

## ***Interactive comment on “Archive of bacterial community in anhydrite crystals from a deep-sea basin provides evidence of past oil-spilling in a benthic environment in the Red Sea” by Yong Wang et al.***

### **Anonymous Referee #2**

Received and published: 20 September 2016

Review for Wang et al.

The manuscript submitted by Wang et al. describes the diversity of organisms in anhydrite crystals and further states a high abundance of *Alcanivorax* microbes. The authors see the specific community composition of the 750 years old crystals as indicator of an oxic oil-spilling benthic environment. Overall, the approach and results are highly interesting and definitely worth reporting in Biogeosciences.

I, however, suggest revising and clarifying several parts of the manuscript before final publication.

C1

#### General remarks:

While I do not doubt the results of the metagenomic investigation per se, I am honestly wondering what the meaning of the quantification of microbes based on traces of DNA is and how you could find intact cells being preserved for 750 years in crystals, which are still traceable using FISH. I am missing a detailed explanation and discussion on that topic as also for me the microscopic pictures are not entirely satisfying. Additionally I would like to see some proof of the assembled genome and a metabolic map. Further, the conclusion on past oil-spilling is a bit too loosely connected to the other parts of the story, both in the abstract and the results and discussion parts. This could be done by introducing geochemical data earlier and by rephrasing some of the respective text parts. The references are not in Copernicus style.

#### Specific comments:

##### Abstract:

First sentence should be rephrased as a hypothesis.

I.33: which binning tool? Replace separated by assembled

I. 35. Remove 'the'

I.33-I.37: you are jumping back and forward between the metagenome and the cell identification

I. 40 sentence is misplaced, here. Should probably be moved upwards.

I. 41-45: As written this seems to be highly speculative

##### Introduction

L. 54: Repetition of previous sentence.

I. 57: I don't understand this sentence.

I. 59: replace 'prediction' by 'interpretation'

C2

- I. 60: replace 'have been' by 'are'
- I. 63: The previous sentence already started with 'although'- replace.
- I. 64: I would like to see a reference for this.
- I. 69: replace 'become' by ' may get' and add a reference for this statement.
- I. 79: the bodies are the biomarkers? Actually lipids, DNA or also pigments may be used as biomarkers. Not sure whether the word 'body' is really the right expression for dead microbial cells.
- I. 81: similar to what?
- I. 95: 'Hence' is contradictory to 'probably'
- I. 97: Which other markers? Why probably? This would actually be the place to explain the geochemical background.
- I. 100-104. This belongs to the methods part

#### Material and methods

- I. 110: A map of the sampling location would be beneficial.
- I. 138: Commonly, all abbreviations should be introduced when used for the first time.
- I. 146: this is a very low amount of DNA, how representative is this for community analysis? In how far is the quantification of OTUs and the genome identification trustworthy based on such low amounts of DNA? What about different degradation patterns of different organisms?
- I. 150: Is this a random amplification?
- I. 156: Which chemistry and protocol?

Some statistics on the sequencing (how many reads per run, how many of the identifiable, how many reads related to 16S rDNA, etc.) would be desirable.

C3

- I. 169: How?
  - I. 169: Based on the low amount of DNA and amplification steps in between, I doubt the quantitative aspects of the analysis.
  - I.175: remove gap after ). Rstudio will not make any visualization without a proper script, so please provide details on your script. Also, provide the correct reference instead of the link to the homepage.
  - I. 181: It is unclear how the draft genome was assembled. What is the completeness? A genome plot would increase the credibility.
  - I. 207: replace 'examine' by 'validate'
  - I. 223 MQ- please spell out
- I doubt that FISH produces a reliable signal if you only get 20pg of DNA out of the crystals.

#### Results

- I.232, 236, 238, 246, 249 : Check numbering of figures. Also, where do you refer to Fig. 5?
- I. 262: archaea, fungi without capital letters
- I. 265: Why is that surprising?
- I. 269 onwards: this needs a genome plot and a Kegg metabolic map.
- I. 278: I don't buy the quantitative aspect, here.
- I. 346: postulate seems to be a bit strong for the line of evidence provided, here.
- I. 346 ff: This information is what you need to put into the abstract to make it convincing

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Interactive comment on Biogeosciences Discuss., doi:10.5194/bg-2016-204, 2016.

C4