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Proceedings of the Conference

Whitebark Pine: A Pacific Coast Perspective

August 27-31, 2006

Ashland, Oregon



Crater Lake Sentinel

Photo by Sandra J. Kegley

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Proceedings of the Conference
Whitebark Pine:
A Pacific Coast Perspective

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Ellen Michaels Goheen and Richard A. Sniezko,
Technical Coordinators

Conference Sponsors

Crater Lake Institute

Crater Lake Natural History Association

Southern Oregon University

USDA Forest Service

USDI National Park Service

Whitebark Pine Ecosystem Foundation

Abstract

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The Conference Whitebark Pine: A Pacific Coast Perspective provided a forum for current research on whitebark pine and other high elevation five-needle pine species with emphasis on the ecology, population structure and genetics, wildlife interactions, disturbance regimes, threats, and challenges of managing these five-needle pines in British Columbia, Washington, Oregon, and California. Papers or abstracts from 38 oral or poster presentations are included.

Keywords: five-needle pines, whitebark pine, *Pinus albicaulis*, white pine blister rust, *Cronartium ribicola*, mountain pine beetle, *Dendroctonus ponderosae*

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Whitebark Pine and Other High Elevation Five-needle Pines in Pacific Coast Ecosystems: A Conference Preface

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Whitebark pine, a hardy, sub-alpine conifer is considered a keystone species throughout its range for its contributions to high elevation ecosystems including its value as a food source for wildlife, its ability to survive harsh environmental conditions, its effect of regulating snowmelt and reducing soil erosion, and for its aesthetic and symbolic values in the high mountains of western North America.

Much has been written about whitebark pine in the Rocky Mountains but comparatively little information has been compiled for whitebark pine along the Pacific Coast. There is, however, substantial interest in the Pacific Coast high elevation ecosystems. From the Coast Range of British Columbia, down through the Cascade Mountains of Washington, Oregon and Northern California, and south through the Sierra Mountains of California and into Mexico, several species of five-needle pines are critical components of alpine and subalpine forests. They are highly-valued for their aesthetic contribution to National Parks and Wilderness areas. High elevation five-needle pines contribute critical elements of habitat for many wildlife species. White pine blister rust, mountain pine beetle, and fire exclusion are having negative effects on these species and are influencing associated ecosystem process and function. The potential influence of climate change is of concern.

Based on these growing concerns for the health of whitebark pine ecosystems in Oregon and Washington, the USDA Forest Service, Pacific Northwest Region, USDI National Parks and others began a more concerted effort to gather information on the status of whitebark pine in Oregon and Washington. Early work included an informal information survey, seed collections to be used to examine natural genetic resistance to white pine blister rust and common garden studies to examine genetic variation, germination tests to examine longevity of seed in cold storage and germination procedures, protocols to grow whitebark pine seedlings, and surveys of the health of whitebark pine ecosystems. Beginning in 2004, this work intensified and a four year '*Pacific Northwest Albicaulis Project*' was initiated. <http://www.fs.fed.us/r6/genetics/publications/albicaulis-project>. During this period studies were also being undertaken in California and British Columbia.

In October 2005, a whitebark pine workshop held at Crater Lake National Park brought together a small group of biologists, geneticists, ecologists, entomologists, and pathologists

from California, Oregon, Washington and British Columbia. The workshop was an opportunity for participants working in Pacific Coast high elevation ecosystems to share information; discuss issues, and concerns; and to help develop a strategy to maintain and restore whitebark and other high elevation five-needle pines in our region. After presentations by Dr. Frank Lang, Emeritus Professor of Biology at Southern Oregon University on the ecology and biology of Pacific Coast high elevation five-needle pines and Dr. Diana Tomback, University of Colorado and the Whitebark Pine Ecosystem Foundation on the concerns and status of whitebark pine in the Rocky Mountains, participants engaged in more focused discussions about the questions that need to be answered regarding these species. Of key concern were issues such as:

Surveys and Data

- The status of the whitebark pines and other high elevation five-needle pine species: Are they healthy, are they reproducing?
- What is the distribution of whitebark pine on the landscape?
- Do we have/use standardized survey methods for blister rust?
- Should we have/use standardized reporting/outputs?

Genetics

- The need to develop a strategy to sample populations for genetic variation
- Long-term seed storage bank for gene conservation plus viability requirements for long-term seed storage
- Establishing Common Garden studies
- Prioritizing work areas/zones based on current status/risk
- Strategy for gene conservation throughout range
- Gene conservation seed collections (seed banking/screening)
- What is the level of white pine blister rust resistance in our native conifers?
- Field validation of resistance
- Climate change- will there be an effect on genetic variability? How many populations may be extirpated?
- Easy/economically feasible method to locate resistance (i.e. markers to complement traditional screening program)

Wildlife Interactions

- The need for surveys on populations of nutcrackers
- How can we provide for the disturbances nutcrackers need for seed caching?
- Impact of wildfire & prescribed fire on habitat of key species in these ecosystems?
- Will pathogens such as west nile virus have a significant impact on nutcracker populations?
- How to get regulatory agencies involved earlier to remove real or perceived barriers in whitebark pine restoration because of listed species?

Insects and Pathogens

- White pine blister rust/mountain pine beetle interaction – how well are they characterized?
- Do we understand host pathogen interactions in these high elevation ecosystems? Where does inoculum come from? What influences host susceptibility to rust or bark beetles?
- What sites are high, moderate, and low rust hazard?
- How may climate change affect rust epidemiology?
- What silvicultural tools are available for managing bark beetles and white pine blister rust?
- How/where/under what circumstances should Verbenone treatments be used? Can we protect whole stands?

- Can we develop a management strategy that reverses downward trend? What tools do we have or need to make a change?

Fire

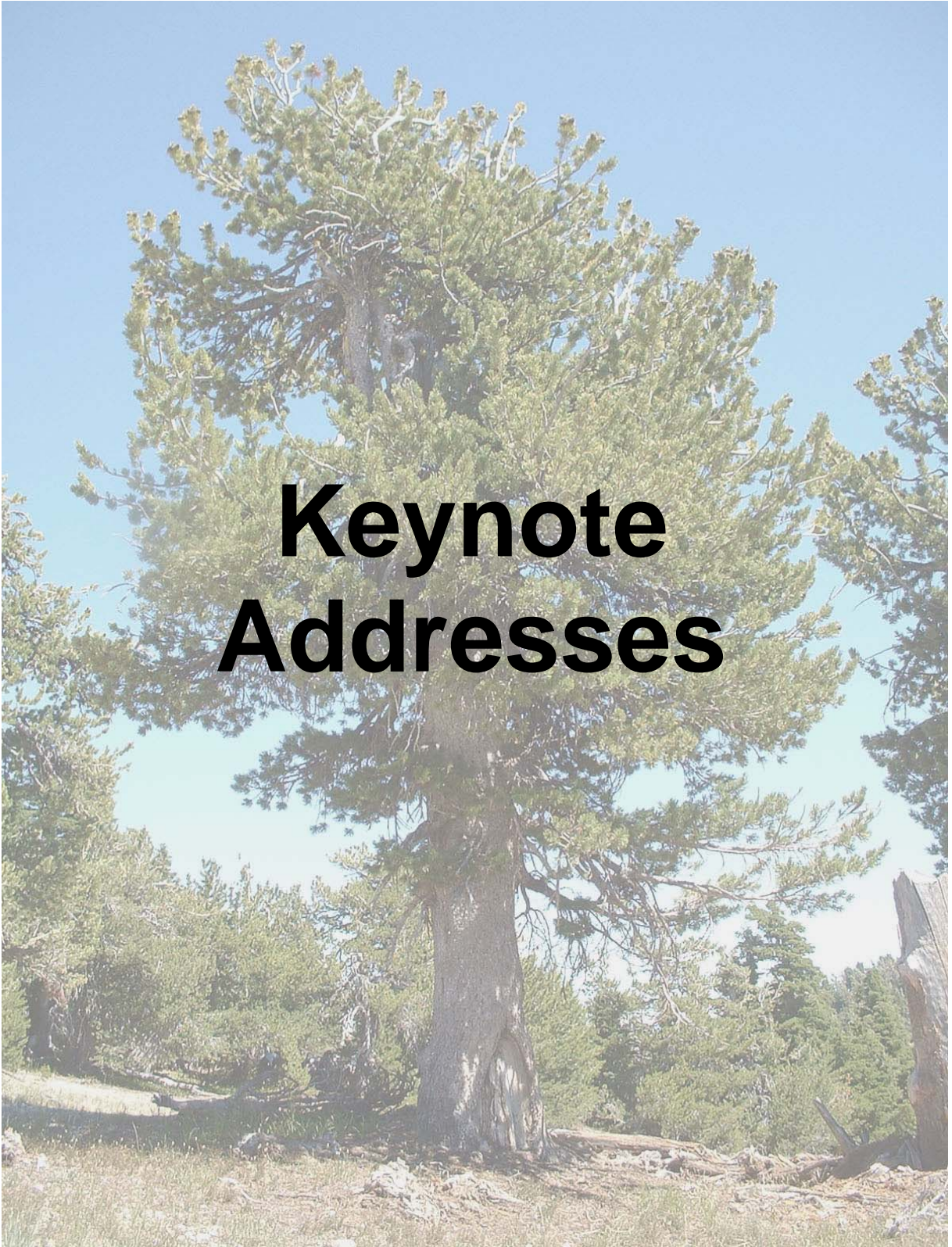
- Interactions between fire and whitebark pine biology (i.e. survival, consequences of fire injury, seed dispersal etc.)
- What are fire regimes & fire history within the range of whitebark pine?
- Do current fire management strategies make things better/worse for whitebark pine?

Natural Resource Policy as it Relates to Restoration

- The need for a restoration/conservation strategy for whitebark pine and other high elevation five needle pines
- Increased education and outreach for policy makers to heighten awareness of the issue
- Recognize white pine blister rust as an invasive species despite its long term establishment
- How to resolve conflicting uses such as Threatened and Endangered Species –what are impediments to conservation?
- Much of the impact from the non-native blister rust and fire management is in wilderness areas or National Parks
- Need to collaborate with other agencies to help shape our management plans/policies
- Increasing multi-disciplinary participation in whitebark conservation
- Develop a common set of best management practices in wilderness.
- Strategies and funding will need to be long-term...

The conference and its proceedings, entitled *Whitebark Pine: A Pacific Coast Perspective* is a direct outcome of the workshop held in 2005. The workgroup concluded that while many of the issues are the same for the whitebark pine on the Pacific Coast and the Rocky Mountain whitebark, there are some unique characteristics which distinguish the Pacific Coast situation from that of the Rocky Mountains. Workshop participants determined that a conference pertaining specifically to the whitebark pine and other high elevation five-needle pines on the Pacific Coast of North America was an important step towards informing a larger audience of the issues and concerns related to these species and in garnering support for restoration efforts. New information, including numerous assessments of the status and health of whitebark pine and other high elevation five-needle pines that had been recently completed were now available pertaining directly to these ecosystems. The charge had been given to develop restoration strategies. The story of these ecosystems, from a Pacific Coast perspective, was waiting to be told.





Whitebark Pine: Ecological Importance and Future Outlook

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Abstract

Whitebark pine (*Pinus albicaulis*) ranges widely throughout the western United States and Canada. On wind-swept sites, it assumes picturesque shapes with sparse canopies and twisted trunks. Whitebark pine is one of eight “five-needled” white pines (Subgenus *Strobus*, Family Pinaceae) found in the West, but the only North American species in Subsection *Cembrae*, a taxon characterized by large, wingless seeds, cones that do not open, and seed dispersal by the nutcrackers. Clark’s nutcracker (*Nucifraga columbiana*, Family Corvidae) is the primary disperser of whitebark pine seeds. The coevolved, mutualistic whitebark pine-nutcracker interaction shapes the population genetic structure of whitebark pine, engineers tree growth forms, and facilitates rapid regeneration of whitebark pine after fire. Whitebark, regarded as both a foundation and keystone species, provides multiple ecosystem services: Its seeds are an important wildlife food; it stabilizes soil and protracts snow melt at high elevations; it initiates community development after fire and serves as a nurse tree for other conifers on stressful sites. These ecosystem services, as well as the biodiversity supported by whitebark pine communities, are diminishing nearly rangewide from the spread of *Cronartium ribicola*, an exotic fungal pathogen that causes white pine blister rust in five-needled white pines. Blister rust cankers kill tree branches, reducing cone production and rendering trees non-reproductive years before trees succumb to the disease. Blister rust infection levels in whitebark pine stands are highest in the northern Rocky Mountains of the United States, with survey averages greater than 80%. Recently, the threat posed by blister rust in this region has been overshadowed by mortality in whitebark pine from outbreaks of mountain pine beetles (*Dendroctonus ponderosae*). Also, in the northern Rocky Mountain and Intermountain ranges, whitebark pine is experiencing successional replacement by shade-tolerant species. Restoration techniques developed for whitebark pine include thinning and burning to reduce competition and prepare seedbeds for natural regeneration or planting seedlings. Seedlings with genetic resistance to blister rust are currently viewed as the most promising approach to restoring whitebark pine communities. Whitebark pine trees with genetic resistance to blister rust are being identified and can be protected against mountain pine beetles. In short, restoration techniques are available, but restoration requires a high level of commitment over time and dependable funding. Without this commitment, we risk the loss of whitebark pine and greatly diminished western forest biodiversity.

Introduction

The dedication of an entire symposium to a single high elevation Pacific Coast conifer, whitebark pine (*Pinus albicaulis*, Family Pinaceae, Subgenus *Strobus*) and its high elevation relatives, the “five-needled” white pines, indicates how our perspectives as forest researchers and managers have evolved over the last quarter century. First of all, the concepts integral to ecosystem management have raised our awareness of the importance of intact forest

ecosystems, not just for their commercial and value but also for the ecosystem services and the biodiversity they comprise. Secondly, research has provided us with better understanding of the role and function of whitebark pine ecosystems, and the current synergism of threats to these ecosystems—invasive disease, pine beetle outbreaks, and advancing ecological succession. We also acknowledge the ecological value of the other Pacific Coast five-needed white pine ecosystems and the similarly uncertain future they face.

The realization that these ecosystems are and will be disappearing from the Pacific mountain landscape over the coming decades raises questions of how well we understand the ecological role of these pines, particularly in this region, and how ecosystems will be affected in their absence. Most importantly, we need to ask whether we understand what management intervention is necessary and sufficient to restore these ecosystems.

When the 20th century history of western forest management is written, whitebark pine should have a special place as a symbol of new priorities. Although the threat posed by the exotic pathogen *Cronartium ribicola* to all five-needed white pines was previously recognized, the looming possibility of regional extirpations of whitebark pine raises concerns about ecological consequences, indicating the importance of these values to management objectives. In some regions, restoration strategies are already being planned. This paper summarizes why whitebark pine has become the focus of our attention: its ecological importance, major threats, current status, and future outlook.

Distribution and taxonomy

Whitebark pine (*Pinus albicaulis*), a tenacious survivor of the harsh upper subalpine zone and treeline ecotone, ranges widely throughout the western United States and Canada (Fig. 1). Its latitudinal range, from about 37° to 55° N, is among the broadest of the *Strobus* pines; whitebark pine ranges longitudinally from 107° to 128° (Olgivie 1990, McCaughey and Schmidt 2001). “In much of its geographic range whitebark pine is primarily a timberline tree, confined to sites that are so cold, snowbound, rocky, or wind-scoured that its competitors are suppressed by the harsh environment...” (Arno 2001). Thus, whitebark pine occupies sites not tolerated by other subalpine conifers, and consequently may range higher in elevation. On wind-swept sites, such as the rim of Crater Lake, it assumes picturesque shapes with sparse canopies and twisted trunks.

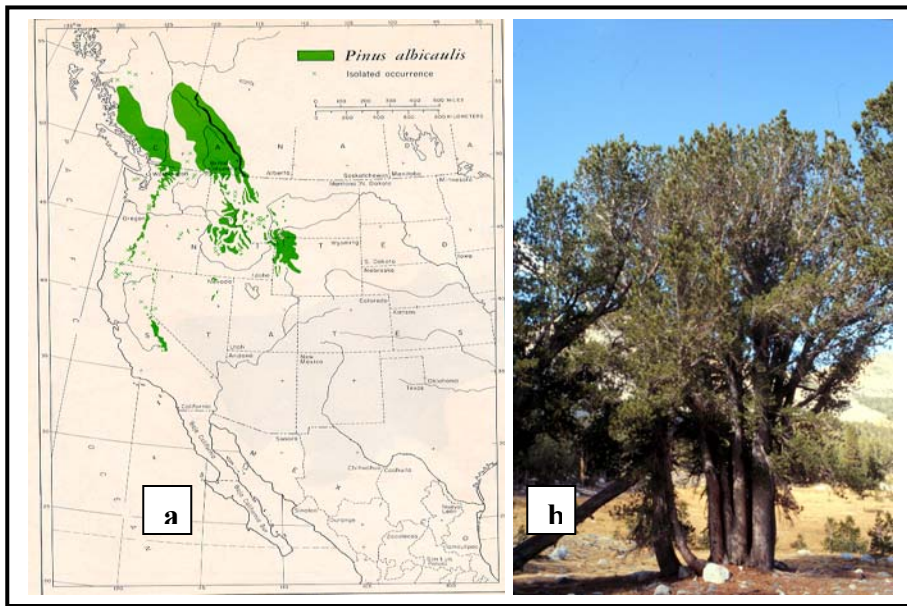


Fig. 1. a. Map of the general distribution of whitebark pine (from Critchfield and Little 1966). The Canadian distribution is patchier than indicated here (see Olgivie 1990). b. Whitebark pine with multiple stems, Tioga Pass, Yosemite National Park, California (Photo: D. F. Tomback).

Whitebark pine is divided into a Rocky Mountain and western or Pacific Coast distribution, connected by a series of ranges in the inter-mountain regions of northeastern Washington and southern British Columbia (Ogilvie 1990, McCaughey and Schmidt 2001). In the Rocky Mountains, whitebark pine ranges in elevation from 3,200 m in western Wyoming to 900 m at its northern limit; and in the west from about 3,660 m in the southern Sierra Nevada to 900 m to its northern limit in the Coast Mountains of British Columbia (Arno and Hoff 1990, Ogilvie 1990).

Whitebark pine has been traditionally classified in Subgenus *Strobus*, Section *Strobus*, Subsection *Cembrae* (Price et al. 1998). According to this scheme, whitebark pine is the only North America member of the *Cembrae*, a taxon considered to be coevolved with nutcrackers (*Nucifraga* spp.). The monophyly of *Cembrae* pines and *Strobus* pine taxonomy, however, are being challenged by recent molecular analyses (e.g., Gernandt et al. 2005)

Community types

Whitebark pine and its high elevation relatives are considered to be “stress-tolerant” pines, capable of extremely slow growth rates under adverse conditions (McCune 1988). Throughout its range, whitebark pine forms climax communities (i.e., self-replacing) on harsh, upper subalpine sites and at treeline, where it usually occurs as krummholz growth forms. Because of slow growth rates, whitebark pine is generally outcompeted on favorable sites, but it is capable of growing throughout the upper subalpine zone in absence of competition (Arno 2001).

Disturbance provides whitebark pine the opportunity to colonize favorable upper subalpine sites: In the northern Rocky Mountains of the United States and southern Canada, and Intermountain Region, where fire at subalpine elevations occurs in both stand-replacement and mixed-severity regimes, whitebark pine forms early seral communities after disturbance

and often persists late in succession (Arno 2001, Tomback et al. 2001, Campbell and Antos 2003). Seral whitebark pine communities in the upper subalpine zone may also be more common in the Pacific Coast ranges than previously realized, as suggested by recent fire history studies in the Cascade Range (e.g., Murray 2006, also this proceedings). This possibility requires further study.

Seed dispersal and ecological impact

Whitebark pine seeds are primarily dispersed by Clark's nutcracker (*Nucifraga columbiana*, Family Corvidae), resulting in an interaction that shapes many aspects of the ecology and population biology of whitebark pine (Tomback and Linhart 1990, Tomback 2001).

Nutcrackers have both behavioral and morphological adaptations for an annual cycle that is based on the year-round use of fresh and stored pine seeds (Vander Wall and Balda 1977, Tomback 1978). Nutcrackers possess a sublingual or "throat" pouch, in which 100 or more whitebark pine seeds may be transported to seed caching sites (Fig. 2a) (Tomback 1978, Hutchins and Lanner 1982). They have a sturdy, long, pointed bill that enables them to dig into unripe whitebark pine cones and tear off cone scales for access to seeds. Their well-developed spatial memory permits each bird to remember the precise locations of thousands of seed caches that it stores in late summer and fall (Vander Wall 1982, Kamil and Balda 1985). Nutcrackers retrieve cached pine seeds aided by their spatial memory in order to feed themselves and their young during winter and spring (Mewaldt 1956). Unretrieved seeds that are exposed to snow melt or summer rains may germinate and lead to whitebark pine regeneration (Tomback 1982). Basic information pertaining to nutcracker harvest, transport, and caching of whitebark pine seeds is reported in Box 1.

Box 1. Summary of typical whitebark pine caching behavior by Clark's nutcrackers. Information based on Tomback(1978, 1982) and Hutchins and Lanner (1982).

- One nutcracker may carry maximum loads of 100 or more seeds, although average pouchloads may contain fewer seeds.
- Caches typically consist of 1-15 seeds, with means ranging from 3 to 5 seeds per cache.
- Nutcrackers typically bury seeds 1 to 3 cm under soil substrate.
- Caching substrate may consist of soil, gravel, pumice, or duff.
- Selected sites for seed caching are both under forest canopy and on open terrain, the latter especially including steep, southerly slopes and recently burned areas.
- Nutcrackers transport seeds from a few meters to 12 km from parent trees.

The adaptations of whitebark pine for interaction with Clark's nutcrackers include large, wingless seeds, which provide nutcrackers an energy-rich food that is obtained efficiently (Fig. 2b) (Lanner and Gilbert 1994, Tomback 1978). Nutcrackers not only gain more nutrition per foraging effort compared to smaller conifer seeds but they also place the harvested seeds directly in their sublingual pouch without the need to remove the seed wings (Tomback 1978). The ripe seeds are retained in cones, which do not open, and seed release depends on nutcrackers. Furthermore, whitebark pine canopy morphology appears to make

cones more visible and accessible to nutcrackers: the cones are horizontally-oriented on the tips of upwardly-directed branches (Tomback 1978, Lanner 1982).



Fig. 2. a. Clark's nutcracker, with a partially filled sublingual pouch, standing on a whitebark pine branchtip. b. Whitebark pine cones, Crater Lake National Park. (Photos: D.F. Tomback)

Seed dispersal by nutcrackers has important consequences for the distribution, successional status, growth form, and population genetic structure of whitebark pine (Tomback and Linhart 1990, Tomback 2001). Where whitebark pine grows is determined by the cache-site preferences of nutcrackers in conjunction with the environmental tolerances of whitebark pine seeds and seedlings. Whitebark pine seedlings are morphologically robust and tolerant of harsh sites (Arno and Hoff 1990, McCaughey and Tomback 2001). Nutcrackers cache whitebark pine seeds widely throughout the upper subalpine zone landscape (Box 1), but also within the treeline ecotone and above treeline, as well as below the lower elevational limit of whitebark pine. Consequently, whitebark can respond to climate warming or cooling with changes in elevational patterns of recruitment. Seed dispersal by nutcrackers after fire leads to rapid establishment and formation of early seral whitebark pine communities (Tomback et al. 2001a).

The population genetic structure of whitebark is influenced on multiple scales by nutcracker seed dispersal (Tomback 2001, Tomback 2005 for detailed overview). First of all, the tendency of nutcrackers to bury more than one seed per cache often results in a "tree cluster" growth form, comprising genetically distinct stems (resembling the tree in Fig. 1b) that tend to be genetic relatives (e.g., Rogers et al. 1999). Stems within clusters tend to be closely related to each other, because nutcrackers typically harvest many seeds from the same parent tree. This growth form leads to a clumped population dispersion pattern for whitebark pine. Furthermore, neighboring trees or tree clusters tend not to be genetically related—a pattern that emerges when different nutcrackers cache seeds from different trees haphazardly within an area. On a regional scale, whitebark pine populations are not geographically genetically differentiated, possible because of long-distance seed dispersal by nutcrackers (e.g., Bruederle et al. 1998). On a rangewide scale, mitochondrial DNA haplotype distribution suggests how nutcrackers impacted whitebark pine migration from Pleistocene glacial refugia (Richardson et al. 2002a, 2002b). There are three distinct mitochondrial DNA haplotypes,

each likely representing a Pleistocene refugium, and the boundaries between haplotypes appear to reflect geographical limits to seed dispersal by nutcrackers.

Whitebark pine as a foundation and keystone species: Why we are concerned about losing whitebark pine.

There is a growing recognition that single species may exert important influences that shape or define the character of entire ecosystems. These important species are considered *foundation species*: “A single species that defines much of the structure of a community by creating locally stable conditions for other species, and by modulating and stabilizing fundamental ecosystem processes” (Dayton 1972, cited in Ellison et al. 2005). Whitebark pine is an important foundation species, providing ecosystem services with respect to community development, watershed stability and hydrology, and treeline community development, as outlined below (Box 2). It may also be regarded as a *keystone species*, which by definition has greater effects on biodiversity than its abundance or density would suggest (e.g., Krebs 2001). Whitebark pine supports biodiversity in part through its large, nutritious seeds, which comprise an important wildlife food, but by also providing wildlife habitat especially at the highest elevations (Box 2). Whitebark pine seeds are high in fat (52% by weight) (Lanner and Gilbert 1994). At least 12 species of birds from five families and mammals from three families are known to feed on whitebark pine seeds—among them the black bear (*Ursus americanus*) and the grizzly bear (*Ursus arctos*), which usually obtain seeds by raiding the middens of red squirrels (*Tamiasciurus hudsonicus*) (Tomback and Kendall 2001). Whitebark pine seeds are a major food for grizzly bears in the Greater Yellowstone Area, and considered important for the long-term viability of the population (Mattson et al. 1992).

Box 2. The foundation and keystone roles of whitebark pine, based on overviews in Tomback and Kendall (2001) and Tomback et al. (2001b) and references therein unless noted otherwise.

- Broad geographic distribution and ecological tolerances lead to a wide array of community types (Arno 2001).
- Seeds are an important wildlife food.
- Found at the highest treeline elevations
- Most often initiates tree island formation in the northern Rocky Mountains (L.M. Resler and D. F. Tomback, unpublished data).
- Grows to the highest elevations in the treeline ecotone, providing wildlife habitat.
- Slows snow melt and run-off in the treeline ecotone (Farnes 1990).
- Reduces soil erosion at high elevations.
- Establishes rapidly after fire and other disturbances (Tomback et al. 20011).
- Tolerates harsh conditions after fire.
- Facilitates community development after disturbance.
- On harsh sites, acts as a “nurse” tree (Callaway 1998).

Losses of whitebark pine from watershed to regional scales thus results in diminished ecosystem services, altered community structure, including greater homogeneity of forests, and reduced carrying capacity for wildlife.

Why whitebark pine is declining

Three factors have caused widespread losses of whitebark pine: the spread and intensification of *Cronartium ribicola*, the introduced pathogen that causes white pine blister rust; recent mountain pine beetle (*Dendroctonus ponderosae*) upsurges; and the successional replacement of whitebark pine by more shade-tolerant species. The threat posed by each factor varies in importance geographically.

Cronartium ribicola, accidentally introduced to western North America in 1910, has spread nearly rangewide in whitebark pine (Fig. 3). The disease has a complex life cycle with five spore stages, requiring two hosts for completion of a cycle (e.g., McDonald and Hoff 2001). One host group, the “five-needled” white pines of subgenus *Strobus*, is perennially infected by the disease, producing aeciospores most years, which then infect alternate hosts. The alternate hosts, which grow new leaves each spring, comprise various species of the genus *Ribes* (Family Grossulariaceae) and, as recently discovered, the herbaceous species *Pedicularis racemosa* and *Castilleja miniata* (Family Orobanchaceae) (McDonald et al. 2006). Blister rust, growing on the leaves of alternate hosts, releases basidiospores in late summer, which start new infections on infected and uninfected pines. The fungus enters the stomates of pine needles and grows into the branch or stem of a tree, producing a sporulating canker within about 3 years.

Trees with blister rust disease are weakened and experience reduced radial growth (L. Daniels, personal communication). Blister rust cankers in the tree canopy kill branches and thus reduce or eliminate seed production years before the trees themselves succumb to the disease. Blister rust cankers that develop on stems usually girdle and kill the tree. However, a small number of trees within each population (< 5%) show genetic resistance to blister rust and are the basis for restoration strategies speeding up natural selection.

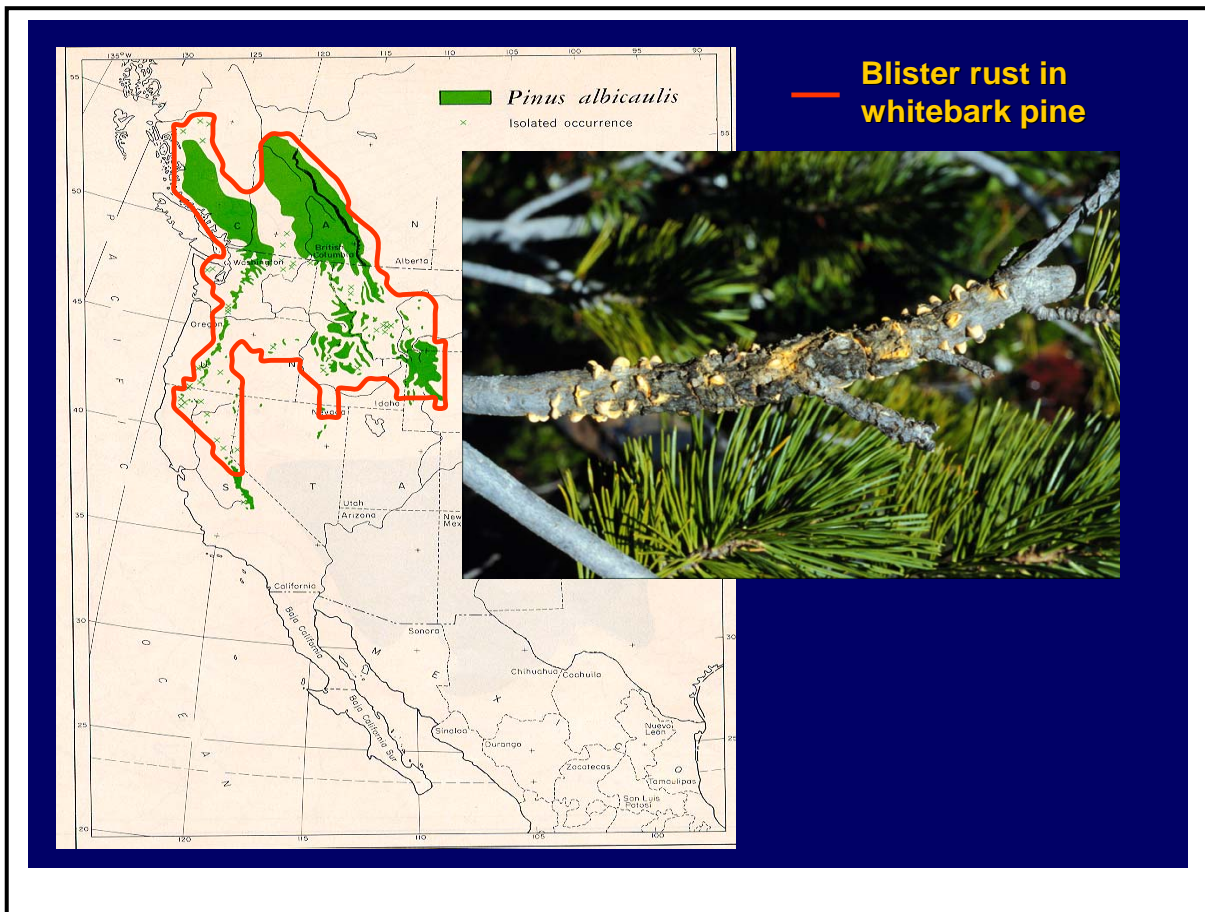


Fig. 3. Map showing the distribution of *Cronartium ribicola* within the range of whitebark pine (modified from Critchfield and Little 1966). Inset picture of a blister rust canker on a whitebark pine branch, with aeciospore sacs (Photo: D.F. Tomback)

Blister rust now infects whitebark pine nearly rangewide (Fig. 3), with the exception of the southern Sierra Nevada (P. Maloney and J. Dunlap, this proceedings) and several Great Basin ranges (J. Guyon, pers. commun.). The highest levels of blister rust infection, mortality, and canopy damage occur in the northern Rocky Mountains of the U.S., including western Montana, Glacier National Park, the Bob Marshall Wilderness Complex, and the Blackfoot Reservation, with average infection levels over 80% (e.g., Keane and Arno 1993; Keane et al. 1994; Kendall et al. 1996). Mean infection levels are now about 20 to 70% in most other regions, including the northernmost limits of whitebark pine’s distribution (Schwandt 2006, D.F. Tomback and P. Achuff, unpublished data). Furthermore, as whitebark pine cone production diminishes, nutcracker numbers may decline, or the birds may avoid subalpine elevations, reducing the possibility of seed dispersal (Tomback and Kendall 2001). Blister rust alone has the potential to cause local, if not regional and even range-wide extirpation of whitebark pine in the absence of management support.

Currently, large-scale outbreaks of mountain pine beetles in whitebark pine are occurring in the northern Rocky Mountains and Intermountain Region of the United States—including areas where blister rust infection levels are high. In 2005 in the Greater Yellowstone Area, aerial flyovers indicated that nearly 720,000 whitebark pine trees had been killed by mountain pine beetles in 2004 (Gibson 2006). Upsurges in mountain pine beetle populations have occurred in the same regions in the past, leaving behind stands of old snags or “grey

ghosts” (e.g., Perkins and Swetnam 1996). These periodic outbreaks may be driven by higher temperatures, which are favorable to beetle development; this does not bode well for global warming trends (Logan and Powell 2001). With mountain pine beetles rapidly killing whitebark pine trees in many populations along with blister rust, the losses of whitebark pine are accelerating. Furthermore, mountain pine beetles are killing trees that are genetically resistant to blister rust.

The third threat to whitebark pine is successional replacement, possibly exacerbated by fire suppression. Studies in the northern Rocky Mountains and Intermountain region have documented shifts to late seral communities (e.g., Keane and Arno 1993, Murray et al. 2000). Historically, the landscape comprised a mosaic of different successional stages, maintaining relatively more whitebark pine than today.

Restoration—the last hope for whitebark pine

The challenge is to prevent local and even regional extirpation of whitebark pine where blister rust infection levels are high, and many trees have already been lost. A major restoration strategy is to speed up natural selection by planting genetically rust-resistant seedlings. The primary objective is to maintain whitebark pine on the landscape until genetic resistance to blister rust is well-distributed throughout populations.

This strategy requires that rust-resistant whitebark pine trees be identified and protected against mountain pine beetles, for example with verbenone pouches (e.g., Bentz et al. 2005), and cones collected to grow seedlings for outplanting. Genetic resistance in whitebark pine has been recently verified: Seedlings were grown from seed collections from 108 potentially rust-resistant whitebark pine trees in the Intermountain and Northern Rocky Mountain region and then screened for blister rust resistance. Overall, 48% of the parent trees demonstrated at least one genetic resistance mechanism (Mahalovich et al. 2006). Seedbed preparation through prescribed fire or thinning may be necessary prior to planting seedlings to reduce competition. Seedlings may also be planted in new and old burns, particularly in areas with little potential for natural whitebark pine regeneration. Direct seeding rather than planting seedlings may be a useful alternative technique that deserves further study, particularly for wilderness areas where disturbance must be minimized. Additional strategies include encouraging natural regeneration by creating “nutcracker openings” in mature forests, and using burning and thinning to reduce competitors of whitebark pine in late seral communities (Keane and Arno 2001).

Whitebark pine restoration is a multi-generational stewardship commitment that should be built into regional National Forest plans, with dependable funding. The goal is the spread of genetic resistance to blister rust throughout whitebark pine populations over time. Without management intervention, the losses of whitebark pine and also its high elevation relatives will have major consequences for western forest biodiversity. Collectively, the high elevation five-needled white pines represent a multiplicity of community types across broad environmental, elevational, topographic, and geographic spectrums. Losing this magnitude of biodiversity would be a national tragedy.

Acknowledgments

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Ecology of Whitebark Pine in the Pacific Northwest

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Whitebark pine (*Pinus albicaulis*) is found in the subalpine zone throughout the Coastal, Olympic, Cascade, and Klamath Mountains, extending well into California at high elevations along the Sierra Nevada. Precipitation is > 300 cm/yr on the western slopes of the Coastal Mountains in British Columbia and the Olympics & Washington Cascades (Franklin and Dyrness 1988). Precipitation generally decreases along the Oregon Cascades moving south, with 50-100 cm/yr common in southern Oregon. The coastal mountain ranges in southern Oregon and Northern California, including the Siskiyou, Trinity, and Klamath Mountain ranges also receive precipitation upwards of 300 cm/yr, decreasing to 150-200 cm/yr throughout much of the California Cascades and northern Sierra Nevada (Schoenherr 1992).

In the coastal ranges and Cascade ranges of British Columbia, Washington, and northern Oregon heavy precipitation leads to persistent snowpack, and therefore whitebark pine is often restricted to exposed ridges of the subalpine zone or drier sites in the rain shadow of these ranges where it commonly grows in association with subalpine fir (*Abies lasiocarpa*), Engelmann spruce (*Picea engelmannii*), and lodgepole pine (*Pinus contorta*). Whitebark pine is also a minor component of moister subalpine sites, or in mixed subalpine forests at lower elevations. Moister subalpine sites west of the Cascade crest in the southern Washington and northern Oregon Cascades have mountain hemlock (*Tsuga mertensiana*) and whitebark pine in association on some of the highest and most exposed slopes (Diaz et al. 1997). Whitebark pine typically loses dominance in the lower subalpine zone, and its dominance being restricted to drier sites may be related to drought tolerance. However, whitebark pine is also considered a seral species with lower shade tolerance than most Pacific subalpine conifers. High elevation whitebark pine populations are typically isolated on mountain peaks, as whitebark pine is outcompeted by other subalpine tree species at lower elevations, therefore forming a metapopulation structure.

The southern Oregon Cascades show a decrease in subalpine fir and Engelmann spruce, and an increase in mountain hemlock, lodgepole pine and Shasta red fir (*Abies magnifica* var. *shastensis*) growing in association with whitebark pine. Westside, wetter slopes and lower subalpine slopes are often dominated by Shasta red fir and mountain hemlock with whitebark pine increasing in abundance on ridges. Mountain hemlock is often the dominant subalpine species in both the number of trees/ha and in basal area on mesic sites, with lodgepole pine and Shasta red fir typically more abundant than whitebark pine; western white pine (*Pinus monticola*) is also present (Goheen et al. 2002). Whitebark pine is common on high elevation ridges in this region, and drier soils lead to greater dominance by whitebark pine on these sites, occasionally forming pure stands in areas east of the Cascade crest. The subalpine zone of the eastern side of the southern Cascades are often more open with

lodgepole pine increasing in importance (Hopkins 1979). Whitebark pine is also common near treeline in the Sierra Nevada, Oregon and California Cascades and coastal mountains, with increasing importance of additional 5-needle pines including *Pinus flexilis*, *Pinus balfouriana*, and *Pinus longaeva* (Schoenherr 1992).

White pine blister rust (*Cronartium ribicola*) is common throughout the Pacific Northwest, generally decreasing toward eastern WA and south through OR and CA; lower infection levels may be related to lower transmission rates on drier sites and/or to a more recent arrival of white pine blister rust on these sites. The spread of blister rust south and east, particularly in California and the Great Basin region, will require additional attention and resources to be allocated to all high-elevation 5-needle pines. There is considerable variability in blister rust infection at finer scales, for example across mountain ranges, and this variability may be related to local variation in the alternate host *Ribes*, microclimate, and/or variation in blister rust resistance among hosts.

Field data used to create a spatially explicit metapopulation model for whitebark pine for Mt. Rainier National Park using RAMAS GIS predicts a rapid decline in whitebark pine in the park, with the population falling below 100 individuals in 148 years (Ettl and Cottone 2004). The proportion of blister rust resistant whitebark pine is unknown, but incorporating resistance into the model only moderately slows population decline (Cottone 2001). Infected trees show relatively low cone production (DelPrato 1999) and therefore even resistant trees, with presumed slow disease progression, are likely to have limited reproduction. The fire return interval is variable in whitebark pine stands across the Pacific Northwest (range ~30-300 years) with wetter sites showing longer return intervals. Furthermore, low fuel availability in open grown high-elevation ecosystems typically leads to poor fire transmission across the landscape. Modeling a fire return interval of 50 years, projects a faster decline for whitebark pine in Mt. Rainier than in the absence of fire (Cottone 2001). It seems likely that mixed species whitebark pine stands would benefit from reduced competition provided by prescribed fire, but it is unclear whether the loss of whitebark pine from fire-related mortality could be compensated for by increased whitebark pine seedling establishment in recently burned areas. Natural progression of white pine blister rust throughout whitebark pine communities puts the species at risk of local extinction without human assistance, although whitebark pine may persist through chance, patchy host distribution, or natural selection. Propagation and planting of resistant whitebark pine hold the best prospects for maintaining whitebark pine in the Pacific Northwest. The spread of blister rust suggests a similar fate for all 5-needle high-elevation pines, and gaps in our knowledge of these systems should be addressed in preparation for the arrival of blister rust.

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Whitebark Pine in Peril: A Rangewide Assessment and Strategies for Restoration

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The following is a brief summary of the results of year-long special assignment for the US Forest Service Washington Office of Forest Health Protection. This assignment was the result of a prior study that found most of our native five needled pines are declining (Samman et al. 2004). My assignment was to:

- Compile a range-wide health assessment of whitebark pine
- Compile restoration strategies
- Describe information needs and challenges to restoration

This was a challenging assignment, but I found a great number of resource specialists from several agencies that willing shared data and experiences with me and my final report, *Whitebark Pine in Peril: A case for restoration* (Schwandt, 2006), is a tribute to their generosity. Copies are available from my office (e-mail: jschwandt@fs.fed.us).

Health Assessment

Although there has been no range-wide systematic health assessment for whitebark pine,

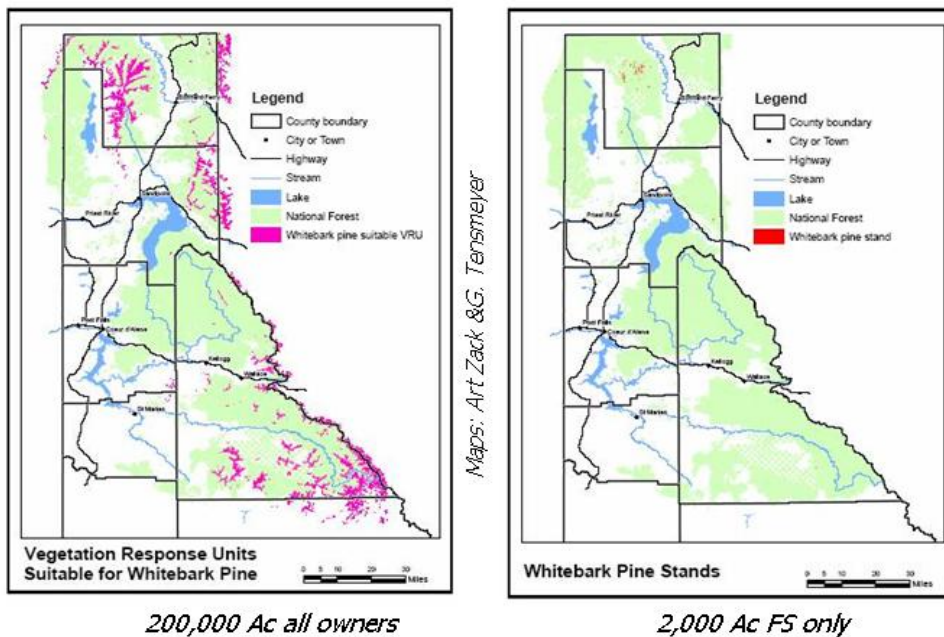


Fig.1 Potential historic and current range of whitebark pine in northern Idaho.

many surveys have found that this species is in peril in much of its range. White bark pine has disappeared from as much as 98% of its potential habitat in northern Idaho (fig 1.) This dramatic decline is due to a combination of several factors including white pine blister rust, competing vegetation, fire, and bark beetle outbreaks. All of these factors may be exacerbated by climate changes. White pine blister rust is a primary concern because this introduced disease has radically altered historical regeneration pathways by quickly killing small trees as well as causing mortality or reducing cone crops in large trees. Rust infected trees have been found in all but the very southern tip of its range in the southern Sierra Nevada in California. Although surveys have found wide variation, infection levels are generally lower in drier habitats, and blister rust continues to spread and intensify (Schwandt 2006). Hopefully additional permanent monitoring plots will help to explain this variation as well as provide additional information on spread, intensification, and mortality rates.

The urgency for restoration in some areas has been increased by recent outbreaks of the mountain pine beetle (MPB) which has killed thousands of mature trees, some of which may carry natural resistance to blister rust (fig. 2). Although mountain pine beetle is a native insect and outbreaks in the past have been documented (Perkins and Sweetnam, 1996), the current outbreaks have been more intense due to warm winters resulting in reduced beetle mortality and more beetles completing their life cycle in a single year rather than two (Logan and Powell 2001).

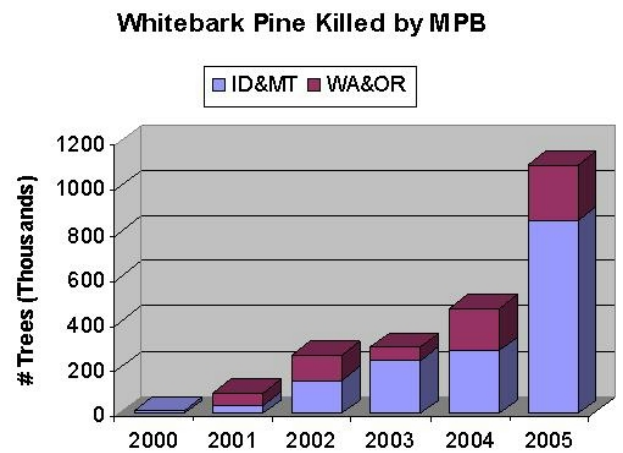


Fig 2. Whitebark pine killed by MPB in the Pacific Northwest over the last several years

Restoration Strategies



The ultimate goal is to increase the proportion of whitebark pine with natural blister rust resistance that will survive in the presence of white pine blister rust. This will require strategies that will protect and enhance existing whitebark pine populations and restore populations where they have been lost. This includes strategies that:

- will evaluate and monitor whitebark health,
- promote selection of natural blister rust resistance,
- reduce competing vegetation,
- enhance regeneration opportunities,
- conserve isolated gene pools, and
- minimize bark beetle losses.

Fig. 3. testing for rust resistance

Surveys continue to show rust spreading and intensifying throughout the range of whitebark pine (Ward et al. 2006, Smith et al. 2006). Fortunately preliminary testing has found some whitebark pine have natural resistance to blister rust (fig. 3.), and efforts are underway across the range to identify trees without blister rust to test for resistance. At the same time it is important to protect existing populations and enhance natural regeneration wherever possible to maintain populations and encourage natural selection.

Whitebark pine is strongly dependent on fires to create planting sites and reduce competing vegetation (especially in the Rocky Mountains, Arno 2001). However, fire suppression in some areas is preventing the natural role of fire and accelerating conversion to competing vegetation. Although wildfires have burned entire populations in some remote areas, prescribed fire and wildland fire are key tools for restoration and need to be encouraged wherever possible.

It is clear that without active management, losses will continue so managers need to prioritize and implement these strategies based on stand conditions, whitebark pine health, and management objectives.

Information Needs and Challenges

Since most whitebark pine occurs in remote or wilderness areas, the logistics of planting seedlings or implementing treatments such as thinning or girdling competing vegetation can be a major challenge. However, it maybe possible to plant seeds instead of seedlings and use wildland fire to enhance natural restoration in remote areas. In addition, logistics of locating and protecting rust resistant trees from mountain pine beetle can be a challenge. However, anti-aggregant pheromones are being developed and are showing promise of protecting high value individual trees from mountain pine beetle attacks (Kegley and Gibson 2004).

There are still many unanswered questions regarding whitebark pine restoration. Answers to these questions will provide practical information that can be used to prioritize stands for restoration based on potential ecosystem impacts. Major information needs include:

- determine frequency of natural resistance across the range of whitebark pine
- identify resistance mechanisms, their heritability, frequency, and distribution
- guidelines for growing, testing, and regenerating seedlings at high elevations
- examine rust epidemiology and variation of infection to develop hazard rating models
- further examination of relationships between whitebark pine and nutcrackers and other wildlife as well as bark beetles, fire, and climate
- further examination of the genetic variability in both the rust and whitebark pine

Conclusion

Since the spread of white pine blister rust appears to be relentless, we must act now to develop and implement strategies to conserve and restore whitebark pine to maintain the diversity and health of whitebark pine ecosystems. If all whitebark are eliminated within the caching range of Clark's nutcrackers (about 20 km), that piece of the whitebark pine range will be permanently lost unless it is artificially replanted. If seed have not been collected from these areas, a small part of the gene pool will be lost forever (Tomback 2001).

Successful restoration will take a long time and will require range-wide, coordinated multi-agency efforts with a long-term commitment. However, if we can establish enough rust resistant whitebark pine in an area, it may be possible for natural processes to eventually resume restoration. Therefore it is critical that implementation of restoration efforts be given high priority by land managers.

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Photo credits:

Figure 3. Richard Sniezko

Assessment of Whitebark Pine Health in Oregon and Washington

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Surveys were conducted in 2005 in 29 whitebark pine (*Pinus albicaulis*) stands on national forests in Oregon and Washington to assess the incidence of white pine blister rust (*Cronartium ribicola*) infection and evidence of recent mountain pine beetle (*Dendroctonus ponderosae*) infestation in whitebark pine. Surveys consisted of fixed-width (10-meter), variable-length belt transects (similar to Whitebark Pine Ecosystem Foundation 2005). Transect length was set at a minimum of 50 meters. All whitebark pine trees greater than 1.4 meters tall within each transect were observed for status (live or dead), white pine blister rust and evidence of mountain pine beetle infestation. Mortality from all causes was 21.3 percent. By transect, the percentage of trees that were dead ranged from 1.1 percent to 61.0 percent, with a median of 19.6 percent. The overall blister rust infection rate in live trees was 23.9 percent. Blister rust infections were observed in 23 of the 29 transects. Infection rates in those 23 transects ranged from 4.7 percent to 73.3 percent of live trees, with a by-transect median of 26.8 percent. The lowest infection rates were east of the Cascades in southern Oregon. Evidence of recent mountain pine beetle infestation was observed in 8 of the 29 transects; the rate of infestation ranged from 1.2 percent to 28.4 percent of trees observed. A survey of seedlings within each transect was also conducted: overall seedling mortality was 3.8 percent, and 4.8 percent of live seedlings were infected with blister rust.

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Status of Whitebark Pine and Other High-elevation Five-Needle Pines with Emphasis on Pacific Coast Ecosystems: What are the Issues and Concerns? Perspective from California

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Abstract

Six of nine five-needle white pine species native to the U.S. are found in California, and all of these are susceptible to the exotic pathogen, white pine blister rust (*Cronartium ribicola*). Since entering California, the rust has spread south over the geographic range of sugar pine, but until recently little was known about its impact on the higher elevation pines. From 1995 to 1999, a survey of five species in Sequoia and Kings Canyon National Parks revealed rust in plots of sugar and western white pine only. In 2004-2005 a survey of the high elevation species over their California ranges revealed rust in plots of western white pine, whitebark, and the northern foxtail populations, but not in limber, southern foxtail, or Great Basin bristlecone pines. Mean incidence of rust across all plots was relatively low (12 to 15%), but variation among plots was high (0 to 92%). Rust was observed in a plot at 3400 m elevation in the southern Sierra. Other stress factors such as mountain pine beetle, fire exclusion and climate change are discussed in relation to their impacts on these pines. Practical issues for future management of these high-elevation pines and their ecosystems are also presented.

Introduction

Six of nine five-needle white pine species native to the U.S. are found in California. They are sugar pine (*Pinus lambertiana*), western white pine (*P. monticola*), whitebark pine (*P. albicaulis*), limber pine (*P. flexilis*), foxtail pine (*P. balfouriana*), and Great Basin bristlecone pine (*P. longaeva*). The six species range in elevation from about 150 to 3700 meters. In the State, sugar pine grows at low to mid elevations, western white pine at mid- to high elevations and the remaining four species at high elevations where they are adapted to harsh mountain environments, provide habitat to wildlife, and are culturally valued for their aesthetic appearance and longevity. All are critical components of their ecosystems. However, both biotic and abiotic factors may be affecting the health of these trees and associated ecological components. White pine blister rust, mountain pine beetle, fire exclusion, and changes in climate are most often mentioned in the literature as affecting high elevation pines elsewhere in the West and are discussed in relation to California.

White Pine Blister Rust

One significant biotic factor is the exotic fungal pathogen, white pine blister rust (*Cronartium ribicola*). The pathogen has had a severe impact on five-needle pines in the western U.S., particularly the northwestern States and the western Canadian provinces (Samman et al. 2003, Schwandt 2006). Moreover, it continues to spread over the geographic distribution of these pines. In California, the rust's migration from the north has led to serious concern over its impact on the health of these species and their ecosystems.

Chronology of the spread of white pine blister rust in California: The history and current distribution of blister rust on the lower elevation sugar pine is well-documented. The species is a significant source of rust inoculum due to its wide distribution in the State. The pathogen first entered California about 1930 (Shelly Creek, Grants Pass-Crescent City highway), spread steadily south on sugar pine, and within 70 years had reached Breckenridge Mountain at the southern Sierra Nevada (Kliejunas and Adams 2003). White pine blister rust has not yet stabilized on sugar pine, and continues to spread and intensify. Sugar pine in the Tehachapi Mountains and southern California remain uninfected. Until recently, the incidence and impact of the rust in high elevation ecosystems in California were largely unknown. The rust was reported on foxtail pine in the Klamath Mountains in 1967. Reports since then suggest that the rust may just be entering the higher elevations.

Surveys of white pine blister rust on high-elevation five-needle pines: An extensive survey of sugar, western white, whitebark, limber, and foxtail pines in Sequoia and Kings Canyon National Parks was performed from 1995 through 1999 (Duriscoe and Duriscoe 2002). A total of 151 permanent monitoring plots were established. Rust was found only in plots with sugar pine (21% average incidence) and western white pine (3% average incidence). The populations of whitebark, limber, and foxtail pines within the plots were not infected by the rust.¹ Incidence and severity of rust was closely associated with elevation: it was rarely found above 2,700 meters, and it was most often found in valley bottoms.

A field project was initiated in 2004 to gather blister rust information on high elevation five-needle white pines in other areas of the State. The objectives were to 1) determine the current incidence and levels of blister rust associated with western white, whitebark, limber, foxtail, and bristlecone pines in California, and 2) to establish a system of permanent plots for long-term monitoring of rust incidence and severity in these pine species. A total of 118 long term monitoring plots were established over two field seasons; 43 in western white, 44 in whitebark, 14 in limber, 12 in foxtail, and 5 in Great Basin bristlecone. Standard plot and tree data were collected as well as information on *Ribes* spp. (the alternate host), white pine blister rust, mountain pine beetle, and other damaging agents.

Rust was present in western white (25 of 43 plots), whitebark (18 of 44 plots), and the northern populations of foxtail (5 of 6 plots). It was not found in limber, Great Basin bristlecone, or the southern populations of foxtail pine (Table 1). Non-aeical evidence of rust

¹ Rust was noted on one foxtail pine in SEKI by a forest pathologist, but that observation was not part of the SEKI survey.

was noted on six trees in one northern and on two trees in one southern foxtail plot.² The mean rust levels were relatively low across plots (12 to 14%), but varied widely from plot to plot (0 to 90%). Moderate incidence was observed in northwest California, north-central Sierra Nevada, and the west side of the southern Sierra Nevada. The absence of rust in limber, the southern populations of foxtail², and Great Basin bristlecone pines may be due mainly to the time factor; the rust pathogen has been present in the southern Sierra (on sugar pine) only since the 1960s. With more time and the continuous nature of the forests, rust on sugar pine and western white pine at lower elevations will provide an inoculum source for *Ribes* spp. and, in turn, inoculum for the higher elevation whitebark, foxtail, and limber pine. In this survey, rust was observed on whitebark pine at about 3400 m. Rust is spreading not only south in latitude, but also upward in elevation.

Mountain Pine Beetle

Another biotic factor affecting high-elevation five-needle pines is the mountain pine beetle (*Dendroctonus ponderosae*). A native to western North America, mountain pine beetle has several main host species, i.e., ponderosa, lodgepole, sugar, and western white pines, but may attack other five-needle pines as well (Gene et al. 1990). In California, the biology of mountain pine beetle is not well-understood for the high-elevation forests. In our survey, beetle activity was present in about 50% of the plots in western white, whitebark, and the northern population of foxtail, but absent from the plots of limber, the southern populations of foxtail, and Great Basin bristlecone pines (Table 1). Overall, mortality levels on the plots were low. Mortality ranged from zero in limber pine to a mean of 4% in western white (Table 1).

Table 1. Incidence of blister rust, mountain pine beetle (MPB) and mortality levels in 118 monitoring plots in California.

Pine Species	No. Plots with Rust	Avg. % Rust Levels (Range)	% of Plots with MPB	% Mortality (Range)
<i>P. monticola</i>	25/43	14 (0 – 90)	53.5	4 (0 – 49)
<i>P. albicaulis</i>	18/44	13 (0 – 76)	62	1 (0 – 12)
<i>P. flexilis</i>	0/14	0	0	0
<i>P. balfouriana</i> N	5/6	12 (2 – 32)	42.8	1 (0 – 6)
<i>P. balfouriana</i> S	0/6	0	0	1.5 (0 – 9)
<i>P. longaeva</i>	0/5	0	0	0 (0)

Fire Exclusion and Climate Effects on High-Elevation Five-Needle Pines

California’s forests have been substantially modified by wildland fire suppression since the early 20th century (McKelvey et al. 1996, Skinner and Chang 1996, Skinner and Stephens 2004). However, in the subalpine zone where high-elevation five-needle pines grow, the impact of fire exclusion is thought to be minimal. Subalpine forests are open stands with compact discontinuous fuel loads and natural intervals between fires are quite long (van

² The presence of aecial blisters was required for a positive confirmation of rust as part of the field data collection protocol. Non-aecial rust symptoms in the plots were considered to be unconfirmed rust.

Wagtendonk and Fites-Kaufman 2006). In the upper montane zones, high-elevation forests with five-needle pines, such as western white pine, are found in denser mixed-species stands. Fire intervals seem to be shorter or regimes more variable across the Sierra and Klamath mountains (Skinner et al. 2006, van Wagtendonk and Fites-Kaufman 2006). Yet, recent long fire-free time periods have been described for areas of the upper montane Sierra and the mid to upper montane Klamath mountains (McKelvey et al. 1996, Skinner and Chang 1996, Skinner et al. 2006). Stand densities of white and red fir-dominated forests have increased in the Klamath and southern Cascades since fire exclusion (Skinner et al. 2006, Skinner and Taylor 2006). To our knowledge, an understanding of the natural fire cycles coupled with fire exclusion is not clear for the upper montane forests.

Climate change is another key factor influencing forest dynamics over decades, centuries, and millennia (Millar 2004). In California, temperatures are projected to increase from 2 to 6°C from the year 2000 to 2100 (Cayan et al. 2006). Warmer temperatures may result in elevation shifts of California's high-elevation five-needle pines. Species may respond by shifting to cooler sites, such as moving latitudinally, while other species disperse up the mountains. Warmer temperatures may also enlarge the area of mountain pine beetle activity from the current lower elevations (Logan and Powell 2001). The dynamics of forest communities in relation to climate change are not simple however. Climate continues to oscillate at multiple time scales while impacting the ecological traits of each species. These interactions may lead to species assemblages that change, disappear, or expand over time (Millar and Brubaker 2006).

Practical Issues for Future Management

In considering the future management of high-elevation five-needle pines and their ecosystems, several practical issues were addressed in the presentation:

- 1) **What's the problem?** The high-elevation pine ecosystems in California have multiple threats, including the native mountain pine beetle, the introduced blister rust, climate change, and other stressors. These high-elevation species also occupy harsh environments and/or are at the margins of their ranges in California, making them more susceptible to the abiotic and biotic stress factors.
- 2) **Should we be concerned about high-elevation pines in California?** Yes, although mortality was low in the plots, the data revealed low to moderate rust infection with large plot-to-plot variation and some mountain pine beetle incidence. Biotic and abiotic stressors will likely lead to more changes in the future. We need to follow through with monitoring in the plots to examine the effects. At this time, we can certainly raise the awareness of the impacts that are occurring elsewhere and the level of severity that could potentially occur in California.

The great majority of California's population is closely tied to the urban environment, and seldom, if ever, becomes aware of non-urban problems, much less about what is happening in often inaccessible high-elevation sites. Decline of high-elevation ecosystems is different from a situation like Sudden Oak Death (caused by *Phytophthora ramorum*), where the public can see and is directly affected by dying

trees and the resulting fire hazard. This public awareness and concern led to strong political support. Thus, efforts to increase public awareness are valuable for initial and long-term support on conservation and restoration of high-elevation pine ecosystems.

- 3) **Will management constraints in high-elevation ecosystems limit effective options?** Most high-elevation pine stands in California are in national parks or wilderness, areas that have constraints on management options and degree of intervention. However, active management intervention may be necessary to maintain the natural character of high-elevation pine ecosystems in the face of the exotic white pine blister rust. Managers have different viewpoints on the extent of management that is appropriate. In some situations, restoration of ecosystems is encouraged, but active management to prevent their decline is not. Because multiple factors will influence a decline of high-elevation pines, an interdisciplinary approach will be necessary to develop effective management options. Historically, multi-disciplinary efforts on managing a problem have not always been successful, but early recognition of the complexity of threats may promote information sharing and the development of integrative conservation strategies.
- 4) **Where's the funding?** Who should be paying to provide management and protection of our high-elevation ecosystems, or paying for genetic conservation efforts? Lack of funding has limited implementation of conservation strategies for high-elevation pine ecosystems in California. Funding for the latest rust survey in California provided general information on the current range and levels of blister rust, and there has been some limited seed banking. However, screening for natural rust resistance of high-elevation pines is not operational and testing remains mainly with research. Financial resources are limited for such work.
- 5) **Who is going to do the work?** Assuming that sufficient funds were available to do the work necessary to maintain and restore high-elevation ecosystems, are there researchers and managers available to do the work? If so, would the work be done separately by Region or merged among Regions; similarly, would this effort be an inter-Agency effort? These concerns are not unique to California or to high-elevation pine ecosystems.

In summary, California has begun to gather information about the impact of blister rust on the high-elevation five-needle pines. Certainly, more information would lead to a better understanding about the other biotic and abiotic factors affecting these pines and the associated ecosystems. Such information would provide a foundation for developing future conservation and management strategies of these ecosystems. An effort towards developing such strategies will depend on the public interest, and availability of multi-disciplinary personnel and financial resources.

Acknowledgments

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Whitebark Pine in British Columbia: Current Conditions and the State of Our Efforts

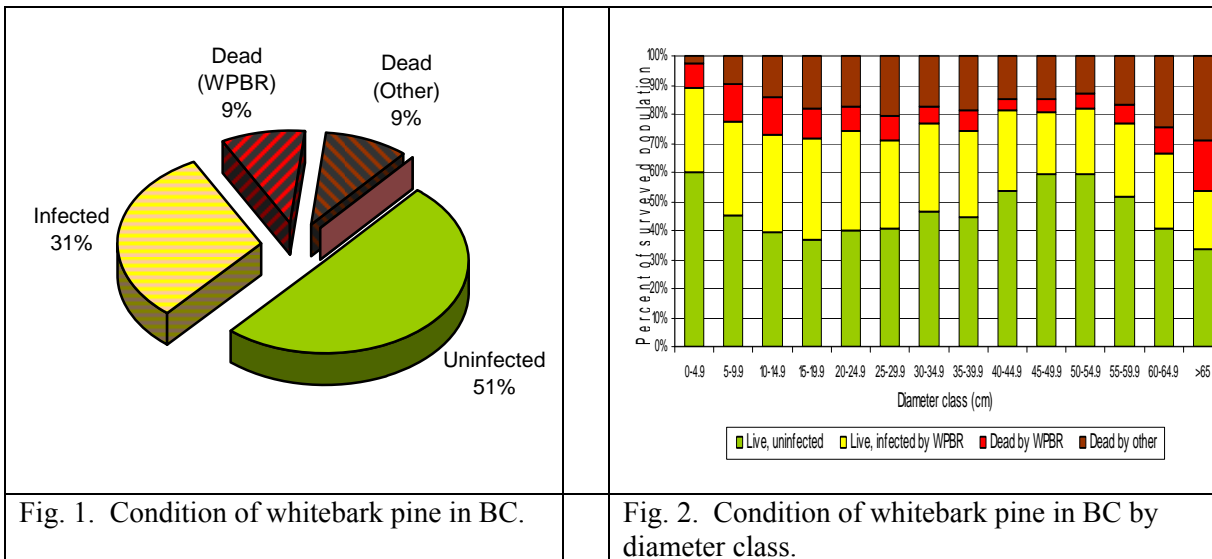
Stefan Zeglen

B.C. Ministry of Forests and Range, Coast Forest Region, Nanaimo, British Columbia, Canada

Over the past few years studies on whitebark pine in British Columbia (BC) have greatly increased our knowledge of the range, habitat and survival of this species. A brief synopsis of the state of whitebark pine is given along with a summary of current research underway in the province.

There is little work published regarding whitebark or limber pines in BC prior to 1967 when Day produced a paper on whitebark pine in the Rocky Mountains. The two most notable points about this paper are that Day berates the foresters of the day for overlooking the utility of whitebark pine in pursuit of other species. Also, there is not one mention of mortality due to white pine blister rust (WPBR) even though by that time the rust must have been present for a couple of decades. It took another 20 years before Ogilvie (1990) produced his reference work on the ecological characteristics of whitebark pine in BC. More recently, a relative flurry of studies have been published starting with Stuart-Smith's (1998) thesis on conservation of whitebark pine in the Canadian Rockies. Campbell and Antos (2000) followed with a broader examination of the ecology and condition of whitebark pine across a large portion of BC. Zeglen (2002) looked specifically at the result of WPBR activity across the range of whitebark pine in the province.

The last two papers quite neatly summarize the recent condition of whitebark pine in its northern range. Overall, almost 1 in 5 whitebark pine in BC are dead, split evenly between mortality from WPBR and other or undetermined factors (see Fig. 1). Another 1 in 3 trees are currently infected by WPBR. Of these, two-thirds have stem cankers, a usual precursor to death by rust. The remaining half of the population shows no infection at this time, although there are the myriad physical defects (e.g., dead or broken tops) that affect the reproductive ability of mature trees.



The incidence of WPBR and mortality varies greatly by location across the province and by diameter of the trees affected. Overall, smaller trees (<5 cm dbh) are less affected by WPBR but that can fluctuate greatly as tree diameter increases (see Fig. 2). Some areas, like Cathedral Provincial Park, showed low WPBR incidence (11.5%) and mortality (1.5%) values across the diameter range while the southern Rockies area was consistently high for both (44.9 and 29.9%, respectively). One troubling aspect of areas with high amounts of WPBR is that often very few live large diameter trees remained signifying that natural repopulation of these areas will be difficult. If one were to imagine a transect from the west to the east across the southern portion of BC, one would note a general trend toward increasing amounts of WPBR infection and mortality as one crossed each of the six mountain ranges from the Coast Range to the Rockies.

Losses to mountain pine beetle, a voracious killer of many native pines in western North America, were found to be low in both studies (<0.5% of trees). However, since the time of these surveys the population of mountain pine beetle in the province has exploded into what is considered the largest recorded insect epidemic ever in North America, covering over 9 million hectares (Westfall 2005). The huge influx of beetles has pushed up the hillsides threatening whitebark pine habitat that is usually insulated from more moderate outbreaks. At this time, the impact of the outbreak on whitebark pine is unknown and will likely not be fully quantified until the outbreak subsides, likely after 2008.

Whitebark pine regeneration (trees <1.3 m dbh) is, for the most part, remarkably uninfected compared to larger trees (see Fig. 3). Unfortunately, the amount of regeneration is often not great with half of the survey plots installed in the Zeglen (2002) study showing no regeneration at all even in areas with mature whitebark pine present. A further third of the plots contained only a few (1-5) trees. This reflects the natural difficulty in getting this species to regenerate (i.e., slow maturity, infrequent cone crops, competition for seed from predators, difficult environment, etc.) overlaid with the additional problem of WPBR and beetles killing mature trees. In many areas the most frequently found regeneration is subalpine fir, a climax species. This suggests that unless some sort of beneficial disturbance occurs, whitebark pine regeneration, and even established trees, will find it difficult to persist on many of these sites.

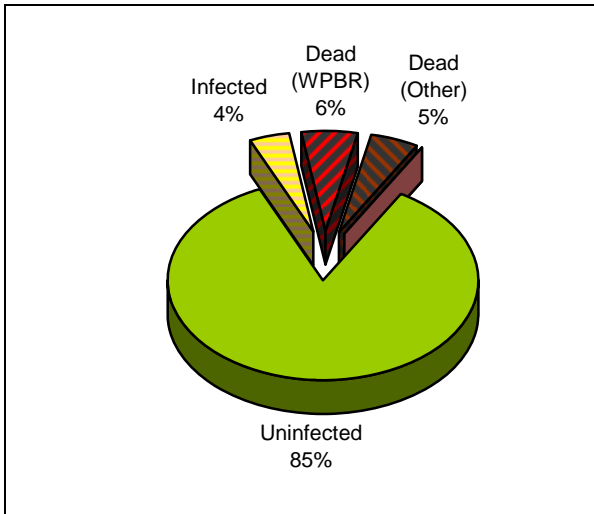


Fig. 3. Condition of whitebark pine regeneration.

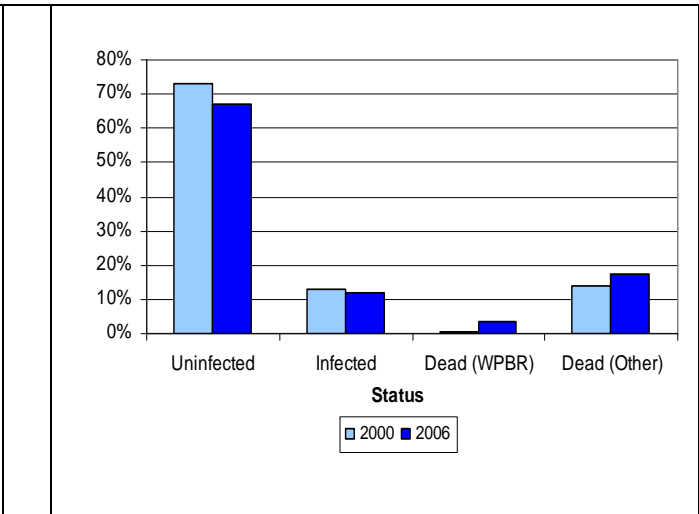


Fig. 4. Preliminary survival data from three monitoring plots in the Coast Range.

While these and other surveys have outlined the current incidence of infection and mortality of whitebark pine, there is little information regarding the rate of mortality. A few long-term monitoring plots have been installed but these are too new to yield much information. A recent examination of some plots that I have has shown a gradual increase in mortality of whitebark pine at three sites along the Coast Range over the last five years (see Fig. 4). The increase works out to roughly 1% per year with most being attributable to the action of WPBR. The apparent leap in the amount of WPBR-caused mortality is mainly due to our ability to definitively identify and track infections rather than having to guess post-mortem. These results are very preliminary and should be taken with a huge grain of salt.

On a positive note, there has been a tremendous amount of interest in studying BC whitebark pine in the new millennium. Most of this effort is centred in universities, often in conjunction with the UBC Centre for Gene Conservation headed by Sally Aitken. Two of her graduate students have recently completed their work; Jodie Krakowski (2003) studied conservation genetics of whitebark pine and Andy Bower (2006) recently completed his PhD studying several aspects of the species that he describes elsewhere in this workshop. Other ongoing studies include Randy Moody's Master's-level study examining the post-fire regeneration of whitebark pine and a PhD-level dendrochronology study of whitebark pine dynamics by Carmen Wong (both from UBC). Elizabeth Campbell (BC Ministry of Forests and Range) is also midway through an evaluation of the impact that mountain pine beetle is having on whitebark pine during the current epidemic. Parks Canada also has an active whitebark pine program in the Rockies headed by Cyndi Smith.

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White Pine Blister Rust

Why Here, Why Now, What Next?

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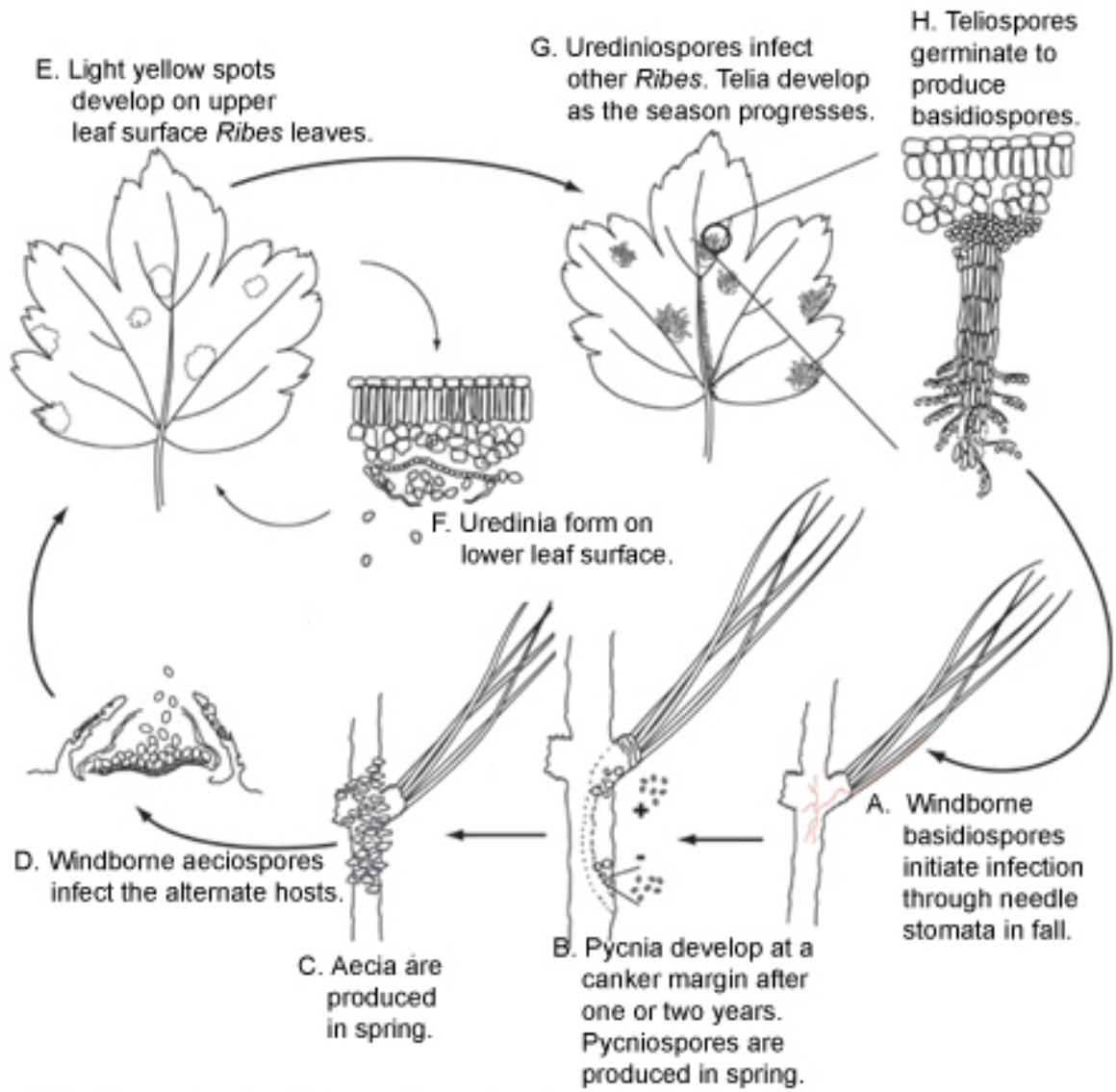
White pine blister rust is caused by the stem rust fungus *Cronartium ribicola*. It infects species of 5-needled pines, and the North American “soft” pines are particularly susceptible. The center of evolutionary origin for *Pinus* is evidently North America, where 69 species are recognized today. Asia is a secondary center of diversity for the genus, with about



38 species. The 5-needled pines of *Pinus* section *Strobus* are also concentrated in North America. Not unexpectedly, North America is the center of diversity for *Cronartium*, the pine stem rusts. These biotrophic pathogens have evolved with their host pines, and as with most co-evolved symbiotic interactions, host and pathogen maintain a rough detente—native rusts do not threaten the ecological standing of the native pines they parasitize.

When transported beyond the constraints of environment and host genetics, however, rust fungi, like many pathogens, have the potential for devastating impact. *C. ribicola* provides textbook example. White pine blister rust is native to eastern Asia. It is reportedly not difficult to find on Asian 5-needled pines, but seldom kills trees in its native home. It was introduced to Europe, probably in the early 1700s, where it proved destructive on *P. strobus* which was widely planted at the time. In the late 1800s or early 1900s it was transported to North America. Twice. Despite the evident threat to North American species, first eastern white pine seedlings and later western white pine seedlings, raised in European nurseries and exposed to blister rust, were imported to this country for forest outplanting. After damage was reported in the United States quarantines were imposed, but too late.

Today, blister rust occurs throughout the range of eastern white pine, and in the west, it has killed most of the western white pine and sugar pine in the northern parts of their ranges. It continues to intensify on the high elevation species, white bark and limber pine, and is still spreading south and east in the western mountains. The endemic white pines of Mexico and Central America are next in line.



White Pine Blister Rust Disease Cycle (Drawing by Vickie Brewster).

The life cycle of the pine rusts, including white pine blister rust, is long and complex. Five separate spore stages, on two unrelated host species, are needed to initiate a new generation. At each stage critical environmental conditions must be met, and host tissue at the appropriate stage of development must be available. It is easy to understand why damage is limited in co-evolved ecosystems. In western North America, however, *C. ribicola* found favorable environments, white pines that generally lacked useful resistance, and new, widely distributed alternate hosts.

Because of the economic as well as the ecological values of the white pines, efforts to reduce losses from blister rust have continued for nearly 100 years. The first quarantines were imposed too late, and were too limited to prevent the first introductions to North America. Quarantine protections are still important, however. There are evidently

different pathogenic races of the pathogen in eastern Asia today, with different alternate hosts, than the races that were introduced here years ago. We don't want those new races; we have trouble enough with the limited rust gene pool that we already face.

The textbook control for long-cycled rusts like wheat stem rust, and blister rust, is removal of the alternate host. The theory is sound, and phenomenal efforts were made, but ribes eradication didn't put a measurable dent in the blister rust epidemic in the west. In the eastern United States, however, where less robust topography makes spore dispersal more predictable, ribes eradication continues to be a practical tool in areas where the overall rust hazard is moderate or low. It may be useful in the west in special cases, perhaps in areas where the environment is marginal for the rust, or where resistant pine selections are deployed.

Pruning also offers useful protection where rust hazard is not extreme. Preventative pruning of lower branches, to remove the tissue most likely to be initially infected, or targeted canker pruning cutting off infected branches before cankers reach the main stem where they will kill the tree, are labor intensive but may be useful tools in high value situations.



Tree resistance is the disease control strategy of choice, however. Individual resistant trees have been found in all of the North American 5-needled pines, and active resistance selection and breeding programs continue for *P. strobus*, *P. monticola*, and *P. lambertiana*. Aggressive efforts are underway to find and exploit resistance in *P. albicaulace*. There have been many frustrations, and even embarrassing failures along the way, but lessons have been learned. While evolution of resistance is the tree's natural strategy, so too is evolution of new virulence genotypes the way of the fungus. Paradoxically, to be durable, that is to be

successful in the long run, tree resistance, and the breeding and outplanting programs that enhance it, must allow for the continued survival of the pathogen as well.



One hundred years of blister rust in North America have spawned 100 years of research, and we understand this disease better than most others. Surprises still come around, though, and new directions hold promise for new and better protections. The recent demonstrations of additional alternate hosts for *C. ribicola* in North America and of hybridization between *C. ribicola* and the native *C. comandrae* on limber pine were dramatic surprises. Characterization and better utilization of “slow rusting” types of resistance in more species of 5-needled pines may allow establishment of pine populations more like the co-evolved populations found in areas where rust and pine are both indigenous. Now, if we can keep whitebark pine from being pushed off the top of the mountains by warming climates.....

The Mountain Pine Beetle and Whitebark Pine Waltz: Has the Music Changed?

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Introduction

The mountain pine beetle, *Dendroctonus ponderosae* Hopkins (Coleoptera: Curculionidae, Scolytinae) (MPB), is a bark beetle native to western North American forests, spanning wide latitudinal and elevational gradients. MPB infest and reproduce within the phloem of most *Pinus* species from northern Baja California in Mexico to central British Columbia in Canada, and their geographic range is dictated by the distribution of both suitable host species and favorable climatic regimes (Safranyik 1978, Logan and Bentz 1999).

Whitebark pine (*Pinus albicaulis* Engelmann), a long-lived species within the pine subsection *Cembrae*, is typically found just below the alpine timberline (McCaughey and Schmidt 2001). MPB has existed at detectable levels in high elevation whitebark pine stands for some time. Paleoeological records suggest that *Dendroctonus spp.* were present in ecosystems dominated by whitebark pine during the Holocene (Brunelle et al. *submitted*). Dendroecological and written records also indicate that MPB population outbreaks occurred in high elevation stands during the 1930 to 1940 time period (Perkins and Swetnam 1996, Furniss and Renkin 2003 and references therein). Periods of beetle-caused mortality during the 20th century were associated with warmer than average temperatures, and documented in historical records as ephemeral. Following periods of warm temperature, MPB populations returned to endemic levels with the return of cooler temperatures. In general, it was believed that cold temperatures in high elevation whitebark pine forests were not conducive to MPB outbreaks, often resulting in life-cycles requiring three years to complete (Amman 1973).

Many life-history strategies that drive MPB outbreak dynamics are strongly influenced by temperature. Successful MPB reproduction requires death of its host. Host trees, however, have evolved effective response mechanisms against bark beetle attacks (Raffa et al. 1993). Almost all trees respond to bark beetle attacks, but only those with a rapid and sustained response survive. Tree defensive mechanisms can be exhausted if many beetles attack the same tree simultaneously. Outcomes of MPB dispersal and colonization attempts, therefore, depend on competing rate reactions regulating both beetle arrival and host tree resin response (Raffa and Berryman 1979). When emergence from brood trees is synchronous, populations are more successful in overcoming the defenses of new host trees. 'Mass attacks', which increase population success, are driven by emergence synchrony which results from temperature control of life-cycle timing.

In low elevation lodgepole pine forests, a temperature regime that results in univoltinism (one generation per year) and synchronous summer emergence is predicted to be the most

successful for sustaining MPB outbreaks (Logan and Bentz 1999). With no known diapause to synchronize lifestages, MPB seasonality appears to be a direct consequence of nonlinear, stage-specific rate curves (relating temperature and rate of development) and thermal periodicity (Jenkins et al. 2001). Using development rate data collected at a range of constant temperatures, a lifestage-specific model of mountain pine beetle phenology was developed (Bentz et al. 1991) and later refined to include individual variability (Gilbert et al. 2004).

Within the past 5-10 years, wide-scale MPB caused mortality has occurred in high elevation pine stands throughout western North America and ongoing climate shifts are blamed (Logan and Powell 2001). Herein we describe MPB phenology in high elevation, seral whitebark pine forests in the Greater Yellowstone Ecosystem. Using field observations and phenology model predictions we explore how temperature changes may be influencing the ecological relationship between MPB and high elevation pines.

Methods

Research Sites and Data Collection

Phenological data were collected at three high elevation whitebark pine sites with MPB population levels ranging from endemic to outbreak (Table 1).

Table 1. Research sites located within the Greater Yellowstone Ecosystem predominated by whitebark pine.

Site Name	District, Forest	Elevation (m)	Coordinates
Black Butte	Madison Ranger District, Beaverhead-Deer Lodge, MT	2743	N 44.92036° W 111.82649°
Sawtell Peak	Island Park Ranger District, Targhee National Forest, ID	2652	N 44.55579° W 111.44157°
Togwotee Pass	Windriver Ranger District, Shoshone National Forest, WY	2926	N 43.74401° W 110.05324°

In June 2004, emergence cages were placed on ten whitebark pine at each site that had been attacked and infested by MPB in 2003. Emergence cages consisted of a flexible screen stapled over a 0.60 x 0.30 m section of the tree bole, centered at approximately 1.37 m height from the ground, one each on the north and south aspect of the bole. A tube attached to the bottom of the screen enclosure collected all adults that emerged from the tree bole within the sample space. In early June 2005, emergence cages were placed on ten trees at each site that had been infested during the summer and fall of 2004. All cages were checked weekly during the summers of 2004, 2005 and 2006. Due to the remoteness and difficult access of all sites, initial collections often did not occur until after emergence had commenced. Air temperature was recorded hourly at each site throughout each year beginning June 2004 and ending 30 September 2006 (Onset Computer, Bourne, MA, U.S.). Following emergence, bark

underneath each cage was removed and a variety of measurements taken, including the number of pupal chambers and parent adult mortality (Schen-Lagenheim unpublished data). The number of pupal chambers (e.g. new brood adults) was compared to number of adults collected from each cage providing a measure of parent adult re-emergence.

Mountain Pine Beetle Phenology Model

The MPB phenology model (MPBphen) describes development rate (the inverse of time required to complete a stadium) of eight MPB lifestages (ovipositional adult, egg, larval instars 1 – 4, pupae and teneral adult) as a function of temperature. The model was developed based on algorithms in Logan (1988) and parameterized using development rate data collected at six constant temperatures in the laboratory (Logan and Amman 1986, Bentz et al. 1991). Ovipositional adult development is based on equations and data described in Logan and Bentz (1999). Variability in individual development was included in the model based on the age-structured McKendrick-von Forester partial differential model (Gilbert et al. 2004). MPBphen is driven by hourly temperature and predicts the proportion of individuals in each lifestage throughout the lifecycle. The model used here is coded in the MATLAB (Math Works 2006) mathematical language. Field validation in low elevation lodgepole pines indicates the model does a very good job of predicting emergence timing (Bentz et al. unpublished data).

Model Evaluation and Simulations

MPBphen requires an input distribution of ovipositional adults and hourly temperature for the duration of the lifecycle (e.g. up to 3 years). To evaluate model predictions of MPB phenology in high elevation whitebark pine, we initiated the model with an input distribution of ovipositional adults based on observed timing of emergence in 2004 from trees infested in 2002 and 2003. Hourly air temperature data collected at each site were used to drive the model. Based on analyses in Bolstad et al. (1997), 1.8°C was added to each hour temperature to accommodate differences between air and phloem temperatures.

We were interested in using MPBphen to predict adult emergence timing at high elevation pine sites for years without observed temperature measurements. Herein, we present methods and results for only one site, Togwotee. Daily maximum and minimum temperatures were acquired for 10 NRCS SNOTEL stations (<http://www.wcc.nrcs.usda.gov/snow>) that were in close proximity and elevation to the Togwotee site. Using mixed model analyses (SAS Institute Inc.), parameters were estimated to predict daily maximum and minimum temperatures any given year using temperature data collected at the Togwotee site in 2004, 2005 and 2006, and data from the 10 SNOTEL sites. Daily maximum and minimum temperatures at the Togwotee site were then predicted for several years in the range of available data at the SNOTEL sites. A sine wave function was used to predict hourly temperatures from daily maximum and minimum temperatures. Field observations suggest that peak flight at the Togwotee site was centered around the middle of July. Therefore, an input distribution of ovipositional adults based on a normal distribution with a mean date = July 13 and standard deviation = 10 d was used for all simulation runs. Additional temperatures were predicted in a similar fashion for a whitebark pine site near Crater Lake in Oregon using SNOTEL stations at ~1820m.

Results and Discussion

Mountain Pine Beetle Emergence Timing

Patterns of adult emergence from trees attacked in 2004 was similar among the three sites. At all sites, adults emerged from individual trees both 1 and 2 years following attack. There was no consistent trend among the trees in proportion adults emerging each year. Some trees had a majority of 1 year beetles (e.g. univoltine) and other trees at the same site produced a majority 2 year beetles (semivoltine). Early peaks in emergence the first year following attack contained a least some proportion of parent adults that re-emerged. We assume these parents will attack new hosts and produce a second brood cohort, although the fate of re-emerged parents has not been studied in the field.

Using emergence cages similar to those used in the current study, Bentz (2006) observed adult emergence from low elevation lodgepole pine sites to be highly synchronized, with >90% of emergence occurring in a 14 d period. In the current study, at high elevation whitebark pine sites, emergence occurred over a period >60 d, particularly 1 year following 2004 attacks. We hypothesize that long emergence periods at high elevation sites is a function of the influence of temperature on lifestage timing, which in turn influences the overwintering lifestage(s). Similar patterns of dispersed summer MPB emergence were observed in western white pine (DeLeon et al. 1934).

Our results suggest that in high elevation whitebark pine a variety of cohorts may be emerging to attack trees at any given time. Cohorts comprising parent beetles, brood adults developing on a 1 year lifecycle and brood adults developing on a 2 year lifecycle could be emerging to attack new host trees at any given time. Given the large amount of whitebark pine mortality observed at our sites (Schen-Langenheim unpublished data), these results suggest that strict univoltinism, as is often observed in low elevation lodgepole pine sites, is not necessarily a requirement for mountain pine beetle population outbreaks at high elevation sites.

MPBphen Simulations

Using observed timing of beetle attacks in 2004 and observed hourly temperatures in 2004, 2005 and 2006, MPBphen did a good job of describing the proportion of beetles emerging 1 and 2 years following attack (Fig. 1).

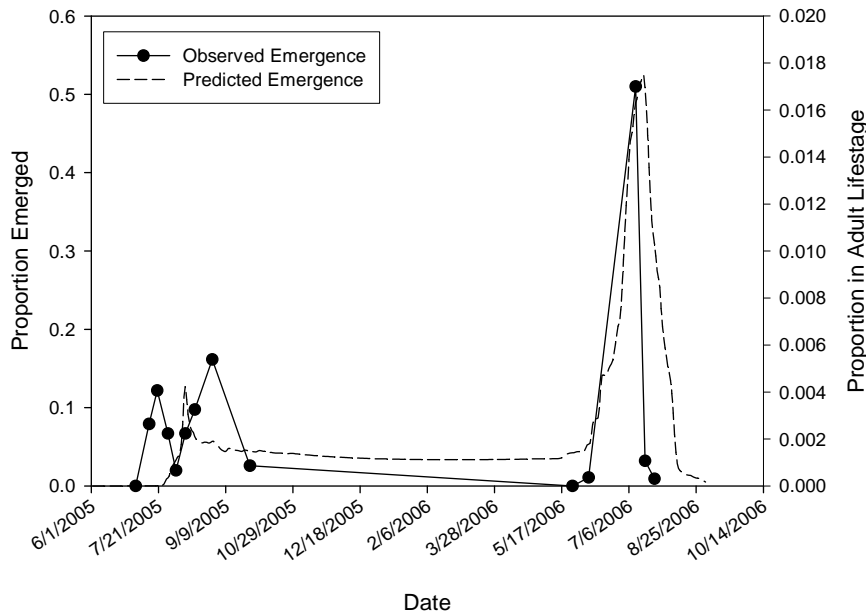


Figure 1. Observed and predicted MPB emergence from whitebark pine attacked in 2004. Proportion MPB emerged predicted using MPBphen and observed hourly temperatures recorded at the Togwotee field site. Observed emergence data collected using emergence cages on infested trees at the Togwotee site.

We then used MPBphen and predicted temperatures to estimate emergence timing at one site, Togwotee, for additional years. Using predicted temperatures to drive MPBphen, results suggest that trees attacked in 2003 would produce 100% 2 year beetles. Field observations indicated that trees attacked in 2004 produced a small proportion of 1 year beetles, with the majority developing in 2 years. However, trees attacked in 2005 were predicted to have a majority of 1 year beetles, with the remainder developing in 2 years. Therefore, MPBphen predictions and observed data suggest that warming temperatures from 2003 through 2006 resulted in a voltinism shift from a majority of MPB developing in 2 years to a majority of the population developing in 1 year. Additional simulations using temperatures predicted for the middle 1970s, when MPB outbreaks in whitebark pine were observed to be at low levels, estimated a large proportion of MPB developing on a 3 year lifecycle (Fig. 2).

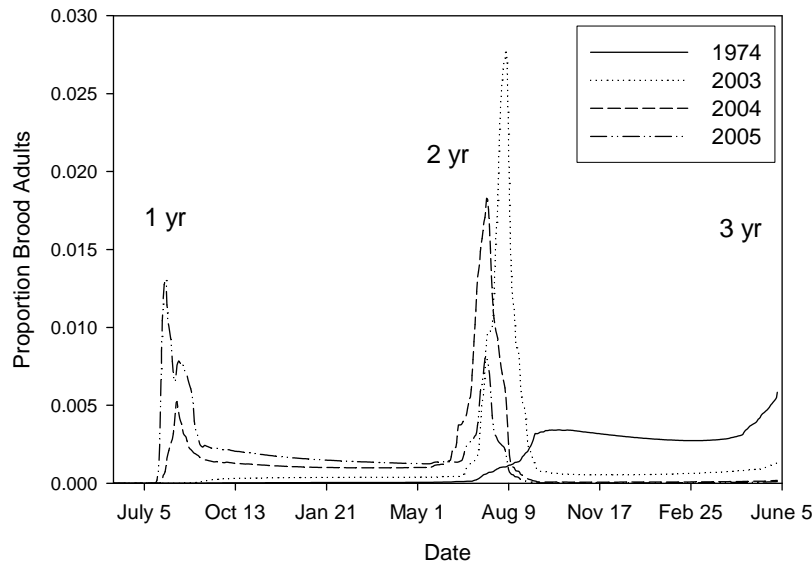


Figure 2. MPBphen predictions of MPB emergence timing at the Togwotee whitebark pine site. Shown are the proportion brood adults emerging in 1 and 2 years by year of tree attack. Brood not emerging in 1 and 2 years are assumed to emerge in 3 years.

Using predicted temperatures for a whitebark pine site at ~1820m near Crater Lake, Oregon, MPBphen predicted trees attacked in 2004 would produce 100% 1 year beetles. Similar results were obtained when MPBphen was run using predicted temperatures for trees attacked in 1989 at the same site. However, USDA Forest Service, Forest Health Protection, Aerial Detection Surveys indicate there has been a significant increase in whitebark pine mortality in Oregon between 1989 and 2004. This suggests that factors in addition to MPB voltinism, such as temperature effects on host trees, are undoubtedly influencing outbreak dynamics in high elevation whitebark pine forests across western North America.

Field observations and model predictions suggest that MPB development at high elevation sites is flexible and highly responsive to changes in temperature. A variety of voltinism pathways allow for population success and subsequent host tree mortality. Additional research is needed to better understand differences in temperature-driven phenology and subsequent attack strategies of MPB at low and high elevation pine sites. Predictions of continued climate change dictate a need for long term research investigating the differential role of temperature and host effects on MPB population success in high elevation whitebark pine ecosystems.

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Fire and Pacific Coast Whitebark Pine

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Abstract

Although whitebark pine ecosystems are widely distributed along the Pacific Coast mountain ranges, their fire ecology is not well known. In the Cascade Range, evidence of historic fires is common and fire return intervals are wide-ranging. Overall, findings from the Pacific Coast ranges indicate that whitebark pine readily recolonize stand-replacing fires. Fire severity is variable, reflecting a mixed severity regime. Perhaps as a result of fire exclusion, late-seral competing species have taken over about 12.5% of former whitebark pine dominated stands in the Cascades. Managers are encouraged to carefully reintroduce fire. Priority should be given to sites characterized by historically frequent non-stand replacing fire. Fire professionals should promote lightning-caused fire whenever possible, plan prescribed fires based on site-specific stand histories, and work with Natural Resource Specialists to protect disease-resistant trees from lethal burning.

Introduction

Since the early 1980's we've become aware of fire's multi-faceted influence on whitebark pine (*Pinus albicaulis*) ecosystems, but nearly exclusively in the U.S. Rocky Mountains. As a keystone process, fire is often responsible for promoting whitebark pine in two fundamental ways. First, fire can limit less fire-hardy competitors (Arno and Hoff 1990). Second, blazes promote regeneration as evinced by Clark's nutcrackers caching seeds in recently burned sites (Tomback and others 1993). Insight regarding fire dynamics and whitebark pine have been revealed primarily through research into fire history, post-fire effects, and succession.

In comparison to the Rocky Mountains, the Pacific Coastal whitebark pine forests support more tree species, endure a different climate, and tend to be more isolated into smaller stands – all important implications for applying fire. Yet little is known about the relationship between fire and these forests.

This paper reviews emerging findings along the Pacific Coast and provides some preliminary recommendations for managing whitebark pine with fire. The conclusions from several studies are offered, including a recent fire history project conducted by Siderius and Murray (2005) which has not yet been published in the scientific literature.

Emerging Results and Discussion

Fire: Commonplace or Rare?

A commonly held view of natural fire, is that it tends to be rare in the high elevations of the Pacific Coast mountains where sparse fuels and lingering snowpacks limit burning. Virtual absence of fire evidence, such as tree scars, is commonly believed – and supports this notion

(Mastrogiuseppe 2003). However, until recently no systematic investigation of fire’s history in the whitebark pine zone had been conducted in this region.

In 2003, we initiated a field study to examine Cascadian fire history and found evidence that fire was a common element of whitebark pine forests in the past (Siderius and Murray 2005). Although fire scars may not be as widespread, compared to the Rocky Mountains, we found a substantial number. The presence of charcoal, either on the ground or on tree relics, was observed at 88% of sites signifying fire’s prevalent influence (fig. 1).



Figure 1— Fire scars and charred wood are common in the whitebark pine zone of the Cascades. (Left: Sawtooth Ridge, Chelan Ranger District (photo by Joel Siderius.) Right: Mission Ridge above Wenatchee, WA (photo by Michael Murray.))

Post-fire Succession

Studies indicate that whitebark pine readily establish the post-burn environment in this region. Perhaps the first formal observation is an

account by Hofmann (1917) on the flanks of Mount Adams, WA. On a large burn (approximately 28,000 hectares) from 1892, whitebark pine were observed reproducing along with noble fir (*Abies procera*) and Pacific silver fir (*A. amabilis*). Firs dominated the new tree cover (66%) followed by whitebark pine (9%), hemlock (*Tsuga* sp.) (9%), and lesser amounts of other species. They formed a fairly uniform distribution.

A fire on Cathedral Peak, Yosemite National Park, CA was examined for post-fire recruitment of whitebark pine (Tomback 1986). This high-severity burn supported significantly more seedlings than the adjacent unburned site. Tomback (1986) surmised that Clark’s nutcrackers were the primary source of regeneration. McDowell (2006) found a negative relationship between fire-induced mortality volume and seedling densities. She also reported higher seedling recruitment at greater distances from the burn edges.

A chronosequence of stand conditions resulting from nine fires was examined in whitebark pine – subalpine fir (*Abies lasiocarpa*) forests of the southern BC Coastal Mountains by Campbell and Antos (2003). They found that both whitebark pine and subalpine fir established well in the post-fire environment. However, by about 269 years post-burn, whitebark was reduced in favor of subalpine fir.

Severity

In examining tree age structure, Campbell and Antos (2003) found both stand-replacing and mixed-severity burns among past fires. The stand-replacing fires were most common. Hofmann (1917) characterized the large Mount Adams Burn as killing “almost all of the timber” except for “occasional patches in such places as near a marsh or spring.” Tomback (1986) observed that the Cathedral Peak fire severely burned a stand of krummholz

whitebark pine. Some live trees were nearly consumed by the flames, leaving only charred fragments.

We examined the most recent fire event at each site we sampled (Siderius and Murray 2005). Based on forty-three fires, we found a mix of high-severity (56%) and non-stand replacing (44%) fires.

Two very small lightning-caused fires at Crater Lake National Park were monitored for mortality after they were suppressed. The Mulligan Fire occurred in a closed-canopy stand of mountain hemlock (*Tsuga mertensiana*) – Shasta fir (*Abies x shastensis*) – whitebark pine in 2002. All five mature whitebark pine in the perimeter were heated by flames. Two were directly killed by torching. One of the three remaining scorched trees was colonized by mountain pine beetle (*Dendroctonus ponderosa*) within days of the fire. By the next year, it was dead.

The Dutton Fire ignited in August 2004 in a timberline mosaic of meadows among small ‘islands’ of whitebark pine supporting a minor component of mountain hemlock and Shasta fir (fig. 2). This fire torched or scorched most trees within the small (.01 ha) island where



Figure 2—The 2004 Dutton Fire, Crater Lake National Park, was a small stand-replacing burn. (Photo by Michael Murray.)

fire was unable to spread in the sparse fuels of the surrounding meadow. Within a month, most of the torched trees were heavily infested with mountain pine beetle, thus a stand-replacing event was the outcome.

Frequency

To date, only one study has examined the historical frequency of natural fires in whitebark pine ecosystems. Siderius and Murray (2005) studied three Cascadian National Parks (Crater Lake NP, Mount Rainier NP, and North Cascades NP) along with adjacent National Forest lands.

Cascadian forests support an impressive range of fire frequency. We detected fire return intervals for every 10-year class up to 160-169 years (fig. 3). Most fires

(67%) occurred at intervals less than 100 years.

However, 18% of plots supported evidence of only a single fire and 14% had only charcoal evidence – suggesting longer (multi-century) fires. Individual burns often fail to leave detectable evidence, so it’s

possible that we missed some events, thus overestimating fire interval lengths. This potential shortcoming is inherent to fire history research (Agee 1993).

Overall, we found a negative correlation between latitude and frequency of non-stand replacing fires. This modest correlation is significant ($r_s = .453$ at $P = 0.10$) and indicates that fire intervals shorten with higher latitudes (fig. 4). This may be explained by lightning pattern and the tendency of northern forests to support more complete understory vegetation cover providing surface fuel.

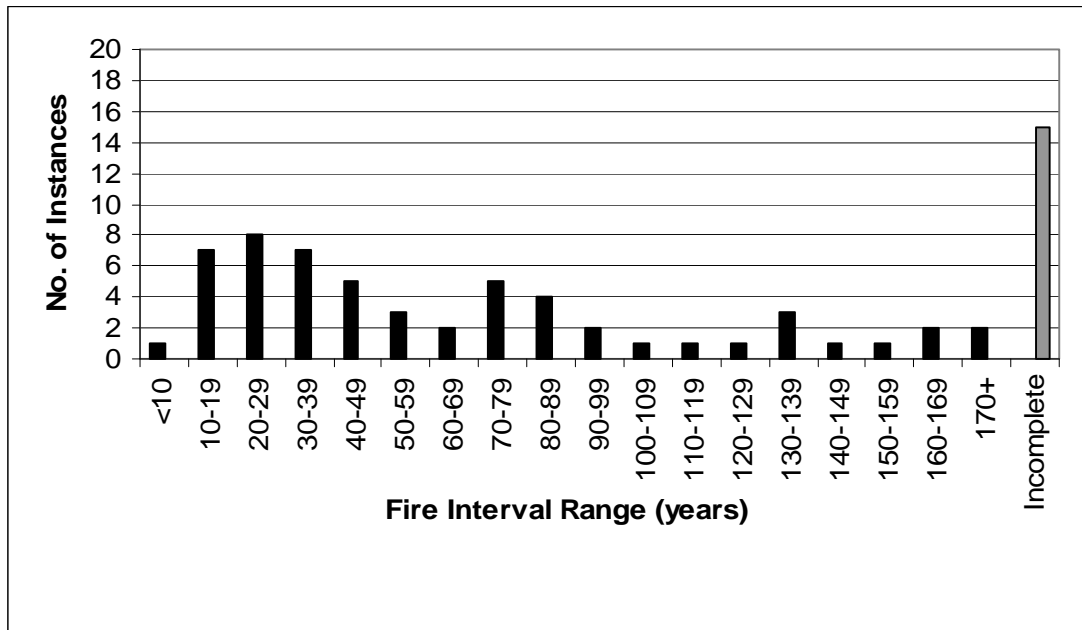


Figure 3—Distributional frequency of fire intervals. Last bar denotes instances where only a single fire was detected – incomplete interval (Siderius and Murray 2005).

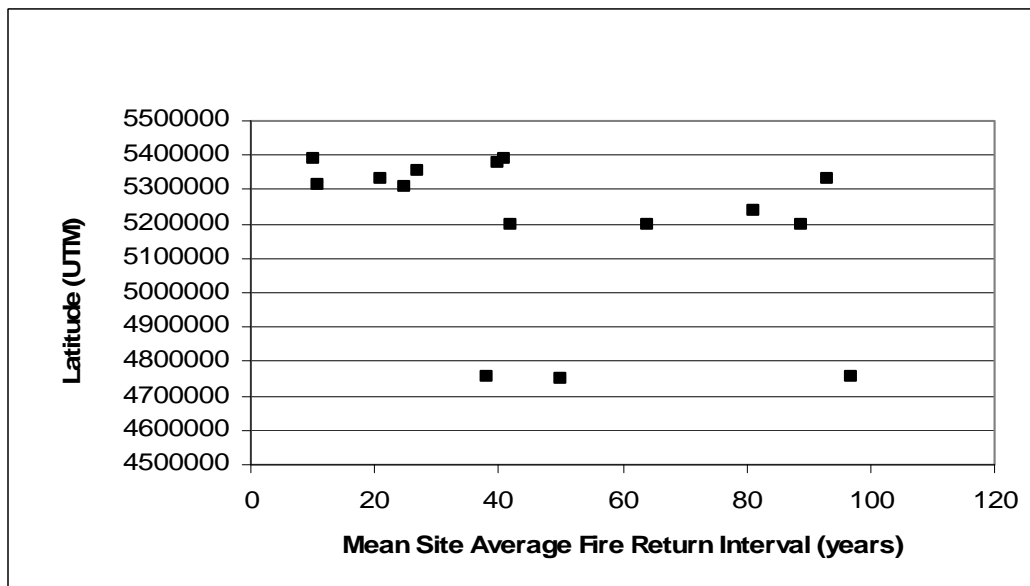


Figure 4—A moderate correlation between latitude and fire intervals in the Cascades based on three study areas (Crater Lake, Mt. Rainier, and North Cascades).

Comparison with Rocky Mountains

In comparing Cascadian fire ecology with the Rocky Mountains, the similarities are notable. The tendency of whitebark pine to readily colonize and develop significant volume in burned sites is well-documented in the Rockies (Arno and Hoff 1989; Murray 1996; Tomback 1994). On most acreage that is not too high and harsh for competitors, late-seral species gradually

increase in proportion to whitebark pine in the absence of disturbance (Keane and Arno 1993; Murray and others 2000). Another similarity is the mixed-severity regime (Arno 2001), although as indicated above (Severity section), Pacific Coast fires tend more towards stand-replacing behavior. Fire return intervals are extremely wide-ranging in both regions. For example, in the Rockies, Barrett (1994) found non-stand replacing events to range between 66 and 204 years. Murray (1996) found these fires every 42 – 256 years. However, Cascadian fires appear to be skewed towards shorter intervals, at least in the north where frequencies *average* as low as 10 years northeast of Winthrop, WA (fig. 5). Examining two similar covertypes, the mean frequencies are alike (Table 1). It should be noted again that many whitebark pine stands have evidence of only a single fire and a small percentage have no dateable events. Whether these instances represent longer intervals or where fire simply failed to leave evidence is not known. In general, mixed severity regimes are especially challenging to sample, characterize, and convey to an audience (Agee 2005).

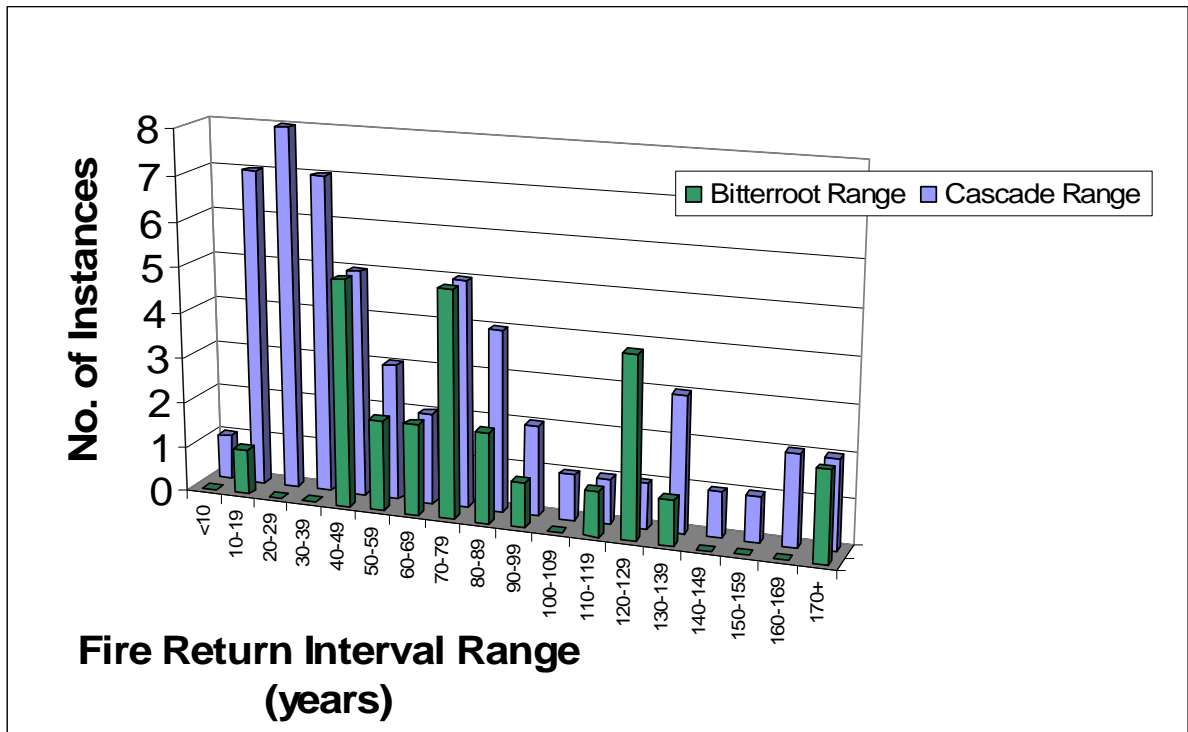


Figure 5—A comparison of fire interval distributions between Bitterroot (Murray 1996) and Cascade (Siderius and Murray 2005) regions.

Table 1—A comparison of average fire return intervals between the Bitterroots (Murray 1996) and Cascades (Siderius and Murray 2005).

Overstory	Location	No. of Plots	Mean Fire Return Interval (years)
Pure Whitebark Pine	Cascades	11	85
Pure Whitebark Pine	Bitterroots	3	96
Whitebark Pine – Lodgepole Pine	Cascades	9	73
Lodgepole Pine – Whitebark Pine – Douglas-fir	Bitterroots	64	92

Are whitebark pine declining due to lack of fire?

Fires have diminished since the early 19th century in the Cascade Range (Siderius and Murray 2005). During the time period of 1800-1900, we found 51 fire events. The period of 1900-2000 supported only 22 fires. This represents a decrease of approximately 57%. Few recent fires (post-1950) were documented in this study.

The marked decline in fire may be yielding an increase in the abundance of fire-sensitive late-seral tree species and a decrease of openings conducive to whitebark pine regeneration. By applying stand reconstruction methodologies, we estimate that late-seral species, especially Shasta and subalpine fir, have increased volume at dramatic levels. Since 1924, these two species have increased 4181% and 2888% respectively. Surprisingly, whitebark pine has also increased about 280%. This result may be due to recovery from widespread fires in the late 1800s, shortcomings in the spreadsheet model, and/or high growth rates for saplings/seedlings. Estimates of stem area volume alone may not be the best measure of historical abundance. Regardless, our results indicate late-seral species have taken over 12.5% of former whitebark pine dominated stands. Therefore, although fire exclusion has not led to the reduction of whitebark pine, it is likely allowing competing species to replace whitebark pine as the dominant species. This trend has already been observed in the Rocky Mountains where subalpine fir is actually reducing whitebark pine through competition (Keane and Arno 1993).

Recommendations for Fire Management

Opportunities for Lightning-caused Fires

With the knowledge that fire has been an important historic component of Pacific Coast whitebark pine forests, it’s time to carefully consider where and how to reintroduce this missing element. When presented with the opportunity, *allowing lightning-ignited fires to burn whitebark pine stands is the preferred scenario for re-introducing fire* (fig. 6). First, the location is determined by nature, and not biased by human selection. Since most whitebark pine is remote from human development, there is rarely a hazard. Second, the pattern of fire will be more natural. Management-ignited fires tend to create a much larger ignition area (e.g. aerial ping-pong balls, drip torch striping) than would occur naturally. This can result in

extensive stand-replacing burns which may not be typical for the target area's fire regime.

Third, lightning fires occur at the time of year when forest communities are best adapted to burn. Thus the appropriate seasonality is achieved. Fourth, lightning-ignited fires are typically much less expensive to manage than prescribed or suppression fires (USDA 2006).



Figure 6—Attendees of this conference's field trip observe a fire at Crater Lake NP as it reaches the whitebark zone. Started by lightning on July 23, 2006, this fire (Bybee Complex) was managed as "wildland fire use for resource benefits." (Photo by Michael Murray.)

Unfortunately, due to management and political constraints, the vast majority of lightning fires, even in remote whitebark pine habitats continue to be suppressed in Canada and the U.S. (CFS 2006; NIFC 2006). Relying solely on the occasional administrative approval of a lightning fire may not reintroduce fire at a meaningful scale. Thus, prescribed fire, if carefully applied, can provide ecological benefits to whitebark pine ecosystems (Arno 2005).

Prescribed Fire

Given the inherent high variability of fire in these ecosystems, local Natural Resource Specialists should tailor prescriptions with Fire Managers to match site-specific regimes. Evaluating fire history and fuel conditions of individual stands prior to any management-ignited burning is critical for the appropriate application of fire in these sensitive forests.

Priority should be given to sites characterized by historically frequent non-stand replacing fire.

It's likely that these stands are most affected by decades of fire exclusion. Moreover, fire behavior is less risky to manage than stand-replacing regimes. Conversely, applying human ignitions in stands with little or no evidence of historic frequency may actually do more harm than good to the ecological integrity of the stand.

To select and define burn units, Natural Resource Specialists should first assess candidate stands for visual clues indicating frequent fire which include extensive grass or shrub cover and fire scars. Stands with low fire frequency are often characterized by mountain hemlock, sparse understory vegetation and/or exposed rocks or mineral soil (often pumice).

Don't Burn Disease Resistant Trees

Fire managers should always be cognizant of the unique status and threats to whitebark pine – a ‘sensitive species.’ Fire-induced mortality of potentially disease resistant trees should be strictly avoided. They are the life-link to the species’ future. Where blister rust infection is high, it’s strongly recommended that pathologists or trained technicians survey for healthy mature trees prior to burning operations or during lightning-fires (safety permitting). Managers can protect these trees from flames by mitigating ladder and surface fuels in their immediate vicinity. Depending on the particular fire incident, other options such as water dropping, wet lining, or foil wraps could be considered.

As an example, during the 2006 Bybee Complex Fire, a two-person crew searched for healthy trees as the fire reached the whitebark pine zone. Twenty-six healthy mature



individuals were identified as possible disease resistant trees as the fire crept close by. Beneath each tree, we removed dead and down fuel, and then established hand lines near each tree crown’s drip line (fig. 7). Fire Managers were very supportive and none of the candidate trees suffered fire injury.

Figure 7—The 2006 Bybee Complex Fire threatened about 26 disease-resistance candidate trees. For this tree, we removed woody debris and constructed a handline beneath its canopy to the unburnable meadow edge. (Photo by Michael Murray.)

Information Gaps

Overall knowledge of fire effects, size, spatial pattern, and seasonality is lacking for the Pacific Coast ranges. Investigation of historical frequency in California, British Columbia, and the Olympic Range will prove useful. Questions remain pertaining to the relationships between fire and blister rust disease incidence, and its alternate hosts. Whitebark pine’s tolerance (survivorship) of fire under various environmental conditions is poorly understood. Improved knowledge of interrelationships between mountain pine beetle and fire is also needed.

Conclusions

Fire is an historical influence on whitebark pine of the Pacific Coast mountain ranges. A mixed-severity regime with a broad range of fire return interval lengths indicate complex relationships between fire and the pine. Late-seral competitors appear to be overtaking whitebark pine at some locations. Re-introducing fire can benefit whitebark pine, but it must be undertaken with extreme care. Managers should promote lightning-caused fire whenever possible and plan prescribed fires based on site-specific stand histories. In the face of a non-native plague, protecting healthy and potentially disease-resistant trees from lethal burning is imperative.

Acknowledgement

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The Coevolution of Whitebark Pine and Clark's Nutcracker

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Coevolution is said to be occurring when two (sometimes more) species interact in such a way that the evolution of both is affected, due to selective pressures each exerts on the other. This process shapes evolutionary changes in such interactions as predator/prey, seed/seed disperser, and parasite/host situations, in which each participant in the interaction exerts natural selective pressures on the other(s).

A coevolutionary relationship can be direct, as when two species affect each other; or diffuse, when a species' evolution is shaped by numerous other species, which are in turn affected by it. The former is exemplified by the coevolved mutualism between whitebark pine (*Pinus albicaulis*) and Clark's Nutcracker (*Nucifraga columbiana*); the latter by the mutualism between the tropical tree *Caesaria corymbosa* and its numerous seed dispersers which include woodpeckers, flycatchers and vireos.

A mutualism is an interaction between species in which each contributes essential benefits to the other. As a default position, it can be argued that mutualisms are of necessity coevolved. Another possibility that might account for a mutualistic relationship is the coadaptation of two species. This implies that the species had already evolved traits that allowed a mutualism to come about between them when they fortuitously encountered each other. However, such a chance meeting would probably rapidly become coevolutionary.

The mutualism of whitebark pine and Clark's Nutcracker exemplifies that which involves the other stone pines of *Pinus* subsection *Cembrae* and the Eurasian Nutcracker, *N. caryocatactes*. The nutcracker regenerates the pine by caching its seeds in the soil as a stored food and failing to eat all of them, or feed them to its young. Those that germinate form the only reliable nucleus of a new generation. In honing the relationship between these species the pine has been caused to deviate from its progenitor white pines in the morphology of its seeds (large and flightless), cones (non-opening, breakaway scales that clasp seeds, sessile attachment), branching (verticalized cone-bearing branches, limb forking to amplify cone production and form a display surface), and in the variable dormancy of its seeds (postponing germination and allowing the nutcracker's food source to remain available).

The other members of subsection *Cembrae* are the Swiss stone pine (*P. cembra*), Siberian stone pine (*P. sibirica*), Korean stone pine (*P. koraiensis*), and Japanese stone pine (*P. pumila*). Except perhaps for *P. pumila*, which some investigators believe is misplaced in this subsection, the other stone pines share with whitebark pine, to a surprising degree, the morphological modifications described above. In addition, no other species in this large and

variable genus share the cone modifications that characterize the stone pines. This is true even of other bird- or mammal-dispersed pines like limber pine (*P. flexilis*) and the pinyons (subsection *Cembroides*) that share some seed and form characters.

The nutcracker has deviated from its *Corvidae* progenitors by evolving an exceptional memory, a powerful bill, a unique sublingual pouch for seed transport, and a suite of behavioral traits that optimize its seed foraging activities. The dependency of the pine on the nutcracker for its regeneration, and of wildlife species not a part of the mutualism for pine seeds, demonstrate the indispensability of biodiversity in maintaining ecosystem integrity. This mutualism also spotlights how a lineage of passerine birds has engendered phylogenetic diversity in an ancient lineage of conifers.

Can the coevolution of whitebark pine and Clark's Nutcracker be demonstrated as actually having occurred, or is it a presumption? Playing Devil's Advocate, I would argue that all relationships viewed as coevolutionary are indeed presumptions which by their nature cannot be demonstrated. That is because the genetic changes required of each interacting species, probably the fixation of mutant genes, cannot be directly observed. And even if they could, each such change could only be presumed to be caused by selective pressure from the opposite participant, and not from some other organism or factor acting in parallel. What basis, then, have we for labeling the pine-corvid relationship coevolutionary?

The answer, I believe, lies in the empirical evidence of modification from white pine progenitors on the one hand, and corvid progenitors on the other. Each of the white pine modifications figures in the foraging by nutcrackers for pine seeds. It appears highly improbable that such a suite of characters could arise one by one over the millions of years of this interaction without a natural selective impetus. The same holds true for the nutcracker characters that depart from those of other corvids. Things fit together too nicely and work too efficiently for us to ascribe them to coincidence with any confidence.

A few words should be said about the ecological effects of the mutualism of corvids and stone pines. Whitebark pine is a pioneering species, usually seeded by nutcrackers in burned areas or other openings. Many are sown by nutcrackers in the shade of the forest as well, but those that begin their lives in the open are more likely to survive to maturity. As a result, the whitebarks that come up in the open are in a position to materially modify their environment, from a treeless area to one supporting a woodland of scattered, broad crowned, low-branched trees or tree clumps with large cone-bearing capacity.

The modified microclimate they create by providing shade, reducing wind speed and raising relative humidity permits the establishment of wildflowers, shrubs and forbs absent from the openings on their margins. Birds and mammals that find the openings inhospitable, move into the woodland groves. Eventually, shade tolerant conifers – firs and spruces – form an understory and eventually replace the whitebark pine canopy. On very exposed sites whitebarks may provide the only tree cover for centuries. Whatever the ultimate outcome, whitebark pine is a species of uncommon value, and its disperser, Clark's Nutcracker, is complicit in all that the pine brings about. Thus it is the landscape and its biodiversity that are the legacy of this instance of coevolution.

Climate Variability in Whitebark Pine Ecosystems: A Paleoperspective

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There is a broad consensus within the scientific community that temperatures over the most recent 50 years have been warmer than in perhaps the past millennium, due at least in part to rising greenhouse gas concentrations brought about by human activity. What remain uncertain are the impacts any potential warming might have on sensitive habitats. In particular, it is likely that increases in temperature will lead to shifts in disturbance regimes such as fire and insect outbreaks. Here I review our current understanding of climate variability with an emphasis on the potential impacts on whitebark pine. I will also discuss the importance of whitebark pine in better understanding climate variations via the development of reconstructions of past climate. In this respect, whitebark pine affords a great deal of opportunity. Since it is long-lived and grows near the physiological limits of tree growth the variations in its ring-widths are sensitive to changes in climate. Whitebark pine has been little used for the reconstruction of climate, but the growth characteristics have been shown to capture decadal scale temperature variability effectively. The application of tree growth information from whitebark pine to infer climate variations also has a number of important challenges. For example, it appears that rising temperatures over the course of the 20th century have resulted in a shift from temperature as a primary limiting growth factor to a more mixed signal whereby moisture conditions have become more limiting. The mechanisms responsible for this shift are currently unclear. It is critical to consider the role of climate variation in development of management plans meant to preserve this critical species and its unique habitat.

Whitebark Pine in the Pacific Northwest: What's Next?

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Abstract

Conservation and restoration of whitebark pine are two important challenges facing us in the immediate future. We will present our approach for a whitebark pine conservation strategy which will focus on Federal lands in Oregon, Washington and California. This management plan will identify the actions needed to reduce the threats to the continued existence of whitebark pine throughout the planning area. Following the completion of the strategy, we will develop a land manager's guide for restoration and managing whitebark pine. This manual will describe site assessments, provide a decision-making guide for setting stand level prioritization, and explain restoration techniques. In our presentation we will review these restoration techniques and discuss the challenges of their application to the highly variable whitebark pine stands in the Pacific Northwest.

Introduction

Conservation and restoration of whitebark pine are two important challenges facing us in the immediate future. Today, our previous speakers have provided us with an overview of whitebark pine ecology, distribution of the species in the Pacific Northwest, and an introduction to its status with respect to white pine blister rust. While we realize that in most of our areas, mortality and infection rates are less severe than in the Rocky Mountains, we still feel that it is time for us to take action to protect and restore these communities.

Whitebark pine is distributed in high-elevation areas just above and below forest line. While this habitat is a very small component of the Pacific Northwest landscape, most of these areas are under federal ownership. In Washington, about 5% of our landscape is above forest line; 66% of these areas are managed by the US Forest Service and 27% by the National Park Service (Rochefort et al. 2006). Although Oregon has much less area in the subalpine and alpine zone, the trend is still the same – 88% is managed by the US Forest Service and 6% by the National Park Service. Based on our joint ownership of these areas, we feel that we (i.e. our agencies) have a primary responsibility to encourage and facilitate research on whitebark pine and to develop strategies for long-term protection of the species. This afternoon, we would like to provide you with a brief overview of some of the inter-agency activities that we have been involved in. We would also like to share with you some of the challenges that we see before us with respect to whitebark pine restoration.

Development of Conservation Strategies

Through the efforts of many individuals, we have conducted health assessments, updated the whitebark pine range map for Oregon and Washington, collected seed from stands across the

region, and conducted molecular genetic analysis and blister rust resistance testing. Our next step will be the development of a conservation strategy for whitebark pine in the Pacific Northwest and a restoration guide for land managers.

Our conservation goal is to provide a high likelihood that whitebark pine and associated species will exist, well-distributed, throughout the Pacific Northwest. There are a number of large scale issues to be considered. Given the critical role that whitebark pine seed plays in the grizzly bear diet, we will put special emphasis on the North Cascades Grizzly Bear Recovery Area (Grizzly Bear Outreach Project 2005). Other regional scale issues include patterns of genetic variation which will be used in determining seed transfer guidelines, delineation of local populations, important for connectivity among populations, and patterns of blister rust resistance across the landscape. The two key objectives of the conservation strategy are to 1) define conservation units, and 2) prioritize these units for conservation and restoration. We were inspired by the approach used by The Nature Conservancy to create a blueprint for conservation (The Nature Conservancy of Canada. No date.). In this approach, a portfolio of sites is selected for the area under consideration that provides an effective and efficient network at the least cost. There are 6 steps to build a site-based conservation strategy:

1. Define the planning area
2. Prepare a comprehensive base map
3. Collect relevant ecological information.
4. Develop criteria to apply to each planning unit.
5. Select a portfolio of sites based on these using a computer model.
6. Critically analyze the results.

The criteria (step 4) will include grizzly bear habitat, wilderness designation, level of blister rust infection, the level or trend of mountain pine beetle infestation, acres burned versus historic, and access (miles from the nearest road or trailhead) among others.

The Natural Conservancy developed a computer model for site-portfolio selection (The Nature Conservancy 1999). An excellent example of the application of this model, called the SITES model, is the plan for conservation of threatened and endangered species along the Santa Clara River (Court et al. 2000). Since the symposium, we have become aware of a model that is even better suited to our goals. The Ecosystem Management Decision Support (EMDS) system, developed by the U.S.D.A. Forest Service is “an application framework for knowledge-based decision support of ecological assessments at any geographic scale” (Redlands Institute. No date.). This approach will allow us to prioritize the sites for conservation and restoration and display the network of sites in GIS. Site prioritization is essential for the allocation of the limited funds available. The conservation strategy will be completed by June 2008.

Once the portfolio of sites has been selected, managers will need information on restoration options and techniques. The *Land Manager’s Guide for Restoring and Managing Whitebark Pine* will provide tools for the conservation and, where appropriate, restoration of whitebark pine. It will be formatted after a landowner’s guide for Oregon white oak (Pacific Wildlife

Research 2004) and will include direction on site assessments, restoration planning and technique selection. The guide will be completed by September 2008.

Development of Restoration Strategies

Management Philosophies

One of the first steps in developing a restoration strategy or plan is to identify broad goals or management philosophies for the area. Setting broad goals for a site regarding plant communities, spatial scales, and ecological processes actions is essential to a successful project. Broad goals establish a foundation upon which to create more detailed objectives. Two basic questions that should be addressed at this stage are:

- 1.) What are your management objectives?
- 2.) What spatial scale are you addressing in your restoration strategy?

In a very broad sense, management objectives are developed following one of two management philosophies: Are you preserving whitebark pine? Or Are you mitigating for anthropogenic influences? While these two philosophies sound similar, they are really defining different desired future conditions of the site. If your goal is to preserve whitebark pine, then you are defining your management goal in terms of a plant community or state. If however, you are removing anthropogenic influences, then you are focusing your management actions on restoring ecosystem processes. National Park Service Management Policies (National Park Service 2006) address these goals by stating: "...The Service will not attempt to solely preserve individual species (except threatened or endangered species) or individual process; rather it will try to maintain all the components and processes of naturally evolving park ecosystems, including the natural abundance, diversity, and genetic and ecological integrity of the plant and animal species native to those ecosystems. Just as all components of a natural system will be recognized as important, natural change will also be recognized as an integral part of the functioning of natural systems." (National Park Service 2006, chapter 4.1).

In the case of whitebark pine and blister rust, these two management objectives are often compatible and in fact subsets of one another. In the Pacific West, the primary threat to whitebark pine survival often is blister rust so, by mitigating the anthropogenic influence (i.e. blister rust introduction) we are actually preserving whitebark pine. However, this is not always the case. To illustrate how your restoration prescription could be very different depending which on management objective you select, I would like you to imagine a whitebark pine site. The site is located in the subalpine parkland and is dominated by subalpine fir (*Abies lasiocarpa*) and whitebark pine (*Pinus albicaulis*). Mature trees are growing in scattered clumps and regeneration is a mixture of whitebark pine and subalpine fir; subalpine fir is the dominant species in both the overstory and regeneration. The open stand is very close to a closed canopy forest and both stands contain dead and infected whitebark pine trees. Approximately 20% of mature whitebark pine trees are infected with blister rust and about 25% appear to have died from it. Many mature whitebark pines still appear in good condition and to be producing cones. The site is a seral whitebark pine site (e.g. rather than a climax whitebark pine site) so, next you try to discern the disturbance regime that resulted in the two stand ages. You cannot find any signs of fire, but you notice

an avalanche chute and believe that the younger stand developed following an avalanche that probably occurred at least 200 years ago. So what is your restoration prescription?

If you are preserving whitebark pine, you will look at the site and ask, is whitebark pine survival threatened on this site? Subalpine fir is dominating both the overstory regeneration. In many areas, the openings between tree clumps are closing and suitable habitat for whitebark pine seedling establishment is decreasing. In addition, blister rust is present and you are not certain if the density of regeneration present is sufficient to sustain the population. In this case, you may decide that you want to maintain the open natural of the mixed-species stand and not allow it to succeed to a subalpine fir, closed-canopy dominated forest. In order to preserve whitebark pine, you decide to: remove small pockets of subalpine fir to create openings for whitebark pine regeneration, propagate and plant whitebark pine seedlings in the openings, and continue to monitor whitebark pine for blister rust infection and mortality. In the future, you may create more openings and hope to identify rust-resistant trees from which to propagate more seedlings.

If you are preserving natural processes and mitigating for anthropogenic influences, you may prescribe a different management strategy for the site. The site is a seral community that will naturally succeed to a closed-canopy subalpine fir forest. The site was originally cleared following an avalanche and you do not believe that current management has decreased the frequency of avalanches. When you ask the question, what are the human influences to the ecosystem, the answer is blister rust. Although the density of whitebark pine seedlings is decreasing you must decide if this is a result of blister rust or due to a reduction in suitable habitat caused by canopy closures. If density is decreasing because of canopy closure, then you may choose to continue to monitor the site and accept that it will succeed to a subalpine fir stand, with little whitebark pine. However, if you determine that seedling density is decreasing because blister rust has caused a reduction in cone production or high mortality of seedlings, then you may decide to plant whitebark pine seedlings, but not create new openings. In this scenario, you are trying to insure that whitebark pine is still in the stand so that when a natural disturbance occurs, the species still has the resilience to respond to the disturbance as it would have without blister rust.

As you can see by these two examples, the decision to protect state or process is not an easy one. While the previous examples highlighted philosophical differences, practical considerations such as funding, adjacent resources, or recreational opportunities must also be considered at each site. Sometimes, your restoration strategy for a site may be the same even with different management goals.

Spatial Scales

In addition to identifying your management philosophies, a restoration plan should also define the spatial scale that you are working on. There are three spatial scales that we feel we work on: landscape or regional, mid-level, and stand level. Landscape scale strategies focus on geographic areas such as the range of whitebark pine or whitebark pine in the Cascade Range. Although the boundaries of these areas may vary with different plans, they are based on ecological attributes rather than ownership or management responsibilities. The Conservation Strategy that Carol is working on is an example of a plan aimed at the

landscape scale. Mid-level spatial scales are management defined rather than based on ecological characteristics. Mid-level scales may reflect a particular agencies “sphere of influence” such as all National Parks in the Pacific West Region or all US Forests in Region 6. Stand level spatial scales are based on both ecological and management considerations. A stand is the scale at which many restoration projects are focused due to our ability to manage a specific area. If we decide to plant whitebark pine seedlings or manually remove competing vegetation to create openings for regeneration, we are probably working on the stand level. If we are restoring fire to an ecosystem, we could be working on any one of these three levels.

Depending on the spatial scale you are addressing, the detail of the information you must address may change. For instance, if you are working on a landscape scale, you may state that you would like to protect naturally evolving levels of genetic diversity in whitebark pine populations. However, when you develop plans for stand level restoration, you may need to address the mechanics of this strategy. If you have a stand that is still producing cones, but no regeneration is survival due to lack of suitable habitat or predation you may decide to propagate seedlings from seeds collected on site. In this case, you determine the number of cones collected and number of trees from which to collect the cones to protect genetic diversity and based on you prediction of survival rates. However, if your site has limited regeneration because there is no cone production, how far from this site to you travel to collect cones? When do you decide that genetically resistant stock is your preferred seed source and how does this influence genetic diversity? This is just one example of how the spatial scale will influence the details in your restoration plan or strategy. Identification of spatial scale is an important factor in the development of restoration strategies and plans because it sets the geographic boundaries by which we will plan and evaluate our actions in the future.

Stand Level Restoration Methods

Initially, you conduct a stand analysis in which basic characteristics of the stand are recorded: species composition, whether it is a seral or climax stand, stand structure, stand health (e.g. blister rust, beetles, other diseases), and disturbance regime (what is the primary disturbance, frequency, and has it been altered). At some point, you should identify your management objectives and philosophies. Theoretically, you should know your management philosophies before you conduct the stand exam (e.g. such as NPS Management Policies), but practically, these often evolve as you conduct the stand exam and identify restoration alternatives. In the site review, you should document what the impediments are to whitebark pine survival – lack of cone production, lack of suitable habitat for regeneration, seedling mortality, cone predation, beetles, blister rust, interrupted fire regime, tree competition. Then you must review what management zone your site is in and how do management policies influence your selection of management goals or restoration methods. Is the site in designated Wilderness or a front country, developed zone? Is the site within endangered species habitat, a historic landscape, or the urban interface? Are there other policies that restrict your selection of restoration strategies? Last, you can select your restoration tools. Some of these tools include removal of competing trees (manually or with fire), cone collection, seeding, planting of seedlings, removal of branches with cankers, and introduction

of fire. All restoration plans should include monitoring and reporting to insure that adaptive management will be informed by scientific data.

Conclusion

This morning, and over the next few days we expect to hear more facts and figures regarding infection and mortality rates. Although we could be discouraged by these numbers, we are also optimistic. We are optimistic because so many people have been able to conduct surveys to provide us with accurate estimates of blister rust infection and mortality rates. We are optimistic because the Pacific West whitebark pine stands are as diverse as our rugged topography. The diversity that our whitebark pine stands encompass includes species composition, successional stage, disturbance regime, and white pine blister rust infection and mortality rates. We are encouraged because there are so many tools for us to use to conserve whitebark pine. We feel that based on the diversity of our stands and individual stand histories, that restoration strategies that are mildly successful in some areas may be very successful in other areas and this will allow us to adapt and develop successful management techniques for all areas. However, we do not want this feeling of optimism to allow us to relax – there is urgency to our mission. Global climate change may facilitate a faster progression of blister rust infection at high elevations than in the past. Beetles life cycles may shorten and allow beetle-caused mortality to increase. Fire regimes are expected to change and we cannot predict how this will influence the blister rust cycles in our areas. This conference has helped to strengthen our whitebark pine networks within the west and we hope that we can continue to share information and develop successful whitebark pine protection strategies.

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Preparing the Landscape for Invasion – Early Intervention Approaches for Threatened High Elevation White Pine Ecosystems

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White pine blister rust is now a permanent resident of North America. The disease continued to cause tree mortality and impact ecosystems in many areas. However, not all high elevation white pine ecosystems have been invaded; the pathogen is still spreading within the distributions of the whitebark, limber, foxtail, Rocky Mountain bristlecone pine and has yet to infect Great Basin bristlecone pines. While the heavily impacted areas are in need of immediate management to restore ecosystem function, management of the threatened areas to position them to avoid development of severe impacts upon invasion is also an immediate need – as the disease intensifies, time is running out (Schoettle 2004b).

Increasing the frequencies of durable resistance or tolerance traits within tree populations is accepted as a promising avenue for the co-existence of native tree species and non-native pathogens. Two of the five strategies for proactive management of high elevation 5-needle pine ecosystems (Schoettle and Snieszko, *in press*) will contribute to evolutionary change toward greater resistance in the pine species:

- (a) Increasing the frequency of resistance through introduction of resistant stock, via artificial regeneration, to sustain forest cover (Option 4 in Schoettle and Snieszko, *in press*) and
- (b) Accelerating natural selection for resistance through age class diversification to sustain populations (Option 5 in Schoettle and Snieszko, *in press*).

Artificial regeneration with resistant pine stock is a well accepted strategy for management of many pathosystems including the white pine blister rust system (Samman et al. 2003). A proactive application of this strategy requires that resistant stock be planted before or soon after pathogen invasion, thereby lowering the risk of ecosystem collapse by reducing the window between overstory mortality and reproductive maturity of the resistant outplantings (Option 4 in Schoettle and Snieszko, *in press*). The resistant genotypes can be identified from areas that have experienced extensive pathogen-caused mortality and transferred to the target management area or identified from screening of on-site genotypes using standard techniques (i.e. Danchok et al. 2003). Both approaches require considerable resources: the first requires a thorough understanding of geographic variation in physiological traits, focused family rust-

resistance screening and validated seed transfer guidelines while the second requires extensive genotype rust-resistance screening.

Early intervention to accelerate the selection for rust resistant individuals while sustaining ecosystem function may be an effective, affordable alternative for high elevation 5-needle pine forests (Option 5 in Schoettle and Sniezko, *in press*). Generation of a mosaic of diverse age classes will provide trees at a highly susceptible life stage (young) to promote rust-resistance selection within that cohort while the older trees hold site occupancy and provide ecosystem services during pathogen invasion (Schoettle, 2004b; Schoettle and Sniezko, *in press*). Accelerating the generation time and natural selection process through silvicultural treatments will reduce the ecological consequences of mortality in any one cohort and increases the potential for development of durable resistance within the population while maintaining broad genetic diversity.

Both of these strategies, applied singly or together, will increase the frequency of resistance on the management unit as well as benefit near-by populations through gene flow. Increasing resistance on accessible portions of the landscape will, over time, improve resistance in the next cohort of pines on neighboring harsh, inaccessible yet highly valuable areas. In heavily impacted areas or high hazard areas, populations may need to be supplemented with resistant stock. However, if sufficient resistance is in the native population, and the hazard or infection level is low, stimulating natural regeneration may be sufficient to increase the proportion of resistance to sustain ecosystem function in the presence of the pathogen.

A program to support the application of these strategies in the threatened Rocky Mountain bristlecone pine and limber pine forests of the Southern Rockies is ongoing. White pine blister rust is in the early stages of invasion in these susceptible high elevation ecosystems (Schoettle 2004a). To aid in the development and testing of silvicultural prescriptions for this objective, spatial and temporal dynamics of regeneration of limber and bristlecone pine are being defined. To improve decision making ability and prioritization of intervention, the frequency of rust resistance is being estimated for bristlecone and limber pine populations of the Southern Rockies. The geographic distribution of adaptive traits is also being studied. This information can be combined with climate change estimates to improve seed transfer guidelines and outplanting procedures. Guidelines for selecting putatively resistant seed trees are being developed for low rust incidence areas. Silvicultural prescriptions to stimulate regeneration are being developed, implemented and tested for limber pine and Rocky Mountain bristlecone pine forests on the San Isabel National Forest (Colorado). Seed collections have been initiated to archive genetic diversity and prepare for artificial regeneration projects to restore impacted areas of the Medicine Bow National Forest (Wyoming).

Both the artificial regeneration and the stimulation of natural regeneration strategies have been recommended for application in whitebark pine ecosystems (Schwandt 2006). Likewise, maximizing population size through natural regeneration is among the recommendations for management of small Port-Orford-cedar stands threatened by *Phytophthora lateralis* in northwest California and southwest Oregon (Frank Betlejewski,

personal communication (Interregional Port-Orford-Cedar Program Manager, SW Oregon Forest Insect and Disease Service Center, Central Point OR)).

As with any intervention strategy and especially for those in the high elevation wildlands that support 5-needle pines, involvement and acceptance by diversity constituents is essential if these threatened ecosystems are to be sustained into the future. An educational website to increase the awareness of the high elevation white pine species, their ecosystems and the threats that face them is now available on-line (Schoettle et al. 2006).

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Structure and Dynamics of Whitebark Pine Forests in the Warner Mountains, Northeastern California

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We examined the structural characteristics and dynamics of whitebark pine (*Pinus albicaulis*) stands along a 640 m elevation gradient in the Warner Mountains of northeastern California. Permanent plots were established along systematically located transects in 1994, and trees within those plots were marked and inventoried.

Whitebark pine forest structure and dynamics vary with elevation. At lower elevations (below approximately 2500 m), stands are characterized by low stem density and basal area, by relatively young, small trees, and by little downed wood and very few snags. Age structure of these stands suggests that whitebark pine began expanding down slope into sagebrush-steppe habitat in the latter half of the 19th century. Higher elevation (above approximately 2500 m) stands generally exhibit structural characteristics that suggest suggesting old, self-perpetuating stands: uneven (approximating “reverse-J” shaped) diameter and age distributions, individual trees of great size and age, occasional snags, and greater amounts of downed wood. Other structural components (density, basal area, tree height and maximum diameter) of these higher stands vary with elevation. Whitebark pine regeneration is occurring at all elevations, but sapling and seedling density are uniformly lower than that of tree density.

Field observations in 1994 suggested that a maximum of 6% of the trees within the study plots were infected with white pine blister rust. Standing, dead stems occurred at a density of 30.1/ha and comprised 2.6% of all stems measured.

In 2006, plots will be revisited and the fate of previously marked trees will be assessed. Preliminary results will be presented regarding tree mortality over the twelve-period and current rates of blister rust infection.

Structural Adaptation in Bird-dispersed Whitebark Pine and Siberian Stone Pine Seeds

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The large and wingless seeds of *Cembrae* pines are dispersed by nutcrackers (Genus *Nucifraga*; Family *Corvidae*), which cache seeds in the soil during late summer and autumn. A high percentage of undeveloped seeds, irregularity of large cone crops, and lack of detailed studies on the anatomy of stone pine seeds hamper effective seedling production. We studied basic structures, maturity, and water imbibition by whitebark pine (*Pinus albicaulis* Engelm.) and Siberian stone pine (*Pinus sibirica* Du Tour) seeds, as well as structural changes during a 90-day multi-step treatment of whitebark pine seeds, using scanning electron (FESEM), transmission electron (TEM) and light microscopy (LM) and histological stainings. The thick and hard seed coat was permeable to water. However, the very small pits of the sclerotesta of the seed coat together with phenol compounds, particularly in both the pigmented sarcotesta and in the multilayered nucellar tissue inside the seed coat, hindered the entrance of stain molecules. These structures apparently constitute an essential protection against drying, if seeds are left uncovered, or against microbes and predators when seeds are cached in the soil. They also partly explain the existence of a *Cembrae* pine soil seed bank, which is a very unusual phenomenon among conifers. Other striking differences compared to many other conifer seeds were clustering of the large, thin-walled megagametophyte cells, and great accumulation of starch in both the megagametophyte and the embryo of the untreated and pre-treated seeds. Protein bodies of the embryo were in early development stages, whereas in the megagametophyte their stages varied. Lipids were in easily soluble form and therefore difficult to observe with LM. Lipid bodies were less abundant and smaller in the embryo than in the megagametophyte. Ultrastructurally, they formed a layer in the periphery of most embryo cells. The megagametophyte cells were packed with lipid bodies of variable form and sizes. Abundance of starch grains and soluble lipid bodies may facilitate removal of reserves needed for the physiological maturation and the structural differentiation of the embryos during the time when seeds are buried in the soil. Microscopy revealed enlargement of the moistened embryo at the beginning of the pre-treatment, and early structural differentiation of the embryo rather than actual growth through cell division and increasing cell numbers until seedling emergence. Although the seed coat was mature, our results indicated that the embryo and the megagametophyte were still immature. The morphological differences and variant chemical maturity of the seed structures may be an advanced

adaptation to bird-dispersal, and well-controlled pre-treatment simulating natural conditions should result in improved germination.

Burn Severity and Whitebark Pine (*Pinus albicaulis*) Regeneration in the North Cascades

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Extensive population declines throughout the range of Whitebark pine due to fire exclusion and an exotic fungus (*Cronartium ribicola*), which produces white pine blister rust, have raised alarm over the fate of this essential subalpine species. This study examined the relationship between post-fire whitebark pine regeneration and burn severity in the North Cascades of Washington. Any increase in whitebark pine seedling success could accelerate the process of natural selection towards blister rust resistance. The relationship between successful whitebark regeneration and burn severity had not previously been established in the North Cascades. Regeneration data was collected eleven years after two 1994 fires, the Boulder Creek Burn in the North Cascades NP and in the Tyee Complex Burn in the Wenatchee NF. This study questioned if there are more whitebark pine seedlings on burned or unburned sites, what level of burn severity produced the most whitebark pine seedlings, and if these relationships are different on moist and dry sites. Burn severity was determined by basal area mortality, char depth in the soil, and char height on the standing trees. This thesis presents an ecological model of the relationships between environmental and biological factors, which can assist in predicting the system's response to fire. The Boulder Creek site showed a strong negative quadratic relationship between basal area mortality and Whitebark pine seedling densities; whereas, the Tyee Complex Fire site showed a strong positive relationship between the Whitebark pine seedling establishment and the distance to the edge of the burn. Understanding the natural regeneration process for whitebark pine and its response to fire is essential for successful implementation of whitebark pine restoration efforts.

The Role of Disease Resistance in the Recovery of Whitebark Pine

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Over the last fifty years, research and development of genetic resistance to *Cronartium ribicola* in North American white pines (subgenus *Strobus*) has gone from what seemed a long-shot in the late fifties and early sixties, to the most widely-accepted option today for protecting host pines from infection and damage by the exotic rust pathogen. Yet, as budgets and personnel shrink in size and number, it is becoming increasingly difficult to muster the long-term will and resources to carry out the R & D tasks required. It is no longer enough to assume that finding and developing resistance to white pine blister rust is a financially- and politically-supportable enterprise, without explicitly documenting the scope of the task and addressing the likelihood of success. Efforts to develop disease-resistance in white pines of commercial value (*Pinus strobus*, *P. monticola*, and *P. lambertiana*), despite allocation of substantial human, financial, and temporal resources, have met with remarkable but incomplete success overall, leading some managers to believe that it is now time to declare victory in the war against white pine blister rust and leave the field of battle. Following a counter-trend, many forest researchers, managers, and advocates, with considerable preliminary success, have turned their attention to whitebark pine (*Pinus albicaulis*), a high-elevation keystone species that is simultaneously threatened by *C. ribicola*, bark beetles, decades of fire suppression, global climate change, and benign neglect. Again, developing resistance to the pathogen is heralded as the best option for whitebark recovery, yet, as a non-commercial species that is appreciated for ecosystem values that are somewhat opaque to politicians and the general public, can we expect research and management efforts to accomplish with whitebark pine what has eluded us so far with the three commercial white pine species? In this talk, I suggest that we consider carefully what we hope to achieve with our resistance efforts, and that we let our past efforts be a guide to how we plan and carry out this monumental task. Just saying, "It has to be done," is not enough. I shall discuss a few of the considerations that must be addressed, some in greater detail than others. Among these are several key questions: Is *C. ribicola* a primary or secondary cause of the observed decline of whitebark pine in Western North America? If it is a primary cause, will deploying resistant whitebark seedlings reverse that decline? Do we have the know-how and technology to deploy whitebark successfully? What are the mechanisms of resistance to blister rust in whitebark pine? Are they sufficiently observable, repeatable, robust, and durable? How are these mechanisms inherited? Is that mode of inheritance sufficient to practically deploy seedlots that comprise an appropriate mix of the desired resistance traits? How long will it take to answer these five questions? How long will it take to develop product (i.e., resistant seedlings), and how will we market it? Do we have the will and stamina to carry out these and other needed research and development efforts over the long-term and with the finances

required? Perhaps most important, do we have the will and foresight to seriously address these and other issues before we leap into the enterprise?

Variation In Resistance to White Pine Blister Rust Among 43 Whitebark Pine Families from Oregon and Washington – Early Results and Implications for Conservation

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Abstract

All nine North American species of white pines are susceptible to the introduced, invasive pathogen *Cronartium ribicola*, the cause of white pine blister rust. Whitebark pine is considered one of the most susceptible species. Genetic resistance is considered a cornerstone for survival to this pathogen. Fortunately, all of the native species of white pines have some level of resistance. Evaluation of resistance in Oregon and Washington families of whitebark pine has only recently begun; currently over 150 seedlots collected from individual parent trees are in resistance testing. This report summarizes differences in responses among 43 seedling families and one bulked seedlot through two years after artificial inoculation with blister rust. Initial infection after inoculation of three-year-old seedlings was very high in the first set of trials; 100% of the seedlings developed needle lesions in the two trials reported in this paper. There were large differences among families in several traits, including percentage of trees with stem symptoms and survival two years after inoculation. The level of resistance present in some families and the frequency of resistance among the 43 families reported here is encouraging. A possible geographic trend in resistance is also noted. It is recommended that at least a subset of families be field planted to validate resistance ratings from this short-term screening. The collection and use of seed from putative resistance parent trees identified through this testing would be a good starting point for restoration efforts.

Introduction

Whitebark pine (*Pinus albicaulis* Engelm.) is one of the nine native species of white pines (also known as five-needle pines) in the U.S. and Canada. All nine species are susceptible to white pine blister rust, a disease caused by the non-native, invasive pathogen *Cronartium ribicola* (J.C. Fisch.). Whitebark pine is considered to be among the most susceptible of the nine North American species of white pines (Hoff and others 1980).

Concerns about the status of whitebark pine in Oregon and Washington began to escalate in the 1990s (Delprato 1999; Snieszko and others 1994). Since then a series of additional field surveys have been undertaken (e.g. Erickson and others, this proceedings; Goheen and others 2002), and an overall summary of these reports has recently been prepared (Ward and others 2006). Blister rust infection and mortality are having heavy impacts on whitebark pine and its associated ecosystems throughout the range of the species, and concerted action is needed

to revitalize these areas (Schwandt 2006). The concerns about the whitebark ecosystems in Oregon and Washington led to the start of a four-year (2005 to 2008) ‘Albicaulis Project’ to further assess the status of this species, to organize additional cone collections, and to develop a conservation plan (see <http://www.fs.fed.us/r6/genetics/programs/whitebark-pine/albicaulis/> for details).

Fortunately, there is at least a low frequency of naturally-occurring genetic resistance in whitebark pine (Hoff and others 2001). Such genetic resistance has been used as a cornerstone for potential restoration and reforestation efforts for other species (Sniezko 2006) and is viewed as an essential element in such efforts with whitebark pine (Samman and others 2003; Schwandt 2006). Cone collections from National Forests, National Parks, and the Confederated Tribes of Warm Springs were started in the mid-1990s to use in rust resistance testing and in studies of genetic variation (For results from early common garden genetic studies see Bower 2006; Hamlin and others, this proceedings.).

The Pacific Northwest Region of the USDA Forest Service (Region 6) has begun testing seedling families of whitebark pine for resistance to white pine blister rust. Over 150 families are now being tested at Dorena Genetic Resource Center (DGRC), and several hundred additional seedlots are scheduled to be sown in early 2007 for testing (many of these latter collections were funded through the ‘Albicaulis’ project). These seedlots represent much of the range of whitebark pine (WBP) in Oregon and Washington (Region 6) (Table 1). Resistance screening of these seedlots will provide baseline data on levels, frequencies and patterns of resistance. Options for conservation and restoration strategies for affected areas will depend upon the level and frequency of resistance and any geographical trends in resistance.

Table 1. Number of Oregon and Washington whitebark pine seedlots sown for operational blister rust screening at Dorena Genetic Resource Center by year.

Organization	# Families Tested by Sow Year ^a					totals
	2001	2002	2004	2005	2007	
Deschutes NF		10		3	47	60
Fremont NF	4	14		7	15	40
Gifford Pinchot NF					14	14
Malheur NF			19		8	27
Mt. Baker-Snoqualmie NF					4	4
Mt. Hood NF		1	19		3	23
Okanogan NF					10	10
Olympic NF					9	9
Umatilla NF		1	27		2	30
Umpqua NF					10	10
Wallowa-Whitman NF					13	13
Wenatchee NF					38	38
Winema NF		4			10	14
Colville NF		13		5	11	29
Crater Lake NP			10		25	35
Mt. Rainier NP			19	9	4	32
Warm Springs CT			7		12	19
Rogue River NF (Mt. Ashland)					1	1
totals	4	43	101	24	236	408

^a Operational trials at DGRC are identified by the year sown; some seedlots have been included in more than one trial. Inoculation of seedlings occurs in September two or three years after sowing.

Inoculation and assessment protocols for whitebark pine have been adapted from those used for over 40 years in testing for resistance in western white pine (*P. monticola* Dougl. ex D. Don.) and sugar pine (*P. lambertiana* Dougl.) in Region 6 (see Kegley and Sniezko 2004 for some background) and refined in a small prototype test with whitebark pine (see Kegley and others 2004). The first operational inoculation of seedling families from 43 candidate trees selected in Region 6 was conducted in September 2004. Some of the early results are reported here; the seedlings will continue to be assessed over the next three years. Additional operational tests will be reported in future years (see Table 1 for other tests underway).

Objectives of this first operational screening were to (1) confirm the transferability of the rust testing protocols utilized for sugar pine and western white pine programs to whitebark pine; (2) produce the first estimates of the frequency and types of resistance in whitebark pine from Region 6 parent trees; (3) examine the influence of two inoculum sources of the pathogen on resistance; (4) compare the relative level and types of resistance of whitebark pine and western white pine; and (5) discuss some of the implications of the findings to whitebark pine conservation. Only the early assessments of these trials were available for this conference – updates will be provided upon final assessment.

Materials and Methods

Plant Material

The first significant inoculation of individual Region 6 whitebark pine (WBP) families occurred in 2004. This trial included a total of 43 WBP families (seedlings from wind-pollinated parent trees located in natural stands) from six different National Forests in Region 6 as well as a bulked WBP seedlot from the Shoshone National Forest (Forest Service Region 2). The bulked Shoshone lot (CDA # 7425) included seed of >50 parent trees southwest of Dubois, WY (~43°30'N ~109°50'W, elevation ~2987 m) (E. Jungck, pers. comm.). There were 2 groups of Shoshone material; one group of seedlings had been grown at the USDA Forest Service nursery in Coeur d'Alene, Idaho, and another group was sown and grown at DGRC with the Region 6 families.

The DGRC-grown seedlings were germinated in March 2002 following the protocols described by Riley and others (this proceedings) and cultured in supercell containers (16.4 cm³) for two growing seasons (see Riley and others, this proceedings). The WBP seedlings were grown in non-replicated family plots for the first two growing seasons and were a composite from several culturing treatments. The seedlings were transplanted into standard wooden frames (0.91 m wide x 1.21 m long x 0.30 m high) at DGRC approximately 14 months before inoculation. Seedling families were randomized across six replications in 10-tree row plots; some families had less than 60 seedlings and were distributed across the replications accordingly. Eight family row plots were included in each frame. The Idaho-grown Shoshone seedlot was represented in four plots in each replication.

In addition to the whitebark pine, eighty seedlots of western white pine were stratified, then sown directly into standard wooden frames at DGRC in March 2003, and cultured for two growing seasons. Both open-pollinated and control-pollinated seedlots were included and represented an array of susceptible and resistant types. As with the whitebark pine, the western white pine (WWP) families were randomized into six replications of up to 10 trees per replication in family row plots (see Kegley and Sniezko 2004 for a description of the basic study design for WWP operational blister rust screening trials). Two WWP families in this trial, one susceptible and one resistant control, were used to compare relative resistance of WBP with WWP.

Inoculation

Two separate trials, each utilizing a different inoculum source, were conducted. Leaves of *Ribes* spp., the alternate host for blister rust, infected with *C. ribicola* at the telial stage were collected for the inoculations. For **Trial 1**, three of the six replications of both WBP and WWP were inoculated concurrently during the week of August 23, 2004 with a source of rust with known virulence to a specific major gene (Cr2) in WWP. Cr2 conditions a hypersensitive reaction (HR) in the needles of WWP (Kinloch and others 2003), and the virulent source of rust (vcr2 source) would be expected to render HR in WWP ineffective (see Kinloch and others 2004 for discussion). In the absence of vcr2, the WWP resistant control is expected to show only 25% of seedlings with stem symptoms. Approximately one week later, three different replications were inoculated (**Trial 2**) with a composite collection of inoculum from several areas outside the geographic areas where vcr2 has been previously documented (Kinloch and others 2004) (AVCr2 source). Trial 1 used *R. bracteosum* Dougl.

ex Hook. collected on the Umpqua National Forest Cottage Grove Ranger District and *R. nigrum* L. and *R. hudsonianum* Richards. var. *petiolare* from the DGRC Ribes Garden. Trial 2 used *R. hudsonianum* var. *petiolare* leaves from eastern Oregon (Wallowa-Whitman, Malheur, and Ochoco NF), central Oregon (Silver Lake), and southern Washington (Gifford Pinchot NF and Ahtunum Creek, which is near existing WBP stands) and *R. bracteosum* from Trout Lake, WA.

Inoculation followed standard DGRC procedure (Danchok and others 2004). The target inoculum density for both species and both inoculations was 3000 basidiospores/cm², which is the standard inoculum density for operational screening of WWP at DGRC. Actual inoculum densities were 2946 and 3482 basidiospores/cm² for Trial 1 and Trial 2, respectively (std err = 73 and 90, for Trial 1 and 2).

Disease Assessment and Analysis

Seedlings were assessed for survival and the presence and number of blister rust symptoms. Number of needle lesions ('spots') was assessed in May 2005 (8.5 months after inoculation). Presence of spots as well as number and type of stem symptoms were assessed in December 2005 (15.5 months after inoculation). Mortality was assessed in early August 2006 (24 months after inoculation). Third-year height (height at time of inoculation) was measured in May 2005. This paper reports percent spotting (% SPOT), number of spots at 8.5 months after inoculation (SPOT1#), percentage seedlings with stem symptoms (% SS2), number of stem symptoms per infected tree (SS2#), percentage infected seedlings surviving (% RSURV2), and percentage seedlings surviving with stem symptoms (% SSAL2).

Only preliminary analyses of the untransformed data have begun; analyses of variance were performed using plot means using SAS Proc GLM, and Pearson product-moment correlations were calculated using family means (SAS Inc. 2006). Trees dead from non-rust causes (notably due to *Phytophthora* spp.) were excluded from the dataset. Family identity information was compromised in one box of eight families in Rep 3, and those observations were also excluded from the analysis.

Results

Whitebark Pine – General Trends

Inoculation was very effective. 100% of the WBP seedlings had needle infections at the May 2005 assessment (8.5 months after inoculation), and the majority (93.9 and 90.0% for the Trial 1 and Trial 2, respectively) still had needle lesions present seven months later, 15 months after inoculation (% SPOT2, Table 2). There were significant differences among families ($F = 3.76$, $p < 0.0001$) and replications ($F = 21.5$, $p < 0.0001$) for number of spots. Number of spots averaged 14.9 and 18.9 for Trials 1 and 2, respectively (SPOT1#, Table 2), while family means ranged from 9.4 to 24.9 over the two trials. In general families tended to perform similarly in both trials (Figure 1, $r = 0.57$, $p < 0.0001$).

Table 2. Population (forest) means for whitebark pine (WBP) and family means for western white pine (WWP) controls after inoculation with two inoculum sources (Trial 1 and Trial 2).

Group	# families	spot1# ^a		% SPOT2 ^b		% SS2 ^b		% RSURV2		% SSAL2	
		Tria 11	Trial 2	Tria 11	Tria 12	Tria 11	Tria 12	Tria 11	Tria 12	Tria 11	Trial 2
All families	48	14.9	18.9	93.9	90.0	87.7	89.8	25.6	24.4	18.3	18.1
Deschutes	10	15.5	18.2	91.1	90.2	92.4	95.2	19.7	12.1	17.5	8.1
Fremont	14	17.1	21.2	91.8	90.1	100.0	97.3	8.8	5.1	8.8	2.7
Mt Hood	1	18.1	18.4	96.7	93.0	45.6	82.0	82.6	81.5	61.9	77.5
Umatilla	1	10.7	15.3	81.7	66.7	100.0	91.7	4.2	0.0	4.2	0.0
Winema	4	13.9	21.1	97.5	96.4	100.0	94.1	6.8	15.8	6.8	11.4
Colville	13	13.1	19.4	97.7	96.6	69.6	76.3	55.3	58.5	35.8	46.0
DGRC-grown Shoshone	1	19.6	16.1	100.0	83.3	85.0	100.0	20.6	10.0	5.6	10.0
CDA-grown Shoshone	4	12.0	11.0	92.8	67.6	87.8	87.8	14.2	15.4	4.5	4.2
WWP susceptible	1	--	--	93.3	93.3	100.0	93.3	100.0	100.0	100.0	100.0
WWP MGR	1	--	--	74.8	53.3	92.6	26.7	96.7	100.0	100.0	100.0

^a 100% of the WBP seedlings and the WWP controls had spots ~8.5 months after inoculation. Actual number of spots was not counted for the WWP; relative number of spots was indicated by assignment into classes (see Kegley and Sniezko 2004 for details of the procedure)

^b Adjusted to exclude Phytophthora mortality

The percentage of seedlings with stem symptoms (% SS2) was similar for the two trials, 87.7% and 89.8% for Trials 1 and 2, respectively. There were significant family and replication differences for % SS2. Family means varied from 39.9 to 100% in Trial 1 and from 23.3 to 100% in Trial 2 (Figure 2). Fifteen of the 43 seedling families showed 100% SS2 in both Trial 1 and 2. Families tended to perform similarly with both inoculum sources ($r = 0.75$, $p < 0.0001$), but a few families showed differences of 30% or more (Figure 2). In general, the Colville families and the single Mt. Hood family had fewer seedlings with stem symptoms relative to the other populations (Table 2, Figure 2). One of the Deschutes families also showed relatively low % SS2. Infection (presence of needle spots or stem symptoms) was 100% in both trials.

Figure 1. Family mean number of spots per seedling (SPOT1#) in Trial 1 (vcr2 inoculum source) vs. Trial 2 (AVCr2 inoculum source) for seedlots from 7 National Forests

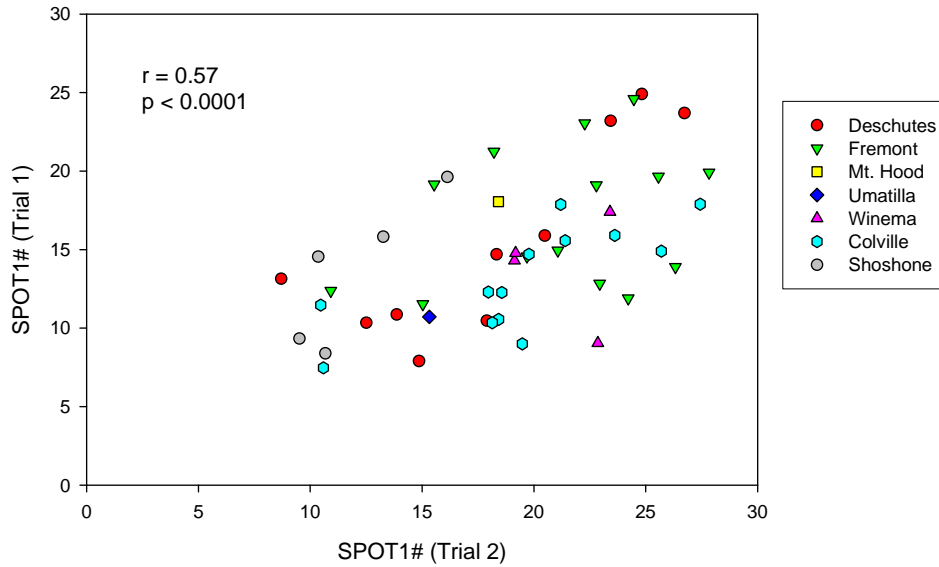
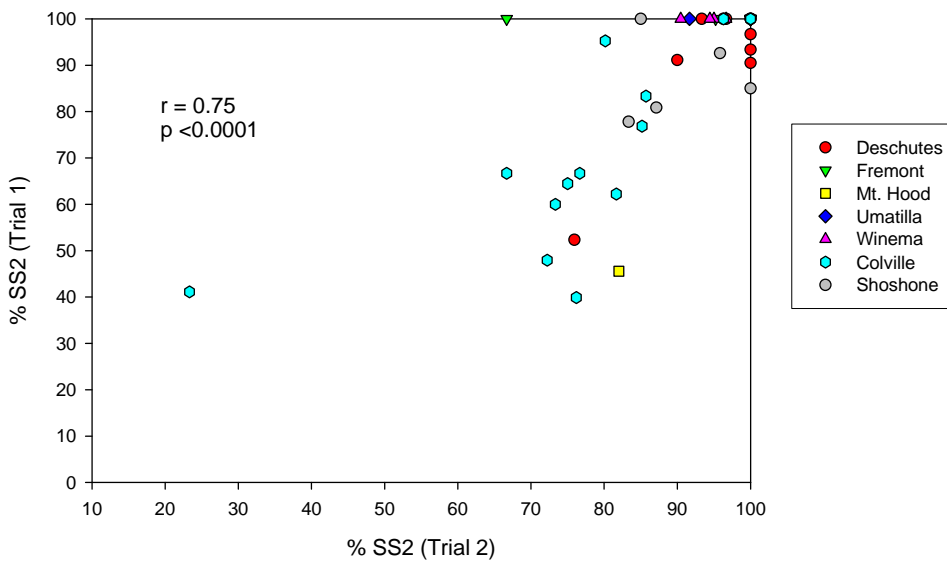
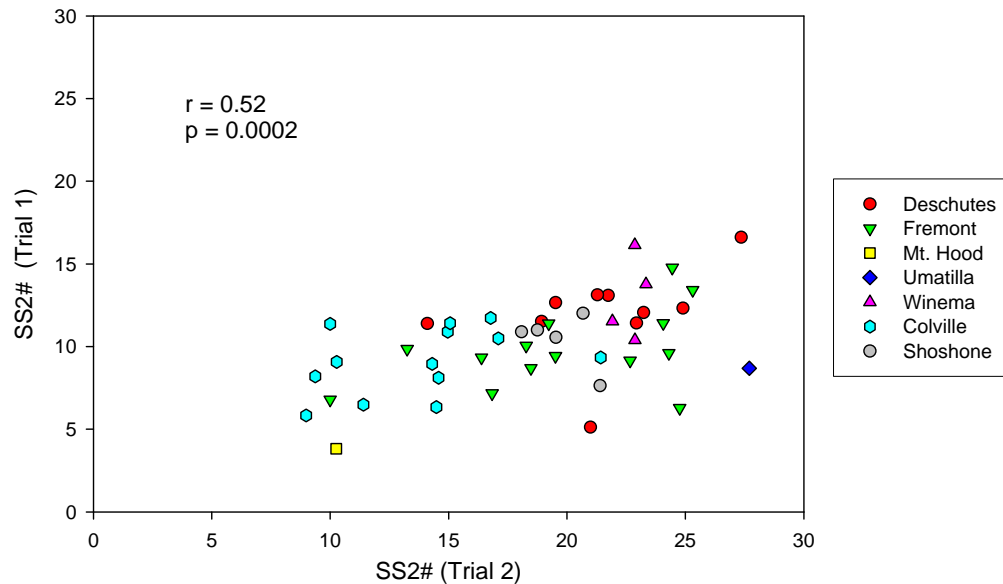


Figure 2. Family mean percentage seedlings with stem symptoms (% SS2) in Trial 1 vs. Trial 2 for seedlots from 7 National Forests



Although overall % SS2 was similar across the two trials, there were significant family, inoculum source, and replication differences in number of stem symptoms per infected seedling (SS2#); mean SS2# was 10.2 for Trial 1 and 18.7 for Trial 2; family mean SS2# ranged from 3.8 to 16.6 for Trial 1 and from 9.0 to 27.7 for Trial 2 (Figure 3). The Mt. Hood family and many of the Colville families had among the lowest SS2#, while the four Winema families had among the highest SS2# in both trials (Figure 3).

Figure 3. Family mean number of stem symptoms (SS2#) in Trial 1 vs. Trial 2 for seedlots from 7 National Forests



Survival of seedlings with stem symptoms two years after inoculation (% SSAL2) averaged around 18% for both trials (Table 2); family means ranged from 0 to nearly 75% for the two trials. Fourteen of the 43 families had at least 15% SSAL2 in both trials, and four of these families had more than 50% SSAL2 in both trials. It was noted that, at least for the first two years after inoculation, the families that had lower % SS2 also tended to have higher % SSAL2 (Figure 4).

Survival of infected seedlings nearly two years after inoculation (% RSURV2) was similar between the two trials, averaging 24.5% for Trial 1 and 22.4% for Trial 2 (Table 2, Figure 5). Dramatic differences in survival were evident among families (Figures 5 and 6a). Family mean survival ranged from 0 to 81.5% for Trial 1 and from 0 to 90% for Trial 2 (Figure 5). Four of the 43 Oregon and Washington families had no survivors in either trial. Two years after inoculation, 13 of the 43 seedling families (30% of families) showed moderate to high levels of survival (>35% seedlings alive), including 11 of the 13 Colville families (Figure 5). The remaining 30 families and the Shoshone bulk lot generally showed much lower levels of survival. Families tended to show similar levels of survival in both trials (Figure 5, $r=0.87$, $p<0.0001$).

Whitebark Pine versus Western White Pine

The WWP susceptible control exhibited 100% and 93.3% SS in Trial 1 and Trial 2, respectively (Table 2). The MGR WWP control family had 92.6% SS in Trial 1 and 26.7% SS in Trial 2 (Table 2). This WWP family was expected and did exhibit a differential response to the two inoculum sources (higher % SS when inoculated with the vcr2 source); the susceptible WWP family and the WBP families did not exhibit this differential response. The susceptible WWP averaged 7.7 and 11.1 SS per infected tree for Trial 1 and Trial 2 respectively. This is slightly lower than the trial averages for all WBP families, despite the WWP being much larger in size. Third-year seedling height of the WBP averaged 9.2 cm in Trial 1 and 8.8 cm in Trial 2; family means ranged from 5.0 to 13.6 cm across the two trials;

whereas the WWP controls averaged between 27.1 and 29.4 cm. Through August 2006, no mortality had occurred in these two WWP seedlots compared with high mortality noted in the whitebark pine (Figure 6b).

Figure 4. Family mean % survival with stem symptoms (% SSAL) vs. percentage seedlings with stem symptoms (% SS) for Trial 1 and Trial 2 for seedlots from 7 National Forests

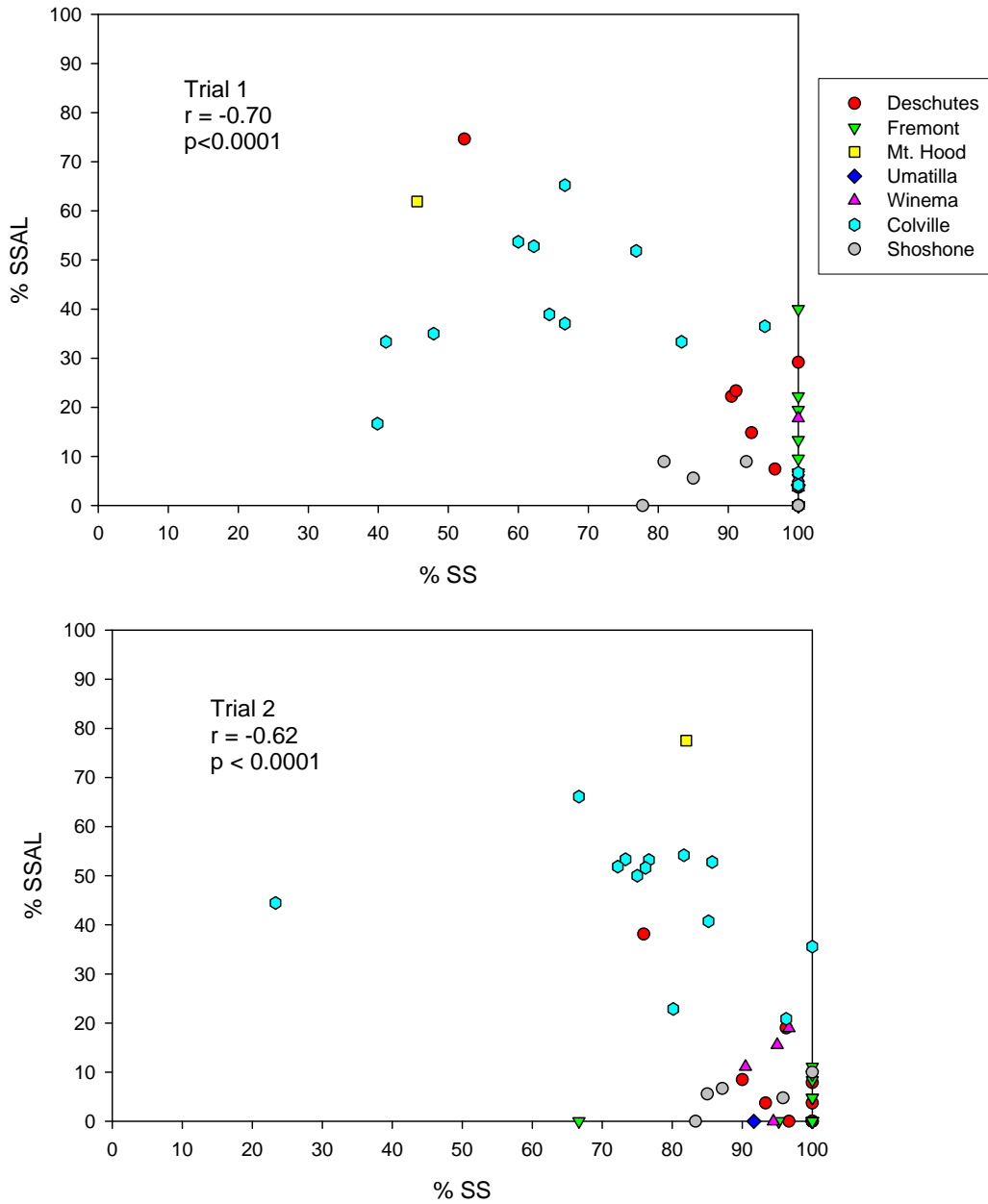


Figure 5. Family mean % survival of infected seedlings (%RSURV2) in Trial 1 vs. Trial 2 for seedlots from 7 National Forests

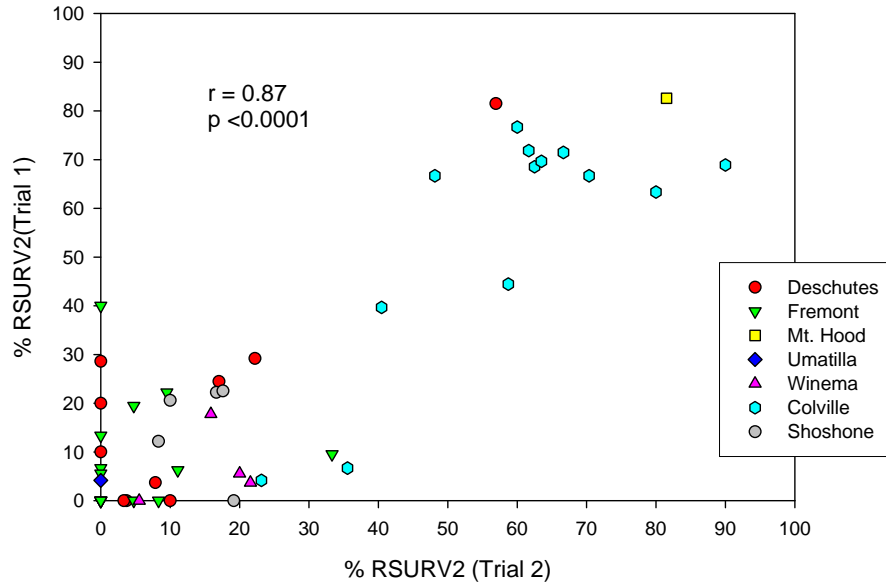
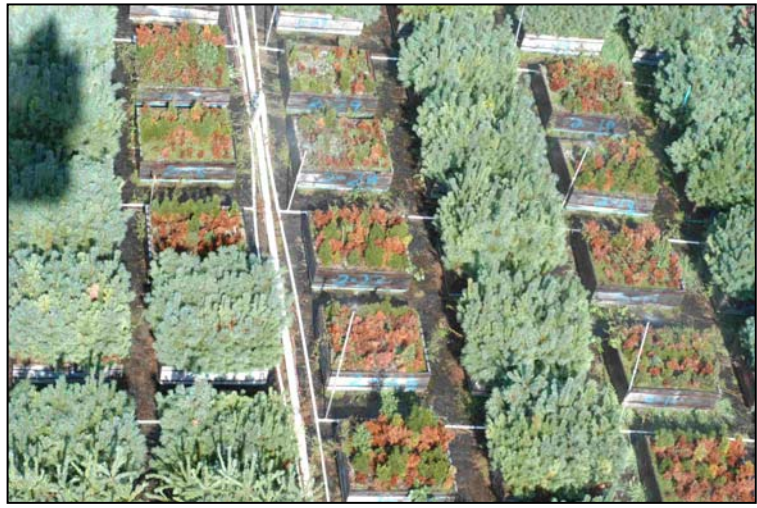


Figure 6. (a—Left) Survival and mortality (red trees) of WBP in May 2006. Note the high survival of one family from the Colville NF. (b—Right) Comparison of WWP (tall green trees) and WBP



Discussion and Summary

It will still be several years before we have final results from these two trials, but at this stage it appears that there is little practical difference between the two inoculum sources used. Similar results were found in a smaller, earlier trial using two different geographic sources of inoculum; the % SS and % survival were similar between the two inoculum sources (Kegley and Snieszko 2004; Kegley and others, this proceedings). If this result holds for the trials presented here, it would imply that easily accessible sources of inoculum could be used to evaluate whitebark pine for resistance. However, the ultimate validation will come from field tests.

The moderate to high frequency of families with low % SS in the 43 field selections tested is higher than that found our rust tests of field selections of WWP or sugar pine (see Kegley and Sniezko 2004). However, the WBP seedlings averaged more SS per tree than the susceptible WWP control, and mortality in WBP proceeded more rapidly than in WWP. The rapid mortality of seedlings with stem symptoms may be a function of several factors, including seedling size or intrinsic susceptibility of the species. From our experience at DGRC, WBP, WWP, sugar pine, and eastern white pine (*P. strobus* L.) are all very susceptible, with sugar pine and eastern white pine showing faster mortality than WWP. However, even through this stage of assessment, the level and frequency of resistance (notably low % SS) found in this first test of Region 6 families was somewhat unexpected.

The Shoshone seedlot was a bulked collection and had been included in an earlier prototype trial to examine the effect of inoculum density and geographic source of inoculum on WBP (Kegley and others 2004; Kegley and Sniezko, this proceedings). As in that earlier trial, the Shoshone seedlot also showed relatively low levels of resistance and low survival (10 and 11.8% for DGRC-grown and Coeur d'Alene-grown seedlings, respectively). In another test, this seedlot ranked 59th for rust resistance among 108 seed sources from the Inland Northwest, using an index of resistance traits (Mahalovich and others 2006).

The level of infection, particularly the number of stem symptoms per seedling, is high in these two trials and should represent a good test of resistance. However, this high level of infection may overwhelm some types of partial resistance in small seedlings, leading to high mortality in some families. In addition some resistant traits (e.g. low # stem symptoms) may not be as effective in very young seedlings as they would be in the field on larger trees. Thus, further examination of traits such as the number of needle spots, number of stem symptoms and time of mortality is needed. Field validation of the effectiveness of these traits is also essential.

The number of resistance mechanisms present in WBP is unknown and may vary by geographic origin. Hoff and others (2001) summarized resistance work in several populations of WBP and found three of the putative resistance mechanisms that have been reported in WWP of Idaho origin: 'Needle shed' and 'Short-shoot', resistances which prevent stem infection, and 'Bark Reaction', a resistance that produces an incompatible stem infection. In a larger study of sources from the Inland Northwest, resistance responses similar to those reported by Hoff were found (Mahalovich and others 2006). In the two DGRC trials reported here, most trees still had spots at the second assessment (15.5 months after inoculation), by which time they were cankered. In the DGRC trials, the few families with lower % SPOT2 had very high levels of stem symptoms and mortality (unpublished data). The underlying mechanism resulting in a no stem symptom phenotype in Oregon and Washington WBP and its inheritance is currently unknown. Through nearly two years after inoculation, the bark reaction response was very infrequent and incomplete (partial) in our trials.

In this study of progeny of Oregon and Washington trees, apparent differences in SPOT1#, SS2#, % SS2, % SSAL2, and % RSURV2 were observed, and further examination of these traits is underway. In general, the families showing the lowest % SS2 were also the families

showing the highest % SSAL2. The parents of these families may have several resistant mechanisms, or seedlings within the families may manifest the resistance slightly differently. These are also the families with the highest survival through August 2006 assessment. These results are still preliminary, especially for the survival traits. Many infected seedlings are heavily cankered but currently alive, SSAL (Figure 7a and 7b), and are expected to die in the next two years.

From the modest number of seedlots sampled here, one of the four geographic areas with multiple families stands out for resistance, the Colville NF. Most of the Colville families had 40 to 80 percent SS and were in the top quartile for survival. In addition, the one seedlot from Mt. Hood NF also was showing high survival at this stage. This early data suggests that in Oregon and Washington, resistance may be in higher frequency in the northern part of the geographic range. This possibility is under further investigation in other trials (Table 1). In a study of Interior Northwest seed sources, principally from Idaho and Montana, an increase in some types of resistance was noted from southeast to northwest (Mahalovich and others 2006). Four families from Colville NF were included in that trial, and two were among the top 10 ranked for resistance and represented the northwestern part of the range sampled in that study.

The higher incidence of rust resistance noted in collections from the northern areas in this study may be due to several factors, including higher incidence of rust in the northern part of the range. At this point in time, information about infection levels from all of the parent tree stands is incomplete. However, recent surveys have noted that northern stands generally have greater levels of rust infection (Shoal and Aubry 2006). The higher incidence of rust in the north would have led to higher rust mortality, thus leaving a higher percentage of trees with rust resistance (see Hoff 1994 for discussion). Hoff (1994) observed higher levels of resistance in WBP families from stands with high mortality due to blister rust and lower levels of resistance in WBP from stands with low to moderate mortality (44% canker-free vs. 4% and 18%, respectively). The Region 6 WBP parent trees represented by seedling families in this test were selected on the basis of cone availability and not necessarily for blister rust resistance. Further work is needed to examine the rust incidence and mortality in areas where seed collections were made.

Recent research suggests that whitebark pine is most closely related to the Eurasian stone pines and is likely the result of migration through the Bering Strait opening more than 1.8 million years ago (Krutovskii and others 1994; A. Liston, pers. comm.). The Eurasian stone pines have shown very high blister rust resistance in screening trials (Hoff and others 1980). It is possible that the retention of at least some ancestral rust resistance genes may help explain the relatively high frequency of resistance in northern populations of WBP. Additional rust screening trials will help clarify the pattern of resistance in WBP.

Short-term resistance screening can be a very valuable tool to efficiently evaluate many parents for resistance. Additional work currently underway with whitebark pine includes examining many additional families for resistance, testing some of the highest surviving families for HR (not yet documented in WBP), examining the effect of inoculum density on resistance, and determining whether even younger seedlings can be used to rate families for

resistance (which could reduce costs and shorten the time period for assessing resistance). However, planting of a subset of seedling families on whitebark pine sites will be essential to confirm that this resistance is also effective under field conditions and to monitor the durability of resistance for this species.

The parent trees of the families rated for resistance in this test can be used as ‘permanent plots’ to follow the continued progression of rust as well as to monitor the durability of field resistance. Consideration should be given to protecting some of these parent trees from mountain pine beetle (*Dendroctonus ponderosae* Hopkins) so that they can serve as both cone producers and as monitors of field resistance. Parent trees of seedling families showing good survival, particularly those showing the lowest levels of SS%, are candidates for seed collection or inclusion in seed orchards (seedlings within these families are also candidates for inclusion in orchards). Families performing well for other traits (for example, families with few stem symptoms) should also be considered. However, additional families will have to be evaluated for resistance to increase the genetic base of any orchards established.

Conservation and restoration possibilities for whitebark pine will depend on a variety of factors, but the frequency and durability of resistance and rust hazard of the site will be two of the most important. Establishing resistant seedlings will be a key step in moderate to high disease areas. Encouraging abundant natural regeneration can also be an important step in maintaining whitebark pine in ecosystems (Schoettle and Sniezko, in press; Schoettle and others, this proceedings). Further development of seed transfer guidelines for whitebark pine within Oregon and Washington would be valuable in helping transfer resistant seed from one area to other areas where there is little or no resistance.

Figure 7. Seedlings surviving with multiple stem symptoms (January 2007)



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Genetic Diversity and Geographic Differentiation in Quantitative Traits, and Seed Transfer Guidelines for Whitebark Pine

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Whitebark pine (*Pinus albicaulis* Englem.) has declined dramatically throughout its range due to white pine blister rust (caused by the fungus *Cronartium ribicola* J.C. Fisch.), successional replacement resulting from fire suppression, and attack by mountain pine beetle (*Dedroctonus ponderosae* Hopkins). Restoration is needed to halt or reverse this decline; however, no information regarding genetic diversity and local adaptation of quantitative traits is available to guide these efforts.

A seedling common garden experiment was employed to assess genetic diversity and geographic differentiation of quantitative traits (Q_{ST}) of whitebark pine and to determine the climatic variables driving local adaptation. Seedlings from 48 provenances from a near range-wide seed collection were grown in raised beds in Vancouver, B.C. for two years in two soil temperature treatments (ambient and cold). Seedlings were measured for second year height increment, total biomass, root:shoot ratio, date of needle flush, fall and spring cold injury, and survival.

Significant differences were found between soil temperature treatments for height growth and survival, with seedlings in the cold treatment performing better. The environment where the test was grown (Vancouver, B.C.) is considerably warmer than the natural habitat of whitebark pine. While most temperate forest trees would likely benefit from the warmer soil temperatures, it appears that this is a stressor for a species adapted to cold, harsh environments.

We observed significant differences among provenance means in most quantitative traits (Table 1), similar to many other widespread North American conifers. Differences among provenances accounted for a substantial proportion of the variance only for cold acclimation traits (date of needle flush and fall cold injury), indicating that provenances are under stronger differential selection for these traits than for growth. In the common garden environment, provenances from higher latitudes and lower winter temperatures flushed earlier in the spring, suffered less cold injury in the fall, and allocated more biomass to shoots. In the subalpine environments where whitebark pine grows, these traits most likely have a larger role than growth traits in determining local fitness and the ability to withstand abiotic stresses associated with local climate.

Table 1. Significance level of provenance effect in ANOVA, proportion of variation accounted for by provenance and family, and genetic differentiation (Q_{ST}) for nine quantitative traits in ambient (A) soil temperature treatment and seven traits in cold (C) treatment.

Variable	Provenance		σ^2_P/σ^2_T	σ^2_F/σ^2_T	Q_{ST}
	F-value	p-value			
A-Height increment^{ab}	1.84	0.01	0.05	0.05	0.14
A-Total dry mass^{ab}	1.69	0.01	0.04	0.09	0.07
A-Root dry mass^{ab}	1.65	0.02	0.03	0.07	0.08
A-Shoot dry mass^{ab}	1.73	0.01	0.04	0.10	0.07
A-Root:shoot^{ab}	1.09	0.35	0.00	0.03	0.00
A-Date of needle flush 2003^a	6.51	<0.001	0.26	0.05	0.47
A-Date of needle flush 2004^a	5.49	<0.001	0.23	0.04	0.47
A-Fall cold injury	2.59	<0.001	0.19	0.06	0.36
A-Spring cold injury	1.11	0.33	0.05	0.07	0.12
C-Height increment^b	1.57	0.04	0.07	0.07	0.13
C-Total dry mass^b	1.24	0.20	0.06	0.11	0.09
C-Root dry mass^b	1.22	0.22	0.05	0.10	0.08
C-Shoot dry mass^b	1.23	0.21	0.06	0.11	0.08
C-Root:shoot^b	1.09	0.36	0.01	0.08	0.01
C-Date of needle flush 2003	3.29	<0.001	0.21	0.05	0.43
C-Date of needle flush 2004	6.36	<0.001	0.33	0.03	0.65

^a A=ambient soil temperature treatment, C=cold soil temperature treatment

^b Natural log transformed

Genetic differentiation (Q_{ST}) was moderate for growth traits (height increment and biomass) and strong for cold adaptation traits (date of needle flush and fall cold injury). For all traits Q_{ST} was greater than previously published estimates for whitebark pine based on range wide studies using molecular markers (F_{ST}) (0.034-0.046), indicating natural selection driving local adaptation.

Canonical correlation analysis was used to examine the relationship of quantitative traits with climatic and geographic variables, and to determine whether variation in quantitative traits among provenances is clinal. The results of this analysis were then used to develop predictive equations for the construction of seed transfer guidelines. Values of significant canonical variables associated with the quantitative traits were regressed on the standardized key climatic variable with the highest loading for that canonical variable. The slope of this regression provides a rate of change in the canonical variable associated with the quantitative traits relative to the climatic variable. Rates of differentiation along climatic gradients were interpreted relative to the least significant difference among provenances at the 20% level (LSD 0.2). This reduces Type II error - accepting no differences among provenances when differences actually exist. The rate of differentiation of the key quantitative traits associated with canonical vectors was determined as the change in the standardized climate variable associated with the LSD value of the canonical variable. The difference in the climate

variable associated with significant genetic differentiation between provenances was calculated as the rate of differentiation multiplied by the standard deviation of the climate variable.

The first pair of canonical variables demonstrates the effects of mean temperature of the coldest month. The positive correlations of date of needle flush and fall cold injury with the first canonical variable (Figure 1) indicates that trees from provenances with higher mean temperature of the coldest month flush later in the spring and suffer higher cold injury in the fall. The second pair of variables demonstrates the effect of the length of the growing season (FFP), with trees from provenances with longer growing seasons growing taller, and producing more biomass.

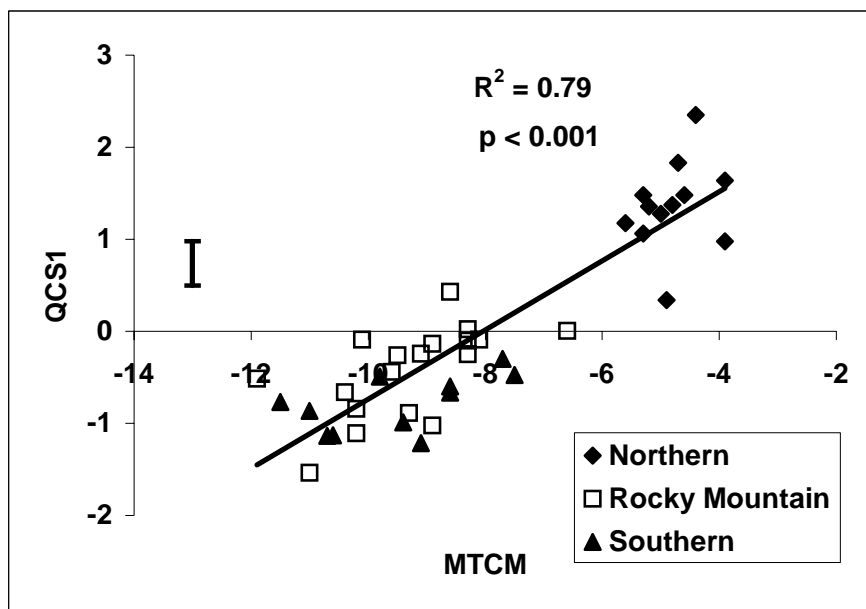


Figure 1. Regression of first quantitative canonical score (QCS1) on standardized mean temperature of the coldest month (MTCM) for 41 whitebark pine provenances in three geographic regions. Axes scales are standard deviations and bracket indicates value of LSD 0.20.

Cold adaptation traits were strongly influenced by mean temperature of the coldest month, while growth traits were influenced by the length of the growing season. Guidelines for seed transfer in restoration programs based on these results indicate that whitebark pine seed can be moved within areas differing by 1.16° in mean temperature of the coldest month or approximately 3° in latitude with minimal risk of maladaptation. The difference in elevation required to distinguish genetically different populations (~700m) exceeds the elevational range of whitebark pine within 3° of latitude, so there should be no elevational restrictions on seed movement.

The role of climate change in the future of whitebark pine is uncertain, and restricting movement of seed to the south may provide a buffer against future climate warming. Therefore, movement of whitebark pine seed should be restricted to 3° to the north and less than 1° to the south in order to minimize maladaptation in current and future environments. The predicted increases in temperature will push whitebark pine beyond the geographic

limits to which is locally adapted and will likely result in a dramatic reduction in suitable habitat, potentially decreasing genetic diversity. Damage and mortality due to blister rust is a primary concern for whitebark pine; however, the potential affects of climate change should not be underestimated. Without restoration and conservation efforts using appropriate seed sources that will be adapted to new climatic conditions, the decline of whitebark pine is likely to be accelerated, with the potential of extirpation in some areas. The data and results presented here are crucial for restoration efforts that will be necessary to maintain whitebark pine as more than a minor component of the ecosystems in which it plays such a vital role.

Genetic Mapping of Genes Controlling Partial Resistance and Major Gene Resistance in Sugar Pine

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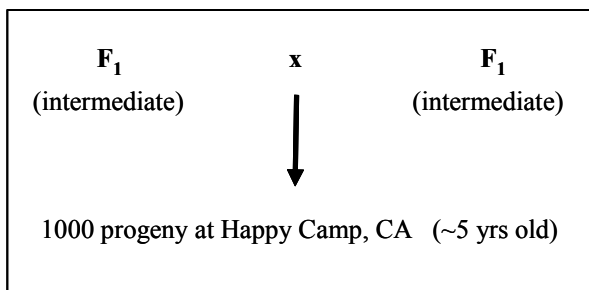
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Pines belonging to the *Pinus* subgenus *Strobus* are susceptible to a fungal pathogen (*Cronartium ribicola*) that was introduced to Northern America in the early 1900s. Surprisingly, several white pines have been shown to possess innate resistance to the rust infection. Two forms of resistance have been observed: 1) monogenic (or qualitative) resistance, segregating as a single dominant gene (major gene resistance – MGR) and 2) polygenic (or quantitative) resistance, showing a wide distribution in disease phenotypes (partial resistance). Breeding programs based on deployment of the major gene of resistance are currently in place for sugar pine (*Pinus lambertiana*), however, resistance controlled by a suite of genes may provide resistance that is more durable over time. The number of genetic factors controlling a polygenic trait influences the rate at which genetic gain can be obtained through artificial selection. Hence, knowledge about the number of genes involved in partial resistance is important for any breeding strategy. Genetic mapping of polygenic traits (spring and fall cold-hardiness, dormancy, dormancy-break, wood density, and fiber angle) have been successful in several species from the Pinaceae, providing information about not only the number of genes controlling a trait, but also the location within the genome of such genes, the size of effect that each gene has on a phenotype and whether its mode of action is additive or dominant (Brown et al. 2003; Chagne et al. 2003; Devey et al. 2004; Jermstad et al. 2001a, 2001b, 2003; Neale et al. 2002; Wheeler et al. 2005)

Using sugar pine as a representative species, we are employing several strategies to develop genomic resources for the five-needled pines. The quantitative trait loci (QTL) involved in partial resistance will be mapped in a large full-sib sugar pine family ($n > 1000$) that is segregating for this type of resistance (**Fig.1**). Currently, only the genomic region containing the

Figure 1. Full-sib sugar pine family designed for mapping QTL for partial resistance to *C. ribicola*.



MGR has been mapped in sugar pine (Harkins et al. 1998), thus the construction of a full genome map is much needed. Single nucleotide polymorphisms (SNPs) identified in 1200 gene sequences (see project description at <https://www.fastlane.nsf.gov/servlet/showaward?award=0638502>) will provide the marker segregation data required for genetic mapping and will also be a useful resource for nucleotide diversity studies and comparative genomics among conifers.

The genome organization among pines and other related conifers is conserved according to the results of comparative mapping studies (Krutovsky et al. 2004; Neale and Krutovsky 2004). Therefore, it is expected that the genomic architecture and gene sequences of the white pines will be very similar and, therefore, map information and DNA sequences will be transferable among them. In addition to mapping QTL for partial resistance, we are also pursuing a positional cloning strategy to isolate the DNA sequence (allele) conferring MGR. Two RAPD markers flanking the MGR in sugar pine (**Fig. 2**) have been successfully converted to Sequence Characterized Amplified Region (SCAR) markers. Analyses of these SCAR markers in

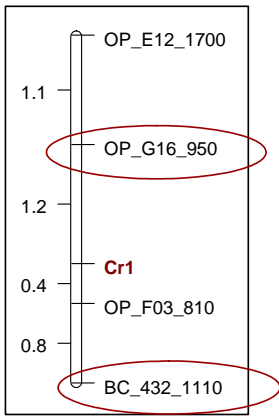
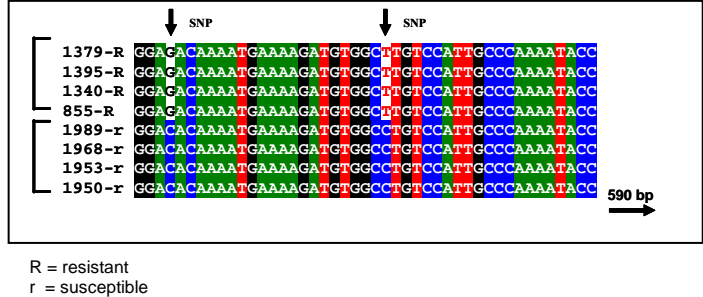


Figure 2. RAPD markers flanking the MGR (*Cr1*) in sugar pine tree 5701 (Harkins et al. 1998). Markers OP_G16_950 and BC_432_1110 are each 1.2 cM (map units) from *Cr1*.

megagametophytes from multiple sugar pine trees revealed SNPs segregating with the disease phenotype (**Fig. 3**), however, each tree examined had a different set of segregating SNPs.

Figure 3. DNA sequences of SCAR marker OPG16₉₅₀ in megagametophytes from tree 5701.



R = resistant
r = susceptible

Hence, these markers would have utility for identification of resistant progeny within specific families but would not be applicable for assays in natural populations.

In an effort to isolate the DNA sequence of *Cr1*, SCAR markers will be utilized as probes for screening a loblolly pine BAC library available (<http://www.pine.msstate.edu/bac.htm>). Isolation of the DNA sequence of the allele conferring MGR in sugar pine (*Cr1*) will provide a precise diagnostic tool for identifying resistant trees in wild populations. Additionally, the isolation of MGR will provide an avenue by which molecular and biochemical interactions between the host and pathogen can be explored.

Because of observed similarity between the genomes of various pines, we expect discoveries pertaining to *C. ribicola* in sugar pine to have direct application in related five-needle pines.

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White Pine Blister Rust and Whitebark Pine Ecosystems in California

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Whitebark pine is a key component of California's subalpine forest ecosystems, contributing to species diversity, trophic interactions, wildlife habitat, and hydrologic functions. While the distribution and effects of white pine blister rust (*Cronartium ribicola*) have been well-documented on sugar pine, less is known about the effects of this introduced pathogen on high-elevation white pines throughout California. In 2004 and 2005 the incidence and impact of white pine blister rust (WPBR) on whitebark pine (*Pinus albicaulis*) was assessed in long-term monitoring plots that were established over the species geographic range in California. Plot data included landscape/stand level data as well as individual tree data. The statewide average for rust on whitebark pine was 12%, ranging from 0 to 71%, with the highest levels in the north-central Sierra Nevada (mean = 25%). Cumulative mortality of whitebark pine was low (mean = 1.1%) in the plots, although causes of mortality in these subalpine woodlands may largely be due to drought, mountain pine beetle, and white pine blister rust.

While we do not see the widespread mortality due to WPBR as in other locations throughout the west, WPBR is affecting the demography of white pine populations. Demographic effects include juvenile mortality, lowered recruitment, and reproductive output. These effects can have negative population and genetic consequences, which will influence how whitebark pine populations respond to other stressors, such as climatic warming. Average regeneration of whitebark pine in uninfected plots was 8.8 seedlings (per 0.15 ha) and 6.3 seedlings in plots where WPBR was present. Reproductive output was higher in uninfected whitebark pine populations (40% of trees bearing cones) compared to WPBR-infected populations (34%). More detailed demographic data as well as quantitative measures of cone production are needed to determine the strength of these patterns.

Blister rust infections were not observed on the alternate host *Ribes* in subalpine forests during the study period; annual infections on *Ribes*, however, are often observed in lower montane mixed-conifer forests. A very weak correlation exists between WPBR incidence and percent cover of *Ribes* in subalpine woodlands ($r = 0.16$) while a much stronger relationship is found between WPBR and *Ribes* cover in lower elevation mixed-conifer forests ($r = 0.55$), where sugar pine is a component. In this survey, preliminary regression analyses of biotic and abiotic factors gave a strong negative relationship with distance to nearest montane mixed-conifer forests, which may be the source of infective propagules to subalpine woodlands. May relative humidity (positive relationship) was also a good predictor of WPBR incidence, i.e., having a moist spring for infections to occur on *Ribes*, as well as mean September minimum temperature with relatively warmer fall temperatures being a

requirement for infections to occur in subalpine forests. Further epidemiological analyses will continue and relate disease incidence to biological and climatological data (PRISM), in order to better understand the epidemiology and disease dynamics of WPBR in whitebark pine forests throughout California.

Interactions among stressors and threats must also be considered e.g., the interaction of climate on WPBR together with mountain pine beetle (MPB) activity, and other anthropogenic influences. Evidence of MPB activity was found in 75% of the whitebark pine plots established, with an average incidence of 7 % (range 0-32%). In high-elevation forests we see evidence of MPB-caused mortality, but at low levels and often associated with protracted drought periods. Once again, these are not at levels of activity seen in other regions of western North America (although there are a few exceptions). So is it a numbers game, in which our region lacks the extensive lodgepole pine forests for large outbreaks and beetle populations to spillover into high- elevation forests or, given the California climatic regime, are whitebark pines in this region relatively more drought tolerant, therefore having a higher threshold before being susceptible to beetle attack? We know little about mountain pine beetle biology and behavior in high elevation forests of California. Clearly, more work is needed in the area of genetics, demography, epidemiology, and MPB ecology to better understand the dynamics of WPBR and other stressors on whitebark pine and how this species will respond to changing environmental conditions in California. Such information would be a valuable asset in developing conservation strategies for maintaining and preserving whitebark pine in the high-elevation subalpine ecosystems.

Verbenone Suppression of Mountain Pine Beetle in Lodgepole Pine at the Sawtooth National Recreation Area in Central Idaho

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The antiaggregation pheromone verbenone was operationally tested for five years to deter mass attack by the mountain pine beetle (MPB) on lodgepole pine in campgrounds and administrative areas surrounding Redfish and Little Redfish Lakes at the Sawtooth National Recreation Area in central Idaho. Each year, five-gram verbenone pouches were placed in an even distribution (approximately 10m apart) within seven of fourteen 0.2 ha plots. During the first two years of the study a median of 12% of the host trees >13cm dbh were attacked and killed on the treated plots, whereas trees on the untreated plots incurred a median mortality of 59%. When approximately 50% of the trees on the untreated plots were killed a detectable beetle response to verbenone on the treated plots dramatically declined. After five years, MPB had killed a median of 87% of the lodgepole pine trees >13 cm in untreated plots and 67% in plots containing verbenone pouches. Beetle pressure was higher on untreated plots in 2000 and 2001, nearly equal between treatments in 2002, higher on verbenone treated plots in 2003 and similar between treatments in 2004. It is hypothesized that the lack of response to verbenone after two years may be related to both population size and spatial scale, i.e. large numbers of vigorous beetles in a local area with a reduced number of preferred large-diameter trees become crowded and stressed, causing a decline in the response to verbenone. The two-year delay in widespread pine mortality caused by verbenone would have given land managers time to use other management tactics to deter catastrophic loss of trees caused by MPB.

Whitebark pine (*Pinus albicaulis*) does not grow in contiguous forest across the landscape like lodgepole pine. Rather, it occurs at high elevations in dispersed small stands or as open-growing individuals. This lack of stand connectivity could help verbenone may be more effective in deterring mountain pine beetle attack in whitebark pine. An active management regime may be the best approach to protect whitebark from attack by mountain pine beetle. The ability of verbenone to divert mountain pine beetles seeking a suitable host may be most effective in isolated stands and where the beetles have an alternate source of attraction. Individual tree protection with verbenone may be of merit for small groups of trees or isolated individuals. In these areas, the application of verbenone in combination with a beetle sink (trap trees or baited traps) and removal of currently infested trees, would most likely show the most promise.

A Comparison of Cone Collecting Techniques for Whitebark Pine

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Abstract

This study's objective was to test and compare the utility of the most popular and emerging methods for collecting cones from whitebark pine: climbing, orchard ladder, ground-based, and tree-tong. A total of 101 cages were installed and retrieved at Crater Lake National Park during 2005. Strengths and weaknesses of each technique are summarized. Brief recommendations are offered.

Introduction

With blister rust well-established in most of whitebark pine's (*Pinus albicaulis*) natural range, tempering the tree's decline requires a long-term commitment. A variety of management techniques are being applied to slow mortality including pruning infected branches, removing fungus-bearing shrubs, and applying fungicides. In order to reverse declines, restoration protocols rely on finding rare naturally occurring disease-resistant trees (Mahalovich and Dickerson 2004). By using these trees as seed sources, breeding programs can regenerate large numbers of trees which can survive the blister rust plague. This protocol essentially accelerates the process of natural selection – thus mitigating the impending bottleneck of whitebark pine numbers. This approach has been successfully applied to western white pine (*P. monticola*) and may be 'the last hope' for whitebark pine (McDonald and Hoff 2001).

The first critical step for developing disease resistance hinges on the ability to identify candidate trees and collect cones. Whitebark pine occurs at some of the most rugged and remote locations in western North America. Most populations are miles from the nearest road, exposed to harsh climate, and upon treacherous terrain. Long rescue times present additional risk. A further challenge is preventing the harvest of cones by marauding birds. These impediments demand careful planning which incorporates the safest and most efficient techniques for collecting cones.

There is very little information available addressing the challenges of whitebark pine cone collection. This study's objective was to test and compare the utility of four popular methods for collecting cones from whitebark pine along with a new technique, a tree-tong, to guide fieldworkers in safe and effective practices.

Methods

During the summer of 2005, twenty-nine whitebark pine trees were chosen for disease-resistance testing at Crater Lake National Park, Oregon. Collecting options were: climbing with rope, free climbing, ground-based, orchard ladder, and tree-tong (fig. 1). Climbing with

rope involved the prusik system with a climbing harness (Davis 2005). Free climbing involved the same safety gear, harness, and lanyard except no rope was used to assist the climber. Ground-based collection was simply performed where cones were within reach of a person standing on the ground. The orchard ladder we used was a commonly available aluminum tripod model (14-ft tall). The tree-tong was developed specifically at Crater Lake National Park for collecting whitebark pine cones in 2005. It consists of an 18-ft long telescoping aluminum pole with a pre-fabricated tong screwed on the end which is opened and closed from a dangling rope. A single two-person crew did all collecting.



Fig 1—Collecting techniques tested include, from left to right, tree-tong, orchard ladder, climbing with rope, and free climbing. (Photos by Michael Murray, except Molly Allen (left.))

Because Clark’s nutcracker (*Nucifraga columbiana*) and rodents are ubiquitous collectors of whitebark pine seeds, we enclosed cones in protective mesh cages in early summer. During return visits in September-October we retrieved each cage and the enclosed ripened cones. Usually, the same collection method was used on the second visit.

Results and Discussion

A total of 101 cages were installed and retrieved. I found strengths and weaknesses of each collection technique (Table 1). Climbing was a relatively slow process and caused damage to the thin tree bark despite our best efforts to the contrary. The tree-tong was the workers’

favorite for its portability and quickness. However, we were unable to adequately close the cage bottom to firmly grasp the branches. As a result, nearly half of these cages blew off before cones were ready for retrieval. In 2006, modifications to the system resulted in improved cage closure ability (Davies and Murray, in press).

Table 1—Comparative summary of cone collection techniques used at Crater Lake National Park, summer 2005.

Technique	Strengths	Weaknesses
Climbing with Rope	<ul style="list-style-type: none"> • Tallest trees can be accessed 	<ul style="list-style-type: none"> • Technical training required • Hazardous • Slow • Costly Equipment • Tree damage unavoidable
Free Climbing	<ul style="list-style-type: none"> • Tallest trees can be accessed • Moderately fast 	<ul style="list-style-type: none"> • Technical training required • Hazardous • Moderately Costly Equipment • Tree damage unavoidable
Ground-based	<ul style="list-style-type: none"> • Fastest • Least Expensive • Safest 	<ul style="list-style-type: none"> • Restricted to cones reachable from ground (can be rare)
Orchard Ladder	<ul style="list-style-type: none"> • Fast 	<ul style="list-style-type: none"> • Ladder can be cumbersome to carry
Tree-tong	<ul style="list-style-type: none"> • Very Portable • Inexpensive • Safe 	<ul style="list-style-type: none"> • Less effective in closing bottom of cage around branch^a

^aImprovements in cage closing ability have been made since 2005 (Davies and Murray, in press).

The most appropriate time to choose a collecting technique is during the initial tree survey. This decision should factor cone height in the tree canopy, the availability of branches for climbing, the distance from a road to the tree, and the exposure of the canopy to strong winds (Murray, in press). Once a technique is selected, corresponding field gear can be prepared for a return trip to install cages. In remote locations requiring long travel, the initial survey can be combined with cage installation, necessitating the transport of all cages and installation equipment. In these instances, a tree-tong plus climbing gear with ropes is recommended.

Acknowledgement

Molly Allen, Biological Technician, Crater Lake National Park, provided significant fieldwork assistance.

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Whitebark Pine Seed Storage and Germination: A Follow-Up Look at Seedlots from Oregon and Washington

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Abstract

During the spring of 2006, 21 seedlots (families), collected from 1996 through 2005 from across the Pacific Northwest range of whitebark pine, were used in 3 trials with the following objectives: to evaluate effects of storage time on germination success and speed of germination; to evaluate differences in embryo length relative to cavity size on germination success and speed of germination; to evaluate effects of x-rays on germination success and speed of germination; and to compare results of tetrazolium (TZ) testing with x-ray results and actual germination in whitebark pine seeds. Two key findings were: 1) following mechanical sanding, seedlots stored as long as 10 years germinated almost as well as seedlots stored for shorter periods of time; and 2) properly scored X-rays can be a good predictor of germination success.

Introduction

Information on seed viability and seed storage periods for whitebark pine has been variable and limited. Whitebark pine seeds contain higher proportions of lipids than most other conifer seeds (Lanner and Gilbert 1994; Lanner 1998), which could affect viability following periods of freezer storage. Some research has shown that seed viability is significantly reduced following storage of periods greater than 2 to 3 years (Burr and others 2001), with earlier studies showing losses down to 1 to 3% total germination after 11 years (McCaughey and Schmidt 1990). However, work in Oregon found that at least some older Oregon and Washington seeds can be stored without significant damage for longer periods of time (Riley and others, this proceedings).

In recent years, there has been a surge in whitebark pine seed collections to be sown and cultured at various seedling nurseries and testing facilities. Proper collection of whitebark pine cones requires caging of the immature cones in the spring to protect from nutcracker and squirrel damage, then actual collection in the fall. Depending on the location and size of the trees, both of these activities may require climbing of individual trees. In addition, whitebark cones may require special extraction procedures once they are transported to a seed extractory. The result can be extremely expensive and valuable seeds, requiring high seed use efficiency in a nursery.

Several factors enter into seed use efficiency and sowing calculations, with seed viability and germination potential as the most important. In order to determine the best use of seeds available for sowing and eventual white pine blister rust (*Cronartium ribicola*) resistance

testing at USDA Forest Service Dorena Genetic Resource Center (DGRC), a study was implemented to:

- 1) evaluate the effects of storage time on germination success and speed of germination;
- 2) evaluate differences in embryo length relative to cavity size, as a subjective estimate of maturity, on germination success and speed of germination;
- 3) evaluate effects of x-ray analysis on germination success and speed of germination to determine if whitebark pine seeds are damaged during the X-ray process; and
- 4) compare results of tetrazolium (TZ) testing with x-ray results and actual germination.

This study was done to follow-up similar small trials of seedlots from Oregon and Washington at DGRC (Riley and others, this proceedings).

Methods And Materials

A total of 21 seedlots, collected from 1996 through 2005 from across the Pacific Northwest range of whitebark pine, were used in trial 1, with a subset of seedlots used in trials 2 and 3 (Table 1). With the exception of those seedlots collected in 2005, all seeds had been stored at DGRC under standard seed-freezer storage conditions. Seeds were air-dried to approximately 7% moisture content, placed in triple-layer foil seed packages, and stored in a seed freezer at -15 to -17 °C.

Table 1. Seedlots used in whitebark pine Trials 1 through 3.

Seedlot #	Coll Year	Origin	% Filled	Prev Germ	# Seeds	Trial 1	Trial 2	Trial 3
1	2005	Crater Lake NP	34		100	X		
2	2005	Crater Lake NP	43		100	X		
3	2005	Giff Pin NF	55		300	X	X	X
4	2005	Okan/Wen NF	32		100	X		
5	2005	Okan/Wen NF	61		300	X	X	X
6	2005	Okan/Wen NF	29		100	X		
7	2005	Deschutes NF	47		100	X		
8	2005	Deschutes NF	51		100	X		
9	2003	Mt Hood NF	100	72	300	X	X	X
10	2003	Mt Hood NF	98	86	100	X		
11	2003	Umatilla NF	96	83	300	X	X	X
12	2003	Umatilla NF	100	85	300	X	X	X
13	2003	Umatilla NF	96	73	100	X		
14	2001	Deschutes NF	90	44/38/53	100	X		
15	2001	Deschutes NF	96	75/64/58	300	X	X	X
16	1999	Fremont NF	34	53/86	100	X		
17	1999	Fremont NF	21	21	300	X	X	X
18	1997	Colville NF	60	25	100	X		
19	1997	Colville NF	56	34	100	X		
20	1996	Colville NF	73	22	100	X		
21	1996	Colville NF	83	46	300	X	X	X

Trial 1—Storage Time and Ratio of Embryo to Cavity Size

A total of 100 randomly selected seeds from each of the 21 single-tree collections were placed on a template and X-rayed. Seeds were then divided into 4 categories as follows:

- a) embryo fills >75% of cavity (Figure 1A);
- b) embryo fills 50% to 75% of cavity (Figure 1B);
- c) embryo fills <50% of cavity (Figure 1C);
- d) embryo cavity empty (unfilled seed) (Figure 1D).

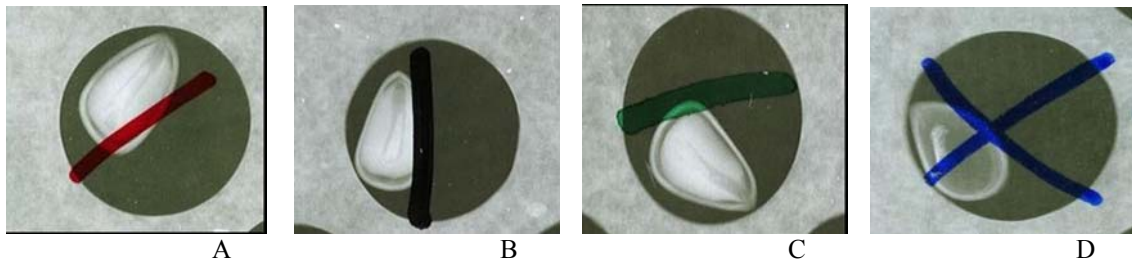


Figure 1. Embryo fills >75% of seed cavity (A); embryo fills 50% to 75% of seed cavity (B); embryo fills <50% of seed cavity (C); unfilled seed (D).

Seeds in each category varied widely by seedlot (Table 2). Seeds in each of categories “A” through “C” were divided into 2 reps for testing.

Table 2. Number of seeds in each category in each seedlot (total 100 seeds/lot).

Seedlot #	Coll Year	Origin	>75% embryo-fill	50% to 75% embryo-fill	<50% embryo-fill	Unfilled embryo
1	2005	Crater Lake NP	15	32	44	9
2	2005	Crater Lake NP	30	28	35	7
3	2005	Giff Pin NF	35	36	20	9
4	2005	Okan/Wen NF	31	31	31	7
5	2005	Okan/Wen NF	24	43	30	3
6	2005	Okan/Wen NF	23	39	31	7
7	2005	Deschutes NF	20	36	40	4
8	2005	Deschutes NF	36	42	19	3
9	2003	Mt Hood NF	29	39	32	0
10	2003	Mt Hood NF	32	49	17	2
11	2003	Umatilla NF	34	24	36	6
12	2003	Umatilla NF	60	26	13	1
13	2003	Umatilla NF	37	22	38	4
14	2001	Deschutes NF	28	36	18	18
15	2001	Deschutes NF	25	39	29	7
16	1999	Fremont NF	0	9	80	11
17	1999	Fremont NF	2	14	68	16
18	1997	Colville NF	32	21	10	37
19	1997	Colville NF	40	18	17	25
20	1996	Colville NF	0	12	53	35
21	1996	Colville NF	11	18	32	39

Trial 2—X-ray Effects

A subset of 8 lots, with either relatively high filled percentages or previous high germination success, were used in trial 2. Seedlots were spread across the geographic range and across collection years. A total of 100 randomly selected seeds that were **not** X-rayed were divided into 2 reps for this trial.

Individual seedlots/ reps for trials 1 and 2 were labeled and placed in mesh stratification bags (Figure 2). Using standard protocols for seed stratification at DGRC, seeds were soaked in 1% H₂O₂ for 24 hours, rinsed, and soaked in water for an additional 24 hours. Bags were then placed in covered plastic tubs for stratification at 10 °C for 30 days, then 1 to 2 °C for an additional 90 days. All seeds were rinsed once per week during stratification.

At the end of the stratification period, seeds were abraded, using 100-grit sandpaper, approximately 1 mm back from the radicle end and along the main line dividing the 2 halves of the seedcoat. This process was done to help maximize germination percentage as well as uniformity of germination. Five seedlots had a percentage of seeds that began to crack during stratification. These seeds were not sanded. All seeds were placed on moistened blotter paper in 10 x 10 cm clear plastic boxes (Figure 3), which were then placed in a germinator maintained at 16 °C night/18 °C day with a 12-hour photoperiod.

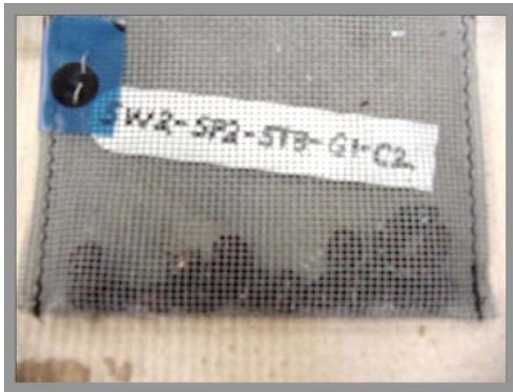


Figure 2. Individually labeled mesh bags for seed stratification.

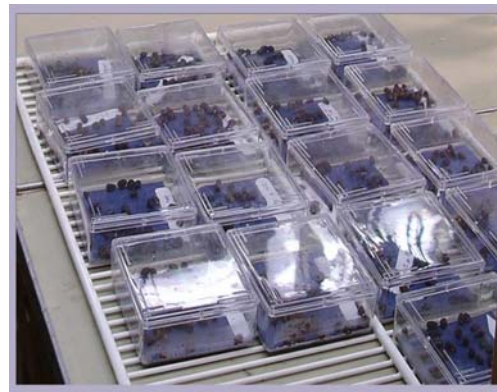


Figure 3. Whitebark pine seeds in individually labeled germination boxes.

All lots were monitored for germination twice per week for 21 days. Seeds were considered germinated when the radicle protruded from the seedcoat to a length of 2 mm and was curved. Germinated seeds were removed from the germination boxes, sown into individually labeled Ray Leach SuperCells® (164 cm³) (Stuwe and Sons, Inc., Corvallis, OR) containing pre-fertilized (180-day Nutricote® control-release fertilizer [18-6-8 with minors]) and pre-moistened media (peat:composted fir bark:perlite:pumice [40:20:20:20]), and covered with nursery grit. Seedlings were cultured under standard protocols for whitebark pine at DGRC (see Riley and others, this proceedings, for further information on seedling culture).

Trial 3—Tetrazolium Testing

A subset of 8 seedlots (same lots as those used in trial 2) were used in this trial. A total of 100 randomly selected seeds were X-rayed and then sent to the USDA Forest Service National Seed Laboratory in Athens, GA for tetrazolium testing to determine viability. (See Grabe (1970) and Peters (2000) for information on tetrazolium testing.)

Results

Trial 1

Germination by collection year

Germination varied widely by seedlot, ranging from 21% to 92%, with an overall average of 62%. Germination percentage was relatively high overall (Figure 4); 81% of the individual seedlots averaged greater than 50% germination, and 67% averaged greater than 70% germination. Although seeds stored for less than 5 years had significantly higher germination rates ($P < 0.05$), it appears that seeds are still viable up to 10 years after collection. Previous germination percentages for most seedlots used in earlier sowings compared favorably to current results, although some varied significantly (Table 3). (Seedlots collected in 2005 did not have previous germination percentages.) Seeds collected in 1999 displayed lower germination presumably due to the large number of immature seeds. Burr and others (2001) found that immature seeds do not store well for long periods of time. Other variations may have been due to early incorrect stratification and seed handling methods (Riley and others, this proceedings).

Table 3. Comparison of germination percentages for seedlots used in previous sowing (no 2005 seedlots).

Seedlot #	Coll Year	Origin	Average Previous germination	Current Study germination
9	2003	Mt Hood NF	72	84
10	2003	Mt Hood NF	86	76
11	2003	Umatilla NF	83	83
12	2003	Umatilla NF	85	76
13	2003	Umatilla NF	73	48
14	2001	Deschutes NF	45	83
15	2001	Deschutes NF	66	68
16	1999	Fremont NF	70	13
17	1999	Fremont NF	21	20
18	1997	Colville NF	25	65
19	1997	Colville NF	34	48
20	1996	Colville NF	22	25
21	1996	Colville NF	46	39

Seeds which were stored less than 5 years (2001 through 2005 collections) germinated more quickly than those stored for longer periods of time (Figure 5). Nearly half of all seeds in these lots germinated within 4 days. However, the majority of seeds for all collection years germinated within 10 days.

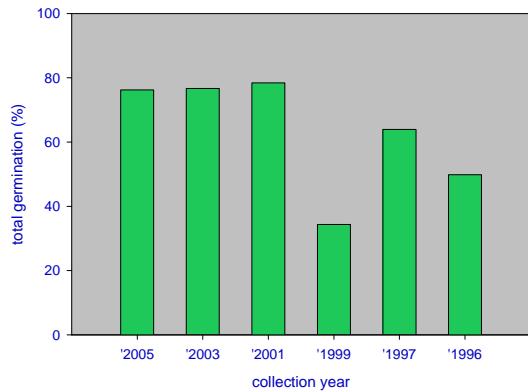


Figure 4. Total germination by collection year (all categories of filled embryos).

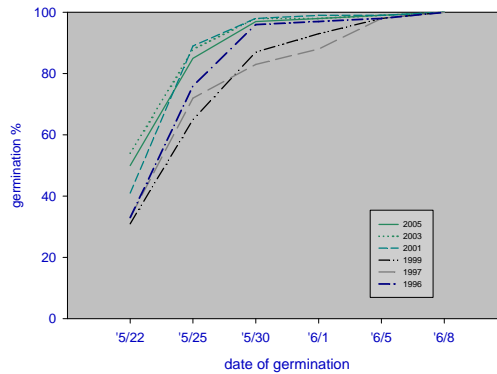


Figure 5. Speed of germination by collection year (all categories of filled embryos).

Germination by embryo-fill category

No significant difference was found in germination percentage between the >75% filled embryo cavity and the 50% to 75% filled embryo cavity categories (Figure 6). Although germination was significantly lower ($P < 0.0004$) in seeds with <50% filled embryo cavity, over half of the seeds in this category germinated.

No significant differences were found in speed of germination between embryo-fill categories (Figure 7).

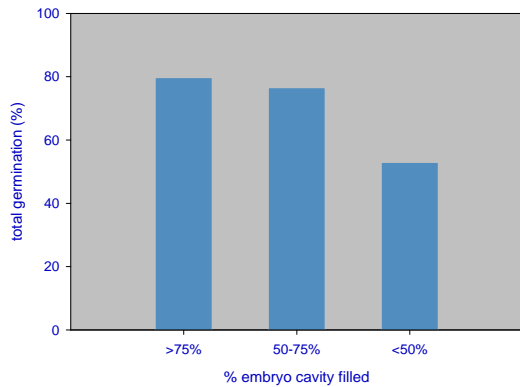


Figure 6. Total germination by embryo-fill category (all collection years).

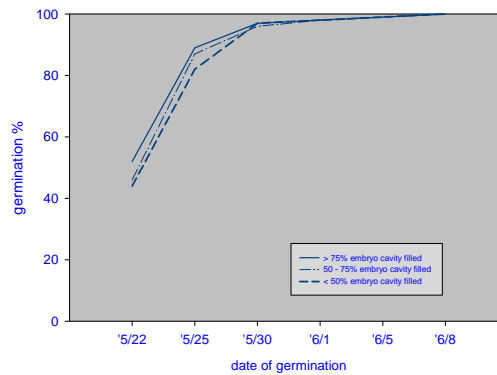


Figure 7. Speed of germination by embryo-Fill category (all collection years).

Trial 2

Effects of X-ray on seed viability

No significant differences were found in germination percentages or speed of germination between X-rayed seeds and seeds which were not X-rayed (Figures 8 and 9).

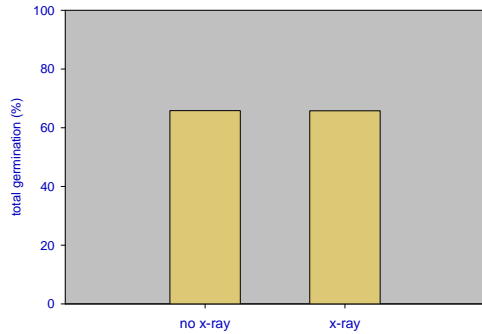


Figure 8. Total germination for X-rayed seeds vs seeds which were not subjected to X-ray (all collection years).

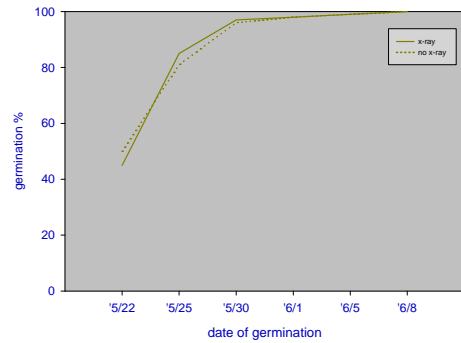


Figure 9. Speed of germination for X-rayed seeds vs seeds which were not subjected to X-ray (all collection years).

Trial 3

Tetrazolium testing vs X-ray and germination

The relationship of tetrazolium testing results with seed X-ray and actual germination percentages varied between seedlots (Figure 10). Older lots (sow numbers 17 and 21) showed more variation between test types than seedlots that had been in storage for shorter periods of time. However, there were no significant differences found overall when test results were averaged over all seedlots.

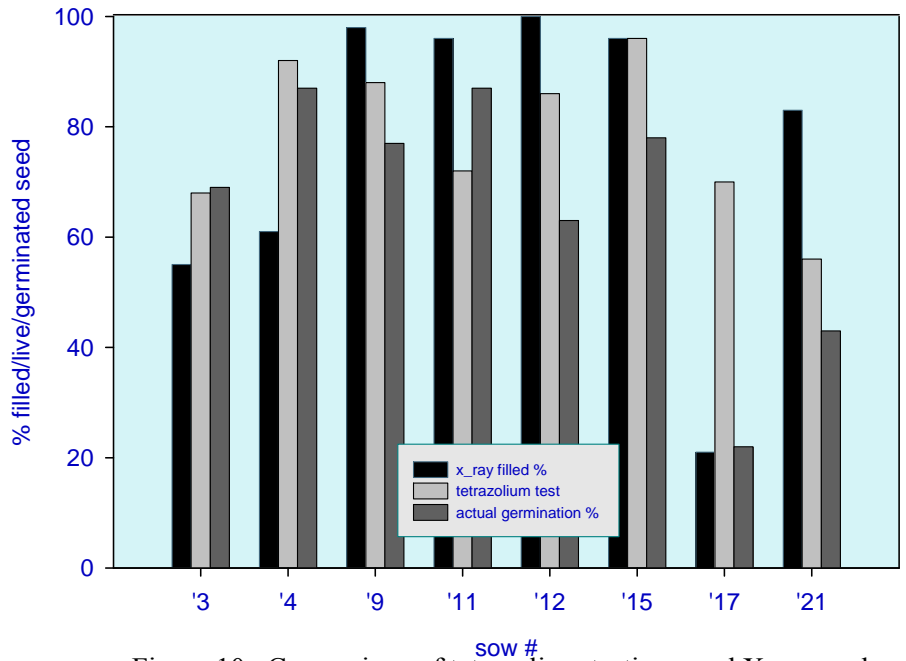


Figure 10. Comparison of tetrazolium testing, seed X-ray, and germination as predictors of seed viability.

Conclusions

The effects of storage time on whitebark pine seed viability has been studied over a number of years, with differing results. Although relatively small numbers of seeds were used in this study, some trends are apparent. Under standard seed storage conditions at DGRC, it appears that whitebark pine seeds from Oregon and Washington can be stored effectively and maintain fairly high viability for up to 10 years. However, it does appear that younger seeds (in storage less than 5 years) do complete germination faster than those stored for longer periods of time.

Seeds in which the embryo fills >50% of the cavity appear to have a high potential for germination. However, even seeds where the embryo cavity has a filled percentage of <50% may still be viable, which is in contrast to seeds of many other conifer species. Leadem (1986) found that maintaining imbibed whitebark pine seeds under warm conditions for 30 to 60 days (warm stratification) effectively promoted the development of immature tissue. That is, seeds with a significant proportion of apparently immature embryos (as compared with other conifer species) may still germinate successfully.

As with other conifer species, X-rays do not appear to affect germination success. In comparison to other testing methods, including laboratory tetrazolium testing or actual germination, properly scored X-rays do appear to be a good predictor of germination potential.

Further study may be needed on the question of storage time, as well as collection date, as further seed collections are completed. Potential future studies could include: 1) the effects of storage time on germination success and speed of germination by testing the same families every 3 years for up to 20 years; 2) the effects of collection year on germination success and speed of germination by collecting from the same trees over time; and 3) the effects of collection date on germination success and speed of germination.

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Seedling Nursery Culture of Whitebark Pine at Dorena Genetic Resource Center: Headaches, Successes, and Growing Pains

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Abstract

The production of whitebark pine (*Pinus albicaulis*) seedlings for white pine blister rust (*Cronartium ribicola*) resistance screening and reforestation has required the development of nursery culturing regimes to optimize both germination and subsequent growth. Germination and culturing studies have been conducted to modify and adapt growing regimes to the environment of USDA Forest Service Dorena Genetic Resource Center. As a result, seedlings of sufficient height and caliper are now produced in 2 years for both blister rust resistance screening and outplanting on USDA Forest Service and USDI National Park Service lands.

Introduction

Production of whitebark pine (*Pinus albicaulis*) seedlings in nurseries, for both disease resistance screening and reforestation, requires overcoming some inherent problems of regeneration in the species. Seed germination is generally both poor and erratic; only 10% to 15% germinate in the first year under natural conditions (McCaughey 1993, cited in Burr and others 2001). The presumed reasons for this include: predation (and caching) of seed crops before embryo maturity by various animals, lack of appropriate substrate and climatic conditions, complex dormancy-release physiological requirements, and extremely hard seedcoats. These factors can be positively adaptive in a natural environment over a period of time by providing a small supply of germinants over a period of 2 to 3 years, given the proper conditions. However, they present serious challenges in a nursery environment.

Depending on various factors, up to 100% germination can be achieved under strict laboratory conditions followed by intensive greenhouse culturing and monitoring (McCaughey 1992; Riley unpublished data). The challenge has been to determine a regime that is reliably successful, in an offsite environment, for a broad range of seedlots. Previous literature on attempts to successfully produce whitebark pine seedlings is not extensive, and determining a routinely satisfactory protocol has been difficult. Stratification regimes and seed pretreatments vary (Krugman and Jenkinson 1974; Arno and Hoff 1989; Rose and others 1998), and little research has been done on culturing regimes. The most successful procedures have been those developed by USDA Forest Service Coeur d'Alene Nursery in Coeur d'Alene, Idaho (Burr and others 2001). However, other nurseries must adapt growing regimes to fit their particular climates. USDA Forest Service Dorena Genetic Resource

Center (DGRC), located in western Oregon at 43°47', -122°58', elevation 285 m, spent several years adapting these regimes to the growing environment at that location.

Initial Attempts at Seedling Culture

In cooperation with the USDA Forest Service and USDI National Park Service, Dorena Genetic Resource Center has sown and cultured approximately 178 families of whitebark pine, from a wide range of locations throughout the Pacific Northwest from 1996 to 2006. Seedlings have been used in white pine blister rust (*Cronartium ribicola*) resistance screening as well as outplanting on Federal lands. Due to limited information resources and a number of environmental variables, early sowings resulted in low germination and few viable seedlings.

Direct Sowing into Rust Testing Frames

Although DGRC has been expanding the rust resistance testing program over the years to include other five-needle pines, western white pine (*Pinus monticola*) and sugar pine (*P. lambertiana*) have been the traditional species sown for standard testing. Nursery protocols for these species have been modified over the years. Following appropriate seed handling and stratification, standard nursery practice includes hand-sowing of both species directly into 1 x 1.2 x 0.3 m testing frames located in an outside growing area and culturing for two years prior to inoculation with rust spores (Figure 1).

In 1996, 2 seedlots of whitebark pine were available for blister rust testing at DGRC. Two hundred seeds from these lots were soaked in 1% H₂O₂ for 24 hours, rinsed, and soaked in water for an additional 24 hours. They were then placed in cold stratification at 1 °C for 90 days, hand-sown directly into rust testing frames with western white pine, and cultured under standard nursery regimes for western white pine and sugar pine. Approximately 5% of the seeds from all 3 lots germinated, resulting in insufficient seedlings for rust testing (Figure 2).



Fig. 1. Hand-sowing of western white pine seeds in frames for blister rust testing.



Fig. 2. Direct-sowing of whitebark pine seeds into rust testing frames with western white pine. Whitebark pine is located in the 3rd row from the left.

Direct Sowing into Seedling Containers

In 2000, the decision was made to sow whitebark pine seeds into containers to culture under greenhouse conditions for rust testing. Two hundred seeds from each of 6 lots of whitebark pine were soaked in 1% H₂O₂ for 24 hours, rinsed, and soaked in water for an additional 24 hours. They were then placed in cold stratification at 1 °C for 90 days, scarified using a razor blade to remove a portion of the seedcoat, and sown directly into Ray Leach SuperCells® (164 cm³) (Stuewe and Sons, Inc., Corvallis, OR) in a greenhouse. As with the 2000 sowing, the resulting germination was extremely low (Figure 3A). Approximately 11% of the seeds in these seedlots did germinate and emerge, but several were damaged by both mice and birds in the greenhouse (Figures 3B and 3C).

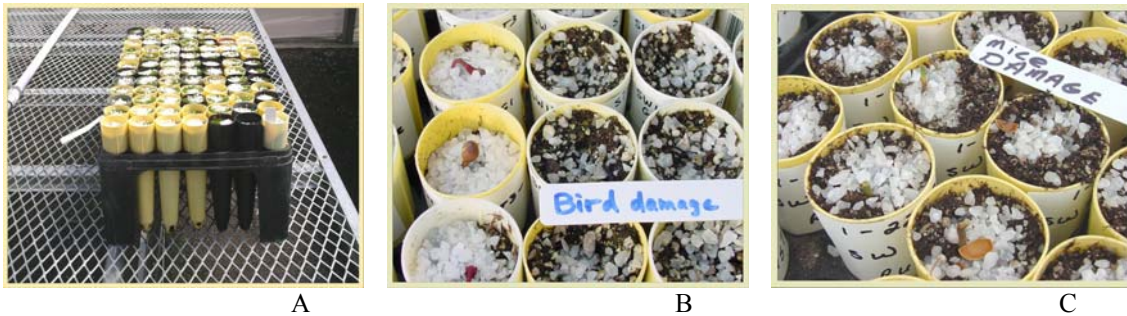


Fig. 3. Extremely low germination resulting from hand sowing whitebark pine seeds directly into containers following stratification and scarification (A). The few germinants that did emerge were damaged by both birds (B) and mice (C).

Culturing Trials

2002 Whitebark Pine Cultural Regimes Study

Due to the failures of the 2 previous attempts to germinate and culture whitebark pine at DGRC, a study was implemented in 2002 to determine seed pre-treatment, stratification, germination, and culturing regimes that would provide optimal conditions for germination and growth. This study was based on the protocols developed by Burr and others (2001) and Burr (2002) at USDA Forest Service Coeur d'Alene Nursery (CDAN). The objective was to compare CDAN protocols with other pre-sowing and post-sowing treatments to determine proper growing procedures in a different growing environment and with seedlots from different provenances.

Materials and Methods

Twenty seedlots of whitebark pine were used in this study. The seedlots ranged in storage time from 0 to 7 years. Nineteen of the lots were individual tree collections from 5 forests in Washington and Oregon; one lot was a bulk seedlot received from CDAN.

Seeds from each lot were X-rayed on a template to extract only filled seeds. A total of 504 filled seeds from each whitebark pine seedlot were divided into a total of 24 treatments.

Seed pretreatment. Seeds from all lots were divided equally into 2 treatments (252 seeds each). In treatment 1 (control as utilized by CDAN), seeds from each lot were placed in labeled mesh bags and soaked for 48 hours in running tap water to remain aerated during water absorption. In treatment 2 (standard procedure at DGRC), seeds from each lot were placed in labeled mesh bags, soaked for 24 hours in a 1% H₂O₂ solution, rinsed, and then soaked for an additional 24 hours in water. All seeds were rinsed once per week with water during stratification, and moldy seeds were rinsed well with 1% H₂O₂

Stratification time. All seeds were placed in warm stratification (20 °C night/22 °C day) for 28 days (Burr and others 2001). Following the warm stratification period, seeds were further subdivided (from different soaking treatments) into 3 cold stratification periods: 30 days, 60 days (control), and 90 days at 1-2 °C (84 seeds each).

Germination temperature. At the conclusion of all stratification periods, seeds were nicked with a scalpel or razor blade, approximately 1 mm from the radicle end, along the main line dividing the 2 halves of the seedcoat and placed on moistened blotter paper in 10 x 10 cm clear plastic boxes. Seeds were further subdivided into 2 treatments (42 seeds each). Boxes containing the control seeds (CDAN) were placed in a germinator maintained at 20 °C night/22 °C day with a 12-hour photoperiod. Boxes containing seeds in treatment 2 were placed in a germinator maintained at 16 °C night/18 °C day with a 12-hour photoperiod.

All lots were monitored for germination every 48 hours. Seeds were considered germinated when the radicle protruded from the seedcoat to a length of 2 mm. Germinated seeds were removed from the germination boxes, planted into Ray Leach SuperCells® (164 cm³) containing pre-fertilized and pre-moistened “seedling mix,” and covered with 0.3 to 0.6 cm nursery grit. Following planting, supercells were maintained at a constant temperature of 21 °C for 7 days with 18 hour photoperiod in a building, and then moved to greenhouses.

Culturing. Two different greenhouse environments were tested during this study. Cells containing germinated seeds were further subdivided (up to 21 seeds each) into either a climate-controlled greenhouse (with daytime temperatures ranging from 21 to 27 °C throughout the growing season) or a standard greenhouse subject to ambient conditions (with daytime temperatures ranging from 21 to 38 °C throughout the growing season).

Results

Overall germination percentage was significantly different ($P < 0.0001$) between seedlots. Although germination percentage based on age of the seedlots was confounded by geographic origin, some older seedlots ranged from 80% to 100% germination in some treatments.

Seed pretreatment. No significant difference was found in total germination percentage between seed pretreatments. However, the standard DGRC pretreatment of 24 hour soak in H₂O₂ resulted in slightly higher overall germination and slightly faster speed of germination.

Stratification time. There were significant differences ($P < 0.0001$) between stratification treatments in overall germination percentages and speed of germination. The 120-day

stratification treatment yielded both higher germination percentages and speed of germination than either the 90-day or 60-day stratification periods. Total germination for seeds stratified for 120 days ranged from 44.6% to 97.0%, with an overall average of 68.9%; seeds stratified for 90 days ranged in germination from 31.5% to 81.5%, with an average of 61.6%; seeds stratified for 60 days ranged in germination from 21.4% to 54.2%, with an average of 36.9%. In addition, depending on the seedlot, the 120-day stratification period resulted in higher numbers of seeds that did not require scarification due to seedcoat splitting.

Germination temperature. No significant difference was found in either total germination percentage or speed of germination between germination temperature regimes. However, the higher night/day temperatures (20 °C/22 °C) resulted in higher numbers of moldy seeds during the monitoring period.

Culturing. Seedlings from all seedlots and germination treatments that were placed in the climate-controlled greenhouse were significantly shorter in height at the end of the growing season than those grown in ambient temperatures. Average heights (by seedlot) for seedlings grown in the climate-controlled house ranged from 2 to 4.5 cm, with an overall average of 3.7 cm; average heights (by seedlot) for seedlings grown at ambient temperatures ranged from 5.2 to 7.1 cm, with an overall average of 6.3 cm. However, observationally, it appeared that budset occurred earlier in seedlings subjected to the higher greenhouse temperatures.

The results of the culturing portion are somewhat counter-intuitive. One explanation for the decreased growth rates in the climate-controlled greenhouse may be that the greenhouse was used for other purposes during that growing season. As a temporary measure, the seedlings were grown on pallets on the floor of the greenhouse directly in front of the cool cells. The resulting cooler temperatures in this location may have precluded the proper release of the control-release fertilizer in the cells.

2006 Whitebark Pine Scarification Study

Depending on the seedlot, many seeds require scarification to increase germination potential. The majority of seedlots sown for testing at DGRC are small lots; each lot is usually comprised of less than 200 seeds. Since scarification methods used for larger bulk lots are impractical, each seed must be hand-nicked if the seedcoat has not cracked during stratification.

Nicking seeds with a scalpel presents concerns for both human safety as well as the possibility of damaging the megagametophyte tissue prior to germination. Nicking is also very slow and labor intensive. For these reasons, a small trial was implemented to determine if light abrasion of the seedcoat caused by sanding would produce similar or better germination potential as nicking seeds with a scalpel.

Materials and Methods

In spring 2006, 8 individual seedlots from Crater Lake National Park and 1 bulk lot from the Deschutes National Forest were used in a scarification study. Fifty seeds from each of the 9 lots were stratified for 30 days at 10 °C and 90 days at 1-2 °C. Following stratification, 25 seeds from each lot were nicked with a scalpel using the standard protocol, and 25 seeds

were lightly hand-sanded using 100-grit sandpaper. Seeds were then placed on moistened blotter paper in labeled 10 x 10 cm clear plastic boxes. Boxes were placed in a germinator maintained at 16 °C night/18 °C day with a 12-hour photoperiod. Germination was monitored twice per week for 4 weeks.

Results

Germination differed significantly between seedlots, with germination percentages ranging from 14% to 84%. However, no significant difference was found between scarification treatments. The seed abrasion treatment appeared to yield similar results to the hand-nicking, but was much faster and easier.

Current Cultural Methods

Culturing regimes for whitebark pine at DGRC have evolved since 2000 based on experience, as well as the results of culturing studies and trials. These regimes may not be applicable to other situations, but appear to work well in the growing climate at DGRC. Current methods used at DGRC are detailed below. For further information on terminology and physiological details of whitebark pine seedling culture, refer to Burr and others (2001).

Stratification

Stratification is defined as a seed pregermination treatment to break seed dormancy and provide for uniform germination, and is accomplished by placing seeds in a moist cool environment (often preceded by a warm period) to hasten afterripening (Schopmeyer 1974; Bonner 1984). Seeds at DGRC are placed in individually labeled mesh bags (Figure 4), soaked for 24 hours in 1% H₂O₂, rinsed, and soaked an additional 24 hours in water to take up water as well as reduce the levels of seedborne pathogenic fungi. Mesh bags are hung on dowels in covered plastic tubs, placed in warm stratification at 10 °C for 30 days, then at 1 to 2 °C for 90 days. The warm stratification temperature is actually cooler than temperatures tested in previous cultural trials. This temperature appears to reduce the occurrence of mold as well as the number of early germinants. All seeds are rinsed once per week with water during stratification, and moldy seeds are rinsed well with 1% H₂O₂.



Fig. 4. Individually labeled mesh bag for seed stratification.

Scarification and Germination

When stratification is complete, the seedcoat of each seed is abraded, using 100-grit sandpaper, at the radicle end (approximately 1 mm back from the tip) and along the main line dividing the 2 halves of the seedcoat (Figure 5A). Care must be taken to avoid sanding too heavily, as damage to the megagametophyte tissue could result. This process is done to help maximize germination percentage as well as uniformity of germination. Seedlots which are not sanded or nicked show lower germination percentage and germinate at a slower rate (Burr and others 2001). A few seedlots have a percentage of seeds that begin to crack during

stratification. These seeds are not sanded. During the 2007 sowing season, a newly designed sanding machine will be used for seedcoat abrasion (Figure 5B) (Spence 2006).

Sanded seeds are placed on moistened blotter paper in 10 x 10 cm clear plastic boxes (Figure 6A), which are then placed in a germinator maintained at 16 °C night/18 °C day with a 12-hour photoperiod (Figure 6B).

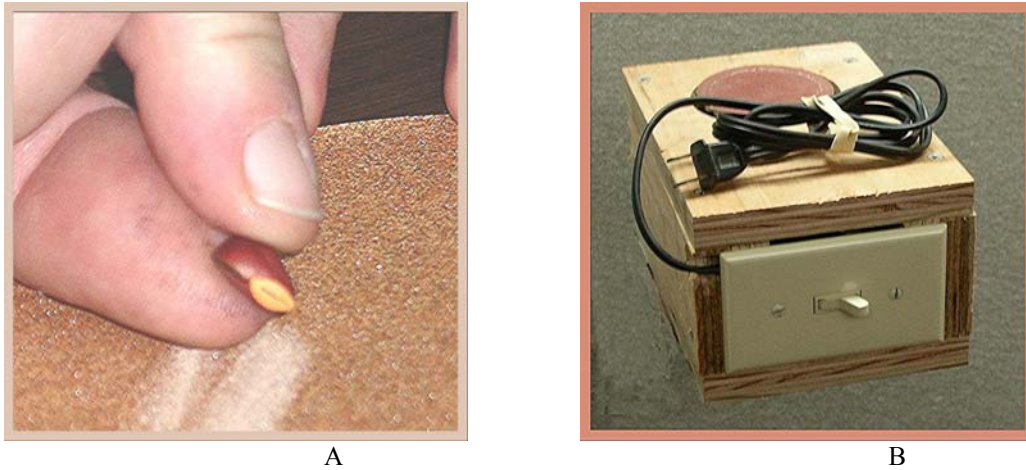


Fig. 5. Seedcoat abrasion using 100-grit sandpaper (A). Sanding machine, designed and built at DGRC, to be used for seedcoat abrasion (Spence 2006) (B).

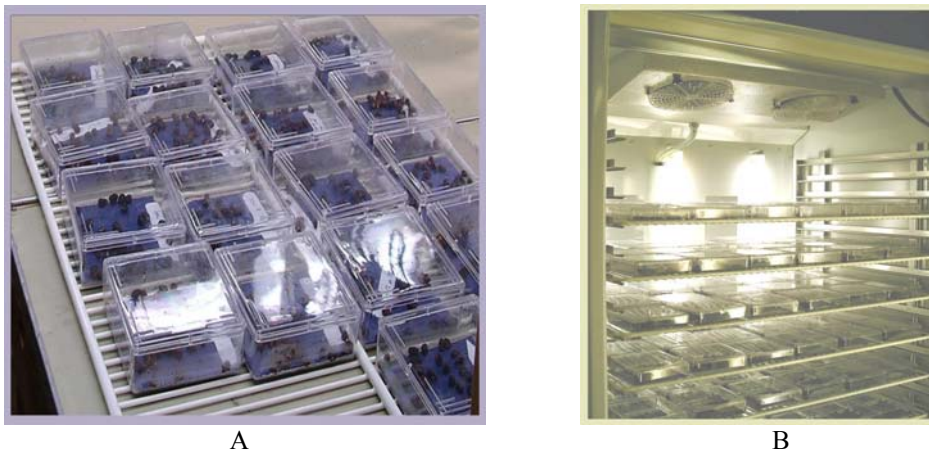


Fig. 6. Whitebark pine seeds in individually labeled germination boxes (A), which are then placed in a germinator maintained at 16 °C night/18 °C day with a 12-hour photoperiod (B).

Seedlots are monitored for germination twice per week for 5 weeks, and are considered germinated when the radicle protrudes from the seedcoat to a length of 2 mm and is curved (Figure 7). Germinated seeds are removed from the germination boxes, sown into individually labeled Ray Leach SuperCells® (164 cm³) containing pre-fertilized (180-day Nutricote® control-release fertilizer [18-6-8 with minors]) and pre-moistened media (peat:composted fir bark:perlite:pumice [40:20:20:20]), and covered with nursery grit (Figure

8). Following sowing, supercells are placed in racks on covered tables inside a heated building and kept at approximately 21 °C (18-hour photoperiod) for 7 days.

At the end of this period, racks are moved to a greenhouse with lights (18-hour photoperiod) for the remainder of the growing season. All tables are covered with a mesh screen prior to rack placement, and racks are covered with mesh screen tops to protect seeds from birds and rodents (Figure 9). Covers remain in place until seedcoats have been shed from the seedlings (approximately 2 weeks following emergence).



Fig. 7. Germinated whitebark pine seeds prior to sowing.

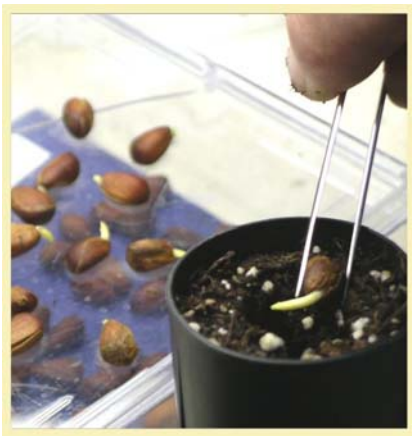


Fig. 8. Sowing germinated seed.



Fig. 9. Seedling protection in the greenhouse.

Culturing—Year One

During the first growing season, supplemental light is used in the greenhouse (18-hour photoperiod) from germination through the middle of October. Supplemental lighting is used to mimic high elevation growing conditions as well as extend the growing season outside the normal range of the species.

Seedling nutrition includes the initial application of control-release fertilizer prior to sowing, as well as a variety of soluble fertilizers, depending on the growth stage, throughout the growing season. Fertilization is initiated when seedcoats have been shed from the seedlings. Low vigor seedlings may have difficulty shedding seedcoats (Burr and others 2001), so it may be necessary to remove seedcoats by hand. Soluble fertilizers are applied once per week through the middle of October.

Seedlings are overwintered in a greenhouse with the sides open to achieve appropriate chilling hours.

Culturing—Year Two



Fig. 10. Fertilization of individual cells at the beginning of the second growing season.

During mid-February of the second growing season, the greenhouse sides are closed, although no supplemental heat is added to the growing environment. Seedlings begin to receive supplemental lighting (18-hour photoperiod), which will continue through the middle of August. Nutricote® control-release fertilizer 140-day (18-6-8 with minors) is added to each cell by hand (Figure 10).

Seedling nutrition includes the application of control-release fertilizer, as well as a variety of soluble fertilizers, applied once per week through the middle of August. During the middle of August, seedlings are moved to an outdoor growing situation for hardening-off prior to either disease resistance testing or outplanting.

Seedling Results

The cultural regimes currently in use at DGRC have resulted in 2-year-old seedlings of sufficient height and caliper for use in either rust testing or outplanting (Figure 11). Two-year seedling heights range from a minimum of 6 cm to a maximum of slightly over 14 cm depending on seed source (Figure 12). To date, 437 whitebark pine seedlots (401 individual tree selections) have been sown and cultured for rust testing and/or outplanting on USDA Forest Service and USDI National Park Service lands. In 2007, an additional 247 seedlots are scheduled to be sown for testing and outplanting in 2008.



Fig. 11. Two-year old seedlings grown at Dorena Genetics Resource Center.

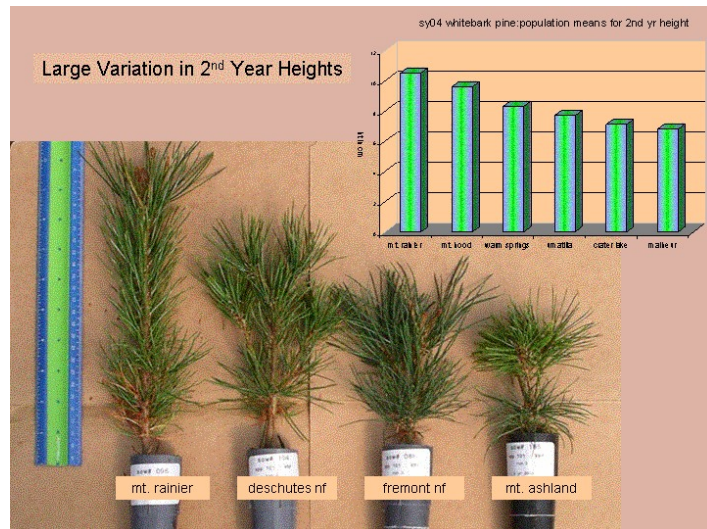


Figure 12. Variation in 2nd year height growth, depending on seed source.

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First Year Results of a Whitebark Pine Seed Planting Trial Near Baker City, Or.

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Abstract

Whitebark pine occurs primarily in roadless areas, national forests, wilderness areas, or national parks and is declining throughout most of its range. Planting seedlings to restore whitebark pine in remote areas may be difficult logistically and not compatible with wilderness use, so a trial planting of 700 whitebark pine seeds was established in a whitebark pine site on Vinegar Hill near Baker City, Oregon. The trial included three treatments to enhance germination (warm stratification, seed scarification, and a combination of both), and three treatments to reduce rodent predation (Thiram, cayenne pepper, and caging). The results after nine months indicate warm that stratification can significantly enhance germination, but treatment differences for rodent repellants were not yet observed.

Introduction

Whitebark pine is a keystone species of high elevation ecosystems throughout western North America (Tomback et al. 2000). Much of this range occurs in remote or relatively inaccessible areas where routine restoration by planting seedlings may not be possible (Keane 2000). Recent studies have found whitebark pine is in peril in much of its range (Kendall and Keane 2000, Schwandt, 2006), so alternatives such as direct planting of seeds need to be investigated. Whitebark pine seeds normally take at least two years to germinate (McCaughey and Tomback 2001, Tomback et al. 2001), but previous tests have indicated it may be possible to shorten the germination period by using a warm stratification period followed by cold stratification or by scarifying the seed coat (Burr et al. 2001). Rodent predation of planted seed may also be a major factor in restoration success (McCaughey pers. com) and needs further investigation.

Therefore a pilot study was designed to determine 1) if seed could be planted effectively and 2) if different seed treatments could be used to enhance germination and reduce rodent predation. This study was implemented on whitebark pine sites with good access, local seed already in storage, and strong interest by local resource professionals.

Methods and Materials

On November 1, 2005 a trial project using a total of 700 whitebark pine seeds from a local seed source was installed in a whitebark pine site at Vinegar Hill, about 35 miles west of Baker City, Oregon. This area of the Malheur National Forest is a prime whitebark pine site, about 7600 ft. in elevation along a southwest facing ridge with scattered clumps of whitebark pine mixed with lodgepole pine and subalpine fir (fig. 1).

The subalpine fir and lodgepole pine had been cut or girdled to enhance growth of existing whitebark pine and encourage natural regeneration. The slash piles had been burned the previous year and the small burned areas were selected to install five experimental blocks or treatment replicates containing seven 20-seed treatments per replicate. The seven treatments included three treatments to enhance germination and three treatments to reduce rodent predation plus a control. Treatments were randomly assigned locations within each of the replicates.



Fig.1. Small burned area and surrounding vegetation on Vinegar Hill prior to installation of seed planting trial.

To test for warm stratification effects, 200 seeds were washed and stored 21 days at 50 °F prior to planting. One hundred of these seeds were individually hand-scarified with sandpaper after the warm stratification. Another one hundred seeds were scarified only, and an additional 100 seeds were used as controls and planted without any scarification or warm stratification. All the treatments to enhance germination were covered with hardware cloth cages to prevent rodent predation since prior tests of direct seeding suffered severe rodent predation (McCaughey 1990).

Cages were made from a standard roll of 19 gauge ½” mesh 24” wide hardware cloth cut into 7 foot lengths with a 6 inch top and 9 inch sides. The lower 3 inches of each side were bent out to create a lip to prevent rodents from burrowing down the side and under the cage (fig



Figure 2. Hardware cloth cages to prevent rodent predation and logs to provide shade

2). Cages were buried 2-3 inches deep and anchored with six inch nail spikes. Treatments using the remaining 300 seeds were not covered with hardware cloth to test efficacy of two rodent repellants, Thiram® and cayenne pepper dust, against an untreated control. Thiram® (tetramethylthiuram disulfide) is used as a seed protectant for fungal diseases and as an animal repellant to protect fruit trees and ornamentals from damage by rabbits, rodents, and deer (Meister, R.T. 1992). Seeds were soaked for 2 minutes in a solution of Thiram diluted 1:1 with tap water, allowed to dry 3 hours, and then soaked a second time and dried 8 hours at room temperature.

Rodent repellants using hot pepper are also commonly recommended for rodent control (Smith, pers. com.).



Fig. 3. Cayenne pepper treatment

Cayenne pepper powder was shaken into a bag with 100 seed and also liberally shaken into and around the planting hole after planting each seed in this treatment (fig 3.)

Seeds in each treatment were sown in a single row, six inches apart, and one inch deep using a flat rubber template with pre-drilled holes (fig. 4). All but 3-4 inches of twelve inch metal stakes were permanently buried three inches from the last seed at both ends of each row to facilitate measurements in the future. Rows were spaced at two foot intervals and logs 8-12 inches in diameter and seven feet long were placed 3 inches from each row of seeds to provide shade. Logs were oriented on a bearing of 300 degrees to allow early morning sun on the treatments but provide maximum shade in the afternoon. Hobo® weather instruments were installed to monitor relative humidity, precipitation, and ambient temperature every hour.

The site was re-visited July 19, 2006 (nine months after planting) check for germination in the various treatments. Seed locations were found using a tape marked at six inch increments and stretching between the two metal stakes used mark both ends of each row (fig 5). Germination at each seed planting location was noted along with notes regarding seedling condition (live, fading, top kill, or dead) and brush encroachment. Results were tested using the Pearson Chi-square test for statistical significance



Figure 4. Planting Thiram coated seed through rubber template with holes at 6 inch intervals



Fig. 5. Whitebark pine seedling 9 months after planting

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Results and Discussion

A total of 94 of the 700 seeds germinated between November 1, 2005 and July 19, 2006.

Germination of the warm stratification treatment seeds was 38%, and warm stratification plus scarification treatment seeds had 25% germination. These treatment differences were highly significant ($P < .001$) from all other treatments (fig. 6).

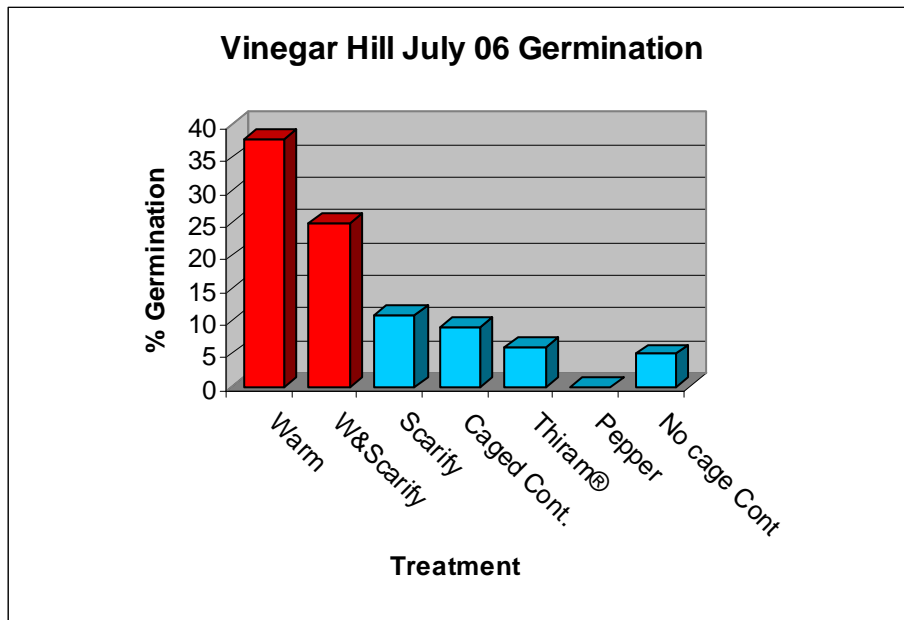


Fig. 6. Percent whitebark pine seed germination after 9 months, by treatment

Germination in the five replicates varied from a total of 7 (5%) to 27 (19.3%) seeds out of 140 seeds per replicate (Table 1.) The warm stratification treatment had significantly greater germination in three of the five replicates while the warm stratification combined with seed scarification had significantly greater germination in one other replicate. Germination was very low for all treatments in the first replicate.

Table 1. Whitebark pine germination in each treatment and replication – July, 2006; significant (*) and highly significant (**) treatment differences.

TREATMENTS\ REPS:	1	2	3	4	5	totals
Warm Stratification (caged)	2	9*	2	12*	13*	38**
Warm Strat. & Scarify (caged)	0	6	12*	1	6	25**
Scarify only (caged)	1	5	2	0	3	11
Caged Control	0	3	5	0	1	9
Pepper (no cage)	0	0	0	0	0	0
Thiram® (no cage)	1	3	2	0	0	6
No cage Control	3	1	1	0	0	5
Total Seeds Germinated	7	27	24	13	23	94
Percent Germination	5.0	19.3	17.1	9.3	16.4	13.4

Since germination normally takes at least two years (McCaughey 1993, Tomback et al. 2001), not much germination was expected in the rodent repellent treatments or controls. None of the 100 seed that were treated with cayenne pepper germinated and only 6 % of the Thiram treated seed germinated. These were not significantly different from the 9% germination in the caged control and the 5% germination in the uncaged control. Four percent of the 94 germinants were dead and a few others were wilted or chlorotic.

Some plots had abundant shrub growth (figs. 7a & b) which may provide additional shading for seedlings. However, it also made it difficult to locate seedlings and could create problems for seedling growth in the future. The wire mesh cages sustained minor animal or snow damage during the winter so additional braces or stiffer gauge wire may be desirable in future installations.

Further monitoring is planned for the fall of 2006 to see how the germinants survived the driest summer in many years plus annual summer and fall inspections are planned the next several years to check for additional germination and to monitor seedling survival over time. A similar test was initiated in the fall of 2006 southwest of Bend, Oregon on Mt. Batchelor and other tests are planned throughout the range of whitebark pine to get a better understanding of the potential of this technique for restoration.



Fig. 7 (A & B). Extensive shrub growth on some plots

Acknowledgements:

This study was a team effort and our sincere thanks to the dedicated group of resource professionals that scouted the area and installed the trial in a driving snow storm. This included: Vicki Erickson, Karen Prudhomme, Vince Novotny, Steve Duncan, Paul Phelps, Victoria Rockwell, Holly Kearns, and Jim Hoffman. We also greatly appreciated advice and assistance from Jerry Berdeen (Dorena Genetic Resource Center) as well as his efforts to stratify and treat the seed in time for planting.

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Modeling Contemporary Climate Profiles of Whitebark Pine (*Pinus albicaulis*) and Predicting Responses to Global Warming

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The Random Forests multiple regression tree was used to develop an empirically-based bioclimate model for the distribution of *Pinus albicaulis* (whitebark pine) in western North America, latitudes 31° to 51° N and longitudes 102° to 125° W. Independent variables included 35 simple expressions of temperature and precipitation and their interactions. These climate variables were derived from a spline climate model that provides point estimates (latitude, longitude, and altitude). The analysis used presence-absence data from more than 119,000 plot locations largely from USDA Forest Inventory and Analysis. Of these plots, 2738 contained whitebark pine. Climate estimates were made for each plot location and assembled into 10 data sets containing all of the plots where whitebark pine was present at a proportion of 40%. The remaining 60% were randomly selected from the database.

The Random Forest regression tree analysis was then applied to each of the 10 data sets to develop a model for predicting the occurrence of whitebark pine from climate, using the general approach described in detail in Rehfeldt et al. (2006). The analyses consisted of constructing 10 forests each with 100 trees. The most parsimonious model was determined through an iterative process of removing variables and repeating the analysis until an increase in out-of-bag error was observed. Output from the analyses included statistics useful for evaluating the importance of input variables. We used importance scores from the Gini index to determine variable importance. To produce mapped projections of the bioclimate model, the estimated climate for 5.9 million pixels of 1 km resolution representing the terrestrial area of western USA were run down the regression tree. Each tree of the 10 forests provided a vote on whether or not the climate of the pixel was within the profile of the species.

The most parsimonious had 9 variables and had an overall error of classification of 2.47%. Among the 9 variables, the 3 most important were degree-days > 5 °C, mean temperature of the warmest month, and Julian date when degree-days > 5 °C reaches 100. When run independently, these 3 variables classified the species occurrence with an overall error of only 4.91%. The mapped prediction for the occurrence of *P. albicaulis* using its climate profile was more accurate than its published range map (Fig 1).

This model was then used to predict responses to global warming. The Hadley and Canadian General Circulation Models (1% increase GGa/yr) were used to estimate the climate of each pixel for the area of analysis. The future climate expected in each pixel was then run down the regression tree to predict the future geographic distribution of *P. albicaulis*' contemporary climate profile. Projections were made for climates expected for the decades beginning in 2030, 2060, and 2090 (Fig 2)

Results indicated a rapid and large scale decline in the total area occupied by the species' contemporary climate profile throughout the century. For the decade beginning in 2030, *P. albicaulis*' contemporary climate profile is projected to decline 70 % while moving upward in altitude by 333 m on average. By the end of the century, its contemporary climate profile is projected to diminish to an area equivalent to less than 3% of its current distribution.

This bioclimate model for *P. albicaulis* provides a tool of high statistical precision that predicts the location of sites having a suitable climate. It can be used by resource managers as a range map and for identify geographic locations climatically suited for establishment and growth of the species. The model may also be used in conjunction with climate change scenarios to identifying populations at risk and future locations climatically suited for conservation efforts.

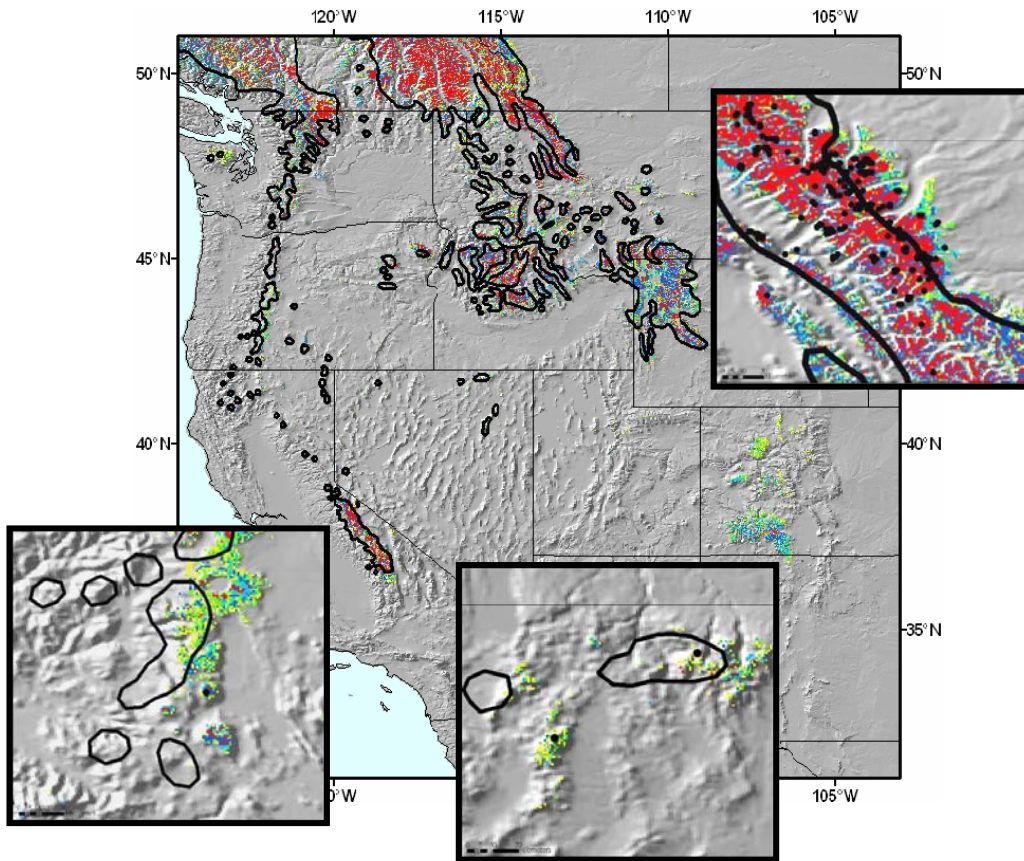


Fig. 1 Modeled bioclimate profile of *Pinus albicaulis* overlaid with the digitized range map of Little (1971). Colors code the proportions of votes received by a pixel in favor of being within the climate profile: 0%–50% no color; yellow 50%–60%; green 60%–70%; light blue 70%–80%; dark blue 80%–

90%; and 90%–100% red. Areas outlined in black depict species boundaries defined by Little (1971). Dots in the panels indicate sites known to be inhabited.

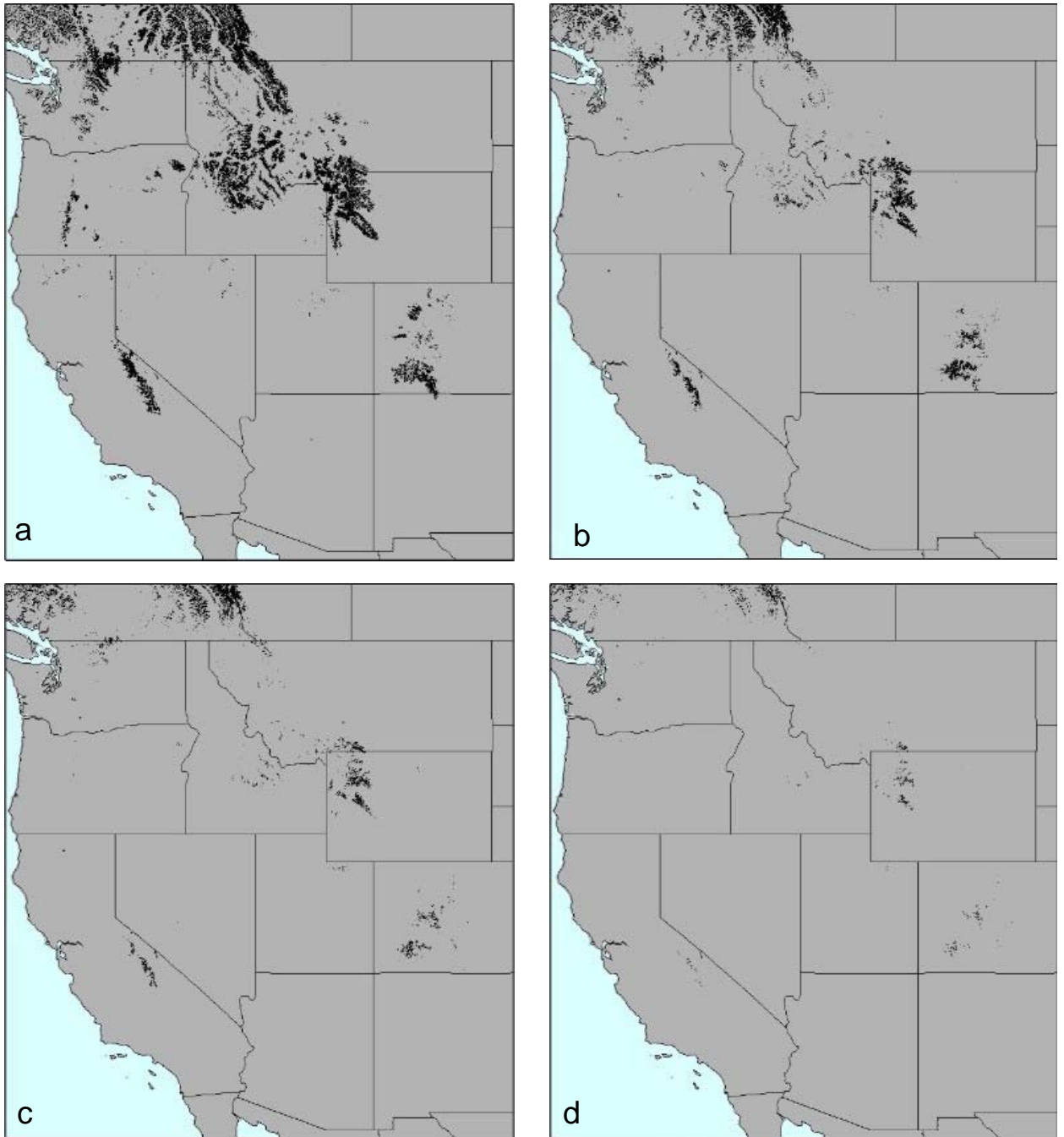


Fig. 2 Modeled bioclimate profile of *Pinus albicaulis* for the present (a) and predicted climate for decades 2030 (b), 2060 (c) and 2090 (d) under climate change scenario using an average of Hadley and CCMA GCM scenarios of 1% per year increase GPa. Black indicates location of pixels receiving $\geq 50\%$ proportion of votes in favor of being within the climate profile.

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Methods for Surveying and Monitoring Whitebark Pine for Blister Rust Infection and Damage.

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Whitebark pine (*Pinus albicaulis*) is declining nearly rangewide primarily as a result of white pine blister rust, which is caused by the exotic fungal pathogen white pine blister rust (*Cronartium ribicola*). In the northern Rocky Mountains and Intermountain Region of the United States, recent outbreaks of mountain pine beetle (*Dendroctonus ponderosae*) are also rapidly reducing populations. In these regions, losses are compounded by a trend towards successional replacement of whitebark pine by shade-tolerant conifers in seral communities. The Whitebark Pine Ecosystem Foundation (WPEF) advocates rangewide surveys and monitoring of blister rust infection levels and other mortality factors for whitebark pine. Information obtained from surveying and monitoring will enable managers to plan and prioritize geographic areas for restoration activities.

Standardized methods were developed by the WPEF in 2003 and revised in 2005 to quantify blister rust infection levels, damage, and mortality, and mortality from other factors, including mountain pine beetle spatially and temporally. Recognizing limits to personnel, time, and budgets, we designed these methods to gather critical information efficiently, with minimal technical training. Methods were developed after reviewing 10 major whitebark pine surveys of blister rust incidence, followed by discussion with resource managers, forest ecologists, and pathologists. Meaningful variables were identified as survey objectives, and are recommended for general use in these or similar survey methods. In June, 2003, we conducted a two-day methods field trial on Sawtell Peak, Caribou-Targhee National Forest, Idaho, with 11 participants, including pathologists, managers, and ecologists, representing different experience levels with blister rust surveys (Fig. 1).

The WPEF developed a methods package/database system in collaboration with David Pillmore and Brent Frakes (Rocky Mountain Network Inventory and Monitoring, USDI National Park Service) called the “White pine blister rust survey database: whitebark pine application,” Beta Test Version 2.0. This database is available at no cost from Rocky Mountain Network Inventory and Monitoring, USDI National Park Service, Ft. Collins, CO

and can be accessed through a link on the WPEF website (www.whitebarkfound.org) under “Resources”. It is a Microsoft Access-based system that stores collected data and calculates summary statistics for plots. This package also interfaces with the Whitebark-Limber Pine Information System (WLIS, developed by the USDA Forest Service, Northern Region, Forest Health Protection, Forest Health Technology enterprise Team, ver. 1.0, March 2006).



Fig. 1. Two-day methods “run-through” in 2003 on Sawtell Peak, Caribou-Targhee National Forest, Idaho, with pathologists, managers, and ecologists from the U.S. and Canada.

In June, 2004, eighty participants from across the range of whitebark pine were trained in the use of the WPEF methods at a workshop held in West Yellowstone, Montana, USA. The training session and ensuing discussion provided feedback enabling further refinement of the methods. In addition, the methods were used in 2003 by C. M. Smith for an extensive assessment in the Canadian Rocky Mountains, providing additional feedback. The revised survey methods are posted on the WPEF website under “Monitoring”

Here, we briefly describe sampling objectives, design, strategy, and key factors concerning the methodology. First of all, when a project is identified, sampling approaches must be determined in advance, with good rationale. The two alternative approaches are “statistical,” which uses a GIS-based, randomized or methodical sampling regime designed in consultation with a statistician, or “relevé,” in which sampling locations are selected based on predetermined criteria by the field crew. The statistical approach should be used when data must be compared using rigorous statistical assumptions and procedures across space and time, but it may require more funding, personnel, and time resources. The relevé approach is used when documentation of ecological changes is more important than statistically valid

estimates of change, and resources are limited. Plots are placed in a representative portion of the sample stand, and the only valid statistical comparisons are between the same plots over time.

Our survey approach is based on a fixed area monumented belt transect plot that is 10 m wide by 50 m in length (500 sq m). Besides detailed geospatial information, the transect description includes: slope, aspect, successional status, habitat or cover type, estimated proportion of each tree species in the overstory, woody and herbaceous plant dominants, and potentially rust resistant candidate trees. Each live or dead whitebark pine tree that is >1.4 m in height within the plot is permanently marked with a sequentially numbered aluminum tag, and the diameter at breast height (DBH) is recorded. Monumenting and tagging ensures that plots can be easily relocated and the same trees resampled to determine change over time. Dead trees are assessed for cause of death; and, each living whitebark pine tree is assessed for blister rust symptoms, canopy kill, bark-stripping, mountain pine beetle infestation, and assigned an overall health status. Blister rust cankers are described as active (sporulating, with new or old aecial spore sacs) or inactive, and recorded for both stem and branch. For branches or stems that appear to have a canker but show no aecial sacs, we recommend that three of five ancillary symptoms be present near the affected area before the canker is identified as definitely caused by blister rust: foliage flagging, rodent chewing/bark stripping, oozing sap, roughened bark, and swelling. Living whitebark pine seedlings <1.4 m in height are counted in the understory survey and further subdivided into two height classes. Active and inactive cankers are recorded for each single stem or clump.

We suggest a number of optional variables which are useful for specific research questions or for obtaining a more detailed picture of either the habitat type or dynamics of spread of blister rust. These variables include: counting stem and branch cankers, counting and identifying species of *Ribes* in the plot, detailed surveys of regeneration (focusing on seedlings and saplings and excluding suppressed trees), identifying other forest trees, and obtaining whitebark pine ages.

Plot data are easily entered into the Access database. Basic summary statistics, such as percentage of live trees with active blister rust stem cankers, are easily calculated for the plots using database queries. If these standardized methods for surveying and monitoring are widely adopted, collected data will provide critical information that can be used for geographic and even temporal comparisons. Such comparisons will provide the tools for managers to prepare regional and even range-wide strategies for whitebark pine restoration, making the most efficient use of resources.

WLIS: The Whitebark-Limber Pine Information System and What It Can Do for You.

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Whitebark and Limber Pine Information System (WLIS) is a database of basic plot information on whitebark and limber pines from the numerous surveys and studies that have been completed in the US and Canada. This compilation of summary data permits rangewide assessments of whitebark and limber pines. Forest Inventory and Analysis (FIA) plot data are part of the database. The data can be queried to provide a spatial summary of the condition of these two species. The US National Park Service has created an interface for simplifying entry of future survey/study data by individuals using the survey design developed by the Whitebark Pine Ecosystem Foundation as a template.

There are three main components associated with WLIS. The first is an interactive interface that allows for the easy entry and validation of data and review of data already in the database. Over 2,200 records have been gathered and entered into the database. Additional data can be entered into the user's own copy of the database. This can either be through direct entry via the interface or by collecting data utilizing the US National Park Service database (Pillmore and Frakes) and importing it directly into WLIS. The second component of WLIS is its query builder. This tool allows the user to easily construct queries of the data. Queries can be built for any of the variables included in the database, either individual elements or combined. The results of a query can be viewed through the mapping application, and also can be exported through the interface into a commercially available spreadsheet. The third component is the GIS mapping ability of the program. Selected plots can be mapped along with other spatial components. The survey plot locations can be viewed along with FIA inventory plot locations. This component of WLIS has limited GIS capabilities, but geospatial data can be exported and used in more advanced GIS software.

WLIS was tested by selected individuals prior to release. Following testing and revisions, it has been made available to those interested. The system is available as a CD from the authors or it can be downloaded from the web (<http://www.fs.fed.us/r1-r4/spf/fhp/prog/programs2.html>). Surveyors and researchers collecting data on whitebark and limber pines are encouraged to enter their data and make it available for inclusion in WLIS. Periodic requests for new data will be made using a mailing list created when users download WLIS or request a CD. Users will be notified as updates of the database are made available through the website.

Those interested in assessing the condition of whitebark and limber pines can use WLIS to more effectively plan surveys and identify priority areas to evaluate. Historical trends can also be assessed as multiple surveys are completed and entered.



Using Verbenone to Protect Whitebark Pine from Mountain Pine Beetle Attack

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Whitebark pine (*Pinus albicaulis* Engelm.) is currently declining in many areas due to a variety of factors including white pine blister rust, fire suppression, forest successional processes, and periodic outbreaks of mountain pine beetle (*Dendroctonus ponderosae* Hopkins).

In 2005 in northern Idaho and Montana, mountain pine beetle killed nearly 630,000 mature whitebark pine trees on about 142,000 acres. Many of these stands have also been impacted by white pine blister rust. Seed is collected from phenotypically blister rust-resistant “plus” trees which are now under threat of mountain pine beetle attack. Silvicultural methods such as thinning, used to manage mountain pine beetle in other hosts, may not be applicable in less-dense whitebark pine stands. Individual, high-value trees can be protected from beetle attack with topical treatments of insecticides applied to tree boles with high-pressure spray equipment. However, spraying is not practical in inaccessible areas or on a large scale. Using beetle pheromones to protect high-value trees from attack has been shown to be a viable management option.

We tested the efficacy of verbenone, an anti-aggregation pheromone for mountain pine beetle, in protecting individual whitebark pine trees from beetle attack for three years. EPA-registered 5-gram verbenone pouches, replaced at mid-season, were tested along with new thicker membrane (longer lasting) pouches, and larger (7.5-gram) pouches. All were compared to untreated controls.

In 2002, we tested verbenone on 150 whitebark pine in northern Idaho. Treatments were: two, 5-gram pouches per tree; four pouches per tree; and untreated controls. To ensure equal beetle pressure, each tree was baited with a mountain pine beetle attractant tree bait. Pouches were replaced mid-season due to their short elution period. After beetle flight, trees were rated as mass attack, strip attack, pitch out, or no attack. “Mass attack” means the tree was overcome by beetles and killed. “Strip attack” is a tree successfully attacked on a portion of its bole, but the tree is not killed. A “pitch out” is one unsuccessfully attacked. “No attack” is an unattacked tree.

All untreated trees were killed while over 90% of treated trees survived. There was no statistical difference between the use of two or four pouches per tree (figure 1).

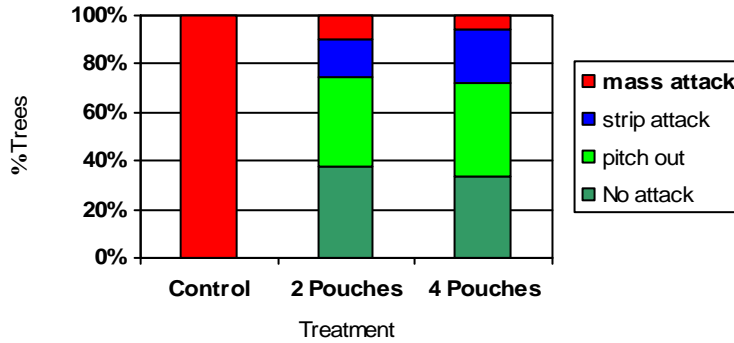


Figure 1. A test of whitebark pine trees treated with two or four verbenone pouches per tree in North Idaho in 2002.

In 2003, we treated 150 whitebark pine in western Montana. Treatments were: two standard 5-gram pouches per tree; two experimental longer-lasting 5-gram pouches per tree; and untreated controls. Only standard pouches were replaced mid-season. Attractant tree baits were placed 10-15 feet from each tree. Results were comparable to 2002. A total of 42% of control trees were killed while over 90% of the treated trees survived (figure 2). There was no significant difference between the two pouches.

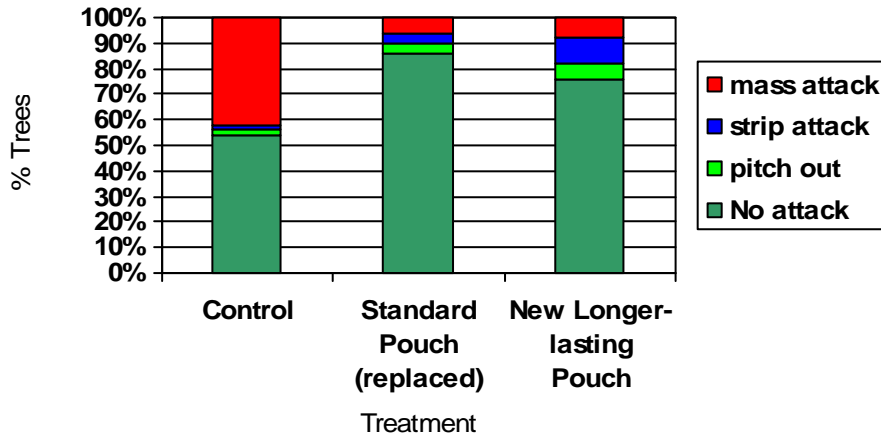


Figure 2. Whitebark pine test in 2003 in western Montana using two standard verbenone pouches that were replaced mid-season, and two experimental longer lasting pouches per tree.

In 2004, 100 whitebark pine were treated in northern Idaho. Treatments were two, experimental 7.5 gram pouches per tree and untreated controls. The pouches were not replaced mid-season. Attractant tree baits were placed 5-10 feet from each tree. Treated trees were protected from mass attack (figure 3). While 77% of control trees were killed, 87% of treated trees survived.

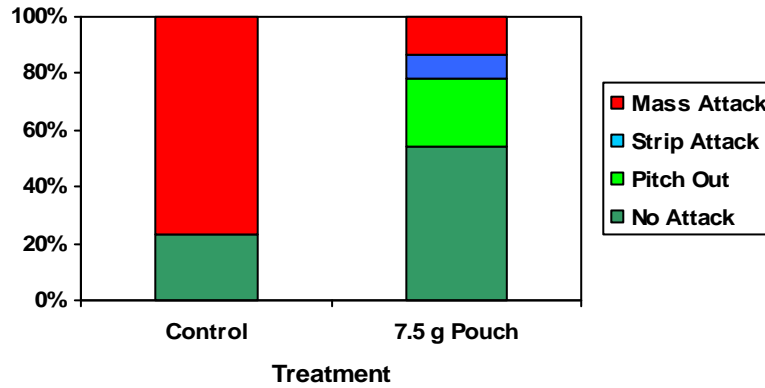


Figure 3. Whitebark pine test in North Idaho in 2004 using two 7.5 gram verbenone pouches per tree.

In summary, most individual whitebark pine trees, subjected to the presence of a mountain pine beetle attractant pheromone, were protected from mass beetle attacks. Treating individual whitebark pine trees with either two registered or two experimental pouches significantly reduced mountain pine beetle attacks when compared to untreated controls. These tests provide strong evidence that individual pine trees can be protected from mountain pine beetle attack using verbenone. Standard (5-gram) verbenone pouches are currently registered and available for use. Used operationally, they have successfully protected whitebark pine plus trees from mountain pine beetle attack, even in areas of high beetle populations.

Condition and Health of Whitebark Pine in the Blue Mountains of Northeastern Oregon

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Introduction

Whitebark pine is a keystone species, regulating snowmelt, preventing soil erosion, and providing an important food source for seed-eating birds and mammals. In the Greenhorn, Elkhorn, and Wallowa Mountains in northeastern Oregon, whitebark pine occurs in small, disjunct island populations at elevations above 2300 meters (Fig. 1). Forming both mixed and nearly pure stands, whitebark pine will often be succeeded by the more shade-tolerant subalpine fir if fire does not intervene.

From 2002-2005, field surveys were conducted to assess trends in the condition and health of whitebark pine populations, specifically: (1) the amount and severity of white pine blister rust within and among populations, (2) the

degree of mortality from whitebark pine blister rust and other causes across age classes, and (3) the amount of stand regeneration. Tree-ring data were also

obtained to assess demographic and recruitment patterns within and among species, and to evaluate the frequency and severity of growth suppression events.

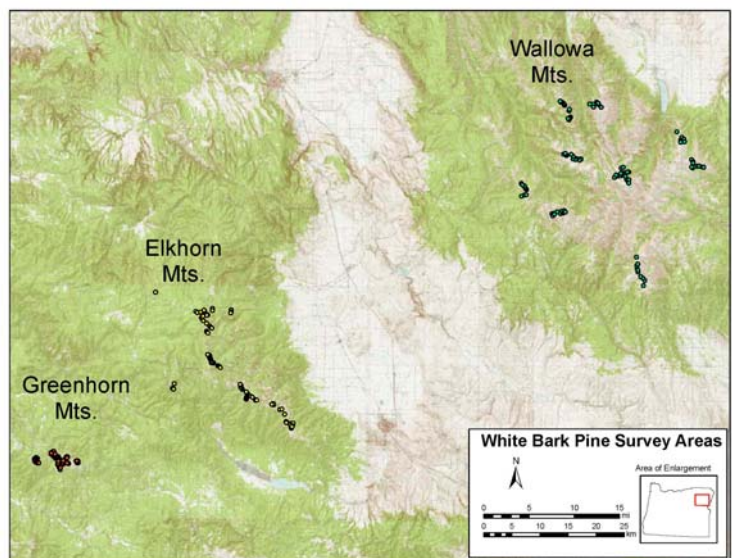


Fig. 1. Location of whitebark pine survey areas (Elkhorn, Wallowa, and Greenhorn Mts. in northeastern Oregon (U.S.A.).

Survey Methods:

In the Greenhorn Mountains, whitebark pine and associated conifer species were surveyed in 153 plots in 30 stands using modified Common Stand Exam (CSE) procedures. Circular 0.04 ha plots were established to sample all whitebark pine (alive and dead) greater than 10 cm DBH; 0.01 ha plots were used to sample seedlings and saplings. In the Elkhorn and Wallowa Mountains, 200 transects (46 m x 9 m) were established following survey methods developed by the Whitebark Pine Ecosystem Foundation. Only whitebark pine trees were assessed in the transects.

Within each plot or transect, increment core samples were taken from the first live standing whitebark pine tree per diameter group (n=8 classes). In the Greenhorn Mountain plots, core samples were obtained from associated species in addition to whitebark pine. Tree ring analysis was completed by J. King (Lone Pine Research, Bozeman, MT).

Survey Results

Of the 4464 whitebark pine surveyed in the three areas, a total of 595 (13%) were dead. Mortality was highest in the Greenhorn Mountains (24%), and lowest in the Elkhorn Mountains (8%) (Fig. 2). The principal cause of mortality, especially among the older trees, was attributed to a mountain pine beetle epidemic that occurred in northeastern Oregon during the 1970's (Fig. 3). White pine blister rust accounted for approximately 15-21% of the observed mortality (Fig. 3), most of which was in the younger age classes (data not shown).

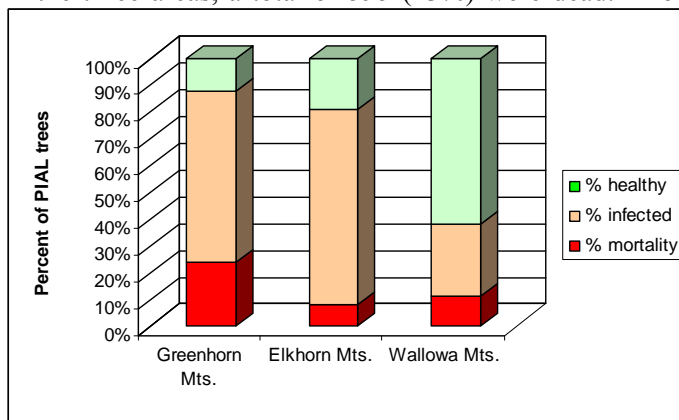
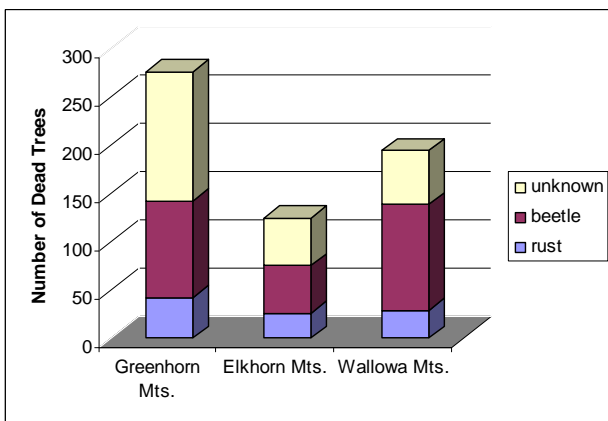


Fig. 2. Percentages of sampled whitebark pine in the Greenhorn Mts., Elkhorn Mts., and Wallowa Mts. that were healthy, infected with white pine blister rust, or dead

The prevalence of blister rust in live trees was extremely high in both the Elkhorn and Greenhorn



Mountains, where infection rates were 73% and 64%, respectively (Fig. 2). In contrast, only 27% of the trees in the Wallowa Mountains were infected. In all areas, the majority of the infected trees (>75%) had either bole cankers or branch infections located within 15 cm of the bole (blister rust severity rating = 3).

Fig. 3. Whitebark pine mortality in the Greenhorn Mts., Elkhorn Mts., and Wallowa Mts as attributed to mountain pine beetle, white pine blister rust, or unknown causes

In each study area, a number of whitebark pine exhibited pronounced growth suppressions in the period following ca. 1980 (e.g., Fig. 4 for Greenhorn Mts.). The cause is unknown, but not likely linked to climate because not all sampled whitebark pine had reduced growth during the same time period. Also, in the Greenhorn Mountains, where associated species were analyzed in addition to whitebark pine, growth suppressions were not entirely synchronous among species as would be expected if climate were a controlling factor (Fig. 4). The oldest living whitebark pine occurred in the Wallowa and Elkhorn Mountains, where a number of trees pre-dated the 1600's (Fig. 5a, b). The oldest (>500 years) and slowest growing (>100 years per inch of radius) living whitebark pine trees occurred in the Wallowa Mountains.

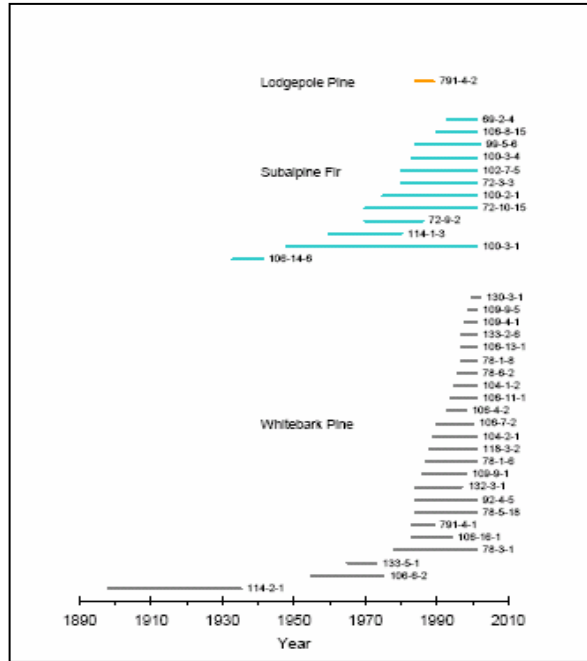


Fig. 4. Timing of tree-ring growth suppression in whitebark pine and associated species in the Greenhorn Mts. Each horizontal line represents a period of distinct growth suppression in an individual tree.

Tree ring analysis revealed several episodes of diminished whitebark pine recruitment and a recruitment peak ca. 1940-1960 (Fig. 5, 6, 7). There was a close coincidence between recruitment pulses and periods of past climate warming (J. King, pers. comm.). These results, combined with the highly synchronous recruitment pulses across the three sampling areas, suggest that regional climate is strongly controlling recruitment patterns.

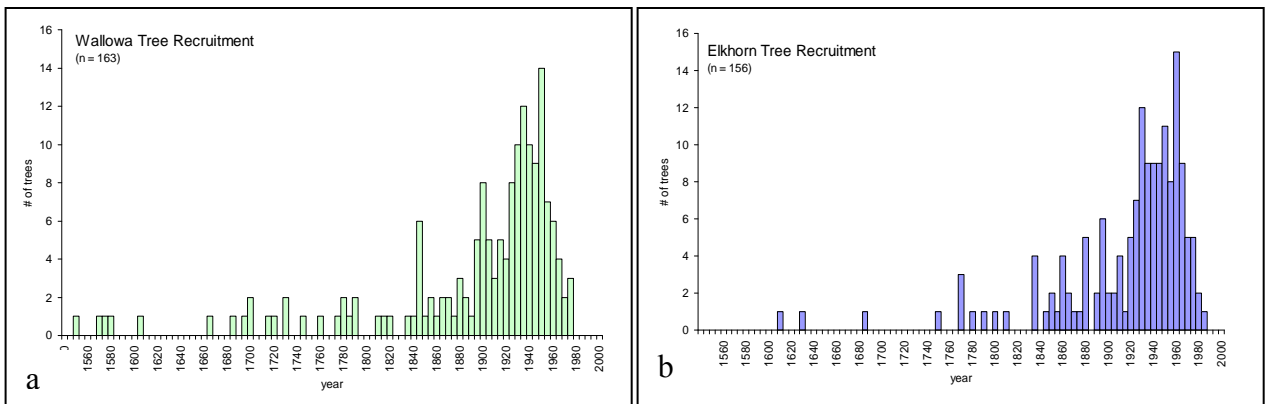


Fig. 5. Whitebark pine recruitment dates in the (a) Wallowa Mts., and (b) Elkhorn Mts.

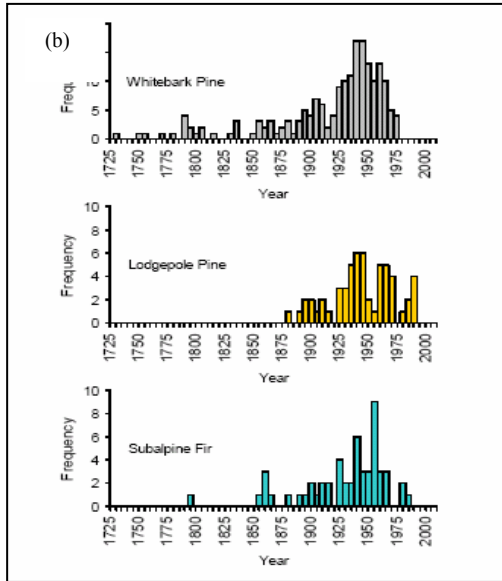


Fig. 6. Recruitment dates for whitebark pine and associated species in the Greenhorn Mts.

Discussion

Information from the 2002-2005 surveys provide reference conditions from which to assess changes in the status of whitebark pine in northeastern Oregon. While data analysis is still ongoing, our impression is that whitebark pine is seriously threatened by white pine blister rust, particularly in the Greenhorn and Elkhorn Mountains. Several measures have been taken to protect and conserve the unique populations occurring in these areas, including: (1) cone collections for blister rust resistance screening at Dorena Genetic Resource Center, (2) a regeneration trial and an outplanting project in the Greenhorn Mts., and (3) mechanical treatments in the Greenhorn and Elkhorn Mts. to reduce competition and to create openings for whitebark pine regeneration (see related abstract, this proceedings). Additional work will be accomplished as resources become available.

Acknowledgments: Funding for this work was provided by the USDA Forest Service, Forest Health Protection Program.

Whitebark Pine Restoration Treatments in the Vinegar Hill Scenic Area of Northeastern Oregon

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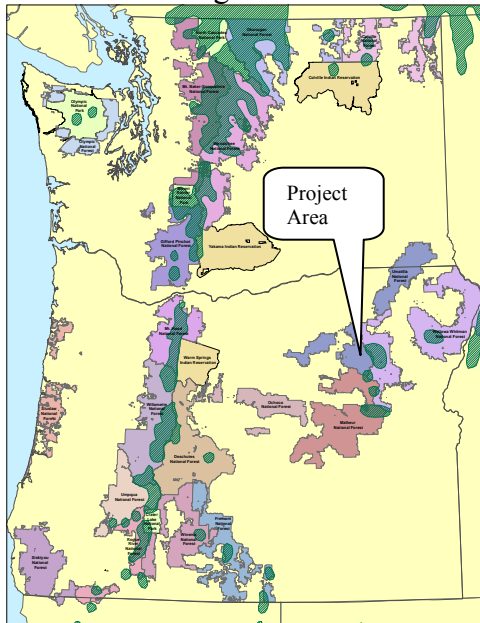
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Introduction

The Vinegar Hill Scenic Area is located in the south central portion of the Blue Mountains in northeastern Oregon. The Scenic Area contains one of the several isolated populations of



whitebark pine known to occur in northeastern Oregon, all of which are well outside the main distribution of the species (see Fig. 1).

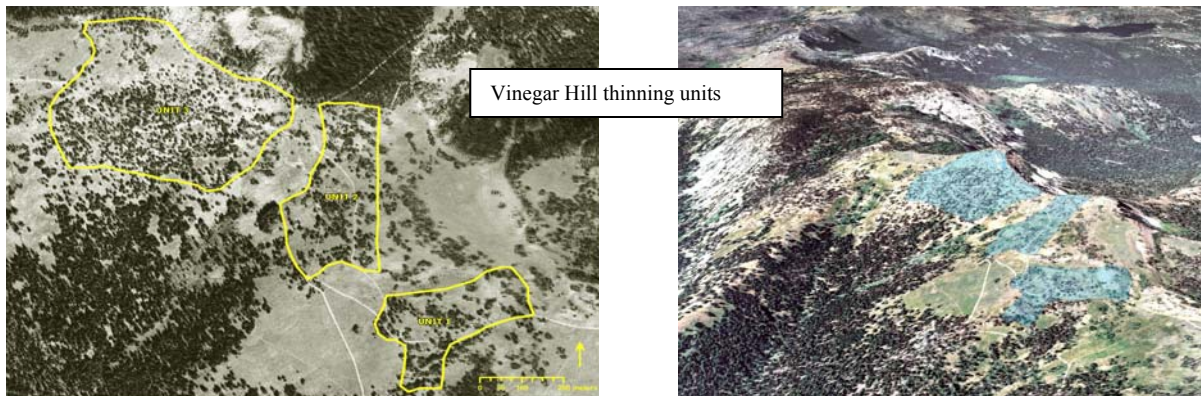
Within the high elevation (2200-2500 m), mixed-conifer forests and meadows of the Scenic Area, approximately 193 ha contain whitebark pine as a major component of the stand. Recent assessments of the condition and health of whitebark pine in these stands provide evidence that the Vinegar Hill population is at risk of local extinction due to mortality caused by white pine blister rust (*Cronartium ribicola*), subalpine fir encroachment and associated overstocking-induced stress, and mountain pine beetle (*Dendroctonus ponderosae*) (see abstract, Condition and Health of Whitebark Pine in the Blue Mountains, this proceedings).

Whitebark Pine Restoration

In 2003, a restoration project was implemented to reduce co-dominant and understory subalpine fir and other competing conifer species to stand density and basal area levels more conducive to whitebark pine growth and vigor (<10m²/ha). An additional objective of the project was to create small openings to encourage whitebark pine seed caching by Clark's nutcracker.



The project area contained 3 units totaling 33 ha of the most accessible stands where there was a predominance of whitebark pine. Within each unit, competing subalpine fir, lodgepole pine, and other conifer species were removed by felling or girdling. The work was completed under a service contract at a cost of \$1927-\$2322 per ha.



Project Specifications

- Live trees of competing species were cut or girdled within a 15 m radius of whitebark pine that were over 5 m in height. A 6 m radius was used if whitebark pine tree height was <5 m.
- Competing trees with a DBH of 3 cm or greater were girdled by 2 overlapping sawcuts below the lowest limb.
- If the tree to be removed was <3 cm DBH, it was cut below the lowest live limb, bucked into manageable pieces, and hand-piled at the edge of the stand for later burning (Fall 2004). Stumps were limited to a maximum height of 2.5 cm.
- Slash piles were restricted to no more than 2.5m x 2.5m x 2 m in size and were located at least 9 m from standing live trees or snags.



Girdled trees



Girdled trees



Burned slash piles

Future monitoring will determine the effectiveness of the thinning and girdling treatments on whitebark pine vigor, health, and regeneration.

Acknowledgments: Funding for this work was provided by the USDA Forest Service, Forest Health Protection Program.



Dead and dying girdled trees (red flagging)

Status of Whitebark Pine on National Forest Lands in Washington and Oregon

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In 2002 to 2004, surveys were conducted in whitebark pine stands on 9 National Forests in USFS Region 6 (Washington and Oregon). The primary objectives of these surveys were to locate and map whitebark pine stands, and to assess the incidence of the damaging agents white pine blister rust (*Cronartium ribicola*) and mountain pine beetle (*Dendroctonus ponderosae*). Overall, the sample included 10,500 individual trees. Mean blister rust incidence was 41.5% (range 0.0 – 100%); mean mountain pine beetle incidence was 4.5% (range 1.1-34.3%); and overall mean mortality from all causes was 33.4% (range 0.0-89.4%).

Impacts of Climate Change on Mountain Pine Beetle Habitat Suitability and Outbreak Risk in Whitebark Pine Forests of British Columbia

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Populations of mountain pine beetle are currently at epidemic levels in British Columbia and observations suggest that infestations are occurring in areas previously considered climatically unfavourable for brood development. Warming climates are expanding the northern limits of mountain pine beetle's range in British Columbia, and probably creating more suitable habitat for the establishment and persistence of beetle populations at higher elevations. This means that high-elevation whitebark pine forests may be more vulnerable to beetle outbreaks than ever before. Whitebark pine is already declining due to white pine blister rust infections and forest management activities, and the added pressure of mountain pine beetle outbreaks is likely to exacerbate its decline, adversely affecting many ecosystem processes and posing a threat to animals that rely on its seed for food.

Little is known about the status of mountain pine beetle populations in whitebark pine forests of British Columbia. The objective of this work was to assess the potential impacts of climate change on the suitability of beetle habitats in whitebark pine forests. Based on empirical relationships of the direct and indirect influences of climate on mountain pine beetle, landscape-wide projections of climatically suitable beetle habitat have been constructed from 1921 to 2070 using historical weather records and future conditions predicted by the CGCM1 global circulation model, which assumes a doubling of atmospheric CO₂ by 2100. We overlaid these projections onto the geographic range of whitebark pine in British Columbia and quantified changes in the distribution of suitable beetle habitat and risk (very low, low, moderate, severe, extreme) of whitebark pine forests to beetle outbreaks. We found that warming climates are generating substantial changes in the risk of whitebark pine forests to beetle outbreaks. From 1921 to 1950, most whitebark pine forests were at very low risk to beetle outbreaks. However, between 1921 and 2070, there were substantial increases in the amount (ha) of whitebark pine forests at moderate and severe risk to beetle outbreaks, with the amount of whitebark pine forest at severe risk doubling between 2001 and 2070.

Influence of Inoculum Source and Density on White Pine Blister Rust Infection and Mortality of Whitebark Pine: 2007 Update on 2001 Inoculations of Shoshone National Forest Seedlings

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Abstract

Artificial inoculation trials to evaluate genetic resistance of whitebark pine have only recently started. Inherent in such trials are questions of how results vary with inoculum source or inoculum density. Seedlings from a bulked collection of cones from trees on the Shoshone National Forest were inoculated in 2001. Treatments consisted of a factorial of two geographic sources of inoculum and three levels (densities) of inoculum. Early results showed high infection levels (needle spots and/or cankers) across all treatments within a year of inoculation. Through March 2006, the percentage of trees with stem symptoms for the six treatment combinations varied from 73.7 to 100 percent, while rust mortality varied from 60.4 to 83.8 percent. Infection and mortality levels were similar for the mid- and high inoculum density treatments. The surviving trees were re-inoculated in September 2005 and will be followed further to examine the level of resistance in this Shoshone population.

Introduction

White pine blister rust is caused by the non-native invasive pathogen *Cronartium ribicola*. All nine North American five-needle pine species (*Pinus* subsection *Strobus*) are susceptible to this disease, with high mortality occurring in natural stands as well as plantings of western white pine (*Pinus monticola*) and sugar pine (*P. lambertiana*). There is little published information about artificial screening of whitebark pine (*P. albicaulis*) seedlings for blister rust resistance. In this trial, we examine whether the amount of basidiospores (inoculum density) or the geographic source of inoculum influence the resistance of whitebark pine. The purpose of this study was to help refine an inoculation protocol for operational screening of whitebark pine at Dorena Genetic Resource Center. This report updates the early results presented in 2004 and focuses on stem infection and rust mortality.

Materials and Methods

Plant Material

Whitebark pine (WBP) seed from a bulked cone collection (CDA # 7425) from the Shoshone National Forest southwest of Dubois, Wyoming (~43°30'N 109°50'W, elevation ~2987 m) was sown in Ray Leach SC-10-Super Cells (16.4 cm³) in April 1999 at the Forest Service's Coeur d'Alene nursery and transported to Dorena Genetic Resource Center (DGRC) in July 2001 just before the September 2001 inoculation.

Inoculation

Treatments consisted of a factorial of 2 sources of inoculum (*Ribes* sp. leaves infected with *Cronartium ribicola* at the telial stage) and 3 targeted inoculum densities (Table 1). Seedlings were divided into six groups with approximately 48 seedlings per group; each group was allocated to one of the six treatment combinations (Table 1). The inoculation treatments were randomly assigned in the inoculation chamber with no replication and were separated to minimize cross-contamination. Inoculation followed standard DGRC procedure (Danchok and others 2004). The seedlings were transplanted into three standard DGRC boxes approximately 3 weeks after inoculation. Each treatment was randomly assigned to five rows in a completely randomized design, with 8-10 seedlings per treatment row.

Table 1. Treatments of whitebark pine inoculated in September 2001.

Ribes Source ^a	Inoculum Density (basidiospores/cm ²)			Spore Germ ^b (%)	Inoc. Time (h) ^c
	Level	Target	Actual (se)		
MA	Low	1000	825 (165.2)	95	15.5
MA	Medium	2500	2500 (248.3)	87	26.5
MA	High	5000	5150 (585.2)	99	40.5
SL	Low	1000	1000 (91.3)	99	11.0
SL	Medium	2500	2625 (342.5)	100	47.0
SL	High	5000	5400 (393.7)	87	37.3

^a *Ribes* sp. leaves collected from Silver Lake, Oregon (SL) and Mt. Adams, Washington (MA)

^b Basidiospore germination

^c Time (h) to reach target inoculum density

Assessments

Seedlings were assessed in June 2002 (~ 9 months after inoculation) for height (cm) and number of needle lesions. Number of needle lesions, number of stem infections, and survival were assessed in March 2003 (~ 18 months after inoculation) and July 2003 (~ 22 months after inoculation). Survival and presence of stem symptoms were assessed approximately 30, 34, 46, and 52 months after inoculation. Only percentage seedlings with stem symptoms, percentage survival with stem symptoms, and percentage rust mortality are reported here.

Analysis

Exploratory analyses of variance using SAS Proc GLM (SAS Inc. 2006) for the percentage seedlings with stem symptoms and percentage seedlings surviving were performed using untransformed plot means. Inoculum source information was lost for 3 of the high density plots; those plots were excluded from this summary.

Results

Stem Symptoms

At 52 months after inoculation percentage seedlings with stem symptoms (% SS) ranged from 73.7 to 100% for the Mt. Adams (MA) inoculum source treatments, and 87.8 to 95.8% for the Silver Lake (SL) treatments (Table 2). For the MA treatments there was an increase in % SS as inoculum density increased (Table 2, Figure 1). For the SL treatments both the

high and low density treatments had slightly higher % SS than the medium density treatment (Figure 1). The overall mean for SL was slightly greater than for MA (92.9 vs. 86.4% SS, respectively). In general most of the seedlings had developed stem symptoms by the 22-month assessment, regardless of treatment (Figure 1).

Table 2. Means by inoculum source and density

Inoculum Source ^a	Inoculum Density	# seedlings ^b	% infected ^c	% SS	% SSAL	% RMORT
MA	Low	46	100.0	73.7	17.0	60.4
MA	Medium	49	96.0	85.6	18.7	69.3
MA	High	28	100.0	100.0	14.4	81.9
SL	Low	47	100.0	95.8	19.8	76.2
SL	Medium	49	97.8	87.8	12.7	75.6
SL	High	37	100.0	95.0	11.8	83.8

^a Where MA = Mt Adams, WA; SL = Silver Lake, OR

^b Inoculum source information was lost for 3 plots inoculated at the high density

^c Percentage seedlings that developed needle lesions or stem symptoms (SS)

Four years after inoculation differences between inoculum sources were not significant for % SS; there were significant differences between inoculum sources at the 18-month assessment only. In contrast the main effect of inoculum density was significant until the 52-month assessment ($F=2.28$, $p=0.0670$).

There was a significant interaction between inoculum source and inoculum density for the last two assessments ($F=3.52$, $p=0.0481$; $F=3.67$, $p=0.0428$). This is likely driven by the high % SS in the SL low density (1000 basidiospores/cm²) treatment and differences in scale between inoculum sources.

The percentage seedlings surviving with stem symptoms at 52 months after inoculation (% SSAL) ranged from 11.8 to 19.8% among the treatments. For the MA treatments % SSAL was similar for the Low and Medium density treatments and slightly lower for the High density treatment (Table 2). However, for the SL treatments, % SSAL was higher for the Low density treatments and lower for the Medium and High density treatments (Table 2).

Mortality

Rust mortality (% RMORT) means ranged from 60.4 to 81.9% for the MA treatments and from 75.6 to 83.8% for the SL treatments at 52 months after inoculation (Table 2, Figure 2). For the MA treatments there was an increase in % RMORT as inoculum density increased (Table 2, Figure 2). % RMORT was relatively similar across inoculum densities for the SL treatments 46 and 52 months after inoculation (Table 2, Figure 2). The SL treatments tended to have slightly higher % RMORT (78.5 vs. 70.5%, respectively), but this difference was not statistically significant. There were significant differences among inoculum sources in % RMORT at the 30-month assessment only ($F=4.57$, $p=0.0445$).

The high density treatment had the highest % RMORT, regardless of inoculum source, at all assessments (Table 2, Figure 2). % RMORT was similar between the Low and Medium

density treatments within a given inoculum source. Although % RMORT is higher in the high density treatments, regardless of inoculum source, this difference was not statistically significant. There were significant differences among inoculum densities at the 30 and 34 month assessments only ($F=5.42, p=0.0127$; $F=6.37, p=0.0069$).

Figure 1. Treatment mean percentage seedlings with stem symptoms (% SS) by inoculum source

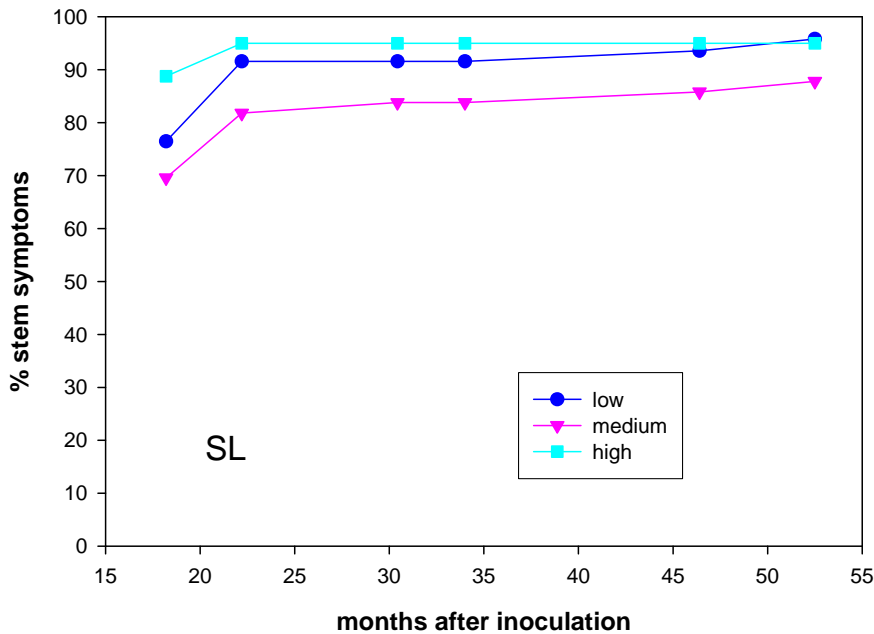
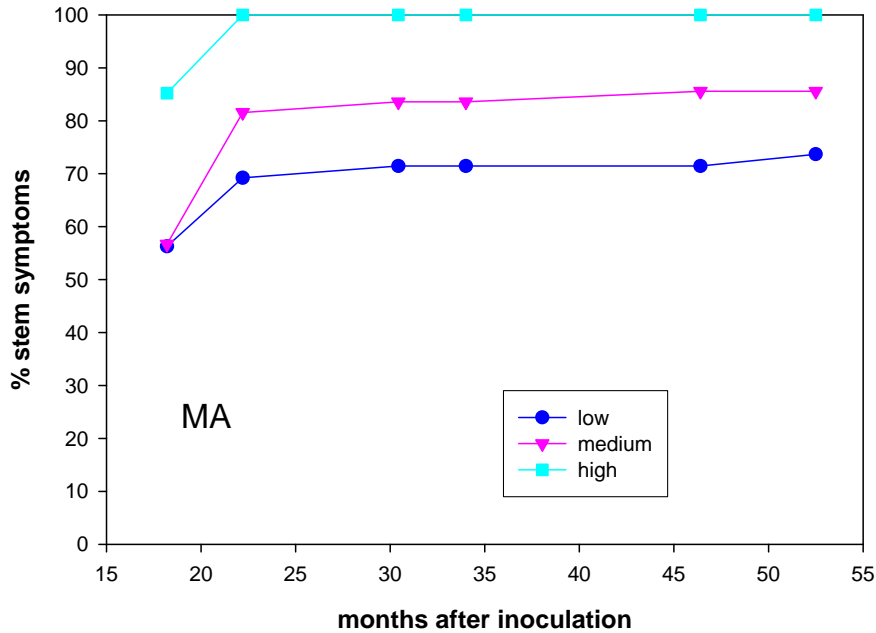
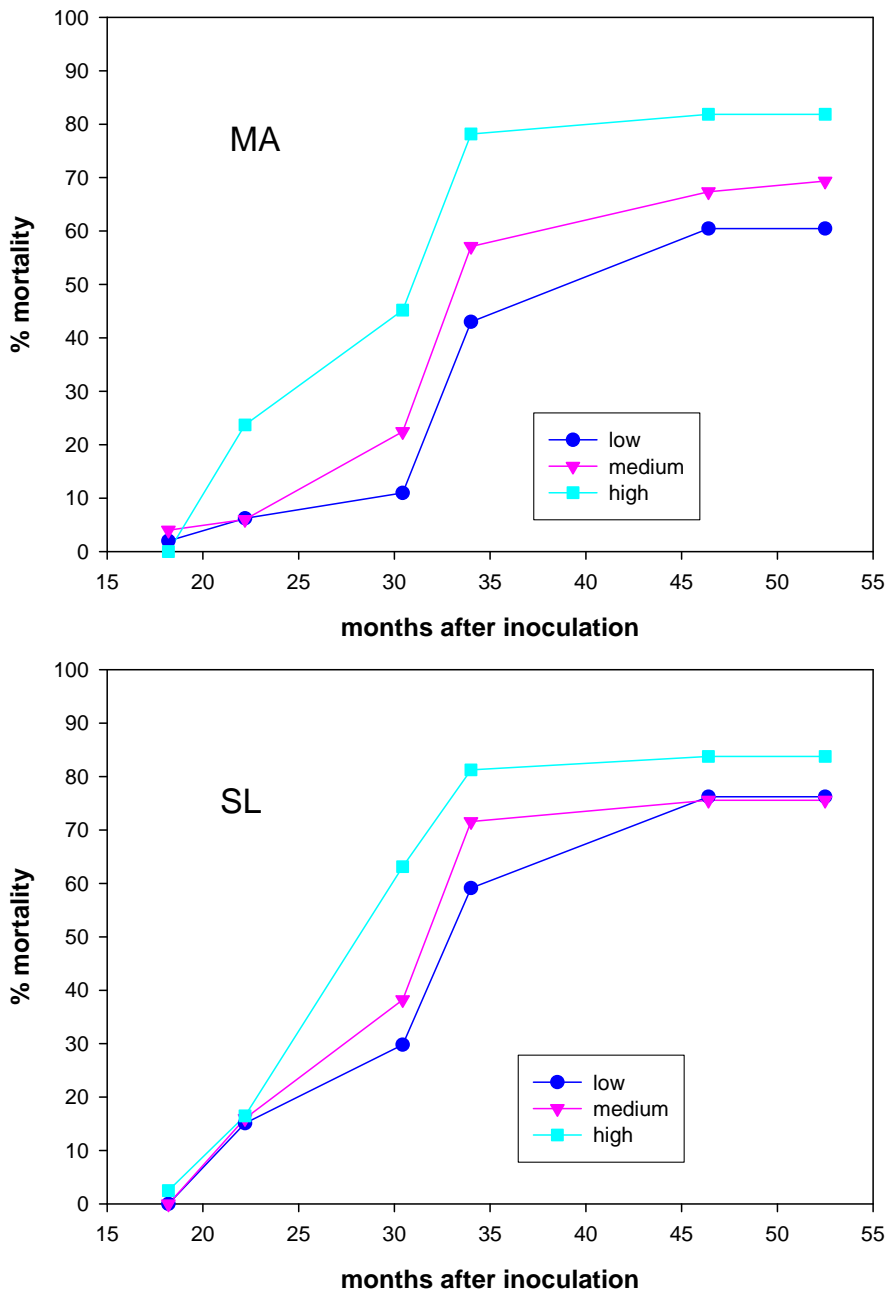


Figure 2. Treatment mean percentage rust mortality by inoculum source



Discussion and Summary

These results indicate that although there may be small but significant initial differences between geographic sources of rust or among inoculum densities, these effects are transitory for the traits % SS, % SSAL, and % RMORT. These results report only the percentage seedlings with stem symptoms or dead of blister rust. It should be noted that there were typically two to three times as many stem symptoms per infected tree in the high density

treatments relative to the low and medium density treatments, regardless of inoculum source (Kegley and others 2004). Although the final ratings may be similar, there may be differences in ontogeny of symptom development among inoculum sources (Kegley and others 2004). The effect of inoculum density was more persistent for % SS, only losing significance at the 52-month assessment.

Mortality only reached ~ 85% for the high density treatments through 52 months after inoculation; this is slightly unexpected given the high infection achieved in the test (>96%). Typically, open-pollinated progeny of phenotypic selections of western white pine and sugar pine exhibit >90% mortality in operational screening at DGRC (unpublished data). Thus, there may be at least a low level of resistance present in the Shoshone WBP population. In a subsequent DGRC trial, the Shoshone bulked seedlot showed relatively low levels of resistance and low survival through 2 years after inoculation (10 and 11.8% survival for DGRC-grown and Coeur d'Alene-grown seedlings, respectively) (Sniezko and others, this proceedings). However, the Shoshone bulked seedlot was ranked 59 of 108 seedlots for blister rust resistance in a test of WBP from the Inland Northwest (Mahalovich and others 2006).

Based on the data from this study, an inoculum density of 3000 basidiospores/cm² will ensure high levels of infection needed to test for resistance, while minimizing potential escapes and the possibility of overwhelming resistances that are manifested at moderate inoculum densities. Geographic source of inoculum appears to have little effect on the final outcome, indicated by rust mortality. This was a relatively small study, with a maximum of 48 seedlings per treatment combination, so the conclusions reached here are merely broad guidelines. Both geographic sources of rust were in the inoculation chamber at the same time, so there was the possibility of contamination among the treatments. However, the results presented here are similar to early results reported by Sniezko and others (this proceedings) for two separate trials; survival and % SS were similar for seedling families exposed to two different sources of inoculum. A trial examining the effect of inoculum density on individual seedling families has also been undertaken (unpublished data). In addition, the survivors of this prototype trial were reinoculated in 2005 and will be assessed for development of disease symptoms and mortality. This will help ascertain whether there were some escapes in the first inoculation.

Acknowledgments

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North American, Non-*Ribes* Alternate Hosts of *Cronartium Ribicola*: Ongoing Studies to Determine their Significance and Impact to Whitebark Pine

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To effectively manage white pine blister rust, caused by *Cronartium ribicola*, it is critical to understand the roles of local alternate hosts in the disease cycle and epidemiology in pine stands. Our ability to predict the progress of epidemics is questionable since the discovery of widespread species in the Orobanchaceae that can act as alternate hosts, i.e., *Pedicularis racemosa* and some *Castilleja* species (McDonald et al. 2006; Zambino et al. 2006). It is presently unknown how and when these hosts were acquired in North America and what influence they have on blister rust epidemiology. For these reasons, it is important to understand the life history of *C. ribicola* in eastern Asia—the pathogen’s putative center of origin—and the relationships among non-*Ribes* hosts in Asia and North America.

In Asia, utilization of alternate host species appears to vary considerably among regions. For example, Hyun and Koo (1981) reported that *C. ribicola* inoculum in Korea was able to infect *P. resupinata* but not a *Ribes* species (*Ribes hudsonianum*, a native of North America); however, native Korean *Ribes* species were not tested. In Japan, inoculation tests have shown rust sources vary in their abilities to infect *Ribes* and *Pedicularis* species. Inoculum from a *Pinus strobus* plantation in Japan infected only *Ribes* while inoculum from *Pinus pumila* at higher elevations infected *Pedicularis* or both genera (Yokota and Uozumi 1976). As *C. ribicola* adapts to diverse environments in North America, similar patterns of host utilization may appear, depending upon co-occurrence of white pines with different combinations of *Ribes*, *Pedicularis*, and *Castilleja* in different environments.

Recent studies have elucidated phylogenetic relationships among many species of *Pedicularis* and sister genera (Bennett and Mathews 2006, Ree 2006, Wolfe et al. 2005). Of relevance to white pine blister rust are the relationships among susceptible *Pedicularis* species in Asia and North America. Analysis of mitochondrial and nuclear DNA sequences showed a close phylogenetic relationship between *P. resupinata* and *P. yesoensis* (Ree 2006). These species are the major hosts in Korea and Japan, respectively. We performed a phylogenetic analysis of different *Pedicularis* species from Asia and North America using previously published DNA sequence (internal transcribed spacer of the nuclear ribosomal DNA) (Ree 2006 and Wolfe et al. 2005). The results indicate that North American *Pedicularis* species are well dispersed among several clades. *Pedicularis racemosa*, native to North America and susceptible to *C. ribicola*, is closely related to *P. yesoensis* and *P. resupinata*. However, in Asia, *C. ribicola* can also utilize species within other clades of

Pedicularis. For instance, *P. chamissonis* is a susceptible species in Japan that is found in a separate clade; it is currently undetermined if this species has close relatives in North America. Thus, *C. ribicola* may have a wider potential host range in North American *Pedicularis* and *Castilleja* than is currently recognized. Current plans are to address fundamental questions regarding utilization of non-*Ribes* hosts. Molecular genetic studies and inoculation experiments are underway to address genetic relationships among rust isolates from different alternate and primary hosts in Asia and North America, phylogenetic relationships among host species, and whether diverse rust sources can utilize *P. racemosa*.

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Radio-Tagging Clark's Nutcrackers: Preliminary Data from a Study of Habitat Use in Washington State.

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Whitebark pine (*Pinus albicaulis*) has coevolved with Clark's nutcracker (*Nucifraga columbiana*) (Lanner 1982, Tomback 1982) and nutcrackers provide the sole mechanism of primary seed dispersal for whitebark pine (Hutchins and Lanner 1982). Nutcrackers forage on the large seeds produced by whitebark pine in autumn and scatterhoard thousands of seeds in subterranean caches to be retrieved later in the year. Seeds that are not retrieved by nutcrackers are able to germinate (Tomback 1982). In response to a range-wide decline in whitebark pine, there is concern that populations of Clark's nutcracker are declining as well. A downward trend in nutcracker populations would have reciprocal effects for whitebark pine population regeneration, and consequently for the viability of subalpine ecosystems in western North America.

Efforts are underway to restore whitebark pine by sowing seeds and planting seedlings in established plots. Natural regeneration is required, however, because it is not possible for land managers to duplicate the unique population genetic structure, successional advantages, and the growth form characteristics enabled by nutcracker dispersal behaviors. To sustain whitebark pine communities, nutcracker populations must be maintained. This requires information on the habitats that nutcrackers use for critical behaviors. To date, research on Clark's nutcracker has been largely restricted to observational studies of harvesting and caching behavior (e.g. Vander Wall and Balda 1977, Tomback 1978, Hutchins and Lanner 1982, Balda and Kamil 1992). There has been no systematic study of habitat use by nutcrackers anywhere in their range. The current study will specifically investigate the following gaps:

- Nutcrackers are known to forage on and cache the seeds of multiple species of conifer in autumn (Giuntoli and Mewaldt 1978, Tomback 1978); yet the relative importance of habitats containing these conifers is unknown (Tomback 1998).
- Nutcrackers store seeds anywhere between 0.001 and 22 km from harvest trees and in a breadth of landscape features, microhabitats, and microsites (Vander Wall and Balda 1977, Tomback 1978, Hutchins and Lanner 1982, Dimmick 1993); a quantitative assessment of variation in cache site selection has not been possible because of limitations in study methodologies.
- Information is lacking on home range and habitat use by nutcrackers during the spring breeding season; in the literature, only 4 nests have been monitored (Mewaldt 1948,

1956), and there is little information on the habitats used by breeding birds (Tomback 1998).

Overall, the proportional use of different habitats by nutcrackers, particularly for critical behaviors such as foraging, seed caching, and reproduction, is speculative because of a paucity of quantitative data.

The objectives of the current study are to quantitatively measure home range size and habitat use, and to determine habitats that are critical for foraging, caching, and breeding in Clark's nutcracker. These objectives can most effectively be addressed using radio telemetry. Throughout this study, nutcrackers will be trapped in mist-nets and noose-carpet traps. All trapped nutcrackers will be banded, aged, and weighed. Adults will be fitted with a 3.9 g (3% of body weight) transmitter (model #A1080, Advanced Telemetry Systems, Isanti, MN) that is secured to the back with a harness. The transmitter battery is expected to last 415 days. Sites for trapping are located on the Okanogan-Wenatchee National Forest in the central Washington Cascade Range. The boundaries of the study area are not fixed, however, and will be determined by the radio-tagged nutcrackers as they move over the landscape.

Individual home ranges will be delineated using fixed kernels estimates (Worton 1989). A Geographic Information System (GIS) will be used to determine habitat attributes at each point of use. Behavior observations at each point (Marzluff et al. 2001, 2004) will enable a quantitative and probabilistic measure of differential habitat use within home ranges and will provide a quantitative means of correlating habitat use with critical behaviors. Variability in cache site selection will be measured by following radio-tagged nutcrackers between harvest stands and cache sites. The locations of harvest trees and cache sites will be marked on GPS units. Discrete choice models (Cooper and Millspaugh 1999, 2001) will be used to calculate the probability that each cache site was selected as a function of specific habitat attributes: dominant cover type, elevation, aspect, slope, proximity to nurse shrubs, soil type, and linear distance to both the harvest tree and spring nest site.

Preliminary data from five months of data collection indicate that there may be considerable variation within populations in summer and autumn home range size. Average summer home range size for four individuals was estimated to be 20.7 km² (range 7.7–38.8 km²). Home range size may be affected by habitat composition. For two individuals with home ranges containing mature whitebark pine, home ranges averaged approximately 10.3 km². Home range size for two individuals without whitebark pine within their home range averaged 31.1 km². While three harvested pine seed up to 35 km from their home range, caching was only known to occur within the two individuals respective home ranges. Detailed observations were made on the harvesting and caching behaviors of two radio-tagged nutcrackers. One-way flights of up to 30.6 km were made by one radio-tagged nutcracker while traveling between harvest stands of whitebark and ponderosa (*P. ponderosa*) pine and cache sites. The two radio-tagged nutcrackers were observed caching in three landscape features; within closed canopy forests, on steep, bare slopes (70-90°), and within forest openings (5-10 ha). For both individuals, the majority of caches were placed within closed canopy forests, followed by steep slopes, and lastly in forest openings.

This study is expected to last for three years and final analysis of data will occur in 2009. Fifty nutcrackers will be radio-tagged over the course of this study. Behavior observations on these additional individuals will enable more accurate assessments of home range size and habitat use.

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An Assessment of Genetic Variation of Whitebark Pine Populations from Oregon and Washington in Relation to Height Increment, Phenology, and Form.

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A complementary study to a planned rust evaluation experiment at the Dorena Genetic Resource Center was undertaken in order to evaluate genetic variation of height growth increment. The study sample included a total of 94 families from 6 provenances located in Oregon and Washington. A set of serial measurements were taken throughout the growing season in order to quantify the phenology patterns, relative growth rates, and total growth increment during the third growing season of the planned experiment. In addition, stem form was assessed. A few preliminary findings from the study are presented.

Eight cumulative height measurements were taken during the course of the growing season in order to assess the relative rates of shoot elongation, phenology, and growing season increment; where time of measurements were designated as days from 1 January, 2006. Least square means for all measured height increments (at days 91, 107, 122, 136, 150, 165, 178 and 201) differed significantly ($P < .05$) among provenances. The cumulative height increment changed rank among provenances until the third periodic measure (day 122 measure), wherein after the provenances general ranking remained the same throughout the rest of the growing season. The more northwestern provenances (Mt. Hood, Mt. Rainier, Warm Springs) formed a general group with the maximum height. The Umatilla provenance was intermediate in height followed by the Malheur and Crater Lake provenance grouping.

Significant genetic (family) variability was also detected at each measurement period for height. Family heritabilities were moderate and ranged from 0.40 – 0.65 for the respective measurement dates. The relationship (Pearson correlation coefficient) between family mean cumulative height increment (day 201 measure) and parent tree source environmental variables of elevation, longitude, and latitude were assessed. Moderate correlations existed between final height and elevation (- 0.46, lesser height with higher elevation), longitude (0.43, greater height with a more western source) and latitude (0.41, greater height with a more northern source).

In addition to height measurements, the percentage of the seasonal growth completed at each measurement date was computed. This variable provides a viewpoint relative to initiation of shoot elongation and/or relative growth rate (in early season growth) and to growth cessation (in late season growth).

Least square means for the derived percent of growth completion differed significantly among provenances for the first two measurement dates in the early season (day 91, 107), and for the last two measurement dates in the late season (day 165, 178). Day 201 was considered to represent 100% of growth completion for all seedlings, and thus there is no comparison among provenances for that measurement date. In the initial two measurement periods, the Warm Springs and Mt. Hood provenances exhibited the lowest percentage of growth completion values. The Malheur provenance exhibited the highest or second highest percentage of growth completion from the second measurement (day 107) on to the end of the growing season. There existed two major sub-sets of provenances at the latter part of the late season measurements (day 150, 165, 178) that were closely aligned. The Malheur and Umatilla provenances exhibited the highest percentage of growth completion, while Mt. Rainier, Mt. Hood, Warm Springs, and Crater Lake exhibited the lesser percentage of growth completion overall. The differences among provenances in both the early and late season would seem to indicate potential differences in either initial shoot elongation, cessation of growth, relative growth rate, or some variable combinations of these attributes.

Significant genetic (family) variability was also detected in all but the last (day 178) measurement for the percent of growth completion. Family heritabilities for this derived trait were low to moderate (range from 0.28 – 0.53) for the first six measurement dates.

The general form of the trees was scored in a subjective manner, where trees were scored as being in one of three form classes:

1. single stem form
2. forked form
3. numerous multi-stems/bush-like form.

Least square means for the transformed class variable differed significantly among provenances. The rank order of the more desirable tree form class to the less desirable tree form class was closely aligned with the rank order (from tallest to shortest) of the final cumulative heights per respective provenance. The northwestern provenances (Mt. Rainier, Mt. Hood, Warm Springs) expressed a more desirable mean tree form class than that of the remaining provenances.

Significant genetic (family) variability was also detected for form class, where the estimated family heritability was 0.67. The relationship (Pearson correlation coefficient) between family mean form class and final cumulative height was -0.63 (greater mean height associated with a more desirable form class). The correlation between family mean form class and parent tree source environmental variables of elevation, longitude, and latitude were also assessed. Low to moderate correlations existed between form class and elevation (0.33, less desirable form with higher elevation), longitude (-0.32 , less desirable form with a more eastern source) and latitude (-0.42 , less desirable form with a more southern source).

The preliminary findings from this study will be posted on the Dorena webpage: <http://www.fs.fed.us/r6/Dorena>. Further detailed analyses will be conducted on this study data, and all results will be posted on the webpage in Spring, 2007.

