

***Interactive comment on “The Arctic picoeukaryote  
Micromonas pusilla benefits from Ocean  
Acidification under constant and dynamic light”  
by Emily White et al.***

**Emily White et al.**

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Abstract: Good Introduction: Good overview of a scattered field. Figures + legends: Good. We are happy the reviewer was content with these parts our manuscript.

Materials & Methods: "well as with macronutrients in Redfield proportions (containing 100  $\mu\text{mol L}^{-1}$  of nitrate and silicate, and 6.2  $\mu\text{mol L}^{-1}$  phosphate)." The goal is to understand what *Micromonas* might do in a changing Arctic ocean. So how does 100  $\mu\text{M}$   $\text{NO}_3^-$  and 6.2  $\mu\text{M}$   $\text{PO}_4^{3-}$  compare to natural levels? We live in an imperfect world, but responses to an increase in  $\text{pCO}_2$  (or to fluctuating light) might be very different under a situation of luxury accumulation of excess protein, vs. nutrient limits on protein

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accumulation etc. Just dimly remembering that 80  $\mu\text{M}$   $\text{NO}_3^-$  is about the equivalent of the Pearl River Delta, so.... pretty high? I know we face compromises in culture work at getting enough biovolume in a reasonable culture volume, but these points might influence/alter/limit the findings? In contrast the fluctuating light regime is nicely justified in terms of realistic approximations of the environment. We fully agree with the reviewer that low nutrient concentrations are a major feature of the Arctic Ocean. Therefore, it would be highly desirable to conduct experiments under realistically low nutrient levels. It is, however, very tricky to achieve constant levels of nutrient limitation, which would be necessary to fulfil the 'Ceteris paribus' principle and deduce causal relationships. To our knowledge, this is only possible in chemostat or turbidostat set-ups that are very complex to establish and run under low biomass (the latter being necessary for stable carbonate chemistry), and therefore were beyond the scope of this study. Furthermore, the strain we are working with was isolated from pre-spring bloom conditions in a fjord of Svalbard under non-nutrient limiting conditions, so that we are confident that our setup is a reasonable approximation of this habitat/environmental setting. Nonetheless, we now stress the fact that nutrient limitation is a very important driver in the Arctic that we have not investigated in the implications section of our manuscript by writing 'Nutrient deficiency was not addressed in this study as the experimental design was aiming to mimic non-nutrient limiting conditions before the spring bloom. Nonetheless, the often limiting nutrient supply in the Arctic sets the trophic status of each region and limits annual productivity (Tremblay et al., 2015), thus is an important factor to consider in future studies.' (L412-415).

Eqn. 3 would benefit from an additional set of parentheses around the denominator terms to clarify the order of operations. Agreed and done.

Eqn. 4 should use  $\sigma_{\text{PSII}}$ , otherwise you are not accounting for any nonphoto-chemical down-regulation of  $\sigma_{\text{PSII}}$  under illumination. If I entered the equation incorrectly in Xu et al. 2017, I apologize, my papers have been filled with equations typos lately. We thank the reviewer for pointing out this typo. We have used  $\sigma_{\text{PSII}}$

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for our calculations, and the equation is corrected in the revised manuscript (L185).

Line 195: Do these dyes enter cells, or stay outside? or both? I am recently learning that superoxide radical has a very short diffusion length, whereas H<sub>2</sub>O<sub>2</sub> can move a fair ways. Both of these dyes enter the cells but also stay in the surrounding seawater (Prado et al. 2012). The HE fluorophore binds DNA after being oxidized by superoxides, while DHR123 localizes in the mitochondria. By using the other flowcytometric characteristics of the cells (forward and side scatter as well as Chla fluorescence), the cell specific levels can be determined and diffusion distances should not play a role. These details are now provided in the text (L199-201).

Results: Line 245 The indicator dyes show the standing pool of reactive oxygen, which is the outcome of production rate - detoxification rate. Picky point, but it is possible the effects result from changes in detoxification, rather than production. Also, standing pool of a ROS species is not necessarily the same as oxidative stress... We fully agree with the reviewer regarding this inaccuracy of our wording, and have replaced 'oxidative stress' by 'ROS' (L255). In the discussion of dynamics light effects, we also slightly changed our wording to clarify such processes, i.e. replacing 'ROS production' by 'ROS accumulation' (L294) while in the discussion of OA effects we already considered these (L345).

Lines 285 etc. increased tau under fluctuating light, compared to decreased flow to POC & growth strongly suggests an induction of dissipative electron transport capacity under fluctuating light, leading to 'dumping' of electrons under the high light periods. Consider that you actually have all the data to estimate the Oxborough proxy for PSII I-1 (based upon F<sub>0</sub>/sigmaPSII). It is far from perfect, but, if you estimated it, and multiplied by your e- PSII-1 s-1, you could get e- I-1 s-1 Then you can compare electron generation rate with growth rate or with POC accumulation and get an electron quotient for growth. I bet it increases under fluctuating light. This is perhaps a more defined re-statement of your lines 292 etc. While there is definitely merit in the calculations of electron transport rates as proposed by Oxborough et al. (2012), we are not confident

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that they outweigh the uncertainty introduced by the estimation of PSII concentrations (see e.g. Schuback et al 2016) in this specific dataset. Following the principle of ‘as simple as possible, as complex as necessary’, we would therefore prefer to keep the simpler e- PSII-1 s-1-based ETR estimate. If the reviewer insists on this point, we would however, be willing to change these calculations.

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