Biogeosciences Discuss., 11, C3471–C3478, 2014
www.biogeosciences-discuss.net/11/C3471/2014/

Biogeosciences Discussions
Open Access

Interactive comment on "Methane related changes in prokaryotic activity along geochemical profiles in sediments of Lake Kinneret (Israel)" *by* I. Bar Or et al.

Anonymous Referee #1

Received and published: 14 July 2014

The manuscript by Bar Or et al discusses the changes in microbial community (composition and diversity) alongside changes in electron donors and acceptors in a depth profile in the sediments of Lake Kinneret in Israel. The main conclusion of the paper is the link between Thaumarchaeota, which are typically known as marine ammonia oxidizers, and anaerobic methane oxidation. Unfortunately this important piece of information is well hidden within the text among other less significant / valid data.

While the above mentioned conclusion by the authors is very interesting, the paper in its current state requires, in the opinion of this reviewer, significant rewriting with respect to organization, language and to some extent data exclusion.

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

By reading the acknowledgments section it appears that this manuscript has already been under review. Since it has not passed through my hands, I understand that some of my comments may go against suggestion made by previous reviewers. Therefore, I will try to explain my comments as much as possible where I find it necessary.

As a general comment to the writing of the paper, there is a substantial amount of discussion in the results section. The results discussion should be kept short and interpretation free. Unfortunately this is not the case here. Please see below specific comments.

A second point which I find surprising is the choice of methodology made by the authors. In a study that focuses on diversity the authors chose to make use of a low throughput sequencing technique, i.e. cloning rather than pyrosequecning or illumine. Additionally the authors present a low number of sequences per sample. The use of cloning would be more understandable had the authors made full use of the method by amplifying full 16S rRNA sequences, to better resolve the taxonomy. Last, despite the low-throughput, the authors chose to present (analyze) single replicate samples. Overall the author choice of methodology makes their data inappropriate for a full diversity study. Therefore the authors should not over-use their data. The calculation of diversity indexes for comparison between the different samples cannot be meaningful with such a low number of sequences and without any biological or technical replicates. I therefore believe the author should discuss the taxa discovered and their potential biogeochemical role without too much weight on their abundance.

Specific comments (The page numbering refers to the page number in the online pdf version)

One major thing that has not been done throughout the paper is writing taxa in italics. This should be applied from the Domain level onwards.

Title

BGD

11, C3471–C3478, 2014

The authors state in the title changes in activity – however activity measurements are not reported. A correct title would include “Changes in microbial community”

Abstract

P9814 L 9: There is no need for the abbreviation LK in the abstract since Lake Kinneret appears only once. The abbreviation should be reintroduced in the main body of the manuscript.

P9814 L 12: Erase the word “possible” you examined the changes and not whether they are possible.

P9814 L 16: Thaumarchaeota are a group of organisms therefore they do not belong to the family of copper contacting membrane bound monooxygenases. I believe the author refer to the ammonium monooxygenase. Please rewrite the sentence to state that Thaumarchaeota contain such enzymes.

P9814 L 18: Do the authors mean that they have discovered/showed that Thaumarchaeota in Lake Kinneret are ammonia oxidizers? Or do they refer to the fact that Thaumarchaeota are typically ammonia oxidizers? If the latter is the case, the use of the word “discovered” is inappropriate.

P9841 L21: I believe that this should be the guiding line throughout the paper: AOM is driven by iron and not by sulfate. Also unless this was the intent of the authors they should stat that AOM is driven by iron and not sulfate and not as currently written that iron drive AOM is not sulfate driven.

Materials and methods

P9816 L 26: The references for nitrate and sulfate are too old specifically the one from 1974. If these data are used in the discussion which I believe they are not, the authors should provide newer references or their own data (perhaps if there is a routine monitoring program it could be referenced).

BGD

11, C3471–C3478, 2014

P9817 L 8-12: The units here are mixed. Total iron is given in 3% - not stated of what dry/wet weight? Manganese is given in μg g-1. Organic carbon is given again in percent. Please be consistent.

P9817 L 11: Can the authors be certain that the Mn concentration measured 43 years ago is still valid??? Don't you provide actual values from your profiles which in fact are much lower?

P9817 L 12: A similar remark as above the concentration and trend in the sediment is from 1978. Surely there has been sedimentation at the lake bottom since then. This is not a valid reference.

P9817 L22: Can you provide the sampling frequency in these 4 years (yearly, monthly weekly).

P9818 L7: Please add the model of the GC.

P9819 L13: Are the primers 87-907R designed by Ben-Dov as suggested here. I believe that they are older. Unless they were modified in the cited paper, please cite the original reference.

P9820: L1-5. This paragraph needs some rewriting. Something like "inserts were amplified from white colonies using the M13F and M14R primers."

P9820: L 10: The second check for chimeras is not clear. To what did the authors refer: when the two halves did not align? Do you refer if they didn't align to the same reference sequence? Do you mean aligned or do you mean their final location in the ARB guide tree? I am not certain this is a good measure for Chimera as a 450 nt sequence from different parts of the 16S molecule may easily end up aligned to a slightly different sequences. Was there a cutoff in the decision to throw out sequences?

P9820: L14: The authors probably refer to the placement in the ARB guide tree rather than alignment.

BGD
11, C3471–C3478, 2014

Results

P9821 L10: Over a dozen can be 13 or 50. Please be specific.

P9821 L11: please explain seasonally. This was missing from the method section as well.

P9821 L13: profiles of... The word of seems to be forgotten from a previous sentence.

P9821 L19-22: the authors provide here data from old references. The result section should present only results obtained during the course of this study. Interpretation or references to previous studies should be left to the discussion.

P9822 L1: The methane profile is in Fig 2B rather than A. The panels are inverted also in the figure caption. Additionally the figures are, at least in my version, of low quality and cannot be read properly.

P9822: This entire section is mixed with results and their interpretation. Any sentence that uses "suggests", "probably", "support"... belongs to the discussion and should be removed from the results section. This entire section can be much shorter and "cleaner".

P9823 L10-13: The decision which samples to sequence belongs to the methods and can be mentioned once more in the discussion.

P9823 L22-25: This is valid to all sequencing methods. As long as direct counts are not available (via FISH), PCR based data should be used cautiously.

P9823 L26: High degree of richness as compared to what?

P9824 L4-5: highly diverse community – this has to be used comparatively to other environments. And belongs to the discussion.

P9824 L16-23: The use of percentage is not valid in my opinion. Over 10% means 4 sequences. This is meaningless. An increase in Nitrospira to a relatively high per-

BGD

11, C3471–C3478, 2014

centage (11%) – One replicate, 4 sequences (11% of 38). You can say they are found in the deeper samples and not in the shallow one but I would refrain from using any percentages.

P9825 L1-3: The authors jump from phyla (Nitrospirae) to family (Nitrospiraceae) to genera (Nitrospira). If it was done intentionally, make use of the prefix family or genera.

P9825 L6: Please specify which families do: “Our Deltaproteobacteria” refer to.

P9825 L26: Rephrase. About 17% . . . could not be classified using SINA and were classified using ARB instead.

P9826 L2: To the 13%-40% refer to % out of the total community or % out of the Thaumarchaeota - specify?

P9826 L 13: closely instead of close related.

Discussion

P9827 L10: ferrous – the word iron is missing.

P9827 L14: and its resemblance (not resemble)

P9827 L17-21: I would avoid making use of the diversity indexes given the limitation of the methods used and samples sizes and numbers.

P9827 L22: Therefore (not Therfor)

P9827 L26: Proteobacteria are the most described phyla of bacteria (especially from environmental samples) therefore it is not a big surprise that it is among the most abundant phyla. The discussion should be held at the family level or higher taxonomic resolution.

P9828 L2: It is more common and correct to say that the sequences were related to or clustered with sequences of. . . rather than aligned to. Specifically since the sequences aligned to other sequences and not to organisms.

BGD

11, C3471–C3478, 2014

P9828 L3: Some sulfate reducers are also iron reducers. This may be relevant to the iron based AOM discussion. Please specify families found.

P9828 L5: upper part of LK. Does this refer to sediment of water column?

P9828 L9: Chloroflexi are usually rather small. You have stated that sulfate reduction was the main process in the upper part of the sediment. How does this fit with your thoughts regarding the role of Chloroflexi.

P9828 L15-23 This entire sections discusses organic matter usage by different groups. Though interesting it deviates from the AOM topic of the paper. Furthermore the discussion does not follow a single line but rather states that all the groups found may be organic matter consumers. I am curious how does the activity attributed to these organisms fit with the relatively deep O2 penetration of 4 mm which was mentioned earlier. As well as the denitrifies which should be anaerobes. My guess is that the 4 mm O2 penetration is seasonal and was not the case during some of the periods discussed here. But all of this should not be left for the reader to assume or guess but rather be clearly stated.

P9828 L23: Archaeal communities are responsible for many environmental processes. This sentence is meaningless unless you specify which processes.

P9828 L29: similarity at the phyla level is almost meaningless and the authors clearly state that this is not valid at the OTU level. Keep the discussion to meaningful data. It does not make sense to provide information regarding similarity of taxonomic units to which one cannot (practically or potentially) assign a defined functional role.

P9829 L3: Why did the authors use such a low cutoff (90%) for their similarity? Please have a look at the paper by Rosello-Mora and Amann: The species concept of prokaryotes FEMS Microbiol Rev. 2001 Jan;25(1):39-67. The paper shows the correlation between DNA-DNA hybridization (i.e. genomic similarity) and 16S similarity. 90% is quite far off to say anything about the functional similarity of the organisms from which the

BGD

11, C3471–C3478, 2014

sequence was obtained.

P9829 L 25-26: The authors make a factual statement citing a reference from 1992. The use of old reference regarding what is happening in the lake during the course of the present study is done quite often in this paper. If the authors believe the lake remained unchanged since the 70' 80' 90' or so, they should provide evidence for this and state this clearly at the beginning of the manuscript.

P9829 L29- It is more common to say that the newer study supports the older one and not vice versa.

P9830 L5: It may be true that generally sulfate reduction outcompete methanogenesis, however the concept of the sequential redox tower has been discussed recently as more and more "miss fitting" bacteria are found in the wrong place e.g. sulfate reducers in areas of oxygenic phototrophy. For the case mentioned here, have a look at MEPS 107, 177-18 (1994) where co culturing of methanogens and sulfate reducers has been shown. P9830 L15: The same comment as above.

P9832 L5-6 The same comment – old reference for an actual value of a substrate in the lake. Don't you provide Mn data yourself in Fig 2?

P9834 L10-15 Too long sentence. Split and write explicitly to which enzyme you refer to.

Figures

Fig 2: Panels A and B are inverted with respect to the text. There is room to move panel D up to the same line as the other panels. At least in my version the figures are of low quality the text is not readable and the fonts too small.

Figures 3 and 4 – should be done at the family level and restricted to main families not all the observed ones. The latter should be supplied as a supplementary table.

Interactive comment on Biogeosciences Discuss., 11, 9813, 2014.