

Interactive comment on “Effects of long-term high CO₂ exposure on two species of coccolithophores” by M. N. Müller et al.

Anonymous Referee #3

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Global warming and ocean acidification are considerable concern to all people. Long-term exposure experiments of coccolithophores held by authors are worthy approach for understanding the response of marine calcareous phytoplankton against ocean acidification. However, I do not understand why authors adopted ‘sub-continuous batch culture’ for this research. Authors wrote that they controlled pCO₂, but not mentioned how they controlled nutrient concentration for 66 and 98 days. So here I write my following comment under the understanding that authors did not control nutrient concentration during their experiments.

In my understanding, physiological and morphological experiments of coccolithophores are usually done while the culture strain is in exponential growth phase to collect healthy cells, and many papers reported culture of *E. huxleyi* reaches to stationary

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phases within 7–15 days in their experiments.

In this paper, authors wrote that nitrate limitation did not occurred during 98-days experiments, but it seems unusual that culture conditions including nutrient concentration were satisfactory through 98-days-long experiments if the strain is enough healthy. Authors wrote that cell density of culture, which has strain started from 100 cells per 0.1 ml, never reached to 5×10⁸ cells per L(-1) level in their 98-day experiments. If nutrient concentration was enough, this result suggests another unknown factor limited healthy exponential growth of the strains.

In my opinion, continuous culture system with nutrient control was more appropriate for the purpose of this study. Or, did authors control other factors (nutrient etc.) in this study although it was not mentioned in the method? If it is so, I should change my comments.

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