The occurrence and activity of sulphate-reducing bacteria in the bottom sediments of the Gulf of Gdańsk

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Abstract

The paper presents the results of investigations concerning the number, distribution and physiological activity of sulphate-reducing bacteria (SRB) inhabiting the bottom sediments of the Gulf of Gdańsk. The numbers of this group of bacteria range between 0.76×10^3 and 1.27×10^4 cells per g wet sediment. The bacterial sulphate reduction rate in bottom sediments of this area of the Baltic Sea varies from 1.89 to 31.6 nM SO_4^{2-} g^{-1} 24 h^{-1}. The numbers of SRB and their physiological activity were subject to considerable seasonal fluctuations, maximum values being noted in summer (June) and minima in spring (April). A direct relationship has been found between the number of SRB and hydrogen sulphide concentrations; there is, however, no such relationship with reference to sulphate concentrations. The numbers and distributions of SRB demonstrated considerable variation in a depth profile of bottom sediments. SRB inhabiting the bottom sediments of the Gulf of Gdańsk were able to use three different organic substrates (lactate, acetate, propionate) as electron donors and as carbon and energy sources.

1. Introduction

Increasing eutrophication of marine coastal areas generates a higher primary production in the photic zone. The organic matter produced is accumulated mainly in the bottom sediments, which provide anaerobic conditions as a result of the active consumption of oxygen by heterotrophic organisms (Takii & Fukui 1991, Imhoff *et al.* 1995). Under such environmental conditions the process of sulphate reduction plays a key part in the mineralisation of organic matter (Parkes *et al.* 1989, Bussmann & Reichardt 1991). According to Jørgensen (1982) and Bharathi & Chandramohan (1985) over 50% of the accumulated organic matter becomes mineralised in coastal and shelf sediments. Marine sulphate-reducing bacteria (SRB), among which the *Desulfovibrio* and *Desulfotomaculum* genera are dominant, make up an ecologically and morphologically heterogeneous group of microorganisms (Battersby 1988). The main property of those either obligate or facultative anaerobic bacteria populations is their active use of sulphate as a final electron acceptor during anaerobic respiration (Keith *et al.* 1982). The final product of this respiration is hydrogen sulphide, which is discharged into the environment. Where concentrations of H_2S are very high, this can penetrate SRB cell membranes and thus impede their metabolic activity (Okabe *et al.* 1995). SRB utilise a very wide spectrum of different low molecular organic compounds (lactate, acetate, proprionate, succinate, pyruvate, ethanol, aliphatic acids, sugars, amino acids, indole, nicotinic acid) as electron donors, and also as carbon and energy sources (Parkes *et al.* 1989, Bak & Pfennig 1991, Caumette 1993). SRB synthesise numerous enzymes that catalyse sulphate reduction. The following enzymes play a major part in sulphate activation and reduction: pyrophosphatase, ATP sulphurylase, bisulphite reductase, desulphoviridin, desulphorubidin and desulphofuscidin (Gibson 1990, Saas *et al.* 1992, Visscher *et al.* 1992). Sulphate and organic matter concentrations, sedimentation rate, turbulence, bioturbulence, temperature, salinity and hydrostatic pressure are the main environmental factors controlling the numbers and distribution of SRB and the rate of bacterial sulphate reduction (Jørgensen 1982, Edenborn *et al.* 1987, Westrich & Berner 1988).

SRB, which generate large amounts of toxic hydrogen sulphide in aquatic ecosystems, are important not only for ecological reasons. They are also vital from the point of view of the economy. This primarily concerns the petroleum industries, which use immense amounts of seawater in their technologies while recovering oil from under the sea bed. A large amount of SRB may cause the oil and gas to acidify, the piping to corrode and technical installations to become clogged (Gibson *et al.* 1987, Battersby 1988,

Peng *et al.* 1994, Okabe *et al.* 1995). Owing to their quite considerable ecological and economic importance, SRB have become recently a popular subject of scientific investigation (Gibson 1990).

Despite the obviously important position of SRB in the functioning of marine ecosystems, information concerning them in the Baltic Sea is scarce. Hence it was the aim of the present study to determine the spatial distribution and seasonal dynamics of SRB populations in the bottom sediments of the Gulf of Gdańsk, southern Baltic Sea.

2. Material and methods

The investigations were carried out in the Gulf of Gdańsk, *i.e.* that part of the southern Baltic receiving the waters of the river Wisła. This inflow of riverine waters substantially modifies environmental conditions in the Gulf of Gdańsk. The Wisła carries *ca* 75% of the pollutants entering the Baltic Sea from Poland. In 1991, for example, a total of 145 million m^3 of waste water were discharged into the Gulf of Gdańsk by the Wisła (Andrulewicz 1996). These waste waters carry large amounts of organic matter which accumulate mainly in the bottom sediments. Three sampling stations were chosen in the Gulf of Gdańsk (No. 1 at 54°27′N–18°57′E, No. 2 at 54°30′N–18°57′E, No. 3 at 54°35′N–18°57′E) (Fig. 1). From on board the r/v 'Oceanograf' 30-cm-long bottom sediment cores were sampled by means of a tube scoop in spring (23 April 1995), summer

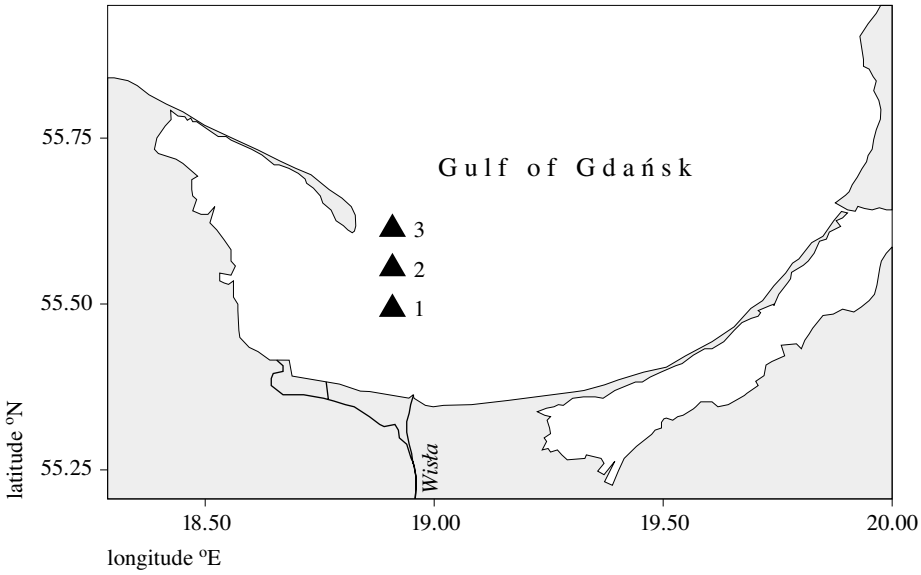

Fig. 1. Map of the study area in the Gulf of Gdańsk with sampling stations

(22 June 1996) and autumn (30 October 1995). The cores were then sectioned into 2.5 cm subsamples to a depth of 15 cm, and from there into 5 cm subsamples to the bottom of the core. The bottom sediment samples were cooled (4°C) and immediately transported to the laboratory: no more than 9 hours elapsed between sampling and microbiological analysis. In order to determine SRB numbers, the sediments were diluted with sterile buffered water (Daubner 1967) and inoculated onto a liquid medium B (Postgate 1966) composed of KH_2PO_4 – 0.5 g, NH_4Cl – 1.0 g, $CaSO_4$ – 1.0 g, $MgSO_4 \cdot 7H_2O$ – 2.0 g, Na-lactate – 3.5 g, yeast extract – 1.0 g, ascorbic acid 1.0 g, $FeSO_4 \cdot 7H_2O$ – 0.5 g, thioglycollic acid – 1.0 g and seawater – 1 dm^3. The medium was prepared with aged seawater. After its pH had been adjusted to 7.0–7.5, the medium was sterilised at 117°C for 20 min. Thioglycollic acid, a reducing agent, which had been separately sterilised on membrane filters, was introduced into the sterilised medium. In parallel to the Postgate medium containing Na-lactate as growth substrate, two other variants of the medium were used with Na-acetate and Na-propionate as growth substrates. The bacterial cultures were incubated at 20°C for 28 days. SRB numbers were determined by the most probable number (MPN) method, and blackening of the medium was regarded as a positive result. Following Jørgensen (1978), it was assumed that one SRB cell reduces 2.5 x 10^{-12} mol SO_4^{2-} 24 h^{-1}, from which the bacterial rate of sulphate-reduction could be calculated. To estimate the rate of carbon mineralisation via sulphate reduction in the sediment, it was assumed that 2 mol of CO_2 are produced per mol of sulphate reduced (Edenborn *et al.* 1987, Takii & Fukui 1991). Sulphate concentration and hydrogen sulphide contents in the bottom sediments were determined by methods routinely used in marine chemistry (UNEP/IOC/IAEA 1988).

3. Results

The data in Table 1 show that the sulphate concentrations in the bottom sediments (155.2–524.l mg SO_4^{2-} dm^{-3}) were highest at station 1, closest to the mouth of the Wisła. At stations 2 and 3, more distant from the river mouth, these concentrations (45.6–198.1 mg SO_4^{2-} dm^{-3}) were much lower. At all three stations the highest sulphate concentrations were recorded in the top layer (0–5 cm) of the bottom sediment (Fig. 2). The sulphate content decreased with depth and the lowest concentrations were recorded in the deepest parts of the sediment cores (20–30 cm).

The hydrogen sulphide contents in these bottom sediments ranged between 0.13 and 2.22 mmol dm^{-3} (Table 1), reaching a maximum in summer (June) and falling to a minimum in autumn (October). An inverse relationship obtained between the contents of sulphate and hydrogen

Table 1. Average values of chemical and bacteriological variables investigated in the Gulf of Gdańsk sediments in different seasons

Season	Station	SO_4^{2-}	H_2S	Number of SRB				Sulphate reduction rate				Organic carbon degraded via sulphate reduction
		[mg dm^{-3}]	[mM dm^{-3}]	[10^3 cells g^{-1} w.s.]				[nM SO_4^{2-} g^{-1} $24h^{-1}$]				[nM C g^{-1} 24 $^{-1}$]
				$\bar{x}$	min	max	SD	$\bar{x}$	min	max	SD	
spring	1	524.1	0.8	0.76	0.24	16.0	4.70	1.89	0.60	4.0	1.20	3.78
	2	198.1	1.48	1.70	0.05	9.2	3.00	4.21	0.12	23.0	7.50	8.42
	3	–	–	–	–	–	–	–	–	–	–	–
summer	1	155.2	1.8	12.7	1.70	35.0	14.50	31.6	4.25	87.5	36.3	63.2
	2	45.6	2.22	0.77	0.18	1.6	0.49	1.93	0.45	4.0	1.2	3.86
	3	47.5	1.84	4.30	1.30	13.0	3.80	10.6	3.25	32.5	9.5	21.2
autumn	1	368.8	0.13	3.60	0.08	16.0	5.40	9.07	0.19	40.0	13.6	18.14
	2	160.1	0.17	5.00	1.10	22.0	6.80	12.6	2.75	55.0	17.0	25.2
	3	115.7	0.27	5.80	0.70	24.0	8.40	14.6	1.75	60.0	21.0	29.2

$\bar{x}$ – average, SD – standard deviation, min – minimum value, max – maximum value.

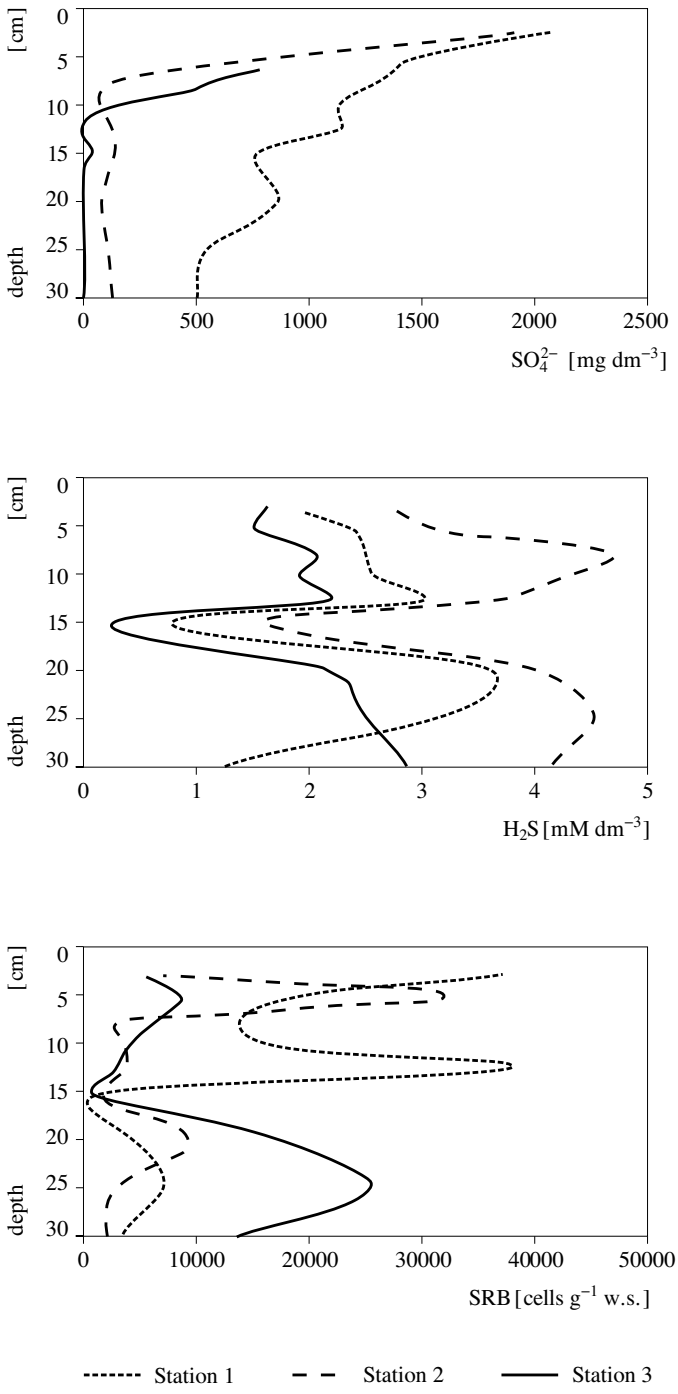

Fig. 2. Vertical distribution of sulphate, hydrogen sulphide and numbers of sulphate-reducing bacteria in the Gulf of Gdańsk sediment

sulphide in the samples. Considerable variations were noted in the hydrogen sulphide concentration in the 30-cm-deep sediment layer. Although H_2S concentrations were lowest at core depths of 15 cm at all three stations, maximum concentrations were recorded at different depths at these stations (Fig. 2).

The data in Table 1 show that mean SRB numbers varied between 0.76 x 10^3 and 1.27 x 10^4 cells per g wet sediment. The bacterial sulphate reduction rate ranged from 1.89 to 31.6 nM SO_4^{2-} g^{-1} 24 h^{-1}, equivalent to mineralisation via sulphate reduction of 3.78–63.2 nM C g^{-1} 24 h^{-1} of organic carbon. The maximum SRB cells count (3.5 x 10^4 g^{-1}) and the fastest rate of bacterial sulphate reduction (87.5 nM SO_4^{2-} g^{-1} 24 h^{-1}) were recorded in summer at station 1 (Table 1).

The data in Table 1 confirm that no direct relation could be found between SRB numbers and sulphate concentration in the bottom sediments during the study period. However, such a relation did exist between SRB numbers and the hydrogen sulphide concentration. In spring and summer a higher hydrogen sulphide concentration corresponded to elevated numbers of SRB, and their higher metabolic activity was reflected in the sulphate reduction rate.

The numbers of SRB in the depth profile of the bottom sediments at particular stations were highly variable, and it was difficult to find any clear distribution pattern (Fig. 2). At each station maximum and minimum SRB counts were noted at different sediment depths.

The data in Fig. 3 indicate that the bacteriological variables under investigation were subject to considerable seasonal fluctuation. The SRB count and the sulphate-reduction rate were highest in summer (June). The smallest number of SRB and a slower rate of bacterial sulphate reduction were recorded in spring (April).

Figure 4 illustrates the results of an analysis whereby SRB on different organic substrates were used as sources of electrons and of energy and carbon in sulphate reduction. The data show that SRB inhabiting the bottom sediments of this particular area of the Gulf of Gdańsk were able to use all three organic substrates (lactate, acetate, propionate) as electron donors and as carbon and energy sources. At the same time, it was found that the SRB inhabiting different layers of the bottom sediment preferred various forms of organic carbon. Lactate was an optimum substrate for the bacteria from the top layer of sediments (0–5 cm). Acetate-utilising bacteria were present in the greatest numbers in the 5–10 cm layer of the sediment, while the bacteria inhabiting the sediments at depths 10–15 cm and 25–30 cm tended to use propionate.

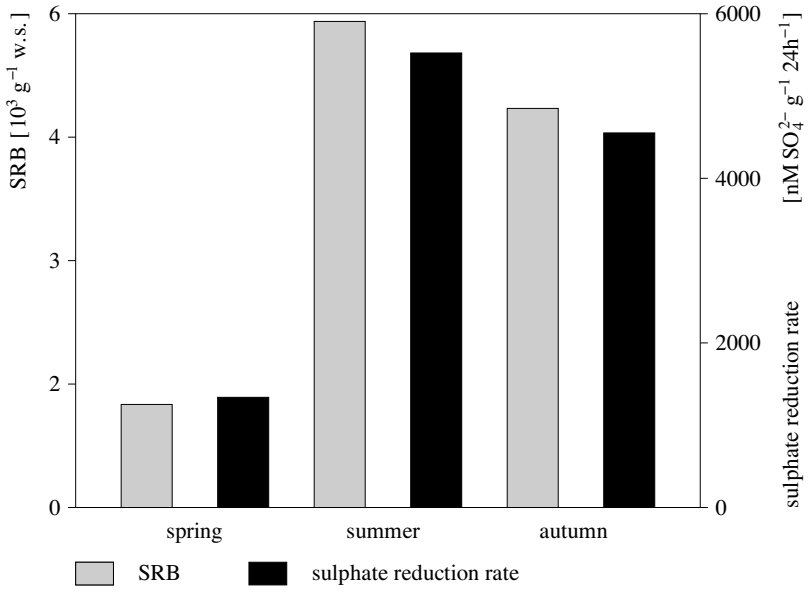

Fig. 3. Seasonal variations in the number of sulphate-reducing bacteria and of sulphate reduction rates

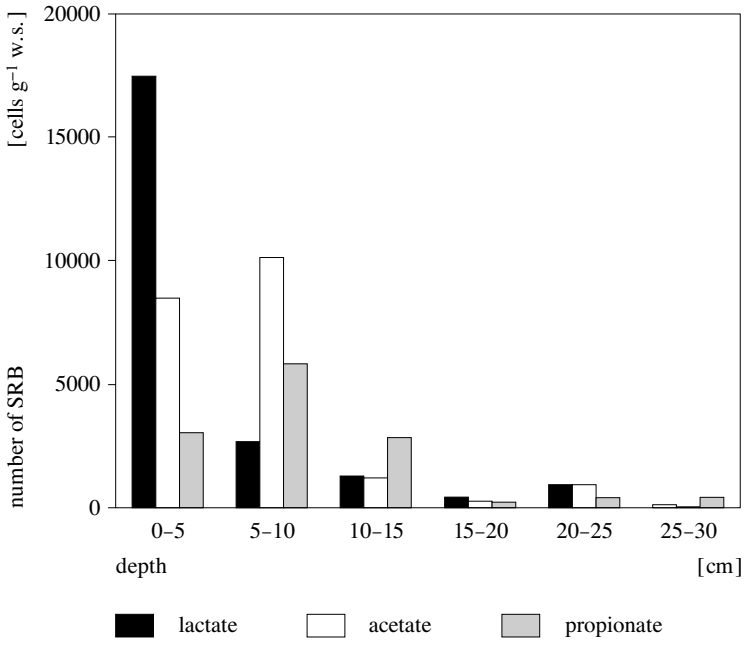

Fig. 4. Depth distributions of lactate-, acetate- and propioniate-utilising SRB

4. Discussion

In marine ecosystems, particularly in coastal zones and estuaries, sulphate-reducing bacteria play a key role in the sulphur cycle and organic matter decomposition (Kohler *et al.* 1984, Fukui & Takii 1990). Marine bottom sediments provide an optimum environment for these micro-organisms. SRB numbers in the bottom sediments of the Gulf of Gdańsk ranged between 76×10^2 and 1.27×10^4 cells per g wet sediment. Schneider (1977) and Bussmann & Richardt (1991) recorded an almost identical number of SRB (4×10^2–7×10^4 cells^{-1} g^{-1}) in the Kiel Bight. In other marine sediments SRB numbers varied from 10^2 to 10^6 cells^{-1} g^{-1} (Jørgensen 1977, 1978, Laanbroek & Pfennig 1981, Bak & Pfennig 1991). In the bottom sediments of both the Gulf of Gdańsk and Limfjorden (Jørgensen 1977), the SRB numbers fluctuated considerably between stations and along their vertical profile. The variations in these bacteriological parameters indicate a significant heterogeneity of these bottom sediments. According to Gibson *et al.* (1987) and Jørgensen & Bak (1991) there are still no optimum methods allowing the SRB count to be determined with precision. As the presently available viable count procedure does not assess bacterial populations accurately enough to yield a result resembling *in situ* numbers, it has been suggested that the SRB count determined on the medium be multiplied by 1000 (Jørgensen 1978).

Numerous investigations (Jørgensen 1977, Kohler *et al.* 1984, Edenborn *et al.* 1987) have indicated a great diversity ($0.4 - 3000$ nM SO_4^{2-} cm^{-3} 24 h^{-1}) in the sulphate reduction rate in marine sediments. According to our data from the Gulf of Gdańsk, this rate lay within the range from 1.89 to 31.3 nM SO_4^{2-} g^{-1} 24 h^{-1} and was greater than in the Kiel Bight by 0.08–8.2 nM SO_4^{2-} g^{-1} 24 h^{-1} (Hartmann & Nielsen 1969). Jørgensen (1982), Ward & Winefry (1985) and Fukui & Takii (1990) conclude that it is organic matter availability and not sulphate concentration that principally limit SRB numbers and the sulphate reduction rate in marine bottom sediments. Our findings fully confirm this hypothesis: SRB numbers and bacterial sulphate reduction rates in the bottom sediments of the Gulf of Gdańsk are independent of the sulphate concentration. At the same time, maximum numbers of SRB were recorded at the Wisła river mouth. This increase in the SRB count and SRB activity in that part of the Gulf of Gdańsk may have been due to Wisła river water flowing into the gulf. This water carries freshwater sulphate-reducing bacteria along with large quantities of organic matter (some 0.6 million tons of organic carbon per year) (Witek 1995) deposited in the bottom sediments. Schneider (1977) and by Hines & Buck (1982) report a similar situation in other aquatic ecosystems. In the bottom sediments examined in the present work, the

numbers of SRB and their activity as indicated by the sulphate reduction rate both reach a maximum in summer and a minimum in spring. This fully corresponds with the results obtained by Bansemir & Rheinheimer (1974) and Schneider (1977) elsewhere in the southern Baltic (Kiel Fjord, Kiel Bight). Temperature is an abiotic parameter governing SRB numbers and the rate of sulphate reduction, especially in shallow marine coastal areas (Westrich & Berner 1988). Fukui & Takii (1989) showed how a temperature increase significantly stimulates SRB reproduction and intensifies bacterial sulphate reduction. At the same time, however, Jørgensen (1977) pointed out that the summer maxima of SRB numbers and their metabolic activity may have been due to the oxygen shortage at that time in the sediments, which may, in turn, have had a disastrous effect on the benthos. The consequent inflow of fresh organic matter stimulates rapid SRB growth. If one takes into account the very large benthos biomass (10 g C m^{-2}) in the bottom sediments of the Gulf of Gdańsk (Witek 1995), this phenomenon could also occur there.

So far it is mainly lactate that has been used as a growth substrate for SRB cultures, as the sole donor of electrons and as sources of carbon and energy. However, new physiological types of SRB have been isolated recently (Keith *et al.* 1982, Gibson *et al.* 1987, Gast & Gocke 1988) from numerous estuarine and marine bottom sediments. Since these bacteria can utilise other organic growth substrates as potential substrates in sulphate reduction, principally acetate and propionate, these must be used as well as lactate if we wish to realistically estimate the population of SRB. Numerous analyses have shown that different physiological groups of SRB in mutualistic relationships with one another may use, though with different intensity, these three organic carbon compounds in their metabolic processes (Laanbroek & Pfeninng 1981, Sorensen *et al.* 1981, Postage 1984, Taylor & Parkes 1985, Parkes *et al.* 1989, Jørgensen & Bak 1991, Visscher *et al.* 1992). Moreover, the SRB inhabiting the bottom sediments of the Gulf of Gdańsk were able to actively use lactate, acetate and propionate as growth substrates. Therefore, determining solely those SRB that use lactate as a growth substrate may provide sufficient information as to their numbers and distribution. At the same time it was found that the SRB inhabiting particular bottom sediment layers chose different growth substrates while reducing sulphates, which would be in line with the earlier findings by Laanbroek & Pfeninng (1981), Bharathi & Chandramohan (1985), Gibson *et al.* (1987).

The results presented in this paper may contribute to the explanation of the role of SRB in the process of organic matter destruction in the bottom sediments of the Gulf of Gdańsk.

References

Andrulewicz E., 1996, *The regional characteristics of the Gdańsk Basin*, Oceanol. Stud., 1–2, 11–16.

Bak F., Pfennig N., 1991, *Sulfate-reducing bacteria in littoral sediment of Lake Constance*, FEMS Microbiol. Ecol., 85, 43–52.

Bansemir K., Rheinheimer G., 1974, *Bakteriologische Untersuchungen über die Bildung van Schwefelwasserstoff in einer Vertiefung der inneren Kieler Förde*, Kiel. Meer., 30, 91–98.

Battersby N. S., 1988, *Sulphate-reducing bacteria*, [in:] *Methods on aquatic bacteriology*, B. Austin, John Wiley & Sons (eds.), 269–299.

Bharathi P. A., Chandramohan D., 1985, *Sulfate reducing potential in estuarine beach*, Ind. J. Mar. Sci., 14, 187–191.

Bussmann I., Reichardt W., 1991, *Sulfate-reducing bacteria in temporarily oxic sediments with bivalves*, Mar. Ecol. Prog. Ser., 78, 97–102.

Caumette P., 1993, *Ecology and physiology of phototrophic bacteria and sulphate-reducing bacteria in marine salterns*, Experientia, 49, 473–481.

Daubner I., 1967, *The Microbiology of Water*, Sloven. Acad. Sci., Bratislava, 447 pp., (in Slovak).

Edenborn H. M., Silverberg N., Mucci A., Sundby B., 1987, *Sulfate reduction in deep coastal marine sediments*, Mar. Chem., 21, 329–345.

Fukui M., Takii S., 1989, *Kinetics of colony formation by sulphate-reducing bacteria*, Bull. Jap. Soc. Microbiol. Ecol., 3, 67–71.

Fukui M., Takii S., 1990, *Seasonal variations of population density and activity of sulfate-reducing bacteria in offshore and reed sediments of a hypertrophic freshwater lake*, Jap. J. Limnol., 51, 63–71.

Gast V., Gocke K., 1988, *Vertical distribution of number, biomass and size-class spectrum of bacteria in relation to oxic/anoxic conditions in the Central Baltic Sea*, Mar. Ecol. Prog. Ser., 45, 179–186.

Gibson G. R., 1990, *Physiology and ecology of the sulphate-reducing bacteria*, J. Appl. Bacteriol., 69, 769–797.

Gibson G. R., Parkes R. J., Herbert R. A., 1987, *Evaluation of viable counting procedures for the enumeration of sulphate-reducing bacteria estuarine sediments*, J. Microbiol. Meth., 7, 201–210.

Hartmann M., Nielsen H., 1969, *$\delta^{34}S$–Werte in rezenten Meeressedimenten und ihre Deutung am Beispiel einiger Sedimentprofile aus der westlichen Ostee*, Geol. Rundschau., 58, 621–655.

Hines M. E., Buck J. D., 1982, *Distribution of methanogenic and suplphate-reducing bacteria in near-shore marine sediments*, Appl. Environm. Microbiol., 43, 447–453.

Imhoff J. F., Schneider A., Podgorsek I., 1995, *Correlation of viable cell count, metabolic activity of sulphur-oxidizing bacteria and chemical parameters of marine sediments*, Helgo. Meer., 49, 223–236.

Jørgensen B. B., 1978, *A comparison of methods for the quantification of bacterial sulphate reduction in coastal marine sediments. III. Estimation from chemical and bacteriological field data*, Geomicrobial. J., 1, 49–64.

Jørgensen B. B., 1977, *Bacterial sulfate reduction within reduced microniches of oxidized marine sediments*, Mar. Biol., 41, 7–17.

Jørgensen B. B., 1982, *Mineralization of organic matter in the sea bed – the role of sulfate reduction*, Nature, 296, 643–645.

Jørgensen B. B., Bak F., 1991, *Pathways and microbiology of thiosulfate transformations and sulfate reduction in a marine sediment (Kattegat, Denmark)*, Appl. Environm. Microbial., 57, 847–856.

Keith S. M., Herbert R. A., Harfoot C. G., 1982, *Isolation of new types of sulphate-reducing bacteria from estuarine and marine sediments using chemostat enrichments*, J. Appl. Bacteriol., 53, 29–33.

Kohler H. P., Ahring B., Albella C., Ingvorsen K., Keweloh H., Laczko E., Stupperich E., Tomei F., 1984, *Bacteriological studies on the sulfur cycle in the anerobic part of the hypolimnion and the surface sediments of Rotsee in Switzerland*, FEMS Microbiol. Lett., 21, 279–286.

Laanbroek H. J., Pfennig N., 1981, *Oxidation of short-chain fatty acids by sulfate-reducing bacteria in freshwater and marine sediments*, Archiv. Microbiol., 128, 330–335.

Okabe S., Nielsen P. H., Jones W. I., Characklis W. G., 1995, *Sulfide product inhibition of Desulfovibrio desulfuricans in batch and continous cultures*, Wat. Res., 29, 571–578.

Parkes R. J., Gibson G. R., Mueller-Harvey I., Buckingham W. J., Herbert R., 1989, *Determination of the substrates for sulphate-reducing bacteria within marine and estuarine sediments with different rates of sulphate reduction*, J. Gen. Microbiol., 135, 175–187.

Peng C. G., Park J. K., Patenaude R. W., 1994, *Statistics-based classification of microbially influenced corrosion in freshwater systems*, Wat. Res., 28, 951–959.

Postgate J. R., 1966, *Media for sulphur bacteria*, Lab. Pract., 15, 1239–1244.

Postgate R. J., 1984, *The Sulphate-Reducing Bacteria*, Cambridge Univ. Press, Cambridge–New York, 208 pp.

Sass H., Steuber J., Kroder M., Kroneck P. M., Cypionka H., 1992, *Formation of thionates by freshwater and marine strains of sulphate-reducing bacteria*, Arch. Microbiol., 158, 418–421.

Schneider J., 1977, *Desulfurication and sulfur oxidation*, [in:] *Microbial ecology of a brackish water environment*, G. Rheinheimer (ed.), Springer Verlag, Berlin–Heidelberg–New York, 25, 244–248.

Sorensen J., Christensen D., Jørgensen B. B., 1981, *Volatile fatty acids and hydrogen as substrates for sulfate-reducing bacteria in anaerobic marine sediments*, Appl. Environm. Microbiol., 42, 5–11.

Takii S., Fukui M., 1991, *Relative importance of methanogenesis, sulfate reduction and denitrification in sediment of the lower Tama river*, Bull. Jap. Soc. Microbiol. Ecol., 6, 9–17.

Taylor J., Parkes R. J., 1985, *Identifying different of sulphate-reducing bacteria within marine sediment systems, using fatty acids*, J. Gen. Microbiol., 131, 631–642.

UNEP/IOC/IAEA, 1988, *Standard chemical methods for marine environmental monitoring. Reference methods for marine pollution studies*, Nairobi–Monaco, 50, 8–50.

Visscher P. T., Prins R. A., Gemerden H., 1992, *Rates of sulfate reduction and thiosulfate consumption in marine microbial mat*, FEMS Microbiol. Ecol., 86, 283–294.

Ward D. M., Winefry M. R., 1985, *Interactions between methanogenic and sulphate-reducing bacteria in sediments*, Adv. Aquat. Microbiol., 3, 141–179.

Westrich J. T., Berner R. A., 1988, *The effect of temperature on rates of sulfate reduction in marine sediments*, Geomicrobiol. J., 6, 99–117.

Witek Z., 1995, *Biological production and its utilization within a marine ecosystem in the western Gdańsk Basin*, Mor. Inst. Ryb., Gdynia, 1–145, (in Polish).