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Interactive comment on "Reviews and syntheses: Processes and functional genes involved in nitrogen cycling in marine environments" *by* Ramiro Ramos and Silvia Pajares

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We would like to thank the Reviewer for the detailed suggestions. We have taken these comments into consideration and we are sure they will strongly help to improve our manuscript. We address each of the Reviewer's points raised below:

1. "In their manuscript "Processes and functional genes involved in nitrogen cycling in marine environments," the authors have tried to assemble a comprehensive review of nitrogen cycling in the ocean. However, multiple recent and excellent reviews exist on this topic, foremost Kuypers et al. 2018, which is also frequently cited in this manuscript. Without wanting to offend the authors, it becomes quite clear when reading

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this manuscript that they cannot (yet) match the knowledge of the authors of some of these recent reviews. This makes me call the purpose of this manuscript into question. The apparent aim here is to present another review of all reactions and microorganisms involved in N cycling in marine systems, for which I must unfortunately say there is no need right now, and the authors lack the necessary expertise in many areas. This becomes apparent as many (recent) studies on different aspects of N cycling in the ocean are missing (e.g., Delmont et al. 2018, showing the abundance of non-cyanobacterial heterotrophic diazotrophs in marine metagenomes), and many pathways and proteins involved are incomplete and partly wrong (like for instance assimilatory nitrate and nitrite reduction)."

We thank the Reviewer for the comments. As we mentioned to Reviewer #1, we understand that writing a review paper on this topic is very risky given the great contributions made by other authors in recent years. However, none of these review papers have covered the main aspects we try to cover with our manuscript. For instance, the recent review of Kuypers et al. (2018) summarizes the current understanding of the microbial nitrogen-cycling network but does not focus on the microbially mediated N processes in marine ecosystems. Additionally, other recent papers reviewing N cycling processes in marine environments are too specific (e.g. Devol et al. in 2015 reviewing marine sediments or Lam and Kuypers in 2011 reviewing OMZs) or focus on other aspects of marine N cycling (e.g. Voss et al. in 2013 focusing on the anthropogenic effects or Zehr and Kudela in 2011 analysing the current understanding and identifying knowledge gaps). Again, while most of these papers cover biochemical, genetic or anthropogenic aspects of N cycling, in our manuscript we try to summarize the current knowledge on the nitrogen processes studied so far in the ocean, as well as the distribution of microorganisms involved in N cycling in marine environments and the factors affecting it, which are aspects that have not been covered in such an integral way so far. We are aware that there has been a great advance in the study of the marine nitrogen cycle in recent years and for that reason we believe it is appropriate to conduct a review in the field including the latest discoveries that have not been covered

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in other review papers. We apologize for not including several recent studies; however, we have included over 50 studies from the last three years. In the case of the study by Delmont et al. 2018, we must say that our manuscript was completed months ago, and it is hard to keep up with every study published in the last months (in fact, Delmont et al. 2018 was published a few days after submitting our article to Biogeosciences). In the revised version we will include it, as well as others that have been published since we first sent the manuscript for revision. Additionally, we will correct and complete the metabolic pathways and proteins.

2. "However, the authors do have and present a fairly good overview of the primer sets available and used for detecting the different steps of the N cycle. In view of this expertise, and the focus of the other available reviews, I would strongly advise the authors to focus this manuscript on a comprehensive review of the available tools to study the functional guilds involved in N cycling, which questions these can answer, and what are their limitations (which is the part I miss the most in the current manuscript). Here I would suggest to include discussions of limited coverage of some (most) primer sets and the existence of multiple pathways for the same reactions (e.g. in both assimilatory [nirA, nirBD, OTR/ONR] and dissimilatory [nirK, nirS, nrfAH] nitrite reduction)."

We appreciate a lot the Reviewer's recommendations. We agree with the Reviewer that the tools used so far to study the functional guilds involved in marine N cycling, the limited coverage of most primer sets and the existence of multiple pathways for the same reactions are interesting topics for our review. Then, we will include these suggestions without deviating from the main purpose of our manuscript.

3. "I would also advise the authors to be as complete on the processes they include into this review as possible. For instance, the nitrification section focuses almost exclusively on AOA and ignores the marine AOB (foremost Nitrosococcus) and especially NOB (Nitrospina and Nitrococcus, but also Nitrospira). On the other hand, comammox is included, even if not observed in marine systems so far. The same goes for N-DAMO, which discusses only the nitrite-dependent NC10 bacteria and only shortly mentions

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the nitrate-dependent archaea. If there is a lack of molecular tools to detect some of these groups (as I earlier advised this should be the focus), this should be stated and discussed. A review like this should then also include a critical discussion of the limitations of any PCR-based study, as many metagenomic-based studies have recently been published showing the amount of novelty that is missed by these approaches."

We appreciate the comments and agree with the criticisms made by the Reviewer. In the next version we will include the missing information on nitrifiers and n-damo. We will also remove the comammox section and include the reviewer's suggestions, again, without deviating from the main purpose of our manuscript.

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