

## ***Interactive comment on “Plankton-derived environmental DNA extracted from abyssal sediments preserves patterns of plankton macroecology” by Raphael Morard et al.***

### **Anonymous Referee #2**

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This paper explores the potential of using eDNA from sediments to infer plankton community structure. The authors analyze already available foraminifera sequence data to address an ecological relevant and timely question. The idea of using the aDNA preserved in sediments as an archive to explore planktonic biogeographic patterns is interesting; however, as the authors focus only on planktonic foraminifera, I do not think that the authors can conclude that the approach is valid for all planktonic taxa, and the title and some conclusions should then be accordingly revised. Overall, the manuscript is well written and the methodology description is detailed and precise, although there are some important concerns that should be clarified or revised before publication. Furthermore, the results do not so strongly support the main conclusion,

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as eDNA and fossil record only produce somewhat similar patterns. The only clear reproducible pattern is the separation between high and low latitude samples. A major issue relates to the large variation in the number of retained reads per sample after filtering (48 to 124,355). The authors conduct all the analyses without subsampling to the lowest number of reads, which undoubtedly bias community comparisons and diversity estimates. As some samples have an extremely low number of reads belonging to planktonic foraminifera, I suggest excluding those samples and reanalyze the dataset equalizing the number of reads per sample. In addition, the authors should clarify how they analyzed data from census counts of microfossils (details of sampling, number of individuals per sample, normalization, etc.) as they use these data to validate the use of eDNA from sediments to infer planktonic foraminifera community structure. Another suggestion is to exclude all the sequences belonging to small (<150 microns) foraminifera from the eDNA dataset, as the census counts are not including this fraction of the community.

#### Specific comments

Title. I suggest revising the title as the authors focus only on planktonic foraminifera. Page 2, lines 13-17. These final statements are too strong. Page 2, lines 26-27. Please explain why high concentrations of DNA in sediments indicates that part derives from planktonic/pelagic organisms. Page 3, lines 25-26 and Page 4, lines 1-2. The authors should keep in mind throughout the manuscript that they are focusing on a taxonomic group that seems to be particularly suited for validating their hypothesis and thus extrapolating to all planktonic or even the entire spectrum of pelagic organisms is not straightforward. Page 7, lines 10-11. Please, provide details on the census counts dataset. Page 7, line 18. Change “ascribed” to “ascribe”. Page 9, lines 21-23. I do not see that the eDNA dataset reproduce separation between Caribbean and Japan samples (only when using absolute numbers of reads due to several order of magnitude difference in number of reads). I do not find appropriate the analyses conducted with absolute read numbers considering the extreme differences among samples. Please

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consider excluding panels a-c in figure 4. Page 10, line 1. I do not see that the patterns are identical, please revise. Page 10, lines 12-13. It looks from data in figure 6 that the correlation is not significant. Moreover, many data are well above or below the 1:1 line. Please provide p-value. Page 11, lines 4-5. As already commented, the patterns are not so consistent. Page 12, line 5. I suggest changing “discover” to “detection”. Page 12, lines 5-7. Are these species present in the fossil record? Please, add a comment on that. Page 12, lines 18-25. If microperforate species are not represented in the fossil record, I suggest excluding these sequences from the eDNA archive in order to compare both datasets. Page 13, line 25. I suggest smoothing this statement.

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