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Effects of acetylene at low concentrations on nitrification, mineralization and microbial biomass nitrogen concentrations in forest soils

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Temperate forest surface soils at the varying distances from main trunks (e.g., *Pinus koraiensis* and *Quercus mongolica*) were used to study the effects of acetylene (C_2H_2) at low concentrations on nitrification, mineralization and microbial biomass N concentrations of the soils, and to assess the contribution of heterotrophic nitrification to nitrous oxide (N_2O) emissions from soils. The use of acetylene at partial pressures within a range from 10 to 100 Pa C_2H_2 in headspace gas gave a significant decrease in N_2O emission at soil moisture of *c*. 45% water-filled porosity space, and the decrease was almost the same in each soil after exposure of C_2H_2 at low concentrations. Heterotrophic nitrification could account for 21%—48% of total N_2O emission from each soil; the contribution would increase with increasing distances from the *Pinus koraiensis* trunks rather than from the *Quercus mongolica* trunks. Under the experimental conditions, the use of C_2H_2 at low concentrations showed no significant influence on soil microbial biomass N, net N mineralization and microbial respiration. However, 100 Pa C_2H_2 in headspace gas could reduce carbon dioxide (CO_2) emissions from soils. According to the rapid consumption of 10 Pa C_2H_2 by forest soils and convenience for laboratory incubations, 50 Pa C_2H_2 in headspace gas can be used to study the origin of N_2O emissions from forest soils under aerobic conditions and the key associated driving mechanisms. The N_2O and CO_2 emissions from the soils at the same distances from the *Quercus mongolica* trunks were larger than those from the *Pinus koraiensis* trunks, and both emissions decreased as the distances from trunks increased. The stepwise regression analysis showed that 95% of the variability in soil CO_2 emissions could be accounted for by the concentrations of soil total C and water soluble organic C and soil pH, and that 72% of the variability in soil N_2O emissions could be accounted for by the concentrations of soil total N, exchangeable NH_4^+-N and microbial biomass N and 25% of the variability in heterotrophic nitrification by the soil microbial biomass N concentration. The emissions of N_2O and CO_2 from forest soils after exposure of C_2H_2 at low concentrations were positively related to the net nitrification of the soils.

heterotrophic nitrification, forest soil, acetylene, mineralization, microbial biomass nitrogen

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Low doses of acetylene (C_2H_2) (normally within a range of 10 to 100 Pa) inhibited autotrophic NH_4^+ oxidation, and then NH_4^+ concentration in soil was increased after exposure of C_2H_2[1–3]. Due to microbial preferential utilization for NH_4^+, this phenomenon may result in an increase in microbial N immobilization in soil[4], which was accompanied by the decrease in soil mineral N concentration. As a result, the change in concentrations of soil N forms due to addition of C_2H_2 is likely to affect the responses of nitrous oxide (N_2O) emissions from soils to addition of C_2H_2 at low concentrations. Hatch et al.[5] showed that the use of acetylene at larger than 10 Pa C_2H_2 in headspace gas tended to increase soil microbial N immobilization. In addition, C_2H_2 can act as a carbon source to heterotrophic soil microorganisms[6], thereby increasing soil microbial N concentration at partial pressures larger than 100 Pa C_2H_2[7]. To date, there is limited knowledge about the effects of C_2H_2 exposure on soil microbial biomass N concentration. This lack would not be beneficial to further studying soil N transformations and the key associated driving mechanisms by using inhibition of C_2H_2.

Earlier few studies have shown that there are reduced rates of mineralization in grassland soils at concentrations of added C_2H_2 from 500 to 10000 Pa C_2H_2, and that 10% C_2H_2 in headspace gas can result in an apparent net immobilization of mineral N[8]. However, we cannot confirm whether the exposure of C_2H_2 at low concentrations affects the change in soil mineral N production. Due to the limitations of measurement, at present there is limited knowledge with regard to heterotrophic nitrification and its relationships to N transformations and N_2O production in acid forest soils[9]. Differences in stem flow of different trees normally result in the change in properties of the soils around trunks, which is likely to affect the origin of N_2O emission from soil and its spatial properties under forest stands. Hence, it is useful and timely to understand the effects of C_2H_2 at low concentrations on microbial N immobilization and net mineralization in soil.

The objectives of this present work were to study the effects of C_2H_2 at low concentrations on nitrification, mineralization and microbial N immobilization in temperate forest surface soils at the varying distances (0, 50, and 100 cm) from coniferous and hardwood trunks. Of addition interest are the contributions of heterotrophic nitrification and the varying distances from trunks to N_2O production in soil. The results would promote one to study the origin of N_2O emissions from acid soils under upland forest ecosystems and the key associated driving mechanisms.

1 Materials and methods

1.1 Soil sampling and measurement of soil properties

Temperate forest surface soils under the Korean pine and broadleaf mixed forest (*Pinus koraiensis* mainly mixed with hardwood trees such as *Quercus mongolica*, >200 years, altitude 740 m) were sampled near the Research Station of Changbai Mountain Forestry Ecology (128°28′E, 42°24′N). Annual mean temperature is *c*. 2.8℃ and precipitation *c*. 750 mm at the bottom of the mountain. Five individual coniferous and broadleaf trees (e.g., *Pinus koraiensis* and *Quercus mongolica*) with a broad diameter of *c*. 80 cm each and with no trees within the range of 10 to 15 m from trunks were respectively selected in July 2006, and at the varying distances (0, 50, and 100 cm) from trunks, forest surface soils to a depth of 7 cm, after removal of litter and humus layers, were sampled by hammering the cores into the soil, and bulked as one sample. Hence, in total 30 bulk soil samples were taken. All samples were kept separately in airtight plastic bags and rapidly transported to the laboratory. The fresh moist soils were sieved (2 mm) to remove small stones and roots, and stored in the dark at 4℃ for use.

The forest soil belongs to Andisols (Food and Agriculture Organization Soil Classification), and the depth of the A-horizon in soil profile is approximately 10 cm. The properties of the soils at the various depths beneath the forest stand were reported by Xu et al.[10]. Duplicate soils were dried at 105℃ for 24 h to determine moisture content. Total C and N concentrations in soil samples were measured with a CN analyzer (MT-700 with an Auto Sampler MTA-600, Yanaco, Kyoto, Japan). Fresh soil pH (soil/water, 1/2.5, w/w) was measured with a portable pH meter. Exchangeable NH_4^+-N and NO_3^--N concentrations were extracted by shaking 5.0-g fresh soil with 25 mL of 1 mol/L KCl solution for 60 min on an end-over-end shaker. The slurry was then filtered into 50-mL plastic bottles, and NH_4^+-N and NO_3^--N concentrations in the extracts were measured colorimetrically via the nitroprusside and hydrazine-reduction methods, respectively[11]. Forest soils (5.0 g) were extracted by shaking with 25 mL of deionized water for 30 min on an

end-over-end shaker. The suspensions were centrifuged at 6400 g for 5 min and then filtered into 50-mL plastic bottles via cellulose-acetate membrane filters (0.45 μm pore size). Soluble organic C and total N concentrations in the soil extracts were measured using a TOC/TN-analyzer (Shimadzu TOC-V_{CSH}/TN, Kyoto, Japan). Concentrations of soluble soil organic N (DON) were considered the differences between total N and mineral N (NH_4^+-N and NO_3^--N) concentrations in the soil extracts. Soil microbial C and N concentrations were measured using the chloroform fumigation-extraction method[12]. Fresh soils (5.0 g) with and without chloroform fumigation were extracted with 25 mL of 0.5 mol/L K_2SO_4 solution, as above, and soluble organic C and total N in the extracts were measured using a TOC/TN-analyzer (Shimadzu TOC-V_{CSH}/TN, Kyoto, Japan). The main soil properties at the varying distances from main trunks are presented in Table 1.

1.2 Incubation experiment

All forest soil samples taken from the varying distances from trunks were used to study the change in soil microbial biomass N and mineral N concentrations and in N_2O and CO_2 emissions from the soils after exposure of C_2H_2 at low concentrations. For each of soil samples, water was added to naturally settled soils (20.0 g) in 250-mL glass bottle to achieve a range of WFPS from 0.40—0.45 m^3 water m^{-3} pore space. The level of the soil in each bottle was marked prior to incubation and the volume of the soil was measured via water following the removal of soil. The WFPS was calculated according to the equation reported by Franzluebbers[13]. At the start of the incubation, a series of acetone-free C_2H_2 (approximately 3% C_2H_2 in N_2, *v/v*) volume was injected into each bottle through a stopcock connected to the needle inserted through the stoppers using air-tight syringes to give concentrations of 10, 50 and 100 Pa C_2H_2; the control with C_2H_2 addition was incubated. The batch incubation was done at 25℃ in the dark for 10 days under oxic conditions. Twenty-five milliliter of headspace gases was sampled from each bottle using 50-mL air-tight syringes 3, 5, 7 and 10 days after C_2H_2 addition and then analyzed for concentrations of CO_2, N_2O and C_2H_2 using a gas chromatograph. After sampling, the rubber stoppers were removed from the bottles for 2 h to refresh the air, and then they were closed again. A series of acetone-free C_2H_2 volume was again injected into each bottle, as above, to receive concentrations of 10, 50 and 100 Pa C_2H_2. Preliminary studies have indicated that 10 Pa C_2H_2 in headspace gas is completely consumed by soil microorganisms within 24 h. Thus, the presence of 10 Pa C_2H_2 in headspace gas can remain through daily injection of standard C_2H_2 into each bottle. At the end of the 10-day oxic incubation, 5-g and 10-g subsamples from each treatment were respectively used to measure the concentrations of soil mineral N (NH_4^+-N and NO_3^--N) and microbial biomass C and N, as above.

1.3 Measurement of CO_2, N_2O and C_2H_2 concentrations

According to the methods reported by Wang et al.[14], concentrations of CO_2, N_2O and C_2H_2 in headspace gases were respectively quantified with a modified gas chromatograph (Agilent 5890) equipped with a flame ionization detector (FID) and with an electron capture detector (ECD). Carbon dioxide, N_2O and C_2H_2 were severally separated by one stainless steel column (2 m length and 2.2 mm i.d.) that was packed with 50—80 mesh Porapack Q, afterwards hydrogen reduced CO_2 to CH_4 in a Nickel catalytic converter at 375℃, and the

Table 1 Main properties of the soils at the varying distances from trunks[a)]

Distances from trunks (cm)	TC	TN	C/N	DOC	DON	Microbial C	Microbial N	NH_4^+-N	NO_3^--N	pH
	(mg g^{-1} dry soil)			(μg g^{-1} dry soil)						(water)
Broadleaf forest soil										
0	129.7(7.5)	11.5(1.6)	11.7(0.8)	621(118)	119(10)	2338(171)	573(27)	28.0(4.6)	2.7(0.8)	6.3(0.1)
50	78.8(3.6)	6.8(0.4)	11.7(0.3)	299(44)	69(6)	1939(187)	441(42)	16.3(3.1)	1.1(0.1)	5.6(0.1)
100	72.8(10.1)	6.3(0.8)	11.4(0.3)	252(30)	59(6)	1920(266)	429(62)	11.4(2.9)	0.9(0.1)	5.4(0.1)
Coniferous forest soil										
0	71.6(17.8)	5.4(0.9)	12.8(0.8)	512(201)	71(15)	1620(261)	369(73)	17.3(5.3)	2.4(0.4)	5.4(0.1)
50	50.6(6.2)	4.1(0.5)	12.4(0.5)	245(38)	46(4)	1239(167)	281(36)	7.9(1.5)	0.9(0.1)	5.5(0.1)
100	46.7(5.1)	3.9(0.4)	12.1(0.4)	192(11)	45(4)	1108(194)	245(45)	7.1(1.6)	0.8(0.1)	5.5(0.1)
$LSD_{0.05}$	28.1	2.7	1.6	289	25	619	145	10.1	1.1	0.2

a) Values are the means of five replicates, and standard errors are shown in parenthesis. Least significant differences (LSD; 5%) were used to assess differences in the soil properties at the varying distances from trunks. Broadleaf tree, *Pinus koraiensis*; Coniferous tree, *Quercus mongolica*. The same below.

CH_4 and C_2H_2 were severally detected by FID. The oven was operated at 55℃, and the FID at 200℃ (ECD at 330℃ for N_2O), respectively, with N_2 as carrier gas at a flow rate of 20 cm^3 min^{-1}. The detector responses were calibrated using certified gas standards, which contain 4.0 mL L^{-1} CO_2 in N_2, 0.37 μL L^{-1} N_2O in N_2, and 100 μL L^{-1} C_2H_2 in N_2, respectively.

1.4 Calculation and statistical analysis

All data were transformed on an oven-dried weight basis. Rates of N_2O and CO_2 emissions from soils were calculated from the increase in gas concentration with time, and were expressed in pmol N_2O-N g^{-1} dry soil d^{-1} and μmol CO_2-C g^{-1} dry soil d^{-1}, respectively. The CO_2 concentrations in headspace gases excluded CO_2 production due to the decomposition of acetylene by soil. Soil net N mineralization rates were calculated by the difference in soil NH_4^+-N and NO_3^--N concentrations after and prior to the incubation divided by time, and were expressed in μg N g^{-1} dry soil d^{-1}. Concentrations of soil microbial biomass C and N were calculated by using the difference in salt-extractable organic C or N concentrations between fumigated and non-fumigated soil extracts divided by 0.45[15,16]. Means and standard errors of five replicates in each treatment were calculated. Least significant differences (LSD) were calculated at the 5% level to assess the differences in N_2O and CO_2 emissions and in soil properties between treatments and soils. Stepwise regression analysis was performed by using SPSS software for Windows (version 13.0) to assess soil properties that can affect the emissions of N_2O and CO_2 from soils and heterotrophic nitrification in forest soils.

2 Results and discussion

2.1 N_2O emissions from soils after exposure of C_2H_2 at low concentrations and heterotrophic nitrification

Table 2 shows the effects of addition of C_2H_2 at low concentrations on N_2O emissions from the soils at the varying distances from trunks, and the contribution of heterotrophic nitrification to the soil N_2O emission. The N_2O emission from the soil at the same distance from the hardwood trunks was larger than that from the coniferous trunks, and it decreased as the distances from trunks increased. The difference in soil N_2O emissions was mainly ascribed to the status of nutrients and microbial biomass C and N concentrations in soil (Table 1). The results were in agreement with the findings reported by Ullah et al.[17], who showed that deciduous forest soils exhibited greater potential of N_2O fluxes than did white pine forest soils in Eastern Canada.

In comparison with the control, the use of acetylene at partial pressures from 10 to 100 Pa C_2H_2 in headspace gases gave a significant decrease in N_2O emission at soil moisture of *c*. 45% water-filled porosity space, and the decrease was the same in each soil after treatment with acetylene at low concentrations (Table 2). Heterotrophic nitrification could account for 21%—48% of total N_2O emission from each soil; the contribution would increase with increasing distances from the coniferous trunks

Table 2 N_2O emissions from the soils after exposure of C_2H_2 at low concentrations and proportion of heterotrophic nitrification in the soils at the varying distances from trunks

Distances from trunks (cm)	N_2O emissions from the soils (pmol N_2O-N g^{-1} dry soil d^{-1}) and proportion of heterotrophic nitrification after exposure of C_2H_2 at low concentrations							$LSD_{0.05}$ [a]	$LSD_{0.05}$ [b]
	0 Pa C_2H_2	10 Pa C_2H_2		50 Pa C_2H_2		100 Pa C_2H_2			
	N_2O emission	N_2O emission	Proportion of heterotrophic nitrification	N_2O emission	Proportion of heterotrophic nitrification	N_2O emission	Proportion of heterotrophic nitrification		
Broadleaf forest soil									
0	133.5(23.4)	31.7(4.0)	0.26(0.04)	25.8(1.7)	0.21(0.02)	31.2(3.1)	0.27(0.06)	36.0	0.14
50	103.2(16.9)	29.1(4.7)	0.30(0.04)	26.8(6.4)	0.26(0.05)	21.0(3.5)	0.21(0.03)	28.4	0.12
100	104.8(18.5)	25.1(3.4)	0.30(0.10)	22.1(2.7)	0.30(0.13)	22.9(2.4)	0.30(0.12)	28.7	0.36
Coniferous forest soil									
0	109.9(5.6)	29.9(6.8)	0.27(0.05)	26.5(5.7)	0.24(0.05)	28.4(3.7)	0.26(0.05)	16.7	0.15
50	73.5(12.1)	22.5(4.1)	0.32(0.05)	15.9(1.2)	0.25(0.05)	18.8(3.8)	0.27(0.05)	20.0	0.15
100	66.6(12.4)	33.6(10.0)	0.48(0.08)	25.1(6.9)	0.37(0.06)	22.3(5.6)	0.34(0.04)	27.1	0.19
$LSD_{0.05}$ [c]	46.2	17.4	0.19	13.7	0.20	10.6	0.19		

a) LSD (5%) for comparing differences in N_2O emissions from each soil after exposure of C_2H_2 at low concentrations; b) LSD (5%) for comparing differences in proportions of heterotrophic nitrification to N_2O emission from each soil between treatments of C_2H_2 at low concentrations; c) LSD (5%) for comparing differences in N_2O emissions and proportions of heterotrophic nitrification between soils after exposure of C_2H_2 at the same concentration.

rather than from the hardwood trunks (Table 2). Xu and Inubushi[7] reported that heterotrophic nitrification among acid forest soils in central Japan accounted for the range from 37% to 76% of the total N_2O production under the controlled aerobic conditions and was variable with forest stands, topographical slope and seasons. Hart et al.[18] also reported that heterotrophic nitrification accounted for 65%—72% of the gross nitrification in two coniferous forest stands of contrasting productivity and with different altitude, and that soil pH (3.9—5.4) and status of substances were important in controlling the significance of heterotrophic nitrification in forest soil. However, using ^{15}N pool dilution-enrichment approach in the laboratory, Barraclough and Puri[19] observed that heterotrophic nitrification contributed $\leqslant$8% of the total gross nitrification rate in an acid woodland soil. There was usually low activity of autotrophic nitrifiers and the predominance of heterotrophic nitrification in acid forest soils at pH < 4.5, although there is information about autotrophic nitrifying bacteria that are capable of ammonium oxidation at pH 4[20]. Pedersan et al.[21] showed that the heterotrophic nitrification accounted for a larger contribution to total gross nitrification rate in coniferous forest soils after clear-cutting compared to that in undisturbed forest soils. Hence, the contribution of heterotrophic nitrification to soil total gross nitrification was dependent on kinds of forest vegetations, soil properties, and seasons.

Acetylene at low concentrations can be considered a particularly useful selective biochemical block of the autotrophic pathway[1,2], which does not inhibit the heterotrophic pathway[3,22]. In order to effectively study heterotrophic nitrification and its contribution to N_2O production from acid forest soils, it is timely and important to understand the effects of acetylene at low concentrations on microbial N immobilization and net mineralization in soil.

2.2 Change in soil net mineralization and microbial biomass N concentrations after exposure of C_2H_2 at low concentrations

The net N mineralization rate in the soil at the same distance from the hardwood trunks was larger than that from the coniferous trunks, and it was larger at 0 cm than at 50 cm and 100 cm distances from each trunk (Table 3). The differences in soil net N mineralization rates can be also ascribed to the status of nutrients and microbial biomass N and C concentrations in soil (Table 1). In comparison with the control, the use of acetylene at partial pressures from 10 to 100 Pa C_2H_2 in headspace gases showed no significant influence on the net N mineralization in the soils at the varying distances from trunks. This was in agreement with the results reported by Ross et al.[23], who showed that the use of 100 Pa C_2H_2 had no significant influence on net N mineralization in forest soils in Northeastern USA. However, Boer et al.[24] reported that the addition of acetylene at partial pressures from 10 to 1000 Pa C_2H_2 in headspace gas decreased net mineral N production in mineral layer rather than in organic layer of an acid oak-beech forest soil. Similarly, Hatch et al.[8] reported that the use of C_2H_2 at elevated concentrations from 1000 to 10000 Pa C_2H_2 significantly decreased net mineral N production

Table 3 Net N mineralization and microbial biomass N concentrations after exposure of C_2H_2 at low concentrations in the soils at the varying distances from trunks

Distances from trunks (cm)	Soil net N mineralization (μg N g^{-1} dry soil d^{-1}) and microbial biomass N concentrations (μg N g^{-1} dry soil) after exposure of C_2H_2 at low concentrations								$LSD_{0.05}$ [a]	$LSD_{0.05}$ [b]
	0 Pa C_2H_2		10 Pa C_2H_2		50 Pa C_2H_2		100 Pa C_2H_2			
	Net N mineralization	Microbial N	Net N mineralization	Microbial N	Net N mineralization	Microbial N	Net N mineralization	Microbial N		
Broadleaf forest soil										
0	7.83(0.90)	573.1(26.8)	7.92(1.14)	597.2(29.4)	7.75(0.94)	594.1(37.1)	7.65(0.79)	613.3(39.0)	2.85	100.4
50	2.77(0.22)	441.2(41.7)	2.69(0.22)	469.5(47.8)	2.58(0.32)	458.0(42.2)	2.93(0.29)	465.3(43.2)	0.81	131.3
100	3.51(0.47)	429.0(61.7)	3.32(0.42)	446.2(63.6)	3.50(0.24)	456.8(69.9)	3.32(0.33)	448.5(66.5)	1.3	196.4
Coniferous forest soil										
0	2.63(0.66)	369.3(72.5)	2.44(0.61)	363.0(67.4)	2.69(0.66)	360.3(67.5)	2.72(0.69)	367.5(67.0)	1.54	205.8
50	2.10(0.17)	281.9(36.2)	1.90(0.34)	284.5(40.6)	1.80(0.26)	279.4(46.7)	1.80(0.27)	284.7(39.5)	0.79	122.7
100	1.68(0.48)	245.5(44.7)	1.69(0.30)	253.6(40.8)	1.65(0.34)	251.6(42.9)	1.74(0.30)	251.4(43.4)	1.08	128.8
$LSD_{0.05}$ [c]	1.51	145.2	1.67	146.1	1.46	153.7	1.34	149.6		

a) LSD (5%) for comparing differences in net N mineralization in each soil after exposure of C_2H_2 at low concentrations; b) LSD (5%) for comparing differences in microbial biomass N concentrations in each soil after exposure of C_2H_2 at low concentrations; c) LSD (5%) for comparing differences in net N mineralization and microbial biomass N concentrations between soils after exposure of C_2H_2 at the same concentration.

in soil and the inhibition was associated with initial soil NO_3^- concentration. Hence, the effects of C_2H_2 addition on soil net N mineralization were dependent on the concentrations and decomposition of C_2H_2 and soil properties such as NO_3^--N concentration.

Soil microbial biomass N concentration at the same distance from the hardwood trunks was much larger than that from the coniferous trunks ($P < 0.05$), and it decreased as the distances from trunks increased. The difference in soil microbial biomass N concentrations could be accounted for by the relatively large concentrations of organic C and water soluble organic C in the soils under hardwood trees rather than under coniferous trees (Table 1). In comparison with the control, the use of acetylene at partial pressures from 10 to 100 Pa C_2H_2 in headspace gases showed no significant influence on microbial biomass N concentration in soil (Table 3). However, Hatch et al.[5] showed that the addition of acetylene at partial pressures larger than 10 Pa C_2H_2 in headspace gas tended to increase microbial N immobilization in fertilized and unfertilized grass soils, but did not influence microbial N concentration in unfertilized grass and clover soil. The oxidizing capacity of the nitrifiers was recognized as one of important factors controlling the increase in soil microbial N immobilization[5]. Xu and Inubushi [7] reported that the addition of C_2H_2 at partial pressures within a range from 500 to 10000 Pa C_2H_2 in headspace gases significantly increased microbial N immobilization in soil. Furthermore, the stimulating effect of C_2H_2 on the increase in soil microbial N immobilization would remain a saturation status after exposure of leveled C_2H_2 concentrations[7]. Hence, the effects of C_2H_2 on soil microbial N immobilization were dependent on the concentrations of C_2H_2, kinds of soils, the oxidizing capacity of the nitrifiers, and the other experimental conditions.

2.3 Change in soil respiration after exposure of C_2H_2 at low concentrations

Soil respiration has been considered one of main methods describing apparent microbial activity. According to the measurement of CO_2 emissions from soils, the present experiment can be used to assess the effects of C_2H_2 at low concentrations on microbial respiration in forest soils. The results and combination with the responses of soil microbial N immobilization and net mineralization to C_2H_2 exposure would promote one to study the origin of N_2O emissions from acid forest soils and the key associated driving mechanisms.

The addition of acetylene at partial pressures from 10 to 100 Pa C_2H_2 in headspace gases showed no significant influence on microbial respiration in forest soils, but the use of 100 Pa C_2H_2 tended to decrease soil respiration (Table 4). The CO_2 emission from the soil at the same distance from the hardwood trunks was larger than that from the coniferous trunks, and it decreased as the distances from trunks increased (Table 4). This trend paralleled with the change in N_2O emissions, net mineralization and microbial biomass N concentrations of the soils with and without addition of acetylene (Tables 2 and 3). Herrmann et al.[25] showed that brief exposure to low atmospheric C_2H_2 concentrations in headspace gas (0.1—1.0 kPa) did not significantly affect C mineralization rates when nitrification was completely inhibited. However, in a wheat soil from Hebei Province, Li et al.[26] reported that the use of 2000 Pa C_2H_2 in headspace gas reduced the respiration of soil heterotrophic microorganisms by 50%. Hence, the use of acetylene at low concentrations does not affect the activity of soil microbial respiration, but the activity can be inhibited by leveled C_2H_2 concentrations. The functioning of the use of acetylene in inhibiting soil carbon mineralization is variable with soil properties and experimental conditions.

2.4 Soil properties affecting N_2O and CO_2 emissions from the soils at the varying distances from trunks

Table 5 shows the coefficients of correlation between N_2O and CO_2 emissions from the soils in the absence of acetylene and soil properties. The N_2O and CO_2 emissions from the soils without C_2H_2 addition were significantly related to the concentrations of soil total C and N, water soluble organic N, and microbial biomass C and N, respectively. Heterotrophic N_2O emissions from the soils were significantly related to the concentrations of water soluble organic C and N and microbial biomass C and N, respectively. Stepwise regression analysis showed that the emissions of CO_2 and N_2O from soils and heterotrophic N_2O emissions could be accounted for by the following equations:

$$y = -(3.6\pm1.2)+(0.023\pm0.003)a+(0.003\pm0.000)b +(0.68\pm0.22)c,$$
$$R^2 = 0.95,\ n = 30,\ P<0.001, \quad (1)$$

where y is the soil CO_2 emission (μmol CO_2-C g^{-1} d^{-1}) and a, b and c represent the concentration of soil total C (mg C g^{-1}) and water soluble organic C (μg C g^{-1}) and

Table 4 Effects of C_2H_2 at low concentrations on CO_2 emissions from the soils at the varying distances from trunks

Distances from trunks (cm)	CO_2 emissions from the soils (μmol CO_2-C g^{-1} dry soil d^{-1}) after exposure of C_2H_2 at low concentrations				$LSD_{0.05}$ [a)]
	0 Pa C_2H_2	10 Pa C_2H_2	50 Pa C_2H_2	100 Pa C_2H_2	
Broadleaf forest soil					
0	5.05(0.35)	4.92(0.33)	5.13(0.26)	4.89(0.26)	0.91
50	2.70(0.18)	2.67(0.23)	2.64(0.22)	2.47(0.17)	0.60
100	2.36(0.26)	2.35(0.24)	2.28(0.20)	2.14(0.25)	0.72
Coniferous forest soil					
0	3.01(0.99)	2.95(0.95)	2.92(0.95)	2.24(0.69)	2.70
50	2.01(0.25)	2.00(0.27)	1.88(0.26)	1.85(0.30)	0.80
100	1.68(0.11)	1.72(0.18)	1.63(0.19)	1.50(0.17)	0.49
$LSD_{0.05}$ [b)]	1.35	1.31	1.28	1.03	

a) LSD (5%) for comparing differences in CO_2 emissions from each soil after exposure of C_2H_2 at low concentrations; b) LSD (5%) for comparing differences in CO_2 emissions from different soils after exposure of C_2H_2 at the same concentration.

Table 5 Coefficients of correlation between emissions of CO_2 and N_2O from the soils without C_2H_2 addition and initial soil properties at the varying distances from trunks[a)]

	N_2O	CO_2	Total C	Total N	C:N ratio	DOC	DON	Microbial C	Microbial N	NH_4^+-N	NO_3^--N	pH	Hetero N_2O
N_2O	1												
CO_2	0.51**	1											
Total C	0.67**	0.92**	1										
Total N	0.78**	0.77**	0.93**	1									
C:N ratio	−0.46**	0.22	−0.01	−0.33*	1								
DOC	0.21	0.85**	0.67**	0.47**	0.45**	1							
DON	0.70**	0.83**	0.84**	0.79**	−0.09	0.62**	1						
Microbial C	0.73**	0.67**	0.84**	0.82**	−0.22	0.40*	0.68**	1					
Microbial N	0.73**	0.72**	0.85**	0.82**	−0.20	0.44**	0.79**	0.97**	1				
NH_4^+-N	0.25	0.73**	0.75**	0.61**	0.24	0.59**	0.59**	0.61**	0.62**	1			
NO_3^-N	0.34*	0.53**	0.38*	0.30	0.11	0.56**	0.50**	0.21	0.29	0.20	1		
pH	0.32*	0.60**	0.56**	0.54**	−0.10	0.32*	0.72**	0.37*	0.50**	0.45**	0.42**	1	
HeteroN_2O	0.37*	0.41*	0.44**	0.36*	0.06	0.32*	0.42*	0.49**	0.52**	0.40*	0.12	0.15	1

a) Hetero N_2O, Heterotrophic N_2O emission; * P<0.05, ** P<0.01.

soil pH, respectively.

$$y = (24.1\pm10.8)+(8.61\pm2.17)a-(1.72\pm0.50)b +(0.12\pm0.05)c,$$
$$R^2 = 0.72,\ n = 30,\ P<0.001, \quad (2)$$

where y is the soil N_2O emission (pmol N_2O-N g^{-1} d^{-1}) and a, b and c represent the concentration of soil total N (mg N g^{-1}) and exchangeable NH_4^+-N (μg N g^{-1}) and microbial biomass N (μg N g^{-1}), respectively, in the soil.

$$y = (9.7\pm4.6)+(0.036\pm0.011)a,$$
$$R^2 = 0.25,\ n = 30,\ P<0.01, \quad (3)$$

where y is the soil heterotrophic N_2O emission (pmol N_2O-N g^{-1} d^{-1}) and a is the microbial biomass N concentration (μg N g^{-1}), respectively, in the soil.

According to the regression models, 95% of the variability in soil CO_2 emissions could be accounted for by the concentrations of soil total C and water soluble organic C and soil pH, and 72% of the variability in soil total N_2O emissions by the concentrations of soil total N, exchangeable NH_4^+-N and microbial biomass N and 25% of the variability in heterotrophic nitrification by the soil microbial biomass N concentration.

There was the nice goodness-of-fit for the regression models of N_2O and CO_2 emissions from the C_2H_2-treated soils (y) against nitrification rate (x), which was described as $y = 118.1x-11.9$ ($R^2 = 0.46$, $n = 12$, $P = 0.01$) and $y = 3.64x+1.45$ ($R^2 = 0.45$, $n = 12$, $P = 0.01$), respectively. The regression models indicated that both N_2O and CO_2 emissions from the soils after exposure of C_2H_2 at low concentrations were positively related to the net nitrification of the soils.

3 Conclusions

The use of C_2H_2 at partial pressures within a range from 10 to 100 Pa C_2H_2 in headspace gas gave a significant decrease in N_2O emissions from forest soils, and the decrease was the same in each soil after exposure of C_2H_2. Heterotrophic nitrification could account for 21%—48% of the soil total N_2O emission; the contribution increased with increasing distances from the coniferous trunks rather than from the hardwood trunks. The

emissions of N_2O and CO_2 from soils decreased as the distances from trunks increased.

The use of C_2H_2 at low pressures from 10 to 100 Pa C_2H_2 in headspace gas did not influence soil microbial biomass N, net N mineralization and microbial respiration, but 100 Pa C_2H_2 in the headspace could decrease CO_2 emissions from soils. According to the rapid consumption of 10 Pa C_2H_2 by forest soils and the convenience for laboratory incubations, 50 Pa C_2H_2 in headspace gas can be used to study the origin of N_2O emissions from acid forest soils under aerobic conditions and the key associated driving mechanisms.

N_2O and CO_2 emissions from forest soils at the varying distances from trunks were associated mainly with the concentrations of soil total C and N, water soluble organic C and N as well as microbial C and N, and both emissions from the C_2H_2-treated soils were positively related to the net nitrification of the soils. Soil microbial biomass N concentration accounted for 25% of the variability in heterotrophic nitrification.

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