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Elucidating soil pore N₂O production and consumption processes using isotope and microbial gene analysis: A depth profile approach

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N₂O is a stratospheric ozone depleting substance and a potent greenhouse gas which significantly contributes to global warming. Although soils are the largest source of N₂O emissions, knowledge gaps in the understanding of N₂O production and reduction processes in soils still exist. Here, we investigated N₂O production and consumption processes along soil depth profiles in a mesocosm experiment using natural-abundance N₂O and NO₃⁻ isotopic signatures as well as abundances of soil microbial genes associated with N₂O production (*nirK*, *nirS*) and reduction (*nosZ*). Soil columns either displayed undisturbed soil stratification (control treatments), or contained an artificial clay layer at 35 cm depth (clay treatment), which acted as a diffusion barrier and thus induced O₂-limited conditions in deeper strata. We collected soil pore gas, soil solution and soil samples at five depths of the soil columns over the course of four weeks. In addition, we continuously monitored N₂O fluxes at the soil surface and soil environmental parameters (oxygen, moisture, temperature) along the soil depth profiles. Microbial gene analysis in soil samples revealed similar abundances of *nirK*, *nirS* and *nosZ* in the two treatments across the entire soil depth profiles. The distribution of the functional genes was thus not indicative of enhanced N₂O production and/or reduction in O₂-limited conditions. However, lowest O₂ concentrations below the clay layer were associated with highest ¹⁵N and ¹⁸O enrichments in both NO₃⁻ and N₂O, indicating N₂O production by denitrification and fractional N₂O reduction. In addition, we found higher N₂O concentrations and surface fluxes for the clay treatment. Our observations imply a dominance of N₂O production over N₂O reduction, even under conditions most favorable for complete denitrification.