

THE USE OF NATIVE ECTOMYCORRHIZAL FUNGI IN THE RESTORATION OF
WHITEBARK PINE

by

Erin Rebecca Lonergan

A thesis submitted in partial fulfillment
of the requirements for the degree

of

Master of Science

in

Plant Sciences

MONTANA STATE UNIVERSITY
Bozeman, Montana

November 2012

©COPYRIGHT

by

Erin Rebecca Lonergan

2012

All Rights Reserved

APPROVAL

of a thesis submitted by

Erin Rebecca Lonergan

This thesis has been read by each member of the thesis committee and has been found to be satisfactory regarding content, English usage, format, citation, bibliographic style, and consistency and is ready for submission to The Graduate School.

Dr. Cathy L. Cripps

Approved for the Department of Plant Sciences and Plant Pathology

Dr. John Sherwood

Approved for The Graduate School

Dr. Ronald W. Larsen

STATEMENT OF PERMISSION TO USE

In presenting this thesis in partial fulfillment of the requirements for a master's degree at Montana State University, I agree that the Library shall make it available to borrowers under rules of the Library.

If I have indicated my intention to copyright this thesis by including a copyright notice page, copying is allowable only for scholarly purposes, consistent with "fair use" as prescribed in the U.S. Copyright Law. Requests for permission for extended quotation from or reproduction of this thesis in whole or in parts may be granted only by the copyright holder.

Erin Rebecca Lonergan

November 2012

ACKNOWLEDGEMENTS

I would like to thank the many people without whom this project may never have been possible. Foremost, I would like to thank my graduate advisor and mentor Dr. Cathy Cripps for her constant support and encouragement, endless editing, fungal expertise, and the opportunity to continue developing as a plant scientist. Extreme gratitude goes to my committee members, Cathy Zabinski for statistical and editing assistance and Tracy Dougher for fertilizer and editing assistance. Many thanks go to Megan Higgs for all her statistical advice. This research would not have been possible without funding from Cyndi Smith with Parks Canada and John Schwandt with the USDA Forest Health Protection. I also thank Cyndi for the wonderful opportunity to work in Waterton-Lakes National Park and John for letting me tag along during some of his field work. I would like to extend my gratitude to the Couer d'Alene Forest Service nursery for providing seedlings used in the greenhouse study and to the Glacier National Park Revegetation crew and all the volunteers who helped plant the seedlings at the Summit Lake study site. Many thanks go to the Whitebark Pine Ecosystem Foundation for their hard work and dedication to restoring whitebark pine forests. I would like to thank Ed Barge for collecting suilloids and for his constant patience in lab. I would like to acknowledge Rosemary Keating and John Mason for volunteering to help collect field data. I owe a debt of gratitude to Don Bachman for his logistical advice and for always volunteering to drive. I would like to thank Elliot Johnson for his realistic advice and for always believing in me. Lastly, I would like to thank my family for years of love, support, and patience and of course my wonderful mutt Poncho for continually inspiring me.

TABLE OF CONTENTS

1. LITERATURE REVIEW	1
Whitebark Pine.....	1
Threats.....	3
Restoration	6
WBP Seed Zones	6
Rust Resistance Screening in WBP Seedlings.....	6
Growing WBP Seedlings	8
Fire and Silviculture.....	10
Monitoring WBP Seedling Survival	12
Waterton Lakes-Glacier International Peace Park WBP Restoration.....	17
Mycorrhizae	19
Mycorrhizae and Whitebark Pine	23
Field Assessment and Monitoring of ECM fungi Associated with WBP	25
Research Objectives.....	28
References.....	30
2. LOW NITROGEN FERTILIZER AS A STRATEGY TO MAINTAIN ECTOMYCORRHIZAL COLONIZATION OF INOCULATED WHITEBARK PINE SEEDLINGS BEFORE OUT-PLANTING	42
Introduction.....	42
Ectomycorrhizal Fungi and Restoration	43
Ectomycorrhizal Fungi and Whitebark Pine.....	45
Research Objectives.....	49
Materials and Methods.....	50
Seedlings	50
Spore Slurry Types	51
Fertilizer Regimes.....	52
Inoculation Methods	53
Assessment of Mycorrhizal Colonization.....	53
Experimental Design.....	56
Statistical Analysis.....	57
Results.....	57
Frequency of Ectomycorrhizal Colonization: Assessments 1, 2 and 3	57
Abundance of Ectomycorrhizal Colonization: Assessment 3.....	58

TABLE OF CONTENTS CONTINUED

Effects of Inoculation on WBP Seedling Parameters	64
Discussion	68
Fertilization	68
Container Length	71
Slurry Type	72
Inoculation Method	74
Seedling Parameters	74
Conclusions	75
References	77
3. EFFECT OF SITE CONDITIONS, MICROSITE, AND INOCULATION WITH NATIVE ECTOMYCORRHIZAL FUNGI ON THE EARLY SURVIVAL OF OUT-PLANTED WHITEBARK PINE SEEDLINGS	87
Introduction	87
Whitebark Pine	87
Restoration	89
Restoration in the International Peace Park	92
Ectomycorrhizae	95
Research Objectives	98
Materials and Methods	99
Study Site	99
Seedlings	102
Spore Slurry and Seedling Inoculation	103
Plot Layout and Design	104
Monitoring Seedlings	109
Statistical Analysis of Seedling Survival and Height	110
Results	113
Overall Seedling Survival	113
Seedling Survival by Site Condition	115
Seedling Survival by Microsite	116
Survival of Individual Seedlings by Mycorrhizal Treatment	117
Un-burned Areas	119
Burned Areas	119
Evaluation of Seedling Survival by Cluster Inoculation Treatment	120
Seedling Parameters	122
Discussion	127
Overall Seedling Survival	127
Survival by Site Conditions	129

TABLE OF CONTENTS CONTINUED

Survival by Microsite.....	136
Ectomycorrhizal Inoculation and Individual Seedling Survival.....	137
Ectomycorrhizal Inoculation and Cluster Survival.....	141
Summary of Conclusions.....	144
References.....	148
LITERATURE CITED.....	162
APPENDIX A: Field Study Plot and Seedling Data	186

LIST OF TABLES

Table	Page
2.1. Number of Whitebark Pine Seedlings in Each Treatment	56
2.2. Results from ANOVA.....	59
2.3. Average Mean and Standard Error for Ectomycorrhizal Abundance	60
2.4. Results from Post Hoc Tukey HSD Test for Fertilizer Effect	61
2.5. Average Mean and Standard Deviation for Root Volume (ml).....	65
2.6. Average Mean and Standard Deviation for Wet Biomass (g)	65
3.1. Number of Whitebark Pine Seedlings Planted in Each Site Condition.....	111
3.2. Number of Whitebark Pine Clusters Planted in Each Site Condition.....	111
3.3. Survival Odds for Individual WBP Seedlings as a Function of Site Condition and Mycorrhizal Treatment	118
3.4. Survival Odds for WBP Clusters as a Function of Site Condition and Cluster Inoculation Treatment	121
3.5. Average Height (cm) of Whitebark Pine Seedlings	124
3.6. Pairwise Comparison of Average Height	125

LIST OF FIGURES

Figure	Page
1.1 Whitebark Pine Range Map	1
2.1 Long and Short Containers, Dried and Fresh Sporocarps, Drip and Inject Inoculation Methods	52
2.2 Timeline of Inoculation and Assessments	54
2.3 Example of Suilloid Fungi Colonizing a WBP Seedling.....	55
2.4 Frequency of ECM Colonization Over Time.....	58
2.5 Mean Percent ECM Colonization as a Function of Fertilizer Regime	61
2.6 Mean Percent ECM Colonization as a Function of Container Length	62
2.7 Mean Percent ECM Colonization as a Function of Fertilizer Treatment, Container Length and Slurry Type	63
2.8 Mean Root Volume and Wet Biomass as a Function of Fertilizer Regime.....	66
2.9 Mean Root Volume and Wet Biomass as a Function of Container Length	66
2.10 Mean Root Development, Foliar Development and Needle Color.....	67
3.1 Location of Study Site	101
3.2 Study Site Snow Data	102
3.3 Plot Locations	105
3.4 Prescribed Burning at Summit Lake Study Site	106
3.5 Diagram of a Plot.....	107
3.6 Photos of Each Site Condition	108
3.7 Average Overall Survival for 2011 and 2012	113

LIST OF FIGURES CONTINUED

Figure	Page
3.8 Seedling Survival by Plot.....	114
3.9 Average Seedling Survival in 2011 and 2012 by Site Condition	115
3.10 Average Overall Survival by Burn Treatment	116
3.11 Average Survival by Site Condition and Microsite	117
3.12 Seedling Survival by Site Condition and Mycorrhizal Treatment of Individual Seedlings	118
3.13 Seedling Survival by Site Condition and Mycorrhizal Treatment of Whole Clusters.....	121
3.14 Seedling Height by Site Condition and Microsite	123
3.15 Seedling Height by Site Condition and Mycorrhizal Treatment of Individual Seedlings	124
3.16 Seedling Health by Site Condition and Mycorrhizal Treatment of Individual Seedlings	126
3.17 Seedling Health by Site Condition and Microsite.....	127

ABSTRACT

Whitebark pine (*Pinus albicaulis*) is an endangered keystone species in western North America. Populations are being decimated by white pine blister rust, mountain pine beetles and fire suppression. Large restoration efforts that include the planting of 200,000 rust-resistant seedlings are ongoing, but survival rates are low. Conifers are routinely inoculated with ectomycorrhizal fungi in the greenhouse to enhance out-planting success, but this has not been tried with whitebark pine. The goal of this project is to examine the use of native ectomycorrhizal fungi in restoration of whitebark pine with a greenhouse and field study. A main goal of the greenhouse study was to determine if low nitrogen fertilizer is conducive to ectomycorrhizal colonization by the native fungus, *Suillus sibiricus*. The effects of dried/fresh inoculum, short or long containers, and the drip/injection method were also tested. Results showed that mycorrhizal colonization was maintained with a low nitrogen fertilizer (4-25-15 NPK), although colonization declined at higher levels. Long containers were more conducive to mycorrhizal colonization, but differences were minimal for other variables. The field study conducted at Summit Lake in Waterton Lakes National Park is part of an effort to combat seriously declining pine populations. One thousand seedlings, half inoculated with *Suillus sibiricus*, were planted in clusters of three in four site condition combinations: burned/unburned areas, with and without beargrass. Survival was higher than for other studies one (95%) and two years (69%) after planting (2010). Results could be due to favorable spring conditions, conducive site conditions (terra-torching), and mycorrhizal inoculation on certain sites. Survival was 24% higher on burns in comparison to unburned sites; microsite increased survival across all sites. Effects of mycorrhizal inoculation were site dependent and survival was increased 17-23% on unburned sites with beargrass; inoculation did not impact seedling survival on burns. Survival was lowest (38%) on poor planting sites (unburned, no beargrass) but these sites benefited greatly from microsite presence. Data suggest site conditions strongly influence early seedling survival and that mycorrhizal inoculation may be beneficial when soil fungi are restricted. Longer term monitoring is necessary to determine how variables affect seedling survival in the future.

CHAPTER 1

LITERATURE REVIEW

Whitebark Pine

Whitebark Pine (*Pinus albicaulis* Engelm.) is a five-needle pine that tends to dominate in upper subalpine and timberline plant communities (Fryer 2002, McCaughey and Schmidt 2001) in both the Pacific Northwest and the Northern Rocky Mountains (Arno and Hoff 1989, Figure 1). It is a long-lived species, often surviving for more than 600 years (Luckman et al. 1984) with the oldest tree recorded at over 1,270 years old when it was discovered in the Sawtooth – Salmon River range, Idaho in 1993 (Perkins and Swetnam 1996).

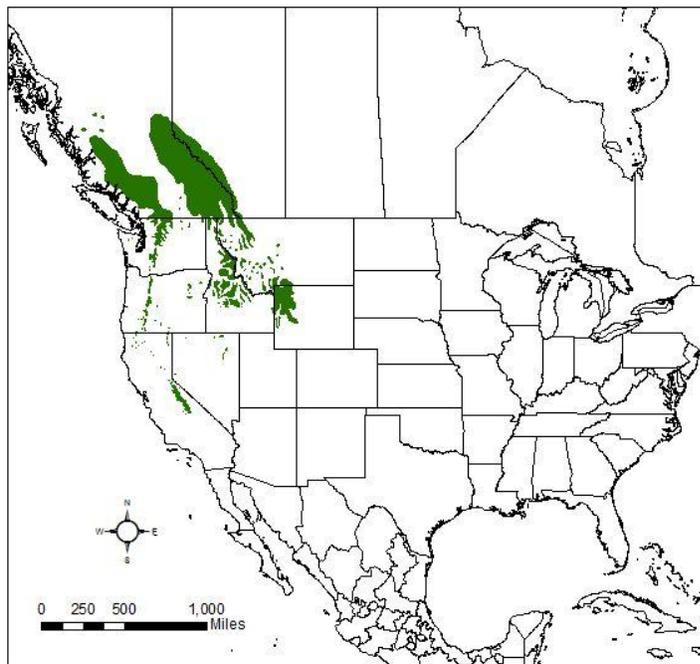


Figure 1. Range wide distribution map for whitebark (*Pinus albicaulis*) generated using data from Little (1971).

Whitebark pine is in the family *Pinaceae*, genus *Pinus*, subgenus *Strobus*, and subsection *Cembrae* (Price et al. 1998). It is a member of the stone pine group which is unique in possessing indehiscent cones that do not open to release seeds; instead they rely on birds for seed dispersal (Lanner 1996). Whitebark pine has co-evolved with Clark's nutcracker and relies heavily on birds for natural regeneration (Tomback 1982, Tomback 2001b). The Clark's nutcracker will pry seeds from whitebark pine cones and temporarily store the seeds until an appropriate place is found to cache the seeds for later consumption (Tomback 1982, Lanner 1996). The Clark's nutcracker will often cache seeds on steep, windswept slopes and in open, burned areas (McCaughey 1990, Tomback 2001b) and on average will bury 3-5 seeds per cache (Tomback 2001b). Seeds not retrieved by the nutcracker germinate in clusters from the cache (McCaughey 1990, Tomback 2001b) and eventually unite to form a composite whitebark pine tree. Many other animals in addition to Clark's nutcracker depend on whitebark pine as a valuable food resource. Their seeds are high in fat and energy and are consumed by numerous animals including endangered grizzly bears, black bears, and small mammals such as the red squirrel (Arno 1986, Mattson et al. 1992).

The upper subalpine is a harsh environment; however a unique dispersal strategy and large meaty seeds allow whitebark pine to be a primary colonizer of these extreme environments (Tomback et al. 2001a). Establishment of whitebark pine in open, disturbed sites often facilitates colonization of other subalpine species, as the pines provide protection from the elements and enhance humidity (Tomback et al. 2001a). Near the timberline portion of its range, where climatic conditions are more severe, whitebark pine

will form stable climax communities and may assume a twisted, dwarfed form called krummholz (Arno 2001). In these timberline areas, whitebark pine plays a pivotal role in watershed dynamics by retaining snowdrifts and delaying snow melt into the mid-summer months (Farnes 1990). The penetrating root systems of whitebark pine coupled with its ability to delay snow melt help stabilize steep mountain slopes and decrease erosion of poorly developed topsoil in subalpine zones (Farnes 1990). Whitebark pine is an integral member of high elevation communities and is considered a keystone and foundation species for its roles in supporting biological diversity and providing environmental stability (Ellison et al. 2005, Lantz 2010).

Threats

Whitebark pine populations are seriously declining and have been reduced by 90% or more in certain areas of its range (Kendall and Keane 2001, Schwandt 2006). The concerted effects of white pine blister rust, mountain pine beetle, and enhanced competition aided by fire exclusion all contribute to this rapid decline (Tomback et al. 2001a, Bockino 2008). The synergistic interactions of these threats pose serious challenges to maintaining viable populations of whitebark pine (Tomback and Achuff 2010).

White pine blister rust (*Cronartium ribicola* J.C. Fisch.), a fungal pathogen native to Eurasia and introduced around 1910, has now spread through most western forests of North America (McDonald and Hoff 2001, Geils et al. 2010). Whitebark pine is highly susceptible to white pine blister rust with the highest infection and mortality levels occurring in the Pacific Northwest and the Rocky Mountains of southern Canada,

Northern Idaho and Montana with an average infection level of 70% (Kendall and Keane 2001, Smith et al. 2008). Damage from white pine blister rust can include branch dieback and diminished reproductive success and mortality (McDonald and Hoff 2001, Tomback and Achuff 2010). Branch dieback usually results in reduced cone production which has been observed to cause a decline in the occurrence of Clark's nutcracker and the subsequent reduction of seed dispersal services they provide, thus hindering the natural reproduction of whitebark pine (McKinney and Tomback 2007, McKinney et al. 2009).

The mountain pine beetle (*Dendroctonus ponderosae* Hopkins) is a native inhabitant of western coniferous forests, however the colder climates generally associated with whitebark pine ecosystems have historically been too extreme for anything more than episodic attacks on whitebark pine populations (Logan et al. 2010). Due to much milder winter temperatures, the mountain pine beetle can now access these previously unavailable whitebark pine populations (Logan et al. 2010). Whitebark pine mortality caused by mountain pine beetles in Montana and the Greater Yellowstone Ecosystem ranges from 38-96% in trees over 5 inches in diameter at breast height which far exceeds that of any other five-needle pine species (Gibson et al. 2008). Mountain pine beetles prefer to attack large diameter mature cone-bearing trees and are also known to attack and kill rust-weakened trees (Hicke et al. 2006, Six and Adams 2007, Gibson et al. 2008). Loss of these mature cone-bearing trees is of high concern since whitebark pine can take 60-100 years to reach full cone production and these mature trees provide the primary source for genetic resistance against white pine blister rust (Snieszko 2006, Schoettle and Snieszko 2007).

Whitebark pine forests in subalpine areas of the Rocky Mountains have a historical fire frequency ranging between 30-300 years (Arno and Hoff 1989, Morgan et al. 1994) and are commonly characterized by mixed severity natural fire regimes, although non-lethal surface fires and stand replacing fires also occur in these ecosystems (Keane and Parsons 2010). Fire helps to maintain whitebark pine forests by reducing competing vegetation (particularly subalpine fir) and creating new openings for seedling establishment (Aubry 2008). Also, Clark's nutcrackers prefer to cache seeds on distinct open sites such as burns (McCaughey 1990). For most of the 20th century Federal Forest Service policy focused on suppressing all wildland fires with the goal of protecting valuable timber resources and rural communities, however this policy failed to take into account the ecological importance of wildfires (Aplet 2006). Fire suppression has favored the establishment of more shade tolerant tree species creating uncharacteristically dense forests that now have reduced habitat for seed caching and seed germination of whitebark pine (Keane and Arno 1993, Morgan et al. 1994).

With more than 90% of whitebark pine populations occurring on public lands, the coordinated efforts of government land management agencies are critical in developing a successful restoration plan (Tomback and Achuff 2010, Keane et al. 2012). Shared knowledge regarding research and restoration efforts will help create a more efficient and successful management strategy.

Restoration

Restoration efforts have been on-going for over 30 years (Schwandt 2006) and have focused on promoting conservation and regeneration of whitebark pine populations (Keane and Schoettle 2011, Keane et al. 2012). These efforts have led to the development and improvement of whitebark pine restoration techniques, including cone collection, nursery techniques, seed germination, rust resistance selection, out-planting methods and genetic conservation.

Whitebark Pine Seed Zones

Whitebark pine seed zones and transfer guidelines have been developed to limit the movement of genetic material from one region to another, with the assumption that seeds genetically adapted to local conditions have a better chance of survival. Seed zones have been developed separately for the Pacific Northwest and the Rocky Mountain region (Keane et al. 2012). In the Rocky Mountain region, five seed zones have been established that divide the range of whitebark pine based on natural mountain ranges and on the level of rust infection (Mahalovich and Hoff 2000); transfer guidelines have been established to restrict movement within seed zones based on stand characteristics (Burns et al. 2008).

Rust Resistance Screening in Whitebark Pine Seedlings

Rust resistance breeding programs have been developed to increase the level of genetic resistance to white pine blister rust in whitebark pine (Hoff et al. 2001, Schoettle and Sniezko 2007). Management of natural resistance provides an avenue for potentially

establishing a balance between the survival of whitebark pine and this naturalized non-native pathogen (Hoff et al. 2001, Aubry et al. 2008). Rust-resistance programs for whitebark pine have been adapted from methods developed for western white pine trees (Hoff et al. 2001). These programs typically follow a two-step process that involves locating and identifying potentially resistant genetic stock followed by artificial rust inoculation trials (McDonald and Hoff 2001 and Mahalovich et al. 2006).

A successful rust-resistance breeding program relies heavily on locating and protecting potentially rust-resistant cone-bearing whitebark pine trees from mountain pine beetle attack (Mahalovich and Dickerson 2004). Genetic conservation is critical for these programs due to the high mortality that has been seen in whitebark pine populations so far (Aubrey et al. 2008). Potentially resistant trees are those displaying little or no signs of infection in locations where the pines have otherwise been decimated by blister rust (Mahalovich and Dickerson 2004) and are termed 'plus' trees. Verbenone, an anti-aggregation pheromone, is being used to protect potentially resistant cone-bearing trees from mountain pine beetle attack (Kegley and Gibson 2004, Bentz et al. 2005). Verbenone pouches applied prior to mountain pine beetle flight have been found to provide at least 80% protection of individual trees from successful mountain pine beetle attack (Kegley and Gibson 2009).

Seeds from these 'plus' trees are collected with the assumption that resistance is heritable (Hoff et al. 2001). Seedlings are then grown from the collected seeds and artificially inoculated with blister rust spores to evaluate variation in resistance levels and to subsequently select genetic stock with the ability to survive repeat inoculations

(Mahalovich et al. 2006). The development of needle lesions, early stem symptoms, bark reaction and canker tolerance are used as indicators in evaluating blister rust resistance levels as these responses are consistently observed on artificially inoculated seedlings (Hoff et al. 2001, Mahalovich et al. 2006). Restoration managers screen cones for genetic resistance at testing centers to ensure that the seedlings grown for restoration purposes can withstand levels of white pine blister rust infection (Sniezko et al. 2011).

Seed orchards are another option for the genetic conservation of rust-resistant whitebark pine (Tomback and Achuff 2010). Whitebark pine seed orchards are created by grafting rust-resistant whitebark pine branches onto mature root-stocks. Trees in these orchards are then able to produce cones within a few years of grafting, subsequently producing an accessible and stable supply of rust-resistant whitebark pine seeds for future restoration efforts (Mahalovich and Dickerson 2004, Aubrey et al. 2008). The Forest Service anticipates that whitebark pine seed orchards will be producing rust-resistant seed for seed zones in Montana and Idaho by 2015 (Scott et al. 2011).

Growing Whitebark Pine Seedlings

The USDA Forest Service Dorena Genetic Resource Center in Cottage Grove, Oregon and the USDA Forest Service Coeur d'Alene Nursery in Coeur d'Alene, Idaho are the primary facilities growing whitebark pine seedlings. Due to the seed characteristics and the slow growing nature of whitebark pine, nursery cultivation of seedlings for restoration has proven to be very challenging.

In order to grow whitebark pine seedlings, it is first necessary to identify mature cone-bearing 'plus' trees and to protect developing cones from predation during

maturation. Cages are usually applied to 2nd year maturing cones in the early summer months (Burr et al. 2001) with cones commonly being collected in the fall (Berdeen et al. 2007, Murray 2007). Once cones are collected and dried, seeds are carefully extracted and a random sample is often examined using an X ray. This is a common, quick and non-destructive procedure used to assess the quality of the seed cleaning, to estimate potential germination rates, to determine if the gametophyte is damaged, and to check for the presence of insect larvae in seeds (Burr et al. 2001).

Whitebark pine seeds normally take two or more years to germinate under natural conditions due to physiological and morphological characteristics of the seeds (Tomback et al. 2001c, McCaughey and Tomback 2001). To hasten germination in the nursery, seed stratification and germination techniques have been adapted specifically for whitebark pine (Mahalovich et al. 2006, Schwandt et al. 2007). The most effective stratification process developed to date requires a month-long warm stratification (22^o C day/20^o C night) followed by a 60-100 day cold stratification (1-2^o C); (Burr et al. 2001, Riley et al. 2007). Following the stratification process, seeds were historically germinated on petri dishes before planting. Today, the Coeur d'Alene Forest Service Nursery in Idaho plants two non-scarified seeds directly into each container (Eggleston 2010).

The whitebark pine seedlings used in reforestation in the Rocky Mountain area are grown almost exclusively at the Coeur d'Alene Forest Service Nursery in Idaho. The nursery has developed a standard protocol for growing high-quality whitebark pine seedlings; however these practices are continually evolving in terms of testing new stratification and germination techniques, growth substrates, and fertilizer regimes.

Whitebark pine seedlings grow at a slow rate and are often not ready for out-planting until 2-3 years of age (Burr et al. 2001).

The planting process is an important step in the survival of whitebark pine seedlings. Out-planting can be a weak link in the restoration process if appropriate planting techniques are not carefully followed. A set of whitebark pine planting guidelines has been developed to assist land managers in their restoration efforts (McCaughey et al. 2009, Scott et al. 2011). These guidelines suggest that healthy, robust seedlings be out-planted in the fall in burned or open areas where they have access to space and light (Izlar 2007, McCaughey et al. 2009, Scott et al. 2011). Planting near a microsite, such as a large stump or rock, that can provide physical protection from the elements is also recommended and thought to increase survival success (Izlar 2007, McCaughey et al. 2009, Scott et al. 2011). Snags and small rocks and logs that could fall on or roll over the seedling should be avoided in microsite selection. Whitebark pine seedling growth and survival may be reduced if planted near quick growing conifer species, in areas with dense understories, in deep, wet soils, or near standing dead trees (Izlar 2007, McCaughey et al. 2009).

Fire and Silviculture

In 1993, the Forest Service initiated a study termed Restoring Whitebark Pine Ecosystems (RWPE), one of the first “formal restoration studies” of whitebark pine ecosystems (Keane and Parsons 2010). The goal of this project was to create a useful set of guidelines to assist land managers in selecting the appropriate restoration treatment for whitebark pine forests. The RWPE was a large scale and long term study that involved

numerous whitebark pine forests in the Rocky Mountains. The study looked at natural whitebark pine regeneration across 19 sites after high, moderate and low severity prescribed burns and mechanical cutting techniques such as fuel enhancement, small clear cuts (1-3 acres), and slashing or thinning of competing vegetation. The planting of whitebark pine seedlings was not a major factor in this study and results depended mostly on natural regeneration; however seedlings were planted at two study sites with a relatively low survival of less than 30% after five years. Use of new planting techniques may improve survival success, and the planting of whitebark pine seedlings is now recommended when using prescribed burns in restoration (Keane and Parsons 2010).

The RWPE study found that while utilization of mechanical cutting techniques without prescribed burns was effective at creating canopy openings, the leftover slash created additional problems for natural whitebark pine regeneration. The slash hindered seed caching by Clark's nutcracker by obstructing access to the ground, provided increased habitat for *Ips* bark beetles, and enhanced the risk of severe wildfire (Keane and Parsons 2010). Combining cutting techniques with prescribed burns was successful at creating openings with reduced competition from more shade tolerant conifers. Caching by Clark's nutcracker was observed after both high and moderate prescribed burn treatments when a sufficient cone crop surrounded the site. However, natural whitebark pine regeneration across all sites was poor and at some sites many whitebark pine trees died due to fire-related factors. Keane and Parsons (2010) offer a few likely reasons for low natural whitebark pine generation. First, they suggest that many cached whitebark pine seeds were re-claimed by Clark's nutcracker due to low levels of seed production

near the study area. They also hypothesize that severe site conditions, such as heavy creeping snow packs and erosive top soil, may have removed growing seedlings. It is also possible that five years may be too short a time for accurate evaluation of regeneration on these unstable high elevation sites (Keane and Parsons 2010).

Monitoring Whitebark Pine Seedling Survival

Hundreds of thousands of seeds and seedlings have been planted in the Western U.S. and Canada, although monitoring of these efforts has been scarce, often limited by minimal personnel and financial resources (Izlar 2007, Keane et al. 2012).

Currently, whitebark pine reforestation relies mainly on the planting of two-year-old nursery grown seedlings. Only two facilities are involved in growing whitebark pine seedlings for restoration, and access to seedlings may prove limiting in future restoration efforts. In addition, “customers” must provide their own cones from ‘plus’ trees found within their seed zone to the nursery. The direct out-planting of whitebark pine seeds into soil is being studied as a practical alternative, especially in remote areas with difficult access; this method is less expensive and less time consuming than planting nursery-grown seedlings (Schwandt et al. 2011). Monitoring has also been conducted on emergence and survival of both planted whitebark pine seeds and nursery seedlings, with a focus on the effects of different site conditions (McCaughey 1990, Mellmann-Brown 2005, Perkins 2004, Izlar 2007, Asebrook 2011, and Schwandt 2011).

In 1990, McCaughey conducted one of the first studies evaluating ecological factors affecting whitebark pine seed establishment. Over 16,000 seeds were planted in the Gallatin National Forest, Montana, and germination rates were evaluated for a variety

of seed sowing depths, predation exclusion levels, light levels, and seedbed conditions. Surface sown seeds had a very low germination rate (6%) compared to buried seeds (45.4%) after accounting for rodent predation (McCaughey 1990). However, rodent foraging was observed at all surface-sown and buried seed locations when protective cages were not applied (McCaughey 1990). Caged seeds averaged much higher germination rates when compared to un-caged seeds and this method appears to better control for rodent predation when compared to rodent repellents such as Thiram, Ropel, and cayenne pepper (McCaughey 1990, Schwandt et al. 2011). In contrast, Mellmann-Brown (2005) monitored emergence and eight-year survival for 2,325 whitebark pine seeds, left all planting sites unprotected from animal foraging and observed no signs of seed predation, although her sites were at timberline.

In terms of other site factors, both McCaughey (1990) and Mellmann-Brown (2005) found shade to be important for the survival of whitebark pine seedlings. This is in contrast to more current recommendations that suggest whitebark pine seedlings are shade intolerant (Izlar 2007, Scott et al. 2011). McCaughey (1990) also found comparable germination rates between both litter (14.5%) and burned (15.3%) seedbeds, although both were low. Tomback et al. (2001b) monitored natural regeneration patterns following the 1988 Yellowstone fires and found fire to have little effect on natural whitebark pine seed germination.

In June of 2001, 3,355 whitebark pine seeds were planted in the Bitterroot Mountains on the Montana-Idaho border to test the effects of fire on seedling establishment, growth and survival (Perkins 2004). These seeds were planted near four

dominant understory plants (*Vaccinium scoparium* Leiberg ex Coville, *Luzula hitchcockii* Hämet-Ahti, *Xerophyllum tenax* (Pursh) Nutt., and *Carex geyeri* Boott.) to test for positive or negative associations with whitebark pine (Perkins 2004). Seeds planted in burned plots had higher germination and seedlings had increased biomass compared to unburned plots. A positive association between *V. scoparium* (grouse whortleberry) and whitebark pine survival was observed; although seedlings planted near either *X. tenax* (beargrass) or *C. geyeri* (Geyer's sedge) had significantly decreased biomass (Perkins 2004). The dense, rhizomatous root mass of both *X. tenax* and *C. geyeri* may have restrictive effects on conifer establishment; in addition both plants show a high degree of resistance to fire and are able to re-sprout quickly from underground rhizomes (Crane 1990, Chadwick 2002, Perkins 2004, Izlar 2007).

Mellmann-Brown (2005) also reports informative associations between vegetative communities and appropriate whitebark pine habitat. Seeds planted near vegetative communities defined by *Salix glauca* L. or dry *Geum rossii* (R. Br.) Ser. turf showed increased germination and survival rates. This is in comparison to seeds planted near vegetative communities defined by either *Carex elynoides* T. Holm or *Silene acaulis* L. Jacq/*Arenaria obtusiloba* (Rydb.) Fernald which showed poor germination and had no surviving seedlings 5-8 years after planting. *Salix glauca* typically grows in sub-alpine and alpine meadows, often on well drained soils near the transition between wetland and upland moisture conditions (Uchytel 1992, Mellmann-Brown 2005). This shrub pioneers disturbed sites and helps facilitate natural conifer regeneration (Uchytel 1992, Mellmann-Brown 2005). Dry *Geum rossii* turf is characterized by intermediate temperature and

moisture regimes and a lack of competing conifers (Mellmann-Brown 2005). Mellmann-Brown (2005) considered these two community types to be good indicators of favorable planting conditions for whitebark pine seeds in the timberline ecotome.

Schwandt et al. (2011) found that a 30-day warm stratification greatly improved germination rates of whitebark pine seeds planted at moderate sites (72% vs. 51%) as well as at harsh exposed sites (43% vs. 17%). However, after three years most seedlings planted at the harsh exposed site had died due to heat exposure and only 2% of the total seeds planted remained alive (Schwandt et al. 2011). Mellmann-Brown (2005) also reports sites with relatively good germination rates (29-80%) but with no seedling survival after 5-8 years. Appropriate site characteristics may improve germination and survival of planted whitebark pine, however site conditions favorable for whitebark pine germination may not be favorable for seedling survival (Tomback et al. 2001b, Mellmann-Brown 2005, Larson and Kipfmueller 2010). Studies indicate that planting sites with warmer average temperatures experience higher germination rates, but also experience higher mortality due to solar radiation and drought (McCaughey 1990, Tomback et al. 2001c, Larson and Kipfmueller 2010). Larson and Kipfmueller (2010) suggest that while planting in cooler sites may have lower germination rates, seedling survival rates could be improved. Another consideration is that soil temperatures are often higher on severe burn sites in comparison to adjacent forests (Trusty 2009).

In 2007, Kay Izlar conducted the first wide-scale monitoring of planted nursery grown whitebark pine seedlings. Izlar consolidated information on whitebark pine plantings on federal lands from 1989 -2005 and monitored the survival of over 100,000

whitebark pine seedlings (Izlar 2007). Overall survival rates for these monitored seedlings averaged about 42%, with a rapid decrease between 1st year survival (74%) and 3rd-15th year survival (38%). Mellmann-Brown (2005) also observed a rapid decrease in survival between the 1st year (36%) and 8th year survival (14%).

Izlar (2007) monitored seedlings planted over a wide range of sites and ecological conditions and found that seedlings planted in mixed severity burns averaged a higher survival rate (52%) compared to seedlings planted in unburned areas (21%); also seedlings planted in dry or mesic conditions averaged a higher survival rate (48%) compared to seedlings planted in wet sites (25%). Seedling survival also appeared to be significantly affected by the presence of a microsite such as large logs or rocks that can provide physical protection from solar radiation, wind, snow drifts, and soil erosion (Izlar 2007, McCaughey et al. 2009). However, Schwandt et al. (2011) found that large shade logs, while important for seedling survival, had little to no effect on seed germination.

Larson and Kipfmueller (2010) inventoried 8,467 naturally regenerating whitebark pine of varying ages across Montana, Idaho, and Oregon to evaluate relationships between site characteristics and regeneration patterns. Comparable regeneration was observed on both burned and unburned sites, suggesting that whitebark pine regeneration is not limited to burns (Larson and Kipfmueller 2010). A positive association between whitebark pine regeneration and mountain pine beetle mortality was also observed, suggesting that beetle-infected stands may possibly be favorable sites for planting nursery grown seedlings (Larson and Kipfmueller 2010); however more data is needed to draw definitive conclusions. This study and others (Izlar 2007, McCaughey et

al. 2009) have found that planting nursery grown seedlings in cold, dry environments could increase survival of whitebark pine seedlings.

Waterton Lakes-Glacier International Peace Park Whitebark Pine Restoration

Waterton Lakes-Glacier International Peace Park (WGIPP), established in 1932, totals approximately 460,000 ha of diverse habitat ranging from prairies to glaciated mountains (World Conservation Monitoring Centre 2011). Research and monitoring of WGIPP's five-needle pines aims at improving conservation, restoration, and management techniques for these tree species.

Whitebark pine populations have dramatically decreased within Glacier National Park (GNP), with mortality estimated at 46% in 2003 (Kendall and Keane 2001, Smith et al. 2008, Asebrook et al. 2011). Monitoring of whitebark pine populations between 1990 and 2010 has shown a continual decline in healthy trees with at least 70% of live trees infected with white pine blister rust (Kendall and Keane 2001, Smith et al. 2008, Asebrook et al. 2011). Whitebark pine restoration within GNP began in 1997 with the first seedlings planted in 2000 (Asebrook et al. 2011). Since then over 7000 seedlings have been planted and restoration efforts have expanded to include monitoring of 'plus' trees, cone collection, out-planting, and monitoring of seedling survival (Asebrook et al. 2011).

Between the years of 2000 and 2007, 6,400 nursery grown whitebark pine seedlings and 723 seeds were planted within GNP (Asebrook et al. 2011). Approximately 21% of the planted seedlings are being monitored and 3-8 year survival rates are

consistent with previous studies averaging 41% (Asebrook et al. 2010, Izlar 2007). However, for planted whitebark pine seeds, first year germination rates were very low, with three seeds germinating and only two surviving the first season (Asebrook et al. 2011). Seeds were neither stratified nor caged, and animal foraging was observed at all monitored sites (Asebrook et al. 2011).

Waterton Lakes National Park (WLNP) has also seen a dramatic and rapid decline in whitebark pine populations, with mortality increasing from 26% in 1996 to 61% in 2009 (Smith et al. 2008, Smith et al. 2011b). White pine blister rust infection levels have also shown a rapid increase from 44% in 1996 to 78% in 2009 (Smith et al. 2008, Smith et al. 2011b). Whitebark pine restoration efforts within WLNP began in 2006 with the first seedlings planted in 2009, since then over 2,900 seedlings have been planted (Smith 2012). Current restoration efforts include the protection of ‘plus’ trees from mountain pine beetle attack, cone collection, out-planting, and monitoring (Parks Canada 2010). Most recently, a terra torch has been used to mimic lightning strikes in order to remove competing vegetation, particularly subalpine fir and Engelmann spruce and to open canopy space to encourage caching of seeds by Clark’s nutcracker (Schwanke and Smith. 2010).

As part of a \$90 million investment to address ecological concerns in National Parks across Canada, \$7.1 million has been invested in the *Restoring Terrestrial Ecosystems* project in Waterton Lakes National Park, which in part addresses declining whitebark pine populations. In 2008, whitebark pine was approved for listing as an endangered species in the province of Alberta (Government of Alberta 2010). The

Committee on the Status of Endangered Wildlife in Canada designated whitebark pine as endangered in 2010 (COSEWIC 2010), and following this designation it was federally listed throughout Canada in July of 2012 (Government of Canada 2012). One focus of the WLNP restoration effort will be to reverse the decline of whitebark pine communities through the collection and propagation of 'plus' seeds and the planting of seedlings, including the use of mycorrhizal fungi critical to their survival (Parks Canada 2010). As of July 18, 2011, the U.S. Fish and Wildlife Service determined that whitebark pine warranted protection under the Endangered Species Act (Nicholas and Katzenberger 2011). The U.S. Fish and Wildlife Service determined that the threats to whitebark populations are imminent and of high magnitude and that this species is in danger of extinction; however it is currently precluded from protection due to priority constraints (Nicholas and Katzenberger 2011).

Mycorrhizae

Whitebark pine, like all pines, is an obligate mycorrhizal associate, and requires ectomycorrhizal fungi to survive in nature (Read 1998, Smith and Read 2008). Seedlings that lack native ectomycorrhizal fungi may have a severe disadvantage in natural ecosystems. Ectomycorrhizal fungi have been shown to enhance conifer survival through increased nutritional uptake, enhanced drought tolerance, and protection from harmful soil biota (Cripps 2004). Therefore, ectomycorrhizal fungi should be considered in restoration efforts and especially when the planting of nursery grown seedlings is involved. Areas that may be devoid of ectomycorrhizal fungi appropriate for whitebark

pine include severe burns, ghost forests (dead forests) and areas not previously inhabited by whitebark pine (Cripps and Grimme 2011).

Mycorrhizae are a mutualistic relationship between certain fungi and the roots of vascular plants. This association provides plants with additional nutrients while the fungi receive a source of carbohydrates (Allen 2003, Smith and Read 2008); mycorrhizae, not roots, are the primary organ involved in nutrient and water acquisition by land plants (Smith and Read 2008). The mycorrhizal association has been estimated to be over 450 million years old and is the natural state of over 85% of all plant species (Allen 2003). Mycorrhizae are thought to have played a pivotal role in the colonization of land by plants and continue to influence the structure of many plant communities (van der Heijden et al. 1998, Wilkinson 2001, van der Heijden et al. 2002).

Ectomycorrhizae are a specific type of mycorrhizal association that is characterized by a mantle, Hartig net (no intracellular penetration), and a mycelial network (Smith and Read 2008). The mantle forms a sheath on the fine roots of the host plant and may provide some root protection (Brundrett et al. 1996, Cripps 2004). The Hartig net is the site of nutrient and carbohydrate exchange. It is formed where hyphae penetrate between cortical cells forming an interface between the plant and fungus (Brundrett et al. 1996). A mycelial network spreads through the soil, dramatically increasing the area available for absorption of water and nutrients (Brundrett et al. 1996).

Ectomycorrhizal (ECM) fungi are found in the Basidiomycota, Ascomycota and Zygomycota phyla and mainly associate with large woody trees (Cripps 2004, Smith and Read 2008). Certain ectomycorrhizal fungi are generalists and form associations with a

wide range of host plants, while others may be more restricted and only form associations with specific host plants (Molina et al. 1992, Smith and Read 2008). Ectomycorrhizal fungi follow successional patterns with different fungi adapted to occupying different niches in forest development (Frankland 1998, Allen et al. 2003). Thousands of species of fungi are thought to form ectomycorrhizal associations (Molina et al. 1992).

Only about 50 species of native ectomycorrhizal fungi are known to occur with whitebark pine in the Greater Yellowstone Area (Cripps and Antibus 2011, Mohatt et al. 2008). Of these, certain suilloid fungi (Basidiomycota) specific to five-needle pines are important ectomycorrhizal associates of both young and mature whitebark pine trees (Tedersoo et al. 2009, Cripps and Antibus 2011). Suilloid fungi are primarily restricted to hosts in the *Pinaceae* and tend to form associations with specific tree genera, subgroups, or species (Bruns et al. 2002). In addition, suilloid mushrooms are an integral part of the food chain in whitebark pine forests and are eaten by squirrels, deer, elk and bears (Fortin 2011, Cripps 2012).

It is now well documented that whitebark pine seeds are an important food source for black bears and female grizzly bears in the Greater Yellowstone Ecosystem (GYE), especially in the months prior to hibernation (Mattson et al 1992, Fortin 2011). However, in years when whitebark pine seed production is minimal, female grizzly bears in the GYE feed almost exclusively on suilloid fungi, specifically *Rhizopogon* species (false-truffles); (Fortin 2011). These ectomycorrhizal suilloid fungi cannot survive in nature without pine trees, and if the host declines so will this alternate food source. Thus, suilloid fungi are linked directly to bears as well as whitebark pine trees (Fortin 2011).

Suilloid fungi also occur with other stone pines in comparable forest ecosystems in the Swiss Alps and the Altai Mountains of Central Asia (Moser 2004). In fact, for over fifty years the Federal Forest Service Nursery in Austria has been successfully inoculating the European Stone pine (*Pinus cembra* L.) with three species of native suilloid fungi adapted to high elevation conditions (Moser 1956, Weisleitner 2008): *Suillus plorans* (Rolland) Kuntze, *S. sibiricus* Singer and *S. placidus* (Bonord.) Singer. Seedlings planted at high elevations have shown a 20-80% increase in survival when inoculated; when inoculation is combined with intensive silviculture techniques, survival increased a dramatic 50-90% (Moser 1956, Schmid 2006). In 2006, molecular techniques were used to identify mycorrhizae on the roots of seedlings from the Federal Nursery planted 25-30 years previously; surprisingly all three of the original suilloid species used to inoculate seedlings were still present and colonizing the root systems (Schmid 2006). The Federal Forest Nursery in Austria has been able to maintain this inoculum through periodic inoculation and the use of biodegradable pots. As the pots break down, roots grow from one pot to the next, subsequently inoculating neighboring seedlings in an outdoor nursery setting. Also, green manuring is used instead of chemical fertilizers to help maintain the ECM association. While this method has worked well in Austria, differences between stone pine species, nursery practices, climate and precipitation suggest that inoculation methods appropriate for the western U.S. need to be developed (Cripps and Grimme 2011).

Mycorrhizae and Whitebark Pine

Although mycorrhizal inoculation is routinely used in reforestation efforts for other conifer species (Khasa et al 2009), whitebark pine seedlings at the Forest Service nursery are not currently inoculated. Commercial inoculum appropriate for whitebark pine seedlings is not available and alien fungi should not be used in these sensitive forest systems (Keane et al. 2012).

Research regarding the inoculation of whitebark pine seedlings with native ectomycorrhizal fungi under standard nursery conditions is being conducted at Montana State University. Cripps and Grimme (2011) screened over twenty-five strains of native fungi from whitebark pine forests as potential inoculum. Initial trials showed that a variety of suilloid fungi that occur with five-needle pines can be used successfully as inoculum for whitebark pine seedlings and that some strains of *Suillus* colonized roots vigorously in initial trials (Mohatt 2008, Cripps and Grimme 2011). Of the inoculation methods tested on the whitebark pine seedlings, spore slurries made from fresh sporocarps were shown to be very effective compared to mycelial inoculum (Cripps and Grimme 2011). However, high elevation sites are prone to drought which prevents fungal fruiting and makes finding fresh sporocarps each year difficult. If dried sporocarps could be used to make spore slurries, there would be potential for the long-term storage of sporocarps for use as inoculum. A small trial showed dried spore powder is effective as an inoculum for limber pine, but this was only for a few seedlings and the powder was not made into slurry but was added directly to seeds (Antibus 2010, pers. comm.). Therefore, more trials are needed to test the possibility of using spore slurries made from

dried, preserved sporocarps so that inoculum can be available when needed. While the use of tissue-cultured fungi (mycelium) also produced mycorrhizae, it was not as effective and methods were labor-intensive (Cripps and Grimme 2011).

Fertilization and particular soil substrates have been shown to suppress mycorrhizal colonization in the nursery (Castellano et al. 1985, Smith and Read 2008, Rincón et al. 2005). The effects of fertilizer and various soil substrates on the mycorrhizal colonization of whitebark pine seedlings have been evaluated to a limited degree (Cripps and Antibus 2011, Cripps and Grimme 2011). Cripps and Grimme (2011) found that certain soil substrates (sunshine mix: soil-less blend of Canadian Sphagnum peat moss and horticultural grade Perlite, buffered with Dolomitic lime, and containing a starter fertilizer charge and wetting agent) and fertilizer regimes (200 ppm of 20:20:20 NPK once/week) seriously suppressed mycorrhizal colonization of whitebark pine seedlings. This might in part be due to a narrow tolerance of pH range by certain isolates of suilloid fungi (Cripps and Antibus 2011) as higher pH's are known to prevent spore germination in slurries (Castellano et al. 1985). Seedling age at the time of inoculation may also be a crucial factor affecting colonization success and should be investigated further.

Fertilizer regimes can affect mycorrhization of nursery-grown conifers (Khasa et al. 2001). Exponential fertilization, achieved by progressively increasing nutrient applications so they correspond with relative seedling growth rates (Ingestad and Lund 1986, McAllister and Timmer 1997, Quoreshi and Timmer 1998), was found to be conducive to ectomycorrhizal colonization of black spruce seedlings (Quoreshi et al. 1998). Western white pine (*Pinus monticola*), a 5-needle relative of whitebark pine, can

also form mycorrhizal associations under exponential fertilizer regimes (Dumroese et al. 2005). In addition, when whitebark pine seedlings were fertilized normally for 16 months and inoculated one month after fertilization was stopped, seedlings were well-colonized in the few months that preceded out-planting (Cripps and Grimme 2011). These results suggest that it may be possible to combine mycorrhizal colonization with a delayed, interrupted or exponential fertilization regime for whitebark pine seedlings. However fungal species and isolates vary in tolerance to soil substrates, fertilizer types, and rates of application, and therefore methods specific to whitebark pine and their native ECM fungi need to be developed.

Field Assessment and Monitoring of ECM Fungi Associated with Whitebark Pine

As previously mentioned, whitebark pine requires ectomycorrhizal fungi to survive in nature and the absence of the appropriate fungi may be detrimental to restoration efforts. Thus, it is important to determine whether planted whitebark pine seedlings have access to native ECM fungi and how effectively these fungi colonize seedling root systems. A road construction project on Dunraven Pass in Yellowstone National Park in 2007 and the subsequent planting of 4000 nursery grown seedlings provided the first opportunity for the assessment of natural mycorrhizal colonization of planted whitebark pine seedlings (Cripps and Trusty 2007). In this small study, a total of 20 seedlings were sacrificed, and a general estimate of the percent mycorrhizal colonization and frequency of each fungal species on roots were recorded for each seedling. Seventy percent of healthy seedlings hosted mycorrhizal fungi and mycorrhizal

colonization was correlated with higher fitness and increased survival two years after planting (Cripps and Trusty 2007, Izlar 2009). The native ectomycorrhizal fungi found on seedling roots were primarily suilloid fungi as detected by molecular techniques ITS sequencing); (Cripps and Trusty 2007). Another destructive study conducted on Scotch Bonnet Mountain in southeast Montana found that seedlings growing in avalanche paths outside the forest canopy hosted different ECM fungi than those seedlings occurring under the forest canopy (Mohatt 2006). Seedlings growing in clusters were also found to host a greater diversity of ECM fungi than seedlings growing alone (Mohatt 2006).

Two studies using minimally destructive techniques were conducted to evaluate natural mycorrhizal colonization of planted and natural whitebark pine seedlings. Minimally-destructive methods require that each seedling be carefully dug up keeping the majority of the root system intact. A total of 25-40 cm of root is removed from the top, middle, and lower portions of each root system before seedlings are replanted (Mohatt et al. 2008, Trusty 2009). One study was conducted in an almost pure whitebark pine stand that was partially burned during the 2001 Fridley wildfire in Gallatin National Forest. There was a significant species shift in ectomycorrhizal fungi on seedling roots between burned and unburned sites. However, whitebark pine seedlings were over 90% mycorrhizal on both burned and unburned sites and on both naturally regenerating and planted seedlings (Trusty 2009). Another study using minimally-destructive sampling was conducted in mixed whitebark pine forests within Waterton Lakes-Glacier International Peace Park (WGIPP). A diversity of suilloid fungi were found colonizing root systems of natural seedlings. ECM fungal diversity was low on seedlings occurring

in areas heavily populated with beargrass (*Xerophyllum tenax*); (Cripps et al. 2008). Both of these studies were conducted in areas hosting current populations of whitebark pine trees and thus appropriate ECM for whitebark pine seedlings were likely present.

A preliminary study evaluating survival success of out-planted, nursery grown whitebark pine inoculated with suilloid ectomycorrhizal fungi has been conducted in Waterton Lakes –Glacier International Peace Park. In Glacier, 300 seedlings were planted in the fall of 2010 immediately after inoculation. Planting sites were located near the Flattop Mountain area and monitoring data is not yet available for this site (Cripps 2011). In Waterton Lakes National Park, 93 inoculated and 397 non-inoculated seedlings were planted near Upper Rowe Lake in 2009 immediately after inoculation. Initial survival data from the Upper Rowe Lake plantings shows little difference between inoculated and un-inoculated seedlings (Smith 2012). Due to time restrictions, these seedlings likely did not have enough time to form mycorrhizae before out-planting; also this site is known to host ectomycorrhizal fungi appropriate for whitebark pine (Cripps and Antibus 2011). Detrimental effects due to an initial carbon drain to fungi were not noted.

Perkins (2004) found that whitebark pine seedlings planted near *Vaccinium scoparium* (grouse whortle-berry) experienced increased growth and survival rates compared to seedlings planted in bare ground. She hypothesized that whitebark pine and grouse whortle-berry may share mycorrhizal associates (Perkins 2004, Scott et al. 2011). Currently, there is no data to support this hypothesis (Mohatt 2006, Trusty 2009, Cripps 2011). Grouse whortleberry forms ericoid mycorrhizae, a distinctive association that occurs between certain plants in the Ericales and fungi in the Ascomycota (Smith and

Read 2008). To date, no ericoid mycorrhizal fungi have been found to form relationships with whitebark pine and no suilloid mycorrhizal fungi have been found to associate with *Vaccinium scoparium* (Largent et al. 1980, Mohatt 2006, Cripps and Eddington 2005, Trusty 2009).

In summary, ectomycorrhizal fungi should be considered in restoration efforts and especially when the planting of nursery grown seedlings is involved. This is particularly applicable for areas that may be devoid of ectomycorrhizal fungi appropriate for whitebark pine, including severe burns, ghost forests (dead forests), and areas not previously inhabited by whitebark pine.

Research Objectives

One overlooked restoration strategy that may enhance long-term seedling survival is inoculation of nursery grown seedlings with native ectomycorrhizal fungi (Cripps 2011). A goal of our lab is to examine the use of ectomycorrhizal fungi as an additional tool for the restoration of whitebark pine communities. My research consists of a greenhouse (inoculation of seedlings) and field study (monitoring seedlings). The main goal of the greenhouse study is to determine whether seedlings inoculated and colonized with *Suillus sibiricus* (a native ECM fungus) can maintain this mutualistic association under certain fertilizer regimes. The overall goal is to enhance seedling growth with fertilization while maintaining mycorrhizal colonization before out-planting. In addition, several other variables will be examined: a) use of dried versus fresh sporocarps for slurries, b) inject versus drip method for application, and c) use of long and short

containers. The latter is of interest because of the difficulty of planting seedlings in long containers in rocky, high elevation sites. These seedlings are slated to be planted in WGIPP if growth is sufficient.

The field study will evaluate whether inoculation with *Suillus sibiricus* (a native ECM fungus) increases survival rates of whitebark pine seedlings out-planted in four different habitat types (burn/non-burned, beargrass/non-beargrass) within Waterton Lakes National Park. Seedlings were planted in burned and unburned areas because it is currently believed that planting in burns improves seedling survival (Izlar 2007, McCaughey et al. 2009). Seedlings were planted in areas with and without the presence of beargrass because it is the dominant understory vegetation at the site and is thought to negatively impact whitebark pine seedling survival (Perkins 2004, Izlar 2007). This will be the first attempt to monitor inoculated whitebark pine seedlings in a detailed manner. Future studies will address the use of native ECM fungi to help reestablish planted whitebark pine seedlings in areas devoid of native ECM fungi, such as ghost forests.

References

- Allen, M.F., Swenson, W., Querejeta, J.I., Egerton-Warburton, L.M., and Treseder, K.K. 2003. Ecology of mycorrhizae: a conceptual framework for complex interactions among plants and fungi. *Annual Review of Phytopathology* 41: 271-303.
- Antibus, R.K. 2010. [Personal communication]. Professor, Department of Science, Bluffton University, Bluffton, Ohio.
- Aplet, G.H. 2006. Evolution of wilderness fire policy. *International Journal of Wilderness* 12(1): 9-13.
- Arno, S.F. 1986. Whitebark pine cone crops - a diminishing source of wildlife food? *Western Journal of Applied Forestry* 1: 92-94.
- Arno, S.F. 2001. Community types and natural disturbance processes. Pp. 74-88. In: Tomback, D.F., Arno, S.F., and Keane, R.E., (eds.). *Whitebark pine communities: ecology and restoration*. Island Press, Washington, DC. 439 p.
- Arno, S.F. and Hoff, R.J. 1990. Silvics of whitebark pine (*Pinus albicaulis*). General Technical Report INT-253. U.S. Department of Agriculture, Forest Service, Intermountain Research Station, Ogden, UT.
- Asebrook, J., Lapp, J., and Carolin, T. 2011. Whitebark and limber pine restoration and monitoring in Glacier National park. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). *The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium*. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Aubry, C., Goheen, D., Shoal, R., Ohlson, T., Lorenz, T., Bower, A., Mehmel, C., and Sniezko, R. 2008. Whitebark pine restoration strategy for the Pacific Northwest region 2009-2013. U.S. Department of Agriculture, Forest Service. 13p.
- Bentz, B.J., Kegley, S., Gibson, K. and Thier, R. 2005. A test of high-dose verbenone for stand-level protection of lodgepole and whitebark pine from mountain pine beetle (Coleoptera: Curculionidae: Scolytinae) attacks. *Journal of Economic Entomology* 98(5): 1614-1621.
- Berdeen, J.C., Riley, L.E., and Sniezko, R.S. 2007. Whitebark pine seed storage and germination: a follow-up look at seedlots from Oregon and Washington. Pp. 113-

121. Proceedings: Conference on Whitebark pine: A Pacific Coast Perspective. USDA Forest Service R6-NR-FHP-2007-01.
- Bockino, N.k. 2008. Interactions of white pine blister rust, host species, and mountain pine beetle in whitebark pine ecosystems in the Greater Yellowstone. Master's Thesis University of Wyoming, Laramie, WY.
- Bower, A.D. and Aitken, S.N. 2008. Ecological genetics and seed transfer guidelines for *Pinus albicaulis* (*Pinaceae*). *American Journal of Botany* 95(1): 66-76.
- Brundrett, M., Bougher, N., Dell, B., Grove, T., and Malajczuk, N. 1996. Working with mycorrhizas in forestry and agriculture. ACIAR Monograph 32.
- Bruns, T.D., Bidartondo, M.I., and Taylor, L. 2002. Host specificity in ectomycorrhizal communities: what do the exceptions tell us? *Integrative and Comparative Biology* 42: 352-359.
- Burns, K.S., Schoettle, A.W., Jacobi, W.R., and Mahalovich, M.F. 2008. Options for the management of white pine blister rust in the Rocky Mountain region. USDA Forest Service General Technical Report RMRS-GTR-206.
- Burr, K., Eramian, A., and Eggleston, K. 2001. Growing whitebark pine seedlings for restoration. Pp. 325-345. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds). *Whitebark pine communities: ecology and restoration*. Island Press, Washington, DC.
- Castellano, M.A., Trappe, J.M., and Molina, R. 1985. Inoculation of container-grown Douglas-fir seedlings with basidiospores of *Rhizopogon vinicolor* and *R. colossus*: effects of fertility and spore application rate. *Canadian Journal of Forest Research* 15: 10-13.
- Chadwick, A.C. 2002. *Carex geyeri*. In: Fire Effects Information System. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station, Fire Sciences Laboratory (Producer). [Online]. Available: <http://www.fs.fed.us/database/feis/> [2011, June 20].
- COSEWIC. 2010. COSEWIC assessment and status report on the Whitebark Pine *Pinus albicaulis* in Canada. Committee on the Status of Endangered Wildlife in Canada. Ottawa. Pp 44. (www.sararegistry.gc.ca/status/status_e.cfm).
- Crane, M. F. 1990. *Xerophyllum tenax*. In: Fire Effects Information System. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station, Fire Sciences Laboratory (Producer). [Online]. Available: <http://www.fs.fed.us/database/feis/> [2011, June 20].

- Cripps, C.L. 2004. Fungi in forest ecosystems: systematics, diversity and ecology. New York Botanical Gardens Press, NY. 363 p.
- Cripps, C.L. 2011. [Personal Communication]. July 15, 2011. Associate Professor, Montana State University, Bozeman, Montana.
- Cripps, C.L. and Antibus, R.K. 2011. Native ectomycorrhizal fungi of limber and whitebark pine: necessary for forest sustainability? In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Cripps, C.L. and Eddington, L.H. 2005. Distribution of mycorrhizal types among alpine vascular plant families on the Beartooth Plateau, Rocky Mountains, U.S.A., in reference to large-scale patterns in arctic-alpine habitats. *Arctic, Antarctic, and Alpine Research* 37(2): 177-188.
- Cripps, C.L. and Grimme, E. 2011. Inoculation and successful colonization of whitebark pine seedlings with native mycorrhizal fungi under greenhouse conditions. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Cripps, C.L., Smith, C., Carolin, T., and Lapp, J. 2008. Ectomycorrhizal fungi with whitebark pine. *Nutcracker Notes*. 14: 12-13.
- Cripps, C.L. and Trusty, P. 2007. Final Report. Whitebark pine restoration, Dunraven Pass: Monitoring mycorrhizal status of seedlings. Greater Yellowstone Coordinating Committee.
- Dumroese, K., Page-Dumroese, D.S., Salifu, K.F., and Jacobs, D.F. 2005. Exponential fertilization of *Pinus monticola* seedlings: nutrient uptake efficiency, leaching fractions, and early out-planting performance. *Canadian Journal of Forest Research* 35: 2961-2967.
- Eggleston, K. 2010. [Personal Communication to Dr. Cathy Cripps]. April 19, 2010. U.S. Department of Agriculture Forest Service Nursery, Coeur D'Alene, Idaho.
- Ellison, A.M., Bank, M.S., Clinton, B.D., Colburn, E.A., Elliott, K., Ford, C.R., Foster, D.R., Kloppel, B.D., Knoepp, J.D., Lovett, G.M., Mohan, J., Orwig, D.A.,

- Rodenhouse, N.L., Sobczak, W.V., Stinson, K.A., Stone, J.K., Swan, C.M., Thompson, J., Von Holle, B., and Webster, J.R. 2005. Loss of foundation species: consequences for the structure and dynamics of forested ecosystems. *Frontiers in Ecology and the Environment* 3: 479–486.
- Farnes, P.E. 1990. SNOTEL and snow course data: describing the hydrology of whitebark pine ecosystems. Pp. 302-304. In: Schmidt, W.C. and McDonald, K.J. (comps.). *Proceedings: Symposium on whitebark pine ecosystems: ecology and management of a high-mountain resource*. USDA Forest Service, General Technical Report INT-270, Intermountain Research Station, Ogden, Utah.
- Fortin, J.K. 2011. Niche separation of grizzly (*Ursus arctos*) and American black bears (*Ursus americanus*) in Yellowstone National Park. Washington State University PhD dissertation, Pullman, WA.
- Frankland, J.C. 1998. Fungal succession – unraveling the unpredictable. *Mycological Research* 102: 1-15.
- Fryer, J.L. 2002. *Pinus albicaulis*. In: Fire Effects Information System, [Online]. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station, Fire Sciences Laboratory (Producer). Available: <http://www.fs.fed.us/database/feis/> [2011, May 23].
- Geils, B.W., Hummer, K.E., and Hunt, R.S. 2010. White pines, *Ribes*, and blister rust: a review and synthesis. *Forest Pathology* 40(3-4): 147-185.
- Government of Alberta. 2010. Species assessed by Alberta's Endangered Species Conservation Committee: short list [online]. Available: <http://www.srd.alberta.ca/BiodiversityStewardship/SpeciesAtRisk/SpeciesSummaries/documents/SpeciesAssessed-EndangeredSpeciesConservationCommittee> [June 3, 2010].
- Government of Canada. 2012. Order amending Schedule 1 to the Species at Risk Act. *Canada Gazette Part II* 146 (14) SOR/2012-113: 1418-1629. [online] Available: http://www.sararegistry.gc.ca/virtual_sara/files/orders/g2-14614i_e.pdf.
- Gibson, K., Skov, K., Kegley, S., Jorgensen, C., Smith, S., and Witcosky, J. 2008. Mountain pine beetle impacts in high-elevation five-needle pines: current trends and challenges. USDA Forest Service, Northern region, Missoula, MT. R1-08-020.
- Hicke, J.A., Logan, J., Powell, J., and Ojima, D.S. 2006. Changing temperatures influence suitability for modeled mountain pine beetle (*Dendroctonus*

ponderosae) outbreaks in the western United States. *Journal of Geophysical Research* (111): 1-12.

- Hoff, R.J., Ferguson, D.E., McDonald, G.I., and Keane, R.E. 2001. Strategies for managing whitebark pine in the presence of white pine blister rust. Pp. 346-366. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds). *Whitebark pine communities: ecology and restoration*. Island Press, Washington, DC.
- Ingestad, T., and Lund, A.B. 1986. Theory and techniques for steady state mineral nutrition and growth of plants. *Scandinavian Journal of Forest Research* 1: 439–453.
- Izlar, K. 2007. Assessment of whitebark pine seedling survival for Rocky Mountain plantings. University of Montana Master's thesis. Missoula, MT.
- Izlar, K. 2009. [Personal Communication to Dr. Cathy Cripps] 2009. U.S. Forest Service, Flathead National Forest, Montana.
- Keane, R.E. and Arno, S.F. 1993. Rapid decline of whitebark pine in western Montana: evidence from 20-year re-measurements. *Western Journal of Applied Forestry* 8: 44-47.
- Keane, R.E. and Parsons, R.A. 2010. Management guide to ecosystem restoration treatments: whitebark pine forests of the northern Rocky Mountains, U.S.A. General Technical Report RMRS-GTR-232. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 133 p.
- Keane, R.E. and Schoettle, A.W. 2011. Strategies, tools, and challenges for sustaining and restoring high elevation five-needle white pine forests in Western North America. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). *The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium*. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Keane, R.E., Tomback, D.F., Aubry, C.A., Bower, A.D., Campbell, E.M., Cripps, C.L., Jenkins, M.B., Manning, M., McKinney, S.T., Murray, M.P., Perkins, D.L., Reinhart, D.P., Ryan, C., Schoettle, A.W., Smith, C.M. 2012. A range-wide restoration strategy for whitebark pine (*Pinus albicaulis*). General Technical Report RMRS-GTR-279. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 108 p.
- Kendall, K.C. and Keane, R.E. 2001. Whitebark pine decline: infection, mortality, and populations trends. Pp. 221-242. In: Tomback, D.F., Arno, S.F., and Keane, R.E.

- (eds.). Whitebark pine communities: ecology and restoration. Island Press, Washington DC.
- Kegley, S. and Gibson, K. 2004. Protecting whitebark pine trees from mountain pine beetle attack using verbenone. Forest Health Protection Report 04-8.
- Kegley, S. and Gibson, K. 2009. Individual-tree tests of verbenone and green-leaf volatiles to protect lodgepole, whitebark and ponderosa pines, 2004-2007. Forest Health Protection Report 09-03.
- Khasa, P.D., Piche, Y., and Coughlan, A.P. (eds.). 2009. Advances in Mycorrhizal Science and Technology. NRC Research Press, Ottawa. 179 p.
- Khasa, P.D., Sigler, L., Chakravarty, P., Dancik, B.P., Erikson, L., and McCurdy, D. 2001. Effect of fertilization on growth and ectomycorrhizal development of container-grown and bare-root nursery conifer seedlings. *New Forests* 22: 179-197.
- Lanner, R.M. 1996. Made for each other: a symbiosis of birds and pines. Oxford University Press. New York, NY.
- Lantz, G. 2010. Whitebark pine: an ecosystem in peril. American Forests Special Report. Norman, OK.
- Largent, D.L., Sugihara, N. and Wishner, C. 1980. Occurrence of mycorrhizae on ericaceous and pyrolaceous plants in northern California. *Canadian Journal of Botany* 58: 2274-2279.
- Larson, E.R. and Kipfmüller, K.F. 2010. Patterns in whitebark pine regeneration and their relationships to biophysical site characteristics in southwest Montana, central Idaho, and Oregon, USA. *Canadian Journal of Forestry* 40: 476-487.
- Little Jr., E.L. 1971. Atlas of United States trees, volume 1, conifers and important hardwoods: U.S. Department of Agriculture Miscellaneous Publication 1146, 9 p., 200 maps.
- Logan, J., McFarlane, W., and Wilcox, L. 2010. Whitebark pine vulnerability to climate driven mountain pine beetle disturbance in the Greater Yellowstone Ecosystem. *Ecological Applications* 20(4): 895-902.
- Luckman, B.H., Jozsa, L.A., and Murphy, P.J. 1984. Living seven-hundred-year-old *Picea engelmannii* and *Pinus albicaulis* in the Canadian Rockies. *Arctic and Alpine Research*. 16(4): 419-422.

- Mahalovich, M.F., Burr, K.E., and Foushee, D.L. 2006. Whitebark pine germination, rust resistance, and cold hardiness among seed sources in the inland northwest: planting strategies for restoration. USDA Forest Service Proceedings RMRS-P-43.
- Mahalovich, M.F. and Dickerson, G.A. 2004. Whitebark pine genetic restoration program for the Intermountain West (USA). Pp. 181-187. In: Sniezko, R., Samman, S., Schlarbaum, S., and Kriebel, H. (eds.). Breeding and genetic resources of five-needle pines: growth, adaptability and pest resistance, IUFRO Working Party 2.02.15, Proceedings. USDA Forest Service, Rocky Mountain Research Station, RMRS-P-32, Ogden, UT.
- Mahalovich M.F. and Hoff, R.J. 2000. Whitebark pine operational cone collection instructions and seed transfer guidelines. Nutcracker Notes 11: 10-13.
- Mattson, D.J., Blanchard, B.M., and Knight, R.R. 1992. Yellowstone grizzly bear mortality, human habituation, and whitebark pine seed crops. Journal of Wildlife Management 56: 432-442.
- McAlister, J.A., and Timmer, V.R. 1998. Nutrient enrichment of white spruce seedlings during nursery culture and initial plantation establishment. Tree Physiology 18: 195-202.
- McCaughey, W.W. 1990. Biotic and microsite factors affecting *Pinus albicaulis* establishment and survival. Ph.D. dissertation Montana State University, Bozeman, MT.
- McCaughey, W.W. and Tomback, D.F. 2001. The natural regeneration process. Pp. 105-120. In: Tomback, D.F.; Arno, S.F.; Keane, R.E. (eds.). Whitebark pine communities: Ecology and restoration. Island Press, Washington, DC.
- McCaughey, W.W. and Schmidt, W.C. 2001. Taxonomy, distribution and history. Pp. 29-40. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds.). Whitebark pine communities, ecology and restoration. Island Press, Washington, DC.
- McCaughey, W., Scott, G.L., and Izlar, K.L. 2009. Whitebark pine planting guidelines. Western Journal of Applied Forestry 24(3): 163-166.
- McDonald, G.I. and Hoff, R.J. 2001. Blister rust: an introduced plague. Pp. 193-220. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds.). Whitebark pine communities, ecology and restoration. Island Press, Washington, DC.

- McKinney, S., Fiedler, C.E., and Tomback, D.F. 2009. Invasive pathogen threatens bird-pine mutualism: implications for sustaining a high-elevation ecosystem. *Ecological Applications* 19(3): 597-607.
- McKinney, S.T. and Tomback, D.F. 2007. The influence of white pine blister rust on seed dispersal in whitebark pine. *Canadian Journal of Forest Research* 37: 1044-1057.
- Mellmann-Brown, S. 2005. The regeneration of whitebark pine in the timberline ecotone of the Beartooth Plateau, U.S.A.: spatial distribution and responsible agents. Pp. 96-116. In: Broll, G. and Keplin, B. (eds.). *Mountain ecosystems: studies in treeline ecology*. Springer-Verlag, Berlin.
- Mohatt, K. 2006. Ectomycorrhizal fungi of whitebark pine (*Pinus albicaulis*) in the Northern Yellowstone Greater Ecosystem. Montana State University Master's Thesis, Bozeman, MT.
- Mohatt, K., Cripps, C.L., and Lavin, M. 2008. Ectomycorrhizal fungi of whitebark pine (a tree in peril) revealed by sporocarps and molecular analysis of mycorrhizae from treeline forests in the Greater Yellowstone Ecosystem. *Botany* 86: 14-25.
- Molina, R., Massicotte, H., and Trappe, J.M. 1992. Specificity phenomena in mycorrhizal symbiosis: community-ecological consequences and practical implications. In: Allen, M.F. (ed.). *Mycorrhizal Functioning*. Chapman and Hall, London, UK.
- Morgan, P., Bunting, S.C., Keane, R.E., and Arno, S.F. 1994. Fire ecology of whitebark pine forests of the Northern Rocky Mountains, U.S.A. Pp 136-141. In: Schmidt, W.C. and Holtmeier, F.K., (comps.). *Proceedings of the international workshop of subalpine stone pines and their environment: the status of our knowledge; September 5-11, 1992; St. Moritz, Switzerland*. General Technical Report INT-GRT-309. Ogden, UT: U.S. Department of Agriculture, Forest Service, Intermountain Research Station.
- Moser, M.M. 1956. Die Bedeutung der Mykorrhiza für Aufforestungen in Hochlagen. *Forstwissenschaftliches Centralblatt* 75: 8-18.
- Moser, M.M. 2004. Subalpine conifer forests in the Alps, the Altai, and the Rocky Mountains: a comparison of their fungal populations. Pp. 151-158. In: Cripps, C.L. (ed.), *Fungi in forest ecosystems: systematics, diversity, and ecology*. New York Botanical Garden Press, NY.
- Murray, M.P. 2007. Cone collecting techniques for whitebark pine. *Western Journal of Applied Forestry* 22(3): 153-155.

- Nicholas, A. and Katzenberger, D. 2011. Whitebark pine to be designated a candidate for endangered species protection. U.S. Dept. of Interior, Fish and Wildlife Service, Press Release, Lakewood, CO.
- Parks Canada. 2010. Parks Canada Invests in Ecological Integrity-Press Release. [online] Accessed: http://www.pc.gc.ca/apps/cp-nr/release_e.asp?bgid=1338&andor1=bg.
- Price, R.A., Liston, A., and Strauss, S.H. 1998. Phylogeny and systematics of *Pinus*. Pp. 49-68. In: Richardson, D.M. (ed.). Ecology and biogeography of *Pinus*. Cambridge University Press, Cambridge, UK.
- Perkins, J. 2004. *Pinus albicaulis* seedling regeneration after fire. University of Montana PhD dissertation, Missoula, MT.
- Perkins, D.L. and Swetnam, T.W. 1996. A dendroecological assessment of whitebark pine in the Sawtooth--Salmon River region, Idaho. Canadian Journal of Forest Research. 26: 2123-2133.
- Quoreshi, A.M. and Timmer, V.R. 1998. Exponential fertilization increases nutrient uptake and ectomycorrhizal development of black spruce seedlings. Canadian Journal of Forestry 28: 674-682.
- Read, D.J. 1998. The mycorrhizal status of *Pinus*. Pp. 324-340. In: Richardson, D.M. (ed.). Ecology and biogeography of *Pinus*. Cambridge University Press, Cambridge, UK.
- Riley, L.E., Coumas, C.M., Danielson, J.F., and Berdeen, J.C. 2007. Seedling nursery culture of whitebark pine at Dorena Genetic Resource Center: headaches, successes, and growing pains. USDA Forest Service Proceedings of the Conference Whitebark Pine: a pacific coast perspective. R6-NR-FHP-2007-01.
- Rincón, A., Parlade, J., and Pera, J. 2005. Effects of ectomycorrhizal inoculation and the type of substrate on mycorrhization, growth and nutrition of containerized *Pinus pinea* L. seedlings produced in a commercial nursery. Annals of Forest Science 62: 817-822.
- Schmid, V. 2006. Entwicklung molekularer methoden für ein schnelles und kostengünstiges monitoring der inokulation von forstpflanzen mit ektomykorrhizasymbionten. University of Innsbruck PhD dissertation, Innsbruck, Austria.
- Schoettle, A.W. and Sniezko, R.A. 2007. Proactive intervention to sustain high-elevation pine ecosystems threatened by white pine blister rust. Journal of Forest Research 12: 327-336.

- Schwanke, R. and Smith, C. 2010. Imitating lightning strikes for whitebark pine restoration. Whitebark Pine Ecosystem Foundation. Nutcracker Notes 19: 12.
- Schwandt, J. 2006. Whitebark pine in peril: a case for restoration. U.S. Department of Agriculture, Forest Service, R1-06-28. Missoula, MT.
- Schwandt, J., Chadwick, K., and Kearns, H. 2011. Whitebark pine direct seeding trials in the Pacific Northwest. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Schwandt, J., Tomback, D.F., Keane, R.E., McCaughey, W.W., and Kearns, H. 2007. First year results of a whitebark pine seed planting trial near Baker City, OR. USDA Forest Service R6-NR-FHP-2007-01.
- Scott, G.L., McCaughey, W.W., and Izlar, K. 2011. Guidelines for whitebark pine planting prescriptions. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Six, D. and Adams, J. 2007. White pine blister rust severity and selection of individual whitebark pine by the mountain pine beetle (*Coleoptera: Curculionidae, Scolytinae*) Journal of Entomological Science 42(3): 345-353.
- Smith, C.M. 2012. [Personal Communication]. Park Ecologist, Waterton-Lakes National Park, Alberta, Canada.
- Smith, C.M., Shepherd, B., Gillies, C., and Stuart-Smith, J. 2011. Re-measurement of whitebark pine infection and mortality in the Canadian Rockies. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Smith, C.M., Wilson, B., Rasheed, S., Walker, R.C., Carolin, T., and Shepard, B. 2008. Whitebark pine and white pine blister rust in the Rocky Mountains of Canada and Northern Montana. Canadian Journal of Forest Research 5: 982-995.

- Smith, S.E. and Read, D.J. 2008. Mycorrhizal symbiosis, 3rd edition. Academic Press, Inc., San Diego, CA.
- Snieszko, R.A. 2006. Resistance breeding against non-native pathogens in forest trees – current successes in North America. *Journal of Plant Pathology* 28: 270-279.
- Snieszko, R.A., Mahalovich, M.F., Schoettle, A.W., and Vogler, D.R. 2011. Past and current investigation of the genetic resistance to *Cronartium ribicola* in high-elevation five-needle pines. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Tedersoo, L., May, T.W., and Smith, M.E. 2009. Ectomycorrhizal lifestyle in fungi: global diversity, distribution, and evolution of phylogenetic lineages. *Mycorrhiza* 20: 217-263.
- Tomback, D.F. 1982. Dispersal of whitebark pine seeds by Clark's Nutcracker: a mutualism hypothesis. *Journal of Animal Ecology* 51: 451-467.
- Tomback, D.F. 2001b. Clark's nutcracker: agent of regeneration. Pp. 89-104. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds.). *Whitebark pine communities, ecology and restoration*. Island Press, Washington, DC.
- Tomback, D.F., Arno, S.F., and Keane, R.E. 2001a. *Whitebark pine communities: ecology and restoration*. Island Press, Washington, DC. 440 p.
- Tomback, D.F., Anderies, A.J., Carsey, K.S., Powell, M.L., and Mellmann-Brown, S. 2001c. Delayed seed germination in whitebark pine and regeneration patterns following the Yellowstone fires. *Ecology*. 82(9): 2587-2600.
- Tomback, D.F. and Achuff, P. 2010. Blister rust and western forest biodiversity: ecology, values, and outlook for white pines. *Forest Pathology* 40: 186-225.
- Trusty, P. 2009. Impact of severe fire on ectomycorrhizal fungi of whitebark pine seedlings. Montana State University Masters Thesis, Bozeman, MT.
- Uchytel, R.J. 1992. *Salix glauca*. In: Fire Effects Information System. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station, Fire Sciences Laboratory (Producer). [Online] Available: <http://www.fs.fed.us/database/feis/> [2011, July 6].

- Van Der Heijden, M.G.A. 2002. Arbuscular mycorrhizal fungi as a determinant of plant diversity: in search for underlying mechanisms and general principles. Pp. 243-265. In: van der Heijden, M.G.A., Sanders, I.R., (eds.). Mycorrhizal ecology. Springer-Verlag, Berlin, Germany.
- Van Der Heijden, M.G.A., Klironomos, J.N., Ursic, M., Moutoglis, P., Streitwolf-Engel, R., Boller, T., Wiemken, A. and Sanders, I.R. 1998. Mycorrhizal fungal diversity determines plant biodiversity, ecosystem variability and productivity. *Nature* 396: 69-72.
- Weisleitner, H. 2008. [Personal Communication to Dr. Cathy Cripps]. Federal Nursery of Austria, Innsbruck, Austria.
- Wilkinson, D.M. 2001. Mycorrhizal evolution. *Trend in Ecology and Evolution* 16: 64-65.
- [UNEP, WCMC] World Conservation Monitoring Centre. 2011. Protected Areas and World Heritage: Waterton-Glacier International Peace Park Alberta, Canada & Montana, United States of America. 13 p.

CHAPTER 2

LOW NITROGEN FERTILIZER AS A STRATEGY TO MAINTAIN
ECTOMYCORRHIZAL COLONIZATION OF INOCULATED
WHITEBARK PINE SEEDLINGS
BEFORE OUT-PLANTING

Introduction

Whitebark pine (WBP) (*Pinus albicaulis* Engelm.) is North America's only native stone pine and is limited to upper sub-alpine and timberline portions of Western North America (Tomback et al 2001). It is a "keystone" and "foundation" species that plays a pivotal role in supporting biological diversity and providing environmental stability in these high elevation communities (Ellison et al. 2005, Lantz 2010). Whitebark pine forests have been reduced to a fraction of their former range by white pine blister rust (*Cronartium ribicola* J.C. Fisch.), mountain pine beetle (*Dendroctonus ponderosae* Hopkins) infestation and fire suppression (Schwandt 2006, Keane and Parsons 2010, Logan et al. 2010, Tomback and Achuff 2010). The U.S. Fish and Wildlife Service has stated that the threats to whitebark pine populations are 'imminent' and 'of high magnitude' and that this species is in danger of extinction (Nicholas and Katzenberger 2011). As of July 18, 2011 the U.S. Fish and Wildlife Service determined that whitebark pine was warranted for protection under the Endangered Species Act; it is currently precluded due to funding and priority constraints (Nicholas and Katzenberger 2011).

The importance of whitebark pine in high elevation ecosystems within its range has long been recognized and restoration efforts have been on-going since the 1990's (McCaughey 1990). Some efforts have focused on the conservation and restoration of whitebark pine habitat through prescribed fire, silviculture techniques, and protection from mountain pine beetle attack (Bentz et al. 2005, Keane and Parsons 2010). Other efforts have focused on reforestation through the out-planting of nursery grown seedlings, which has led to the development of rust-resistance screening techniques, cone collection methods, seed germination methods, and improved out-planting guidelines (Burr et al. 2001, Mahalovich and Hoff 2001, Mahalovich and Dickerson 2004, Mahalovich et al. 2006, Murray 2007, McCaughey et al. 2009). The Coeur d'Alene nursery has developed a standard protocol for growing high-quality potentially rust-resistant whitebark pine seedlings. Currently, it is common practice to combine habitat restoration with the out-planting of potentially rust-resistant nursery-grown seedlings. Nonetheless, whitebark pine populations continue to decline and both natural seedling regeneration and out-planted seedling survival rates remain low (Perkins 2004, Izlar 2007, Keane and Parsons 2010). Although restoration of whitebark pine forests continues to be a difficult task, concerned citizens, forest managers, and scientists are exploring new ways to maintain and restore these valuable forests. One previously overlooked tool is the use of native ectomycorrhizal fungi to enhance out-planted seedling survival.

Ectomycorrhizal Fungi and Restoration

Ectomycorrhizal fungi enhance the survival of woody plants through increased nutrient uptake, increased drought tolerance, and protection from harmful soil biota

(Cripps 2004, Smith and Read 2008). The inoculation of container grown conifers with ectomycorrhizal fungi during greenhouse production is recommended for seedlings to be used in restoration, especially in areas devoid of native ectomycorrhizal fungi (Wiensczyk et al. 2002). Inoculation of seedlings with ectomycorrhizal fungi can enhance survival in the field (Steinfeld et al. 2003, Quoreshi et al. 2009, Lehto and Zwiazek 2011) and reduce fertilizer, irrigation, and pesticide expense while protecting against root pathogens in the nursery (Whipps 2004). However, results vary depending on the host plant, fungi involved, soil type and site conditions. The use of ectomycorrhizal fungi to enhance seedling performance has been employed with other container grown pines, but it has not been used in whitebark pine production (Eggleston 2010).

The European stone pine is a close relative of whitebark pine and provides similar ecological services in high elevation communities in Europe and Asia (McCune 1988, Tomback and Linhart 1990, Gernandt et al. 2005). Over sixty years ago the Federal Nursery in Austria began inoculating European stone pine (*Pinus cembra* L.) seedlings with native ectomycorrhizal fungi to assist in reforestation of high elevation avalanche paths (Heumader 1992, Weisleitner 2008). The Austrian nursery developed specific cultivation practices aimed at the long-term production of seedlings colonized with native ectomycorrhizal fungi that are adapted to high elevation conditions. Seedling transplant beds were originally inoculated in the 1950's with mycelium from three native species of suilloid fungi adapted to high elevations: *Suillus placidus* (Bonord.) Singer, *Suillus plorans* (Rolland) Kuntze, and *Suillus sibiricus* Singer. The long-term cultivation of cembran pines has resulted in nursery soil that is now full of native mycelium and re-

inoculation has not been necessary for many years (Heumader 1992). Inoculation, along with improved silviculture techniques, enhanced seedling survival in the field 50-90%. The planted cembran pines are now over fifty years old and are flourishing on the Austrian slopes (Peintner 2012).

Ectomycorrhizal Fungi and Whitebark Pine

Whitebark pine seedlings used in reforestation in the Rocky Mountain region are grown almost exclusively at the Coeur d'Alene Forest Service Nursery in Idaho; currently they are not inoculated with native ectomycorrhizal fungi (Eggleston 2010). Commercially available mycorrhizal inoculum is not appropriate for whitebark pine seedlings and introduction of alien fungi into these sensitive forest systems is not recommended (Schwartz et al. 2006, Keane et al. 2012).

About 50 species of native ectomycorrhizal fungi have been found to associate with whitebark pine forests in the Rocky Mountain region, including some of the same or related species that are used by the Austrian Federal nursery (Mohatt 2006, Cripps et al. 2008, Mohatt et al. 2008). Over twenty-five strains of these native ectomycorrhizal fungi have been screened as potential inoculum for whitebark pine seedlings (Cripps and Grimme 2011). As with the European stone pine, suilloid fungi adapted to high elevations and specific to five-needle pines successfully colonized whitebark pine seedlings under nursery conditions (Cripps and Grimme 2011, Cripps and Antibus 2011). In these early screening trials, *Suillus sibiricus* out-performed other ectomycorrhizal fungi in the formation of ectomycorrhizae on whitebark pine seedling roots in the greenhouse; this species was subsequently selected for further evaluation as fungal

inoculum (Cripps and Grimme 2011). *Rhizopogon* species are commonly used in commercial inoculum applied in U.S. nurseries (Amaranthus 2002), however the native species of *Rhizopogon* tested with whitebark pine lagged significantly behind *Suillus* species in mycorrhizal formation in the greenhouse and they are not as host specific (Cripps and Grimme 2011).

Fungal species and isolates vary in their ability to colonize root systems and in their tolerance to particular soil substrates and fertilizer regimes (Trappe 1977, Marx 1980, Khasa et al. 2001). Some nursery practices are not conducive to root colonization by *native* ectomycorrhizal fungi (Cordell and Marx 1994, Quoreshi et al. 2009). Greenhouse conditions often result in colonization by an array of “nursery” fungi adapted to regular watering and fertilizer regimes (Danielson 1991, Khasa et al. 2009), however ‘nursery’ fungi provide minimal long-term benefits to seedlings following out-planting (Danielson 1991, Bruns et al. 2002). Furthermore, there is evidence that host specific high elevation adapted fungi do promote the survival of 5-needle pines (Heumader 1992, Weisleitner 2008, Peintner 2012).

If inoculation of whitebark pine seedlings with native ectomycorrhizal fungi is to be an effective nursery technique, cultivation methods specific to whitebark pine seedlings and their associated native ectomycorrhizal fungi need to be developed (Cripps and Grimme 2011). The effects of inoculum type, soil substrate and fertilizer regime on the colonization of whitebark pine by native ectomycorrhizal fungi have been assessed to a limited degree in preliminary trials (Cripps and Grimme 2001). Additional factors such as watering, inoculation method, slurry type, and container style also need to be explored

to optimize the timely formation of ectomycorrhizae under greenhouse conditions (Landis et al. 1989b, Khasa et al. 2009, Quoreshi et al. 2009, Repáč 2011).

In limited, preliminary trials, colonization by native *Suillus* species was shown to be more prolific when whitebark pine seedlings were inoculated with spore slurries made from fresh sporocarps in comparison to the addition of a mycelial inoculum (Cripps and Grimme 2011). Other studies have found similar results (Gonzalez-Ochoa et al. 2003, Brundrett et al. 2005). However, fresh sporocarps are needed to make fresh spore slurries. High elevation sites are prone to drought which can often prevent fungal fruiting which makes finding fresh sporocarps for inoculum difficult in some years. Also, inoculation is usually necessary in the spring before the fall fruiting season occurs. An alternative is to make spore slurries from dried sporocarps, which could potentially be stored for long-term periods before use in inoculum (Parladé et al. 1996, Rincón, et al. 2005, Chen et al. 2006). However, it is necessary to test the effectiveness of slurries made with dried sporocarps of ectomycorrhizal fungi associated with whitebark pine before large scale production of spore slurries.

Soil substrates have been shown to significantly affect mycorrhizal colonization of nursery grown seedlings (Shaw et al. 1981, Brundrett et al. 1996, Rincon et al. 2005, Oliveira et al. 2010). The effect of different substrates on the mycorrhizal colonization of containerized whitebark pine seedlings has been evaluated to a limited degree (Cripps and Grimme 2011). Substrates that are conducive to colonization of whitebark pine seedlings by native suilloid fungi include: peat:vermiculite, peat:composted bark, and a soil mixture containing MSU soil (loam soil, Canadian Sphagnum peat moss, and washed

concrete sand (1:1:1), with an AquaGro 2000 G wetting agent applied at a rate of one lb./cubic yd) vermiculite and peat in a 2:2:1 by volume ratio (termed Soil Mix 3) (Cripps and Grimme 2011). Substrates containing peat:un-composted bark or Sunshine Mix (a soil-less blend of Canadian Sphagnum peat moss and horticultural grade Perlite, buffered with Dolomitic lime, and containing a starter fertilizer charge and wetting agent) suppress mycorrhizal colonization of whitebark pine seedlings by native fungi, possibly due to changes in pH and/or the narrow pH range of certain isolates of suilloid fungi (Cripps and Antibus 2011, Cripps and Grimme 2011). A high pH has been shown to prevent spore germination in spore slurries (Castellano et al. 1989).

Fertilization can also inhibit mycorrhizal colonization in the nursery (Shaw et al. 1981, Castellano et al. 1985, Rincón et al. 2005, Smith and Read 2008). High NPK fertilization levels can suppress colonization by native mycorrhizal fungi while promoting colonization by “nursery” fungi (mainly E-strain and *Thelephora sp.*) in whitebark pine seedlings (Cripps and Grimme 2011). However, reduced or altered fertilizer regimes can allow for ectomycorrhizal colonization of nursery grown seedlings by native fungi (Gagnon et al. 1988, Quoreshi et al. 1998, Khasa et al. 2001, Meikle and Amaranthus 2008). Khasa et al. (2001) found that reducing fertilization by 33% allowed for both mycorrhization and good seedling growth in a variety of conifer seedlings. For black spruce seedlings (*Picea mariana* (Mill.) Britton, Sterns & Poggenb.) in particular, exponential fertilization, achieved by progressively increasing nutrient applications so they correspond with relative seedling growth rates, did support ectomycorrhizal colonization in the nursery (Quoreshi et al. 1998). In addition, whitebark pine seedlings

that were fertilized for 16 months and inoculated one month after fertilization was stopped, were well-colonized in the few months that preceded out-planting (Cripps and Grimme 2011). These results suggest that it may be possible to develop a fertilization regime specific for whitebark pine seedlings and their associated native ectomycorrhizal associates.

Research Objectives

The main objective of this experiment is to determine whether WBP seedlings inoculated with native ectomycorrhizal fungi can maintain this mutualism when a low nitrogen (N) fertilizer is subsequently applied in the greenhouse. It was hypothesized that a low N fertilizer would be less likely to interfere with the established mutualism since *S. sibiricus* (and other suilloid fungi) are involved primarily with enhancing N uptake in plants (Keller 1996, Cripps and Antibus 2011). This is analogous to the use of a low phosphorous fertilizer for arbuscular fungi which allows mycorrhizal colonization in the greenhouse; this ultimately enhanced survival of several plant species in field trials (Meikle and Amaranthus 2008). If successful with WBP, this would allow fertilization to continue before out-planting, thus enhancing the stature and health of seedlings by two strategies, fertilization and mycorrhizal inoculation. This approach has application for other container-grown conifers as well.

In addition, three other variables were examined for optimizing mycorrhizal formation on WBP seedlings in the greenhouse: two types of spore slurries (fresh and dried), two methods of inoculation (injection or drip), and long and short containers were compared in the experiment. It is often difficult to plant seedlings grown in long

containers in the shallow, rocky soils found at the high elevation restoration sites and so container length was included as a variable.

Materials and Methods

Seedlings

Approximately 500 fourteen-month-old rust-resistant WBP seedlings, grown from seeds collected at Preston Park (48°43'45"N, 113°39'03"W, 8907' elevation) in Glacier National Park, were obtained from the USDA Forest Service Nursery in Coeur d'Alene, Idaho. At the Idaho nursery, seeds were sown (February 2009) in long Ray Leach cone-tainers (3.8 cm x 21 cm) and grown under standard nursery conditions in a substrate mix of Canadian Sphagnum peat moss and composted bark (7:3). Seedlings were fertilized every 8-12 days with a 4-25-35 NPK fertilizer and STEM (soluble trace element mix, Peter's Professional) micronutrients (Eggleston 2010). When seedlings were moved to the Plant Growth Center (Montana State University-MSU, Bozeman, MT) fertilization was stopped and seedlings were grown under standard greenhouse conditions (22 °C day and 18 °C night temperatures, 16-hour photoperiod). Seedlings were re-planted in Soil Mix 3 which is composed of MSU mix, vermiculite, and sifted Canadian Sphagnum peat moss (2:2:1) with an average pH of 5.66. The MSU mix component contains loam soil, Canadian Sphagnum peat moss, and washed concrete sand (1:1:1), with an AquaGro 2000 G wetting agent applied at a rate of one lb./cubic yd. The MSU mix is steam pasteurized at 70 °C for 60 minutes before addition to Soil Mix 3. Approximately half (N = 254) of the WBP seedlings were re-planted in short Ray Leach cone-tainers (3.8 cm x

14 cm, 115 ml) and the other half (N = 260) were re-planted in the original long Ray Leach cone-tainers (3.8 cm x 21 cm, 164 ml) from the Idaho Nursery (Figure 2.1).

Spore Slurry Types

Sporocarps (mushrooms) of one species, *Suillus sibiricus*, were collected in WBP forests in Montana in September 2010. Two large collections were used for slurries, one from Gallatin (2800 m elevation) and the other from Flathead County (1000 m elevation). Two inoculum types were made: one slurry was made from fresh sporocarps and the other from dried sporocarps (Figure 2.1). Sporocarps for the dried slurry were dried on a dehydrator and stored in plastic bags for one month prior to use. The fresh slurry was stored in the refrigerator for one month until it was used. To make the slurries, sporocarps were cleaned and the hymenium (pore surface) removed, cut into small pieces and ground for approximately 1 minute in a coffee grinder with 10 ml of sterile distilled water. The resulting ground-up material was strained into approximately 400 ml of sterile distilled water and stored in glass bottles in the refrigerator for one month. The spore content of each slurry was counted using a hemocytometer and further diluted to a spore count of approximately 1×10^6 spores/ml.



Figure 2.1. Top: Fresh and dried sporocarps of *S. sibiricus*, inject and drip inoculation methods, and long and short containers. Bottom: Whitebark pine seedlings in the greenhouse.

Fertilizer Regimes

WBP seedlings were not fertilized until six months after inoculation and just after vernalization. To simulate a natural vernalization period, seedlings were placed in a cold room at approximately 4 °C for 2 months.

After vernalization, whitebark pine seedlings were fertilized for 22 weeks with Phosgard 4-25-15 liquid NPK fertilizer from JH Biotech, Inc, a low N fertilizer. All

seedlings were fertilized from the same solution of 13.1-35.8-40.8 parts per million of NPK, respectively (13.1 ppm N, 81.9 ppm P₂O₅, 49.1 ppm K₂O). Fertilizer was applied during normal watering periods and seedlings were soaked to saturation. All seedlings were watered three times per week. Fertilizer was applied once a week for seedlings in the ‘high’ treatment and once every other week to seedlings in the ‘low’ treatment. Seedlings in the ‘none’ category were also watered three times per week but fertilizer was not applied to seedlings.

Inoculation Methods

Approximately half of the 21-month-old WBP seedlings were inoculated (November 2010) with either the ‘fresh’ or ‘dry’ slurry by one of two methods. For the ‘drip method’ of inoculation, seedlings were removed from containers and 5 ml of spore slurry was dripped onto the exposed roots of the seedlings using a glass pipette and seedlings were placed back in the containers. For the ‘inject method’ of inoculation, 5 ml of spore slurry was injected directly onto the growing medium of containerized seedlings using an Allflex 50 ml repeat syringe commonly used for multiple dosing of livestock (Figure 2.1). Each seedling received approximately 5×10^6 spores with either inoculation method.

Assessment of Mycorrhizal Colonization

WBP seedlings were assessed by non-destructive techniques (visual observation) for mycorrhizal colonization three times throughout the experiment (Figure 2.2). Assessments 1 and 2 (January and March 2011, respectively) were used to determine

whether inoculum was viable and whether mycorrhizal colonization was occurring prior to initiating the fertilizer treatments.

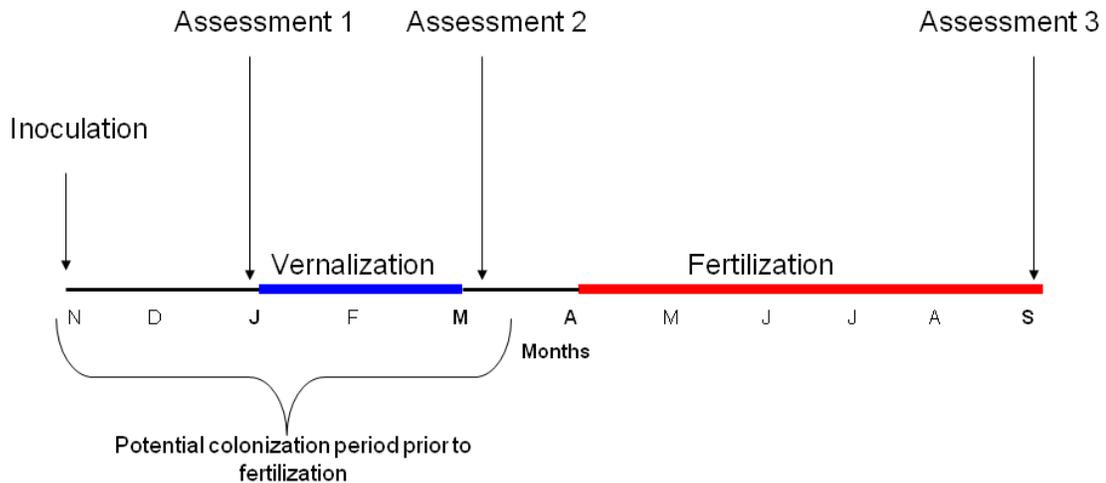


Figure 2.2: Timeline of inoculation and assessments. The experiment started in November which is designated as Month 1 in diagram. The three assessments are of mycorrhizal colonization of seedlings.

The percent of WBP seedlings (frequency) with suilloid ectomycorrhizae was assessed for each treatment combination of slurry type, inoculation method, and container size. For Assessment 1, approximately half of the seedlings (203) as determined by a random number generator were assessed for the presence/absence of suilloid ectomycorrhizae. All of the seedlings were examined for presence/absence of suilloid ectomycorrhizae in Assessments 2 and 3. It was necessary to use non-destructive methods since seedlings were to be out-planted.

At the end of the fertilization treatments, (Assessment 3, November 2011) both frequency (percent of seedlings with suilloid ectomycorrhizae) and average abundance of ectomycorrhizal colonization (percent of roots covered) across all treatments were

evaluated. To assess abundance, roots of each seedling were immersed in distilled water and soil particles removed by gentle agitation. Roots were then assessed under a dissecting scope to determine percent colonization of suilloid ectomycorrhizae. Suilloid ectomycorrhizae are recognized by characteristics typical of *S. sibiricus* (Figure 2.3), in particular the presence of a white plectenchymatous mantle, a coralloid branching pattern, weak rhizomorphs, and a lack of clamp connections (Treu 1990). Periodic checks under the compound microscope helped confirm identification.



Figure 2.3. Example of suilloid fungi colonizing the roots of a whitebark pine seedling.

In addition, plant parameters that included wet biomass, root volume, needle color, canopy development, and root development were measured. It was necessary to use non-destructive measures, since seedlings were to be transplanted. Biomass was measured by weighing bare-root seedlings rinsed of soil. Root volume was assessed by measuring the volume of water displaced by each root system after it was

rinsed of soil. Needle color, canopy development and root development were measured using a scale system. Needle color was recorded as dead, >50% necrosis, <50% necrosis, chlorotic, or green. Canopy development and root development were recorded as poor, moderate, or robust.

Experimental Design

A full factorial design with three fertilizer treatments, two container lengths, two slurry types and two inoculation methods for a total of 24 treatments was used. N values for each combination of treatments are shown in Table 1. WBP seedlings were placed in container trays by treatment and individual trays were rotated each month to compensate for variation in light and other greenhouse conditions. Prior to setting up the experiment, seedlings were checked for colonization by nursery fungi and those that appeared to be colonized were removed. At the end of the experiment only living seedlings were assessed since mortality was minimal and random across treatments.

Table 2.1: Number of living whitebark pine seedlings in each treatment at completion of experiment.

Container Length	Slurry Type	Inoculation method	High Fertilizer	Low Fertilizer	No Fertilizer
Long	Fresh	Drip	18	17	5
		Inject	21	16	8
	Dried	Drip	13	14	7
		Inject	18	18	8
Short	Fresh	Drip	12	18	5
		Inject	22	19	9
	Dried	Drip	9	15	6
		Inject	16	13	6

Statistical Analysis

The mean percent ectomycorrhizal colonization (abundance) of inoculated WBP seedlings was analyzed using a 4-way analysis of variance (ANOVA) in SPSS General Linear Model Univariate in SPSS (IBM 2011) that included four main effects (3 fertilizer groups, 2 container lengths, 2 slurry types and 2 inoculation methods) and all possible interactions. Levine's test of equal variance and box plot analysis indicated that the assumption of equal variance was not met and data were transformed (square root) prior to analysis. Any statistical differences detected among treatment combinations were analyzed further using pairwise comparisons. Means for each fertilizer treatment were separated by Tukey's honest significance difference (HSD); ($\alpha = 0.05$).

Results

Frequency of Mycorrhizal Colonization: Assessments 1, 2, and 3

Three months after inoculation (Assessment 1), 91% of WBP seedlings inoculated with slurry made from fresh sporocarps showed signs of ectomycorrhizal colonization whereas this was true for only 29% of seedlings inoculated with slurry made from dried sporocarps (Figure 2.4). Five months after inoculation and following vernalization (Assessment 2), 80% of seedlings inoculated with 'fresh' slurry showed signs of ectomycorrhizal colonization whereas only 34% of seedlings inoculated with 'dried' slurry had ectomycorrhizae (Figure 2.4). However, at the end of the fertilizer treatment (Assessment 3), 92% of the seedlings inoculated with 'dried' slurry had ectomycorrhizae and 97% of those inoculated with fresh slurry had ectomycorrhizae (Figure 2.4). Thus, a

high percentage of seedlings had suilloid ectomycorrhizae at the completion of the experiment.

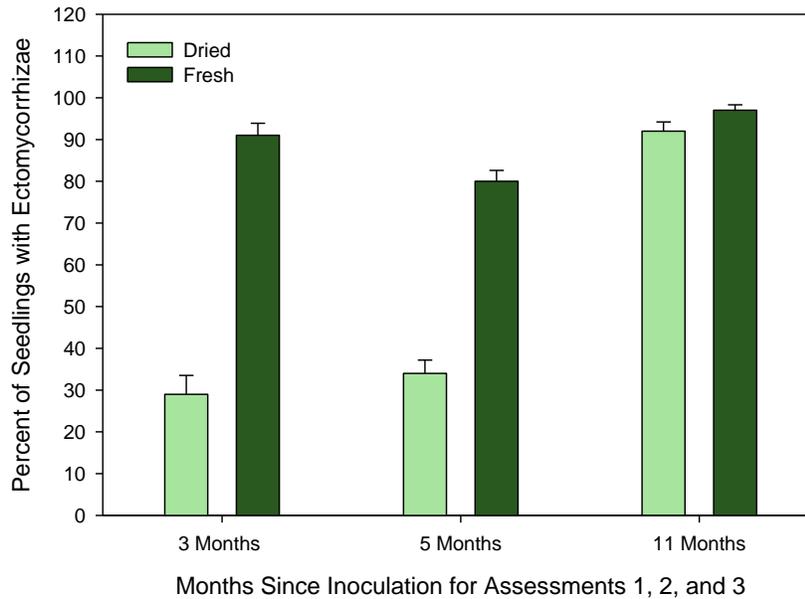


Figure 2.4: Frequency of seedlings with ectomycorrhizae for Assessments 1, 2, and 3. Graphs include 95% confidence bars.

Abundance of Ectomycorrhizal Colonization: Assessment 3

Results from the ANOVA are shown in Table 2.2 and means for each treatment combination are in Table 2.3. Ectomycorrhizal colonization of WBP seedlings was maintained at some level under both low and high fertilizer regimes and on seedlings that were not fertilized (Figure 2.4). However, significant effects were observed for fertilizer treatment ($p = <0.001$), container length ($p = <0.001$), and for three interactions: fertilizer x slurry ($p = 0.049$); container x slurry ($p = 0.006$); and fertilizer x container x slurry ($p=0.002$); (Table 2.2).

Table 2.2: Four-way Analysis of Variance table for ectomycorrhizal colonization (square root transformed) as a function of fertilizer group, container length, slurry type, inoculation method and all possible interactions. Significant effects are shown in bold.

Source	Type III SS	df	F-stat	Sig.
Intercept	7264.756	1	1696.21	0.854
Fertilizer group	172.613	2	20.151	0.000
Container length	100.406	1	23.443	0.000
Slurry type	11.795	1	2.754	0.098
Inoculation method	11.655	1	2.721	0.100
Fertilizer group* Slurry type	26.131	2	3.051	0.049
Fertilizer group* Container length	4.766	2	0.556	0.574
Fertilizer group* Inoculation method	1.557	2	0.182	0.834
Container length * Slurry type	32.738	1	7.644	0.006
Container length* Inoculation method	0.135	1	0.032	0.859
Slurry type* Inoculation method	0.011	1	0.002	0.960
Fertilizer group* Container length* Slurry type	53.031	2	6.191	0.002
Fertilizer group* Container length* Inoculation method	7.195	2	0.840	0.433
Fertilizer group* Slurry type* Inoculation method	15.440	2	1.802	0.167
Container length* Slurry type* Inoculation method	1.111	1	0.260	0.611
Fertilizer group* Container length* Slurry type* Inoculation method	6.027	2	0.704	0.496

Table 2.3. Effects of fertilizer treatment, container length, slurry type, and inoculation method on the mean abundance of ectomycorrhizal colonization of whitebark pine seedlings by the native fungus *Suillus sibiricus* in the greenhouse.

Fertilizer Treatment	Container length	Slurry type	Inoculation method	Mean (%)	Std. Error
High	Long	Dried	Drip	22.31	5.66
			Inject	23.94	4.81
		Fresh	Drip	26.78	4.81
			Inject	41.91	4.45
	Short	Dried	Drip	20.00	6.80
			Inject	13.88	5.10
		Fresh	Drip	20.83	5.89
			Inject	23.64	4.35
Low	Long	Dried	Drip	24.64	5.46
			Inject	43.61	4.81
		Fresh	Drip	39.41	4.95
			Inject	46.88	5.10
	Short	Dried	Drip	19.00	5.27
			Inject	23.85	5.66
		Fresh	Drip	26.17	4.81
			Inject	30.79	4.69
None	Long	Dried	Drip	41.43	7.72
			Inject	50.00	7.22
		Fresh	Drip	74.00	9.13
			Inject	61.25	7.22
	Short	Dried	Drip	48.33	8.33
			Inject	60.00	8.33
		Fresh	Drip	16.00	9.13
			Inject	26.67	6.80

The abundance of ectomycorrhizal colonization on WBP seedlings differed among fertilizer treatments when averaged across container length, slurry type, and inoculation method ($f_{2, 289} = 20.15$, $p < 0.001$); (Figure 2.5). Post hoc comparisons indicated that the mean abundance of mycorrhizal colonization differed among all fertilizer comparisons (Table 2.4). Seedlings that were not fertilized had an average of 46.7% of the root system colonized, followed by seedlings in the low fertilizer group

(32.3%) and the high fertilizer group (25.2%). However, differences in mycorrhizal colonization among fertilizer treatments were influenced by container length and slurry type ($f_{2, 289} = 6.19$, $p = 0.002$).

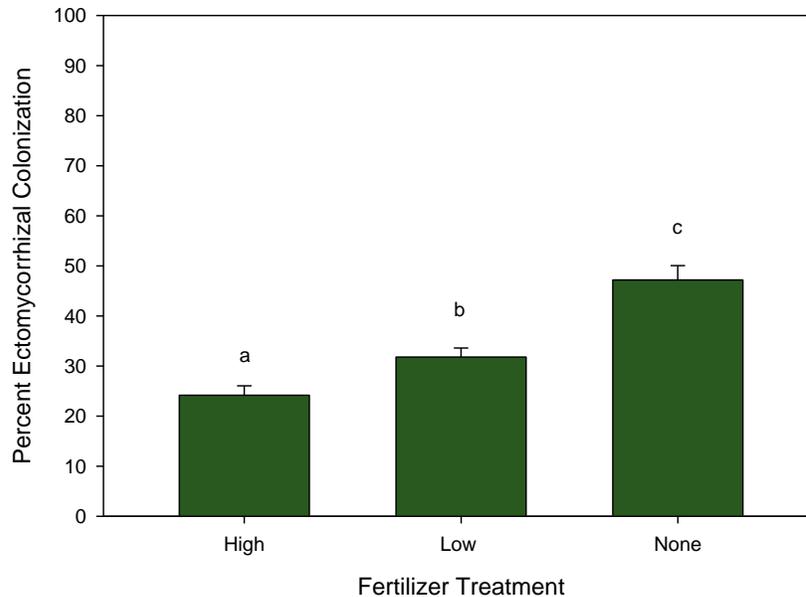


Figure 2.5: Mean percent ectomycorrhizal colonization of whitebark pine seedlings as a function of fertilizer treatment with 95% confidence intervals. Fertilizer treatment was 12.5-31-37.5 ppm NPK once a week for high and once every other week for low.

Table 2.4: Post hoc Tukey HSD test shows highly significant differences in ectomycorrhizal colonization among all comparisons for high, low and no fertilizer treatments of whitebark pine seedlings averaged across container length, slurry type, and inoculation treatment.

Fertilizer Treatments	None	Low	High
High	-	0.007	-
Low	0.001	-	-
None	-	-	<.0001

Overall, seedlings planted in long containers had a higher percentage of their roots systems colonized by suilloid fungi (41.3%) in comparison to seedlings planted in short containers (27.4%); (Figure 2.6). Root systems were assessed for *percent*

colonization, so these figures are proportional to the size of the root system. However, differences in colonization between container sizes were influenced by fertilizer treatment and slurry type ($f_{2, 289} = 6.19$, $p = 0.002$); (Table 2.2, Figure 2.7).

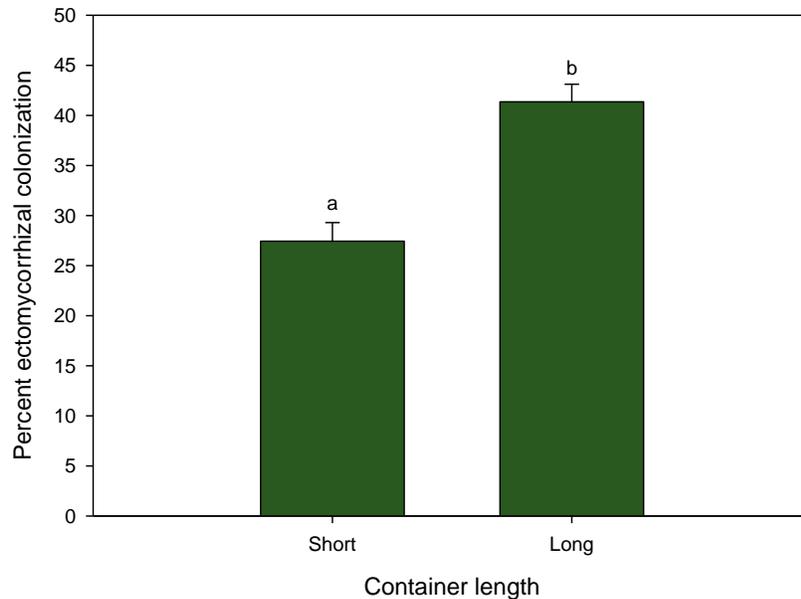


Figure 2.6 Mean percent ectomycorrhizal colonization of whitebark pine seedlings inoculated with *Suillus sibiricus* as a function of container length.

In general, seedlings in *long containers* followed the same pattern for abundance of mycorrhizal colonization for fertilizer treatments across both slurry types (Figure 2.7). This pattern was also true for seedlings in short containers inoculated with slurry made from dried sporocarps. However, seedlings in short containers inoculated with slurry made from fresh sporocarps showed a different pattern, with no statistical differences among fertilizer treatments for this group ($f_{2, 289} = 1.12$, $p = 0.36$). In general, colonization was low across the board for seedlings in short containers with one exception (unfertilized seedlings inoculated with dried slurry); (Figure 2.7).

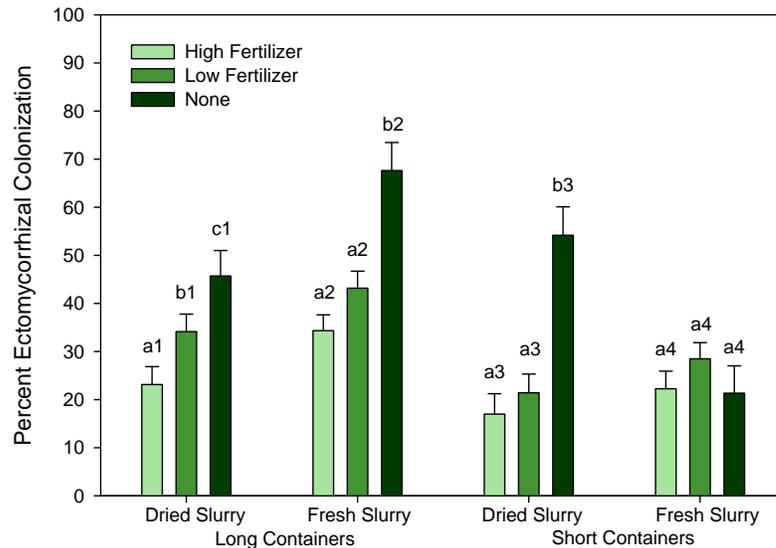


Figure 2.7: Mean percent ectomycorrhizal colonization of whitebark pine seedlings inoculated with *Suillus sibiricus* as a function of fertilizer group, container length and slurry type with 95% confidence bars. Data from inoculation method are pooled since no significant difference was observed.

When seedlings are planted in long containers and inoculated with slurry made from either fresh ($f_{2, 289} = 8.54$, $p < 0.001$) or dried sporocarps ($f_{2, 289} = 6.39$, $p = 0.002$), evidence indicates that there is a difference in mean colonization among fertilizer treatments. When inoculated with slurry made from fresh sporocarps, un-fertilized seedlings had the highest percent ectomycorrhizal colonization (67%) and were on average 33% more colonized than seedlings in the high fertilizer treatment and 24% more colonized than seedlings in the low fertilizer treatment, with associated confidence intervals of 20-46% and 11-38%, respectively. When inoculated with slurry made from dried sporocarps, un-fertilized seedlings still had the highest percent ectomycorrhizal colonization (45%) and were on average 22% more colonized than seedlings in the high

fertilizer treatment and 11% more colonized than seedlings in the low fertilizer treatment, with associated confidence intervals of 9-35% and 1-24%, respectively.

For seedlings planted in short containers, there is evidence of a difference in mean ectomycorrhizal colonization between fertilizer treatments when seedlings are inoculated with slurry made from dried sporocarps ($f_{2, 289} = 13.20$, $p < 0.001$); there is no evidence of a difference when seedlings are inoculated with slurry made from fresh sporocarps ($f_{2, 289} = 1.12$, $p = 0.36$). When inoculated with slurry made from dried sporocarps, unfertilized seedlings also had the highest percent ectomycorrhizal colonization (54%) and were on average 37% more colonized than seedlings in the high fertilizer treatment and 32% more colonized than seedlings in the low fertilizer treatment, with associated confidence intervals of 20-46% and 11-38%, respectively.

Overall, WBP seedlings planted in long-containers inoculated with fresh slurry that were not fertilized had the highest ectomycorrhizal colonization (67.6%) when averaged over inoculation method. Seedlings planted in short containers inoculated with dried slurry that received the highest fertilizer rate had the lowest ectomycorrhizal colonization (16.9%) when averaged over inoculation method. Data for 'inoculation method' are pooled on Figure 2.7 since no significant differences were observed.

Effects of Inoculation on Whitebark Pine Seedling Parameters

Statistical analyses of wet biomass and root volume were not conducted due to high variability in the data, but results are shown in table 2.5 and 2.6. Root volume and wet total biomass were graphed to show trends (Figure 2.8a and b, Figure 2.9a and b).

A trend between average root volume and fertilizer group is observed as shown in Figure 2.8a. Seedlings in the high and low fertilizer treatments had an average root volume that was 17% larger than seedlings that were not fertilized. A slight trend in average wet biomass by fertilizer group is observed in Figure 2.8b. The wet biomass of seedlings in the high and low fertilizer group was on average 13% larger than that of seedlings that were not fertilized.

Table 2.5: Mean and standard deviation for root volume (ml) of whitebark pine seedlings inoculated with *Suillus sibiricus* in each treatment.

Container Length	Slurry Type	Inoculation method	High Fertilizer	Low Fertilizer	No Fertilizer
Long	Fresh	Drip	5.64 ± 3.57	6.35 ± 2.17	5.20 ± 1.92
		Inject	6.33 ± 1.56	6.06 ± 2.08	5.63 ± 0.87
	Dried	Drip	5.73 ± 2.87	5.79 ± 1.12	4.71 ± 1.11
		Inject	6.81 ± 3.33	7.44 ± 1.75	4.88 ± 2.03
Short	Fresh	Drip	5.29 ± 1.63	4.06 ± 0.87	4.00 ± 1.58
		Inject	5.20 ± 1.76	3.95 ± 1.26	4.67 ± 1.22
	Dried	Drip	5.28 ± 2.71	6.40 ± 1.67	5.00 ± 0.32
		Inject	6.06 ± 2.66	5.38 ± 1.19	4.33 ± 1.03

Table 2.6: Mean and standard deviation for wet biomass (g) of whitebark pine seedlings inoculated with *Suillus sibiricus* in each treatment.

Container Length	Slurry Type	Inoculation method	High Fertilizer	Low Fertilizer	No Fertilizer
Long	Fresh	Drip	6.57 ± 2.89	7.60 ± 1.66	6.15 ± 0.99
		Inject	6.45 ± 1.16	6.26 ± 1.45	6.37 ± 1.09
	Dried	Drip	6.44 ± 2.41	6.43 ± 1.42	5.58 ± 1.56
		Inject	7.60 ± 2.89	7.69 ± 1.48	5.81 ± 1.32
Short	Fresh	Drip	6.21 ± 1.76	5.25 ± 1.01	5.42 ± 0.94
		Inject	6.01 ± 1.65	5.02 ± 1.16	5.4 ± 0.94
	Dried	Drip	6.56 ± 2.71	7.03 ± 1.83	5.59 ± 0.64
		Inject	7.17 ± 2.69	6.08 ± 1.38	5.73 ± 0.75

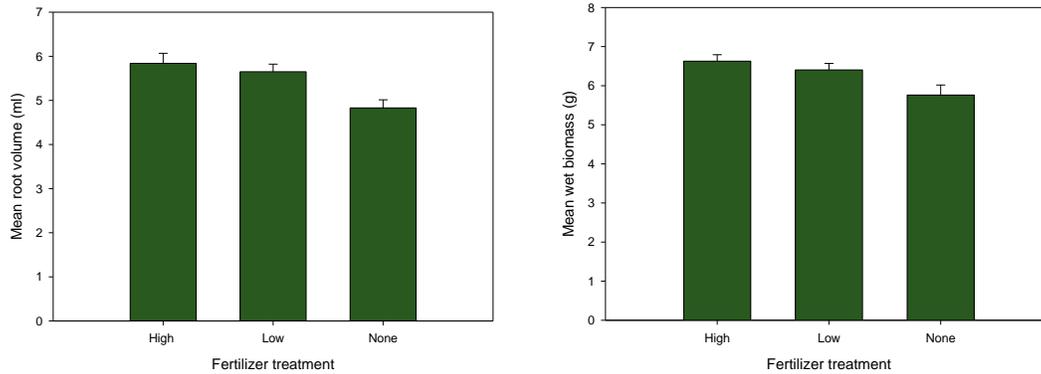


Figure 2.8: Seedling parameters for a) mean root volume (ml) of whitebark pine seedlings inoculated with *Suillus sibiricus* as a function of fertilizer treatment with 95% confidence bars, b) mean wet biomass (g) of whitebark pine seedlings inoculated with *Suillus sibiricus* as a function of fertilizer treatment with 95% confidence bars.

Container length appeared to affect both root volume and wet biomass. A comparison of average root volume and container size is observed in Figure 2.9a. Seedlings in long containers had an average root volume 18% larger than seedlings in short containers. Also, there was a slight trend for seedlings in long containers to have a higher average wet biomass (Figure 2.9b). Seedlings in long containers were on average 11% larger than seedlings in short containers.

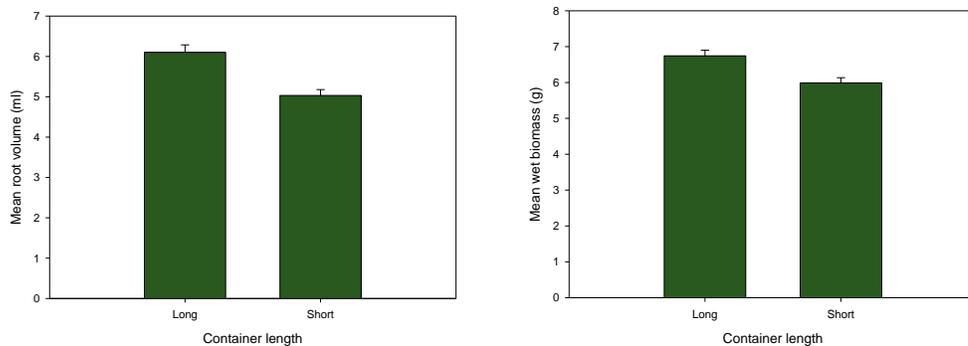


Figure 2.9: Parameters of whitebark pine seedlings inoculated with *Suillus sibiricus* as a function of container length for a) mean root volume (ml), b) mean wet biomass (g). Graphs include 95% confidence bars.

Root development, foliar development and needle color were also graphed to show trends although differences were minimal (Figure 2.10a, b, and c). Statistical analysis was not conducted as differences were negligible.

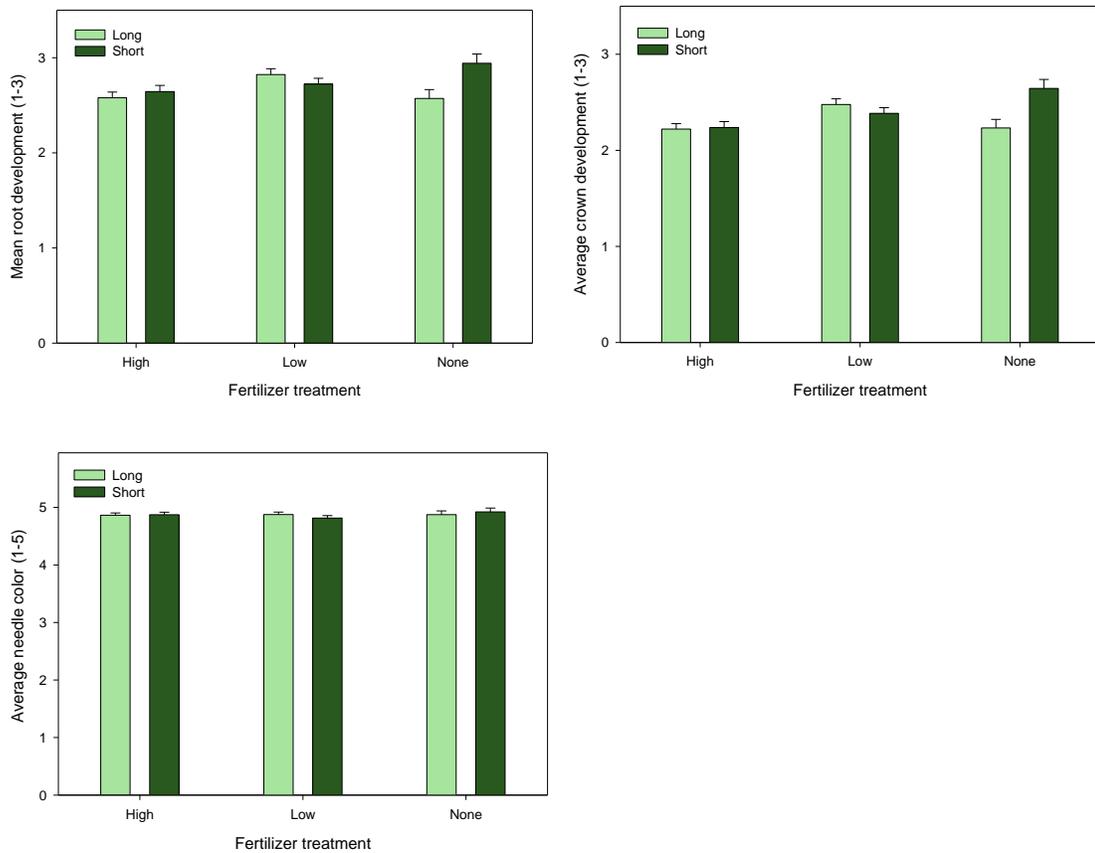


Figure 2.10: Effects of fertilizer treatment and container length on whitebark pine seedlings inoculated with *Suillus sibiricus* a) average root development (poor, moderate, robust), b) average foliar development (poor, moderate, robust), c) average needle color (dead, necrotic, partial necrosis, chlorotic, green). Graphs include 95% confidence bars.

Discussion

Fertilization

Numerous studies have shown that fertilization, especially with N, can reduce or eliminate ectomycorrhizal colonization of inoculated conifers in the greenhouse (Castellano et al. 1985, Gagnon et al. 1988, Arnebrant 1994, Smith and Read 2008). This is also true for WBP where weekly applications of 200 ppm of NPK (Scotts® Peters General Purpose 20-20-20) fertilizer resulted in negligible mycorrhizal colonization on seedling roots (Cripps and Grimme 2011). The low N fertilizer (Phosgard 4-25-15) used in this study did allow ectomycorrhizal colonization to be maintained on WBP seedlings across treatments, but colonization was lowest for heavy application, higher for light application and higher still for non-fertilized controls. The percent of the roots systems colonized are comparable with other greenhouse inoculations, where seedlings are not all typically colonized possibly due to differences in container conditions or genetic variation in seedlings (Brundrett et al. 2005). Low levels of ectomycorrhizal colonization are not functionally sufficient in some cases (Marx and Cordell 1988), although the minimum needed to enhance whitebark pine survival in the field is not known.

However, fertilization is necessary for the production of container-grown pines (Landis et al. 1989a) and virtually all whitebark pines are currently grown in containers. High fertilization levels can promote nursery fungi such as *Thelephora*, E-strain and root pathogens (Quoreshi et al. 2009); whitebark pine seedlings can host a variety of fungal pathogens in the greenhouse (James and Burr. 2000, Dumroese 2008). Increased substrate pH with fertilization and watering may be an inhibitory factor for most native

ectomycorrhizal fungi and particularly for spore germination (Castellano 1996, Rincón et al. 2005).

High N in particular can have detrimental effects on ectomycorrhizal formation on roots (Wallender and Nyland 1991, Brunner and Brodbeck 2001). In the greenhouse high N fertilizers can reduce ectomycorrhizal colonization on *Pinus contorta* Douglas ex Loudon (Ekwebelam and Reid 1983), *Pinus halepensis* Mill. (Diaz et al. 2010), and *Picea mariana* (Mill.) DuRoi (Gagnon et al. 1988) seedlings, to give a few examples. Many of the native fungi used in inoculations, including those used on WBP, are known to be involved with N acquisition, so results could be more applicable to these ectomycorrhizal fungi (Keller 1996).

Exponential fertilization can be more conducive to mycorrhizal colonization in the greenhouse and is achieved by progressively increasing nutrient application to correspond with seedling growth rates. Ectomycorrhizal colonization of *Picea mariana* was increased significantly in the greenhouse with exponential fertilization in comparison to conventional fertilization (Quoreshi and Timmer 1998). For the five-needle western white pine (*Pinus monticola* Rybd.), a lower nutrient application rate with exponential fertilization reduced overall fertilization by 45% compared to conventionally fertilized seedlings (Dumroese et al. 2005). These seedlings were not inoculated in the greenhouse, but the exponentially fertilized seedlings had higher ectomycorrhizal colonization the year following out-planting.

Nitrogen loading before out-planting can increase survival of container-grown conifers under some circumstances (Van Den Driessche 1987, Timmer 1996, Rikala et al.

2004). A more recent study reports that *Pinus halepensis* seedlings grew larger with added N, but that survival was unaffected by nitrogen rate, at least above a certain N threshold (Jackson et al. 2012). Inoculation with native ectomycorrhizal fungi may be an alternative to nutrient loading of conifer seedlings prior to out-planting. Foliar N can be enhanced by ectomycorrhizal fungi depending on the conifer species and fungal isolates involved (Gagnon et al. 1988, Chakravarty and Chatarpaul 1989, Heumader 1992, Quoreshi and Timmer 1998, Amaranthus et al. 2005, Rincón et al. 2005). For example, significant increases in N uptake have been reported for inoculated seedlings of *Pinus halepensis* (Rincón et al. 2007) and *Picea mariana* (Quoreshi and Timmer 1998, Gagnon et al. 1988) in comparison to non-inoculated controls. More applicably, European stone pine seedlings associated with *Suillus placidus* (Bonord.) Singer in the nursery had significantly higher foliar N compared to non-inoculated seedlings (Heumader 1992). Green manure is used to maintain this mycorrhizal association *in lieu* of chemical fertilizer and seedlings are planted in biodegradable pots (Heumader 1992). In the experiment reported here, foliar N was not assessed.

Ectomycorrhizal inoculation of nursery grown conifers has been shown to improve seedling survival in the field on sites where natural inoculum is lacking (Wiensczyk et al. 2002, Steinfeld et al. 2003, Parladé et al. 2004, Gagne et al. 2006, Menkis et al. 2007). In areas where native ectomycorrhizal fungi are present, nursery inoculation can benefit seedling survival by enhancing access to nutrients and water during the critical establishment period (Ortega et al. 2004, Quoreshi et al. 2009, Lehto and Zwiazek 2011). For example, survival of *Pinus ponderosa* P. Lawson & C. Lawson

seedlings planted on harsh, dry sites increased 30-56% when seedlings were inoculated with *Rhizopogon* species (Steinfeld et al. 2003). Similarly, 3.5 years after out-planting, survival of *Pinus pinea* L. seedlings was 20% higher for seedlings inoculated with *Rhizopogon roseolus* (Corda) Th. Fr. and this increase was enough to justify inoculation expenses (Parladé et al. 2004). Survival of European stone pine seedlings in Austria was increased 50-90% after inoculation with native ectomycorrhizal fungi in combination with intensive silviculture techniques (Moser 1956, Schmid 2006).

The ectomycorrhizal fungi used in inoculation of seedlings do not always persist after out-planting and can be replaced by indigenous fungi. However, the original inoculation can benefit seedling establishment by increasing a seedling's ability to rapidly access water and nutrients following out-planting (Ortega et al. 2004, Quoreshi et al. 2009, Lehto and Zwiazek 2011). The use of appropriate native fungi in the greenhouse may circumvent the need for fungal replacement in the field. In Austria 50 years after inoculation of European stone pine seedlings with three indigenous species of *Suillus*, including *S. sibiricus*, molecular techniques identified all three original *Suillus* species as still present and colonizing root systems (Schmid 2006).

Container Length

Container size can play an important role in shaping the morphology and physiology of seedling root systems. Long, narrow containers are typically recommended for growing native plants for restoration in order to reduce the effects of limiting soil moisture (Landis et al. 1990, Dominguez-Lerena et al. 2006, Landis et al. 2010b). However, it has proven difficult to plant WBP seedlings with long root systems in the

shallow, rocky soils often associated with high elevations sites. Shallow plantings can lead to frost heaving and increased out-planting mortality due to desiccation.

Seedlings planted in short containers had a smaller percentage of roots covered with suilloid ectomycorrhizae. However, in an effort to continue exploration of options for planting in shallow, rocky soils, it may be worth testing an intermediate container size that is either wider or longer than the short containers used in this experiment, such as Stuewe and Sons large dee-pots (6.4 cm x 17.8 cm, 444 ml). Container length should be considered an important factor even when seedlings are not inoculated. In one study, roots of WBP seedlings retained their long container shape for at least five years after out-planting (Trusty and Cripps 2011). This study showed that roots of non-inoculated nursery stock grown in long containers may not grow out into the native soil in a timely manner.

Slurry Type

Successful inoculation of containerized nursery grown seedlings has been achieved through the use of a variety of inoculum types (Castellano et al. 1985, Boyle and Robertson 1987, Repáč 1996a, 1996b, 2007, 2011). In a large experiment, spore slurries were more effective, less costly, and more efficient than mycelial suspensions overall (Brundrett et al. 2005). For example, Rincon and others (2007), found spore slurries of *Suillus collinitus* (Fr.) Kuntze to be a more effective mycorrhizal inoculum for *Pinus halepensis* seedlings compared to mycelial suspensions. For WBP, spore slurries made from fresh sporocarps were more effective than mycelial inoculum under greenhouse conditions in small trials (Cripps and Grimme 2011). In some cases the

viability of suilloid spore slurries has been maintained with refrigeration up to 3 years (Castellano and Molina 1989). However, viability declined in 90-180 days when slurries were frozen or refrigerated in another study (Torres and Honrubia 1994). Declines could reflect dormancy and not death, so that results may depend on the assessment method.

In the WBP experiment, the ‘inoculation potential’ of spore slurries made with dried sporocarps was compared to that of slurries made with fresh sporocarps to determine whether dried sporocarps that had been stored for a length of time could subsequently be used as inoculum. Mycorrhizal frequency, assessed prior to vernalization and fertilization, was lower when spore slurries were made from dried sporocarps. However, at the completion of the experiment, there was no practical difference in the abundance of suilloid mycorrhizae for the two types of inoculum. A latency period brought on by drying of the sporocarps or dormancy factors associated with suilloid spores could explain the lower colonization early on and perhaps dried spores need more time or vernalization to germinate (Aime and Miller 2002).

Results from this experiment suggest that dried fruiting bodies of *Suillus sibiricus* can be stored and subsequently used in spore slurry inoculation of five-needle pines. However, seedlings should be inoculated at least 5 months prior to out-planting to allow sufficient time for colonization and it is not known if vernalization is required. The use of dried sporocarps in inoculum could help alleviate timing conflicts between suilloid sporocarp production in nature (Aug.-Oct.) and inoculation time in the nursery (spring) and help remedy the problem of locating fruiting bodies in drought prone high elevation sites every year.

Inoculation Method

For each type of fungal inoculum, whether mycelial or spores, an array of effective application methods exist (Repáč 2011). For spore inoculum in particular, the most common inoculation method used is the application of spores suspended in water (spore slurry) to the seedling soil substrate through drenching, irrigation, or injection.

Inoculation and colonization of whitebark pine seedlings was previously successful using an Allflex 50mL repeat syringe to inject spore slurry into the soil surrounding seedlings (Cripps and Grimme 2011). In the current study, this injection method was compared to an alternative method where seedlings were carefully slipped out of the containers and spore slurry dripped over the whole root system. It was hypothesized that a more uniform application of spores might increase mycorrhizal colonization. At the end of the experiment, there were no differences in the abundance of mycorrhizae for the two methods of inoculation. From a logistical viewpoint, the injection method was more efficient and the method of dripping spores on the roots could potentially spread disease due to extra handling of seedlings.

Seedling Parameters

As shown by many studies, the effects of mycorrhizal inoculation and fertilization on seedling performance are variable and results are dependent on many factors (Castellano 1996, Quoreshi and Timmer 1998, Khasa et al. 2001, Campbell et al. 2003, Dumroese et al. 2005, Schwartz et al. 2006, Quoreshi et al. 2008, Khasa et al. 2009). The goal of fertilization is to produce healthy seedlings for out-plantings. Fertilized seedlings in this experiment had a larger root volume and wet biomass in comparison to un-

fertilized seedlings. In addition, seedlings in long containers had a larger root volume and wet biomass than seedlings in short containers. This could be expected as larger containers provide more space for root development (Landis et al. 2010a). In a separate analysis comparing only actually colonized seedling, (not just inoculated) to un-colonized seedlings, colonized seedlings had larger root systems regardless of container treatment (Data not presented). Inoculation with mycorrhizal fungi often produces ‘fuller’ root systems that may be better equipped for soil exploration after out-planting (Khasa et al. 2009).

No trend in crown development or needle color was observed regardless of fertilizer treatment or container length. Whitebark pine is a slow growing conifer and growth parameters are not as informative as for fast growing tree species. For whitebark pine, seedling survival is still the main concern, since survival typically decreases three years after out-planting to an average of 38% (Izlar 2007).

Conclusions

Currently, our data suggests it is possible to combine low N fertilization with ectomycorrhizal inoculation of WBP seedlings in the nursery. However, further research is necessary to refine and optimize the fertilization regime and inoculation methods so that the two strategies work synergistically. Production of seedlings similar in size to those produced with conventional fertilization can be achieved by combining lower fertilization rates with mycorrhizal colonization (Khasa et al. 2001). In another small trial with older WBP seedlings, fertilization was stopped one month prior to inoculation and 3-5 months were allowed for seedling colonization to occur before out-planting. These

seedlings quickly became colonized with ectomycorrhizal fungi, showing that it is possible to continue a typical fertilizer regime for two years which is then interrupted and followed by inoculation before out-planting (Cripps and Grimme 2011). Seedlings were of robust size but N levels were not analyzed. Inoculated seedlings of WBP are now out-planted on high elevation sites and are currently being monitored.

Nursery managers should consider inoculating seedlings when out-planting in areas that may lack native ectomycorrhizal fungi specific for whitebark pine, such as ghost forests or severe burns, and in areas where out-planting survival has been low (Keane et al. 2012). Regionally-appropriate native ectomycorrhizal fungi should be used for nursery inoculation of whitebark pine seedlings and the introduction of alien fungi into these sensitive forest ecosystems should be avoided (Keane et al. 2012). Inoculation with native ectomycorrhizal fungi may not be necessary if sufficient and appropriate native ectomycorrhizal fungi are available in the soil. In this case, it is recommended that un-inoculated seedlings be planted near inoculum sources such as live whitebark pine trees or be quickly planted in areas recently inhabited by whitebark pine (Keane et al. 2012).

References

- Aime, M.C. and Miller Jr., O.K. 2002. Delayed germination of basidiospores in temperate species of *Crepidotus* (Fr.) Staude. *Canadian Journal of Botany* 80(3): 280-287.
- Amaranthus, M.P. 2002. Around the world nursery inoculations and conifer establishment using *Rhizopogon* mycorrhizal fungi. In: Dumroese, R.K., Riley, L.E., and Landis, T.D., (coords. and eds.). National proceedings: forest and conservation nursery associations-1999, 2000, and 2001. Proceedings RMRS-P-24. Ogden, UT: U.S. Department of Agriculture Forest Service, Rocky Mountain Research Station. 226 p.
- Amaranthus, M.P., Nair, M.G., Reid, T.C., and Steinfeld, D. 2005. Improved *Rhizopogon* mycorrhizal colonization and foliar nutrient levels in Ponderosa pine and Douglas-fir with myconate (r). *Journal of Sustainable Forestry* 20(3): 1-14.
- Arnebrant, K. 1994. Nitrogen amendments reduce the growth of extramatrical ectomycorrhizal mycelium. *Mycorrhiza* 5(1): 7-15.
- Bentz, B.J., Kegley, S., Gibson, K., and Thier, R. 2005. A test of high-dose verbenone for stand-level protection of lodgepole and whitebark pine from mountain pine beetle (Coleoptera: Curculionidae: Scolytinae) attacks. *Journal of Economic Entomology* 98(5): 1614-1621.
- Boyle, C.D. and Robertson, W.J. 1987. Use of mycelial slurries of mycorrhizal fungi as inoculum for commercial tree seedling nurseries. *Canadian Journal of Forest Research* 17(12): 1480-1486.
- Brundrett, M., Bougher, N., Dell, B., Grove, T., and Malajczuk, N. 1996. Working with mycorrhizas in forestry and agriculture, ACIAR Monograph 32. 374 p.
- Brundrett, M., Malajczuk, N., Mingquin, G., Daping, X., Snelling, S., and Dell, B. 2005. Nursery inoculation of Eucalyptus seedlings in Western Australia and Southern China using spores and mycelial inoculum of diverse ectomycorrhizal fungi from different climatic regions. *Forest Ecology and Management* 209: 193-205.
- Brunner, I. and Brodbeck, S. 2001. Response of mycorrhizal Norway spruce seedlings to various nitrogen loads and sources. *Environmental Pollution* 114: 223-233.
- Bruns, T.D., Bidartondo, M.I., and Taylor, L. 2002. Host specificity in ectomycorrhizal communities: what do the exceptions tell us? *Integrative and Comparative Biology* 42: 352-359.

- Burr, K., Eramian, A., and Eggleston, K. 2001. Growing whitebark pine seedlings for restoration. Pp. 325-345. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds.). Whitebark pine communities: ecology and restoration. Island Press, Washington, DC. 440 p.
- Campbell, D.B., Jones, M.D., Kiiskila, S. and Bulmer, C. 2003. Two-year field performance of lodgepole pine seedlings: effects of container type, mycorrhizal fungal inoculants, and site preparation. B.C. Journal of Ecosystem Management 3(2): 1-11.
- Castellano, M.A. 1996. Outplanting performance of mycorrhizal inoculated seedlings. Pp. 223-301. In: Mukerji, K.G. (ed.). Concepts in Mycorrhizal Research. Kluwer Academic Publishers, Netherlands. 371 p.
- Castellano, M.A. and Molina, R. 1989. Mycorrhizae. Pp. 101-167. In: Landis, T.D., Tinus, R.W., McDonald, S.E., and Bamett, J.P. (eds.). The Container Tree Nursery Manual vol. V, USDA Forest Service, Public Affairs Office: Washington, DC.
- Castellano, M.A., Trappe, J.M., and Molina, R. 1985. Inoculation of container-grown Douglas-fir seedlings with basidiospores of *Rhizopogon vinicolor* and *R. colossus*: effects of fertility and spore application rate. Canadian Journal of Forest Research 15: 10-13.
- Chakravarty, P. and Chataropaul, L. 1989. Effect of fertilization on seedling growth, ectomycorrhizal symbiosis, and nutrient uptake in *Larix laricina*. Canadian Journal of Forest Research 20: 245-248.
- Chen, Y.L., Dell, B., and Malajczuk, N. 2006. Effects of *Scleroderma* spore density and age on mycorrhiza formation and growth of containerized *Eucalyptus globulus* and *E. urophylla* seedlings. New Forests 31(3): 453-467.
- Cordell, C.E. and Marx, D.H. 1994. Effects of nursery cultural practices on management of specific ectomycorrhizae on bareroot tree seedlings. Pp. 133-152. In: Pflieger, F.L. and Linderman, R.G. (eds.). Mycorrhizae and Plant Health. APS Press, St. Paul, Minnesota. 344 p.
- Cripps, C.L. 2004. Fungi in forest ecosystems: systematics, diversity and ecology. New York Botanical Gardens Press, NY. 363 p.
- Cripps, C.L. and Antibus, R.K. 2011. Native ectomycorrhizal fungi of limber and whitebark pine: necessary for forest sustainability? Pp. 37-44. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the

High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.

- Cripps, C.L. and Grimme, E. 2011. Inoculation and successful colonization of whitebark pine seedlings with native mycorrhizal fungi under greenhouse conditions. Pp. 312-322. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Cripps, C.L., Smith, C., Carolin, T. and Lapp, J. 2008. Ectomycorrhizal fungi with whitebark pine. *Nutcracker Notes* 14: 12-14.
- Danielson, R.M. 1991. Temporal changes and effects of amendments on the occurrence of sheathing (ecto-) mycorrhizas of conifers growing in oil sands tailings and coal spoil. *Agriculture Ecosystems & Environment* 35: 261-281.
- Diaz, G., Carrilla, C., and Honrubia, M. 2010. Mycorrhization, growth, and nutrition of *Pinus halepensis* seedlings fertilized with different doses and sources of nitrogen. *Annals of Forest Science* 67(405): 1-9.
- Dominguez-Lerena, S., Herrero Sierra, N., Carrasco Manzano, I., Ocana Bueno, L., Penuelas Rubira, J.L., and Mexal J.G. 2006. Container characteristics influence *Pinus pinea* seedling development in the nursery and field. *Forest Ecology and Management* 221(1): 63-71.
- Dumroese, R.K. 2008. Observations on root disease of container whitebark pine seedlings treated with biological controls. *Native Plants Journal* 9(2): 92-97.
- Dumroese, R.K., Page-Dumroese, D.S., Salifu, K.F., and Jacobs, D.F. 2005. Exponential fertilization of *Pinus monticola* seedlings: Nutrient uptake efficiency, leaching fractions, and early out-planting performance. *Canadian Journal of Forest Research* 35: 2961-2967.
- Eggleston, K. 2010. [Personal Communication with Dr. Cathy Cripps] April 19, 2010. USDA Forest Service Nursery, Coeur d'Alene, Idaho.
- Ellison, A.M., Bank, M.S., Clinton, B.D., Colburn, E.A., Elliott, K., Ford, C.R., Foster, D.R., Kloeppe, B.D., Knoepp, J.D., Lovett, G.M., Mohan, J., Orwig, D.A., Rodenhouse, N.L., Sobczak, W.V., Stinson, K.A., Stone, J.K., Swan, C.M., Thompson, J., Von Holle, B., and Webster, J.R. 2005. Loss of foundation species:

consequences for the structure and dynamics of forested ecosystems. *Frontiers in Ecology and the Environment* 3: 479–486.

Ekwebelam, S.A. and Reid, C.P.P. 1983. Effects of light, nitrogen fertilization, and mycorrhizal fungi on growth and photosynthesis of lodgepole pine seedlings. *Canadian Journal of Forest Research* 13(6): 1099-1106.

Gagne, A., Jany, J.L., Bousquet, J., Khasa, J., and Gamase, P. 2006. Ectomycorrhizal fungal communities of nursery-inoculated seedlings out-planted on clear-cut sites in northern Alberta. *Canadian Journal of Forest Research* 36: 1684-1694.

Gagnon, J., Langlois, C.G., and Fortin, J.A. 1988. Growth and ectomycorrhiza formation of containerized black spruce seedlings as affected by nitrogen fertilization, inoculum type, and symbionts. *Canadian Journal of Forest Research* 18: 922-929.

Gernandt, D.S., López, G.G., García, S.O., and Liston, A. 2005. Phylogeny and classification of *Pinus*. *Taxon* 54(1): 29-42.

González-Ochoa, A.I., Heras, J., Torres, P., and Sánchez-Gómez, E. 2003. Mycorrhization of *Pinus halepensis* Mill. and *Pinus pinaster* Aiton seedling in two commercial nurseries. *Annals of Forest Science* 60: 43-48.

Heumader, J. 1992. Cultivation of cembran pine plants for high-elevation afforestations. International workshop on subalpine stone pines and their environment: the status of our knowledge, St. Moritz, Switzerland.

IBM Corporation. Released 2011. IBM SPSS Statistics for Windows, version 20.0 Armonk, NY: IBM Corp.

Izlar, K. 2007. Assessment of whitebark pine seedling survival for Rocky Mountain plantings. University of Montana Master of Science thesis, Missoula, MT.

Jackson, D.P., Dumroese, R.K., and Barnett, J.P. 2012. Nursery response of container *Pinus palustris* seedlings to nitrogen supply and subsequent effects on outplanting performance. *Forest Ecology and Management* 265: 1-12.

James, R.L. and Burr, K.E. 2000. Diseases associated with whitebark pine seedling production. Coeur d'Alene, Idaho: USDA Forest Service Nursery, Forest Health Protection, May 2000, Report 00-8: 1-8.

Keane, R.E. and Parsons, R.A. 2010. Management guide to ecosystem restoration treatments: Whitebark pine forests of the northern Rocky Mountains, U.S.A. General Technical Report RMRS GTR-232. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 133 p.

- Keane, R.E., Tomback, D., Aubry, C., Bower, A., Campbell, E., Cripps, C., Jenkins, M., Manning, M., McKinney, S., Murray, M., Perkins, D., Reinhart, D., Ryan, C., Schoettle, A., Smith, C. 2012. A range-wide restoration strategy for whitebark pine (*Pinus albicaulis*). General Technical Report RMRS-GTR-279. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station.
- Keller, G. 1996. Utilization of inorganic and organic nitrogen sources by high-subalpine ectomycorrhizal fungi of *Pinus cembra* in pure culture. *Mycological Research* 100: 989-998.
- Khasa, P.D., Piche, Y., and Coughlan, A.P. (eds.). 2009. *Advances in mycorrhizal science and technology*. NRC Research Press, Ottawa. 197 p.
- Khasa, P.D., Sigler, L., Chakravarty, P., Dancik, B.P., Erickson, L., and McCurdy, D. 2001. Effect of fertilization on growth and ectomycorrhizal development of container-grown and bare-root nursery conifer seedlings. *New Forests* 22: 179-197.
- Landis, T.D., Dumroese, R.K., and Haase, D.L. 2010a. Seedling processing, storage, and outplanting, vol. 7. *The container Tree Nursery Manual*. Agric. Handbook, 674. U.S. Department of Agriculture Forest Service, Washington, D.C. 200 p.
- Landis, T.D., Seinfeld, D.E., and Dumroese, R.K. 2010b. Native plant containers for restoration projects. *Native Plants Journal* 11(3): 341-348.
- Landis, T.D., Tinus, R.W., McDonald, S.E., and Barnett, J.P. 1990. Containers and growing media, vol. 2. *The container Tree Nursery Manual*. Agric. Handbook, 674. U.S. Department of Agriculture Forest Service, Washington, D.C. 88 p.
- Landis, T.D., Tinus R.W., McDonald S.E., Barnett J.P. 1989a. Seedling nutrition and irrigation, vol. 4. *The container Tree Nursery Manual*. Washington (D.C.): USDA Forest Service, Agriculture Handbook. 119 p.
- Landis, T.D., Tinus, R.W., McDonald, S.E., Barnett, J.P. 1989b. The biological component: nursery pests and mycorrhizae, vol. 5. *The container Tree Nursery Manual*. Washington (D.C.): USDA Forest Service, Agriculture Handbook. 674. 171 p
- Lantz, G. 2010. *Whitebark pine: An ecosystem in peril*. American Forests Special Report. Norman, OK. 8 p.
- Lehto, T. and Zwiazek, J.J. 2011. Ectomycorrhizas and water relations of trees: a review. *Mycorrhiza* 21(2): 71-90.

- Logan, J., Mcfarlane, W., and Wilcox, L. 2010. Whitebark pine vulnerability to climate driven mountain pine beetle disturbance in the Greater Yellowstone Ecosystem. *Ecological Applications* 20(40): 895-902.
- Mahalovich, M.F., Burr, K.E., and Foushee, D.L. 2006. Whitebark pine germination, rust resistance, and cold hardiness among seed sources in the inland northwest: planting strategies for restoration. USDA Forest Service Proceedings RMRS-P-43.
- Mahalovich, M.F. and Dickerson, G.A. 2004. Whitebark pine genetic restoration program for the Intermountain West (USA). Pp. 181-187. In: Sniezko, R., Samman, S., Schlarbaum, S., and Kriebel, H. (eds.). *Breeding and genetic resources of five-needle pines: growth, adaptability and pest resistance*, IUFRO Working Party 2.02.15, Proceedings. USDA Forest Service, Rocky Mountain Research Station, RMRS-P-32, Ogden, UT.
- Mahalovich M.F. and Hoff, R.J. 2001. Whitebark pine operational cone collection instructions and seed transfer guidelines. *Nutcracker Notes* 11: 10-13.
- Marx, D. H. 1980. Ectomycorrhiza fungus inoculations: A tool for improving forestation practices. Pp. 13–71. In: Mikola, P. (ed.). *Tropical Mycorrhiza Research*. Oxford University Press, Oxford. 270 p.
- Marx, D.H. and Cordell, C.E. 1988. Specific ectomycorrhizae improve reforestation and reclamation in eastern United States. In: Lalonde, M. and Piché, Y., (cords. and eds). *Symposium Proceedings, Canadian Workshop on Mycorrhizae in Forestry*; May 1-4, 1988; Centre de recherche en biologie forestière, Facult de foresterie et de géodesie, Université Laval, Sainte-Foy, Qué.
- McCaughey, W.W. 1990. Biotic and microsite factors affecting *Pinus albicaulis* establishment and survival. Montana State University PhD dissertation, Bozeman, MT.
- McCaughey, W.W., Scott, G.L., and Izlar, K.L. 2009. Whitebark pine planting guidelines. *Western Journal of Applied Forestry* 24(3): 163-166.
- McCune, B. 1988. Ecological diversity in North American pines. *American Journal of Botany* 75(3): 353-368.
- Meikle, T.W. and Amaranthus, M. 2008. The influence of fertilization regime and mycorrhizal inoculum on outplanting success: a field trial of containerized seedlings in Oregon. *Native Plants Journal* 9(2): 107-116.
- Menkis, A., Vasiliauskas, R., Taylor, A.F.S., Senlid, J., and Finlay, R. 2007. Afforestation of abandoned farmland with conifer seedlings inoculated with three

ectomycorrhizal fungi- impact on plant performance and ectomycorrhizal community. *Mycorrhiza* 17(4): 337-348.

Mohatt, K.R. 2006. Ectomycorrhizal fungi of whitebark pine (*Pinus albicaulis*) in the Greater Yellowstone Ecosystem. Montana State University Master of Science thesis, Bozeman, MT.

Mohatt, K.R., Cripps, C.L., and Lavin, M. 2008. Ectomycorrhizal fungi of whitebark pine (a tree in peril) revealed by sporocarps and molecular analysis of mycorrhizae from treeline forests in the Greater Yellowstone Ecosystem. *Botany* 86: 14-25.

Moser, M.M. 1956. Die Bedeutung der Mykorrhiza für Aufforestungen in Hochlagen. *Forstwissenschaftliches Centralblatt* 75: 8-18.

Moser, M.M. 2004. Subalpine conifer forests in the Alps, the Altai and the Rocky Mountains: a comparison of their fungal populations. Pp. 151-158. In: Cripps, C.L. (ed.). *Fungi in forest ecosystems: systematics, diversity, and ecology*. New York Garden Botanical Press, N.Y. 378 p.

Murray, M.P. 2007. Cone collecting techniques for whitebark pine. *Western Journal of Applied Forestry* 22(3): 153-155.

Nicholas, A. and Katzenberger, D. 2011. Whitebark pine to be designated a candidate for endangered species protection. U.S. Dept. of Interior, Fish and Wildlife Service, Press Release, Lakewood, CO.

Oliveira, R.S., Franco, A.R., Vosatka, M., and Castro, P.M.L. 2010 Management of nursery practices for efficient ectomycorrhizal fungi application in the production of *Quercus ilex*. *Symbiosis* 52(2-3): 125-131.

Ortega, U., Dunabeitia, M., Mendez, S., Gonzalez-murua, C., and Majada, J. 2004. Effectiveness of mycorrhizal inoculation in the nursery on growth and water relations of *Pinus radiata* in different water regimes. *Tree Physiology* 24: 65-73.

Parladé, J., Luque, J., Pera, J., Rincón, A.M. 2004. Field performance of *Pinus pinea* and *P. halepensis* seedlings inoculated with *Rhizopogon* spp. and outplanted in formerly arable land. *Annals of Forest Science* 61: 507-514.

Parladé, J., Pera, J., and Alvarez, F. 1996. Inoculation of containerized *Pseudotsuga menziesii* and *Pinus pinaster* seedlings with spores of five species of ectomycorrhizal fungi. *Mycorrhiza* 6(4): 237-245.

Peintner, U. 2012. [Personal Communication to Dr. Cathy Cripps]. Professor of Biology, University of Innsbruck, Innsbruck, Austria.

- Perkins, Judy. 2004. *Pinus albicaulis* seedling regeneration after fire. University of Montana PhD dissertation, Missoula, MT.
- Quoreshi, A.M., Kernaghan, G., and Hunt, G.A. 2009. Mycorrhizal fungi in Canadian forest nurseries and field performance of inoculated seedlings. Pp. 115-128. In: Khasa, P.D., Piche, Y., and Coughlan, A.P. (eds.). *Advances in Mycorrhizal Science and Technology*. NRC Research Press, Ottawa. 197 p.
- Quoreshi, A. M., Piche, Y., and Khasa, D.P. 2008. Field performance of conifer and hardwood species 5 years after nursery inoculation in the Canadian Prairie Provinces. *New Forests* 35: 235-253.
- Quoreshi, A.M. and Timmer, V.R. 1998. Exponential fertilization increases nutrient uptake and ectomycorrhizal development of black spruce seedlings. *Canadian Journal of Forest Research* 28: 674-682.
- Repáč, I. 1996a. Inoculation of *Picea abies* (L.) Karst. seedlings with vegetative inocula of ectomycorrhizal fungi *Suillus bovinus* (L.:Fr.) O. Kuntze and *Inocybe lacera* (Fr.) Kumm. *New Forests* 12: 41-54.
- Repáč, I. 1996b. Effects of forest litter on mycorrhiza formation and growth of container - grown Norway spruce (*Picea abies* [L.] Karst.) seedlings. *Lesnictví* 42: 317-324.
- Repáč, I. 2007. Ectomycorrhiza formation and growth of *Picea abies* seedlings inoculated with alginate-bead fungal inoculum in peat and bark compost substrates. *Forestry* 80(5): 517-530.
- Repáč, I. 2011. Ectomycorrhizal Inoculum and Inoculation Techniques. Pp 43-63. In: Rai, M. and Varma, A. (eds.). *Diversity and Biotechnology of Ectomycorrhizae*, *Soil Biology* 25, Springer - Verlag, Heidelberg, Berlin. 459 p.
- Rikala, R., Heiskanen, J., and Lahti, M. 2004. Autumn fertilization in the nursery affects growth of *Picea abies* container seedlings after transplanting. *Scandinavian Journal of Forest Research* 19(5): 409-414.
- Rincón, A., de Felipe, M.R., and Fernández-Pascual, M. 2007. Inoculation of *Pinus halepensis* Mill. with selected ectomycorrhizal fungi improves seedling establishment 2 years after planting in a degraded gypsum soil. *Mycorrhiza* 18: 23-32.
- Rincón, A., Parlade, J., and Pera, J. 2005. Effects of ectomycorrhizal inoculation and the type of substrate on mycorrhization, growth and nutrition of containerized *Pinus*

pinea L. seedlings produced in a commercial nursery. *Annals of Forest Science* 62: 817-822.

- Schmid, V. 2006. Entwicklung molekularer methoden für ein schnelles und kostengünstiges monitoring der inokulation von forstpflanzen mit ektomykorrhizasymbionten. University of Innsbruck PhD dissertation, Innsbruck, Austria.
- Schwandt, J. 2006. Whitebark pine in peril: A case for restoration. U.S. Department of Agriculture, Forest Service, Forest Health Protection R1-06-28. Missoula, MT. 20 p.
- Schwartz, M.W., Hoeksema, J.D., Gehring, C.A., Johnson, N.C., Klironomos, J.N., Abbott, L.K, and Pringle, A. 2006. The promise and the potential consequences of the global transport of mycorrhizal fungal inoculum. *Ecology Letters* 9(5): 501-515.
- Shaw, C.G., Molina, R., Walden, J. 1981. Development of ectomycorrhizae following inoculation of containerized Sitka and white spruce seedlings. *Canadian Journal of Forest Research* 12: 191-195.
- Smith, S.E. and Read, D.J. 2008. Mycorrhizal symbiosis, 3rd edition. Academic Press, Inc., San Diego, CA. 787 p.
- Steinfeld, D., Amaranthus, M., and Cazares, E. 2003. Survival of ponderosa pine (*Pinus ponderosa* Dougl. Ex Laws.) seedlings out-planted with *Rhizopogon* mycorrhizae inoculated with spores at the nursery. *Journal of Arboriculture* 29(4): 197-208.
- Timmer, V.R. 1996 Exponential nutrient loading: a new fertilization technique to improve seedlings performance on competitive sites. *New Forests* 13: 275-295.
- Tomback, D.F. and Achuff, P. 2010. Blister rust and western forest biodiversity: ecology, values, and outlook for white pines. *Forest Pathology* 40: 186-225.
- Tomback, D.F., Arno, S.F., and Keane, R.E. 2001. Whitebark pine communities: ecology and restoration. Island Press, Washington, DC. 440 p.
- Tomback, D.F. and Linhart, Y.B. 1990. The evolution of bird-dispersed pines. *Evolutionary Ecology* (4): 185-219.
- Torres, P. and Honrubia, M. 1994. Basidiospore viability in stored slurries. *Mycological Research* 98(5): 527-530.

- Trappe, J.M. 1977. Selection of fungi for ectomycorrhizal inoculation in nurseries. *Annual Review of Phytopathology* 15: 203–222.
- Treu, R. 1990. *Suillus sibiricus*. In: Colour atlas of ectomycorrhizae, plate 47, Agerer, R. (ed.). Einhorn-Verlad, Schwabisch Gmund.
- Trusty, P. and Cripps, C.L. 2011. Influence of fire on mycorrhizal colonization of planted and natural whitebark pine seedlings: Ecology and management implications. Pp. 198-202. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Van Den Driessche, R. 1987. Nursery growth of conifer seedlings using fertilizers of different solubilities and application time, and their forest growth. *Canadian Journal of Forest Research* 18: 172-173.
- Wallander, H. and Nylund, J.E. 1991. Effects of excess nitrogen on carbohydrate concentration and mycorrhizae development of *Pinus sylvestris* seedlings. *New Phytologist* 119: 405-441.
- Weisleitner, H. 2008. [Personal Communication with Dr. Cathy Cripps]. Federal Nursery of Austria, Innsbruck, Austria.
- Whipps, J.M. 2004. Prospects and limitations for mycorrhizas in biocontrol of root pathogens. *Canadian Journal of Botany* 82: 1198-1227.
- Wiensczyk, G., Durall, D. Jones, M. and Simard, S. 2002. Ectomycorrhizae and forestry in British Columbia: a summary of current research and conservation strategies. Extension Note. *B.C. Journal of Ecosystems and Management* 2(1):1-20.

CHAPTER 3

EFFECT OF SITE CONDITIONS, MICROSITE, AND INOCULATION WITH
NATIVE ECTOMYCORRHIZAL FUNGI ON THE EARLY SURVIVAL
OF OUT-PLANTED WHITEBARK PINE SEEDLINGSIntroductionWhitebark Pine

Whitebark Pine (*Pinus albicaulis* Engelm.) is a long-lived, slow growing, high elevation, five-needle pine species that is in peril. It is distinct as the only stone pine (subgenus *Strobus*, subsection *Cembrae*) in North America and is limited to the western portion of the continent (Tomback 1982). Stone pines produce indehiscent cones that do not open to release seeds and instead rely on birds (Clark's nutcracker) for seed dispersal (Lanner 1996, Price et al. 1998).

In western North American, whitebark pine is limited to upper sub-alpine and timberline vegetation zones where it is considered a 'keystone' and 'foundation' species (McCaughey and Schmidt 2001, Fryer 2002, Ellison et al. 2005, Lantz 2010). Whitebark pines' unique seed dispersal strategy and its large meaty seeds allow it to be a primary colonizer of high elevation ecosystems (Tomback et al. 2001a). Establishment of whitebark pine in these open, disturbed sites often facilitates colonization of other subalpine species by mitigating harsh environmental conditions (Tomback et al. 2001a). In the northern Rocky Mountains east of the continental divide, whitebark pine is the

most common conifer involved in the formation of timberline 'tree islands' (Resler 2004).

Whitebark pine plays a pivotal role in supporting biological diversity and providing environmental stability in these high elevation communities (Ellison et al. 2005, Lantz 2010). The extensive, radiating root system helps stabilize steep mountain slopes and slows erosion of poorly developed top soils; this is especially true in the upper subalpine and timberline portions of its range (Farnes 1990). In areas where it forms extensive pure stands, canopy shading delays snowmelt which subsequently regulates downstream water flow (Farnes 1990). Whitebark pine seeds are a valuable high fat and high energy food source for many animals including grizzly bears, black bears, small mammals, and birds (Arno 1986). Prior to hibernation, grizzly and black bears rely heavily on whitebark pine seeds as a protein-rich food source; low cone production has been correlated with an increase in grizzly bear mortality (Arno 1986, Mattson et al. 1992, Mattson and Reinhart 1994, Felicetti et al. 2003).

Whitebark pine populations are in serious decline and have been reduced by 90% or more in certain areas of its range (Izlar 2007). White pine blister rust (*Cronartium ribicola* J.C. Fischer), mountain pine beetle (*Dendroctonus ponderosae* Hopkins) infestations exacerbated by fire suppression, and global climate change are major contributors to the rapid decline of whitebark pine populations (Keane and Arno 1993, McKinney and Tomback 2007, Keane and Parsons 2010, Logan et al. 2010, Tomback and Achuff 2010). The synergistic effects of these factors have been devastating (Logan et al. 2010). The U.S. Fish and Wildlife Service has stated that threats to whitebark pine

populations are ‘imminent’ and ‘of high magnitude,’ and that this species is in danger of extinction (Nicholas and Katzenberger 2011). As of July 18, 2011 the U.S. Fish and Wildlife Service determined that whitebark pine was warranted for protection under the Endangered Species Act; it is currently precluded due to funding and priority constraints (Nicholas and Katzenberger 2011).

Restoration

The importance of whitebark pine in the West has been recognized and restoration efforts have been on-going for more than 30 years (Tomback et al. 2001c). The Range Wide Restoration Strategy for Whitebark Pine (Keane et al. 2012) lays out a plan that focuses on genetic conservation aiding natural regeneration, and on restoration accomplished by the planting of rust-resistant, nursery grown seedlings (Keane et al. 2012). This has led to the development and improvement of methods for rust resistance screening (Mahalovich and Hoff 2001, Mahalovich and Dickerson 2004, Mahalovich et al. 2006), cone collection (Burr et al. 2001), seed germination (Murray 2007), nursery seedling production, and out-planting methods (McCaughey et al. 2009, Scott et al. 2011). To date, over 200,000 nursery grown seedlings have been planted in Western North America (Izlar 2007).

The use of rust-resistant seedlings is a crucial component in whitebark pine restoration. Management of this partial natural resistance provides an avenue for potentially establishing a balance between the survival of whitebark pine and this naturalized non-native pathogen (Hoff et al. 2001, Aubry et al. 2008). Rust-resistance screening programs for whitebark pine have been adapted from methods developed for

western white pine trees (*Pinus monticola* Douglas ex D. Don), which were seriously hit with white pine blister rust beginning around 1910 (Hoff et al. 2001). The whole process of locating and confirming genetically resistant parent stock, including identification of potential ‘plus’ trees through cone collection, seed germination, and artificial rust inoculation trials, can take five years or longer (Mahalovich and Dickerson 2004).

Production of whitebark pine seedlings for restoration has also proven to be time consuming and challenging. Mature cone-bearing ‘plus’ trees must first be identified, the cones collected, and seeds harvested before being entered into the USFS genetic resistance screening program (Burr et al. 2001). Whitebark pine cones take two years to mature and cages are applied in early summer of the 2nd year to protect maturing cones from bird and squirrel predation (Burr et al. 2001, Leslie and Wilson 2011). The collected cones are dried and seeds extracted with an automated cleaning machine (Burr et al. 2001). Seeds are then given a seed lot number that includes information on the parental stock and this determines the zones where subsequent seedlings can be planted (Mahalovich and Dickerson 2004). The Rocky Mountain region is comprised of six seed zones, and transfer restrictions are primarily aimed at limiting movement between areas with different rust infection levels and areas with different cold hardiness thresholds (Mahalovich et al. 2006, Bower and Aitken 2008, Burns et al. 2008).

Due to delayed seed maturation and dormancy factors, whitebark pine seeds normally take two or more years to germinate under natural conditions. Seed stratification and germination techniques have been developed to hasten germination in the nursery (Burr et al. 2001, McCaughey and Tomback 2001). Following stratification

procedures, seeds are sown directly into long containers and the germinated seedlings are regularly fertilized and irrigated at the indoor nursery (Eggleston 2010). Whitebark pine seedlings are typically not out-planted until 2-3 years of age (Burr et al. 2001, Eggleston 2010). Currently, only two facilities are involved in growing whitebark pine seedlings for restoration: the USDA Forest Service Dorena Genetic Resource Center in Cottage Grove, Oregon and the USDA Forest Service Coeur d'Alene Nursery in Coeur d'Alene, Idaho.

Hundreds of thousands of potentially rust-resistant seedlings have been planted in the Western U.S. and Canada; however survival data is limited due to the scarce resources available for monitoring (Izlar 2007, Keane et al. 2012). The first wide-scale monitoring of the survival of out-planted nursery grown whitebark pine seedlings was conducted in 2007 (Izlar 2007). Approximately 100,000 whitebark pine seedlings planted over a wide range of sites and ecological conditions on federal lands in the Rocky Mountains from 1989-2005 were assessed for survival (Izlar 2007). Survival rates were low and averaged around 42%, with a decline between 1st year survival (74%) and 3rd-15th year survival (38%). Mellmann-Brown (2005) monitored the survival of 2,325 whitebark pine seedlings out-planted at tree-line in Montana and Wyoming and reported a rapid decline between 1st (36%) and 8th year survival (14%). These studies as well as others identified a few factors that might correlate with higher seedling survival including: planting in mixed severity burns, in mesic areas, and near microsites such as large logs or rocks that can provide physical protection from solar radiation, wind, snow drifts, and soil erosion (Mellmann-Brown 2005, Izlar 2007, McCaughey et al. 2009, Scott et al. 2011). Additional studies have sought to show that natural regeneration is also

enhanced in burns but data are inconclusive (Tomback et al. 2001b, Keane and Parsons 2010, Larson and Kipfmüller 2010). The presence of understory plants that show a high degree of resistance to fire and an ability to re-sprout quickly from underground rhizomes, such as beargrass and Geyer's sedge, have been correlated with a decrease in the survival of whitebark pine seedlings (Perkins 2004, Izlar 2007).

The actual planting process can be a weak link in the restoration process and detailed whitebark pine planting guidelines have been developed (McCaughey et al. 2009, Scott et al. 2011). The guidelines suggest that healthy, robust seedlings be out-planted in the fall in burned or open areas where they have access to space and light (Izlar 2007, McCaughey et al. 2009, Scott et al. 2011). Planting in a microsite where seedlings are provided physical protection from the elements is also recommended and is thought to increase survival success (Izlar 2007, McCaughey et al. 2009, Scott et al. 2011). Planting is not recommended near quick growing conifer species, in areas with dense understories, in deep, wet soils, or near snags (Izlar 2007, McCaughey et al. 2009, Scott et al. 2011). Snags may appear to provide seedling protection, but they eventually fall and can crush or up-root planted whitebark pine seedlings. Planting near broken off snags, stumps, large downed trees, or rocks is believed to provide better protection for developing whitebark pine seedlings (McCaughey et al. 2009, Scott et al. 2011).

Restoration in the International Peace Park

Whitebark pine populations have dramatically decreased within both Waterton Lakes and Glacier National Parks. In Glacier National Park (GNP), whitebark pine mortality was estimated at 44% in 2003 and populations have continued to decline with at

least 78% of live trees now infected with white pine blister rust (Kendall and Keane 2001, Smith et al. 2008, Asebrook et al. 2011). In Waterton Lakes National Park (WLNP), whitebark pine mortality has increased from 26% in 1996 to 61% in 2009 (Smith et al. 2008, Smith et al. 2011b). White pine blister rust infection levels have also shown a rapid increase from 43% in 1996 to 78% in 2009 in WLNP (Smith et al. 2008, Smith et al. 2011b).

Waterton Lakes-Glacier International Peace Park has an active, multi-faceted whitebark pine restoration program in place. Whitebark pine restoration began in GNP in 1997 with seed collection from potential ‘plus’ trees. Park personnel began growing seedlings in 1998 and the first seedlings were planted in 2000 (Asebrook et al. 2011). Between 2000 and 2007, 6,400 nursery grown whitebark pine seedlings and 723 seeds were planted within GNP (Asebrook et al. 2011). Approximately 21% of the planted seedlings are being monitored and 3-8 year survival rates are consistent with previous studies averaging 41% (Izlar 2007, Asebrook et al. 2011). Seven hundred twenty-three whitebark pine seeds were planted directly into the soil. The first year germination rate of these whitebark pine seeds was very low, with only 3 out of 723 seeds germinating and only two surviving after the first season (Asebrook et al. 2011). Other studies also found germination and subsequent survival rates of directly planted whitebark pine seeds to be low (McCaughey 1990, Perkins 2004, Mellman-Brown 2005, Schwandt et al. 2011). Research trials outside of the national park that aim at improving germination of directly planted seeds using warm stratification and scarification techniques are on-going

(Schwandt et al. 2011). Cone collection, seedling production, and the out-planting of nursery grown seedlings continue in GNP.

Whitebark pine restoration efforts within WLNP began in 2006 with the proactive protection of potentially rust-resistant ‘mother’ trees from mountain pine beetle attack. The trees are protected using Verbenone, an anti-aggregation pheromone, and green-leaf volatiles which fool the beetles into believing the tree is not a suitable host plant (Fettig et al. 2008, Smith 2009, Smith 2012). Seeds from 15 potentially rust-resistant ‘parent’ trees have been collected from cones and entered into the USFS genetic testing program for white pine blister rust-resistance. The first whitebark pine seedlings derived from these parent trees were planted in WLNP in 2009: since then, over 2,900 seedlings have been planted and approximately 70% of the planted seedlings are being monitored (Smith 2009, Smith 2012). First year survival rates averaged 64% (Smith 2012).

In 2008, whitebark pine was approved for listing as an endangered species in the province of Alberta (Government of Alberta 2010). The Committee on the Status of Endangered Wildlife in Canada (COSEWIC), an advisory body to the Species at Risk Act (SARA), designated whitebark pine as endangered in 2010 (COSEWIC 2010) and following this designation it was federally listed throughout Canada in July of 2012 (Government of Canada 2012). As part of a \$90 million investment to address ecological concerns in National Parks across Canada, \$7.1 million has been invested in the *Restoring Terrestrial Ecosystems* project in Waterton Lakes National Park. One focus of the WLNP restoration effort will be to reverse the decline of whitebark pine communities through the collection, propagation, and out-planting of ‘plus’ seedlings (Smith 2008).

Current restoration efforts include the protection of ‘plus’ trees from mountain pine beetle attack, cone collection, out-planting of seedlings, and monitoring (Parks Canada 2010).

Ectomycorrhizae

With whitebark pine populations continuing to decline and with out-planting success low, new strategies for maintaining and restoring these valuable forests are under exploration. One previously overlooked tool is the use of native ectomycorrhizal fungi to enhance whitebark pine seedling survival in the field following out-planting (Mohatt 2006, Cripps et al. 2008, Cripps and Antibus 2011, Cripps and Grimme 2011).

Inoculation with native ectomycorrhizal fungi can enhance the survival of out-planted nursery grown conifer seedlings by increasing their ability to rapidly access water and nutrients (Ortega et al. 2004, Quoreshi et al. 2009, Lehto and Zwiazek, 2011) and is recommended during the greenhouse production of seedlings to be used in restoration, especially in areas devoid of native ectomycorrhizal fungi (Wiensczyk et al. 2002).

Mycorrhizal inoculation is routinely used in reforestation efforts for other conifer species and has been shown to improve survival under certain conditions (Moser 1956, Wiensczyk et al. 2002, Steinfeld et al. 2003, Parladé et al. 2004, Gagne et al. 2006, Menkis et al. 2007, Khasa et al. 2009), but whitebark pine seedlings grown at the Forest Service nurseries are not currently inoculated with ECM fungi (Eggleston 2010).

Currently, there is no commercial inoculum appropriate for whitebark pine and introduction of alien fungi into these sensitive forest systems is not recommended (Keane et al. 2012).

Whitebark pine is known to associate with around 50 species of ectomycorrhizal fungi in the Rocky Mountains, including several species of suilloid fungi (Mohatt 2006, Mohatt et al. 2008, Cripps and Antibus 2011). Suilloid fungi are a group of ectomycorrhizal fungi primarily restricted to hosts in the Pinaceae (Hunt 1992, Menkis et al. 2005, El Karkouri et al. 2005). Within Pinaceae, specific suilloid fungi tend to form associations with specific tree genera, sub-genera or species (Bruns et al. 2002). Suilloid fungi are in the phylum Basidiomycota, family Boletaceae, and the primary taxa are *Suillus* and *Rhizopogon* species. *Rhizopogon* species are important in conifer establishment and are commonly used for nursery inoculation of conifer seedlings within the U.S. (Castellano 1996, Amaranthus 2002). However, Cripps and Grimme (2011) found that *Suillus* species specific to five needle pines are also important ectomycorrhizal partners for both young and mature whitebark pine trees. Some of these same *Suillus* species also occur with other stone pines in comparable forest ecosystems in the Swiss Alps and the Altai Mountains of Central Asia (Ronikier et al. 2002, Moser 2004). In addition, mushrooms produced by suilloid fungi are an integral part of the food chain in whitebark pine forests and are eaten by squirrels, deer, elk and bears (Fortin 2011, Cripps 2012).

The European stone pine is a close relative of whitebark pine and provides similar ecological services in high elevation communities in Europe and Asia (McCune 1988, Tomback and Linhart 1990, Gernandt et al. 2005). The Federal Nursery in Austria has been growing European stone pines (*Pinus cembra* L.) for reforestation since the 1950's and cultivation practices have been aimed at the production of seedlings colonized by 3

Suillus species adapted to high elevation conditions (Moser 1956, Heumader 1992, Weisleitner 2008). These restoration efforts using native ECM fungi have resulted in flourishing forests of cembra pines in the Austria Alps (Weisleitner 2008). However, the effects of mycorrhizal inoculation on the out-planting performance of conifer seedlings are variable and tend to be highly site specific (Castellano 1996, Brundrett et al. 2005, Menkis et al. 2007, Quoreshi et al. 2008).

One site-specific variable that has played a significant role in whitebark pine ecology and restoration is fire. Fire plays an important role in the health and maintenance of whitebark pine forests (Keane and Parsons 2010) and a majority of whitebark pine seedlings planted for restoration purposes have been on federal land burned by prescribed or wildfire (Izlar 2007). Although the aboveground influence of fire on whitebark pine regeneration has been studied (Tomback et al. 2001b, Perkins 2004, Keane and Parsons 2010, Larson and Kipmueller 2010), there is limited information on how fire impacts the availability of ectomycorrhizal fungi in the soil for establishing whitebark pine seedlings.

Fire is known to affect established mycorrhizal communities of other conifers, but impacts are variable and dependent on many factors including: severity, intensity and size of a fire, as well as on time since the fire occurred, tree species, forest stand age, climate, and soil moisture (Neary et al. 1999, Cairney and Bastias 2007). Fire typically causes a considerable alteration in soil microbial composition (Neary et al. 1999, Certini 2005, Cairney and Bastias 2007) and the abundance of ectomycorrhizal fungi can decrease following high intensity wildfire (Neary et al. 1999). In whitebark pine forests, high intensity wildfire has been reported to decrease the diversity and species richness of

ectomycorrhizal communities on seedlings (Trusty 2009). This study found a significant difference in ectomycorrhizal species on natural and planted whitebark pine seedlings regenerating on a severe burn in comparison to non-burned areas. There was also a lowered abundance in suilloid fungi on the whitebark pine seedlings in the burn four years after the wildfire (Trusty 2009). This suggests there was a reduced availability of fungi in the soil on the burn. The potential elimination of fungal propagules is one reason inoculation is recommended for seedlings planted on severe burns (Wiensczyk et al. 2002). If the availability of mycorrhizal fungi is reduced in the soil, it suggests there may be a need for inoculation of seedlings before they are out-planted on these sites.

Other site characteristics that can affect the mycorrhizal community and the ability of fungal species to colonize host trees are: soil composition, moisture, temperature, pH, cation exchange capacity, climate, planting date, and human mediated stresses such as soil compaction and the presence of pesticides and heavy metals (Entry et al. 2002, Wiensczyk et al. 2002). The presence of competing herbaceous vegetation has been noted to decrease the abundance of ectomycorrhizal fungi on host roots as well (Sylvie and Jarstfer 1997, Wiensczyk et al. 2002). This may be of particular importance to whitebark pine restoration in areas where certain understory plants such as beargrass could re-sprout quickly from underground rhizomes not sufficiently burned by fire.

Research Objectives

This field study evaluated whether inoculation with *Suillus sibiricus* Singer (a native ECM fungus) increased survival rates of whitebark pine seedlings out-planted in 2010 in four different site conditions (burn/non-burned, beargrass/non-beargrass) within

Waterton Lakes National Park. Seedlings were planted in burned and unburned areas because burns are currently believed to improve seedling survival (Izlar 2007, McCaughey et al. 2009). Seedlings were planted in areas with and without beargrass because it is the dominant understory vegetation at the site and is thought to negatively impact whitebark pine seedling survival (Crane 1990, Chadwick 2002, Perkins 2004, Izlar 2007). Seedlings were planted in clusters of three to mimic the natural planting strategy of Clark's nutcracker and because in high elevation areas it is recommended that seedlings be planted in clusters to provide protection from harsh climatic conditions (Heumader 2000, Schönenberger 2001). Each cluster contained 0-3 inoculated seedlings (4 treatments) to evaluate the minimum number of inoculated seedlings needed for subsequent plantings. The survival, health, and height of each seedling were measured in the fall of 2011 and 2012. This was the first attempt to monitor whitebark pine seedlings inoculated with ectomycorrhizal fungi in a detailed manner.

Material and Methods

Study Site

The field study site is located near Summit Lake on the Carthew-Alderson trail in Waterton Lakes National Park (WLNP), Alberta Canada at latitude 49° 0' 43" North and longitude 114° 1' 30" West (Figure 3.1). The Summit Lake study area is located on a morainal saddle between 1,954 (6,41 ft) and 1,999 meters (6,558 ft) in elevation; the site is relatively flat with gentle slopes throughout. The climate is characterized by deep, persistent snow packs and short summers. The average yearly temperature is 5 °C (41

°F); average summer temperature is 14 °C (57 °F) and the average winter temperature is -4 °C (24 °F). The average annual precipitation is 152 cm; in 2011 and 2012 spring precipitation near the Summit Lake study area was approximately 30-40 cm above average (Government of Alberta 2012, Figure 3.2). Winds are frequent and strong averaging 30km/hr with gusts of over 100km/hr being common in the fall and winter (Government of Alberta 2012). The soil composition is an orthic humo-ferric podzol derived from glacial till; it varies between 10-70% coarse fragments (Coen and Holland 1976). The soil texture is loam to gravelly loam (Coen and Holland 1976).

The over-story on the site consists of mixed coniferous forest comprised mainly of subalpine fir (*Abies lasiocarpa* (Hook.) Nutt.) and Engelmann spruce (*Picea engelmannii* Parry ex Engelm) with scattered whitebark pine trees. The dominant understory vegetation is comprised of thick mats of beargrass (*Xerophyllum tenax* (Pursh) Nutt.) and scattered rusty menziesia (*Menziesia ferruginea* Sm.) and huckleberry shrubs (*Vaccinium membranaceum* Douglas ex Torr.); (Achuff et al. 2002). White pine blister rust infection and mountain pine beetle attack have caused severe mortality in the mature whitebark pine trees at the site. Many of the naturally regenerating whitebark pine seedlings at the site are also dead or dying due to white pine blister rust and competitive exclusion from faster growing, shade tolerant conifers (Smith et al. 2008).

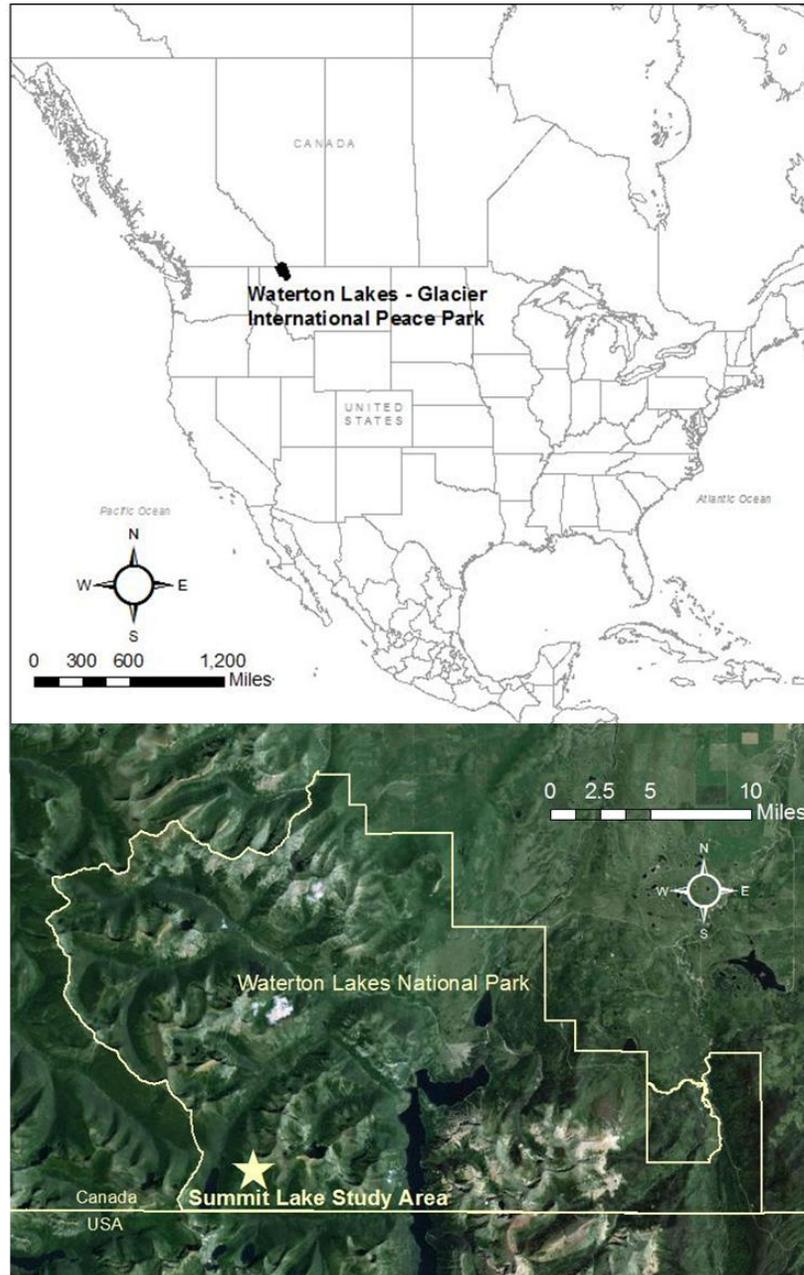


Figure 3.1: Location of study site in Waterton Lakes National Park located in Southern Alberta, Canada.

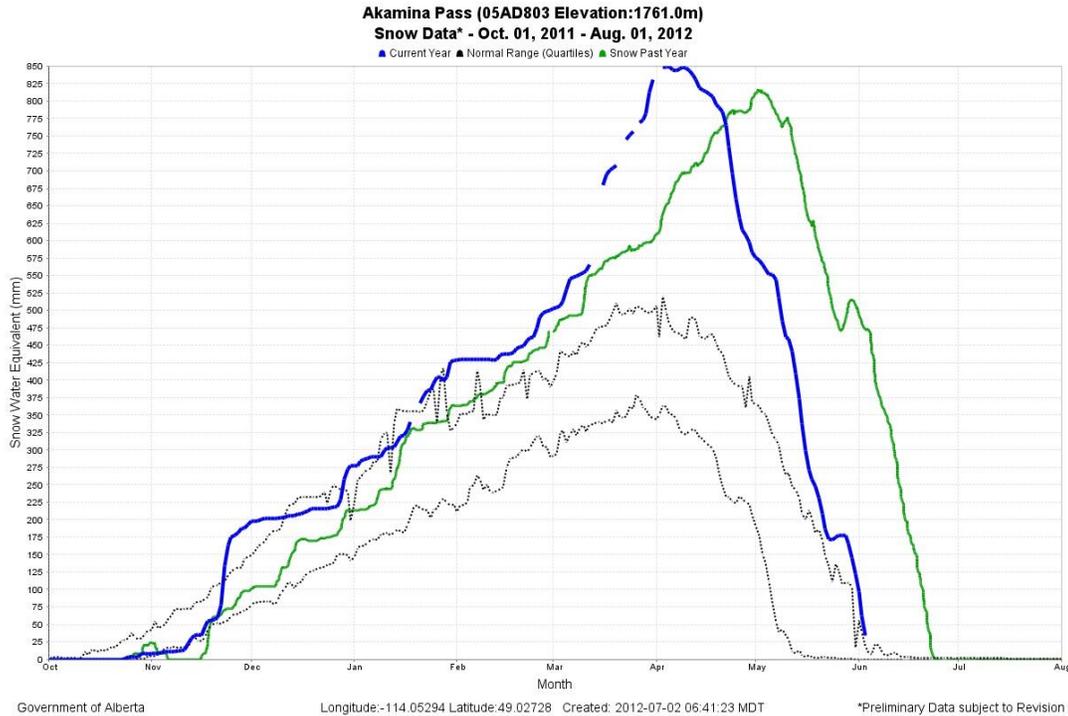


Figure 3.2: Snow data provided by the Government of Alberta. Normal quartile ranges for the snow depth in the Akamina Pass area of Waterton Lakes National Park are lower dotted lines and data for 2011 and 2012 are upper solid lines (Government of Alberta 2012).

Seedlings

Nine-hundred and ninety two 16-month-old whitebark pine seedlings were obtained from the USDA Forest Service nursery in Coeur d'Alene, Idaho. Seedlings selected for this study were grown from potentially rust-resistant seeds collected at Preston Park in Glacier National Park. Cones from the potentially resistant 'mother' tree have been entered into the USFS genetic testing program for white pine blister rust (Smith 2008, Smith 2012). At the Forest Service nursery seedlings were grown under standard nursery conditions in a substrate mix of 70% Canadian Sphagnum peat moss and 30% composted bark in long Ray Leach cone-tainers (3.8 cm x 21 cm). Seedlings

were fertilized every 8-12 days with a 4-25-35 NPK fertilizer at approximately 24-66-74 parts per million, respectively, and with STEM (Soluble Trace Element Mix) micronutrients.

Seedlings were transferred to the nursery at Glacier National Park (GNP) in April 2010 and fertilization was stopped. At the GNP nursery seedlings were placed in an outdoor nursery setting subject to natural environmental conditions. Seedlings were irrigated with a Rainbird automatic irrigation system and were watered until containers were saturated. Seedlings were inoculated at the outdoor nursery on August 19, 2010 and transported to Waterton Lakes National Park on September 27, 2010 by the GNP Revegetation Crew. For ease of transport, seedlings were removed from their containers, laid on plastic bubble wrap and bundled into groups of ten before transportation to Waterton Lakes National Park. Inoculated and un-inoculated seedlings were bundled separately to avoid exposing un-inoculated seedlings to mycorrhizal treatment.

Spore Slurry and Seedling Inoculation

The native ectomycorrhizal fungus *Suillus sibiricus* (CLC 2640) was selected for this experiment, because it is an obligate mycorrhizal associate of five-needle pines and a highly effective colonizer of whitebark pine seedlings in the greenhouse (Cripps and Grimme 2011). Spore slurries were made from cleaned fresh sporocarps. The hymenium was removed, cut into small pieces and ground for approximately 1 min in a coffee grinder with 10 ml of sterile distilled water. The resulting ground-up material was strained into approximately 400 ml of sterile distilled water and stored in glass bottles in a refrigerator with an average temperature of 4 °C. The spore content was then counted

using a hemocytometer and the slurry diluted to a spore count of approximately 1×10^6 spores/ml.

On August 19, 2010, 477 of the 992 whitebark pine seedlings were inoculated with slurry made from fresh sporocarps while the seedlings were at the Glacier National Park nursery. Inoculation was accomplished by injecting spore slurry directly onto the soil of containerized seedlings using an Allflex 50 ml repeat syringe commonly used for multiple dosing of livestock. Each seedling received approximately 3 ml of slurry with a spore content of 1×10^6 spores/ml for a total of 3 million spores/seedling. The remaining 515 seedlings were not inoculated. Seedlings were then tagged as inoculated or as controls.

Plot Layout and Design

Twenty one plots were established in the Summit Lake study area (Figure 3.3) as part of the whitebark pine restoration burn plan for Waterton Lakes National Park first outlined in 1999 (Schwanke and Smith 2010). Plots were selected by randomly generating GPS points within the study boundaries. Each GPS point marked the center of a circular plot with a 25-meter radius. Whitebark pine seedlings were planted in these previously monumented plots so that their survival could be monitored. Plots were used for the convenience of finding seedlings for monitoring purposes and not to delineate treatments.

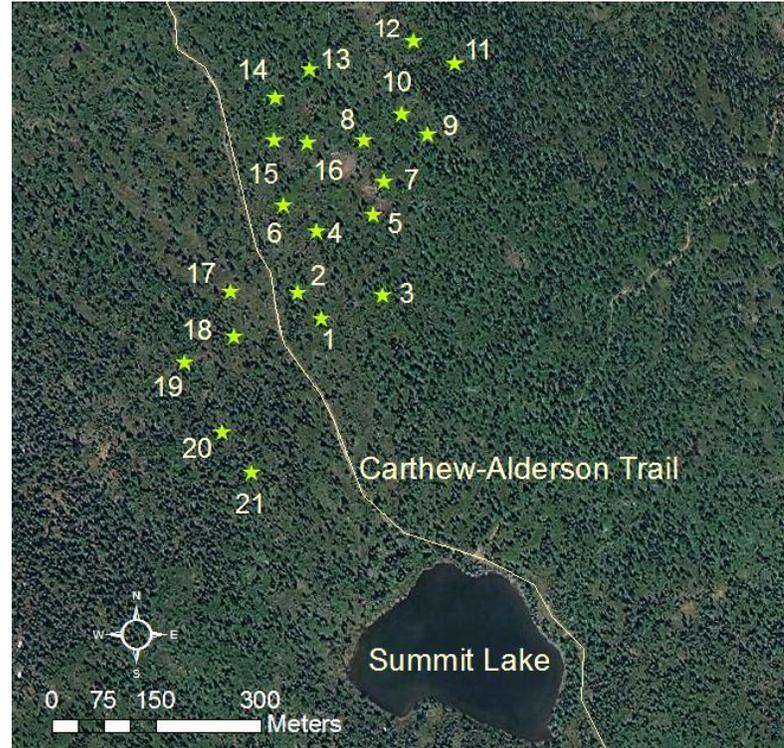


Figure 3.3: Field study site with locations of each plot within WLNP. Plots 1-21 are located between 1,950 and 2,000 meters in elevation. Map was generated using ArcGIS (ESRI 2011).

During the fall of 2009 and 2010, prescribed burns were used to simulate lightning strikes in 21 of these plots (Schwanke and Smith 2010). A custom built terra-torch, designed by the Park's Fire Specialist, was used to burn all competing vegetation within a 15-meter diameter of the torch, focusing on large Engelmann's spruce and subalpine fir (Figure 3.4). Effort was taken to avoid burning mature and regenerating whitebark pine trees and prescribed burns were only conducted during periods of low fire danger. Each plot contained a mosaic of burned and unburned areas (Schwanke and Smith 2010). Plots 1-12 were burned in 2009 and plots 13-21 in 2010. The goal of these simulated lightning strikes, as outlined in the 1999 plan, was to create openings in the canopy and reduce competing vegetation such as subalpine fir and Englemann spruce to

encourage natural WBP seedling regeneration and seed caching by Clark's nutcracker (Schwanke and Smith 2010).



Figure 3.4: Park personnel using a terra torch to simulate lightning strikes at the Summit Lake study area. Photo Courtesy of Cyndi Smith, Ecologist with Parks Canada.

On September 28, 2010, all 992 whitebark pine seedlings were planted in the 21 partially burned plots. Seedlings, still bundled in plastic bubble wrap, were transported to the study site via horseback and separated into two piles: inoculated and un-inoculated. At the study site, the Glacier revegetation crew with the help of volunteers randomly grouped the seedlings into clusters of three. Seedlings were planted in clusters of three to mimic the natural planting strategies of Clark's nutcracker. Each cluster of three included 0, 1, 2 or 3 inoculated seedlings. Different numbers of inoculated seedlings were included in each cluster to evaluate the minimum number of inoculated seedlings needed to

improve overall cluster survival. Seedling clusters were then planted in burned and unburned areas with and without the presence of beargrass (Figure 3.5, Figure 3.6).

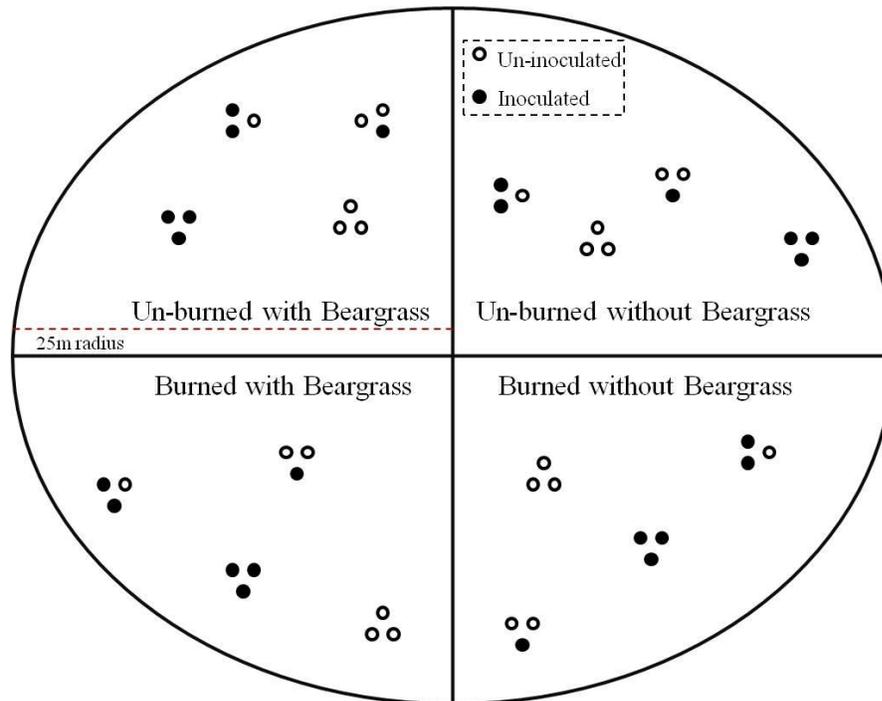


Figure 3.5: Schematic diagram of a plot showing site conditions and inoculation treatments.

Each plot typically contained all four site conditions and approximately four replicates of each of the four inoculation treatments were planted in each plot, for an average of 16 clusters per plot (Figure 3.5). A total of 321 clusters containing 992 seedlings were planted.



Figure 3.6: Four site conditions: a) burned without the presence of beargrass, b) burned with the presence of beargrass, c) un-burned without the presence of beargrass and d) un-burned with the presence of beargrass.

Plantings did not adhere to any strict layout, although seedlings were preferentially planted in areas appropriate for whitebark pine seedling survival. These ‘appropriate’ planting areas typically contained a microsite such as a stump, live whitebark pine tree, large rock, or large log that could potentially provide the planted seedlings with physical protection from solar radiation, wind, snow drifts, and soil erosion (Izlar 2007, McCaughey et al. 2009, Scott et al. 2011). An effort was made to ensure cluster treatments were distributed among the four site conditions, although unburned sites without beargrass were difficult to find.

During initial plot setup, the distance and azimuth from the center stake of the plot to each individual cluster was recorded using a meter tape and compass to aid in relocating the seedlings during monitoring. Data regarding the elevation, slope, aspect, and the year of each prescribed spot burn was also recorded for each plot (See appendix).

Monitoring Seedlings

Seedling survival was monitored in August of 2011 and 2012 for all 992 seedlings. Survival and health data were collected for individual seedlings as well as for clusters. The height of each individual seedling was also recorded. The type of burn, presence of beargrass, and the presence and type of microsite were also recorded for each cluster. Within each cluster, seedlings were designated as either A, B, or C. For example, with the assessor’s back to the center pin of the plot, seedling A is the left-most seedling in the cluster with B and C following in a clockwise direction.

For survival, individual seedlings were assessed as ‘dead’ or ‘alive’. Seedlings were recorded as dead when all the needles were brown. A health scale was used to rank

those seedlings still considered alive. Live seedlings were ranked as: almost dead (2), having no new growth (3), having some new growth (4), or having lots of new growth (5). Seedlings were recorded as missing when no trace of the seedling could be found, typically due to avalanches, frost heaving, or rodent activity. Seedling height (cm) was measured from the ground to the top of the canopy. Differences in seedling heights were reported from measurements taken from the base of the canopy to the top in order to account for inconsistencies in planting depth for each seedling as well as for soil movement during winter months.

For clusters, survival was recorded as 0/3 seedlings alive, 1/3 seedlings alive, 2/3 seedlings alive, or 3/3 seedlings alive. 'Type of burn' was recorded as burned or unburned. Burned sites fell into a range of severities which were categorized as low, moderate, high-low, high-moderate, or high-high. A complete description of these rankings can be found in the appendix. Clusters were recorded as being planted in beargrass if beargrass was found growing within 30 cm of a planted cluster. Clusters were recorded as being planted near a microsite if a live tree, stump, large rock, or large log was located within 30 cm of the planted cluster.

Statistical Analysis of Seedling Survival and Height

Data collection for this project focused on survival, height and health for each mycorrhizal inoculation treatment and site condition. For individual seedlings, mycorrhizal treatment was recorded as un-inoculated, exposed, or inoculated (Table 3.1). Seedlings were only considered as actually 'inoculated' if they initially received spore

slurry at the GNP out-door nursery. Un-inoculated seedlings in clusters containing 1 or 2 inoculated seedlings were considered ‘exposed’ to mycorrhizal treatment.

Table 3.1: The number of un-inoculated, exposed, or inoculated whitebark pine seedlings planted in each site condition.

Treatment	Unburned No Beargrass	Unburned Beargrass	Burned No Beargrass	Burned Beargrass
Un-inoculated	27 seedlings	54 seedlings	87 seedlings	92 seedlings
Exposed	16 seedlings	47 seedlings	99 seedlings	85 seedlings
Inoculated	41 seedlings	70 seedlings	174 seedlings	191 seedlings

For seedling clusters, mycorrhizal treatment was recorded as 0, 1, 2, or 3 seedlings inoculated/cluster (Table 3.2). Site condition was recorded as either burned with beargrass, burned without beargrass, unburned with beargrass, or unburned without beargrass. Microsite was recorded as present or absent. Mycorrhizal treatment, site condition, and microsite were used as explanatory variables in relation to the response variable for either survival or height.

Table 3.2: The number of whitebark pine seedling clusters planted in each site condition. Treatments consist of 0, 1, 2 or 3 inoculated seedlings per cluster.

Treatment	Unburned No Beargrass	Unburned Beargrass	Burned No Beargrass	Burned Beargrass
0	9 Clusters	18 Clusters	30 Clusters	31 Clusters
1	5 Clusters	17 Clusters	31 Clusters	27 Clusters
2	6 Clusters	13 Clusters	33 Clusters	32 Clusters
3	8 Clusters	10 Clusters	28 Clusters	33 Clusters

To analyze survival, generalized linear models were constructed in the statistical program R (R Development Core Team 2008). The response variable of whether a

seedling survived or not is denoted by either a 1 or 0 respectively, and is thus considered a binary response variable (binomial distribution). Graphical exploration of the data revealed varying relationships between seedling survival and site conditions, microsite and inoculation treatments. Interaction terms between all variables were added to the model to further investigate the relationships between these variables. Selective backwards elimination of terms was then used to determine an appropriate model. Using Wald's z-test followed by drop-in deviance tests for further confirmation, it was determined that interaction terms for burn (B) and beargrass (bg), microsite (ms) and beargrass, microsite and burn, inoculation treatment (I1, I2, I3) and burn, and inoculation treatment and beargrass were necessary. Logistic regression for a binary response variable was used to analyze the data with the following models:

1. $\text{logit}(\text{odds of survival}) = B_0 + B_1\text{burn} + B_2\text{bg} + B_3\text{ms} + B_4\text{I1} + B_5\text{I2} + B_6\text{I3} + B_7\text{burn*bg} + B_8\text{burn*ms} + B_9\text{bg*ms} + B_{10}\text{burn*I1} + B_{11}\text{burn*I2} + B_{12}\text{burn*I3} + B_{13}\text{bg*T1} + B_{14}\text{bg*T2} + B_{15}\text{bg*T3}$
2. $\text{logit}(\text{odds of survival}) = B_0 + B_1\text{burn} + B_2\text{bg} + B_3\text{ms} + B_4\text{exposed} + B_5\text{burn*bg} + B_6\text{burn*ms} + B_7\text{bg*ms} + B_8\text{burn*exposed} + B_9\text{bg*exposed}$

The mean height of whitebark pine seedlings was analyzed as a function of site condition, microsite, and inoculation treatment using a 4-way Analysis of Variance (IBM 2011). The 4-way ANOVA included 4 main effects – burn, beargrass, microsite, and inoculation treatment and all possible interaction effects. Levene's test of equal variance as well as box plots both indicated that the assumption of equal variance was adequately met and transformation was not necessary.

Results

Overall Seedling Survival

In 2011, the first year after out-planting, whitebark pine seedling survival averaged 95% across all treatments. In 2012, the second year after out-planting, whitebark pine seedling survival averaged 69% across all treatments; this represents a 26% decrease in seedling survival between the 1st and 2nd year after out-planting (Figure 3.7).

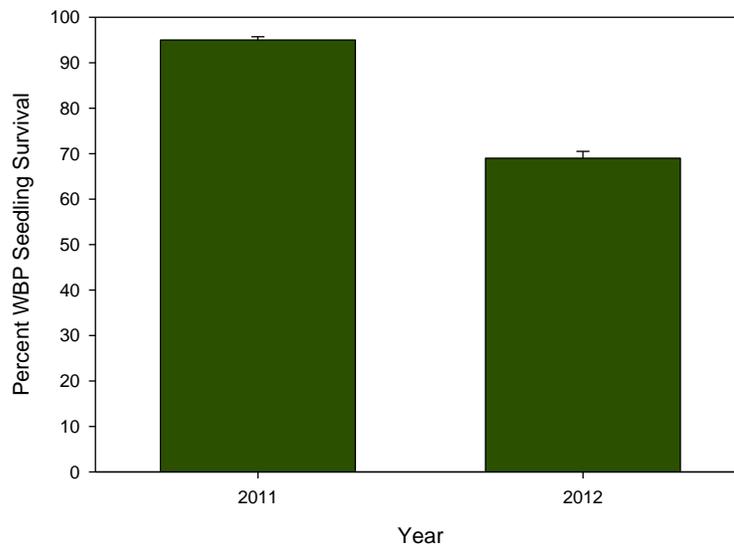


Figure 3.7: Average percent whitebark pine seedling survival one (2011) and two (2012) years after out-planting at the Summit Lake Study site, WLNP. Graph includes 95% confidence bars.

Seedling survival was assessed for each plot to determine whether there were any spatial effects. Seedling survival was generally consistent across all plots in 2012, except for plots 2, 12 and 21 which had noticeably lower survival rates than all the other plots (Figure 3.8a). Plots 2 and 21 had a higher proportion of seedlings planted in un-burned

areas (Figure 3.8b) and plot 21 had few clusters planted in it. Plots 2, 12, and 21 also had fewer seedling planted near microsities in comparison to other sites.

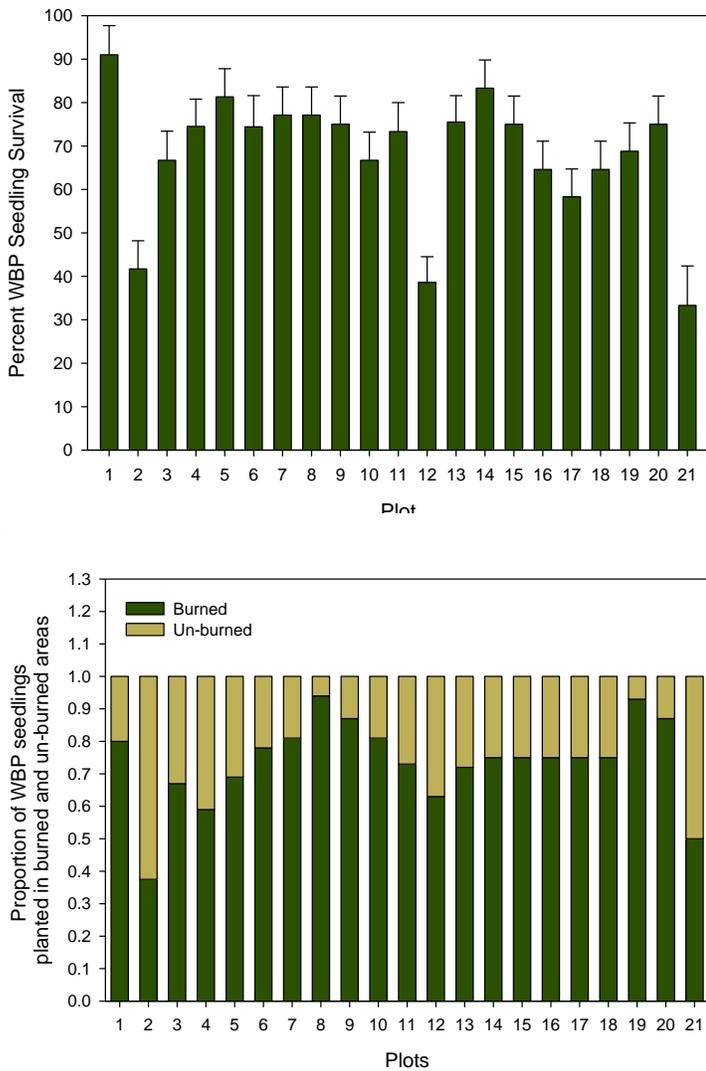


Figure 3.8: a) Percent whitebark pine seedling survival averaged across clusters. Plots 1-12 were burned in 2009 and plots 13-21 were burned in 2010 and assessment is for 2012. b) Proportion of whitebark pine seedlings planted in burned and unburned areas for each plot. Plots 2 and 21 had a higher portion of seedlings planted in un-burned areas. Plot 21 also had a low number of seedling clusters. Graph (a) includes 95% confidence bars.

Seedling Survival by Site Condition

Survival was high (> 90%) across all site conditions after one year; however a drop in survival occurred across all site conditions from year one to two, but the drop was generally greater on un-burned sites (Figure 3.9). The survival of whitebark pine seedlings was affected by both burn treatment and the presence of beargrass (Figure 3.9). After two years, seedlings planted in burned areas without beargrass had a higher survival rate (~ 82%) than seedlings planted in burned areas near beargrass (~66%). However, in un-burned areas seedlings planted near beargrass had a higher survival rate (~ 62%) than seedlings not planted near beargrass (~ 38%).

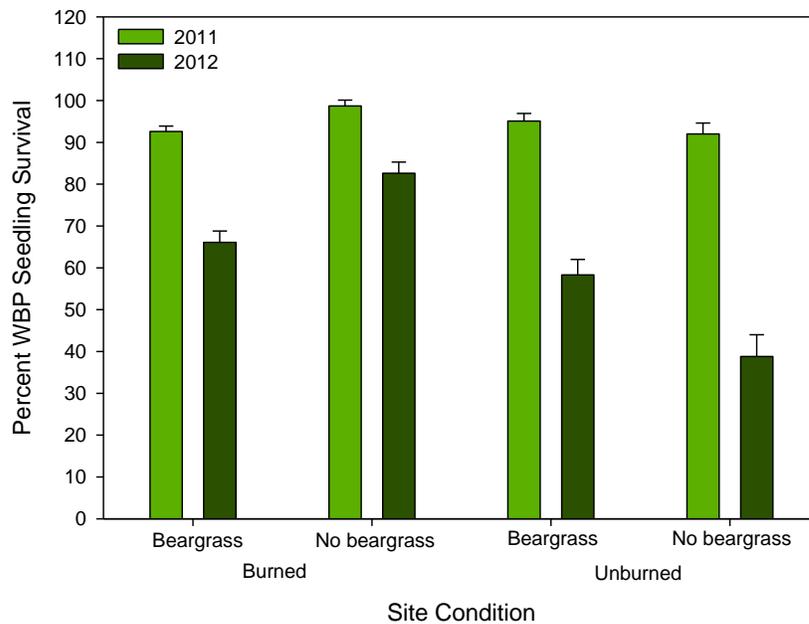


Figure 3.9: Average whitebark pine seedling survival for the 1st (2011) and 2nd (2012) year after out-planting as a function of site condition at the Summit Lake study site, WLNP. Graph includes 95% confidence bars.

In general, two years after planting, whitebark pine seedling survival was higher for seedlings planted in burned areas (Figure 3.10). Out-planted whitebark pine seedling survival in burned areas averaged 70.3% whereas seedlings planted in un-burned areas had a survival average of 51% (Figure 3.10). Overall, differences in survival for seedlings planted with and without beargrass were slight (8.5%), and strongly dependent on site condition (burned/unburned).

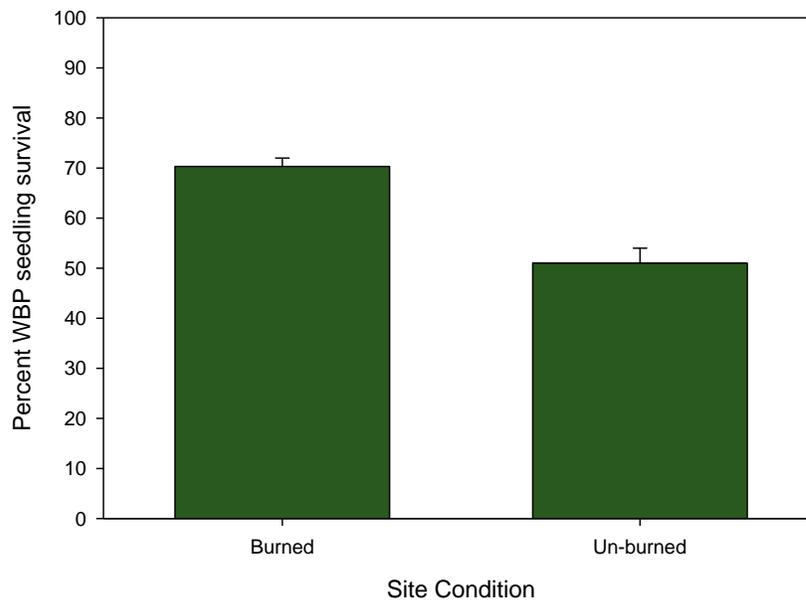


Figure 3.10: Average whitebark pine seedling survival as a function of burn treatment. Graph includes 95% confidence bars.

Seedling Survival by Microsite

On all four site conditions the presence of a microsite significantly increased seedling survival (Figure 3.11). On burned areas, the presence of a microsite increased survival 10% when beargrass was present ($p=0.05$ from DinD on 1 d.f.) and 12.6% when beargrass was not present ($p=0.002$ from DinD on 1 d.f.). On unburned areas, the

presence of a microsite increased survival 10.7% when beargrass was present (two-sided p-value = 0.06 from DinD on 1 d.f.) and 34.5% when beargrass was not present (two-sided p-value = 0.001 from DinD on 1 d.f.).

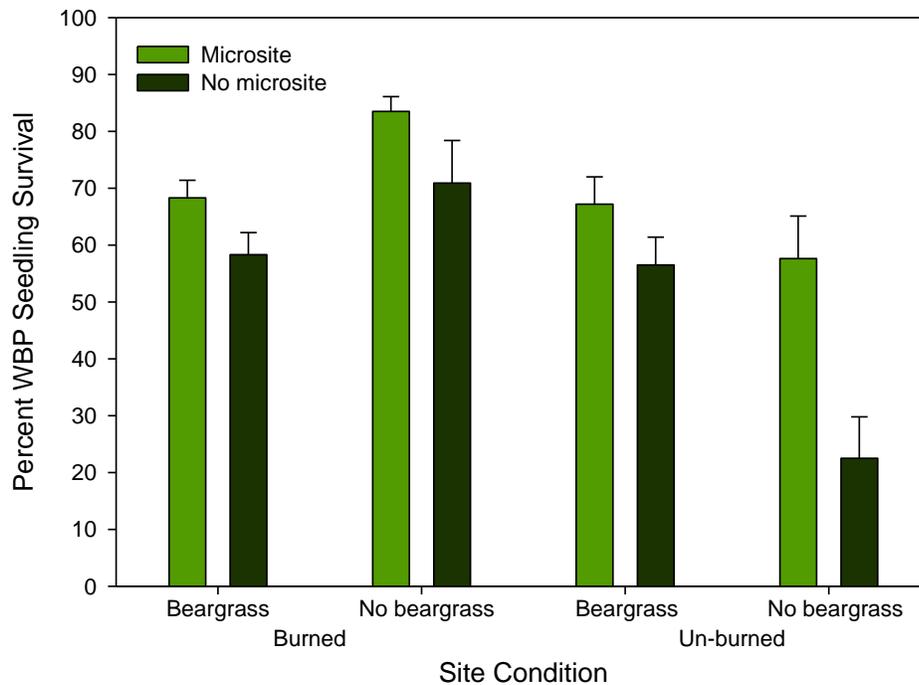


Figure 3.11: Average whitebark pine seedling survival as a function of site condition and microsite. Graph includes 95% confidence bars.

Survival of Individual Seedlings by Mycorrhizal Treatment

Results show that the effect of inoculation treatment on seedling survival is dependent on site condition and significant interactions between burn, beargrass, and inoculation treatment were observed (Figure 3.12, Table 3.3).

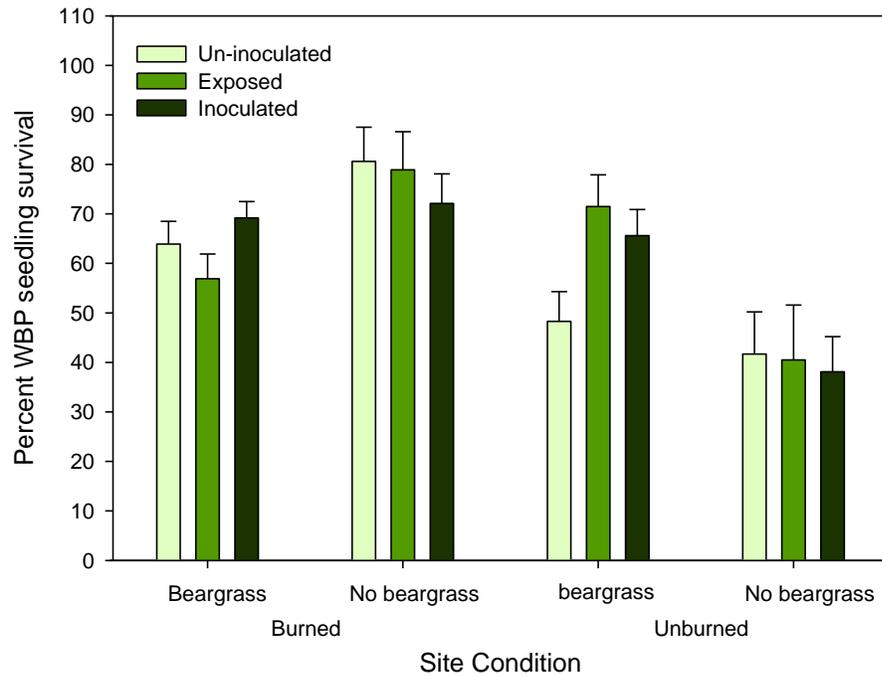


Figure 3.12: Average percent whitebark pine seedling survival as a function of site condition and mycorrhizal treatment assessed after two years. ‘Exposed’ indicates seedlings planted in clusters next to inoculated seedlings. Graph includes 95% confidence bars.

Table 3.3: The odds of survival and significance values for inoculated and exposed in comparison to survival of un-inoculated whitebark pine seedlings as a function of site condition.

Site Condition	Treatment	Survival Odds	p-value	Average % Survival
Un-burned with Beargrass	Exposed	2.22	0.03	71.5
	Inoculated	1.83	0.07	65.6
Un-burned without Beargrass	Exposed	1.64	0.29	40.5
	Inoculated	0.99	0.92	38.1
Burned with Beargrass	Exposed	0.75	0.33	56.9
	Inoculated	1.33	0.26	69.2
Burned without Beargrass	Exposed	0.56	0.11	78.9
	Inoculated	0.70	0.28	72.1

Unburned Areas: When seedlings are out-planted in un-burned areas near beargrass, evidence indicates that the odds of survival differ significantly between un-inoculated whitebark pine seedlings and those exposed to mycorrhizal treatment (p-value = 0.03, from DinD on 1 d.f.); (Table 3.3). In un-burned areas near beargrass, survival increased 23.2% when seedlings were exposed to the mycorrhizal treatment (Figure 3.12). This corresponds to a 2.22 times increase in the odds of survival when whitebark pine seedlings are out-planted in un-burned areas near beargrass, with an associated confidence interval of 1.06 - 4.70 times greater (Table 3.3). There is also some evidence of a difference between un-inoculated and inoculated seedlings (p-value = 0.07, from DinD on 1 d.f.); inoculated seedlings showed an increase in survival of 17.3% corresponding to 1.83 times increase in the odds of survival.

However, when seedlings were planted in un-burned areas without the presence of beargrass there was no evidence of a difference in survival between either un-inoculated seedlings and those exposed to mycorrhizal treatment or un-inoculated and inoculated seedlings (p-value = 0.29 and 0.92, from DinD on 1 and 1 d.f. respectively). Overall, seedlings planted in un-burned areas without the presence of beargrass had the lowest average survival rate at 40% regardless of mycorrhizal treatment (Figure 3.12).

Burned Areas: The difference in survival between un-inoculated seedlings and those either inoculated or exposed to mycorrhizal treatment was minimal regardless of the presence of beargrass on burned areas (Figure 3.12). There is no evidence that the odds of survival differ between un-inoculated seedlings and either inoculated seedlings or those exposed to mycorrhizal treatment when whitebark pine seedlings are out-planted in

burned areas with or without the presence of beargrass (two-sided p-value=0.47 and 0.26, from DinD on 1 and 1d.f. respectively). Overall, seedlings planted in burned areas without the presence of beargrass had the highest average survival rate at 77% regardless of mycorrhizal treatment after two years.

Evaluation of Seedling Survival by Cluster Inoculation Treatment

Figure 3.13 shows no consistent trends for mycorrhizal treatments from I0 to I3 across the four sets of site conditions. For clusters planted in unburned areas near beargrass, survival was higher when clusters contained any number of inoculated seedlings in comparison to un-inoculated clusters (Table 3.4). When clusters contained two inoculated seedlings survival significantly increased 24.7% in un-burned areas with beargrass ($p= 0.02$ from DinD on 1 d.f.); (Figure 3.13, Table 3.4). This corresponds to a 2.51 times increase in the odds of survival when clusters containing 2 inoculated seedlings were out-planted in un-burned areas near beargrass, with an associated confidence interval of 1.16-5.59 times greater. Overall, clusters with ectomycorrhizal inoculation or exposure to inoculated seedlings showed a 19.3% increase in survival compared to un-inoculated clusters planted in un-burned areas near beargrass.

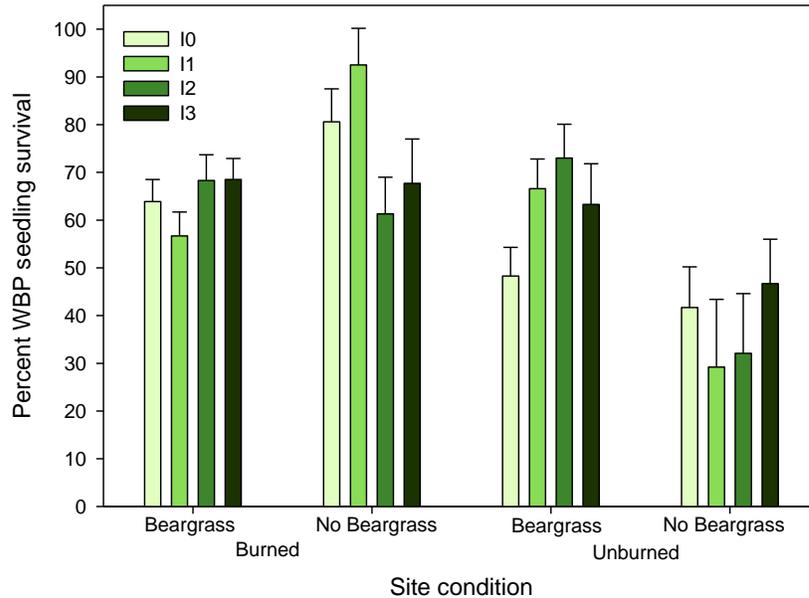


Figure 3.13: Average WBP seedling survival as a function of site condition and cluster inoculation treatment. Graph includes 95% confidence bars.

Table 3.4: The odds of survival and significance values comparing un-inoculated whitebark pine clusters to clusters with either 1, 2, or 3 inoculated seedlings as a function of site condition. Results in bold are significant at the $\alpha = 0.05$ level.

Site Condition	Inoculation Treatment	Survival odds	P-value	Average % Survival
Un-burned with Beargrass	I1	1.66	0.16	66.6
	I2	2.51	0.02	73.0
	I3	1.87	0.13	63.3
Un-burned without Beargrass	I1	1.74	0.24	29.2
	I2	0.79	0.62	32.1
	I3	1.17	0.74	46.7
Burned with Beargrass	I1	0.80	0.41	56.7
	I2	1.37	0.30	68.3
	I3	1.23	0.47	68.5
Burned without Beargrass	I1	0.84	0.87	92.5
	I2	0.43	0.02	61.3
	I3	0.77	0.50	67.7

The survival of all seedling clusters planted in un-burned areas without the presence of beargrass from I0 to I3 was lower than for all other treatments.

Proportionally, more whole clusters died in these areas (37%) in comparison to other site conditions and dead clusters typically lacked microsites. There were no significant differences at the $\alpha=0.05$ level for I1-I3 compared to un-inoculated seedlings, although the odds of survival for clusters with one inoculated seedling was almost twice (1.74) those of un-inoculated seedlings ($p=0.24$ from DinD on 1 d.f.).

Survival was higher for clusters planted in burned areas without beargrass for I0, I1 and I3 than for any of the other treatments. There was no evidence of a difference in survival between un-inoculated clusters and clusters with either 1 or 3 inoculated seedlings ($p=0.65$ and 0.50 from DinD on 1 and 1 d.f. respectively). However, the odds of seedling survival are lower in clusters with two inoculated seedlings and this value is significant when compared to seedlings in clusters with no inoculated seedlings ($p=0.020$ from DinD on 1 d.f.); (Figure 3.13). Overall survival was 12.2% less when 2 of 3 seedlings were inoculated. For clusters with 2 inoculated seedlings this corresponds to a 0.43 times decrease in the odds of survival when out-planted in un-burned areas near beargrass, with an associated confidence interval of 0.21-0.86 times less (Table 3.4).

Seedling Parameters

Average seedling height was increased under all site conditions when a microsite was present, although the increase was only significant in burned areas (Figure 3.14). In burned areas, the presence of a microsite increased average height 1.1 cm when beargrass was present ($f_{1, 959}=4.66$, $p=0.005$) and 1.3 cm when beargrass was not present ($f_{1, 959}=$

8.07, $p=0.03$) with associated confidence intervals of 0.99-1.28 cm and 0.51-2.09 cm greater, respectively.

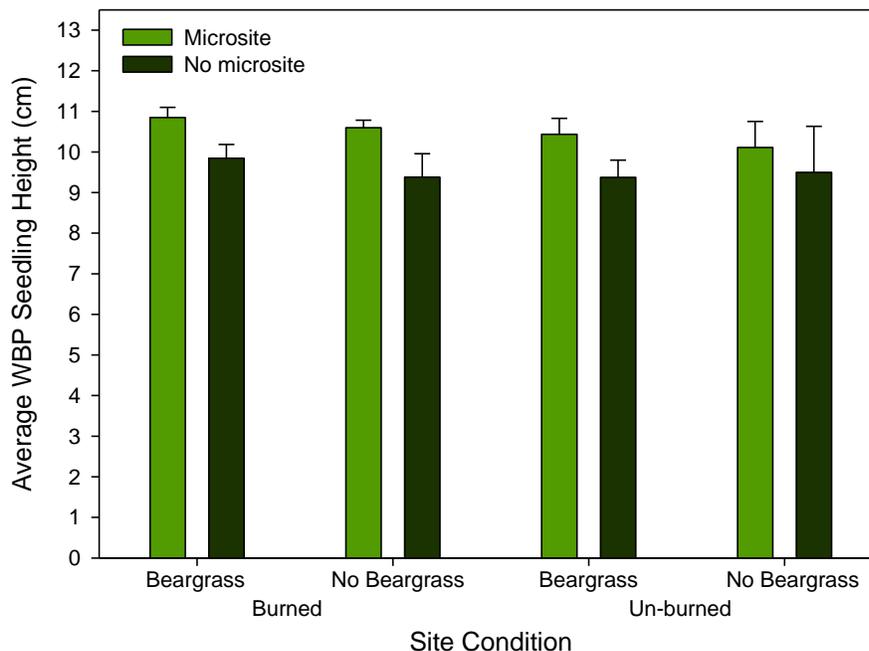


Figure 3.14: Average height of surviving seedlings (cm) as a function of site condition and presence of a microsite. Graph includes 95% confidence bars.

Differences in height were minimal across mycorrhizal treatments (Figure 3.15, Table 3.5) with only four of twelve values showing significance (Table 3.6). In all four cases inoculated seedlings showed a height increase over un-inoculated or exposed seedlings. For seedlings planted in un-burned areas near beargrass, there were no differences in height, except between exposed and inoculated seedlings ($p=0.03$ from LSD on 1 d.f.); (Figure 3.15). Inoculated seedlings were an average of 1.40 cm taller than ‘exposed’ seedlings, with an associated confidence interval of 1.26 - 1.53 cm taller (Table 3.5). When seedlings are planted in un-burned areas without the presence of beargrass

there is no evidence of a difference in average height between mycorrhizal treatments (Figure 3.15, Table 3.6).

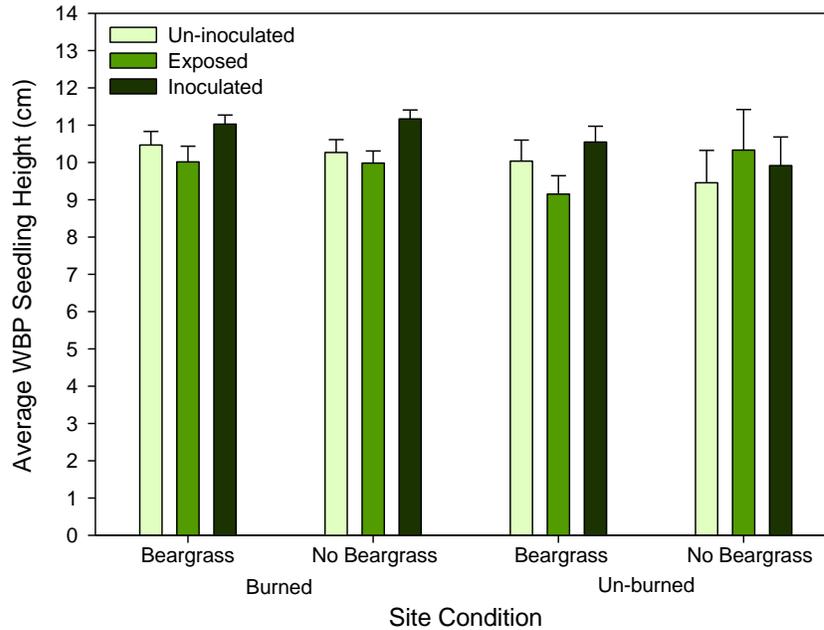


Figure 3.15: Average whitebark pine seedling height (cm) as a function of site condition and mycorrhizal treatment. Graph includes 95% confidence bars.

Table 3.5: The average height (cm) for un-inoculated, exposed, and inoculated whitebark pine seedlings as a function of site condition.

Site Condition	Inoculation Treatment	Average Height (cm)
Un-burned with Beargrass	Un-inoculated	10.04
	Exposed	9.15
	Inoculated	10.55
Un-burned without Beargrass	Un-inoculated	9.46
	Exposed	10.33
	Inoculated	9.91
Burned with Beargrass	Un-inoculated	10.46
	Exposed	10.01
	Inoculated	11.03
Burned without Beargrass	Un-inoculated	10.27
	Exposed	9.98
	Inoculated	11.17

Table 3.6: Pairwise comparison of average height (cm) between un-inoculated, exposed, and inoculated whitebark pine seedlings as a function of site condition.

Site Condition	Comparison	Significance Value
Un-burned with Beargrass	Un-inoculated vs. Exposed	0.24
	Un-inoculated vs. Inoculated	0.47
	Inoculated vs. Exposed	0.03
Un-burned without Beargrass	Un-inoculated vs. Exposed	0.53
	Un-inoculated vs. Inoculated	0.69
	Inoculated vs. Exposed	0.76
Burned with Beargrass	Un-inoculated vs. Exposed	0.42
	Un-inoculated vs. Inoculated	0.20
	Inoculated vs. Exposed	0.04
Burned without Beargrass	Un-inoculated vs. Exposed	0.55
	Un-inoculated vs. Inoculated	0.03
	Inoculated vs. Exposed	0.003

For seedlings planted in burned areas near beargrass, there were no differences in height except between exposed and inoculated seedling ($p = 0.04$ from LSD on 1 d.f.); (Figure 3.15). Inoculated seedlings were on average 1.02 cm taller than seedlings only exposed to mycorrhizal treatment for this site condition, with an associated confidence interval of 0.67-1.36 (Table 3.5 and 3.6). For whitebark pine seedlings planted in burned areas without the presence of beargrass there is evidence of a difference in the average height between mycorrhizal treatments ($p = 0.007$, $f\text{-stat} = 5.040$ on 2 d.f.); (Figure 3.15). In burned areas without the presence of beargrass, inoculated whitebark pine seedlings were on average 0.90 cm taller than un-inoculated seedlings and 1.19 cm taller than seedlings exposed to mycorrhizal treatment, with associated confidence intervals of 0.70-1.10 cm and 1.02-1.35 cm, respectively (Table 3.5 and 3.6).

Whitebark pine seedlings planted in burned areas tended to have a slightly higher average health score in comparison to seedlings planted in un-burned areas (Figure 3.16). However, there is no obvious health trend for whitebark pine seedlings as a function of mycorrhizal treatment within site conditions (Figure 3.16). Seedling health was also somewhat higher in burned areas when seedlings were planted near microsites, but this was not true on un-burned sites (Figure 3.17).

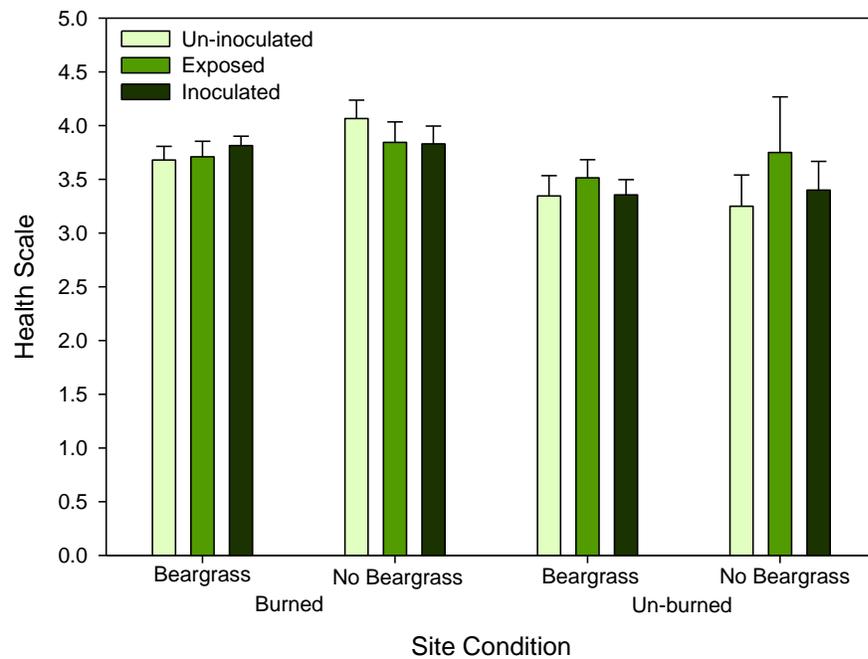


Figure 3.16: Average health of surviving of whitebark pine seedlings as a function of mycorrhizal treatment and site condition. Graph includes 95% confidence bars.

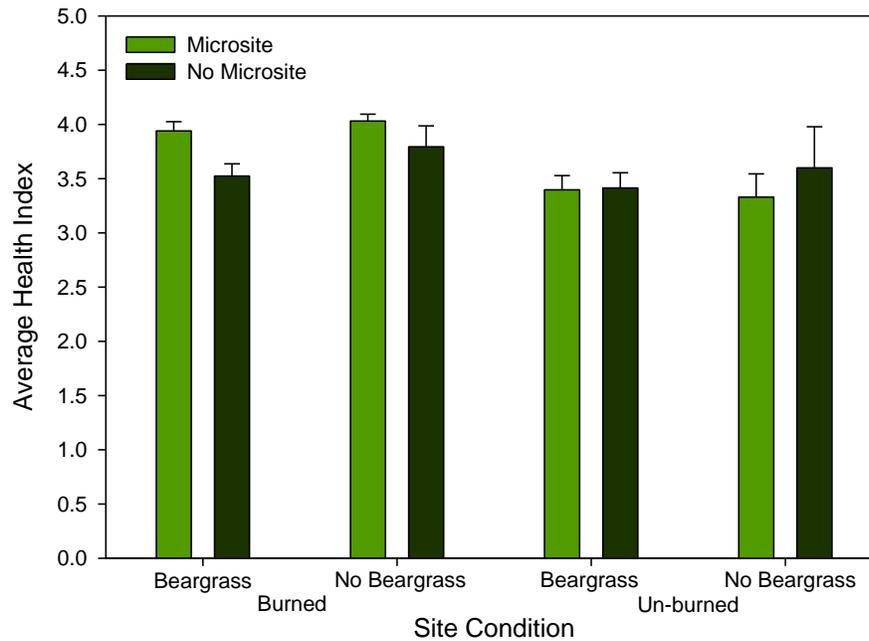


Figure 3.17: Average health of surviving of whitebark pine seedlings as a function of microsite and site condition. Graph includes 95% confidence bars.

Discussion

Overall Survival

Currently, whitebark pine reforestation relies on the planting of potentially rust-resistant, two-year old nursery grown seedlings; hundreds of thousands of seedlings have been planted in the Western U.S. and Canada (Izlar 2007, Keane et al. 2012). Monitoring efforts have been scarce, often limited by financial resources and only a few studies have reported on the survival of out-planted nursery grown whitebark pine seedlings in the Rocky Mountain region (Carolin 2006, Izlar 2007, Asebrook et al. 2011, Keane et al. 2012). The current study is the only one to document the survival of whitebark pine seedlings intentionally inoculated with ectomycorrhizal fungi.

In our field trial, first year survival for out-planted whitebark pine seedlings was higher than reported by other studies in the Rocky Mountain region, averaging 95% one year after out-planting. In a much larger study, first year survival rates for out-planted whitebark pine seedlings averaged 74% for over 100,000 nursery-grown, whitebark pine seedlings planted on ridge tops, mountain slopes, high mountain valleys, moraines, and benches in 37 plots ranging in elevation from 5,045 to 9,500 feet and located across 9 national forests, 2 national parks, and 1 BLM district in Montana, Idaho, and Wyoming (Izlar 2007). In a smaller out-planting study on Dunraven Pass in Yellowstone National park, first year survival rates averaged 68% (Izlar 2007). Similarly, 1st year survival rates for whitebark pine seedlings out-planted in the Red Eagle (79%) and Flattop Mountain (52%) areas of Glacier National Park were lower than that observed at the Summit Lake study Site (Asebrook et al. 2011). The high first-year survival rate at the Summit Lake study site could be in part due to wet and favorable spring conditions followed by a mild summer in 2011. Average spring precipitation was 30-40 cm above average in 2011 and the summer season was short and relatively cool (Government of Alberta 2012). Other studies have reported higher mortality rates at sites with high sun exposure, suggesting that hot and dry conditions may not be conducive to seedling survival (Izlar 2007, Asebrook et al. 2011).

In 2012, the second year after out-planting, whitebark pine seedling survival dropped dramatically from 95% to 69%. This result is consistent with other studies which observed a rapid decrease in whitebark pine seedling survival between the 1st and 3rd-15th years after out-planting (Mellmann-Brown 2005, Izlar 2007, Asebrook et al. 2011).

Survival rates of out-planted whitebark pine seedlings appear to decrease quickly the first 3-5 years after out-planting and then typically stabilize (Izlar 2007, Asebrook et al. 2011). Four years after out-planting, survival rates for seedlings planted in GNP dropped to 38%, however subsequent mortality averaged only 1% per year for the next three years (Asebrook et al. 2011). In Izlar's (2007) study, survival rates for the 3rd-15th years after out-planting also averaged around 38% (Izlar 2007). Whitebark pine seedling survival (69%) assessed after two years at the Summit Lake study site was still higher than that reported by other whitebark pine monitoring studies for this time period (Carolin 2006, Izlar 2007, Asebrook et al. 2011).

This study summarizes seedlings survival during the early acclimation phase of out-planted whitebark pine seedlings at year one and two. Valuable insights can be gained on the early establishment of whitebark pine seedlings planted for restoration purposes; however, the ultimate impact of restoration treatments on whitebark pine seedling survival may not be apparent until later years. Whitebark pine is a slow growing conifer and long term monitoring will be necessary to adequately assess the effectiveness of restoration techniques, such as ectomycorrhizal inoculation, microsite, and site conditions on seedling survival.

Survival by Site Conditions

The survival of whitebark pine seedlings was about 20% higher when seedlings were planted in burned patches (70.3%) rather than in un-burned areas (51%). The average seedling health index also increased slightly when seedlings were planted in burned areas instead of un-burned areas (Health Index in Appendix). Izlar (2007) also

reported that whitebark pine seedlings planted in mixed severity burns had a higher survival rate (52%) than seedlings planted in unburned areas (21%). Directly planted whitebark pine seeds have been reported to have higher germination rates and increased biomass when planted in burned areas (Perkins 2004). However, McCaughey (1990) found germination rates of directly planted seeds to be comparable between litter soils and burned areas. The health and survival of limber pine seedlings (*Pinus flexilis* James), another five-needle pine of the Rocky mountain region, has also been shown to increase when seedlings are planted in burned areas (Smith et al. 2011a).

The effects of wild and prescribed fire have been evaluated for natural whitebark pine regeneration as well (Tomback et al. 2001b, Keane and Parsons 2010, Larson and Kipfmüller 2010). Seven years after the 1988 Yellowstone fires, natural whitebark pine seed germination rates were similar on burned and unburned sites, although germination was lowest on the moist, unburned sites (Tomback et al. 2001b). Other studies also found natural regeneration rates on burns from wildfires to be similar to that on unburned sites in Montana, Idaho, Oregon, and the Canadian Rockies (Tomback et al. 2001b, Moody 2006, Larson and Kipfmüller 2010). For prescribed burning, natural regeneration has been low for at least five years following treatment; however prescribed burns have been effectively used to create canopy openings that reduce competition from shade tolerant conifers (Keane and Parsons 2010). In addition, Clark's nutcrackers have been observed caching seeds on both high and moderate prescribed burn treatments when a sufficient cone crop surrounded the site (Keane and Parsons 2010). However, in relatively small

burned areas, nutcrackers can retrieve a majority of cached seeds for consumption (Keane and Parsons 2010).

Historically, whitebark pine forests in the Rocky Mountains are characterized by mixed severity burns with a fire frequency ranging between 30-300 years (Arno and Hoff 1990, Morgan et al. 1994). However, until recently Forest Service policy favored fire suppression which encouraged the establishment of more shade tolerant tree species subsequently creating uncharacteristically dense forests that now have reduced habitat for seed caching and seed germination of whitebark pine (Keane and Arno 1993, Morgan et al. 1994). Fire plays a critical role in the maintenance and regeneration of whitebark pine forests by reducing competition from shade tolerant conifers, creating areas for nutcracker caching, and by creating open areas for seedling growth (Tomback et al. 2001c, Keane and Parsons 2010). Fire may also benefit planted whitebark pine seedlings by improving soil properties and in essence lengthening the growing season because of raised soil temperatures (Tomback et al 2001b, Izlar 2007). In addition, soils typically experience a pulse of higher mineral concentrations following a fire, specifically ammonium, nitrate and phosphorus; thus recently burned soils could offer more available nutrients to developing seedlings (Certini 2005, Wan et al. 2011).

At the Summit Lake study area a 'terrestrial torch' was used to burn small plots of mature subalpine fir and Engelmann spruce to mimic lightning strikes (Schwanke and Smith 2010). A truck-mounted terra-torch has been used previously in whitebark pine restoration, primarily to create openings for seed caching and natural regeneration (Keane and Parsons 2010). Nutcracker activity is most abundant in disturbed or non-forest

patches between 1 and 40 acres (Norment 1991) and most previous prescribed burns for whitebark pine restoration fit this size scale (Keane and Parsons 2010). The use of a terrestrial torch at the Summit Lake study area was unique in that burned patches were typically small, averaging less than 25 meters in diameter. The use of a terrestrial torch to create small burned patches that mimic lightning strikes may be a viable restoration technique if long-term results support early high survival rates resulting from this technique.

The presence of beargrass also influenced the survival of out-planted whitebark pine seedlings, but results differed between burned and un-burned areas at the Summit Lake study site. Beargrass is a dominant understory plant at the site and dense stands cover most areas; the above-ground biomass was greatly reduced in burned areas although the rhizomatous root system was likely still intact. The presence of beargrass (or at least the presence of its rhizomatous root mass) did have a negative impact on whitebark pine seedling survival on burned sites, although survival rates were still high (66%). This is consistent with other studies that found a decrease in survival for whitebark pine seedlings planted near beargrass (Perkins 2004, Izlar 2007). Perkins (2004) reported that whitebark pine seedlings also had a lower biomass when planted near Geyer's sedge (*carex geyeri*), another understory plant found with whitebark pine in the Rocky Mountain region. Competition from woody and herbaceous plants for light, water, and nutrients can seriously threaten the survival of young conifer trees (Wagner et al. 1989, Lieffers et al. 1993, Landhauser et al. 1996, Hangs et al. 2002, Parladé et al. 2004). Currently, whitebark pine planting guidelines suggest avoiding planting seedlings

near beargrass or other highly competitive vegetation so that competition for nutrients and soil moisture can be minimized (Scott et al. 2011).

Both beargrass and Geyer's sedge are capable of reproducing vegetatively from underground rhizomes following fire (Fischer and Bradley 1987). In fact, native basket weavers annually burned beargrass gathering sites to maintain its productivity (Shebitz et al. 2009). Beargrass has a stout shallow rhizome system that allows it to re-sprout quickly following light broadcast burns, but the species can be decreased by high severity fires that burn the duff layer (Arno et al. 1985, Hollingsworth 2010). However, one study found a significant increase in beargrass seedling establishment and vegetative reproduction following high intensity prescribed fire on the Olympic peninsula (Shebitz et al. 2009). Populations of Geyer's sedge typically increase following fire (Fischer and Bradley 1989). Geyer's sedge is highly adapted to fire and tends to rapidly invade burned areas forming dense stands (Bradley et al. 1992). The ability of both beargrass and Geyer's sedge to tolerate fire may be of concern in whitebark pine restoration as both species have dense rhizomatous root systems that may have restrictive effects on conifer establishment (Crane 1990, Bradley et al. 1992, Perkins 2004, Izlar 2007). Subsequent monitoring at the Summit Lake study site can help determine how stimulation of certain understory plants with burning affects pine establishment in the future.

Other herbaceous species with rhizomatous growth habits have also been shown to suppress conifer establishment (Amaranthus and Perry 1989, Amaranthus et al. 1993, Landhausser et al. 1996). Bluejoint reedgrass (*Calamagrostis Canadensis* (Michx.) Beauv.) is a highly competitive native grass species that is an important competitor of

white spruce (*Picea glauca* (Moench.) Voss) and poses serious problems for conifer reforestation (Lieffers et al. 1993, Landhauser et al. 1996). Similarly, the survival of sugar pine seedlings (*Pinus lambertiana*) out-planted in Oregon following a wildfire was reduced due to competition from seeded grasses (Amaranthus et al. 1993). In addition, weed competition can lead to a dramatic reduction in pine root development for at least three years after out-planting (Sylvie and Jarstfer 1997).

In un-burned areas, the presence of beargrass appeared to result in higher whitebark pine seedling survival (62%) in comparison to sites where it was not present two years after out-planting (38%). At planting, a small circular section of the rhizomatous root system was removed at the spot where seedlings were planted using a hoedad. It is possible that the surrounding beargrass on these un-burned sites contributed to higher seedling survival because the vegetation mat helped retain soil, nutrients, and moisture creating suitable microclimate for whitebark pine seedlings (Coop and Schoettle 2009, Smith et al. 2011a). Also, beargrass may have served to protect seedlings against harsh environmental conditions, such as heavy creeping snow packs and desiccation from solar radiation and winds during the initial stages of seedling establishment (Castro et al. 2002). However, mycorrhizal inoculation appears to have played a significant and positive role in seedling survival on these site conditions. Long term monitoring is needed to determine if this benefit is persistent or if competition from the beargrass will eventually negate results. In addition, results are based on a comparison to un-burned areas without beargrass.

Un-burned sites lacking beargrass resulted in the lowest survival rate for whitebark pine seedlings of all site conditions (38%). It should be noted that un-burned areas devoid of beargrass were difficult to find since beargrass forms a dense understory at the Summit Lake study site. These sites were typically in full sun, devoid of most other vegetation, and had shallow and rocky soils. The shallow, rocky soils made planting difficult which could have led to increased frost heaving and seedling desiccation. Indeed, there was a substantial loss of whole clusters on these sites. The lack of vegetation and other established root systems may have promoted the loss of soil nutrients through leaching during periods of high precipitation (Smith et al. 2011a). Furthermore, because these sites were much more exposed, seedlings could have experienced greater temperature extremes, stronger winds, and heavier snow packs. It is possible that extreme abiotic conditions created an inhospitable environment for seedling establishment as well as fungal survival on un-burned sites lacking beargrass. However, there was a strong microsite effect for this site condition with the presence of a microsite dramatically increasing seedling survival (34.5%) as discussed in a subsequent section.

Whitebark pine is a long-lived seral species of the *Pinus albicaulis/Vaccinium scoparium* habitat type (Arno and Hoff 1990). Grouse whortleberry (*Vaccinium scoparium*) is consistently the most dominant shrub in whitebark pine communities throughout the Rocky Mountains (Weaver and Dale 1974, Arno and Hoff 1990). Planting guidelines suggest that this habitat type requires little if any site preparation prior to out-planting whitebark pine seedlings (Scott et al. 2011). Survival rates have been reported to increase when whitebark pine seedling are planted near Grouse whortleberry (Perkins

2004). However, it is not known if the increase in survival of whitebark pine seedlings planted in the presence of grouse whortleberry is due to biotic interaction or if the two species simply co-exist on the same sites due to a need for the same set of environmental conditions. It has been hypothesized that this increase in survival could be due to shared mycorrhizal associates between whitebark pine and grouse whortleberry (Perkins 2004, Scott et al. 2011). Currently, there is no data to support this hypothesis and *Vaccinium* typically hosts ericoid fungi while pines host ectomycorrhizal fungi (Mohatt 2006, Trusty 2009, Keane et al. 2012).

Survival by Microsite

The presence of a shelter object (log, stump, rock, snag within 30 cm) positively and significantly influenced whitebark pine seedling survival on all site conditions at the Summit Lake study area, with the greatest effect being on un-burned sites without beargrass (34.5%). The use of a shelter object (microsite) has previously been considered important to whitebark pine seedling survival; however, this is the first study to provide clear statistical evidence supporting this hypothesis. In a small study using a subset of 100 monitored seedlings, Izlar (2007) reported that the presence of a shelter object/microsite significantly increased whitebark pine seedling survival in the first year after out-planting; however, samples were heavily biased towards micrositied seedlings. Average seedling height also increased at the Summit Lakes study area when a shelter object was present and this increased significantly in burned areas. Izlar (2007) also observed a correlation between the presence of a shelter object/microsite and average seedling height during the early establishment phase of planted whitebark pine seedlings.

Additionally, naturally regenerating whitebark pine seedlings are often associated with a shelter object, and a majority of naturally establishing seedlings have been reported as within 15cm of a shelter object/microsite (Tomback et al. 1993, McCaughey and Tomback 2001). Izlar (2007) suggests that large logs and rocks located uphill or to the side of planted whitebark pine seedlings create the most favorable shelter objects, while live trees and shrubs are the least beneficial (Izlar 2007). Planting in favorable microhabitats has been hypothesized to increase seedling survival and growth because it protects developing seedlings from wind, soil erosion, snow movement, and increased solar radiation often associated with high elevation sites (McCaughey and Tomback 2001, Izlar 2007, Scott et al. 2011).

Ectomycorrhizal Inoculation and Individual Seedling Survival

The effects of ectomycorrhizal inoculation on seedling performance after out-planting have been shown to be highly variable and site specific (Castellano 1996, Campbell et al. 2003, Schwartz et al. 2006, Quoreshi et al. 2008, Khasa et al. 2009). In this study, the effect of inoculation was dependent on site conditions. Inoculation with native ectomycorrhizal fungi did improve seedling survival 17-23% on un-burned sites that had beargrass two years after out-planting. It is possible that the dense rhizomatous root system of beargrass restricted control seedlings from associating with local ectomycorrhizal fungi in the soil on a microsite level. In a small study in Waterton-Lakes National Park, only a few ectomycorrhizae were found on whitebark pine seedlings regenerating naturally in beargrass mats; and the diversity of ectomycorrhizal fungi on

the roots of these seedlings was low (Cripps et al. 2008). Competition from weeds will reduce the number of ectomycorrhizal root tips on conifer seedlings (Sylvie and Jarstfer 1997). In this study, inoculation may have given an advantage to seedlings isolated from local ectomycorrhizal fungi by the rhizomatous beargrass mat. Beargrass negatively affects the survival of planted whitebark pine seedlings (Perkins 2004), and here inoculation appears to overcome this limitation. However, the long-term dynamics between seedlings, ectomycorrhizal fungi, and beargrass are not known at this point.

In un-burned areas lacking beargrass there was no clear mycorrhizal effect, and microsite appears to be the dominant factor positively influencing seedling survival. As mentioned previously, these are poor sites with shallow rocky soils that are more exposed to extreme environmental conditions. While inoculation might be expected to ameliorate these harsh conditions, it is possible that the harsh abiotic conditions (soil, moisture, pH) were simply not conducive to establishment of either seedlings or fungal associates. These sites had the lowest survival overall and the absence of other plant life could indicate poor planting areas for whitebark pine seedlings, inoculated or not. Results suggest that these kinds of sites should be either avoided in future plantings or the use of microhabitats should be emphasized.

No clear mycorrhizal effect was observed on burned sites. This was surprising since it is generally thought that the effect of inoculation might be most apparent on burns, however, the influence of fire on ectomycorrhizal communities is known to be site specific and results depend on many factors including: frequency, intensity, and season of burning, as well as climate, soil moisture, fuel load, and forest type (Cairney and Bastias

2007). At the Summit Lake study site, survival in burned areas was still high for all treatments two years after out-planting and the effects of ectomycorrhizal inoculation may not yet be apparent, if they occur at all. In an assessment of the natural mycorrhizal colonization of planted whitebark pine seedlings on Dunraven Pass in Yellowstone National Park there was little correlation between survival and mycorrhizal colonization until the 3rd year after out-planting (Izlar 2007, Cripps and Lonergan 2011). In the third year, survival was higher (43-100%) on sites where suilloid ectomycorrhizal fungi had been observed on roots in the first year after out-planting in comparison to sites where ectomycorrhizal fungi were absent (25%); (Cripps and Lonergan 2011). In addition, initial nutrient release typically associated with burned soils may have contributed to the high survival rates masking any inoculation effect.

Another possibility as to why there was no effect from inoculation in burned areas is that native ectomycorrhizal fungi could already be present and available in the soil. This would obscure any potential advantage of nursery inoculation. Living, mature whitebark pine trees are present on the Summit Lake study site and could provide a source of inoculum to un-inoculated seedlings. Pre-existing mycelial networks are the primary mode of colonization for many ectomycorrhizal fungi and can remain intact in forest systems even after fire (Amaranthus and Perry 1989, Deacon and Flemming 1992, Horton et al. 1998). Sites at Summit Lake burned by the terrestrial torch were small and patchy and the burns were of a mixed severity ranging from 'no lethal fire' to 'greater than 50% consumption of live tree canopies'.

Certain ectomycorrhizal fungi can survive fire as infected root tips or resistant spores (Cairney and Bastias 2007). In fact, resistant propagules (spores) of *Rhizopogon* species (a suilloid) were suggested to be the primary inoculum source for naturally regenerating bishop pine (*Pinus muricata* D. Don) seedlings following a stand-replacing wildfire on the California coast (Baar et al. 2002). It is also important to recognize that burned sites are conducive to natural mycorrhization by burn-adapted fungi, such as *Amphinema byssoides* (pers.) J. Erikss. or *Pseudotomentella nigra* (P. karst). Both inoculated and un-inoculated seedlings could pick up these fungi leading to higher ectomycorrhizal diversity which could be an advantage. Four years after the stand-replacing Fridley fire in the Gallatin National Forest, the root systems of planted and naturally regenerating whitebark pine seedlings were 90-99% colonized by local, burn adapted fungi (Trusty 2009, Trusty and Cripps 2011). In addition, the animals that can serve as vectors for moving inoculum from un-burned sites onto burns are present at Summit Lake.

However, there does appear to be some indication of an early mycorrhizal effect in that inoculated seedlings were on average slightly taller than un-inoculated seedlings for most site conditions and were significantly taller (0.90 cm) on burned areas without beargrass. This provides some possible evidence that inoculation treatment is starting to affect the performance of whitebark pine seedlings on most site conditions. This result was unexpected since whitebark pine is a slow growing conifer and growth parameters are not typically as informative as they are for fast growing tree species. For example, Rincón et al. (2007) found a significant increase in the height of *Pinus halepensis*

seedlings when they were inoculated with *Suillus collinitus* and similarly, Quoreshi et al. (2008) found that inoculation with ectomycorrhizal fungi significantly increased the stem volume of out-planted black spruce, white spruce, and lodgepole pine seedlings. However, the influence of ectomycorrhizal inoculation on conifer growth is variable and results are dependent on many abiotic and biotic factors (Senstrom and Ek 1990. Castellano 1996, Steinfield et al. 2003, Teste et al. 2004, Rincón et al. 2007, Quoreshi et al. 2008). At this point, it is not known whether increased seedling height correlates with survival. Also, on three of the sites, inoculated seedlings were significantly taller than ‘exposed’ seedlings. It is possible that initiation of colonization on ‘exposed’ seedlings has caused a carbon drain and reduced heights.

Ectomycorrhizal Inoculation and Cluster Survival

Whitebark pine seedlings germinate naturally in small, dense clusters from seeds cached by Clark’s nutcracker (Tomback 1982, Tomback et al. 2001a). The planting of seedlings in clusters was advantageous to limber pine seedlings in Waterton-Lakes National Park. Three years after out-planting, limber pine seedlings planted in clusters of three and five seedlings had higher survival rates than individually planted seedlings (Smith et al. 2011a). This result, as well as patterns of natural regeneration, provided the basis for planting whitebark pine seedlings in clusters. Clusters of several seedlings are better able to withstand extreme environmental conditions because canopies entwine, effectively sheltering seedlings and producing a favorable microclimate (Heumader 2000, Smith et al. 2011a). Furthermore, the larger root mass associated with multiple seedlings may anchor clusters in place reducing seedling mortality due to erosion and avalanches

(Mohatt 2006). In a small study on Scotch Bonnet Mountain, Montana, whitebark pine seedling clusters also hosted a greater diversity of ectomycorrhizal fungi than individual seedlings (Mohatt 2006).

Seedlings at the Summit Lake study site were planted in clusters to mimic the patterns of natural regeneration. Each cluster contained 0, 1, 2 or 3 inoculated seedlings in order to evaluate the minimum number of inoculated seedlings needed to increase overall cluster survival. There were no clear trends between seedling survival and cluster inoculation treatment (T1-T3) across all treatments and again survival was influenced strongly by site condition.

In un-burned areas with beargrass, seedling survival was increased whether clusters contained one (18.3%), two (24.7%) or three (15.0%) inoculated seedlings. In fact, seedling survival for inoculated clusters was comparable to overall survival rates observed in burned areas with beargrass. This suggests that on un-burned sites with beargrass, seedlings could benefit from nursery inoculation and that a minimum of only one inoculated seedling is necessary to increase cluster survival, although survival was higher yet when two seedlings in a cluster were inoculated and further monitoring is necessary to confirm this result.

In this applied study, there were also many factors that could not be controlled. We know that soil properties, climate, and resource competition can potentially influence the effects of ectomycorrhizal inoculation on cluster survival. However, logistical considerations might also be of importance. Seedlings in this study were inoculated only one month prior to planting and it was not feasible to assess them for mycorrhizal

colonization before out-planting as seedlings were taken immediately to the planting site at Waterton-Lakes National Park. It is recommended that seedlings be inoculated 3-5 months prior to out-planting to allow sufficient time for ectomycorrhizal colonization to occur (Loneragan and Cripps 2012, in ed.). Half of the seedlings were inoculated and had access to suilloid fungi, and additional seedlings were ‘exposed’ to ectomycorrhizal fungi by being planted in clusters with inoculation seedlings. However, it is not known how far the ectomycorrhizal colonization process had progressed on inoculated seedlings prior to out-planting, and how far it progressed on ‘exposed’ seedlings afterwards, since it was not possible to dig up seedlings and directly assess the root systems. In addition, it is known from previous assessments of the root systems of planted whitebark pine seedlings that the container shape can persist for years (Trusty 2009, Schwandt et al. 2012 unpublished data). Therefore, at Summit Lake the assessment is likely for this “containerized” condition. Future studies hope to address the use of ectomycorrhizal inoculation to help re-establish planted whitebark pine seedlings in areas known to be devoid of native ectomycorrhizal fungi, such as ghost (dead) forests, large severe burns, and areas not previously in whitebark pine.

Extrapolation to a larger scale could be illustrative for future restoration strategies that involve ‘cluster’ planting. While not directly comparable to the ‘seedling clusters’ that result from nutcracker plantings, the cluster-planting of seedlings in small islands has been examined as an afforestation strategy in Austria (Heumader 2000, Schönenberger 2001). Cluster afforestation has shown many advantages compared to uniform planting systems and has been successfully used to restore pine forests on extreme sites, such as

avalanche paths and torrent catchments, where traditional uniform planting failed (Schönenberger 2001). A long-term, large-scale restoration study intended to mimic the clustering patterns of natural seedling regeneration was implemented over 50 years ago in the Austrian Alps with the intention of creating independent mature “tree islands” (Heumader 2000, Schönenberger 2001). In this study, 20-30 seedlings were “clustered” by being planted in plots 2-4 meters in diameter (Schönenberger 2001). This approach avoided excess waste of seedlings by preferentially planting seedling clusters in the best available microsites while avoiding unfavorable locations such as depressions, hot exposed sites, and areas with densely established herbaceous vegetation (Schönenberger 2001). Uniform plantings were found to be susceptible to wind-throw when they reached a certain age; however, tree islands with open areas in-between allowed for movement of wind and snow (Schönenberger 2001). Tree islands also allowed for better penetration of light, ameliorated temperature fluctuations, and improved moisture conditions (Schönenberger 2001). This method could offer some valuable insights for high-elevation whitebark pine reforestation.

Summary of Conclusions

Assessment of the early survival of whitebark pine seedlings out-planted at the Summit Lake site in Waterton Lakes National Park with implications for future plantings found:

- Overall survival was high (95%) after one year and dropped to 69% in year two. Survival rates were higher than for most other studies and a number of factors may have contributed to high overall survival, including favorable spring

conditions, conducive site conditions (burning) and mycorrhizal inoculation (for some sites).

- The seedling survival rate was highest in burned areas (70%) and lower on unburned areas (51 %) after two years. The average health of seedlings planted on burned sites was also higher than that of seedlings planted in unburned areas. Results suggest that the combination of terra-torching small areas (in this case where living whitebark pines were present) and planting seedlings is a useful restoration treatment in some areas.
- The presence of microsites increased seedling survival around 10% on burned sites and unburned sites with beargrass, and 35% on unburned sites without beargrass
- The unburned areas without beargrass had the lowest survival rate (38%). One third of all clusters died on these planting sites with shallow, rocky soils. These poor planting sites should either be avoided in future plantings or consideration should be taken to plant seedlings in appropriate microsites.
- The effect of ectomycorrhizal inoculation on seedling survival was dependent on site conditions. On unburned sites with beargrass present, inoculated (65.6%) and exposed seedlings (71.5%) had a higher survival rate than un-inoculated controls (48.3%). Inoculated seedlings were also slightly taller on these sites. Inoculation raised survival rates up to that of burned areas without beargrass. Rhizomatous beargrass mats could have restricted control seedlings from associating from

native fungi in the soil and local conditions might have been conducive to the fungi used in inoculation.

- Inoculation did not appear to have an effect on seedling survival in burned areas where survival is still high after two years. However, inoculated seedlings were slightly taller on burned areas, possibly an early indication of a mycorrhizal effect. Longer term monitoring is needed here, since mycorrhizal effects may not be apparent early on, on some sites.
- Determining the minimum number of inoculated seedlings needed in each cluster to improve survival proved more complicated. Analysis of the odds-of-survival for seedlings planted in clusters showed no real trend from T1 through T3 across treatments. However, on unburned sites with beargrass, all three inoculation treatments (T1, T2, and T3) had a positive effect on survival, suggesting that a minimum of one inoculated seedling is sufficient for a response. The odds-of-survival for seedlings in these clusters was improved 1.85, 2.54 and 1.85 times, with p values of 0.09, 0.02 and 0.13, respectively. There was basically no mycorrhizal effect for clusters on other sites (with one positive and one negative exception).
- Complicating factors are that it was not possible to assess the actual mycorrhizal status of inoculated and un-inoculated seedlings and also whitebark pine seedlings can retain their container shape for years and are likely being assessed in this condition.

- Longer term monitoring is necessary to determine the effect of site conditions and mycorrhizal inoculation on subsequent seedling survival.
- Future studies also hope to address the use of ectomycorrhizal inoculation in the greenhouse to help whitebark pine seedlings establish in areas truly devoid of native ectomycorrhizal fungi such as ghost (dead) forests, large severe burns, and areas not previously in whitebark pine.

References

- Achuff, P.L., McNeil, R.L., Coleman, M.L., Wallis, C., and Wershler, C. 2002. Ecological land classification of Waterton Lakes National Park, Alberta, Vol. I: integrated resource description. Unpublished Technical Report, Parks Canada, Waterton Park, AB.
- Amaranthus, M. 2002. Around the world inoculation and conifer establishment using *Rhizopogon* mycorrhizal fungi. In: Dumroese, R.K., Riley, L.E., and Landis, T.D. (coords). National Proceedings: forests and conservation nursery associations-1999, 2000, and 2001. Proceedings RMRS-P-24. Ogden, UT: U.S. Department of Agriculture Forest Service, Rocky Mountain Research Station. 226 p.
- Amaranthus, M.P. and Perry, D.A. 1989. Interaction effects of vegetation type and Pacific madrone soil inocula on survival, growth, and mycorrhiza formation of Douglas-fir. *Canadian Journal of Forest Research* 19: 550-556.
- Amaranthus, M.P., Trappe, J.M., and Perry, D.A. 1993. Soil moisture, native revegetation, and *Pinus lambertiana* seedling survival, growth, and mycorrhiza formation following wildfire and grass seedling. *Restoration Ecology* 1: 188-195.
- Arno, S.F. 1986. Whitebark pine cone crops--a diminishing source of wildlife food? *Western Journal of Applied Forestry* 1: 92-94.
- Arno, S.F. and Hoff, R.J. 1990. Silvics of whitebark pine (*Pinus albicaulis*). General Technical Report INT-253. U.S. Department of Agriculture, Forest Service, Intermountain Research Station, Ogden, UT.
- Arno, S.F.; Simmerman, D.G.; and Keane, R.E. 1985. Forest succession on four habitat types in western Montana. General Technical Report INT-177. U.S. Department of Agriculture, Forest Service, Intermountain Forest and Range Experiment Station, Ogden, UT. 74 p.
- Asebrook, J., Lapp, J., and Carolin, T. 2011. Whitebark and Limber pine restoration and monitoring in Glacier National park. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Aubry, C., Goheen, D., Shoal, R., Ohlson, T., Lorenz, T., Bower, A., Mehmel, C., and Sniezko, R. 2008. Whitebark pine restoration strategy for the Pacific Northwest region 2009-2013. U.S. Department of Agriculture, Forest Service. 13p.

- Baar, J., Horton, T.R., Kretzer, A.M., and Bruns, T.D. 2002. Mycorrhizal colonization of *Pinus muricata* from resistant propagules after a stand-replacing wildfire. *New Phytologist* 43(2): 409-418.
- Bower, A.D. and Aitken, S.N. 2008. Ecological genetics and seed transfer guidelines for *Pinus albicaulis* (Pinaceae). *American Journal of Botany* 95(1): 66-76.
- Bradley, A.F., Noste, N.V., and Fischer, W.C. 1992. Fire ecology of forests and woodlands in Utah. General Technical Report INT-287. U.S. Department of Agriculture, Forest Service, Intermountain Research Station, Ogden, UT. 128 p.
- Brundrett, M., Malajczuk, N., Mingquin, G., Daping, X., Snelling, S., and Dell, B. 2005. Nursery inoculation of Eucalyptus seedlings in Western Australia and Southern China using spores and mycelial inoculum of diverse ectomycorrhizal fungi from different climatic regions. *Forest Ecology and Management* 209: 193-205.
- Bruns, T.D., Peay, K.G., Boynton, P.J., Grubisha, L.C., Hynson, N.A., Nguyen, N.H., and Rosenstock, N.P. 2009. Inoculum potential of *Rhizopogon* spores increases with time over the first 4 yr of a 99-yr spore burial experiment. *New Phytologist* 181(2): 463-470.
- Burns, K.S., Schoettle, A.W., Jacobi, W.R., and Mahalovich, M.F. 2008. Options for the management of white pine blister rust in the Rocky Mountain region. General Technical Report RMRS-GTR-206, U.S. Department of Agriculture, Forest Service.
- Burr, K., Eramian, A., and Eggleston, K. 2001. Growing whitebark pine seedlings for restoration. Pp. 325-345. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds.). *Whitebark pine communities: ecology and restoration*. Island Press, Washington, DC. 440 p.
- Cairney, J.W. and Bastias, B.A. 2007. Influences of fire on soil fungal communities. *Canadian Journal of Forest Research* 307: 207-215.
- Campbell, D.B., Jones, M.D., Kiiskila, S. and Bulmer, C. 2003. Two-year field performance of lodgepole pine seedlings: effects of container type, mycorrhizal fungal inoculants, and site preparation. *B.C. Journal of Ecosystem Management* 3(2): 1-11.
- Carolin, T. 2006. Whitebark and limber pine restoration in Glacier National Park: monitoring results. *Nutcracker Notes* 10: 14.

- Castellano, M.A. 1996. Outplanting performance of mycorrhizal inoculated seedlings. Pp. 223-301. In: Mukerji, K.G. (ed.). Concepts in Mycorrhizal Research. Kluwer Academic Publishers, Netherlands. 371 p.
- Castro, J., Zamora, R., Hódar, J.A., and Gómez, J.M. 2002. Use of shrubs as nurse plants: a new technique for reforestation in Mediterranean Mountains. *Restoration Ecology* 10(2): 297-305.
- Certini, G. 2005. Effects of fire on properties of forest soils: a review. *Oecologia* 143: 1-10.
- Chadwick, A.C. 2002. *Carex geyeri*. In: Fire Effects Information System [online]. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station, Fire Sciences Laboratory (Producer). Available: <http://www.fs.fed.us/database/feis/>.
- Coen, G.D. and Holland, W.D. 1976. Soils of Waterton Lakes National Park, Alberta. Alberta Institute of Pedology S-73-33, Information Report NOR-X-65. 114 p.
- Coop, J.D. and Schoettle, A.W. 2009. Regeneration of Rocky Mountain bristle cone pine (*Pinus aristata*) and limber pine (*Pinus flexilis*) three decades after stand replacing fires. *Forest Ecology and Management* 257: 893-903.
- COSEWIC. 2010. Assessment and status report on Whitebark Pine (*Pinus albicaulis*) in Canada [online]. Committee on the Status of Endangered Wildlife in Canada. Ottawa. 44 p. Available: www.sararegistry.gc.ca/status/status_e.cfm.
- Crane, M. F. 1990. *Xerophyllum tenax*. In: Fire Effects Information System [online]. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station, Fire Sciences Laboratory (Producer). Available: <http://www.fs.fed.us/database/feis/>.
- Cripps, C.L. 2012. [Personal Communication]. July 15, 2011. Associate Professor, Montana State University, Bozeman, Montana.
- Cripps, C.L. and Antibus, R.K. 2011. Native ectomycorrhizal fungi of limber and whitebark pine: necessary for forest sustainability? In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.

- Cripps, C.L. and Grimme, E. 2011. Inoculation and successful colonization of whitebark pine seedlings with native mycorrhizal fungi under greenhouse conditions. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Cripps, C.L. and Lonergan, E. 2011. Whitebark pine and ectomycorrhizal fungi: summary and update on various projects. Whitebark Pine Ecosystem Foundation Meeting, Cody, WY.
- Cripps, C.L., Smith, C., Carolin, T., and Lapp, J. 2008. Ectomycorrhizal fungi with whitebark pine. Nutcracker Notes. 14: 12-13.
- Cripps, C.L. and Trusty, P. 2007. Final Report. Whitebark pine restoration, Dunraven Pass: monitoring mycorrhizal status of seedlings. Greater Yellowstone Coordinating Committee.
- Deacon, J.W. and Flemming, L.V. 1992. Interactions of ectomycorrhizal fungi. Pp. 249-300. In: Allen, M.F. (ed.). Mycorrhizal functioning: an integrative plant-fungal process. Chapman and Hall, New York.
- Eggleston, K. 2010. [Personal Communication with Dr. Cathy Cripps] April 19, 2010. USDA Forest Service Nursery, Coeur d'Alene, Idaho.
- El Karkouri, K., Martin, F., Douzery, J.P., and Mousain, D. 2005. Diversity of ectomycorrhizal fungi naturally established on containerized *Pinus* seedlings in nursery conditions. Microbiological Research 160: 47-52.
- Ellison, A.M., Bank, M.S., Clinton, B.D., Colburn, E.A., Elliott, K., Ford, C.R., Foster, D.R., Kloeppel, B.D., Knoepp, J.D., Lovett, G.M., Mohan, J., Orwig, D.A., Rodenhouse, N.L., Sobczak, W.V., Stinson, K.A., Stone, J.K., Swan, C.M., Thompson, J., Von Holle, B., and Webster, J.R. 2005. Loss of foundation species: consequences for the structure and dynamics of forested ecosystems. *Frontiers in Ecology and the Environment* 3: 479-486.
- Entry, J.A., Rygielwicz, P.T., Watrud, L.S., Donnelly, P.K. 2002. Influence of adverse soil conditions on the formation and function of arbuscular mycorrhizas. *Advances in Environmental Research* 7910: 123-138.
- ESRI 2011. ArcGIS Desktop: Release 10. Redlands, CA: Environmental Systems Research Institute.

- Farnes, P.E. 1990. SNOTEL and snow course data: describing the hydrology of whitebark pine ecosystems. Pp. 302-304. In: Schmidt, W.C. and McDonald, K.J. (comps.). Proceedings: Symposium on whitebark pine ecosystems: ecology and management of a high-mountain resource. USDA Forest Service, General Technical Report INT-270, Intermountain Research Station, Ogden, Utah.
- Felicetti, L.A., Schwartz, C.C., Rye, R.O., Haroldson, M.A., Gunther, K.A., Phillips, D.L., and Robbins, C.T. 2003. Use of sulfur and nitrogen stable isotopes to determine the importance of whitebark pine nuts to Yellowstone grizzly bears. *Canadian Journal of Zoology* 81(5): 763-770.
- Fettig, C.J., Dabney, C.P., McKelvey, S.R., and Huber, P.W. 2008. Non-host angiosperm volatiles and verbenone protect individual ponderosa pines from attack by western pine beetle and red turpentine beetle (Coleoptera: Curculionidae, Scolytinae). *Western Journal of Applied Forestry* 23(1): 40-45.
- Fischer, W.C. and Bradley, A.F. 1987. Fire ecology of western Montana forest habitat types. General Technical Report INT-223. U.S. Department of Agriculture, Forest Service, Intermountain Research Station, Ogden, UT. 101 p.
- Fortin, J.K. 2011. Niche separation of grizzly (*Ursus arctos*) and American black bears (*Ursus americanus*) in Yellowstone National Park. Washington State University PhD dissertation, Pullman, WA.
- Fryer, J.L. 2002. *Pinus albicaulis*. In: Fire Effects Information System. [online]. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station, Fire Sciences Laboratory (Producer). Available: <http://www.fs.fed.us/database/feis/>.
- Gagne, A., Jany, J.L., Bousquet, J., Khasa, J., and Gamase, P. 2006. Ectomycorrhizal fungal communities of nursery-inoculated seedlings out-planted on clear-cut sites in northern Alberta. *Canadian Journal of Forest Research* 36: 1684-1694.
- Gernandt, D.S., López, G.G., García, S.O., and Liston, A. 2005. Phylogeny and classification of *Pinus*. *Taxon* 54(1): 29-42.
- Government of Alberta. 2010. Species assessed by Alberta's Endangered Species Conservation Committee: short list [online]. Available: <http://www.srd.alberta.ca/BiodiversityStewardship/SpeciesAtRisk/SpeciesSummaries/documents/SpeciesAssessed-EndangeredSpeciesConservationCommittee>.
- Government of Alberta. 2012. Environment and Sustainable Resource Development [online]. Available:

<http://www.environment.alberta.ca/apps/basins/DisplayData.aspx?Type=Figure&BasinID=14&DataType=4&StationID=AKAM>.

- Government of Canada. 2012. Order amending Schedule 1 to the Species at Risk Act. Canada Gazette Part II 146 (14) SOR/2012-113: 1418-1629. [online]. Available: http://www.sararegistry.gc.ca/virtual_sara/files/orders/g2-14614i_e.pdf.
- Hangs, R.D., Knight, J.D., and Van Rees, K.C.J. 2002. Interspecific competition for nitrogen between early successional species and planted white spruce and jack pine seedlings. *Canadian Journal of Forest Research* (32): 1813-1821.
- Heumader, J. 1992. Cultivation of cembran pine plants for high-elevation afforestations. International workshop on subalpine stone pines and their environment: the status of our knowledge, September 5-11, St. Moritz, Switzerland.
- Heumader, J. 2000. High elevation afforestation and regeneration of subalpine forest stands experiences in Austria. *Internationales Symposium Interpraevent 2000 – Villach/Österreich Tagungspublikation 2*: 29-40.
- Hoff, R.J., Ferguson, D.E., McDonald, G.I., and Keane, R.E. 2001. Strategies for managing whitebark pine in the presence of white pine blister rust. Pp. 346-366. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds). *Whitebark pine communities: ecology and restoration*. Island Press, Washington, DC.
- Hollingsworth, L.T. 2010. *Davies Fire: fire behavior and fire effects analysis*. Fire Modeling Institute, Missoula Fire Sciences Laboratory, Rocky Mountain Research Station.
- Horton, T.R., Cázares, E., and Bruns, T.D. 1998. Ectomycorrhizal, vesicular-arbuscular and dark septate fungal colonization of bishop pine (*Pinus muricata*) seedlings in the first 5 months of growth after wildfire. *Mycorrhiza* 8: 11-18.
- Hunt, G. 1992. Effects of mycorrhizal fungi on quality of nursery stock and plantation performance in the southern interior of British Columbia. FRDA Report 185. Forestry Canada and the British Columbia Ministry of Forests.
- IBM Corporation. Released 2011. *IBM SPSS Statistics for Windows, version 20.0*. Armonk, NY: IBM Corp.
- Izlar, K. 2007. Assessment of whitebark pine seedling survival for Rocky Mountain plantings. University of Montana Master of Science thesis, Missoula, MT.

- Keane, R.E. and Arno, S.F. 1993. Rapid decline of whitebark pine in western Montana: evidence from 20-year re-measurements. *Western Journal of Applied Forestry* 8: 44-47.
- Keane, R.E. and Parsons, R.A. 2010. Management guide to ecosystem restoration treatments: whitebark pine forests of the northern Rocky Mountains, U.S.A. General Technical Report RMRS-GTR-232. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 133 p.
- Keane, R.E., Tomback, D., Aubry, C., Bower, A., Campbell, E., Cripps, C., Jenkins, M., Manning, M., McKinney, S., Murray, M., Perkins, D., Reinhart, D., Ryan, C., Schoettle, A., Smith, C. 2012. A range-wide restoration strategy for whitebark pine (*Pinus albicaulis*). General Technical Report RMRS-GTR-279 Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station.
- Kendall, K.C. and Keane, R.E. 2001. Whitebark pine decline: infection, mortality, and populations trends. Pp. 221-242. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds.). whitebark pine communities: ecology and restoration. Island Press, Washington DC.
- Khasa, P.D., Piche, Y., and Coughlan, A.P. (eds.). 2009. Advances in mycorrhizal science and technology. NRC Research Press, Ottawa. 197 p.
- Landhausser, S., Stadt, K., and Lieffers, V. 1996. Screening for control of a forest weed: early competition between three replacement species and *Calamagrostis canadensis* or *Picea glauca*. *Journal of Applied Ecology* 33: 1517-1526.
- Lanner, R.M. 1996. Made for each other: a symbiosis of birds and pines. Oxford University Press. New York, NY. 166 p.
- Lantz, G. 2010. Whitebark pine: an ecosystem in peril. American Forests Special Report. Norman, OK.
- Larson, E.R. and Kipfmüller, K.F. 2010. Patterns in whitebark pine regeneration and their relationships to biophysical site characteristics in southwest Montana, central Idaho, and Oregon, USA. *Canadian Journal of Forest Research* 40: 476-487.
- Lehto, T. and Zwiazek, J.J. 2011. Ectomycorrhizas and water relations of trees: a review. *Mycorrhiza* 21(2): 71-90.
- Leslie, A. and Wilson, B. 2011. No free lunch: observations on seed predation, cone collection and controlled germination of whitebark pine from the Canadian Rockies. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.).

The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.

Lieffers, V.J., Macdonald, S.E. and Hogg, E.H. (1993) Ecology of and control strategies for *Calamagrostis canadensis* in boreal forest sites. Canadian Journal of Forest Research 23: 2070-2077.

Little, E.L. Jr., 1971. Atlas of United States trees, volume 1, conifers and important hardwoods: U.S. Department of Agriculture Miscellaneous Publication 1146. 9 p. and 200 maps.

Logan, J., Mcfarlane, W., and Wilcox, L. 2010. Whitebark pine vulnerability to climate driven mountain pine beetle disturbance in the Greater Yellowstone Ecosystem. Ecological Applications 20(40): 895-902.

Lonergan, E. and Cripps, C.L. (in ed.). Low nitrogen fertilizer as a strategy to maintain ectomycorrhizal colonization of inoculated whitebark pine seedlings before out-planting. Native Plants Journal, in process.

Mahalovich, M.F., Burr, K.E., and Foushee, D.L. 2006. Whitebark pine germination, rust resistance, and cold hardiness among seed sources in the inland northwest: planting strategies for restoration. U.S. Department of Agriculture, Forest Service Proceedings RMRS-P-43.

Mahalovich, M.F. and Dickerson, G.A. 2004. Whitebark pine genetic restoration program for the Intermountain West (USA). Pp. 181-187. In: Snieszko, R., Samman, S., Schlarbaum, S., and Kriebel, H. (eds.). Breeding and genetic resources of five-needle pines: growth, adaptability and pest resistance, IUFRO Working Party 2.02.15, Proceedings. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station, RMRS-P-32, Ogden, UT.

Mahalovich M.F. and Hoff, R.J. 2001. Whitebark pine operational cone collection instructions and seed transfer guidelines. Nutcracker Notes 11: 10-13.

Mattson, D.J., Blanchard, B.M., and Knight, R.R. 1992. Yellowstone grizzly bear mortality, human habituation, and whitebark pine seed crops. Journal of Wildlife Management 56: 432-442.

Mattson, D.J. and Reinhart, D.P. 1994. Bear use of whitebark pine seeds in North America. Pp. 212-220. U.S. Department of Agriculture, Forest Service General Technical Report INTGTR-309.

- Marx, D.H. 1991. The practical significance of ectomycorrhizae in forest establishment. Pp. 54–90. In: Hägglund, B. (ed.). Ecophysiology of Mycorrhizae of Forest Trees. Proceedings. The Marcus Wallenberg Foundation, Stockholm, Sweden.
- McCaughey, W.W. 1990. Biotic and microsite factors affecting *Pinus albicaulis* establishment and survival. Ph.D. dissertation Montana State University, Bozeman, MT.
- McCaughey, W.W. and Schmidt, W.C. 2001. The biology of whitebark pine: taxonomy, distribution and history. Pp. 29-40. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds.). Whitebark pine communities, ecology and restoration. Island Press, Washington, DC. 440 p.
- McCaughey, W., Scott, G.L., and Izlar, K.L. 2009. Whitebark pine planting guidelines. *Western Journal of Applied Forestry* 24(3): 163-166.
- McCaughey, W.W. and Tomback, D.F. 2001. The natural regeneration process. Pp. 105-120. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds.). Whitebark pine communities: ecology and restoration. Island Press, Washington, DC. 440 p.
- McCune, B. 1988. Ecological diversity in North American pines. *American Journal of Botany* 75(3): 353-368.
- McKinney, S.T. and Tomback, D.F. 2007. The influence of white pine blister rust on seed dispersal in whitebark pine. *Canadian Journal of Forest Research* 37: 1044-1057.
- Mellmann-Brown, S. 2005. The regeneration of whitebark pine in the timberline ecotone of the Beartooth Plateau, U.S.A.: spatial distribution and responsible agents. Pp. 96-116. In: Broll, G. and Keplin, B. (eds.). Mountain ecosystems: studies in treeline ecology. Springer-Verlag, Berlin.
- Menkis, A., Vasiliauskas, R., Taylor, A.F.S., Stenlid, J., and Finlay, R. 2005. Fungal communities in mycorrhizal roots of conifer seedlings in forest nurseries under different cultivation systems, assessed by morphotyping, direct sequencing, and mycelial isolation. *Mycorrhiza* 16: 33-41.
- Menkis, A., Vasiliauskas, R., Taylor, A.F.S., Stenlid, J., and Finlay, R. 2007. Afforestation of abandoned farmland with conifer seedlings inoculated with three ectomycorrhizal fungi- impact on plant performance and ectomycorrhizal community. *Mycorrhiza* 17(4): 337-348.

- Mohatt, K. 2006. Ectomycorrhizal fungi of whitebark pine (*Pinus albicaulis*) in the Northern Yellowstone Greater Ecosystem. Montana State University Master's Thesis, Bozeman, MT.
- Mohatt, K.R., Cripps, C.L., and Lavin, M. 2008. Ectomycorrhizal fungi of whitebark pine (a tree in peril) revealed by sporocarps and molecular analysis of mycorrhizae from treeline forests in the Greater Yellowstone Ecosystem. *Botany* 86: 14-25.
- Moody, R.J. 2006. Post-fire regeneration and survival of whitebark pine (*Pinus albicaulis* Engelm.). University of British Columbia Master's Thesis, Vancouver, British Columbia.
- Morgan, P., Bunting, S.C., Keane, R.E., and Arno, S.F. 1994. Fire ecology of whitebark pine forests of the Northern Rocky Mountains, U.S.A. Pp 136-141. In: Schmidt, W.C. and Holtmeier, F.K. (comps.). Proceedings of the international workshop of subalpine stone pines and their environment: the status of our knowledge; September 5-11, 1992; St. Moritz, Switzerland. General Technical Report INT-GRT-309. U.S. Department of Agriculture, Forest Service, Intermountain Research Station Ogden, UT.
- Moser, M.M. 1956. Die Bedeutung der Mykorrhiza für Aufforestungen in Hochlagen. *Forstwissenschaftliches Centralblatt* 75: 8-18.
- Moser, M.M. 2004. Subalpine conifer forests in the Alps, the Altai and the Rocky Mountains: a comparison of their fungal populations. Pp 151-158. In: Cripps, C.L. (ed.). *Fungi in forest ecosystems: Systematics, diversity, and ecology*. New York Garden Botanical Press, NY. 378 p.
- Murray, M.P. 2007. Cone collecting techniques for whitebark pine. *Western Journal of Applied Forestry* 22(3): 153-155.
- Neary, D.G., Klopatek, C.C., DeBano, L.F., Ffolliott, P.F. 1999. Fire effects on belowground sustainability: a review and synthesis. *Forest Ecology and Management* 122: 51-71.
- Nicholas, A. and Katzenberger, D. 2011. Whitebark pine to be designated a candidate for endangered species protection. U.S. Dept. of Interior, Fish and Wildlife Service, Press Release July 18, Lakewood, Colorado.
- Norment, C.J. 1991. Bird use of forest patches in the subalpine forest-alpine tundra ecotone of the Beartooth Mountains, Wyoming. *Northwest Science* 65(1): 1-10.

- Ortega, U., Dunabeitia, M., Mendez, S., Gonzalez-murua, C., and Majada, J. 2004. Effectiveness of mycorrhizal inoculation in the nursery on growth and water relations of *Pinus radiata* in different water regimes. *Tree Physiology* 24: 65-73.
- Parks Canada. 2010. Parks Canada Invests in Ecological Integrity. Press Release. [online] Available: http://www.pc.gc.ca/apps/cp-nr/release_e.asp?bgid=1338&andor1=bg.
- Parladé, J., Luque, J., Pera, J., Rincón, A.M. 2004. Field performance of *Pinus pinea* and *P. halepensis* seedlings inoculated with *Rhizopogon* spp. and outplanted in formerly arable land. *Annals of Forest Science* 61: 507-514.
- Perkins, J. 2004. *Pinus albicaulis* seedling regeneration after fire. University of Montana PhD dissertation, Missoula, MT.
- Price, R.A., Liston, A., and Strauss, S.H. 1998. Phylogeny and systematics of *Pinus*. Pp. 49-68. In: Richardson, D.M. (ed.). *Ecology and biogeography of Pinus*. Cambridge University Press, Cambridge, UK.
- Quoreshi, A.M., Kernaghan, G., and Hunt, G.A. 2009. Mycorrhizal fungi in Canadian forest nurseries and field performance of inoculated seedlings. Pp. 115-128. In: Khasa, P.D., Piche, Y., and Coughlan, A.P. (eds.). *Advances in Mycorrhizal Science and Technology*. NRC Research Press, Ottawa. 197 p.
- Quoreshi, A. M., Piche, Y., and Khasa, D.P. 2008. Field performance of conifer and hardwood species 5 years after nursery inoculation in the Canadian Prairie Provinces. *New Forests* 35: 235-253.
- R Development Core Team. 2008. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL <http://www.R-project.org>.
- Resler, L.M. 2004. Conifer establishment sites on a periglacial landscape, Glacier National Park, Montana. Texas State University Ph.D. dissertation, San Marcos, TX. 186 p.
- Rincón, A., de Felipe, M.R., and Fernández-Pascual, M. 2007. Inoculation of *Pinus halepensis* Mill. with selected ectomycorrhizal fungi improves seedling establishment 2 years after planting in a degraded gypsum soil. *Mycorrhiza* 18: 23-32.
- Ronikier, M., Miskiewicz, A., and Mleczko, P. 2002. Presence and distribution of *Suillus plorans* in the Polish Tatra Mountains (Western Carpathians). *Acta Societatis Botanicorum Poloniae* 71(3): 235-242.

- Schönenberger, W. 2001. Cluster afforestation for creating diverse mountain forest structures – a review. *Forest Ecology and Management* 145: 121-128.
- Schwandt, J., Chadwick, K., and Kearns, H. 2011. Whitebark pine direct seeding trials in the Pacific Northwest. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). *The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.*
- Schwandt, J., DeMastus, C., Cripps, C.L. 2012. Whitebark pine direct seeding trials, unpublished data. Yellowstone Club, Montana.
- Schwanke, R. and Smith, C. 2010. Imitating lightning strikes for whitebark pine restoration. *Nutcracker Notes* 19: 12-13.
- Schwartz, M.W., Hoeksema, J.D., Gehring, C.A., Johnson, N.C., Klironomos, J.N., Abbott, L.K, and Pringle, A. 2006. The promise and the potential consequences of the global transport of mycorrhizal fungal inoculum. *Ecology Letters* 9(5): 501-515.
- Scott, G.L., McCaughey, W.W., and Izlar, K. 2011. Guidelines for whitebark pine planting prescriptions. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). *The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.*
- Senstrom, E. and Ek, M. 1990. Field growth of *Pinus sylvestris* following nursery inoculation with mycorrhizal fungi. *Canadian Journal of Forest Research* 20: 914-918.
- Shebitz, D.J., Reichard, S.H., and Dunwiddie, P.W. 2009. Ecological and cultural significance of burning beargrass habitat on the Olympic peninsula, Washington. *Cultural Restoration* 27(3): 306-319.
- Smith, C.M. 2009. Restoration of whitebark and limber pine: first steps on a long road in Waterton Lakes National Park. *BC Forest Professional*, May-June 14-15.
- Smith, C.M. 2012. [Personal Communication]. *Park Ecologist*, Waterton-Lakes National Park, Alberta, Canada.
- Smith, C.M., Poll, G., Gillies, C., Praymak, C., Miranda, E., and Hill, J. 2011a. Limber pine seed and seedling experiment in Waterton Lakes National Park, Canada. In:

- Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Smith, C.M., Shepherd, B., Gillies, C., and Stuart-Smith, J. 2011b. Re-measurement of whitebark pine infection and mortality in the Canadian Rockies. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Smith, C.M., Wilson, B., Rasheed, S., Walker, R.C., Carolin, T., and Shepard, B. 2008. Whitebark pine and white pine blister rust in the Rocky Mountains of Canada and Northern Montana. *Canadian Journal of Forest Research* 5: 982-995.
- Smith, S.E. and Read, D.J. 2008. *Mycorrhizal symbiosis*, 3rd edition. Academic Press, Inc., San Diego, CA.
- Steinfeld, D., Amaranthus, M., and Cázares, E. 2003. Survival of ponderosa pine (*Pinus ponderosa* Dougl. Ex Laws.) seedlings out-planted with *Rhizopogon* mycorrhizae inoculated with spores at the nursery. *Journal of Arboriculture* 29(4): 197-208.
- Sylvie, D.M. and Jarstfer, A.G. 1997. Distribution of mycorrhiza on competing pines and weeds in a southern pine plantation. *Soil Science Society of America Journal* 61: 139-144.
- Teste, F.P., Schmidt, M.G., Berch, S.M., Bulmer, C., and Egger, K.N. 2004. Effects of ectomycorrhizal inoculants on survival and growth of interior Douglas-fir seedlings on reforestation sites and partially rehabilitated landings. *Canadian Journal of Forest Research* 34: 2074–2088.
- Tomback, D.F. 1982. Dispersal of whitebark pine seeds by Clark's Nutcracker: a mutualism hypothesis. *Journal of Animal Ecology* 51: 451-467.
- Tomback, D.F. 2001a. Clark's nutcracker: agent of regeneration. Pp. 89-104. In Tomback, D.F., Arno, S.F., and Keane, R.E. (eds.). *Whitebark pine communities: ecology and restoration*. Island Press, Washington, DC.
- Tomback, D.F. and Achuff, P. 2010. Blister rust and western forest biodiversity: ecology, values, and outlook for white pines. *Forest Pathology* 40: 186-225.

- Tomback, D.F., Anderies, A.J., Carsey, K.S., Powell, M.L., and Mellmann-Brown, S. 2001b. Delayed seed germination in whitebark pine and regeneration patterns following the Yellowstone fires. *Ecology* 82(9): 2587-2600.
- Tomback, D.F., Arno, S.F., and Keane, R.E. 2001c. Whitebark pine communities: ecology and restoration. Island Press, Washington, DC. 440 p.
- Tomback, D.F. and Linhart, Y.B. 1990. The evolution of bird-dispersed pines. *Evolutionary Ecology* (4): 185-219.
- Tomback, D.F., Sund, S.K., and Hoffmann, L.A. 1993. Post-fire regeneration of *Pinus albicaulis*: height –age relationships, age structure, and microsite characteristics. *Canadian Journal of Forest Research* 23: 113-119.
- Trusty, Paul. 2009. Impact of severe fire on ectomycorrhizal fungi of whitebark pine seedlings. Montana State University Master's Thesis, Bozeman, MT.
- Trusty, P. and Cripps, C.L. 2011. Influence of fire on mycorrhizal colonization of planted and natural whitebark pine seedlings: Ecology and management implications. Pp. 198-202. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Wagner, R.G., Peterson, T.D., Ross, D.W., and Radosevich, S.R. 1989. Competition thresholds for the survival and growth of ponderosa pine seedlings associated with woody and herbaceous vegetation. *New Forests* 3(2): 151-170.
- Wan, S., Hui, D., and Luo, Y. 2001. Fire effects on nitrogen pools and dynamics in terrestrial ecosystems : a meta-analysis. *Ecological Applications* 11(5): 1349-1365.
- Weaver, T. and Dale, D. 1974. *Pinus albicaulis* in central Montana: environment, vegetation, and production. *American Midland Naturalist* 37: 395-401.
- Weisleitner, H. 2008. [Personal Communication with Dr. Cathy Cripps]. Federal Nursery of Austria, Innsbruck, Austria.
- Wiensczyk, G., Durall, D. Jones, M. and Simard, S. 2002. Ectomycorrhizae and forestry in British Columbia: a summary of current research and conservation strategies. Extension Note. *B.C. Journal of Ecosystems and Management* 2(1): 1-20.

LITERATURE CITED

-
- Achuff, P.L., McNeil, R.L., Coleman, M.L., Wallis, C., and Wershler, C. 2002. Ecological land classification of Waterton Lakes National Park, Alberta, Vol. I: integrated resource description. Unpublished Technical Report, Parks Canada, Waterton Park, AB.
- Aime, M.C. and Miller Jr., O.K. 2002. Delayed germination of basidiospores in temperate species of *Crepidotus* (Fr.) Staude. *Canadian Journal of Botany* 80(3): 280-287.
- Allen, M.F., Swenson, W., Querejeta, J.I., Egerton-Warburton, L.M., and Treseder, K.K. 2003. Ecology of mycorrhizae: a conceptual framework for complex interactions among plants and fungi. *Annual Review of Phytopathology* 41: 271-303.
- Amaranthus, M.P. 2002. Around the world nursery inoculations and conifer establishment using *Rhizopogon* mycorrhizal fungi. In: Dumroese, R.K., Riley, L.E., and Landis, T.D., (coords. and eds.). National proceedings: forest and conservation nursery associations-1999, 2000, and 2001. Proceedings RMRS-P-24. Ogden, UT: U.S. Department of Agriculture Forest Service, Rocky Mountain Research Station. 226 p.
- Amaranthus, M.P., Nair, M.G., Reid, T.C., and Steinfeld, D. 2005. Improved *Rhizopogon* mycorrhizal colonization and foliar nutrient levels in Ponderosa pine and Douglas-fir with myconate (r). *Journal of Sustainable Forestry* 20(3): 1-14.
- Amaranthus, M.P. and Perry, D.A. 1989. Interaction effects of vegetation type and Pacific madrone soil inocula on survival, growth, and mycorrhiza formation of Douglas-fir. *Canadian Journal of Forest Research* 19: 550-556.
- Amaranthus, M.P., Trappe, J.M., and Perry, D.A. 1993. Soil moisture, native revegetation, and *Pinus lambertiana* seedling survival, growth, and mycorrhiza formation following wildfire and grass seedling. *Restoration Ecology* 1: 188-195.
- Antibus, R.K. 2010. [Personal communication]. Professor, Department of Science, Bluffton University, Bluffton, Ohio.
- Aplet, G.H. 2006. Evolution of wilderness fire policy. *International Journal of Wilderness* 12(1): 9-13.
- Arnebrant, K. 1994. Nitrogen amendments reduce the growth of extramatrical ectomycorrhizal mycelium. *Mycorrhiza* 5(1): 7-15.

- Arno, S.F. 1986. Whitebark pine cone crops - a diminishing source of wildlife food? *Western Journal of Applied Forestry* 1: 92-94.
- Arno, S.F. 2001. Community types and natural disturbance processes. Pp. 74-88. In: Tomback, D.F., Arno, S.F., and Keane, R.E., (eds.). *Whitebark pine communities: ecology and restoration*. Island Press, Washington, DC. 439 p.
- Arno, S.F. and Hoff, R.J. 1990. Silvics of whitebark pine (*Pinus albicaulis*). General Technical Report INT-253. U.S. Department of Agriculture, Forest Service, Intermountain Research Station, Ogden, UT.
- Arno, S.F.; Simmerman, D.G.; and Keane, R.E. 1985. Forest succession on four habitat types in western Montana. General Technical Report INT-177. U.S. Department of Agriculture, Forest Service, Intermountain Forest and Range Experiment Station, Ogden, UT. 74 p.
- Asebrook, J., Lapp, J., and Carolin, T. 2011. Whitebark and limber pine restoration and monitoring in Glacier National park. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). *The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium*. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Aubry, C., Goheen, D., Shoal, R., Ohlson, T., Lorenz, T., Bower, A., Mehmel, C., and Sniezko, R. 2008. Whitebark pine restoration strategy for the Pacific Northwest region 2009-2013. U.S. Department of Agriculture, Forest Service. 13p.
- Baar, J., Horton, T.R., Kretzer, A.M., and Bruns, T.D. 2002. Mycorrhizal colonization of *Pinus muricata* from resistant propagules after a stand-replacing wildfire. *New Phytologist* 43(2): 409-418.
- Bentz, B.J., Kegley, S., Gibson, K. and Thier, R. 2005. A test of high-dose verbenone for stand-level protection of lodgepole and whitebark pine from mountain pine beetle (Coleoptera: Curculionidae: Scolytinae) attacks. *Journal of Economic Entomology* 98(5): 1614-1621.
- Berdeen, J.C., Riley, L.E., and Sniezko, R.S. 2007. Whitebark pine seed storage and germination: a follow-up look at seedlots from Oregon and Washington. Pp. 113-121. *Proceedings: Conference on Whitebark pine: A Pacific Coast Perspective*. USDA Forest Service R6-NR-FHP-2007-01.

- Bockino, N.k. 2008. Interactions of white pine blister rust, host species, and mountain pine beetle in whitebark pine ecosystems in the Greater Yellowstone. Master's Thesis University of Wyoming, Laramie, WY.
- Bower, A.D. and Aitken, S.N. 2008. Ecological genetics and seed transfer guidelines for *Pinus albicaulis* (*Pinaceae*). *American Journal of Botany* 95(1): 66-76.
- Boyle, C.D. and Robertson, W.J. 1987. Use of mycelial slurries of mycorrhizal fungi as inoculum for commercial tree seedling nurseries. *Canadian Journal of Forest Research* 17(12): 1480-1486.
- Bradley, A.F., Noste, N.V., and Fischer, W.C. 1992. Fire ecology of forests and woodlands in Utah. General Technical Report INT-287. U.S. Department of Agriculture, Forest Service, Intermountain Research Station, Ogden, UT. 128 p.
- Brundrett, M., Bougher, N., Dell, B., Grove, T., and Malajczuk, N. 1996. Working with mycorrhizas in forestry and agriculture. *ACIAR Monograph* 32.
- Brundrett, M., Malajczuk, N., Mingquin, G., Daping, X., Snelling, S., and Dell, B. 2005. Nursery inoculation of Eucalyptus seedlings in Western Australia and Southern China using spores and mycelial inoculum of diverse ectomycorrhizal fungi from different climatic regions. *Forest Ecology and Management* 209: 193-205.
- Brunner, I. and Brodbeck, S. 2001. Response of mycorrhizal Norway spruce seedlings to various nitrogen loads and sources. *Environmental Pollution* 114: 223-233.
- Bruns, T.D., Bidartondo, M.I., and Taylor, L. 2002. Host specificity in ectomycorrhizal communities: what do the exceptions tell us? *Integrative and Comparative Biology* 42: 352-359.
- Bruns, T.D., Peay, K.G., Boynton, P.J., Grubisha, L.C., Hynson, N.A., Nguyen, N.H., and Rosenstock, N.P. 2009. Inoculum potential of *Rhizopogon* spores increases with time over the first 4 yr of a 99-yr spore burial experiment. *New Phytologist* 181(2): 463-470.
- Burns, K.S., Schoettle, A.W., Jacobi, W.R., and Mahalovich, M.F. 2008. Options for the management of white pine blister rust in the Rocky Mountain region. USDA Forest Service General Technical Report RMRS-GTR-206.
- Burr, K., Eramian, A., and Eggleston, K. 2001. Growing whitebark pine seedlings for restoration. Pp. 325-345. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds). *Whitebark pine communities: ecology and restoration*. Island Press, Washington, DC.

- Cairney, J.W. and Bastias, B.A. 2007. Influences of fire on soil fungal communities. *Canadian Journal of Forest Research* 307: 207-215.
- Campbell, D.B., Jones, M.D., Kiiskila, S. and Bulmer, C. 2003. Two-year field performance of lodgepole pine seedlings: effects of container type, mycorrhizal fungal inoculants, and site preparation. *B.C. Journal of Ecosystem Management* 3(2): 1-11.
- Carolin, T. 2006. Whitebark and limber pine restoration in Glacier National Park: monitoring results. *Nutcracker Notes* 10: 14.
- Castellano, M.A. and Molina, R. 1989. Mycorrhizae. In: Landis, T.D., Tinus, R.W., McDonald, S.E., and Bamett, J.P., (cords. and eds.). *The Container Tree Nursery Manual*, vol. V. USDA Forest Service, Public Affairs Office, Washington, DC. p 101-167.
- Castellano, M.A., Trappe, J.M., and Molina, R. 1985. Inoculation of container-grown Douglas-fir seedlings with basidiospores of *Rhizopogon vinicolor* and *R. colossus*: effects of fertility and spore application rate. *Canadian Journal of Forest Research* 15: 10-13.
- Castellano, M.A. 1996. Outplanting performance of mycorrhizal inoculated seedlings. Pp. 223-301. In: Mukerji, K.G. (ed.). *Concepts in Mycorrhizal Research*. Kluwer Academic Publishers, Netherlands. 371 p.
- Castro, J., Zamora, R., Hódar, J.A., and Gómez, J.M. 2002. Use of shrubs as nurse plants: a new technique for reforestation in Mediterranean Mountains. *Restoration Ecology* 10(2): 297-305.
- Certini, G. 2005. Effects of fire on properties of forest soils: a review. *Oecologia* 143: 1-10.
- Chakravarty, P. and Chataropaul, L. 1989. Effect of fertilization on seedling growth, ectomycorrhizal symbiosis, and nutrient uptake in *Larix laricina*. *Canadian Journal of Forest Research* 20: 245-248.
- Chadwick, A.C. 2002. *Carex geyeri*. In: *Fire Effects Information System*. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station, Fire Sciences Laboratory (Producer). [Online]. Available: <http://www.fs.fed.us/database/feis/> [2011, June 20].
- Chen, Y.L., Dell, B., and Malajczuk, N. 2006. Effects of *Scleroderma* spore density and age on mycorrhiza formation and growth of containerized *Eucalyptus globulus* and *E. urophylla* seedlings. *New Forests* 31(3): 453-467.

- Coen, G.D. and Holland, W.D. 1976. Soils of Waterton Lakes National Park, Alberta. Alberta Institute of Pedology S-73-33, Information Report NOR-X-65. 114 p.
- Coop, J.D. and Schoettle, A.W. 2009. Regeneration of Rocky Mountain bristle cone pine (*Pinus aristata*) and limber pine (*Pinus flexilis*) three decades after stand replacing fires. *Forest Ecology and Management* 257: 893-903.
- Cordell, C.E. and Marx, D.H. 1994. Effects of nursery cultural practices on management of specific ectomycorrhizae on bareroot tree seedlings. Pp. 133-152. In: Pfleger, F.L. and Linderman, R.G. (eds.). *Mycorrhizae and Plant Health*. APS Press, St. Paul, Minnesota. 344 p.
- COSEWIC. 2010. COSEWIC assessment and status report on the Whitebark Pine *Pinus albicaulis* in Canada. Committee on the Status of Endangered Wildlife in Canada. Ottawa. Pp 44. (www.sararegistry.gc.ca/status/status_e.cfm).
- Crane, M. F. 1990. *Xerophyllum tenax*. In: Fire Effects Information System. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station, Fire Sciences Laboratory (Producer). [Online]. Available: <http://www.fs.fed.us/database/feis/> [2011, June 20].
- Cripps, C.L. 2004. *Fungi in forest ecosystems: systematics, diversity and ecology*. New York Botanical Gardens Press, NY. 363 p.
- Cripps, C.L. 2011. [Personal Communication]. July 15, 2011. Associate Professor, Montana State University, Bozeman, Montana.
- Cripps, C.L. and Antibus, R.K. 2011. Native ectomycorrhizal fungi of limber and whitebark pine: necessary for forest sustainability? In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). *The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium*. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Cripps, C.L. and Eddington, L.H. 2005. Distribution of mycorrhizal types among alpine vascular plant families on the Beartooth Plateau, Rocky Mountains, U.S.A., in reference to large-scale patterns in arctic-alpine habitats. *Arctic, Antarctic, and Alpine Research* 37(2): 177-188.
- Cripps, C.L. and Grimme, E. 2011. Inoculation and successful colonization of whitebark pine seedlings with native mycorrhizal fungi under greenhouse conditions. In:

- Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Cripps, C.L. and Lonergan, E. 2011. Whitebark pine and ectomycorrhizal fungi: summary and update on various projects. Whitebark Pine Ecosystem Foundation Meeting, Cody, WY.
- Cripps, C.L., Smith, C., Carolin, T., and Lapp, J. 2008. Ectomycorrhizal fungi with whitebark pine. *Nutcracker Notes*. 14: 12-13.
- Cripps, C.L. and Trusty, P. 2007. Final Report. Whitebark pine restoration, Dunraven Pass: Monitoring mycorrhizal status of seedlings. Greater Yellowstone Coordinating Committee.
- Danielson, R.M. 1991. Temporal changes and effects of amendments on the occurrence of sheathing (ecto-) mycorrhizas of conifers growing in oil sands tailings and coal spoil. *Agriculture Ecosystems & Environment* 35: 261–281.
- Deacon, J.W. and Flemming, L.V. 1992. Interactions of ectomycorrhizal fungi. Pp. 249-300. In: Allen, M.F. (ed.). *Mycorrhizal functioning: an integrative plant-fungal process*. Chapman and Hall, New York.
- Diaz, G., Carrilla, C., and Honrubia, M. 2010. Mycorrhization, growth, and nutrition of *Pinus halepensis* seedlings fertilized with different doses and sources of nitrogen. *Annals of Forest Science* 67(405): 1-9.
- Dominguez-Lerena, S., Herrero Sierra, N., Carrasco Manzano, I., Ocana Bueno, L., Penuelas Rubira, J.L., and Mexal J.G. 2006. Container characteristics influence *Pinus pinea* seedling development in the nursery and field. *Forest Ecology and Management* 221(1): 63-71.
- Dumroese, R.K. 2008. Observations on root disease of container whitebark pine seedlings treated with biological controls. *Native Plants Journal* 9(2): 92-97.
- Dumroese, K., Page-Dumroese, D.S., Salifu, K.F., and Jacobs, D.F. 2005. Exponential fertilization of *Pinus monticola* seedlings: nutrient uptake efficiency, leaching fractions, and early out-planting performance. *Canadian Journal of Forest Research* 35: 2961-2967.
- Eggleston, K. 2010. [Personal Communication to Dr. Cathy Cripps]. April 19, 2010. U.S. Department of Agriculture Forest Service Nursery, Coeur D'Alene, Idaho.

- Ekwebelam, S.A. and Reid, C.P.P. 1983. Effects of light, nitrogen fertilization, and mycorrhizal fungi on growth and photosynthesis of lodgepole pine seedlings. *Canadian Journal of Forest Research* 13(6): 1099-1106.
- El Karkouri, K., Martin, F., Douzery, J.P., and Mousain, D. 2005. Diversity of ectomycorrhizal fungi naturally established on containerized *Pinus* seedlings in nursery conditions. *Microbiological Research* 160: 47-52.
- Ellison, A.M., Bank, M.S., Clinton, B.D., Colburn, E.A., Elliott, K., Ford, C.R., Foster, D.R., Kloeppel, B.D., Knoepp, J.D., Lovett, G.M., Mohan, J., Orwig, D.A., Rodenhouse, N.L., Sobczak, W.V., Stinson, K.A., Stone, J.K., Swan, C.M., Thompson, J., Von Holle, B., and Webster, J.R. 2005. Loss of foundation species: consequences for the structure and dynamics of forested ecosystems. *Frontiers in Ecology and the Environment* 3: 479-486.
- Entry, J.A., Rygielwicz, P.T., Watrud, L.S., Donnelly, P.K. 2002. Influence of adverse soil conditions on the formation and function of arbuscular mycorrhizas. *Advances in Environmental Research* 7910: 123-138.
- ESRI 2011. ArcGIS Desktop: Release 10. Redlands, CA: Environmental Systems Research Institute.
- Farnes, P.E. 1990. SNOTEL and snow course data: describing the hydrology of whitebark pine ecosystems. Pp. 302-304. In: Schmidt, W.C. and McDonald, K.J. (comps.). *Proceedings: Symposium on whitebark pine ecosystems: ecology and management of a high-mountain resource*. USDA Forest Service, General Technical Report INT-270, Intermountain Research Station, Ogden, Utah.
- Felicetti, L.A., Schwartz, C.C., Rye, R.O., Haroldson, M.A., Gunther, K.A., Phillips, D.L., and Robbins, C.T. 2003. Use of sulfur and nitrogen stable isotopes to determine the importance of whitebark pine nuts to Yellowstone grizzly bears. *Canadian Journal of Zoology* 81(5): 763-770.
- Fettig, C.J., Dabney, C.P., McKelvey, S.R., and Huber, P.W. 2008. Non-host angiosperm volatiles and verbenone protect individual ponderosa pines from attack by western pine beetle and red turpentine beetle (Coleoptera: Curculionidae, Scolytinae). *Western Journal of Applied Forestry* 23(1): 40-45.
- Fischer, W.C. and Bradley, A.F. 1987. Fire ecology of western Montana forest habitat types. General Technical Report INT-223. U.S. Department of Agriculture, Forest Service, Intermountain Research Station, Ogden, UT. 101 p.

- Fortin, J.K. 2011. Niche separation of grizzly (*Ursus arctos*) and American black bears (*Ursus americanus*) in Yellowstone National Park. Washington State University PhD dissertation, Pullman, WA.
- Frankland, J.C. 1998. Fungal succession – unraveling the unpredictable. *Mycological Research* 102: 1-15.
- Fryer, J.L. 2002. *Pinus albicaulis*. In: Fire Effects Information System, [Online]. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station, Fire Sciences Laboratory (Producer). Available: <http://www.fs.fed.us/database/feis/> [2011, May 23].
- Gagne, A., Jany, J.L., Bousquet, J., Khasa, J., and Gamase, P. 2006. Ectomycorrhizal fungal communities of nursery-inoculated seedlings out-planted on clear-cut sites in northern Alberta. *Canadian Journal of Forest Research* 36: 1684-1694.
- Gagnon, J., Langlois, C.G., and Fortin, J.A. 1988. Growth and ectomycorrhiza formation of containerized black spruce seedlings as affected by nitrogen fertilization, inoculum type, and symbionts. *Canadian Journal of Forest Research* 18: 922-929.
- Geils, B.W., Hummer, K.E., and Hunt, R.S. 2010. White pines, *Ribes*, and blister rust: a review and synthesis. *Forest Pathology* 40(3-4): 147-185.
- Gernandt, D.S., López, G.G., García, S.O., and Liston, A. 2005. Phylogeny and classification of *Pinus*. *Taxon* 54(1): 29-42.
- González-Ochoa, A.I., Heras, J., Torres, P., and Sánchez-Gómez, E. 2003. Mycorrhization of *Pinus halepensis* Mill. and *Pinus pinaster* Aiton seedling in two commercial nurseries. *Annals of Forest Science* 60: 43-48.
- Government of Alberta. 2010. Species assessed by Alberta's Endangered Species Conservation Committee: short list [online]. Available: <http://www.srd.alberta.ca/BiodiversityStewardship/SpeciesAtRisk/SpeciesSummaries/documents/SpeciesAssessed-EndangeredSpeciesConservationCommittee> [June 3, 2010].
- Government of Alberta. 2012. Environment and Sustainable Resource Development [online]. Available: <http://www.environment.alberta.ca/apps/basins/DisplayData.aspx?Type=Figure&BasinID=14&DataType=4&StationID=AKAM>.
- Government of Canada. 2012. Order amending Schedule 1 to the Species at Risk Act. *Canada Gazette Part II* 146 (14) SOR/2012-113: 1418-1629. [online] Available: http://www.sararegistry.gc.ca/virtual_sara/files/orders/g2-14614i_e.pdf.

- Gibson, K., Skov, K., Kegley, S., Jorgensen, C., Smith, S., and Witcosky, J. 2008. Mountain pine beetle impacts in high-elevation five-needle pines: current trends and challenges. USDA Forest Service, Northern region, Missoula, MT. R1-08-020.
- Hangs, R.D., Knight, J.D., and Van Rees, K.C.J. 2002. Interspecific competition for nitrogen between early successional species and planted white spruce and jack pine seedlings. *Canadian Journal of Forest Research* (32): 1813-1821.
- Heumader, J. 1992. Cultivation of cembran pine plants for high-elevation afforestations. International workshop on subalpine stone pines and their environment: the status of our knowledge, St. Moritz, Switzerland.
- Hicke, J.A., Logan, J., Powell, J., and Ojima, D.S. 2006. Changing temperatures influence suitability for modeled mountain pine beetle (*Dendroctonus ponderosae*) outbreaks in the western United States. *Journal of Geophysical Research* (111): 1-12.
- Hoff, R.J., Ferguson, D.E., McDonald, G.I., and Keane, R.E. 2001. Strategies for managing whitebark pine in the presence of white pine blister rust. Pp. 346-366. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds). *Whitebark pine communities: ecology and restoration*. Island Press, Washington, DC.
- Hollingsworth, L.T. 2010. *Davies Fire: fire behavior and fire effects analysis*. Fire Modeling Institute, Missoula Fire Sciences Laboratory, Rocky Mountain Research Station.
- Horton, T.R., Cázares, E., and Bruns, T.D. 1998. Ectomycorrhizal, vesicular-arbuscular and dark septate fungal colonization of bishop pine (*Pinus muricata*) seedlings in the first 5 months of growth after wildfire. *Mycorrhiza* 8: 11-18.
- Hunt, G. 1992. Effects of mycorrhizal fungi on quality of nursery stock and plantation performance in the southern interior of British Columbia. FRDA Report 185. Forestry Canada and the British Columbia Ministry of Forests.
- IBM Corporation. Released 2011. *IBM SPSS Statistics for Windows, version 20.0*. Armonk, NY: IBM Corp.
- Ingestad, T., and Lund, A.B. 1986. Theory and techniques for steady state mineral nutrition and growth of plants. *Scandinavian Journal of Forest Research* 1: 439–453.
- Izlar, K. 2007. Assessment of whitebark pine seedling survival for Rocky Mountain plantings. University of Montana Master's thesis. Missoula, MT.

- Izlar, K. 2009. [Personal Communication to Dr. Cathy Cripps] 2009. U.S. Forest Service, Flathead National Forest, Montana.
- Jackson, D.P., Dumroese, R.K., and Barnett, J.P. 2012. Nursery response of container *Pinus palustris* seedlings to nitrogen supply and subsequent effects on outplanting performance. *Forest Ecology and Management* 265: 1-12.
- James, R.L. and Burr, K.E. 2000. Diseases associated with whitebark pine seedling production. Coeur d'Alene, Idaho: USDA Forest Service Nursery, Forest Health Protection, May 2000, Report 00-8: 1-8.
- Keane, R.E. and Arno, S.F. 1993. Rapid decline of whitebark pine in western Montana: evidence from 20-year re-measurements. *Western Journal of Applied Forestry* 8: 44-47.
- Keane, R.E. and Parsons, R.A. 2010. Management guide to ecosystem restoration treatments: whitebark pine forests of the northern Rocky Mountains, U.S.A. General Technical Report RMRS-GTR-232. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 133 p.
- Keane, R.E. and Schoettle, A.W. 2011. Strategies, tools, and challenges for sustaining and restoring high elevation five-needle white pine forests in Western North America. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). *The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.*
- Keane, R.E., Tomback, D.F., Aubry, C.A., Bower, A.D., Campbell, E.M., Cripps, C.L., Jenkins, M.B., Manning, M., McKinney, S.T., Murray, M.P., Perkins, D.L., Reinhart, D.P., Ryan, C., Schoettle, A.W., Smith, C.M. 2012. A range-wide restoration strategy for whitebark pine (*Pinus albicaulis*). General Technical Report RMRS-GTR-279. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 108 p.
- Keller, G. 1996. Utilization of inorganic and organic nitrogen sources by high-subalpine ectomycorrhizal fungi of *Pinus cembra* in pure culture. *Mycological Research* 100: 989-998.
- Kendall, K.C. and Keane, R.E. 2001. Whitebark pine decline: infection, mortality, and populations trends. Pp. 221-242. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds.). *Whitebark pine communities: ecology and restoration*. Island Press, Washington DC.

- Kegley, S. and Gibson, K. 2004. Protecting whitebark pine trees from mountain pine beetle attack using verbenone. Forest Health Protection Report 04-8.
- Kegley, S. and Gibson, K. 2009. Individual-tree tests of verbenone and green-leaf volatiles to protect lodgepole, whitebark and ponderosa pines, 2004-2007. Forest Health Protection Report 09-03.
- Khasa, P.D., Piche, Y., and Coughlan, A.P. (eds.). 2009. Advances in Mycorrhizal Science and Technology. NRC Research Press, Ottawa. 179 p.
- Khasa, P.D., Sigler, L., Chakravarty, P., Dancik, B.P., Erikson, L., and McCurdy, D. 2001. Effect of fertilization on growth and ectomycorrhizal development of container-grown and bare-root nursery conifer seedlings. *New Forests* 22: 179-197.
- Landhausser, S., Stadt, K., and Lieffers, V. 1996. Screening for control of a forest weed: early competition between three replacement species and *Calamagrostis canadensis* or *Picea glauca*. *Journal of Applied Ecology* 33: 1517-1526.
- Landis, T.D., Dumroese, R.K., and Haase, D.L. 2010a. Seedling processing, storage, and outplanting, vol. 7. The container Tree Nursery Manual. Agric. Handbook, 674. U.S. Department of Agriculture Forest Service, Washington, D.C. 200 p.
- Landis, T.D., Seinfeld, D.E., and Dumroese, R.K. 2010b. Native plant containers for restoration projects. *Native Plants Journal* 11(3): 341-348.
- Landis, T.D., Tinus, R.W., McDonald, S.E., and Barnett, J.P. 1990. Containers and growing media, vol. 2. The container Tree Nursery Manual. Agric. Handbook, 674. U.S. Department of Agriculture Forest Service, Washington, D.C. 88 p.
- Landis, T.D., Tinus R.W., McDonald S.E., Barnett J.P. 1989a. Seedling nutrition and irrigation, vol. 4. The container Tree Nursery Manual. Washington (D.C.): USDA Forest Service, Agriculture Handbook. 119 p.
- Landis, T.D., Tinus, R.W., McDonald, S.E., Barnett, J.P. 1989b. The biological component: nursery pests and mycorrhizae, vol. 5. The container Tree Nursery Manual. Washington (D.C.): USDA Forest Service, Agriculture Handbook. 674. 171 p
- Lanner, R.M. 1996. Made for each other: a symbiosis of birds and pines. Oxford University Press. New York, NY.
- Lantz, G. 2010. Whitebark pine: an ecosystem in peril. American Forests Special Report. Norman, OK.

- Largent, D.L., Sugihara, N. and Wishner, C. 1980. Occurrence of mycorrhizae on ericaceous and pyrolaceous plants in northern California. *Canadian Journal of Botany* 58: 2274-2279.
- Larson, E.R. and Kipfmüller, K.F. 2010. Patterns in whitebark pine regeneration and their relationships to biophysical site characteristics in southwest Montana, central Idaho, and Oregon, USA. *Canadian Journal of Forestry* 40: 476-487.
- Lehto, T. and Zwiazek, J.J. 2011. Ectomycorrhizas and water relations of trees: a review. *Mycorrhiza* 21(2): 71-90.
- Leslie, A. and Wilson, B. 2011. No free lunch: observations on seed predation, cone collection and controlled germination of whitebark pine from the Canadian Rockies. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). Little Jr., E.L. 1971. Atlas of United States trees, volume 1, conifers and important hardwoods: U.S. Department of Agriculture Miscellaneous Publication 1146, 9 p., 200 maps.
- Lieffers, V.J., Macdonald, S.E. and Hogg, E.H. (1993) Ecology of and control strategies for *Calamagrostis canadensis* in boreal forest sites. *Canadian Journal of Forest Research* 23: 2070-2077.
- Little, E.L. Jr., 1971. Atlas of United States trees, volume 1, conifers and important hardwoods: U.S. Department of Agriculture Miscellaneous Publication 1146. 9 p. and 200 maps.
- Logan, J., McFarlane, W., and Wilcox, L. 2010. Whitebark pine vulnerability to climate driven mountain pine beetle disturbance in the Greater Yellowstone Ecosystem. *Ecological Applications* 20(4): 895-902.
- Lonergan, E. and Cripps, C.L. (in ed.). Low nitrogen fertilizer as a strategy to maintain ectomycorrhizal colonization of inoculated whitebark pine seedlings before out-planting. *Native Plants Journal*, in process.
- Luckman, B.H., Jozsa, L.A., and Murphy, P.J. 1984. Living seven-hundred-year-old *Picea engelmannii* and *Pinus albicaulis* in the Canadian Rockies. *Arctic and Alpine Research*. 16(4): 419-422.
- Mahalovich, M.F., Burr, K.E., and Foushee, D.L. 2006. Whitebark pine germination, rust resistance, and cold hardiness among seed sources in the inland northwest: planting strategies for restoration. USDA Forest Service Proceedings RMRS-P-43.

- Mahalovich, M.F. and Dickerson, G.A. 2004. Whitebark pine genetic restoration program for the Intermountain West (USA). Pp. 181-187. In: Sniezko, R., Samman, S., Schlarbaum, S., and Kriebel, H. (eds.). Breeding and genetic resources of five-needle pines: growth, adaptability and pest resistance, IUFRO Working Party 2.02.15, Proceedings. USDA Forest Service, Rocky Mountain Research Station, RMRS-P-32, Ogden, UT.
- Mahalovich M.F. and Hoff, R.J. 2000. Whitebark pine operational cone collection instructions and seed transfer guidelines. Nutcracker Notes 11: 10-13.
- Mattson, D.J., Blanchard, B.M., and Knight, R.R. 1992. Yellowstone grizzly bear mortality, human habituation, and whitebark pine seed crops. *Journal of Wildlife Management* 56: 432-442.
- Mattson, D.J. and Reinhart, D.P. 1994. Bear use of whitebark pine seeds in North America. Pp. 212–220. U.S. Department of Agriculture, Forest Service General Technical Report INTGTR-309.
- Marx, D. H. 1980. Ectomycorrhiza fungus inoculations: A tool for improving forestation practices. Pp. 13–71. In: Mikola, P. (ed.). *Tropical Mycorrhiza Research*. Oxford University Press, Oxford. 270 p.
- Marx, D.H. 1991. The practical significance of ectomycorrhizae in forest establishment. Pp. 54–90. In: Hägglund, B. (ed.). *Ecophysiology of Mycorrhizae of Forest Trees*. Proceedings. The Marcus Wallenberg Foundation, Stockholm, Sweden.
- Marx, D.H. and Cordell, C.E. 1988. Specific ectomycorrhizae improve reforestation and reclamation in eastern United States. In: Lalonde, M. and Piché, Y., (eds. and eds). *Symposium Proceedings, Canadian Workshop on Mycorrhizae in Forestry; May 1-4, 1988; Centre de recherche en biologie forestière, Facult de foresterie et de géodesie, Université Laval, Sainte-Foy, Qué.*
- McAlister, J.A., and Timmer, V.R. 1998. Nutrient enrichment of white spruce seedlings during nursery culture and initial plantation establishment. *Tree Physiology* 18: 195–202.
- McCaughey, W.W. 1990. Biotic and microsite factors affecting *Pinus albicaulis* establishment and survival. Ph.D. dissertation Montana State University, Bozeman, MT.
- McCaughey, W.W. and Schmidt, W.C. 2001. Taxonomy, distribution and history. Pp. 29-40. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds.). *Whitebark pine communities, ecology and restoration*. Island Press, Washington, DC.

- McCaughey, W., Scott, G.L., and Izlar, K.L. 2009. Whitebark pine planting guidelines. *Western Journal of Applied Forestry* 24(3): 163-166.
- McCaughey, W.W. and Tomback, D.F. 2001. The natural regeneration process. Pp. 105-120. In: Tomback, D.F.; Arno, S.F.; Keane, R.E. (eds.). *Whitebark pine communities: Ecology and restoration*. Island Press, Washington, DC.
- McCune, B. 1988. Ecological diversity in North American pines. *American Journal of Botany* 75(3): 353-368.
- McDonald, G.I. and Hoff, R.J. 2001. Blister rust: an introduced plague. Pp. 193-220. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds.). *Whitebark pine communities, ecology and restoration*. Island Press, Washington, DC.
- McKinney, S.T. and Tomback, D.F. 2007. The influence of white pine blister rust on seed dispersal in whitebark pine. *Canadian Journal of Forest Research* 37: 1044-1057.
- McKinney, S., Fiedler, C.E., and Tomback, D.F. 2009. Invasive pathogen threatens bird-pine mutualism: implications for sustaining a high-elevation ecosystem. *Ecological Applications* 19(3): 597-607.
- Meikle, T.W. and Amaranthus, M. 2008. The influence of fertilization regime and mycorrhizal inoculum on outplanting success: a field trial of containerized seedlings in Oregon. *Native Plants Journal* 9(2): 107-116.
- Mellmann-Brown, S. 2005. The regeneration of whitebark pine in the timberline ecotone of the Beartooth Plateau, U.S.A.: spatial distribution and responsible agents. Pp. 96-116. In: Broll, G. and Keplin, B. (eds.). *Mountain ecosystems: studies in treeline ecology*. Springer-Verlag, Berlin.
- Menkis, A., Vasiliauskas, R., Taylor, A.F.S., Stenlid, J., and Finlay, R. 2005. Fungal communities in mycorrhizal roots of conifer seedlings in forest nurseries under different cultivation systems, assessed by morphotyping, direct sequencing, and mycelial isolation. *Mycorrhiza* 16: 33-41.
- Menkis, A., Vasiliauskas, R., Taylor, A.F.S., Senlid, J., and Finlay, R. 2007. Afforestation of abandoned farmland with conifer seedlings inoculated with three ectomycorrhizal fungi- impact on plant performance and ectomycorrhizal community. *Mycorrhiza* 17(4): 337-348.
- Mohatt, K. 2006. Ectomycorrhizal fungi of whitebark pine (*Pinus albicaulis*) in the Northern Yellowstone Greater Ecosystem. Montana State University Master's Thesis, Bozeman, MT.

- Mohatt, K., Cripps, C.L., and Lavin, M. 2008. Ectomycorrhizal fungi of whitebark pine (a tree in peril) revealed by sporocarps and molecular analysis of mycorrhizae from treeline forests in the Greater Yellowstone Ecosystem. *Botany* 86: 14-25.
- Molina, R., Massicotte, H., and Trappe, J.M. 1992. Specificity phenomena in mycorrhizal symbiosis: community-ecological consequences and practical implications. In: Allen, M.F. (ed.). *Mycorrhizal Functioning*. Chapman and Hall, London, UK.
- Moody, R.J. 2006. Post-fire regeneration and survival of whitebark pine (*Pinus albicaulis* Engelm.). University of British Columbia Master's Thesis, Vancouver, British Columbia.
- Morgan, P., Bunting, S.C., Keane, R.E., and Arno, S.F. 1994. Fire ecology of whitebark pine forests of the Northern Rocky Mountains, U.S.A. Pp 136-141. In: Schmidt, W.C. and Holtmeier, F.K., (comps.). *Proceedings of the international workshop of subalpine stone pines and their environment: the status of our knowledge; September 5-11, 1992; St. Moritz, Switzerland*. General Technical Report INT-GRT-309. Ogden, UT: U.S. Department of Agriculture, Forest Service, Intermountain Research Station.
- Moser, M.M. 1956. Die Bedeutung der Mykorrhiza für Aufforestungen in Hochlagen. *Forstwissenschaftliches Centralblatt* 75: 8-18.
- Moser, M.M. 2004. Subalpine conifer forests in the Alps, the Altai, and the Rocky Mountains: a comparison of their fungal populations. Pp. 151-158. In: Cripps, C.L. (ed.), *Fungi in forest ecosystems: systematics, diversity, and ecology*. New York Botanical Garden Press, NY.
- Murray, M.P. 2007. Cone collecting techniques for whitebark pine. *Western Journal of Applied Forestry* 22(3): 153-155.
- Nicholas, A. and Katzenberger, D. 2011. Whitebark pine to be designated a candidate for endangered species protection. U.S. Dept. of Interior, Fish and Wildlife Service, Press Release, Lakewood, CO.
- Norment, C.J. 1991. Bird use of forest patches in the subalpine forest-alpine tundra ecotone of the Beartooth Mountains, Wyoming. *Northwest Science* 65(1): 1-10.
- Oliveira, R.S., Franco, A.R., Vosatka, M., and Castro, P.M.L. 2010 Management of nursery practices for efficient ectomycorrhizal fungi application in the production of *Quercus ilex*. *Symbiosis* 52(2-3): 125-131.

- Ortega, U., Dunabeitia, M., Mendez, S., Gonzalez-murua, C., and Majada, J. 2004. Effectiveness of mycorrhizal inoculation in the nursery on growth and water relations of *Pinus radiata* in different water regimes. *Tree Physiology* 24: 65-73.
- Parks Canada. 2010. Parks Canada Invests in Ecological Integrity-Press Release. [online] Accessed: http://www.pc.gc.ca/apps/cp-nr/release_e.asp?bgid=1338&andor1=bg.
- Parladé, J., Luque, J., Pera, J., Rincón, A.M. 2004. Field performance of *Pinus pinea* and *P. halepensis* seedlings inoculated with *Rhizopogon* spp. and outplanted in formerly arable land. *Annals of Forest Science* 61: 507-514.
- Parladé, J., Pera, J., and Alvarez, F. 1996. Inoculation of containerized *Pseudotsuga menziesii* and *Pinus pinaster* seedlings with spores of five species of ectomycorrhizal fungi. *Mycorrhiza* 6(4): 237-245.
- Peintner U. 2012. [Personal Communication to Dr. Cathy Cripps]. Professor of Biology, University of Innsbruck, Innsbruck, Austria.
- Perkins, D.L. and Swetnam, T.W. 1996. A dendroecological assessment of whitebark pine in the Sawtooth--Salmon River region, Idaho. *Canadian Journal of Forest Research*. 26: 2123-2133.
- Perkins, J. 2004. *Pinus albicaulis* seedling regeneration after fire. University of Montana PhD dissertation, Missoula, MT.
- Price, R.A., Liston, A., and Strauss, S.H. 1998. Phylogeny and systematics of *Pinus*. Pp. 49-68. In: Richardson, D.M. (ed.). *Ecology and biogeography of Pinus*. Cambridge University Press, Cambridge, UK.
- Quoreshi, A.M., Kernaghan, G., and Hunt, G.A. 2009. Mycorrhizal fungi in Canadian forest nurseries and field performance of inoculated seedlings. Pp. 115-128. In: Khasa, P.D., Piche, Y., and Coughlan, A.P. (eds.). *Advances in Mycorrhizal Science and Technology*. NRC Research Press, Ottawa. 197 p.
- Quoreshi, A. M., Piche, Y., and Khasa, D.P. 2008. Field performance of conifer and hardwood species 5 years after nursery inoculation in the Canadian Prairie Provinces. *New Forests* 35: 235-253.
- Quoreshi, A.M. and Timmer, V.R. 1998. Exponential fertilization increases nutrient uptake and ectomycorrhizal development of black spruce seedlings. *Canadian Journal of Forestry* 28: 674-682.

- R Development Core Team. 2008. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL <http://www.R-project.org>.
- Read, D.J. 1998. The mycorrhizal status of *Pinus*. Pp. 324-340. In: Richardson, D.M. (ed.). Ecology and biogeography of *Pinus*. Cambridge University Press, Cambridge, UK.
- Repáč, I. 1996a. Inoculation of *Picea abies* (L.) Karst. seedlings with vegetative inocula of ectomycorrhizal fungi *Suillus bovinus* (L.:Fr.) O. Kuntze and *Inocybe lacera* (Fr.) Kumm. New Forests 12: 41-54.
- Repáč, I. 1996b. Effects of forest litter on mycorrhiza formation and growth of container - grown Norway spruce (*Picea abies* [L.] Karst.) seedlings. Lesnictví 42: 317-324.
- Repáč, I. 2007. Ectomycorrhiza formation and growth of *Picea abies* seedlings inoculated with alginate-bead fungal inoculum in peat and bark compost substrates. Forestry 80(5): 517-530.
- Repáč, I. 2011. Ectomycorrhizal Inoculum and Inoculation Techniques. Pp 43-63. In: Rai, M. and Varma, A. (eds.). Diversity and Biotechnology of Ectomycorrhizae, Soil Biology 25, Springer - Verlag, Heidelberg, Berlin. 459 p.
- Resler, L.M. 2004. Conifer establishment sites on a periglacial landscape, Glacier National Park, Montana. Texas State University Ph.D. dissertation, San Marcos, TX. 186 p.
- Rikala, R., Heiskanen, J., and Lahti, M. 2004. Autumn fertilization in the nursery affects growth of *Picea abies* container seedlings after transplanting. Scandinavian Journal of Forest Research 19(5): 409-414.
- Riley, L.E., Coumas, C.M., Danielson, J.F., and Berdeen, J.C. 2007. Seedling nursery culture of whitebark pine at Dorena Genetic Resource Center: headaches, successes, and growing pains. USDA Forest Service Proceedings of the Conference Whitebark Pine: a pacific coast perspective. R6-NR-FHP-2007-01.
- Rincón, A., de Felipe, M.R., and Fernández-Pascual, M. 2007. Inoculation of *Pinus halepensis* Mill. with selected ectomycorrhizal fungi improves seedling establishment 2 years after planting in a degraded gypsum soil. Mycorrhiza 18: 23-32.
- Rincón, A., Parlade, J., and Pera, J. 2005. Effects of ectomycorrhizal inoculation and the type of substrate on mycorrhization, growth and nutrition of containerized *Pinus*

- pinea* L. seedlings produced in a commercial nursery. *Annals of Forest Science* 62: 817-822.
- Ronikier, M., Miskiewicz, A., and Mleczko, P. 2002. Presence and distribution of *Suillus plorans* in the Polish Tatra Mountains (Western Carpathians). *Acta Societatis Botanicorum Poloniae* 71(3): 235-242.
- Schmid, V. 2006. Entwicklung molekularer methoden für ein schnelles und kostengünstiges monitoring der inokulation von forstpflanzen mit ektomykorrhizasymbionten. University of Innsbruck PhD dissertation, Innsbruck, Austria.
- Schoettle, A.W. and Sniezko, R.A. 2007. Proactive intervention to sustain high-elevation pine ecosystems threatened by white pine blister rust. *Journal of Forest Research* 12: 327-336.
- Schönenberger, W. 2001. Cluster afforestation for creating diverse mountain forest structures – a review. *Forest Ecology and Management* 145: 121-128.
- Schwandt, J. 2006. Whitebark pine in peril: a case for restoration. U.S. Department of Agriculture, Forest Service, R1-06-28. Missoula, MT.
- Schwandt, J., Tomback, D.F., Keane, R.E., McCaughey, W.W., and Kearns, H. 2007. First year results of a whitebark pine seed planting trial near Baker City, OR. USDA Forest Service R6-NR-FHP-2007-01.
- Schwandt, J., Chadwick, K., and Kearns, H. 2011. Whitebark pine direct seeding trials in the Pacific Northwest. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). *The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium*. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Schwandt, J., DeMastus, C., Cripps, C.L. 2012. Whitebark pine direct seeding trials, unpublished data. Yellowstone Club, Montana.
- Schwanke, R. and Smith, C. 2010. Imitating lightning strikes for whitebark pine restoration. Whitebark Pine Ecosystem Foundation. *Nutcracker Notes* 19: 12.
- Schwartz, M.W., Hoeksema, J.D., Gehring, C.A., Johnson, N.C., Klironomos, J.N., Abbott, L.K, and Pringle, A. 2006. The promise and the potential consequences of the global transport of mycorrhizal fungal inoculum. *Ecology Letters* 9(5): 501-515.

- Scott, G.L., McCaughey, W.W., and Izlar, K. 2011. Guidelines for whitebark pine planting prescriptions. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Senstrom, E. and Ek, M. 1990. Field growth of *Pinus sylvestris* following nursery inoculation with mycorrhizal fungi. Canadian Journal of Forest Research 20: 914-918.
- Shaw, C.G., Molina, R., Walden, J. 1981. Development of ectomycorrhizae following inoculation of containerized Sitka and white spruce seedlings. Canadian Journal of Forest Research 12: 191-195.
- Shebitz, D.J., Reichard, S.H., and Dunwiddie, P.W. 2009. Ecological and cultural significance of burning beargrass habitat on the Olympic peninsula, Washington. Cultural Restoration 27(3): 306-319.
- Six, D. and Adams, J. 2007. White pine blister rust severity and selection of individual whitebark pine by the mountain pine beetle (*Coleoptera: Curculionidae, Scolytinae*) Journal of Entomological Science 42(3): 345-353.
- Smith, C.M. 2009. Restoration of whitebark and limber pine: first steps on a long road in Waterton Lakes National Park. BC Forest Professional, May-June 14-15.
- Smith, C.M. 2012. [Personal Communication]. Park Ecologist, Waterton-Lakes National Park, Alberta, Canada.
- Smith, C.M., Poll, G., Gillies, C., Praymak, C., Miranda, E., and Hill, J. 2011a. Limber pine seed and seedling experiment in Waterton Lakes National Park, Canada. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Smith, C.M., Shepherd, B., Gillies, C., and Stuart-Smith, J. 2011. Re-measurement of whitebark pine infection and mortality in the Canadian Rockies. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.

- Smith, C.M., Wilson, B., Rasheed, S., Walker, R.C., Carolin, T., and Shepard, B. 2008. Whitebark pine and white pine blister rust in the Rocky Mountains of Canada and Northern Montana. *Canadian Journal of Forest Research* 5: 982-995.
- Smith, S.E. and Read, D.J. 2008. *Mycorrhizal symbiosis*, 3rd edition. Academic Press, Inc., San Diego, CA.
- Snieszko, R.A. 2006. Resistance breeding against non-native pathogens in forest trees – current successes in North America. *Journal of Plant Pathology* 28: 270-279.
- Snieszko, R.A., Mahalovich, M.F., Schoettle, A.W., and Vogler, D.R. 2011. Past and current investigation of the genetic resistance to *Cronartium ribicola* in high-elevation five-needle pines. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). *The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium*. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Steinfeld, D., Amaranthus, M., and Cazares, E. 2003. Survival of ponderosa pine (*Pinus ponderosa* Dougl. Ex Laws.) seedlings out-planted with *Rhizopogon* mycorrhizae inoculated with spores at the nursery. *Journal of Arboriculture* 29(4): 197-208.
- Sylvie, D.M. and Jarstfer, A.G. 1997. Distribution of mycorrhiza on competing pines and weeds in a southern pine plantation. *Soil Science Society of America Journal* 61: 139-144.
- Tedersoo, L., May, T.W., and Smith, M.E. 2009. Ectomycorrhizal lifestyle in fungi: global diversity, distribution, and evolution of phylogenetic lineages. *Mycorrhiza* 20: 217-263.
- Teste, F.P., Schmidt, M.G., Berch, S.M., Bulmer, C., and Egger, K.N. 2004. Effects of ectomycorrhizal inoculants on survival and growth of interior Douglas-fir seedlings on reforestation sites and partially rehabilitated landings. *Canadian Journal of Forest Research* 34: 2074–2088.
- Timmer, V.R. 1996 Exponential nutrient loading: a new fertilization technique to improve seedlings performance on competitive sites. *New Forests* 13: 275-295.
- Tomback, D.F. 1982. Dispersal of whitebark pine seeds by Clark's Nutcracker: a mutualism hypothesis. *Journal of Animal Ecology* 51: 451-467.

- Tomback, D.F. 2001. Clark's nutcracker: agent of regeneration. Pp. 89-104. In: Tomback, D.F., Arno, S.F., and Keane, R.E. (eds.). Whitebark pine communities, ecology and restoration. Island Press, Washington, DC.
- Tomabck, D.F. and Achuff, P. 2010. Blister rust and western forest biodiversity: ecology, values, and outlook for white pines. *Forest Pathology* 40: 186-225.
- Tomback, D.F., Anderies, A.J., Carsey, K.S., Powell, M.L., and Mellmann-Brown, S. 2001c. Delayed seed germination in whitebark pine and regeneration patterns following the Yellowstone fires. *Ecology*. 82(9): 2587-2600.
- Tomback, D.F., Arno, S.F., and Keane, R.E. 2001a. Whitebark pine communities: ecology and restoration. Island Press, Washington, DC. 440 p.
- Tomback, D.F. and Linhart, Y.B. 1990. The evolution of bird-dispersed pines. *Evolutionary Ecology* (4): 185-219.
- Tomback, D.F., Sund, S.K, and Hoffmann, L.A. 1993. Post-fire regeneration of *Pinus albicaulis*: height –age relationships, age structure, and microsite characteristics. *Canadian Journal of Forest Research* 23: 113-119.
- Torres, P. and Honrubia, M. 1994. Basidiospore viability in stored slurries. *Mycological Research* 98(5): 527-530.
- Trappe, J.M. 1977. Selection of fungi for ectomycorrhizal inoculation in nurseries. *Annual Review of Phytopathology* 15: 203–222.
- Treu, R. 1990. *Suillus sibiricus*. In: Colour atlas of ectomycorrhizae, plate 47, Agerer, R. (ed.). Einhorn-Verlad, Schwabisch Gmund.
- Trusty, P. 2009. Impact of severe fire on ectomycorrhizal fungi of whitebark pine seedlings. Montana State University Masters Thesis, Bozeman, MT.
- Trusty, P. and Cripps, C.L. 2011. Influence of fire on mycorrhizal colonization of planted and natural whitebark pine seedlings: Ecology and management implications. Pp. 198-202. In: Keane, R.E., Tomback, D.F., Murray, M.P., and Smith, C.M., (eds.). The future of high-elevation, five-needle white pines in Western North America: Proceedings of the High Five Symposium. 28-30 June 2010; Missoula, MT. Proceedings RMRS-P-63. Fort Collins, CO: U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station. 376 p.
- Uchytel, R.J. 1992. *Salix glauca*. In: Fire Effects Information System. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station, Fire Sciences

Laboratory (Producer). [Online] Available: <http://www.fs.fed.us/database/feis/> [2011, July 6].

- Van Den Driessche, R. 1987. Nursery growth of conifer seedlings using fertilizers of different solubilities and application time, and their forest growth. *Canadian Journal of Forest Research* 18: 172-173.
- Van Der Heijden, M.G.A., Klironomos, J.N., Ursic, M., Moutoglis, P., Streitwolf-Engel, R., Boller, T., Wiemken, A. and Sanders, I.R. 1998. Mycorrhizal fungal diversity determines plant biodiversity, ecosystem variability and productivity. *Nature* 396: 69-72.
- Van Der Heijden, M.G.A. 2002. Arbuscular mycorrhizal fungi as a determinant of plant diversity: in search for underlying mechanisms and general principles. Pp. 243-265. In: van der Heijden, M.G.A., Sanders, I.R., (eds.). *Mycorrhizal ecology*. Springer-Verlag, Berlin, Germany.
- Wagner, R.G., Peterson, T.D., Ross, D.W., and Radosevich, S.R. 1989. Competition thresholds for the survival and growth of ponderosa pine seedlings associated with woody and herbaceous vegetation. *New Forests* 3(2): 151-170.
- Wallander, H. and Nylund, J.E. 1991. Effects of excess nitrogen on carbohydrate concentration and mycorrhizae development of *Pinus sylvestris* seedlings. *New Phytologist* 119: 405-441.
- Wan, S., Hui, D., and Luo, Y. 2001. Fire effects on nitrogen pools and dynamics in terrestrial ecosystems : a meta-analysis. *Ecological Applications* 11(5): 1349-1365.
- Weaver, T. and Dale, D. 1974. *Pinus albicaulis* in central Montana: environment, vegetation, and production. *American Midland Naturalist* 37: 395-401.
- Weisleitner, H. 2008. [Personal Communication to Dr. Cathy Cripps]. Federal Nursery of Austria, Innsbruck, Austria.
- Whipps, J.M. 2004. Prospects and limitations for mycorrhizas in biocontrol of root pathogens. *Canadian Journal of Botany* 82: 1198-1227.
- Wiensczyk, G., Durall, D. Jones, M. and Simard, S. 2002. Ectomycorrhizae and forestry in British Columbia: a summary of current research and conservation strategies. Extension Note. *B.C. Journal of Ecosystems and Management* 2(1):1-20.
- Wilkinson, D.M. 2001. Mycorrhizal evolution. *Trend in Ecology and Evolution* 16: 64-65.

[UNEP, WCMC] World Conservation Monitoring Centre. 2011. Protected Areas and World Heritage: Waterton-Glacier International Peace Park Alberta, Canada & Montana, United States of America. 13 p.

APPENDIX A

FIELD STUDY PLOT AND SEEDLING DATA

Description of plots 1-21 at the Summit Lake study area

Plot	Latitude	Longitude	Elevation (m)	Aspect	Slope	Burn Year	# of Clusters	# of Seedlings
1	49.0100	-114.0269	1960.9	SW	5%	2009	15	45
2	49.0103	-114.0272	1959.1	SW	5%	2009	16	48
3	49.0102	-114.0261	1964.2	S-SW	5%	2009	15	45
4	49.0108	-114.0270	1964.0	W	8%	2009	17	51
5	49.0109	-114.0263	1968.1	W	8%	2009	16	48
6	49.0110	-114.0274	1964.5	S-SW	5%	2009	16	48
7	49.0112	-114.0261	1973.1	W	3%	2009	16	48
8	49.0115	-114.0264	1973.5	W	13%	2009	16	47
9	49.0116	-114.0256	1954.7	W-SW	14%	2009	15	45
10	49.0118	-114.0259	1983.2	W-SW	14%	2009	16	48
11	49.0122	-114.0252	1999.1	W	16%	2009	15	45
12	49.0124	-114.0257	1995.2	W	8%	2010	19	57
13	49.0121	-114.0271	1976.7	W	8%	2010	18	53
14	49.0119	-114.0275	1969.2	SW-W	5%	2010	16	48
15	49.0115	-114.0275	1966.3	W	5%	2010	16	48
16	49.0115	-114.0271	1969.0	SW-W	8%	2010	16	48
17	49.0103	-114.0281	1960.2	N	5%	2010	16	48
18	49.0099	-114.0281	1961.8	N	8%	2010	16	48
19	49.0097	-114.0287	1964.5	N-NE	5%	2010	16	48
20	49.0091	-114.0282	1962.5	NE-N	8%	2010	16	48
21	49.0087	-114.0278	1961.8	NE	10%	2010	8	24

Fire Effects Descriptors**Unburned = UB**

- These areas were not burned by prescribed fire

Low Severity = L

- Standing Trees
 - Non lethal fire
 - Lightly scorched
- Surface fuels
 - Unburned (ie. grass, juvenile trees < 2 meters in height and dead/down material)

Moderate Severity = M

- Standing trees
 - Lethal fire – not immediate
 - Moderately scorched
 - Crown fraction burn <50%
- Surface fuels
 - Unburned

High-Low Severity = HL

- Standing trees
 - Lethal fire – immediate
 - Highly scorched
 - Crown fraction burn >50%
- Surface fuels
 - Unburned

High-Moderate Severity = HM

- Standing trees
 - Lethal fire – immediate
 - Highly scorched
 - Crown fraction burn >50%
- Surface fuels
 - Grass <50% burned
 - Juvenile trees (<2 meters) <50% burned
 - Dead/down material - <50% consumption

High-High Severity = HH

- Standing trees
 - Lethal fire – immediate
 - Highly scorched
 - Crown fraction burn >50%
- Surface fuels
 - Grass >50% burned
 - Juvenile trees (<2 meters) >50% burned
 - Dead/down material >50% consumption

Health Index

0 = Seedling dead



1 = Seedling missing



2 = Seedling almost dead, over 50% needle mortality



3 = Seedling has no new growth



4 = Seedling has some new growth



5 = Seedling has lots of new growth and all needles are dark green



Seedling Data**Abbreviations**

S ID = Seedling identification number = plot.cluster/seedling

Dis = Distance from center stake to seedling cluster

Az = Azimuth from center stake to seedling cluster

Burn = Clusters planted in burned areas = 1, clusters planted in un-burned areas = 0

BT = Burn Treatment, abbreviations are listed with the Fire Effects Descriptors

BG = Beargrass, 1=present, 0=absent

CT = Cluster inoculation treatment

I = Inoculated, 1=yes, 0=no

MS = Microsite

YR1 S = Seedling survival at year 1, 1=alive, 0=dead

YR2 H = Health at year 2, definitions are listed in the Health Index

YR2 S = Seedling survival at year 2, 1=alive, 0=dead

YR2 HT = Seedling height at year 2

S ID	Dis	Az	Burn	BT	BG	CT	I	MS	YR1 S	YR2 H	YR2 S	YR2 HT(cm)
1.01a	10	340.00	1	HM	1	T0	0	Snag	1	5	1	16.5
1.01b	10	340.00	1	HM	1	T0	0	Snag	1	4	1	8
1.01c	10	340.00	1	HM	1	T0	0	Snag	1	5	1	12.5
1.02a	10.12	18.00	1	M	0	T0	0	Log	1	4	1	7.5
1.02b	10.12	18.00	1	M	0	T0	0	Log	1	4	1	11
1.02c	10.12	18.00	1	M	0	T0	0	Log	1	4	1	7.2
1.03a	8.74	27.00	1	M	1	T0	0	Snag	1	4	1	4
1.03b	8.74	27.00	1	M	1	T0	0	Snag	1	5	1	10
1.03c	8.74	27.00	1	M	1	T0	0	Snag	1	5	1	7
1.04a	10.56	173.00	0	UB	1	T3	1	Log	1	0	0	na
1.04b	10.56	173.00	0	UB	1	T3	1	Log	1	4	1	12
1.04c	10.56	173.00	0	UB	1	T3	1	Log	1	4	1	13.5
1.05a	13.58	195.00	1	HM	1	T2	0	Log	1	3	1	5.5
1.05b	13.58	195.00	1	HM	1	T2	1	Log	1	5	1	11.5
1.05c	13.58	195.00	1	HM	1	T2	1	Log	1	4	1	8
1.06a	18.65	184.00	1	HH	0	T3	1	Snag	1	4	1	7.5
1.06b	18.65	184.00	1	HH	0	T3	1	Snag	1	5	1	12.5
1.06c	18.65	184.00	1	HH	0	T3	1	Snag	1	5	1	12.5
1.07a	12.6	202.00	1	HH	0	T1	1	Snag	1	5	1	7
1.07b	12.6	202.00	1	HH	0	T1	0	Snag	1	5	1	9
1.07c	12.6	202.00	1	HH	0	T1	0	Snag	1	5	1	9.7
1.08a	3.9	216.00	0	UB	1	T2	1	NA	1	2	1	4
1.08b	3.9	216.00	0	UB	1	T2	1	NA	1	0	0	na
1.08c	3.9	216.00	0	UB	1	T2	0	NA	1	0	0	na
1.09a	18.49	204.00	1	HH	0	T2	1	Log	1	5	1	8
1.09b	18.49	204.00	1	HH	0	T2	1	Log	1	5	1	10
1.09c	18.49	204.00	1	HH	0	T2	0	Log	1	5	1	9
1.10a	19.1	218.00	1	HM	1	T3	1	Snag	1	4	1	8.7
1.10b	19.1	218.00	1	HM	1	T3	1	Snag	1	4	1	7.5
1.10c	19.1	218.00	1	HM	1	T3	1	Snag	1	5	1	10
1.11a	16.52	222.00	1	HH	0	T3	1	Snag	1	5	1	15
1.11b	16.52	222.00	1	HH	0	T3	1	Snag	1	5	1	10.5
1.11c	16.52	222.00	1	HH	0	T3	1	Snag	1	5	1	9
1.12a	7.65	222.00	0	UB	1	T1	0	Log	1	3	1	6.5
1.12b	7.65	222.00	0	UB	1	T1	1	Log	1	4	1	10.2
1.12c	7.65	222.00	0	UB	1	T1	0	Log	1	3	1	8
1.13a	9.72	278.00	1	M	0	T1	1	Snag	1	5	1	9.5
1.13b	9.72	278.00	1	M	0	T1	0	Snag	1	5	1	8.5

1.13c	9.72	278.00	1	M	0	T1	0	Snag	1	2	1	5
1.14a	6.19	306.00	1	HH	0	T2	0	Snag	1	4	1	10
1.14b	6.19	306.00	1	HH	0	T2	1	Snag	1	3	1	7
1.14c	6.19	306.00	1	HH	0	T2	1	Snag	1	2	1	7
1.15a	7.5	325.00	1	HM	0	T1	1	Log	1	3	1	5
1.15b	7.5	325.00	1	HM	0	T1	0	Log	1	4	1	8
1.15c	7.5	325.00	1	HM	0	T1	0	Log	1	0	0	na
2.01a	7.10	187.00	0	UB	1	T2	1	NA	1	2	1	5.5
2.01b	7.10	187.00	0	UB	1	T2	1	NA	1	0	0	na
2.01c	7.10	187.00	0	UB	1	T2	0	NA	1	0	0	na
2.02a	8.20	210.00	1	HH	0	T1	0	Snag	1	4	1	7.5
2.02b	8.20	210.00	1	HH	0	T1	0	Snag	1	4	1	9.5
2.02c	8.20	210.00	1	HH	0	T1	1	Snag	1	5	1	11.4
2.03a	5.15	212.00	0	UB	0	T3	1	?	0	0	0	na
2.03b	5.15	212.00	0	UB	0	T3	1	?	0	0	0	na
2.03c	5.15	212.00	0	UB	0	T3	1	?	0	0	0	na
2.04a	9.32	223.00	1	HH	0	T0	0	Snag	1	5	1	16.5
2.04b	9.32	223.00	1	HH	0	T0	0	Snag	1	5	1	9.5
2.04c	9.32	223.00	1	HH	0	T0	0	Snag	1	5	1	9.3
2.05a	9.47	314.00	1	L	0	T3	1	NA	1	0	0	na
2.05b	9.47	314.00	1	L	0	T3	1	NA	1	0	0	na
2.05c	9.47	314.00	1	L	0	T3	1	NA	1	2	1	4
2.06a	12.34	310.00	0	UB	1	T0	0	NA	1	3	1	7.5
2.06b	12.34	310.00	0	UB	1	T0	0	NA	1	2	1	6
2.06c	12.34	310.00	0	UB	1	T0	0	NA	0	0	0	na
2.07a	13.80	321.00	1	HM	0	T2	1	Log	1	3	1	7
2.07b	13.80	321.00	1	HM	0	T2	1	Log	1	5	1	12.5
2.07c	13.80	321.00	1	HM	0	T2	0	Log	1	5	1	12
2.08a	10.92	337.00	1	HM	1	T1	0	NA	1	3	1	5.5
2.08b	10.92	337.00	1	HM	1	T1	1	NA	0	0	0	na
2.08c	10.92	337.00	1	HM	1	T1	0	NA	0	0	0	na
2.09a	15.95	344.00	1	HM	1	T3	1	NA	1	0	0	na
2.09b	15.95	344.00	1	HM	1	T3	1	NA	1	0	0	na
2.09c	15.95	344.00	1	HM	1	T3	1	NA	1	0	0	na
2.10a	22.50	345.00	0	UB	0	T0	0	NA	1	3	1	6
2.10b	22.50	345.00	0	UB	0	T0	0	NA	1	0	0	na
2.10c	22.50	345.00	0	UB	0	T0	0	NA	1	0	0	na
2.11a	18.70	349.00	0	UB	1	T1	1	Log	1	3	1	5
2.11b	18.70	349.00	0	UB	1	T1	0	Log	1	4	1	8
2.11c	18.70	349.00	0	UB	1	T1	0	Log	1	3	1	7

2.12a	16.49	356.00	0	UB	0	T2	1	NA	1	3	1	9
2.12b	16.49	356.00	0	UB	0	T2	1	NA	1	0	0	na
2.12c	16.49	356.00	0	UB	0	T2	0	NA	1	0	0	na
2.13a	16.35	18.00	0	UB	1	T1	0	NA	1	2	1	4
2.13b	16.35	18.00	0	UB	1	T1	1	NA	1	0	0	na
2.13c	16.35	18.00	0	UB	1	T1	0	NA	1	0	0	na
2.14a	12.90	19.00	0	UB	0	T3	1	NA	1	0	0	na
2.14b	12.90	19.00	0	UB	0	T3	1	NA	1	0	0	na
2.14c	12.90	19.00	0	UB	0	T3	1	NA	0	0	0	na
2.15a	17.59	32.00	0	UB	0	T2	1	NA	0	0	0	na
2.15b	17.59	32.00	0	UB	0	T2	0	NA	1	0	0	na
2.15c	17.59	32.00	0	UB	0	T2	1	NA	1	0	0	na
2.16a	14.40	43.00	0	UB	0	T0	0	NA	1	0	0	na
2.16b	14.40	43.00	0	UB	0	T0	0	NA	0	0	0	na
2.16c	14.40	43.00	0	UB	0	T0	0	NA	1	0	0	na
3.01a	4.00	179.00	0	UB	0	T3	1	NA	1	3	1	7
3.01b	4.00	179.00	0	UB	0	T3	1	NA	1	0	0	na
3.01c	4.00	179.00	0	UB	0	T3	1	NA	1	2	1	7.5
3.02a	16.40	179.00	1	HH	0	T0	0	Log	1	4	1	10.5
3.02b	16.40	179.00	1	HH	0	T0	0	Log	1	3	1	8
3.02c	16.40	179.00	1	HH	0	T0	0	Log	1	4	1	5
3.03a	19.50	200.00	1	HH	0	T3	1	Rock	1	3	1	6.5
3.03b	19.50	200.00	1	HH	0	T3	1	Rock	1	3	1	8.5
3.03c	19.50	200.00	1	HH	0	T3	1	Rock	1	3	1	6.5
3.04a	17.83	211.00	1	HM	0	T1	0	Log	1	2	1	5.5
3.04b	17.83	211.00	1	HM	0	T1	1	Log	1	0	0	na
3.04c	17.83	211.00	1	HM	0	T1	0	Log	1	0	0	na
3.05a	11.50	273.00	1	HH	0	T2	1	Log	1	0	0	na
3.05b	11.50	273.00	1	HH	0	T2	0	Log	1	5	1	7
3.05c	11.50	273.00	1	HH	0	T2	1	Log	1	0	0	na
3.06a	5.75	329.00	0	UB	1	T2	1	Log	1	3	1	7
3.06b	5.75	329.00	0	UB	1	T2	0	Log	1	3	1	6.5
3.06c	5.75	329.00	0	UB	1	T2	1	Log	1	3	1	6
3.07a	7.11	346.00	0	UB	0	T1	1	Log	1	0	0	na
3.07b	7.11	346.00	0	UB	0	T1	0	Log	1	3	1	8
3.07c	7.11	346.00	0	UB	0	T1	0	Log	1	2	1	5.5
3.08a	4.83	7.00	1	HM	1	T3	1	NA	1	4	1	8.5
3.08b	4.83	7.00	1	HM	1	T3	1	NA	1	4	1	10.5
3.08c	4.83	7.00	1	HM	1	T3	1	NA	1	3	1	6.5
3.09a	8.80	39.00	0	UB	0	T0	0	Log	1	0	0	na

3.09b	8.80	39.00	0	UB	0	T0	0	Log	1	0	0	na
3.09c	8.80	39.00	0	UB	0	T0	0	Log	1	0	0	na
3.10a	12.65	40.00	1	HM	1	T2	1	Snag	1	4	1	12.5
3.10b	12.65	40.00	1	HM	1	T2	0	Snag	1	4	1	8.5
3.10c	12.65	40.00	1	HM	1	T2	1	Snag	1	0	0	na
3.11a	9.00	86.00	1	M	1	T3	1	Log	1	0	0	na
3.11b	9.00	86.00	1	M	1	T3	1	Log	1	3	1	7
3.11c	9.00	86.00	1	M	1	T3	1	Log	1	3	1	6.5
3.12a	15.50	87.00	1	M	1	T2	1	Log	1	5	1	10
3.12b	15.50	87.00	1	M	1	T2	0	Log	1	5	1	13.5
3.12c	15.50	87.00	1	M	1	T2	1	Log	1	5	1	10.5
3.13a	28.50	93.00	0	UB	1	T0	0	Log	1	2	1	5
3.13b	28.50	93.00	0	UB	1	T0	0	Log	1	3	1	7
3.13c	28.50	93.00	0	UB	1	T0	0	Log	1	0	0	na
3.14a	17.15	131.00	1	HL	0	T1	1	Log	1	0	0	na
3.14b	17.15	131.00	1	HL	0	T1	0	Log	1	0	0	na
3.14c	17.15	131.00	1	HL	0	T1	0	Log	1	0	0	na
3.15a	14.15	150.00	1	HM	1	T1	0	Log	1	3	1	7
3.15b	14.15	150.00	1	HM	1	T1	1	Log	1	3	1	6
3.15c	14.15	150.00	1	HM	1	T1	0	Log	1	4	1	6
4.01a	5.00	338.00	0	UB	1	T2	0	Log	1	4	1	7
4.01b	5.00	338.00	0	UB	1	T2	1	Log	1	3	1	6
4.01c	5.00	338.00	0	UB	1	T2	1	Log	1	3	1	7
4.02a	10.69	345.00	0	UB	1	T1	0	Rock	1	0	0	na
4.02b	10.69	345.00	0	UB	1	T1	1	Rock	1	0	0	na
4.02c	10.69	345.00	0	UB	1	T1	0	Rock	1	3	1	8
4.03a	14.30	10.00	1	HH	0	T0	0	Stump	1	5	1	9
4.03b	14.30	10.00	1	HH	0	T0	0	Stump	1	5	1	8.5
4.03c	14.30	10.00	1	HH	0	T0	0	Stump	1	3	1	4
4.04a	13.00	67.00	1	HH	0	T1	1	Snag	1	5	1	7.5
4.04b	13.00	67.00	1	HH	0	T1	0	Snag	1	2	1	2
4.04c	13.00	67.00	1	HH	0	T1	0	Snag	1	4	1	6
4.05a	8.30	118.00	1	HH	0	T2	0	Log	1	4	1	8
4.05b	8.30	118.00	1	HH	0	T2	1	Log	1	5	1	10
4.05c	8.30	118.00	1	HH	0	T2	1	Log	1	5	1	10
4.06a	8.25	132.00	1	HH	0	T0	0	NA	1	5	1	9.5
4.06b	8.25	132.00	1	HH	0	T0	0	NA	1	5	1	8
4.06c	8.25	132.00	1	HH	0	T0	0	NA	1	5	1	8
4.07a	14.20	162.00	0	UB	1	T1	1	NA	1	0	0	na
4.07b	14.20	162.00	0	UB	1	T1	0	NA	1	0	0	na

4.07c	14.20	162.00	0	UB	1	T1	0	NA	1	0	0	na
4.08a	10.50	178.00	0	UB	1	T1	0	Log	1	2	1	3
4.08b	10.50	178.00	0	UB	1	T1	1	Log	1	0	0	na
4.08c	10.50	178.00	0	UB	1	T1	0	Log	1	0	0	na
4.09a	7.58	194.00	1	HM	0	T2	0	Snag	1	5	1	14.5
4.09b	7.58	194.00	1	HM	0	T2	1	Snag	1	5	1	12
4.09c	7.58	194.00	1	HM	0	T2	1	Snag	1	5	1	10.5
4.10a	9.15	249.00	1	HH	0	T2	1	Log/snag	1	5	1	10.5
4.10b	9.15	249.00	1	HH	0	T2	0	Log/snag	1	5	1	10.5
4.10c	9.15	249.00	1	HH	0	T2	1	Log/snag	1	4	1	7
4.11a	13.40	254.00	1	M	0	T3	1	Log	1	3	1	9
4.11b	13.40	254.00	1	M	0	T3	1	Log	1	4	1	10
4.11c	13.40	254.00	1	M	0	T3	1	Log	1	3	1	5.5
4.12a	14.15	264.00	1	M	0	T2	1	Log	1	4	1	9.5
4.12b	14.15	264.00	1	M	0	T2	0	Log	1	0	0	na
4.12c	14.15	264.00	1	M	0	T2	1	Log	1	4	1	8
4.13a	3.59	272.00	0	UB	0	T3	1	NA	1	4	1	10.5
4.13b	3.59	272.00	0	UB	0	T3	1	NA	1	4	1	6
4.13c	3.59	272.00	0	UB	0	T3	1	NA	1	4	1	6
4.14a	4.95	276.00	0	UB	0	T2	1	Log	1	0	0	na
4.14b	4.95	276.00	0	UB	0	T2	1	Log	1	0	0	na
4.14c	4.95	276.00	0	UB	0	T2	0	Log	1	4	1	14
4.15a	8.41	283.00	1	HM	0	T3	1	Log	1	3	1	8
4.15b	8.41	283.00	1	HM	0	T3	1	Log	1	2	1	5
4.15c	8.41	283.00	1	HM	0	T3	1	Log	1	5	1	10
4.16a	7.22	306.00	0	UB	1	T0	0	NA	1	2	1	5
4.16b	7.22	306.00	0	UB	1	T0	0	NA	1	4	1	5
4.16c	7.22	306.00	0	UB	1	T0	0	NA	1	0	0	na
4.17a	16.50	314.00	1	HH	0	T2	1	Log/stump	1	2	1	4
4.17b	16.50	314.00	1	HH	0	T2	0	Log/stump	1	0	0	na
4.17c	16.50	314.00	1	HH	0	T2	1	Log/stump	1	0	0	na
5.01a	5.40	31.00	1	HH	0	T3	1	ROCK	1	5	1	12
5.01b	5.40	31.00	1	HH	0	T3	1	ROCK	1	5	1	16
5.01c	5.40	31.00	1	HH	0	T3	1	ROCK	1	3	1	8
5.02a	14.60	93.00	1	HM	0	T3	1	ROCK/STUMP	1	4	1	12
5.02b	14.60	93.00	1	HM	0	T3	1	ROCK/STUMP	1	4	1	10.5
5.02c	14.60	93.00	1	HM	0	T3	1	ROCK/STUMP	1	4	1	9.5
5.03a	11.15	113.00	0	UB	0	T1	1	ROCK	1	4	1	7
5.03b	11.15	113.00	0	UB	0	T1	0	ROCK	1	5	1	9
5.03c	11.15	113.00	0	UB	0	T1	0	ROCK	1	5	1	11

5.04a	15.50	120.00	1	HL	1	T2	0	ROCK	1	5	1	13
5.04b	15.50	120.00	1	HL	1	T2	1	ROCK	1	4	1	10.5
5.04c	15.50	120.00	1	HL	1	T2	1	ROCK	1	4	1	7.5
5.05a	21.24	139.00	1	HL	1	T1	1	ROCK	0	0	0	na
5.05b	21.24	139.00	1	HL	1	T1	0	ROCK	1	4	1	6.5
5.05c	21.24	139.00	1	HL	1	T1	0	ROCK	1	3	1	9.5
5.06a	2.42	189.00	0	UB	0	T3	1	ROCK	1	0	0	na
5.06b	2.42	189.00	0	UB	0	T3	1	ROCK	1	2	1	6.5
5.06c	2.42	189.00	0	UB	0	T3	1	ROCK	1	3	1	8
5.07a	9.00	189.00	1	HM	0	T1	0	ROCK	1	0	0	na
5.07b	9.00	189.00	1	HM	0	T1	1	ROCK	1	0	0	na
5.07c	9.00	189.00	1	HM	0	T1	0	ROCK	1	0	0	na
5.08a	16.25	189.00	1	HH	0	T2	0	ROCK	1	0	0	na
5.08b	16.25	189.00	1	HH	0	T2	1	ROCK	1	3	1	8.5
5.08c	16.25	189.00	1	HH	0	T2	1	ROCK	1	0	0	na
5.09a	10.80	206.00	0	UB	0	T0	0	ROCK	1	3	1	6.5
5.09b	10.80	206.00	0	UB	0	T0	0	ROCK	1	3	1	7
5.09c	10.80	206.00	0	UB	0	T0	0	ROCK	1	4	1	11.5
5.10a	22.60	220.00	1	HM	1	T0	0	LOG	1	4	1	10.5
5.10b	22.60	220.00	1	HM	1	T0	0	LOG	1	4	1	11
5.10c	22.60	220.00	1	HM	1	T0	0	LOG	1	4	1	9
5.11a	15.26	240.00	1	HH	0	T1	0	LOG	1	4	1	10
5.11b	15.26	240.00	1	HH	0	T1	1	LOG	1	5	1	14
5.11c	15.26	240.00	1	HH	0	T1	0	LOG	1	5	1	11.5
5.12a	14.55	253.00	1	HM	0	T2	1	LOG	1	3	1	5
5.12b	14.55	253.00	1	HM	0	T2	1	LOG	1	3	1	7
5.12c	14.55	253.00	1	HM	0	T2	0	LOG	1	1	0	na
5.13a	19.05	335.00	1	HH	0	T0	0	ROCK/LOG	1	5	1	10
5.13b	19.05	335.00	1	HH	0	T0	0	ROCK/LOG	1	5	1	10
5.13c	19.05	335.00	1	HH	0	T0	0	ROCK/LOG	1	5	1	6.5
5.14a	17.77	344.00	1	HH	0	T3	1	ROCK	1	5	1	10
5.14b	17.77	344.00	1	HH	0	T3	1	ROCK	1	0	0	na
5.14c	17.77	344.00	1	HH	0	T3	1	ROCK	1	4	1	10.5
5.15a	25.60	125.00	0	UB	1	T0	0	ROCK	1	3	1	10
5.15b	25.60	125.00	0	UB	1	T0	0	ROCK	1	3	1	10
5.15c	25.60	125.00	0	UB	1	T0	0	ROCK	1	3	1	7
5.16a	14.10	176.00	0	UB	1	T2	1	ROCK	1	3	1	7
5.16b	14.10	176.00	0	UB	1	T2	1	ROCK	1	4	1	8
5.16c	14.10	176.00	0	UB	1	T2	0	ROCK	1	4	1	7.5
6.01a	4.90	342.00	1	HM	0	T2	0	STUMP	1	2	1	3

6.01b	4.90	342.00	1	HM	0	T2	0	STUMP	1	2	1	3
6.01c	4.90	342.00	1	HM	0	T2	0	STUMP	1	3	1	4.5
6.02a	14.60	356.00	1	HM	0	T1	0	LOG	1	5	1	12
6.02b	14.60	356.00	1	HM	0	T1	0	LOG	1	5	1	10.5
6.02c	14.60	356.00	1	HM	0	T1	0	LOG	1	5	1	11
6.03a	19.60	30.00	0	UB	0	T0	0	LOG	1	0	0	na
6.03b	19.60	30.00	0	UB	0	T0	0	LOG	1	4	1	11
6.03c	19.60	30.00	0	UB	0	T0	0	LOG	1	4	1	9
6.04a	15.50	43.00	1	HL	0	T1	0	LOG	1	4	1	9
6.04b	15.50	43.00	1	HL	0	T1	1	LOG	1	4	1	10
6.04c	15.50	43.00	1	HL	0	T1	0	LOG	1	4	1	10.5
6.05a	8.70	66.00	1	L	1	T1	1	LOG	1	0	0	na
6.05b	8.70	66.00	1	L	1	T1	0	LOG	1	0	0	na
6.05c	8.70	66.00	1	L	1	T1	0	LOG	1	0	0	na
6.06a	16.00	108.00	1	HL	1	T0	0	LOG	1	3	1	7.5
6.06b	16.00	108.00	1	HL	1	T0	0	LOG	1	0	0	na
6.06c	16.00	108.00	1	HL	1	T0	0	LOG	1	2	1	6.5
6.10a	21.80	152.00	1	HL	1	T2	1	STUMP	1	4	1	9
6.10b	21.80	152.00	1	HL	1	T2	0	STUMP	1	0	0	na
6.10c	21.80	152.00	1	HL	1	T2	1	STUMP	1	3	1	7
6.11a	13.30	152.00	0	UB	1	T2	1	LOG	1	0	0	na
6.11b	13.30	152.00	0	UB	1	T2	1	LOG	1	3	1	5.5
6.11c	13.30	152.00	0	UB	1	T2	0	LOG	1	2	1	4
6.12a	11.90	194.00	1	HM	0	T3	1	LOG	1	4	1	7
6.12b	11.90	194.00	1	HM	0	T3	1	LOG	1	5	1	8
6.12c	11.90	194.00	1	HM	0	T3	1	LOG	1	0	0	na
6.13a	14.00	204.00	1	M	0	T3	1	STUMP	1	3	1	5
6.13b	14.00	204.00	1	M	0	T3	1	STUMP	1	5	1	10
6.13c	14.00	204.00	1	M	0	T3	1	STUMP	1	0	0	na
6.14a	10.30	231.00	1	HL	0	T1	1	STUMP	1	5	1	7
6.14b	10.30	231.00	1	HL	0	T1	0	STUMP	1	4	1	9
6.14c	10.30	231.00	1	HL	0	T1	0	STUMP	1	5	1	9.5
6.15a	10.20	265.00	1	HM	0	T0	0	STUMP	1	5	1	15.5
6.15b	10.20	265.00	1	HM	0	T0	0	STUMP	1	3	1	9
6.15c	10.20	265.00	1	HM	0	T0	0	STUMP	1	0	0	na
6.16a	12.40	277.00	1	HL	0	T0	0	LOG	1	5	1	12
6.16b	12.40	277.00	1	HL	0	T0	0	LOG	1	5	1	13
6.16c	12.40	277.00	1	HL	0	T0	0	LOG	1	5	1	14
7.01a	8.70	277.00	1	HL	1	T1	0	LOG	1	3	1	6
7.01b	8.70	277.00	1	HL	1	T1	0	LOG	1	4	1	9

7.01c	8.70	277.00	1	HL	1	T1	1	LOG	1	3	1	8.5
7.02a	14.40	4.00	1	HL	1	T3	1	SNAG	1	5	1	9
7.02b	14.40	4.00	1	HL	1	T3	1	SNAG	1	5	1	8.5
7.02c	14.40	4.00	1	HL	1	T3	1	SNAG	1	5	1	10.5
7.03a	4.60	79.00	1	L	1	T3	1	LOG	1	0	0	na
7.03b	4.60	79.00	1	L	1	T3	1	LOG	1	0	0	na
7.03c	4.60	79.00	1	L	1	T3	1	LOG	1	0	0	na
7.04a	10.00	93.00	1	HH	0	T0	0	STUMP	1	5	1	13
7.04b	10.00	93.00	1	HH	0	T0	0	STUMP	1	5	1	11
7.04c	10.00	93.00	1	HH	0	T0	0	STUMP	1	5	1	11
7.05a	9.70	107.00	1	HL	0	T0	0	LOG	1	4	1	9
7.05b	9.70	107.00	1	HL	0	T0	0	LOG	1	3	1	7.5
7.05c	9.70	107.00	1	HL	0	T0	0	LOG	1	5	1	10.5
7.06a	13.10	111.00	1	HL	1	T3	1	LOG	1	4	1	7
7.06b	13.10	111.00	1	HL	1	T3	1	LOG	1	5	1	16
7.06c	13.10	111.00	1	HL	1	T3	1	LOG	1	5	1	11.5
7.07a	11.10	162.00	1	HL	0	T2	1	SNAG	1	5	1	16
7.07b	11.10	162.00	1	HL	0	T2	1	SNAG	1	5	1	13
7.07c	11.10	162.00	1	HL	0	T2	0	SNAG	1	4	1	10.5
7.08a	4.50	162.00	0	UB	1	T2	1	NA	1	3	1	9
7.08b	4.50	162.00	0	UB	1	T2	1	NA	1	3	1	10
7.08c	4.50	162.00	0	UB	1	T2	0	NA	1	3	1	10
7.09a	10.60	168.00	1	HL	0	T0	0	STUMP	1	4	1	11
7.09b	10.60	168.00	1	HL	0	T0	0	STUMP	1	4	1	7
7.09c	10.60	168.00	1	HL	0	T0	0	STUMP	1	4	1	8
7.10a	11.50	168.00	1	HL	0	T3	1	SNAG	1	5	1	14
7.10b	11.50	168.00	1	HL	0	T3	1	SNAG	1	5	1	11
7.10c	11.50	168.00	1	HL	0	T3	1	SNAG	1	5	1	10
7.11a	3.60	197.00	0	UB	1	T1	0	NA	1	3	1	4
7.11b	3.60	197.00	0	UB	1	T1	1	NA	1	3	1	7
7.11c	3.60	197.00	0	UB	1	T1	0	NA	1	3	1	8
7.12a	7.10	222.00	0	UB	1	T0	0	NA	1	0	0	na
7.12b	7.10	222.00	0	UB	1	T0	0	NA	1	0	0	na
7.12c	7.10	222.00	0	UB	1	T0	0	NA	1	0	0	na
7.13a	11.00	215.00	1	HL	0	T2	1	LOG	1	4	1	7.5
7.13b	11.00	215.00	1	HL	0	T2	0	LOG	1	5	1	10
7.13c	11.00	215.00	1	HL	0	T2	1	LOG	1	5	1	7.5
7.14a	8.50	254.00	1	HL	1	T1	1	NA	1	4	1	9
7.14b	8.50	254.00	1	HL	1	T1	0	NA	1	0	0	na
7.14c	8.50	254.00	1	HL	1	T1	0	NA	1	4	1	11

7.15a	14.90	190.00	1	HM	1	T0	0	NA	1	2	1	5
7.15b	14.90	190.00	1	HM	1	T0	0	NA	1	0	0	na
7.15c	14.90	190.00	1	HM	1	T0	0	NA	1	3	1	8
7.16a	14.20	254.00	1	HL	1	T2	1	NA	1	0	0	na
7.16b	14.20	254.00	1	HL	1	T2	1	NA	1	0	0	na
7.16c	14.20	254.00	1	HL	1	T2	0	NA	1	0	0	na
8.01a	11.20	210.00	1	HL	0	T2	1	SNAG	1	5	1	11
8.01b	11.20	210.00	1	HL	0	T2	1	SNAG	1	5	1	10
8.01c	11.20	210.00	1	HL	0	T2	0	SNAG	1	4	1	7
8.02a	8.00	250.00	1	HL	1	T2	1	SNAG	1	1	0	na
8.02b	8.00	250.00	1	HL	1	T2	0	SNAG	1	1	0	na
8.02c	8.00	250.00	1	HL	1	T2	1	SNAG	1	1	0	na
8.03a	10.90	327.00	1	HL	1	T0	0	SNAG	0	4	1	6
8.03b	10.90	327.00	1	HL	1	T0	0	SNAG	1	5	1	10.5
8.03c	10.90	327.00	1	HL	1	T0	0	SNAG	1	4	1	7
8.04a	13.80	14.00	1	M	0	T0	0	LOG	1	3	1	7.5
8.04b	13.80	14.00	1	M	0	T0	0	LOG	1	3	1	5.5
8.04c	13.80	14.00	1	M	0	T0	0	LOG	1	0	0	na
8.05a	5.90	68.00	1	HL	1	T1	0	STUMP	1	5	1	9.5
8.05b	5.90	68.00	1	HL	1	T1	0	STUMP	1	0	0	na
8.05c	5.90	68.00	1	HL	1	T1	1	STUMP	1	0	0	na
8.06a	9.20	105.00	1	HL	0	T3	1	LOG	1	2	1	4
8.06b	9.20	105.00	1	HL	0	T3	1	LOG	1	4	1	5
8.06c	9.20	105.00	1	HL	0	T3	1	LOG	1	5	1	7.5
8.07a	9.00	115.00	1	HL	1	T2	1	LOG	1	2	1	6.5
8.07b	9.00	115.00	1	HL	1	T2	0	LOG	1	3	1	8
8.07c	9.00	115.00	1	HL	1	T2	1	LOG	1	4	1	8.5
8.08a	4.20	125.00	0	UB	1	T1	0	NA	1	2	1	6
8.08b	4.20	125.00	0	UB	1	T1	1	NA	1	0	0	na
8.08c	4.20	125.00	0	UB	1	T1	0	NA	1	0	0	na
8.09a	13.60	145.00	1	M	1	T0	0	NA	1	0	0	na
8.09b	13.60	145.00	1	M	1	T0	0	NA	1	0	0	na
8.09c	13.60	145.00	1	M	1	T0	0	NA	1	0	0	na
8.10a	9.40	172.00	1	HL	0	T3	1	SNAG	1	3	1	5.5
8.10b	9.40	172.00	1	HL	0	T3	1	SNAG	1	4	1	10
8.10c	9.40	172.00	1	HL	0	T3	1	SNAG	1	2	1	6.5
8.11a	11.60	172.00	1	HL	1	T0	0	SNAG	1	3	1	7.5
8.11b	11.60	172.00	1	HL	1	T0	0	SNAG	1	3	1	7.5
8.11c	11.60	172.00	1	HL	1	T0	0	SNAG	1	2	1	7
8.12a	11.90	183.00	1	HL	1	T3	1	SNAG	1	4	1	9.7

8.12b	11.90	183.00	1	HL	1	T3	1	SNAG	1	4	1	5
8.12c	11.90	183.00	1	HL	1	T3	1	SNAG	0	5	1	12
8.13a	17.70	196.00	1	HL	1	T2	1	SNAG	1	2	1	5
8.13b	17.70	196.00	1	HL	1	T2	1	SNAG	1	2	1	4
8.13c	17.70	196.00	1	HL	1	T2	0	SNAG	1	2	1	4
8.14a	12.00	217.00	1	HL	1	T1	0	SNAG	1	4	1	7.3
8.14b	12.00	217.00	1	HL	1	T1	1	SNAG	1	4	1	8.5
8.14c	12.00	217.00	1	HL	1	T1	0	SNAG	1	4	1	10
8.15a	12.60	212.00	1	HL	1	T3	1	LOG	1	5	1	14
8.15b	12.60	212.00	1	HL	1	T3	1	LOG	1	3	1	7.5
8.15c	12.60	212.00	1	HL	1	T3	1	LOG	1	5	1	10.5
8.16a	11.30	321.00	1	HL	0	T1	0	STUMP	1	4	1	6.3
8.16b	11.30	321.00	1	HL	0	T1	0	STUMP	1	3	1	7
8.16c	11.30	321.00	1	HL	0	T1	1	STUMP	1	5	1	8
9.01a	9.90	322.00	1	L	1	T2	1	ROCK	1	3	1	6
9.01b	9.90	322.00	1	L	1	T2	0	ROCK	1	0	0	na
9.01c	9.90	322.00	1	L	1	T2	1	ROCK	1	1	0	na
9.02a	15.20	38.00	1	HM	0	T1	0	LOG	1	5	1	11.5
9.02b	15.20	38.00	1	HM	0	T1	1	LOG	1	5	1	9.5
9.02c	15.20	38.00	1	HM	0	T1	0	LOG	1	4	1	7.5
9.03a	19.30	41.00	1	HH	0	T0	0	STUMP	1	4	1	10
9.03b	19.30	41.00	1	HH	0	T0	0	STUMP	1	4	1	9
9.03c	19.30	41.00	1	HH	0	T0	0	STUMP	1	4	1	6.5
9.04a	9.20	54.00	1	HM	0	T2	0	LOG	1	5	1	7
9.04b	9.20	54.00	1	HM	0	T2	1	LOG	1	5	1	8.3
9.04c	9.20	54.00	1	HM	0	T2	1	LOG	1	5	1	8.5
9.05a	16.90	108.00	1	HM	0	T0	0	ROCK	1	0	0	na
9.05b	16.90	108.00	1	HM	0	T0	0	ROCK	1	5	1	13.7
9.05c	16.90	108.00	1	HM	0	T0	0	ROCK	1	5	1	19
9.06a	8.10	106.00	1	HL	0	T3	1	STUMP	1	0	0	na
9.06b	8.10	106.00	1	HL	0	T3	1	STUMP	1	0	0	na
9.06c	8.10	106.00	1	HL	0	T3	1	STUMP	1	4	1	6.5
9.07a	12.50	144.00	1	HM	1	T1	0	LOG	1	0	0	na
9.07b	12.50	144.00	1	HM	1	T1	1	LOG	1	0	0	na
9.07c	12.50	144.00	1	HM	1	T1	0	LOG	1	0	0	na
9.08a	9.80	171.00	1	HM	0	T2	1	STUMP	1	2	1	5.5
9.08b	9.80	171.00	1	HM	0	T2	0	STUMP	1	5	1	21
9.08c	9.80	171.00	1	HM	0	T2	1	STUMP	1	5	1	13
9.09a	13.80	184.00	1	UB	0	T3	1	NA	1	4	1	10
9.09b	13.80	184.00	1	UB	0	T3	1	NA	1	4	1	11.5

9.09c	13.80	184.00	1	UB	0	T3	1	NA	1	0	0	na
9.10a	9.80	207.00	0	UB	0	T1	0	LOG	0	2	1	4
9.10b	9.80	207.00	0	UB	0	T1	1	LOG	1	4	1	9.5
9.10c	9.80	207.00	0	UB	0	T1	0	LOG	0	0	0	na
9.11a	6.10	212.00	0	UB	1	T0	0	STUMP	1	4	1	10
9.11b	6.10	212.00	0	UB	1	T0	0	STUMP	1	4	1	11.5
9.11c	6.10	212.00	0	UB	1	T0	0	STUMP	1	0	0	na
9.12a	14.90	236.00	0	HM	1	T0	0	STUMP	1	5	1	11.5
9.12b	14.90	236.00	0	HM	1	T0	0	STUMP	1	5	1	12.5
9.12c	14.90	236.00	0	HM	1	T0	0	STUMP	1	1	0	na
9.13a	17.20	251.00	1	HM	0	T1	0	SNAG	1	4	1	12
9.13b	17.20	251.00	1	HM	0	T1	0	SNAG	1	4	1	9.5
9.13c	17.20	251.00	1	HM	0	T1	1	SNAG	1	4	1	12.5
9.14a	6.40	257.00	1	HM	1	T3	1	SNAG	1	5	1	9.5
9.14b	6.40	257.00	1	HM	1	T3	1	SNAG	1	5	1	12.5
9.14c	6.40	257.00	1	HM	1	T3	1	SNAG	1	5	1	14
9.15a	14.00	291.00	1	HL	1	T3	1	SNAG	1	5	1	11
9.15b	14.00	291.00	1	HL	1	T3	1	SNAG	1	5	1	9
9.15c	14.00	291.00	1	HL	1	T3	1	SNAG	1	5	1	10.5
9.16a	11.00	170.00	1	HH	1	T2	1	LOG	1	3	1	7
9.16b	11.00	170.00	1	HH	1	T2	0	LOG	1	3	1	9.5
9.16c	11.00	170.00	1	HH	1	T2	1	LOG	1	4	1	10.5
10.01a	5.90	170.00	1	M	1	T0	0	STUMP	1	4	1	8
10.01b	5.90	170.00	1	M	1	T0	0	STUMP	1	0	0	na
10.01c	5.90	170.00	1	M	1	T0	0	STUMP	1	4	1	7
10.02a	17.10	207.00	1	HL	0	T0	0	SNAG	1	5	1	9.7
10.02b	17.10	207.00	1	HL	0	T0	0	SNAG	1	5	1	10
10.02c	17.10	207.00	1	HL	0	T0	0	SNAG	1	5	1	9.5
10.03a	16.00	225.00	1	M	1	T0	0	STUMP	1	5	1	11
10.03b	16.00	225.00	1	M	1	T0	0	STUMP	1	5	1	8.5
10.03c	16.00	225.00	1	M	1	T0	0	STUMP	1	5	1	9
10.04a	8.70	279.00	1	HL	1	T3	1	LOG	1	4	1	9.5
10.04b	8.70	279.00	1	HL	1	T3	1	LOG	1	4	1	10
10.04c	8.70	279.00	1	HL	1	T3	1	LOG	1	4	1	9.5
10.05a	11.70	288.00	1	HM	1	T2	1	LOG	1	5	1	15
10.05b	11.70	288.00	1	HM	1	T2	1	LOG	1	5	1	13.3
10.05c	11.70	288.00	1	HM	1	T2	0	LOG	1	5	1	10
10.06a	15.50	294.00	1	HL	0	T3	1	LOG	1	5	1	11.5
10.06b	15.50	294.00	1	HL	0	T3	1	LOG	1	3	1	7.3
10.06c	15.50	294.00	1	HL	0	T3	1	LOG	1	4	1	11.5

10.07a	12.60	301.00	1	HM	0	T3	1	ROCK	1	0	0	na
10.07b	12.60	301.00	1	HM	0	T3	1	ROCK	1	2	1	6.5
10.07c	12.60	301.00	1	HM	0	T3	1	ROCK	1	0	0	na
10.08a	1.00	301.00	0	UB	1	T2	1	ROCK	1	2	1	7.5
10.08b	1.00	301.00	0	UB	1	T2	0	ROCK	1	4	1	5.3
10.08c	1.00	301.00	0	UB	1	T2	1	ROCK	1	0	0	na
10.09a	8.70	353.00	1	HM	0	T1	0	SNAG	1	4	1	8
10.09b	8.70	353.00	1	HM	0	T1	1	SNAG	1	4	1	7.3
10.09c	8.70	353.00	1	HM	0	T1	0	SNAG	1	3	1	5
10.10a	9.90	358.00	1	HM	1	T1	1	LOG	0	5	1	9.5
10.10b	9.90	358.00	1	HM	1	T1	0	LOG	1	4	1	4
10.10c	9.90	358.00	1	HM	1	T1	0	LOG	1	3	1	3
10.11a	10.40	22.00	1	HM	0	T1	1	STUMP	1	4	1	8.5
10.11b	10.40	22.00	1	HM	0	T1	0	STUMP	1	0	0	na
10.11c	10.40	22.00	1	HM	0	T1	0	STUMP	1	3	1	7.3
10.12a	9.50	49.00	1	M	1	T1	0	NA	1	0	0	na
10.12b	9.50	49.00	1	M	1	T1	0	NA	1	0	0	na
10.12c	9.50	49.00	1	M	1	T1	1	NA	1	0	0	na
10.13a	13.60	78.00	0	UB	1	T3	1	LOG	1	0	0	7.5
10.13b	13.60	78.00	0	UB	1	T3	1	LOG	1	2	1	na
10.13c	13.60	78.00	0	UB	1	T3	1	LOG	1	0	0	5
10.14a	12.30	137.00	1	M	1	T2	1	LOG	1	2	1	10.3
10.14b	12.30	137.00	1	M	1	T2	1	LOG	1	4	1	na
10.14c	12.30	137.00	1	M	1	T2	0	LOG	1	0	1	9.5
10.15a	19.40	154.00	1	HM	0	T2	1	STUMP	0	3	1	na
10.15b	19.40	154.00	1	HM	0	T2	0	STUMP	1	0	0	na
10.15c	19.40	154.00	1	HM	0	T2	1	STUMP	1	0	0	na
10.16a	15.20	42.00	0	UB	1	T0	0	LOG	1	1	0	na
10.16b	15.20	42.00	0	UB	1	T0	0	LOG	1	1	0	na
10.16c	15.20	42.00	0	UB	1	T0	0	LOG	1	1	0	na
11.01a	3.00	38.00	0	UB	1	T2	0	ROCK	1	4	1	10.7
11.01b	3.00	38.00	0	UB	1	T2	1	ROCK	1	4	1	9
11.01c	3.00	38.00	0	UB	1	T2	1	ROCK	1	4	1	10
11.02a	4.87	338.00	1	HM	0	T3	1	STUMP	1	4	1	8
11.02b	4.87	338.00	1	HM	0	T3	1	STUMP	1	4	1	7.5
11.02c	4.87	338.00	1	HM	0	T3	1	STUMP	1	4	1	10.7
11.03a	7.50	12.00	1	HM	1	T3	1	LOG	1	5	1	13
11.03b	7.50	12.00	1	HM	1	T3	1	LOG	1	4	1	6.5
11.03c	7.50	12.00	1	HM	1	T3	1	LOG	1	5	1	12
11.04a	8.32	360.00	1	HM	0	T1	0	LOG	1	4	1	8

11.04b	8.32	360.00	1	HM	0	T1	0	LOG	1	4	1	9
11.04c	8.32	360.00	1	HM	0	T1	1	LOG	1	4	1	7
11.05a	15.90	4.00	1	HM	1	T1	0	NA	1	4	1	40.5
11.05b	15.90	4.00	1	HM	1	T1	0	NA	1	5	1	11
11.05c	15.90	4.00	1	HM	1	T1	1	NA	1	0	0	na
11.06a	15.00	21.00	1	HM	0	T2	1	NA	1	4	1	8
11.06b	15.00	21.00	1	HM	0	T2	1	NA	1	4	1	7.3
11.06c	15.00	21.00	1	HM	0	T2	0	NA	1	4	1	7.5
11.07a	13.87	27.00	1	HM	0	T3	1	LOG	1	4	1	8.5
11.07b	13.87	27.00	1	HM	0	T3	1	LOG	1	4	1	10.5
11.07c	13.87	27.00	1	HM	0	T3	1	LOG	1	5	1	10
11.08a	11.17	64.00	1	HM	0	T0	0	SNAG	1	5	1	8.5
11.08b	11.17	64.00	1	HM	0	T0	0	SNAG	1	5	1	7.5
11.08c	11.17	64.00	1	HM	0	T0	0	SNAG	1	4	1	7.5
11.09a	6.58	95.00	0	UB	1	T3	1	ROCK	1	5	1	11.5
11.09b	6.58	95.00	0	UB	1	T3	1	ROCK	1	4	1	9.5
11.09c	6.58	95.00	0	UB	1	T3	1	ROCK	1	2	1	5
11.10a	19.40	103.00	1	HM	1	T0	0	NA	1	4	1	8.5
11.10b	19.40	103.00	1	HM	1	T0	0	NA	1	4	1	8.5
11.10c	19.40	103.00	1	HM	1	T0	0	NA	1	4	1	7
11.11a	13.72	165.00	1	HM	1	T3	1	NA	1	0	0	na
11.11b	13.72	165.00	1	HM	1	T3	1	NA	1	0	0	na
11.11c	13.72	165.00	1	HM	1	T3	1	NA	1	0	0	na
11.12a	16.30	219.00	1	HM	0	T2	0	LOG	1	0	0	na
11.12b	16.30	219.00	1	HM	0	T2	1	LOG	1	4	1	5.5
11.12c	16.30	219.00	1	HM	0	T2	1	LOG	1	0	0	na
11.13a	7.73	308.00	1	HM	0	T2	0	SNAG	1	4	1	11.5
11.13b	7.73	308.00	1	HM	0	T2	1	SNAG	1	0	0	na
11.13c	7.73	308.00	1	HM	0	T2	1	SNAG	1	4	1	13
11.14a	18.54	124.00	0	UB	1	T0	0	NA	1	1	0	na
11.14b	18.54	124.00	0	UB	1	T0	0	NA	1	1	0	na
11.14c	18.54	124.00	0	UB	1	T0	0	NA	1	1	0	na
11.15a	10.36	143.00	0	UB	1	T1	0	LOG	0	3	1	7
11.15b	10.36	143.00	0	UB	1	T1	0	LOG	1	0	0	na
11.15c	10.36	143.00	0	UB	1	T1	1	LOG	1	0	0	na
12.01a	4.40	75.00	0	UB	1	T1	0	LOG	1	4	1	9
12.01b	4.40	75.00	0	UB	1	T1	0	LOG	1	4	1	11.5
12.01c	4.40	75.00	0	UB	1	T1	1	LOG	1	3	1	4.5
12.02a	5.60	41.00	0	UB	1	T2	0	NA	1	2	1	6.5
12.02b	5.60	41.00	0	UB	1	T2	1	NA	1	0	0	na

12.02c	5.60	41.00	0	UB	1	T2	1	NA	1	0	0	na
12.03a	14.19	40.00	1	HL	0	T1	0	NA	1	4	1	8
12.03b	14.19	40.00	1	HL	0	T1	0	NA	0	3	1	7
12.03c	14.19	40.00	1	HL	0	T1	1	NA	1	3	1	6
12.04a	14.00	46.00	1	HL	0	T3	1	SNAG	1	3	1	6.5
12.04b	14.00	46.00	1	HL	0	T3	1	SNAG	1	4	1	10
12.04c	14.00	46.00	1	HL	0	T3	1	SNAG	1	0	0	na
12.05a	14.20	59.00	0	HL	0	T2	0	NA	1	4	1	7
12.05b	14.20	59.00	0	HL	0	T2	1	NA	1	0	0	na
12.05c	14.20	59.00	0	HL	0	T2	1	NA	1	0	0	na
12.06a	16.00	50.00	0	HL	1	T0	0	NA	1	4	1	9
12.06b	16.00	50.00	0	HL	1	T0	0	NA	1	4	1	5.7
12.06c	16.00	50.00	0	HL	1	T0	0	NA	1	0	0	na
12.07a	11.51	60.00	1	HL	1	T0	0	NA	1	3	1	4.5
12.07b	11.51	60.00	1	HL	1	T0	0	NA	1	2	1	3.3
12.07c	11.51	60.00	1	HL	1	T0	0	NA	1	0	0	na
12.08a	12.47	90.00	1	HL	1	T3	1	NA	1	0	0	na
12.08b	12.47	90.00	1	HL	1	T3	1	NA	0	0	0	na
12.08c	12.47	90.00	1	HL	1	T3	1	NA	0	0	0	na
12.09a	10.52	100.00	0	UB	1	T0	0	ROCK	1	0	0	na
12.09b	10.52	100.00	0	UB	1	T0	0	ROCK	1	0	0	na
12.09c	10.52	100.00	0	UB	1	T0	0	ROCK	0	3	1	4.5
12.10a	12.52	120.00	1	HL	1	T2	1	ROCK	1	1	0	na
12.10b	12.52	120.00	1	HL	1	T2	0	ROCK	1	0	0	na
12.10c	12.52	120.00	1	HL	1	T2	1	ROCK	0	0	0	na
12.11a	9.00	115.00	1	HL	1	T1	1	ROCK	0	0	0	na
12.11b	9.00	115.00	1	HL	1	T1	0	ROCK	0	0	0	na
12.11c	9.00	115.00	1	HL	1	T1	0	ROCK	1	0	0	na
12.12a	14.32	2.00	1	HL	0	T2	1	NA	1	0	0	na
12.12b	14.32	2.00	1	HL	0	T2	1	NA	1	0	0	na
12.12c	14.32	2.00	1	HL	0	T2	0	NA	1	0	0	na
12.13a	16.25	7.00	1	HL	1	T0	0	NA	1	0	0	na
12.13b	16.25	7.00	1	HL	1	T0	0	NA	0	3	1	7
12.13c	16.25	7.00	1	HL	1	T0	0	NA	0	4	1	10
12.14a	21.20	3.00	0	UB	1	T1	0	NA	0	0	0	na
12.14b	21.20	3.00	0	UB	1	T1	0	NA	0	3	1	4
12.14c	21.20	3.00	0	UB	1	T1	1	NA	0	4	1	10
12.15a	20.48	355.00	1	HL	1	T3	1	NA	0	0	0	na
12.15b	20.48	355.00	1	HL	1	T3	1	NA	1	0	0	na
12.15c	20.48	355.00	1	HL	1	T3	1	NA	0	0	0	na

12.16a	26.15	283.00	1	HL	0	T2	1	SNAG	1	1	0	na
12.16b	26.15	283.00	1	HL	0	T2	0	SNAG	1	1	0	na
12.16c	26.15	283.00	1	HL	0	T2	1	SNAG	1	1	0	na
12.17a	21.95	300.00	1	HL	0	T0	0	NA	1	0	0	na
12.17b	21.95	300.00	1	HL	0	T0	0	NA	1	0	0	na
12.17c	21.95	300.00	1	HL	0	T0	0	NA	1	0	0	na
12.18a	24.48	310.00	1	HL	1	T1	1	ROCK	0	0	0	na
12.18b	24.48	310.00	1	HL	1	T1	0	ROCK	1	2	1	2
12.18c	24.48	310.00	1	HL	1	T1	0	ROCK	0	2	1	7.5
12.19a	17.55	310.00	0	UB	0	T2	0	NA	1	0	0	na
12.19b	17.55	310.00	0	UB	0	T2	1	NA	1	0	0	na
12.19c	17.55	310.00	0	UB	0	T2	1	NA	1	3	1	4
13.01a	4.17	281.00	0	UB	1	T2	1	NA	1	2	1	5
13.01b	4.17	281.00	0	UB	1	T2	0	NA	1	0	0	na
13.01c	4.17	281.00	0	UB	1	T2	1	NA	1	4	1	9.5
13.02a	4.19	307.00	1	M	0	T1	1	STUMP	1	3	1	6
13.02b	4.19	307.00	1	M	0	T1	0	STUMP	1	4	1	9.7
13.02c	4.19	307.00	1	M	0	T1	0	STUMP	1	3	1	6.5
13.03a	13.36	322.00	1	HM	1	T1	1	NA	1	4	1	7.5
13.03b	13.36	322.00	1	HM	1	T1	0	NA	1	0	0	na
13.03c	13.36	322.00	1	HM	1	T1	0	NA	1	0	0	na
13.04a	10.45	328.00	0	UB	1	T2	0	NA	1	0	0	na
13.04b	10.45	328.00	0	UB	1	T2	1	NA	0	3	1	6.5
13.04c	10.45	328.00	0	UB	1	T2	1	NA	1	4	1	9
13.05a	12.06	337.00	0	UB	1	T0	0	NA	1	3	1	6.5
13.05b	12.06	337.00	0	UB	1	T0	0	NA	1	0	0	na
13.05c	12.06	337.00	0	UB	1	T0	0	NA	1	0	0	na
13.06a	7.60	340.00	1	HL	1	T3	1	NA	1	3	1	7.5
13.06b	7.60	340.00	1	HL	1	T3	1	NA	0	3	1	9.5
13.06c	7.60	340.00	1	HL	1	T3	1	NA	0	3	1	8
13.07a	10.65	35.00	1	HM	1	T0	0	NA	1	0	0	na
13.07b	10.65	35.00	1	HM	1	T0	0	NA	1	3	1	4.5
13.07c	10.65	35.00	1	HM	1	T0	0	NA	1	4	1	7.3
13.08a	13.52	59.00	1	M	1	T2	1	NA	1	4	1	9
13.08b	13.52	59.00	1	M	1	T2	0	NA	0	2	1	4
13.08c	13.52	59.00	1	M	1	T2	1	NA	0	3	1	7
13.09a	13.07	66.00	1	M	1	T3	1	NA	1	4	1	9
13.09b	13.07	66.00	1	M	1	T3	1	NA	1	4	1	11.5
13.09c	13.07	66.00	1	M	1	T3	1	NA	1	5	1	15
13.10a	17.52	73.00	0	UB	1	T1	0	NA	1	4	1	7.5

13.10b	17.52	73.00	0	UB	1	T1	1	NA	1	5	1	13
13.10c	17.52	73.00	0	UB	1	T1	0	NA	1	4	1	7
13.11a	13.94	129.00	1	HM	1	T2	0	NA	1	0	0	na
13.11b	13.94	129.00	1	HM	1	T2	1	NA	1	2	1	4
13.11c	13.94	129.00	1	HM	1	T2	1	NA	1	2	1	6
13.12a	11.33	127.00	1	HH	1	T1	0	NA	1	4	1	6.5
13.12b	11.33	127.00	1	HH	1	T1	0	NA	1	0	0	na
13.12c	11.33	127.00	1	HH	1	T1	1	NA	1	3	1	7.5
13.13a	14.33	134.00	1	HH	1	T3	1	NA	1	0	0	na
13.13b	14.33	134.00	1	HH	1	T3	1	NA	1	4	1	12
13.13c	14.33	134.00	1	HH	1	T3	1	NA	1	4	1	10.5
13.14a	11.27	143.00	1	HH	1	T0	0	NA	0	5	1	8
13.14b	11.27	143.00	1	HM	1	T0	0	NA	1	5	1	8
13.14c	11.27	143.00	1	HM	1	T0	0	NA	1	5	1	7
13.15a	19.55	156.00	0	UB	1	T3	1	LOG	1	0	0	na
13.15b	19.55	156.00	0	UB	1	T3	1	LOG	1	0	0	na
13.15c	19.55	156.00	0	UB	1	T3	1	LOG	1	2	1	5.5
13.16a	20.00	173.00	1	HH	1	T0	0	STUMP	1	5	1	9
13.16b	20.00	173.00	1	HH	1	T0	0	STUMP	1	5	1	13
13.17a	16.26	250.00	1	HH	0	T3	1	LOG	1	5	1	9.5
13.17b	16.26	250.00	1	HH	0	T3	1	LOG	1	3	1	5.7
13.17c	16.26	250.00	1	HH	0	T3	1	LOG	1	4	1	12
13.18a	18.18	255.00	1	HH	0	T0	0	STUMP	1	3	1	5
13.18b	18.18	255.00	1	HH	0	T0	0	STUMP	1	0	0	na
13.18c	18.18	255.00	1	HH	0	T0	0	STUMP	1	3	1	5.5
14.01a	1.25	201.00	0	UB	1	T0	0	LOG	1	0	0	na
14.01b	1.25	201.00	0	UB	1	T0	0	LOG	1	0	0	na
14.01c	1.25	201.00	0	UB	1	T0	0	LOG	1	0	0	na
14.02a	9.09	214.00	1	HM	0	T2	1	LOG	1	5	1	11
14.02b	9.09	214.00	1	HM	0	T2	1	LOG	1	5	1	11
14.02c	9.09	214.00	1	HM	0	T2	0	LOG	1	5	1	13
14.03a	6.57	300.00	1	HL	1	T3	1	NA	1	5	1	10
14.03b	6.57	300.00	1	HL	1	T3	1	NA	0	4	1	9
14.03c	6.57	300.00	1	HL	1	T3	1	NA	1	5	1	11
14.04a	11.54	325.00	0	UB	1	T1	0	LOG	1	2	1	5
14.04b	11.54	325.00	0	UB	1	T1	1	LOG	1	0	0	na
14.04c	11.54	325.00	0	UB	1	T1	0	LOG	1	0	0	na
14.05a	7.43	346.00	0	UB	1	T2	1	LOG	1	5	1	13
14.05b	7.43	346.00	0	UB	1	T2	1	LOG	1	5	1	10.5
14.05c	7.43	346.00	0	UB	1	T2	0	LOG	1	2	1	10.5

14.06a	16.88	349.00	1	HH	0	T1	0	SNAG	1	4	1	8
14.06b	16.88	349.00	1	HH	0	T1	0	SNAG	1	5	1	8.5
14.06c	16.88	349.00	1	HH	0	T1	1	SNAG	1	4	1	10
14.07a	11.84	354.00	1	HH	0	T2	0	SNAG	1	3	1	4.5
14.07b	11.84	354.00	1	HH	0	T2	1	SNAG	1	4	1	6
14.07c	11.84	354.00	1	HH	0	T2	1	SNAG	1	4	1	7
14.08a	16.54	357.00	1	HH	0	T0	0	SNAG	1	0	0	na
14.08b	16.54	357.00	1	HH	0	T0	0	SNAG	1	3	1	6
14.08c	16.54	357.00	1	HH	0	T0	0	SNAG	1	4	1	4
14.09a	10.63	11.00	1	HH	0	T3	1	SNAG	1	5	1	13
14.09b	10.63	11.00	1	HH	0	T3	1	SNAG	1	5	1	17
14.09c	10.63	11.00	1	HH	0	T3	1	SNAG	1	5	1	9
14.10a	10.36	52.00	1	HH	1	T0	0	SNAG	1	3	1	6.5
14.10b	10.36	52.00	1	HH	1	T0	0	SNAG	1	4	1	13
14.10c	10.36	52.00	1	HH	1	T0	0	SNAG	1	4	1	8.5
14.11a	7.29	98.00	1	HM	1	T1	0	SNAG	1	0	0	na
14.11b	7.29	98.00	1	HM	1	T1	1	SNAG	1	4	1	9.3
14.11c	7.29	98.00	1	HM	1	T1	0	SNAG	1	3	1	8.5
14.12a	14.48	118.00	1	HM	1	T2	1	SNAG	1	5	1	10
14.12b	14.48	118.00	1	HM	1	T2	0	SNAG	1	4	1	11
14.12c	14.48	118.00	1	HM	1	T2	1	SNAG	1	4	1	9.3
14.13a	6.48	149.00	0	UB	0	T3	1	NA	1	2	1	12
14.13b	6.48	149.00	0	UB	0	T3	1	NA	1	0	0	na
14.13c	6.48	149.00	0	UB	0	T3	1	NA	1	2	1	10.3
14.14a	9.10	208.00	1	HM	1	T2	1	LOG/snag	0	5	1	10.5
14.14b	9.10	208.00	1	HM	1	T2	1	LOG/snag	1	5	1	8
14.14c	9.10	208.00	1	HM	1	T2	0	LOG/snag	1	5	1	13
14.15a	16.00	192.00	1	HH	1	T3	1	SNAG	1	4	1	7.5
14.15b	16.00	192.00	1	HH	1	T3	1	SNAG	1	4	1	12
14.15c	16.00	192.00	1	HH	1	T3	1	SNAG	1	4	1	8
14.16a	13.59	198.00	1	HH	0	T0	0	SNAG	1	5	1	11
14.16b	13.59	198.00	1	HH	0	T0	0	SNAG	1	5	1	10.5
14.16c	13.59	198.00	1	HH	0	T0	0	SNAG	1	5	1	9.5
15.01a	21.67	73.00	0	UB	1	T3	1	LOG	1	0	0	na
15.01b	21.67	73.00	0	UB	1	T3	1	LOG	1	3	1	7
15.01c	21.67	73.00	0	UB	1	T3	1	LOG	1	3	1	9.5
15.02a	7.81	82.00	1	HH	1	T0	0	SNAG	1	4	1	8.5
15.02b	7.81	82.00	1	HH	1	T0	0	SNAG	1	4	1	12.5
15.02c	7.81	82.00	1	HH	1	T0	0	SNAG	1	4	1	11
15.03a	12.10	98.00	1	HH	1	T3	1	NA	1	4	1	10.5

15.03b	12.10	98.00	1	HH	1	T3	1	NA	1	4	1	7.7
15.03c	12.10	98.00	1	HH	1	T3	1	NA	1	4	1	8
15.04a	20.80	108.00	1	HH	1	T2	1	LOG	1	5	1	10
15.04b	20.80	108.00	1	HH	1	T2	1	LOG	1	4	1	9
15.04c	20.80	108.00	1	HH	1	T2	0	LOG	1	0	0	na
15.05a	15.86	121.00	1	HH	0	T1	0	LOG	1	3	1	9.5
15.05b	15.86	121.00	1	HH	0	T1	1	LOG	1	4	1	12
15.05c	15.86	121.00	1	HH	0	T1	0	LOG	1	4	1	14.5
15.06a	14.76	142.00	1	HH	1	T2	1	SNAG	1	4	1	11
15.06b	14.76	142.00	1	HH	1	T2	0	SNAG	1	0	0	na
15.06c	14.76	142.00	1	HH	1	T2	1	SNAG	1	5	1	7.5
15.07a	16.09	142.00	1	HH	1	T3	1	NA	1	3	1	5
15.07b	16.09	142.00	1	HH	1	T3	1	NA	1	3	1	7
15.07c	16.09	142.00	1	HH	1	T3	1	NA	1	5	1	11.5
15.08a	17.80	155.00	1	HH	1	T1	0	NA	1	5	1	13
15.08b	17.80	155.00	1	HH	1	T1	0	NA	1	5	1	7
15.08c	17.80	155.00	1	HH	1	T1	1	NA	1	5	1	12
15.09a	20.08	162.00	1	HH	0	T0	0	ROCK	1	0	0	na
15.09b	20.08	162.00	1	HH	0	T0	0	ROCK	1	4	1	6
15.09c	20.08	162.00	1	HH	0	T0	0	ROCK	1	2	1	5.5
15.10a	20.60	185.00	1	HH	0	T0	0	LOG	1	3	1	5.5
15.10b	20.60	185.00	1	HH	0	T0	0	LOG	1	2	1	2
15.10c	20.60	185.00	1	HH	0	T0	0	LOG	1	2	1	4
15.11a	14.44	227.00	0	UB	1	T1	1	LOG	1	2	1	5.5
15.11b	14.44	227.00	0	UB	1	T1	0	LOG	1	3	1	10
15.11c	14.44	227.00	0	UB	1	T1	0	LOG	1	2	1	5
15.12a	5.84	248.00	1	HM	1	T1	0	LOG	1	0	0	na
15.12b	5.84	248.00	1	HM	1	T1	1	LOG	1	0	0	na
15.12c	5.84	248.00	1	HM	1	T1	0	LOG	1	0	0	na
15.13a	7.50	350.00	1	HH	1	T2	1	LOG	1	4	1	7.5
15.13b	7.50	350.00	1	HH	1	T2	1	LOG	1	4	1	7
15.13c	7.50	350.00	1	HH	1	T2	0	LOG	1	0	0	na
15.14a	7.55	290.00	1	HH	1	T3	1	LOG	1	0	0	na
15.14b	7.55	290.00	1	HH	1	T3	1	LOG	1	0	0	na
15.14c	7.55	290.00	1	HH	1	T3	1	LOG	1	0	0	na
15.15a	24.82	104.00	0	UB	1	T0	0	NA	1	3	1	7
15.15b	24.82	104.00	0	UB	1	T0	0	NA	1	3	1	7.5
15.15c	24.82	104.00	0	UB	1	T0	0	NA	0	1	0	na
15.16a	29.06	104.00	0	UB	1	T3	1	NA	1	4	1	7.5
15.16b	29.06	104.00	0	UB	1	T3	1	NA	1	4	1	11

15.16c	29.06	104.00	0	UB	1	T3	1	NA	1	4	1	10.3
16.01a	4.70	36.00	1	M	1	T1	1	NA	1	5	1	12.5
16.01b	4.70	36.00	1	M	1	T1	0	NA	1	4	1	9.5
16.01c	4.70	36.00	1	M	1	T1	0	NA	0	5	1	11.5
16.02a	8.20	44.00	1	L	1	T2	0	NA	1	0	0	na
16.02b	8.20	44.00	1	L	1	T2	1	NA	1	2	1	5.5
16.02c	8.20	44.00	1	L	1	T2	1	NA	1	0	0	na
16.03a	10.10	70.00	1	HH	1	T3	1	SNAG	1	5	1	13.5
16.03b	10.10	70.00	1	HH	1	T3	1	SNAG	1	5	1	8.7
16.03c	10.10	70.00	1	HH	1	T3	1	SNAG	1	5	1	10
16.04a	14.20	126.00	0	UB	1	T3	1	NA	1	0	0	na
16.04b	14.20	126.00	0	UB	1	T3	1	NA	1	0	0	na
16.04c	14.20	126.00	0	UB	1	T3	1	NA	1	0	0	na
16.05a	13.60	140.00	0	UB	1	T1	0	NA	1	5	1	10
16.05b	13.60	140.00	0	UB	1	T1	0	NA	1	4	1	6
16.05c	13.60	140.00	0	UB	1	T1	1	NA	1	0	0	na
16.06a	14.50	156.00	0	UB	1	T1	1	NA	1	4	1	13.7
16.06b	14.50	156.00	0	UB	1	T1	0	NA	1	4	1	2.5
16.06c	14.50	156.00	0	UB	1	T1	0	NA	1	4	1	8.5
16.07a	12.70	185.00	1	HM	1	T2	1	NA	0	3	1	8.5
16.07b	12.70	185.00	1	HM	1	T2	0	NA	0	0	0	na
16.07c	12.70	185.00	1	HM	1	T2	1	NA	0	0	0	na
16.08a	7.50	191.00	1	HH	1	T1	0	NA	1	3	1	8
16.08b	7.50	191.00	1	HH	1	T1	1	NA	1	0	0	na
16.08c	7.50	191.00	1	HH	1	T1	0	NA	1	2	1	6
16.09a	10.20	194.00	1	HH	1	T0	0	NA	1	5	1	12.5
16.09b	10.20	194.00	1	HH	1	T0	0	NA	1	3	1	8
16.09c	10.20	194.00	1	HH	1	T0	0	NA	1	0	0	na
16.10a	5.10	219.00	0	UB	1	T3	1	NA	1	4	1	11.5
16.10b	5.10	219.00	0	UB	1	T3	1	NA	1	4	1	9.5
16.10c	5.10	219.00	0	UB	1	T3	1	NA	1	4	1	9.5
16.11a	19.40	246.00	1	HL	1	T2	1	NA	1	2	1	4.5
16.11b	19.40	246.00	1	HL	1	T2	0	NA	1	3	1	8
16.11c	19.40	246.00	1	HL	1	T2	1	NA	1	2	1	7.5
16.12a	12.90	256.00	1	HM	1	T3	1	NA	1	4	1	9
16.12b	12.90	256.00	1	HM	1	T3	1	NA	1	4	1	11.5
16.12c	12.90	256.00	1	HM	1	T3	1	NA	0	4	1	8
16.13a	16.70	262.00	1	M	1	T0	0	NA	1	0	0	na
16.13b	16.70	262.00	1	M	1	T0	0	NA	1	3	1	7.5
16.13c	16.70	262.00	1	M	1	T0	0	NA	1	3	1	7.3

16.14a	10.10	276.00	1	HL	1	T1	0	NA	1	0	0	na
16.14b	10.10	276.00	1	HL	1	T1	0	NA	1	0	0	na
16.14c	10.10	276.00	1	HL	1	T1	1	NA	1	0	0	na
16.15a	5.40	86.00	1	HL	1	T0	0	NA	1	4	1	11
16.15b	5.40	86.00	1	HL	1	T0	0	NA	1	4	1	9.7
16.15c	5.40	86.00	1	HL	1	T0	0	NA	1	1	0	na
16.16a	17.90	145.00	1	HL	1	T0	0	NA	1	0	0	na
16.16b	17.90	145.00	1	HL	1	T0	0	NA	1	0	0	na
16.16c	17.90	145.00	1	HL	1	T0	0	NA	1	4	1	14
17.01a	12.20	315.00	1	HM	0	T2	1	SNAG	1	4	1	8
17.01b	12.20	315.00	1	HM	0	T2	1	SNAG	1	0	0	na
17.01c	12.20	315.00	1	HM	0	T2	0	SNAG	0	3	1	5
17.02a	7.60	332.00	0	UB	0	T0	0	NA	1	0	0	na
17.02b	7.60	332.00	0	UB	0	T0	0	NA	1	0	0	na
17.02c	7.60	332.00	0	UB	0	T0	0	NA	1	0	0	na
17.03a	9.70	8.00	1	HH	1	T0	0	LOG	0	5	1	9
17.03b	9.70	8.00	1	HH	1	T0	0	LOG	1	4	1	7
17.03c	9.70	8.00	1	HH	1	T0	0	LOG	1	4	1	6.5
17.04a	7.10	55.00	1	HH	0	T3	1	LOG	1	4	1	9
17.04b	7.10	55.00	1	HH	0	T3	1	LOG	1	4	1	11.5
17.04c	7.10	55.00	1	HH	0	T3	1	LOG	1	4	1	10
17.05a	15.60	61.00	0	UB	1	T0	0	LOG	1	2	1	3.5
17.05b	15.60	61.00	0	UB	1	T0	0	LOG	1	0	0	na
17.05c	15.60	61.00	0	UB	1	T0	0	LOG	1	5	1	10.5
17.06a	13.10	84.00	1	HH	0	T2	1	SNAG	1	4	1	8
17.06b	13.10	84.00	1	HH	0	T2	1	SNAG	1	4	1	8
17.06c	13.10	84.00	1	HH	0	T2	0	SNAG	1	4	1	6
17.07a	16.00	105.00	1	HH	0	T1	0	SNAG	1	4	1	9
17.07b	16.00	105.00	1	HH	0	T1	0	SNAG	1	5	1	12
17.07c	16.00	105.00	1	HH	0	T1	1	SNAG	1	5	1	13.5
17.08a	5.40	122.00	0	UB	0	T1	0	NA	1	0	0	na
17.08b	5.40	122.00	0	UB	0	T1	1	NA	1	0	0	na
17.08c	5.40	122.00	0	UB	0	T1	0	NA	1	0	0	na
17.09a	9.50	124.00	0	UB	0	T3	1	NA	1	0	0	na
17.09b	9.50	124.00	0	UB	0	T3	1	NA	1	0	0	na
17.09c	9.50	124.00	0	UB	0	T3	1	NA	1	0	0	na
17.10a	18.90	113.00	1	HH	0	T1	0	LOG	1	4	1	7.5
17.10b	18.90	113.00	1	HH	0	T1	0	LOG	1	5	1	7
17.10c	18.90	113.00	1	HH	0	T1	1	LOG	1	5	1	7
17.11a	18.60	128.00	1	HH	0	T3	1	LOG	1	0	0	na

17.11b	18.60	128.00	1	HH	0	T3	1	LOG	1	0	0	na
17.11c	18.60	128.00	1	HH	0	T3	1	LOG	1	0	0	na
17.12a	12.20	139.00	1	HH	1	T0	0	NA	1	4	1	8.5
17.12b	12.20	139.00	1	HH	1	T0	0	NA	1	0	0	na
17.12c	12.20	139.00	1	HH	1	T0	0	NA	1	0	0	na
17.13a	10.10	176.00	1	HM	1	T2	1	SNAG	1	4	1	8
17.13b	10.10	176.00	1	HM	1	T2	1	SNAG	1	5	1	10.5
17.13c	10.10	176.00	1	HM	1	T2	1	SNAG	1	5	1	9.5
17.14a	11.50	211.00	1	HM	1	T3	1	SNAG	1	0	0	na
17.14b	11.50	211.00	1	HM	1	T3	1	SNAG	1	2	1	7.5
17.14c	11.50	211.00	1	HM	1	T3	1	SNAG	1	0	0	na
17.15a	8.50	212.00	1	HM	0	T1	0	SNAG	1	2	1	5.3
17.15b	8.50	212.00	1	HM	0	T1	1	SNAG	1	4	1	7.3
17.15c	8.50	212.00	1	HM	0	T1	0	SNAG	1	2	1	8
17.16a	13.10	275.00	1	HM	1	T0	0	LOG	1	0	0	na
17.16b	13.10	275.00	1	HM	1	T0	0	LOG	1	0	0	na
17.16c	13.10	275.00	1	HM	1	T0	0	LOG	1	3	1	7.7
18.01a	4.44	227.00	1	HM	1	T1	0	SNAG	1	0	0	na
18.01b	4.44	227.00	1	HM	1	T1	1	SNAG	1	3	1	9
18.01c	4.44	227.00	1	HM	1	T1	0	SNAG	1	0	0	na
18.02a	10.72	252.00	1	HM	1	T3	1	SNAG	1	4	1	7.5
18.02b	10.72	252.00	1	HM	1	T3	1	SNAG	1	4	1	6.3
18.02c	10.72	252.00	1	HM	1	T3	1	SNAG	1	0	0	na
18.03a	10.60	273.00	1	HH	1	T2	1	SNAG	1	5	1	8
18.03b	10.60	273.00	1	HH	1	T2	0	SNAG	1	5	1	9.5
18.03c	10.60	273.00	1	HH	1	T2	1	SNAG	1	5	1	11.5
18.04a	13.40	277.00	1	HM	1	T2	1	SNAG	1	4	1	7
18.04b	13.40	277.00	1	HM	1	T2	1	SNAG	1	5	1	8
18.04c	13.40	277.00	1	HM	1	T2	0	SNAG	1	5	1	10
18.05a	12.90	323.00	1	HH	0	T3	1	SNAG	1	5	1	15.3
18.05b	12.90	323.00	1	HH	0	T3	1	SNAG	1	2	1	2
18.05c	12.90	323.00	1	HH	0	T3	1	SNAG	1	5	1	9
18.06a	4.80	345.00	0	UB	0	T1	1	LOG	1	0	0	na
18.06b	4.80	345.00	0	UB	0	T1	0	LOG	1	0	0	na
18.06c	4.80	345.00	0	UB	0	T1	0	LOG	1	0	0	na
18.07a	12.50	362.00	1	HM	0	T0	0	LOG	1	0	0	na
18.07b	12.50	362.00	1	HM	0	T0	0	LOG	1	2	1	5.7
18.07c	12.50	362.00	1	HM	0	T0	0	LOG	1	2	1	6
18.08a	11.90	9.00	1	HM	0	T1	0	SNAG	1	4	1	6.7
18.08b	11.90	9.00	1	HM	0	T1	1	SNAG	1	4	1	6

18.08c	11.90	9.00	1	HM	0	T1	0	SNAG	1	4	1	9.3
18.09a	8.50	15.00	1	HM	1	T3	1	SNAG	1	4	1	12.3
18.09b	8.50	15.00	1	HM	1	T3	1	SNAG	1	3	1	4
18.09c	8.50	15.00	1	HM	1	T3	1	SNAG	0	4	1	9
18.10a	10.10	16.00	1	HM	0	T2	1	SNAG	1	0	0	na
18.10b	10.10	16.00	1	HM	0	T2	1	SNAG	1	4	1	9.5
18.10c	10.10	16.00	1	HM	0	T2	0	SNAG	1	0	0	na
18.11a	6.50	44.00	0	UB	0	T0	0	NA	1	2	1	4
18.11b	6.50	44.00	0	UB	0	T0	0	NA	1	0	0	na
18.11c	6.50	44.00	0	UB	0	T0	0	NA	1	3	1	8
18.12a	11.50	45.00	0	UB	0	T0	0	LOG	1	0	0	na
18.12b	11.50	45.00	0	UB	0	T0	0	LOG	1	3	1	9.5
18.12c	11.50	45.00	0	UB	0	T0	0	LOG	1	0	0	na
18.13a	9.50	80.00	1	HH	0	T1	0	SNAG	1	0	0	na
18.13b	9.50	80.00	1	HH	0	T1	1	SNAG	1	5	1	13
18.13c	9.50	80.00	1	HH	0	T1	0	SNAG	0	4	1	4
18.14a	15.80	92.00	1	HH	0	T0	0	SNAG	1	3	1	9
18.14b	15.80	92.00	1	HH	0	T0	0	SNAG	0	5	1	8.5
18.14c	15.80	92.00	1	HH	0	T0	0	SNAG	1	5	1	8.5
18.15a	10.80	147.00	0	UB	0	T3	1	NA	1	0	0	na
18.15b	10.80	147.00	0	UB	0	T3	1	NA	1	0	0	na
18.15c	10.80	147.00	0	UB	0	T3	1	NA	1	2	1	7
18.16a	10.10	172.00	1	HM	0	T0	0	SNAG	1	0	0	na
18.16b	10.10	172.00	1	HM	0	T0	0	SNAG	1	3	1	6.5
18.16c	10.10	172.00	1	HM	0	T0	0	SNAG	1	0	0	na
19.01a	3.60	279.00	1	HM	1	T2	1	NA	1	2	1	6
19.01b	3.60	279.00	1	HM	1	T2	0	NA	1	4	1	9
19.01c	3.60	279.00	1	HM	1	T2	1	NA	1	4	1	8.5
19.02a	8.10	298.00	1	HM	0	T1	0	SNAG	1	4	1	8.5
19.02b	8.10	298.00	1	HM	0	T1	1	SNAG	1	3	1	6.5
19.02c	8.10	298.00	1	HM	0	T1	0	SNAG	1	0	0	na
19.03a	5.70	302.00	1	HH	1	T0	0	SNAG	1	0	0	na
19.03b	5.70	302.00	1	HH	1	T0	0	SNAG	1	0	0	na
19.03c	5.70	302.00	1	HH	1	T0	0	SNAG	1	0	0	na
19.04a	6.90	346.00	1	HM	1	T3	1	NA	1	0	0	na
19.04b	6.90	346.00	1	HM	1	T3	1	NA	1	0	0	na
19.04c	6.90	346.00	1	HM	1	T3	1	NA	1	2	1	9.5
19.05a	6.90	33.00	1	HM	0	T1	1	NA	1	5	1	10.5
19.05b	6.90	33.00	1	HM	0	T1	0	NA	1	2	1	2.5
19.05c	6.90	33.00	1	HM	0	T1	0	NA	1	4	1	9.5

19.06a	13.60	25.00	1	HH	1	T1	0	NA	1	4	1	6
19.06b	13.60	25.00	1	HH	1	T1	0	NA	1	4	1	6.5
19.06c	13.60	25.00	1	HH	1	T1	1	NA	1	4	1	8
19.07a	18.70	48.00	1	HH	0	T2	0	NA	1	0	0	na
19.07b	18.70	48.00	1	HH	0	T2	1	NA	1	2	1	7
19.07c	18.70	48.00	1	HH	0	T2	1	NA	1	0	0	na
19.08a	6.30	59.00	1	HM	1	T0	0	LOG	1	0	0	na
19.08b	6.30	59.00	1	HM	1	T0	0	LOG	1	0	0	na
19.08c	6.30	59.00	1	HM	1	T0	0	LOG	1	0	0	na
19.09a	6.80	90.00	1	M	1	T3	1	NA	1	3	1	6.5
19.09b	6.80	90.00	1	M	1	T3	1	NA	1	3	1	7.5
19.09c	6.80	90.00	1	M	1	T3	1	NA	1	4	1	9
19.10a	10.80	102.00	1	HM	1	T2	0	NA	1	0	0	na
19.10b	10.80	102.00	1	HM	1	T2	1	NA	1	3	1	6.5
19.10c	10.80	102.00	1	HM	1	T2	1	NA	1	0	0	na
19.11a	11.50	176.00	1	M	1	T1	0	NA	1	5	1	12
19.11b	11.50	176.00	1	M	1	T1	0	NA	1	0	0	na
19.11c	11.50	176.00	1	M	1	T1	1	NA	1	5	1	8.5
19.12a	10.00	192.00	1	M	1	T0	0	SHRUB	1	4	1	7.5
19.12b	10.00	192.00	1	M	1	T0	0	SHRUB	1	4	1	9.5
19.12c	10.00	192.00	1	M	1	T0	0	SHRUB	1	4	1	5
19.13a	12.30	207.00	1	HL	1	T2	1	NA	1	4	1	8.5
19.13b	12.30	207.00	1	HL	1	T2	1	NA	1	4	1	10.5
19.13c	12.30	207.00	1	HL	1	T2	0	NA	1	4	1	7
19.14a	10.40	241.00	1	HM	1	T3	1	NA	1	3	1	8
19.14b	10.40	241.00	1	HM	1	T3	1	NA	1	0	0	na
19.14c	10.40	241.00	1	HM	1	T3	1	NA	1	3	1	7
19.15a	13.00	87.00	0	UB	1	T1	1	LOG	1	3	1	9.5
19.15b	13.00	87.00	0	UB	1	T1	0	LOG	1	3	1	6
19.15c	13.00	87.00	0	UB	1	T1	0	LOG	1	3	1	6
19.16a	18.50	64.00	1	HH	0	T2	1	STUMP	1	5	1	11.3
19.16b	18.50	64.00	1	HH	0	T2	0	STUMP	1	4	1	8
19.16c	18.50	64.00	1	HH	0	T2	1	STUMP	1	5	1	12
20.01a	2.90	288.00	1	HM	0	T2	1	SNAG	1	4	1	7.3
20.01b	2.90	288.00	1	HM	0	T2	1	SNAG	1	2	1	4.2
20.01c	2.90	288.00	1	HM	0	T2	0	SNAG	1	4	1	7.5
20.02a	17.80	319.00	1	HH	0	T0	0	LOG	1	3	1	3.5
20.02b	17.80	319.00	1	HH	0	T0	0	LOG	1	4	1	5.5
20.02c	17.80	319.00	1	HH	0	T0	0	LOG	1	0	0	na
20.03a	15.40	355.00	1	HH	0	T1	1	SNAG	1	4	1	12

20.03b	15.40	355.00	1	HH	0	T1	0	SNAG	1	3	1	6
20.03c	15.40	355.00	1	HH	0	T1	0	SNAG	1	0	0	na
20.04a	6.90	5.00	1	HL	0	T0	0	LOG	1	2	1	6
20.04b	6.90	5.00	1	HL	0	T0	0	LOG	1	0	0	na
20.04c	6.90	5.00	1	HL	0	T0	0	LOG	1	3	1	5
20.05a	5.80	18.00	1	HL	0	T1	1	LOG	1	3	1	6
20.05b	5.80	18.00	1	HL	0	T1	0	LOG	1	3	1	5.5
20.05c	5.80	18.00	1	HL	0	T1	0	LOG	1	3	1	8.5
20.06a	5.40	108.00	0	UB	1	T3	1	NA	1	0	0	na
20.06b	5.40	108.00	0	UB	1	T3	1	NA	1	3	1	4
20.06c	5.40	108.00	0	UB	1	T3	1	NA	1	3	1	8.5
20.07a	7.15	157.00	0	UB	0	T2	1	NA	1	0	0	na
20.07b	7.15	157.00	0	UB	0	T2	1	NA	1	0	0	na
20.07c	7.15	157.00	0	UB	0	T2	0	NA	1	0	0	na
20.08a	11.15	163.00	1	HH	1	T2	1	SNAG	1	4	1	7
20.08b	11.15	163.00	1	HH	1	T2	0	SNAG	1	3	1	6
20.08c	11.15	163.00	1	HH	1	T2	1	SNAG	1	4	1	9.5
20.09a	9.50	195.00	1	HM	1	T1	1	NA	1	4	1	6.5
20.09b	9.50	195.00	1	HM	1	T1	0	NA	1	2	1	2.5
20.09c	9.50	195.00	1	HM	1	T1	0	NA	1	3	1	8.5
20.10a	11.50	209.00	1	HM	0	T3	1	LOG	1	4	1	8.3
20.10b	11.50	209.00	1	HM	0	T3	1	LOG	1	3	1	6
20.10c	11.50	209.00	1	HM	0	T3	1	LOG	1	4	1	12.3
20.11a	13.50	211.00	1	HH	0	T1	0	NA	1	5	1	5
20.11b	13.50	211.00	1	HH	0	T1	1	NA	1	5	1	9
20.11c	13.50	211.00	1	HH	0	T1	0	NA	1	4	1	5.5
20.12a	12.45	217.00	1	HH	0	T0	0	NA	1	4	1	5
20.12b	12.45	217.00	1	HH	0	T0	0	NA	1	4	1	7.5
20.12c	12.45	217.00	1	HH	0	T0	0	NA	1	3	1	3
20.13a	8.90	217.00	1	HM	0	T0	0	NA	1	3	1	7.5
20.13b	8.90	217.00	1	HM	0	T0	0	NA	1	3	1	7.5
20.13c	8.90	217.00	1	HM	0	T0	0	NA	1	3	1	6
20.14a	13.20	224.00	1	HH	1	T3	1	NA	1	2	1	5.5
20.14b	13.20	224.00	1	HH	1	T3	1	NA	1	0	0	na
20.14c	13.20	224.00	1	HH	1	T3	1	NA	1	0	0	na
20.15a	10.70	226.00	1	HH	0	T2	1	SNAG	1	5	1	8.7
20.15b	10.70	226.00	1	HH	0	T2	0	SNAG	1	2	1	5
20.15c	10.70	226.00	1	HH	0	T2	1	SNAG	1	5	1	8
20.16a	7.95	231.00	1	HM	1	T3	1	SNAG	1	0	0	na
20.16b	7.95	231.00	1	HM	1	T3	1	SNAG	1	0	0	na

20.16c	7.95	231.00	1	HM	1	T3	1	SNAG	1	0	0	na
21.01a	7.20	120.00	0	UB	1	T0	0	NA	1	1	0	na
21.01b	7.20	120.00	0	UB	1	T0	0	NA	1	1	0	na
21.01c	7.20	120.00	0	UB	1	T0	0	NA	1	0	0	na
21.02a	15.32	221.00	1	HM	1	T0	0	SNAG	1	1	0	na
21.02b	15.32	221.00	1	HM	1	T0	0	SNAG	1	1	0	na
21.02c	15.32	221.00	1	HM	1	T0	0	SNAG	1	1	0	na
21.03a	17.00	233.00	1	HM	1	T2	1	SNAG	1	0	0	na
21.03b	17.00	233.00	1	HM	1	T2	1	SNAG	1	0	0	na
21.03c	17.00	233.00	1	HM	1	T2	0	SNAG	1	0	0	na
21.04a	4.44	280.00	0	UB	1	T0	0	NA	1	3	1	10.5
21.04b	4.44	280.00	0	UB	1	T0	0	NA	1	3	1	8
21.04c	4.44	280.00	0	UB	1	T0	0	NA	1	0	0	na
21.05a	7.90	288.00	0	UB	0	T0	0	NA	1	3	1	8.5
21.05b	7.90	288.00	0	UB	0	T0	0	NA	1	0	0	na
21.05c	7.90	288.00	0	UB	0	T0	0	NA	1	2	1	6
21.06a	11.09	312.00	0	UB	1	T0	0	NA	1	4	1	11.5
21.06b	11.09	312.00	0	UB	1	T0	0	NA	1	3	1	8.5
21.06c	11.09	312.00	0	UB	1	T0	0	NA	1	2	1	4.5
21.07a	9.40	8.00	1	HL	1	T0	0	NA	1	1	0	na
21.07b	9.40	8.00	1	HL	1	T0	0	NA	1	1	0	na
21.07c	9.40	8.00	1	HL	1	T0	0	NA	1	1	0	na
21.08a	17.94	325.00	1	M	1	T0	0	NA	1	3	1	6
21.08b	17.94	325.00	1	M	1	T0	0	NA	1	0	0	na
21.08c	17.94	325.00	1	M	1	T0	0	NA	1	0	0	na