Environmental Pollution 159 (2011) 363–367

Contents lists available at ScienceDirect

Environmental Pollution

journal homepage: www.elsevier.com/locate/envpol

Nitrogen isotope variations in camphor (*Cinnamomum Camphora*) leaves of different ages in upper and lower canopies as an indicator of atmospheric nitrogen sources

Hua-Yun Xiao*, Liang-Hong Wu, Ren-Guo Zhu, Yan-Li Wang, Cong-Qiang Liu

State Key Laboratory of Environmental Geochemistry, Institute of Geochemistry, Chinese Academy of Sciences, No. 46, Guanshui Road, Guiyang 550002, China

Nitrogen isotope in camphor leaves indicating atmospheric nitrogen sources.

ARTICLE INFO

Article history:
Received 7 September 2010
Received in revised form
8 November 2010
Accepted 11 November 2010

Keywords:
$\delta^{15}N$
Atmospheric nitrogen
Soil nitrogen
Age
Canopy
Leaf
Camphor

ABSTRACT

Nitrogen isotopic composition of new, middle-aged and old camphor leaves in upper and lower canopies has been determined in a living area, near a motorway and near an industrial area (Jiangan Chemical Fertilizer Plant). We found that at sites near roads, more positive $\delta^{15}N$ values were observed in the camphor leaves, especially in old leaves of upper canopies, and $\Delta\delta^{15}N = \delta^{15}N_{upper} - \delta^{15}N_{lower} > 0$, while those near the industrial area had more negative $\delta^{15}N$ values and $\Delta\delta^{15}N < 0$. These could be explained by two isotopically different atmospheric N sources: greater uptake from isotopically heavy pools of atmospheric NO_x by old leaves in upper canopies at sites adjacent to roads, and greater uptake of ^{15}N-depleted NH_y in atmospheric deposition by leaves at sites near the industrial area. This study presents novel evidence that ^{15}N natural abundance of camphor leaves can be used as a robust indicator of atmospheric N sources.

1. Introduction

Due to the high population density and intensive industries, anthropogenic N emissions from cities have inevitably become prominent sources for elevated regional N pollution, which influences the air quality of cities and poses threats to surrounding ecosystems (Jung et al., 1997; Krupa, 2003). Therefore, it is important and urgent to identify their sources.

Identification of atmospheric N sources by physical methods is a complex task because a wide range of N compounds occurred in the gas phase, in aerosols and in precipitation, which has made it very difficult and expensive to undertake long-term instrumental monitoring, especially in remote areas (Solga et al., 2005; Pitcairn et al., 2006). Owing to the scarcity of physical monitoring, information of atmospheric N deposition and major atmospheric N sources is still lacking or sparse in many regions. Therefore, a less costly alternative to physical measurement of N deposition is needed. One potentially reliable approach is to use plant as a bio-monitor, which has been believed an easy and low-cost way to specifically shed light on integrative and long-term N deposition (Jung et al., 1997; Pitcairn et al., 2001). Mosses, lacking a cuticle and absorbing soluble and insoluble mineral nutrients only from ambient air and precipitation, have often been used and been shown to be outstanding bio-indicators in a wide range of air pollution studies (Hutchinson and Scott, 1988; Nygaard and Abrahamsen, 1991; Hicks et al., 2000; Pitcairn et al., 2001; Liu et al., 2007; Xiao et al., 2010a,b). However, mosses will not grow where there is heavy pollution.

Isotopic data for atmospherically derived N are increasingly used in identifying the sources of atmospheric inputs to a wide variety of soil and plant environments (Evans and Ehleringer, 1993; Durka et al., 1994). Moss $\delta^{15}N$ was also believed as a reliable tool to distinguish N emission sources (Liu et al., 2008) because isotopic fractionation during N uptake has been assumed to be absent or very low for mosses (Bragazza et al., 2005). Differently, $\delta^{15}N$ of soil-grown plants provides a synthesis of the $\delta^{15}N$ of the N sources (soil N, atmospheric N, and so on), N forms taken up (organic N, NH_4^+, and NO_3^-), fractionation occurs during and after N uptake by plants and by different mycorrhizal associations, differential translocation of nitrogen isotopes within the tree (Nadelhoffer et al., 1996; Högberg, 1997). So, researches on using nitrogen isotopic signature of soil-grown plants as an indicator of atmospheric N sources are very limited. Some studies available are concentrated on coniferous trees. For example, Jung et al. (1997) studied atmospheric N input in an industrially

* Corresponding author.
E-mail address: xiaohuayun@vip.skleg.cn (H.-Y. Xiao).

0269-7491/$ – see front matter © 2010 Elsevier Ltd. All rights reserved.
doi:10.1016/j.envpol.2010.11.011

polluted area in Germany based on the $\delta^{15}N$ values of pine needles. Ammann et al. (1999) found that ^{15}N abundance in Norway spruce needles provided an indication of traffic-derived NO_2.

In this study, camphor tree (*Cinnamomum Camphora*), a broad-leaved tree that widespread distributes in southern China, is selected to show whether their foliage $\delta^{15}N$ can be used as a effective bio-monitor of atmospheric N sources. Specific questions addressed are (1) How does the foliage $\delta^{15}N$ vary with ages and what influences it? (2) Are there any differences in $\delta^{15}N$ values between soil N and camphor leaves and between upper and lower canopy? (3) What are the main sources of atmospheric N deposition based on the foliage $\delta^{15}N$?

2. Materials and methods

2.1. Sample collection and treatment

We selected three areas (living area, motorway and industrial area) in Nanchang city for leaf sampling because they differ to some extent from each other as regards isotopic signatures of atmospheric nitrogen sources. Two camphor trees (*Cinnamomum Camphora*) were chosen in a living community: one is located near a road (TH2) and the other is far away from any roads (TH1). At sites with a distance of 10 m (MW1) and 160 m (MW2), respectively, from a motorway (Changzhang Motorway), two camphor trees were selected for comparisons. The traffic flow on the motorway is about 5,000 vehicles per day in the year 2009. Three camphor trees at 20 m (JA1), 100 m (JA2) and 160 m (JA3) from the Jiangan Chemical Fertilizer Plant (JCFP) were chosen. In the JCFP, about 110,000 tons ammonia fertilizer and 160,000 tons urea were produced per year. All the camphor trees chosen for leaf sampling are about at the age of ten and about five meters in height in order to reduce the influences from physiological differences between these sampling trees and different soil depths that tree roots reached. And camphor trees possibly disturbed by other N sources were avoided.

For all the selected camphor trees, about 10 g of new leaves ($<10\ cm^2$ surface area), middle-aged leaves (10 to 30 cm^2 surface area) and old leaves ($>30\ cm^2$ surface area) was collected in 2009 from both the upper and lower canopies. Only those camphor leaves growing in 2009 were taken from the selected trees. We collected 5 to 10 samples for each kind of leaves. The leaf materials were washed and dried for 24 h at 80 °C. Then they were ground into a very fine power using a mortar and pestle prior to chemical analyses. At each site and just at the same time when camphor leaves were collected, rhizospheric soil was sampled by taking about 100 g of the topmost 10 cm soil layer and bulked. Leaf litters and roots were removed from the soil samples. Soil samples were dried for 24 h at 80 °C and homogenized using a mortar and pestle.

2.2. Chemical analyses

Foliage N concentrations were measured by an elemental analyzer (Model PE-2400 II, USA) with an analytical precision of 1%. Calibration of the instrument with cystine standard (N141-0324, provided by Perkin Elmer) was carried out. Accuracy and recovery of N concentrations were checked by analysing a sample of this standard material after each set of eight camphor leaf analyses.

For each sample, nitrogen isotope analyses were performed by burning 10 mg leaf materials. After high purification with liquid N, nitrogen isotope ratios of camphor leaves and soil samples were determined on a Finnigan MAT 252 mass spectrometer. The $\delta^{15}N$ values are given in permil units (‰), in reference to air international standard. Analysis of potassium nitrate standard (MOR2386-01) provided by Shoko Co., Ltd., Tokyo, Japan (+1.9‰) gave a mean (±SD) $\delta^{15}N_{air}$ value of 1.9 ± 0.2‰ ($n = 5$). High purity N_2 reference gas was run with each analysis. Three to five replicate measurements per sample were carried out, and values were presented as the average of these measurements. The analytical precision (±SD, $n = 5$) for $\delta^{15}N$ was ±0.2‰.

2.3. Statistical analysis

Statistical analysis was conducted by SPSS 11.5, and graphs were mainly created with SigmaPlot 2000 software (both SPSS Science, Chicago, USA). Differences in concentrations and isotopic values between leaf ages or sampling sites were tested for significance by using a one-way analysis of variance (ANOVA) procedure, and least significant differences (LSD) were used by a Tukey-HSD test to compare significant differences (at the 95% confidence level). Differences were considered significant at $P < 0.05$.

3. Results

3.1. Nitrogen concentrations in camphor leaves and rhizospheric soils

The nitrogen concentrations of camphor leaves were in the range of 1.29% to 5.54% (Table 1) and varied considerately with leaf ages. As expected, the highest concentrations were found in the new leaves and the lowest in the old leaves. But for leaves of the same age, there are no significant differences in N concentrations between camphor trees growing at different sites ($p > 0.05$).

Total N concentrations of rhizospheric soils varied from 0.22% to 0.46% (Table 1), with the average of 0.35 ± 0.09%. No significant differences in soil N concentrations were observed between sampling sites ($p > 0.05$).

3.2. Nitrogen isotope ratios in camphor leaves and rhizospheric soils

The $\delta^{15}N$ values of camphor leaves in Nanchang City were found to vary depending on their specific location by a factor of up to one order of magnitude (−11.8‰ to +2.5‰) (Table 1). The camphor leaves with positive $\delta^{15}N$ values were observed near roads (TH2 and MW1) and negative $\delta^{15}N$ values near the industrial area (JA1, JA2 and JA3) and at the site 160 m from the motorway. And for the three camphor trees near the industrial area, more negative $\delta^{15}N$ values occurred in camphor leaves on sites at smaller distances from the Jiangan Chemical Fertilizer Plant (JCFP).

Although the range of foliage $\delta^{15}N$ values was very large, rhizospheric soil $\delta^{15}N$ values within these sampling sites were limited from +0.6‰ to +5.1‰ (Table 1). For all the sampling sites, the foliage $\delta^{15}N$ values were significantly lower than the soil $\delta^{15}N$ values ($p < 0.001$).

3.3. Nitrogen isotope variations of camphor leaves of different ages

From new, to middle-aged then to old camphor leaves, an increase in $\delta^{15}N$ values was observed at sites in the living area (Fig. 1a) and adjacent to the motorway (Fig. 1b), while an opposite trend (i.e. decrease with leaf ages) was found at sites near the industrial area (Fig. 1c). The trend of the latter was similar to that in spruce needles in Sweden (Gebauer and Schulze, 1991).

Both the two camphor trees in the living area showed positive $\delta^{15}N$ values in old leaves and negative values in new leaves. The age

Table 1
The range and average of N concentrations and $\delta^{15}N$ values in camphor leaves and rhizospheric soils.

			Living area		Motorway		Industrial area (JCFP)		
			TH1	TH2	MW1	MW2	JA2	JA1	JA3
Camphor leaves	N conc. (%)	Range	1.82~5.25	1.29~5.54	2.63~3.41	2.40~2.91	2.08~5.36	1.74~5.34	1.55~5.16
		Average	3.36	3.05	2.99	2.73	3.31	3.05	3.10
	$\delta^{15}N$ (‰)	Range	−0.8~+0.3	−1.8~+2.5	−1.0~+2.0	−3.9~−2.8	−11.8~−7.8	−11.1~−8.0	−5.9~−1.5
		Average	−0.1	+0.3	+0.1	−3.3	−9.	9−9.8	−4.3
Rhizospheric soils	N conc. (%)		0.26	0.22	0.41	0.44	0.46	0.29	0.33
	$\delta^{15}N$ (‰)		+0.6	+3.2	+3.0	+3.9	+3.1	+5.1	+3.3

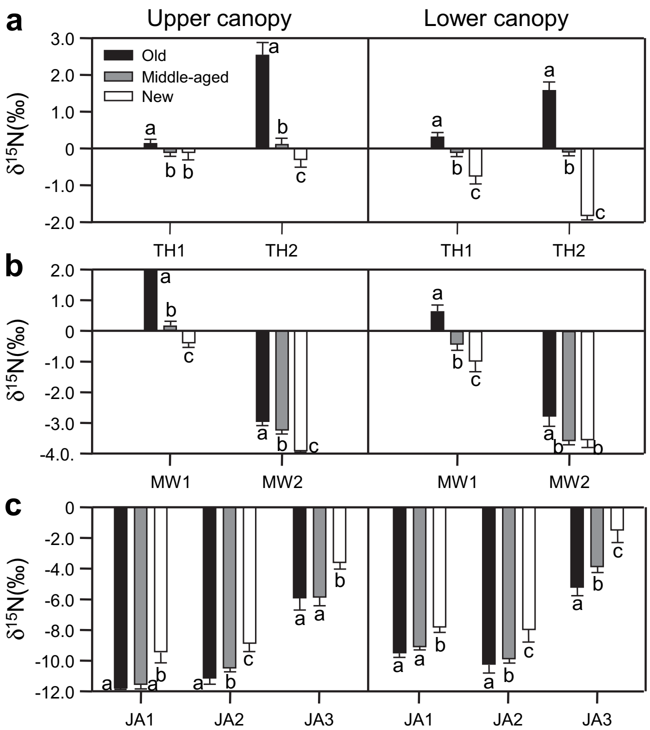

Fig. 1. The $\delta^{15}N$ values of old, middle-aged and new camphor leaves in the upper (left column) and lower canopies (right column) at sites in a living area (a), near a motorway (b) and near an industrial area (c) in Nanchang City. Different uppercase letters denote means found to be statistically different by the Tukey-HSD test with LSD between leaf ages. TH2 and MW1 are located near roads and JA1 nearer the chemical fertilizer plant (JCFP).

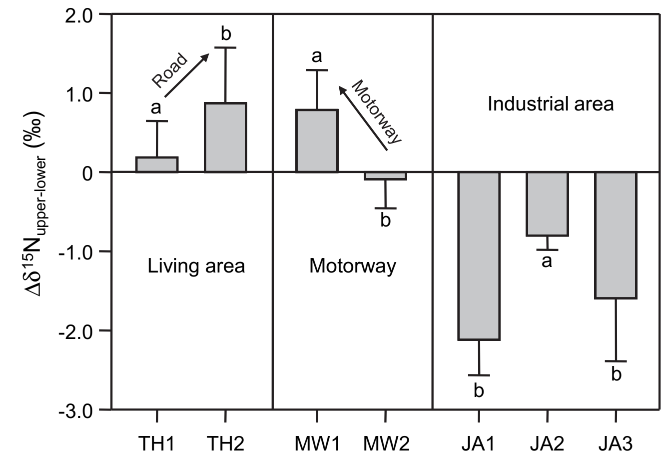

Fig. 2. The $\Delta\delta^{15}N$ values ($\Delta\delta^{15}N = \delta^{15}N_{upper} - \delta^{15}N_{lower}$) of camphor leaves between upper and lower canopies in Nanchang City. Different uppercase letters denote means found to be statistically different by the Tukey-HSD test with LSD between sampling sites.

pattern of foliage $\delta^{15}N$ values at site TH2 (near a road) is similar to that at site MW1, which lies only 10 m from the motorway. The foliage $\delta^{15}N$ values of the two camphor trees at sites MW1 and MW2 (160 m from the motorway) are significantly different ($p < 0.001$).

The differences of $\delta^{15}N$ values between old and new leaves are large (≈ 3‰) at sites near roads (TH2 and MW1) whereas small (<1‰) at sites TH1 and MW2 (Fig. 1a, b). The difference in foliage $\delta^{15}N$ values for all the three camphor trees growing near the industrial area (JCFP) is about 2‰ between old and new leaves (Fig. 1c).

3.4. Foliage nitrogen isotopes of upper and lower canopies

Foliage $\delta^{15}N$ values of upper canopies ranged from -11.7‰ to $+2.5$‰, whereas those of lower canopies were in the range of -10.2‰ to $+1.6$‰. At sites near roads (TH2 and MW1), camphor leaves in upper canopies had higher ^{15}N contents than those in lower canopies ($\Delta\delta^{15}N = \delta^{15}N_{upper} - \delta^{15}N_{lower} > 0$) while near the industrial area (JCFP), $\Delta\delta^{15}N$ was found < 0 (Fig. 2).

4. Discussion

4.1. Nitrogen isotope variations during soil and plant processes

In this study, camphor foliage $\delta^{15}N$ values were found lower than those of rhizospheric soils at all sampling sites, especially in new leaves (Table 1). This pattern is consistent with many other studies (Gebauer and Schulze, 1991; Garten, 1993; Garten and Van Miegroet, 1994; Miller and Bowman, 2002). Usually, soil-grown plant $\delta^{15}N$ is determined by (1) the $\delta^{15}N$ values of atmospheric N, (2) the $\delta^{15}N$ values of soil N, (3) the N forms in soils, (4) isotopic fractionations during soil N transformation, (5) isotopic fractionations during plant processes (e.g. nitrogen uptake, basipetal translocation), etc (e.g. Garten, 1993; Nadelhoffer and Fry, 1994; Jung et al., 1997; Högberg, 1997). Although the isotope ratios in camphor leaves may be affected by different soil depths that the camphor roots can reach, influences from soil-depth differences between these sampling trees are small. This is because the trees we selected are similar in heights and so they usually have roots of similar lengths. Because the new camphor leaves have a small surface area of <10 cm^2 and are exposed to atmospheric N for very short time (<1–2 days), the obvious difference in $\delta^{15}N$ values between soil N and new camphor leaves may be the result of a number of factors, which include the above 2 to 5 aspects. Some of these aspects have been discussed as follows.

N transformation is the most important process in soils that can affect the $\delta^{15}N$ values of soil N. If ammonium is nitrified in soils, isotopic discrimination can lead to the production of soil nitrate with lower $\delta^{15}N$ values than residual ammonium, particularly if ammonium pools are not totally nitrified (Nadelhoffer and Fry, 1994). So, nitrification in soils can lead to soil-grown plants strongly ^{15}N-depleted if plants derive most of their N from nitrate (Shearer et al., 1974). It is possible that the camphor leaves (especially new leaves) in this study may rely on NO_3^- with low $\delta^{15}N$ values for their N nutrition to a greater degree than NH_4^+ and organic N. Another important factor that may lead to ^{15}N-depleted plants is in association with mycorrhizal fungi in rhizospheric soils. Several studies had shown that mycorrhizal fungi deliver isotopically depleted nitrogen to plants (e.g., Michelsen et al., 1996, 1998).

The assumptions that isotope ratio of source nitrogen is preserved during nitrogen absorption, assimilation and translocation, and that the $\delta^{15}N$ values of leaf tissues reflect that of the nitrogen source in the rhizospheric soils are invalid because plant processes such as different nitrogen uptake mechanisms, different pathways of assimilation, and recycling of nitrogen in the plant can discriminate against ^{15}N. Available data suggest that under natural conditions, nitrogen isotope fractionation accompanying uptake of nitrogen by the roots is relatively small or zero (Karamanos and Rennie, 1980; Mariotti et al., 1982). Isotopic discrimination was observed, however, when plant nitrogen demand was relatively low, compared with the nitrogen available in solution (Evans, 2001). According to this information, isotopic discrimination during nitrogen uptake will occur in most urban ecosystems, where external concentrations are high and nitrogen supply will exceed plant demand. Additionally, after N enters into plants, nitrate reductase and amino acid biosynthesis both fractionate against ^{15}N; observed discrimination by nitrate reductase and amino acid biosynthesis is 15‰ and 17‰, respectively (Handley and Raven, 1992; Yoneyama et al., 1993). This fractionation event causes the organic nitrogen product ^{15}N-depleted compared with the original soil N source. Isotopic fractionation associated with basipetal translocation prior to autumn leaf fall could cause the nitrogen stored in trees to become isotopically more negative over time. But a contrary issue proposed by Garten (1993) shows that the isotope fractionation accompanying the analogous translocation of nitrogen from senescing foliage before leaf abscission is small, with residual nitrogen typically enriched by ≤0.5‰. Therefore, he believed that isotopic signature of foliage nitrogen was for the most part preserved throughout the growing season.

Through the above discussion, we interpret the more negative $\delta^{15}N$ values in camphor leaves than in rhizospheric soils as reflecting isotopic fractionation during soil and plant processes, most of which reactions discriminate against ^{15}N. Even so, we can derive some information about atmospheric N sources from the isotopic comparisons between leaves of different ages at different sampling sites and between upper and lower canopies.

4.2. Foliage isotope variations related to atmospheric N sources

Usually, epiphyte such as mosses is believed to be the best bio-monitor of atmospheric N deposition because they derive N only from the atmosphere. For instance, Pearson et al. (2000) reported that mosses growing near roadsides in London had an average $\delta^{15}N$ value of +3.7‰, significantly different to the value (−7.8‰) reported for mosses near agricultural sources. Although $\delta^{15}N$ of soil-grown plants is influenced by many factors such as soil N and isotopic fractionation occurs during and after N uptake by plant, and differential translocation of nitrogen isotopes within the tree (Högberg, 1997), some previous studies on coniferous trees (Jung et al., 1997; Ammann et al., 1999) showed that atmospheric N input could be recorded by the $\delta^{15}N$ values of soil-grown tree leaves. Saurer et al. (2004) found that current-year needles had an average value of +1.3‰ close to the motorway, decreasing to −2.6‰ at the intermediate site and to −4.4‰ at 1000 m distance. Pine needles in heavily polluted areas of Germany varied by one order of magnitude depending on the plants proximity to emission sources (Jung et al., 1997). Vegetation from the heavily polluted industrial area of Cubatão (SE Brazil) was strongly ^{15}N-depleted compared to plants at remote sites (Stewart et al., 2002). Ammann et al. (1999) believed that leaves of plants growing in close proximity to an emitter reflected the $\delta^{15}N$ values of the atmospheric pollutant N.

As camphor trees do not fix atmospheric N_2, the external sources of nitrogen at these studied sites are either direct canopy uptake of N (NO_x or NH_y) in the atmosphere or N uptake from the rhizospheric soil by the roots. A number of studies have measured the isotopic composition of atmospheric NO_x and NH_y and have shown different $\delta^{15}N$ values between them (e.g., Heaton, 1986; Garten, 1992; Xiao and Liu, 2002). For example, the $\delta^{15}N$ values of atmospheric NH_y at sites near NH_y emitters of petrochemical and fertilizer factory were observed at about -40.82‰ (Stewart et al., 2002), typically isotopically lighter than those of atmospheric NO_x (+5.7‰) measured on a highway (Ammann et al., 1999). According to these, we suspected that atmospheric NH_y near the industrial area (JCFP) may have a low ^{15}N abundance whereas the $\delta^{15}N$ values of atmospheric NO_x at sites adjacent to roads (TH2 and MW1) will be high. Therefore, while greater uptake of atmospheric NO_x could lead to relatively higher foliage $\delta^{15}N$ values, reliance upon atmospheric NH_y to meet N uptake requirements could lead to lower foliage $\delta^{15}N$ values, suggesting that ^{15}N natural abundance of camphor leaves can be used as a reliable indicator of atmospheric N sources. This can be further proofed by the following three aspects.

Firstly, more positive $\delta^{15}N$ values were observed in the camphor leaves at sites near roads (TH2 and MW1) whereas those leaves near the industrial area and at the site 160 m from the motorway had more negative $\delta^{15}N$ values (Fig. 1). Isotopic fractionation during soil and plant processes as discussed above cannot result in the site-specific variations since the $\delta^{15}N$ values of soil N are somewhat similar at different sites. On one hand, although the ^{15}N abundances of N from soils and NO_x from the atmosphere are not significantly different in this study, the positive isotopic values of soil N turned to be negative after reaching the leaves (Table 1). Because tree leaves contact closely atmospheric N, the isotope fractionation during uptake of atmospheric N by leaves may be smaller relative to the uptake of soil N by roots. Therefore, the only possible event that causes the more positive foliage $\delta^{15}N$ values at sites near roads (TH2 and MW1) is greater uptake of ^{15}N-enriched atmospheric NO_x sources. On the other hand, assimilation of isotopically light atmospheric N results in ^{15}N-depletion of the camphor leaves near the industrial area and at the site 160 m from the motorway. It is clear that the strongly ^{15}N-depleted camphor leaves appear directly associated with the industrial activities of JCFP. Vitousek et al. (1989) suggested that inputs of ^{15}N-depleted nitrogen from precipitation would cause the strongly negative $\delta^{15}N$ values in non-nitrogen fixing plants.

Secondly, with time of exposure (i.e. leaf ages) the ^{15}N abundances of the camphor leaves should increasingly be influenced by the $\delta^{15}N$ values of the atmospherically deposited N. Although N would move out from old leaves prior to leaf abscission, the isotope fractionation accompanying the analogous translocation of nitrogen is small (Garten, 1993). Thus, the foliage $\delta^{15}N$ values would increase with increasing leaf age if N uptake from atmospheric deposition were enriched in ^{15}N while if they are exposed to isotopically light atmospheric N, a decrease with leaf ages would be expected. The isotopic decrease with leaf ages was also reported by Gebauer and Schulze (1991) and Gebauer et al. (1994). The age-dependant trend of foliage $\delta^{15}N$ values confirming the above assumption was indeed found at all these sites in this study (Fig. 1).

Thirdly, camphor leaves in the upper canopy usually derive more N from the atmosphere (in particulate and rain) than those in the lower canopy. Thus, if atmospheric N was ^{15}N-riched, more positive foliage $\delta^{15}N$ values would be found in the upper canopy ($\Delta\delta^{15}N > 0$). On the contrary, camphor leaves in the upper canopy would be more ^{15}N-depleted ($\Delta\delta^{15}N < 0$) if isotopically light atmospheric N was absorbed (e.g. near the industrial area). The positive and negative $\Delta\delta^{15}N$ values occurred at sites near roads (TH2 and MW1) and at sites near the JCFP (Fig. 2) were suggestive of two isotopically different atmospheric N sources, respectively.

5. Conclusions

Three important results could be drawn from the comparisons of $\delta^{15}N$ values between leaves of different ages at different sampling sites, and between upper and lower canopies.

(1) More positive $\delta^{15}N$ values were observed in the camphor leaves at sites near roads (TH2 and MW1) while those near the industrial area (JCFP) and at the site 160 m from the motorway had more negative $\delta^{15}N$ values.
(2) With time of exposure (i.e. leaf ages), the ^{15}N abundance of the camphor leaves at sites near roads increased whereas those at sites near the industrial area (JCFP) decreased.
(3) More positive foliage $\delta^{15}N$ values were found in the upper canopies than in the lower canopies at sites near roads whereas at sites near the industrial area (JCFP), the upper canopies showed more negative foliage $\delta^{15}N$ value.

Although more negative $\delta^{15}N$ values in camphor leaves than in rhizospheric soils reflected isotopic fractionation during soil and plant processes, the above results indicated that there appeared to be two isotopically different atmospheric N sources affecting the ^{15}N abundance of camphor leaves. They are (1) greater uptake from isotopically heavy pools of atmospheric NO_x by plants at sites adjacent to roads, and (2) greater uptake of isotopically light NH_y in atmospheric deposition by plants at sites near the industrial area (JCFP). Therefore, this study presents novel evidence that ^{15}N natural abundance of camphor leaves can be used as a robust indicator of atmospheric N sources.

Acknowledgments

This study work was kindly supported by the National Natural Science Foundation of China through grants 41073016 and 40573006 (H.Y. Xiao) and 40721002 (C.Q. Liu), and by West Light Foundation of The Chinese Academy of Sciences (H.Y. Xiao).

References

Ammann, M., Siegwolf, R., Pichlmayer, F., Suter, M., Saurer, M., Brunold, C., 1999. Estimating the uptake of traffic-derived NO_2 from ^{15}N abundance in Norway spruce needles. Oecologia 118, 124–131.

Bragazza, L., Limpens, J., Gerdol, R., Grosvernier, P., Hájek, M., Hájek, T., Hajkova, P., Hansen, I., Iacumin, P., Kutnar, L., Rydin, H., Tahvanainen, T., 2005. Nitrogen concentration and $\delta^{15}N$ signature of o *mbrotrophic Sphagnum* mosses at different N deposition levels in Europe. Global Change Biology 11, 106–114.

Durka, W., Schulze, E.D., Gebauer, G., Voerkellust, S., 1994. Effects of forest decline on uptake and leaching of deposited nitrate determined from ^{15}N and ^{18}O measurements. Nature 372, 765–767.

Evans, R.D., 2001. Physiological mechanisms influencing plant nitrogen isotope composition. Trends in Plant Science 6, 121–126.

Evans, R.D., Ehleringer, J.R., 1993. A break in the nitrogen cycle in arid lands? Evidence from $d^{15}N$ of soils. Oecologia 94, 313–317.

Garten, C.T., 1992. Nitrogen isotope composition of ammonium and nitrate in bulk precipitation and forest throughfall. International Journal of Analytical Chemistry 47, 33–45.

Garten, C.T., 1993. Variation in foliage ^{15}N abundance and the availability of soil nitrogen on Walker Branch watershed. Ecology 74, 2098–2113.

Garten, C.T., Van Miegroet, H., 1994. Relationships between soil nitrogen dynamics and natural ^{15}N abundance in plant foliage from Great Smoky Mountains National Park. Canadian Journal of Forest Research 24, 1636–1645.

Gebauer, G., Giesemann, A., Schulze, E.D., Jäger, H.J., 1994. Isotope ratios and concentrations of sulfur and nitrogen in needles and soils of *Picea abies* stands as influenced by atmospheric deposition of sulfur and nitrogen compounds. Plant Soil 164, 267–281.

Gebauer, G., Schulze, E.D., 1991. Carbon and nitrogen isotope ratios in different compartments of a healthy and a declining *Picea abies* forest in the Fichtelgebirge, NE Bavaria. Oecologia 87, 198–207.

Handley, L.L., Raven, J.A., 1992. The use of natural abundance of nitrogen isotopes in plant physiology and ecology. Plant, Cell & Environment 15, 965–985.

Heaton, T.H.E., 1986. Isotopic studies of nitrogen pollution in the hydrosphere and atmosphere: a review. Chemical Geology 59, 87–102.

Hicks, W.K., Leith, I.D., Woodin, S.J., Fowler, D., 2000. Can the foliage nitrogen concentration of upland vegetation be used for predicting atmospheric nitrogen deposition? Evidence from field surveys. Environmental Pollution 107, 367–376.

Högberg, P., 1997. ^{15}N natural abundance in soil-plant systems. New Phytologist 137, 179–203.

Hutchinson, T.C., Scott, M.G., 1988. The response of the feather moss, *Pleurozium schreberi*, to 5 years of simulated acid precipitation in the Canadian boreal forest. Canadian Journal of Botany 66, 82–88.

Jung, K., Gebauer, G., Gehre, M., Hofmann, D., Weissflog, L., Schuurmann, G., 1997. Anthropogenic impacts on natural nitrogen isotope variations in *Pinus sylvestris* stands in an industrially polluted area. Environmental Pollution 97, 175–181.

Karamanos, R.E., Rennie, D.A., 1980. Variations in natural nitrogen-15 abundance as an aid in tracing fertilizer nitrogen transformations. Soil Science Society of America Journal 44, 57–62.

Krupa, S.V., 2003. Effects of atmospheric ammonia (NH_3) on terrestrial vegetation: a review. Environmental Pollution 124, 179–221.

Liu, X.Y., Xiao, H.Y., Liu, C.Q., Li, Y.Y., 2007. $\delta^{13}C$ and $\delta^{15}N$ of moss (*Haplocladium microphyllum* (Hedw.) Broth) for indicating environment variations and canopy retention on atmospheric nitrogen deposition. Atmospheric Environment 41, 4897–4907.

Liu, X.Y., Xiao, H.Y., Liu, C.Q., Li, Y.Y., Xiao, H.W., 2008. Stable carbon and nitrogen isotopes of the moss *Haplocladium microphyllum* in an urban and a background area (SW China): the role of environmental conditions and atmospheric nitrogen deposition. Atmospheric Environment 42, 5413–5423.

Mariotti, A., Mariotti, F., Champigny, M.L., Amerger, N., Moyse, A., 1982. Nitrogen isotope fractionation associated with nitrate reductase activity and uptake of NO_3^- by Pearl Millet. Plant Physiology 69, 880–884.

Michelsen, A., Quarmby, C., Sleep, D., Jonasson, S., 1998. Vascular plant ^{15}N natural abundance in health and forest tundra ecosystems is closely correlated with presence and type of mycorrhizal fungi in roots. Oecologia 115, 406–418.

Michelsen, A., Schmidt, I.K., Jonasson, S., Quarmby, C., Sleep, D., 1996. Leaf ^{15}N abundance of subarctic plants provides field evidence that ericoid, ectomycorrhizal and non- and arbuscular mycorrhizal species access different sources of soil nitrogen. Oecologia 105, 53–63.

Miller, A.E., Bowman, W.D., 2002. Variation in nitrogen-15 natural abundance and nitrogen uptake traits among co-occurring alpine species. Oecologia 130, 609–616.

Nadelhoffer, K.J., Fry, B., 1994. Nitrogen isotopes in forest ecosystems. In: Lajtha, K., Michener, R.H. (Eds.), Stable Isotopes in Ecology and Environmental Science. Blackwell, Oxford, pp. 22–44.

Nadelhoffer, K., Shaver, G., Fry, B., Giblin, A., Johnson, L., McKane, R., 1996. ^{15}N natural abundances and N use by tundra plants. Oecologia 107, 386–394.

Nygaard, P.H., Abrahamsen, G., 1991. Effects of long-term artificial acidification on the ground vegetation and soil in 100 year-old stand of Scots pine (*Pinus sylvetris*). Plant and Soil 131, 151–160.

Pearson, J., Wells, D.M., Seller, K.J., Bennett, A., Soares, A., Woodall, J., Ingrouille, M.J., 2000. Traffic exposure increases natural 15N and heavy metal concentrations in mosses. New Phytologist 147, 317–326.

Pitcairn, C.E.R., Fowler, D., Leith, I.D., Sheppard, L., Tang, S., Sutton, M., Famulari, D., 2006. Diagnostic indicators of elevated nitrogen deposition. Environmental Pollution 144, 941–950.

Pitcairn, C.E.R., Leith, I.D., Sutton, M.A., Fowler, D., Hargreaves, K.J., Moghaddam, M., Kennedy, V.H., Grannat, L., 2001. Foliage nitrogen as an indicator of nitrogen deposition and critical loads exceedance on a European scale. Water, Air, Soil Pollution 130, 1037–1042.

Saurer, M., Cherubini, P., Ammann, M., De Cinti, B., Siegwolf, R., 2004. First detection of nitrogen from NO_x in tree rings: a $^{15}N/^{14}N$ study near a motorway. Atmospheric Environment 38, 2779–2787.

Shearer, G., Duffy, J., Kohl, D.H., Commoner, B., 1974. A steady state model of isotopic fractionation accompanying nitrogen transformation in soil. Soil Science Society of America Proceedings 38, 315–322.

Solga, A., Burkhardt, J., Zechmeister, H.G., Frahm, J.P., 2005. Nitrogen content, ^{15}N natural abundance and biomass of the two pleurocarpous mosses *Pleurozium schreberi* (Brid.) Mitt and *Scleropodium purum* (Hedw.) Limpr. in relation to atmospheric nitrogen deposition. Environmental Pollution 134, 465–473.

Stewart, G.R., Aidar, M.P.M., Joly, C.A., Schmidt, S., 2002. Impact of point source pollution on nitrogen isotope signatures ($\delta^{15}N$) of vegetation in SE Brazil. Oecologia 131, 468–472.

Vitousek, P.M., Shearer, G., Kohl, D.H., 1989. Foliage ^{15}N natural abundance in Hawaiian rainforest: patterns and possible mechanisms. Oecologia 78, 383–388.

Xiao, H.Y., Liu, C.Q., 2002. Sources of nitrogen and sulfur in wet deposition at Guiyang, Southwest China. Atmospheric Environment 36, 5121–5130.

Xiao, H.Y., Tang, C.G., Xiao, H.W., Liu, X.Y., Liu, C.Q., 2010a. Mosses indicating atmospheric nitrogen deposition and sources in the Yangtze River drainage basin, China. Journal of Geophysical Research-Atmosphere 115, D14301. doi:10.1029/2009JD012900.

Xiao, H.Y., Tang, C.G., Xiao, H.W., Wang, Y.L., Liu, X.Y., Liu, C.Q., 2010b. Tissue S/N ratios and stable isotopes ($\delta^{34}S$ and $\delta^{15}N$) of epilithic mosses (*Haplocladium microphyllum*) for showing air pollution in urban cities in Southern China. Environmental Pollution 158, 1726–1732.

Yoneyama, T., Kamachi, K., Yamaya, T., Mae, T., 1993. Fractionation of nitrogen isotopes by glutamine synthetase isolated from spinach leaves. Plant and Cell Physiology 34, 489–491.