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Distribution of saprophytic bacteria in the southern Drake Passage and in the Bransfield Strait (February— March 1981, BIOMASS-FIBEX)*

ABSTRACT: Numbers of saprophytic bacteria were determined by the plate count in samples obtained at 45 oceanographic stations, from six standard depths between 10 and 150 m. Depending on the sampling place, the numbers of bacteria fluctuated between 0.8×10^2 to $4.3 \times 10^4 \cdot l^{-1}$ and 1.2×10^7 to 1.3×10^8 , in a water column under 1 m² sea surface. Most of saprophytic bacteria were observed at stations located south and south-east of the King George Island, and also north and north-west of the Anvers Island. Fewer numbers were found in areas of large krill swarms in the Bransfield Strait, between 58°30' and 62°30' W, and in the north-western part of the research area, far away from the South Shetland Islands.

Key words: Antarctic, saprophytic bacteria distribution

1. Introduction

Saprophytic bacteria are quantitatively not a large but nevertheless an important group of bacteria occurring in marine ecosystems (Bölter 1977, Rheinheimer 1977). The total numbers of all bacteria in Antarctic waters are of the order of 10^8 cells l^{-1} (Hodson et al. 1981), while the numbers of saprophytic bacteria are 10^2 – $10^4 \cdot l^{-1}$ (Sieburth 1965). Because of their high metabolic activity and the ability of a fast degradation of proteins and carbohydrates, saprophytic bacteria play an important role in the mineralization of dead plant and animal matter. An extensive development of saprophytic bacteria populations occurs during the processes of biodegradation of organic matter (Rheinheimer 1977). In Antarctica, this has been confirmed by the results of research on the decomposition of *Euphausia superba* Dana (Zdanowski 1981). During exposure of dead krill in the waters of the Admiralty Bay, the numbers of saprophytic bacteria increased from 3.1×10^5 per lg of wet weight at the moment of krill catching

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to 2.4×10^9 per 1g wet krill weight, on the 15th day of the field experiment. Saprophytic bacteria may also be indicators of hydrological phenomena in marine ecosystems (Kriss, Lebedeva and Mitzkevich 1960). Their numbers depend on the amounts of nutrients and other characteristics of the environment such as climate, salinity, water depth, distance from the shore, etc. (Sieburth 1960, Rheinheimer 1981).

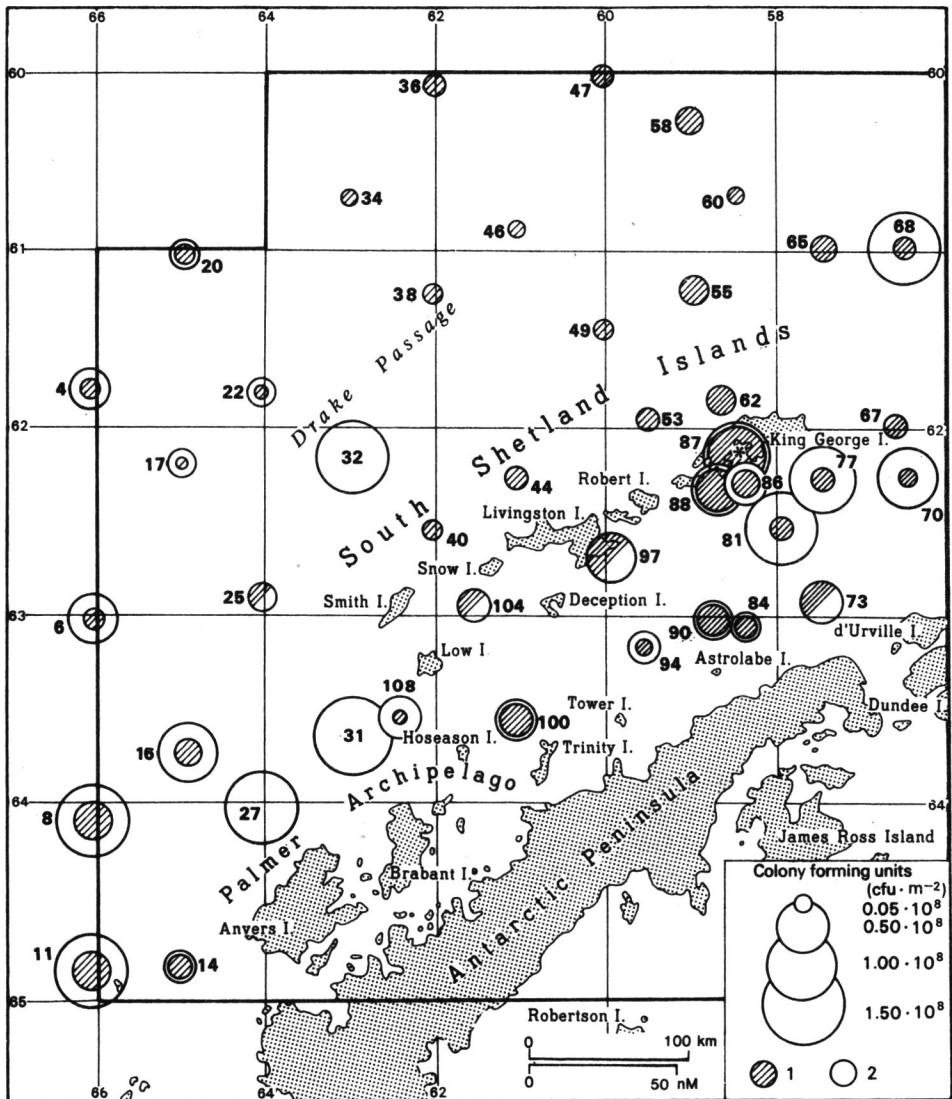


Fig. 1. Distribution of saprophytic bacteria in the southern Drake Passage and in the Bransfield Strait

Colony forming units in sea water column under 1 m^2 ($\text{cfu} \cdot \text{m}^{-2}$) 1 mm of diameter = $0.008 \times 10^8 \text{ cfu} \cdot \text{m}^{-2}$, 1 — cfu determined after 10 days of incubation at 9°C on nutrient agar, 2 — cfu determined after 15 days, * stations 136, 137 and 138 are located in Admiralty Bay (King George Island).

The purpose of this work has been to determine the quantities of saprophytic bacteria in the region of complex ecological investigations in the Drake Passage and the Bransfield Strait. The purpose has also been to relate the occurrence of large krill swarms to the distribution of bacteria in the research area.

2. Materials and methods

Vertical and horizontal distribution of saprophytic bacteria was investigated in the "A" area of the Drake Passage and the Bransfield Strait (Fig. 1), designated by the international BIOMASS-FIBEX programme. The work was carried out on board of the r/v "Profesor Siedlecki" between 15th February and 12th March 1981. Locations of the oceanographic stations, the time and depths of sampling are given in another report of the expedition (Rakusa-Suszczewski 1982).

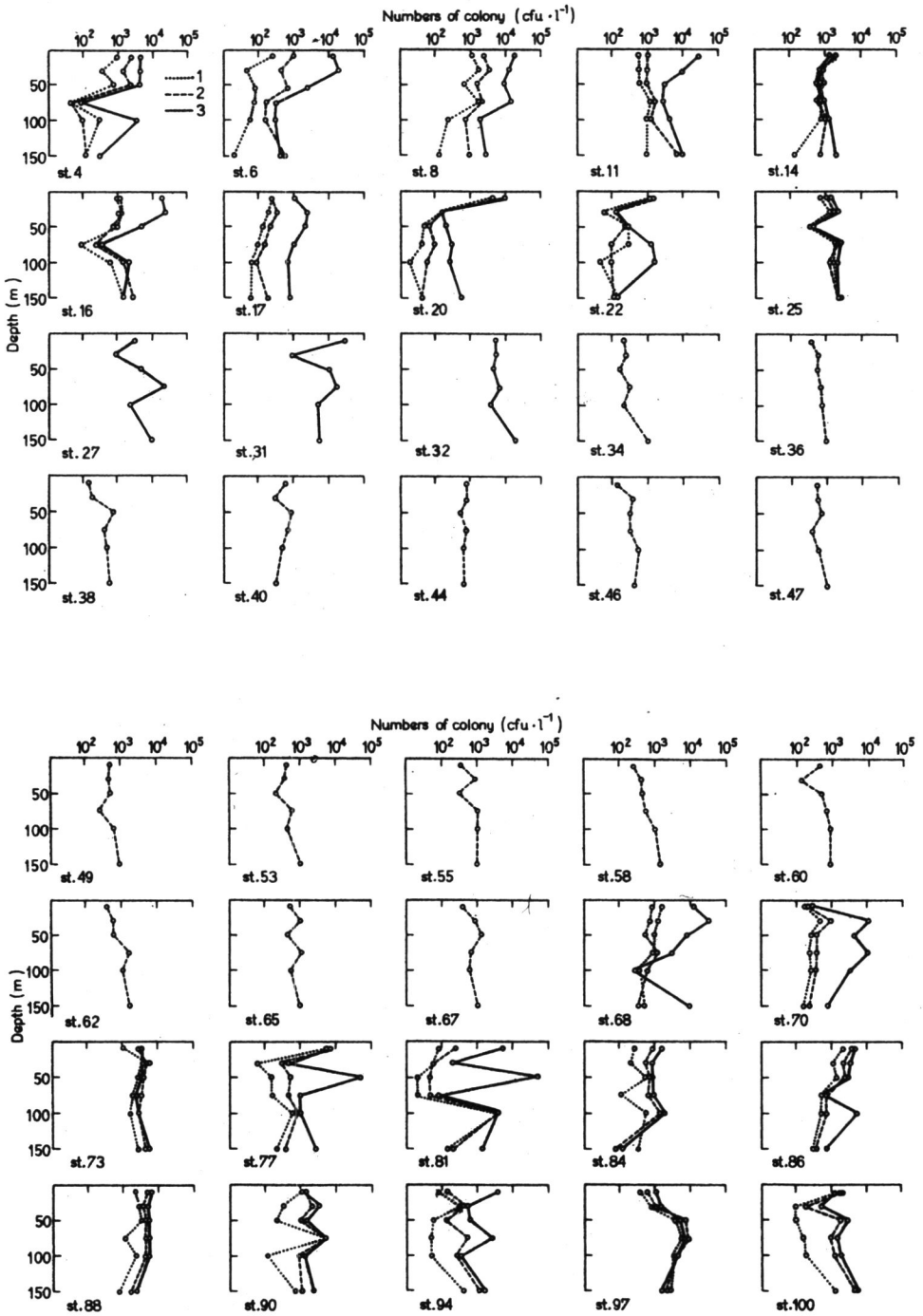
2.1. Sampling

Water samples were taken at 45 stations at standard depths of 10, 30, 50, 75, 100 and 150 m. A Van Dorn type bottle (six litre capacity), sterilized with 96% ethanol (Kriss, Misustina and Lebedeva 1969) was used to obtain the samples. Additional water samples were collected at one station near the northern shore of the Elephant Island (61°02,8' S, 55°07, 0'W): at 15, 65 and 95 m (total depth 100 m). Samples for the determination of the numbers of colony forming units (*cfu*) were prepared immediately after their collection.

2.2. Preparation of samples

50 ml water samples were filtered through sterile Millipore 47 mm HA filters (pore size 0.45 μm). Filters with bacteria were incubated at the optimum growth temperature of 9°C (Inoue 1977) on a nutrient agar of pH 7 which contained: Bacto-beef extract, Bacto-tryptone, Bacto-agar and sea water collected *in situ* (Fenchel and Hemmingsen 1974). Colonies (*cfu*) were counted after 5, 10, and 15 days of incubation. The value of *cfu* reaches a maximum after 15 days, and this has been considered as the basis for the general estimation of bacteria numbers. Exception are stations 34 to 67 and st. 135 (Figs. 1 and 2) where *cfu* were counted only after 15 days of incubation. These values do not include the part of population which forms colonies between the 10th and 15th day of culture, and makes up, on the average, 40% of the total numbers of colonies after 15 days of incubation.

The obtained values are shown as $\text{cfu} \cdot \text{l}^{-1}$, and as integrated values (by the trapezium method) of $\text{cfu} \cdot \text{m}^{-2}$ contained in a water column of 10–150 m under 1 m² sea surface. Observations of colonies were done in



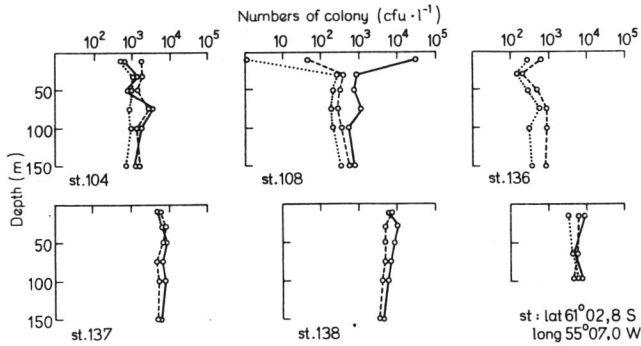


Fig. 2. Vertical distribution of saprophytic bacteria on investigated stations Determined after: 1) 1–5 days, 2) 2–10 days, 3) 3–15 days of culturing at 9 C.

a light passing through the bacteria cultures and reflected, of a changeable intensity. Colonies were distinguished macroscopically based on such characteristics as rate of growth, pigment formation, and shape and size of the colonies.

3. Results and discussion

The quantities of saprophytic bacteria sampled during the Antarctic summer of 1981 in the research area of the Drake Passage and the Bransfield Strait (Fig. 1), and determined after 15 days of incubation, were found to fluctuate between 0.8×10^2 and 4.3×10^4 $cfu \cdot l^{-1}$ (Fig. 2). In the vertical distribution the highest numbers of bacteria were noted at depths of 10 and 30 meters which corresponded to a water layer above a pycnocline (stations 4, 6, 11, 16, 20, 68). This tendency had not been noticed in areas of strong mixing of waters such as the Bransfield Strait and the south-west regions of the investigated "A" area (Fig. 1 and 2). Generally, a "mosaic" type of distribution was characteristic of the abundances of saprophytic bacteria, with low $cfu \cdot l^{-1}$ values found in a close neighbourhood of high values.

The integrated cfu values (Fig. 1 and Table I) in a water column under $1 m^2$ sea surface ranged between 1.2×10^7 and 1.3×10^8 $cfu \cdot m^{-2}$. The highest amounts of bacteria (6.4×10^7 — 1.3×10^8 $cfu \cdot m^{-2}$) were found in the Admiralty Bay (King George Island), south-east of the King George Island, west of the Elephant Island, and also north and north-west of the Anvers Island (stations: 6, 70, 16, 137, 11, 138, 27, 77, 68, 81, 8, 32, 31), (Fig. 1). Small numbers of saprophytic bacteria (1.5×10^7 — 6.0×10^7 $cfu \cdot m^{-2}$) occurred in the Bransfield Strait between $58^\circ 30'$ and $62^\circ 30'$ W. This region was characterized by small quantities of net-collected phytoplankton (Kopczyńska and Ligowski, 1982) and by the occurrence of very large krill swarms. On the other hand, a reverse relationship in the amounts of krill and bacteria was found in another area, the vicinity of the Elephant Island where both, extremely large krill swarms and high numbers of bacteria were found. In the north-western part of the area investi-

Table I.
Number of saprophytic bacteria (as colony forming units — *cfu*)

Station	Integrated value (<i>cfu</i> × 10 ⁷ · m ⁻²) after following days of incubation:		
	5	10	15
4	0.5	1.1	3.6
6	0.1	0.6	6.4
8	1.2	2.5	12.0
11	1.2	3.3	9.8
14	1.0	1.3	1.7
16	1.2	1.9	8.5
17	0.2	0.3	1.9
20	0.5	1.1	1.4
22	0.3	0.3	1.2
25	2.3	2.3	2.3
27	—	—	11.0
31	—	—	13.0
32	—	—	12.0
34	—	0.5	—
36	—	1.0	—
38	—	0.7	—
40	—	0.8	—
44	—	1.0	—
46	—	0.6	—
47	—	0.9	—
49	—	0.8	—
53	—	0.8	—
55	—	1.3	—
58	—	1.2	—
60	—	0.9	—
62	—	1.5	—
65	—	1.2	—
67	—	1.2	—
68	—	1.0	—
70	0.3	0.5	7.2
73	3.5	4.7	4.7
77	0.9	1.3	11.0
81	1.5	1.6	12.0
84	0.5	1.2	1.5
86	1.2	1.9	4.2
88	2.7	4.5	5.8
90	1.5	2.4	3.1
94	0.2	0.6	1.9
97	4.7	6.0	6.0
100	0.7	2.8	3.5
104	1.2	2.4	2.4
108	0.3	0.4	4.3
136	0.5	1.0	—
137	—	8.1	9.6
138	—	6.8	10.0
E.I.	3.4	5.0	5.0*)

*) Station near Elephant Island (61°02.8 S; 55°07.0 W); Integrated value in sea water column calculated on the basis of numbers for 15, 65, 95 m.

gated, far away from the South Shetland Islands, both the numbers of bacteria and of krill were low.

The numbers of saprophytic bacteria determined after ten days of incubation ranged between 0.4×10^2 and 1.0×10^4 $cfu \cdot l^{-1}$ (Fig. 2), and from 0.3×10^7 to 8.1×10^7 $cfu \cdot m^{-2}$ (Fig. 1 and Table I). Highest values (6.0×10^7 — 8.1×10^7 $cfu \cdot m^{-2}$) were observed at stations located in the Admiralty Bay and near the southern shore of the Livingston Island. Low quantities (0.5×10^7 — 1.3×10^7 $cfu \cdot m^{-2}$) were noted in the Drake Passage between $57^\circ 30'$ and $62^\circ 00' W$. Here the numbers of bacteria were determined only after ten days of incubation on nutrient agar. Because of an uncompleted development of bacteria on agar, counts obtained after ten days of incubation have less meaning for the interpretation of the total numbers of bacteria. They illustrate the quantities of fast growing bacteria, which on the average make up 60% of the investigated population.

In the process of determining the numbers of bacteria with the plate method, a development of bacteria on a nutrient agar have been observed. There have been distinguished early colonies, formed between the second and fifth day of culture, and late-formed colonies, noticeable between the 10th 15th day. The former group contributed on the average 37% and the latter 40% to the total populations. Among the early-formed colonies, five to ten groups have been distinguished which differed macroscopically and reached 3–10 mm in diameter. White, creamy-yellow and creamy-pink colonies were dominant; their shape was round, the surface and edges smooth, and the profile was convex. Within the late—formed colonies, numerous red-orange, and fewer white ones have been observed. They had a characteristic small size of 0.2–0.4 mm, which remained unchanged even after prolongation the time of culture to next 15 days. Preliminary microscopical analysis revealed in the colonies the presence of long (5–15 μ m), coloured, bent rods resembling *Myxobacteriae*.

The present results corroborate earlier data (Kriss, Lebedeva and Mitzkevitch 1960, Sieburth 1965, Kriss, Misustina and Lebedeva (1969) of the low saprophytic bacteria content in the Antarctic marine waters. No regions devoid of saprophytic bacteria were observed during the present investigations. This is in contrast to the frequent observations of Lebedeva (1958) in the Indian Ocean section of the Antarctic, and of Mitzkevich (1975) in the south west Atlantic section of Antarctic, where no saprophytic bacteria were found. Our results showed an increase of bacteria numbers in the near shore areas; this had been previously noted by Lebedeva (1958). Exception were the nearshore areas of the Bransfield Strait characterized by the occurrence of large krill swarms, and the associated with them, low amounts of saprophytic bacteria (Fig. 1).

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4. Резюме

Во время антарктической экспедиции БИОМАСС-ФИБЭКС (15 февраля—12 марта 1981) на корабле “Профессор Седлецки” было определено распределение сапрофитных бактерий в проливе Дрейка и проливе Брансфилда.

Пробы воды взятые на 45 океанографических станциях из шести стандартных глубин в пределе 10—150 м, были отфильтрованы на миллиметровых фильтрах НА с диаметром поров 0,45 мкм. Бактерии на фильтрах инкубировались на питательном агаре в течение 15 дней в температуре +9°C. Определено число колоний в литре воды ($cfu \cdot l^{-1}$) и в столбе воды под 1 м² ($cfu \cdot m^{-2}$).

Бактерии образовали несколько групп отличающихся внешне колоний. Среди них были выделены ранние колонии, образующиеся на второй до пятого дня инкубации. Они образовали 37% популяции, определённой на 25 день инкубации. Поздние колонии образующиеся между 10 и 15 днём инкубации составляли 40%.

Количество сапрофитных бактерий, после 15 дней экспозиции колебалось в пределах от 0.8×10^2 до $4.3 \times 10^4 cfu \cdot l^{-1}$ и от 1.2×10^7 до $1.3 \times 10^8 cfu \cdot m^{-2}$ в зависимости от места и глубины. Обнаружено, что большое количество этих бактерий наиболее часто встречается на глубинах 10 и 30 м. Однако в пространственном распределении бактерии наблюдалась “мозаичность”, заключающаяся в том, что большие и малые величины $cfu \cdot l^{-1}$ были перемешаны. Наибольшие интегральные величины были обнаружены на станциях расположенных на юг и юго-восток от острова Кинг Джордж, а также на север и северный-запад от острова Анверс. Более низкие величины были обнаружены в северо-западной части исследуемого района, издали от архипелага Южных Шетландов. Также низкие величины были обнаружены в проливе Брансфилда, в районе больших концентраций криля.

5. Streszczenie

Podczas wyprawy antarktycznej BIOMASS-FIBEX (15 lutego—12 marca 1981) na statku r/v “Profesor Siedlecki” określono rozmieszczenie bakterii saprofitycznych w Cieśninie Drake’a i Cieśninie Bransfielda.

Próbki wody pobrane z 45 stacji oceanograficznych, z sześciu standardowych głębokości, zawartych pomiędzy 10 i 150 m, filtrowano przez sączi Millipore HA, o średnicy porów 0,45 μm. Bakterie na filtrach inkubowano przez 15 dni w temperaturze +9°C na agarze odżywczym. Oznaczono liczbę kolonii w litrze wody ($cfu \cdot l^{-1}$) i w słupie wody pod 1 m² ($cfu \cdot m^{-2}$).

Bakterie tworzyły kilkanaście grup zróżnicowanych zewnętrznie kolonii. Wśród nich wyróżniono wczesne kolonie powstające między 2 i 5 dniem inkubacji. Stanowiły one średnio 37% populacji oznaczanej w 15 dniu inkubacji. Późne kolonie powstające między 10 i 15 dniem inkubacji stanowiły średnio 40%.

Liczebność bakterii saprofitycznych, po 15 dniach ekspozycji, wahała się od 0.8×10^2 do $4.3 \times 10^4 cfu \cdot l^{-1}$ i od $1,2 \times 10^7$ do $1,3 \times 10^8 cfu \cdot m^{-2}$ w zależności od stanowiska i głębokości. Stwierdzono tendencję do częstszego występowania wysokich wartości na 10 i 30 m głębokości. Jednak w przestrzennym rozmieszczeniu bakterii obserwowano „mозаиковоść”, polegającą na przemieszaniu niskich i wysokich wartości $cfu \cdot l^{-1}$. Najwyższe zintegrowane wartości stwierdzono na stanowiskach zgrupowanych na południe i południowy-wschód od Wyspy Króla Jerzego oraz na północ i północny-zachód od Wyspy Anvers. Niższe wartości stwierdzono w północno-zachodnim rejonie obszaru badań, z dala od Archipelagu Szetlandów Południowych. Także niskie wartości stwierdzono w Cieśninie Bransfielda w miejscu występowania dużych skupień kryla.

6. References

1. Bolter M. 1977 — Numerical taxonomy and character analysis of saprophytic bacteria isolated from the Kiel Fjord and the Kiel Bight (In: *Microbial Ecology of a Brackish Water Environment*. Ed. G. Rheinheimer) — Springer — Verlag, Berlin: 148–178.
2. Fenchel T., Hemmingsen B. B. 1974 — *Manual of Microbial Ecology* — Akademisk Forlag Studentlitteratur, 235 pp.
3. Hodson R. E., Azam F., Carlucci A. F., Fuhrman J. A., Karl D. M., Holm-Hansen O. 1981 — Microbial uptake of dissolved organic matter in McMurdo Sound, Antarctica — *Mar. Biol.*, 61: 89–94.
4. Inoue K. 1977 — Effect of temperature on growth of obligately psychrophilic bacteria — *J. Gen. Appl. Microbiol.*, 23: 53–63.
5. Kopczyńska E. E., Ligowski R. 1982 — Phytoplankton abundance and distribution in the southern Drake Passage and the Bransfield Strait, Antarctic (February–March 1981 FIBEX-BIOMASS investigations) — *Pol. Polar Res.*, 3 (3–4): 193–202.
6. Kriss A. E., Lebedeva M. N., Mitzkevich I. N. 1960 — Micro-organisms as indicators of hydrological phenomena in seas and oceans — II. Deep-Sea Research, 6: 173–183.
7. Kriss A. E., Mišustina I. E., Lebedeva M. N. 1969 — Plotnost' bakterjalnogo naselenija (geterotrofov) v vodnoj tolšče Južnogo i Indijskogo okeanov — *Mikrobiologija*. 38: 511–517.
8. Lebedeva M. N. 1958 — Količestvennoe raspredelenie geterotrofnich mikroorganizmov v Indijskom Okeane i priliegajuščich morjach Antarktiki — *Dokl. Akad. Nauk SSR*, 121: 557–560.
9. Mickevič I. N. 1975 — Mikrobiologičeskije issledovanija v II-m rejsie nis. "Akademik Kurcatov" — *Trudy Instituta Okeanologii im. P. P. Širšova* 103: 48–59.
10. Rakusa-Suszczewski S. 1982 — Report on the r/v "Profesor Siedlecki" expedition to the Antarctic in 1981 during the international FIBEX-BIOMAS programme — *Pol. Polar Res.*, 3 (3–4): 137–141.
11. Rheinheimer G. 1981 — *Mikrobiologie der Gewässer* — G. Fischer, Jena, 251 pp.
12. Rheinheimer G. 1977 — Regional and seasonal distribution of saprophytic and coliform bacteria (In: *Microbial ecology of a Brackish water environment*, Ed. G. Rheinheimer) — Springer verlag, Berlin 121–137.
13. Sieburth J. McN. 1960 — Soviet aquatic bacteriology: a review of the past decade — *Quart. Rev. Biol.* 35: 179–205.
14. Sieburth J. McN. 1965 — Microbiology of Antarctica (In: *Biogeography and ecology in Antarctica*, Eds. P. Van Oye and J. van Mieghem) — *Monogr. Biol.* 15, Dr W. Junk Publ., The Hague, 267–295.
15. Zdanowski M. K. 1981 — Growth of bacteria in the course of decomposition of *Euphausia superba* Dana — *Bull. Ac. Pol.: Biol. Sci.* 29: 155–161.

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