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Interactive comment on "High temperature decreases the PIC / POC ratio and increases phosphorus requirements in *Coccolithus pelagicus* (Haptophyta)" *by* A. C. Gerecht et al.

Anonymous Referee #1

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The manuscript by Gerecht et al. is an important contribution to the debate on coccolithophore's response to nutrient limitation. The experimental setup is appropriate and the data are robust. There can be no doubt that this dataset should eventually be published in Biogeosciences. However, the presentation of the data leaves room for improvement and the interpretation is not doing justice to the data. The main issues are these: 1) The interpretation of the PIC/POC ratios is confusing, because it is neither consistent with the author's own data, nor data in the literature. 2) There are many disagreements between statements in the text and data given in table 2. Even within table 2 there are inconsistencies. 3) This is actually the first dataset showing an increase in malformations in response to P limitation. The authors are apparently not aware of

that remarkable fact. On the contrary, they cite Ehux literature allegedly showing that this is also the case in Ehux. The cited papers do, in fact, show no such thing. I'll explain these points in more detail in the specific comments: P 1022 L 12: is braarudii a species or a sub-species? P 1022 L 16: please give the temperatures rather than just the difference between them P 1022 L 18-19: this is not in accordance with the data. In pelagicus 15C, P-limitation does affect PIC/POC. Under low P, temperature does not affect PIC/POC P 1025 L 8: Ian Probert is an author. So I don't think that a recommendation by him needs to be mentioned. P 1025 L 19: How was the acclimation done in case of P-limitation? The latter is in batch mode a transient stage, so it is difficult to imagine how one can acclimatise a culture to a non-constant condition. P 1025 L 22: the light intensity probably means light limitation. Why was that chosen? P 1026 L 4: growth rate is calculated by exponential regression of the cell density vs time curve, not linear regression. P 1032 L 5: The PIC/POC response in Ehux is strain-specific, and probably method-specific. In B92/11 for instance, PIC/POC decreases in batch (Langer et al. 2013, JEMBE 443, 155-161) and remains constant in continuous culture (Borchard et al. 2011, JEMBE 410, 61-71). There are also unpublished results on Ehux showing a decrease as well as no change in PIC/POC. P 1033 L 12: statement not correct. In pelagicus 15C, P-limitation caused PIC/POC to rise by almost a factor of 2 P 1033 L 14: not correct. In fact PIC/POC is below unity in pelagicus 15C only. P 1033 L 16: not correct. POC quotas decreased in both high and low P. P 1033 L 17: not correct: In high P, elevated temperature caused POC production to decrease by 22% P 1033 L 18: PIC production declined by 58%. So that's ca. 60%, rather than ca. 50% I strongly advise the authors to have another look at the tables. It is highly confusing if the statements in the text are in disagreement with the data. As a reader one wonders whether the text or the table is correct. Also table 2 seems to have internal problems. The PIC/POC ratios are correct relative to the quotas in pelagicus 15C only. P 1034 L 5: temperature-stress treatment is supposed to mean pelagicus 15C, I guess. Please be precise and avoid fuzzy terms. P 1034 L 5-19: the discussion is not convincing, because if P limitation affects primarily POC production, the PIC/POC ratio

should always increase, which it clearly does not (not even in the dataset presented here, let alone in other datasets in the literature). P 1034 L 8: which offset? P 1034 L 18: what does “calcification” mean? The correct term is PIC quota. Also the statement about the Riegman paper is not correct. In the latter PIC quota increased by a factor of ca 4, and POC quota increased by a factor of ca 2. P 1034 L 22: The hypothesis in the cited paper was tested in Langer et al. 2013 (JEMBE 443, 155-161). Please cite also the latter paper here. P 1034 L 23: not correct. PIC/POC increases in pelagicus 15C in response to P-limitation. As mentioned above (comment on P 1032 L 5), the response in Ehux is by no means uniform. By the way, the C leptoporus strain tested in Langer et al. 2012 (JEMBE 413, 131-137) showed no change in PIC/POC. So the discussion P 1034 L 25 - P 1035 L 12 is completely beside the point. If one wanted to know what the response to P limitation in terms of PIC/POC of coccolithophores as a group is, the best approach would be to throw the dice, I’m afraid. As for pelagicus it is obvious, but not easily explained, that temperature changes the PIC/POC response to P limitation. It strikes me that the pelagicus high P 15C sample shows the exceptional PIC/POC (0.5, while all others are ca 1, if the quotas are correct in table 2), not the low P 15C. But I’m afraid I can’t really make much sense of that. The higher P quota and lower cell yield at higher temperature is fascinating, but also difficult to understand. Could it be that pelagicus suffers from heat damage at 15C? The constant growth rate is a caveat here, though. The higher percentage of malformations at 15C, however, points indeed to heat damage (see also Langer et al. 2010 J. Phycol. 46, 1252–1256). Why would higher than optimum temperature increase P requirement? Just a wild idea: in chickens higher levels of inorganic P in food increased the levels of heat shock protein mRNA in response to heat stress (Mahmoud et al. 2004 Comp Biochem Physiol C Toxicol Pharmacol. 137(1):11-8). Maybe heat stress reactions consume extra-P? P 1036 L 11: Culture artefacts can, at present, not be explained entirely satisfactorily, but there has been some progress since 2009. It was shown that high cell densities and a lack of mixing lead to higher percentages of malformations in Ehux (Langer et al. 2013 Helgoland Marine Research 67, 359-369). The latter authors also

argued that the effects should be more pronounced in species such as leptoporus or braarudii/pelagicus. P 1036 L 14-17: This comparison nicely illustrates another fact about culture artefacts, i.e. that they are not constant over time (compare the case of leptoporus reported in Langer et al. 2013 Helgoland Marine Research 67, 359-369). P 1036 L 17-19: The statement about Ehux is not correct. Neither of these studies show that Ehux increases the percentage of malformations in response to P limitation (see also discussion in Langer et al. 2012 JEMBE 413, 131-137). The latter paper also shows that leptoporus does not increase malformations in response to P limitation (or N limitation). N limitation apparently does not induce malformations in braarudii (Benner 2008, PhD thesis, University of Bremen). This does not necessarily represent a discrepancy to the findings reported here, because, first, it is another type of limitation, and, second, there are cell density issues in Benner 2008 (see discussion in Langer et al. 2012 JEMBE 413, 131-137). To sum up, the present study is the first one to report macro-nutrient limitation induced malformations. This is quite remarkable and it would be very interesting to see what other species are doing. Another interesting question is, of course, what is Coccolithus’ response to N-limitation? Such an experiment can be done using the setup employed by the authors. But I am by no means suggesting that it should be part of the present study. P 1036 L 22: by “outside” the authors probably mean “above the optimum”. I perfectly agree (see comments above). The temperature effect on morphology strongly points to heat damage (see also discussion in Langer et al. 2010 J. Phycol. 46, 1252–1256). That’s why heat damage should be considered in possible explanations of other temperature effects. P 1037 L 7: increased PIC/POC is only true for some Ehux strains (see above). P 1037 L 9: there might in principle be an upper constraint but this is definitely not the point here (see above for details). P 1037 L 12: not true for pelagicus at low P, if one believes the table, precisely the quota data in the table and not the calculated PIC/POC, because the latter does not tally with the quota data. P 1037 L 20: in Ehux there are even strain-specific differences (see above). Table 1 pelagicus T0 low P 15C: the CO2 numbers are swapped I guess. Table 2: Check numbers please. PIC/POC ratios more often than not don’t follow from PIC

and POC quotas. Please put all data on the cells in the table, including loose and attached coccoliths and morphology. Alternatively add more tables. Especially the data on morphology need to be in a table, because the figure does not contain standard deviation. I would very much like to see a discussion on the ratio of loose to attached coccoliths. Is that ratio higher at 15C in pelagicus? If so that might be another indicator of heat damage. Figure 1: how is n=6 possible with triplicate incubations? On the whole, this is a very interesting dataset, which deserves a decent discussion. I strongly recommend publication, but only after the manuscript underwent appropriate revision.

Interactive comment on Biogeosciences Discuss., 11, 1021, 2014.