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# Substrate-driven microbial response: A novel mechanism contributes significantly to temperature sensitivity of $N_2O$ emissions in upland arable soil

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## ABSTRACT

The mechanism by which temperature sensitivity (TS) of soil $N_2O$ emissions is increased by agricultural management with application of nitrogen fertilizer (AMN) is unclear. We hypothesized that a higher TS of $N_2O$ emission induced by AMN is the result of the faster growth of specific microorganisms in response to faster nitrogen (N) mineralization at higher temperatures. To test this hypothesis, we used reciprocal transplants to separate the contributions of abiotic and microbial components to the TS of $N_2O$ emissions in an arable soil receiving organic and inorganic fertilizers and its neighboring natural grassland soil treated with two levels of N. $N_2O$ sources were separated with acetylene, and the abundances of $N_2O$-producing microbes were assessed by quantifying the copy numbers of the associated functional genes. Compared with natural soil, only changes in abiotic properties increased the $Q_{10}$ (the factor by which the rate increases with a 10 °C rise in temperature) by 105.7%, while changes in both abiotic and the microbiome increased the $Q_{10}$ by 225.2%. Higher TS of $N_2O$ emission in the arable soil induced by a microbiome shift was associated with faster N mineralization, increased proportion of nitrification-$N_2O$ emission, and faster growth of ammonia-oxidizing bacteria at higher temperatures. Addition of ammonium nitrate further enhanced the TS of $N_2O$ emissions, the proportion of nitrification-$N_2O$ emission, and the increased extent of the growth of ammonia-oxidizing bacteria in the soil with AMN compared to the natural grassland soil. Substrate-driven growth of ammonia-oxidizing bacteria with higher substrate preference contributes significantly to the higher TS of $N_2O$ emission caused by AMN.

## 1. Introduction

Nitrous oxide ($N_2O$) is the third most important greenhouse gas (IPCC, 2007) and is a major cause of stratospheric ozone depletion (Ravishankara et al., 2009). About 60% of anthropogenic $N_2O$ is emitted from agricultural soils, mostly derived from applications of synthetic fertilizer N and animal manure (Smith, 2017; Zhou et al., 2017). Besides by N sources, the emission of $N_2O$ is controlled by many other factors, including contents of soil water and carbon and temperature (Weier et al., 1993; Conrad, 1996; Avrahami et al., 2003; Baggs and Philippot, 2010; Blagodatskaya et al., 2014; Smith, 2017). Of these, temperature is particularly significant because it influences current $N_2O$ emissions and also impacts future $N_2O$ emissions via positive or negative feedback (Smith, 1997). We analyzed the results in the review by Smith (1997) and subsequent studies and found that N fertilization increases the $N_2O$-emission rate and also its temperature sensitivity (TS). The $Q_{10}$ levels in soils with AMN are higher than in soils without N fertilizer in both disperse studies and studies with paired N and no N treatments (Fig. 1, Table S1). However, the mechanisms underlying the AMN-enhanced TS of soil $N_2O$ emission are not well known.

The variation in TS of soil $N_2O$ emission has been attributed mostly to anaerobic-zone development (AZD) in aggregated soils (Smith, 1997, 2017). This explanation suggests that a temperature increase leads to an

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