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Biogeosciences Discussions

Interactive comment on "Competition for inorganic and organic forms of nitrogen and phosphorous between phytoplankton and bacteria during an *Emiliania huxleyi* spring bloom (PeECE II)" *by* T. Løvdal et al.

T. Løvdal et al.

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Referee #2

Referee comment: A difficulty in the interpretation of the data lies in the fact that due to the high abundance of particle associated bacterial the two size fractions (0.2-0.8 μm and $>$0.8 μm) do not allow a clear separation between bacteria and phytoplankton. To account for this, the authors correct for the contribution of particle-associated bacteria to the $>$0.8 μm size fraction. A crucial assumption for this is "that particle-associated bacteria had the same affinity for uptake as free bacteria" (page 3353, lines 4-5). Given the large difference in bacterial size between free and attached bacteria, and given the

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differences in substrate quality and quantity that the two groups are likely to experience, this assumption is questionable. For the same reasons, it is also unlikely that temporal changes in nutrient uptake affinities were the same for free and attached bacteria throughout the experiment. Obviously, assumptions regarding the uptake affinity of particle-associated bacteria also affect the calculations of phytoplankton uptake rates. This approach greatly compromises the robustness of the results reported here.

Authors response: - We acknowledge that the separation of attached bacteria from phytoplankton is indeed difficult. Therefore, we have presented uptake data for each size fractions prior to the corrected data. We also agree that the assumption of similar affinity (per biomass) in free and attached bacteria and of the same temporal variations, is questionable. To account for the latter, data on the corrected estimates have been divided into two periods, with statistics only presented for the period after the bloom with stable TNH4 $<$ 10 h and a stable but low DIN:SRP ratio. During the revision of the manuscript (ms), we have tried to elevate the biomass-specific affinity of particle-attached bacteria to the point before the confidence limits of the statistical tests reported have been changed. This time, the assumption is that particle associated bacteria have higher affinity for all substrates than free living bacteria, as expected from the references given in the original ms. According to our correction method, however, for leucine and all the P-substrates, an affinity increase of only about 10% relative to free bacteria during the N-limited period, leaves the phytoplankton with a negative net uptake. Thus, the null hypothesis is most probably valid assuming a net uptake by phytoplankton. In the case of NH4, the null hypothesis can not be rejected as long as the biomass specific affinity of particle associated bacteria is $<$ 3.3 times the biomass specific affinity of free bacteria. Considering the theoretical maximum affinity by the diffusion model (Fig. 7 in the revised ms, Løvdal et al., 2008), it is unrealistic that the biomass specific affinity of particle associated bacteria would be higher than this value during N-limitation. This has been discussed and explained in the discussion of the revised ms ("Algal -bacterial competition; implications for the microbial food web"). For clarification: from the assumptions given in the revised ms, we have calculated

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that free-living bacteria (size 0.2 μm3) would have a maximum biomass-specific affinity (a max) for NH4 and PO4 of 0.0013 L nmol-C-1 h-1 assuming bacteria are perfect spheres. This value will increase if the cells are non-spherical (Løvdal et al., 2008). Particle associated bacteria (size 1.5 μm3) would have an a max of 0.0009 L nmol-C-1 h-1 if they are perfect spheres. It is fair to assume that the majority of the smaller bacteria were coccoid, and that larger bacteria were elongated. Most likely, big bacteria have a higher frequency of becoming rod-shaped than smaller ones; a particle-associated bacterium (size 1.5 μm3) that is rod-shaped with a length to width ratio of 5 would for example have a max of 0.0021 L nmol-C-1 h-1. These calculations were performed using revised estimates of (C) biomass as explained later. There should be no doubt, however, that the resulting experimental estimates of algal and bacterial biomass-specific affinity are rough estimates, and must be viewed in connection with all other parameters presented.

Specific comment: Authors responses: 1. Year of experiment has been included in the revised ms. 2. The reference suggested has been included in the revised ms. 3. During the revision of the ms, all proxies for biomass have been reviewed. One consequence of this is that affinity is now normalized to C-biomass. By doing so, some uncertainty related to additional conversion factors is avoided. The revised phytoplankton C-biomass is based on counting and conventional conversion factors converting from cell volume to C content in phytoplankton. This valuable work has been performed by Véronique Martin-Jézéquel, thus her name has been added to the author list. The revised phytoplankton C-biomass correlates to the original Chl a C-biomass (r=0.897, n=9), whereby the revised biomass is on average ˜ 40% higher than the original estimates. Bacterial C-biomass has been revised using a constant C cell-1 factor only for the smallest of bacteria (Loferer-Krössbacher et al., 1998) and the allometric factor for all other bacteria (see the original ms). The revised bacterial C-biomass correlates to the original bacterial C-biomass (r=0.961, n=10), whereby the revised biomass is on average 16% higher. The revised total C-biomass correlates to POC (Engel et al., 2007) (r=0.914, n=8), and to TPC (our own unpublished data; r=0.526, n=9), but POC

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is more than two-fold and TPC 2-4 fold higher, respectively, than our revised estimates. From that we assume that POC measured in the present study has included a significant fraction of detritus. Perhaps detritus has been introduced by addition of deep, high salinity water into the mesocosm, because at that time TPC has dramatically increased. It may be interesting to note that these TPC data correlates well with TPC measured elsewhere (U. Riebesell, pers. comm.) (r=0.995, n=7), whereby the former TPC values were on average ˜ 10% higher, most likely because we have used 0.8 μm silver membrane filters, whereas GF/F filters have been used in the latter study.

4. This section has been deleted from the revised ms. 5. In the section "methodological considerations" in the revised ms, we have discussed the potential effect of this treatment. See also responses to referee #1. 6. We have replaced "C-limitation" with the less strict term "C-stress".Although we agree with referee #2 that C-stress of heterotrophic processes may be unlikely, we find it quite puzzling that, although DIN and SRP decreased rapidly, the turnover times of the respective organic substrates were rather short compared to the inorganic ones. This may indicate that bacteria were not deficient in inorganic N or P during this phase, but rather hydrolyzed the organic substrates for "something else", most likely C. 7. We did not use high DIN:SRP ratios as an indicator for P-limitation (as repeatedly pointed out throughout the ms), but rather have mentioned that this is traditionally a measure of P-limitation.

On behalf of the authors, Trond Løvdal

References:

Engel, A., Schulz, K., Riebesell, U., Bellerby, R., Delille, B., and Schartau, M.: Effects of CO2 on particle size distribution and phytoplankton abundance during a mesocosm bloom experiment (PeECE II), Biogeosciences Discuss., 4, 4101-4133, 2007.

Loferer-Krössbacher, M., Klima, J., and Psenner, R.: Determination of bacterial cell dry mass by transmission electron microscopy and densitometric image analysis, Appl. Environ. Microbiol., 64, 688-694, 1998.

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Løvdal, T., Skjoldal, E. F., Heldal, M., Norland, S., and Thingstad, T. F.: Changes in morphology and elemental composition of Vibrio splendidus along a gradient from carbon-limited to phosphate-limited growth, Microb. Ecol., 55, 152-161, 2008.

Interactive comment on Biogeosciences Discuss., 4, 3343, 2007.