

Interactive comment on “Effect of CO₂ on the properties and sinking velocity of aggregates of the coccolithophore *Emiliana huxleyi*” by A. Biermann and A. Engel

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Response to Anonymous Referee #2 (Referee comments in quotation marks)

We thank the reviewer for his/her thoughtful comments on the general relevance of perturbation/ acidification studies and also for his/her compliments on the technical performance of this study. We agree that the issue of applicability of results from experiments simulating a future ocean scenario to the real future ocean is too often neglected. One obvious reason is that we know little about the real future, including the potential of organisms to adapt to specific changes such as acidification. But we can investigate the mechanisms how a changing environment may alter processes in the ocean. In

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fact, this is what we did during this study, i.e. to investigate how acidification-induced changes in the degree of calcification could alter the physical properties of aggregates. The effect of ballast on aggregate formation and sinking is not straightforward as several non-linear processes are involved. To our current knowledge it is unlikely that calcification will cease completely in the future as long as calcite under-saturation does not occur. Therefore, we need to know how sensitive aggregate properties are to changes in the degree of ballasting. Therefore this study clearly differs from the earlier experiments by Engel et al. (2009). We will emphasize this difference between the studies in the revised version.

Referee comment: “Plopping unadapted organisms into a high CO₂ environment...does not test their response to a high CO₂ ocean...but rather measures their response to stressful environmental conditions.” “Acclimation for 13 days, as done in this paper, is not enough for adaptation” Response: Adaptation refers to the genetically response of organisms to environmental conditions, whereas acclimation describes the cell’s physiological response. Whereas 13 days, roughly 13 generations (~ 7 generations until stationary phase) are surely not enough to include a potential adaption to changes in pCO₂, it has been suggested to be enough for the acclimation of cells (see LaRoche, J., Rost, B. and A. Engel: Bioassays, batch culture and chemostat experimentation. In: Guide to best practice in ocean acidification research, edited by Riebesell, U., Fabry, V. J. and Gattuso, J.-P., submitted). In previous studies, generation times of *E. huxleyi* ranged between 7 and 12 generations (Riebesell et al. 2000, Zondervan et al. 2001, 2002). Müller et al. (2009, BGD) found that a gradual increase in CO₂ concentration up to 1150 μ atm for *E. huxleyi*, grown over 152 generations, did not change the cell’s sensitivity to CO₂ or pH, particular the changes in PIC/POC ratio, compared to the shorter termed response. Barcelos e Ramos et al. (2009, BGD) report for *E. huxleyi* of a calcification decrease and organic carbon fixation increase with increasing CO₂ concentrations already within 8 hours. The authors conclude that the (physiological) acclimation state is a matter of hours only.

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As far as we know, there have been no experiments identifying time scales of adaptation of *E. huxleyi* to pCO₂ so far, or whether or not the cells will adapt at all. Therefore, it is not known whether future adaptation in situ would enhance or weaken the responses identified in shorted termed experimental studies. We agree that more work has to be done on the adaptation potential of coccolithophores in Ocean Acidification experiments. However, including timescales of adaptation was beyond the scope of this experiment. We will include these aspects in the revised discussion.

Referee comment: “Phytoplankton under stress from many causes often produces more exudates material (polysaccharides) that would generate more mucous, fluffy aggregates.” Response: The experiments were carried out after a sufficient acclimation time (see above). We therefore do not assume that changes in PIC/POC ratios are the result of a stress reaction, in the sense of a sudden change of environment. Moreover, fluffy aggregates were not observed in the low CO₂ treatment.

Referee comment: “...differences in the composition and characteristics of coccolithophorid aggregates with varying amounts of calcified and non-calcified *E. huxleyi* ...has already been published (Engel et al. 2009).” As also pointed out in our response to reviewer #1, Engel et al. (2009) did not study the effect of different CO₂ concentrations on aggregate properties on *E. huxleyi*, nor of varying amounts of calcified cells. They investigated aggregate formation of totally calcified and completely naked *E. huxleyi*.

Specific comments: We will improve the text according to the reviewers’ suggestions, e.g. include a reference for lower calcification rates of coccolithophorids and clarify the sections concerning the decreased calcification or probable dissolution of calcite for naked cells.

Referee comment: Methods: “...began aggregation experiments 7 days after the start of the stationary phase.” “... also unnatural,...the high concentrations of coccolithophores...above expected concentrations in nature.” Response: We agree

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that due to experimental constraints the cell concentrations and numbers of aggregates per litre volume were above what is reported for in nature. This, however, does not disprove the observed changes of individual aggregates. Moreover, the results of the present study are in line with previous studies, with regards to decreased calcification and enhanced organic carbon production in cells when increasing CO₂ concentrations for *E. huxleyi* (Barcelos e Ramos, Müller, Zondervan, Riebesell). Those experiments were conducted with lower cell densities.

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