

The authors have studied how the effects of ocean acidification, growth temperature and UV exposure interact in terms of growth, photosynthesis, and calcification in the coccolithophor, *Emiliana huxleyi*. The study complements several related studies of how these processes in *E. huxleyi* respond to various combinations of pCO₂, growth irradiance and UV Treatments. The novel aspect of this study is the evaluation of these effects at temperatures chosen to bracket this strain's apparent optimum growth temperature at 20°C. The results of this study have the potential to advance our understanding of how this key species responds to the multiple changes occurring in the aquatic environment. To realize this potential, these results need to be better integrated with previous studies. In particular, the present discussion does not do much to show how these results fit into the bigger picture of how *E. huxleyi* responds to ocean acidification and UV. Several speculations are advanced which relate to specifics of this study which I did not consider justified as explained below. Thus, the discussion will require substantial revision to address these issues.

Major Comments:

Line 80 The sentence refers to the possibility that ocean warming will lead to enhanced stratification and mixed layer shoaling. It cites a reference on UV-temperature interaction in coral reefs (Courtial et al., 2017), that reference presents no direct information on the relationship between ocean warming and mixed layer depth. Courtial et al. (2017) do cite two review papers (now outdated) as sources for a presumed increase in UV exposure due to mixed layer shoaling and claim that as a result corals would be exposed to higher visible and UV radiation. It is difficult to understand this statement given that corals are benthic organisms that do not experience vertical motion. In any case, more recent studies have refuted the assumed relationship between ocean warming and mixed layer shoaling, see Somavilla et al. (2017).

Line 189 “ratios ... were about 30% higher”

What is the source for the UV:PAR relationship? The percentage looks about right for comparison of the solar-simulator output to incident, midday, near-solstice irradiance in the subtropics but the UV:PAR ratios for incident radiation will be lower than that averaged over the day and over seasons (as I assume was done to estimate the PAR mean light level). Also as a fraction of incident, mean UV in the mixed layer will be less than mean PAR in the mixed layer since UV is attenuated more strongly than PAR.

Line 231 “two sample paired t-tests”

If these are comparisons between specific treatments included in the ANOVA analysis, post-hoc multiple comparisons should be applied to test for the significant effects.

Lines 292-361 (Sections 3.5 and 3.6)

These two sections read as if they are two different data sets whereas in reality it is just the same data viewed in two different ways. There is substantial redundancy between the two

sections and it should be condensed into one section. For example, lines 303 – 305 state that photosynthesis was reduced by 33.4% in HC cells and 19.9% in LC at 20°C, essentially the same result is presented in lines 334-335 in the next section. If differences between treatments (e.g. PA and PAB) are not significant, then the inhibition percentage should not be considered significantly different from zero.

It makes sense to discuss the Cal/Pho ratio either as being either increased or decreased in a treatment vs the ratio in the control. On the other hand, the terms “inhibition” and “stimulation” are appropriate for the rates themselves but not the ratio of rates. From that standpoint, it seems that everything that needs to be said about treatment effects of Cal/Pho are covered in lines 322-330, and lines 355-361 can be dropped.

Line 395ff Explanations of higher resistance to UV in 24°C HC

Here the authors argue that increased cellular nitrogen in this treatment was responsible for the lower sensitivity to UVA via enhanced defense/repair mechanisms. I question the rationale here, because the enhanced PON is accompanied by larger cell volume and more POC, so there is no indication that the C:N ratio has changed. An increase in the content of nitrogen-intensive defense mechanisms, relative to other components of the photosynthetic apparatus, sufficient to change sensitivity to UV would be expected to lower the C:N ratio, compare (for example) with results of Lesser et al. (1994)

Estimating from the figures for example, I get these POC: PON (mass:mass) ratios
15° HC POC:PON = 11.9/2.1=5.67 (6.6 molar)
24°C HC 13/2.25 =5.78 (6.7 molar)

These are not significantly different. Apart from this, I question whether mycosporine amino acid (MAA) accumulation could be involved in the enhanced resistance, particularly since no measurements of absorbance or pigments were made. Xing et al found that UV absorbing compounds (UVAC) were accumulated when cells were grown in the presence of UV, but no UVAC were accumulated during PAR only exposure as used in this study. Moreover, Xing et al never definitively identified the UVAC as MAAs. Other studies of *E. huxleyi* have found only low or trace quantities of MAAs.

Lines 422 – Better explanation needed for the discrepancy between different studies – how do differences in growth irradiance and treatment exposure explain why UVA sometimes stimulates vs inhibits calcification rate?

Lines 431ff Speculation on reason for UVA-stimulation

Here the authors invoke UVA-dependent bicarbonate uptake as a mechanism to account for the enhanced calcification under the PA treatment. The arguments advanced are not justified at the irradiance levels used in the experiment. At the PAR levels used in the exposure, both photosynthesis and calcification rate are irradiance saturated (cf. Jin et al. 2017). So the

additional photon energy coming from UVA is unlikely to increase calcification. Likewise, if irradiance effects are saturated, differences in coccolith layer thickness are unlikely to have an effect mediated by light transmission.

Line 468 This implies the presence of UVR could compensate...

Don't understand the reasoning here, episodic exposure to UV as used in the experiment is already occurring in natural assemblages and will continue in the future, so what is the potential for UVR effects to compensate for future increases in temperature and acidification?

Minor Comments

Line 180 elemental samples

Better – "samples for elemental analysis"

Line 181: ...90 mL quartz tubes (volume 100 mL)

Are the numbers are reversed? Obviously, 100 mL can't fit in a 90 mL tube

Line 183 – How many tubes were incubated in each treatment?

Line 186 "PAM2100"

The model number is incorrect, I expect what was meant was PMA2100, which is a logger that can be used with several types of sensors, so specify which sensors were used for each channel.

Line 212ff Methods formulas state that Inh was calculated as a percentage, but figure (6) contains only fractions

Line 496 Misspelling – Author is Banaszak

Line 651 "Response of growth and photosynthesis of *Emiliania huxleyi* to visible and UV irradiances under different light regimes."

Citation is incorrect year 2015, vol 91:343-9

There are a number of other typos in the reference list

References:

Lesser, M. P., J. J. Cullen, and P. J. Neale. 1994. Carbon uptake in a marine diatom during acute exposure to Ultraviolet B radiation: relative importance of damage and repair. *Journal of Phycology* **30**: 183-192.

Somavilla, R., C. González-Pola, and J. Fernández-Díaz (2017), The warmer the ocean surface, the shallower the mixed layer. How much of this is true?, *J. Geophys. Res. Oceans*, 122, doi:[10.1002/2017JC013125](https://doi.org/10.1002/2017JC013125).