

## Eukaryotic diversity in late Pleistocene marine sediments around a shallow methane hydrate deposit in Japan Sea

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Eukaryotic DNA has been detected from detrital organisms such as planktonic remains, spores, cysts, and pollen from marine and lacustrine sediments, associated with the intense upwelling and the high primary productivity. However, a large proportion of deposited eukaryotic DNA is aerobically biodegraded in shallow marine sediments. Cold seep sediments are often anaerobic near the sediment–water interface, so eukaryotic DNA in such sediments is expected to be preserved. As the abundance and biodiversity of eukaryotic DNA in deeply buried marine sediments are largely unknown, we investigated marine sediments at depths up to 31.0 meters below the seafloor (mbsf; 97 ka) around a shallow methane hydrate deposit in the eastern Japan Sea. Quantitative PCR analysis revealed the reproducible recovery of eukaryotic DNA in deep marine sediments in the vicinity of the methane hydrate deposit. Pyrosequencing of an 18S rRNA gene variable region generated total 9,366 reads from 5 samples, which was sufficient to cover the biodiversity based on rarefaction curves. Phylogenetic analysis revealed that most of the eukaryotic DNA originated from radiolarian genera of the class Chaunacanthida, which have SrSO<sub>4</sub> skeletons, the seagrass genus *Zostera*, and the seaweed genus *Sargassum*. Eukaryotic DNA originating from planktonic fauna and land plants were also detected. Diatom sequences closely related to *Thalassiosira* spp., known as indicator species for cold climates, were obtained from sediments deposited during the last glacial period (MIS-2). Land plant sequences of the genera *Alnus* and *Ulmus* were found in sediments deposited during the warm interstadial period (MIS-3). These results suggest the long-term persistence of eukaryotic DNA from terrestrial and aquatic sources in marine sediments associated with cold seeps, and that the genetic information from eukaryotic DNA from deeply buried marine sediments associated with cold seeps can be used to reconstruct environments and ecosystems from the past.

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