

## Author response to RC2

Natividad and others explore the carbon balance and metabolism of a restored seagrass meadow. The analysis is largely interesting and methods sound, but a number of aspects of the presentation need to be improved and I have concerns about how the in situ results are translated to the real world environment.

Response: We sincerely thank the reviewer for the thorough and constructive evaluation of our manuscript “Estimation of Metabolic of Restored Seagrass Meadows in a Southeast Asia Islet Insights from Ex Situ Benthic incubation.

We acknowledge the reviewer’s concern regarding the translation of ex situ results to real-world in situ conditions. In this study, we used ex situ benthic incubation as a practical and widely used method to quantify seagrass metabolism, especially in subtidal systems where in situ measurements are logistically challenging. This method has been used in several studies and provide successful and valuable dataset. While we recognize that ex situ conditions may differ from natural underwater environments, we carefully designed our setup to mimic field conditions.

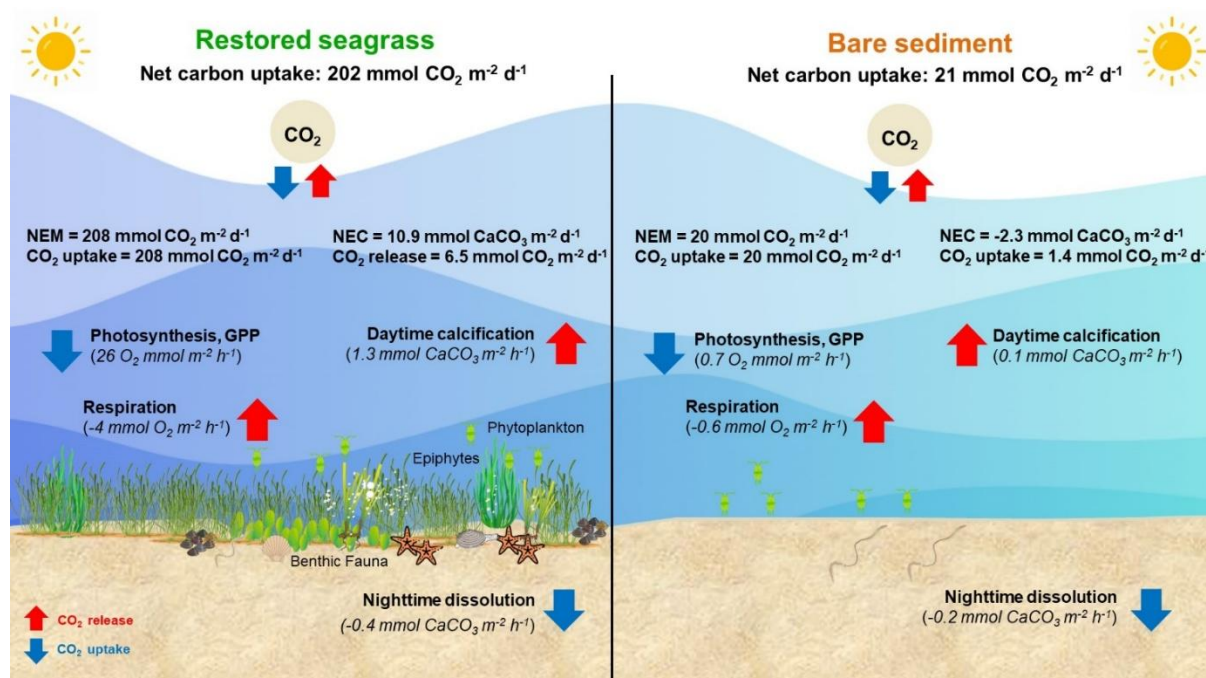
We have now revised the manuscript to clarify this methodological approach and added a statement discussing its strengths and limitations, including the need for future validation with in situ data under varying environmental conditions.

Line 122-125 *“This method offers a feasible approach for quantifying seagrass metabolism, especially in subtidal systems where in situ measurements are often logistically challenging. While ex situ conditions may differ from natural underwater environments, we carefully designed our setup to closely replicate field conditions, including natural light exposure and ambient temperature, to ensure ecological relevance.”*

Graphical abstract: units are needed on some terms (namely calcification).

Response: Thank you for the comment. We have revised the graphical abstract to include appropriate units (“mmol”), and the calcification rates are now clearly labeled as  $\text{mmol m}^{-2} \text{h}^{-1}$ . (Line 38)

During the revision process, we also identified an error in the previously reported values for calcification and dissolution. Specifically, although the units were indicated as  $\text{mmol m}^{-2} \text{h}^{-1}$ , the values reflected daily rates. We have now converted and corrected these values to accurately report hourly rates ( $\text{mmol m}^{-2} \text{h}^{-1}$ ), as originally intended. These changes do not affect the overall interpretation or conclusions of the study.



41: are there 72 species globally? Are species-level differences relevant here? Does this apply to just the grasses?

Response: Yes, there are approximately 72 seagrass species globally (Fourqurean et al, 2012; Short et al., 2011). Species-level differences are relevant, particularly in studies of productivity and metabolism, as seagrass species vary in morphology, physiology, and ecological function. These differences can influence carbon and carbonate dynamics, and thus are important to consider in site-specific assessments. To clarify, the term “seagrasses” here specifically refers to marine angiosperms, which are taxonomically distinct from terrestrial grasses.

145: did this reflect the PAR making it to the grasses in the natural environment with a larger water column on the order of 2-4 m? (see also line 197 and elsewhere). This feeds into the explanation on line 305: is PAR actually higher at leaf level in situ?

Response: We thank the reviewer for this insightful comment. In our study, PAR sensors were deployed in air to record incident solar radiation. However, based on our laboratory tests, when sediment cores were transferred to the incubation tanks, the light intensity decreased by approximately 50% due to attenuation through the water column and incubation setup. This attenuation resulted in light levels within the incubation tanks that were comparable to those in the natural seagrass meadows where samples were collected.

While we acknowledge that some differences remain between ex-situ and in-situ light environments. We believe that our approach provides a reasonable approximation of field conditions in the absence of in-situ incubations. Indeed, previous research has shown that ex-situ and in-situ incubations can yield comparable metabolic estimates, supporting the validity of our approach (Maher and Eyre 2011). We have added this clarification to the limitation of ex situ incubation and future research section to acknowledge this limitation and highlight the value of future in situ incubations for more accurately capturing the light environment experienced by seagrass leaves in their natural habitat.

Line 451-457

*“Moreover, we suggest validating the ex situ results with in situ data to ensure comparability with natural conditions, particularly the effects of light attenuation. Our measurements were obtained under ex-situ conditions in a shallow water column, which likely exposed the cores to higher irradiance than would be encountered in situ at different seagrass depths (2–4 m). While previous research has shown that ex situ and in situ incubations can yield comparable metabolic estimates, supporting the validity of our approach (Maher and Eyre, 2011), we acknowledge the need for future in situ incubations to more accurately capture the natural light environment experienced by seagrass leaves.”*

Ref: Maher, D., & Eyre, B. D. (2011). Benthic carbon metabolism in southeast Australian estuaries: Habitat importance, driving forces, and application of artificial neural network models. *Marine Ecology Progress Series*, 439, 97-115. (Line 637-639)

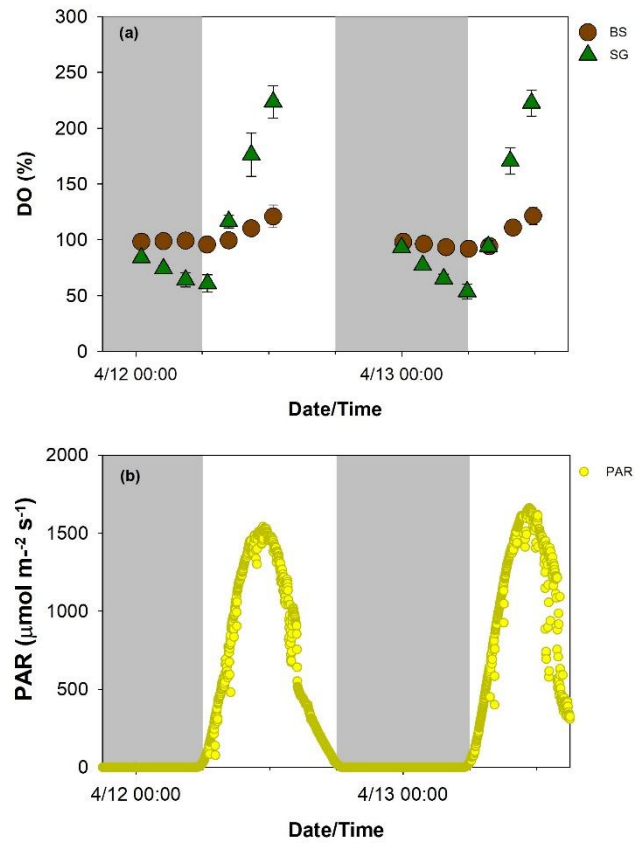
a bit critical of using hours for measurements because this is not an SI unit. Aggregating to days is a different story because this is an aggregation. I know that the community often uses hours, but a lot can happen in an hour. Perhaps note that these units are to compare against other studies.

Response: We acknowledge the reviewer’s point that hours are not SI units. However, regarding the temporal resolution of measurements, hourly measurements are standard practice in marine and plant metabolism studies because they capture fine-scale variations in light availability that are critical to photosynthetic processes.

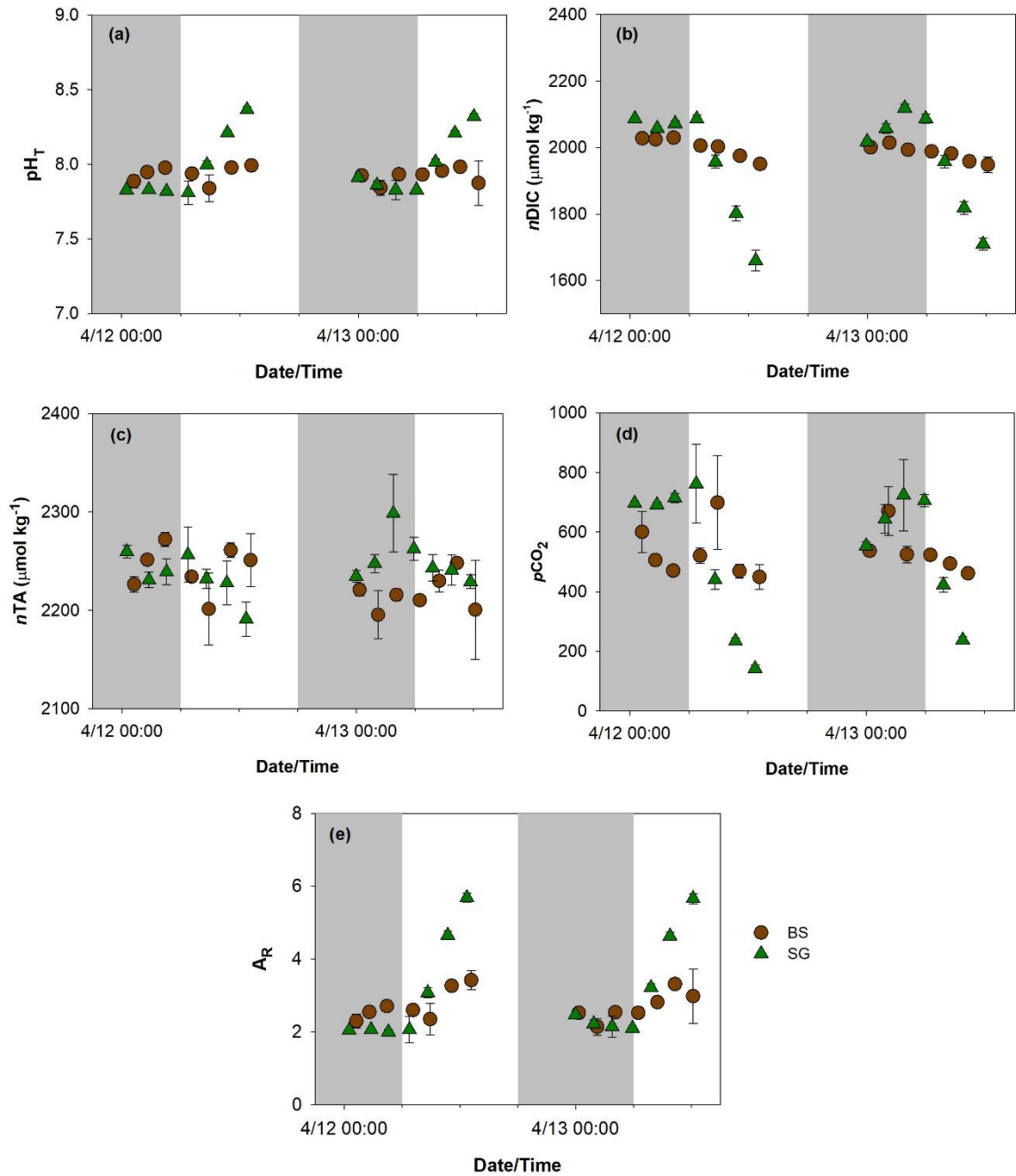
We agree that substantial changes can occur within an hour; therefore, we reported both hourly and daily rates in the main text. Hourly rates were used to examine diel variations in metabolic processes between bare and seagrass habitats, while daily rates were presented as net values to provide an integrated perspective on the overall carbon dynamics. This approach captures detailed variability across time scales (from hourly to daily). Moreover, several previous studies have reported data in a similar manner, facilitating comparisons across different systems and contributing to broader synthesis efforts. To clarify this, we have added the following statement to the method section on Benthic flux rates calculation Line 196-198: *“In this study, both hourly and daily rates were reported. Hourly rates allow us to examine diel variations in metabolic processes, while daily rates provide an integrated view of overall carbon dynamics, facilitating comparison with existing literature.”*

Fig. 3: does this pass a colorblindness check? At a minimum use differently shaped symbols. Also Fig. 4. And especially Figure 8. This figure would not be interpretable if printed in black and white, and not everyone has a color printer.

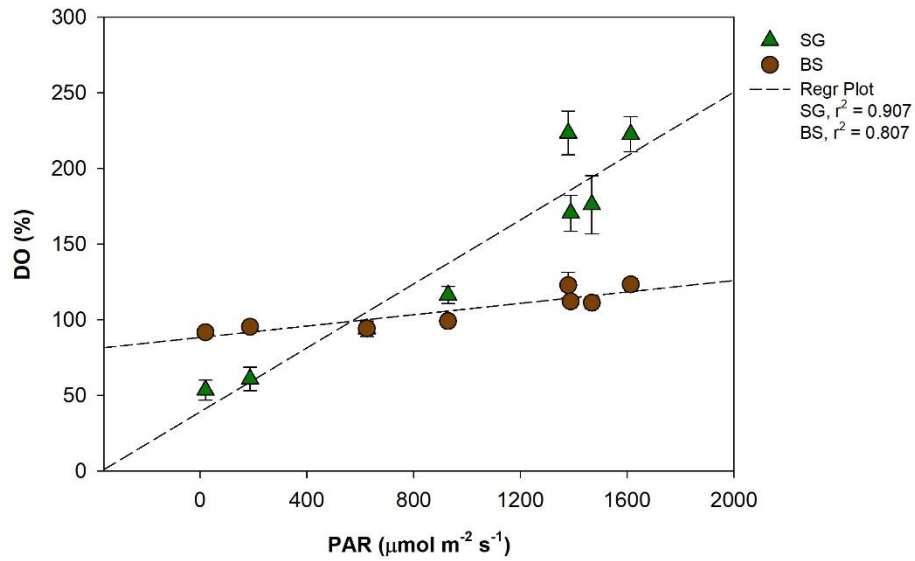
Response: Thank you for the helpful suggestion. We have revised Figure 3 (Line 230), Figure 4 (Line 2234), Figure 7 (Line 335), and Figure 8 (Line 386) to incorporate distinct shapes (e.g., circles and triangles) along with color, ensuring they are accessible for colorblind readers and interpretable in black-and-white printing.



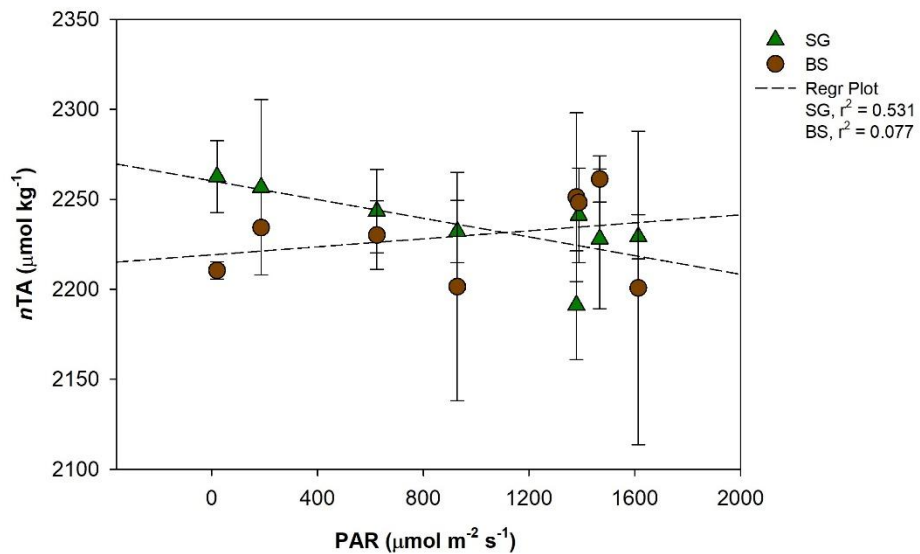
**Figure 1: Diurnal pattern of dissolved oxygen (DO, a) in replanted seagrass (SG, green triangle) and bare sediment (BS, brown circle) ( $n=9$ , mean  $\pm$  SD), and photosynthetically active radiation (PAR, b) during the two-day (April 12-13, 2024) incubation.**



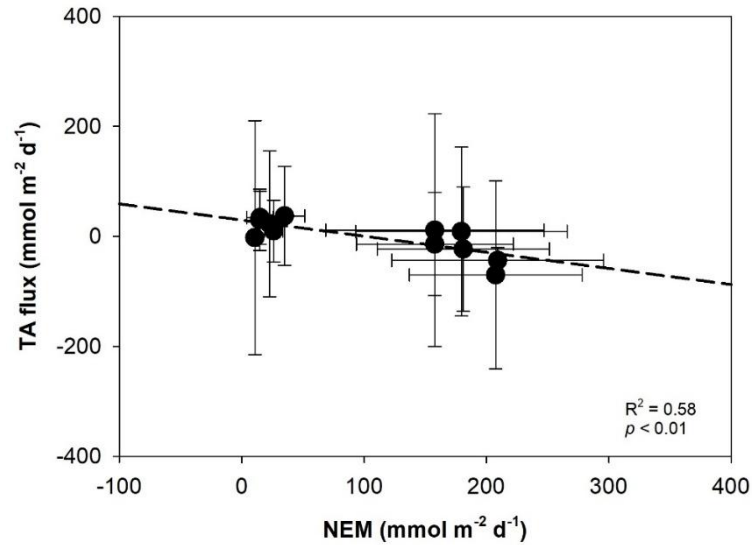
**Figure 2: Total scale pH ( $pH_T$ , a), normalized dissolved inorganic carbon ( $nDIC$ , b), normalized total alkalinity ( $nTA$ , c), partial pressure of carbon dioxide ( $pCO_2$ , d), and aragonite saturation state ( $\Omega_{AR}$ , e) in replanted seagrass (SG, green triangle) and bare sediment (BS, brown circle) during the two-day (April 12-13, 2024) incubation.  $n=3$ , mean  $\pm$  SD.**



**Figure 3: Regression plot between photosynthetically active radiation (PAR,  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ) vs dissolved oxygen (DO, %) in restored seagrass (SG, green triangle) and bare sediment (BS, brown circle). Error bars represent standard deviation (SD).**



**Figure 4: Regression plot between photosynthetically active radiation (PAR,  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ) vs normalized total alkalinity (nTA,  $\mu\text{mol kg}^{-1}$ ) in restored seagrass (SG, green triangle) and bare sediment (BS, brown circle). Error bars represent standard deviation (SD).**



**Figure 5: Linear regression showing the relationship between total alkalinity (TA, mmol m<sup>-2</sup> d<sup>-1</sup>) flux and net ecosystem metabolism (NEM, mmol m<sup>-2</sup> d<sup>-1</sup>) in restored seagrass meadows and bare sediment. Error bars represent standard deviation (SD).**

From the discussion, why is R suppressed in the seagrass ecosystem? The full mechanisms might be clear but it's important to explain what is happening to the best that the data will allow.

Response: We appreciate the reviewer's observation and agree that further explanation of the suppressed respiration (R) in the seagrass (SG) area is important, even if the full mechanisms cannot be entirely resolved with the current dataset. In response, we have revised the discussion to elaborate on the most likely factors contributing to the lower R values observed.

Specifically, we added that the SG beds are located in carbonate-rich sediments, which typically contain lower organic matter than siliciclastic or muddy sediments (Belshe et al., 2018; Kindeberg et al., 2018). This limits the availability of labile substrates for microbial decomposition. Furthermore, the organic matter derived from seagrass detritus is generally more refractory and less labile, which reduces its accessibility for microbial breakdown and thus suppresses heterotrophic respiration. While seagrasses can transport oxygen to belowground tissues via internal aerenchyma (Borum et al., 2006), supporting aerobic respiration, the combination of low organic content and substrate quality appears to constrain microbial activity and oxygen consumption.

We have incorporated this explanation into the revised manuscript (Lines 324-333), clarifying the likely mechanisms of respiration rates.