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Archaeal community composition based on tetraether lipids and 16S rRNA in Holocene methane pockmarks of the Baltic Sea

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Pockmarks are concave depressions in seabed and lakes, found worldwide. These geomorphological structures, ranging in size from tenths to hundreds of metres, are often indicators of hydraulic seabed activity. Their formation begins with a pressure build-up in the pores of fine-grained surface sediments with low permeability. Hence, the location of pockmarks is often associated with shallow gas systems, sometimes accompanied by submarine groundwater discharge (SGD), as seen in the south-eastern Baltic Sea. Consequently, the wide variety of pockmarks provides diverse habitats for microbial niches.

This study concentrates on the Holocene methane-bearing sediments of the Gulf of Gdańsk. The investigation of archaeal presence and the composition of the glycerol dialkyl glycerol tetraethers (GDGTs) has not been conducted here until now.

Core sediments (95 cm thick) were obtained in October 2019. The archaeal community was investigated by analysing free core lipids using ultra-high-pressure liquid chromatography coupled with high-resolution mass spectrometry (HPLC-MS) and 16S rRNA metagenomic sequencing. Cores retrieved from the methane-bearing sediments and the reference sites (i.e. outside methane-bearing structures) are characterised by very high TOC content (on average, 13 and 9 mmol/g, respectively) and contain all targeted iso- and OH-GDGTs (GDGT-0, -1, -2, -3, crenarchaeol, crenarchaeol isomer, OH-0, -1, -2). Methane-bearing sediments exhibit higher concentrations of GDGTs than non-gassy sediments, but crenarchaeol remains dominant in both, followed by GDGT-0. OH-GDGTs display similar trends, with OH-GDGT-0 being the main component. The archaeal community composition varies significantly between non-gassy and methane-charged sediments, with the latter showing higher absolute abundance. Nanoarchaeia dominate in both. Methanogenic archaea, (e.g. *Methanomicrobium*, *Methanosarcinia*), along with Thermococci and Altiarchaeia, were prevalent in methane-rich sediments, whereas non-gassy sediments exhibit a more diverse structure, including a higher abundance of Thermoplasmata, Bathyarchaeia, the Deep Sea Euryarchaeotic Group, Lokiarchaeia, and Archaeoglobi. These results open up an interesting discussion on the complexity of the studied environment and the application of GDGT clustering in relation to Archaea.