Biogeosciences Discuss., 10, C1034–C1039, 2013
www.biogeosciences-discuss.net/10/C1034/2013/

Biogeosciences Discussions

Interactive comment on "Anaerobic ammonium oxidation, denitrification and dissimilatory nitrate reduction to ammonium in the East China Sea sediment" *by* G. D. Song et al.

Anonymous Referee #3

Received and published: 17 April 2013

The paper presents a study of the rates and relative importance of denitrification, anammox, and nitrate ammonification (DNRA) along a depth gradient in the East China Sea, based on slurry incubations with 15N nitrate and ammonium. All three processes are found to be active and important. The presence of DNRA calls for modification of the standard approach for calculation of denitrification and anammox rates. Furthermore, a correction is made for the liberation of nitrate from an apparent intracellular nitrate pool.

The novelty of the study lies in it being the first to report this type of data from the East China Sea and in the detail of treatment of how to interpret data in the presence

Full Screen / Esc

Printer-friendly Version

Interactive Discussion

Discussion Paper

of DNRA and internal nitrate pools. In contrast to the impression left by this paper, it is, however, not the first to demonstrate experimentally the simultaneous activity of anammox, denitrification, and DNRA in sediments, nor the first to propose formula for resolving rates of the processes based on 15N incubations (see below).

The paper is carefully conducted and clearly written, but the discussion of the methodological aspects is too long while the discussion of the environmentally relevant results could be expanded. Moreover, there are flaws in the interpretation of the data.

1) The most serious issue is that rates determined in slurries from specific depth intervals in the sediment, amended with high concentrations of nitrate, are added together to obtain area-based rates. One issue here is that this relies on an accurate determination of the zone of nitrate consumption in situ, and it is not clearly specified how this was done. More importantly, however, the entire approach is not valid as clearly evident from loads of previous work on these processes in aquatic sediments. Sediment homogenization and slurrying generally stimulates activity, and at the high concentrations of nitrate the rates measured in the slurries are potential rates. Hence summing them up results in stark overestimation of the integrated in situ rate. This is easily seen in studies that determined rates in both slurries and intact sediment cores (e.g., see review by Trimmer and Engström 2011, in Ward et al. (eds.): Nitrification) and in the study by Sokoll et al. frequently cited in the present paper. It is correct, as suggested by Sokoll et al. that in situ rates may lie somewhere between whole-core and slurry rates when denitrification of intracellular nitrate plays a large role, since this is not captured with the isotope pairing technique. But the area-based rates obtained in the present study are 1-2 orders of magnitude higher than typical rates in continental sediments. Claiming such rates to be realistic is extraordinary and hence requires extraordinary evidence. Yet, the authors do not at all reflect on this matter. I strongly recommend that the depth integration exercise be abandoned altogether. It will be a serious setback if this type of rates makes it into the databases.

2) Another major issue has to do with the calculations and interpretations concerning

intracellular nitrate pools. As presented now, there are several apparent inconsistencies, which need to be discussed (see comments below).

3) The method used to determine 15N-labeling of ammonium to the best of my knowledge also converts organic N, which means that it is not possible to discriminate assimilatory and dissimilatory nitrate reduction to ammonium, ANRA and DNRA. This weakens the conclusion that DNRA is important in these sediments.

Specific comments: 4674, 18-20: Novelty overstated. It is not correct that previous reports on the coexistence of denitrification, anammox, and DNRA are only from flux measurements. Dong et al. 2009, referenced later in the MS, did this in incubations. Also Trimmer and Nicholls 2009 (referenced) detected all three processes in experimental incubations, although DNRA rates were very low.

4675, 1-2: Novelty overstated. It is not correct that there is currently no such model. Spott and Stange (2007) developed a model for this, and Jensen et al. 2011 (referenced and including authors of this paper!) present equations for situations where anammox and DNRA co-occur.

4677, 15-17: The hypobromite method as applied by Preisler and here to whole slurries oxidizes organic amines as well as ammonium. This is why a distillation step was introduced by some workers. Thus, it is not possible to distinguish assimilatory and dissimilatory nitrate reduction to ammonium. This is particularly serious in a setting like this where it is argued that the sediment may contain algae that will reduce nitrate for assimilation.

4678, 8 + 19 + 4680, 1: This is very confusing and seems self-contradicting. There are direct observations of nitrate accumulation, and nitrate release is assumed to happen only initially due to mixing, which is happens long before N-15 tracer is added, such that Fn does not change during incubation. In this case, it should be easy to correct for the release by simply using the nitrate concentration measured at the beginning. So why is there need for complicated calculations to be able to ”conclude” that nitrate

BGD
10, C1034–C1039, 2013

Full Screen / Esc
Printer-friendly Version
Interactive Discussion
Discussion Paper

release occurs? Surely, the best indication of this must be that it is directly measured? The only reason to do the complicated calculation must be that nitrate may be released gradually. What is the justification for the assumption that it is not? In any case, I find ”conclude” to be too strong a word here, since one may think of other factors that could lead to F*N < FN.

4681, 6-17: How is nitrate penetration depth defined. This is important for the integrated rates later. How does the depth integration deal with isolated subsurface peaks? And with the presence of nitrite?

4683, 19-25: Some discussion of the depth distributions of these rates within the sediment is warranted, including the fact that they must be considered potential rates. The increase in DNRA with depth is consistent with the general finding of DNRA increasing in importance in more reducing sediments and that DNRA has often been observed when new nitrate is mixed into reducing, nitrate-free sediments (Nishio et al. 1982 etc.).

4686, 4-5: Again, the novelty is overestimated. Glud et al. (2009) did determine anammox in sediments with nitrate-storing forams and managed to discern the rates. Prokopenko et al. (2005) EPSL, suggested that anammox bacteria may receive nitrate/nitrite from nitrate-storing Thioploca. And in many other cases the good agreement between added and measured initial nitrate concentrations (e.g. early studies by Dalsgaard and others) indicate that there are no large hidden nitrate pools. There is no reason to suggest that earlier studies have all been ignorant about this, which is the impression given now.

4686, 11-13: Forams are present in most sediments. The important question is whether the species composition of the foram community hints at nitrate storage. And correct “similar TO areas”.

4686, 15: ”using conservatively”?

BGD

10, C1034–C1039, 2013

Interactive Comment

4686, 19-20: The fact that nitrate release only ocurred in slurries with 15N nitrate addition is an important, and highly confusing result and therefore should be mentioned in Results. According to p4682, 14-16, nitrate was already released during the preincubation, i.e. before 15N-nitrate was added. So how did the organisms know which amendments were to come? And what about the incubations with 14N-nitrate amendment? If, contrary to what I read on p4682 nitrate was released after amendment, what is the justification of assuming that Fn did not change during the incubation?

4687, 16: I suppose that "the effect of DNRA on denitrification and anammox" means its effect on the measurement of rates of these processes?

4687, 20-21 + Table 5: This is a useful table but it neglects a good number of studies that did not find DNRA to be of any significance (e.g., Binnerup et al. 1998, AEM;. Rysgaard et al. 1993 AEM, Dalsgaard and Thamdrup 2002, Engstrom et al. 2009 L&O).

4688, 5-8: The % DNRA depends critically on accurate determination of the zone of nitrate consumption and on the assumption that nitrate is not limiting for this process in situ (Km so low that potential and in situ rates are the same). These caveats should be discussed.

4688, 10 on: As discussed above, this discussion neglects the fact that slurrying generally stimulates rates. Also, a wide selection of half saturation constants can be found in the literature, often within the range of pore water concentrations. Why are those determined in permeable sediments particularly appropriate here? And obviously nitrate concentrations MUST have been below the Km in part of the nitrate consumption zone. Otherwise nitrate would not be depleted. Rates exceeding 10 and up to 33 mmol/m2/d are extreme, higher than most benthic N loss rates from marine settings. The loss rates reported here are NOT "a reasonable estimation of benthic N-loss on the ECS shelf"!

4889, 25-26: The relative contribution of denitrification and anammox are interdependent variables. A correlation of the two makes no statistical sense.

BGD

10, C1034–C1039, 2013

Interactive comment on Biogeosciences Discuss., 10, 4671, 2013.

BGD

10, C1034–C1039, 2013

Interactive Comment

Full Screen / Esc

Printer-friendly Version

Interactive Discussion

Discussion Paper