



The effects of climate change on inorganic nitrogen cycling communities in grasslands

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Soil microbial communities play a major role in driving elemental fluxes in terrestrial ecosystems. Nitrogen (N) is a fundamental element for all living organisms and changes to the N cycle can have dramatic consequences in terms of feedbacks to climate change. It is therefore pivotal to precisely assess the effects of future climatic conditions on the microbial communities controlling its cycling, in order to increase our ability to predict climate change consequences. Here we report a study on inorganic nitrogen transformation from a multifactorial Climate Change experiment (ClimGrass – Grassland Carbon and Nutrient Dynamics in a Changing Climate) in a managed montane grassland in the central Austrian Alps which aims to assess the individual and combined effects of warming, elevated CO₂ and drought. Samples were collected at peak growing season in July 2017 from field plots subjected to either ambient climate or simulated future climate conditions (combined +3 °C and +300 ppm CO₂) and drought in a full factorial design. Gross nitrification rates were measured by isotope pool dilution assays with the goal of linking them to the identity and abundance of different nitrifier groups. Five functional genes responsible for both NH₄⁺ and NO₂⁻ oxidation were quantified by qPCR at both DNA and cDNA levels (indicating presence and expression of each gene). In addition, we sequenced the same set of functional genes (Illumina MiSeq) with the goal of assessing changes in nitrifier community composition induced by climate change treatments. Our results demonstrate a strong effect of drought on the transcription of ammonia monooxygenase subunit A (amoA) genes of comammox clade B Nitrospira (comaB) and ammonia-oxidizing archaea (AOA), with gene copy numbers being significantly lower in drought treatments compared to the controls, while combined warming and elevated CO₂ had no effect. Ammonia-oxidizing bacteria (AOB) and nitrite-oxidizing bacteria (NOB) were affected by the future climate treatment only, suggesting shifts in functional microbial community composition. Additionally, the future climate treatment did not significantly affect inorganic N pools or process rates, whereas drought significantly increased NH₄⁺ concentrations and gross nitrification rates. The positive relationship between NH₄⁺ concentrations and gross nitrification rates under drought suggest that the nitrification process was rather limited by substrate availability than by microbial activity. In conclusion this study demonstrates that climate change affects the diversity and differential gene expression of functional microbial communities responsible for inorganic N cycling, but that gross nitrification process rates responded to different triggers than nitrifier abundance.