

General comments

The authors present a comprehensive set of short-term carbonate chemistry manipulations of natural phytoplankton communities from coastal and pelagic stations acquired during 3 different cruises. The manuscript is generally well written and clearly structured, and presents a large dataset. I have, however, major concerns about the interpretation of the data.

Response: We appreciate these comments very much, particularly for understanding the difficulty in obtaining a large number of data through in situ investigation and deck-based incubation covering a large area.

Firstly, the data is interpreted as if these were acclimated OA responses and not short term low pH assays investigating instantaneous CO₂/pH effects. These are two very different things than doing acclimated OA experiments, and show the response of the physiological machinery under current CC to short-term elevation on pCO₂, so an interpolation to future OA is invalid. Also, cells are probably stressed by change in environment, especially as 100% incoming light was used in this study, which is really high. Both of these aspects need to be made clear in abstract, discussion and conclusion.

Response: We have revised the manuscript based on the two points. Please see the specific response below.

My major concern is related to your interpretation of nutrient effects. While I would agree with your general hypothesis, this dataset is not at all suited to show and conclude this: In the methods section, the analytical limits for the nitrate and nitrite measurements are given as 0.1 and 0.04 $\mu\text{mol L}^{-1}$, respectively (L112-113), adding up to 0.14 for NO_x. This value is considerably higher than most of the NO_x values shown in Figure 6, potentially even higher than all except 2 or 3 values. It is not clear to me whether these are the surface values, or the ones from the bottom of the upper mixed layer the authors refer to in the results section (LL210-211). Regarding the former, this is an invalid approach. The samples you have collected and experimented with are from surface waters, so you cannot use the nutrient values from the bottom of the mixed layer as a reference. The latter have nothing to do with the physiological rates occurring in your short-term incubation assays! Also, you cannot conclude that the regional differences you observed are driven by nutrients if you have only sampled for nutrients on one of the three cruises (L115-117). Accordingly, large parts of the discussion and conclusions have to be completely re-written, and the other

environmental drivers (e.g. PAR and salinity effects, that are way more significant according to Table S1!) have to be considered more carefully.

Response: We agree with the reviewer that the nutrient values from the bottom of the mixed layer cannot be used to explain primary productivity in surface seawater. We have thus removed those data. As suggested, we have re-written the discussion and conclusions, focusing the effects of pH, PAR and salinity. Please see relevant sections for details.

Seasonality vs regionality: In L92-93 the timing of the three cruises is mentioned. While the Taiwan Strait cruise occurred in July, the other two cruises took place in September. These differences in the seasonal timing of sampling are not at all considered in the interpretation and discussion of the results. This should be included and discussed. Specifically, to which extent could the regional differences you find be caused by the differences in timing of sampling? Also the term 'region' and the separation between nearshore vs pelagic is not clear, you need to be more specific on how these were defined.

Response: We have added the discussion of seasonality and it reads "It is worth noting that seasonality may also lead to the differential effects of SA on primary productivity since the Taiwan Strait cruise was conducted in July and the cruises of the South China Sea basin and the West South China Sea were conducted in September. The SST and solar PAR intensity of the Taiwan Strait in July was 2–3 °C and $22 \pm 22 \text{ W m}^{-2} \text{ s}^{-1}$ higher than that in September (Zhang et al., 2008, 2009; Table S3). Although the effects of SA were not related to temperature as shown in this study (Table S2), the higher solar radiation in July may contribute to the positive effect of SA on primary productivity" at line 367. We have also redefined the "region" as suggested, we now use the terms of "the continental shelf (0–200 m, 22 stations) and the slope (200–3400 m, 44 stations) and the vast deep-water basin (> 3400 m, 35 stations). In the continental shelf, the areas with depth < 50 m are defined as coastal zones (9 stations)" instead of nearshore and pelagic at line 105.

Zhang C, Zhang X, Zeng Y, Pan W, Lin J. Retrieval and validation of sea surface temperature in the Taiwan Strait using MODIS data. *Acta Oceanologica Sinica*, 30:153-160, 2008.

Zhang C, Ren Y, Cai Y, Zeng Y, Zhang X. Study on local monitoring model for SST in Taiwan strait based on modis data. *Journal of Tropical Meteorology*, 25:73-81, 2009.

Currently the carbonate chemistry data from the incubations is missing, so that it is impossible to judge if the treatments were successful. Without them, an interpretation of the results is not possible.

Response: We have added these data and the relevant description reads "A series of onboard CO₂-enrich experiments in the investigated regions were conducted during three cruises. In the high CO₂ treatments, pH_{total} had a decrease of 0.34–0.43 units, while pCO₂ and CO₂ had an increase of 676–982 μatm and 17–25 μM kg⁻¹ SW, respectively (Table S1). Carbonate chemistry parameters after 24 h of incubation were stable (Δ pH < 0.06, Δ TA < 53 μmol kg⁻¹ SW), indicating the successful manipulation (Table S1)" at line 215.

Specific comments

L1-3: include inf that this is short-term exposure and not acclimation into the title

Response: The title has been revised to "Contrasting responses of phytoplankton productivity between coastal and offshore surface waters in the Taiwan Strait and the South China Sea to short-term seawater acidification"

L25-28: As explained above, this statement is not valid

Response: The ocean acidification has been corrected to seawater acidification throughout the text.

L28-32: I don't think you can make such general statements on long-term OA effects based on 24h incubations that did not allow for any acclimation. This need to be rewriting accordingly.

Response: It has been corrected to "Contrasting responses of phytoplankton productivity in different areas suggest that SA impacts on marine primary productivity are region-dependent and regulated by local environments" at line 30.

L36-38: I suggest including that this process is ongoing and likely intensifying.

Response: Corrected.

L102ff: In Biogeosciences, total alkalinity is usually abbreviated as A_T. Please use this term throughout the manuscript.

Response: We presume that the reviewer's suggestion is to change "TALK" to "TA". We have corrected it throughout the text.

L126-129: A lot more info needs to be given regarding the methods of OA manipulation. Was the CO₂-saturated seawater taken from the same location as the sample? If yes, how much time passed between sampling and start of incubation? How was the carbonate system manipulated (e.g. TA or DIC manipulation?), the decrease of pH units by approx. 0.4 units sounds like a unprecise approach. You need to at least provide a table in the appendix with pH values at the start and the end of each incubation to prove that your OA treatments were successful, ideally a fully constrained carbonate system with measured TA and DIC values.

Response: As suggested, the text has been clarified to "Seawater that was collected from the same location as PP and filtered by cellulose acetate membrane (0.22 μm) was used to make the CO₂-saturated seawater, which was made by directly flushing with pure CO₂ until pH reached around 4.50. When saturated-CO₂ seawater was added to the HC treatment, equivalent filtered seawater (without flushing with CO₂) was also added to the AC treatment as a control. The ratios of added saturated-CO₂ seawater to incubation seawater were about 1:1000. Seawater was incubated within half an hour after they were collected" at line 143. We have also supplied a table of carbonate system as suggested. The relevant description reads "A series of onboard CO₂-enrich experiments in the investigated regions were conducted during three cruises. In the high CO₂ treatments, pH_{total} had a decrease of 0.34–0.43 units, while pCO₂ and CO₂ had an increase of 676–982 μatm and 17–25 μM kg⁻¹ SW, respectively (Table S1). Carbonate chemistry parameters after 24 h of incubation were stable (Δ pH < 0.06, Δ TA < 53 μmol kg⁻¹ SW), indicating the successful manipulation (Table S1)" at line 215.

L133: 100% incident irradiance is really high, as samples don't get mixed down in an incubator. Please consider and discuss if OA effects may be driven by high light stress in those incubations with high PAR intensity.

Response: We have added this point and it reads "It is worth noting that the samples were not mixed down in the water bath and the 100 % incident solar irradiances may have high light stress on cells. Lower incident solar irradiances or some devices can be used to simulate seawater mixing in future studies" at line 303.

L133-134: On many ships, underway seawater supply still ends up being considerable warmer than SST due to the water running through a ship. Can you provide measurements of incubator temperature and offset to SST?

Response: That is true. The following information has been supplied "Due to heating by the deck, the temperatures in the water bath were 0-2 °C higher than in situ surface seawater temperatures" at line 155.

L178: Results from nutrient measurements are missing

Response: Nutrient levels in the surface water at most stations were undetectable. That is why we used the values at the bottom of upper mixing layers. We removed the data of NO_x since it is invalid to use them in explaining PP of surface seawater.

L192-193: please adjust to 'we observed that instantaneous effects of elevated pCO₂'

Response: Corrected.

L194: not sure which regions were compared. Please clarify.

Response: This is a general description of OA effects in all investigated regions. It has been clarified to "It was observed that instantaneous effects of elevated pCO₂ on primary productivity of surface phytoplankton community in all investigated regions ranged from -88% (inhibition) to 57% (promotion), revealing significant regional differences (ANOVA, $F_{(100, 404)} = 4.103$, $p < 0.001$, Fig. 5). Among 101 stations, 70 stations showed insignificant SA effects. SA increased PP at 6 stations and reduced PP at 25 stations" at line 220.

L197: 'was' should read 'were'

Response: Corrected.

L199: should read 'approaching the Mekong River plume' and 'A reduction'

Response: Corrected.

L203-206 and elsewhere: Again this region-related effect is not 100% clear, be more precise here, please. How did you define pelagic vs. near-shore, and is this the same definition you always use when talking about regions?

Response: We have redefined the regions and it reads "Investigation areas include the continental shelf (0–200 m, 22 stations) and the slope (200–3400 m, 44 stations), and the vast deep-water basin (> 3400 m, 35 stations). In the continental shelf, the areas

with depth < 50 m are defined as coastal zones (9 stations)" at line 104. Therefore, the text here has been revised to "Overall, the elevated pCO₂ had neutral or positive effects on primary productivity in the continental shelf and slope regions, while having adverse effects in the deep-water basin" at line 233.

L210-211: As explained above, this NO_x statistics approach is not valid

Response: We have removed the NO_x data as suggested.

L219-220: Based on your data this statement cannot be made!

Response: We have removed the information of nutrient and it reads now "Our results suggested that the enhanced effects of the SA treatment on photosynthetic carbon fixation depend on regions of different physicochemical conditions, including pH, light intensity and salinity" at line 247.

L234: should read 'reducing the removal rate'

Response: Corrected.

L237: RuBisCO abbreviation written differently in introduction. Please make consistent

Response: Corrected.

L245-276: all of this needs to be removed as you cannot conclude anything on nutrient effects based on your dataset.

Response: We have removed all of this as suggested.

L295-299: This statement cannot be made based on your dataset, rewrite following your actual data.

Response: We have rewritten it that reads "By investigating the impacts of the elevated pCO₂ on PP in the Taiwan Strait and the SCS, we demonstrated that such short SA-treatments induced changes in PP were mainly related to pH, light intensity and salinity based on Pearson correlation coefficients" at line 376.

Reviewer's comments for

Ms. Ref No.: bg-2021-326

Title: Contrasting responses of phytoplankton productivity between coastal and offshore surface waters in the Taiwan Strait and the South China Sea to future CO₂-induced acidification

Authors: Guang Gao et al.

Submitted to: Biogeosciences

General comments

This paper examines the impacts of ocean acidification on primary productivity using a number of deck incubation experiments. A dataset of 101 experiments is unique and valuable. I consider that an approach using multiple experiments under a protocol is reasonable to assess phytoplankton responses to ocean acidification. The dataset can be useful to draw some response patterns of primary productivity to ocean acidification under any key environmental conditions. However, I found some issues in the current manuscript, so these should be clarified before publishing this paper in the journal Biogeosciences.

[Response: We very much appreciate these comments and have revised the manuscript based on the reviewer's suggestions.](#)

Major comments

1. The authors claim that higher NO_x concentrations are responsible for the primary productivity enhancement under ocean acidification conditions. However, I consider that we cannot differentiate direct effect of NO_x levels on phytoplankton physiology from the effect of different phytoplankton community structures derived from different NO_x availabilities. Furthermore, NO_x data are very few relative to other parameters observed in this study. NO_x data are not at the surface but at the bottom of surface mixing layers. No direct link between the surface PP and NO_x at the bottom of surface mixing layer are not explained in the manuscript. I do not consider that a conclusion is not reasonably supported by the results presented in this study.

[Response: We have realized this issue and removed the data of NO_x as suggested by the two reviewers.](#)

Specific comments

2. L17 and L46. The authors mention “multiple drivers”, but this study treats only one driver of ocean acidification (pH). This mention does not fit to this study’s introduction.

Response: We did treat only one driver of seawater acidification but investigate its effects in different environments (temperature, salinity, solar irradiances, nutrients, etc.). To take the reviewer's suggestion, we have revised the text to "In order to examine effects of SA in changing environments" at line 17 and "it is important to understand the responses of the key players of marine biological CO₂ pump, the phytoplankton, to seawater acidification" at line 53.

3. L23-24. Are the digits of four reasonable?

Response: We have changed to two digits throughout the text.

4. L95-96. It is helpful to show the number of stations examined in each category of the coastal zone, continental shelf, slope, and deep basin.

Response: It has been revised to "Investigation areas include the continental shelf (0–200 m, 22 stations) and the slope (200–3400 m, 44 stations), and the vast deep-water basin (> 3400 m, 35 stations). In the continental shelf, the areas with depth < 50 m are defined as coastal zones (9 stations)" at line 104.

5. We need to know how the pH electrode was treated after the calibration. Just after the calibration using NBS buffers, electrodes usually do not work appropriately in seawaters because of the quite different ion intensity between the NBS buffers and seawaters. Such electrodes may be kept in a seawater for a certain period.

Response: What the reviewer mentioned is exactly right. We usually keep the electrode in seawater for half an hour before the measurement. This information has been added and it reads "After the calibration, the electrode of pH meter was kept in surface seawater for half an hour and then the formal measurements were conducted" at line 114.

6. How did the authors define the quantitative limits?

Response: It was determined by compared measured values with certified reference. However, this part was removed since the reviewer 1 does not think it make sense.

How were the nutrient samples stored until analysis?

Response: The nutrient samples were measured within 24 h upon sampling and stored in a 4 °C refrigerator before analysis. However, this part was removed since the reviewer 1 does not think it make sense.

7. Was the CO₂-saturated seawater filtered through 0.22 um before saturating? How was the CO₂-saturated seawater made? The authors should describe the typical volume of the added CO₂-saturated seawaters with the volume of pre-filtered seawater for incubations.

Response: It has been clarified to "Seawater that was collected from the same location as PP and filtered by cellulose acetate membrane (0.22 μm) was used to make the CO₂-saturated seawater, which was made by directly flushing with pure CO₂ until pH reached around 4.50. When saturated-CO₂ seawater was added to the HC treatment, equivalent filtered seawater (without flushing with CO₂) was also added to the AC treatment as a control. The ratios of added saturated-CO₂ seawater to incubation seawater were about 1:1000" at line 143.

8. The authors should explain the reason of selecting 0.4 pH unit.

Response: It has been clarified to "To study the impacts of projected SA (dropping by ~0.4 pH) by the end of this century (RCP8.5) on marine primary productivity in different areas (Gattuso et al., 2015)" at line 987.

9. In my understandings, whether 14C incubation for 24 h measures net production or not is under debate. In such condition, CO₂ enrichment in HC treatments may alter 14C incorporation manner in phytoplankton.

Response: We would like to take this suggestion and remove the term of "net". In HC treatments, there are more CO₂ and HCO₃⁻ and less CO₃²⁻ compared to AC treatments, which may alter 14C incorporation manner. However, 14C exists in all carbonate forms and theoretically the ratio of 14C to 12C is consistent in all forms. Therefore, this alteration of carbonate system may not affect the accuracy of carbon fixation using 14C technique.

10. chlorophylls a --> chlorophyll a

Response: Corrected.

11. The unit for Talk of umol/L?

Response: It has been corrected to " $\mu\text{mol kg}^{-1} \text{ SW}$ ".

12. The unit for PAR of $\text{W/m}^2/\text{s}$?

Response: The unit of $\text{W/m}^2/\text{s}$ can be used for light intensity, particularly for solar radiation (Wild et al., 2005; Yuan et al., 2021; Proutsos et al., 2022)

Proutsos, N., Alexandris, S., Liakatas, A., Nastos, P. and Tsiros, I.X., 2022. PAR and UVA composition of global solar radiation at 72a high altitude Mediterranean forest site. *Atmospheric Research*, 269, 106039

Wild, M., Gilgen, H., Roesch, A., Ohmura, A., Long, C.N., Dutton, E.G., Forgan, B., Kallis, A., Russak, V. and Tsvetkov, A., 2005. From dimming to brightening: Decadal changes in solar radiation at Earth's surface. *Science*, 308(5723), 847-850.

Yuan, M., Leirvik, T. and Wild, M., 2021. Global trends in downward surface solar radiation from spatial interpolated ground observations during 1961–2019. *Journal of Climate*, 34(23), 9501-9521.

13. The results of manipulation in carbonate systems should be described first. The readers need to know actual pH reduction, pCO_2 and CO_2 increase. The 0.4 pH reduction, even if this is identical for all experiments, would result different pCO_2 increase depending on ambient temperature, salinity, and carbonate system.

Response: We have supplied relevant description and it reads "A series of onboard CO_2 -enrich experiments in the investigated regions were conducted during three cruises. In the high CO_2 treatments, pH_{total} had a decrease of 0.34-0.43 units, while pCO_2 and CO_2 had an increase of 676-982 μatm and 17-25 $\mu\text{M kg}^{-1} \text{ SW}$, respectively (Table S1). Carbonate chemistry parameters after 24 h of incubation were stable ($\Delta\text{pH} < 0.06$, $\Delta\text{TA} < 53 \mu\text{mol kg}^{-1} \text{ SW}$), indicating the successful manipulation (Table S1)" at line 215.

14. The samplings of the bottom of upper mixing layers are not described in Materials and Methods section.

Response: This part has been removed based on the comments of reviewer #1.

15. Regardless of the importance of NO_x availability in this study (L220), spatial NO_x distribution was not shown. Furthermore, NO_x data are shown only 15, few data relative to other parameters, and this should be explained in the Materials

and Methods section. Six out of 15 data are located out of 95% confidence intervals in Fig. 6. Does this situation actually support your conclusion?

Response: We have realized this issue and removed the data of NO_x.

16. The paper shows nutrient data only for NO_x. We would like to know phosphate, silicic acid, ammonium, and probably iron levels.

Response: We agree with the reviewer that other nutrient elements may also affect the effects of OA on primary productivity. However, due to the limit of manpower and finance, other nutrient elements were not measured because most of them have very low levels in surface seawater of the South China Sea, requiring advanced instruments and well-trained technicians.

17. This discussion with a positive effect on diatoms is inconsistent with the discussion on a negative effect on diatoms in L237-238. Interpretations of the impact of OA on diatoms are not so straightforward as discussed here.

Response: Honestly, we are not sure which part is inconsistent with the discussion on a negative effect on diatoms in L237-238. In this study, OA showed positive, neutral and negative effects on primary productivity. We discussed the possible reasons that resulted in different effects. To clarify it, we have added some words and it reads now "Therefore, the net impact of OA depends on the balance between its positive and negative effects, leading to enhanced, inhibited or neutral influences, as reported in diatoms (Gao et al., 2012, Li et al., 2021) and phytoplankton assemblages in the Arctic and subarctic shelf seas (Hoppe et al., 2018), the North Sea (Eberlein et al., 2017) and the South China Sea (Wu and Gao 2010, Gao et al., 2012). The balance of positive and negative effects of OA could be regulated by other factors, including pH, light intensity, salinity, population structure, etc. (Gao et al., 2019a, b; Xie et al., 2022)." at line 268.

18. The "harmful algal blooms" is sudden and unacceptable, not discussed in the Discussion section.

Response: It has been removed.

19. 2. I am not sure whether the unit of total alkalinity is $\mu\text{mol/L}$ and that of PAR is $\text{W/m}^2/\text{s}$.

Response: As responded above, we have changed the unit of TA to $\mu\text{mol kg}^{-1} \text{SW}$ and hope to keep the unit of PAR as $\text{W/m}^2/\text{s}$.

20. 5 caption. L534-536. This is described only for this figure and can be removed because of the repetition of the Result section.

Response: It has been removed as suggested.