Biogeosciences Discuss.,
https://doi.org/10.5194/bg-2017-126-RC5, 2017

Biogeosciences Discussions
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Interactive comment on "Phosphorus limitation and heat stress decrease calcification in *Emiliania huxleyi*" *by* Andrea C. Gerecht et al.

Anonymous Referee #5

Received and published: 7 July 2017

General comments

The manuscript -Phosphorus limitation and heat stress decrease calcification in Emiliania huxleyi - by Gerecht et al. addresses and try to answer to an essential question in the coccolithophores community : How coccolithophores physiology will change in a environmental conditions changing ocean ? With this wider question in mind, this work focused on a more specific question : How Emiliania huxleyi will react to futur nutrient (P) and temperature conditions in ocean in term of calcite production, nutrient (P) requirement and morphology? The two different experimental methods used allows the comparison between the response of a strain of Emiliania huxleyi cultivated in a closed (batch) system and in semi-continuous cultures. This latter allow a determination of calcite and POC production and a lower nutrient limitation in contrary of a batch

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approach for which the calculation of production is impossible due to a decrease of growth rate and which led to a strong limitation of cells. The conclusions of this paper are mainly the negative feedback on carbon sequestration by the decreasing of cell density of Emiliania huxleyi in a warmer ocean due to a higher P-requirement under heat stress.

These both experimental approaches are a good way to have indication on the method dependence for a given response. Therefore, this work brings new insights on the response of Emiliania huxleyi to a P-limitation combined with a heat stress as well as new data to the literature known about Emiliania huxleyi physiology for both batch and semi continuous approach and for both P-limitation and temperature stress conditions. However, the choice of the cultures conditions, P limitation and temperature stress, need to be express with more details in the introduction or in the methodology part concerning the choice of the values. The paper is well written and has a clear and easy-to-read structure. This manuscript will fit perfectly to the journal Biogeosciences after few corrections and comments from reviewers. The figures and tables are well presented and show essential results that you discuss in this manuscript.

Specific comments

p. 2 ; Ln 9 : You need to add the word particulate to present for the first time in your manuscript the PIC and POC terms.

p. 3 ; 19 : What does inter alia mean?

p.3 ; Ln 29 & 35 : Did you test different temperature on the strain (before these experiment) to find the optimal temperature at the selected light dose (12 :12, 100μmol photons.m-^{2}.s-1) ? Or did you have an idea (or reference) of the optimal temperature of your strain before selecting your two temperature conditions ?

p.3 ; Ln 34-39 : What is the name of your strain ? Have you done some acclimations of your strain to the temperature and P concentration conditions before starting your

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experiment ? You should indicate in your method if you did. What is the initial nitrate concentration in your medium ? Is it the standard concentration of NO3 in a K medium ? If not, the value of the concentration need to be indicate in this method part. You also need to add the value of the salinity of your medium.

p. 5 ; Ln 21-26 : Did you take into account that Emiliania huxleyi can have multiple layers of coccoliths ? It is clearly visible on your figure 1 that your strain can have several layers of coccoliths depending of the cultures conditions. How did you take this particularity in your coccoliths counts ? What is the standard deviation on your counts ? At least, you have triplicate so you need to specify the standard deviation on your number. What about coccospheres diameter on your SEM images ? Did you think about doing coccopheres measurements ? If not, is it because of the high number of detached coccoliths on your filter ? It should be interesting to discuss the PIC content related to your coccolith number and the thickness of your coccolith layer.

p. 6 ; Ln 24-34 : Final cell density for batch cultures were being push really high in order to get the P-limitation. However, respect to LaRoche et al. 2010, inorganic carbon system changes need to be kept below 5% to avoid carbon system changing due to the high cell density. This meens that nutrient limitation experiment for batch system have to attain a reasonable final cell density in order to keep a DIC system quasi-constant through the experiment and to avoid a inorganic carbon limitation before the P limitation in this case. In your P-limited cultures, DIC changes that you get at the end of the experiment led to a low calcite saturation constant. Therefore observations in morphology and calcite content may be due to this changes rather than the P-limitation and heat stress effect. You took into account this changes in your discussion but it will be useful to indicate why you did batch experiments in this way. Could you justify this choice in the methodology of your batch experiment ? Did you think about carrying the experiment with a lower initial P concentration rather than push the cell density so high ? Or did you have a target initial P concentration that you wanted to test ? In this last case, a comment on the 2.1 part will be necessary.

p. 7 ; Ln 27 : What is the normal temperature? Even if you explain it later (p.8 Ln 26), you should describe here that the normal temperature is 19°C if you want to use this term. It is not obvious for readers.

p. 8 ; Ln 1-5 : same comments that previously on the DIC system

Figures & Tables

Figure 2 : You need to add the color explanations : blue is your schematic initial cell, red is the schematic effect observed at the end of your experiment.

Figure 4 : You should add the error bar on triplicate somewhere in your legend.

Tables 1, 2 & 3 : You should add the standard deviation in your legend and your n number (for example, n = 3 if it is triplicate).

Table 2 : You need to clarify in the legend of your table if the cell volume has been calculated with measurements of cell diameter from the harvest day or from an average of daily measurements.

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