

Interactive comment on “High tolerance of protozooplankton to ocean acidification in an Arctic coastal plankton community” by N. Aberle et al.

Anonymous Referee #2

Received and published: 4 December 2012

Review of Aberle et al. 'High tolerance of protozooplankton to ocean acidification...' submitted to BGD

The work described in the manuscript focus on potential effects on ocean acidification on heterotrophic dinoflagellates and ciliates. Emphasis is given to estimates of direct effects on community/diversity and their implications in stoichiometry/phenology/carrying capacity. The topic is of great research interest, especially as the response is addressed in a more complex mesocosm environment. The authors found little evidence for any major effects and conclude that protozooplankton is relatively insensitive to acidification. Although I can follow this interpretation, I find the

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data basis quite poor. Addressing the raised questions requires a frequent sampling and thorough estimate of sampling variability, which is not given here. Often the interpretation is based on only 1-2 sampling points during one of the 3 bloom phases and conclusions of 'increasing'/'decreasing' or 'peak biomass' are made without knowledge of sampling variability. This is highly critical considering the low abundance of protists and its inherent counting variability. I have my doubts if any quantitative conclusions can be made at all. Statistics regarding quantitative implications are completely lacking.

Apart from this principal problem I have with the manuscript, there are many details to be improved (in case the manuscript is accepted):

The introduction is biased towards negative impacts of OA on organisms, despite a large body of literature suggesting at least the opposite (e.g., Hansen papers). The two hypotheses read nice, but it remains unclear how the authors can find separate answers to them. Stoichiometric changes are not addressed at all later (hypothesis 2). The material and methods lack some considerable detail about sampling and analysis. The discussion lacks critical evaluation of the sampling scheme and lacking estimates of variability. In addition, the discussion on food web aspects (top-down, food quality) is very speculative as no data is presented that would support conclusions. In addition, the authors also need to discuss the new data in the context of interactions between food size, food abundance and food quality which is presently not the case and – as I feel it – beyond the focus of the manuscript. Besides this, important literature on trophic upgrading and sources of fatty acids in the food web are ignored.

Detailed comments:

Introduction:

P13033 line 1: This is not logic: Open ocean plankton communities are expected to be more vulnerable due to larger variations in pH in coastal areas, so why you argue then that 'therefore, one of the central questions...was whether arctic coastal

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plankton is vulnerable...'. What is the rationale behind the investigation when coastal plankton is likely to experience large variation in pH? In addition, Hinga's review (2002) emphasizes the importance of high pH in causing the large variability in coastal areas.

P13033 line 5: Protozooplankton is used synonymously with heterotrophic protists and protozoa, which is confusing, similarly to the reduction of protozooplankton to ciliates and heterotrophic dinoflagellates. What about mixotrophic protists, which are part of the PZP? Are they excluded from the analysis?

P13033 line 13: The description of potential effects on plankton is considerably biased towards negative effects. Apart from the cited literature, there is quite some literature available that clearly points out that many groups may not be affected. These should be cited and a more balanced description of potential implications for OA effects should be given.

P13033 line 20: The two hypotheses appear clear on the first look. However, one wonders whether and how these hypotheses can be separately addressed and analyzed in a complex mesocosm experiment. Furthermore, is there any evidence that changes in phytoplankton stoichiometry can alter phenology (in addition, no data on stoichiometry is presented here)? The knowledge or evidence underlying the second hypothesis is not adequately described in the introduction. In addition, large protists can be subject to considerable predation by larger zooplankton, the potential interaction of altered prey community and mesozooplankton grazing pressure needs to be addressed in such a complex set-up. Furthermore, changes in stoichiometry are not addressed later at all.

P13034 line 1: Apparently, the experiment was started after the spring bloom. Although this is not in the primary focus of the experiment, have the authors considered a potential bias by the fact that the pelagic community already went through a spring cycle associated with a pH shifting from low to high? A considerable decrease in pH due to manipulation after the bloom is uncommon and might influence the protist composition.

Methods:

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P13034 line 14: How were the mesocosms filled, by pumping water into them? This is important to know (potential disruption of delicate organisms).

P13035 line 4: If I understood correctly, the authors want not only to study potential changes in the community composition, but also in phenology. How can both be addressed when samples were taken only once per week? Ciliate blooms can be highly ephemeral (Moritz et al. 2006, Fileman & Leakey 2005, . . .), so part of the development is missed.

P13035 line 4-25: The sampling lacks many details. At which depths were water samples taken? Volume per depth? Total sample volume? Since only heterotrophs are addressed in the study, how were mixotrophs separated? Is this possible at all? Some of the taxa included in Table 1 can be mixotrophic (*Laboea strobolia* for instance)! Any real subsamples? The analysis lacks details on counting procedures and numbers of counts per taxonomic group. Literature is missing in the references.

P13036 line 4: Again, details are missing: where *Chla* samples also taken from integrated water samples?

P13036 line 10: The conducted test doesn't appear appropriate for the purpose. Samples were taken only once per week. Any small-term shift in the timing of development of the protists (phenology) smaller than 6 days is not recognized; in addition, how are maximum concentrations defined (compare results)? The grazing pressure by mesozooplankton might be different between mesocosms, potentially influencing composition, abundance and timing as well. Because of these interactions, one cannot simply compare samples taken at a time point x, but need to compare the dynamic development in the different mesocosms. The regression analysis needs to be specified.

Results:

P13036 line 15: It would be helpful to see the pH/CO₂ results as this is central to the understanding of any effects. From the material and methods, it is not clear whether the

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pH was manipulated only at the beginning of repeatedly. In Figure 1, PZP biomass bars should be confined to the day of sampling instead of covering several days; symbols don't fit with legend. Arrows could indicate timing of events (e.g., nutrient addition).

P13037 line 1: Is this trend significant? Is there an opposite trend in the second bloom?

P13037 line 9: This is a bit contradictory: Dinoflagellates dominated the 3rd bloom, with higher levels at higher CO₂. In contrast, the Chl_a was higher at lower CO₂ in this phase. What makes the difference?

P13037 line 13: The counts for different Mesocosms reveal a 3 fold difference in the t₀ values for PZP, directly after filling. This can influence the response to manipulation. In addition, I miss estimates of within mesocosm variation. This puts statements as 'decreased' or 'increased' into doubt. Table 1 provides a detailed list: does it contain all taxonomic groups, or what about unidentifiable groups such as ciliates? The biomass is partly low, and one wonders on how many cell counts the biomass estimate is based? Which test has been used for correlation analysis?

P13038 line 13: The description of 'response in biomass' and 'succession' is based on the 'analysis of maximal two consecutive samples without estimates of within mesocosm variability. Conclusions as 'strongest positive biomass response' and 'peak biomasses' are doubtful with regard to the sampling frequency. How do the authors know when peak biomass was achieved?

Discussion:

I miss a general discussion on the limitation of the sampling scheme to identify responses of heterotrophic dinoflagellates and ciliates to OA. Quite important appears also that the experiment was apparently started following the spring bloom, a phase in which the pH of the system has likely been increased due to biological activity. P13039 line 22: When the heterotrophic dinoflagellates and ciliates are undoubtedly sandwiched between primary producers and predators, should the dynamics of these

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organisms not be evaluated in response to both pressures? I assume abundant zooplankton was around, which consume dinoflagellates and ciliates.

P13040 line 3: Before judging 'that no direct effects on PZP composition and diversity were observed' authors should analyze if the set-up was suitable to answer this question with regard to sampling frequency and analysis. Still unclear to me is the actual values of pH during the experiment, have they been drifting or were pH levels kept constant? This of course has implications for the interpretation.

P13041 line 1: The conclusion on lacking effects on carrying capacity and phenology are invalid considering the sampling frequency. How do the authors define maximum biomass and what are the appropriate temporal scales for analyzing phenology?

P13041 line 12: The statement of a strong decline contrasts with the biomass described for the first two sampling events in Fig 3, which were the dominant species. In addition, lacking estimates of the biomass variation within each sampling date make the conclusion doubtful. Many of the groups had a low biomass, and considering their large size, thus a low abundance, suggesting a potentially high variability is inherent in the methodology.

P13042 line 1: Instead of speculating about potential top down control, authors might consider to calculate the potential pressure from the abundance of zooplankton which is available. (Niehoff et al.)

P13042 line 18: The original literature for trophic upgrading should be cited. For both top-down control and trophic upgrading no data is presented and the discussion here is very speculative. Why should food quality of autotrophs decrease as inferred here? Considering the composition of the protists, increasing zooplankton egg production might simply results from the changing size distribution. I find this very speculative here, as no data is presented.