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Interactive comment on "Ocean acidification and nutrient limitation synergistically reduce growth and photosynthetic performances of a green tide alga *Ulva linza*" *by* Guang Gao et al.

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This paper reports results from an interesting study aiming to test the effects of ocean acidification and nutrients limitation on Ulva. The study is pretty straightforward: adult and juvenile algae were exposed to different conditions of CO2 and nutrients and their physiological response was investigated. While this study is rather "classical", the originality comes from the use of nutrient limitation, while most studies have used so far nutrients addition. The results are rather interesting and demonstrate that the interaction between pCO2 and nutrient limitations are not straightforward. I find the discussion a bit complex and hard to read given the quantity of physiological parameters

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discussed. It might be worth considering adding a figure that would summarize all the results. Maybe a schematic representing the physiological impact of nutrients and carbon could be added.

I have listed below some specific comments.

Response: We appreciate these comments very much and a schematic figure has been added to summarize all the results.

Abstract: indicate the duration of the experiment

Response: it has been clarified to “We cultured Ulva linza for 9-16 days” at line 6.

L55: Wrong reference for Cornwall et al. 2017, they looked at coralline algae not phytoplankton.

Response: We are grateful for this comment and this reference has been removed.

L63-64: reformulate this sentence

Response: It has been revised to “By analyzing the literatures, it is found that life stage can affect the effects of ocean acidification on growth of Ulva species” at lines 63-65.

L119: “LCHNHP” is a bit hard to read/understand but I guess it’s not really used later on.

Response: It has been revised to “The treatment of lower pCO2, higher nitrate and higher phosphate (LCHNHP) was set as the control.” at lines 121-122.

L130: How does this light level compare to in situ?

Response: The samples were collected in March 2017 and the light density of 300 photons m-2 s-1 used for the cultures was close to the ambient light level in situ. This information has been added to lines 134-136.

L132: What was the size of the tanks? Did you use any pumps, etc , to create water motion? This is critical as it could affect the capacity of the organisms to uptake

nutrients.

Response: The thalli were grown in 1-L balloon flasks containing 900 mL of media. The cultures were bubbled with ambient or CO2-enriched air at a rate of 300 mL min-1 to make the thalli roll up and down. Please see lines 140-141.

L133: Any reason to have chosen these durations? 9 days is rather short.

Response: The cultures had been finished before the thalli became reproductive as the aim of this study focused on the growth and photosynthesis. Different cultivation periods were used because the periods were different for sporeling and adult to become reproductive. These cultivation periods are enough for Ulva linza's acclimation to ocean acidification (Eggert, 2012; Gao et al., 2016). This information has been added to lines 147-151.

L156: What were those fragments? Just a piece of algae? I always have problem with this method, as I highly doubt it represents the response of the entire organism. When where the incubations done, at the end of the experiment? How many replicates were used?

Response: The text has been specified to "Algal individuals were cut into 1-cm-long segments with a scissor. Approximately 0.02 g segments were randomly selected and transferred to the oxygen electrode cuvette with 8 ml of media from the culture flask." at lines 175-178 and " The following parameters were measured at the end of the culture periods for each flask under each treatment." at lines 151-152.

L176: This was also done at the end of the 9 d?

Response: Yes.

Results: I would favour indicating the actual p-values rather than < 0.05 or >0.05

Response: We have used the actual P-values for most cases, with $P < 0.001$ for those where actual P-values were less than 0.001. Meanwhile, we hope we can keep P <

0.05 or > 0.05 for some cases where there are too many comparisons in one sentence.

L-314-315: Any reason why the algae would do that? If they have more carbon available why would they reduce their photosynthesis? It doesn't make much sense from an organismal point of view.

Response: We appreciate these comments. The explanation has been specified to "Meanwhile, the saved energy due to down-regulation of CCMs in thalli grown under HC combined with higher light density used in this study may depress PSII activity and thus reduce net photosynthetic rate (Gao et al., 2012)." at lines 340-344.

L 331-332: Could this be due to pH rather than carbon?

Response: Yes, there is possibility that the change of NRA was due to pH. The following information has been added to the text "Meanwhile, the change of NRA under different pCO2 levels might be also caused by varying pH as pH could affect NRA in seaweeds (Lopes et al., 1997)" at lines 363-364.

Lopes P F, Oliveira M C, Colepicolo P. Diurnal fluctuation of nitrate reductase activity in the marine red alga Gracilaria tenuistipitata (Rhodophyta). Journal of Phycology, 1997, 33: 225-231.

L344-345: CCM activity has often been linked to the light level. Could it explain some of these results?

Response: Yes, there are connections among CCM activity, CO2 and light. The related discussion has been added to the text and it reads " The potential reason is that the saved energy from down-regulated CCMs under higher CO2 levels could be used for growth at lower light levels but could inhibit PSII activity and thus growth at higher light levels" at lines 383-385.

As explained before, I think that an additional figure to summarize all of those results (and mostly the link between each other) would be highly valuable.

Response: It has been done. Please see section 4.4 for details.

L392-393: Could the seaweed culture also be affected by those limitations?

Response: The text has been revised to “This may hinder the occurrence of green tides and Ulva cultivation in future ocean.” at lines 430-431.

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