Evolutionary Ecology Research, 2018, **19**: 407–421

Communities and diversities of bacteria and Archaea in Arctic seawater

Jia-Pei Liu*, Ying-Ping Fan*, Xue-Zhu Liu and Jian-Xin Wang[#]

Marine Microorganism Ecological and Application Laboratory, Zhejiang Ocean University, Zhejiang, China

ABSTRACT

Questions: (1) What is the diversity and abundance of bacteria and Archaea in the surface seawater off the Svalbard Archipelago? (2) What are some prokaryotic functions in this region?

Methods: We sampled five different locations in the Svalbard Sea (Arctic Ocean). We used high-throughput sequencing based on 16S rRNA genes to investigate the diversity and abundance of bacteria and Archaea. We also used PICRUSt analysis to determine the functional sequence copies in the surface seawater.

Results: Proteobacteria were the dominant taxon in all samples, followed by Bacteroidetes, Actinobacteria, Planctomycetes, Verrucomicrobia, Cyanobacteria, Chloroflexi, NKB19, Acidobacteria, Firmicutes, and Armatimonadetes. α-Proteobacteria and γ-Proteobacteria dominated among the Proteobacteria. At site BJ7, β-Proteobacteria and γ-Proteobacteria were more abundant than the other Proteobacteria. Crenarchaeota was the most abundant phylum of Archaea. Functional analyses showed that the copy reads of functional genes that pertain to carbon cycling are the most abundant, with methane metabolism dominant; the fewest reads appeared in phosphonate and phosphinate metabolism.

Keywords: Archaea, Arctic Sea, bacteria, biotic community, function, Svalbard Archipelago.

INTRODUCTION

Prokaryotes play an important role in the marine ecosystem (Bacelar-Nicolau *et al.*, 2003). They have a huge impact on the metabolism and function of the ecosystem (Gasol and Duarte, 2000), as well as its biogeochemical cycles (Tortell *et al.*, 1999). Prokaryotes, as the main primary producers, can influence climate change, control the transfer of nutrients and energy in marine environments, provide medicine and food for humans, and impact global environmental change (Kasting and Siefert, 2002). Prokaryotes are also crucial in the marine food chain and in interactions in marine environments (Karl, 2002).

The Arctic Ocean, with its extreme environmental conditions, is one of the most distinctive regions on Earth (Dong *et al.*, 2016). This ocean is hydrographically complex, with double

* These authors contributed equally to this study and share first authorship.
[#]Correspondence: J.-X. Wang, Marine Microorganism Ecological and Application Laboratory, Zhejiang Ocean University, No. 1 South Haida Road, Zhejiang 316022, China. email: jxwang@zjou.edu.cn

inflows of Pacific (through the Bering Strait) and Atlantic (through the Fram Strait) water (Han *et al.*, 2015). The Arctic Ocean has a permanent covering of sea ice, which is an obstacle to studies of biodiversity and ecology (Bachy *et al.*, 2011).

The Svalbard Archipelago lies at the intersection of the northern North Atlantic and Arctic Ocean basins, where warm Atlantic water and cold, less saline Arctic Ocean water mix. Throughout the Holocene, the relative locations of this significant boundary, along with associated sea-ice feedbacks, has varied and influenced Svalbard's climate (De Wet *et al.*, 2017).

In this study, our aims were to: (1) investigate the diversity and community structure of bacteria and Archaea in Svalbard seawater at five different locations; (2) determine the abundance and distribution of bacteria and Archaea at those same locations; and (3) analyse prokaryotic functions in this region.

The Arctic is experiencing dramatic climate change and will be most affected by future warming (ACIA, 2004). One, already visible sign of global warming is the rapid retreat of glaciers. Glacial meltwater inflow, with its particulate matter and freshwater, affects microbial community composition in Arctic fjords (Piquet *et al.*, 2010; Urbanski *et al.*, 2017). These changes have implications for both science and human livelihoods. In recent years, intensive development of no-till cultivation techniques has contributed significantly to the expansion of knowledge on microbial diversity and community composition in the environment (Zengler, 2009; Piquet *et al.*, 2016). Sinha *et al.* (2017b) used high-throughput sequencing to examine bacterial diversity in Kongsfjorden during the summer and fall of 2012. They assigned 11,999 operational taxonomic units (OTUs) to 19 known phyla and five genera *incertae sedis*. Proteobacteria was the most abundant phylum (55.9–61.0%). Pedrós-Alió *et al.* (2015) reviewed recent research on the diversity of planktonic microorganisms in the Arctic Ocean based on the technique of high-throughput sequencing. They found that the Arctic Ocean is similar to the other oceans in terms of the abundance and general composition of microbial communities. However, bacterial communities in the Arctic vary significantly across regions and seasons. Justifiably, the variety of microbial communities has attracted attention and deserves further study.

MATERIALS AND METHODS

Sample collection

We collected samples during a 2014 expedition (24–31 July) from five locations around the Svalbard archipelago (Fig. 1). These sites are located between 11°05′–15°36′E and 78°12′–79°33′N (Table 1). The samples, labelled BJ2, BJ5, BJ6, BJ7, and BJ8, were subpackaged and stored in airtight sterile plastic bottles in a −20°C freezer during the expedition and then at −80°C upon our return to the laboratory.

DNA extraction, amplification and sequencing

We filtered the samples through 0.22-μm pore-size polycarbonate nucleopore membranes (Merck Millipore Ltd., Billerica, MA) using a vacuum pump suction filter, at a vacuum of 20 kPa (Porter and Feig, 1980). We then extracted DNA from the filters using the extraction protocol for the MOBIO PowerWaterfi DNA Isolation Kit (No filters) (Mo Bio Laboratories, Carlsbad, CA). All operational steps were carried out in accordance with

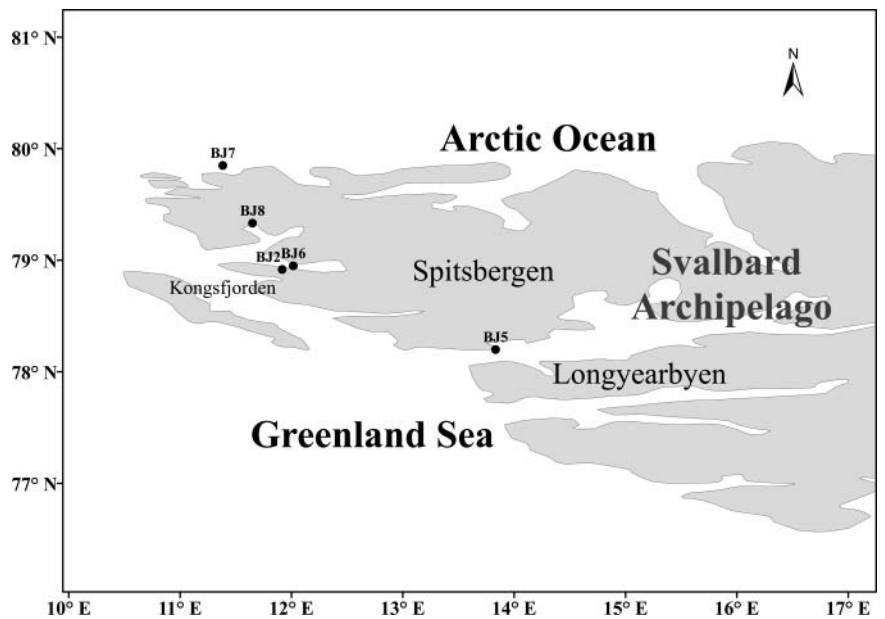

Fig. 1. Map of sampling sites around the Svalbard Archipelago.

the manufacturer's instructions. Using a protein nucleic acid detector (Bio-Rad, Hercules, CA), we measured the concentration of purified DNA. The last elution step of the extraction protocol for DNA resulted in five tubes containing 60 μL of DNA extract for each sample. We sent the DNA extractions to the Novogene Company for amplification and sequencing. They performed Amplicon PCR on variable regions 4 and 5 of the 16S rRNA gene. The bacterial 16S rRNA gene of the extracted DNA was amplified using the universal primers 519F (5′-GTGCCAGCMGCCGCGGTAA-3′) and 915R (5′-CCGTCAATTCCTTTGAGTTT-3′) (Xiao *et al.*, 2016). The archaeal 16S rRNA gene was amplified using primers 519F (5′-CAGCCGCCGCGGTAA-3′) and 915R (5′-GTGCTCCCCCGCCAATTCCT-3′) (Masoud *et al.*, 2011).

Sequence analysis

Sequence data were quality filtered using the QIIME 1.9.1 script split_libraries_fastq.py (Caporaso *et al.*, 2010). De novo OTU picking (Hauptmann *et al.*, 2014) in QIIME was done by assigning similar sequences to a single OTU at a 97% similarity threshold. We used the results for the subsequent diversity and function analyses. The diversity analyses included counts of OTUs (number of unique tag sequences) and estimates of the number of species, rarefaction curves, community structures and heatmap.

For analyses, we used the Shannon (Shannon-Wiener diversity) index, Simpson index, and the Abundance Coverage Estimator (ACE) (Thompson *et al.*, 2017). We used the Simpson index and ACE (Rosenzweig *et al.*, 2010; Wang *et al.*, 2017) to estimate the number of species. The Shannon index measures not only the number of species but also the evenness of their abundances (Thompson *et al.*, 2017). PD_whole_tree was used to obtain averages of the phylogenetic diversity (Faith, 1992).

To display the community structures of bacteria and Archaea at the five locations, we used Origin 9.0 to draw the composition plot at different levels. The heat map was compiled using the VEGAN package in R (Oksanen *et al.*, 2009). In order to analyse the microbial predictive functions, PICRUSt (phylogenetic investigation of communities by reconstruction of unobserved states) was used to detect 16S rRNA marker gene sequences.

RESULTS

Microbial sequence information

We obtained 266,840 and 279,497 qualified reads from bacteria and Archaea respectively (Table 1). At site BJ8, bacteria yielded the largest number of qualified reads, Archaea the least.

When we focused on observed OTUs, we found differing results. We obtained a total of 5043 bacterial OTUs from the five samples. Site BJ5 yielded the largest number of OTUs, while BJ2 yielded the second largest. There was no significant difference between the rest of the sites. The rarefaction curves of bacteria (Fig. 2) show the same result; the curves of

Table 1. Sample locations, sequence information and raw diversities in the Arctic Ocean Survey

	Locations		Bacteria		Archaea	
Site	Longitude	Latitude	Qualified reads	Observed OTUs	Qualified reads	Observed OTUs
BJ2	11°55′01″E	78°55′01″N	53,509	1471	61,339	6
BJ5	13°49′58″E	78°12′00″N	53,708	1868	60,162	7
BJ6	12°01′01″E	78°57′00″N	51,148	787	51,287	7
BJ7	11°22′58″E	79°50′60″N	45,895	326	57,319	9
BJ8	11°39′00″E	79°19′58″N	62,580	591	49,390	8

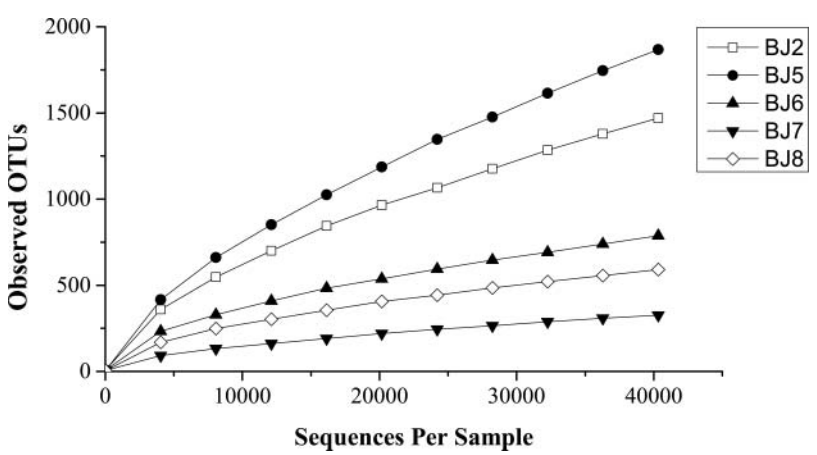

Fig. 2. Rarefaction curves of bacteria from the five sampling sites.

BJ2 and BJ5 rise much more steeply than those of the other three. In Archaea, most reads were bacterial, or unassigned and filtered with QIIME (these data are not shown). Altogether, we obtained 37 OTUs for Archaea. Site BJ7 had the most (nine) and BJ2 had the least (six). Site BJ5 yielded the largest number of OTUs (1868), while BJ2 yielded the second largest (1471). There was no significant difference between the rest of the sites (Table 1).

Measures of diversity

Table 2 shows the results of microbial diversity for each of the sampling sites. No dominant pattern emerges. In both bacteria and Archaea, both the Shannon index and the Simpson index were highest at site BJ2 and lowest at BJ8. The Abundance Coverage Estimator (ACE) was highest at BJ5 and lowest at BJ7. The phylogenetic diversity of bacteria was also highest at site BJ5 and lowest at BJ7. In contrast, the phylogenetic diversity of Archaea was highest at BJ2 and lowest at BJ8.

Bacterial community structure

We found bacterial sequences belonging to 12 known phyla (Fig. 3): Proteobacteria (62.13%), Bacteroidetes (32.23%), Actinobacteria (1.91%), Planctomycetes (1.25%), Verrucomicrobia (1.04%), Cyanobacteria (0.25%), Chlamydiae (0.18%), NKB19 (0.15%), Acidobacteria (0.15%), Firmicutes (0.13%), Armatimonadetes (0.10%), and others (0.49%). The group 'others' included bacteria that were unassigned and less than 0.1% of the total. Proteobacteria included α-Proteobacteria (71.40%), β-Proteobacteria (7.60%), γ-Proteobacteria (20.5%), δ-Proteobacteria (0.45%), and ε-Proteobacteria (0.05%). Even though the major component of each site was not the same, Proteobacteria and Bacteroidetes always dominated. Among the Proteobacteria, α-Proteobacteria was the major component except at site BJ7, where β-Proteobacteria and γ-Proteobacteria both exceeded α-Proteobacteria. Other phyla like Cyanobacteria, NKB19, Acidobacteria, and Armatimonadetes were absent from some samples.

The bacterial heatmap (Fig. 4) confirms the results shown in Fig. 3. Most phyla had low diversity except Proteobacteria and Bacteroidetes. Furthermore, the heatmap also reveals the relationships of these five sites. They could be divided into three groups: BJ5 and BJ6; BJ2 and BJ8; and BJ7.

Table 2. Indices and estimates of microbial diversity

	Bacteria				Archaea			
Site	Shannon	Simpson	ACE	PD_whole_tree	Shannon	Simpson	ACE	PD_whole_tree
BJ2	5.75	0.94	4330	72.55	2.50	0.92	2500	1.79
BJ5	4.91	0.83	5967	96.15	2.17	0.79	3118	1.56
BJ6	4.59	0.85	2001	48.46	1.53	0.81	1906	1.41
BJ7	3.51	0.84	909	22.29	2.38	0.81	1101	1.62
BJ8	3.50	0.80	1610	35.86	1.28	0.77	1532	1.81

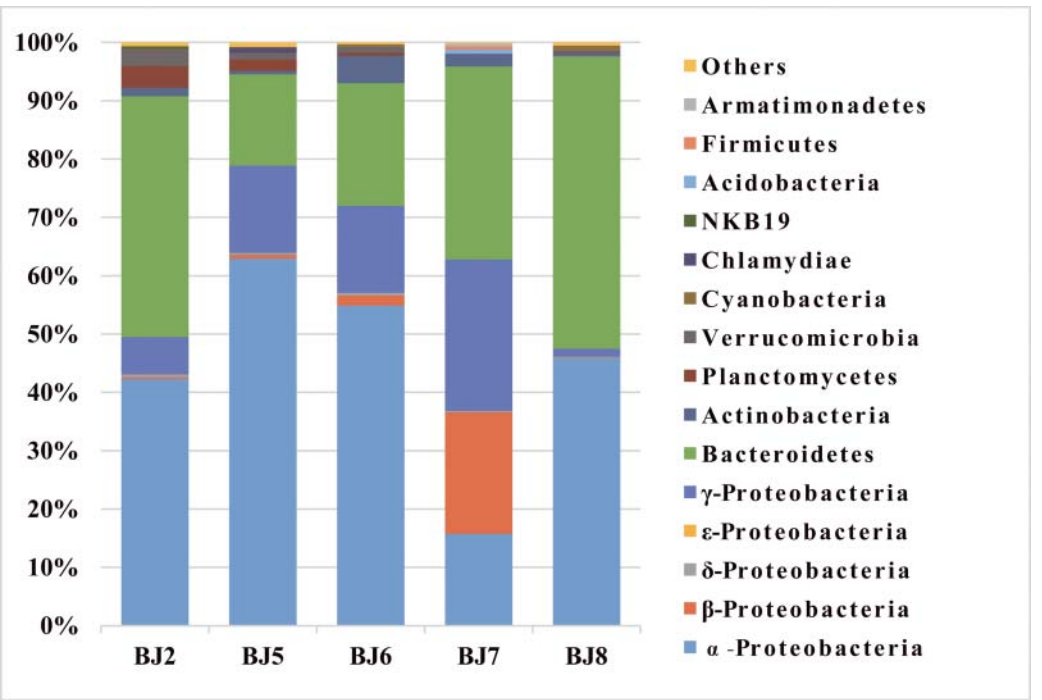

Fig. 3. Composition structure of bacteria from five Arctic seawater sites. Community information to the level of phylum (e.g. Bacteroidetes) and class (e.g. α-Proteobacteria).

Archaeal community structures

We obtained just 37 archaeal OTUs from all qualified reads, i.e. reads after we deleted 2741 OTUs of bacteria and unassigned species. All 37 OTUs belonged to one of three phyla: Crenarchaeota, Euryarchaeota, and Parvarchaeota. All Archaea could be divided into six orders. Table 3 displays the details.

Figure 5 displays the community structure of Archaea at order level. Nitrososphaerales and Halobacteriales dominated. The two orders appeared at all sites, although their

Table 3. Archaeal taxonomic ranks from five sites

Phylum	Class	Order
Crenarchaeota (63.14%)	Thaumarchaeota (63.14%)	Cenarchaeales (1.33%) Nitrososphaerales (61.81%)
Euryarchaeota (34.01%)	Halobacteria (33.32%) Methanobacteria (0.69%)	Halobacteriales (33.32%) Methanobacteriales (0.69%)
Parvarchaeota (2.84%)	Parvarchaea (2.84%)	WCHD3-30 (2.50%) YLA114 (0.34%)

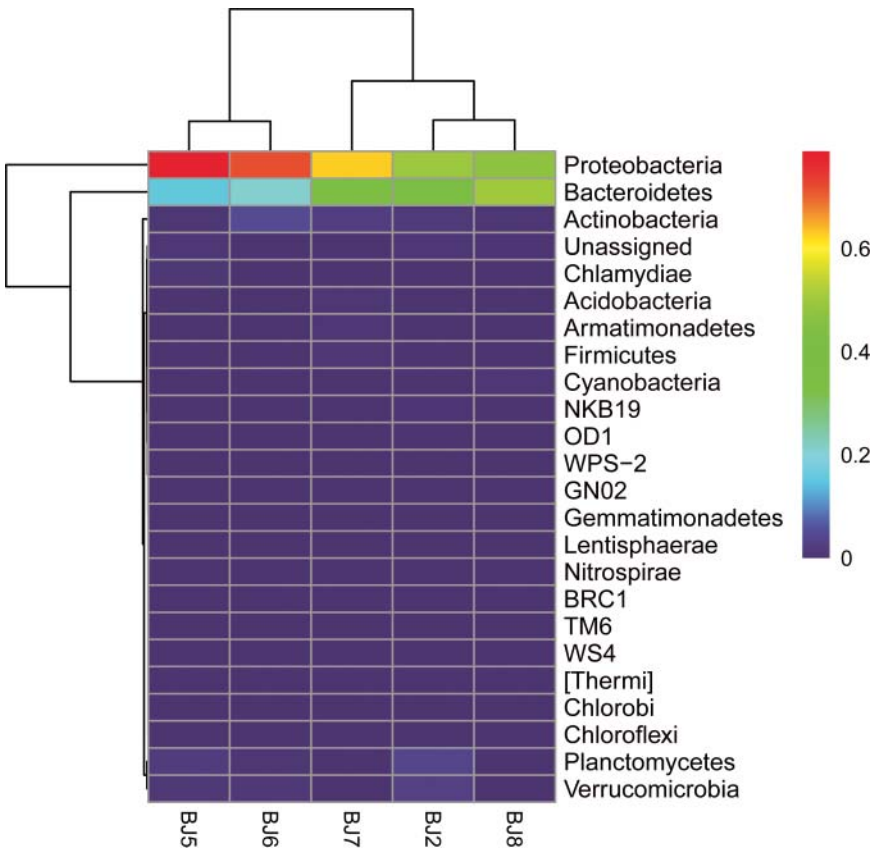

Fig. 4. Heatmap of bacteria at the five sampling sites.

proportional abundances did vary from site to site. Nitrososphaerales predominated at sites BJ2, BJ5, and BJ6. But at BJ8 it formed a much smaller proportion of the Archaea. There, Halobacteriales dominated. At BJ7, Halobacteriales and Nitrososphaerales contributed equally.

Function prediction of microbiome

We further tested and optimized the genome prediction step of PICRUSt using additional information from sequenced reference genomes. The total number of functional genes was 330, with 217 different metabolism-related genes. We selected eight metabolism genes, including carbon cycling (contained genes relating to the carbon fixation pathway in prokaryotes, methane metabolism, starch and sucrose metabolism, and photosynthesis), nitrogen cycling, and other often discussed elements that contribute to biochemical cycling. Copy reads of functional genes relating to carbon cycling and nitrogen cycling were the most abundant (Fig. 6). Methane metabolism had the most reads, followed by carbon fixation pathways in prokaryotes. The fewest reads were obtained for phosphonate and phosphinate metabolism. Among all the sites, BJ8 had the most functional copies and BJ6 had the least.

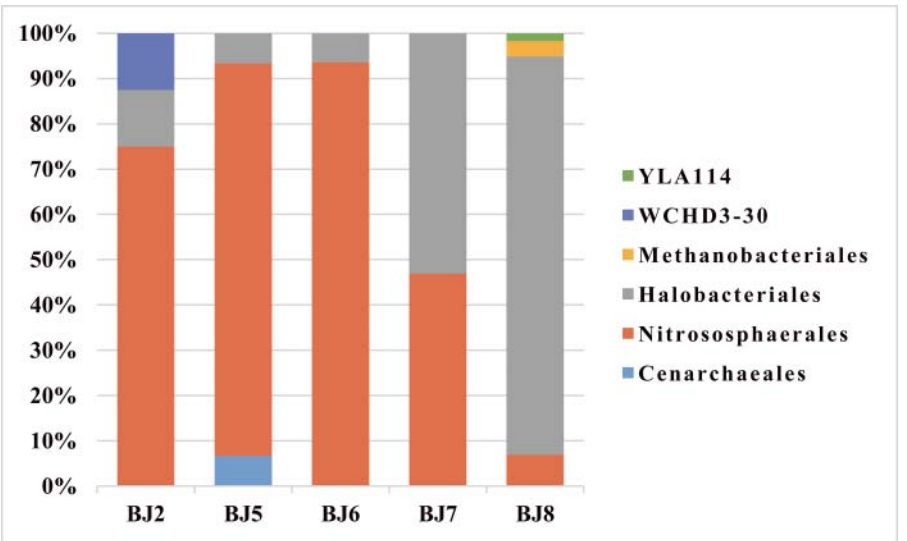

Fig. 5. Composition of Archaea at the five sampling sites.

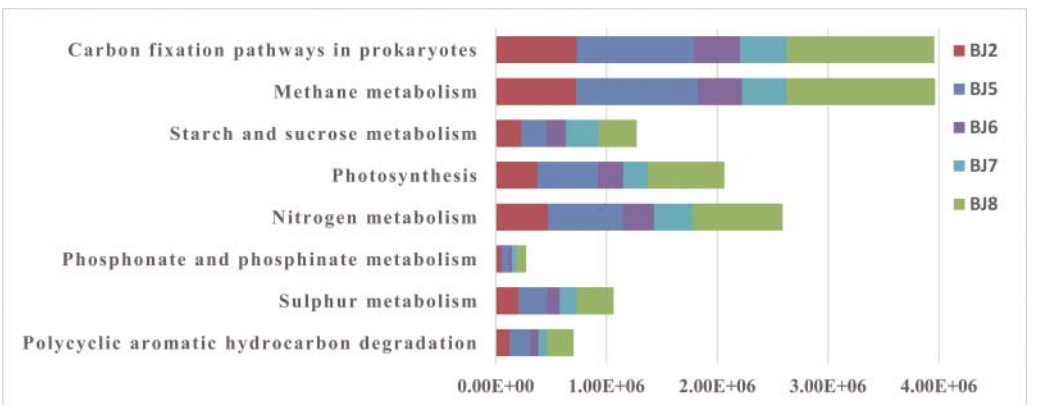

Fig. 6. Microbial gene functions based on 16S rRNA gene sequences related to KEGG pathways.

DISCUSSION

The diversity and community structure of bacteria

Of the bacterial communities, Proteobacteria, Bacteroidetes, and Actinobacteria were the dominant phyla at all five studied sites. Several other papers have also shown these three types of bacteria dominate around the Svalbard Archipelago, although their percentages were found to differ (Górniak *et al.*, 2017; Papale *et al.*, 2017; Sinha *et al.*, 2017a).

In our study, Proteobacteria were found to predominate, with α-Proteobacteria (71.40%) and γ-Proteobacteria (20.5%) most common. Similar results have been reported in Arctic water, ice, and sediments (Han *et al.*, 2015; Soltwedel *et al.*, 2016; Conte *et al.*, 2018). These studies

all indicated that α-Proteobacteria were abundant in many Arctic ecosystems, accounting for more than 50% of the bacterial biomass. This result is not unexpected since α-Proteobacteria are among the most abundant and ecologically relevant marine bacteria (Geng and Belas, 2010). The phylum Proteobacteria has been reported to dominate in oil-contaminated seawater (Gomes *et al.*, 2016; Dong *et al.*, 2018). Proteobacteria (especially the γ-subclass) was identified as the key organism, playing a major role in the degradation of petroleum hydrocarbons (Harayama *et al.*, 2004; Mapelli *et al.*, 2017; Shinde *et al.*, 2017). This raises the possibility that our sampling sites might have been polluted by oil (Vergeynst *et al.*, 2018).

Bacteroidetes and Actinobacteria are also well distributed across our sampling sites. Bacteroidetes included mainly Flavobacteria, Cytophagia, and Sphingobacteria. Flavobacteria are found in a number of aquatic environments. They have several cold-active enzymes (Reddy *et al.*, 2009) and display biodegradative activity (Markúsdóttir *et al.*, 2013). Thus these bacteria offer good prospects for further research. However, the community of Flavobacteria can shift with changes in temperature, light, dissolved organic matter (DOM), and other factors (Bunse and Pinhassi, 2017). Sphingobacteria can produce sphingolipids (Boone *et al.*, 2001; Lindstrom, 2002). This class can be found in multidrug-resistant water (Narcisodarocha and Manaia, 2016) and polluted seawater (Nagaraj *et al.*, 2017). We believe that people have influenced and polluted the Arctic Ocean. Actinobacteria are widely distributed in marine environments, and are believed to be an important source of bioactivators and potential drug compounds (Bull *et al.*, 2005). The appearance of all these species indicates that the sampling regions may have been polluted by humans. Greater attention must be paid to environmental change.

The diversity and community structure of Archaea

In this study, Crenarchaeota was the most abundant archaeal phylum in seawater (accounting for 63.14%), followed by Euryarchaeota (34.01%). This result is similar to that of Galand *et al.* (2008), who found that in the Arctic shelf ecosystem almost all archaeal reads in seawater belonged to the Crenarchaeota. Gantner (2011) found that the clone libraries featured both Euryarchaeota and Crenarchaeota in variable proportions in the Arctic Ocean. Galand *et al.* (2008) showed that these two kinds of Archaea are cosmopolitan. Massana *et al.* (2000) compared the phylogenetic compositions of marine planktonic archaeal populations in different marine provinces. They collected samples from eight different environments at two depths (surface and aphotic zone) and found that most of the archaeal clones were affiliated with one of the two groups of marine Archaea: group I Crenarchaeota and group II Euryarchaeota. Recent studies have identified that characteristic Crenarchaeota might be the most abundant Archaea in the marine environment (Madigan and Martinko, 2005), especially in the cold, deep ocean (Karner *et al.*, 2001). Their ability to oxidize ammonia has major implications for global biogeochemical cycling (Könneke *et al.*, 2005; Wuchter *et al.*, 2006). The numerical and functional importance of Archaea in the ocean demands better understanding of the factors structuring their diversity.

Nitrososphaerales and Halobacteriales are the most prominent orders in cold environments. Makhalanyane *et al.* (2016) found that Nitrososphaerales dominated in both Antarctic and Arctic soils. Nitrososphaerales has a high affinity for concentrations of ammonia and oxygen (Ferrera and Sánchez, 2016). So the appearance of Nitrososphaerales might indicate eutrophic and oxygen-deficient conditions in this region. Meanwhile, according to Antony *et al.* (2016), archaeal sequences in the coastal snow pack are dominated by Halobacteriales.

The presence of Halobacteriales could be a result of low oxygen (Bandekar *et al.*, 2017). Thus the prominence of Nitrososphaerales and Halobacteriales in our samples might indicate anoxic marine environments. We found Euryarchaeota and Crenarchaeota to be dominant in the Arctic Ocean, the most abundant orders being Nitrososphaerales and Halobacteriales. We suspect that these prokaryotes are cold adapted.

Microbial predictive functions and ecology

The copy reads of carbon cycling (including genes relating to the carbon fixation pathway in prokaryotes, methane metabolism, starch and sucrose metabolism, and photosynthesis) were the most abundant. In light of rapid climate change, the carbon cycle in the Arctic Ocean is of global importance. Such processes as regional warming, wind-induced mixing, and higher inputs of terrigenous organic matter perturb the region's carbon cycle in unquantified ways (MacGilchrist *et al.*, 2014). Some studies have shown that terrigenous carbon (Dittmar and Kattner, 2003; Oleg *et al.*, 2012; Harris *et al.*, 2018) and algal blooms (Chen *et al.*, 2017) that occur in the Arctic play important roles in the carbon cycle. So carbon cycle components could be one to two orders of magnitude higher than previously thought. Anthropogenic CO_2 increase results from fossil fuel consumption and land use. The increase causes glaciers to melt and release carbon (Retelletti Brogi *et al.*, 2018), resulting in changes to seawater chemistry (Matsuoka *et al.*, 2017), ocean pH levels (Rérolle *et al.*, 2016), and biological communities (Conte *et al.*, 2018). Based on all this research, it is unsurprising that the number of carbon cycle-related gene copies is so high.

Note that the copies of methane metabolism and carbon fixation in the carbon cycle function were dominant. Methane (CH_4) is a critical atmospheric greenhouse gas. Methane emissions from Arctic environments had been predicted to increase with a warmer climate, thus acting as a positive feedback to global warming (Douglas *et al.*, 2016; Piskozub, 2017). Zhang *et al.* (2014) found that several episodic oil and gas spill events increased surface methane concentration and raised the local methane outgassing rate. Olefeldt *et al.* (2013) showed that rising temperatures lead to increases in methane production and additional carbon release from permafrost-affected soils. All of these activities could increase the methane content, influencing methane metabolism in the ocean. Methane metabolism was also related to microbial communities. Studies have shown that methanogens (Barbier *et al.*, 2012) – such as Methanobacteriales in our study – and methanotrophs (Martineau *et al.*, 2014) – including γ-Proteobacteria, α-Proteobacteria, and Verrucomicrobia in our study – are crucial in methane metabolism. Such results offer an explanation for the predominance of methane metabolism genes.

Prokaryotic carbon fixation was also a major component of the carbon cycle. Carbon fixation utilizes CO_2 through several CO_2 assimilation pathways, converting CO_2 into value-added chemicals or inducing calcium carbonate ($CaCO_3$) precipitation (Hicks *et al.*, 2017). Currently, there are six known pathways of CO_2 fixation (Hügler and Sievert, 2011). It is known that several prokaryotes have the ability to assimilate CO_2, including Proteobacteria, Actinobacteria, Firmicutes, Cyanobacteria, Chloroflexi, Euryarchaeota, and Crenarchaeota. Those prokaryotes have different carbon fixation pathways (Berg, 2011; Håvelsrud *et al.*, 2013; Hicks *et al.*, 2017). Berg (2011) found that some prokaryotes, such as γ-Proteobacteria and Euryarchaeota, have two different autotrophic pathways, presumably giving them an advantage in varying environmental conditions. The Proteobacteria, Actinobacteria, Planctomycetes, and Firmicutes that we detected in this survey have the potential ability

to fix CO_2 via various pathways (Wang and Sun, 2018). Furthermore, some heterotrophic bacteria in the ocean could fix CO_2 through various carboxylation reactions (Moran *et al.*, 2004). A wide variety of bacteria and Archaea might incorporate CO_2 in the dark as part of their metabolism, including chemoautotrophs, heterotrophs, and even the photosynthetic bacteria (Zhou *et al.*, 2017). To date we are yet to discover other carbon pathways in marine ecosystems (Berg *et al.*, 2010; Hügler and Sievert, 2011). All of this knowledge (from our work and previous work) helps to explain the large amount of carbon-fixation pathway copies.

Conclusions

We used high-throughput and PICRUSt approaches to investigate the diversity, abundance, and function of Arctic bacteria and Archaea. Proteobacteria were the dominant taxon in all samples, with α-Proteobacteria and γ-Proteobacteria the most abundant subtypes. The phylum Crenarchaeota was the most abundant archaeal community. The copy reads of functional genes leading to carbon and nitrogen cycling were the most abundant.

ACKNOWLEDGEMENTS

We wish to thank Professor Ri-Xin Wang from Ningbo University and Professor Ge Shi from Zhejiang Ocean University for their help with sample collection. This work was supported financially by the National Natural Science Foundation of China (#31270160).

REFERENCES

ACIA. 2004. *Impacts of a Warming Arctic – Arctic Climate Impact Assessment*. Cambridge: Cambridge University Press.

Antony, R., Sanyal, A., Kapse, N., Dhakephalkar, P.K., Thamban, M. and Nair, S. 2016. Microbial communities associated with Antarctic snow pack and their biogeochemical implications. *Microbiol. Res.*, **192**: 192–202.

Bacelar-Nicolau, P., Nicolau, L.B., Marques, J.C., Morgado, F., Pastorinho, R. and Azeiteiro, U.M. 2003. Bacterioplankton dynamics in the Mondego estuary (Portugal). *Acta Oecol.*, **24**: S67–S75.

Bachy, C., Lopez-Garcia, P., Vereshchaka, A. and Moreira, D. 2011. Diversity and vertical distribution of microbial eukaryotes in the snow, sea ice and seawater near the north pole at the end of the polar night. *Front. Microbiol.*, **2**: 106.

Bandekar, M., Ramaiah, N., Jain, A. and Meena, R.M. 2017. Seasonal and depth-wise variations in bacterial and archaeal groups in the Arabian Sea oxygen minimum zone. *Deep Sea Res. Part II: Topical Stud. Oceanogr*. [https://doi.org/10.1016/j.dsr2.2017.12.015].

Barbier, B.A., Dziduch, I., Liebner, S., Ganzert, L., Lantuit, H., Pollard, W. *et al.* 2012. Methane-cycling communities in a permafrost-affected soil on Herschel Island, Western Canadian Arctic: active layer profiling of mcrA and pmoA genes. *FEMS Microbiol. Ecol.*, **82**: 287–302.

Berg, I.A. 2011. Ecological aspects of the distribution of different autotrophic CO_2 fixation pathways. *Appl. Environ. Microbiol.*, **77**: 1925–1936.

Berg, I.A., Kockelkorn, D., Ramos-Vera, W.H., Say, R.F., Zarzycki, J., Hügler, M. *et al.* 2010. Autotrophic carbon fixation in archaea. *Nat. Rev. Microbiol.*, **8**: 447–460.

Boone, D.R., Castenholz, R.W., Garrity, G.M., Staley, J.T. and Bergey, D.H. 2001. *The Archaea and the Deeply Branching and Phototrophic Bacteria*. Dordrecht: Springer.

Bull, A.T., Stach Jeward, A.C. and Goodfellow, M. 2005. Marine actinobacteria: perspectives, challenges, future directions. *Antonie Van Leeuwenhoek*, **87**: 65–79.

Bunse, C. and Pinhassi, J. 2017. Marine bacterioplankton seasonal succession dynamics. *Trends Microbiol.*, **25**: 494–505.

Caporaso, J.G., Kuczynski, J., Stombaugh, J., Bittinger, K., Bushman, F.D., Costello, E.K. *et al.* 2010. QIIME allows analysis of high-throughput community sequencing data. *Nature Meth.*, **7**: 335.

Chen, M., Nam, S.-I., Kim, J.-H., Kwon, Y.-J., Hong, S., Jung, J. *et al.* 2017. High abundance of protein-like fluorescence in the Amerasian Basin of Arctic Ocean: potential implication of a fall phytoplankton bloom. *Sci. Total Environ.*, **599/600**: 355–363.

Conte, A., Papale, M., Amalfitano, S., Mikkonen, A., Rizzo, C., De Domenico, E. *et al.* 2018. Bacterial community structure along the subtidal sandy sediment belt of a high Arctic fjord (Kongsfjorden, Svalbard Islands). *Sci. Total Environ.*, **619/620**: 203–211.

De Wet, G.A., Balascio, N.L., D'Andrea, W.J., Bakke, J., Bradley, R.S. and Perren, B. 2017. Holocene glacier activity reconstructed from proglacial lake Gjøavatnet on Amsterdamøya, NW Svalbard. *Quat. Sci. Rev.*, **183**: 188–203.

Dittmar, T. and Kattner, G. 2003. The biogeochemistry of the river and shelf ecosystem of the Arctic Ocean: a review. *Mar. Chem.*, **83**: 103–120.

Dong, C., Sheng, H., Wang, W., Zhou, H. and Shao, Z. 2016. Bacterial distribution pattern in the surface sediments distinctive among shelf, slope and basin across the western Arctic Ocean. *Polar Biol.*, **40**: 423–436.

Dong, W.L., Lee, H., Lee, A.H., Kwon, B.O., Khim, J.S., Yim, U.H. *et al.* 2018. Microbial community composition and PAHs removal potential of indigenous bacteria in oil contaminated sediment of Taean coast, Korea. *Environ. Pollut.*, **234**: 503–512.

Douglas, P.M.J., Stolper, D.A., Smith, D.A., Walter Anthony, K.M., Paull, C.K., Dallimore, S. *et al.* 2016. Diverse origins of Arctic and Subarctic methane point source emissions identified with multiply-substituted isotopologues. *Geochim. Cosmochim. Acta*, **188**: 163–188.

Faith, D.P. 1992. Conservation evaluation and phylogenetic diversity. *Biol. Conserv.*, **61**: 1–10.

Ferrera, I. and Sánchez, O. 2016. Insights into microbial diversity in wastewater treatment systems: how far have we come? *Biotechnol. Adv.*, **34**: 790–802.

Galand, P.E., Lovejoy, C., Pouliot, J. and Vincent, W.F. 2008. Heterogeneous archaeal communities in the particle-rich environment of an arctic shelf ecosystem. *J. Mar. Syst.*, **74**: 774–782.

Gantner, S., Andersson, A.F., Alonso-Sáez, L. and Bertilsson, S. 2011. Novel primers for 16S rRNA-based archaeal community analyses in environmental samples. *J. Microbiol. Meth.*, **84**: 12–18.

Gasol, J.M. and Duarte, C.M. 2000. Comparative analyses in aquatic microbial ecology: how far do they go? *FEMS Microbiol. Ecol.*, **31**: 99–106.

Geng, H. and Belas, R. 2010. Molecular mechanisms underlying roseobacter–phytoplankton symbioses. *Curr. Opin. Biotechnol.*, **21**: 332–338.

Gomes, M.B., Gonzales-Limache, E.E., Sousa, S.T.P., Dellagnezze, B.M., Sartoratto, A., Silva, L.C.F. *et al.* 2016. Exploring the potential of halophilic bacteria from oil terminal environments for biosurfactant production and hydrocarbon degradation under high-salinity conditions. *Int. Biodeterior. Biodegr*. [http://dx.doi.org/10.1016/j.ibiod/2016.08.014].

Górniak, D., Marszałek, H., Kwaśniak-Kominek, M., Rzepa, G. and Manecki, M. 2017. Soil formation and initial microbiological activity on a foreland of an Arctic glacier (SW Svalbard). *Appl. Soil Ecol.*, **114**: 34–44.

Han, D., Ha, H.K., Hwang, C.Y., Lee, B.Y., Hur, H.-G. and Lee, Y.K. 2015. Bacterial communities along stratified water columns at the Chukchi Borderland in the western Arctic Ocean. *Deep Sea Res. Part II: Topical Stud. Oceanogr.*, **120**: 52–60.

Harayama, S., Kasai, Y. and Hara, A. 2004. Microbial communities in oil-contaminated seawater [Review]. *Curr. Opin. Biotechnol.*, **15**: 205–214.

Harris, C.M., McTigue, N.D., McClelland, J.W. and Dunton, K.H. 2018. Do high Arctic coastal food webs rely on a terrestrial carbon subsidy? *Food Webs*, **15**: e00081.

Hauptmann, A.L., Stibal, M., Bælum, J., Sicheritz-Pontén, T., Brunak, S., Bowman, J.S. *et al.* 2014. Bacterial diversity in snow on North Pole ice floes. *Extremophiles*, **18**: 945–951.

Håvelsrud, O.E., Haverkamp, T.H.A., Kristensen, T., Jakobsen, K.S. and Rike, A.G. 2013. Metagenomics in CO_2 monitoring. In Proceedings of the International Conference on Greenhouse Gas Technologies 2012 (GHGT-11) (T. Dixon and K. Yamaji, eds.). *Energy Procedia*, **37** (Pt. 5): 4215–4233.

Hicks, N., Vik, U., Taylor, P., Ladoukakis, E., Park, J., Kolisis, F. *et al.* 2017. Using prokaryotes for carbon capture storage. *Trends Biotechnol.*, **35**: 22–32.

Hügler, M. and Sievert, S.M. 2011. Beyond the Calvin cycle: autotrophic carbon fixation in the ocean. *Annu. Rev. Mar. Sci.*, **3**: 261.

Karl, D.M. 2002. Microbiological oceanography: hidden in a sea of microbes. *Nature*, **415**: 590–591.

Karner, M.B., Delong, E.F. and Karl, D.M. 2001. Archaeal dominance in the mesopelagic zone of the Pacific Ocean. *Nature*, **409**: 507–510.

Kasting, J.F. and Siefert, J.L. 2002. Life and the evolution of Earth's atmosphere. *Science*, **296**: 1066.

Könneke, M., Bernhard, A.E., de la Torre, J.R., Walker, C.B., Waterbury, J.B. and Stahl, D.A. 2005. Isolation of an autotrophic ammonia-oxidizing marine archaeon. *Nature*, **437**: 543–546.

Lindstrom, K. 2002. International Committee on Systematics of Prokaryotes: Subcommittee on the taxonomy of Agrobacterium and Rhizobium. Minutes of the meeting, 7 September 2010, Geneva, Switzerland. *Int. J. Syst. Evol. Microbiol.*, **61**: 3089–3093.

MacGilchrist, G.A., Garabato, A.C.N., Tsubouchi, T., Bacon, S., Torres-Valdés, S. and Azetsu-Scott, K. 2014. The Arctic Ocean carbon sink. *Deep-Sea Res. Part I: Oceanogr. Res. Pap.*, **86**: 39–55.

Madigan, M.T. and Martinko, J.M. 2005. *Brock Biology of Microorganisms*, 11th edn. Englewood Cliffs, NJ: Prentice-Hall.

Makhalanyane, T.P., Van Goethem, M.W. and Cowan, D.A. 2016. Microbial diversity and functional capacity in polar soils. *Curr. Opin. Biotechnol.*, **38**: 159–166.

Mapelli, F., Scoma, A., Michoud, G., Aulenta, F., Boon, N., Borin, S. *et al.* 2017. Biotechnologies for marine oil spill cleanup: indissoluble ties with microorganisms. *Trends Biotechnol.*, **35**: 860–870.

Markúsdóttir, M., Heiðmarsson, S., Eyþórsdóttir, A., Magnússon, K.P. and Vilhelmsson, O. 2013. The natural and anthropogenic microbiota of Glerá, a sub-arctic river in northeastern Iceland. *Int. Biodeterior. Biodegr.*, **84**: 192–203.

Martineau, C., Pan, Y., Bodrossy, L., Yergeau, E., Whyte, L.G. and Greer, C.W. 2014. Atmospheric methane oxidizers are present and active in Canadian high Arctic soils. *FEMS Microbiol. Ecol.*, **89**: 257–269.

Masoud, W., Takamiya, M., Vogensen, F.K., Lillevang, S., Alsoud, W.A., Sørensen, S.J. *et al.* 2011. Characterization of bacterial populations in Danish raw milk cheeses made with different starter cultures by denaturating gradient gel electrophoresis and pyrosequencing. *Int. Dairy J.*, **21**: 142–148.

Massana, R., Delong, E.F. and Pedrós-Alió, C. 2000. A few cosmopolitan phylotypes dominate planktonic archaeal assemblages in widely different oceanic provinces. *Appl. Environ. Microbiol.*, **66**: 1777–1787.

Matsuoka, A., Boss, E., Babin, M., Karp-Boss, L., Hafez, M., Chekalyuk, A. *et al.* 2017. Pan-Arctic optical characteristics of colored dissolved organic matter: tracing dissolved organic carbon in changing Arctic waters using satellite ocean color data. *Remote Sens. Environ.*, **200**: 89–101.

Moran, M.A., Buchan, A., González, J.M., Heidelberg, J.F., Whitman, W.B., Kiene, R.P. *et al.* 2004. Genome sequence of *Silicibacter pomeroyi* reveals adaptations to the marine environment. *Nature*, **432**: 910–913.

Nagaraj, V., Skillman, L., Ho, G., Li, D. and Gofton, A. 2017. Characterisation and comparison of bacterial communities on reverse osmosis membranes of a full-scale desalination plant by bacterial 16S rRNA gene metabarcoding. *npj Biofilms Microbiomes*, **3**: 13.

Narcisodarocha, C. and Manaia, C.M. 2016. Multidrug resistance phenotypes are widespread over different bacterial taxonomic groups thriving in surface water. *Sci. Total Environ.*, **563/564**: 1–9.

Oksanen, J., Kindt, R., Legendre, P., O'Hara, B., Simpson, G.L., Sólymos, P. *et al.* 2009. Package 'vegan': Community Ecology package. R package version 1.13-1 [https://www.r-project.org].

Olefeldt, D., Turetsky, M.R., Crill, P.M. and McGuire, A.D. 2013. Environmental and physical controls on northern terrestrial methane emissions across permafrost zones. *Global Change Biol.*, **19**: 589–603.

Oleg, S.P., Viers, J., Dupré, B., Chabaux, F., Gaillardet, J., Audry, S. *et al.* 2012. Biogeochemistry of carbon, major and trace elements in watersheds of northern Eurasia drained to the Arctic Ocean: the change of fluxes, sources and mechanisms under the climate warming prospective. *Comptes rendus – Géoscience*, **344**: 663–677.

Papale, M., Giannarelli, S., Francesconi, S., Di Marco, G., Mikkonen, A., Conte, A. *et al.* 2017. Enrichment, isolation and biodegradation potential of psychrotolerant polychlorinated-biphenyl degrading bacteria from the Kongsfjorden (Svalbard Islands, High Arctic Norway). *Mar. Poll. Bull.*, **114**: 849–859.

Pedrós-Alió, C., Potvin, M. and Lovejoy, C. 2015. Diversity of planktonic microorganisms in the Arctic Ocean. *Progr. Oceanogr.*, **139**: 233–243.

Piquet, M.T., Scheepens, J.F., Bolhuis, H., Wiencke, C. and Buma, A.G.J. 2010. Variability of protistan and bacterial communities in two Arctic fjords (Spitsbergen). *Polar Biol.*, **33**: 1521–1536.

Piquet, M.T., Maat, D.S., Confurius-Guns, V., Sintes, E., Herndl, G.J., Poll, W.H.V.D. *et al.* 2016. Springtime dynamics, productivity and activity of prokaryotes in two Arctic fjords. *Polar Biol.*, **39**: 1749–1763.

Piskozub, J. 2017. Svalbard as a study model of future High Arctic coastal environments in a warming world. *Oceanologia*, **59**: 612–619.

Porter, K.G. and Feig, Y.S. 1980. The use of DAPI for identifying and counting aquatic microflora. *Limnol. Oceanogr.*, **25**: 943–948.

Reddy, P.V.V., Sailaja, B., Kavya, B., Manorama, R.R., Singh, S.M., Srinivas, T.N.R. *et al.* 2009. Bacterial diversity and bioprospecting for cold-active enzymes from culturable bacteria associated with sediment from a melt water stream of Midtre Lovenbreen glacier, an Arctic glacier. *Res. Microbiol.*, **160**: 538–546.

Rérolle, V., Ruiz-Pino, D., Rafizadeh, M., Loucaides, S., Papadimitriou, S., Mowlem, M. *et al.* 2016. Measuring pH in the Arctic Ocean: colorimetric method or SeaFET? *Meth. Oceanogr.*, **17**: 32–49.

Retelletti Brogi, S., Ha, S.-Y., Kim, K., Derrien, M., Lee, Y.K. and Hur, J. 2018. Optical and molecular characterization of dissolved organic matter (DOM) in the Arctic ice core and the underlying seawater (Cambridge Bay, Canada): implication for increased autochthonous DOM during ice melting. *Sci. Total Environ.*, **627**: 802–811.

Rosenzweig, M.L., Donoghue, J., II, Li, Y.M. and Yuan, C. 2010. Estimating species density. *Biological Diversity: Frontiers in Measurement and Assessment* (A. Magurran and B. McGill, eds.), pp. 276–288. Oxford: Oxford University Press.

Shinde, V.L., Suneel, V. and Shenoy, B.D. 2017. Diversity of bacteria and fungi associated with tarballs: recent developments and future prospects. *Mar. Poll. Bull.*, **117**: 28–33.

Sinha, R.K., Krishnan, K.P., Hatha, A.A.M., Rahiman, M., Thresyamma, D.D. and Kerkar, S. 2017a. Diversity of retrievable heterotrophic bacteria in Kongsfjorden, an Arctic fjord. *Braz. J. Microbiol.*, **48**: 51–61.

Sinha, R.K., Krishnan, K.P., Kerkar, S. and Thresyamma, D.D. 2017b. Influence of glacial melt and Atlantic water on bacterioplankton community of Kongsfjorden, an Arctic fjord. *Ecol. Indic.*, **82**: 143–151.

Soltwedel, T., Bauerfeind, E., Bergmann, M., Bracher, A., Budaeva, N., Busch, K. *et al.* 2016. Natural variability or anthropogenically-induced variation? Insights from 15 years of multidisciplinary observations at the arctic marine LTER site HAUSGARTEN. *Ecol. Indic.*, **65**: 89–102.

Thompson, L.R., Sanders, J.G., McDonald, D., Amir, A., Ladau, J., Locey, K.J. *et al.* & Earth Microbiome Project, C. 2017. A communal catalogue reveals Earth's multiscale microbial diversity. *Nature*, **551**: 457–463.

Tortell, P.D., Maldonado, M.T., Granger, J. and Price, N.M. 1999. Marine bacteria and biogeochemical cycling of iron in the oceans. *FEMS Microbiol. Ecol.*, **29**: 1–11.

Urbanski, J.A., Stempniewicz, L., Węsławski, J.M., Dragańskadeja, K., Wochna, A., Goc, M. *et al.* 2017. Subglacial discharges create fluctuating foraging hotspots for sea birds in tidewater glacier bays. *Sci. Rep.*, **7**: 43999.

Vergeynst, L., Wegeberg, S., Aamand, J., Lassen, P., Gosewinkel, U., Fritt-Rasmussen, J. *et al.* 2018. Biodegradation of marine oil spills in the Arctic with a Greenland perspective. *Sci. Total Environ.*, **626**: 1243–1258.

Wang, H.-L. and Sun, L. 2018. Comparative metagenomic analysis of the microbial communities in the surroundings of Iheya north and Iheya ridge hydrothermal fields reveals insights into the survival strategy of microorganisms in deep-sea environments. *J. Mar. Syst.*, **180**: 102–111.

Wang, S.-B., Wang, J.-X., Liu, X.-H., Fan, Y.-P., Jiang, R., Liu, M.-H. *et al.* 2017. Diversity and abundance of bacteria in the surface seawater of the Changjiang Estuary and its adjacent areas. *Evol. Ecol. Res.*, **18**: 515–530.

Wuchter, C., Abbas, B., Coolen, M.J., Herfort, L., Van, B.J., Timmers, P. *et al.* 2006. Archaeal nitrification in the ocean. *Proc. Natl. Acad. Sci. USA*, **103**: 12317–12322.

Xiao, E., Krumins, V., Tang, S., Xiao, T., Ning, Z., Lan, X. *et al.* 2016. Correlating microbial community profiles with geochemical conditions in a watershed heavily contaminated by an antimony tailing pond. *Environ. Pollut.*, **215**: 141–153.

Zengler, K. 2009. Central role of the cell in microbial ecology. *Microbiol. Mol. Biol. Rev.*, **73**: 712–729.

Zhang, Y., Zhao, H.-D., Zhai, W.-D., Zang, K.-P. and Wang, J.-Y. 2014. Enhanced methane emissions from oil and gas exploration areas to the atmosphere: the central Bohai Sea. *Mar. Pollut. Bull.*, **81**: 157–165.

Zhou, W., Liao, J., Guo, Y., Yuan, X., Huang, H., Yuan, T. *et al.* 2017. High dark carbon fixation in the tropical South China Sea. *Cont. Shelf Res.*, **146**.