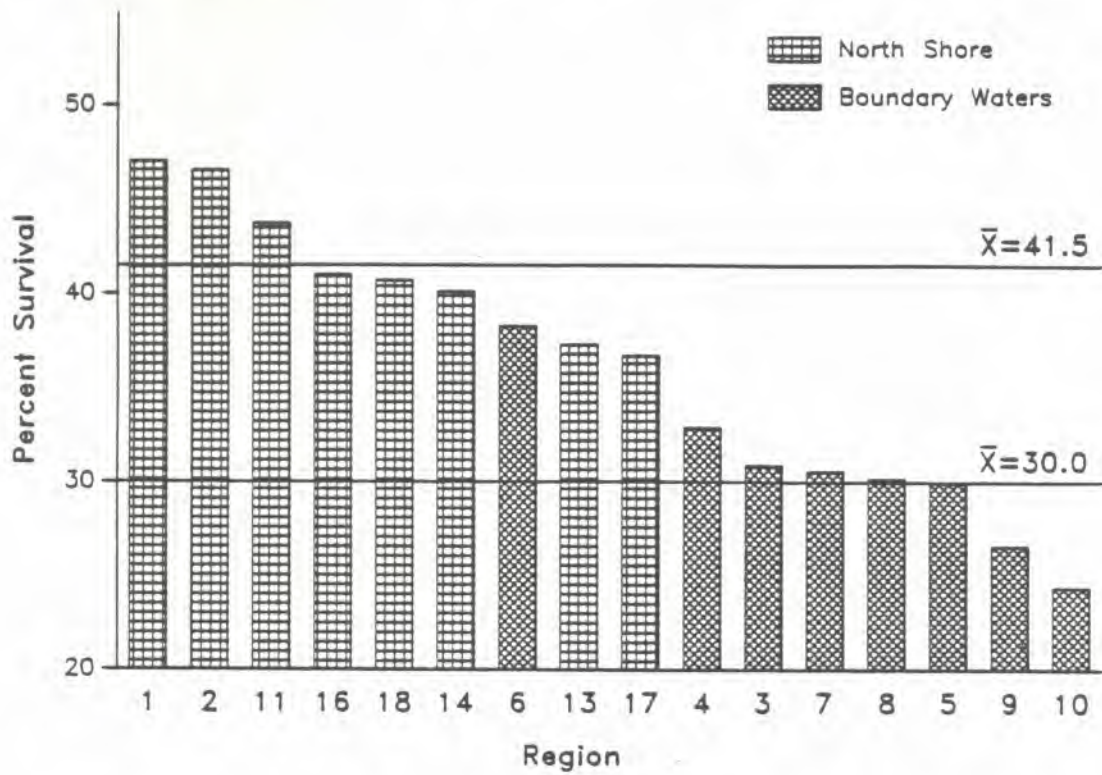
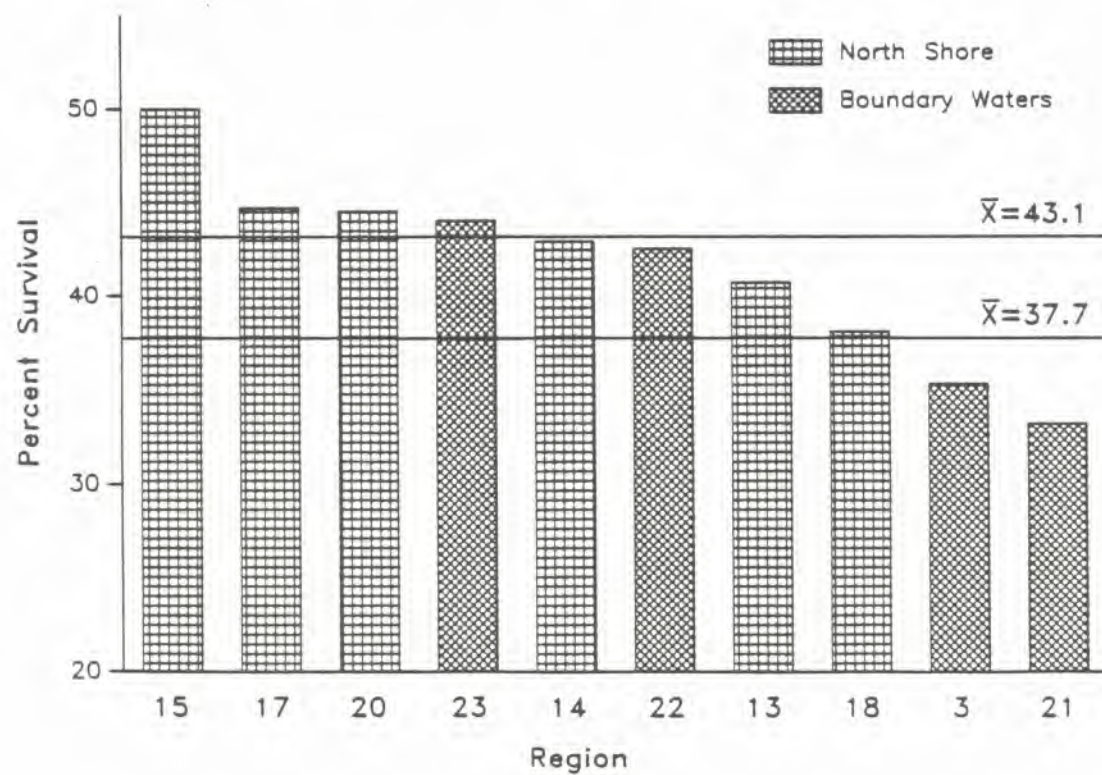


Figure 7. Distribution of 1974 Regional Mean Survival



non-infected materials from this test are being grafted into a breeding arboretum at the University of Minnesota. These materials will be retested to substantiate the presence of resistance. Depending upon the outcome of these evaluations, studies on the mode of inheritance of resistance will be undertaken using either the individuals at Tofte or the grafts.

Comparison of 1972 and 1974 Results

The lower survival of the 1972 material may simply reflect its longer exposure to the rust fungus. The distribution of family mean survival was broader and flatter in 1972 than in 1974, suggesting an age effect. There is a high percentage of severely infected stems in the 1974 planting that will undoubtedly succumb to the fungus in the next several years. It is likely that survival will continue to decrease in this test probably more rapidly in the 1974 planting.

The geographic effect associated with survival in 1972 was not found in the 1974 material. The lack of a geographic effect for 1974 may also reflect the age difference between the two plantings. As more mortality occurs in the 1974 planting, geographic differences may appear.

The higher non-infection rate in the 1972 planting may be due to a number of factors. The inoculated Ribes plants were established at the same time as the 1972 planting. There probably was more inoculum present when the 1974 materials were established than in 1972. Because there is an age effect associated with resistance to the blister rust fungus, (Patton 1961) the difference in non-infected individuals between the plantings may have resulted from the 1974 material being exposed to a greater amount of inoculum at a more susceptible age than the 1972 planting.

Another possible explanation for the difference in non-infected rates also involves the age difference between the two plantings. Some of the surviving stems in the 1972 planting had reached heights in excess of 20 feet. In this portion of the study, the canopy has closed and some lower branches have been shed. It is likely that some of the trees in the 1972 planting scored as non-infected were previously infected, but the evidence of infection was lost when the branch was shed. In a similar manner, some of the infected 1974 trees may be non-infected in the future.

It has been indicated several times that age differences between the two plantings produced some difference in observed response. This is an important point especially considering the emphasis on early screening for disease resistance. A number of resistance factors such as inactivation of cankers (Hungerford, 1972), formation of wound periderm (Struckmeyer and Riker, 1951), and long term tolerance of the rust fungus can not currently be evaluated adequately at the seedling stage.

From a forestry standpoint, we are more interested in producing a stand of trees reaching rotation age without significant loss caused by the fungus than we are in growing completely non-infected trees. Early evaluation of disease resistance may eliminate some genotypes susceptible to, but tolerant of, the fungus. In this situation, it could be desirable to postpone selection until some of these other factors can be evaluated.

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