RESEARCH ARTICLE

Key Points:
- Intensive timber harvest leads to reductions of soil carbon and nitrogen in the upper 1 m of the soil profile that can persist for decades
- Seasonal variations in soil carbon and nitrogen are attributable to concurrent variations in root biomass and forest floor mass
- Long-term reductions in soil carbon may influence rates of biogeochemical processes that influence soil fertility and forest productivity

Supporting Information:
- Supporting Information S1
- Table S1
- Table S2
- Data Set S1

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Decadal-scale changes in forest soil carbon and nitrogen storage are influenced by organic matter removal during timber harvest

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Abstract This study investigates whether different intensities of organic matter removal associated with timber harvest influence decadal-scale storage of soil organic carbon (SOC) and total nitrogen (TN) in the top 1 m of mineral soil 18 years postharvest in a *Pinus taeda* L. forest in the Gulf Coastal Plain. We quantified forest harvest-related changes in SOC, TN, microbial biomass carbon (MBC), and nitrogen (MBN) pools (0–100 cm) in unharvested control stands and in two organic matter removal treatment stands subjected to either (i) merchantable bole/stem-only harvest or (ii) whole-tree harvest + forest floor removal. In addition, δ^{13}C of SOC and δ^{15}N of TN were measured in mineral soil to provide insights regarding mechanisms that might explain changes in SOC and TN pool sizes. Soils were sampled seasonally for 1 year. Increasing organic matter removal intensity reduced SOC, TN, MBC, and MBN relative to the unharvested control. Furthermore, soils from whole-tree harvest + forest floor removal stands had lower δ^{13}C and higher δ^{15}N values, suggesting that increasing organic matter removal may decrease heterotrophic activity as well as increase rates of N loss. Seasonal variabilities in SOC and TN were correlated to changes in forest biological properties such as root biomass and forest floor mass. These results indicate that more intensive harvest methods may lead to decade-scale decreases in SOC and TN storage in surface and subsurface soils which could influence rates of biogeochemical processes, the availability of soil nutrients, and potential forest productivity.

1. Introduction

The global soil organic carbon (SOC) pool stores approximately 1200–1550 Pg C in the upper 1 m of soil, which represents about 75% of the terrestrial carbon (C) pool [*Jobbagy and Jackson*, 2000; *Houghton*, 2007; *Batjes*, 2014]; congruently, soil total nitrogen (TN) stocks are estimated at 133–140 Pg N within that same depth interval [*Batjes*, 2014]. In forest soils, the upper 1 m stores 353–413 Pg C which is nearly 30% of global SOC and accounts for more C than is stored in above and belowground live biomass, deadwood biomass, and litter biomass [*Pan et al.*, 2011]. Furthermore, it has been noted that most forest ecosystems store between 2 and 12 Mg N ha^{-1} in mineral soil [*Johnson and Turner*, 2014], which is equivalent to 8–48 Pg N across all forestlands [*Food and Agriculture Organization*, 2015]. *Hansen et al.* [2010] recently showed that the global rate of gross forest cover loss due to natural and anthropogenic perturbations between the years 2000 to 2005 was $>1 \times 10^6$ km^2, equivalent to 3.1% of global forest cover in 2000. Given the geographic dimensions of forest disturbance, the magnitude of soil C and N stores in forest soils, and the important roles of these elements in global biogeochemistry and the climate system, it is important to understand how forest soils might respond to disturbance.

Responses of SOC and TN stocks in surface and subsurface mineral soils may be particularly relevant in the context of timber harvesting, whose legacy effects may last for decades to centuries [*Chen et al.*, 2013; *Kellman et al.*, 2014; *Prest et al.*, 2014; *Dean et al.*, 2016]. Both SOC and TN are important indicators of soil quality due to their ability to influence soil structure, nutrient concentrations, water-holding capacity, and microbial activity [*Lal*, 2004; *Bationo et al.*, 2007]. Removal or redistribution of forest biomass during timber harvest has the potential to alter SOC and TN stocks and modify rates of nutrient cycling processes [*Chen et al.*, 2013; *Dangal et al.*, 2014; *Vario et al.*, 2014], potentially jeopardizing forest production and sustainability. Furthermore, forest biomass removal may also substantially affect climate change through the conversion of forests from carbon sinks to carbon sources [*Pan et al.*, 2011; *Chen et al.*, 2013]. For these reasons, the accurate quantifications of SOC and TN are essential for determining the long-term

influence of intensive timber harvest regimes. Studies investigating the effect of forest disturbance on SOC and TN have generally been limited to the top 10 cm of the soil profile; however, it has become apparent that deeper (i.e., >10 cm depth) soil C and N stocks need to be quantified in order to more fully characterize the effects of forest harvest [*Diochon et al.*, 2009; *Slesak et al.*, 2011; *Buccholz et al.*, 2014; *James et al.*, 2014; *James et al.*, 2015]. Furthermore, SOC and TN temporal dynamics at depth are poorly understood and should be investigated to determine what factors maintain C and N in subsurface soil horizons.

Investigations on changes in SOC storage in response to differing timber harvest methods have been shown to result in divergent conclusions. Studies from single locations or regions sometimes report reductions in SOC with increasing timber harvest intensity [*Li et al.*, 2003; *Jones et al.*, 2011; *Huang et al.*, 2013], while other report no differences [*Johnson and Todd*, 1998; *Zerpa et al.*, 2010]. However, most meta-analyses that incorporate studies from across a range of abiotic and environmental gradients consistently report that intensive timber harvest results in a general reduction in SOC [*Johnson and Curtis*, 2001; *Nave et al.*, 2010; *Achat et al.*, 2015a]. Specifically, *Achat et al.* [2015a] showed that intensive timber harvest methods involving removal of harvest residues can reduce SOC content in mineral soil by 10%. These reductions have been shown to be positively correlated to both the amount of harvested biomass removed and the C and N stores in that biomass [*Hazlett et al.*, 2014; *Kellman et al.*, 2014; *Vario et al.*, 2014; *Achat et al.*, 2015a, 2015b]. A recent literature review and modeling study has suggested that the observed wide range of SOC responses to forest harvest is likely due to variability in (a) the time interval between the harvest event and SOC sampling and (b) the number of prior logging cycles at a given site [*Dean et al.*, 2016]. It should also be noted that soil C has been shown to take several decades to recover to preharvest levels following harvest, with some soil orders taking upward of 75 years to recover [*James and Harrison*, 2016]; however, continued research is needed to further investigate the mechanisms governing soil C recovery, especially at depth.

In addition to SOC and TN stocks, differing timber harvest methods can also influence the size of the soil microbial biomass pool and the rates of the biogeochemical processes that they mediate. Microbial biomass C (MBC) and N (MBN) are fundamental components of SOC and TN as well as indicators of biogeochemical potential in surface and subsurface soil [*Wardle*, 1992; *Gallardo and Schlesinger*, 1994; *Zak et al.*, 1994] and should be investigated when quantifying SOC and TN stocks. It has been shown that the magnitude of microbial biomass is highly correlated with pool sizes of SOC and TN [*Wardle*, 1992; *Allen and Schlesinger*, 2004]; therefore, any changes in the size of SOC and TN pools following disturbance may negatively impact the microbial biomass, even at depth. Previous studies have shown that surface soil microbial biomass (i.e., 0–10 cm) can be affected by timber harvest [*Busse et al.*, 2006; *Foote et al.*, 2015]; however, no study has investigated whether this trend persists in deeper portions of the soil profile.

δ^{13}C values of SOC and δ^{15}N values of TN can add insight into the relative magnitude of C and N inputs versus losses from the soil following disturbance [*Ehleringer et al.*, 2000; *Robinson*, 2001; *Pataki et al.*, 2003; *Diochon and Kellman*, 2008; *Schlesinger*, 2012]. The few studies that have investigated changes in δ^{13}C values in response to different levels of timber harvest showed enrichment in δ^{13}C as harvest intensity increases [*Diochon and Kellman*, 2008; *Huang et al.*, 2011]. Congruently, disturbances that accelerate ecosystem N losses through higher rates of denitrification or nitrification can result in enrichment of bulk soil δ^{15}N [*Nadelhoffer and Fry*, 1994; *Högberg*, 1997; *Bai et al.*, 2013]. Few studies have utilized δ^{13}C and δ^{15}N values to better understand the mechanisms that might lead to changes in soil C and N stores in either surficial or deep soils.

The purpose of this study was to quantify the long-term (decade-scale) consequences of different timber harvest methods on C and N pool sizes in mineral soil and in the soil microbial biomass throughout the upper 1 m of the profile. We hypothesized that (1) SOC, TN, and microbial biomass would be lowest in harvest treatments with the highest levels of organic matter removal, and these decreases would be evident throughout the entire soil profile; (2) soil δ^{13}C and δ^{15}N values would be more enriched in the more intensively harvested treatment due to accelerated C and N losses; and (3) SOC, TN, and microbial biomass compartments would vary seasonally in the upper portions of the soil profile in response to intra-annual variation in forest floor and root inputs but not in deeper portions of the profile where organic matter inputs are more limited.

2. Materials and Methods

2.1. Study Site and Experimental Design

Research was conducted in Davy Crockett National Forest near Groveton, Texas, USA (31°06′32.48″N, 95°09′59.15″W) at a site that is part of the Long-term Soil Productivity (LTSP) network [*Powers*, 2006; *Ponder et al.*, 2012]. Topography is nearly flat with slopes of 1–3% and elevation ranging from 101 m to 110 m. Soil across the study area is classified as a fine-loamy, siliceous, thermic Oxyaquic Glossudalf in the Kurth series which developed in loamy coastal plain sediments of the Yegua and Whitset geological formations [*U.S. Department of Agriculture/Natural Resources Conservation Service*, 2003]. The climate is subtropical with a mean annual temperature of 18.7°C and mean annual precipitation total of 1107 mm (1950–2010). Rainfall is evenly distributed throughout the year with May and September as the wettest months. Potential annual evapotranspiration is approximately 1200 mm [*Norwine et al.*, 2005]. Climate data during the study period were obtained from the NOAA National Climatic Data Center weather stations in Crockett, TX, and Lufkin, TX, 38 km northwest and 48 km northeast of the study site, respectively, and averaged to obtain mean values for the study area.

The experimental design used in this study includes (i) unharvested control forest stands composed of trees 60–80 years of age, (ii) merchantable bole/stem-only harvest stands, and (ii) whole-tree harvest + forest floor removal stands. The unharvested control stands have been thinned intermittently; however, the last thinning was before the treatment plots were established (i.e., >20 years ago). Each treatment was composed of three replicates, and each replicated plot was approximately 0.2 ha (i.e., 63 m × 32 m). In 1996, trees in harvested plots were hand-felled and lifted off of the plots with a loader. Forest floor removal was accomplished by hand-raking all aboveground organic matter from the whole-tree + forest floor removal treatment plots. Containerized *P. taeda* L. seedlings of 10-half sib families from the U.S. Forest Service seed orchards were hand-planted on a 2.5 m × 2.5 m spacing in 1997.

2.2. Sample Collection

Soil cores were collected at approximately 4 month intervals from June 2014 through March 2015 by using a JMC Environmentalist Sub-Soil Probe PLUS, 2.8 cm diameter × 120 cm length coring tube (Clements Associates Inc., Newton, IA, USA). Soil cores were taken at 1.8 m from the base of a randomly selected *P. taeda* L. individual with a diameter at breast height (DBH) between 18 and 24 cm. A three tree buffer from the outside of the plots was not sampled to avoid edge effects. At each sample point, forest floor materials were collected down to the mineral soil from a 0.25 × 0.25 m quadrat followed by the extraction of a soil core. In this study the term forest floor is equivalent to the O-horizon (Oi + Oe + Oa). Soil sampling followed a stratified random sampling design in which four cores were taken from each plot and pooled by depth increment to increase sample mass and reduce error introduced by environmental heterogeneity. Specifically, each soil core was partitioned into four depth increments in the field (0–10, 10–30, 30–60, and 60–100 cm), pooled together with the other replicated cores, and individual depths were analyzed separately. Samples were transported on ice packs from the field to the lab on the same day they were taken from the ground. Soil samples were aseptically homogenized in the lab and stored at 4°C until analyzed.

2.3. Soil Chemical and Physical Characterization

Soil pH was determined by using an Accumet Basic pH meter (Denver Instrument, Arvada, CO, USA) on a 1:2 solution of soil in a 0.01 M $CaCl_2$ solution [*Minasny et al.*, 2011]. A 50 g aliquot of field-moist soil was dried at 105°C for 48 h to calculate bulk density and volumetric soil moisture. The remaining soil was passed through a 2 mm sieve to homogenize the soil and to remove large organic fragments and roots. Roots were saved for biomass quantification. A 25 g aliquot of sieved soil was then dried at 60°C for 48 h and finely ground into powder by using a TE250 ring pulverizer (Angstrom, Inc., Belleville, MI, USA). The pulverized soil was used to determine the concentration and isotopic composition of C and N. An additional 125 g aliquot of sieved soil was dried for 48 h at 105°C for texture analysis by using the hydrometer method [*Ashworth et al.*, 2001].

2.4. Carbon and Nitrogen Concentrations, Densities, and Isotopic Composition

Soils were analyzed for SOC and TN concentrations, as well as their $\delta^{13}C$ and $\delta^{15}N$ values, in the Stable Isotopes for Biosphere Science Laboratory at Texas A&M University. Analyses were conducted on a Carlo Erba EA-1108 elemental analyzer (CE Elantech, Lakewood, NJ, USA) interfaced with a Thermo Fisher Delta

Plus isotope ratio mass spectrometer (Thermo Fisher Scientific, Inc., Waltham, MA, USA) in continuous flow mode. Carbon and N isotope ratios were reported in delta notation:

$$\delta^{xx}E(\text{‰}) = \left[\left(R_{\text{sample}} - R_{\text{standard}}\right)/R_{\text{standard}}\right] \times 1000$$

where E is the element (either C or N), R_{sample} is the ratio of either ^{13}C:^{12}C or ^{15}N:^{14}N in the sample, and R_{standard} is the ratio of ^{13}C:^{12}C of the international standard Vienna Peedee belemnite [*Coplen et al.*, 2006] or ^{15}N:^{14}N of the international atmospheric N$_2$ standard [*Mariotti*, 1983]. Soil C and N stocks (g m^{-2}) were computed as the product of the elemental concentration and soil bulk density for each soil depth [*Ellert and Bettany*, 1995]. Carbon to nitrogen ratios (C:N) were calculated as the proportion of SOC to TN on a g kg^{-1} basis.

2.5. Microbial Biomass

Microbial biomass carbon (MBC) and microbial biomass nitrogen (MBN) were determined on homogenized soil subsamples by using the chloroform fumigation extraction method [*Vance et al.*, 1987]. Two, 10 g, field fresh aliquots of each sample were placed into separate 50 mL glass beakers. One aliquot served as a nonfumigated control and was immediately extracted with 40 mL of 0.5 M K$_2$SO$_4$, shaken for 1 h, centrifuged at 700×g for 10 min, and filtered over preleached (0.5 M K$_2$SO$_4$) #5 Whatman filter paper, and the filtrate was analyzed for dissolved organic C (DOC) and dissolved organic N (DON) by using a Shimadzu TOC-V$_{\text{CSH}}$ with a TNM-1 module (Shimadzu Corp., Kyoto, Japan) set for 5x dilution [*Chen et al.*, 2005]. The second 10 g aliquot was fumigated at field moisture in a dark vacuum desiccator for 24 h in the presence of ethanol-free chloroform. Following the incubation, DOC and DON from each fumigated sample were extracted and analyzed by using the same procedure used for the nonfumigated control. MBC and MBN were calculated by using the following formula:

$$\text{MBC} = \left(\text{C}_{\text{fumigated}} - \text{C}_{\text{control}}\right)/k_{\text{EC}}; \text{and}$$
$$\text{MBN} = \left(\text{N}_{\text{fumigated}} - \text{N}_{\text{control}}\right)/k_{\text{EN}}.$$

Because extraction efficiencies for DOC and DON are less than 100%, extraction coefficients for carbon (k_{EC}) of 0.45 [*Potthoff et al.*, 2009; *Joergensen et al.*, 2011] and nitrogen (k_{EN}) of 0.54 [*Brookes et al.*, 1985] were used to calculate soil microbial biomass carbon and nitrogen, respectively.

2.6. Vegetation, Roots, and Forest Floor Quantification

Diameter at breast height (DBH) and tree height were measured for 150 and 30 randomly selected individuals per treatment, respectively. Understory vegetation cover was measured by using the line intercept method; specifically, six randomly placed parallel transects that measured 63 m in length were placed in each of the plots, and the horizontal linear length of each understory plant that intercepts each line was noted as well as the identity of the plant. Those values were added together, divided by the total length of the transect and then multiplied by 100 to obtain an understory vegetation percentage. The understory percentages were averaged by plot, and those values were used to compare treatments by using a student's *t* test. Roots collected during sieving and all forest floor materials were dried at 60°C until stable mass was achieved and then weighed.

2.7. Statistical Analyses

All data and statistical analyses were performed by using JMP Pro 11 (SAS Institute, Inc., Cary, NC, USA) or OriginPro (OriginLab, Inc., Northhampton, MA, USA). All data sets were tested for normality by using Shapiro-Wilk's test. When data were not of normal distribution, log transformations were applied. Edaphic variables were statistically analyzed by using a linear mixed model analysis of variance (ANOVA). Because of the inherent autocorrelation between differing soil depths, a split plot experimental design with repeated measures was employed with harvest treatment as the fixed main plot, soil depth designated as the fixed split plot, and time incorporated as the repeated measure. Replicated plots were nested within harvest treatment and were considered a random effect. Results from the mixed model ANOVA were compiled into Table 1. When differences were significant, Tukey's honest significant difference test was performed to assess posthoc contrasts with significance inferred at $\alpha \leq 0.05$. Results from posthoc analysis were compiled into Table S1 in the supporting information. Spearman's correlation analysis for all sample points including time, harvest treatment, and soil depth was used to assess connections between physicochemical properties.

Table 1. Effects of Harvest Treatment, Soil Depth, Time, and Their Interactions on Soil Chemical, Physical, and Biological Properties

	Harvest Treatment (OMR)	Soil Depth (SD)	Sampling Time (T)	OMR × SD	OMR × T	SD × T	OMR × SD × T
			F-Ratio				
VWC (%)	3.5	22.8***	70.7***	2.6*	2.0	1.1	0.6
Sand (%)	2.1	25.3***	0.5	4.3***	0.1	0.3	0.1
Silt (%)	12.6**	16.3***	13.0***	1.8	0.5	0.4	0.4
Clay (%)	1.5	58.3***	9.9***	3.0*	0.2	0.3	0.4
Soil pH	4.6*	24.2***	0.1	8.3***	1.0	0.2	0.2
Bulk density (g cm^{-3})	2.7	113.9***	2.1	1.3	2.2	0.8	0.9
Roots (g m^{-2})	2.1	46.5***	5.7**	2.2*	0.3	1.1	0.4
[a]Forest floor (g m^{-2})	1296.7***	–	39.4***	–	8.4***	–	–
SOC (g C m^{-2})	14.1**	378.5***	0.6	3.6**	2.9*	1.2	1.2
TN (g N m^{-2})	2.8	128.9***	3.8*	0.9	0.8	0.5	0.4
SOC (g C kg^{-1})	9.2*	207.5***	1.2	2.7*	1.1	0.8	0.7
TN (g N kg^{-1})	2.4	64.9***	5.9**	0.9	0.4	0.4	0.4
C:N	0.3	143.9***	3.8*	2.1	3.0*	0.6	0.9
δ^{13}C (‰)	1.1	89.4***	0.4	1.1	0.3	0.1	0.4
δ^{15}N (‰)	4.5	112.1***	37.1***	0.8	0.4	1.6	0.6
MBC (μg g^{-1})	9.2*	258.6***	39.4***	4.8***	2.7*	9.5***	1.6
MBN (μg g^{-1})	1.9	192.3***	42.1***	1.3	0.8	17.9***	0.9

[a]Forest floor mass was statistically analyzed for harvest treatment and time by using a two-way ANOVA.
*$p < 0.05$.
**$p < 0.01$.
***$p < 0.001$.

3. Results

3.1. Climate and Soil Characteristics

Precipitation in 2014 was 1136 mm which was similar to the 60 year average of 1107 ± 33 mm (mean ± standard error); however, the first 6 months (January–June) of 2015 recorded a total of 1152 mm, which was 50% higher than the 60 year average of 577 ± 50 mm over that same monthly interval (Figure 1). From January 2014 through June 2015, temperatures did not deviate appreciably from the 60 year average (Figure 1). Soil volumetric water content (VWC) varied significantly with time and depth but not harvest treatment (Figure 1). VWC was highest in June 2014 and March 2015, reflecting the distribution of rainfall during the duration of the study.

Soil texture in all treatments was a uniform sandy loam from 0–60 cm, consisting of 685.8 ± 14.2 g kg^{-1} sand, 179.5 ± 12.1 g kg^{-1} silt, and 134.7 ± 12.4 g kg^{-1} clay. From 60 to 100 cm, soil texture was a sandy clay loam consisting of 594.9 ± 17.5 g kg^{-1} sand, 146.8 ± 11.1 g kg^{-1} silt, and 258.3 ± 21.8 g kg^{-1} clay (Figure 2). When averaged across all depths, controls (227.1 ± 6.9 g kg^{-1}) possessed a significantly higher proportion of silt than the bole-only harvest treatment (124.7 ± 45.7 g kg^{-1}) or the whole-tree harvest + forest-floor removal treatment (162.3 ± 6.2 g kg^{-1}). Generally, soil pH decreased with depth and increased with increasing timber harvest intensity (Figure 2) but was not altered by time. Soil from the whole-tree harvest + forest floor removal plots were significantly higher (pH = 4.02 ± 0.07) than soil from the bole-only harvest stands (pH = 3.77 ± 0.09) and unharvested control stands (pH = 3.57 ± 0.07) over the entirety of the 1 m soil core (Figure 2). Bulk density increased significantly with soil depth ($p < 0.001$), ranging from 1.03 ± 0.03 g cm^{-3} at 0–10 cm to 1.60 ± 0.03 g cm^{-3} at 60–100 cm (Figure 2).

3.2. Vegetation Composition, Root Biomass, and Forest Floor Mass

When the two harvest treatments were compared by using a single factor ANOVA (i.e., bole-only harvest versus whole-tree harvest + forest floor removal), mean DBH and tree height were statistically larger in bole-only harvest stands (Table 2). Unharvested control stands were not included in this analysis because of the differences in tree age; however, mean DBH and tree height for the unharvested control stands were larger than either of the treatment plots (Table 2). Average understory cover per plot was 33.8 ± 1.4% and did not vary by treatment (Table 2). Nearly 90% of the understory was attributed to *Ilex vomitoria* (Table 2). Root biomass did not vary with respect to harvest intensity; however, significant differences occurred between soil depths (Figure 3). Specifically, root biomass was highest in the surface
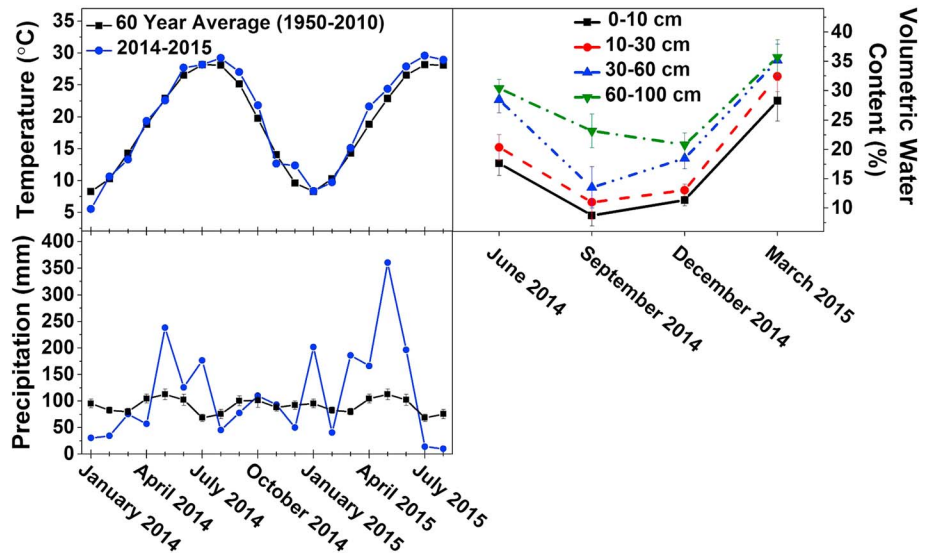
Figure 1. Monthly distribution and 60 year mean of air temperature and total precipitation from 5 months prior, during, and 5 months after the study as well as volumetric water content for each depth of interest during the four sampling points. The error bars for the 60 year average indicate standard error and due to low standard error for the temperature are not observable on this figure. Data for air temperatures and total precipitation are from the National Oceanic and Atmospheric Administration sites in Crockett, TX, and Lufkin, TX. Temperature and precipitation were averaged from the two sites to obtain an overall mean for the entire area.

soil (1079.5 ± 52.6 g m^{-2}) and decreased to 166.8 ± 13.9 g m^{-2} at 1 m. Root biomass in the 10–30 cm increment (475.3 ± 55.5 g m^{-2}) was not significantly different than what was found in the 30–60 cm increment (611.4 ± 81.7 g m^{-2}). Forest floor mass significantly varied with harvest intensity and time.
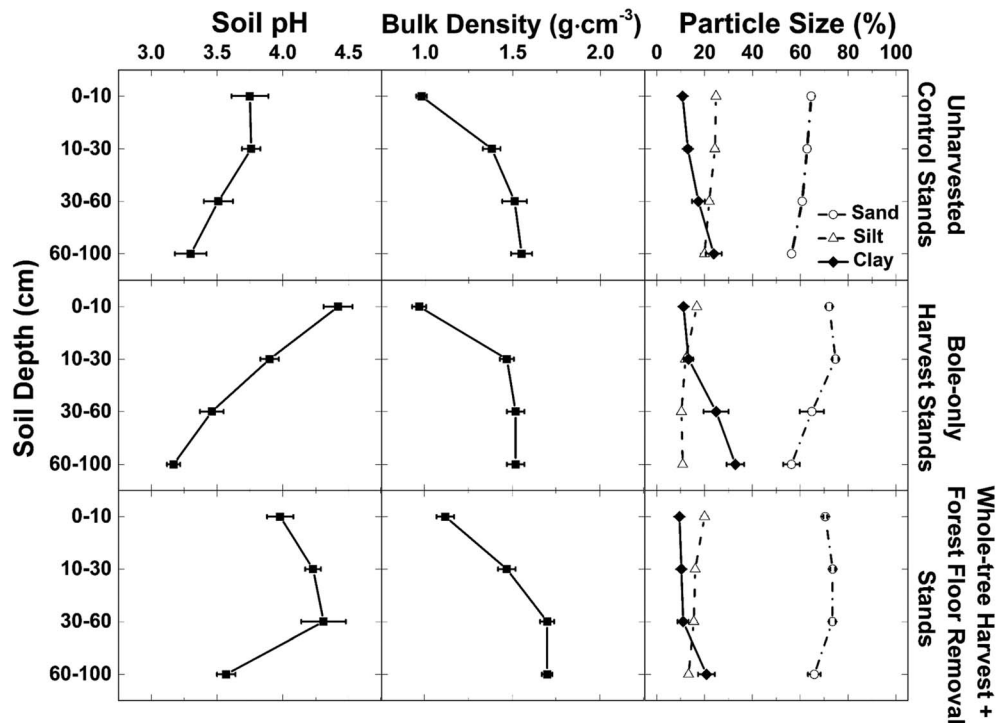

Figure 2. Physical and chemical characteristics of soil from different timber harvest methods over a range of soil depth increments (0–100 cm). Each point is the mean ± standard error of 12 replicates. The symbols used for particle size distribution: white circle: sand, white triangle: silt, black diamond: clay.

Table 2. Forest Vegetation and Biological Properties for Each Harvest Treatment[a]

	P. taeda	P. taeda	Total Understory	I. vomitoria	Forest Floor	Roots
	DBH (cm)	Height (m)	Cover (%)	Cover (%)	(g m^{-2})	0–100 cm (g m^{-2})
Harvest treatment	N = 150	N = 30	N = 18	N = 18	N = 12	N = 48
Unharvested control stands	27.89 (1.23)	22.92 (2.16)	34.07 (2.92)	31.04 (2.70)	2369.36 (107.94)	661.47 (69.80)
Bole-only harvest stands	20.66 (0.36)a	17.43 (0.65)a	34.78 (2.18)a	32.06 (2.13)a	1633.42 (34.75)a	690.25 (73.27)a
Whole-tree harvest + forest floor removal stands	17.59 (0.30)b	14.34 (0.54)b	32.81 (2.01)a	29.91 (1.77)a	1056.17 (32.04)b	398.03 (51.56)b

[a]A single factor ANOVA was used to compare the two harvest treatments with significance inferred at $\alpha \leq 0.05$. Values are listed as mean (standard error). Different letters following values indicate significant difference.

When averaged across all time points, forest floor mass was highest for the unharvested control stands (2369.4 ± 107.9 g m^{-2}) and lowest in the whole-tree harvest + forest floor removal stands (1056.2 ± 32 g m^{-2}). When averaged across all treatments, mean forest floor mass was highest in March (1902.4 ± 262.9 g m^{-2}) and lowest in December (1520.4 ± 159.4 g m^{-2}).

3.3. SOC, TN, and C:N Ratio

Mean SOC concentrations, aggregated from all depths and sampling times, in unharvested control stands (8.1 ± 1.2 g kg^{-1}) and the bole-only harvest stands (7.4 ± 1 g kg^{-1}) did not differ from each other, but both possessed significantly larger concentrations of SOC than did the whole-tree harvest + forest floor removal stands (4.7 ± 0.8 g kg^{-1}) (Figure 4). SOC concentrations, averaged across all harvest treatments and sampling times, in the 0–10 cm (17.9 ± 1 g kg^{-1}) and 10–30 cm (4.4 ± 0.3 g kg^{-1}) increments were significantly different from all other depths. The 30–60 cm (2.3 ± 0.2 g kg^{-1}) and 60–100 cm (2.3 ± 0.1 g kg^{-1}) depth increments
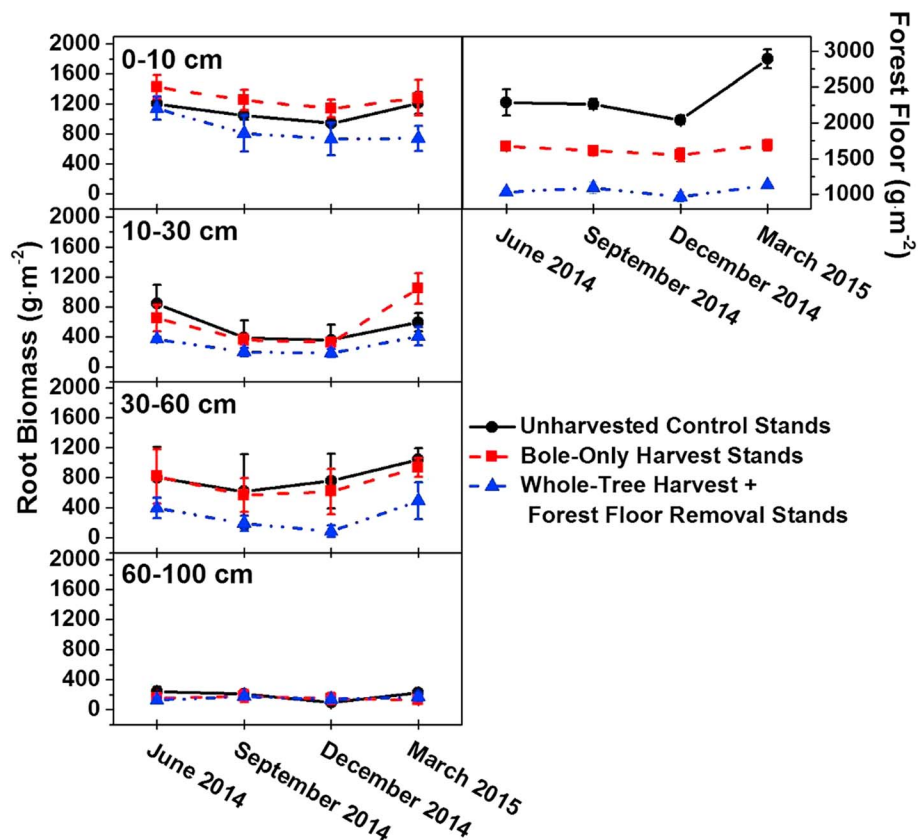

Figure 3. Variations in root biomass and forest floor mass (g m^{-2}) for each sampling point. Root biomass is separated into depth increments with each point being the mean ± standard error of three replicates. Singular points for forest floor mass represent the mean ± standard error of three replicates.
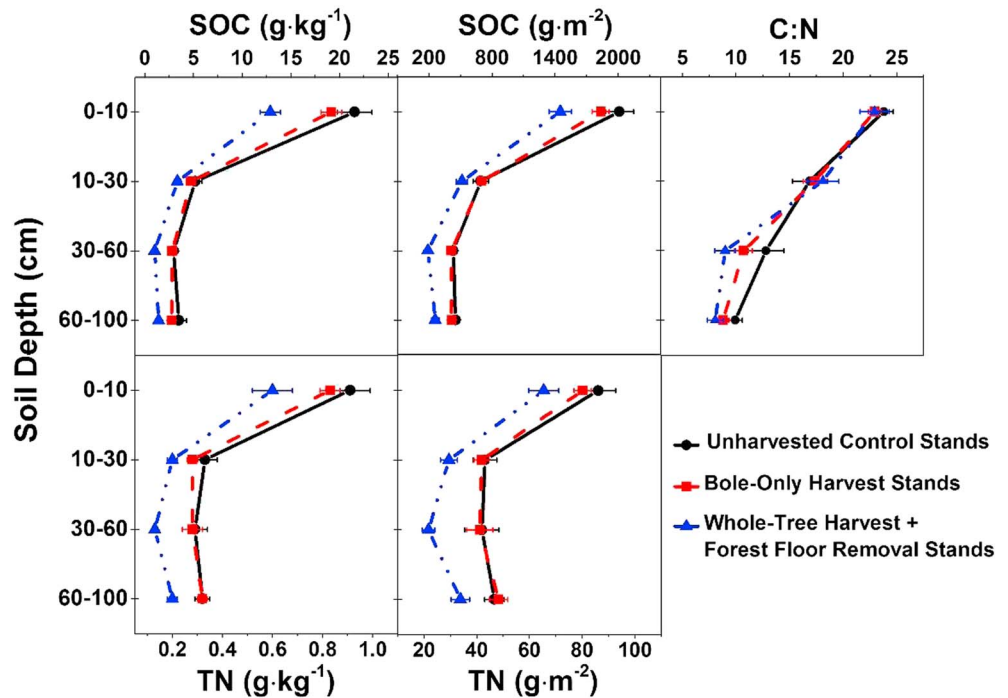
Figure 4. Soil organic carbon (SOC) and total nitrogen (TN) stocks and concentrations as well as ratios of SOC to TN (C:N) over the top 1 m of the soil profile. Each time point is the mean ± standard error of 12 replicates without regard for seasonal differences.

contained statistically smaller concentrations of SOC than the shallower depths ($p < 0.001$) (i.e., 0–10 and 10–30 cm); however, they did not vary from each other. When summed SOC stocks (g C m^{-2}) from all depth increments are averaged across all sampling time points and analyzed for treatment differences by using a one-way ANOVA, we observe significantly higher stocks in the unharvest control (3681.5 ± 283.3 g C m^{-2}) and bole-only harvest (3366.7 ± 191.9 g C m^{-2}) than the whole-tree harvest + forest floor removal treatment (2417.8 ± 217.6 g C m^{-2}).

There was no statistical difference in TN concentration between the three treatments (Figure 4); however, TN was significantly altered by soil depth (0–10 cm: 0.78 ± 0.04 g kg^{-1}; 10–30 cm: 0.27 ± 0.02 g kg^{-1}; 30–60 cm: 0.23 ± 0.03 g kg^{-1}; 60–100 cm: 0.28 ± 0.02 g kg^{-1}) and significantly varied over time (June: 0.40 ± 0.04 g kg^{-1}; September: 0.38 ± 0.05 g kg^{-1}; December: 0.34 ± 0.04 g kg^{-1}; March: 0.44 ± 0.06 g kg^{-1}). Soil TN in the 0–10 cm increment contained a higher concentration of TN than any other depth ($p < 0.001$), but there were no significant differences between the deeper depths (i.e., 10–30, 30–60, and 60–100 cm). When integrated over the entire 1 m depth, the unharvested control (217.9 ± 21.5 g N m^{-2}) possessed significantly more TN that the bole-only harvest treatment (211.5 ± 14.7 g N m^{-2}) and whole-tree harvest + forest floor removal treatment (150.3 ± 14.7 g N m^{-2}).

Temporal variabilities in SOC and TN stocks were strongly correlated with concurrent changes in root biomass and forest floor mass; however, the extent to which either SOC or TN was correlated to root biomass or forest floor mass was depth dependent. Specifically, temporal variability in SOC and TN stocks in the 0–10 cm depth increment (for all treatments) was significantly correlated with temporal variability in forest floor mass (Table 3). In contrast, SOC and TN stocks in the 30–60 cm and 60–100 cm depth increments were correlated with changes in root biomass (Table 3).

The carbon to nitrogen ratio (C:N) of mineral soil decreased significantly with respect to depth (0–10 cm: 23.2 ± 0.6; 10–30 cm: 17.5 ± 0.8; 30–60 cm: 10.8 ± 0.7; 60–100 cm: 9.2 ± 0.3) when averaged across all stands and time (Figure 4); furthermore, all depths were statistically different from one another. C:N was not impacted by harvest treatments. The C:N ratio varied significantly with time; however, posthoc contrasts reveal that the only difference was between September 2014 (16.1 ± 1.1) and March 2015 (14.1 ± 1.1).

Table 3. Spearman's Correlation Analysis Among SOC and TN With Root Biomass, Forest Floor Mass, MBC, and MBN at Different Soil Depths for Each Harvest Treatment[a]

Harvest Treatment	Soil Depth (cm)		Spearman's Rho Roots (g m^{-2})	Forest Floor (g m^{-2})	MBC (µg C g^{-1})	MBN (µg N g^{-1})
Unharvested Control	0–10	SOC (g m^{-2})	0.37	0.84***	0.72**	0.46
		TN (g m^{-2})	0.18	0.64*	0.44	0.4
	10–30	SOC (g m^{-2})	−0.24	0.51	0.56	0.34
		TN (g m^{-2})	0.34	0.62*	0.80**	0.60*
	30–60	SOC (g m^{-2})	0.75**	0.4	0.19	0.13
		TN (g m^{-2})	0.71**	0.38	−0.03	0.08
	60–100	SOC (g m^{-2})	0.79**	0.18	0.17	0.43
		TN (g m^{-2})	0.64*	0.25	0.08	0.23
Bole-only Harvest	0–10	SOC (g m^{-2})	0.53	0.73**	0.15	−0.05
		TN (g m^{-2})	0.55	0.75**	0.29	0.1
	10–30	SOC (g m^{-2})	0.02	0.13	0.22	0.09
		TN (g m^{-2})	0.76**	0.59*	0.75**	0.65*
	30–60	SOC (g m^{-2})	0.83**	0.22	0.43	0.08
		TN (g m^{-2})	0.89***	0.4	0.64*	0.66*
	60–100	SOC (g m^{-2})	0.79**	−0.41	0.15	0.22
		TN (g m^{-2})	0.66*	−0.36	0.36	0.3
WT harvest + FF removal	0–10	SOC (g m^{-2})	0.1	0.58*	0.11	−0.03
		TN (g m^{-2})	0.01	0.67*	0.41	0.45
	10–30	SOC (g m^{-2})	0.1	0.3	0.55	0.31
		TN (g m^{-2})	0.29	0.54	0.70*	0.58*
	30–60	SOC (g m^{-2})	0.66*	0.06	0.2	0.29
		TN (g m^{-2})	0.63*	0.24	0.71*	0.57*
	60–100	SOC (g m^{-2})	0.71*	−0.35	0.23	0.3
		TN (g m^{-2})	0.71**	0.18	0.41	0.28

[a]Data from all treatments, soil depths, and sampling times were included in these calculations.
*$p < 0.05$.
**$p < 0.01$.
***$p < 0.001$.

3.4. Soil δ^{13}C and δ^{15}N Analysis

Bulk soil δ^{13}C values were not statistically impacted by harvest treatment or time; however, they became significantly more enriched with soil depth (0–10 cm: −27.8 ± 0.1‰; 10–30 cm: −25.9 ± 0.2‰; 30–60 cm: −23.8 ± 0.3‰; 60–100 cm: −23.1 ± 0.3‰) (Figure 5). Most soil depth increments varied from each other with the exception of the 30–60 cm increment not being statistically different from the 60–100 cm increment. When averaged across all soil depths and sample times, soil δ^{13}C values in the whole-tree harvest + forest floor removal stands (−25.7 ± 0.3‰) were more depleted than the bole-only harvest treatment (−24.9 ± 0.4‰) and the unharvested control treatments (−24.8 ± 0.3‰). Although the effect of organic matter removal intensity had no effect on δ^{13}C in the initial mixed model ANOVA, a posthoc contrast comparing the combined effects of the unharvested control treatment and the bole-only harvest treatment against the whole-tree harvest + forest floor removal treatment, we do observe a significant difference ($p < 0.01$).

No statistical differences due to harvest treatment were observed in soil δ^{15}N ($p = 0.06$); however, mean values varied significantly through time (June: 4.9 ± 0.3‰; September: 4.5 ± 0.3‰; December: 2.9 ± 0.3‰; March: 4.9 ± 0.3‰) and with depth (0–10 cm: 1.9 ± 0.2‰; 10–30 cm: 4.6 ± 0.2‰; 30–60 cm: 5.6 ± 0.3‰; 60–100 cm: 5.0 ± 0.2‰) (Figure 5). Soil δ^{15}N values across all depths and sampling times were most depleted in the unharvested control stands (mean: 3.7 ± 0.2‰). In contrast, whole-tree harvest + forest floor removal treatments (mean: 4.6 ± 0.3‰) were most enriched.

3.5. Microbial Biomass

When averaged across all soil depths and time points, microbial biomass carbon (MBC) was significantly higher in the unharvested control (113.7 ± 21.7 µg g^{-1}) and bole-only harvest treatment (103.5 ± 18.1 µg g^{-1}) than the whole-tree harvest + forest floor removal treatment (76.4 ± 14.9 µg g^{-1}); however, microbial biomass nitrogen (MBN) did not vary between harvest treatments. Mean MBC did vary with time
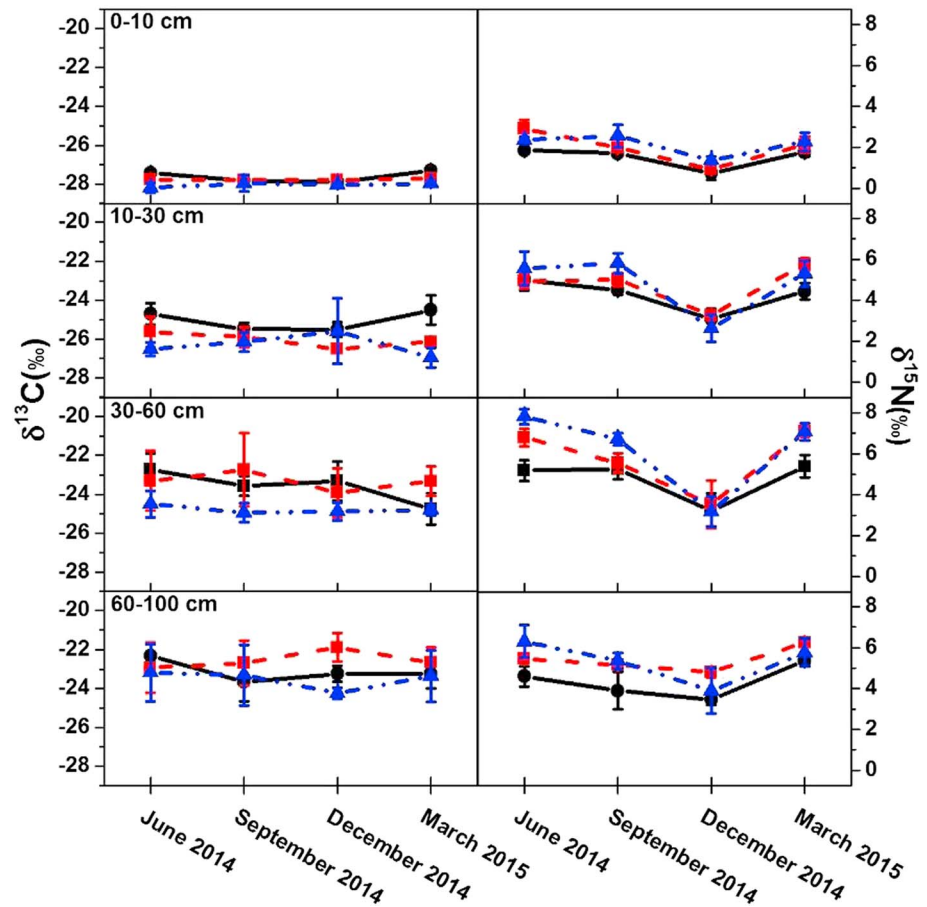
Figure 5. δ^{13}C of SOC and δ^{15}N of TN by treatment, separated into different depth increments and depicting seasonal trends. Each point is the mean ± standard error of three replicates. The symbols used for treatment differentiation: black circle: unharvested old-growth stands, black square: bole-only harvest stands, black triangle: whole-tree harvest + forest floor removal.

(June: 109.9 ± 23.7 µg g^{-1}; September: 68.7 ± 16 µg g^{-1}; December: 54.4 ± 12.8 µg g^{-1}; March: 158.5 ± 26.1 µg g^{-1}) and decreased drastically with depth (0–10 cm: 275.6 ± 22.6 µg g^{-1}; 10–30 cm: 64.2 ± 8.7 µg g^{-1}; 30–60 cm: 24.9 ± 3.9 µg g^{-1}; 60–100 cm: 26.7 ± 4.4 µg g^{-1}) (Figure 6). The same statistical differences were observed for MBN in regard to both time (June: 13.1 ± 2.7 µg g^{-1}; September: 4.4 ± 0.9 µg g^{-1}; December: 7.9 ± 1.9 µg g^{-1}; March: 19.8 ± 3.9 µg g^{-1}) and depth (0–10 cm: 32.6 ± 3.4 µg g^{-1}; 10–30 cm: 6.6 ± 0.9 µg g^{-1}; 30–60 cm: 3.1 ± 0.4 µg g^{-1}; 60–100 cm: 3.1 ± 0.4 µg g^{-1}). Roughly 70% of the total MBC and MBN (0–100 cm) was observed in the 0–10 cm depth increment. When averaged across all time points and depth increments, MBC in the unharvested control stands (113.7 ± 21.7 µg g^{-1}) was 9% higher than in the bole-only stands (103.5 ± 18.1 µg g^{-1}) and 39% more than the whole-tree harvest + forest floor removal stands (76.4 ± 14.9 µg g^{-1}). MBN was 8% lower in unharvested controls stands (11.6 ± 2.3 µg g^{-1}) when compared to bole-only treatment stands (12.6 ± 2.4 µg g^{-1}); however, the whole-tree harvest + forest floor removal stands (9.7 ± 2.4 µg g^{-1}) possessed 17% less MBN than the control stands.

Variations in MBC and MBN across all treatments and soil depths were correlated with both SOC (MBC: $r = 0.74$, $p < 0.001$; MBN: $r = 0.68$, $p < 0.001$) and TN (MBC: $r = 0.72$, $p < 0.001$; MBN: 0.66, $p < 0.001$). However, when MBC and MBN are analyzed by harvest intensity and depth, these variables are correlated primarily with TN in the 10–30 and 30–60 cm depth increments (Table 3). When we estimate the percent difference, based on samples from all treatments and depths, in MBC between December 2014 (lowest mean MBC) and March 2015 (highest mean MBC), we note an 80% difference that for the 0–10 cm compared to a 156%
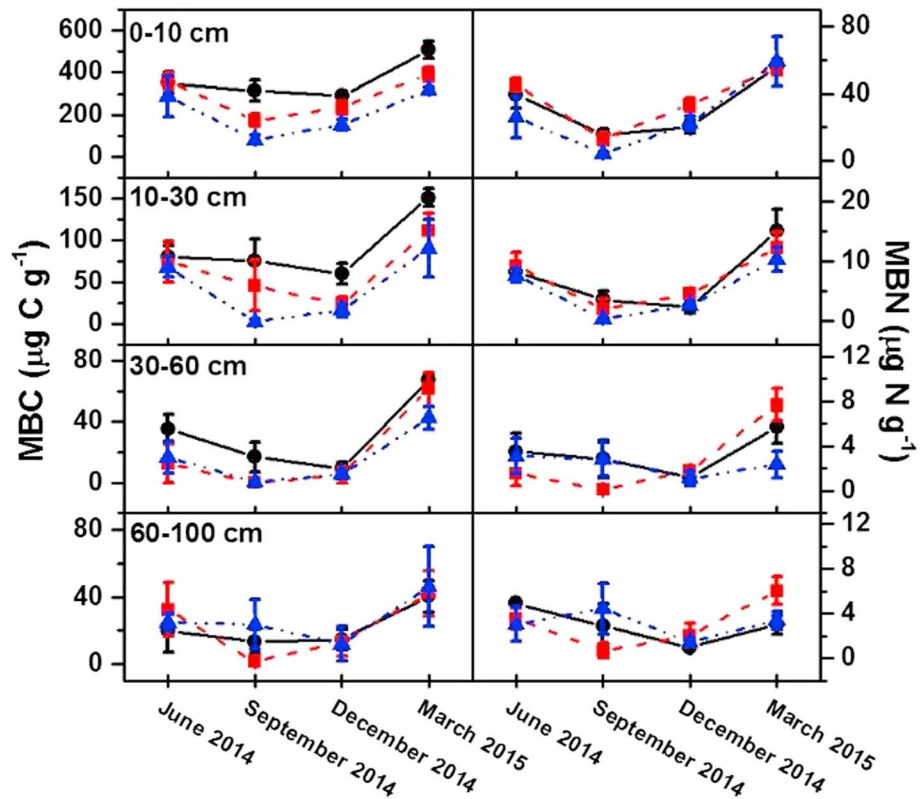
Figure 6. Microbial biomass carbon (MBC) and nitrogen (MBN) by treatment, separated into different depth increments and depicting seasonal trends. Each point is the mean ± standard error of three replicates. The symbols used for treatment differentiation: black circle: unharvested old-growth stands, black square: bole-only harvest stands, black triangle: whole-tree harvest + forest floor removal.

difference for the 30–60 cm increment. The other two depths fell in between these values; however, they both exceeded 100% difference. For each depth, MBC and MBN were correlated with soil water content (Table S2). The ratio of MBC to SOC (MBC/SOC) showed significant variation by time with the highest ratios, averaged from all treatments and soil depths, in March (2.9 ± 0.3) and the lowest in December (0.7 ± 0.1).

4. Discussion

4.1. Organic Matter Removal Influences Soil C and N Storage on Decadal Time Scale

The impact of differing levels of organic matter removal associated with timber harvest on long-term soil C and N storage and forest productivity has been of great interest globally as reflected in the numerous publications over a wide range of forest ecosystems [*Johnson and Curtis*, 2001; *Scott et al.*, 2004; *Powers et al.*, 2005; *Hansen et al.*, 2010; *Nave et al.*, 2010; *Slesak et al.*, 2011; *Thiffault et al.*, 2011; *Chen et al.*, 2013; *Scott et al.*, 2014; *Foote et al.*, 2015; *Achat et al.*, 2015a, 2015b]. However, the extent to which these differing harvest methods influence decadal-scale storage, in surface and subsurface mineral soils, has not been studied extensively, or has been inconclusive [*Jones et al.*, 2008; *Slesak et al.*, 2011]. This can be partially attributed to the expensive (monetarily and temporally) task of maintaining and sampling experimental sites.

Almost two decades after aboveground organic matter removal and replanting of *P. taeda*, SOC and TN in the upper 1 m of the profile remained lower in whole-tree harvest + forest floor removal compared to unharvested controls and bole-only harvest. SOC stocks, across all soil depths and sampling points, were 39% lower in the whole-tree harvest + forest floor removal treatment when compared to the unharvested control stands; congruently, the concentration of TN was 37% lower when comparing the same treatments. In the top 10 cm of the soil profile, SOC was 28% lower and TN was 24% lower. This is consistent with *Scott et al.* [2004] and *Foote et al.* [2015] in which measurements were taken at 5 years and 15 years post harvest,

respectively, using the same study area. Our results are also similar to *Mack et al.* [2014] who reported that the removal of forest floor material can lead to long-term (15 years) general reductions in mineral soil C and N at another site along the Gulf Coastal Plain. This long-term evidence suggests that when intensive organic matter removal is employed (i.e., whole-tree harvest + forest floor removal), diminished SOC and TN occur within the first 5 years of organic matter removal and can persist for decades, throughout the upper 1 m of the soil profile. In comparison, a 6% reduction in SOC and a 1% reduction in TN were observed when comparing bole-only harvest treatment to unharvested control treatment. The reduction in SOC is similar in scale as *Laiho et al.* [2003], *Smaill et al.* [2008], and *Huang et al.* [2011], and TN values are within ranges of studies by *Thiffault et al.* [2011], *Zummo and Friedland* [2011], *Prest et al.* [2014], and *Kellman et al.* [2014]. Our values are also consistent with a meta-analysis by *Achat et al.* [2015b] in which the effect of harvest intensity on soil organic matter and nutrient stocks was compiled from 140 articles and 168 experimental forest sites.

The decadal-scale decrease in SOC and TN in the whole-tree harvest + forest floor removal treatment may have multiple causes, some being categorized as initial ($<$5 years after harvest) and others as sustained (5+ years after harvest). The main initial decrease in SOC and TN may be attributed to a large portion of potential SOC and TN inputs being removed in the form of aboveground biomass and forest floor materials during organic matter removal, coupled with relatively low rates of above and belowground organic matter inputs from regrowing forest compared to older forest. This original disturbance would also theoretically allow for increased radiant energy reaching the soil surface as well as increased moisture infiltration. These conditions would result in a situation that is conducive for increased rates of C and N cycle processes. Sustained decreases in SOC and TN are likely driven by long-term soil organic matter destabilization arising from alterations in the biophysical conditions that influence the stability of soil organic matter (i.e., aggregation), especially in sandy soils. As mentioned previously, similar studies have placed emphasis on environmental controls such as temperature and moisture availability [*Bormann and Likens*, 1979; *Johansson et al.*, 1995; *Paul et al.*, 2003; *Kellman et al.*, 2014; *Solly et al.*, 2014] that regulate decomposition rates, SOC and TN transport, and organomineral interactions. In regard to the loss of SOC and TN at depth, it has been shown that changes in the structure of the forest floor can increase infiltration of labile organic matter to deeper depths, possibly creating a priming effect and increasing decomposition rates of deep roots and other forms of recalcitrant organic matter [*Fontaine et al.*, 2007; *Blagodatskaya and Kuzyakov*, 2008]. This priming effect may have occurred shortly after harvest leading to the observed reduction at year 5 [*Scott et al.*, 2004] and has since been unable to recover. The inability of SOC and TN in whole-tree harvest + forest floor removal stands to recover to preharvest conditions, even 18 years post harvest, illustrates the importance of C and N stores in the forest floor and slash residues in maintaining SOC and TN in surface and subsurface mineral soil post harvest.

Observed temporal variability in SOC and TN was correlated with concurrent variation in root biomass and forest floor mass. In the surface soil, variation in SOC and TN were significantly correlated with changes in forest floor mass, while in subsurface soils (i.e., 30–60 and 60–100 cm) those variables were instead correlated with changes in root biomass. This trend was consistent regardless of harvest treatment. Both root biomass and forest floor mass were largest in March. Previous studies have noted similar patterns [*Gill et al.*, 1999; *Wuest*, 2014]. These depth-dependent correlations suggest that forest floor inputs are strong determinants of SOC and TN in the surface soil, while root inputs are the important drivers of those pool sizes in deeper portions of the profile.

C:N ratios were not statistically impacted by differing timber harvest intensities and are similar to those reported by *Smaill et al.* [2008], *Diochon et al.* [2009], *Zummo and Friedland* [2011], and *Prest et al.* [2014]. This could be explained by proportional SOC and TN losses following timber harvest where increased rates of biogeochemical cycling following harvest result in C loss through increased heterotrophic respiration, while a proportional amount of N is simultaneously being mineralized, oxidized, and subsequently lost through leaching or volatilization.

4.2. Microbial Biomass Varies With Harvest Treatment, Time, and Depth

There were large reductions in MBC and MBN when the unharvested control treatment was compared to the whole-tree + forest floor removal treatment; however, only MBC was significantly different. The magnitude of MBC and MBN for the 0–10 cm depth increment was similar to those found in a North Carolina pine plantation [*Busse et al.*, 2006] and a boreal coniferous forest [*Wardle*, 1992]. MBC and MBN values obtained at the

Groveton-LTSP site 4 years earlier [*Foote et al.*, 2015] indicated statistically significant differences in MBC and MBN; however, this study only investigated the 0–10 cm depth increment. When we analyze only our 0–10 cm data, we also see a statistical difference in MBC and MBN between treatments; however, for MBN, this was not observed in any of the deeper increments which impacted the lack of significance in the overall model. We observed that roughly 30% of MBC and 28% of MBN in the upper 1 m of the soil profile occurred below 10 cm, which is similar in magnitude to *Fierer et al.* [2003], suggesting a high potential for biogeochemical activity at depth.

We initially hypothesized that MBC and MBN would show more pronounced seasonal variation in the surface soils versus deeper soil due to larger seasonal organic matter inputs that are concentrated in upper portions of the profile. However, we observed that seasonal fluctuations in deeper portions of the profile were comparable in magnitude to those found throughout the entire 1 m of the profile. Within any given soil depth increment and treatment, MBC and MBN varied by approximately two to eightfold across all time points in this 1 year study. This may be partly due to the availability and movement of pulses of substrate to deeper depths during the wetter, warm months, and reductions of those substrates during the drier colder months. In contrast, the reduced variability in the surface soil may be attributed to the more sustained availability of substrates year-round; however, variation is still observed. We generally observed more microbial biomass during the warmer months (i.e., June, September, and March) than December which is consistent with *Bååth and Söderström* [1982] who showed fungal biomass is highest in the warmer, summer months. Contrary to our observation of variation in MBC and MBN over time, both *Holmes and Zak* [1994] and *Blume et al.* [2002] note that microbial population size is generally stable over time. Also, our temporal pattern of microbial biomass differs from *Maithani et al.* [1996] in which the highest values of microbial biomass were observed in the winter. Many studies have suggested that in subtropical forest soils, soil moisture is the major controlling factor of microbial biomass [*Diaz-Raviná et al.*, 1995; *Yang et al.*, 2010]. Similar to *Yang et al.* [2010], we observed that seasonal dynamics in microbial biomass were correlated with variation in soil water content; both of these variables were highest in spring and lowest in the winter. The seasonal variations in soil moisture may be responsible for the variation in MBC and MBN at each depth, which is similar to studies of microbial biomass in other pine plantations [*Chen and Li*, 2003; *Yang et al.*, 2010].

The MBC/SOC ratio or microbial quotient has been widely used as an indicator of the changes in organic matter status due to alterations of soil conditions [*Sparling*, 1992]. Although no differences were observed for harvest treatment or depth, we used seasonal differences to evaluate trends in substrate availability and the proportion of total SOC immobilized in microbial biomass. The ratio of MBC/SOC was lowest in December and highest in March, suggesting a possible decrease in microbial immobilization during the winter months, which is consistent with reports from a Chinese pine plantation [*Yang et al.*, 2010].

4.3. Proposed Mechanisms of C and N Cycling Following Timber Harvest Inferred From Soil δ^{13}C and δ^{15}N

Following timber harvest, accelerated biogeochemical transformations may occur, resulting in altered stocks of SOC and TN [*Thiffault et al.*, 2011; *Achat et al.*, 2015a, 2015b]. These processes may leave behind isotopic signatures which can be used to infer the mechanisms behind the associated gains or losses [*Amundson et al.*, 2003; *Garten et al.*, 2007; *Templer et al.*, 2007; *Diochon and Kellman*, 2008; *Hobbie and Quimette*, 2009; *Craine et al.*, 2015]. Thus, δ^{13}C and δ^{15}N values may offer insights regarding forest ecosystem responses to organic matter removal during tree harvest events.

During the decomposition of soil organic matter, CO_2 released from soil due to heterotrophic respiration tends to be depleted in ^{13}C, while the stabilized residual soil organic matter is enriched in ^{13}C, relative to the original substrate [*Mary et al.*, 1992; *Santruckova et al.*, 2000]. With timber harvest having the potential to increase rates of decomposition, δ^{13}C values could be used to indicate the relative influence of different timber harvest intensities on heterotrophic activity. We hypothesized that following timber harvest, bulk soil C in the whole-tree harvest + forest floor removal stands would have higher δ^{13}C values due to higher rates of C-cycling and heterotrophic activity. Contrary to our hypothesis, soil δ^{13}C values were more depleted in the whole-tree + forest floor removal treatment, suggesting that more intensive organic matter removal methods may actually decrease rates of heterotrophic activity and carbon mineralization by reducing new and relatively labile organic matter inputs to the soil. Alternatively, in the whole-tree + forest floor removal

treatment, the replanted forest may be delivering new litter inputs to the soil that are more ^{13}C-depleted relative to the old forest floor material that was removed at the time of the harvest event. In forest ecosystems, δ^{13}C values generally increase in the order: leaves < fresh litter < older litter < soil [*Balesdent et al.*, 1993; *Garten et al.*, 2000]. Thus, removing the old forest floor materials and replacing it with more recent and more ^{13}C-depleted litter could potentially cause a reduction in soil δ^{13}C values.

As we predicted, bulk soil δ^{15}N values of surface and subsurface soils were consistently most depleted in the unharvested control stands and most enriched in the whole-tree + forest floor removal stands. Specifically, whole-tree harvest + forest floor removal and bole-only harvest stands were 0.9‰ and 0.8‰ more enriched in δ^{15}N, respectively, when compared to the unharvested control stands, over the entire 1 m profile. It is likely that the higher δ^{15}N values in the more severe organic matter removal treatments are at least in part attributable to higher rates of N losses due to acceleration of nitrification and denitrification that result in higher δ^{15}N values for the residual ecosystem N. Increases in solar radiation reaching the soil surface, decreases in transpiration and rainfall interception, and increases in the amount of precipitation reaching and infiltrating the forest floor and into the soil would favor higher rates of N losses during the time interval between harvest and stand recovery. Significant temporal differences were observed at all depth increments, with some depth increments producing greater than 50% reduction in enrichment when comparing summer 2014 to winter 2014. Winter tends to be the least biologically active season and can result in the accumulation of ^{14}N-enriched organic matter which would reduce δ^{15}N values. Our results are consistent with other studies that have shown that δ^{15}N values of soil total N can be increased for as long as several decades following tree harvesting [*Pardo and Nadelhoffer*, 2010; *Kellman et al.*, 2014].

5. Conclusions

In this study we report the long-term effect of intensive timber harvest practices on soil biogeochemical properties and processes. Specifically, we quantified SOC and TN stocks, microbial biomass, and soil δ^{13}C and δ^{15}N values in the upper 1 m of the soil profile for three treatments of increasing harvest intensity including an unharvested control treatment, a bole-only harvest treatment, and a whole-tree harvest + forest floor removal treatment. We found that 18 years after timber harvest, SOC and TN have not recovered to preharvest conditions in the upper 1 m of the soil profile. Furthermore, alterations in the natural abundance of δ^{13}C and δ^{15}N from bulk soil indicate that rates of key biogeochemical processes may be affected for a large portion of the mineral soil profile. The reduction in soil organic matter and alterations to the biogeochemical potential of this system may lead to diminished mineralization of limiting nutrients and potentially reduce the productivity of subsequent rotations. Our results suggest that harvest practices aimed at retaining the forest floor and residual slash should be employed to conserve SOC and TN to sustain the productivity and economic output of southeastern forestlands.

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