We thank Reviewer 2 for the detailed and constructive feedback. The text in red indicates our response and the proposed modifications to the manuscript.

Reviewer 2:

"Perturbation increases source-dependent organic matter degradation rates in estuarine sediments" by Guangnan Wu et al. is a comprehensive and well-executed study investigating the chemical characteristics of sediment organic matter in Rotterdam harbour and the potential response of sediment carbon stocks to human disturbances (i.e. dredging). The study employs a range of techniques in the basic characterization of samples along a salinity transect (bulk sediment C/N, del13C, BIT index and pyrolysis-GCMS analysis of macromolecular organic matter) followed by a set of incubation experiments (both classical whole-core and homogenized sediment subaerial set-ups) to show how remineralization processes respond to disturbance. The data is of high quality and is generally processed and interpreted well, with a high degree of integration between the various lines of evidence. Overall, the outcome is convincing and should be published. However, I outline below one key issue related to the data processing that requires more careful consideration in the text, and several minor comments that should be addressed.

We sincerely thank the reviewer for their positive recommendation as well as the constructive feedback on our manuscript.

Key issue:

In processing the results of the whole-core incubations, ammonium fluxes are used to estimate the fraction of the DIC flux that is derived from organic matter remineralization (results shown in Fig. 5e). This calculation assumes Redfield stocihiometry for the degrading material, i.e. ammonium and DIC from remineralization are released in the ratio 106:16. This may be valid for the marine end-member site 115 but not necessarily for riverine end-member site 21A. Considering the bulk sediment C/N ratio data it is likely that site 21A releases more DIC per mole ammonium, even if the most reactive fraction of the sedimentary organic matter is relatively nitrogen rich. I suggest that this uncertainty is somehow included in the calculations, for example by presenting additional bars in Fig. 5e or a summary table. This issue is important because the quantification of remineralization-derived DIC production in the whole-core incubations is used later in the study when comparing with rates in the subaerial incubations, ie. assessing by how much dredging of sediment onto land would stimulate remineralization.

We appreciate the reviewer for pointing this out. We propose to make changes to Fig. 5e (see below). Indeed, the organic matter (OM) composition and C/N ratio differ between site 115 and site 21A (Table 2 below). Therefore, a different C/N ratio should be used when converting ammonium flux to OM-mineralized DIC flux (Fig. 5e). We will use different C/N ratios (with literature-reported ranges, Table 2) to account for OM compositional variability between site 115 and 21A and their corresponding uncertainties will be presented in a summary table (as Table 2). Besides the modification of Fig. 5e, we will carefully check and change the quantifications associated with Fig. 5e throughout the manuscript.

Proposed new Fig. 5, supplemented by Table 2:

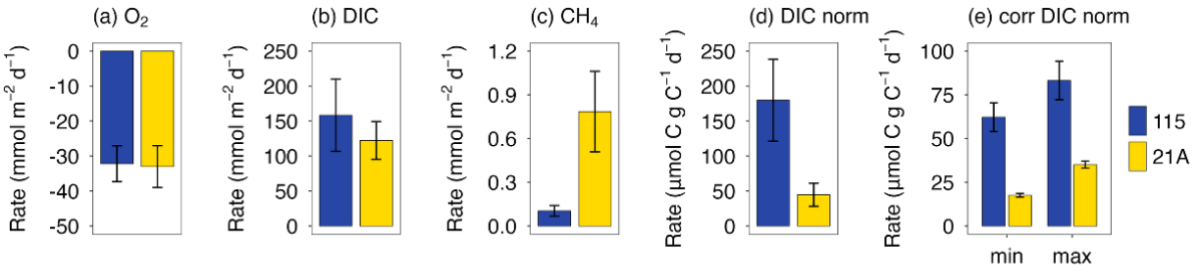

Fig. 5. *Benthic fluxes of dissolved O_2 (a), DIC (b), and CH_4 (c) determined from whole-core incubation. Positive and negative rates represent efflux (from sediment into overlying water) and influx (from overlying water into sediment), respectively. Sediment TOC-normalized DIC (DIC norm) is presented in panel (d) with TOC content being 2.2 wt.% for 115 and 5.0 wt.% for 21A. Panel (e) shows the DIN-corrected DIC fluxes (representing OM-derived DIC), normalized by sediment TOC (corr DIC norm), using minimum (min) and maximum (max) C:N ratios from literature (see Table 2). Error bars represent the standard deviation from triplicate core incubations.*

Table 2. *The bulk C/N ratios at sites 115 and 21A compared to values reported in literature in marine and riverine sediments. The literature-reported ratios were used to calculate DIN-corrected DIC fluxes (Fig. S4c) and TOC-normalized, DIN-corrected DIC fluxes (Fig. 5e) for sites 115 and 21A.*

Sediment	*Site 115 (marine)*	*Site 21A (riverine)*
Measured bulk C:N ratio	*7.2*	*9.9*
Literature-reported bulk C:N ratio in marine/riverine sediments	*6.625 (Redfield, 1958), 7.4 (Martiny et al., 2014), 5.6–7.5 (Körtzinger et al., 2001)*	*12 (Benoit et al., 2006), 13.1 (They et al., 2017), 7.1–13 (Maranger et al., 2018), 10.4–14.2 (Hecky et al., 1993)*
Measured DIN flux	*9.8 ± 1.3 mmol m^{-2} d^{-1}*	*7.0 ± 0.9 mmol m^{-2} d^{-1}*
Min C:N ratio for conversion	*5.6*	*7.1*
Max C:N ratio for conversion	*7.5*	*14.2*
DIN-corrected DIC flux (Fig. S4c), min mean (± SD)–max mean (± SD)	*54.7 (±7.2)–73.3 (±9.7) mmol m^{-2} d^{-1}*	*49.8 (±6.5)–99.7 (±12.9) mmol m^{-2} d^{-1}*
DIN-corrected DIC flux with TOC normalization (Fig. 5e), min mean (± SD)–max mean (± SD)	*62.2 (±8.2)–83.2 (±11.0) µg C g^{-1} day^{-1}*	*17.6 (±1.0)–35.1 (±2.0) µg C g^{-1} day^{-1}*

Minor comments:

Line 81-82: No need to highlight CH4 specifically. The point is valid for the balance between any given set of remineralization pathways.

We agree there is no need to highlight CH_4 specifically in this sentence, but it is important to mention somewhere that shifting salinity affects CH_4 dynamics (suggested by Reviewer 1). So, we propose that we delete "particularly for CH_4" in this sentence to keep this transition more general and smoother (Line 81–82) and introduce salinity-CH_4 dynamics in the following sentence. The revision is as:

> *This can lead to a strong spatial variability in OM degradation pathways and carbon dynamics~~, particularly for CH_4~~ (Cao et al., 2021). For example, SO_4^{2-} (usually higher in saline seawater) inhibits CH_4 production via multiple mechanisms (e.g. anaerobic oxidation of CH_4), which can lead to lower CH_4 emissions (Lovley et al., 1987; Egger et al., 2018).*

Line 84-87: Consider rewording the sentence ("estuaries" appears to be used in both a general and a specific way in the same sentence) and add the reference.

Will be changed.

Line 88: Should be "penetration depth".
Line 99: Make sure to use brackets correctly.
Line 123: Should be "dredged sediment".
Line 137-139: Should be "bottle incubations"
Line 139: Should be "Our results show..." or "We show..."
Line 141: Should be "the properties of OM influence..."
Line 213: Remove "an"
Line 383: Should be "overlying water".
Line 438: Should be "thus" not "this"
Line 478: Should be "may explain the observation that".
Line 607: Check the formatting of the citation.
Line 620: Should be "10 times faster than..."
We will double check and correct the typos and grammar throughout the manuscript.

Line 162: Use "end-members" or similar, rather than "realm"
Will be changed.

Line 220-229: It would be useful for a reader trying to reproduce this method to know how the HF was disposed of during the protocol. Is it evaporated at some stage? This is a dangerous chemical and lab protocols need to be carefully designed.
The HF waste was collected in a high-density polyethylene plastic container designated for HF waste, further handled by the EHS team (i.e. ARBO staff in the Netherlands). Details will be added in the revised manuscript.

Line 229: Clarify that "MOM" here refers to the residue of the previous steps.
Clarification will be made.

Line 283: It is not clear what is meant by "60% water-filled volume". It is difficult to estimate the porosity of freeze-dried and homogenized sediment after re-wetting due to changes in grain size distribution during these processing steps.
We thank the reviewer for the insightful comment. Here, the “60% water-filled pore space” refers to 60% of the porosity of the freeze-dried, homogenized sediment. We agree that the grain size distribution may change after rewetting, which could further affect the sediment porosity. To avoid confusion, we propose to simply describe the materials as “rewetted sediments”. For clarity, we will specify that the added volume of artificial rainwater corresponds to 60% of the porosity of the freeze-dried sediment.

Line 345: "Asterisk/stars" not "asteroids". Also unclear what the arrows in the plot indicate, should they be pointed in the opposite direction to link the numerical labels to the 2D fields in the x-y plot?

Will be changed.

Line 245: Should be Table S3, not S2.
We will double check the ordering of tables and figures.

Fig. 6: Please add the information about "marine" vs. "riverine" stations to the plot or legend.
Will be added.

Fig. 7: What do the stipple vs. solid lines indicate? Also show which sites are "marine" and "riverine".
Different line styles were intended to differentiate two sediment textures (i.e. sand-rich sediment vs. silt-rich sediment). To avoid confusion, we will use the solid lines for all subplots in Fig. 7. We will also specify in the new plot that site 115 is the “marine” site, and site 21A is the “riverine” site.

Line 451: This is a valid consideration rather than a confounding factor.
Will be changed.

Line 517: It is not unusual that soil OM contains similar chemical signatures to the higher plants that grow and decay in them. It is perhaps misleading to state that the correlation between BIT and the EMMM results implies low contribution of vegetation input, rather it should be stated that, as you conclude later, the plant signals are transferred via the soils (where they also pick up the BIT signal).
Thank you for noticing. The text will be revised as follows:

> *Its strong correlation with the modelled non-marine OM (encompassing soil OM, riverine OM, and terrestrial vegetation input) suggests that vegetation input was not a major component of the modelled non-marine OM contribution~~. Plant-derived OM, however, was suggested to be a major MOM constituent, with an abundance of lignin-derived products of up to 40% (Fig. 8b). Possibly, the lignin-derived products were mainly from eroded soils carrying plenty of OM debris from the plants previously growing on them, or the amount of vegetation input scaled proportionately with the amount of soil input.~~, or if not, vegetation OM was transferred from the soils, where they pick up the (enhanced) BIT signal. The latter assumption was supported by the MOM constituents, where the relative abundance of lignin-derived products (indicating higher plant signals) was up to 40% (Fig. 8b).*

Line 551-552: The comparison with ocean margin trends is not useful here as the reactivity gradients in near-shore areas is more likely to be controlled by terrestrial inputs than by water-depth fractionation of sinking OM.
We agree with the reviewer. This comparison will be removed in the revised manuscript.

Line 564: Remove "ancient". There is no information about age of the material so this is not relevant.
Will be removed.

Line 567-568: Interpretation of carbonate dissolution needs to be expanded. Is there evidence for this in the data or literature?

Yes, the DIC flux/DIN flux was ~16, larger than the C/N ratio (5.6–14.2; Table 2) of marine and riverine OM. This suggests that other pathways, besides OM mineralization, contributed to the carbon release. In this case, carbonate dissolution was a likely process. We propose the following changes in the revised text.

> *The respiratory quotient (RQ), determined as the ratio between DIC outflux and O_2 influx, was notably higher in the investigated estuarine sediments (3.75–5) than the typical range observed in marine sediments (0.69–1.31; Jørgensen et al., 2022), probably because carbonate dissolution enhances the DIC flux suggested by the DIC/DIN ratios (~16; Fig S4b), which was larger than the literature-reported C/N ratio of marine and riverine OM (5.6–14.2; Table 2), particularly for site 115 (marine).*

Line 577: The higher CH4 flux in a whole core incubation at a more freshwater site can be due to the higher position in the sediment column of the SMTZ. See e.g. the diagram in Fig. 7 of https://esd.copernicus.org/articles/13/633/2022/esd-13-633-2022.html.
Modifications will be made.

Line 633: Also refer to Keil et al. (1994).
Will be added.

Line 677-678: Separate the statements about global AOM estimates and dredging impacts, these are not obviously connected.
We propose the following revisions to the text in this paragraph:

> *Methane, a strong greenhouse gas, is often oversaturated in the OM-rich coastal sediments where CH_4 bubbles are formed. Most CH_4 is trapped below the sulfate-methane transition (SMTZ), and approximately 71% of the CH_4 is removed via anaerobic oxidation of methane (AOM) in marine sediments (Gao et al., 2022), but the removal efficiency is usually lower in estuarine sediments due to lower SO_4^{2-} concentrations. Dredging, which can remove the SMTZ completely or partially, is likely to impact the functioning of the AOM filter, consequently leading to a short-term CH_4 emission peak by increasing diffusion and ebullition (Maeck et al., 2013; Nijman et al., 2022).* ~~*Estuarine systems are characterized by a strong salinity gradient with a large variability of the depth of the sedimentary methanic zone. Anaerobic oxidation of methane consumes approximately 71% of the CH_4 in marine sediments (Gao et al., 2022), while dredging will inevitably disrupt anaerobic methane oxidation.*~~ *While dredging intensity and the SMTZ position can differ greatly within estuaries, further research should quantify the effect of dredging on CH_4 emission under realistic*~~*, large scale*~~ *and site-specific dredging practices.*

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