Interactive comment on "Physiological responses of coastal and oceanic diatoms to diurnal fluctuations in seawater carbonate chemistry under two CO$_2$ concentrations" *by* Futian Li et al.

Anonymous Referee #1

General comments

This manuscript presents data on the effects of Ocean Acidification on coastal and oceanic diatom species under constant and fluctuating pH regimes. This is a very relevant and timely issue, and the results are very interesting. I am particularly excited about the differences between the coastal and oceanic species investigated here. Before publishing this manuscript, however, the statistics and some parts in the description/ discussion of data need to be changed. Unfortunately, I also see two potentially significant problems with this dataset, which hopefully can be resolved by the authors: Firstly, a second parameter of carbonate system is missing to fully constrain carbonate chemistry. Secondly, even though not clearly mentioned, from the description of the

methods and data it sounds like the distinct measurements were conducted from the same incubation bottles (as the authors speak about replicate "samples" but not "incubations" or "replicates"). If this would be true, no statistical analysis or any kind of interpretation would be meaningful to conduct based on this data. I hope this is rather a misunderstanding from my side, because otherwise the authors would have to repeat the experiment with proper replication.

Specific comments

P2 L20: I suggest changing the beginning of the sentence from "Diel or seasonal" to "Diel and seasonal"

P2 L 22: I suggest changing the sentence from "natural carbonate buffer system" to "natural dynamics in the carbonate buffer system"

P3 L54-55: Not clear if the statement on "fluctuations in coastal seawater" refers to current or future conditions.

P4 L69-72: The first and second part should be spilt in two separate sentences. Furthermore, something seems to be missing here.

P6: In the description of the manipulation of and measurements of carbonate chemistry, only pH measurements are mentioned. To constrain carbonate chemistry, however, a second parameter of the carbonate system is critically needed (cf. best practice guide; Riebesell et al 2010). While I understand that it is probably not feasible to measure other parameters as frequently as needed for the fluctuating pH regime, the authors still need to show that they properly controlled carbonate chemistry, e.g. by presenting AT data from the beginning and the end of the experiment.

P6 L 120-121: The time points of measurements are defined differentially throughout the manuscript. It would be good to have these more consistent. Here for example, also the number of hours after onset of light should be mentioned.

P7 L 136: Rather than filter size, the pore size seems to be the more relevant informa-

tion.

P8 L147: How similar was the light? Please be more specific here.

P8 L150-152: Light exposure for 15s is very short, I do not think that NPQ can be robustly estimated under these assay conditions. The authors need to provide evidence for their statement that they really "provide estimates on the kinetics of NPQ development".

P8 L 164-165: Standard errors or deviations of the pH values are missing.

P9 L169: I do not agree with the way the statistics have been done. From my perspective, you have two independent variables (i.e. LC vs. HC and steady vs. fluctuating) and not one, so the data should have been analyzed using a two-way instead of a one-way ANOVA.

P8 L171-172: The authors state that all data is reported as "mean value of triplicate samples". Does this mean that there was no true replication in the experiments, and samples were taken from the same incubation bottles? This needs to be clarified. If the latter is the case, statistical analysis is not possible, as this would mean n=1.

P9 L177: I would still prefer to see the error bars.

P11 L 210-216: I find the structure of the results section partially confusing (especially in this section). I would try to structure it more clearly, e.g. by always describing the responses of T. weissflogii before those of T. oceanica.

P12 L 239: Can cells "have a decrease" in something? Consider revising.

P12 L241-249: I find this section also quite confusing, also because the time points are sometimes described with hours and sometimes descriptive (e.g. middle of photoperiod).

P13 L 251: I think this should read "while the fluctuating regime had no detectable effect".

P13 L 263-258: Given the limited usefulness of these super short RLCs, do you really need this data for your argumentation?

P15 L 296-298: The authors state that "diatoms may have reduced silicon requirements per carbon fixed under an OA scenario than under ambient pCO_2 condition, and so has implications for changes in local and global silicon budgets". Despite improvable grammar in this sentence, I find the use of the term "silicon requirements" in this context rather misleading because BSi per cell is only affected by OA in one out of four situations and the change in BSi:POC ratio is rather driven by changes in POC quota (Figure 3).

P15 L308: Consider changing "C3-C4 intermediate (Roberts et al. 2007) photosynthesis" to "C3-C4 intermediate photosynthesis (Roberts et al. 2007)".

P16 L314: Details on CCM characteristics were not "shown here", but rather hypothesized.

P16 L319-322: I do not like the use of the word "sacrifice" in this context. This sounds like an active decision by the algae, rather than a process where evolution is acting upon an organism.

P17 L337: Consider changing from "calcification of corals benefit" to "calcification of corals can benefit".

P17 L 346-349: I don't think the authors can claim that "all of the members" of a natural diatom community" have been investigated in this species (e.g. cf. Schaum et al. 2012 for intraspecific plasticity).

P18 L360-364: UV comes in as a bit of a surprise here and I am not convinced it really feeds into the argumentation/story of this manuscript.

P18 L367-268: I do not find data that would show that "elevated CO2 mitigated the limited availability of pCO_2 that occurred at the end of photoperiod under the LCf condition" in this manuscript.

P18 L371-372: It may be worth mentioning that this is even more so in coastal compared to oceanic environments.

P19 L377: I don't think the responses really classify as "poor physiological performance".

P19 L385: I don't understand the last part of this sentence, what is meant by "factors that will help to decide the spatial distribution patterns of species"?

P20 L397: Something went wrong with this citation.

P30 L 599-600: The used pH scale and error estimates are missing. Furthermore, it should be mentioned if this is 1) an average over all days, or an example and 2) averaged over the biological replicates (I assume were used) or just one bottle.

P30 L611 and L618: For consistency, I would also mention the number auf ours after start of the photoperiod in these captions.

P32 Table 1: The differences in cell size between both species are an interesting aspect that should be discussed in terms of their implications for surface:volume ratios, carbon acquisition and pH homeostasis. Similarly, also the R:P ratio is an interesting parameter (e.g. the significantly higher ratio in T. oceanica under LCf), that is currently not discussed in the manuscript. Furthermore, units of ratios are missing.

P33 Table 2: The irradiance level used for these measurements should be mentioned in the caption. For clarity, I would furthermore call the time point really "time point rather than "time" and add a "h" after the number of hours.