

Signatures of anaerobic methanotrophic communities in gas hydrate-bearing sediments of the Chukchi Sea



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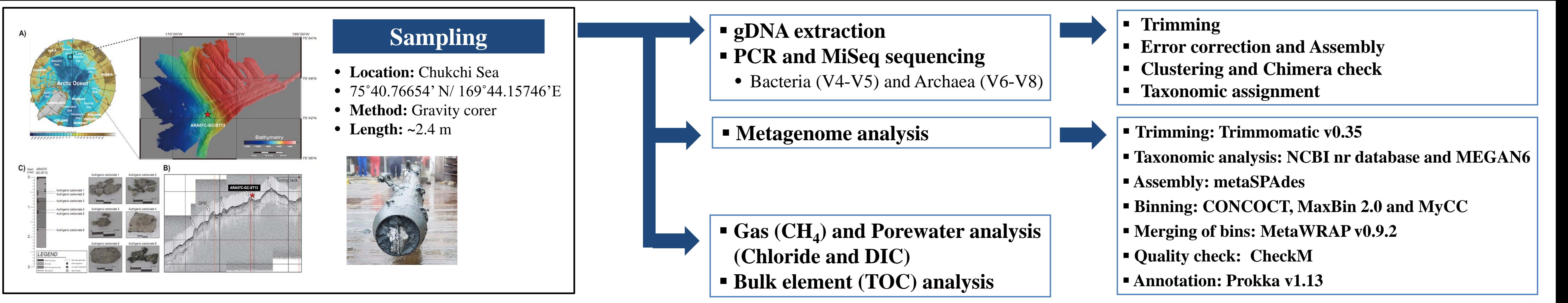
Anaerobic oxidation of methane with sulfate-reduction by anaerobic methanotrophic archaea (ANME) and sulfate-reducing bacteria (SRB) is an important process in the methane cycle of marine environments, which is regarded as a major sink of the potential greenhouse gas. However, little is known about metabolic relationship between ANMEs and their sulfate-reducing partners due to the lack of culture representatives. We investigated the microbial communities of gas hydrate-bearing sediments of the Chukchi basin along the depth in combination with geochemical analysis. ANME-1 and sulfate-reducing Deltaproteobacteria were predominant accounting for 97.3% of archaeal communities and 44.3% of bacterial communities, respectively at sulfate-methane transition zone (~120 cm below seafloor). To link metabolic functions with specific microbial populations, genome-centric metagenomic analysis was performed. The assembled metagenomic sequence data generated 1.52 Mb and 2.22 Mb genomes of ANME-1 and sulfate-reducing Deltaproteobacteria. The completeness of ANME-1 and sulfate-reducing Deltaproteobacteria analyzed by single copy marker gene were 80.1% and 86.2%, respectively. ANME-1 genome contains genes for key metabolic enzymes of reverse methanogenesis pathway (except N⁵,N¹⁰-methylene tetrahydromethanopterin reductase) and sulfate-reducing Deltaproteobacteria possesses a complete set of genes for dissimilatory sulfate reduction pathway. We will further investigate the syntrophy and microbial consortia formation to confirm the metabolic coupling between ANME1 and SRB in controlling methane removal.

Abstract

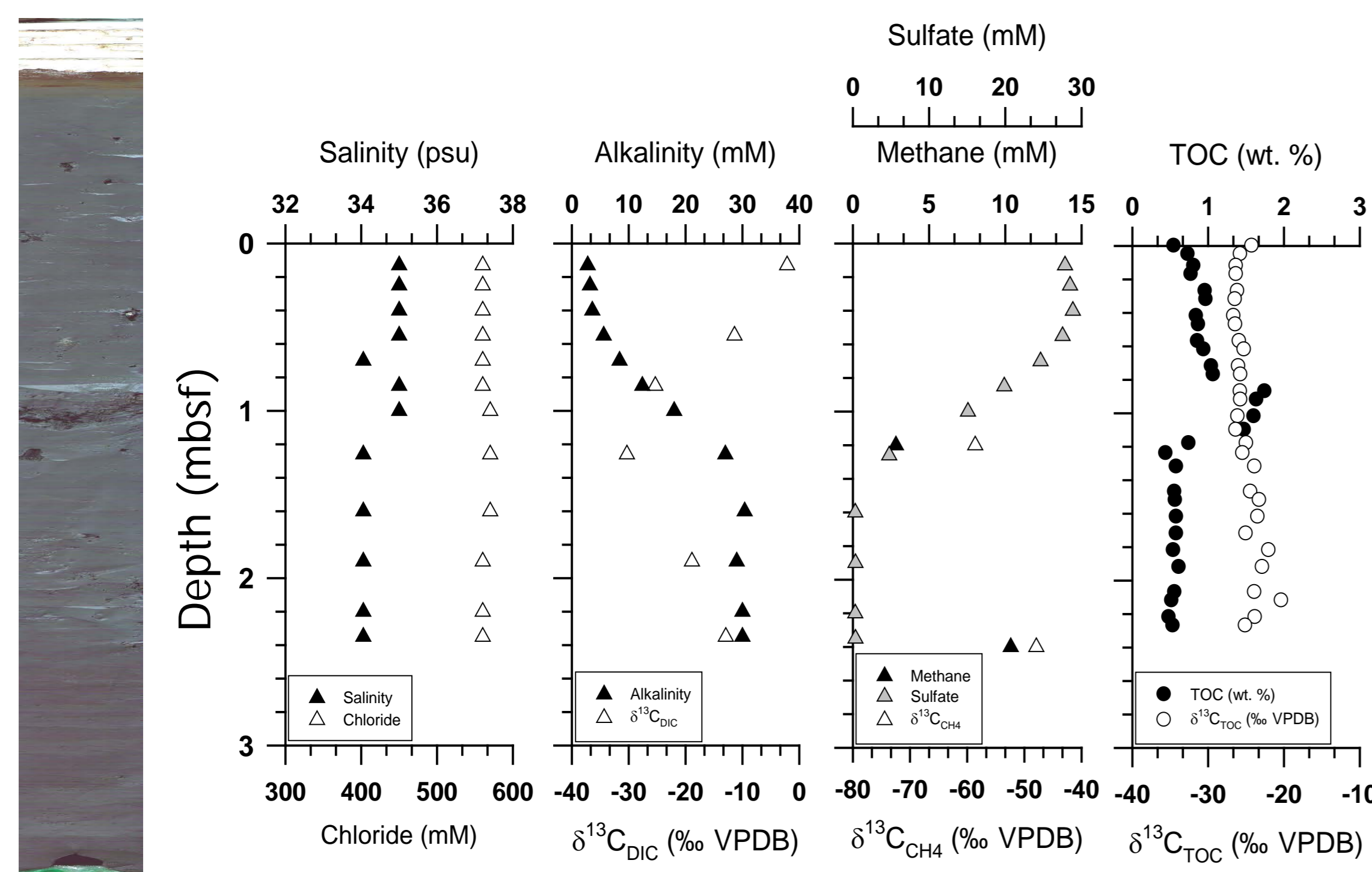
Materials & Methods

Results

Conclusion



1. Profiling of environmental factors



Depth profiles of gas (methane), porewater (chloride, alkalinity, and dissolved inorganic carbon), and bulk element (total organic carbon)

- Sulfate – Methane Transition Zone (SMTZ) was detected at 118 cmbsf.
- The isotopic values of methane (- 58.6‰ at 1.18 m and - 47.9‰ at 2.4 m) indicated the source of methane is thermogenic.

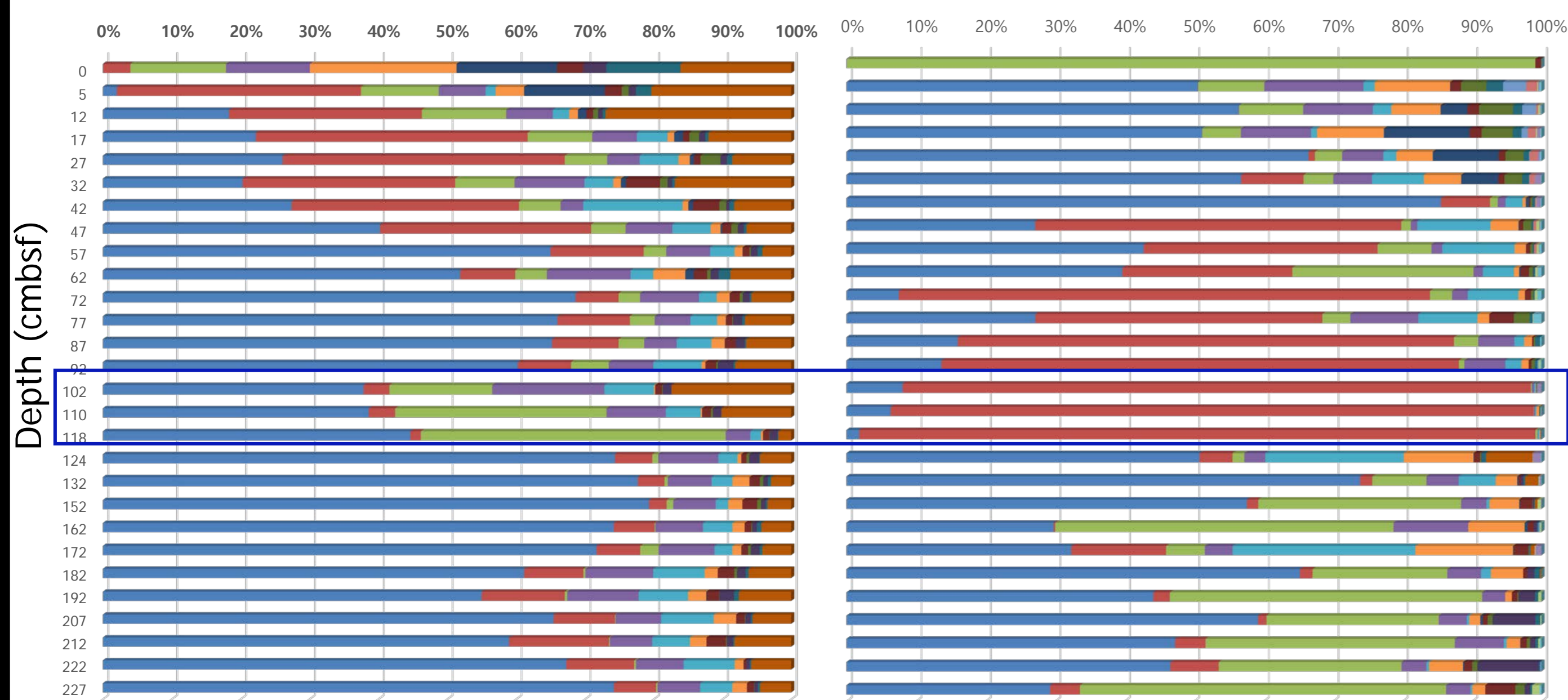
2. Microbial composition

Bacterial communities

Archaeal communities

Relative abundance

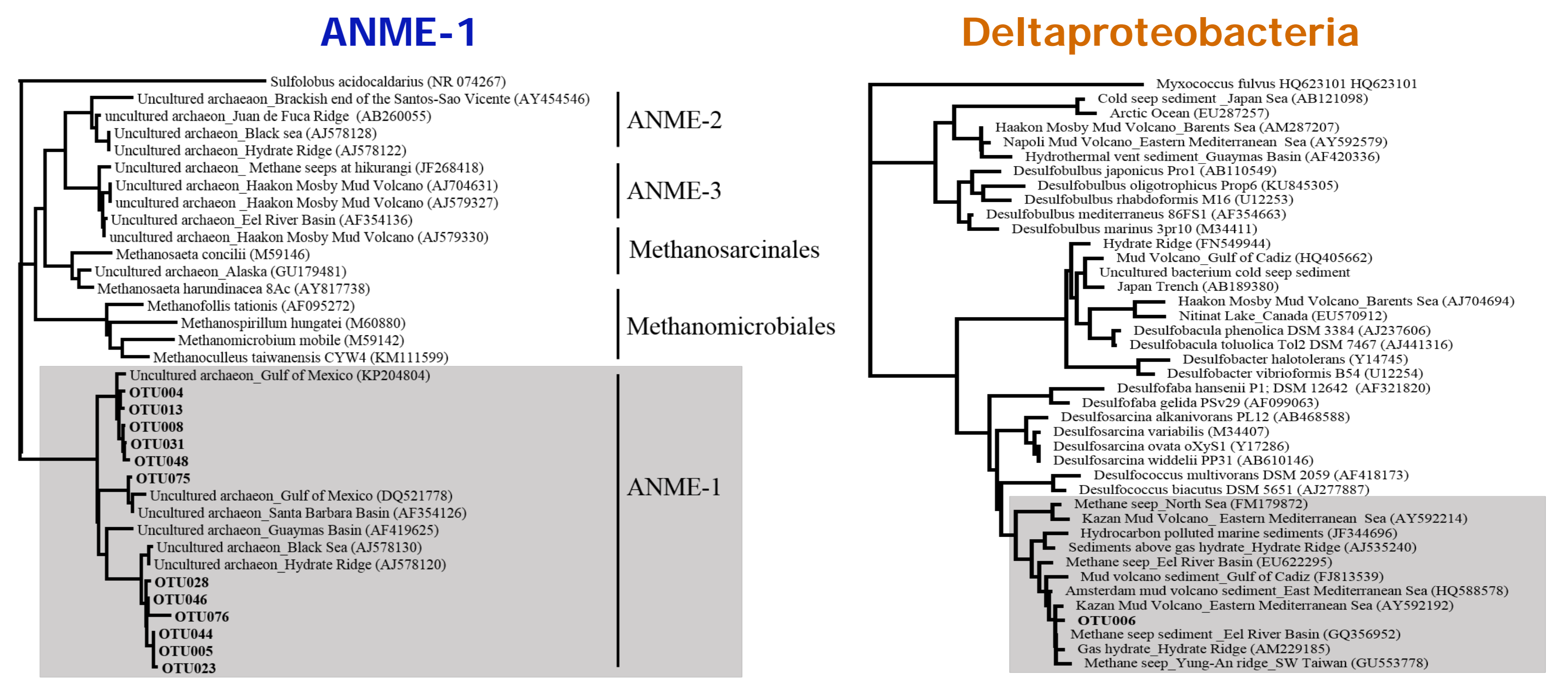
Relative abundance



- JS1
- Planctomycetes
- Acidobacteria
- Actinobacteria
- Chloroflexi
- Aminicenantes_OP8
- Bacteroidetes
- Alphaproteobacteria
- Deltaproteobacteria
- AY835409_o
- Gammaaproteobacteria_AF424768_o
- Methanomassiliococcales
- EF446193_o
- DQ841218_o
- AB019718_o
- ANME-1_o
- Methanosarcinales
- Others
- Archaea_unclassified
- DQ522931_o
- DQ522934_o
- Cenarchaeales
- Group3_o
- Thermoplasmata_unclassified
- AY835425_o
- DQ522931_o
- AJ578125_o

Relative abundance of bacterial taxa (at phylum level and class level of Proteobacteria) and archaeal taxa (at class level) along the core depths.

3. Phylogenetic analysis of major OTUs and their abundance

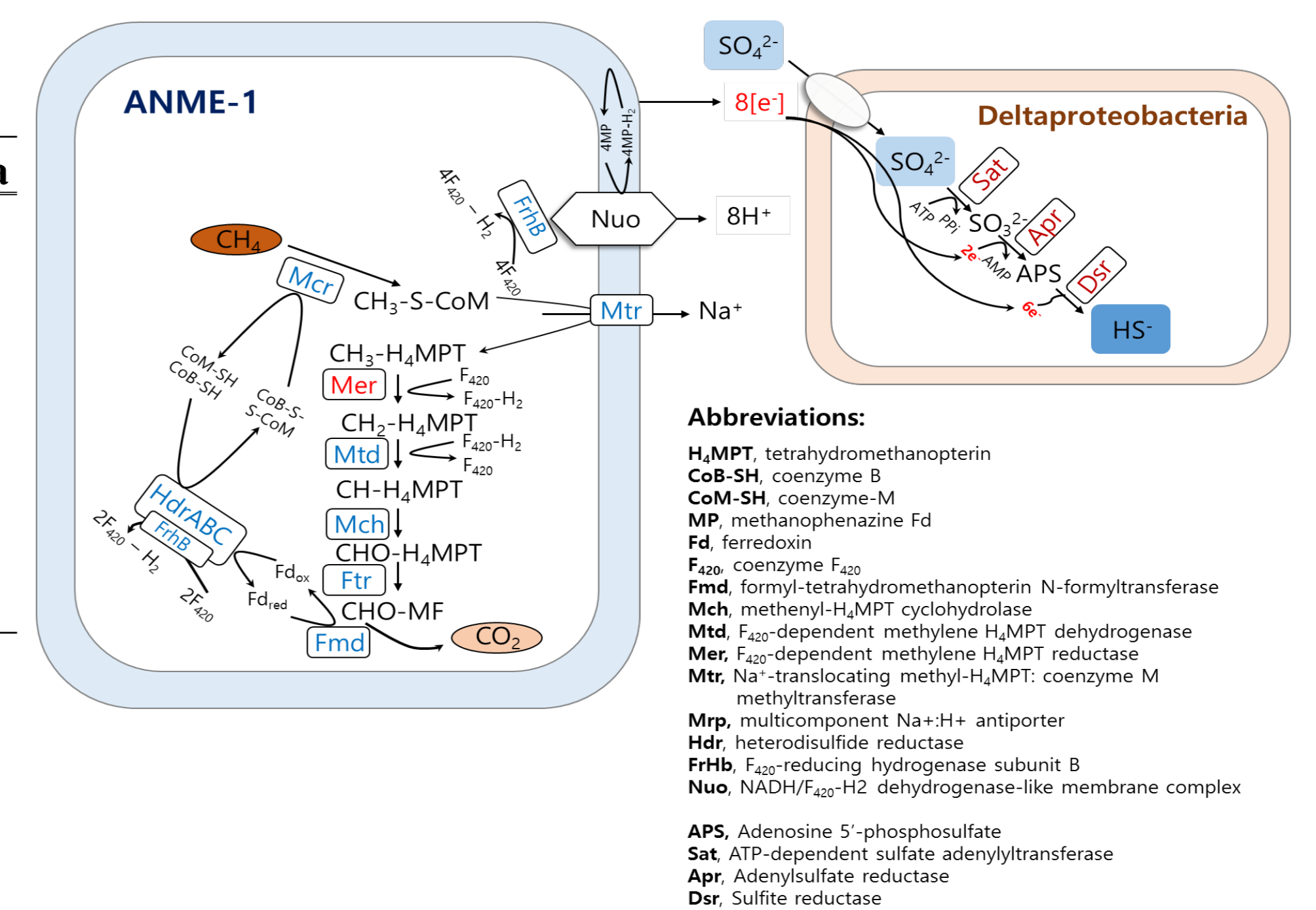


Heat map of major OTUs (>3%) of ANME-1 and Deltaproteobacteria

Domain	Class	OTU_ID	Depth (cmbsf)																													
			0	5	12	17	27	32	42	47	57	62	72	77	87	92	102	110	118	124	132	152	162	172	182	192	207	212	222	227		
Archaea	ANME-1	OTU004	0.0	0.0	0.0	0.0	0.0	0.3	2.8	1.9	0.8	0.6	3.3	5.0	10.5	24.5	22.9	0.0	0.0	0.0	0.0	0.1	0.2	0.0	0.0	0.2	0.2	0.0	0.0	0.0		
		OTU005	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	13.3	7.8	5.9	22.7	4.8	1.7	2.1	31.7	30.1	35.9	0.6	0.2	0.7	0.0	3.9	0.2	0.3	0.0	0.5	0.0	0.0
		OTU008	0.0	0.0	0.0	0.0	0.1	1.2	0.9	0.4	0.4	1.1	2.4	5.6	11.4	13.9	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.3	
		OTU013	0.0	0.0	0.0	0.0	0.1	0.8	0.5	0.2	0.2	0.7	1.1	3.2	7.9	8.9	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.0	0.0	
		OTU023	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.7	0.7	0.8	1.3	0.6	0.1	0.5	5.1	4.1	16.3	0.2	0.1	0.3	0.0	2.6	0.0	0.9	0.0	0.2	0.0	0.0	
		OTU028	0.0	0.0	0.0	0.0	0.0	0.0	0.0	2.9	1.8	1.0	4.5	1.2	0.4	0.4	6.7	6.7	6.2	0.2	0.0	0.1	1.0	0.0	0.2	0.0	0.2	0.0	0.0	0.0	0.0	
		OTU031	0.0	0.0	0.0	0.0	0.0	0.1	0.1	0.1	0.0	0.2	0.3	0.9	1.8	2.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.3	
		OTU044	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.1	0.6	0.6	2.1	0.4	0.2	0.3	3.5	2.7	5.8	0.0	0.1	0.0	0.0	0.7	0.7	0.0	0.0	0.0	0.1	0.0	0.0	
		OTU046	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.7	0.4	0.6	1.2	0.2	0.0	0.2	3.0	2.3	6.8	0.2	0.1	0.1	0.0	1.1	0.0	0.2	0.0	0.2	0.2	0.0	0.0	
		OTU048	0.0	0.0	0.0	0.0	0.2	0.1	0.1	0.1	0.2	0.4	1.0	1.9	1.9	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	
		OTU075	0.0	0.0	0.0	0.0	0.0	0.4	0.5	3.2	2.3	0.1	0.6	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
		OTU076	0.0	0.0	0.0	0.0	0.0	0.0	2.8	1.6	1.7	4.0	0.6	0.3	0.3	1.5	2.8	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.0	0.0	0.0	0.1	0.2	0.3	0.0	
		Bacteria	Deltaproteobacteria	OTU006	0.0	0.0	0.1	0.2	0.1	0.1	0.1	0.2	0.5	0.2	0.1	0.1	0.1	1.1	8.6	26.1	43.3	0.4	0.2	0.3	0.0	2.3	0.0	0.0	0.0	0.0	0.0	0.0

4. Genomic features of methanotrophic groups

Attribute	ANME-1	Deltaproteobacteria
Genome size (bp)	3,669,952	2,220,637
% GC content	42.3	42.2
contigs	510	358
N50	11,811	10,196
Total genes	3,795	2,070
Protein coding genes	3,741	2,023
RNA genes	54	47



- ¹³C-depleted methane at SMTZ (~118 cm) is proposed to be produced by the anaerobic methane oxidation.
- Genomes of ANME-1 and Deltaproteobacteria assembled from metagenome sequences of SMTZ depth with high portion of those methanotrophic groups suggest that methane is oxidized anaerobically by coupling of sulfate-reducing bacteria.