VARIATION IN THE FLOOD TOLERANCE OF THREE MIDWESTERN OAK SPECIES

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The undersigned, appointed by the Dean of the Graduate School, have examined the thesis entitled:

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a candidate for the degree of Master of Science, and hereby certify that in their opinion it is worthy of acceptance.

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CHAPTER 1

INTRODUCTION

Purpose

The purpose of this study is to evaluate the flood tolerance of selected oaks from seed sources collected along a topographic gradient to improve seed collection strategies for the purposes of growing seedlings for outplanting in flood-prone areas of Missouri and elsewhere. One of the regional guidelines for the Missouri Department of Conservation calls for the restoration and maintenance of riparian habitat as bottomland hardwood forests and wetlands, thereby underscoring the importance of flood tolerance evaluation. Most bottomlands in Missouri were cleared for crop production during the last century and this forest clearing and conversion to agriculture has eliminated up to 95% of bottomland forests in the Missouri and Mississippi River basins (Dey and others 2001). The devastating late summer flood of 1993 destroyed over 325,000 ha of cropland in the lower Missouri River floodplain by scouring holes and depositing sand in agricultural fields (Grossman and others 2003). Restoration efforts are underway and there is a desire to restore more natural floodplain vegetation and hydrologic regimes (Dey and others 2001). Public land managers and private land owners have a strong interest in regenerating native oak (*Quercus* sp.) on what had been converted to largely agricultural floodplains (Dey and others 2006). Attempts to establish oak and other hard mast species in bottomland fields have often failed despite the best efforts of land managers (Grossman and others 2003).

Bottomland forests subject to periodic flooding are extremely productive in terms of wood production and wildlife habitat. These forests contain bottomland hardwood species well-adapted to periodic flooding (Hodges 1997). The ability of a tree to tolerate periodic flood events is dependent upon a number of factors including soil characteristics, topography, season of flooding, and its genetic makeup. Although there have been numerous studies describing the tolerances to flooding and other environmental stresses of the oak species, little is known about genetic variation within and among bottomland oak species for these characteristics. A central issue during restoration is how well is the preservation and restoration of the original biodiversity, especially the genetic diversity of the woody component, and whether appropriate seed sources are being used.

Currently the oak seedlings produced at the MDC state nursery originate from acorns most likely collected from upland seed sources, because of the ease of collection. It is unclear to what extent tree planting failures on bottomland sites and within riparian forest buffer strips which are subject to periodic flooding may in part be due to the use of maladapted planting stock from upland seed sources. This is of particular concern for the oak species that inhabit both uplands and bottomlands such as bur and swamp white oak. Establishment problems have been observed due to poor seed sources and prescribing species that were not adapted to the site (Stanturf and others 2001). Seedlings transplanted (outplanted) for bottomland forest restoration (afforestation) must be able to survive intermittent flooding that occurs frequently in bottomland forests (Anderson and Pezeshki 1999). Other challenges arise due to differences in flooding depth, frequency and duration as a function of microtopographic variation within the floodplains (Battaglia and Sharitz 2004). Though the following research project that will be presented is limited to oak families (seed sources) native to Missouri, the research stands to benefit all those who wish to improve seed collection strategies for restoration of riparian corridors (e.g., the Mississippi River Valley bisecting other states). This research will benefit land owners and managers if the research is able to elucidate physiological, morphological, and genetic characteristics of oak seedlings that allow them to tolerate periodic flooding characteristic of bottomland sites.

Objectives

The objectives of this study are: a) To determine differences in survival, recovery, and growth of oak seedlings grown from acorns collected from contrasting topographic positions subject to different flood treatments; and b) To determine the extent of genetic variation to flood tolerance among open-pollinated families of swamp white, bur, and white oaks from trees from contrasting topographic positions.

Hypothesis

The following hypotheses have been developed for this research project: a.) There is no difference in the survival, recovery, and growth of potted seedlings of swamp white, bur, and white oak to flood treatments; b.) There are no differences in the genetic variations to flood tolerance among open-pollinated families from contrasting topographic positions; and c.) There are no differences in the flood tolerances between seedlings grown from upland and bottomland seed sources.

CHAPTER 2

LITERATURE REVIEW

Flood Tolerance

Riparian and bottomland forest ecosystems are important for their high productivity, biodiversity, and ecological services , e.g. mitigation of floods and erosion, removal of nutrients from agricultural runoff, alleviation of pollution effects, and creation of habitats for birds and mammals (Kozlowski and Pallardy 2002). Flooding can positively and negatively influence the growth and survival of woody plants. In flood-intolerant plants, flooding reduces transpiration and photosynthesis (Striker and others 2005). In contrast, those same parameters may be enhanced in flood-tolerant plants to allocate biomass to the above-ground (above-flood) components (Naidoo and Naidoo 1992). The degree to which these events occur depends on the individual flood tolerance of the selected species (Striker and others 2005).

Flood tolerance has been described by numerous researchers and professionals in the field. Flood tolerance is largely the physiological adaptation of plants to anoxic conditions, toxic substances, and other associated changes in soil properties induced by flooding (Unger and others 2007). Flood-tolerant tree species recover net photosynthetic potential after acclimating to anoxic soil conditions, and net photosynthesis recovers more quickly and to a higher extent for flood-tolerant tree species when drained compared to flood-intolerant species

(Anella and Whitlow 1999). Flooding tolerance refers to a species' ability, from seedling stage to maturity, to tolerate soil saturation or inundation during the growing season (Hook 1984). The responses of woody plants to flooding vary with species and genotype, plant age, condition of the floodwater, and time and duration of flooding (Kozlowski and Pallardy 1997). Flooding-intolerant plants can withstand anoxia (lack of oxygen) temporarily, but not for prolonged periods (Taiz and Zeiger 2002). In contrast, some flood-tolerant plants, specifically bald cypress (*Taxodium distichum* L.), can withstand partial inundation for one to two years.

The ability of a tree to tolerate periodic flood events is dependent upon a number of factors including soil characteristics, topography, degree of flooding (season, depth, etc.), and inherent genetic makeup. Flood tolerance is associated with the ability to survive and recover, refoliate, and initiate new shoot growth after flooding (Anderson and Pezeshki 1999). A major contributing factor to flood intolerance is probably the lack of dissolved oxygen in highly saturated soils. Water-filled pores lead to decreased oxygen concentrations in the soil that primarily affect root systems that, in turn, affect the above-ground components of plants (Pigliucci and Kolodynska 2002). With water inundation of soils, the activity of roots and microorganisms decline quickly with decreasing oxygen availability (Striker and others 2005).

Major adaptations for flood tolerance of some woody plants include the capacity to produce adventitious roots that physiologically compensates for the decay of original roots under soil anaerobiosis, facilitation of oxygen up-take through stomata and newly formed lenticels, the formation of aerenchyma tissue, and metabolic adjustments (Kozlowski and Pallardy 1997). The development of arenchyma tissue in response to flooding creates large intercellular spaces to facilitate oxygen diffusion (Coutts and Phillipson 1978; Kozlowski and Pallardy 2002). The formation of adventitious roots is positively correlated with flood tolerance (Sena Gomes and Kozlowski 1980). Plants can undergo biochemical or metabolic adaptations to facilitate flood tolerance that enables them to survive under anaerobic conditions by maintaining a glucose supply and avoiding accumulation of toxic compounds (Crawford and Finegan 1989; Kozlowski 2002). These adaptations induced by soil anaerobiosis may also negatively affect trees by altering respiration, photosynthesis, protein synthesis, mineral nutrition and hormone relations (Kozlowski and Pallardy 2002).

Oxygen uptake through plant-root systems is essential for respiration. Dissolved oxygen diffuses so slowly in (flood) water that only a few centimeters of water near the surface remain oxygenated (Taiz & Zeiger 2002). Under anaerobic soil conditions, as is the case with flooding, the formation of hypertrophied lenticels on submerged portions of the stems and roots may occur (Kozlowski 1984). Flood-induced lenticels allow for the exchange of dissolved gasses in flood water (Chirkova and Gutman 1972). The gasses (air) enter through lenticels on woody stems and roots, and travel by molecular diffusion, or by convection driven by small pressure gradients (Taiz and Zeiger 2002). Some flood-tolerant species absorb oxygen through stomata or lenticels and transport it to the roots, from which it diffuses to the rhizosphere (Kozlowski & Pallardy

2002). Unger and others (2007) found no difference in dissolved oxygen levels between flowing and stagnant water.

These and other aspects of flood tolerance have been examined in the past through case studies and more recently through replicated field studies and replicated pot studies. All of the flood oriented studies have positive and negative characteristics associated with each study type and within each study itself. The goal is to learn from past studies and to further elaborate on specific research objectives under a highly controlled system.

Case Studies

Most of the early work evaluating the flood tolerances of plants was carried out via un-replicated natural flooding events. These case studies provide useful information but were difficult to quantify. Lack of control in terms of the amount and duration of flooding exist with many case studies. Since case studies are not replicated in time and space, these preclude the use of parametric statistical analysis.

Bell and Johnson (1974) evaluated flooding effects of high reservoir levels around Rend Lake and Lake Shelbyville in Southern Illinois on mature trees. Flooding was facilitated by the abnormally high water levels caused by the spring and early summer flooding of 1973. Flooding on the Mississippi River further affected lake levels by inhibiting drainage from the lake into the river system. As a result, 24 species of trees were partially inundated and their respective flood tolerance ratings were determined by survival throughout the growing season. Survival was measured during the periods of flooding and post-flooding recovery.

Results from the study indicated that 30 days of flooding during the growing season was not significant to kill any established tree. Mortality among the predominantly upland species became apparent when flood conditions extended to more than two months. Species of the floodplain, for example bur oak (*Quercus macrocarpa* Michx.), were found to be completely tolerant of the partial inundation. All of the bottomland species successfully completed their annual growth cycle under partial inundation. By examining Bell and Johnson's (1974) study, it is clear that the amount and duration of flooding could not be controlled by the researchers. The lack of a non-flooded control treatment in this study creates difficulties for making comparisons with other studies. Their results indicate the need for a more controllable experimental design to elucidate specific research objectives related to determining the flood tolerances of younger trees (seedlings) in shorter duration flooding treatments.

Kabrick and others (2004) evaluated the effects of bedding on soil properties and on the early survival and growth of pin oak (*Quercus palustris* Muenchh.) and swamp white oak (*Quercus bicolor* Willd.). The study was carried out in two state conservation areas (Plowboy Bend CA and Smoky Waters CA) located within and outside of a 100-year levee in the Missouri River Floodplain in central Missouri. The area within the levee did not flood until the third year of the study. Initial data collections included total height and basal diameter at 2.5 cm above the ground level and data were collected following each growing season. Seedling survival and the presence of animal damage were determined each year as well. Because the sites did not flood during the first two years of this study, this research demonstrates the variation that can arise in natural flooding regimes. The Smoky Waters CA site was flooded twice in the first four years of establishment. Dey and others (2006) collected the data on both conservation areas and further analyzed their findings. In 2001 and 2002, the study site experienced flooding for up to three weeks in June with variable depths reaching a maximum of up to 1.5 m. This flooding depth was deep enough to fully inundate bareroot seedlings. There were not significant differences in survival of the oaks associated with flooding depths; however, difference in survival of the redtop ground cover was related to elevation and duration and depth of flooding (Dey, personal communication). Results of Dey and others (2006) may fortify the need for a more controlled manner in which to carry out these studies and in a replicated manner. Many researchers investigating plant response to flooding have conducted replicated studies in the field and under greenhouse conditions which may enable them to attain a higher degree of control over each study.

Replicated Field Studies

There have been several replicated field studies to qualify the flood tolerances of woody tree species. Several of these were carried out in constructed, flood-tolerance laboratories (FTLs) also called flooding research facilities (FRFs). One of these facilities is located in New Franklin, Missouri and the other in Skarkey County, Mississippi, USA. Large, field facilities are needed to evaluate and compare flood tolerances among large numbers of hardwood seedlings.

Van Sambeek and others (2007) described initial implementation of the University of Missouri Center for Agroforestry's FTL located at the University of Missouri Horticulture and Agroforestry Research Center in New Franklin, Missouri, USA. Construction of the FTL began in 1999 and modifications to the FTL have occurred several times since. The FTL consists of twelve parallel 6-m wide by 180-m long independent channels. Soils within the channels consisted of moderately-well drained Nodaway silt loams and varied with distance from Supher Creek. Water pumped from a nearby constructed retention pond can be pumped independently into each channel to control timing, depth, and duration of either stagnant or flowing flood water. Extra care was taken to maintain the original bottomland soils that occurred on this riparian site located in the Missouri River floodplain along Sulphur Creek. Each channel was created by constructing 6-m wide by 2-m high berms around each channel using soil excavated for the retention pond. Problems have been encountered in this study that have caused complications for some of the research. The majority of the problems arise from the lack of control over extreme weather events. Heavy rains can create excessively saturated conditions in the non-flooded control channels. During one study year, up to forty-five centimeters of rain fell in one month and caused back flow from the adjacent Sulphur Creek to flood the facility. The ability to remove the water and return soil moisture below field capacity has been problematic. Control channels that were not subject to flooding received seepage water from under the berms from adjacent flooded channels and results in saturated soils. Micro-site differences have been found in soil properties (e.g., soil pH and structure) across the channels moving away from the creek. This publication reveals the need for a more controllable study where natural weather conditions, seepage, and changes in soil properties will not play such a significant role in the reported results.

Lockhart and others (2006) evaluated the flooding research facility (FRF) located in Sharkey County, Mississippi, USA on the Theodore Roosevelt National Wildlife Refuge Complex. It is adjacent to the headwaters of the Dowling Bayou that flows into the Big Sunflower River. The site was constructed by the United States Fish and Wildlife Service in 1994 and has undergone several improvements and is fully operational. The FRF contains twelve cells that can be artificially and independently flooded to specified levels and durations. Each cell is 18.3-m-wide by 201-m-long with a 2.4-m-tall levee on each side with a 3-m-tall levee surrounding the entire site. A 3-m-tall hog fence surrounds the entire area as feral hogs are located in the area. The large size of each cell allowed for the construction of shade houses within each channel to evaluate not only flood tolerances of plants, but the interactions between and among light and flooding. Problems associated with the FRF are shared with the FTL in Missouri. Difficulties exist in removing the flood waters in a timely manner. Both flood tolerance facilities are needed for rapid routine evaluation of large numbers of plants and for conducting multiple studies simultaneously.

The studies that have been carried out in these two facilities have been valuable to help describe the flood tolerances of relatively large numbers of seedlings under near-natural conditions for specific plant species. Researchers have used these facilities to conduct research relating to flood tolerances of plants (Van Sambeek and others 2007) and even soil responses to flooding using soil monitoring equipment (Unger and others 2007). Kabrick and others (2007) used the FTL in New Franklin, Missouri to evaluate the flood tolerances of bottomland hardwood seedlings. In their study, twenty five 1-0 seedlings from six commonly planted bottomland species, including bur oak and swamp white oak were planted in all twelve channels. Within blocks, single treatments were randomly assigned to each channel as a five-week stagnant flood, a five-week flowing flood, a three-week flowing flood, and a non-flooded control. Flooding depth was twenty to twenty-five centimeters and seedlings were evaluated for survival, shoot growth or dieback, and seedling health at the end of the growing season. They found that flooding decreased the survival, growth, and health of all species compared to the control. A five-week flowing and stagnant treatment was used to determine if either treatment would result in higher mortality related to differences in soil anoxia. Results indicated there was little differentiation between flowing and stagnant flooding treatments.

Kabrick and others (2007) indicated consistencies and discrepancies with published flood tolerance ratings for the evaluated species. Of the oak species used in this study, bur oak seedlings suffered the most through reductions in survival and growth and the presence of chlorotic leaves at the end of the growing season. Findings suggest that swamp white oak seedlings were much more tolerant to flooding than any of the other oaks examined. The researchers suggest future studies were needed to evaluate stock of known genetic origin and ecotype (upland vs. bottomland) to determine their role in governing flood tolerance.

Using the FTL Coggeshall and others (2007) investigated the genotypic variation in the flood tolerances of three southern bottomland oaks and black walnut (*Juglans nigra* L.). Seed from seed sources in western Tennessee were collected and sown in replicated nursery beds in the Flint River State nursery located in Byromville, Georgia, USA. Seedlings were lifted, sorted, and individually evaluated by root collar diameter, number of first order lateral roots, and stem length. Seedlings were transported to Missouri and planted in the Flood Tolerance Laboratory in the nine channels. Using a split-split-plot design with three blocks, three flooding treatments were applied: a 15 cm-deep flowing, a 15 cm-deep stagnant, and a non-flooded control treatment. Flooding responses included percent survival, total number of new shoots, total length of new shoots, and percent of surviving seedlings with basal sprouts. Non-flooded control channels were subject to soil saturation due to seepage and backflow flooding of nearby Sulphur Creek resulting in saturated soil conditions for more than seven weeks (Van Sambeek and others 2007). Therefore the results may be difficult to interpret.

Lockhart and others (2006) have used the FRF in Mississippi to evaluate flood tolerance of pondberry (*Lindera melissifolia* Walt.), an endangered shrub species. A three year study to evaluate flooding and light regimes is currently underway and will assess survival, morphology, physiology, growth, and biomass accumulation. Flood treatments consist of a non-flooded control and partial inundation (five to ten cm) for a 45-day flood, and a 90-day flood. Shade structures within each channel provided 5, 37, and 100 percent of full sunlight. The main problems that arose in the FRF was the inability to drain the channels in a timely manner (Lockhart, personal communication).

Other flood tolerance research has been carried out without the use of a large elaborate outdoor flooding research facility in specially designed greenhouse facilities. Angelov and others (1996) researched long- and short-term flooding effects of swamp-adapted trees including two species of oak. This experiment was conducted at the Santee Experimental Forest in Berkley County, South Carolina, USA. This research was carried out in a hydroedaphytron which is comprised of twelve concrete tanks 1.8-m in all dimensions and contains a specific profile of rock below local alluvial soils. The research investigated the flooding responses of four bottomland species. Five-month-old seedlings grown from locally collected seed sources were subject to three flood treatments. A pond-type treatment exposed seedlings to stagnant-flood water to a depth of 15-centimeters above the soil surface for the duration of the study; a swamp-bottomland treatment exposed seedlings to a flowing flood water treatment to a depth of fifteen centimeters followed by dry cycles at three week intervals; and an upland-type treatment as the non-flooded control. The study was conducted over two years and flood responses were evaluated through periodic data-collections of seedling height, root collar diameter, survival, and visible morphological changes. Results indicated that flooding had a negative effect on seedling survival and formation of new leaves (leaf areas) for the flooded seedlings when compared to seedlings in the non-flooded control treatment. Flooding resulted in the formation of hypertrophied lenticels, and the presence of chlorotic leaves for the flooded seedlings.

Mortality of oak seedlings occurred in phases associated with periods of major vegetative growth, specifically after spring bud break and stem elongation and after summer stem elongation (Angelov and others 1996). For both oaks, after flood waters partially inundating the seedlings were drained, leaves formed during the dry period had three to four times greater leaf areas than those formed during flooding periods. Continuous flooding caused several morphological changes including the formation of lenticels and the presence of chlorotic leaves.

Replicated Pot Studies

Replicated pot studies have been used to investigate the flood tolerance of select plant species. Using replicated pot studies the researcher is allowed a high degree of control over many aspects of the study. Replicated pot studies can be carried out using plants grown in individual containers together with a uniform soil medium. Control of floodwater depth and duration is more frequently achieved and a quicker return to soil field capacity. These studies can occur in a greenhouse with excellent control of environment or in an outdoor laboratory.

Cao and Connor (1998) investigated the flood tolerances of 60-day-old cottonwood (*Populus deltoides* Bartr. ex Marsh.) seedling clones from cuttings for reforestation (afforestation) projects in China. Trees were grown under greenhouse conditions for 60 days in 15 x 25 cm plastic nursery pots containing a commercial growing mix (Metro-Mix 220®). Plants were subject to three treatments: a watered non-flooded control, a shallow flood with cuttings inundated to a depth of three centimeters above the soil line, and a deep flood with the complete inundation of cuttings. Flooding duration lasted 42 days and seedlings were evaluated for height, leaf area, transpiration rate, and dry weight of roots and above ground components.

Cao and Connor (1998) found that flooding caused reduced leaf areas and reduced root and shoot growth when compared to the non-flooded control treatment seedlings. Leaf size, leaf area, and number of leaves were all reduced for all flooded plants relative to watered-only plants. Flooding inhibited root growth and caused some deterioration of the original root system and also induced hypertrophied lenticels on the flooded portion of the stem. Flooding inhibited leaf initiation in their study and leaf chlorosis and abscission occurred. The diverse responses to the measured parameters suggested that there is a potential for enhancing tree productivity by selecting the more flood tolerant (cottonwood) clones for planting in (China's) wetlands.

Tang and Kozlowski (1982) investigated the responses of bur oak seedlings to flooding and specifically examined morphological changes and ethylene production. Forty two families of locally collected seed were used in this study. Four-week old seedlings were assigned to have 24 seedlings flooded to a depth of 4 cm above the soil line for 28 days and 18 seedlings assigned to a non-flooded control treatment. Data collection occurred after 7, 14, and 28 days following the initiation of flooding treatments and included height and number of leaves. Final data collections examined dry weights of stems, leaves, and roots.

Tang and Kozlowski (1982) found a negative effect on biomass accumulation and root and shoot growth for the flooded seedlings when compared to the non-flooded control-treatment seedlings. The presence of hypertrophied lenticels appeared by the fifth day of flooding and increased in quantity thereafter with continued inundation. Results indicated that the dry weights of flooded plants were twenty five percent less than the dry weights of non-flooded plants. Leaf growth was only slightly inhibited and stem growth was intermediately affected; while growth of the roots was affected the most by flooding.

Conner and others (1998) investigated the survival and growth of four bottomland oak species in response to flooding and salinity. One-year-old nursery-grown, bareroot seedlings were transplanted into plastic nursery pots fifteen centimeters in diameter and twenty centimeters tall. Seedlings were subjected to various treatments of fresh and saline water for approximately one year. Survival and height growth was taken biweekly and root and stem components were dried and analyzed at the end of the study.

Connor and others (1998) found a negative effect due to flooding on seedling morphology which consisted of reduced growth for the flooded seedlings when compared to the non-flooded control-treatment seedlings. Flooded plants grew significantly less in diameter and overall height than seedlings in the non-flooded control. In turn, the dry weight of root and stem components were also less for flooded seedlings.

Pezeshki and others (1999) investigated the effects of intermittent flooding on seedlings of nuttall oak (*Quercus nuttallii* Palm.) and cherrybark oak (*Q. pagoda* Raf.). Seed was collected, germinated, and transplanted into thirty by twenty-two centimeter plastic nursery pots containing a locally-collected silt loam soil. Flooding treatments consisted of an intermittent flooding treatment of five days flooded followed by five days of no flooding and a non-flooded control treatment. Data collection included stomatal conductance, net photosynthetic rate, and leaf, root, and stem biomass. Data were recorded at the initiation of the study and following the end of each flooding and dry period.

Pezeshki and others (1999) found flooding had a negative effect on all variables for seedlings of both oak species for the flooded seedlings when compared to the non-flooded control seedlings. Reduced stomatal conductance and net photosynthetic rates were observed for intermittently flooded oak seedlings. There were significant reductions in the leaf, root, and shoot biomass of seedlings flooded when compared to the non-flooded control treatment.

Colin-Belgrand and others (1991) investigated the responses of English oak (*Quercus robur*), northern red oak (*Q. rubra*), and pin oak (*Q. palustris*) to flooding. Acorns were collected, stored, stratified, and germinated. Individual seedlings were placed in five-liter, twenty-five-centimeter-deep plastic nursery pots containing a sandy loam soil. Flooding treatments consisted of saturating the soils to a pseudo water table of six centimeters below the soil line, and a non-flooded control. Soil saturation treatments lasted seven weeks followed by two weeks of post-flooding recovery. Responses to flooding were measured by biomass accumulation, water status, nutrient content, xylem sap composition, presence of hypertrophied lenticels, and degree of root senescence.

Colin-Belgrand and others (1991) results indicated that saturating the soils had a significant negative effect on the root morphology of the flooded seedlings when compared to the non-flooded control treatments. There was necrosis of the tap and lateral roots and formation of hypertrophied lenticels on the submerged portion of the seedlings. Soil flooding did not have a significant effect on shoot growth and leaf morphology. Minor changes in micro- and macro-nutrient content was observed in flooded seedlings.

Anella and Whitlow (1999) investigated photosynthetic responses of red maple (*Acer rubrum* L.) seedlings grown from seed sources from wet (bottomland) and dry (upland) sites. Seeds were collected from three upland and three bottomland sites, grown in plastic nursery pots and eventually transplanted into large tubs using the same greenhouse mixture through the entire study. Flooding treatments were administered in the spring and repeated with new seedlings in the summer. Flooding treatments consisted of inundating the seedlings to a depth of five to eight centimeters above the soil line for a period of twenty two days and a non flooded control. Flooding responses were measured by net photosynthesis, relative chlorophyll content of the leaves, leaf areas, and shoot and root dry weights.

Anella and Whitlow (1999) results showed flooding reduced net photosynthesis, growth, and chlorophyll content in seedlings from both hydrologic positions. Seedlings from bottomland seed sources had higher photosynthetic rates when compared to seedlings from upland seed sources across all treatments. Control seedlings of bottomland origin were significantly larger and had higher photosynthetic rates and chlorophyll content than seedlings of upland origin. Although there were significant differences among seedlings collected from bottomland sites when compared to seedlings collected from upland sites, the flooding treatments caused a near cessation of growth. The authors concluded that there was a habitat (topographic position) by treatment interaction occurring in their study and with the selected families of seedlings obtained from contrasting topographic regimes.

Bauerle and others (2003) also investigated ecophysiology of red maple seedlings from contrasting hydrologic habitats: dry (upland) and wet (bottomland) sites. Seeds were collected, germinated and allowed to grow for one growing season in a greenhouse potting mix in individual plastic nursery pots under greenhouse conditions. Seedlings were carried over through winter in large walk-in coolers. One-year-old seedlings were subject to a well-watered control treatment and a drought treatment. Watering consisted of filling the nursery pots to field capacity. Data collection measured gas exchange, abscisic acid concentration, stable carbon isotope determination, and growth in term of height and diameter of seedlings.

Bauerle and others (2003) found that wet-site seed sources grown under well-watered conditions grew faster and had consistently higher net photosynthesis, leaf conductance, maximum carboxylation rate, and other gas exchange variables when compared to dry-site seed sources. Under conditions of low soil-water availability, only dry-site seed sources responded with decreased osmotic potential. Wet-site seed sources responded with a reduction in absolute growth rate, gas exchange, and a greater increase in abscisic acid concentrations than dry-site seed sources. Their results indicated that red maple has two physiologically distinct ecotypes that differ in their adaptation to soil water availability.

Keeley (1979) investigated population differentiation along a flood frequency gradient for blackgum (*Nyssa sylvatica* Marsh.) seedlings. Seeds were collected from three contrasting seed source site classes: Upland seed sources were from well-drained sites in the mountains of northern Georgia, floodplain seed sources were from alluvial sites in the piedmont, and swamp seed sources were from more or less waterlogged sites. Each geographic locale had at least two populations and each population had a total of two to fifteen trees. Seeds were cold stratified for six months in order to meet germination requirements of blackgum seeds from contrasting elevations. Seeds were removed from cold storage, germinated in fifteen-centimeter-tall clay pots, and grown under greenhouse conditions. Treatments were carried out in 32-cm-tall tubs inside of the greenhouse. Treatments consisted of partial inundation for sixteen months at approximately five centimeters above the soil line and a non-flooded control treatment. Data collection measured root nutrient content, aerobic respiration, alcohol fermentation, ethanol production, alcohol dehydrogenase (ADH) activity, malic acid concentration, and rates of oxygen transport.

Keeley (1979) found that seedlings from upland seed sources had lower survival in the flooded treatment (27%) when compared to the non-flooded control treatment (73%). In contrast, seedlings from swamp bottomland seed sources had lower survival in the non-flooded control treatment (65%) when compared to the flooded treatment (98%). Seedlings from floodplain bottomland seed sources had no difference in survival in the non-flooded control treatment (100%) when compared to the flooded treatment (94%). In terms of biomass accumulation, seedlings from upland seed sources had 2/3 lower biomass in the flooded treatment (4.11g) when compared to the non-flooded control treatment (10.84g). Seedlings from swamp bottomland and floodplain bottomland seed sources had higher biomass accumulations when compared to the seedlings from upland seed sources across both treatments. Seedlings from swamp bottomland seed sources had greater biomass accumulation than seedlings from all other seed sources in both the non-flooded control treatment and flooded treatment.

Nielsen and Jorgensen (2003) investigated phenology and diameter increment in seedlings of European beech (*Fagus sylvatica* L.) under contrasting flooding regimes. The entire study was carried out inside a greenhouse in large containers containing a greenhouse potting mix. Seeds of fourteen beech seed sources were collected, stratified, germinated and seedlings transplanted in rows inside each of the containers. Soil water treatments consisted of maintaining soil water at thirty-eight, fifty-five, and eighty percent of field capacity. Data collections included root collar diameter at three centimeters above the soil line, height, and leaf flushing characteristics measured periodically throughout the growing season.

Nielsen and Jorgensen (2003) found an interaction for seedling growth responses between provenances of origin and the soil water treatments. Root collar diameter of seedlings and overall heights were smaller in the low soil water treatments when compared to the seedlings in the medium and high soil water treatments.

Upon examination of the results of replicated pot studies, replicated field studies, and case studies, there are clear positives and negatives associated with all methods. The use of a replicated pot study provides for the best control over many factors. Replicated pot studies may not mimic natural conditions as well as replicated field studies and case studies because replicated pot studies typically use better-drained soils than found in the latter. Replicated pot studies, however can focus directly on species responses to flooding without the concern over extreme and/or variable environmental factors, such as temperature, precipitation, and degree of flooding (depth and duration). Uncontrollable natural events, such as extreme weather, may not be fully controlled but are more likely to be mitigated to the point where these events do not confound the results.

These environmental factors, though naturally occurring, may have contributed to confounding results if they are not taken into account during the analysis stage.

Use of Shade Structures

The use of shade to reduce sunlight and the resultant soil warming of potted nursery crops is a time-honored cultural practice. Landis (2005) reviewed the use of shade structures and reports that shade cloth can be used to both shade and protect seedlings by reducing the amount of incoming solar radiation. Studies with pots in large tanks under full sunlight have shown the system (flooding facilities) tends to absorb substantial solar radiation and results in undesirable water heating. Coggeshall (unpublished) observed substantial mortality on potted oak seedlings partially inundated in stock tanks after only five weeks with little or no mortality of seedlings of the same families within the FTL. For this reason it was deemed optimal to implement the use of a shade structure in my study to achieve more natural floodwater temperatures by reducing the amount of solar radiation. As most oak seedlings reach their light compensation point for photosynthesis at or below fifty percent of full sunlight, potted plant studies can be done under shade (Kramer and Kozlowski 1979).

Flood Tolerance Variation among Oaks

A number of oak species have been evaluated using multiple procedures and given qualitative descriptions in response to flooding (Table 1). The information is compiled from several authors involving flood tolerance research of oak seedlings and/or mature trees. Bell and Johnson (1974) and Whitlow and

Harris (1979) used mature oak trees to differentiate their flood tolerance ratings. Hook (1984) utilized the results of physiological and empirical field studies and field observations to classify flood tolerance ratings. Kabrick and Dey (2001) used seedlings of several oak species while it is unclear how Keeland and others (2001) distinguished their flood tolerance ratings.

Swamp white, bur and other oaks will grow on both poorly drained, wet bottomland soils and on dryer soils typical of upland ridges (Settergren and McDermott 2000). Bur oak (*Q. macrocarpa*) exhibits wide ecological amplitude and occurs on dry rocky cliffs, hill crests, glacial moraines, mesic uplands, and moist bottomlands (Tang and Kozlowski 1982). Swamp white oak (*Q. bicolor*) occurs in bottomland forests in valleys and on rich, lower slopes, in wet-ground bordering streams, and oxbow lakes of floodplain and stream meanders, and along streams and can withstand drought conditions once it is established (Kurz 2003).

White oak is not commonly found on bottomland sites and typically cannot withstand inundation by floodwaters. Several species, such as white and black oak, are seldom found on sites subjected to periodic flooding.

Past studies reveal that oak species naturally occupying predominantly upland sites will succumb to flood induced mortality before oak species found on bottomland sites (Bell and Johnson 1974). For the species growing on both upland and bottomland sites, it is unknown how much the intraspecific genetic variation for flood tolerance exists in these species. Specifically, to what degree are seedlings grown from seeds collected from upland sites maladapted to

Table 1: Flood tolerance ratings from 1 (intolerant) to 4 (tolerant) for 14 species of *Quercus*.

Species	Common Name	Bell & Johnson 1974[1]	Whitlow & Harris 1979[2]	Hook, 1984[3]	Kabrick & Dey 2001[4]	Keeland & others 2001[5]
Quercus bicolor	Swamp white oak	4	2	---	3	3
Q. alba	White oak	2	1	1	1	1-2
Q. macrocarpa	Bur oak	4	4	---	3	1
Q. velutina	Black oak	1	1	---	---	---
Q. palustris	Pin oak	4	3	3	2	3
Q. imbricaria	Shingle oak	3	3	---	---	---
Q. pagoda	Cherrybark oak	---	---	2	---	1-2
Q. rubra	Northern red oak	2	1	---	---	---
Q. lyrata	Overcup oak	---	4	4	3	3
Q. michauxii	Sw chestnut oak	---	---	2	1	2
Q. phellos	Willow oak	---	2	3	3	2-3
Q. shumardii	Shumard oak	---	1	2	2	2
Q. muehlenbergii	Chinquapin oak	---	---	---	---	---
Q. nigra	Water oak	---	1	---	3	2-3

[1] Expected flood tolerances where 1 = intolerant species with severe effects in less than 50 days of flooding; 2 = slightly tolerant species where most individuals survive greater than 50 days of flooding but less than 100 days of flooding; 3 = somewhat tolerant species where some individuals are killed by less than 90 days of flooding and some individuals survived greater than 150 days of flooding; and 4 = tolerant species where most individuals survived greater 150 days of inundation (Bell and Johnson 1974).

[2] Expected flood tolerances where 1 = intolerant species that are unable to survive more than a few days of flooding during the growing season with significant mortality; 2 = somewhat tolerant species that are able to survive flooding or saturated soils for 30 consecutive days during the growing season; 3 = tolerant species that are able to survive deep flooding for one growing season, with significant mortality occurring if flooding is repeated the following year; and 4 = very tolerant species that are able to survive deep, prolonged flooding for more than 1 year (Whitlow and Harris 1979).

[3] Expected flood tolerances where 1 = least tolerant species that are capable of living from seedling through maturity in soils that are occasionally flooded for a few days only; 2 = weakly tolerant species that are capable of living from seedling to maturity in soils that are temporarily flooded for durations of 1 to 4 weeks; 3 = moderately tolerant species that are capable of living from seedling to maturity in soils flooded 50 % of the time; and 4 = highly tolerant species that are capable of living from seedling to maturity in soils flooded about 50 % of the time (Hook 1984).

Continuation of footnotes for Table 1:

[4] Expected flood tolerance ratings where 1 = intolerant species that cannot withstand growing season flooding; 2 = moderately tolerant species that can survive growing season flooding lasting 1 to 3 months; 3 = tolerant species that can withstand flooding for more than 1 growing season; and 4 = very tolerant species that can withstand flooding for periods of two or more growing seasons (Kabrick and Dey 2001).

[5] Expected flood tolerance ratings where 1 = intolerant species that are not able to survive even short periods of soil saturation or flooding during the growing season; 2 = weakly tolerant species that are able to survive saturated or flooded soils for relatively short periods of a few days to a few weeks during the growing season; 3 = moderately tolerant species that are able to survive saturated or flooded soils for several months during the growing season, but mortality is high if flooding persists or reoccurs for several consecutive years; and 4 = tolerant species that are able to survive and grow on sites where soil is saturated or flooded for long periods during the growing season (Keeland and others 2001).

outplanting on bottomland sites that are prone to flooding? Seeds collected by the public for sowing in tree nurseries probably originate from upland sites for several reasons revolving around the ease of collection. Upland sites are usually more accessible and more likely to be open grown, highly precocious trees in open park-like settings.

Genetic Variation in Flood Tolerance

Little published information exists regarding the intraspecific and interspecific genetic variation in terms of flood tolerance in Quercus species. Especially important is a lack of information on intraspecific variation for flood tolerance associated with seed sources originating from contrasting topographic positions for *Quercus* species. Flood tolerance of oak seedlings may vary not only between and among contrasting topographic positions, but also between and among individual families (seed sources).

Some scientific research has been conducted on the subject of genetic variation of tree seedlings grown from seed sources originating from different topographic positions. Anella and Whitlow (1999) found ecotypic differentiation with wet-site (bottomland) more flood tolerant than dry-site (upland) populations of red maple near Ithaca, New York. Bauerle and others (2003) also report that red maple has two physiologically distinct ecotypes that differ in their adaptation to soil water availability. Keeley (1979) found that blackgum seedlings collected from upland seed sources were very intolerant of flooded soils; in contrast, seedlings from bottomland seed sources were quite tolerant of flooded soils.

This past research indicates seedlings from upland seed sources will respond to periods of inundation by flooding differently than seedlings of bottomland origin.

Genetic variation is expected in an oak seedlings ability to tolerate inundation by floodwater. This variation may occur between individual families or seed sources independent of any moisture or stress gradient. Coggeshall and others (2007) suggested that within the oak species, there is a strong opportunity to make significant genetic gains in field survival through selection of more flood tolerant seedling families. This is further supported by Kabrick and others (2007) who suggested variation among seed sources for flood tolerance.

With the extent of genetic variation that may be present in the flood tolerance of oaks between and among families and contrasting topographic positions, additional research is required. This flood tolerance research may serve to benefit research foresters and land managers in attaining their management objectives.

CHAPTER 3

MATERIALS AND METHODS

Acorn Collection

Acorns were collected under 24 individual mother trees (seed source) between 22 September 2005 and on 14 November 2005 (Table 2). There was no evidence to support that any of the study trees were planted trees. Acorn collections were divided into two topographic positions, denoted as upland or bottomland, based on the occurrence any known flooding or ponding of water on the site in modern time. Bottomland sites were locations that currently experience some form of flooding, either short term (local creek floodplain) or long term (Missouri River Floodplain). Data collected at the time of collection from the parent tree included species, diameter at breast height (DBH), topographic position, and geographic location. Acorns showing signs of decay, acorn weevil damage, or presence of non-abscised acorn caps were not collected. Information for each site was taken from the cares.missouri.edu website. Percent slope, flooding frequency, soil series, drainage class and refined longitude and latitudes were reported directly from tools available on the interactive site using the map room and were used to complete tables 2 and 3. Families (seed sources) were denoted by a special code, e.g. BN21B; the first two letters denote county where collected (Boone), followed by stand number (2), then tree number (1), and finally by species (B).

Table 2: Geographic location, DBH, and collection date for the mother trees of all 24 families used in this study.

Family	Species	County	DBH	Collection Date	Latitude	Longitude
BN12S	SWO	Boone	91cm	11 Oct 2005	38°58’05”N	92°13’48”W
BN13S	SWO	Boone	81cm	11 Oct 2005	38°58’05”N	92°13’48”W
BN22S	SWO	Boone	43cm	13 Oct 2005	38°53’56”N	92°20’31”W
AD12S	SWO	Adair	56cm	10 Oct 2005	40°10’20”N	92°34’35”W
AD14S	SWO	Adair	58cm	10 Oct 2005	40°10’20”N	92°34’35”W
PK11S	SWO	Pike	122cm	1 Oct 2005	39°31’35”N	91°11’35”W
PK21S	SWO	Pike	53cm	1 Oct 2005	39°22’19’N	91°9’59”W
CA21S	SWO	Callaway	76cm	14 Oct 2005	30°2’39”N	91°53’43”W
CA22S	SWO	Callaway	81cm	14 Oct 2005	30°2’39”N	91°53’43”W
CO11S	SWO	Cole	137cm	13 Oct 2005	38°29’45”N	92°0’44”W
AD11B	BRO	Adair	76cm	10 Oct 2005	40°10’20”N	92°34’35”W
BN11B	BRO	Boone	36cm	23 Sep 2005	38°56’44”N	92°21’46”W
BN12B	BRO	Boone	66cm	23 Sep 2005	38°56’44”N	92°21’46”W
SH11B	BRO	Shannon	69cm	14 Nov 2005	37°26’02”N	91°35’51”W
CA11B	BRO	Callaway	102cm	27 Oct 2005	38°55’49”N	92°0’35”W
HW11B	BRO	Howard	152cm	29 Sep 2005	39°0’31”N	92°42’06”W
BN31B	BRO	Boone	165cm	30 Sep 2005	38°55’52”N	92°18’46”W
BN21B	BRO	Boone	234cm	28 Oct 2005	38°53’57”N	92°27’53”W
BN12W	WHO	Boone	79cm	22 Sep 2005	38°55’02”N	92°20’01”W
BN21W	WHO	Boone	61cm	30 Sep 2005	38°55’48”N	92°19’15”W
BN23W	WHO	Boone	122cm	30 Sep 2005	38°54’26”N	92°17’48”W
CO11W	WHO	Cole	102cm	4 Oct 2005	38°29’45”N	92°0’44”W
CO12W	WHO	Cole	114cm	4 Oct 2005	38°29’45”N	92°0’44”W
OS12W	WHO	Osage	46cm	3 Oct 2005	38°27’15”N	92°0’35”W

Table 3: Description of topographic site conditions for the mother trees of all 24 families used in this study.

Family	Topographic Position	Slope Position	% Slope	Drain Class[1]	Flooding Frequency[2]	Soil Series[3]
BN12S	Bottomland	Floodplain	0-2	VPD	Frequently	Perche L
BN13S	Bottomland	Floodplain	0-2	VPD	Frequently	Perche L
BN22S	Upland	Summit	2-9	MWD	None	Weller
AD12S	Upland	Shoulder	9-14	MWD	None	Gara L
AD14S	Upland	Shoulder	9-14	MWD	None	Gara L
PK11S	Bottomland	Terrace	0-3	VPD	Rarely*	Okaw SL
PK21S	Bottomland	Floodplain	0-2	VPD	Occasionally	Twomile SL
CA21S	Upland	Summit	1-5	SPD	None	Mexico SL
CA22S	Upland	Summit	1-5	SPD	None	Mexico SL
CO11S	Bottomland	Floodplain	0-2	VPD	Occasionally*	Moniteau SL
AD11B	Upland	Shoulder	9-14	MWD	None	Gara L
BN11B	Upland	Shoulder	2-9	WD	None	Weller
BN12B	Upland	Shoulder	2-9	WD	None	Weller
SH11B	Upland	Footslope	5-9	WD	None	Viration GSL
CA11B	Bottomland	Floodplain	0-2	SPD	Frequently	Belknap SL
HW11B	Bottomland	Floodplain	0-2	SPD	Occasionally*	Hayne SL
BN31B	Bottomland	Floodplain	0-3	MWD	Frequently	Haymond SL
BN21B	Bottomland	Floodplain	0-2	VPD	Occasionally*	Darwin Silty CL
BN12W	Upland	Summit	5-9	MWD	None	Keswick-Urban Land Complex
BN21W	Upland	Shoulder	0-3	WD	None	Jamerson SL
BN23W	Upland	Summit	5-9	SPD	None	Armstrong L
CO11W	Bottomland	Floodplain	0-2	VPD	Occasionally*	Moniteau SL
CO12W	Bottomland	Floodplain	0-2	VPD	Occasionally*	Moniteau SL
OS12W	Upland	Summit	3-8	MWD	None	Gravois-Gatewood Complex

[1] Drainage class abbreviations: VPD = very poorly drained, SPD = somewhat poorly drained, MWD = moderately well drained, WD = well drained

[2] An asterisk denotes the tree was flooding in 1993 and/or 1995

[3] Soil abbreviations: L = loam, S = silt, C = clay, G = Gravelly.

Acorn Storage

Once the collections were completed, acorns were stored in plastic zipper bags with holes punched for aeration. Acorns were stored on open shelves at 4º C in the University of Missouri Department of Forestry's walk-in cooler (Room 45) in the Anheuser Busch Natural Resources Building in Columbia, MO. The acorns remained in this location until 15 November 2005 when they were transported to the walk-in coolers at the University of Missouri Horticulture and Agroforestry Research Center (HARC) in New Franklin, MO. Storage conditions at both locations were a constant 4º C. Acorns of white oak families were direct seeded on 28 November 2005 and stored on germination trays because of their fall germination.

Acorn Preparation

Acorns were soaked for 48 hours beginning on 28 November 2005 in large stock tanks at HARC to ensure adequate moisture content in the seeds. Once the soaking procedure was completed, the acorns were float-tested by family in the stock tanks. Each seedlot was placed free of bags into the stock tanks and all acorns that floated on the surface or suspended near the surface were removed and discarded. Water was removed after each family was float-tested and the acorns that sank were returned to cold storage in plastic zipper bags. For each family, 25 acorns were randomly chosen cut in half to detect the presence of insects and/or decay to determine the number of acorns to germinate.

Acorn Germination

Seedlings of all species were germinated and grown using a variant of the Root Production Method© developed by Wayne Lovelace of Forrest Keeling Nursery in Elsberry, MO (Lovelace, 1998). *Q. alba* acorns germinate in the fall and require different methods for propagation than *Q. macrocarpa* and *Q. bicolor.* All potting media consisted of a pine bark, peat, perlite, vermiculite, and sand mix in a 15:2:5:2:2 vol/vol ratio, supplemented with 0.15% slow release 38-0-0 NPK, 0.2% slow release 13-13-13 NPK, and 0.1% Scott's® micronutrients (V/V).

White Oak Acorn Germination

After the soaking procedure, only white oak acorns with emerging radicals were chosen for sowing on 28 November 2005. Acorns were sown with radicals placed in holes created with a pen in a 5 to 8 cm deep layer of potting medium within mesh bottom 40 X 40 X 12.7 cm propagation flats (Anderson Die and Manufacturing Co., Portland, OR). Acorns were sown at densities of 150 to 200 acorns per tray. The mesh bottom seed trays were chosen in order to air-prune the taproots of the germinants. Loose potting medium was spread over the acorns and the trays were saturated thoroughly. After sowing, trays were put into individual polyurethane bags and returned to cold storage at 4° C. On 14 February 2006 acorns were removed from cold storage and placed in a greenhouse at 23º C. Propagation flats were saturated and watering was carried out as needed 2 to 3 times weekly. Seedlings were allowed to grow in propagation flats until all seedlings had attained at least one complete flush.

Bur and Swamp White Oak Acorn Germination

Acorns were removed from cold storage on 16 February 2006 and sown the same day. Acorns were placed in Anderson open-mesh trays on top of 5 to 8 cm of potting medium. Loose media was placed over the acorns and saturated thoroughly. Watering was continued as needed 2 to 3 times per week. Planting densities in seed trays varied due to the differences in acorn sizes both within species and family. Bur oak densities ranged from 50 to 100 acorns per tray and swamp white oak densities ranged from 100 to 150 acorns per tray. Trays were randomly placed on movable open mesh benches in the greenhouse together with white oak seed trays at 23º C. Seedlings were allowed to grow in propagation flats until all seedlings had attained at least one complete flush.

Transplanting Seedlings

One-flush seedlings were transplanted over a two-week period beginning on 20 March 2006 to individual pots. Approximately seventy of the largest, single-stemmed seedlings were chosen from the propagation trays of each family for transplanting. Seedlings were transplanted into 23-cm-tall 1.65 L “short one” tree pots (Stuewe and Sons Inc., Corvallis, Oregon). Each tree was placed into its own container and the same potting medium was used to fill and encase the roots of the seedlings. The same soil line on the stems was used as the soil line in transplanted stock. Soil lines were apparent by the pink color tissues under the soil line. Generally the soil lines were 2.5 cm below the top of the pot to facilitate future diameter measurements. Soil was compacted before and after the placement of seedlings into pots. The seedlings were watered before and after transplanting and watered 2 to 3 times per week or as needed thereafter. All seedlings were moved back into the greenhouse in 8 cm by 8 cm spacing on the open mesh movable benches and allowed to resume growth. Seedlings underwent labeling using a four-digit numbered plastic label. The labels were wrapped around the bases of seedlings in a manner that would not restrict diameter growth.

Seedlings were moved on 10 May 2006 to the outdoor shade structure covered with a 50% shade cloth in order to adapt to ambient relative humidity, temperature, and wind. During this period, plants were irrigated daily each morning using an automatic overhead impact drive rotary sprinkler heads. Irrigation water was pumped from a rainwater catchment pond located at HARC. Seedlings were sorted by species and family. The 36 largest seedlings were selected from each family with 3 seedlings randomly distributed to the 12 stock tanks used in the experiment. Three seedlings of each family were chosen for each stock tank. Of the remaining seedlings, the largest five seedlings of each seed source were selected for destructive sampling.

Flooding Design

The 12 galvanized 1,126 L stock tanks used in the study were 61 cm in height, 92 cm in width, and 244 cm in length. Tanks were set up with four replications each having three treatments (Figure 1). The control, 4-week flood, and 8-week flood treatments were spatially arranged in a Latin-square pattern across the four replications. An assemble built from plastic seed trays and plastic pots was used to raise the seedlings to a level within each tank where the tops of the seedling containers were at least 7 to 10 cm below the top of the tank. Seedlings for a species were placed in stock tanks at random within each species. Special attention was focused to ensure the white oak seedlings were placed on either end of the stock tank to prevent excess shading by the, taller seedlings of swamp white oak and bur oak. Stock tanks were placed inside the outdoor shade structure on landscape fabric over a bed of gravel. A 50% shade cloth was suspended over the experiment area to alleviate solar radiation inputs to the tanks. Past studies indicated that stock tanks in direct sunlight may have resulted in elevated water temperatures and increased mortality of normally-flood tolerant seedlings (Coggeshall, personal communication).

Flooding Treatment

Flooding treatments consisted of an 8-week flood, a 4-week flood, and a non-flooded control. Flooding began on 29 May 2006 in tanks numbered 1 through 8 with tanks numbered 9 through 12 designated as controls (figure 2).

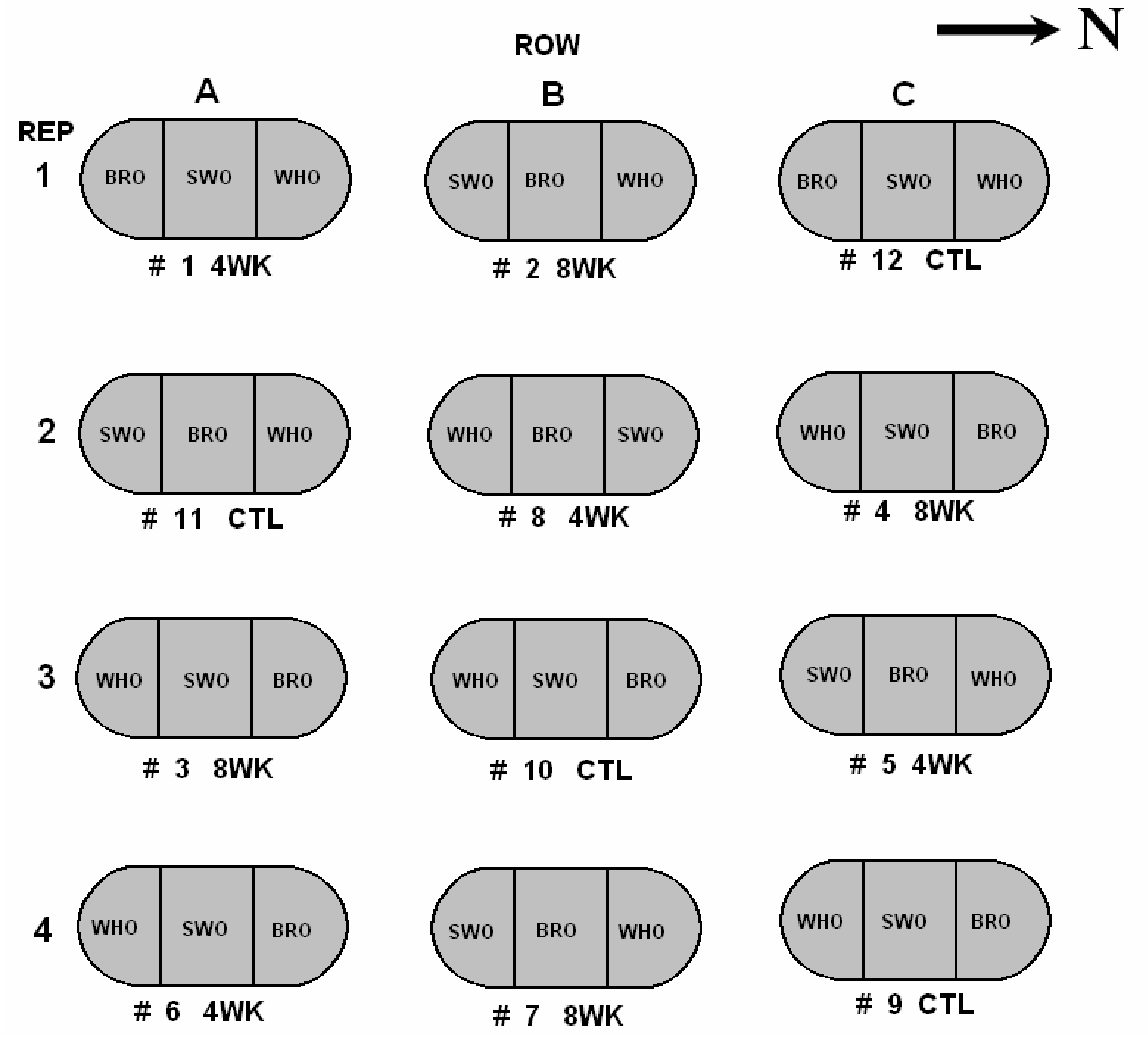

Figure 1: Latin-square arrangement of three flood treatments within four replications of stock tanks.

15 September 2005- 31 October 2005	Acorn collecting from 24 families of oak
↓↓↓	---
28 November 2005- 14 February 2006	Acorn stratifying at 4º C in walk-in coolers
↓↓↓	---
15 February 2006- 20 March 2006	Producing 1-flush seedlings in climate-controlled greenhouse
↓↓↓	---
20 March 2006- 10 May 2006	Transplanting seedlings into individual nursery pots
↓↓↓	---
10 May 2006- 29 May 2006	Acclimating seedlings to outdoor-growing conditions
↓↓↓	---
29 May 2006	Initiating all flooding treatments with data collection
↓↓↓	---
26 June 2006	Ending 4-week flooding treatment with data collection
↓↓↓	---
24 July 2006	Ending 8-week flooding treatment with data collection
↓↓↓	---
21 August 2006	Ending post-flood recovery period with data collection
↓↓↓	---
2 October 2006	All seedlings destructively sampled with data collection

Figure 2: Timeline demonstrating tasks completed beginning on 15 September 2005 and ending on 2 October 2006.

Flooding for the four-week-flood treatment ended for tanks numbered 1, 5, 6, and 8 on 26 June 2006. Flooding for the eight-week flood treatment ended for tanks numbered 2, 3, 4, and 7 on 24 July 2006. When not flooded, seedlings were manually irrigated with pond water 2 to 3 times per week or as needed depending on climatic conditions (Figures 3 and 4). Daily weather observations were taken from the HARC NOAA weather station number 23-6012-1, New Franklin, Missouri. Water in the flooded tanks was replenished manually with pond water during all watering sessions and seedlings remained partially inundated throughout the flooding process. In order to facilitate data collections, after all flooding treatments had ended; seedlings were removed from stock tanks and arranged in the same spatial orientation on top of the landscape fabric until the onset of destructive sampling. Seedlings were manually irrigated with pond water 2 to 3 times per week or as needed.

Data Collection

Data collection occurred on 27 May 2006, on 26 June 2006, on 24 July 2006, and on 21 August 2006 and included survival, flush stage, basal diameter, leaves per flush, total stem length, presence of lenticels, and presence of basal sprouting. Flush stages according to a modified version of Hanson and others (1986) *Quercus* Morphological Index were collected from each seedling. Seedlings at each measurement date were assigned a whole number corresponding to their number of completed flushes. A 0.3 was added to the

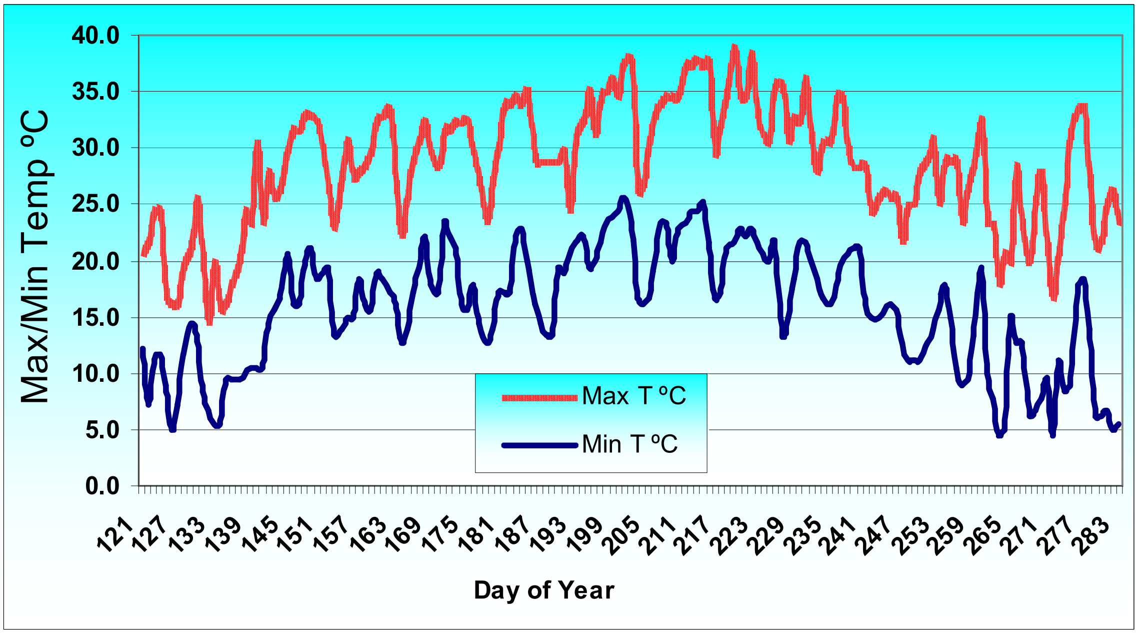

Figure 3: Minimum and maximum recorded temperatures for the duration of the experiment that occurred outdoors.

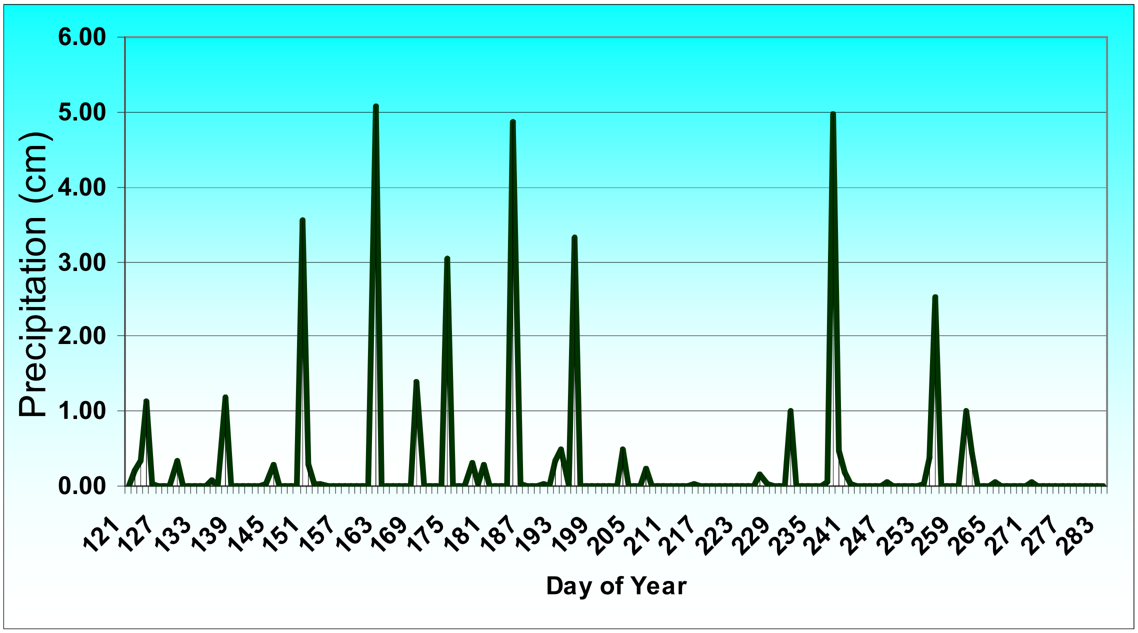

Figure 4: Daily precipitation for the duration of the experiment that occurred outdoors.

number of completed flushes if a new flush had begun and was in the process of stem elongation. A 0.6 was added if the newest flush had completed stem elongation and growth was focused on leaf elongation. Only when all stem and leaf elongation had been completed was a whole number for the number of completed flushes given. A seedling that had completed its first three flushes and had just begun its fourth flush would be assigned a 3.3. Total stem height was determined during each data collection. Measurements were taken from soil line to tip of bud or elongating shoot to the nearest 0.5 cm. Stem diameters were taken at approximately 2.5 cm above the soil line. Measurements were to the nearest 0.1 cm using a Model SC-6 digital caliper (Mitutoyo© Corporation, Aurora, Ill). The numbers of leaves per flush were counted and including any new emerging leaves on new flushes were also counted as they became visible. Any mortality of seedlings was documented at each measurement date as were the presence of lenticels on the stem and basal sprouting. Basal sprouting consisted of all emerging new shoots below level of the floodwater.

Destructive sampling of the seedlings occurred 84 days following the onset of flooding in the flood treatments. All seedlings of all species and treatments were removed from containers and all loose potting medium was shaken off the roots. Due to the fibrous nature of most of the roots and their tendency to retain potting medium, all roots were thoroughly rinsed with pond water until nearly all debris was removed. Seedlings were individually placed in brown paper bags and returned to cold storage at the Forestry Department and/or HARC cold storage units.

Over the next month all seedlings were measured and destructively sampled. Basal diameters were measured at approximately 2.5 cm above the soil line. The stem was then separated from the roots at the root collar. Root collar diameters were attained by visually locating the average diameter and recording it to the nearest tenth cm using the same digital caliper. The distance from the root collar to the location of the basal diameter measurement was recorded to the 0.5 cm.

Leaves per flush were counted and leaf areas per flush were measured to the nearest cm^2 using an LI-3000 portable area meter attached to a belt driven stand (Li-Cor© Inc., Lincoln, NE). Leaves were then stacked by flush and held together by three standard staples and flush number was written on the leaves with a permanent marker. Stem length of each flush was measured to the nearest 0.5 cm. Total stem length was measured to the nearest 0.5 cm. If basal sprouting was present the total number and cumulative length of the sprouts were measured to the nearest 0.5 cm. Leaves on basal sprouts were collectively counted and leaf areas were measured to the nearest cm^2. If lateral branching was present the total number and cumulative length of the branches were measured to the nearest 0.5 cm. Leaves on lateral branches were collectively counted and measured to the nearest cm^2. Root length was measured from the root collar to the tip of the longest taproot or longest first order lateral root (FOLR) to the nearest whole cm. Roots were stretched to attain each length. Root fibrosity was measured on the following scale: 1= the least fibrous and most likely dead, 2= having few if any FOLR's fine roots, 3= having moderate numbers of

FOLR's and fine roots, 4= having many FOLR's and fine roots, and 5= having numerous FOLR's and fine roots.

Seedlings were returned to their individual paper bags before placing in a Model 600 constant temperature oven (Yamato© Inc., Tokyo, Japan) at 60º C for at least 72 hours. Some seedlings were removed after the 72 hours weighed, returned to the oven for an additional 24 hours and weighed once again. On all re-weighed seedlings the difference in dry weights was less the 1 % of the total weight, thus the 72 hour time period was deemed adequate. Leaves were weighed by flush using a Model TSXB620M digital scale (Thomas Scientific© Inc., Swedesboro, NJ) to the nearest 0.01 gram. The weight of the three standard staples (0.096 g) was rounded to 0.1 g and subtracted from the dry weight of each leaf bundle per flush. Stem weight per flush was measured to the nearest hundredth gram. Total stem weight and root weight was measured and recorded to the nearest hundredth gram. Basal sprouting and lateral branching were handled as separate flushes. Dry weights of stems were collectively recorded to the 0.01 g. Dry weights of leaves were collectively recorded to the 0.01 g. Seedlings sampled prior to flooding underwent the same data collection procedures.

Data Analysis

Data were checked for missing entries and keyboarding errors using SAS proc univariate. Treatment means for each family within each stock tank were created in SAS. When data were normally distributed, family means by species and flood treatment were analyzed for statistical differences using an ANOVA with a split-split plot design (Table 4). Main effects were tested for significance at $\alpha = 0.05$ using Duncan's new multiple range test for mean separations. Interactions were tested for significance at $\alpha = 0.01$ using Fisher's unprotected least significant difference for mean separations. When data were not normally distributed, and all classes exceeded 10% of total (Snedecor and Cochran 1967), Chi-square analysis was used to analyze the count and percentage data.

Table 4: ANOVA Table for a split-split plot design used to analyze the effects of three flooding regimes on three species of oak seedlings averaging 4 half-sib families per species within two topographic positions.

	Source	Df
MAIN PLOT	Block	3
	Flood	2
	Block X Flood = **(ERROR A)**	6
SUB PLOT	Species	2
	Flood X Species	4
	Block X Species (Flood) = **(ERROR B)**	18
SUB SUB PLOT	Position (Species)	3
	Flood X Position (Species)	6
	Family (Species X Position)	18
	Flood X Family (Species X Position)	36
	Residual = **(ERROR C)**	189
	Total	287

CHAPTER 4

RESULTS

Pre-Flood Analysis

In order to identify differences in the study, basal diameters, heights above the root collar, and total number of leaves were examined at the time container-grown seedlings were placed in the stock tanks for flooding. Seedlings at this time had grown to two-flush seedlings from acorns removed from stratification 103 days earlier.

Seed source differences existed between upland and bottomland seed sources for all three species for one or more of the variables aforementioned (Table 5). There were significant differences in pre-flood mean family heights in all three species sorted by topographic position of the seed source. With the exception of upland seed sources of swamp white oaks, all species sorted by topographic position had significant differences in pre-flood basal diameters and number of leaves. Seedlings of bur oak families from upland and bottomland seed sources had the highest variation in average height recorded at the initiation of flood treatments. Seedlings of bur oak families from upland and bottomland seed sources and seedlings of swamp white oak families from bottomland seed sources had the highest variation in basal diameter recorded at the initiation of the flood treatments. In contrast, seedlings of swamp

Table 5: Pre-flood average height above root collar, basal diameter, and total number of leaves by families for species and position from contrasting topographic positions.

			Dependent Variable[3]		
Family	Species[1]	Position[2]	Height (cm)	Diameter (mm)	Leaves (no.)
BN21B	BRO	BOT	44.5 ± 3.2	5.2 ± 0.6	11.9 ± 1.5
BN31B	BRO	BOT	40.4 ± 1.8	5.0 ± 0.5	12.9 ± 2.7
CA11B	BRO	BOT	35.5 ± 4.3	4.4 ± 0.6	9.8 ± 1.3
HW11B	BRO	BOT	35.8 ± 3.1	4.7 ± 0.6	11.6 ± 1.4
AD11B	BRO	TOP	35.3 ± 2.6	4.5 ± 0.3	10.5 ± 1.2
BN11B	BRO	TOP	42.8 ± 2.9	5.2 ± 0.5	11.6 ± 1.3
BN12B	BRO	TOP	39.3 ± 3.9	4.7 ± 0.5	10.6 ± 1.5
SH11B	BRO	TOP	26.0 ± 2.6	3.9 ± 0.3	9.3 ± 1.5
BN12S	SWO	BOT	33.7 ± 1.8	4.7 ± 0.4	11.6 ± 1.6
BN13S	SWO	BOT	33.0 ± 2.3	4.8 ± 0.4	10.6 ± 1.0
CO11S	SWO	BOT	33.8 ± 3.1	4.5 ± 0.5	10.7 ± 0.9
PK11S	SWO	BOT	25.2 ±1.8	3.7 ± 0.2	8.9 ± 1.0
PK21S	SWO	BOT	27.5± 2.2	4.1 ± 0.4	9.5 ± 0.8
AD12S	SWO	TOP	28.3 ± 1.2	4.3 ± 0.3	9.9 ± 0.7
AD14S	SWO	TOP	27.0 ± 1.0	4.2 ± 0.4	9.7 ± 0.7
BN22S	SWO	TOP	29.5 ± 2.5	4.5 ± 0.3	10.1 ± 1.4
CA21S	SWO	TOP	29.2 ± 3.1	4.4 ± 0.3	10.7 ± 1.5
CA22S	SWO	TOP	30.1 ± 2.5	4.4 ± 0.3	10.6 ± 0.9
CO11W	WHO	BOT	17.1 ± 3.0	2.9 ± 0.2	7.7 ± 1.0
CO12W	WHO	BOT	21.7 ± 1.9	3.4 ± 0.2	10.9 ± 1.6
BN12W	WHO	TOP	23.4 ± 1.9	3.4 ± 0.4	11.1 ± 1.1
BN21W	WHO	TOP	17.6 ± 1.7	3.2 ± 0.4	8.3 ± 1.6
BN23W	WHO	TOP	25.8 ± 2.5	3.8 ± 0.3	12.7 ± 1.9
OS12W	WHO	TOP	19.6 ± 2.3	3.3 ± 0.2	9.9 ± 1.7
Significance LSD $_{0.01}$ =			**2.7**	**0.3**	**1.4**

SOURCE	P > F	P > F	P > F
Flood	0.3252	0.0014	0.8690
Species	0.0001	0.0001	0.0008
Flood * Species	0.7011	0.0002	0.6389
Position (Species)	0.0001	0.0001	0.0001
Flood * Position (Species)	0.6828	0.0750	0.1365
Family (Position * Species)	0.0001	0.0001	0.0001
Flood * Family (Position * Species)	0.0397	0.5668	0.0189

[1] Species codes are BRO = bur oak, SWO = swamp white oak, WHO = white oak

[2] Position codes are TOP = upland seed sources, BOT = Bottomland seed sources

[3] Family means and standard deviations are an average of 36 seedlings

white oak families from upland seed sources had less variation in basal diameter than all species from all other positions. Seedlings of white oak families from upland seed sources had the highest amount of variation in the total number of leaves of all species at the initiation of flood treatments. Seedlings of swamp white oak families from upland seed sources had the least amount of variation in the total number of leaves at the initiation of flood treatments.

Additionally, seedlings of one bur oak seed source was taller than seedlings of all other bur oak families and a greater basal diameter than seedlings of five of the seven other bur oak families. This seed source, labeled family BN21B, from the Missouri State Champion Bur Oak located in McBaine, MO and is now the Co-National Champion Bur Oak as well.

There was a highly significant interaction between seedling heights above the root collar, basal diameter, and/or total number of leaves for bur oak, swamp white oak, and white oak between the seedlings from upland and bottomland seed sources (Table 6). Bur and swamp white oak seedlings from bottomland seed sources were taller than seedlings from upland seed sources. In contrast, white oak seedlings from upland seed sources were taller than seedlings from bottomland seed sources. Bur oak seedling basal diameters and total number of leaves from bottomland seed sources were significantly greater than for seedlings from upland seed sources. In contrast, white oak seedling basal diameters and total number of leaves from upland seed sources were significantly larger than seedlings from bottomland seed sources. Swamp white

Table 6: Pre-flood average height above root collar, basal diameter, and total leaves per seedling of bur, swamp white, and white oak species from upland and bottomland sites.

	Position		Significance
Response & Species	Upland	Bottomland	Pr > | t |
HEIGHT ABOVE ROOT COLLAR	-------centimeters----------		
Bur oak	35.8	39.0	0.0001
Swamp white oak	28.8	30.6	0.0001
White oak	21.6	19.4	0.0006
BASAL DIAMETER	--------millimeters----------		
Bur oak	4.6	4.8	0.0002
Swamp white oak	4.3	4.4	0.6423
White oak	3.4	3.1	0.0007
LEAVES PER SEEDLING	----------number------------		
Bur oak	10.5	11.6	0.0001
Swamp white oak	10.3	10.2	0.7752
White oak	10.5	9.3	0.0004

oak seedlings from bottomland seed sources had no significant difference in basal diameter and leaves per seedling when compared to swamp white oak seedlings from upland seed sources.

Although seedlings were randomly assigned to flood treatments prior to the onset of flooding, a significant interaction existed for basal diameters among flood treatments and species. Bur and swamp white oak seedlings that were assigned to the non-flooded control treatment had mean basal diameters of 5.1 and 4.6 mm, respectively. In contrast, bur and swamp white oak seedlings that were assigned to the two flood treatment had mean basal diameters of 4.5 and 4.2 mm, respectively. Mean basal diameters for white oak seedlings assigned to all three flood treatments was 3.3 mm.

Post-Flood Analysis

Survival

Family means for percent survival were not normally distributed and therefore could not be analyzed using the proposed analysis of variance model. This was in large part because few seedlings actually died. Seedlings that were labeled dead are those that showed either signs of epinasty and/or completely defoliated without any refoliating.

In the non-flooded control treatment, no seedling of any species died without eventual reflushing. Eleven seedlings died in the four-week flood treatment which included 7 white oak, 2 bur oak, and 2 swamp white oak seedlings. Eight of these seedlings from upland seed sources while three from bottomland seed sources. Twenty nine seedlings died in the eight-week flood treatment which included 24 white oak, 4 bur oak, and 1 swamp white oak seedling(s). Twenty two of these seedlings from upland seed sources while 7 from bottomland seed sources. Of the 25 white oak seedlings from upland seed sources that died in the four- and eight-week flood treatments, 16 were from 2 white oak families (OS12W and BN21W).

Only white oak seedling survival data for the eight-week flood could be tested with a Chi-square analysis for differences between topographic positions. Chi-Square analysis indicated that there were significant differences in white oak seedling mortality in the eight-week flood treatment (Table 7). White oak seedlings from bottomland seed sources had a greater number of seedlings that survived flooding treatments when compared to white oak seedlings from upland seed sources.

Bur and swamp white oak seedling survival data could not be tested with a Chi-square analysis for species response between topographic positions

Table 7: Chi-Square analysis of white oak seedling survival by three flooding treatments sorted for two topographic positions through the entire growing season.

White Oak Mortality

		Total	Live	Dead	% Live	$\aleph^2$	P
Control	Upland	48	48	0	100		
	Bottom	24	24	0	100		
	Total	72	72	0	100	NA[1]	---
4-Week	Upland	48	42	6	87.5		
	Bottom	24	23	1	95.8		
	Total	72	65	7	90.3	NA[1]	---
8-Week	Upland	48	29	19	60.4		
	Bottom	24	19	5	79.2		
	Total	72	48	24	66.7	44.57	< 0.001
Species	Upland	144	119	25	82.6		
	Bottom	72	66	6	91.7		
	Total	216	185	31	85.6	NA[1]	---

[1] Chi-square analysis inappropriate because of too few seedlings in one or more classes

independent of treatment and for any of the flooding treatments. All mortality percentages were below the 10% threshold (0-6.2%). With such low mortality, no significant conclusion can be made regarding survival of either species.

Some seedlings had lost all their leaves and were recorded as dead at the end of the eight-week flooding treatment. However, fifteen seedlings reflushed at some point before the end of the growing season. A bur oak from an upland seed source completely defoliated in the non-flooded control treatment and eventually added an entirely new flush before the end of the growing season. Five seedlings in the four-week flood treatment defoliated and reflushed before the end of the growing season. All seedlings were from upland seed sources and none of seedlings that reflushed exhibited basal sprouting. Nine defoliated seedlings reflushed in the eight-week flood treatment which included five bur oak and four white oak seedlings. The reflushing mechanism for three bur and white oak seedlings was basal sprouting. Basal sprouts emerged above the root collar and in some cases, above the soil line and below the water level during inundation. Across both flood treatments and independent of species, reflushing during the post-flood recovery period occurred in eleven defoliated seedlings from upland seed sources and three seedlings from bottomland seed sources.

Height Growth and Flushing Response

Family means for the percent of seedlings that had completed one or more flushes and the change in seedling height after initiation of the flood treatments through the end of the growing season was not normally distributed and could not be analyzed using the proposed analysis of variance model. There were 201 seedlings of 864 seedlings that initiated one or more new flushes during or post-flooding. When flushing percentage exceeded 10% for both topographic positions, count data was subject to Chi-square analysis. Only seedlings that exhibited a flush stage change by undergoing shoot elongation incurred a change in overall height.

A higher percentage of swamp white oak seedlings produced one or more flushes than did bur oak and white oak seedlings across all treatments from the initiation of flooding treatments through the end of the growing season (Figure 5). A higher percentage of swamp white and bur oak seedlings flushed in the non-flooded control treatment than did seedlings in both flood treatments. White oak seedlings tended to have a low percentage of seedlings that flushed across all flood treatments.

There was no difference in white oak seedling flushing response data for the control treatment between topographic positions. White oak seedling flushing response data could not be tested with a Chi-square analysis for species response between topographic positions independent of treatment, the four-week, and eight-week flood treatments because too few seedlings flushed.

Flushing response of bur oak seedlings was tested with a Chi-square analysis for differences between topographic positions for the overall species response and for the control treatment (Table 8). Bur oak seedlings from upland

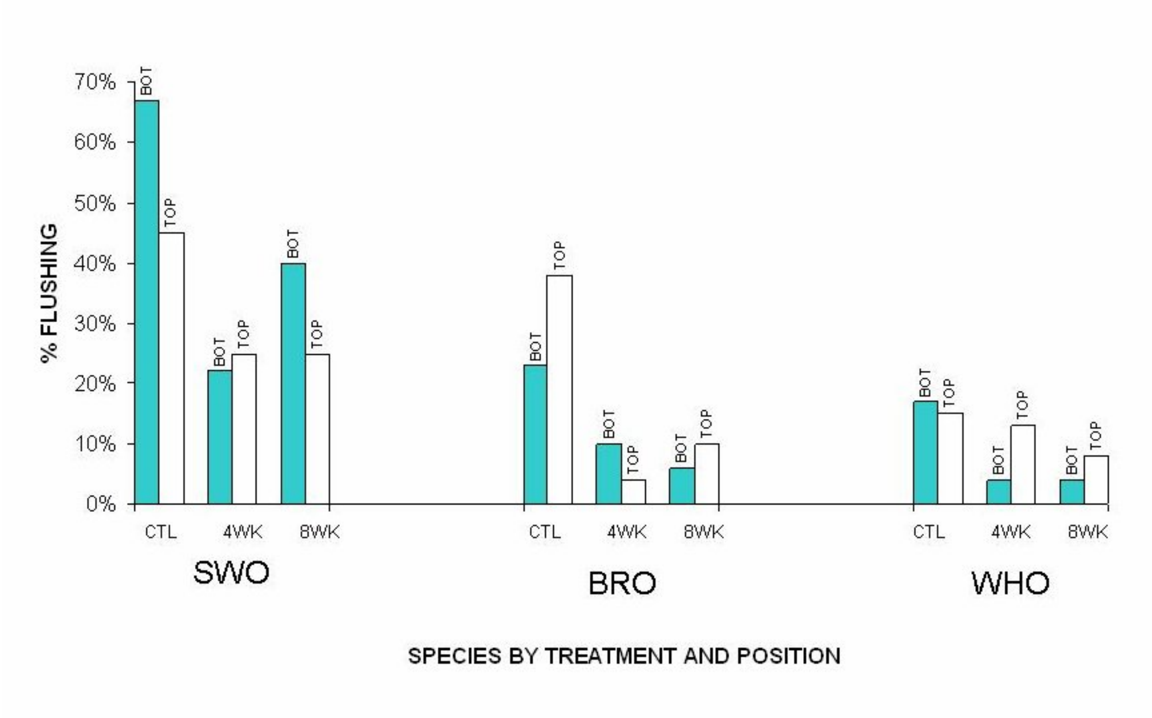

Figure 5: Percent of seedlings that flushed one or more times after initiation of flood treatments through the end of the growing season for upland (TOP) and bottomland (BOT) seed sources of swamp white oak (SWO), bur oak (BRO), and white oak (WHO).

seed sources had a greater flushing percentage than bur oak seedlings from bottomland seed sources. This finding does not support the proposed hypothesis that bottomland seed sources may be maladapted for outplanting as seedlings on bottomland sites. Bur oak seedling flushing response data could not be tested with a Chi-square analysis for the four-week and eight-week flooding treatments because too few seedlings flushed.

Swamp white oak seedling flushing response data tested with a Chi-square analysis for the four-week treatment indicated no significant difference between topographic positions. Post-flood flushing response of swamp white oak seedlings tested with a Chi-square indicated significant differences between topographic positions for overall species response, the non-flooded control, and the eight-week flooding treatment, but not the four-week flooding treatment (Table 9). Swamp white oak seedlings from bottomland seed sources had a greater flushing percentage when compared to swamp white oak seedlings from upland seed sources. This finding supports the hypothesis that upland seed sources of this species may be maladapted to outplanting as seedlings on bottomland sites.

Table 8: Chi-Square analysis of bur oak seedlings that completed one or more flushes through the end of the growing season by two topographic positions within and across the three flooding treatments.

Bur Oak Flushing

		Total	Flushed	No Flush	% Flush	$\aleph^2$	P
Control	Upland	48	18	30	37.5		
	Bottom	48	11	37	22.9		
	Total	96	29	67	30.2	36.23	< 0.001
4-Week	Upland	48	2	46	4.2		
	Bottom	48	5	43	10.4		
	Total	96	7	89	7.3	NA	---
8-Week	Upland	48	5	43	10.4		
	Bottom	48	3	45	6.3		
	Total	96	8	88	8.3	NA	---
Species	Upland	144	25	119	17.4		
	Bottom	144	19	125	13.2		
	Total	288	44	244	15.3	21.13	< 0.001

[1] Chi-square analysis inappropriate because of too few seedlings in one or more classes

Table 9: Chi-Square analysis of swamp white oak seedlings that completed one or more flushes through the end of the growing season by two topographic positions within and across the three flooding treatments.

Swamp White Oak Flushing

		Total	Flushed	No Flush	% Flush	$\aleph^2$	P
Control	Upland	60	27	33	45.0		
	Bottom	60	40	20	66.7		
	Total	120	67	53	55.8	87.27	< 0.001
4-Week	Upland	60	15	45	25.0		
	Bottom	60	13	47	21.7		
	Total	120	28	92	23.3	1.047	0.33
8-Week	Upland	60	15	45	25.0		
	Bottom	60	24	36	40.0		
	Total	120	39	81	32.5	47.41	< 0.001
Species	Upland	180	57	123	31.7		
	Bottom	180	77	103	42.8		
	Total	360	134	226	37.2	75.73	< 0.001

[1] Chi-square analysis inappropriate because of too few seedlings in one or more classes

Diameter Growth and Lenticel Frequency

Analysis of the change in diameter of the seedlings from the initiation of the flooding treatment through the end of the growing season indicated there was a significant (P = 0.0083) flooding by family interaction (Table 10). Family mean changes in basal diameters of bur oak seedlings showed four of the eight families in the four-week flooding treatment had a greater growth than seedlings in the non-flooded control treatment; however, only one family had statistically greater diameter growth than seedlings in the non-flooded control. The same is true for the eight-week flood treatment; however the increases are not consistent by family. A single family of bur oak (HW11B) had a significantly larger increase in basal diameter growth for seedlings in the eight-week flood treatment than that of all other bur oak families across all treatments. Of this family of bur oak, twenty five of thirty six seedlings had diameter growth exceeding 3.0 mm or more across all flooding treatments. Nineteen of twenty five seedlings of family HW11B with 3.0 mm or greater diameter growth were found in the four- and eight-week flood treatments. Five of ten families of swamp white seedlings in the eight-week flood treatment showed a significantly greater diameter growth than diameter growth of seedlings in the non-flooded control. Family mean basal diameter growth of swamp white oak seedlings showed that four of ten families increased in basal diameter in the four-week flood treatment and nine of ten families increased in the eight-week flood treatment when compared to the non-

Table 10: Average change in seedling basal diameter (Δ D), and percent of seedlings with hypertrophied lenticels (% LN) for families by species and contrasting topographic position.

			CONTROL		4-WEEK FLOOD		8-WEEK FLOOD	
Family	**Species**[1]	**Position**[2]	**Δ D**	**% LN**	**Δ D**	**% LN**	**Δ D**	**% LN**
BN21B	BRO	BOT	1.81	0	2.19	100	2.13	100
BN31B	BRO	BOT	1.78	0	1.66	75	1.64	100
CA11B	BRO	BOT	1.57	0	2.01	83	1.80	100
HW11B	BRO	BOT	2.08	0	2.38	100	3.42	100
AD11B	BRO	TOP	1.88	0	1.73	100	2.03	100
BN11B	BRO	TOP	1.74	0	1.80	100	1.92	100
BN12B	BRO	TOP	2.16	0	1.83	92	2.07	100
SH11B	BRO	TOP	1.76	0	1.22	100	1.13	88
BN12S	SWO	BOT	2.27	0	2.85	92	2.73	92
BN13S	SWO	BOT	2.09	0	2.48	92	3.19	100
CO11S	SWO	BOT	1.97	0	2.52	92	3.18	100
PK11S	SWO	BOT	1.03	0	1.46	67	2.40	100
PK21S	SWO	BOT	1.76	0	1.53	92	2.45	92
AD12S	SWO	TOP	1.48	0	2.18	83	2.63	100
AD14S	SWO	TOP	1.56	0	1.53	83	2.78	100
BN22S	SWO	TOP	1.56	0	1.86	92	2.45	100
CA21S	SWO	TOP	1.58	0	2.18	83	2.44	100
CA22S	SWO	TOP	1.95	0	2.33	100	2.63	100
CO11W	WHO	BOT	1.19	0	1.07	75	0.92	83
CO12W	WHO	BOT	1.92	0	1.58	83	0.83	83
BN12W	WHO	TOP	2.06	0	1.40	75	1.08	50
BN21W	WHO	TOP	1.21	0	0.68	67	0.57	50
BN23W	WHO	TOP	2.45	0	1.88	100	1.37	83
OS12W	WHO	TOP	2.51	0	1.00	75	0.83	42
Significance $LSD_{(r=12)\ 0.01}$ =					**0.52**	**14.16**[3]		

SOURCE	P > F	P > F
Flood	0.5342	0.0001
Species	0.0001	0.0005
Flood * Species	0.0001	0.0061
Position (Species)	0.0003	0.0537
Flood * Position (Species)	0.0285	0.0093
Family (Position * Species)	0.0001	0.0270
Flood * Family (Position * Species)	0.0083	0.4831

[1] Species codes are BRO = bur oak, SWO = swamp white oak, WHO = white oak

[2] Position codes are TOP = upland seed sources, BOT = Bottomland seed sources

[3] Data for lenticels was not normally distributed.

flooded control. Four of six families of white oak seedlings showed significant less growth in basal diameters in the eight-week flood treatment than the diameter growth for seedlings in the non-flooded control treatment. All families of white oak showed less growth in basal diameter with increased flood period.

The percent of seedlings with lenticels was significantly higher in both the four- and eight-week flood treatments when compared to seedlings in the non-flooded control treatment. The presence of lenticels may have accounted for some of the basal diameter growth of flooded seedlings. Mean basal diameters of seedlings with lenticels appeared to be larger for seedlings of bur and swamp white oak seedlings in the four- and eight-week flood treatments when compared to the non-flooded control treatment. In contrast, seedlings of white oak showed a trend of a lower number of seedlings having lenticels in the eight-week flood treatment than the four-week flood treatment. This could be explained by the number of white oak seedlings that appeared to have died over the course of the flooding treatments. Identification of lenticels may have been confounded on the dying and/or dead white oak seedlings due to the decay of the stem and root systems incurred during the flooding treatments.

There was a significant interaction among seedlings from bottomland seed sources when compared to seedlings from upland seed sources for bur and swamp white oak seedlings (Table 11). Bur and swamp white oak seedlings from bottomland seed sources had greater diameter growth when compared to basal diameter growth of bur and swamp white oak seedlings from

Table 11: Average seedling basal diameter growth (mm) and percent of seedlings with hypertrophied lenticels from the initiation of flood treatments through the end of the growing season.

		TREATMENT						POSITION BY SPECIES MEANS
		CONTROL		4-WEEK FLOOD		8-WEEK FLOOD		
SPECIES[1]	POSITION[2]	Δ D[3]	% LN[4]	Δ D[3]	% LN[4]	Δ D[3]	% LN[4]	Δ D[3]
SWO	TOP	1.63	0	2.02	88	2.59	100	2.08*
	BOT	1.82	0	2.17	87	2.79	97	2.26*
BRO	TOP	1.89	0	1.65	98	1.79	97	1.78*
	BOT	1.81	0	2.06	90	2.25	100	2.04*
WHO	TOP	2.06	0	1.24	79	0.96	56	1.42^{ns}
	BOT	1.56	0	1.33	79	0.88	83	1.26^{ns}
MEANS BY FLOOD TREATMENT		1.81	0	1.81	88	2.03	90	1.88

[1] Species codes are BRO = bur oak, SWO = swamp white oak, and WHO = white oak;
[2] Position codes are TOP = upland seed sources and BOT = bottomland seed sources;
[3] Δ D = seedling diameter growth (mm) from initiation of flood to end of growing season
[4] % LN = % of seedlings with lenticels from initiation of flood to end of growing season

upland seed sources. There was no difference in diameter growth of white oak seedlings from bottomland seed sources when compared to white oak seedlings from upland seed sources.

Dry Weights

Mean dry weights of roots had significant differences at the family ($P < 0.0001$) and position ($P < 0.0001$) levels and an interaction between flood treatment and species ($P < 0.0115$) (Table 12). Family mean dry weights of roots were quite variable across all species and positions with the largest being 23.84 g for the “McBaine Bur Oak” and the smallest being 4.58 g for white oak family BN21W. Analysis of dry weight of roots by topographic position within species showed a larger mean weight of bur and swamp white oak seedlings from bottomland seed sources when compared to bur and swamp white oak seedlings from upland seed sources (Table 13). There were no significant differences in dry weights of roots between seedlings from upland and bottomland seed sources for white oak. There was a significant interaction among species and flood treatment for root dry weight (Figure 6). Seedlings of all species showed an overall trend of decreasing root dry weights with increasing flood, however, unlike bur and white oak seedlings, swamp white oak seedlings had no difference between four and eight weeks of flooding.

Table 12: Average seedling dry weights of root, stem, and leaf components and leaf and specific leaf areas for families by species and position from contrasting topographic positions.

			DRY WEIGHTS[1]			LEAF AREAS[2]	
FAMILY	**SPECIES**	**POSITION**	**ROOT**	**STEM**	**LEAF**	**LA**	**SLA**
BN21B	BRO	Bottom	23.84	7.32	3.93	666.8	176.3
BN31B	BRO	Bottom	19.03	6.36	5.73	656.8	175.1
CA11B	BRO	Bottom	17.36	5.32	3.58	604.2	167.8
HW11B	BRO	Bottom	18.98	6.79	4.00	619.3	157.4
AD11B	BRO	Upland	15.78	5.08	3.62	564.1	157.1
BN11B	BRO	Upland	20.08	7.26	3.40	597.3	173.8
BN12B	BRO	Upland	18.68	7.13	4.21	689.9	167.4
SH11B	BRO	Upland	9.25	3.43	3.00	503.8	168.2
BN12S	SWO	Bottom	16.58	7.59	3.20	558.2	168.9
BN13S	SWO	Bottom	17.57	6.88	3.49	557.4	162.6
CO11S	SWO	Bottom	15.79	6.21	2.86	472.0	166.3
PK11S	SWO	Bottom	6.51	3.25	1.70	313.4	186.5
PK12S	SWO	Bottom	11.10	4.39	2.93	483.7	162.1
AD12S	SWO	Upland	11.36	4.71	2.51	420.2	170.8
AD14S	SWO	Upland	9.77	3.95	2.11	370.4	173.6
BN22S	SWO	Upland	12.99	5.12	2.82	465.2	165.5
CA21S	SWO	Upland	12.28	5.21	2.61	434.1	171.1
CA22S	SWO	Upland	12.72	5.64	2.35	386.0	167.8
CO11W	WHO	Bottom	5.73	1.90	1.63	229.6	145.3
CA12W	WHO	Bottom	8.20	2.75	1.80	273.2	157.8
BN12W	WHO	Upland	9.23	3.55	2.49	399.6	166.6
BN21W	WHO	Upland	4.58	1.58	1.08	157.5	153.0
BN23W	WHO	Upland	11.59	4.52	2.79	407.7	149.4
OS12W	WHO	Upland	7.00	2.68	1.80	232.1	145.5
Significance $LSD_{0.01\,(r=12)}$ =			**3.34**	**1.19**	**0.81**	**125.5**	**15.1**
SOURCE			P > F	P > F	P > F	P > F	P > F
Flood			0.0001	0.0001	0.1123	0.0366	0.1260
Species			0.0001	0.0001	0.0001	0.0001	0.0001
Flood * Species			0.0115	0.0001	0.6915	0.7580	0.0002
Position (Species)			0.0001	0.0001	0.0097	0.0029	0.7841
Flood * Position (Species)			0.1770	0.2759	0.2242	0.1877	0.7144
Family (Position * Species)			0.0001	0.0001	0.0001	0.0001	0.0001
Flood * Family (Position * Species)			0.0563	0.2364	0.2364	0.3126	0.5958

[1] Dry weights measured in grams

[2] Leaf area (LA) measure in cm^2 and specific leaf area (SLA) measure in cm^2/g

Table 13: Average seedling dry weights of root, stem, and leaf components and leaf and specific leaf areas for species and position from contrasting topographic positions.

		DRY WEIGHTS[1]			LEAF AREAS[2]	
SPECIES	POSITION	ROOT	STEM	LEAF	LA	SLA
Bur oak	Bottomland	19.80	6.45	4.31	636.8	169.2
Bur oak	Upland	15.95	5.73	3.56	588.8	166.6
Swamp white oak	Bottomland	13.51	5.66	2.84	476.9	169.3
Swamp white oak	Upland	11.82	4.93	2.48	414.2	169.7
White oak	Bottomland	6.97	2.33	1.72	251.4	151.5
White oak	Upland	8.10	3.08	2.04	299.2	153.6
Significance $LSD_{0.01(r=48)}$ =		**1.67**	**0.60**	**0.40**	**62.7**	**7.6**

[1] Dry weights measured in grams

[2] Leaf area (LA) measure in cm^2 and specific leaf area (SLA) measure in cm^2/g

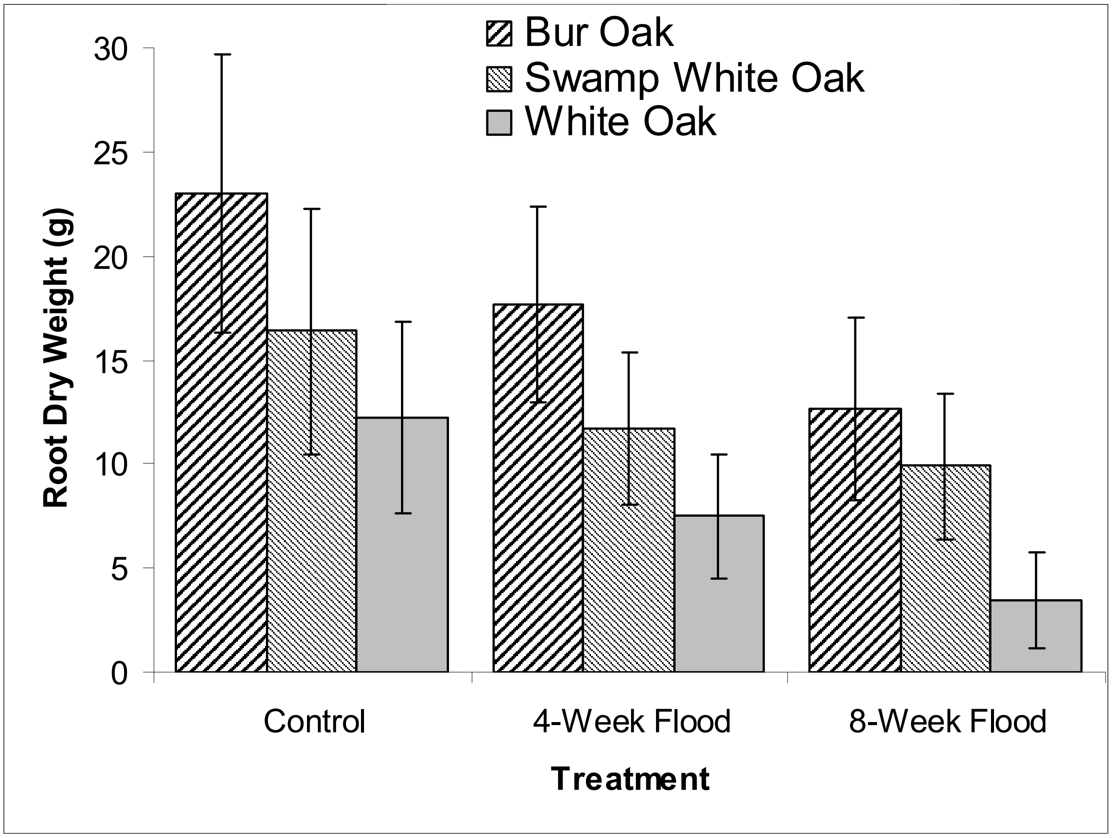

Figure 6: Average dry weight (± SD) by flood treatment of roots from bur, white, and swamp white oak seedlings.

Mean dry weights of stems had significant differences at the family (P < 0.0001) and position (P < 0.0001) levels and an interaction between flood treatment and species (P < 0.0001) (Table 12). Family mean dry weights of stems were quite variable across all species and positions with the largest being 7.59 g for swamp white oak family BN12S and the smallest being 1.58 g for white oak family BN21W. Analysis of dry weight of roots by topographic position within species showed that bur and swamp white oak seedlings originating from bottomland seed sources had significantly larger dry weights of stems when compared to seedlings originating from upland seed sources (Table 13). In contrast, white oak seedlings from upland seed sources had significantly larger stem dry weights than white oak seedlings from bottomland seed sources. There was a significant interaction for shoot dry weight among species and flood treatment (Figure 7). Seedlings of bur and white oaks showed an overall trend of decreasing stem dry weights with increasing flood duration, in contrast to swamp white oak seedlings that had no difference between any of the flooding treatments.

Mean dry weights of leaves had significant differences at the family (P < 0.0001), position (P < 0.0097), and species (P < 0.0001) levels (Table 12). Family mean dry weights of leaves were quite variable across all species and positions with the largest being 5.73 g for bur oak family BN31B and the smallest being 1.08 g for white oak family BN21W. Analysis of dry weights of leaves by position within species showed a larger mean weight for bur oak seedlings

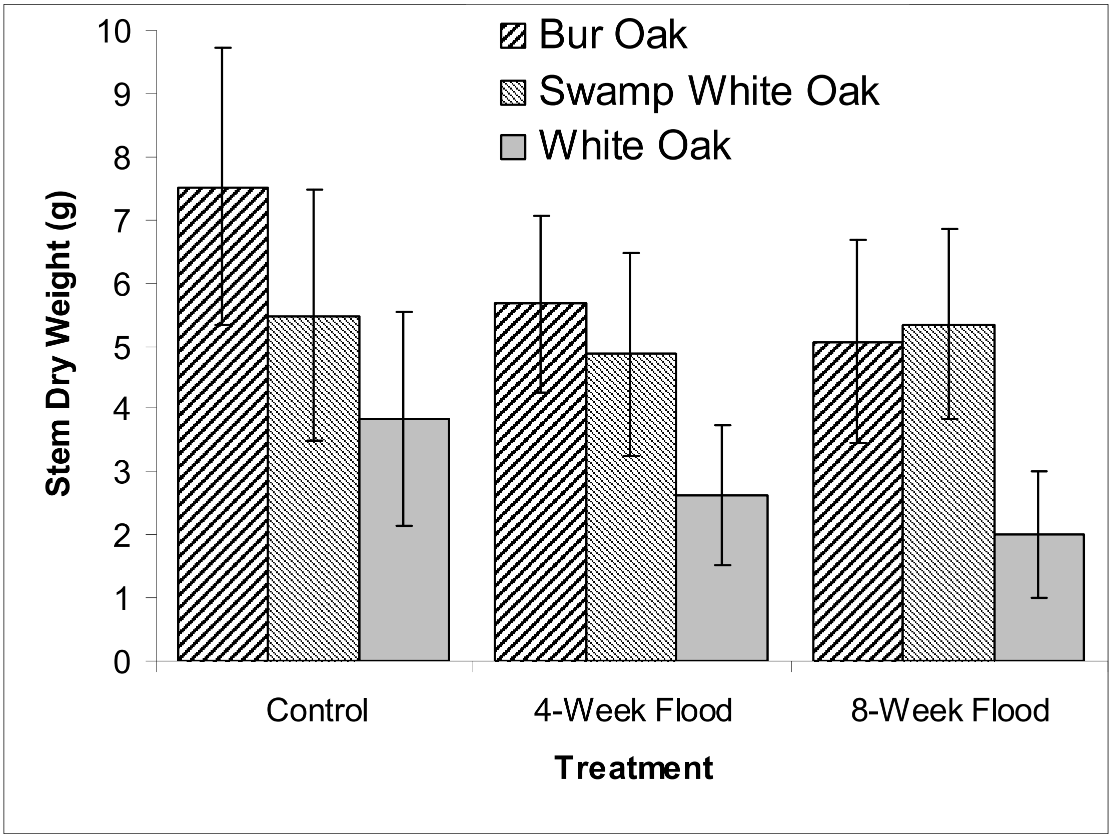

Figure 7: Average dry weight (±SD) by flood treatment of stems from bur, white, and swamp white oak seedlings.

from bottomland seed sources when compared to bur oak seedlings from upland seed sources (Table 13). There were no significant differences in dry weights of leaves between seedlings from upland and bottomland seed sources for swamp white or white oaks. Dry weights of leaves in bur oak were higher than dry weights of leaves for swamp white and white oak seedlings (Figure 8). Also, swamp white oak seedling leaf dry weights were greater than white oak seedlings.

Leaf Areas

Mean leaf areas per seedlings had significant differences at the family ($P < 0.0001$), position ($P < 0.0029$), species ($P < 0.0001$), and flood ($P < 0.0366$) levels (Table 12). Family mean leaf areas were quite variable across all species and positions with the largest being 690 cm^2 for bur oak family BN12B and the smallest being 158 cm^2 for white oak family BN21W. Swamp white oak seedlings from bottomland seed sources had larger leaf areas than swamp white oak seedlings from upland seed sources with no difference due to topographic position for seedlings of bur and white oaks (Table 13). Mean leaf areas of bur oak seedlings were larger than leaf areas of swamp white and white oak seedlings (Figure 9). Also, swamp white oak seedling leaf areas were larger than white oak seedlings. Mean leaf area of seedlings of all species in the non-flooded control treatment (526 cm^2) was significantly greater than mean leaf area of seedlings of all species in the four-week flooding treatment (438 cm^2) and eight-week flooding treatment (419 cm^2).

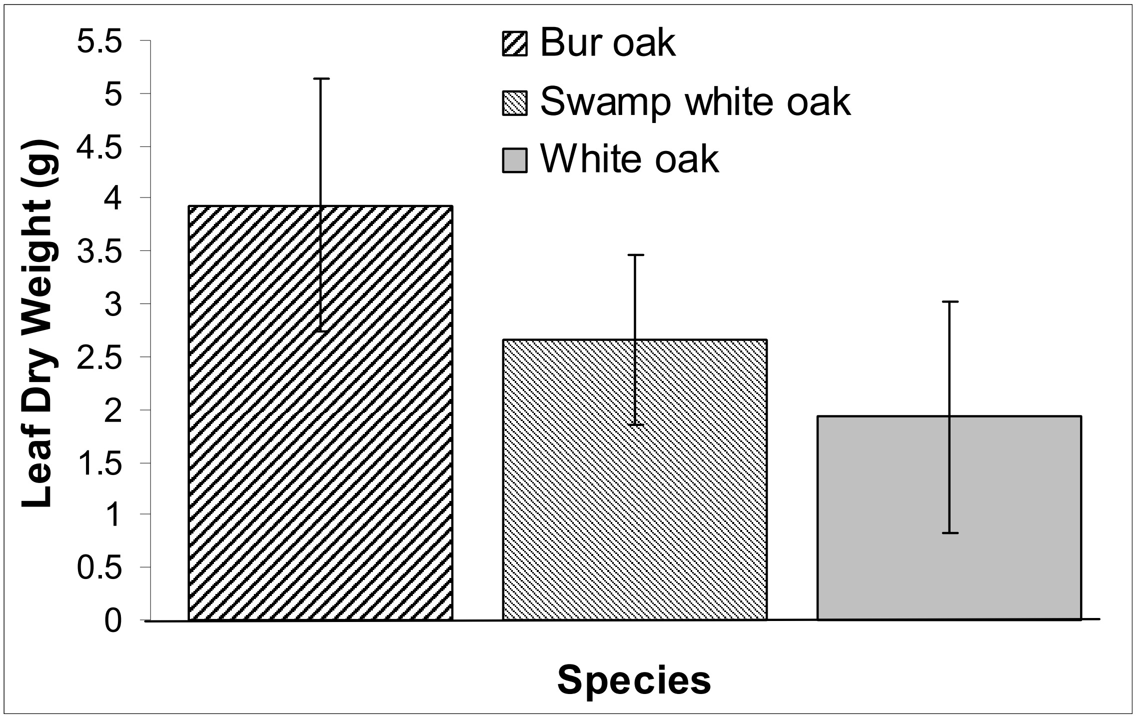

Figure 8: Average leaf dry weight (± SD) from bur, swamp white, and white oak seedlings.

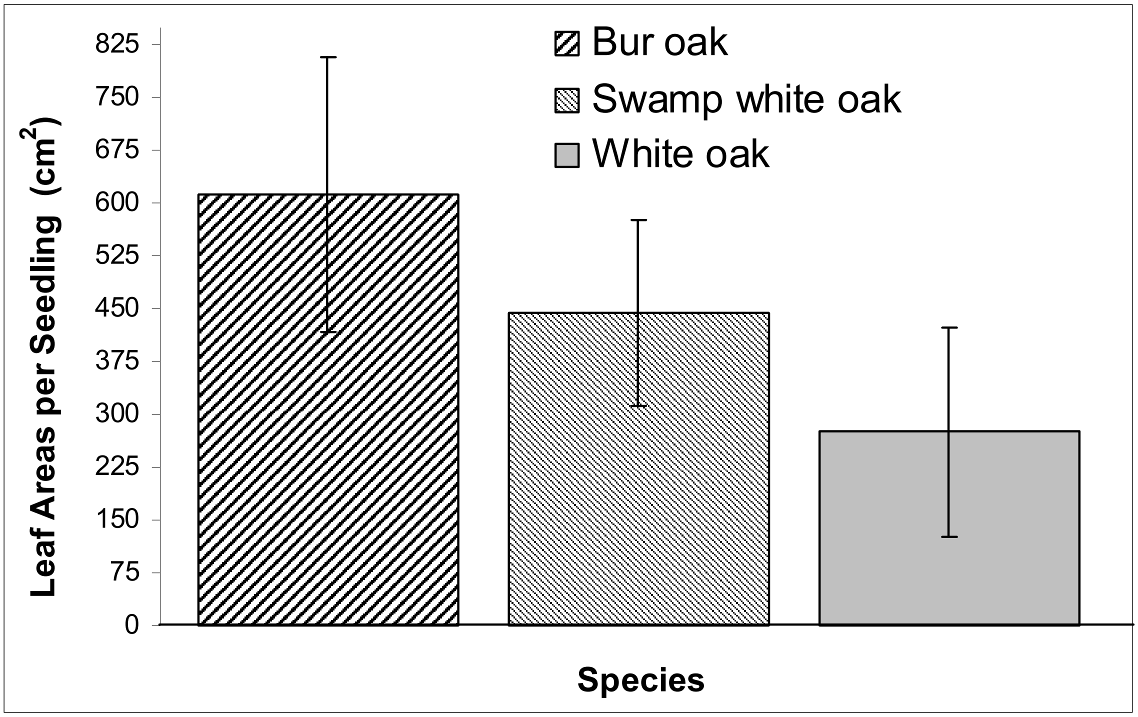

Figure 9: Average leaf area per seedling (± SD) from bur, swamp white, and white oak seedlings.

Mean specific leaf areas had significant differences at the family ($P < 0.0001$) level and an interaction between flood treatment and species ($P < 0.0002$). Family mean leaf areas were quite variable across all species and positions with the largest being 176 cm^2/g for the “McBaine Bur Oak” and the smallest being 145 cm^2/g for white oak family CO11W. There was a significant interaction for specific leaf area among species and flood treatments (Figure 10). Seedlings of all species showed a trend of decreasing specific leaf areas in the four-week flood treatment when compared to the non-flooded control treatment. Bur and white oak seedlings showed a trend of increasing specific leaf areas in the eight-week flood treatment when compared to seedlings in the non-flooded control and four-week flood treatment. There was no difference in the specific leaf areas of swamp white oak seedlings in the eight-week flood treatment when compared to seedlings in the four-week flood treatment.

Root Fibrosity

Root fibrosity scores for bur oak seedlings were compared to distributions of root fibrosity for the destructively sampled seedlings at the time of flooding and in the non-flooded control treatment seedlings at the end of the growing season. Chi-Square analysis indicated a significant difference ($P < 0.001$) in the root fibrosity of bur oak seedlings in all three flooding treatments when compared to the root fibrosity of pre-flood destructively sampled bur oak seedlings (Table 14). There were a higher percentage of seedlings scoring a 5 before initiating the flood treatments than at the end of the growing season for seedlings in all three

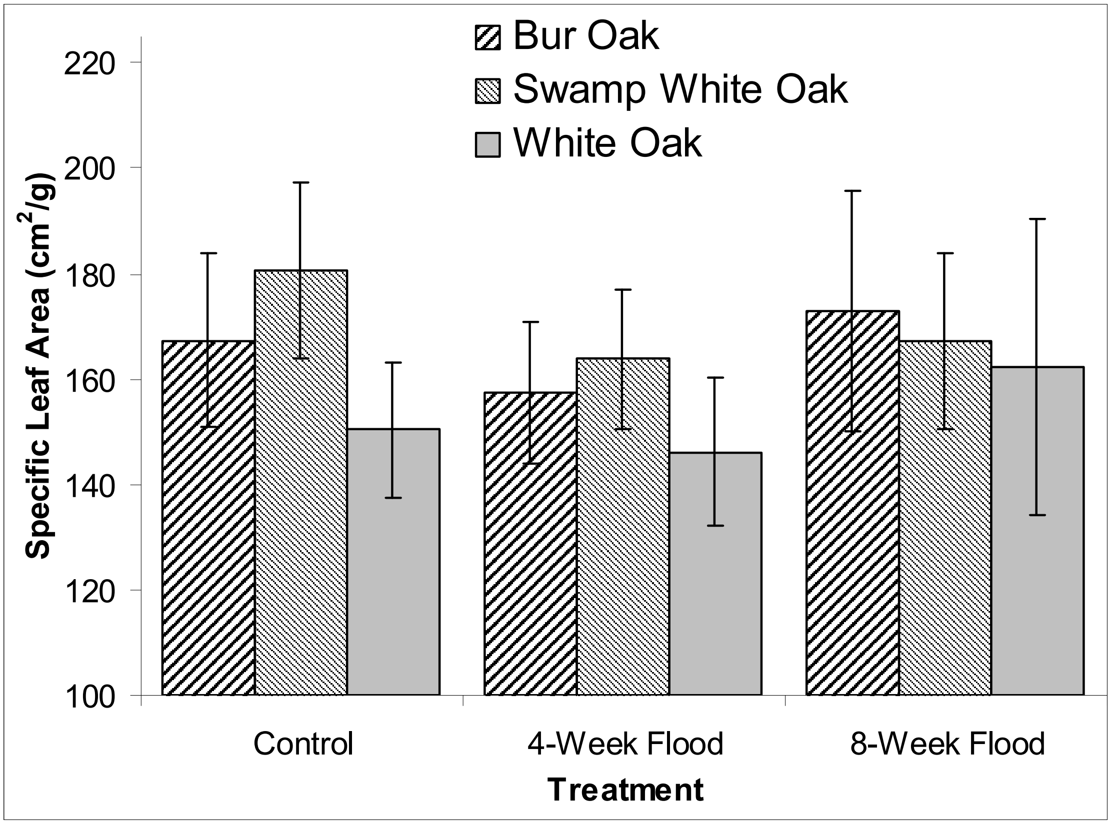

Figure 10: Average specific leaf area (±SD) by flood treatment from bur, white, and swamp white oak seedlings.

Table 14: Pre-flood and post-flood distribution of bur oak seedlings by root fibrosity classes.

		ROOT FIBROSITY[1]				א^2	
Treatment	N	1+2	3	4	5	Pre-Flood[2]	Control[3]
Pre-Flood	50	3	7	9	21	----	----
Control	120	1	19	64	12	32.3*	----
4-Week	120	1	18	64	13	31.4*	0.07^{ns}
8-Week	120	2	18	58	17	22.6*	1.51^{ns}

[1] Root fibrosity classes ranging from 1 (least fibrous) to 5 (very fibrous)

[2] Chi-Square analysis results using seedlings from pre-flood destructive sampling (df = 3)

[3] Chi-Square analysis results using seedlings from non-flooded control treatment (df = 3)

flood treatments. There was no difference in the distribution of root fibrosity scores of bur oak seedlings that had been flooded for four or eight weeks when compared to the distribution of root fibrosity scores for bur oak seedlings from the non-flooded control treatment.

Changes in the distribution of root fibrosity scores for swamp white oak seedlings were also compared to distributions of root fibrosity for the destructively sampled seedlings at the time of flooding and seedlings in the non-flooded control treatment at the end of the growing season. Distribution of root fibrosity classes of swamp white oak seedlings were altered by the four-week flooding treatment ($P < 0.01$) but not the eight-week flooding treatment ($P < 0.10$) when compared to the non-flooded control treatment (Table 15). There were a higher percentage of seedlings scoring a 3 and a lower percentage of seedlings scoring a root fibrosity score of a 4 in the four-week flooding treatment when compared to the non-flooded control treatment. In contrast to bur oak seedlings, there was no difference in the root fibrosity of the pre-flood destructively sampled seedlings when compared to seedlings in the three flooding treatments at the end of the growing season.

There were no significant differences in the root fibrosity of pre-destructively sampled white oak seedlings when compared to seedlings in the three flooding treatments. There was also no difference in the root fibrosity of seedlings in the four- and eight-week flooding treatments when compared to the non-flooded control treatment. There was a general trend for more white oak seedlings falling into the 1-2 root fibrosity class more so than the 3-5 root fibrosity

Table 15: Pre-flood and post-flood distribution of swamp white oak seedlings by root fibrosity classes.

		ROOT FIBROSITY[1]				$\aleph^2$	
Treatment	N	1+2	3	4	5	Pre-Flood[2]	Control[3]
Pre-Flood	50	7	25	16	2	----	----
Control	120	13	47	44	16	4.40^{ns}	----
4-Week	120	21	65	23	11	4.14^{ns}	12.3*
8-Week	120	22	53	28	17	4.94^{ns}	6.26^{ns}

[1] Root fibrosity classes ranging from 1 (least fibrous) to 5 (very fibrous)

[2] Chi-Square analysis results using seedlings from pre-flood destructive sampling (df = 3)

[3] Chi-Square analysis results using seedlings from non-flooded control treatment (df = 3)

class in pre-destructively sampled seedlings and in all three of the flooding treatments.

CHAPTER 5

DISCUSSION

By choosing a flood-intolerant oak species (white oak) and two intolerant to moderately tolerant oak species (bur and swamp white oak) the study was expected to produce significant interactions among flood treatments and species, position within species, and/or family within position within species. Of the seven variables that could be analyzed using analysis of variance, one or more of these interactions were found with all of the variables.

Flood by Species Interactions

The interaction between flood by species showed significant differences for diameter growth after the initiation of flooding treatments through the end of the growing season, percent of seedlings with hypertrophied lenticels, and end of growing season root dry weight, stem dry weight, and specific leaf area. Swamp white and bur oak seedlings in the four- and eight-week flooding treatments had the largest increase in diameter growth with increased flood duration while the white oak seedlings in the four- and eight-week flooding treatments exhibited the least basal diameter growth with increased duration of flooding. This implies that of the three oak species studied, swamp white and bur oak may be the best suited for outplanting on bottomland sites. This is validated by Kabrick and others (2007) who found that swamp white oak seedlings maintained a high growth rate (basal diameter) with three and five weeks of flooding in their flooding study. They also reported a moderate increase in basal diameters of bur oak seedlings in their flooding study. In this study, there was a general trend of increasing change in basal diameter with increased flooding for seedlings of bur and swamp white oaks. This could be attributed to the laying down of arenchyma tissue in response to flooding which creates large intercellular spaces to facilitate oxygen diffusion (Coutts and Phillipson 1978; Kozlowski and Pallardy 1997). Connor and others (1998) reported flood by species interactions for diameter growth in overcup oak (*Quercus lyrata*), swamp chestnut oak (*Q. michauxi*), water oak (*Q. nigra*), and nuttall oak (*Q. nuttallii*).

The formation of hypertrophied lenticels may have contributed to the increase in basal diameter; however, swelling of the submerged portion of the stem could not be completely explained by lenticel formation. All three species examined produced hypertrophied lenticels over the duration of the study. Lenticels were limited to forming on seedlings in the four- and eight-week flooding treatments with a higher number of seedlings initiating lenticel formation on swamp white and bur oak than on white oak. The formation of lenticels on white oak seedlings was difficult to quantify due to mortality and subsequent decay of the taproot and submerged portions of the stem. Tang and Kozlowski (1983) noted the formation of hypertrophied lenticels on bur oak seedlings after five days of seedling inundation by floodwaters and an increase thereafter. The formation of hypertrophied lenticels has been noted for swamp tupelo (*Nyssa* *aquatica*), eastern cottonwood, English oak (*Quercus robur*), northern red oak (*Q. rubra*), and pin oak (*Q. palustris*) following inundation by floodwaters (Angelov and others 1996; Cao and Connor 1998; Chirkova and Gutman 1972; Colin-Belgrand and others 1990). The basal swelling and subsequent change in diameter growth is a function of the development of arenchyma tissue and the formation of lenticels, which has been indicated by Tang and Kozlowski (1982) as adaptations for flood tolerance. White oak seedlings that are generally believed to be intolerant of flooding had an overall decrease in basal swelling with increasing flood duration, which supports this idea.

End of growing season root dry weights showed an interaction between flooding and species. Bur and white oak seedlings responded identically with a decrease in root dry weight with flooding duration. However, swamp white oak seedling root dry weight decreased from the non-flooded control treatment to the four-week flooding treatment with no change in root dry weights from the four-week flooding treatment to the eight-week flooding treatment. The ability of swamp white oak seedlings to continue to add biomass to their root systems independent of flooding for four- or eight-weeks indicates it is a well-suited species for outplanting on bottomland sites that are prone to flooding for periods of four to eight weeks during the growing season. Connor and others (1998) reported flood by species interactions for root dry weights in overcup, swamp chestnut, and nuttall oaks. Pezeshki and others (1999) also found significant flood by species interactions for root dry weights in seedlings of nuttall and cherrybark oaks (*Q. pagoda*).

End of growing season stem dry weights showed an interaction between flooding and species. As was the case in root dry weights, bur and white oak seedlings showed a similar trend of a decrease in stem dry weight with an increase in flooding duration. However, swamp white oak seedlings showed no difference in root dry weights across all three flood treatments. The ability of swamp white oak seedlings to maintain similar stem dry weights regardless of flooding treatment confirms that swamp white oak is well-suited for outplanting on bottomland sites that are prone to flooding. Connor and others (1998) reported flood by species interactions for stem dry weights in overcup, swamp chestnut, and nuttall oaks. Pezeshki and others (1999) also found significant flood by species interactions for stem dry weights in seedlings of nuttall and cherrybark oaks.

Specific leaf areas calculated at the end of the growing season indicated there a flood by species interaction. As was the case with root and stem dry weights, bur and white oak seedlings responded similarly across treatments while swamp white oak seedlings responded differently across treatments. Specific leaf areas of bur and white oak seedlings exhibited a decrease from the non-flooded control treatment to the four-week flooding treatment and exhibited an increase from the four-week flooding treatment to the eight-week flooding treatment. In contrast, swamp white oak seedlings responded with no difference in specific leaf area in the four-week flooding treatment when compared to the eight-week flooding treatment, however, swamp white oak seedlings followed the same pattern of specific leaf areas decreasing in the four-week flooding treatment when compared to swamp white oak seedlings in the non-flooded control treatment.

Flood Effects

The effect of flooding was significant only for end of growing season leaf areas. All other variables that appeared to be affected by flooding exhibited a flooding by species interaction and were discussed in the previous section. Flooding significantly reduced the amount of leaf area of seedlings of all species in the four- and eight-week flooding treatments when compared to the non-flooded control treatment. Several authors reported reduced leaf areas as a result of flooding in eastern cottonwood seedlings derived from cuttings and in seedlings of nuttall, cherrybark, and swamp chestnut oaks (Cao and Connor 1999; Pezeshki and others 1999; Angelov and others 1996).

Species Effects

There was a species effect for pre-flood height above the root collar, end of growing season leaf dry weight, and end of growing season leaf area. Heights above the root collar before initiation of flood treatments showed variation across species. Bur oak seedlings exhibited the tallest mean two-flush seedling heights (37.4 cm) followed by two-flush seedlings originating from swamp white oak seed sources (29.7 cm) and the shortest two-flush seedlings were from the white oak seed sources (20.9 cm). Since all of the seedlings were removed from cold storage and placed in the greenhouse at the same time, it is obvious that differences exist among species of oak in terms of initial seedling growth rates. This would be more important in the regeneration of oak seedlings naturally via parent trees in flooded settings where initial height growth is dependent upon the local environment and light availability. It is clear that seedlings grown in a nursery that are taller may have a better chance of tolerating flooded conditions than smaller seedlings.

Leaf dry weights and area varied by species. Seedlings from bur oak seed sources exhibited the largest mean leaf dry weights (3.94 g) followed by seedlings from swamp white oak seed sources (2.66 g) and the lightest were seedlings from the white oak seed sources (1.93 g). Seedlings from bur oak seed sources exhibited the largest mean leaf area (613 cm^2) followed by seedlings from swamp white oak seed sources (446 cm^2) and the smallest were seedlings from the white oak seed sources (275 cm^2).

Flood by Position within Species Interactions

There was a flood by position within species interaction only for percent of seedlings that produced hypertrophied lenticels. A high percentage of seedlings from all species produced lenticels in the four- and eight-week flooding treatments while no seedlings of any species produced lenticels in the non-flooded control treatment. The interaction occurred because white oak seedlings in the four- and eight-week flooding treatments resulted in fewer seedlings with lenticels when compared to bur and swamp white oak seedlings in the same flooding treatments. Many white oak seedlings either died or exhibited stem and/or taproot decay, thereby masking lenticel formation, while few bur and swamp white oak seedlings died or exhibited stem and/or taproot decay.

Position within Species Effects

There was a position within species effect for pre-flood heights, diameter growth from the initiation of flooding treatments through the end of the growing season, and end of growing season root dry weight, stem dry weight, leaf dry weight, and leaf area. Prior to initiation of flooding treatments bur and swamp white oak seedlings from bottomland seed sources had significantly greater seedling heights above the root collar when compared to bur and white oak seedlings from upland seed sources. The opposite was true for white oak seedlings. This infers height serves as some adaptation of flood tolerance. Rapid seedling growth resulting in constituent foliage remaining above the level of the floodwater may serve to facilitate survival. Bauerle and others (2003) found that red maple seedlings from bottomland sites were taller than the same age seedling from upland sites.

There was a position within species effect for diameter growth from the initiation of flooding treatments through the end of the growing season for bur and swamp white oak seedlings. This provides evidence that the bottomland seed sources may be better adapted to tolerate periods of inundation. The increase in diameter, whether it is due to basal swelling caused by the formation of aerenchyma tissue or by the formation of hypertrophied lenticels on the submerged portions of the stems, it is an adaptation to flood tolerance. The basal swelling may also be attributed to the production of new cells in the xylem, which could also be an indicator of flood tolerance because the tree is responding to partial inundation by floodwaters with normal growth.

There was a position within species effect for end of growing season root dry weight. Bur and swamp white oak seedlings from bottomland seed sources had larger root dry weights when compared to bur and swamp white oak seedlings from upland seed sources. This is supported in the literature with bottomland seed sources of blackgum producing heavier root systems under flooded conditions when compared to upland seed sources of blackgum (Keeley 1979). With a larger root system there may be more of a capacity within the seedling root system to produce first-order lateral roots which may in turn increase their respective flood tolerances. White oak seedlings showed no difference in root dry weight between seedlings from upland seed sources and seedlings from bottomland seed sources.

There was a position within species effect for end of growing season stem and leaf dry weight. Bur and swamp white oak seedlings from bottomland seed sources had larger stem and leaf dry weights when compared to bur and swamp white oak seedlings from upland seed sources. This finding is supported in the literature with bottomland seed sources of blackgum producing heavier aboveground component weights under flooded conditions when compared to upland seed sources of blackgum (Keeley 1979). Having more biomass allocated to the aboveground portion of the seedlings may give seedlings a better chance at surviving periods of inundation. White oak seedlings showed no difference in leaf dry weight between seedlings from upland seed sources and seedlings from bottomland seed sources. Stem dry weights of white oak seedlings from upland seed sources were greater than stem dry weights of white oak seedlings from bottomland sites. This is difficult to understand and has occurred in other variables at different levels of analysis for white oak seedlings.

There was a position within species effect for end of growing season leaf areas. Bottomland bur and swamp white oak seed sources had greater leaf areas than upland seed sources. This in combination the aforementioned position within species significant variables shows that bottomland seed sources of bur and swamp white oak have the capability to outperform their upland counterparts. This is of critical importance in ensuring that seedlings of oak (and other) species used in the restoration of bottomland sites are to survive.

CHAPTER 6

CONCLUSIONS

The findings of this study confirmed the flood tolerance ratings for swamp white oak that had been compiled by several authors indicating it ranged from somewhat tolerant to tolerant (Table 1). Seedlings of swamp white oak had the highest flood tolerance of the three species evaluated. Similarly, the study confirmed the previously reported flood tolerance ratings for bur oak compiled from the same authors with the exception of the intolerant rating by Keeley and others (2001). While bur oak seedlings were not as tolerant of flooding as were swamp white oak seedlings, they still were moderately-tolerant of flooding. This study did not confirm Keeley and others (2001) flood tolerance rating of intolerant.

The flood tolerance ratings for white oak ranged from intolerant to slightly tolerant of flooding according to published sources. In this study, white oak seedlings responded to flooding poorly when compared to swamp and bur oak seedlings. White oak seedlings suffered more mortality, had decreased growth, and showed more root decay than seedlings of swamp white and bur oak. However, this study showed that some white oak seedlings were able to tolerate flooding, contradicting most of the preciously reported flood tolerance ratings. As a result of this study, it was confirmed that swamp white oak was the most flood tolerant, while white oak was the least flood tolerant.

Are upland seed sources maladapted for outplanting on bottomland sites? In this study bottomland seed sources of swamp white and bur oak outperformed their upland counterparts in diameter growth and end of growing season leaf area, root dry weight, and stem dry weight. Bottomland seed sources of bur oak outperformed their upland counterparts for the same variables as swamp white oak with the addition of end of growing season leaf dry weight. The lack of an interaction between flooding and position within species supports the finding that bottomland seed sources of bur and swamp white oak are better suited for outplanting not only on bottomland sites, but on any site. This is not the case for bottomland seed sources of white oak. Although the occurrences of bottomland white oak trees are not common, study findings indicate that upland seed sources should continue to be used in seedling production over using seed from bottomland seed sources of white oak.

Are certain families of each oak species better suited for outplanting on bottomland sites? A land manager would benefit by selecting one or more families, such as the “McBaine Bur Oak” (family BN21B) based on diameter growth. Based on comments by nursery managers across Missouri, excellent height and diameter are desirable. There are countless steps that go into the production of seedlings in a nursery to ensure that the highest quality seedlings are propagated and sold. By selecting a superior seed source for nursery growing stock, there should be a larger amount of high quality seedlings produced for outplanting on bottomland sites or elsewhere. By recommending that seed collectors concentrate on bottomland sites for the flood tolerant oak species, there exists the capacity to improve public and private nursery growing stock available in this state and others for the benefit of everyone.

Future research on flood tolerance should concentrate on bottomland oak species. More containerized seedling stock tank studies should be carried out using different treatments, which could include longer duration flooding. Future research should select the best families from this and any other studies that have examined oaks in Missouri and use them in future pot studies. Future studies could use growing medium as a treatment and look at any possible differences between horticultural growing medium and actual riparian soil, only if a homogeneous source is used. Future studies should continue to use shade cloth as a treatment as well. By repeating the study under different light regimes, it may be possible to isolate specific families that will thrive in open sun for afforesting a large field. Isolating a family that will thrive under heavy shade could help with regeneration issues of oak species in green tree reservoirs, such as Mingo National Wildlife Refuge, which relies on its hard mast producing oak species for countless migrating bird species.

It would be beneficial to expand this research to other oak species that are found on Missouri bottomland sites (cherrybark oak) or found on both upland and bottomland sites (pin oak). Research has begun in some of these areas using acorns collected from the same mother trees in the same seed year. Seedlings have been grown in various nurseries and have been outplanted in the Flood Tolerance Laboratory in New Franklin, MO and at the USDA Plant Materials Center in Elsberry, MO. The same families have also been placed into another containerized seedling study using 1-0 seedlings rather than ½-0 seedlings that were used in this study (Mark Coggeshall and Jerry Van Sambeek, personal communication).

Future research should benefit for focusing on fewer variables. In this study, it was found that diameter growth, and dry weights seemed to be the best indicators of a seedling's tolerance to and recovery from flooding. The measurement of stem diameters needs to look at above and below the floodwaters to better assess basal swelling and diameter growth as a measure of flood tolerance.

CHAPTER 7

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