

MICROBIAL TAXONOMIC AND FUNCTIONAL DIVERSITY IN DEEP SUBSURFACE METHANE HYDRATE BEARING SEDIMENTS. C. B. Kretz¹, B. K. Reese², P. R. Girguis³, F. J. Stewart⁴, and J. B. Glass^{1,4*}

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Methane is a product of and substrate for microbial metabolisms in the deep subsurface, an important analog for potentially habitable extraterrestrial targets, but relatively little is known about microbial metabolisms in deep methane hydrate-bearing sediments. In this study, we analyzed microbial community gene diversity relative to geochemical gradients in subsurface sediments beneath Hydrate Ridge, offshore Oregon (ODP Leg 204 Site 1244). We targeted four geochemically distinct depths: near surface (2 mbsf), sulfate-methane transition zone (4 and 8 mbsf), iron-manganese reduction zone (18 and 20 mbsf) and deep subsurface (35 and 68 mbsf). DNA yields ranged from 15 ng/g sediment at 2 mbsf to 1 ng/g sediment at 68 mbsf.

Taxonomic analysis of the microbial 16S rRNA gene revealed large depth-specific differences in community composition (Figure 1). Near-surface samples were dominated by Archaea (57% total sequences), while deeper sediments were dominated by Bacteria (51-90% total sequences). In the near-surface, archaeal sequences were dominated by Marine Benthic Group B (MBGB; 22-29%) and Miscellaneous Crenarchaeotal Group (MCG or Bathyarchaeota; 20%). In contrast, sequences from deeper zones were dominated by OP9 candidate phylum JS1 or Atribacteria (60-87%). The microbial community in the iron-manganese reduction zone was significantly different, with a resurgence of uncultivated archaeal groups (10% South African Gold Mine Euryarchaeotic Group and 16% MBGB).

Metagenomic sequencing yielded 336 million reads from genomic DNA subjected to single cell multiple displacement amplification followed by Illumina HiSeq sequencing. The taxonomic affiliations of metagenomic sequences corroborated the trend of increasing OP9 candidate phylum JS1 or Atribacteria genes with depth. Dominant archaeal gene sequences were associated with uncultured archaea in the iron-manganese reduction zone, whereas Euryarchaeota in the class Methanomicrobia dominated at other depths. According to KEGG pathway analysis, 60-75% of genes of energy metabolism were involved in methane metabolism, with the most abundant being acetyl-CoA decarbonylase/synthase complex (*cdhB*), followed by di/trimethylamine (*dmd*, *tmd*), formaldehyde (*fdhA*) and formate (*fdoH*) dehydrogenases. Investigation of functional gene diversity in deep subsurface sediments is ongoing.

Figure 1. Geochemical data for dissolved methane, sulfate, iron and manganese in sediment porewaters (left) and 16S rRNA taxonomic composition (right) at ODP Site 1244, Hydrate Ridge from ODP Leg 204.

