

**Physiology and Growth of a 6-year-old Loblolly Pine (*Pinus taeda* L.) Plantation in
Response to Rainfall Exclusion and Fertilization Treatments**

by

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Abstract

The increase in atmospheric carbon dioxide (CO₂) is expected to change climate in the southeastern United States. A hotter, drier climate is anticipated over the next century and could have detrimental effects on the productivity of southern forests. Loblolly pine (*Pinus taeda* L.) is the most widespread planted southern pine in the region and knowledge of how this species will respond to future climate is critical. To gain insight on how the growth of loblolly pine may be affected by changes in ambient precipitation, growth and physiological responses of a 6-year-old loblolly pine plantation to rainfall manipulation across a nutritional gradient were studied over a one year period. The experiment was a 2 x 2 factorial design with two levels of rainfall manipulation (ambient and 30% reduction) and two levels of fertilization (none and operational). Fertilization had the greatest influence on leaf area index (LAI) and intercepted photosynthetically active radiation (IPAR). During a drought in 2012, fertilization increased LAI by 17%, while the rain exclusion treatment decreased light saturated net photosynthesis (P_{net}) and stomatal conductance (g_s) by 12% and 21% respectively and increased stomatal limitations of P_{net} . Average soil moisture and predawn water potential (Ψ_L) decreased in 2012 in the rain exclusion treatment. In 2013, a year with high ambient precipitation, fertilization increased LAI by 49%, while the rain exclusion treatment caused no reductions in leaf physiology. No interactions between rainfall manipulation and fertilization treatments were observed for LAI, IPAR, or leaf physiology. The primary effect of the fertilization treatment was on leaf area production and the primary effect of the rain exclusion treatment was on leaf-level physiology.

Reductions in P_{net} and g_s in the rain exclusion treatment indicate that under future hotter, drier climates reduced carbon gain may occur. No interactive effects were observed between the rain exclusion and fertilization on LAI, IPAR and physiological processes, but the greatest gains in LAI in response to fertilization were observed in 2013 when drought was alleviated.

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List of Abbreviations

BA	Basal area
BAI	Basal area increment
CAI	Current annual increment
CO ₂	Carbon dioxide
DBH	Diameter at breast height
Fv/Fm	Efficiency of the photosystem II
GE	Growth efficiency
g _s	Stomatal conductance
ha	Hectare
IPAR	Intercepted photosynthetically active radiation
K	Potassium
LAI	Leaf area index
L _g	Stomatal limitation
N	Nitrogen
P	Phosphorus
PDSI	Palmer Drought Severity Index
P _{net}	Light saturated net photosynthesis
SAS	Statistical Analysis System
SLA	Specific leaf area
Vol	Standing stem volume
Ψ _L	Leaf water potential

1.0 Project Background and Objectives

The southeastern United States is comprised of Atlantic and Gulf coastal states from Texas to Virginia. This region is considered the wood basket of the world, comprising 60% of U.S. timber production and 16% of world production (Wear and Greis, 2002). As of 2010, there were 87 million hectares of forested timber land in the southeastern United States (Weir and Greis, 2011). Of that acreage, 16 million ha or 17% of the forested land is planted in southern pine and that percentage is expected to increase in the next 50 years (Wear and Greis, 2011). This timber production helps drive the wood products sector that accounts for 5.5% of jobs and 7.5% of total industry output in the region (Weir and Greis, 2002). Loblolly pine (*Pinus taeda* L.) is the most expansively grown southern pine in the region because of its adaptability to diverse sites and its favorable response to silvicultural practices (Schultz, 1997). Intensive forest management practices have improved growth and yield in loblolly pine, with productivity increasing three fold through competition control, fertilization, and superior genotypes in the last half century (Jokela et al., 2010; Aspinwall et al., 2011). Potential changes in climate could affect the most important commercial timber species of the region.

Increases in atmospheric carbon dioxide (CO₂) have the potential to influence the climate of every region of the world. As atmospheric CO₂ nears 400 ppm (Monastersky, 2013), global temperatures are expected to continue to rise throughout the next century (Christensen et al., 2007). This rise in overall temperature is expected to be accompanied by more intense and longer duration of heat-waves (Christensen et al., 2007). In the southeastern United States, average annual temperature is expected to rise in every season, with the greatest increases in spring and summer months (Karl et al., 2009). Winter seasons are expected to receive more precipitation while the summer is expected to experience more severe droughts, with extreme predictions of a

30% reduction in annual rainfall (Christensen et al., 2007; Karl et al., 2009). These shifts in climate could potentially have detrimental effects on forest productivity and health (Field et al., 2007; Allen et al., 2010).

Carbon management through existing forests is one potential solution for mitigating atmospheric CO₂ levels. Forests can potentially mitigate rising CO₂ levels and reduce the effect of climate change by acting as large carbon sinks. According to Ryan et al., (2010) there are many solutions in forestry that can aid in the management of carbon. Strategies used by forest managers to increase carbon accumulation include: afforestation, biomass energy, carbon storage through forest products, forest management to decrease carbon loss, reduction of fire risk and the focus of this study, increased growth through improved silvicultural practices. Forests have the ability to sequester large amounts of carbon in woody biomass. The southeastern United States holds 12 Pg of carbon, which is nearly 40% of the carbon in the conterminous United States (Tuner et al., 1995). The southeastern United States has been estimated to sequester 210 Tg of carbon annually in pine plantations (Johnsen et al., 2001). This amount of carbon sequestered in pine plantations has the potential to be further increased through intensive management practices, such as fertilization (Albaugh et al., 2012) and superior genetics (Aspinwall et al., 2011).

With the predicted climatic shift in the southeastern United States, forest managers require new information on how pine plantations will respond under future climatic conditions and the consequences for implementing specific silvicultural practices. The possibility of increased temperatures and more frequent droughts could have a negative effect on growth and ultimately the region's ability to sequester large amounts of carbon in woody biomass to help

offset carbon emissions. To investigate the potential mitigation of CO₂ through improved silvicultural practices in southern pine forests, PINEMAP was created in 2011.

PINEMAP: Pine Integrated Network: Education, Mitigation, Adaptation Project is funded by an Agricultural and Food Research Initiative Competitive Grant under the National Institute of Food and Agriculture in the United States Department of Agriculture. PINEMAP focuses on the 8 million hectares of planted pine forest managed by private landowners in the Atlantic and Gulf coastal states from Virginia to Texas, plus Arkansas and Oklahoma. PINEMAP is integrating research, extension, and education to enable forest landowners to manage forest to increase carbon sequestration, increase the efficiency of nitrogen inputs and adapt forest management approaches to increase forest resiliency and sustainability under variable climate. Project goals are to create and disseminate the knowledge that enables landowners to: manage forests to increase carbon sequestration by 15% by 2030 and increase the efficiency of nitrogen and other fertilizer inputs by 10% by 2030. The overall project will develop extension and education programs to relay knowledge gained from research to landowners and managers as quickly as possible.

There are six Aims involved in the project: The first is the Ecophysiology and Silviculture aim that has established a region wide, three-tiered monitoring system based on existing cooperative research trials and will develop standardized methods to quantify carbon, water, nutrient storage and flux baselines and responses to climate and management. The second is the Modeling Aim which will apply a multi-scaled modeling system incorporating data gathered from the monitoring system created by the Ecophysiology and Silviculture Aim. The third is the Genetics Aim which will analyze the genetics of breeding and natural populations to discover alleles in genes controlling important adaptation and mitigation traits. The fourth is the

Economics and Policy Aim that will conduct comprehensive life cycle analyses of regional forest management systems and multi-scale policy and economic analysis to assess adoption of alternative approaches by private landowners. The fifth is the Education Aim which will create educational resources and training programs for teachers and extension agents to convey the value and relevance of southern forests and climate change impacts. Finally, the Extension Aim will develop extension programming that combines regional climate expertise and forest management outreach to deliver resources and management decision support tools to forest landowners, resource managers and policy makers.

Our specific project is incorporated with the Ecophysiology and Silviculture aim, focusing on one tier III site. The three tiered monitoring system consist of Tier I (Legacy sites), Tier II (Active sites) and Tier III (Fertilization and Rainfall Manipulation sites). Tier I sites were selected from an extensive network of growth and yield cooperative plots that encompass the entire region. These plots have repeated tree inventory measurements and provide information on the spatial and temporal variability of productivity across the region. Tier II sites were chosen from existing cooperative field studies and planted pine AmeriFlux installations across the entire region, most of which include replicated silvicultural treatments. New measurements will also be implemented at Tier II sites to assist in quantifying carbon and nitrogen pools. Tier III sites consist of four experiments established near the edge of loblolly pine range, with the experiments consisting of rainfall manipulation and fertilization treatments. The approach taken in this investigation is to replicate an intensively managed plantation combined with future predictions of precipitation and gain insight on how the physiological processes controlling growth, and consequently carbon sequestration, of loblolly pine will be influenced by the combination of future climate and silviculture practices.

The main objectives of this study are to:

1. Examine how a 30% reduction in ambient precipitation will influence leaf level physiology, leaf area development and light interception of *Pinus taeda* L.
2. Determine whether fertilization will have a greater impact on leaf area and intercepted radiation than on leaf level physiology.
3. Determine if increased LAI and growth in fertilized stands will increase susceptibility to drought.

2.0 Physiology and growth of a 6-year-old loblolly pine plantation in response to rainfall exclusion and fertilization treatments

2.1 Introduction

Loblolly pine is the most extensively planted and managed commercial pine species in the southeastern United States because of its adaptability to diverse sites and its favorable response to silvicultural practices (Schultz, 1997). In the last half century the productivity of loblolly pine has increased three fold through the use of competition control and fertilization and deployment of superior genotypes (Jokela et al., 2010; Aspinwall et al., 2011). Fertilization increases productivity by increasing leaf area, light interception and photosynthetic capacity (Fox et al., 2007; Will et al., 2005; Samuelson et al., 2008; Pangle et al., 2009). For example, Samuelson et al., (2008) studied the growth potential of loblolly pine in response to long term resource management, and reported that fertilization increased productivity by 62%. Jokela et al., (2004) summarized the production dynamics of managed loblolly pine from seven long-term experiments across the southeastern United States and found that leaf area index (LAI) was strongly correlated with stem wood biomass increment and that nutrients rather than water availability had the greatest effect on LAI. Productivity has also been shown to be positively and linearly related to the amount of photosynthetically active radiation (PAR) absorbed by the canopy (Cannel et al., 1987; Dalla-Tea and Jokela 1991; Will et al., 2005; Allen et al., 2005), which is directly related to LAI. Fertilization may also increase growth efficiency (GE), the amount of stemwood produced per unit leaf area, by increasing leaf-level photosynthetic rates and decreasing partitioning of biomass belowground (Murthy et al., 1996; Albaugh et al., 1998, 2004; Gough et al., 2004; Samuelson et al., 2004; Fox et al., 2007).

Enhancement of foliage and stem biomass production by fertilization can be limited by drought (Gholz et al., 1990) as a result of decreased foliage production (Hennessey et al., 1992;

Ellsworth 2000; Vose and Allen 1988; Tang et al., 2004) and a downward shift in photosynthetic rate (Seiler and Johnson 1985; Samuelson, 1998). For example, Tang et al., (2004) reported an interactive effect of throughfall rain exclusion and fertilization treatments on LAI in 18-year-old loblolly pine: LAI was increased by fertilization only in ambient rainfall plots. In addition, Tang et al., (2004) found that the throughfall rain exclusion treatment decreased whole canopy net photosynthesis and leaf level stomatal conductance (g_s). Improvements in growth have been observed when water is made non-limiting in forest stands (Stape et al., 2010; Allen et al., 2005; Samuelson 2004). Campoe et al., (2013) found that fertilization increased above ground net primary production (ANPP) of a 9-year-old loblolly pine plantation by 2-fold and that adding irrigation with fertilization resulted in a tripling of ANPP.

Climate models predict a 2-5 °C rise in average annual temperatures across the United States over the next century (Christensen et al., 2007). In the southeastern United States, warming is expected during every season, but the largest temperature increases are projected to occur in summer, accompanied by a reduction in precipitation primarily in the spring and summer (Karl et. al., 2009). The increase in temperature combined with reduced rainfall across the southeastern United States can lead to reduced forest productivity and tree mortality (Field et al., 2007; Allen et al., 2010). For example, Noormets et al., (2010) studied carbon fluxes in a coastal plain loblolly pine forest and found net primary production (NPP) was closely linked to precipitation. Moisture limitations increased the sensitivity of canopy stomatal conductance to high evaporative demands by decreasing tree hydraulic conductivity during drought.

To better understand the potential impacts of reduced precipitation combined with silvicultural practices on loblolly pine productivity, we examined the interactive effects of rainfall exclusion and fertilization treatments on growth, foliage production, intercepted radiation

and leaf-level physiology in a 6-year-old loblolly pine plantation. The influence of fertilization treatments (none and operational) and rainfall exclusion treatments (ambient or a 30% reduction) on LAI, IPAR, leaf-level gas exchange, leaf water potential (Ψ_L), chlorophyll fluorescence and foliar $\delta^{13}\text{C}$ were measured over a one year period. Throughfall exclusion trays were installed to intercept 30% of ambient rainfall. Similar throughfall exclusion trays have been used in studies in native forests of Brazil (Nepstad et al., 2002) and loblolly pine in Louisiana (Tang et al., 2004). We hypothesized that: (1) water availability would have a greater impact on leaf level physiology than on leaf area development and light interception; (2) fertilization would have a greater impact on LAI and IPAR than on leaf level physiology; (3) higher LAI in fertilized stands will increase susceptibility to drought. This project was conducted in conjunction with a large integrated network of research on loblolly pine productivity under changing climate known as PINEMAP (Pine Integrated Network: Education, Adaptation Project (www.pinemap.org)). The overall goals of PINEMAP are to increase carbon sequestration by loblolly pine plantations by 15% by 2030, increase the efficiency of nitrogen and other fertilizer inputs by 10%, and adapt forest management approaches to increase forest resilience and sustainability under variable climate.

2.2 Material and Methods

2.2.1 Site description

The study was conducted in a loblolly pine plantation located in Taliaferro County, Georgia, owned by Plum Creek Timber Company. The study site is located approximately 12 km northeast of Crawfordville, GA at an elevation of 152 m and a latitude $33^{\circ}37'32.61''$ N and longitude $82^{\circ}47'56.54$ W. Average daily maximum and minimum temperature is 22.7°C and 10.1°C with an average annual precipitation of 110.8 cm (<http://www.ncdc.noaa.gov/cdo-web/datasets/ANNUAL/locations/ZIP:30673/detail>, accessed January 2013). The Palmer Drought Severity Index (PDSI) during 2012 and 2013 was collected for Climate Division 3 in the state of Georgia (<http://www1.ncdc.noaa.gov/pub/data/cirs/drd964x.pdsi.txt/> accessed July 2013). The site is comprised of two different soil series, Lloyd and Cecil series. The variation in soil series between and among blocks was minimal. The site comprised mostly of the Lloyd series and only a small portion as the Cecil series. The Lloyd series is a fine, kaolintic, thermic Rhodic, while the Cecil series is fine, kaolinitic, thermic Typic Kanhapludults (Daniel Markewitz, University of Georgia Warnell School of Forestry and Natural Resources, personal communication). These soils are common to the gently sloping to moderately steep uplands of the Piedmont and tend to be well drained with medium to rapid runoff and moderate permeability (www.soilseries.sc.egov.usda.gov/ accessed June 2013). The site was hand planted in 2006 with loblolly pine seedlings from an open-pollinated family at a density of 1544 trees ha^{-1} and an approximate spacing of 3 m x 2 m. The site received herbaceous weed control during planting (Oust extra, 219 ml ha^{-1}).

2.2.2 Experimental Design

The study was designed as a 2 x 2 factorial with four blocks and two levels of precipitation, ambient and 30% reduction, and two levels of fertilization, none and operational. Each plot was comprised of a 0.03 ha measurement plot, 0.10 ha treatment plot and a 6.1 m buffer. To achieve a target 30% reduction in ambient precipitation, rain throughfall exclusion trays were installed to cover 30% of the ground area and transport rainfall off the treatment plot and into the buffer areas. A supporting structure was built to fit between rows in the treatment plot and measured approximately 3 m wide, and the height of the trays varied from 1 m to 3 m across plots. Two rainfall collections trays were secured on top of the supporting structure and the trays were separated by a 30.5 cm opening. Trays were covered with clear Poly Scrim 12 that consist of two layers of U.V. stabilized coextruded polyethylene and high strength cord. Trays were installed in May of 2012. The operational fertilization treatment included 224 kg N ha⁻¹, 28 kg P ha⁻¹, 56 kg K ha⁻¹, and a micronutrient blend. Nitrogen and phosphorus were applied as a mix of urea and diammonium phosphate (DAP), and potassium was applied using potassium chloride (KCl). Fertilizer was evenly broadcast across each plot by hand in March of 2012.

2.2.3 Growth

Stand level inventories were conducted at study initiation (December 2011) and after one year of treatment (December 2012). Individual tree stem volume was calculated using equations from Van Deusen et al., (1981) and individual tree stem weight was calculated using equations from Bullock and Burkhart, (2003). Conversion of green weight to dry weight followed Samuelson et al., (2008). Growth efficiency was calculated as the ratio between current annual increment (CAI) and maximum leaf area index (LAI).

2.2.4 Leaf Area Index and Intercepted Photosynthetically Active Radiation

Leaf area index was measured monthly using optical sensors (LAI- 2000 Plant Canopy Analyzer LI-COR Inc., Li-COR Lincoln, NE, USA). Measurements were made in diffuse sunlight, within an hour of either dusk or dawn, using two sensors. One sensor was placed in the opening near the site and the other was used to take readings below the canopy using the 90° view cap. All measurements were made above the exclusion trays, with heights of trays ranging from 1 m to 3 m, along randomly selected, diagonal transects across the inter row space. Three trees, each from a different row, were randomly selected before each sampling period. Starting at each selected tree, LAI was measured every 0.5 m along each transect, totaling eight points on each transect with the direction of each transect randomly selected. At each of the eight points the sensor was pointed parallel with the row towards plot center.

Intercepted photosynthetically active radiation (IPAR) was measured between 1100 and 1600 h during cloud free or mostly sunny days using a line quantum sensor (Licor Inc., Lincoln NE, USA). Measurements were made monthly from May 2012 to September 2012 and from March 2013 through July 2013 when the zenith angle was between 10° and 30°. The proportion of radiation intercepted by each plot was calculated as the difference between above canopy radiation and below canopy radiation measured simultaneously. Three permanent transects were established between rows in each measurement plot. Along each transect, ten randomly selected permanent sample points were marked. Three non-overlapping measurements were made at each permanent sample point to capture the light environment across the entire inter row space (tree to tree). The three measurements made at each permanent sample point were averaged to generate a total of 30 measurements for each plot. Intercepted radiation was then calculated as:

$$\text{IPAR}=1 - (\text{under canopy PAR} / \text{incident PAR}) \quad (1)$$

Daily IPAR interpolation and conversion to cumulative IPAR intercepted followed Allen et al., (2005).

2.2.5 Soil Moisture

Soil moisture was measured using the Hyrdosense II which uses the dielectric permittivity technique (Campbell Scientific, Logan, UT, USA). Soil moisture was measured to a 12 cm depth during leaf gas exchange measurements. At each measurement tree, soil moisture was measured at two locations: one equidistance between trees and one equidistant between rows, tree side and row were chosen randomly. Soil moisture measurements made between rows in rain exclusion plots were made directly under rain exclusion trays.

2.2.6 Leaf Physiology

Gas Exchange

Light-saturated net photosynthesis (P_{net}) and stomatal conductance (g_s) were measured using a portable photosynthesis system (Licor 6400-40 portable photosynthesis system Licor Inc., Lincoln NE, USA) equipped with a fluorescence chamber. Stomatal limitation (L_g) was calculated as $(C_a - C_i) / C_a$, where C_a is ambient CO_2 and C_i is the intercellular CO_2 concentration. A shoot from the upper third of the canopy of three trees per plot was detached using a pole pruner. Gas exchange measurements were made on two fascicles from each shoot within one minute of detachment (Samuelson et al., 2001). Trees were randomly selected each measurement session. Measurements were made monthly, with the exception of December 2012 and January 2013, beginning July 2012 and concluding July 2013. Starting in July 2012 to May 2013, the first flush of the 2012 was measured. Beginning June 2013, the first flush produced in 2013 was measured. Measurements were made by block over a two day period between 0900

and 1400 hours. The measurement order of blocks and plots within blocks was randomly selected.

Within the leaf chamber, temperature and humidity were allowed to fluctuate with ambient conditions. Photosynthetically active radiation and CO₂ concentration were held constant inside the chamber at 1800 $\mu\text{mol m}^{-2} \text{ s}^{-1}$ and 400 ppm, respectively. After measurements, the total area of the two fascicles from each measurement was calculated following Samuelson et al., (1992). Needles were then dried, at 70° C for at least 72 hours and weighed. Specific leaf area (SLA) was then calculated as the ratio between foliage area and dry mass.

Chlorophyll Fluorescence

Minimum fluorescence (F_0) and maximum fluorescence (F_m) were measured on six dates from June 2012 to July 2013 using a portable fluorometer (Licor 6400-64 fluorescence chamber Licor Inc., Lincoln, NE, USA). Foliage was dark adapted for a minimum of 20 minutes *in situ* to determine the maximum efficiency of photosystem II (F_v/F_m). Dark adapted fluorescence measurements made immediately following gas exchange measurements on foliage adjacent to gas exchange samples. All measurements of fluorescence were made on the first flush produced in 2012, because in April 2013 the first flush of the current year was not yet fully expanded.

Leaf Water Potential

Predawn and midday leaf water potential (Ψ_L) were measured using a pressure chamber (PMS, Instrument Corp., Corvallis, OR, USA). Midday Ψ_L measurements were made during photosynthesis measurements. One fascicle was removed from each shoot detached for leaf gas exchange measurements. Predawn Ψ_L were measured between 0400 and 0600 hours.

2.2.7 Foliar Nutrients and $\delta^{13}\text{C}$ Concentration

Foliage was collected October 2012 for foliar N and $\delta^{13}\text{C}$ analyses from three trees per plot. Each shoot was collected from the upper third of the canopy and the first and second flush of 2012 was measured. The second flush was initiated in late July and was much smaller than the first flush. Total needle area was measured as described previously and foliage was then dried at 70° C for at least 72 hours and weighed. Tissue was then ground to a fine powder. Samples were sent to the Duke Environmental Stable Isotope Laboratory (biology.duke.edu/Jackson/devil) for $\delta^{13}\text{C}$ analysis, which uses the Finnigan MAT Delta Plus XL continuous flow mass spectrometer (Thermo Scientific, Waltham, MA, USA). Foliar nutrients, nitrogen (N), phosphorus (P), and potassium (K) were analyzed at the Soil Testing Laboratory (www.aces.edu/anr/soillab/) at Auburn University by using inductively coupled argon plasma spectrophotometry (ICAP).

2.2.8 Statistical Analyses

Measurements were separated by year for analysis. Data were averaged by month, block, and plot. The main and interactive effects of fertilization, water availability and month of measurement, where appropriate, were tested using repeated measures analysis of variance with block as a random factor and treatments as fixed factors (PROC MIXED, SAS, Cary, NC, USA). Analyses of basic stand measurements for 2011 and 2012, peak LAI and IPAR, cumulative PAR, foliar nutrients and foliar $\delta^{13}\text{C}$ were performed using PROC GLM procedure in SAS (Statistical Institute, Cary, NC, USA). Both main and interaction effects were considered significant at $\alpha=0.05$.

2.3 Results

2.3.1 Climate

During 2012, monthly precipitation ranged from 17.3 mm in October to 125.2 mm in July and totaled 849.4 mm for the year (Figure 2.3.1). Average daily minimum temperatures ranged from 2.8 °C to 23.8 °C and average daily maximum temperatures ranged from 15.1 °C to 34.9 °C (Figure 2.3.1). The Palmer Drought Severity Index (PDSI) indicates there was a significant drought throughout 2012 (Figure 2.3.1). Average leaf temperature during leaf gas exchange measurements ranged from 15.4 °C in November to 35.9 °C in July. The average leaf to air vapor pressure deficit (VPD) during gas exchange measurements ranged from 1.1 kPa in November to 2.9 kPa in July (Figure 2.3.1).

Between January 2013 and July 2013, average daily maximum temperatures ranged from 16.4 °C to 30.8 °C and average daily minimum temperatures ranged from 2.6 °C to 22.2 °C. Monthly precipitation ranged from 55.4 mm in January to 201.9 mm in June. The PDSI indicated that drought began to dissipate at the beginning of 2013 due to high rainfall at the beginning of the year (Figure 2.3.1). Average leaf temperature during leaf gas exchange measurements ranged from 17.4 °C in March to 33.2 °C in June, and average VPD ranging from 0.8 kPa in February to 1.8 kPa in June (Figure 2.3.1).

2.3.2 Growth

No significant interactive or main effects of the rain exclusion or fertilization treatments were observed for pre-treatment DBH, Ht, BA, and Vol measured December 2011 (data not shown). In 2012, no significant interactive effects of rain exclusion and fertilization treatments on growth were detected (Table 2.3.1). Fertilization significantly increased basal area increment

(BAI) and current annual increment (CAI) and a trend ($p=0.100$) for increased basal area (BA) was detected. Rain exclusion decreased BAI, while having no significant effects on any other variables (Table 2.3.1). Growth efficiency (GE) was not affected by fertilization or rain exclusion treatments and averaged $7.1 \text{ m}^3 \text{ LAI}^{-1}$ across all treatments (Table 2.3.2). During the study period a total of five trees died. Four of the five dead trees were removed from the rain exclusion x fertilization treatment in block 1, with two being removed in April 2013 and two removed in August 2013. In July 2013, the top of one tree in the rain exclusion x fertilization treatment in block 2 was blown out.

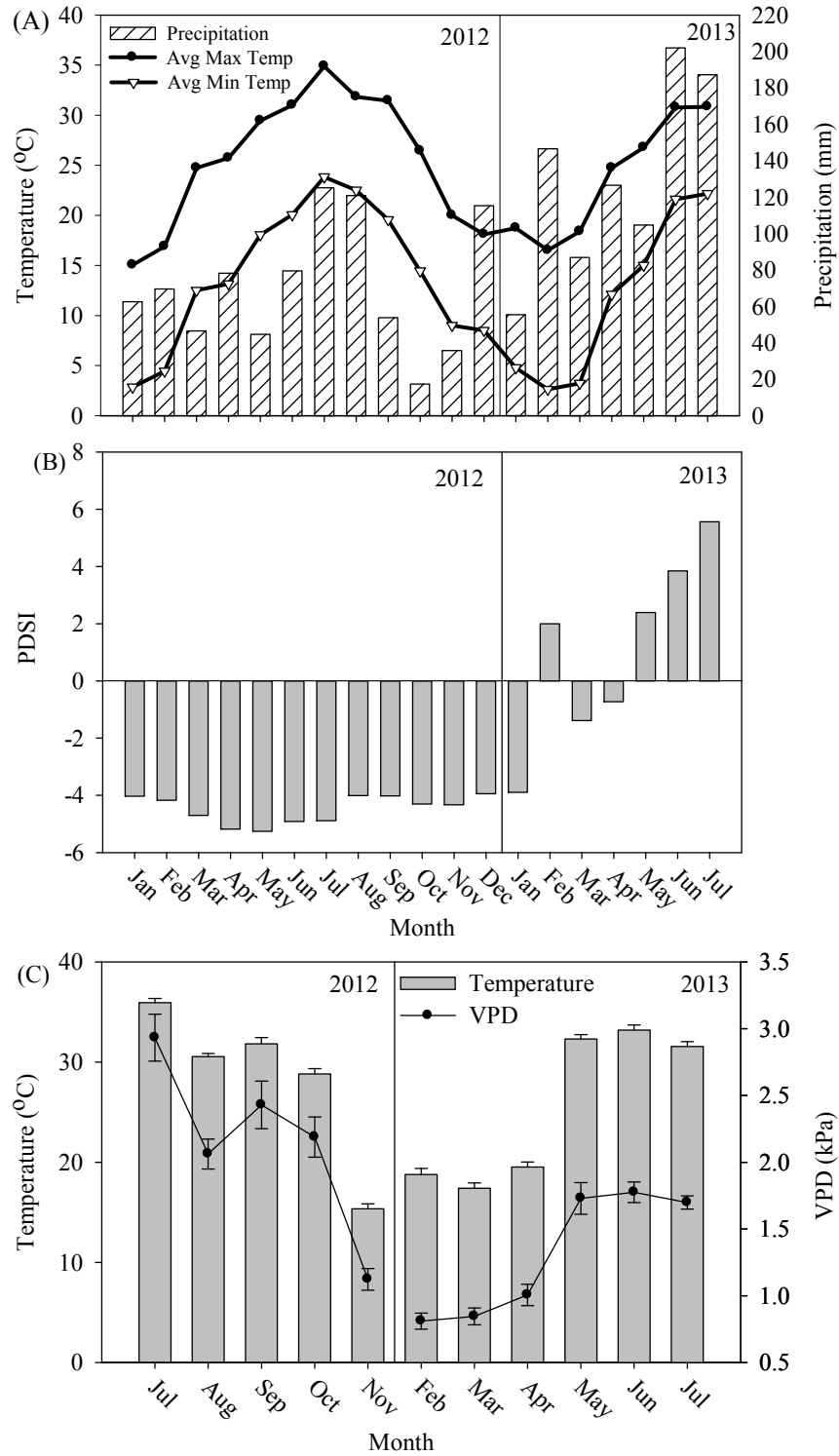


Figure 2.3.1. Total monthly precipitation and average minimum and maximum temperatures (A), the Palmer Drought Severity Index (PDSI) during 2012 and 2013 (B), and monthly average leaf temperature and leaf vapor pressure deficit (VPD) averaged across all treatments during leaf gas exchange measurements (C).

Table 2.3.1. Influence of rain exclusion and fertilization treatments on mean (\pm SE) stand characteristics of 6-year-old loblolly pine plantation. Diameter at breast height (DBH), height (Ht), basal area (BA), standing stem volume (Vol), current annual increment (CAI), and basal area increment (BAI) were measured in December 2012, after one year of treatment.

Treatment	DBH (cm)	Ht (m)	BA (m ² ha ⁻¹)	Vol (m ³ ha ⁻¹)	CAI (m ³ ha ⁻¹ yr ⁻¹)	BAI (m ² ha ⁻¹ yr ⁻¹)
Ambient Rain	10.6 (0.3)	7.2 (0.2)	12.5 (0.7)	40.6 (2.4)	14.3 (0.7)	3.8 (0.2)
Rain Exclusion	10.4 (0.3)	7.2 (0.2)	12.1 (0.6)	41.0 (2.9)	12.7 (0.9)	3.3 (0.2)
No Fertilization	10.4 (0.3)	7.1 (0.2)	11.8 (0.6)	39.2 (2.6)	12.1 (0.8)	3.1 (0.1)
Fertilization	10.7 (0.3)	7.3 (0.2)	12.9 (0.6)	42.4 (2.6)	14.9 (0.6)	4.0 (0.2)
<i>P>F</i>						
Rain Exclusion	0.368	0.668	0.493	0.946	0.067	0.006
Fertilization	0.129	0.433	0.076	0.252	0.014	<0.001
Rain Exclusion x Fertilization	0.776	0.944	0.946	0.309	0.283	0.918

2.3.3 LAI and IPAR

In 2012, no interactions between rain exclusion and fertilization treatments were detected for peak LAI or IPAR (Table 2.3.2). In 2012, peak LAI was reached in September and LAI increased from $1.78 \text{ m}^2 \text{ m}^{-2}$ with no fertilization to $2.13 \text{ m}^2 \text{ m}^{-2}$ with fertilization (Figure 2.3.2). Peak IPAR was significantly increased by fertilization and coincided with peak LAI in September (Table 2.3.2, Figure 2.3.2). Fertilization treatment increased IPAR by 13% compared to the no fertilization treatment. The rain exclusion treatment had no effect on peak LAI and IPAR (Table 2.3.2). Neither rain exclusion or fertilization treatment had a significant effect on cumulative PAR interception (Table 2.3.2).

In 2013, no interactions between rain exclusion and fertilization treatments were detected for peak LAI or IPAR in July (Table 2.3.2). Fertilization increased LAI in July by 49%, from $2.14 \text{ m}^2 \text{ m}^{-2}$ with no fertilization to $3.18 \text{ m}^2 \text{ m}^{-2}$ with fertilization (Figure 2.3.2). In July, peak IPAR was increased 17% by fertilization (Table 2.3.2) from 70% in no fertilization treatment to 82% in the fertilization treatment (Figure 2.3.2). The rain exclusion treatment had no effect on LAI or IPAR in 2013.

Table 2.3.2. Influence of rain exclusion and fertilization treatments on growth efficiency (GE), cumulative intercepted photosynthetically active radiation (Cumulative IPAR) for 2012, peak leaf area index (LAI), and intercepted photosynthetically active radiation (IPAR) for 2012 and 2013, in a 6-year-old loblolly pine plantation. Growth efficiency was calculated for 2012 as the ratio between CAI and peak LAI. Peak LAI and IPAR for 2012 occurred in September and our observed peak LAI and IPAR in 2013 occurred in July. Cumulative IPAR is calculated as interpolated daily IPAR summed for 2012.

Treatment	GE (m ³ LAI ⁻¹)	Cumulative IPAR (MJ m ⁻² yr ⁻¹)	2012 LAI (m ² m ⁻²)	2013 LAI (m ² m ⁻²)	2012 IPAR (%)	2013 IPAR (%)
Ambient Rain	7.4 (0.3)	1901.6 (101.2)	1.9 (0.1)	2.7 (0.2)	74 (0.02)	75 (0.04)
Rain Exclusion	6.8 (0.4)	1916.8 (76.9)	1.9 (0.2)	2.6 (0.2)	78 (0.02)	78 (0.04)
No Fertilization	6.8 (0.3)	1833.1 (75.8)	1.8 (0.1)	2.1 (0.1)	72 (0.02)	70 (0.03)
Fertilization	8.0 (0.4)	1985.4 (93.7)	2.1 (0.2)	3.2 (0.2)	80 (0.01)	82 (0.03)
<i>P>F</i>						
Rain Exclusion	0.205	0.868	0.627	0.414	0.098	0.552
Fertilization	0.225	0.119	0.013	<0.001	0.002	0.039
Rain Exclusion x Fertilization	0.834	0.409	0.116	0.954	0.531	0.949

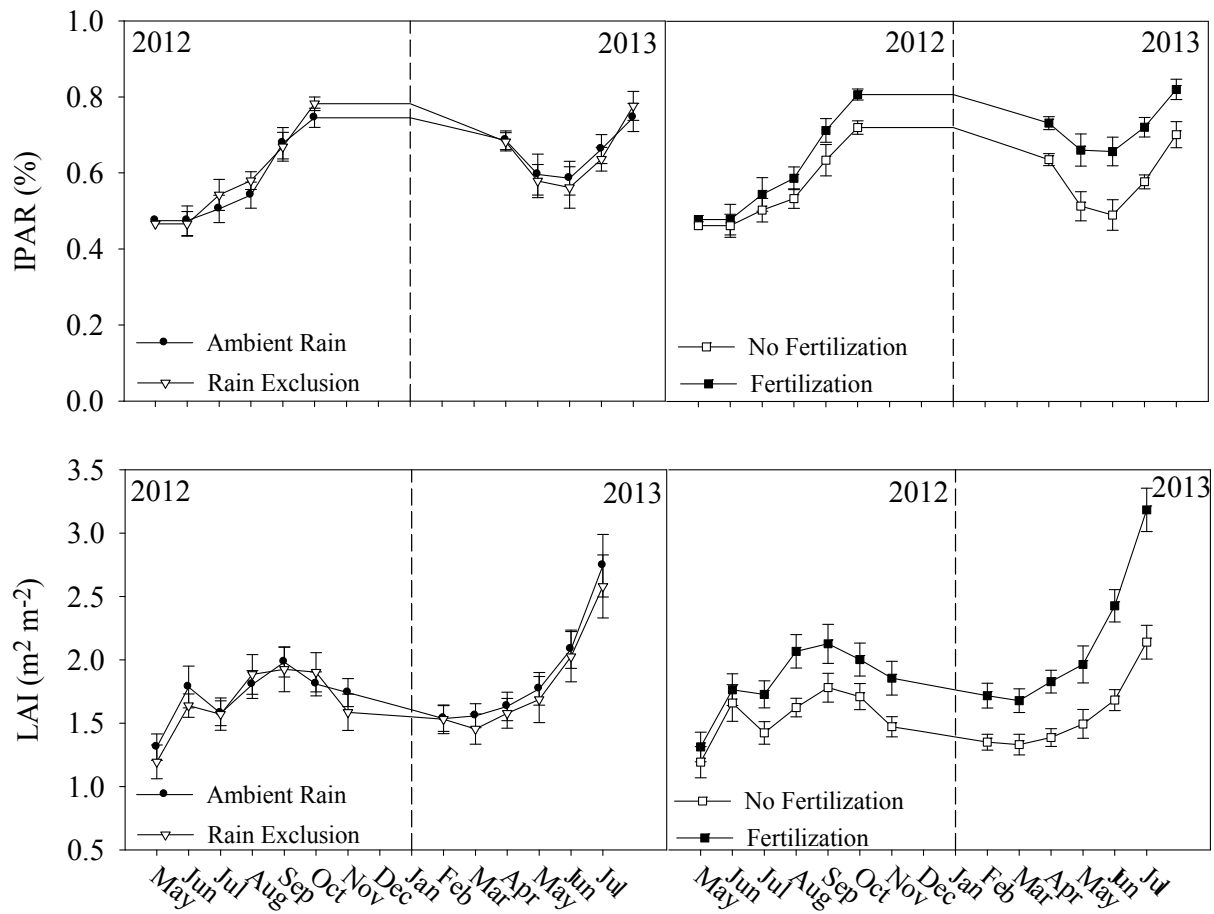


Figure 2.3.2. Intercepted photosynthetically active radiation (IPAR) and leaf area index (LAI) in response to rain exclusion and fertilization treatments in a 6-year-old loblolly pine plantation. Values of IPAR between November and March were interpolated following Allen et al. (2005). Dotted line represents start of 2013.

2.3.4 Leaf Physiology

During 2012, there were no interactive effects of rain exclusion and fertilization treatments and date of measurement on P_{net} , g_s , L_g , F_v/F_m , Ψ_L , or soil moisture during leaf gas exchange measurements (Table 2.3.3). The rain exclusion treatment decreased g_s by 21%, from 56.2 $\text{mmol m}^{-2} \text{s}^{-1}$ in the ambient rain treatment to 44.6 $\text{mmol m}^{-2} \text{s}^{-1}$ in the rain exclusion treatment (Table 2.3.3, Figure 2.3.3), and increased the L_g from 37% to 41% (Table 2.3.3, Figure 2.3.4). When averaged across all months in 2012, P_{net} decreased from 4.2 $\mu\text{mol m}^{-2} \text{s}^{-1}$ in the ambient rain treatment to 3.7 $\mu\text{mol m}^{-2} \text{s}^{-1}$ in the rain exclusion treatment (Table 2.3.3, Figure 2.3.3). Maximum efficiency of photosystem II (F_v/F_m) was not affected by either fertilization or rainfall treatments (Table 2.3.3, Figure 2.3.5). Predawn Ψ_L was decreased from -0.67 MPa in ambient rain treatment to -0.71 MPa in the rain exclusion treatment, but midday was not affected (Figure 2.3.6). Fertilization treatment had no significant influence on predawn or midday Ψ_L (Table 2.3.3). Soil moisture measured in-between rows was reduced by fertilization treatment, and soil moisture measured in-between trees was reduced by fertilization treatment and rain exclusion treatment (Table 2.3.3, Figure 2.3.7). In general in 2012, soil moisture was lower between rows under the rain exclusion trays than between trees.

In 2013, there was no interaction between treatments or between date of measurement and treatments for P_{net} , g_s , F_v/F_m , L_g , soil moisture, and midday Ψ_L (Table 2.3.3). There were no effects of treatment on P_{net} , g_s , F_v/F_m observed in 2013. The fertilization treatment increased L_g by 10% compared to the non-fertilized treatment and a trend ($p=0.100$) for reduced g_s with fertilization was detected (Table 2.3.3, Figure 2.3.3). A significant interaction between date of measurement and rain exclusion treatment was observed for predawn Ψ_L . The rain exclusion treatment decreased predawn Ψ_L only in June 2013. Midday Ψ_L was decreased by the rain

exclusion treatment in 2013 (Table 2.3.3). When averaged across all months in 2013, midday Ψ_L decreased from -1.07 MPa in the ambient rain treatment to -1.25 MPa in the rain exclusion treatment (Figure 2.3.6). Soil moisture measured in-between rows was decreased by fertilization, from 21.4% in the no fertilization treatment to 18.6% in the fertilization treatment. (Table 2.3.3, Figure 2.3.7). Soil moisture measured in-between trees was reduced from 18.1% in the no fertilization treatment to 16.4% in the fertilization treatment.

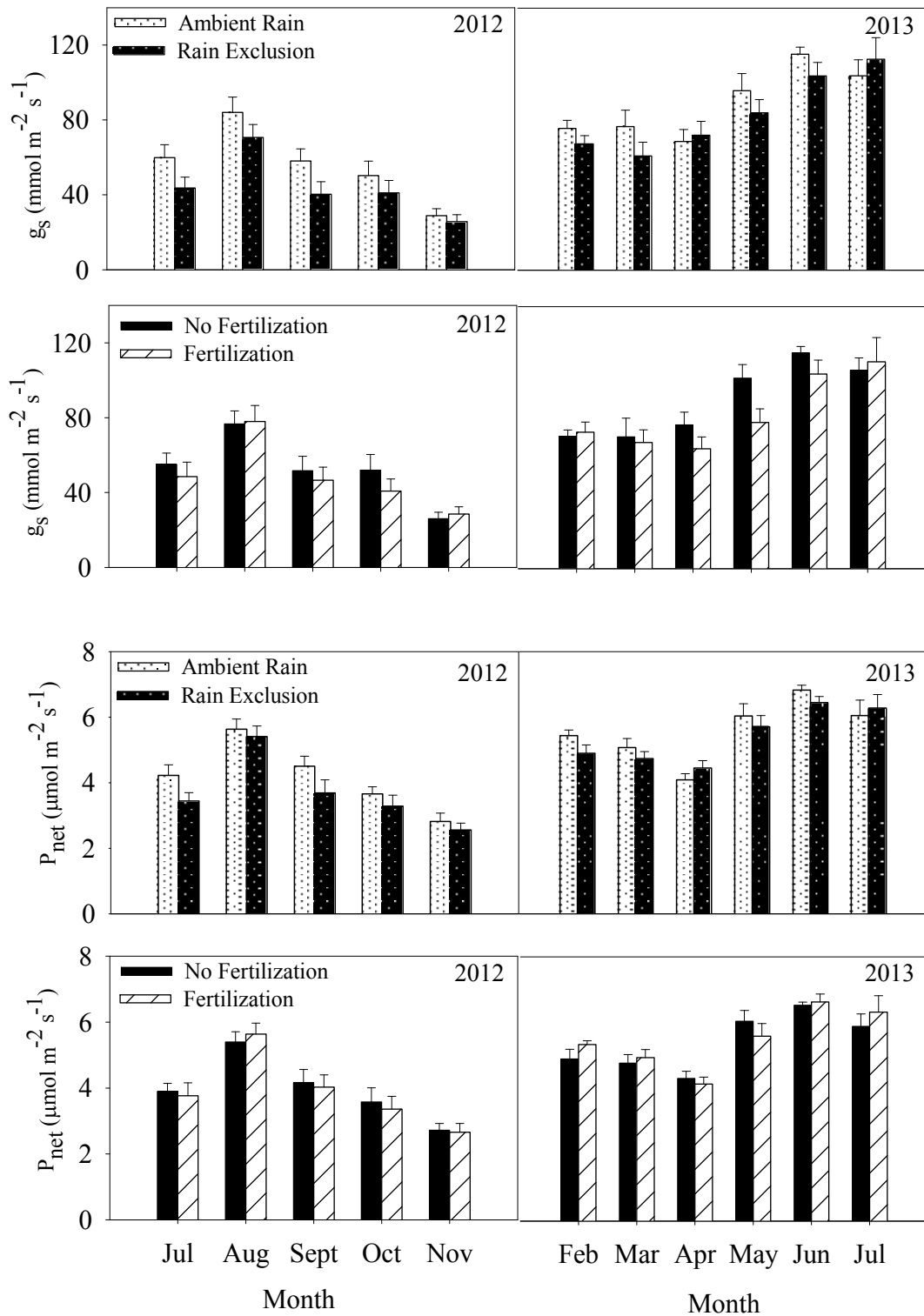


Figure 2.3.3. Mean (\pm SE) light-saturated net photosynthesis (P_{net}) and stomatal conductance (g_s) by month in response to rain exclusion and fertilization treatments in a 6-year-old loblolly pine plantation.

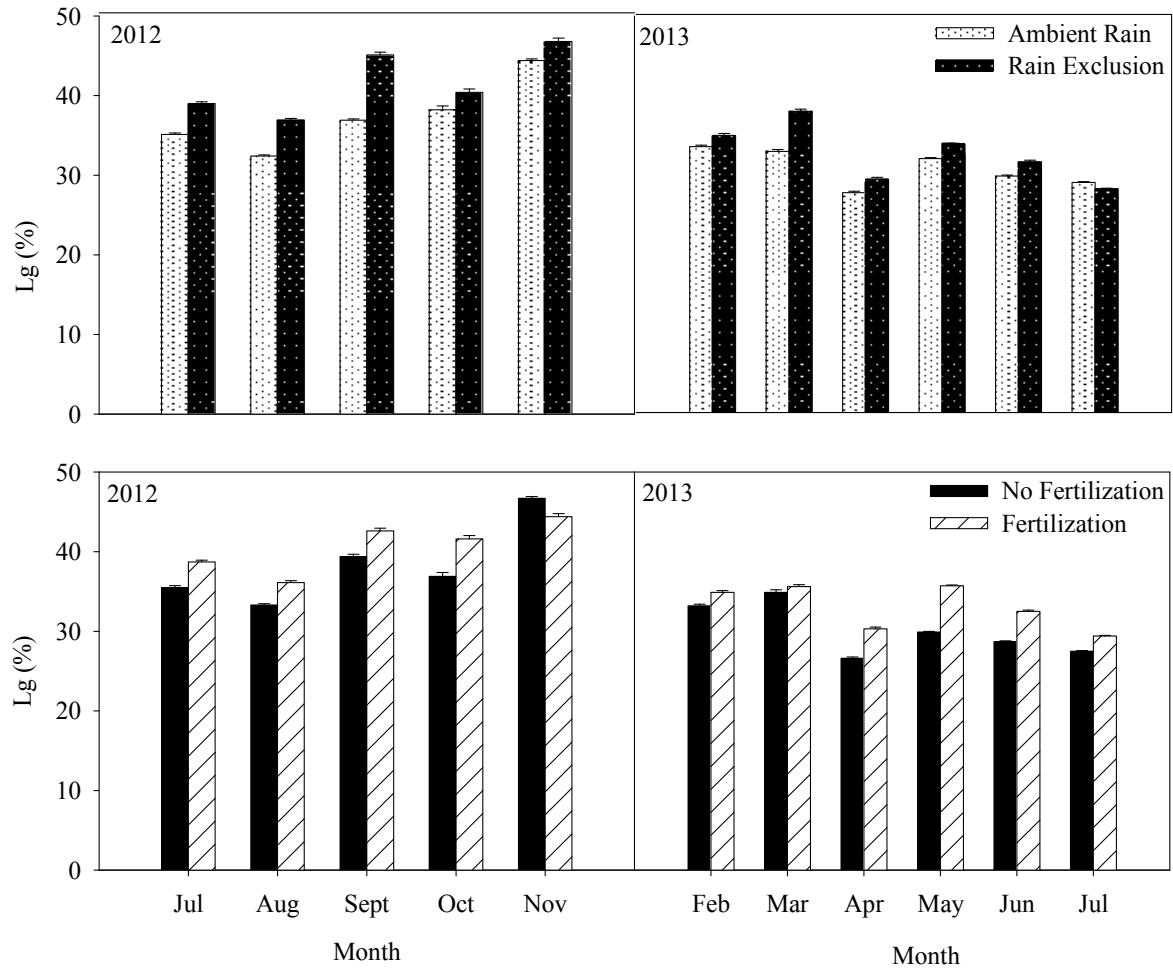


Figure 2.3.4. Mean (\pm SE) stomatal limitation (L_g) in response to rain exclusion and fertilization treatments by month in a 6-year-old loblolly pine plantation.

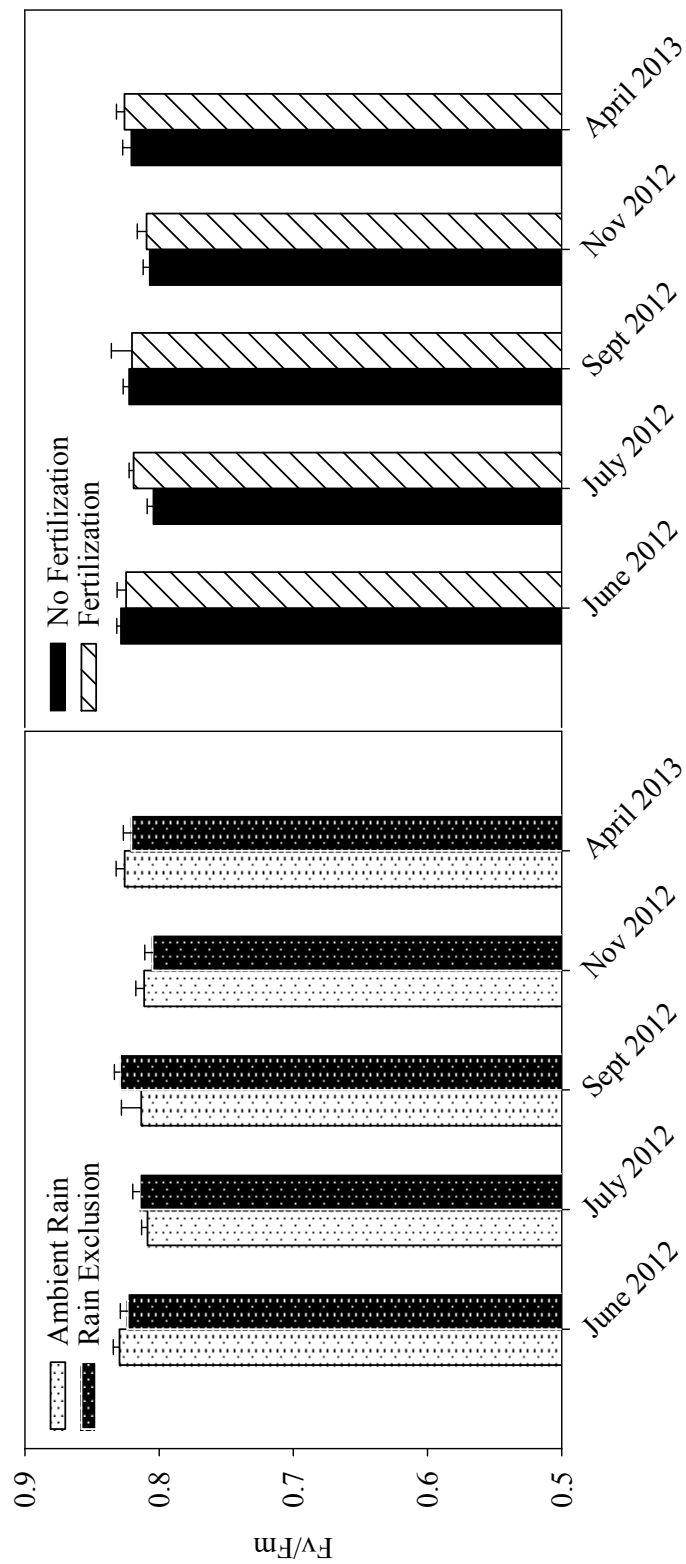


Figure 2.3.5. Monthly mean (\pm SE) of the maximum efficiency of the photosystem II (Fv/Fm) in response to rain exclusion and fertilization treatments in a 6-year-old loblolly pine plantation.

Table 2.3.3. Observed probability values for the effects of rain exclusion and fertilization treatments, month of measurement and interactions between treatments and month of measurement on light-saturated net photosynthesis (P_{net}), stomatal conductance (g_s), soil moisture between rows (SM_{br}), soil moisture between trees (SM_{bt}), efficiency of the photosystem II (Fv/Fm), stomatal limitation (L_g) and predawn and midday leaf water potential (Ψ_L) measured in 2012 and 2013 in a 6-year-old loblolly pine plantation.

	P_{net}	g_s	Predawn Ψ_L	Midday Ψ_L	SM_{br}	SM_{bt}	Fv/Fm	L_g
2012								
Month	<0.001	<0.001	<0.001	0.031	<0.001	<0.001	0.006	0.005
Rain Exclusion	0.005	0.003	0.002	0.981	0.081	0.040	0.725	0.014
Fertilization	0.630	0.213	0.978	0.283	0.021	0.020	0.687	0.176
Rain Exclusion x Fertilization	0.913	0.577	0.233	0.067	0.871	0.203	0.892	0.409
Month x Rain Exclusion	0.817	0.656	0.571	0.940	0.319	0.209	0.222	0.847
Month x Fertilization	0.926	0.846	0.778	0.902	0.889	0.068	0.255	0.811
Month x Rain Exclusion x Fertilization	0.980	0.740	0.604	0.964	0.184	0.844	0.317	0.499
2013								
Month	<0.001	<0.001	0.002	0.043	<0.001	<0.001	-	<0.001
Rain Exclusion	0.329	0.181	0.066	0.006	0.068	0.608	0.458	0.136
Fertilization	0.627	0.097	0.532	0.698	0.045	0.009	0.458	0.007
Rain Exclusion x Fertilization	0.998	0.944	0.414	0.261	0.249	0.643	0.807	0.520
Month x Rain Exclusion	0.443	0.287	0.041	0.925	0.235	0.886	-	0.429
Month x Fertilization	0.526	0.293	0.310	0.909	0.851	0.293	-	0.616
Month x Rain Exclusion x Fertilization	0.485	0.841	0.923	0.645	0.555	0.643	-	0.192

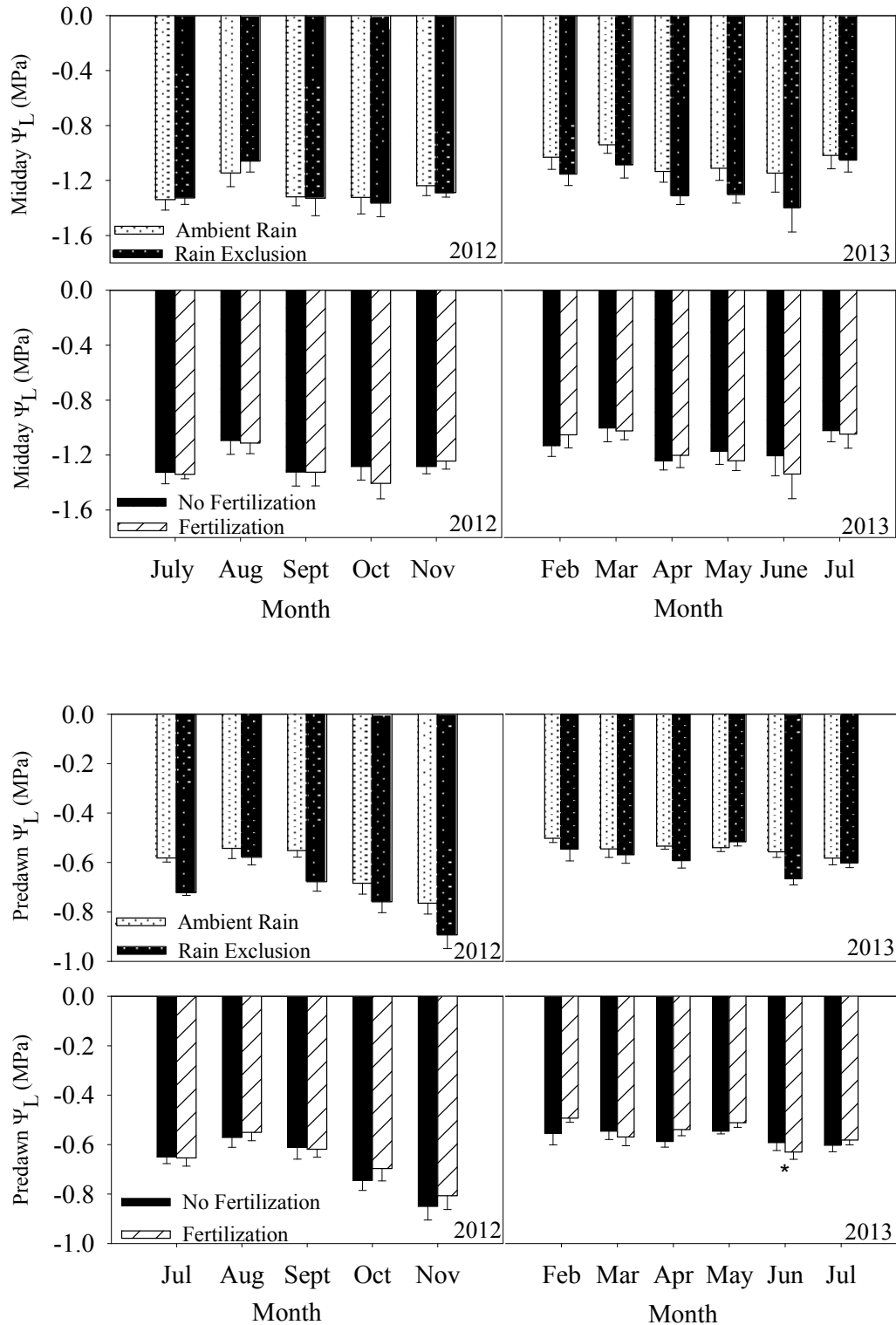


Figure 2.3.6. Mean (\pm SE) predawn and midday leaf water potential (Ψ_L) in response to rain exclusion and fertilization treatments by month in a 6-year-old loblolly pine plantation. Asterisks indicate a significant treatment effect within a month when a treatment by date interaction was detected.

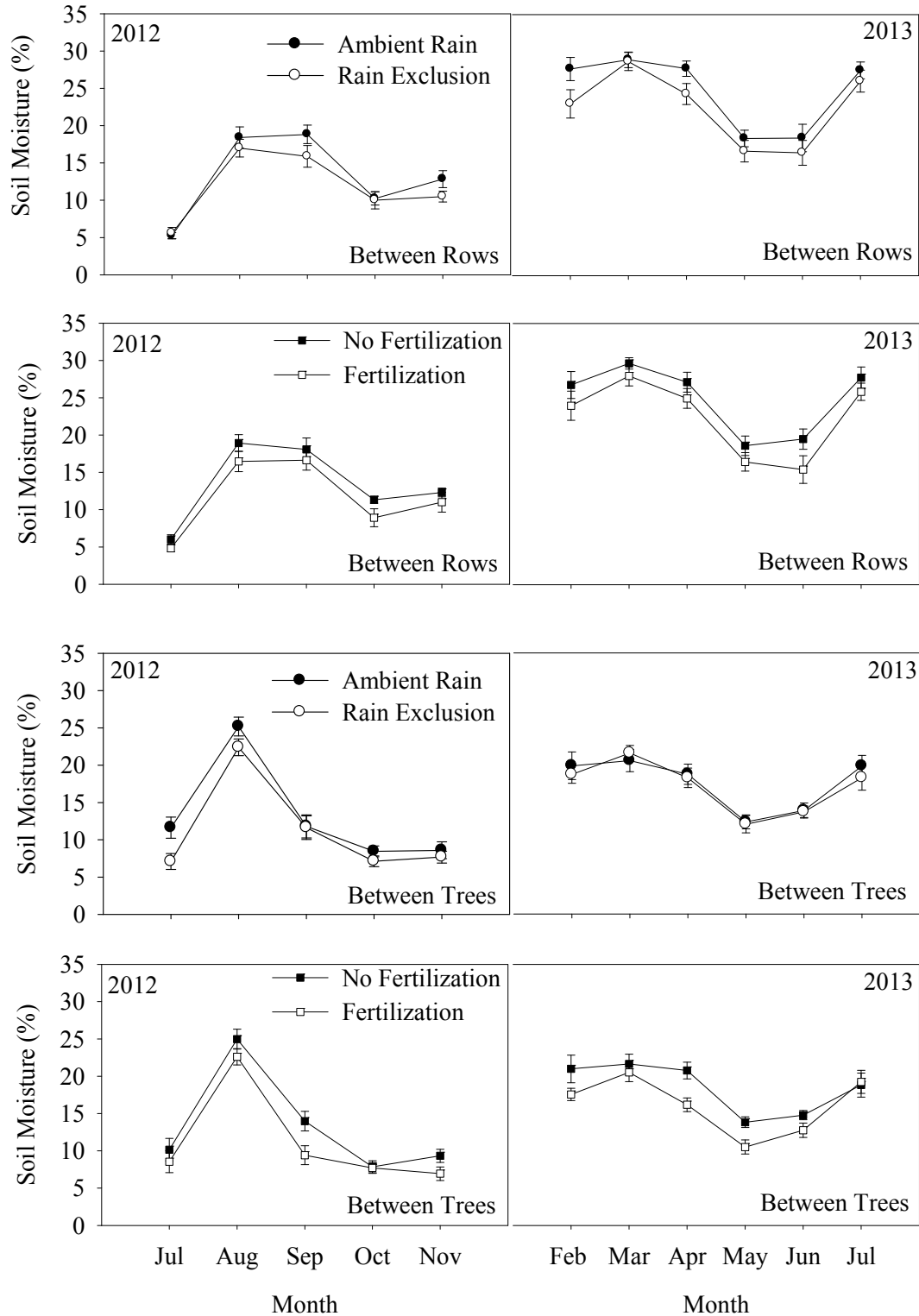


Figure 2.3.7. Mean (\pm SE) soil moisture measured to a 12 cm depth in-between trees and in-between rows in response to rain exclusion and fertilization treatments by month in a 6-year-old loblolly pine plantation. Soil moisture in-between rows was measured directly under rain exclusion trays.

2.3.5 Foliar Nutrients, $\delta^{13}\text{C}$ and Specific Leaf Area

Foliar $\delta^{13}\text{C}$ measured in 2012 was significantly increased by fertilization and rain exclusion treatment (Table 2.3.4). Fertilization increased $\delta^{13}\text{C}$ from -28.94‰ to -28.63‰ in the fertilization treatment, while rain exclusion treatment increased $\delta^{13}\text{C}$ from -29.03‰ to -28.54‰ (Table 2.3.4) indicating less discrimination of Rubisco to the heavier isotope in the rain exclusion and fertilization treatment.

Foliar nitrogen (N) was significantly higher in the first versus second flush of 2012, but no interactive effects of treatment and flush on foliar N were observed (Table 2.3.4). Fertilization increased foliar N from 13.8 mg g⁻¹ with no fertilization to 16.7 mg g⁻¹ with fertilization, while the rain exclusion treatment decreased foliar N from 15.7 mg g⁻¹ in the ambient rain treatment to 14.6 mg g⁻¹ in the rain exclusion treatment (Table 2.3.4). A significant interaction between flush, fertilization and rain exclusion treatments were observed for potassium (K) and phosphorus (P) (Table 2.3.4). Only in the second flush and in plots receiving the ambient rain treatment, did fertilization increase P. The rain exclusion treatment decreased foliar K only in the second flush and in plots receiving fertilizer.

Significant interactive effects between rain exclusion treatment and flush and between fertilization and rain exclusion treatments were observed for SLA (Table 2.3.4). The rain exclusion treatment increased SLA only in the second flush, from 56.9 cm² g⁻¹ to 61.9 cm² g⁻¹. Within the rain exclusion treatment, fertilization significantly increases SLA.

Table 2.3.4. Influence of rain exclusion and fertilization treatments on foliar content of nitrogen (N), potassium (K), phosphorus (P), foliar $\delta^{13}\text{C}$, and specific leaf area (SLA) in response to fertilization and rain exclusion treatments in a 6-year-old pine plantation. Means are averaged across first and second flush. Specific leaf area is reported on a projected area basis. Foliage was collected in October 2012.

Treatments	N (mg g^{-1})	P (mg g^{-1})	K (mg g^{-1})	$\delta^{13}\text{C}$ (‰)	SLA ($\text{cm}^2 \text{g}^{-1}$)
Ambient Rain	15.8 (0.08)	1.1 (0.003)	6.5 (0.03)	-29.0 (0.1)	54.0 (1.0)
Rain Exclusion	14.6 (0.07)	1.0 (0.003)	6.1 (0.03)	-28.5 (0.1)	56.7 (1.6)
No Fertilization	13.8 (0.06)	0.9 (0.002)	6.5 (0.03)	-28.6 (0.1)	54.6 (1.0)
Fertilization	16.6 (0.07)	1.1 (0.004)	6.1 (0.03)	-28.9 (0.1)	56.1 (1.6)
<i>P</i> < <i>F</i>					
Flush	<0.001	0.589	0.002	0.619	<0.001
Rain Exclusion	0.016	0.035	0.206	0.003	0.015
Fertilization	<0.001	<0.001	0.263	0.049	0.132
Rain Exclusion x Fertilization	0.508	0.004	0.197	0.185	0.046
Flush x Rain Exclusion	0.508	0.115	0.693	0.542	0.032
Flush x Fertilization	0.112	0.057	0.206	0.853	0.124
Flush x Rain Exclusion x Fertilization	0.137	0.045	0.033	0.619	0.980

2.4 Discussion

We hypothesized that nutrient availability would have a greater influence on LAI and IPAR rather than on leaf level physiology. In support of this hypothesis, fertilization did significantly increase foliar nitrogen concentrations, LAI and IPAR. Previous studies have shown that increased nutrient availability can have a significant effect on leaf area production, IPAR and growth of loblolly pine (Vose and Allen 1988; Albaugh et al., 1998, 2004; Allen et al., 2005; Will et al., 2005; Samuelson et al., 2008). Similar to prior studies, significant increases in LAI and IPAR were accompanied with increases in CAI and BAI in response to fertilization.

The rainfall manipulation treatments had no significant effect on LAI and IPAR, though foliar nitrogen concentrations were decreased in the rain exclusion treatment, most likely due to a reduction in soil moisture and transpiration which drives the uptake of nutrients from the soil through diffusion and mass flow. While leaf production and productivity in loblolly pine have been shown to increase with irrigation (Campoe et al., 2013; Samuelson et al., 2008), the increases from irrigation in these studies were small (6% to 31%) compared to the increases in response to fertilization (62% to 72%). Nutrient availability, rather than water availability, has been demonstrated to be the primary driver of production in loblolly pine (Herbert and Jack, 1998; Jokela et al., 2004, Albaugh et al., 2004). Jokela et al., (2004) provided a summary of seven long-term productivity experiments spread across the range of loblolly pine, from Florida to Oklahoma. Across all sites, a strong relationship between stemwood biomass increment and LAI was observed with LAI increasing in response to soil nutrient availability rather than site water balance. In this study, while fertilization had the greatest influence on LAI, IPAR and growth, BAI was sensitive to the rain exclusion treatment during a drought, most likely because the rainfall exclusion treatment decreased P_{net} and g_s in 2012.

No significant differences between treatments were observed for GE or cumulative IPAR. Growth efficiency ($7.1 \text{ m}^3 \text{ LAI}^{-1}$) and cumulative IPAR ($1900 \text{ MJ m}^2 \text{ yr}^{-1}$) are comparable to values reported by Albaugh et al., (1998) and Will et al., (2005) for loblolly pine stands of similar age and density. For example, Albaugh et al., (1998) found a range in GE from $7.1 \text{ m}^3 \text{ LAI}^{-1}$ to $9.2 \text{ m}^3 \text{ LAI}^{-1}$ in an 8-year-old loblolly pine plantation with a density of $1260 \text{ stems ha}^{-1}$, while Will et al., (2005) found a cumulative IPAR of $1568 \text{ MJ m}^2 \text{ yr}^{-2}$ for a 4-year-old loblolly pine plantation with a density of $1480 \text{ trees ha}^{-1}$. The most likely explanation for the lack of response in cumulative IPAR to fertilization is the time since treatment application. Fertilization was applied in March 2012 and increases in IPAR from fertilization were not detected until September 2012. Shifts in GE in response to fertilization can be attributed to changes in biomass allocation or rate of photosynthesis (Gough et al., 2004; Albaugh et al., 1998). No influence of fertilization on P_{net} was observed, which is likely why GE was similar between fertilization treatments. The lack of a significant response of P_{net} to fertilization could be due to leaf nitrogen concentration. The lowest foliar N level observed at our study occurred in the control (13.8 mg g^{-1}), which is well above the sufficiency level of 11 mg g^{-1} for loblolly pine (Allen, 1987).

Based on the PDSI, every month in 2012 was categorized as having severe or extreme drought conditions. Total rainfall for 2012 was 849 mm which is a 24% decrease from the 30 year average for Washington, GA. In 2013, conditions were described by the PDSI as moderately to extremely moist. The increase in rainfall seen at the end of 2012 and beginning of 2013 likely alleviated soil moisture reductions seen in all treatments during the previous year. Total precipitation for 2013 as of July was 520 mm which is 52% of the 30 year average annual

rainfall for Washington, GA. Soil moisture, averaged across all treatments, increased from 12.3% in 2012 to 20.5% in 2013.

Rain exclusion trays were designed to collect and transport 30% of throughfall rain off the plots. The amount of water that was captured and transported off the plots was not quantified, and the reduction of ambient precipitation in the rain exclusion treatment was possibly less due to partitioning of rainfall between throughfall and stemflow (Staelens et al., 2008). No response variable, including soil moisture measured to 12 cm depth, exhibited a 30% reduction in response to the rain exclusion treatment. On average, soil moisture was reduced by 13% in the rain exclusion treatment compared to the ambient rain treatment in 2012, but no significant reduction in soil moisture was observed in 2013. Cregg and Dougherty (1988) examined a 10-year-old loblolly pine plantation in southeastern Oklahoma subject to repeated drought and found that during a dry year predawn Ψ_L averaged nearly -0.75 MPa and during a relatively wet year predawn Ψ_L did not decline below -0.6 MPa. At our site in 2012, average predawn Ψ_L was -0.71 in the rain exclusion treatment, suggesting trees were water stressed, while in 2013 predawn Ψ_L averaged -0.5 MPa in the rain exclusion treatment providing evidence that the drought had been alleviated.

In 2012 during a severe drought, a decrease in P_{net} , g_s and predawn Ψ_L in response to the rain exclusion treatments was observed. The negative effect of water stress on gas exchange has been documented for loblolly pine (Seiler and Johnson, 1985; Teskey et al., 1986). Reductions in soil moisture availability can decrease the rate of photosynthesis through closure of stomata, and total plant photosynthesis by reductions in leaf expansion and increases in leaf shedding. The reduction in photosynthesis combined with reductions in leaf carbohydrate storage and leaf weight (Radoglou and Teskey, 1997) are likely responsible for the increase in SLA observed for

the second flush of 2012 in the rain exclusion treatment. Photosynthesis can be limited by non-stomatal limitations as well as stomatal limitations (Pallardy, 2008) and in loblolly pine stomatal limitations have been shown to range from 20% to 35% (Teskey et al., 1986; Samuelson et al., 2001). Severe drought can potentially cause photoinhibition by impairing photosystem II (PSII) activity (Cornic and Massacci, 1996). No evidence that damage to the PSII had occurred, because the efficiency of photosystem II (F_v/F_m) fell within the normal range, 0.75-0.85 (Maxwell and Johnson 2000). The reduction in P_{net} was most likely caused by stomatal closure. The L_g was higher in the rain exclusion treatment compared to the ambient rain treatment and values were higher (35% to 45%) compared to Teskey et al., (1986). The increase in foliar $\delta^{13}C$ in response to the rain exclusion treatment also supports stomatal limitation to P_{net} . Similarly, Choi et al., (2005), found that enhanced water availability increased the discrimination against foliar $\delta^{13}C$.

In addition to higher precipitation and the alleviation of drought in 2013, a shift in fine root production from the in-between row to in-between tree space or an increase in root production at depth where water may be more readily available may have occurred. Shifts in fine root production and greater production of roots at depth have been observed in response to changes in resource availability (Gower et al., 1992; Albaugh et al., 1998; Torreano and Morris 1998). Torreano and Morris, (1998) studied loblolly pine seedling root growth and distribution under water stress and observed that when upper layers of the soil rhizotron dried, root growth increased in the lower depths. Also, Gower et al., (1992) observed in rocky mountain Douglas-fir (*Pseudotsuga menziesii* var. *glauca* (Mayr) Franco) that fine root production was greater in plots where resource availability was limiting. Although no data on root production are available, it is

possible that fine root growth increased in the rain exclusion treatment. A shift to root production may also explain the decrease in BAI observed in the rain exclusion treatment.

Tang et al., (2004) examined the interactive effects of throughfall rain exclusion and fertilization on an 18-year-old loblolly pine plantation in southwest Louisiana. They reported decreases in P_{net} , g_s and predawn Ψ_L in response to the rain exclusion treatment, and also observed a significant interaction between rain exclusion and fertilization treatments for total crown foliar biomass and whole-crown photosynthesis. They observed within the rain exclusion treatment that total crown foliar biomass and whole-crown photosynthesis showed little response to fertilization, whereas in the ambient rain treatment total crown foliar biomass and whole-crown photosynthesis increased in response to fertilization. We observed interactive effects between rain exclusion and fertilization treatments on foliar nutrients. Foliar P in the second flush was increased only in the ambient rain treatment when receiving fertilization, while foliar K was reduced in the second flush only in the rain exclusion treatment when fertilizer was applied. The limited uptake of foliar nutrients caused by reduced soil moisture in the rain exclusion treatment could reduce the effectiveness of fertilization on leaf production and growth in the future.

While no interactive effects of rain exclusion and fertilization treatments on LAI, IPAR and leaf level physiology were observed, the rain exclusion and fertilization treatments had an effect on water use and availability. Midday Ψ_L became more negative in the rain exclusion treatment in 2013. As LAI increased in all plots from 2012 to 2013, this increase probably resulted in increased transpiration causing a more negative midday Ψ_L . Soil moisture was also reduced by the fertilization treatment in both 2012 and 2013. The small increase in LAI observed in 2012 (17%) was probably not large enough to cause greater susceptibility to drought. In 2013,

LAI increased by 49% but high ambient precipitation likely offset the potential for drought stress associated with increased LAI. Diminished soil moisture under conditions of more normal precipitation with increased LAI could lead to interactions between the rain exclusion and fertilization treatments in the future.

2.5 Conclusions

In summary, reductions in water availability affected leaf level physiological processes, reducing the rate of photosynthesis through stomatal closure, but nutrient availability remained the primary driver of growth through increases in LAI. Our study demonstrated that reduced water availability can also have a negative effect on growth in loblolly pine. In 2012 during a drought, the rain exclusion treatment caused a reduction in soil moisture, predawn Ψ_L , g_s , and P_{net} , and BAI. Fertilization had the largest influence on growth and despite the severe drought in 2012, fertilization increased growth, but water availability may have played a role in the efficiency of the fertilization treatment. Increase in LAI in 2012 was relatively low compared to increases observed in 2013. In 2012, LAI increased by 17% in response to fertilization, while in 2013, a year with above average rainfall, LAI was increased by 49%, suggesting that the increase in LAI with fertilization was influenced by precipitation. Growth in 2013 will determine if differences in LAI between 2012 and 2013 resulted in an equally substantial difference in growth. Based on observations from this study, future response to fertilization in loblolly pine plantations could be limited by water availability and, depending on the timing of drought the increase in LAI with fertilization may exacerbate the effect of reduced precipitation on growth and ultimately the ability to sequester CO₂.

Pine plantations are estimated to cover 16 million ha in the southeastern United States (Wier and Greis 2011) and account for 17% of forestland in the region. Southern pine plantations sequester as much as 210 Tg C yr⁻¹ (Johnsen et al. 2001), with the ability to increase this amount through silvicultural practices such as fertilization (Albaugh et al., 2012). Small declines in loblolly pine annual growth rates in response to shifts in climate could have a detrimental effect on the amount of CO₂ sequestered across millions of hectares. If increased growth responses due to fertilization are diminished by future predicted reductions in precipitation, other viable options, such as the deployment of more drought tolerant seed sources and lower planting densities will need to be examined in order to sustain the growth and carbon sequestration of this species in the face of changing climate.

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