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Interactive comment on "The fate of N₂O consumed in soils" *by* B. Vieten et al.

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We are aware that nitrogenase activity of free living organisms is very low. As referee #2 mentioned, a km value of 24kPa was determined for purified component proteins from *Klebsiella pneumoniae* (Jensen and Burris, 1986). Even when we consider that km values regarding N2O consumption can vary by a factor up to 2000 between soils, organisms and the methods used (Conrad, 1996), we would not expect nitrogenase to substantially contribute to N2O consumption within the range of concentrations applied in our experiment. However, other - currently unknown - pathways of reduction of N2O to NH3 may exist and discoveries of new processes and responsible organisms continue to occur in our times (e.g.: Strous et al., 1999).
The objective in our study was to clarify whether assimilatory N2O reduction is an ecologically relevant process occurring in soil. Considering the observed large rates of

N2O consumption in our experiments, the thermodynamical advantage of N2O over N2 as substrate for the production of NH3 (Shestakov and Shilov, 2001), and the evidence of direct N2O to NH3 reduction as a biological process (Yamazaki et al., 1987), albeit only for a low affinity enzyme (Jensen and Burris, 1986) so far, the negative result regarding N2O assimilation is unexpected. One would expect natural selection to have favoured organisms assimilating N2O rather than N2.
We agree with referee #2 that in not finding any evidence for a fourth pathway of assimilatory reduction of N2O to NH3 does not prove the absence of this process. It rather shows that such a process is most likely not of ecological importance in our soils under the investigated environmental conditions.

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