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Nitrogen deposition and carbon sequestration in alpine meadows

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Abstract. Nitrogen deposition experiments were carried out in alpine meadow ecosystems in Qinghai-Xizang Plateau in China, in order to explore the contribution of nitrogen deposition to carbon sequestration in alpine meadows. Two methods were used in this respect. First, we used the allocation of ^{15}N tracer to soil and plant pools. Second, we used increased root biomass observed in the nitrogen-amended plots. Calculating enhanced carbon storage, we considered the net soil CO_2 emissions exposed to nitrogen deposition in alpine meadows. Our results show that nitrogen deposition can enhance the net soil CO_2 emissions, and thus offset part of carbon uptake by vegetation and soils. It means that we have to be cautious to draw a conclusion when we estimate the contribution of nitrogen deposition to carbon sequestration based on the partitioning of ^{15}N tracer in terrestrial ecosystems, in particular in N-limited ecosystems. Even if we assess the contribution of nitrogen deposition to carbon sequestration based on increased biomass exposed to nitrogen deposition in terrestrial ecosystems, likewise, we have to consider the effects of nitrogen deposition on the soil CO_2 emissions.

Introduction

Since the onset of Industrial Revolution, human activities have altered the global nitrogen (N) cycle significantly. A large amount of reactive N originating from fossil-fuel emissions (Galloway et al. 1994) and fertilizer consumption (Matthews 1994) and biomass burning (Crutzen and Andreae 1990) has been deposited into all kinds of ecosystems on Earth. The consequences of N deposition have been a concern widely (Matson et al. 2002; Vitousek et al. 1997) because N limitation of terrestrial productivity is widespread on Earth (Vitousek and Howarth 1991). Accordingly, N deposition has been applied to explain 'missing' carbon (C) sink (Peterson and Melillo 1985; Schindler and Bayley 1993; Galloway et al. 1995; Schimel 1995; Townsend et al. 1996; Holland et al. 1997). Actually, many studies demonstrate that N deposition can result in enhanced C storage in boreal forests through stimulating plant growth (Peterson and Melillo 1985; Schindler and Bayley 1993; Apps et al. 1994; Mäkipää 1995; Turner et al. 1995; Speicker et al. 1996; Nissinen and Hari

1998), and increase C sequestration in both humus layer and mineral soil in boreal forests (Mäkipää 1995).

Besides, N deposition has many dramatic effects on other ecological processes in terrestrial ecosystems, such as root respiration, soil microbial respiration and litter decomposition (including dead roots). Recent research has shown that N addition can enhance (Hunt et al. 1988; Berg and Tamm 1991; Fenn 1991; Prescott et al. 1992; Kuperman 1996), depress (Söderström et al. 1983; Persson and Wirén 1989; Berg and Tamm 1991) or have no effect on decomposition rates (Prescott 1995; Prescott et al. 1999). Possible mechanisms have been put forward, in order to interpret the phenomena above, e.g. it is suggested that decomposition is stimulated by N addition during early stages, while depressed during later stages, in particular during the stage of lignin decomposition (Fogg 1988; Berg and Matzner 1997). Subsequently, it is proven by many studies on the effects of N deposition on extracellular enzyme activities (Hobbie 2000; Osono and Takeda 2001; Saiya-Cork et al. 2002; Vestgarden 2001). Of them, Saiya-Cork et al. (2002) find that N deposition can enhance litter decomposition and depress soil organic matter decomposition. Furthermore, it shows that N additions significantly accelerate the decomposition of light soil C fractions, whereas further stabilize soil C compounds in heavier, mineral-associated fractions (Neff et al. 2002). Long-term N additions can reduce decomposition rates, along with increased lignin accumulation (Magill and Aber 1998). Meanwhile, other studies indicate that N deposition can increase (Månsson and Falkengren-grerup 2003; Gallardo and Schlesinger 1994), reduce (Arnebrant et al. 1996; Aerts and de Caluwe 1999; Berg and Matner 1997; Fog 1988; Persson and Wiren 1989; Persson et al. 2000) or have no significant influences on soil CO_2 emissions (Castro et al. 1994).

This means that N deposition might have an impact on the net soil CO_2 evolution through affecting both litter and soil organic matter decomposition, and thus increase or decrease C storage in terrestrial ecosystems. However, most studies (Holland 1997; Nadellhoffer et al. 1999; Peterson and Melillo 1985; Schindler and Bayley 1993; Townsend et al. 1996) within this research field are restricted to estimate C uptake by vegetation, but few consider whether N deposition can enhance litter decomposition and thus increase soil CO_2 emissions and counteract any increased C uptake by vegetation in terrestrial ecosystems, e.g. Nadellhoffer et al. (1999) suggest that N deposition makes a minor contribution to C sequestration in temperature forests, based on the partitioning of ^{15}N tracer in forests. They do not consider the effects of N deposition on the soil CO_2 emissions. As a result, their budget suffers from some uncertainties.

Most studies about the effects of N deposition on plant growth and litter decomposition and CO_2 emissions are, respectively, conducted in forest ecosystems, because boreal forests have been regarded as significant CO_2 sinks. Moreover, the research on soil CO_2 emissions is only based on soil incubation experiment in the laboratory. Few experiments are done to assess the potential for enhanced C storage in the field in grasslands or in alpine ecosystems, which

function as a potential sink for atmospheric CO_2 (Van Ginkel 1999; Cao et al. 2004). Alpine meadows occupy more than 35% of the Qinghai-Xizang Plateau which extends over 2.5 million km^2. They are very fragile and sensitive to environmental changes due to huge altitude of the Plateau, and are experiencing N deposition of 7.2–10 kg N hm^{-2} yr^{-1} through rain (Zuo et al. 1986), where plant growth is also limited by available N (Cao and Zhang 1999). Hence, N input through rain can enhance C sequestration in alpine meadows. However, little is known about the contribution of N deposition to C sequestration in alpine meadows in the Plateau.

The purposes of this research are to quantify the amount of enhanced C sequestration in alpine meadows exposed to N deposition. Two methods were used in this research: first, we estimated the contribution of N deposition to C sequestration based on the allocation of ^{15}N tracer to soil and plant pools, and second based on increased root biomass observed in the N-amended plots. Calculating the enhanced C sequestration, we considered the net soil CO_2 emission in alpine meadows exposed to N deposition, in order to demonstrate whether N deposition can enhance the net soil CO_2 emission and offset part of C uptake by alpine meadows. Furthermore, we attempt to make sure which is better for estimating the contribution of N deposition to C sequestration in terrestrial ecosystems using the partitioning of ^{15}N tracer in the ecosystem or increased biomass caused by N deposition.

Materials and methods

Sites description

The study area is located in an alpine meadow ecosystem zone in the Qinghai-Xizang Plateau, China, where is characterized by a typical alpine meadow climate: warm and rainy summers and cold, dry and windy winters. Mean annual precipitation is 560 mm and annual temperature averages −1.7 °C at Haibei Research Station from 1976 to 2001. The soil is classified as Mat Crygelic Cambisol. Detailed descriptions about soil and soil microorganisms are available elsewhere (Bao et al. 1995; Chinese Soil Taxonomy Research Group 1995; Jiang et al. 1995; Wang and Li 1995). Total N, inorganic N, C/N ratios, soil moisture and pH values in soils are shown in Table 1. The study area is dominated by *Kobresia* grasses. The dominant species are *Kobresia pygmaea*, *Ptilagrostis concinna*, *Saussurea superba*, *Potentilla nivea*, *Potentilla bifurca*, *Gentiana straminea*, *Leontopodium nanum* and *Thalictrum alpinum*.

In the summer of 2000, three sites, uniform in species composition and cover, were selected along altitude gradients. Site I is located at Haibei Alpine Meadow Ecosystem Station of the Chinese Academy of Sciences, Qinghai Province (37°36′N,101°19′ E, 3215 a.s.l.). Site II is located 16 km northwest of the station (37° 52′ N, 101° 02′ E, 3515 m a.s.l.). Site III is located at the south

Table 1. Total N and inorganic N in soil before N additions. Soil moisture, C/N ratio and pH are also shown

Date 25 Jul	Total N %	Inorganic N g m^{-2}	Soil moisture %	C/N	pH
Site I	0.53 ± 0.04	0.87 ± 0.10	18.7 ± 0.9	18.2 ± 0.7	7.5
Site II	0.50 ± 0.06	0.68 ± 0.20	31 ± 0.01	18.7 ± 0.9	7.8
Site III	0.48 ± 0.01	1.1 ± 0.16	23.4 ± 1.1	18.2 ± 0.7	7.5

All data are for the top 15 cm of soil in the field, with means ± SD of the three sites per treatment.

slope (< 30°) of Oboling, 24 km northwest of the station (38° 00′ N, 100° 55′ E, 3755 m a.s.l.).

^{15}N additions

In the July of 2000, three 9 m^2 plots were established at each site. On the July 26 of 2000, ^{15}N tracers were dissolved in H_2O and sprayed on two of the three plots, respectively. $Na^{15}NO_3$ (99.26 atom%) was applied at 4.4 kg N hm^{-2} yr^{-1}, and $(^{15}NH_4)_2SO_4$ (99.40 atom%) at 5.6 kg N hm^{-2} yr^{-1}. Another amount of H_2O was sprayed on each plot in order to prevent more ^{15}N from being absorbed on the leaves. The total amount of H_2O was equivalent to 2 mm of rain. At the same time, equivalent H_2O was sprayed on the third plot as the control. ^{15}N tracers were added to alpine meadows only for one growth season. Samples of soils, gases and biomass were collected one day before ^{15}N additions and 4 weeks and 8 weeks after ^{15}N additions at each site, and 11–13 months following ^{15}N additions at the site III.

Sampling and analysis

Soil sampling

At each sampling, 15 soil cores (2.7 cm in diameter, 15 cm in depth) were collected randomly on each plot. Every 5 soil cores were combined as one sample. The samples were put into an ice-box and brought into the laboratory. Immediately the samples were mixed well and sieved (< 2 mm). The sieved soils were analyzed for soil moisture, pH, total N, $^{15}N/^{14}N$ ratio and organic C.

Soil moisture was determined gravimetrically and pH was measured by a glass electrode using a 1:2 soil-to-water ratio. Soil organic C was measured by the dichromate digestion method (Kalembasa and Jenkinson 1973). Total N of the samples was determined by Kjeldahl digestion with a salicylic acidification (Pruden et al. 1985). Microbial biomass N was estimated by a chloroform fumigation-direct extraction method (Brookes et al. 1985; Davidson et al. 1989). NH_4^+–N and NO_3^-–N in K_2SO_4 extracts were measured by stream distillation with MgO, using Dewarda's alloy to reduce NO_3 to NH_4 (Bremner 1965).

Plants material sampling

Root biomass was estimated from 2.7 cm diameter soil cores collected to 15 cm depth because more than 95% of the roots are concentrated in this depth in *Kobresia* meadows (Zhou 1982). At each sampling, 15 cores were collected on each plot. Live roots, carefully removed from the soil cores, were rinsed with water. Aboveground biomass was measured by harvesting a 25×25 cm squares ($n = 3$). All plant material was dried in an oven at 60 °C over 48 h. Ground plant material was analyzed for total N, $^{15}N/^{14}N$ ratios and C content.

CO_2 sampling

We only measured soil CO_2 emissions using a closed chamber method in this research. Three square iron frames with channels were inserted 5 cm inside the soils on each plot, respectively. The frame provided a base to mount a closed chamber (40 cm × 40 cm × 25 cm). When we measured soil CO_2 emissions, the channel was filled with water for a tight fit, and the chambers were kept for one and a half hours. Two small battery-operated fans were installed on the chamber ceiling to circulate the air within the chamber. Three-way stopcocks were fitted at the top of the chambers to collect gas samples. CO_2 emissions from the control and N-amended plots at the same site were measured simultaneously. We measured the chamber temperature with a glass thermometer inserted from the top of the chamber. Air temperature and soil temperatures at depths of 0, 5, 10, 15 and 20 cm were also determined. CO_2 concentrations were measured using a Li-Cor6252 CO_2 infrared analyzer. CO_2 emission rates were calculated following methods described by Zhang et al. (2001).

It is impossible to finish all the measurements of CO_2 emissions at all the sites during the same period in one day because it is difficult work to carry out these experiments at over 3200 m above sea level. Previous studies showed that soil CO_2 emission rates measured both from 9:00 to 11:00 and from 15:00 to 17:00 were close to the mean values determined during a-24 h period with 2-h intervals (Cao et al. 2003). Hence, in our experiment most CO_2 emission measurements were finished from 9:00 to 11:00 in the morning, while others were done from 15:00 to 17:00 in the afternoon. Our results also showed that mean CO_2 emission rates of the control plots were compared to those observed in other alpine meadows (Liu et al. 2001; Cao et al. 2004).

^{15}N analysis and calculation

All the samples for ^{15}N analysis followed the methods described by Buresh et al. (1982) and Pruden et al. (1985), except NH_4^+–N and NO_3^-–N samples.

H_2O-free alcohol was distilled after each digest was distilled. The $^{15}N/^{14}N$ ratios were measured on a Finnigan MAT-251 mass spectrometer.

The ^{15}N recovered in plants or inorganic N (NH_4^+-N + NO_3^--N) was measured by multiplying the N concentration of the pool by the mass of the component per square meter and its atom% excess ^{15}N. The ^{15}N recovered in soil microbial biomass was calculated as the difference in the ^{15}N recovered in non-fumigated and fumigated soil samples. The ^{15}N recovered in soil organic matter was calculated as the difference between the ^{15}N recovered in bulk soil and the ^{15}N recovered in the soil microbial biomass and extractable inorganic-N pools (Zogg et al. 2000). Standard deviation of the mean (SD) was calculated to provide a measure of the variance for ^{15}N recovered in different components, root biomass, and soil CO_2 emission measurements.

Results

Soil CO_2 emissions

CO_2 emission rates were in the range of -0.5 to 3.0 μmol mm^{-2} s^{-1} during the 2 months in alpine meadows (Table 2). CO_2 emission rates of the control, added NO_3^- and NH_4^+ plots averaged 1.20, 1.32 and 1.44 μmol m^{-2} s^{-1}, respectively. Because soil temperature has often been described as important variable determining CO_2 emission rates (Braswell et al. 1997), here we have to distinguish which accounts for observed CO_2 differences between soil temperature and N deposition. In our research CO_2 emission rates were in positive correlation with soil temperatures at depth of 5 cm (Figure 1), but we found there're no significant temperature differences between treatments. It indicated that soil CO_2 emissions were mainly enhanced by N deposition in alpine meadows.

Table 2. The effects of N deposition on soil CO_2 emission rates in alpine meadows

Sites	Treatment	CO_2 emission rates (μmol m^{-2} s^{-1})	
		20 Aug	20 Sep
Site I	Control	1.18 ± 0.20	1.43 ± 0.13
	NO_3^-	0.92 ± 0.13	2.07 ± 0.13
	NH_4^+	1.52 ± 0.12	1.36 ± 0.10
Site II	Control	1.89 ± 0.55	−0.47 ± 0.02
	NO_3^-	1.71 ± 0.46	−0.17 ± 0.05
	NH_4^+	3.08 ± 0.44	−0.20 ± 0.03
Site III	Control	2.21 ± 0.75	0.77 ± 0.10
	NO_3^-	2.97 ± 0.54	0.34 ± 0.06
	NH_4^+	1.93 ± 0.40	0.92 ± 0.20

Means ± SD of sites I–III are shown, with three chambers as averaged sub-samples per site. Only soil CO_2 emissions were measured.

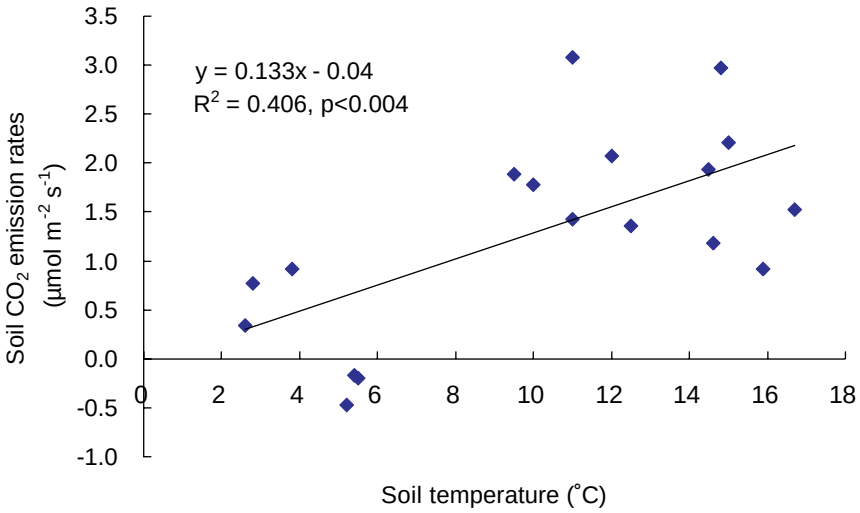

Figure 1. Correlation between soil CO_2 emission rates and soil temperatures at depth of 5 cm.

Based on their soil CO_2 emission rates, the amount of CO_2-emitted from the control, added NO_3^- and NH_4^+ plots was 74.65, 82.11 and 89.58 g C m^{-2} during the 2 months. In the research of Cao et al. (unpublished data) in *Kobresia* meadows they found that CO_2 emission during both August and September was close to 30% of the annual emission. Hence, the annual CO_2 emission from the control, added NO_3^- and NH_4^+ plots was estimated to be about 248.83, 273.70 and 298.60 g C m^{-2}, respectively. Here these values are the gross soil CO_2 source, which includes root respiration and microbial respiration. However, root respiration was considered when we calculated net primary production. So root respiration should be subtracted from the gross soil CO_2 source when the C balance is calculated. Many studies found that the contribution of root respiration to total soil CO_2 source averaged from 10 to 90% depending on vegetation type and season of the year (Hanson et al. 2000). The contribution of live root respiration to the total soil emissions is very high in cold, northern biomes, varying from 50 to 93% in arctic tundra (Billings et al. 1977, 1978; Chapin et al. 1980) and from 62 to 89% in boreal forests (Bonan 1993; Ryan et al.1997), but Hu et al. (2004) found it was about 35 ± 29% in alpine meadows using a basal method (Marshall and Perry 1987). Here we assumed that the contribution of root respiration to the total soil CO_2 source averaged 40% in alpine meadows. As a result, the annual microbial respiration from the control, NO_3^- and NH_4^+ plots was estimated to be about 111.97, 123.17 and 134.37 g C m^{-2}. The area of alpine meadows is about 35% of the whole Qinghai-Xizang Plateau. The net CO_2 emissions exposed to deposited NO_3^- and NH_4^+ in alpine meadows were estimated to be 13.1 and 26.1 Tg C yr^{-1}, respectively. So the total net CO_2 emission could be estimated to be 39.2 Tg C yr^{-1} exposed to N deposition in alpine meadows.

Enhanced C sequestration based on ^{15}N tracers to soil and plant pools

Short-term fate of ^{15}N-labelled ammonium and nitrate was shown in Table 3, which demonstrated that short-term fate of added ammonium and nitrate was significantly different in alpine meadows. More ^{15}N was taken up by both roots and soil organic matter. Plants and soil micro-organisms took up more $NO_3^- - ^{15}N$ than $NH_4^+ - ^{15}N$, while soil organic matter retained more $NH_4^+ - ^{15}N$ than $NO_3^- - ^{15}N$. Detailed short-term fate of ^{15}N tracers at Site III was available (Xu et al. 2003). Long-term fate of ^{15}N tracers at Site I was also available (Xu et al. 2004). It showed that soil organic matter, soil micro-organisms and plants retained 26.3, 26.6 and 39.2% of added $NO_3^- - ^{15}N$, while they recovered 32.3, 15.9 and 14.5% of applied $NH_4^+ - ^{15}N$ after one year following N additions. This implied that the ability to retain NO_3^- was higher than to retain NH_4^+ in alpine meadows. Compared to their short-term fate, more NO_3^- was taken up by plants while more added NH_4^+ was retained in soil organic matter. Here we used short-term allocation of ^{15}N tracers to soil and plant pools in alpine meadows because there're no significant differences between shot-term and long-term allocation of ^{15}N tracers to soil organic matter and plant pools. The amount of input N with rain was about 7.2–10 kg N hm^{-2} yr^{-1} in the alpine meadow in Qinghai-Xizang Plateau (Zuo et al. 1986). We assumed that the amount of N input through rain was 10.0 kg–hm^{-2} yr^{-1}. So the magnitude of deposited N in alpine meadows in Qinghai-Xizang Plateau was estimated to be about 0.88 Tg yr^{-1}. Following the methods described by Nadelhoffer et al. (1999), the CO_2 uptake by alpine meadows caused by N deposition was estimated to be about 16.7 Tg C yr^{-1} (Table 4).

Enhanced C sequestration based on enhanced root biomass exposed to N deposition

Heer and Körner (2002) showed that high elevation pioneer plants were sensitive to mineral nutrient addition, and exhibited massive biomass stimulations at 2500 m a.s.l. in the Swiss Central Alps. A similar result was obtained in our research. It showed that N addition had a big boost on biomass in alpine meadows over 3200 m a.s.l. in the Qinghai-Xizang Plateau, and added N played a different role in stimulating aboveground and root biomass (Figures 2 and 3). Here we just considered root biomass because it was significantly higher than aboveground biomass, and N deposition stimulated it more than aboveground biomass in alpine meadows. Root biomass decreased to the minimum On September 6. 11–13 months after ^{15}N additions, both aboveground and belowground biomass in N-amended plots increased substantially (Figures 4 and 5). It indicated that it is not temporary for the effects of N deposition on biomass in N-limited alpine meadows.

Table 3. Proportion of ^{15}N recovered in soil and plant components 8 weeks following ^{15}N additions

			Plant components			Soil components			
Sites	Location	Treatment	Root	Green	Litter	MB	SOM	IN	Total recovery
Site I	37°36′N,	20 Sep							
	101°19′E	$^{15}NO_3^-$	36.6 ± 2.5	2.8 ± 2.3	0.21 ± 0.1	9.3 ± 3.1	0.7 ± 2.4	0.26 ± 0.04	49.9 ± 10.4
		$^{15}NH_4^+$	22.5 ± 2.4	8.8 ± 0.8	1.3 ± 0.4	11.6 ± 2.2	9.5 ± 9.0	0.04 ± 0.01	53.7 ± 14.8
Site II	37°52′N,	20 Sep							
	101°02′E	$^{15}NO_3^-$	45.9 ± 4.5	4.7 ± 1.7	0.43 ± 0.16	12.3 ± 2.2	24.5 ± 3.7	0.07 ± 0.00	87.5 ± 9.1
		$^{15}NH_4^+$	34.1 ± 0.3	2.4 ± 0.6	1.0 ± 0.3	8.4 ± 2.4	25.5 ± 7.2	0.24 ± 0.12	70.5 ± 10.3
Site III	38°00′N,	20 Sep							
	100°55′E	$^{15}NO_3^-$	34.7 ± 1.3	5.7 ± 1.5	0.3 ± 0.2	8.5 ± 0.6	18.0 ± 3.4	0.14 ± 0.04	67 ± 12.3
		$^{15}NH_4^+$	21.3 ± 2.2	2.3 ± 1.0	0.4 ± 0.2	5.9 ± 0.5	19.5 ± 1.2	0.09 ± 0.01	49.3 ± 5.3

Means ± SD of sites I–III are shown, with three cores as averaged sub-samples per site.
MB – Microbial biomass, SOM – soil organic matter, IN – inorganic nitrogen pool.

Table 4. Estimated CO_2–C uptake by alpine meadows due to nitrogen deposition

Alpine meadow pool (1)	Nitrogen deposition allocated to pool (%) (2)	C/N (3)	N deposition in pool (Tg) (=0.88 Tg×(2)) (4)	CO_2–C taken up (Tg) (=(3) × (4)) (5)
Root	32.5	34.9	0.29	10.1
Green	6.1	17.7	0.05	0.9
Litter	0.6	21.8	0.005	0.1
Soil	32.6	19.4	0.29	5.6

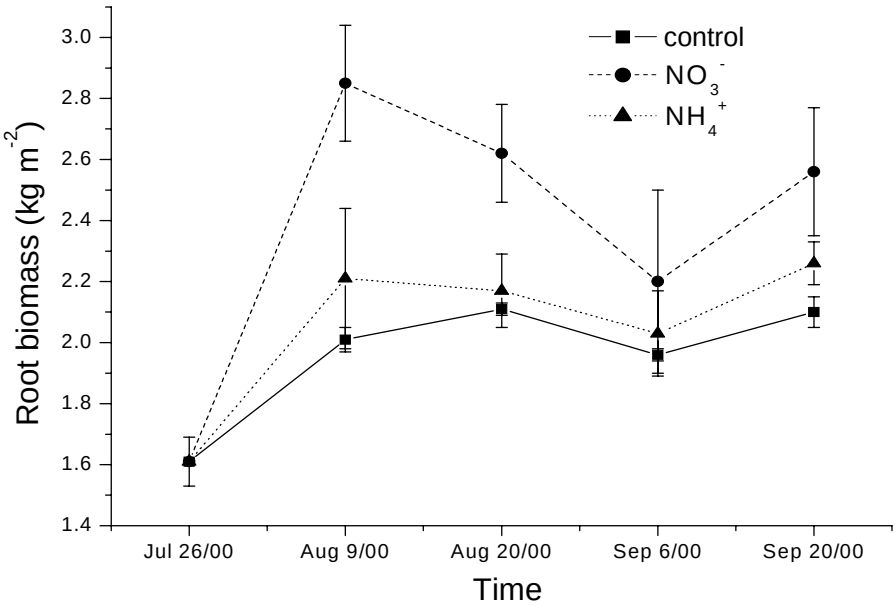

Figure 2. Root biomass exposed to enhanced N deposition in an alpine meadow. Vertical bars indicate the standard deviation of the measurement mean (±SD) for three sites per treatment. An increase in root biomass after Sep 6 in 2000 originates from the growth of cold-tolerant species.

The amount of C sequestration caused by N deposition was calculated by multiplying C content of roots biomass and the difference of that between the fertilized and non-fertilized. Therefore, the CO_2 uptake by alpine meadows due to NO_3^- + NH_4^+ deposition was estimated to be about 256 Tg C yr^{-1}. The turnover of live roots is about 3–4 years (Cao and Zhang 2001). So about 30% of roots will be dead in the coming year. Decomposition experiment using a buried-bag incubation technique showed that 16.25% of dead roots was lost due to decomposition during the first year and 70% of dead roots detritus could reside in upper soils for a long time (Cao and Zhang 2001). Here we assumed that 50% of dead roots resided in the soils in the form of soil organic

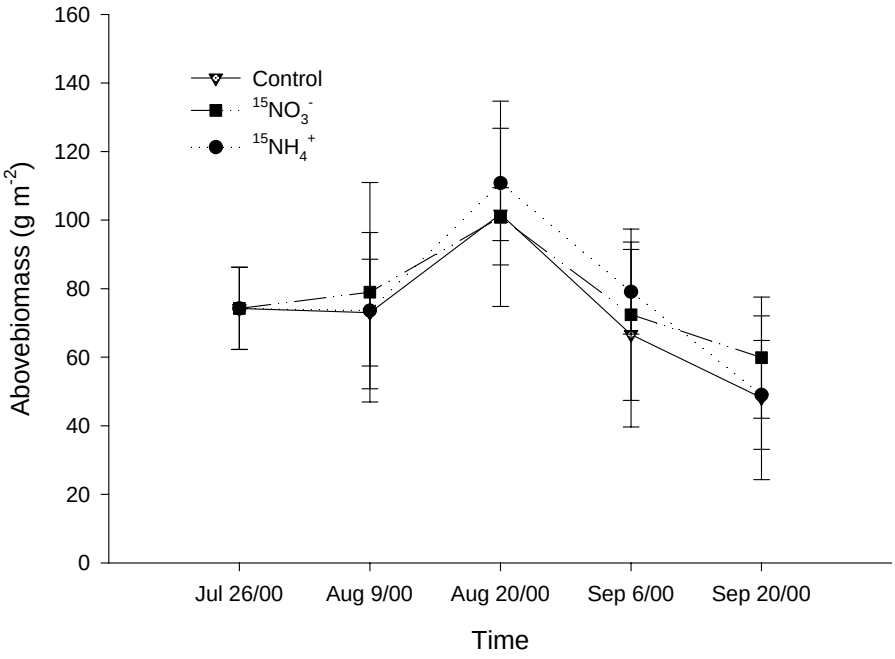

Figure 3. Aboveground biomass exposed to enhanced N deposition in an alpine meadow. Vertical bars indicate the standard deviation of the measurement mean for three sites per treatment.

matter. So the enhanced C sequestration by alpine meadows caused by N deposition should be estimated to be about 217.6 Tg C yr^{-1}.

Discussions

Nadelhoffer et al. (1999) show that N deposition makes a minor contribution to C sequestration in temperate forests through the partitioning of added ^{15}N in forests. However, our results imply that their budget is experiencing some uncertainties. First, the contribution of N deposition to C sequestration in alpine meadows is 16.7 Tg C yr^{-1} based on the allocation of ^{15}N tracer to the soil and plant pools, and 217.6 Tg C yr^{-1} based on increased root biomass observed in the N-amended plots. The latter is an order of magnitude greater than the former. Apparently, the contribution of N deposition to C sequestration can be seriously underestimated using the partitioning of ^{15}N tracers in terrestrial ecosystems. Second, N deposition has a great influence on the soil CO_2 evolution. In our research N deposition can enhance the net soil CO_2 emissions. It means we should subtract it from the enhanced C storage caused by N deposition. It is 178.4 Tg C yr^{-1} when we subtract the net soil CO_2 emission from the increased root biomass caused by N deposition, which implies that the

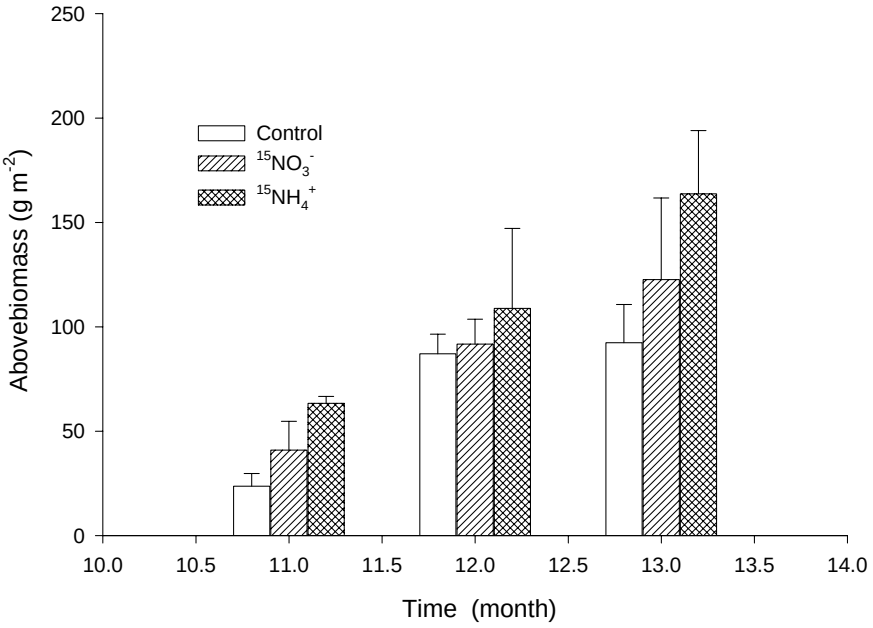

Figure 4. Aboveground biomass after 11–13 months following ^{15}N additions. Vertical bars indicated the standard deviation of the measurement mean for three sub-samples per treatment at site III.

enhanced C uptake by vegetation exposed to N deposition can be overestimated in alpine meadows if the effects of N deposition on soil CO_2 emissions are not taken into account. Regard to another estimate of C uptake by alpine meadows, the amount of the net CO_2-C emission caused by N deposition exceeds the value estimated using the allocation of ^{15}N tracer to soil and root pools. It indicates that N deposition might have an impound impact on all the aspects of terrestrial ecosystems. Many researchers (Legg and Stanford 1967; Saphozhnikov 1969; Broadbent and Nakashima 1971; Westerman and Kurtz 1973, 1974) find that added N can apparently increase mineralization of native organic N following ^{15}N additions both under laboratory and field conditions. Bowman and Steltzer (1998) show that N deposition cause replacement of the dominant plant species in alpine tundra, and lead to an eightfold increase in the net mineralization and nitrification. Therefore, we suspect that deposited N result in an increase in the net N mineralization in alpine meadows, then deposited N and mineralized N stimulate plant growth together. It means that we have to be cautious to draw a conclusion when we estimate the contribution of N deposition to C sequestration based on the partitioning of ^{15}N tracers in terrestrial ecosystems, in particular in N-limited ecosystems. Even if we assess the contribution of N deposition to C sequestration using increased terrestrial biomass or increased C in soils caused by N deposition, likewise, we have to be cautious. The reason is

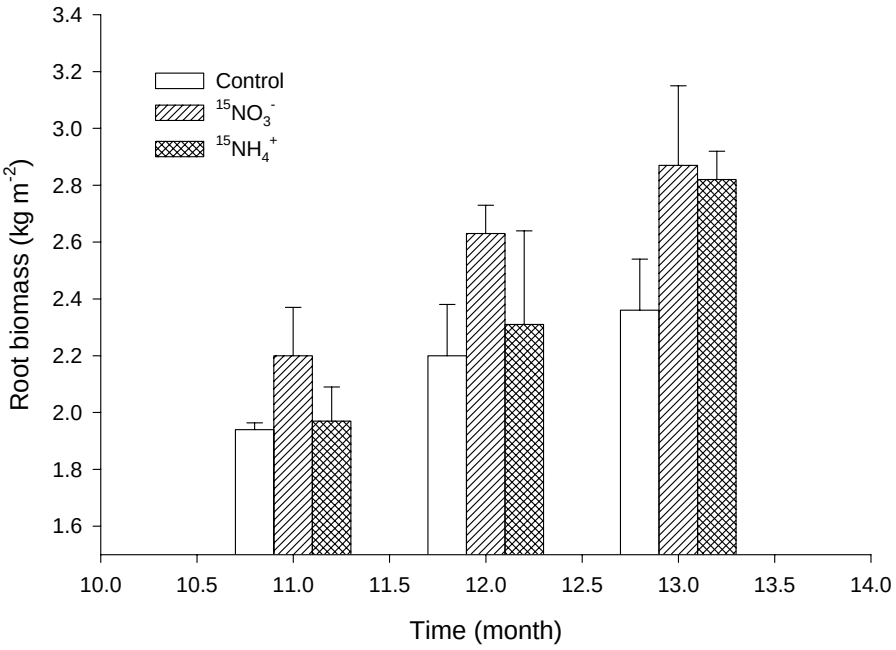

Figure 5. Root biomass after 11–14 months following ^{15}N additions. Vertical bars indicate the standard deviation of the measurement mean for three sub-samples per treatment at sites III.

that N deposition can have great influence on other ecological processes related to C sequestration, such as litter decomposition and soil CO_2 emissions, which affects our estimate of the contribution of N deposition to C sequestration. Our results shows that it is a better method to assess the contribution of N deposition to C sequestration in terrestrial ecosystems using increased biomass and increased C in soils caused by N deposition, along with considering the effects of N deposition on the net soil CO_2 evolution.

Fang et al. (2001) have shown that the annual forest C storage in China is estimated to be about 0.021 Pg in China during the past two decades. Cao et al. (2003) has reported that net ecosystem production in China during the period of 1981–2000 varies between -0.32 and 0.25 Pg C yr^{-1}, with a mean value of 0.07 Pg C yr^{-1}. Compared to the annual forest C storage and net ecosystem production in China, C sequestration caused by N deposition is very high in alpine meadows in the Qinghai-Xizang Plateau in China. It can be estimated to be 0.18 Pg C yr^{-1}, which can compared to C uptake (0.29 to 0.35 Pg C yr^{-1}) stimulated by anthropogenic N deposition in North America (Holland and Brown 1999). What's more, plant growth is still limited by available N supplies in alpine meadows, with an output of 159.35 kg hm^{-2} yr^{-1} and an input of 84.73 kg hm^{-2} yr^{-1} (Cao and Zhang 1999). Additionally, the decomposition of dead roots and soil organic matter is very slow in alpine meadows due to

lower temperature. Most of soil organic matter derived from dead roots can reside in soils for decades. It means that N deposition can simulate C sequestration in the coming decades, even hundreds of years. As a result, N deposition might make a great contribution to C sequestration in alpine meadows in the Qinghai-Xizang Plateau.

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References

Aerts R. and De Caluwe H. 1999. Nitrogen deposition affects on carbon dioxide and methane emissions from temperate peatland soils. Oikos 84: 44–54.

Apps M.J. and Kurtz W.A. 1994. In: Kanninen M. (ed.), Carbon Balance of World's Forested Ecosystem Towards a Global Assessment. Academy of Finland, Helsinki.

Arnebrant K., Bååth E., Söderström B. and Norhrstedt H.-ö. 1996. Soil microbial activity in eleven Swedish coniferous forests in relation to site fertility and nitrogen fertilization. Scand. J. Forest Res. 11: 1–6.

Bao X.K., Cao G.M. and Gao Y.X. 1995. Accumulation of organic matter in Cryo-sod soil. Approach on Chinese Soil Tax. pp.152–160.

Braswell B.H., Schimel D.S. and Linder E. et al. 1997. The response of global terrestrial ecosystems to interannual temperature variability. Science 278: 870–872.

Berg B. and Matzner E. 1997. Effect of N deposition on decomposition of plant litter and soil organic matter in forest systems. Environ. Rev. 5: 1–25.

Berg B. and Tamm C.O. 1991. Decomposition and nutrient dynamics of litter in long-term optimum nutrition experiments. I. Organic matter decomposition in *Picea abies* needle litter. Scand. J. Forest Res. 6: 305–321.

Billings W.D., Peterson K.M. and Shaver G.R. 1977. Growth, turnover, and respiration rates of roots and tillers in tundra graminoids. In: Tieszen L.L. (ed.), Vegetation and Production Ecology of an Alaskan Arctic Tundra. Springer-Verlag, New York, pp. 415–434.

Billings W.D., Peterson K.M., Shaver C.G. and Trent A.W. 1978. Root growth, respiration, and carbon dioxide evolution in an arctic tundra soil. Arc. Alp. Res. 9: 129–137.

Bonan G.B. 1993. Physical controls of the carbon balance of boreal forest ecosystems. Can. J. Forest Res. 23: 1453–1471.

Bowman W.D. and Steltzer H. 1998. Positive feedbacks to anthropogenic nitrogen deposition in Rocky Mountain alpine tundra. Ambio 27: 514–517.

Bremner J.M. 1965. Inorganic forms of nitrogen. In: Black C.A. (ed.), Methods of Soil Analysis Vol. 2. American Society of Agronomy, Madison, pp. 1179–1237.

Broadbent F.E. and Nakashima T. 1971. Effect of added salts on nitrogen mineralization in three California soils. Soil Sci. Soc. Am. Proc. 35: 457–460.

Brookes P.C., Landman A., Pruden G. and Jenkinson D.S. 1985. Chloroform fumigation and the release of soil nitrogen: a rapid direct extraction method to measure microbial biomass nitrogen in soil. Soil Bio. Biochem. 17: 837–842.

Buresh R.J., Austin E.R. and Creswell E.T. 1982. Analytical methods in ^{15}N research. Fer. Res. 3: 37–47.

Cao G.M. and Zhang J.X. 1999. The nitrogen cycle in an alpine meadow ecosystem. Acta Eco. Sin. 19: 509–513.

Cao G.M. and Zhang J.X. 2001. Soil nutrition and substance cycle of *Kobresia* meadow. In: Zhou X.M. (ed.), Alpine Kobresia Meadows in China. Science Press, Beijing, pp. 188–216.

Cao G.M., Tang Y.H., Mo W.H., Wang Y.S., Li Y.N. and Zhao X.Q. 2004. Grazing intensity alters soil respiration in an alpine meadow on the Tibetan plateau. Soil Bio. Biochem. 36: 237–243.

Cao G.M., Zhang J.X., Zhao X.Q., Li Y.N. and Zhou X.M. 2003. Methods of estimating CO_2 emissions from Mat Cry-sod soil. ACTA Pedologica Sinica 39(2): 261–267.

Cao M.K., Prince S.D., Li K.R., Tao B., Small J. and Shao X.M. 2003. Response of terrestrial carbon uptake to climate interannual variability in China. Glob. Change Biol. 9: 536–546.

Castro M.S., Peterjohn W.T., Melillo J.M. and Steudler P.A. 1994. Effects of nitrogen fertilization on the fluxes of N_2O, CH_4, and CO_2 from soils in a Florida slash pine plantation. Can. J. Forest Res. 24: 9–13.

Chapin F.S. III, Miller P.C., Billings W.D. and Coyne P.I. 1980. Carbon and nutrient budgets and their control in coastal tundra. In: Brown J., Miller P.C., Tieszen L.L. and Bunnell F.L. (eds), An Arctic Ecosystem: The Coastal Tundra at Barrow Alaska. Dowden, Hutchinson and Ross, Stroudsburg, P.A., U.S.A., pp. 458–482.

Chinese Soil Taxonomy Research Group 1995. Chinese Soil Taxonomy. Science Press, Beijing, pp. 58–147.

Crutzen P.J. and Andreae M.O. 1990. Biomass burning in the tropics: impact on atmospheric chemistry and biogeochemical cycles. Science 250: 1669–1678.

Davidson E.A., Eckert R.W., Hart S.C. and Firestone M.K. 1989. Direct extraction of microbial biomass nitrogen from forest and grassland soils of California. Soil Bio. Biochem. 21: 773–777.

Fang J.Y., Chen A.P., Peng C.H., Zhao S.Q. and Ci L.J. 2001. Changes in forest biomass carbon storage in China between 1949 and 1998. Science 292: 2320–2322.

Fenn M. 1991. Increased sited fertility and litter decomposition rate in high-pollution sites in San Bernardino Mountains. Forest Sci. 37: 1163–1181.

Fog K. 1988. The effect of added nitrogen on the rate of decomposition of organic matter. Biol. Rev. 63: 433–462.

Gallardo A. and Schlesinger W.H. 1994. Factors limiting microbial biomass in the mineral soil and forest floor of a warm-temperate forest. Soil Bio. Biochem. 26: 1409–1415.

Galloway J.N., Levy H. and Kasibhatla P.S. 1994. Year 2020: consequences of population growth and development on deposition of oxidized nitrogen. Ambio 23: 120–123.

Galloway J.N., Schlesinger W.H., Levy H., Michael A. and Trolier M. 1995. Nitrogen fixation: Anthropogenic enhancement-environmental response. Glob. Biogeochem. Cycle 9: 235–252.

Hanson P.J., Edwards N.T., Carten C.T. and Andrews J.A. 2000. Separating root and soil microbial contribution to soil respiration: a review of methods and observations. Biogeochemistry 48: 115–146.

Heer C. and Körner C. 2002. High elevation pioneer plants are sensitive to mineral nutrient addition. Basic Appl. Ecol. 3(1): 39–47.

Hobbie S.E. 2000. Interactions between litter lignin and soil nitrogen availability during leaf litter decomposition in a Hawaiian montane forest. Ecosystems 3: 484–494.

Holland E.A. and Brown S. 1999. North American carbon sink. Science 283: 1815.

Holland E.A., Braswell B.H., Lamasque T.F., Townsend A., Sulzman T., Muller T.F., Dentener F., Brasseur G., Levy II H., Penner T.E. and Roelofs G.T. 1997. Variations in the predicted spatial distribution of atmospheric nitrogen deposition and their impact on carbon uptake by terrestrial ecosystems. J. Geophys. Res. 102: 15849–15866.

Hu Q.W., Cao G.M., Wu Q., Li D. and Wang T.S. 2004. Comparative study on CO_2 emission rate from different alpine ecosystems during grass exuberance period. J. Geographys. Sci. 14: 167–176.

Hunt H.W., Ingham E.R., Coleman D.C., Elliott E.T. and Reid C.P.P. 1988. Nitrogen limitation of production and decomposition in prairie, mountain meadow, and pine forest. Ecology 69: 1009–1016.

Jiang W.B., Wang Q.L., Yang T. and Geng B.W. 1995. Seasonal dynamics of cellulose decomposition in alpine meadow soil. Alpine Meadow Eco. 4: 187–187.

Kalembasa S.J. and Jenkinson D.S.A 1973. Comparative study of titrimetric and gravimetric methods for determination of organic carbon in soil. J. Sci. Food Agr. 24: 1085–1090.

Kuperman R.G. 1996. Relationships between soil properties and community structure of soil macroinvertebrates in oak-hickory forests along an acidic deposition gradient. Appl. Soil Eco. 4: 125–137.

Legg J.Q. and Stanford G. 1967. Utilization and fertilizer by oats in relation to the available N status of soils. Soil Sci. Soc. Am. Proc. 31: 215–219.

Liu Y.F., Ouyang H., Cao G.M., Luo J., Zhang X.Z., Zhao X.Q. and Yang Q.W. 2001. Soil carbon emission from ecosystems of eastern Qinghai-Tibet Plateau. J. Nat. Res. 16(2): 152–160.

Magill A.H. and Aber J.D. 1998. Long-term effects of experimental nitrogen additions on foliar litter decay and humus formation in forest ecosystems. Plant Soil 203: 301–311.

Mäkipää R. 1995. Effect of nitrogen input on carbon accumulation of boreal forest soils and ground vegetation. For. Eco. Manage. 79: 217–226.

Månsson K.F. and Falkengren-Grerup U. 2003. The effect of nitrogen deposition on nitrification, carbon and nitrogen mineralization and litter C:N ratios in oak (*Quercus robur* L.) forests. For. Eco. Manage., 179: 455–467.

Marshall J.D. and Perry D.A. 1987. Basal and maintenance respiration of mycorrhizal and non-mycorrhizal root systems of conifers. C. J. Forest Res. 17: 872–877.

Matson P., Lohse K.A. and Hall S.J. 2002. The globalization of nitrogen deposition: consequences for terrestrial ecosystems. Ambio 31: 113–119.

Matthews E. 1994. Nitrogenous fertilizers: global distribution of consumption and associated emissions of nitrous oxide and ammonia. Glob. Biogeochem. Cycle 8: 411–439.

Nadelhoffer K.J., Emmett B.A., Gundersen P., Kjønaas O.J., Koopmans C.J., Schleppi P., Tietema A. and Wright R.F. 1999. Nitrogen deposition makes a minor contribution to carbon sequestration in temperate forests. Nature 398: 145–148.

Neff J.C., Townsend A.R., Gleixner G., Lehman S., Turnbell J. and Bowman W.D. 2002. Variable effects of nitrogen additions on stability and turnover of soil carbon. Nature 419: 915–917.

Nissinen A. and Hari P. 1998. Effects of nitrogen deposition on tree growth and soil nutrients in boreal Scott pine stands. Environ. Pollu. 102: 61–68.

Osono T. and Takeda H. 2001. Effects of organic chemical quality and mineral nitrogen addition on lignin and holocellulose decomposition of beech leaf litter by *Xylaria* sp. Eur. J. Soil Biol. 37: 17–23.

Persson T. and Wirén A. 1989. Microbial activity in forest soils in relation to acid/base and carbon/nitrogen status. Meddelelser fra Norsk institutt for skogforskning 42(1): 83–94.

Persson T., Karlsson P.S., Seyferth U., Sjöberg R.M. and Rudebeck A. 2000. Carbon mineralization in European forest soils. In: Schulze and E.-D. (ed.), Ecological Studies—Carbon and Nitrogen Cycling in European Forest Ecosystems. Springer, Berlin, pp. 257–275.

Peterson B.J. and Melillo J.M. 1985. The potential storage of carbon caused by eutrophication of the biosphere. Tellus 37B: 117–127.

Prescott C.E. 1995. Does nitrogen availability control rates of litter decomposition in forests? Plant Soil 168: 83–88.

Prescott C.E., Kabzams R. and Zabek L.M. 1999. Effects of fertilization on decomposition rate of *Populus tremuloides* foliar in a boreal forest. Can. J. Forest Res. 29: 393–397.

Proscott C.E., Corbin J.P. and Parkinson D. 1992. Immobilization and availability of N and P in the forest floors of fertilized Rocky Mountain coniferous forests. Plant and Soil 143: 1–10.

Pruden G., Powlson D.S. and Jenkinwson D.S. 1985. The measurement of ^{15}N in soil and plant material. Fert. Res. 6: 205–218.

Ryan M.G., Lavigne M.G. and Gower S.T. 1997. Annual carbon cost of autotrophic respiration in boreal forest ecosystems in relation to species and climate. J. Geophys. Res. 102(D24): 28871–28884.

Saiya-Cork K.R., Sinsabaugh R.L. and Zak D.R. 2002. The effects of long term nitrogen deposition on extracelluar enzyme activity in an *Acer saccharum* forest soil. Soil Bio. Biochem. 34: 1309–1315.

Saphozhnikov N.A., Nesterova E.I., Rusinova I.P., Sirota L.B. and Vanova T.K.L. 1969. The effect of fertilizer nitrogen on plant uptake of nitrogen from different podzolic soils. Trans. Int. Congr. Soil Sci. 9th, 2: 467–474.

Schimel D. 1995. Terrestrial ecosystems and carbon cycle. Glob. Change Biol. 1: 77–91.

Schindler D.W. and Bayley S.E. 1993. The biosphere as an increasing sink for atmospheric carbon: estimates from increased nitrogen deposition. Glob. Biogeochem. Cycle 7(4): 717–733.

Söderström B., Bååth E. and Lundgren B. 1983. Decrease in soil activity and biomass owing to nitrogen amendments. Can. J. Microbiol. 29: 1500–1506.

Speicker H., Mielikäinen K., Köhl M. and Skovsgaard J.P. 1996. Growth Trends in European Forests. Springer, Berlin.

Townsend A.R., Braswell B.H., Holland E.A. and Penner J.E. 1996. Spatial and temporal patterns in terrestrial carbon storage due to deposition of fossil fuel nitrogen. Eco. Appl. 6: 806–814.

Turner D.P., Koerper G.J., Harmon M.E. and Lee J.J. 1995. A carbon budget for forests of the conterminous United States. Ecol. Appl. 5: 421–436.

Van Ginkel J.H., Whitmore A.P. and Gorissen A. 1999. Lolium perenne grasslands may function as a sink for atmospheric carbon dioxide. J. Environ. Qual. 28: 1580–1584.

Vestgargen L.S. 2001. Carbon and nitrogen turnover in the early stage of Scots pine (*Iinus sylvestris* L.) needle litter decomposition: effects of internal and external nitrogen. Soil Bio. Biochem. 33: 465–474.

Vitousek P.M. and Howarth R.W. 1991. Nitrogen limitation on land and sea: how can it occur? Biogeochemistry 5: 7–34.

Vitousek P.M., Aber J.D., Howarth R.W., Likens G.E., Matson P.A., Schindler D.W., Schlesinger W.H. and Tilman D.G. 1997. Human alteration of the global nitrogen cycle: sources and consequences. Ecol. Appl. 7(3): 737–750.

Wang Q.L. and Li J.Z. 1995. Seasonal dynamics of fungal biomass in different vegetation soil of alpine meadow ecosystem. Alpine Meadow Eco. 4: 169–178.

Westerman R.L. and Kurtz L.T. 1973. Priming effect of ^{15}N-labelled fertilizers on soil nitrogen in field experiments. Soil Sci. Soc. Am. Proc. 37: 725–727.

Westerman R.L. and Kurtz L.T. 1974. Effects of salts and salts plus nitrogen-15-labelled ammonium chloride in mineralization of soil nitrogen nitrification, and immobilization. Soil Sci. Soc. Am. Proc. 38: 602–605.

Xu X.L., Ouyang H., Pei Z.Y. and Zhou C.P. 2003. Fate of ^{15}N labeled nitrate and ammonium salts added to an alpine meadow in the Qinghai-Xizang Plateau, China. Acta Bot. Sin. 45(3): 276–281.

Xu X.L., Ouyang H., Pei Z.Y. and Zhou C.P. 2004. Long-term partitioning of ammonium and nitrate among different components in an alpine meadow ecosystem. Acta Bot. Sin. 46(3): 279–283.

Zhang J.X., Cao G.M. and Zhou D.W. 2001. Duel and seasonal changes of carbon dioxide emission from mollic-cryic cambisols on degraded grassland. Acta Pedol. Sin 38(1): 32–39.

Zhou X.M. 1982. Charateristics and main types of *Kobresia* meadows on the Qinghai-Tibet Plateau. ACTA Biol Plateau Sin. (1): 151–160.

Zogg G.P., Zak D.R., Pregiter K.S. and Bruton A.J. 2000. Microbial immobilization and the retention of anthropogenic nitrogen in a northern hardwood forest. Ecology 81: 1858–1866.

Zuo K.C. et al. 1986. Preliminary research on the content of plant nutrients of precipition in haibei alpine meadow ecosystem, Qinghai. ACTA Bio. Plateau Sin. (5): 35–43.