

Persistent seasonal deoxygenation alters benthic metabolic processes and accelerates hypoxia feedbacks in the Baltic Sea

Corresponding Author: Professor Mirjam Perner

This file contains all reviewer reports in order by version, followed by all author rebuttals in order by version.

Version 0:

Reviewer comments:

Reviewer #1

(Remarks to the Author)

The manuscript "Persistent seasonal deoxygenation alters benthic metabolic processes and accelerates hypoxia feedbacks" by Alampalli et al, describes a 15 month long study, where the authors use Bocknis Eck in the Baltic Sea as a natural laboratory to investigate benthic responses to varying oxygen concentrations in the overlying water. They find that under well oxygenated winter conditions, benthic pathways for complex organic substrate breakdown, fermentation, aerobic respiration, nitrate- and iron-reduction are enhanced compared to low oxygen conditions in August/September, when organic matter degradation is diminished and sulfate reduction dominates. Intriguingly, they also show that re-oxygenation of the overlying waters in October doesn't immediately lead to a resumption of the winter state, but a period of enhanced aerobic metabolisms in the reduced sediments. The approach taken in the manuscript is extremely elegant, and an excellent way to gain insights into how benthic coastal systems respond to changing water column oxygenation; an important research question given global change. Overall, I found this manuscript a very enjoyable and informative read and was particularly impressed by the interdisciplinary nature of the research. I think there are few other such in depth studies addressing this question. As such, the manuscript will certainly be of interest to a broad community of benthic and pelagic marine researchers.

Given the above, I only have a few, relatively minor comments regarding the manuscript.

First, given the complex nature of the data presented, I think that the manuscript would really benefit from some kind of illustrative overview figure at the end to bring together the various different findings and their implications for increasingly hypoxic systems. I find that this would increase the reach of the findings to audiences who are maybe not so familiar with how to interpret the varying types of data presented in the main figures.

Moreover, I think that the authors could and should extend on the final paragraphs at the end of the manuscript, more clearly highlighting what these results might imply, in general, for a changing coastal ocean. Given the extensiveness of the dataset and the care given in analyzing it, I think that you have the grounds for some well supported and warranted hypothesising. For example, the manuscript title includes the words "accelerates hypoxia feedbacks" but the word feedback is never actually used in the discussion. It would improve the reach of the manuscript beyond a biogeochemical audience if you spell out the implications more clearly.

Reviewer #2

(Remarks to the Author)

This is an interesting study system that is ideal for addressing the globally important issues of coastal eutrophication/deoxygenation, so the potential impact of this manuscript is consistent with the scope of Comms E and E. This is also an outstanding dataset that combines excellent geochemical data, modeling, and an exquisite omics dataset--including 16S sequencing, as well as paired metagenomes and metatranscriptomes from 4 sampling periods. As a result, I found this manuscript to have great potential.

However, and unfortunately, this manuscript is clearly unpublishable in present form. I make multiple recommendations below to rectify this, but could not be completely comprehensive because there are several overarching issues that appear repeatedly throughout in the manuscript. My recommendation is that the authors rip apart and refocus the present manuscript on a coherent combined geochemical/omic story. They clearly have the necessary data to then produce an additional omics-oriented paper. I could envision direct gene-centric comparison between metagenomes and metatranscriptomes over time, along with MAG-based analysis that provides information on the 99 MAGs assembled here and maps reads back to these MAGs. This additional manuscript could cover all of the pathways and processes that are touched on here, but not directly relevant to the current manuscript (e.g., nitrogen cycle, methane, etc.). For the current manuscript, the overarching issues are the following:

(1) The abstract and end of the paper present a focused 'story' on Fe and S, yet the manuscript includes an enormous amount of information that is not relevant for this. I recommend reorganizing and refocusing around a series of hypotheses or expectations that are evaluated using the data. As part of this, I recommend including a conceptual figure of some kind that illustrates the changes documented by the authors. It is possible that something like this was provided in the Supplemental Info, but I did not review all of the SI content because...

(2) The main body of the manuscript does not contain enough information to stand on its own, particularly in the Methods. There are a series of text notes for each of the Methods; the choice to do this is strange and out of step. To be frank, I am not going to waste time clicking through all of these. For example, the omic Methods in the paper include less important info on how the samples were sequenced, but the far more important bioinformatic analysis is in a Supp Info text note. I also found the fact that Fe(III) data might be untrustworthy to be problematic. Personally I do not think that compromised data should be included in a manuscript.

(3) The Results and Discussion are divided, but the Results section includes some discussion and interpretation of the data as they are presented. I am not particularly militant about strictly separating the Results and Discussion, but the discussion in the Results section in the present manuscript creates confusion because it is too brief and not detailed enough (and see my comment below about the Discussion section itself). Just the very first paragraph of the Results provides several examples, but this occurs throughout the Results: on line 155, data "suggest... a highly reduced sediment matrix"; on line 157, data "are indicative of rapid removal..."; on line 159, data "may be related to high rates of carbonate dissolution..." Each of these statements needs more context and support in a combined R and D format, or should likely be removed—and again, there are many more such examples, this is just the first paragraph of the Results.

(4) Results that are not relevant for a more focused manuscript should also be removed. The most obvious examples of this issue are: the MAGs (99 bins, which could be an entire paper on their own!), the N cycle omic results (which seem interesting, but are not discussed), the CH₄ cycle omic results (which seem inconsistent), and the last paragraph of the Results. The authors should scrutinize what is or is not necessary to make their points.

(5) The Discussion has several strange portions where data are presented, then the discussion uses extensive information from the literature to infer what the data mean, but the two are not connected. There is also a lack of clarity about exactly which data are relevant for the ensuing discussion, esp. 16S vs. meta'omic. The text from 355-365 is an example of these issues; see also text on 387-392 and 398-403, but there are other examples. I recommend that this type of text could be converted into text in the Introduction formulated as expectations/hypotheses that are then tested by the data.

(6) Finally, there are some language/punctuation issues that should be addressed.

I apologize that I cannot be more encouraging about the manuscript in its present form, and want to re-emphasize the quality of the underlying work. I think that moderate editing and reorganizing could quickly transform the paper.

Reviewer #3

(Remarks to the Author)

I read with pleasure this article in which, through a combined approach of biogeochemistry and molecular biology, the authors analyze sedimentary processes in a coastal area of the Baltic Sea subject to stratification and depletion of dissolved oxygen in the waters adjacent to the sediment. The article is generally clear, well-written, and integrates many years of studies in which the effects of anoxia have been investigated in detail. The most interesting aspect of the results concerns, on one hand, the partial circularity of microbial processes and physicochemical characteristics during phases of full circulation and deep oxygenation alternating with phases of stratification, anoxia, and hypoxia; and on the other hand, the feedbacks that anoxia triggers on the benthic system in the medium term. What emerges is that anoxia erodes, over time, the sediments' capacity to buffer sulfide production and the rise of the sedimentary horizon in which these sulfides can be detected. In this sense, persistent anoxia affects biogeochemical dynamics and precludes a full recovery of the benthic system in biogeochemical terms, making it progressively more vulnerable. For this article, which seems well-supported by a large amount of work both on the traditional biogeochemical front (e.g., profiles, pools, in situ incubations, sulfate reduction measurements, modeling) and the molecular front (biochemical aspects, microbial diversity, genes expression and activity), I have some observations of a more philosophical than substantial nature, reported below.

Hypothesis: The abstract hints at a working hypothesis that is not explicitly stated in the final part of the introduction. I believe it is always important for a scientific study to explicitly state a hypothesis, even more so when relating to the effects of anoxia, an extremely well-studied topic.

Meio and Macrofauna: In the introduction and throughout most of the article—except for a single sentence (line 370)—there is no reference to meio- and macrofauna. It could be a valuable discussion point to include these groups of organisms in the

general interpretation of the results. Meio- and macrofauna play a crucial role in reoxidation processes and the distribution of labile particulate matter in sediments. It is possible that a feedback of hypoxia and anoxia also involves a community shift toward more tolerant forms with much lower ventilation rates, which has implications for the recovery of oxidized pools in the sediments.

Discussion vs. Results: In some sections (e.g., the paragraph starting on line 313), the discussion seems like a repetition of the results without citations, thus offering little actual discussion. The authors might consider revising this part to make it more of a discussion.

Integration of Approaches: The work is built on a temporal dynamic where biogeochemical and biochemical aspects are analyzed in detail. I would have expected to find at least one paragraph in the discussion where a discrepancy between a biogeochemical result and a molecular result was addressed. A small example is in lines 345-347: in a discussion, an author should explore why, in some cases, the two approaches are not aligned. I believe it is often through such misalignments that new pathways, organisms, and processes are discovered.

Long-term Consequences: Ultimately, the change triggered by anoxia affects iron and sulfur dynamics and the depletion of the sediments' buffer capacity against sulfides. The authors demonstrate this by comparing their data with previous research. Are these results aligned with those from perennially anoxic ecosystems where oxygen has been absent for decades? What do analyses of microbial diversity and biochemical processes in constantly anoxic sediments indicate? I felt the article lacked a concluding consideration projecting the consequences of sulfide upwelling on the diversity and functioning of microbial communities. This in turn has implications on benthic functioning, carbon retention, nutrient regeneration and stoichiometry, algal blooms...A starting point for such an observation could be drawn from the literature on permanently anoxic and sulfidic systems.

Version 1:

Reviewer comments:

Reviewer #1

(Remarks to the Author)

I am very happy with the revision to the manuscript. The changes to better explain the concept of feedback loops in the intro and at the end of the paper have strengthened it. Likewise, the addition of the final figure is very much appreciated and I think the authors have done an excellent job summarising their study with it.

Reviewer #2

(Remarks to the Author)

I reviewed an earlier version of this manuscript and the authors have successfully addressed nearly all of my comments. I appreciate the work that went into this revision and think this paper is a good fit for Comms E and E.

Reviewer #3

(Remarks to the Author)

Dear authors, I believe you have done an excellent job with the revision and I believe your contribution is important to the literature on oxic-anoxic transitions and buffer mechanism in aquatic systems. Furthermore, I believe it is a convincing, modern and cross-disciplinary contribution.

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Dear Reviewers,

We would like to express our gratitude to the reviewers for their constructive, thorough, and insightful feedback on our manuscript. We are pleased that the Reviewers recognized the elegance, interdisciplinary nature and potential impact of this comprehensive 15 month dataset. All changes were highlighted by “tracked changes” and are referenced by line number in the response below.

Major editorial requirements were addressed (details found under reviewer comments) and are summarized here:

1) Reframe the study around testable hypotheses and provide a solid discussion of your findings in the context of relevant literature and the potential implications

Response: Hypotheses were included in the introduction. Discussions have been expanded according to reviewers' suggestions and potential implications have been added to the final section.

2) Clearly describe all aspects of your methods to guarantee the reproducibility of the study and allow a sound interpretation of the results (including potential discrepancies between results from different methodological approaches)

Response: The methodology of omic analyses was moved from supplement to the main text. More details on the sequential Fe extraction were included as well as some more information on the sampling of seawater and sediments.

On “potential discrepancy between results from different methodological approaches” We have also included a section on the discrepancy between high Sox rates but lacking transcripts associated with sulfur oxidation in late October. We have also elaborated in more detail on elevated methane concentrations in the lower water column vs. the absence of known methane oxidizers/functional genes encoding for methane oxidation.

3) Consider the inclusion of a conceptual model that summaries your findings

Response: A conceptual model was included, see new Figure 8.

In summary, we have carefully considered all comments and structural recommendations. To address them effectively, we have introduced a new summary figure to visually map out our findings, integrated critical targeted terminology, such as explicitly incorporating the word 'feedback', directly into the discussion, and thoroughly expanded on the long-term consequences of sediment resistance decline by comparing our data with perennial anoxic systems. While incorporating these essential expansions, we have firmly and respectfully defended the holistic, multi-omic scope of our study against Reviewer 2's suggestion to split the paper, instead ensuring that the manuscript's focus is sharpened without sacrificing its broad ecosystem value.

Below, we provide a point-by-point response detailing the specific modifications made to the text.

Reviewer #1

Comment 1: “...the manuscript would really benefit from some kind of illustrative overview figure at the end to bring together the various different findings and their implications for increasingly hypoxic systems.”

Response: We enthusiastically agreed with this suggestion. To synthesize our complex multi-omic, geochemical and modeling datasets for a broader oceanographic audience, we have created a comprehensive conceptual diagram (Figure 8 mentioned in the Discussion page 28 lines 17 and page 29 lines 11 and 20). This figure visually summarizes the seasonal progression of the benthic community, tracking the ecosystem's transition from a well-mixed water column and benthic ferruginous winter state to a benthic degrading, sulfidic fall state over time. It integrates the key geochemical, microbial and biogeochemical processes identified in this study and places them into a coherent conceptual framework. We also added a comprehensive figure caption describing the underlying datasets, the seasonal progression, and the proposed mechanisms, thereby providing readers with a concise synthesis of our main findings and their broader implications.

Comment 2: *“...the manuscript title includes the words “accelerates hypoxia feedbacks” but the word feedback is never actually used in the discussion. It would improve the reach of the manuscript beyond a biogeochemical audience if you spell out the implications more clearly.”*

Response: The word feedback (including relevant processes) is now specifically mentioned throughout the manuscript. We have expanded the final section of the Discussion to explicitly detail these self-reinforcing mechanisms. The text now clearly spells out the biological and biogeochemical dynamics of the “hypoxia feedback loop” (page 29, lines 19ff).

Reviewer #2

Comment 1 & 4: *“The abstract and end of the paper present a focused 'story' on Fe and S, yet the manuscript includes an enormous amount of information that is not relevant for this. I recommend reorganizing and refocusing around a series of hypotheses or expectations that are evaluated using the data. As part of this, I recommend including a conceptual figure of some kind that illustrates the changes documented by the authors. It is possible that something like this was provided in the Supplemental Info, but I did not review all of the SI content because....*

Response: We have added hypotheses in the introduction (page 5, lines 14ff) and a conceptual overview figure (Figure 8).

Results that are not relevant for a more focused manuscript should also be removed. The most obvious examples of this issue are: the MAGs (99 bins, which could be an entire paper on their own!), the N cycle omic results (which seem interesting, but are not discussed), the CH₄ cycle omic results (which seem inconsistent), and the last paragraph of the Results. The authors should scrutinize what is or is not necessary to make their points”

Response: We respectfully disagree with the recommendation to remove these components or split the study into separate papers. The core novelty of our work lies in providing a holistic, ecosystem-level view of benthic responses to seasonal deoxygenation. In coastal sites like Boknis Eck, the iron and sulfur cycles do not operate in a vacuum; they are fundamentally coupled to alternative electron acceptors (the nitrogen cycle, cf. page 21 lines 13-15) and terminal carbon mineralization pathways (methane dynamics, inconsistency discussed page 22 lines 4-6). Removing the 99 MAGs and these interconnected cycles would reduce a comprehensive ecosystem study into isolated chemical silos, stripping the paper of its multidisciplinary value.

Comment 2: *“The main body of the manuscript does not contain enough information to stand on its own, particularly in the Methods. There are a series of text notes for each of the Methods; the choice to do this is strange and out of step. To be frank, I am not going to waste time clicking through all of these. For example, the omic Methods in the paper include less important info on how the samples were sequenced, but the far more important bioinformatic analysis is in a Supp Info text note. I also found the fact that Fe(III) data might be untrustworthy to be problematic. Personally I do not think that compromised data should be included in a manuscript.”*

Response: To comply with standard journal length constraints and optimize readability for a broad audience, highly technical bioinformatic parameters were originally placed in the Supplemental Information. However, to ensure the main text stands completely on its own, we have migrated the essential bioinformatic pipelines, assembly thresholds and mapping criteria directly into the main Methods section (compare page 8 onwards). We also added some more information on the sampling of seawater and sedimentary properties section, cf. page 6-7.

Regarding the Fe(III) data: We have revised the Methods section to provide a more complete description of the sample handling and clarified the wording regarding the potential artefacts Fe(III) data. Our intention was not to suggest that these data are untrustworthy, but rather that they should be interpreted with appropriate caution because some oxidation during sample handling cannot be excluded. To avoid overinterpretation, we have removed discussion based on small Fe(III) signals that could potentially reflect handling artefacts. The remaining interpretations rely only on robust patterns and we are confident that the conclusions drawn from the Fe data are supported by the dataset despite this acknowledged limitation (e.g. page 6 line 23ff, page 13 lines 15ff).

Comment 3: *“The Results and Discussion are divided, but the Results section includes some discussion and interpretation of the data as they are presented. I am not particularly militant about strictly separating the Results and Discussion, but the discussion in the Results section in the present manuscript creates confusion because it is too brief and not detailed enough (and see my comment below about the Discussion section itself). Just the very first paragraph of the Results provides several examples, but this occurs throughout the Results: on line 155, data “suggest... a highly reduced sediment matrix”; on line 157, data “are indicative of rapid removal...”; on line 159, data “may be related to high rates of carbonate dissolution...” Each of these statements needs more context and support in a combined R and D format, or should likely be removed--and again, there are many more such examples, this is just the first paragraph of the Results.”*

Response: We understand the reviewers' perspectives on strictly separating data presentation from literature contextualization and reduced the discussion part in the results section where possible, while retaining only minimal contextual references needed for interpretation of the data (e.g., page 12, line 11ff; page 13, lines 10-12 and 20-22; page 15 lines 12ff, page 16, 17 and 18).

Comment 5: *“The Discussion has several strange portions where data are presented, then the discussion uses extensive information from the literature to infer what the data mean, but the two are not connected. There is also a lack of clarity about exactly which data are relevant for the ensuing discussion, esp. 16S vs. meta'omic. The text from 355-365 is an example of these issues; see also text on 387-392 and 398-403, but there are other examples. I recommend that this type of text could be converted into text in the Introduction formulated as expectations/hypotheses that are then tested by the data.”*

Response: we have added a hypothesis to the introduction as suggested (page 5, lines 14ff). We have strengthened the connection between our results and the supporting literature by explicitly relating each interpretation to the corresponding findings from our dataset. Throughout the revised manuscript, each interpretation is now directly connected to the corresponding results and supporting (if available) literature (please compare to extensive changes made throughout Discussion). We have also added explicit figure references throughout the Discussion to guide the reader to the relevant dataset.

Comment 6: “...there are some language/punctuation issues that should be addressed.”

Response: The entire manuscript has undergone a rigorous copyediting pass to correct minor punctuation errors, fix formatting artifacts, and optimize sentence structures for maximum clarity.

Reviewer #3

Comment 1: “The abstract hints at a working hypothesis that is not explicitly stated in the final part of the introduction. I believe it is always important for a scientific study to explicitly state a hypothesis...”

Response: We completely agree that explicitly stating a guiding hypothesis strengthens the narrative framework of the study. Following the reviewer's recommendation, we have updated the final part of the Introduction. We now explicitly state our working hypothesis that “...persistent seasonal deoxygenation drives a shift toward sustained anaerobic microbial pathways and depletes reactive Fe-mineral inventories, which progressively reduced the sediment's capacity to buffer hydrogen sulfide and accelerates self-reinforcing feedbacks that reduce long-term sedimentary resilience.” (page 5, lines 14ff).

Comment 2: “In the introduction and throughout most of the article... there is no reference to meio- and macrofauna. It could be a valuable discussion point to include these groups of organisms in the general interpretation of the results.”

Response: We thank the reviewer for highlighting the ecological importance of macro- and meiofauna in benthic systems. We agree that these organisms play a vital role cf. bioturbation and bioirrigation. While both processes were mentioned in the previous version, we have now explicitly made a point of highlighting their role in the seasonal shift (e.g., page 5, line 6ff; page 23, line 6ff).

Comment 3: “Discussion vs. Results: In some sections (e.g., the paragraph starting on line 313), the discussion seems like a repetition of the results without citations...”

Response: We appreciate this critique and made a concerted effort to better integrate observations and interpretations in the Discussion

Comment 4: “Integration of Approaches... I would have expected to find at least one paragraph in the discussion where a discrepancy between a biogeochemical result and a molecular result was addressed. A small example is in lines 345-347...”

Response: We completely agree that scientific discovery and deeper ecosystem understanding often thrive on the misalignments between independent methodologies. These discrepancies highlight the inherent logistical and biological limitations of different methods, particularly the unique challenges of extracting, processing and capturing highly transient environmental RNA via metatranscriptomics

versus detecting stable, accumulated geochemical porewater profiles. We made a conscious scientific choice not to over-interpret or artificially force our distinct datasets into a perfectly synchronized narrative. Instead, we chose to present our empirical findings with absolute transparency exactly as they were captured and elaborate on these method's caveats in the supplementary material.

We have also included a section on the discrepancy between high Sox rates but lacking transcripts associated with sulfur oxidation in late October (see page 26, line 15ff). We have also elaborated in more detail on elevated methane concentrations in the lower water column vs. the absence of known methane oxidizers/functional genes encoding for methane oxidation (page 22 line 4-6).

Comment 5: *“Long-term Consequences... Are these results aligned with those from perennially anoxic ecosystems where oxygen has been absent for decades?... I felt the article lacked a concluding consideration projecting the consequences of sulfide upwelling on the diversity and functioning of microbial communities.”*

Response: We have significantly expanded our final concluding synthesis to contextualize our findings within long-term global ocean trends (page 29, line 21ff).

We believe these comprehensive updates fully address the reviewers' critiques while preserving the structural integrity and multidisciplinary scope of our study. Thank you again for your time and effort in evaluating our work.

Sincerely,

Mirjam Perner on behalf of all co-authors

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Dear COMMSERV Editorial Board,

September 15, 2026

Thank you for the opportunity to submit a final revision of our manuscript entitled "Persistent seasonal deoxygenation alters benthic metabolic processes and accelerates hypoxia feedbacks in the Baltic Sea".

We have carefully reviewed all editorial requirements and guidance for authors listed in the provided table and have implemented the requested changes throughout the manuscript. The table also provides a point-by-point overview of the changes made, with further details provided in the "Author comments" column.

The three reviewers had no further comments or concerns regarding the scientific content of the manuscript, and we are very grateful for their positive assessment.

As requested, we have submitted a marked-up version of the revised manuscript with all changes highlighted, as well as a clean version of the revised manuscript.

We thank you and the reviewers for the careful assessment of our manuscript and hope that the revised version is now suitable for publication.

Kind regards,
Stefanie Böhnke-Brandt
on behalf of Mirjam Perner

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Manu- script In- for- mation	Guidance for Authors	Author Comments
Title	Please note that manuscript titles should be 15 words or less, written in the English language, and should not contain punctuation (including colons and dashes), idioms, or puns (these do not translate well internationally).	The revised title is within the 15-word limit.
Title	Suggested new manuscript title: Persistent seasonal deoxygenation alters benthic metabolic processes and accelerates hypoxia feedbacks in the Baltic Sea	Yes, we have included the "in the Baltic Sea" as suggested.
Abstract	The abstract should be accessible to non-specialists and less than ~160 words long. It should include the background and context of the work, "Here we show" or an equivalent phrase, and then the major results and conclusions of the paper in the present tense. It must not contain references. Acronyms and element symbols should be avoided. In your abstract, please try to make the abstract more layman-friendly, including explaining phrases like eutrophication and hypoxia, for example.	Yes, we have revised the abstract to improve accessibility for non-specialists while retaining the scientific detail. We have clarified terms such as eutrophication and hypoxia, removed element symbols and acronyms, and revised the abstract to present the major results and conclusions in the present tense. The abstract is now 158 words and contains no references.
Main text	The paper should be structured as follows: An "Introduction" section that provides sufficient background and rationale for the work. The final paragraph of the Introduction should summarize the major findings and conclusions of the work. A "Results" section with up to two layers of sub-headings, the first of which should appear after the 'Results' heading. This should be followed by a 'Discussion' section, with no sub-headings. A combined "Results and Discussion" section could be used instead, with sub-headings. You may wish to include a "Conclusions" section	We have revised the manuscript structure to comply with the requested format. The Methods section has been moved to follow the Discussion. We would prefer to retain the sub-headings in the Discussion, as they provide important structure and help readers navigate the discussion, particularly given the length and complexity of the study. We kindly ask whether an exception to this formatting requirement would be possible.

	at the end, with heading. A “Methods” section after the Discussion or Conclusions. Sufficient detail should be given to allow the work to be reproduced. If the Methods are central to the study, this section can be placed in between the Introduction and the Results (or Results and Discussion) section. Section numbers are not supported by our formatting style. Please make sure not to reference sections by their number in the text (or the Supplementary Information).	
Main text	Please revise the manuscript to avoid novelty claims and exaggerated language when describing the results. In particular, we ask that you refrain from using words such as “new,” “novel,” “for the first time”, “unique” and phrases including “previously undiscovered”, “previously unknown”, “previously not shown”. Novelty should be made clear from the context. Additionally, please remove exaggerated or promotional descriptors such as “extremely,” “outstanding,” “exceptional”, “unprecedented”, “unparalleled”, “comprehensive”, and replace them with precise, evidence-based language.	We have revised the manuscript to remove novelty claims and exaggerated or promotional language. The only instance requiring revision was in the Materials and Methods section, where “entirely new phyla or lineages” has been replaced with more precise, neutral wording.
Display items	Tables larger than a single page should instead be provided either within the Supplementary Information PDF or in Excel (or a similar format) and be described as Supplementary Data.	No tables are included in the main manuscript. Table S1 (now Data S8) is provided in the Supplementary Information PDF, while Tables S2–S7 (now Data S1–S7) are provided as Supplementary Data in an Excel file with six worksheets.
Display items	For all graphs depicting a single point value (e.g., mean) with error bars, you must add individual data points or convert the graph to a box-plot or dot-plot. This applies to bar graphs and line graphs. You may wish to refer to this blog post about representing data distribution in	Figure 3 has been revised to include individual data points alongside the mean values and error bars.

	plots (particularly for small datasets). https://ecrlife420999811.wordpress.com/2018/07/10/beyond-bar-graphs-free-tools-and-resources-for-creating-more-transparent-figures-for-small-datasets/ . We strongly encourage the same for plots with multiple time courses depicted. See the Comms Bio Editorial (June 2019) for more details about this policy. Example plots are shown here: https://www.nature.com/articles/s42003-019-0489-0	
Display items	Using rainbow colour plots can be visually misleading (see links below). Please change the colour schemes used in your plots to perceptually uniform ones. https://www.nature.com/articles/s41467-020-19160-7 https://www.climate-lab-book.ac.uk/2014/end-of-the-rainbow/ https://root.cern.ch/rainbow-color-map	Figure 5 has been revised using the colour-blind-friendly Okabe-Ito palette to improve colour distinction and accessibility.
Data availability	At the time of publication, the minimal dataset necessary to interpret, replicate and build upon the methods or findings reported in the article should be available in a publicly accessible repository. The data should be uploaded to a repository before the manuscript is accepted. If this is not possible, for example, for legal or ethical concerns, this must be clearly stated in the Data Availability statement. The statement ‘data are available upon request’ or making data available solely within the paper or Supplementary Information is not acceptable, data should be placed in a publicly accessible repository.	The source data underlying the main figures are publicly available in permanent repositories, as detailed in the Data Availability statement. The only data not previously included were the iron extraction data, which have now been added to the previously listed PANGAEA dataset (https://doi.pangaea.de/10.1594/PANGAEA.987156).

	We distinguish model output from data; we encourage (but do not require) authors to deposit model output in a publicly accessible repository. All source data, i.e. the numerical results underlying the graphs and charts presented in the main figures must also be made available via a permanent repository (e.g., Figshare or Dryad). Please provide details on access in the Data Availability statement.	
Data	Authors should cite any datasets stored in external repositories that are mentioned within their manuscript. For previously published datasets, we ask authors to cite both the related research articles and the datasets themselves. For more information on how to cite datasets in submitted manuscripts, please see our submission guidelines.	Yes, all external datasets mentioned in the manuscript are appropriately cited and referenced. We have added citations to the two previously published PAN-GAEA datasets used in this study; the related research articles were already cited in the manuscript.
Geological samples	Details of any geological samples and/or any palaeontological specimens must be provided, including clear provenance information. Information regarding the requisite permission obtained must be placed in the methods section. Palaeontological and type specimens should be deposited in a recognised museum or collection.	Not applicable
Checklist	For papers reporting the development or use of Machine Learning, please complete the following checklist and upload it with the resubmission as a 'Related manuscript file': https://www.nature.com/documents/ML-reporting-summary.pdf	Not applicable
Code availability	If code is not fully open, where code is a key advance within the paper, please provide details here of your alternative approach to describing the code advance, to make this aspect of value to the research community.	Not applicable

Software	Please indicate the explicit version of the software used for any simulation or data analysis in the relevant point of the Methods section.	Yes, the software versions used for data analysis are already specified at the relevant points in the Methods section.
Competing Interest	If any author is also on the editorial board of this journal, please include the following text within the Competing Interests statement: XXX is an Editorial Board Member for Communications (journal name) but was not involved in the editorial review or peer review, nor in the decision to publish this article.	None of the authors is an Editorial Board Member of this journal.
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Supplementary Information	Supplementary Information must be provided as a single, separate file in PDF format. Other supplementary files such as Movies, Videos, Audio, and Data files may be provided separately. These should be clearly labelled as Supplementary Movie/Audio/Data 1, etc., and must be accompanied by legends in a separate file (not in the Supplementary Information file). Please rename the Supplementary tables as Supplementary data and cite them accordingly in the main article file.	The supplementary tables have been renamed from Table S1–S8 to Supplementary Data S1–S8 and the corresponding references in the main manuscript have been updated. The relocation of the Methods section also resulted in changes to the numbering of some supplementary items, which have been adjusted accordingly throughout the manuscript.

Supplementary Information	We notice that the captions for your Supplementary Figures/Schemes/Tables are included in your main manuscript file. Please remove these and place them directly into your Supplementary Information PDF.	done
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	is not acceptable, data should be in a publicly accessible repository. Making data available solely within the paper or Supplementary Information is not acceptable, data should be placed in a publicly accessible repository. The Data Availability statement should be placed after the Methods section and must include: * The data types used in the study and how they can be accessed * Accession codes for deposited data * Other unique identifiers (such as DOIs and hyperlinks for any other datasets) * If applicable, a statement explaining any restrictions on data access * If no data were used or generated in the study, the Data Availability statement should be as follows: 'Data sharing not applicable to this article as no datasets were generated or analysed during the current study.' Where community repositories are available, we require data sharing through such repositories. Where such repositories are not available, datasets may be hosted in general data repositories such as Figshare, Dryad or Zenodo. See this editorial for more details. http://www.nature.com/sdata/policies/repositories	
Statistics	Legend Information - Wherever statistics have been derived (e.g. error bars, box plots, statistical significance) the legend needs to provide and define the n number. The n number must be provided as a precise value (not a range), using the wording "n=X biologically independent	The Figure 3 legend has been revised to specify the sample size as n = 3 technical replicates, except for 25 February 2022 (1-2 cm), for which n = 6 for both the 0.5M and 6M extractions; 25 October 2026 (0-1 cm), for which n = 7 for the

	samples/animals/independent experiments” etc. as applicable. Please note that this information is missing in the legend of figure 3.	0.5M and 6M and 25 February 2022 (0-1cm) were n = 2.
Statistics	Please note that the error bars/error bands need to be defined in the legend of figure 3.	The Figure 3 legend has been revised to define the error bars and specify that they represent $\pm$ SD of technical replicates.