

Effects of warming and nitrogen addition on soil enzyme activities

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Abstract. Due to the intensification of human activities, global warming and increased atmospheric nitrogen deposition have become important driving factors of global change. In order to understand the effects of long-term warming and nitrogen application on soil physical and chemical properties and enzyme activities, the effects of warming and nitrogen application on grassland soil enzyme activities were measured. The aim was to reveal the response of enzyme activity in an Inner Mongolian desert steppe to long-term warming and nitrogen addition. The results were as follows. In 2018, nitrogen fertilization significantly increased soil dehydrogenase activity, and soil depth significantly affected soil urease, β -Glucosidase, phosphatase and dehydrogenase activities. In 2019, the four soil enzyme activities only showed significant differences at different soil depths, while the effects of warming, nitrogen addition and their interactions on soil enzyme activities were not significant. In general, the enzyme activities of desert grassland soil were not affected by warming, while nitrogen addition had a greater impact on the soil enzyme activities. However, large difference in soil moisture between the two years of the experiment was also a key factor leading to large fluctuations between the two years, which indicated that in the semi-arid desert grassland, the change of soil nutrient characteristics is also affected by water content.

Introduction

Since the Industrial Revolution, the intensification of human activities and the over exploitation and utilization of natural resources, such as deforestation, increasing use of artificial fertilizers, industrial waste gas emissions, and the gradual increase in agricultural land use, have led to a series of global climate change issues such as an increase in the Earth's surface temperature, increased atmospheric nitrogen deposition, changes in precipitation patterns, and the destruction of the ozone layer. Among them, the most important factors that cause global change are temperature rise and atmospheric nitrogen deposition, and a series of global ecological and environmental problems caused by these factors have become of great concern to ecologists and the public. Soil enzymes are mainly formed due to the metabolic process of soil microorganisms, the secretion of soil animals and plant roots and the decomposition of residues. The catalytic mineralization of organic carbon, nitrogen, phosphorus and other substances is the main way in which extracellular enzymes obtain nutrients. The activity of soil extracellular enzymes can represent the catalytic history of microorganisms and indicate the response of microorganisms to environmental changes. Therefore, soil extracellular enzymes can not only be used to estimate the nutrient requirements of microorganisms, but also be used as sensitive indicators to reflect changes in the soil environment. In addition, soil enzymes can decompose organic nutrients in soil into available nutrients, which can be directly used by plants and microorganisms (Waring et al., 2014). Therefore, in order to better understand the soil nutrient cycle and the response of the microbial community to warming and nitrogen addition, it is particularly important to study changes in soil enzyme activity under warming and nitrogen addition. In this study of *Stipa breviflora*—a constructive species in Inner Mongolian desert grassland—we used an experimental

platform with simulated warming and nitrogen deposition treatments over 14 years to study the effects of warming and nitrogen input on the distribution and activity of enzymes.

Methods and Study Site

Soil samples were collected on the long-term warming and nitrogen application experimental platform of the Comprehensive Experiment and Demonstration Center of the Institute of Agriculture and Animal Husbandry (111 ° 53 ' E, 41 ° 46 ' N, elevation 1 450 m) located in the south-central area of Siziwang Banner, Ulanqab City, Inner Mongolia Autonomous Region. The study area is located in an area with a typical continental monsoon climate in the mid-temperate zone. The winter is long and cold, and summer precipitation is limited but more concentrated, and the intra-annual temperature difference is large. The soil in this area is classified as light chestnut soil with sandy soil texture and is a relatively poor soil. The soil type is characterized by rich potassium content, limited nitrogen and phosphorus content, and low organic matter content in the soil. The experiment with elevated temperature and nitrogen application started in early May 2006 and is still ongoing. The experiment uses a 2 × 2 factor split plot design, with elevated temperature as the main treatment factor and nitrogen application as the side treatment factor. Under the main treatment, there are 2 temperature treatments (control and warming), and each treatment is set with 6 repetitions. There were 4 treatments in the whole experiment, namely control (C), warming (W), nitrogen addition (N) and warming+nitrogen addition (W+N). Each treatment was set with 6 repetitions, making a total of 24 plots. In the six main heating plots, infrared radiators were used to heat the main quadrats continuously throughout the year. The annual average increase in surface temperature reached 1.3 °C. In order to reduce or eliminate the error caused by radiator shading or other factors, a "false heater" of the same size and shape as the radiator was installed at the same position in each control plot. At the end of June to the beginning of July of each year, ammonium nitrate (NH₄NO₃) was used as the nitrogen source to apply nitrogen to the nitrogen application sub-plots once a year, at a rate of 10 g · m⁻² · yr⁻¹ actual nitrogen. Soil samples (0-10 cm, 10-20 cm and 20-30 cm) for determination of soil enzyme activity were collected in August 2018 and 2019. After passing through a 2 mm soil sieve, plant roots were removed, and part of the soil was put into a vacuum bag for low-temperature preservation and were quickly returned to the laboratory, and stored in a 4 °C refrigerator.

Results and Discussion

There is a strong relationship between soil environment and enzyme activity. Soil temperature can not only have a direct impact on soil enzyme activity, but can also have an indirect impact on soil enzyme activity by influencing the microorganism releasing enzymes or dynamic characteristics. In this study, the effects of the treatments on different enzyme activities also differed between the two study years. In 2018, warming increased urease and phosphatase activities and decreased β-Glucosidase and dehydrogenase activities, but the effects were not significant. In 2019, warming increased urease and β-Glucosidase activity, reducing phosphatase and dehydrogenase activity, but again the effects were not significant. Some studies have shown that soil urease activity will gradually increase with warming, but the effect of temperature on soil urease activity will gradually weaken with longer treatment time. However, soil urease activity is not limited by temperature alone. Some studies have found that soil water content is also a key factor affecting soil urease activity. Previous studies have confirmed the promotion of soil enzyme activity by warming (Stone et al. 2012). However, since soil enzyme activity is sensitive to a small increase in temperature, and enzyme activity under a small

increase in temperature depends on change in soil moisture, when drought is caused by warming, the dissolution and diffusion rates of soil nutrients are low, and enzyme activity is limited. Warming had no significant effects on soil urease, phosphatase β -Glucosidase or dehydrogenase in the two years of the experiment, indicating that these four enzyme activities were not very sensitive to soil warming in the study area. Warming reduced the diversity of the bacterial community, but had no significant impact on the soil fungal community, and thus had no significant impact on most soil enzyme activities. The effect of nitrogen addition on soil enzyme activity showed an upward trend in the two years, but the effects on urease, phosphatase and β -Glucosidase activity were not significant, and the effect on dehydrogenase activity was only significant in the 0-10 cm soil layer and 20-30 cm soil layer in 2018. Nitrogen addition can increase the content of nitrogen in the soil, thus increasing the activity of soil urease, which may be related to the participation of urease in the decomposition of urea and the nitrogen cycle in the soil. The research results of Wang *et al.* (2014) showed that an appropriate amount of nitrogen addition (N_{25} , N_{50}) can provide nitrogen sources for soil microorganisms, alleviate nitrogen competition between plants and microorganisms, and promote microbial activity. Under high nitrogen conditions (N_{200}), soil pH may be significantly reduced, and pH is generally regarded as the main factor limiting soil enzyme activity.

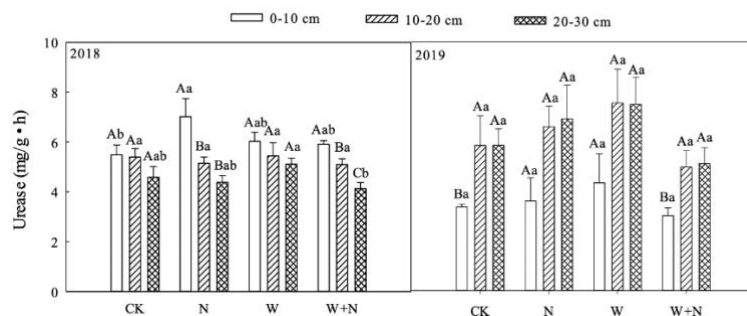


Fig.1 Soil urease activity of different soil layers under each treatment in 2018 and 2019

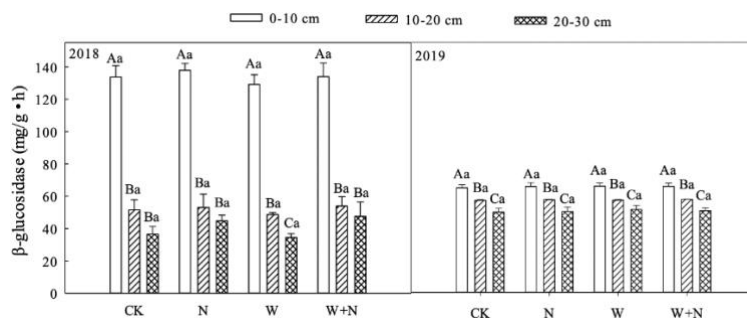


Fig.2 Soil β -glucosidase activity of different soil layers under each treatment in 2018 and 2019

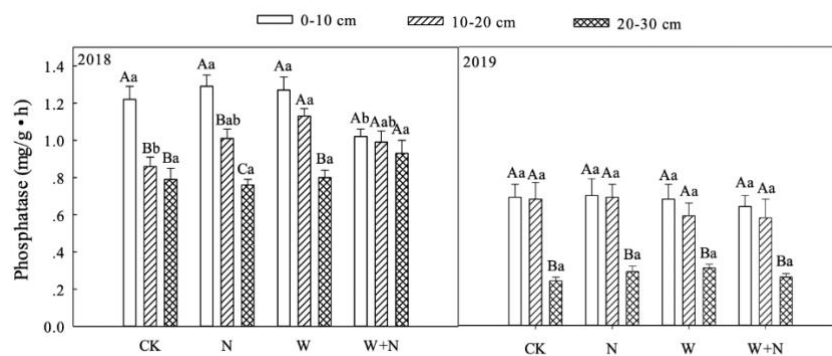


Fig.3 Soil Phosphatase activity of different soil layers under each treatment in 2018 and 2019

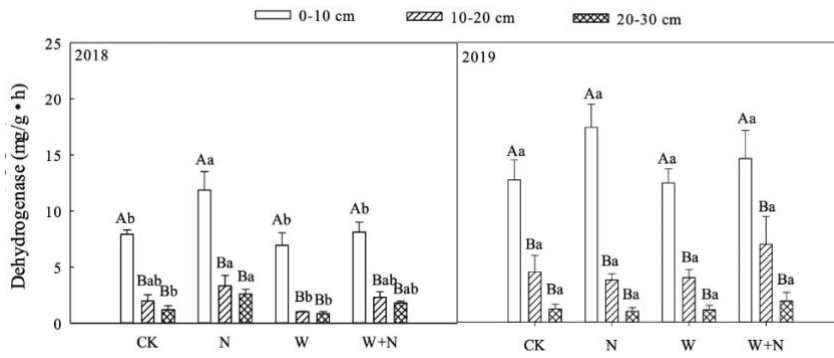


Fig.4 Soil Dehydrogenase activity of different soil layers under each treatment in 2018 and 2019

Conclusions

The effect of elevated temperature on soil physical and chemical properties and enzyme activities was relatively limited, while the effect of nitrogen application on soil physical and chemical properties and enzyme activities was greater than that of temperature. However, due to differences in the timing of soil sampling during the two-year experiment, there were large differences in soil water content, which indicates that water also has an important impact on soil enzyme activities.

References

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