

1 Trimborn et al. evaluate in a full-factorial experimental design how the
2 interaction between ocean acidification and high light alter the photophysiology,
3 stoichiometry, production and growth of a model Antarctic Cryptophyte at short-term
4 scales. The topic assessed fits with the *Biogeoscience*'s scope, and is novel as currently
5 information related with the interactive effect of both factors on Antarctic
6 phytoplankton is still scarce. The experiment is well-designed and performed, and the
7 results are appropriate to being published in the Journal.

8 Below, authors will find a point by point revision with the main issues found
9 over the manuscript, and suggestions which I hope that they find useful.

10 Respectfully submitted,

11 Marco J. Cabrerizo

12 **Title:** I suggest including in the title two variables also quantified by authors:
13 production and photophysiology of the species. On potential title could be: "*Ocean*
14 *acidification and high irradiance stimulate the photophysiology, growth and production*
15 *in the Antarctic cryptophyte Geminigera cryophila*".

16 **Introduction:** What are the individual effects of HL and OA on phytoplankton?.
17 I suggest that authors add some general information about the individual effects of light
18 and OA on primary producers, and then they focus such impacts on Antarctic
19 phytoplankton and cryptophytes. Through a general view, a potential reader can identify
20 the gaps of knowledge related with the quantification of such impacts on phytoplankton,
21 and the scarcity of experimental studies testing their interaction on this key group.

22 **Lines 27-29:** If it was found a contrasted response pattern in southern WAP, it
23 means that there were both a positive as a negative effect. In such case, it partially
24 agrees with those results reported in the northern part. Please rewrite.

25 **Lines 29-30:** Break this sentence to separate both ideas.

26 **Line 45:** Reference?

27 **Line 69:** I think that it would be nice that authors include a hypothesis work
28 about what do they expect based on the previous information known?.

29 **Material and Methods:**

30 **Culture conditions:** This subsection is confusing at the present state. I propose
31 you modifying it as follow: "*Before to being used in experimentation (two weeks),*
32 *triplicate semi-continuous cultures of the Antarctic cryptophyte G. cryophila (CCMP*
33 *2564) were grown (and maintained in mid-exponential growth) at 2°C in sterile -filtered*
34 *(0.2 µm) Antarctic seawater (salinity 30.03) enriched with phosphate (final*
35 *concentration 100 µmol L⁻¹) and nitrate (final concentration 6.25 µmol L⁻¹) (N:P ratio*
36 *of 16:1, Redfield 1963), as well as, trace metals and vitamins according to F/2 medium*

(Ref). Cells were exposed under a 16h L: 8h D cycle and three constant light intensities (20, 200 and 500 $\mu\text{mol m}^{-2} \text{s}^{-1}$) (LL, ML and HL, respectively)".

Once you have already described the pre-acclimation phase, you can describe your experimental setup, and finally, all the details related with the CO₂ enrichment. For example, "*To assess the interactive effect of light and OA on the photophysiology, metabolism and growth of G. cryophila, a 3x2 full factorial matrix (in triplicate) was implemented with: (a) three light levels (LL, ML and HL) and two pCO₂ treatments (ambient and OA)*"...

Line 15: Cells in exponential or mid-exponential phase. Please clarify.

Statistics: Were ANOVA assumptions checked?. Was the interaction light and OA significant in all response variables? Please clarify. Over the results section, I cannot see if Light×OA was or not significant.

Results: I propose authors to change the ordering of the results. First, they should show PSII variables, then pigments, metabolism and stoichiometry and finally growth. The rationale behind my proposal is related with the fact that changes at PSII level occurs in temporal scales ranging msec to minutes, whereas those related with the other variables need more time.

Line 30: In my humble opinion, I think that authors should consider the effect of light under ambient pCO₂. Authors state that "*growth rates remained unchanged in cells grown under ambient pCO₂*", however, according to the figure 1, increasing light had a negative effect on cells. In fact, they did not grow.

Line 38: Change significantly influenced by negatively influenced

Chl a fluorescence (Lines 12-13): I would rewrite this paragraph in a more direct style. For example, it could be rewritten as follow: "*Regarding to the individual effects of both factors on PSII_{max}, it was found that whereas light conditions decreased it (particularly at HL), OA increased (ca. 17% at LL) or did not affect it (i.e. at ML). Noticeably, lowest values were found under the Light×OA interaction (ca. 0.35) (Fig. 2A)*".

Chl a fluorescence (Lines 20-21): This paragraph is a little bit confusing. To avoid any misunderstanding I would rephrase this section as follow: "*F_v/F_m recovery was not influenced neither by light nor pCO₂ treatment, excepting under ambient pCO₂ and ML treatments in which the quantum yield's recovery was maxima (Fig. 2A)*".

Chl a fluorescence (Lines 22-25): Similarly than mentioned in the previous paragraphs, such idea could be simplified as: "*For [RCII], increasing light conditions reduced a 39% the functional RC; however, OA did not show a clear response pattern, as it both decreased (~XX% at LL) as increased (~44% at ML) the functional RC (Fig. 3A)*".

Chl a fluorescence (Lines 26-34): In my humble opinion, in this paragraph is difficult extracting the main finding, and what is the effect of light, OA and their interaction. Also, I sincerely think that authors should present this dataset at the same way than for figure 1-3, in a bar chart. If I see the table 3 presented, I would summarize the findings as follow: *"In relation to the individual effects of light and OA on the PSII photophysiology it was found that increasing light conditions exerted a mostly stimulatory effect, excepting on connectivity (i.e. P) where it was inhibitory. OA had a significant positive effect at LL on σ PSII, ETRmax and Ik, but significantly inhibitory on α at ML. Light and OA, as a single factors, did not exert any significant effect on P and τ Qa. At the same way than mentioned above, the LightxOA interaction had a contrasting effect on the PSII-photosynthesis, being stimulatory on σ PSII, ETRmax and Ik, and inhibitory on P and τ Qa. The interactive effect of both factors did not alter α ".*

Chl a fluorescence (Lines 37): I suggest that figure 4 could be moved to supplementary information, as the core information related with this section/variable is contained in Table 3. Thus, and similarly than mentioned above to authors, Table 3 should be presented as figure instead table. Regarding to the Fig.4, authors can highlight into the text, the stimulatory effect of OA on ETR under LL, and the absence of effect by OA under ML. Also, that such stimulatory effect was coupled with significant? lower NPQ values (i.e. LL) or with not significant differences between amb and OA treatments (i.e. ML). Maxima ETR and NPQ values were found under HL.

Regarding to the figure 5, I suggest that it could be also moved to supplementary information together with figure 4, and merge both figures into a single one but with 6 panels.

Finally, despite the main goal of this study was assess the interactive effect of HL and OA, authors did not explicitly quantified such impacts on the response variables assessed. Considering that only the OA effect could be calculated, as cells did not grow under HL, I suggest authors calculating the size effect in percentage (or the log response ratio) at least for the HLxOA interaction and for all variables, and show them together in a final figure. Using this approach could strengthen the message of your work, and easily showing to a potential reader if the HLxOA interaction was synergistic/antagonistic (or stimulatory/inhibitory), and on what variable(s).

Discussion: What is the main finding of your study?. It would be nice highlighting to a potential reader in a sentence or a paragraph what is the gap that this study fills (or contributes).

Lines 28-29: Based on the fact that significant higher ETRmax, Ik and alpha values were not coupled with higher POC, and ultimately growth, could it be plausible that cells were less efficient than under LL? Or more damaged?. Thus, this lower photosynthetic efficiency (or higher damage) would be consistent with a 2-fold increase in the NPQ, and ultimately, could support the higher Fv/Fm recovery (%).

Line 34: I think that "high light" should be modified to avoid any misunderstanding with the HL treatment.

Line 40: As the inability for growing of the target specie is a surprising result, and previous results have found that such phytoplankton group grow under HL and stratified conditions, I suggest that authors discuss the potential mechanism(s) or reason(s) that impeded that cell grown.

Lines 46 and 69: Statistical information can be omitted in discussion section

Implications: On one hand, it is true that the interaction HLxOA stimulated all assessed processes in *G. cryophila*, however, such responses were based on a short-term scale. It could be plausible that such beneficial effects would be accentuated at long-term scales by an adaptation of the populations, or by contrast, reduced. Authors could briefly discuss such issue in the context of their study.

On the other hand, in my humble opinion, I think that this idea presented at the end of the manuscript should be also highlighted into the Introduction section, as it represents the core about why assess the interactive effects of both global-change factors on Antarctic phytoplankton communities. Currently, we do not know how communities will respond to such changes, if such changes will lead to a diatoms or flagellates-dominated community, and as a consequence, if it will boost or reduce the C-sink capacity of this area.

Minor comments:

Tables 1-3: Please, include what lowercase letters mean. Are they representing posthoc comparisons?. Data represent means and SD?. And OA?.

Introduction (line 54): Change almost nothing by little is known