Answers to os-2019-113_SC1

Changes in the composition of marine and sea-ice diatoms derived from sedimentary ancient DNA of the eastern Fram Strait over the past 30,000 years

By Heike H. Zimmermann, Kathleen R. Stoof-Leichsenring, Stefan Kruse, Juliane Müller, Rüdiger Stein, Ralf Tiedemann, and Ullrike Herzschuh

Dear Henriette Kolling,

thank you very much for your comments and questions which helped to improve the quality of this manuscript. Please, find below your comments and questions in bold letters, while our answers are placed below and changes that will be made in the text are underlined. New references that will be included are placed at the end of this document. Line numbers are referring to changes made in the revised manuscript.

The term "richness" is used very often, and I have difficulties understanding that term. Could you please add some information on this term, as you use it very often.

We included the following sentence:

L149-150: "As a measure of alpha-diversity we calculated richness of (1) ASVs (number of amplicon sequence variants) and (2) unique taxonomic names (number of grouped ASVs that were assigned to the same taxonomic name)."

Abstract: The Abstract is missing some information on the investigated material/samples.

We included the information in the abstract.

L17: "By amplifying a short, partial *rbcL* marker <u>on sediment core MSM05/5-712-2</u>, […]"

Introduction: I have the feeling other sea ice proxies should be mentioned. Further, some information on the advantages of the sedaDNA study you present here is missing. It is not clear why sedaDNA may be of advantage for sea ice reconstructions, as the sea ice biomarker IP25 is not as prone to dissolution as microfossils.

We agree. We moved the paragraph further below after we introduced ancient DNA and have re-written it.

Chapter 6: I feel this chapter needs more detail. The mismatch of IP25 and sedaDNA was to be expected, as you do not investigate the IP25 producers. Could you elaborate more on the reasons for the mismatch of biomarker and sedaDNA record? What about seasonality of N. frigida and IP25 production? From SW Greenland, Krawzcyk et al (2015; Polar Biology) found that N. frigida is most abundant in late winter before the main spring bloom – which is expected to be the main production season of IP25 in Fram Strait. What about habitats (under the ice, inside the ice) between the different species? And finally what are your recommendations for future work or when using sedaDN for sea-ice reconstructions?

We have re-structured section 6 according to comments made by the other reviewers and discussed more about the ecology of *Nitzschia frigida* in this context.

At the end of our conclusions, we added the following recommendation for future work:

"Recommendations for future work with sedimentary ancient DNA in the context of sea ice reconstructions involve the preparation of reference genomes and a more targeted enrichment, for example of genes that help species to adapt to sea ice and allow them to cope with rapidly changing environmental conditions."

L36 Krawczyk et al., 2017 should be added here

We added the reference.

L59 & L60 overuse of whether

We exchanged the first occurrence with if.

L82 I find the description of the sea ice condition in the working area confusing.

We changes the sentence to:

"Today, the site is located south of the winter and summer sea-ice margin and is ice-free year round […]"

L82 mentioning past sea ice variability feels wrong here, maybe add this information with some more detail to the introduction

We moved the sentence to L64 and changed the sentence to:

"As previous work indicates variability in the past sea-ice cover (Falardeau et al., 2018; Müller et al., 2012; Müller and Stein, 2014; Werner et al., 2011, 2013), samples were chosen according to high, medium and low concentrations of the diatom produced sea-ice biomarker IP_{25} (Müller et al., 2012; Müller and Stein, 2014) and we expect associated changes in the taxonomic composition."

L86 should say Epp et al. (2019), L116 should say Callahan et al. (2017), L120 should say Dulias et al. (2017) and Stoof-Leichsenring et al. (2012)

We have changed them accordingly.

L134 I do not understand this Quote.

We changed the sentence to:

L143-144: "We resampled the dataset 100 times to the minimum number of sequences available (25,601 counts), then, for each replicate, we calculated the mean number of sequence counts for each ASV across the 100 resampling steps (code available at: https://github.com/StefanKruse/R_Rarefaction (Kruse, 2019))."

L150-159 I feel the information on lake studies has too much detail whereas the information on marine studies is too short as the presented study is marine.

Here we want to show, that only few studies exist that focus on ancient DNA from diatoms. As a couple of weeks ago a study about diatom and foraminiferal ancient DNA was published (Pawłowska et al., 2020) and we will add this to the list as well as a study about resurrection ecology.

We included the underlined statement, to make this clearer:

We used *sed*aDNA metabarcoding by applying the diatom-specific *rbcL_76* marker (Stoof-Leichsenring et al., 2012) which has already proved successful in low-productivity lakes of northern Siberia (Dulias et al., 2017; Stoof-Leichsenring et al., 2014, 2015), but so far has not been tested on marine sediments.

L198 This is a major problem. However cannot be changed for your study but I welcome this comment. For future studies the parallel investigation of biomarkers and diatoms in the microfossil and genetic record may be a very promising approach.

Yes, we agree.