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## EXPERIMENTAL ARTICLES

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# Microbiological Explorations in the Northern Part of the Barents Sea in Early Winter

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**Abstract**—The total number of microorganisms and rates of microbial processes of the carbon cycle were determined in snow, sea ice, water, and seafloor sediments of the northern part of the Barents Sea from September to October, 1998. The explorations were carried out in two areas: along the transection from Franz Josef Land to Victoria Island and along the continental slope region covered with solid ice at latitude 81°–82° N and longitude 37°–39° E. At the time of study, the ice cover was represented by thick one-year old ice (up to 1.2 m), perennial ice (up to 1.85 m), and pack ice. The number of bacteria in the snow cover, sea ice, and seawater was 12 to 14, 50 to 110, and 10 to 240 × 10<sup>3</sup> cells/ml, respectively. Rates of dark CO<sub>2</sub> assimilation, glucose utilization, and methane oxidation by bacteria were determined. The highest rate of microbial processes was found in samples of the lowermost newly formed sea ice. The lowest level of activity for all processes was observed in melted snow water. A direct relation was shown between the concentration of C<sub>org</sub>, the bacterial biomass, and the values of δ<sup>13</sup>C<sub>org</sub> in mixtures of melted snow and ice. The number of microorganisms and rates of microbial processes in seafloor sediments measured at the stations on the continental slope are comparable to those in the central part of the Barents Sea and the northern part of the Kara Sea.

**Key words:** microbial processes, microbial count, carbon cycle, methane oxidation, sulfate reduction, Barents Sea

The first studies of microorganisms of the Barents Sea are typically attributed to B.L. Issatchenko, who was the first to establish the wide distribution of bacteria in water and seafloor sediments of the arctic seas [1]. Two decades later, from 1928–1931, V.S. Butkevich estimated the number of bacteria in the water of the southeastern and southwestern parts of the Barents Sea. He noted an increased number of bacteria in the region where warm Gulf Stream water combines with cold arctic waters [2]. In the 1970s, studies attempting to evaluate the number of bacterioplankton in the Barents Sea in different seasons were carried out [3]. From 1983 to 1994, works by Baitaz *et al.* [4] in open areas of the Barents Sea determined the number of bacterioplankton as a function of water depth for several water column profiles. The total bacterial biomass was thus evaluated.

Even with the fairly representative body of experimental data regarding the number, total biomass, and cell size of bacterioplankton in the Barents Sea, the rates and regularities of important microbial processes remain virtually uninvestigated.

Of all the arctic seas, the Barents Sea is the best studied. There are, however, two important facts to be noted: (1) most of the data on the Barents Sea relate to

areas south of 80° N latitude and (2) almost all explorations were carried out during short summer seasons. This applies to all processes associated with the inflow of substances into the water column from the atmosphere and to all processes related to algal and bacterial activities.

The following characteristics of biogeochemical processes in the arctic basin distinguish it from most areas of the World Ocean: (1) a large flow of atmospheric aerosol that is washed out from the atmosphere by the snow masses and accumulates on the surface of the ice cover [5]; (2) the occurrence of two extra phase barriers formed at the top and bottom surfaces of the ice cover; and (3) the seasonal variation of all biological processes.

It is evident that all processes involving the activities of microorganisms are strongly influenced by these factors.

Sea ice is related to water and its microflora is to a certain extent derived from seawater microflora. It has been noted by several workers that the bulk of bacteria occurs in the deepest and youngest layers of sea ice in channels filled with nonfreezing salt brine [6, 7] where ice algae, dominated by diatoms, develops. Growth of bacteria is stimulated by organic metabolites excreted

by algae [8]. Brown algobacterial association that developed in one-year-old ice was studied by Grossi *et al.* [9]. The number of bacteria in this ice increased proportionally to the number of diatomic algae in illuminated ice regions. Around 30% of the bacterial biomass was composed of epiphytic bacteria associated with algae. The distinctive features of this bacterial group were larger cells and a faster population growth. There are only a few works showing seasonal variations in the number of bacteria in sea ice. An increase in the number of bacteria in the Arctic Ocean was observed when the population of algae was stable or decreasing. The number and the biomass of the bacteria (45.5 mg C/m<sup>2</sup>) were maximal at the end of the season when the algal biomass decreases [10].

Detailed studies of the bacterial community in pack ice were carried out during the arctic summer voyage of *Polarstern* in 1986. The distribution of microorganisms proved to be extremely uneven and showed large variation. The highest population of bacteria was found in the deepest and youngest ice layers [7].

To understand the details of microbial processes in the carbon cycle, it is important to establish the origin of particulate organic matter of snow, ice, and seawater. It may contain both autochthonous organic matter (represented by phytoplankton and bacterioplankton) and allochthonous matter supplied from the mainland. The suspended matter found in the arctic seas contains large quantities of mineral and organic aeolian material. The organic material is mainly represented by plant tissue fragments, flower pollen, diatomic algae, etc. The organic carbon ( $C_{org}$ ) of this material contains more of the light isotope  $^{12}C$  than marine phytoplankton in mid-latitudes. In arctic regions, the value of  $\delta^{13}C_{org}$  of phytoplankton might vary from -24 to -28‰ [11–13]. The particulate organic matter contained is the substrate for the development of bacterial processes. By measuring  $\delta^{13}C_{org}$  of the suspended matter along with the bacterial biomass and rates of microbial processes, the nature of particulate  $C_{org}$  can be determined.

The objective of this work was to determine the total number and the total biomass of bacteria in the beginning of the winter season, and to quantitatively assess the part played by microbial processes in organic matter synthesis and in the transformations of snow, sea ice, water and modern seafloor sediments.

## MATERIALS AND METHODS

The material for this study was collected during a comprehensive expedition aboard the research vessel *Academician Fedorov* in the Barents Sea from September through October, 1998.

Samples of seafloor sediments were collected with the *Ocean* grab, a boxcorer, and a UT-73 gravity tube. Samples of water were obtained with a 200-l glass bathometer and the Rozette bottles. Samples of silt and water were taken immediately as the seafloor sediments

were lifted aboard the ship. The siet samples were placed in plastic squirts (5 cm<sup>3</sup>) and closed with butyl rubber stoppers. The water samples were placed in glass bottles, allowing extra water to spill over to prevent the capture of air oxygen, and closed with gas-tight rubber stoppers.

All experiments with the sediment and water were performed in the first few hours after sampling at temperatures close to in situ temperatures, ranging between -1.4 and 1.5°C.

To study the potential specific rates of different microbial processes in the snow and ice samples, the samples were melted.

Samples of snow were taken from the surface of the ice with a plastic bucket on the windward side of the ship and placed in large polyethylene bags. To minimize contamination with fuel combustion products from the ship, samples of the uppermost five cm of the snow were taken separately. Snow samples were thawed at 8–10°C in the sample bags, and the resulting water was collected in prewashed containers. As soon as the melting stopped, samples of water to be used for the assay of microbial process rates were poured into 25-ml penicillin bottles, leaving no head space, and sealed with air-tight rubber stoppers.

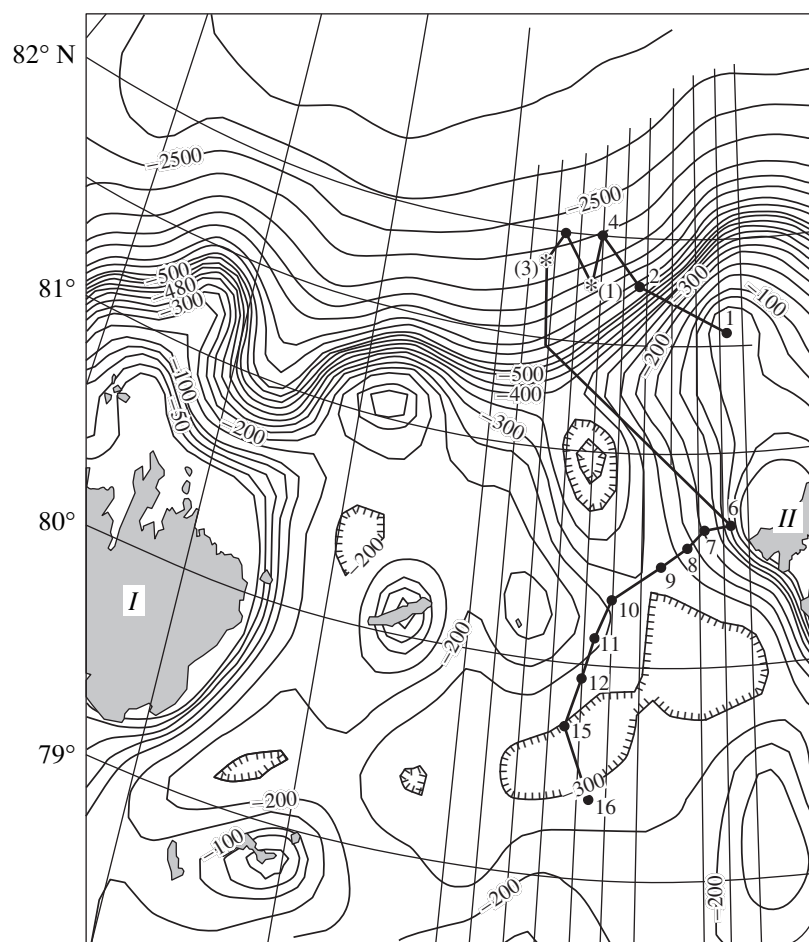
Ice core samples were obtained by the field team using a hand-driven ice bore (core diameter, 180 mm). Ice core samples were transferred to the laboratory in polyethylene bags. The cores were divided into samples from the different horizons (top, middle, and bottom) depending on the measurements of temperatures of the layers in the cores. Each core horizon was melted separately. The ice-melted water was treated the same as the snow-melted water. Samples of both types were incubated at a temperature of -0.5 to -1.4°C in a reservoir filled with seawater on the deck of the ship.

To determine the total concentration of the suspended matter, water was filtered through GF/F 47-mm diameter fiberglass filters that were preliminarily calcined and weighed to an accuracy of  $\pm 0.05$  mg, after which the filters were dried at 60°C.

The number of microorganisms in the water samples was determined by direct counting under a luminescent microscope. A fixed volume (35–50 ml) of the water sample was filtered through lavsan-based filters (made in Dubna, Russia) with 0.19  $\mu$ m a pore diameter that were preliminarily treated with Sudan black. The collected material was fixed with 96% ethanol and stained with a fluorescent dye (acridine orange or DAPI). After staining and drying, the filters were placed in nonfluorescent immersion oil and examined in a LUMAM-3 luminescent microscope at  $\times 900$  magnification. Cells were counted in 20 fields.

The total number of bacteria  $N$  in 1 ml of water was estimated by the following relationship:

$$N = n \times 10^6 \text{ } S \text{ } s^{-1} \text{ } V^{-1},$$



**Fig. 1.** Location of stations where snow, ice and seawater samples were collected. 1–16 mark the stations used to sample seawater and bottom sediments; (1)\*–(3)\* are stations where snow and ice were sampled. I is Svalbard and II is Franz Josef Land.

where  $n$  is the mean number of bacteria in one field of view;  $S$  is the area of the filtering surface,  $\text{mm}^2$ ;  $s$  is the area of the field of view,  $\mu\text{m}^2$ ; and  $V$  is the volume of filtered water, ml.

The rate of methane oxidation was determined radioisotopically in experiments involving short-term sample incubation (12–36 h) with  $^{14}\text{CH}_4$  dissolved in sterile gas-free water [14, 15]. A 0.2-ml portion of labeled methane solution (1–2  $\mu\text{Ci}$  per water or silt sample) was introduced with a syringe. After incubation, the samples were fixed with a 2 M solution of KOH (1 ml) or with Lugol solution (1 ml). Later, in the laboratory, the samples were further processed by the modified method described in [15]. The method determines the amount of carbon dioxide resulting from microbial methane oxidation and the amount of methane carbon incorporated in the bacterial biomass and extracellular organic exometabolites. The rate of microbial production of the organic exometabolites was determined as the difference in values yielded by the complete oxidation of the total organic matter with

potassium (ammonium) persulfate and the biomass deposited on the filters.

The rate of  $\text{CO}_2$  assimilation by the bacteria was determined using sodium bicarbonate containing  $^{14}\text{C}$  (10–20  $\mu\text{Ci}$  per sample) dissolved in sterile seawater. This solution (0.1–0.2 ml) was added to the samples, which were then incubated for 12 to 36 h at close to in situ temperatures. At the end of incubation, the amount of carbon dioxide fixed in the bacterial biomass and the total organic matter were determined by complete oxidation with potassium persulfate [16].

The rate of microbial utilization of  $^{14}\text{C}$ -glucose (the heterotrophic potential) was measured by short-term incubation (12–24 h) at close to in situ temperatures with the addition of 0.1–0.2  $\mu\text{l}$  (5–10  $\mu\text{Ci}$ ) of labeled glucose solution per sample. Then, the amount of glucose carbon, both oxidized to carbon dioxide and fixed in the biomass, was measured.

The rate of sulfate reduction was estimated from the production of hydrogen sulfide, pyrite, and elemental and organic sulfur from  $\text{Na}_2^{35}\text{SO}_4$  (0.1–0.2 ml, 25–50  $\mu\text{Ci}$  per sample) in tests similar to those described above [16].

**Table 1.** Microbiological and biogeochemical characteristics of seawater samples from the water column of the Barents Sea

Station no., horizon, m	Coord- inates	Alk, mg-equiv/l	Total number of bacteria, 10 <sup>3</sup> cells/ml	Bacterial biomass, µg/l	Glucose utilization, µg C/(l day)	Assimilation of CO <sub>2</sub> , µg C/(l day)	CH <sub>4</sub> , nl/l	Oxidation of CH <sub>4</sub> , nl/(l day)
1	2	3	4	5	6	7	8	9
St. 1,	81°34' N							
40	44°18' E	2.6	110	23.1	8.1	25	130	0.3
130		2.6	130	27.3	7.2	23	140	0.08
245		2.6	90	19.9	55	33	150	0.2
St. 2,	81°47' N							
5	40°46' E	2.7	80	16.8		15	120	0.12
25		2.6	20	4.2		14	130	0.2
50		2.7	65	13.7	8.0	11	160	0.07
100		2.8	150	31.5	6.5	14	130	0.08
200		2.8	80	16.8	9.0	14	140	0.04
400		3.0	190	40	8.0	13	160	0.02
St. 4,	82°00' N							
2	38°58' E	2.6	16	3.7	22	22	130	0.02
20		2.6	44	9.2	16	27	160	0.04
120		2.5	70	14.7	44	13	180	0.02
500		2.9	10	2.1	55	22	110	0.07
Near bottom, 750		2.8			76	17	100	0.09
St. 5,	82°00' N							
25	37°33' E	2.6	17	3.6	60	18	150	0.01
40		2.5	13	2.7	14	10	150	0.08
100		2.8	50	11	27	19	160	0.3
Near bottom, 1050		2.9	80	16.8	9.3	10	170	0.01
St. 6,	80°38' N							
25	44°26' E	2.6	34	7.1	21	21	70	0.06
50		2.6	70	14.7	29	16	80	0.06
275		3.0	200	42	25	22	50	0.07
St. 8,	80°28' N							
Surface	42°08' E	2.6	20	4.2	49	20	80	0.08
15		2.5	10	2.1	9.0	18	80	0.09
100		2.6	15	3.2	49	21	90	0.07
Near bottom, 150		2.9	100	21	30	22	100	0.07
St. 9,	80°24' N							
Surface	41°34' E	2.6	20	4.2	8.0	22	90	0.02
20		2.5	20	4.2	7.0	19	100	0.28
50		2.6	30	6.3	14	16	100	0.14
170		2.9	10	2.1	15	6.8	90	0.08
Near bottom, 340		2.9	16	3.4	18	4.4	110	0.02
St. 10,	80°23' N							
Surface	40°30' E	2.6	105	22.1	46	12	100	0.01
25		2.6	95	20	17	22	110	0.02
90		2.6	120	25.2	15	26	120	0.2
180		2.9	80	16.8	27	21	120	0.16
Near bottom, 250		2.9			90	23	120	0.08

**Table 1.** (Contd.)

1	2	3	4	5	6	7	8	9
St. 11,	80°19' N							
Surface	39°59' E	2.6	240	50.4	9.2	11	100	0.18
20		2.5	90	19	19	14	110	0.25
100		2.9	90	19	17	8.4	110	0.25
Near bottom, 250		2.8	105	22.1	35	18	120	0.16
St. 12,	80°17' N							
Surface	39°21' E	2.6	90	19	24	16	100	0.1
25		2.5	65	13.7	48	21	90	0.06
40		2.6	70	16	14	12	110	0.14
60		2.7	85	18	32	12	120	0.25
Near bottom		2.8	90	19	30.5	7.8	120	0.02
St. 15,	79°39' N							
Surface	38°38' E	2.6	10	2.1	9.6	16	100	0.15
60		2.7	13	2.7	30	15	130	0.23
100		2.8	90	19	16	15	130	0.13
180		2.9	10	2.1	14.3	20	150	0.1
Near bottom, 310		2.9	20	4.2	35.5	26	100	0.23

**Table 2.** Microbiological and biogeochemical characteristics of snow and ice samples

Station no. and coordinates	Sample source	Total number of bacteria, 10 <sup>3</sup> cells/ml	Bacterial bio-mass, µg/l	Content of CH <sub>4</sub> , nl/l	Oxidation of CH <sub>4</sub> , nl/(l day)	CO <sub>2</sub> -assimilation, µg C/(l day)	Utilization of glucose, µg C/(l day)
St. 1, 81°47' N 40°46' E	Snow-1	14	4.9	80	0.88	3	64.6
St. 2, 82°01' N 39°05' E	Snow-2	12	4.2			5	
St. 2, 82°01' N 39°05' E	Ice, tube no. 7, topmost 60 cm	80	28	80	0.18	13	67
	Ice, tube no. 7; 60 cm in center part	70	26	80	0.56	20	78
	Ice, tube no. 7, bottommost 60 cm	110	39	80	2.52	45	115
St. 3, 82°00' N 37°33' E	Ice-3, topmost 85 cm	50	18	80	0.79	8	207
	Ice-3, bottommost 85 cm	100	36	80	1.1	30	108

The radioactivity of fixed samples was determined with a RacBeta liquid scintillation counter (LKB, Sweden). Rates of microbial processes were calculated using the relationship previously described [15].

To determine the isotopic composition of C<sub>org</sub>, samples were heated and treated with 8% HCl to remove carbonates and then burnt in a vacuum circulation

apparatus in a quartz tube at 900°C. The CO<sub>2</sub> formed in this process was sealed in ampoules. The C<sub>org</sub> content of the suspension was determined chromatographically from the amount of CO<sub>2</sub> evolved. The isotopic composition of carbon was measured on a MI-1201B mass spectrometer equipped with a three channel gas input system (SNG-3). The measurement error was ±0.5%.

**Table 3.** Biogeochemical characteristics of suspended matter and  $\delta^{13}\text{C}_{\text{org}}$  in snow, sea ice and water column samples

Station no., depth, m	Suspended matter content, mg/l	$\text{C}_{\text{org}}$ content of suspension, $\mu\text{g C/l}$	$\delta^{13}\text{C}_{\text{org}}$ , ‰
Snow samples			
Snow 1	2.6	49	–25.4
Snow 2	2.8	41	–24.7
Snow 3	4.3	55	–25.8
Ice samples			
Ice 1, top	9.0	220	–24.5
Ice 2, bottom	4.5	290	–23.8
Ice 3, top	4.0	189	–24.6
Ice 3, bottom	3.4	310	–24.1
Seawater samples			
St. 1, 130	1.45	66	–24.9
St. 2, 100	1.05	54	–23.2
St. 2, under ice	2.0	160	–22.6
St. 2, 400	0.90	35	–24.6
St. 5, 100	1.21	54	–24.7
St. 5, 1050	1.41	90	–21.9
St. 6, 25	1.40	60	–24.3
St. 6, 50	1.49	50	–24.3
St. 6, 275	1.53	50	–24.9
St. 8, 15	1.00	69	–23.2
St. 8, 100	1.02	57	–23.8
St. 8, 150	1.05	49	–23.4
St. 10, 25	1.09	77	–24.7
St. 10, 90	1.11	97	–25.3
St. 10, 250	1.28	71	–25.5
St. 12, near surface	1.03	109	–24.3
St. 12, near sea floor	1.13	97	–24.9
St. 15, near surface	1.69	60	–25.5
St. 15, 100	1.19	103	–25.6
St. 15, 310	1.20	81	–24.1

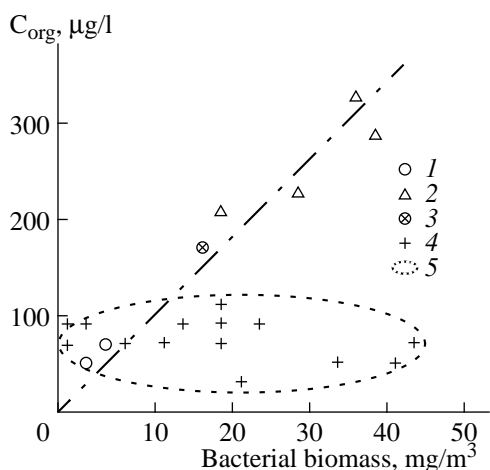
To determine the concentration of methane in the samples, water and silt samples were placed into Balch tubes with a fixed head space. Methane was determined by the phase equilibrium degassing of water and silt samples on a Khrom-5 gas chromatograph with a flame ionization detector.

#### *Brief Description of the Exploration Region*

The explorations were carried out in two areas: along the transection from Franz Josef Land to Victoria Island and in the ice-covered continental slope region of the Barents sea at  $81^{\circ}$ – $82^{\circ}$  N latitude and  $37^{\circ}$ – $39^{\circ}$  E

longitude (Fig. 1). This region is partially free of ice during the short summer season and is typically covered with solid ice during the rest of the year. At the time of our studies, the ice cover was represented by thick one-year-old ice (up to 1.2 m), perennial ice (up to 1.85 m), and pack ice. The snow cover consisted of fresh snow and snow carried over by snowstorms. At the sample sites, it did not exceed 25 cm in depth.

Hydrological and hydrochemical conditions in the water column varied considerably. Mixing of warm Atlantic waters, present at depths from 80 to 200 m, with cold Arctic waters gives rise to variations in water density and considerable vertical stratification. The



**Fig. 2.** Relation between  $C_{org}$  suspended in melt water from snow and ice and the total bacterial biomass in the exploration region at lat.  $81^{\circ}30' - 82^{\circ} N$ : (1) snow; (2) ice; (3) water column under ice; (4) water column; (5) values for water column south of lat.  $81^{\circ} N$ .

seawater salinity is also not constant. This is caused by the concentration of brines during ice formation and their submersion in the water column. The temperature of seawater was minimal,  $-1.7$  to  $-1.8^{\circ}C$ , in the layer just under the ice, and increased gradually with depth to  $1.8^{\circ}C$  at 50–200 m, and then dropped down again at the seafloor horizons.

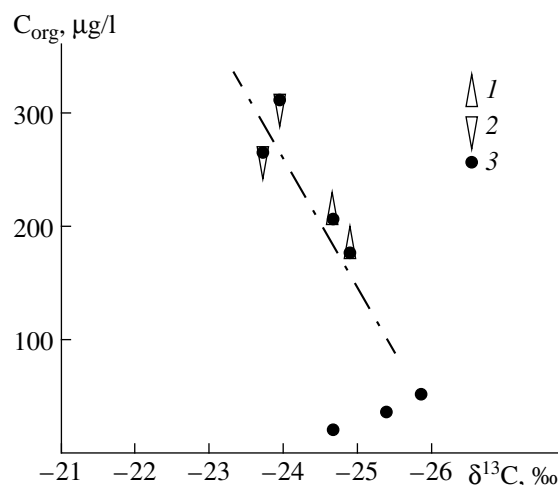
In the surface horizon, the obtained sediment columns were represented by semiliquid pelitic silt. From the surface to a depth of 0.4 to 0.6 m, the sediment was oxidized,  $Eh = +100$  to  $+400$  mV. Over 0.5 m, the redox potential of the sediment was close to zero, and in some silt regions, even slightly negative. The underlying layers contained sand, fine gravel, streaks of siltstone, and inclusions of hydrotroilite.

## RESULTS AND DISCUSSION

### *Abundance and Biomass of Bacteria*

The number and the biomass of bacterioplankton along the transection from Franz Josef Land to Victoria Island were determined at seven stations (nos. 6–15, Table 1). Samples were taken and bacteria were counted from three to five horizons. The data show considerable variation between stations and at different depths. For most of the stations, the highest population numbers were found in the near-surface and near-seafloor samples, except for stations 10 and 15, at which the local population maximum occurred at the 90 and 100 m horizons. These are likely to be the boundaries between water layers with different temperatures and salinities.

The number of bacteria in the seawater samples in the northern exploration area (st. 1–5, Table 1) ranged between 10000 and 190000 cells/ml. Local increases in population were found at a depth of 100 m at station 2 and



**Fig. 3.** Relation between  $\delta^{13}C-C_{org}$  and the  $C_{org}$  content of meltwater from snow and ice in the exploration region lat.  $81^{\circ}30' - 82^{\circ} N$ : (1) ice, top; (2) ice, bottom; (3) snow.

at 120 m depth at station 4. At the same depths, increased water temperature and salinity were also observed.

The numbers of bacterial cells obtained were several times lower than those reported by Baitaz *et al.* [4]. The total number of cells they found in surface water samples taken in August 1984 was  $(561 \pm 21) \times 10^3$  cells/ml, and their biomass measured  $112 \pm 3.0$  mg/m<sup>3</sup>. These researchers worked in the Barents Sea area south of  $76^{\circ} - 78^{\circ} N$  and made special mention of the fact that the number of cells decreased to the north. According to Kriss, the bacterial population density established by direct microscopy in the region close to the North Pole never exceeded 10000 cells/ml [17]; i.e., it was close to the lower boundary of the population range that we obtained.

The numbers of bacterial cells in snow cover and sea ice samples were determined for three stations (Table 2). Results of bacterial counts within the mass of sea ice confirmed the recognized notion of elevated biological activity in the lowermost ice layer, where the maximum density of bacteria,  $(100 - 110) \times 10^3$  cells/ml, was found. In addition to bacterial cells, single-cell algae and flagellants were also observed in this layer, giving it its typical grayish brown color. The number of diatomic algae in these samples was as high as  $(0.2 - 3.0) \times 10^3$  cells/ml. Using light microscopy, bacterial cells from the sea ice were shown to be greater than in the water samples, and, accordingly, the same was true of their biomasses. By contrast, in the snow samples, the number of bacteria was fairly low  $((12 - 14) \times 10^3$  cells/ml). Short rods and cocci were predominant in the preparations. Microscopic observation of snow-melted water revealed a large number of mineral and organic particles lacking the radiation characteristic of live cells. These particles are derived from the aeolian suspension sorbed by snow

from the atmosphere. It is likely that the bacteria in the snow samples were also components of the suspended aeolian matter.

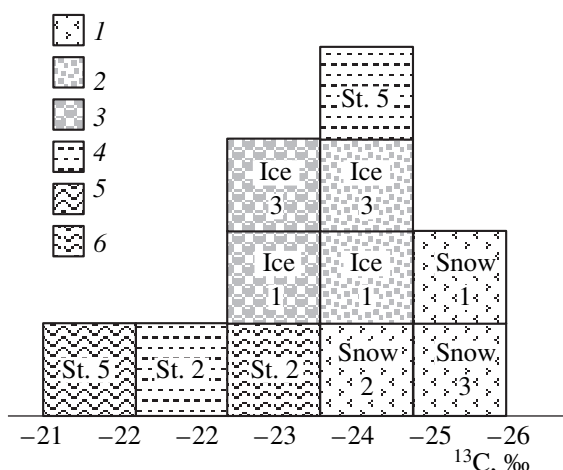
#### *Rates of CO<sub>2</sub> Assimilation, Glucose Utilization, and Methane Oxidation*

Rates of dark assimilation of <sup>14</sup>C-carbon dioxide give a general idea of the overall biogeochemical activity in the samples. Our experiments (Tables 1, 2) showed that the lowest rates of CO<sub>2</sub> assimilation occur in samples of snow-melted water (3–5 µg C/(l day)). The total alkalinity of snow-melted water was low (Alk ≤ 0.2 µg-equiv). The rate of <sup>14</sup>CO<sub>2</sub> assimilation in water samples varied between 10 and 33 µg C/(l day)). Similar rates of <sup>14</sup>CO<sub>2</sub> assimilation (11 µg C/(l day)) were recorded in the central part of the Barents Sea in January 1981 [18]. In our tests, the highest values of the total biogeochemical activity were observed in snow-melted water samples obtained from the lowermost ice layer (45 µg C/(l day)).

The rate of glucose utilization in snow- and ice-melted water varied between 65 and 207 µg C/(l day) (Table 2). In samples obtained from the water column, it ranged from 6.5 to 90 µg C/(l day), with a maximum intensity observed in horizons near the seafloor (stations 4, 10, 11, and 15).

The methane concentrations in the samples studied ranged between 50 and 180 nM. At the stations studied, the rate of methane oxidation varied from 0.01 to 0.3 nM/(l day) with the horizon. Local peaks of methane oxidation activity were observed in 100 to 200 m horizons which for several stations were exactly the horizons with increased CO<sub>2</sub> assimilation. To compare, in the northern part of the Kara Sea, the rate of methane oxidation in seawater was 0.4–8.8 nM CH<sub>4</sub>/(l · day), and in the coastal area of Dixon Island, it was as high as 8.9–26.1 nM CH<sub>4</sub>/(l · day) [19].

In the analysis of the data, it should be noted that the bacterial activity within the ice column actually only takes place in the liquid phase within the intercrystal space [20]. The concentration of salts in such cells normally exceeds that in seawater. The relation between the liquid phase (brines) and the solid phase in sea ice is known to depend on the age of the ice and its temperature. For example, at –2°C, which is the temperature of the lowermost ice layer next to the seawater, the liquid phase accounts for 0.7 of the ice volume; at –4°C, it is 0.35; and at –8°C, it is about 0.1 [21]. When the samples were thawed, liquid water (brine) from the intercrystal cells, where mineral and organic components are dissolved and all biogenic processes occur, was combined with very fresh crystal ice water. As a result, the obtained numbers of microorganisms and rates of processes measured represent the values averaged over the entire ice column rather than the actual figures for the brines where activity takes place. The true process rates can be calculated by using correction



**Fig. 4.** Histogram of values of  $\delta^{13}\text{C-C}_{\text{org}}$  in melt water from snow and ice and in the water column, obtained in the exploration region lat. 81°30'–82° N: (1) snow cover; (2) ice cover (top); (3) ice cover (bottom); (4) water column; (5) water near seafloor; (6) water directly under ice.

coefficients determined by sea ice temperature in the horizons of interest.

In discussing rates of microbial processes in snow- and ice-melted water, it should be noted that the results do not fully relate to the actual processes of in situ snow and sea ice. Rather, these data provide a quantitative measure of the ability of live cells to develop activity in water. The development of cell activity is primarily conditioned by a change in the aggregate state of samples (water transformation from the crystalline to the liquid state). A higher temperature of the samples is not a very important source of discrepancy between the obtained readings and the real process rates, because samples of snow and ice melted water were exposed to –1.4°C. At this temperature, samples from the lowermost ice layer persisted in the liquid state, where less saline samples of snow and the topmost ice layer started to freeze at the very beginning of the exposition process.

#### *Characteristics of Suspended Matter, the Content of C<sub>org</sub>, and the Value of $\delta^{13}\text{C}_{\text{org}}$*

Twenty seven suspension samples were studied (Table 3). The highest concentrations of suspended matter were observed in ice (3.4–9.0 mg/l) and snow (2.6–4.3 mg/l). In seawater, an elevated concentration of suspended matter was found only in the water layer directly under the ice (st. 2, under ice, Table 3). In the rest of the samples, it ranged from 0.9 to 1.69 mg/l. The maximum concentration of C<sub>org</sub> (189–310 µg C<sub>org</sub>/l) was found in the ice samples and in water directly under the ice (160 µg/l). The concentration of C<sub>org</sub> in snow (41–55 µg/l) is 5–7 times lower than in ice, and somewhat lower than in the water column (Table 3). In the northernmost region (81°30'–82° N), we obtained a



**Table 4.** Physicochemical and microbiological characteristics of bottom sediments of the Barents Sea

Station no. (depth, m) Horizon, cm	Station coordi- nates	Eh, mV	SO <sub>4</sub> <sup>2-</sup> , g/l	Alk, mg-equiv/l	Rates of microbial processes				Number of bacteria, cells × 10 <sup>8</sup> /g wet silt	Methane content, μl/dm <sup>3</sup>
					1*	2**	3***	4****		
St. 4 (785)										
0–2	82°01' N	400			19	14	9.4	0	6.3	3.1
20–35	39°05' E	270	2.31	3.5	58	25	10	0	4.4	3.5
46–52		160	2.38	3.6	11	37	6.2	0	2.1	3.6
65–80		60	2.34	3.7	27	16	6.6	11	1.5	3.3
95–110		–40	2.32	3.8	13	21	6.2	4.5	1.5	3.5
110–130		–20	2.53	3.3	10	22	11	2.5	1.0	3.8
145–160		–10	2.38	4.0	6.4	9.8	9.0	0	0.8	4.0
St. 7 (365)										
0–5	80°32' N	180	–	–	75	24	4.2	0	6.0	1.0
10–20	43°41' E	120	3.18	3.5	8.6	19	5.8	210	2.2	2.3
40–47		100	2.93	3.0	7.0	31	12	538	0.5	3.9
70–77		80	2.83	3.2	8.8	18	96	112	0.8	7.0
150–157		30	2.55	5.5	7.2	38	28	0	0.8	9.5
St. 9 (350)										
0–2	80°24' N	440	2.81	3.1	67	16	98	0	5.5	15.0
5–9	41°34' E	200			22	5.1	26	0	3.0	9.7
9–21		120	2.60	3.1	13	9.0	40	261	3.3	9.5
45–60		–20	2.76	3.2	11	25	26	4.8	2.5	9.8
85–110		–10		3.3	9.8	30	51	137	2.1	19.3
120–135		–10	2.45	3.4	9.0	17	24	190	2.3	9.2
165–170		–30	–	–	6.0	19	12	0	2.0	4.5
St. 16 (336)										
0–2	79°21' N	370	2.74	3.1	124	13	28	0	1.7	4.7
15–30	39°34' E	190	2.78	3.5	43	12	6.4	0	0.35	2.2
65–80		–130	2.56	3.4	24	7	3.0	74	0.25	1.5
110–130		–15	2.55	3.6	8	11	14	36	0.21	5.9

\* Glucose consumption, μg C/(dm<sup>3</sup> day).\*\* Dark <sup>14</sup>CO<sub>2</sub> fixation μg C/(dm<sup>3</sup> day).\*\*\* Oxidation of <sup>14</sup>C–methane, nl/(dm<sup>3</sup> day).\*\*\*\* Bacterial sulfate reduction, μg S/(dm<sup>3</sup> day).

direct relationship between the concentration of C<sub>org</sub> in the suspension and the biomass of bacteria in the ice and snow samples (Fig. 2). This fact suggests that the development of algae stimulates both microbial processes and bacterial ice colonization.

In the same region, the value of δ<sup>13</sup>C<sub>org</sub> turned out to depend upon the concentration of C<sub>org</sub> in snow and ice, i.e., upon the total bacterial biomass in these samples (Fig. 3). In snow samples, where C<sub>org</sub> (also in the form of bacterial biomass) is low, the isotopic composition of organic carbon is deficient in the <sup>13</sup>C heavy isotope. The matter suspended in snow is mostly composed of material windborne from the mainland, where the values of δ<sup>13</sup>C<sub>org</sub> are known to range between –24.7 to –25.8‰.

In autumn, active blooming of algae takes place in the ice, especially in its lower horizons, and communities of heterotrophic microorganisms develop. It is precisely here that a relatively high intensity of CO<sub>2</sub> assimilation is observed (Table 2). The values of δ<sup>13</sup>C<sub>org</sub> for samples of suspended matter in ice and in the seawater horizon underlying the ice at station 2 ranged from –22.6 to –24.4‰.

In other words, active biogeochemical processes (algal blooming and bacterial activities) cause isotopic fractionation of carbon and its enrichment with the heavier isotope as compared to that of C<sub>org</sub> in the snow cover, which is dominated by allochthonous organic matter.

The isotopic composition of the suspended  $C_{org}$  reflects the influence of the three major sources of organic matter suspended in the seawater column: aeolian, phytoplanktonogenic, and bacterial. Active biogeochemical processes in ice and in the water column give rise to isotopic fractionation and enrichment with the heavier isotope compared to aeolian and other allochthonous organic matter in the Barents Sea (Fig. 4). The isotopic composition of organic carbon suspended in the Barents Sea is notably depleted in heavier isotopes compared to that of  $C_{org}$  in the water column in the midlatitudes ( $40^{\circ}$  S to  $40^{\circ}$  N), where  $\delta^{13}C_{org}$  is known to vary between  $-22$  and  $-18\text{‰}$  [12]. In high latitudes, the deficit of the heavier isotope  $^{13}C$  in seawater isotopic composition can amount to  $5$ – $10\text{‰}$  and be due to a number of factors, including fractionation in the course of photosynthesis and bacterial processes, and also as a result of a higher concentration of lipids in phytoplankton and the presence of mainland allochthonous material containing lighter isotopes. The relative contributions of each of these factors will be estimated in future studies.

#### *Abundance of Bacteria and Rates of Microbial Processes in Bottom Sediments*

The first data on the number of bacteria and rates of microbial processes in bottom sediments in the central Arctic, north of  $81^{\circ}$  N, are given in Table 4. The number of bacteria in the top horizon was as high as  $(1.7\text{--}6.3) \times 10^8$  cells/g wet silt. At a depth of 20 to 130 cm, the number of bacteria dropped to  $(0.25\text{--}2.3) \times 10^8$  cells/g sediment. Virtually the same population number was observed in bottom sediments in the central and southern parts of the Barents Sea during studies we conducted aboard the research vessel *Academician Vavilov* in 1997.

Rates of bacterial sulfate reduction were determined in four bottom sediment columns (Table 4). In the sediment column from the continental slope (station 4, 785 m depth), sulfate reduction was active in horizons at the interface between the aerobic and anaerobic zones, amounting to  $11 \mu\text{g S}/(\text{dm}^3 \text{ day})$ . In the underlying horizons, the rate of this process declined. At the same time, the sulfate content of the pore water in sediments showed virtually no variation with column depth. The low rate of bacterial sulfate reduction in the lower horizons is apparently related to the depletion of available organic matter. In bottom sediment columns sampled along the Franz Joseph Land–Victoria Island section (stations 7, 9, and 16, 336–365 m depth), the rate of sulfate reduction in some horizons was quite high,  $112\text{--}538 \mu\text{g S}/(\text{dm}^3 \text{ day})$ . The obtained figures are close to the sulfate reduction rates in Barents Sea bottom sediments established on voyage 11 of the research vessel *Academician Vavilov* (up to  $800 \mu\text{g S}/(\text{dm}^3 \text{ day})$ ) [22].

The rate of  $^{14}CH_4$  oxidation in the sediments was determined at four stations. With the content of meth-

ane equal to  $3.1\text{--}19.3 \mu\text{l}/\text{dm}^3$  wet silt, the rate of its oxidation was  $3.0\text{--}98 \text{ nl } CH_4/(\text{dm}^3 \text{ day})$ . The obtained rates of bacterial oxidation of methane are higher than those reported by Gal'chenko *et al.* [15], who showed that, in Bering Sea sediments, these rates varied between 1.5 and  $20 \text{ nl } CH_4/(\text{dm}^3 \text{ day})$  and the total methane content was  $0.24\text{--}8.0 \mu\text{l}/\text{dm}^3$ . In our study, the maximum rate of methane oxidation was found in the top oxidized horizons. In studies conducted by Gal'chenko *et al.*, the methane oxidation process was most active in subsurface reduced sediment horizons with  $Eh = -110$  to  $-440$  mV. In comparison, the rate of methane oxidation in sediments from the northern part of the Kara Sea was  $4.5\text{--}33.8 \text{ nl } CH_4/(\text{kg day})$  [19], which is close to our data.

To determine the overall heterotrophic potential, the rate of  $^{14}C$ -glucose utilization was measured. The peak rate of  $^{14}C$ -glucose utilization was observed in the top-most horizons of bottom sediments (stations 7, 9, and 16). In sediments at station 4, the process was most active in the 20–35 cm horizon.

Above, the first evidence for the real rates of microbial processes in Barents Sea sediments north of  $80^{\circ}$  N was presented. The key factor determining the intensity of such processes is the availability of organic matter, while the low temperatures and the presence of the year-round ice cover have no significant effect on the activity of microorganisms in bottom sediments. The quantitative data on the activities of microorganisms also show that the arctic ice cover acts as a complex biogeochemical barrier, in which the processes of biosynthesis and microbial destruction of organic matter are in balance.

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