



## **Quantification of nitrous oxide (N<sub>2</sub>O) uptake in boreal forest soils by combining isotopic and microbial approaches**

Nina Welti, Henri Siljanen, Christina Biasi, and Pertti Martikainen

University of Eastern Finland, Environmental Science, Biogeochemistry Research Group, Kuopio, Finland (nina.welti@uef.fi)

The amount of nitrous oxide (N<sub>2</sub>O) produced during denitrification is highly regulated by the function of the last reductase enzyme (nitrous oxide reductase; nosZ) which is known to be inhibited by oxygen, low pH and low temperature, which are typical characteristics of boreal peatlands and some forest soils. Denitrification can be a sink for N<sub>2</sub>O, if the last step of the process is very efficient. Generally, the N<sub>2</sub>O sink potential of soils is poorly constrained; while uptake rates were often observed in field studies, the data was rejected as analytical errors or artifacts. This led to the question: when and by which mechanisms does N<sub>2</sub>O uptake occur in natural boreal forests?

In order to answer this question, we established a <sup>15</sup>N<sub>2</sub>O tracer experiment where the production of <sup>15</sup>N<sub>2</sub> and consumption of <sup>15</sup>N<sub>2</sub>O were quantified in aerobic and anaerobic conditions followed by abundance analyses of genes and transcripts. The laboratory incubations were complemented with molecular approaches which linked the N<sub>2</sub>O dynamics with individual microbial species and transcriptomics. The abundance of denitrifying functional genes and gene transcripts reducing nitrous oxide (nosZ) were quantified throughout the experiment with sacrificial sampling in order to solve the role of typical and atypical denitrifying populations on N<sub>2</sub>O consumption.

For this study, a Finnish boreal spruce forest and peatland were selected where previous field measurements have revealed negative N<sub>2</sub>O fluxes (i.e. N<sub>2</sub>O uptake). Soil horizons were selected in both the organic layer and uppermost mineral soil layer and in the peat layers 0-10 cm and 10-20 cm, where oxygen is limited and N<sub>2</sub>O uptake occurs at the field scale. <sup>15</sup>N-N<sub>2</sub>O (99 AT %) was added to an initial N<sub>2</sub>O concentration of 1.7 ppm. All soils were flushed with 100% helium prior to the N<sub>2</sub>O addition to ensure that the NO<sub>3</sub> stocks were reduced, leaving the added N<sub>2</sub>O as the sole activator of N<sub>2</sub>O uptake and primary N source.

Aerobic N<sub>2</sub>O uptake was quantified in the mineral horizon of the spruce forest soil (20% from initial concentrations) and both layers of the peat (average 35%). Anaerobic N<sub>2</sub>O uptake was quantified in both the peat (85- 98%) and spruce forest soils (89-95%) at each layer at a much higher rate than aerobic uptake, indicating that the microbial community responsible for N<sub>2</sub>O uptake is present and can be activated within the soil and peat layers. The majority of the N<sub>2</sub>O uptake occurred within the first 48 hours after N<sub>2</sub>O additions, indicating that this is a quickly occurring reaction when N stocks are limited to only N<sub>2</sub>O. Only typical denitrifier transcripts of nosZ were found in the soils layers under the conditions consuming N<sub>2</sub>O in both spruce forest soils and boreal peatland, suggesting that typical denitrifiers are primarily responsible for N<sub>2</sub>O uptake.