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Interactive comment on "Macroalgal metabolism and lateral carbon flows create extended atmospheric CO_2 sinks" *by* Kenta Watanabe et al.

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Author response to RC1 by Albert Pessarrodona Silvestre

We thank to your constructive comments. Below is reviewer's comments and our response to them.

Comment #1: This is a great study that provides some highly valuable and relevant new insights about the potential transport of macroalgal carbon. Although the export of DOC below the mixed layer is believed to be the main pathway through which macroalgal carbon gets sequestered in the ocean, our understanding of the fate of macroalgal DOC after its release is very limited. This study presents tempting evidence of its potential export to offshore waters (but see some concerns below), which is an important

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step to verify the role of macroalgae in oceanic carbon sequestration. Overall, I found the study to be well conducted and well written. The authors provide a set of comprehensive measurements of different carbon compartments and forms, which I applaud. Although I am not familiar with some of the more technical protocols of the sample analysis, further reading and consulting suggest that they are standard.

Response: Thank you for your comments and suggestions. We have modified the manuscript considering your suggestions. Please see details below.

Change: We have modified the manuscript considering your suggestions. Please see details below.

Comment #2: One of my principal concerns is that the authors have not yet established a direct transport link between the water exported from the macroalgal bed and the waters at the offshore site. The authors found that (1) water near the macroalgal bed had different properties (namely: lower DIC, fCO2 and higher DOC concentrations) than the water offshore except for February, when DOC concentrations were not significantly different. They then used mass balance models to simulate the diurnal changes in the carbonate and DOC system of the macroalgal bed (ln. 148); incorporating water exchange into their models helped better explain their readings (ln. 218, 245), which suggests that (2) there is water inflowing and outflowing at both the macroalgal bed and offshore site. There is however no direct demonstration that it is specifically the macroalgal bed water the one that reaches the offshore waters. This is a very important nuance, as the water that lowers the CO2 concentrations and enhances atmospheric CO2 uptake at the offshore site could come from other habitats that "produce" low DIC, high DOC water (e.g. seagrass meadow). Characterizing the DOC profile of both waters could help shed light on the provenance of that water.

Response: We agree with your comment. Our results show that low CO2 and high DOC water is exported from the macroalgal bed but this finding does not directly demonstrate that the macroalgal bed water reaches the offshore waters and affects

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carbon dynamics although we did not find such a low CO2 and high DOC water body mass around. Thus, we have rephrased the sentences about this claim throughout the manuscript. We have also added the discussion about the necessity of future study about the origin of water bodies affected by coastal vegetation.

Change: We have rephrased the sentences about this claim in Abstract, section 4.1, and section 5 as that macroalgal beds “potentially” create areas of adjacent water that serve as CO2 sinks. We have also added the discussion about the necessity of future study about the origin of water bodies affected by coastal vegetation.

Comment #3: The mass balance models only consider changes due to processes related to macroalgal metabolism, but some could argue that they are missing some parameters. For example, volatile and semi-volatile compounds can be an important fraction of the DOC, and can be volatilize to the atmosphere (Ruiz-Halpern, Vaquer-Sunyer, & Duarte, 2014) instead of remaining in the water column as assumed here. Similarly, some of the other processes that can affect the DIC pools (e.g. dissolution, chemical addition; (Langdon et al., 2003)), are not considered. If the authors consider that those fluxes are negligible that is fine, but they should provide evidence to back their approach.

Response: In this study, “DOC” did not contain volatile fraction of DOC because we measured DOC as non-purgeable organic carbon according to the well-established method. Because the model only calculated non-purgeable DOC, the volatilization of DOC can be ignored. About the DIC pools, carbonate dissolution and calcification were included in the mass balance model (Eq. 4).

Change: We have specified that our DOC was non-purgeable organic carbon.

Comment #4: It is very valuable that the study measurements were conducted at two separate time points albeit in the same season which gives an idea of the variability associated with the carbon flows estimated in the study. For instance, both the amount of DOCM and its constituents (as suggested by the different decomposition rates) were

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different across months (Wada et al., 2008). These points should be further elaborated to produce a rich and interesting discussion section. It would also be worth discussing how other species of macroalgae may differ in the production and characteristics of their DOC, as S. horneri was not the dominant species in the bed. Another limitation worth discussing is that DOC incubations for the degradation experiments were also maintained at a constant temperature (22), which may not necessarily reflect conditions in the field.

Response: The difference in the initial DOCM concentrations of macroalgae bags between February and March would be caused by the differences in the biomass and water volume in the experimental bags. We have added the discussion about this point. We have added the discussion about seasonal and interspecific variations in the release rates of refractory DOC by referring previous works. In this study, we used room temperature (22âĎČ), which is higher than in situ temperature, for both study periods to compare the quality of organic matter. We have added the discussion about the effect of temperature on the microbial degradation of DOC in the discussion section.

Change: We have added the discussion about this point as follows: “The difference in the initial DOCM concentrations of macroalgae bags between February and March would be caused by the differences in the biomass and water volume in the experimental bags (Fig. 4a, b). The variation of DOC concentration may affect the degradation rates via resource limitations for microbial activity (e.g., Arrieta et al., 2015). Understanding the fate of macroalgal DOC will be supported by assessing physical and biochemical factors regulating the microbial degradation of DOC.” We have added the discussion about seasonal and interspecific variations in the release rates of refractory DOC as follows: “Phlorotannin contents in macroalgal thalli have variations among seasons, growth phases, and species (Steinberg, 1989; Kamiya et al., 2010), which may regulate seasonal and interspecific variations in the biological recalcitrance of macroalgal DOC.” In this study, we used room temperature (22âĎČ), which is higher than in situ temperature for both study periods to compare the quality of organic mat-

ter. We have added the discussion about the effect of temperature on the microbial degradation of DOC in the discussion section.

Comment #5: Finally, some of the sections of the manuscript also need to be further clarified, as it is difficult for the reader to grasp how some very key parameters where calculated. For example, it is unclear how the gross community production, respiration and calcification were calculated from the DOC bag experiments (ln. 160), all of which are key parameters in the model. It is also not very clear how the tidal water exchange (EXtide) rate was estimated from changes in depth (ln. 169)

Response: We agree with your comment.

Change: We have added the equations and explanations for metabolic parameter estimation in Materials and methods section. We have added the equation for calculating EXtide for clarification and temporal changes in EX with water height in Fig. 5.

Specific comments Comment #6: Ln 33: Add “far” before “been”

Response: We agree with your comment.

Change: We have added “far” before “been” as per your suggestion.

Comment #7: Ln. 37 Add “more” before “efficiently”

Response: We agree with your comment.

Change: We have added “more” before “efficiently” as per your suggestion.

Comment #8: Ln 45: stored where? In the sediments, water column...? Also, consider citing here (Queirós et al., 2019), which provides an example of macroalgal-sediment connectivity.

Response: We agree with your comment.

Change: We have added “stored in sediments and water column” and the citation “Queirós et al., 2019” as per your suggestion.

Comment #9: Ln. 52: I suggest making the topic sentence of the paragraph the fact that DOC is believed (at least according to (Krause-Jensen & Duarte, 2016)) to be the principal pathway of macroalgal carbon sequestration (although). This will highlight more the relevance of this study, as more empirical support is needed to demonstrate the assumptions of (Krause-Jensen & Duarte, 2016)

Response: We agree with your comment.

Change: We have made the topic sentence as follows: “The export and persistence of macroalgal dissolved organic carbon (DOC) is proposed to be one of the principal pathways of macroalgal carbon sequestration but more empirical support is needed to quantify this carbon flow.”

Comment #10: Ln. 55: This paragraph feels a bit out of place here, you are talking about DOC and all of a sudden start talking about the carbonate system. Consider rearranging/rewriting.

Response: We agree with your comment.

Change: We have added the following sentence at the top of this sentence: “Even though macroalgal beds have the significant function of assimilating organic carbon, they could also be net CO2 emitters via air–water CO2 exchange depending on the chemical equilibrium in carbonate system in water columns.”

Comment #11: Ln. 61: The sentence gives the impression that the effects of macroalgal metabolism in their surrounding waters have not been studied, which is not the case (the authors provide plenty of examples). What is truly novel is examining its effects on other water bodies. I suggest deleting “both macroalgal beds and”

Response: We agree with your comment.

Change: We have deleted this phrase as per your suggestion.

Comment #12: Ln 67: Sargassaceous algae sounds a bit strange to me, perhaps

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just use Sargassum beds? Sargassums are also commonly found in tropical regions (Fulton et al., 2019), so I would suggest changing for “we focused on Sargassum beds because they are one of the dominant macroalgal habitats in temperate and tropical regions).

Response: We agree with your comment.

Change: We have replaced “Sargassaceous” to “Sargassum” through the manuscript. Also, we have changed the sentence as per your suggestion.

Comment #13: Ln. 69 The issue of carbon sequestration was not directly addressed in this paper, as no evidence that the carbon measured is locked away from the atmosphere for very long periods of time (decades-centuries) is presented. Although some of the DOC did not decompose after 150 days under constant experimental conditions, it is not known how long it would remain in the field or whether it could reach the mixed layer. I suggest cutting similar claims made throughout the ms

Response: The elucidation of the mechanisms of long-term CO2 sequestration by macroalgae is a final goal of our study but the present study addressed the mechanism of CO2 uptake by macroalgae. We have modified this point through the manuscript as per your suggestion.

Change: We have clarified our research goal as the assessment of the key mechanisms of CO2 uptake by the macroalgal bed in Abstract and Introduction section. (See also comment #2)

Comment #14: Ln. 75. Given that the water inflowing and outflowing from the bed is so important for this study, the readers would appreciate more details about the water movements around the study area (e.g. tidal characteristics, exposure)

Response: We agree with your comment.

Change: We have added the following sentence as per your suggestion: “The study site is characterized by relatively high tidal amplitude (<4 m) and adjacent to a deep

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strait (∼60 m)."

Comment #15: Ln. 79. This sentence is a bit redundant from the one in Ln 76. Consider merging them.

Response: We agree with your comment.

Change: We have modified this sentence as follows: "Surface water samples for analyses of DIC, TAlk, and DOC were collected from a research vessel three times during the daytime in both survey at five stations (H1–H5)."

Comment #16: Ln. 96. Is that the volume of seawater in the bag?

Response: Yes, this is the volume of seawater in each bag.

Change: We have added the sentence "in each bag" here.

Comment #17: Ln. 109. Please indicate the pore size of the filter. Was the filtering pressurized?

Response: We agree with your comment.

Change: We have added the information of pore size (0.7 μm) and filtering process ("under reduced pressure").

Comment #18: Ln. 127. What concentration of KHPh?

Response: We have added the concentration of Potassium hydrogen phthalate (83, 166, and 332 μM).

Change: We have added the concentration of Potassium hydrogen phthalate.

Comment #19: Ln 140. At what height was the wind speed measured at Agenosho?

Response: The altitude was 6.5 m.

Change: We have added this information.

Comment #20: Ln 143. Delete “that”

Response: We agree with your comment.

Change: We have deleted “that” as per your suggestion.

Comment #21: Ln. 148. Using the active voice is more readable in this instance. “We simulated the diurnal changes and budgets of the carbonate system and DOC in the macroalgal bed using mass balance models”

Response: We agree with your comment.

Change: We have modified the sentence as per your suggestion.

Comment #22: Ln. 151. This sentence seems to indicate that you changed the depth of the macroalgal bed. Please rewrite. Was the tide simulated by changing water height over the bed?

Response: We simulated the tide by changing water height over the bed.

Change: For clarification, we have modified the sentence as follows: “..., and the tide was simulated by changing water height along with the observed tide.”

Comment #22: Ln 152. The average Sargassum biomass used was derived from the field surveys, right? Please state so

Response: Yes, it is right.

Change: We have added the sentence “the average biomass of Sargassum algae obtained from the field survey”.

Comment #23: Ln 157. The amount of formulas, acronyms and parameters used in the manuscript can be a bit overwhelming. I encourage the authors to consider making a first figure with a schematic diagram of the different carbon pools and fluxes, as well as different carbon forms (e.g. POC, PIC, DOC, DIC...) and the processes that affect them (e.g. primary production, calcification...). That figure could include the formulas

in lines 157-159 to show how they were calculated in the mass balance models. I think this could be very useful to the reader.

Response: We agree with your comment.

Change: We have added the new figure with a schematic diagram of the different carbon pools, fluxes and processes (Fig. 2 in the revised manuscript).

Comment #24: Ln. 160. It is very unclear how all these parameters where calculated. Did you use some sort of relationship between DOC release and productivity? Please provide further details.

Response: We agree with your comment.

Change: We have added the equations and explanations for metabolic parameter estimation in Materials and methods section.

Comment #25: Lns 165-166. They can be just one sentence

Response: We agree with your comment.

Change: We have modified the sentences as per your suggestion.

Comment #26: Lns 192-193. They can be just one sentence

Response: We agree with your comment.

Change: We have modified the sentences as per your suggestion.

Comment #27: Ln 205. The use of “g WW” is more standard. Also wet weight (WW) needs to be abbreviated somewhere in the paper.

Response: We agree with your comment.

Change: We have replaced “g-ww” to “g WW” through the manuscript and added the abbreviation in Materials and methods.

Comment #28: Ln. 208-209. Please provide statistical evidence that the decrease in

time is statistically significant.

Response: We agree with your comment. We have conducted a Welch's two-sample t-test to detect the differences between the initial and final concentrations of DOC during degradation experiments. The conclusion has not been changed.

Change: We have conducted a Welch's two-sample t-test to detect the differences between the initial and final concentrations of DOC during degradation experiments. We have added the analytical process in the Materials and methods section and the results of this analysis in the Results section.

Comment #29: Ln 210. Perhaps it would be informative to include those final percentages in Fig. 4, as the decrease is a bit hard to observe in some panels (e.g. 4b)

Response: We agree with your comment.

Change: We have added these final percentages in Fig. 4.

Comment #30: Ln. 218. Please provide an index of how well the model fits the data. This way you can say that a model improves or worsens by adding/removing water exchange.

Response: We have added the explanation for the model improvement (the change in the RMSEs of every parameters) by considering water exchange in this paragraph and the legend of Fig. 5. In the previous version of our manuscript, model fitting was performed by minimizing RMSEs solely for DIC model but it may cause the uncertainty in other parameters (i.e., TAlk, DOC, and fCO2). We have modified this model fitting method as follows: "EXr was determined by fitting the models so as to minimize the root mean squared error (RMSE) compared with the observed values. RMSEs were calculated for the z-scores of DIC, TAlk, DOC, and fCO2 values, which were standardized anomalies from the mean observed values divided by the standard deviations. The EXr value that minimize the averaged RMSEs for these four parameters was determined for each survey." This modification has changed the results of water exchange rate and

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carbon budgets but the conclusion has not been changed.

Change: We have added the explanation for the model improvement (the change in the RMSEs of every parameters) by considering water exchange in this paragraph and the legend of Fig. 5. We have modified the model fitting method and the related results (Fig. 5 and Table 3).

Comment #31: Ln 238. Add “For example”, before DIC uptake”

Response: We agree with your comment.

Change: We have added “For example,” before DIC uptake” as per your suggestion.

Comment #32: Ln. 168: The estimation of water exchange is crucial for the aims of this paper. I am having a bit of trouble understanding how you EXtide was estimated from changes in depth. Is that referring to tidal height? It could be helpful if some example values are provided (e.g. is the number greater on spring tides, what is the maximum value it can attain? 1? What would that mean)

Response: We agree with your comment.

Change: We have added the equation for calculating EXtide for clarification and temporal changes in EX with water height in Fig. 5.

Comment #33: Ln. 256. I wonder how seasonality will affect the fate of the DOC released as well. How do oceanographic conditions vary in the study area?

Response: In this study, we did not collect the seasonal data for macroalgal DOC and oceanographic conditions, but we have added the discussion about this point (temperature and oceanographic conditions).

Change: We have added the discussion about the fate of macroalgal DOC depending on temperature and oceanographic conditions.

Comment #34: Ln 274. You may also be interested in the extensive work of Sophie

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Martin in maerl beds e.g. (Martin, Clavier, Chauvaud, & Thouzeau, 2007)

Response: We agree with your comment.

Change: We have added this citation and its NCP value in the revised manuscript.

Comment #35: Ln. 296-297. These two statements seem contradictory

Response: We agree with your comment.

Change: For better clarification, we have modified the sentence as follows: “Although the DOC release rates were similar between our two surveys, the percentages were very different between February (34 %) and March (6 %) (Fig. 6).”

Comment #36: Ln. 306. Very interesting find!

Response: Thank you!

Change: We have added the discussion about seasonal and interspecific variations in the release rates of refractory DOC by referring previous work. (See also comment #4)

Comment #37: Ln. 320. Insert “considered as” before “are”

Response: We agree with your comment.

Change: We have modified this sentence as per your suggestion.

Comment #38: Ln. 321. Consider “[...] export of particulate macroalgal carbon (e.g. entire thalli and fragments) to the deep sea [...]”

Response: We agree with your comment.

Change: We have modified this sentence as per your suggestion.

Comment #39: Figure 4. Consider stating the percentage of DOC remaining in each of the treatments of panels 4a and 4b as it is a big hard to tell how much remained sometimes. Also consider shading the area between the two treatments and indicating that it corresponds to the macroalgal DOC (DOCM; ln. 121).

Response: We agree with your comment.

Change: We have added the final percentage values of DOC in each treatment inside the panels in Fig. 4. We have also shading the area between the two treatments and indicating that it corresponds to the DOCM.

Comment #40: Figure 5. I think that plotting the value of EX in this graphs would be very valuable, as it would help the reader understand what is the water doing (inflow or outflow), and how this affects the readings at the macroalgal bed and offshore sites. The mass balance model should also predict the observations at the offshore site; please plot those ones as well.

Response: We agree with your comment about EX. The mass balance model cannot predict the observations at the offshore site because the values of the offshore site were used as endmember of inflowing water.

Change: We have added the plot of EX along with water height in Fig. 5. We have added the explanation for clarifying that the values of the offshore site were used as endmember of inflowing water in the Materials and methods section.

Comment #41: Figure 6. I suggest putting a dashed line through the middle of the panes to clearly delineate the offshore waters from the macroalgal bed. Also, put the titles of "Offshore" and "Macroalgal bed" at the very top so it is easier to read. I think that using symbols instead of the photo of the macroalgal bed would declutter the figure and make it more understandable. For instance, the ones at https://ian.umces.edu/imagelibrary/displayimage-search-0-4529.html are freely available (with attribution) and make for very appealing figures.

Response: We agree with your comment.

Change: We have modified figure 6 as per your suggestion. We have put dashed lines for delineating the offshore waters from the macroalgal bed. We have also put the titles of "Offshore" and "Macroalgal bed" at the top. We have made symbols of macroalgae

and used them.

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