

## Relationship of N<sub>2</sub>O emission and environmental factor between different vegetation in a cool temperature

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Nitrous oxide (N<sub>2</sub>O) is a greenhouse gas, mainly produced by soil microbial processes (i.e., denitrification and nitrification) within nitrogen (N) cycle. The N<sub>2</sub>O emissions from soil are controlled by factors such as soil temperature, moisture, pH, and N availability in soil. However, the relative contribution of these environmental factors to N<sub>2</sub>O emission vary across different types of ecosystem and climate. Also, microbial functional genes driving N cycle are important as the controlling factors of N<sub>2</sub>O emission from soil. We hypothesized that driving factors for N<sub>2</sub>O emissions from soils of different vegetation are influenced not only litter quality (CN ratio, residual organic matter) but also factors related to the functional gene abundance of soil microbes and soil related environmental factors (temperature and soil moisture). In this study, we aimed to clarify the spatial and temporal patterns and their driving factors including both environmental and microbial variables (i.e., functional genes in soils) in a cool temperate forest in eastern Hokkaido, northern Japan. We established study plots in two forest types: broad-leaved forest dominated by *Quercus crispula* with *Sasa nipponica* as understory vegetation; and evergreen needle-leaved forest dominated by *Abies sachalinensis* (i.e., total 6 plots = 2 vegetation x 3 replicates). We observed N<sub>2</sub>O emission flux from soil using a closed chamber method from April to October in 2019. Soil temperature and moisture were also observed in surface soil near the chamber. We collected soil samples for analysis of inorganic N (NH<sub>4</sub>-N and NO<sub>3</sub>-N), dissolved organic carbon (DOC), soil pH, and abundance of bacterial and archaeal *amoA* (AOB and AOA), *nirK*, *nirS*, *nosZ1* and *qnorB* (functional genes related to nitrification and denitrification) as copy number in surface soil. We used generalized additive model (GAM) to investigate the driving factors influencing the N<sub>2</sub>O emissions in each forest type.

Average N<sub>2</sub>O emission rates were 8.75 ug N m<sup>-2</sup> h<sup>-1</sup> (± 4.45 SD) in the broad-leaved forest, and 7.41 ug N m<sup>-2</sup> h<sup>-1</sup> (± 3.51 SD) in the needle-leaved forest. The highest N<sub>2</sub>O emission rates were observed in July, when soil temperature was the highest. The GAM analysis indicated that NH<sub>4</sub>-N, *nosZ1* and DOC were selected in the best models in the broad-leaved forest, while soil temperature, soil moisture and *nirK* were selected in the best model in the coniferous forest. DOC is an energy source of soil microbes in the denitrification process. Therefore, it is assumed that it was chosen with *nosZ1* as the best model. Since *nosZ1* is a functional gene that reduces N<sub>2</sub>O to N<sub>2</sub>, it is considered that N<sub>2</sub>O was reduced to N<sub>2</sub> by complete denitrification, resulting in a decrease in N<sub>2</sub>O emission. On the other hand, factors related to the environment surrounding the soil, such as soil moisture and soil temperature, were selected as the best model for the coniferous forest. From this, it is considered that the change in the physical environment surrounding the coniferous forest soil is relatively large. Since the coniferous forest is an evergreen conifer, the forest canopy is closed, so the soil temperature and soil moisture change greatly in early spring due to the influence of temperature. It is considered that *nirK* involved in nitrite reduction was selected together with soil moisture because it was inhibited when soil moisture was low.

In our result, there was no significant difference in N<sub>2</sub>O emission between two forest types, but the results of this study suggested that the factors affecting N<sub>2</sub>O emissions differed between two forest types. Soil chemical indicators such as resources for soil microbes (nitrogen and carbon) affect the broad-leaved forest, and physical factors such as soil temperature and soil moisture content and physical factors affect

in the coniferous forest.

Keywords: Nitrous oxide, Forest soil, Soil microbes