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# Urea- and nitrapyrin-affected $N_2O$ emission is coupled mainly with ammonia oxidizing bacteria growth in microcosms of three typical Chinese arable soils

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ABSTRACT

It is unclear how inhibition of nitrous oxide ($N_2O$) emissions by nitrification inhibitor (NI) is regulated through the ammonia oxidizing bacteria (AOB) or archaea (AOA) in arable soils. In this study, we investigated effects of 2-chloro-6-(trichloromethyl)-pyridine (Nitrapyrin, NP) on $N_2O$ emissions, and characterized the ammonia oxidizing microbial community in three arable soils typical of northern China. In alluvial, black, and paddy soils, average $N_2O$ emission rates were increased by addition of urea by 3.5, 0.7 and 2.1 pM $N_2O$ $g^{-1}$ soil $h^{-1}$, respectively, but were reduced by 2.9, 0.4 and 2.2 pM $N_2O$ $g^{-1}$ soil $h^{-1}$ when urea was applied with NP. The stimulation and suppression of $N_2O$ emission by urea and NP occurred alongside fluctuation in the growth of AOB in alluvial and paddy soils ($P < 0.01$). Weak stimulation and suppression of $N_2O$ emissions by urea and NP corresponded with weak effects on AOB abundances in the black soil. Changes in $N_2O$ emissions were not significantly correlated with AOA abundances in any of the three soils. The results showed that differential responses of $N_2O$ emission to urea and NP application in arable soils can be mainly explained by differences in growth of ammonia oxidizing bacteria.



## 1. Introduction

Application of nitrogen (N) fertilizer, one of the major causes of agricultural source $N_2O$, contributes to over half of the anthropogenically induced $N_2O$ emission (Mosier et al., 1998). It is estimated that up to 2.5% of fertilizer nitrogen can be converted to $N_2O$ on a global scale (Davidson, 2009). Also in China, a great proportion of $N_2O$ emission results from nitrogen fertilizer use. For example, annual fertilizer-induced $N_2O$ was estimated to be 210.5 Gg $N_2O$–N $year^{-1}$ in the 1990s (Zou et al., 2010). Nitrification is an essential process in agricultural ecosystems, oxidizing ammonium fertilizer nitrogen into nitrate and releasing $N_2O$ as a by-product. Simultaneous application of nitrification inhibitors (NI) with mineral-N fertilizer offers a promising approach retarding nitrification and mitigating $N_2O$ emission from arable soils (Wolt, 2004). The relative reduction of $N_2O$ emission by this approach is variable (Wolt, 2004; Watanabe, 2006; Snyder et al., 2009), and previous studies have focussed on the role of adsorption and hydrolysis of NIs to explain this variation (Jacinthe and Pichtel, 1992; Vannelli and Hooper, 1992). However, natural variations in the soil microbes which control this process at a biological level have not been fully addressed.

Many NIs, including dicyandiamide (DCD) and NP, inhibit nitrification by interfering with the ammonia monooxygenase (AMO) enzyme which catalyzes ammonia oxidation, the first and rate-limiting step of soil nitrification (McCarty, 1999; Kowalchuk and Stephen, 2001). In soil, discrete groups of bacteria and archaea are both known to be involved in ammonia oxidation (Kowalchuk and Stephen, 2001; Spang et al., 2010; Zhang et al., 2010). By characterizing the conserved *amoA* gene, which encodes a subunit of the AMO enzyme, researchers have shown that

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