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Soil microbial nitrogen cycling and nitrous oxide emissions from urban afforestation in the New York City Afforestation Project

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ABSTRACT

The establishment of stands of trees in previously unforested areas (afforestation) is active in cities around the world. Given the complex and often degraded state of urban soils, there is great interest in soil biological processes that support plant growth but may also produce greenhouse gases in these new forests. We measured soil to atmosphere fluxes of nitrous oxide (N_2O) in order to determine how the presence/absence of shrubs and compost in urban afforestation site preparation affects the emission of this potent greenhouse gas. To complement the measurement of N_2O flux, microbial biomass carbon (C) and nitrogen (N), potential net N mineralization and nitrification, microbial respiration, and soil inorganic N were measured in experimentally afforested plots in New York City, USA. Results suggest that afforestation with shrubs and trees stimulates smaller fluxes of N_2O from soils than afforestation without shrubs and trees. The range of N_2O flux observed from recently afforested plots was -0.031–0.641 ng N cm^{-2} h^{-1}. There were no significant differences in N_2O fluxes and microbial biomass C between sites with shrubs and/or one–time application of compost. The results suggest that afforestation efforts to create natural vegetation structure (i.e. canopy trees with understory plants) and foster a functional microbial community through additions of organic matter may not increase emissions of N_2O to the atmosphere. Rather, this method of afforestation site preparation may tighten C and N cycles and leave N_2O emissions in these urban ecosystems unchanged.

1. Introduction

Afforestation, the establishment of stands of trees in previously unforested areas, is active in cities around the world with intentions of reducing storm water runoff, sequestering atmospheric carbon dioxide (CO_2), reducing urban ambient temperatures and providing green spaces to improve local quality of life (Kitha and Lyth, 2011; Pataki et al., 2011; Jansson, 2013; Schäffler and Swilling, 2013). As city governments seek to increase "green infrastructure" to attain observed environmental and health benefits for city residents, there is a need to evaluate the effects of creating urban green spaces on ecosystem processes (Guo and Gifford, 2002; Churkina, 2008; Jansson, 2013; Oldfield et al., 2014). Soil processes involved in carbon (C) and nitrogen (N) cycling are critical for tree establishment and growth and influence the impact of the urban forest ecosystem on the atmosphere and hydrology (Cogliastro et al., 2003; Pouyat et al., 2006; Nowak et al., 2013; Oldfield et al., 2014). Understanding the interactions among soil biogeochemical cycles and afforestation processes is critical to meeting the diverse goals of urban afforestation policies.

The natural N cycle is greatly altered by direct anthropogenic inputs of reactive N from the combustion of fossil fuels, application of agricultural fertilizers, and conversion of landscapes from natural to human-dominated spaces (Neff and Hooper, 2002; Galloway et al., 2003; Howarth, 2004; Filoso et al., 2006) and these changes may create positive feedbacks to climate change (Hungate et al., 2003; Zaehle et al., 2010). Nitrogen is made bioavailabile in soils through the microbial processes of mineralization and nitrification through which organic N is converted to ammonium (NH_4^+) and then to nitrate (NO_3^-) (Crawford and Glass, 1998) with the greenhouse gas (GHG) nitrous oxide (N_2O) emitted as a by-product. This process is linked to the removal of N from the soil system by the microbial process of denitrification through which NO_3^- is reduced to N_2O (with some release to the environment) and finally to chemically inert dinitrogen (N_2) gas.

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Spatial and temporal variability in the production of N_2O from soils creates great uncertainty in estimating fluxes from natural and anthropogenic ecosystems (Groffman and Tiedje, 1989; Groffman et al., 2009). The environmental characteristics known to drive N_2O production include soil moisture, the presence of oxygen, availability of C substrates and temperature, though the interactions of these and other factors is a topic of current investigation (Morse et al., 2015; Powell et al., 2015). Relevant environmental parameters are often difficult to characterize in human-dominated systems where conditions can be altered in unknown ways (Kaye et al., 2006; Pickett and Cadenasso, 2008; Raciti et al., 2011; Pickett et al., 2013). In suburban and urban landscapes, changes to soil structure, nutrient inputs, species composition, irrigation and impervious surfaces have been shown to alter N cycling rates and N_2O fluxes to the atmosphere (Zhu et al., 2004; Kaye et al., 2006; O'Driscoll et al., 2010; Groffman et al., 2014). As research increasingly shows that human decisions in urban and exurban planning can have net negative impacts on the environment, managers and planners have sought to design more ecologically sustainable spaces (Seyfang and Smith, 2007; Kitha and Lyth, 2011; Felson et al., 2013; Baró et al., 2014). There is a strong need to determine whether efforts to mitigate climate change through improved design of urban greenspaces have positive or negative effects on N_2O emissions from urban soils.

Studies which have tested the effects of afforestation of soils with different land use histories on rates of net N mineralization and nitrification and soil inorganic N content generally suggest that afforestation decreases these variables, likely due to a changing relationship between soil organic C and N over time since afforestation (Templer et al., 2005; Luo et al., 2006; Gelfand et al., 2012; Deng et al., 2014). Singh et al. (2011) provide evidence that shifts in microbial community structure alter soil N cycling and decrease N_2O fluxes as a result of afforestation. There is a body of evidence which suggests that organic C and N inputs to soil from afforestation as well as the microbial community response to afforestation may drive changes in N cycling and alter the production of N_2O.

In the US, the The MillionTreesNYC (MTNYC) initiative is a large-scale effort to establish one million new trees, and "healthy, multi-story forests with native trees, shrubs and herbaceous layer" in particular, in New York City (Lu et al., 2014). The MTNYC initiative is a component of New York City's PlaNYC2030 sustainability agenda (www.milliontreesnyc.org). Because of the scale of its planting goals, the initiative is among the most ambitious municipal efforts to increase urban canopy and green space. The New York City Afforestation Project (NY-CAP) is a component of the MTNYC initiative developed as a designed-experiment approach to address the effects of urban environmental stressors and management on the physical attributes of soils and tree performance in urban sites (Felson et al., 2013). The NY-CAP plots were varied in shrub presence/absence, tree species richness and soil amendment with compost. To determine the importance of stand structure and initial inputs of organic matter to N_2O emissions from urban afforestation sites, we evaluated the effects of presence/absence of a shrub and herbaceous understory species and compost addition on N_2O flux in the NY-CAP. We hypothesized that shrub planting and compost application would lead to greater availability of C, higher microbial biomass and higher rates of microbial respiration that would increase rates of N uptake by microbes (immobilization) and lower rates of N transformations (nitrification and denitrification), thus decreasing N_2O emissions.

2. Methods

This research was conducted in Kissena Corridor Park (KCP) (40.749824°N, −73.823136°W) in the Flushing neighborhood of Queens County, New York, USA. Queens, NY experiences a temperate humid climate and receives an average annual rainfall of 113 cm (NOAA, 2012). The area of the study site is approximately 6.6 ha, and land use in the surrounding area is a mixture of high-density residential and commercial buildings. Soils in Kissena Corridor Park are classified as Inwood–Laguardia–Ebbets complex (Typic udorthents), well-drained loamy-skeletal to coarse-loamy fill, with ∼35% coarse fragments (NRCS, 2009).

Table 1
Treatment description, and number of plots sampled for N_2O flux per month and ambient temperature at time of sampling in December 2012 and March and November 2013.

Treatment	November (10.0 °C)	December (2.8 °C)	March (4.4 °C)
Shrubs/Herbs + Compost	$n = 4$	$n = 0$	$n = 1$
Shrubs/Herbs only	$n = 4$	$n = 4$	$n = 4$
Compost only	$n = 4$	$n = 2$	$n = 0$
No additions	$n = 6$	$n = 4$	$n = 4$

The NY-CAP research sites employed a factorial design to vary three factors in afforestation site preparation: presence/absence of shrubs and herbaceous plants, presence/absence of soil amendment with compost, and high/low tree species richness (Felson et al., 2013). This study used a subset of experimental plots ($n = 12$) within KCP to make a factorial comparison of presence/absence of shrubs and herbs and presence/absence of compost. The subset of plots in this study represented the high species richness treatment group of the NY-CAP, thus tree species richness was consistent throughout the 12 plots sampled. All plots included in this study were embedded within parklands that prior to experimental preparation were overgrown with and dominated by a small number of mostly invasive, herbaceous species such as mugwort (*Artemesia vulgaris*) and phragmites (*Phragmites australis*) as well as goldenrod (*Solidago canadensis*) and other native weeds (Oldfield et al., 2014).

In 2009, the plots were weeded and rototilled, and compost treatment plots had compost applied at a rate of 2.5 cm^3 per 100 cm^3 of soil to 15 cm depth by a landscaping company contracted by the New York City Department of Parks and Recreation. The compost had a pH of 6.3, a bulk density of 457 kg m^3, and nutrient composition was 60% C, 3.2% N, 3.7% P and 0.44% K (dry weight basis) (Oldfield et al., 2014). In 2010, each experimental plot was planted with 56 individual trees (3–5 year old saplings, 0.6–1.2 m in height), 41 individual shrubs and 672 individual herbaceous seedlings (Oldfield et al., 2014). The tree species planted in these plots were *Tilia americana*, *Quercus rubra*, *Carya* sp., *Prunus serontina*, *Quercus alba*, and *Celtis occidentalis*. Shrub species included were *Cornus racemosa*, *Hamamelis virginiana*, *Lindera benzoin*, *Sambucus canadensis*, and *Viburnum dentatum* (see Felson et al., 2013 for more planting details). Herbaceous species included were *Apocynum cannabinum, Asclepias syriaca, Elymus canadensis, Euthamia graminifolia, Eupatorum purppurium, Panicum virgatum, and S. canadensis.* The experimental plots were surrounded by playing fields, foot paths, roads and high-rise buildings and were accessed by park patrons. Some of the 12 treatment plots were lost due to vandalism and storm damage, thus treatment replicates were uneven (see Table 1).

Soil cores (2–5 per plot; 15 cm length and 5 cm diameter) were taken with beveled-edge PVC corer in December 2012 from each experimental plot and composited. Soil samples were homogenized by hand to remove large and fine roots, rocks and woody organic debris. Microbial biomass C and N and were measured using the chloroform fumigation incubation method (Gundersen et al., 2012; Jenkinson et al., 2004). Test samples of 7.5 (±0.05) g were fumigated with chloroform ($CHCl_3$) for ∼12 h, reinoculated with 0.20 (±0.05) g of unfumigated fresh soil and incubated for 10 days

Table 2
Means (±SE) for one-time measurement of microbial biomass carbon and nitrogen content, potential net nitrogen mineralization and nitrification, microbial respiration and soil inorganic N (NH_4^+ and NO_3^-) and moisture content in soils from four treatments in Kissena Corridor Park sampled in December 2012. There were no significant differences between treatments for any variable ($p > 0.05$).

Variable	Unit	Shrubs + Compost (n = 8)	Shrubs only (n = 8)	Compost only (n = 7)	No additions (n = 10)
Microbial biomass C	g C/m^2/day	228.0 (73.42)	321.42 (138.61)	292.02 (177.07)	217.47 (91.21)
Microbial biomass N	g N/m^2/day	5.54 (2.32)	7.24 (1.26)	5.13 (1.83)	3.51 (2.31)
Soil respiration	g C/m^2/day	8.52 (5.20)	28.7 (4.22)	18.59 (2.10)	3.71 (1.15)
Total inorganic N	g N/m^2	3.77 (3.05)	7.28 (1.75)	5.54 (6.77)	2.88 (3.20)
NO_3^-	g N/m^2/day	2.90 (2.51)	5.15 (1.01)	4.52 (4.53)	1.79 (2.64)
NH_4^+	g N/m^2/day	0.89 (0.53)	2.13 (0.77)	2.77 (2.28)	1.09 (0.58)
Potential net N mineralization	g N/m^2/day	0.21 (0.20)	0.43 (0.11)	−0.43 (0.55)	0.04 (0.26)
Potential net nitrification	g N/m^2/day	0.21 (0.12)	0.19 (0.12)	0.09 (0.26)	0.02 (0.21)
Volumetric water content	% (g dry soil/g field moist soil)	43	53	42	51
Bulk density	g/cm^3	1.42 (0.07)	1.42 (0.07)	1.42 (0.07)	1.42 (0.07)

at laboratory temperature (~20 °C) in 1 L glass jars fitted with rubber septa to allow for sampling of headspace gas (10 mL). Control samples not treated with $CHCl_3$ were incubated in parallel with the fumigated samples. These incubations provided estimates of potential net N mineralization and nitrification and microbial respiration. Ammonium and $NO^-{}_3$ were extracted from initial samples with 2 M potassium chloride (KCl) prior to incubation and incubated samples after 10 days and quantified colorometrically using continuous flow injection analysis (Lachat QuikChem Automated Flow Injection Analysis System) (Lachat, Loveland, CO). Headspace gas was sampled from fumigated and unfumigated samples before and after incubations and CO_2 was quantified with on a Shimadzu GC 14 gas chromatograph equipped with a thermal conductivity detector. Accumulation of CO_2 in the headspace of the fumigated samples was assumed to be directly proportional to the microbial biomass C content of the soil (corrected by a proportionality constant of 0.45). Accumulation of CO_2 in the headspace of the unfumigated samples provided estimates of microbial respiration. Accumulation of NH_4^+ (measured as described above) was assumed to be directly proportional to the microbial biomass N content of the soil (no proportionality constant was used). Accumulation of NH_4^+ plus NO_3^- in the unfumigated samples was used to calculate potential net N mineralization and accumulation of NO_3^- alone in these samples provided an estimate of potential net nitrification. Volumetric water content was calculated on a dry weight basis from one oven dried subsample from bulked soil cores for each plot (Table 2).

Nitrous oxide fluxes from each plot were sampled using an in situ static chamber design identical to the design described by Bowden et al. (1991). Measurements were made once per month in December 2012, March 2013 and November 2013. Two chambers consisting of 278 mm-diameter by 40mm-high PVC lids fitted on top of permanently established base rings with the same measurements were established randomly within each plot. Headspace gas was allowed to accumulate for 30 min, with samples taken at 10 min intervals. The samples were taken from the headspace of the chamber with syringes from ports fitted with septa. The samples (10 mL) were then transferred to evacuated glass vials fitted with lids and Teflon septa and were stored at room temperature. Gases were later analyzed for N_2O and CO_2 concentration with a Shimadzu GC 14 gas chromatograph equipped with electron capture and thermal conductivity detectors. Fluxes were calculated from the slope of the regression of N_2O concentrations with time over the 30 min incubation.

We used publicly available data from a mature urban forest site in Leakin Park, Baltimore, MD, USA (39.306389°N, −76.690833°W) from the Baltimore Ecosystem Study (BES) as an undisturbed reference site (Groffman, 2012) for comparison with our KCP site. BES sites have undisturbed vegetation dominated by *Quercus* spp. and *Liriodendron tulipifera* (Groffman et al., 2009). Though the KCP and BES plots differ in their dominant vegetation and soil, BES serves as a regional comparison within the context of urban

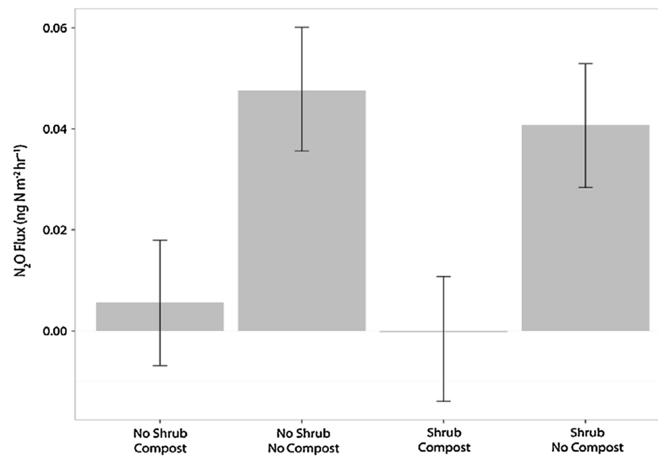

Fig. 1. Least squares means with standard error for N2O flux from soils across four treatments in Kissena Corridor Park. Sampling occurred in December 2012, March 2013 and November 2013. Treatments are no shrubs/no compost (No.Sh.No.Com, n = 14), shrubs/no compost (Sh/No.Com, n = 16), no shrubs/compost (No.Sh/Com, n = 6), and shrubs/compost (Sh/Com, n = 5).

forest N_2O production in the eastern United Sates. N_2O flux data from the BES sites collected from 1998–2010 (n = 185) in March were averaged and compared to the N_2O flux data collected from KCP in March 2013. Repeated measures analysis of variance (ANOVA) was used to compare the response of N_2O flux to the treatments. Least squares means of N_2O flux were calculated for each treatment for the KCP plots due to the small sample sizes of this dataset and the variable number of replicates among treatments and among sampling dates. The N_2O flux data from the BES Leakin Park forest sites and from this study were tested for normality using the Shapiro–Wilk test ($\alpha = 0.05$). All statistical analyses were carried out in R 3. 0. 3 (R Core Team, 2014).

3. Results

N_2O fluxes were highest in the plots not amended compost (Fig. 1) and were higher in March compared to November and December (ANOVA, df = 41, $p = 0.0541$) (Fig. 2). Fluxes were non-normally distributed ($p < 0.05$), and while the ANOVA of the mean N_2O flux values for each treatment indicates a marginal difference in the flux response of at least one treatment (ANOVA, df = 41 $p = 0.048$), it was not possible to discern specific significant differences between treatments. The mean N_2O flux values from KCP during December 2012, March 2013 and November 2013 were lower (but not significantly) than the mean N_2O flux from March, December and November (1998–2012) from the BES reference site in Baltimore ($p = 0.849$) (Fig. 3).

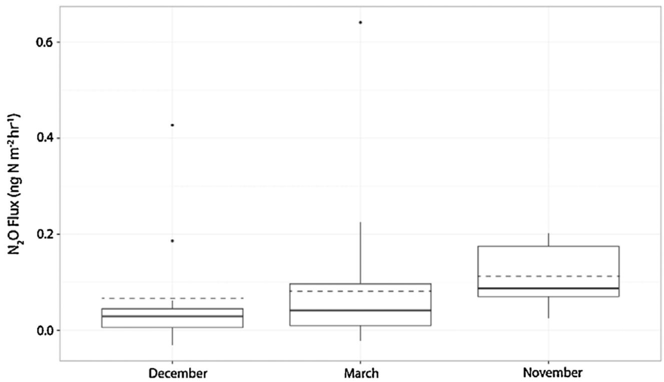

Fig. 2. Nitrous oxide flux from soils (with standard error, December $n = 19$; March $n = 19$, November $n = 15$) in December 2012, March 2013 and November 2013 in Kissena Corridor Park averaged across four treatments. Medians are shown as dashed lines, means are shown as bold, solid lines and outliers are shown as black dots.

The shrubs/no compost treatment had the highest values for nearly all soil variables related to microbial activity among the four treatments (Table 2). In contrast, the no shrubs/no compost treatment had the lowest values among the treatments for nearly all soil variables measured (Table 2). Both compost treatments had high values for microbial biomass C and N, respiration, inorganic N and nitrification compared to the treatments without compost. None of the differences were statistically significant ($\alpha = 0.05$).

4. Discussion

We expected to find the lowest N_2O fluxes from the KCP plots with shrubs and compost. This expectation was based on the idea that inputs of C and N from compost and from leaf litter in the shrub and compost treatments would lead to greater availability of C, higher microbial biomass and higher rates of microbial respiration. These increases, due to greater availability of C, would increase rates of N uptake by microbes (immobilization) and thus lower rates of N transformations that lead to N_2O emission; nitrification and denitrification. Alternatively, these inputs could stimulate denitrification and associated N_2O production by providing labile C for denitrification (Morley and Baggs, 2010). Plant-derived C has been shown to stimulate rhizosphere microbial activity, leading to higher rates of nitrification and denitrification, which produce N_2O (Marschner et al., 2004). Consistent with our original expectation, the highest N_2O fluxes we observed were from the no shrub/no compost treatment, but the differences were not significant due to high variability. Further, understory plants may have multiple influences on N_2O fluxes via effects on soil microclimate, chemistry and water balance. Shrubs and herbaceous understory plants, as well as compost, have positive effects on soil moisture which in turn can regulate denitrification, leading to differences in flux rates (Gray et al., 2002; Wang et al., 2011). Treatment effects, including soil moisture effects, on soil N_2O flux were not detectable in this study, however low sample size and low frequency of measurements for each treatment may have masked treatment effects due to high natural variability in soil N_2O flux (Groffman et al., 2009; Raciti et al., 2011).

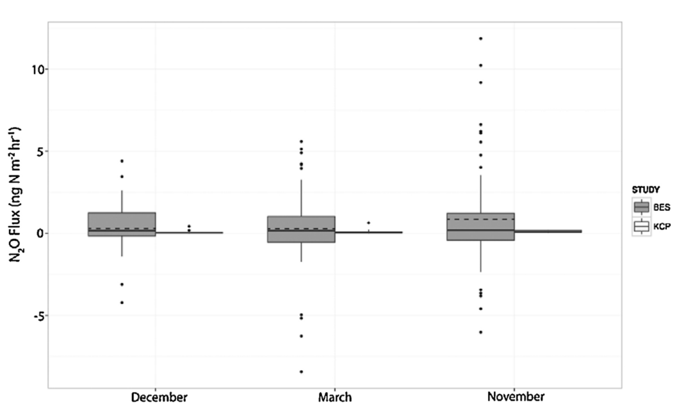

Fig. 3. Nitrous oxide fluxes from Baltimore Ecosystem Study (BES) and Kissena (KCP) sites in December, March, and November. BES values are represent $n = 185$ chamber measurements with standard error. For KCP values (with standard error), December $n = 19$; March $n = 19$, November $n = 15$. Means are shown as dashed lines ($p = 0.266$).

The trend (non-significant) of higher N_2O flux from the no shrub/no compost treatment suggests that plant and microbial uptake of inorganic N may be one regulator/inhibitor of N_2O loss in urban afforestation sites planted with shrubs, herbaceous plants, and trees through limitation of N availability and labile C substrate for N_2O production. This, however, cannot be confirmed without consideration of other variables, including temporal dynamics of soil moisture, direct measurement of labile C compounds, pH, and other factors across treatments which we did not include in this study. We suggest that the trend toward lower microbial biomass C, potential net N mineralization and nitrification in the absence of shrubs and compost may have been related to lower C inputs from shrub litter, compost and rhizodeposition, which are necessary for soil N transformations. The positive relationship between labile C availability and indices of microbial activity has been demonstrated in other studies but was not directly tested at the NY-CAP sites (Groffman et al., 2006). Lower microbial biomass N in the no shrub/no compost treatment suggests reduced immobilization of N in microbial biomass which may have fostered the flow of NH_4^+ to nitrifiers and NO_3^- to denitrifiers, facilitating N_2O production by nitrification and denitrification (Table 2). Nitrifiers are poor competitors with plants and other microbes for access to NH_4^+ and are known to thrive where root and microbial demand for N are low (Zak et al., 1990; Kaye and Hart, 1997; DeBoer and Kowalachuk 2001; Kuzyakov and Xu, 2013). The results from this study suggest that site preparation with compost and establishment of a plant community including both canopy and understory species may be one among other drivers of N_2O fluxes from soils in afforestation projects, though the significance of this site preparation remain unclear. Given the known effects of C substrate availability on denitrification activity (Senbayram et al., 2012) and the notion that natural plant community structure leads to more aerated soils, and fewer anaerobic microsites, afforestation site preparation with shrubs and compost may reduce soil habitats for denitrification (Hoben et al., 2011), though further research is necessary in order to draw conclusions that inform management decisions.

The effects of compost amendments and shrub litter inputs were most strongly seen in the no shrub/no compost treatment, where values of microbial biomass and activity were lowest (Table 2). Because the availability of organic matter has been shown to significantly influence total microbial biomass, microbial community activity and belowground C storage (Brant et al., 2006; Pandey et al., 2007; Leff et al., 2012), this relationship may also explain the comparatively high values for microbial biomass, soil respiration (CO_2 efflux) and total inorganic N (TIN) in the presence of compost (Favoino and Hogg, 2008; Ball et al., 2014). This further demonstrates that compost additions may be effective for improving soil quality and plant growth, and for minimizing N_2O emissions, in urban afforestation projects.

In addition to determining how afforestation site preparation affects soil N_2O flux, this study aimed to compare fluxes from an old (~70 years) urban forest stand to fluxes from a recently afforested urban site. This comparison allowed for assessment of how recent afforestation affects N_2O production from urban forests compared to older stands within the eastern United States. Though this comparison is not a direct one due to differences in soil texture and moisture, dominant vegetation and precipitation, we aimed to provide some context for the magnitude of N_2O flux from a more established urban forest. When compared to the BES plots, KCP plots had lower average N_2O fluxes during all months sampled, with the difference between sites not significantly different across months ($p = 0.26$). Though this reference site is not a perfect comparison due to differences in stand age, soil type and vegetation, the comparison does suggest that site preparation practices at the MTNYC sites are not causing higher fluxes of N_2O compared to fluxes from a well-established forest embedded in a comparable urban matrix. To verify this result, more frequent sampling for a longer periods of time would help to add statistical power to this comparison. Previous studies have observed increased rates of N_2O emissions from soils after afforestation (Maljanen et al., 2012) as well as reduced emissions following afforestation (Allen, 2009). Maljanen et al. (2012) concluded that low C: N ratios (<20) in soils yield the highest N_2O emissions, suggesting that additions of labile organic matter that raise soil C:N ratio can be important to mitigating N_2O emission from afforested soils. Other research indicates that stand age after afforestation significantly affects the emission of N_2O from soil (Mushirov 2010; Gundersen et al., 2012); specifically that a transition from low to high levels of GHG emissions from soils occurs within the first four decades of afforestation. The relatively lower N_2O flux from KCP plots compared to BES plots may therefore be a result of its young age compared to more established urban forests, though differences between the plots and the statistical power of the two datasets make it difficult to draw these conclusions. There is a clear need for continued monitoring of GHG fluxes at KCP and other afforestation sites to establish whether a temporal pattern in N_2O emissions exists following urban afforestation. This and other studies of the effects of urban afforestation on soil N_2O production would benefit from comparisons to other land-uses and afforestation in other ecosystems, however comparable measurements are currently scarce in the literature.

5. Conclusions

Of the four treatments tested, we found no significant effects on losses of N_2O in this study. The results suggest that the absence of a natural vegetation structure, including canopy, shrub and herbaceous species, and compost at the time of site preparation may be one among other drivers of N_2O flux to the atmosphere and the variables measured in this study were not sufficient to discern the main influences of N_2O production under urban afforestation conditions. We conclude that efforts to design afforestation projects to support natural vegetation structure with amendments of compost may tighten C and N cycles in urban ecosystems by increasing internal cycling of N and reducing N losses via N_2O emissions, however the data presented here are not sufficient to draw conclusions for site preparation decisions. Losses of N_2O from the NY-CAP and other urban afforestation projects should be monitored in the future to determine the particular mechanisms controlling N_2O production in differing urban forests. Further monitoring of soil biological processes, along with labile C availability, soil moisture, N_2O flux, and emissions of other GHG over longer time periods will help provide clearer results and provide a better understanding of the dynamics of newly created forests that are appearing in cities around the world.

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