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Methane uptake in forest soils along an urban-to-rural gradient in Pearl River Delta, South China

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We investigated soil CH_4 fluxes from six forests along an urban-to-rural gradient in Guangzhou City metropolitan area, South China. The most significant CH_4 consumption was found in the rural site, followed by suburban, and then urban forest sites. The rates of CH_4 uptake were significantly higher (by 38% and 44%, respectively for mixed forest and broadleaf forest) in the rural than in the urban forest site. The results indicate that soil water filled pore space (WFPS) is the primary factor for controlling CH_4 consumption in subtropical forests. The reductions of soil CH_4 uptake in urban forests were also influenced by the higher rates of atmospheric nitrogen (N) deposition and increases in soil nitrate (NO_3^-) and aluminum (Al^{3+}) contents as a result of urbanization. Results from this work suggest that environmental changes associated with urbanization could decrease soil CH_4 consumption in subtropical forests and potentially contribute to increase of atmospheric CH_4 concentration.

Methane contributes up to 30% to total net anthropogenic radiative forcing of the atmosphere[1]. Atmospheric CH_4 has increased 151% since 1750 as a result of an imbalance between increasing sources and decreasing sinks[2]. The largest biological sink of CH_4 is consumption by soil aerobic bacteria[3]. Upland soils (e.g., forest, grassland, and desert) have been shown to be significant sinks for CH_4[4], contributing approximately 6% of the total consumption[5]. Among upland soils, forest is probably the most efficient CH_4 sink[4]. Urbanization has increased rapidly worldwide and 70% of the world population will be urban by 2050[6]. The urban population of China rose from 18% in 1978 to 45% in 2005, and is projected to be 60% by 2030[7]. Urbanization is generally accompanied with environmental changes in chemistry and climate, which consequently impact the capacity of CH_4 consumption in urban forests[8,9].

Previous studies showed that CH_4 uptake was commonly lower in urban than in suburban and rural forests[8,10,11]. However, there was disagreement regarding the mechanisms underlying the reductions of CH_4 uptake. Goldman et al. ascribed the suppressed CH_4 uptake to lower rates of organic matter degradation and nutrient cycling caused by air pollution (especially as O_3)[8]. Groffman and Pouyat reported that increases in N deposition and CO_2 levels might be the possible mechanisms[10]. Costa and Groffman found that the differences in N cycling associated with urbanization led to a reduction in the microbial populations responsible for CH_4 uptake[12]. So far, the effects of urbanization on CH_4 uptake in tropical and subtropical forest soils remain unclear.

The objective of this study was to investigate how CH_4 fluxes from subtropical forests varied along an urban-to-rural gradient in Guangzhou City metropolitan area, and to determine what factors underlie urbanization effects on CH_4 uptake. We hypothesized that CH_4 consumption would be lowest in urban forests, followed by the suburban and rural forest sites, due to the exposure to urbanization-induced environmental changes of the former.

Results

Soil CH_4 uptake. Heishiding (HSD, rural site) forests had the highest rates of CH_4 uptake, followed by Dinghushan (DHS, suburban site) and then Maofengshan (MFS, urban site) forests (Fig. 1 a, b, all $P < 0.05$). For pine and broadleaf mixed forests (MF), the mean rate of CH_4 uptake in HSD was higher by 38% than in MFS

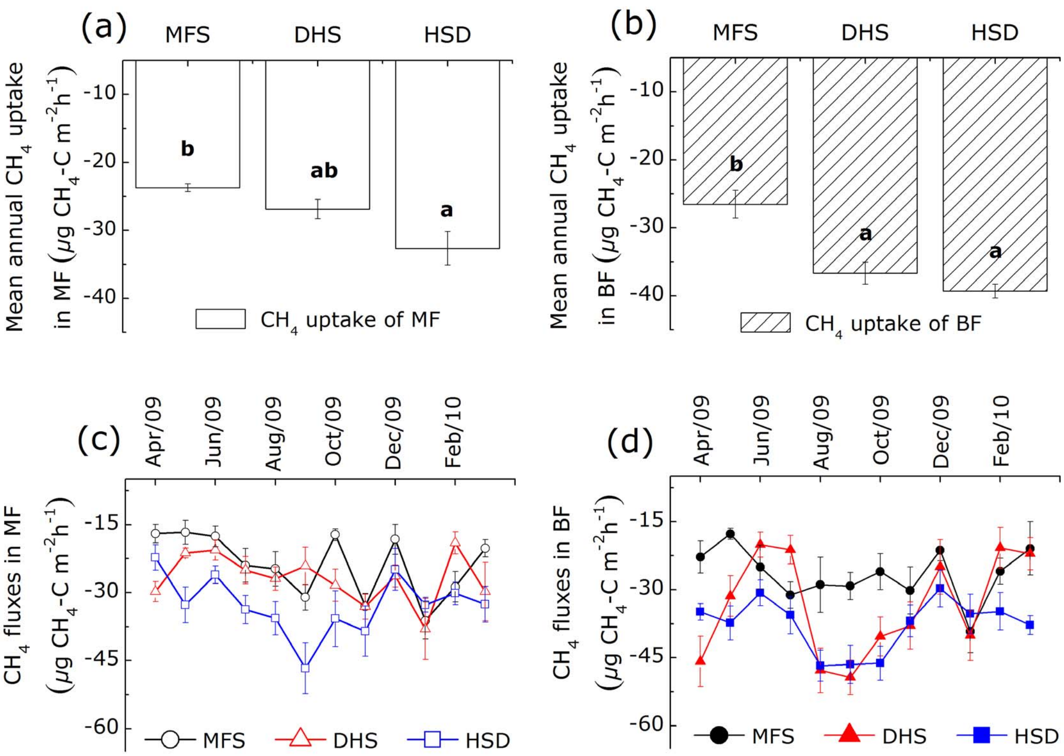

Figure 1 | Comparisons of the mean rates and seasonal patterns of CH_4 uptake. (a) and (b) depict the mean rates of CH_4 uptake, and (c) and (d) show the seasonal patterns of soil CH_4 uptake (Mean value ± 1 standard error, n = 5); Different letter "a" and "b" denote significant differences ($P < 0.05$) for the same forest type. MF, mixed forest; BF, broadleaf forest; MFS, Maofengshan; DHS, Dinghushan; HSD, Heishiding.

site (Fig. 1 a). In broadleaf forests (BF), the average rate of CH_4 uptake was higher by 37% in DHS, and 44% in HSD than that of MFS site (Fig. 1 b, all $P < 0.05$). In DHS forest site, average CH_4 uptake for BF was 37% higher than that of MF ($P = 0.04$). However, there were no differences between MF and BF stands at other forest sites. Monthly mean CH_4 uptake in all forests showed a similar seasonal pattern, with the highest consumption occurring in the fall (August to October) (Fig. 1 c, d).

Biotic and abiotic variables. The amount of N deposition in rainfall was higher in MFS and DHS than in HSD site ($P < 0.05$), with no significant difference between MFS and DHS (Table 1). Soil temperature and WFPS exhibited clear seasonal patterns (Fig. 2 a, b), similar to those of air temperature and rainfall at each site (Fig. S1). Higher soil WFPS and temperatures were found in MFS forests than that of HSD site (Fig. 2 c, d, all $P < 0.05$).

For both forest types, soil NO_3^- contents were higher in HSD than that of MFS and DHS sites, whereas soil NH_4^+ and total N (TN) showed an opposite pattern (Table 2). Soil bulk density and pH values were significantly lower in MFS and DHS than in HSD forests, conversely, soil Al^{3+} contents were higher in MFS and DHS sites (Table 2). Soil CH_4 uptake showed negatively related with soil NO_3^- and Al^{3+} contents (Fig. 3 a, c), and positively related to pH values (Fig. 3 b). Soil microbial biomass C (MBC) contents were lowest in MFS, followed by DHS and HSD for the same forest stands, whereas microbial biomass N (MBN) had no significant differences across the gradient sites (Table 2).

Discussion

Methane fluxes at all sites were predominantly negative during the study period, indicating CH_4 uptake from the atmosphere. While many studies have quantified differences in CH_4 cycling under the

Table 1 | Site characteristics along an urban-to-rural gradient in southern China

Characteristic	Site		
Site code	MFS (urban)	DHS (suburban)	HSD (rural)
Latitude and longitude	23°18′5.87″N; 113°27′0.57″E	23°8′57.27″N; 112°31′3.07″E	23°27′42.85″N; 111°54′19.78″E
Distance from urban core (km)	36.7	97.8	169.3
Altitude (m)	150	225	395
Mean slop (°)	21.2	24.5	27.3
Annual precipitation (mm)	1742	1625	1690
Average annual temperature (°C)	22.1 (0.5)	22.2 (0.4)	21.0 (0.5)
N deposition in rainfall (kg N $ha^{-1}yr^{-1}$)	32.4 (1.1) a	40.2 (1.3) a	18.6 (0.7) b
NH_4^+ : NO_3^- ratio	0.47 b	0.61 b	1.23 a
Soil type	Lateritic red earth	Lateritic red earth	Lateritic red earth

Data of temperature and precipitation were collected from nearby meteorological stations (from April 2009 to March 2010). Values of average annual temperature and N deposition in rainfall are presented as means with SE in parentheses (n = 12 and 5, respectively for temperature and N deposition). Different letter within a row presented significantly different (one-way ANOVA, $P < 0.05$). NH_4^+ : NO_3^- ratio, the ratio of NH_4^+ : NO_3^- in rainfall; MFS, Maofengshan; DHS, Dinghushan; HSD, Heishiding.

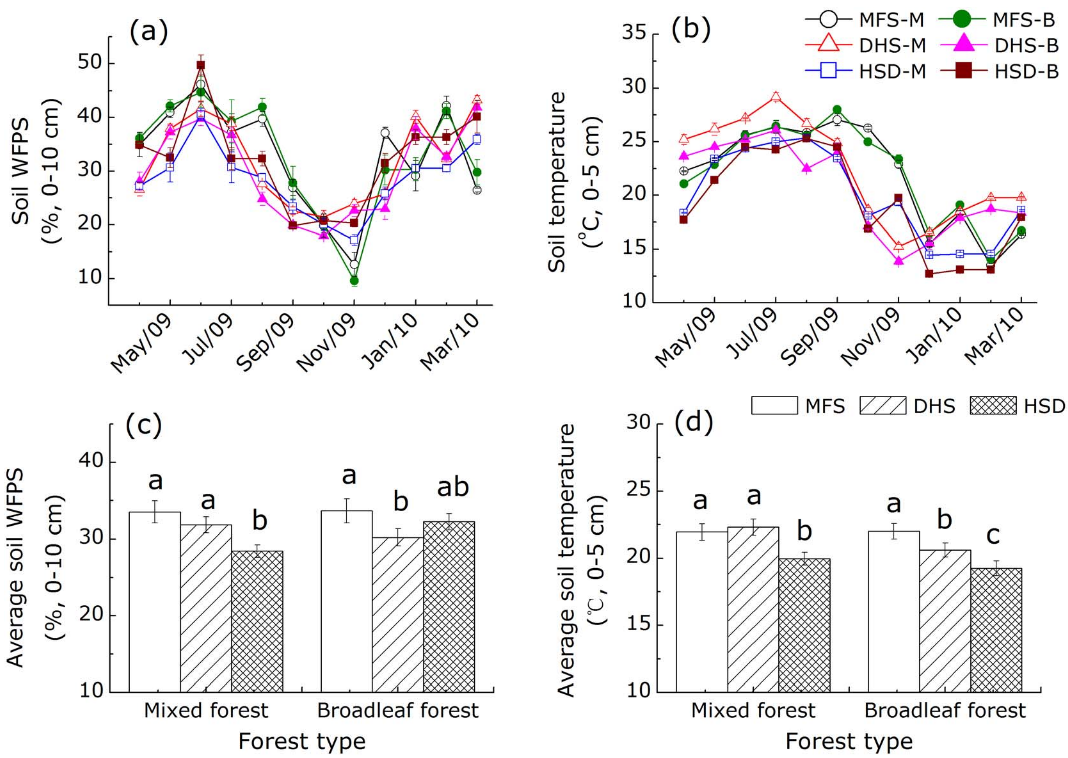

Figure 2 | Soil temperature and WFPS at forests along the urban-to-rural gradient. (a) Seasonal patterns of soil WFPS at 0–10 cm depth; (b) seasonal patterns of soil temperature at 5 cm depth; (c) annual mean soil WFPS; and (d) annual mean soil temperature (Mean value ± 1 standard error, n = 5). In panel (a) and (b), letters M and B following the site abbreviations denote mixed forest and evergreen broadleaf forest, respectively. Different letter "a" and "b" denotes significant difference between forests for the same forest type ($P < 0.05$). MFS, Maofengshan; DHS, Dinghushan; HSD, Heishiding.

changes of land uses[11], there is considerable uncertainty about the unmanaged or intact forests within urban areas acting as CH_4 sinks that have been observed[8,10,12]. The rates of CH_4 uptake in the suburban and rural forests (2.4 to 3.3 kg CH_4-C ha^{-1} yr^{-1}) were comparable with previous studies in tropical and subtropical regions of southern China (2.5 to 4.3 kg CH_4-C ha^{-1} yr^{-1})[13–15]. In the suburban (DHS) site, the soil of BF oxidized more CH_4 than that of MF stand, a trend observed in previous studies[13,16]. The BF is an old-growth forest (more than 400 years), which represents a forest type in an advanced successional stage within the study region[17]. Forest composition, species assemblages and soil density (Table S1) are more favorable than MF, leading to increased CH_4 consumption in this BF stand[4,13,16].

Consistent with our previous hypothesis, the urban forest soils had the lowest capacity of CH_4 consumption compared to rural and suburban sites. Our results were comparable with previous reports that CH_4 uptake would be markedly lower in urban than in rural forest soils. For example, Goldman et al. found that CH_4 uptake rates in intact forests in urban center of New York City (NY, USA) were 30% lower than that of rural forest soils[8]. Similar results were found in urban forests in the Baltimore metropolitan area (MD, USA)[10,11]. These results have suggested that there is an urban atmospheric effect on CH_4 consumption in urban forests[12]. In the present study, the reduction of CH_4 uptake in urban forests might be influenced by several factors as follows.

Firstly, the reduction of CH_4 uptake was primarily influenced by the lower diffusion of CH_4 into urban forest soils. Multiple regression analyses indicated that soil WFPS accounted for 52% of the variance in CH_4 uptake for both MF and BF stands (Table 3). Higher soil WFPS and bulk density in the urban forests could increase resistance to atmospheric CH_4 and O_2 transport into the soils, reducing the

Table 2 | Soil properties (0–10 cm depth) of forests along the urban-to-rural gradient

Forest type	MF				BF			
Soil properties	MFS	DHS	HSD	*Significance*	MFS	DHS	HSD	*Significance*
NH_4^+ (mg kg^{-1})	6.4(0.5) b	8.5(1.6) b	17.2(1.9) a	$P < 0.01$	9.0(1.1) b	8.8(1.0) b	16.7(2.2) a	$P < 0.01$
NO_3^- (mg kg^{-1})	17.5(1.4) a	21.7(1.5) a	8.1(1.3) b	$P = 0.01$	23.1(2.3) a	22.5(2.3) a	11.7(1.4) b	$P < 0.01$
Total N (g kg^{-1})	1.7(0.1) b	2.2(0.1) a	2.4(0.2) a	$P = 0.01$	2.2(0.1) b	2.7(0.2) b	3.2(0.1) a	$P < 0.01$
MBC (mg kg^{-1})	189(25) b	278(30) ab	294(15) a	$P = 0.02$	389(18) b	515(37) a	509(38) a	$P = 0.05$
MBN (mg kg^{-1})	31.2(4)	30.8(4)	22.7(2)	$P > 0.05$	44.2(5)	45.7(7)	37.4(4)	$P > 0.05$
pH	3.87(0.01)b	3.97(0.07) ab	4.14(0.04) a	$P < 0.01$	3.86(0.08) b	3.73(0.03) b	4.22(0.03) a	$P = 0.01$
Al^{3+} (mmol kg^{-1})	85(2) a	82(2) a	59(1) b	$P = 0.04$	101(4) a	102(3) a	69(1) b	$P = 0.03$
Bulk density (g cm^{-3})	1.2(0.1)a	1.2(0.1) a	1.0(0.2) b	$P = 0.02$	1.2(0.1) a	1.1(0.0) ab	1.0(0.2) b	$P = 0.01$

Soil samples were collected in July 2009. Values are presented as mean with SE in parentheses (n = 5). Different letters within the same row from the same forest type denote significant difference between the three sites (one-way ANOVA with Tukey's HSD, $P < 0.05$). MBC, microbial biomass C; MBN, microbial biomass N; MF, mixed forest; BF, broadleaf forest; MFS, Maofengshan; DHS, Dinghushan; HSD, Heishiding.

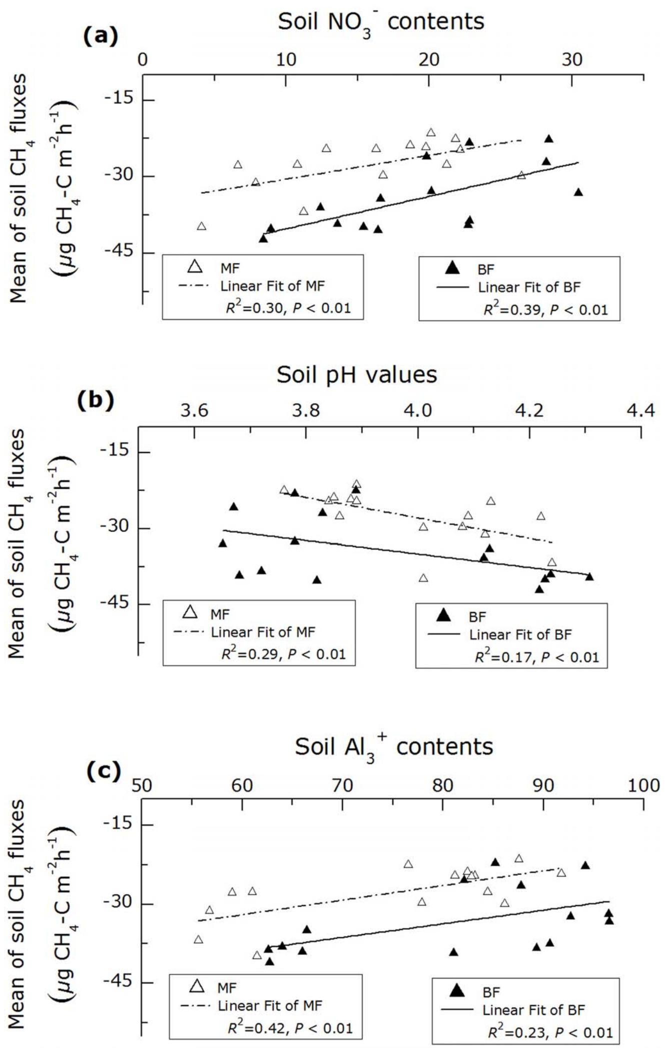

Figure 3 | Linear regressions between annual CH_4 uptake rates and soil properties. (a) soil NO_3^- contents and CH_4 fluxes, (b) soil pH values and CH_4 fluxes, and (c) soil Al^{3+} contents and CH_4 fluxes. CH_4 fluxes used in this figure were measured in field at the same day of soil sampling (July 2009). MF, mixed forest; BF, evergreen broadleaf forest.

activity of methanotrophic bacteria[4,18]. The seasonal changes in CH_4 uptake were also best explained by gas diffusion, and were most related with lower soil WFPS during the fall. There was no significant relationship between CH_4 uptake and soil temperature, which was consistent with previous studies in this region[13,15,19]. These results suggest that soil temperature is not the key factor for controlling CH_4 consumption in subtropical forests of southern China.

Secondly, higher N cycling rates and soil NO_3^- contents, which are caused by high rates of atmospheric N deposition, significantly decreased soil CH_4 uptake in the urban forests. Soil NO_3^- contents accounted for 30% of the variance in CH_4 uptake (Table 3). Previous studies have shown that NO_3^- and/or NO_2^- produced from the NO_3^- reduction were possibly toxic to CH_4-oxidizing microbes[20,21], which might be a potential role for the reduction of CH_4 consumption in our urban forests. Although we did not measure net N mineralization and nitrification rates, the fact that these results were consistent with patterns of N deposition and soil N cycling already published for this urban-to-rural gradient[22], strongly suggest that higher rates of N cycling underlie the reductions of CH_4 consumption in the urban forests[10,12]. We found that soil NH_4^+ and TN contents tended to be higher in the rural than that in suburban and urban forest sites, which was in conflict with the generally accepted idea that soil N contents tend to be higher in urban than rural forests[23]. However, our result was comparable with previous studies within the same region[22,24]. The cause may be a higher rate of N losses in the urban site[22].

Thirdly, CH_4 uptake might also be reduced by the changes in soil pH value and Al^{3+} content of the urban forests. A decrease of soil pH in urban forests might lead to decreased CH_4 uptake[25], which was noted in the Broadbalk Experiment in southeastern England[26]. Soil acidification might cause a release of heavy metals, such as Al^{3+}, inhibiting soil CH_4 uptake[26]. Al^{3+} toxicity could inhibit CH_4 uptake[27], which potentially explains the results from our study.

Finally, the suppressive effect of urbanization-induced environmental changes on CH_4-consuming bacteria might be another mechanism for the CH_4 reduction. Urban environments might decrease CH_4 uptake indirectly through changes in the habitat of the methanotrophic bacteria[28]. Long-term N deposition could cause a decrease in methanotrophic populations via niche competition with nitrifiers[29], leading to a reduction in CH_4 consumption[30]. In the present study, a significant decrease in soil MBC at the urban site compared to the rural and suburban forest sites, might negatively affect the size, composition and activity of the CH_4 oxidizing community[10,31]. The urbanization-induced environmental changes might have profound implications for CH_4 consuming communities and further research should be conducted.

In summary, the largest consumption of CH_4 was found in the rural, followed by the suburban and urban forest sites. The data presented here strongly indicated that the reduction of CH_4 uptake in urban forests was due to higher soil WFPS, and adverse effects of urbanization-induced environmental changes, such as atmospheric N deposition, higher soil NO_3^- content and Al^{3+} toxicity. This is the first *in situ* study to attempt to clarify the effects of urbanization-induced environmental changes on soil CH_4 uptake in tropical and subtropical forests. Our results suggest that the projected environmental changes associated with urbanization would decrease CH_4 consumption in urban forest soils of subtropical regions, and this reduction of CH_4 consumption needs to be considered in global CH_4 budgets.

Table 3 | Responses of CH_4 uptake rates to soil variables: multiple regression analysis and partitioning the variance in R^2

Forest type		MF			BF			MF + BF		
Y variable	X variable	R^2	P	% of R^2	R^2	P	% of R^2	R^2	P	% of R^2
CH_4 uptake	All variables	0.74	0.02		0.76	0.01		0.50	0.01	
	WFPS	0.30		41	0.41		54	0.26		52
	NO_3^-	0.32		43	0.16		21	0.15		30
	Al^{3+}	0.10		13	0.14		18	0.05		10
	pH	0.02		3	0.05		7	0.04		8

All sampling times were combined on July 2009. The relative importance of each ''X'' variable in the regression is indicated by the % of R^2 for which it accounts. WFPS are the soil water content when the soil cores were harvested. MF, mixed forest; BF, broadleaf forest. MF + BF, combined the data of the six forests.

Methods

Study area. The study sites were located along a 150 km gradient that extends from MFS (an urban site near Guangzhou City), through DHS (a suburban site), to HSD (a rural site) within the Pearl River Delta (PRD) region of Guangdong Province, South China (Fig. S2). Some characteristics of the study sites were presented in Table 1. The PRD region is one of the regions experiencing rapid urbanization with its population increasing nearly two fold from 1982 (54 million) to 2010 (104 million)[32]. The study region has a subtropical monsoon climate. There was a gradient for environmental variables from Guangzhou City to its surroundings (Table 1).

At each site, a MF and a BF were selected for experimental plots. Several criteria were used in study site selection to ensure comparability among the forests: (1) no disturbance after planting (such as fire, insect infestations, logging, and fertilization); (2) forest age between 50 and 70 year (excluding the BF in DHS); (3) soil of lateritic red earth (Table S1). The dominant species and plant characteristics are described in Table S1. Within each forest, five 10 m × 10 m plots were randomly established at least 100 m from the edge to avoid "edge effects".

Measurement of CH_4 fluxes. Soil CH_4 fluxes were measured from April 2009 to March 2010 using the static chamber method. Gas samples were collected biweekly during the growing season (April to September) and monthly at other times. The chamber design and the measurement procedure were adopted from Zhang et al[13]. Gas samples were collected with 60 ml plastic syringes at 0, 15 and 30 min after the cover chamber closure, and immediately injected into special gas bags (Yile CO., LTD, Shanghai, China). The samples were transferred to a lab and analyzed within 2–3 days by a gas chromatography with flame ionization detection (FID). Atmospheric pressure, air temperature (inside chamber), soil temperature (5 cm depth) and moisture (0–10 cm depth) were measured during each sampling event. Soil moisture was converted to WFPS (%). Details of the measurement were referenced from Zhang et al.[13].

Soil sampling and analysis. Soil samples (0–10 cm depth) were collected on July 2009. Three soil cores (3.5 cm internal diameter, ID) were collected randomly from each plot and combined to one composite sample. The samples were passed through a 2 mm sieve and divided to two parts. One subsample was used for the analysis of NH_4^+, NO_3^-, MBC, and MBN. The other was air dried at 25°C for the estimation of other chemical parameters. Gravimetric water content was determined through oven drying at 105°C for 48 h.

Soil NH_4^+ and NO_3^- contents were determined by extraction with 2 M KCl solution followed by colorimetric analysis on a flow-injection autoanalyzer (Lachat Instruments, Milwaukee, USA). TN content was determined by micro-Kjeldahl digestion[33], followed by detection of ammonium with a UV-8000 Spectrophotometer (Metash Instruments Corp., Shanghai, China). Soil MBC and MBN contents were estimated by chloroform fumigation-extraction method[34]. For each sample, two fresh soil subsamples (10 g dry soil equivalent) were prepared. One subsample was fumigated with chloroform for 24 h at 25°C. The other was treated as control. Soil MBC and MBN contents were calculated as the difference in extractable C, N between fumigated and non-fumigated subsamples, using the conversion factors of 0.33 and 0.45 for MBC and MBN, respectively[35,36].

Atmospheric N deposition. We used ion-exchange resin (IER) columns to monitor N deposition in precipitation at the three sites during the study period. The IER columns and the measurement procedure were adopted from Fang et al.[22]. The resin columns were collected with interval for three months. The concentrations of NH_4^+ and NO_3^- were determined by 2 M KCl extraction as described above.

Statistical analysis. Repeated Measures Analysis of Variance (ANOVA) was used to examine the differences of soil CH_4 fluxes among the three sites. One-way ANOVA was performed to compare the differences in soil properties, MBC, and MBN within the same forest type. Linear regression analysis was performed to quantify the relationships between CH_4 fluxes and individual soil variables. Multivariate linear regression analysis was performed using CH_4 uptake rate as dependent variable, and soil WFPS, NO_3^-, Al^{3+} contents, and pH values as independent variables. R^2 was partitioned to quantify the importance of each independent variable. All statistical analyses were conducted using SPSS 16.0 for windows (SPSS Inc., Chicago, IL, USA). Statistically significant difference was set at $P \leq 0.05$ unless otherwise stated. Mean values ± 1 standard error were reported.

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