SITE PREPARATION FOR LONGLEAF PINE RESTORATION ON HYDRIC SITES:

STAND DEVELOPMENT AND GROUND FLORA RESPONSES 15 YEARS AFTER PLANTING

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by

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DEDICATION

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ABSTRACT

Longleaf pine (*Pinus palustris* Mill.) restoration is an important land management goal throughout the Southeast. On hydric sites within the Atlantic Coastal Plain, restoration may involve site preparation prior to planting in order to overcome challenges to seedling establishment, such as abundant competition and poor soil drainage. Short-term seedling responses to site preparation on these sites are well understood, with site preparation typically improving longleaf pine seedling growth but not survival. Investment in site preparation assumes that treatments will result in long-term benefits to stand establishment, yet lasting impacts of site preparation on longleaf pine are not well understood. Additionally, longleaf pine ecosystem restoration seeks not only to establish longleaf pine but also to maintain or improve the understory plant community. Little research has been conducted on the long-term effects of site preparation on vegetation in flatwoods sites. To fill these gaps in scientific understanding, we sampled longleaf pine plantations in Onslow County, North Carolina through three years and at 15 years after site preparation and planting. The eight study treatments we tested include an untreated control, six combinations of two vegetation control treatments (chopping or herbicide) with three soil manipulation treatments (flat [no treatment], mounding, or bedding), and an herbicide-chopping-bedding treatment. Our objectives were to determine the long-term impacts of site preparation on longleaf pine stand development and on the surrounding ground flora.

Our findings indicate that site preparation significantly improved long-term establishment and growth of longleaf pine on hydric flatwoods sites. Specifically, herbicide resulted in greater growth, higher survival, and earlier grass stage emergence compared to chopping. Similarly, soil manipulation treatments resulted in improved stand establishment outcomes relative to flat-planting (no treatment). Differences in survival and growth between treated and untreated areas increased through time, which demonstrates that site preparation had lasting, long-term impacts on longleaf pine stand development. Additionally, stem analysis indicated that increased height growth after grass stage emergence seemed to contribute more directly to long-term stand establishment outcomes than did the timing of grass stage emergence. In terms of understory responses to site preparation, we found that the chop-only treatment resulted in the greatest plant species diversity and cover of bunchgrasses and wiregrass. In the first three years after application, treatments with herbicide, particularly those crossed with soil manipulation, appeared be moving sites towards the natural vegetation community by reducing shrub cover without reducing bunchgrass cover. However, by year 15 these treatments had reverted away from the natural community due to long-term reductions in cover of bunchgrasses and wiregrass. We hypothesize that this may be due to a fire return interval that was too infrequent and interruption of fire continuity on soil manipulation treatments, both of which may have contributed to the failure to maintain the early improvements obtained from site preparation. Taken together, intermediate treatments, such as chopping and bedding, chopping and mounding, or herbicide alone, may provide the optimal balance between establishment and growth of planted longleaf pine with maintenance and improvement of the understory on hydric flatwoods sites.

Chapter 1: Literature Review

History and Current State of the Longleaf Pine Ecosystem

Historical Extent and Decline

When European settlers first arrived in the southeastern United States in the 16th century, they would have stumbled into a seemingly endless and remarkably diverse swath of forested land dominated by longleaf pine (*Pinus palustris* Mill.). All along the Southeast, from Virginia to Texas, longleaf pine occupied an estimated 37 million hectares (Frost, 2006). However, by 2000, one of the most extensive forest ecosystems in North America had been reduced to approximately 2% of its pre-settlement range, occupying an estimated 1 million ha (Frost, 2006). This reduction was due to a number of anthropogenic changes after European settlement, including widespread timber harvest and turpentine production, grazing by hogs and other livestock, land conversion for urban and agricultural uses, and, perhaps most importantly, fire exclusion (Frost, 2006; Landers et al., 1995).

The most recent estimate by Oswalt et al. (2012) found that, although the geographic extent of longleaf pine continued to shrink, the total hectarage had increased. Using forest inventory and analysis (FIA) data from 2010, Oswalt et al. (2012) estimated that longleaf pine occupied over 1.7 million ha, which is around 4.5% of its pre-settlement range. This total includes over 1.3 million ha of longleaf pine forest and almost 400,000 ha of mixed longleaf pine and oak (*Quercus* spp.) forest (Oswalt et al., 2012). Although this increase in hectarage is an encouraging sign, the character and extent of longleaf pine forests have been altered dramatically since European settlement. Today's longleaf pine ecosystems remain only a vestige of what were once the most dominant feature of the southeastern landscape.

Contemporary Longleaf Pine Restoration

This dramatic reduction in areal extent of longleaf pine, along with its economic and ecological values, have made longleaf pine ecosystem restoration an important land management objective throughout the Southeast. Economically, longleaf pine forests can be profitable and versatile resources for landowners. Longleaf pine produces high quality timber, abundant pine straw, and ideal habitat for hunting (Brockway et al., 2005; Landers et al., 1995). Moreover, longleaf pine is fire tolerant, can thrive on harsh sites, and is more resistant to insects and disease and less susceptible to damage from ice and wind than other southern pines (Johnson and Gjerstad, 2006; Landers et al., 1995; Mitchell et al., 2006).

From an ecological perspective, longleaf pine ecosystems include some of the most diverse plant communities outside of the tropics (Walker and Silletti, 2006), with the richest savannas containing upwards of 40 vascular plant species in 0.25 m^2 (Walker and Peet, 1983). Many plant species within longleaf pine ecosystems also demonstrate high endemism and rarity. Forty percent of the 1600+ plant species in the Atlantic and Gulf Coastal Plains are restricted to longleaf pine ecosystems (Walker, 1998), and Walker (1993) described 187 rare plant species associated with longleaf pine ecosystems. That count has since been updated to include an additional 256 species, reflecting the continued and increasing endangerment of longleaf pine associates

(Glitzenstein et al., 2001). Diversity of longleaf pine ecosystems manifests itself not only within communities but also between them. From xeric, rocky slopes in Alabama to hydric, sandy flatwoods in North Carolina, longleaf pine occupies a wide range of site types. This diversity among longleaf pine communities is perhaps best represented by the 135 longleaf pine associations described by Peet (2006).

Longleaf pine forests may be managed simultaneously for both timber and biodiversity (Freeman and Jose, 2009; Mitchell et al., 2006). The removal of large overstory trees, which provide high quality timber, also serves to increase light penetration into the species-rich understory (Van Lear et al., 2005). Similarly, frequent fire promotes dominance of longleaf pine (Mitchell et al., 2009), increases understory species richness (Glitzenstein et al., 2003; Walker and Peet, 1983), and creates open habitat for game species, such as bobwhite quail (*Colinus virginianus*) and white-tailed deer (*Odocoileus virginianus*), that generate hunting revenue (Landers et al., 1995). Landers et al. (1995) suggested that this pairing of economic and ecological benefits would be one of the keys to successful restoration of longleaf pine ecosystems across the landscape.

Since that assessment over two decades ago, longleaf pine restoration has been embraced across the Southeast by scientists, public agencies, and private landowners alike. Land management objectives and silvicultural practices have expanded to include conservation of biodiversity in addition to sustained timber yield (Mitchell et al., 2006). More than 20 agencies and organizations worked together in 2009 to publish a range-wide conservation plan for longleaf pine (Darden et al., 2009). The plan’s ultimate goal was to increase longleaf acreage by almost 2 million ha in 15 years. In 2010, the US Departments of Agriculture, Defense, and the Interior signed a memorandum of understanding to support that goal (Oswalt et al., 2012). Federal support for longleaf pine restoration is critical because the federal government has been able to provide financial incentives to private landowners, who own over 80% of the land in longleaf pine's historical range (Oswalt et al., 2012).

This commitment to longleaf pine restoration has underscored the need for research-based guidelines that encompass the wide array of site conditions on which restoration may take place. The range of site types that longleaf pine can occupy, paired with the variety of pre-restoration conditions of those sites, means that site-specific restoration approaches may be needed. Ultimately, though, the final goals of these approaches tend to be structurally and compositionally similar because they all seek to restore a complete, fully functioning longleaf pine ecosystem. To better understand what restoration entails, it is important to understand the structure, composition, and function of the longleaf pine ecosystem.

Longleaf Pine Ecosystem Description

Ecosystem Structure and Composition

The longleaf pine ecosystem is characterized by an open overstory dominated by longleaf pine, a sparse midstory containing little more than ascending longleaf pine, and an understory rich in grasses, forbs, and shrubs (Brockway et al., 2005). While this framework is straightforward, the natural variation among longleaf pine communities is wide. Structurally, longleaf pine can exist naturally in both even-aged and uneven-aged stands, and these stands can range from closed canopy forests to open savannas (Landers et al., 1995). Compositionally, understory species vary widely due to the expansive geographic extent of longleaf pine, the gradient of soil moisture and texture on which longleaf pine communities exist, and the frequency with which communities are burned (Frost, 2001; Peet, 2006).

Overview of Site Types

The most recent comprehensive description of longleaf pine vegetation was developed by Peet (2006), who classified longleaf pine communities into six main ecological groups based upon soil type and vegetation cover. These six groups are (1) xeric sand barrens and uplands, (2) subxeric sandy uplands, (3) silty uplands, (4) clayey and rocky uplands, (5) flatwoods, and (6) savannas and seeps.

Xeric sand barrens and uplands are characterized by well-drained sand and consist of scattered overstory longleaf pine with an oak understory. The open ground layer may contain some grass and low shrubs. Subxeric sandy uplands have topographic relief and deep, sandy soils that are consistently dry. Upland oak species are common in the understory, and the ground layer is dominated by continuous grass cover. Silty uplands are well-drained sites with high herbaceous diversity, a ground layer dominated by bluestem (*Andropogon* spp. and *Schizachyrium* spp.) grasses, and oaks present beneath the longleaf pine canopy. Silty uplands with natural longleaf pine vegetation are rare because most of them have been converted to agricultural land. Clayey and rocky uplands contain longleaf pine on exposed ridges and south-facing slopes. Shortleaf pine (*Pinus echinata* Mill.), oaks, and other upland hardwoods are found on these sites, and the herbaceous layer is dominated by wiregrass (*Aristida stricta* Michx.). Flatwoods are characterized by low, flat terrain and poorly drained, sandy soils. Wiregrass, bracken fern (*Pteridium aquilinum* (L.) Kuhn), and saw palmetto (*Serenoa repens* (W. Bartram) Small) may dominate the understory along with a diverse array of forbs. Savannas and seeps occur on seasonally saturated, fine-textured soils and have a very open canopy dominated by longleaf pine or other southern pines. These sites are extremely diverse, featuring an array of grasses and forbs, including numerous orchids (e.g., *Calopogon, Cleistes, Platanthera, Pogonia, Spiranthes*), lilies (e.g., *Aletris, Lilium, Tofieldia, Zigadenus*), and insectivorous plants (e.g., *Drosera, Dionaea, Pinguicula, Sarracenia, Utricularia*).

Flatwoods and Savannas

The study that is the focus of this thesis took place on sites that fit Peet's (2006) descriptions of both flatwoods and savannas, so these vegetation communities will be reviewed in greater depth than the other four. Although “flatwoods” and “savannas” are terms that can have many meanings, Peet (2006) provided precise definitions for these community types. Flatwoods are fire-maintained pine woodlands over flat, Coastal Plain landscapes, and savannas are open pine woodlands on seasonally saturated, fine-textured soils. Clearly, there is room for overlap between these two ecological groups.

Both flatwoods and savannas may have understories that contain a diverse collection of forbs, most notably insectivorous plants (Peet, 2006). These two communities are also similar because wiregrass may be a dominant feature in flatwoods, and savannas are characterized by a grassy understory (Peet, 2006). However, the primary difference between the two communities is that flatwoods often have a high proportion of shrubs (Peet, 2006). Across the geographic range of flatwoods, shrubs in the heath family (e.g., *Gaylussacia*, *Lyonia*, *Vaccinium*) are common, and along the southern Atlantic and eastern Gulf Coastal Plains, saw palmetto often dominates the understory (Peet, 2006). Variations in fire frequency might further obscure the differences between flatwoods and savannas. Glitzenstein et al. (2003) hypothesized that several decades of reduced fire frequency may convert wet, grass-dominated savannas into wet, shrub-dominated flatwoods. This hypothesis relates closely to the concept of alternative community states as explored by Martin and Kirkman (2009), who suggested that depending upon a site's fire regime, it will exhibit one of two community states (species-rich herbaceous community or hardwood dominated community with little groundcover). Martin and Kirkman (2009) discussed how alternative state theory ties into restoration, which will be explored later in this review.

From a soil standpoint, the study sites align closely with flatwoods because they are situated on spodic soils. Spodosols are the primary soil order of flatwoods and are characterized by their sandy, acidic, and infertile nature (Brockway et al., 2005; Peet, 2006). Specifically, the study sites are located on Leon sand (sandy, siliceous, thermic Aeric Alaquod), a poorly drained Spodosol with a fluctuating water table that may be at or near the surface during the summer rainy season (Barnhill, 1992; NRCS, 2014). Leon sand is one of the most extensive soil series on Camp Lejeune, the military installation on which this study’s sites were located, (Barnhill, 1992; Frost, 2001) and is of large extent throughout the Southeast (NRCS, 2014).

From a vegetation standpoint, the line between savanna and flatwoods is less clear on the study sites. Frost (2001) extensively mapped pre-settlement vegetation and fire regimes on Camp Lejeune. Frost's (2001) work indicated that the pre-settlement community on Leon sand would have been a diverse, wet savanna dominated by longleaf pine in the overstory and wiregrass in the understory. The understory would also have contained a rich collection of other grasses, sedges, forbs, and fire-dwarfed shrubs. Like the savannas that Peet (2006) described, these sites would have featured a number of insectivorous plants, including sundews (*Drosera* spp.), pitcher plants (*Sarracenia* spp.), and Venus flytrap (*Dionaea muscipula* Ellis). Today, Camp Lejeune supports the world’s largest remaining population of Venus flytraps, which can only be found where fires burn frequently. In fact, Frost (2001) suggested that Venus flytrap is the most fire dependent species known because populations disappear when the fire return interval exceeds three years. This is just one of many examples demonstrating how frequent fire shapes the longleaf pine ecosystem.

Longleaf Pine Ecosystem Function

Fire’s Role in the Ecosystem

Fire is arguably the most important ecological process in the longleaf pine ecosystem. Frequent fire is associated with high species richness in longleaf pine communities (Glitzenstein et al., 2003; Walker and Peet, 1983), and an estimated 65% of native rare plants and animals in the Southeast are dependent upon fire to create or maintain their habitat (Frost, 2001). Accordingly, Mitchell et al. (2006) suggest that fire frequency is the most important factor for sustaining native Southeastern ecosystems. Fire exclusion in the 20th century was one of the primary causes of the decline of longleaf pine ecosystems (Frost, 2006), and returning fire to the landscape is recognized as one of the critical features of longleaf pine restoration (Van Lear et al., 2005).

Longleaf pine and bunchgrasses together facilitate frequent surface fire, which results in a positive feedback loop as frequent fire promotes dominance of both longleaf pine and bunchgrasses (Brockway et al., 2005; Mitchell et al., 2009). The typical bilayered structure of longleaf pine communities is a result of this frequent fire regime. The tight coupling of fuels and fire goes both ways, though, because fire exclusion creates a positive feedback of its own (Martin and Kirkman, 2009). Without fire, a dense midstory develops that, in turn, alters the fire regime making fire less frequent and more severe (Mitchell et al., 2006).

Fire impacts not only the community's structure but also its composition. Frequent burning promotes understory species richness by limiting the growth of woody plants (Martin and Kirkman, 2009) and reducing grass and sedge foliage that shade out smaller grasses and forbs (Walker and Peet, 1983). It has been generally assumed that the relationship between fire and richness is mediated by light availability, with frequent fire increasing light penetration to the understory (Van Lear et al., 2005). However, recent studies suggest that shade cast by shrubs and hardwoods may not be the true mechanism that limits species richness in the absence of fire. Instead, accumulation of leaf litter and development of organic soil horizons in the absence of fire are the factors that reduce understory diversity (Hiers et al., 2007; Veldman et al., 2014). Regardless of the mechanism, the importance of fire in maintaining natural longleaf pine vegetation communities cannot be overstated.

Longleaf Pine Fire Adaptations and Natural History

Longleaf pine, too, benefits from this disturbance because it tolerates frequent fire better than its competitors. A novel fire adaptation of longleaf pine is the tree's grass stage. After germination, seedlings remain stemless, making them highly resistant to fire-induced mortality (Boyer, 1990). This fire resistance occurs because the seedling's root collar is kept at the soil surface and the apical meristem is protected by bud scales and a thick tuft of needles (Wang et al., 2016). The stemless grass stage can last for 2-20 years, during which seedlings develop extensive root systems and build up carbohydrate reserves that will later be utilized for rapid height growth (Brockway et al., 2006). This rapid growth stage, known as "bolting," is thought to minimize the amount of time that seedlings are susceptible to damage by surface fire (Boyer, 1990).

A recent study by Wang et al. (2016) found that rapid height growth was not an important factor for survival of longleaf pine regeneration. After emergence from the grass stage, it took 3-4 years for seedlings to reach a critical height of 90 cm, below which they would be susceptible to damage from fire. This height growth was no faster than loblolly pine (*Pinus taeda* L.), even though longleaf pine seedlings had much higher survival than loblolly pine under a regime of frequent surface fire (Wang et al., 2016). Although this study contradicts one of the accepted mechanisms by which longleaf pine seedlings survive frequent fire, it supports the fact that they are resistant to fire-induced mortality. A third critical fire adaptation of longleaf pine is its thick bark (Brockway et al., 2005), which could be the mechanism that explains the findings of Wang et al. (2016).

When frequent fire is absent, longleaf pine struggles to regenerate. Difficulties regenerating longleaf pine are a major reason for the tree’s historical decline (Brockway et al., 2005), and the challenges associated with establishing longleaf pine remain a primary barrier to restoration (Johnson and Gjerstad, 2006). Longleaf pine seed production is erratic, its large seeds have limited dispersal, and seedling survival is often low (Brockway et al., 2006; Landers et al., 1995). Perhaps the greatest impediment to successful regeneration is the tree’s intolerance to competition (Boyer, 1990; Brockway et al., 2006). Grass stage seedlings are particularly sensitive to competition, with high levels of competition resulting in later emergence from the grass stage (Boyer, 1990).

These natural history characteristics hold true across site types, but hydric sites, such as flatwoods, amplify the challenges associated with regeneration. Competition from woody plants is especially abundant on wet sites, and poor soil drainage can inhibit seedling growth and survival (Brockway et al., 2006). In order to overcome these challenges, longleaf pine regeneration must be accompanied by some form competition control (Brockway et al., 2006). Longleaf pine would have naturally regenerated on hydric sites under a regime of frequent surface fire, and prescribed fire is often a suitable solution because of the natural relationship between frequent fire and competition reduction. However, sites with heavy competition from woody plants and a history of fire suppression will often require mechanical or chemical site preparation to successfully establish planted longleaf pine seedlings (Brockway et al., 2006).

Longleaf Pine Site Preparation

Overview of Site Preparation

Due to the widespread reduction in areal extent of longleaf pine, restoration efforts often occur on sites where mature longleaf pine is no longer present or is a small component of the overstory. In such cases, artificial regeneration is necessary to reestablish longleaf pine. This practice has become so common that over a quarter of the modern longleaf pine forest – almost 450,000 ha – was initiated by planting (Oswalt et al., 2012). Artificial regeneration can take two forms: direct seeding or planting seedlings. Generally, planting seedlings is more viable because the cost of seed for successful regeneration is higher than the cost of seedlings (Johnson and Gjerstad, 2006). There are also two types of seedlings: bareroot or container-grown. In a synthesis of over 20 studies comparing the two seedling types, South et al. (2005) found that average survival of container-grown seedlings was over 22% greater than that of bareroot seedlings. This difference tends to be greater on adverse sites (South et al., 2005), which supports Boyer's (1988) suggestion that container-grown seedlings may be more resistant to environmental stresses such as drought, competition, poor planting, and herbicide exposure. Therefore, container-grown seedlings are typically the most desirable method of artificial regeneration for longleaf pine restoration.

Preparation of a site prior to planting is critical for successful longleaf pine seedling establishment (Brockway et al., 2006). Primary objectives of site preparation include controlling competing vegetation and ameliorating adverse soil conditions in order to enhance early survival and growth of planted seedlings (Lowery and Gjerstad, 1991). It is helpful to categorize the wide variety of site preparation techniques that exist into two main categories based upon their primary purpose: vegetation control or soil manipulation (Morris and Lowery, 1988). Site preparation treatments designed to reduce competing vegetation can be either chemical or mechanical and include herbicide application, chopping, and shearing (Johnson and Gjerstad, 2006). Bedding, mounding, and disking are mechanical treatments that may also provide some vegetation control, but their primary objective is to manipulate the soil to overcome adverse soil conditions, such as poor drainage and a high water table (Johnson and Gjerstad, 2006). Prescribed fire is often used in combination with one or more of these treatments to further control competition and remove debris prior to planting (Lowery and Gjerstad, 1991). The study that is the focus of this thesis studied the effects of two soil manipulation treatments (bedding and mounding) and two vegetation control treatments (chopping and herbicide), so each of these treatments will be explored in greater depth.

Tree Responses to Soil Manipulation Treatments

Although bedding and mounding are different techniques that are commonly utilized in different regions, their objectives are quite similar. Bedding is the formation of a continuous low ridge, or bed, of soil using a plow or other similar machinery (Lowery and Gjerstad, 1991). This site preparation technique is commonly used on flat poorly drained sites in the Southeast (Johnson and Gjerstad, 2006). Unlike bedding, mounds are not continuous. Instead, an excavator is used to scoop soil and deposit it into discontinuous mounds across the site (Londo and Mroz, 2001). Mounding is regularly used in the Lake States region and is designed to imitate the pit and mound microtopography that is common in wetlands throughout that region (Londo and Mroz, 2001).

The objectives of mounding and bedding are to improve survival and growth of planted seedlings on wet sites by improving soil drainage, raising soil temperatures, and incorporating organic matter into the soil (Johnson and Gjerstad, 2006; Londo and Mroz, 2001; Lowery and Gjerstad, 1991). Soils on top of beds and mounds are drier, better aerated, and quicker to warm in the spring, all of which promote a good rooting environment for planted seedlings (Johnson and Gjerstad, 2006; Londo and Mroz, 2001; Lowery and Gjerstad, 1991). An ancillary benefit of these soil manipulation treatments is that they provide some degree of vegetation control by damaging root stocks of residual woody vegetation (Johnson and Gjerstad, 2006). However, there are concerns that intense site preparation treatments like these may negatively impact herbaceous vegetation (Brockway et al., 2005; Johnson and Gjerstad, 2006), which is a drawback that will be explored in a later section.

Due to the widespread practice of intensely managing southern pine plantations, the effects of bedding – and most other site preparation treatments – have been studied much more extensively on commercially grown southern pines, such as loblolly and slash (*Pinus elliottii* Engelm.), than on longleaf pine. For slash pine seedlings planted on hydric sites, bedding has been shown to improve both survival (Pritchett, 1979) and growth (Outcalt, 1984; Pritchett, 1979). Pritchett (1979) found that seedling survival was three times greater and mean height was 1.25 m taller on bedded plots compared to controls eight years after planting. Similarly, Outcalt (1984) found that high beds (38 cm above ground line) produced a greater mean height and diameter than low beds (15 cm above ground line) at age 10; however, by age 15 those differences no longer existed. One study that did explore the effects of bedding on longleaf pine, albeit on a well-drained site, found that bedding reduced survival of seedlings by 11% after the first year (Loveless et al., 1989). However, bedding did hasten grass stage emergence, resulting in 25% more trees in height growth in the second year, and increased seedling height by almost 0.5 m after four years of growth (Loveless et al., 1989).

There is a paucity of studies that examine the response of southern pines to mounding because the technique was developed in a different region and, thus, is utilized much less frequently than bedding. Theoretically, though, mounding is a logical technique to apply to flat, poorly drained sites in the Southeast and presents a potential alternative to bedding. Haywood (1987) studied the response of slash pine on a flatwoods site to two mounding treatments – high (75 cm above ground line) and low (32 cm above ground line) – and an untreated control. Both mounding treatments performed similarly, and there was no difference in survival among treatments. However, mounding did improve seedling root collar diameter by 38% and seedling height by 25% compared to the untreated control.

The study that is the focus of this thesis is a follow up to research conducted by Knapp et al. (2006, 2008), and these earlier studies serve as a helpful reference for how site preparation impacted early survival and growth of the longleaf pine seedlings. Seedling survival through 20 months after planting did not differ among the three soil manipulation treatments: bedding, mounding, and a flat-planted control (Knapp et al., 2006). However, there were differences among the treatments in seedling growth. Bedding (19.8 mm, 8.1%) and mounding (18.6mm, 8.6%) produced similar mean values for root collar diameter and percentage of seedlings in height growth (> 15 cm tall), and both treatments outperformed the flat-planted control (15.7 mm, 2.2%). Additionally, Knapp et al. (2008) found that both bedding and mounding resulted in a reduction in soil moisture at 6 cm and an increase in soil temperature at 15 cm relative to the control. These findings suggest that two of the hypothesized benefits of soil manipulation treatments, namely drier and warmer soil, did in fact play a role in promoting growth of seedlings.

Tree Responses to Vegetation Control Treatments

Vegetation control treatments for site preparation can be applied mechanically or chemically. There are numerous mechanical approaches to vegetation control and even more choices of herbicide, but the focus of this review will be narrowed to the two treatments utilized in this study: chopping and herbicide application (of imazapyr and triclopyr). Chopping, which is also referred to as crushing, utilizes a heavy metal drum ribbed with blades to chop stems and roots of vegetation (Lowery and Gjerstad, 1991). This technique is desirable because it causes minimal soil disturbance, has a low impact on herbaceous vegetation, and allows fuels to dry out before a fire (Johnson and Gjerstad, 2006). Although chopping immediately reduces standing woody vegetation, it often promotes sprouting of woody growth, which can be problematic for successful establishment of planted seedlings (Lowery and Gjerstad, 1991). Lowery and Gjerstad

(1991) suggested that the reduced height and diameter of woody sprouts facilitate a follow-up treatment directly after chopping, and Johnson and Gjerstad (2006) even recommended chopping a second time in later summer or early fall when carbohydrate reserves have diminished.

The use of chemical vegetation control has increased in recent decades because herbicide application typically does not result in sprouting of woody competition (Lowery and Gjerstad, 1991). Other advantages of herbicides include relatively low cost of application, minimal soil disturbance, high increases in tree productivity, and the ability of herbicides to target specific plant groups (Johnson and Gjerstad, 2006; Lowery and Gjerstad, 1991; Walker and Silletti, 2006). Imazapyr, which is one of the herbicides used in this study, targets broadleaf plants, hardwoods, and grasses and is commonly used in longleaf pine restoration because of its versatility across soil types (Addington et al., 2012; Johnson and Gjerstad, 2006). The other herbicide utilized in this study is triclopyr, which targets hardwoods and broadleaf weeds and effectively reduces waxy species common in the Coastal Plain (Johnson and Gjerstad, 2006; Lowery and Gjerstad, 1991). Just like chopping commonly precedes other site preparation treatments, herbicide application is almost always followed by a prescribed fire because burning further stresses targeted species and clears debris (Lowery and Gjerstad, 1991).

The widespread acceptance of herbicide application as a form of site preparation has resulted in a sizeable amount of research exploring the response of longleaf pine to herbicide treatment. Some studies have found that longleaf pine seedling survival is not affected by herbicide application (Addington et al., 2012; Knapp et al., 2006; Loveless et al., 1989), while others have observed reduced survival following herbicide (Boyer, 1988; Freeman and Jose, 2009). On the other hand, studies have consistently observed increased longleaf pine seedling growth when herbicide is applied prior to planting (Addington et al., 2012; Boyer, 1988; Freeman and Jose, 2009; Knapp et al., 2006). Both Boyer (1988) and Freeman and Jose (2009) found that herbicide treatment reduced time spent in the grass stage. Of the four herbicides that Freeman and Jose (2009) tested, imazapyr showed the highest percentage of seedlings in height growth (> 12 cm tall) relative to the control (65% vs. 30%). Addington et al. (2012) tested a mix of imazapyr and glyphosate, which increased longleaf pine seedling mean root collar diameter by almost 2 cm and height by over 1 m, relative to the untreated control.

Unlike herbicides, there has been relatively little research conducted studying the effects of chopping. A study by Miller (1980) tested the effects of two vegetation control treatments – chopping and windrowing (also called shearing and piling) – on a loblolly pine plantation. Miller (1980) found that windrowing reduced woody vegetation by 55% compared to chopping two years after treatment application. However, soil erosion was still apparent two years after windrowing, while no such erosion was present on the chopping treatment. The poor vegetation control of chopping may be due to the fact that only a single pass of the chopper was applied. Boyer (1988) found that a second pass of chopping or harrowing resulted in significantly higher survival of longleaf pine seedlings three years after treatment application compared to a single pass (73% vs. 58%). Two passes also resulted in a greater number of seedlings in height growth after two years, but by the end of the third year there was no longer a difference between one and two passes.

The previous study from Knapp et al. (2006) is unique from the rest of the literature because it directly compares chopping and herbicide. Longleaf pine seedling survival was statistically similar between chopping and herbicide 20 months after planting. However, the herbicide treatment resulted in greater root collar diameter and percentage of seedlings in height growth (19.0 mm, 9.0%) compared to chopping (17.0 mm, 3.6%).

Vegetation Responses to Site Preparation

Restoration of the longleaf pine ecosystem includes not only establishment of a longleaf pine overstory but also restoration of the ground layer plant community (Walker and Silletti, 2006). In addition to promoting survival and growth of longleaf pine seedlings, site preparation may also enhance the plant community by reducing woody species that would otherwise decrease herbaceous richness and diversity (Litt et al., 2001). However, there is widespread concern that intense mechanical site preparation treatments, such as bedding and mounding, may damage native understory plants (Brockway et al., 2005, 2006; Mitchell et al., 2006; Platt et al., 2015; Van Lear et al., 2005). Therefore, Johnson and Gjerstad (2006) suggest that when restoration of the entire longleaf pine ecosystem is the objective, there must be balance between maximizing site preparation that promotes seedling establishment and minimizing detrimental impacts of those treatments on native flora and fauna. Van Lear et al. (2005) argue that, ultimately, the benefits of active management far outweigh the cost of doing nothing and that without such active management, the longleaf pine ecosystem is doomed.

There has been a fair amount of research on the effects of site preparation on plant communities within longleaf pine ecosystems. Bedding and mounding have been shown to reduce grass cover (Knapp et al., 2008; Pritchett, 1979), which is concerning because of the critical role that grasses play in carrying surface fire through longleaf pine communities. Knapp et al. (2008) found that mounding had even more intense impacts than bedding. On mounds, cover of graminoids was less than a third and cover of shrubs less than half of that on the control. In terms of vegetation control treatments, Knapp et al. (2008) found that herbicide resulted in a greater reduction of shrub cover than did chopping, while graminoid and forb cover were similar for both treatments. Herbicide treatments also successfully reduced hardwood stem density and height through six years after application in a study from Fort Benning, GA (Addington et al., 2012). However, unlike the other studies mentioned, herbaceous vegetation, including bunchgrasses, was not negatively impacted by herbicides, and species richness was similar among herbicide treatments and the control. It is important to note that the effectiveness of and potential side effects from herbicide treatments are influenced by a number of factors, including the type of chemical used, application method (e.g., broadcast vs. spot-spray) and application rate. Therefore, it can be challenging to find commonality of results across many studies (Litt et al., 2001).

Freeman and Jose (2009) found that species richness, herbaceous cover, and wiregrass cover were higher on plots treated with imazapyr than on untreated controls four years after application. In the same study, early shrub control provided by herbicide was no longer detectable after four years, but Freeman and Jose (2009) suggested that the initial woody reduction was key for allowing herbaceous plants to establish and persist even after shrubs rebounded. This supports the conceptual framework of restoration thresholds put forth by Martin and Kirkman (2009). When attempting to restore one community state to another (e.g., from hardwood-dominated to species-rich herbaceous community), intensive intervention early on may be necessary to overcome a threshold between the community states. Once that initial threshold is overcome (i.e., the positive feedback between fine fuels and frequent fire is re-established), then regular prescribed fire is the only treatment needed to maintain the restored, desired community state (Martin and Kirkman, 2009). This framework is helpful to consider when evaluating how long-term restoration can be achieved through early site preparation treatments.

Long-Term Effects of Site Preparation

Investment in site preparation assumes that treatments will result in long-term benefits to stand establishment, yet many studies on site preparation effects report on short-term responses within a few years after treatment. For longleaf pine, this may not even be enough time to know if seedlings will emerge from the grass stage. Moreover, the trajectory of responses to site preparation may change through time. Three possible long-term growth responses to site preparation have been described (Morris and Lowery, 1988; Nilsson and Allen, 2003). Type A response occurs when growth gains from initial treatment continue to increase over time. Type B response occurs when initial growth gains from the treatment are maintained through time but do not continue to increase. Type C response occurs when growth gains from initial treatment are subsequently lost. Nilsson and Allen (2003) described a fourth trajectory, Type D, which occurs when growth of an untreated stand surpasses that of the treated stand. It is not clear how site preparation affects longleaf pine stand development over the long term. Short-term studies have generally found that site preparation improves growth, but not survival, of planted longleaf pine. However, some studies (e.g., Boyer, 1988; Outcalt, 1984) have found that growth responses to site preparation diminished through time. Given the unique grass stage growth form of longleaf pine seedlings, it is not known if short-term benefits of site preparation result in earlier grass stage emergence followed by sustained improvements in growth or if growth patterns following grass stage emergence are independent of site preparation treatment. These uncertainties illustrate the importance of monitoring long-term responses to site preparation.

Longleaf pine ecosystem restoration includes restoration of both the longleaf pine overstory and the native, ground-layer plant community (Walker and Silletti, 2006). In traditional plantation silviculture, site preparation is intended to maximize survival and growth of planted seedlings. However, maximized growth of planted longleaf pine seedlings may not be necessary for restoration. Instead, site preparation may only be needed to promote sufficient grass stage emergence to create a future stand of a desired structure (e.g., open savanna or closed-canopy forest). Understory plant communities are also expected to change through time, so for restoration it would be desirable if they moved toward a specified reference or desired condition (Kirkman et al., 2013). This reference condition will vary depending upon site type and geographic location, but re-establishment of frequent fire is a consistent objective for restoring ground flora in longleaf pine communities (Martin and Kirkman, 2009). Therefore, site preparation treatments must sustain a frequent fire regime that is continuous in both time and space (Mitchell et al., 2009).

Conclusion

Areas for Future Research

The lack of long-term studies on longleaf pine and ground flora responses to site preparation highlights a major research need. As discussed in the previous section, short-term increases in seedling growth from site preparation may diminish over time (Morris and Lowery, 1988; Nilsson and Allen, 2003), so longer-term studies will provide a more accurate representation of long-term treatment success. An accurate evaluation of site preparation is especially critical for wet sites and sites with soil resource limitations, for which there is a general lack of guidelines for longleaf pine establishment (Oswalt et al., 2012). There is also a lack of research exploring long-term responses of ground layer plant communities to site preparation treatments (Addington et al., 2012; Litt et al., 2001; Walker and Silletti, 2006). Oswalt et al. (2012) cite the need for more research evaluating the influence of silvicultural treatments on the understory and the impact of herbicides on nontarget herbaceous species. To that end, Litt et al. (2001) emphasized the lack of studies that report findings on responses of individual species of special concern, such as wiregrass. Long-term data on site preparation responses in hydric flatwoods sites will be critical for a wholistic evaluation of restoration success.

Project Overview

This study served as a longer-term follow up to a project initiated in 2003 that examined the responses of planted longleaf pine seedlings and ground layer vegetation to site preparation treatments. The initial project (Knapp et al., 2008, 2006) was novel because of the breadth and intensity of treatments tested. Few studies, if any, have tested the effects of three levels of soil manipulation treatments (including a flat-planted control) and two levels of vegetation control treatments on both longleaf pine seedlings and ground layer vegetation. Additionally, the project's emphasis on planting longleaf pine for restoration, rather than for maximized productivity, on wet, flatwoods sites set it apart from previous research on site preparation for longleaf pine. Measurements for the initial study ceased after three growing seasons, yet questions remained about how longleaf pine and ground layer vegetation respond to site preparation treatments in the longer term.

This study sought to answer those questions by returning to the study sites and re-measuring the planted longleaf pine and surrounding ground flora. In Chapter 2 we report results on the effects of site preparation on longleaf pine stand development through 15 years after planting. This includes both tree-level and stand-level responses to site preparation treatments. In Chapter 3 we present results on ground-layer vegetation responses to site preparation through 15 years after treatment application. Both chapters will integrate data from the initial study to quantify the effect of time and to determine whether early responses to site preparation during the seedling establishment phase were maintained into stand development.

Chapter 2: Long-Term Longleaf Pine Responses to Site Preparation for Restoration on Hydric Sites

Introduction

Restoration of longleaf pine (*Pinus palustris* Mill.) ecosystems is an important land management objective throughout the southeastern United States. Prior to European settlement, longleaf pine occupied an estimated 37 million hectares (Frost, 2006). By 2010, one of the most extensive forest ecosystems in North America had been reduced to approximately 4.5% of its pre-settlement range (Oswalt et al., 2012). This reduction was due to a number of anthropogenic changes after European settlement, including widespread timber harvest and turpentine production, land conversion for urban and agricultural uses, grazing by hogs and other livestock, and, perhaps most importantly, fire exclusion (Frost, 2006; Landers et al., 1995).

In addition to the dramatic reduction in areal extent of longleaf pine, its economic and ecological values are driving interest in restoration. Longleaf pine produces high quality timber, abundant pine straw, and ideal habitat for hunting (Brockway et al., 2005; Landers et al., 1995). Moreover, longleaf pine is fire tolerant, can thrive on harsh sites, and is more resistant to insects and disease and less susceptible to damage from ice and wind than other southern pines (Johnson and Gjerstad, 2006; Landers et al., 1995; Mitchell et al., 2006). From an ecological perspective, longleaf pine ecosystems include some of the most diverse plant communities outside of the tropics (Walker and Silletti, 2006), with the richest savannas containing upwards of 40 vascular plant species in 0.25 m^2 (Walker and Peet, 1983). Many plant species within longleaf pine ecosystems also demonstrate high endemism and rarity. Forty percent of the 1600+ plant species in the Atlantic and Gulf Coastal Plains are restricted to longleaf pine ecosystems (Walker, 1998), and 443 rare plant species are associated with longleaf pine (Glitzenstein et al., 2001; Walker, 1993). Longleaf pine forests may be managed simultaneously managed for both timber and biodiversity (Freeman and Jose, 2009; Mitchell et al., 2006). This pairing of economic and ecological benefits has been critical to increasing interest in longleaf pine restoration across the Southeast (Landers et al., 1995; Mitchell et al., 2006).

Widespread interest in longleaf pine restoration has underscored the need for research-based guidelines that encompass the wide array of site conditions on which restoration may take place. The range of site types that longleaf pine can occupy, paired with the variety of pre-restoration conditions of those sites, means that site-specific restoration approaches may be needed. Restoration efforts often occur on sites where mature longleaf pine is no longer present or is a small component of the overstory. In such cases, artificial regeneration is necessary to reestablish longleaf pine on the site. This practice has become so common that over a quarter of the modern longleaf pine forest – almost 450,000 ha – was regenerated artificially (Oswalt et al., 2012).

A major impediment to successful establishment of longleaf pine on any site is its intolerance to competition, particularly as a grass stage seedling (Boyer, 1990). This challenge is amplified on wet sites, such as flatwoods, where competition from woody plants is especially abundant. Poor soil drainage can also inhibit seedling growth and survival (Brockway et al., 2006). In order to overcome these challenges, longleaf pine regeneration must be accompanied by some form competition control (Brockway et al., 2006). Longleaf pine would have naturally regenerated on hydric sites under a regime of frequent surface fire, and prescribed fire is often a suitable solution because of the natural relationship between frequent fire and competition reduction. However, sites with heavy competition from woody plants and a history of fire suppression will often require site preparation to successfully establish planted longleaf pine seedlings (Brockway et al., 2006).

Site preparation techniques can be divided into two categories based upon their primary objective: vegetation control or soil manipulation (Morris and Lowery, 1988). Treatments designed to reduce competing vegetation include herbicide application and mechanical chopping (Johnson and Gjerstad, 2006). Bedding and mounding are mechanical treatments that may also provide some vegetation control (Johnson and Gjerstad, 2006). However, their primary objectives are to manipulate the soil to overcome adverse conditions, such as poor drainage and a high water table, and to improve the rooting environment for planted seedlings by increasing soil temperature and aeration (Johnson and Gjerstad, 2006; Londo and Mroz, 2001; Lowery and Gjerstad, 1991).

Due to the intense management of southern pine plantations, the effects of bedding – and most other site preparation treatments – have been studied more extensively on commercially grown southern pines, such as loblolly (*Pinus taeda* L.) and slash (*Pinus elliottii* Engelm.), than on longleaf pine. Bedding has been shown to increase survival of slash pine seedlings planted on hydric sites (Pritchett, 1979) but not of longleaf pine seedlings planted on well-drained sites (Loveless et al., 1989) or poorly-drained flatwoods (Knapp et al., 2006). Mounding has been insufficiently studied in the Southeast because it is used more commonly in wetlands of the Lake states region (Londo and Mroz, 2001). However, mounding has not been shown to increase survival of slash (Haywood, 1987) or longleaf pine seedlings (Knapp et al., 2006) planted on flatwoods sites. Bedding and mounding have been shown to improve growth of slash pine (Haywood, 1987; Outcalt, 1984; Pritchett, 1979) and longleaf pine seedlings (Knapp et al., 2006; Loveless et al., 1989).

The use of chemical vegetation control has increased in recent decades because, unlike mechanical treatments, herbicide application typically does not result in sprouting of woody competition (Lowery and Gjerstad, 1991). This widespread acceptance has resulted in a sizeable amount of research exploring the response of longleaf pine to herbicide treatment. Some studies have found that longleaf pine seedling survival is not affected by herbicide application (Addington et al., 2012; Knapp et al., 2006; Loveless et al., 1989), while others have observed reduced survival following herbicide (Boyer, 1988; Freeman and Jose, 2009). On the other hand, studies have consistently observed increased longleaf pine seedling growth when herbicide is applied prior to planting (Addington et al., 2012; Boyer, 1988; Freeman and Jose, 2009; Knapp et al., 2006).

Unlike herbicides, there has been little research conducted studying the effects of chopping. Knapp et al. (2006) found that chopping resulted in reduced root collar diameter growth and a lower proportion of grass stage emergence than herbicide. Miller (1980) found that windrowing, an alternative vegetation control treatment, reduced woody vegetation by 55% compared to chopping. The poor performance of chopping in both studies may be due to the fact that only a single pass of the chopper was applied. Boyer (1988) found that a second pass of chopping or harrowing resulted in higher survival of longleaf pine seedlings and increased grass stage emergence compared to a single pass.

Investment in site preparation assumes that treatments will result in long-term benefits to stand establishment, yet many studies on site preparation effects report on short-term responses within a few years after treatment. For longleaf pine, this may not even be enough time to know if seedlings will emerge from the grass stage. Moreover, the trajectory of responses to site preparation may change through time. Three possible long-term growth responses to site preparation have been described (Morris and Lowery, 1988; Nilsson and Allen, 2003). Type A response occurs when growth gains from initial treatment continue to increase over time. Type B response occurs when initial growth gains from the treatment are maintained through time but do not continue to increase. Type C response occurs when initial growth gains from the treatment are subsequently lost. Nilsson and Allen (2003) described a fourth trajectory, Type D, which occurs when growth of an untreated stand surpasses that of the treated stand. It is not clear how site preparation affects longleaf pine stand development over the long term. Short-term studies have generally found that site preparation improves growth, but not survival, of planted longleaf pine. However, some studies (e.g., Boyer, 1988; Outcalt,

1984) have found that growth responses to site preparation diminished through time. Given the unique grass stage growth form of longleaf pine seedlings, it is not known if short-term benefits of site preparation result in earlier grass stage emergence followed by sustained improvements in growth or if growth patterns following grass stage emergence are independent of site preparation treatment. These uncertainties illustrate the importance of monitoring long-term responses to site preparation.

The objective of this study was to quantify the long-term responses of planted longleaf pine to site preparation treatments on poorly drained, hydric sites. The study served as a 15-year follow up to an earlier study by Knapp et al. (2006, 2008) that monitored seedling growth and survival through three years after planting. Using data collected during the phases of seedling establishment and stand development, we sought to address three specific questions: (1) How does site preparation affect longleaf pine stand-level size and density 15 years following planting? (2) Are early gains obtained from site preparation maintained beyond the seedling establishment phase and into stand development? (3) Are stand-level differences in tree size and density among treatments the result of earlier emergence from the grass stage, increased growth rates following emergence from the grass stage, or a combination of the two?

Methods

Study Area

This study was conducted on Marine Corps Base Camp Lejeune (34°36'N 77°24'W) in Onslow County, North Carolina. Camp Lejeune is located within the Atlantic Coastal Flatwoods section of the Outer Coastal Plain Mixed Forest province (McNab et al., 2007). The study sites were located on Leon sand (sandy, siliceous, thermic Aeric Alaquod), a poorly drained Spodosol with a fluctuating water table that may be at or near the surface (Barnhill, 1992; NRCS, 2014). Spodosols are the primary soil order of flatwoods and are characterized by their sandy, acidic, and infertile nature (Brockway et al., 2005; Peet, 2006). Leon sand is one of the most extensive soil series on Camp Lejeune (Barnhill, 1992; Frost, 2001) and is of large extent throughout the Southeast (NRCS, 2014). Timber production has been a primary land use objective throughout Camp Lejeune's history (MCBCL, 2015), and the sites selected for this study previously contained planted slash and loblolly pine, which were harvested six months to two years prior to treatment installation.

The natural vegetation community on frequently burned Leon sand in this area is species-rich longleaf pine wet savanna (Frost, 2001). This bilayered community consists of an overstory dominated by longleaf pine and an understory containing a diverse array of grasses, sedges, forbs, and fire-dwarfed shrubs (Frost, 2001). Wiregrass (*Aristida stricta* Michx.) dominates the herbaceous layer, and bluestems (*Andropogon* spp. and *Schizachyrium* spp.) are also common. The estimated pre-settlement fire return interval was 1-3 years, and with such frequent fire Leon sand supports rare species such as roughleaf loosestrife (*Lysimachia asperulifolia* Poir.) and Venus flytrap (*Dionaea muscipula* Ellis) (Frost, 2001). These sites also typically contain other insectivorous plants, such as pitcher plants (*Sarracenia* spp.) and sundews (*Drosera* spp.). Common shrubs include inkberry (*Ilex glabra* (L.) A. Gray), dwarf and blue huckleberries

(*Gaylussacia dumosa* and *frondosa* (Andrews) Torr. & A. Gray), and a few blueberry species (*Vaccinium* spp.).

Table 1. Summary of site preparation treatments implemented in the study.

Treatment	Vegetation Control		Soil Manipulation		
	Chop	Herbicide	Flat	Mound	Bed
Flat [control] (F)			X		
Chop/flat (CF)	X		X		
Herbicide/flat (HF)		X	X		
Chop/mound (CM)	X			X	
Herbicide/mound (HM)		X		X	
Chop/bed (CB)	X				X
Herbicide/bed (HB)		X			X
Chop/herbicide/bed (CHB)	X	X			X

Experimental Design

This study utilized a randomized complete block design, consisting of eight treatments replicated across five blocks for a total of 40 experimental units. Study treatments were randomly assigned to experimental units with dimensions of 54.6 m x 86.3 m (almost 0.5 ha), with 20 m buffers between units to prevent treatment overlap and reduce edge effects. Study treatments consist of combinations of two vegetation control treatments (herbicide and chopping) and three soil manipulation treatments (mounding, bedding, and flat-planting [no additional treatment]) to create a 2 x 3 factorial design of six treatments. The two additional study treatments were an untreated check (no vegetation control and flat-planting) and a bedding treatment with both levels of vegetation control. The eight resulting study treatments (Table 1) are often referred to by their initials: F (flat-planting and no vegetation control), HF (herbicide and flat-planting), CF (chopping and flat-planting), HM (herbicide and mounding), CM (chopping and mounding), HB (herbicide and bedding), CB (chopping and bedding), and CHB (chopping, herbicide, and bedding).

Prior to site preparation, all blocks were harvested and sheared to remove standing vegetation. Study treatments were then applied in August 2003. The chopping treatment was implemented using a 2.4 m Lucas Drum Chopper pulled by a TD15 Dresser crawler tractor. The herbicide treatment included 1.54 lb/ha of imazapyr and 1.24 lb/ha of triclopyr, which were mixed and broadcast at a rate of 280 l/ha. Mounds approximately 1.2 m wide were created using a New Forest Technology custom mounding bucket on a Caterpillar 320BL excavator. For consistency, the mounds were created in rows, rather than in the discontinuous, random distribution that is commonly associated with mounding site preparation. Beds approximately 2.1-2.4 m wide were created using a Rome 6-disc Bedding Harrow with three discs on each side. All blocks were burned in October or November 2003 to remove remaining vegetation and further prepare the sites for planting. Container-grown longleaf pine seedlings from locally collected seed were hand planted in December 2003. Each block was burned three times after planting at roughly a five-year fire return interval.

Data Collection

After planting, a full census was conducted to record the number of seedlings planted in each experimental unit. A sub-sample of 45 seedlings from each experimental unit was then randomly selected for repeated growth and survival measurement. Survival was measured through two years after planting, with two measurements periods in August 2004 and 2005. Root collar diameter (RCD) was measured through three years after planting, with four measurement periods occurring in June and December 2004, December 2005, and December 2006.

In summer 2018, a sample population of trees was selected from each experimental unit for growth and density measurement. Five circular subplots were systematically located within each rectangular experimental unit, with one subplot located at the center and each of the other four subplots spaced between each respective corner and the center. Each subplot had a radius of 10 m, resulting in approximately a third of each experimental unit being sampled. Every tree within the subplot was identified to species and measured for height and diameter at breast height (DBH). There was some seeding in of slash and pond pine (*Pinus serotina* Michx.); however, our analyses focused only on the planted longleaf pine.

In summer and winter 2017, we conducted stem analysis on a random sample of longleaf pine trees from each experimental unit to quantify growth responses of individual trees to site preparation. We established a transect running longways across the middle of each rectangular experimental unit and established six equally spaced points along that transect. We then located the longleaf pine nearest each point to be destructively sampled for stem analysis. Each tree was cut into sections every 10 cm from the ground line to 1.5 m and every 25 cm thereafter. These intervals gave us a fine resolution toward the base of the tree, where transitions between growth years were less spaced out. To obtain an accurate count of growth rings, we followed the Missouri Tree-Ring Lab's standard protocol for sanding (J. Marschall, personal communication, 2017) and used a stereo microscope for counting annual rings.

Data Analysis

We fit linear mixed models to each stand-level response variable of interest (i.e., height, DBH, trees per hectare, grass stage seedlings per hectare, and basal area), treating treatment as a fixed effect and the hierarchical, nested sampling design (i.e., subplots within experimental units within blocks) as a random effect. We used a height of 15 cm (from ground line to terminal bud) as the threshold to classify grass stage emergence (Boyer, 1988; Wang et al., 2016). We then conducted one-way analysis of variance (ANOVA) on each of the fitted mixed models to test for differences among the eight study treatments for each response variable of interest. When one-way ANOVA indicated that treatments significantly differed ($\alpha = 0.05$), post-hoc Tukey-adjusted pairwise comparisons were used to determine which treatments significantly differed. Additionally, we used a 2 x 3 factorial ANOVA, excluding the F and CHB treatments, to determine the effect of individual factors (e.g., chopping vs. herbicide) on each response variable of interest and to test for interactions between the vegetation control treatments and soil manipulation treatments. When factorial ANOVA indicated that main effects (i.e., vegetation control or soil manipulation treatments) were significant ($\alpha = 0.05$) and the interaction was not, post-hoc Tukey-adjusted pairwise comparisons were used to determine which individual factors significantly differed.

For the stem analysis data, we determined the age of each tree section by counting annual rings and adjusting internode tree heights using Carmean's (1972) method (Dyer and Bailey, 1987). From this data, we computed two response variables of interest: age of emergence from the grass stage and rate of height growth after emergence. Age of emergence from the grass stage was determined by the number of years since a tree had reached 15 cm in height. The rate of growth after emergence was determined by computing the mean annual height growth after the tree exceeded 15 cm in height. Similar to the stand-level data, we fit linear mixed models for both age of emergence and post-emergence growth rate. We then conducted one-way ANOVA, followed by Tukey-adjusted pairwise comparisons, on both mixed models to test for differences among the eight study treatments. We also conducted a 2 x 3 factorial ANOVA, excluding the F and CHB treatments and followed by Tukey-adjusted pairwise comparisons, to determine the effects of individual treatment levels on the response variables.

To understand how longleaf pine responses to site preparation changed through time, we integrated the stand-level measurements taken 15 years after planting with data collected in the original study, which measured seedlings through three years after planting. We computed two response variables of interest: survival and relative diameter. Survival in the initial two-year measurement period was calculated by applying the survival rates from sub-sampled seedlings to the number of planted seedlings recorded in the initial census. Survival at 15 years was calculated by scaling longleaf pine density within the sub-plots up to the experimental unit level and then dividing those estimates by the number of seedlings planted in each experimental unit. To integrate root collar diameter measurements from the seedling study with diameter at breast height measurements recorded at age 15, we calculated diameter for each treatment relative to the control (F). Relative diameter was calculated for each measurement period using the following equation:

$$relative\ diameter = \frac{treatment\ mean - control\ mean}{control\ mean}$$

Therefore, a treatment mean diameter that is twice as large as the control mean diameter results in a relative diameter value of one. We fit linear mixed models to both survival and relative diameter and then used repeated measures ANOVA to test for differences among treatments and through time and to test for interactions between treatment and time. Similar to one-way and factorial ANOVA tests, significant main effect and interaction terms ($\alpha = 0.05$) from repeated measures ANOVA were followed by post-hoc Tukey-adjusted pairwise comparisons to determine which treatments and time periods significantly differed.

Data were analyzed in R (R Core Team, 2018). The *lme* function in the *nlme* package (Pinherio et al., 2018) and the *glmer* function in the *lme4* package (Bates et al., 2015) were used for fitting linear mixed models, with the former being used in most cases and the latter being used to specify a Poisson distribution where necessary. The *anova* function in the *stats* package (R Core Team, 2018) was used for running ANOVA tests. The *lsmeans* function from the *emmeans* package (Lenth, 2019) and the *CLD* function from the *multcompView* package (Graves et al., 2015) were used for post-hoc, Tukey-adjusted pairwise comparisons. When factorial or repeated measures ANOVA resulted in a significant ($\alpha = 0.05$) interaction term, we used the *testInteraction* function in the *phia* package (De Rosario-Martinez, 2015) to conduct post-hoc interaction significance tests. The *dplyr* package (Wickham et al., 2019) was used for data manipulation, the *ggsurvplot* function in the *survminer* package (Kassambara and Kosinski, 2018) was used for plotting survival, and the *ggplot2* package (Wickham, 2016) was used to create all other figures.

Results

Stand-Level Responses to Site Preparation

Fifteen years after planting, site preparation resulted in significant differences in longleaf pine height ($p < 0.001$) and diameter at breast height ($p < 0.001$) among the eight study treatments (Fig. 1A and 1B). CHB, HM, and HB resulted in the greatest tree size at age 15, with mean heights of more than 5 m and mean DBH values upwards of 8 cm. These three treatments resulted in significantly greater height than HF, CF, and F and significantly greater DBH than CF and F. CF and F, the treatments with the poorest growth, produced mean heights of less than 2 m and mean DBH values of less than 5 cm. Based on factorial ANOVA, both soil manipulation and vegetation control treatments affected height ($p < 0.001$) and DBH ($p \leq 0.001$) of planted longleaf pine (Table 2), and there was no significant interaction between the treatment types for height ($p = 0.469$) or DBH ($p = 0.773$). Bedding and mounding resulted in greater height and DBH than flat-planting (no soil manipulation), with differences in height and DBH exceeding 2 m and 2 cm, respectively. Herbicide resulted in greater height and DBH than chopping, with differences in height and DBH exceeding 1 m and 1 cm, respectively.

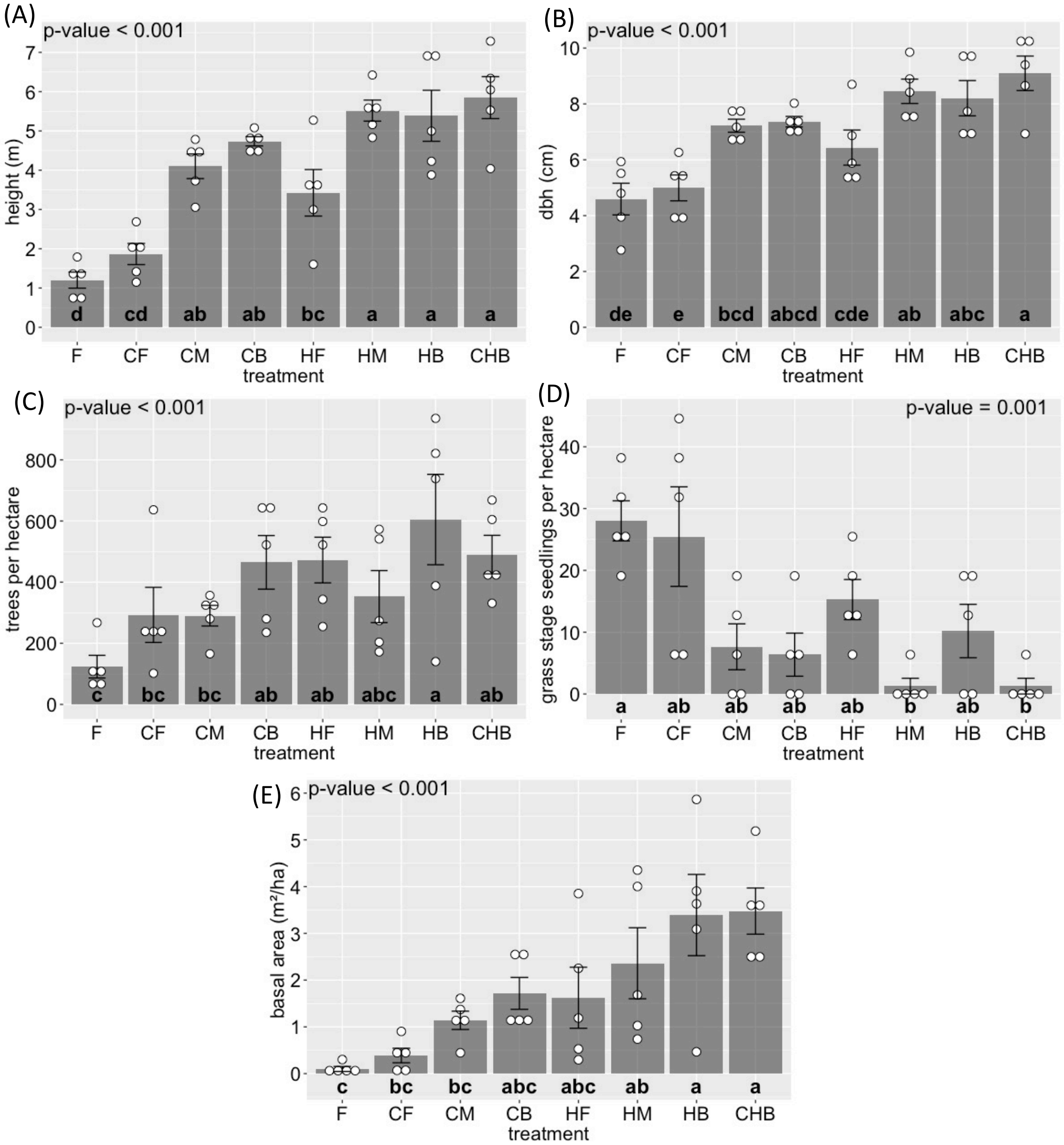

Figure 1. Longleaf pine stand-level responses to site preparation study treatments 15 years after application. Treatments are defined in Table 1. Bars show treatment means and standard errors; points show means of each experimental unit. Same letters indicate no significant difference ($\alpha = 0.05$) among treatments within a response variable based upon Tukey-adjusted pairwise comparisons; *p*-values are from one-way ANOVA tests.

Table 2. Least square means of longleaf pine stand-level responses to individual site preparation treatment levels 15 years after application.

Treatment	Mean height (m)	Mean diameter at breast height (cm)	Mean trees per hectare	Mean grass stage seedlings per hectare	Mean basal area (m^2/ha)
Flat	2.59 b	5.77 b	383 ab	20.4 a	1.01 b
Mounding	4.79 a	7.79 a	321 b	8.3 b	1.75 ab
Bedding	5.02 a	7.78 a	535 a	4.5 b	2.55 a
p-value	< 0.001	< 0.001	0.010	0.006	0.009
Chopping	3.50 b	6.46 b	349 b	13.2 a	1.08 b
Herbicide	4.76 a	7.76 a	477 a	8.9 a	2.46 a
p-value	< 0.001	0.001	0.026	0.264	0.001

Same letters indicate no significant difference ($\alpha = 0.05$) within a treatment type and response variable based upon Tukey-adjusted pairwise comparisons; *p*-values are from 2 x 3 factorial ANOVA tests.

Site preparation treatments also resulted in significant differences in longleaf pine tree density ($p < 0.001$) and grass stage seedling density ($p = 0.001$) at age 15. HB, CHB, CB, and HF had greater than three times the number of trees per hectare than F (Fig. 1C), while F resulted in more grass stage seedlings per hectare than CHB and HM (Fig. 1D). F treatments averaged 28 grass stage seedlings per hectare, while grass stage seedlings were absent from all but one experimental unit for CHB and HM. Based on factorial ANOVA, soil manipulation ($p = 0.010$) and vegetation control treatments ($p = 0.026$) affected trees per hectare, and there was no significant interaction between the treatment types ($p = 0.659$). Bedding resulted in greater tree density than mounding, and herbicide resulted in greater tree density than chopping 15 years after planting (Table 2). In terms of grass stage seedling density, differences among soil manipulation treatments were significantly different ($p = 0.006$), but there was no significant difference between vegetation control treatments ($p = 0.264$) and no significant interaction between treatment types ($p = 0.162$). Flat-planting resulted in greater grass stage seedlings per ha than mounding and bedding (Table 2).

Basal area, which integrates tree size and density at the stand level, was also significantly different among the eight study treatments ($p < 0.001$). Mean basal area for CBH and HB, which approached 3.5 m^2/ha, was significantly greater than that of CM, CF, and F, which were near or below 1 m^2/ha (Fig. 1E). Soil manipulation ($p = 0.009$) and vegetation control treatments ($p = 0.001$) affected basal area, and there was no significant interaction between the treatment types ($p = 0.848$). Bedding resulted in significantly greater basal area than flat-planting, and herbicide resulted in significantly greater basal area than chopping 15 years after planting (Table 2).

Responses to Site Preparation through Time

Repeated measures ANOVA indicated that time significantly affected longleaf pine survival ($p < 0.001$) through 15 years after planting, while treatment did not ($p = 0.281$). Survival decreased through time, and mean survival across all treatments was 72.5% at eight months after planting, 59.1% at 20 months, and 32.7% at 15 years. The interaction between time and treatment approached significance ($p = 0.061$), likely because survival did not significantly differ among treatments after the first and second growing seasons (Knapp et al., 2006) but did differ in year 15 according to one-way ANOVA ($p = 0.002$). At this measurement period, the herbicide treatments – HB, HM, HF, and CHB – resulted in higher survival compared to the untreated control (Fig. 2).

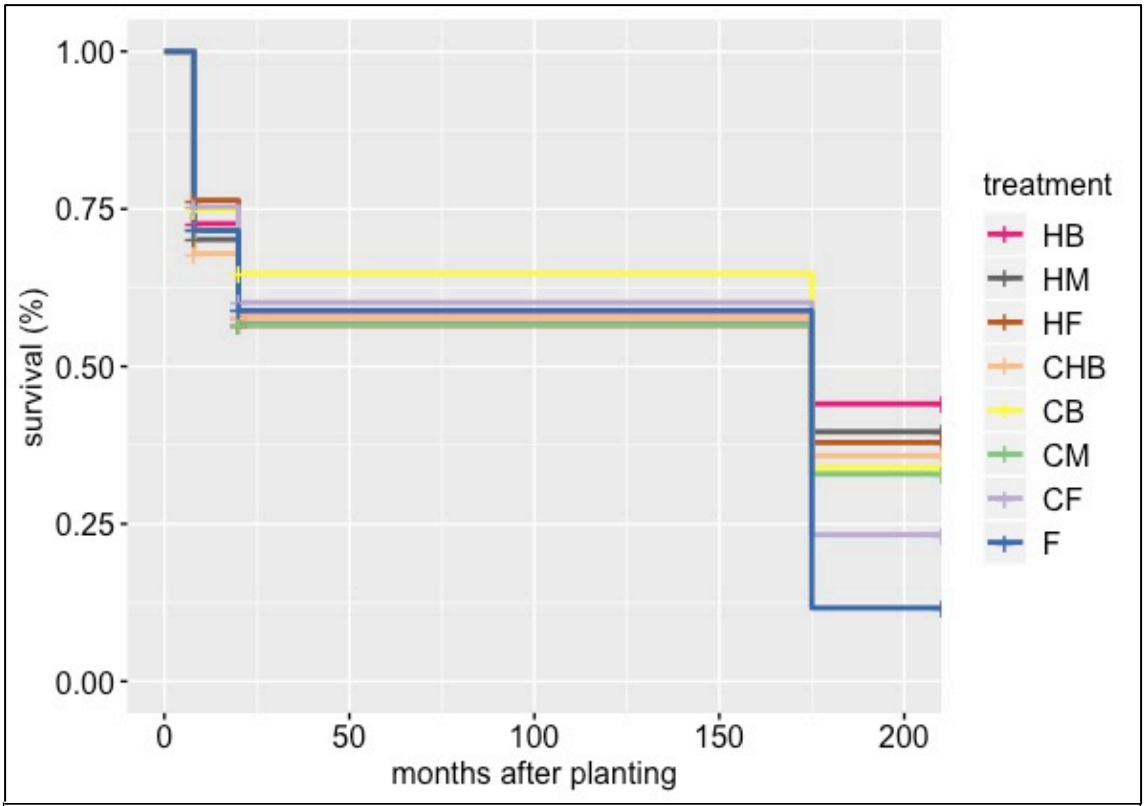

Figure 2. Survival for each study treatment (defined in Table 1) at three points in time: 8, 20, and 175 months after planting.

The interaction between treatment and time was significant ($p < 0.001$) for relative diameter of longleaf pine through 15 years after planting. Post-hoc interaction tests indicated that there were no significant differences among treatments at seven or 12 months after planting ($p \geq 0.855$), but there were differences at two, three, and 15 years after planting ($p < 0.001$). After two years, CHB and HB consistently had greater relative diameter than CF and F (Fig. 3). By year 15, CHB, HM, HB, CB, and CM had significantly greater relative diameter than CF and F. Notably, there were no significant changes in relative diameter for any individual study treatment after year two ($p = 0.382$).

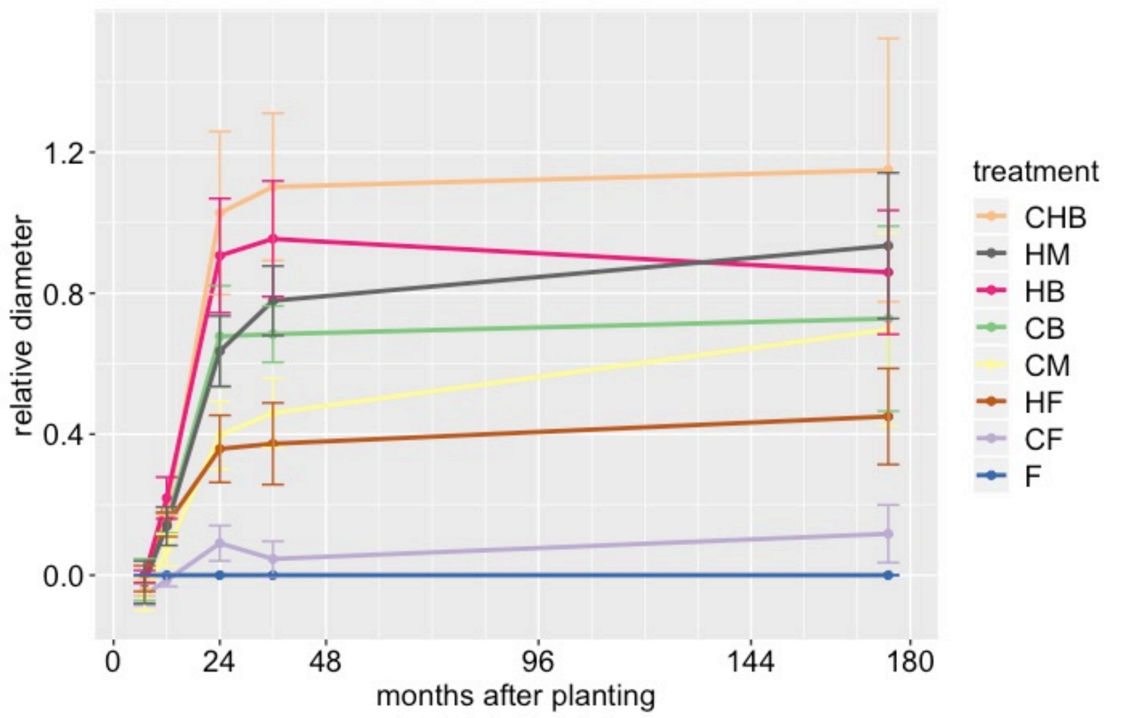

Figure 3. Diameter of each study treatment (defined in Table 1) relative to the control (F) at five points in time, from seedling establishment into stand development.

Tree-Level Responses to Site Preparation

One-way ANOVA indicated that there were significant differences in age of emergence from the grass stage among the eight study treatments ($p < 0.001$). CF, which had a mean emergence age of 9.25 years after planting, emerged significantly later than the other seven treatments (Fig. 4A). The other treatments had emergence ages ranging from 3.72 to 5.29 years and did not significantly differ from each other. Based on factorial ANOVA, soil manipulation ($p < 0.001$) and vegetation control ($p < 0.001$) affected emergence age. The interaction between the treatment types approached significance ($p = 0.054$) because CF resulted in much later emergence than any of the other treatments. Bedding and mounding resulted in earlier grass stage emergence than flat-planting, while herbicide resulted in earlier emergence than chopping (Table 3).

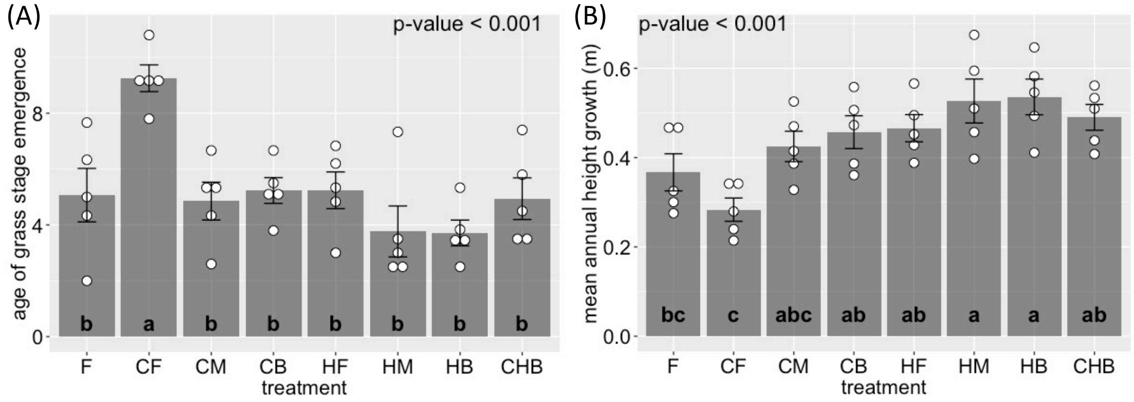

Figure 4. Tree-level responses to site preparation study treatments 15 years after application. Treatments are defined in Table 1. Same letters indicate no significant difference ($\alpha = 0.05$) among treatments within a response variable based upon Tukey-adjusted pairwise comparisons; *p*-values are from one-way ANOVA tests.

One-way ANOVA also found significant differences in mean annual height growth after grass stage emergence among the eight study treatments ($p < 0.001$). HM and HB, with mean height growth exceeding 0.5 m/year, grew significantly faster than F and CF, which had mean growth rates of 0.33 m/year and 0.23 m/year, respectively (Fig. 4B). CF had significantly lower annual height growth than all treatments except for F and CM. Based on factorial ANOVA, soil manipulation ($p < 0.001$) and vegetation control ($p < 0.001$) significantly

Table 3. Least square means of tree-level responses to individual site preparation treatment levels 15 years after application.

Treatment	Age of grass stage emergence	Mean annual height growth (m/yr)
Flat	7.31 a	0.330 b
Mounding	4.33 b	0.459 a
Bedding	4.48 b	0.486 a
p-value	< 0.001	< 0.001
Chopping	6.47 a	0.357 b
Herbicide	4.27 b	0.493 a
p-value	< 0.001	< 0.001

Same letters indicate no significant difference ($\alpha = 0.05$) within a treatment type and response variable based upon Tukey-adjusted pairwise comparisons; *p*-values are from 2 x 3 factorial ANOVA tests.

impacted post-emergence growth, while the interaction between the two treatment types was insignificant (*p* = 0.195). Bedding and mounding resulted in a greater annual growth rates than flat-planting, and herbicide produced greater annual growth than chopping (Table 3).

Stem analysis allowed us to reconstruct height growth of each sampled longleaf pine through time. Fig. 5 shows this reconstruction summarized by treatment, and the growth patterns displayed represent actual mean growth patterns for each treatment. The year in which each treatment line begins on the x-axis indicates the earliest grass stage emergence age of trees sampled in that treatment. The y-axis represents cumulative height growth after grass stage emergence. Stem analysis also allowed us to reconstruct mean annual height growth after grass stage emergence for each treatment (Fig. 6). HB and HM had significantly greater annual growth through 10 years after grass stage emergence than CF, and aside from slight overlap in confidence interval envelopes in years 5-8, so did HF, CHB, and CB. Annual height growth continued to increase slightly over time for the control, while growth generally levelled off after 4-5 years for the other treatments.

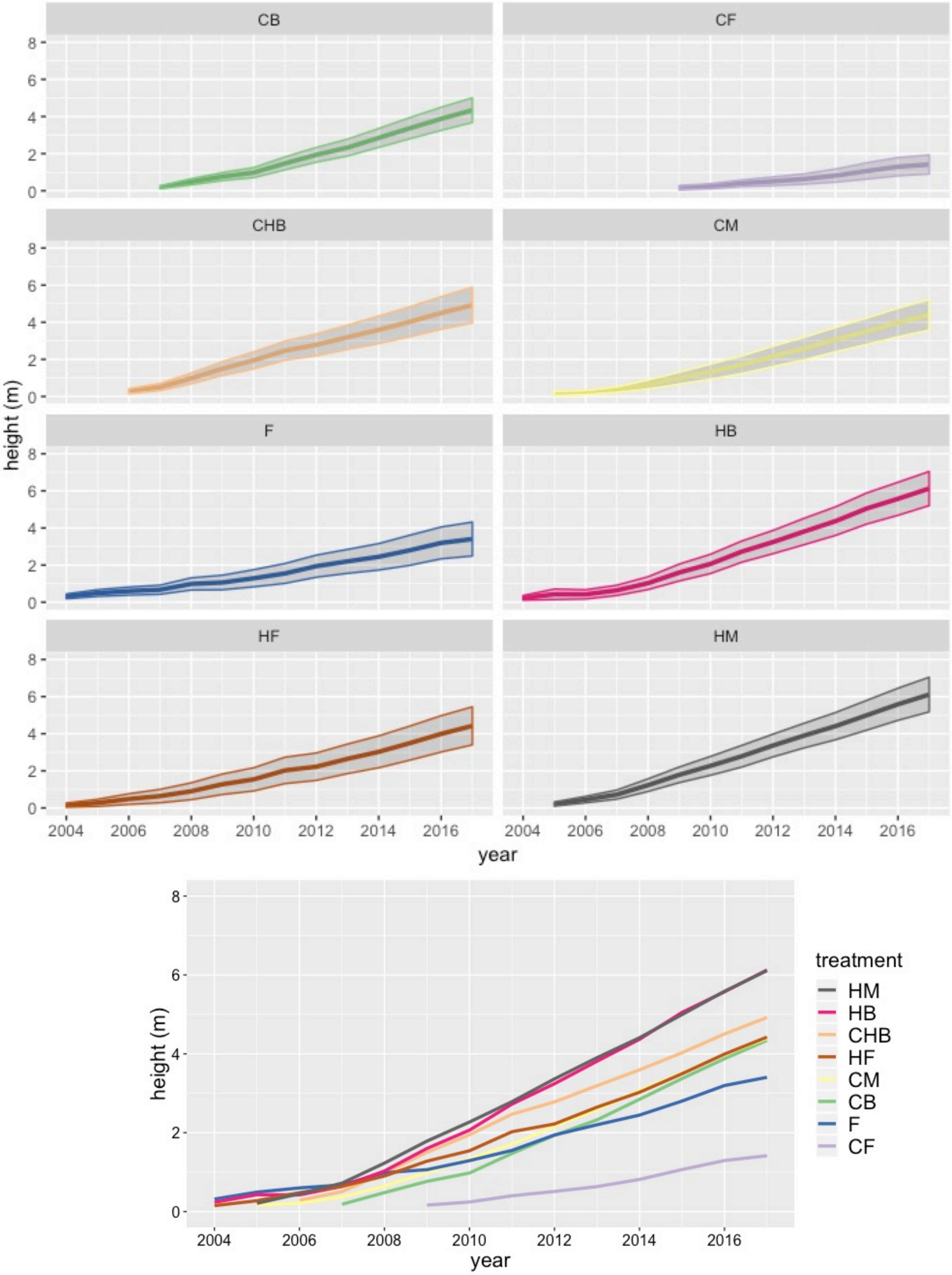

Figure 5. Reconstruction of mean height growth after grass stage emergence for each study treatment (defined in Table 1). Individual treatment panels include a line for the treatment mean and an envelope of the 95% confidence interval. The plot with all eight treatments shows only treatment means.

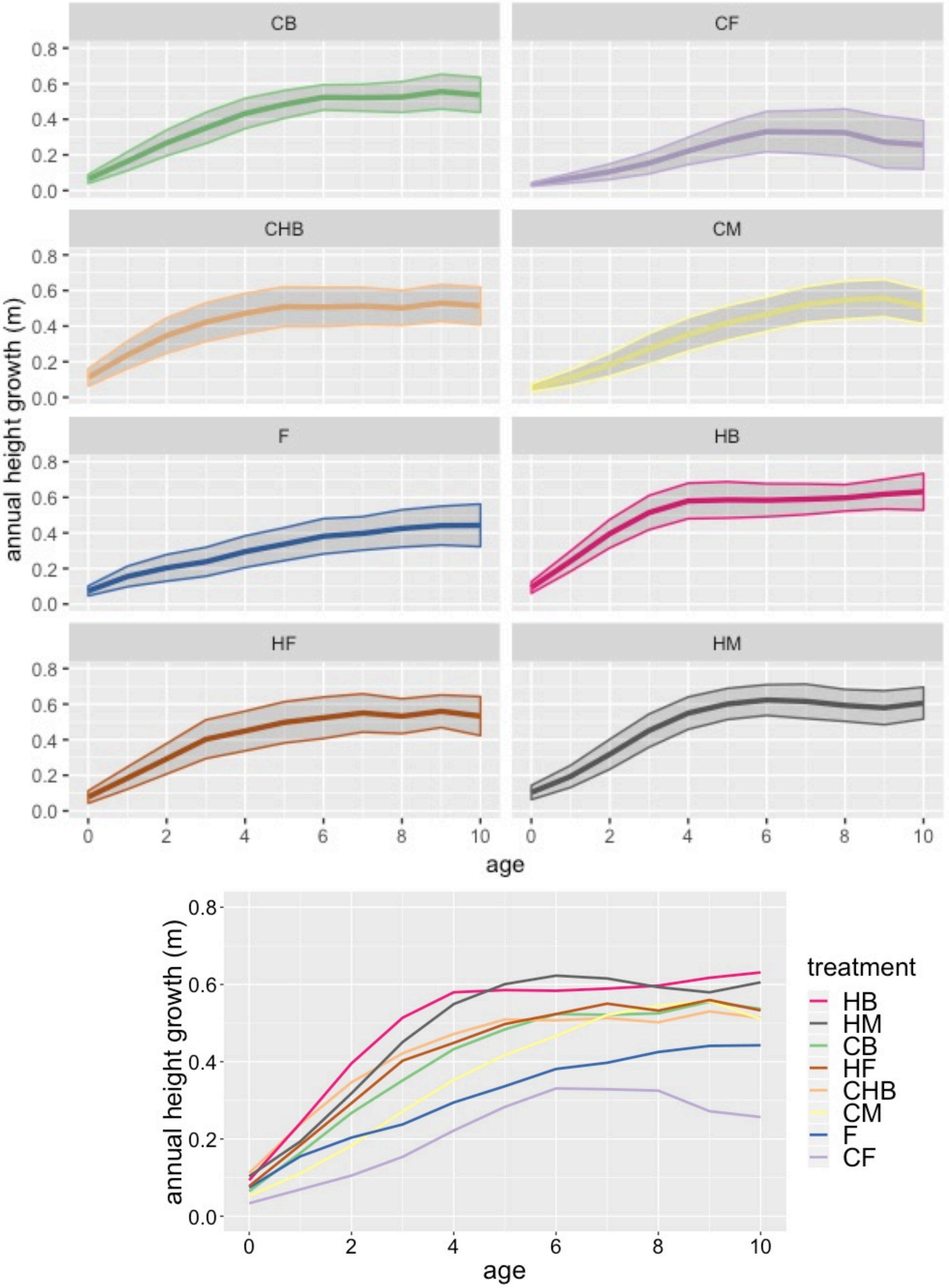

Figure 6. Mean annual height growth after grass stage emergence for each study treatment (defined in Table 1). Individual treatment panels include a line for the treatment mean and an envelope of the 95% confidence interval. The plot with all eight treatments shows only treatment means. Lines are smoothed on a three-year mean to reduce noise.

Discussion

Our results show that site preparation had strong effects on longleaf pine stand development through 15 years after treatment. The untreated control (F) resulted in the smallest height and DBH, the lowest density, and the greatest number of seedlings still in the grass stage compared to the other seven study treatments, although not every treatment was statistically different from the control for all responses. This suggests that any form of site preparation, aside from chopping alone, may improve long-term longleaf pine stand establishment compared to no treatment at all. Other studies have also found significantly reduced growth of planted slash (Haywood, 1987) and longleaf pine seedlings (Addington et al., 2012; Freeman and Jose, 2009; Knapp et al., 2006) on untreated sites compared to those with site preparation, which further supports the positive relationship between site preparation and longleaf pine growth. Our results also demonstrate a pattern of increasing magnitude of response with increasing site preparation intensity, which is consistent with other studies that have examined longleaf pine responses to vegetation control and soil manipulation (Knapp et al., 2006; Loveless et al., 1989). Study treatments that resulted in the greatest size and density included both herbicide and a soil manipulation treatment, while treatments that produced intermediate size and density included only herbicide or soil manipulation. The treatments that resulted in the smallest size and lowest density – CF and F – included neither herbicide nor soil manipulation.

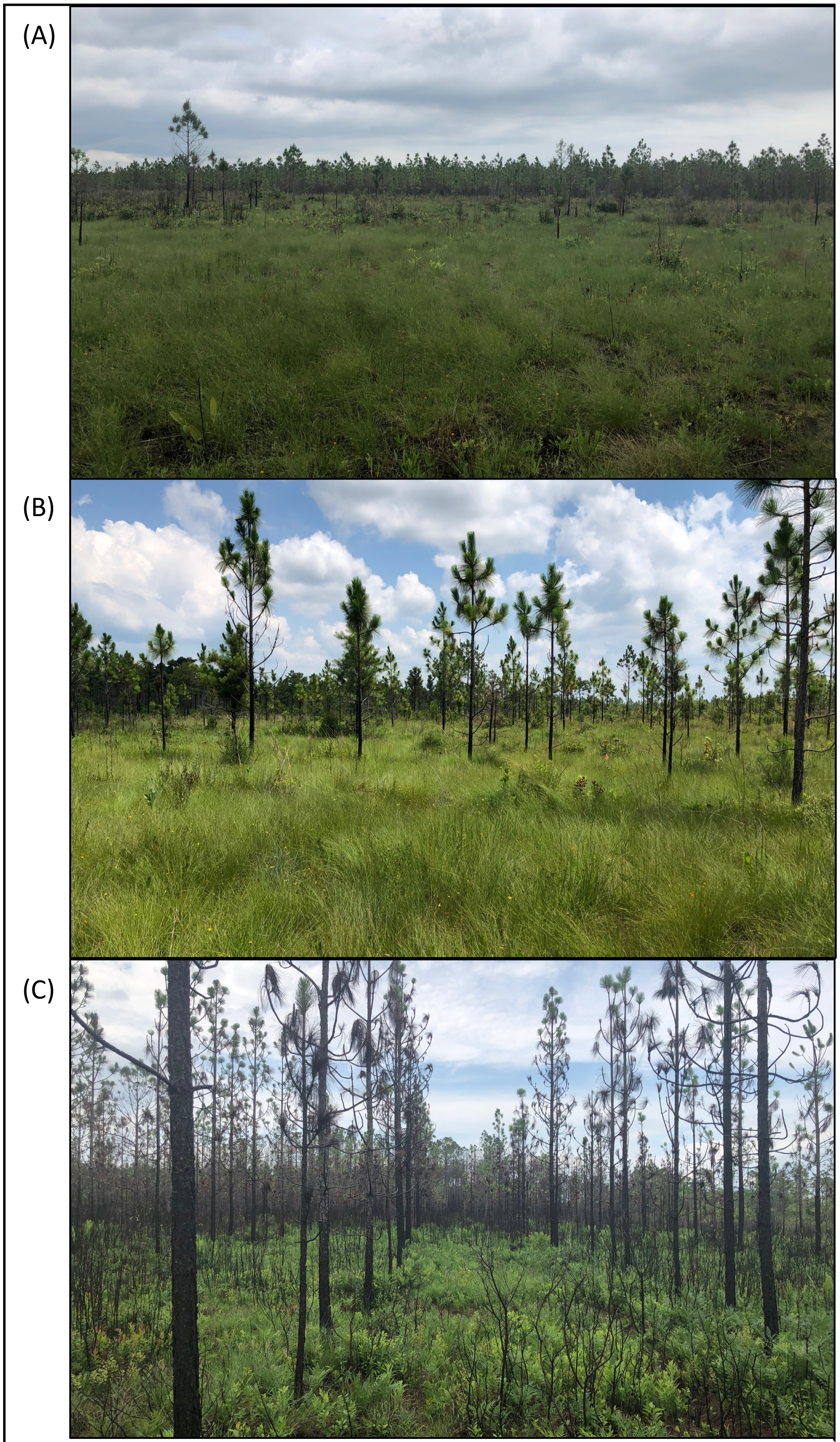

Figure 6. Photos to contrast the stand structure of (A) an untreated control, (B) a CB treatment, and (C) an HB treatment.

In our study, chopping was not effective for improving conditions for longleaf pine establishment. Chopping resulted in significantly lower longleaf pine size and density compared to herbicide. This is consistent with other studies, which have found that chopping resulted in reduced longleaf pine seedling growth (Freeman and Jose, 2009; Knapp et al., 2006) and increased woody vegetation (Miller, 1980) compared to other vegetation control treatments. Freeman and Jose (2009) suggested that chopping plus fire may be sufficient for longleaf pine establishment in flatwoods, even though growth may be slow. However, our findings indicate that chopping followed by fire was not sufficient for long-term establishment of longleaf pine on similar flatwoods sites. Our study found no significant differences between F and CF and between CHB and HB, emphasizing the ineffectiveness of chopping, alone or in combination with other treatments. Although chopping causes minimal soil disturbance, has low impact on herbaceous vegetation, and immediately reduces standing woody vegetation, a major drawback is that it promotes sprouting of woody species (Lowery and Gjerstad, 1991). On hydric sites where woody competition is abundant, a single pass of chopping prior to planting may be ineffective at reducing competition, which could explain the poor stand establishment responses produced by chopping in this study.

In contrast, herbicide was present in the three treatments combinations – CHB, HB, and HM – that produced the most successful, long-term stand establishment outcomes. Studies have consistently observed increased longleaf pine seedling growth (Addington et al., 2012; Boyer, 1988; Freeman and Jose, 2009; Knapp et al., 2006; Loveless et al., 1989) and earlier grass stage emergence (Boyer, 1988; Freeman and

Jose, 2009; Loveless et al., 1989) as a result of herbicide application. Importantly, none of these studies tracked responses beyond six years after planting, yet our findings indicate that improved growth of planted longleaf pine as a result of herbicide application can be maintained into stand development. Additionally, studies have generally found that longleaf pine seedling survival is not affected by herbicide (Addington et al., 2012; Knapp et al., 2006; Loveless et al., 1989) or is reduced following herbicide application (Boyer, 1988; Freeman and Jose, 2009). Survival in our study did not differ among treatments through two years after planting (Knapp et al., 2006), but by year 15, significant differences existed, with herbicide being present in the four treatments with the highest survival. This finding emphasizes the importance of a longer-term perspective when evaluating responses to site preparation.

In our study, herbicide resulted in both earlier emergence from the grass stage and greater annual height growth after emergence compared to chopping. Therefore, herbicide application prior to planting appears to have provided both an immediate (earlier grass stage emergence) and long-term (increased height growth) advantage for longleaf pine establishment. The former is consistent with our expectation that reduced competition should promote earlier grass stage emergence and improve seedling growth, but the long-term advantage obtained from herbicide is less intuitive. Competition control provided early on by herbicide likely did not persist indefinitely, as we would expect competitors to recover through time. On flatwoods sites, Freeman and Jose (2009) found no difference in shrub cover among herbicide treatments and the control four years after treatment, which provides a rough estimation of when we might expect competitors to recover. Yet, stem analysis indicated that annual growth rates on herbicide treatments remained higher through time than treatments with chopping. Boyer (1990) suggested that duration of the grass stage has a critical impact on longleaf pine growth, which could explain why herbicide application in our study had a long-term, positive impact on longleaf pine growth.

Similar to herbicide, bedding resulted in improved long-term longleaf pine stand-level size and density compared to flat-planting (no treatment). Other studies have found that bedding increased growth of longleaf seedlings (Knapp et al., 2006; Loveless et al., 1989) and longer-term survival and growth of other southern pines (Outcalt, 1984; Pritchett, 1979). In our study, mounding produced responses similar to bedding, except for trees per hectare, where density was significantly lower on mounded treatments. This is likely due to lower initial planting density on mounds (mean = 886 tph) compared to beds (mean = 1,371 tph) and flat-planted treatments (mean = 1,188 tph). The similarities in survival at year 15 between bedded and mounded treatments further implies that lower stand densities on mounded treatments at year 15 were the result of lower planting density.

The significant improvement in long-term longleaf pine growth and survival as a result of bedding and mounding demonstrates the importance of ameliorating adverse conditions on productive, hydric sites. On the same sites, Knapp et al. (2008) found that bedding and mounding resulted in a reduction in soil moisture at 6 cm and an increase in soil temperature at 15 cm relative to the control through two years after treatment application. This suggests that two of the hypothesized benefits of soil manipulation treatments, namely drier and warmer soil, did in fact play in role in promoting seedling establishment and perhaps continued to contribute to longer-term stand development. Stem analysis indicated that, similar to herbicide, bedding and mounding provided short- (earlier grass stage emergence) and long-term (increased height growth) advantages for planted longleaf pine compared to flat-planting. The soil in mounds is expected to settle out over time (Londo and Mroz, 2001), though, and it is logical to expect that soil on beds would do the same. However, it has been estimated that bedding may alter a site's hydrology for more than 30 years (Brockway et al., 2005), which may explain why bedded treatments still had greater growth even 15 years after application in our study. Longleaf pine on mounded treatments, too, continued to have greater height growth 15 years after treatment, suggesting that even if mounds had begun to settle, longleaf pine was still benefiting from the soil manipulation treatment.

Treatments that resulted in the greatest stand size and density had the highest post-emergence height growth rates, and the opposite was also true. This suggests that growth rate after grass stage emergence may play a more significant role in determining longer-term stand establishment outcomes than the timing of grass stage emergence, because patterns between timing of emergence and stand-level responses were less consistent. This inconsistency is best demonstrated by the fact that the untreated control (F) had statistically similar timing of grass stage emergence as all treatments except CF, yet ultimately resulted in the poorest long-term stand establishment outcomes. To understand why this inconsistency occurred, we must consider the survivorship bias that was implicit in our sampling approach. By studying a random sample of trees 15 years after planting, we were limited to studying only those trees that were still alive after 15 years. As a result, the conclusions we draw are based only on trees that managed to survive. This is particularly evident on the untreated control, which experienced a high level of mortality, likely due to abundant competition and poor soil drainage. Stem analysis results indicate that for trees to survive on these harsh hydric sites, they must emerge from the grass stage early. If they fail to do so, competitors have time to recover and will presumably overtake the grass stage seedlings, resulting in suppression and eventually mortality. Because competition and poor drainage were not improved on the control, post-emergence growth remained low for trees that did manage to survive.

Using the site preparation response trajectories proposed by Morris and Lowery (1988) and Nilsson and Allen (2003), growth and survival of longleaf pine in our study exhibited a type A response, meaning the difference between treated and untreated areas increased through time. Treatments had statistically similar survival during seedling establishment, but by year 15, significant differences existed. Differences in relative diameter among treatments became significant much earlier – in year two. Even though the relative diameter remained consistent from year two to 15, the true difference between treated and untreated sites continued to grow because relative diameter is a multiplier. For example, if a treatment’s relative diameter is 1, then it is twice as large as the control. When the control is 2 cm, the treatment is 4 cm, but when the control is 8 cm, the treatment is 16 cm. Similarly, stem analysis indicated that annual height growth remained consistently higher through time on treated sites compared to the untreated control and CF. The fact that survival, relative diameter, and height growth all exhibited type A trajectories demonstrates the importance of monitoring long-term responses to site preparation. The longer-term perspective allowed us to understand that treatment responses continued to grow through time, whereas a shorter-term perspective may have underestimated the impacts of site preparation. Going forward, the type A trajectory of these stands indicates that the difference between treated and untreated sites should continue to grow, further emphasizing the substantial impact of site preparation on these hydric sites.

Understanding long-term responses to site preparation is critical for evaluating treatment success and informing future management activity, regardless of the management objective. Historically, site preparation has most commonly been used and studied in the context of plantation forestry, where maximizing tree productivity throughout a stand's rotation is the primary objective. Restoration is an entirely different objective, and site preparation treatments for restoration must be evaluated using different criteria than those of traditional plantation forestry. In restoration scenarios, a threshold often exists between the pre-restoration community (e.g., loblolly or slash pine-dominated stand lacking overstory longleaf pine or natural regeneration) and the desired community (e.g., longleaf pine savanna) (Martin and Kirkman, 2009). Intensive intervention early on, such as site preparation, may be necessary to overcome the threshold between the two community states, but once the "restoration threshold" is overcome (e.g., longleaf pine is successfully established), less intense treatment (e.g., regular prescribed fire) may be all that is required to maintain the restored, desired community state (Martin and Kirkman, 2009). Therefore, site preparation treatments must promote sufficient grass stage emergence and survival to create a future stand of desired structure (e.g., longleaf pine savanna or forest) and sustain a frequent fire regime that is continuous in both time and space (Mitchell et al., 2009). An evaluation of the study treatments' impacts on fire and fuels ecology is beyond the scope of this study, but this study does provide insight into the long-term structural development of longleaf pine as a result of site preparation on hydric sites.

Longleaf pine restoration may encompass a range of structural outcomes and management objectives, from an open savanna with an emphasis on restoring the understory plant community to a woodland with an emphasis on dual management of timber and biodiversity. For the former, no site preparation at all may be sufficient. Although the untreated control in this study resulted in the slowest growth and highest mortality, it still resulted 100 longleaf pine trees per ha in year 15, which may be sufficient to eventually produce an open longleaf pine savanna. Slow growth could even be considered beneficial in some restoration efforts because slower tree growth is correlated increased longevity (Black et al., 2008). Forgoing intense site preparation might also be desirable because of the potential deleterious impact that mechanical treatments may have on native understory plants, such as wiregrass (Brockway et al., 2006, 2005; Clewell, 1989; Mitchell et al., 2006; Platt et al., 2015; Van Lear et al., 2005).

An alternative restoration objective could be to maximize longleaf pine growth so that time to maturity is minimized and red-cockaded woodpecker (*Leuconotopicus borealis*) habitat is established quickly (Shaw and Long, 2007). In this scenario, site preparation would be critical for improving establishment and long-term growth of planted seedlings. Our findings indicate that stand size and density at age 15 is maximized by combining herbicide and soil manipulation. However, in the context of restoration, these additional gains may not be worth the added financial and ecological cost, and all that may be necessary is either application of herbicide or some form of soil manipulation. Our future work explores understory responses to site preparation on these same hydric sites and should provide more context for evaluating site preparation's role in longleaf pine ecosystem restoration.

Conclusion

Site preparation significantly improved long-term establishment and growth of longleaf pine on hydric sites. Specifically, herbicide resulted in greater growth, higher survival, and earlier grass stage emergence compared to chopping. Similarly, soil manipulation treatments resulted in improved stand establishment outcomes relative to flat-planting (no treatment), with bedding tending to slightly outperform mounding. Differences in survival and growth between treated and untreated areas increased through time, which demonstrates that site preparation had lasting, long-term impacts on longleaf pine stand development. Additionally, stem analysis indicated that increased height growth after grass stage emergence seemed to contribute more directly to long-term stand establishment outcomes than did the timing of grass stage emergence.

Restoration may encompass a range of objectives, and if improved long-term growth or survival of planted seedlings is an important aspect of a restoration effort, then our findings indicate that herbicide or soil manipulation may be necessary on hydric sites. Combining herbicide and soil manipulation maximized stand size and density, but in the context of restoration, this intense approach may not be worth the ecological cost. On the other hand, if restoration seeks only to establish an open longleaf pine savanna, our findings indicate that a planting density upwards of 1,000 trees per ha without site preparation may result in sufficient long-term establishment. This approach may result in high mortality, but it does not risk deleterious impacts on the understory as a result of intense mechanical site preparation.

Understanding understory responses to site preparation is critical for conducting a holistic evaluation of longleaf pine ecosystem restoration. Longleaf pine ecosystem restoration typically seeks to not only establish an overstory of longleaf pine but also maintain or promote the ground layer plant community (Walker and Silletti, 2006). Therefore, a balance must be struck between maximizing site preparation that promotes longleaf pine establishment and minimizing detrimental impacts of those treatments on native flora and fauna (Johnson and Gjerstad, 2006). The next chapter of this thesis seeks to address this tradeoff by exploring the long-term effects of site preparation on vegetation in longleaf pine plantations.

Chapter 3: Long-Term Vegetation Responses to Site Preparation for Longleaf Pine Restoration on Hydric Sites

Introduction

In recent decades, longleaf pine (*Pinus palustris* Mill.) restoration has become an important land management objective throughout the Southeast, due in large part to the ecological value of the longleaf pine ecosystem. This ecosystem includes some of the most diverse plant communities outside of the tropics (Walker and Silletti, 2006), with the richest savannas containing upwards of 40 vascular plant species in 0.25 m^2 (Walker and Peet, 1983). Many plant species within longleaf pine ecosystems also demonstrate high endemism and rarity. Forty percent of the 1600+ plant species in the Atlantic and Gulf Coastal Plains are restricted to longleaf pine ecosystems (Walker, 1998), and Walker (1993) described 187 rare plant species associated with longleaf pine ecosystems. That count has since been updated to include an additional 256 species, reflecting the continued and increasing endangerment of longleaf pine associates (Glitzenstein et al., 2001). Diversity of longleaf pine ecosystems manifests itself not only within communities but also between them. From xeric, rocky slopes in Alabama to hydric, sandy flatwoods in North Carolina, longleaf pine occupies a wide range of site types. This diversity among longleaf pine communities is perhaps best represented by the 135 longleaf pine associations described by Peet (2006).

Longleaf pine forests may be managed simultaneously for both timber and biodiversity (Freeman and Jose, 2009; Mitchell et al., 2006). The removal of large overstory trees, which provide high quality timber, also serves to increase light penetration into the species-rich understory (Van Lear et al., 2005). Similarly, frequent fire promotes dominance of longleaf pine (Mitchell et al., 2009), increases understory species richness (Glitzenstein et al., 2003; Walker and Peet, 1983), and creates open habitat for game species, such as bobwhite quail (*Colinus virginianus*) and white-tailed deer (*Odocoileus virginianus*), that generate hunting revenue (Landers et al., 1995). Landers et al. (1995) suggested that this pairing of economic and ecological values would be one of the keys to successful restoration of longleaf pine ecosystems across the Southeast. Since that assessment over two decades ago, land management objectives and silvicultural practices have expanded to include conservation of biodiversity in addition to sustained timber yield (Mitchell et al., 2006).

Widespread interest in longleaf pine restoration has underscored the need for research-based guidelines that encompass the wide array of site conditions on which restoration may take place. The range of site types that longleaf pine can occupy, paired with the variety of pre-restoration conditions of those sites, means that site-specific restoration approaches may be needed. Restoration efforts often occur on sites where mature longleaf pine is no longer present or is a small component of the overstory. In such cases, artificial regeneration is necessary to reestablish longleaf pine on the site. This practice has become so common that over a quarter of the modern longleaf pine forest – almost 450,000 ha – was regenerated artificially (Oswalt et al., 2012).

A major impediment to successful establishment of longleaf pine on any site is its intolerance to competition, particularly as a grass stage seedling (Boyer, 1990). This challenge is amplified on wet sites, such as flatwoods, where competition from woody plants is abundant and soil drainage is poor. In order to overcome these challenges, longleaf pine regeneration must be accompanied by some form competition control (Brockway et al., 2006). Longleaf pine would have naturally regenerated on hydric sites under a regime of frequent surface fire, and prescribed fire is often a suitable solution because of the natural relationship between frequent fire and competition reduction. However, sites with heavy competition from woody plants and a history of fire suppression will often require site preparation to successfully establish planted longleaf pine seedlings (Brockway et al., 2006).

Site preparation techniques can be divided into two categories based upon their primary purpose: vegetation control or soil manipulation (Morris and Lowery, 1988). Treatments designed to reduce competing vegetation include herbicide application and mechanical chopping (Johnson and Gjerstad, 2006). Bedding and mounding are mechanical treatments that may also provide some vegetation control (Johnson and Gjerstad, 2006). However, their primary objectives are to manipulate the soil to overcome adverse conditions, such as poor drainage and a high water table, and to improve the rooting environment for planted seedlings by increasing soil temperature and aeration (Johnson and Gjerstad, 2006; Londo and Mroz, 2001; Lowery and Gjerstad, 1991).

Although site preparation is applied with planted seedlings in mind, it is critical that we understand and account for the impacts of site preparation on the plant community. In addition to promoting survival and growth of planted longleaf pine seedlings, site preparation may also enhance the plant community by reducing woody species that would otherwise decrease herbaceous richness and diversity (Litt et al., 2001). However, there is widespread concern that intense mechanical site preparation treatments, such as bedding and mounding, may negatively impact native understory plants (Brockway et al., 2005, 2006; Mitchell et al., 2006; Platt et al., 2015; Van Lear et al., 2005). Similarly, herbicides are designed to control certain plants but may also affect non-target plants. While we expect short term reductions in vegetation abundance, deleterious, and even perhaps long-term, impacts on non-target species may occur. Therefore, Johnson and Gjerstad (2006) suggest that when restoration of the entire longleaf pine ecosystem is the objective, there must be balance between maximizing site preparation that promotes seedling establishment and minimizing detrimental impacts of those treatments on native flora and fauna. Van Lear et al. (2005) argue that, ultimately, the benefits of active management far outweigh the cost of doing nothing and that without such active management, the longleaf pine ecosystem is doomed.

There is a paucity of studies that examine vegetation responses to mechanical site preparation treatments in longleaf pine ecosystems. Bedding has been shown to reduce grass cover in hydric slash pine (*Pinus elliottii* Engelm.) plantations (Pritchett, 1979), which is consistent with the findings of Knapp et al. (2008) on the same hydric longleaf pine plantations as those in this study. This reduction in grasses is concerning because of the critical role that grasses play in carrying surface fire through longleaf pine communities. In fact, Knapp et al. (2008) found that mounding had even more intense impacts than bedding. On mounds, cover of graminoids was less than a third and cover of shrubs less than half of that on the control two years after treatment application. In the same study, least square mean values of graminoid and shrub cover on chop treatments were comparable to those of bedding. Another poorly understood, yet important, aspect of soil manipulation treatments is their local impact on vegetation. Because untreated areas remain after bedding and mounding, impacts on species richness or diversity at the broader scale may go undetected. Therefore, understanding local responses on treated versus untreated areas will provide a more accurate understanding of how the plant community responds to soil manipulation treatments. Quantifying local responses to soil manipulation is especially important for evaluating impacts on species of concern, such as wiregrass, which may be particularly sensitive to intensive site preparation.

In contrast to mechanical site preparation, there are more studies that examine vegetation responses to herbicide application. A literature review by Litt et al. (2001) found only a few studies have examined vegetation response to herbicide in natural flatwoods, but those that have consistently observed reductions in species richness and cover of herbaceous and woody plants following herbicide application. On flatwoods plantations, Knapp et al. (2008) found that herbicide resulted in a greater reduction of shrub cover than did chopping, while graminoid and forb cover were similar for both treatments. On upland sites, herbicide treatments have been shown to reduce hardwood stem density and height through six years after application (Addington et al., 2012). In the same study, herbaceous vegetation, including bunchgrasses, was not negatively impacted by herbicides, and species richness was similar among herbicide treatments and the control. Freeman and Jose (2009) found that species richness, herbaceous cover, and wiregrass cover were actually higher on plots treated with imazapyr than on untreated controls four years after application in a flatwoods plantation. In the same study, early shrub control provided by herbicide was no longer detectable after four years, but Freeman and Jose (2009) suggested that the initial woody reduction was key for allowing herbaceous plants to establish and persist even after shrubs rebounded.

Investment in site preparation assumes that treatments will result in long-term benefits to the site, yet many studies on site preparation effects report on short-term responses within a few years after treatment. This phase is enough to capture initial vegetation responses to site preparation, but it might not be long enough to explore if and how the plant community recovers. This is particularly important in the context of restoration, where it is desirable for the plant community to move toward a specified reference, or desired, condition over time (Kirkman et al., 2013). This reference condition will vary depending upon site type and geographic location, but re-establishment of frequent fire is a consistent objective for restoring ground flora in longleaf pine communities (Martin and Kirkman, 2009). Thus, site preparation treatments must sustain a frequent fire regime that is continuous in both time and space (Mitchell et al., 2009).

In addition to fire, direct effects of site preparation on longleaf pine may have indirect effects on the understory plant community. Longleaf pine and bunchgrasses together facilitate frequent surface fire, which results in a positive feedback loop that further promotes their dominance (Brockway et al., 2005; Mitchell et al., 2009). However, this facilitation will likely be weaker at first when seedlings have little foliage and needle litter is sparse. On the other hand, herbaceous species have been shown to respond positively to increased understory light transmittance beneath a longleaf pine canopy (Platt et al., 2006). In both cases, the effects of longleaf pine on the understory change through time – as they shed more needles and develop wider crowns – demonstrating the importance of understanding how site preparation's impacts on overstory structure might relate to understory composition.

The objective of this study was to quantify long-term ground layer responses to site preparation treatments on poorly drained, hydric sites. Using data collected through three years and at 15 years after plantation establishment, we sought to address three specific questions: (1) How does site preparation impact diversity, abundance, and composition of the plant community through 15 years after treatment application? (2) What are the long-term local effects of bedding on understory vegetation? (3) How does site preparation impact overstory canopy structure and, thus, understory light transmittance?

Methods

Study Area

This study was conducted on Marine Corps Base Camp Lejeune (34°36'N 77°24'W) in Onslow County, North Carolina. Camp Lejeune is located within the Atlantic Coastal Flatwoods section of the Outer Coastal Plain Mixed Forest province (McNab et al., 2007). The study sites were located on Leon sand (sandy, siliceous, thermic Aeric

Alaquod), a poorly drained Spodosol with a fluctuating water table that may be at or near the surface (Barnhill, 1992; NRCS, 2014). Spodosols are the primary soil order of flatwoods and are characterized by their sandy, acidic, and infertile nature (Brockway et al., 2005; Peet, 2006). Leon sand is one of the most extensive soil series on Camp Lejeune (Barnhill, 1992; Frost, 2001) and is of large extent throughout the Southeast (NRCS, 2014). Timber production has been a primary land use objective throughout Camp Lejeune's history (MCBCL, 2015), and the sites selected for this study previously contained planted slash and loblolly pine (*Pinus taeda* L.), which were harvested six months to two years prior to treatment installation.

The natural vegetation community on frequently burned Leon sand in this area is species-rich longleaf pine wet savanna (Frost, 2001). This bilayered community consists of an overstory dominated by longleaf pine and an understory containing a diverse array of grasses, sedges, forbs, and fire-dwarfed shrubs (Frost, 2001). Wiregrass (*Aristida stricta* Michx.) dominates the herbaceous layer, but other common graminoids include bluestems (*Andropogon* spp. and *Schizachyrium* spp.), rosette and panic grasses (*Dichanthelium* spp.), and yellow-eyed grasses (*Xyris* spp.). The estimated pre-settlement fire return interval was 1-3 years, and with such frequent fire Leon sand supports rare species such as roughleaf loosestrife (*Lysimachia asperulifolia* Poir.) and Venus flytrap (*Dionaea muscipula* Ellis) (Frost, 2001). These sites also typically contain other insectivorous plants, such as pitcher plants (*Sarracenia* spp.) and sundews (*Drosera* spp.). Common shrubs include inkberry (*Ilex glabra* (L.) A. Gray), dwarf and blue huckleberries (*Gaylussacia dumosa* and *frondosa* (Andrews) Torr. & A. Gray), piedmont staggerbrush (*Lyonia mariana* (L.) D. Don), and a collection of blueberry species (*Vaccinium* spp.). Wet longleaf pine savannas are associated on downslopes by pond pine (*Pinus serotina* Michx.) pocosin communities, which have persistent shrub cover beneath a sparse overstory of pond pine (*Pinus serotina* Michx.) (Frost, 2001).

Table 1. Summary of site preparation treatments implemented in the study.

Treatment		Vegetation Control		Soil Manipulation		
		Chop	Herbicide	Flat	Mound	Bed
Flat [control]	(F)			X		
Chop/flat	(CF)	X		X		
Herbicide/flat	(HF)		X	X		
Chop/mound	(CM)	X			X	
Herbicide/mound	(HM)		X		X	
Chop/bed	(CB)	X				X
Herbicide/bed	(HB)		X			X
Chop/herbicide/bed	(CHB)	X	X			X

Experimental Design

This study utilized a randomized complete block design, consisting of eight treatments replicated across five blocks for a total of 40 experimental units. Study treatments were randomly assigned to experimental units with dimensions of 54.6 m x 86.3 m (almost 0.5 ha), with 20 m buffers between units to prevent treatment overlap and reduce edge effects. Study treatments consist of combinations of two vegetation control treatments (herbicide and chopping) and three soil manipulation treatments (mounding, bedding, and flat-planting [no additional treatment]) to create a 2 x 3 factorial design of six treatments. The two additional study treatments were an untreated check (no vegetation control and flat-planting) and a bedding treatment with both levels of vegetation control. The eight resulting study treatments (Table 1) are often referred to by their initials: F (flat-planting and no vegetation control), HF (herbicide and flat-planting), CF (chopping and flat-planting), HM (herbicide and mounding), CM (chopping and mounding), HB (herbicide and bedding), CB (chopping and bedding), and CHB (chopping, herbicide, and bedding).

Prior to site preparation, all blocks were harvested and sheared to remove standing vegetation. Study treatments were then applied in August 2003. The chopping treatment was implemented using a 2.4 m Lucas Drum Chopper pulled by a TD15 Dresser crawler tractor. The herbicide treatment included 1.54 lb/ha of imazapyr and 1.24 lb/ha of triclopyr, which were mixed and broadcast at a rate of 280 l/ha. Mounds approximately 1.2 m wide were created using a New Forest Technology custom mounding bucket on a Caterpillar 320BL excavator. For consistency, the mounds were created in rows, rather than in the discontinuous, random distribution that is commonly associated with mounding site preparation. Beds approximately 2.1-2.4 m wide were created using a Rome 6-disc Bedding Harrow with three discs on each side. All blocks were burned in October or November 2003 to remove remaining vegetation and further prepare the sites for planting. Container-grown longleaf pine seedlings from locally collected seed were hand planted in December 2003. All blocks were burned three times (2006, 2011-12, and 2016-18) after planting at roughly a five-year fire return interval.

Data Collection

We used stratified random sampling to locate 12 1 m x 1 m quadrats within each experimental unit. We did so by establishing four equally spaced transects that ran longways across each rectangular experimental unit and then randomly locating three quadrats along each transect. Every vascular plant within the quadrat was identified to species, except for a few cases where discrimination in the field was impossible. Percent cover of each species or group of species was recorded using Carolina Vegetation Survey cover classes (Peet et al., 2012). These data were collected each summer for three years after plantation establishment (2004, 2005, and 2006), and the summer of 2018, which was 15 years after plantation establishment. The same quadrats were used for the first three years, but at year 15 new quadrats were randomly established following the same protocol because the original quadrats could not be re-located.

In summer 2018, each quadrat in bedded treatments was paired with an adjacent quadrat to determine the local effects of bedding. One quadrat was positioned on the bedding row, where trees were planted, and the other quadrat was positioned between bedding rows. The area between bedding rows had unaltered soil and will henceforth be referred to as the interbed area. The same sampling protocol outlined above was used for quadrats on both the beds and the interbed areas.

In summer 2018, a hemispherical photograph was taken at the center of each quadrat to quantify light transmittance to the understory. We positioned a camera at 1.5 m above the ground with the lens parallel to the ground, and we used a fisheye lens to capture a 180° image. All photographs were taken on cloudy days to reduce direct sun reflection. We used the HemiView Forest Canopy Image Analysis System (Delta-T Devices, Cambridge, UK) to process the photographs and obtain estimates of gap light index and gap fraction for each quadrat. Gap light index estimates the percentage of incident photosynthetically active radiation (PAR) through a gap to a point in the understory (Canham, 1988), while gap fraction estimates the percentage of sky unobstructed by the canopy from a point in the understory (Welles and Cohen, 1996). Both gap light index and gap fraction have been shown to accurately estimate canopy light transmittance in longleaf pine ecosystems (Battaglia et al., 2003).

Data Analysis

We obtained percent cover of functional groups by obtaining the mean value of the recorded cover class range (e.g., 7.5% for a range of 5-10%) and summarizing those values for all species within the functional group. We categorized species into six main functional groups: ferns, forbs, graminoids, shrubs, trees, and vines. We also further divided forbs into annuals and perennials, graminoids into bunchgrasses, sedges and rushes, and other graminoids, and shrubs into tall and short shrubs. The only species that we analyzed individually was wiregrass because of its ecological value and functional role in longleaf pine ecosystems. We fit linear mixed models to each response variable of interest (i.e., percent cover of each functional group), treating treatment and time as fixed effects and the hierarchical, nested sampling design (i.e., experimental units within blocks) as a random effect. We then used repeated measures analysis of variance (ANOVA) to test for differences among treatments and through time and to test for interactions between treatment and time. When repeated measures ANOVA indicated that main effect or interaction terms were significant ($\alpha = 0.05$), post-hoc Tukey-adjusted pairwise comparisons were used to determine which treatments and time periods significantly differed. Additionally, for the measurements taken in 2018, we used a 2 x 3 factorial ANOVA, excluding the F and CHB treatments, to determine the effect of individual factors (e.g., chopping vs. herbicide) on each response variable and to test for interactions between the vegetation control treatments and soil manipulation treatments. When factorial ANOVA indicated that main effects (i.e., vegetation control or soil manipulation treatments) were significant (α = 0.05) and the interaction was not, post-hoc Tukey-adjusted pairwise comparisons were used to determine which individual factors significantly differed.

We calculated three measures of diversity using the data collected in 2018: species richness, Shannon entropy transformed into a diversity index (or Hill number), and evenness. Unlike for cover, we did not integrate the most recent diversity measurements with those from the first three years because some species were lumped into groups differently during the two time periods. Species richness was calculated by counting the total number of plant species present in each 1 m^2 quadrat. We used relative cover of each species in each quadrat as a proxy for abundance when calculating Shannon entropy and evenness. Therefore, Shannon entropy (H) was calculated using the following equation, where p_i is the relative cover of a species.

$$H = \sum_{i=1}^{s} p_i \, ln \, p_i$$

To convert Shannon entropy into a diversity index, or Hill number, we took the exponent of Shannon entropy (Jost, 2006). To calculate evenness, we divided Shannon entropy by the natural logarithm of species richness, so that each species is assigned a value between 0 and 1, where 1 indicates a completely even distribution of all species. For each of these three measures of diversity, we fit linear mixed models, treating treatment as a fixed effect and block as a random effect. We then conducted one-way ANOVA on each of the fitted mixed models to test for differences among the eight study treatments for each measure of diversity. When one-way ANOVA indicated that treatments significantly differed (α = 0.05), post-hoc Tukey-adjusted pairwise comparisons were used to determine which treatments significantly differed. We also conducted a 2 x 3 factorial ANOVA, excluding the F and CHB treatments and followed by post-hoc Tukey-adjusted pairwise comparisons, to determine the effects of individual treatment levels on richness, diversity, and evenness.

We also fit linear mixed models to gap light index and gap fraction estimates obtained from hemispherical photographs following the same approach outlined for vegetation cover and diversity. We conducted one-way ANOVA to test for differences among the eight study treatments and 2 x 3 factorial ANOVA, excluding F and CHB, to determine effects of individuals treatment levels on canopy cover and gap light index. When one-way or factorial ANOVA indicated significant differences among treatments or factors, post-hoc Tukey-adjusted pairwise comparisons were used to determine which treatments or factors significantly differed. To test for differences in vegetation between bedding rows and interbed areas, we conducted paired *t*-tests for all of the cover, diversity, and light transmittance response variables outlined above.

Data were analyzed in R (R Core Team, 2018). The *lme* function in the *nlme* package (Pinherio et al., 2018) was used for fitting linear mixed models. The *anova* function and the *t.test* function in the *stats* package (R Core Team, 2018) were used for running ANOVA and paired *t*-tests, respectively. The *lsmeans* function from the *emmeans* package (Lenth, 2019) and the *CLD* function from the *multcompView* package (Graves et al., 2015) were used for post-hoc pairwise comparisons. When factorial or repeated measures ANOVA resulted in a significant ($\alpha < 0.05$) interaction term, we used the *testInteraction* function in the *phia* package (De Rosario-Martinez, 2015) to conduct post-hoc interaction significance tests. The *dplyr* package (Wickham et al., 2019) was used for data manipulation and the *ggplot2* package (Wickham, 2016) was used for figure creation.

Results

Vegetation Cover

Based on repeated measures ANOVA, the interaction between treatment and time was significant ($p < 0.001$) for total vegetation cover through 15 years after treatment application. Post-hoc interaction significance tests indicated that there were significant differences in total cover among the eight study treatments for the first two years after application ($p < 0.001$) (Fig. 8A). In 2004, F and CF had higher total cover than HM, HB, and CHB, and in 2005, F, CF, and CB had higher cover than HM, HB, and CHB. However, in years three and 15, those differences were no longer significant ($p \geq 0.246$). On all treatments, total cover in 2005 and 2018 was significantly higher than in 2004 ($p < 0.001$). For most treatments, total cover during 2006 was also significantly higher than in 2004; however, F and CF had similar total cover during the first and third years, likely due to the prescribed burn that occurred in 2006.

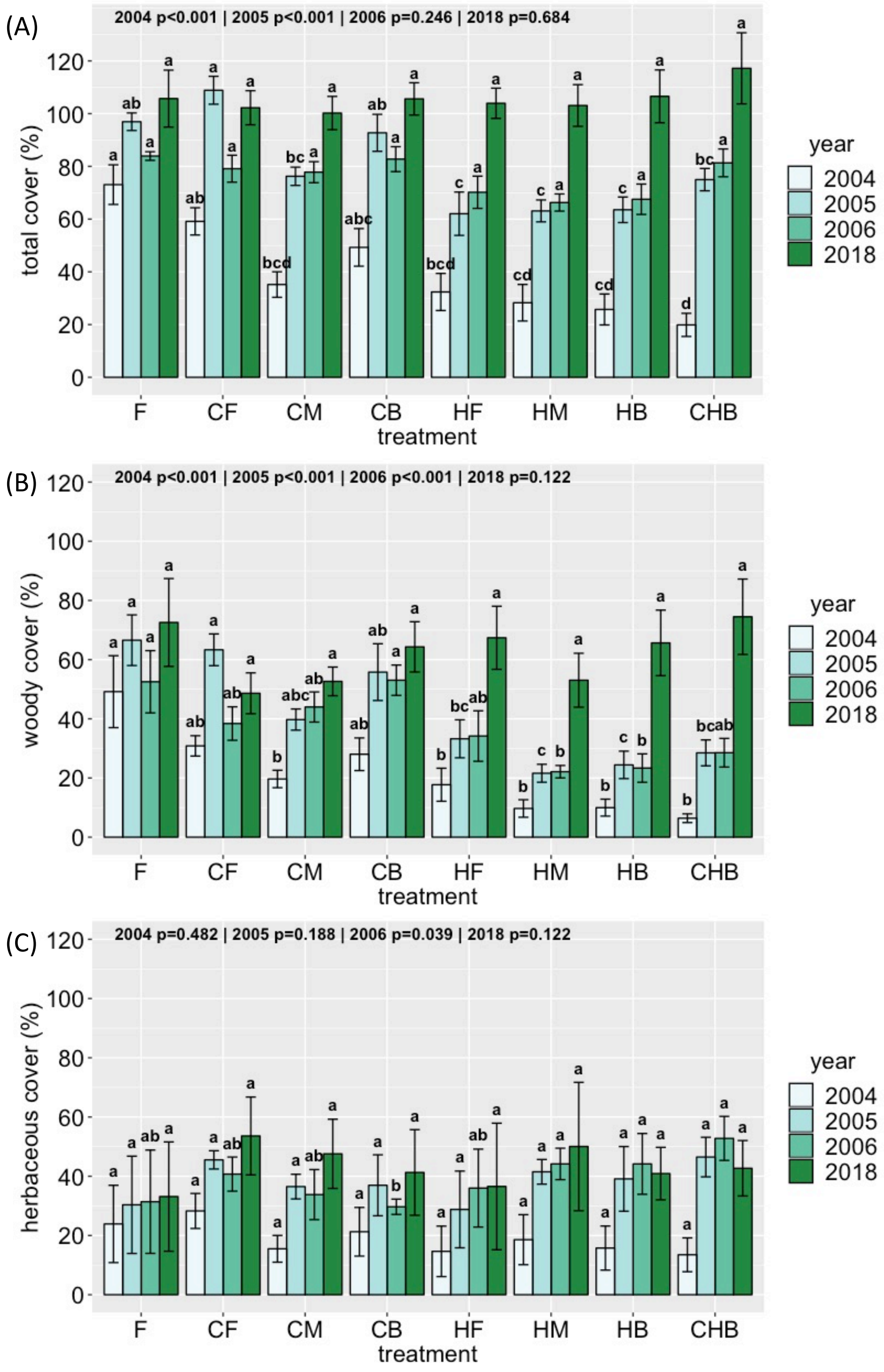

Figure 8. Total, woody, and herbaceous plant cover through 15 years after site preparation. Treatments are defined in Table 1. Same letters indicate no significant difference ($\alpha = 0.05$) among treatments within each measurement year and response based upon Tukey-adjusted pairwise comparisons; *p*-values are from repeated measures ANOVA post-hoc interaction significance tests.

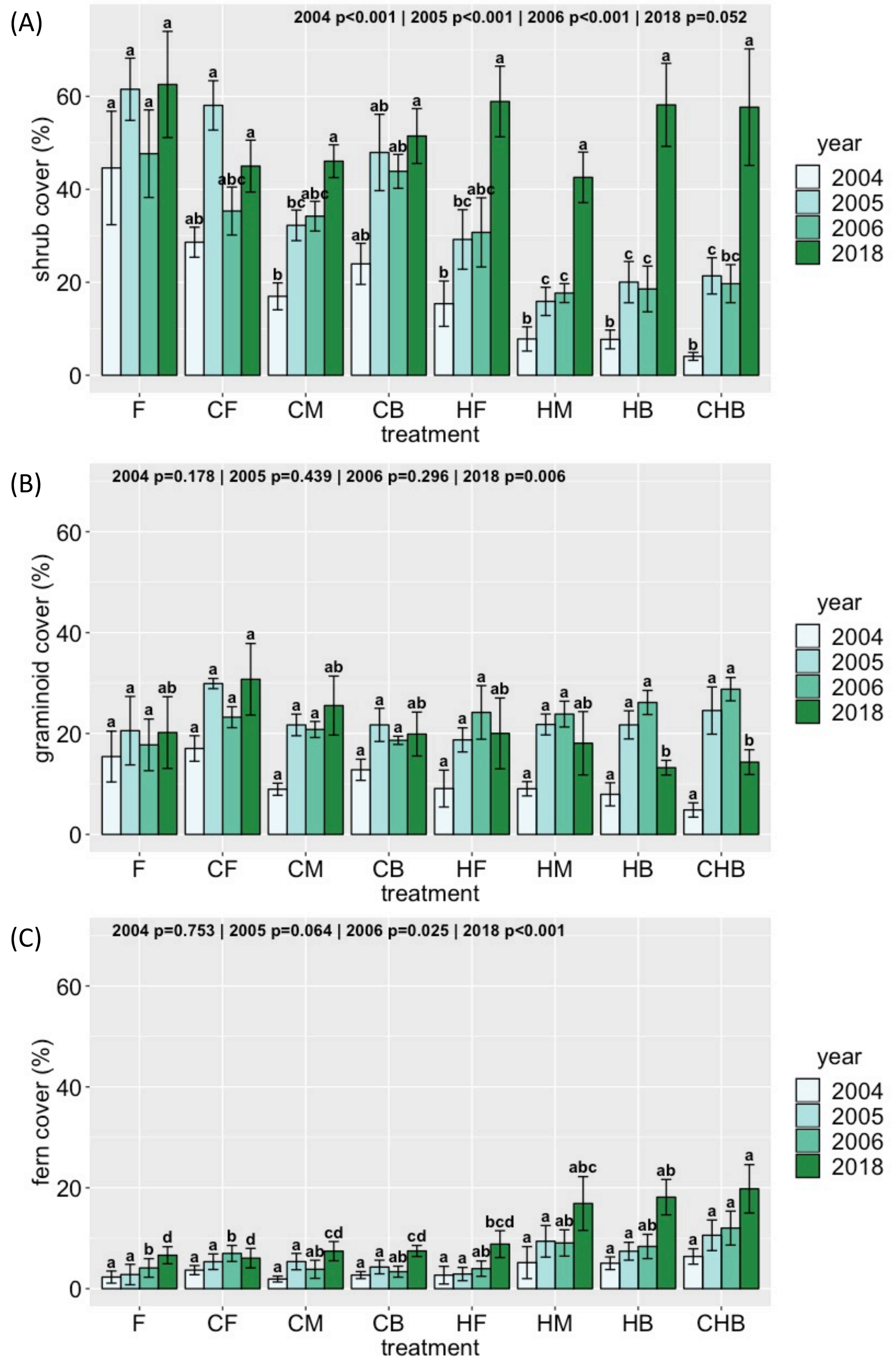

Figure 9. Cover of shrubs, graminoids, and ferns through 15 years after site preparation. Treatments are defined in Table 1. Same letters indicate no significant difference (α = 0.05) among treatments within each measurement year and response based upon Tukey-adjusted pairwise comparisons; *p*-values are from repeated measures ANOVA post-hoc interaction significance tests.

The interaction between treatment and time was also significant for cover of woody plants ($p < 0.001$) and shrubs ($p < 0.001$). Because shrubs made up the majority of woody plants in this study, the patterns between cover of woody plants and shrubs were similar (Fig. 8B and Fig. 9A). Significant differences in both woody ($p < 0.001$) and shrub ($p < 0.001$) cover existed among the study treatments during the first three years. In 2004, the untreated control had greater woody and shrub cover than all other treatments, except for CF and CB. In 2005, F and CF had greater woody and shrub cover than HF, CHB, HB, and HM. And in 2006, F and CB had greater cover than HB and HM. However, by year 15, differences in woody and shrub cover among treatments were no longer significant ($p \geq 0.052$). Similar to total cover, both woody and shrub cover tended to increase through time, with a few exceptions after the prescribed burn in 2006. The four treatments with herbicide had dramatic increases in woody and shrub cover between year three and 15, whereas increases in the non-herbicide treatments were more gradual, with the greatest increases in woody and shrub cover typically occurring between the first and second years after treatment.

Repeated measures ANOVA also indicated that the interaction between treatment and time was significant for herbaceous cover ($p = 0.026$), graminoid cover ($p = 0.023$), and fern cover ($p = 0.014$). For herbaceous cover, the only year in which treatments significantly differed was 2006 ($p = 0.039$), at which point HM, HB, and CHB had greater cover than CB (Fig. 8C). For graminoid cover, the only year in which treatments significantly differed was 2018 ($p = 0.006$), when CF had greater cover than CHB and HB (Fig. 9B). Significant differences in fern cover existed in 2006 ($p = 0.025$) and

2018 ($p < 0.001$). In 2006, fern cover was higher on CHB than F and CF, and in 2018, CHB and HB had greater cover than CB, CM, CF, and F (Fig. 9C). Again, both herbaceous and fern cover tended to increase through time, with some exceptions in 2006. However, graminoid cover did not follow this trend. While some treatments did have reduced graminoid cover in 2006, others also experienced significant reductions in graminoid cover between years three and 15. HB and CHB had significantly reduced graminoid cover between years three and 15 by over 13%, and HM and HF experienced reductions of just over 4%, although they were insignificant.

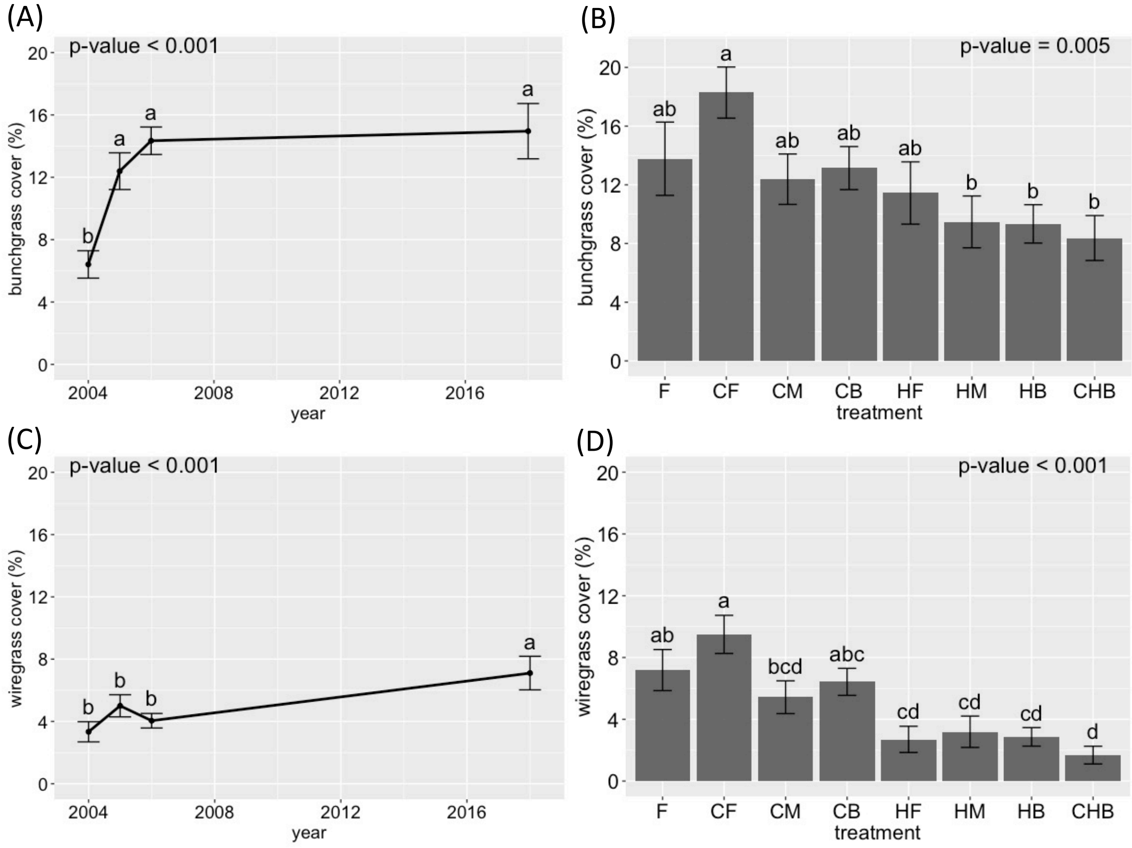

Figure 10. Bunchgrass and wiregrass cover through 15 years after site preparation and among study treatments (defined in Table 1). Same letters indicate no significant difference ($\alpha = 0.05$) among measurement periods or treatments within each panel based upon Tukey-adjusted pairwise comparisons; *p*-values are from repeated measures ANOVA.

To further explore the impacts of treatment and time on graminoids, we also conducted repeated measures ANOVA tests on cover of bunchgrasses and wiregrass. Treatment ($p \leq 0.005$) and time ($p < 0.001$) significantly affected cover of both bunchgrasses, in general, and wiregrass, specifically, and the interaction between treatment and time was insignificant in both cases ($p \geq 0.073$). Bunchgrass cover was significantly greater in 2005, 2006, and 2018 than in 2004 (Fig. 10A), and wiregrass cover was greater in 2018 than during the first three years after treatment application (Fig. 10C). CF resulted in significantly greater bunchgrass cover than HM, HB, and CHB (Fig. 10B), while CF and F resulted in significantly greater wiregrass cover than all four herbicide treatments (Fig. 10D).

Cover of the other three main functional groups we explored – forbs, trees, and vines – also significantly increased through time ($p < 0.001$). Forb cover increases leveled off after year two (Fig. 11A), while vine cover increases leveled off after year three (Fig. 11E). Tree cover increased from year one to year 15 (Fig. 11C). Treatment did not significantly affect forb or vine cover ($p \geq 0.352$) (Fig. 11B and Fig. 11F), but there were significant differences among treatments in tree cover ($p = 0.031$), with CB having significantly greater cover than CF (Fig. 11D). For all three of these functional groups, the interaction between treatment and time was insignificant ($p \geq 0.107$).

Based on factorial ANOVA, vegetation control treatments resulted in significant differences in cover of ferns ($p = 0.001$), forbs ($p = 0.014$), and graminoids ($p = 0.015$) 15 years after treatment application (Table 4). Herbicide resulted in greater cover of ferns compared to chopping, while chopping resulted in greater cover of forbs and graminoids. Both annual and perennial forbs were significantly greater on chopped treatments than herbicide treatments ($p \leq 0.036$). The reduced cover of graminoids as a result of herbicide can be attributed to a significant reduction in bunchgrasses

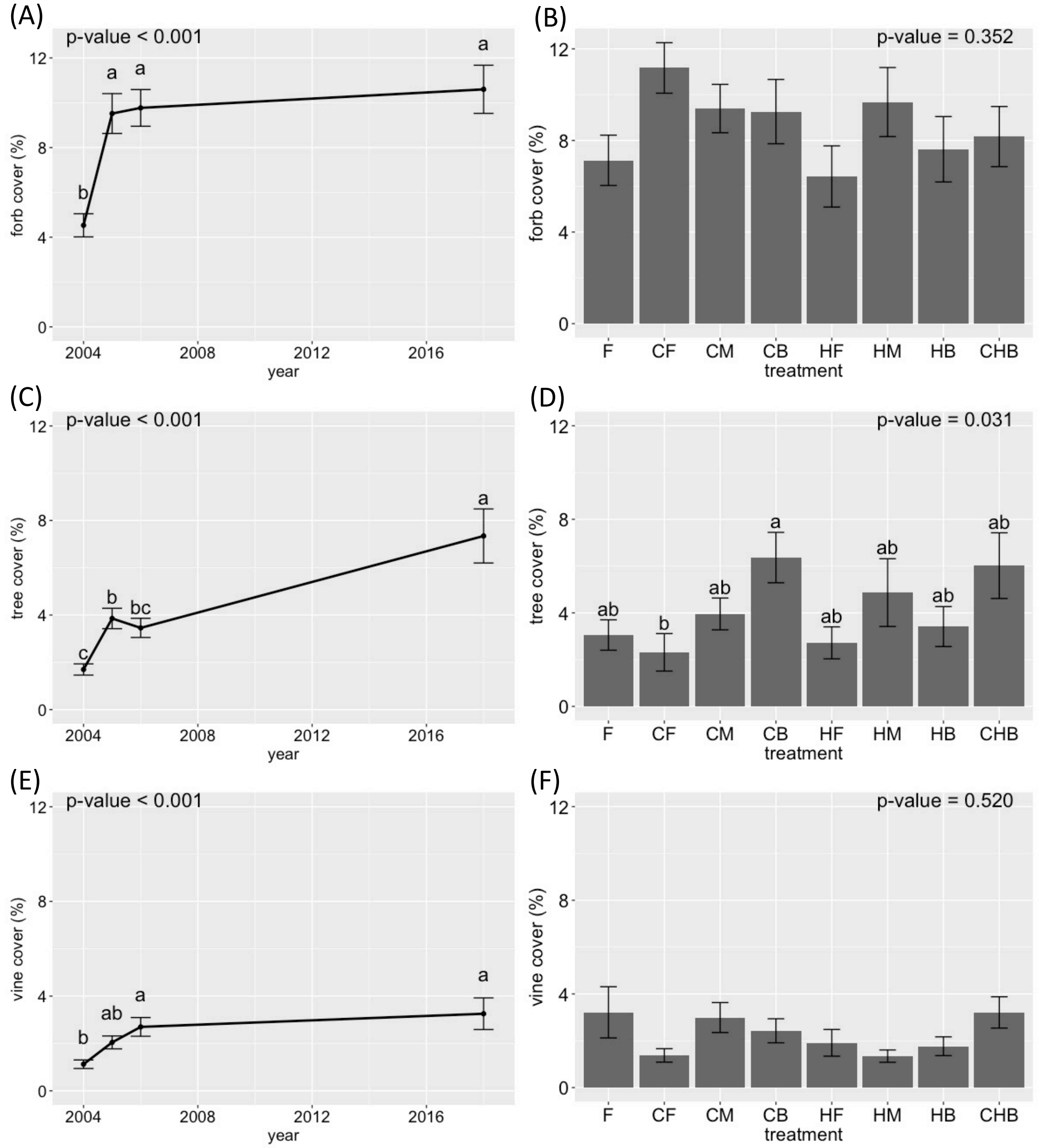

Figure 11. Forb, tree, and vine cover through 15 years after site preparation and among study treatments (defined in Table 1). Same letters indicate no significant difference (α = 0.05) among measurement periods or treatments within each panel based upon Tukey-adjusted pairwise comparisons; *p*-values are from repeated measures ANOVA.

compared to chopping (p =0.003), and much of the bunchgrass reduction is accounted for by a reduction in wiregrass ($p = 0.002$). Total cover and cover of all the other functional groups we explored did not differ between chopping and herbicide ($p \geq 0.083$).

Factorial ANOVA also indicated that cover of shrubs ($p = 0.034$) and wiregrass ($p = 0.024$) significantly differed among soil manipulation treatments 15 years after application (Table 4). Shrubs had significantly greater cover on bedded treatments than on mounded treatments, with flat-planted treatments not differing significantly from bedding or mounding. Wiregrass cover was significantly higher on flat-planted treatments than mounded and bedded treatments by over 4%. Total cover and cover of all other functional groups tested did not significantly differ among soil manipulation treatments 15 years after application ($p \geq 0.085$).

Additionally, there were no significant interactions between vegetation control and soil manipulation treatments for cover of any functional group tested ($p \geq 0.238$), although interactions for cover of wiregrass ($p = 0.051$), shrubs ($p = 0.099$), and tall shrubs ($p = 0.057$) approached significance. Greater wiregrass cover on flat-planted treatments compared to mounded and bedded treatments was largely due to high cover on flat-planted treatments crossed with chopping. When flat-planting was crossed with herbicide, wiregrass cover was similar to mounding and bedding with herbicide ($p = 0.750$). Similarly, interaction significance tests indicated that chopping and herbicide only differed on flat-planted treatments ($p < 0.001$), not on mounded ($p = 0.710$) or bedded treatments ($p = 0.079$), although the latter approached significance.

Table 4. Least square means of vegetation responses to individual site preparation treatment levels 15 years application.

	Soil Manipulation				Vegetation Control		
	Flat	Mound	Bed	*p*-value	Chop	Herbicide	*p*-value
Cover							
Total	103	102	106	0.775	103	105	0.723
Herbaceous	45.1	48.8	41.1	0.480	47.5	42.5	0.340
Woody	58.0	52.8	65.0	0.144	55.2	62.0	0.171
Ferns	7.4	12.2	12.8	0.085	7.0 b	14.6 a	**0.001**
Forbs	12.0	13.3	10.4	0.446	14.4 a	9.4 b	**0.014**
Annual forbs	0.72	0.34	0.31	0.260	0.71 a	0.21 b	**0.036**
Perennial forbs	10.9	12.6	9.9	0.469	13.4 a	8.9 b	**0.020**
Graminoids	25.4	21.8	16.6	0.089	25.4 a	17.1 b	**0.015**
Bunchgrasses	19.5	15.7	12.2	0.067	19.8 a	11.9 b	**0.003**
Wiregrass	10.6 a	6.2 b	6.3 b	**0.024**	10.1 a	5.3 b	**0.002**
Sedges and rushes	3.1	3.6	2.2	0.469	3.1	2.8	0.782
Other graminoids	2.8	2.4	2.1	0.812	2.4	2.4	0.981
Shrubs	51.9 ab	44.3 b	54.8 a	**0.034**	47.5	53.2	0.083
Tall shrubs	33.1	30.2	36.3	0.425	30.9	35.4	0.236
Short shrubs	18.9	14.1	18.6	0.279	16.6	17.8	0.654
Trees	4.6	7.3	8.7	0.303	5.9	7.8	0.366
Vines	1.8	2.9	2.8	0.477	2.7	2.4	0.707
Diversity							
Richness	17.3	16.5	15.8	0.409	17.3 a	15.7 b	0.079
Diversity (Hill number)	7.18	7.41	6.78	0.376	7.85 a	6.40 b	**< 0.001**
Evenness	0.67	0.69	0.68	0.260	0.70 a	0.66 b	**< 0.001**
Light Transmittance							
Gap light index	0.90	0.86	0.86	0.142	0.92 a	0.83 b	**< 0.001**
Gap fraction	0.74	0.67	0.69	0.058	0.76 a	0.66 b	**< 0.001**

Same letters indicate no significant difference ($\alpha = 0.05$) within a treatment type and response variable based upon Tukey-adjusted pairwise comparisons; *p*-values are from 2 x 3 factorial ANOVA tests.

Plant Species Diversity

Fifteen years after site preparation, plant species richness did not significantly differ among the eight study treatments ($p = 0.259$) (Fig. 12A). Across all treatments, mean richness was 16.1 species per 1 m^2. Similarly, factorial ANOVA indicated that neither soil manipulation ($p = 0.409$) nor vegetation control treatments ($p = 0.079$)

significantly impacted species richness (Table 4), and there was no significant interaction between the two treatment types ($p = 0.446$). In contrast, there were significant differences in diversity ($p = 0.006$) and evenness ($p = 0.022$) among the eight study treatments at year 15. Diversity was significantly greater on CF, which had a mean of 8.22, than F, HF, and HB, which had mean diversities of slightly greater than 6 (Fig. 12B). Evenness was significantly greater on CF, which had a mean of 0.71, than HF, which had a mean of 0.63 (Fig. 12C). Differences in diversity and evenness among vegetation control treatments were significant ($p < 0.001$), with chopping resulting in higher diversity and evenness than herbicide (Table 4). There were no significant differences in soil manipulation treatments ($p \geq 0.260$) and no significant interaction ($p \geq 0.160$) for either diversity or evenness.

Light Transmittance

Site preparation treatments also produced significant differences in understory light transmittance 15 years after application ($p < 0.001$). Gap light index and gap fraction were significantly greater on F and CF than on HF, HM, HB, and CHB, with differences in gap light index exceeding 0.10 (Fig. 13A) and in gap fraction approaching 0.15 (Fig. 13B). Vegetation control treatments significantly affected both measures of light transmittance ($p < 0.001$), with chopping resulting in more light than herbicide (Table 4). Differences in soil manipulation treatments were not significant for either gap light index ($p = 0.142$) or gap fraction ($p = 0.058$), although the latter approached significance. Additionally, the interaction between treatment types was insignificant for both measures of light transmittance ($p \geq 0.123$).

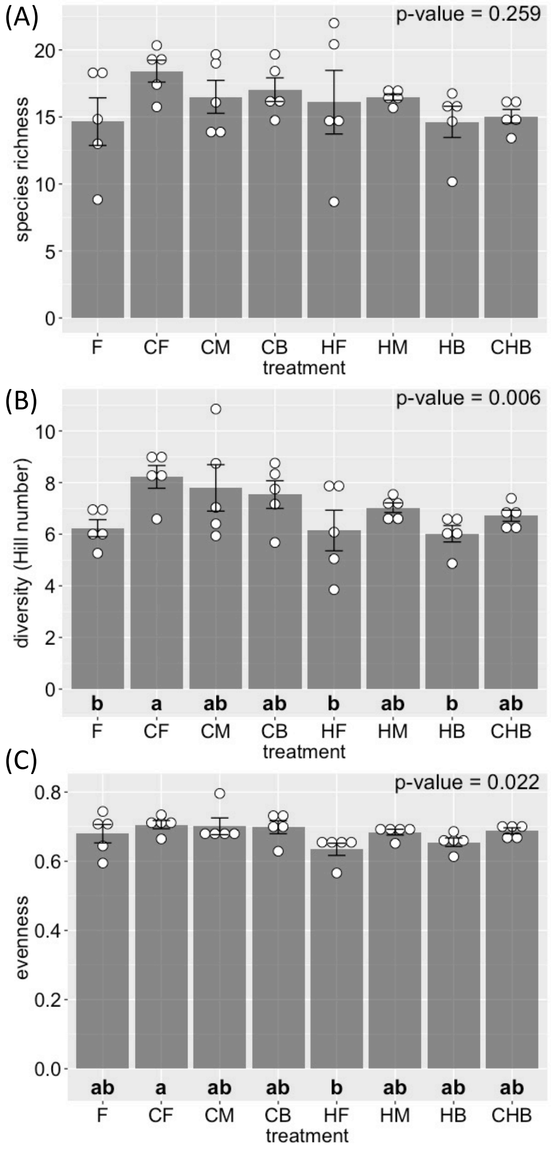

Figure 12. Measures of plant species diversity 15 years after site preparation. Treatments are defined in Table 1. Same letters indicate no significant difference (α = 0.05) among treatments within a response variable based upon Tukey-adjusted pairwise comparisons; *p*-values are from one-way ANOVA tests.

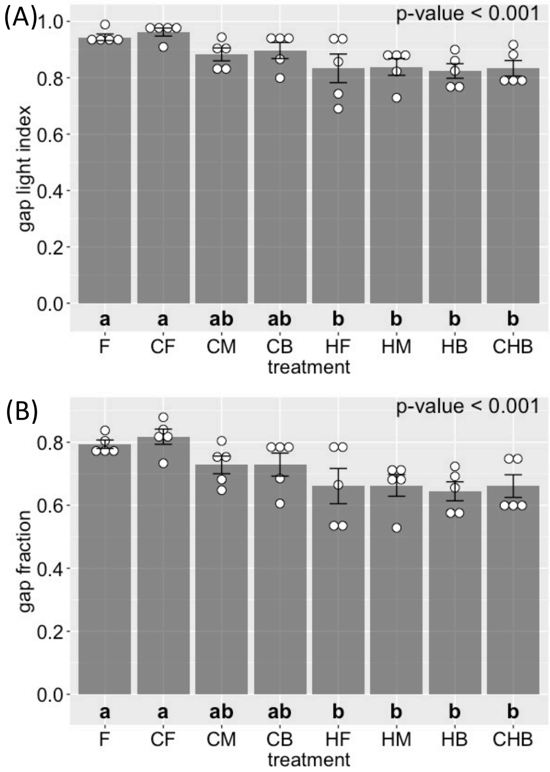

Figure 13. Estimates of (A) gap light index and (B) gap fraction obtained from hemispherical photos taken 15 years after site preparation. Treatments are defined in Table 1. Same letters indicate no significant difference (α = 0.05) among treatments within a response variable based upon Tukey-adjusted pairwise comparisons;

Local Effects of Bedding

Paired *t*-tests indicated that total vegetation cover and cover of some functional groups differed significantly between the bedding rows and interbed areas (Table 5).

Total plant cover was almost 10% higher on beds compared to interbed areas (p = 0.014). This difference was accounted for by significantly greater cover of woody species on beds (72.6%) compared to interbeds (58.8%) (p < 0.001). The difference in cover of herbaceous species between the two locations approached significance (p = 0.055), with interbed areas (44.5%) having slightly higher mean cover than beds (39.6%). In terms of the five main functional groups we explored, four of them – ferns, forbs, shrubs, and vines – did have significantly different cover between bedding rows and interbed areas. The difference in shrub cover approached significance (p = 0.077), but the other three groups did not ($p \geq 0.193$).

The other two main functional groups – trees and graminoids – did have significantly different cover on beds compared to interbeds (Table 5). Tree cover on beds was over 8% greater compared to interbeds (p < 0.001), while graminoid cover was over 3% greater on interbeds compared to beds (p = 0.023). The significant difference in graminoid cover can be attributed to bunchgrasses, and wiregrass in particular, as both bunchgrasses as a whole and wiregrass in particular had about 3% higher cover on interbeds compared to beds ($p \leq 0.021$). Annual forbs had significantly higher cover on interbeds compared to beds (p = 0.011), although the mean difference was less than 0.2%. There were no significant differences in cover for perennial forbs, tall shrubs, or short shrubs ($p \geq 0.130$).

Paired *t*-tests also indicated that there were no significant differences in plant species richness, diversity, or evenness between bedding rows and interbed areas ($p \geq$ 0.511) (Table 5). There were, however, significant differences in understory light transmittance. Gap light index and gap fraction were higher on interbeds than on beds ($p \leq 0.009$), although the mean differences for both estimates were relatively small in magnitude (Table 5).

Table 5. Means and standard errors for measures of diversity, percent cover, and light transmittance on bedded rows and interbed areas 15 years after treatment application.

	Bedding Row		Interbed Area		
	Mean	SE	Mean	SE	*p*-value
Cover					
Total	112.2	3.6	103.3	2.8	**0.014**
Herbaceous	39.6	1.9	44.5	2.0	0.055
Woody	72.6	3.5	58.8	2.9	**< 0.001**
Ferns	15.6	1.4	14.9	1.4	0.629
Forbs	7.3	1.1	9.1	0.9	0.193
Annual Forbs	0.15	0.03	0.34	0.08	**0.011**
Perennial Forbs	6.8	1.1	8.5	0.9	0.200
Graminoids	15.1	1.2	18.7	1.4	**0.023**
Bunchgrasses	9.9	1.0	13.3	1.3	**0.005**
Wiregrass	4.3	0.8	7.2	1.1	**0.021**
Sedges and rushes	2.3	0.5	2.9	0.6	0.376
Other graminoids	3.0	0.4	2.5	0.5	0.437
Shrubs	56.9	2.6	52.1	2.5	0.077
Tall shrubs	34.0	2.2	31.9	2.1	0.424
Short shrubs	22.9	2.0	20.2	1.8	0.130
Trees	13.8	1.8	5.2	1.2	**< 0.001**
Vines	3.6	0.5	3.2	0.5	0.427
Diversity					
Richness	15.6	0.3	15.5	0.3	0.708
Diversity (Hill number)	6.54	0.17	6.69	0.20	0.511
Evenness	0.67	0.01	0.67	0.01	0.747
Light transmittance					
Gap light index	0.84	0.01	0.86	0.01	**0.007**
Gap fraction	0.67	0.01	0.68	0.01	**0.009**

The *p*-values are from paired *t*-tests between bedding rows and interbed areas; significant differences ($\alpha = 0.05$) are shown in bold.

Discussion

Restoration of longleaf pine ecosystems typically intends to move a site towards a specified reference condition (Kirkman et al., 2013). The understory composition of this reference will vary widely depending upon site type and geographic location (Peet, 2006), but the reference has consistent structural and functional conditions, such as a longleaf pine overstory, an understory dominated by grasses, and a frequent fire regime. For this study, the natural vegetation community described by Frost (2001) for Leon sand on Camp Lejeune can be used as a reference condition: a wet savanna dominated by longleaf pine in the overstory and wiregrass in the understory, with a diverse array of other grasses, sedges, forbs, and fire-dwarfed shrubs. We did not identify or quantify a specific reference community against which to compare the communities in our study treatments; however, the natural vegetation community for this site type can still be used as a general reference against which to compare the restoration outcomes in our study treatments.

Herbicides are commonly used as a form of site preparation because of their ability to target specific plant groups (Walker and Silletti, 2006), and through the first three years, herbicide reduced cover of woody plants and shrubs without significantly reducing cover of herbaceous plants and graminoids. These early reductions in woody plant and shrub cover without reductions in herb and grass abundance are consistent with findings of other studies (Addington et al., 2012; Freeman and Jose, 2009). However, herbicide effects on non-target species in longleaf pine ecosystems are poorly understood (Litt et al., 2001), and in our study, herbicide resulted in long-term deleterious impacts on the plant community. By year 15, early reductions in shrub cover as a result of herbicide were no longer significant, while a reduction in graminoid cover was. On similar flatwoods sites, Freeman and Jose (2009) observed a recovery of shrubs within four years of herbicide application, yet in that study wiregrass cover remained significantly higher on sites treated with imazapyr. In our study, though, herbicide resulted in significant long-term reductions in bunchgrass and wiregrass cover. Additionally, herbicide resulted in lower species diversity, evenness, and forb cover compared to chopping in year 15. Herbaceous plant cover and species richness in year 15 were not reduced by herbicide though, which is consistent with shorter-term findings of others (Addington et al., 2012; Freeman and Jose, 2009).

In contrast to herbicide, the impacts of chopping in our study were fairly tame, particularly in the long term. Although chopping is designed to immediately reduce standing woody vegetation, it often promotes sprouting of woody growth (Lowery and Gjerstad, 1991). In our study, chopping did little to reduce woody or shrub cover in the first three years, especially compared to herbicide. Similarly, Miller (1980) found that chopping resulted in 55% more woody vegetation than windrowing, an alternative mechanical vegetation control treatment. The weak impact of chopping in both that study and ours may be due to the fact that only a single pass of the chopper was applied. A second pass of chopping has been shown to increase growth of longleaf pine (Boyer, 1988), presumably as a result of reduced competition, and Johnson and Gjerstad (2006) recommend chopping a second time in late summer or early fall when carbohydrate reserves in woody plants have diminished.

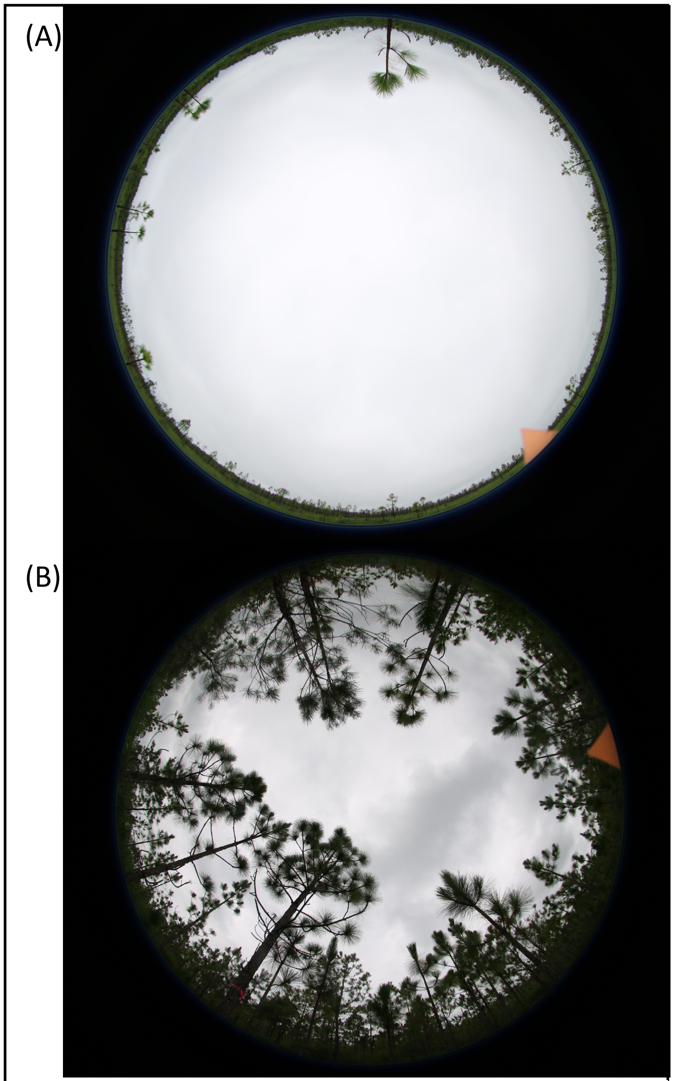

Figure 14. Hemispherical photos to contrast the canopy closure and light transmittance of (A) CF and (B) HB treatments.

Interestingly, the chop-only (CF) treatment in our study resulted in the greatest bunchgrass and wiregrass cover and was among the treatments with the greatest diversity. The advantages of chopping include minimal soil disturbance, low impact on herbaceous vegetation, and drier fuels for fire (Johnson and Gjerstad, 2006). Frequent fire promotes dominance of bunchgrasses (Brockway et al., 2005; Mitchell et al., 2009) and is associated with high species richness in longleaf pine communities (Glitzenstein et al., 2003; Walker and Peet, 1983), so perhaps this last benefit – improved surface fire – could explain why bunchgrass and wiregrass were most abundant and diversity was highest on the CF treatment. Additionally, chopping resulted in significantly more understory light transmittance than herbicide, which could have also contributed to higher diversity on chopping treatments in general and the CF treatment in particular. A third consideration is that CF provided a slight reduction in early shrub cover compared to the untreated control, specifically in the first year after treatment application and in year three after fire. This early shrub reduction could have provided opportunity for herbs and graminoids to develop, as has been suggested after herbicide application (Freeman and Jose, 2009).

Bedding and mounding treatments disrupt the soil surface and may have lasting impacts on the plant communities, especially perennial plants that are physically uprooted. Our findings indicate that mounding and bedding did impact plant communities, particularly when used in conjunction with herbicide. Overall, shrubs, graminoids, and ferns appeared to be most dramatically impacted by soil manipulation treatments. Shrub cover was lowest on the soil manipulation treatments crossed with herbicide – HM, HB, and CHB – through three years after planting. However, by year 15 these three treatment combinations were statistically similar to the other five, demonstrating the long-term recovery of shrubs. Similarly, fern cover significantly increased from years 1-3 to year 15 on treatments where soil manipulation was crossed with herbicide. Interestingly, mounding had significantly lower shrub cover than bedding 15 years after treatment, while flat-planting was similar to both.

Site preparation appeared to have lasting impacts on bunchgrasses and wiregrass, which are considered important fuels in the longleaf pine ecosystem (Mitchell et al., 2009). The eight study treatments had similar graminoid cover through three years after treatment, but by year 15, HB and CHB had significant declines in graminoid cover, and HM also experienced a slight, yet insignificant, decline. Similarly, these three soil manipulation plus herbicide treatments had the lowest bunchgrass and wiregrass cover through time, and in year 15, both bedding and mounding resulted in significantly lower wiregrass cover than no soil manipulation. Wiregrass appeared to be particularly sensitive to site preparation and still had not fully recovered 15 years after treatment. Wiregrass is easily uprooted, rarely reproduces via seed, and propagates itself primarily by clump expansion, all of which make wiregrass ill-suited for recovery after soil disturbance (Clewell, 1989). Locally, bedding rows had significantly lower graminoid, bunchgrass, and wiregrass cover compared to the undisturbed interbed areas at year 15. Had no undisturbed interbed area been maintained between bedding rows, treatment-level reductions in wiregrass would likely have been even more dramatic, which emphasizes the long-term deleterious impact bedding has on wiregrass abundance.

Aside from the reduced cover of graminoids, bunchgrasses, and wiregrass on bedding rows, long-term differences between beds and interbed areas were minimal. Cover of woody plants was significantly higher on beds than interbeds 15 years after treatment, but this difference was largely due to the significantly greater cover of trees on beds, as would be expected because that is where trees were planted. Shrub cover was greater on beds too, although the difference between beds and interbed areas was insignificant. Notably, there were no significant long-term local bedding differences in cover of herbaceous plants or in any of the three measures of plant species diversity. There were also no significant long-term differences in diversity measures between bedding treatments and flat-planted controls, which indicates a recovery of the plant community 15 years after bedding, aside from wiregrass which still had significantly lower cover on bedded and mounded treatments compared to the flat-planted controls.

Gap light index and gap fraction were significantly higher on interbeds compared to beds, which would also be expected because trees were planted on the beds. These differences were extremely small, though, so their biological significance could be questioned. Significant differences in understory light transmittance also existed at the treatment level. Gap light index and gap fraction were significantly higher on F and CF than on HF, HM, HB, and CHB. These findings further emphasize the differences in longleaf pine establishment among treatments, with reduced understory light transmittance occurring on treatments that resulted in more successful longleaf pine survival and growth.

To tie our findings back to restoration and examine how site preparation could move sites toward a reference condition, the conceptual framework of restoration thresholds is helpful to consider (Martin and Kirkman, 2009; Suding and Hobbs, 2009). In restoration scenarios, a threshold often exists between the pre-restoration community (e.g., slash pine-dominated stand lacking overstory longleaf pine or natural regeneration) and the desired community (e.g., the natural vegetation community described earlier). Intensive intervention early on, such as site preparation, may be necessary to overcome the threshold between the two community states, but once the "restoration threshold" is overcome (e.g., longleaf pine is successfully established and shrubs are controlled), less intense treatment (e.g., regular prescribed fire) may be all that is required to maintain the restored, desired community state (Martin and Kirkman, 2009).

During the first three years after plantation establishment, study treatments with herbicide appeared to significantly reduce cover of shrubs and woody plants while cover of graminoids and herbaceous plants remained similar to the untreated control. Compared to the control and chop treatments, herbicide treatments appeared to be moving the understory closer toward the reference condition, with shrub cover levelling off and graminoid cover continuing to slightly increase after the fire that occurred prior to year three measurements. However, by year 15, woody and shrub cover among all treatments was similar, and the most intense treatments – HM, HB, and CHB – had significantly lower bunchgrass cover. Therefore, treatments that had appeared to overcome the restoration threshold and were moving toward the reference early on actually regressed by year 15 and moved further away from the reference compared to the control and less intense treatments – CF, CM, and CB.

There are two likely explanations for this regression: a fire return interval that was too long and negative impacts on fire behavior as a result of site preparation. The first burn occurred two years after site preparation, but the next two burns occurred at 5-7-year intervals, depending upon the block. The presettlement fire return interval for these sites would have been 1-3 years (Frost, 2001), which indicates that these sites were likely not burned frequently enough between years three and 15. Furthermore, flatwoods have a high shrub component, so the need for frequent fire, especially during restoration, may be emphasized on these sites because even a slight reduction in frequency could promote shrub encroachment (Glitzenstein et al., 2003). Similarly, the direct effects of site preparation on ground layer vegetation in the short-term may have important implications for fire behavior that result in indirect, long-term effects. An analysis of fire behavior during the prescribed fire in 2006 indicated that bedding and mounding interfered with fire behavior (Walker et al., 2009). Bedding rows in particular interrupted the spread of fire because of their minimal vegetation cover and the wet troughs that are adjacent to the rows. Mounding also produced patches of minimal vegetation with adjacent wet pits, but their discontinuous spatial arrangement allowed for more fire continuity. The long-term impact of these alterations to fire behavior may have been increased abundance of shrubs.

In either case, early shrub reductions as a result of site preparation were ultimately lost, suggesting that site preparation may have initially overcome the restoration threshold, but infrequent fire or interrupted fire behavior failed to maintain the early shrub reductions. As a result, the sites appeared to be reverting away from an understory dominated by bunchgrasses to one dominated by shrubs. This is further supported by the fact that year 15 bunchgrass and wiregrass cover on all eight study treatments were much lower than the reference natural vegetation community. In a typical longleaf pine savanna on Leon sand under a frequent fire regime, wiregrass cover is around 50-75% with minimal additional cover from other bunchgrass species (Frost, 2001). In our study, mean bunchgrass and wiregrass cover on all treatments in year 15 were 15.0% and 7.1%, respectively. Even on the chop-only treatment, which had the greatest bunchgrass and wiregrass abundance, mean bunchgrass cover was less than 20% and wiregrass cover was less than 10%. Similarly, mean species richness across all treatments in our study was 16.1 species per 1 m^2, which is much lower than wet longleaf pine savannas that have not undergone site preparation, where richness exceeds 30 species per 1 m^2 (Walker and Peet, 1983). It is unclear why bunchgrass and wiregrass cover and species richness in our study were so low relative to comparable natural vegetation communities, but shrub encroachment and fire behavior are likely explanations. In either case, the importance of frequent fire in restoring and maintaining longleaf pine-grassland ecosystems cannot be overstated.

Conclusion

Understory responses to site preparation in hydric longleaf pine plantations varied significantly among treatments and through time. In the first three years after treatment, herbicide reduced shrubs and other woody competition, particularly when crossed with mounding or bedding, without reducing cover of bunchgrasses. However, by year 15 differences in shrub cover among the eight study treatments were no longer significant, while differences in graminoid and bunchgrass cover were. In the long term, herbicide and soil manipulation treatments had negative impacts on bunchgrasses and especially wiregrass, which is concerning because of the critical role these grasses play in carrying surface fire through the longleaf pine ecosystem. The highest long-term plant species diversity and cover of bunchgrasses and wiregrass were observed on the chop-only treatment, perhaps due to improved surface fire, high understory light transmittance, or a slight, early shrub reduction.

While site preparation appeared to be moving the understory toward the natural vegetation community early on, by year 15 the most intense treatments – those that combined herbicide and mechanical soil manipulation – were furthest from the natural vegetation community due to deleterious impacts on bunchgrasses and wiregrass. It is not clear why this regression occurred, but we hypothesize that it may be due to a fire return interval that was not frequent enough to maintain early reductions in shrub cover or due to treatment effects on fire continuity. Testing similar site preparation treatments as those included in this study under a more frequent fire interval could provide insight into whether fire frequency or behavior may have caused the long-term regression from the natural vegetation community. Additionally, more frequent sampling during the intervening 12-year time period would have provided a better understanding of the trajectory of the understory community. Because fire plays such an important role in the function and maintenance of longleaf pine communities, future research could directly quantify the effects that site preparation, particularly soil manipulation treatments, have on fire behavior.

Chapter 4: Conclusions and Management Implications

Site preparation significantly improved long-term establishment and growth of longleaf pine on hydric sites. Specifically, herbicide resulted in greater growth, higher survival, and earlier grass stage emergence compared to chopping. Similarly, soil manipulation treatments resulted in improved stand establishment outcomes relative to flat-planting (no treatment), with bedding tending to slightly outperform mounding. Differences in survival and growth between treated and untreated areas increased through time, which demonstrates that site preparation had lasting, long-term impacts on longleaf pine stand development. Stem analysis indicated that these long-term stand establishment outcomes are likely the result of increased height growth after emergence from the grass stage, rather than earlier grass stage emergence.

Herbicide and soil manipulation treatments also had long-term impacts on the understory plant community. In the first three years after treatment, herbicide reduced shrubs and other woody competition, particularly when crossed with mounding or bedding, without reducing cover of herbaceous plants or graminoids. Thus, early results suggested that treatments had potential for controlling woody plants to favor herbaceous vegetation. However, by year 15 differences in shrub cover among the eight study treatments were no longer significant, while differences in graminoid and bunchgrass cover were. In the long term, herbicide and soil manipulation treatments had negative impacts on bunchgrasses, especially wiregrass. The greatest long-term plant species diversity and cover of bunchgrasses and wiregrass were observed on the chop-only treatment, perhaps due to improved surface fire, high understory light transmittance, or a slight, early shrub reduction.

Restoration may encompass a range of objectives, but whatever the specific goals may be, a balance must be struck between maximizing site preparation that promotes longleaf pine establishment and minimizing detrimental impacts of those treatments on understory ground flora (Johnson and Gjerstad, 2006). If improved long-term growth or survival of planted seedlings is an important aspect of a restoration effort, then our findings indicate that herbicide or soil manipulation may be necessary on hydric sites. Combining herbicide and soil manipulation maximized stand size and density, but in the context of ecosystem restoration, this intense approach may not be worth the ecological cost. While site preparation appeared to be moving the understory toward the natural vegetation community early on, by year 15 the most intense treatments – those that combined herbicide and mechanical soil manipulation – were furthest from the natural vegetation community because of bunchgrass and wiregrass reductions. The failure to maintain early improvements obtained from herbicide and/or soil manipulation treatments may be due to a fire interval that was too infrequent or interruption of fire continuity as a result of soil manipulation treatments.

On the other hand, if restoration seeks only to establish an open longleaf pine savanna, our findings indicate that a planting density upwards of 1,000 trees per ha preceded only by chopping may result in sufficient long-term establishment. This approach may result in high mortality, as chopping provided little benefit for longleaf pine establishment in our study. However, it may result in the most desirable long-term understory restoration outcomes, as the chop-only treatment did in our study. While the roughly five-year fire return interval proved sufficient for longleaf pine restoration, perhaps more frequent fire would have further improved understory restoration. An evaluation of the treatments' impacts on fire and fuels ecology is beyond the scope of this study, but our findings underscore the importance of incorporating frequent fire into longleaf pine restoration.

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APPENDIX

Appendix 1. All species recorded in 1 m² sampling quadrats. ID period indicates the measurement period(s) (years 1-3, year 15, or both) during which each species was identified.

Family	Genus	Species	Woody/ Herb	Functional Groups	Sub Groups	ID Period
Anacardiaceae	Rhus	copallinum	woody	shrub	tall shrub	both
Anacardiaceae	Toxicodendron	radicans	woody	vine		year 15
Apiaceae	Centella	asiatica	herb	forb	perennial	year 15
Aquifoliaceae	Ilex	coriacea	woody	shrub	tall shrub	both
Aquifoliaceae	Ilex	glabra	woody	shrub	tall shrub	both
Aquifoliaceae	Ilex	myrtifolia	woody	shrub	tall shrub	both
Aquifoliaceae	Ilex	opaca	woody	tree		both
Asclepiadaceae	Asclepias	pedicellata	herb	forb	perennial	year 15
Asteraceae	Acer	rubrum	woody	tree		both
Asteraceae	Ambrosia	artemisiifolia	herb	forb	annual	years 1-3
Asteraceae	Aster	spp.	herb	forb	perennial	years 1-3
Asteraceae	Baccharis	halimifolia	woody	shrub	tall shrub	both
Asteraceae	Carphephorus	spp.	herb	forb	perennial	years 1-3
Asteraceae	Carphephorus	tomentosus	herb	forb	perennial	both
Asteraceae	Coreopsis	falcata	herb	forb	perennial	year 15
Asteraceae	Coreopsis	linifolia	herb	forb	perennial	year 15
Asteraceae	Erechtites	hieraciifolius	herb	forb	annual	both
Asteraceae	Erigeron	spp.	herb	forb	annual	years 1-3
Asteraceae	Eupatorium	album	herb	forb	perennial	years 1-3
Asteraceae	Eupatorium	capilliofolium	herb	forb	perennial	years 1-3
Asteraceae	Eupatorium	leucolepis	herb	forb	perennial	both
Asteraceae	Eupatorium	mohrii	herb	forb	perennial	both
Asteraceae	Eupatorium	pilosum	herb	forb	perennial	both
Asteraceae	Eupatorium	rotundifolium	herb	forb	perennial	both
Asteraceae	Eupatorium	spp.	herb	forb	perennial	years 1-3
Asteraceae	Eurybia	paludosa	herb	forb	perennial	both
Asteraceae	Euthamia	caroliniana	herb	forb	perennial	both
Asteraceae	Euthamia	galetorum	herb	forb	perennial	years 1-3
Asteraceae	Hieracium	gronovii	herb	forb	perennial	year 15
Asteraceae	Ionactis	linariifolia	herb	forb	perennial	both
Asteraceae	Liatris	pilosa	herb	forb	perennial	both
Asteraceae	Liatris	spicata var. resinosa	herb	forb	perennial	year 15
Asteraceae	Marshallia	graminifolia	herb	forb	perennial	year 15
Asteraceae	Pityopsis	graminifolia	herb	forb	perennial	both
Asteraceae	Pterocaulon	pycnostachyum	herb	forb	perennial	year 15
Asteraceae	Sericocarpus	tortifolius	herb	forb	perennial	year 15
Asteraceae	Solidago	fistulosa	herb	forb	perennial	both
Asteraceae	Solidago	spp.	herb	forb	perennial	years 1-3
Asteraceae	Solidago	verna	herb	forb	perennial	years 1-3
Asteraceae	Solidago	nemoralis	herb	forb	perennial	year 15
Asteraceae	Solidago	odora	herb	forb	perennial	both

Family	Genus	Species	Woody/ Herb	Functional Groups	Sub Groups	ID Period
Asteraceae	Solidago	pulchra	herb	forb	perennial	both
Asteraceae	Symphyotrichum	dumosum	herb	forb	perennial	year 15
Asteraceae	Symphyotrichum	walteri	herb	forb	perennial	both
Asteraceae	Taraxacum	spp.	herb	forb	perennial	years 1-3
Asteraceae	Trilisa	odoratissima	herb	forb	perennial	both
Asteraceae	Trilisa	paniculata	herb	forb	perennial	year 15
Blechnaceae	Woodwardia	areolata	herb	fern		both
Blechnaceae	Woodwardia	virginica	herb	fern		both
Campanulaceae	Lobelia	nuttallii	herb	forb	perennial	both
Caprifoliaceae	Viburnum	nudum	woody	shrub	tall shrub	year 15
Cistaceae	Lechea	pulchella var. ramosissima	herb	forb	perennial	year 15
Clethraceae	Clethra	alnifolia	woody	shrub	tall shrub	both
Clusiaceae	Hypericum	cistifolium	woody	shrub	short shrub	year 15
Clusiaceae	Hypericum	crux-andreae	woody	shrub	short shrub	both
Clusiaceae	Hypericum	densiflorum	woody	shrub	short shrub	years 1-3
Clusiaceae	Hypericum	hypericoides	woody	shrub	short shrub	both
Clusiaceae	Hypericum	reductum	woody	shrub	short shrub	years 1-3
Clusiaceae	Hypericum	spp.	woody	shrub	short shrub	years 1-3
Clusiaceae	Hypericum	tenuifolium	woody	shrub	short shrub	year 15
Cornaceae	Nyssa	biflora	woody	tree		year 15
Cuscutaceae	Cuscuta	compacta	herb	forb	perennial	year 15
Cuscutaceae	Cuscuta	spp.	herb	forb	perennial	years 1-3
Cyperaceae	Carex	striata	herb	graminoid	sedge/rush	both
Cyperaceae	Cyperus	spp.	herb	graminoid	sedge/rush	years 1-3
Cyperaceae	Fimbristylis	puberula	herb	graminoid	sedge/rush	both
Cyperaceae	Rhynchospora	breviseta	herb	graminoid	sedge/rush	years 1-3
Cyperaceae	Rhynchospora	cephalantha	herb	graminoid	sedge/rush	year 15
Cyperaceae	Rhynchospora	chalarocephala	herb	graminoid	sedge/rush	both
Cyperaceae	Rhynchospora	chapmanii	herb	graminoid	sedge/rush	year 15
Cyperaceae	Rhynchospora	ciliaris	herb	graminoid	sedge/rush	both
Cyperaceae	Rhynchospora	fascicularis	herb	graminoid	sedge/rush	both
Cyperaceae	Rhynchospora	galeana	herb	graminoid	sedge/rush	year 15
Cyperaceae	Rhynchospora	inexpansa	herb	graminoid	sedge/rush	year 15
Cyperaceae	Rhynchospora	pallida	herb	graminoid	sedge/rush	both
Cyperaceae	Rhynchospora	plumosa	herb	graminoid	sedge/rush	both
Cyperaceae	Rhynchospora	rariflora	herb	graminoid	sedge/rush	year 15
Cyperaceae	Rhynchospora	spp.	herb	graminoid	sedge/rush	years 1-3
Cyperaceae	Scleria	ciliata var. glabra	herb	graminoid	sedge/rush	both
Cyperaceae	Scleria	minor	herb	graminoid	sedge/rush	both
Cyperaceae	Scleria	muehlenbergii	herb	graminoid	sedge/rush	both
Cyperaceae	Scleria	pauciflora var. caroliniana	herb	graminoid	sedge/rush	year 15
Cyrillaceae	Cyrilla	racemiflora	woody	shrub	tall shrub	both
Dennstaediaceae	Pteridium	aquilinum var. pseudocaudatum	herb	fern		both
Diaspensiaceae	Pyxidanthera	barbulata	herb	shrub	short shrub	year 15
Droseraceae	Dionaea	muscipula	herb	forb	perennial	both
Droseraceae	Drosera	brevifolia	herb	forb	perennial	years 1-3

Family	Genus	Species	Woody/ Herb	Functional Groups	Sub Groups	ID Period
Droseraceae	Drosera	capillaris	herb	forb	perennial	both
Droseraceae	Drosera	spp.	herb	forb	perennial	years 1-3
Droseraceae	Drosera	intermedia	herb	forb	perennial	year 15
Ebenaceae	Diospyros	virginiana	woody	tree		both
Ericaceae	Chamaedaphne	calyculata	woody	shrub	short shrub	both
Ericaceae	Eubotrys	racemosus	woody	shrub	tall shrub	year 15
Ericaceae	Gaylussacia	dumosa	woody	shrub	short shrub	both
Ericaceae	Gaylussacia	frondosa	woody	shrub	short shrub	both
Ericaceae	Kalmia	carolina	woody	shrub	tall shrub	both
Ericaceae	Lyonia	ligustrina	woody	shrub	short shrub	both
Ericaceae	Lyonia	lucida	woody	shrub	tall shrub	both
Ericaceae	Lyonia	mariana	woody	shrub	short shrub	both
Ericaceae	Rhododendron	atlanticum	woody	shrub	short shrub	both
Ericaceae	Vaccinium	corymbosum	woody	shrub	tall shrub	years 1-3
Ericaceae	Vaccinium	crassifolium	woody	shrub	short shrub	both
Ericaceae	Vaccinium	formosum	woody	shrub	tall shrub	year 15
Ericaceae	Vaccinium	fuscatum	woody	shrub	tall shrub	both
Ericaceae	Vaccinium	stamineum	woody	shrub	short shrub	year 15
Ericaceae	Vaccinium	tenellum	woody	shrub	short shrub	both
Ericaceae	Zenobia	pulverulenta	woody	shrub	tall shrub	both
Eriocaulaceae	Eriocaulon	compressum	herb	forb	perennial	years 1-3
Eriocaulaceae	Eriocaulon	decangulare	herb	forb	perennial	year 15
Eriocaulaceae	Eriocaulon	spp.	herb	forb	perennial	years 1-3
Eriocaulaceae	Lachnocaulon	anceps	herb	forb	perennial	both
Eriocaulaceae	Lachnocaulon	spp.	herb	forb	perennial	years 1-3
Fabaceae	Lespedeza	cuneata	herb	forb	perennial	years 1-3
Fagaceae	Quercus	falcata	woody	tree		year 15
Fagaceae	Quercus	nigra	woody	tree		year 15
Fagaceae	Quercus	spp.	woody	tree		years 1-3
Fagaceae	Quercus	virginiana	woody	tree		year 15
Gentianaceae	Bartonia	virginica	herb	forb	annual	both
Gentianaceae	Gentiana	autumnalis	herb	forb	perennial	year 15
Gentianaceae	Sabatia	difformis/ quadrangula	herb	forb	perennial	year 15
Gentianaceae	Sabatia	spp.	herb	forb	perennial	years 1-3
Grossulariaceae	Itea	virginica	woody	shrub	tall shrub	year 15
Haemodoraceae	Lachnanthes	caroliniana	herb	forb	perennial	both
Hamamelidaceae	Liquidambar	styraciflua	woody	tree		both
Iridaceae	Iris	spp.	herb	forb	perennial	years 1-3
Iridaceae	Iris	verna	herb	forb	perennial	year 15
Juncaceae	Juncus	biflorus	herb	graminoid	sedge/rush	year 15
Juncaceae	Juncus	pelocarpus	herb	graminoid	sedge/rush	year 15
Juncaceae	Juncus	spp.	herb	graminoid	sedge/rush	years 1-3
Juncaceae	Juncus	scirpoides	herb	graminoid	sedge/rush	year 15
Lauraceae	Persea	borbonia	woody	shrub	tall shrub	years 1-3
Lauraceae	Persea	palustris	woody	shrub	tall shrub	year 15
Lentibulariaceae	Pinguicula	pumila	herb	forb	perennial	year 15
Lentibulariaceae	Utricularia	juncea	herb	forb	perennial	years 1-3

Family	Genus	Species	Woody/ Herb	Functional Groups	Sub Groups	ID Period
Lentibulariaceae	Utricularia	subulata	herb	forb	perennial	year 15
Liliaceae	Aletris	aurea	herb	forb	perennial	year 15
Liliaceae	Aletris	farinosa	herb	forb	perennial	years 1-3
Liliaceae	Amianthium	muscotoxicum	herb	forb	perennial	years 1-3
Liliaceae	Hypoxis	wrightii	herb	forb	perennial	year 15
Liliaceae	Lilium	catesbaei	herb	forb	perennial	year 15
Liliaceae	Pleea	tenuifolia	herb	forb	perennial	both
Liliaceae	Triantha	racemosa	herb	forb	perennial	year 15
Liliaceae	Zigadenus	densum	herb	forb	perennial	both
Liliaceae	Zigadenus	glaberrimus	herb	forb	perennial	year 15
Loganiaceae	Gelsemium	sempervirens	woody	vine		both
Lycopodiaceae	Lycopodiella	alopecuroides	herb	fern		year 15
Magnoliaceae	Magnolia	virginiana	woody	tree		both
Melastomataceae	Rhexia	alifanus	herb	forb	perennial	both
Melastomataceae	Rhexia	lutea	herb	forb	perennial	years 1-3
Melastomataceae	Rhexia	mariana	herb	forb	perennial	years 1-3
Melastomataceae	Rhexia	nashii	herb	forb	perennial	both
Melastomataceae	Rhexia	petiolata	herb	forb	perennial	both
Melastomataceae	Rhexia	spp.	herb	forb	perennial	years 1-3
Myricaceae	Morella	caroliniensis	woody	shrub	tall shrub	both
Myricaceae	Morella	cerifera	woody	shrub	tall shrub	both
Onagraceae	Ludwigia	alternifolia	herb	forb	perennial	years 1-3
Onagraceae	Ludwigia	maritima	herb	forb	perennial	both
Orchidaceae	Calopogon	pallidus	herb	forb	perennial	year 15
Orchidaceae	Cleistes	spp.	herb	forb	perennial	years 1-3
Orchidaceae	Cleistesiopsis/ Pogonia	C.divaricatas/ C. oricamporum/ P. ophioglossoides	herb	forb	perennial	year 15
Orchidaceae	Platanthera	blephariglottis	herb	forb	perennial	year 15
Orchidaceae	Platanthera	spp.	herb	forb	perennial	years 1-3
Orchidaceae	Spiranthes	laciniata	herb	forb	perennial	year 15
Orchidaceae	Spiranthes	spp.	herb	forb	perennial	years 1-3
Osmundaceae	Osmundastrum	cinnamomeum	herb	fern		both
Pinaceae	Pinus	elliottii	woody	tree		both
Pinaceae	Pinus	palustris	woody	tree		both
Pinaceae	Pinus	serotina	woody	tree		both
Pinaceae	Pinus	spp.	woody	tree		years 1-3
Pinaceae	Pinus	taeda	woody	tree		both
Poaceae	Amphicarpum	amphicarpon	herb	graminoid	other graminoid	both
Poaceae	Andropogon	gyrans	herb	graminoid	bunch	year 15
Poaceae	Andropogon	spp.	herb	graminoid	bunch	years 1-3
Poaceae	Andropogon	virginicus var. 1 "smooth variant"	herb	graminoid	bunch	year 15
Poaceae	Andropogon	capillipes	herb	graminoid	bunch	both
Poaceae	Andropogon	glaucopsis	herb	graminoid	bunch	year 15
Poaceae	Andropogon	glomeratus var. glomeratus	herb	graminoid	bunch	year 15
Poaceae	Andropogon	virginicus	herb	graminoid	bunch	both
Poaceae	Aristida	palustris	herb	graminoid	bunch	year 15

Family	Genus	Species	Woody/ Herb	Functional Groups	Sub Groups	ID Period
Poaceae	Aristida	stricta	herb	graminoid	bunch	both
Poaceae	Aristida	virgata	herb	graminoid	bunch	year 15
Poaceae	Arundinaria	tecta	herb	shrub	tall shrub	both
Poaceae	Coleataenia	rigidula	herb	graminoid	other graminoid	year 15
Poaceae	Ctenium	aromaticum	herb	graminoid	bunch	year 15
Poaceae	Dichanthelium	chamaelonche	herb	graminoid	other graminoid	year 15
Poaceae	Dichanthelium	consanguineum	herb	graminoid	other graminoid	year 15
Poaceae	Dichanthelium	dichotomum	herb	graminoid	other graminoid	year 15
Poaceae	Dichanthelium	ensifolium	herb	graminoid	other graminoid	both
Poaceae	Dichanthelium	erectifolium	herb	graminoid	other graminoid	year 15
Poaceae	Dichanthelium	latifolium	herb	graminoid	other graminoid	years 1-3
Poaceae	Dichanthelium	mattamuskeetense	herb	graminoid	other graminoid	year 15
Poaceae	Dichanthelium	scabriusculum	herb	graminoid	other graminoid	year 15
Poaceae	Dichanthelium	scoparium	herb	graminoid	other graminoid	year 15
Poaceae	Dichanthelium	spp.	herb	graminoid	other graminoid	years 1-3
Poaceae	Dichanthelium	strigosum var. leucoblepharis	herb	graminoid	other graminoid	year 15
Poaceae	Dichanthelium	strigosum var. strigosum	herb	graminoid	other graminoid	year 15
Poaceae	Dichanthelium	webberianum	herb	graminoid	other graminoid	year 15
Poaceae	Dichanthelium	leucothrix	herb	graminoid	other graminoid	year 15
Poaceae	Dichanthelium	portoricense	herb	graminoid	other graminoid	year 15
Poaceae	Dichanthelium	tenue	herb	graminoid	other graminoid	year 15
Poaceae	Eragrostis	refracta	herb	graminoid	bunch	year 15
Poaceae	Eragrostis	spp.	herb	graminoid	bunch	year 15
Poaceae	Muhlenbergia	expansa	herb	graminoid	bunch	years 1-3
Poaceae	Panicum	spp.	herb	graminoid	other graminoid	years 1-3
Poaceae	Panicum	verrucosum	herb	graminoid	other graminoid	both
Poaceae	Paspalum	setaceum	herb	graminoid	other graminoid	year 15
Poaceae	Poa	spp.	herb	graminoid	bunch	years 1-3
Poaceae	Schizachyrium	scoparium	herb	graminoid	bunch	both
Poaceae	Sporobolus	brevipilis	herb	graminoid	bunch	year 15
Poaceae	Sporobolus	pinetorum	herb	graminoid	bunch	both
Polygalaceae	Polygala	brevifolia	herb	forb	annual	year 15
Polygalaceae	Polygala	lutea	herb	forb		both
Polygalaceae	Polygala	nana	herb	forb	annual	years 1-3
Polygalaceae	Polygala	ramosa	herb	forb	annual	year 15
Polygalaceae	Polygala	cruciata	herb	forb	annual	both
Rosaceae	Aronia	arbutifolia	woody	shrub	tall shrub	both
Rosaceae	Crataegus	spp.	woody	shrub	tall shrub	year 15

Family	Genus	Species	Woody/ Herb	Functional Groups	Sub Groups	ID Period
Rosaceae	Rubus	cuneifolius	woody	shrub	tall shrub	year 15
Rosaceae	Rubus	pensilvanicus	woody	shrub	tall shrub	year 15
Rosaceae	Rubus	spp.	woody	shrub	tall shrub	years 1-3
Rubiaceae	Mitchella	repens	herb	forb	perennial	both
Sarraceniaceae	Sarracenia	flava	herb	forb	perennial	both
Sarraceniaceae	Sarracenia	purpurea	herb	forb	perennial	both
Scrophulariaceae	Agalinis	purpurea/ virgata	herb	forb	annual	year 15
Scrophulariaceae	Gratiola	pilosa	herb	forb	perennial	year 15
Scrophulariaceae	Seymeria	cassioides	herb	forb	annual	years 1-3
Smilacaceae	Smilax	bona-nox	woody	vine		years 1-3
Smilacaceae	Smilax	glauca	woody	vine		both
Smilacaceae	Smilax	laurifolia	woody	vine		both
Smilacaceae	Smilax	rotundifolia	woody	vine		years 1-3
Smilacaceae	Smilax	spp.	woody	vine		years 1-3
Symplocaceae	Symplocos	tinctoria	woody	shrub	tall shrub	both
Theaceae	Gordonia	lasianthus	woody	tree		both
Violaceae	Viola	primulifolia	herb	forb	perennial	year 15
Vitaceae	Muscadinia	rotundifolia	woody	vine		year 15
Xyridaceae	Xyris	caroliniana	herb	forb	perennial	both
Xyridaceae	Xyris	platylepis	herb	forb	perennial	both
Xyridaceae	Xyris	ambigua	herb	forb	perennial	both
Xyridaceae	Xyris	baldwiniana	herb	forb	perennial	both
Xyridaceae	Xyris	brevifolia	herb	forb	perennial	both