

Dear Editor,

Now, we are sending back the reworked manuscript "Effects of CO<sub>2</sub> perturbation on phosphorus pool sizes and uptake in a mesocosm experiment during a low productive summer season in the northern Baltic Sea" by Nausch et al. The manuscript is revised according to the comments. Our responses to all comments are listed below.

We are grateful to all reviewers that they have spent their time to evaluate the manuscript and for the many helpful comments. We thank especially referee #1 and referee #3 for the indication of mistakes in the text. We would like to thank referee #2, emphasizing the importance of this work. All reviewers recommended to shortening the manuscript. In particular Referee #1 (major comment 1) suggested to focus only on CO<sub>2</sub>-related effects and to delete all chapters describing the P-dynamics in the fjord. We discussed this topic with the co-authors. Some of them had the opinion that the processes within the mesocosms cannot be understood without the knowledge of the dynamics in the area of investigation. Therefore, the chapters about the fjord in the "Results" and "Discussion" were shortened, but, not removed completely, except Table 5 that is deleted now. Data obtained from the fjord have been also eliminated from figures 9 and 10.

In addition, some changes in the verbalization are made by all (co)-authors to improve the understanding. Thus, e.g., the sentence in the discussion (page 17565, line 27) "It is hard to assess the short-term effects that we have found in phase I" is replaced by "While in phases II/III elevated CO<sub>2</sub> caused a change in the PP and PO<sub>4</sub> pools for about 22 days, changes lasting only 2 days have been observed at the beginning of phase I (Fig. 7a), but, shorter effects cannot be excluded."

Response to comments of referees:

- **Referee #1**, comment 2:

Mesocosm fCO<sub>2</sub>. Figure 2a clearly shows that fCO<sub>2</sub> was dramatically changed during the experiment. fCO<sub>2</sub> in high CO<sub>2</sub> treatment decreased from over 1600 ppm in phase I to less than 1000 ppm in phase III which is lower than the fCO<sub>2</sub> of 821 ppm mesocosm in phase I. fCO<sub>2</sub> variations are similar between the untreated and 497 ppm mesocosms. Some analyses are conducted for the whole experimental period between the untreated, intermediate, and high CO<sub>2</sub> mesocosms. Is this really appropriate analyses? The fCO<sub>2</sub> conditions in Fig. 2a simply look two CO<sub>2</sub> treatment, lower (365, 368, and 497 ppm) and higher (821, 1007, 1231 ppm).

*Response:*

The classification proposed by the referee is an option. However, the 497ppm treatment deviated significantly in mean from the control (see also pH in Fig 2b) and can potentially alter the behavior of organisms. The whole researcher group, participating in the mesocosm experiment, decided to take the same symbols for the respective mesocosms for all manuscripts to be submitted to this special issue. However the assignation to intermediate and high CO<sub>2</sub> levels done in chapter 3.1.1 is now omitted.

- Referee #1, comment 3:

Abstract. Although most of this part is devoted to describing P pool sizes and P uptake dynamics, readers would like to know whether the pool size and uptake dynamics are altered under elevated CO<sub>2</sub> conditions. Please show what is the conclusion of this study. The abstract can be written in a single paragraph.

*Response:*

The Abstract was adapted to the new version focusing more on the mesocosm experiments to aim for a clear description of CO<sub>2</sub> effects on the phosphorus cycle. The following conclusions are included

now: It can be deduced from the results, that visible effects of CO<sub>2</sub> on P pools are coupled to phytoplankton growth when the transformation of PO<sub>4</sub> into POP was stimulated. The transformation of PO<sub>4</sub> into DOP on the other hand does not seem to be affected. Additionally, there were some indications that cellular mechanisms of P regulation might be changed under CO<sub>2</sub> elevation changing the relationship between cellular constituents.

- Referee #1, comment 4:

Introduction P17546L25-27: TP pool has been recognized to be composed of PO<sub>4</sub>, DOP, particulate organic P (POP), and particulate inorganic P (PIP) (Loh and Bauer, 2000; Yoshimura et al., 2007). Since PIP composes a significant part of particulate P pool, ignoring PIP is not correct to describe P cycle in the ocean. In this study PIP did not measured, so the term particulate P (PP) or total particulate P (TPP) have to be used instead of the POP.

*Response:*

We agree. PIP should be not ignored and thus it is included in the introduction now.

According to our experience, PIP is of minor importance in open waters of the Baltic Sea, and it can be assumed for the mesocosm experiments that the P-dynamics in them is driven by organisms. However the used method does not exclude PIP. Therefore the term "POP" has been corrected by changing into "PP" throughout the ms.

- Referee #1, comment 5:

Introduction. P17547L6-8. I agree with the author's view. Since many centric and pennate diatom species showed an increase in C:P ratio in response to increases in pCO<sub>2</sub> (e.g., Sun et al., 2011; Sugie and Yoshimura, 2013), P metabolism in phytoplankton may be easily affected by an increase in CO<sub>2</sub>. Yoshimura et al. (2013, 2014) may report some changes in DOP dynamics in natural plankton communities under elevated CO<sub>2</sub> conditions. These also can become a motivation to study impacts of CO<sub>2</sub> increase on P cycle.

*Response:*

Many thanks for the suggestion to the very informative papers about the response to elevated pCO<sub>2</sub> of specific diatoms or diatom dominated population in the sub-polar region. The respective references are now included in the introduction and in the discussion:

Introduction: In CO<sub>2</sub> manipulation experiments, particulate phosphorus dynamics were studied to determine effects on C:P stoichiometry of phytoplankton (Riebesell and Tortell, 2011; Sugie and Yoshimura, 2013)

Discussion: An interaction of CO<sub>2</sub> effects with phosphorus and iron availability has been found by Sun et al. (2011) and Yoshimura et al. (2014) for a the diatom *Pseudo-nitzschia multiseriata* and for a diatom dominated subarctic plankton community.

- Referee #1, comment 6:

Sampling strategy. P17548L25-27. Seawater samples were collected for integrated entire 17m depth, but I imagine that the depth of thermocline (i.e., surface mixed layer) varied day by day. Is this method appropriate to observe temporal variations in P pool and P uptake dynamics in the mesocosms?

*Response:*

The referee is right, the thermocline was sometimes above 17m depth. The 0-17m layer was sampled to see the reaction of the whole mesocosm. As reported by (Paul et al., 2015 b) samples were also taken in the 0-10 m layer that was always above the thermocline. In contrast to Paul et al. (2015 b), P-pool sizes and uptake rates did not show significant differences between the two sampling depths. Therefore the results are not given in the manuscript.

- Referee #1, comment 7:

P uptake experiments. While  $\text{PO}_4$  uptake was measured under light conditions, ATP uptake was measured under dark conditions. ATP uptake by phytoplankton can be altered under light and dark conditions. Please explain whether the balance between phytoplankton and bacterial ATP uptake is altered under light vs. dark conditions.

*Response:*

ATP can be taken up by organisms only after degradation via nucleotidase that is an enzyme of only heterotrophic bacteria. Potentially, it is possible that phytoplankton can take up phosphate released from ATP. However, according to Tamminen (1989) the affinity to release P is much lower in phytoplankton than in heterotrophic bacteria under *in situ* light conditions. Thus the uptake of released P within an incubation time of 2h applied in our experiments should be insignificant. Incubation under light conditions should therefore not alter the obtained results.

- Referee #1, comment 8:

Discussion. This paper discusses temporal changes in P pools and uptakes in the mesocosms to show the impacts of  $\text{CO}_2$ . In addition to this, to reveal  $\text{CO}_2$  impacts on P cycle, I would like to know whether temporal changes in e.g. PP/Chl-a and PC/PP differ among the mesocosms in each phase. Changes in these ratios under elevated  $\text{CO}_2$  can alter biogeochemical cycles of bioactive elements dramatically in the future.

*Response:*

The PP/Chl-a and the PC/PP ratios did not differ among the treatments in each phase supporting the conclusion in the discussion that changes in PP were mediated by biomass formation. PC/PP ratios are discussed in detail by Paul et al. (2015 b) in detail. Therefore it was only mentioned shortly here to avoid repetitions.

- Referee #1, comment 9:

P17549L24. Is this a colorimetric method?

*Response:*

Many thanks for this hint. It is corrected by replacing coloumetric instead of colorimetric

- Referee #1, comment 10:

P17550L4. A method for silicate analysis is not described in this paper.

*Response:*

The description of the carbonate chemistry is rewritten and silicate is not anymore mentioned in the ms..

- Referee #1, comment 11:

L24. at 20\_C ==> at -20\_C?

*Response:*

It is corrected to -20°C now. Many thanks!

- Referee #1, comment 12:

L24-26. I like to see the reference for the microwave method for DOP analysis.

*Response:*

The reference (Johnes and Heathwaite, 1992) has been added.

- Referee #1, comment 13:

P17551L7. Why the subsamples need to be filtered through 0.2  $\mu\text{m}$  filter in addition to through GF/F?

*Response:*

Direct filtration of the required water quantity through 0.2 $\mu\text{m}$  filters was very difficult because the pores rapidly clogged. Some picoplankton passes GF/F-filters. Therefore, the water was pre-filtered

through GF/F-filters to remove large particles and then through 0.2µm filters to remove the picoplankton that passes the GF/F filters. The respective note is included:

“For all analyzed components, subsamples were pre-filtered through pre-combusted (6 h, 450°C) filters (Whatman GF/F) to remove larger particles followed by filtration through 0.2µm cellulose acetate filters to remove picoplankton.”

- Referee #1, comment 14:

L13. Bjorkman ==> Björkman

*Response:*

Many thanks for the hint. It is corrected now.

- Referee #1, comment 15:

P17552L2. 2.5 pmol ml<sup>-1</sup> = 2.5 nmol l<sup>-1</sup>?

*Response:*

The unit was changed into nmol l<sup>-1</sup>.

- Referee #1, comment 16:

1P17553L21-22. I like to see the reference for the pressure cooker method for PP analysis. Is there any reason why you use Oxisolv here, not potassium peroxydisulfate as in DOP analysis?

*Response:*

The use of a pressure cooker for organic phosphorus and nitrogen (POP, DOP, PON, DON) analysis is mentioned by Grasshoff et al. (1983), a standard manual for seawater analyses, including modifications for the Baltic Sea. The pressure cooker was replaced by a microwave later because the Teflon bottles used in the pressure cooker partly leaked influencing the reproducibility of measurements. “Grasshoff et al. (1983)” has been mentioned already in the chapter.

For PP analysis, different methods are possible. Potassium peroxydisulfate, recommended by Grasshoff et al. (1983) is generally used in our laboratory. But Oxisolv is provided by the companies to make handling easier and is applied in several labs. Using both methods simultaneously, we were able to compare them and we found no significant differences. This may be a useful piece of information for other researchers as well. Therefore, we have written in the text, that potassium peroxydisulfate was used in the aqueous method like for DOP:

“Particulate phosphorus (PP) was analyzed using two methods in parallel. In the “aqueous method”, 40 ml of unfiltered subsamples were frozen at -20°C and analyzed as described for DOP using the potassium peroxydisulfate digestion (Grasshoff et al., 1983).”

- Referee #1, comment 17:

L26. Could you show the detection limit for PO<sub>4</sub> analysis?

*Response:*

The detection limit of 0.02 µmol l<sup>-1</sup> is now given.

- Referee #1, comment 18:

18. P17554L1. Does the PC include particulate organic and inorganic carbon

*Response:*

The term PC is used because particulate inorganic carbon cannot be completely excluded. See also (Paul et al., 2015 b).

- Referee #1, comment 19:

Could you show the light intensity for the laboratory incubation, and the light condition correspond to which depth in the mesocosms?

*Response:*

Unfortunately, we have not measured the light intensity in the laboratory. However, when I started with radiotracer experiments years ago, I have compared different light conditions for the incubations. It was found that different light conditions do not influence the results. In the literature, different opinions are reported. Some researchers found an influence and others not.

- Referee #1, comment 20:

20. P17555L6. Please use “Bq (SI unit)” not “Ci”.

*Response:*

Ci is replaced by Bq now:

“Triplicates and a formalin-killed control were incubated with  $^{14}\text{C}$ -Leu ( $7.9\text{GBq mmol}^{-1}$ ; Hartmann Analytic GmbH, Germany) at a final concentration of  $165\text{ nmol l}^{-1}$ , which ensured saturation of uptake systems of both free and particle-associated bacteria.”

- Referee #1, comment 21:

L19. I like to see the reference for the “factor of 2”.

*Response:*

The reference of Simon and Rosentstock is added:

An intracellular isotope conversion factor of 2 has been used according to Simon and Rosenstock (1992).

- Referee #1, comment 22:

P17556L13-15. M1 and M5 etc. (probably mesocosm#1) are not defined in any part of this paper.

*Response:*

As already done on this page, it is written at the beginning of the results that M1 and M5 are the untreated levels. The arrangement of the mesocosms has been changed following the comments of the referee and avoiding a classification:

M1 365  $\mu\text{atm } f\text{CO}_2$ , pH 8.08

M5 368  $\mu\text{atm } f\text{CO}_2$ , pH 8.07

M7 497  $\mu\text{atm } f\text{CO}_2$ , pH 7.95

M6 821  $\mu\text{atm } f\text{CO}_2$ , pH 7.74

M3 1007  $\mu\text{atm } f\text{CO}_2$ , pH 7.66

M8 1231  $\mu\text{atm } f\text{CO}_2$ , pH 7.58

M1 and M5 were the untreated mesocosms and served as controls.

- Referee #1, comment 23:

23. L17. Table 1 shows that minimum temperature was 7.82, not 7.81 here.

*Response:*

It is changed in the text now. Many thanks for these hints.

- Referee #1, comment 24:

P17558L4. POC ==> PC

*Response:*

POC has been changed into PC. The present sentence: “PP developed in parallel with PC.”

- Referee #1, comment 25:

L4 and L6. Fig. 6b ==> Fig. 5b?

*Response:*

Fig. 6b is changed into 5b: “.....(Figs. 5b, 7).”

- Referee #1, comment 25:

L6. Table 5 ==> Table 2?

*Response:*

Table 5 has been changed into Table 2: "PP developed in parallel with PC. The two parameters were positively correlated in the untreated and the intermediate CO<sub>2</sub> treatments, but not in the high CO<sub>2</sub> treatments (Table 2)."

- Referee #1, comment 27:  
P17559L28. PO<sub>4</sub> uptake rates ==> PO<sub>4</sub> turnover times?

*Response:*

Yes the referee is right. It is rewritten now: "ATP turnover times of 0.2 to 3.6 days (mean 0.94±0.74 days, n=90) were much shorter than the PO<sub>4</sub> turnover times and did not vary between the treatments."

- Referee #1, comment 28:  
28. P17560L1-4. I do not understand this. Does this agree with Fig. 9d?

*Response:*

Many thanks pointing to this mistake. It is shown in Fig 9c as corrected in the text. The sentence is not essential. Thus, it is omitted in the reworded ms:

"ATP turnover times of 0.2 to 3.6 days (mean 0.94±0.74 days, n=90) were much shorter than the PO<sub>4</sub> turnover times and did not vary between the treatments (Fig. 9c)."

- Referee #1, comment 29:  
29. L14. Table 2 ==> Table 5?

*Response:*

Table 5 is deleted in the manuscript and not mentioned anymore in the present ms.

- Referee #1, comment 30:  
P17561L10. Fig. 6b ==> Fig. 5b?

*Response:*

The numbers of figures are corrected in the present ms.

- Referee #1, comment 31:  
P17563L5-7. Comparing Fig. 9a and b, I consider that the shortest turnover times in days 15-17 correspond to the highest uptake rates in days 15-17.

*Response:*

By shortening the manuscript, PO<sub>4</sub> turnover times and uptake rates are not shown in the present ms.

- Referee #1, comment 32:  
L13-14. I do not understand the two number "0.02 and 0.46 nmol (µg Chl a)-1h-1".

*Response:*

This chapter is deleted in the manuscript and the comment is not applicable in the present ms.

- Referee #1, comment 33:  
Table 5. In "Variable" pCO<sub>2</sub> ==> fCO<sub>2</sub>?

*Response:*

Table 5 is omitted in the reworked ms.

- Referee #1, comment 34:  
Figure 2. fCO<sub>2</sub> (µmol L<sup>-1</sup>)? Put "b" on the bottom figure.

*Response:*

The unit of measurement is changed into µatm like in Table 2. The "b" is included now.

- Referee #1, comment 35:

Figure 5c. Put a dotted line.

*Response:*

Fjord data are eliminated from all figures.

❖ **Referee #2** comment:

Is Tvärminne northern Baltic as stated by the authors?

*Response:*

The Gulf of Finland is attributed to the northern Baltic Sea.

**Mentioned literature here:**

Grasshoff, K., Ehrhardt, M., and Kremling, K. (Eds.): Methods of seawater analysis, Verlag Chemie, Weinheim, 419 pp., 1983.

Johnes, P. and Heathwaite, A. L.: A procedure for the simultaneous determination of total nitrogen and total phosphorus in freshwater samples using persulfate microwave digestion., *Water Res.*, 26, 1281-1287, 1992.

Paul, A. J., Bach, L. T., Schulz, K.-G., Boxhammer, T., Czerny, J., Achterberg, E. P., Hellemann, D., Trense, Y., Nausch, M., Sswat, M., and Riebesell, U.: Effect of elevated CO<sub>2</sub> on organic matter pools and fluxes in a summer, post spring-bloom Baltic Sea plankton community, *Biogeosciences*, 12, 6181-6203, 2015 b.

Simon, M. and Rosenstock, B.: Carbon and nitrogen sources of planktonic bacteria in Lake Constance studies by the composition and isotope dilution of intracellular amino acids., *Limnol. Oceanogr.*, 37, 1496-1511, 1992.

Sugie, K. and Yoshimura, T.: Effects of pCO<sub>2</sub> and iron on the elemental composition and cell geometry of the marine diatom *Pseudo-nitzschia pseudodelicatissima* (Bacillariophyceae), *J. Phycol.*, 49, 475-488, 2013.

Sun, J., Hutchins, D. A., Feng, Y. Y., Seubert, E. L., Caron, D. A., and Fu, F. X.: Effects of changing pCO<sub>2</sub> and phosphate availability on domoic acid production and physiology of the marine harmful bloom diatom *Pseudo-nitzschia multiseries*, *Limnol. Oceanogr.*, 56, 829-840, 2011.

Tamminen, T.: Dissolved organic phosphorus regeneration by bacterioplankton: 5'-nucleotidase activity activity and subsequent phosphate uptake in a mesocosm enrichment experiment, *Mar. Ecol.-Prog. Ser.*, 58, 89- 100, 1989.

Yoshimura, T., Sugie, K., Endo, H., Suzuki, K., Nishioka, J., and Ono, T.: Organic matter production response to CO<sub>2</sub> increase in open subarctic plankton communities: Comparison of six microcosm experiments under iron-limited and -enriched bloom conditions, *Deep-Sea Res. Part I-Oceanogr. Res. Pap.*, 94, 1-14, 2014.

❖ **Referee #3, Comment 1:**

Referee 3 is wondering why this location has been chosen for our experiment. Mesocosm experiments as done in this study with about 40 participants requires a well-elaborated management. The required conditions in connection with the Zoological station Tvärminne were given in this area where mesocosms experiments were performed already before. No other location with comparable preconditions could be found in the Baltic Sea. There is no permanent upwelling at the Finish coast of the Gulf of Finland. Depending on the wind direction, upwelling can also occur at the coasts of Estonia and Latvia. Thus, the intense upwelling during our study period could not be foreseen.

P17545,ln23-28.The statement that the “significant relationships”..”vanished” in the CO<sub>2</sub> treatments seems contradictory to the next sentence- “Consequently, it can be hypothesized that..” Please clarify.

Response:

The abstract has been rewritten according to the comments of all referees. The specific sentence, mentioned here, has the following verbalization: “In addition, observed significant relationships (e.g., between PP and Chl<sub>a</sub>) in the untreated mesocosms disappeared under increased fCO<sub>2</sub> conditions”.

Referee #3, Comment 2:

P17548, ln 16. “CO<sub>2</sub> treatment started on day 0 and was repeated on subsequent 4 days.” This is a bit unclear.

Was CO<sub>2</sub> injected during **the** “subsequent 4 days”, i.e., day 1, 2, 3 and 4? Or was it one additional injection on Day 4? I assume the former.

Response: The sentence is rewritten now: “CO<sub>2</sub> treatment was injected at day 0 and at the subsequent 4 days by pumping various quantities of 50-µm-filtered and CO<sub>2</sub>-enriched fjord water into seven of the mesocosms as described by Riebesell et al. (2013)”.

Referee #3, Comment 3:

P17553, ln24-28. This sentence is hard to understand as written. I suggest omitting the “both” and rearranging the sentence slightly..”significant differences between the two methods..however, the difference between the means for the filter method and the aqueous method (0.19±0.03 µmol l<sup>-1</sup> and 0.16±0.04 µmol l<sup>-1</sup> respectively) where near the detection limit of the methods.

Response: The sentence is rewritten as:

“Paired t-test revealed significant differences between the two methods, however, the difference between the means of the filter method and of the aqueous method (0.19 ± 0.03 µmol l<sup>-1</sup> and 0.16 ± 0.04 µmol l<sup>-1</sup>, respectively) were near the detection limit (0.02 µmol l<sup>-1</sup>) of the methods.”

Referee #3, Comment 4:

P 17556, ln 13-15. What was the rational of making three groups of two mesocosms each, rather Than two groups with three each, or even just Control (M1,M5) versus high fCO<sub>2</sub>, low pH (M3, M8)? It seems to me that the middle group’s (M7, M6) values are farther from one another in terms of fCO<sub>2</sub> or pH, than to either the lower or higher groups.

Response:

The classification proposed by the referee is an option. However, the 497ppm treatment deviated significantly in mean from the control (see also pH in Fig 2b) and can potentially alter the behavior of organisms. The whole group of researchers has decided to take the same symbols for the respective mesocosms used in this manuscript for all manuscripts to be submitted to this special issue. However the assignation to intermediate and high CO<sub>2</sub> levels in chapter 3.1.1 is omitted now.



Referee #3, Comment 5:

P17557, ln4-6. This sentence is confusing to me. It seems to say that an increase by 24% was statistically significant in phase III but 0.27 was not? What is the 24% in reference to? In Fig 4 it does not look much happens to the higher CO<sub>2</sub> mesocosms between day24 (midway into phase II) and day 40 (phase III)..may be this is only because the figure is busy(?)

Response:

I agree with the referee and changed the sentence: The increase in Chl<sub>a</sub> in the high CO<sub>2</sub> mesocosms by 0.27 µg l<sup>-1</sup> in phase III was only marginal. Nevertheless according to Paul et al. (2015 b), this represents an increase of 24% which is a significant difference compared to the untreated mesocosms.

Referee #3, Comment 6:

P 17565, ln24. Why is the P-uptake rate a measure of gross uptake? And why would a change in net modify the retention in the POP? Isn't it likely that the size, and community structure changed (i.e. larger phytoplankton) and gross also increased i.e. the flux of P increased, and more biomass contained more P? I am not sure how you can distinguish gross versus net here if DOP production and PO<sub>4</sub> recycling weren't measured.

Response: The general method for uptake rates measurements is the detection of the straight slope of radiotracer incorporation into biomass. Therefore, the measured uptake is a gross uptake. At longer incubations release of radiotracers again from the biomass can be observed. New PP formation at an unchanged uptake rate can only occur when more P is retained in the biomass. This process should be the same in all organisms. The text has been changed now into: However, the elevated transformation of PO<sub>4</sub> into PP was not reflected in the PO<sub>4</sub> uptake rates which can be seen as gross uptake rates. But, an increase of PP, caused by biomass formation, while the PO<sub>4</sub> uptake remained unchanged can only occur when the P release from the organisms is reduced. Thus, it is likely that not the gross uptake but rather the net uptake was modified under CO<sub>2</sub> elevation.

Referee #3, Comment 7:

P17546,ln7. "...predicted to rise to 750-> 1000 ppm.." What does the ">" mean? Up to?

Response: It is rewritten into:

Atmospheric CO<sub>2</sub> is predicted to rise to **750 - 1000 ppm and higher** in 2100 (IPCC, 2001) corresponding with a decrease in pH by 0.3-0.5 units (Caldeira and Wickett, 2005) from the present pH of 8.1.

Referee #3, Comment 8:

P17550, ln19-20. Is a p-value of 0.026 not significantly different for the two methods used for PO<sub>4</sub> analysis?

Response: It is right, we are grateful for the hint. The p value is 0.26.

It is corrected now.

Referee #3, Comment 9:

P17550,ln24. Should this be -20°C? (now it read 20°C).

Response: That is right, now it is corrected:

For the determination of DOP, duplicate 40-ml subsamples were filtered through pre-combusted (6 h, 450 °C) glass fiber filters (Whatman GF/F) and stored in 50-ml vials (Falkon) at -20°C until further processing

Referee #3, Comment 10:

P17551,ln16. 2ml to 200ml is 1% v/v.

Response: Many thanks for the hint. It is corrected now:

The blank was obtained by the addition of formaldehyde (1% final concentration) 10 min before radiotracer addition, in order to poison the samples

Referee #3, Comment 11:

P17554,ln 20. Of what materials were the 0.2 and 0.3µm filters?

Response: it is made more clear that PC-filters were used. Now it is written:

At defined time intervals within the incubation, 5-ml subsamples were taken from each of the parallel samples and filtered onto polycarbonate (PC) filters pre-soaked with a cold 20 mM PO<sub>4</sub> solution to prevent non-specific [<sup>33</sup>P]PO<sub>4</sub> binding..... PC-filters of 0.2 and 3 µm pore sizes (Whatman and Millipore, respectively) were used to determine uptake by the whole plankton community and the size fraction >3µm, respectively.

Referee #3, Comment 12:

P17557,ln17, and 27. Does the 116 nmol L<sup>-1</sup>, 0.12 µmol L<sup>-1</sup> and 0.06 µmol L<sup>-1</sup> have propagated Error estimates?

Response:

Standard deviations are included now as indicated here: Thus, the loss of phosphorus (116 ± 34 nmol l<sup>-1</sup>) from the 17-m layer during the 29-day measurement period was calculated to be 4.0 nmol l<sup>-1</sup> day<sup>-1</sup>.

and

Averaged over all mesocosms, TP decreased by 0.12 ± 0.03 µmol l<sup>-1</sup>, whereas PP declined only by 0.06 ± 0.01 µmol l<sup>-1</sup> during this period.

Referee #3, Comment 13:

P17558,ln6. Should this be table 2? (not 5?)

Response:

Table 5 has been changed into Table 2:

Referee #3, Comment 13:

P 17558,ln 24. What is meant with “variations only in the nanomolar range”?

Perhaps state something like number of standard deviations instead, or ±x.

Response:

The sentence is rewritten into: **Phosphate (PO<sub>4</sub>)** concentrations ranged between 0.06 and 0.21 µmol l<sup>-1</sup>, with differences of 0.01 - 0.06 µmol l<sup>-1</sup> between the mesocosms.

Referee #3, Comment 14:

P 17559, pg 3.1.4. Perhaps also use the median values here, where the range is

Large but the means seem to be skewed.

Response: The use of mean or median values depends from the viewpoint of the author. Sometimes the use of median values might be more appropriate, but, it is not essential for this manuscript.

I decided to use means and standard deviations throughout the text. If the means would be replaced by medians here than an inconsistency would be in the text. Means and median values are mostly similar, as the comparison showed.:

Mean 4.0 - median 3.84;

mean 1.7-median, 1.5;

mean 41.3 –median 36.0;

mean 86.5 – median 86.5

mean 13.3 median 12.6

Referee #3, Comment 15:

P17560,ln2. What is meant by degraded? Total hydrolysis of ATP, or P-incorporation

Into cells?

Response:

The sentence is rewritten: Between 0.05 and 0.36 nmol ATP l<sup>-1</sup> h<sup>-1</sup> (mean 0.14±0.08 nmol l<sup>-1</sup> h<sup>-1</sup>, n=36) were **hydrolysed**, corresponding to a P supply of 0.14 and 1.08 nmol l<sup>-1</sup> h<sup>-1</sup> (mean 0.44±0.25 nmol l<sup>-1</sup> h<sup>-1</sup>, n=36).

Referee #3, Comment 16:

Table 5. What does the -, and +signs mean here?

Response:

Table 5 is omitted in the reworked ms due to shortening of the ms.