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Supplementary Materials for

Patterns of eukaryotic diversity from the surface to the deep-ocean sediment

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The PDF file includes:

Figs. S1 to S13 Tables S1 and S4 Legends for tables S2, S3, S5 to S8 References

Other Supplementary Material for this manuscript includes the following:

Tables S2, S3, S5 to S8

mean %Reads

Figure S1: Normalized richness and distribution of eukaryotic ASVs within and across oceanic realms (pelagic euphotic, pelagic aphotic and sediment). Each realm dataset was aggregated into single vector prior to subsampling 1000 times at a depth of one million reads. The top panel barplot shows the mean proportion of ASVs and reads abundance within and across realms (standard deviation across 1000 draws is indicated on each bar). The left panel shows the mean normalized overall ASV richness (and standard deviations) for each realm. The bottom pie charts indicate the average contribution of each realm in number of reads to realm intersections.

Figure S2: Effect of size fractionation of pelagic samples on the alpha- and beta-diversity. Left panel: ASVs accumulation curves as a function of sampling effort. Pelagic aphotic samples are in dark blue and pelagic euphotic are in light blue, and size fractions are indicated at the tips of the curves. The nano fraction of the aphotic samples is particularly richer than any other size fraction. Right panel: Non-metric multidimensional scaling ordination of the pelagic samples. The ordination was performed on a Bray-Curtis dissimilarity matrix computed from normalized reads counts using the cumulative sum scaling method. The stress value is indicated on the plot. The size fractions are clustering separately on the ordination.

Figure S3: Structure of eukaryotic communities across realms (pelagic euphotic, pelagic aphotic with only nano and pico fractions, and deep sediment) and as a function of geographic basin. The ordination is a Non-metric multidimensional scaling of the Bray-Curtis dissimilarity matrix computed from normalized reads counts using the cumulative sum scaling method. The red and black lines on the ordination represent, respectively, the absolute latitude and depth as fitted surfaces to the ordination.

Figure S4: Taxonomic composition (ASVs abundance) of eukaryotic groups in the pelagic euphotic, pelagic aphotic, and deep-ocean sediment realms (see Table S3 for details).

Figure S5: Taxonomic composition (ASVs richness) between the pelagic and the benthic communities (euphotic and aphotic are grouped and unassigned ASVs discarded).

Figure S6: Number of Operational Taxonomic Units (OTUs) formed from unassigned eukaryotic ASVs as a function of similarity cutoff. The number of ASVs before clustering is indicated on the left side of the plot.

Figure S7: Cumulative proportion of ASVs and reads abundance as a function of similarity with best hit with a reference sequence in PR² for selected benthic taxonomic groups.

Figure S8: Benthic alpha diversity variation along gradients of latitudes, primary productivity, POC export from the surface and POC reaching the seafloor. In red is the normalized richness obtained by rarefying the ASV table and in green is the Shannon diversity. Blues lines are fitted generalized additive models (s=3) and shades are 95% confidence intervals. The explained deviance for each combination is indicated on the top of the plots as well as the significance of the relationship (ns: non-significant, *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$).

Figure S9: Distance decay of Sørensen similarities (log transformed) as a function of distance separation between samples of the pelagic euphotic, pelagic aphotic and benthic communities. Similarity matrices were averages over ten rarefactions draws of community matrices.

Figure S10: Neutral community assembly models fitting pelagic euphotic, pelagic aphotic and benthic eukaryotic communities. Datasets have been aggregated at the station level prior fitting. Goodness of fit (R^2) are indicated in the titles of the plots. Insets within main plots represent the distribution of ASVs across the more geographically widespread category (in red) the neutral category (in blue) and the less geographically widespread category (in green) than expected by the model. The bottom panel below main plots display the distribution of ASVs across category for selected taxonomic groups.

Figure S11: Correlation of relative abundance profiles (log transformed) between the sinking pelagic ASVs from the euphotic or aphotic realms and their respective abundance profile in the deep-ocean sediment (in orange). In blue is the correlation between euphotic and aphotic relative abundance profiles of pelagic ASVs also detected in the pelagic aphotic zone. The titles above each plot indicate the number of sinking ASVs compared to the number of ASVs of a given taxonomic group in the 'source realm' and the relative abundance within this source realm those sinking ASVs represent. For instance, 204 ASVs of diatoms out of the 2665 detected in the

Euphotic zone are detected in the sediment and these 204 ASVs represent 80.2% of diatom reads in the euphotic zone. Adjusted R^2 and significance of linear models are reported (ns: nonsignificant, *: $p < 0.05$, **: $p < 0.01$, ***: $p < 0.001$).

Figure S12: Comparative analysis of sediment DNA and RNA datasets obtained from the deep_sea project samples. A) Number of ASVs and their relative abundances detected in both DNA and RNA libraries and only in DNA or RNA. A total of 89.82% of the sediment reads

represent ASVs present in both DNA and RNA V9 libraries, even though numerous rare ASVs are present either only in DNA or RNA. B) Proportion of sediment reads in DNA and RNA libraries as a function of their source (benthic vs planktonic). The proportion of plankton reads is higher in the DNA (22.7%) compared to RNA (14.9%). C) Relative abundances of taxonomic groups within the sinking plankton and benthic fractions in both DNA and RNA sediment libraries. It shows variations between DNA and RNA, mostly for the plankton fraction in the sediment (e.g. diatoms & dinoflagellates are more abundant in DNA than in RNA, whereas fungi and kinetoplastids are more abundant in the RNA. For the benthic fraction, the relative abundances of groups are much more consistent between DNA and RNA, with the notable exception of amoebae that are more abundant in the RNA. D) PCoA ordination of DNA and RNA samples as a function of geographical origin (using a Bray-Curtis dissimilarity matrix computed from normalized reads counts using the cumulative sum scaling method). Above the ordination are indicated the results of a permutational analysis of variance model, testing for the effects of biogeography and type of molecule (and their interaction) on the structure of sediment communities. The ordination shows that the biogeographic pattern is mostly mirrored between DNA and RNA, and although the interaction term between biogeography and molecule (DNA/RNA) is significant, it explains only 2.9% of the variation. Taken together, these results indicate that the diversity and the biogeographic signals are mostly similar between DNA and RNA.

Figure S13: Length distribution of the ASVs. ASVs are colored after their taxonomic annotation on the SILVA v138 database. We removed prokaryotes, non-18S ASVs and any unassigned ASVs shorter than 116 bp long, because they may represent unassigned prokaryotes ASVs. For downstream analysis, we also kept only ASVs that match an eukaryotic reference sequence in PR2 with at least 20% similarity and that contain the 'GTCG' sequence motif in the first four nucleotides in the 5' end (see methods).

Table S1. Deep-ocean sediment dataset. Material origin, number of stations and biological samples

Table S2: Summary statistics of the 18S V9 sequencing data processing. #raw_reads refers to the number of reads in raw files. #good reads refers to the number of reads that passed quality filtering. #analysed reads refers to the number of eukaryotic reads analysed in this study. References of the published datasets analysed in this study are reported

See the file 'Supp_Tables_S2_S3_S5_S6_S7_S8.xlsx'

Table S3: Aggregated eukaryotic richness and abundance in the pelagic euphotic, pelagic aphotic, and sediment biomes (also split into benthic and sinking plankton fractions only)

See the file 'Supp_Tables_S2_S3_S5_S6_S7_S8.xlsx'

Table S4: Explained compositional variance of benthic communities by a selection of environmental variables (stepwise dbRDA model building using the *ordiR2step* function of the vegan R package)

Table S5: Mantel correlation between eukaryotic communities dissimilarity matrices and geographic distance. Distance decay parameters, initial similarity within 1 km distance, slope of linear models and halving distances, i.e. the distance after which the initial similarity is halved (95% confidence intervals values are indicated between brackets)

See the file 'Supp_Tables_S2_S3_S5_S6_S7_S8.xlsx'

Table S6: Pelagic ASVs detected in sediments that are associated with POC export from the surface or POC seafloor, based on a sparse partial least square (sPLS) regression. Their taxonomic and functional annotation is reported.

See the file 'Supp_Tables_S2_S3_S5_S6_S7_S8.xlsx'

Table S7: Full metadata table

See the file 'Supp_Tables_S2_S3_S5_S6_S7_S8.xlsx'

Table S8: Manual curation of the metazoan fraction of the pelagic ASVs sinking to the deepocean sediment

See the file 'Supp_Tables_S2_S3_S5_S6_S7_S8.xlsx'

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