15N natural abundance of Hinoki cypress and the available nitrogen in soils

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The natural abundance of stable N isotopes has been used for interpreting N cycles in forest ecosystems (Robinson *et al*.2001). Because isotopic fractions during microbial transformations (mineralization, nitrification, denitrification) creates N pools with distinct isotopic signatures (Nadelhoffer and fry 1994). In other words, measuring the isotopic signatures of N pools has the possibility of understanding N transformation. Because the primary N source of plants is soil, it has been conceived that plant (foliar) isotopic signatures is reflected the isotopic signatures of soil (Hogberg *et al*. 1997). Some studies on foliar isotopic signatures assumed that foliar isotopic signatures can reflect the isotopic signatures of available nitrogen the plant utilize in soils. However, the measurements of isotopic signatures of the available N in soil have been labor-intensive, which prevents us from complete understanding of N cycle in plant-soil systems by using isotopic signatures. Therefore, we examined the relationships between foliar isotopic signatures and the isotopic signatures of available nitrogen in soil. The aim of this study was to understand what factors were determined foliar isotopic signatures of Hinoki cypress (*Chamaecparis obtusa* Endlicher).

Foliar and soil sampling were carried out at three regions in Japan. The regions were Norikura in Nagano Prefecture, Okutama, in Tokyo and FM tama, which is the experimental forest of Tokyo University of Agriculture and Technology. We chose seven plots of Hinoki cypress in all of our study areas. Three plots were in Norilkura and two plots were in Okutama, the other plots were in FM tama. In 2008, we collected five samples of foliage of Hinoki cypress from each stand and three subsamples of mineral soil (A horizon, 0-10 cm depth), which were adjacent to the stands of Hinoki cypress. Apart from the forests of Hinoki cypress, we collected only mineral soil at another four plots (three plots were in Norikura, the other was in FM tama). Soil subsamples were composited for heterogeneity and extracted with 2M KCl solution. Extracts were measured the concentration and the isotopic signatures value of the available N (NH_4^+, NO_3^-, DON). We measured isotopic signatures- NO_3^- by denitrifer method (Sigman *et al*.2001; Casciotti *et al*. 2002) and isotopic signatures-TDN (NH_4^+ + NO_3^- + DON) by denitrifer method after persulfate oxidation (Knapp et al.2005). isotopic signatures- NH_4^+ was measured by the combination diffusion method (Holmes *et al*.1998), persulfate oxidation, and denitrifer method. isotopic signatures-DON and isotopic signatures-DIN (NH_4^+ + NO_3^-) were calculated by mass balance equations. We also measured total isotopic signatures and foliar isotopic signatures of Hinoki cypress.

Foliar isotopic signatures were 15N enriched by 1.5 per mil compared with isotopic signatures-DIN in soil and there was a strong relationship between these. This suggested that isotopic signatures-DIN in soil determines foliar isotopic signatures of Hinoki cypress. Generally, the order of soil isotopic signatures was consistently DON, total N, NH_4^+, NO_3^- in order of high values for each plot. The fact that foliar isotopic signatures were 15N enriched in relation to isotopic signatures-DIN may implies that Hinoki cypress uses DON as a nitrogen source at least. However, calculation with Iso-Source model (Phillips et al. 2003) estimated, the contribution of DON to Total N uptake by Hinoki cypress was less than 25 percents. Consequently, it was suggested that Hinoki cypress primarily uses inorganic N in soil as N sources and the foliar isotopic signatures of it is determined by isotopic signatures-DIN in soil.