Review of bg-2019-132 Major role of ammonia-oxidizing bacteria in N2O production in the Pearl River Estuary

August 7, 2019

Dear Dr. Zhang

Thanks for submitting responses to reviewers of bg-2019-132 "Major role of ammoniaoxidizing bacteria in N_2O production in the Pearl River Estuary". Based on those and my own reading I think the article is promising due to sound data and interpretation but it is not publishable in the present form in Biogeosciences since it lacks some structure as pointed out by Reviewer 1. This aspect precludes full understanding of your work in mainly two aspects:

- 1) Mixing of results and discussion such as in Figure 2 that shows field data combined with panels i and j that, according to text, ΔN_2O is calculated for incubations so, they are probably results from incubations. Since there is no explanation of experimental treatments with variable O_2 concentration, I would assume that those are derived from field sampling, right? If that were the case, results are mixed with discussion in this figure and text in page 8, paragraph starting in L24. Alternatively, oxygenation conditions of incubations are missing in the method section.
- 2) Better explanation is needed regarding "concentration-based "rate" measurements ..." (Reviewer 2) that not only refers to "... changing of the nutrients can be sensitively detected during incubations" as you pointed out, but also to multiple and simultaneous sources and sinks, therefore at the most you obtain a net rate since it is likely that these nutrients are simultaneously removed.

In any case, this aspect is not clear in the text. Please clearly show how you interpret each of rates (net production or decay/ incubation time) of Table 1:

 $\Delta N_2 O \text{ (nmol } L^{-1}h^{-1}\text{)}$

 ΔNO_3^- (µmol L⁻¹h⁻¹)

 ΔNO_2^- (µmol L⁻¹h⁻¹)

 Δ (NH₃ + NH₄⁺) (µmol L⁻¹h⁻¹). You mean NH₃ + NH₄⁺ instead of NH₃/NH₄⁺ (a ratio), don't you?

A cartoon similar to the one below may help to explain your rationale for interpreting these rates and better sustain conclusions such as "... results clearly indicate that nitrification occurred during the entire P01 incubations, and suggest that denitrification may be present in the ending phase... " (Page 10, L11-12).



In addition, please consider the following:

- 1. Pag. 1. Line 19. All "N₂O parameters". Do you mean N₂O-related parameters?
- 2. P. 1, Line 22-25 "Taken together, the in situ incubation experiments, N₂O isotopic composition and concentrations, and gene datasets suggested that the high concentration of N₂O (oversaturated) is mainly produced from strong nitrification by the relatively high abundance of AOB in the upper reaches as the major source of N₂O emitted to the atmosphere in the whole estuary."

What is the evidence for the whole estuary? What about seasonal variability?

- 3. Pag. 3, L.12. "anaerobic particle interiors". Do you mean anoxic particle interiors?
- 4. Page 3, L 20 (de)nitrification. Why using ()?
- 5. Page 4, L25. "Temperature and salinity were continuously measured with the CTD system." Define continuously and in detail depths
- 6. Page 4. L24. "2.2 Biogeochemical parameters, N2O emissions, and isotopic analysis" Detail whether this is in the water column in your 11 sites or in experiments, or both

Figure 1. Enlarge symbols + and * (or change colors). There is an extra red dot, isn't?Table 1. a) Fix typos such as "Liner Equation". Use either regression or equationb) Since this is regression, R2 is the coefficient of determination!

Figure 6. Since there is an equation (y = f(x)) with a line, it is a regression analysis (one independent and one dependent variable) whereas in correlation there are not dependent or independent variable. Coefficient must be R2 for regression.

Page 7, L 19. Explain this please Page 10, L10. Why is there a "... but..." here? This sentence is not clear.

Sincerely your

Silvio Pantoja Associate Editor