Response to referee comment by Tom Jilbert on “Biogeochemical controls on carbonate dynamics driven by methane and freshwater inputs in shallow sediments of a brackish continental shelf sea” by Łukawska-Matuszewska et al.

We would like to thank Prof. Tom Jilbert for the careful consideration of our manuscript and for all constructive and helpful comments. We will make every effort to revise the manuscript according to your suggestions. Below we respond to each point individually and describe the revisions we will make to address the concerns raised.

Interactive comment on “Biogeochemical controls on carbonate dynamics driven by methane and freshwater inputs in shallow sediments of a brackish continental shelf sea” by Łukawska-Matuszewska et al.

General

This paper presents an extremely comprehensive analysis of sediment biogeochemistry at three locations in the Gdansk Deep. The authors should be commended for the integrated approach to study solid-phase and porewater chemistry, which yields valuable insights into coupled primary and secondary diagenetic processes. In particular the work focuses on the factors controlling the rates of diagenetic mineral formation and burial (especially dolomite and pyrite) but touches on many other topics due to the breadth of the data.

I agree with most of the conclusions of the study but there are some interpretations that I would suggest to modify because their implications may be misunderstood if taken at face value. For example I do not think it is justified to state unilaterally that methanogenic sediments favor authigenic carbonate formation. I would also like to see some clarification of the separate measurements of del13-DIC and del 13C-CO2 included in the porewater analysis and how these results should be interpreted, and some more details about the incubation experiments and connected data processing. Overall I would say that the breadth of the approach introduces some difficulties to maintain coherence in the manuscript. Some rewriting of the introduction would help to guide the reader towards the rationale of the study in a more logical way. My Line by Line comments contain all the relevant points to be addressed. I wish the authors success in their revisions.

Thank you for the acknowledging the comprehensive nature of our work. We note that many of your substantive comments concern the Methods; we apologize for the lack of clarity in our descriptions. Because we measured a very large number of parameters, the original Methods became excessively long and we shortened it for the main text; in doing so several important procedural and analytical details were unfortunately omitted. We will expand the Methods to include more detailed description of procedures so the results can be unambiguously interpreted. Below we explain how we will address the main issues you raised, as well as other smaller comments.

Line by Line

Line 24: “In” rather than “At” the methane-free sediment

Answer: We will change “At the methane-free sediment” to “In the methane-free sediment.”

Lines 25 and 29: DIC production rates are given here in two separate units. For the sake of readability it would be sensible to use the same unit.

Answer: After the sentence "The role of methane-related processes became increasingly important in sediments influenced by freshwater input, where pore water freshening reduces sulfate availability, shallows the sulfate-methane transition and stimulates exceptionally high AOM rates reaching up to

1077 µmol dm^{-3} d^{-1}, resulting in a DIC production rate of 331 mmol dm^{-3} d^{-1}, more than fivefold higher than OSR.", we propose adding a follow-up sentence giving integrated rates over the upper 100 cm of sediment, expressed in µmol m^{-2} d^{-1} (to ensure consistent units and improve readability). The next sentence about authigenic dolomite burial will be left unchanged: "This environment facilitates significant authigenic dolomite burial (ranging from 467 to 994 µmol m^{-2} d^{-1}), illustrating efficient inorganic carbon sequestration within these sediments."

Lines 34-80 (general comments on Introduction): Lines 72-80 do a good job of outlining the eventual rationale for the study, but some of the earlier text is harder to read. I suggest to try to introduce the key biogeochemical processes to be studied in a more consistent way, e.g. treat primary and secondary diagenetic processes separately, then introduce the effects of salinity and submarine groundwater discharge, and finally focus on those processes that control alkalinity production as a driver of carbonate precipitation. The current flow jumps between these concepts and is not always consistent.

Answer: Thank you for this suggestion. We will revise the Introduction to follow the suggested, more consistent structure. Specifically, we will reorganize it according to the following outline:

1. Continental shelf seas significance in the global carbon cycle. Anthropogenic nutrient loading and coastal eutrophication.
2. Input and fate of organic matter to sediments; redox cascade of sedimentary organic matter oxidation (sequence of electron acceptors and transitions from aerobic to anaerobic respiration).
3. Organoclastic sulfate reduction in coastal sediments - prevalence and contribution to organic matter oxidation.
4. Methanogenesis and methane turnover in sediments - conditions favoring methanogenesis and pathways of methane oxidation.
5. Impacts of OM remineralization on DIC and total alkalinity (production, implications for seawater buffering, DIC-driven supersaturation and long-term burial mechanisms of authigenic carbonate precipitation).
6. Role of reoxidation and authigenic mineral formation in alkalinity dynamics - alkalinity consumption by reoxidation, alkalinity gain from mineral preservation (e.g., pyrite formation).
7. Regional context - shallow shelf seas and the Baltic Sea example: close benthic–pelagic coupling, high sedimentation rates, freshening effects, enhanced anaerobic remineralization, and shallow SMT dynamics.
8. Study scope and objectives: environments compared, measurements made (pore-water chemistry, sediment geochemistry, mineralogy, isotopes), rate experiments, and conceptual model development.

Lines 45-48: OSR should not lower pH or enhance carbonate dissolution because there is 2:1 production of HCO_3^- vs. H^+. Please check and rephrase.

Answer: We will revise and reorganize the Introduction as suggested; during that process we will carefully check for and correct any errors (including the pH/carbonate-dissolution statement) and rephrase the passage accordingly.

Line 91: Does this mean 7 cores in July 2023 and another 7 in February 2024? Please be specific.

Answer: We will clarify that seven sediment cores were collected during each sampling campaign: seven cores in July 2023 and seven cores in February 2024, and we will update the Methods text to state these dates and sample numbers explicitly.

Line 94: This should read "CH4, CO2 and their stable isotopes..." (but see later comments about needed clarifications about this methodology).

Answer: We will correct the text to read "CH_4, CO_2 and their stable isotopes..." as suggested. The methodological clarification is provided in our response to the comment about Supplement Fig. S3 (labelled important) on page 7 of this document.

Line 99: How was the rhizon sampling performed under anoxic conditions, was the setup placed in a glove bag?

Answer: Thank you for this remark. The current porewater sampling description given in lines 99–101 ("Pore waters were extracted from intact, sealed cores using Rhizon® samplers and 20 cm^3 syringes under anoxic conditions. Samplers were inserted through pre-drilled holes at 5 cm intervals. The uppermost sampler (1–4 cm above the sediment surface, depending on station) collected bottom water overlying the core.") is too brief and omits important details. We propose to replace it with the following text: Pore waters were extracted from intact, sealed cores using Rhizon® samplers (0.15 µm pore size). For each core liner, 4 mm holes matching the Rhizon® sampler fitting were pre-drilled at 5 cm intervals and sealed with a tape. After cores retrieval aboard, the tape was pierced and samplers inserted; to minimize contact with air, cores remained sealed throughout porewater sampling. The uppermost sampler (1–4 cm above the sediment surface, depending on a station) collected bottom water overlying the core. Porewater was drawn into attached 20 cm^3 plastic syringes. To minimize oxidation artefacts, all subsequent subsampling and treatment were performed immediately after porewater sampling.

Line 107: At what stage was a headspace injected into the methane sample vials? This should be stated.

Answer: Wet sediment aliquots were subsampled directly into gastight serum vials containing 2.5% NaOH to halt microbial activity and preserve dissolved gases. Vials were then sealed and a headspace was created and equilibrated immediately by shaking; headspace gas was subsampled and analyzed for CH_4 concentration.

Line 160: The term *IG* in equation (2) is not defined.

Answer: We will define IG in Equation (2) on first use as the loss on ignition (LOI) in percent of wet sediment.

Line 163-172: this section concerns porewater analyses so it is not clear why it is placed here in the sediment analyses section.

Answer: This section describes gas-phase analyses and was placed following the CH_4 concentration methods for logical flow. If the Reviewer prefers, we will relocate the entire description (CH_4 analysis plus C and H isotopes of CH_4 and C isotopes of CO_2 described in lines 153–172) to section 2.2.1 for clearer organization.

Line 168: What was the detection limit in terms of CH4 concentrations to obtain reliable delC13 data? Was a pre-concentration unit used?

Answer: Answer: The detection limit for obtaining reliable $\delta^{13}C$ data was 0.1% CH_4. No pre-concentration unit was used. We will add this information in Methods section.

Line 178: How were thin sections produced? Was the sediment embedded and if so with which resin?

Answer: Undisturbed air-dried samples of relevant horizons were embedded in a Technovit EPOX resin. A relevant information will be added to the Methods section (this will be inserted in line 179, after "SEM analyses of powders…").

Line 186: Clarify that *d* in equation 3 is the wet bulk density.

Answer: We will clarify that *d* in Equation (3) refers to the wet bulk density - we will change the wording to: "where d is the wet sediment bulk density (g cm^{-3})."

Line 190: Was the PXRD quantification achieved using the Rietveld method? Please comment on the uncertainty for estimates of dolomite and pyrite content in the ranges calculated in this study.

Answer: Mineral composition was quantified by Rietveld full-pattern X-ray diffraction fitting technique using SIROQUANT analysis of randomly oriented powders. A relevant information will be added to the Methods section in line 176.

The uncertainty was estimated from standard deviations on individual scale factors for each phase. These uncertainties are (in wt.%) 0.3 for pyrite, 0.5 for dolomite and 0.4 for calcite. We will add this in the Methods section.

Line 198: If the measured product is H_2S, how does this method to determine OSR exclude H_2S produced by SO_4-AOM?

Answer: Thank you for this comment. In the organoclastic sulphate reduction assays, lactate was supplied as a dedicated electron donor for sulphate-reducing bacteria at concentrations largely exceeding the potential availability of methane introduced with the 1 cm^3 sediment slurry. The sediment was pre-diluted with sterile seawater enriched with lactate to enable accurate handling of dense sediment, which further ensured an excess of organic substrate relative to methane. Therefore, sulphate reduction activity measured as H_2S production was driven by organoclastic sulphate reduction rather than sulphate-dependent anaerobic oxidation of methane. We will add in methods section: Lactate was added as an electron donor for sulphate-reducing bacteria during sample pre-treatment to ensure that measured H_2S production primarily reflected organoclastic sulphate reduction.

Line 237: 3.2 has the same heading as 3.1 (!)

Answer: We will correct the heading so Section 3.2 reads: "3.2. Composition of pore water and bottom water."

Line 286: This clarification of the incubation approach should also be stated in the Methods.

Answer: We will move the clarification of the incubation approach into the Methods section.

Line 296 (Table 1): The raw data used to calculate the rates needs to be included in the Supplement. Also it is interesting to note that MET-2 and ZGG are very similar in terms of experimentally-derived process rates. This does not seem to match with the porewater profiles, in which AOM appears much more important at MET-2.

Answer: Thank you for this suggestion. We will include the raw data used to calculate the rates in the Supplementary Material. Specifically, we will provide the measured H_2S concentrations obtained during incubations at 0.1 M sulphate concentration, which were used for rate calculations. In addition, we will include in the Supplement the fitted kinetic parameters derived from the rate versus substrate concentration curves. We also will add the parameters and conditions for determining CO_2 concentration as a result of methane oxidation (AOM).

The experimentally-derived AOM rates represent the potential activity of the microbial community under substrate-replete conditions rather than the actual in situ process rates. Therefore, the similar experimentally-derived rates at MET-2 and ZGG indicate a comparable microbial potential for AOM at both sites. In contrast, the porewater profiles reflect in situ conditions, where methane availability differs substantially between the sites. At MET-2, methane is present and supports active AOM, which is reflected in the porewater profiles. At ZGG, methane concentrations are negligible, so AOM appears less significant in situ despite the presence of a microbial community capable of performing this process if methane became available.

Line 308: Can this variable volume of "sediment matrix" be related to changes in water content downcore? The images imply so, but it depends partly on how the thin sections were prepared, see earlier comment and adjust accordingly.

Answer: The values presented here were obtained from planimetric analysis using an optical microscope. Up to half of the sediment was a mineral-organic matrix, grouped together due to the inability to distinguish individual components. Since thin sections were prepared from air-dried sediments, water content should not have been a significant factor.

Line 328: Where are the SEM-EDS results?

Answer: Initially, we decided not to include EDS spectra in the text due to its length. Representative spectra of pyrite, dolomite and calcite will be added to the Supplementary Materials.

Line 334 (Fig. 4): "c" appears to be pyrite and not calcite as stated in the caption.

Answer: Thank you for pointing this out. We noticed that there are a few mistakes here and hence the entire caption will be corrected as follows: Examples of SEM-BSE images of authigenic sulfides and carbonates: (a) pyrite framboids, MET2 (40–45 cm); (b) pyrite framboid and disseminated pyrite crystals, ZGG (10–15 cm); (c) pyrite framboids within a diatom shell, ZGG (40–45 cm); (d) pyrite framboid, diatom, dolomite, MET1-MP (40–45 cm: thin section); (e) euhedral and subhedral dolomite crystals, MET1-MP (10–15 cm; thin section); (f) euhedral dolomite, MET1-MP (10–15 cm; thin section); (g) dolomite, MET1-MP (40–45 cm); (h) calcite surrounded by clay minerals, ZGG (40–45 cm); (i) siderite, MET2 (40–45 cm).

Line 398: The diagenetic reactions are given different codes (e.g. R1, R2) in the main text and in the supplement. Please be consistent throughout.

Answer: We will revise the Introduction and resolve the reaction-numbering inconsistency. Specifically, we will move all diagenetic reaction equations to the Supplementary Information and refer to them from the main text (ensuring a single, consistent reference for each reaction), which avoids mismatched codes between text and tables. We will also reorganize the reactions in the Table S1 (please see table below) so they appear logically: (1) organic-matter degradation (primary redox reactions), (2) reoxidation of reduced by-products and AOM (secondary redox reactions), and (3) mineral precipitation/dissolution.

Line 426-427: Is this a reference to the incubation results of the present study? If so please be clear.

Answer: Yes, this refers to incubation results from the present study. We will clarify that by revising the sentence to: "Experiments with labeled substrates in the present study indicate the potential for AOM at ZGG, with rates up to ~10 µmol dm^{-3} d^{-1}."

Line 435: For completeness should denitrification be considered here?

Answer: In the reducing sediments we analyzed, porewater nitrate concentrations were consistently below the detection limit, and nitrite occurred only at trace levels, indicating that their availability was insufficient to support measurable denitrification under the study conditions. Given the redox conditions and the absence of oxidized nitrogen forms, the contribution of denitrification is likely negligible and was therefore not included in the quantitative DIC budget. We agree that nitrogen-cycling processes are important in shaping microbial redox pathways; however, these aspects are addressed in detail in a separate, complementary manuscript focused on the microbiological characterization of the studied sediments, which includes a comprehensive analysis of nitrogen-related processes. In our present work we propose to add a sentence: "Besides Mn(IV) reduction, organic matter oxidation in marine sediments can proceed via denitrification. However, in the reducing sediments examined here nitrate was consistently below detection limit and nitrite occurred only at trace levels (data not shown), so denitrification was not quantified. While denitrification can produce DIC, we have no evidence for substantial availability of oxidized nitrogen species in these samples and therefore expect its contribution to be negligible."

Line 439: Refer to Table 2 again showing the low values of dolomite at this site.

Answer: We will add a reference to Table 2 in the sentence: Despite thermodynamic supersaturation, carbonate contents remain low (Table 2).

Line 446-447: This logic is not quite correct, because as stated earlier in the manuscript, carbonate alkalinity, as HCO3- + 2(CO3 2-) is not quite equivalent to DIC. I am not against using these type of approximations to derive useful information from the dataset, but all assumptions should be made clear.

Answer: Thank you for the suggestion. We decided to use the DIC/TA ratio rather than CA/TA because DIC/TA is far more commonly used in the literature. At first use of DIC/TA we will add the clarification that DIC = $[CO_2^*] + [HCO_3^-] + [CO_3^{2-}]$ and $[CO_2^*] = [CO_2] + [H_2CO_3]$ (Dickson et al., 2007).

Dickson, A.G., Sabine, C.L. and Christian, J.R. (Eds.) 2007. Guide to best practices for ocean CO_2 measurements. PICES Special Publication 3, 191 pp.

Line 499: Should the reference to Fig. 4 actually be to Fig. 5?

Answer: We will change the in-text reference from Fig. 4 to Fig. 5 and verify all figure references for consistency.

Line 544-545 (important): I think the interpretation is more nuanced. Generally the formation of carbonates will be a function of the overall rate of the primary diagenetic reactions producing DIC and thus raising the IAP of dolomite and calcite. There is no mechanistic reason why methanogenic sediments (in the absence of sulfate) should favor carbonate precipitation; on the contrary OSR-dominated systems would be expected to have higher rates of diagenesis generally, due to greater energy yield of OSR. However, in systems where both process occur but overall diagenesis rates are so high that sulfate is exhausted in the uppermost decimeters (e.g. MET-1, MET-2 here), DIC will be observed to be higher overall (thus favoring carbonate formation), and by coincidence a methanogenic zone will be observed in short sediment cores that would otherwise be confined to deeper horizons. So I would suggest that one of the main outcomes of this study is to highlight how topographic features such as pockmarks influence the sedimentation regime such that demand for electron acceptors is concentrated in specific areas, leading to knock-on effects in secondary processes such as mineral formation (with contrasting outcomes for sulfides and carbonates).

Answer: Thank you for this remark. We will remove implication that methanogenesis per se causes carbonate precipitation and reframe the interpretation to emphasize that carbonate formation is controlled primarily by the overall rate of primary diagenetic reactions producing DIC (higher DIC → higher IAP → greater potential for carbonate precipitation), rather than by the identity of a single terminal electron-accepting process. We will specify that in methane-bearing sediments, methanogenesis and AOM are additional sources of DIC, whose importance is greater in organic-rich sediments affected by freshwater seepage that limits sulfate availability, as shown for pockmark MET1-MP.

We propose remove the sentence in lines 544-545: “These data indicate that methane-bearing sediments with active methanogenesis constitute a more effective sink for inorganic carbon than anoxic sediments dominated solely by OSR.". We propose to add, from line 542, that: Intense diagenesis leads to a strong increase in DIC, which raises porewater saturation with respect to carbonates and thereby promotes precipitation of authigenic phases.

We have reviewed the entire manuscript and will make corresponding revisions elsewhere. In lines 576–580, where we describe the pockmark as a sediment trap accumulating OM, we will add that the physical–sedimentary character of this system drives the discussed processes because OM accumulation increases demand for electron acceptors; as a result of intense diagenesis sulfate can be exhausted in the upper decimeters and the methanogenesis zone shifts toward the sediment surface. We will state that intense diagenesis is responsible for the high DIC, and we will link this to the statement that the very high concentration of ammonium and phosphate is consequence of the same intense diagenetic activity (lines 581–582). We will highlight the importance of methanogenesis and AOM as additional sources of DIC.

Also in Conclusions and Abstract we will clarify that authigenic carbonate formation is promoted where intense diagenesis elevates porewater DIC and saturation. We will provide a more comprehensive and accurate description of the sediments, rather than the brief term "methane-bearing sediments.

Line 973: Whiticar (1999) is missing from the bibliography. Check all references.

Answer: We will add

Whiticar, M.J., 1999. Carbon and hydrogen isotope systematics of bacterial formation and oxidation of methane. Chem. Geol. 161, 291–314. https://doi.org/10.1016/10.1016/S0009-2541(99)00092-3

to the bibliography and will check all references for completeness and consistency, correcting any missing or incorrect entries.

Supplement Fig. S3 (**important**): This point concerns the methodology for stable carbon isotopic analysis of dissolved inorganic carbon species used in the study, and how the data should be interpreted. Samples appear to have been taken in two separate ways: 1. in connection with the CH4 sampling, for which wet sediments were stored in 2.5%NaOH and then headspace gas was extracted for del13C-CO2; and 2. in connection with the DIC sampling, for which extracted porewaters were acidified with phosphoric acid and all DIC converted to CO2 for determination of del13C-DIC. A couple of queries here:

- for method 1, do the authors assume the data is reporting the del13C of only the dissolved CO2 (i.e. not the HCO3- and CO3(2-) derived fractions)?
- if so, how should those data differ from the del13C-DIC, if the entire DIC pool is in equilibrium? I checked the Whiticar (1999) paper and it seems that there is some interchangeable use of the terms del13C-CO2 and del13C-DIC in that study. For example in Fig. 7 of Whiticar the axis states “del13C-CO2” but the caption indicates “carbon isotopes of bicarbonate”. But if the authors are aware of literature of isotopic fractionation within the carbonate system in porewaters they should cite it here and explain how it impacts on the observations.
- please comment on how it was possible to achieve sufficient recovery of CO2 by method 1, if the treatment with NaOH shifted the equilibrium in the carbonate system towards HCO3- and CO3(2-)? Is it a question of how quickly the headspace samples were taken?

Answer: Thank you for highlighting this issue. We did not describe the sampling procedures for isotopic analyses in gases and pore water, which understandably creates uncertainty about data interpretation. We apologize for this. We acknowledge that additions of NaOH or acid alter carbonate-system equilibria; therefore we collect isotope samples separately (as described in our earlier works, for example in **Brodecka-Goluch A., Łukawska-Matuszewska K.**, Kotarba M.J., **Borkowski A.**, Idczak J., Bolałek J. (2022) Biogeochemistry of three different shallow gas systems in continental shelf sediments of the South-Eastern Baltic Sea (Gulf of Gdańsk): carbon cycling, origin of methane and microbial community composition. Chem Geol 597:120799). We will add a detailed description of the sampling procedures for isotopic analyses in gases and pore water to the Methods section.

1. For δ^{13}C-CO$_2$ (and δ^{13}C-CH$_4$): samples were collected in the same manner as the CH$_4$ samples but the procedure was carried out inside an N$_2$ glove box. Instead of adding NaOH, vials were pre-filled with Milli-Q water (this need to be added to section 2.1 on sampling, immediately after the description of methane-sampling procedure given in lines 105-107).

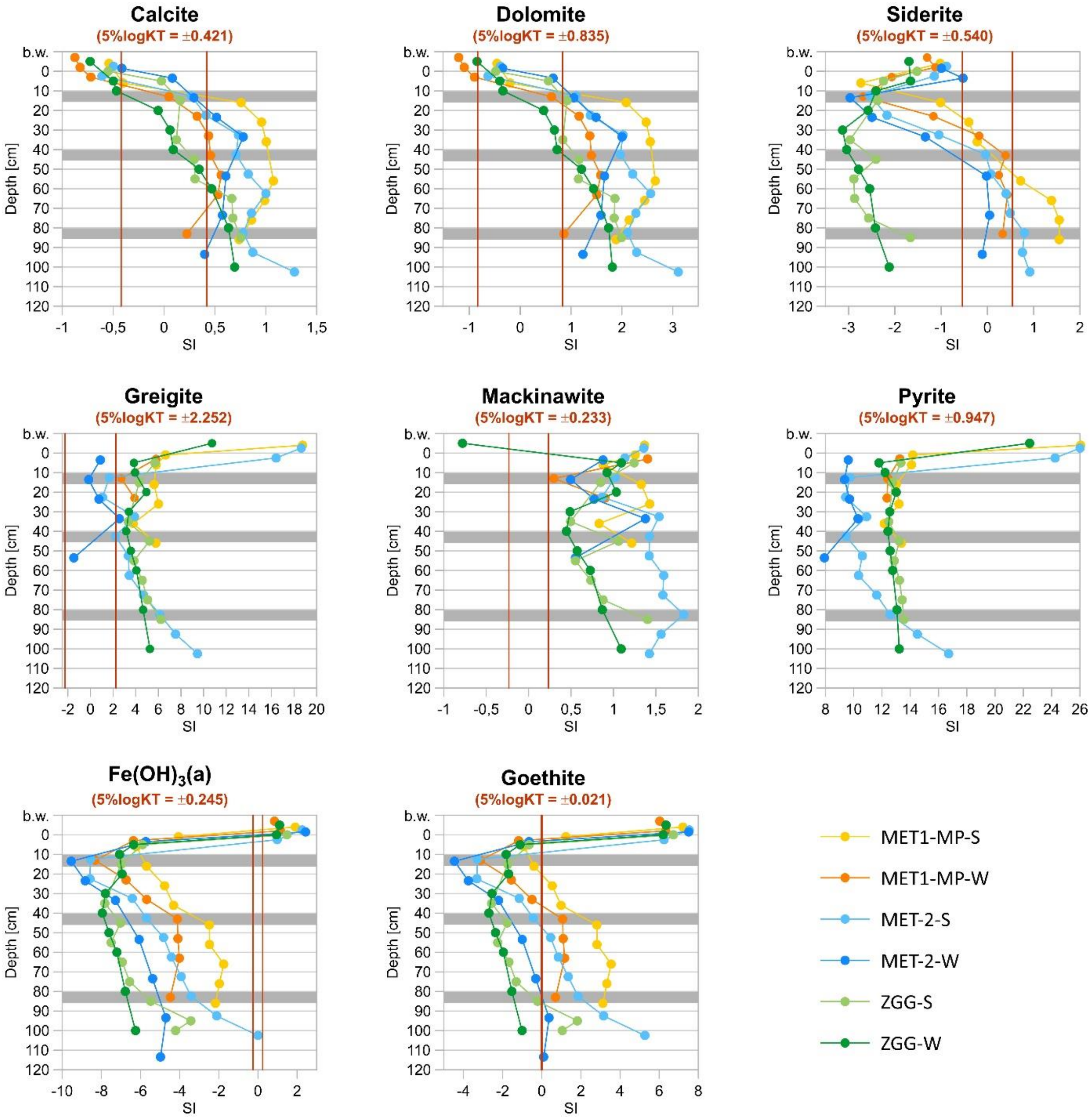
No NaOH was added to the vials used for the δ^{13}C-CO$_2$ headspace samples to avoid shifting the carbonate-system equilibrium. Samples were collected in an N$_2$ glove box into vials pre-filled with Milli-Q water (no alkali), sealed with butyl septa, and equilibrated immediately by gentle shaking to allow CO$_2$ to partition into the headspace; headspace aliquots were then taken promptly for isotope analysis. These steps (no NaOH, anaerobic handling, rapid equilibration and sampling) ensured efficient recovery of CO$_2$ into the headspace for isotope measurement.

2. For δ^{13}C-DIC: sampling was carried out in an N$_2$ glove box; porewater samples were collected into 2 mL septum vials that were filled by injecting porewater with one needle while displacing N$_2$ through a second needle; vials were completely filled with porewater and stored at 4°C prior to analysis (this need to be added to section 2.1 on sampling, right after the description of DIC-sampling procedure given in lines 102-104).

During analysis, samples were reacted with H$_3$PO$_4$ for 24 h at 25°C and the evolved CO$_2$ was analyzed for carbon isotope composition using a Finnigan MAT 253 isotope ratio mass spectrometer coupled to a Thermo GasBench II under continuous helium flow (this is described in lines 125-127). Therefore the data is reporting the δ^{13}C of CO$_2$ as well as bicarbonate and carbonate derived fractions (δ^{13}C-DIC).

Supplement Fig. S9: There is no legend so the color coding is not clear.

Answer: We will add a legend to Supplementary Figure S9 explaining the color coding as presented below.


Calcite
(5%logKT = ±0.421)
Dolomite
(5%logKT = ±0.835)
Siderite
(5%logKT = ±0.540)
Greigite
(5%logKT = ±2.252)
Mackinawite
(5%logKT = ±0.233)
Pyrite
(5%logKT = ±0.947)
$Fe(OH)_3(a)$
(5%logKT = ±0.245)
Goethite
(5%logKT = ±0.021)
Depth [cm]
b.w.
0
10
20
30
40
50
60
70
80
90
100
110
120
SI
-1
-0,5
0
0,5
1
1,5
2
3
-3
-2
4
6
8
10
12
14
16
18
20
22
24
26
-10
-8
-6
-4
MET1-MP-S
MET1-MP-W
MET-2-S
MET-2-W
ZGG-S
ZGG-W