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# Interactive comment on "Temporal biomass dynamics of an Arctic plankton bloom in response to increasing levels of atmospheric carbon dioxide" by K. G. Schulz et al.

## K. G. Schulz et al.

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We thank both anonymous referees for their positive and constructive reviews which will significantly enhance the scientific quality of our manuscript entitled 'Temporal biomass dynamics of an Arctic plankton bloom in response to increasing levels of atmospheric carbon dioxide'. Below please find our comments to the more general and then specific issues raised by the referees.

### **General Comments**



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**1.** 'The intro needs more background information... The authors reference one review paper on calcifying organisms, but do not give credit to previous studies that have directly examined phytoplankton community responses...And the authors need to end intro with justification for the research.'

We will add more background information, giving credit to several previous studies investigating the effect of ocean acidification on phytoplankton community structure, especially of non-calcifyers. We will also clarify the research justification in the introduction.

**2.** 'This manuscript needs a concluding paragraph, summarizing their results in the context of the breadth of knowledge of OA impacts on phytoplankton community structure...'

We will add a concluding paragraph, summarizing the main findings on direct and indirect effects of  $CO_2$  on marine phytoplankton and potential implications for future food-web structure and biogeochemical element cycling.

#### Specific comments of referee #1

**1.** What is the rationale for adding 100, 20 and 70 individuals of pteropods at day 4, 5 and 6?'

Pteropod occurrence in the fjord is extremely patchy and depends on wind and current conditions. Thus, it took several attempts and three collection days to find sufficient numbers. We will add information on that to the text.

**2.** 'The estimation of seawater volume with salinity increases should be further explained using an equation (an example).'

We will be more specific.

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3. 'For nutrient addition, 50  $\mu$ m filtered seawater might have contained phytoplankton of different amounts for each addition or mesocosm. How was this overcome to avoid the unexpected effects?'

Since we adjusted the amount of nutrient stock solution to be added, ranging between 21.95 and 23.78 kg per mesocosm, depending on actual volume determined by the salt addition, exactly the same amount of nutrients and also biomass/phytoplankton were added to each liter of mesocosm water. Furthermore, the dilution factor was about 1:2000, limiting a potential effect of added phytoplankton on community composition inside the mesocosms.

**4.** 'POC filters were treated with fuming HCl in a desiccator for 2h before drying and analysis, is this exposure to HCl fume long enough?'

Tests with pure coccolithophore cultures with a particulate organic to inorganic carbon ratio of about 1, i.e. much more calcite in relation to particulate organic carbon than found in most oceanic regions, showed that already half an hour of exposure to fuming HCl is enough to remove all calcite from the filters. With time and use the fuming acid 'ages', losing its ability to quickly and completely remove particulate inorganic carbon. That's why freshly-prepared acid was used.

**5.** Phase II and Phase III apparently showed differential impacts of elevated  $CO_2$  on biomass density in terms of ChI a. As discussed, the authors attributed this mainly to different responses of different groups, such as Micromonas-like phylotypes, to elevated  $CO_2$ ... Given the fact most of the phytoplankton species investigated so far operate CCMs, focusing on Micromonas alone seems unfair.

Mass balance considerations from typical phytoplankton ChI a to carbon relations and absolute ChI a biomass of different phytoplankton groups show that a significant portion of the phytoplankton biomass during phase II was in prasinophytes, especially at higher  $CO_2$  levels. Thus, the question is why did they profit? The reasoning behind our line of

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argument is that since probably all phytoplankton species employ some sort of carbon concentrating mechanism (CCM), the ones to profit from higher  $CO_2$  availability are most likely those with the least efficient CCM. The smaller a cell, the less it obviously has to invest to actively concentrate  $CO_2$  at the site of carboxylation because of more favorable surface to volume ratios and reduced diffusive boundary layer in comparison to bigger cells. We will clarify this in the revised version of our manuscript, but also see our answer to comment #17 of referee #2.

**6.** 'Accumulated energy inputs ... into the mesocosms, over Phase II and III, seemed to differ a lot (by up to 30%?? higher in Phase II). The total energy inputs of Phase II and III should be given and discussed. Interactions of light energy with CO<sub>2</sub> and nutrients might be the key reason responsible for the community structure change and primary production.'

Indeed, average daily light intensity (PAR) was 586 during phase II in comparison to 392  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup> during phase III. We will add this information to figure 4. We also agree that changing amounts of incident light energy might interact with other factors such as nutrient availability, grazing and viral pressure, and species interaction to ultimately shape observed phytoplankton community assemblage dynamics. We will discuss this in more detail in the revised version of the manuscript.

7. 'It is a nice approach to compare dynamics in the fjord and that of the mesocosms... but I feel hard to be persuaded in the direction that light or energy input is identical. In view of the low transparency of the material (polyethylene??) used for the mesocosms, daily or phase-integrated light energy inputs to the mesocosms must be much lower than that in the fjord. Since it is easily be estimated using the light attenuation factor and transparency of the bags, how vertical profiling of light or total energy input would differ inside from that outside the mesocosms it should be provided.'

Light attenuation coefficients (kd) were calculated for each mesocosm and the fjord on

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ten days, by fitting an exponential function,  $f(x) = e^{(-kd x)}$ , to the measured vertical light profiles. The percentage of light reaching a certain depth in comparison to the surface was nearly identical between mesocosms and the fjord. While lowest values were found on day t4 at about 0.23, kd ranged between about 0.30 and 0.40 from then on.

Furthermore, apart from light attenuation, also absolute amounts of PAR must have been quite similar between the fjord and the mesocosms. On day t27 kd was about 0.37, meaning that PAR at 3.7 m depth should be about a forth of incident light. And indeed, during the continuous light measurements inside mesocosm M1 for 40h on the following day four to six times less PAR was measured at 3.7 m water depth in comparison to direct measurements at air. The observed variability was probably connected to variations in cloud cover and solar elevation. Thus, it seems that the energy input into the watercolumn of the fjord and the mesocosms are quite comparable and that shading by the thermoplastic polyurethane bags and the dome-shaped hoods of the mesocosms are lower than one might expect. We will include this information in the revised version of the manuscript.

#### Specific comments of referee #2

1. 'Name study location in the abstract'

Will be done.

**2.** 'Page 3: "Therefore, the Arctic is projected to be the first ocean region to become under-saturated..." I am most familiar with the Orr et al. (2005) model predictions, which suggest that undersaturation will occur in the Southern Ocean first due to the low existing saturation levels of carbonate minerals, uniform temperatures and the extent of mixing in the water column. Please clarify here.'

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The models used in the 2005 study by Orr et al. did not include the Arctic Ocean above 60 degrees North. Thus, they projected undersaturation to occur in the Southern Ocean first, in the year 2050. The model study by Steinacher et al. (2009) includes the Arctic Ocean and projected undersaturation to occur already about 20 years earlier in the Arctic than in the Antarctic. We will clarify this in the revised version of the manuscript.

**3.** 'Page 3: "ocean acidification can cause aragonite under-saturation already today (Bates et al., 2009;.." I suggest changing "can cause" to "has caused".'

Will be done.

**4.** 'Page 4: Again, name study location here or at least a more specific one than the current "in the Arctic".'

Will be done.

**5.** 'Page 5: "Adult pteropods.." Why did you add different numbers of pteropods and why on those specific days?'

See answer to comment #1 of referee #1.

**6.** 'Page 6: "t-4 and t4..." It would be helpful to add in a one sentence explanation regarding the timeline of your experiment at the beginning of the methods section and move Fig 2 up as well, to aid in understanding of your labeled days "t-4" vs "t4" for example.'

Will be done.

7. 'Page 7, Section 2.3: You need to, early on in the methods, state which mesocosms served as controls. I assumed it was the 2 lowest  $pCO_2$  mesocosms, but I didn't see

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this stated until results section 4.2 (discussion)... Additionally, when I think of a "control" in OA studies, I typically think of somewhere near average ambient atmospheric  $pCO_2$  (380- 390  $\mu$ atm), but the controls in this study were at significantly lower CO<sub>2</sub> levels...'

We will add more information on the control treatments to the methods section 2.3 and also  $pCO_2$  and pH levels of the fjord to table 1. We will also discuss the relatively low control treatments in more detail. Basically, they reflect post-bloom conditions, typical for Kongsfjorden at this time of the year.

**8.** 'Page 7, Section 2.3: What is the justification for the chosen  $pCO_2$  levels? And why a gradient and not replicates of certain preselected  $pCO_2$  levels?'

With the relatively low amount of possible replicates statistical power of linear regressions are the same, if not superior, than compared to ANOVA based analyses. Also, a gradient approach is less vulnerable to the potential loss of one or two mesocosm units in comparison to a replicated design. There are more advantages, nicely summarized in Havenhand et al. (2010). The initial  $pCO_2$  were chosen to cover with seven out of nine mesocosms levels projected until the end of this century. As primary production and air/sea gas exchange at  $pCO_2$  levels higher than those in the atmosphere shift carbonate chemistry speciation towards lower  $pCO_2$  and higher saturation states with respect to aragonite, the two highest  $pCO_2$  levels were chosen to keep two treatments at the end of the experiment still under-saturated with respect to aragonite. Will will include these aspects in the revised version of the manuscript.

**9.** 'Page 7, Section 2.4: What is the justification for these specific concentrations of nutrients?'

The nutrient addition was meant to simulate the upwelling of deeper, nutrient-rich waters to a nutrient depleted surface. Phosphate concentrations at about 30 m depth in the fjord were measured at about 0.6  $\mu$ mol kg<sup>-1</sup>, about twice our addition. The other nutrients were scaled according to Redfield proportions. We will add this information

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to the revised version of the manuscript.

**10.** 'Page 8, Section 2.5: "water samples were brought back to shore and stored at in situ water temperature in the dark until processing..." State the length of time between sampling and processing.'

Sampling was between 9:00 and 11:00. Depending on parameter, sample processing was within an hour after samples had been brought ashore. We will add this information to the revised version of the manuscript.

**11.** 'Page 16, Section 2.5: "While silica concentrations in phase II displayed a statistically significant negative correlation to CO<sub>2</sub>, those of biogenic silica were positively correlated (compare Fig. 7A and B, and Tab. 2)." Is silica actually supposed to be silicate?'

Yes. Will be changed.

**12.** "DOC, starting at about 70-80  $\mu$ mol I<sup>-1</sup> in all mesocosms, increased before nutrient addition during phase 0 and I, resulting in higher concentrations at higher CO<sub>2</sub> in phase II." Did this affect total alkalinity?'

The DOC increase was too small to affect alkalinity. The measured increase in alkalinity of about 10  $\mu$ mol kg<sup>-1</sup> (compare Bellerby et al. in the same special issue) until the end of the experiment can entirely be explained by nutrient utilization and evaporation. We also realized that we had presented and analyzed only half of the measured DOC values. This will be changed in the revised version of the manuscript. Note that now there will still be a tendency towards higher DOC concentrations at higher CO<sub>2</sub> in phase II, although, not statistically significant.

**13.** 'Page 20, Section 3.8: "An exception were prasinophytes and dinoflagellates, important contributors to autotrophic standing stocks in all mesocosms during phase II

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and III, having insignificant contributions in the fjord during this time." Why the discrepancy between mesocosm and field?'

Here we can only speculate. Because of significant water mass variability in the fjord, as seen by significant changes in salinity, we would have expected much more pronounced differences between phytoplankton assemblages in the fjord and in the mesocosms in the first place. Also, there was significant meltwater input, especially towards the end of the experiment, reducing salinities in surface waters significantly. Thus, it came as a surprise to us that general bloom patterns were quite similar between the fjord and the mesocosms. The question should be re-phrased, not asking why there was a discrepancy between mesocosm and field but rather why there was quite some similarity. We will discuss this in more detail in the revised version of the manuscript.

**14.** 'Page 20, Section 3.9: " During phase III carbon biomass by diatoms was higher at lower  $CO_2$  levels, a trend found to be statistically significant." What kind of diatoms? Were they bloom-forming species? Why did diatoms not respond to higher  $CO_2$ ? Low nutrients?'

Yes, this was a pure nutrient effect, or an indirect CO<sub>2</sub> effect as detailed in the discussion section. The dominant diatoms were chain-forming. SEM analyses are under way for species identification and information will be included in the revised version of the manuscript.

**15.** 'Page 20-21, Section 3.9: " An exception were autotrophic dinoflagellates with insignificant and chrysophytes with higher carbon biomass in comparison to the meso-cosms at certain times." Again, why the discrepancy between mesocosm and field?

See answer to the referee's comment #13.

16. 'Page 21, Section 4.10: "REALTE" should be "RELATE".'

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#### Will be changed.

**17.** 'Page 25, Section 4.2.1: "However, its relatively small size (less than 2  $\mu$ m in diameter) could make the extensive operation of active CO<sub>2</sub> and HCO<sub>3</sub><sup>-</sup> uptake, like in most bigger phytoplankton species (compare e.g. Giordano et al. (2005) and references therein) unnecessary, as the diffusive boundary layer can be considered relatively small (Riebesell et al., 1993)." This is the opposite of what is suggested by Tortell et al. (2008) who states: Larger chain-forming Chaetoceros species may be at a competitive disadvantage for C uptake under low CO<sub>2</sub> conditions which induce an upregulation of cellular C transport (Figure 1), and favor small cells such as Pseudonitzschia with high surface area to volume ratios." This needs to be addressed, and I am genuinely interested in your thoughts on the matter. I suggest you include your reasoning for the opposing ideas.'

This is actually a very interesting point. Terms such as small or large, however, are always relative. Although Pseudo-nitzschia is smaller than Chaetoceros, both are much larger than what we refer to as small cells (smaller 2  $\mu$ m in diameter). In case that both diatoms possess a similar carbon concentrating mechanism (CCM), the smaller one could have a competitive advantage at low CO<sub>2</sub> concentrations as suggested by Tortell et al. 2008. If, however, cells become so small that carbon supply can at least be partly met by diffusion without active carbon transport, what could be considered an inefficient or poorly-regulated CCM, then those small cells could benefit from higher CO<sub>2</sub> levels. And although Pseudo-nitzschia is smaller than Chaetoceros, it most likely doesn't belong to that category, also evident from high bicarbonate to carbon dioxide uptake ratios. In other words, only those species which are currently not able to saturate photosynthesis might directly profit from increasing seawater CO<sub>2</sub> concentrations. And this could be the very small ones (smaller 2  $\mu$ m) as suggested here and the very big ones as suggested by Tortell et al. (2008). We will include such a discussion in the revised version of the manuscript.

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**18.** 'Page 26, section 4.2.2: "During phase III of the experiment, after termination of the second bloom by viral infection (see Brussaard et al. (2012) for details)..." Even though you cite Brussaard here, this definitely needs expanded upon because it directly relates to your results. Did you add viruses to the mesocosms or were they natural viral infections. Did the infections affect all phytoplankton taxa similarly?'

No additional viruses were added to the mesocosms. Viral infection is actually highly host-specific and also depends on virus and plankton abundances. Flowcytometry identified two distinct virus clusters of which one correlated with the decline of the dominant nanophytoplankton in phase I and the other with the dominant picophytoplankton in phase II. We will elaborate on this in the revised version of the manuscript.

**19.** 'Page 26, section 4.2.2: "This is most likely an indirect  $CO_2$  effect as after the collapse of the second bloom in phase II, more inorganic nutrients were available at lower  $CO_2$  concentrations (compare Figs. 6B and D)." If there were inorganic nutrients in the lower  $CO_2$  mesocosms in phase II, then why did the second bloom collapse? Was it due mainly to viral infection?'

Yes (see also answer to comment #18 above). We will mention this in the revised version of the manuscript.

**20.** 'Page 26, section 4.2.2: It would help to have some background ecological information about this fjord to help interpret some of your results regarding phytoplankton community composition. Are the small phytoplankton species you saw during your manipulation typical? Or in contrast are there typically large diatom blooms in this fjord, but are maybe more prevalent during times of higher nutrient concentrations? Are the diatoms you saw bloom forming species? I am just trying to wrap my head around why higher diatom biomass would occur under lower  $CO_2$  conditions.'

Usually a quite typical spring bloom, dominated by diatoms, develops in Kongsfjorden. Afterwards, into the summer season, the phytoplankton community becomes 9, C6222–C6234, 2012

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more diverse and dinoflagellates, cryptophytes and flagellates, sometimes described as 'small', dominate the community (Hop et al. 2002, Halldal & Halldal 1973). While Keck et al. (1999) reported on the occurrence of unidentified flagellates smaller than 3  $\mu$ m, it is not clear whether the abundant 'small' flagellates described in previous studies belong to the group of prasinophytes as identified here. However, the general pattern of phytoplankton species composition after the spring bloom in our study seems to be typical for this time of the year. Higher diatom derived biomass in phase III of the experiment is most likely the result of an indirect CO<sub>2</sub> effect. See also our answer to comment #14. We will add some more back ground information on phytoplankton species composition in the fjord to the revised version of the manuscript.

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