RESPONSE OF IMPROVED WESTERN LARCH SEEDLINGS TO DROUGHT STRESS AND SITE QUALITY

A Thesis
Presented in Partial Fulfillment of the Requirements for the
Degree of Master of Science
with a
Major in Natural Resources
in the
College of Graduate Studies
University of Idaho

By
Kelsie M. Grover

Major Professor: Andrew Nelson, Ph.D.
Committee Members: Mark Coleman, Ph.D., Dan Johnson, Ph.D.
Department Administrator: P. Charles Goebel, Ph.D.

May 2019

AUTHORIZATION TO SUBMIT THESIS

This thesis of Kelsie Grover, submitted for the degree of Master of Science with a Major in Natural Resources and titled "RESPONSE OF IMPROVED WESTERN LARCH SEEDLINGS TO DROUGHT STRESS AND SITE QUALITY," has been reviewed in final form. Permission, as indicated by the signatures and dates given below, is now granted to submit final copies to the College of Graduate Studies for approval.

Major Professor: ______________________________ Date: ________________
Andrew Nelson, Ph.D.

Committee Members: ______________________________ Date: ________________
Mark Coleman, Ph.D.

______________________________ Date: ________________
Dan Johnson, Ph.D.

Department Administrator: ______________________________ Date: ________________
P. Charles Goebel, Ph.D.

ABSTRACT

Tree improvement remains a valuable and effective way to increase growth and yield in commercially valuable timber species. Significant results have been found in the manipulation of genetics for *Pseudotsuga menziesii* in the northwestern U.S, *Pinus taeda* in the southeastern U.S, *Pinus raidata* in New Zealand, and *Pinus sylvestris* L. in Europe. Unfortunately, it still remains unclear how western larch performs in genetics trials. Endemic to the Inland northwest and valued primarily for its rapid growth and superior wood quality, western larch is widely planted and managed with intensive silvicultural regimes. In an effort to gain a better understanding of the performance of improved western larch in the field, members of the Inland Empire Tree Improvement Cooperative (IETIC) established a western larch seed orchard in 2007 with families from superior-rated trees. The orchard serves to produce open-pollinated seed for reforestation of cooperative member lands and identify desirable candidates for continued breeding. Seed from seven individual half-sib families in the orchard, selected across a range of regional climates, was collected in 2016. One open-pollinated bulk orchard lot was also collected for this study. Seed was sent to the University of Idaho Pitkin Forest Nursery in 2017 where it was grown and then out-planted the following year for field testing against various site climates and qualities. In 2018, individual families were grown from seed in a greenhouse study to examine genetic by soil moisture effects on seedling morphology and physiology.

The greenhouse drought study was arranged as a Complete Randomized Block Design, with four treatments of increasing drought severity including a well-watered control. Seedlings were germinated in germ cloth, transplanted into D25L Deepots, and then well-watered until secondary leaves flushed. From second leaf flush, the drought trial was applied for 10 total weeks. Seedlings were held at specific gravimetric water contents based on a moisture release curve for the growing media. Three rounds of gas exchange and destructive sampling were conducted in order to obtain photosynthesis, transpiration, specific leaf area, above ground biomass, belowground biomass, and root mass fraction. Height and stem diameter were measured weekly. Results from this study revealed a family and treatment effect on all morphological features, indicating superiority of certain families for drought tolerance during early seedling development. Aboveground development, in mass, height, and stem diameter, and belowground root development were reduced with increased drought intensity. Physiology followed common trends with decreasing photosynthetic and transpiration rates with increased drought severity. Specific leaf area also decreased with increasing drought intensity, indicating increased tissue density. Overall, there was no mortality which could be based on various factors, one of which is high overall water use efficiency in improved stock. Family differences in the morphology indicated some level of drought tolerance within certain seed lots, which could prove advantageous for out-planting performance at drier sites.

The field study was arranged as a Random Complete Block Design, ranging across three different sites each belonging to members of the IETIC and the Intermountain Forestry Cooperative. Each site was selected based on varying soil parent material and moisture regimes. Data collected from the out-planting trial included seedling height, root collar diameter, soil volumetric water content, soil bulk density, soil chemistry, competing vegetation cover, vertical vegetation next to seedlings, and slash loading. Site, soil moisture, and competing vegetation were the most significant factors effecting height growth. There was no family effect on height or diameter. Seedlings at the drier site, Stimson, outperformed seedlings at the wetter site, Molpus, however, Molpus was also the last to be planted and had lower June through August average air temperatures. Total forbs had a negative correlation to height growth as seedlings competed for soil moisture and nutrients. Height of nearby competing vegetation had a positive correlation with seedling height growth. The results from this experiment should be considered short-term findings, however, as it stands they indicated that there is no superiority between families and that half-sib breeding control does not outperform an open-pollinated bulk lot based on site conditions.

ACKNOWLEDGEMENTS

I am thankful to have had the chance to complete a graduate degree at the University of Idaho. I would like to give special thanks to my major advisor Andrew Nelson for giving me the opportunity to further my knowledge in the field of forestry. His careful guidance and support has been invaluable. Also special thanks to my committee members Dan Johnson and Mark Coleman, who provided me with equipment and guidance needed to use those tools in the field, as well as valuable insight into the implications of my project.

Special thanks to the National Science Foundation-Center for Advanced Forestry Systems for funding my project, Inland Empire Tree Improvement Cooperative members and Intermountain Forestry Cooperative members for resources, Pitkin Forest Nursery and Center for Forest Nursery and Seedling Research. I would like to thank Don Regan and the nursery staff at Pitkin Forest Nursery for assuring the seedlings for my study were grown successfully and taken care of. I would also like to thank Stimson Lumber Company, Molpus Timberlands Management, Llc, and PotlatchDeltic for providing the field sites for this study and for coordinating plantings crews.

Lori Mackey, Lauren Goss, Jon Cherico, Gus Reely, and many others provided valuable assistance to my project in some way from collecting data to site installment. Without the help of these people and countless others this project would have been completely unmanageable. This has been the most immersive and comprehensive piece of work I have ever completed and I am looking forward to continuing my career in forestry.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

CHAPTER 1: ROLE OF TREE IMPROVEMENT FOR INCREASING FOREST PRODUCTIVITY AND ADAPTATION

1.1. History of tree improvement

Tree improvement gained popularity in the United States in the early 1900s, following the success of coastal Douglas-fir (*Pseudotsuga menziesii*) genetic trials (Wheeler et al 2015). Initial interest in forest genetics was prompted by a combination of the rapid decline of available commercial timber in the U.S. and by the rediscovery of Gregor Mendel's research pertaining to heredity in plant breeding (Perkins 1997; Prudham 2003). The concept of provenance and geographic constraints on Douglas-fir seed sources along the Pacific Coast was first introduced by U.S. Forest Service scientist Thornton Munger in 1913 (Munger & Morris 1936; Bordelon 1988; Prudham 2003). Soon after, foresters began implementing hybridization trials in poplar and pine, and by the 1980's significant amounts of federal grant money was being directed to forest genetics and to the hiring of specialized genetics faculty in universities (Wheeler et al 2015).

Coordination between industry and university in the 1950's gave rise to cooperative tree improvement programs which aimed to utilize the collaboration to further genetics research and guide breeding programs (Wheeler et al 2015). Cooperators provided parent tree selection, progeny testing, and seed from seed orchards, while university personnel conducted field trials to assess clone performance, analyzed the data, and provided guidance for tree breeding strategies (Cooperative Forest Genetics Research Program 2010). Some of the earliest programs included the Western Gulf Forest Tree Improvement Program at Texas A&M University, the Cooperative Forest Genetics Research Program at University of Florida, and the North Carolina

State University-Cooperative Tree Improvement all of whom focused their research efforts on loblolly (*Pinus taeda*) and slash pine (*Pinus elliottii*). Early established programs in the Pacific Northwest (PNW) included the Northwest Tree Improvement Cooperative at Oregon State University, and the Inland Empire Tree Improvement Cooperative at the University of Idaho which focused on Douglas-fir, western white pine (*Pinus monticola*), and western larch (*Larix occidentalis*) (Wheeler et al 2015).

Tree improvement is based on the process of selecting individuals of a species located within a geographic region that display desirable characteristics such as increased wood volume productivity and quality, and disease resistance (Ruotsalainen 2014). The process of tree breeding begins by selecting phenotypically favorable parent trees, also called plus trees, from either a natural forest environment or from a plantation (Ruotsalainen 2014). After which, sexually mature tree limbs, called scions, are collected from the plus tree, grafted onto root stock and out-planted for field testing (Zobel & Talbert, 1984). Breeding values for the plus trees are evaluated by the performance of first-generation progenies produced by the grafted scions, and will indicate potential candidates for continued second generation progeny testing and scion collection (Fig 1.1) (Ruotsalainen 2014). Multiple generations of progeny testing provides geneticists with an indication of rotation end production traits and for candidacy into seed orchards (Andersson, Gull & Mullin 2013). Once genetic material has been integrated into a seed orchard, breeding can be controlled to varying levels of severity, ranging from the highest level of control (full sibling), partial control (halfsibling), or the least control open-pollinated. Full siblings will have a known father and mother tree, while half-siblings know only the mother tree (Zobel & Talbert, 1984). Open-pollinated families, also known as bulk orchard lots, are comprised of a random combination of seed with unknown mother and father trees from within the orchard (Zobel & Talbert, 1984).

One aspect of tree breeding that has been thoroughly researched has been the genotype by environment interaction. The genotype-environment (GE) interaction is the effect of environmental components such as soil, climate, nutrients, photoperiod, and competition, on the expression of a tree’s genotypic performance (Shelbourne & Campbell 1976). Common garden experiments are traditionally used to assess interactions and the results of heterogeneous variances or rank changes within genotypes (Ukrainetz et al 2018). Heterogeneous variances are found when there are significant family differences between two planting environments (Ukrainetz et al 2018). Ranks are assigned to families based on their overall genetic superiority expressed over the life of a crop (Ukrainetz et al 2018; Shelbourne & Campbell 1976). Campbell (1992) assessed the effects of GE interactions on genetic-gains of coastal Douglas-fir in Oregon and found significant interaction effects and heavy shifts in rankings between planting sites. Ultimately, tree breeders are looking for a high ranking genotype within an improvement program that performs well across a variety of site conditions, regardless of GE effects.

1.1.1. Economics of tree improvement

Many factors need to be addressed when calculating potential economic gains within a tree breeding program. Balancing seed yield with plantation size, and level of selection intensity directly affect the productivity of a program (Ledig & Porterfield, 1982). Selection intensity is a unit of measure based on the phenotypic standard deviation of the selected superior parent trees in relation to the population mean (Marshall 2006). Selection intensity decreases when a larger proportion of the population is used for genetic material, thus a higher selection intensity indicates a smaller range of genotypic diversity (Marshall 2006). Although it is more costly to select from a larger population, it is generally undesirable to have a high selection intensity in a breeding program. Ledig and Porterfield (1982) suggested using a lower selection intensity in order to reduce biological risk of decreasing genetic diversity within the orchard. They also recommend that orchards produce enough seed to support an entire plantation, while not overproducing seed that cannot be used.

Economic gains have been found within various North American species such as Douglas-fir, loblolly pine, western white pine, and red pine. Collective results from provenance-progeny expected yield studies indicate juvenile height growth gains of approximately 14.7% at 15 years for black spruce, 7.4% at 21 years for jack pine, 12.4% at 20 years for white spruce, and 8.0% at 15 years for red pine (Newton 2003). There are issues of climate and geographic restrictions for long-rotation western conifers that do not apply to short-rotation conifers in the south.

Long-term yield results for first year provenance-progeny experiments are still limited in western larch. In order to estimate performance of improved western larch plantations, models have been used to predict growth and yield under specific management regimes. Moore and Fins (1984) determined theoretical biological yields from genetically improved and unimproved stock of western larch using a simulation model called the Prognosis Model for Stand Development (Stage 1973; Wykoff et al. 1982). They used the subroutine called CHEAPO (Medema and Hatch, 1979) in conjunction with Prognosis, and accounted for costs associated with a tree improvement program, as well as depreciating or appreciating costs and prices for discount rates and stumpage rates. Results indicated that profitability of a western larch breeding program was guaranteed as long as seed orchards maintained high rates of seed production and planting programs were large enough to utilize all of the produced seed (Moore & Fins 1984).

1.2. Western larch: silviculture and management

Known for its deciduousness and for being the largest North American larch, western larch (Larix occidentalis) is coveted by timber companies for its high-quality wood and rapid early growth (Schmidt et al 1976). Western larch has a high resin content and dense, decay resistant wood make this species desirable for flooring, cabinetry, railroad ties, and utility poles (Schmidt & Shearer 1995; Sibley 2009). The wood also contains a usable percentage of arabinogalactan, a water soluble gum used in paint products, ink, food, and for pharmaceuticals (Boe 1958). High heating value makes the wood from this species the best for fuel in the northwestern U.S. (Scher 2002).

Often intermixing with other conifer species, western larch very rarely occurs in pure stands unless artificially planted. Western larch is highly shade-intolerant and can be relatively selective in regards to growing conditions. Exclusive to the Inland Northwest and Canada, western larch is typically found in montane valleys along a northern or eastern aspect, within an elevation range of 2,000 to 5,000 feet in its northern range (Schmidt & Shearer, 1995), and up to 7,000 feet in its southern range of central Idaho and north-central Oregon (Boe 1958). Temperature limits the geographic and elevation distribution of western larch, particularly because early bud break makes this species more susceptible to frost damage (Schmidt et al 1976).

Western larch are characterized by their distinct straight trunks and short pyramidal shape crowns, often growing anywhere from 30-55 m tall to over 60 m in some cases (Schmidt et al 1976). Foliage is bright green with clusters of soft needles longer than those found on American larch (*Larix laricina*) (Sibley 2009). Cones are small and positioned upright, and mature in a single growing season (Sibley 2009). Unfortunately, cone crops are sporadic and can produce few seeds, which is why seed orchards and nursery cultivation are heavily relied on for reforestation (Schmidt & Shearer 1991). During good seed production years, natural germination rates occur best on microsites of exposed mineral soils or recently burned soils, with low vegetation competition (Boe 1958). Cutting methods that remove most or all overhead vegetation are favored for natural regeneration, including seed-tree, shelter-wood, or clear-cutting (Boe 1958). The two highest causes of first-year seedling mortality are fungi and insolation, with the latter only being a factor in full-sun or partial-sun areas, followed by drought as the next leading abiotic cause of mortality (Schmidt & Shearer 1991). Larch Dwarf Mistletoe (Arceuthobium laricis) infections can substantially affect western larch in the Inland Northwest. About 47% of western larch stands in Oregon and Washington have been infected, 30% of the larch stands in Idaho, and about 50% in Montana (Bolsinger 1978; Mathiasen 1998; Drummond 1982). Removal of infected overstory larch before new regeneration reaches seven years old or 0.9 m tall is the recommended management practice.

Since western larch are deciduous, their physiologic response is different from other conifer species. According to a study conducted by Rosenthal and Camm (1996), chloroplasts continue to function until the final stages of senescence, which they characterized by a decline in the chlorophyll a/b ratio and decreased carbon dioxide assimilation. Initial chlorophyll decline was triggered by decreased air temperature, irradiance, and photoperiod, as well as an increase in the relative humidity. Stomatal conductance was found to be unaffected by photoperiod and soil temperature but was more correlated with irradiance levels and differences in leaf-to-air vapor pressure (Benecke et al. 1981, Sandford and Jarvis 1986, Matyssek and Schulze 1988). Western larch has also been shown to have a relatively high resistance to cavitation under a water deficit, meaning it is fairly drought tolerant compared to other species like ponderosa pine, Douglas-fir, and western red cedar (Pinol & Sala 2000).

Western larch is a prime candidate for tree improvement programs, coupled with the fact that it is already heavily utilized within commercial practices and that it has superior overall growth. Aside from inconsistent cone crops and minor pest issues, western larch is a hardy species that grows best following common timber harvest practices. Climatic restrictions may prove to be the most influential dictator of improved seed lot performance, given western larch's sensitivity to colder temperatures and frost. In which case, clonal trials might help determine if specific seed lots can be isolated and bred for planting in less desirable sites. The long-term goal of this study is to determine exactly that, whether specific seed lots significantly differ by site climate and quality.

Figures

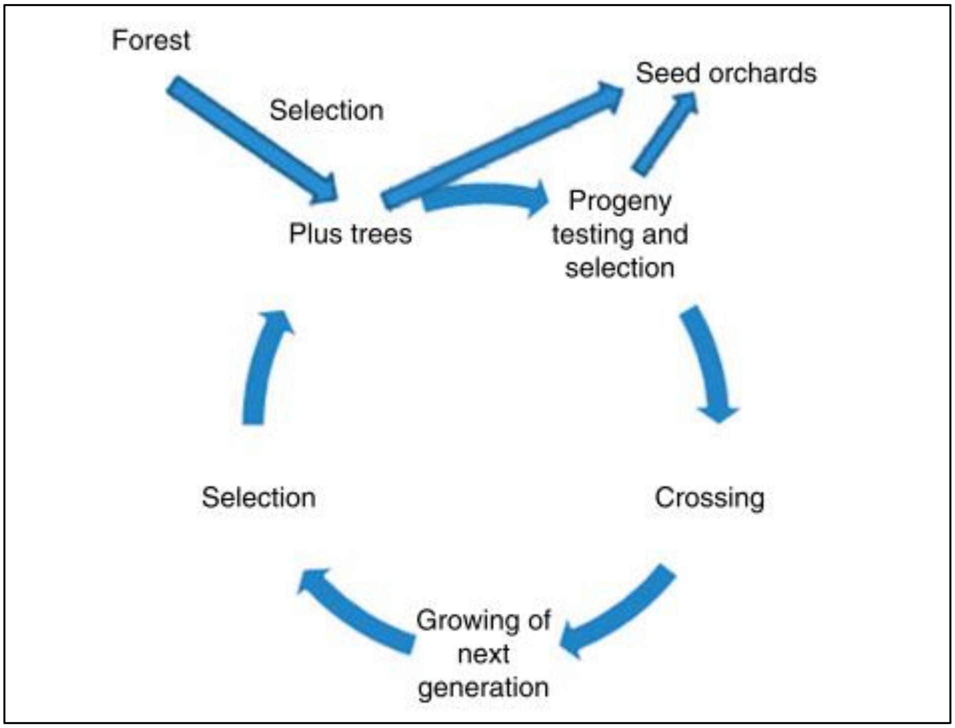

Figure 1.1. Process of testing and selection of seed orchard genetic material from plus tree selection to orchard integration (*From:* Ruotsalainen 2014).

References

Benecke, U., Schulze, E. D., Matyssek, R., and Havranek, W.M. 1981. Environmental control of CO2-assimilation and leaf conductance in Larix decidua Mill. Oecologia 50: 54-61.

Boe, K. N. 1958. June 1958 silvics of western larch. Forest Service U.S. Department of Agriculture Reed W. Bailey, Director (16).

Bolsinger, C. L. 1978. The extent of dwarf mistletoe in six principal soft- woods in California, Oregon, and Washington, as determined from forest survey records. Proceedings of the symposium on dwarf mistletoe control though forest management. USDA Forest Service, General Technical Report. PSW-31. 190.

Bordelon, M. A. 1988. Genetic improvement opportunities. In Assessment of Oregon's forests 1988, ed. G. J. Lettman, 231–39. Salem: Oregon State Department of Forestry.

Campbell, R. K. 1992. Genotype * environment interaction - a case study for Douglas-Fir in western Oregon. USDA Forest Service Pacific Northwest Research Station Research Paper. (455): 1.

Cooperative Forest Genetics Research Program. 2010. University of Florida, School of Forest Resources and Conservation. http://www.sfrc.ufl.edu/cfgrp/logIn/logIn.shtml#research

Drummond, D. B. 1982. Timber loss estimates for the coniferous forests of the United States due to dwarf mistletoes. USDA Forest Service, Forest Pest Management, Methods Application Group, No. 83-2.

Moore, J. A., & Fins, L. 1984. Economic Analysis of a Tree Improvement Program for Western Larch Tree. Journal of Forestry, (November). 675–679.

Ledig, F. T. and Porterfield, R. L. 1982. Tree improvement in western conifers: Economic aspects. Society of American Foresters. Journal of Forestry 80(10): 653-657.

Marshall, K (2006). Response to selection I. IAEA Regional training course on selective breeding and gene technologies, Korea.

Mathiasen, R. L. 1998. Infection of young western larch by larch dwarf mistletoe in Northern Idaho and Western Montana. Western Journal of Applied Forestry, 13(2): 41-46.

Matyssek, R., and Schulze, E. D. (1988). Carbon uptake and respiration in above-ground parts of a *Larix decidua* × *leptolepis* tree. Trees. 2. 233-241.

Medema, E.L., and Hatch, C.R. 1979. Computerized help for the economic analysis of prognosis-model outputs (CHEAPO). Research report pre- pared for the Intermountain Forest and Range Experimental Station, Ogden, UT. Prepared by the College of Forestry, Wildland, and Range Sciences, University of Idaho, Moscow. 71.

Munger, T. T., Morris, W.G. 1936. Growth of Douglas-fir trees of known seed source. Technical Report no. 37. Portland, OR: Pacific Northwest Forest Experiment Station, U.S. Department of Agriculture.

Newton, P. F. 2003. Systematic review of yield responses of four North American conifers to forest tree improvement practices, Forest Ecology and Management 172(1): 29-51.

Perkins, J. H. 1997. Geopolitics and the green revolution: Wheat, genes, and the Cold War. New York: Oxford University Press.

Piñol, J., & Sala, A. 2000. Ecological implications of xylem cavitation for several *Pinaceae* in the Pacific Northern USA. Functional Ecology, 14(5): 538–545.

Prudham, S. 2003. Taming trees: Capital, science, and nature in pacific slope tree improvement. Annals of the Association of American Geographers, 93(3): 636–656.

Rosenthal, S. I., and Camm, E. L. 1997. Photosynthetic decline and pigment loss during autumn foliar senescence in western larch (Larix occidentalis). Tree Physiology, 17(12): 767-775.

Ruotsalainen, S. 2014. Increased forest production through forest tree breeding. Scandinavian Journal of Forest Research, 29(4): 333–344.

Sandford, A.P., and Jarvis, P.G. 1986. Stomatal responses to humidity in selected conifers. Tree Physiology 2. 89-103.

Scher, Janette S. 2002. Larix occidentalis. In: Fire Effects Information System. U.S. Department of Agriculture, Forest Service, Rocky Mountain Research Station, Fire Sciences Laboratory.

Schmidt, W.C. & Shearer, R.C. 1991. Larix Occidentalis Nutt. In R. M. Burns & B. H. Honkala (Ed.), Silvics of North America, Washington: U.S. Department of Agriculture, Forest Service. 160-170.

Schmidt, W.C. and Shearer, R.C. 1995. *Larix occidentalis*: A pioneer of the North American

West. Ecology and management of Larix forests: A look ahead. Symposium Proceedings. General Technical Report. GTR-INT-319. Ogden, UT. USDA Forest Service Intermountain Research Station. 33–37.

Schmidt, W. C., Shearer, R. C., Roe, A. L. 1976. Ecology and silviculture of western larch forests. U.S. Department of Agriculture, Technical Report 1520. Washington, DC. 96.

Shelbourne, C. J. A., & Campbell, R. K. 1976. The impact of genotype-environment interactions on tree improvement strategy. In: Proceedings of the IUFRO, Joint Meeting on Advanced Generation Breeding, Bordeaux, June 14 - 18, 1976, 73–93.

Sibley, D. A. 2009. The Sibley guide to trees. Alfred A. Knopf. New York

Stage, A. R. 1973. Prognosis model for stand development. USDA Forest Service Res. Paper INT-137. 32.

Ukrainetz, N. K., Yanchuk, A. D., & Mansfield, S. D. 2018. Climatic drivers of genotype–environment interactions in lodgepole pine based on multi-environment trial data and a factor analytic model of additive covariance. Canadian Journal of Forest Research, 48(7): 835–854.

Wheeler, N. C., Steiner, K. C., Schlarbaum, S. E., & Neale, D. B. 2015. The Evolution of Forest Genetics and Tree Improvement Research in the United States. Journal of Forestry, 113(5): 500–510.

Wykoff, W.R., Crookston, N.L., and Stage, A.R. 1982. User's guide to the stand prognosis model. USDA Forest Service General Technical Report INT-133. 112.

Zobel, B. and Talbert, J. 1984. Applied forest tree improvement. Wiley. New York

CHAPTER 2: FIRST YEAR IMPROVED WESTERN LARCH SEEDLING RESPONSE TO SIMULATED DROUGHT

2.1. Abstract

Moisture stress can significantly impact first year seedling morphology and physiology. Under optimal conditions, seedlings will allocate energy to stem height growth and root development, while stressed seedlings may exhibit drastic departures from growth and physiology. The goal of this study was to subject seven half-sibling improved western larch (*Larix occidentalis*) families to four levels of moisture stress during the first year of development from seed and monitor the growth and physiology in a complete randomized block design. Moisture stress treatments were based on the specific water holding capacity of the growing medium and expressed as volumetric water content (VMC): 25% VWC, 15%, 10%, and 5% respectively. Treatments began 4 weeks after transplanting and continued for 10 weeks. Total height and root collar diameter were measured weekly for each individual seedling. Blocks were randomly selected for destructive sampling at weeks 6, 8, and 10, from which photosynthesis, transpiration, specific leaf area, root mass fraction, aboveground and below ground biomass were measured. Statistical analysis indicated that a drought intensity by week effect was significant for all growth variables. Family and the drought intensity by family interaction effects were significant for all morphological variables but not for physiological variables. Drought intensity was significant to all morphological and physiological variables except specific leaf area. All of these factors had a negative correlation to drought severity except for specific leaf area and root mass fraction. This indicates that moisture stress has a significant impact on seedling development and is an extremely limiting factor for first year

seedlings. Genotype showed a strong response to moisture stress which implies higher tolerance to drought in certain families over others.

2.2. Introduction

Water is a critical resource that plants require to function, yet less than one percent of water taken up by a plant is retained as biomass (Lambers et al. 2008). The majority of water that plants use is released back to the atmosphere through various physiological functions. Plants pull water through the roots, upward through the stem, and into the leaves via a continuous negative pressure chain. Moisture within the leaves is then released as water vapor through the process of transpiration which is regulated by small pores called stomata. Trees manipulate the opening of stomata in response to water availability to limit water loss when atmospheric demands are high or soil moisture supply is low, a process that also impacts carbon assimilation (Reddy et al 2004). Trees also adjust morphological growth when faced with prolonged water deficits, which can be expressed as decreased height growth, early bud set, or increased specific leaf area (Okali & Dodoo 1973). First year seedlings are especially vulnerable to environmental stress and can easily be killed by insolation incurred by extreme microclimate conditions, and seasonal drought (Haig et al 1941).

One way researchers have been able to test seedling response to drought is by manipulating moisture availability in a controlled greenhouse setting. By controlling soil moisture, researchers can test genotypes for variability in drought response to determine whether certain seed sources have higher tolerance to moisture deficits and how they respond morphologically (McIver et al 2014). Vance and Running (1985) used varying levels of moisture stress and light reduction on nursery grown western larch seedlings to examine their effects on biomass, height, and root collar diameter (RCD). They found western larch was highly responsive to photoperiod duration and moisture availability which controlled timing of when active shoot growth ceased. Greenhouse studies have also been used to assess genotype by drought interactions. Ponton et al (2002) tested two *Quercus* species against a well-watered and severe drought regime and varying levels of irradiance, and found no significant genotype by environment interaction in response to the drought. However, they concluded that initial growth rates between the two species had an inverse relationship with long-term drought survival, and that seedlings with slower initial growth rates were better adapted for drought and had higher water use efficiency overall.

Microsite can be critical for protecting seedlings from drought during the first year of development. The concept of a safe site was proposed by Harper et al. (1961) as the area around a seedling that has the potential to impact growth initiation, germination, and protection from predators and harsh environmental conditions. A safe site can exist within a range of microsite conditions created by surrounding vegetation, rocks, and residual slash for forest operations (Oswald & Neuenschwander 1993). Microsite conditions can also be defined by unique soil conditions like temperature, moisture, and nutrient availability (Beatty and Stone 1986). Variations in microsites are therefore extremely effective at impacting seedling growth as they are unique mini-climates existing at the individual seedling level. One microsite could allow for significantly warmer, wetter soil than another even though they are separated by a short distance.

Western larch prefers specific site conditions for successful germination. Natural germination of western larch is reported to begin when soil temperatures reach 16 ^{o}C and will increase rapidly when the soil climbs to 21-27 ^{o}C (Boe 1958). Although soil temperature is important for germination and early development, insolation is ranked as the major cause of death in first year seedlings. Therefore western larch is almost never planted on a south aspect, due to poor survival. Insolation can still remain a problem for seedlings in full-sun exposure on a north aspect. The second leading cause of mortality among first year seedlings is drought, followed by fungal diseases (Haig 1936). Despite this, western larch is reported to germinate under a wide variety of site conditions, preferring ashy, bare mineral soils and a north or east aspect (Fisher 1935; Oswald & Neuenschwander 1993; Boe 1958).

Root development can aid in seedling survival during early establishment. In the region where western larch is native, soil moisture is plentiful immediately after germination through the early summer, followed by a 2-3 month summer drought. It is critical for seedlings to develop elongated roots that penetrate to the lower soil profile to withstand this drought. Western larch is typically a deeper-rooted species, lending them an advantage toward drought tolerance if they can favorably develop deep roots within the first year (Haig 1936). Higher root density has been positively correlated to moisture availability in western larch (Aghai et al 2014). This can create an urgent situation as seedlings need to establish roots while moisture is available, but they need a strong root system to withstand drought conditions.

The goal of this study was to examine first-year genotypic response of seven improved western larch families across a gradient of drought severity. Specific objectives included examining effects of drought severity, family, and time since treatment on: (1) height, diameter, and biomass allocation, and (2) leaf-level gas exchange including photosynthesis and transpiration.

2.3. Materials and methods

Members of the Inland Empire Tree Improvement Cooperative (IETIC) established a western larch seed orchard in 2007 in Kalamalka, British Columbia with families from superior-rated trees throughout eastern Washington and northern Idaho. For this investigation, seed was collected from seven mother trees in the orchard that produced high cone yields in 2016 and spanned a geographic and climatic gradient in the region (Table 2.1). Cones from each mother tree were processed separately at the British Columbia Tree Seed Centre yielding seven half-sib families. The seed was sent to the University of Idaho Franklin H. Pitkin Forest Nursery in spring 2017, where it was stratified in a cooler for 30 days and then rinsed for 48 hours. The rinsed seed was germinated in damp cloth in clear trays where it was misted three times a day with distilled water for seven days until germination. Germinates were transplanted into pots filled with soil medium while the root radical was a centimeter or less in length. Three germinates per cell were transplanted into D25L Deepots® (Stuewe and Sons, Inc. Tangent, OR) containers (410 ml) filled with SunGro forestry mix #1 professional horticulture growing media comprising of 40-50% Canadian sphagnum peat, 30-40% vermiculite, 10-20% bark, 4.5-5 pH, and 70-160 EC (Sun Gro Horticulture, MA). Containers were placed in trays with a capacity of 50 cells.

The average volume of medium used per container was 409.6 cm^3. Within each container 1.2 g of Osmocote Smart-Release® Plant Food Plus Outdoor & Indoor slow release fertilizer (15-9-12 + micronutrients) (The Scotts Company, Marysville, OH) was incorporated into the medium before transplanting based on the manufacturer's recommended rate. Dry media weight per container was 48.2 g and the weight at field capacity was ~325 g. Field capacity was determined by fully saturating the media until water drained from the bottom of the pot then weighing the pot after it drained for one hour. Germinates were covered in 13 g of grit following transplanting and then moved into a greenhouse at the Pitkin Nursery when max air temperate was 26.7^{o}C, and minimum air temperature was 15^{o}C. A moisture release curve for SunGro professional horticulture mix was created using ECH$_2$O EC-5 volumetric soil moisture probe (ECM-5) and a MPS-6 calibrated water potential sensor (Meter Group, Pullman, WA) (Fig. 2.1). The purpose of creating the moisture release curve was to determine the drought stress treatments based on actual water holding capacity of this specific growing media. Gravimetric water content was measured by subtracting the dry media weight from the wet media weight and then dividing by the dry media weight.

Transplants received three passes with an overhead boom biweekly for four weeks until the stems turned woody and secondary leaves appeared. Experimental drought treatments were applied from this point forward for 10 weeks. Seedlings were weighed once to twice per week to keep track of gravimetric moisture. If the gravimetric moisture was below the target for the treatment, water was added with a Wheaton 374301 Unispense® peristaltic pump liquid dispenser to bring the moisture back up to the target weight. Trays were kept on a table in an enclosed greenhouse at the University of Idaho Franklin H. Pitkin Forest Nursery.

2.3.1. Experimental design

The design for this experiment was a Complete Randomized Block Design, with seven blocks (individual tray) (b = 7), four drought treatments (t = 4, control [25% volumetric water content (VWC)], low [15% VWC], moderate [10% VWC], and high [5% VWC]), and seven half-sib families. Soil VWC (W_v) was calculated using the gravimetric water content (GWC) (W_d), which was obtained by calculating *Container weight with wet soil (g) – Empty container weight (g) = Wet soil weight (g)*. Which was then used to calculate the W_d with equation: *(Wet* *soil weight [g] – Dry soil weight [g]) / Dry soil weight (g) = GWC* (W_d). Soil bulk density for the growing media needed to be calculated using the equation: *Bulk density* (g/cm^3) *= Dry soil weight (g) / Soil volume* (cm^3). Then the W_v could be calculated: W_d ** Soil bulk density* (g/cm^3) $= W_v$.

Each block contained one replicate of each drought × family combination randomly assigned to a cell location. Trays were spread in a single row across a greenhouse table from the sidewall of the greenhouse to the center walkway. Levels of drought stress were determined based on points along the previously mentioned moisture release curve for SunGro professional horticulture mix. The well-watered treatment was selected near the field capacity GWC point on the graph and the severe drought was selected at the point where the curve sharply plateaued.

2.3.2. Data collection

Total height (cm) and stem diameter (mm) were measured once a week for each seedling using a measuring tape and caliper. At week 6, two random blocks were selected and measured for leaf-level gas exchange followed by destructive harvest to measure shoot and root biomass. Two additional blocks were sampled at week 8 for the same measurement and the remaining three blocks were measured at week 10.

Leaf-level gas exchange was measured using a LI-6400XT® portable photosynthesis system (LI-COR Biosciences, Lincoln, NE) with a 6400-22L lighted conifer chamber head. The leaf temperature within the chamber head was set to 25 ^{o}C and the flow rate set to 400 µmol s^{-1}. The flow meter and the CO_2 mixer were calibrated with the scrub and desiccant set to full scrub and then the ParIn and IRGA's were zeroed. The CO_2 reference was set to 400 ppm, the lamp set to 1200 µmol m^{-2} s^{-1} photosynthetically-active radiation, and the desiccant valve turned to bypass to maintain the empty chamber-reference relative humidity at 30%. Ambient incoming light within the greenhouse at the time of measurement was measured using a PAR sensor attached to the chamber head. The terminal top of each seedling was placed into the chamber head and left undisturbed for a minimum of four minutes or until photosynthesis and stomatal conductance measurements stabilized before recording the measurement. The IRGA's were matched after every two seedlings to reduce shifts in measurements.

To correct gas exchange measurements for the actual leaf area within the LI-6400 XT chamber, foliage within the chamber was removed and placed in a plastic bag and then put in a freezer. Frozen needles were analyzed for area with WinSEEDLE^{TM} and a STD4800 flat-bed scanner (Regent Instruments Inc., Quebec, Canada). Needles were placed within a clear plastic tray ensuring no overlap and then scanned at 800 DPI and analyzed for projected (one-sided) leaf area using the dual-lighted option to minimize needle shadow. The leaf area values were adjusted in the exported LI-COR file so that true gas exchange measurements could be obtained. Needles used for leaf area calculations were placed in envelopes and put in the oven at 65^{o} C for 48 hours or until constant mass and added to the foliage weight of the entire seedling.

2.3.3. Statistical analysis

Data was analyzed using R software (R Core Team 2013). Using the 'nlme' package (Pinheiro et al 2018), a two-way linear mixed-effects model was used to examine the main effects of drought intensity, week, family and their interaction. Drought intensity and family were considered fixed effects and block within week as a random effect. Separate analysis of variance (ANOVA) models were analyzed using the 'doBy' package (Højsgaard & Halekoh 2018) for height growth, RCD growth, belowground biomass, aboveground biomass, root mass fraction, photosynthesis, transpiration, and specific leaf area (SLA). Week 8 physiology data was removed from the analysis because of errors in the data collection. Linear mixed-effect residual plots were examined for symmetry and heterogeneity. Least-squared means with $\alpha=0.05$ were calculated for all morphological and physiological parameters using the ‘lsmeans’ package (Lenth 2016). The package used for the graphing of the least-squared means was ‘ggplot2’ (Wickham 2016). Package ‘qqplotr’ (Almeida et al 2017) was used to check normality to determine best fit models. Some models had to be adjusted due to heteroscedasticity in the plots. Individual models for belowground biomass and aboveground biomass included the exponent of height as a weighted covariate. The model for root mass fraction included the exponent of RCD as a weighted covariate. Individual models for height, SLA, photosynthesis, transpiration, and RCD did not need adjustment based on residual plots and non-significant model fit tested with likelihood ratio tests (Pinheiro and Bates 2000).

2.4. Results

2.4.1. Absolute height and root collar diameter

Absolute height differed significantly by drought intensity ($p \leq 0.001$), family ($p \leq 0.001$), week ($p \leq 0.001$), drought intensity × week ($p \leq 0.001$), and family × drought intensity ($p \leq 0.001$) (Table 2.2). This means that as drought intensity progressed over an extended period of time, there was a significant impact on height, and that there were also family differences within the drought intensities. Seedling height decreased as drought intensity increased (Fig. 2.2). Height of seedlings in the control averaged 29.7 ± 0.7 cm at week 10, 23.6 ± 0.7cm in the low drought treatment, 14.6 ± 0.7 cm in the moderate drought, and 7.2 ± 0.6 cm in the severe drought. Family 3 had the highest overall average height of 13.2 ± 0.3 cm and family 4 had the lowest average height of 10.6 ± 0.3 cm. Average height across all treatments and families went from 3.5 ± 0.3 cm at week 1 to 18.8 ± 0.4 cm at week 10. Total height decreased across all families as drought severity increased (Fig. 2.3).

RCD significantly differed by drought intensity ($p \leq 0.001$), family ($p \leq 0.001$), week ($p \leq 0.001$), drought intensity × week ($p \leq 0.001$), and family × drought intensity ($p \leq 0.001$) (Table 2.3). Average RCD was 5.11 ± 0.09 mm at week 10 across all families in the control, 4.19 ± 0.09 mm for the low drought, 3.61 ± 0.09 mm for the moderate drought, and 2.19 ± 0.09 mm in the severe drought. Family 2 seedling displayed the highest overall RCD across all treatments with 2.33 ± 0.03 mm, and family 7 had the lowest RCD of 2.09 ± 0.03 mm. Average stem diameter decreased as drought severity increased (Fig. 2.4) and was significantly different between families (Fig. 2.5).

2.4.2. Above and belowground biomass

Aboveground biomass significantly differed by drought intensity ($p \leq 0.001$), family ($p = 0.001$), week ($p = 0.001$), drought intensity × week ($p \leq 0.0001$), drought intensity × family ($p \leq 0.001$), family × week ($p = 0.025$), and drought intensity × family × week ($p \leq 0.001$) (Table 2.4). This was the only measured variable to have a significant three-way interaction, indicating that certain families adjusted their aboveground development differently than others across drought intensity, and that the more prolonged the drought intensities were the more significantly they responded. Average aboveground mass was 2.42 ± 0.24 g for the control at week 10, 1.73 ± 0.14 g in the low drought, 1.00 ± 0.05 g in the moderate drought, 0.38 ± 0.02 g in the severe drought. Average overall aboveground biomass increased from week 6 (0.64 ± 0.02 g) to week 10 (1.38 ± 0.07 g), however, values decreased with increasing drought intensity

(Fig. 2.6). Family 3 had the highest average aboveground biomass (1.06 ± 0.14 g), and family 6 had the lowest (0.93 ± 0.06 g).

Belowground biomass significantly differed by drought intensity ($p \leq 0.001$), family ($p=0.001$), week ($p=0.003$), drought intensity × week (≤ 0.001), and family × week ($p=0.002$) (Table 2.5). Average root mass was 1.31 ± 0.16 g for the control at week 10, 1.09 ± 0.11 g in the low drought, 0.82 ± 0.05 g for the moderate drought, and 0.50 ± 0.03 g for the severe drought. Average belowground biomass increased from week 6 (0.43 ± 0.03 g) to week 10 (0.93 ± 0.05 g), however, values decreased with increasing drought intensity (Fig. 2.7). Family 4 had the highest average root mass (0.76 ± 0.04 g), and family 7 had the lowest (0.61 ± 0.05 g).

Root mass fraction significantly differed by drought intensity ($p \leq 0.001$), family ($p \leq 0.001$), and drought intensity × week ($p \leq 0.001$) (Table 2.6) (Fig. 2.8). Average root mass fraction significantly varied between all treatments within weeks 6, 8, and 10. However, only the differences between weeks 6 and 10 ($p=0.029$), and weeks 6 and 8 ($p=0.038$) within the severe drought were significant. This means that although root mass fraction changed with increasing drought severity, it only significantly changed over time within the most severe drought. Average root mass fraction for the control was 0.22 ± 0.01 g at week 10, 0.38 ± 0.01 g for the low drought, 0.45 ± 0.01 g for the moderate drought, and 0.54 ± 0.01 g for the severe drought. Family 4 and 5 had the highest root mass fractions (0.45 ± 0.01 g), and family 1 had the lowest (0.40 ± 0.01 g). Total dry mass was also significant to root mass fraction ($p=0.023$) and had a positive correlation (Fig. 2.9).

2.4.3. Specific leaf area

SLA significantly differed by week (p=0.022) and treatment × week (p=0.005) (Table 2.7). As drought intensity increased for a longer period of time there was a significant effect on SLA. Average SLA went from 100.3 ± 6.07 g/cm^2 at week 6 to 78.8 ± 6.12 g/cm^2 at week 10 in the control (p=0.640) (Fig. 2.9). Seedlings from the low drought went from 103.7 ± 6.07 g/cm^2 at week 6 to 79.4 ± 5.80 g/cm^2 at week 10 (p=0.847). Seedlings from the moderate drought went from 84.7 ± 6.07 g/cm^2 at week 6 to 76.1 ± 5.49 g/cm^2 at week 10 (p=0.816). Change in SLA was significant in the severe drought from 80.4 ± 6.07 g/cm^2 at week 6 to 88.2 ± 5.31 g/cm^2 at week 10 (p=0.022).

2.4.2. Leaf gas exchange

Photosynthesis significantly differed by drought severity treatment (p≤0.001) and treatment × week (p=0.017) (Table 2.8). Average photosynthesis for week 6 in the control was 11.37 ± 1.30 µmol m^{-2}s^{-1} and increased to 15.29 ± 1.21 µmol m^{-2}s^{-1} by week 10 (p=0.119) (Fig. 2.10). The same trend was observed in the low drought with 10.98 ± 1.30 µmol m^{-2}s^{-1} at week 6, increasing to 12.51 ± 1.26 µmol m^{-2}s^{-1} by week 10 (p=0.958). Photosynthesis began to decrease in the moderate drought from 12.65 ± 1.30 µmol m^{-2}s^{-1} at week 6, to 11.92 ± 1.18 µmol m^{-2}s^{-1} by week 10 (p=1.000), and in the severe drought from 8.33 ± 1.30 µmol m^{-2}s^{-1} to 7.03 ± 1.12 µmol m^{-2}s^{-1}, though the change was not statistically significant (p=0.987).

Transpiration significantly differed by drought severity treatment (p≤0.001) and treatment × week (p=0.022) (Table 2.9). Average transpiration at week 6 in the control was 1.73 ± 0.35 mmol m^{-2} s^{-1}, and increased to 2.95 ± 0.35 mmol m^{-2} s^{-1} at week 10 (p=0.224) (Fig. 2.11). Within the low drought treatment, transpiration increased from 1.63 ± 0.35 mmol m^{-2} s^{-1}

to 2.25 ± 0.31 mmol m^{-2} s^{-1} (p=0.884). Average transpiration increased from 1.62 ± 0.34 mmol m^{-2} s^{-1} to 2.22 ± 0.30 by week 10 for the moderate drought (p=0.884). Average transpiration was lowest in the severe drought and remained consistent from week 6 (1.11 ± 0.33 mmol m^{-2} s^{-1}) to week 10 (1.13 ± 0.28) (p=1.000).

2.5. Discussion

2.5.1. Effects of moisture stress on morphology

As drought severity increased, seedlings either slowed or completely shut down aboveground growth by ceasing terminal elongation. This is a common trend in seedling morphology that height growth is significantly reduced under prolonged moisture deficits (Beckage & Clark 2003). Since treatment, family, and their interaction were significant for both height and stem diameter, it was important to analyze the differences between genotypes. Family 3 seedlings had significantly higher average heights at week 10 for the control, and higher heights in the severe drought as well. Initial well-watered conditions were clearly advantageous for seedlings in family 3 as they displayed rapid early height growth compared to seedlings from families 5, 6, and 7. Seedlings from these families grew slowly even under initial well-watered conditions and sustained a slower growth rate despite being in the control treatment. However, families 5, 6, and 7 also saw less drastic shifts in heights between drought intensities which could be considered an indicator of drought tolerance (Brink & Bruun 2011). Seedlings more adapted for dry conditions have been reported to grow slower despite exposure to wetter conditions (Cregg & Zang 2001). This makes sense as all three families came from mother trees where scion was collected from individuals at lower elevations and higher associated mean maximum temperature. Root collar diameter was not drastically different between the families, however it followed the same trend as height growth. As drought severity increased, average stem diameter decreased. This is a result of the seedlings not being able to allocate energy to diameter growth while under a moisture deficit. Stem diameter is directly correlated with water availability and growth rates have been found to drastically slow or stop completely under drought conditions (Rötzer et al 2017).

2.5.2. Effects of moisture stress on biomass allocation

Seedlings more adapted to drought will allocate growth toward belowground biomass to increase fine root production which maximizes the probability for their roots to locate water throughout the soil profile (Cregg 1994; Haig 1936; Rötzer et al 2017). In fact, limited water supply often encourages seedlings to promote root growth when moisture is a limiting factor (Rötzer et al 2017; Brenchley 1916; Maximov 1929). However, root biomass in a drought stressed seedling will never be greater than in a well-watered seedling as water is still required for overall growth and other physiological activities. Analysis of the data revealed a decrease in biomass with increased drought intensity, particularly a shift from aboveground allocation to belowground growth, however, that trend is slightly misleading. Severe drought seedlings were not decreasing their aboveground biomass per se, but instead were preventing aboveground development entirely while maintaining root development. This is why root mass fraction was used to assess allocation instead of root to shoot ratio, because it is a better method for measuring biomass of each part of the plant relative to the total mass (Poorter & Nagel 2000). Root mass fraction values typically remain higher in water stresses plants compared to well-watered ones, however, Poorter and Nagel (2000) found that changes in allocation are relatively small in response to moisture stress compared to other factors like nutrient deficiencies and irradiance. This could explain why there was only a significant difference in root mass fraction between weeks within the severe drought. Leaf and root development are costly expenditures for seedling development and will only be carried out when resources are available.

Although height and stem diameter do positively impact aboveground biomass, there were significant trends in the tradeoff between stem and foliage mass. Foliage dry mass was far greater than stem dry mass for the majority of families in the control, low drought, moderate drought, and the severe drought in week 6. That trend continued through week 10 where values started to even out between stem and foliage mass in all drought intensities except for the control, where stem mass became greater than foliage mass. This was due to the increase in stem diameter rather than a decrease in foliage production. There are trade-offs associated between productivity and drought tolerance, where productivity is compromised for the sake of increased drought tolerance (Cregg 2001; Long 1980; Teskey et al 1987). Genotypes originating from drier sites typically maintain a predisposition to slower growth even under well-watered conditions (Cregg 2001). This could explain the family differences in biomass allocation, particularly between family 6 and family 3. In week 10, severe drought, family 6 displayed low aboveground biomass (0.31 g), low height (6.3 cm), and low belowground biomass (0.37 g). Meanwhile for the same treatment and week, family 3 had high aboveground biomass (0.43 g), the highest height (8.7 cm), and the highest belowground biomass (0.63 g). Although family 3 seedlings had superior aboveground growth compared to family 6 seedlings, this does not guarantee true drought tolerance. In fact, simply displaying characteristics that indicate increased water use efficiency does not necessarily indicate long-term drought tolerance (Zhang et al 1997).

2.5.3. Effects of moisture stress on gas exchange

Species with indeterminate growth, such as western larch, continue producing new foliage and subsequently maintain higher rates of gas exchange as long as conditions are favorable and moisture is readily available. This is why we found higher photosynthesis and transpiration for the control and low drought treatment seedlings, compared to the moderate and severe drought. However, when moisture is limiting it can encourage plants to not only change leaf structure, but to also make physiological changes in order to reduce cell damage and mitigate water loss (Lambers 2008). In addition, water availability influences stomatal conductance and thus carbon dioxide uptake and carbon assimilation (Reddy 2004). Therefore, decreased photosynthetic values were found in seedlings exclusively in the moderate and severe drought intensities. Photosynthesis and transpiration also differed among the treatments as the drought intensity progressed, possible response of increasing drought effects on seedling physiology.

Treatment alone was not significant for specific leaf area, but the treatment by week interaction showed a negative correlation in photosynthesis and specific leaf area meaning as drought intensity increased and for a longer period of time, both variables decreased. It has been found in research pertaining to moisture stress, that a lower specific leaf area is indicative of a conservative growth strategy common in drought tolerant plants, a greater photosynthetic capacity per unit leaf area, and lower drought induced mortality (Brink & Bruun 2011; Bowes et al 1972; Nautiyal et al 2002; Greenwood et al 2017). This adaptive strategy of reducing surface area allows seedlings to maximize photosynthetic output without having to produce more foliage. Reduction in specific leaf area has also been linked to species with slower relative growth rates and positively correlates to potential growth rates of seedlings, and annual aboveground biomass production (Reich et al 1997).

There appears to be some conflicting data from this study in regards to specific leaf area values across drought intensity. In week 6, specific leaf area decreased with increasing drought severity. This would coincide with the literature on lower leaf area values commonly found in drought stressed plants. However, in week 10 the values reversed and there was a positive correlation between drought severity and specific leaf area. Low specific leaf area was found in *Quercus* sp. seedlings placed under low-light and high drought conditions, and it was revealed that the leaf area had no impact on photosynthetic performance; in fact these seedlings were able to produce higher photosynthetic rates than those under full-sun (Quero et al 2006). Light availability appears to have a much more significant impact than moisture availability in regard to leaf structural traits, however, reduction in specific leaf area should still be considered a water-use efficiency tactic (Quero et al 2006; Brink & Bruun 2011). While smaller surface area allows plants to reduce loss from evapotranspiration, it also serves as a reduction in aboveground dry mass (Liu et al 2016).

2.5.4. Conclusion

The objectives of this study were to examine trends in first year western larch morphology and gas exchange in response to simulated drought and to examine if responses differed by half-sib families. A strong treatment × family response was observed with the greatest reductions in seedling height, diameter, and biomass production found in the severe drought treatment. Seedling development also followed a predictable trend by drought treatment with seedlings exposed to well-watered conditions produced the greatest biomass both aboveground and belowground, while seedlings in the severe drought treatment allocated a greater proportion of their total biomass belowground. Higher root mass fraction is a phenomenon observed in plants when stressed by moisture limitations and believed to be a strategy to scavenge for water at the expense of creating more respiring aboveground tissue. Even though the response of photosynthesis response to drought intensity did not differ by week, drought still had an effect, suggesting reduced gas exchange under water limited conditions led to reduced biomass allocation. Interestingly, height, RCD, and biomass production all differed by family and exhibited family-specific responses to drought intensity. Family 3, where the mother tree originated from a site with cool maximum temperature and low precipitation performed the best when water was a limiting while family 6 from the wettest hottest site performed worst among families in growth under high drought conditions. Although only a study of first year seedlings, it is interesting to observe the genotypic responses that suggest parent-tree climate may influence early seedling growth under a range of limited moisture regimes.

2.6. Figures

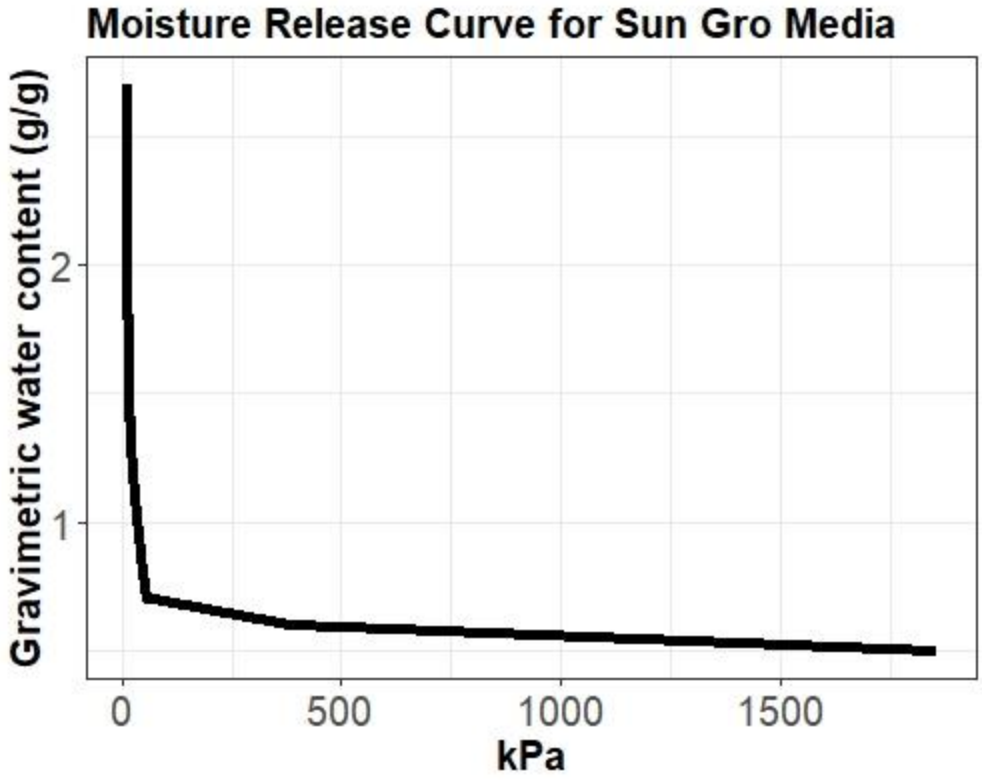

Figure 2. 1. Moisture release curve created specifically for SunGro professional horticulture growing mix comprised of 40-50% Canadian sphagnum peat, 30-40% vermiculite, and 10-20% bark, 4.5-5 pH, 70-160 EC.

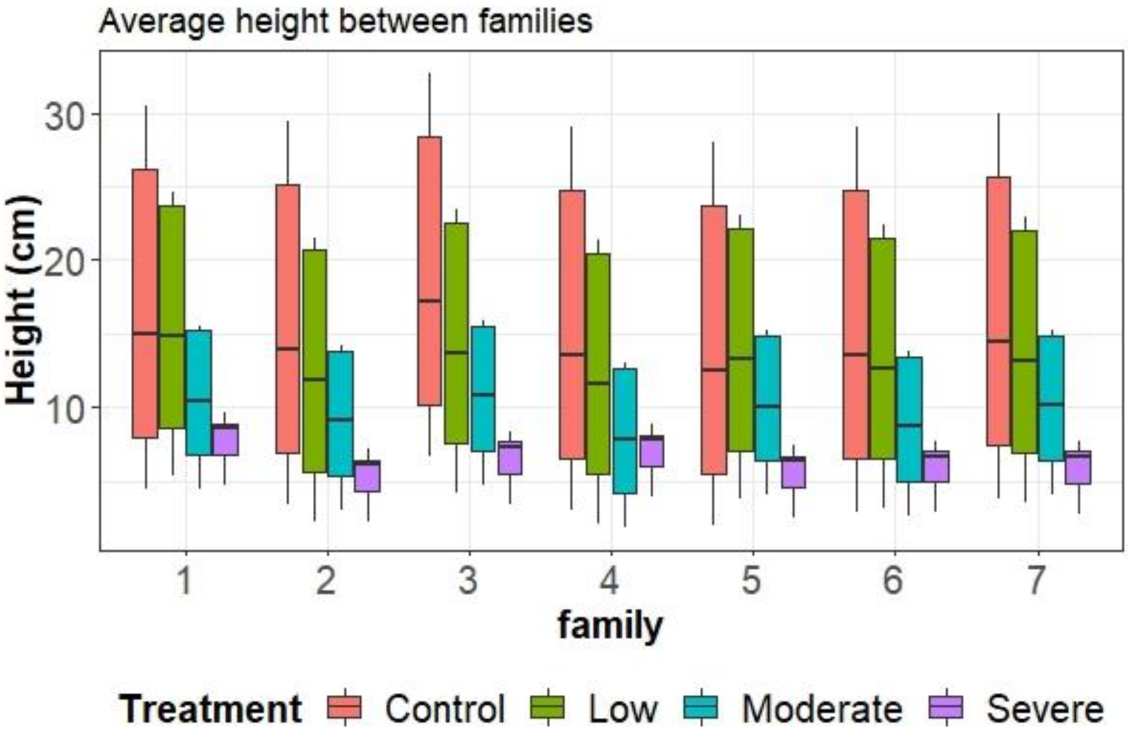

Figure 2. 2. Seedling final height averaged over all families compared between drought treatments using the linear mixed-effects model data. The line in the center of each box is the median, the bottom of the box is the 25th quartile, the top of the box is the 75th quartile, and the whiskers extending outside the box are the full range of heights.

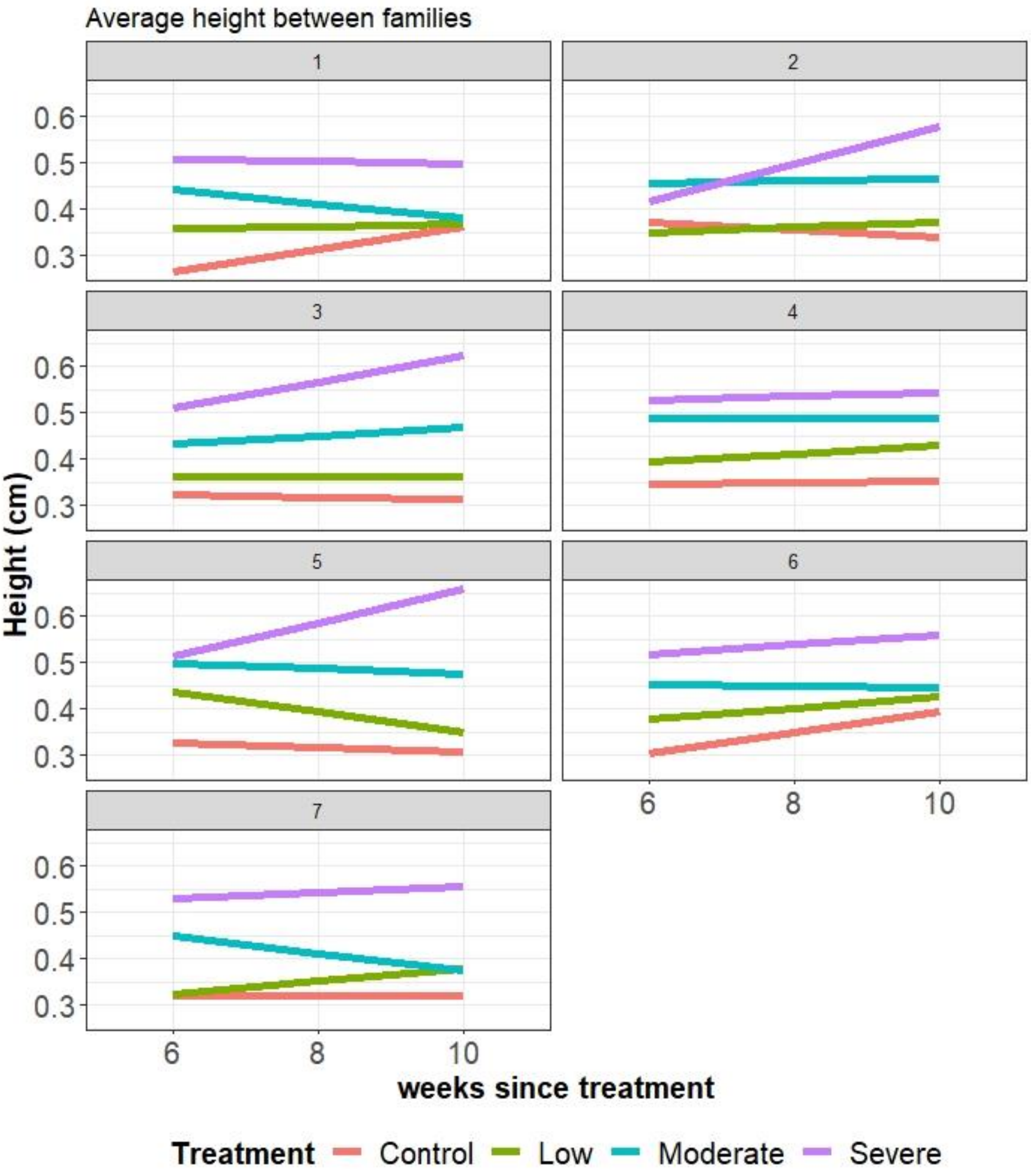

Figure 2. 3. Average seedling final heights by family and treatment effects using least-square means from the linear mixed-effects model.

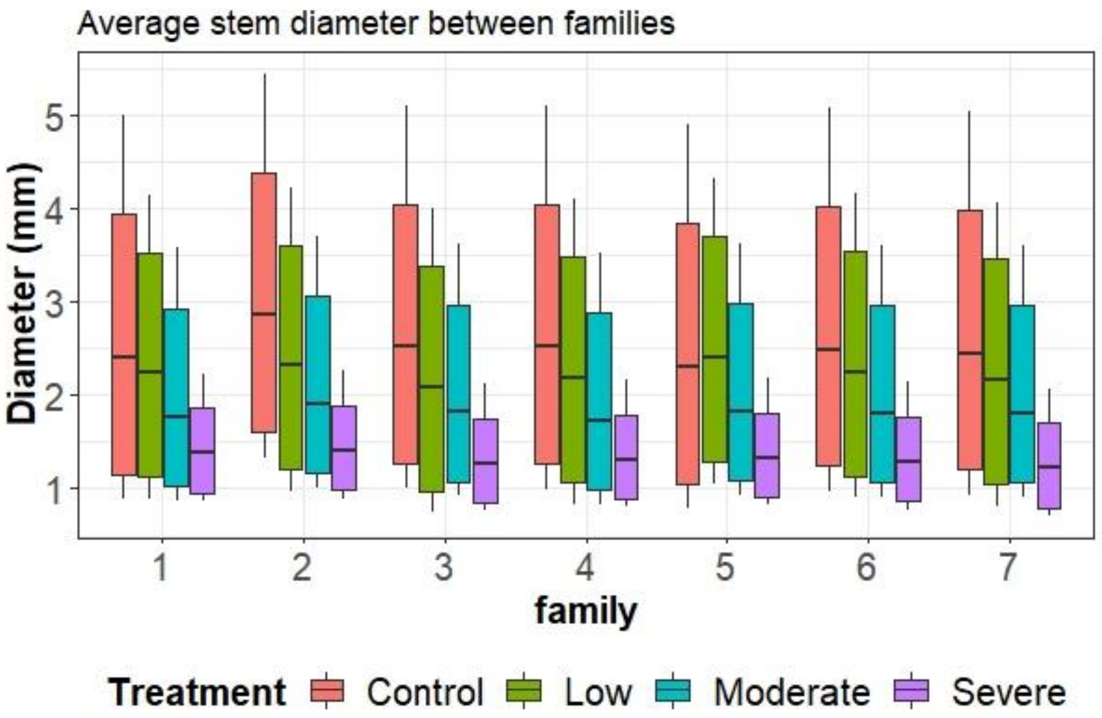

Figure 2. 4. Seedling final diameter averaged over all families compared between drought treatments using the linear mixed-effects model data. The line in the center of each box is the median, the bottom of the box is the 25th quartile, the top of the box is the 75th quartile, and the whiskers extending outside the box are the full range of diameters.

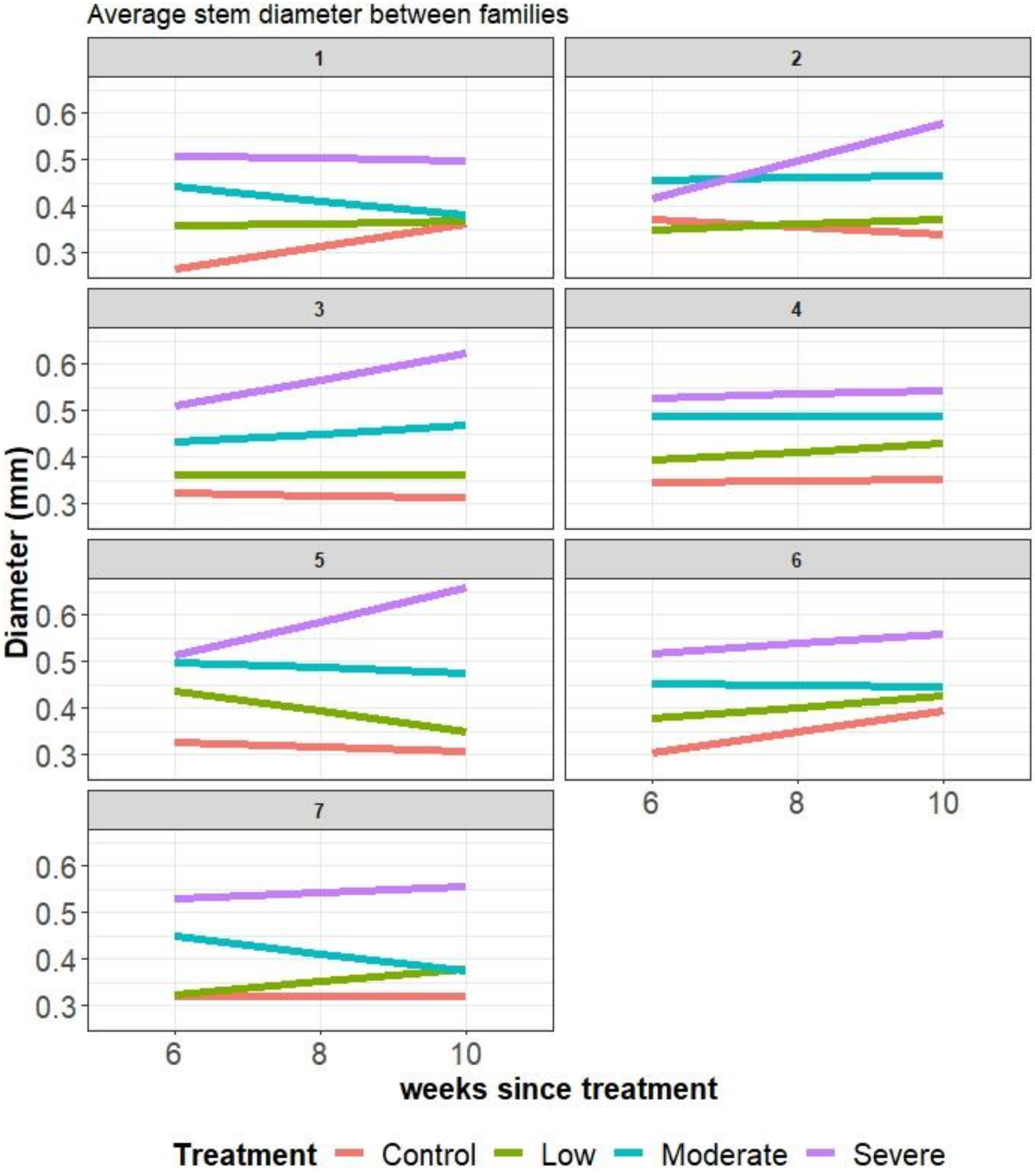

Figure 2. 5. Average final seedling root collar diameter by family and treatment effects using least-square means from the linear mixed-effects model.

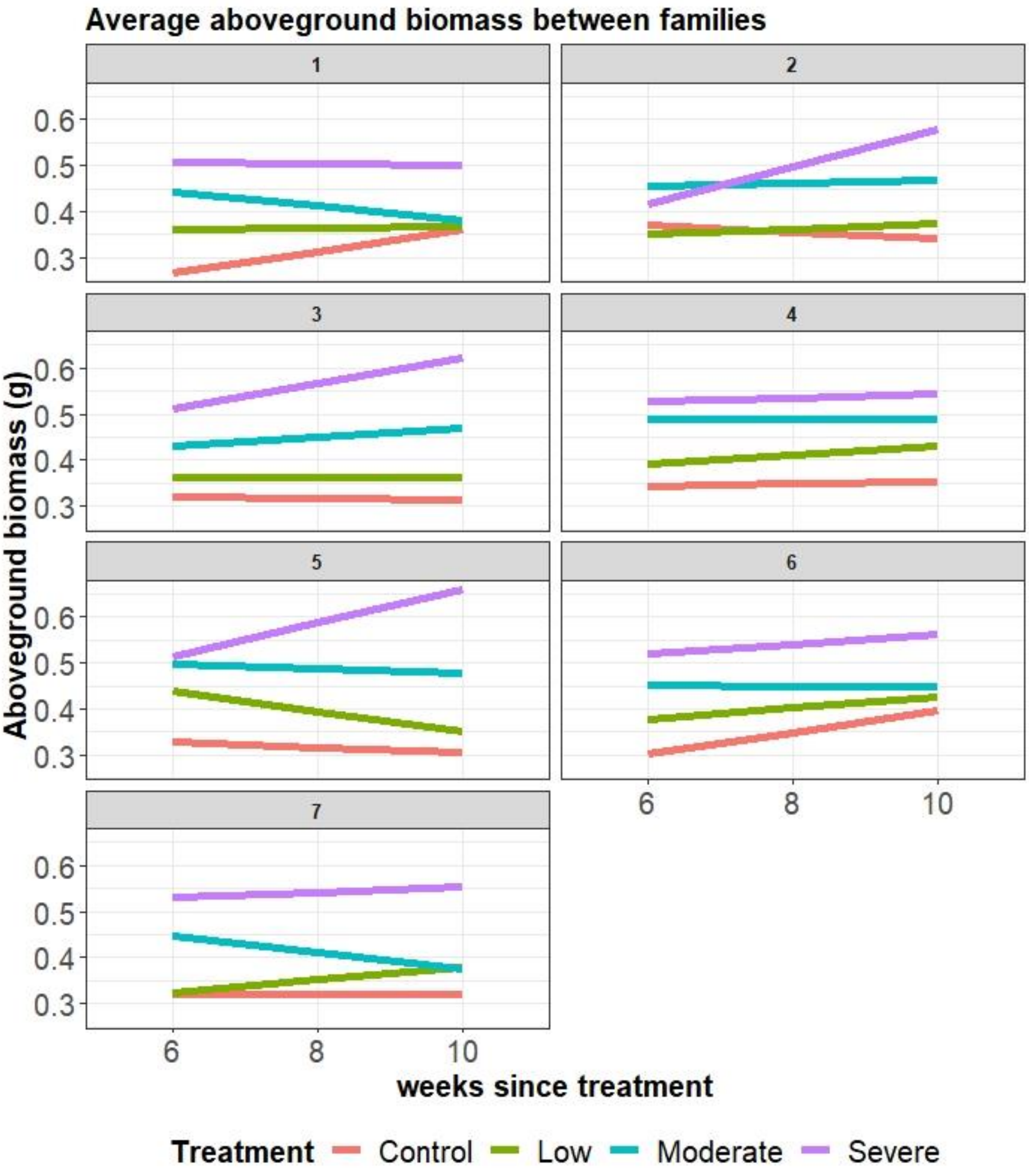

Figure 2. 6. Average aboveground biomass (foliage + stem) by family and treatment effects using least-square means from the linear mixed-effects model.

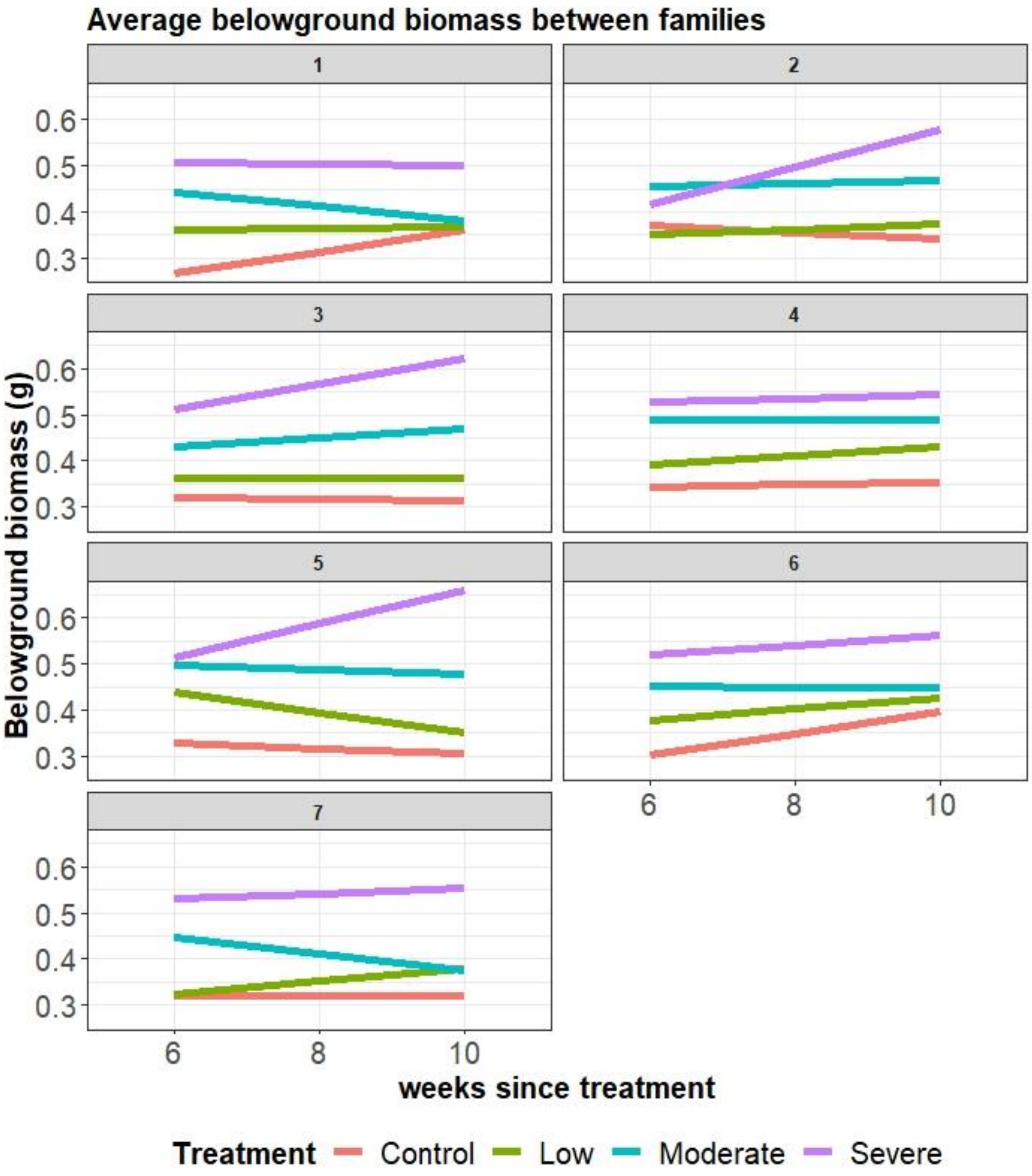

Figure 2. 7. Average belowground biomass by family and treatment effects using least-square means from the linear mixed-effects model.

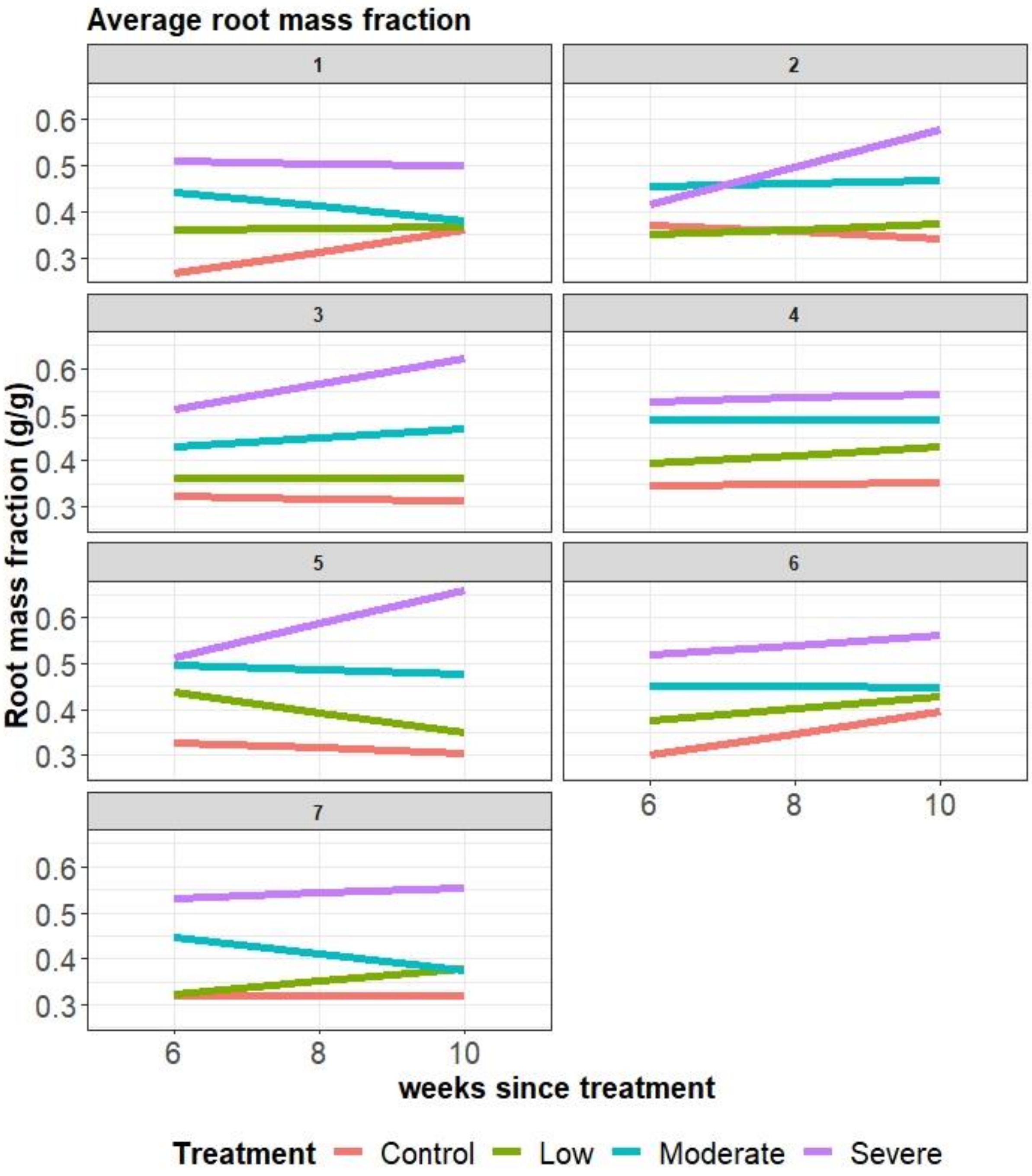

Figure 2. 8. Average seedling root mass fraction by family and treatment effects using least-square means from the linear mixed-effects model.

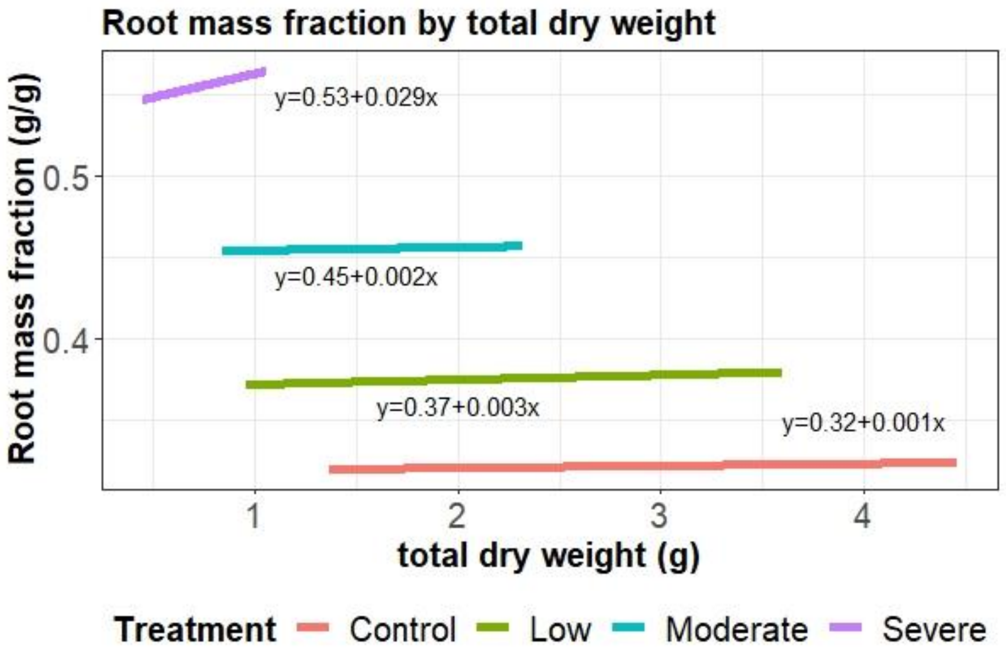

Figure 2. 9. Average seedling root mass fraction by total dry weight and treatment effects using least-square means from the linear mixed-effects model.

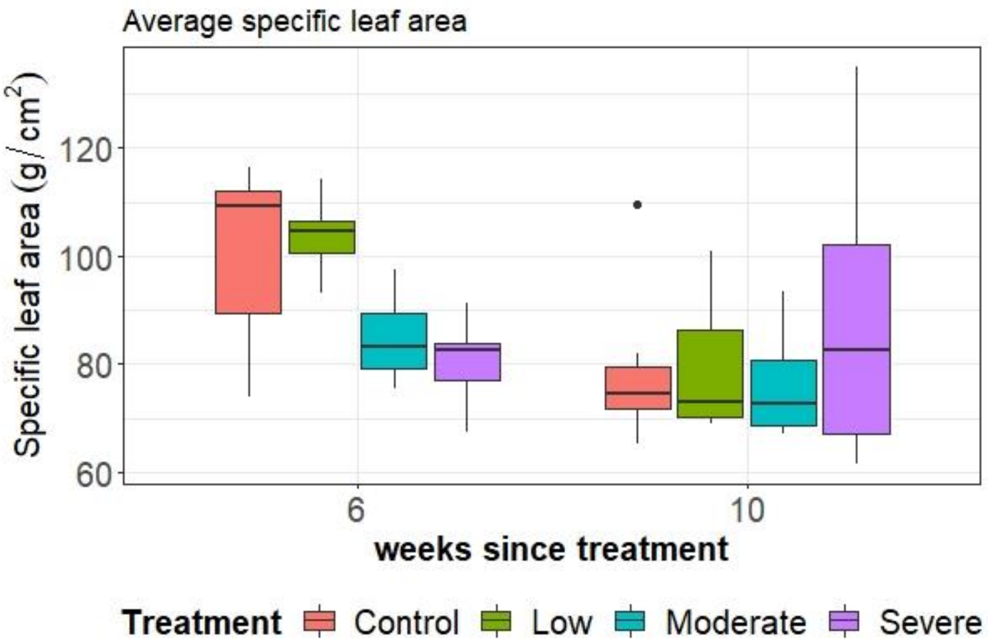

Figure 2. 10. Seedling photosynthesis averaged over sampling weeks compared between drought treatments using the linear mixed-effects model data.

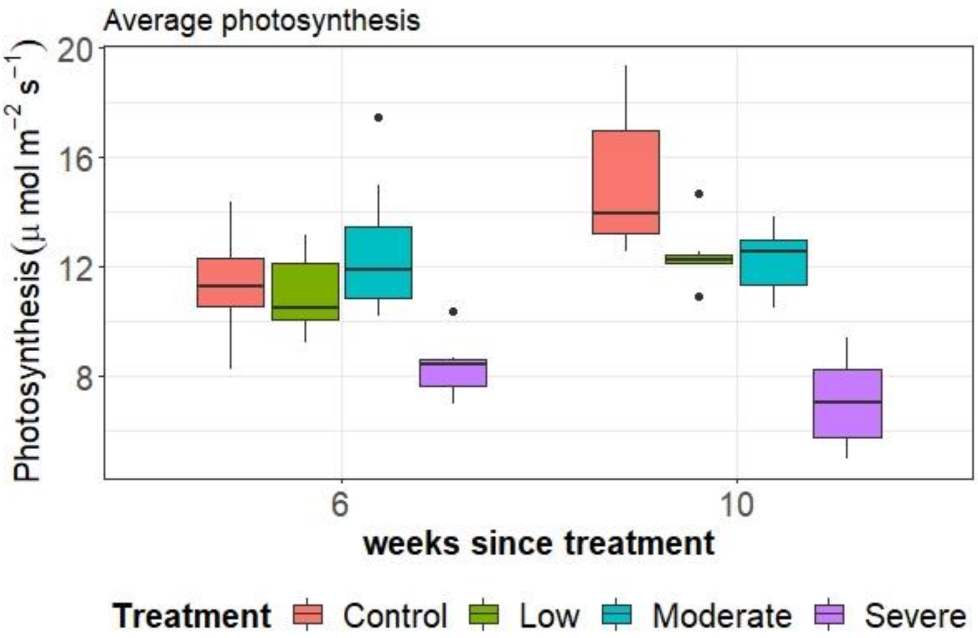

Figure 2. 11. Seedling photosynthesis averaged over sampling weeks compared between drought treatments using the linear mixed-effects model data.

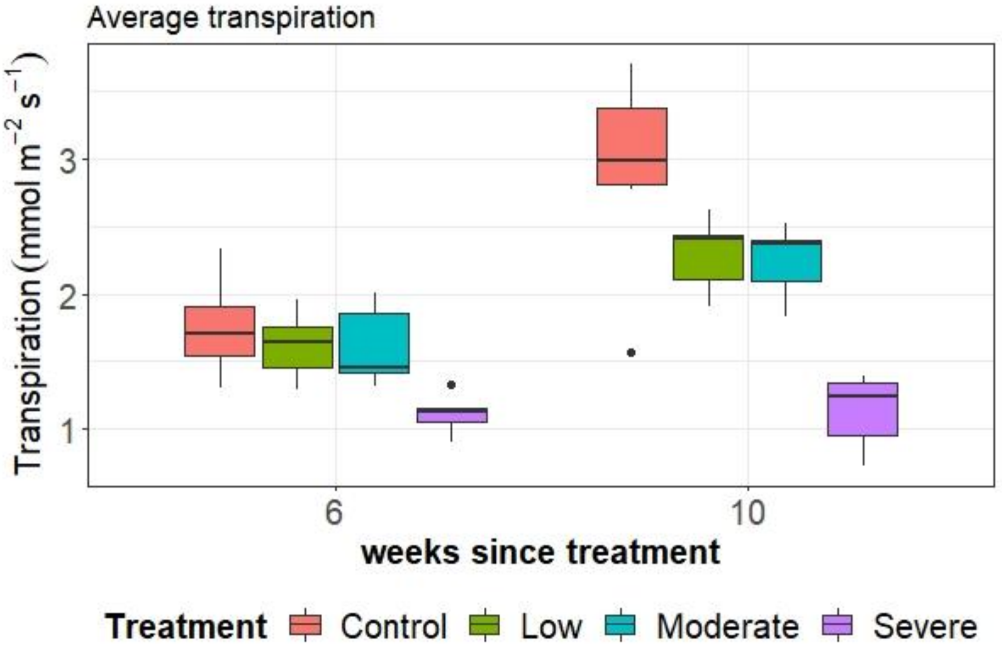

Figure 2. 12. Seedling transpiration averaged over sampling weeks compared between drought treatments using the linear mixed-effects model data.

2.7. Tables

Table 2. 1. Climate data for parent tree locations where Family refers to the experimental designated mother tree, and mmax refers to mean max temperature for the warmest month (ºC), and map refers to mean annual precipitation (cm).

Family	**elevation (m)**	**mmax** (ºC)	**map (cm)**
1	1280	24.2	60.6
2	1250	24.5	65.8
3	1265	23.2	70.7
4	945	25.6	87
5	1158	25	126.2
6	792	28.1	117.9
7	1036	25.4	78.9

Table 2. 2. ANOVA results for seedling height with F-value statistics and associated p-values at $\alpha = 0.05$.

Source	**DF**	**F-value**	**p-value**
(Intercept)	1	2219.5	<0.001
Drought intensity	3	675.8	<0.001
Family	6	26.4	<0.001
Week	9	585.1	<0.001
Drought × Family	18	5.6	<0.001
Drought × Week	27	62.2	<0.001
Family × Week	54	1.2	0.205
Drought × Family × Week	162	0.7	0.998

Table 2. 3. ANOVA results for seedling RCD with F-value statistics and associated p- values at $\alpha = 0.05$.

Source	**DF**	**F-value**	**p-values**
(Intercept)	1	6128.9	<0.001
Drought intensity	3	843.8	<0.001
Family	6	12.0	<0.001
Week	9	1148.4	<0.001
Drought × Family	18	4.4	<0.001
Drought × Week	27	58.8	<0.001
Family × Week	54	1.1	0.352
Drought × Family × Week	162	0.7	0.998

Table 2. 4. ANOVA results for seedling aboveground biomass with F-value statistics and associated p- values at α = 0.05.

Source	**DF**	**F-value**	**p-values**
(Intercept)	1	2834.6	<0.001
Drought intensity	3	189.8	<0.001
Family	6	4.1	0.001
Week	2	52.7	0.001
Drought × Family	18	21.7	<0.001
Drought × Week	6	23.1	<0.001
Family × Week	12	2.1	0.025
Drought × Family × Week	36	3.6	<0.001

Table 2. 5. ANOVA results for belowground biomass with F-value statistics and associated p- values at α = 0.05.

Source	**DF**	**F-value**	**p-values**
(Intercept)	1	1186.1	<0.001
Drought intensity	3	57.8	<0.001
Family	6	5.7	<0.001
Week	2	40.8	0.002
Drought × Family	18	6.4	<0.001
Drought × Week	6	11.1	<0.001
Family × Week	12	2.7	0.003
Drought × Family × Week	36	1.38	0.109

Table 2. 6. ANOVA results for seedling root mass fraction with F-value statistics and associated p- values at α = 0.05.

Source	**DF**	**F-value**	**p-values**
(Intercept)	1	15218.7	<0.001
Drought intensity	3	1165.3	<0.001
Family	6	4.7	0.003
Week	2	6.4	0.057
Drought × Family	18	1.4	0.152
Drought × Week	6	4.3	0.001
Family × Week	12	1.0	0.448
Drought × Family × Week	36	1.2	0.264

Table 2. 7. ANOVA results for specific leaf area with F-value statistics and associated p- values at α = 0.05.

Source	DF	F-value	p-values
(Intercept)	1	828.9	<0.001
Drought intensity	3	1.8	0.164
Family	6	2.1	0.070
Week	1	4.8	0.116
Drought × Family	18	1.3	0.215
Drought × Week	3	5.4	0.002
Family × Week	6	1.4	0.231
Drought × Family × Week	18	1.1	0.352

Table 2. 8. ANOVA for photosynthesis with F-value statistics and associated p- values at α = 0.05.

Source	DF	F-value	p-values
(Intercept)	1	464.7	<0.001
Drought intensity	3	11.1	<0.001
Family	6	1.4	0.240
Week	1	0.7	0.471
Drought × Family	18	0.5	0.957
Drought × Week	3	2.4	0.076
Family × Week	6	0.6	0.721
Drought × Family × Week	18	0.4	0.982

Table 2. 9. ANOVA results for transpiration with F-value statistics and associated p- values at α = 0.05.

Source	DF	F-value	p-values
(Intercept)	1	52.9	<0.001
Drought intensity	3	19.8	<0.001
Family	6	1.8	0.112
Week	1	1.3	0.342
Drought × Family	18	0.5	0.967
Drought × Week	3	3.4	0.022
Family × Week	6	0.4	0.895
Drought × Family × Week	18	0.4	0.987

2.8. References

Almeida, A., Loy, A., and Hofmann, H. 2017. qqplotr: Quantile-Quantile Plot Extensions for 'ggplot2', R package version 0.0.3 initially funded by Google Summer of Code 2017, https://github.com/aloy/qqplotr.

Aghai, M. M., Pinto, J. R., & Davis, A. S. 2014. Container volume and growing density influence western larch (*Larix occidentalis* Nutt.) seedling development during nursery culture and establishment. New Forests, 45(2): 199–213.

Beatty, S., & Stone, E. 1986. The variety of soil microsites created by tree falls. Canadian Journal of Forest Research, 16(3): 539-548.

Beckage, B., & Clark, J. S. 2003. Seedling Survival and Growth of Three Forest Tree Species : The Role of Spatial Heterogeneity. Ecology, 84(7): 1849–1861.

Boe, K. N. 1958. June 1958 silvics of western larch by Forest Service U. S. Department of Agriculture Reed W. Bailey, Director, (16).

Bowes, G., Orgen, W.L., Hageman, R.H. 1972. Light saturation, photosynthesis rate, RUDP carboxylase activity and specific leaf weight in soybeans grown under different light intensities. Crop Science. 12, 77-79.

Brenchley W.E. 1916. The effect of the concentration of the nutrient solution on the growth of barley and wheat in water cultures. Annals of Botany 30, 77–91 p.

Brink, D. J., & Bruun, H. H. 2011. Seedling stage strategies as a means of habitat specialization in herbaceous plants. PLoS ONE, 6(7): 1–10.

Duba, S. E., Goggans, J. F., & Patterson, R. M. 1972. Seed Source Testing of Alabama Loblolly Pine: Implications for Seed Movements and Tree Improvement Programs.

Cregg, B.M. 1994. Carbon allocation, gas exchange and needle morphology of *Pinus ponderosa* genotypes known to differ in growth and survival under imposed drought. Tree Physiology. 14, 883-898.

Cregg, B. M., & Zhang, J. W. 2001. Physiology and morphology of *Pinus sylvestris* seedlings from diverse sources under cyclic drought stress. Forest Ecology and Management, 154(1–2): 131–139.

Fisher, G. M. 1935. Comparative Germination of Tree Species on Various Kinds of Surface-Soil Material in the Western White Pine Type. Ecology, 16(4): 606.

Haig, I. T. 1936. Factors Controlling Initial Establishment of Western White Pine and Associated Species. Yale School of Forestry Bulletin 41. viii, 149.

Harper, J., Clatworthy, J., McNaughton, I., & Sagar, G. 1961. The evolution and ecology of closely related species living in the same area. Evolution, 15(2): 209-227.

Harrington, C. A., & St.Claire, B. S. 2017. The Douglas-fir Seed-Source Movement Trial Yields Early Results, (October), 22–24.

Højsgaard, S., and Halekoh, U. 2018. doBy: Groupwise Statistics, LSmeans, Linear Contrasts, Utilities. R package version 4. 6-2. https://CRAN.R-project.org/package=doBy.

Greenwood, S., Ruiz-Benito, P., Martínez-Vilalta, J., Lloret, F., Kitzberger, T., Allen, C. D., Jump, A. S. (2017). Tree mortality across biomes is promoted by drought intensity, lower wood density and higher specific leaf area. Ecology Letters, 20(4): 539–553.

Lambers, H., Chapin, F. S. I., & Pons, T. L. 2008. Plant Physiological Ecology. New York, NY.

Lenth, R. V. 2016. Least-Squares Means: The R Package lsmeans. Journal of Statistical Software, 69(1): 1-33. doi:10.18637/jss.v069.i01

Liu L.-B., Wu Y.-Y., Hu G., Zhang Z.-H., Cheng A.-Y., Wang S.-J., Ni J. (2016). Biomass of karst evergreen and deciduous broad-leaved mixed forest in central Guizhou province, southwestern China: a comprehensive inventory of a 2 ha plot. Silva Fennica, 50(3): 1492.

Long, E.M. 1980. Texas and Louisiana loblolly pine study confirms importance of local seed sources. Southern Journal of Applied Forestry. 4, 127-132.

Maximov N.A. 1929. The plant in relation to water. Allen and Unwin, London.

McIvor, I. R., Sloan, S., & Pigem, L. R. 2014. Genetic and environmental influences on root development in cuttings of selected *Salix* and *Populus* clones - a greenhouse experiment. Plant and Soil, 377(1–2): 25–42.

Missoorten, M., Turcsán, A., Mijnsbrugge, K. V., Mees, G., Steppe, K., Sárközi, E., & Erdélyi, É. 2016. Early summer drought stress during the first growing year stimulates extra shoot growth in oak seedlings (*Quercus petraea*). Frontiers in Plant Science, 7(February), 1–9.

Nautiyal, P. C., Rachaputi, N. R., & Joshi, Y. C. 2002. Moisture-deficit-induced changes in leaf-water content, leaf carbon exchange rate and biomass production in groundnut cultivars differing in specific leaf area. Field Crops Research, 74(1): 67–79.

Ohashi, Y., Nakayama, N., Saneoka, H., & Fujita, K. 2006. Effects of drought stress on photosynthetic gas exchange, chlorophyll fluorescence and stem diameter of soybean plants. Biologia Plantarum, 50(1): 138–141.

Okali, D., & Dodoo, G. 1973. Seedling growth and transpiration of two West African mahogany species in relation to water stress in the root medium. Journal of Ecology, 61(2): 421-438.

Oswald, B. P., & Neuenschwander, L. F. 1993. Microsite variability and safe site description for western larch germination. Bulletin of the Torrey Botanical Club, 120(2): 148–156.

Pinheiro J, Bates D, DebRoy S, Sarkar D, R Core Team. 2018. _nlme: Linear and Nonlinear Mixed Effects Models_. R package version 3.1-137, <URL: https://CRAN.R-project.org/package=nlme>.

Ponton, S., Dupouey, J. L., Bréda, N., & Dreyer, E. 2002. Comparison of water-use efficiency of seedlings from two sympatric oak species: Genotype x environment interactions. Tree Physiology, 22(6): 413–422.

Poorter, H., & Nagel, O. 2000. The role of biomass allocation in the growth response of plants to different levels of light, CO2, nutrients and water: a quantitative review. Australian Journal of Plant Physiology, 27(6): 12.

Quero, J. L., Villar, R., Marañón, T., & Zamora, R. 2006. Interactions of drought and shade effects on seedlings of four Quercus species: Physiological and structural leaf responses. New Phytologist, 170(4): 819–834.

Reddy, A. R., Chaitanya, K. V., & Vivekanandan, M. 2004. Drought-induced responses of photosynthesis and antioxidant metabolism in higher plants. Journal of Plant Physiology, 161(11): 1189–1202.

Reich, P. B., Walters, M. B., & Ellsworth, D. S. 1997. From tropics to tundra: Global convergence in plant functioning. Proceedings of the National Academy of Sciences of the United States of America, 94(December), 13730–13734.

Rötzer, T., Biber, P., Moser, A., Schäfer, C., & Pretzsch, H. 2017. Stem and root diameter growth of European beech and Norway spruce under extreme drought. Forest Ecology and Management, 406(December 2016), 184–195.

Teskey, R.O., Bongarten, B.C., Cregg, B.M., Dougherty, P.M., Hennessey, T.C. 1987. Physiology and genetics of tree growth in response to moisture and temperature stress: an examination of the characteristics of loblolly pine (*Pinus taeda L*.). Tree Physiology. 3, 41-61.

Vance, N., & Running, S. 1985. Light reduction and moisture stress: Effects on growth and water relations of western larch seedlings. Canadian Journal of Forest Research, 15(1): 72-77.

Vivekanandan, A.S., Gunesena, H.P.M. 1976. Lysimetric studies on the effect of soil moisture tension on the growth and yield of maize (Zea mays L.) and groundnut (*Arachis hypogaea* L.). Beitr. Trop. Landwirtsch Veterianacrmedizin 14, 369-378.

Wickham, H. 2016. ggplot2: Elegant Graphics for Data Analysis. Springer-Verlag New York.

Zhang, J.W., Feng, Z., Cregg, B.M., Schumann, C.M. 1997. Carbon isotopic composition, gas exchange, and growth of three populations of ponderosa pine differing in drought tolerance. Tree Physiology. 17, 461-466.

CHAPTER 3: GENOTYPIC RESPONSE OF PLANTED IMPROVED WESTERN LARCH SEEDLINGS TO SITE QUALITY

3.1. Abstract

Western larch (*Larix occidentalis*) is valued for timber production in the Inland Northwest, yet there is little research on efforts to improve this species to increase growth and value. This study planted improved western larch seedlings from seven half-sibling families and one bulk orchard lot across three sites located throughout the northern Idaho that differed in climate and soil parent material. This experiment used a Randomized Complete Block Design with three blocks at each site. Along with seedling height and diameter measurements, competing vegetation cover and height around seedling, as well as volumetric soil moisture, soil chemistry, and soil bulk density were collected during the first year of growth. Soil moisture, bulk density, and forb cover were all correlated with height growth, while stem diameter growth was not affected by any measured variables. Family showed no significant effect on growth. Lack of family significance may be a result of first year establishment where seedlings are allocating energy toward adapting to the environment. Ultimately, soil moisture, and the presence of forbs were the most influential to performance. Seedlings showed larger heights at the drier site, but overall higher averages of height growth at the wetter site because of consistent available moisture. Competition for resources with other vegetation, for water, nutrients, and light, had negative and positive effects on growth.

3.2. Introduction

Since its inception, manipulation of forest genetics has been heavily researched for a variety of reasons ranging from commercial, climatic, and purely scientific. From a commercial standpoint, the purpose of an improvement program is to increase wood quality and productivity in commercial timber species through the use of applied research and provenance trials (Wheeler et al 2015). Tree improvement gained popularity in the early 1900s following the rediscovery of Gregor Mendel's plant breeding trials, and was heavily supported in the face of impending timber shortages across the U.S. (Prudham 2003). Early tree improvement work by U.S. Forest Service Scientist Thornton Munger focused mainly on Douglas-fir (*Pseudotsuga menziesii*) and it was not until the mid-1970s that trials were performed using western larch (*Larix occidentalis*) (Prudham 2003; Jaquish & El-Kassaby 1998). Models of western larch tree breeding trials have shown a potential for considerable gains in growth, yet publications on actual field performance on improved western larch are still uncommon (Moore 1984). Thus there currently remain questions pertaining to cost-effectiveness for tree improvement programs and the level of control required to obtain economic gains, whether through open-pollinated orchard seed lots or controlled breeding.

Western larch is endemic to the inland northwestern US and is highly valued for timber production (Schmidt & Shearer 1991). Landowners traditionally see high rates of success when planting on favorable site conditions particularly on clear-cuts where slash is reduced and vegetation is controlled (Boe 1958). Indeterminate growth gives western larch an advantage over other tree species because they can continue to grow if conditions are favorable, whereas other species like Douglas-fir and western white pine (*Pinus monticola*) set bud earlier and are limited to predetermined growth restrictions. Western larch seedlings can still experience high mortality within the first few years post-planting if vegetation control is poor, planted on drought-prone sites, damage from animal browsing or trampling. These issues are important to consider, especially matching genotypes to site conditions, to ensure investment in advanced genetics is not lost.

Current research on western larch improvement focuses on the logistics of tree breeding programs and associated costs with maintenance, orchard size, and increasing seed production. Some of those concerns result from the potential imbalance of genetic variation among open-pollinated orchard clones, the inconsistent quality and quantity of cone-crops with western larch, and most importantly the costs associated with running a large orchard while maintaining high genetic diversity (Ledig & Porter 1982; Funda et al.2009). Preliminary data from out-planting trials shows evidence that western larch responds positively to one generation of selective breeding for height growth and that the amount of genetic gain displayed through progenies is mostly determined by the quality of the selected parent trees (Rehfeldt 1991; Funda et al. 2009). Although tree improvement is advantageous for western larch and it remains important to select high quality parent trees for genetic material, it is still uncertain how parent tree climate effects western larch genotypes when planted in diverse site conditions.

One way to assess genetic variation within a genotype is through provenance testing. Provenance zones indicate the range in which seedlings can move without deleterious effects on growth and survival (Kreyling et al. 2012). Provenance testing can be used to assess to what degree progenies can be moved from their original seed source. Harrington and St. Claire (2017) found significant differences in mean height of Douglas-fir depending on provenance climate by age 8, where coastal varieties outperformed inland low elevation and high elevation seed sources in all testing locations from California, southern Oregon, central Oregon, and Washington. Provenance trials in southern Alabama found loblolly pine seedlings originating from a southern seed source significantly outperformed native sources in mean height growth by age 1 up to 14 when planted across a southern, central, and northern climate gradient. (Duba et al 1984). Rehfeldt (1991) found western larch seed lots originating from higher elevations showed lower average growth and earlier dormancy when planted at lower elevations, while seed lots from those same lower elevations grew faster for longer and were larger overall by age 8 at 730 m elevation. Assessing the response of genotypes to provenance zones can take years, as establishment conditions at the planting sites can overwhelmingly affect seedlings prior to genetic expression of performance (Isik et al. 2005).

A collection of seven half-sib improved western larch seed lots selected across a climate gradient based on mean annual temperature and mean annual precipitation was tested for this study along with an open-pollinated orchard bulk lot. The objectives of the study were to (1) examine first year growth and survival after planting effects of family by site quality and (2) examine the effects of soil moisture and competing vegetation on seedling morphology.

3.3. Materials and methods

In 2007 the Inland Empire Tree Improvement Cooperative (IETIC) established a western larch seed orchard containing families from superior-rated trees which originated from Eastern Washington, Western Montana, and Northern Idaho. The orchard at Kalamalka, British Columbia serves to produce open-pollinated seed for reforestation of cooperative member lands and to test superiority of clones for future breeding programs. Parent trees for this project were specifically selected across a climate gradient, with varying mean annual precipitations and elevations, in order to capture a broad range of different genotypes (Chapter 2). Cone producing limbs were collected from the parent trees and grafted onto trees that were planted within an orchard at Kalamalka, British Columbia.

Seed was isolated from seven individual half-sib families and one open pollinated bulk lot in late summer 2016. Seed was sent to the University of Idaho Franklin H. Pitkin Forest

Nursery where it was put in cold storage and then stratified for 28 days. Seed was rinsed for 48 hours following stratification and then sown into 130 ml cell Styroblocks® (Beaver Plastics). Seedlings were grown from April to October of 2017 using the standard western larch cultural practices used at the Pitkin Nursery (Dumroese and Wenny 1995). Seedlings were grown in SunGro professional horticulture growing media was used, comprising of 40-50% Canadian sphagnum peat, 30-40% vermiculite, and 10-20% bark, 4.5-5 pH, and 70-160 EC. Height and stem diameter were measured biweekly to track growth by family (Table 3.1). Seedlings were packed in November 2017 and placed in cold storage until May 2018. Landowners for each site provided professional planting crews for planting in May and June 2018. Trees within the measured plot were individually tagged with aluminum tags in the summer of 2018.

3.3.1. Site description

The three sites chosen for this study were provided by members of the University of Idaho Intermountain Forestry Cooperative (Fig. 3.1). Sites were selected across a range of different soil types and climates in northern Idaho (Table 3.2). The most southern site called Threebear is owned by PotlatchDeltic Corporation and is located near Elk River, Idaho. The elevation of Potlatch is about 914 m and receives on average 87 cm of precipitation annually (US Climate Data 2019). The annual high temperature is 14 ^{o}C and the annual low temperature is 0.4 ^{o}C (Fig 3.2). Soils at Potlatch are 92% Norwidge-Threebear complex and 8% Riswold-Grangemont complex (USDA 2006) with a parent material of volcanic ash over basalt. Potlatch has a 5-30% slope and is characterized by a western red cedar (*Thuja plicata*) and queencup beadlily (*Clintonia uniflora*) habitat type.

The second site, Low Crutch, is owned by Stimson Lumber Company and is located south of Plummer, Idaho. This site is located at 1,025 m elevation and receives 86 cm of precipitation per year (US Climate Data 2019). The annual high temperature is 13 ^{o}C and the annual low is 2 ^{o}C (Fig. 3.3). The soil map for Stimson is the most diverse of all the sites and is comprised of 12% Ardenvoir-Huckle complex, 3% Huckle ashy silt loam, 32% Tigley moist-Humus complex, and 53% Ardenvoir-Huckle-Saint Maries complex (USDA 2006). Stimson has a 35- 65% slope with the plots located on the back-slope position. The soils are well drained and the parent material is volcanic ash over metasedimentary rock. The most predominant habitat types are grand fir (*Abies grandis*), western red cedar, and western hemlock (*Tsuga heterophylla*), with queen-cup bead lily and ninebark (*Physocarpus malvaceus*).

The most northern site Scary Berry is located north of Sandpoint, Idaho and is managed by Molpus Timberlands Management, Llc. The elevation is 1,100 m and is on a 35-65% linear back slope. Average annual precipitation is 107 cm, and average annual high temperature is 10 ^{o}C and the average low is -0.4 ^{o}C (Fig. 3.4) (US Climate Data 2019). The site is classified as Priestlake gravelly sandy loam. The parent material is volcanic ash over glacial till. Molpus is unique from the other sites because it is influenced by glacial events dominated by the retreating of the Cordilleran ice sheet 7,000 years ago. The predominant habitat type at this site is western hemlock and queen-cup bead lily (USDA 2006).

Soil nutrient analysis was provided by AgSource Harris Laboratories, Nebraska and was utilized to determine soil chemistry and soil texture. Stimson and Molpus were similar in nutrient makeup. Stimson had only slightly higher amounts of potassium (K), magnesium (Mg), nitrogen (N), and calcium (Ca). Molpus was determined to be mainly sandy loam, and Stimson and Potlatch were textured as silt loam (Table 3.3).

3.3.2. Experimental Design

At each site, a randomized complete block design, with three blocks each with eight treatment plots randomly located within each block were installed (Appendix). Each block contained two rows of four blocks with the long-end of the block spanning across the slope contour. Each plot was 27.4 × 27.4 m and contained a single family or the orchard bulk lot with 81 trees (9 x 9 rows) and a 49-tree measurement plot nested in the center (Figure 3.5). Trees were planted approximately 3.1 m apart to align with operational planting standards. A 6.1 m buffer was maintained between plots. Blocks were all placed on either a north or east aspect and within elevation ranges where western larch would naturally grow. Some plots had to be repositioned to work around slash piles and skid trails (Appendix A).

3.3.2. Field data collection

Initial height and root collar diameter (RCD) were measured in June 2017 for all seedlings within the measurement plots. Height, diameter, and survival were then measured at the end of the growing season in late September 2017. Percent volumetric soil water content was measured three times during the growing season within the top 15 cm of mineral soil using a HydroSense II soil moisture probe (Campbell Scientific, Logan, UT). The first sample period was early June, the next in mid-July, and the last in mid-October to examine the decline and rebound in soil moisture characteristic of the seasonal drought in North Idaho. Moisture measurements were collected from 10 randomly selected spots within each plot and were averaged to obtain plot level estimates. Random sample locations were determined by the sampler tossing a marked stick over the shoulder without looking. This was repeated for each measurement.

Competing vegetation was measured in mid-June immediately after the first measurement of soil moisture. A 1-m^2 PVC pipe frame was placed at five systematic points within each plot, resulting in a 1.13% sample within a plot. A 50 m tape was used to measure 34.4 m across the diagonal of the measurement plot from tree 1 to tree 49. The first vegetation plot was 8.6 m along the diagonal, followed by 17.2 m (center of the plot), and the third at 25.8 m (Fig. 3.6). The remaining two plots were placed at 8.6 m towards the plot center from the corners along the perpendicular diagonal. Percent vegetation to the nearest 5% level accuracy was measured within each frame by specific shrub and forb species known to be common and highly competitive and also broader vegetation groups. Vegetation groups included slash, bare ground, detritus, perennial grass, annual grass, snowberry (*Symphoricarpos albus*), ninebark (*Physocarpus opulifolius*), ocean spray (*Holodiscus discolor*), Oregon grape (*Mahonia aquifolium*), ground shrubs, fern, golden pea (*Thermopsis montana*), bull thistle (*Cirsium vulare*), Canada thistle (*Cirsium arvense*), yarrow (*Achillea millefolium*), mullein (*Verbascum thapsus*), other forbs, other shrubs, trees, and bryophytes.

Mineral soil bulk density was measured in July 2018 to assess soil compaction. Three samples of bulk density cores were collected per plot using a bulk density soil sampler (AMS, American Falls, ID). Sample one was collected at the plot center, and then two more samples were collected 8.6 m from the center in opposite directions along the gradient of the plot. The sampler canister contained two cylinders each 90.59 cm^3 in volume. After removing the organic layer around the sampling location, the auger head was driven into the ground with a slide hammer attachment. The cylinders were carefully removed from the auger head, placed in a holding platform, and then separated using a sharp knife. The knife was also used to shave off excess soil from the open ends of the cylinder. Care was taken to maintain the soil integrity within the cylinder and not lose or compact the sample. One cylinder full of soil was emptied into a pre-weighed autoclave bag which was then brought back to the lab and placed in an oven for 48 hours. The dried soil was weighed after drying and the pre-weighed bag weight was subtracted from the total weight in order to determine soil weight in grams. Bulk density was then calculated as the ratio of weight to the known volume in the canister. Bulk density samples were averaged to obtain plot level estimates.

Soil chemistry samples were collected at the same time as the bulk density samples. Five samples were collected per plot using a regular soil auger (AMS, American Falls, ID). After removing the organic layer around the sampling location, the auger head was twisted into the ground using the top bar handles. One sample was collected at plot center, followed by two samples 8.6 m distance from the center point moving along the gradient. Two more samples were then collected perpendicular to the gradient line 8.6 m from the center, similar to the vegetation sampling. Soil cores were removed from the auger, thoroughly combined in a bucket, and then placed into a gallon plastic bag until full. Samples were brought back to the lab and finely sieved to remove as much debris as possible. One cup of soil was then placed back into a labelled plastic bag and left open to air dry before sending off to AgSource Harris Laboratories, Nebraska for analysis. Samples were not used in the data analysis but they were used to describe plot level soil chemistry.

3.3.3. Statistical analysis

Separate linear mixed-effect analysis of covariance (ANCOVA) models were used to analyze the effects of soil variables and competing vegetation and their interactions with site and family as fixed effects on the change in height and diameter growth. Block was a random effect in the models. R software was used to analyze the data, including the 'nlme' package which was used to calculate linear mixed-effects models (R Core Team 2013; Pinheiro et al 2018). Least-squared means were calculated for all morphological parameters against environmental and biological factors using the 'lsmeans' package (Lenth 2016). The package used for the graphing of the least-squared means was 'ggplot2' (Wickham 2016). The soil parameter models tested the effects of soil bulk density and VWC in July. The vegetation competition models tested all vegetation groups.

Non-significant variables and their interactions were removed from the full models to find parsimonious models with only significant variables. Seedlings counted as dead, missing, or with negative height growth or RCD growth were removed from the analysis. Those with negative height growth were most likely browsed, damaged by frost, or lost their terminal shoot in some other way. Data collected from the Potlatch site was omitted from moisture, soil BD, and vegetation analysis due to June frost damage that stunted seedling growth significantly and would have skewed the data. Potlatch height growth, RCD growth, and survival were analyzed by site and by clone. Survival was calculated as a percentage of remaining live trees at the end of the growing season out of 49 initially planted. The vegetation competition models dropped Molpus because the site contained very low amounts of competing vegetation, thus skewing the results. There were no issues of heteroscedasticity in the residuals plots for any of the data analyzed for this experiment. Package 'qqplotr' (Almeida et al 2017) was used to check normality to determine best fit models. Data appeared normally distributed for all the models according to the standardized residuals in the QQplot.

3.4. Results

3.4.1. Height and stem diameter growth

Average seedling height growth significantly differed by site (p=0.001), July soil VWC (p=0.030) (Fig. 3.7), and soil bulk density (BD) (p=0.005), but not by family (p=0.814) (Table 3.4). Average height growth at Stimson was 30.2 ± 2.6 cm and 17.1 ± 2.6 cm at Molpus ($p \leq 0.001$). Although not analyzed against environmental factors, average height growth at Potlatch was 15.1 ± 2.6 cm. July VWC had a positive correlation with height growth (Fig. 3.8). No environmental factors were significant for average RCD growth (Table 3.5). Average RCD growth was 4.25 ± 0.3 mm at Stimson, and 4.25 ± 0.3 mm at Molpus (p=0.192). Molpus experienced a more consistent soil moisture level throughout the summer starting at 27% VWC in June, 22% VWC in July, and 22% VWC in October, whereas there was a noticeable dry down at Stimson with 31% VWC in May, 18% VWC in July, and 13% VWC in October.

3.4.3. Survival

Survival significantly differed by site (p=0.018) (Table 3.6). Overall survival was highest at Molpus (99.7 ± 0.005%), second highest at Stimson (98.9 ± 0.005%), and lowest at Potlatch (96.5 ± 0.005%) (Table 3.7). When tested as an interaction for clone by site, there are no significant differences. However, Family 2 had a significant difference in survival between Molpus (100%) and Potlatch (95.2%) (p=0.033), and Family 6 had a significant difference between Potlatch (95.2%) and Stimson (99.3%) (p=0.033).

3.4.4. Competing Vegetation

Average RCD growth slightly differed by total forb percent (p=0.060), but was not significant to any other factors (Table 3.8). The same trend was found in average height growth, with only total forb slightly significant (p=0.073) (Table 3.9). Height growth was negatively correlated with total forb (Fig. 3.9). Average vegetation group estimates for Stimson were 35% slash, 41% detritus, 9% bare ground, 2% tree regeneration (mainly aspen), 1% thistle, and 1% perennial grasses.

3.5. Discussion

3.5.1. Family and site effect

While it has been supported in many field trials that improved families significantly outperform unimproved clones over time, in some cases 30% volume gains in loblolly pine (Isik et al. 2005), the results from this experiment on improved western larch are strictly limited to seedling in their first growing season after planting and no family effects were detected. Thus it is not wise to speculate beyond the current data and assume that family will have a significant effect on morphological growth in the future. In some cases genotypic variation may not present itself at all as it has been reported in species such as loblolly pine that genetic variation among half-sib and full-sib families showed no significance for morphologic growth traits (Emhart et al. 2006). The most probable explanation for why family was not significant is that the seedlings are still adjusting to the new environmental conditions. C effects, which are non-genetic effects common (C) to a group of individuals, could explain why there were no noticeable family differences in the field trial (Lerner 1958). Homogenous nursery conditions, a qualifying C effect, can both bias post-planting seedling morphology (Baltunis et al 2007). However, C effects have been found to lessen over time revealing phenotypic family differences in measurable growth traits (Libby and Jund 1962). Noticeably higher amounts of foliage and rapid height and diameter growth displayed in certain families during nursery propagation may have been C effects. An alternative explanation for lack of family significance could be that the clones are all of matching quality, including the bulk orchard lot, which implies that there is no benefit to controlling to the half-sib level over open-pollinated seedlots.

Technically, the seed lots for this study are not exact genetic clones of the original parent trees, so there will inherently be variation in phenotypic response based on the unique genetic makeup of the seed orchard. In some cases it is speculated that an 80/20 rule is in effect, in which 80% of the seed-crop is produced by only 20% of the parent trees within the seed orchard (Funda et al. 2009). For this experiment, the mother tree is known for all seed lots except the bulk orchard lot, but the father tree is unknown. Influence of the parent trees may have some effect on phenotype later on, as shown in a 5 year study of loblolly pine in Georgia (Paul et al. 1997). Paul et al. (1997) reported that significance in height growth from a male x female parent interaction gradually increased throughout the measured period up to age 5. They speculated that delayed significance was due to interspecific competition that typically exists during post-planting establishment. However, the parent tree interaction only accounted for about 2% of total variation in height, while environmental effects accounted for nearly 81%. This supports the results found in our experiment, in which environmental factors and site had a significant effect on seedling performance while family did not.

3.5.2. Soil moisture and soil bulk density

The positive correlation between height growth and VWC was not surprising as it is common for seedlings to display increased height growth in well-watered conditions (Chapter 2, Fortin et al. 2018; Batkhuu et al. 2006). The results for the analysis of soil moisture and height growth were interesting because seedlings grown at the drier site Stimson had significantly higher average height growth compared to the wetter site Molpus. This could indicate water was not a limiting factor, but instead could have been temperature driven. Although soil temperature was not measured in this study, cooler temperatures can result in less height growth (Lafleur et al 2015).

Results suggested that the late frost event at Potlatch had a significant impact on total average height growth compared to the other sites (Table 3.10). This was also reflected in the survival, which was lowest in Potlatch and highest at Molpus. Family 6 and 2 seedlings were significantly impacted in terms of survival, both having lowest survival at Potlatch. Of all the parent trees, family 6 originated from the lowest elevation, southernmost latitude while family 2 originated from a high elevation, at the northernmost latitude. Provenance latitude has been found to significantly impact springtime frost tolerance in Siberian larch (*L. sibrica*), where results showed northern provenances had longer frost tolerance than southern ones (Eysteinsson et al 2009). However, this contradicts results found in other larch studies conducted in Canada, where Carswell and Morgenstern (1995) found seedlings from northern and higher elevation provenances were more susceptible to early frost than southern ones. It is certain that the frost event did significant damage to terminal shoots, thus reducing height growth. However, without having data on initial bud break and foliage mass, it is too difficult to claim that certain families were specifically impacted by the frost event and that their survival was not a factor of vegetation competition or moisture.

Stem diameter growth is always a secondary growth to stem elongation. Seedlings allocate resources to height growth upon bud break, and if they have a surplus of energy and conditions are favorable they will accumulate significant RCD growth. Otherwise, diameter growth is simply a function of creating new xylem tissue for water transport and for structural support for a taller stem. Site became a significant factor when Potlatch was included in the analysis. The frost event most likely affected diameter growth because seedlings had to use most of their energy to recover from terminal shoot damage. It is common for RCD to remain unaffected by moisture following out-planting. A study conducted on early seedling growth in Olga Bay larch (*Larix olgensis* Henry) showed no significant differences in RCD growth between stock type sizes from year one to year three following out-planting (Li et al. 2011). A two-year study on the growth of red pine seedlings (*Pinus resinosa* Ait.) also found no significant effect of soil moisture on RCD until year two (Strothmann 1967). Significant effects of environmental factors will most likely present themselves in future measurements, however, the seedlings from this experiment were still relying on holdover nutrients from the previous year's nursery regime.

3.5.2. Vegetation competition effect

Results revealed a negative correlation between height growth and total forb cover, indicating that height growth decreased as total forb increased. In most cases, herbaceous competition will have a negative effect on height growth during early seedling establishment because there is direct competition for water and nutrients in the soil (Balandier et al 2006).

Competition control studies on coastal Douglas-fir showed a negative correlation between total vegetation cover and overall volume growth at age 5 (Maguire et al. 2009). However, once the trees reached a substantial volume they were able to outcompete the vegetation and competition was no longer significant factor. Another study on maple and birch species found, within two years, that the relationship between competing vegetation and seedling height was highly negative in early successional communities (Berkowitz et al. 2014). Jensen and Löf (2017) found young oaks were significantly affected by interspecific woody competition by the second year of growth, whereupon competitive growth slowed significantly. Western larch does have somewhat of a competitive advantage in the fact that it has indeterminate growth. Where most forbs reach peak growth for the season in July and die back in August, western larch can continue to grow into mid to late September depending on favorable conditions. However, overall growth is still significantly affected by the presence of vegetation competition, which is why site preparation with herbicide is crucial to initial seedling performance.

3.5.3. Conclusion

The objectives of this study were to examine first year growth and survival after planting effects of family by site quality, and examine the effects of soil moisture and competing vegetation on seedling morphology. As improved seed, all families plus the bulk orchard lot displayed superior overall growth. Survival was high across all sites despite a late frost in June which did have a significant negative impact on seedling morphology. The most significant factor to first-year seedling growth was soil moisture. Higher amounts of moisture did not result in larger trees which could indicate that the seed lots are able to cope with drier site conditions. However, since temperature was not recorded, it could be possible that the cooler temperatures at Molpus negatively affected seedling height growth. Competing vegetation regeneration barely had a significant effect on morphology, not surprising as western larch’s rapid and indeterminate growth allows it to outcompete other vegetation. Even though no family differences were detected this could change in the next 5 years of growth potentially revealing the gains from improvement of this high valued conifer species. Parent tree provenance effects could still present themselves in the morphology. This portion of the study should be viewed as a long-term experiment, and current results should not be considered final indicators of genotypic performance.

3.6. Figures

Figure 3. 1. Map of site locations across the Idaho panhandle.

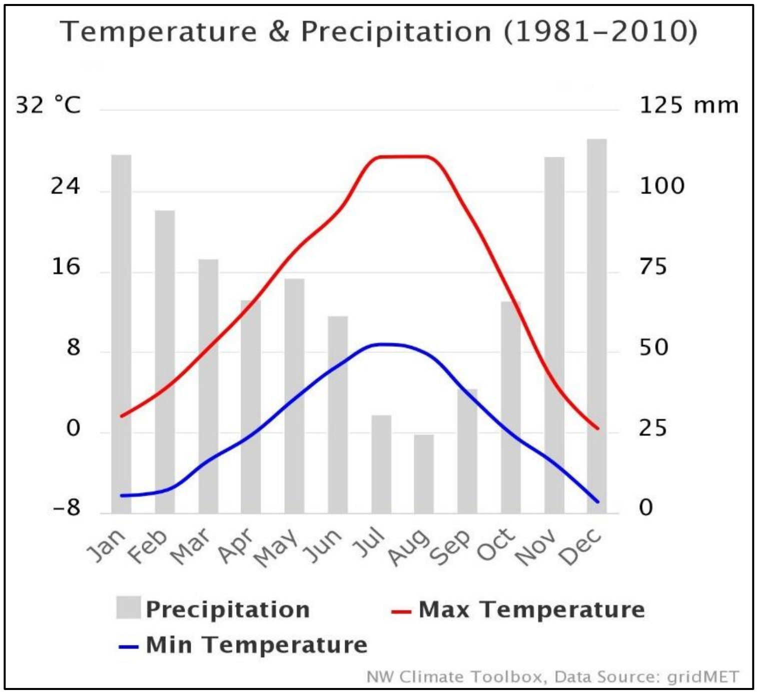

Figure 3. 2. Potlatch annual climate data including max temperature, minimum temperature, and precipitation.

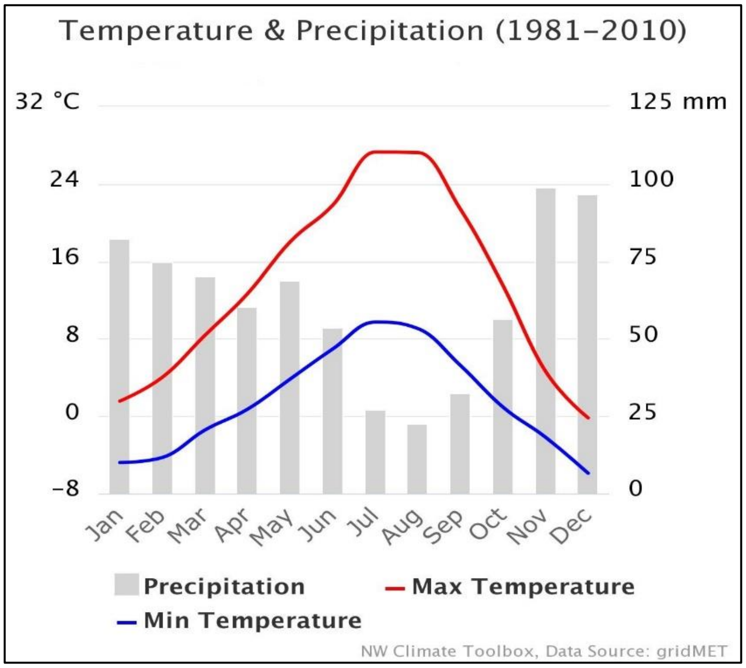

Figure 3. 3. Stimson annual climate data including max temperature, minimum temperature, and precipitation.

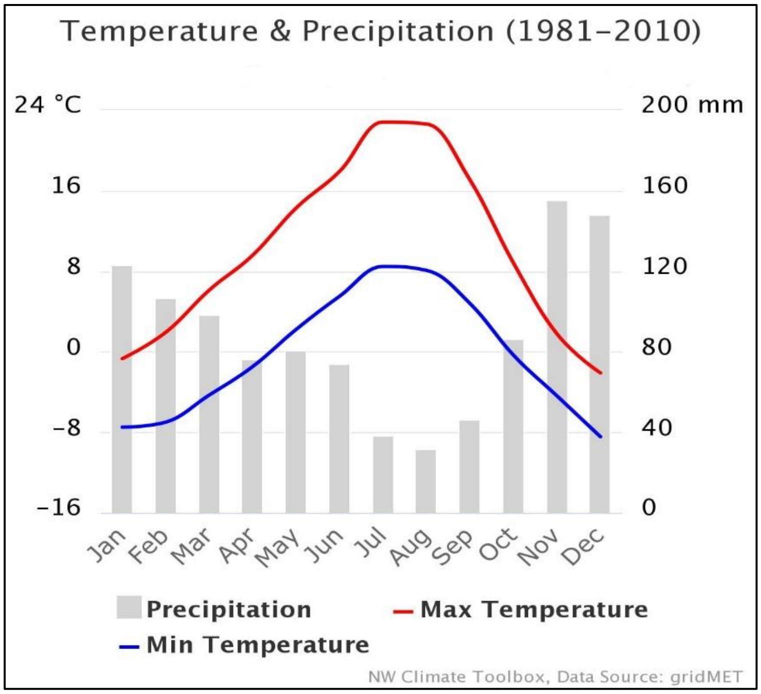

Figure 3. 4. Molpus annual climate data including max temperature, minimum temperature, and precipitation.

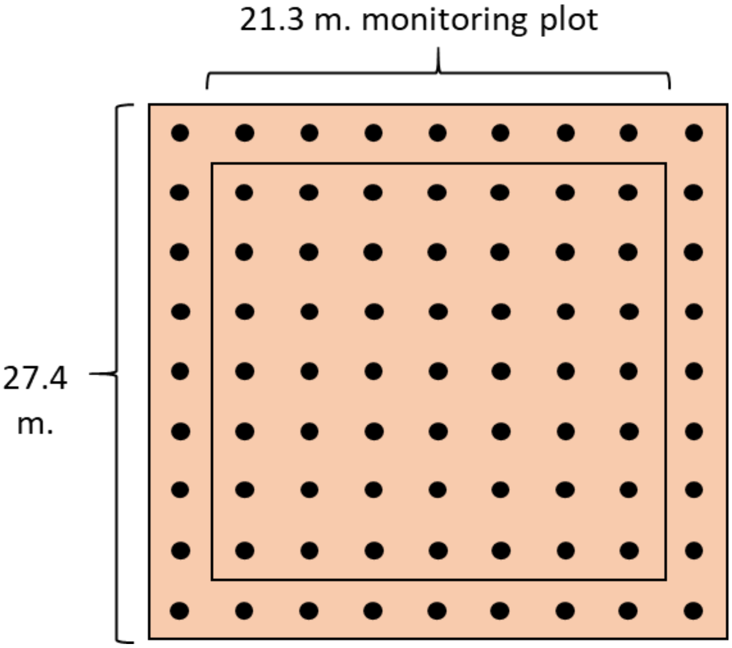

Figure 3. 5. Individual 27.4 × 27.4 m plot design containing a single family or the orchard bulk lot with 81 trees (9 x 9 rows) with a 49-tree measurement plot nested in the center.

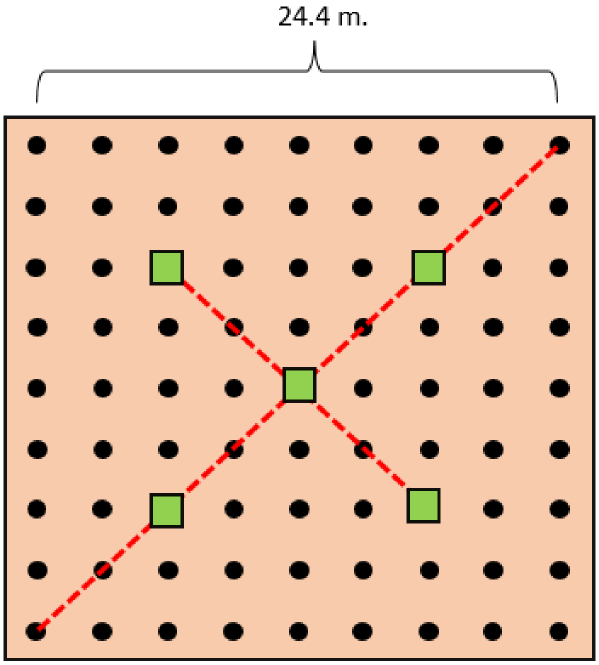

Figure 3. 6. Diagram of the vegetation group sample plots. Each green square represents a m^2 sample plot, each distanced 8.6 m apart.

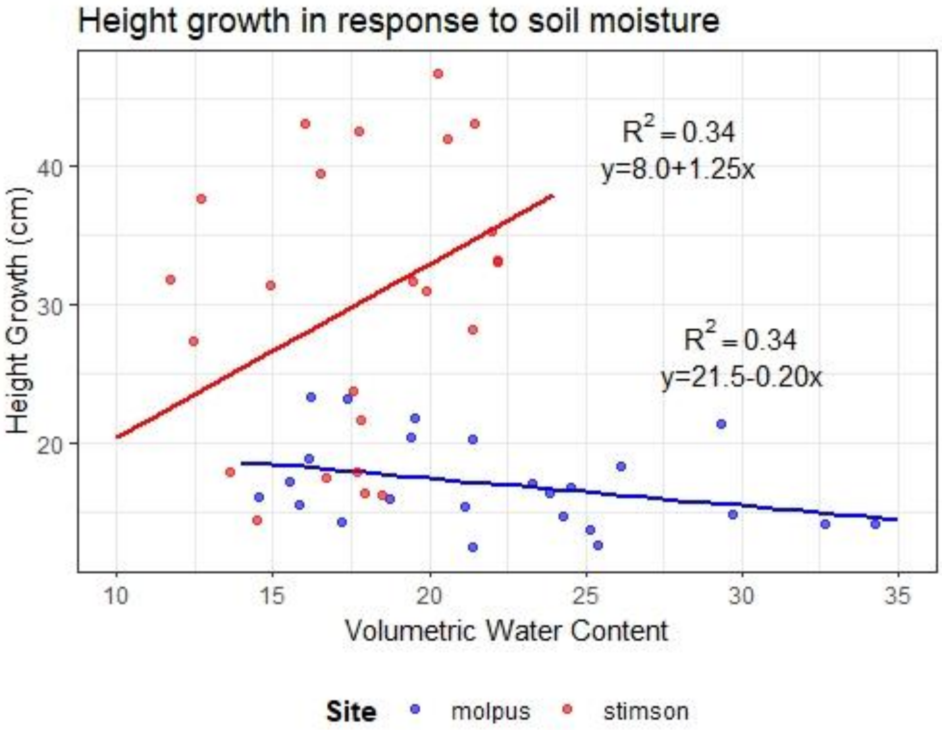

Figure 3. 7. Change in average height growth in response to soil volumetric water content (%) measured in July.

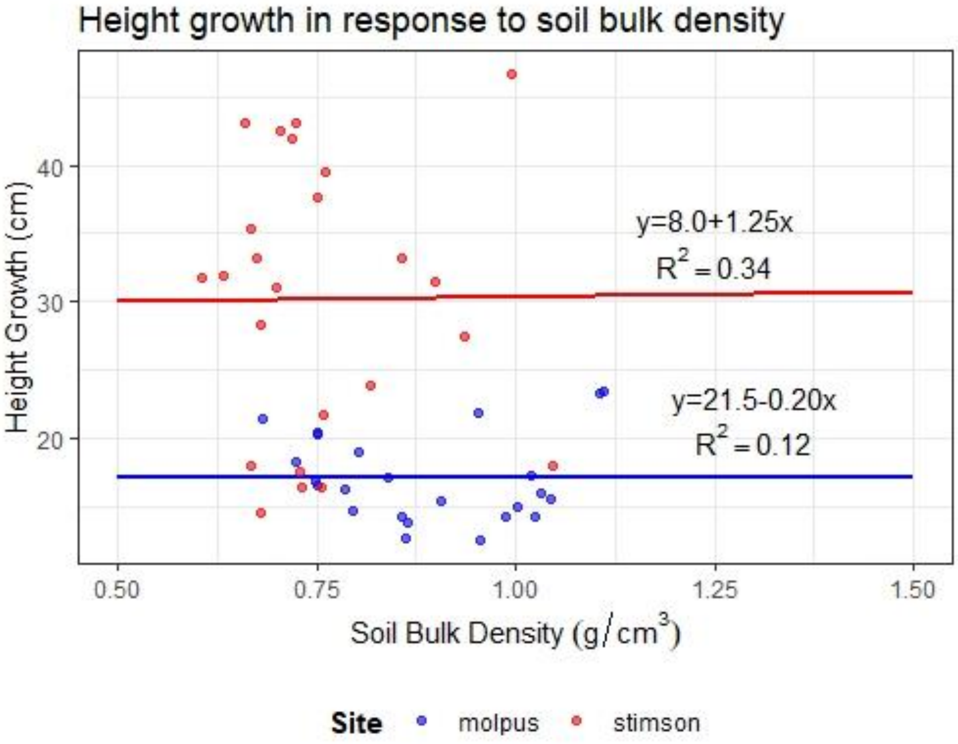

Figure 3. 8. Change in average height growth in response to soil bulk density.

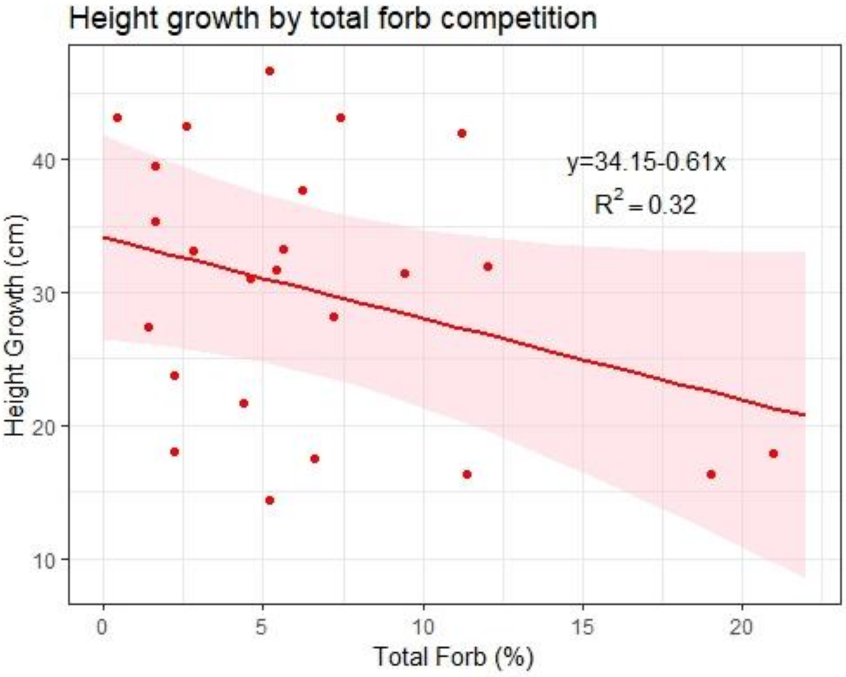

Figure 3. 9. Change in average height growth in response to total forb at the plot level only at the Stimson site.

3.7. Tables

Table 3. 1. Seedling height and stem diameter measured at weeks 1, 9, and 25 for the nursery crop.

	Average Height (cm)			**Average RCD (mm)**		
	Week			**Week**		
Family	**1**	**9**	**25**	**1**	**9**	**25**
1	8.4	26.9	27.1	1.62	3.18	4.46
2	8.9	26.3	26.6	1.70	3.31	4.53
3	8.7	28.4	26.3	1.56	3.06	4.30
4	8.7	27.3	27.4	1.65	3.16	4.33
5	7.2	27.8	28.0	1.61	3.17	4.31
6	8.3	26.9	27.4	1.70	3.25	4.49
7	6.9	25.1	24.9	1.51	3.00	4.26
Bulk	8.0	26.3	28.6	1.64	3.17	4.21

Table 3. 2. Site elevation, climate, and soil type. Elevation is in (m), MAP is mean annual precipitation (cm), and temperatures are annual high and annual low (^{o}C).

Site	**Elevation**	**MAP**	**Temp$_{high}$**	**Temp$_{low}$**	**Soil Texture**	**Parent Material**
Potlatch	914	87.0	13	0.4	silt loam	volcanic ash over basalt
Stimson	1,025	86.0	13	2	silt loam	volcanic ash over metasedimentary rock
Molpus	1,100	107.0	10	-0.4	sandy loam	volcanic ash over glacial till

Table 3. 3. Soil chemistry analysis for the three sites, results provided by AgSource Harris Laboratories, NE**.**

	% OM	K ppm	Mg ppm	N ppm	Ca ppm	pH	CEC	% Sand	% Silt	% Clay
Molpus										
Block 1	7	103	63	15	624	6	12	57	38	5
Block 2	8	94	47	13	452	6	12	57	38	5
Block 3	5	114	68	8	637	6	12	64	30	6
Stimson										
Block 1	6	195	104	12	1363	6	11	46	49	6
Block 2	5	269	103	12	1417	7	11	42	53	5
Block 3	6	218	104	20	1299	6	10	40	56	4
Potlatch										
Block 1	7	194	65	13	637	6	13	37	58	5
Block 2	6	217	60	7	756	6	12	37	58	5
Block 3	6	218	102	5	941	6	13	34	60	7

Table 3. 4. ANOVA results for height growth environmental conditions with F-value statistics and associated p- values at α = 0.05.

Source	**DF**	**F-value**	**p-values**
(Intercept)	1	138.3	<0.001
VWC (July)	1	3.9	0.058
Bulk Density	1	6.6	0.016
Site	1	20.7	0.001
Family	7	0.5	0.814
Site × Family	7	0.1	0.994

Table 3. 5. ANOVA results for RCD growth environmental conditions with F-value statistics and associated p- values at α = 0.05.

Source	**DF**	**F-value**	**p-values**
(Intercept)	1	347.7	<0.001
VWC (July)	1	0.0	0.964
Bulk Density	1	1.6	0.217
Site	1	1.7	0.205
Family	7	0.9	0.514
Site × Family	7	0.2	0.977

Table 3. 6. ANOVA results for survival with F-value statistics and associated p-values at α = 0.05.

Source	DF	F-value	p-values
(Intercept)	1	132,317.5	<0.001
Site	2	12.9	<0.001
Family	7	0.2	0.993
Site × Family	14	0.5	0.901

Table 3. 7. Survival percentage of seedlings at the plot level within site.

	Family								
Site	1	2	3	4	5	6	7	Bulk	Total
Molpus	99.3 a	100 a	99.3 a	99.3 a	100 a	99.3 a	100 a	100 a	99.7 b
Stimson	99.3 a	99.3 a	98.6 a	99.3 a	97.3 a	100 a	98.0 a	99.3 a	98.9 b
Potlatch	96.6 a	95.2 a	98.0 a	98.0 a	97.3 a	95.2 a	96.6 a	95.9 a	96.5 a

Table 3. 8. ANOVA results for RCD growth biological conditions with F-value statistics and associated p- values at α = 0.05.

Source	DF	F-value	p-values
(Intercept)	1	67.9	0.001
Family	7	0.7	0.685
Total forb	1	4.1	0.082
Total thistle	1	1.5	0.261
Total shrub	1	0.3	0.617
Totforb × Totthistle	1	0.2	0.644
Totforb × Totshrub	1	0.3	0.622
Totthistle × Totshrub	1	0.5	0.502
Totforb × Totthistle × Totshrub	1	1.2	0.304

Table 3. 9. ANOVA results for height growth biological conditions with F-value statistics and associated p- values at α = 0.05.

Source	DF	F-value	p-values
(Intercept)	1	64.1	0.001
Family	7	0.4	0.851
Total forb	1	4.4	0.073
Total thistle	1	0.3	0.623
Total shrub	1	2.0	0.196
Totforb × Totthistle	1	0.6	0.456
Totforb × Totshrub	1	0.2	0.651
Totthistle × Totshrub	1	0.8	0.397
Totforb × Totthistle × Totshrub	1	1.8	0.218

Table 3. 10. Average initial and final seedling height (cm) for families within site.

	Molpus		**Stimson**		**Potlatch**		**Totals**	
Family	*Initial*	*Growth*	*Initial*	*Growth*	*Initial*	*Growth*	*Initial*	*Growth*
1	28.4	17.8	26.8	33.4	27.1	14.2	27.4	21.8
2	27.3	16.2	27.8	29.6	28.4	11.2	27.8	19.0
3	30.3	16.9	29.1	35.4	29.7	11.5	29.7	21.3
4	29.2	15.1	28.8	27.5	28.4	11.5	28.8	18.0
5	29.2	17.5	28.9	27.7	27.9	16.2	28.7	20.5
6	29.3	14.3	27.9	25.6	27.9	15.7	28.4	18.5
7	26.2	19.1	27.3	32.8	25.6	12.2	26.4	21.4
Bulk	28.5	15.1	27.0	28.4	27.0	16.8	27.5	20.1
Total	**28.6**	**16.5**	**28.0**	**30.1**	**27.8**	**13.7**	**28.1**	**20.1**

Table 3. 11. Average initial and final seedling stem diameter (mm) for families within site.

	Molpus		**Stimson**		**Potlatch**		**Totals**	
Family	*Initial*	*Growth*	*Initial*	*Growth*	*Initial*	*Growth*	*Initial*	*Growth*
1	4.02	3.85	4.17	4.21	4.29	1.64	4.16	3.23
2	4.01	3.70	4.28	4.68	4.10	1.87	4.13	3.42
3	3.75	4.14	4.12	5.29	4.17	1.96	4.01	3.80
4	3.87	3.27	4.17	3.64	4.08	1.49	4.04	2.80
5	3.64	3.78	3.98	3.73	4.26	1.80	3.96	3.10
6	4.03	3.24	4.09	3.55	4.29	2.13	4.14	2.97
7	3.85	3.90	4.09	4.49	3.97	1.92	3.97	3.44
Bulk	3.83	3.87	4.16	3.99	4.24	1.88	4.08	3.25
Total	**3.88**	**3.72**	**4.13**	**4.20**	**4.18**	**1.84**	**4.06**	**3.25**

3.8. References

Almeida, A., Loy, A., and Hofmann, H. 2017. qqplotr: Quantile-Quantile Plot Extensions for 'ggplot2', R package version 0.0.3 initially funded by Google Summer of Code 2017, https://github.com/aloy/qqplotr.

Balandier, P., Collet, C., Miller, J.H., Reynolds, P.E., and Zedaker, S.M. 2006. Designing forest vegetation management strategies based on the mechanisms and dynamics of crop tree competition by neighbouring vegetation. Forestry 79(1): 3-27.

Baltunis, B. S., Huber, D. A., White, T. L., Goldfarb, B., & Stelzer, H. E. 2007. Genetic analysis of early field growth of loblolly pine clones and seedlings from the same full-sib families. Canadian Journal of Forest Research, 37(1): 195–205.

Batkhuu, N.-O., Lee, D. K., Tsogtbaatar, J., Dae, P. Y., Bat-Amgalan, R., & Eun, P. G. 2006. Variation in Seed Quality, Seedling Growth and Biomass Allocation of One-year-old Siberian Larch (*Larix sibirica* Ledeb.) Seedlings Grown in Different Conditions from Diverse Seed Sources of Mongolia. Mongolian Journal of Biological Sciences, 4(2): 15–23.

Berkowitz, A. R., Canham, C. D., & Kelly, V. R. 2014. Competition vs. facilitation of tree seedling growth and survival in early successional communities, 76(4): 1156–1168.

Carswell, C. L. & Morgenstern, E. K. 1995. Phenology and growth of nine larch species and hybrids tested in New Brunswick, Canada. In W. C. Schmidt & K. J. McDonald (Eds.), Ecology and management of Larix forests: A look ahead. Ogden, UT: Intermountain Research Station. General Technical Report, GTR-INT-319, 318-322.

Dumroese, R.K., and Wenny, D.L. 1995. Growing western larch in a container nursery. *In* Ecology and management of *Larix* forests: A look ahead. Edited by W.C. Schmidt and K.J. McDonald, Gen. Tech. Rep. GTR-INT-319. Ogden, UT: U.S. Department of Agriculture Forest Service, Intermountain Research Station. 213-219.

Burdett, A. N. 1983. Quality Control in the Production of Forest Planting Stock by. Forestry, (June), 132–138.

Emhart, V. I., Martin, T. A., White, T. L., & Huber, D. A. 2006. Genetic variation in basal area increment phenology and its correlation with growth rate in loblolly and slash pine families and clones. Canadian Journal of Forest Research, 36(4): 961–971.

Eysteinsson, T., Karlman, L., Fries, A., Martinsson, O., & Skulason, B. 2009. Variation in spring and autumn frost tolerance among provenances of Russian larches (*Larix* Mill.). Scandinavian Journal of Forest Research, 24(2): 100–110.

Fortin, M., Couwenberghe, R. Van, Perez, V., & Piedallu, C. 2019. Evidence of climate effects on the height-diameter relationships of tree species, 1–20.

Funda, T., Lstibůrek, M., Lachout, P., Klápště, J., & El-Kassaby, Y. A. 2009. Optimization of combined genetic gain and diversity for collection and deployment of seed orchard crops. Tree Genetics and Genomes, 5(4): 583–593.

Grossnickle, S. C., & MacDonald, J. E. 2018. Why seedlings grow: influence of plant attributes. New Forests, 49(1): 1–34.

Højsgaard, S., and Halekoh, U. 2018. doBy: Groupwise Statistics, LSmeans, Linear Contrasts, Utilities. R package version 4.6-2. https://CRAN.R-project.org/package=doBy

Isik, F., Goldfarb, B., Lebude, A., Li, B., & Mckeand, S. 2005. Predicted genetic gains and testing efficiency from two loblolly pine clonal trials, 1766(1990): 1754–1766.

Jaquish, B., & El-Kassaby, Y. A. 1998. Genetic variation of western larch in British Columbia and its conservation. Journal of Heredity, 89(3): 248–253.

Jensen, A. M., & Löf, M. 2017. Effects of interspecific competition from surrounding vegetation on mortality, growth and stem development in young oaks (*Quercus robur*). Forest Ecology and Management, 392, 176–183.

Kreyling, J., Wiesenberg, G. L. B., Thiel, D., Wohlfart, C., Huber, G., Walter, J., Anke, J.,Konnert, M., Beierkuhnlein, C. 2012. Cold hardiness of *Pinus nigra* Arnold as influenced by geographic origin, warming, and extreme summer drought. Environmental and Experimental Botany, 78, 99–108.

Lafleur, B., Cazal, A., Leduc, A., & Bergeron, Y. 2015. Soil organic layer thickness influences the establishment and growth of trembling aspen (*Populus tremuloides*) in boreal forests. Forest Ecology and Management, 347, 209–216.

Ledig, F. T., & Porter, R. L. 1982. Tree Improvement in Western Conifers : Economic Aspects.

Lenth, R. V. 2016. Least-Squares Means: The R Package lsmeans. Journal of Statistical Software, 69(1): 1-33.

Lerner, I. M. 1958. The genetic basis of selection. John Wiley & Sons, Inc., New York.

Li, G. L., Liu, Y., Zhu, Y., Yang, J., Sun, H. Y., Jia, Z. K., & Ma, L. Y. 2011. Influence of initial age and size on the field performance of Larix olgensis seedlings. New Forests, 42(2), 215–226 p.

Libby, W.J., and Jund, E. 1962. Variance associated with cloning. Heredity, 17: 533–540.

Maguire, D. A., Mainwaring, D. B., Rose, R., Garber, S. M., & Dinger, E. J. 2009. Response of coastal Douglas-fir and competing vegetation to repeated and delayed weed control treatments during early plantation development. Canadian Journal of Forest Research, 39(6): 1208–1219.

Paul, A. D., Foster, G. S., Caldwell, T., & Mcrae, J. 1997. Trends in genetic and environmnental parameters for height, diameter, and volume in a multilocation clonal study with loblolly pine. Society of American Foresters, i(1), 87–98.

Pinheiro J, Bates D, DebRoy S, Sarkar D, R Core Team. 2018. _nlme: Linear and Nonlinear Mixed Effects Models_. R package version 3.1-137, <URL: https://CRAN.R-project.org/package=nlme>.

Prudham, S. 2003). Taming trees: Capital, science, and nature in pacific slope tree improvement. Annals of the Association of American Geographers, 93(3): 636–656.

R Core Team. 2013. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL http://www.R-project.org/.

Rehfeldt, G. E. 1991. Breeding Strategies for *Larix occidentalis*: Adaptations to the biotic and abiotic environment in relation to improving growth. Journal of Experimental Psychology: General, 136(1): 23–42.

Schmidt, W.C. & Shearer, R.C. 1991. Larix Occidentalis Nutt. In R. M. Burns & B. H. Honkala (Ed.), Silvics of North America, Washington: U.S. Department of Agriculture, Forest Service. 160-170.

South, D. B., & Miller, J. H. 2007. Growth response analysis after early control of woody competition for 14 loblolly pine plantations in the southern U.S. Forest Ecology and Management, 242(2–3): 569–577.

Strothmann, R. O. 1966. The influence of light and moisture on the growth of red pine seedlings in Minnesota. Science.

United States Department of Agriculture, Natural Resources Conservation Service. 2006. Land resource regions and major land resource areas of the United States, the Caribbean, and the Pacific Basin. U.S. Department of Agriculture Handbook 296.

US Climate Data. 2019. Version 2.3. Programming and Design by Your Weather Service. Retrieved from https://www.usclimatedata.com/

Wheeler, N. C., Steiner, K. C., Schlarbaum, S. E., & Neale, D. B. 2015. The evolution of forest genetics and tree improvement research in the United States. Journal of Forestry, 113(5): 500–510.

Wickham, H. 2016. ggplot2: Elegant Graphics for Data Analysis. Springer-Verlag New York.

Appendix A: Out-planting site maps

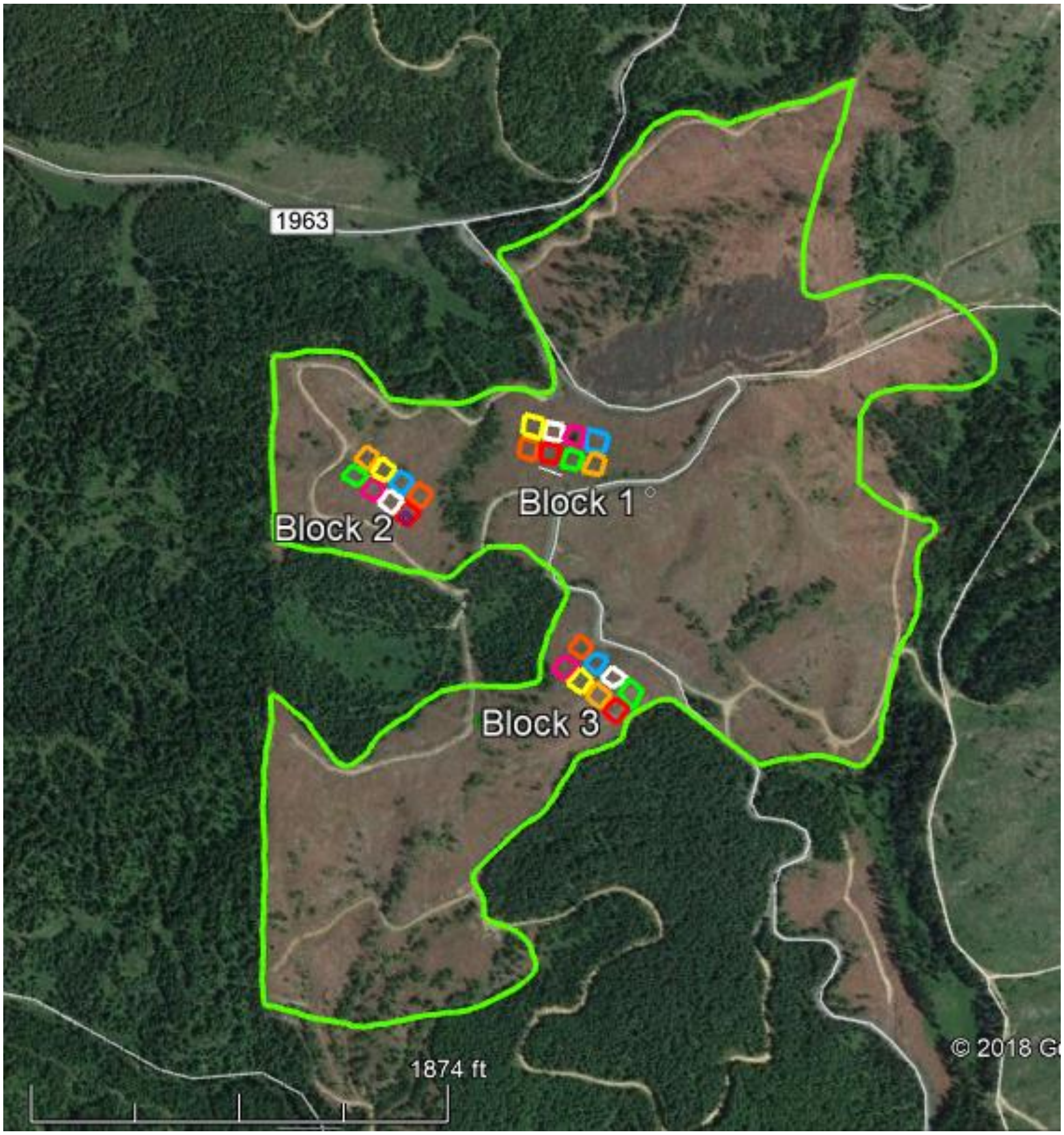

Figure A. 1. Map of block locations at Threebear, PotlatchDeltic site.

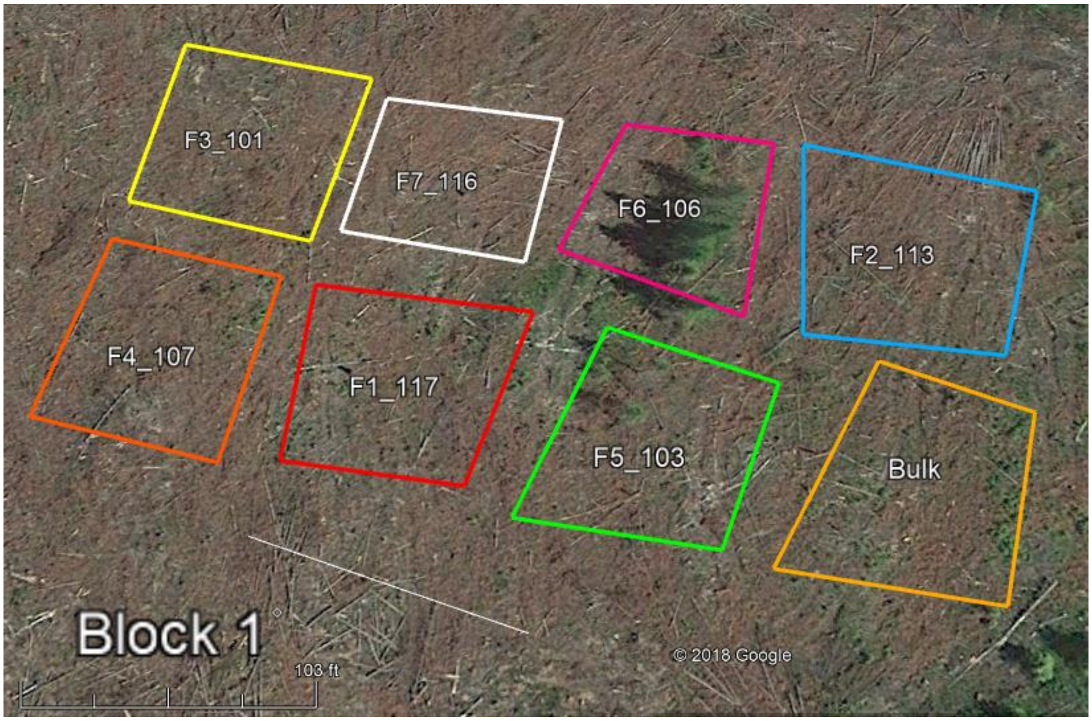

Figure A. 2. Potlatch block 1 plot map with corresponding colors, family code and number.

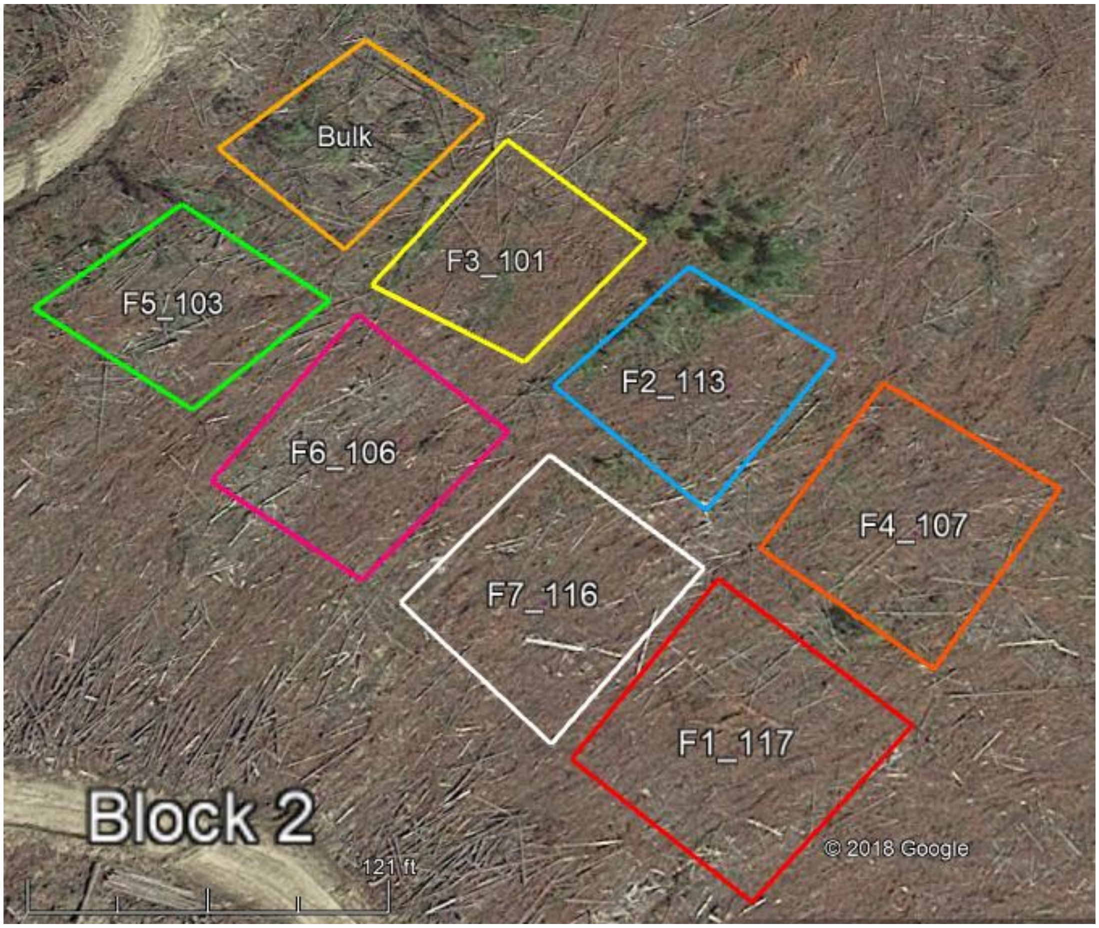

Figure A. 3. Potlatch block 2 plot map with corresponding colors, family code and number.

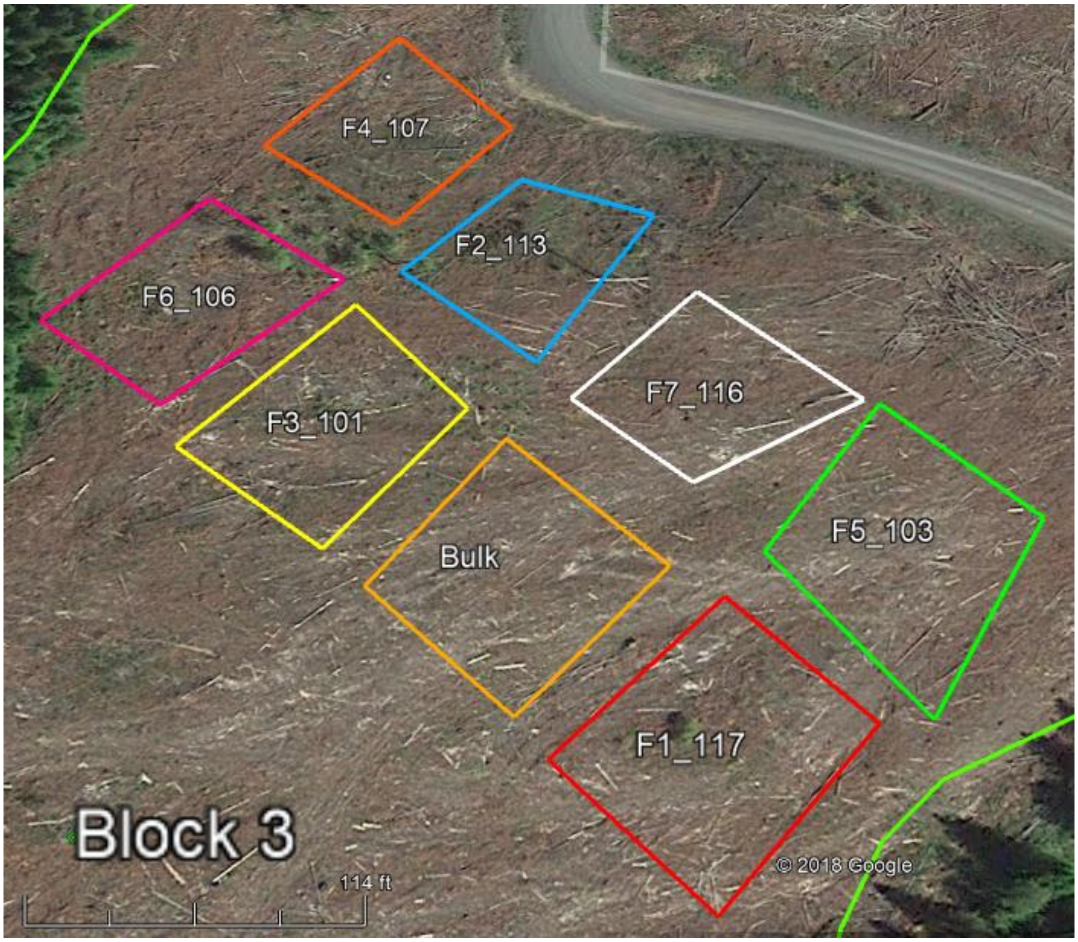

Figure A. 4. Potlatch block 3 plot map with corresponding colors, family code and number.

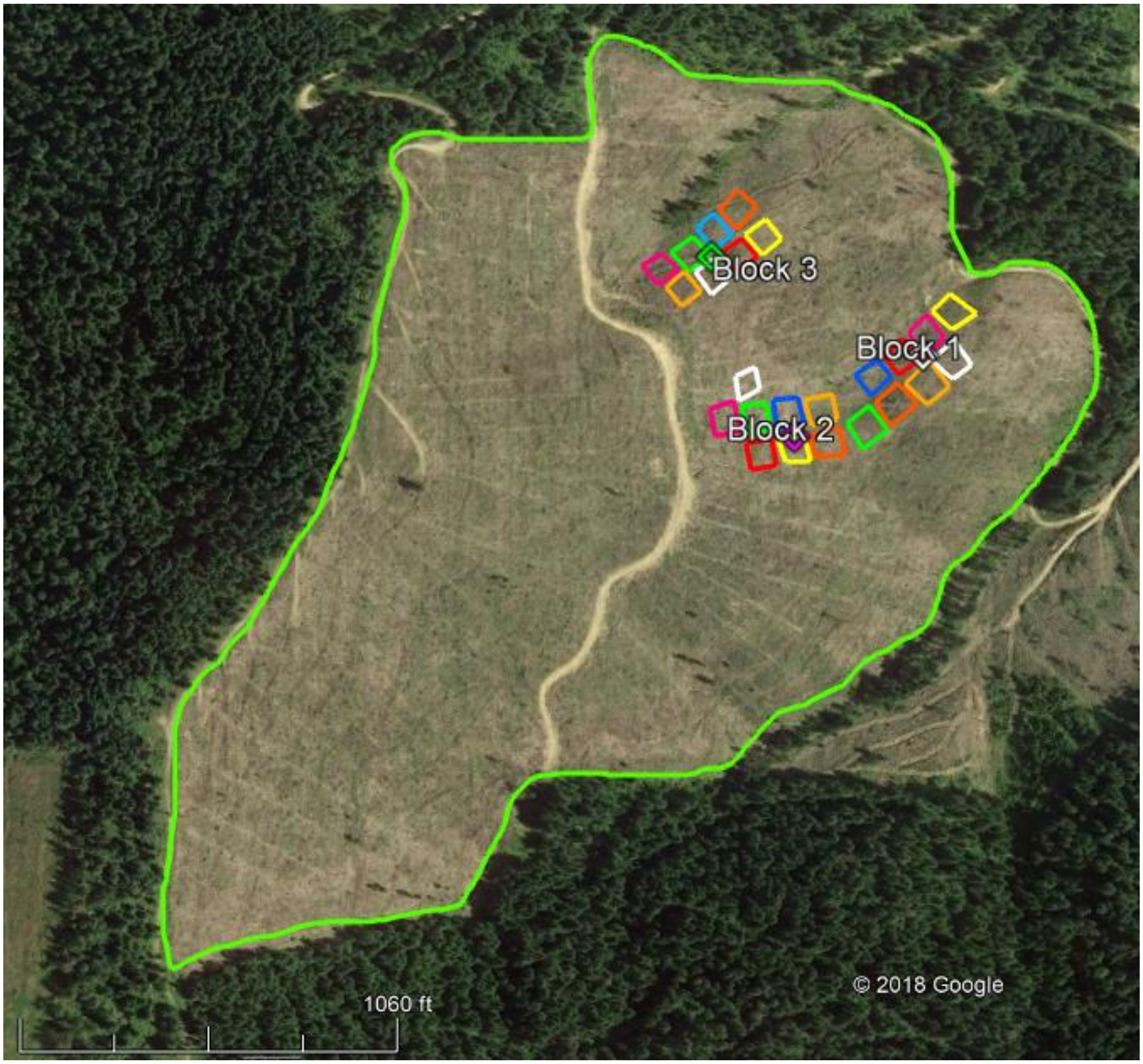

Figure A. 5. Map of block locations at Low Crutch, Stimson Lumber Co. site.

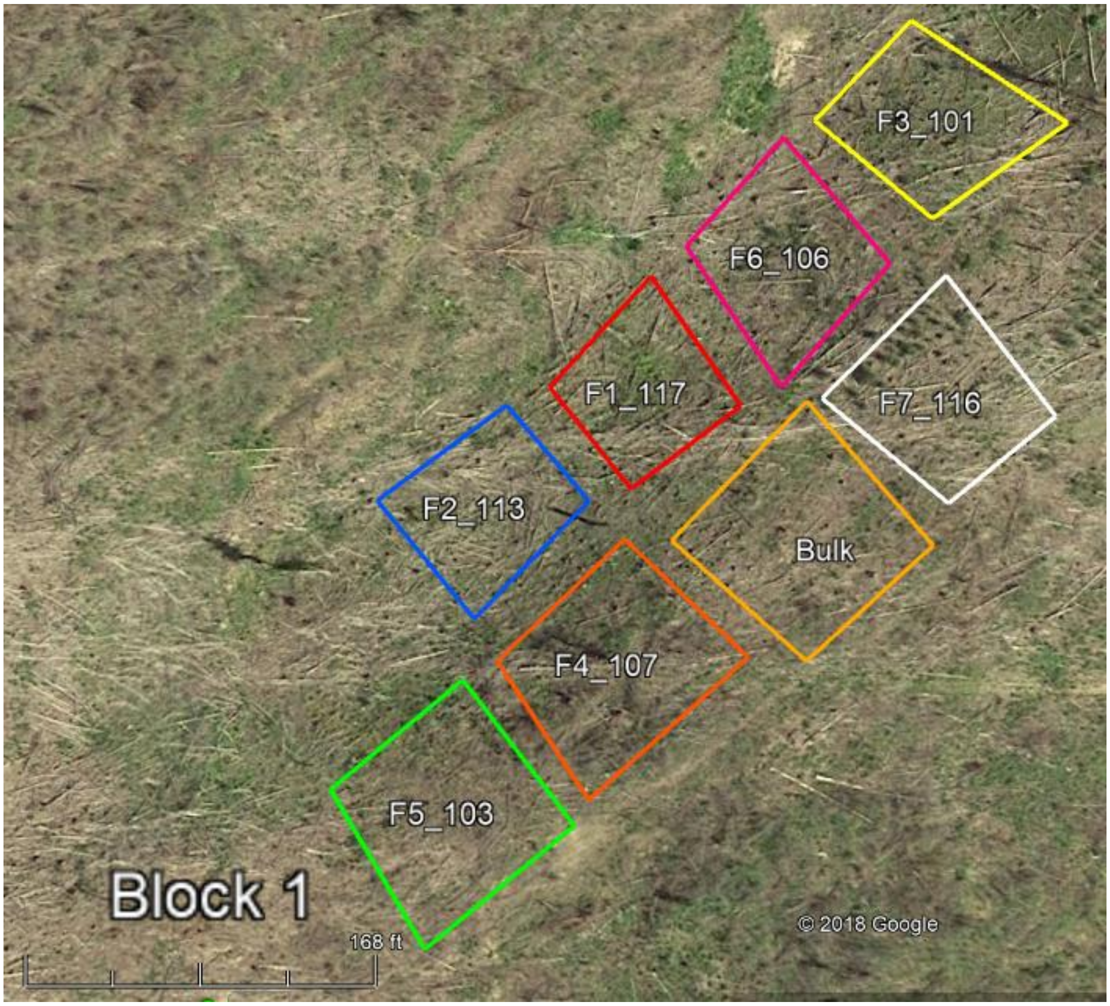

Figure A. 6. Stimson block 1 plot map with corresponding colors, family code and number.

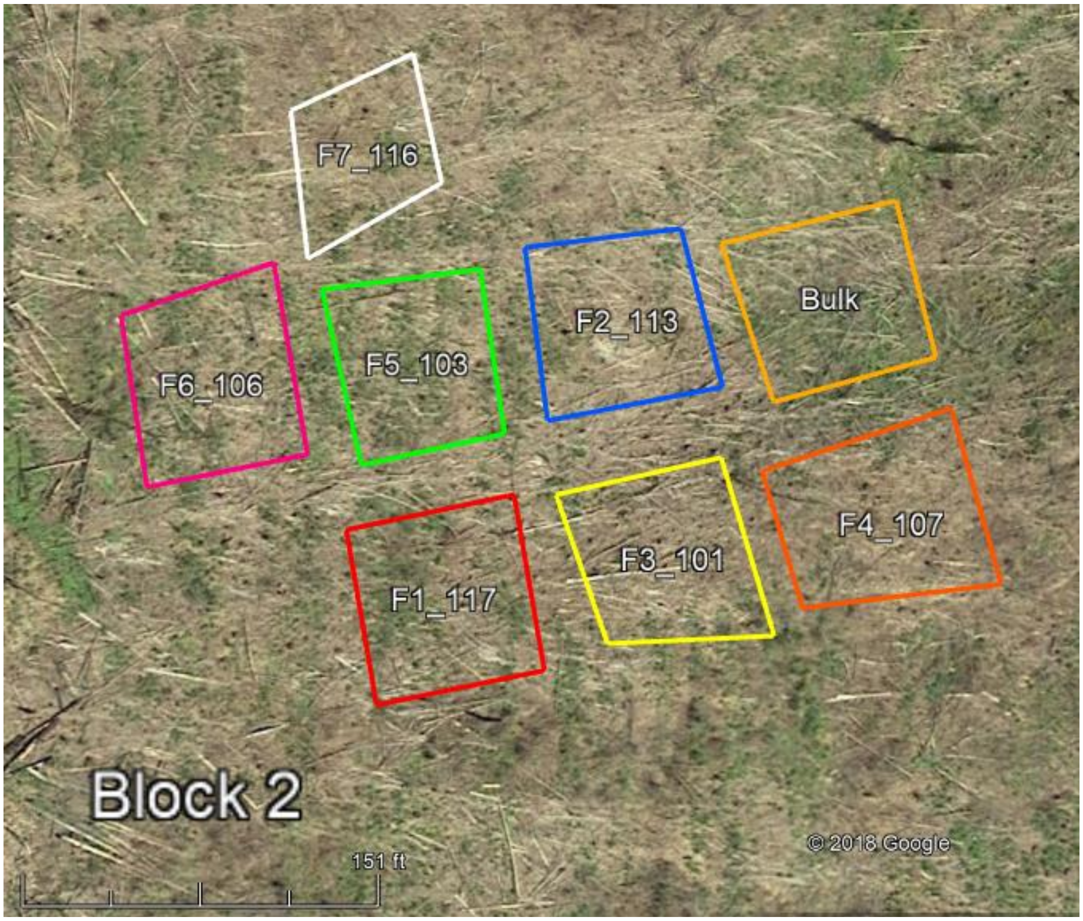

Figure A. 7. Stimson block 2 plot map with corresponding colors, family code and number.

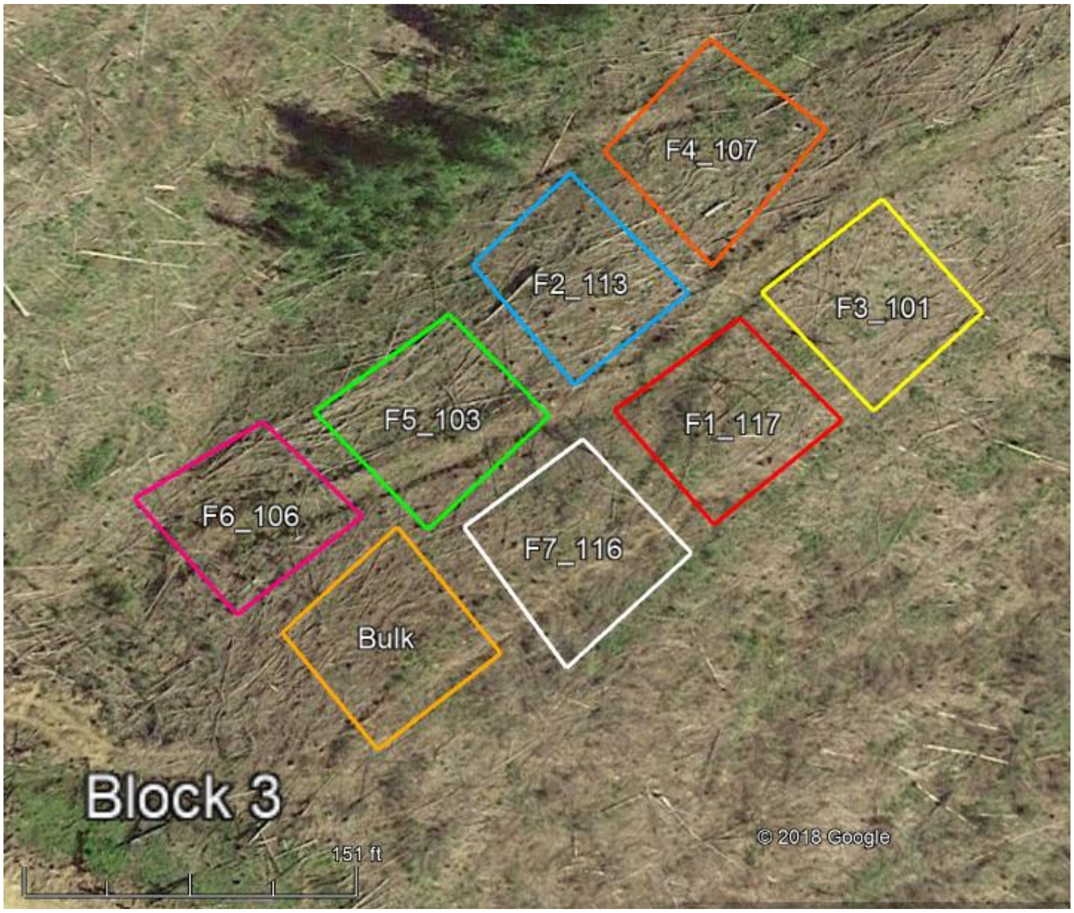

Figure A. 8. Stimson block 3 plot map with corresponding colors, family code and number.

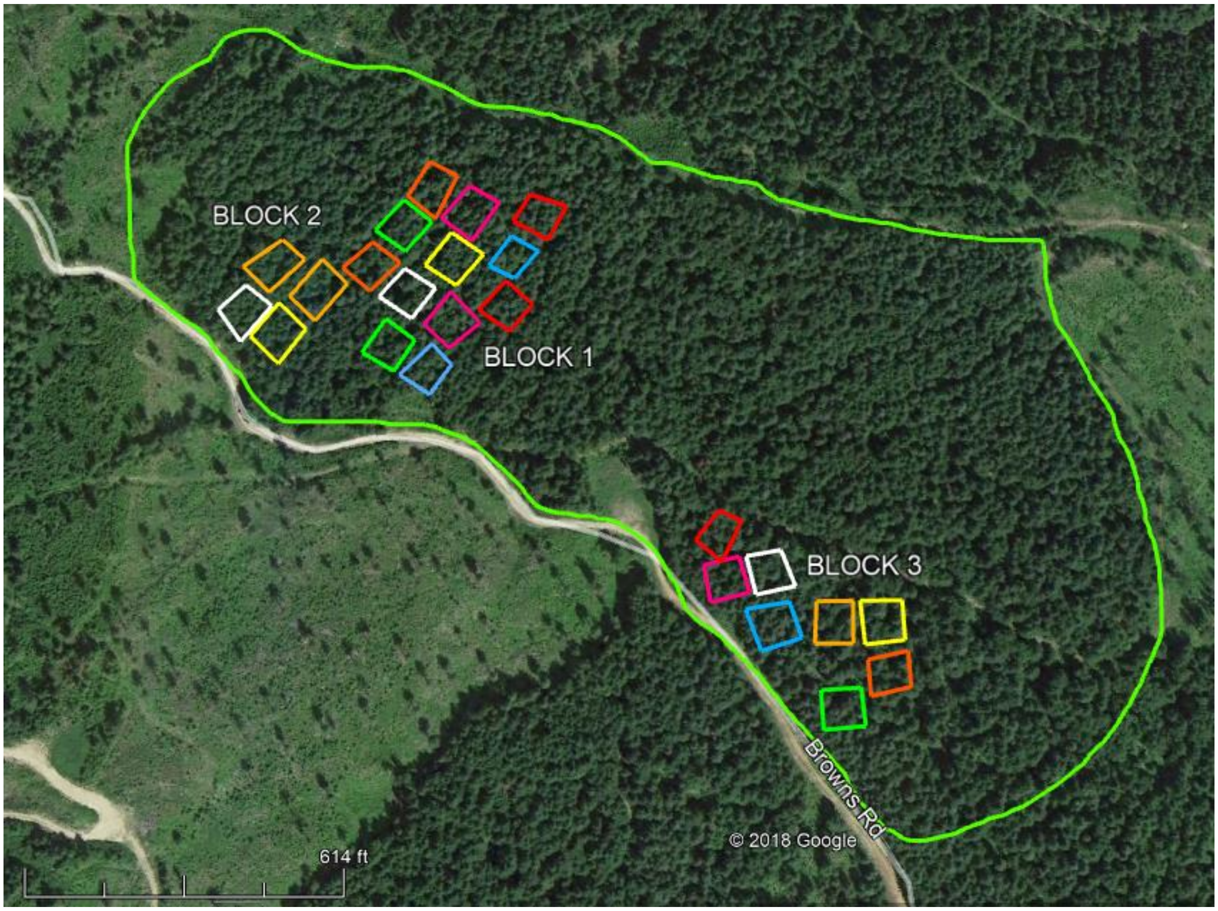

Figure A. 9. Map of block locations at Scary Berry, Molpus Woodlands Group, LLC site.

Figure A. 10. Molpus block 1 plot map with corresponding colors, family code and number.

Figure A. 11. Molpus block 2 plot map with corresponding colors, and family codes.

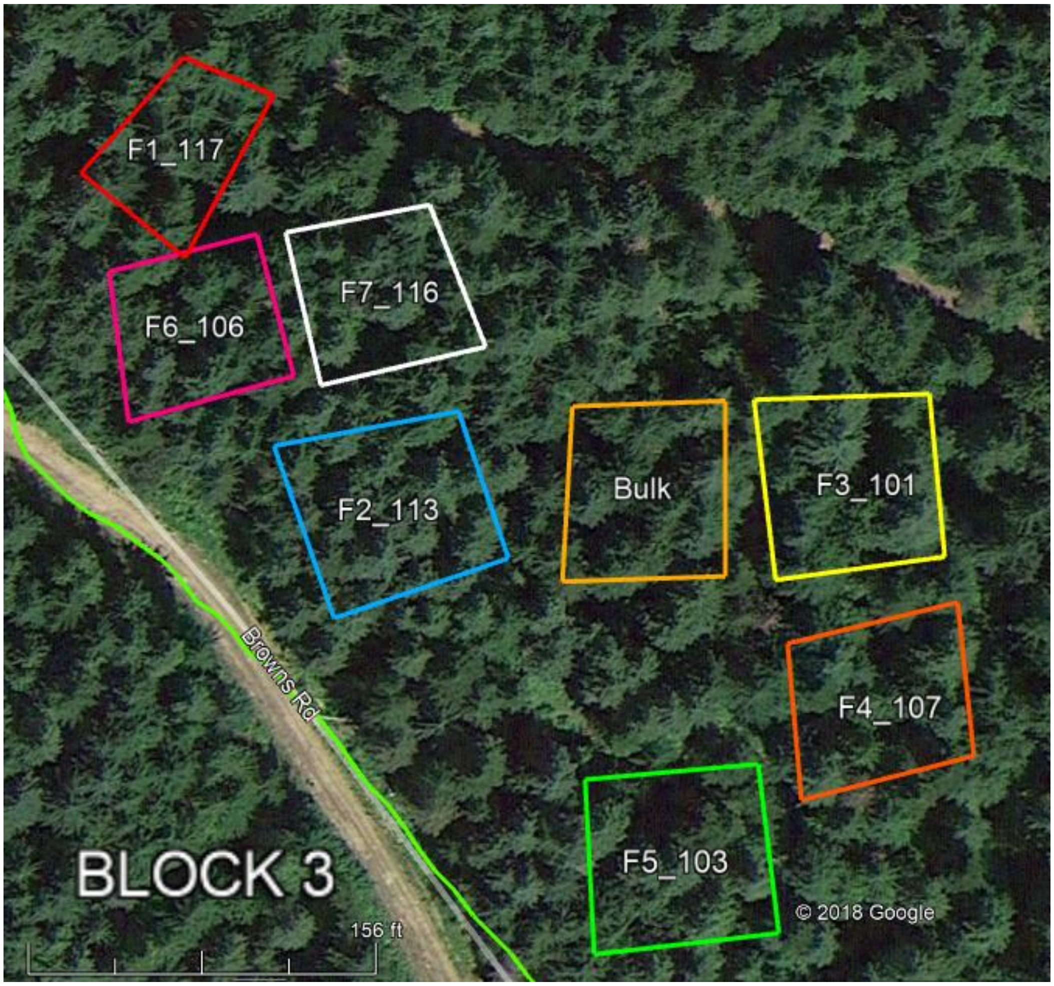

Figure A. 12. Molpus block 3 plot map with corresponding colors, and family codes.

EPILOGUE

The results from Chapter 2 and Chapter 3 revealed interesting trends when viewed as separate works, but they tell a more compelling story when viewed together. Chapter 2 results were particularly interesting given that family was a significant factor to all morphological aspects, and despite having no family effect on the out-planting seedlings, there were still cohesive findings in the morphology that correlate to the drought experiment. In terms of height growth, family 6 displayed significantly different growth than the other families. Not only did it have significantly lower absolute height in the well-watered treatment and the severe drought treatment, but it also had the lowest average height growth at Molpus and Stimson. On the other hand, when it came to Potlatch, family 6 had the highest average height growth but the lowest survival. The parent tree for this family originated from an area of low elevation, high mean max temperature, and high annual precipitation. Based on the literature, it would appear that slow growth is an adaptation for long-term drought tolerance, and is also a common trend of seed lot originating from low elevation provenances. However, the fact that family 6 had low survival at Potlatch and low growth in the severe drought treatment, more so indicates that family 6 is a poor performing seed lot regardless of conditions and may even be frost intolerant. The implications of this could support discontinued breeding of this genotype within the Kalamalka seed orchard.

Growth results were drastically different for family 3 seedlings in both the drought experiment and in the out-planting, which may be an estimation of future field performance. Family 3 seedlings had highest height growth at both Molpus and Stimson, and had high absolute heights in the well-watered and severe drought treatments. The parent tree for this seed lot originated form an area of high elevation, low mean max temperature, and low mean annual precipitation. This is why family 3 seedlings outperformed all others when placed under favorable conditions, as well as drier conditions. Family 3 seedlings are able to take advantage of available moisture and maintain rapid height growth and biomass production. Even under drought conditions they can maintain higher averages. All of this suggests that this genotype will eventually outperform the other seed lots in the field trial.

Ultimately, all of these assumptions are limited to the results of first year growth. Not only were seedlings still influenced by the nursery holdover effect, but the 2018 growing conditions were extremely mild compared to previous years. If conditions had been like those during the 2015 summer drought, we might have seen drastically different results. Based on the nursery experiment, it can be presumed that family would have been a significant factor to morphology in the field trial. Drought conditions were able to encourage genotypic differences in first year growth, and most likely would have done the same to second-year seedlings. I believe that family 3 would have out-performed the other seed lots if it had been an extreme summer. It is important for landowners to know how a seed lot performs under stressful conditions, more than mild ones, because a crop is a long-term investment. That is why this study will have big implications for landowners and why the current results should be viewed as the preliminary findings in a long-term study.