

The effect of soil freeze-thaw on nitrogen transformation through the root litter changes

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Soil freeze-thaw cycles are considered to alter soil nitrogen (N) cycle through physical disturbances of soil, changes in root litter quality, inhibition of microbial N immobilization and others. However, these mechanisms have not been well elucidated yet. Plant litter is important substrate for N mineralization by soil microbes. It has been reported that root litter mass is quantitatively comparable to those of leaf litter in various forest ecosystems. Previous studies suggested that physical disruption of root litter by freeze-thaw cycle in winter affect soil N dynamics through the change in substrate availability. In this study we aimed to clarify that effect of root litter on the rates of soil N mineralization and nitrification under various conditions of soil freeze-thaw.

This study was conducted at Shibeche experimental forest, Kyoto University located in Eastern Hokkaido, Japan. The study site is dominated by Natural oak (*Quercus crispula*) with dense dwarf bamboo (*Sasa niponica*) as understory vegetation. Soil is Humic Andosol. In July 2013, we collected 0-10cm mineral soil and fine root (<2mm) of oak in the 2500 cm² square plot. The collected soil was sieved to 2mm to remove coarse gravel and coarse organic matter. Fine roots for incubations were separated manually from the organic matter. The fine roots were added to 25g soil as 0, 5 and 15 mg g soil⁻¹, respectively. The soil were exposed to three different freeze-thaw treatments: +5 °C ~-5 °C, -5 °C ~0 °C, -5 °C constant and +5 °C constant for 7days in low temperature incubator. After these freeze-thaw treatments, the soil were incubated at +5 °C for 2 days. For the samples exposed at +5 °C ~-5 °C and -5 °C constant were also incubated at +5 °C for 7 days and at +10 °C for 2 and 7 days. Each treatment had four replications. Soils were extracted using potassium chloride (KCl) before and after incubations, and were measured for ammonium (NH₄) and nitrate (NO₃) concentrations in the extracts. Net production rates of NH₄ and NO₃ were calculated as differences of NH₄ and NO₃ contents in soil between before and after the incubations. After the freeze-thaw treatment, roots were extracted using distilled water, and were measured for dissolved organic nitrogen (DON) in the extracts.

Root litter addition significantly increased the net NH₄ production incubated at 5 °C for 2 days after all freeze-thaw treatments (15 mg added >0mg added) with maximum at -5 °C ~0 °C treatment followed by -5 °C constant treatment. However, these effect were not observed in higher incubation temperature (10 °C) and longer incubation period (7days), rather dominated by N immobilization in those treatments. Similarly, the root litter additions significantly increased the net NO₃ production rate (nitrification rate) incubated at 5 °C for 2 days after all freeze-thaw treatments (15 mg added >0mg added) with maximum at +5 °C ~-5 °C treatment. However, these effect were not observed in higher incubation temperature (10 °C) and longer incubation period (7days) as same as the effects to the NH₄ production. The DON supply by water extraction from root litter tended to be large at -5 °C ~0 °C treatment.

These results indicated that increases of soil freeze-thaw cycles with root litter addition increased the net NH₄ production and nitrification. It was suggested that DON supply from root litter by soil freeze-thaw cycle related to these impacts. These effect seems to be remarkable in shorter period (2 days) and lower temperature (5 °C incubation). Furthermore, net NH₄ production, the sum of net NH₄+NO₃ productions and DON supply from root rate were higher at -5 °C ~0 °C treatment than those at +5 °C ~-5 °C treatments, implying that magnitude (temperature ranges) of freeze-thaw cycle was not simple explain variables to impact of freeze-thaw on the microbial NH₄ production and nitrification activities.