Algal pigments in Hornsund (Svalbard) sediments as biomarkers of Arctic productivity and environmental conditions

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Abstract: Pigments (chloropigments-a and carotenoids) in sediments and macroalgae samples, collected in Hornsund, in July 2015 and July 2016, were analysed (HPLC) in this work. In spite of the aerobic conditions and the periodic intensive solar irradiation in the Arctic environment, neither of which favour pigment preservation in water column and surface sediments, our results indicate that these compounds can provide information about phytoplankton composition, primary production and environmental conditions in this region. The sum of chloropigments-a, a marker of primary production, in the Hornsund sediments varied from 0.40 to 14.97 nmol/g d.w., while the sum of carotenoids ranged from 0.58 to 8.08 nmol/g d.w. Pheophorbides-a and pyropheophorbides-a made up the highest percentage in the sum of chloropigments-a in these sediments, supplying evidence for intensive zooplankton and/or zoobenthos grazing. Among the carotenoids, fucoxanthin and its derivatives (19'-hexanoyloxyfucoxanthin and 19'-hexanoyloxy-4-ketofucoxanthin) contributed the highest percentage, which points to the occurrence mainly of diatoms and/or haptophytes in the water. The pigment markers show that the input of macroalgae to the total biomass could be considerable only in the intertidal zone.

Key words: Arctic, Spitsbergen, pigments, sediments, HPLC.

Introduction

Pigments (chloropigments and carotenoids) are natural compounds widespread in aquatic environments. They occur mainly in phytoplankton, macroalgae and bacteria but also, together with their derivatives, in detritus sinking through the water and settling on the seabed (Jeffrey *et al*. 1997). As a result, sediments may contain a large variety of pigments. The sediment pigments are universal markers of productivity, phytoplankton taxonomy and environmental conditions

in the adjacent water (Louda *et al*. 2002; Bianchi *et al*. 2002; Fietz *et al*. 2007; Freiberg *et al*. 2011; Szymczak-Żyła *et al*. 2011). The sum of chloropigments-a, like β-carotene, is an indicator of primary production (Schüller *et al*. 2013; Szymczak-Żyła *et al*. 2017), while specific chlorophyll-a derivatives are markers of environmental conditions (Szymczak-Żyła *et al*. 2011). As pigments may be degraded to their derivatives by both abiotic and biotic factors, *i.e*. by light, heat, oxygen, herbivore grazing or microorganism activity, these compounds provide information about conditions in the water column and sediments (Welschmeyer and Lorenzen 1985; Bianchi *et al*. 1988; Louda *et al*. 1998, 2002; Szymczak--Żyła *et al*. 2006). Carotenoids together with chlorophylls-c and chlorophyll-b are markers of the main phytoplankton groups (Jeffrey and Vesk 1997).

Pigment records in sediments are the averaged indicators of what has happened over a period of time in a basin (Meyers 1997). Pigments in sediments give an information about changes in past algae blooms, sources of organic matter, ecosystem productivity and environmental, preservation conditions (Meyers 1997; Kowalewska *et al*. 1999). The great advantage of studying pigments in sediments in order to gauge the average annual situation is that samples can be collected *e.g*., once a year or less often. This is in contrast to water, which has to be sampled frequently, since pigment concentrations in water are variable in time and space (Szymczak-Żyła and Kowalewska 2007; Dobrzyn *et al*. 2009). Hence, the analysis of pigments in sediments helps to understand the trends of processes taking place in an environment.

It is known that only about 10% of total productivity leaves the euphotic zone and is transported to sediments (Tribovillard *et al*. 2006). It has been estimated as well that c. 10% of chlorophyll-a produced in water is transferred to the sediments of the Gulf of Gdansk, which is an eutrophic area (Szymczak--Żyła and Kowalewska 2007). The majority of chlorophyll-a is degraded in water column and in uppermost sediment layer to colourless product (Reuss *et al*. 2005). The concentrations of pigments in sediments can be interpreted on the basis of primary production, its sources and environmental conditions in study area, *e.g*., photo-chemical oxidation, organisms grazing or oxygen concentration.

There are numerous papers concerning pigments in the sediments of lakes, open seas, shelf areas and estuaries (*e.g*., Leavitt *et al*. 1997; Chen *et al*. 2001; Reuss *et al*. 2005; Szymczak-Żyła and Kowalewska 2007; Canuel *et al*. 2009; Tani *et al*. 2009; McGowan *et al*. 2012 and references therein). However, only a few authors have studied these compounds in Arctic regions. Spitsbergen fjords could be interesting areas for studying pigments so long as these are not decomposed to colourless products under conditions unfavourable to pigment preservation. Contemporary global warming has advanced, and its effects are more apparent in Arctic than in mid latitudes (Manabe and Stouffer 1980; Morata and Renaud 2008; Węsławski *et al*. 2010; Kay *et al*. 2012; Gervais *et al*. 2016); environmental conditions in high latitudes have been greatly influenced as a result (Overpeck *et*

al. 1997; Post *et al*. 2009; Wassmann *et al*. 2011). The large inflow of freshwater from glacier melting lowers water salinity and increases turbidity, which may in turn affect phytoplankton composition (Wiktor and Wojciechowska 2005). The usual approach of microscopically identifying the phytoplankton composition in Arctic regions, also in the Hornsund fjord (Wiktor and Wojciechowska 2005; Piwosz *et al*. 2009) is time consuming and costly (Paerl *et al*. 2003). Most of the few papers on pigments in Arctic aquatic environments relate to seawater (Dobrzyn *et al*. 2009; Pettersen *et al*. 2011) or lacustrine sediments (Guillizzoni *et al*. 2006; Anderson *et al*. 2008; Jiang *et al*. 2011; Florian *et al*. 2015); those authors detected chloropigments predominantly by fluorometry, not HPLC. The latter method enables the qualitative and quantitative analysis of pigments and their derivatives (Jeffrey and Wright 1997; Morata and Renaud 2008).

The aim of this work was to find out whether algal pigments in the Arctic fjord sediments could be used as biomarkers for these marine environments relating to productivity, phytoplankton composition, sources of organic matter and environmental changes. The fieldwork was carried out in Hornsund, one of the Spitsbergen fjords, which is being impacted by changing environmental conditions and is one of the most thoroughly explored by scientific expeditions.

Study area

Hornsund, a cold-water fjord (mean depth 90 m, max. depth 260 m) on the western side of the southernmost tip of Spitsbergen, is connected with the Greenland Sea (Fig. 1). During summer, Hornsund may be under the influence of warm Atlantic waters (West Spitsbergen Current) or cold Arctic waters (Sørkapp Current) (Swerpel 1985). Hornsund is characterized by different water masses between -1.5°C and 3°C in temperature, and from 34 to 35 in salinity (Cottier *et al*. 2005). The water in the vicinity of the glaciers is stratified as a result of glacial melting and freshwater inflows to the fjord, but there is no stratification nearer the fjord mouth (Swerpel 1985). The volume of freshwater in Hornsund in summer is about 0.79 km^3, but only 40–50% of this water enters the fjord as glacier melt water. The remaining freshwater comes from Arctic Water transported by the East Spitsbergen Current (Beszczyńska-Möller *et al*. 1997). Hornsund has the highest oxygen uptake in sediments of several fjords along Spitsbergen's west coast (Jørgensen *et al*. 2005). The sediment accumulation rate may be as fast as 35 cm/year in the inner part of Hornsund (Brepollen) but only 0.1 cm/year in the outer part (Görlich *et al*. 1987). In the central basin, glacier melting processes release mainly mineral and some organic particles to fjord water and are responsible for high sedimentation rate in this area (~1 cm/yr). High water turbidity reduces the euphotic zone to 5–10 m and as a result primary production is limited there (Wróbel *et al*. 2013). In outer part of the fjord, sedimentation

rates are lower (~0.1 cm/yr), the euphotic zone – deeper (20–55 m) and primary production – higher (Görlich *et al.* 1987; Wróbel *et al.* 2013). The dominant sediments, mostly silt, in Hornsund are very fine-grained and rather homogeneous (Grzelak and Kotwicki 2012; Drewnik *et al.* 2016). The organic carbon content was estimated from 0.8 to 1.8% (Drewnik *et al.* 2016).

Materials and methods

Sample collection. — Sediments were sampled at seven stations in Hornsund (Fig. 1). The sediments at stations H1, H2, H3 were collected in July 2015, those at stations H4, H5, H6, H7 in July 2016. The sampling sites were selected in such a way as to cover a wide range of environmental conditions in the study area, *i.e.* different water depth, salinity, dissolved oxygen levels and distance from the coastline and glaciers. Station H1 was located in the eastern part of Burgerbukta, H7 at the entrance to Burgerbukta, H2 and H6 in Brepollen, the innermost part of Hornsund, and H3 in Isbjørnhamna, near the Polish Polar Station. Station H5 was situated in the central part of the fjord – this was the deepest of the stations. Station H4 was situated in the outer part of Hornsund. The stations were characterized by differences in salinity, temperature and oxygen concentration in the near-bottom water (Table 1). Salinity varied between 33.3 (station H3) and 35.4 (station H6). Near-bottom temperatures ranged from -1.1°C (H2) to 4.2°C (H6), and oxygen concentrations between 11.03 mg/l (H5) and

Fig. 1. Map of Spitsbergen (**A**) and bathymetric map of the Hornsund fjord (**B**) with location of the sampling sites. Glaciers are shown in light grey, while land areas without glaciers are shown in dark grey.

Table 1

Characteristics of the sampling stations.

Station	Coordinates	Collection date	Water depth [m]	Parameters of near-bottom water[*]		
				salinity	temp. [°C]	oxygen [mg/l]
H1	77°02.373' N 16°01.473' E	July 2015	71	34.5	1.6	14.78
H2	77°01.358' N 16°32.041' E	July 2015	92	35.1	-1.1	12.52
H3	77°00.570' N 15°37.319' E	July 2015	50	33.3	3.5	13.39
H4	76°56.098' N 15°21.059' E	July 2016	134	33.8	2.2	12.00
H5	76°58.953' N 15°45.821' E	July 2016	228	34.8	3.0	11.03
H6	77°00.591' N 16°29.263' E	July 2016	165	35.4	4.2	11.55
H7	77°00.696' N 16°00.929' E	July 2016	107	35.0	3.5	12.00

[*]measured with a ProfiLine Multi 197i WTW meter

14.78 mg/l (H1). The sediments were obtained with a GEMAX twin-core sampler during cruises of r/v *Oceania*. The cores were sliced into intervals of 0–1, 1–5, 5–10, 10–15 and 15–20 cm and frozen on board immediately after collection. Surface sediment samples (0–1 cm) were also taken at two locations in the intertidal zone (stations I1 and I2) in July 2015. Macroalgae (*Laminaria digitata*, *Desmaresta aculeata*, *Fucus* sp., *Acrosiphonia* sp.) were collected along the coast of the fjord in the intertidal zone, near the River Revelva, in July 2015. All the samples were stored at -20°C until analysis. The salinity, temperature and oxygen concentration were measured with an SBE19 probe (vertical profiles in the water column) and with a ProfiLine Multi 197i WTW meter (near-bottom water).

Pigment analysis. — Pigments were extracted from the sediment samples according to the procedure used by the Marine Pollution Laboratory of the Institute of Oceanology, Polish Academy of Sciences (Szymczak-Żyła and Kowalewska 2009). A frozen sediment sample (0.5–10 g) was placed in a glass centrifuge tube and left to thaw. Water was removed by centrifugation (6 min, 2500 rpm). Samples were mixed with acetone, sonicated (2–3 min), centrifuged again and the

extract decanted. The extraction was repeated until the supernatant was colourless (max. three times). The pooled acetone extracts were transferred to a separating funnel for liquid–liquid extraction in the acetone extract:benzene:water system (15:1:10 v/v/v). The benzene layer was transferred to a glass vial, evaporated to dryness in a stream of argon, and stored frozen (-20°C) until HPLC analysis. The extracted sediment was dried at 60°C and weighed. The pigment content was calculated per dry sediment weight (nmol/g).

Acetone was added to the frozen macroalgae, which were then crushed with a glass rod. The samples were centrifuged (6 min, 2500 rpm) and injected into the HPLC system. Pigments in macroalgae were determined only qualitatively in order to identify the pigment composition.

Samples dissolved in acetone were injected into a Lichrospher column 100RP-18 (250 × 4 mm, 5 μm, 100 Å; Merck, Germany) through a guard column (Lichrospher 100RP-18 endcapped, 4 mm × 4 mm; Merck, Germany) using an HPLC-DAD system (Knauer, Germany). The HPLC system consisted of an autosampler (Optimas), three pumps (Smartline 100), a degasser (Smartline), column thermostat (Smartline) and detector (DAD-K-2800). Different gradient systems were used for separating chloropigments and carotenoids.

Chloropigments-a, chlorophyll-b and pheophytin-b were separated using a two-solvent (A – acetone, B – 80:20 acetone:water v/v) gradient system, at a flow rate of 1.00 ml/min. The mobile phase and gradient system was according to that in Szymczak-Żyła *et al.* (2008). The chromatograms were recorded at 660 nm and 650 nm.

Chlorophylls-c were analysed using an HPLC system (Knauer, Germany) equipped with a fluorescence detector (FL, RF-20Axs, Shimadzu, Japan). The analysis was carried out in the same two-solvent (A – acetone, B – 80:20 acetone:water v/v) gradient system at a flow rate of 0.5 ml/min. The excitation and emission wavelengths were 440 and 630 nm, respectively. The chlorophylls-c contents were obtained according to the procedure described by Kowalewska *et al.* (1996).

Carotenoid separations were carried out using a mobile phase consisting of three solvents (A – 85:15 methanol: 0.5 M ammonium acetate, aq. v/v, B – 90:10 acetonitrile: water v/v, C – ethyl acetate) in a gradient system and a flow rate of 1.00 ml/min. The mobile phase/gradient system was described in Krajewska *et al.* (2017). The chromatograms were recorded at 450 nm.

Absorption spectra were measured over the 360–750 nm range. The solvents (HPLC grade, VWR International Sp. z o.o.) were filtered and degassed with helium before analysis. Identification of individual pigments was based on the retention times, and absorbance spectra were compared with individual pigment standards (DHI, Denmark). Pigments were quantified according to the method given in Kowalewska *et al.* (1996) and Krajewska *et al.* (2017).

Statistical analysis. — The results were statistically processed using STATISTICA 12.5 software (StatSoft, Poland). Principal component analysis (PCA) was applied with the standardization of chlorophylls-c, chlorophyll-b and additional parameters.

Results and interpretation

Pigment distribution and primary production. — The pigment concentrations in different sediment layers at the fjord stations (H1–H7) and in the surface sediment layer at stations I1 and I2 in the intertidal zone are shown in Fig. 2. The sum of chloropigments-a ($\sum$Chlns-a) in the fjord sediments
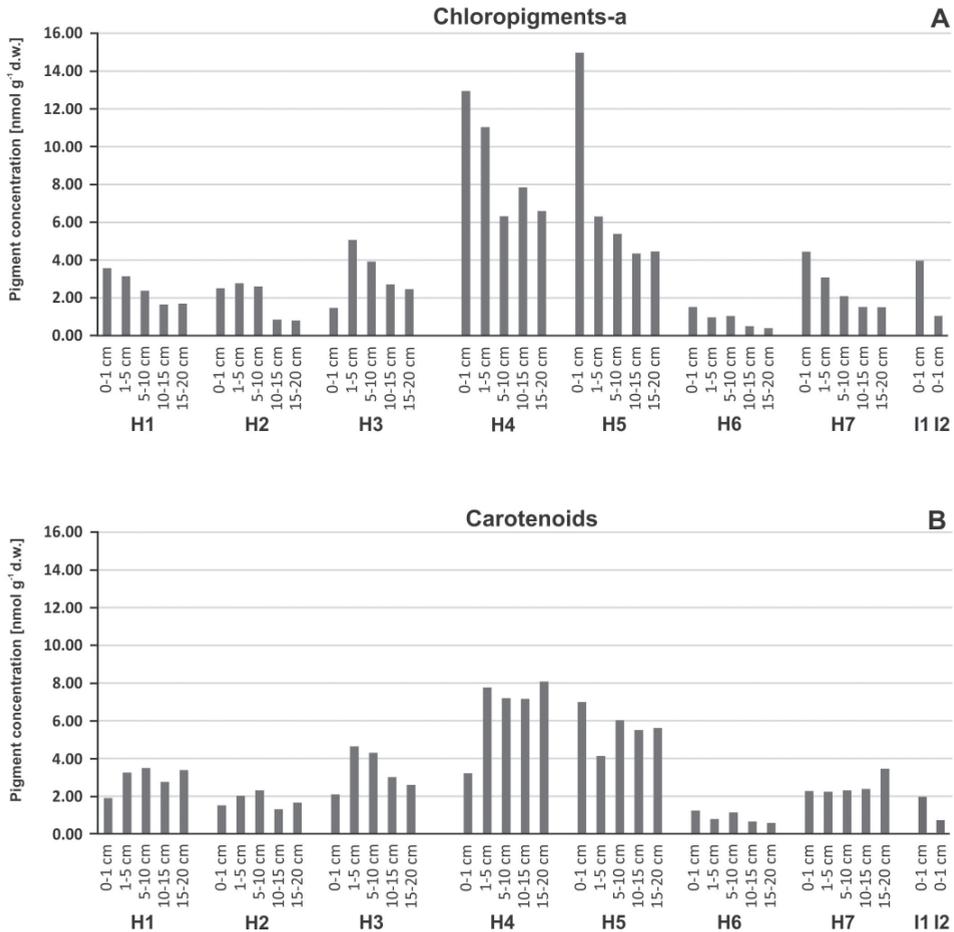
Fig. 2. Contents of the sum of (**A**) chloropigments-a and (**B**) carotenoids.

(H1–H7) varied from 0.4 (station H6, 15–20 cm) to 14.97 nmol/g d.w. (station H5, 0–1 cm), whereas total carotenoids ranged from 0.58 (station H6, 15–20 cm) to 8.08 nmol/ g d.w. (station H4, 15–20 cm). Despite the aerobic conditions in the near-bottom water (Table 1) and the potentially intensive solar irradiation in summer, which can speed up pigment degradation in water column, considerable amounts of pigments are transferred to Hornsund sediments and preserved. Although the concentrations are not high, they are comparable with values obtained for other coastal areas of different latitudes where aerobic conditions also prevailed, *e.g.*, coastal areas of the Gdansk Basin, the Pomeranian Bay, the Venice Lagoon, and the coasts of Scotland (Szymczak-Żyła and Kowalewska 2007; Szymczak-Żyła *et al.* 2011), Trondheimsfjord (Cibic *et al.* 2007) and India (Rasiq *et al.* 2016).

The highest concentrations of chloropigments-a and carotenoids were determined in sediments in the outer part of the fjord (H4, H5) (Fig. 2). The high pigment concentration in these areas might have resulted from a higher level of primary production, which increases from the inner to the outer fjord (from 14.00 to 86.65 mg C m^{-2} h^{-1}) (Piwosz *et al.* 2009) or from low accumulation of mineral particles (Zaborska 2017; Zaborska *et al.* 2017). However, the percentage of undecomposed chlorophyll-a in the surface sediments (0–1 cm) was from 0.5% (H5) to 23% (H3) (Fig. 3). Such a low percentage of chlorophyll-a additionally confirms that ∑Chlns-a is a better marker of primary production than the commonly used chlorophyll-a (Szymczak-Żyła *et al.* 2017).

Environmental conditions. — Pheophorbides-a and pyropheophorbides-a made up the highest percentage among chlorophyll-a derivatives in the sediments at all fjord stations except H3 (Isbjørnhamna) (Fig. 3). Pheophorbides-a are relatively less stable derivatives of chlorophyll-a and indicate the presence of fresh organic matter. Pyropheophorbides-a are markers of zooplankton and/or zoobenthos grazing (Szymczak-Żyła *et al.* 2011). The high percentage of pyropheophorbides-a in the 0–1 cm layer of the Hornsund sediments at the stations in Burgerbukta and Brepollen (H1, H2, H6 and H7) indicates that chlorophyll-a decomposes there mainly as a result of biotic factors, *i.e.* zooplankton/zoobenthos grazing.

Based on results of ^{210}Pb analysis (Zaborska 2017; Zaborska *et al.* 2017, in press), we can suppose that there was no sediment mixing at stations in Burgerbukta (H1), entrance to Burgerbukta (H7), at one station in Brepollen (H6) and in outer part of fjord (H4, H5), which is in agreement with the decreasing profile of ∑Chlns-a (Fig. 2) in sediments and there was a slight mixing at stations H2 and H3. At stations H1, H7 and H6, the high percentage of pyropheophorbides-a suggests intensive zooplankton activity. The majority of zooplankton in Hornsund consists of herbivorous species (Piwosz *et al.* 2009): the copepod *Calanus glacialis*, known to be a key phytoplankton grazer in the
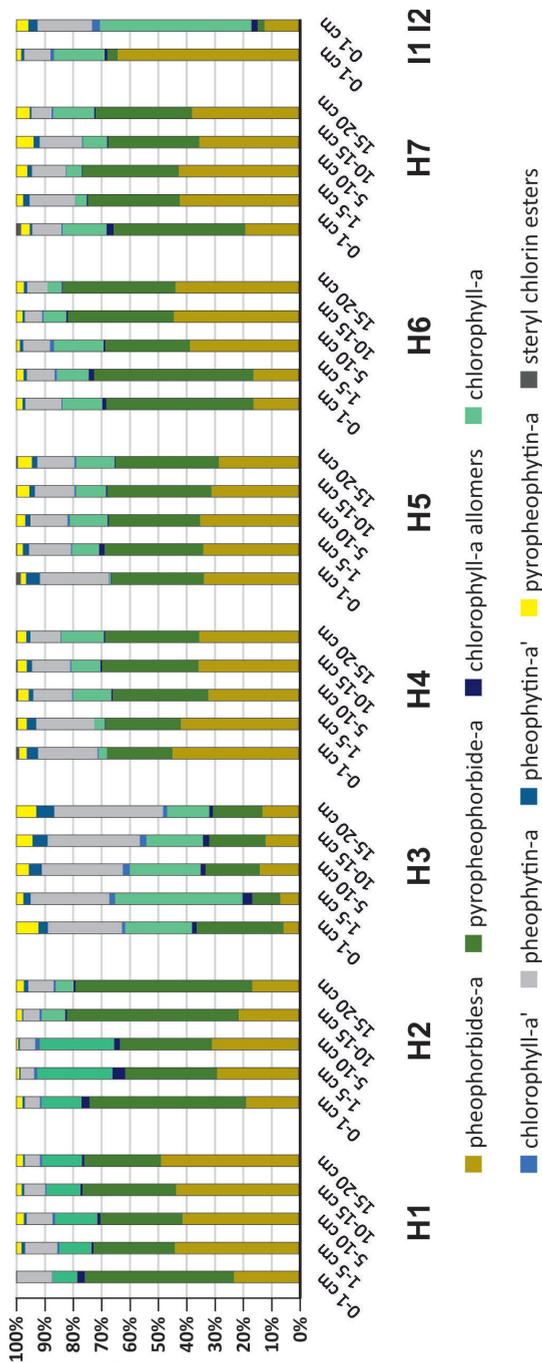
Fig. 3. Percentage of particular chloropigments-a in their sum ($\sum$Chlns-a).

Arctic, is a major contributor to the Hornsund zooplankton (Weydmann *et al*. 2016). The slight mixing (~3 cm) at station H2, stated by Zaborska 2017, is confirmed by our results, *i.e.* the higher concentration of ∑Chlns-a in deeper than in surface sediment layer. The high percentage of pyropheophorbides-a at station H2 suggests that benthic activity is responsible for sediment mixing at this site, which tallies with the information that mixing in the surface sediment layers of Hornsund is caused mainly by bioturbation (Zajączkowski *et al*. 2010; Włodarska-Kowalczuk *et al*. 2013).

The situation at station H3 (Isbjørnhamna) is different. The sediments there contained more undecomposed chlorophyll-a (Fig. 3), *i.e.* up to 45% in the 1–5 cm sediment layer. At this station (H3), the percentage of pheophytin-a, the marker of senescent/slightly decomposed phototrophic matter in sediments, was also high (~30%), indicating that another set of factors was influencing organic matter decomposition. At H3, the sediment was probably mixed (~2 cm) (Zaborska 2017), but contained a smaller percentage of pyropheophorbides-a than sediments from the stations in Burgerbukta and Brepollen (H1, H2, H6 and H7). This suggests that slight mixing at station H3 may be caused by abiotic factors, *e.g*., currents or activity of glaciers. Station H3 was situated near the Hansbreen glacier, so mixing was most likely due to glacier recession. In addition, the small amount of pyropheophorbides-a (a marker of grazing) is linked with the high mortality of zooplankton communities resulting from freshwater outflow from the glacier (Węsławski and Legeżyńska 1998). A high percentage of fucoxanthin, a marker of diatoms, was also recorded at station H3 (Fig. 4). Fucoxanthin contains an epoxy group and is regarded as an unstable carotenoid (Itoh *et al*. 2003; Patoine and Leavitt 2006), as it is highly degraded in sediments (Reuss and Conley 2005). Consequently, the high levels of fucoxanthin and undecomposed chlorophyll-a in the sediments at station H3 indicate that fresh organic matter was present there. Indeed, the 20 cm long core at this station was formed during about 23 years (Zaborska 2017).

The large amounts of pigments at the outer-fjord stations (H4, H5) suggest that conditions there were favourable for pigment preservation in sediments. Station H5 is the deepest one and the oxygen concentration was the lowest of all such values measured in near-bottom water (Table 1). The surface sediment layers (0–1 cm) at stations H4 and H5 contained a high percentage of pheophorbides-a and pheophytin-a, but very small amounts of chlorophyll-a. These sediments differ from those collected at station H3 (Isbjørnhamna) in the percentage of chlorophyll-a and pheophorbides-a, but have a similar pheophytin-a percentage. Based on the accumulation rate, we can estimate that the 0–1 cm layer was formed during about 1 year (H3), 4 years (H5) (Zaborska 2017) and 6 years (H4) (Zaborska *et al*. 2017). The same surface sediment layer at the outer stations (H4 and H5) was formed during a longer period of time than at station H3, which could have caused the decomposition of chlorophyll-a to pheophorbides-a.
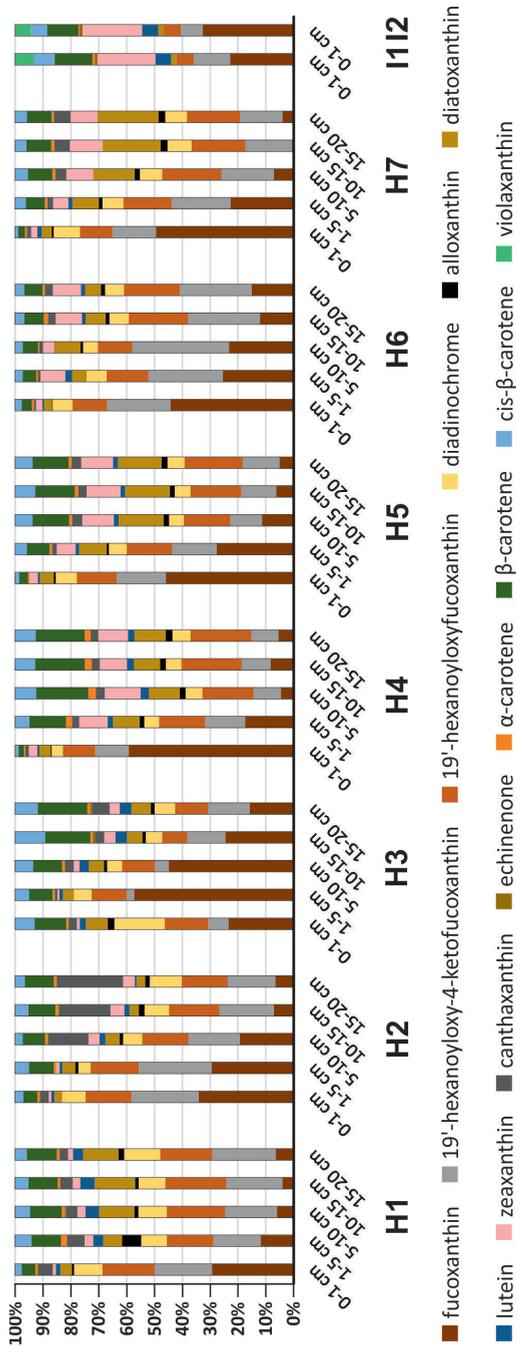
Fig. 4. Percentage of particular carotenoids in their sum.

At stations H4 and H5 grazing was also less intensive than at stations H2, H6 (Brepollen) and H1, H7 (Burgerbukta, entrance to Burgerbukta), which is in agreement with the information that the percentage of herbivorous zooplankton increased from the outer to the inner part of Hornsund (Piwosz *et al.* 2009).

Taking into account the percentage of the most abundant chloropigments-a, carotenoids and standardized parameters, *i.e.* concentrations of chlorophylls-c and chlorophyll-b, as well as additional parameters (station depths, accumulation rates, near-bottom water parameters – salinity, temperature, oxygen concentration) for the Hornsund stations (H1–H7), the results of the statistical analysis show that the stations divide into 4 groups (Fig. 5). This confirms our earlier conclusions, *e.g.*, regarding the exceptionally high amount of undecomposed chlorophyll-a at station H3. The stations in Burgerbukta and Brepollen (H1, H2, H6 and H7) have a high percentage of pyropheophorbides-a and differ from H4 and H5 in the concentrations of chlorophylls-c and chlorophyll-b. Stations H1, H2 differ from H6, H7, for example, in the percentage of fucoxanthin derivatives.

Recent sediments (0–1 cm) were also collected in the intertidal zone (Fig. 1). They differed in the percentages of chlorophyll-a and pheophorbides-a (Fig. 3), but both contained fresh material, because pheophorbides-a are the initial products of chlorophyll-a decomposition under good oxygen conditions.

Phytoplankton taxonomy. — Carotenoid concentrations in sediments give an idea of which phytoplankton species are present. At each station (H1–H7), fucoxanthin and its derivatives (19'-hexanoyloxyfucoxanthin, 19'-hexanoyloxy-4-ketofucoxanthin) were the predominant carotenoids in the sediments (Fig. 4). Analysis of the first 0–1 cm layer (H1, H4-H7) and the carotenoid level in the 0–1 cm and 1-5 cm layers at stations H2 and H3 (averaged due to sediment mixing) indicates that the sum of 19'-hexanoyloxyfucoxanthin and 19'-hexanoyloxy-4-ketofucoxanthin in the sediments at stations H1 (Burgerbukta) and H2 (Brepollen) made up a higher percentage than of fucoxanthin. This is indicative of the presence of haptophytes, since fucoxanthin, 19'-hexanoyloxyfucoxanthin and 19'-hexanoyloxy-4-ketofucoxanthin are haptophyte carotenoid markers (Zapata *et al.* 2004). Wiktor and Wojciechowska (2005) determined haptophytes (*Phaeocystis puchetii*, *Chrysochromulina* sp.) in water from Hornsund.

The percentage of fucoxanthin was higher at stations H3–H7 than that of its derivatives, suggesting that diatoms were the predominant species there. The occurrence of diatoms is also confirmed by the presence of diatoxanthin and diadinochrome in the sediments. Diadinoxanthin, a diatom marker, is an unstable carotenoid, diadinoxanthin is ranked 3 on a four-point scale: 1-most stable, 4-least stable (Leavitt and Hodgson 2001), and was not recorded in the sediments. Instead, diadinochrome was found. Diadinochrome is a rearrangement product of diadinoxanthin, which can occur in some diatoms (Borodina and Ladygina 2013).
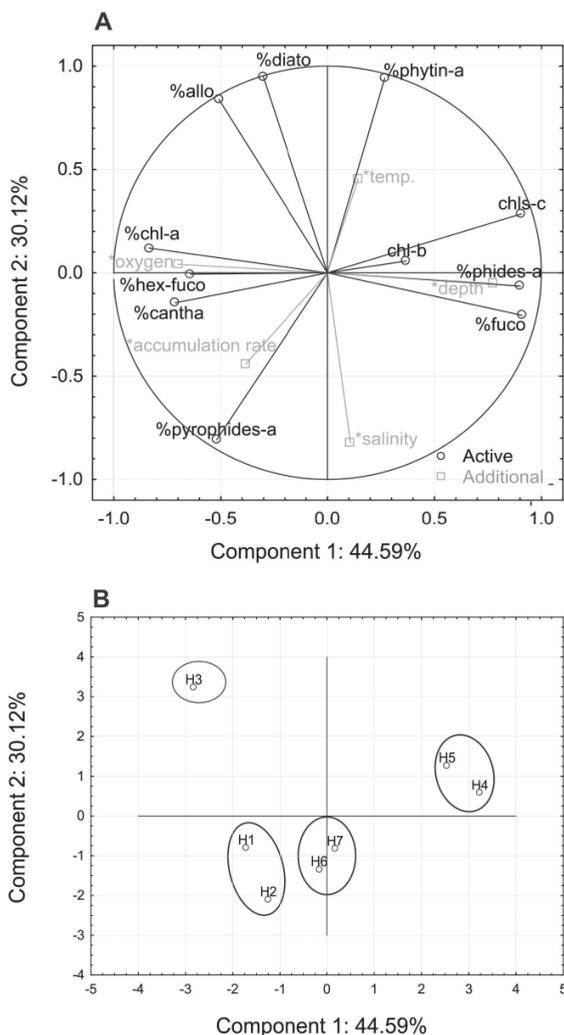
Fig. 5. Results of the statistical analysis: (**A**) scatter plot of principal component loading by individual variables; (**B**) scatter plot of principal component object scores by sampling sites, based on characteristic of the surface (0–1 cm) sediment layer: the percentage of main carotenoids and chloropigments-a, concentration of chlorophyll-b, concentration of chlorophylls-c and as additional parameters: accumulation rates and near-bottom water parameters (salinity, temperature and oxygen concentrations).

Diatoxanthin is more often determined in sediments than diadinoxanthin, because in sediments the latter decomposes rapidly to the former (Reuss *et al*. 2005).

At stations H1 and H2, by contrast, intensive zooplankton grazing might have reduced the abundance of diatoms, the major food item of *Calanus* spp. (Falk-Petersen *et al*. 2002, 2009; Wold *et al*. 2011), so most of the carotenoids

at H1 and H2 derived from nanoplankton, like haptophytes, have been preserved in the sediments.

There was also a high percentage of fucoxanthin at the stations in Brepollen (H6) and at the entrance to Burgerbukta (H7), despite the high level of pheophorbides-a. The considerable fucoxanthin content at station H6 implies that accumulation rate was higher there (~0.66 cm/yr) than at H4 (~0.17 cm/yr) and H5 (~0.23 cm/yr) (Zaborska 2017; Zaborska *et al.* 2017) and that fucoxanthin was not completely decomposed in the water. At station H7, the accumulation rate (0.23 cm/yr, Zaborska *et al.* in press) was not so high as at H6, but despite intensity of zooplankton grazing, primary production of diatoms was also evidently higher.

According to Piwosz *et al.* (2009), diatoms made up the majority of the total biomass in the whole of Hornsund. It was observed that diatoms were present when cold water flows in from the Barents Sea (Wiktor and Wojciechowska 2005). This is in agreement with our results. The concentration of chlorophylls-c, a diatom marker, was higher at stations close to the entrance to Hornsund (H4, H5) than in its inner part (Fig. 6). In Arctic environment, diatoms can originate mainly from pelagic or ice blooms (Leu *et al.* 2015), however organic carbon produced by sea ice autotrophs constitute only from 1 to 12.5% of total primary production in this region (Arrigo *et al.* 2008). Ice algal blooms are often dominated by diatoms, which are the first food sources for sympagic, pelagic and benthic herbivores, after long winter time (Leu *et al.* 2015). Actually, there are no available observation of ice flora in Hornsund. Newetherless, it is known that due to small ice cover in the fjord, especially in recent years in connection with contemporary global warming, ice-associated algae biomass is low (Wiktor and Wojciechowska 2005; Smoła *et al.* 2017). Observations conducted by Muckenhuber *et al.* (2016), in years 2000–2014, show that maximum fast ice cover, *i.e.* sea ice attached to the coastline, exceed about 40% of the fjord area in all studied years, except for 2012 and 2014, when ice season was shorter and the maximum ice cover stayed below 20%. The other potential source of ice diatoms can be ice transported from Barents Sea with the Sørkapp Current (Hegseth 1992; Wiktor and Wojciechowska 2005).

Fucoxanthin may origin also from nanoflagellates, like haptophytes, and these taxa were observed as dominant at some water depths in Hornsund (Piwosz *et al.* 2005). However, these organisms are difficult to identify under the microscope due to their small size and delicate structures (Wiktor and Wojciechowska 2005), while our pigment analysis indicated that they may make up a large proportion of the biomass in Hornsund. Thus, pigment analysis in sediments can facilitate the identification of phytoplankton in this area.

A high percentage of canthaxanthin (almost 24% of the sum of all carotenoids identified) was recorded in the 15–20 cm sediment layer at H2 (Fig. 4). Canthaxanthin is a marker of the nitrogen-fixing cyanobacteria of the order
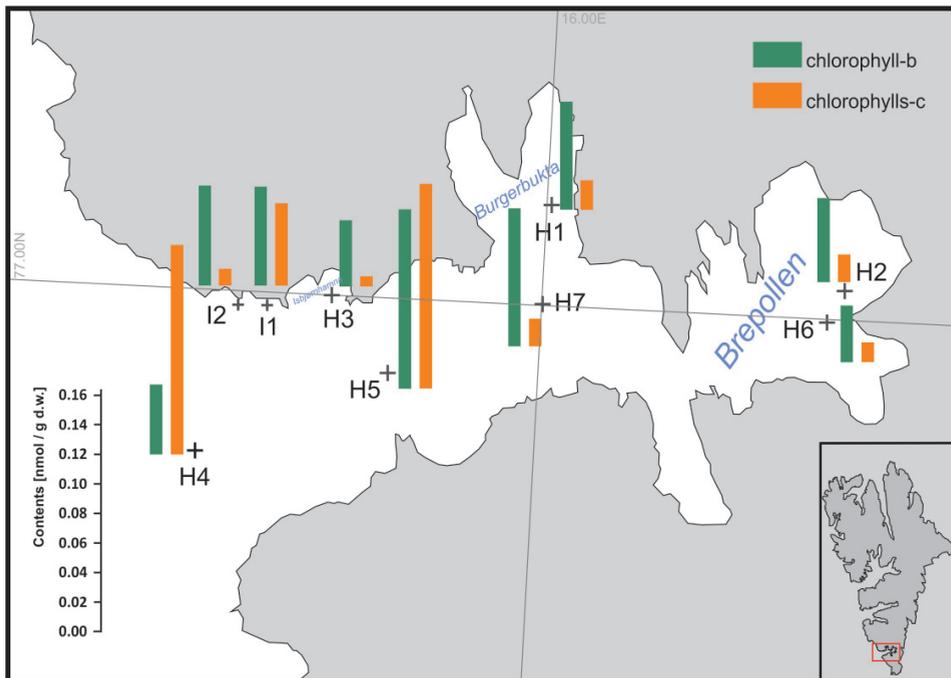
Fig. 6. Contents of chlorophyll-b and chlorophylls-c in 0-1 cm sediment layer.

Nostocales (Desphande *et al*. 2014). In polar environments, cyanobacteria are a major source of nitrogen, as they can fix this gas from the air (Chapin *et al*. 1991; Birkemoe and Liengen 2000; Stewart *et al*. 2011).

Sources of organic matter. — The most abundant pigments in the macroalgae samples (*Laminaria digitata, Desmaresta aculeata, Fucus* sp.) were chlorophylls-c, chlorophyll-a, fucoxanthin, violaxanthin, antheraxanthin, zeaxanthin and β-carotene. The presence of zeaxanthin, violaxanthin and antheraxanthin show that one of the xanthophyll cycles has taken place in macroalgae, a process protecting photoautotrophs against photooxidation (Louda *et al*. 2002). In addition to the above-mentioned pigments, lutein and chlorophyll-b were detected in *Acrosiphonia* sp., a green algae.

Recent sediments (0–1 cm) from the intertidal zone (I1, I2) contained from 0.6 to 1.6 nmol/g of carotenoids (Fig. 2). Besides fucoxanthin and its derivatives, these sediments also contained violaxanthin and more zeaxanthin (Fig. 4) than the sediments from the fjord (H1–H7). This suggests an input from macroalgae, which contain these latter two pigments. The pigments in the intertidal zone sediments probably originated from macroalgae, in contrast to the sediments from the fjord stations (H1–H7), where the organic matter originated mainly from

phytoplankton (diatoms, haptophytes), as described in the previous section. The higher ratio of chlorophyll-b to chlorophylls-c (Fig. 6), at stations closer to the Hornsund shoreline (H1–H3, H6, H7), suggests that some of this organic matter may have terrigenous origin, because glacier melt water contains chlorophyll-b (Aliani *et al*. 2003).

Conclusions

The main results can be summarized as follows:
- the sum of chloropigments-a ($\sum$Chlns-a) is a better marker of primary production in Hornsund than chlorophyll-a,
- macroalgae may be the main source of organic matter only for sediments in the intertidal zone; the organic matter in Hornsund was derived mainly from phytoplankton,
- diatoms and haptophytes constitute the majority of the plant biomass in Hornsund,
- zooplankton grazing is the main factor decomposing organic matter in Hornsund.

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