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E-mail:hyxbe@263.net

Bacterial and archaeal community structures in the Arctic deep-sea sediment

LI Yan[1, 3], LIU Qun[2], LI Chaolun[1], DONG Yi[1], ZHANG Wenyan[1], ZHANG Wuchang[1], XIAO Tian[1]*

[1] Key Laboratory of Marine Ecology and Environmental Sciences, Institute of Oceanology, Chinese Academy of Sciences, Qingdao 266071, China

[2] State Field Research Station of Jiaozhou Bay Marine Ecology, Institute of Oceanology, Chinese Academy of Sciences, Qingdao 266071, China

[3] Center for Transport Phenomena, Energy Research Institute, Shandong Academy of Sciences, Jinan 250014, China

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Abstract

Microbial community structures in the Arctic deep-sea sedimentary ecosystem are determined by organic matter input, energy availability, and other environmental factors. However, global warming and earlier ice-cover melting are affecting the microbial diversity. To characterize the Arctic deep-sea sediment microbial diversity and its relationship with environmental factors, we applied Roche 454 sequencing of 16S rDNA amplicons from Arctic deep-sea sediment sample. Both bacterial and archaeal communities' richness, compositions and structures as well as taxonomic and phylogenetic affiliations of identified clades were characterized. Phylotypes relating to sulfur reduction and chemoorganotrophic lifestyle are major groups in the bacterial groups; while the archaeal community is dominated by phylotypes most closely related to the ammonia-oxidizing Thaumarchaeota (96.66%) and methanogenic Euryarchaeota (3.21%). This study describes the microbial diversity in the Arctic deep marine sediment (>3 500 m) near the North Pole and would lay foundation for future functional analysis on microbial metabolic processes and pathways predictions in similar environments.

Key words: Arctic, deep-sea sediment, microbial community structure, pyrosequencing

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1 Introduction

The Arctic deep-sea sediments represent extreme environments characterized by permanently low temperature, organiccarbon limitation, and increasing hydrostatic pressure with depth. Microorganisms in such environments are surviving the rough conditions (Jannasch and Taylor, 1984; Deming, 1986). However, extensive shifts in terms of warmed surface waters, shrinking sea ice coverage and modified freezing-melting cycles of Arctic ice are occurring in the Arctic ecosystem due to global warming. Research on how deep-sea sedimentary microbial community structures respond to global warming is pivotal to the understanding of Arctic ecosystem's future.

Geographically, the Arctic Ocean is basins surrounded by continental shelf seas (Chukchi, Beaufort, Laptev, etc.). The major source of organic matter are river-derived / terrestrial – originated nutrient particles (Stein and MacDonald, 2004), while *in situ* primary and secondary production are comparatively at low levels (Wheeler et al., 1996). The input and availability of organic matter are the shaping forces in the sedimentary microbial communities of deep-sea ecosystem (Gooday et al., 1990). The global warming had resulted in earlier spring phytoplankton blooms (Hinzman et al., 2005), increased under-ice primary production (Boetius et al., 2013) and nutrient input coming into the Arctic Ocean (Peterson et al., 2002). Such changes are affecting nutrient particles sedimentation and the sedimentary microbial communities (Kirchman et al., 2009).

Investigations on microbial population diversity, richness, and composition of Arctic sediments have discovered distinct features in community composition, taxonomic diversity, and metabolic complexity (Ravenschlag et al., 1999). Bacteria diversity analyses have been stressing on psychrophilic sulfate-reducing bacteria, specific as well as ubiquitous microbial lineages were targeted and quantified (Ravenschlag et al., 1999; Sahm et al., 1999; Ravenschlag et al., 2001). Microbial diversity from coastal area sediments (Kongsfjorden, Svalbard) (Tian et al., 2009), multiple sites of northern Bering Sea (Zeng et al., 2011) and regional scale distribution at the Arctic Long-Term Ecological Research (LTER) site HAUSGARTEN (Eastern Fram Strait) (Jacob et al., 2013) were examined by either PCR-denaturing gradient gel electrophoresis (PCR-DGGE) or rRNA gene clone library. Other studies tried to resolve the microbial community composition with respect to extra-cellular enzymes activity to determine the relationship between community structure and potential function (Arnosti, 2008; Forschner et al., 2009; Teske et al., 2011). These

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* Corresponding author, E-mail: txiao@qdio.ac.cn

investigations have provided us with insights into the microbial assemblage of Arctic marine sediment. However, the microbial community structure of deep sea sediment with water depth more than 3 500 m and close to the North Pole is still unknown.

In 2010, during the 4th Chinese National Arctic Research Expedition, the ice cover thawed at summer time, so our cruise reached area near the Arctic Pole. The objectives of the current work were to characterize microbial community composition, including both bacterial and archaeal, of Arctic deep marine sediment by Roche 454 pyrosequencing, and try to link microbial diversity with environment factors by examining the sediment composition. A more specific goal is to document the baseline composition of benthic microbial ecosystem at the onset of polar ice sheet melting in the high Arctic, which would freshen our understanding of the microbial life within this deep biosphere.

2 Materials and methods

2.1 *Sample collection*

Sediment sample was collected during the 4th Chinese National Arctic Research Expedition in 2010. The sampling location was at 88°23′639″N, 176°37′702″W. Specifically, a single sample was collected by means of a box corer (with a maximum breakthrough of 60 cm and an effective sampling area of 0.25 m^2) at water depth 3 995.7 m. The sediment temperature was 1.6°C, the surface sea water temperature was 2.3°C and salinity was 33.08. The sediment sample was transferred to sterile plastic bag, quickly frozen in liquid nitrogen and then stored at −80°C until analysis.

2.2 *Environmental parameters analysis*

Measurement of chlorophyll and phaeophytin pigments was done as previously described (Boetius and Damm, 1998). Sediment total volatile solids content (VS, w/w, %) or the organic matter percentage (OM, %) was determined by weight difference of the dried and ashed (550°C for 4 h) sediment sample. The content of total soluble polysaccharide was measured by anthrone-sulfuric acid colorimetric method.

Four basic elements, carbon, nitrogen, hydrogen and sulphur concentrations in the Arctic deep sea sediment were analyzed by ELEMENTAR's TOC/TNb analyzers Liqui Toc Ⅱ (S. V. Instruments Analytica Pvt. Ltd.).

2.3 *DNA extraction and pyrosequencing*

Surface 0–10 cm layer of the sediment sample was retrieved in a sterilized hood. Total community genomic DNA was extracted directly from 0.5 g wet weight mixed sediment using FastDNA Spin Kit for Soil (QBIOgene, USA) with Fast Prep™ FP120 cell disrupter (Thermo Electron Corp, USA) at speed of 4.5m/s for 30 s. Recovered DNA was analyzed by 1% (w/v) agarose gel electrophoresis. Before PCR, DNA quantity was spectrophotometrically adjusted with a NanoVue™ Plus Spectrophotometer (GE Healthcare UK Limited, UK).

The genomic DNA was confirmed as containing bacterial and archaeal 16S rDNA by PCR before pyrosequencing. The16S rDNA V3 hypervariable region was amplified with the bacterial primers 8F (5'-AGAGTTTGATCCTGGCTCAG-3') /533R (5'-TTACCGCGGCTGC TGGCAC-3'), and with the archaeal primers Arch344F (5'- ACGGG GYGCAGCAGGCGCGA -3') /Arch915R (5'- GTGCTCCC CCGCC AATTCCT-3'). PCR product was checked by 1% (w/v) agarose gel electrophoresis. DNA fragment was recovered by gel purification (AxyPrep™ DNA Gel Extraction Kit, Axygen Biosciences, Union City, CA 94587 USA). Pyrosequencing was carried out on a Roche GS FLX Titanium pyrosequencer (454 Life Sciences, Branford, CT, USA) under the manufacture's protocol. All sequences in this study are available from the NCBI sequence read archive under run accession number SRR543702 (Bacteria) and SRR546475 (Archaea).

2.4 *Pyrosequencing data processing and operational taxonomic units (OTUs) definition*

After pyrosequencing, the reads were processed by Chopseq (Shanghai Majorbio developed sequence processing software) to trim adapters and primer, resolving in reads with lengths between 420–560 nt for bacteria and between 460–580 nt for archaea. Sequencing noise, such as non-specific amplification, chimeric sequences, ambiguous nucleotides and homologue region was further removed to obtain high-quality reads. In total, 24 777 optimized reads were obtained for bacteria and 5 303 optimized reads were obtained for Archaea. All reads were cluste-red and aligned as compared with Silva database (http://www.arb-silva.de/) release 111 (Pruesse et al., 2007). The microbial ecology community software program Mothur (Schloss et al., 2009) (http://www.mothur.org/wiki/Downloadmothur) was applied to define OTUs at 100% (unique), 97%, 93%, and 90% similarity.

2.5 *Diversity estimations and statistical analysis*

Mothur was also applied to estimate microbial diversity as described (Bowman et al., 2011). The Chao1 estimator and the ACE estimator were used to calculate community richness. The Shannon calculator and the Simpson calculator returned community diversity indices for OTU definition. The coverage calculated the Good's coverage for OTU definition. OTUs were assessed at different similarity levels: unique, 97% (0.03), 95% (0.05), 90% (0.10). Rarefaction curve for the number of observed OTUs as a function of distance between sequences and the number of sequences sampled (see http://www.mothur.org/wiki/Mothur_manual).

2.6 *Taxonomy classification and community structure*

Each OTU's taxonomic classification was determined by alignment with SILVA database (Version 106) (Quast et al., 2013) and using Mothur classification tool (>80% credibility). Community structure of the sample was compared at each different taxonomic level from "phylum" to "genus". Taxons with percentage over 1% were labeled separately on the pie charts. Taxons that contributed less than 1% to the pyrosequencing dataset were grouped into the "others" category.

2.7 *Bacteria taxonomic analysis*

MEGAN was applied to interactively explore the bacterial dataset (Huson et al., 2011) (http://ab.inf.uni-tuebingen.de/softnware/megan/). Sequence reads were aligned to the NCBI taxonomy database (ftp://ftp.ncbi.nih.gov/pub/taxonomy/) for taxonomic assignment. Each node is labeled by a taxon and the number of reads assigned to the taxon. The size of a node is scaled logarithmically to represent the number of assigned reads. MEGAN allows inspection of the assignment of reads to a specific node.

2.8 *Archaeal phylogenetic tree construction*

FastTree software was used to construct archaeal phylo-genetic tree (Price et al., 2010) (http://www.microbesonline.org/fasttree/). Approximately-maximum-likelihood algo-rithm was used to infer newick-formatted phylogenetic tree, and the output profile could be visualized in R project (R Core Team, 2014) (http://www.r-project.org/).

3 Results

3.1 *Environmental parameters*

Chlorophyll pigment content in the sediment was around

0.006 μg/g; while the phaeophytin was at 0.048 μg/g. Carbon, nitrogen, hydrogen and sulphur percentage for the sediment was 1.4%, 0.07%, 0.525% and 0.447% respectively. The C/N ratio was calculated at 19.80. Volatile solid percentage was estimated at 2.45% and the polysaccharide concentration was below detection limit.

3.2 *Microbial community richness and diversity*

To target different levels of diversity within the Arctic deep-sea sediment habitat of the microbial communities, we investigated community richness and diversity of both bacterial and archaeal groups. Totally, we sequenced around 27 000 reads for bacterial group and 5 500 reads for archaeal group. After optimization, we got 24 684 reads for bacteria and 5 303 reads for Archaea, respectively.

Community richness was estimated by the Chao1 and Ace index at different similarities. Shannon index was 4.30 for bacteria community and 3.63 for archaea community (97% similarity). The Good's coverage was 0.973 and 0.979 (97% similarity) for bacteria and archaea respectively (Table 1). Rarefaction curves for bacteria and archaea groups were provided in Appendix Fig. A1. Phylogenetic assignments of sequence reads are listed in Appendix Tables A1 and A2 for bacteria and archaea, respectively. A summary of the bacteria community structure at taxo-nomic class level of Arctic sediment microbial diversity studies was listed in Appendix Table A3.

Table 1. Estimates of richness and diversity for multiple OTU definitions for the bacterial and archaeal communities

Label	Chao1		Ace		Shannon		Simpson		Coverage	
	Bacteria	Archaea	Bacteria	Archaea	Bacteria	Archaea	Bacteria	Archaea	Bacteria	Archaea
Unique	7 751 (7 027, 8 591)	2 192 (1 717, 2 863)	14 550 (13 863, 15 280)	3 809 (3 455, 4 207)	4.71 (4.68, 4.74)	4.26 (4.21, 4.31)	0.048 (0.046, 0.049)	0.037 (0.035, 0.039)	0.929	0.926
0.03	2 504 (2 276, 2 786)	422 (344, 553)	3 282 (3 088, 3 496)	597 (520, 696)	4.30 (4.27, 4.33)	3.63 (3.59, 3.67)	0.055 (0.053, 0.056)	0.053 (0.051, 0.056)	0.973	0.979
0.05	1 607 (1 472, 1 782)	169 (144, 223)	2 010 (1 883, 2 156)	220 (187, 271)	4.01 (3.98, 4.04)	3.19 (3.15, 3.23)	0.070 (0.068, 0.072)	0.071 (0.068, 0.073)	0.982	0.992
0.10	996(898, 1 132)	71 (53, 126)	1 227 (1 135, 1 337)	100 (75, 144)	3.65 (3.63, 3.68)	2.26 (2.23, 2.28)	0.079 (0.077, 0.081)	0.141 (0.137, 0.146)	0.989	0.997

3.3 *Bacteria community structure*

The bacterial phyla Proteobacteria and Bacteroidetes constituted the largest proportions (47.25% and 46.02%) of the total bacterial reads; the Chloroflexi accounted for 3.34%, and the Actinobacteria and other low-abundance phyla accounted for less than 1% of the bacterial sequence dataset. Cyanobacteria was presented at very low level (around 0.02%) in this sediment sample.

At the class level, Flavobacteria was most abundant in the community, comprised 46.05% of total bacterial reads. The Proteobacteria, α-proteobacteria and γ-proteobacteria comprised 22.77% and 21.34% of total reads, while β-and δ-proteobacteria were less represented in the community (1.96% and 0.98% respectively). SAR202_clade, a bacteria class widely spread in marine environment, comprised 0.92% of total reads. Opitutae and Verrucomicrobiae were presented at very low levels in our deep-sea sediment reads (0.02% and 0.01% of total bacterial reads) (Fig. 1).

Flavobacteriales continued to be dominant at order level, comprised 45.41% of bacterial reads. Rhodobacterales was the second dominant and came up with 20.41%, followed by Alteromonadales (8.55%), Oceanospirillales (5.6%), Pseudomonadales (2.27%). No_rank (not classified) reads comprised 5.38% of the community at this taxonomic level. Other clades with percentage over 1% were Pseudomonadales (2.27%), Thiotrichales (1.92%), Burkholderiales (1.81%), Xanthomonadales (1.60%) and Rhizobiales (1.07%). SAR324_clade (Marine_group_B) was presented at a low level of 0.13%.

Flavobacteriaceae and Rhodobacteraceae ranked first and second at family level, accounting for 45.4% and 20.41% of the reads respectively. No_rank (not classified) comprised 7.04% of the community at this level. Alcanivoracaceae which could degrade alkane was present at 2.50% of all reads. SAR406_clade (Marine_group_A) was present at quite low level of 0.03%.

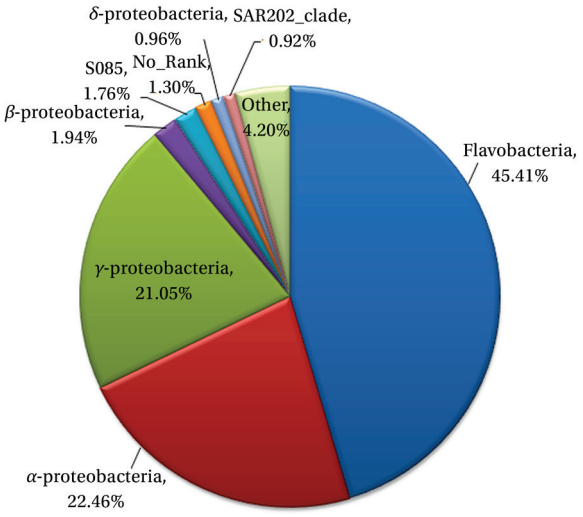

Fig. 1. Numerically dominant clades of the bacteria community (at class level). clades with percentage less than 0.5% were grouped as others (4.2%).

At the genus level (the lowest level assigned), totally 118 genus of bacteria were identified, with 35.21% of the reads unclassified. The top 10 genera were listedin supplemental Table S1. Maribacter (29.44% of the reads) was the most represented clade at this level, followed by Colwelliarepresenting 4.06% of the reads and Pseudoalteromonas comprised of 3.08%. Another important lineage, Roseobacters, were also found widely distributed in the Arctic deep-sea sediment (Table 2).

The bacterial community distributions at order, family, genus levels were shown in Appendix Fig. A2.

Table 2. Roseobacters found in this study

Genus	OTU	Reads
Dinoroseobacter	1	1
Leisingera	1	1
Loktanella	9	637
Marinosulfonomonas	3	64
Octadecabacter	9	238
Roseobacter_clade_DC5-80-3_lineage	1	3
Roseovarius	1	4
Sulfitobacter	38	666

3.4 *Archaea community structure*

Archaea communities were primarily comprised of Thaumarchaeaota and Euryarchaeota. Thaumarchaeaota was overall the most abundant group, representing 96.66% of total archaeal reads, while Euryarchaeota comprised only 3.21% of the reads. No_Rank (not classified) comprised 0.13% of total archaeal reads.

At class level, Marine_Group_I (MGI) from Thaumarchaeaota was numerically dominant, representing 96.38% of all reads. Marine_Benthic_Group_A was the second in Thaumarchaeaota group and comprised of 0.28% of the reads. In Euryarchaeota group, Halobacteria and Methanomicrobia ranked first and second, accounting for 3.07% and 0.13% of the reads respectively (Fig. 2).

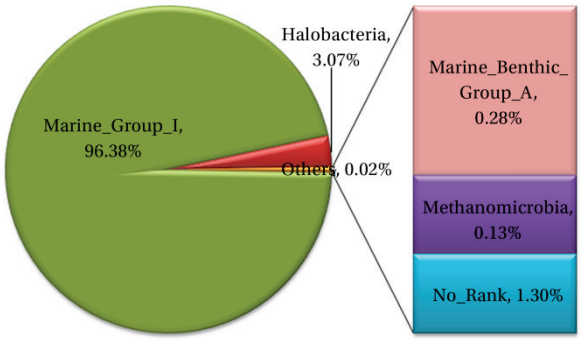

Fig. 2. Numerically dominant clades in the archaea community (at class level). clades with percentage less than 0.5% were grouped as others (0.54%).

The archaeal community distributions at order, family, genus and species levels were shown in Appendix Fig. A3.

About 10 species were identified for archaeal group, including 8 species from Thaumarchaeaota, including *Marine_Group_I uncultured marine_group_I crenarchaeote* (8.41%), *Marine_Group_I uncultured sediment archaeon* (6.28%) and *uncultured_Ce-narchaeum_sp*. (2.85%) etc., in which *uncultured marine_grou-p_I crenarchaeote* (1.75%) belong to *Candidatus_Nitrosopumilus*. Only 2 species, *Methanosaeta uncultured methanogenic archaeon* (0.13%, belong to Methanosaeta) and *Deep_Sea_Hydrothermal_Vent_Gp_6 (DHVEG-6) marine_metagenome* (0.04%) were from Euryarchaeota group (Appendix Table A2). No_Rank comprised of 78.52% of all reads at this level.

3.5 *Bacterial taxonomic analysis*

Bacterial taxonomic analysis was done by MEGAN4 software. Bacteria were classified as 22 phyla, 36 classes, 72 orders, 87 families and 118 genera. The taxonomic affiliation of each clade was clustered according to its relative taxonomic cascade position (Appendix Fig. A4).

3.6 *Archaeal phylogenetic tree analysis*

Since the known archaeal taxonomic information is not as comprehensive as bacterial library, besides taxonomic classification, we aligned the sequenced fragments with NCBI known reference sequences and constructed phylogenetic tree by FastTree (Fig. 3a, 3b). Detailed information of blasted NCBI hit sequenc-es were listed in Appendix Table A4.

4 Discussion

4.1 *Bacterial diversity*

As we were focusing on Arctic deep sea sediment near the North Pole, in order to gain a stratified view of the distribution of bacterial assemblage along the Arctic Polar axis, we compared this study with Bowman et al. (2011) study, which was sampling the Arctic Polar multiple year ice (MYI) and nearby surface sea water (Bowman et al., 2011). At 97% similarity, the bacteria diversity Shannon's index was 4.30 in our study. In Bowman et al. (2011), the Shannon index (mean value) was 3.38 for MYI and 3.72 for seawater. In general, the bacteria community diversity of our study was higher than that of MYI and seawater, regardless of the actual sequencing reads obtained.

For bacterial community composition, Flavobacteria (45.41%) was the dominant clade in our study, followed by α-proteobacteria (22.77%) and γ-proteobacteria (21.34%). In comparison, they found the dominant bacteria communities were γ-proteobacteria (62.3%) for MYI and α-proteobacteria (49%) for sea water; Flavobacteria was 19.9% and 4.6% for MYI and sea water respectively (A list of the class level lineages between this study and other Arctic sea ice/sea water, deep-sea sediment studies could be found in supplemental Table S5). Proteobacteria and the Cytophaga-Flavobacter cluster (Bacteroidetes) are typically abundant bacteria in marine systems (Rappe et al., 1997; Suzuki et al., 1997). To explain for the composition discrepancy between our study and Bowman et al. (2011), we might have to look at the different biological and physicochemical characteristics (primary production, substrate type and accessibility and available electron acceptor), especially the distribution of dissolved organic matter (DOM) between surface sea ice-water and deep sea sediment (Teske et al., 2011). The supply of DOM is an important factor determining rates of microbial activity. And difference in utilization of various DOM may be the underlying reason for the distribution of microbial community groups. The upper layer sea water, especially the euphotic zone, is active in primary production. High concentrations of dissolved organic carbon (DOC) and dissolved organic nitrogen (DON) were observed in Arctic multiyear ice floes, suggesting favorable conditions for bacteria (Thomas et al., 1995). These low-molecular-weight DOM, especially DOC, were quickly taken up and catabolized by microorganism. Only the passive and hard-to-degrade substances (i.e., refractory organic matter, ROM) were transported to the interior ocean and deposit in sediment. So the distribution of DOM/DOC is stratified in different marine systems. And the DOM categories in upper layer sea water are relatively simple compared to that of deep sea sediment (Cottrell and Kirchman, 2000). This may explain why MYI and surface sea water bacterial diversity were lower than that of deep-sea sediment.

Fig. 3a. Phylogenetic tree of archaea community (FastTree) – Upper part.

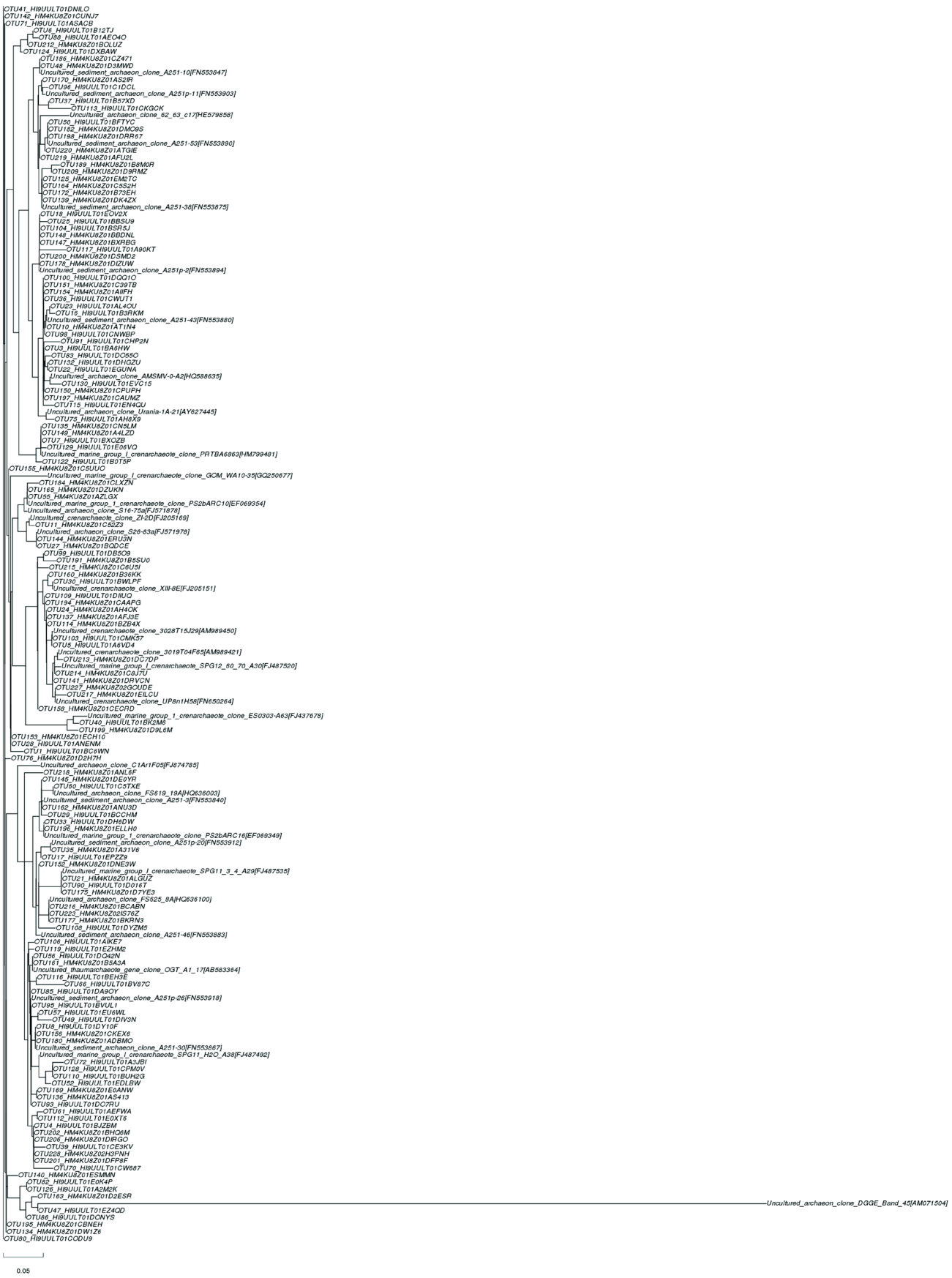

Fig. 3b. Phylogenetic tree of archaea community (FastTree) – Lower part.

On the other hand, organic matter in the marine sediment was controlled by multiple environmental processes such as sea-ice distribution, terrigenous sediment supply, oceanic currents, and surface-water productivity (Stein et al., 1994; Fahl and Stein, 1997). The Arctic deep sea sediment received relatively low input of marine-derived organic matter, because the region is largely covered by multi-year ice, and primary production is highly seasonal and at low average (Wheeler et al., 1996; Gosselin et al., 1997). Considerable amount of terrestrial ROM came from the large Russian and North American rivers. Such inputs shaped the organic component of the Arctic sediments from the shelves to along the slopes and into the basins (Macdonald et al., 1998). Therefore, the fraction of terrigenous organic carbon preserved in Arctic Ocean sediments was much higher than that of marine organic carbon (Stein and MacDonald, 2004). Numerous studies had suggested that high C/N atomic ratios in the sediments of the Arctic deep sea indicate that most of the organic matter was terrigenous derived material (Stein et al., 1994; Fahl and Stein, 1997; Clarke, 2003). We found the C/N ratio of our sample was estimated at 19.8, indicating the organic matter in Arctic sediment was mostly terrigenous originated (Perdue and Koprivnjak, 2007). Such variation in DOM composition also implicated different metabolic processes between sea water and marine sediment habitats.

In bacterial genus taxa, we found that *Maribacter* was the most abundant lineage among the taxonomically-defined genera, representing 29.49% of bacterial reads (7268 out of 24684 reads) in our study, but was representing only 28 out of 12336 bacteria reads (0.23%) in Bowman et al. (2011). *Maribacter* is widely distributed among marine environment (Nedashkovskaya et al., 2004), and we found that this genus also flourished in the Arctic deep-sea sediment. *Maribacter* belong to the *Cytophaga-Flavobacteria* cluster (Bacteroidetes), which is notable for a chemo-organotrophic metabolism style. Various lines of evidence suggest bacteria in this cluster are proficient at degrading high-molecular-weight polymeric organic matter, such as cellulose, chitin, and protein(Nedashkovskaya et al., 2004); while proteobacterias seem to digest low-molecular-weight DOM more quickly (Cottrell and Kirchman, 2000; Kirchman, 2002). This DOM consumption preference explained for the presence of *Maribacter* in Arctic deep-sea sediment. Evidence from study of extracellular enzyme subcellular location also showed that *Flavobacteria* form a unique cluster among abundant marine bacteria groups, indicating the unique metabolism capacity of this clade (Luo, 2012).

As for *Roseobacter*, they also play important roles for the global carbon and sulfur cycle and even the climate (Wagner-Döbler and Biebl, 2006). Genera *Sulfitobacter* and *Loktanella* belong to *Roseobacter*, they show traits of aerobic anoxygenic photosynthesis, oxidizing the greenhouse gas carbon monoxide, and producing the climate-relevant gas dimethylsulfide through the degradation of algal osmolytes.

There are other genera which show specific material metabolizing capacity. For example, *Alcanivorax* is an alkane-degrading marine bacterium that naturally propagates and becomes predominant in crude-oil-containing seawater. They are currently thought to be the world's most important oil-degrading organisms (Kasai et al., 2002). *Oleispira*, with a preference for aliphatic hydrocarbons, is a typical marine hydrocarbonoclastic bacteria like *Alcanivorax* (Wang et al., 2012). J*TB255 marine benthic group*, which was also found in Antarctic marine benthic sediment, was thought to have putative sulfide-oxidizing capability (Bowman and McCuaig, 2003). *Leucothrix* is strictly marine originated and widespread as an epiphyte of marine algae. They show capability to oxidize sulphide, sulfur and thiosulphate, all compounds being oxidized to sulphate (Bland and Brock, 1973).

In addition, given the Arctic deep sea perennial cold environment, a good part of the bacteria found in this study were either obligate or ubiquitous psychrophile marine bacterium,like *Colwellia*, *Pseudoalteromonas* and *Pseudomonas* (Methé et al., 2005). Also this environment is largely suboxic or anoxic compared to the surface layer,anaerobic processes, mainly sulfate reduction, could account for over 50% of total carbon mineralization in marine sediment (Jørgensen, 1982). In this sample, we found members of the sulfate-reducing subclades from δ-proteobacteria, including Desulfobacterales and Desulfovibrionales, and from Firmicutes, including *Desulfosporosinus*.

4.2 *Archaea diversity*

Archaea are now thought to be widely distributed and phylogenetically diverse as bacteria, although the relative abundance of Archaea in the same environment tend to be lower than that of bacteria (Aller and Kemp, 2008; Auguet et al., 2009). As a unique prokaryote assemblage in the Arctic, researches on Archaea have drawn many attentions (Ravenschlag et al., 2001; Bano et al., 2004; Galand et al., 2009; Bowman et al., 2011). In Galand et al. (2009), the archaeal assemblage of western Arctic Ocean water mass from different sampling site was quite diverse. Compared to tagging the V3 region of 16S rDNA, they were sequencing the V6 region, which might affect the taxonomic assignment of the acquired reads (Galand et al., 2009). Bowman et al. (2011) found Archaea was mostly present in Arctic seawater (14%) and Euryar-chaeota was dominant. Since they did not use archaeal specific primers, unspecific amplification might occur which accounted for the small percentage of Archaea. In the present study, we sequenced Archaea group separately and did not found Archaea presence in Bacteria group.

For Archaea taxonomy definition, MGI was now grouped into the newly proposed phylum — Thaumarchaeota. Members of Thaumarchaeota show ammonia-oxidizing capacity and adaptation to autotrophic or possibly mixotrophic lifestyles (Pester et al., 2011). Here, the genus *Candidatus_Nitrosopumilus*, identified in Thaumarchaeota, might be related with Arctic deep sea nitrogen cycling. The other genus *Methanosaeta*, which is capable of *methanogenesis*, is usually found in marine habitats where electron acceptors, such as oxygen, nitrate, and sulfate, are strictly limted (Smith and Ingram-Smith, 2007; Mori et al., 2012). Thus, *Methanosaeta* may play a role in carbon cycles in the Arctic deep-sea environment.

We noticed that at genus level over 96.0% of archaeal reads were defined as No_Rank (unclassified). One possible reason might be that most members in this group still could not be cultured under laboratory condition. A large amount of archaeal OTUs identified in this study might not be previously reported or included in the database. Therefore, there may be novel archaea lineages present in the Arctic deep sea sediment. It is in great need of improved and/or newly-developed archaea isolation and cultivation methods for further investigations.

The limitation of this study was that we did not examine the RNA expression levels of the Arctic deep-sea sediment as did in Sahm and Berninger (1998). Therefore, we lack the information of RNA/DNA ratio to deduce which genes were active or which metabolism processes were dominating in the Arctic deep-sea sediment. Future works in this field could provide with more complete information of microbial life in the oligotrophic and cold Arctic deep-sea sediment.

5 Conclusions

In this article, we extensively examined the bacterial and arc-

haeal community's diversity, richness, and composition of the Arctic deep-sea sediment, and compared our findings with that of other studies on Arctic microbial diversity. We found that the microbial composition of our sample diverged greatly from that of the sea-ice and sea water in the Arctic and from other deep-sea areas. The high percentage of Flavobacteria at class level or *Maribacte*r at genus level in bacterial group stated the terrigenous source of the sediment. The identification of *Candidatus_Nitrosopumilus* and *Methanosaeta* connected archaeal community with carbon and nitrogen cycling in the Arctic deep-sea environment. This study could serve as a fundamental investigation of microbial assembly in the Arctic deep-sea sediment and close to the North Pole.

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Appendix:

Table A1. Classification of reads at each taxonomic level of the Bacteria community

Bacteria	**Abundance**
All	24 684
Phylum	
No_Rank	95
Proteobacteria	11 607
Bacteroidetes	11 305
Chloroflexi	820
Actinobacteria	241
Acidobacteria	120
Planctomycetes	107
Gemmatimonadetes	98
Candidate_division_OD1	75
Firmicutes	32
TM6	32
Cyanobacteria	30
Deferribacteres	24
Trimed	24
Candidate_division_OP3	17
Candidate_division_WS3	8
Verrucomicrobia	8
Nitrospirae	7
BD1-5	6
NPL-UPA2	6
Spirochaetes	6
Candidate_division_BRC1	5
Lentisphaerae	5
Chlorobi	2
Candidate_division_OP11	1
Fibrobacteres	1
MVP-21	1
SM2F11	1
Class	
No_Rank	321
Flavobacteria	11 209
α-proteobacteria	5 543
γ-proteobacteria	5 195
β-proteobacteria	478
S085	434
δ-proteobacteria	238
SAR202_clade	228
Acidimicrobiia	158
Anaerolineae	108
Gemmatimonadetes(class)	98
Acidobacteria(class)	88
Epsilonproteobacteria	81
Cytophagia	77
Phycisphaerae	58
Thermoleophilia	44
JTB23	40
Actinobacteria(class)	39
Planctomycetacia	34
JG30-KF-CM66	26
Chloroplast	25
Deferribacteres(class)	24
Trimed	24
RB25	22
Bacilli	20
Clostridia	12
Holophagae	8
Nitrospira	7
Sphingobacteria	7
OM190	6
Spirochaetes(class)	6
Lentisphaeria	5
Opitutae	5
Caldilineae	2
Ignavibacteria	2
KD4-96	2
Pla3_lineage	2
Verrucomicrobiae	2
Bacteroidia	1
FS118-62B-02	1
Fibrobacteria	1
Napoli-4B-65	1
OPB35_soil_group	1
SC3-20	1
Order	
No_Rank	1 329
Flavobacteriales	11 209
Rhodobacterales	5 038
Alteromonadales	2 111
Oceanospirillales	1 383
Pseudomonadales	561
Thiotrichales	473
Burkholderiales	447
Xanthomonadales	396
Rhizobiales	264
Rhodospirillales	159
Acidimicrobiales	158
Sh765B-TzT-29	122
Anaerolineales	108
Campylobacterales	81

to be continued

Continued from Table A1

Cytophagales	75
Gemmatimonadales	57
Chromatiales	55
Gammaproteobacteria_Order_Incertae_Sedis	51
Sphingomonadales	44
Planctomycetales	34
Solirubrobacterales	34
BPC102	33
SAR324_clade(Marine_group_B)	33
Deferribacterales	24
Trimed	24
BPC015	20
Phycisphaerales	20
PAUC43f_marine_benthic_group	19
C86	18
BD2-11_terrestrial_group	16
Myxococcales	15
DA023	14
Bacilli_Bacillales	13
CCM11a	13
Rhodocyclales	13
Clostridiales	12
Nitrosomonadales	12
Desulfobacterales	11
Enterobacteriales	11
Frankiales	11
Legionellales	11
Propionibacteriales	11
AKIW543	10
KF-JG30-18	9
Bacilli_Lactobacillales	7
GR-WP33-30	7
Nitrospirales	7
Sphingobacteriales	7
Caulobacterales	6
Micrococcales	6
Spirochaetales	6
Corynebacteriales	5
JG37-AG-116	5
Puniceicoccales	5
Hydrogenophilales	4
MSB-3A7_sediment_group	4
Sva0725	4
Acidithiobacillales	3
BD7-8_marine_group	3
DA052	3
Desulfovibrionales	3
EC3	3
MNG3	3
vadinBA30_marine_sediment_group	3
Bdellovibrionales	2
Caldilineales	2
Desulfuromonadales	2
Ignavibacteriales	2
JH-WHS99	2
Order_II_Incertae_Sedis	2
PAUC26f	2
Rickettsiales	2
Verrucomicrobiales	2
Bacteroidales	1
Fibrobacterales	1
Holophagales	1
MD2904-B13	1
NKB17	1
NKB5	1
Neisseriales	1
R76-B128	1
Vibrionales	1
WCHB1-41	1
Family	
No_Rank	1
Flavobacteriaceae	11
Rhodobacteraceae	5
Colwelliaceae	1
Pseudoalteromonadaceae	760
Oceanospirillaceae	629
Alcanivoracaceae	616
Pseudomonadaceae	551
Sinobacteraceae	350
Alteromonadaceae	302
Thiotrichaceae	241
Piscirickettsiaceae	232
Comamonadaceae	205
Burkholderiaceae	187
Rhodospirillaceae	157
Halomonadaceae	134
Anaerolineaceae	108
Brucellaceae	97
OCS155_marine_group	91
Cyclobacteriaceae	59
Gemmatimonadaceae	57
Rhizobiaceae	56
Chromatiaceae	55
Alcaligenaceae	53
Gammaproteobacteria_Order_Incertae_Sedis_Family_Incertae_Sedis	51
Acidimicrobiaceae	48

to be continued

Continued from Table A1

Helicobacteraceae	46
Xanthomonadaceae	46
Psychromonadaceae	39
Planctomycetaceae	34
Campylobacteraceae	33
Elev-16S-1332	32
Sphingomonadaceae	31
Phyllobacteriaceae	27
Rhodobiaceae	27
Trimed	24
Phycisphaeraceae	20
Methylobacteriaceae	17
Rhodocyclaceae	13
PAUC34f	12
Enterobacteriaceae	11
Propionibacteriaceae	11
Sporichthyaceae	11
Coxiellaceae	10
Erythrobacteraceae	10
Moraxellaceae	10
Nitrosomonadaceae	10
Staphylococcaceae	9
Flammeovirgaceae	8
Hyphomicrobiaceae	8
Acidimicrobiales_	7
SAR406_clade(Marine_group_A)	7
Desulfobulbaceae	6
Nitrospiraceae	6
Shewanellaceae	6
Spirochaetaceae	6
Cystobacterineae_	5
Deferribacterales_Family_Incertae_Sedis	5
Puniceicoccaceae	5
Bradyrhizobiaceae	4
Caulobacteraceae	4
Hydrogenophilaceae	4
JG37-AG-15	4
MBAE14	4
Microbacteriaceae	4
Mycobacteriaceae	4
Ruminococcaceae	4
Saprospiraceae	4
Acidithiobacillaceae	3
Bacillales_Bacillaceae	3
Carnobacteriaceae	3
Clostridiales_Family_XI_Incertae_Sedis	3
Cytophagaceae	3
Desulfovibrionaceae	3
Nannocystaceae	3
Nitrospinaceae	3
Bdellovibrionaceae	2
Caldilineaceae	2
Chitinophagaceae	2
Clostridiaceae	2
Desulfobacteraceae	2
Gallionellaceae	2
Hyphomonadaceae	2
Intrasporangiaceae	2
Lactobacillaceae	2
Lactobacillales_Streptococcaceae	2
PHOS-HE36	2
Rhodothermaceae	2
TK34	2
Acetobacteraceae	1
Bacillales_Family_XII_Incertae_Sedis	1
Beijerinckiaceae	1
DEV007	1
Family_XIII_Incertae_Sedis	1
Fibrobacteraceae	1
GR-WP33-58	1
Geobacteraceae	1
Holophagaceae	1
Lachnospiraceae	1
Legionellaceae	1
M.0319-6G20	1
Neisseriaceae	1
OPB95	1
Peptococcaceae	1
Rubritaleaceae	1
S.288-2	1
S.480-2	1
S24-7	1
Sva0996_marine_group	1
Vibrionaceae	1
Genus	
No_Rank	8 692
Maribacter	7 268
Colwellia	1 003
Pseudoalteromonas	760
Sulfitobacter	666
Loktanella	637
Alcanivorax	616
Pseudomonas	551
Oleispira	352
JTB255 marine benthic group	350
Leucothrix	241

to be continued

Continued from Table A1

Octadecabacter	238	*Maritimimonas*	10
Aestuariicola	230	*Nitrosomonas*	10
Alteromonas	227	*Polaribacter*	9
Spongiispira	210	*Coxiella*	8
Delftia	189	*Planctomyces*	8
Methylophaga	187	*Staphylococcus*	8
Sediminicola	186	*Flavobacteriaceae*	7
Rhodobacteraceae	139	*Ilumatobacter*	7
Burkholderia	134	*OM60(NOR5)_clade*	7
Halomonas	134	*Shewanella*	6
Formosa	103	*Winogradskyella*	6
Ochrobactrum	97	*Aquabacterium*	5
Lutibacter	64	*Caldithrix*	5
Marinosulfonomonas	64	*Gemmatimonadaceae*	5
Defluviicoccus	60	*Nitrospiraceae_Nitrospira*	5
Pibocella	60	*Phycisphaera*	5
Cyclobacterium	58	*Pir4_lineage*	5
Rhizobium	56	*Acinetobacter*	4
Nitrosococcus	55	*Bradyrhizobium*	4
Rhodospirillaceae	47	*Brevundimonas*	4
Sulfurovum	45	*Eudoraea*	4
Ralstonia	43	*Flavobacterium*	4
Stenotrophomonas	40	*Mycobacterium*	4
Psychromonas	39	*Porticoccus*	4
Oceanobacter	38	*Roseovarius*	4
Pusillimonas	36	*Acidithiobacillus*	3
Algibacter	35	*Alcaligenes*	3
Piscirickettsiaceae	35	*Bacillaceae_Bacillus*	3
Arcobacter	33	*Desulfobulbus*	3
Marinicella	30	*Desulforhopalus*	3
Candidatus_Endobugula	28	*Desulfovibrio*	3
Leeuwenhoekiella	28	*Enhydrobacter*	3
Ahrensia	27	*Flammeovirgaceae*	3
Novosphingobium	27	*Granulicatella*	3
Rhodobium	27	*Hymenobacter*	3
Thalassolituus	24	*Methyloversatilis*	3
Trimed	24	*Rhodopirellula*	3
Arenibacter	23	*Roseobacter_clade_DC5-80-3_lineage*	3
Glaciecola	23	*Sphingomonas*	3
Arenicella	21	*Thalassospira*	3
Methylobacterium	17	*Thiomicrospira*	3
Olleya	17	*Ancalomicrobium*	2
Acidimicrobiaceae	16	*Aquicella*	2
Achromobacter	14	*Bdellovibrio*	2
Urania-1B-19_marine_sediment_group	14	*Blastopirellula*	2
Escherichia-Shigella	11	*Caldilineaceae*	2
Propionibacterium	11	*Candidatus_Entotheonella*	2
Cupriavidus	10	*Clostridium*	2

to be continued

Continued from Table A1

Dechloromonas	2
Hyphomonas	2
Janibacter	2
Lactobacillus	2
Marinobacter	2
Peptoniphilus	2
Psychrobacter	2
Rubricoccus	2
Ruminococcaceae	2
Spirochaeta	2
Streptococcaceae_Streptococcus	2
Thiobacillus	2
Variovorax	2
Actibacter	1
Algoriphagus	1
Anaerolineaceae	1
Beijerinckiaceae	1
Belnapia	1
Comamonas	1
Curvibacter	1
Desulfosporosinus	1
Erythrobacter	1
Exiguobacterium	1
Fibrobacteraceae	1
Finegoldia	1
Flavisolibacter	1
Gaetbulibacter	1
Geoalkalibacter	1
Haliea	1
Hydrogenophaga	1
Hydrotalea	1
Jeotgalicoccus	1
Legionella	1
Leisingera	1
Lentimonas	1
Limnohabitans	1
Microbacterium	1
Neisseria	1
Nitrospiraceae	1
Paracoccus	1
Persicobacter	1
Petrobacter	1
Planctomycetaceae	1
Rubritalea	1
Sphingobium	1
Sulfuricella	1
Sulfuricurvum	1
Tenacibaculum	1
Ulvibacter	1
Vibrio	1
Yonghaparkia	1

Table A2. Classification of reads at each taxonomic level of the Archaea community

Archaea	**Abundance**
All	5 303
Phylum	
Thaumarchaeota	5 126
Euryarchaeota	170
No_Rank	7
Class	
Marine_Group_I	5 111
Halobacteria	163
Marine_Benthic_Group_A	15
Methanomicrobia	7
No_Rank	7
Order	
No_Rank	5 133
Halobacteriales	163
Methanosarcinales	7
Family	
No_Rank	5 134
Deep_Sea_Hydrothermal_Vent_Gp_6 (DHVEG-6)	161
Methanosaetaceae	7
Deep_Sea_Euryarchaeotic_Group (DSEG)	1
Genus	
No_Rank	5 110
Candidatus_Nitrosopumilus	186
Methanosaeta	7
Species	
No_Rank	4 164
Marine_Group_I_uncultured_marine_group_I_crenarchaeote	446
Marine_Group_I_uncultured_sediment_archaeon	333
uncultured_Cenarchaeum_sp.	151
uncultured_marine_group_I_crenarchaeote	93
Marine_Group_I_uncultured_archaeon	82
Marine_Benthic_Group_A_uncultured_marine_group_1_crenarchaeote	10
Marine_Group_I_uncultured_marine_group_1_crenarchaeote	10
Methanosaeta_uncultured_methanogenic_archaeon	7
Marine_Benthic_Group_A_uncultured_crenarchaeote	5
Deep_Sea_Hydrothermal_Vent_Gp_6 (DHVEG-6)_marine_metagenome	2

Table A3. Summary of Arctic sedimentary microbial diversity studies

Sampling site (depth)	Methods	Main results	Reference
Tromsø (northern Norway) Spitsbergen, Svalbard (115–329 m)	DAPI-staining; rRNA slot-blot hybridization	Prokaryotes number at 2×10^8 to 4×10^9 cells/cm^3; Bacteria was dominant (>96%); Archaea was less presented.	Sahm and Berninger (1998)
Hornsund (155 m) , Storfjord (175 m)	rRNA slot-blot hybridization; 16SrDNA DGGE-analysis	Mainly psychrophilic sulfate-reducing, belong to δ-Proteobacteria.	Sahm et al. (1999)
Same as above	16SrDNA clone library	δ-Proteobacteria (43.4%); γ-Proteobacteria (18.1%); α- Proteobacteria (0.6%).	Ravenschlag et al. (1999)
Smeerenburg fjord, Svalbard (218 m)	Fluorescence in situ hybridization; rRNA slot blot hybridization	Bacteria were about 65.4% ±7.5%; Archaea was about 4.9% ± 1.5%.	Ravenschlag et al. (2001)
Kongsfjorden, Svalbard (20 m)	16SrDNA clone library	Bacteria around 71.3%; Archaea around 26.7%. In bacteria, Proteobacteria was about 60.4%, others were Bacteroidetes. Planctomycetes and Verrucomicrobia were detected at 1.9%. In Archaea, Crenarchaeota (99.3%); Euryarchaeota (0.7%).	Tian et al. (2009)
Smeerenburg fjord, Svalbard (211 m)	16S rDNA clone; sequencing	In surface sediment (1–2 cm), γ-Proteobacteria was dominant (49%), followed by Bacteroidetes, δ-Proteobacteria and Acidobacteria. In subsurface sediment (3–9 cm), δ-Proteobacteria was dominant (20%), Sphingobacteria and Bacteroides was at 14%， γ-Proteobacteria was at 9%.	Teske et al. (2011)
Siberian continental margin, Laptev Sea (37–3427 m)	454 massively parallel tag sequencing	Surface sediment (0–1 and 1–2 cm), totally 10 samples. At Phylum level, Proteobacteria (51%), Actinobacteria (10%), Acidobacteria (9%); At class level, γ-proteobacteria (26%), δ-proteobacteria (14%), Actinobacteria (10%), α-proteobacteria (7%), Acidobacteria (6%).	Bienhold et al. (2011)
Arctic Mid-Ocean Ridge system (about 2 000 m) (Two 3 m-long sediment cores retrieved from 15 km southwest (SW) and 15 km northeast (NE) of the Loki's Castle Vent Field)	Pyrosequencing	http://services.cbu.uib.no/supplementary/jorgensen2012	Jorgensen et al. (2012)

Table A4. NCBI blasted hit sequences of pyrosequencing results for Archaea

OUT name	OUT size	Hit_id	Hit_definition
OTU2	772	gi|285310361|emb|FN650243.1|	Uncultured crenarchaeote partial 16S rRNA gene, clone RD3Ti183
OTU4	532	gi|218664886|gb|FJ487492.1|	Uncultured marine group I crenarchaeote SPG11_H2O_A38 16S ribosomal RNA gene, partial sequence
OTU136	345	gi|348591359|emb|FN553918.1|	Uncultured sediment archaeon partial 16S rRNA gene, clone A251p-26
OTU3	334	gi|315441116|gb|HQ588635.1|	Uncultured archaeon clone AMSMV-0-A2 16S ribosomal RNA gene, partial sequence
OTU44	296	gi|285310361|emb|FN650243.1|	Uncultured crenarchaeote partial 16S rRNA gene, clone RD3Ti183
OTU19	237	gi|285310389|emb|FN650271.1|	Uncultured crenarchaeote partial 16S rRNA gene, clone VP8N1B80
OTU5	234	gi|251730599|emb|AM989450.1|	Uncultured crenarchaeote partial 16S rRNA gene, clone 3028T15J29
OTU146	175	gi|285310389|emb|FN650271.1|	Uncultured crenarchaeote partial 16S rRNA gene, clone VP8N1B80
OTU12	173	gi|310787841|gb|HQ287130.1|	Uncultured archaeon clone GL81-A001 16S ribosomal RNA gene, partial sequence
OTU10	155	gi|348591316|emb|FN553880.1|	Uncultured sediment archaeon 16S rRNA gene, clone A251-43
OTU81	127	gi|251730594|emb|AM989445.1|	Uncultured crenarchaeote partial 16S rRNA gene, clone 3026T90H79
OTU9	113	gi|71979695|emb|AM071504.1|	Uncultured archaeon partial 16S rRNA gene, clone DGGE Band 45
OTU138	102	gi|285310389|emb|FN650271.1|	Uncultured crenarchaeote partial 16S rRNA gene, clone VP8N1B80
OTU141	99	gi|251730599|emb|AM989450.1|	Uncultured crenarchaeote partial 16S rRNA gene, clone 3028T15J29
OTU7	94	gi|301177898|gb|HM799481.1|	Uncultured marine group I crenarchaeote clone PRTBA6863 small subunit ribosomal RNA gene, partial sequence
OTU37	83	gi|348591339|emb|FN553903.1|	Uncultured sediment archaeon partial 16S rRNA gene, clone A251p-11
OTU134	72	gi|285310361|emb|FN650243.1|	Uncultured crenarchaeote partial 16S rRNA gene, clone RD3Ti183
OTU24	65	gi|251730599|emb|AM989450.1|	Uncultured crenarchaeote partial 16S rRNA gene, clone 3028T15J29
OTU27	61	gi|220898623|gb|FJ571978.1|	Uncultured archaeon clone S26-83a 16S ribosomal RNA gene, partial sequence
OTU18	58	gi|348591330|emb|FN553894.1|	Uncultured sediment archaeon partial 16S rRNA gene, clone A251p-2
OTU34	55	gi|220898456|gb|FJ571811.1|	Uncultured archaeon clone S16-8a 16S ribosomal RNA gene, partial sequence
OTU150	49	gi|50727780|gb|AY627445.1|	Uncultured archaeon clone Urania-1A-21 16S ribosomal RNA gene, partial sequence
OTU8	48	gi|348591303|emb|FN553867.1|	Uncultured sediment archaeon 16S rRNA gene, clone A251-30
OTU28	43	gi|220898523|gb|FJ571878.1|	Uncultured archaeon clone S16-75a 16S ribosomal RNA gene, partial sequence
OTU33	42	gi|118344706|gb|EF069349.1|	Uncultured marine group 1 crenarchaeote clone PS2bARC16 16S ribosomal RNA gene, partial sequence
OTU17	36	gi|316980963|dbj|AB583374.1|	Uncultured thaumarchaeote gene for 16S rRNA, partial sequence, clone: OGT_A4_47
OTU139	36	gi|348591311|emb|FN553875.1|	Uncultured sediment archaeon 16S rRNA gene, clone A251-38
OTU135	36	gi|301177898|gb|HM799481.1|	Uncultured marine group I crenarchaeote clone PRTBA6863 small subunit ribosomal RNA gene, partial sequence
OTU156	33	gi|348591303|emb|FN553867.1|	Uncultured sediment archaeon 16S rRNA gene, clone A251-30
OTU21	27	gi|218664929|gb|FJ487535.1|	Uncultured marine group I crenarchaeote SPG11_3_4_A29 16S ribosomal RNA gene, partial sequence
OTU167	26	gi|329757553|gb|JF747711.1|	Uncultured archaeon clone MT5A48 16S ribosomal RNA gene, partial sequence

Table A5. Bacterial community compositions between studies (class level)

Bacteria (Class level)	Our study (3 995.7 m)	Arctic sediments (Bienhold et al., 2011)	Arctic MYI (Bowman et al., 2011)	Arctic seawater (Bowman et al., 2011)
Flavobacteria	45.41%	–	19.9%	4.6%
α-proteobacteria	22.46%	7%	4%	49%
γ-proteobacteria	21.05%	26%	62.3%	3.5%
β-proteobacteria	1.94%	–	–	–
S085	1.76%	–	–	–
δ-proteobacteria	0.96%	14%	<1%	7%
SAR202_clade	0.92%	–	–	–
Acidimicrobia	0.64%	–	–	–
Acidobacteria	0.36%	6%	–	–
Actinobacteria	0.16%	10%	–	–
Opitutae	0.02%	–	2%	<1%
Cyanobacteria	0.02%	–	7%	5.8%
Unclassified	No_Rank, 1.3%	–	3%	29%

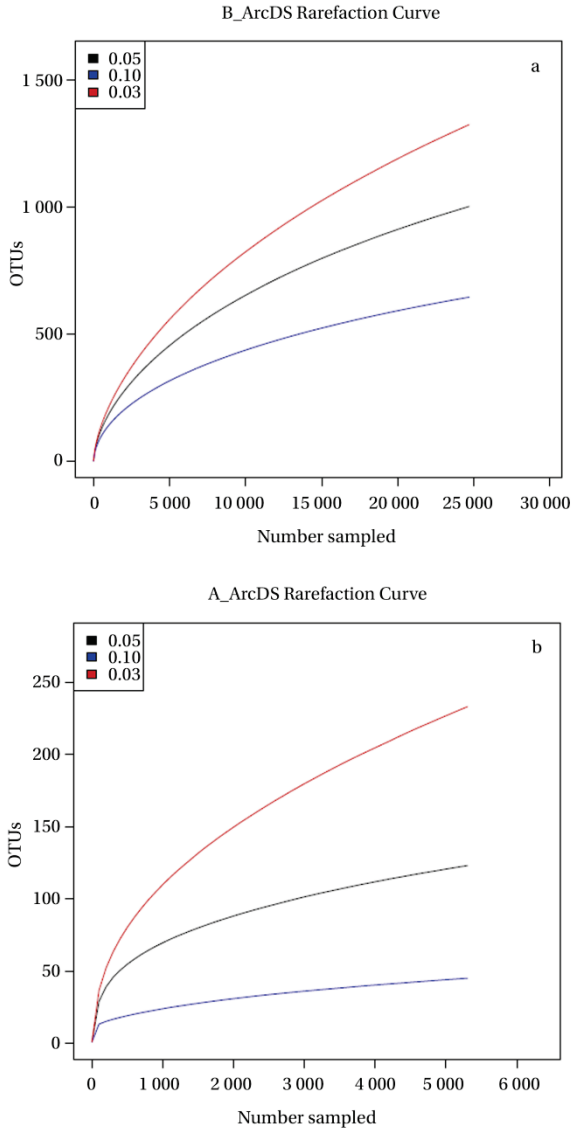


Fig. A1. Rarefaction curves for bacteria (a) and archaea (b) communities from the arctic deep-sea sediment. OTU similarity levels were set at 97%, 95%, and 90%.

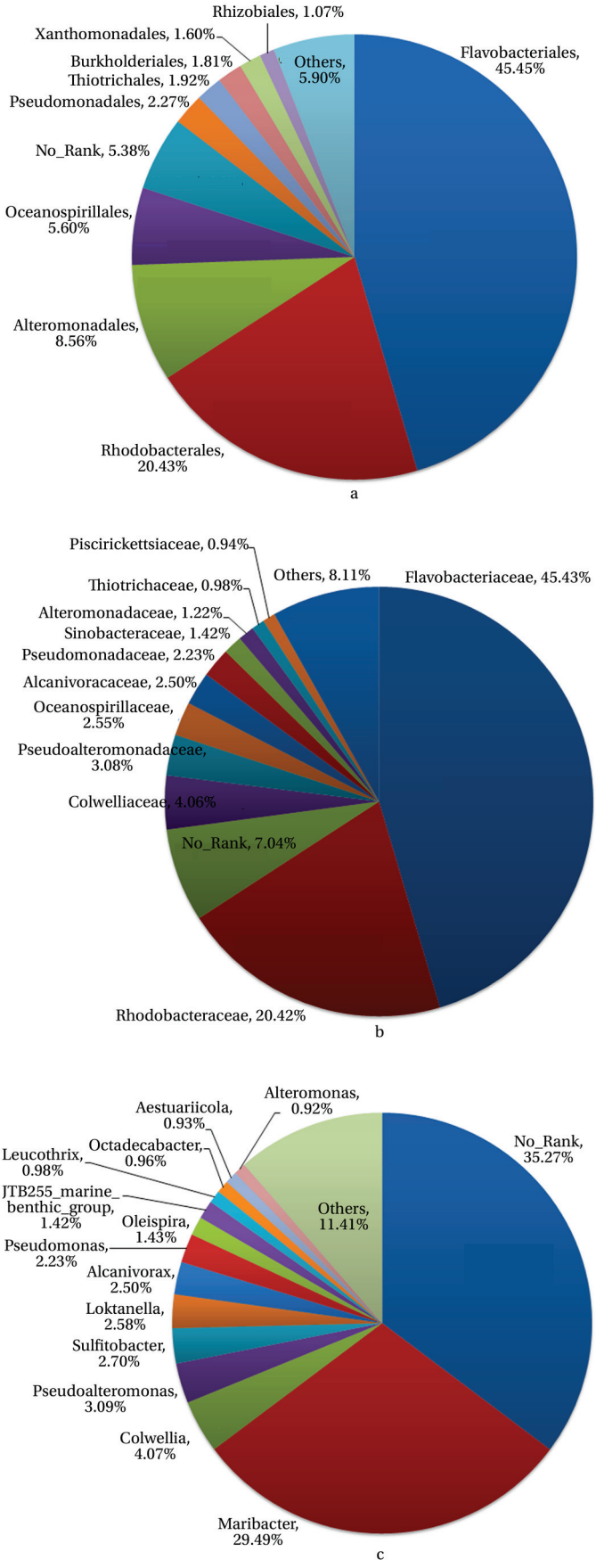
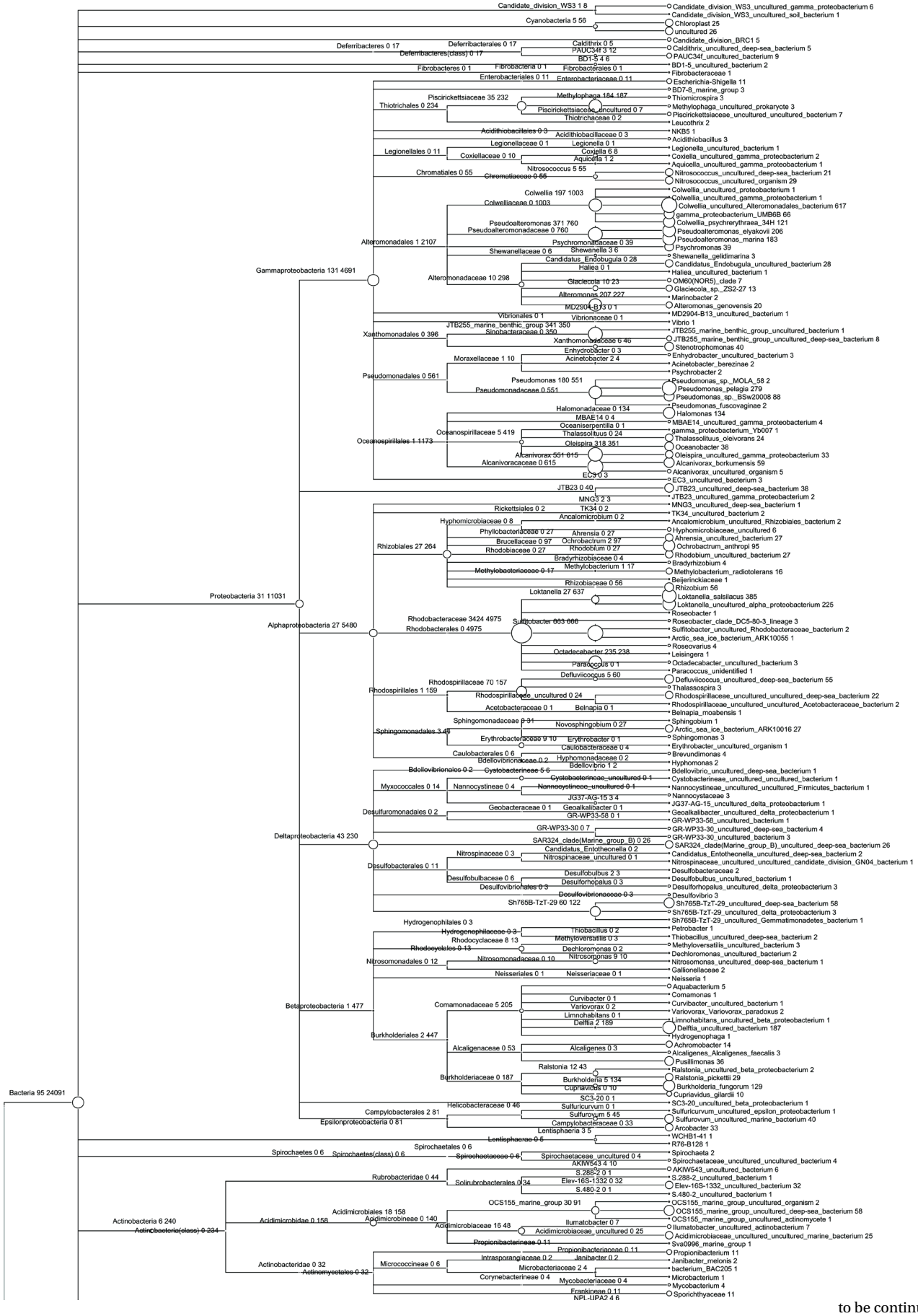


Fig. A2. Bacterial community distributions at order (a), family (b) and genus (c) levels.

Fig. A3. Archaeal community distributions at order (a), family (b), genus (c) and species (d) levels.



to be continued

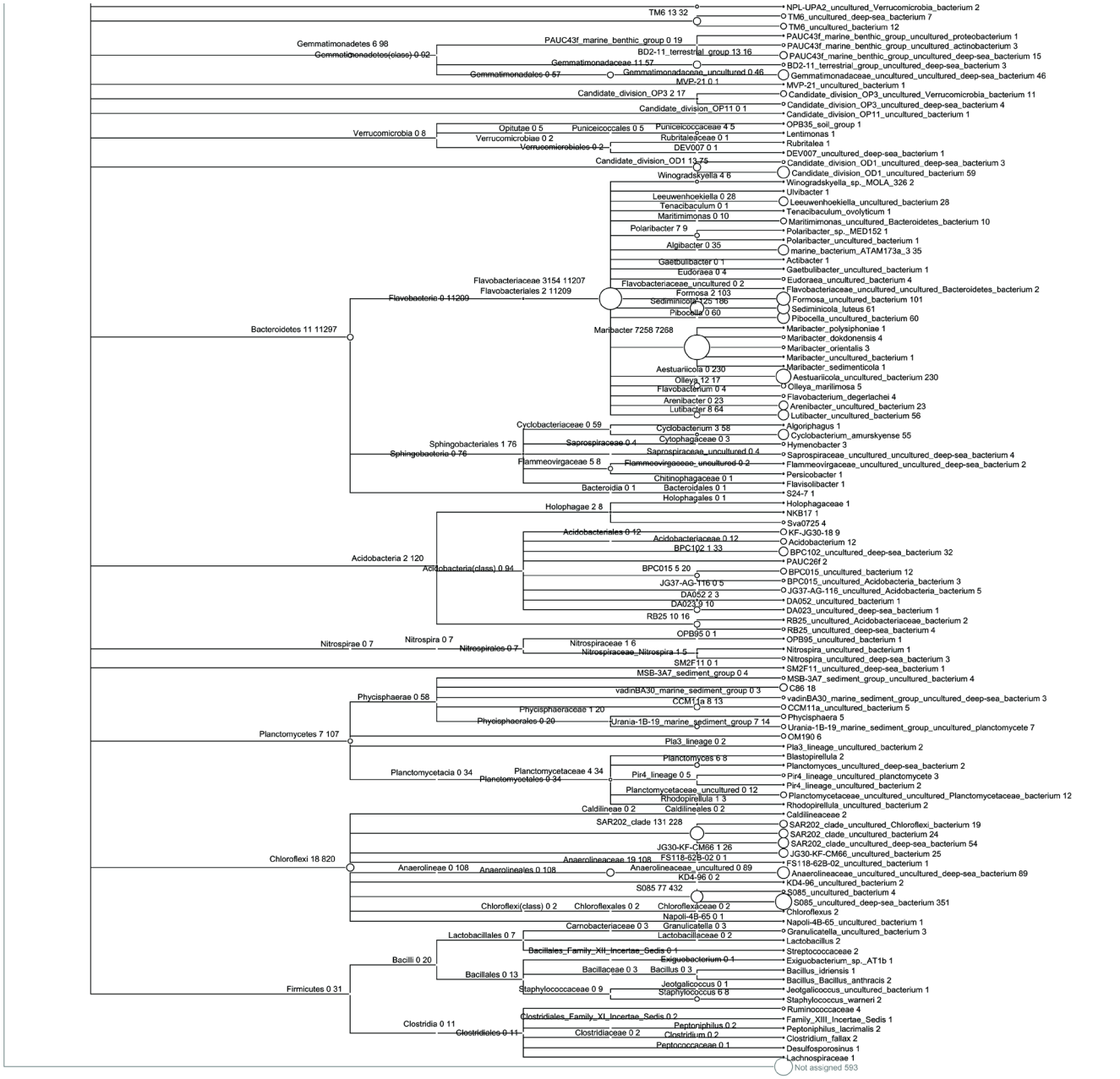


Fig. A4. Bacteria taxonomic affiliation of each identified clade (Megan).