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Biogeosciences Discussions

Interactive comment on "Availability of phosphate for phytoplankton and bacteria and of labile organic carbon for bacteria at different pCO_2 levels in a mesocosm study" *by* T. Tanaka et al.

T. Tanaka et al.

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Interactive comments on “Availability of phosphate for phytoplankton and bacteria and of labile organic carbon for bacteria at different pCO2 levels in a mesocosm study” by Tanaka et al.

Reply to Referee #3

[Referee comment] If we accept that there are no significant differences due to pCO2 levels, then this paper has a huge problem in trying to extract something positive out of negative results (the lack an effect)… The lack of an effect perhaps is worthwhile pointing out in a short note, but not such a long paper. Perhaps there are other ways of organizing the data that would result in more interesting observations. [Author

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response] We did not detect statistically significant time-effects caused by the different pCO2 treatments in particulate P concentrations, turnover times of phosphate and glucose, and APA between the different pCO2 treatments during the experimental period (ANCOVA test, P$>$0.05). We also found no bacterial growth limitation by the availability of labile DOC but P-deficiency at three different pCO2 levels with a less available phosphate pool in the 3xCO2 mesocosm toward the end of the experiment. These results suggest that higher loss of organic carbon from the surface mixed layer with increasing pCO2 (Riebesell, et al., 2007) was likely because a smaller pool of available inorganic nutrients reduced the bacterial capacity to degrade organic carbon. We therefore believe that this ms is worthwhile publishing in a full paper format. In order to clarify these findings and arguments, we have thoroughly revised the manuscript (see the revised ms).

[Referee comment] It is perhaps interesting to compare the glucose uptake with the results of Riebesell et al. (2007, Nature). That paper reported that increasing pCO2 results in higher production of transparent extracellular polymers (TEP). Since one may expect TEP to be glucose-rich, it is surprising that glucose uptake was not apparently affected. [Author response] As the referee suggested, this fresh DOC can be composed of glucose-rich exudates (e.g. Grossart, et al., 2006). Riebesell et al. (2007) report higher production of DOC in increasing pCO2 but do not specify higher production of TEP in increasing pCO2.

[Referee comment] I wonder if there are some effects of pCO2 on at least the P properties. … but I find these types of graphs difficult to interpret. [Author response] We improved the text in order to clarify effects of pCO2 on the P properties. Figures 1 and 3 present the data in unit of μmol L-1 and nmol-P L-1 h-1, respectively, while Figure 2 (bottom) presents the data in unit of % due to the 33P uptake method used in this study. We improved the pattern presentation in Figures 1-3.

[Referee comment] The authors point out that because they analyzed samples from only one of the three mesocosms they did, the power of their statistical tests is limited.

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Perhaps others would object and call this pseudo-replication, but it seems that the authors can use analytical errors and the differences over time to make conclusions about pCO2 effects. I suspect that the analytical errors and the temporal variation, which the authors have measured, are larger than the variations among mesocosms. [Author response] A sophisticated statistical test is interesting, however, we think it is not essential for our argument. Samples of dissolved and particulate nutrients, biomass and production of phytoplankton and bacteria were measured from all nine mesocosms (3 different pCO2 treatments in triplicate), but no significant differences between samples of each pCO2 level were found for temporal changes in these parameters (ANCOVA test, $P>0.05$: Allgaier, et al., 2007; Egge, et al., 2007; Paulino, et al., 2007; Schulz, et al., 2007). With regard to the analytical errors and the temporal variations, we mention that such small differences could be explained by variance between parallel treatments rather than different treatments (Martínez-Martínez, et al., 2006).

[Referee comment] 1. page 3938, line 20 [Author response] We improved the statement in the revised ms: A significant negative correlation between specific glucose affinity and concentration of dissolved organic carbon (DOC) suggests that the temporal changes in DOC concentration were largely related to those of the labile DOC fraction (glucose). Specific glucose affinity of bacteria behaved similarly at the three different pCO2 levels with measured specific glucose affinities being consistently much lower than the theoretical maximum predicted from the diffusion-limited model. This suggests that bacterial growth was not limited by the availability of labile dissolved organic carbon.

[Referee comment] 2. page 3940, line 10 and later [Author response] We added the definition of the specific affinity in the revised ms: The specific affinity is the slope of the specific uptake rate versus a substrate concentration curve, and is analogous to a specific clearance rate, the volume cleared for food (substrate) per unit biomass and unit time. In specific substrate uptake vs. substrate concentration relationship such as Michaelis-Menten type, specific affinity is the slope between the origin and a

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specific uptake rate at a given substrate concentration. When substrate concentration approaches to zero, specific affinity approaches to its maximum.

[Referee comment] 3. page 3942, line 4 [Author response] We added a final concentration of 33PO4 in samples and incubation times in the revised ms: Carrier-free 33P-orthophosphate (Amersham, 370 MBq ml-1) was added to samples at a final concentration of 125 pmol L-1…Incubation time varied between 5 minutes and 4 hours: short enough to assure a linear relationship between the fraction of isotope adsorbed vs. the incubation time but it was long enough to reliably detect isotope uptake above background levels.

[Referee comment] 4. page 3942, line 19 [Author response] We added a final concentration of 14C-glucose in samples in the revised ms: D-[U-14C]-glucose (Amersham, 7.4 MBq ml-1) was added to samples at a final concentration of 100 nmol L-1.

[Referee comment] 4a. I suspected the added 14C glucose concentration was much higher than ambient concentration. [Author response] We discussed a potential effect of added 14C-glucose on turnover time of glucose in Results and Discussion section of the revised ms: The phytoplankton bloom resulted in an increase in bulk DOC of 25-30 μmol L-1 (Schulz, et al., 2007), which to a large degree can be composed of glucose-rich exudates (Grossart, et al., 2006). Unfortunately, we did not measure glucose concentration in this study. Since the 14C-glucose concentration (100 nmol L-1) might not always have been at tracer level in this experiment, this could to some extent have caused an overestimation of glucose turnover time.

[Referee comment] 5. page 3942, line 24 [Author response] We agree that there are not many reports of % respiration of added 14C-glucose. But this is not important in our ms, and we do not report these data in the revised ms.

[Referee comment] 6. page 3944, section 2.4 [Author response] As suggested, we delete this section.

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[Referee comment] 7. page 3944, line 4; 7a. [Author response] We modified the unit of specific glucose affinity from P to C.

[Referee comment] 8. page 3947 bottom and top of page 3948 [Author response] As suggested, we made a table (Table 1 in the revised ms).

[Referee comment] 9. page 3948, line 24 [Author response] As suggested, we corrected in the revised ms: However a significant negative correlation between specific glucose affinity and DOC concentrations ($r=-0.625$, $P<0.001$, $n=26$) suggests that the temporal changes in DOC concentration were largely related to those of the labile DOC fraction (glucose). Since we noticed that the significant negative correlation between specific glucose affinity and DOC concentrations can sufficiently be mentioned in the text, we deleted Figure 6 (bottom) presented in the original ms.

[Referee comment] 10. page 3950, line 12 [Author response] We already mentioned the comparison between the measured specific glucose affinity and the theoretical maximum predicted from the diffusion model in the original ms. We improved the description for extrapolating from glucose uptake results to labile DOC in the revised ms: It may be very difficult to extrapolate results of specific affinity of a single compound to the available pool of other compounds. However a significant negative correlation between specific glucose affinity and DOC concentrations ($r=-0.625$, $P<0.001$, $n=26$) suggests that the temporal changes in DOC concentration were largely related to those of the labile DOC fraction (glucose). This can be explained by increased production of labile DOC through phytoplankton DIC over-consumption (Toggweiler, 1993; Riebesell, et al., 2007).

[Referee comment] 11. Figure 6B [Author response] As mentioned above, we mentioned our interpretation of the negative relationship between specific glucose affinity and DOC concentration in the revised ms: However a significant negative correlation between specific glucose affinity and DOC concentrations ($r=-0.625$, $P<0.001$, $n=26$) suggests that the temporal changes in DOC concentration were largely related

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to those of the labile DOC fraction (glucose). We explained relationships between glucose turnover time and DOC concentration and between glucose turnover time and bacterial abundance in the revised ms: A significant correlation was found between glucose turnover time and bacterial abundance (r=-0.645, $P<0.001$, n=26) but not between glucose turnover time and DOC concentration (r=-0.282, $P>0.1$, n=26). This suggests that glucose was not a major component of bulk DOC. On the other hand, since we noticed that the significant negative correlation between specific glucose affinity and DOC concentrations can sufficiently be mentioned in the text, we deleted Figure 6 (bottom) presented in the original ms.

[Note by the authors] We re-arranged the author list according to the revision of our manuscript.

We thank Referee #3 for helpful comments.

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