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Stimulation of nitrogen turnover due to nutrients release from aggregates affected by freeze-thaw in wetland soils

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ABSTRACT

The freeze-thaw phenomenon will occur more frequently in mid-high latitude ecosystems under climate change which has a remarkable effect on biogeochemical processes in wetland soils. Here, we used a wet sieving procedure and a barometric process separation (BaPS) technique to examine the responses of wetland soil aggregates and related carbon and nitrogen turnover affected by the freeze-thaw treatment. Wetland soil samples were divided into a treatment group and a control group. The treatment group was incubated at temperatures fluctuating from 10 °C to −10 °C, whereas the control group was incubated at the constant temperature of 10 °C. A 24 h process was set as the total freeze-thaw cycle, and the experiment had 20 continuous freeze-thaw cycles. In our results, the freeze-thaw process caused great destruction to the >2 mm water-stable aggregates (WSA) fraction and increased the <0.053 mm WSA fraction. The dissolved organic carbon (DOC) content was stimulated during the initial freeze-thaw cycles followed by a rapid decline, and then still increased during subsequent freeze-thaw cycles, which was mainly determined by the soil organic carbon (SOC). The NH_4^+ and NO_3^- content, respiration rate and gross nitrification rate were all significantly improved by the freeze-thaw effect. Because the amount of NH_4^+ and NO_3^- expressed prominent negative responses to the content of >2 mm WSA fraction and the gross nitrification rate can be stimulated at the initial freeze-thaw cycles, nutrients and substrates may play a leading role in the freeze-thaw treatment regardless of the minimal influences on microbial biomass pools.

1. Introduction

The seasonal freeze-thaw process is a significant meteorological phenomenon in mid-high latitude and high altitude areas (Yu et al., 2011). The IPCC has reported that atmospheric temperature in the spring would fluctuate remarkably under global climate change, and there would be thinner overwinter snow cover in some regions (IPCC., 2007). Snow cover as a thermal insulating layer can prevent soil from directly experiencing severe atmospheric temperatures (Yi et al., 2015). Changes in the duration and thickness of snow cover will make the freeze-thaw process occur more frequently in the future (Makoto et al., 2014).

Freeze-thaw cycles can significantly disturb biogeochemical processes by influencing the soil structure, plant roots and microorganisms (Koponen et al., 2006; Kreyling et al., 2010; Matzner and Borken, 2008). Previous studies have indicated that the stimulated nitrogen transformation rate mainly resulted from active microorganism communities, abundant energy supply and reaction substrates from the available organic matter (Henry, 2007). Wetlands are one of the most important terrestrial carbon and nitrogen pools (Gorham, 1991; Jia et al., 2017). Boreal wetlands are considered to be especially vulnerable to freeze-thaw processes with low temperature and high water availability (Dise, 2009; Essl et al., 2012). Considering that boreal wetlands store approximately one-third of the organic carbon in worldwide soils (Bridgham et al., 1999; Zhao et al., 2016), we should clearly determine the key processes of wetland soil carbon and nitrogen turnover under the freeze-thaw effect.

Recent articles have reported that the freeze-thaw process can alter soil nitrogen pools and stimulate nitrogen turnover, such as mineralization, nitrification and emission of nitric gas in forest ecosystems, tundra ecosystems, crop lands and meadows (Henry,

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2007; Kielland et al., 2006; Ludwig et al., 2006; Miao et al., 2014). However, the nitrogen turnover process in wetland soils is still poorly understood. Due to the restrictions of research methods, many studies have not reported the gross nitrogen transformation rate. The net nitrogen transformation rate alone cannot reflect the real nitrogen turnover. We cannot distinguish whether alterations in the net nitrification result from changes in mineralization, denitrification or assimilation.

Moreover, soil aggregates are the basic units of soil structure and contain most of the organic matter (Jastrow, 1996). The content of water-stable aggregates determines the potential of soil runoff and erosion. Some articles have reported that the freeze-thaw process can destroy soil macro-aggregates whose stability has shown to be inversely proportional to soil moisture content in general (Kværnø and Øygarden, 2006; Lehrsch, 1997). Because the soil moisture content of wetlands is usually much higher than that of other soils, the freeze-thaw process may lead to severe destruction of aggregates. The relationship between the disruption of water-stable aggregates in wetlands and the availability of readily decomposable substrates for nitrogen turnover may play a key role under freeze-thaw stress (van Bochove et al., 2000).

In this study, we used a wet sieving procedure to perform a careful analysis of different fractions of aggregates and related soil organic carbon changes during the simulated freeze-thaw treatment. Furthermore, we measured the gross rates of nitrification, denitrification and respiration of wetland soils using the barometric process separation (BaPS) technique after every two freeze-thaw cycles. The objectives of this study were to examine how the freeze-thaw process influences wetland soil aggregates and clarify the relationship between changes in aggregates and wetland nitrogen turnover to a certain degree.

2. Materials and methods

2.1. Study site description and soil sampling

The study site is located in the National Nature Wetland Reserve of Xingkai (Khanka) Lake of Sanjiang Plain (45°21.875′N, 132°18.372′E), which is the largest expanse of freshwater lacustrine wetlands in China. The annual mean precipitation is 561 mm and the air temperature ranges from −39 °C in January to 36 °C in July with a mean of 3.5 °C; these data are based on 54 years of meteorological records from 1957 to 2010 (Zou et al., 2014). There is no standing water on the study site, with predominant flora of *Calamagrostis angustifolia* and *Zizania latifolia*. The soil type was silty clay meadow marsh soil. We collected the surface layer (0–10 cm) followed by a “S” shaped route with 3 repetitions. The roots in the soil were not removed in order to maintain the original condition of soils. After the soil samples were homogenized, we transported them to the laboratory at the Northeast Institute of Geography and Agroecology, CAS, where they are awaiting further laboratory simulations. The major soil properties of the study site are shown in Table 1.

2.2. Experimental design

After measuring the initial soil moisture content, the soil samples were divided into the control group and the treatment group, and 60 sub-samples were put into aluminum bottles. The bottles were covered with plastic film with three pinholes each for gas exchange and to minimize water evaporation from the soil. We also weighed the bottles every day and added de-ionized water to keep the soils at the initial moisture content during the freeze-thaw incubation. The control group was incubated at a constant 10 °C, which was measured by a thermostat. The treatment group was simulated with daily freeze-thaw frequencies according to predicted spring ambient air temperatures, with fluctuations from 10 °C to −10 °C. The specific temperature setting is shown in Fig. 1. A total of 24 h of the procedure from thawing to freezing to thawing was regarded as a freeze-thaw cycle. There was a total of 20 freeze-thaw cycles in the incubation period. After every two freeze-thaw cycles, we took out 3 sub-samples each from the control group and the treatment group for further measurements of aggregate fractions and variables related to wetland carbon and nitrogen turnover.

2.3. Stability measurements of water-stable aggregates (WSA)

Soils in wetlands are always supersaturated or covered with standing water. Therefore, it is more suitable to measure the stability of aggregates using a wet-sieving apparatus (Denef et al., 2001, 2007; Klute, 1986; Yoder, 1936). The soil samples were passed through a 5-mm sieve and then air-dried. After that, 16 g of a soil sample were immersed in water, on the top of a nest of sieves with 2.0 mm, 0.25 mm and 0.053 mm meshes for 10 min, and the process was repeated in triplicate. The nest of sieves was then raised and lowered with a 30-mm amplitude to ensure that the top meshes were not exposed to air. After the wet sieving procedure, the aggregates on each sieve were collected and dried at 50 °C to constant weight. The WSA sizes were classified into four fractions (>2.0 mm, 0.25–2.0 mm, 0.053–0.25 mm and <0.053 mm) based on the size of the sieves. The <0.053 mm WSA fraction was calculated by the residual value (i.e., the total mass of the soil minus the mass of other three WSA fractions). The changes in the stability of the aggregates were expressed as the percentage of each WSA fraction relative to the initial amount of the soil.

2.4. Measurement of carbon and nitrogen turnover

We used the BaPS technique to determine the soil heterotrophic respiration, gross nitrification and denitrification rates. Nitrification is a process that decreases pressure because of a net consumption of O_2; denitrification is a process that increases pressure because of the net production of NO, N_2O, N_2 and CO_2; soil respiration is a neutral pressure process if the respiration quotient equals 1. This method is based on the measurement of change of CO_2, O_2 and gas pressure in an isothermal gas tight soil system, as shown in Equation (1). Temperature adjustment and CO_2 dissolution in the soil water are also considered in the BaPS instrument (UMS GmbH Inc., Munich, Germany). For further details, including discussions and comparisons between the ^{15}N dilution methods, see Breuer et al. (2002), Ingwersen et al. (1999, 2008) and Müller et al. (2004). The BaPS technique was suitable for incubating well-aerated soils. Therefore, we chose relatively drier soils in wetlands in our

Table 1
Major soil properties of the study site in the Xingkai lacustrine wetlands. Values shown are the mean ± SE (n = 3).

Clay content (%)	pH (H_2O)	Total C (mg/g)	Total N (mg/g)	C:N	Water content (%)
64.3 ± 7.02	5.40 ± 0.02	40.79 ± 3.56	0.67 ± 0.01	61.39 ± 6.37	46.09 ± 0.48

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