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Interactive comment on “Ocean acidification mediates photosynthetic response to UV radiation and temperature increase in the diatom *Phaeodactylum tricornutum*” by Y. Li et al.

Anonymous Referee #3

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The study from Li et al. “Ocean acidification mediates photosynthetic response to UV radiation and temperature increase in the diatom *Phaeodactylum tricornutum*” deals with the interactive effects of an increase in CO₂ and temperature, and the exposure to UV radiation on the diatom *Phaeodactylum tricornutum*. In order to assess the effects on this species the authors acclimate cultures to different CO₂ concentrations (1000 ppmv vs. 390 ppmv) at 20 °C and PAR (70 μmol photons m⁻² s⁻¹) during >20 generations. After this period, they expose subsamples to higher irradiances including PAR, UVA and UVB (PAR, 290 μmol photons m⁻² s⁻¹) at 15, 20 and 25 °C for approximately 1–2 h depending on the technique. The techniques included measurements for the assessment of the photosynthetic quantum yield with a Xe-PAM fluorometer and carbon

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fixation with ^{14}C at three different spectral treatments (PAR (P), PAR+UVA (PA) and PAR+UVA+UVB (PAB)). The results from the study show that growth at elevated CO_2 increased non-photochemical quenching (NPQ) and partially counteracted the harm to PSII caused by UVR. Such an effect was less pronounced under increased temperature levels. In addition there were not a big significance in carbon fixation rates between cells grown under high CO_2 or had slightly higher rates under low CO_2 conditions.

The topic of the study is interesting. However in my opinion there are substantial points related to the effect of CO_2 that need to be addressed with more detail. There is also a lack of consistency between the results and conclusions from this study and those published previously from authors belonging to the same group that are vaguely discussed in the manuscript (Wu et al 2010, Gao et al. 2007). For example, in the paper from Wu, Gao and Riebesell (2010) CO_2 -induced seawater acidification affects physiological performance of the marine diatom *Phaeodactylum tricornutum*. Biogeosciences 7, the authors conclude (working with the same strain of *Phaeodactylum* and exactly similar growth conditions) that in the high CO_2 grown cells, the electron transport rate from PSII was photoinhibited to a greater extent at high levels of PAR, while NPQ was reduced compared to low CO_2 grown cells. In addition, high CO_2 grown cells were downregulated, and growth and photosynthetic carbon fixation rates were enhanced by 5% and 12% respectively compared to cells grown at ambient CO_2 . As stated above, opposite conclusions are shown in the present manuscript without showing a clear explanation for the differences: The main argument shown by the authors is that the decrease in photoinhibition might be due to UV stimulation of the external carbonic anhydrase (CAe) by UVR. However, it is expected that an acclimation to high CO_2 levels similar to that performed for the experimental conditions shown in this paper (> than 20 generations of acclimation to high CO_2 levels) would downregulate CA, as also demonstrated in Wu et al. 2010. Under downregulated conditions no activation of CA should be observed, independently of the presence/absence of UVR. Is it possible that despite the long acclimation to high CO_2 levels *Phaeodactylum* cells would not be completely downregulated? It would be interesting to see some results demonstrating

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the activity of the CAs or other CCMs. Information about growth rates under high/low CO₂ conditions would be also helpful. They can give some insights to explain if cells were completely acclimated, and therefore CCMs downregulated.

Are the subsamples independent replicates? The authors used triplicate samples to measure carbon fixation and photochemical efficiency under the different spectral treatments and temperatures. However it is not clear to me if the subsamples come from only one or several cultures. Which is the volume of the culture/s?

The authors say that PAM measurements were carried out under 300 $\mu\text{mol photons m}^{-2} \text{ s}^{-1}$ PAR for all the treatments, including those with UVA and UVA+UVB. I thought that the Xe-PAM had the capability of exposing samples to PAR and UVR. I understand that samples were measured quite fast after they were collected, however if UVR exposed samples are exposed to PAR we should assume some recovery of photoinhibition caused by UVR. Please clarify why UVA and UVB were not included during the Xe-PAM measurements for the samples coming from the PA and PAB treatments, respectively. Which is the reason for growing cultures at low light (70 $\mu\text{mol photons m}^{-2} \text{ s}^{-1}$) and expose them for assessment of the photosynthetic characteristics at high light (PAR, 290 $\mu\text{mol photons m}^{-2} \text{ s}^{-1}$)?

It would be useful to have absolute rates of damage and repair instead the ratios

The introduction and discussion sections could be highly improved.

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