

## Urbanization effects on soil nitrogen transformations and microbial biomass in the subtropics

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**Abstract** As urbanization can involve multiple alterations to the soil environment, it is uncertain how urbanization effects soil nitrogen cycling. We established 22–0.04 ha plots in six different land cover types—rural slash pine (*Pinus elliottii*) plantations ( $n=3$ ), rural natural pine forests ( $n=3$ ), rural natural oak forests ( $n=4$ ), urban pine forests ( $n=3$ ), urban oak forests ( $n=4$ ) and urban lawns ( $n=5$ ) to investigate how net soil nitrogen mineralization rates and soil microbial biomass differed between urban forests and rural forests and between urban forests and urban lawns in the Florida panhandle. Urban forest sites have 2.5 times as much net total nitrogen mineralized than rural forest sites based on the mean daily rates averaged over the 2 years study (2010–2012). Urbanization may increase soil microbial biomass and activity (potential carbon mineralization rates) and this may be influencing the soil nitrogen mineralization rates in the forest sites. To include an urban lawn (turfgrass) component in the study, one time measurements of soils from the aforementioned forest sites and from urban lawn sites (no fertilization, no irrigation) were collected in 2012. Urban forest sites and urban lawns sites do not differ in their potential carbon mineralization rates, potential net total nitrogen mineralization rates or microbial biomass carbon and nitrogen contents. However, lawns have a higher potential net nitrification rate compared to urban forests.

**Keywords** Nitrogen mineralization · Nitrification · Microbial biomass · Urbanization · Turfgrass · Forest

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## Introduction

The south is forecasted to lose between 4.4 and 9.3 million hectares of forest land to urban uses from 1997 to 2060 (Wear 2013). It is uncertain how much of this new urban land will be dominated by urban greenspace (urban lawns and urban forests), but it is likely significant. Turfgrass land cover (residential, commercial, and institutional lawns, parks, golf courses and athletic fields) accounts for an estimated 1.9 % of the total continental United States area (Milesi et al. 2005), whereas urban lands account for an estimated 3.5 to 4.9 % (Nowak et al. 2001). Since nitrogen (N) is a necessary plant nutrient as well as a potential water contaminant, it is important to understand how urbanization impacts N cycling and, specifically, soil N mineralization and immobilization rates.

One of the most important microbial mediated processes in surface soils is N mineralization and immobilization. Soil net N mineralization rates indicate the availability of nitrogen to plants and are strongly correlated to plant productivity (Reich et al. 1997). Furthermore, the net production of nitrate can result in ecosystem loss of N since nitrate can be lost via leaching or denitrification. Microbial immobilization of N has been implicated in conserving this nutrient when plant growth is minimal during the spring in the northern United States (i.e., the “vernal dam” hypothesis, Zak et al. 1990) and after disturbance from timber harvesting (Vitousek and Matson 1984) and hurricanes (Rice et al. 1997) in the southeast United States.

The focus of this research is to gain insight into how urbanization impacts net N mineralization, immobilization and nitrification in surface soils. The effects of urbanization on soils and nutrient cycling can be divided into indirect and direct effects (Pouyat et al. 2003). The indirect effects of urban land use change involve changes in the abiotic and biotic environment, such as higher temperatures, increased carbon dioxide concentrations, N deposition, and invasion of exotic species. Indirect effects of urbanization can leave soils and vegetation communities in an urban matrix physically undisturbed. The direct effects of urbanization can drastically modify soils through removal of the topsoil, additions of fill material and clearing of native vegetation. Another direct effect of urbanization is in post-development management practices of urban land cover, such as the mowing, irrigation, and fertilization of lawns (Pouyat et al. 2003). Our approach is to study the indirect effects of urbanization by comparing unmanaged forest fragments within the urban matrix (i.e., urban forests) to nearby rural forests and to study the direct effects of urbanization by comparing urban forests to urban turfgrass dominated land cover (i.e., urban lawns).

Currently, no consensus exists on how soil N mineralization and nitrification rates differ between urban and rural forests. Urban remnant forest soils in the New York City Metropolitan Area have similar (Pouyat et al. 1997; Zhu and Carreiro 1999; Pouyat and Turechek 2001) and higher (Zhu and Carreiro 2004) net N mineralization rates compared to rural forest soils. Furthermore, net nitrification rates within these urban forest soils were found to be similar (Pouyat et al. 1997) and higher (Zhu and Carreiro 1999, 2004; Pouyat and Turechek 2001) compared to rural forest soils. Comparisons were made using rates expressed on a mass of soil organic matter (SOM) basis, i.e.,  $\text{mg N kg}^{-1} \text{ SOM day}^{-1}$ . Differences between urban and rural sites in these studies were likely driven by the higher abundance of exotic earthworms in the urban sites. In Asheville, North Carolina, the higher net total N mineralization rates in urban forests were attributed to warmer temperatures in the urban sites compared to rural forested sites (Pavao-Zuckerman and Coleman 2005). However, net nitrification rates between the urban and rural forested sites were similar. Lastly, nitrogen cycling in remnant forests within

and surrounding the Baltimore Metropolitan area was determined more by soil parent material and land use history than the extent of urbanization (Groffman et al. 2006).

Potential net nitrification in the surface soils of turfgrass systems has been found to be significantly higher than nearby urban remnant forests in studies from Baltimore, Maryland (Groffman et al. 2009) and Pinehurst, North Carolina (Shi et al. 2006). Furthermore, urban grasslands in Baltimore lose more N via nitrate leaching compared to urban forests (Groffman et al. 2009). However, lawn soils can have an incredible capacity to retain N (Raciti et al. 2008; Groffman et al. 2009). Therefore, the conversion of land into turfgrass dominated systems has the potential to be a powerful driver of local and regional change in the N cycle (Groffman et al. 2009).

Very little research exists on how urbanization alters net N mineralization and immobilization in the subtropical regions of southeastern United States. This study examined how urbanization alters N cycling in sandy soils along the Florida panhandle when urbanization occurs at low population densities. To investigate the indirect effects of urbanization on N cycling, microbial biomass, net soil N mineralization rates, net nitrification rates, and net ammonium production were measured in-situ over a 2 years period in the surface soils of urban and rural forested land covers. In addition, one time measurements of forested soils were taken to measure potential net N mineralization, potential carbon (C) mineralization rates (a metric of microbial activity) and microbial biomass under optimal moisture and temperature conditions in the laboratory. To examine the direct effects of urbanization, one time measurements of surface soils from urban lawn sites were collected at the same time as those from the aforementioned forested sites. Potential net N and C mineralization rates and microbial biomass were also measured on the urban lawn soils. In the context of this study, rural forests include both naturally regenerating forest (hereafter “natural forest”) and pine plantation land covers. Natural forest and urban land covers were categorized by their dominant overstory vegetation (oak or pine), as pine flatwoods and hardwood hammocks are both common plant communities in the area.

## Methods

### Study area and site description

The study was conducted within and outside the city limits of Apalachicola and Eastpoint, Franklin County, Florida, from 2010 to 2012. In 2010, Apalachicola and Eastpoint had low population densities (less than 500 people km<sup>-2</sup>) and had populations of 2,242 and 2,337 people, respectively (US Census Bureau 2010). Both cities were within watersheds that had impervious surface percentages that did not exceed 15 % of the watershed land area (Nagy et al. 2012). Apalachicola historically receives 143.5 cm of precipitation per year (National Climatic Data Center 2012); however, drought conditions persisted throughout the region from 2010 to 2012 (Enloe et al. 2015).

The land covers chosen for this study are a subset of land covers previously described for the region (Nagy et al. 2014) that occur on an urbanization gradient (urban lawn, urban forest, natural forest). The land cover of pine plantation is also included, as it is the dominant land cover by area in the region (Nagy et al. 2014). In July 2010 seventeen circular plots (0.04 ha) were established in the study area that included urban forest oak dominated ( $n=4$ ), urban forest pine dominated ( $n=3$ ), natural forest oak dominated ( $n=4$ ), natural forest pine dominated ( $n=3$ )

and pine plantation ( $n=3$ ) land covers. Five sites were added in June 2012 to incorporate the urban lawn land cover into the study. Urban lawn and urban forest sites are located within residential areas of Apalachicola and Eastpoint. Natural forest and pine plantation sites are located in rural forested areas between 2.4 and 6.5 km outside of city limits.

Sites within each land cover were located across a similar range of hill slope positions and soil drainage classifications typical of the area (Enloe et al. 2015). All study sites exhibited no recent soil disturbance and were located on marine terraces (no floodplain locations). Soils are derived from sandy marine sediments (Sasser et al. 1994) and are a mix of Entisols, Inceptisols and Spodosols (Enloe et al. 2015). All forest sites had complete canopy closure and the natural forest and pine plantation sites had not been burned in the last 5 years. Six of the urban forest sites are likely urban remnant forests (never been cleared for urban use) (Zipperer 2002). The seventh urban forest site (oak dominated) is a regenerated urban forest and had been cleared for residential use, then abandoned, and reverted to a tree-covered site by 1969 (Enloe et al. 2015).

Slash pine (*P. elliotii*) is the dominant overstory tree on the pine dominated forested sites, whereas live oak (*Quercus virginiana* Miller), sand live oak (*Q. geminata* Small), laurel oak (*Q. hemisphaerica* Bartram ex Willdenow), swamp laurel oak (*Q. laurifolia* Michaux) and/or water oak (*Q. nigra* Linnaeus) are common overstory species on the oak dominated sites. Camphortree (*Cinnamomum camphora* (L.) J. Presl) is a non-native tree species found within the urban forests and not within the natural forests and pine plantations observed in this study. Urban lawns are a mix of warm season turfgrass and weedy broadleaf species and contained no more than 6.3 tree stems  $\text{ha}^{-1}$ . The two dominant turfgrass species in the urban sites are Bermudagrass (*Cynodon dactylon* (L.) Pers.) and St. Augustinegrass (*Stenotaphrum secundatum* (Walter) Kuntze). At the time of sampling, the lawn sites were not irrigated, fertilized or limed, but were mowed approximately once every month.

A description of forest characteristics and surface soil properties for the study sites are given in Enloe et al. (2015) and Enloe (2014). Standing crop biomass, stem density, aboveground net primary productivity (aNPP), surface soil C and N content and soil pH data (Supplementary Table 1) were measured during the study period and reported in these studies.

### Soil net N mineralization rates

Samples to measure soil N mineralization rates for the forested sites were processed over a 2 years period from July 2010 through May 2012. Lawn sites were not sampled during this time period. Net N-mineralization and nitrification were determined using an in-situ, buried bag method (Hart et al. 1994b). Within each of the forested sites, two soil cores were extracted to a depth of 7.5 cm. Soil from each core was split into two polyethylene bags. One of these bags was immediately reburied and incubated for 25 to 30 days and the other bag was brought back to Auburn University. Pre- and post-incubation samples were immediately processed for ammonium and nitrate. Processing consisted of passing the soil through a 2 mm sieve and extracting 10 g of the sieved field moist soil in 100 mL of 2 mol  $\text{L}^{-1}$  KCl. The extracted KCl solutions were filtered and then frozen until the extracts were analyzed using standard colorimetric techniques and a microplate reader (Sims et al. 1995). Bulk density for the surface (0 to 7.5 cm) soil depth was calculated for each of the forested sites (Blake and Hartge 1986).

Soil net N mineralization rates were calculated as the accumulation of nitrate and ammonium during the incubation and were reported in terms of N as ammonium ( $\text{NH}_4\text{-N}$ ) mineralized, N as nitrate ( $\text{NO}_3\text{-N}$ ) produced and total N mineralized ( $\text{NH}_4\text{-N} + \text{NO}_3\text{-N}$ ) on a gram N

hectare<sup>-1</sup> day<sup>-1</sup> (g N ha<sup>-1</sup> day<sup>-1</sup>) basis. In-situ net nitrogen mineralization rates were measured 12 times over the study period and were averaged across all sampling periods.

### Potential soil carbon and net nitrogen mineralization rates

Potential C and net N mineralization rates were determined using procedures of Robertson et al. (1999) and Hopkins (2008). Two surface soil samples (0–7.5 cm) were collected from forested and urban lawn sites in June 2012. Soils were passed through a 2 mm sieve. In July 2012, field moist sub-samples (35 g on an oven dried basis) were moistened to field capacity and incubated in the dark at 25 °C for 30 days in 1 L canning jars that also contained a 20 mL vial of deionized water to maintain humidity. To estimate potential net N mineralization rates, soil ammonium and nitrate were measured on pre- and post-incubated samples using the same techniques for the in-situ net N mineralization estimates. As for the C mineralization rates, 1 mol L<sup>-1</sup> NaOH traps were added after a one week settling period had passed in the incubation chamber and potential C mineralization was measured over 23 days. The settling time was incorporated into this study in order to avoid the flush of carbon dioxide (CO<sub>2</sub>) that occurs from sieving and moistening soil (Hopkins 2008). Evolved CO<sub>2</sub> was determined by titration of the alkali trap with 1 mol L<sup>-1</sup> HCl. Potential C mineralization rates were calculated as the amount of C (as CO<sub>2</sub>) evolved over the 23 days (g C-CO<sub>2</sub> kg<sup>-1</sup> soil).

### Soil microbial biomass carbon and nitrogen

Soil microbial biomass was estimated using the chloroform-fumigation method (Vance et al. 1987). Soil sub-samples were taken from the sieved, field moist pre-incubated soil samples collected for in-situ net N mineralization measurements (October 2010 through May 2012) of the forested sites and for the potential net N mineralization measurement (June 2012) of the forested and lawn sites. Fumigated samples were exposed to chloroform for 24 h. Fumigated and unfumigated samples (18.5 g) were extracted with 100 mL 0.5 mol L<sup>-1</sup> K<sub>2</sub>SO<sub>4</sub>. The extracts were filtered and then frozen. After thawing, the samples were analyzed for organic C and N using a Shimadzu TOC-V and total N combustion analyzer (Shimadzu Scientific Instruments, Columbia, MD). The differences between fumigated and unfumigated samples represented microbial N and C (in g g<sup>-1</sup> soil). For the forested sites, soil microbial biomass C and N were measured six times over the study period and were averaged across all sampling periods to obtain a 2 years average for each parameter. A one-time measurement of soil microbial biomass was taken for the urban lawn soils in June 2012.

### Statistical analysis

All data were analyzed by SAS version 9.2 (SAS Institute, Cary, NC). Treatment effects on soil net N mineralization rates, microbial biomass C and N content, and potential C and N mineralization rates were tested using analysis of variance with land cover as the primary treatment variable. Contrast statements were used for the following planned comparisons for differences among the forest sites (a–e) and for differences among the forested and lawn sites (f).

- a. Urban forest pine versus natural forest pine;
- b. Urban forest oak versus natural forest oak;

- c. Natural forest pine versus pine plantation;
- d. Urban sites (oak+pine) versus rural sites (natural oak+natural pine+pine plantation);
- e. Pine (urban+natural+plantation) versus oak (urban+natural); and
- f. Urban forest (oak+pine) versus urban lawn.

Contrast f is the only contrast reported for comparisons of microbial biomass C and N among land covers for the one-time soil sampling in June 2012 that included the lawn soils. Microbial biomass data from the June 2012 collection are used in the calculation of the 2 years average microbial biomass content of the forested sites.

A Pearson product–moment correlation was used to measure the association between potential C mineralization rates and both microbial biomass C and net N mineralization rates for the one-time soil sampling of forest sites and urban lawns. Pearson correlations and differences among means for the planned comparisons were considered statistically significant at  $p \leq 0.05$ . Differences with  $p \leq 0.10$  were reported for informational purposes. Normality was tested under the null hypothesis (no differences in land use categories). Normality tests failed to reject the null hypothesis. Therefore, it is reasonable to assume that each land cover is normally distributed. In select circumstances, the data were logarithmically transformed to meet the assumptions of equality of variance.

## Results

### Urban and rural forests

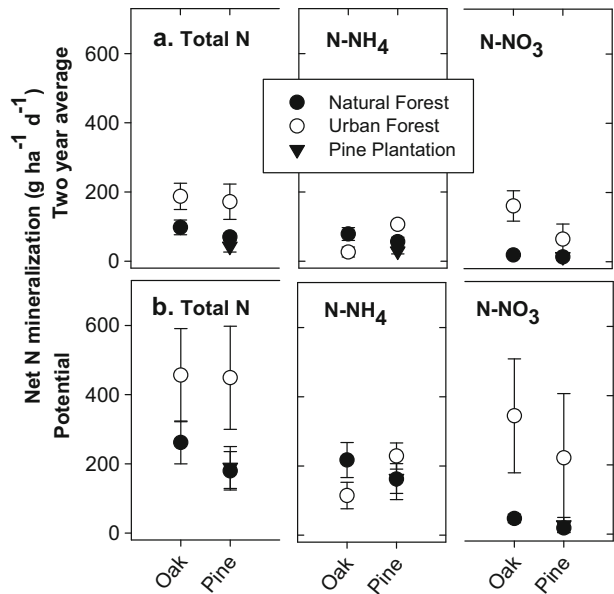
The 2 years average total net N mineralization rates in the upper 0 to 7.5 cm of soil range from 42.8 g ha<sup>-1</sup> day<sup>-1</sup> for pine plantation sites up to 187.2 g ha<sup>-1</sup> day<sup>-1</sup> for urban forest oak dominated sites (Table 1, Fig. 1). The total net N mineralization rates for slash pine plantation sites in this study are similar to those rates estimated for surface soils in slash pine plantations in Louisiana using in-situ soil incubations (44.2 g ha<sup>-1</sup> day<sup>-1</sup>, Discus and Dean 2008). Total net N mineralization rates, net production of ammonium and net nitrification rates did not differ ( $p > 0.10$ ) between pine plantation sites and natural forest pine dominated sites in this study.

**Table 1** Soil net N mineralization rates and microbial biomass averaged over a 2 years period (2010–2012) for each of the forested land covers located in western Florida. Standard errors of the means are in parentheses

Land cover	Net N mineralization rate <sup>a</sup>			Microbial biomass <sup>a</sup>		
	(g N ha <sup>-1</sup> day <sup>-1</sup> )			(μg g <sup>-1</sup> soil)		Ratio
	N-total	N-NH <sub>4</sub>	N-NO <sub>3</sub>	C	N	C to N
Natural forest oak	97.5 (21.2)	78.4 (18.6)	19.1 (3.4)	135.3 (18.6)	16.9 (2.1)	9.3 (2.4)
Urban forest oak	187.2 (38.1)	26.4 (14.6)	160.8 (44.0)	238.6 (48.7)	42.3 (13.5)	6.4 (1.0)
Natural forest pine	69.8 (10.8)	56.2 (12.8)	13.5 (3.1)	277.9 (19.8)	38.1 (6.3)	7.6 (1.0)
Urban forest pine	171.6 (51.0)	106.2 (8.2)	65.3 (43.4)	474.0 (129.2)	54.7 (18.5)	8.7 (0.8)
Pine plantation	42.8 (16.4)	28.6 (7.7)	14.2 (9.3)	276.0 (73.4)	24.8 (7.4)	13.6 (4.0)

<sup>a</sup> Measured in the 0 to 7.5 cm mineral soil

**Fig. 1** Net N mineralization rate ( $\text{g ha}^{-1} \text{ day}^{-1}$ ) as total N mineralized, N as ammonium ( $\text{NH}_4\text{-N}$ ) and N as nitrate ( $\text{NO}_3\text{-N}$ ) for forest soils (a) averaged over 2 years and (b) during a laboratory incubation study. Data from lawn sites are not shown. Significant differences are not shown. Vertical bars represent one standard error from the mean



Both urban forest oak dominated sites and urban forest pine dominated sites are trending towards higher total net N mineralization rates compared to their natural forest oak ( $p=0.0503$ ) and natural forest pine ( $p=0.0537$ ) dominated counterparts, respectively (Table 2). In particular, urban forest oak sites have significantly higher net nitrification rates compared to natural forest oak sites ( $p=0.0033$ ). Nitrification is a main component of total N mineralized in the urban forest oak sites, with 79 % of the net N mineralized being nitrified in the urban forest oak sites. In comparison, twenty one percent of the net N mineralized is nitrified in the natural forest sites (both oak and pine dominated) and 31 % in the urban forest pine sites. In regard to ammonium mineralization rates, the pine and the oak sites have opposing urbanization trends. The oak sites exhibit a significant decrease in net ammonium production rates in the urban forests compared to the natural forests ( $p=0.0169$ ), whereas the urban forest pine sites have significantly higher net ammonium production rates compared to the natural forest pine dominated sites ( $p=0.0396$ ). Similar trends are observed among the forested covers for the potential net N mineralization rates as those for the 2 years mean net N mineralization rates (Table 2, Fig. 1).

Urban forest oak dominated forests are trending towards higher microbial biomass C and N contents (2 years average) compared to natural forest sites ( $p=0.0650$  and  $p=0.0550$ , respectively; Tables 1 and 2). Urban forest pine sites have higher mean values of microbial biomass C and N content, however the differences are not significant. No significant differences are observed between pine and oak dominated forested sites and between natural forest pine sites and pine plantation sites in regards to microbial biomass contents (2 years average) and potential C mineralization rates (one-time measurement). Potential C mineralization rates in this study ( $5.8$  to  $10.2 \text{ mg C-CO}_2 \text{ kg}^{-1} \text{ soil day}^{-1}$ , Supplementary Table 2) are similar to those found for surface soils in pine plantations ( $3.8 \text{ mg C-CO}_2 \text{ kg}^{-1} \text{ soil day}^{-1}$ ), upland forests ( $4.0 \text{ mg C-CO}_2 \text{ kg}^{-1} \text{ soil day}^{-1}$ ) and urban areas ( $5.8 \text{ mg C-CO}_2 \text{ kg}^{-1} \text{ soil day}^{-1}$ ) in northern Florida (Ahn et al. 2009). Potential C mineralization rates measured for all of the sites (forested



**Table 2** Results of the contrast statements for differences among land covers in western Florida for net N mineralization rates, potential net N mineralization rates, microbial biomass C and N content, and potential C mineralization rates

Response Variable	Contrast	Point Estimate <sup>a</sup>	<i>p</i> -value
Net N mineralization rate, 2 years average (g N ha <sup>-1</sup> day <sup>-1</sup> )			
N-total	UFO-NFO	89.6	0.0503
	UFP-NFP	101.8	0.0537
	Urban forest - Rural forest	109.4	0.0027
N-NH <sub>4</sub>	UFO-NFO	-52.0	0.0169
	UFP-NFP	50.0	0.0396
N-NO <sub>3</sub>	UFO-NFO	141.6	0.0033
	UFP-NFP	51.3	0.0775
	Urban forest - Rural forest	97.4	0.0029
Potential net N mineralization rate, incubation study (g N ha <sup>-1</sup> day <sup>-1</sup> )			
N-total	UFP-NFP	269.0	0.0314
	Urban forest-Rural forest	242.2	0.0063
N-NH <sub>4</sub>	UFO-NFO	-102.0	0.0818
	Lawn-Urban forest	-178.5	0.0013
N-NO <sub>3</sub>	UFO-NFO	296.8	0.0529
	Urban forest-Rural forest	251.4	0.0229
	Lawn-Urban forest	298.9	0.0222
Microbial biomass, 2 years average (μg g <sup>-1</sup> soil)			
C	UFO-NFO	103.3	0.0650
	Urban forest-Rural forest	126.5	0.0522
	Pine forest - Oak forest	155.8	0.0058
N	UFO - NFO	25.5	0.0550
	Urban forest-Rural forest	21.9	0.0519
Microbial biomass, incubation study <sup>b</sup> (μg g <sup>-1</sup> soil)			
C to N ratio	Lawn-Urban forest	-2.04	0.0125
Potential C mineralization, incubation study (mg C kg <sup>-1</sup> day <sup>-1</sup> )			
	Urban forest-Rural forest	3.77	0.0966

*NFO* natural forest oak, *NFP* natural forest pine, *PP* pine plantation, *UFO* urban forest oak, *UFP* urban forest pine

Only contrasts with a *p*-value ≤ 0.10 are reported

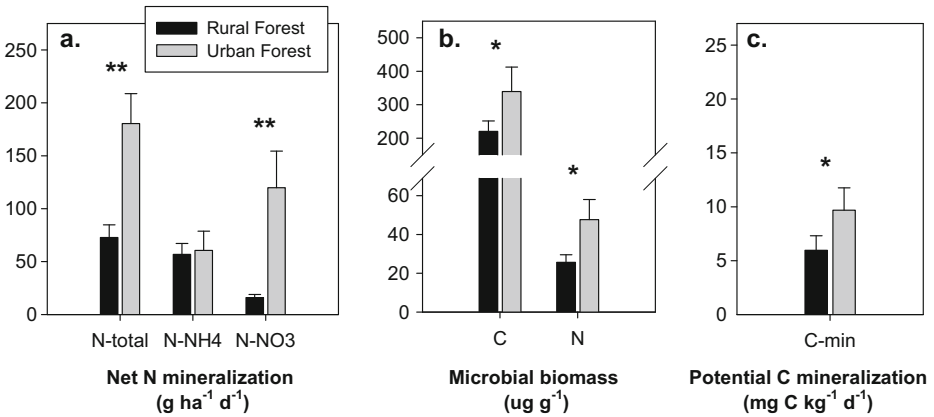
<sup>a</sup> Point estimate of the difference between land covers

<sup>b</sup> Includes the contrasts between urban lawn and urban forest only

and lawn) in this study are strongly and positively correlated with potential net N mineralization rates ( $r=0.8555$ ,  $p<0.0001$ ) of incubation soil samples.

To understand overall distinctions between urban and rural sites, urban forest sites (pine and oak dominated) are compared to rural forest sites (pine plantation and natural oak and pine dominated forests). The total net N mineralization rates and net nitrification rates for urban forested sites are significantly larger than those of rural sites ( $p=0.0027$ ;  $p=0.0029$ , respectively) (Table 2, Fig. 2a). Based on the 2 years averages of daily net N mineralization rates, urban forest sites are found to have 2.5 times as much net total N mineralized and 7.5 times as



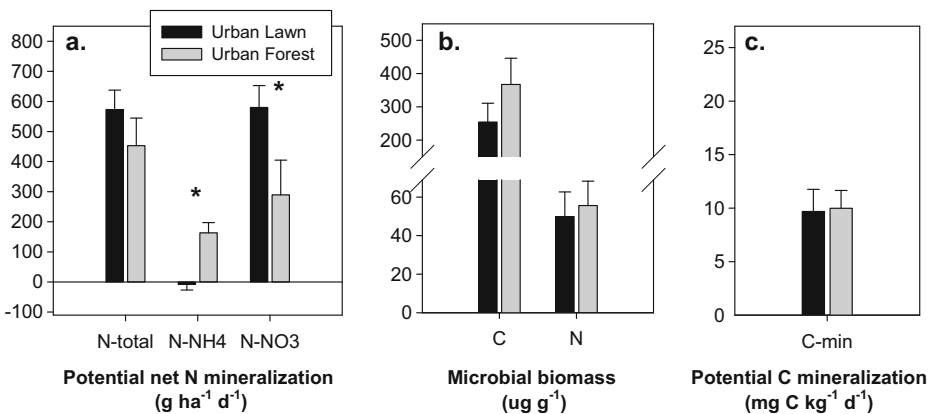


**Fig. 2** Results of contrast statements comparing rural forests (natural forest oak and pine and pine plantation) to urban forests (oak and pine dominated) for (a) net N mineralization rates and (b) microbial biomass C and N content averaged over the study period and for (c) potential C mineralization rates measured during a laboratory incubation study. Significance is indicated by asterisks:  $p \leq 0.05$  (\*) and  $0.05 < p < 0.10$  (\*\*). Vertical bars represent one standard error from the mean

much N as nitrate produced than rural forest sites. Urban forested sites are trending towards larger microbial biomass C and N contents in the upper 7.5 cm of soil compared to the rural sites ( $p=0.0522$  and  $p=0.0519$ , respectively) based on an analysis of the 2 years averages (Table 2, Fig. 2b). One time measurements of potential C mineralization rates indicate that urban forests are trending towards higher rates of microbial activity compared to rural forests ( $p=0.0966$ , Table 2, Fig. 2c).

**Urban lawns**

Urban lawns have similar potential net total N mineralization rates (Fig. 3a), similar microbial biomass C and N contents (Fig. 3b) and similar potential C mineralization rates (Fig. 3c,



**Fig. 3** Results of contrast statements comparing urban lawns to urban forests (oak and pine dominated) for (a) potential net N mineralization rates, (b) microbial biomass C and N content, and (c) potential C mineralization rates measured during a laboratory incubation study. Significance is indicated by astrisks:  $p \leq 0.05$  (\*) and  $0.05 < p < 0.10$  (\*\*). Vertical bars represent one standard error from the mean

Supplementary Table 2) to urban forest (pine and oak dominated) soils. Urban lawns have higher microbial C to N ratios than the urban forest sites ( $p=0.0125$ , Table 2). Compared to urban forest sites, urban lawns have significantly lower rates of potential net ammonium production ( $p=0.0013$ ) and higher rates of potential net nitrate production ( $p=0.0222$ , Table 2, Fig. 3a). Urban lawns displayed little to no potential net ammonium production, which indicates that soil microorganisms immobilized ammonium that was mineralized and/or converted ammonium to nitrate.

## Discussion

### Indirect effects of urbanization: Urban versus rural forests

Urban forest sites are found to have 2.5 times as much net total N mineralized than rural forest sites. This indicates that plant available N in the surface soils of urban forests is significantly higher than that of rural forest soils. The urban forests sites have significantly higher above-ground net primary productivity compared to rural forest sites (Supplementary Table 1, Enloe et al. 2015). As most of the N stored in soil is in an organic form and unavailable for plant uptake, soil microorganisms are key to the production of plant available N and to plant productivity. Urban forests are trending towards higher microbial biomass contents (2 years average) and activity (potential C mineralization rates, one time measurement) compared to rural forests ( $p<0.10$ ).

Urbanization can alter the abiotic and biotic soil environment through several means, including the urban heat island effect, N deposition, and the introduction of exotic species, which may have contrasting effects on microbial activity and function. As this project was designed to measure the impacts of urbanization, we can only speculate as to the drivers of these impacts. Urban forested sites in this study have warmer winter temperatures and narrower temperature ranges in the forest floor compared to rural sites (Enloe et al. 2015). The urban heat island effect has been thought to increase net N mineralization rates in urban forest soils compared to rural forest soils (Pavao-Zuckerman and Coleman 2005; Chen et al. 2010). It is unclear whether urban sites in this study received additions of N from dry and wet deposition. Increased inputs of N to soils have been found to reduce soil microbial biomass and activity (Fisk and Fahey 2001; Ramirez et al. 2012). In contrast, soil nitrification rates commonly increase after invasions by exotic plants (Ehrenfeld 2003). In this study, the presence of the invasive camphortree is more apparent in the urban oak versus the urban pine dominated forests. Camphortree was not observed in the natural forest sites. Net nitrification rates are higher in urban forest oak sites compared to natural forest oak sites ( $p=0.0033$ ), but are only trending towards higher rates in the urban forest pine sites compared to the natural forest pine dominated sites ( $p=0.0775$ ).

Cause and effect interpretations regarding N mineralization rates and microbial activity and factors that can increase microbial activity (i.e., urban heat island) should be made with some caution as net N mineralization is a net process whereas C mineralization is a gross process. Net N mineralization rates do not account for the N that is released and then immobilized within the microbial biomass and therefore may not always correlate with C mineralization rates (Giardina et al. 2001; Hart et al. 1994a) as gross N mineralization rates do (Hart et al. 1994a). Potential C mineralization and potential net N mineralization rates are strongly and positively correlated in this study ( $r=0.8555$ ,  $p<0.0001$ ). Additional research would be needed to clarify how gross N mineralization rates and immobilization rates change due to urbanization.

## Direct effects of urbanization: Urban forests versus urban lawns

A change in vegetation cover from forest to lawn did not alter potential soil total net N mineralization rates, but it did strongly impact net nitrification rates and microbial C to N ratios. The lower C to N ratio of microbial biomass in the surface soils of the urban lawns compared to those in the urban forests may indicate a more active N-cycling soil microbial community in the urban lawns (Raciti et al. 2011b). Potential net nitrification rates in the unfertilized lawns (based on current management practices) in this study are significantly higher than those in urban forests ( $p=0.0274$ ). Urban lawn soils have been found to have higher rates of potential net nitrification compared to urban remnant forest soils in Baltimore, Maryland (Groffman et al. 2009; Raciti et al. 2011b). Soils collected from golf course fairways in Pinehurst, North Carolina, also had higher potential net nitrification rates than soils in nearby undisturbed pine (*Pinus palustris* Miller) stands (Shi et al. 2006). The urban lawns in these studies from Baltimore and Pinehurst included sites that were fertilized with N. However, fertilizer addition was not a significant predictor of available nitrate in lawn soils in Baltimore (Raciti et al. 2011b).

It is unclear if urban lawns soils have higher rates of net nitrification compared to urban forests due to higher rates of gross nitrification or to lower rates of microbial nitrate immobilization or a combination of both. There is some indication that the rates of gross nitrification may be higher in these sites, as urban lawn sites have a mean surface soil pH of 5.8 (Supplementary Table 1), which is on average  $1.8\pm 0.5$  (95 % confidence interval,  $p<0.0001$ ) units higher than those found for urban forest soils (Enloe 2014). The population of chemoautotrophic nitrifiers expands rapidly when soil acidity is corrected (Fisher and Binkley 2000), and this may be the cause of the higher potential net nitrification rates in the urban lawn sites compared to the urban forest sites.

The lawn sites in this study have been under this land cover since the early 1950's (Enloe 2014). Before the 1970's, local fill material was commonly used in urban areas, especially for roads and home foundations and driveways, in the study area. Local fill material was largely composed of oyster (*Crassostrea virginica*) shells and came from two sources: (1) shell middens that were scattered throughout the study area prior to European settlement and (2) waste shells from the local seafood industry (Joseph Schuster personal communication). Currently, there is little visual evidence of shell material in the lawn soils. Coarse fragments composed less than 5 % by volume of the soil to a depth of 90 cm. Although it is unclear what past management practice may have caused the higher soil pH in the lawn sites, it is possible that it may have come from the incorporation of these fill materials into the urban lawn soils.

Urban lawns can have a considerable capacity for N retention (Raciti et al. 2008; Groffman et al. 2009). Nitrogen retention in lawn soils is dependent on high levels of soil organic matter and high rates of C cycling, as indicated by potential C mineralization rates and microbial biomass (Groffman et al. 2009). Other research comparing soil C cycling of lawns (often fertilized) to remnant forests in the eastern United States suggests that urban lawns can strongly resemble urban forests in soil organic C and microbial C levels (Shi et al. 2006; Groffman et al. 2009; Raciti et al. 2011a, b). Urban forest sites and urban lawns sites in this study are not significantly different in their potential C mineralization rates or one-time measurements of microbial biomass C and N contents. The urban lawn sites and urban forest sites have statistically similar soil C and N contents in their surface soils (Supplementary Table 1, Enloe 2014). Additional research in the study area also found that urban lawns have similar soil C and N contents in the surface soil (0 to 7.5 cm) compared to urban forests (Nagy et al. 2014). Urban lawns in this study may have a similar potential to that of urban forest soils

to retain N. However, their higher rates of nitrification compared to urban forests combined with their sandy soil texture imply that urban lawn soils can be susceptible to nitrate leaching.

## Summary and conclusions

Urbanization appears to have a distinct impact on net N-mineralization and nitrification rates in surface soils of a developing region in the Florida panhandle. In a one-time sampling of urban forest and urban lawn soils, lawn soils have significantly higher potential net nitrification rates compared to urban forest soils. Urban forest sites are found to have 2.5 times as much net total N mineralized than rural forest sites which indicates that plant available N in the surface soils of urban forests is significantly higher than that of rural forest soils.

Cities have a distinctly different biogeochemistry than natural systems (Kaye et al. 2006). As mentioned in the introduction, there is no consensus on how soil N mineralization differs between urban and rural forests. Soil properties and biogeochemical cycling in urban forests and rural forests have been found to be different for three cities in the eastern US that ranged over three orders of magnitude in population size. However, the degree and nature of that impact varied from city to city (Pavao-Zuckerman 2008). Hence, there is a need for city specific ecological knowledge (see Pavao-Zuckerman 2008), especially in regards to the indirect impacts of urbanization.

Nonetheless, the urban ecosystem convergence hypothesis implies that urbanization “drives ecosystem structure and function . . . toward a range of similar endpoints over time regardless of ecosystem life zone starting points” (Pouyat et al. 2003). This hypothesis has been supported by research investigating the indirect effects of urbanization on soil carbon content of urban lawns compared to native land covers or to urban remnant forests (see Pouyat et al. 2009). Work from this study as well as from Maryland (Groffman et al. 2009) and North Carolina (Shi et al. 2006) indicates that urban lawns have higher potential nitrification rates compared to urban forests. This suggests that anthropogenic drivers (i.e., management) can dominate natural controlling factors (i.e., soil parent material, climate). The lawns in this study are considered low maintenance, with minimal weed control and no current fertilizer or irrigation additions. Therefore, further work needs to be done in investigating the role of management intensity on the convergence of urban soil properties over regional scales.

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