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EXPERIMENTAL ARTICLES

Bacterial Abundance and the Activity of Microbiological Processes in the Bay of Tugur of the Sea of Okhotsk

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Abstract—This paper presents the results of investigation of the total abundance and the biomass of bacterioplankton, the abundance of heterotrophic bacteria, and the activity of microbiological processes involved in the carbon cycle in the water of the Bay of Tugur of the Sea of Okhotsk. In different regions of the bay, the total abundance of bacterioplankton was found to vary from 0.51×10^6 to 2.54×10^6 cells/ml; the bacterioplankton biomass, from 8.5 to 46.5 μg C/l; the abundance of heterotrophic bacteria, from 0.06×10^3 to 2.12×10^3 cells/ml; the bacterial assimilation of CO_2, glucose, acetate, and protein hydrolysate, from 0.8 to 6.3, from 0.11 to 1.88, from 0.07 to 0.56, and from 0.01 to 0.22 mg C/(m^3 day), respectively; the degradation of organic matter ranged from 28 to 221 mg C/(m^3 day); and the intensity of methane oxidation, from 0.0005 to 0.17 μl CH_4/l. The spatial pattern and the functional characteristics of bacterioplankton in the Bay of Tugur were found to be dependent on the tidal dynamics.

Key words: bacterioplankton, microbiological processes, tidal dynamics.

The specific hydrologic and hydrochemical conditions and high tidal range in the Bay of Tugur (up to 7 m) [1] have led to the formation of a unique ecosystem, in which both marine and freshwater organisms are present [2]. Little is still known about the functioning of this ecosystem in general and its bacterial constituent in particular [3].

The aim of this work was to study the microbiological parameters of the water of the Bay of Tugur and to assess the role of bacterioplankton in the carbon cycle in the local situation.

MATERIALS AND METHODS

The Bay of Tugur (137° E, 54° N) is located in the western part of the Sea of Okhotsk and extends longitudinally 120 km from the Ptichii island to the settlement of Tugur at the mouth of the Tugur River (Fig. 1). The neck in the middle of the bay divides it into two parts (northern and southern), which differ in their morphometric parameters and hydrologic conditions. The open northern part of the bay is exposed to the influence of the waters of the Sea of Okhotsk, so that only the nearshore waters of this area are slightly freshened. In contrast, waters in the southern part of the bay are freshened by the river runoff, particularly during falling tides [1].

The water of the Bay of Tugur was sampled during July–August 1990 from on board the research vessel *Brig* at 12 stations located mostly in the central and southern parts of the bay (Fig. 1), where hydrodynamic conditions are very complex. Water samples, taken two times a day (at high and low waters) at different depths, were analyzed for several important physicochemical characteristics, as well as for the abundance, vertical and horizontal distribution, and biochemical activity of bacterioplankton.

Water was sampled either with a Ruttner bottle made of Plexiglas (for physicochemical analysis) or using a Frantsev sampler equipped with sterile bottles. Water samples were appropriately treated on board the vessel within 1–2 h after sampling and then thoroughly analyzed in the laboratory. The temperature and oxygen content of the water were measured using a KL-115 oxygen-meter equipped with a thermistor. The concentration of dissolved methane was determined by the method of partial proportions [4] using a Chrom-5 gas chromatograph equipped with a Porapak N column and a flame ionization detector. The total abundance and the size of bacterioplankton cells were determined by the epifluorescence method with acridine orange [5] and 0.2-μm nucleopore filters. Heterotrophic bacteria were enumerated using SPA : 10 agar plates [6]. The dark assimilation of CO_2, glucose, acetate, and protein hydrolysate and the photosynthetic activity of phytoplankton were assayed with radionuclides, by which the intensity of β radiation was measured with a Mark-2 scintillation counter [6]. The intensity of methane oxidation was measured by gas chromatography [4]. The results obtained were quantitatively analyzed assuming

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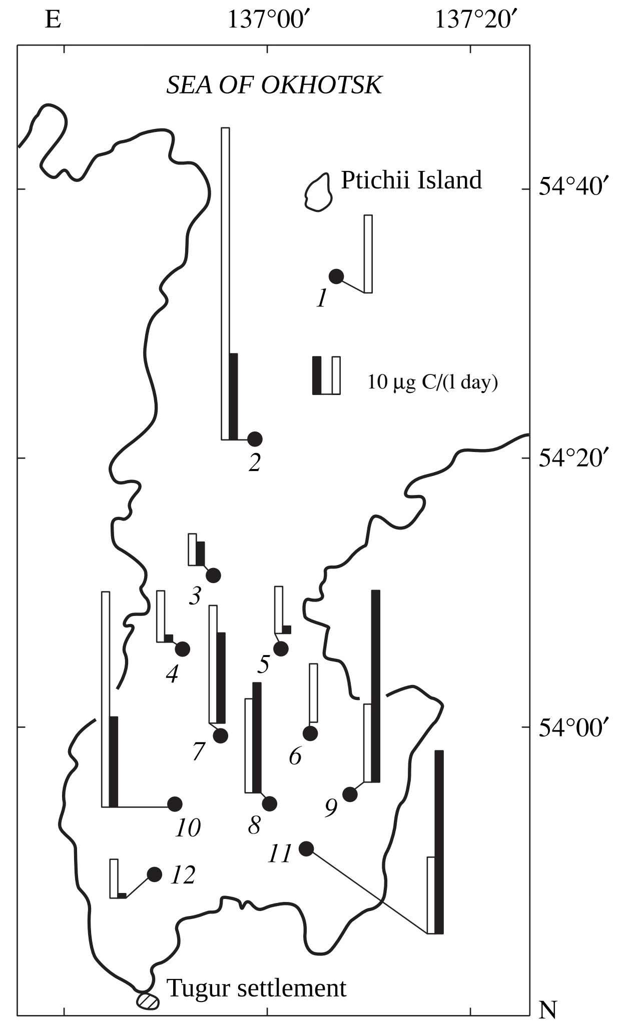

Fig. 1. Map showing the location of stations *1–12* in the Bay of Tugur. Open and dark bars illustrate the photosynthetic activity of phytoplankton in the surface water at high and low sea levels, respectively.

that dark assimilation comprises 6% (in terms of carbon) of bacterial productivity, that the degradation of organic matter is 2.13 times the bacterial productivity, and that the carbon content of the wet biomass comprises 7% [7].

RESULTS AND DISCUSSION

Investigations showed that the water of the Bay of Tugur is heterogeneous, both vertically and horizontally, in salinity (the experimental data of A.V. Ivanov) and other physicochemical parameters (Table 1), as well as in the photosynthetic activity of phytoplankton (Fig. 1). The distribution pattern of the parameters measured turned out to be different at high and low tidal waters.

In the northern part of the bay (stations 1, 2), where the effect of the waters of the Sea of Okhotsk is profound, the temperature stratification of water was observed neither at high nor at low water. Similar results were obtained for the shallow water off the southeastern coast of the bay (stations 9, 11, 12). In spite of the temperature homogeneity of water in these areas, the vertical distribution of biogenic gases there was not uniform (Table 1). In the north, the content of dissolved oxygen varied from almost 12 mg O_2/l (about 140% of saturation) in the surface water to 0.8–1.1 mg O_2/l in the bottom water. At some stations in the south, the content of methane in the bottom water was 1 to 2 orders of magnitude higher than that at the other stations and reached 4–12 μl CH_4/l, which is comparable with the methane content in the anaerobic basins of the Baltic Sea [8]. The zone of the bay neck (stations 3–5), where the interaction between the water of the Sea of Okhotsk and the freshened water from the southern part of the bay is maximum [2], was characterized by intense gradients (both vertical and horizontal) of temperature and hydrochemical parameters. The concentration of dissolved CH_4 in this area was low throughout the water column (Table 1).

As compared to the water of the Sea of Okhotsk [9], the total abundance of bacterioplankton in the surface water of the Bay of Tugur was typically higher and more variable (Table 2). The only exception was station 1, where the density of bacterioplankton in the surface water horizon (0–1 m) was as low as 0.61×10^6 cells/ml, which is on the same order as in the pelagic (open-water) zones of seas [7, 10]. Near the southern and southeastern coasts of the bay, the abundance of bacterioplankton was $(1.94–2.54) \times 10$ cells/ml, which is a typical value for eutrophic seawaters [11].

The biomass of bacterioplankton, which depends both on the abundance and the size of bacterial cells [12], varied from 9.5 to 46.5 μg C/l. The spatial distribution of the bacterial biomass differed from that of the total bacterial abundance. The minimal bacterial biomasses were observed in the central and northern parts of the bay, where cocci and minute rods were dominant microorganisms. The maximum bacterial biomasses were detected at stations 2, 9–11, where the water was characterized by intense photosynthesis (Fig. 1). Here, the fraction of cocci decreased, whereas that of large rods increased. Some water samples contained chains of cells and filamentous bacterial forms. In the southern part of the bay (station 12), where the effect of runoff from the Tugur River is maximum [2], the total abundance of bacterioplankton considerably increased at low tidal water. The increase was mainly due to the presence of minute bacterial forms in the river runoff [13].

The abundance of heterotrophic bacteria in the surface water varied from 60–110 cells/ml in the northern and central regions of the bay to $(2–3) \times 10^3$ cells/ml in the southern and southeastern nearshore waters (Table 2). Similar results were obtained during the 1988 summer expedition in the Bering and Chukchi Seas, the maxi-

Table 1. The physicochemical characteristics of water in the Bay of Tugur

Station	Water level	Depth, m	Transparency, m	T, °C	Salinity, ‰	O_2, mg/l	CH_4, μl/l
1	High	23	7.0	$\frac{8.0}{7.9}$	$\frac{30.2}{30.2}$	$\frac{10.6}{9.8}$	$\frac{0.08}{0.09}$
2	High	21	6.2	$\frac{10.2}{10.1}$	$\frac{30.0}{30.1}$	$\frac{11.9}{9.8}$	$\frac{0.26}{0.12}$
2	Low	17	5.5	$\frac{12.2}{12.0}$	$\frac{28.1}{29.4}$	$\frac{10.2}{9.8}$	$\frac{0.22}{0.12}$
3	High	21	6.0	$\frac{12.8}{12.2}$	$\frac{29.2}{29.4}$	$\frac{9.9}{9.2}$	$\frac{0.15}{0.19}$
3	Low	16	5.0	$\frac{14.6}{13.6}$	–	$\frac{9.2}{9.0}$	$\frac{0.18}{0.21}$
4	High	13	5.5	$\frac{13.8}{12.8}$	$\frac{29.1}{29.2}$	$\frac{10.1}{9.6}$	$\frac{0.17}{0.26}$
4	Low	5	4.5	$\frac{15.6}{15.4}$	$\frac{26.4}{-}$	$\frac{8.7}{-}$	$\frac{0.25}{-}$
5	High	15	5.5	$\frac{14.6}{13.6}$	$\frac{28.1}{28.3}$	$\frac{9.6}{8.9}$	$\frac{0.17}{0.22}$
5	Low	12	4.5	$\frac{15.2}{14.6}$	$\frac{26.0}{27.8}$	$\frac{8.4}{8.2}$	$\frac{0.29}{0.21}$
6	Mean	7	6.2	$\frac{14.8}{14.0}$	–	$\frac{9.2}{8.9}$	$\frac{0.28}{0.59}$
7	High	13	5.0	$\frac{13.0}{12.2}$	$\frac{26.8}{28.0}$	$\frac{10.3}{9.1}$	$\frac{0.25}{0.22}$
7	Low	8	4.0	$\frac{14.8}{14.2}$	$\frac{22.1}{28.0}$	$\frac{9.1}{8.9}$	$\frac{0.28}{0.37}$
8	High	14	5.2	$\frac{14.6}{13.8}$	$\frac{26.5}{27.1}$	$\frac{8.9}{8.7}$	$\frac{0.26}{0.29}$
8	Low	7	4.8	$\frac{16.1}{15.5}$	–	$\frac{8.9}{8.4}$	$\frac{0.47}{0.41}$
9	High	7	5.0	$\frac{16.4}{15.4}$	$\frac{23.3}{24.0}$	$\frac{8.4}{8.2}$	$\frac{0.76}{5.21}$
9	Low	3	2.8	17.2	$\frac{21.1}{-}$	8.9	11.6
10	High	14	5.0	$\frac{14.6}{14.2}$	$\frac{25.4}{26.1}$	$\frac{10.1}{9.1}$	$\frac{0.37}{0.31}$
10	Low	8	4.8	$\frac{16.8}{15.6}$	$\frac{22.3}{26.0}$	$\frac{8.4}{7.8}$	$\frac{0.56}{2.83}$
11	High	6	5.0	$\frac{16.6}{15.6}$	$\frac{22.9}{25.0}$	$\frac{8.4}{8.2}$	$\frac{0.68}{2.11}$
11	Low	3	2.8	17.1	$\frac{15.5}{20.1}$	9.3	12.4
12	High	9	4.0	$\frac{17.0}{16.6}$	$\frac{20.8}{21.4}$	$\frac{8.4}{7.4}$	$\frac{0.61}{2.33}$
12	Low	4	2.5	$\frac{17.6}{17.0}$	$\frac{10.3}{-}$	$\frac{8.0}{7.4}$	$\frac{0.82}{6.44}$

Note: Numbers in the numerators and denominators refer to the surface and bottom waters, respectively. The sign "–" indicates the absence of data.

Table 2. The total abundance of bacterioplankton (BA), its biomass (BB), and the abundance of heterotrophic bacteria (HBA) in the surface waters (0–1 m) of the Bay of Tugur

Station	BA, 10^6 cells/ml	BB, μg C/l	HBA, 10^3 cells/ml
1	0.61 –	9.5 –	0.32 –
2	1.23 0.89	26.3 16.8	1.21 0.64
3	0.81 0.79	15.3 16.3	0.11 0.06
4	0.83 1.05	13.1 16.6	0.48 0.52
5	0.81 0.89	14.4 13.1	0.24 0.12
6	0.83 –	13.9 –	0.11
7	0.86 0.91	16.3 17.6	0.46 0.28
8	1.07 1.14	20.5 24.3	0.27 0.54
9	1.17 2.54	26.1 46.5	0.79 2.47
10	1.56 1.32	33.1 26.7	1.82 1.18
11	1.22 2.24	24.7 45.5	1.01 2.12
12	1.28 2.02	22.1 33.5	0.37 1.22

Note: Numbers in the numerators and denominators refer to the surface and bottom waters, respectively. Heterotrophic bacteria were counted on SPA : 10 medium.

mum amount of heterotrophic bacteria being detected in the high-productivity and, especially, nearshore waters [14].

The essential role of bacteria in aquatic ecosystems is due to their involvement in the carbon cycle. The dark assimilation of CO_2 (one of the most general functional characteristics of bacterioplankton) in the surface water was 0.8–6.3 μg C/(l day), which corresponds to an intermediate productivity of seawater [7, 15]. The assimilation rate of organic compounds, predominantly glucose, varied from 0.02 μg C/(l day) (the central and northern regions of the bay) to 1.88 μg C/(l day) (the southeastern nearshore waters) (Table 3). At southern stations 8, 9, and 11, the functional activity of bacterioplankton was maximum at low sea level (Table 3), although the bacterioplankton contained inactive and dead microbial cells of terrigenic origin. In general, high bacterial activity is typical of the nearshore water areas with high river runoff or where plant detritus is abundant [13, 14, 16].

The intensity of the bacterial oxidation of methane in the surface waters of the bay commonly did not exceed 0.0005–0.003 μl CH_4/(l day), which corresponds to the maximum values of bacterial methane oxidation in open seas [17]. The only exception was shallow waters in the south of the bay, where the effect of the relatively warm river runoff is high [1] and the concentration of dissolved methane in water is maximum (Table 1). The rate of methane oxidation in these shallow waters reached 0.08–0.17 μl CH_4/(l day), i.e., values typical of the waters of the Yenisey Gulf of the Kara Sea and the Baltic Sea in winter [8, 17].

The microbial degradation of organic matter, which was estimated as the value of dark assimilation, greatly differed in different areas of the bay, being minimum (28 mg C/(m^3 day)) in the north of the bay and maximum near the southeastern coast (221 mg C/(m^3 day)). In the north, dark assimilation was maximum at high water, when photosynthetic activity was also at a maximum (Fig. 1). In contrast, in the southeastern shallow waters, dark assimilation was maximum at low water (Table 3). This fact can be explained by the warming of shallow waters and their enrichment with allochthonous organic substances. Bacterial productivity in the surface waters of the bay ranged from 13 to 101 μg C/(l day), being on the same order as in the open Baltic Sea but lower than in the bays and polluted areas of this sea [11]. In general, the bacterial productivity in the Bay of Tugur has a medium level [7].

Thus, the bay water turned out to be horizontally inhomogeneous in their physicochemical and microbiological parameters. The vertical profiling of the bay water was carried out at stations 2, 7, and 11, which were located in particular areas of the bay, either deep or shallow. At station 2, located in the north of the bay, the water was vertically homogeneous in most of the physicochemical parameters during high and low water, except that the surface layer (0–2 m) exhibited oxystratification due to high photosynthetic activity. The vertical distribution of bacterioplankton and the activity of microbiological processes at this station differed during high and low water, reflecting the specificity of primary productive processes in this area (data not presented). At high water, maxima of bacterial abundance and dark assimilation were observed in the middle of the euphotic zone (Fig. 2a), while the maximum number and activity of heterotrophic bacteria were detected near the water surface. At low water, the microbiological parameters, like photosynthetic activity, exhibited two maxima, which were located in the subsurface layer and at the lower boundary of the photic zone (Fig. 2b).

At station 7, located in the deep southern part of the bay subject to the influence of freshwater runoff from the river, the water at the maximum sea level exhibited a narrow vertical variation in its physicochemical

Table 3. The activity of microbiological processes in the surface waters (0–1 m) of the Bay of Tugur

Station	Dark assimilation (μg C/(l day)) of				Bacterial productivity, μg C/(l day)	Methane oxidation, 10^{-2} μl/(l day)	Organic matter degradation, μg C/(l day)
	CO_2	glucose	acetate	protein hydrolysate			
1	0.8/–	0.22/–	0.09/–	0.02/–	13/–	<0.1/–	28/–
2	2.6/1.9	0.75/0.41	0.17/0.11	0.09/0.04	42/30	0.3/0.2	91/66
3	1.3/1.2	0.12/–	0.07/–	0.01/–	21/19	0.3/0.2	46/42
4	2.4/3.5	0.19/–	0.18/–	0.12/–	38/56	0.2/–	84/123
5	2.0/1.6	0.32/–	0.11/–	0.06/–	32/27	0.2/0.2	70/56
6	0.9/–	0.11/–	0.07/–	0.02/–	15/–	0.3/–	32/–
7	1.0/0.9	0.28/0.31	0.12/0.11	0.06/0.07	16/15	0.2/0.3	35/32
8	1.2/2.3	0.22/–	0.09/–	0.04/–	19/37	0.3/0.6	42/81
9	2.2/6.3	0.55/0.81	0.22/0.46	0.08/0.02	37/101	0.8/16	81/221
10	2.7/1.9	0.36/–	0.26/–	0.11/–	45/30	0.4/0.6	95/66
11	1.8/4.1	0.42/1.88	0.18/0.56	0.08/0.18	29/66	0.8/17	63/144
12	2.1/1.7	0.24/–	0.12/–	0.04/–	34/27	0.5/7.9	74/60

Note: Numbers in the numerators and denominators refer to the surface and bottom waters, respectively.

parameters. In contrast, at the minimum sea level, the water at this station exhibited high thermo- and halostratification. The stratification was caused by the superposition of the warmed and freshened water from the southern part of the bay onto the cold and saline bottom seawater. The stratified waters differed not only in their physicochemical parameters but also in the composition of aquatic organisms [2, 3]. In general, the vertical distribution of bacterioplankton and the activity of microbiological processes in this area (Fig. 2) reflected the activity of photosynthetic processes, indicating that labile autochthonous organic matter plays a crucial part in the functioning of the bacterial component of the bay ecosystem. Due to the high inflow of organic matter into the southern part of the bay, its effect on the composition of bacterioplankton and degradative microbiological processes is more pronounced in this region than in the central and northern regions of the bay (Tables 2, 3).

The dynamic and inhomogeneous southeastern shallow waters of the bay (station 11) are subject to the action of the so-called shoreline factor. The vertical inhomogeneity of these waters in physicochemical and microbiological parameters reached a maximum during ebb tide (Fig. 2) and was caused by factors other than those responsible for the inhomogeneity of deep waters. Indeed, the surface waters of the bay are enriched in allochthonous organic matter due to their mixing with the riverine waters, while the waters of the bay bottom are enriched in the organic matter and products of its degradation that are released from the bottom sediments. This is evident from the high concentration of CH_4 (Table 1), bacterial abundance, and activity of microbiological processes in the bottom waters (Fig. 2b).

Analysis of the activity of microbiological processes in the Bay of Tugur showed that its ecosystem may be considered midproductive [7, 11]. The water area of the bay can be divided into four distinct ecolog-

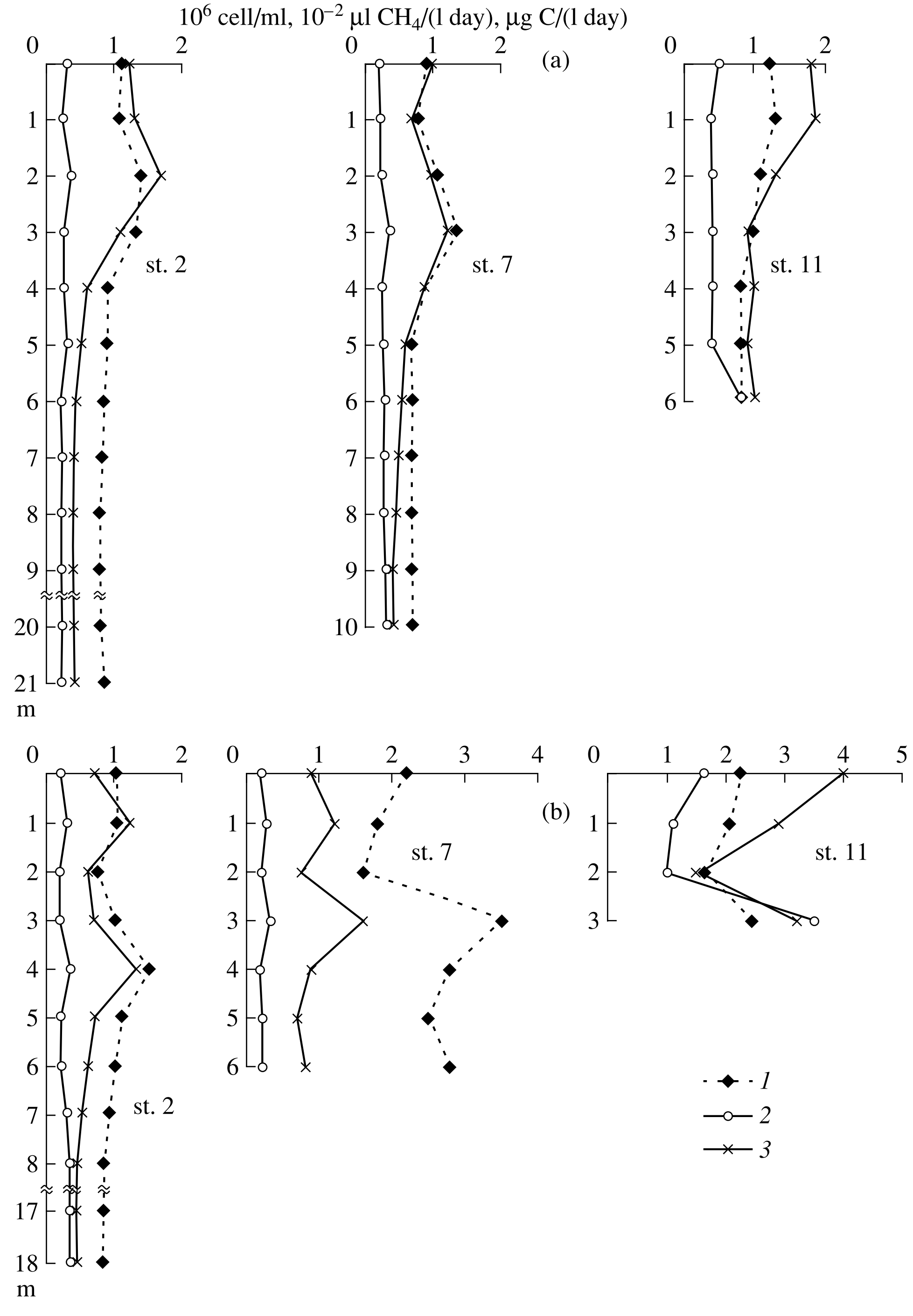

Fig. 2. The vertical distribution of the activity of microbiological processes in the northern (station 2), southern (station 7), and shallow (station 11) zones of the Bay of Tugur at (a) high and (b) low water: (*1*) the total abundance of bacterioplankton (BA) expressed in 10^6 cells/ml, (*2*) methane oxidation rate (MO) expressed in µl CH_4/(l day), and (*3*) the dark assimilation of CO_2 (DA) expressed in µg C/(l day).

ical zones according to the productive and degradative capabilities of their bacterial communities (Table 4). Bacterioplankton in the deep northern region of the bay had maximum degradative capability, whereas bacterioplankton in the shallow nearshore freshened waters exhibited maximum abundance and productivity. Allochthonous organic matter is important for the functioning of the ecosystem of the bay, particularly in its central area and in the southeastern shallows.

Thus, the bacterioplankton in the Bay of Tugur is structurally and functionally inhomogeneous. This is typical of transboundary areas, where there is interaction between waters of different genesis [7, 10, 11], and the nearshore and estuarine areas with a considerable

Table 4. The microbiological parameters of water in different ecological zones of the Bay of Tugur

Ecological zone	BA, 10^6 cells/ml	HBA, 10^3 cells/ml	BB, µg C/l	Dark assimilation of CO_2		Bacterial productivity		Organic matter degradation		Methane oxidation, 10^{-2} µl/(l day)
				µg C/(l day)	mg C/(m^2 day)	µg C/(l day)	g C/(m^2 day)	µg C/(l day)	g C/(m^2 day)	
Open northern zone	0.51–1.23	0.08–1.21	9.5–26.3	0.8–2.6	9–29	13–42	0.15–0.49	28–91	0.32–1.02	0.05–0.15
Central zone	0.81–1.05	0.06–0.68	13.1–16.6	1.2–3.5	13–18	19–56	0.22–0.30	42–123	0.42–0.62	0.1
Deep southern zone	0.83–1.56	0.04–1.82	13.9–26.7	0.9–2.7	8–21	15–45	0.14–0.35	32–95	0.28–0.74	0.1–0.3
Shallow south-eastern zone	1.17–2.24	0.19–3.12	22.1–46.5	1.7–6.3	9–20	27–101	0.15–0.34	60–221	0.28–0.69	0.3–8.5

Note: Heterotrophic bacteria were counted on SPA : 10 medium.

intake of allochthonous matter [13, 14, 16]. In the activity of microbiological processes, the ecosystem of the Bay of Tugur corresponds to midproductive seas [7]. The high tidal range of this bay and several other specific hydrologic features give rise to four distinct ecological zones in the bay. The bacterioplankton of its deep northern region has typical marine features [3, 16]. The central region of the bay is characterized by the mixing of water from the Sea of Okhotsk with the freshened water of the bay and by the degradation of marine cenoses [2, 3]. Photosynthetic activity in this zone is low (Fig. 1), while the role of bacterioplankton in the functioning of the ecosystem is essential [2]. The third zone, located in the deep southern part of the bay, is characterized by destructive microbiological processes with the involvement of allochthonous organic matter. The properties of the fourth shallow zone off the southeastern shore of the bay are determined by the profound effect of the river runoff and the great intake of allochthonous material. This unstable zone is characterized by a large contribution of bacterioplankton to the carbon cycle.

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