

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

**Anonymous Referee #2**

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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 Kuyper et al. 2018, which is also frequently cited in this manuscript. Without wanting to offend the authors, it becomes quite clear when reading 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 be-

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comes 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). 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). 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 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.

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