Replies to second round of reviews

A boost on the final stretch: intense river metabolism and wetland discharge increase aquatic CO_2 dynamics in the Danube Delta

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Public justification (visible to the public if the article is accepted and published):

Dear Authors,

Thank you for conducting a comprehensive revision in response to the reviewers' comments and suggestions.
The two reviewers from the first round have assessed the revised version and provided additional specific comments that would help enhance its clarity. Based on their reports, I recommend 'Reconsider after minor revisions' before the final version can be accepted. Please carefully revise your manuscript by incorporating the reviewers' comments and suggestions enclosed below.
When submitting the revised manuscript, please ensure that all changes are clearly shown in a track-changed version. Additionally, please provide a point-by-point response to the reviewers' comments, specifying the relevant line numbers in the revised text.
We look forward to receiving your finalized manuscript.

Sincerely,

Ji-Hyung Park
Associate Editor, Biogeosciences

Dear Editor,

Thank you for the opportunity to resubmit our manuscript after minor revisions. We have carefully considered all comments and revised the manuscript accordingly. Below, we show the reviewers comments in bold face and provide a detailed point-by-point response in plain text with quotes of the changed sentences in italics. Line numbers refer to the revised manuscript.

Sincerely,
The Authors

Review 1- second round

This paper is a revision of one that I reviewed before. Overall the paper is much better with a clear narrative structure around three questions. I very much like the structure of this paper.

Thank you for this positive assessment. We appreciate the reviewer's constructive comments during the first review round, which helped us to substantially improve the structure, focus, and overall presentation of the manuscript.

All three reviewers recommended not plotting exO2 vs exCO2 but rather plotting exO2 vs exDIC. The authors chose not to do so and with and explanation that I did not fully understand. It does not matter how much DIC is varying, the calculation is simply the excess DIC relative to what it would be if CO2 were at equilibrium, and not DIC by itself. I understand that the paper is for the authors, and not the reviewers, to write. But 3 reviewers saying the same thing requires stronger justification

We appreciate the reviewer's concern regarding the use of $exCO_2$ rather than exDIC. During the first review round, Reviewer 2 suggested the addition of DIC results to complement the CO_2 data: "Both the initial and community reviewers indicated the limitations of CO_2 data in high-alkalinity systems. While I recognize that adding new measurements to this already comprehensive manuscript would be challenging, calculating DIC to complement the CO_2 data would directly address these concerns."

We followed this recommendation and added DIC calculations to complement the exCO2 data in the revised manuscript of the last round. To emphasize the broad scope of our additional DIC analyses, we list some major changes made during the major revision after the first round of reviews.

- Line 91: "Analyzing the temporal dynamics of paired O_2-DIC measurements offers the advantage of removing buffer effects and simplifying carbon budget calculations (Shangguan et al., 2025)."
- Line 107 "Time series of dissolved O_2, CO_2 and DIC obtained from these campaigns allowed us to address the following specific research questions:"
- Line 222 "we included the DIC timeseries and paired exO_2-DIC measurements in our analysis"
- Line 262: "The inflowing Danube at Tulcea showed DIC minima of 2.6 mM in the warm season and maxima of 3.5 mM during the cold months (Fig. C1)."
- Line 274 "Compared to CO_2 and O_2, the 24-h DIC curves showed more stochastic deviations from the sinusoidal pattern, likely reflecting the rapidly changing DIC concentration in the inflow (Figs. C1, C2)".
- Line 296: "A similar pattern was observed for the median DIC amplitudes with 13 and 12 mM in Tulcea and Sulina, 50 and 23 in St. George and Chilia and elevated values within the delta: 23, 32 and 40 mM at Puiu-Rosu, Balanova and Busurca, respectively. Using DIC as an indicator, metabolic activity within the delta was 2-3 times higher than at the upstream Danube station. The results demonstrate how river metabolism intensified in the slow-flowing waters of the delta."
- Line 447 "Additional analyses of daily DIC variability support this result, although the DIC patterns exhibit higher stochasticity due to hydrological fluctuations."
- Line 474 "Consequently, the 24-h amplitudes of exO_2, $exCO_2$ and DIC all follow the sequence St. George > Chilia > Sulina consistent with decreasing lateral connectivity (Fig. 4)."

- Line 490: "The timing of daily cycles indicates a preferred sampling window around noon for obtaining representative values of O_2, CO_2 and DIC, whereas the early hours of 5-8 am and the late afternoon of 3-6 pm would introduce the largest sampling biases in the case of the lower Danube aquatic ecosystems (Fig. 3, Fig D1)."
- Line 518 "A quantitative assessment of the different lateral interactions affecting the three Danube River reaches was based on monitoring the average monthly difference in DIC concentrations between the downstream stations and the inflow at Tulcea (Fig. C3). Integrating these differences from April to December 2016 reveals an annual DIC addition of 0.5 and 1.2 mol m^{-3} yr^{-1} at Sulina and St. George, respectively."
- Line 611 "Balancing DIC increases along the final stretches of the main Danube branches revealed clear differences between low-carbon inflow from lakes in Ukraine and high-carbon wetland discharge in the southern part of the Delta."

All these additional results and discussions in first revision were based on DIC analyses which we illustrated with new figures (Fig. 4 and Figs. C1–C3). We thank the reviewers for advocating the merits of DIC calculations.

However, we decided to retained the exO_2-$exCO_2$ Figures 5,6, and 7, because they illustrate the gas exchange signal discussed throughout the study. The additional DIC analyses in Appendix C (line 681 ff) provide complementary information and allow readers to evaluate both approaches.

Consider a time reversal counterfactual: If Vachon et al. had written their excellent and influential paper plotting exO2 and exDIC would there be any reason at all to modify their analysis to plot exO2 vs exCO2? Would today one say "Vachon et al. 's approach of plotting exDIC does not work, thus we plotted vs exCO2"? I ask this question because if the answer is "no", then we cannot let being first in time to dictate what is in fact correct. And all three reviewers suggest that exDIC is correct.

We appreciate the reviewer's argument. However, we do not believe that the choice between exDIC and $exCO_2$ can be resolved through a time reversal counterfactual. Rather, the most appropriate representation depends on the scientific question being addressed.

In principle, our dataset allows us to relate measured O_2 to several components of the carbonate system, including dissolved CO_2, HCO_3^-, CO_3^{2-}, and DIC. All these concentrations can be calculated correctly and provide complementary perspectives on the coupled oxygen–carbon dynamics. We fully agree that exDIC is the preferred variable when addressing questions related to inorganic carbon balances and total inorganic carbon processing within the aquatic system.

In the present study, however, a key interest is the dynamics of CO_2 which determines emission rates. In this context, exO_2–$exCO_2$ plots provide a direct representation of CO_2 supersaturation and its relationship to biological and hydrological processes. To address the concerns raised during the first round of reviews, we additionally calculated and presented DIC as outlined in the list of bullet points above. Importantly, the DIC analyses support the same overall conclusions as the exCO2-based approach.

Table 1. Delete the correlation coefficient. Correlation and regression are related, but represent different analyses and should not be done together.

We agree and have removed the correlation coefficient from Table 1 (Line 350).

350. Methods in results. Instead, start with a finding that answers the clearly stated questions in the introduction.

Done. We revised the opening of this paragraph to begin with a key finding rather than methodological information. The paragraph now starts by highlighting the observed differences in the exO2–exCO2 covariance patterns between the Danube River reaches and the delta stations (Line 355):
"The following analyses address our second hypothesis that lateral inputs modify the downstream O_2-CO_2 dynamics. The ex-O_2-exCO_2 patterns reveal significant differences in the stretch of the 95% covariance ellipses between the main reaches of the Danube in 2016 and the stations within the delta in 2018 (Fig. 6)".

Table 2 Transpose so columns have common units.

Done. Table 2 has been transposed as suggested (Line 415 ff).

Table 3 Ditto. Great table!

Done. Table 3 has been transposed (Lines 433 ff). We thank the reviewer for the positive assessment of this table.

431. Seems like intro material. Findings would be better.

As suggested by the reviewers, we structured the Discussion around the guiding questions introduced earlier in the manuscript. In this paragraph, we provide context that helps interpret the findings related to this question. We agree, however, that the original text was unnecessarily long. We shortened it as follows (Lines 439 ff):

"Why is it relevant to study the differences in river metabolism between lowland river reaches and wetland channels? Our study provides evidence for significantly faster metabolic processes in the slowly moving waters of the delta wetlands compared to the main river reaches of the Danube. Monthly averages of 24-h amplitudes and the stretch of exO_2-exCO_2 covariance ellipses consistently indicate a 4-8 times more intense aquatic ecosystem metabolism within the delta. This distinction is particularly relevant in the context of ongoing wetland restoration efforts across the Lower Danube (Csagoly et al., 2018), where changes in hydrological connectivity may substantially alter carbon processing, nutrient retention, and greenhouse-gas dynamics (Hemes et al., 2018; Tschikof et al., 2022)."

577. Not for this paper but it seems like eddy covariance CO2 exchange would be needed to close the carbon budget of this wetland.

We agree with the reviewer. While a full carbon budget is beyond the scope of the present study, eddy-covariance measurements would provide valuable constraints for quantifying ecosystem-scale CO_2 exchange and closing the carbon budget of the wetland. We now acknowledge this point in the manuscript and note that wetland carbon budgets are often based on eddy-covariance observations (Line 591 ff). The paragraph ends now as follows:

"A complete assessment of the carbon budget would require ecosystem-scale CO_2 and CH_4 exchange measurements, for example using eddy-covariance approaches, which have provided the basis for many wetland carbon budgets (Zou et al., 2022)."

606. Intriguing idea of these CO2 emitting wetlands as net sink. Sink as in storing the excess NEP in sediments? Or exporting from the delta as OC? (Lovett et al. 2006, Ecosystems)

Good point. We addressed carbon accumulation and export in previous studies. We have therefore revised the sentence to distinguish between carbon storage within the wetland and organic carbon export pathways (Line 621 ff):

'' In summary, the well-connected delta wetlands provide a boost in ecosystem metabolism which leaves clear signals of the wetland CO_2 pump in the lateral discharge reaching the main Danube branches. At the landscape scale, however, there is evidence from the literature that the highly productive wetland areas may act as a net carbon sink, which retains organic carbon in the sediments and exports POC and DOC to the Black Sea (Durisch-Kaiser et al., 2010; Maier et al., 2021).

Reviewer 2 – second round

I recommend major revisions to this paper. This paper has been substantially altered from its initial submission and some improvements to the data analysis are evident. However, I fail to connect the three "guiding" questions: "1) How does the intensity of river metabolism differ between the open waters of the delta compared to the upstream Danube River? (2) How significant is the contribution of lateral inflows from lake systems and reedbeds to the O2 and CO2 dynamics of the main river reaches crossing the delta? (3) What are the overall effects of river metabolism and wetland discharge on aquatic CO2 emission rates in the Danube Delta?"

Thank you for highlighting the difficulty in connecting the three guiding questions. We agree that the links between them can be improved. To address this concern, we now provide a brief explanation in the Introduction describing how the questions are connected within a common conceptual framework (lines 112 ff).

"The first question addresses the intensity and characteristics of coupled O_2-CO_2 dynamics as indicators for differences in metabolic activity between river reaches and deltaic waters. The second question analyses O_2, CO_2 and DIC indicators for lateral inputs from lakes, channels and reed stands. Building on the evidence for metabolic activity and wetland discharge, the third question evaluates the consequences for aquatic supersaturation of CO_2 and resulting emission rates. By addressing these questions our study proceeds from the analysis of local O_2-CO_2 and DIC patterns and processes to ecosystem-scale emission rates. We show how high-frequency observations …"

First, metabolism is not measured, and so question one cannot be answered in the study.

We respectfully disagree with this comment. While we do not directly quantify gross primary production (GPP) and ecosystem respiration (ER), our analysis is specifically designed to investigate metabolic processes through the coupled dynamics of excess O_2 and CO_2. For instance, the review by Battin et al. (2023) emphasizes that ecosystem metabolism represents the integrated biological processing of carbon and oxygen within aquatic ecosystems and can be explored through a variety of observational approaches. The answer to our first research question is based on the comparison of the intensity and signatures of metabolically driven O_2–CO_2 dynamics between the Danube River and aquatic environments within the delta rather than direct measurements of GPP and ER.

Battin, T.J., Lauerwald, R., Bernhardt, E.S. *et al.* River ecosystem metabolism and carbon biogeochemistry in a changing world. *Nature* **613**, 449–459 (2023). https://doi.org/10.1038/s41586-022-05500-8

Second, "significant" has little meaning here, especially because lateral inflows are not measured.

We respectfully disagree with the statement of meaningless significance, but we clarified the text to address the indirect nature of the evidence. While lateral inflows were not quantified directly as water fluxes, the coupled O_2–CO_2 observations provide consistent evidence for their biogeochemical influence. The observed exCO_2 enrichments deviate strongly from the metabolic 1:1 O_2 :CO_2 stoichiometry expected for oxic river waters, and the covariance patterns, centroid shifts, and seasonal trajectories shown in Figs. 5–7 indicate CO_2-rich, O_2-depleted inputs characteristic of wetland discharge. This interpretation is further supported by previous studies in the Danube Delta and other wetland-connected river systems (Maier et al., 2021, 2022; Canning et al., 2021; Borges et al., 2019).
To avoid ambiguity, we have revised the second guiding question and clarified in the Discussion that our interpretation is based on the biogeochemical signature of wetland-derived inputs rather than on direct quantification of lateral water fluxes (lines 110-111).

"(2) How do lateral inputs from lake systems and reedbeds modify the O_2 and CO_2 dynamics of the main river reaches crossing the delta?'"

In addition, we explain this indicator approach in the discussion on lines 533 ff:

"Although lateral water fluxes were not measured directly, the combination of CO_2 enrichment, O_2 depletion, centroid shifts, and deviations from the metabolic 1:1 O_2–CO_2 stoichiometry provides a consistent biogeochemical signature of wetland-derived inputs. (Borges et al., 2019; Maier et al., 2021; Zuijdgeest et al., 2016)"

Third, wetland discharge is not measured. Instead, there are many figures/results related to O2 and CO2 dynamics. That's fine, but the focus of the paper would need to be revised substantially to align with the presented results.

We agree that wetland discharge was not measured directly and that no lateral water fluxes were quantified. However, we respectfully disagree that this prevents us from assessing its influence. Many hydrological and biogeochemical studies infer source contributions from characteristic chemical signatures rather than direct flow measurements. We cite Borges et al. (2019), Van der Knaap et al. (2025) and Xue et al. (2025) in the section of wetland discharge (Lines 499 ff). In our study, the coupled O_2–CO_2 dynamics provide a distinct biogeochemical fingerprint that is consistent with wetland-derived inputs. The observed CO_2 enrichments, O_2 depletions, covariance patterns, centroid shifts, and deviations from the metabolic 1:1 O_2–CO_2 stoichiometry collectively indicate the influence of CO_2-rich waters originating from connected wetlands and reedbeds.

To avoid ambiguity, we have clarified this point in the revised manuscript and now explicitly distinguish between quantifying wetland water discharge and identifying its biogeochemical effects. As already mentioned, we revised the second guiding question from

- "How significant is the contribution of lateral inflows…" to
- "How do lateral inputs from lake systems and reedbeds modify the O_2 and CO_2 dynamics of the main river reaches crossing the delta?" (Lines 110-111).

I insist on developing clear, testable hypotheses to clarify this work. What did the authors predict and how do the data compare to those predictions?

We agree that the hypotheses and their connection to the results should be stated as clearly as possible. We note, however, that the revised manuscript already presented three explicit and testable hypotheses in both the Abstract (lines 35 ff) and the Introduction (lines 69 ff).

"(1) Ecosystem metabolism intensifies when river water enters the slower flow paths through channels and lakes of a delta. (2) Wetland discharge from a delta significantly alters the oxygen and carbon dynamics in a river. (3) In such a case, average aquatic CO_2 emissions increase on the final stretch before a river reaches the sea."

To further improve clarity, we now explain the logical flow of the resulting research questions in more detail starting on line 112 as explained above.

Each figure should clearly relate to the questions and hypotheses.

We agree with this principle and note that all figures in the manuscript were designed to address the guiding questions and hypotheses. To improve clarity, we have revised the manuscript to make these connections more explicit.

- We added a link to our first hypothesis at the beginning of section 3.1 on river metabolism (Line 261) *"The results allow testing of the hypothesis that aquatic metabolism is more intense within the delta compared to the main river reaches. "*

- Similarly, we now introduce section 3.2 on wetland discharge with the sentence *" The following analyses address our second hypothesis that lateral inputs modify the downstream O_2-CO_2 dynamics."* (Line 355)

- We also introduce section 3.3 with our third hypothesis: *"Finally, we address the third hypothesis that a boost in aquatic metabolism in the delta and additional wetland discharge will increase the average aquatic CO_2 emissions on the final stretch before a river reaches the sea.»* (Line 407)

A conceptual figure could be helpful.

Good point. We added Fig 8 to the discussion section (Line 563).