N_2O AND CH_4 EFFLUX FROM LOBLOLLY AND SLASH PINE PLANTATIONS: RESPONSE TO THROUGHFALL EXCLUSION BY FERTILIZER MANIPULATION, AND THINNING, FERTILIZATION AND HERBICIDE, PLANTING DENSITY, CULTURAL TREATMENTS

by

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(Under the Direction of Daniel Markewitz)

ABSTRACT

Nitrous oxide (N_2O) and methane (CH_4) are greenhouse gases that might be released or consumed in response to planting density, fertilization, vegetation control, or thinning of southern pine plantations. In a throughfall exclusion x fertilization manipulation, over two years, fertilization increased N_2O but not CH_4 efflux. Efflux of CH_4 was significantly decreased (increased consumption) in response to throughfall exclusion. A test of N fertilizers (urea, ammonium nitrate, or diammonium phosphate) also found significantly increased N_2O efflux from all forms. N_2O and CH_4 efflux were also measured over one year in three sites of a Lower Coastal Plain Culture x Density (CD) study and three sites of a Mid-Rotation Treatment (MRT) study. In CD there was no main effect of Culture or Density on N_2O or CH_4 efflux. In the MRT, where N fertilization had recently occurred, there was an effect of thin, herb, and fert on N_2O but not on CH_4.

INDEX WORDS: Loblolly Pine, Slash Pine, Nitrous Oxide, N_2O, Methane, CH_4, Fertilization, Greenhouse Gas Emissions, Silviculture

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CHAPTER 1

INTRODUCTION

Greenhouse gases (GHGs) of CO_2, N_2O, and CH_4 are increasing in the Earth's atmosphere and will likely impact the Earth's energy budget and global climate (Solomon et al. 2007). Globally, forests play a substantial role in GHG budgets thus understanding the cycles of GHGs through forested ecosystems is essential for understanding and mitigating global climate change. One third of the world's land surface is covered in forest, and these forests are both sources and sinks for CO_2, N_2O and CH_4 (Smith et al. 2003; Pilegaard et al. 2006; World Bank 2011). Compared to CO_2, the predominant GHG, CH_4 has a warming potential of 28-36 times over an approximately 100-year period and accounts for at least 30% of the Earth's energy balance (Lelieveld et al. 1998; USEPA 2014). Similarly, N_2O is estimated to have a global warming potential of 265-298 times of CO_2 over the same 100-year period (USEPA 2014) and is estimated to have a global net flux to the atmosphere of 12 x 10^{12} g N yr^{-1} (Ganzeveld et al. 2002). Of this global N_2O flux, ~35% is released to the atmosphere in response to fertilizer applications on cultivated fields (Shepherd et al. 1991). Nitrous oxide has also been shown to increase with nitrogen (N) fertilization within forest ecosystems (Shrestha et al. 2015). Relative to the numerous studies in agricultural systems (Smith et al. 2001; Bouwman et al. 2002; Akiyama et al. 2010; Rafique et al. 2011), however, the role of fertilizer use and GHG emissions has been far less extensively researched in forested ecosystems.

In this thesis I focus specifically on the fluxes of N_2O and CH_4 from managed forest plantations that are most likely to receive fertilizer inputs as well as other silvicultural treatments (i.e., site preparation, various initial planting densities, herbicide applications, mid-rotation thinning). In the southeastern United States about 87 million hectares are forestland, which includes 10 million hectares of naturally regenerated pine and about 14 million hectares of pine plantations (Zhao et al. 2016). Loblolly pine (*Pinus taeda* L.) and slash pine (*Pinus elliottii*) are common plantation species native to the southeastern USA, growing from eastern Texas to Florida, and as far north as Maryland (Cunningham et al. 1990).

Given a large demand for industrial roundwood (~110 million dry tons annually) throughout the southeastern USA, there has been increasing amounts of silvicultural treatments within managed loblolly pine plantations (Fox et al. 2007). For example, nitrogen-rich fertilizers, of which urea (CH_4N_2O) is the most common, are applied to pine plantations 1 to 3 times during a 25-to 30-year rotation with an accumulations of ~500 kg N ha^{-1} (Allen et al. 1990). Other fertilizers such as diammonium phosphate (DAP; $(NH_4)_2HPO_4$) and ammonium nitrate (NH_4NO_3) are also used to increase the amount of N and P within these ecosystems (Fox et al. 2007a). The application of fertilizers typically increases loblolly pine growth. For example, in one study across the southeastern USA that included 23 installations, fertilized stands grew an additional ~4 m^3 ha^{-1} $year^{-1}$, on average over an 8-year period (Carlson et al. 2014). Although this enhanced growth increases CO_2 sequestration from the atmosphere, N fertilization also increases N_2O release to the atmosphere. In a slash pine (*Pinus elliottii*) plantation in Florida, for example, N fertilization with 180 kg ha^{-1} of urea-N increased N_2O efflux by 6-800 times (12-74 ug N_2O-N m^2 h^{-1}) compared to a control (Castro et al., 1994). As such, there is continued interest in understanding conditions affecting N_2O efflux under managed pine.

In addition to effecting N_2O efflux in pine plantations, N fertilization has also been shown to alter CH_4 efflux (Castro et al., 1994; Shrestha et al., 2014). Methane exchange between the soil and atmosphere is regulated by the balance between methanogenesis, the production of CH_4 (Aronson et al. 2013), and methanotrophy, the consumption of CH_4 (Schlesinger and Bernhardt 2013). Under anaerobic conditions, methanogenic bacteria consume organic matter releasing CH_4, as such many freshwater wetlands are a known source of CH_4 to the atmosphere (Bridgham et al 2013). In contrast, well-drained upland forest soils have methanotrophic bacteria that consume CH_4 and at a global scale upland forest soils are an important CH_4 sink (Dutaur and Verchot, 2007). Upland forest soils, however, have also been found to be a CH_4 source during high rainfall events or within wet seasons (Shrestha et al. 2015). Water-filled pore space, N-fertilization of soil, and soil temperature all play key roles in the regulation of CH_4 uptake or emission from forest soils (Shrestha et al. 2015). In a review, 10 of 12 forest soil studies found a 5-95% inhibition of N fertilizers on CH_4 uptake, although only one of these studies was within southeastern pine (Shrestra et al., 2015). In one tropical forest, the addition of N during a dry period stimulated soil CH_4 uptake (Veldkamp et al. 2003). In southeastern USA pine plantations N fertilization may interact strongly with soil moisture conditions to convert sites to a stronger CH_4 sink or source.

Silvicultural practices such as the initial planting density, application of fertilizers, competing vegetation suppression (herbicide), and thinning all impact the edaphic conditions of the stands as well as the growth of trees (Allen, 2001). High planting density (>3000 trees ha^{-1}), for example, increases early (<5 yr) stand productivity and canopy closure increasing demand on soil water and nutrients (Zhao et al. 2011). In contrast, a decrease in the amount of competitors through herbicide application within these stands allows for the crop trees to gain more access to water, nutrients, and light. In the same way, the act of thinning reduces intraspecies competition belowground and also allows for more light into the canopy. Thinning is an important practice in plantation management that produces significant changes within the growth dynamics of stands (Short and Burkhart 1992; Zhang et al. 1997; Sharma et al. 2006). The interactions of these silvicultural practices and fertilization on fluxes of N_2O and CH_4 have not been well-investigated.

Similar to the effects of silvicultural practices on edaphic condition, changes in climate may also have an important impact on soil temperature and moisture and thus important interactions on N_2O and CH_4 efflux (Potter et al., 1996). Increasing soil temperature from -5 to 10ºC was related to increasing CH_4 consumption in red pine and mixed hardwood forests of the northeast USA while increasing water filled pore space in these same forests limited CH_4 consumption (Castro et al. 1995). Increasing water filled pore space was also positively correlated with N_2O emissions across a range of tropical forests (Davidson et al. 2000). In the southeastern USA mean annual temperature is expected to rise and summertime precipitation may decline by 10 to 30% (Meehl et al. 2007, Solomon et al. 2007). The impact of these climatic changes and the interaction with silvicultural management is of current interest in the region (Will et al., 2015).

The prominence of silviculture including fertilization in southeastern USA pine management (Fox et al. 2007b) and the potential for changing climate has created a need for better quantification of N_2O and CH_4 emissions from ecosystems. In Chapter II of this thesis I focus on N_2O and CH_4 efflux under a fertilization x throughfall exclusion experiment within a loblolly pine plantation in Taliaferro County, GA. This is a multi-year field study with quarterly measures of GHG efflux. To capture short-term N_2O efflux response to N fertilization I also conducted a 1-year experiment within another loblolly pine plantation applying three different commonly used fertilizers (urea, ammonium nitrate, and diammonium phosphate). The measurements in this study included 7 dates in the first 90 days to a final measurement at one year. In Chapter III, I quantify the N_2O and CH_4 efflux within two different silvicultural studies of the Plantation Management Research Cooperative both within the Lower Coastal Plain in Georgia and Florida. The Culture x Density (CD) study included three initial loblolly pine planting densities under two levels of fertilization and herbicide treatments. The Mid-Rotation Treatment (MRT) study included a thinning treatment, thinning with herbicide, thinning with fertilizer, and thinning with fertilizer and herbicide in slash pine plantations. In both studies efflux measurements were made twice under wet soil conditions (Feb. 2015, Mar. 2016) and twice under drier conditions (June 2015, June 2016). In Chapter IV I briefly discuss the importance of this research.

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CHAPTER II

N_2O AND CH_4 EFFLUX WITHIN A LOBLOLLY PINE (*Pinus taeda* L.) PLANTATION: RESPONSE TO THROUGHFALL EXCLUSION AND FERTILIZATION MANIPULATION

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Abstract

Globally, forests play a substantial role in greenhouse gas budgets and can be both sources and sinks of GHGs, including nitrous oxide (N_2O) and methane (CH_4). N_2O has been shown to increase with N fertilization in forest ecosystems and forest fertilization is particularly prevalent in Southeast USA loblolly pine plantations. Presently, there is also concern about changing climate in this region (i.e., reduced summer rainfall) and thus interest in the interaction of fertilization with changing climate. This study investigated N_2O and CH_4 efflux in a throughfall exclusion x fertilization manipulation study in a Georgia loblolly pine plantation with samples taken quarterly from 2012-2015. We also quantified N_2O efflux response among common fertilizers of urea, ammonium nitrate, and diammonium phosphate. N_2O efflux increased ($p<0.001$) in response to fertilization but not throughfall exclusion while CH_4 efflux did not respond to fertilization but declined ($p=0.057$) in response to throughfall exclusion. In the test of N fertilizer forms all increased N_2O efflux ($p < 0.001$) within the first 6 weeks but no response was measureable at 1 year. Nitrogen fertilization of loblolly pine plantations increases N_2O efflux, but only for a short duration, while reduced precipitation could lead to an increase in CH_4 and N_2O sinks.

Keywords: Loblolly pine, CH_4, N_2O, fertilization, throughfall exclusion, urea, NH_4NO_3, DAP

Introduction

Globally, forests play a substantial role in GHG budgets thus understanding the cycles of GHGs through forested ecosystems is essential for understanding and mitigating global climate change. One third of the world's land surface is covered in forest, and forests can be both sources and sinks of GHGs, including nitrous oxide (N_2O) and methane (CH_4; Smith et al. 2003; Pilegaard et al. 2006; World Bank 2011). Methane has a global warming potential 28-36 times that of CO_2 over an approximately 100-year period and accounts for at least 30% of the Earth's energy balance (Lelieveld et al. 1998; USEPA 2014). N_2O is estimated to have a global warming potential of 265-298 times that of CO_2 over the same period (USEPA 2014). Nitrous oxide also has a longer half-life than CH_4 and thus remains in the atmosphere longer. Although industrial factories are the main contributor to GHG emissions, agriculture is responsible for approximately 7% of the U.S. GHG emissions (Parton et al. 2011). In 2012, CH_4 accounted for 9% and N_2O for 6% of all United States GHG emissions, with 75% of the total attributed to agricultural soil management (USEPA 2014).

Emissions of GHGs from agricultural fertilization are a recognized and growing concern in regulating global warming potential. It is estimated that on average an additional 10 kg N_2O-N is emitted to the atmosphere for every 1,000 kg of N rich fertilizer applied to agricultural soils (Soloman et al. 2007). Many agricultural studies have investigated the role of fertilizer use in GHG emissions in row crops (Smith et al. 2001; Bouwman et al. 2002; Akiyama et al. 2010; Rafique et al. 2011) but research on GHG emissions from forested systems is less extensive. In a few cases, N_2O has been shown to increase with N fertilization in forest ecosystems (Shrestha et al. 2015) with unfertilized forests producing 20-500% less N_2O than fertilized forests (Shrestha et al. 2014).

Forest fertilization is particularly prevalent in the southeastern USA where between 1969 and 2004 fertilized plantation area increased to over 6 million hectares (Albaugh et al. 2007). Loblolly pine (*Pinus taeda* L.) is the most prevalent plantation pine species in the Southeast and is native to the region, growing from parts of Texas into Florida, and as far north as Maryland (Cunningham et al. 1990). Loblolly pine productivity over the last sixty years has increased due to silvicultural practices, including fertilization (Fox et al. 2007b). Nitrogen-rich fertilizers, of which urea (CH_4N_2O) is the most common, are applied to pine plantations 1 to 3 times during a 25 to 30-year rotation at rates of ~50-225 kg N ha^{-1} per application (Allen et al. 1990). Diammonium phosphate (DAP; $(NH_4)_2HPO_4$) and ammonium nitrate (NH_4NO_3) are two other types of fertilizers that are used to increase the amount of N and P within these ecosystems (Fox et al. 2007a). In one study across the southeastern USA that included 23 installations, on average over an 8-year period fertilized stands grew an additional ~4 m^3 ha^{-1} $year^{-1}$ (Carlson et al. 2014).

Increased stand growth from N-rich fertilization can remove additional CO_2 from the atmosphere but increased N_2O efflux can partially offset this GHG benefit (Shrestha et al. 2014). Nitrous oxide exchange between soil and atmosphere is partly regulated by nitrification and denitrification from microbial activity (Schlesinger and Bernhardt 2013). Increasing readily available plant N can increase microbially-mediated nitrification and denitrification, thus increasing N_2O emissions from forests (Zhixin et al. 2014). Microbial activity increases not only with N addition (Phillips et al. 2009) but also with soil drying and rewetting (Davidson et al 1991b; Davidson et al. 1993; Hartley and Schlesinger 2000; Beare et al. 2009; Kim et al. 2012). Moisture facilitates the development of anaerobic microsites where microbial-mediated denitrification can occur (Inselsbacher et al. 2010). For example, the experimental addition of water with N in a steppe pasture of China was shown to increase soil N_2O emissions within 2-3 days from 11 fold at a low N application rate to 165 fold at a high N application rate (Xinchao et al. 2015). In contrast, a decline in rainfall as predicted in the southeastern USA driven by climate change (Sánchez-Lugo 2015; Voes et al. 2015) could result in soil becoming a greater N_2O sink (Inselsbacher et al. 2010), as moist microsites decrease in number (Butterbach-Bahl et al. 2013). A study in the Amazonian rainforest determined that experimental reduction in throughfall (i.e., simulated drought) lowered N_2O annual emission by 33% and increased the amount of CH_4 uptake by a factor of three (Davidson et al. 2008).

As demonstrated in the Amazon, southeastern USA pine plantations might also convert to a stronger CH_4 sink under predicted climate change scenarios. Methane exchange between soil and atmosphere is regulated by the balance between methanogenesis (Aronson et al. 2013) and methanotrophy (Schlesinger and Bernhardt 2013), with methanotrophy occurring under the aerobic conditions likely to increase in the absence of precipitation. Globally about 5% of the total removal of atmospheric CH_4 is taken up by upland soils (Reeburg 2003), although upland soils have also been found to be a CH_4 source during high rainfall events or within wet seasons (Keller and Reiners; 1994, Shrestha et al. 2015). Water filled pore space, N-fertilization of soil, and soil temperature play key roles in the regulation of CH_4 uptake and emission (Shrestha et al. 2015). The addition of N to a tropical forest ecosystem during a dry period was found to stimulate soil CH_4 uptake (Veldkamp et al. 2003).

The prominence of fertilization in pine plantation management (Fox et al. 2007b) and the potential for changing precipitation patterns in the southeastern USA requires investigation of this interaction on N_2O and CH_4 emission. The objective of this study was to quantify N_2O and CH_4 efflux under a throughfall exclusion x fertilization experiment in a multi-year field study within a loblolly pine plantation. As a secondary objective, we quantify short-term N_2O efflux among three fertilizers (urea, NH_4NO_3, and DAP) within a small-scale loblolly pine plantation study. Our hypothesis is that fertilization in the absence of throughfall exclusion (i.e. in the presence of higher soil moisture) will have the greatest N_2O and CH_4 efflux. Also, we predicted that as seen with the application of urea to agricultural systems (Kissel et al. 2009), forest fertilization with urea will have the greatest short-term N_2O efflux.

Methods

Site description and sample design

Two planted loblolly pine (*Pinus taeda* L.) stands located in Taliaferro and Clarke counties within the Piedmont Region of Georgia were utilized for this study (Figure 1). The Taliaferro County site (33°37’32.61”N / 82°47’56.54”W) was used for the throughfall exclusion x fertilization manipulation and one of four sites used for a larger regional collaborative research project (see Will et al. 2015 for a full description of the PINEMAP project). The plantation in Whitehall Forest in Clark County (33°88’67.85”N / 83°35’66.57”W) was used to test short-term response of N_2O flux to different fertilizer types.

Throughfall Exclusion x Fertilization Experiment

The Taliaferro County plantation was established by Plum Creek in 2006. Trees were spaced at 2.1 m within the row and 3.5 m between the rows. The soils of the site are a Lloyd-Cecil complex (Kanhapludults), which are very deep and well drained. In 2012, four blocks were established based on existing tree height and basal area and four 21 x 14 m treatment plots were established within each block with a 12-m buffer zone between the plots. Treatments included:

control, fertilization, throughfall exclusion, and throughfall exclusion and fertilization (Samuelson et al. 2014). The throughfall exclusion treatments, installed in May 2012, included a rigid plastic guttering system held up by a wood frame (about 1.8 meters in height) to divert 30% of throughfall from the plots (Samuelson et al. 2014). Pelletized fertilizer consisting of approximately 224 kg N ha^{-1}, 28 kg P ha^{-1}, and 56 kg K ha^{-1} was applied in March 2012. An early spring fertilization was selected to take advantage of saturated soil conditions and to avoid application during periods of heavy precipitation.

N Fertilizer Form Experiment

Whitehall Forest is a 300-ha property in Clarke County managed by the University of Georgia. The pine plantation used within Whitehall Forest was planted in 1996 with trees planted 2 m apart within the rows and 3 m between the rows for a density of ~1450 trees ha^{-1}. Soils consist of Cecil and Pacolet series; both Fine, kaolinitic, thermic Typic Kanahpludults. The site was previously fertilized with 140 kg ha^{-1} DAP (18-21% N) and 432 kg ha^{-1} urea (45-46% N) in the spring of 2000.

Two plots (1 x 1 m) per treatment were randomly assigned in each of three blocks (12.60 x 5.90 m) and treated with different fertilizers in May 2013. Fertilizer treatments consisted of urea (109 kg N ha^{-1}), DAP (124 kg N ha^{-1}), and ammonium nitrate (115 kg N ha^{-1}), all compared to a control (no fertilizer). The total amount of N differed slightly due to the dissolution of solid fertilizer forms. Given the small plot size, pelletized fertilizer was dissolved into deionized water and applied from a backpack sprayer at the above rates, which are comparable to rates used in pine forest management applications (Allen et al. 1990).

Forest floor and mineral soil C and N concentration

In Taliaferro County, forest floor and mineral soil samples were collected within the large treatment plots pre-fertilization. In Whitehall Forest, pre-fertilization soil samples were taken inside the blocks but outside of the small treatment plots. In Taliaferro County, a 35 x 35 cm square wooden frame was used to delineate the subplots while in Whitehall Forest a 1 x 1 m frame was utilized. In each location the frame was randomly placed on the forest floor and forest floor material was removed and separated by O_i (litter) and O_e + O_a (duff). Surface mineral soil was augered after forest floor removal from 0-10 cm using a 6 cm diameter open bucket auger. In Taliaferro County a total of 8 samples were collected and composited into two samples per plot. In Whitehall a single sample was collected in each block. Organic horizons were oven dried (65^{o}C) for 72 hours, passed through a Wiley mill (Thomas Scientific, Swedesboro, NJ, USA) with a 2-mm sieve, and then pulverized using a ball mill grinder. The mineral soil samples were air-dried, sieved through a 2-mm sieve, and similarly pulverized. All samples were analyzed for total C and N concentration using a Thermo Flash 2000 (Waltham, MA, USA). The soil samples from both locations were analyzed for pH within the 0-10 cm using 10g soil to 20 ml of DI water. Data are reported in Table 2.1.

N_2O and CH_4 collection and analysis

Static chambers (Hutchinson and Livingston 2002) consisting of 15.24 cm (diameter) x 25.4 cm (height) PVC collars were used to collect gas samples. On sampling dates, static chamber collars were covered with PVC caps configured with a collection septum and a ventilation port to allow equalization between internal and external atmospheric pressures. In Taliaferro County four chambers were randomly distributed within the plots and collars were installed 24 hours before sampling. In Whitehall Forest, collars were placed in the center of 1 x 1 meter plots and remained in the plots for the duration of the experiment. During sampling, caps remained on the collars until sample completion. Samples were extracted using a 10 mL syringe and transferred into pre-evacuated, glass vials (Exetainers, Labco Inc., UK) at 0, 10, 30, 50, and 70 minutes. In Taliaferro County, sampling started in March 2012 and continued approximately quarterly through February 2015. In Whitehall Forest, gas measurements began July 9, 2013 a week before fertilization and proceeded on days 1, 3, 7, 14, 21, 35, and 365 after fertilization.

Samples (1 ml) for the Taliaferro County and Whitehall sites were initially analyzed for N_2O on a Shimadzu GC-14A gas chromatograph using an electron capture detector (ECD) (Shimadzu Corporation GC-14A, Kyoto, Japan) until September 2013 when a Shimadzu GC-2014 was acquired. From November 2013 to February 2015 N_2O and CH_4 were analyzed with an electron capture detector and flame ionization detector (FID), respectively. The carrier gas was Nitrogen (280 kPa) and flow within the column was 24.31 ml min^{-1}. Compressed air and hydrogen gas was the make up for the FID. The ECD make up consisted of a P-5 (95% Argon and 5% Methane) gas mixture.

Precipitation

Precipitation records were obtained from the University of Georgia Weather Network (georgiaweather.net). Using six locations within 90 km of each site, total precipitation input was summed for the week preceding each gas sample collection date.

2.5 Data Analysis

To estimate rates of gas efflux, the slope of linear regressions between measured concentrations and time of collection (i.e., $\frac{\Delta C}{\Delta t}$) were calculated. All data were initially graphed to visually identify any outliers that may weaken the linear model; values that were outside the 95% confidence level were considered outliers and were removed from the data set (<10% of the data). To convert slopes to efflux the following equation was used:

$$F = \left(\frac{\Delta C}{\Delta t}\right)\left(\frac{V}{A}\right)\kappa \qquad \text{Eq 1}$$

where F is the gas flux (mass of gas m^{-2} d^{-1}), $\Delta C/\Delta t$ is the slope that defines the rate that the gas (N_2O or CH_4) concentration changes (μL L^{-1} min^{-1}), V is the volume inside the chamber (m^3), A is the soil surface area covered by the chamber (m^2), and κ is a conversion factor converting volume of gas to mass (temperature corrected ideal gas law) and minutes to day. The regressions were run in R v3.2.3 (R Core Team 2016).

Gas fluxes were analyzed using a repeated measures mixed model in SAS 9.4 (Proc MIXED; SAS Institute Inc.). ANOVA assumptions were evaluated. The data from the N fertilization manipulation in the Whitehall Forest were natural log transformed and retained an underlying autoregressive correlation matrix, so date was treated as a repeated measure. The N_2O and CH_4 throughfall exclusion by fertilization manipulation data were assessed in a 2-factor ANOVA employing an underlying autoregressive correlation matrix. The pre-fertilization data were excluded from the statically tests for this study because not all plots were measured on this date. *Post hoc* comparisons between treatment level effects applied a t-test with non-pooled standard deviation; the False Discovery Rate adjustment minimized Type I errors without the stringent conditions found in family-wise comparisons, making this a more powerful comparison (R Core Team 2016; Benjamini and Hochberg 1995; Benjamini and Yekutieli 2001). The p-values that are <0.05 were considered statically significant.

Results

Throughfall exclusion x fertilization manipulation

The N_2O efflux was significantly ($p<0.001$) increased by fertilization (Table 2.2) with overall mean efflux from unfertilized plots being -0.011 mg N_2O-N m^{-2} d^{-1} and fertilized plots being 0.088 mg N_2O-N m^{-2} d^{-1} (Figure 2.2a). There was no significant effect of exclusion or fertilization by exclusion interaction (Table 2.2). Day of efflux measurement was significant ($p<0.001$) and there was a day by fertilization interaction significant at $p=0.1$. The interaction reflected a difference between non-fertilized and fertilized plots earlier in the experiment that declined with time (Figure 2.2a and 2.2b). The N_2O efflux rates over all treatments and dates ranged from -0.95 – 3.61 mg N_2O-N m^{-2} d^{-1}. Measurements within the control treatment varied as much as 3-fold (April 2012 to June 2012, Figure 2.2a) but generally were low and positive (<0.06 mg N_2O-N m^{-2} d^{-1}) (Figure 2.2a).

Methane efflux was not affected by fertilization (Table 2.2) but did decline (increased CH_4 consumption) in response to throughfall exclusion ($p=0.057$). The non-exclusion treatments mean was -0.034 mg CH_4–C m^{-2} d^{-1} while that for exclusion treatments was -0.161 mg CH_4–C m^{-2} d^{-1}. There was no interaction between fertilization and throughfall exclusion. Day of measurement was significant ($p<0.001$) as CH_4 efflux increased and decreased over the years but there was no interaction between day and fertilization or throughfall exclusion. The CH_4 efflux over all treatments and dates ranged from -1.95 to 2.41 mg CH_4–C m^{-2} d^{-1}. For the first 14 months of the study, the soil was a CH_4 sink (Figure 2.3). Between September 2014 and November 2014 when cumulative precipitation increased 8-fold, all treatments switched to a source (Figure 2.3).

N fertilizer forms

All treatments (i.e., forms of fertilizer) resulted in an increase in N_2O efflux compared to the control (Table 2.2; Figure 2.4). There was also a significant date and date by treatment interaction (Table 2.2), which reflected a decline in N_2O efflux after the first six dates of measurement in the first six weeks after fertilization. In the final measurement at 1 year there was no difference among treatments (Figure 2.4). The N_2O flux from all treatments ranged from -0.337 to 5.62 mg N_2O-N m^{-2} d^{-1}. Treatment averages over all dates where 0.128, 0.515, 0.586, and 0.546 mg N_2O-N m^{-2} d^{-1} for control, DAP, NH_4NO_3, and urea, respectively. On the final date of measurement, approximately one year after fertilization, the average across all treatments for the date was 0.026 mg N_2O-N m^{-2} d^{-1} (Figure 2.4).

Discussion

Nitrogen fertilization

One consistent result from across both the Throughfall Exclusion x Fertilization (Figure 2.2a) and N Fertilizer Form (Figure 2.4) experiments was an increase in N_2O efflux with N fertilization (Table 2.2). Fertilization from any source (i.e., urea, NH_4NO_3, or DAP) resulted in significantly higher N_2O efflux than the unfertilized treatments. Contrary to our hypothesis, however, efflux from urea was not statistically greater than that from the other fertilizer types. Our Whitehall Forest site had not received any fertilizer inputs for 13 years prior to our experiment and upon fertilization with N-rich sources resulted in as much as a 16% increase in instantaneous N_2O efflux rates.

These results are consistent with previous research in agricultural (Mosier et al., 1998) and forest (Shrestha et al., 2015) ecosystems as well as a few studies in southern pine plantations (Castro et al., 1994; Shrestha et al., 2014). In these other southern pine studies, fertilization was with urea (or coated urea) and efflux rates increased from 4 to 600-fold above controls. Based on the hole-in-the-pipe model (Davidson et al., 1993) that identifies N_2O "leaking" from the pipe either during nitrification or denitrification, these increased N_2O efflux rates after application suggest that either nitrifying or denitrifying bacteria may have been N-limited. Southern pine plantations often have limited nitrification or net N mineralization, partly as a response to low soil pH (Table 2.1), but these rates have been shown to increase with N fertilization (Gurlevik et al., 2004). Loblolly pine plantation managers may apply fertilizer, typically after canopy closure, every 4-6 years at roughly100 to 200 kg N ha^{-1} (Fox et. al 2007a) so release of N_2O may occur periodically over the rotation.

In the throughfall exclusion x fertilization study, there was no significant response in CH_4 efflux to fertilization (Figure 2.3; Table 2.2). This result is contrary to our hypothesis and to much previous research showing an inhibitory effect of N fertilization on CH_4 efflux such that CH_4 consumption declines (Stuedler et al., 1989; Castro et al., 1994; Shrestha et al., 2015). In our study site, which is a well-drained, upland soil, but with high clay (>40%) contents in the B_t horizon (Qi 2016), CH_4 efflux was mostly negative indicating consumption at this site (Figure 2.3). It is not clear why N inhibition of methanotrophy was not observed, although measurement of CH_4 efflux was delayed until 18 months after fertilization (Mar 2012 to Oct 2013). As such, it is quite likely the N availability had declined by the time of first measurement. This speculation is somewhat supported by lack of an N_2O response at these measurement times.

Throughfall Exclusion

Although CH_4 efflux did not respond to fertilization, CH_4 efflux did decline (i.e., greater consumption) under throughfall exclusion (Table 2.2). Increased CH_4 consumption was also observed in a throughfall exclusion study in a tropical rainforest (Davidson et al., 2008). Increased efflux into the soil is driven by greater rates of gas diffusion as soils dry.

The present study also found that the throughfall exclusion treatment alone was often associated with a negative N_2O efflux, suggesting that the soil can serve as a N_2O sink (Schlesinger 2013), although N_2O efflux was not significantly altered by throughfall exclusion. Investigation of N_2O efflux under different soil drainage classes (i.e., poorly drained, moderately drained, and well-drained) in southeastern pine clearly demonstrated greater efflux with poorer drainage (Shrestha et al., 2014). Comparisons between N_2O efflux and local precipitation measurements at our site showed greater efflux measurements in non-throughfall exclusion associated with greater precipitation during the week prior to sampling (Table 2.3); a result consistent with studies that have found a positive correlation between soil moisture and N_2O efflux in forested systems (Beare et al. 2009; Kim et al. 2012; Buchkina et al. 2013; Shrestha et al. 2014; Liu et al. 2015). It is possible that the occasional periods of high precipitation activate formation of anaerobic microsites (Zhixin, et al. 2014) that facilitate denitrification. Other studies have also indicated that the effects of N_2O emissions from N fertilizers have been triggered by rainfall events and high soil moisture (Borken and Beese 2005; Zona et al. 2013).

Our work suggests that loblolly pine plantations will become an N_2O source only if fertilizer is applied. If the climate predictions within the southeastern USA are correct, however, in that rainfall will decline during the summer growing season (Sánchez-Lugo 2015; Vose et al 2015), forest soils could provide both N_2O and CH_4 storage from the atmosphere (Inselsbacher et al. 2010). As such, these ecosystems could serve as a GHG sink if left unfertilized.

Conclusion

Nitrogen fertilization of loblolly pine plantations increases N_2O efflux, but only for a short duration. Furthermore, urea, the most common fertilizer used within loblolly pine plantations, releases the equivalent amount of N_2O as other common fertilizers (NH_4NO_3 and DAP). Throughfall exclusion decreased CH_4 efflux (i.e., increased consumption) but not N_2O efflux. Both N_2O and CH_4 efflux were responsive to precipitation inputs becoming sources during wetter periods. Nitrogen fertilization will increase N_2O efflux while lack of precipitation could lead to an increase in CH_4 and N_2O sinks.

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Table 2.1 Mean pre-fertilization soil C and N concentration and pH in the Throughfall Exclusion x Fertilization experiment (N=16 plots) in Taliaferro County, GA in 2012 and the N Fertilizer Form experiment (N=3 blocks) in Clark County, GA in 2013.

Treatment	Soil Layer	C	SE	N	SE	C:N	pH_{water}
		%		%			
Throughfall Exclusion x Fertilization	Oi	41.7	3.94	0.83	0.33	58.6	
	Oe + Oa	20.3	2.88	1.73	1.40	13.7	
	0-10cm	1.77	0.41	0.09	0.02	23.7	5.46
N-Fertilizer Form							
	Oi	46.2		0.91		59.5	
	Oe + Oa	23.9		0.91		30.8	
	0-10cm	2.38		0.15		18.0	4.33

Table 2.2. Gas efflux ANOVA summary statistics from the Throughfall Exclusion by Fertilizer and N-Fertilizer Form experiments. Values shown in bold are statistically significant.

	F-Stat	P-value
Throughfall Exclusion x Fertilization		
(n=4)		
Nitrous Oxide		
Fertilization	11.85	**<0.001**
Throughfall Exclusion	0.01	0.911
Fert x TFE	1.20	0.274
Date	6.17	**<0.001**
Date x Fertilization	1.61	0.100
Date x TFE	0.23	0.994
Methane		
Fertilization	0.30	0.583
Throughfall Exclusion	3.65	**0.057**
Fert x TFE	2.11	0.147
Date	9.65	**<0.001**
Date x Fertilization	1.40	0.225
Date x TFE	0.54	0.745
Nitrogen Fertilizer Forms		
(n=3)		
Nitrous Oxide		
Treatment	12.53	**<0.001**
Date	6.69	**<0.001**
Date x Trt	2.06	**0.011**

Table 2.3. Pearson's correlation coefficient between cumulative precipitation one week prior to gas sampling and N_2O efflux for each of the four treatments.

Treatment	Pearson's Correlation
Control	0.25
Throughfall Exclusion	-0.46
Fertilizer	0.46
TFE x Fert	0.10

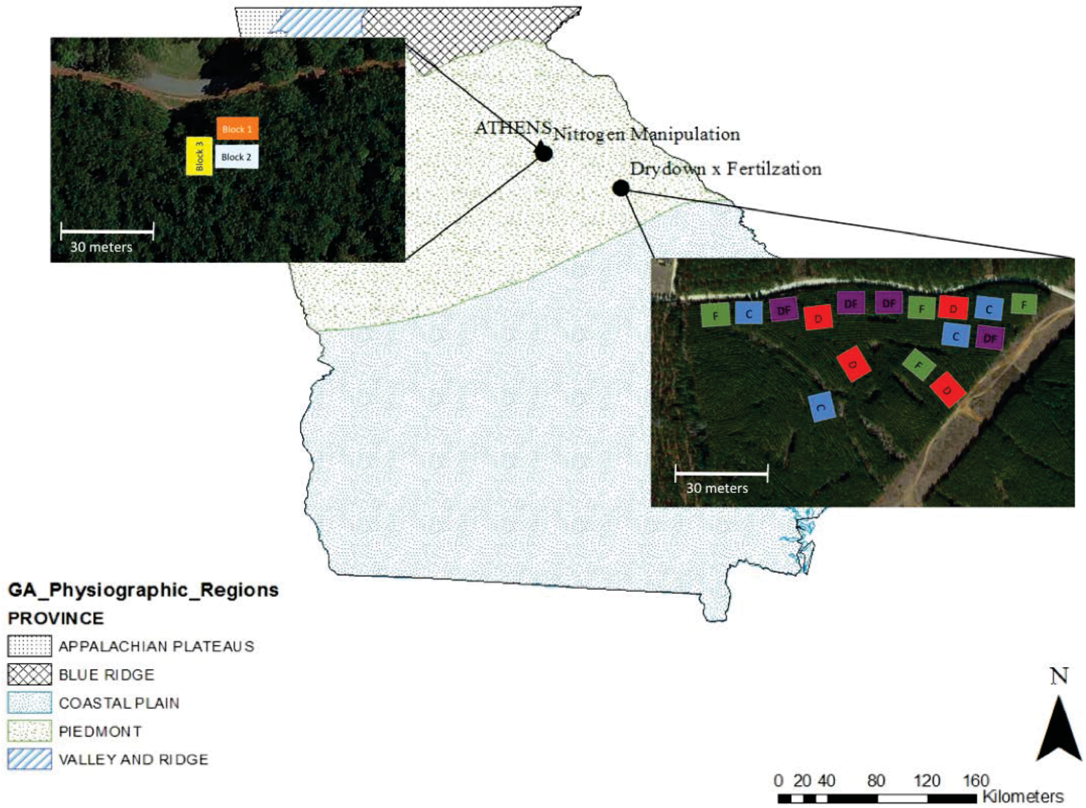

Figure 2.1. Location of study sites within Georgia Piedmont. The left inset is the Fertilizer Form experiment site with three blocks. The inset on the right is the throughfall exclusion x fertilization site that contained four blocks of four treatments: Control-C, Fertilizer-F, Throughfall exclusion- D, and Throughfall exclusion x Fertilization- DF.

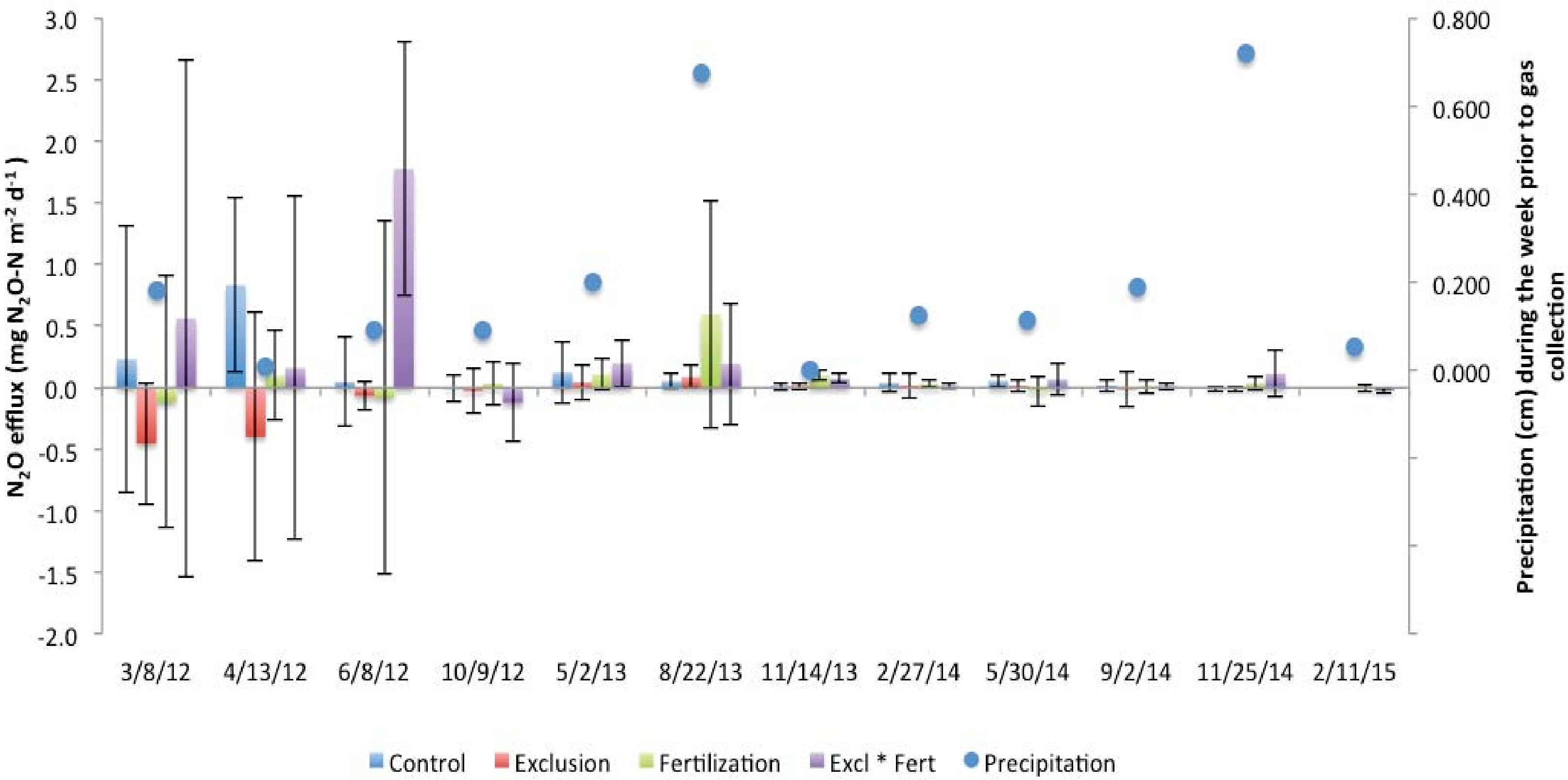

Figure 2.2a. N_2O emissions over time by throughfall exclusion and fertilizer treatments in Taliaferro County, GA. Precipitation data during the week preceding sample collection were gathered from the Georgia weather website (georgiaweather.net), using records averaged for 6 stations found within 90 km of the site. Fertilizers were applied in March 2012 and throughfall exclusion began in May 2012.

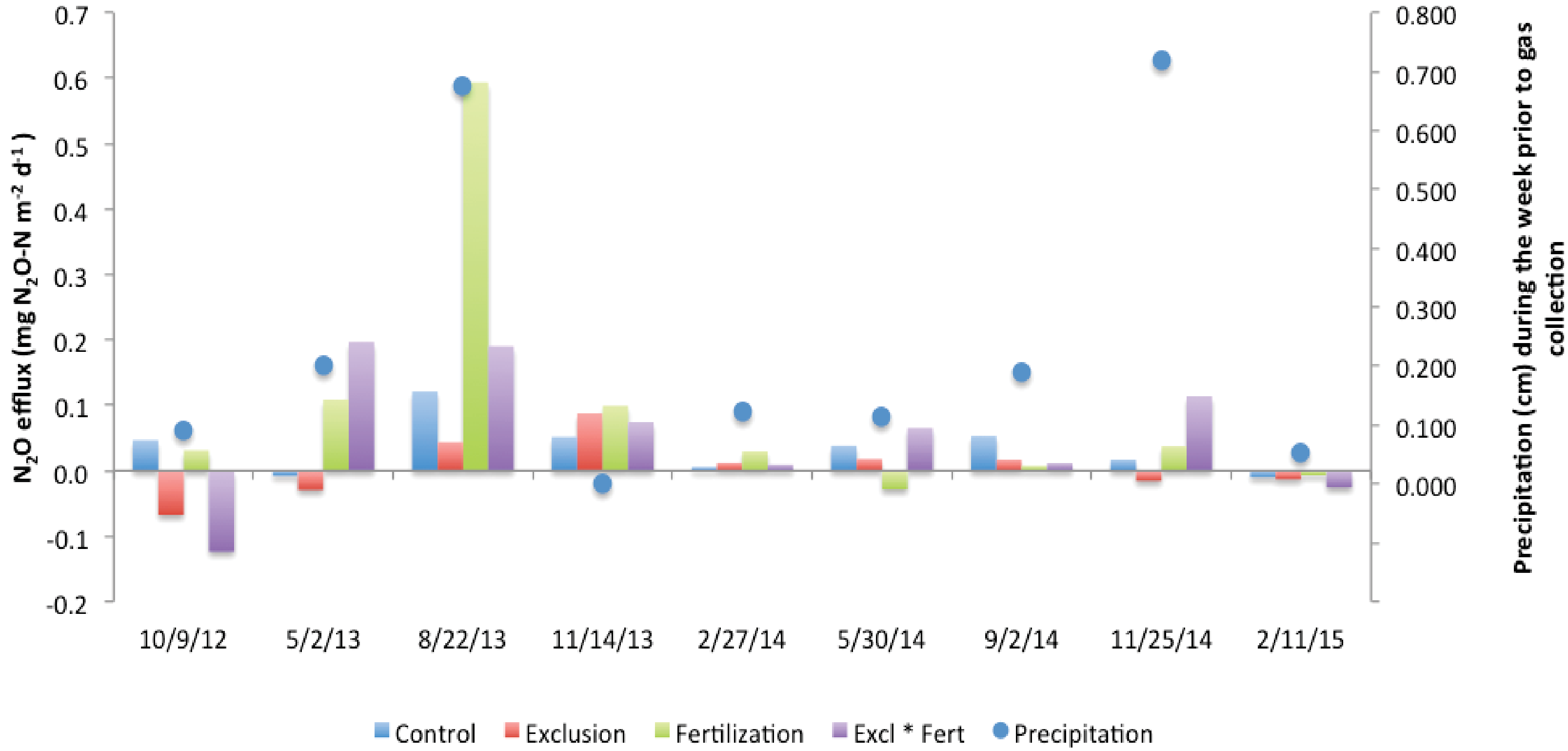

Figure 2.2b. (inset of Figure 2a to increase resolution after June 2012) N_2O emissions over time by throughfall exclusion and fertilizer treatments in Taliaferro County, GA. Precipitation data during the week preceding sample collection were gathered from the Georgia weather website (georgiaweather.net), using records averaged for 6 stations found within 90 km of the site. Fertilizers were applied in March 2012 and throughfall exclusion began in May 2012.

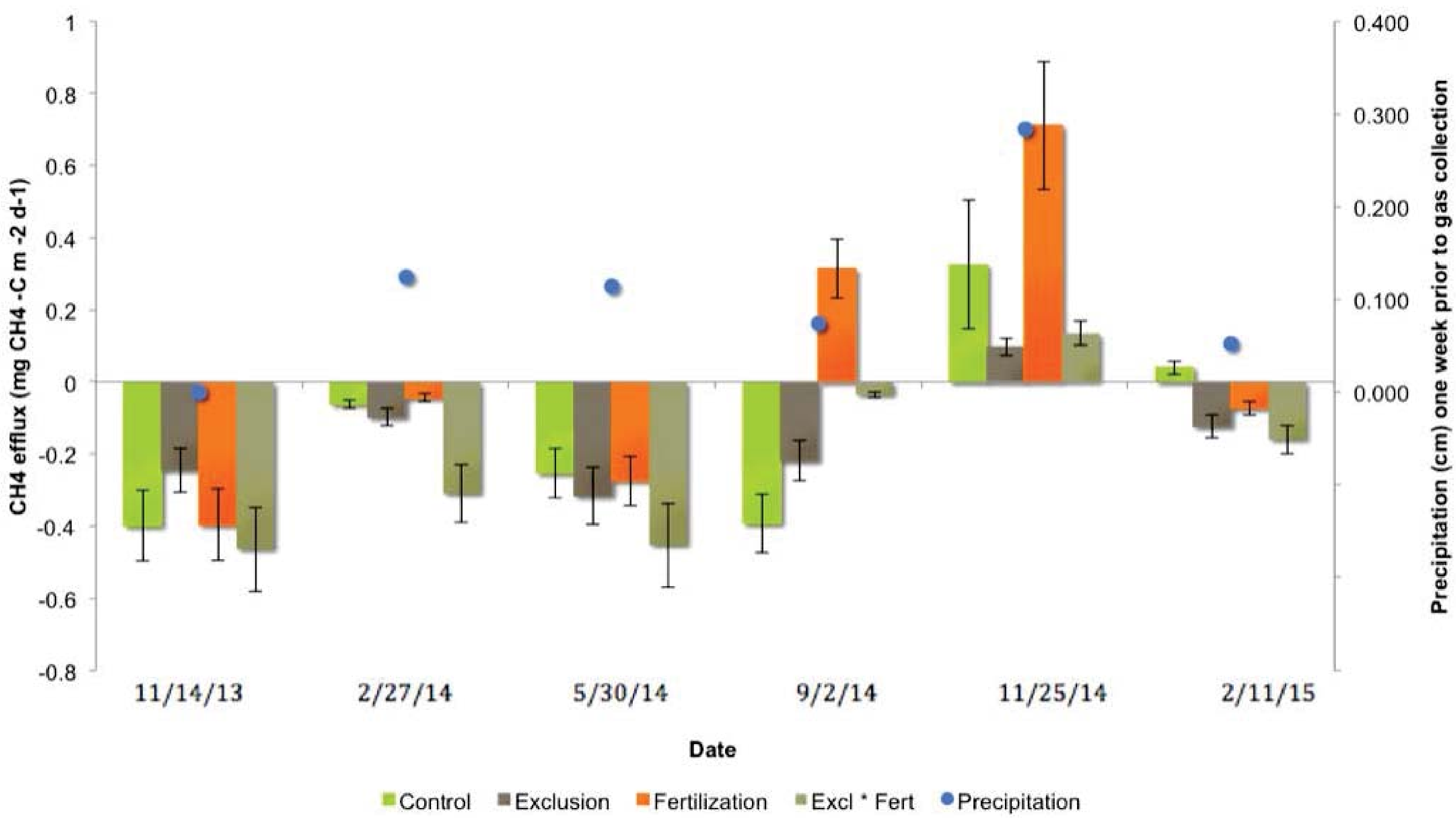

Figure 2.3. CH_4 emissions over time by throughfall exclusion and fertilizer treatments in Taliaferro County, GA. Precipitation data during the week preceding sample collection were gathered from the Georgia weather website (georgiaweather.net), using records for 6 stations found within 90 km of the site. Fertilizers were applied in March 2012 and throughfall exclusion began in May 2012.

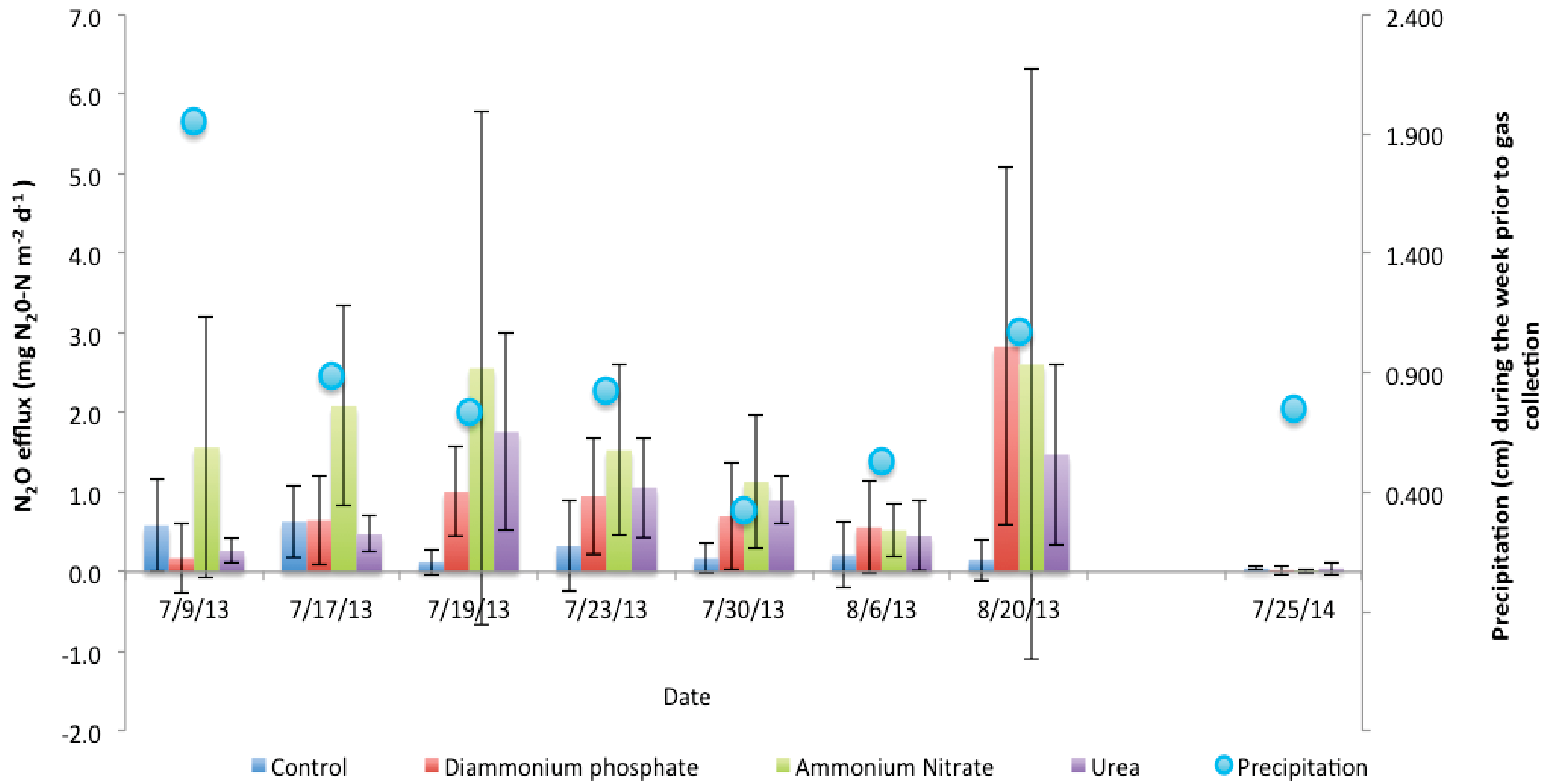

Figure 2.4. N_2O efflux following fertilization with three fertilizer types at the nitrogen fertilizer form experiment (Clark County, GA Whitehall Forest). Precipitation data were gathered from the Georgia weather website (georgiaweather.net) within 90 km of the site. Cumulative precipitation was averaged from 6 stations for the 7 days preceding each sampling event. Fertilizers were applied July 16, 2013.

CHAPTER III

N_2O AND CH_4 EFFLUX WITHIN LOBLOLLY AND SLASH PINE PLANTATIONS: RESPONSE TO PLANTING DENSITY, CULTURE, THINNING, FERTILIZATION, AND HERBICIDE TREATMENTS

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Abstract

Nitrous oxide (N_2O) and methane (CH_4) are greenhouse gases that can be released or consumed in response to silvicultural management of southern pine plantations. Nitrogen fertilization can enhance release of N_2O while planting density, vegetation control, or thinning may alter CH_4 efflux due to changes in site edaphic conditions. The amount of N_2O and CH_4 efflux in plantations is important for understanding the impact of wood and bioenergy production on atmospheric warming potential. N_2O and CH_4 effluxes were measured over one year in three sites of a loblolly pine Culture x Density (CD) study and in three sites of a slash pine Mid-Rotation Treatment (MRT) study both in the Lower Coastal Plain of Georgia and Florida. Sampling using a static chamber approach began in spring 2015 and continued until the summer of 2016. The results indicate that in the CD study neither the operational or intensive levels of culture (both had fertilizers and herbicide) or initial planting densities altered N_2O or CH_4 efflux. In the MRT study there was a significant main effect of thinning, herbicide, and fertilization on N_2O efflux with all activities increasing efflux. Average efflux from the thin-only plots (0.023 mg N_2O-N m^{-2} d^{-1}) was 8-fold less than the thin+herb+fert plots (0.164 mg N_2O-N m^{-2} d^{-1}). There was, however, no significant effect on CH_4 efflux. Under both experiments, methane efflux was largely negative indicating CH_4 consumption within these plantations. Extractable NH_4-N was correlated with N_2O efflux suggesting that any silvicultural activities that increase soil N cycling may have a potential to increase N_2O efflux.

Keywords: Loblolly, Slash, CH_4, N_2O, soil temperature, extractable ammonium, volumetric water content (VWC)

Introduction

Forests play a large role in the regulation of greenhouse gases (GHG), including nitrous oxide (N_2O) and methane (CH_4) (Smith et al. 2003; Pilegaard et al. 2006; World Bank 2011). N_2O is estimated to have a global warming potential of 265-298 times that of CO_2 over a 100-year period (USEPA 2014) and is estimated to have a global net flux to the atmosphere of 12 x 10^{12} g N yr^{-1} (Ganzeveld et al. 2002). Of this global N_2O flux, ~35% is released to the atmosphere in response to fertilizer applications on cultivated fields (Shepherd et al. 1991). CH_4 has a global warming potential of 28-36 times that of CO_2 over the same 100-year period and accounts for at least 30% of the Earth's energy balance (Lelieveld et al. 1998; USEPA 2014). In the global CH_4 budget, forest soils provide an important biotic sink for atmospheric CH_4 (Dutaur and Verchot, 2007). Understanding how silvicultural treatments such as fertilization, thinning, and harvest of managed plantation forests may alter N_2O and CH_4 efflux is necessary in estimating the net GHG benefit of wood and bioenergy production from forest and in managing the role of forests in a changing climate.

Agricultural studies have investigated the roles of fertilizer use and GHG emissions in crops (Smith et al. 2001; Bouwman et al. 2002; Akiyama et al. 2010; Rafique et al. 2011), but the amount of research in forested systems is less extensive. Loblolly (*Pinus taeda* L.) and slash (*Pinus elliottii*) pine plantations are intensively managed forests that cover ~14 million ha in the Southeast, USA. A large demand for industrial roundwood in this region has led to increasing silvicultural practices such as site preparation, competing vegetation suppression (i.e., herbicides), the application of fertilizers, and thinning, which have all led to a large increase in productivity over the last sixty years (Allen et al., 2001; Fox et al. 2007b). Although this enhanced growth increases CO_2 sequestration from the atmosphere, N fertilization or other silvicultural activities may increase N_2O or CH_4 release to the atmosphere partially offsetting this GHG benefit (Castro et al., 1994; Stuedler et al., 1989).

The addition of nitrogen (N) to pine plantations has been shown to increase N_2O emissions (Castro et al., 1994; Papen et al. 2001; Shrestha et al., 2014). In a 26-yr-old slash pine plantation on poorly drained soil in Florida, for example, N fertilization with 180 kg ha^{-1} of urea-N increased N_2O efflux by 6-800 times (12-74 ug N_2O-N m^2 h^{-1}) compared to a control (Castro et al., 1994). Increase in N_2O efflux with N fertilization has generally been a consistent result in N-limited forest soils (Shrestha et al., 2015). In N-rich sites response may differ, such as one study in an 80-yr old mixed deciduous forest in Pennsylvania where fertilization with 100 kg N ha^{-1} yr^{-1} in the form of NH_4NO_3 for 6 years did not increase N_2O efflux (Bowden et al. 2000).

The addition of N fertilization also impacts CH_4 efflux. In the slash pine plantation noted above, CH_4 uptake was 5-20 times lower than the control in response to N fertilization (Castro et al. 1994). Inhibition of CH_4 uptake by N fertilization has been demonstrated in a range of forest ecosystems (Stuedler et al., 1989; Shrestha et al., 2015), including the N-rich, deciduous forest in Pennsylvania where in the five-month period following fertilization a 24% reduction was observed in total CH_4 uptake (Bowden et al., 2000).

Nitrous oxide and CH_4 efflux response to silvicultural treatments other than fertilization has been rarely studied. These other silvicultural activities, however, can impact edaphic factors such as N concentration, soil temperature, or soil moisture that influence efflux. For example, thinning is an important silvicultural practice in the southeastern USA that has been shown to produce significant changes in the growth of stands (Short and Burkhart 1992; Zhang et al. 1997; Sharma et al. 2006). Thinning allows more light to penetrate the canopy thus allowing for the soil to increase in temperature (Selig et al., 2008). Changes in soil temperature can directly impact efflux as demonstrated in a 66-yr old red pine (*Pinus resinosa*) stand, where CH_4 uptake increased from 0 to 0.12 mg CH_4-C m^2 ha^{-1} as temperatures increased from -5 to 10ºC (Castro et al. 1995). Relationships among soil temperature, moisture, and soil N have also been described for N_2O efflux (Davidson et al., 2000). Generally, increasing soil temperature and soil N availability increase N_2O efflux while increasing soil moisture can decrease gas diffusion limiting positive or negative efflux. Although many studies have looked at how different silvicultural practices affect tree growth (e.g., Zhao et al. 2011) investigations on trace gas fluxes are lacking.

Concern over the role of forest management on N_2O and CH_4 efflux in regards to mitigating GHGs and in understanding the net balance of GHG uptake and release during timber bioenergy feedstock production has prompted the following research on planting density, cultural intensity, thinning, herbicide, and fertilizer impacts. Two studies established by the Plantation Management Research Cooperative (PMRC) at the University of Georgia are utilized: the Culture x Density (CD) within loblolly pine and Mid-Rotation Treatment (MRT) within slash pine. Both studies utilize multiple sites within the Lower Coastal Plain of the southeastern USA. The objective is to quantify the effects of silviculture on N_2O and CH_4 efflux. It is hypothesized that N_2O and CH_4 efflux will not differ between the different initial planting densities of the CD study but N_2O

efflux will increase and CH_4 efflux will decrease with the higher fertilize/herbicide inputs of the intensive cultural treatments. We also hypothesize that the thinning treatments in the MRT study will lead to greater N_2O efflux due to soil warming but lower CH_4 consumption due to increased soil moisture with the loss of transpiring canopy. Response in the MRT study to fertilization and herbicide is expect to be the same as the CD study, an increase in N_2O efflux and a decrease in CH_4 efflux.

Methods

Site Description

This research utilized two studies (Figure 3.1) previously established by the Plantation Management Research Cooperative (PMRC), based in the Warnell School of Forestry and Natural Resources at The University of Georgia. Typically, as in these two experiments, studies are established on cooperator land but installation, management, and measurements are the responsibility of the PMRC.

Cultural x Density (CD) Study

Three installations of the Culture x Density study in the Lower Coastal Plain of Florida were utilized (Table 3.1). These installations were planted during the 1995/1996 dormant season. Two levels of silvicultural intensity were tested: operational and intensive (Table 3.2). Both levels included herbicide and fertilization treatments but in the intensive treatment complete and sustained control of competing vegetation was achieved and total N inputs were nearly twice that of the operational treatment (approximately 700 vs 1500 kg-N ha^{-1} through 2015). The most recent fertilization was in early spring 2015. The cultural main plots were split into planting density sub-plots of 741, 1483, 2224, 2965, 3706, and 4448 trees ha^{-1} in a randomized split-plot design. This study only utilized the 1483, 2965, and 4448 trees ha^{-1} plots at the three different locations. Replications of the treatments were not established at each site; the different site locations serve as blocks for the study.

Mid-Rotation Treatment (MRT) Study

The Mid-Rotation Treatment study was established in existing stands that were operationally planted and may have had slightly different site preparation, planting density, and cultural treatments prior to the study installation. For inclusion in the study, a stand should have had initial planting of ~1000 to 2000 trees ha^{-1}, be well stocked, and not have been fertilized within five years. The three installations utilized (Table 3.1) for this research were planted in 1997, 1999, and 2000 (Table 3.3), and had pre-thinning basal areas of 24 to 34 m^2 ha^{-1} (Table 3.4). A total of five plots were established at each location: control (no thin, no herbicide, no fertilize), thin only, thin + fertilize, thin + herbicide, and thin + fertilize + herbicide. Basal area on thinned plots was reduced to ~16 m^2 ha^{-1} in 2014 using a fifth row and below combination thinning. The study was designed to be a randomized 2 x 2 factorial of post-thinning fertilization and herbicide release with treatments applied from March to May in 2015 (Table 3.3). Replications of the treatments were not established at the sites; the different site locations serve as blocks for the study.

Forest floor and mineral soil collections and analysis

The forest floor and soil samples from the CD sites were collected 1-year earlier in a previous study (Ward 2015). In the MRT study the same methods were followed. Forest floor samples were collected (February 2015) in eight locations in each plot within each of the three MRT sites. A 35 x 35 cm wooden square frame was used to collect forest floor (O_i, O_e, and O_a) down to the mineral soil. Within each plot, eight locations were randomly sampled and four randomly selected locations were composited providing two within-plot replicates. Samples were returned to the laboratory, placed into paper bags, and dried at 65°C to a constant weight. The forest floor material was passed through a Wiley Mill Model 4 (Thomas Scientific, Swedesboro, NJ) with a 2-mm sieve and then pulverized using a ball mill grinder (SPEX 8000). A subsample of the ground material was used for an ash correction for soil mineral contamination (SSSA, Ben-Dor and Banin 1989). All pulverized samples were analyzed for total C and N concentration using a Thermo Flash 2000 (Waltham, MA, USA).

Mineral soil samples were collected after the removal of the forest floor samples. Samples were collected using a 6.5 cm diameter, open-bucket soil auger to a depth of 1 meter in the following layers: 0-10, 10-20, 20-50, and 50-100 cm. Soil samples from each depth were composited consistent with forest floor samples. Two sampling locations per plot were selected for deeper sampling from 100-150 and 150-200 cm depth to provide complete profile descriptions. Samples were air dried then passed through a 2-mm sieve to remove the roots and rocks.

Soil samples were analyzed for pH, C and N, exchangeable acidity, and macro-nutrients (P, K, Ca, and Mg). Soil pH_{CaCl_2} and pH_{H_2O} were measured using a 1:2 ratio of soil to 0.01 M $CaCl_2$ or deionized (DI) water following Thomas (1996). Exchangeable acidity was determined by shaking 5 g soil with 50 mL of 1 M KCl then filtering through a Whatman 42 filter. Samples were kept in a cooler until titrated to a pH of 8.2 with 0.02 M NaOH using an auto-titrator (Mettler-Toledo, Columbas, OH) (Bertsch and Bloom, 1996). A subsample of mineral soil from each depth was ball mill ground to a powder (SPEX 8000) and analyzed for total C and N content using a Thermo Flash 2000 (Waltham, MA, USA). Soils from the sites sampled down to 2-m were extracted with Mehlich 1 (0.05 N H_2SO_4, 0.025 N HCl) solution (Mehlich, 1953) and analyzed on a Spectro Arcos Inductively Coupled Plasma (ICP-OES) analyzer (Kleve, Germany) for P, K, Ca, and Mg.

N_2O and CH_4 collection and determination

N_2O and CH_4 collections were made in Feb. 2015, June 2015, Mar. 2016, and June 2016. At each date, static chambers made out of 12.7 x 15.2 cm (5 x 6 inch) PVC collars and caps were used to collect gas samples (Hutchinson and Livingston 2002). Four permanent collars were installed in each MRT plot and three were installed in each CD plot. Collars were randomly distributed within each plot of the CD study and within the MRT control plots. Within thinned MRT plots a random stratified placement between thinned tree rows (n=2) and the un-thinned tree rows (n=2) was utilized. On sampling dates, static chamber collars were covered with PVC caps that possessed a collection septum and a ventilation port to allow equalization between internal and external atmospheric pressures. Caps remained on the collars during sample completion. Samples were extracted using a 10mL syringe at 0, 10, 20, 40, and 60 minutes and transferred into pre-evacuated, glass vials (Exetainers, Labco Inc., UK). The gas samples were collected within four hours at any site to minimize temperature differences. Samples were returned to the laboratory and analyzed on a gas chromatograph using an electron capture detector for N_2O and a flame ionization detector for CH_4 (Shimadzu GC-2014, Kyoto, Japan). The carrier gas was Nitrogen (280 kPa) and flow within the column was 24.31 ml min^{-1}. Compressed air and hydrogen gas was the make up for the FID. The ECD make up consisted of a P-5 (95% Aragon and 5% Methane) gas mixture.

During gas sampling and starting in June 2015, soil moisture and temperature were measured at each chamber. Soil moisture was measured with a Hydrosense time domain reflectometry probe (Cambell Scientific, Utah, USA) for 0-15 cm and soil temperature was measured from 0-10 cm (AcuRite Digital Thermometer, Wisconsin, USA). In addition, a soil punch tube (2.54 cm) was used to collect 0-10 cm mineral soil. The soil samples were kept cool until a 2M KCl extraction for exchangeable ammonium could be performed in the laboratory (Bremner 1965). A Flow Solution 3000 (OI Analytical, College Station, TX) flow injection instrument was used to determine the concentration of ammonium within the extracted sample.

Data Analysis

The forest floor and mineral soil data from the MRT sites were analyzed in SAS 9.4 using a mixed model with site as a random blocking factor (Proc MIXED: SAS Institute Inc.). ANOVA assumptions were evaluated and data were transformed as needed. Mineral soil depth was treated as a spatially repeated measurement factor as measurements with depth are autocorrelated.

To estimate rates of gas efflux, the slope of linear regressions between measured concentrations and time of collection (i.e., $\frac{\Delta C}{\Delta t}$) were calculated. All data were initially graphed to visually identify any outliers (outside the 95% confidence interval) that may weaken the linear model; the points that were determined as outliers were removed from the data set (<10% of the data). To convert slopes to efflux the following equation was used:

$$F = (\frac{\Delta C}{\Delta t})(\frac{V}{A})\kappa \qquad \text{Eq 1}$$

where F is the gas flux (mass of gas m^{-2} d^{-1}), $\Delta C/\Delta t$ is the slope that defines the rate that the gas (N_2O or CH_4) concentration changes (μL L^{-1} min^{-1}), V is the volume inside the chamber (m^3), A is the soil surface area covered by the chamber (m^2), and κ is a conversion factor converting volume of gas to mass (temperature corrected ideal gas law) and minutes to day. The regressions were run in R v3.2.3 (R Core Team 2016).

Gas fluxes were analyzed for each experiment separately using a repeated measures mixed model (Proc mixed in SAS 9.4 SAS Institute Inc.). Data normality and ANOVA assumptions were evaluated. In both CD and MRT data, the N_2O efflux was natural log transformed (after adding 0.4 to make all values positive) but the CH_4 efflux did not require transformation. The collars within the MRT sites were stratified between thinned and un-thinned tree rows but were not weighted as area in the two regions were similar. CD had a split-plot design with culture split into densities and repeated measures over the dates. In MRT, to capture information from the unthinned control, data were initially analyzed as five treatments (unthinned control (1), thinned only (2), thin+herb (3), thin+fert (4), thin+herb+fert (5)). If a significant treatment effect was observed, contrasts of unthinned vs thinned (1 vs (2-3-4-5), no herb vs herb (2-4 vs 3-5), and no fert vs fert (2-3 vs 4-5) were analyzed.

Results

MRT forest floor and mineral soil

Soils at the MRT sites were strongly acidic (pH<4) throughout the profiles and were of low base status (Table 3.5). Soil P was also relatively low (<10 μg g^{-1}) throughout except in the surface of the Raiford location. Soil C was somewhat elevated in the 0-10 cm layer (>2%) but generally declined with depth. The Fargo site was an exception having high carbon throughout the profile. Soil N was not similarly elevated in Fargo and was <0.1% throughout all sites.

The ANOVA among treatments across all the MRT sites found no significant effect (p>0.1) by treatment or treatment x depth interaction with either the forest floor mass or soil C and N concentrations (Table 3.6). The average forest floor mass was 62.3±3.6 Mg ha^{-1} while the range among treatments means for C concentrations was 48.5 to 52.1% and for N was 0.36 to 0.58%.

MRT Study- N_2O and CH_4 efflux

There was a significant effect of treatment on N_2O efflux at p=0.11 (Table 3.7 and Figure 3.2) with the thin+herb+fert treatment exceeding all others (p<0.05) and thin+fert exceeding thin only (p<0.07). Date was also significant with Jun16>Mar16. There was a significant treatment by date interaction (p<0.01) as all treatments did not increase or decrease with date consistently (Figure 3.2). All contrasts were significant (Table 3.7) for N_2O efflux with thin>no thin, herb>no herb, and fert>no fert. The N_2O efflux among all the experimental units ranged from -0.191 to 9.774 mg N_2O-N m^{-2} d^{-1} throughout the entire study. On average efflux from the thin only plots (0.023 mg N_2O-N m^{-2} d^{-1}) was 8-fold less than from the thin+herb+fert plots (0.164 mg N_2O-N m^{-2} d^{-1}).

Methane efflux in the MRT study showed no significant effect of treatment ($p>0.1$) but did vary with date ($p=0.003$; Table 3.7) as Jun15 had greater CH_4 consumption (i.e., negative efflux) than Mar16 (Figure 3.2). The CH_4 efflux ranged from -6.60 to 6.68 mg CH_4 –C $m^{-2}d^{-1}$ throughout the study period (Figure3.2).

Site environmental attributes of soil temperature, soil moisture, and extractable NH_4-N varied by sampling date but only soil temperature and extractable NH_4-N were significantly impacted by the MRT treatments ($p=0.064$ and 0.023, respectively; Table 3.8). Average soil temperature was greater under thin+herb+fert (23.3°C) and thin+herb (22.7°C) than the control (21.7°C) and surface temperature ranged from 15.5 to 31.1°C. Average extractable NH_4-N in thin+herb+fert (0.16 μg NH_4-N g^{-1}) was greater than all other treatments (0.007 to 0.023 μg NH_4-N g^{-1}). Extractable NH_4-N ranged from 0.007 to 17.26 μg NH_4-N g^{-1}. VWC ranged from ~15 to 31 cm^3 cm^{-3} with the wettest period being March 2016. Contrasts were consistent with the above results in finding thinned>non-thinned for soil temperature and extractable NH_4-N ($p<0.001$ and 0.003, respectively; Table3.8), fert>no fert for extractable NH_4-N ($p<0.001$), and herb>non-herb for soil temperature and extractable NH_4-N ($p<0.008$ and 0.005, respectively).

CD Study- N_2O and CH_4 efflux

There was not a significant difference between N_2O efflux by culture or density and there was no interaction (Figure 3.3 and Table 3.9). The N_2O efflux ranged from -0.43 to 1.59 mg N_2O-N m^{-2} d^{-1} throughout the entire study. There was a significant effect of date ($p= 0.041$) on N_2O efflux as the Jun16 N_2O efflux was greater than that for the other dates (Figure 3.3).

Methane efflux did not show a significant effect of culture or density and there was no interaction (Figure 3.3 and Table 3.9). The CH_4 efflux ranged from -7.21 to 5.36 mg CH_4–C m^{-2} d^{-1} throughout the study period. There was no significant effect of date ($p>0.3$) on CH_4 efflux and during most dates the majority of the efflux measurements were negative indicating CH_4 is being consumed from the atmosphere.

Culture and density had no significant effects on the edaphic attributes of soil temperature (range of 16-26.3℃), extractable ammonium (range of 0.007-0.010 ug NH_4-N g^{-1}), or volumetric water content (range of 1-20 cm^3 cm^{-3}). All attributes did vary by date ($p<0.001$; Table 3.10). Soil temperature, for example, was lower in Mar16 than Jun16. Extractable NH_4-N was greater in Jun15 and Jun16 as compared to Mar16. Volumetric water content was greater in Mar16 and least in Jun15.

Discussion

Forest floor and mineral soil

No increase was found in the CD study in total N concentration within the forest floor or the mineral soil (Ward, 2015; Table 3.5). In the earlier sampling at the CD study, forest floor mass was greater with more intensive culture such that the content of N (i.e., mass x concentration) in the forest floor was greater. Increases in forest floor mass and N content in fertilized pine plantations has been previously observed (Borders et al., 2004; Kiser and Fox 2012). For example, in 13 and a 24-yr-old loblolly pine stands the addition of ~1,000 kg N ha^{-1} increased the forest floor N by 300 kg N ha^{-1} (Kiser and Fox 2012). Despite the increase in forest floor N observed by Kiser and Fox (2012), no increase was observed in mineral soil N, a result consistent with the current finding. The current studies also found no decrease in mineral soil C with the combined fertilizer and herbicide treatments. Herbicide treatments alone have previously been observed to decrease soil C (Sartori et al. 2007; Rafai et al., 2010). In the MRT study treatments had only been applied months before sampling so this may have reduced the ability to measure responses. Any changes in forest floor or mineral soil C or N could impact CH_4 and N_2O effluxes. For example, increases in litterfall N (presumably leading to increased forest floor N) have been associated with greater N_2O efflux (Davidson et al., 2000)

MRT Study - N_2O and CH_4

The results from the MRT study indicated an impact of not only fertilization, but also herbicide treatments and thinning on N_2O efflux. Fertilizer was applied just prior to the start of measurements thus increasing the likelihood of observing a short-term response in N_2O efflux. As a pulse of fertilizer N is moving through the forest ecosystem an increase in N mineralization, possibly through microbial turnover (Guverlik et al., 2004), can enhance N_2O efflux. As noted above, an increase in N_2O efflux in response to N fertilization is commonly observed (Shrestha et al., 2015), although is somewhat unexpected in the acidic, low nitrifying soil as observed in these plantations.

The increase in N_2O efflux in response to herbicide treatments or thinning appears best explained by the concurrent increases in soil temperature and soil extractable NH_4-N under these treatments (Table 3.8). Extractable N (both NO_3-N + NH_4-N) across a broad range of tropical forests has been positively associated with N_2O efflux (Davidson et al., 2000). In the absence of fertilization, the increases in extractable NH_4-N in these plantations may reflect an increase in soil N mineralization with temperature. Such an increase in N cycling, or as described by Davidson et al. (2000), N flowing through the pipe, would increase N_2O efflux.

The MRT study found no response in CH_4 efflux (Table 3.8). This result is contrary to that from the Florida slash pine plantation that found lowered CH_4 uptake in response to fertilization with a reduction 5-20 times lower than the control plot (Castro et al. 1994). Changes within the N-cycle after the application of fertilization have been proposed to cause the microbial population of CH_4-oxidizing bacteria to become NH_4-oxidizing, thus reducing the amount of CH_4 uptake (Castro et al. 1994, Shrestha et al. 2015). Similarly, ammonification or ammonium based N fertilizers can compete with CH_4 for methane reduction (Schnell and King 1994). In the current study, however, it appears that the lack of change in VWC with treatments (Table 3.8) may have resulted in a lack of CH_4 efflux response (Keller and Reiners 1994).

CD Study - N_2O and CH_4

The CD study did not find a response in N_2O or CH_4 efflux to culture or density. The last fertilization within these 18-year-old stands was in 2015, although both operational and intensive culture received the same input (224 kg N ha^{-1} as urea). As such, it may not be surprising that no significant differences were detected. On the other hand, greater input of N over time in the intensive treatment relative to operational (i.e.,1500 vs 700 kg-N ha^{-1}) was expected to increase N cycling and thus increase N_2O efflux but this was not observed. The lack of response to initial planting density may also reflect the homogenization of standing biomass in the different treatments as stands have self-thinned (Zhao et al., 2015). The lack of response in CH_4 efflux is also consistent with the lack of observed differences in volumetric water content, soil temperature, or extractable NH_4-N (Table 3.10). In the absence of any differences in driving edaphic variables there is no reason to expect differences in N_2O or CH_4 efflux. It is surprising that these Lower Coastal Plain plantations that often are observed to have water tables within 30 cm of the soil surface consistently consumed CH_4 (Figure 3.3).

Conclusion

Results indicated that silvicultural practices can impact N_2O efflux if edaphic variables are impacted by the treatments. In the MRT study, where N fertilization occurred at study initiation, N fertilization elicited an increase in N_2O efflux. In contrast, in the CD study where all plots were fertilized prior to measurements no cultural treatment differences in N_2O efflux were observed. In the MRT study, recent herbicide and thinning treatments also resulted in warmer soil temperatures and higher extractable NH_4 that were associated with higher N_2O efflux. In the CD study, where by age 18 large earlier stand differences resulting from planting density and cultural regime had homogenized to similar standing biomass and canopy characteristics as a result of self-thinning dynamics, soil temperature, moisture, and extractable NH_4 were indistinguishable as was N_2O efflux. No CH_4 efflux treatment related changes were detected, which was consistent with a lack of change in volumetric water content among silvicultural treatments. This study suggests that silvicultural practices that alter N cycling and availability, either through direct inputs from N fertilization or through impacts on edaphic variables (i.e., temperature, extractable N) may enhance N_2O efflux.

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Table 3.1. Locations and soil series for the Culture x Density (CD) and Mid-rotation Treatment (MRT) studies of the Plantation Management Research Cooperative (PMRC) utilized for N_2O and CH_4 efflux investigations.

Study	Install #	Latitude	Longitude	Location	Soil Series
CD	1	30°15'17.92"N	82°18'52.22"W	Olustee, FL	Sapelo
CD	9	30°42'08.64"N	81°46'34.33"W	Hilliard, FL	Ocilla
CD	11	30°32'47.57"N	81°57'46.12"W	Callahan, FL	Ridgewood
MRT	51	30°49'11.82"N	82°36'30.73"W	Fargo, GA	Mascotte
MRT	54	30°06'14.81"N	82°17'04.87"W	Raiford, FL	Mascotte
MRT	55	31°22'17.19"N	81°53'45.96"W	Jesup, GA	Leon

Table 3.2. Silvicultural treatments implemented for operational and intensive cultural regimes for the Culture by Density (CD) study sites.

Treatment	Operational (OPER)	Intensive (INT)
Chemical Site Preparation	Late summer/fall high-rate banded	Late summer/fall high-rate broadcast
Mechanical Site Preparation	Bedding	Bedding
Fertilization	1996: 561 kg ha $^{-1}$ of 10-10-10	1996: 561 kg ha^{-1} of 10-10-10
		1998: 673 kg ha^{-1} of 10-10-10 + 131 kg ha^{-1} NH_4NO_3 + micronutrients
		1999: 336 kg ha^{-1} NH_4NO_3
		2001: 336 kg ha^{-1} NH_4NO_3
	2003, 2007, 2015: 224 kg ha^{-1} N (from urea) + 28 kg ha^{-1} P	2003, 2005, 2007, 2009, 2013, 2015: 224 kg ha^{-1} N (from urea) + 28 kg ha^{-1} P
Herbicide (weed control)	1996: 0.280 kg ha^{-1} Oust banded + direct spraying for hardwood control	1996: 0.280 kg ha^{-1} Oust banded + direct spraying for complete competing vegetation control, 0.235 kg ha^{-1} Arsenal broadcast. Repeated direct spraying for complete vegetation control

Table 3.3. The Mid-Rotation Treatment (MRT) site history for the application of thinning, fertilizer, and herbicide treatments. The fertilizer consisted of 224 kg ha^{-1} N (urea) and 28 kg ha^{-1} P (DAP). The herbicide consisted of Garlon 3A (10 oz ha^{-1}) and glyphosate (10 oz ha^{-1}) with an over the top broadcast spray and a basal stump treatment of Garlon 4 (25%) and bark oil (75%).

Location	Installation	Planted	Thinned	Fertilization	Herbicide
Fargo	51	Jun-00	Nov-14	Mar-15	May-15
Raiford	54	Jun-97	Dec-14	Mar-15	May-15
Jesup	55	Jun-99	Nov-14	Mar-15	May-15

Table 3.4. Basal area and height of the Mid-Rotation Treatment (MRT) sites prior to thinning treatment. The target thinning was to 16 m^2 ha^{-1}.

Installation	**Plot**	**Basal area (m^2 ha^{-1})**	**Height (meters)**
Fargo, GA 51	Control	28.0	17.6
	Thin	25.2	16.6
	Thin+Herb	26.1	16.3
	Thin+Fert	26.9	17.1
	Thin+Herb+Fert	26.2	16.7
Raiford, FL 54	Control	33.8	17.4
	Thin	31.7	17.3
	Thin+Herb	30.0	17.1
	Thin+Fert	31.5	17.5
	Thin+Herb+Fert	33.0	17.9
Jesup, GA 55	Control	27.5	15.3
	Thin	26.0	15.1
	Thin+Herb	24.7	15.6
	Thin+Fert	24.5	15.3
	Thin+Herb+Fert	25.0	14.9

Table 3.5. Average (± SE) pH_{CaCl_2}, C, N, P, K, Ca, Mg, exchangeable acidity and effective cation exchange capacity (ECEC) for the Mid-Rotation Treatment (MRT) sites. N=2 per site and samples were collected in December 2015 after thinning and post fertilization and herbicide application.

Site Location	Depth (cm)	pHw	C %	N %	P (ug/g)		K (cmolc/kg)		Ca (cmolc/kg)		Mg (cmolc/kg)		Exch Ac (cmolc/kg)		ECEC (cmolc/kg)
Fargo, GA	0-10	3.4±0.1	2.2±1.1	0.042±0.010	11.9	±3.84	0.018	±0.004	0.032	±0.003	0.052	±0.011	2.53	±1.466	2.63
	10-20	3.6±0.1	2.0±1.0	0.017±0.006	4.3	±1.51	0.014	±0.003	0.017	±0.005	0.045	±0.010	2.65	±0.877	2.73
	20-50	3.6±0.1	1.8±1.9	0.014±0.002	1.5	±0.08	0.006	±0.000	0.012	±0.006	0.018	±0.000	1.15	±0.392	1.19
	50-100	3.9±0.1	1.3±8.3	0.013±0.001	1	±0.07	0.005	±0.006			0.017	±0.012	1.05	±0.441	1.07
	100-150	3.9±0.1	2.8±0.3	0.013±0.002	3.4	±0.60	0.008	±0.002	0.018	±0.002	0.025	±0.007	2.19	±1.007	2.24
	150-200	3.8±0.2	3.0±1.3	0.011±0.001	12.8	±6.50	0.01	±0.004	0.03	±0.007	0.031	±0.013	1.91	±0.049	1.98
Jesup, GA	0-10	2.8±0.0	2.5±0.6	0.078±0.014	24.4	±7.35	0.061	±0.014	0.232	±0.098	0.195	±0.045	4.27	±0.755	4.76
	10-20	3.3±0.1	1.5±0.2	0.047±0.006	7.6	±2.10	0.023	±0.001	0.055	±0.019	0.074	±0.003	0.99	±0.314	1.14
	20-50	3.7±0.1	0.6±0.0	0.021±0.001	1.7	±0.20	0.01	±0.001	0.019	±0.001	0.033	±0.005	1.98	±0.998	2.04
	50-100	3.5±0.2	0.2±0.0	0.011±0.001	2.2	±0.33	0.008	±0.000	0.012	±0.000	0.025	±0.001	0.63	±0.189	0.67
	100-150	3.5±0.1	0.3±0.2	0.011±0.002	17.3	±12.21	0.018	±0.004	0.042	±0.028	0.059	±0.012	0.37	±0.246	0.49
	150-200	3.5±0.0	0.1±0.0	0.011±0.002	28.2	±21.08	0.017	±0.009	0.056	±0.042	0.054	±0.029	0.61	±0.245	0.74
Raiford, FL	0-10	3.5±0.1	3.9±1.1	0.095±0.013	67	±9.86	0.075	±0.018	1.047	±0.313	0.24	±0.057	2.34	±0.539	3.70
	10-20	3.7±0.3	1.3±0.4	0.037±0.012	15.1	±4.73	0.021	±0.004	0.339	±0.238	0.068	±0.013	1.01	±0.187	1.44
	20-50	4.1±0.2	0.8±0.2	0.026±0.004	4.7	±0.87	0.012	±0.003	0.103	±0.058	0.038	±0.009	1.4	±0.160	1.55
	50-100	4.2±0.2	0.3±0.1	0.012±0.002	3.6	±1.26	0.009	±0.003	0.057	±0.023	0.029	±0.009	1.12	±0.102	1.22
	100-150	4.2±0.0	0.2±0.1	0.013±0.001	11.7	±2.89	0.011	±0.001	0.025	±0.007	0.035	±0.002	1.92	±0.166	1.99
	150-200	4.2±0.1	0.2±0.0	0.010±0.001	20.6	±6.60	0.014	±0.002	0.03	±0.004	0.044	±0.006	3.79	±0.090	3.88

Table 3.6. Average (± SE) percent carbon and nitrogen, and total organic matter by treatment and soil depth at the Mid-Rotation Treatment (MRT) sites. The C, N, and organic mass is calculated on an ash free basis. Samples collected in December 2015. The thinning occurred in November 2014, fertilization in March 2015, and herbicide application in May 2015.

Treatment	Depth	C		N		Organic Matter Mass
	cm	%		%		Mg ha^{-1}
Control	Organic	47.48	±0.87	0.357	±0.05	66.0±4.7
	0-10	2.02	±0.29	0.066	±0.01	
	10-20	1.38	±0.44	0.042	±0.01	
	20-50	0.67	±0.10	0.022	±0.00	
	50-100	0.42	±0.10	0.015	±0.00	
Thin	Organic	52.18	±2.51	0.583	±0.09	59.6±8.7
	0-10	3.03	±0.84	0.085	±0.02	
	10-20	1.17	±0.60	0.030	±0.01	
	20-50	0.69	±0.08	0.020	±0.00	
	50-100	0.60	±0.10	0.019	±0.00	
Thin+Herb	Organic	48.53	±2.11	0.424	±0.06	64.4±7.5
	0-10	2.88	±0.79	0.074	±0.02	
	10-20	0.99	±0.25	0.029	±0.01	
	20-50	0.58	±0.10	0.019	±0.00	
	50-100	0.27	±0.03	0.011	±0.00	
Thin+Fert	Organic	48.81	±1.17	0.481	±0.04	64.1±5.3
	0-10	2.24	±0.32	0.066	±0.01	
	10-20	1.38	±0.63	0.032	±0.01	
	20-50	0.92	±0.27	0.024	±0.00	
	50-100	0.46	±0.07	0.014	±0.00	
Thin+Herb+Fert	Organic	50.59	±2.89	0.525	±0.06	57.4±1.4
	0-10	3.34	±0.88	0.086	±0.02	
	10-20	1.16	±0.38	0.030	±0.01	
	20-50	0.77	±0.14	0.024	±0.00	
	50-100	0.36	±0.06	0.012	±0.00	

Table 3.7. Trace gas efflux ANOVA summary statistics from the Mid-Rotation Treatment (MRT) study.

		DF	F	P-value
N_2O	Treatment	4	2.79	0.101
	Date	3	3.45	**0.018**
	Treatment * Date	12	2.26	**0.011**
	Contrasts			
	NoThin vs Thin	1	4.52	**0.035**
	NoFert vs Fert	1	17.57	**<0.001**
	NoHerb vs Herb	1	11.82	**0.007**
CH_4	Treatment	4	0.13	0.96
	Date	3	6.53	**0.003**
	Treatment *Date	12	0.7	0.75
	Contrasts			
	NoThin vs Thin	1	0.21	0.65
	NoFert vs Fert	1	0.42	0.58
	NoHerb vs Herb	1	1.24	0.34

Table 3.8. ANOVA summary statistics (p-values) for soil temperature, volumetric water content (VWC), and extractable NH_4-N from the Mid-Rotation Treatment (MRT) sites.

Factors	**DF**	**F**	**Temperature**	**VWC**	**NH_4-N**
Treatment	4	3.45	**0.004**	0.98	**0.023**
Date	2	917	**<0.001**	**<0.001**	**<0.001**
Treatment *Date	8	0.83	0.57	0.55	0.46
Contrasts					
NoThin vs Thin	1	16.74	**<0.001**	0.83	**0.003**
NoFert vs Fert	1	1.08	0.19	0.38	**<0.001**
NoHerb vs Herb	1	7.15	**0.008**	0.24	**<0.005**

Table 3.9. Trace gas efflux ANOVA summary statistics from the Culture x Density (CD) study.

Gas	Factor	**DF**	**F**	**P-value**
N_2O	Culture	1	2.30	0.269
	Density	2	0.23	0.803
	Culture*Density	2	2.00	0.250
	Date	3	2.82	**0.041**
	Date*Culture	3	0.99	0.401
	Date*Density	6	0.30	0.937
CH_4	Culture	1	1.56	0.338
	Density	2	2.29	0.217
	Culture*Density	2	1.98	0.252
	Date	3	1.15	0.332
	Date*Culture	3	0.96	0.415
	Date*Density	6	2.05	0.062

Table 3.10. ANOVA summary statistics (p-value) for soil temperature, volumetric water content (VWC), and soil extractable NH_4-N from the culture x density (CD) study.

	DF	Temperature P-value	VWC P-value	NH_4-N P-value
Culture	1	0.989	0.063	0.569
Density	2	0.240	0.838	0.417
Culture*Density	2	0.129	0.219	0.430
Date	3	**<0.001**	**<0.001**	**<0.001**
Date*Culture	3	0.091	0.854	0.851
Date*Density	6	0.303	0.563	0.932

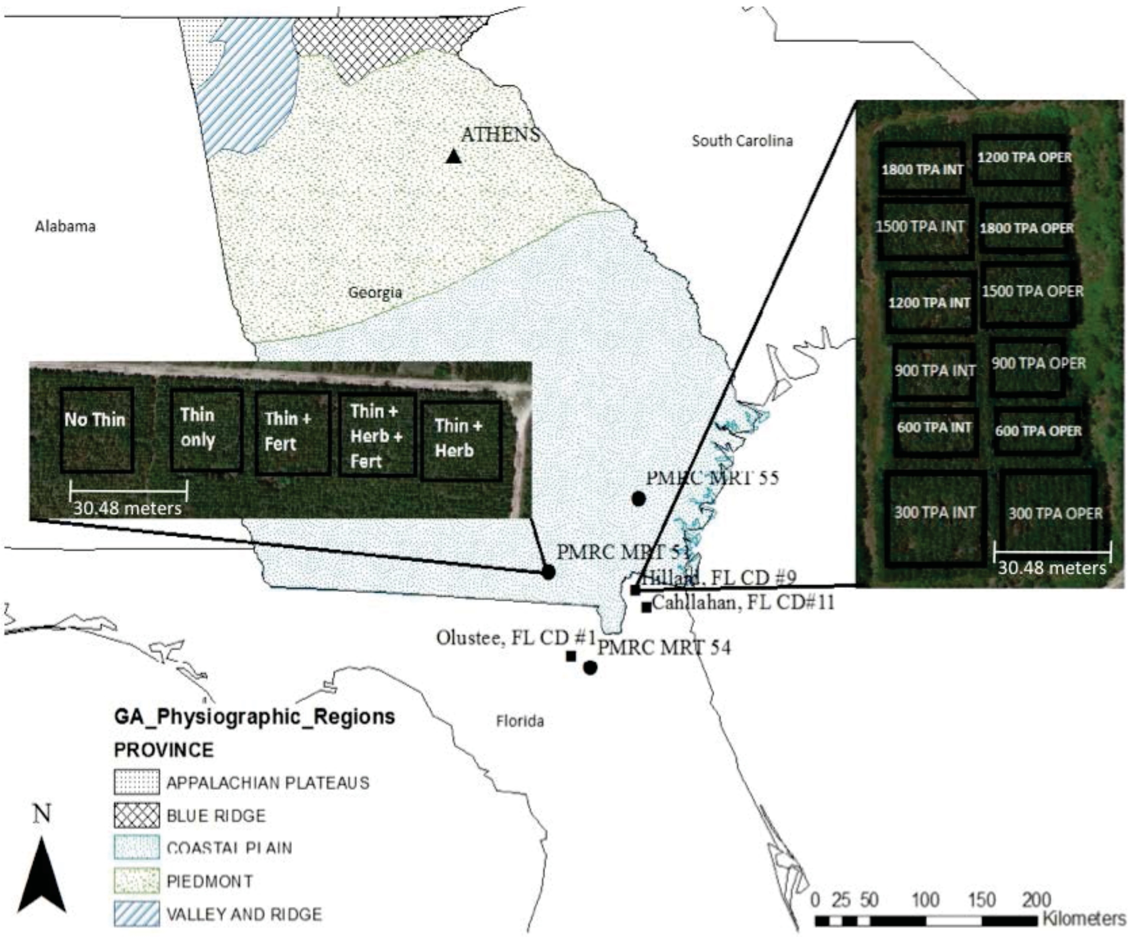

Figure 3.1. The Culture x Density (CD) and Mid-Rotation Treatment (MRT) site locations in Georgia and Florida, Lower Coastal Plain. An inset of the different layouts and treatments. The sites consisted of both loblolly and slash pine.

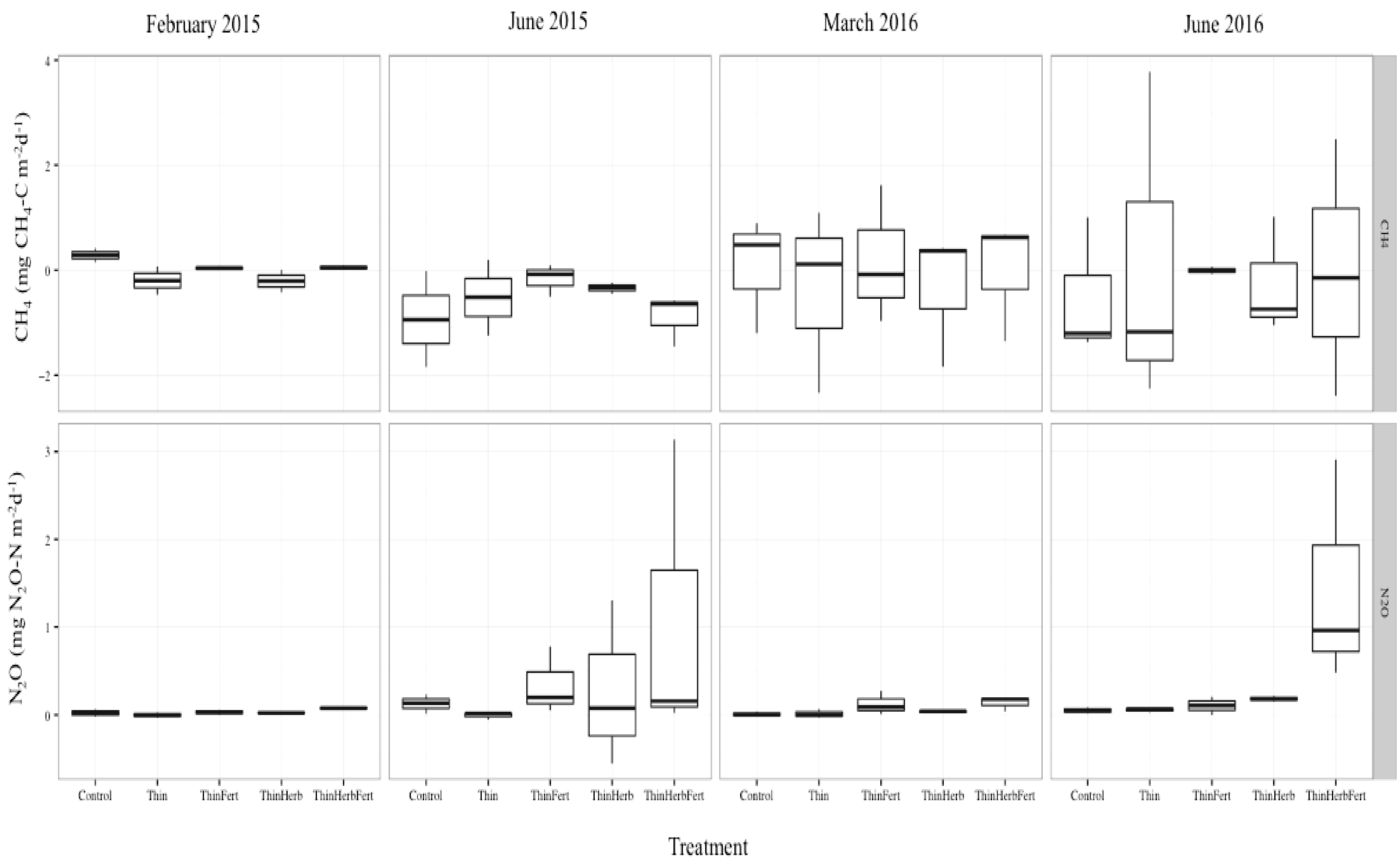

Figure 3.2. The Mid-Rotation Treatment (MRT) CH_4 and N_2O efflux from February 2015 to June 2016 (mean ± SE), n=4. Thinning occurred in November 2014, fertilization in March 2015, and herbicide application in May 2015.

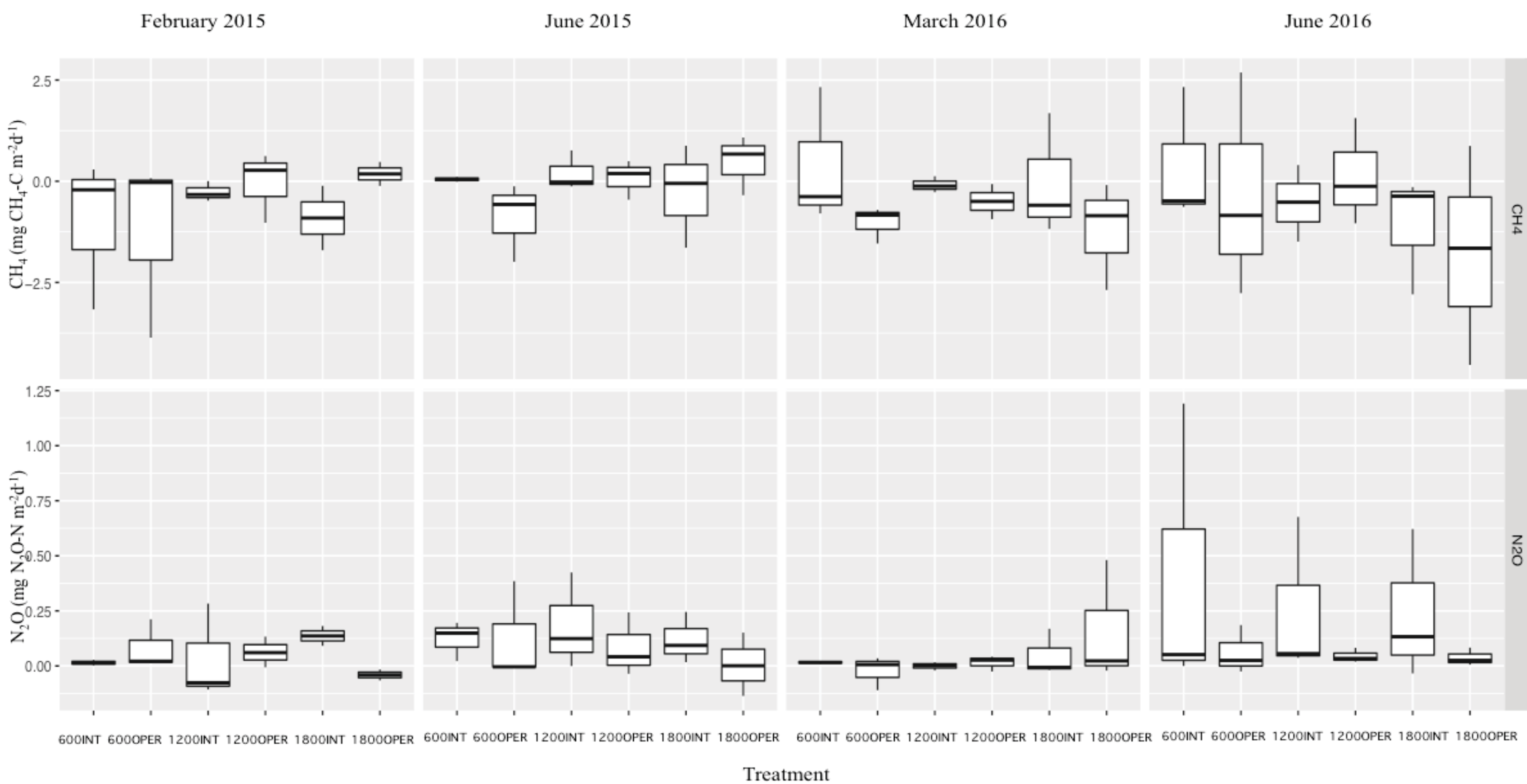

Figure 3.3. The Culture by Density (CD) CH_4 and N_2O efflux from February 2015 to June 2016 (mean ± SE), n=3. All plots were fertilized shortly after the February 2015 sampling.

CHAPTER IV

CONCLUSION

The amount of CH_4 and N_2O efflux from pine plantations within the southeastern United States is an important issue relative to global climate change. An increase in the area and management of pine plantations has resulted in an increase in fertilizer use and other silvicultural treatments. Fertilizers increase the growth rate of pine trees and shorten rotation lengths. Herbicides and thinning allow for nutrients to be re-allocated to the remaining trees within the stand thus also increasing growth. Understanding how these silvicultural treatments within southern pine plantations effect not only growth (and CO_2 uptake) but also CH_4 and N_2O efflux is critical for assessing the net impact on GHG fluxes.

The throughfall exclusion by fertilization experiment reported on in this thesis found that fertilization of loblolly pine plantations increases N_2O efflux, but only for a short duration. Results also indicated that urea, the most common fertilizer used within loblolly pine plantations, releases similar amounts of N_2O as other common fertilizers. Throughfall exclusion did not affect N_2O efflux but increased CH_4 consumption. Methanotrophy occurs under aerobic conditions thus increasing in the absence of precipitation. Both N_2O and CH_4 emissions increased months after fertilization in the presence of adequate precipitation changing GHG sinks to sources. This throughfall exclusion by fertilization study demonstrated that both silvicultural management and a drier regional climate can have important impacts on the net balance of N_2O and CH_4 efflux in southern pine plantations.

The Culture x Density (CD) study reported on indicated that planting density did not have an impact on CH_4 or N_2O efflux after 18 years. Differences in efflux between cultural treatments with fertilizers of herbicides as operational or intensive were also not evident, although both cultural treatments received equal amounts of N fertilization in the year of the study. The Mid-Rotation Treatment (MRT) study observed that the application of fertilizers increased N_2O but not CH_4 efflux. Thinning and herbicide treatments in the MRT study were also found to increase N_2O but not CH_4 efflux. Thinning and herbicide treatments were associated with increases in soil temperature and extractable NH_4. CH_4 and N_2O efflux were positively correlated with soil temperature throughout the duration of the study. CH_4 efflux was also positive correlated with soil moisture. Soil extractable ammonium was positively correlated with N_2O efflux. Both the CD and MRT studies demonstrated that soil temperature, amount of available N, and soil moisture are all factors controlling N_2O and CH_4 efflux. These studies both suggest that silvicultural practices that alter N cycling and availability, either through direct inputs from N fertilization or through impacts on edaphic variables (i.e., temperature, extractable N) may enhance N_2O efflux.