

## ***Interactive comment on “Mineral formation induced by cable bacteria performing long-distance electron transport in marine sediments” by Nicole M. J. Geerlings et al.***

**Anonymous Referee #2**

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The manuscript addresses bio mineralization associated with cable bacteria, a newly discovered group of filamentous bacteria within the Desulfobulbaceae family that performs electrogenic sulfur oxidation. Using a series of advanced microscopic techniques, the authors investigate the minerals formed within cells in the filaments or on the exterior of cable bacteria harvested from sediment-based enrichment cultures. The authors identify the presence of polyphosphates within the cells, the presence of external coatings composed by EPS and (probably) clay minerals; and the presence of external encrustations of iron oxy hydroxides. The findings are discussed primarily in relation to the eco-physiology of cable bacteria, which makes the paper relevant for the community of researchers dealing with these aspects. However, the work also

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addresses more general aspects of biomineralization, through the focus on model organisms, which through its peculiar metabolism has significant influence on many geochemical pathways. More over this organism has been shown to be abundant in many aquatic sediments and therefore I think that the study would be relevant and interesting also for readers of BG, that are not directly associated to cable bacteria research. In general, I find the manuscript well prepared, with (mostly) proper citing (see my point 3) and credit to related work. To my knowledge, the methodology used is sound and I find no reason to doubt the results and the interpretations of raw data. I therefore recommend publication of the manuscript.

I have a few suggestions and questions for the authors to consider. 1. When discussing of encrustation the authors refer to the LPS of gram-negative bacteria, as the cable bacteria are gram-negative bacteria. My question to this end is what is known about the composition of the outer membrane of cable bacteria i.e. the common membrane that encapsulates all individual cells in the filament? Is there any evidence that this membrane is composed from LPS? Note that we can easily imagine that the individual cells in the filament has both an inner and an outer membrane composed as for gram-negative prokaryotes, and that the common outer membrane is composed differently than from that?; Until more knowledge about the composition of the outer membrane is known, I do not think that the authors cannot make firm conclusions about the relationship between iron precipitations and the membrane properties and I encourage the authors to tone this discussion down. 2. In the discussion on the mechanism behind the formation of iron (oxy) hydrates encrustations on the cable bacteria the work of Otte et al. 2018 is used as a model for explanation of the crust formation. This model assumes direct electron transfer between cable bacteria and iron oxidizers present in anoxic sediment strata and as a consequence formation of iron (oxy) hydrates in the absence of oxygen. Perhaps this can occur but is really documented sufficiently well to be used as an explanation for the observation that some cable bacteria are covered with iron (oxy) hydrates? I do not think so. More over as I read the methods section, cable bacteria for this analysis were collected from the oxic zone of the sediments

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and there you do not need anything more than well known geochemistry to explain the formation iron (oxy) hydrates. So I suggest that the author tone down the more exotic explanations and choose the most simple model: that the iron (oxy) hydrates are formed through well-known reactions between O<sub>2</sub> and Fe<sup>2+</sup> in the oxic zone. 3. There are some references to unpublished work (e.g. Cornelissen. subm. ) and I suggest that these are taken out of the manuscript. In my view the information the Cornelissen. et al. subm. Paper, as cited in the manuscript does not contribute to an understanding of the data as it apparently deals with the internal structure of cable bacteria. Encrustation (the topic of the paragraph) is related to the external structure – i.e. the outer membrane. Please also be aware that all information related to this is sufficiently well described in the Pfeffer et al 2012 paper, and that the Meysman 2018 paper, which also is cited along the line of description of the cellular structures (l.453) does appear in the reference list. Here only Meysman 2017 appears and this is a review that does not add more information to the topic, than already described in the primary literature.

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