EXPLAINING THE APPARENT RESILIENCY OF LOBLOLLY PINE PLANTATION TO ORGANIC MATTER REMOVAL

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Abstract -- We utilized 15-year measurements from an organic matter manipulation experiment in a loblolly pine plantation in the Upper Coastal Plain of Alabama to examine the apparent resiliency of a loblolly pine stand to organic matter removal. Treatments included complete removal of harvest residues and forest floor (removed), doubling of harvest residues and forest floor (added), and a standard harvest residue management (reference). Mineral soil and O horizons were sampled and analyzed for carbon (C) and nitrogen (N) using dry combustion. The $\delta^{15}N$ and CuO oxidation biomarkers were assessed in year 15. At year 15, there was no difference in volume between the reference and removed treatments while the added treatment exhibited higher volume. The $\delta^{15}N$ composition of the mineral soil from the removed site was enriched, suggesting that the removed treatment has experienced higher rates of N mineralization. Biomarkers from the CuO oxidation procedure support these assertions. These results have implications for long-term site productivity since recovery of labile N after severe removal of organic matter may be slow, and productivity of subsequent rotations may be negatively impacted if not ameliorated through management.

INTRODUCTION

Forest harvesting intrinsically removes organic matter and associated nutrients; these exports may impact long-term soil productivity and soil carbon (C) stores of managed forests. The Energy Independence and Security Act (EISA) of 2007 set a goal of 36 billion gallons per year of biofuels produced for consumption in the United States by 2022 and, in general, set goals and regulations that reduce U.S. dependency on foreign oil sources (EISA 2007). There are also developing biomass markets for electricity generation. In total, this means more intensive utilization of harvested materials from managed forests. In the U.S., the focus has been on whole-tree harvesting (i.e. boles, branches, tops, needles) (Powers and others 2005) which is associated with higher levels of nutrient and C removal than with standard harvesting operations and may impact long-term soil productivity and soil C stores.

Litter and slash are sources of soil organic matter (SOM), which is a potential long-term sink for atmospheric carbon dioxide. SOM plays a significant role in soil, enhancing cation exchange capacity, soil structure, aeration, water-holding capacity, and soil strength. Decomposing SOM provides the majority of mineralized forms of nitrogen (N) and phosphorus (P). Collectively, these characteristics of SOM play an integral role in sustaining site productivity (Fisher and Binkley 2000).

Most studies of organic matter removal associated with whole-tree harvesting have shown no significant effect on stand productivity (Ponder 2008, Powers and others 2005, Zerpa and others 2010). The North American Long-Term Soil Productivity (LTSP) network found no significant differences in tree productivity at age 10 following whole-tree harvesting and forest floor removal on 18 sites across North America (Powers and others 2005). To date, there has been surprisingly little research on mechanisms of the lack of negative response to stand productivity as a result of a relatively extreme treatment, such as slash and O-horizon removals. This apparent resiliency is the focus of this paper. Huang and others (2011) demonstrated that light-fraction (LF) C concentration can be reduced after whole-tree harvesting of *Pinus radiata* D. Don plantations. While not reported, there was probably a parallel response to LF (or labile) N concentration, suggesting that a stand may resist changes in growth if there is enough labile N to compensate for the losses. However, this response is not universal. Crow and others (2009) report that after 5 years of no above ground litter inputs (belowground inputs still allowed), there was no

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significant effect on LF C concentration relative to the control at the H.J. Andrews Experimental Forest in Oregon. However, when roots and litter were excluded for 5 years, there was a significant decline in LF C and likely a significant impact on labile sources of nutrients associated with organic matter (i.e. N). While there is much uncertainty regarding factors that control resiliency of stand productivity to organic matter removal, we hypothesize that the ability of a soil to be resilient to severe organic matter removals is a site property that could be assessed and monitored by measuring the labile pool of nutrients.

The objective of this research was to assess changes in site productivity by examining the effect of manipulating forest floor and harvest residue inputs. Specifically, we evaluated how these manipulations affected indicators of nutrient cycling and organic biomarkers in the context of intensive forest management.

MATERIALS AND METHODS

Site Description

The Millport Organic Matter Study is located in Lamar County, AL (33° 32' 22.87" N, 88° 77.53" W). The site is located on the Upper Coastal Plain physiographic province. Soils are classified as deep, well-drained Ruston series fine-loam, siliceous, semiactive, thermic Typic Paleudults (NRCS 2011). The nearest weather station with at least 23 years of precipitation and temperature data was in Tuscaloosa, AL (60 kilometers southeast). Average annual temperature from 1987 to 2010 was 16.9 ^{o}C and ranged from 6.3 ^{o}C in January to 27.1 ^{o}C in July (NOAA 2010). Mean annual precipitation was 1320 mm with the wettest month occurring in February (137 mm) and driest month in September (80 mm) (NOAA 2010). Columbus Air Force Base weather station was located 17.7 kilometers northwest of the study site, so it was be used to characterize precipitation and temperatures occurring during the period of study.

A 34-year-old loblolly pine stand was clearcut in 1994 preceding establishment of the current stand. Three treatments were established at the site: added, removed, and reference. Whole trees were harvested, and all slash and the entire forest floor was removed from the removed treatment. The added treatment had bole-only harvest with the slash and forest floor from the removed treatment transferred from an adjacent removed plot (e.g., plot 1 forest floor and residues were transferred to plot 2). This transfer of material preserved the order of O horizons (i.e. Oi, Oe, and Oa). The reference treatment had a bole-only harvest and standard harvest-residue management. A randomized complete block design was utilized containing three treatments and four replicates. Loblolly pine was planted on 4.3- by 3-m spacing in each 0.16-ha plot.

Site and Soil Sample and Data Collection

Stand volume was determined every 3 years through age 15. Diameter at breast height (d.b.h.) was measured on all trees, while 10 randomly selected trees per plot were subsampled to determine average plot height (10 percent of all trees) (Burkhart 1977).

Each plot contained five sampling locations consisting of the four plot corners and the plot center where soil and organic-horizon (O-horizon) data were collected. O-horizon was collected at age 15 from five points per plot and composited. Samples were oven dried at 55 to 60 ^{o}C for 24 hours. O-horizon bulk density was determined by multiplying the surface of the area collected (412 cm^2) by the average thickness of the horizon collected at this point. O-horizon composites were mixed thoroughly, sub-sampled, ground, and analyzed for C and N.

Mineral soils were sampled one time during the fall/winter of 2010-2011 at five locations per plot at 0- to 20-cm, 20- to 40-cm, and 40- to 60-cm depth with a hammer corer. Five samples were composited by plot for a total of 12 soil samples for each depth (48 total soil samples). Soil samples were oven dried at 55 to 60 ^{o}C. Composite samples at each depth were then sub-sampled, ground (Dyna-Crush Soil Grinder, Customer Laboratory Inc.), and analyzed for C and N. Additional soil samples were collected for bulk density determination. Bulk density samples were oven dried at 105 °C for 48 hours.

Density fractionation of mineral soil is a common procedure to determine dynamics of SOM. We utilized it to examine relative lability or recalcitrance of organic forms of N. A low-density (light) fraction in the mineral soil indicates a more labile N pool, while a higher density (heavy) fraction (HF) indicates more recalcitrant forms of N (Six and others 1999). Collected mineral soil samples were separated into LF and HF with a 1.64 g/cm^3 density sodium polytungstate (SPT) solution. Oven-dried soil samples (3 g each) were mixed with 5 g of SPT solution and put in a centrifuge at 3,000 rpm for 10 minutes. After each centrifuge run, LF material floated to the top of the vial and was aspirated and collected in a separate vial. This process was done six times to assure all of the < 1.64g/cm^3 particles were collected. The SPT solution containing LF was filtered through 0.47-µm combusted (3 hours at 350 ^{o}C) glass-fiber filters, oven dried overnight (55 to 60 ^{o}C), processed by randomly punching holes in the filters, and analyzed for total C and N. HF samples were lyophilized and analyzed for total C and N.

Laboratory Analyses

Subsamples of oven-dried O horizon were ground with a Thomas Wiley Laboratory Mill Model 4 using a 60-mesh sieve prior to C and N analysis. Dried mineral soils were subsampled and ground with a mortar and pestle. Organic and mineral samples were analyzed for C and N using dry combustion (Costech ECS 4010).

Samples were subjected to a variety of analyses designed to characterize the composition of the SOM. Details of all analytical techniques used in this study are provided by Hatten and others (2012). The stable isotopic compositions of C and N in the whole soils, and fractions of the 0- to 20-cm horizon were determined by isotope ratio mass spectrometry after high-temperature combustion of pre-acidified samples. In soils, the lighter forms of N are mineralized and taken up or leached, leaving behind the residual heavier N. This leads to a steady enrichment in ^{15}N with depth. We expected that if mineralization rates were impacted by organic matter removal or addition, the alteration would be reflected in the δ^{15}N signature. We hypothesized that if there were higher mineralization rates (faster cycling N) in mineral soil of the removed plot, it would have an enriched stable isotopic N signature relative to the reference. Lower mineralization rates on the added plot would lead to a relatively depleted signature relative to the reference.

We used alkaline CuO oxidation to determine concentrations of organic compounds derived from different biochemical precursors. In this paper we only report lignin-derived vanillyl phenols and parahydroxybenzenes, but many other biomarkers were measured. Vanillyl phenols are derived only from vascular plants while parahydroxybenzoic acids can be derived from both vascular plants and microorganisms. For this analysis, we used the ratio of parahydroxybenzoic acids to vanillyl phenols to examine the contribution of microorganisms to the organic C pool of the soils. Higher activities of microorganisms relative to the total organic C pool should result in a higher ratio.

Statistical Analysis

Analysis of variance (ANOVA) using a general linear models approach on a completely randomized block design was used to assess treatment effects of all parameters collected once during the duration of the study. A critical value of $\alpha = 0.05$ was used to test for significant differences. All ANOVA and Pearson correlation tests were run using SPSS (IBM SPSS Statistics, Version 21).

RESULTS AND DISCUSSION

Stand Volume and Periodic Annual Increment

Standing tree volume at age 15 in the added treatment was 31 and 22 percent higher than the removed and reference, respectively. No significant difference was found between the removed and reference treatments (fig. 1a). There was no significant difference between the treatments in the net volume increment from the last time these stands were measured at age 12. However, volume increment was significantly different between the ages of 5 and 8.

Whole Soils and Stand Productivity

Before describing the trends between treatments with respect to N, we needed to demonstrate that N is a limiting nutrient on these sites. Total N in the O (fig. 2a) and A (fig. 2b) horizons correlates significantly with stand volume at age 15. This suggests that N on these sites has some control over forest productivity. However,

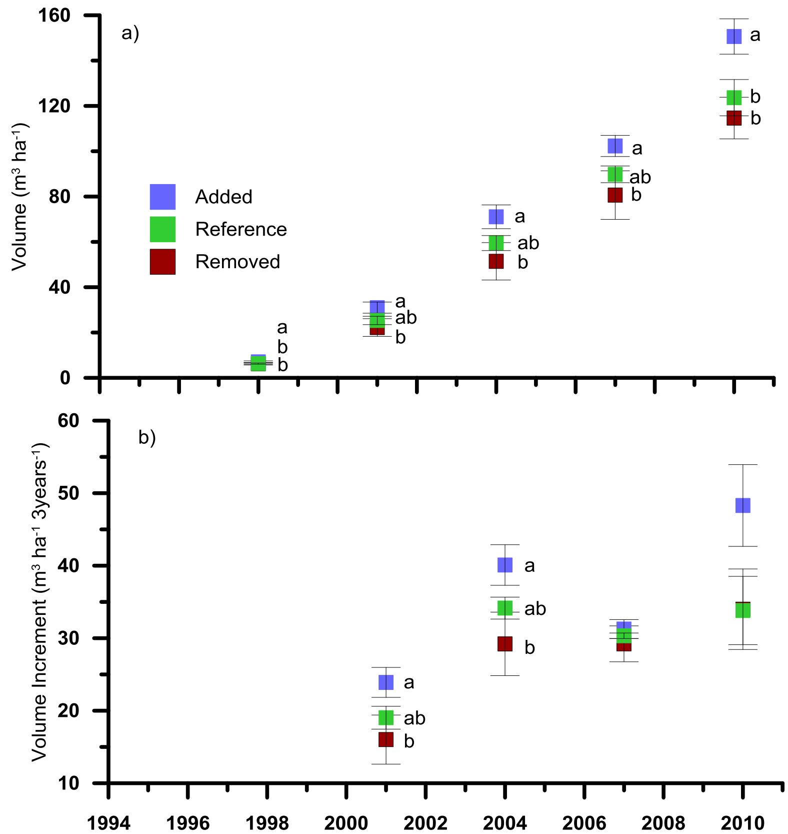

Figure 1--(a) Total bole volume, and (b) periodic annual increment from tree measurements made every year since stand initiation in 1994.

whole soil N content did not differ between treatments (total soil p = 0.395; fig. 3), although the added plots showed a trend of higher N content and the removed plots consistently showed a trend of lower N relative to the reference plots to a depths of 40 cm in the soil profile. It is possible that these small changes in N content result in trends in productivity.

By examining differences in the added and removed plots relative to the reference plots, we found that there was a significant relationship between the change in O-horizon N content and the change in productivity, suggesting that there was an increase in growth on the added treatments and a possible decrease in growth on some of the removed treatments (fig. 4). However, neither the change in volume nor the change in O-horizon N content was significantly different from zero, while only the change in volume was significantly different from zero for the added treatment.

We did not find any significant differences among the mass of LF N as we had hypothesized (data not shown). There were significant differences in HF N between added and removed treatments within the 0- to 20-cm horizon (fig. 5). However, neither treatment was different from the reference. This is possibly a result of recovery that has occurred since treatment application. The addition/removal of N in the slash and O horizon of the added/removed treatment was approximately ±300 kg/ha. In just the HF of 0 to 20 cm, the difference between the removed and reference treatments was about 151 kg/ha or 51 percent of the treatment effect. The HF of the 0- to 20-cm horizon of the added treatment contained 222 kg/ha more than the reference or 74 percent of the added N.

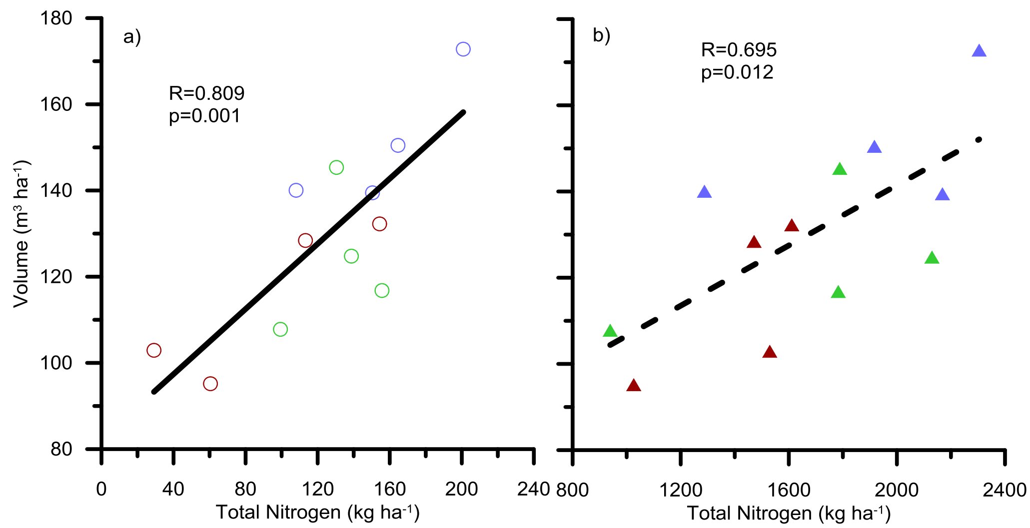

Figure 2--Relationship of total N in: (a) O horizon, and (b) 0- to 20-cm horizon. Colors of symbols represent the same treatments as in figure 1.

This suggests that the HF was at least partly responsible for resiliency of the stand, and this fraction should contain the signal of what has occurred in the soil since treatment.

In soils, depleted forms of N are mineralized and taken up by organisms or leached. The heavier isotope (^{15}N) is left behind leading to enrichment with depth. We expected a similar process to lead to enriched stable isotopic signatures in the removed treatment if N was mineralized faster. This faster rate of N mineralization may have partially relieved a N limitation caused by removing organic matter. Table 1 shows that there was not a significant difference in the HF stable isotopic N signature at $\alpha = 0.05$; however it was significant at $\alpha = 0.10$. In concordance with our hypothesis, HF from the removed treatments was enriched in ^{15}N, suggesting N was cycled faster on the removed plots. Interestingly, the added plots displayed a depleted stable isotopic signature relative to the reference, suggesting that N was cycled at a relatively slower rate. It should be noted that the O horizon has a depleted ^{15}N signature, so removal or addition of this material (i.e. the treatments) could have resulted in these trends. Additional measurements would be needed to determine if the signal in ^{15}N was a direct effect of the treatments or a result of a change in N mineralization rates and a mechanism for soil resiliency.

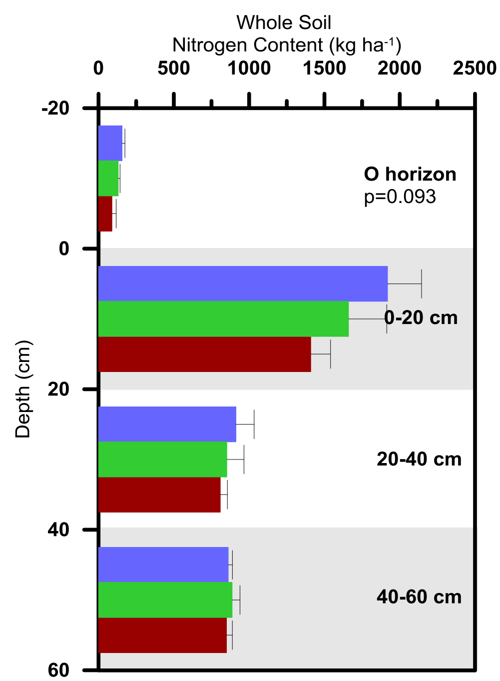

Figure 3--Total N in each soil horizon. Colors of bars represent the same treatments as in figure 1.

The ratio of parahydroxybenzoic acid to vannillyl is an indicator of microbial contributions to SOM. Elevated ratios suggest that there are higher rates of microbial activity relative to the total SOM pool. We found that the stable isotopic signature of the 0- to 20-cm whole soil and the parahydroxybenzoic acid to vannilyl ratio were

Table 1--Stable isotopic N signature of the HF from the 0 to 20 cm horizon. Enriched values of $\delta^{15}N$ suggest faster rates of N cycling

Treatment	$\delta^{15}N$
	ppm
Added	-0.67±0.38
Reference	0.17±0.17
Removed	1.07±0.85
p	0.07

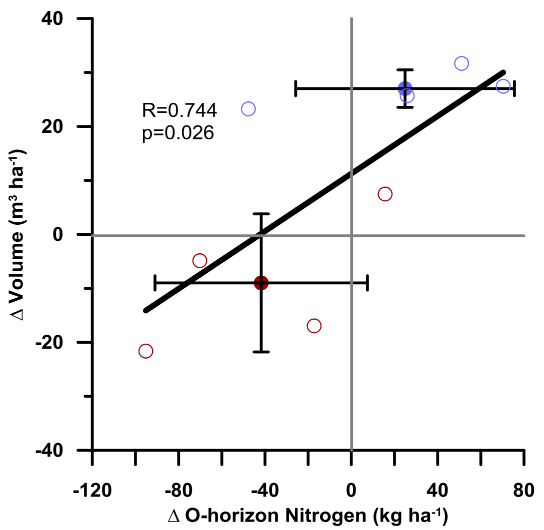

Figure 4--Relationship of the change in O-horizon N content and change in volume relative to the reference plot within each block. Solid circles represent averages with 95 percent confidence interval. Colors of circles represent the same treatments as in figure 1.

significantly correlated, suggesting that a portion of the stable isotopic signature was a result of higher rates of microbial activity (fig. 6a). We also found that there were significant differences in this indicator ratio among the treatments at α = 0.10 (fig. 6b). This trend is not the result of the addition or removal of organic matter since O horizons had a much higher ratio of parahydroxybenzoic acids to vannilyl ratio. This result was similar across several other biomarkers including fatty acids, diacids, and benzene dicarboxylic acids (all markers of relative contributions of microbes to SOM).

We found evidence for a slight nutrient limitation in the removed treatment early in stand history, but this nutrient limitation was alleviated by higher rates of mineralization of soil N pools. There is the possibility that the removed treatment could have been affected by higher competition due to the favorable seedbed conditions created by the removal of slash and O-horizon material. The added treatment responded to the addition of nutrients by increasing growth rates relative to the control; however this could have also been confounded by differences in competition early on in the stand.

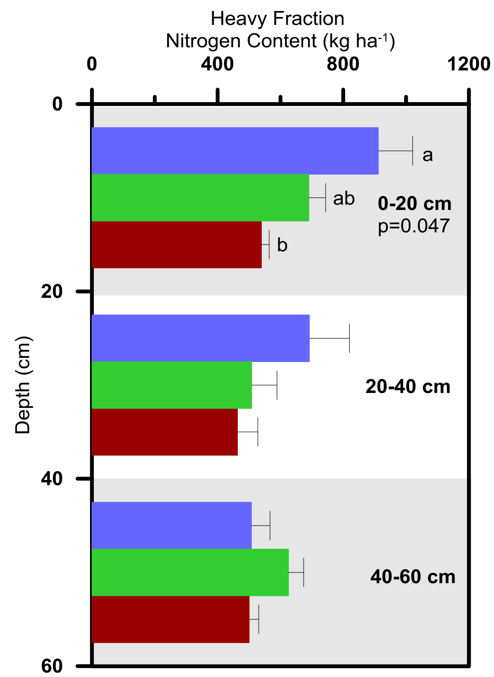

Figure 5--Total heavy fraction N in each soil horizon. The HF represents what is thought to be a slow cycling pool of N in the soil. Colors of bars represent the same treatments as in figure 1.

Despite uncertainties regarding the levels of competition experienced by the developing stands, we found compelling evidence of a mechanism for resiliency in these soils that includes the mineralization of what is thought to be a recalcitrant pool of organic nutrients. What drove the mineralization of this recalcitrant pool? Possibly changes in soil moisture and temperature as a result of exposing the mineral soil directly to precipitation and sunlight, changes to the microbial community, or possibly some other conceived mechanism. More research is necessary to more certainly determine the mechanisms of this resiliency.

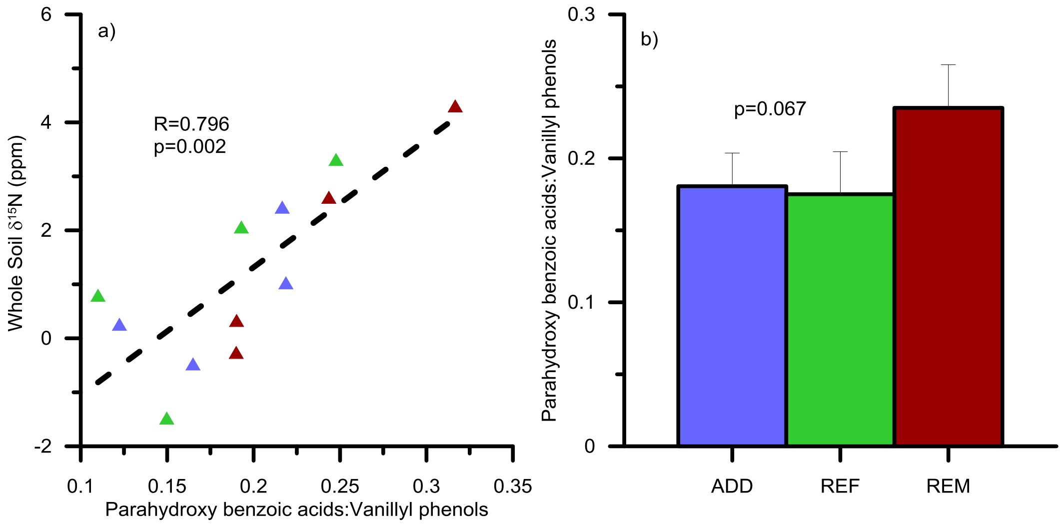

Figure 6--(a) Relationship of 0 to 20 cm $\delta^{15}N$ and parahydroxy benzoic acids in the 0- to 20-cm soil horizon; and (b) ratio of parahydroxy benzoic acids to vanilyl phenols in the 0- to 20-cm horizon. Parahydroxy benzoic acids are a biomarker of microbial contributions to SOM. Colors of symbols represent the same treatments as in figure 1.

CONCLUSIONS

We found evidence for a mechanism that alleviated a possible nutrient limitation in a loblolly pine plantation with a severe organic-matter-removal treatment. This mechanism appeared to be higher rates of mineralization of slow cycling soil N pools. These results have implications for long-term site productivity since the recovery of labile N after severe removal of organic matter may be slow, and productivity of subsequent rotations may be negatively impacted if not ameliorated through management.

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