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Interactive comment on "High denitrification and anaerobic ammonium oxidation contributes to net nitrogen loss in a seagrass ecosystem in the central Red Sea" *by* Neus Garcias-Bonet et al.

Anonymous Referee #2

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Overall, the paper is well-written and the internal logic is consistent. However, I was disappointed that the authors paired state of the art Unisense probes with outdated methods for denitrification and N-fixation measurements, and ignored DNRA altogether. DNRA is an important nitrate loss pathway in seagrasses (Aoki & McGlathery 2017; An and Gardner 2002), but in contrast to denitrification, it returns N to the system as NH4, rather than removing excess N to the gaseous form. Thus it competes with denitrification and potentially exacerbates eutrophication. DNRA could have easily been measured as NH4 from the slurries in this experiment (and could still if there are samples in the freezer).

The helium purging and 15NO3 IPT method is most appropriate for anoxic water columns where nitrification is not expected to play a significant role. Unfortunately, there are severe limitations with using slurries and helium purging when coupled nitrification-denitrification is likely to be important, as it is expected to be near the oxic-anoxic interface or near vegetated roots that actively pump down oxygen. There is a vast literature indicating that coupled nitrification-denitrification is important in coastal sediments (Christensen et al. 1987; Laursen and Seitzinger 2002), including in hypoxic conditions (Gardner and McCarthy 2009). Anoxic slurries destroy natural redox gradients and prevent nitrification (Eyre et al. 2002), which is often the primary NO3- source for denitrification (e.g., Laursen and Seitzinger 2002). The method used here may underestimate actual denitrification rates where there was in situ coupled nitrification and denitrification (van Lujin et al. 1996). Given that the authors did not report ambient NO3, NO2, or NH4 concentrations, it's difficult to know whether direct denitrification played an important role in situ (if there are water samples in the freezer, I would advise running them for nutrient concentrations). In the future, measuring 28N2 fluxes, or using a MIMS to measure 28, 29, and 30N2 from intact sediment cores is more likely to account for coupled nitrification-denitrification as well as direct denitrification. By underestimating denitrification, the authors may also have overestimated the importance of anammox. The authors need to acknowledge these shortcomings and try to address them. Although the experimental design has shortcomings, the authors may be able to use the equations in McTigue et al. (2016) to try to correct for their underestimate, although the experimental designs were different.

As Reviewer 1 mentioned, there have been many documented issues with N-fixation from ARA, including shifting the microbial community (Fulweiler et al. 2015) and potentially altering rates. The authors should acknowledge these shortcomings.

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