Dear Editor,

Now, we are sending back the reworked manuscript "Effects of CO_2 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.
Following the recommendations of the reviewers, the manuscript was revised in parallel with the answers to the reviewers that were sent to you before. It is therefore possible to submit it immediately. In addition to the recommendations, some changes in the verbalization are made by all (co)-authors to improve the understanding.
Together with the detailed response to the reviewer comments, we are sending two versions of the reworked manuscript: one in which all changes are annotated and a second in which the changes are not indicated.

Sincerely yours
Monika Nausch

Response to referees

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_2-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_2 caused a change in the PP and PO_4 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."

We are sending two versions of the manuscript: one in which all changes are annotated and a second in which the changes are not indicated.

Response to comments of referees:

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

Mesocosm fCO_2. Figure 2a clearly shows that fCO_2 was dramatically changed during the experiment. fCO_2 in high CO_2 treatment decreased from over 1600 ppm in phase I to less than 1000 ppm in phase III which is lower than the fCO_2 of 821 ppm mesocosm in phase I. fCO_2 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_2 mesocosms. Is this really appropriate analyses? The fCO_2 conditions in Fig. 2a simply look two CO_2 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_2 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_2 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_2 effects on the phosphorus cycle. The following conclusions are included now: It can be deduced from the results, that visible effects of CO_2 on P pools are coupled to phytoplankton growth when the transformation of PO_4 into POP was stimulated. The transformation of PO_4 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_2 elevation changing the relationship between cellular constituents.

- Referee #1, comment 4:

Introduction P17546L25-27: TP pool has been recognized to be composed of PO_4, 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_2 (e.g., Sun et al., 2011; Sugie and Yoshimura, 2013), P metabolism in phytoplankton may be easily affected by an increase in CO2. Yoshimura et al. (2013, 2014) may report some changes in DOP dynamics in natural plankton communities under elevatedCO2 conditions. These also can become a motivation to study impacts of CO2increase on P cycle.

Response:

Many thanks for the suggestion to the very informative papers about the response to elevated pCO_2 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_2 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_2 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 multiseries* and for a diatom dominated subarctic plankton community.

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- 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 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 CO_2. In addition to this, to reveal 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 CO_2 can alter biogeochemical cycles of bioactive elements dramatically in the future.

Response:

The PP/Chla 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 µm filter in addition to through GF/F?

Response:

Direct filtration of the required water quantity through 0.2µm 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^{-1} = 2.5 nmol l^{-1}?

Response:

The unit was changed into nmol l^{-1}.

- 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_4 analysis?
Response:
The detection limit of 0.02 µmol l^{-1} is now given.

- Referee #1, comment 18:

18. P17554Ll. 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}C-Leu (7.9GBq $mmol^{-1}$; Hartmann Analytic GmbH, Germany) at a final concentration of 165 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 µatm fCO_2, pH 8.08
M5 368 µatm fCO_2. pH 8.07
M7 497 µatm fCO_2, pH 7.95
M6 821 µatm fCO_2, pH 7.74
M3 1007 µatm fCO_2, pH 7.66
M8 1231 µatm fCO_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_2 treatments, but not in the high CO_2 treatments (Table 2)."

- Referee #1, comment 27:

P17559L28. PO_4 uptake rates ==> PO_4 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_4 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_4 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_4 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_2 ==> fCO_2?
Response:
Table 5 is omitted in the reworked ms.

- Referee #1, comment 34:

Figure 2. fCO_2 (umol L^{-1})? 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 CO2 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 pCO2 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(2) 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 CO2 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 choosen 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 CO2 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*a*) in the untreated mesocosms disappeared under increased fCO_2 conditions".

Referee #3, Comment 2:
P17548, ln 16. "CO_2 treatment started on day 0 and was repeated on subsequent 4 days." This is a bit unclear.
Was CO_2 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_2 treatment was injected at day 0 and at the subsequent 4 days by pumping various quantities of 50-µm-filtered and CO_2-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^{-1} and 0.16±0.04 µmol l^{-1} 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^{-1} and 0.16 ± 0.04 µmol l^{-1}, respectively) were near the detection limit (0.02 µmol l^{-1}) 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 fCO2, 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_2 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_2 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 CO2 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*a* in the high CO_2 mesocosms by 0.27 µg l^{-1} 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 PO4 recycling weren't measured.
Response: The general method for uptake rates measurements is the detection of the strait 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_4 into PP was not reflected in the PO_4 uptake rates which can be seen as gross uptake rates. But, an increase of PP, caused by biomass formation, while the PO_4 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_2 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_2 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 PO4 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 where 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_4 solution to prevent non-specific [^{33}P]PO_4 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-1, 0.12 µmol L-1 and 0.06 µmol L-1 have propagated Error estimates?
Response:
Standard deviations are included now as indicated here: Thus, the loss of phosphorus (116 ± 34 nmol l^{-1}) from the 17-m layer during the 29-day measurement period was calculated to be 4.0 nmol l^{-1} day^{-1}.
and
Averaged over all mesocosms, TP decreased by 0.12 ± 0.03 µmol l^{-1}, whereas PP declined only by 0.06 ± 0.01 µmol l^{-1} 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_4)** concentrations ranged between 0.06 and 0.21 µmol l^{-1}, with differences of 0.01 - 0.06 µmol l^{-1} 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^{-1} h^{-1} (mean 0.14±0.08 nmol l^{-1} h^{-1}, n=36) were **hydrolysed**, corresponding to a P supply of 0.14 and 1.08 nmol l^{-1} h^{-1} (mean 0.44±0.25 nmol l^{-1} h^{-1}, 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.

Effects of CO_2 perturbation on phosphorus pool sizes and uptake in a mesocosm experiment during a low productive summer season in the northern Baltic Sea

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Abstract

Studies investigating the effect of increasing CO_2 levels on the phosphorus cycle in natural waters are lacking although phosphorus often controls phytoplankton development in many aquatic systems. The aim of our study was to analyze effects of elevated CO_2 levels on phosphorus pool sizes and uptake. ~~Therefore, we conducted~~ the phosphorus dynamic was followed in a CO_2-manipulation mesocosm experiment in the Storfjärden (western Gulf of Finland, Baltic Sea) in summer 2012~~.~~ and was also ~~We compared the phosphorus dynamics in different mesocosm treatments but also~~ studied ~~them outside the mesocosms~~ in the surrounding fjord water.

In ~~the~~ all mesocosms as well as in surface waters of Storfjärden, dissolved organic phosphorus (DOP) concentrations of 0.26±0.03 and 0.23±0.04 µmol l^{-1}, respectively, formed the main fraction of the total P-pool (TP), whereas phosphate (PO_4) constituted the lowest fraction with mean concentration of 0.15 ±0.02 µmol l^{-1} in the mesocosms and 0.17±0.07 µmol l^{-1} ~~in the mesocosms and~~ in the fjord~~, respectively~~. Transformation of PO_4 into DOP appeared to be the main pathway of PO_4 turnover. About 82% of PO_4 was converted into DOP whereby only 18% of PO_4 was transformed into particulate phosphorus (PP). ~~Uptake of~~ PO_4 uptake rates measured in the mesocosms ranged between 0.6 and 3.9 nmol l^{-1} h^{-1}. ~~of which ~~ About 86% ~~(mesocosms) and ~72% (fjord~~) of the uptake rates were realized by the size fraction <3 µm. Adenosine triphosphate (ATP) uptake revealed that additional P was supplied from organic compounds accounting for 25-27% of P provided by PO_4 only.

CO_2 additions did not cause significant changes in phosphorus (P) pool sizes, DOP composition, and uptake of PO_4 and ATP when the whole study period was taken into account. ~~About 18% of PO_4 was transformed into POP, whereby the major proportion (~82%) was converted into DOP suggesting that the conversion of PO_4 to DOP is the main pathway of the PO_4 turnover.~~ However, significant short-term effects were observed for PO_4 and PP pool sizes in CO_2 treatments >1000 µatm during periods when phytoplankton biomass increased

In addition,we ~~We observed that~~ found ~~s~~significant relationship~~s~~ (e.g., between P~~O~~P and Chl*a*) in the untreated mesocosms which were not observed under high ~~vanished under increased~~ *f*CO_2 conditions. Consequently, it can be hypothesized that the relationship between P~~O~~P formation and phytoplankton growth changed ~~under elevated CO_2 conditions~~ with CO2 elevation. ~~Significant short-term effects were observed for PO_4 and particulate organic phosphorus (POP) pool sizes in CO_2 treatments >1000 µatm during periods when phytoplankton started to grow.~~

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It can be deduced from the results, that visible effects of CO_2 on P pools are coupled to phytoplankton growth when the transformation of PO_4 into OP was stimulated. The transformation of PO_4 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_2 elevation changing the relationship between cellular constituents.

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Kea words
ocean acidification, phosphorus cycling, DOP composition, PO_4 uptake, ATP uptake, Baltic Sea

1. Introduction

Increasing emissions of anthropogenic CO_2 into the atmosphere and subsequent acidification of the ocean can potentially affect the diversity of organisms and the functioning of marine ecosystems (Eisler, 2012). The rise ~~of the~~ in atmospheric CO_2 ~~content~~ concentrations was accelerated from 3.4±0.2 PgC yr^{-1} in the 1980s to 4.0±0.2 PgCyr^{-1} in the 2000s leading to increases of CO_2 ~~elevation~~ in ocean surface waters ~~the same~~ at a similar rate (IPCC, 2013) . Atmospheric CO_2 is ~~predicted~~ projected to rise to 750 ~~-~~ >1000 ppm and higher ~~ppm~~ in 2100 (IPCC, 2001) corresponding with a decrease in open ocean pH by 0.3-0.5 units (Caldeira and Wickett, 2005) from the present level ~~pH~~ of ~8.1. Although this process is of global significance and all parts of the oceans are at risk, there will be regional differences in the degree of acidification (Borges et al., 2005). Thus, to determine the CO_2-related changes in the oceans, multiple studies in different regions are required. Semi-enclosed coastal regions, such as the Baltic Sea, ~~can be more sensitive~~ react with with higher changes in pH to CO_2 elevation than open ocean waters due to high freshwater inputs resulting in a reduced buffer capacity (Orr, 2011).

In the Baltic Sea, several studies of CO_2 effects ~~are done~~ have been undertaken on the organism~~ic~~ level of fish (Frommel et al., 2013), zooplankton (Pansch et al., 2012; Vehmaa et al., 2012), macrophytes (Pajusalu et al., 2013), benthic ~~organisms~~ species (Hiebenthal et al., 2013; Stemmer et al., 2013), and filamentous cyanobacteria (Czerny et al., 2009; Eichner et al., 2014; Wannicke et al., 2012). Studies on the ~~The~~ impacts of elevated CO_2 at the ecosystem level, however, have thus far been limited to ~~the~~ Kiel ~~Bay~~ Bight in the western Baltic Sea (Engel et al.,

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2014; Rossoll et al., 2013; Schulz and Riebesell, 2013), which may fundamentally differ from other parts of the Baltic Sea.

Next to nitrogen, phosphorus (P) controls the productivity of phytoplankton in the ocean (Karl, 2000; Sanudo-Wilhelmy et al., 2001; Tyrrell, 1999) and is ~~is the~~ a limiting factor in some regions (Ammerman et al., 2003). The total phosphorus (TP) pool comprises phosphate (PO_4), dissolved organic phosphorus (DOP), and particulate organic ~~phosphorus~~ (POP) and inorganic (PIP) phosphorus. There is a continuous transformation of phosphorus between these P species due to their uptake, conversion, and release by organisms as well as by interaction with minerals. While PO_4 is the preferred P-species of phyto- and bacterioplankton, DOP can become~~s~~ an important P source ~~in particular~~ when PO_4 is depleted (Llebot et al., 2010; Lomas et al., 2010). DOP includes nucleic acids, phospholipids, and adenosine triphosphate (ATP) (Karl and Björkman, 2002) which are structural and functional components ~~in~~ of all living cells, but, can be also released into the surrounding water.

In general, there is little knowledge on how the P cycle is affected by ocean acidification and how related changes in P availability influence the response of organisms to CO_2 elevation. In CO_2 manipulation experiments, particulate phosphorus dynamics were studied to determine effects on C:P stoichiometry of phytoplankton (Riebesell and Tortell, 2011; Sugie and Yoshimura, 2013) and PO_4 concentration dynamics to estimate its utilization (Bellerby et al., 2008). CO_2 effects on phosphorus pool sizes and PO_4 uptake have so far ~~only~~ been studied by Tanaka et al. (2008) in the Raunefjorden, Norway and by Unger et al. (2013) and Endres et al. (2013) in laboratory experiments with cultures of *Nodularia spumigena*. ~~Thus, there is a gap of knowledge on how the phosphorus cycle may be affected under future CO_2 conditions~~. In order to reduce the gap of knowledge, ~~We~~ we ~~therefore~~ studied the impact of elevated CO_2 on phosphorus pool sizes, the DOP composition, and PO_4 uptake of a northern Baltic Sea plankton community. These measurements provide important information on potential changes in P cycling under ~~future conditions~~ increasing CO_2 levels and ~~thus will~~ contribute to a better understanding of ~~potential impacts of increased CO_2 levels~~ the P cycle in brackish water ecosystems.

2. Material and methods

2.1 Experimental design and CO_2 manipulation

The study was conducted in the northwestern Gulf of Finland, in the proximity of the Tvärminne Zoological Station (TZS) (Fig. 1), between June 17 and August 4, 2012, using the KOSMOS

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mesocosm system (Riebesell et al., 2013). Nine mesocosms (M1 - M9) were moored in the open waters of the Storfjärden (5951.5 N, 2315.5 E) at a water depth of ~30 m. Only six of them were included throughout the whole study period since leakages in the remaining three rendered them unusable. Equipment and deployment procedures are described in detail by Paul et al. (2015b). Briefly, polyurethane enclosure bags of 2 m in diameter and 18.5 m in length were mounted in floating frames and lowered in such a way that ~17 m of each bag were immersed in the water column and ~1.5 m remained above the water surface. Large organisms were excluded from the mesocosms by a 3-mm mesh installed at the top and bottom of the bags before closure. The mesocosms were deployed 10 days prior to CO_2 manipulation to rinse the bags and for full water exchange. Sediment traps were mounted on the lower ends to close them water tight, while the upper ends were raised above the water surface to prevent water entry during wave action. The mesocosms were covered with a dome shaped roof to prevent nutrient input by bird and potentially significant fresh water input by rain. Salinity gradients were removed by bubbling the mesocosms with compressed air for 3.5 min, so that 5 days before the start of the experiment (day −5) the water body was fully homogeneous.

CO_2 ~~treatment started on day 0 and was repeated on subsequent 4 days~~ was injected at day 0 and the subsequent 4 days by pumping various quantities of 50-µm-filtered and CO_2-enriched fjord water into seven of the mesocosms as described by Riebesell et al. (2013). The intended CO_2 and pH gradients were reached after the last treatment on day 4. Details are described in Paul et al. (2015b). For the two untreated (control) mesocosms, only filtered fjord water was added to adjust the water volume to that of the treated mesocosms. To compensate for outgassing, the CO_2 manipulation was similarly repeated in the upper 7 m layer of the mesocosms on day 16.

2.2 Sampling

Daily sample collection started 3 days before the first CO_2 injection (day−3). Parallel samples were taken from the surrounding fjord. Sampling over the entire 17 m depth was carried out using an integrating water sampler (IWS HYDROBIOS -KIEL) that was lowered slowly on a cable by hand. The sampling frequency differed depending on the parameter to be observed as shown in the overview by Paul et al. (2015b).

Phosphorus pool parameters and uptake rates were determined every second day, except for dissolved organic phosphorus (DOP) components, which were measured every 4 days. Termination of the measurements varied due to logistical constrains. Thus, total phosphorus (TP) and DOP were sampled only until day 29 whereas other parameters were sampled until day 43.

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The collected water was filled in HCl-cleaned polyethylene canisters that had been pre-rinsed with sample water. All containers were stored in the dark. Back on land, subsamples were processed immediately for each P-analysis. The other analyses were carried out within a few hours of sample collection and sample storage in a climate room at *in situ* temperature.

2.~~2~~3 Analytical methods

2.~~3~~2.1 Temperature, salinity, and carbonate chemistry

Measurements in the fjord and in each mesocosm were conducted using a CTD60M memory probe (Sea and sun technology, Trappenkamp, Germany) lowered from the surface to a depth of 17 m at about 0.3 m s^{-1} in the early afternoon (1:30 - 2:30 pm). For these parameters, ~~the~~ depth-integrated mean values are presented here.

The carbonate system is described in detail in Paul et al. (2015b). The pHT ~~w~~(total scale) was determined using ~~the~~ a spectrophotometric method (Dickson et al., 2007) ~~using~~ on a Cary 100 (Varian) ~~spectro-photometer~~ and the dye ~~M~~m-cresol as indicator. Extinction was measured at 578 nm (E1) and 434 nm (E2) in a 10-cm cuvette. The pH was calculated from the ratio of E1 and E2 (Clayton and Byrne, 1993).

DIC was measured using a ~~colorimetric~~ coulometric AIRICA system (MARIANDA, Kiel) measuring the infrared absorption after N_2 purging of the sample and calibration with certified reference material (CRM; Dr. A. Dickson, University of California, San Diego).

~~A_T was determined using potentiometric titration and a Gran type data analysis following Dickson et al (2007). The quality of the measurement was verified with the same reference material as used for the DIC measurements~~.

The fCO_2 was calculated from DIC, pHT, salinity and~~,~~ ~~temperature, phosphate, and silicate data~~ using the ~~CO_2SYS program (Pierrot and Wallace, 2006)~~ stoichiometric equilibrium constant for carbonic acid of (Mehrbach et al., 1973)~~{Mehrbach, 1973 #2799}~~ as refitted by (Lueker et al., 2000).

2.~~3~~2.2 Chlorophyll and inorganic nutrients

Subsamples of 500 ml were filtered onto GF/F-filters, ~~which were then homogenized~~. Chl*a* was extracted in acetone (90 %) in plastic vials by homogenisation of the filters for 5 min. in a cell mill using glass beads ~~in a cell mill~~. After centrifugation (10 min., 800 x g, 4°C) the supernatant was analysed on a fluorometer (TURNER 10-AU) at an excitation of 450 nm and an emission of 670 nm to determine Chl *a* concentrations (Jeffrey and Welschmeyer, 1997).

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A segmented continuous-flow analyzer coupled with a liquid-waveguide capillary flow-cell (LWCC) of 2 m length was used to determine phosphate (PO_4) and the sum of nitrite and nitrate (NO_2+NO_3) at nanomolar precision (Patey et al., 2008). The PO_4 determination was based on the molybdenum blue method of Murphy and Riley (1962), and NO_3+NO_2 on the method of Morris and Riley (1963). PO_4 concentrations from the same subsample were also measured manually using a 5-cm cuvette (Grasshoff et al., 1983). In most of the samplings PO_4 data obtained from both methods did not differ significantly (paired t-test: p=0.0262, t=1.127, n=109).

2.32.3 Dissolved organic phosphorus (DOP)

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. The thawed samples were oxidized in a microwave (MARS*X*press, CEM, Matthews, USA)(Johnes and Heathwaite, 1992) after the addition of potassium peroxydisulfate in an alkaline medium (Bhaya et al., 2000). The P concentration, measured as PO_4 in a 10-cm cuvette, represents the total dissolved phosphorus (DP) concentration. DOP was calculated as the difference between the DP concentrations in the filtered and digested samples and the corresponding PO_4 concentration analyzed as described above.

2.23.4 Dissolved organic phosphorus compounds

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 0.2-µm cellulose acetate filters to remove picoplankton. Subsamples were prepared for storage according to the specific method used for each compound. After the analyses, the phosphorus content of measured DOP compounds was summed and the amount subtracted from the total DOP concentration. The difference is defined as the uncharacterized DOP.

Dissolved ATP

The method of Björkman and Karl (2001), adapted to Baltic Sea conditions (Unger et al., 2013), was used to determine dissolved adenosine triphosphate (dATP). A $Mg(OH)_2$ precipitate, including the co-precipitated nucleotides, was obtained by treating 200 ml of the filtrate with 2 ml of 1 M NaOH (0.51% v/v). The precipitate was allowed to settle overnight and then centrifuged at 1000 g for 15 min. The supernatant was discarded and the precipitate was transferred into 50-ml Falcon tubes, centrifuged again (1.5 h, 1680 x g). The resulting pellet was dissolved by drop-wise addition of 5 M HCl. The samples were frozen at -20°C until further

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processing. The pH of the thawed samples was adjusted to 7.2 by the addition of TRIS buffer (pH 7.4, 20 mM). The final volume was recorded. The dATP concentrations were measured in triplicate using the firefly bioluminescence assay and a Sirius luminometer (Berthold Detection Systems Pforzheim, Germany), as described by Unger et al. (2013). Standard concentrations were prepared as described above, using aged Baltic Sea water and six ATP concentrations (adenosine 50-triphosphate disodium salt hydrate, Sigma-Aldrich, A2383) ranging from 1 to 20 nmol l^{-1}. The detection limit of the bioluminescence assay was 2.5 npmol ~~ml~~L^{-1}. The fluorescence slope of the standard concentrations was used to calculate dATP concentrations, correcting for the final sample volume. The P-content of the ATP (ATP-P) was calculated by assuming that 1 mol of ATP is equivalent to 3 mol P.

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Dissolved phospholipids

The phosphate content of the dissolved phospholipids (PL-P) was analyzed using a modified method of Suzumura and Ingall (2001, 2004). Briefly, 400-ml subsamples of the filtrate were stored at -20°C until further processing. The samples were then thawed in a water bath at 30°C and extracted twice with 100 ml of chloroform. The chloroform phase was collected, concentrated to 5 ml in a rotary evaporator (Heidolph Instruments, Schwabach, Germany), and then transferred into microwave tubes. The chloroform was completely evaporated by incubating the tubes in a 60°C water bath overnight. After the addition of 20 ml of deionized water (Milli-Q, Millipore), the samples were digested with potassium peroxydisulfate in alkaline medium and microwaved as described for the DOP analysis. Six standard concentrations of phospholipids, ranging from 0 to 125 µg l^{-1}, were prepared by adding the respective amounts of a stock solution containing 5 mg of L-phosphatidyl-DL-glycerol sodium salt (PG, Sigma Aldrich, P8318) ml^{-1} to the aged seawater. The detection limit was 0.8 nmol l^{-1}. The blanks contained only chloroform and were processed as for the samples.

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Dissolved DNA and RNA

Dissolved DNA and RNA (dDNA and dRNA) concentrations were determined according to Karl and Bailiff (1989) and as described by Unger et al. (2013). For each sample, 200 ml of the filtrate was gently mixed with the same volume of ethylene-diamine-tetracetic acid (EDTA, 0.1 M, pH 9.3, Merck, 1.08454) and 4 ml of cetyltrimethyl-ammonium bromide (CTAB, Sigma-Aldrich, H5882) and stored frozen at –20°C for at least 24 h. After thawing the samples, the precipitate was collected onto combusted (450°C, 6 h) glass fiber filters (25 mm, GF/F Whatman), placed into annealed vials, and stored frozen at –80°C until further analysis.

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DNA concentrations were measured using a fluorescence-spectrophotometer (Hitachi F 2000), and RNA concentrations using a dual-beam UV/VIS-spectrophotometer U3010 (Hitachi). Coupled standards (DNA + RNA) containing 1–10 µg DNA (Sigma Aldrich, D3779) l^{-1} and 20–120 µg RNA (Sigma Aldrich, R1753) l^{-1} were prepared in aged seawater as described above. A reagent blank served as the reference and aged seawater as the background control. The P-contents of the DNA and RNA were calculated by multiplying the measured values by a factor of 2.06 nmol P per µg dDNA and 2.55 nmol P per µg dRNA. The latter values were determined by the microwave digestion of standard substrates.

2.~~2~~3.5 Particulate organic phosphorus, carbon, and nitrogen

Particulate ~~organic~~ phosphorus (P~~O~~P) 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). The measured PO_4 concentration represents total phosphorus (TP). PP is the difference between the total PO_4 concentration in the unfiltered digested sample and the sum of DOP+PO_4. In the "filter-method", 500 ml subsamples were filtered onto pre-combusted GF/F-filters that were then placed into Schott bottles containing 40 ml of deionised water. PP was digested to PO_4 by the addition of oxidizing decomposition reagent (Oxisolv®, Merck) followed by heating in a pressure cooker for 30 min. The PO_4 concentrations of the cooled samples were determined spectrophotometrically according to Grasshoff et al. (1983) . Paired t-test revealed significant differences between ~~both~~ two methods; however, ~~both~~ the difference between the means ~~of 0.19 ± 0.03 µmol l^{-1} for~~ of the filter method and ~~of 0.16 ± 0.04 µmol l^{-1} for~~of the aqueous method (0.19 ± 0.03mmol l-1and 0.16 ± 0.04µmol l-1, respectively) ~~differed~~ were near the detection limit (0.02 µmol l^{-1}) of the methods. Thus, solely the mean values obtained from both measurements are used in the following.

Particulate carbon (PC) and nitrogen (PN) were analyzed by filtering 500 ml samples onto pre-combusted (450°C, 6 h) glass fiber filters (Whatman GF/F), which were then stored frozen at -20°C. PC and PN concentrations were measured by flash combustion of the dried (60°C) filters using an EuroEA elemental analyser coupled with a Conflo II interface to a Finnigan Delta^{Plus} mass spectrometer and include organic and inorganic matter.

2.~~2~~3.6 Phosphate and ATP uptake

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311 PO_4 uptake was measured by addition of radioactively labeled phosphate [^{33}P]PO_4 (specific
312 activity of 111 TBq mmol^{-1}, Hartmann Analytic GmbH) at concentrations of 50 pmol l^{-1} to 50
313 ml subsamples, which were then incubated under laboratory light and the *in situ* temperatures for
314 ~2 h. For each mesocosm, three parallel samples and a blank were prepared. The blank was
315 obtained by the addition of formaldehyde (1% final concentration) 10 min before radiotracer
316 addition, in order to poison the samples. At defined time intervals within the incubation, 5-ml
317 subsamples were taken from each of the parallel samples and filtered onto polycarbonate (PC)
318 filters pre-soaked with a cold 20 mM PO_4 solution to prevent non-specific [^{33}P]PO_4 binding. The
319 filters were rinsed with 5 times 1 ml of particle-free bay water and placed in 6-ml scintillation
320 vials. Scintillation liquid (4 ml IrgaSafe; Perkin Elmer) was added and the contents of the vials
321 were mixed using a vortex mixer. After allowing the samples to stand for at least 2 h, the
322 radioactivity on the filters was counted in a Perkin Elmer scintillation counter. PC-~~F~~filters of 0.2
323 and 3 µm pore sizes (Whatman and Millipore, respectively) were used to determine uptake by
324 the whole plankton community and the size fraction >3µm, respectively. Picoplankton uptake
325 was calculated as the difference between the activity on the 0.2-µm and 3-µm filters.
326
327 [γ^{33}P]ATP (specific activity of 111 TBq mmol^{-1}, Hartmann Analytic GmbH) was added to
328 triplicate 10-ml samples and a blank, each in a 20-ml vial, at a concentration of 50 pmol l^{-1}. The
329 samples were incubated in the dark at the *in situ* temperature for 1 h. The uptake was stopped by
330 addition of 200 µl of a cold 20 mM ATP solution to the samples, which were then filtered and
331 processed as described for the PO_4 uptake measurements.
332
333 **2.3~~2~~.7 Bacterial production (BPP)**
334 Rates of bacterial protein production (BPP) were determined by incorporation of ^{14}[C]-leucine
335 (^{14}C-Leu, Simon and Azam, 1989) according to Grossart et al. (2006). Triplicates and a
336 formalin-killed control were incubated with ^{14}C-Leu (~~213 mCi~~ 7.9 GBq mmol^{-1}; Hartmann
337 Analytic GmbH, Germany) at a final concentration of 165 nmol l^{-1}, which ensured saturation of
338 uptake systems of both free and particle-associated bacteria. Incubation was performed in the
339 dark at *in situ* temperature (between 7.8°C and 15.8°C) for 1.5 h. After fixation with 2%
340 formalin, samples were filtered onto 5.0 µm (attached) nitrocellulose filters (Sartorius, Germany)
341 and extracted with ice-cold 5% trichloroacetic acid (TCA) for 5 min. Thereafter, filters were
342 rinsed twice with ice-cold 5% TCA, once with ethanol (96% v/v), and dissolved with
343 ethylacetate for measurement by liquid scintillation counting. Afterwards the collected filtrate
344 was filtered on 0.2 µm (free-living) nitrocellulose filters (Sartorius, Germany) and processed in

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the same way as the 5.0 µm filters. Standard deviation of triplicate measurements was usually <15%. The sum of both fractions (free-living bacteria and attached bacteria) is referred to total BPP. The amount of incorporated ^{14}C-Leu was converted into BPP by using an intracellular isotope dilution factor of 2 (Simon and Rosenstock, 1992). A conversion factor of 0.86 was used to convert the protein produced into carbon (Simon and Azam, 1989).

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2.~~2.8~~4 Statistical analyses

The Grubbs test, done online (graphpad.com/quickcalcs/Grubbs1.cfm) was applied to identify outliers in all data sets. The outliers were removed from further statistical analyses.

Spearman Rank correlations were carried out to describe the relationship between the development of the parameters over time in the mesocosms and in the fjord using Statistica 6 software.

Short-term CO_2 effects on P~~O~~P concentrations at days 0-2 and 23-43 between the CO_2 treatments were verified with an ANCOVA analysis using the SPSS software. The "days" were treated as a covariate interacting with the treatments. Paired t-test was applied to check the differences in PO_4 concentrations between the treatments.

3. Results

3.1 Development in the mesocosms

3.1.1 CO_2, pH, temperature and salinity

The different mesocosms were characterized based on their averaged fCO_2 and pH values from day 1 until day 43 (Fig.2a,b):

~~M1 and M5: 365 and 368 µmol l^{-1} atm fCO_2, pH 8.08 and 8.07 untreated levels;~~

~~M7 and M6: 497 and 821 µatmmol l^{-1} fCO_2, pH 7.95 and 7.74 intermediate fCO_2;~~

~~M3 and M8: 1007 and 1231 µatmmol l^{-1} fCO_2, pH 7.66 and 7.58 high fCO_2.~~

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M1 365 µatm fCO_2, pH 8.08

M5 368 µatm fCO_2, pH 8.07

M7 497 µatm fCO_2, pH 7.95

M6 821 µatm fCO_2, pH 7.74

M3 1007 µatm fCO_2, pH 7.66

M8 1231 µatm fCO_2, pH 7.58

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

Temperature development in the mesocosms ~~was determined through temperature variations~~ closely followed that in the fjord ranging ~~-~~from 7.8~~1~~2°C to 15.86°C. Based on this compare Paul et al. (2015 for details), the experiment was divided into four phases (Fig. 3): phase 0: day -3 to day 0; phase I: days 1–16, phase II: days 17–30 and phase III: day 31 until the end of the measurements. Temperature dropped from 8.71°C to 7.82°C in phase 0 and rose from 8.07°C at the ~~end~~ start of phase ~~0~~I to the maximum of 15.86°C by the end of this phase ~~I~~. During phase II, the temperature decreased to 7.89°C interrupted by a short reversal on days 22 and 23. During phase III, the temperature increased to 12.61°C (Table 1).

~~The s~~Salinity ~~of (~~5.69±0.01~~)~~ remained relatively stable in ~~the~~ all mesocosms throughout the entire experimental period (Fig. 3).

3.1.2 Phytoplankton biomass

Chlorophyll *a* (Chl*a*) reached maximum concentrations of 2.06–2.48 µg l^{-1} at day 5 (Fig. 4). Average concentrations of 1.94±0.23 µg l^{-1} in phase I exceeded those in phases II and III when Chl*a* decreased to a mean of 1.08±0.16 µg l^{-1}. The increase in Chl*a* in the high CO_2 mesocosms by ~~24%~~ 0.27µg l^{-1} in phase III was ~~statistically significant (Paul et al. 2015b), and differences of 0.27 µg l^{-1} were only~~ marginal for Baltic Sea summer conditions. According to Paul et al (2015), this represents an increase of 24% which is a significant difference compared to the controls.

We observed a significant relationship between Chl*a* and PO_4 in the untreated and intermediate treated mesocosms that ~~was~~ diminished with increasing fCO_2 as indicated by lower p-values. The statistical significance ~~got~~ was lost in the highest fCO_2 mesocosms (Table 2).

3.1.3 Phosphorus Pools

Total phosphorus (TP) concentrations in the mesocosms ranged between 0.49 and 0.68 µmol l^{-1} (Fig. 5a) during the experiment without statistically significant differences between the ~~different~~ CO_2 treatments. Shortly after the bags were closed, the decline in TP concentrations began and continued until the beginning of phase II. On average, TP concentrations decreased from 0.63±0.02 µmol l^{-1} on day −3 to 0.51±0.01 µmol l^{-1} on day 21. Thereafter, the mean TP remained constant at 0.54±0.03 µmol l^{-1} until the end of the measurements. Thus, the loss of phosphorus (116 ± 34 nmol l^{-1}) from the 17-m layer during the 29-day measurement period was calculated to be 4.0 nmol l^{-1} day^{-1}. The decline in TP can be explained by loss through sedimentation of P~~O~~P (Paul et al., 2015b).

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Particulate ~~organic~~ phosphorus (P~~O~~P) concentrations varied from 0.10 to 0.23 µmol l^{-1} in all CO_2 treatments (Fig. 5b, Fig. 6). We expected that the decrease in TP was reflected in P~~O~~P. However, parallel changes occurred only periodically. P~~O~~P concentrations increased during the first 5 days after the bags were closed. This increase was stimulated by ~~the~~ CO_2 ~~treatments~~ additions from day 0 to day 2 (ANCOVA: p=0.004, F=20.811) (Fig. 7a). Subsequently, P~~O~~P declined in parallel with TP until day 21, albeit with a lower amount. Averaged over all mesocosms, TP decreased by 0.12 ± 0.03 µmol l^{-1}, whereas P~~O~~P declined only by 0.06 ± 0.01 µmol l^{-1} during this period. From day 23 until the end of the measurements, P~~O~~P ~~leveled off and~~ remained at relatively constant concentrations; however, P~~O~~P concentrations in the high CO_2 treated mesocosms exceeded those in the other mesocosms significantly (ANCOVA: p<0.0001, F=11.99) (Figs. ~~6~~5b, 7). P~~O~~P developed in parallel with P~~O~~C. The two parameters were positively correlated in the untreated and the intermediate CO_2 treatments, but not in the high CO_2 treatments (Table ~~5~~2). Figures 3 and 6b show that the increase in Chl*a* was delayed by 2–3 days compared to the increase in P~~O~~P during the first growth event. A correlation between P~~O~~P and Chl*a* was detected only for the untreated mesocosms (Table 2).

Dissolved organic phosphorus (DOP) concentrations in the mesocosms ranged between 0.18 and 0.36 µmol l^{-1} constituting 32–71% of the TP pool (Fig. 5). DOP did not change significantly in response to the CO_2 perturbations, and were similar to the concentrations in fjord water. Concentrations of ≥0.3 µmol l^{-1} were measured on days 6 and 7 (phase I) and on day 23 (phase II); the high DOP value in the intermediate CO_2 treatment at day 19 was identified as an outlier according to ~~(~~Grubbs test~~)~~ (Fig. 5c).

In phase I, DOP initially increased in parallel with Chl*a* and BPP but reached its maximum 1–2 days later, after which it decreased only marginally until the end of this phase, independent of changes in BPP and Chl*a* (Fig. 8c, d). In phase II, the peak conformed to that of BPP. DOP correlated with temperature only in the high fCO_2 mesocosms (Table 2). In addition, the composition of DOP did not change with increasing CO_2 (Fig.10). The sum of RNA (~47%) plus the unidentified fraction constituted 98–99% of the DOP pool whereas the other measured compounds ~~delivered~~ contributed only 1–2% (Table 3).

Phosphate (PO_4) concentrations ranged between 0.06 and 0.21 µmol l^{-1}, with ~~variations~~ deviations between the mesocosms ~~occurring~~ only in ~~the~~ nanomolar range. The mean contribution of PO_4 to TP was 25±6%, which was the lowest among all TP fractions (Fig. 6). From the start of the measurements to day 13, PO_4 declined by 0.06 µmol l^{-1} (or 3.5 nmol l^{-1}

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445 day^{-1}) from initial values of 0.16±0.01 µmol l^{-1} (Fig. 5d). Subsequently, concentrations
446 increased again, by an average of 2.6 nmol l^{-1} day^{-1}, until the end of the experiment. There were
447 no significant differences between CO_2 treatments until day 23, when high CO_2 concentrations
448 led to slightly lower PO_4 concentrations (Fig. 5d). Afterwards, PO_4 concentrations in the high
449 *f*CO_2 mesocosms were significantly lower than those in the untreated mesocosms (t=6.51,
450 p=0.0003). This observation is in accordance with the dynamics of P~~O~~P and Chl*a*
451 concentrations, which were significantly elevated in the high CO_2 treatments. Thus, the
452 transformation of PO_4 to P~~O~~P via stimulated biomass formation may have been promoted under
453 high CO_2 conditions in phase III.
454 Since PO_4 was never fully exhausted, phosphorus limitation of phyto- and bacterioplankton can
455 be excluded. This interpretation is supported by the P~~O~~C:P~~O~~P ratios, which varied between 84.4
456 and 161.1 in all treatments (Paul et al.,2015b) deviating only slightly from the Redfield ratio.
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458 ### 3.1.4 Uptake of PO_4 and ATP

459 PO_4 turnover times of 1.5–8.4 days (mean 4.0±1.2 days, n= 112) in all mesocosms indicated no
460 dependency on the CO_2 treatment (Fig. 9a). Gross PO_4 uptake rates were in the range of 0.6–3.9
461 nmol l^{-1} h^{-1} (mean 1.7±0.6 nmol l^{-1} h^{-1}, n=112), or 14.3–94.4 nmol l^{-1} day^{-1} (mean 41.3±13.8
462 nmol l^{-1} day^{-1}) (Fig. 9b, Table 4). The rates were highest on days 4 and 9 (phase I) and
463 decreased thereafter until day 15, followed by an increase to a mean maximum rate of 2.3±0. 5
464 nmol l^{-1} h^{-1} (n=6) at day 27. The size fraction <3 µm was responsible for 59.1 to 98.4% of the
465 total PO_4 uptake (mean 86.5±7.6%) whereas the size fraction >3 µm accounted for only 1.6–
466 40.9% (mean 13.5±7.4%). Thus, PO_4 was taken up mainly by picoplankton. However, only the
467 uptake rate by the size fraction >3 µm was positively related to Chl*a* and inversely related to the
468 P content of the biomass (Table 2). Thus the PO_4 uptake was obviously stimulated when the
469 phytoplankton biomass increased and at simultaneous decrease of the cellular P. The relationship
470 between PO_4 uptake by this fraction and Chl*a* became evident only in the CO_2-amended
471 conditions indicating that the interaction between P uptake, cellular P-content and growth of
472 phytoplankton was stimulated under elevated CO_2 conditions.
473 ATP turnover times of 0.2 to 3.6 days (mean 0.94±0.74 days, n=90) were much shorter than the
474 PO_4 ~~uptake rates~~ turnover times and did not vary between the treatments (Fig. 9c). ~~Turnover~~
475 ~~times were longest on day 9 and shortest on day 4 (Fig. 9c).~~ Between 0.05 and 0.36 nmol ATP
476 l^{-1} h^{-1} (mean 0.14±0.08 nmol l^{-1} h^{-1}, n=36) were ~~degraded~~ hydrolysed, corresponding to a P
477 supply of 0.14 and 1.08 nmol l^{-1} h^{-1} (mean 0.44±0.25 nmol l^{-1} h^{-1}, n=36). Thus, phosphorus
478 additionally supplied from ATP accounted for ~25% of that provided by PO_4. The picoplankton

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size fraction (<3 µm) was responsible for 90–99% of ATP uptake, with only a marginal portion (1.6–9.5%) attributable to the phytoplankton fraction >3 µm (Table 4).

3.~~2~~ ~~Development in the Fjord~~ Hydrography and pool sizes in the fjord

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~~3.2.1 *In situ* CO_2 and pH conditions~~

Large variations in fCO_2 and pH occurred in fjord water during the period of investigation (Table 1). The relationship of fCO_2 with temperature and salinity indicated that the CO_2 conditions were influenced predominantly by changes in the water masses, specifically by upwelling which affected both the relationship of fCO_2 with PO_4 and probably the correlation of fCO_2 with Chl*a* and PC (Table 2). fCO_2 ranged from 207 µatm (Fig. 2a) at days 12-16 when temperatures were highest to 800 µatm at day 33 when deep water input occurred which was indicated by ~~low~~ pH ~~(>~~ below 7.75~~)~~.

~~3.2.2 Phytoplankton biomass~~

Chl*a* concentrations ~~in the fjord~~ were between 1.12 and 5.46 µg l^{-1} (mean 2.29 ± 1.11 µg l^{-1}; n=38), with distinct phases ~~similar to those of~~ correlating with temperature, ~~and~~ salinity and pH. However, the Chl*a* maximum occurred at the beginning of phase II, which was 1-2 days after the maximum temperature. Shortly thereafter, Chl*a* decreased to its lowest level before it increased again, albeit only marginally to 1.93 µg l^{-1} during phase III (Fig. 4). ~~Chl*a* concentrations correlated positively with temperature and pH (Table 5). The correlations of Chl*a* with PC and BPP suggested that phytoplankton determined the development of PC and was associated with bacterial growth.~~

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~~3.2.3 Phosphorus Pools~~

TP concentrations from day -3 until day 29 ranged between 0.54 and 0.70 µmol l^{-1} (mean 0.61±0.04 µmol l^{-1}; n=19) (Figs. 5a, 6a). ~~The progression of TP differed from that of the hydrographic parameters or the Chl*a* concentrations~~. With a general decreasing tendency, TP undulated with a frequency of about 10 days in the period of phases 0 to the first half of phase I and of 6 days in the second half of phase I to II. For the period under investigation, the TP fractions had the following characteristics:

P~~O~~P concentrations varied from 0.13 to 0.30 µmol l^{-1} (mean 0.20±0.04 µmol l^{-1}; n=29), thus accounting for 23.4–51.8% (mean 34.7±7.9%; n=19) of the TP pool. The development of P~~O~~P over time did not follow that of TP (Fig. 6b). P~~O~~P concentrations were highest between days 8 and 19, when the accumulation of P~~O~~P in the biomass was reflected in declining C:P ratios from

180 to 107 (Paul et al. 2015) and thereafter remained at the low ratio until the end of the measurements. The P~~O~~P increase in phase III occurred in parallel to Chl*a* and to the PO_4 decrease (Fig. 6) ~~(Table 5)~~. Thus PO_4 was transformed into P~~O~~P via biomass production. The calculated P content of phytoplankton was 0.05–0.15 (mean 0.1) µmol P~~O~~P (µg Chl*a*)$^{-1}$.

DOP substantially contributed (26–45%) to the TP pool (Fig. 6). Concentrations ranged between 0.19 and 0.29 µmol l^{-1} (mean 0.24±0.03 µmol l^{-1}; n=17), with high concentrations occurring in parallel to those of TP in phases I and II (Fig. 5c). The very low DOP value of 0.11 µmol l^{-1}, on day 29, was an outlier (Grubbs test) ~~and was excluded from the calculation~~. For the whole study period, DOP concentrations correlated positively with ~~both~~ P~~O~~P (p= 0.034, n=17).and ~~PO$_4$ turnover times and~~ inversely with PO_4 concentrations (p=0.005, n=17) ~~(Table 5)~~. A similar behavior between DOP and Chl*a* was restricted to phases 0 and I, whereas the relationship was inverse in phase II (Fig. 8b) ~~indicating that upwelling of deep water did not change the DOP concentrations in surface water~~. As shown in Figure 8a, the DOP and BPP levels alternated with the same rhythm, but inversely, in phases 0 and I and changed to a parallel development in phase II. Statistical analysis was not feasible because DOP and BPP were not always sampled on the same day and only very few data pairs were available.

~~Phosphorus, derived from the sum of ATP, PL, RNA, and DNA, constituted 42.8–72.0% (mean 59.7±10.7%; n=7) of the DOP pool (Table 3). Thus, 27.8–57.2% of the DOP remained unidentified. Concentrations of 1.4–4.6 nmol ATP l^{-1}, 0.6–4.5 nmol PL l^{-1}, 42.2–163 µg RNA l^{-1}, and 0.03–0.06 µg DNA l^{-1} were measured, yielding 3.1–13.8 nmol ATP-P l^{-1}, 0.6–4.5 nmol PL-P l^{-1}, 42.2–163 nmol RNA-P l^{-1}, and 0.06–0.13 nmol DNA-P l^{-1}. Thus, the contribution of RNA to the DOP pool was the highest, whereas the contributions of ATP, PL, and DNA were relatively small (Table 3). The changes in all of these components over time were not related to changes in the total DOP pool (Fig. 10).~~

PO_4 concentrations ranged between 0.06 and 0.41 µmol l^{-1} (mean 0.21±0.09 µmol l^{-1}, n=21), thus comprising 24.3±11.2% (n=21) of the TP pool (Fig. 6). With a few exceptions, PO_4 concentrations declined from the beginning of the study period until the end of phase I and increased during phase II and the beginning of phase III. These changes were caused by upwelling of PO_4 enriched deep water of higher salinity and lower temperatures. The subsequent decline in PO_4 between days 33 and 40 was caused by the stimulation of phytoplankton production, as indicated by the increase in Chl*a* concentration (Fig. 4). ~~For the whole~~

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experimental period, the Spearman rank correlation showed an inverse relationship between PO_4 and particulate organic matter such as Chl*a*, PC, and PN (Table 5).

3.2.4. Uptake of PO_4 and ATP

Applying [^{33}P]PO_4, PO_4 turnover times in the fjord were in the range of 30–379 h (1.2–15 days) (mean 139±98 h, n=18) (Fig. 9a), corresponding to uptake rates of 0.73–3.37nmol l^{-1} h^{-1} (mean 1.64±0.82 nmol l^{-1} h^{-1}, n=18) (Table 4). These rates were influenced by multiple factors, including temperature, phytoplankton biomass, and DP, POP, and PO_4 concentrations as deduced from Table 5. Despite the paucity of data pairs, the total PO_4 uptake rate correlated with total BPP and with BPP in the fraction <5 µm (r=0.886, p=0.0188; n=6 for each relationship).

Within the experimental period, the turnover times shortened on days 15–17 (Fig.9a), when temperature and Chl*a* (Figs. 3, 4) reached a maximum and PO_4 concentrations were lowest (Figs. 5d). Although the shortest turnover times were expected to be coupled with the highest uptake rates, the latter were estimated 2 days later, between days 17 and 19. The day-to-day variations conformed to the small changes in temperature and PO_4 concentrations. Uptake was dominated by the size fraction <3 µm in most of the measurements (Table 4), which accounted for 17.4–92.3% (mean 72.2±20.6%) of the total uptake rate. The mean contribution of the size fractions >3 µm was 27.8±20.6%. Assuming that autotrophic organisms were largely responsible for the uptake by this fraction, the specific PO_4 uptake rates of phytoplankton, calculated from the size fraction >3 µm and the Chl*a* concentration, were 0.02 and 0.46 nmol (µg Chl*a*)$^{-1}$ h^{-1}.

The turnover times of ATP (7.5–62 h, or 0.3–2.6 days; mean 23±15 h or 0.96±0.59 days; n=15) were significantly shorter than those of PO_4 (Fig. 9c), without any apparent relationship between the two. The longest ATP turnover times of 2.6 and 1.7 days occurred on days 9 and 15, respectively, when PO_4 turnover times were short. Based on measured ATP concentrations, 0.03–0.15 nmol ATP l^{-1} h^{-1} was converted, thus delivering 0.13–0.46 nmol P l^{-1} h^{-1} (Table 4). The size fraction <3 µm utilized 85.0–97.8% (mean 92.4±4.7%) of the ATP whereas only a small portion (2.2–15%) could be attributed to the size fraction >3 µm. ATP uptake rates and concentrations did not correlate with any of the other measured parameters (data not shown), with the exception that ATP turnover time correlated with BPP >5 µm (r=0.943, p=0.048, n=6).

4. Discussion

An increase in CO_2 in marine waters and the associated acidification may potentially have multiple effects on organisms and biogeochemical element cycling (Gattuso and Hansson, 2011).

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~~However, r~~Reported findings indicate wide ranging responses, probably depending on the investigated species and growth conditions. For example, CO_2 stimulation as well as lack of stimulation were found for primary production and carbon fixation (Beardall et al., 2009; Boettjer et al., 2014), DOC release (Engel et al., 2014; MacGilchrist et al., 2014) and phytoplankton growth (Riebesell and Tortell, 2011). An interaction of CO_2 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 multiseries* and for a diatom dominated subarctic plankton community, respectively. Thus, ~~the~~ responses of organisms and ecosystems to enhanced CO_2 concentrations are complex and still poorly understood. The present study is the first to determine the effects of increased CO_2 levels on ~~the~~ phosphorus cyclinge in a brackish water ecosystem.

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4.1 Response of P-pools and P-uptake to enhanced CO_2 in the mesocosms

The Finish ~~side~~ coast of the Gulf of Finland is one of the most important upwelling regions in the Baltic Sea. During our investigation in 2012, surface temperatures, obtained from the NOAA satellite (Siegel and Gerth, 2013) showed that upwelling persisted during the whole study period but with varying intensity. The intensity of upwelling shaped the pattern of temperature not only in the fjord ~~and~~ but also in the mesocosms varying from 7.8 to15.9°C. Such variations in temperature influence the phosphorus transformation and interleave with CO_2 effects.

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While nutrients were added in previous ~~mesocosms~~ CO_2 enrichment experiments (Riebesell et al., 2008; 2013; Schulz et al., 2008), no amendments were undertaken in this study in order to be close to natural conditions. Initial PO_4 concentrations of only 0.17 ± 0.01 µmol l^{-1} were measured, however, PO_4 was never exhausted (Figs. 5, 6). Cellular C:P and N:P ratios were close to the Redfield ratio. Therefore, phosphorus limitation unlikely occurred in this experiment. Simultaneous low nitrate and ammonium concentrations (Paul et al. 2015b) formed nutrient conditions that benefit the growth of diazotrophic cyanobacteria. However, a~~ny~~ cyanobacteria bloom failed to appear, despite the low-level presence of *Aphanizomenon* sp. and ~~*Anabaen*~~ *Dolichospermum*~~a~~ sp. (Paul et al., 2015a) as potential seed stock. For Baltic Sea summer conditions, the phytoplankton development with maximum Chl*a* concentrations of 2.2–2.5 µg l^{-1} remained relatively low with the highest contribution of cryptophytes and chlorophytes in phase I and at the beginning of phase II. Picoplankton was mostly the dominating size fraction, amounting ~20-70% of Chl*a* in phase I and ~~rising~~ up to ~85% in phase III (Paul et al., 2015b). However, a positive correlation of fCO_2 with ~~the~~ Chl*a* was observed only for the size

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fraction >20 µm ~~was estimated~~. The abundance of diatoms that could be a part of this fraction increased from ~day 23 to day 30 and might have an influence on this relationship.

Against this background, the CO_2 perturbation did not cause significant changes in phosphorus pool sizes, DOP composition, and P-uptake rates from PO_4 and ATP when the whole study period was considered. However, small but ~~nevertheless~~ yet significant~~,~~ short-term effects on PO_4 and P~~O~~P pool sizes were observed in phases I, ~~and~~ III and partially in phase II (Fig. 7). CO_2 elevation stimulated the formation of P~~O~~P until day 3 (Fig. 5b) when chlorophytes, cyanobacteria, prasinophytes and the pico-cyanobacteria started to grow (Paul et al., 2015b).

The effects of CO_2 addition on PO_4 and P~~O~~P pool sizes were evident from day 23 onwards (Figs. 5b, 7). PO_4 concentrations were slightly, but significantly lower in the high CO_2 treatment than in the untreated mesocosms, accompanied by significantly elevated P~~O~~P concentrations. This indicates ~~indicating~~ that the transformation of PO_4 into P~~O~~P was likely stimulated under high CO_2 conditions. Since Chl*a* was also elevated ~~as well~~ at similar P~~O~~P:Chl*a* ratios, the PO_4 taken up was used for new biomass formation. However, the elevated transformation of PO_4 into P~~O~~P was not ~~detected~~ reflected in the PO_4 uptake rates which can be seen as gross uptake rates. But, an increase of PP, caused by biomass formation, while the PO_4 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~~, e.g. via reduction in P release from biomass~~ under CO_2 elevation.

~~It is hard to assess the short-term effects that we have found in phase I~~While in phases II/III elevated CO_2 caused a change in the PP and PO_4 pools for about 22 days, achanges lasting only 2 days have been observed at the beginning of phase I (Fig.7a), but, shorter effects cannot be excluded. Uptake and release are assumed to be continuous processes and can alter the P pool sizes on timescales shorter than one day. Thus, variations and differences in the treatments can be overseen at daily sampling. Unger et al. (2013) demonstrated that an accelerated PO_4 uptake by the cyanobacterium *Nodularia spumigena* under elevated CO_2 incubations could only be observed during the first hours. Thereafter, the differences were balanced and the same level of radiotracer labeling was reached in all treatments. An acceleration in the ~~-~~formation of particulate P ~~concentrations~~ under CO_2 elevation without any changes of PO_4 turnover times was also observed by Tanaka et al. (2008). They observed an increase of the P~~O~~P amount and an earlier appearance of the P~~O~~P maximum under CO_2 elevation.

Correlations calculated by using the Spearman rank test between P pools or uptake rates and other parameters for each mesocosm are presented in Table 2. The relationships between P~~O~~P

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and TP with Chl*a* disappeared at elevated fCO_2, whereas correlations developed between P~~O~~P and PC as well as between the PO_4 uptake by phytoplankton in the >3 µm size class and the P~~O~~P:Chl*a* ratio (Table 2). These shifts could be caused by changes in the phytoplankton composition deduced from CO_2 effects on the pigment composition (Paul et al., 2015b).

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Independent of the CO_2 treatment, TP decreased by 2.6 nmol l^{-1} day^{-1} in all mesocosms over the course of the experiment, in agreement with the measured sedimentation rates (Paul et al., 2015b). The strongest decrease (~3.2 nmol l^{-1} day^{-1}) occurred during phase I. Of the total TP removal during this phase (48 nmol l^{-1}), 84% (~40.5 nmol l^{-1}) could be explained by the decrease in P~~O~~P and 16% (~ 8 nmol l^{-1}) by changes in the dissolved pool. However, the PO_4 decline (~34.5 nmol ~~l~~$^{-1}$) was stronger than that of the total dissolved P pool since DOP increased in parallel by ~26.5 nmol ~~l~~$^{-1}$. Thus, about 77% of the PO_4 reduction was retrieved as DOP and remained in the dissolved P-pool ~~being~~ as the main pathway of PO_4 transformation.

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4.~~3~~2 Phosphorus dynamics in the Storfjärden

~~Measurements of P-pool sizes and P uptake in the fjord provided new information about the phosphorus dynamics in a Baltic Sea upwelling system and in times when diazotrophic cyanobacteria did not dominate the phytoplankton community.~~ Nutrients ~~conditions were mainly determined by~~ in upwelled water~~s~~ during our study ~~, which~~ were depleted in dissolved inorganic nitrogen and enriched in PO_4, as reported for other upwelling areas of the Baltic Sea (Lass et al., 2010). Thus, ammonium and NO_2 ~~+~~ NO_3 concentrations in the surface water were only in the nanomolar range (Paul et al., 2015b). PO_4 increased in parallel with the increase in salinity and decrease in temperature~~, indicating their coupling with upwelling (Table 5)~~. Maximum PO_4 concentrations of 0.33 µmol l^{-1} and 0.42 µmol l^{-1} (Figs 5, 6) were observed at the end of the upwelling events in phases 0 and II, respectively. The correlation with Chl*a* and P~~O~~P indicated that PO_4 was utilized during plankton growth in the subsequent relaxation phases I and III. ~~However, PO_4 was not fully depleted (Fig. 6d) which can be attributed to "low" P-demand of organisms as deduced from the relatively low PO_4 uptake rates in fjord water and in the mesocosms. As in the mesocosms~~ Due to PO_4 input into surface water, the phytoplankton community was unlikely P-limited indicated by PC:P~~O~~P ratios of 86–189 (mean 125, n=23) (Paul et al., 2015b). ~~The close correlation of POP with Chl*a* indicated a large contribution of phytoplankton to particulate~~ P. However, the PO_4 availability might be not the only reason for the good P-nutritional status of the plankton. It can be deduced from the long PO_4 turnover times

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in the mesocosms, where external input was excluded, that the P demand of the plankton community might be low.

~~However, its~~ The P content deduced from P~~O~~P:Chl*a* ratios of 0.05–0.15 µmol P (µg Chl*a*)$^{-1}$ was somewhat lower than those observed during an upwelling event along the east coast of Gotland, where ~~the~~ ratios ~~were~~ between 0.1 and 0.2 µmol P (µg Chl*a*)$^{-1}$ were estimated (Nausch et al., 2009).

P~~O~~P concentrations of 0.13–0.3 µmol l^{-1} were in the range typically observed in the Baltic Proper (Nausch et al., 2009; Nausch et al., 2012~~)~~. However, P~~O~~P concentrations in the Gulf of Finland may reach higher values, as was the case in the summer of 2008, when the observed P~~O~~P concentration was 0.35 ± 0.07 µmol l^{-1} (Nausch and Nausch, 2011).

DOP concentration of 0.27 ± 0.02 µmol l^{-1} during our study was similar to that detected in the Gulf of Finland in the summer of 2008 (Nausch and Nausch, 2011). In the Baltic Sea, DOP exhibits vertical gradients with maximum concentrations in the euphotic surface layer ~~(Nausch and Nausch, 2011)~~ and lower than 0.1 µmol l^{-1} at depths below 25 m. Thus, the observed DOP dynamics in surface water during our study can be assumed to be the result of release, consumption and mineralization by organisms or input from land. ~~The DOP increase in phase I coincided with increases in Chl*a* and initially BPP (Fig. 8), while the development of DOP and BPP showed opposing trends in the second part of phase I and thereafter. Thus, the increased DOP concentrations in phase I were due to release by phytoplankton supplemented by bacterial release exceeding the consumption or degradation. During phase II, phytoplankton biomass was low and DOP release should thus be minor. Since the small mesozooplankton increased in the fjord similar to those reported for the mesocosms in phases II and III (Paul et al. 2015b) DOP could be released during grazing combined with the observed temporal offset of BPP and DOP maxima.~~ The relationship of DOP with Chla and BPP (Fig. 8) indicated that the increased DOP concentrations in phase I were due to release by phytoplankton supplemented by bacterial release. DOP can be accumulated in water only when the release exceeded the consumption or degradation. During phase II, phytoplankton biomass was low and DOP release should thus be minor. Since the small mesozooplankton increased in the fjord similar to those reported for the mesocosms in phases II and III (Paul et al. 2015b) DOP could be released during grazing combined with the observed temporal offset of BPP and DOP maxima. Thus the observed DOP variations may be the result of processes in the surface water.

~~The DOP concentration of 0.27 ± 0.02 µmol l^{-1} during our study was similar to that detected in the Gulf of Finland in the summer of 2008 (Nausch and Nausch, 2011). On average, more than~~

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~~half (59.1%) of the DOP consisted of the measured compounds ATP, PL, DNA, and RNA; the other sources remained uncharacterized. ATP levels in the Storfjärden were approximately ten times higher than in the surface water of the subtropical Pacific (Björkman and Karl, 2001) but were similar to those measured during a spring bloom in the Antarctic (Nawrocki and Karl, 1989).~~

~~The contribution of PL to the DOP pool was in the same range as reported by Suzumura (2005) whereas the contribution of DNA was relatively small. DNA concentrations were much lower than either those measured in the northern Baltic Sea (Riemann et al., 2009) or those reported by Karl and Bailiff (1989) for the Pacific Ocean. RNA, however, was the dominant DOP component, contributing about half of the DOP pool in this study. In studies of Karl and Bailiff (1989) in various marine systems RNA concentrations were 6–10 times higher than DNA concentrations. However, they have measured such high RNA concentrations as detected in our study only in a pond.~~

~~The uptake of phosphorus from radioactively labeled ATP is used to monitor DOP utilization (Karl and Björkman, 2002). However, ATP is a component of the labile P fraction and is thus preferred over other substrates (Siuda and Chrost, 2001). The mean ATP turnover times of 23 ±14 h were similar to those measured in the Gotland Basin in May and June 2001 (Nausch et al. 2004), when temperatures were below 12°C. During mesocosm experiments at the Tvärminne station in July 2003 (Lovdal et al., 2007), ATP turnover times at temperatures >18°C were between 2 and 6 h. In our study, 0.04–0.51 nmol ATP-P l^{-1} h^{-1} were taken up mainly (85–98%) by pico-sized organisms, providing them with ~15% of their P requirement; instead the main phosphorus source of phytoplankton during our study was PO_4. In their study of the Sargasso Sea, Michelou et al. (2011) found that, on a cellular basis, more ATP was utilized by the cyanobacterium *Synechococcus* than by heterotrophic bacteria. However, Casey et al. (2009) found that *Prochlorococcus* and *Synechococcus* accounted for only 3–20% of the total ATP uptake. In our study, ATP uptake rates correlated with the contribution of the fraction <2 µm to total Chl*a* whereas no such correlation could be established for BPP. These observations might be an indication that autotrophic picoplankton dominates the ATP uptake.~~

~~PO_4 turnover times varied between 1 and 5 days. Longer turnover times (10–15 days) occurred only in phase 0 while the shortest turnover time (1 day) was at the end of phase I, when temperatures and phytoplankton biomass were highest and PO_4 concentrations lowest. Nevertheless, a 1 day turnover time indicated no P limitation, as under P limited conditions reported turnover times are <1 h (Nausch et al., 2004). The PO_4 uptake rates of 0.9–2.8 nmol l^{-1}~~

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~~h^{-1} and the specific uptake rates of 0.02–0.46 nmol (µg Chl*a*)$^{-1}$ h^{-1} were similar to the rates measured in the eastern Gotland Basin in the summers of 2007 and 2009 (Nausch et al., 2009; 2012). As in the summer of 2001, two thirds of the PO_4 uptake was realized by the size fraction <3 µm, although with progressive PO_4 depletion this percentage may rise to ~90% (Nausch et al., 2004). This result suggests that picoplankton has an advantage in the competition for phosphorus at low concentrations. The close correlation of total PO_4 uptake and BPP <5 µm (Table 2) suggests that free-living heterotrophic bacteria were the main consumers of PO_4. This relationship was probably determined by the 3- to 5 µm size fraction, since there was no apparent correlation between BPP <5 µm and PO_4 uptake by the size fraction <3 µm (Table 2).~~

~~The phosphorus demand of heterotrophic bacteria is influenced by carbon and nitrogen availabilities. The shorter turnover times of ATP, DNA, and PO_4 following nitrogen and carbon amendments in the mesocosm experiments of Lovdal et al. (2007) in July 2003 suggested that the bacterioplankton community at Tvärminne station is C- and N-limited. This may also have been the case during our study. Thus, the relatively long PO_4 turnover times might have been caused not only by low temperatures but also by the reduction in bacterioplankton activities due to C and N limitations and could be the reason that PO_4 was not depleted completely in the mesocosms and in fjord water.~~

5. Conclusions

Surface water in Storfjärden showed highly variable fCO_2 conditions and reached levels of up to 800 µatm, which is similar to that expected in ca. 100 years from now. Deduced from the high frequency of upwelling events ~~there~~, organisms ~~are confronted~~ experience ~~with~~ elevated fCO_2 more or less regularly ~~and are used to high fCO_2 variability~~. ~~This could explain the minimal response of the phytoplankton community~~. ~~A~~ Thus, a general impact of fCO_2 on P pools and P uptake rates in the mesocosms could not be identified for the overall period of investigation. However, ~~temporary~~ short-term responses to fCO_2 elevation lasting only few days were observed for the transformation of PO_4 into P~~O~~P that ~~,~~ was linked with stimulation of phytoplankton growth. Although statistically significant, it is difficult to assess if the differences between the treatments are of ecological relevance. ~~Potentially, s~~Such short-term variations are possible in the phosphorus dynamics since ~~the transformation can take place on hourly scales,~~ the pools size can be transformed within hours and ~~transformations~~ there changes are in the nanomolar concentration range. There are also indications that relationships of P pool sizes or uptake with Chl*a* and PC can change as fCO_2 increases, but the underlying mechanisms are still

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unclear. ~~This would have an effect on biogeochemical cycles. This study also provides information on the phosphorus cycle in an upwelling driven ecosystem of the Baltic Sea. P pool sizes were in the range characteristic for spring and cooler summers when low temperatures inhibit cyanobacteria bloom formation.~~ The transformation of PO_4 into DOP was not affected by CO2 elevation. It may be the major pathway of phosphorus cycling under hydrographical and phytoplankton growth conditions as occurred in our experiment.

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Tables and figures

Table 1: Minimum, maximum and mean values of hydrographical parameters and fCO_2 for the different phases in the fjord. Temperatures in the mesocosms were identical with those in surrounding fjord water.

phase	min	max	mean
	water temperature (°C)		
0	7.82	8.71	8.20
I	9.66	15.86	12.27
II	7.89	14.79	11.68
III	8.35	12.61	10.83
	salinity		
0	5.72	5.85	5.78
I	5.46	5.85	5.65
II	5.67	6.04	5.82
III	5.9	6.05	5.98
	pH		
0	8.09	8.23	8.16
I	8.11	8.30	8.17
II	7.81	8.30	8.00
III	7.75	7.93	7.83
	fCO_2 (µatm)		
0	250	347	298
I	207	336	283
II	208	679	465
III	521	800	668

Table 2: Mesocosms in which the Spearman Rank correlation between P-pools or uptake rates and other parameters was significant. The relationship of POP with TP and Chl*a* was significant only in the untreated mesocosms while the correlation to PC was also significant in the mesocosms with intermediate CO_2 levels. DOP was related to temperature only in the high CO_2 treatments. Under high fCO_2 conditions, the PO_4 uptake in the size fraction >3µm correlated with Chl_a and the P content of phytoplankton.

Relationship between	***f*CO$_2$ (µatm)**	**significant responses** r	p	n
POP-TP	365	0.599	0.008	18
	368	0.515	0.029	18
POP - Chl*a*	365	0.479	0.0130	25
	368	0.584	0.0022	25
PO_4 - Chl*a*	365	-0.832	<0.0001	21
	368	-0.756	0.0011	20
	497	-0.674	0.0008	21
	821	-0.524	0.0147	21
	1007	-0.634	0.0027	20
POP -PC	365	0.542	0.0061	24
	368	0.625	0.0011	24
	497	0.404	0.0490	24
	821	0.551	0.0052	24
DOP -temperature	1007	0.488	0.0470	17
	1231	0.525	0.0310	17
PO_4 uptake>3µm - Chl*a*	497	0.743	0.0056	12
	821	0.674	0.0081	14
	1231	0.476	0.0310	14
PO_4 uptake>3µm - POP/Chl*a*	497	-0.601	0.0380	12
	821	-0.631	0.0160	14
	1231	-0.626	0.0165	14

Relationship between	fCO_2 (µatm)	significant responses r	p	n
PP-TP	365	0.599	0.008	18
	368	0.515	0.029	18
PP - Chl*a*	365	0.479	0.0130	25
	368	0.584	0.0022	25
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	821	-0.631	0.0160	14
	1231	-0.626	0.0165	14

Table 3: Contribution of different phosphorus components to DOP in the mesocosms and in the fjord.

fCO_2	contribution to DOP (%)					
(µatm)	ATP-P	PL-P	DNA-P	RNA-P	sum	unidentified P
Fjord	0.7	0.7	0.04	69.4	70.84	29.16
365	0.7	0.5	0.03	44.1	45.33	54.67
368	0.6	0.5	0.03	46.9	48.03	51.97
497	0.6	0.4	0.04	49.5	50.54	49.46
821	0.6	0.4	0.03	41.8	42.83	57.17
1003	0.8	0.4	0.04	60.1	61.34	38.66
1231	0.5	0.4	0.03	48.6	49.53	50.47

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Table 4: PO_4- and ATP uptake rates in the fjord and in the mesocosms. Minimum, maximum and mean values as well as the contribution of the size fraction <3 µm to the total activity are given for the whole period of investigation (each: n= 16 for PO_4 and n=6 for ATP uptake).

fCO_2	total PO_4 uptake ($nmol l^{-1} h^{-1}$)			portion (%)	total ATP-P uptake ($nmol l^{-1} h^{-1}$)			portion (%)
(µatm)	min	max	mean	<3µm	min	max	mean	<3µm
Fjord	0.87	2.81	1.63 ± 0.58	76 ± 15	0.04	0.51	0.26 ± 0.15	92 ± 5
365	0.82	3.89	1.67 ± 0.82	81 ± 11	0.14	1.08	0.43 ± 0.33	96 ± 2
368	0.65	2.74	1.61 ± 0.58	86 ± 7	0.16	0.97	0.47 ± 0.27	96 ± 2
497	0.61	3.03	1.52 ± 0.59	86 ± 6	0.20	1.07	0.54 ± 0.28	96 ± 2
821	0.91	2.83	1.60 ± 0.59	88 ± 8	0.14	0.71	0.36 ± 0.21	97 ± 2
1003	0.67	3.79	1.73 ± 0.85	86 ± 6	0.22	0.69	0.39 ± 0.15	97 ± 1
1231	0.87	2.23	1.53 ± 0.43	87 ± 6	0.17	0.67	0.44 ± 0.17	97 ± 2

~~Table 5: Significance level (p) deduced from of Spearman Rank correlations that were calculated between the parameters in fjord water listed in the table.~~

Variable	T	S	fCO_2	PO_4	POP	DOP	Chla	PC	C/P	PO_4 TO-time	total PO_4 uptake	>3µm PO_4 uptake	<3µm PO_4 uptake	total BPP	> 5µm BPP	0.2-5µm BPP
T (°C)		0.0001$^-$	0.0061$^-$	<0.0001$^-$	<0.0001$^+$	n.s.	<0.0001$^+$	0.0083$^+$	n.s.	0.0001$^-$	n.s.	n.s.	n.s.	0.0005$^+$	0.0088$^+$	0.0012$^+$
S	0.0001$^-$		<0.0001$^+$	<0.0001$^+$	0.0030$^-$	0.0445$^-$	<0.0001$^-$	<0.0001$^-$	0.0181-	<0.0001$^+$	0.0244$^-$	0.0284$^-$	n.s.	0.0037$^-$	0.0370$^-$	0.0072$^-$
pCO_2	0.0061$^-$	<0.0001$^+$		<0.0001$^+$	0.0288$^-$	n.s.	<0.0001$^-$	<0.0001$^-$	0.0104$^-$	0.0250$^+$	0.0403$^-$	0.0006$^-$	n.s.	n.s.	n.s.	n.s.
PO_4 (µmol L^{-1})	<0.0001$^-$	<0.0001$^+$	<0.0001$^+$		<0.0001$^-$	0.0049$^-$	<0.0001$^-$	0.0008$^-$	n.s.	<0.0001$^+$	0.0455$^-$	n.s.	n.s.	0.0018$^-$	0.0353$^-$	0.0041$^-$
POP (µmol L^{-1})	<0.0001$^+$	0.0030$^-$	0.0288$^-$	<0.0001$^-$		0.0345$^+$	<0.0001$^+$	0.0016$^+$	n.s.	0.0048$^-$	n.s.	n.s.	n.s.	0.0022$^+$	0.0056$^+$	0.0102$^+$
DOP (µmol L^{-1})	n.s.	0.0445$^-$	n.s.	0.0049$^-$	0.0345$^+$		0.0271$^+$	n.s.	n.s.	0.0099$^+$	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
Chla (µg L^{-1})	<0.0001$^+$	<0.0001$^-$	<0.0001$^-$	<0.0001$^-$	<0.0001$^+$	0.0271$^+$		0.0003$^+$	n.s.	0.0006$^-$	n.s.	n.s.	n.s.	<0.0001$^+$	0.0027$^+$	<0.0001$^+$
PC (µmol L^{-1})	0.0083$^+$	<0.0001$^-$	<0.0001$^-$	0.0008$^-$	0.0016$^+$	n.s.	0.0003$^+$		0.0014$^+$	n.s.	n.s.	0.0386$^+$	n.s.	0.0092$^+$	0.0479$^+$	0.0114$^+$
PN (µmol L^{-1})	0.0007$^+$	<0.0001$^-$	<0.0001$^-$	0.0001$^-$	<0.0001$^+$	n.s.	<0.0001$^+$	<0.0001$^+$	0.0156$^+$	0.0165$^-$	n.s.	n.s.	n.s.	0.0092$^+$	0.0479$^+$	0.0114$^+$
C/P	n.s.	0.0181$^-$	0.0104$^-$	n.s.	n.s.	n.s.	n.s.	0.0014$^+$		n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.
PO_4-TO time (d)	0.0001$^-$	<0.0001$^+$	0.0250$^+$	<0.0001$^+$	0.0048$^-$	0.0099$^+$	0.0006$^-$	n.s.	n.s.		<0.0001$^-$	n.s.	n.s.	n.s.	n.s.	n.s.
PO_4 total uptake (nmol $L^{-1}h^{-1}$	n.s.	0.0244$^-$	0.0403$^-$	0.0455$^-$	n.s.	n.s.	n.s.	n.s.	n.s.	<0.0001$^-$		n.s.	n.s.	0.0188$^+$	n.s.	0.0188$^+$
PO_4uptake >3µm (nmol $L^{-1}h^{-}$	n.s.	0.0284$^-$	0.0006$^-$	n.s.	n.s.	n.s.	n.s.	0.0386$^+$	n.s.	n.s.	n.s.		n.s.	n.s.	n.s.	n.s.
PO_4uptake <3µm (nmol $L^{-1}h^{-}$	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.		n.s.	n.s.	n.s.
BBP total (µg C $L^{-1}h^{-1}$)	0.0005$^+$	0.0037$^-$	n.s.	0.0018$^-$	0.0022$^+$	n.s.	<0.0001$^+$	0.0092$^+$	n.s.	n.s.	0.0188$^+$	n.s.	n.s.		<0.0001$^+$	<0.0001$^+$
BPP >5µm (µg C $L^{-1}h^{-1}$)	0.0088$^+$	0.0370$^-$	n.s.	0.0353$^-$	0.0056$^+$	n.s.	0.0027$^+$	0.0479$^+$	n.s.	n.s.	n.s.	n.s.	n.s.	<0.0001$^+$		0.0001$^+$
BPP 0.2-5µm (µg C $L^{-1}h^{-1}$)	0.0012$^+$	0.0072$^-$	n.s.	0.0041$^-$	0.0102$^+$	n.s.	<0.0001$^+$	0.0114$^+$	n.s.	n.s.	0.0188$^+$	n.s.	n.s.	<0.0001$^+$	0.0001$^+$	

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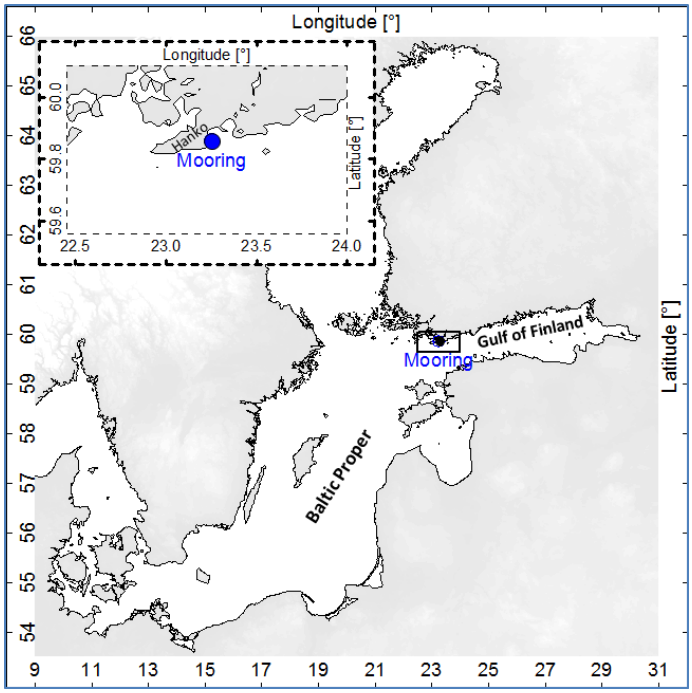


Figure 1: The Baltic Sea and the location near the peninsula Hanko in the western Gulf of Finland where the mesocosms were deployed

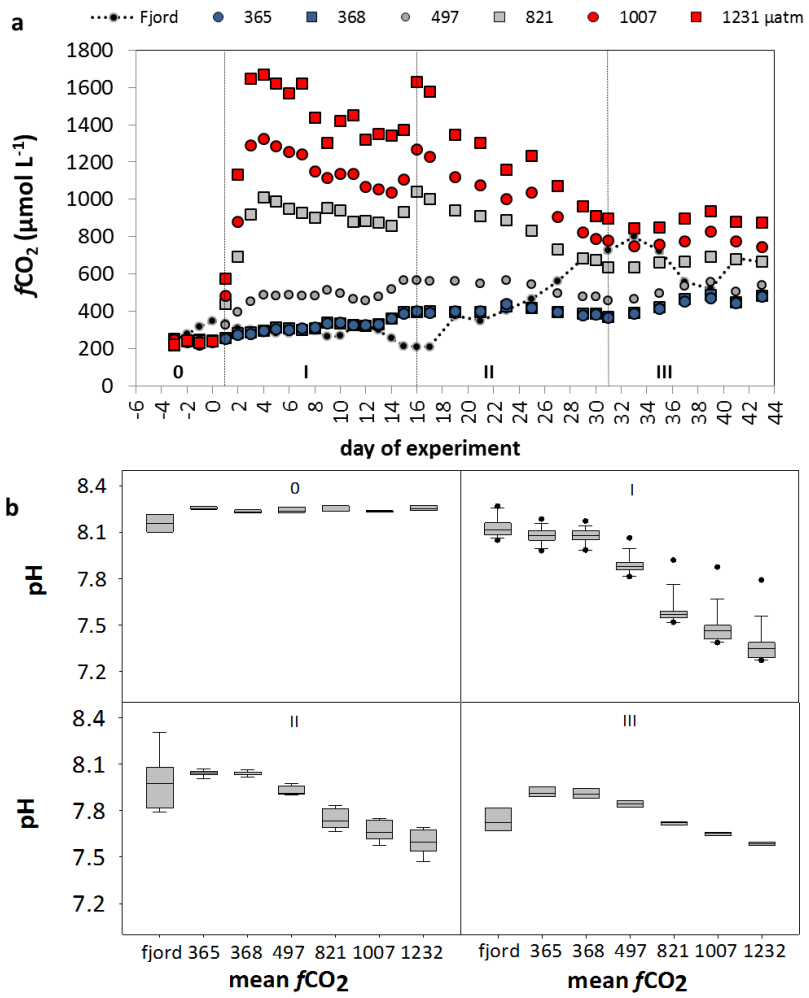


Figure 2a: fCO_2 values in the mesocosms and in the fjord throughout the experiment. Small black dots show the fCO_2 in the ambient fjord water. Treatment of the mesocosms with CO_2 saturated fjord water at the beginning of the experiment (days 0-4) created different fCO_2 levels in the mesocosms: blue symbols represents the untreated mesocosms, grey the intermediate, and red the high CO_2 treated mesocosms. The treatment was repeated at day 16.

Figure 2b: Corresponding pH ranges in the mesocosms during the four phases. Despite decreasing trend over time, a gradient between the mesocosms was kept over the whole period.

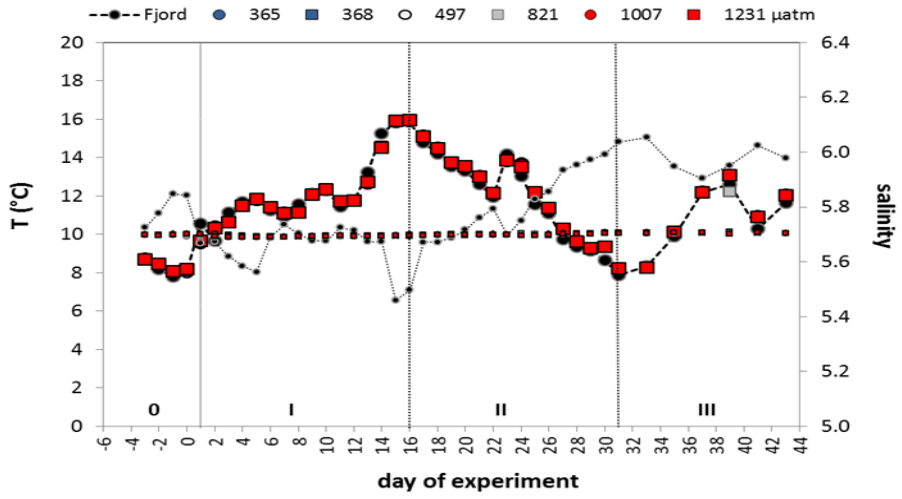


Figure 3: Temperature and salinity averaged over the 17 m surface layer of the mesocosms and the fjord. The data were obtained from daily CTD casts. Large symbols represent temperature and the small symbols salinity. Fjord water is shown as black dots with broken line while blue symbols denote untreated, grey intermediate and red high fCO_2 levels in the mesocosms. According to the temperature regime, the experimental period can be divided into four phases (phases 0, I, II and III).

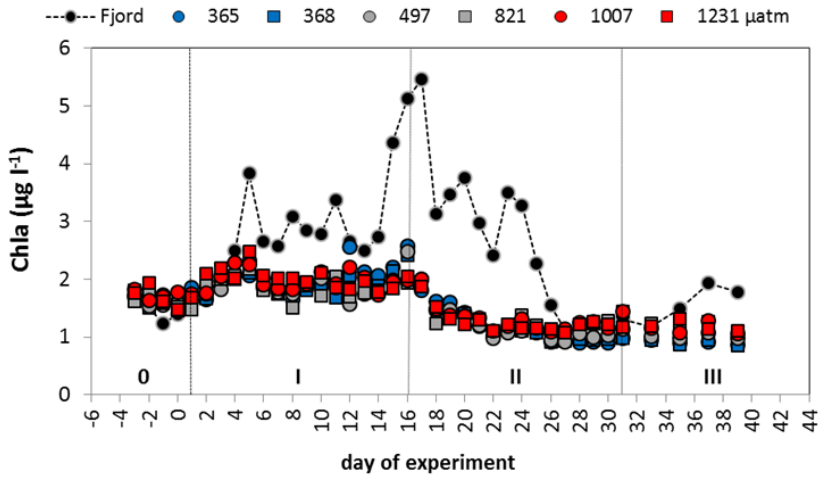
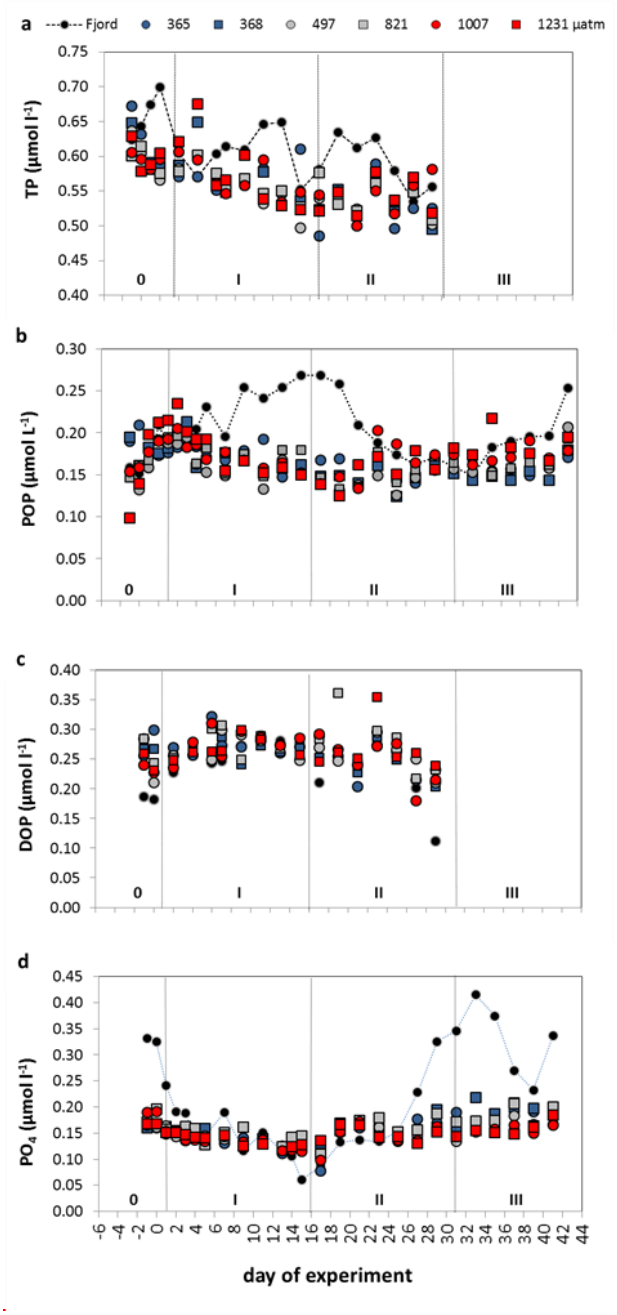


Figure 4: Chl*a* concentrations in fjord water and in the mesocosms with different fCO_2 conditions. The development over time can be divided into three phases as well. Blue represent untreated, grey intermediate, and red highly treated fCO_2 levels. Black dots are the Chl*a* concentrations in the fjord water.


a
Fjord
365
368
497
821
1007
1231 µatm
TP (µmol l-1)
b
POP (µmol L-1)
c
DOP (µmol l-1)
d
PO4 (µmol l-1)
0
I
II
III
day of experiment

1280

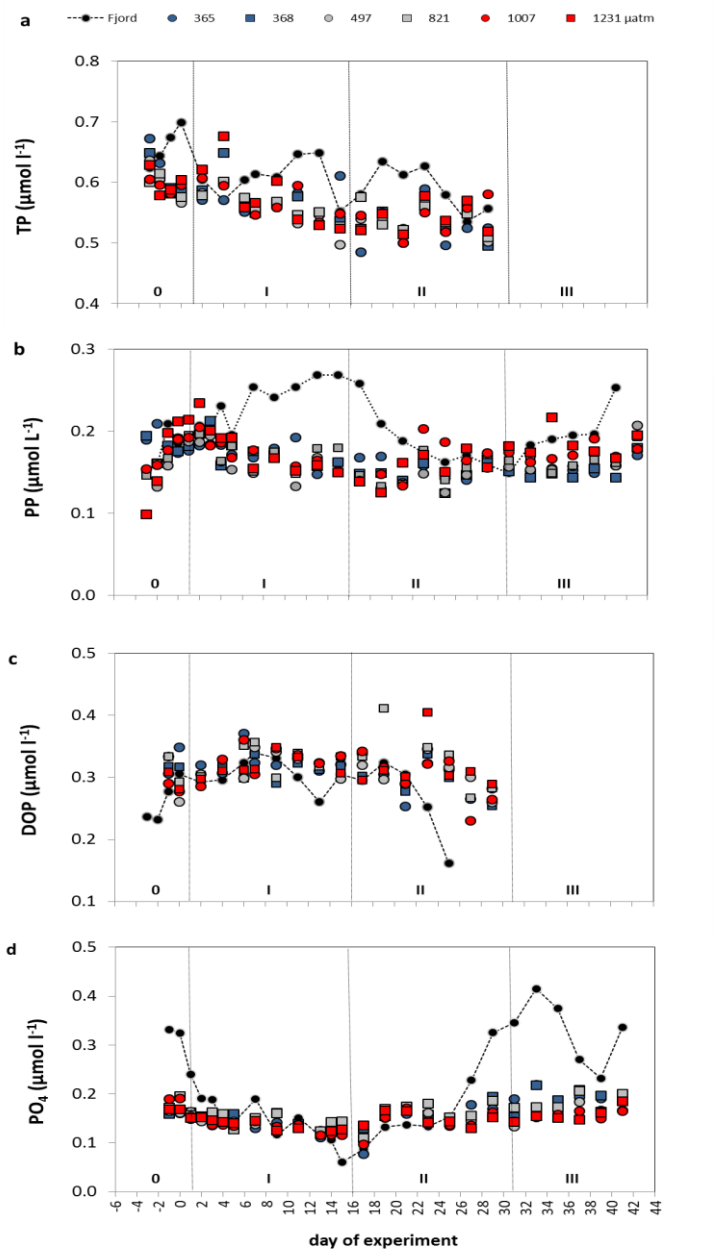


Figure 5a-d: Development of total phosphorus (TP) and the three measured P-fractions in fjord water (black dots with dotted line) and in the mesocosms over time. Blue represents untreated, grey intermediate and red high fCO_2 treatment levels.

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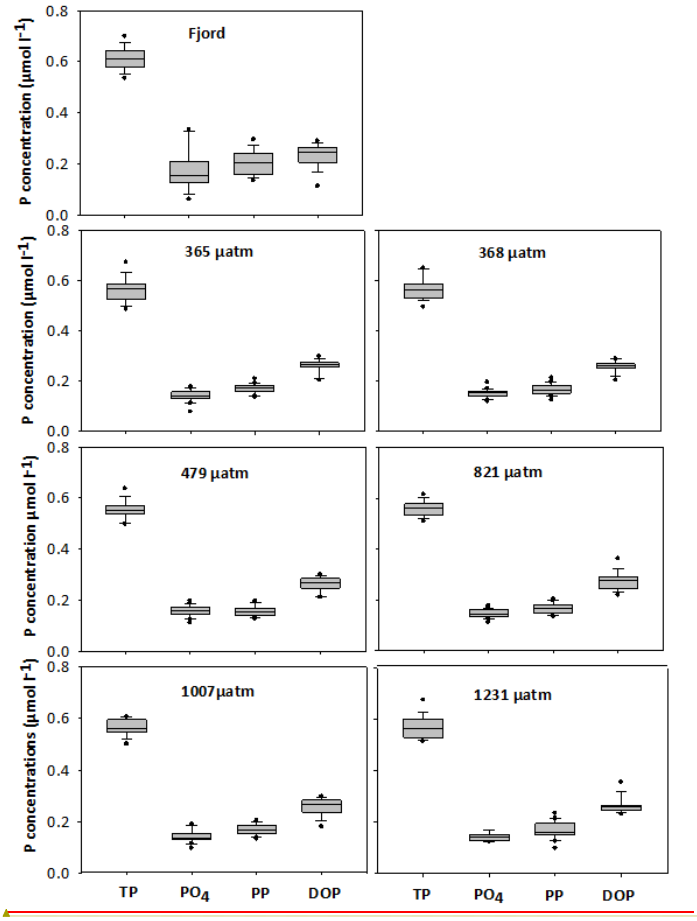

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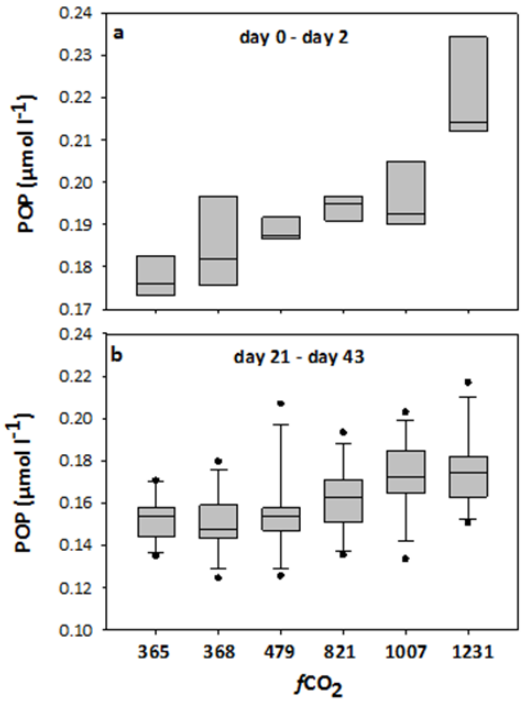
Figure 6: Contribution of the individual P-fractions to TP in fjord water and in the respective mesocosms. The data are averaged for the period when TP measurements were done (day -3 – day 29).



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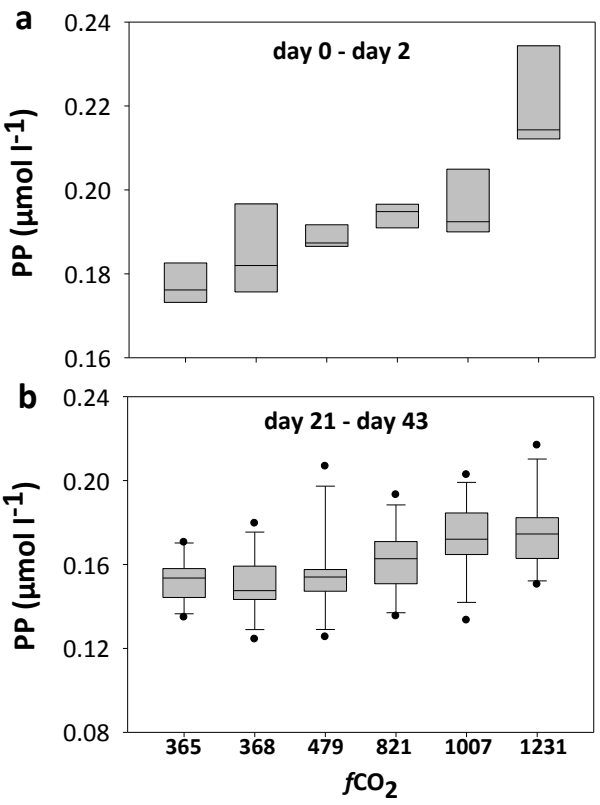


Figure 7: P~~O~~P concentration in the mesocosms during the initial phase from day 0 to day 2 (a) and from day 23 until the end (b) of experiment.

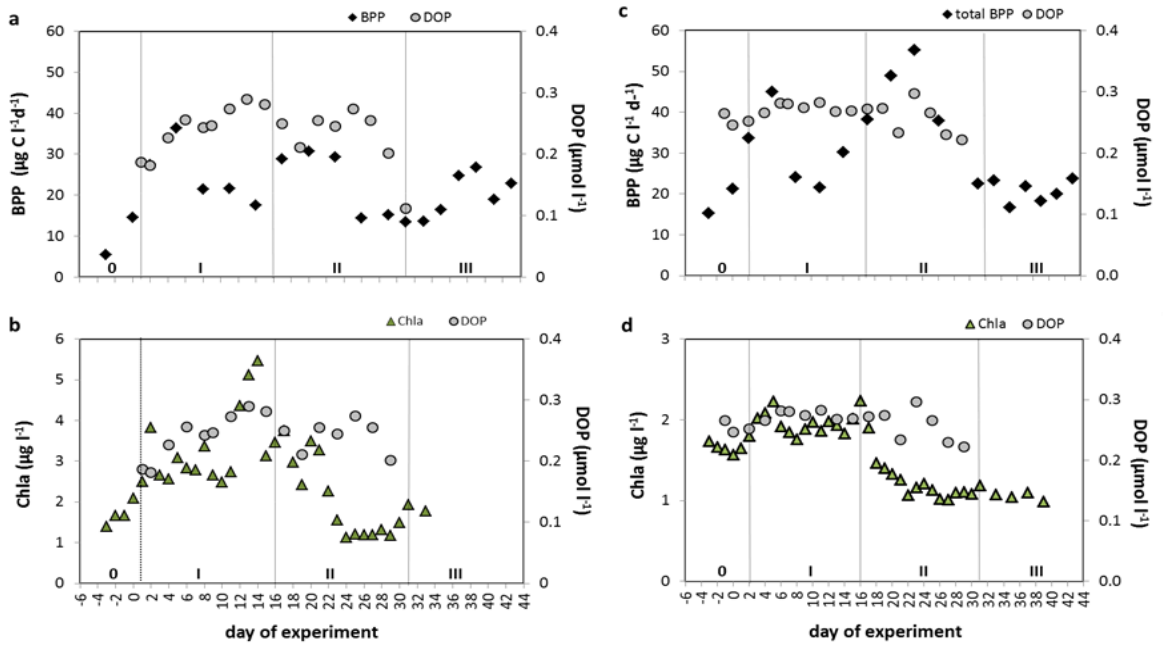
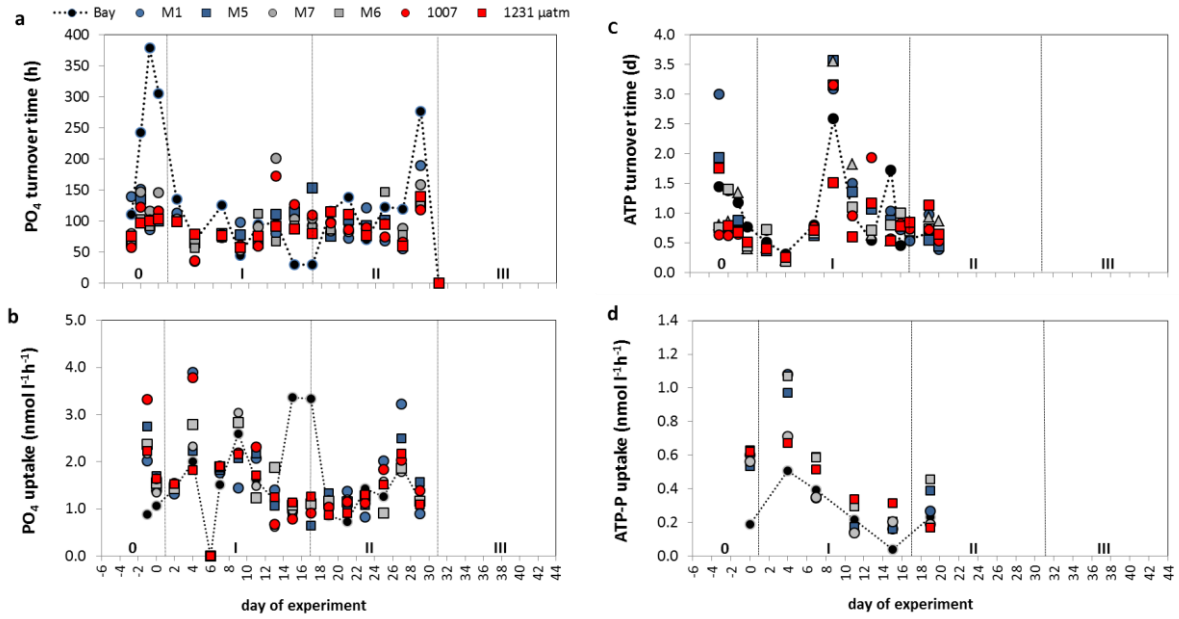


Figure 8: Development of DOP in relation to bacterial production (BPP) and phytoplankton biomass (Chl*a*) in the fjord (a, b) and in the mesocosms (c, d). For mesocosms, mean values averaged over all treatments are given.



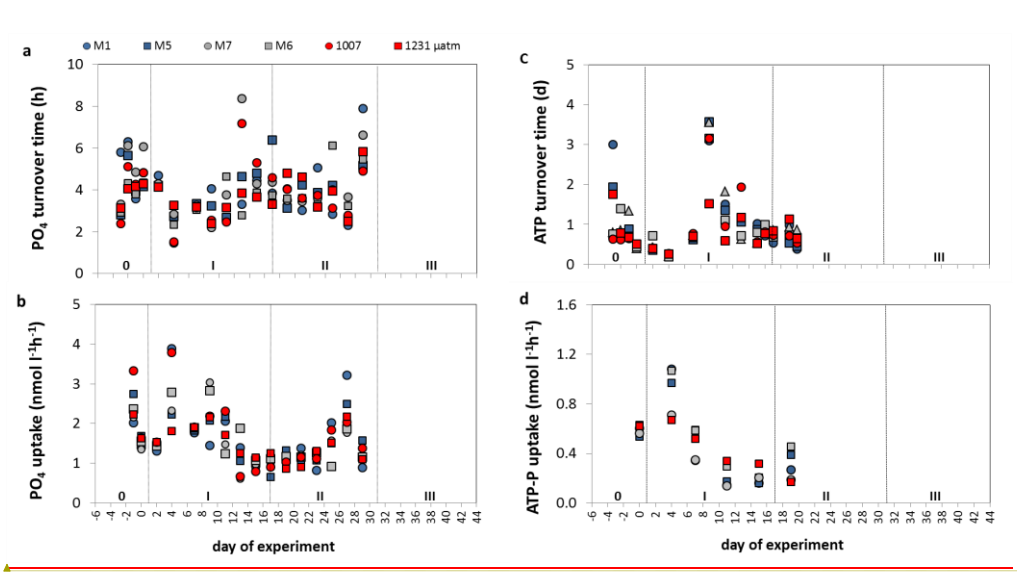


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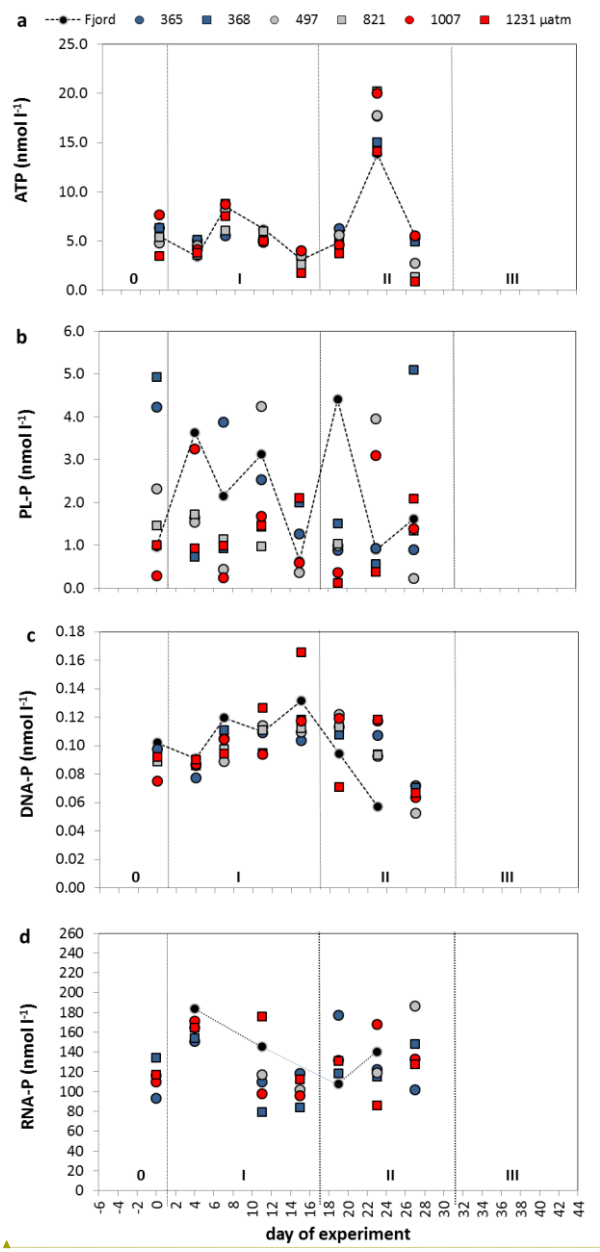
Figure 9: Turnover times of PO_4 (a) and ATP (c) ~~in fjord water and~~ in the mesocosms as well as the respective uptake rates (b, d).



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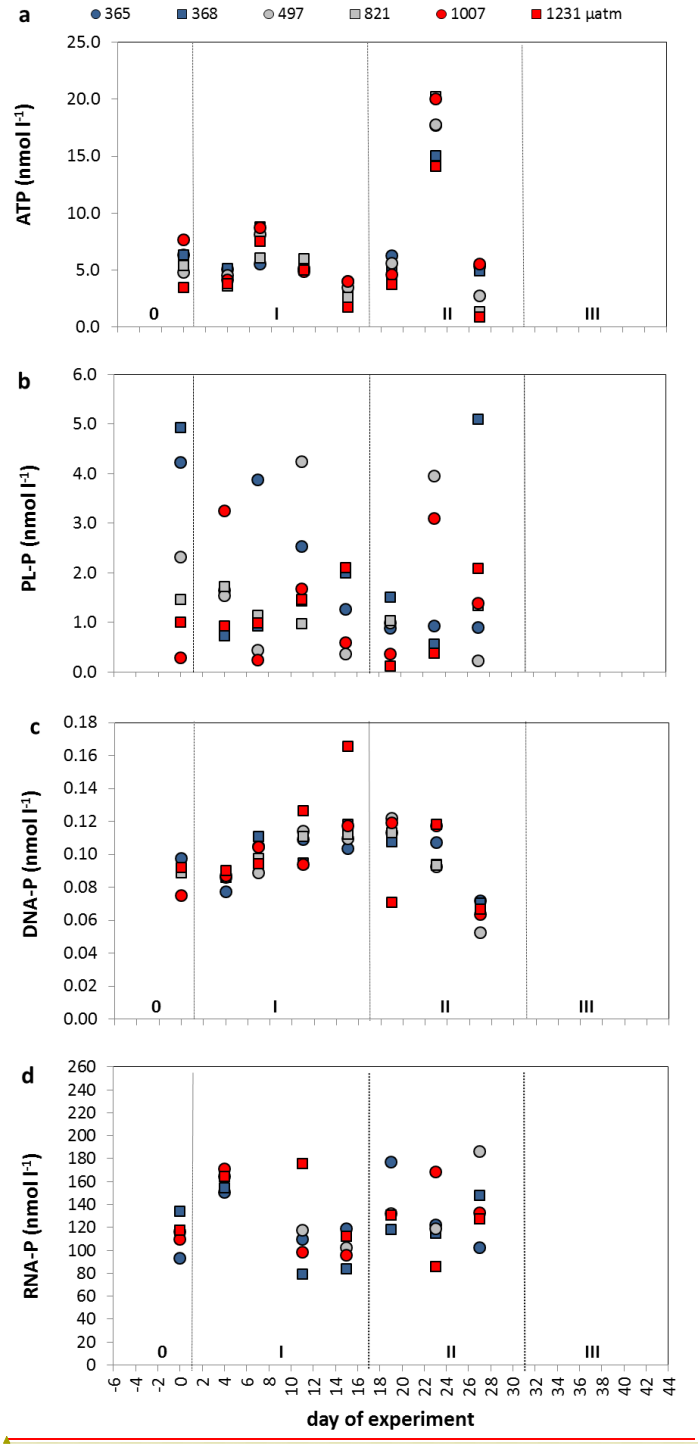

Figure 10: Development of DOP compounds in the mesocosms ~~and in the fjord~~ from day 0 to day 27.

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