

ABSTRACT

MAYNOR, JESSICA ANN. Provenance Variation in Biomass Potential for Improved Loblolly Pine (*Pinus taeda* L.) Families at a Piedmont Site in North Carolina (Under the direction of Dr. Steven E. McKeand and Dr. Fikret Isik).

Loblolly pine is the most economically important forest tree in the Southeastern United States. In its extensive range, it has natural variation across many different environments, and many geological races or provenances have evolved. Loblolly pine is grown on millions of hectares across the Southeast, because of its rapid growth and responsiveness to silvicultural and management inputs. Considerable genetic differences within the species exist for growth, stem form, and wood quality traits that influence biomass/biofuel production. By planting genetically superior trees with desirable biomass/biofuel traits, it is possible to dramatically increase the amount of biomass and sawtimber produced at any given site. Ten of the fastest growing loblolly pine families from each of two provenances, Atlantic Coastal Plain and Piedmont, were tested for their bioenergy potential. Although gains can be achieved, there are some associated risks with seed source movement. This study compared loblolly pine families from two provenances in a field trial near Butner, NC.

At age three years, a major ice storm hit the study site, and as expected, the more southern, Atlantic Coastal Plain seedlings had higher frequency of main stem breakage (0.09) than the local Piedmont families (0.06). Coastal families also had higher incidence of crown damage (branch breakage) (0.33) than most Piedmont families (0.25). Odds ratio was used to examine these differences in damage. The biggest factor affecting main stem breakage was incidence of forking. Forked trees were 2.5 time more likely to experience damage than a non-forked tree, and the Coastal trees had higher incidence of forking (0.26) than the Piedmont

trees (0.19). Wood quality measurements, wood density and outer wood stiffness, did not explain any additional variation in ice damage.

At age six years, Coastal families produced significantly more total volume (3.3 m³/ha) than Piedmont families. However, when examining the sawtimber potential of the trees, Piedmont families produced significantly more wood with potential for sawtimber (3.7 more m³/ha) than Coastal families. Differences in volume were also significant at the family level (Pr = 0.023). Appropriate genotype selection will depend on the management and harvest regimes. For biomass plantations, risks can be minimized due to shorter rotation length, allowing for a high-risk Coastal genotype to capture the greater gains in volume. For a sawtimber stand, more conservative genotype selection should be used to ensure good stem quality at the end of a longer rotation. Understanding the trade-offs with different genotypes and harvest regimes are essential to ensure profitable returns from the plantation.

Height estimation from Unmanned Aerial Systems (UAS) is an ever-increasing interest in natural resource fields. With the use of PhoDAR from drone imagery in combination with LiDAR data, we produced high-resolution digital surface models over two test sites. After individual tree identification, we computed individual tree heights on a young biomass plantation of loblolly pine. PhoDAR derived heights were compared with field measurements. Of the 7,200 study trees, the software was able to detect 7,143 trees. Overall correlations between field measured heights and PhoDAR derived heights was low (0.18). Site 1 (reps 1-4) had a very low correlation (-0.10), but rep 5 at Site 2 had a strong correlation (0.79). The difference in correlation was attributed the “bowl effect” commonly noted in some digital elevation models from vertical UAS imagery. With proper ground

control and techniques to mitigate such broad-scale deformations, the use of PhoDAR derived measurement can become an efficient method for assessing field trials.

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Provenance Variation in Biomass Potential for Improved Loblolly Pine (*Pinus taeda*)
Families at a Piedmont Site in North Carolina

by
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DEDICATION

For my family for always supporting me and believing in my ability to take on anything in life. Dad, I found my love for the outdoors with you as a child. I can still remember every trip to the hunting club, walk around the woods, and my favorite, going to work with you. I work so hard to make you proud. You are forever my hunting buddy. Maddie, growing up with you as my big sister taught me so much of what I still carry with me today. I saw your strength, determination, and need for adventure, and I strived to be just like you. Mom, there are no amount of words I could write to express my love for you. I will forever be your baby, and you will forever be my home.

For my loving husband, Billy, who has been my rock since sophomore year of my undergraduate career, you are my biggest supporter. This work would not be possible if it was not for the encouragement and unconditional love you provided throughout this journey. For that, I am happy to be spending my life with you. Thank you.

BIOGRAPHY

Jessie grew up in the Piedmont of North Carolina, in a small rural town. Her father was a County Ranger with the Forest Service, while her mother was a horticulturist at the North Carolina Zoological Park. Little did she know playing outside as a young girl, put her career path in place.

Jessie obtained her bachelor's degree of science in Fisheries, Wildlife, and Conservation Biology at North Carolina State University in Raleigh, NC. As an undergraduate she explored many areas of natural resources through internships and studies abroad, working with local land trusts, red-cockaded woodpeckers (*Leuconotopicus borealis*), northern bobwhite quail (*Colinus virginianus*), the wildlife resources commission, and traveling to Andros Island studying mosquitofish (*Gambusia hubbsi*). Her love for wildlife was complemented with her love for forests that house these species, leading her to this journey.

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Chapter 1. Review of Literature on Bioenergy, Loblolly Pine Potential as a Biomass Source, and Loblolly Pine Provenance Variation

1.1. Introduction to Bioenergy

With the ever-accelerating rate of population growth, the demand for energy increases as well. The world population at the beginning of the Industrial revolution was 700 million; today the population is 7 billion (Chu and Majumdar 2012) and is expected to exceed eight billion by year 2030 (United Nations 2005). With this growth, energy demand in the Southeastern United States is expected to increase at a rate of 1.5% annually until 2030 (EIA 2006).

Currently, most energy demands are being met with the use of fossil fuels such as oil, coal, and gas. The harvest of these fuels is expensive and is having detrimental effects. The Intergovernmental Panel on Global Change (IPCC 2001; 2007) has confirmed that anthropogenic factors are directly correlated with increase greenhouse gases in the atmosphere (Dincer 2000). These issues in combinations with the 1970s energy crisis has led to a call for a switch to alternative forms of energy, particularly renewable resources (Kantavichai et al. 2014).

Renewable energy sources are energy resources that can be derived from nature and will replenish their reserves over time. These energy sources include solar, wind, biomass, geothermal, ocean currents, and hydropower energy (National Renewable Energy Laboratory

2008). Currently, renewable energy is marketed towards supplying energy for electricity, heating, and transportation (Ellabban et al. 2014 and Galik et al. 2009).

Solar energy is the most abundant renewable energy source available today. While solar energy is free from sunlight, power generation cost from the harnessed energy is exceptionally high (Panwar et al. 2011). Another major disadvantage of renewable energy such as solar, wind, hydropower, etc. is the reliability of the source. Renewable energy often relies on the weather to generate power (UNDP 2000). For example, wind energy needs winds to turn the turbine, solar energy produces the most energy on sunny days, and hydropower relies on an adequate water source.

Another form of renewable energy comes from biomass, a carbon-neutral energy source that includes agricultural crops and trees, wood and wood residuals, grasses, and other plant-derived matter (Galik et al. 2009 and Stöcker 2008). The harvesting of this biomass can produce electricity, liquid biofuel, cellulosic ethanol, and residential heating (Galik et al. 2009). Currently, the most commonly used biobased energy source in the United States is corn-based ethanol (Scott and Tiarks 2008). This first-generation bioethanol production system utilizes plants rich in carbohydrates such as sugars and starches (Stöcker 2008). These first-generation biofuel feedstocks include corn, sugar cane, wheat, barely, potato, and sugar beet (Stöcker 2008). In 2005, 96.2 million hectares of agronomic crops were harvested with corn, bean, and small grain representing roughly 87% of this area (USDA-NASS 2006). However, corn, wheat, and soybean are primary food-crops that are vital to world food security, leading to increasing debate of whether these high-input, highly-valued food crops should be used as an energy source (Hill et al. 2006). Today's standard for any biofuel to be a

viable and sustainable substitute is that it must be economical to produce in large quantities without jeopardizing food supplies, provide more energy than is used to produce it, and provide some environmental benefit such as reduction of green-house gas emissions (Johnson et al. 2007).

Trees have been studied extensively as a potential biomass substitute for corn. This focus on woody perennials as energy crops includes softwoods and hardwoods, also known as second-generation lignocellulose biomass crops (Johnson et al. 2007 and Stöcker 2008). Wood-based biomass is widely available in most parts of the world in large quantities and is inexpensive. In 2008, approximately 43 billion kWh of electricity was generated from woody biomass (US DOE 2009). Most woody biomass electricity comes from the forest products sector, and it is projected to increase to 218 billion kWh by 2030 (White 2010 and USD DOE 2009). Cellulose and hemicellulose from woody plants can produce bioethanol, and lignin offers can be converted into a broad spectrum of valuable chemicals and transportation fuels (Stöcker 2008).

A challenge to producing biofuels from biomass comes from the mechanisms used to create these biofuels. Woody biomass in production of second-generation biofuels are more expensive than first-generation (Stöcker 2008). Current conversion technologies into fuels include extraction, hydrolysis, gasification, pyrolysis, fermentation, and hydro thermal upgrading for HTU diesel (Stöcker 2008 and McKendry 2002). Hydrolysis, the most extensively studied process is used to make ethanol from hemicellulose found in woody biomass. The process requires high temperatures which increases the rate of hemicellulose sugar decomposition and equipment corrosion results in lower yields (Galbe and Zacchi 2002

and Jones and Semrau 1984). A preferred conversion method for ethanol production is pyrolysis, but the process requires high water and oxygen input, low stability, and high acidity (Stöcker 2008 and McKendry 2002). Biofuels processes need to be further improved before deployment into industry can be considered. Current fossil fuel energy production is too efficient and cheap to have a market for any biofuel production plant (Galbe and Zacchi 2002).

Hardwoods grown in short rotations typically include poplars and aspens (*Populus* sp.), *Eucalyptus* sp., sycamore (*Platanus* sp.), maple (*Acer* sp.), and sweetgum (*Liquidambar* sp.), among others (Johnson et al. 2007). General yield figures for short rotation woody crops using contemporary planting stock can produce about 11 to 27 dry megagrams/hectare per year (5 to 12 dry tons per acre per year) (Adegbidi et al. 2001 and Volk et al. 2006). The *Populus* genus (poplars, aspens, and cottonwoods) are among the most researched hardwood species for biomass production. *Populus* species may be an option for biomass in the Southeast but have high establishment costs (Tuskan 1998), require large amounts of water input (Scott and Cunningham 2011), are planted with clones that may be susceptible to insects disease on given sites (Tuskan 1998), and regeneration from coppicing may result in reduced productivity in volume production (Perala and Alban 1982 and Cobb et al. 2008). Furthermore, besides willow, coppicing is no longer recommended for any short rotation woody crop hardwood species (Tuskan 1998). Hardwood regeneration by coppice has decreased establishment costs by 13-26%, but the gains are lost on longer rotations of 6-10 years compared to replanting. In the Southeast, where loblolly pine is most prevalent,

periodic and recurrent drought may limit *Populus*' productivity in comparison to the native pine (Scott and Cunningham 2011).

Another option for a biomass crop in the Southeast may be softwoods, such as loblolly pine (*Pinus taeda*), given its abundance in the area. Of the 1 billion tree seedlings planted in the South in 2015-2016, 77% were loblolly pine (Enebak 2017). Given the importance of loblolly pine in the Southeast, it is sensible to explore it as a potential for biofuel (Li et al. 1999). Softwoods are the dominant source of lignocellulose in the Northern Hemisphere containing nearly 65% cellulose and hemicellulose that can be converted into sugars for fuels (Galbe and Zacchi 2002 and Frederick et al. 2008). Few studies have been conducted comparing hardwood and softwood for biomass production. However, Cobb et al. (2008) tested four species to compare biomass performances, loblolly pine, slash pine (*Pinus elliotti*), sweetgum (*Liquidambar styraciflua*), and sycamore (*Platanus occidentalis*). These species were tested on several sites and conditions, and from a biomass stand point, loblolly produced more volume than the others. Perala and Alban (1982) found similar results to Cobb. These studies and others created an interest on improving biomass production from loblolly pine for tree improvement programs in the Southeast. Advances in breeding and selection from tree improvement programs have improved the species' potential as a bioenergy feedstock (Johnson et al. 2007 and Talbert et al. 1985). From only two breeding cycles, tree improvement programs have made significant gains in volume, height, straightness, and reducing fusiform rust (caused by the fungus *Cronartium quercuum* f. sp. *fusiforme*) over the wild native stock (Li et al. 1999). Through selective breeding, gains as

high as 12 percent more volume per hectare and 32 percent in harvest values were seen over the wild stock in the first generation alone (Zobel and Talbert 1984 and Talbert 1982).

Loblolly pine has been identified as a species of interest for biomass production, but the feasibility needs to be examined. With current market conditions, biomass stumpage prices are not financially attractive. Research exploring the feasibility of planting loblolly pine solely as a biomass crop resulted in negative financial returns under almost every scenario (Munsell and Fox 2010). Loblolly pine's high value for timber means that biomass prices will have to rise before landowners and forestry companies consider planting the species solely as a bioenergy crop. However, mixed management stands have been tested showing promising results. When plantations are managed for both timber and biomass, maximum returns can be achieved (Scott and Tiarks 2008).

Another option is using harvest residuals as biofuel. The South generates the greatest volumes of residuals, and with the predominance of coal-fired power plants in the East, woody biomass residuals may be useful (White 2010). For many forest landowners, intensively managed pine stands have become routine practice. Returns after harvest need to cover establishment costs and also provide profit to give incentive for landowners to continue growing pines. Given the current market conditions, there is little to no incentive for a landowner to plant loblolly as a dedicated energy crop.

Although, there is no market for loblolly as biomass currently, market shifts can occur abruptly. The production from lignocellulosic to usable fuels poses another challenge to the biomass market. It is anticipated, however, that climate change policies will result in a carbon price or some form of incentive will encourage expansion of short rotation woody

crops (White 2010). That is why further research in studying loblolly pine as a potential energy source should continue. Research has identified loblolly as a potential source for biomass (Cobb et al. 2008 and Perala and Alban 1982), but quantifying such biomass still needs to be accomplished for different seed sources.

1.2. Wood Density

The most common way to quantify biomass production is through harvesting trees and oven drying for dry weight estimates, which are labor intensive and destructive to the study site (Isik and Li 2003). Zobel et al. (1969) mentioned that obtaining specific gravity estimates from wood samples allows for creation of dry weight yield tables; this is much less destructive than traditional methods, but is still labor intensive. Specific gravity is the ratio of a substance's density to the density of pure water at 4°C (Zobel et al. 1969). Traditional means for determining specific gravity of a tree involves extracting wood increment cores and measuring the volume and weight of dry wood (ASTM 1985). Although this method is accurate, it is both time consuming and expensive, because many sample trees are needed to estimate a mean for a family or for a stand of trees. An even less destructive method comes from an electronic drill that can be used as an indirect measure of wood density. The Resistograph uses a thin needle (1.5 to 3.0 mm) to drill into a live tree measuring the drill resistance from bark to bark (Isik and Li 2003). Drill resistance is given on a relative scale of 0 to 100% of maximum amplitude. These drill resistance values can be used in equations as described by Walker et al. 2018 to calculate specific gravity. This method is by far the

fastest, least labor-intensive method for obtaining relative estimates of wood density. This allows for periodic biomass estimates of a site throughout the lifecycle of the study.

1.3. Provenance Variation in Loblolly Pine

Natural variation in biomass production comes from loblolly's extensive range, from Virginia down to Florida west to Texas, which has created many geographic races (Zapata et al. 2015). Seed source testing of loblolly pine first began in 1927 with Philip C. Wakeley leading the research of seed transfer studies (Lambeth et al. 2005 and Wakeley 1944). The previous ideology was that local seed was better than seed from distant sources, with the recommendation that seed from within 100 miles of the plantation was the best (Lambeth et al. 2005). The results of Wakeley's pioneering study were contradictory to this previous line of thinking. Wakeley's work influenced the establishment of region-wide, collaborative trials throughout the Southeast (Ford 2017). The objective of these trials was to examine these profound differences among seed sources and exploit them to meet forest industry demands for better seeds (Ford 2017).

From Wakeley's findings, the Southwide Southern Pine Seed Source Study (SSPSSS) by the Southern Forest Tree Improvement Committee in 1951 was created (Lambeth et al. 2005). This study represented the cooperative efforts of tree improvement programs to understand the natural variation in growth and adaptability of loblolly pine. The early results of the SSPSSS study gave an indication that local seed sources may not be the best for growth and disease resistance (Wakeley 1961, Wells 1983, Wells and Wakeley 1966).

From this research, a map of the different provenances was created based on plant hardiness zones (U.S. Department of Agriculture 1990). There were 7 planting zones or provenances known of loblolly pine (Lantz and Kraus 1987). This information served as a guide for transfer of plant material (Lambeth et al. 2005, Schmidting, 2001). The zones were Virginia, North Carolina Coastal Plain, Piedmont North Carolina to Northern Mississippi, South Carolina to Mississippi Coastal Plain, East Texas to West Louisiana, South Arkansas to Southeast Oklahoma, and Western Tennessee, Western Kentucky, and Southern Illinois. These regions were designed based on geographic variation, physiographic boundaries, climate, vegetation, and most importantly plantation performance (Lantz and Kraus 1987). There were real advantages of using nonlocal seed sources to further gain, without any breeding involved (Ford 2017).

For moving loblolly pine material north to south or vice versa the most important climatic variable is yearly average minimum temperature (Schmidting 2001). The more common trends of seed sources come from moving loblolly pine material south to north and east to west. For this study we examine the effect of moving a southern Coastal source more inland to the Piedmont region for potentially higher productivity. Some trends found with desired traits have been growth rate is highest in the southern most seed sources, stem straightness is better in northern more slow growing sources, fusiform rust resistance is greatest in sources west of the Mississippi River, drought resistance is highest in western sources, and cold tolerance is greatest in northern most sources (Wakeley 1961, Wells and Wakeley 1966, Grigsby 1973, Wells and Lambeth 1983, and Schmidting 2001, Lambeth et al. 1984, Lantz and Kraus 1987).

There are potential benefits from planting nonlocal seed sources at a given location. The most common benefit comes from the movement of seed sources north for faster growth. For this study, we anticipate better growth rate and volume from the Coastal seed source families over the native Piedmont source. This movement does involve some risk, but can bring larger returns if successful (Lambeth et al. 2005). Risk can be as minimal as broken limbs from snow and ice or as severe as tree mortality from severe cold. Broken tops from ice or snow damage can reduce quality from a saw log to a short-length, low-grade product. The potential benefits and risks must be evaluated in context of the intended products being grown. For instance, a landowner growing loblolly pine for sawtimber may be more cautious of moving southern material too far north given the risks of cold or ice damage reducing the quality of wood harvested. While, a landowner growing wood solely as a biomass energy crop or pulp wood may make that movement for the increased growth and will be less concerned with stem quality and more concerned with decreasing stem mortality.

The impact of cold entails many aspects working together such as late growing season stress, mid-fall and early winter temperatures, and rapid falling winter temperatures accompanied by high wind and ice (Lambeth et al. 2005). Severity of ice storms can be viewed in terms of ice accumulation, the duration of accumulation, and the resulting damage (Ireland 2000). Ice accumulation, also known as glaze, is one of the most damaging in temperate regions (Bragg 2003). While ice storms are not frequent in the South, when they happen it can be devastating to millions of acres of forestlands (Halverson and Guldin 1995, White 1994, and Forgive 2001). Although loblolly pine is more hardy than many other southern pines, it remains susceptible to ice damage, some provenance more than others.

Several studies have observed the impacts of cold temperatures on Coastal seed sources in loblolly pine. Jones and Wells (1969) showed that warmer seed sources tended to have higher frequency of cold damage than did the colder inland sources. Although ice damage was heaviest among the trees from the warmer seed sources, these trees still produced greater volume than trees from the inland seed sources at 15 years. This study, as well as others, found very little mortality in both seed sources, but a significant reduction in tree form in the warmer sources (Jones and Wells 1969 and Wiesehuegel 1955).

1.4. Wood Bending Strength and Ice Damage

Jones and Wells (1969) observed significant differences in damage between sources and suggested wood strength may be a reason for those difference. Bending strength is one factor affecting ice damage susceptibility. Modulus of elasticity (MOE) or stiffness of wood is a measure of how much a material changes shape per unit of applied stress (Panshin and de Zeeuw 1970). This is especially true since wood that is cold and green or less dense has notably less resistance to breakage than warmer, more dense wood of the same species (Panshin and Zeeuw 1970 and Bragg 2003). Modulus of elasticity has been correlated to lumber grades and quality, but little research has been done correlating MOE to storm damage (Ekhard et al. 2010). The relationship among bending strength and ice damage may provide ability to predict how a provenance or family will respond ice. In loblolly pine, MOE is affected by wood density, corewood micro-fibril angle and stiffness are under genetic control (Roth et al. 2007, Aubrey et al. 2007, and Raymond et al. 2007). By selecting families or provenances based on stiffer wood, could ice storm damage be reduced?

Traditional measurements of MOE involve a static bending test in which a wood section of fixed dimensions and moisture content is subjected to a known stress load (Mora et al. 2009). This method is costly and requires destructive sampling of trees (Raymond et al. 2007). Measurements of MOE with acoustic wave devices such as TreeSonic™ are a good alternative to traditional bending tests (Schimleck et al. 2003). The Tree Sonic™ measures acoustic stress wave time of flight (ToF) from one probe to another on a standing tree. Time of flight can be converted to velocity in meters per second (m/s) to be used as an indirect measure of outer wood stiffness i.e. modulus of elasticity; a higher velocity implies greater wood stiffness (Schimleck et al. 2003). A greater modulus of elasticity means the wood will bend more allowing for recovery after an ice storm (Amateis and Burkhart, 2015). Ekhard et al. (2010) found a strong genetic correlation between time of flight measurement from Tree Sonic and MOE (0.73) similar to others results (Krumar et al. 2002 and Matheson et al. 2002). This means acoustic velocity measurements can be used as nondestructive evaluation of wood stiffness in standing trees (Mora et al. 2009).

1.5. Other Characteristics Affecting Ice Damage

Ice damage susceptibility may also be affected by diameter class, particularly related to main stem breakage. Loblolly is most susceptible to main stem breakage when diameter range is between 12-25 cm (Bragg et al. 2002, 2003, 2004, Amateis and Burkhart 1996). Trees with smaller diameters experience stem bending by being root-sprung, when roots loosen and allow the tree to lean from the base (Amateis and Burkhart 1996). Whereas the larger trees will more likely snap than bend, experiencing branch loss and terminal leader

breakage, but little main stem breakage (Amateis and Burkhart 1996). Correlations between smaller trees and higher modulus of elasticity have been reported in Bragg 2003.

Stem forking can also affect a tree's susceptibility to ice damage as well. A fork is a bifurcation in the trunk of a tree that gives rise to two terminal leaders roughly equal in size and diameter (Zobel and Talbert 1984, Xiong et al. 2010). Jones and Wells (1969) noted that after an ice storm trees with forks tended to have more damage, but they did not take measurements to verify their observations. Amateis and Burkhart (1996) did test this theory, finding that forked trees were more likely to be damaged than single-stemmed trees. They attributed their results to the irregularly shaped crowns and weaker upper stems that made them more susceptible to ice damage (Amateis and Burkhart 1996). It is reasonable to assume that predisposing problems like crooked stems, branch forks, and fusiform rust stem galls magnified the severity of damage given the wood quality is already damaged or weakened (Irland 1998).

Another factor that may contribute to ice storm damage is crown shape. Aubrey et al. (2007) found that damage from an ice storm was more frequent in trees that were taller with larger diameters and crown shape. Guo (1999) found similar results that crown diameter was positively correlated with the likelihood of breakage. This may be due to increased surface area for ice to accumulate or more growth in the crown means less biomass is being allocated to the roots (Boerner et al. 1988). Ireland (2000) noted that lopsided crowns as well as large crowns also affect ice storm damage. Individual trees most likely to survive ice accumulation have symmetrical crowns (Bragg et al. 2003). Crowns may vary due to provenance and environment and may help explain differences in ice damage frequency. Existing methods

for measuring crown diameter and density use leaf area. Leaf area for individual trees are estimated through the use of allometric relationships between stem diameter and cross-sectional area (Roberts et al. 2005). However, this method is expensive and time consuming making this method to sample large areas impractical. A new technology to estimate crown dimensions is using photo derived lidar. Diaz-Varela et al. (2015) was able to assess crown characteristics of olive tree using high-resolution UAV imagery. This technology is expensive, but the amount of area that can be measured is much greater than traditional means and requires less time. Crown density and shape can be assessed using photo derived lidar that may provide insight into ice storm susceptibility among different provenances of trees.

Disease may affect susceptibility of ice damage through weakening wood quality or putting added stress on the tree. One of the most common and damaging disease affecting loblolly pine is fusiform rust (Geron and Hafley 1988). Fusiform rust creates enlarged galls on the lower stem and branches weakening the wood and affecting the quality of the lumber. There is very little literature testing the affect fusiform rust on wood strength. Belanger and others (1996) found that there was little association between ice breakage and occurrence of rust galls on the trees; noting most of the breakage was high in the canopy and rust was low on stems. The study trees were 19 to 22 years old and may have been too tall and too old to be affected by the ice. Younger trees may be more affected by the disease than older trees. Breakage at the rust gall is common and can amount to large financial losses in a plantation setting (Powers et al. 1974). Tree wounds from ice damage may permit infection from

diseases and other damaging agents further reducing the quality of the trees (Bragg et al. 2003).

1.6. Summary

Loblolly pine has the potential to be a biomass source based on its value and extensive variation. Provenances show variation in growth rates (Lambeth, et al. 2005), susceptibility to fusiform rust (Schmidting, 2001 and Li et al. 1999), specific gravity (Isik and Li 2003), and cold tolerance (Lantz and Kraus 1987). There may be potential benefits from planting nonlocal seed sources for biomass production, but there is also associated risks from cold/ice damage.

The primary objective of this study is to evaluate the two provenances (Atlantic Coastal Plains and Piedmont) for their biomass production. We expect Coastal families to grow faster, but at this colder Piedmont site near Butner, NC, the risk of cold damage is higher (Schmidting 2001). Growth, stem quality, and wood quality measurements, will be used to compare biomass productivity among provenances and families.

Another objective of this research project is to analyze the effects an ice storm had on the trees from two provenances of loblolly pine, Atlantic Coastal Plains and Piedmont. We hypothesize that the Coastal provenance will have higher incidence of damage given that it is less adapted to the Piedmont site. Additionally, we hope to explain the observed damage between the two provenances by assessing bending strength by taking Tree Sonic acoustic measurements for wood stiffness and analyzing stem traits such as presence of forking and

fusiform rust. This will allow for assessment on risk/reward tradeoffs of planting faster-growing provenance that is not well-adapted to the region.

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Chapter 2. The Response of Coastal and Piedmont Seed Sources of *Pinus taeda* to Ice Storm Damage at age Three Years

Abstract

We compared the response of 20 loblolly pine families, 10 each from the Atlantic Coastal Plain and Piedmont provenances, to an ice/snow storm that occurred after the third growing season. The study was a randomized split plot experimental design composed of 10 experimental blocks. Each family was nested within provenance and randomly assigned to a 36-tree square sub-plot. As expected, the more southern Atlantic Coastal Plain families had higher frequency of main stem breakage (0.09) than the local Piedmont families (0.06) caused by the ice and snow. Coastal families also had higher incidence of crown damage from branch breakage (0.33) than Piedmont families (0.25). The biggest factor affecting damage was incidence of stem forking. Forked trees were 2.5 time more likely to be damaged than non-forked trees, and trees with fusiform rust galls were 68% more likely to have a main stem break and 60% more likely to have crown damage than trees without rust galls. Wood density and outer wood stiffness did not explain any additional variation in ice damage. This study provides further evidence that ice storms can cause significant damage to southern pine plantations if they are planted outside of their natural range, such as in northern and or/inland sites. Although ice storms are unpredictable events, there are some variables that landowners can control to reduce the risks of damage. The selection of families with low forking and good rust resistance will likely reduce the risk of damage given a major storm event.

Keywords: improved loblolly, cold tolerance, risks/rewards

2.1. Introduction

Loblolly pine (*Pinus taeda* L.) is the most economically important tree species in the southeastern United States, accounting for 77% of the 1 billion pine seedlings planted in the South in 2015-2016 (Enebak 2017). This can be attributed to its fast growth, wide adaptability, and responsiveness to silvicultural treatments. Its economic importance in the South makes loblolly pine a prime candidate for breeding programs to improve the value of the planting stock. From only two breeding cycles, tree improvement programs have made significant gains in volume, height, straightness, and fusiform rust resistance (caused by the fungus *Cronartium quercuum* f. sp. *fusiforme*) over the wild native stock (Enebak 2017 and Li et al. 1999). Through selective breeding, gains as high as 12% more volume per hectare and 32% in harvest values were observed over the wild stock in the first generation alone (Zobel and Talbert 1984).

The earliest efforts of improvement programs in the South were to understand the natural variation in growth and adaptability of loblolly pine. Seed source testing of loblolly pine first began in 1927 (Wakeley 1961 and Lambeth et al. 2005). From decades of research, seven provenances were identified stretching from Virginia down to Florida over to Texas (Lantz and Kraus 1987 and Zapata et al. 2015). These regions were designated based on difference in geology, elevation, soils, physiographic boundaries, climate, vegetation and most importantly plantation performance (Lantz and Kraus 1987). Since the early trials, most southern pine seed source studies have shown that the most important factor influencing growth and survival within natural ranges is average yearly minimum temperature at the seed source (Schmidtling 1997).

There can be significant advantages of using nonlocal seed sources to further gain, without any breeding involved. The natural variation in these seed sources can and are often exploited to capture greater gains in many traits (Lambeth et al. 1984). For example, Atlantic Coastal Plain sources have been noted for their relatively faster growth over the more northern and inland sources such as those from the Piedmont (Kegley et al. 2004). Piedmont sources, however, tend to have better stem form and increased tolerance to cold than its southern counterparts (Wakeley 1961, Wells and Wakeley 1966, Grigsby 1973, Wells and Lambeth 1983, and Schmidting 2001, Lambeth et al. 1984, Lantz and Kraus 1987).

The movement of Coastal source inland does involve some risks, but it can bring larger returns if successful (Lambeth et al. 2005). Risk can be as minimal as broken limbs from snow and ice or as severe as tree mortality from severe cold. Broken tops from ice or snow damage can reduce tree quality from a saw log to a short-length, low-grade product. The potential benefits and risks must be evaluated in context of the intended products being grown. For instance, a landowner growing loblolly pine for sawtimber may be more cautious of moving southern material too far north, given the risks of cold or ice damage that may reduce the quality of timber harvested. While, a landowner growing wood solely as a biomass energy crop or for pulpwood may take that risk for the increased growth since stem quality is less of a concern.

Cold damage can be caused by several factors including late-season growing stress, mid-fall and early winter temperatures, and rapid falling winter temperatures accompanied by high wind and ice (Lambeth et al. 2005). Severity of ice storms can be viewed in terms of ice accumulation, the duration of accumulation, and the resulting damage (Irland 2000). Ice

accumulation, also known as glaze, can be one of the most damaging climatic factors in temperate regions (Bragg et al. 2003). While ice storms are not frequent in the South, when they occur, they can be devastating (Halverson and Guldin 1995, White 1994, and Forgive 2001). Although loblolly pine is hardier than many other southern pines, some provenances are particularly susceptible to ice damage. Several studies have observed the impacts of ice storms on Coastal seed sources in loblolly pine. Jones and Wells (1969) showed that warmer seed sources tended to have higher frequency of ice damage than did the colder inland sources. Although ice damage was heaviest among the trees from the warmer seed sources, these trees still produced greater volume than trees from the inland seed sources at 15 years. This study as well as others found very little mortality in both seed sources, but warmer sources had a significant reduction in form (Jones and Wells 1969, Wieseuegel 1955).

Many factors can contribute to the type and severity of damage in forest trees (Bragg 2003). Loblolly pine is most susceptible to main stem breakage when the diameter range is between 12 and 25 cm (Bragg et al. 2002, 2003, 2004, Amateis and Burkhart 1996). Forking is more likely to affect a tree's susceptibility to ice damage as well (Zobel and Talbert 1984, Xiong et al. 2010, Amateis and Burkhart 1996 and Jones and Wells 1969). Forking may contribute to irregularly shaped crowns and weaker upper stems making trees more susceptible to ice damage (Amateis and Burkhart 1996).

Jones and Wells (1969) observed significant differences in damage between sources and suggested wood strength may be a reason for those difference. Bending strength is one factor affecting ice damage susceptibility. Modulus of elasticity (MOE) or stiffness of wood is a measure of how much a material changes shape per unit of applied stress (Panshin and de

Zeeuw 1970). Modulus of elasticity has been correlated to lumber grades and quality, but little research has been done correlating MOE to storm damage (Ekhard et al. 2010). These correlations among bending strength and ice damage may allow us to predict how a provenance or family will respond to ice.

Traditional measurements of MOE involve a static bending test in which a wood section of fixed dimensions and moisture content is subjected to a known stress load (Mora et al. 2009). This method is costly and requires destructive sampling of trees (Raymond et al. 2007). Surrogate measurements of MOE with acoustic wave devices such as TreeSonic™ are a good alternative to traditional bending test (Schimleck et al. 2003). The TreeSonic™ measures acoustic stress wave time of flight (ToF) from one probe to another on a standing tree. Time of flight can be converted to velocity as an indirect measure of outer wood stiffness, i.e. MOE, with a higher velocity indicating greater wood stiffness (Schimleck et al. 2003). A greater modulus of elasticity means the wood will bend more allowing for recovery after an ice storm. Ekhard et al. (2010) found a strong genetic correlation between time of flight measurement from TreeSonic and MOE (0.73) similar to others (Krumar et al. 2002 and Matheson et al. 2002) indicating that acoustic measurements can be used as a nondestructive evaluation of wood stiffness in standing trees (Mora et al. 2009).

Density of wood may have an impact on susceptibility to cold damage. Trees with less dense wood have a higher probability of damage than trees with more dense wood (Bragg 2003). Density of wood is related to age of wood as well, with younger trees with more juvenile wood having lower densities than older trees with more mature wood (Zobel and Talbert 1984).

Disease may also affect susceptibility of ice damage through weakening wood quality or putting added stress on the tree. One of the most common and damaging diseases affecting loblolly pine and other southern pines is fusiform rust (Geron and Hafley 1988). Breakage at the rust gall is common and can amount to large financial losses in a plantation setting (Powers et al. 1974). Susceptibility to cold damage can be a multi-faceted issue affected not only by predisposing problems such as forking, disease presence, and stem characteristics, but also seed source adaptability (Irland 1998).

2.1.1. Objectives

The objective of this research is to compare loblolly pine families from the Atlantic Coastal Plain and Piedmont provenances for ice storm damage that occurred after the third growing season. Additionally, we assess stem bending strength, wood density, stem forking, incidence of fusiform rust, and growth to determine if any of these traits could explain the underlying causes of ice damage.

2.2. Methods and Materials

2.2.1. Experimental Design and Data Collection

For this experiment, 20 open-pollinated families of loblolly pine were planted at the NC Department of Agriculture & Consumer Services field site in Butner, NC (Figure 1). Because biomass yield is an important trait for biofuel, we selected 10 of the fastest growing families adapted to the Piedmont region and 10 of the fastest growing families adapted to the North Carolina and South Carolina Coastal Plain region. The average minimum winter temperature from where the Coastal families originated is -8.8°C , and the average minimum winter temperature for the Piedmont families is -11.3°C . By incorporating the higher-risk Coastal material, we will be able to evaluate the risk/reward balance of planting faster-growing material that is not as well-adapted as the Piedmont families.

The experiment was planted in the winter/spring of 2011-12. The study was a randomized split-split plot experimental design composed of five experimental blocks, each with a thin and harvest treatment as main plots, provenances (Atlantic Coastal Plain and Piedmont) were assigned to split plots, and families (ten-open-pollinated families per provenance) were randomly assigned to 36-tree plots nested in provenance plot. Since the thinning and harvesting treatments had not been imposed, there were effectively 10 blocks in the study. A spacing density of 1.8m x 2.1m (2645 trees per hectare) was used at the site (Figure 2). Number of experimental seedlings planted was 7,200, but a total of 12,408 trees were planted in the study including buffer and border trees.

Herbicide was applied to the field to control herbaceous woody competition. The chemical imazapyr was applied to the experimental blocks at a rate of 2 ounces per acre to

control for unwanted vegetation. A 0.02% solution of bifenthrin, an insecticide, was used to prevent damage from tip moth, sawflies, and other pests that could compromise the trees. Given that the study site was formerly an agriculture field, there were issues related to soil nutrition. Site specific applications of 2% chelated iron mix solutions and ammonium sulfate were applied to targeted reps to improve soil fertility.

2.2.2. Ice Storm Events and Data Collection

Height (m), presence of fusiform rust galls, and stem forking incidence were recorded at year three before the storm damage occurred. No diameters were recorded, since many of the trees had not reached breast height (1.37 m) at the time of assessment.

During February 2015 at age three years, two severe ice/snow events hit the study site accompanied by extremely low temperatures (between 0°C and -15°C) over multiple days. Additionally, precipitation in the form of ice (ranging from 1.3 to 2.5 cm thick) created added stress on the study trees. Both provenances experienced prolonged periods of ice and cold that resulted in damage in the form of broken limbs and main stems and severe stem lean.

After the storms, damage across the site was assessed by measuring stem and branch breakage on a four-point scale. A score of 1 represented no damage (main stem intact), a score of 2 representing minor damage (loss of a branch or two), a score of 3 representing more serious damage (multiple branches broken), but the tree is still merchantable, and a score of 4 representing main crown/stem broken out (tree is no longer merchantable) (see details in Figure 3). Two characteristics of interest were created based on the breakage

scores; crown damage (yes or no) created from a storm score 2, 3, or 4, and main stem breakage created from a storm score 4 only (yes or no).

After the sixth growing season, wood quality assessments were made when trees were large enough to measure. Trees were not assessed at age three due to the small diameters and multiple branches preventing a clear side to take measurements; therefore, assessments were taken at age six. To determine modulus of elasticity of each tree, TreeSonic™ was used. The tool measures acoustic stress wave as time of flight (ToF) from one probe to another on a live standing tree. Time of flight converted to velocity in meters per second (m/s) can be used as an indirect measure of outer wood stiffness i.e. modulus of elasticity. A higher velocity implies greater wood stiffness (Schimleck et al. 2003).

An electronic drill, Resistograph, was used as an indirect measure of wood density (Isik and Li 2003). The Resistograph uses a thin needle (1.5 to 3.0 mm) to drill into a live tree with a constant force, measuring the drill resistance from bark to bark (Isik and Li 2003). Studies show there is a positive correlation between the wood density and the drill resistance (e.g. Schimleck et al. 2003 and Ekhard et al. 2010). We hypothesized that tree with denser wood would experience fewer incidence of damage than trees with less dense wood.

2.2.3. Statistical Analysis

All response variables were examined through distribution analysis. Descriptive statistics were produced, and least square means of each response variable for family and provenance were examined. The response variables height (m) and acoustic velocity (m/s),

and drill resistance (amplitude) were continuous variables while rust and forking were categorical (yes or no) variables.

The following linear mixed model was fit to understand the differences between seed sources for traits with Gaussian distribution: height, acoustic velocity assessed by TreeSonic™, and the drill resistance assessed by Resistograph. This model was also used to examine the effect of fusiform rust and stem forking on acoustic velocity (outer wood stiffness) between seed sources.

$$Y_{ijkl} = \mu + \beta_1 K + \beta_2 R + B_i + P_j + F_k + BP_{ij} + BF_{ik} + \varepsilon_{ijkl} \quad Eq. 1$$

Where Y_{ijkl} is l -th measurement of family k , in seed source j , in block i ; μ is the intercept followed by covariate stem forking (K) and fusiform rust (R). Block effect was denoted with B_i ; P_j is the i -th provenance effect, F_k is the k -th family effect, and interaction of block with provenance and block with family were modeled with BP_{ij} and BF_{ik} ; and ε_{ijkl} is the error term associated with each observation. In the model, the block effect, block by provenance, and block by family interactions were considered random. The model assumes that the residuals are normally distributed, independent of one another, with constant variance $e \sim N(0, \sigma^2)$. The model was run using the GLIMMIX procedure of SAS software (SAS Institute Inc. 2014).

Stem breakage and crown damage were assumed to have binomial distribution. For the categorical traits, a generalized linear mixed model was used to test differences between seed sources and families. The probability of stem breakage and crown damage was modeled

while accounting for covariates tree height, fusiform rust disease occurrence, stem forking, acoustic velocity, and drill resistance.

$$\log[\pi/(1 - \pi)] = \mu + \beta_1 H + \beta_2 K + \beta_3 R + B_i + P_j + F_k + BP_{ij} + BF_{ik} + \varepsilon_{ijkl} \quad \text{Eq. 2}$$

Where, π is the probability of main stem breakage, $\log[\pi/(1 - \pi)]$ is the logits, μ is the intercept or reference level followed by covariate height (H), stem forking (K) incidence, and fusiform rust (R) incidence. Acoustic velocity and drill resistance were not significant covariates and therefore were dropped from the final model. Block effect was denoted with B_i ; P_j is the i -th provenance effect, F_k is the k -th family effect, and interaction of block with provenance and block with family effects were modeled with BP_{ij} and BF_{ik} ; and ε_{ijkl} is the error term associated with each observation. The β_i are coefficients for the covariates. In the model, the block effect was considered random; block by provenance and block by family interactions were also considered random. Residuals of response variables were normally distributed and independent with constant variances $e \sim N(0, \sigma^2)$. The models were run using the GLIMMIX and LOGISTICS procedures of SAS software (SAS Institute Inc. 2014).

The seed sources (Coastal and Piedmont) were contrasted using least squares means on the probability scale and odds ratio, which is the ratio of the probability of the outcome to the probability of no outcome (SAS Institute Inc. 2014). To examine the probability of main stem breakage as a function of tree height, the probability of stem breakage as a function of tree height was plotted.

The model fit statistic receiver operating characteristic curve was produced to understand the percent of variance explained. This curve describes the diagnostic ability of a

binary response variable, in this study being main stem breakage and crown damage. The receiver operating characteristic curve is similar to the coefficient of determination statistics in a linear regression model (SAS Institute Inc. 2014). The model did not converge when block effect and its interaction terms were included in the model, because it did not explain any variance (zero variances). These terms were dropped from the final model.

2.3. Results

2.3.1. Initial Damage

The overall mortality was low in the experiment; the Coastal provenance had 3.1% mortality and the Piedmont provenance had 3.6% mortality. There were significant differences between seed sources for height, stem forking, and acoustic velocity and marginally significant differences for fusiform rust incidence ($Pr = 0.0638$), but not for drill resistance ($Pr = 0.4188$) (Table 1). The Coastal seed source was on average 0.2 meters taller than the Piedmont seed source and tended to have a higher proportion of forked trees (26.7%) than Piedmont families (19.6%) (Table 2). Incidence of fusiform rust occurrence was higher in Piedmont families (18%) than most Coastal families (13.6%).

Variance components for height, acoustic velocity, and drill resistance explained by random family effect are presented in Table 3. Family effect explained significant variation for all the traits based on the log likelihood ratio tests. Parameter estimates for stem forking was 0.0616 ($SE = 0.026$) and for fusiform rust incidence was 0.176 ($SE = 0.068$); given the ratio of estimate to standard errors, family effect explains considerable variation for stem forking and fusiform rust.

F tests for provenance effects and covariates are presented in Table 4. When adjusted for provenance effect, tree height, presence of stem forking, and fusiform rust disease incidence had significant effect ($Pr < 0.0001$) on both stem breakage and crown damage. Keeping the effect of tree height, stem forking, and fusiform rust incidence constant, Coastal and Piedmont seed sources were significantly ($Pr = 0.0163$) different for main stem breakage, but not significant for crown damage ($Pr = 0.2370$) (Table 4). The average stem breakage for

Coastal families was 9.0% and for Piedmont families was 6.4%. For crown damage, Coastal families experienced more crown damage (33%) than most Piedmont families (25%), but the difference were not statistically significant (Figure 4).

2.3.2. Odds Ratio

The odds ratios of stem breakage and for crown damage are presented in Tables 5 and 6. Fusiform rust incidence had a significant impact on occurrence of crown and stem damage. Trees with fusiform rust galls were 68% more likely to have a main stem break (Table 5) and 60% more likely to have crown damage (Table 6) than trees without rust galls.

The biggest factor affecting stem breakage was presence of forking (Table 5 and 6). The odds ratio of stem breakage and crown damage for trees with forking was almost 2.5 times higher than the odds of stem breakage or crown damage for non-forked trees. Coastal families had more damage overall (Figure 4). Coastal families had a significantly higher occurrence of stem forking (26.7%) than Piedmont families (19.6%) (Figure 5).

Height significantly affected the occurrence of crown and stem damage. With an increase in one meter of height, the probability of stem breakage increased about 37% (Table 4). When examining the probability of main stem breakage (Figure 6) as a function of tree height, the relationship follows an exponential curve. The probability of stem breakage increased as trees got taller, but smaller trees tended to experience little or no damage when they were short with small crowns. As trees get taller, e.g. after 3 meters, the probability of stem breakage increases substantially. Both the Coastal and Piedmont seed sources follow the same trend. They do not seem to be different when trees are short, but the difference

becomes noticeable when trees get taller. For example, a four-meter-tall tree from Coastal seed source has a 22% chance of stem breakage whereas a Piedmont tree of the same height would only have a 17% chance of damage.

The odds ratio for provenance effect for stem breakage was 1.249, significantly different than 1, with a confidence interval of 1.042, 1.497. When adjusting for all other covariate, trees from the Coastal seed source were approximately 25% more likely to experience stem damage than Piedmont seed source.

The traditional R^2 fit statistic is not appropriate for generalized linear models, so the Receiver Operating Curve (ROC) was used as the model fit statistic to assess the variation captured by the model. The area under the curve is the ability of the model to correctly classify trees with and without breakage. The ROC was 0.71 for the models tested, meaning approximately 71% of the variation among main stem breakage and crown damage was accounted for by the models (See Appendix B).

2.3.3. Wood Quality

To further explain the underlying reasons for differences in stem breakage between provenances, acoustic velocity as a surrogate for wood stiffness of family means is presented in Figure 7. Acoustic velocity was significantly ($Pr < 0.0001$) different between seed sources and families ($Pr < .0001$). The Coastal provenance had average of 4.48 km/s^2 , and Piedmont provenance had average of 4.12 km/s^2 (Table 2). Coastal families had higher acoustic velocity readings than did Piedmont families suggesting stiffer wood. However, acoustic

velocity did not explain any additional variation in main stem breakage or crown damage ($Pr = 0.8487$) (Table 4).

The linear mixed model (Eq. 1) was fit to investigate the effect of stem forking and fusiform rust incidence on wood quality. Seed source differences for wood density and stiffness might provide additional information about seed source differences for stem breakage and crown damage. The results showed that stem forking did not have an effect on acoustic velocity ($Pr = 0.1299$). Trees with fusiform rust incidence also did not have effect on acoustic velocity ($Pr = 0.3938$).

There was significant variation in drill resistance measurements among families ($Pr < .0001$) (Figure 8), but there were no statistical significances for drill resistance at the provenance level ($Pr = 0.139$) (Table 1, 3). Additionally, drill resistance did not explain any additional variation in main stem breakage or crown damage ($Pr = 0.7271$) (Table 4).

2.4. Discussion

Similar to other studies, little mortality was observed after the cold events (<5%) (Jones and Wells 1969, Wieseuegel 1955), but significant crown and main stem breakage was observed between sources. Coastal families had 25% more main stem breakage than Piedmont families (Figure 4). This is similar to results from Jones and Wells (1969) that showed the warmer seed sources tended to have higher frequency of cold damage than the more cold-tolerant inland sources.

Forking was the most important factor affecting the stem and crown damage. Trees with forks were 2.5 times more likely to experience stem and crown damage than those without forks. These results were similar to Amateis and Burkhart (1996) who noted that forked trees experienced higher probability of damage than single-stemmed trees. The Coastal provenance had an average of 26% forking, which was 6% greater than the Piedmont provenance average. Of the 20 families in this study, the top eight families with highest proportion of forking were from the Coastal seed source (Figure 5). This greater proportion of forked trees from the Coastal seed source could explain the differences in main stem breakage and crown damage observed in this study. The higher damage observed for forked trees could be attributed to the irregularly shaped crowns and weaker upper stems (Amateis and Burkhart, 1996).

In this study, forked and non-forked trees did not have a significant difference in outer wood relative stiffness measured by the acoustic velocity ($Pr = 0.1299$), meaning there was no differences in wood strength between forked and non-forked trees (Table 5). Although, outer wood stiffness was only taken at breast height and not necessarily where a

break occurred. Perhaps, if outer wood stiffness was taken further up the bole, differences in strength may have been detected.

The presence of fusiform rust had a significant impact on the occurrence of ice damage ($Pr < 0.0001$). This finding differs from Belanger and others (1996) who found little association between ice damage and occurrence of rust galls, noting that most breaks were high in the canopy, and rust was low on the stem. Unlike this study, Belangers' average stand age was 20 years old, whereas this study was only age three when the ice damaged occurred, possibly explaining the difference in findings. However, in our study no data were collected on the location of the break, but field crews commented that many breaks occurred at or near a fusiform rust gall, suggesting that infected trees had weaker wood and therefore broke more frequently. Although, wood stiffness was not significantly different ($Pr = 0.3938$) for trees with and without fusiform rust (Table 5), measurements were only taken on clear, non-disturbed sides, possibly preventing from differences being detected.

TreeSonic was used as a surrogate for modulus of elasticity (wood stiffness), which is the ability of a material to recover its original shape and size after a stress is removed (Panshin and de Zeeuw 1970). Families and provenances were significantly different for acoustic velocity. However, relative wood stiffness did not explain any additional variation in main stem breakage or crown damage. Wood density was not significantly different between provenances and did not explain any additional variation in main stem breakage or crown damage. This suggests that factors other than wood strength (e.g. stem forking and presence of fusiform rust galls) may have greater influence on susceptibility to stem or crown breakage.

Modulus of rupture, which measures the point at which a material can no longer support a stress and breaks, may be a better strength trait to associate with stem breakage given that modulus of elasticity is the bending strength. This assumption is supported by Amateis and Burkhart (1996) who found that stout boles are more likely to break rather than bend once stress load has exceeded its limit. Measuring modulus of rupture would have been destructive to the study trees and therefore was not assessed.

This study provides further evidence that ice storms can cause significant damage to southern pine plantations especially if Coastal seed sources are planted outside of their natural range, such as in northern and or/inland sites. Seed source selection may be influenced by different management and harvest regimes. For instance, a landowner growing loblolly pine for sawtimber may be more cautious of moving southern material too far north given that the risks of cold or ice damage may reduce the quality of wood harvested. While, a landowner growing wood solely as a biomass energy crop or for pulpwood may accept the risk of cold damage in exchange for the greater yield and will be less concerned with stem quality.

Although ice storms are not predictable events, there are some variables that landowners can control to reduce the risks of damage. The selection of families with low forking and good fusiform rust resistance will likely reduce the risk of damage given a major storm event. For a landowner, understanding the risks associated with seed source movement is crucial to mitigate damage and obtain profitable returns.

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2.6. Tables

Table 1. Analysis of variance for tree height, stem forking, fusiform rust incidence, acoustic velocity (surrogate for wood stiffness), and drill resistance (surrogate for wood density).

There were significant differences between seed sources for height, stem forking, and acoustic velocity and marginally significant differences for fusiform rust incidence, but not for differences in drill resistance.

Trait	Den DF	F Value	Provenance effect (Pr < F)
Tree height	6735	8.42	0.0037
Stem forking	7180	10.27	0.0014
Fusiform rust	7180	3.44	0.0638
Acoustic velocity	1175	9.92	0.0017
Drill resistance	1216	0.65	0.4188

Table 2. Provenance means (range for family means in parentheses) for tree height, forking, fusiform rust incidence, acoustic velocity, and drill resistance. Coastal families on average were taller, had higher proportion of forking, and greater acoustic velocities (higher wood stiffness). Piedmont families had higher incidence of fusiform rust.

Trait	Piedmont	Coastal
Tree height (m)	2.6 (2.5, 2.8)	2.8 (2.7, 2.9)
Stem forking (%)	19.6 (16.9, 22.3)	26.7 (23.3, 30.0)
Fusiform rust (%)	18.0 (14.0, 22.0)	13.6 (10.1, 16.3)
Acoustic velocity (km/s) ²	4.1 (3.9, 4.4)	4.5 (4.2, 4.8)
Drill resistance (amplitude)	12.5 (12.2, 12.9)	12.4 (12.0, 12.7)

Table 3. Variance components for family effects for continuous variables. The estimates were significantly different from zero based on the log likelihood ratio tests. Family effect explained significant variation in height, acoustic velocity, and drill resistance.

Trait	Estimate	Standard Error	Estimate/SE (t-value)	Pr > t
Height	0.0095	0.0056	1.68	<.0001
Acoustic velocity	0.0465	0.0196	2.38	<.0001
Drill resistance	0.1274	0.0689	1.85	<.0001

Table 4. Type III F-tests for fixed effects in the model. Height, forking, and fusiform rust incidence were significant as covariates for the stem breakage and crown damage. Seed sources effect was significant for stem breakage but not significant for crown damage.

Effect	DF	Main Stem Breakage		Crown Damage	
		F	Pr<F	F	Pr<F
Provenance	1	5.77	0.0163	1.40	0.237
Height	1	124.86	<.0001	251.71	<.0001
Forking	1	100.73	<.0001	91.84	<.0001
Fusiform rust	1	23.68	<.0001	15.10	0.0001

Table 5. Odds ratios for seed source and covariates for main stem breakage. Forking was the biggest factor affecting stem breakage. A forked tree was about 2.5 times more likely to experience damage than a non-forked tree.

Effect	Odds Ratio	95% Confidence Limits	
Provenance	1.249	1.042	1.497
Height	1.368	1.295	1.446
Forking	2.492	2.085	2.978
Fusiform rust	1.680	1.363	2.071

Table 6. Odds ratio for crown damage for provenance, stem height, forking, and fusiform rust incidence. Provenance effect was not statistically significant for crown damage, as shown by the confidence limit, which includes 1. Forking was the biggest factor affecting crown damage, followed by fusiform rust incidence and tree height.

Effect	Odds Ratio	95% Confidence Limits	
Provenance	1.186	0.984	1.430
Height	1.341	1.267	1.419
Forking	2.347	1.952	2.823
Fusiform rust	1.602	1.290	1.990

2.7. Figures

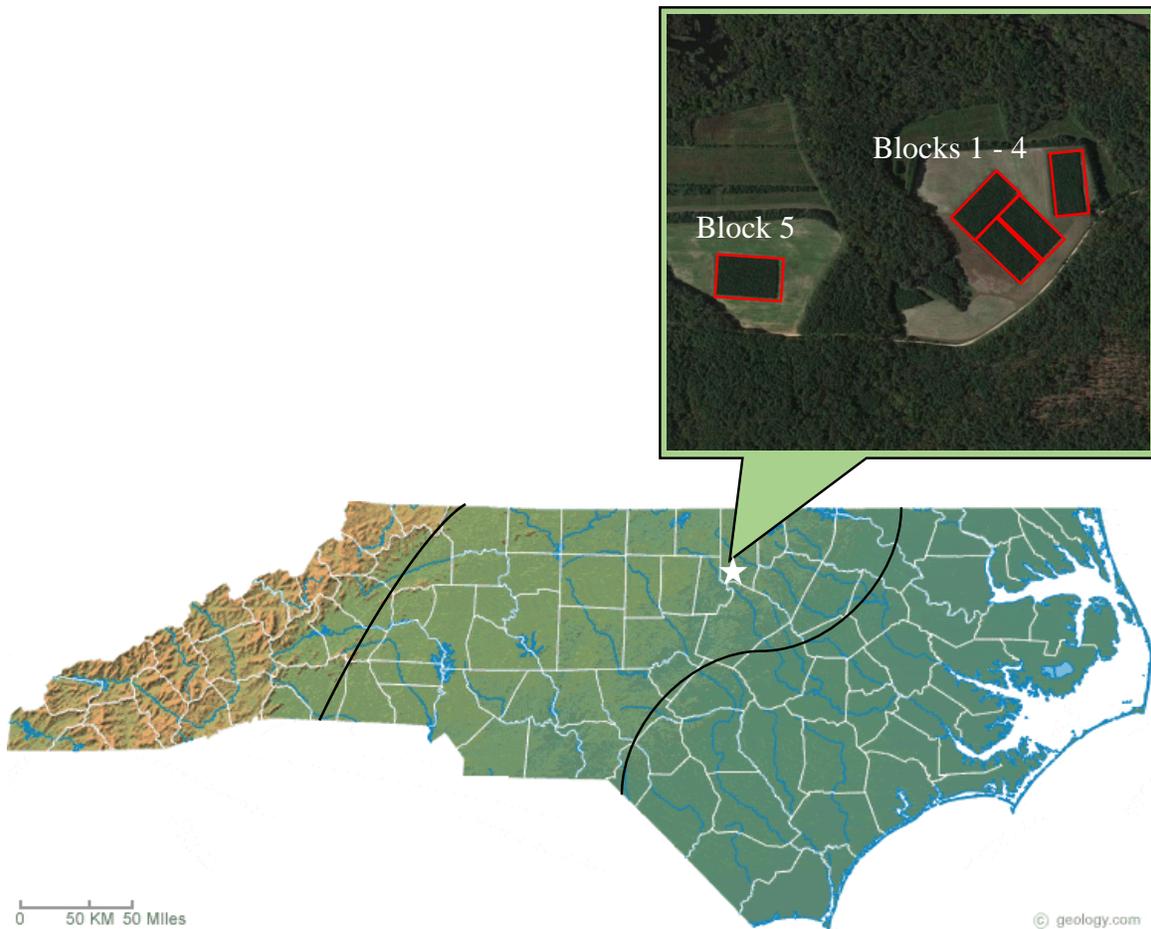


Figure 1. Layout of blocks and general location of Butner field trial in Granville County, Piedmont of North Carolina. The test site is located on the NC Department of Agriculture & Consumer Services Umstead Farm. The lines designate the division of the zones (left to right): Mountain region, Piedmont region, and Coastal region.

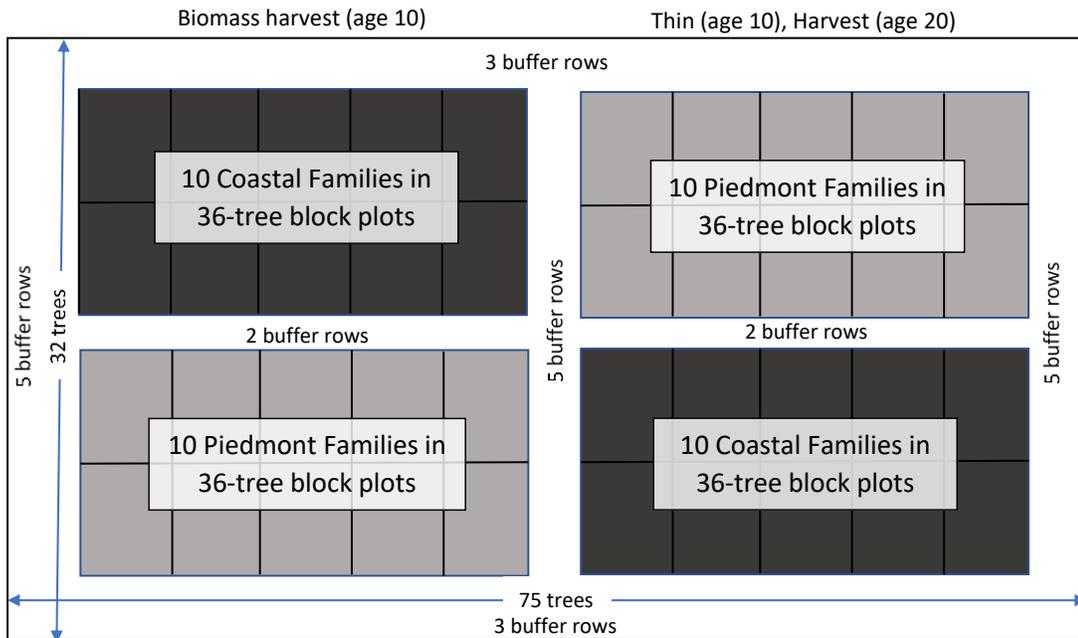


Figure 2. Layout of one experimental block of the split-split-plot experiment. Main plots were the thin and harvest treatments, and provenance was the split plot within the main plot. Ten families of each provenance were randomly assigned to 36-tree family rectangular sub-plots within each provenance plot. There were five blocks in the study, but note that the thinning and harvesting treatments have not yet occurred. Hence, there were effectively 10 blocks in the study.

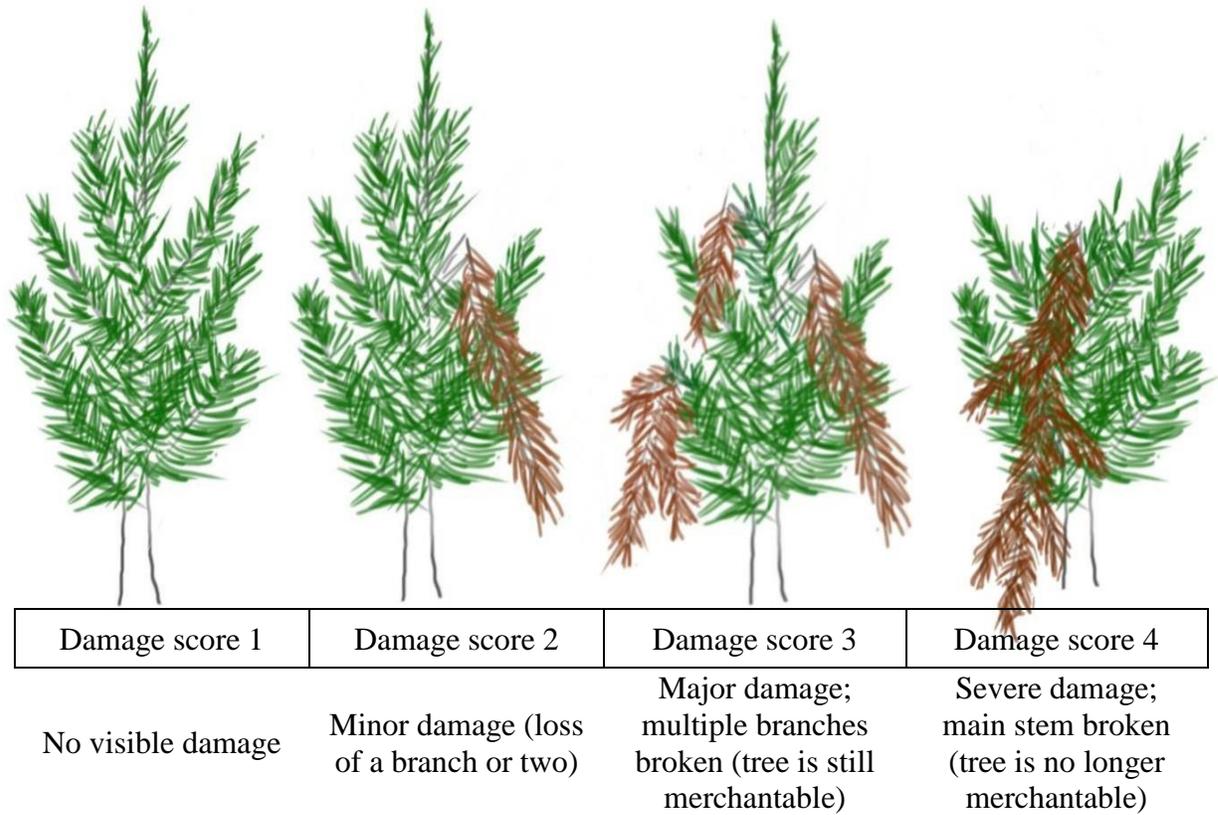


Figure 3. Storm damage scores 1 through 4 with descriptions of each classification. The categorical scoring was used to determine the degree of ice damage observed. The scoring was on a 4-point scale with 1 being the least amount of damage to 4 being severe damage.

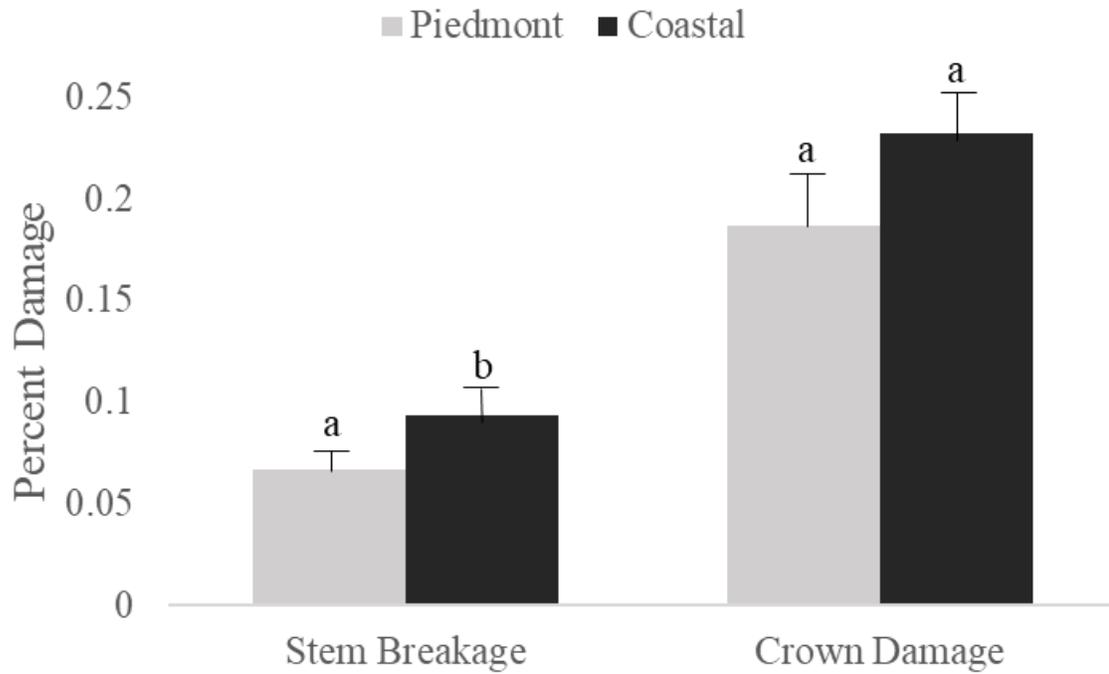


Figure 4. Percent main stem breakage and crown damage by provenance. For both crown damage and main stem breakage, the Coastal provenance had a higher incidence of damage than the Piedmont provenance, but only stem breakage was significantly different.

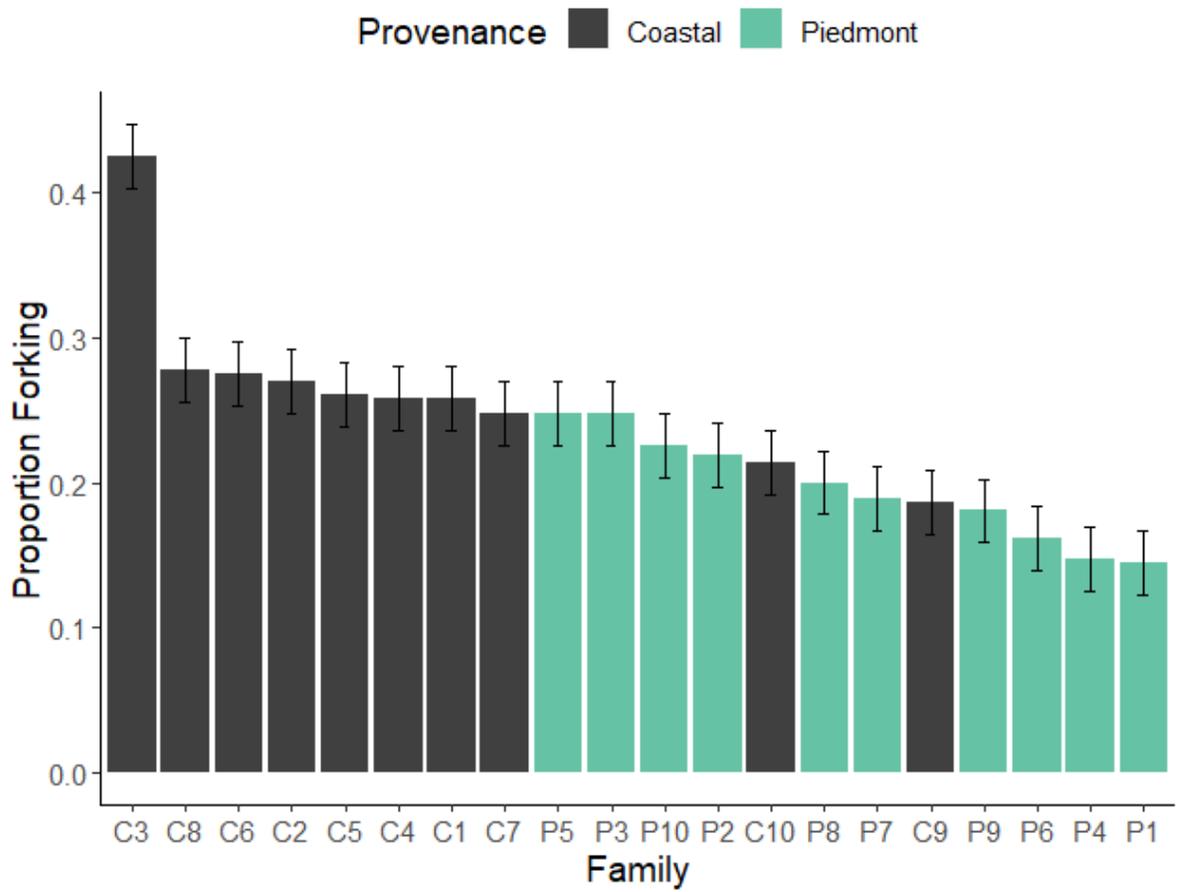


Figure 5. Coastal and Piedmont family means ordered for forking means with standard error bars. The higher forking tendency of Coastal families is apparent in the plot.

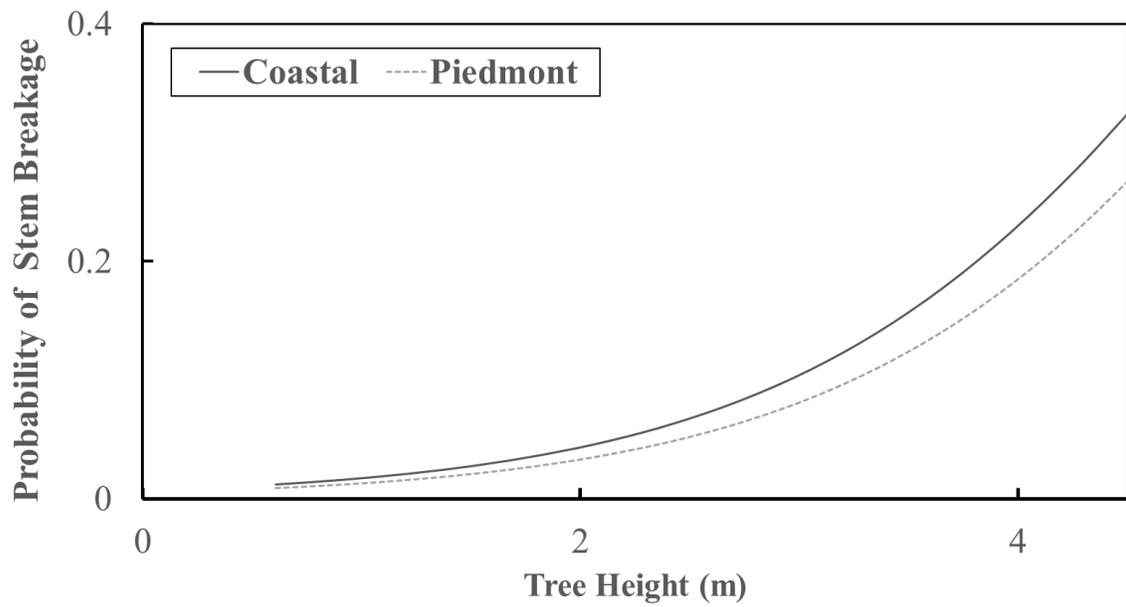


Figure 6. Probability of main stem breakage (y-axis) as a function of tree height (x-axis) for Piedmont and Coastal provenances. The probability of stem breakage increases as the trees get taller, and the difference between seed sources increases with the increased height.

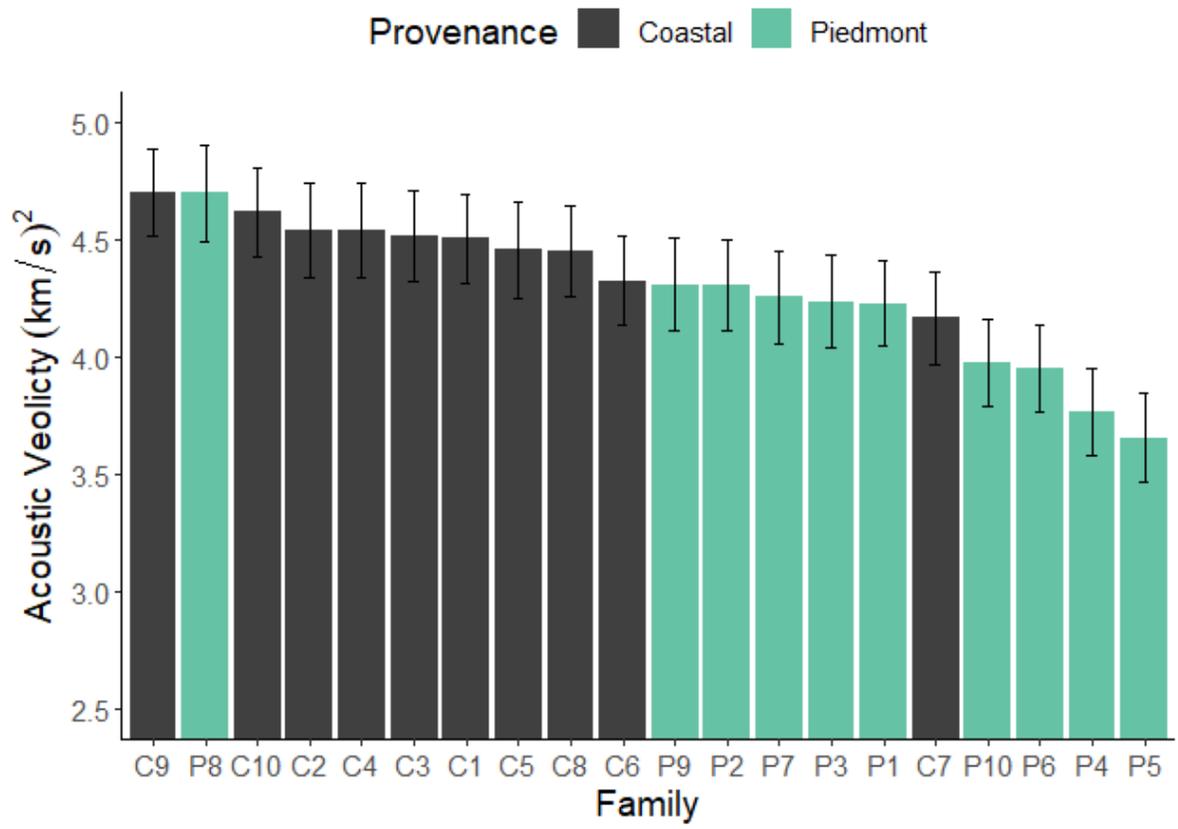


Figure 7. Acoustic velocities mean for families from two provenances. Coastal families had less variation and significantly ($Pr = 0.0017$) higher velocities than did Piedmont families.

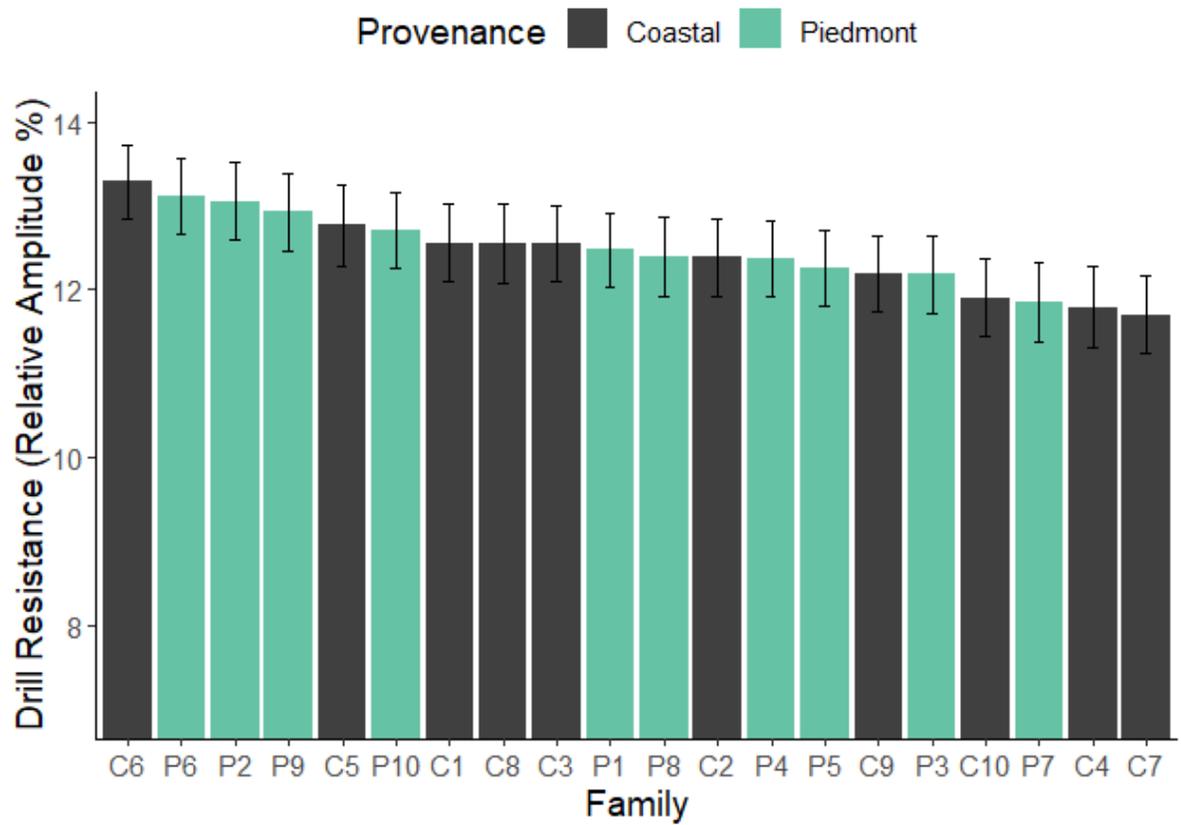


Figure 8. Relative amplitude family means for Coastal and Piedmont seed sources. There was considerable and significant variation among families, but differences between seed sources were not significant.

Chapter 3. Provenance variation in biomass potential of loblolly pine (*Pinus taeda*) families in the Piedmont of North Carolina

Abstract

Considerable genetic differences within loblolly pine (*Pinus taeda* L.) exist for growth, stem form, and wood quality traits that influence biomass/biofuel production. By planting genetically superior trees with desirable biomass/biofuel traits, it is possible to dramatically increase the amount of biomass and as well as sawtimber produced from plantations.

Ten of the fastest growing loblolly pine families from two provenances, Atlantic Coastal Plain and Piedmont, were tested for their biomass potential in North Carolina on a Piedmont site. On average, Coastal families produced 3.3 m³/ha more wood than Piedmont families. However, when examining the sawtimber potential of the trees, Piedmont families produced 3.7 more m³/ha of sawtimber potential wood than Coastal families. Differences in volume were also significant at the family level (Pr = 0.023). For biomass plantations, risks can be minimized due to shorter rotation length, allowing for a higher-risk genotype to capture the greater gains in terms of volume. For a sawtimber stand, more conservative family deployment should be considered to ensure good stem quality at the end of the rotation. Understanding the trade-offs with different genotypes and harvest regimes are essential to ensure profitable returns from the pine plantations.

Key word(s): loblolly pine, biomass, bioenergy, wood quality

3.1. Introduction

As the rate of population growth accelerates in the world, the demand for energy increases as well. The population at the beginning of the Industrial revolution was 700 million; today the population is approximately 7 billion (Chu and Majumdar 2012). The world population is expected to exceed eight billion by year 2030 (United Nations 2015). In the Southeastern United States alone, the energy demand is expected to increase at a rate of 1.5% annually until 2030 (EIA 2006). Currently, most energy demands are being met with the use of fossil fuels such as oil, coal and gas. The harvest and use of these fuels are having detrimental effects on the planet. The Intergovernmental Panel on Global Change (IPCC 2001, 2007) has confirmed that anthropogenic factors are directly correlated with increase greenhouse gases in the atmosphere (Dincer 2000). These issues, in combinations with the 1970s energy crisis has led to a call for a transition to alternative forms of energy, particularly renewable resources (Kantavichai et al. 2014).

Renewable energy sources occur naturally and replenish their reserves over time. These energy sources include solar, wind, biomass, geothermal, ocean currents, and hydropower energy (National Renewable Energy Laboratory 2008). A major disadvantage is the reliability of some types of renewable energy, such as solar and wind power. These energy sources rely on the weather to generate power (Goldemberg 2000). Additionally, the power generation cost from these harnessed energy sources is high. Other sources of renewable energy need to be explored (Panwar et al. 2011).

Another form of renewable energy comes from biomass, a carbon-neutral energy source that includes agricultural crops and trees, wood and wood residuals, grasses, and other plant-derived matter (Galik et al. 2009 and Stöcker 2008). The harvesting of this biomass can produce electricity, liquid biofuel, and residential heating (Galik et al. 2009). Currently, the most used biomass crop is corn (Scott and Tiarks 2008). This first-generation bioethanol production system utilizes plants rich in carbohydrates such as sugars and starches (Stöcker 2008). However, corn and other bioenergy feedstocks are primary food-crops that are vital to world food security, leading to increasing debate of whether these high-input and highly-valued food crops should be used as an energy source (Hill et al. 2006). The standard for any biofuel to be considered a viable and sustainable substitute, it must be economical to produce in large quantities without jeopardizing food supplies, provide more energy than is used to produce the crop, and provide some environmental benefit such as reduction of greenhouse gas emissions (Johnson et al. 2007).

Trees as a potential biomass source have been studied extensively. The studies that focus on woody perennials as energy crops includes softwoods and hardwoods, also known as second-generation lignocellulose biomass crops (Johnson et al. 2007 and Stöcker 2008). Wood-based biomass is widely available in large quantities in most parts of the world. Hardwoods grown in short rotation typically include poplars and aspens (*Populus* spp.), *Eucalyptus* sp., sycamore (*Platanus* sp.), maple (*Acer* spp.), willow (*Salix* spp.), and sweetgum (*Liquidambar* sp.), among others (Johnson et al. 2007). *Populus* sp. may be an option for biomass production in the Southeast, but have high establishment costs (Tuskan 1998), require large amounts of water input (Merkle and Cunningham 2011), are often

planted with clones that may be susceptible to insect disease on given sites (Tuskan 1998), and regeneration from coppicing may result in reduced productivity (Perela and Alban 1982 and Cobb et al. 2008).

Another option for a biomass crop in the Southeast may be softwoods, particularly loblolly pine (*Pinus taeda* L.), given its abundance in the area. Loblolly pine is a heavily researched and economically valuable species in the Southeast. Loblolly pine produces 212 million m³ of wood each year resulting in 4.3 billion US dollars of stumpage value (Li et al. 1999). After only two breeding cycles, tree improvement programs have made significant gains in volume, height, straightness, and reduced fusiform rust incidence (caused by the fungus *Cronartium quercuum* f. sp. *fusiforme*) over the wild native stock (Li et al. 1999 and Johnson et al. 2007). Through selective breeding, gains as high as 12 percent more volume per hectare and 32 percent in harvest values were seen over the wild stock in the first generation alone (Enebak 2017 and Li et al. 1999).

Studies examining the genetic variation within loblolly pine for timber rotations are numerous (McKeand et al. 2006, Farjat et al. 2017, Li et al. 1999 and Talbert et al. 1985). Provenances, or regions, of loblolly pine show variation in growth rates (Lambeth et al. 2005), susceptibility to rust (Schmidting 2001 and Li et al. 1999), wood specific gravity (Isik and Li 2003), and cold tolerance (Lantz and Kraus 1987). Tree improvement programs have identified families within provenances that exhibit superior production and value for sawtimber production (McKeand et al. 2006).

There are potential benefits from planting non-local seed sources at a given location. For example, moving a southern source such as families from the Atlantic Coastal Plain into

the Piedmont region, can result in faster growth and greater productivity (Schmidting 2001). This movement does involve some risk from cold/ice damage, but this may be less important for short-rotation biomass production than for sawtimber rotations, as bole form is less important (Lambeth et al. 2005) and shorter rotations decreases the risks from periodic extreme weather events. Few studies have focused on the variation among families and provenances for biomass production.

Assessing both biomass and sawtimber potential is critical for forestry professionals and landowners to understand harvest options and to make sound management decisions. Given what we know about provenance variation, we expect the Coastal families to grow faster, but being planted outside of their range may put them at a disadvantage on a northern or interior site (Schmidting et al. 2001). Although, Coastal families may have a higher probability of damage given a major storm event, this may not be a significant concern for a biomass rotation.

3.1.1. Objectives

The objective of this study is to evaluate 20 loblolly families from the Atlantic Coastal Plain and Piedmont provenances for their biomass production and sawtimber potential on a Piedmont site. This study will allow for assessment of the risk/reward tradeoffs of planting a faster-growing, but less adapted provenance on a high-risk site. By taking height, and diameter at breast height (DBH), volume estimates, and relative wood density measurements we will be able to compare biomass productivity among provenances and families.

3.2. Methods and Materials

3.2.1. Experimental Design and Data Collection

For this experiment, 20 wind-pollinated families were planted at a NC Department of Agriculture field site in Butner, NC (Figure 1). Because biomass yield is an important trait for biofuel, we selected 10 of the fastest growing families adapted to the Piedmont region and 10 of the fastest growing families adapted to the Coastal Plain region. The average minimum winter temperature from where the Coastal families originated is -8.8°C , and the average minimum winter temperature for the Piedmont families is -11.3°C . By incorporating the higher-risk Coastal families, we will be able to evaluate the risk/reward balance of planting faster-growing material that is not as well-adapted as the Piedmont families.

The experiment was planted in the spring of 2012. The experimental design was randomized complete blocks with split plots. A block was split into four main plots and two seed sources Piedmont and Coastal were randomly assigned to the main plots. Two of the plots were to be for biomass and two for sawtimber thinning/harvest management. Ten families of each provenance were randomly assigned to 36-tree square sub-plots within each plot. There were five blocks in the study, but since the thinning and harvesting regimes had not been implemented, there were effectively 10 experimental blocks in the study. A planting density of 1.829 m x 2.133 m (2563 trees per hectare) was used at the site (Figure 2). Number of seedlings planted at the beginning was 7,200, including buffer and border trees a total of 12,408 trees were planted in the study.

Herbicide was applied to the field to control herbaceous woody competition. The chemical imazapyr was applied to the experimental blocks at a rate of 2 ounces per acre to

control for unwanted vegetation. A 0.02% solution of bifenthrin, an insecticide, was used to prevent damage from Tip moth, sawflies, and other pests that could compromise the trees. Given that the study site was formerly an agriculture field, there were issues related to soil nutrition. Site specific applications of 2% chelated iron mix solutions and ammonium sulfate were applied to targeted reps to improve soil fertility.

3.2.2. Data Collection

The site was measured at age six. Standard measurements included height (m), diameter at breast height (DBH) (cm.), survival (alive or dead), presence of fusiform rust (yes or no), and forking (yes or no). Tree height was recorded using a vertex hypsometer. Diameter at breast height was recorded at 1.4 meters from the base of the tree using a diameter tape.

Incidence of fusiform rust was recorded with 1 presence of galls and 0 indicating no gall. Presence of stem forking was indicated with a 1, no fork was given 0.

Resistograph, a tool used as a surrogate for wood density was taken on a subset of the total 7,200 trees in the study (Isik and Li, 2003). Of the 36-family block plots, the inner 9 trees were assessed for wood density. In the subset, the sample size per family was 90 sample trees, the total subset was 1800 trees.

3.2.3. Biomass Potential

Traditional biomass yields are estimated through harvesting trees and oven drying for dry weight volumes, but these methods are labor intensive and destructive to the site. Standing biomass was calculated from measurements of bole volume and woody density.

Sherrill et al. (2011) developed outside-bark bole volume equation for improved loblolly pines through age 9, which was used to calculate bole volumes at both individual tree and plot levels.

$$\hat{V}_{tob} = 0.020571 + 0.00237(D^2H) \quad Eq. 1$$

Where \hat{V}_{tob} is total outside-bark stem volume (ft³), followed by coefficients, D is diameter at breast height (4.5ft above ground level) in inches, H is total stem height in feet. Since, Sherrill (2011) volume estimates are given in English units, for this study volume estimate per tree was converted to cubic meters (1 ft³ = 0.0283168 m³). Volume estimates per hectare for each plot were calculated by summing the total volume per tree in each plot (36-tree family plots) and dividing by plot size (0.0140469 hectare/plot).

To obtain relative measures of wood density, the Resistograph (drill resistance) was used (Isik and Li 2003). Resistograph uses a thin needle (1.5 to 3.0 mm) to drill into a live tree with a constant force, measuring the drill resistance from bark to bark (Isik and Li 2003). A subset of the total study trees was assessed for wood density due to time constraints. For each 36-tree family plots, the inner 9 trees were sampled for wood density and an average wood density per plot was calculated. Resistographs measurements have shown high genetic correlations with the wood specific gravity and dry weight volumes (Isik and Li 2003 and Eckard et al. 2010). To convert Resistograph values to specific gravity, an equation developed by Walker et al. (2019) was used.

$$SG = SG_{sp} + 0.005508R \quad Eq. 2$$

Where SG is specific gravity, SG_{sp} is specific gravity of species, for this study we used a general specific gravity of 0.41 (Jett et al. 1991), followed by a coefficient multiplied by Resistograph measurements, R .

Next, we converted the specific gravity estimates to wood density in kg/m^3 by multiplying each value by 1000. Wood density was then multiplied by volume per hectare estimates for each plot to give in dry weight index for each plot of the stand. We converted dry weight index from kg/ha to a more recognized metric of Mg/ha . We consider this as an index of dry weight yield since we only had wood density estimates at breast height and not throughout the tree. Previous work has shown that breast height estimates for wood density are highly correlated with whole tree estimates (e.g. Aspinwall et al. 2010 and Zobel et al. 1960), hence, the use of the index.

3.2.4. Sawtimber Potential

Sawtimber potential score was assessed to help identify families with good biomass potential as well as sawtimber potential. Sawtimber potential was measured using a categorical scale suggested by Cumbie et al. (2012). Sawtimber potential was on a 4-point scale, 1 being the best to 4 being the worst, where

1 = Sawtimber: no quality defects in the stem in the first 1.5 logs (7m) of the tree.

2 = Some sawtimber: minor defects in the first 1.5 logs but still likely to be a sawtimber tree. Minor defects may include small or few ramicorn branches, high fork (above first log), or minor sweep.

3 = Pulpwood: defects present, such as low forks, large or multiple ramicorn branches, or large sweep.

4 = Pulpwood or non-merchantable: major stem defects such as stem rust, multiple forks, or extremely poor growth and stem characteristics.

For analysis, sawtimber potential scores 1 and 2 were combined into a sawtimber potential category and score 3 and 4 were combined into a biomass only or pulpwood category, creating a categorical variable.

3.2.5. Statistical Analysis

All response variables were examined through distribution analysis. Descriptive statistics were produced, and least square means of each response variable for family and provenance were examined. The following linear mixed model was fit to understand the differences between seed sources for traits with Gaussian distributions: tree height (m) and diameter at breast height (cm) and volume per tree (dm^3/tree). The plot volume (m^3/ha), plot dry weight indices (Mg/ha), and plot drill resistance (% relative amplitude) were analyzed using a reduced model.

$$Y_{ijkl} = \mu + B_i + P_j + F_k + BP_{ij} + BF_{ik} + \varepsilon_{ijkl} \quad \text{Eq. 3}$$

Where Y_{ijkl} is l -th measurement of family k , seed source j in block i . μ is the intercept, B_i is the i -th block effect, P_j is the j -th provenance effect, F_k is the k -th family effect, and BP_{ij} and BF_{ik} are the interaction between block and provenance, and block and family, respectively. ε_{ijkl} is the residual error associated with individual-tree observations. The

model assumes that the residuals are normally distributed, independent of one another, with constant variances, $e \sim N(0, \sigma^2)$. In the model, the block, block by provenance, and block by family interactions were considered random with expectations of overall zero mean and identical and independent variances. The model was run using the MIXED procedure of SAS software (SAS Institute Inc. 2014). The model did not converge when block by provenance interaction term was included in the model, because it did not explain any variance (zero variances). These terms were dropped from the final model. In order to test the significance of variance component explained by family effect, log likelihood ration tests were carried out by comparing Log Likelihood of a model with and without family effect (Isik. et al. 2017).

Stem forking and presence of rust galls were assumed to have binomial distribution. For the categorical traits such as forking and incidence of rust galls, a generalized linear mixed model was used to test differences between seed sources and families.

$$\log[\pi/(1 - \pi)] = \mu + B_i + P_j + F_k + BP_{ij} + BF_{ik} + \varepsilon_{ijkl} \quad \text{Eq. 4}$$

Where the probability of stem forking or occurrence of rust is modeled using the logits $\log[\pi/(1 - \pi)]$ where π is the probability of outcome, μ is the intercept or reference level, B_i is the i-th block effect, P_j is the j-th provenance effect, F_k is the k-th family effect, and BP_{ij} and BF_{ik} are the interaction between block and provenance and block and family, respectively. ε_{ijkl} is the residual error associated with individual observations. In the model, the block effect was considered random, block by provenance and block by family interactions were also considered random. Residuals of response variables were normally distributed and independent of one another with constant variances. The models were run

using the GLIMMIX and LOGISTICS procedures of SAS software (SAS Institute Inc 2014)
and logit link function.

3.3. Results

After the third growing season, the site experienced two severe winter storms with accumulation of 1.3 to 2.5 cm of ice on the site for several days. Ice storm damage was assessed across the site. Coastal provenance experienced higher frequency of damage in the form of main stem breakage and crown damage than did the Piedmont provenance (Chapter 2).

3.3.1. Biomass Potential

Provenances differed significantly for tree height ($Pr = 0.018$), but not for DBH, stem volume, plot volume, dry weight index, and drill resistance (Tables 1 and Table 2). For the random family effect, variance components for DBH, height, drill resistance, stem volume, stand volume, dry weight index and acoustic velocity are presented in Tables 2. Family effect explained significant variation for all the traits based on log likelihood ratio tests.

The Coastal seed source was 23 centimeters taller than Piedmont seed source, with a population mean of 7.36 meters and Piedmont 7.19 meters (Table 3). Diameter at breast height for the Coastal seed source was 11.24 cm and was not significantly different from the Piedmont seed source mean of 11.19 cm ($Pr = 0.6898$). There was a significant family effect for DBH ($Pr < 0.001$).

Resistograph measurements, recorded as drill resistance and used as a surrogate for wood density, had a large amount of family variation (Table 2, Figure 2) that was significant ($Pr = 0.0004$), but there were no significant differences between seed sources ($Pr = 0.4188$).

Variance components for the family effect for stem forking was 0.07556 (SE = 0.0295) and for fusiform rust incidence was 0.1558 (SE = 0.0571); given the large ratio of estimate to standard errors (>1.5), family effect explains considerable variation for stem forking and fusiform rust.

Dry weight index was calculated by multiplying the volume per hectare estimates by average wood density at the plot level. The dry weight index for the Coastal seed source was 46.6 Mg/ha (SE = 1.94), which was not statistically (Pr = 0.222) different from the Piedmont seed source average of 44.9 Mg/ha (SE = 1.94). There were significant differences between families (Pr < 0.0001) (Figure 3).

There were significant differences (Pr = 0.0189) between seed sources for stem forking incidence, but not for incidence of fusiform rust galls (Pr = 0.2287) (Table 4). Coastal seed source had an average of 63% forked trees in the study and Piedmont seed source had 55% forked trees. The odds ratio for the probability of forking was 1.36, indicating that Coastal families had 36% more likely to have a fork than Piedmont families. For fusiform rust incidence, Piedmont (0.34) and Coastal (0.30) seed source had similar probability values (Table 4).

3.3.2. Sawtimber Potential

The Coastal provenance produced 3.82 more cubic meters of total volume per hectare than the Piedmont provenance, but this difference was not significant at the provenance level (Table 1). There were significant differences at the family level (Pr < 0.0001) (Table 2). The same trend was seen for individual tree volumes, with the Coastal seed source producing

more volume per tree than the Piedmont seed source; the differences were not significant at the provenance level, but were significant at the family level (Table 1 and 2).

Volume was also affected by the ice storm at age three. Coastal families experienced more volume damage in the form of broken tops than most Piedmont families. Even with the damaged volume, some Coastal families produced more non-damaged volume than Piedmont families (Figure 4).

Although some Coastal families still produced more volume at the stand level than some Piedmont families, when classified at age six for sawtimber potential and biomass potential, Coastal families had more biomass potential trees than Piedmont families (Figure 5). Sawtimber potential was significantly different between seed source ($P < 0.0001$). Piedmont seed source had 55% of the trees in the study classified as sawtimber quality potential, while Coastal seed source had 50%. For biomass potential trees, Coastal seed source had roughly 5% more trees classified as biomass potential than the Piedmont seed source average of only 45%.

When examining the amount of volume in sawtimber potential trees compared to biomass potential trees, the Piedmont seed source had a greater proportion of volume in sawtimber potential trees than did the Coastal seed source (Figure 6). Piedmont had 57.1 cubic meters per hectare of sawtimber potential volume and only 36.4 cubic meters per hectare of biomass quality volume. Coastal families had significantly less sawtimber potential volume with 53.4 cubic meters per hectare and 44.1 cubic meters per hectare of biomass potential volume.

3.4. Discussion

3.4.1. Biomass Potential

Diameter at breast height did not differ between the Coastal and Piedmont provenances, but did differ among families. Height was significant by family and seed source. Coastal families were taller than most Piedmont families by about 0.25 m. Coastal seed source having a faster growth rate is well documented (Schmidting, 2001), but there are risks of cold damage and damage from snow or ice. Given the short rotation of a biomass regime, the probability of occurrence would be greatly reduced compared to a much longer sawtimber rotation (e.g. 25 to 40 years).

Wood density was estimated using the Resistograph as a surrogate measurement. The Resistograph measures drill resistance in units of percent relative amplitude. A greater resistance implies denser wood. For biomass, a high-density tree is ideal over trees with high water content and lower density. There were no significant differences between seed sources for wood density, but there was a significant family effect on wood density. Even with ice damage, Coastal families out grew Piedmont families and some even had higher density. Our expectation was that Piedmont families would have higher wood density than the Coastal Source as suggested by others (Belonger et al. 1996 and Jayawckrama et al. 1997), but that was not the case. We believe there were no differences in wood density between seed sources because the families selected were not a representative sample from each seed source. The families from each provenance in this study were selected for being the “best of the best” due to their high growth rate and volume production.

For dry weight indexes, similar to Resistograph measurements, least square means were not significantly different between seed sources, but there were significant differences among families. The top three families with the largest dry weight indexes were from the Coastal provenance. These results suggest that for a biomass rotation, selecting a seed source or genotype not typically recommended due to associated risk, may offer enough added benefit that it may be considered for this type of harvest regime.

3.4.2. Sawtimber Potential

Total volume production at age six was not significantly different between seed sources on individual tree level and plot level. This may be due to the ice storm at age three causing Coastal families to take longer to recover, since they experienced a higher occurrence of damage than most Piedmont families (Chapter 2). There was a plot level effect on volume production, which may be due to the differences in damage across the site (e.g. storm damage is rarely uniform across a landscape).

Sawtimber potential score was used to predict the sawtimber quality of trees at the end of rotation (e.g. Cumbie et al. 2012). Piedmont families had more trees classified as sawtimber quality than Coastal families. This is likely the result of decreased stem form quality in Coastal trees than in Piedmont trees due to the damage from the ice storm. Coastal families had higher frequency of breakage, affecting the overall stem quality (Chapter 2).

There were also differences in sawtimber volume with Piedmont trees having roughly 4 cubic meter per hectare more in sawtimber compared to trees in the Coastal seed source. This difference in volume may be attributed to ice storm damage, but may also be due to forking. On average, the Coastal seed source had 8% higher forking across the study than the

Piedmont source. Forked trees usually resulted in a sawtimber quality score of 3, classifying the tree as only biomass quality. This larger difference in forking may have caused the greater difference in sawtimber quality between the two seed sources.

3.5. Summary

This study examined the effect of family genotypes and seed source on the effect of biomass and sawtimber production. The overall productivity of the site was high with some significant differences among families and between provenances in relations to phenotypic traits. The results highlight the importance of proper genotype selections based on site location and management objectives.

The importance of genetics and management selection cannot be overstated. This study has highlighted the associated risk and benefits of using a nonlocal seed source for two harvest regimes: a sawtimber plantation and a biomass plantation. For biomass plantations, risks can be minimized due to shorter rotation length, allowing for a higher-risk genotype to capture the greater gains in volume. For a sawtimber stand, more conservative genotype selection should be used to ensure good stem quality at the end of rotation. Understanding the trade-offs with different genotypes and harvest regimes are essential to ensure profitable returns from the plantation.

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Zobel, B.J., F. Henson, and C. Webb. 1960. Estimation of certain wood properties of loblolly and slash pine trees from breast-height sampling. *For. Sci.* 6(2):155–162.

3.7. Tables

Table 1. F tests for provenance effect. There was a significant provenance effect for height, but not for the other traits measured.

Trait	Den. DF	F Value	Pr > F
DBH (cm)	6759	0.16	0.6898
Height (m)	6760	5.60	0.0180
Volume (dm ³ /tree)	6757	1.45	0.2279
Volume (m ³ /ha)	100	1.69	0.1966
Dry Weight Index (Mg/ha)	100	1.51	0.2220
Drill Resistance (amplitude)	100	0.59	0.4460

Table 2. Variance components for family effects for biomass related traits. The estimates were significantly different from zero based on log likelihood ratio tests.

Trait	Estimate	Standard Error	Estimate/SE	Pr > Z
DBH (cm.)	0.0089	0.00431	2.06	<.0001
Height (m)	0.0163	0.00876	1.86	<.0001
Volume (dm ³ /tree)	3.4938	1.7508	2.00	<.0001
Volume (m ³ /ha)	29.6084	44.6994	2.04	<.0001
Drill resistance (amplitude)	0.1315	0.0689	1.91	0.0004
Dry Weight Index (Mg/ha)	7.1038	3.4836	2.04	<.0001

Table 3. Provenance means and the standard error of the means for traits with Gaussian distributions. Trees from the Coastal seed source on average were significantly taller than the Piedmont seed source.

Trait	Costal source Mean (SE)	Piedmont source Mean (SE)
DBH (cm)	11.24 (0.149)	11.19 (0.149)
Height (m)	7.36 (0.150)	7.19 (0.150)
Volume (dm/tree)	39.79 (1.38)	38.51 (1.38)
Volume (m ³ /ha)	97.48 (4.04)	93.66 (4.04)
Drill resistance (amplitude)	12.37 (0.171)	12.53 (0.171)
Dry Weight Index (Mg/ha)	46.64 (1.94)	44.87 (1.94)

Table 4. F tests of provenance effect for forking and fusiform rust disease. Coastal had a significantly higher proportion of forked trees than Piedmont. There was no provenance effect on the occurrence of fusiform rust.

Trait	Provenance	F	Pr > F	Mean (Frequency)	SE
Rust (%)	Piedmont	1.45	0.2287	0.34	0.029
	Coastal			0.30	0.027
Forking (%)	Piedmont	5.51	0.0189	0.63	0.0219
	Coastal			0.56	0.0230

3.8. Figures

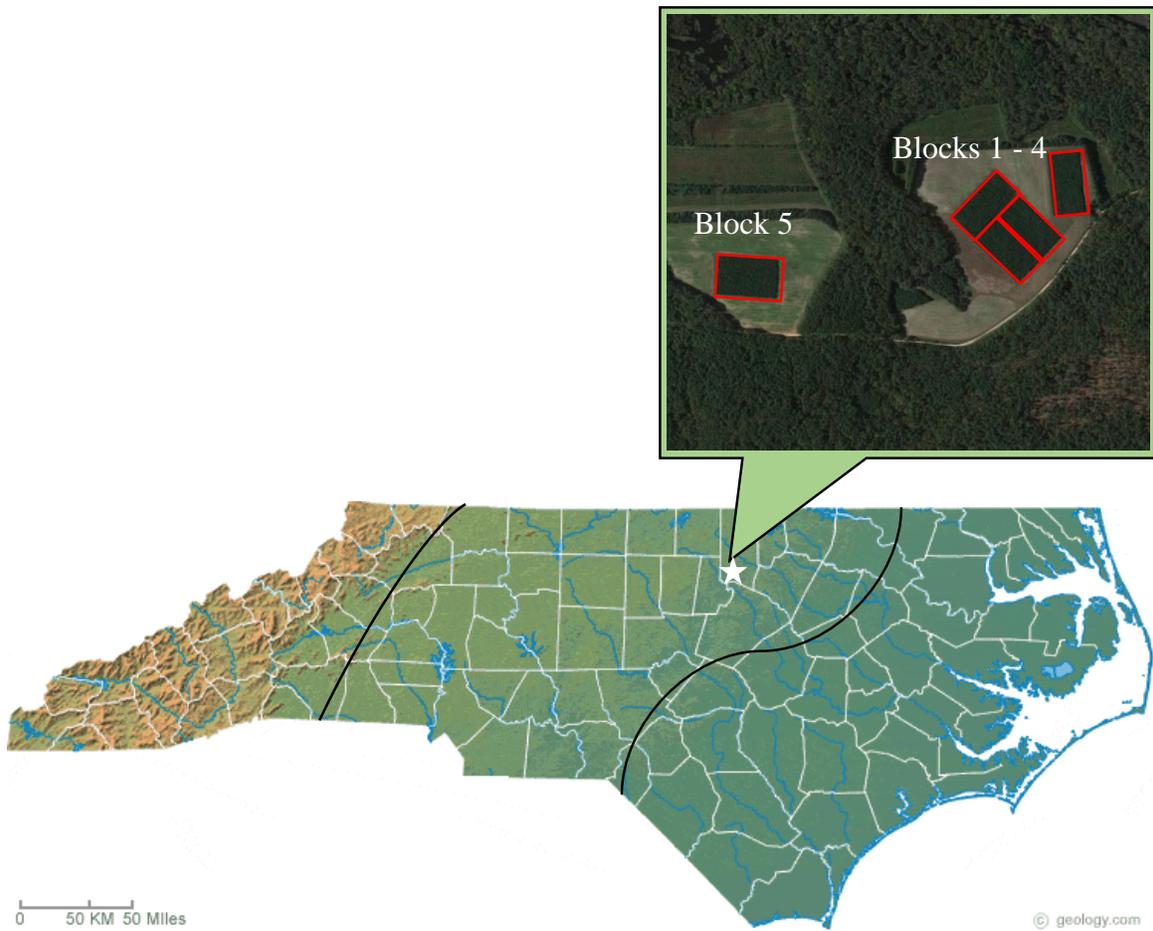


Figure 1. Location of Butner field trial in Granville County, in the Piedmont of North Carolina. The test site is located on the NC Department of Agriculture & Consumer Services Umstead Farm. The lines designate the division of the zones (left to right): Mountain region, Piedmont region, and Coastal region.

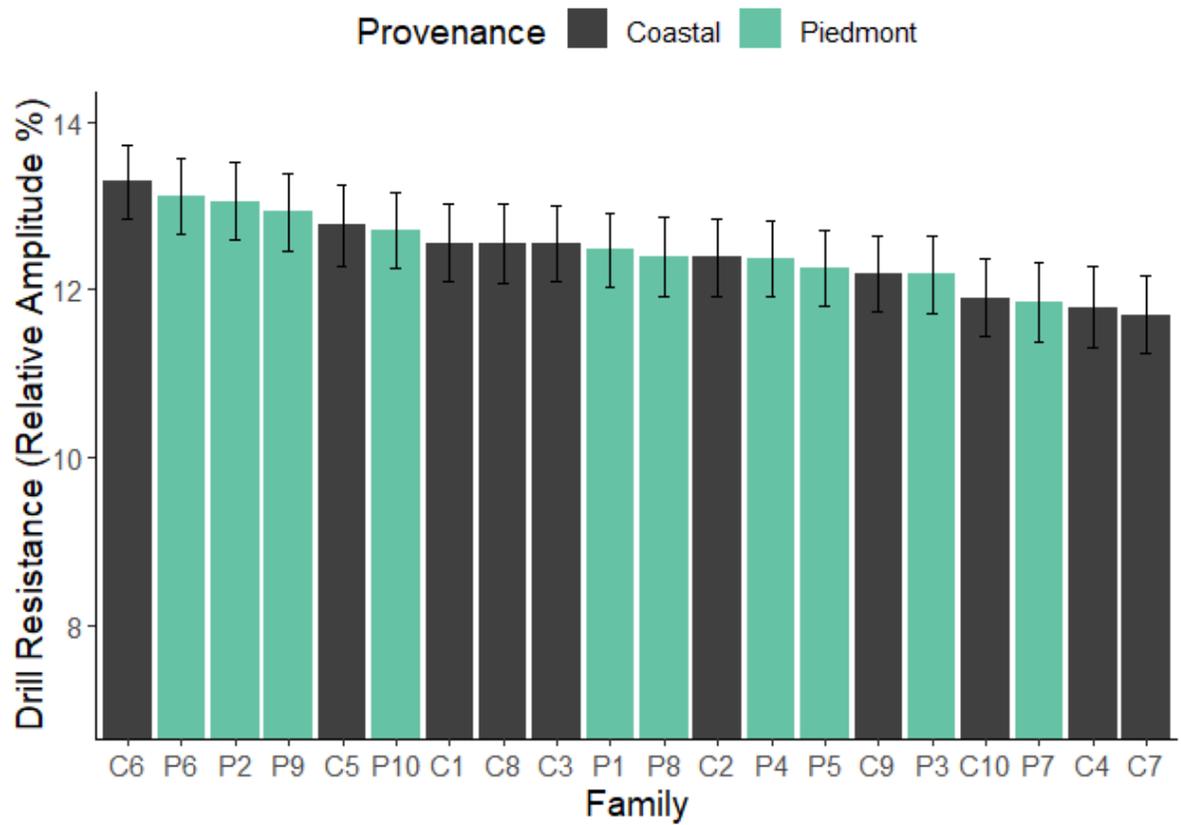


Figure 2. Family means and their 95% confidence intervals for drill resistance. There was a significant family effect on drill resistance, but no significant seed source effect. A greater resistance implies denser wood.

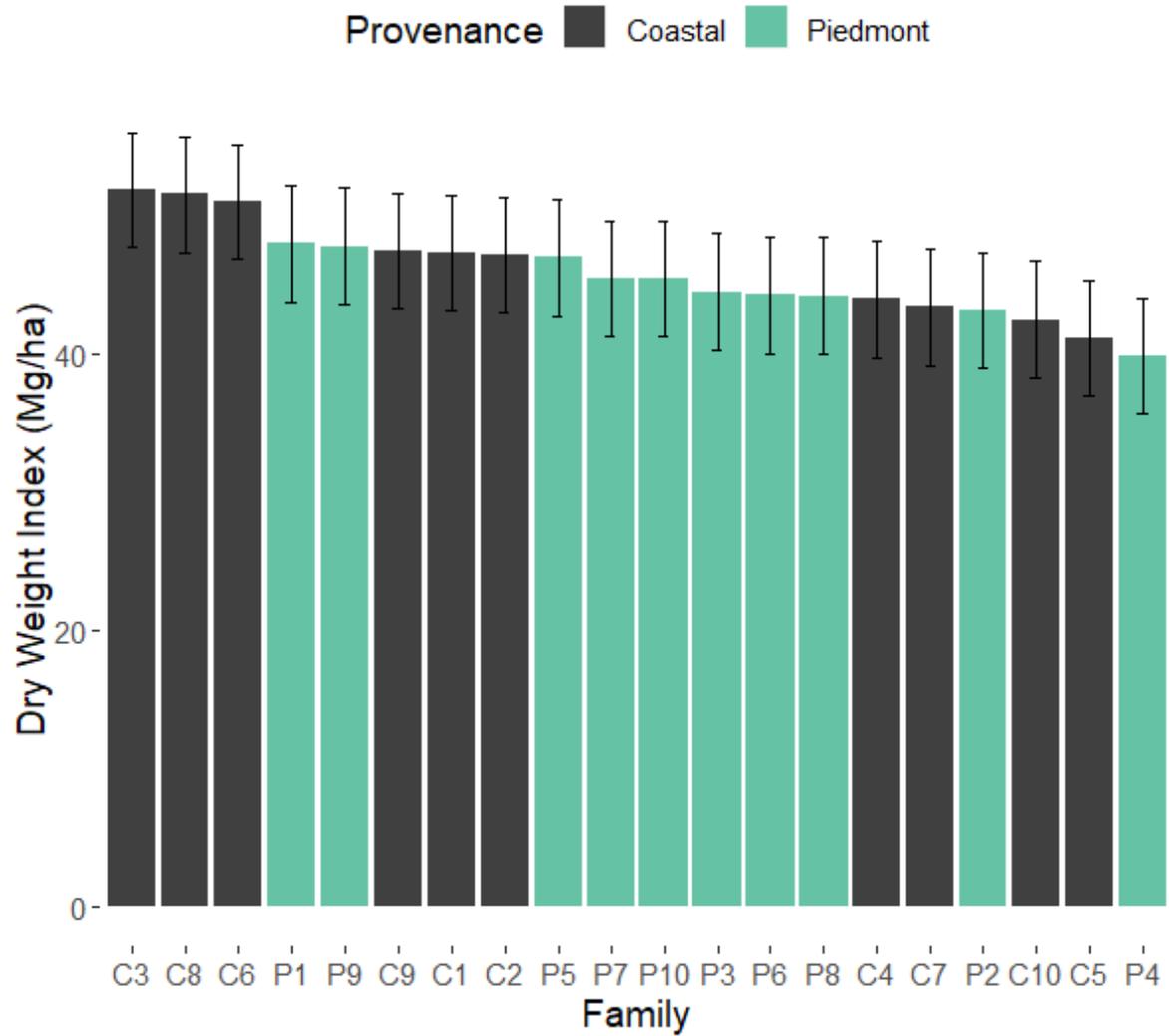


Figure 3. Family means and their 95% confidence intervals for dry weight index (Mg/ha).

There was a significant family effect on dry weight per hectare, but no significant seed source effect.

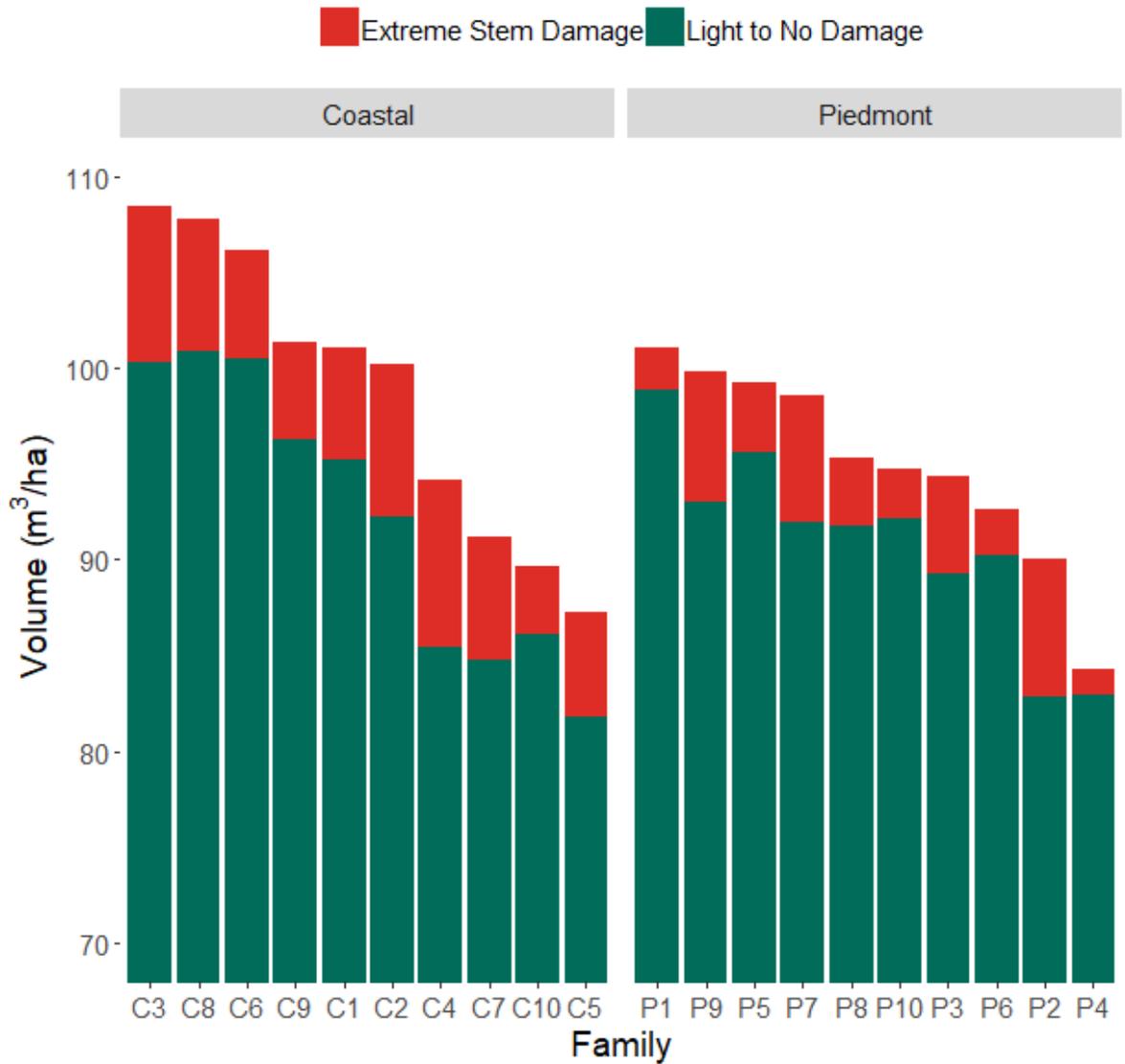


Figure 4. Total volume by damage type, light to no damage or extreme stem damage. For total mean stem volume, Coastal families had greater damage than most Piedmont families. Additionally, some Coastal families also had greater light to no damage than some Piedmont families.

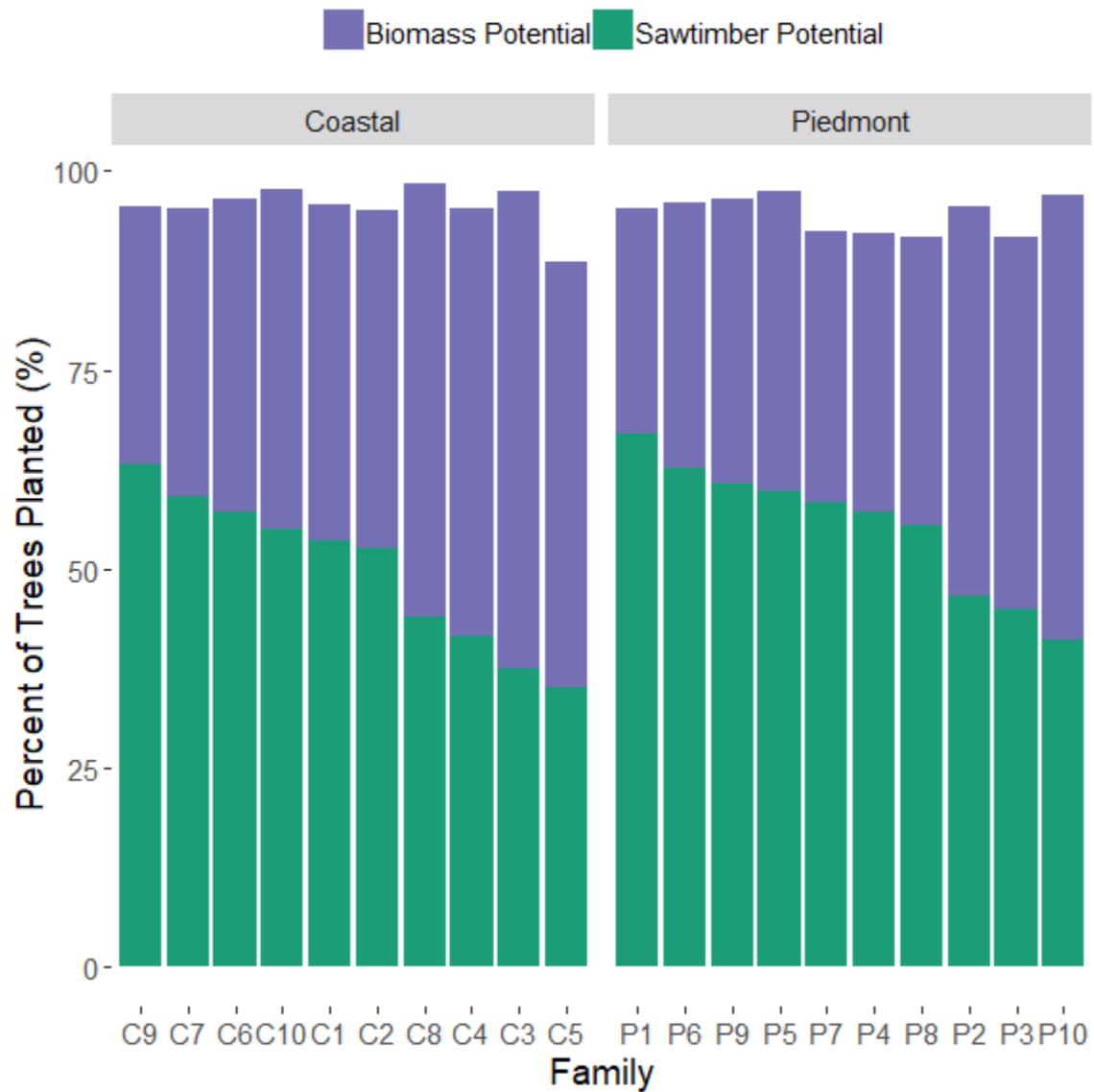


Figure 5. Proportion of trees in each family classified as either sawtimber potential (green) or biomass/pulp potential (purple). On average, Piedmont families had a greater proportion of trees classified as sawtimber potential than did Coastal families.

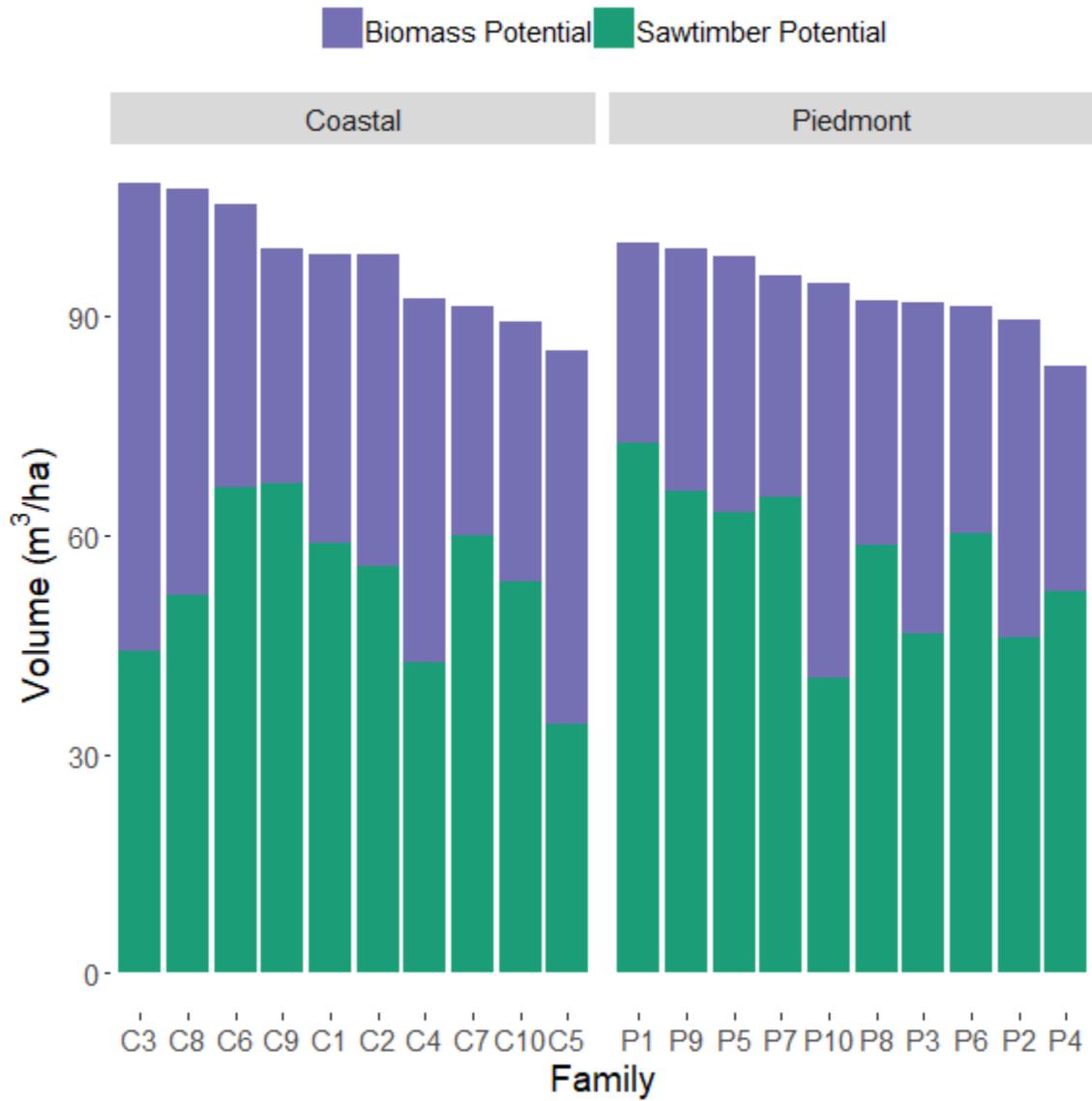


Figure 6. Proportion of volume that is in sawtimber potential trees and biomass trees. Piedmont families had more volume in sawtimber potential trees (55%) than in biomass trees. On average, the Coastal seed source had 50% of the mean volume in sawtimber potential and biomass potential.

Chapter 4. Predicting individual tree heights in loblolly pine biomass plantation using drone imagery and LiDAR-derived measurements

Abstract

Height estimation from Unmanned Aerial Systems (UAS) is an ever-increasing interest in natural resource fields. With the use of PhoDAR from drone imagery in combination with LiDAR data, we produced high-resolution digital surface models over two test sites. After individual tree identification, we computed individual tree heights on a young biomass plantation of loblolly pine. PhoDAR derived heights were compared with field measurements. Of the 7,200 study trees, the software was able to detect 7,143 trees. Overall correlations between field measured heights and PhoDAR derived heights among sites was low (0.18). Site 1 had a very low correlation (-0.10), but Site 2 had a strong correlation (0.79). The difference in correlation was attributed the “bowl effect” commonly noted in some digital elevation models from vertical UAS imagery (James and Robson, 2014). With proper ground control and techniques to mitigate such board-scale deformations, the use of PhoDAR derived measurement can become an efficient method and might replace traditional field methods.

Keywords: loblolly pine, drone imagery, othromosaic, liDAR

4.1. Introduction

Forest management relies on estimation of a forest attribute at stand level such as composition, density, and overall forest health and individual tree level such as tree height, diameter at breast height (DBH), and other phenotypic characteristics (Chang 2008). Current field-based methods of obtaining forest inventories are expensive, labor intensive, and inefficient (Hyypä 2001). To obtain an individual tree characteristic such as height from remote sensing data requires individual tree segmentation (Seul et al. 2015).

Methods for individual tree extraction can be obtained from aerial photography, airborne LiDAR (Light Detection and Ranging) or combined data from both (Chang et al. 2012). Aerial photography, however, is limited to only two-dimensional (2-D) data that often lack the points to generate vertical structures of forest canopies (Hall et al. 2005). Additionally, with only surface images, no vertical insight, ground models are difficult to impossible to construct for height calculation (Seul et al. 2015). LiDAR offers insight into the vertical structure of forests with laser pulse technology mapping the forest layers (Díaz-Varela et al. 2015). This method is widely accepted but has a variety of shortcomings. Most of which include its relative high costs and difficulties in the retrieval of high density returns required for accurate screening or phenotyping, particularly on small plots with high density trees (Díaz-Varela et al. 2015 and Zarco-Tejada et al. 2014). Underestimation of tree height is another disadvantage to using LiDAR for height extraction. This underestimation is often due to LiDAR striking the sides of crowns and missing the apex of the tree (Roberts et al. 2005).

This need for a faster method to measure large plantations has sparked a recent interest in natural resource fields exploring the use of various remote sensing devices, mounted on Unmanned Ariel Systems (UAS) to collect data (Howell 2017). Until recently, this technology was too expensive to use regularly, but with advancements, there has been a reduction in cost and size allowing for more accessibility for users (Howell 2017).

Photodetection and ranging (PhoDAR) is a combination of ‘photography’ and ‘LiDAR’ techniques that may offer an alternative to traditional aerial photography and LiDAR (Howell 2017). PhoDAR often generated from structure from motion (SfM) algorithms is able to generate a reliable 2-D and/or 3-D image with the collection of many multi-angle images with a high level of overlapping (Howell 2017 and Díaz-Valera et al. 2015). Software application such as Pix4D, Agisoft Photoscan, UAS Master, and Visual SFM stitch overlapped photographs by matching known objects to generate 3-D point clouds, surface models, terrain models, mosaics, and orthophotos (Howell 2017). This method based on photo reconstruction can create high resolution digital surface models and orthophograph mosaics with high spatial and temporal resolution (Fritz et al. 2013 and Gini et al. 2014). SfM point clouds are analogous to the first returns data from aerial LiDAR. This creates challenges when trying to extract tree heights with no ground elevation model. Using PhoDAR in combination with public LiDAR data may offer a solution to combat the disadvantages of both methods. Using public LiDAR data reduces costs of obtaining data and using PhoDAR allows for high resolution imagery of small dense study sites.

The study aims to develop, test, and analyze the use of color images by using a low-cost camera mounted on a remote-controlled drone and LiDAR data for the estimation of

heights from a high-density biomass plantation. We used SfM image reconstruction techniques to derive surface models and ortho-mosaics and combined with elevation models from public LiDAR data to estimate heights of six-year-old loblolly pine trees in a biomass study to test ability and accuracy of the combined methods. We tested the performance of the method against traditional field measurements.

4.1.1. Objectives

The objective of this study was to use drone imagery and LiDAR data to identify and extract tree heights from a loblolly pine (*Pinus taeda* L.) biomass plantation. This study focus is on developing a method for extraction and compare results to traditional field-based measurements. Additionally, this study includes a cost analysis on both methods to better inform future management decisions.

4.2. Method and Data Collection

4.2.1. Experimental Design and Study Site

For this experiment, 20 open-pollinated families of loblolly pine were planted at a NC Department of Agriculture field site in Butner, NC (Figure 1). Because biomass yield is an important trait for biofuel value, we selected 10 of the fastest growing families adapted to the Piedmont region and 10 of the fastest growing families from the Coastal Plain region.

The site was planted in the spring of 2012. Establishment of plantations with short-rotation, woody crops have been suggested at a density of 1200-1400 stems ha⁻¹, grown on a 6-10 year rotation (Johnson et al. 2007; Tuskan 1998). Our study design was a split block-plot with trees planted in 36-tree block plots of each family, at a density of 1.8m x 2.1m (2,645 trees per hectare) (Figure 1). The study consists of 10 replications containing 720 trees (20 families x 36 trees per family), replicated 10 times on ~20 acres totaling 7,200 trees in the study. See details in Chapters 2 and 3.

4.2.2. Drone Technology and Specs

The site is ideal for evaluating recent innovations in drone image processing given the even level topography and location of study sites. The test site was a biomass plantation composed of high density spacing (1.8 x 2.1 m or 2,645 trees per hectare) of loblolly pine. Ten reference points were used for ground control, six point at site A and four points at site B, using a Trimble R6 high accuracy survey GPS. The drone used was a DJI Matrice 100 Quadcopter with a DJI Zenmuse X5 camera capable of 16 megapixels with a 70-degree field of view. The drone was automatically controlled, flying on a grid pattern over the site, five

hectares. The flight height was 61m with a spatial resolution of approximately three centimeters per pixel. Pix4D was used to process the approximately 750 images to generate a high resolution orthomosaic and extract a high-density 3-D point cloud (Figure 3).

4.2.3. LiDAR Data

LiDAR data for the area was acquired from the North Carolina Spatial Data Download (<http://sdd.nc.gov/sdd/DataDownload.aspx>) by selecting the area of interest and downloading the appropriate tiles. The two tiles combined cover the extent of the study area. The data were downloaded as a standard .las file.

4.2.4. Height Estimation Process

In this study, we used the method described by Seul et al. (2015) to extract individual tree crowns for high resolution DSM and ortho-image mosaics. The overall experimental flow chart is illustrated in Fig. 2. Drone imagery was used to make RGB orthomosaic images and generate LiDAR like point clouds (PhoDAR) containing X, Y, and Z coordinates of the study areas. The LiDAR data from the NC SDD was used in combination with the PhoDAR to create a nDSM (new Digital Surface Model) from DSM (Digital Surface Model) of tree heights from PhoDAR. Segmentation, a method of partitioning raster images into several segments on the basis of pixel values and locations (Seul et al. 2015), was downloaded using the *lidR* (Roussel and Auty, 2018) package in R (R Core Team, 2017) to identify trees within the 3-D point cloud data. Pixels that are spatially associated are grouped into a group representation individual tree. From this segmentation method tree positions and heights can be extracted from the point cloud by using a “lasso” technique to find the tallest Z value in a

1.8 meter radius of the tree center. The experimental results were compared to field collected height, measured using a Vertex hypsometer, to assess accuracy.

4.3. Results

4.3.1. Individual Tree Identification

The drone imagery derived ortho-mosaics (Figure 2) clearly shows the study site and individual trees. The study sites were planted at a high density so there were some overlapping areas.

LiDAR imagery was used to construct a ground surface layer. **Figure 5** shows the 2014 LiDAR data for that study site corresponding to when the study site was only 3 years of age, with most trees visible from the 3d point cloud.

The digital terrain model was created by removing all, but the last return from the LiDAR data and then interpolating the ground surface (Figure 6) using the *lidR* package in R (Roussel and Auty, 2018). This allowed for a more accurate estimate because small changes in the surface, too small to affect elevation, were detected from this model.

A function was developed in R to estimate the height for a given tree using its planting location (from ESRI shapefile) and using a radius of 6 feet to subset the 3d point cloud and approximate the tree height by finding the highest Z coordinate in the subset of points close to the tree center. This was done for every tree detected (Figure 7). Of the 7,200 study trees, the function was able to detect heights for 7,143 trees.

Since this is a biomass study site, a collection of different genotypes is being used. To connect a given tree detection to from the LidR function to its specific genotype an attribute table from ESRI Arc GIS was connected to each detected tree. **Figure 8** shows the tree tops detected (red dots) with the known tree positions (black dots). Tree detection and tree

position typically matched well with each tree, with some variation among sites and replications.

The detected tree tops were placed on top of the DTM and height was calculated by difference (Figure 9). The estimation was calculated by subtracting the trees highest Z coordinate (tree top) from an average of Z coordinates below the tree top from the DTM. The resulting value is the estimated tree height.

4.3.2. Tree height estimation

In this study, we compared the tree heights determined using the drone imagery to the field measurements. Minimum, maximum, ranges, and averages of difference in measured height and PhoDAR height were observed (Table 1). Across both sites, replication 4 had the smallest minimum difference of 0.0005 m. Replication 3 had the largest difference range from -5.53 to 4.43, while replication 5 had the smallest range from -1.91 to 1.23. The absolute value of the average differences resulted in replication 5 with the smallest average difference of 0.53 m.

The average measure height and PhoDAR was variable across site and replication (Figure 10). For replications 2 and 5, the estimated height using PhoDAR was higher than the measured heights by roughly a half to one meter. Replications 3 and 4 had the closest averages between measured heights and PhoDAR heights.

The correlation between field measured heights and PhoDAR derived heights were 0.18 (Figure 11). However, correlations varied considerably by replication. The correlation between measured heights and PhoDAR heights for Site 1, replications 1 through 4 was very

low with a value of -0.10 (Figure 12). Site 2, containing replication 5, however, had a very large correlation, 0.79, among field-measured heights and PhoDAR heights (Figure 13).

4.3.3. Cost Analysis

A cost comparison for field-based measurements and UAS was done for this study. Up front cost for equipment is slightly higher for PhoDAR measurements. The Matrice 100 Quadcopter (\$3,299) and DJI Zenmuse X5 camera (\$1,679) totaled roughly \$5,000. For the field-based measurements three vertex hypsometers were used to measure heights of all the study trees. Cost per unit of vertex hypsometers are \$1,489; for three that is \$4,467.

The mileage to and from the site, days spent measuring, and number of workers was significantly more for the field-based method in comparison to the PhoDAR method (Table 2). Measuring heights using the drone took one day, to and from the site (60 miles), with two workers. Field measurements took 540 miles (60 miles round trip x 8 days), 8 days to measure study site, and 8 workers.

When comparing costs of only labor between methods, PhoDAR is a cheaper method. The price for a company to fly the drone and processing the data was \$2,000. The price for measuring and processing the field measurements (8 workers x \$12/hour x 277) for the entire study was \$3,324.

The total cost, equipment plus labor and processing, was cheaper for the PhoDAR method. The total cost using the traditional field-based method was \$7,891. Whereas the total cost using the PhoDAR method was \$5,458.

4.4. Discussion

4.4.1. Tree Identification

In this study, we tested the performance of low-cost systems based on UAS imagery acquired from RGB cameras and image reconstruction in conjunction with LiDAR data for detecting individual trees and height measurements. Tree detection was very high and height estimations were significantly variable by site and replication. The data suggest promising results for high density stands with crown overlap.

Overall tree detection was high in this study with 98% detection from the 7,200 study trees across both sites (7,143 trees were detected). The PhoDAR data had limited ground detection because the data are generally all first returns, with no last returns. Gaps in the study replications provided some ground points, but overall ground surface could not accurately calculate tree heights. The integration of LiDAR data for nDTM helped improve height estimations greatly. The DTM derived from LiDAR was able to detect not only changes in elevation, but also small changes across the ground surface resulting in a more accurate height estimation.

4.4.2. Tree height estimation

Height estimation compared to field measurements had a low correlation of 0.18. This could have been due to the high density spacing of the stands. Also, the larger proportion of forked trees may have affected the detection of tree top. The average proportion of forked trees over both sites was 59%. Close spacing in conjunction with high proportion of forking may have affected tree top detection and accurate height estimation.

Tree positions from ArcGIS joining with tree top detection could have affected height estimation. From **Figure 8** there is less variation between tree top detection (red dots) and known tree positions (black dots). Site 2, the tree tops and tree positions are on top of one another, whereas on Site 1, there is more variation. This difference in tree position and tree top could be a result of the tight spacing or the high proportion of forked trees affecting the overall height estimation.

Correlations across site varied greatly with Site 1 having a correlation of -0.10 and Site 2 having a correlation of 0.79. The difference in correlation may be due to number of reference points. Site 1 had four replications, with replication 1, 2 and 3 in a group and replication 4 separated to the side. A total of 6 reference points were placed around all four replications (See Figure 1). Whereas Site 2, with only one replication had reference point on all four corners creating a smaller more uniform reference field. The wide reference point placement on Site 1 created a gradient of accuracy, with points near the reference plots very accurate and the farther away points less accurate, creating a “bowl” in the imagery (James and Robson, 2014). This bowl could explain the differences in accuracy from Site 1 to Site 2. For future application, ways to eliminate or minimize the “bowl effect” in the processed imagery is collect oblique imagery or to add suitable control points.

Another possible source of error may have been from the Vertex hypsometer height estimates from the traditional field-based method. In a dense stand, such as this study, often heights are over or under estimated because of the difficulty in seeing the tops of trees. This source of error could have contributed to the variation we found between PhoDAR method and the traditional field-based method.

4.4.3. Cost analysis

For this study, using PhoDAR technology was cheaper than traditional field-based measuring. The difference in cost for equipment plus labor was roughly \$1,000. Not only is there an added cost benefit, but there are also some considerable advantages to using UAS technology over traditional field-based methods.

The advantage to using UAS methods compared to traditional methods are the increased efficiency. UAS technology allows for measuring larger areas that can often be difficult to access in a shorter period of time.

However, as seen in this study, measurements can be grossly under- or overestimated depending on site. One way to correct this is to use more ground control points will take more time and effort. Another way to increase accuracy is to combine the PhoDAR imagery with LiDAR data which increases costs greatly. However, for a site or area where monitoring or rough estimates will suffice, PhoDAR derived imagery is the cheapest more reliable method.

The results of this study not only show the promising results of PhoDAR imagery, but also highlight the common issues with such technology. However, correlation between field measured heights and PhoDAR derived heights from Site 2 are very promising for measuring high-density stands. Further research could focus on the isolation of error e.g., by adding more ground control points or oblique imagery to the nDMS process.

4.5. Conclusions

In conclusion, PhoDAR methods are not mature enough to replace ground-based manual assessment of tree height in genetic field tests. Genetic field tests of loblolly pine are measured at earlier ages (four to six) in the southern US. The assessments of the genetic field tests are carried out before crown closer. The PhoDAR methods should be tested in such young progeny trials to collect more evidence about the utility of this method in progeny tests assessments.

4.6. Literature Cited

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4.7. Tables

Table 1. Minimum, maximum, and average difference (in meters) between measured heights and PhoDAR derived heights. Absolute value was used for minimum difference and average difference. Replication 5 (Site 2) had the smallest range in difference than the other replications on Site 1.

	Replication	Minimum Difference	Maximum Difference	Average Difference
Site 1	1	0.0016	-3.43, 4.93	1.34
	2	0.0064	-5.80, 2.92	1.42
	3	0.0007	-5.53, 4.43	1.00
	4	0.0005	-4.59, -4.24	0.92
Site 2	5	0.0016	-1.91, 1.23	0.53

Table 2. Cost of measuring the study including mileage to site, days spent measuring site, number of workers used, and hours spent measuring and processing data. Measuring heights from PhoDAR derived imagery is roughly \$1,500 less than tradition field-based methods.

Method	Mileage	Days	Workers	Hours	Labor cost (US \$)
Field – based method	540	8	8	265	3,180
Data processing			1	12	244
Total					3,424
UAS method	60	1	2	4	1,500
Data processing			1	2	500
Total					2,000

4.8. Figures



Figure 1. Layout of study sites. Site 1 has four replications, whereas Site 2 has one replication. The red triangles indicate ground control points (GCP) used to collect GPS coordinates.

a)



b)



c)



Figure 2. Orthomosaic derived from drone photography. Site 1, Replications 1-4 (a), Site 2, Replication 5 (b), closeup showing edge of Replication 4 and drone flying process (c).

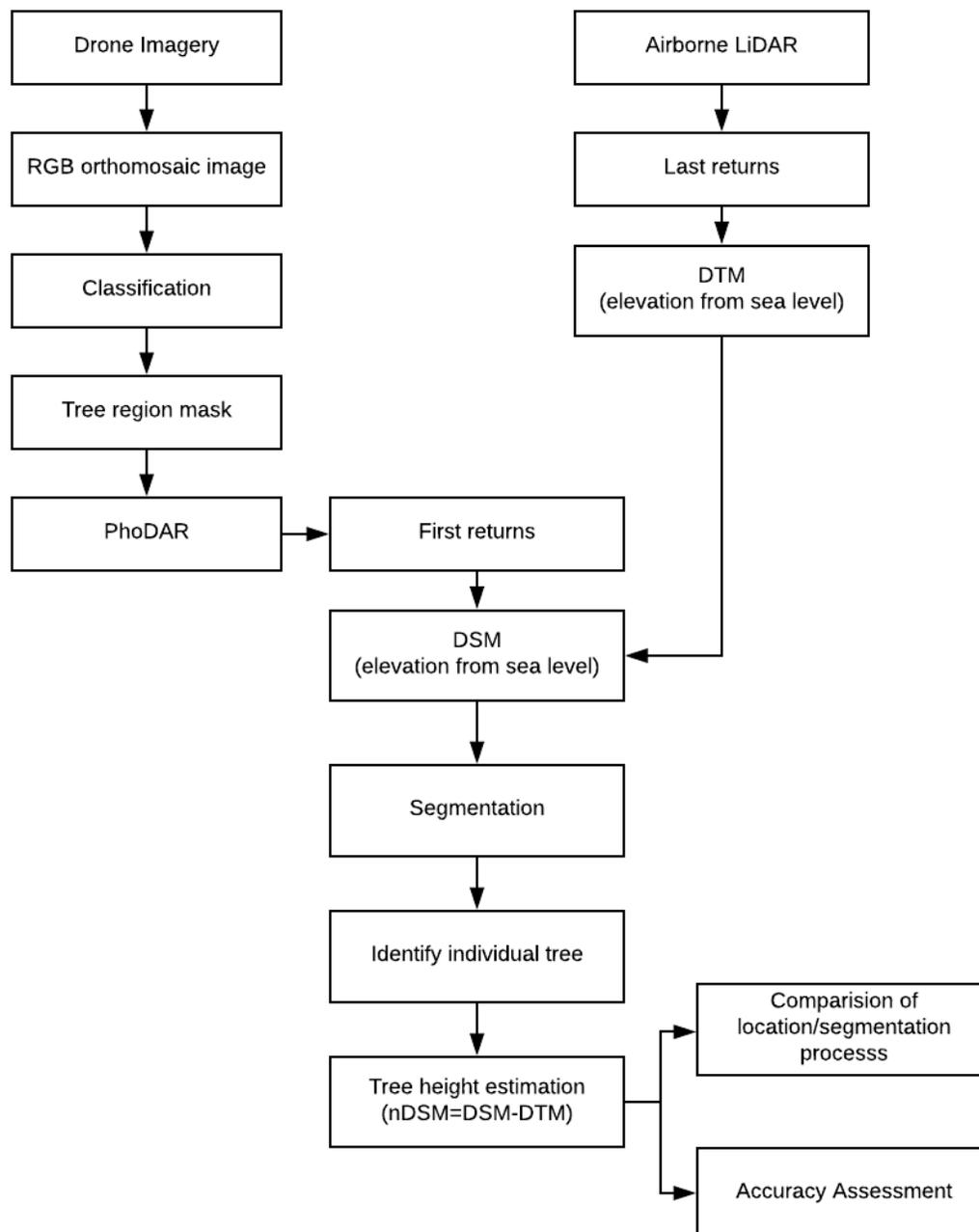


Figure 3. Flowchart of the experimental design used to collect, test, and analyze the methods used.

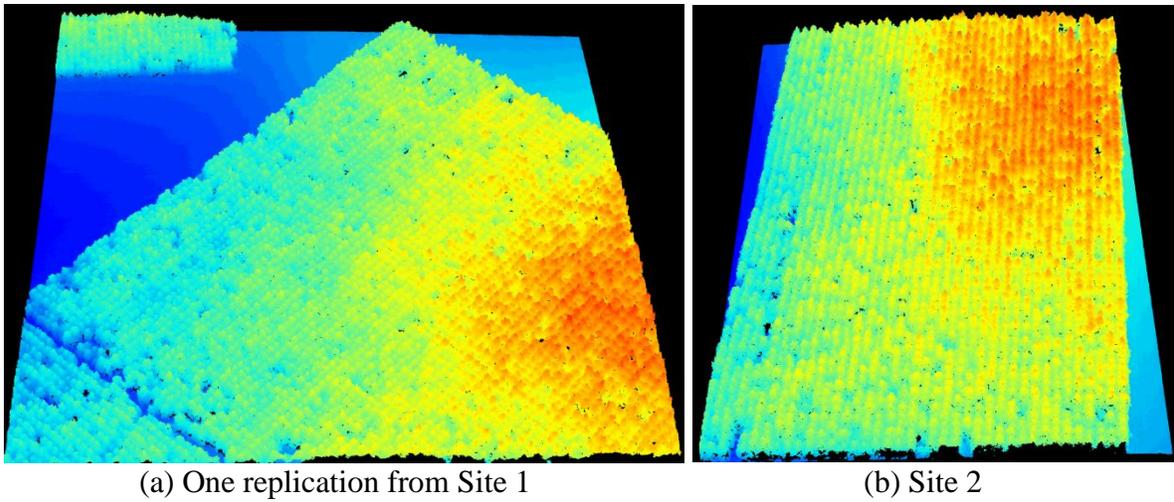
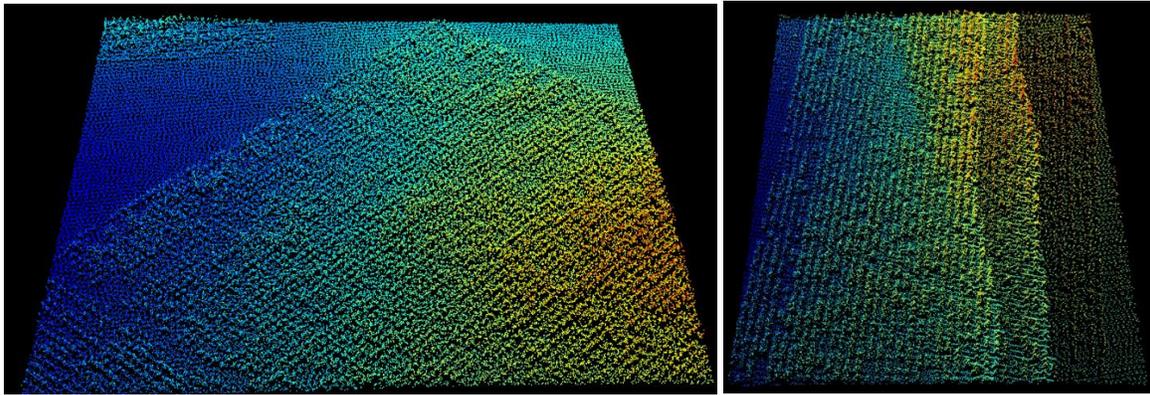


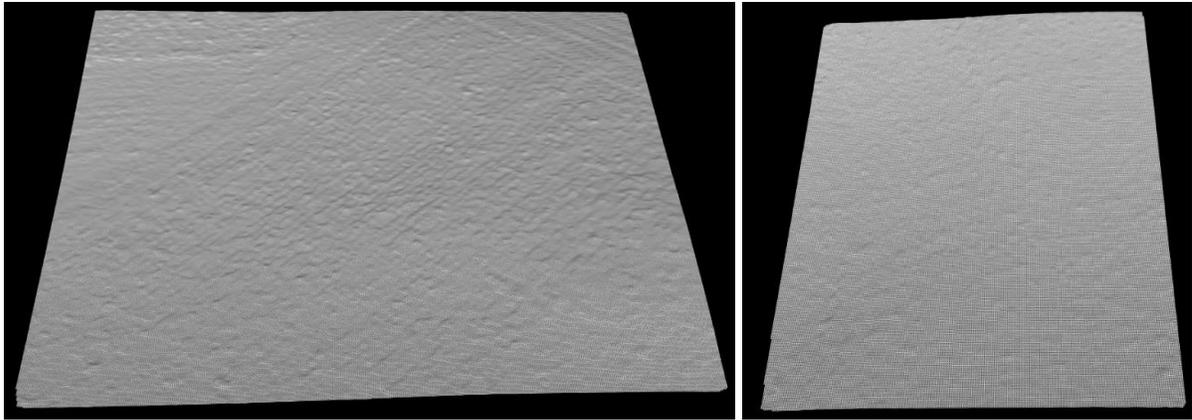
Figure 4. Extracted 3d point cloud derived from orthomosaic (PhoDAR) displayed in R using *lidR* package. Image *a* is a replication from site one and image *b* is site 2. The warmer colors depict higher elevation than the cooler colors.



(a) One replication from Site 1

(b) Site 2

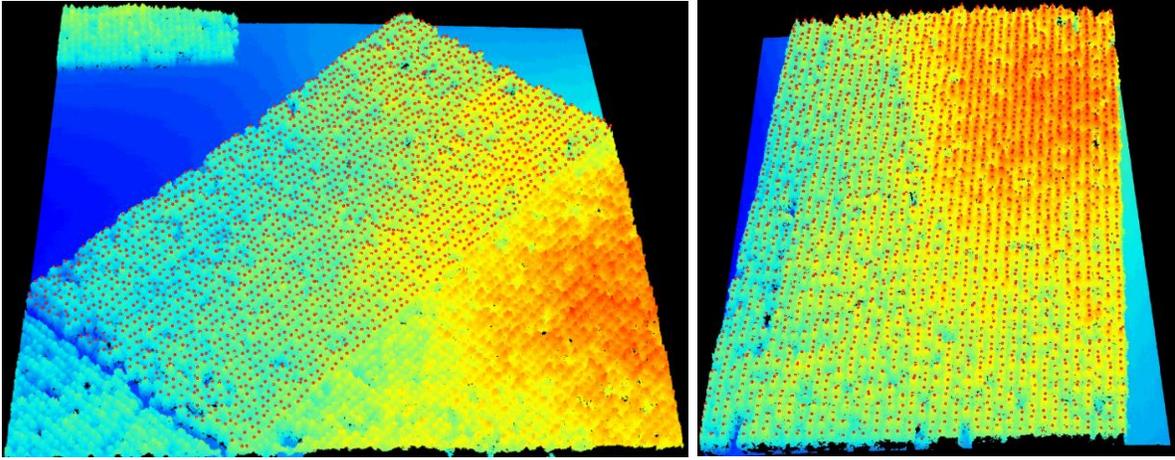
Figure 5. LiDAR point cloud displayed using lidR package. LiDAR data downloaded from NC SDD was from 2014, corresponding to experiment age 3.



(a) One replication from Site 1

(b) Site 2

Figure 6. Digital Terrain Model derived from 2014 LiDAR data by ground filtering and then interpolating to create the surface model. This DTM shows small topography changes for a more accurate estimate.

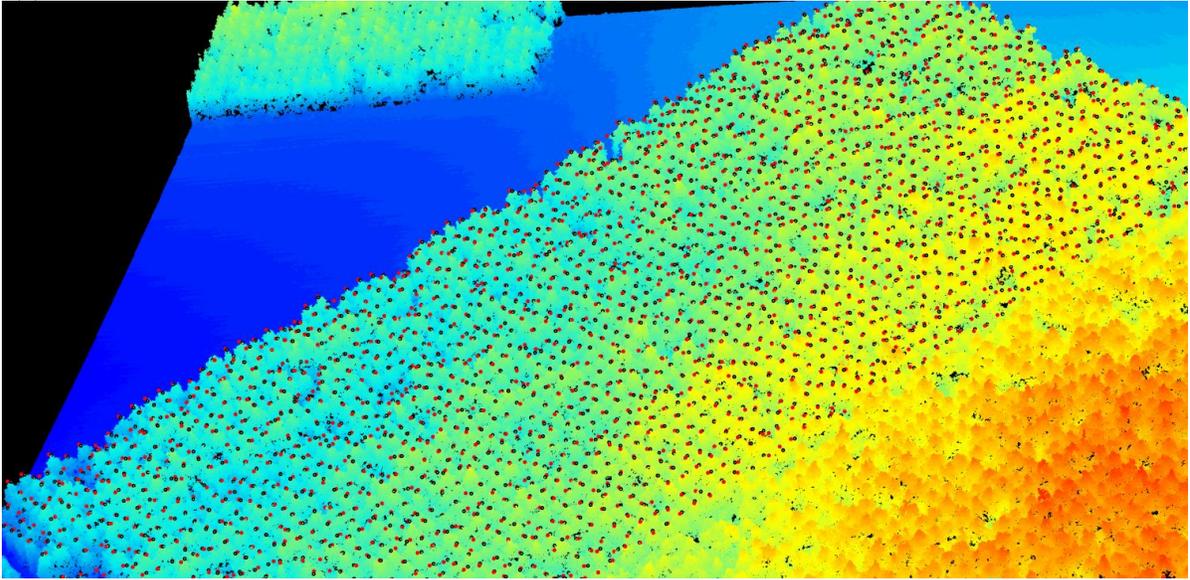


(a) One replication from Site 1

(b) Site 2

Figure 7. Tree height (red dots) from PhoDAR displayed over 3d point cloud.

(a) Site 1



(b) Site 2

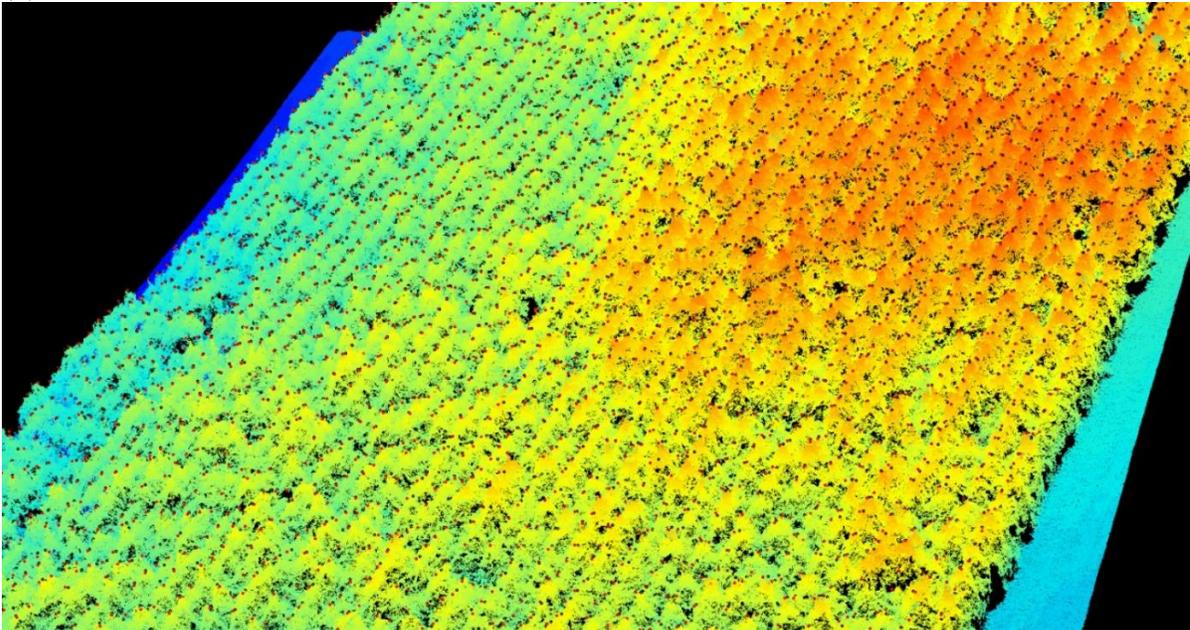
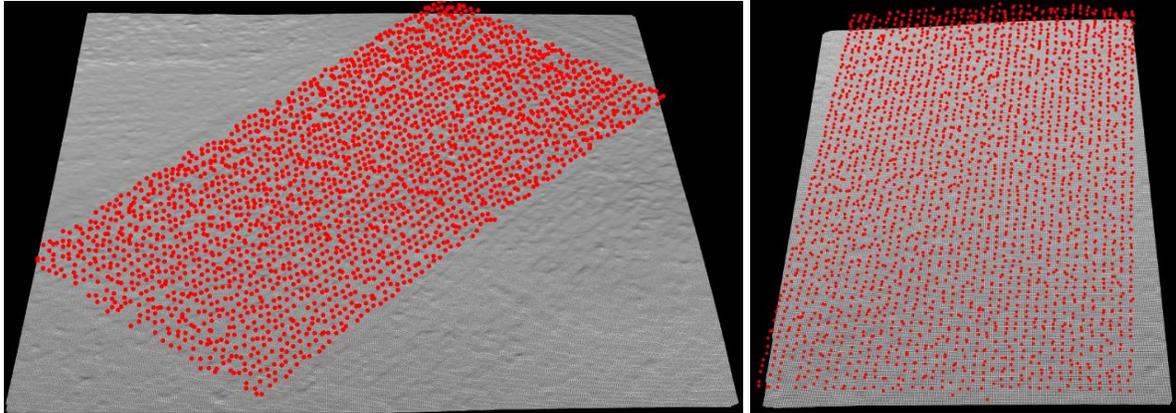


Figure 8. Detected tree heights (red dots) and the known tree positions (black dots). Overall, tree tops and tree position matched, with slight variation across replications.



(a) Site 1

(b) Site 2

Figure 9. Detected tree heights displayed over DTM. The points (red dots) minus the DTM value under that point is the height estimate.

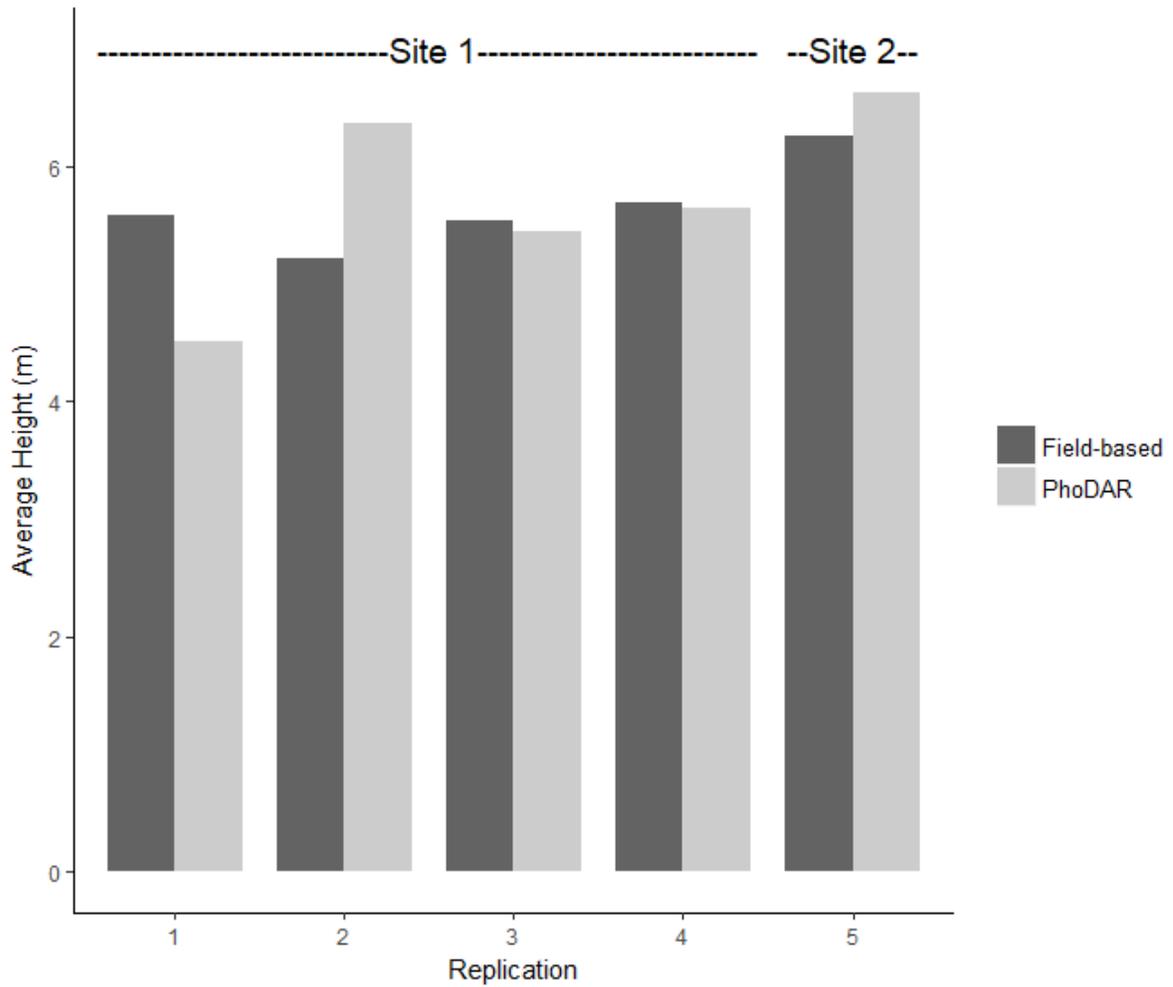


Figure 10. The average height across site and replication for field measured heights and PhoDAR derived heights. Replication 3 and 4's height averages were the most similar while replication 1 and 2 differed the most.

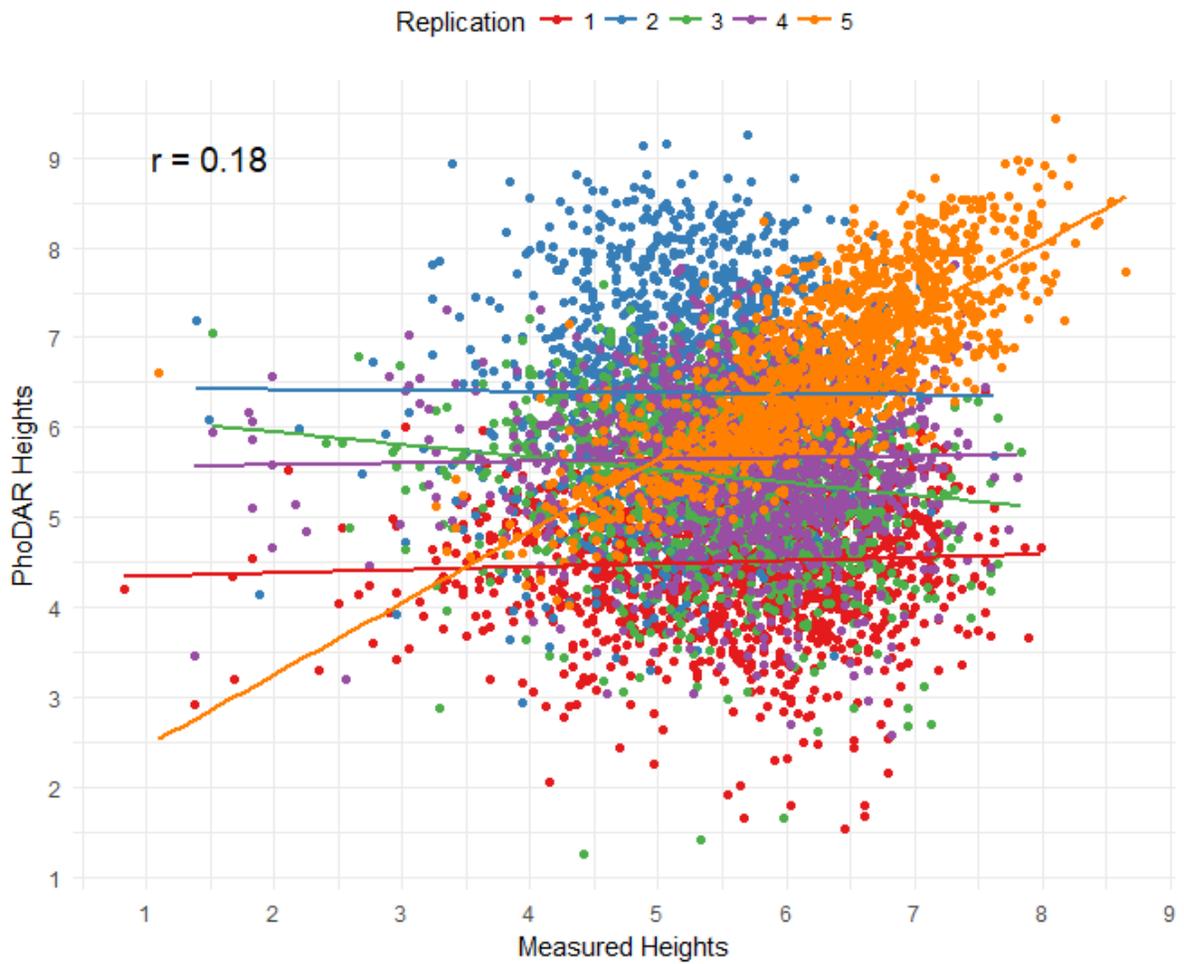


Figure 11. Overall correlation between measured heights (x-axis) and PhoDAR derived heights (y-axis) of all replications (1 – 5). Each color represents a different replication with its own regression line. The overall correlation of all replications was low. However, replication 5 had a strong positive correlation ($r = 0.79$).

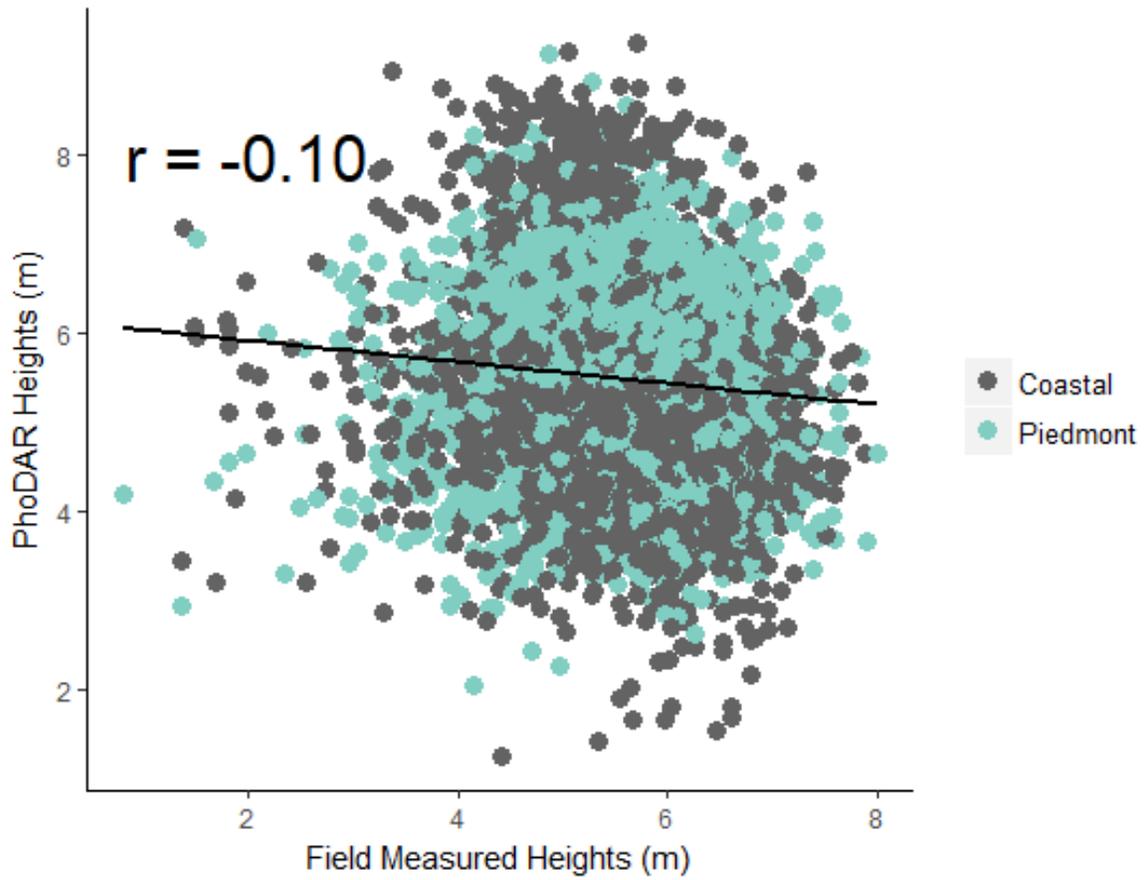


Figure 12. Correlation between field measured heights (x-axis) and PhoDAR derived heights (y-axis) for site 1. There was very little correlations on site 1.

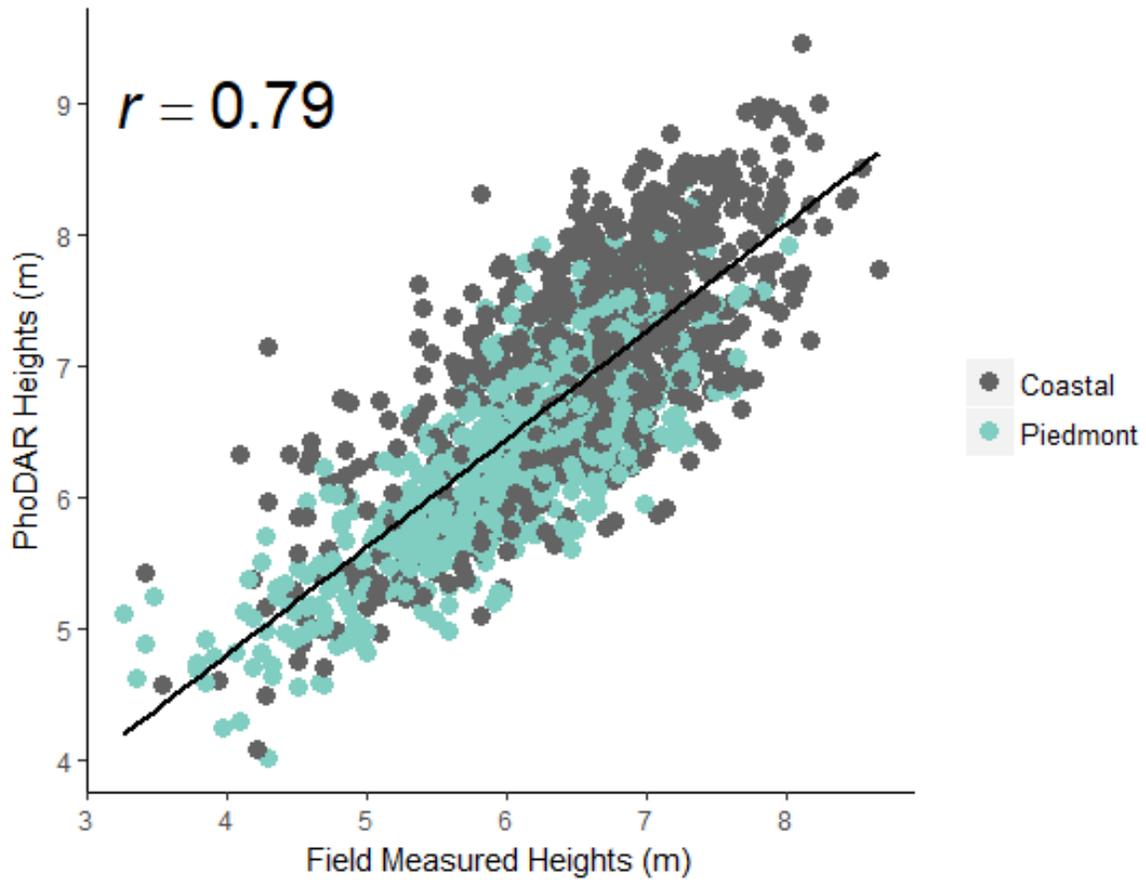


Figure 13. Correlation between field measured heights (x-axis) and PhoDAR derived heights (y-axis) for site 2. The correlation for Site 2 was very strong, higher than any other replications or sites in the study.

APPENDICES

Table A.1 List of families within study by seed source origin. The family identifiers have been shortened for readability.

Piedmont		Coastal	
Family Name	ID	Family Name	ID
TIP2274493	P1	TIP216059	C1
TIP224675	P2	TIP138245	C2
TIP1139704	P3	TIP2440610	C3
TIP1234675	P4	TIP2120526	C4
TIP1439243	P5	TIP275099	C5
TIP1386410	P6	TIP1256535	C6
TIP1460510	P7	TIP1162449	C7
TIP1743328	P8	TIP1711512	C8
TIP1265956	P9	TIP1417199	C9
TIP1929303	P10	TIP1192343	C10

Figure A.1 Receiver operating characteristic (ROC) curve for the model used as a model fit statistic. This plot illustrates the diagnostic ability of a binary response variable. The ROC for the model is roughly 71%, meaning the model predicts the correct response 71% of the time.

