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Post-fertilization physiology and growth performance of loblolly pine clones

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Summary The physiological processes leading to enhanced growth of loblolly pine (*Pinus taeda* L.) following fertilization are not clearly understood. Part of the debate revolves around the temporal response of net photosynthetic rate (A_n) to fertilization and whether the A_n response is always positive. We measured light-saturated photosynthetic rate (A_{sat}), dark respiration rate, growth and crown silhouette area in eight clones of loblolly pine before and after nitrogen (N) fertilization (112 kg ha^{-1}) to track the initial physiological changes prior to any changes in growth. Overall, there were positive photosynthetic and growth responses to fertilization; however, there were pronounced physiological and growth differences among clones, even among clones with the same parents. Clones 4, 6 and 7 showed large volume growth and A_{sat} responses to fertilization. Clone 1 and Clone 8 (a full-sibling of Clone 7) mainly showed a volume growth response, whereas Clone 2 (full-sibling of Clone 1) showed an A_{sat} response only. Clone 5 (full-sibling of Clone 6) showed little response to fertilization, whereas Clone 3 (full-sibling of Clone 4) showed a negative A_{sat} response. Thus, within-family variation warrants further study to ensure that relatively expensive clonal material is used efficiently.

Keywords: crown area, gas exchange, net photosynthesis, nitrogen, Pinus taeda.

Introduction

Over the last several decades, forest managers have striven to increase forest productivity by silvicultural techniques, including fertilization (Fox et al. 2007). In many areas, nutrients are a major factor limiting forest productivity. Nitrogen (N) and phosphorus are considered crucial to the productivity of forest tree species (Helms 1976). Loblolly pine stands in the southeastern USA are often fertilized with positive results (Jokela and Stearns-Smith 1993, Colbert and Allen 1996). Over 480,000 ha of planted pine were fertilized in 2004 (Fox et al. 2007).

Although fertilization often results in increased loblolly pine tree biomass, the physiological mechanisms underlying this response are unclear. Leaf photosynthetic rate, leaf respiration rate, leaf area and tree surface area govern net carbon gain (Teskey et al. 1987), but which of these factors responds to fertilization and results in increased growth is unclear. Several studies have focused on gas exchange and leaf area in fertilized forests, but the results have been inconsistent. Some studies have suggested that productivity of loblolly pine stands increases following fertilization because of increased leaf area and stem wood growth (Teskey et al. 1987, Vose and Allen 1988). Other studies have shown that fertilization increases net photosynthetic rates (A_n) and have partially attributed increased growth to this mechanism (Murthy et al. 1996, Kellomaki and Wang 1997, Samuelson 2000). Similarly, studies on the relationship between respiration and fertilization have yielded conflicting results. For example, several studies have demonstrated a positive relationship between fertilization and respiration (Strand 1997, Kellomaki and Wang 1997, Zhang et al. 1997), whereas other studies have shown there is no relationship (Schaberg et al. 1997, Roberntz and Stockfors 1998, Lavigne et al. 2001). Some of the differences among studies may have resulted from varying sampling times, sampling intensities, tree age, degree of nutrient deficiency or other experimental factors.

Because loblolly pine grows over a broad geographic range in the southern USA, its geographic variability has been well studied (Sierra-Lucero et al. 2002). There are several provenances (e.g., Atlantic Coastal Plain, Central Florida, Gulf Coastal Plain, Lost Pines in central Texas), and their differences in growth are generally well characterized. For example, eastern and southern provenances are faster growing than western provenances such as the Lost Pines. The western provenance has higher drought and disease tolerance than the eastern provenances (Wells 1985). Thus, selective tree breeding may result in superior families that out-produce their unimproved counterparts because of higher tolerance of environmental stress, higher biomass allocation or higher growth efficiency. Most loblolly pine propagation is by seed, which tends to create high genetic variation among trees (Zobel and Talbert 1991).

There is variation in genotypic responses to different treatments and environmental conditions (Teskey et al. 1987,

McKeand et al. 2006). Varying genotypic responses in growth and physiological characteristics have been documented in loblolly pine (Li et al. 1991, McCrady and Jokela 1996, Roberts et al. 2003, Roth et al. 2007). There are reports that fertilized loblolly pine clones may differ in their responses. For example, Li et al. (1991) found a significant family and family × N interaction on loblolly pine tree biomass in a 20-week greenhouse study with low (5 μmol mol^{-1}) and high (50 μmol mol^{-1}) N treatments. In 5-year-old loblolly and slash pine, Roth et al. (2007) found genotype × location and genotype × silvicultural treatment intensity interactions on volume and basal area. Samuelson (2000), however, showed little if any family × N interaction on P_n, R_d, stomatal conductance (g_l) or foliar N in seedlings of loblolly and slash pine (*Pinus elliottii* Englm. var. *elliottii*), and there were no significant two-way interactions on fine root allocation in either species during the greenhouse study.

These findings emphasize the need to determine the range of variation and inheritance of important physiological and growth characteristics to improve future loblolly pine breeding. We also need to determine how these characteristics change with increasing silvicultural intensity (McKeand et al. 2006). For example, how much variability in growth and physiological responses to fertilization exists between clones with common parents? The objectives of our study were to: (1) clarify the short-term physiological mechanisms underlying the increase in loblolly pine biomass in response to fertilization; and (2) investigate the differences among several full-sib clonal varieties in gas exchange and aboveground biomass changes shortly after fertilization.

Materials and methods

Study site

The study site was located in Patrick County, Virginia (36°40′ N, 80°10′ W) at the Reynolds Homestead Forestry Research Center on the upper Piedmont province, where the topography consists of gently rolling hills. Elevation varies between 300 and 350 m. The site has a temperate climate with warm humid summers and cool wet winters. Mean monthly minimum temperature is –4.0 °C, and mean monthly maximum temperature is 29.3 °C. Precipitation is evenly distributed throughout the seasons and averages 1.3 m $year^{-1}$ (Hoare 2005).

The soils consist of Lloyd clay loam (fine, kaolinitic, thermic Rhodic Kanhapludults), Louisa loam (loamy, micaceous, thermic, shallow Ruptic-Ultic Dystrudepts) and Hiwassee loam series (very-fine, kaolinitic, thermic Rhodic Kanhapludults). They are well-drained deep Ultisols originating from granite, schist and gneiss parent material. The site was heavily farmed for two centuries, which destroyed most of the original A horizon. There is a truncated profile, with a surface Ap horizon and clayey B horizons mixed in below.

The study site consists of four blocks, each divided into two 338.2 m^2 plots containing 25 loblolly pine clones that were planted on May 19, 2003. One ramet of each clone was planted in each plot. The clones were planted at a 3.0 × 2.4 m spacing. A buffer row of seedlings surrounds and separates the eight study plots. The clones were provided by the Forest Biology Research Cooperative (FBRC; University of Florida, Gainesville, FL). The clones were rooted cuttings and each family was from different plants in the same full-sib family. The ortets for each clone were selected from the Loblolly Pine Lower Gulf Elite Breeding Population, which contains both Atlantic Coastal and Florida provenances.

Site preparations included a treatment of Roundup (glyphosate), ripping and shallow cultivation in the planting rows. The plots were mowed and manually weeded periodically during the growing seasons. Oust (sulfometuron methyl) and Roundup herbicides were used to remove weed competition from the seedling rows. No other environmental variables were controlled to allow the seedlings to experience typical field conditions.

Nutrient application

Within each block, one plot was randomly chosen for fertilization and the other served as the unfertilized control plot. Fertilization consisted of 224 kg ha^{-1} of diammonium phosphate (DAP) and 184 kg ha^{-1} of ammonium nitrate. Therefore, 112 kg ha^{-1} of elemental N and 53 kg ha^{-1} of elemental phosphorous were applied. Fertilizer was spread by hand by a banded application technique over the four fertilization treatment plots on May 6, 2004.

Measurements

Before fertilization, growth measurements including height and ground-line diameter were collected on all seedlings. Seedling growth was monitored periodically during the growing season by tracking changes in height and ground-line diameter. Any dead or dying seedlings were noted and excluded from the study. Three pre-fertilization height measurements were taken starting on December 29, 2003, and two pre-fertilization ground-line diameter measurements were taken starting on March 5, 2004. An index of stem volume was calculated by multiplying the measured height by the measured ground-line diameter squared for each seedling on the dates when both data were collected. From the 25 loblolly pine clones planted, eight clones were chosen for intensive physiological and growth measurements. Table 1 shows the parents of each of the eight selected clones (note that there are pairs of clones produced from ortets with the same male and female parents).

To characterize the short-term effects of fertilization on loblolly pine physiology, measurements were taken on April 30 and May 6, 2004, before fertilization, to establish a base of comparison for subsequent measurements after fertilizer application. Net photosynthesis and foliar carbon (C) and N analysis were measured on one new, fully expanded needle fascicle per seedling, and R_d was measured on two fascicles per seedling.

Net photosynthesis was measured in saturating light (A_{sat}) with a Model LI-6400 portable photosynthesis system (Li-Cor) on a fascicle of needles immediately after detachment from the seedling (< 3 min). Measurements of A_{sat} are

Table 1. Clone ID of the eight loblolly pine (*Pinus taeda*) clones measured and their associated parents.

Clone	Maternal parent	Paternal parent
1	A	B
2	A	B
3	C	D
4	C	D
5	E	F
6	E	F
7	A	G
8	A	G

generally unaffected by twig removal from the tree for up to 30 min (Ginn et al. 1991), and a sample of 10 needles from 10 trees in a preliminary study showed no change in A_{sat} following detachment for up to 30 min. Once A_{sat} became stable, it was recorded three times per fascicle over a period of 10 s and the mean of the three samples was used for subsequent data analysis. Thus, 64 mean A_{sat} values (eight clones × two fertilization treatments × four blocks) were used for data analysis. All chamber conditions remained at ambient with the exception of photosynthetically active radiation (PAR) = 1600 µmol m^{-2} s^{-1} and carbon dioxide concentration ($[CO_2]$) = 360 µmol mol^{-1}. The data collected were reflective of the environmental conditions experienced by the field-grown seedlings, whereas the set chamber values served as a basis for comparison with the study of Gough et al. (2004*a*). We calculated A_{sat} on a per leaf area basis (Ginn et al. 1991):

$$A_{sat,\,area} = nld + \pi dl$$

where n = number of needles per fascicle, l = needle length in the chamber and d = fascicle diameter. After the A_{sat} measurements, the diameter of each removed fascicle was measured with digital calipers and the fascicles were enclosed in an envelope and dried at 65 °C for later tissue nutrient analysis.

Dark respiration measurements, which were made by the same basic method as the A_{sat} measurements, were conducted monthly between 2300 and 0100 h, within 12 h of measuring A_{sat} on six of the same clones. Chamber conditions were maintained as described for the A_{sat} measurements except that the Li-Cor blue/red chamber light was turned off (PAR = 0 µmol m^{-2} s^{-1}) and carbon dioxide concentration = 400 µmol mol^{-1}. The higher CO_2 concentration was chosen based on previous field measurements of CO_2 concentrations between 400 and 500 µmol mol^{-1} during the night. For each clone, R_d was measured in two fascicles, and the diameter of each removed fascicle was measured with digital calipers.

Fascicles measured for A_{sat} were dried at 65 °C and ground in a Wiley mill before C and N analysis at the USDA Forest Service Southern Research Station laboratory (Research Triangle Park, NC) with a Carlo-Erba elemental analyzer (Model NA-1500, Fison Instruments, Danvers, MA).

Post-fertilization measurements of A_{sat} were conducted immediately following a soaking rain within a week of fertilizer application. Measurements were conducted as for the pre-fertilization measurements. Initially, measurements were conducted twice a week so that any short-term physiological changes in foliar C and N concentrations and A_{sat} would be captured. Measurements were taken less frequently later in the season. Measurements of A_{sat} ceased in April 2005, one year after treatment initiation, and R_d measurements were taken through most of the growing season until October 2004.

Total soil nitrogen

Samples for measurement of total soil N concentration were collected to a depth of 10 cm with a 2.5-cm diameter push-tube at the base of the same seedling clones measured for R_d. Soil samples were collected weekly just after fertilization and then monthly once the total soil N concentration began to stabilize during the fall and winter. Samples were dried, ground and analyzed for total N by dry combustion at the USDA Forest Service Southern Research Station laboratory (RTP, NC) with a Carlo-Erba elemental analyzer (Model NA-1500, Carlo Erba/Thermo Electron, Milan, Italy).

Crown silhouette area

Nondestructive estimates of crown silhouette area (CSA) of each seedling were made periodically during the growing season. Digital photographs were taken following each A_{sat} sampling period, with an Olympus C2500L camera, between 1300 and 1600 h when the sun was overhead or at a slight angle. A red plywood backdrop was used as a uniform background. A small white square measuring 100 cm^2 was taped to the red backdrop for use as a reference area in subsequent CSA calculations. Photographs were taken from the north side of the seedlings looking south at about 0.5 to 1.5 m from the seedling depending on its height and width. All photographs were taken in the same order by block on each sampling date between June and December 2004. Initially, photographs were taken every two weeks to track any immediate changes in crown area and then monthly with the onset of winter.

Computer analysis involved removing the uniform red background to leave only the seedling stem and canopy as well as the small white reference square. The numbers of pixels in the isolated seedling and the reference square were determined separately. The pixel counts were converted to an area value (cm^2) by cross-multiplying the two pixel counts by the reference square area of 100 cm^2.

Crown efficiency

Mean crown efficiency of each seedling was estimated based on the photographic CSA value and seedling volume. Seedling volume at the end of the growing season was divided by its corresponding CSA value to determine how much stem volume could be produced given a certain crown area. The values provided an estimate of which seedlings produced more stem volume with less crown area from June until December 2004. Initially, these measurements were taken every 2 weeks and then monthly with the onset of winter.

Statistical analysis

The study had a randomized block design with a split plot. Significant differences in the sampled variables during and at the end of the growing season were evaluated by analysis of variance (ANOVA). We conducted time series analysis of the effects of fertilization, time and clone and their interactions on the variables measured over several time periods. An $\alpha = 0.05$ was used for all tests except for the mean separation test used on the crown efficiency variable, which had an $\alpha = 0.1$. Appropriate transformations were used if the variance assumptions were not met in the A_{sat}, R_d and growth data.

Results

Gas exchange response

Time series analysis of the 24 sample dates showed significant clone × date ($P < 0.0001$) and fertilizer × date ($P = 0.0026$) interactions on A_{sat}, but no significant response for R_d (Table 2). Nine sampling dates during the growing season showed significant differences between at least two clones at $\alpha = 0.05$ (Figure 1). A Duncans MRT at $\alpha = 0.05$ showed that Clone 6 had a significantly higher mean A_{sat} than at least one other clone on six of those dates (data not shown). Among clones, Clone 6 typically had the highest mean A_{sat}, and Clones 5 (full-sib of Clone 6) and Clone 7 both had at least one date with a significantly higher A_{sat} ($P = 0.05$). In general, Clone 2 had the lowest mean A_{sat} over the same period. In the other clones, A_{sat} tended to vary over time with few significant differences among sampling dates compared with Clones 5–7. Averaged across the clones, A_{sat} was higher in fertilized seedlings on three occasions: May 25 ($P = 0.034$), June 16 ($P = 0.078$) and September 24 ($P = 0.064$) (Figure 2), with mean A_{sat} generally being higher in fertilized seedlings than in unfertilized seedlings for most of the growing season.

Volume growth response

Time series analysis of volume growth showed a significant date × fertilization × clone interaction ($P = 0.0003$; Table 2). Increases in volume growth in response to fertilization differed in magnitude among clones (Figure 3). For example, Clone 4 was the smallest clone at the end of the growing season in control plots; however, it showed a 154% increase in volume growth with fertilization. Volume growth of Clones 6 and 7 also responded positively to fertilization (73 and 112%, respectively). In contrast, volume growth of Clones 1, 2, 3, 5 and 8 responded little or not at all to fertilization (22, 0, 19, –3 and 16%, respectively). Clones 1 and 2 were the only full-siblings with relatively similar volume growth responses to fertilization. Full-sib pairs 3 and 4, 5 and 6, and 7 and 8 all had a large volume growth response to fertilization in one clone, but not in the other.

The relationship between each clone's seasonal mean A_{sat} and its volume at the end of the year demonstrated high clonal variation to fertilization. Some clones showed a large response in both variables, whereas others showed only a volume growth response, an A_{sat} response, or no response (Figure 4).

Table 2. The partial ANOVAs from the time series analysis and *P* values for A_{sat}, R_d, CSA, foliar N, crown efficiency and volume growth across both treatments sampled from May 2004 to April 2005 excluding the two pre-fertilization dates.

Source	df	*F*	*P*
Light-saturated net photosynthesis			
Fert	1	7.06	0.0766
Clone	7	3.27	0.0076
Clone × Fert	7	1.87	0.10
Date	23	463.83	< 0.0001
Date × Fert	23	2.05	0.0026
Date × Clone	161	1.60	< 0.0001
Date × Fert × Clone	161	0.71	0.996
Dark respiration			
Fert	1	0.53	0.5178
Clone	5	1.77	0.15
Clone × Fert	5	2.02	0.11
Date	4	71.23	< 0.0001
Date × Fert	4	1.26	0.288
Date × Clone	20	0.63	0.8851
Date × Fert × Clone	20	0.77	0.7475
Foliar N concentration			
Fert	1	4.96	0.112
Clone	5	2.48	0.032
Clone × Fert	5	1.26	0.29
Date	19	26.54	< 0.0001
Date × Fert	19	1.47	0.089
Date × Clone	133	0.83	0.912
Date × Fert × Clone	133	0.68	0.9968
Crown silhouette area			
Fert	1	2.18	0.235
Clone	7	3.1	0.01
Clone × Fert	7	2.14	0.06
Date	11	182.28	< 0.0001
Date × Fert	11	6.92	< 0.0001
Date × Clone	77	1.74	0.0003
Date × Fert × Clone	77	1.19	0.139
Volume			
Fert	1	0.55	0.512
Clone	7	1.5	0.19
Clone × Fert	7	0.5	0.83
Date	21	1524.83	< 0.0001
Date × Fert	21	9.83	< 0.0001
Date × Clone	147	1.63	< 0.0001
Date × Fert × Clone	147	1.5	0.0003
Crown efficiency			
Fert	1	0.04	0.85
Clone	7	2.49	0.032
Clone × Fert	7	0.53	0.80
Date	11	133.95	< 0.0001
Date × Fert	11	2.06	0.022
Date × Clone	77	0.85	0.814
Date × Fert × Clone	77	0.97	0.562

For example, Clones 4, 6 and 7 showed relatively large volume and A_{sat} responses to fertilization, and these clones were largely responsible for the overall trend seen in Figure 4. Clones 1

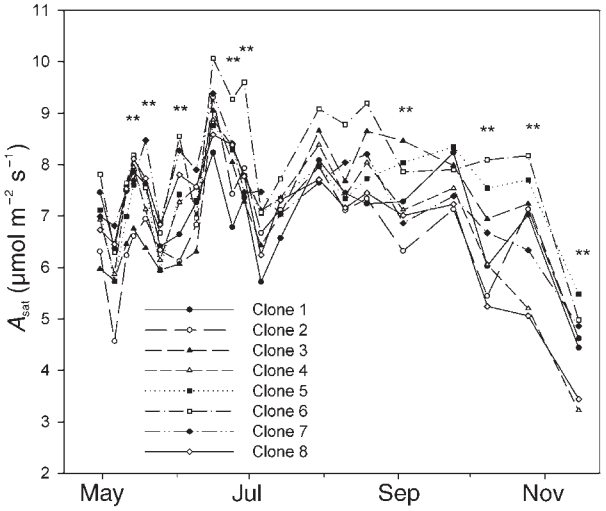

Figure 1. Clonal light-saturated photosynthetic rates (A_{sat}) from April to December 2004 in a 2-year-old loblolly pine (*Pinus taeda*) plantation in the Piedmont of Virginia. Significant differences among clones are indicated by asterisks: *, $P < 0.1$; and **, $P < 0.05$.

and 8 mainly showed a volume growth response to fertilization, whereas Clone 2 showed an A_{sat} response only. For Clone 5, which has the same parents as Clone 6, fertilization had no effect on either variable, whereas Clone 3, which has the same parents as Clone 4, showed a large negative A_{sat} response to fertilization.

Foliar nitrogen concentration, crown silhouette area and crown efficiency

Time series analysis of foliar N concentrations showed a significant clonal effect ($P = 0.03$; Table 2). Clone 6 had a significantly higher foliar N concentration than all other clones (Figure 5). Furthermore, Clone 6 had the highest mean foliar N concentration over most of the growing season, whereas the other clones had similar but slightly lower N concentrations. Clones 4 and 7 had comparatively low foliar N concentrations, and responded well to fertilization. Linear regression analysis across all clones revealed a small, but significant positive correlation between foliar N concentration and A_{sat} ($r^2 = 0.09$, $P < 0.0001$). Figure 6 shows that, compared with unfertilized seedlings, fertilized seedlings had slightly higher A_{sat} (7.70 versus 7.45 µmol m^{-2}s^{-1}) and slightly higher foliar N concentrations (1.7 versus 1.4% N).

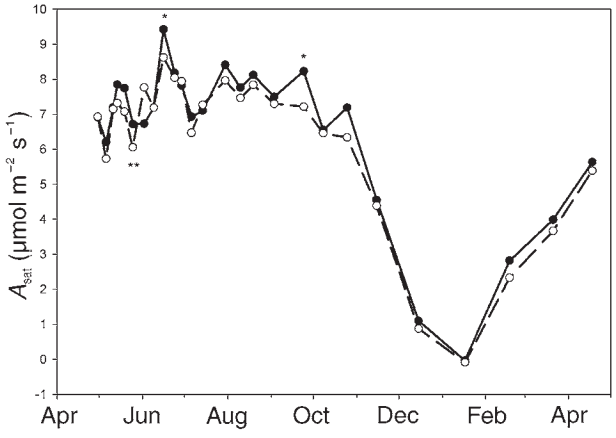

Figure 2. Mean ($n = 8$) light-saturated photosynthetic rates (A_{sat}) for fertilized (●) and unfertilized (○) seedlings from April 2004 to April 2005 in a 2-year-old loblolly pine (*Pinus taeda*) plantation in the Piedmont of Virginia. Seedlings were fertilized May 6, 2004. Significant differences between treatments are indicated by asterisks: *, $P < 0.1$; and **, $P < 0.05$.

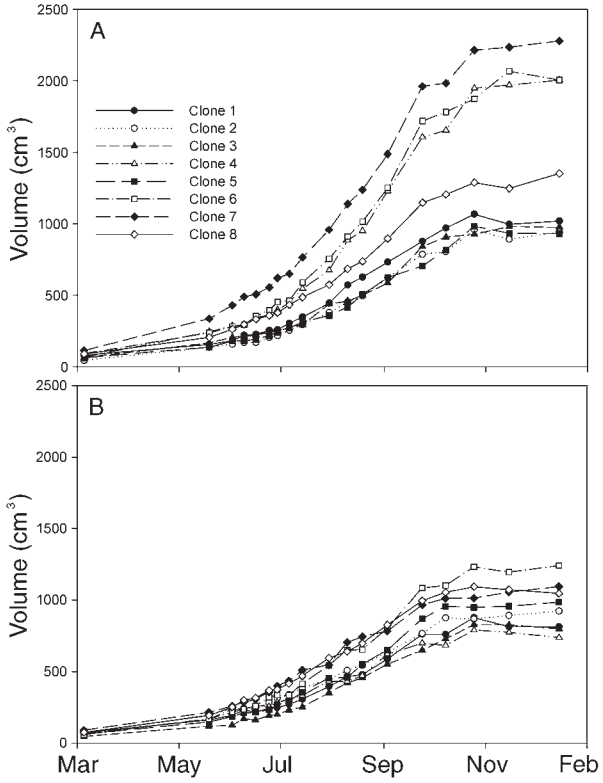

Figure 3. Mean clonal volumes for (A) fertilized and (B) unfertilized seedlings from April to December 2004. Seedlings were fertilized May 6, 2004.

Time series analysis of CSA revealed significant interactions for clone × date ($P = 0.0003$), clone × fertilization ($P < 0.06$) and fertilization × date ($P < 0.0001$; Table 2). The clone × date interaction on CSA showed significant differences ($P = 0.05$) between at least one clone for all sampling dates except the first one (Figure 7). The trend in CSA among clones over time mimicked the pattern seen in volume growth with Clones 4, 6, 7 and 8 showing the greatest CSA values for most sampling dates. Clones 1, 2, 4 and 5 were grouped together with distinctly lower CSA values and on three dates were all significantly different ($P = 0.05$) from at least one of the top four clones. Further examination of the clone × fertilization interaction revealed Clone 4 had a highly significant CSA response to fertilization ($P = 0.02$) by the end of the growing season (Figure 8). A comparison of the CSA responses to fertilization between the other clones yielded no significant results. Clones 1, 6 and 7 had comparatively larger

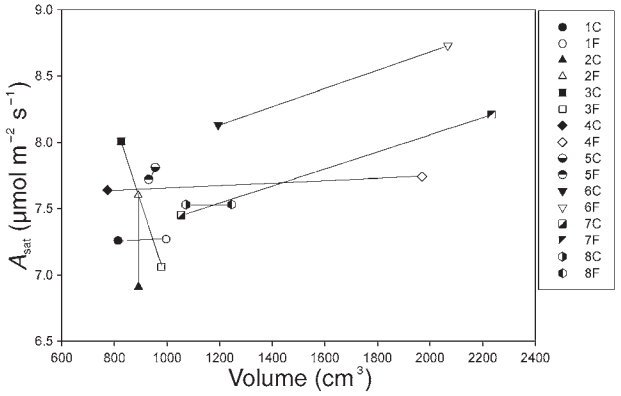

Figure 4. Mean (n = 68) clonal light-saturated photosynthetic rates (A_{sat}) during the growing season as a function of the end of year mean clonal volume. Abbreviations: C denotes control; and F denotes fertilized.

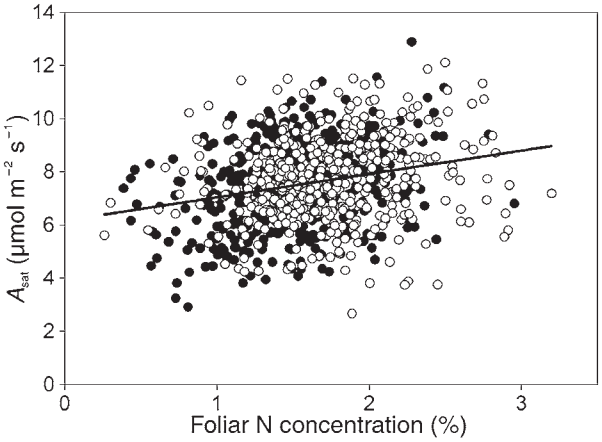

Figure 6. Relationship between light-saturated photosynthetic rate (A_{sat}) and foliar nitrogen (N) concentration for fertilized (○) and unfertilized (●) seedlings for most of the growing season (May to September 2004).

fertilizer responses than Clones 2, 3, 5 and 8, which had little or no response over the growing season. The fertilization × date interaction was the result of a steady increase in CSA in fertilized seedlings over the growing season, being 30.5% higher at the end of the growing season than in control seedlings.

Time series analysis showed a significant clonal effect (P = 0.032) and a fertilizer × date interaction (P = 0.022) on mean crown efficiency (Table 2). When the clonal effects were examined at the end of the year, a Duncans MRT test (α = 0.1) showed that Clones 6 and 7 were significantly more efficient than Clones 8 and 1 (Figure 9). Clones 7 and 8, which share the same parents, differed in crown efficiency by 21%. There was a fertilizer × date interaction on crown efficiency because the fertilized seedlings had a higher crown efficiency by the end of the growing season than the unfertilized seedlings.

Discussion

Many studies have shown that fertilization increases growth or photosynthesis or both in loblolly pines (Teskey et al. 1994, Murthy et al. 1996, 1997, Samuelson 2000, Lavigne et al. 2001, Munger et al. 2003, Gough et al. 2004*b*) by increasing leaf areas (Vose and Allen 1988). However, the physiological mechanisms behind this initial increase in leaf area remain unresolved. Our data suggest that there is more than one mechanism whereby loblolly pine achieves increased volume growth after fertilization. For example, the data for Clones 6 and 7 support the findings of Gough et al. (2004*a*) that fertilization quickly and temporarily increases photosynthesis, leading to a buildup of photoassimilate used to create larger leaf areas and ultimately more growth. Among the clones we tested, Clones 6 and 7 showed the largest increases in A_{sat} and volume growth in response to fertilization. Furthermore, these clones tended to have the highest A_{sat} during the growing season, which most likely led to the differences seen in CSA between the fertilized and unfertilized clones. Clones 6 and 7 also had the highest crown efficiency ratings, and they were significantly higher

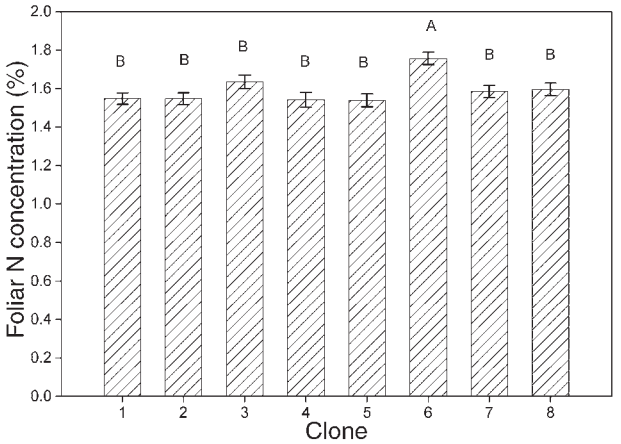

Figure 5. Mean clonal foliar nitrogen (N) concentration over the sampling period. Different letters denote significant differences (P < 0.05) among clones. Error bars represent standard error (n = 128).

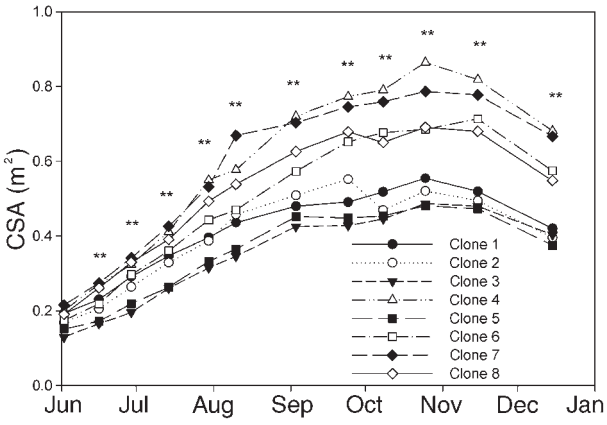

Figure 7. Mean (n = 8) clonal crown silhouette area (CSA) from June to December 2004. Two asterisks (**) denote significant differences (P < 0.05) among clones.

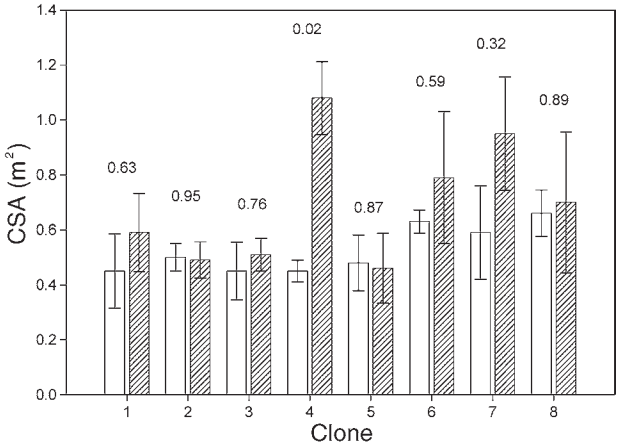

Figure 8. Mean clonal crown silhouette area (CSA) for fertilized (hatched bars) and unfertilized (open bars) seedlings at the end of the year (November 15, 2004). Error bars represent standard error ($n = 4$).

than those of Clones 1 and 8. This result was unexpected because Clones 7 and 8 share the same parents, but lends credence to past studies showing genetic inheritance instability among families (Li et al. 1991, McCrady and Jokela 1996, Roberts et al. 2003, Chang 2003) and genotype × environment interactions (Roth et al. 2007).

The other clones included in our study showed a range of growth responses for which the physiological basis was often unclear. Clones 1, 4 and 8 showed a volume growth response to N fertilization but with almost no response in A_{sat}. Clone 4 showed an extremely large (> 1000 cm^3) volume growth increase after fertilization, whereas its net photosynthetic rate increased only moderately and crown efficiency was the third lowest of the clones tested. Clone 4 was the only clone to show significant difference in CSA between fertilized and unfertilized trees, and it also had the highest CSA value at the end of the growing season relative to the other clones. The large effect of fertilization on volume growth in Clone 4 could therefore be attributed to the large increase in CSA in the fertilized seedlings because A_{sat} increased only slightly. A similar pattern was seen in Clones 1 and 8, although the differences were more subtle. Fertilization increased volume growth in Clones 1 and 8; however, these clones had significantly lower crown efficiencies than the two top-performing clones, and showed only relatively moderate increases in A_{sat} and CSA in response to fertilization. Changes in allocation patterns, although not measured in our study, need to be investigated because they likely contributed to the increase in CSA found in these fertilized clones.

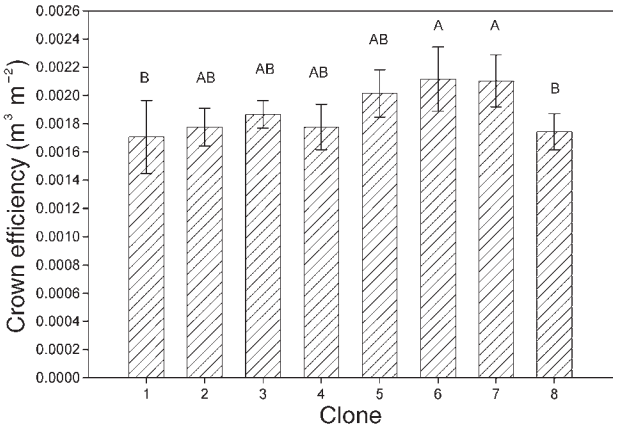

Figure 9. Mean clonal crown efficiencies at the end of the year (November 15, 2004). Different letters denote significant differences ($P < 0.1$) among clones. Error bars represent standard error ($n = 8$).

Increased allocation to aboveground components in loblolly pine after fertilization has been found in several studies. At the nutrient-poor Southeast Tree Research and Education Site (SETRES) in the North Carolina sandhills region, several joint studies examined the biomass production of loblolly pine given nutrient and irrigation treatments over a 4-year period. Albaugh et al. (1998) quantified the biomass partitioning patterns and growth efficiency of the loblolly pines. King et al. (1999) determined stand-level allometric relationships among plant parts. All treatments increased the biomass of stems, branches, foliage, taproots, and coarse roots. The only plant part that showed no increases in accumulated biomass was fine roots. Albaugh et al. (1998) noted that biomass partitioning may be responsible for at least some of the increases in growth efficiency during the study. The other growth efficiency increases seen may have been a result of increased allocation to foliage and decreased allocation to fine roots (high maintenance respiration tissue). King et al. (1999) found that fertilization increased partitioning to coarse roots, branches, and taproots, but decreased partitioning to foliage and fine roots. They concluded that biomass partitioning changes with varying resource availability and is under ontogenic control. Differences in biomass partitioning is one possible explanation for the varying growth responses to fertilization that we observed among clones.

Although not measured as intensively, our study did not demonstrate such a clear biomass growth response to fertilization as the SETRES study. Clone 3 showed a slight positive volume growth response to fertilization but a negative A_{sat} response. This clone exhibited a somewhat higher CSA in fertilized trees than in unfertilized control trees, which may partially explain the increase in volume growth, given that Teskey et al. (1987) noted that there can be a negative relationship between carbon gain and A_{sat} as a result of increases in leaf shading on trees with larger leaf areas. Clone 5 also had a negative A_{sat} response to fertilization, but differed from Clone 3 in having a negative volume growth response as well. Clone 2 showed a slight A_{sat} response to fertilization, but no volume growth response. It is difficult to determine a clear physiological reason for these responses based on the data we collected. We speculate that the high fertility of our soil (Tyree 2005) contributed to the variable clonal responses to fertilization. On a more nitrogen-limited site, perhaps all clones would have responded positively. Our data may indicate that critical foliar N concentrations differ among clones.

There is an increasing likelihood that clones or selected full-sib families will have genotype × environment (G × E) in-

teractions in the future because more forest areas are being intensively managed (Roth et al 2007). Although we did not obtain data across different sites or species, we found clonal variation in response to an intensive silvicultural treatment. Roth et al. (2007) reported significant genotype × location, genotype × silvicultural treatment intensity, and silvicultural treatment intensity × density interactions for basal area (BA) and stem volume in 5-year-old loblolly and slash pine clones. Studies with other species have shown similar clonal variation in response to treatment, location, or treatment intensity (Orlovic et al. 1998, Weih 2001, Chang 2003, Karacic and Weih 2006). For example, Orlovic et al. (1998) found significant genotype × environment interactions and interclonal variation in physiological (P_n and R_d) and growth characteristics (leaf area, height, and diameter) among eight black poplar clones (*Populus deltoides* Bartr. and *Populus* × *euramericana* (Dode) Guinier). Weih (2001) studied several willow clones (*Salix* spp.) subjected to multiple fertilization, irrigation and temperature regimes and observed clone × treatment interactions for some growth parameters and significant differences between clones in leaf area and water-use efficiency. The hybrid willow clone produced 25% more shoot biomass than the natural clone when supplied with high rates of fertilizer and irrigation (Weih 2001). In a sweetgum (*Liquidambar styraciflua* L.) half-sib clonal study, Chang (2003) found significant genotypic effects on leaf area and basal diameter growth, although the diameter growth response could be explained by the differential increase in leaf area expansion between families. Karacic and Weih (2006) found significant clone × treatment interactions on growth parameters of eight poplar clones (*Populus balsamifera* L., *P. trichocarpa* Hook. and *P. trichocarpa* × *P. deltoids*) under different irrigation and fertilization regimes in Sweden. They found relative growth rate and leaf N varied among clones, irrigation and fertilization, although biomass production only responded to clones and irrigation.

In conclusion, early and intensive sampling of loblolly pine revealed that clones differed dramatically in their photosynthetic and growth responses to fertilization. Nighttime needle respiration was unrelated to any fertilization or clonal differences. In response to fertilization, some clones increased photosynthetic rates and growth, whereas others showed only slight increases in photosynthesis but relatively large increases in growth. These large genetic × environment interactions—which even occurred between clones that shared the same parents—suggest that clones may respond quite differently to varying management strategies and inherent soil fertility. The forest industry will need to understand and identify such interactions to ensure the efficient use of relatively expensive clonal material.

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