Effects of nitrogen addition on fluxes and concentrations of dissolved organic matter and inorganic nitrogen under a temperate old-growth forest in northeast China

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Abstract
Soil solutions at 15 cm and 60 cm under a Korean pine and broadleaf mixed forest (>200 years old) at Changbai mountain, northeast China, were sampled using porous ceramic cups from July 2006 to October 2008, to study the effects of nitrogen (N) addition on fluxes and concentrations of dissolved organic matter and inorganic N. The soil net N mineralization and wet atmospheric depositions of N and dissolved organic carbon (C) were also measured. The addition of N sources such as $(NH_4)_2SO_4$, NH_4Cl and KNO_3 at rates of 2.25 and 4.5 g N/m^2 each year tended to increase concentrations and fluxes of inorganic N and dissolved organic N in soil solutions at 15 cm and 60 cm depths and soil net N mineralization, and it reduced leaching losses of soil dissolved organic C. The concentration ratios of dissolved organic C to dissolved organic N and special UV absorbance values in the soil solutions at both depths were smaller under the N-fertilized forest plots than under non-fertilized plots. Soil net N mineralization under the N-fertilized forest plots can contribute to the leaching losses of inorganic N from the soil. Our observations indicate that N inputs to temperate forest floors can affect the status of N and C processes in underlying forest soils.

Key words
Dissolved organic carbon and nitrogen, inorganic nitrogen leaching, forest soil, nitrogen mineralization

Introduction
Soil solution chemistry can be considered a sensitive indicator of biogeochemical processes under forest stands, responding quickly to disturbances or stresses like nitrogen input (e.g. McDowell *et al.* 2004; Pregitzer *et al.* 2004; Michel *et al.* 2006). Hence, it is important to study the dynamics of fluxes and concentrations of dissolved organic matter and inorganic nitrogen (N) in soil solutions at the various depths under different forest stands.
The addition of N sources to temperate forest floors can usually increase N leaching as DON and inorganic N from forest topsoils (e.g. McDowell *et al.* 1998; Michalzik *et al.* 2001; Pregitzer *et al.* 2004). The N inputs from forest topsoils may increase the activities of soil microorganisms and the mineralization of carbon (C) in underlying soils, thus releasing CO_2 into the soil solution (Xu *et al.* 2009). Hence, the fluxes and concentrations of dissolved organic matter (DOM) and inorganic N in soil solutions at the various depths would promote our understanding of C and N processes in the underground canopy under N-fertilized forest stands.
At present, there were many contrasting results regarding N effects on DON and DOC dynamics in forest soil solution in field and laboratory studies (McDowell *et al.* 1998, 2004; Magill and Aber, 2000; Pregitzer *et al.* 2004; Michel *et al.* 2006). Furthermore, these earlier studies mainly focused on the dynamics of DOC, DON and inorganic N concentrations in forest soil solutions sampled using zero-tension lysimeters rather than using suction cups. There are rather limited reports about the concentrations and fluxes of dissolved organic matter and inorganic N in soil solutions at the various depths under N-fertilized forest stands, especially in northeast Asia.
In this study, soil solutions at 15 cm and 60 cm under a Korean pine and broadleaf mixed forest (>200 years old) at Changbai mountain, northeast China, was sampled using porous ceramic cups from July 2006 to October 2008, to study the effects of N addition on the fluxes and concentrations of DOM and inorganic N. The soil net N mineralization and wet atmospheric depositions of N and dissolved organic C were also measured. The objectives of this work were to 1) study the effects of N addition on the fluxes and concentrations of DOM and inorganic N in forest soil solution; and 2) to assess the contribution of soil net N mineralization and wet atmospheric C and N depositions to these fluxes. The results would improve our understanding of N and C processes in underlying forest soils due to the increase in atmospheric N inputs.

Materials and methods

Forest stand site and soil properties

Field experiment was located under a Korean pine and broadleaf mixed forest (*Pinus koraiensis* mainly mixed with hardwood trees such as *Tilia amurensis, Fraxinus mandsburica* and *Quercus mongolica*, >200 years old, altitude 738 m above sea level) nearby the National Research Station of Changbai Mountain Forestry Ecology, northeast China (128°6′E, 42°24′N). The area around the mountain is a temperate, continental climate, with a long-term cold winter and warm summer. The annual mean temperature is approximately 4.1^{o}C, and precipitation averages approximately 855 mm at the bottom of the mountain, with more than 80% of rainfall from May to August. The dark brown forest soil belongs to Andosols (Food and Agriculture Organization soil classification), and the depth of litters and A-horizons is approximately 3-5 cm and 10 cm, respectively. The main properties of the soils at the various depths and groundwater table levels were reported by Xu et al. (2007, 2009).

Effects of N addition on soil solution chemistry under forest stand

Twenty-eight individual plots with 3 m x 3 m each were selected on the flatness under the mixed forest stand. Aqueous solutions of N sources such as $(NH_4)_2SO_4$, NH_4Cl and KNO_3 were respectively sprayed on the ground within four individual plots in equal monthly doses at rates of 2.25 and 4.5 g per m^2 each year, during the growing season from June to October in 2006-2008, corresponding with 5.0 mm rainfall each; tap water was added only to the control. The N addition experiments at high and low doses started from July 2006 and June 2007, respectively. According to the depth of A-horizons and the distribution of tree roots in underlying soil, two sets of porous ceramic suction cups (3.1 cm in diameter and 7 cm in height) were installed at 15 cm and 60 cm depths, respectively, to collect soil solutions of organic layers and beyond root zones (Vandenbruwane *et al.* 2008). To eliminate the disturbance of soil, soil auger with a diameter of 3.3 cm was used to establish the holes down to 15 cm and 60 cm depths, respectively, and the suction cups connected to PVC tubes were fixed closely inside the holes. The pressure inside each tube within a week at water-filled pore space more than 70% or within 24 hours after heavy rainfall was brought to approximately -70 kP_a by a portable vacuum/pressure pump (Mityvac4010, Missouri, USA). Over the years from July 2006 to October 2008, soil solutions were sampled via the stopcock attached to each tube to avoid degassing, using 100-ml plastic syringes equipped with a stopcock, and the volume of soil solution was measured simultaneously. Considering initial effects of installing the suction cups, the early two collections were discarded. These samples were rapidly transported to the laboratory and were frozen prior to the analysis of dissolved organic C, total N, NH_4^+-N and NO_3^--N concentrations.

Measurement of wet atmospheric C and N depositions and soil net N mineralization

Monthly wet atmospheric C and N depositions under such forest stand were sampled at an irregular interval dependent on intensity of precipitation using self-made rainfall collections during the whole experimental period. Dynamics of net N mineralization fluxes of the soil at 15 cm depth under all the experimental plots were measured in 2006-2008 using in-situ resin-core incubation method (Han *et al.* 2009).

Measurement of concentrations of dissolved organic C, total N, NH_4^+-N and NO_3^--N

Concentrations of DOC and total N in soil solutions were measured using a TOC/TN-analyzer (Shimadzu TOC-V_{CSH}/TN, Kyoto, Japan). Both NH_4^+-N and NO_3^--N concentrations of solutions were measured colorimetrically via the nitroprusside and hydrazine-reduction methods, respectively (Kim, 1995). Concentrations of dissolved organic N (DON) were calculated as the differences between total N and mineral N (NH_4^+-N and NO_3^--N) concentrations of solutions.

Calculation and statistical analysis

Wet atmospheric C and N depositions, and fluxes of dissolved organic matter and inorganic N in the soil solution were calculated via multiplying corresponding volume mean concentrations of solution C and N forms by amount of water flux. Means and standard errors of soil solution chemistry for each sampling date were calculated. Correlation coefficients between the tested properties of soil solutions at 15 cm and 60 cm depths were calculated using SPSS software for Windows (version 13.0). The multivariate tests and paired-sample T tests were performed using SPSS software for Windows to compare the differences in concentrations and fluxes of DOC, DON, NH_4^+ and NO_3^- in soil solutions between treatment, sampling date and soil depth.

Results and discussion

Effect of N addition on concentrations of DOC, DON and inorganic N in soil solution

NH_4^+-N concentrations in the soil solutions at 15 cm and 60 cm depths under N-fertilized and non-fertilized plots were mostly below 0.2 µg N/ml, which were much smaller than NO_3^--N concentrations in soil solutions (Figure 1). The addition of N increased the accumulation of NH_4^+-N, NO_3^--N and DON in the soil solutions at both depths (Figure 1). Thus, the export of inorganic N and DON from such forest stand was significantly increased by addition of N.

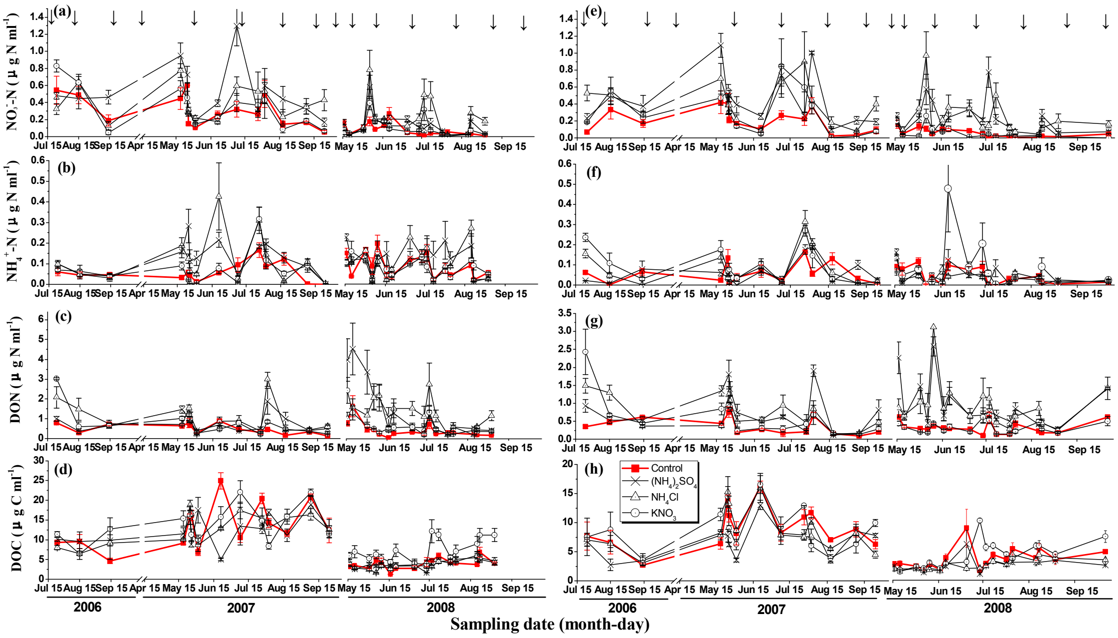

Figure 1. Dynamics of dissolved organic matter and inorganic N concentrations in forest soil solutions in 2006-2008 upon addition of N at a rate of 4.5 g /m^2 each year. a-d: soil solution at 15 cm depth; e-h: soil solution at 60 cm depth. Arrows indicate date of N addition.

The DOC concentrations in soil solutions at 15 cm depth (1.2 to 27.8 µg C/ml) throughout the period of the experiment were significantly larger than those at 60 cm depth (1.1-16.5 µg C/ml) (Figure 1d,h). The addition of N sources such as $(NH_4)_2SO_4$ and NH_4Cl tended to decrease DOC concentrations in the soil solutions at 60 cm depth (Figure 1h) and showed a small change in concentrations at 15 cm depth (Figure 1d). DOC concentrations in the soil solutions decreased with increasing soil depth under all the experimental plots to a greater degree than did DON, especially under N-fertilized plots. This phenomenon may decrease rates of DOC to DON concentrations in soil solutions at both depths upon N addition. Our observations indicated that the mechanisms for DOC dynamics under N-fertilized forest stands on a plot-scale differed from those for DON.

Effect of N addition on fluxes of DOC, DON and inorganic N in soil solution

Monthly DOC fluxes in soil solutions at 15 cm depth (0.2 to 25.0 g C/m^2) throughout the period of the experiment were significantly larger than those at 60 cm depth (0.1-16.3 g C/m^2). The addition of N sources such as $(NH_4)_2SO_4$ and NH_4Cl tended to decrease monthly DOC fluxes in soil solutions at 60 cm depth and showed a small change in fluxes at 15 cm depth. Probably, there was high amount of DOC retained in underlying mineral soils (15-60 cm) under N-fertilized forest plots. The addition of N increased fluxes of NH_4^+-N and NO_3^--N and DON in the soil solutions at both depths and tended to decrease the proportion of DON in total N fluxes.

Effect of N addition on special UV absorbance of forest soil solution

There was a relatively small special UV absorbance of forest soil solution at 60 cm depth under all experimental plots in 2007-2008 compared to the solution at 15 cm depth (Figure 2). This indicated that DOM leached from deep soil layers is characterized by high decomposability. The addition of N tended to decrease special UV absorbance of soil solutions at 15 cm and 60 cm depths, especially at the latter. Probably, N inputs to forest floors can affect the stability of DOM in soil solution.

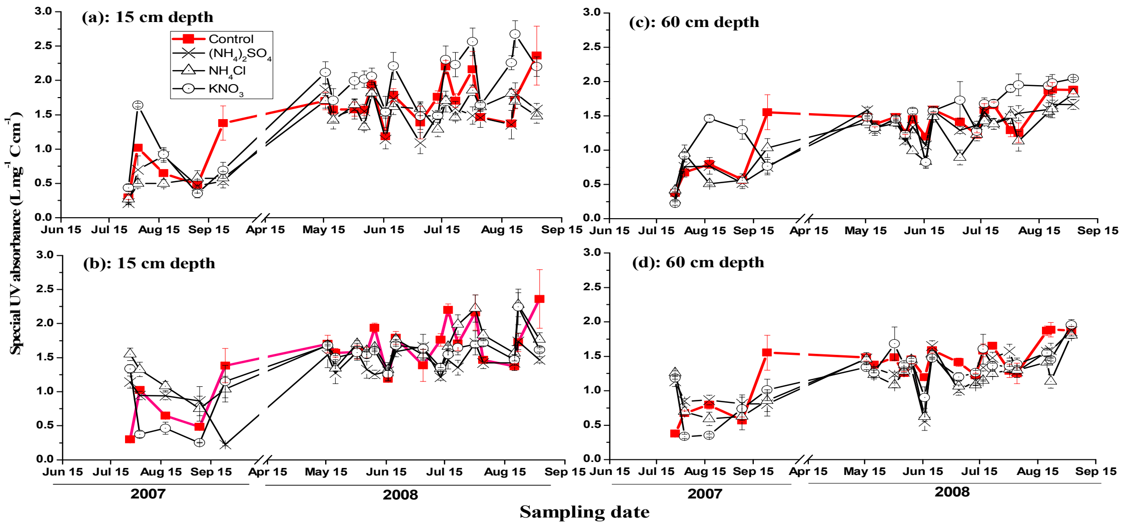

Figure 2. Dynamics of special UV absorbance of soil solutions at 15 cm and 60 cm depth in 2007-2008. a and c: N addition at a rate of 4.5 g /m^2 each year; b and d: N addition at a rate of 2.25 g /m^2 each year.

Contribution of N inputs and soil net N mineralization to N fluxes in soil solution

In combination with previous published studies, our data can be used to assess the contribution of N inputs and soil net N mineralization to N fluxes in soil solution under forest ecosystems.

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