

Contents lists available at ScienceDirect

Soil Biology and Biochemistry



journal homepage: http://www.elsevier.com/locate/soilbio

Wheat rhizodeposition stimulates soil nitrous oxide emission and denitrifiers harboring the *nosZ* clade I gene

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ARTICLE INFO

Keywords: Nitrous oxide (N₂O) nosZ Rhizodeposition Stable isotope probing (SIP) Wheat

ABSTRACT

Stimulatory effects of growing plants on nitrous oxide (N₂O) emissions have been widely reported in terrestrial ecosystems, but the potential mechanisms responsible for these effects remain unclear. This study revealed that wheat can induce a 3.5–9.2-fold increase in N₂O emissions under different soil fertility levels, and that this "plant" source of N₂O occurs in the rhizosphere. Moreover, plants induced soil niche differentiation between denitrifiers harboring the nitrous oxide reductase genes *nosZ*I and *nosZ*II. Pulse labeling of wheat demonstrated that 67% of ¹³C-labeled *nosZ*I-type denitrifiers, but no *nosZ*II-type denitrifiers, were more abundant in the rhizosphere than in bulk soil. Furthermore, a higher percentage of bacterial genomes containing nitrite reductase genes was found within plant-associated *nosZ*I-type denitrifiers than *nosZ*II-type denitrifiers, favoring NO₂⁻ to N₂O conversion. Overall, this study revealed a strong selective stimulating effect of wheat on soil denitrifiers through root-derived carbon and a key role of the *nosZ*I-type community in rhizosphere denitrification.

1. Introduction

Nitrous oxide (N_2O) is one of the most important greenhouse gases and ozone-depleting substances on earth (Ravishankara et al., 2009; Tian et al., 2016), and agricultural fields with large nitrogen (N) inputs contribute about 30% of the total terrestrial emissions worldwide (Syakila and Kroeze, 2011). The atmospheric N_2O concentration has persistently increased at nearly 0.75 ppb per year since 1970 (IPCC, 2014). The only known N_2O sink is its enzymatic reduction to dinitrogen (N_2) by denitrifiers harboring the nitrous oxide reductase gene (*nosZ*), which consists of two distinct clades (*nosZ*II- and *nosZ*II-type denitrifiers). The proportion of these two denitrifier types in soil can have substantial consequences on net N_2O emissions (Jones et al., 2014).

The stimulatory effect of growing plants on N_2O emissions has been widely reported (Bowatte et al., 2014; Hakata et al., 2003; Zou et al., 2005), and there may be several potential mechanisms that contribute to the "plant emissions". First, N₂O could be produced directly from plant organs or tissues through NO₂ assimilation (Bruhn et al., 2014; Hakata et al., 2003; Smart and Bloom, 2001). For example, it has been reported that wheat leaf N₂O emissions were correlated with leaf nitrate assimilation activity and occurred during photoassimilation of NO₂⁻ in the chloroplast (Smart and Bloom, 2001). Second, plants can play the role of a "conduit" to facilitate the transport of N₂O from rhizosphere to atmosphere (Baruah et al., 2012; Bowatte et al., 2014; Yan et al., 2000). Scanning electron microscopy revealed that N₂O emissions were correlated with stomatal frequency of leaf and leaf sheaths (Baruah et al., 2012). Third, some plant-inhabiting microbes in (or on) leaves and roots have the capacity to produce N₂O just as in soil (Ai et al., 2017; Bowatte et al., 2015). Bacteria such as *Nitrosospira* sp. on pasture grass can convert 0.12% of the oxidized ammonia to N₂O (Bowatte et al., 2015).

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https://doi.org/10.1016/j.soilbio.2020.107738

Received 5 September 2019; Received in revised form 21 January 2020; Accepted 29 January 2020 Available online 1 February 2020 0038-0717/© 2020 Elsevier Ltd. All rights reserved.

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