Nitrifying Community and N-cycle Activity are Altered by Increasing Timber Harvest Intensity in Surface and Subsurface Soils of the Western Gulf Coastal Plain

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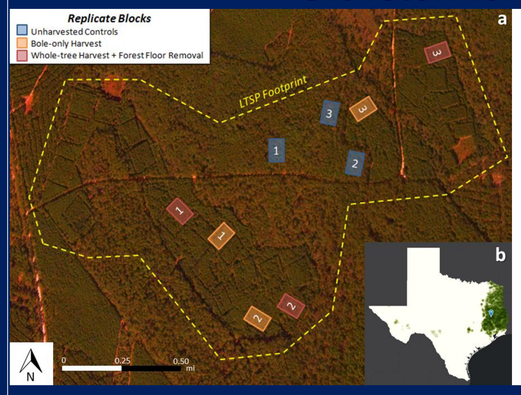
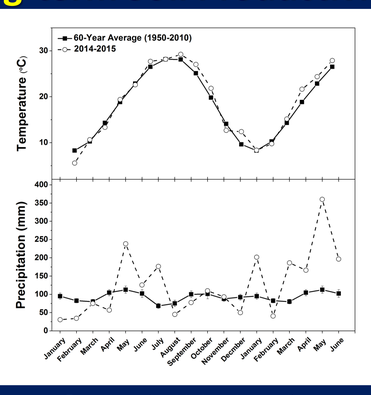
Introduction

Timber harvest has been shown to alter N-biogeochemistry through alterations in soil N stocks; however, soil pools of plant available N have not been quantified, especially in deeper horizons. Furthermore, microbes that mediate N-transformations are likely to be affected by increased harvest practices. The purpose of this study was to determine the effect of differing forest harvest intensities (i.e., no harvest, bole-only harvest, and whole-tree harvest + forest floor removal), 18-years post harvest, on microbial ecology associated with N-biogeochemistry. The Groveton, TX Long-term Soil Productivity site was utilized for this study We hypothesize that microbial biomass and gene copy number of *amoA* will be significantly reduced by increased harvest intensity resulting in smaller pools NO_3^-.

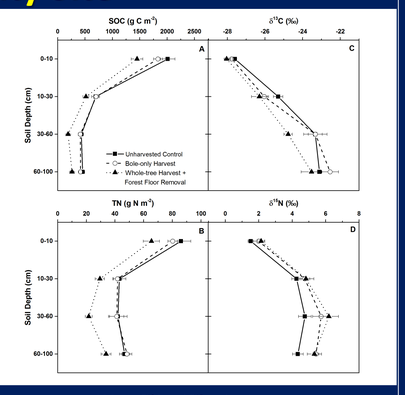
Groveton Long-term Soil Productivity Site



a) Groveton-LTSP plots. b) Extent of the pine forest ecosystem in Texas (green). Blue dot indicates the location of the Groveton-LTSP site.



Temperature and precipitation regimes over the course of 2014-2015 and the 60-year mean.



SOC, TN, $\delta^{13}C$, and $\delta^{15}N$ values in respect to treatment and soil depth. (n=12)

Harvest Method	*Description*
Unharvested Control	No harvest methods employed. Tree age: 55+ years
Bole-only Harvest	Tree boles were removed 18 years ago. Crowns, felled woody and herbaceous understory and forest floor retained
Whole-tree Harvest + Forest Floor Removal	All aboveground biomass was removed 18 years ago. Bare soil exposed

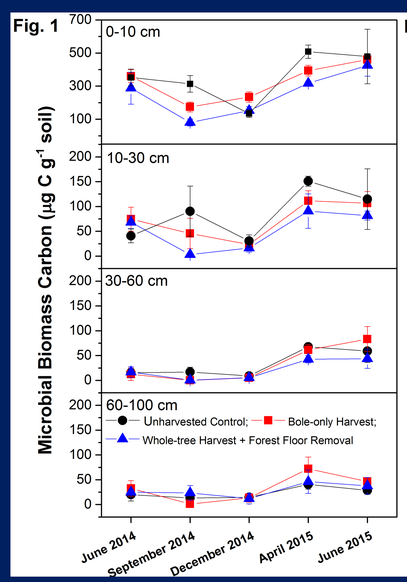
Soil Physicochemical Characteristics

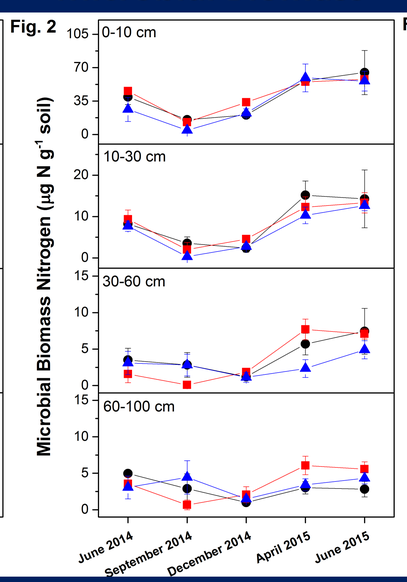
	Bulk Density (g cm^{-3})	Soil pH	Soil Texture
Soil Depth (cm)	*Averaged across all treatments & sampling points (n=45)*		
0-10	1.02 ± 0.03	4.05 ± 0.08	Sandy Loam
10-30	1.44 ± 0.03	3.96 ± 0.05	Sandy Loam
30-60	1.58 ± 0.03	3.76 ± 0.09	Sandy Loam
60-100	1.59 ± 0.03	3.34 ± 0.05	Sandy Clay Loam

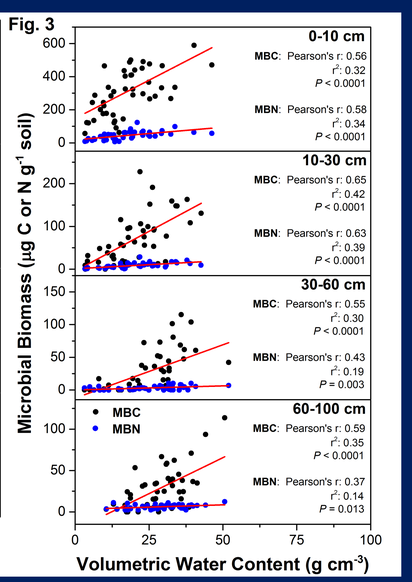
Methods

Soil was sampled to a depth of 1-meter, seasonally for five sequential time points. All analyses were carried out either in the Stable Isotopes for Biosphere Sciences Laboratory or the Soil and Aquatic Microbiology Laboratory, both located on the campus of Texas A&M University. Microbial Biomass C and N was extracted using the chloroform fumigation method and quantified on a Shimadzu TOC/TN Analyzer. NH_3/NH_4^+ and NO_3^- were extracted from soil using 2M KCl and analyzed on a Seal Analytical discrete chemistry analyzer. Copy number of archaeal and bacterial *amoA* were quantified on an Eppendorf Masterplex using SYBR florescent chemistry. Primers used: Arch-amoAF & Arch-amoAR (AOA); amoA-1F & amoA-2R (AOB).

Microbial Biomass







Microbial Biomass Carbon

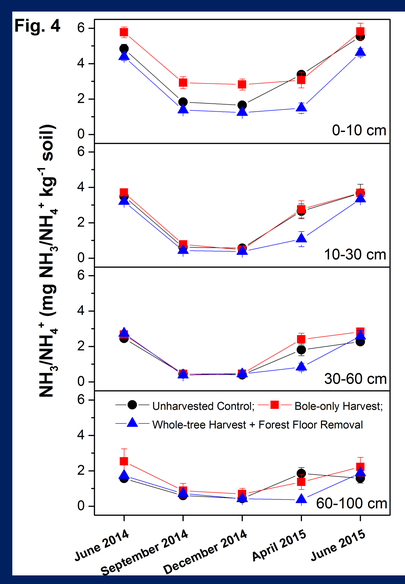
(µg C g^{-1} soil)	Harvest Method	Season	Harvest Method * Season
Soil Depth (cm)[a]		*p*-values[b]	
0-10a	0.0291*	<0.0001***	0.4889
10-30b	0.1565	<0.0001***	0.6659
30-60b	0.2537	<0.0001***	0.6300
60-100b	0.5037	<0.0001***	0.7679

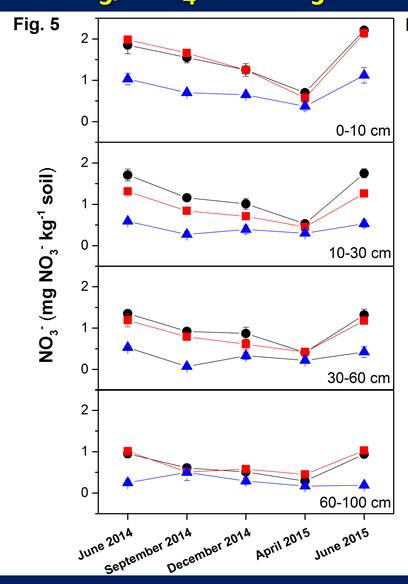
Microbial Biomass Nitrogen

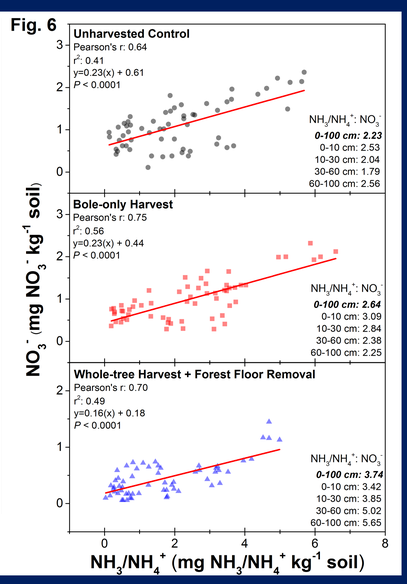
(µg N g^{-1} soil)	Harvest Method	Season	Harvest Method * Season
Soil Depth (cm)[a]		*p*-values[b]	
0-10a	0.4113	<0.0001***	0.8987
10-30b	0.4219	<0.0001***	0.9798
30-60b	0.3634	0.0003***	0.2577
60-100b	0.6771	0.0318*	0.1497

[a] Different letters indicate statistical difference (p<0.05) between soil depth (Tukey Kramer). [b] *p<0.05; **p<0.01; ***p<0.001

Soil NH_3/NH_4^+ & NO_3^- Pools







Soil NH_3/NH_4^+ Pool

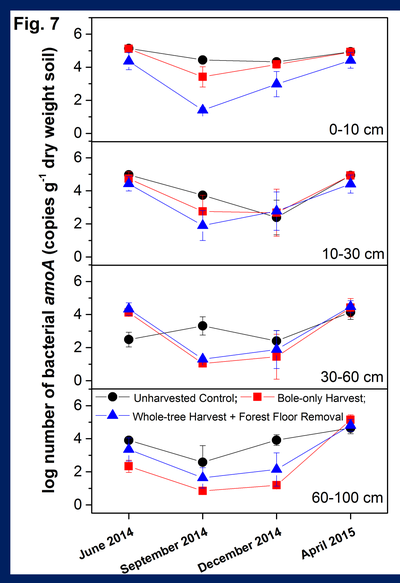
(mg NH_3/NH_4^+ kg^{-1} soil)	Harvest Method	Season	Harvest Method * Season
Soil Depth (cm)[a]		*p*-values[b]	
0-10a	<0.0001***	<0.0001***	0.1019
10-30b	0.0018**	<0.0001***	0.1013
30-60bc	0.0236*	<0.0001***	0.0102*
60-100c	0.0500	<0.0001***	0.1739

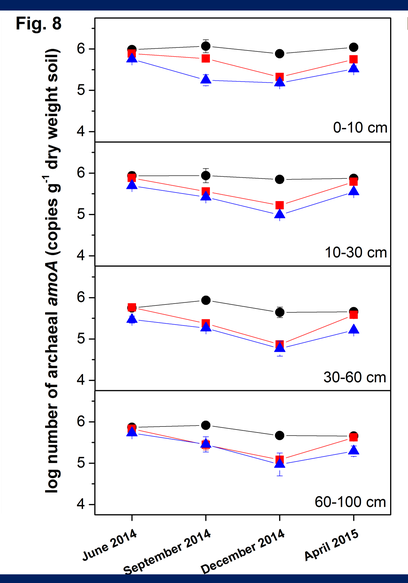
Soil NO_3^- Pool

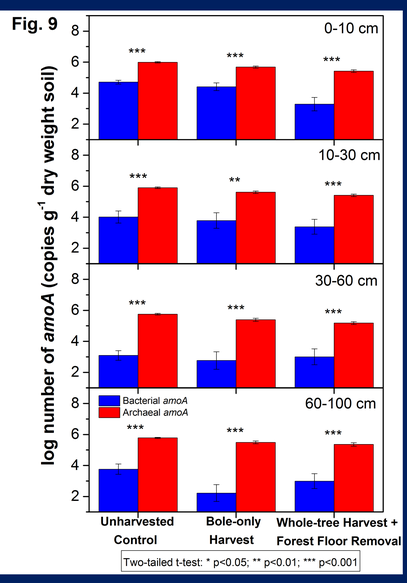
(mg NO_3^- kg^{-1} soil)	Harvest Method	Season	Harvest Method * Season
Soil Depth (cm)[a]		*p*-values[b]	
0-10a	<0.0001***	<0.0001***	0.0230*
10-30b	<0.0001***	<0.0001***	<0.0001***
30-60bc	<0.0001***	<0.0001***	0.0141*
60-100c	<0.0001***	<0.0001***	0.0007***

[a] Different letters indicate statistical difference (p<0.05) between soil depth (Tukey Kramer). [b] *p<0.05; **p<0.01; ***p<0.001

Bacterial & Archaeal *amoA* Quantification







Bacterial amoA

log (copies g^{-1} dry soil)	Harvest Method	Season	Harvest Method * Season
Soil Depth (cm)[a]		*p*-values[b]	
0-10a	<0.0001***	<0.0001***	0.0597
10-30ab	0.5064	0.0014**	0.8912
30-60b	0.7559	<0.0001***	0.0812
60-100b	0.0116*	<0.0001***	0.3060

Archaeal amoA

log (copies g^{-1} dry soil)	Harvest Method	Season	Harvest Method * Season
Soil Depth (cm)[a]		*p*-values[b]	
0-10a	<0.0001***	<0.0001***	0.0097**
10-30ab	<0.0001***	<0.0001***	0.0030**
30-60b	<0.0001***	<0.0001***	0.0005***
60-100ab	0.0001***	<0.0001***	0.1106

[a] Different letters indicate statistical difference (p<0.05) between soil depth (Tukey Kramer). [b] *p<0.05; **p<0.01; ***p<0.001

Conclusions and Future Directions

18-years post harvest, microbial biomass carbon and nitrogen were generally unaffected by increasing timber harvest intensity; however, NH_3/NH_4^+ pools, NO_3^- pools, and *amoA* copy number were significantly affected. Even though the relative biomass of the microbial community has generally stabilized, the quantity of archaeal and bacterial nitrifiers has been reduced by more intensive timber harvest methods, thus reducing the soil NO_3^- pool and possibly leading to the increased ratio of NH_3/NH_4^+ to NO_3^-. This not only has implications for current plant growth, but could affect the viability of future rotations. In regards to a broader microbial ecology perspective, this is one of the first studies to quantify ammonia oxidizers, in surface and subsurface soils, in the southeastern United States. Archaea have been implicated as having a greater impact on ammonia oxidation than bacteria, and our results indicate that this could be true, especially in acidic forest soils. Current efforts are underway to sequence bacterial and archaeal ammonia oxidizer communities from the Groveton-LTSP site as well as quantify their relative activity.