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Interactive comment on “Influence of CO₂ and nitrogen limitation on the coccolith volume of *Emiliana huxleyi* (Haptophyta)” by M. N. Müller et al.

Anonymous Referee #2

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GENERAL COMMENTS

The sensitivity of calcite producing plankton to pCO₂ is an important topic within the current debate over the fate of oceanic carbonate production in a high CO₂ and warming ocean. Müller et al. present results from a series of culture experiments of a strain of *Emiliana huxleyi* under different pCO₂ levels and conditions of nitrogen availability (N⁻, N⁺). The focus of this manuscript is on changes in the size of the cell, coccosphere and coccolith under these conditions. It is worth highlighting that such (culture) studies are not only the remit of palaeo-oceanographic research, but of significant importance and relevance to the interpretation of trends in present-day coccolithophore

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populations.

Missing from the article (e.g. abstract) is a significant conclusion that can be drawn from Figures 2 and 3 (scatter plots of pCO₂ and coccosphere/cell/coccolith diameter): the sensitivity of cell/coccosphere/coccolith to nutrient (nitrogen in this case) availability is much greater than the sensitivity to pCO₂. For example, coccolith volume (um³) changes from 0.76-0.89 under nutrient deplete conditions to 2.09-3.43 under nutrient replete conditions (Fig. 3, Table 4). This observation should be included in the abstract and conclusions. As ocean acidification research continues it is worth noting the relative sensitivity of different processes to multiple stressors, especially when we are dealing with a multivariate ocean environment.

The 3-4 fold differences in coccolith volume (and estimated mass) and calcification rates between nutrient treatments (2-2.4 vs. 5.7-9.3 pg CaCO₃ coccolith-1 and 2-4 vs. 12-21 pg C cell-1 d-1, respectively) is another very interesting observation. Such modifications of cellular levels of calcite and coccolith production rates have implications for how we interpret the mass of individual coccoliths (either in the modern ocean or fossil record). At this time there are relatively few observations of such a trend (i.e., changes in coccolith mass under different environmental conditions which are independent of changes in species/morphotype). Following on from this, the authors should include a few SEM images from the different nitrogen treatments to support this finding (i.e. showing large (>4.5 um) coccoliths in the nutrient replete cultures) and also indicating the level of malformation in the culture experiments.

SPECIFIC COMMENTS

1. Role of irradiance. In terms of determining growth rates of oceanic populations, light availability is as important as nutrient supply/availability and the introduction should better reflect this.
2. Cell quota Although it may be obvious, there is no mention in the methods or results of how the cell quota (of PIC, POC, POP, TPN etc) was calculated. This is a central

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part of the calculation of various parameters (including calcification rates) and it would be good if the authors commented on how it was performed and the possible errors associated with it.

3. SEM images The SEM images are a key part of this manuscript, and although no quantitative analysis is possible (due to preservation problems?), a few representative images would support various conclusions. This is especially important in the large scale differences in coccolith volume (driven by differences in coccolith size, as concluded by the authors) between nutrient conditions/pCO₂ - images showing >4.5 μm coccoliths would support their observations. The images (Fig. 5) are difficult to interpret from only the pCO₂ treatments. Images from the nutrient replete and deplete treatments are needed.

4. Cell shrinkage The authors used an acid treatment to dissolve off the coccoliths from the cells and then measured the cell size. How did they know that (a) all the coccoliths dissolved, and (b) there was no associated shrinkage of the inner organic cell with acidification?

5. Treatment of POC filters Although minor, it would be good for future reference to know how the GFF filters for POC analysis were "treated with HCl" - rinsing? a few drops? overnight fuming? What concentration and what was the duration. There are several different methods of removing the PIC from filters for POC analysis.

6. Changes in cell diameter and coccosphere diameter On pg 4990, lns 3-5: These values are averages and although the standard errors are reported in the Table (Table 4) it would be useful if they were reported in the text as well. This makes it clearer to the reader that these are significant differences.

7. Morphotype E Following Young et al. (2003), what is morphotype E? In Beaufort et al. (2011) this is referred to as morphotype R. Changing terminology is confusing.

TECHNICAL CORRECTIONS

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Generally the article needs proof reading for grammar.

Abstract – best studied or most studied?

pg 4981, ln 4 – missing "as" between "referred to" and "ocean carbonation/acidification".

Table 4 – use correct terminology for final column, "volume of coccoliths".

Correct spelling of Young in Poulton et al. (2011) reference.

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