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Using picoeukaryote communities to indicate the spatial heterogeneity of the Nordic Seas

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ABSTRACT

Picoeukaryotes are an important, diverse and spatially variable component of marine microbial communities. However, little is known of their distribution in response to environmental heterogeneity. In this study, to understand the Nordic Seas picoeukaryotic community, eleven surface samples from different water bodies were collected in June 2015. Archaeplastida, mainly Prasinophyceae, was present in all samples and was the largest component in cold waters, while Rhizaria and Alveoata were most abundant in the samples influenced by warm waters. Multivariate analyses showed that samples could be discriminated into groupings, each with its specific dominant species and community structure could precisely reflect the environmental heterogeneity caused by different water masses. This study details the relationships between the picoeukaryotes and complex currents in the Nordic Seas, and provides insight for application of using picoeukaryotes as indicator in future bioassessment for arctic or boreal waters.

1. Introduction

Single celled eukaryotes in the size range from 0.22 μm to 3 μm are called picoeukaryotes. They are known to be ubiquitous in surface waters of the oceans and dominate protist assemblages of oligotrophic waters (Vaulot et al., 2008; Katja Metfies et al., 2016). Picoeukaryotes have a high biodiversity composed of multiple metabolic types, including phototrophs, phagotrophs, and parasites (de Vargas et al., 2015; Xu et al., 2017), while they are well adapted to harsh polar environmental conditions (Sherr et al., 2003; Kilias et al., 2014a; Katja Metfies et al., 2016). Over the past decade, 18S rDNA based molecular approaches (such as Sanger-based sequencing of clone libraries, 454 pyrosequencing and Illumina MiSeq platform sequencing) have provided broad insights into picoeukaryotic diversity in many areas, including the Arctic Ocean (Lovejoy et al., 2011), sea ice and melt-ponds (Kilias et al., 2014a,b). However, to date, no study has been published on the distribution of picoeukaryotes in the Nordic Seas.

The region between north of the Greenland-Scotland Ridge and south of a Fram Strait-northern Norway transect is defined as the Nordic Seas. They have a complex bathymetry with shallow shelves, deep basins, mid oceanic ridge systems and steep slopes (Drange et al., 2005). In summer Nordic Seas surface waters were affected by both the warm Norwegian Atlantic Current (NwAC), which flows along the Norwegian continental slope to the north, and the cold East Greenland Current (EGC) which flows from Fram Strait to the south (Swift and Aagaard, 1981; Hansen and Østerhus, 2000; Rossby et al., 2009). Due to the dynamic and complex nature of the Nordic Seas currents that is difficult to characterize by spot measurements, comprehensive bioassessment is needed. Although most previous investigations on marine planktonic eukaryotes have shown that they can be used to indicate integrated physicochemical changes in the local environment (Jiang et al., 2011, 2013, 2016; Xu et al., 2016a,b; Liu et al., 2018), picoeukaryotes have the potential to contribute to the interpretation of environmental variations as molecular methods are independent of size and morphological features. Thus, picoeukaryotes have the potential to contribute to the interpretation of environmental variations.

The primary objectives of this study were to: (1) characterize the composition and distribution of picoeukaryotes in the Nordic Seas; (2)

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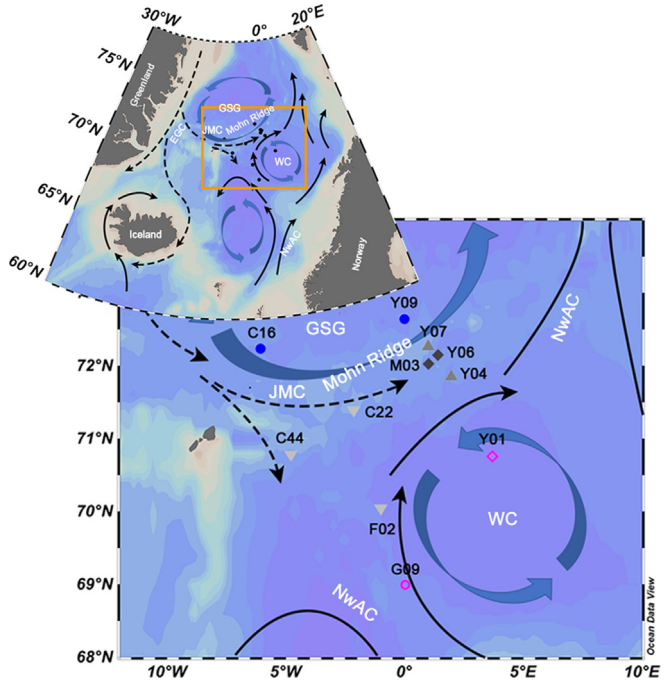

Fig. 1. A schematic illustration of the current system and the position of eleven sampling stations in the Nordic Seas during June 2015. JMC, the Jan Mayen Current; EGC, major branches of the cold East Greenland Current; NwAC, the warm Norwegian Atlantic Current; WC, warm current; GSG, the Greenland Sea Gyre.

investigate linkages between spatial patterns of community structure and the complex Nordic Sea Currents; and (3) reveal the potential for using picoeukaryotic communities as bioindicators of interpreting the environmental heterogeneity in polar marine ecosystems.

2. Materials and methods

2.1. Sample collection

Surface seawater samples (1 L) at 1 m depth were collected from eleven stations in the Nordic Seas during June 2015. These are from the Jan Mayen Current (JMC), major branches of the cold East Greenland Current (EGC), the warm Norwegian Atlantic Current (NwAC), warm current, transitional waters (TW) of the warm and cold currents, and the Greenland Sea Gyre (GSG) (Fig. 1). In Fig. 1, arrows with dot lines indicate the outflow of polar water in the East Greenland Current, while those with solid lines show the warm Atlantic water (Orvik and Niiler, 2002; Blindheim and Rey, 2004). Each water sample was immediately filtered through a 3 μm pore-sized polycarbonate filter followed by a 0.22 μm filter (Whatman, Piscataway, NJ, USA) using a gentle vacuum pump (< 20 cm Hg). Each filter, together with its size-fractionated contents, was carefully placed into a 5 mL tube with 2 mL of cetyltrimethylammonium bromide (CTAB) buffer. The tubes were then quickly frozen with liquid nitrogen and stored at −80 °C for DNA extraction and further study. Seawater temperature and salinity were recorded with a SBE19-CTD profiler.

2.2. DNA extraction and PCR amplification

DNA was extracted in a 1:1 phenol: chloroform mixture (Song et al., 2017). Specific primers with barcode were designed to amplify V4 regions of the 18S SSU rDNA: 3NDF (5′-GGCAAGTCTGGTGCCAG-3′) and V4 (5′-ACGGTATCT(AG)ATC(AG)TCTTCG-3′) (Bråte et al., 2010). PCR reactions were performed in a triplicate 20 μL mixture containing 4 μL of 5 × FastPfu Buffer, 2 μL of 2.5 mM dNTPs, 0.8 μL of each primer (5 mM), 0.4 μL of FastPfu Polymerase (Promega, USA) and 10 ng of template DNA. PCR amplification was performed using an ABI GeneAmp® 9700 under the following conditions: 35 cycles of 95 °C for 30 s, 55 °C for 30 s, 72 °C for 45 s, and a final extension at 72 °C for 10 min.

2.3. High-throughput sequencing

PCR amplicons were checked by 2% agarose gels electrophoresis, purified using the AxyPrep DNA Gel Extraction Kit (Axygen Biosciences, USA) and quantified using QuantiFluorTM -ST (Promega, USA). The concentration of these purified DNA extracts was measured with a Qubit 2.0 fluorometer (Thermo Fisher Scientific Inc., USA). The purified amplicons were then pooled in an equimolar concentration for paired-end sequencing on an Illumina Miseq PE300 platform (http://www.illumina.com.cn/systems/miseq/workflow.aspx). Raw reads in fastq files with low quality (Q < 20 or length < 200 bp) were discarded using QIIME (Version 1.17) (Caporaso et al., 2010). Tags were obtained by merging the paired reads according to their overlaps using COPE (Connecting Overlapped Pair-End, V1.2.3.3) (Liu et al., 2012), after cutting off the sequences of barcodes and primers. High quality pair-wised sequences were obtained following the standards below: (i) bases with ASCII value below 33 were screened out; (ii) a minimum overlap of 19 bp between reads; (iii) no more than one mismatch was accepted while cutting off the sequences of primers. OTU clustering was performed at a minimum sequence similarity of 97% using QIIME (Version 1.8.0) (Caporaso et al., 2010). Chimeric sequences were screened out through UCHIME (Edgar et al., 2011). Each representative OTU after clustering was compared against the Silva (SSU115) 18S rRNA database using a confidence threshold of 70% for taxonomic affiliations. Sequence data generated in this study have been deposited in the NCBI Sequence Read Archive (SRA) database (accession numbers, PRJNA503499).

2.4. Data processing

R software (version 3.2.2, http://cran.r-project.org) was employed to analyse the OTUs dataset of relative abundances. For example, non-parametric species richness ACE, Chao, and diversity indices (Shannon and Simpson) were computed with the "Vegan" package (Dixon, 2003). A heat map of locally abundant OTUs was generated by the "pheatmap" package with corresponding taxonomy to each OTU and abundant OTUs in a given sample refer to OTUs with sequence relative abundance over 1%. (Amaral-Zettler, 2013). Hierarchical clustering and similarity profile permutation tests (SIMPROF) were generated by PRIMER v6.1 (Clarke and Gorley, 2006). The spatial differences in picoeukaryotic communities were summarized using the submodule CAP (canonical analysis of principal coordinates) of PERMANOVA + with Bray-Curtis similarities from log-transformed OTU–abundance data (Clarke and Gorley, 2006; Anderson et al., 2008). PERMANOVA was conducted to test differences among groups (Anderson et al., 2008). Similarity percentage (SIMPER) analysis defines OTUs that contributed to separate each two groups and was conducted among samples in different water bodies (Liu et al., 2018).

Phylogenetic trees were constructed according to the methods reported in Li et al. (2013). Sequences were aligned using MUSCLE (Edgar, 2004). Ends were trimmed and the ambiguous sites were refined manually using BIOEDIT 7.0 (Hall, 1999). The program MrModeltest v. 2 (Nylander, 2004) selected the general time-reversible + invariable sites (=0.4746) + gamma distribution (=0.263) (GTR + I + G) as the best model using the Akaike information criterion, which was then used for both Bayesian inference (BI) analyses. The BI tree was performed with MrBayes 3.1.2 (Ronquist and Huelsenbeck, 2003) using the Markov chain Monte Carlo algorithm. The program was run for 1,000,000 generations, sampling every 100 generations, with the first 2500 trees being discarded as burn-in. Phylogenetic trees were visualized with TreeView v. 1.6.6 (Page, 1996) and

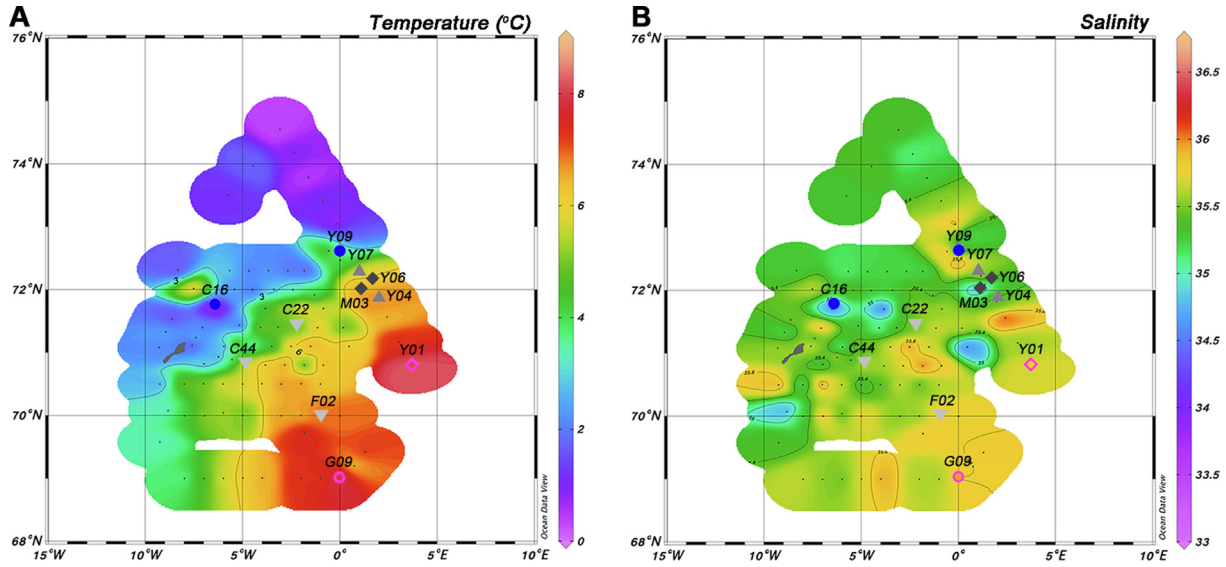

Fig. 2. Surface water temperature (A) and salinity (B) of the Nordic Seas in June 2015.

MEGA 4.0 (Tamura et al., 2007).

3. Results

3.1. Surface temperature and salinity

All samples during the cruise were collected from the Nordic Seas in June 2015. Based on the measurements of temperature and salinity, stations C16 and Y09 were located at GSG, station G09 at NwAC, station Y01 at warm current, stations Y04, Y06, Y07 and M03 at JMC and stations F02, C44 and C22 at transitional waters (Figs. 1 and 2). GSG stations were on average characterized by lowest temperature (1.76 °C) and lowest salinity (34.87). While, stations in JMC and NwAC exhibited higher temperature than it in GSG. Transitional waters properties were intermediate between those of JMC and NwAC. Station in warm current showed the highest temperature (8.20 °C).

3.2. Alpha diversity of picoeukaryote

A total of 388 phylogenetically different OTUs with a mean length of 440 bp were recovered from the 11 samples (97% similarity). The numbers of paired-end sequences in each library varied from 36,233 to 82585. Among the 11 samples, sample Y09 yielded the fewest OTUs (45) while sample F02 had the most (2 2 3) (Table 1). The Shannon diversity index showed different patterns between the various water masses. Shannon diversity index was higher for picoeukaryotic communities collected at the stations in transitional waters. Diversity index Chao and ACE also higher in transitional waters (Table 1).

Table 1
Summary of the estimated operational taxonomic unit (OTU) richness and diversity indices of the 18S rRNA gene libraries for clustering at 97% identity, as obtained from pyrosequencing analysis of the Nordic Seas picoeukaryote communities.

Sample	Sequences	OTUs	ACE	Chao	Shannon	Simpson	Water mass
C16	82,585	60	125	104	0.94	0.67	GSG
C22	37,531	183	196	198	2.24	0.25	TW
C44	77,193	159	189	200	1.53	0.55	TW
F02	36,233	223	244	244	2.78	0.18	TW
G09	70,236	120	201	166	1.08	0.61	NwAC
M03	49,676	82	175	149	2.55	0.14	JMC
Y01	63,876	69	173	145	0.79	0.68	WC
Y04	66,065	90	119	101	0.57	0.78	JMC
Y06	49,953	45	141	145	2.10	0.23	JMC
Y07	41,494	109	103	101	1.31	0.52	JMC
Y09	43,333	108	78	80	0.50	0.81	GSG

3.3. Composition and distribution of picoeukaryotic assemblages

Nine super groups (Alveolates, Stramenopiles, Hacrobia, Archaeplastida, Rhizaria, Opisthokonta, Amoebozoa, Apusozoa and Metazoa) and 30 groups at the class or phylum level (e.g. Prasinophyceae, Dinophyceae, Ciliophora, MASTs, Fungi, Bacillariophyta etc.) were detected in the samples. Relative abundance of OTUs is shown in Fig. 3A and B. At the super group taxonomic level: Alveolata and Archaeplastida appeared in all samples; Archaeplastida dominated in most of samples, such as the samples in GSG (C16, Y09), in JMC-2 (Y04, Y07), and those in transitional waters (C44, C22, F02). Conversely, samples in NwAC (G09), JMC-1 (M03) and warm current (Y01) were dominated by Rhizaria, Alveolata and Metozoa respectively. However, the composition of JMC-1 (Y06) sample was dominated by Archaeplastida, Stramenopiles, Alveolata (Fig. 3A). At the group taxonomic level: Prasinophyceae contributed most to the samples in GSG (C16, Y09), warm current (Y01), JMC-2 (Y04) and transitional waters (C44, C22, F02); Retaria and Dinophyceae respectively were most abundant in NwAC (G09) and JMC-1 (M03); Ciliophora appeared in all samples (Fig. 3B).

3.4. Most abundant OTUs and phylogenetic diversity of picoeukaryotes

Hierarchical clustering (Bray-Curtis similarity index) of the relative abundance of OTUs of the 11 samples is shown in Fig. 4. The clustering result shows that the samples are clustered into six groups. A PERMANOVA analysis showed that there is a significant difference among the six groups (pseudo-F = 4.5684, P = 0.001). Moreover, the SIMPROF result showed that both NwAC (G09) and warm current (Y01) were different from other groups; these are identified by the black lines on the dendrogram (Fig. 4). The other four groups contained their own specific community structures and these branches are shown in red dot lines, indicating that the SIMPROF analysis could find no statistical evidence for any further separation within each group. Notably, the similarities between the sample from NwAC (G09) and the other samples were lower. Locally abundant OTUs (relative abundances > 1% at a given site) are highlighted in the heatmap, and clustered as 13 taxonomic groups (Fig. 4). OTU156 with a BLAST result of *Micromonas pusilla* (100% identity) had the highest relative abundances. Meanwhile

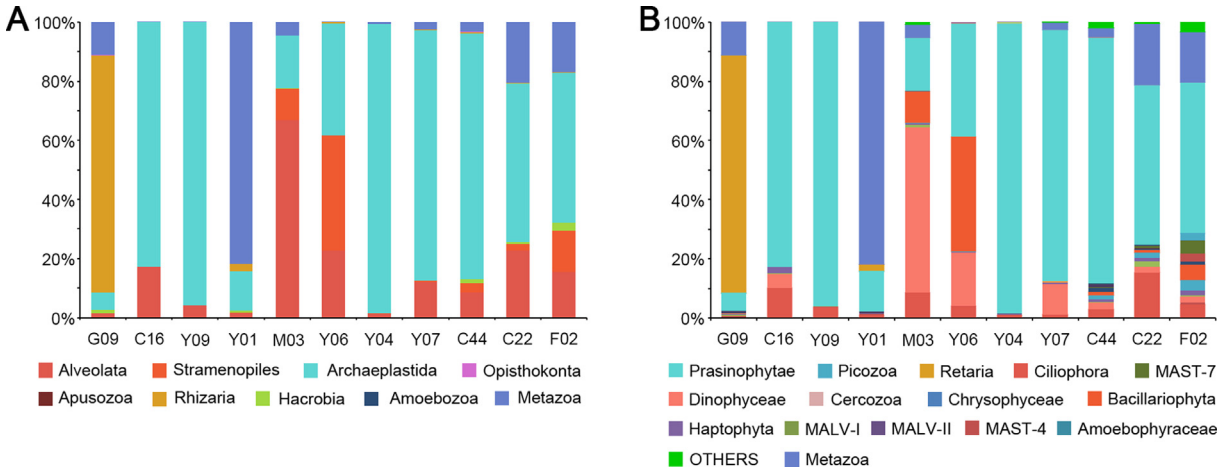

Fig. 3. Relative abundances of picoeukaryotes at the super group (A) and group (B) level for eleven samples. 'Others' includes several lower abundance groups such as Fungi, Labyrinthulomycetes, Pelagomonadales, MASTs, and Phragmoplastophyta.

OTU246 (*Sphaerozoum punctatum*) and OTU140 (*Micromonas* sp.) also had the high relative abundances. Prasinophyceae was widely distributed in all samples. The most abundant OTUs, giving rise to the difference between these five ocean currents, was determined by SIMPER analyses (Table S1). OTU156 (*Micromonas pusilla*) and OTU140 (*Micromonas* sp.) were both presented at all the sites with the former having the higher abundances in transitional waters, warm current, JMC and NwAC, the latter was dominated GSG. While OTU246 (*Sphaerozoum punctatum*) was particularly abundant in NwAC, it was hardly found in the other ocean currents. OTU323 (*Amphidinium*), OTU141 (*Chaetoceros brevis*), OTU270 (*Gyrodinium*), OTU300 (*Chaetoceros*) and OTU362 (Dinophyceae) are the main contributors in sites M03 and Y06, but had lower abundances at other sites.

Based on the phylogenetic analyses of the 30 most abundant OTUs (Fig. 4), it was possible to characterize the phylogenetic diversity of the Nordic Seas. The Ciliophora was the most diverse group in 30 most abundant OTUs. The sequences of Ciliophora have been shown to be abundant in cold water (GSG). Dinophyceae and Bacillariophyta peaked in station M03 and Y06.

3.5. Spatial patterns in picoeukaryotic communities

Discrimination among the 11 samples was plotted using a canonical correspondence analysis of principal coordinates (CAP) using the Bray-Curtis similarities from log-transformed OTU-abundance data (Fig. 5). The plot shows a clear spatial pattern, and the communities could be separated into six groups (Fig. 5). The analysis produced a large first squared canonical correlation ($\delta_1^2 = 0.996$). The first canonical axis separated picoeukaryotic communities in the NwAC (G09), warm current (Y01), and GSG (Y09 and C16) (upper) from communities in the other stations in JMC and transitional waters, while the second canonical axis, which had a similarly high eigenvalue ($\delta_2^2 = 0.992$), discriminated picoeukaryotic communities in the NwAC (G09), warm current (Y01), and JMC-1 (M03 and Y06) (on the left of the plot) from the communities of other stations in GSG, JMC-2 and transitional waters (lower; Fig. 5). A PERMANOVA test demonstrated a significant difference among the six groups (pseudo-$F = 4.5684$, $P = 0.001$).

4. Discussion

So far, some researches have been conducted to monitor environmental variation by using unicellular eukaryotes in temperate or tropical waters (Jiang et al., 2014; Cabello et al., 2016; Pasulka et al., 2016; Zoccarato et al., 2016; Xu et al., 2017, 2018; Liu et al., 2018). However, because of the limitation of sampling in polar or high latitude regions, there is few works focusing on relationship between picoeukaryotes and sea waters. In present study, the response of picoeukaryotic community to the environmental heterogeneity caused by complex ocean currents in the Nordic Seas was investigated using Illumina MiSeq platform sequencing of the 18S rDNA (V4 region). It is still scant for prior investigation of the picoeukaryotic diversity by using molecular approaches in the Nordic Seas (Kilias et al., 2014a). A total of 388 (45 to 223, mean 113) phylogenetically different OTUs (97% similarity), with a mean length of 440 bp, was recovered from the eleven samples. However, it is not possible to compare the data reported herein with previous studies as most previous arctic research have focused on lager nano- and micro-sized eukaryotes (> 2μm) (e.g., Lovejoy, 2014; Kilias et al., 2013; Zoccarato et al., 2016). Kilias et al. (2014a) found that picoeukaryote OTU richness ranged from 164 to 301 (mean 233) in Fram Strait. So, picoeukaryoteic communities in Nodic Seas are not species-poor.

The Nordic Seas, encompassing the Greenland, Iceland and Norwegian, act as a buffer zone between the warm and saline waters of the North Atlantic Ocean, and the cold and fresh waters of the Arctic Ocean. (Jakobsson et al., 2003; Furevik et al., 2007). The Nordic Seas, with their complex topography, water mass distribution and flow regimes, are recognized as a key area for the production of dense bottom water, which has global implications (Furevik et al., 2007). Previous studies have shown that one branch of NwAC tends to the Nordic Seas. This current tends to follow the topographic slope of the Vøring Plateau towards Jan Mayen, and turns northeastward along the slope of the Mohn Ridge. It then turns northwest and continues along the Knipovich Ridge toward Fram Strait (Poulainet et al., 1996, Orvik and Niiler, 2002). In the present study, the picoeukaryotic community structure differed between the samples from stations M03, Y06 and those from Y07, Y04 (Fig. 6). Phylogenetic analysis also showed difference between stations M03, Y06 and stations Y07, Y04. Dinophyceae and all of Bacillariophyta dominated in stations M03 and Y06, while Prasinophyceae dominated in stations Y04 and Y07. This mesoscale structure in the distribution of picoeukaryotic community results from the significant exchange of water masses across the frontal system along Mohn ridge where eddy activities are vigorous, especially in the east side (Raj et al., 2015; Richards and Straneo, 2015). Previous studies have shown the strong relationship between microbiol eukaryotic communities and environmental conditions (Jiang et al., 2013; Song et al., 2017; Xu et al., 2017, 2018; Liu et al., 2018). As regards the research herein, the picoeukaryotic communities around Mohn Ridge showed a clear spatial pattern which could discriminate the complex ocean currents, and proved that they might be used as a robust indicator for understanding hydrological variation.

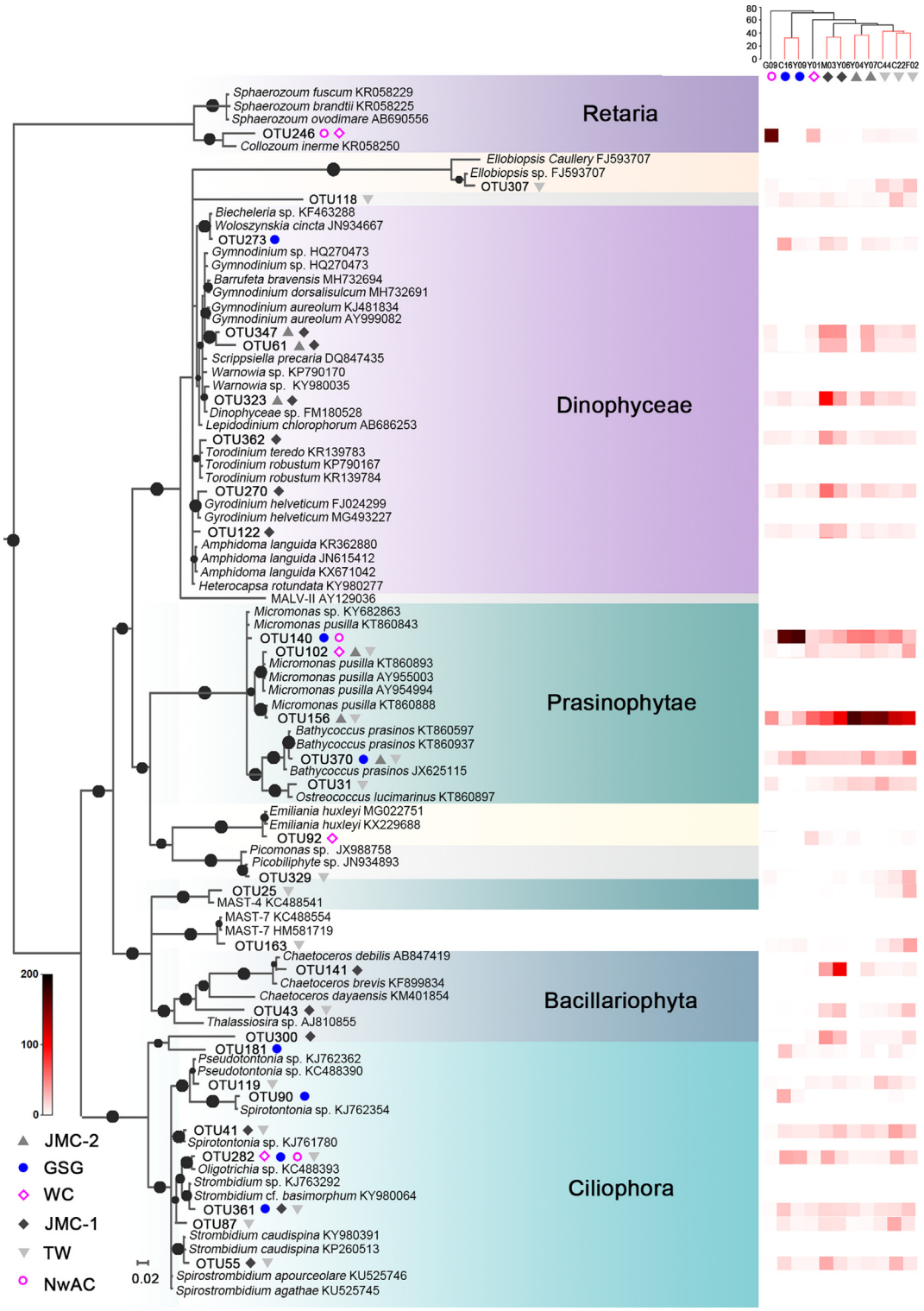

Fig. 4. BI phylogenetic tree of the V4 region of 18S rDNA sequences of top 30 OTUs. BI bootstrap values > 50% (1000 replicates) are shown at the nodes using solid circles. Heatmap of abundant OTUs and hierarchical clustering of the eleven samples are on the right.

The CAP analysis separated the JMC, NwAC, warm current, transitional waters, GSG. In addition, the PERMANOVA analysis provided further support for this finding. All 11 samples from the five regions were clearly separated from each other and the communities within a region were highly similar but significantly different from those in other regions. The dominated water in the JMC is generated from the bifurcation of the EGC and thus is relatively fresh and cold. While the water in the NwAC is advected from further south along the core of Norwegian Atlant Current with a warm and saline feature. That of GSG in the surface mixed layer of Greenland Basin is formed from the mixture of Polar Water and Atlantic Water in the Fram Strait and along EGC. As for warm current in the surface mixed layer of Lofton Basin, it is mainly formed by the cooling of warm Atlantic Water and partly mixed with fresh coastal waters. Within the region of transitional waters, there exists a transitional character in water property, suggesting that water is formed by the process of surface intrusion and mixing

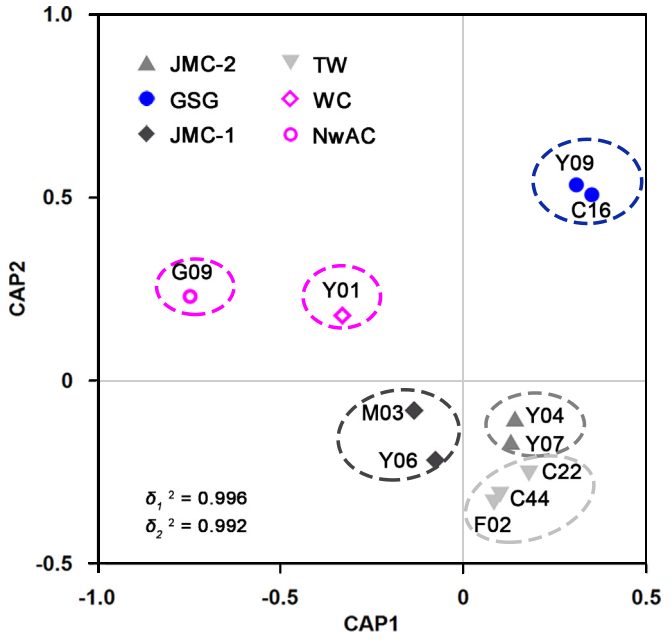

Fig. 5. Canonical analysis of principal coordinates on Bray–Curtis similarities from log-transformed OTU-abundance data of eleven samples in the Nordic Seas, during June 2015.

across the Arctic Front. These results demonstrate that picoeukaryotic community structure differed among regions.

The hierarchical clustering showed the same result as the CAP analysis, revealing that the spatial patterns of the picoeukaryotic communities were consistent with those of the ocean currents. Moreover, the SIMPROF analysis exhibited high similarities between the three transitional waters sites (C22, C44 and F02) and somewhat lower similarities between the JMC sites (M03 and Y06, Y04 and Y07).

The heatmap provided evidence that the samples had different biological properties in the different regions. OTU323 and OTU141 dominated in samples M03 and Y06 respectively. These two sites are located in JMC. OTU323, with an affiliation to *Warnowia* sp., belongs to Dinophyceae. In this study, genus *Warnowia* was only abundant in the cold ocean current, while the warm ocean current was dominated by the heterotroph *Sphaerozoum punctatum* (OTU246), which belongs to the Retaria group. OTU141, from *Chaetoceros brevis*, was the most abundant species in Y06 (JMC-1). A previous study has shown that *C. brevis* is widely distributed in Arctic seas (Gogorev and Samsonov, 2016). OTU156, with a BLAST result of *Micromonas pusilla*, was the most abundant contributor in all cold water samples and has been reported as a cold-adapted ecotype (Lovejoy et al., 2007). And cellular abundance of the prasinophyte *Micromonas* has reportedly increased in the Arctic due to climate-induced changes (van Baren et al., 2016). Previous study showed that the optimum temperature of *Micromonas pusilla* was 2–6 °C (Hoppe et al., 2018). Studies herein, the cold water samples affected by EGC were characterized by low temperatures (0–4 °C). So, we supposed that the lower temperatures might be the main reason why prasinophyte were particularly successful in this region.

The hydrography of this region is characterized by the inflow of warm and saline AW and by the outflow of cold and low salinity Polar Water (PW) via the EGC (Kilias et al., 2013). These differences have the potential to change or explain the observed differences in the protist community's composition and to promote the occurrence of species that are especially adapted to the local environment (Kilias et al., 2014b; Sakshaug and Slagstad, 1991; Li et al., 2009; Tremblay et al., 2009). In this study, picophytoplankton was dominant in transitional waters (stations F02, C44 and C22) which affected by NwAC and EGC. And among five regions, almost all diversity parameters (except Simpson) and sequences had maximum values in the transitional waters stations. The species richness of picoeukaryotes in the transitional waters stations was highest in present study suggesting that in the mixing water in the transition region might supply a better living condition for

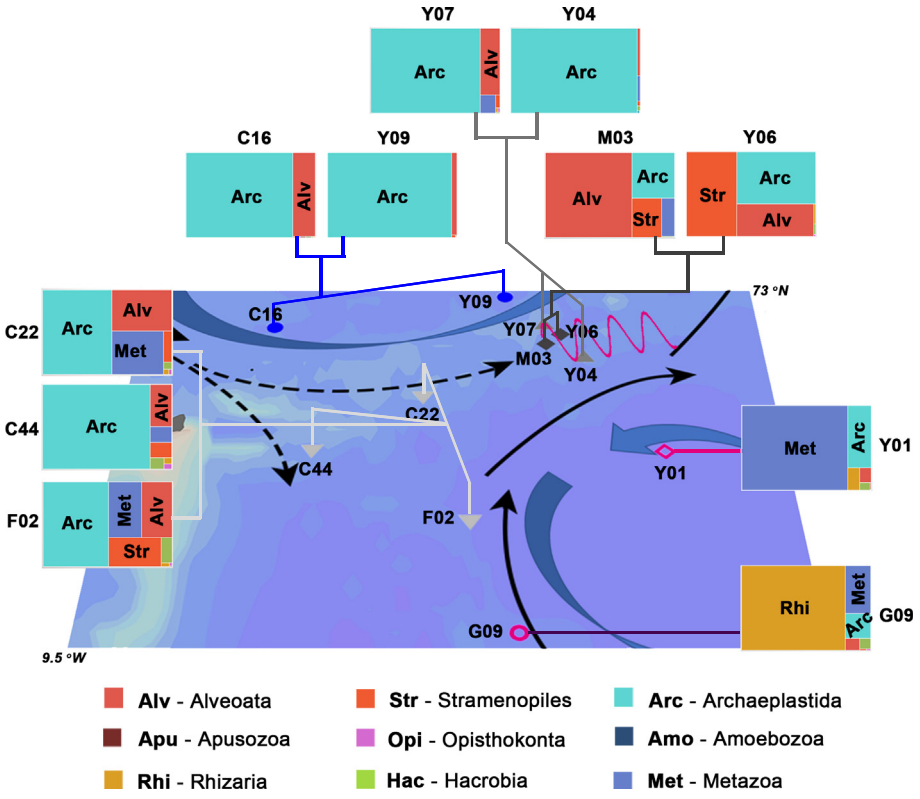

Fig. 6. Map of the stations sampled in Nordic Seas during June 2015, showing the spatial pattern and taxonomic components in picoeukaryote communities.

picoeukaryotes. Previous studies showed that in the vicinity of the polar front and in coastal waters the abundance of picoeukaryotes was higher (Bell and Kalff, 2001; Not et al., 2005). This is probably a consequence of higher nutrient concentrations in these areas (Not et al., 2005). And maybe small cells with faster rates of nutrient uptake and lower metabolic requirements are better able to adapt to changes and hence dominate the Nordic Seas (Kogeler and Rey, 1999; Rat'kova and Wassmann, 2002; Lovejoy et al., 2007).

Marine microbial communities are usually composed of a few of locally abundant species and many rare species (Bowman et al., 2012; de Sousa et al., 2019). The abundant species or groups dominate the community not only by having large numbers of individuals but also by performing the major ecosystem functions (Logares et al, 2014). In this study, a number of abundant groups and species were found that were responsible for much of the differences between the communities and water masses. The super groups SAR (i.e. Stramenopiles, Alveolata and Rhizaria) and Archaeplastida have been widely reported as the dominant picoplankton groups in a variety of surface waters (Massana, 2011). Stramenopiles, which includes diatoms, are bloom formers and are ecological important. Alveolates are subdivided into Ciliophora, Dinophyceae, MALV-I, MALV-II etc. These groups are very abundant in the marine environment. In particular, dinoflagellates within the order Syndiniales have been found in recent molecular surveys to have enormous biodiversity, including parasitic forms (Guillou et al., 2008; Caron et al., 2012; Kilias et al., 2013). Rhizaria are the most recently recognized supergroup of eukaryotes. The three main groups of Rhizaria are Cercozoa, Foraminifera and Radiolaria. Archaeplastida includes land plants, chlorophytes, glaucophytes and red algae, although few of these are single-celled (Caron et al., 2012). In the research herein, Archaeplastida was present in all samples and dominated in most, especially in cold water and mixed water. As in previous studies the picoplankton communities of the Arctic Ocean were mainly dominated by Archaeplastida (Kilias et al., 2014a, c; Metfies et al., 2016). Retaria, which has previously been reported from the surface waters of the Norwegian Sea (Romari and Vaulot, 2004) was most abundant at station G09 (NwAC).

In this study, more than half of the variability in the community composition could be explained by the presence of Prasinophyceae, one green algae related to Archaeplastida. Among the Prasinophyceae, most of the sequences corresponded to two genera *Micromonas* and *Bathycoccus,* both of which belong to the order Mamiellales. *Micromonas pusilla* had the highest relative abundances. Despite the fact that *Micromonas* is considered ubiquitous, only a limited number of field studies have actually recorded its presence, probably because it can only be identified using either living samples or by transmission electron microscopy (Thomsen and Buck, 1998; Romari and Vaulot, 2004). *M. pusilla* has also been found to dominate picophytoeukaryote communities in polar waters (Not et al., 2005; Worden, 2006; Metfies et al., 2016). *Bathycoccus* was initially described from a culture isolated from the bottom of the euphotic zone in the Mediterranean Sea (Eikrem and Throndsen, 1990) and has been little reports since. It has been recorded from the northeastern Atlantic (Johnson and Sieburth, 1982), and from Arctic waters which reached significant abundances at coastal stations and in polar front waters (Not et al., 2005). In the present study, the most number of sequences of *Bathycoccus* were found from station F02, which is in water of the transition region influenced by EGC and NwAC.

5. Conclusions

In summary, the results of this study demonstrate that picoeukaryotic communities in the Nordic Seas are diverse and dominated by Prasinophyceae, Bacillariophyta, and Dinophyceae; the abundance of which varied between water bodies. Meanwhile, the picoeukaryotic community structure showed a clear spatial pattern that could be correlated with ocean currents. The data presented herein provides a better understanding of the spatial distribution of picoeukaryotes in different ocean current trends in the Nordic Seas. And it suggested that the picoeukaryotic communities might be used as an indicator for assessing environmental heterogeneity in arctic or subarctic marine ecosystems.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ecolind.2019.105582.

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