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Biogeosciences Discussions

Interactive comment on "Effects of CO_2 on particle size distribution and phytoplankton abundance during a mesocosm bloom experiment (PeECE II)" *by* A. Engel et al.

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- Use of size information from Flow Cytometry: The referee asks why we did not make use of the size information gained by flow cytometry. Flow cytometry gives size information based on the light scattering of particles. This can in principle be used, after calibration, for size information. However, for calcifying cells such as Emiliania huxleyi information on absolute size can be prone to error since calcification affects the scattering of the cells. Moreover, variations of absolute size of particles have been observed for Flow Cytometer data when measurements were performed over a longer time period, i.e. several days, since size information may change slightly with the flow rate of the sample and sheath fluids (Davey et al. 1993). Therefore, we regard Coulter Counter information on absolute size as more robust.

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- Potential underestimation of the abundance of large particles: Our particle size analysis was based on a triplicate volume of 2ml to determine particle abundance of up to 60 μm ESD. Particle abundance above 10 μm was very low, above 20 μm negligible. However, in order to test whether we missed larger phytoplankton by using a small subsample or an orifice of 100 μm, we determined the abundance of larger particles during the experiment by using a 280 μm orifice (giving a size range of 6.5-168 μm ESD) and a triplicate volume of 20ml; thus counts of a total of 60 ml were used for calculating average counts per ml per μm. The results of these measurements confirmed our observations with the 100 μm/2ml approach and were therefore not included in the present manuscript. In the revised manuscript we now state that larger particles were determined occasionally using a 280 μm orifice, and that these measurements showed no significant abundance of larger particles ($>$60 μm ESD). Although we cannot exclude that phytoplankton cells $>$ 168 μm ESD did occur during the study, we assume that it is unlikely that these cells contributed significantly to the phytoplankton population during this study. Abundance of such large cell must have been apparent from microscopy, which was not the case.

- We modified figure 3 in order to make the differences between the treatments more visible.

- Use of median vs. mean: It is not comprehensible to us why the referee got so confused with the statistics in our manuscript. Because we observed no Gaussian size distribution of particles, we always referred to the median size, and not to the mean size of particles. To compare the CO2 treatments, we calculated the mean value of three realisations (three mesocosms assigned to a treatment). This is clearly stated in the text, method section, and in the caption of Fig. 4. Moreover section 2.4, Statistical treatment of data, states 'Average values are given by the statistical mean and its standard deviation (SD).' And the figure captions say 'Error bars denote +/- 1 SD'. The careful reader will notice that the error bars denote the standard deviation.

- Merit of size distribution analysis with respect to community and ecosystem changes:

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4, S2611–S2613, 2008

We do not agree with the referee in this point. It is speculative to say that the differences in size distribution observed will have no influence on the community or ecosystem level, when no information is currently available on how large size differences in phytoplankton communities need to be in order to have an effect. Furthermore, it is the median size of particles that varied from 3.2 to 4.2 μm ESD between the treatments. As can be seen from Figure 3, there was a clear difference in the size distributions of particles between the treatments at the height of the bloom. These size distributions in fact represented almost the entire nano- and microphytoplankton community. Because microzooplankton grazer do not differ much in size from their prey, changes in particle size spectrum as observed here, i.e. a doubling by ESD, may affect the response of the heterotrophic ecosystem. Moreover, we show that differences in the size range considered can lead to differences in CO2 supply, with potential consequences on cell physiology and elemental cycling. We discuss several potential effects of the observed changes in size distribution on ecosystem structure in chapter 4.4.

- Minor suggestions were adopted in thanks, except that we left Eq. 2 and Fig. 2 unchanged. We think that Eq. 2 without cancellations is more comprehensible to the reader. The comparison between Coulter Counter and Flow Cytometer data as bar chart (Fig. 2) shows nicely that the two dataset agree generally during the pre-bloom and bloom phase, indicating that most of Coulter counts were due to phytoplankton cells during these periods.

Interactive comment on Biogeosciences Discuss., 4, 4101, 2007.