

Response to second round of Referee #2 suggestions:

Dear Referee,

we appreciate your additional suggestions to our first response and integrated them into our current manuscript. We outlined our point to point reply below (in bold).

General comments:

Thanks, I realise it will be a small % but important to acknowledge. It would also be good to add a sentence in the discussion to suggest that DNRA (& anammox) are measured in addition in future work to rule out potential artefacts – there are now several (sediment) papers on the artefacts of the co-occurrence of NO_3^- reducing processes on the IPT assumptions.

We added the importance of measuring N_2 production in future studies in the discussion:
Line 574: “In future ^{15}N -labelling studies, DNRA should be measured to rule out potential pool dilution by the co-occurrence of NH_4^+ production. “

Line 482- 485: “ N_2 production measurements (from anammox and denitrification) were not performed in this study, but should be carried out in future studies to account for potential artefacts by co-occurring NO_3^- reduction processes.”

I think you just need to change “In the vicinity of DNRA in $^{15}\text{NO}_3^-$ incubations...” to “In relation to DNRA in $^{15}\text{NO}_3^-$ incubations...”

We rephrased the whole sentence in the results section to: “In $^{15}\text{NO}_3^-$ incubations, active DNRA produces $^{15}\text{NO}_2^-$ and $^{15}\text{NH}_4^+$ from $^{15}\text{NO}_3^-$ which can contribute to $^{46}\text{N}_2\text{O}$ production by AO.”

I meant that in Bristow et al. and Dalsgaard et al that a lot of their measurements are concentrated below 1-2 μM oxygen and fewer concentrations in the ‘higher’ 10-20 μM range... i.e. focusing on the concentrations where the inhibition/regulation really ‘happens’. But I understand the reasons you describe above given the standard deviations of O_2 measurements and without more sensitive sensors it would be difficult to designate concentrations, I agree. I appreciate that Dalsgaard et al do have a nice reactor/microcosm set up which I realise is very specialised for precisely these experiments and with larger volumes than the serum vials – also that it is a lot of work with these types of experiments. I think it would still be good to add a sentence/statement as to why ‘your’ oxygen concentrations were chosen (e.g. given the reasons above, SD in measurements etc) if possible.

We think that the standard deviations of the different oxygen levels explain why we did not resolve the lower end better and did not add anything there. However, we agree with the referee that it does not become clear why a larger range was applied for the $^{15}\text{NO}_3^-$ treatments, so we explained that better:

Line 167-169: “For the ^{15}N - NO_3^- incubations two more O_2 treatments with 21.5 ± 2.8 and $30.2 \pm 3.35 \mu\text{M}$ O_2 were carried out to extend the range of a previous study in which N_2O production from $^{15}\text{NO}_3^-$ did not decrease up to O_2 concentration of $7 \mu\text{M}$ (Ji et al. 2018).”

But if there is the same amount of particles in all vials/O₂ manipulations then there is potential for some anoxic processes to be ‘unaffected’ by O₂ additions - with some changes in anoxic microsite volume with O₂ diffusion into particles. I realise this is hard to rule out – especially as you collect small particles from the water column to use, indicating that they are there. I think it would be important to write something shortly about why you consider it unlikely that any (significant) anoxic niches occur.

We do not consider it unlikely that anoxic niches occur, but we do think that anoxic niches do not explain the large difference in response of N₂O production at high oxygen levels in the depth profiles (no to little N₂O production) compared to the manipulated oxygen treatments (very high N₂O production), because the potential for anaerobic microsites is given in all incubations. We added the potential for anaerobic processes inside microniches in line 530 – 534: „It further indicates that high N₂O production from NO₃⁻ in high oxygen treatments is unlikely an effect of anoxic micro niches. While anoxic micro niches in batch incubations can never be fully ruled out, there is no reason why they should systematically change N₂O production in NO₃⁻ from NO₂⁻ incubations at the same oxygen treatment. “

Shortly suggest/indicate benefits of also measuring other end products (e.g. ¹⁵N-N₂ and maybe also ¹⁵NH₄⁺ from DNRA) in the text (i.e. how does the ‘efficiency’ of denit change with changing O₂)

We added the advantage of measuring several potential end products in line 574 about DNRA and in line 482- 485 about anammox and denitrification (see first comment). The advantage of having production rates of N₂O and N₂ together is already discussed starting in line 485, and also starting in line 532, where we highlight the value of having the N₂O yields. The different responses/efficiency of denitrification to oxygen is extensively discussed in lines 523 onwards.

Some kind of ‘conclusion’ is needed at the end of the last sentence in relation to your study. Papers referring to ‘cryptic’ biogeochemical cycling in ODZ waters would also be nice to include in relating to ‘hidden’ processes.

As suggested, we added a conclusion to in line 581: “Even if hybrid N₂O production rates are overestimated, it remains the major N₂O production mechanisms of AO in this study.”

In this paragraph we want to explain the occurrence of hybrid N₂O formation rather than hidden process – so we did not add papers on cryptic cycling there.