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Gas kinetics and stoichiometry from four fungi incubated under conditions favouring denitrification

Lena Rohe[1], Reinhard Well[1], Shahid Nadeem[2], and Peter Dörsch[2]
[1]Thünen Institute of Climate-Smart Agriculture, Braunschweig, Germany (lena.rohe@thuenen.de)
[2]Norwegian University of Life Sciences (NMBU), Department of Environmental Sciences (IMV), Ås, Norway

Even though the ability of fungi to produce the greenhouse gas nitrous oxide (N_2O) during denitrification has been demonstrated, the proportion N_2O emissions from fungal denitrification in soils cannot yet be determined or predicted. In order to develop methods for estimating the fungal proportion, N_2O must be partitioned to bacterial and fungal denitrification. The denitrification regulatory phenotype (DRP) is well described for a number of bacterial strains (Bergaust et al. 2010, Bergaust et al. 2011), but to our knowledge there are only few data relating to the fungal DRP in terms of oxygen (O_2) tension in fully stirred cultures at which they start producing N_2O. The aim of this study was to analyse the kinetics of fungal denitrification combined with analysis of the isotopic composition of N_2O. In particular, the ^{15}N site preference of N_2O ($SP\text{-}N_2O$) is known to be a promising tool to differentiate between N_2O produced during bacterial and fungal denitrification.

Four fungal species (*Fusarium oxysporum*, *Fusarium decemcellulare*, *Fusarium solani* fsp. *pisi* and *Chaetomium funicola*) were incubated as batch cultures in a robotized incubation system (Molstad et al. 2007) for 165h. Batch cultures were incubated in 120 ml flasks containing 50 ml of growth medium amended with ample amounts of carbon and nitrate in a He atmosphere with 2 vol%O_2. To test for pH effects, a complex medium (Shoun et al. 1992) with pH values adjusted to 6.9 and 7.4 as well a minimal medium (Dox 1910) with a pH value of about 7.9 were used. O_2 consumption and production of nitric oxide (NO), N_2O, dinitrogen (N_2) and carbon dioxide (CO_2) were monitored at high temporal resolution while isotopic composition of N_2O was analysed in samples taken manually at selected time points.

All four fungal cultures quickly consumed O_2. NO production increased strongly before O_2 was completely consumed and was followed by immediate N_2O production. The kinetics of N_2O production differed to published kinetics of denitrifying prokaryotes by showing a lower sensitivity to O_2. This could result in a larger share of fungal denitrification under microaerobic conditions in soil.

Isotopic analysis of N_2O confirmed previous results of specifically high $SP\text{-}N_2O$ values of fungal produced N_2O. We further showed that $SP\text{-}N_2O$ values of fungal N_2O are quite stable and do not depend on denitrification kinetics. Likewise, incubation conditions such as pH of the medium had little impact on $SP\text{-}N_2O$ values. These findings support the usage of $SP\text{-}N_2O$ values for partitioning

N_2O soil fluxes and provide a tool to study the biology of fungal denitrification under field conditions, which is needed to develop mitigation strategies of N_2O from fungal denitrification.

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