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COMPETITION FOR APPLIED ¹⁵N FERTILIZER IN A LONGLEAF PINE/NATIVE WOODY ORNAMENTAL INTERCROPPING SYSTEM

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Abstract: The cultivation of ornamentals to produce woody floral products – the fresh or dried stems that are used for decorative purposes – may be an attractive option for southeastern landowners looking to generate income from small landholdings. Since many shrubs native to the understory of the longleaf pine (*Pinus palustris* Mill.) ecosystem have market potential, one possibility is the intercropping of select species in the between-row spacing of young longleaf pine plantations. The objective of this study was to evaluate how interspecific competition affects the fate of ¹⁵N fertilizer when American beautyberry (*Callicarpa americana* L.), wax myrtle (*Morella cerifera* (L.) Small) and inkberry (*Ilex glabra* (L.) A.Gray) are intercropped with longleaf pine. Nitrogen derived from fertilizer (NDF), utilization of fertilizer N (UFN) and recovery of fertilizer N (RFN_{soil}) were compared between agroforestry and monoculture (treeless) treatments to assess the effect of competition. Results varied by species, with NDF being higher for *C. americana* foliage and lower for all *M. cerifera* tissues in the agroforestry treatment. No effect was observed for *I. glabra*. UFN was lower for all species in the agroforestry treatment. RFN_{soil} was higher in the agroforestry treatment for *I. glabra*, but no treatment effects were observed for *C. americana* or *M. cerifera*. Overall, while it is clear that interspecific competition was present in the agroforestry treatment, the inefficiency of fertilizer use suggests that nitrogen was not the most limiting resource. Management interventions, particularly those that address competition for water, will likely be critical to the success of this system.

Keywords: ¹⁵N recovery, *Callicarpa americana*, competition, fertilizer use efficiency, *Ilex glabra*, *Morella cerifera*, *Pinus palustris*, woody florals

INTRODUCTION

The longleaf pine (*Pinus palustris* Mill.) ecosystem of the southeastern U.S.A. is one of the most threatened ecosystems in North America (Barnett 1999). Considerable attempts have been made in the recent past in increasing the forest cover under longleaf pine (Jose et al. 2006). For example, approximately 100,000 ha of longleaf pine plantations were established under the Conservation Reserve Program (CRP) in the Southeast during

1999-2000 (Hains et al. 2000). The diversity of groundlayer vegetation is what makes the longleaf pine ecosystem one of the most species-rich plant communities outside of the tropics (Walker and Silletti 2006). Interestingly, many of these species have potential as woody floral products – the fresh or dried stems used for decorative purposes such as wreaths and flower arrangements (Stamps et al. 1998; Venrick 2003; Josiah et al. 2004). While concerns over sustainability would likely rule out the wild harvest of stem material, the intercropping of select species in the between-row spacing of a longleaf pine plantation could be an attractive option for some landowners. Such a system would not only generate income, it would also provide an incentive to reintroduce native understory species, thereby enhancing the biodiversity of plantation forests (Hartley 2002).

By optimizing the use of resources in space and time, a well-designed intercropping system can be highly productive as well as ecologically and economically sustainable. Achieving this balance, however, is dependent upon the understanding, and subsequent management, of the interspecific interactions that affect the productivity of its component species. Of particular importance is minimizing competition, the interaction that occurs when both species simultaneously seek the same limiting resource. Competition in intercropping systems commonly occurs aboveground in the form of shading, or belowground in the form of overlapping zones of resource depletion in the rhizosphere (Schroth 1999; Jose et al. 2004).

The open canopy and high light transmittance that is characteristic of longleaf pine suggests that properly selected shrubs, when intercropped in the alleys between tree rows, would be minimally affected by shading (Battaglia et al. 2003; Hagan et al. in press). The extensive lateral root system of this species, however, creates the potential for interspecific competition belowground (Hagan et al. in press). Competition for nitrogen, typically the most limiting macronutrient in temperate cropping systems (Jose et al. 2004), could be intense under these conditions. This competition could be ameliorated or avoided, to some extent, through fertilization and by selecting species with patterns of nutrient uptake that differ, spatially or temporally, from longleaf pine. Ideally, such complementarity between root systems would create a “safety net” effect, in which the deeply rooted trees capture nutrients which leach beyond the shallow rooting zone of the shrubs (van Noordwijk et al. 1996; Allen et al. 2004b; Jose et al. 2006; Zamora et al. 2008). Increasing the fertilizer use efficiency of a system in this manner effectively reduces the amount of nitrogen that leaches down into groundwater – a common problem with ornamental production systems (Ristvey et al. 2004), which typically require higher fertilization rates than do conventional agronomic crops.

In this study, the nitrogen dynamics in a longleaf pine - native woody ornamental intercropping system were examined using ^{15}N -labeled ammonium sulfate ($(\text{NH}_4)_2\text{SO}_4$) fertilizer. Despite limited use in temperate intercropping applications, ^{15}N labeling techniques have proven to be effective means of tracing the movement of nitrogen in the tree-crop-soil system (Jose et al. 2000; Allen et al. 2004a; Allen et al. 2004b; Zamora et al. 2008). Knowledge of how fertilizer is cycled, in turn, can be used to determine what, if any, management interventions must be implemented to improve both crop yield and ecological sustainability.

The specific objective of this study was to examine how competition from longleaf pine would affect fertilizer uptake and use efficiency by three common native shrub species: American beautyberry (*Callicarpa americana* L.) (Verbenaceae), wax myrtle (*Morella cerifera* (L.) Small) (Myricaceae), and inkberry (*Ilex glabra* (L.) A.Gray) (Aquifoliaceae). We hypothesized that interspecific competition would force the shrubs to derive a greater percentage of their nitrogen from fertilizer and would leave less fertilizer remaining in the soil at the end of the growing season. Reduced fertilizer uptake or use efficiency by the shrubs (if observed) would likely be due to differences in biomass brought about by other resource limitations.

MATERIALS AND METHODS

Study Site and Experimental Design

This study was conducted on a private 15-year-old-longleaf pine plantation in Santa Rosa County, Florida, USA (30°37' N, 87°2' W). The climate of the region is classified as temperate, with mild winters and hot, humid summers. Mean annual precipitation is 1645 mm. The soil is an ultisol and classified as a Fuquay sand (loamy, kaolinitic, thermic Arenic Plinthic Kandiudult), a deep, well-drained sand over loamy marine or fluviomarine deposits.

Trees in the study site were uniformly spaced, with approximately 3 meters between rows and 1.5 meters between stems within the row. Mean diameter at breast height (DBH) at the initiation of the study (December 2005) was 8.3 cm. Mean basal area was 14 m² ha⁻¹. In December 2005, containerized native woody ornamental shrubs identified as having market potential were incorporated into the existing between-row spacing of the site, and as an equivalently spaced monoculture treatment in an adjacent open field. Selected species were American beautyberry (*Callicarpa americana*), wax myrtle (*Morella cerifera*) and inkberry (*Ilex glabra*). Shrubs were given a year for proper establishment with dead or dying shrubs being replaced in the winter of 2006, prior to the initiation of the study.

The effect of intercropping on the nitrogen dynamics of this system was assessed via comparisons with the monoculture treatment. The trial was laid out as a split-plot completely randomized design with treatment (monoculture or intercropped) as the whole plot factor and shrub species as the split-plot factor. There were four replications, each consisting of six subplots (one for each species by treatment combination) with eight shrubs each. Subplots were 2 alleys wide (or equivalent distance in the monoculture) with shrubs planted in two rows of four at a spacing of 3 meters. As a control, four subplots of the same dimensions were established in the plantation and not planted with shrubs.

Fertilizer Application and Plot Maintenance

To assess competition for nitrogen between component species, ¹⁵N ammonium sulfate ((NH₄)₂SO₄) at 5% atom enrichment was applied to two shrubs within each subplot. To

simulate the effect of a slow-release fertilizer, three applications at $146.5 \text{ kg N ha}^{-1}$ were applied at approximately 60 day intervals in a circular area of 325 cm^2 at the base of each shrub. The six remaining shrubs in each plot received non-enriched $(\text{NH}_4)_2\text{SO}_4$ at the same application rate. The first application was on 21 March 2007, shortly after bud swelling and new leaf development were first observed. Pesticide and herbicide application, along with manual weed removal, were conducted as needed throughout the growing season. Plots were non-irrigated, but supplemental water was uniformly provided to all shrubs when at least 20% showed signs of extreme drought stress. This occurred on 3 occasions during a particularly dry period from mid May to early June 2007.

Harvest and Sampling

At the end of the growing season, 90 cm soil cores were taken using a manual soil auger at the site of each plant that received labeled fertilizer. Cores were subdivided into 30 cm segments and subsamples of approximately 5g were taken. Additionally, each plant that received ^{15}N fertilizer was harvested and separated into leaf, stem, root and (when applicable) fruit components. *C. americana* was harvested in mid September, prior to leaf senescence while *I. glabra* and *M. cerifera* were harvested with the onset of cool weather (end of the growing season) in late October. Also following the growing season, pine foliar samples were harvested with a telescoping pruning saw from the four trees (two on each row) closest to the site of ^{15}N application. For this, the canopy was visually divided into upper and lower halves and samples collected from the four cardinal directions in each half (8 total samples/tree). Needles from the four trees were composited into a single sample.

All plant material was dried to constant weight at 70°C , weighed, subsampled and ground with a coffee grinder to a fine ($< 1 \text{ mm}$) particle size. The grinder was thoroughly cleaned and dried between samples to prevent cross-contamination. Soil subsamples were ground with a mortar and pestle until they reached a flour-like consistency.

All tissue and soil samples were analyzed by the Stable Isotope Facility at the University of California Davis (Davis, California, USA). Analyses were conducted using a PDZ Europa ANCA-GSL elemental analyzer interfaced to a PDZ Europa 20-20 isotope ratio mass spectrometer (Sercon Ltd., Cheshire, UK). The results of these analyses were then used to calculate percent plant nitrogen derived from fertilizer (NDF), percent utilization of fertilizer nitrogen (UFN) and percent recovery of fertilizer nitrogen in soil (RFN_{soil}).

Percent plant nitrogen derived from fertilizer (NDF), a measure of the amount of fertilizer that a plant obtains from labeled fertilizer was calculated using the following formula (Allen et al. 2004a):

$$\text{NDF (\%)} = 100 * (a-b)/(c-d),$$
 where $a = \%^{15}\text{N}$ abundance in plant tissue;
 $b =$ percent abundance in control (unlabeled) plant tissue;
 $c = \%^{15}\text{N}$ abundance of fertilizer (5%); and
 $d =$ natural abundance of ^{15}N (0.3663%).

Percent utilization of fertilizer nitrogen (UFN), a measure of fertilizer use efficiency, was calculated using the following formula (Allen et al. 2004a):

$$\text{UFN (\%)} = (\% \text{NDF} * S)/R,$$
 Where %NDF = the percentage of plant nitrogen derived from fertilizer;
 $S =$ the amount of nitrogen (g) in plant tissue; and
 $R =$ the amount of nitrogen (g) applied to each plant.

Percent recovery of fertilizer ^{15}N in soil (RFNsoil), a measure of ^{15}N fertilizer remaining in soil at the end of the growing season was determined at each depth using the following formula (Allen et al. 2004a):

$$\text{RFNsoil (\%)} = 100 * ((a-c)/(b-c)) * (Np/Nf)$$
 Where $a = \%^{15}\text{N}$ abundance in soil that received ^{15}N fertilizer;
 $b = \%^{15}\text{N}$ abundance in fertilizer (5%);
 $c =$ background ^{15}N abundance in unfertilized soil;
 $Np =$ total N of soil sample (g); and
 $Nf =$ total amount (g) of ^{15}N applied to soil as fertilizer.

Data Analysis

Data were analyzed using the PROC GLM procedure in SAS 9.1 (SAS Institute, Cary, NC, USA) within the framework for a split-plot completely randomized experimental design. Between-species comparisons were conducted for all isotopic analyses to determine which species were most effective at capturing and utilizing applied fertilizer. Logarithmic or arcsin transformations were performed when necessary to improve data normality. Tukey's HSD or Dunnett's post hoc tests were used for pairwise comparisons and differences between means were declared significant at $\alpha < 0.05$.

RESULTS

Biomass

Biomass production was significantly lower in the intercropping treatment compared to the monoculture (Table 1). Total biomass in the intercropping treatment was lower by 76.9%, ($P = 0.0030$), 53.8% ($P = 0.0200$) and 67.4% ($P = 0.0012$) for *C. americana*, *M. cerifera* and *I. glabra*, respectively. For foliage, reductions (in the above order) were 83.0% ($P = 0.0002$), 56.9% ($P = 0.0331$) and 76.0% ($P = 0.0030$). Stem biomass was reduced by 72.3% ($P = 0.0065$) for *C. americana* and 75.2% ($P = 0.0023$) for *I. glabra*. The 59.1% reduction observed for *M. cerifera* was not statistically significant. Root

biomass was 67.2% lower for *C. americana* ($P = 0.0052$) and 60.7% lower for *I. glabra* ($P = 0.0098$) in the intercropping treatment. The 43.2% reduction observed for *M. cerifera* was not statistically significant. Comparisons of fruit biomass for *C. americana* and *I. glabra* were not possible as only one individual from each species produced fruit in the intercropping treatment.

Table 1. Leaf, stem, root and total biomass for *C. americana*, *M. cerifera* and *I. glabra* in a longleaf pine-native shrub intercropping system in Florida, USA. Means and standard errors.

Species	Treatment	Biomass (g/plant)				
		Foliage	Stems	Roots	Fruits	Total
<i>C. americana</i>	Monoculture	52.9±9.0	43.2±10.8	87.8±11.6	20.0±5.1	206.3±29.8
<i>C. americana</i>	Intercropped	8.7±11.1 (0.0002) ¹	12.0±14.5 (0.0030)	28.8±15.8 (0.0052)	0.2	50.9±37.4 (0.0030)
<i>M. cerifera</i>	Monoculture	30.6±9.7	68.6±11.6	82.9±11.6		182.0±32.3
<i>M. cerifera</i>	Intercropped	13.2±8.3 (0.0331)	29.6±10.8 (0.0520)	47.0±11.7 (0.0768)		89.8±27.5 (0.0200)
<i>I. glabra</i>	Monoculture	42.0±8.3	86.0±11.6	92.1±12.6	4.2±5.6	220.6±29.8
<i>I. glabra</i>	Intercropped	9.7±8.2 (0.0030)	21.3±11.7 (0.0145)	36.2±11.6 (0.0052)	0.5	68.9±29.9 (0.0012)

¹ P -values given in parentheses.

Tissue Nitrogen Concentrations and Content

Comparisons of nitrogen concentration showed no treatment differences, for any tissue, for either *M. cerifera* or *I. glabra*. A significant treatment effect ($P = 0.0015$), however, was observed for *C. americana* roots, which had 1.01% nitrogen in the intercropping treatment compared to 2.55% in the monoculture (Figure 1).

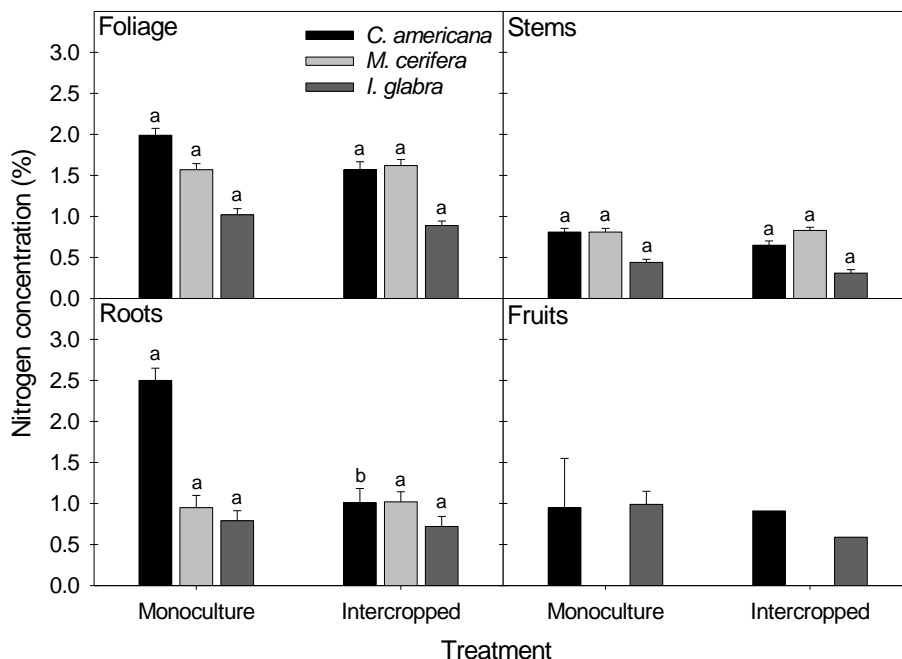


Figure 1. Mean nitrogen concentration (by tissue) for *C. americana*, *M. cerifera* and *I. glabra*. Between-treatment means (by species) with different letters are statistically different at $\alpha < 0.05$.

For *C. americana*, tissue nitrogen content was lower in the intercropping treatment for leaves ($P = 0.0049$), stems ($P = 0.0183$) and roots ($P = 0.0173$). A similar pattern was observed for *I. glabra* ($P = 0.0155$, $P = 0.0014$ and $P = 0.0318$, respectively). Nitrogen content for *M. cerifera* was lower in the intercropping treatment for leaves ($P = 0.0148$) and stems ($P = 0.0036$), but no statistically significant difference was observed for roots (Figure 2).

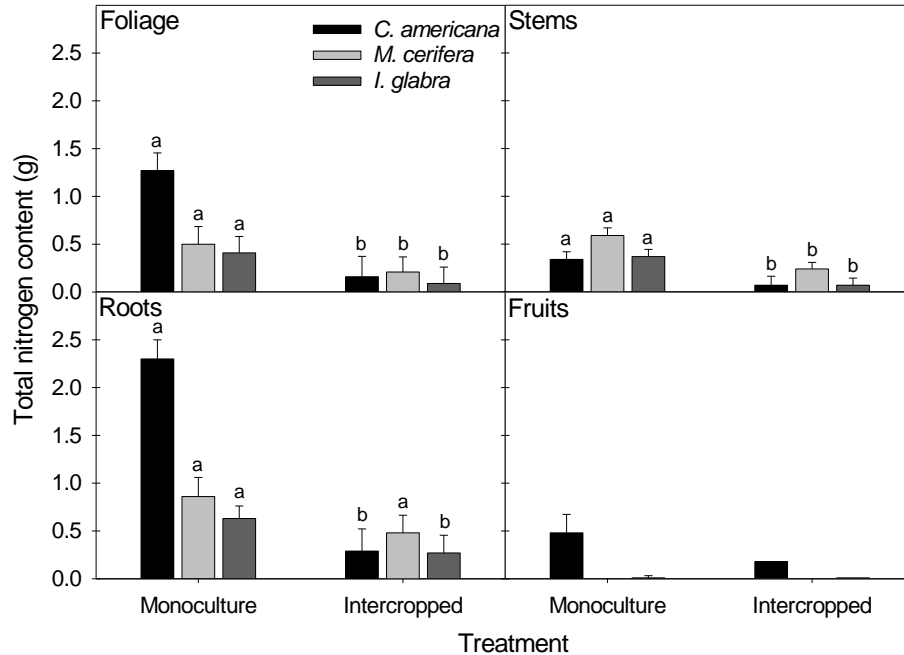


Figure 2. Total nitrogen content (by tissue) for *C. americana*, *M. cerifera* and *I. glabra*. Between-treatment means (by species) with different letters are statistically different at $\alpha < 0.05$.

Foliar nitrogen derived from fertilizer (NDF) values (by species) were 50.8% higher for *C. americana* ($P = 0.0482$) and 66.7% lower for *M. cerifera* ($P = 0.0294$) in the intercropping treatment compared to the monoculture. A similar pattern was observed for *M. cerifera* stems ($P = 0.0494$) and roots ($P = 0.0190$), which had significantly lower NDF values in the intercropping treatment (63.7% and 64.4%, respectively). No significant treatment differences were observed for *I. glabra* for any tissue. Within treatments, *M. cerifera* was the only species that was significantly different, with NDF values for all tissues being significantly lower than *C. americana* and *I. glabra* in the intercropping treatment (Figure 3).

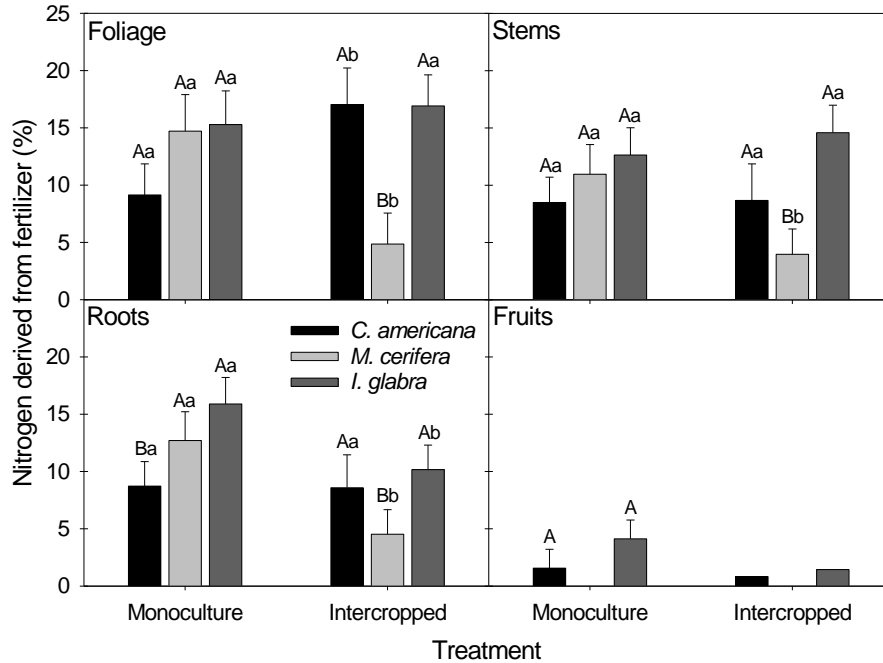


Figure 3. Percent nitrogen derived from fertilizer (NDF) (by tissue) for *C. americana*, *M. cerifera* and *I. glabra*. Within-treatment means with different uppercase letters are statistically different at $\alpha < 0.05$. Between-treatment means (by species) with different lowercase letters are statistically different at $\alpha < 0.05$.

Total utilization of fertilizer nitrogen (UFN) (all tissues combined) was significantly lower in the intercropping treatment ($P < 0.0001$), with species-wise reductions of 82.2%, 81.7% and 78.3% for *C. americana*, *M. cerifera* and *I. glabra*, respectively. Significant reductions in UFN were observed for all species, and were 77.8%, 71.4% and 83.1% for *C. americana*, 85.7%, 81.7% and 81.9% for *M. cerifera*, and 76.9%, 79.0% and 76.4% for *I. glabra*, for foliage, stems and roots, respectively. Within treatments, no significant differences in UFN were observed between species (Figure 4).

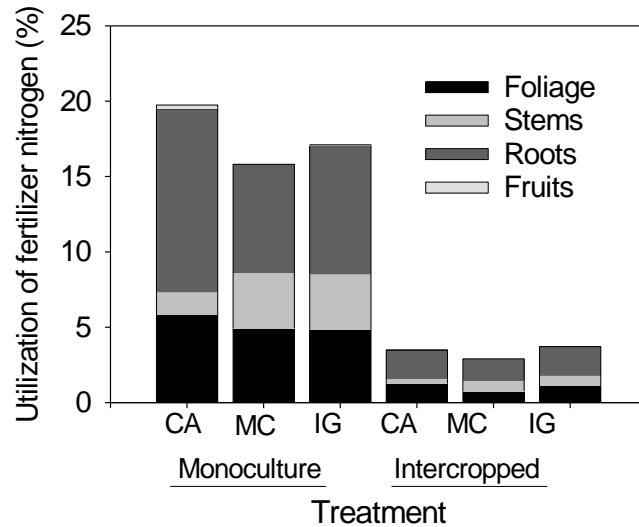


Figure 4. Percent utilization of fertilizer nitrogen (UFN) (by tissue) for *C. americana* (CA), *M. cerifera* (MC) and *I. glabra* (IG).

Recovery of Fertilizer N in Soil

RFN_{soil} varied by species ($P = 0.0007$) and depth ($P < 0.0001$) and there was an interaction between treatment and species ($P = 0.0001$). For *C. americana*, no differences between treatments were observed at any depth. For *M. cerifera*, no treatment differences were observed at 30 or 90 cm, but RFN_{soil} was higher in the intercropping treatment at the 60 cm depth. RFN_{soil} was significantly higher, at all depths, for *I. glabra* in the intercropping treatment, with differences of 61.9% at 30 cm and 79.1% at 60 cm. The magnitude of the difference at 90 cm (while significant) could not be determined, as negative RFN_{soil} values were observed in the monoculture – indicating a lower level of ¹⁵N enrichment than background soil. Between species, no significant differences in RFN_{soil} were observed, at any depth, in the intercropping treatment (Figure 5).

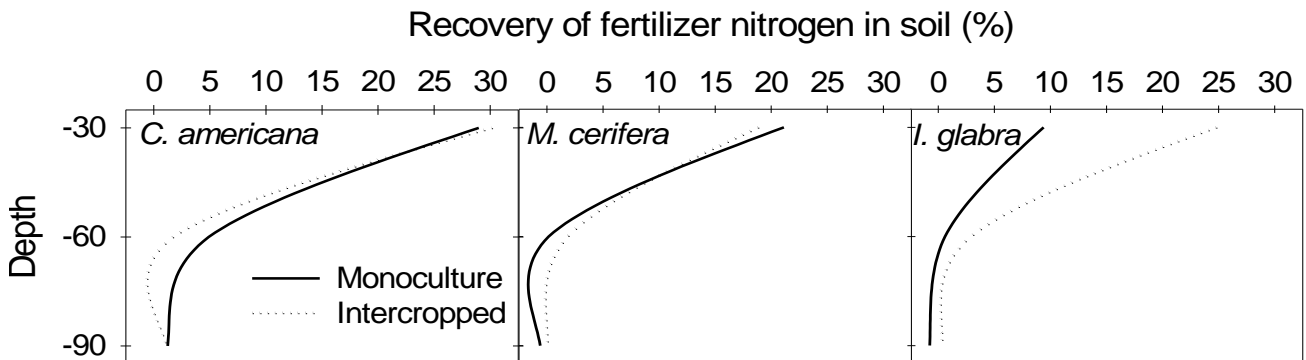


Figure 5. Recovery of fertilizer nitrogen in soil (RFNsoil) at three depths (30, 60 and 90 cm) for *C. americana*, *M. cerifera* and *I. glabra*.

In terms of total RFN_{soil} (RFN summed across the three depths) there was a significant species effect ($P = 0.0265$) and an interaction between treatment and species ($P = 0.0290$). The 33.2% RFN_{soil} observed for *C. americana*, was 41.2% higher than that observed for *I. glabra* and 37.7% higher than *M. cerifera*. *I. glabra* was the only species for which a significant treatment difference was observed, having 3.2 times more fertilizer remaining in the intercropping treatment than in the monoculture. Overall, mean RFN_{soil} in the intercropping treatment was 31.3% for *C. americana*, 20.6% for *M. cerifera* and for 29.7% *I. glabra*. Differences between species were not statistically significant.

Fertilizer N and Total N in Pine Foliage

Mean nitrogen concentration for pine foliage was 1.58%, 0.27% of which was derived from fertilizer. Extrapolating to canopy level as described by Baldwin and Saucier (1983) revealed that 20.9% of applied fertilizer N was in pine foliage at the end of the growing season. There was no effect of shrub species on nitrogen concentration or NDF in pine foliage.

DISCUSSION

Biomass production and allocation patterns observed in our study suggest that competition for resources was an important determinant of productivity for all three shrub species in the intercropping system. All species in the intercropping treatment produced less biomass and exhibited reduced carbon allocation to aboveground tissues and increased allocation to roots, a pattern which suggests that the limiting resources in this system were belowground (Chapin et al. 2002).

Reductions in biomass due to belowground interspecific competition are common in temperate intercropping systems, particularly those involving crops intercropped with large trees with shallow lateral root systems. In a pecan (*Carya illinoensis* K. Koch)-cotton (*Gossypium hirsutum* L.) alleycropping system in NW Florida, a 58% reduction in aboveground cotton biomass (compared to a root barrier treatment) was attributed to competition for nitrogen and water (Allen et al. 2004b; Wanvestraut et al. 2004). Similar effects were observed for maize (*Zea mays* L.) when intercropped with black walnut (*Juglans nigra* L.) and red oak (*Quercus rubra* L.) (Jose et al. 2000). While longleaf pine is known for its deep tap root, it too has an extensive lateral root system (Brockaway and Outcalt 1998). In a companion study conducted in the same plantation, Hagan et al. (in press) found that 81.1% of longleaf pine fine roots were in the uppermost 30 cm of soil – the same horizons exploited by the root systems of the intercropped shrubs. Interspecific competition for water was apparent in the companion study, which had the most pronounced effects on *M. cerifera* and *I. glabra* productivity.

Treatment effects on tissue nitrogen concentrations were observed only in *C. americana* roots – a fact that suggests that this species was more adversely affected by interspecific competition than the other two. Nitrogen stored in roots at the end of the growing season serves as an important reserve for spring tissue development (Chapin et al. 1990),

especially in deciduous species (Lamaze et al. 2003). Deciduous *C. americana* likely has a high early-season nitrogen requirement which, due to this decrease in storage, may have forced them to tap soil N sources, thus increasing the likelihood of competition with longleaf pine. While differences in nitrogen concentration between treatments were minimal, total nitrogen content was lower for all species, and most tissues, in the intercropping treatment. The only exception was *M. cerifera* roots, for which there was no treatment effect for either biomass or total nitrogen content. The observed treatment differences were primarily due to the reductions in total biomass.

It is generally believed (and was the basis of our hypothesis) that competition, by depleting native soil nitrogen levels, encourages intercropped plants to derive a greater proportion of their nitrogen from fertilizer sources. Substantial increases in NDF in this manner were observed in the above-mentioned intercropping systems (Jose et al. 2000; Allen et al. 2004b; Zamora et al. 2008). *C. americana*, however, was the only species in this system which supported our hypothesis, deriving 50.8% more of its foliar nitrogen from fertilizer in the intercropping treatment than in the monoculture. Since leaf development for this species was complete by May 15, it is possible that the most intense interspecific competition occurred in the weeks immediately following the first fertilizer application (March 21), thereby resulting in increased fertilizer utilization. The reasons why this pattern did not hold true for *C. americana* stems and roots, which had no treatment differences, is unknown.

The fact that no treatment differences were observed for any *I. glabra* tissue suggests that interspecific competition for nitrogen was less severe for this species. Perhaps this was the result of spatial or temporal complementarity between shrubs and trees or evidence of greater competitive ability (Schaller et al. 2003). The large decrease in NDF (despite no differences in N concentration) observed for all *M. cerifera* tissues in the intercropping treatment could be evidence of another trend. As a nitrogen-fixing actinorhizal shrub (Young 1992), *M. cerifera* derives a percentage of its nitrogen from the atmosphere, which at 0.3663% atom enrichment has a “dilution” effect on tissue ¹⁵N concentrations (Busse 2000; Robinson et al. 2001). Perhaps interspecific competition led this species to derive a greater percentage of its nitrogen from atmospheric sources instead of soil – resulting in reduced NDF. This is an intriguing possibility that deserves further study.

Differences in UFN for all species were likely functions of biomass differences, with plants in the intercropping treatment generally having lower values due to reduced growth, possibly caused by competition for water (Wanvestraut et al. 2004). As we hypothesized, these reductions in growth likely inhibited the shrubs’ ability to take up fertilizer thus further magnifying treatment differences for UFN (Allen et al. 2004b).

The thick E horizon characteristic of the Fuquay soil series has a very low cation exchange capacity and thus very little ability to retain applied fertilizer. Soil cores from all species/treatment combinations confirmed this, illustrating a pattern of decreasing fertilizer RFN_{soil} concentrations with depth. The combined effects of competition and spatiotemporal differences in nutrient uptake between species typically result in lower RFN_{soil} values in intercropping systems (Allen et al. 2004a; Allen et al. 2004b), which

was the foundation of our hypothesis. This pattern, however, was not observed. In this system, no treatment differences in total RFN_{soil} were observed for *C. americana* or *M. cerifera* and the opposite (lower RFN_{soil} in monoculture) was observed for *I. glabra*. It is possible that the decreased uptake (lower UFN) observed for shrubs in the intercropping treatment was offset by uptake by longleaf pine. Unfortunately this scenario, while plausible for *C. americana* and *M. cerifera*, does not adequately explain the differences observed for *I. glabra*. Perhaps this can be attributed to ecophysiological differences between species. In the aforementioned companion study, Hagan et al. (in press) found *I. glabra* to be the only species that was adversely affected, in terms of carbon assimilation, by both shading and competition for water. This combined with the period of late season growth (August – September) observed for this species in the monoculture, but not the intercropping treatment (Hagan et al. in press), may have contributed to the large disparity in total RFN_{soil} between treatments.

In the intercropping treatment, 83.9% more fertilizer ended up in pine foliage than in the shrubs themselves. This, however, is a conservative estimate of total fertilizer uptake, given the likelihood of storage in pine roots, stems and branches, which were not sampled. On one hand, fertilizer uptake by the pines represents nitrogen that was not lost to leaching, and therefore may be considered evidence of a “safety-net” effect – one of the most commonly touted benefits of intercropping (van Noordwijk et al. 1996; Allen et al. 2004a; Jose et al. 2006). However, given the shallow fine root distribution of the longleaf pines, it is likely that much of the fertilizer was obtained via interspecific competition, not by spatial complementarities between root systems. Any benefit that the trees received from this secondary fertilization, therefore, could potentially have been at the expense of shrub productivity. There was no evidence, based on RFN_{soil} data, of significant fertilizer uptake by trees in the deeper soil depths, although it is possible that some uptake occurred at depths greater than 90 cm.

It is clear that the effective management of belowground processes is essential for the viability of a longleaf pine/native woody ornamental intercropping system. Ideally this could be done in a manner that minimizes the deleterious effects of competition while retaining, to the greatest extent possible, the environmental benefits of intercropping. Of particular importance is the need to improve our understanding of how the nutrient requirements of selected species differ in space and time. This knowledge would not only aid in species selection, but also with nutrient management, as fertilizer applications could be better synchronized with demand, thus decreasing competition, increasing fertilizer use efficiency and decreasing loss. In this system, for example, only 24.3% of applied fertilizer could be accounted for in at the end of the growing season (3.4% in shrubs, 20.9% in trees). The remaining 75.7% either remained in soil or was lost, most likely due to leaching. It deserves reiteration, however, that the most commonly reported signs of competition for nitrogen (increased NDF and reduced RFN_{soil}) were, for the most part, not observed. This, combined with the low utilization of fertilizer nitrogen suggests that nitrogen was likely not the main determinant of productivity in this system. Future research should seek to further elucidate the effect of interspecific competition for water and other belowground resources on shrub growth.

CONCLUSIONS

Results indicate that competition with longleaf pine had a deleterious effect on the growth and productivity of three common understory shrub species, with biomass allocation patterns suggesting that the strongest competitive vectors were belowground. Increased NDF, which would suggest competition for nitrogen, was observed only in *C. americana*. Perhaps the fact that this species had a lower nitrogen concentration in roots in the intercropping treatment forced it to take up more fertilizer during leaf development rather than relying on stored reserves. No treatment effect for NDF was observed for *I. glabra*. Interestingly, NDF for all *M. cerifera* was lower in the intercropping treatment. This suggests, since nitrogen concentrations were not different between treatments, that this species obtained its nitrogen from another source – possibly biological nitrogen fixation. NDF for *M. cerifera* in the intercropping treatment was significantly lower than the other two species. UFN was higher for all species in the monoculture, reflecting the differences in biomass and indicating greater fertilizer use efficiency in the absence of competition. RFN_{soil} decreased with increasing depth, with little to no treatment differences observed for *C. americana* and *M. cerifera*, and significantly greater recovery, at all depths, for *I. glabra* in the intercropping treatment. Fertilizer uptake for pines, as a percentage of fertilizer applied, was estimated at 20.9%. Overall, while it is clear that interspecific competition was present in the intercropping system, the inefficiency of fertilizer use suggests that nitrogen was not the most limiting resource. Management interventions, particularly those that address competition for water, are likely critical to the success of this system.

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