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Soil resource availability impacts microbial response to organic carbon and inorganic nitrogen inputs

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Abstract: Impacts of newly added organic carbon (C) and inorganic nitrogen (N) on the microbial utilization of soil organic matter are important in determining the future C balance of terrestrial ecosystems. We examined microbial responses to cellulose and ammonium nitrate additions in three soils with very different C and N availability. These soils included an organic soil (14.2% total organic C, with extremely high extractable N and low labile C), a forest soil (4.7% total organic C, with high labile C and extremely low extractable N), and a grassland soil (1.6% total organic C, with low extractable N and labile C). While cellulose addition alone significantly enhanced microbial respiration and biomass C and N in the organic and grassland soils, it accelerated only the microbial respiration in the highly-N limited forest soil. These results indicated that when N was not limited, C addition enhanced soil respiration by stimulating both microbial growth and their metabolic activity. New C inputs lead to elevated C release in all three soils, and the magnitude of the enhancement was higher in the organic and grassland soils than the forest soil. The addition of cellulose plus N to the forest and grassland soils initially increased the microbial biomass and respiration rates, but decreased the rates as time progressed. Compared to cellulose addition alone, cellulose plus N additions increased the total C-released in the grassland soil, but not in the forest soil. The enhancement of total C-released induced by C and N addition was less than 50% of the added-C in the forest soil after 96 d of incubation, in contrast to 87.5% and 89.0% in the organic and grassland soils. These results indicate that indigenous soil C and N availability substantially impacts the allocation of organic C for microbial biomass growth and/or respiration, potentially regulating the turnover rates of the new organic C inputs.

Keywords: cellulose; inorganic nitrogen; microbial biomass; microbial activity; carbon sequestration

Introduction

Soil contains nearly twice as much carbon (C) as the atmosphere (about 735 Gt C) and makes up about 65% of the terrestrial ecosystem C (about 2060 Gt C) (Schlesinger, 1996). Whether soil will act as a C sink or a source under future land management regimes and climatic conditions will profoundly impact the CO_2 concentration in the atmosphere (Zhang, 2004a). Recently, various management practices have been recommended to facilitate soil C sequestration by increasing C inputs or reducing decomposition in managed ecosystems (Zhang, 2004b; Kucharik, 2001). These practices include no-tillage (Balesdent, 2000), residue or manure application (Witt, 2000), surface mulching (Jacinthe, 2002), waste application (Borken, 2002), and nitrogen (N) fertilization (Curtin, 2002). In natural ecosystems, predicted CO_2 elevation (Strain, 1987) and N deposition (Holland, 1999) will likely enhance C inputs to soils by stimulating plant primary production. However, whether and how these C and N inputs will affect long-term C storage in soils is not well clear, because they may either stimulate (Woods, 1987) or inhibit (Ågren, 2001) soil indigenous organic matter (SOM) decomposition. Microbes dominate SOM decomposition and nutrient mineralization (Paul, 1996; Zhang, 2004b). Therefore, the impacts on the long-term C storage in soils may depend on microbial responses to the newly added C and N.

Carbon additions to soil usually enhance microbial activities because soil microbes are often energy limited (Paul, 1996). Elevated microbial activities can increase C release by accelerating SOM decomposition, but the net effects on soil C balance varies. Campbell *et al*. (Campbell, 1991) found that C addition in the form of legume green manure had few effects on soil organic C storage in an agricultural soil with a high indigenous organic C level, suggesting that most of the added C was released due to the enhanced microbial activities. McCarl and Schneider (McCarl, 2001) stated that more C from added plant materials could be sequestrated in soils with a low then high indigenous C availability. In addition, increased microbial activity may be due to two different processes, one accompanied with an increase in microbial biomass and one without net increase. For example, Ambus and Jensen (Ambus, 1997) found that C addition stimulated microbial activities with an increase in biomass in agricultural soils after incorporating barley and pea crop residues. Ekblad and Nordgren (Ekblad, 2002) also found that microbial respiration increased with an increase in microbial biomass in boreal forest soils after sucrose addition. However, Vance and Chapin (Vance, 2001) found that newly added-C stimulated microbial respiration without increase in microbial biomass in taiga forest floors.

The effects of mineral N inputs on microbes may be dependent on the specific environment and the process examined. Mineral N can directly enhance microbial activities, since microbes can be N-limited in some environments (Kaye, 1997). For example, Ekblad and Nordgren (Ekblad, 2002) found that NH_4Cl addition stimulated microbial respiration in boreal forest soils. In addition, it has been well demonstrated that N addition increased the decomposition of crop residue with a low-N content, such as wheat straw (Mary, 1996). However, Entry (Entry, 2000) found that the addition of 200 or 400 kgN/hm^2 as NH_4NO_3 decreased cellulose and lignin degradation in wetland soils after 10 and 15 weeks of incubation. In another incubation experiment, Bardgett *et al*. (Bardgett, 1999) found that N addition had no consistent effect on microbial biomass or activities in a semi-natural grassland soil. Furthermore, the effects of N addition may depend on the decay substrates. Carreiro *et al*. (Carreiro, 2000) showed that NH_4NO_3 addition increased cellulase activity in decaying leaf litter but reduced the activity of lignin-degrading

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phenoloxidase. Neff *et al*. (Neff, 2002) also found that the effects of N additions differentially affect soil C pools. They demonstrated that long-term N addition significantly accelerated decomposition of light soil carbon fractions while further stabilizing soil carbon compounds in heavier, mineral-associated fractions. The chemical reaction of added mineral N with soil organic C may contribute to observed inconsistent N effects, because there is high variation in the chemical structure of C substrates in different soils (Berg, 1997). For example, N-added can combine with phenolic and other dissolved organic C to form recalcitrant organic compounds in soils (Hodge, 1953; Fog, 1988; Davidson, 2003).

Soil C sequestration through effective residue, manure or waste management has drawn recent attention (Witt, 2000; Borken, 2002; Eghball, 2002; Six, 2002), but the efficiency of these practices is not exactly known. One major unknown is how soil physical and chemical characteristics impact microbial response to newly added C. Many studies on the effects of organic material amendments have primarily focused on microbial response in a single soil (Mary, 1996; Ambus, 1997; Gagnon, 2001; Borken, 2002; Eghball, 2002), making the evaluation of "soil factors" difficult. It was proposed that the potential of long-term soil C sequestration positively correlated with the loss of organic C during the previous land use (McCarl, 2001). This concept implies that highly degraded soils with low organic matter (e.g. intensively farming lands) may have greater sequestration potential than fairly undisturbed soils with high organic matter (e.g. native forest soils). However, Six *et al*. (Six, 2002) suggested that native soil C levels did not necessarily represent an upper limit in soil C stocks. They further proposed that physicochemical characteristics inherent to soils defined the maximum protective capacity of SOM pools. In addition, animal manure and wastes often contain high levels of available N (Borken, 2002; Eghball, 2002), likely leading to interactive effects of C and N on soil microbes and subsequent C dynamics. Since N stimulation of SOM decomposition in agricultural ecosystems has been well documented, it is of significance to understand how N availability impacts the retention of newly added C inputs in soils.

We hypothesized that microbial responses to C and N additions significantly depend on indigenous soil characteristics, particularly the status of C and N. In the present study, we performed an incubation experiment using three soils with different levels of C and N to examine microbial responses to newly added C (as cellulose) and N (as NH_4NO_3). Incubation methodology enables the experimental conditions to be sufficiently controlled and allow us to effectively address the differences in microbiological responses to substrate amendments in soils. Cellulose is the largest component of plant litterfall and the residue that enters the soil (Richmond, 1991). NH_4^+ and NO_3^- are the main active N components of N depositions and mineral N fertilizers. Our aims were to (1) quantify whether new C and N inputs facilitate C release from soils, and (2) examine whether and how differences in indigenous resource availability impact microbial responses to C and N inputs.

1 Methods and materials

1.1 Soil and site description

Soils used in this study were collected from the surface layer (0—15 cm) of three different terrestrial ecosystems in the spring of 2002. The organic soil (muck, with 58% sand, 33% silt, 9% clay and 14.2% organic C) was collected from a farmland in Plymouth, North Carolina, USA. This farmland supported an irrigated cropping system of corn-soybean rotation. About 200 kg soil was obtained from the field. Although it contained the highest organic C and extractable N, it had very low microbial biomass (Table 1) and labile C (3.69 mg/kg soil). The forest soil was collected from a Binghamton University Nature Preserve area, Binghamton, New York, USA. This soil is in the Lordstown series, which is coarse-loamy, mixed, mesic Typic Dystrudepts (surface A layer soil contains 30% sand, 60% silt, 10% clay and 4.7% organic C). Soils were collected from 5 randomly chosen locations in a secondary growth mixed forest, composed and shipped to North Carolina State University. This soil had very low extractable N, but it contained the highest labile C (12.75 mg/kg soil) and microbial biomass (Table 1). The grassland soil (cecil sand clay loam, with 53% sand, 18% silt, 29% clay and 1.6% organic C) was collected from a grassland ecosystem in Raleigh, NC, USA. It contained the lowest labile C (2.52 mg/kg soil), but had an intermediate amount of extractable N compared to the other two (Table 1).

Table 1 Initial characteristics of the three soils

Property	Soil type		
	Organic soil	Forest soil	Grassland soil
pH	4.5 ± 0.01	4.8 ± 0.01	5.3 ± 0.02
Water holding capacity (WHC), %	70.4 ± 4.4	64.1 ± 5.2	32.8 ± 2.1
Total organic C, %	14.2 ± 0.01	4.7 ± 0.01	1.6 ± 0.02
Total N, %	0.74 ± 0.00	0.27 ± 0.00	0.13 ± 0.00
Extractable inorganic N, mg/kg soil	299.2 ± 2.6	5.7 ± 0.3	42.7 ± 0.6
Microbial biomass C, mg/kg soil	675.2 ± 49.4	1476.9 ± 20.1	635.0 ± 21.4
Microbial biomass N, mg/kg soil	81.5 ± 6.6	251.9 ± 3.5	81.9 ± 2.4

Note: Value was mean of three replicates ± 1 *SE*

1.2 Experimental design and treatments

The experiment was a 3 × 3 × 3 factorial design with three soil types, three treatments (control, C addition, and C + N addition) and three incubation stages (14 d, 24 d and 71 d). Each treatment combinations contained four replicates. The N addition was in the form of NH_4NO_3 and the C addition was in the form of powdered cellulose (CF11, Whatman Co., Maidstone, England).

Soils were passed through a sieve (4 mm) and kept in plastic bags at room temperature (22 ± 2℃) for one week. Then the moisture of each soil was adjusted evenly to 50% of the water holding capacity (WHC) by fine spraying with deionized (DI) water, and kept in a covered plastic box in the dark at room temperature (22 ± 2℃) for another one week before being used for the incubation. Three subsamples of each soil type were collected from the plastic box before

treatments were imposed, and were kept in a refrigerator at 4℃ as original soil for the determination of general soil characteristics(Table 1).

The three treatments were (1) an untreated control (CONT), (2) a cellulose treatment(CELL, 10 g cellulose powder(kg soil), and (3) a cellulose plus N treatment (NCELL, 10 g cellulose and 80 mg N of NH_4NO_3/kg of soil). Cellulose powder was mixed into soils that had been conditioned for the CELL and NCELL treatments. NH_4NO_3 solution was added to the NCELL treatments using a fine spray to bring soil moisture to 70% WHC. Deionized water was added to the CONT and CELL to achieve the same soil moisture as in the NCELL. For the measurements of soil microbial biomass and available N, soil (15.0 g dry soil equivalent) was incubated in the dark at room temperature in plastic cups and glass flasks, respectively. The cups and flasks were covered with aluminum foil, and ten holes were made with a needle to allow for O_2 movement. Soil moisture was adjusted weekly by adding DI water. For the determination of soil respiration, 20.0 g dry soil equivalent was incubated in mason jars in the dark at room temperature (22 ± 2℃) and CO_2 production was quantified.

1.3 Measurement methods

Soil samples were sampled on day 14, 24, and 71 for soil microbial biomass C (MBC) and N (MBN), and extractable NH_4-N and NO_3-N. MBC and MBN were determined using the method of fumigation-extraction (48 h fumigation; Vance, 1987). Soil extractable organic C before and after the fumigation was quantified using a Shimadzu total organic C analyzer(Shimadzu TOC-5050A, Shimadzu Co., Kyoto, Japan). Total extractable-dissolvable soil N(TDN) of the non-fumigated and fumigated soils were measured on a Lachat flow injection analyzer(Lachat Quickchem Systems, Milwaukee, WI) after alkaline persulfate digestion(Cabrera, 1993). The differences in organic C and TDN between the fumigated and non-fumigated soils were assumed to be released from lysed soil microbes. The released C and N were converted to MBC and MBN, respectively, using k_{ec}-0.33 (Sparling, 1988) and k_{en}-0.45 (Jenkinson, 1988). Extractable soil inorganic N (NH_4-N and NO_3-N) was determined in extracts of the non-fumigated samples.

Soil microbial respiration was measured by determining CO_2 production at three to six-day intervals during the incubation. Respired CO_2 was captured in 5.0 ml of 0.5 mol/L NaOH contained in a beaker suspended inside each mason jar(Hu, 1997). The NaOH solution was removed and titrated to determine the amount of CO_2 evolved. The soil microbial respiration was expressed as CO_2 mg/(kg · d) by averaging each interval-period data and accumulated CO_2-C evolution was used as a measurement of total C release over the incubation period. Soil labile C was estimated as the total released-C during the first two-week incubation.

1.4 Statistical analysis

A three-way factorial analysis of variance(ANOVA) with GLM(general linear model) was used to detect the effects of soil types, C and N treatments, and sample times on MBC, MBN, and soil extractable inorganic N. A multiple comparison was performed to tease apart the treatment factors for each soil of each sample date, using LSD (least significant difference) test at $P = 0.05$. A two way ANOVA with GLM was used to test the between-subjects effects of soil types and treatments on total CO_2-C release. For all statistical analyses, SPSS V. 10.0 (SPSS Inc., Chicago, Illinois) software package was used. Unless otherwise noted, all the data were calculated based on oven-dried soils (105℃, 24 h).

2 Results

2.1 Microbial respiration and total carbon evolution

Carbon dioxide productions and their temporal variations differed among the three soils throughout the incubation period(Fig. 1). In the organic soil, CO_2 evolution rates in the CELL and NCELL treatments continued to increase during the first 24 d, and were persistently higher than that in the CONT during most of the incubation, and decreased to the level of CONT by the end of the incubation(Fig. 1). No significant differences of CO_2 production were observed between the CELL and NCELL treatments(averaging at 174.0 and 175.6 mgCO_2/(kg soil·d), respectively) in this soil. In the forest soil, microbes responded quickly to C and N additions. CO_2 evolution rate in the CELL increased to a level of 207.57 mgCO_2/(kg soil·d) during the first nine days and then sustained at that level to the end of the incubation. In the NCELL, the rate peaked quickly by day 15, and then decreased to the level of CONT by day 64. The average rate in the NCELL was 362.6 mgCO_2/(kg soil·d) from 9—40 d, 42.8% higher than that in the CELL. However, it decreased to 165.4 mgCO_2/(kg soil · d) from day 40 to the end of incubation, 38.8% less than that in the CELL. For the grassland soil, CO_2 evolution rate continued to increase in the CELL during the first two weeks of incubation(Fig. 1). On the day 21 of incubation, it showed a slight decrease, and then it stabilized at around 139.1 mgCO_2/(kg soil·d) to the end of the incubation. In the NCELL of the grassland soil, the rate increased rapidly during the first two weeks, and then it decreased sharply and was close to the CONT by the end of incubation. From 12 to 64 d, the average rate in the NCELL was 264.0 mgCO_2/(kg soil·d), 66.1% higher than that in the CELL. However, from day 64 to the end of the

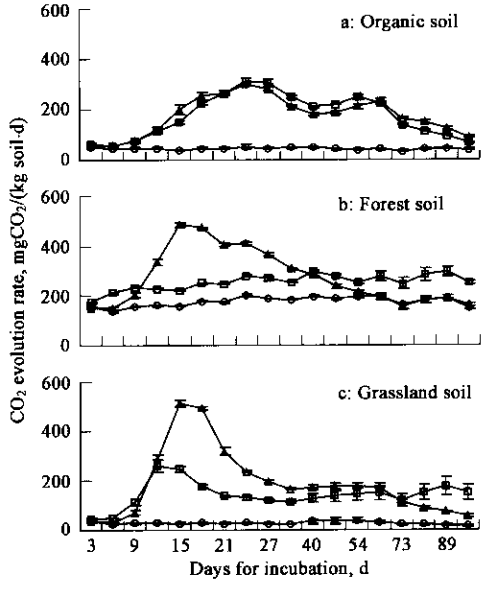

Fig. 1 CO_2 evolution rate as influenced by cellulose and N additions in the three soils

○: CONT; □: CELL; △: NCELL; values ± 1 *SE*; when the error bars are absent, the *SE* is smaller than the symbol

incubation, the average rate was 83.3 mgCO_2/(kg soil·d) in the NCELL, 44.65% less than that in the CELL.

The amounts of total C release were significantly higher ($P < 0.01$) from the CELL and NCELL than from the CONT in all three soils during the incubation period(Fig.2). Also, significant difference in C release was found among the three soils($P < 0.01$; Fig.2). In the organic soil, the amount of released C showed no difference between the CELL and NCELL. The increments (difference of total-released C between the treatments and the control at the end of incubation) were 68.79 and 70.03 mg in the CELL and NCELL, equivalent to 86.0% and 87.5% respectively, of the added cellulose-C. For the forest soil, the amount of C released was significantly less($P < 0.01$) in the CELL than in the NCELL from 15—73 d. At the end of incubation, however, the amounts of C release were similar between the CELL and NCELL. The increments of total C release from the CELL and NCELL were 37.86 and 33.75 mg, accounting for 47.3% and 42.2% respectively, of the added cellulose-C. In the grassland soil, released C was less in the CELL than in the NCELL from day 15 to the end of incubation. The increments of total released-C for the CELL and NCELL were 59.97 and 71.18 mg, amounting to 75.0% and 89.0% of the added cellulose-C, respectively.

2.2 Soil microbial biomass carbon and nitrogen

Statistical tests showed that there were significant differences in MBC and MBN between soil types, treatments and sampling dates(Table 2). MBC and MBN showed similar responses to cellulose and N addition (Table 3). In the organic soil, MBC and MBN were significantly higher on the day 71 in both the CELL and NCELL than in the CONT following the order: NCELL ≥ CELL > CONT. In the forest soil, MBC and MBN showed little change in the CELL throughout the incubation period(Table 3). However, MBC and MBN increased significantly in the NCELL($P < 0.01$), while they decreased slightly in the CONT during the incubation. At the end of incubation (day 71), MBC and MBN in both the CELL and the NCELL were significantly higher than that in the CONT following an order of NCELL > CELL > CONT. For the grassland soil, there were significant increases($P < 0.01$) of MBC and MBN in the CELL and NCELL during the incubation, but no significant change was found in the CONT(Table 3). On the day 24 and 71, MBC and MBN were significantly higher in both the CELL and NCELL than in the CONT according to the following rank: NCELL > CELL > CONT($P < 0.01$).

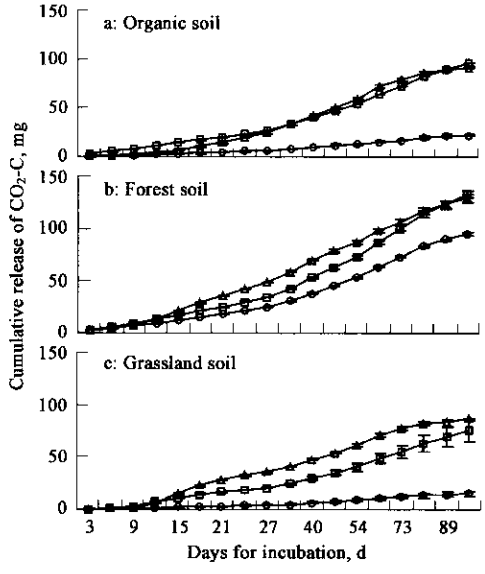

Fig.2 Cumulative release of CO_2-C as influenced by cellulose and N additions in the three soils

○: CONT; □: CELL; △: NCELL; values ± 1 *SE*; when the error bars are absent, the *SE* is smaller than the symbol

Table 2 ***F*-values (with *P* values in parentheses) of a three-way factorial ANOVA on soil microbial biomass C (MBC), biomass N (MBN) and extractable inorganic N(EN)**

Source	*df*	MBC	MBN	EN
Soil type	2	1200.4(<0.01)	2344.5(<0.01)	32003.6(<0.01)
Treatment	2	44.3(<0.01)	27.2(<0.01)	1205.6(<0.01)
Date	2	19.9(<0.01)	7.4(<0.01)	133.3(<0.01)
Soil × treatment	4	9.6(<0.01)	4.3(<0.01)	146.0(<0.01)
Soil × date	4	1.0(0.41)	3.8(<0.01)	95.7(<0.01)
Treatment × date	4	15.8(<0.01)	7.0(<0.01)	272.4(<0.01)
Soil × treatment × date	8	1.2(0.31)	1.0(0.48)	29.8(<0.01)
Residual(error)	81			

2.3 Soil extractable inorganic nitrogen

There existed significant differences in the amount of soil extractable N between soil types, treatments and sampling dates(Table 2). Significant interaction effects were also found between soil type and treatment, soil type and sampling date, treatment and sampling date, and among the three factors, respectively (Table 2). Cellulose addition significantly decreased extractable inorganic N($P < 0.01$) at every stage during the incubation in all three soils(Table 3). During the whole incubation period, the average extractable inorganic N in the CELL of the organic, forest and grassland soils reduced by 27%, 97% and 99% respectively, of that in the CONT. Nitrogen additions initially(day 14) increased the level of extractable inorganic N in the NCELL than in the CONT in all soils, but day 71, extractable soil inorganic N was lower in the NCELL than in the CONT in all the three soils.

3 Discussion

3.1 Effects of carbon input on carbon release

Carbon inputs likely stimulate C release via stimulating microbial activities because soil microbes are usually C-limited. In our experiment, although cellulose enhanced C release in all three soils, the magnitude of the enhancement was significantly different among soils(Fig.2). This difference can be explained by the structure of microbial communities (Paul, 1996) and the status of indigenous N availability(Zak, 2000). In the organic and grassland soils, cellulose dominates the organic C inputs, leading to the dominance of cellulose-based organisms in the microbial communities (Henriksen, 1999). Cellulose addition may, therefore, quickly stimulate the activity (Fig. 1) and population growth (Table 3) of cellulyotic microorganisms when the available N was not limited, leading to greater elevated C release(equal to 85% and 75% of the added-carbon in the two soils, respectively). However, in the forest soil, lignin-based microorganisms likely dominate the microbial community due to the existing lignin-rich substance from litterfall and root material(Keyser, 1978; Yeates, 1997). Stimulation of cellulyotic microbes(Fig.1) in this soil seems to be very limited, resulting a significantly lower elevation of C release(47% of the added C) than in other two soils.

Table 3 Soil microbial biomass carbon (MBC) and nitrogen (MBN) and extractable inorganic nitrogen (EN) as affected by cellulose and N additions in the three soils

Treatments	Incubation stage, d								
	MBC, mgC/kg soil			MBN, mgN/kg soil			EN, mgN/kg soil		
	14	24	71	14	24	71	14	24	71
Organic soil									
CONT	344.8 ± 44.7	382.3 ± 41.4	360.7 ± 10.1	52.6 ± 14.1	51.6 ± 8.1	36.5 ± 17.9	308.0 ± 5.5	324.4 ± 2.5	347.2 ± 5.5
CELL	385.9 ± 58.6	487.4 ± 111.2	492.7 ± 39.4*	45.8 ± 0.9	65.0 ± 12.5	51.6 ± 9.3*	284.4 ± 0.6*	223.3 ± 1.8*	213.2 ± 1.8*
NCELL	372.6 ± 42.1	478.1 ± 88.3	585.1 ± 41.9*	43.7 ± 16.2	65.3 ± 14.4	67.0 ± 3.8*	370.1 ± 6.6*	305.2 ± 1.4*	278.8 ± 1.8*
Forest soil									
CONT	1446.1 ± 13.8	1342.3 ± 91.7	1273.5 ± 17.6	275.7 ± 6.3	262.3 ± 2.3	251.8 ± 6.7	9.0 ± 0.7	13.5 ± 0.7	79.3 ± 4.2
CELL	1396.0 ± 138.9	1436.1 ± 63.3	1536.7 ± 56.6*	281.0 ± 6.3	278.4 ± 3.5	282.9 ± 4.4*	1.0 ± 0.1*	1.2 ± 0.1*	1.1 ± 0.01*
NCELL	1266.2 ± 138.0	1410.1 ± 79.3	1620.4 ± 31.3*	261.6 ± 7.8	286.8 ± 8.0*	297.8 ± 9.8*	49.9 ± 1.6*	21.5 ± 0.2*	32.5 ± 3.1*
Grassland soil									
CONT	572.6 ± 77.9	479.2 ± 18.6	485.0 ± 11.8	98.4 ± 13.9	114.4 ± 10.5	119.9 ± 10.0	47.7 ± 3.6	51.0 ± 1.4	70.4 ± 3.2
CELL	653.0 ± 69.6	708.9 ± 15.4*	857.8 ± 33.3*	127.2 ± 3.3*	127.0 ± 3.5*	139.9 ± 0.9*	0.9 ± 0.1*	0.7 ± 0.04*	0.4 ± 0.1*
NCELL	685.5 ± 18.8	872.9 ± 18.4*	1073.4 ± 112.4*	130.9 ± 6.1*	148.4 ± 2.2*	171.0 ± 2.7*	52.8 ± 2.6*	1.4 ± 0.2*	6.7 ± 0.3*

Notes: * Significant difference between control and treatment ($P < 0.01$); values are means of four replicates ± 1 *SE*

3.2 Effects of nitrogen addition on carbon release in carbon amended soils

Effects of mineral N on soil microbial decomposition are still in debate. Although results from agricultural soils generally showed a stimulating effect(Ekblad, 2002), others found that N addition adversely impacted microbial activities and biomass growth(Keyser, 1978; Carreiro, 2000). In an extensive review, Fog (Fog, 1988) summarized that N addition had stimulating effects only on easily degradable organic compounds but had either no effect or a negative effect on the decomposition of complex organic matter with high C:N ratio(e.g. straw, wood, etc.). Results from our experiment indicated that N addition into different soils had no effect, a stimulating effect, or a retarding effect on soil microbes, respectively. It is possible that additional N inputs to a highly N-enriched soil will not further enhance microbial respiration and biomass C and N, as observed in the organic soil in our experiment (Fig.1, Table 3). However, N-limitation cannot explain the distinct results of respiration (Fig. 1) and total C release obtained in the C plus N treatment between the grassland(ca. 90% stimulation) and forest soils (< 50% stimulation). Both the chemical composition of C substrates and the structure of microbial communities in the native soils may contribute to the observed differences in C release (Bergand, 1997; Ågren, 2001; Davidson, 2003). First, compared to the organic and grassland soils, the forest soil may have much more lignin or lignin-like substances, such as phenolic compounds(Berg, 1997). Added-N can combine with phenolic and other dissolved organic C(Hodge, 1953; Dail, 2001), leading to the formation of highly polymerized products. These compounds can provide chemical barriers (Hodge, 1953; Fog, 1988; Davidson, 2003), which could significantly prevent the major organic C from decomposing in the forest soil(Currie, 1996; Kaiser, 1996). Second, mineral N inputs stimulate cellulolytic microorganisms that are dominant in grassland soils (Ekblad, 2002), but may negatively influence ligninolytic microbes that are dominant in forest soils(Keyser, 1978; Carreiro, 2000), particularly the brown rot and white rot fungi(Keyser, 1978). This difference may explain why additional N input to cellulose-amended soils significantly increased CO_2 release in the grassland soil(Fig. 1), but not in the forest soil. The difference in the enhancement of released-C induced by C and N inputs among the three soils may have important implications for crop residue and waste compost management. Crop residues and waste compost have long been applied in agriculture(Witt, 2000), ecosystem restoration(Lal, 2002) and reclamation of mining areas(Brown, 2003). Our results suggest that newly added-C may be better retained in forest soils than in organic and grassland soils.

3.3 Responses of microbial activity and biomass to carbon and nitrogen additions

New C inputs to soil can stimulate microbial respiration with(Williams, 2000; Ekblad, 2002) or without(Groffman, 1999; Gagnon, 2001; Vance, 2001) an increase in microbial biomass. Results from the present experiment indicate that the status of indigenous N plays a key role in determining which process contributes mainly to the increase in microbial respiration. Cellulose addition stimulated CO_2 evolution(Fig.1) along with increasing MBC and MBN in the organic and grassland soils where N was available(Table 3). In the highly N-limited forest soil, however, cellulose addition accelerated microbial respiration (Fig. 1) but not MBC and MBN because no significant changes in these two properties were found among the three sampling dates in the CELL(Table 3). Addition of cellulose plus inorganic N into this soil stimulated not only CO_2 evolution(Fig.1), but also MBC and MBN (Table 3). These results indicate that N availability mediates the enhanced microbial respiration under new C inputs. This phenomena has been demonstrated in taiga forest floors(Vance, 2001), northern hardwood forest soils(Groffman, 1999), and a degraded potato soil(Gagnon, 2001). The significant differences in the increments of C released under new C addition among the three soils (Fig.2) suggest that soil indigenous N availability could strongly affect soil C dynamics. Stimulated respiration without biomass growth implies that there likely is a high turnover rate of microbial biomass. This high turnover rate may indirectly cause easily degradable C being protected in soils by transferring it to humus compounds, as partially supported by the common evidence that humus and recalcitrant compounds accumulate in soils during SOM decomposition(Billet, 1990; Berg, 1997).

4 Conclusions

The results from the present study indicate that microbes responded to organic C(cellulose) and inorganic N additions differently among soils. Carbon and/or N induced CO_2 release was significantly higher in organic and grassland soils than in the forest one. Nitrogen addition to C-amended soil stimulated microbal CO_2 release in the organic and grassland

soils but reduced it in the forest one. The availability of indigenous C and N can partially explain the differences in microbial responses to resource amendment. Other factors, indigenous resource quality and the microbial community composition in particular, may also profoundly contribute to the dissimilar responses. These results suggest that both resource availability and microbial community in the native soil are important in defining the potential of soil C retention. The results also showed that organic C inputs stimulate microbial activities in all three soils despite significantly different indigenous C and N statuses. However, effects of resource amendments on microbial biomass greatly depend on the indigenous C and N availability. Furthermore, indigenous soil C and O availability substantially impacts the allocation of organic C for microbial biomass growth and/or respiration, potentially regulating the turnover rates of the new organic C inputs.

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