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Seasonal variation in sulfate reduction and methanogenesis in peaty sediments of eutrophic Lake Loosdrecht, The Netherlands

ANJA J. C. SINKE[1], ADI A. CORNELESE[1],
THOMAS E. CAPPENBERG[1] & ALEXANDER J. B. ZEHNDER[2]
[1] *Netherlands Institute of Ecology, Centre for Limnology, Rijksstraatweg 6, NL-3631 AC Nieuwersluis, The Netherlands;* [2] *Department of Microbiology, Agricultural University, Hesselink v. Suchtelenweg 4, NL-6703 CT Wageningen, The Netherlands*

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Abstract. During one year, concentration profiles of sulfate and methane were measured in sediment cores of eutrophic Lake Loosdrecht. Sulfate concentrations decreased exponentially with depth towards a constant threshold value of 7.6 ± 6.1 μM. Concentration profiles were used to calculate fluxes of sulfate and methane and to estimate the anaerobic mineralization rate. Anaerobic mineralization was highest in autumn which was probably due to an increased sedimentation of easily degradable organic carbon. At high rates (>600 μmol organic carbon.m^{-2}.h^{-1}), sulfate reduction appeared to be limited by sulfate and methanogenesis accounted for over 80% of the anaerobic mineralization. At low anaerobic mineralization rates, measured in winter and spring, sulfate reduction was predominant.

There was little methanogenesis below 5 cm depth in the sediment which indicated a rapid decrease of degradable organic matter with depth. There was a remarkable difference, especially in winter, between methane fluxes which were measured in batch experiments and those calculated from the concentration profiles in the sediment. These differences may be due to methane diffusing upward from deep layers.

Introduction

Sulfate reduction and methanogenesis are regarded as alternative degradation reactions that compete for common substrates such as acetate (Winfrey & Zeikus 1977) and hydrogen (Lovley et al. 1982; Robinson & Tiedje 1984). The extent to which sulfate reducers can outcompete methanogens depends on the availability of sulfate. In marine and salt marsh systems with sulfate concentrations higher than 1 mmol.l^{-1}, sulfate reduction accounts for the majority of anaerobic respiration (Capone &

Kiene 1988). However, in fresh water systems sulfate concentrations in the overlying water are much lower, typically between 0.01 and 0.2 mmol.l^{-1}, which results in a very steep gradient of sulfate in the sediment (Winfrey & Zeikus 1979; Lovley & Klug 1983; Cappenberg et al. 1984; Adams & Van Eck 1988). As sulfate reduction in freshwater sediments was demonstrated to be sulfate-limited at concentrations below 0.1 mmol.l^{-1} (Ingvorsen et al. 1981; Lovley & Klug 1983), this process is probably limited by the availability of sulfate during most of the year. As a consequence, most of the anaerobic mineralization is channeled via methanogenesis (Kelly et al. 1982; Cappenberg et al. 1984; Adams & Van Eck 1988).

The availability of sedimenting organic matter can be expected to be an important factor in determining the pathway of the terminal carbon metabolism, especially in fresh water ecosystems. Most studies on the role of sulfate reducers and methanogens have been carried out in deep (> 10 m) lakes (Winfrey & Zeikus 1977, 1979; Lovley et al. 1982; Lovley & Klug 1983; Cappenberg et al. 1984; Kuivila et al. 1989) in which most of the organic matter from the primary production probably is degraded in the water column. However, in shallow lakes, processes in the water and in the sediment may interfere more intensely. So, in shallow eutrophic lakes, the large seasonal fluctuations in primary production and temperature may affect the role of sulfate reduction and methanogenesis in the terminal carbon flow during the year. The present study was carried out to elucidate the seasonal and spatial variation in the importance of sulfate reduction versus methanogenesis in peaty sediments in a shallow eutrophic lake.

Methods

Sampling

The research was carried out in eutrophic Lake Loosdrecht. This lake was made artificially by peat mining in the 17th century when the 7000 years old *Sphagnum* peat was excavated, dried and used as fuel (Van Liere 1986). Primary production in the lake, which is 30—45 mol $\text{carbon.m}^{-2}.\text{y}^{-1}$ on a yearly basis, sometimes exceeds 400 mmol $\text{carbon.m}^{-2}.\text{d}^{-1}$ during summertime (Van Liere et al. 1986). As a result of the morphometry (area $=$ 14.5 km^2, mean depth $=$ 1.8 m) and continuous wind-induced turbulence, the surface layer of the sediment remains aerobic throughout the year. The oxygen penetration depth into the sediment is as shallow as 1.6 mm in summer and increases to 5.4 mm in winter (Sweerts &

Cappenberg 1988). The sediment pore water is slightly alkaline. The pH decreases from pH = 8.3 in the surface layer to pH = 7.3 at greater depths.

Sediment cores were obtained using a hand-driven stainless steel corer containing a 40 cm long inner core of PMMA (polymethylmetacrylate) with an internal diameter of 5 cm. The sharpened edges of the inner core extended well below the metal parts to minimize disturbance of the sediment. Samples were collected in 1988 every fourth week at two representative locations. One sampling station is situated in the open lake and the other, Kievitsarea, is more or less sheltered from wind influence. The sediment at both stations consists of relatively loose organic debris and the organic matter content is very high, 55%—65% on a dry weight basis (Sinke et al. 1990). Temperature was measured just above the sediment surface. Chlorophyll-a and primary production in the lake water were measured by the routine programme of the "Water Quality Research Loosdrecht Lakes" (Van Liere 1986; Van Liere et al. 1986). All sediment cores were placed in a nitrogen-filled glove box (Coy Laboratory Products Inc. Michigan). Two duplicate cores were used for pore water extraction. Half or 1.0 cm thick layers were scooped off and interstitial water was pressed out over 0.45 μm filters using a Reeburgh type squeezer (Reeburgh 1967) at a maximal N_2 pressure of 10^5 Pascal. Two cores were sliced at one cm intervals for methane measurements and subsamples were put in 30 ml Serum bottles immediately. During processing of the sediment cores some methane-loss occurred. Time series extracting subsamples from one layer every 10 seconds, demonstrated that the methane-loss during handling was approximately 10% and data were adjusted for these losses. Methane was measured in the headspace after vigorous shaking.

The bottles containing sediment used for methane measurements were incubated in dark at *in situ* temperature. From the third week on, the accumulation of methane in the headspace was measured every second week for a period of 6—8 weeks. The detection limit for methane production measured this way was 1 $\text{nmol.g}^{-1}\text{d.}^{-1}$.

Analytical procedures

Sulfate was measured with HPLC (Hordijk et al. 1985). Methane was measured using a Packard model 428 gas chromatograph equipped with a Porapak Q column and flame ionization detector (at 160 °C). The carrier gas was helium at a flow rate of 20 ml.min^{-1}.

Diagenetic model

The theoretical model developed by Berner (1964, 1980) is applied to describe both sulfate reduction and methanogenesis in the sediment. The model describes changes in concentration of sulfate and methane with time as a function of diffusion, sedimentation and sulfate reduction or methanogenesis:

$$\delta C/\delta t = D_s \delta^2 C/\delta z^2 - w \delta C/\delta z - f(x)$$

with C: concentration of sulfate or methane at z cm depth (nmol.cm^{-3}), D_s: the whole sediment diffusion coefficient (cm^2.s^{-1}), w: sedimentation rate (cm.s^{-1}) and $f(x)$ = sulfate reduction rate or methane production rate (nmol.cm^{-3}.s^{-1}). The measured concentration profiles were fitted according to:

$$C = (C_0 - C_\infty) \exp(a z) + C_\infty$$

with z: depth (cm), C_0: concentration sulfate or methane in overlying water (nmol.cm^{-3}), C_∞: concentration sulfate or methane at infinite depth (nmol.cm^{-3}) a: attenuation constant (cm^{-1}). The attenuation constant is defined as the ratio of the first order reaction rate constant for sulfate reduction or methanogenesis and the net sedimentation (Berner 1980). The measurements of duplicate cores were pooled. C_0, C_∞ and a could be estimated by fitting of the model to the data by a least squared residual minimization routine. For these calculations BMDPAR (BMDP Statistical Software Inc. Los Angeles, USA) was used (Ralston 1988). The diffusive fluxes of SO_4^{2-} into the sediment and CH_4 towards the sediment surface, were calculated according to Fick's first law of diffusion using the tangents to the fitted equations of the concentration gradients at the sediment-water interface:

$$J = \phi/\theta^2 * D_s * \delta C/\delta z$$

with J: flux of sulfate or methane (nmol.cm^{-2}.sec^{-1}), D_s: whole sediment diffusion coefficient for either sulfate or methane (cm^2. sec^{-1}), ϕ: porosity (dimensionless), θ: tortuosity (dimensionless) and $\delta C/\delta z$ = concentration gradient of either sulfate or methane (nmol.cm^{-4}). D_s were calculated from the molecular diffusion coefficients (D_0 sulfate: 8.9×10^{-6} cm^2.sec^{-1}, at 18 °C, Li and Gregory 1974; D_0 methane: 15.3×10^{-6} cm^2.sec^{-1}, at 15 °C, Broecker & Peng 1974) that were corrected for *in situ* temperature and sediment characteristics. Porosity of the sediment was taken as 0.95 and the square of tortuosity as 1.40 (J.-P. Sweerts, pers. comm.).

The relative importance of sulfate reduction and methanogenesis in the anaerobic mineralization was determined on the basis of the calculated fluxes. The total anaerobic mineralization was approximated as twice the sum of sulfate reduction and methanogenesis.

Statistical analyses

A recent version of CANOCO (Ter Braak 1987a) was used to analyse data with Redundancy Analysis (RDA). RDA (Rao 1964) is a technique which relates a set of multivariate data to explanatory variables. RDA combines the features of Principal Component Analysis, summarizing multivariate data, and multiple regression analysis, relating data (fluxes and rates) to environmental variables. RDA selects successive linear combinations of environmental variables that give the smallest residual sum of squares. The environmental variables used in the analysis were temperature (TEMP), chlorophyll-a concentration and primary production rate in the lake water (CHLA, PROD) and the overall change in temperature and primary production rate between two successive sampling dates (δTEMP, δPROD). Environmental variables were selected stepwise, maximizing the explained sum of squares with the minimum number of environmental variables. The results are presented as a correlation biplot (Gabriel 1971; Ter Braak 1987b). In a biplot the length and the cosine of the angle between vectors of two variables is an indication of the correlation between them. The variables which were analysed were: total anaerobic mineralization rate (TOTAL), methane flux and sulfate reduction rate calculated from the concentration profiles (CH_4, SO_4^{2-}), the relative importance of sulfate reduction expressed as percentage of the total anaerobic mineralization (%SO_4) and the methane production in batches (BATCH). To get an overall insight in the response of the sediment, the data sets of the two sampling points were pooled. To compensate for the differences between the two sampling stations, the location itself was introduced as a 0/1 explanatory variable.

Results

Sulfate reduction and methanogenesis in the sediment

Sulfate concentrations decreased exponentially with depth in the top 3 or 4 cm and remained at a constant low level ($SO_4\infty$) at greater depths (Fig. 1A). The value for $CSO_4\infty$ fluctuated between 0 and 17.7 μM with an average value of 7.6 ± 6.1 μM. During most of the year the $CSO_4\infty$ was

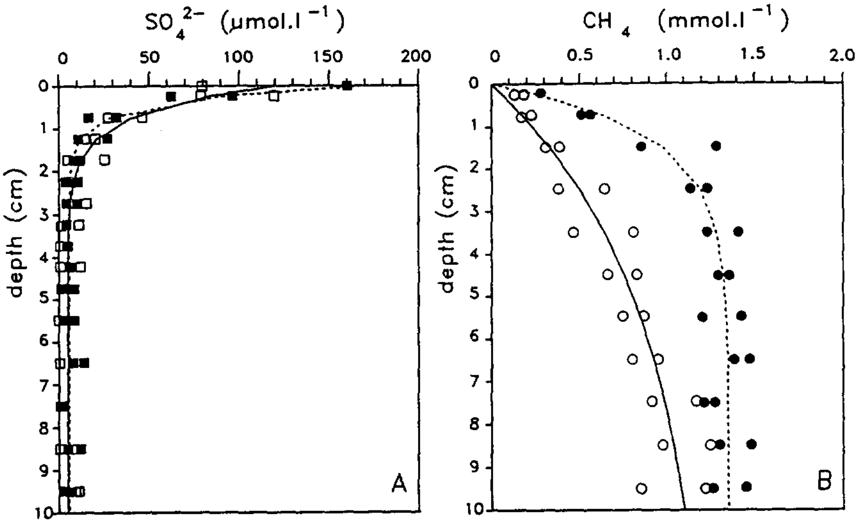

Fig. 1. Concentration profiles of sulfate (A) and methane (B) in sediment cores of station Kievitsarea in September (closed symbols) and in February (open symbols). Lines represent fitted values (September -----, February ———).

reached within 4 cm from the sediment-water interface. However, in winter and early spring sulfate penetrated as deep as 8 cm in some cases. Concentration profiles of sulfate could be well described by the models which were used, p being in most cases < 0.01 (Table 1). The value for the attenuation constant for sulfate reduction showed large seasonal variations with high values in autumn and ranged from 0.07 to 2.88 cm^{-1}.

Methane concentration increased with depth towards a constant level (Fig. 1B). The average value of the methane concentrations at the sediment-water interface (CCH_4o) did not deviate significantly from zero, and calculations were repeated with $CCH_4o = 0$. The measured methane concentrations could be described reasonably by the model except for cores with low values for a, especially in winter and early spring. In these cores methane concentrations increased linearly with depth and could only be fitted when $CCH_4\infty$ was allowed to increase to extreme over-saturating values (Table 1). The values for the attenuation constant for the methane profiles ranged from 0.01 to 0.85 cm^{-1} and were always lower than the values for the sulfate profiles.

Sulfate reduction rates ranged from 3.2 to 44.6 $\mu mol.m^{-2}.h^{-1}$ and were somewhat lower in winter and early spring. During the rest of the year, the sulfate reduction rates were relatively constant between 20—40 $\mu mol.m^{-2}.h^{-1}$. The calculated CH_4 flux towards the surface layer showed

Table 1. Temperature, chlorophyll-a and primary production rate in the water and fitted values for the attenuation constant (*a*) and concentrations at infinite depth (C∞) for the sulfate and methane concentration profiles in the open lake and Kievitsarea for each sampling data.

sampling date	temp. °C	chl-a $\mu g.l^{-1}$	prim. prod. $mmol.m^{-2}.d^{-1}$	OPEN LAKE SO_4^{2-} profiles		CH_4 profiles		KIEVITSAREA SO_4^{2-} profiles		CH_4 profiles	
				a	C∞	a	C∞	a	C∞	a	C∞
03.15.88	6	93	35	0.24	0 ***	0.01	#4.20***	1.86	22.1***	0.19	1.23***
04.12.88	10	93	68	N.D.	N.D.	0.01	1.42**	0.07	0	0.01	1.42***
05.10.88	16	63	98	0.93	5.3	0.24	0.69***	1.32	5.2***	N.D.	N.D.
06.07.88	16	156	124	0.61	0 **	0.04	#2.71***	1.75	17.8***	0.03	#3.53***
07.05.88	16	113	180	0.88	10.8***	0.30	0.62***	0.88	7.9**	0.08	#2.11**
08.02.88	19	156	200	1.10	17.9**	0.31	0.50***	0.72	0.7***	0.34	0.79**
08.30.88	18	108	172	0.83	5.7***	0.77	0.41**	0.95	4.7***	0.34	0.62**
09.27.88	14	98	88	1.38	8.0***	0.62	0.88***	1.08	6.1***	0.50	0.93**
10.25.88	12	103	50	2.88	6.3***	0.24	1.11***	2.71	5.8***	0.85	1.35**
11.22.88	5	92	24	2.45	17.7***	0.42	0.69***	2.37	14.2***	0.47	1.21**
12.13.88	5	90	18	1.70	7.9***	0.01	5.54***	1.88	5.6**	0.02	#6.04**
01.10.89	6	117	43	1.08	4.9***	0.05	2.11***	1.60	4.9***	0.20	1.28**
02.07.89	6	80	48	0.23	0 ***	0.01	#3.95***	1.67	2.0***	0.35	0.95**

** $p < 0.1$, *** $p < 0.01$, # extreme oversaturation, N.D. not determined large variations and ranged from 5.5 to 342 μmol.m^{-2}.h^{-1}. In the open lake station a clear seasonal trend in the methane fluxes could be seen with high values in autumn and low values in winter and early spring (Fig. 2). Values for Kievitsarea followed the same pattern (not shown).

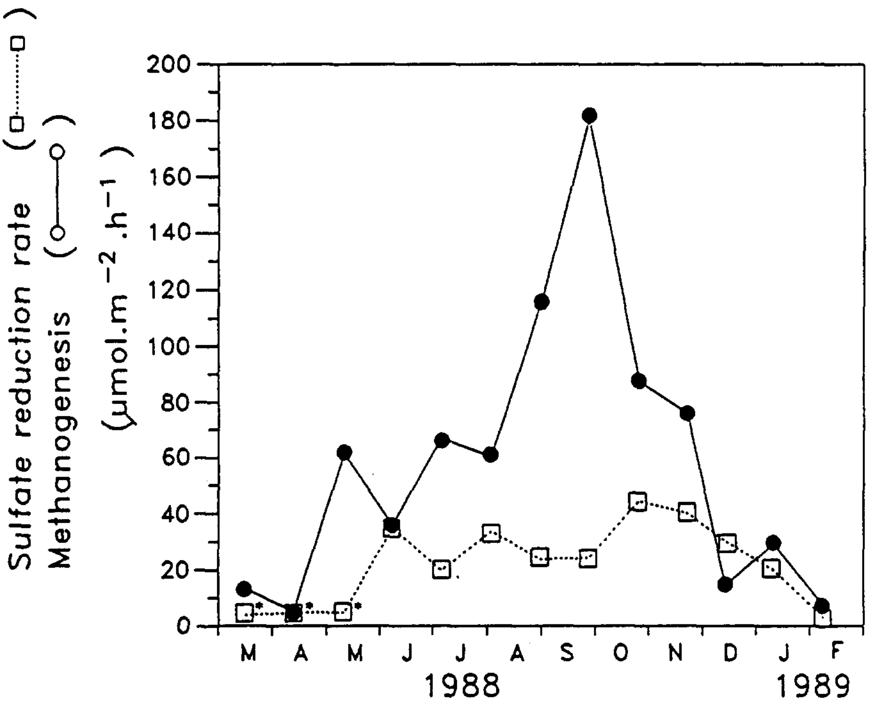

Fig. 2. Seasonal variation in the open lake in sulfate reduction rate (□) and methanogenesis (○) calculated from the fitted concentration profiles. *represent unreliable results (see Table 1).

The total anaerobic mineralization of organic carbon ranged from 0.7 mol.m^{-2}.y^{-1} in the open lake to 1.2 mol.m^{-2}.y^{-1} at Kievitsarea. At high total anaerobic mineralization rates, the contribution of methanogenesis became increasingly important (Fig. 3). In autumn when anaerobic mineralization was at a maximum of 750 μmol.m^{-2}.h^{-1}, methanogenesis accounted for approximately 80% of the total anaerobic mineralization. Sulfate reduction rates levelled off at a constant rate of 40 μmol.m^{-2}.h^{-1}, (Fig. 3). During this period the primary production rates decreased form 200 to 18 mmol carbon.m^{-2}.d^{-1} (Table 1). The chlorophyll-a content of the water also showed a decrease from 156 to 90 μg.l^{-1} (Table 1).

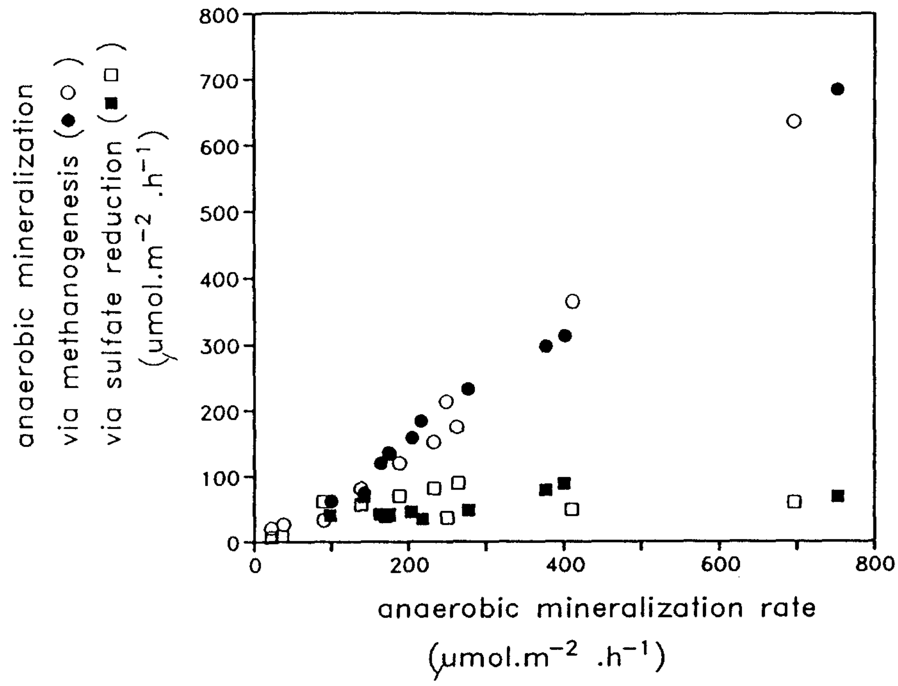

Fig. 3. Sulfate reduction rates (□) and methane fluxes (○) in Lake Loosdrecht (open symbols: open lake, closed symbols: Kievitsarea) as a function of the total anaerobic mineralization.

Methane production in laboratory experiments

The methane production rate in batches was linear with time. The production rate in the sediment decreased exponentially with depth. The highest values were measured in August (120 nmol.g^{-1}.d^{-1}). Production rates were high in late summer and at or below detection limit in winter (Fig. 4). The methane production rate in batches was integrated over depth (Table 2). The 0—0.5 cm layer was excluded from the integration to allow for a better comparison with the calculated fluxes. *In situ*, oxygen and sulfate are present in the surface layer and probably reduce methanogenesis strongly. The integrated methane production was dependent on temperature ($p < 0.01$) with an average Q_{10} value in the range from 10—20 °C of 2.4. In general the integrated methane production in batches was lower than calculated fluxes (Table 2). Especially at temperatures below 10 °C hardly any significant production was measured in batches while the fluxes that were calculated from the concentration gradients ranged from 5.5—91.6 µmol.m^{-2}.h^{-1}.

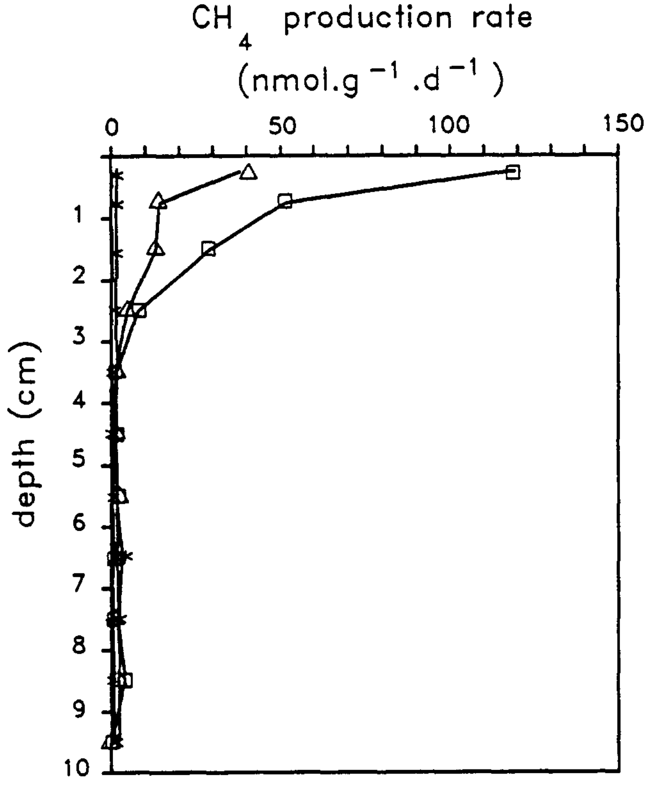

Fig. 4. Methane production in batches of different sediment layers of sediment columns sampled in August (□), September (△), and December (*) in the open lake.

Statistical analysis

The independent variables which sequentially entered the redundancy analysis and the proportion of variance in the dependent variables which additionally could be explained are listed in Table 3. The value of 0.21 for TEMP for additional variance explained, implies that 21% of the variance in the complete data-set can be explained by temperature. The total percentage of variance explained was 54%. The major fraction of the total explained variance was described by the first two axes (35.7% on the first and 8.2% on the second axis). The residual fraction of 9.1% was described by axes with a higher dimension. Addition of the primary production rate and chlorophyll-a content as explaining variables to the RDA gave no significant improvement of the explained variance. This is due to the high correlation of the primary production rate and chlorophyll-a content of

Table 2. Methane fluxes (μmol.m^{-2}.h^{-1}) towards the sediment-water interface, in situ values calculated from concentration profiles and integrated batch values, for the open lake and Kievitsarea from each sampling date.

sampling date	Open lake in situ	batch	Kievitsarea in situ	batch
03.15.88	13.3	1.0	64.6	4.0
04.12.88	N.D.	N.D.	5.5	13.5
05.10.88	61.6	16.4	N.D.	115
06.07.88	35.9	65.1	40.3	59.0
07.05.88	67.2	6.6	60.5	37.8
08.02.88	60.2	28.1	106	118
08.30.88	115	54.7	79	165
09.27.88	181	30.9	155	168
10.25.88	87.5	63	342	91.7
11.22.88	76.0	B.D.	149	B.D.
12.13.88	14.6	10.0	25.0	B.D.
01.11.89	29.3	2.4	67.1	7.2
02.07.89	67.6	B.D.	91.6	B.D.

N.D. not determined
B.D. below detection limit (1 μmol.m^{-2}.h^{-1})

Table 3. Sequence and additional proportion of variance explained of the environmental variables used in the Redundancy Analysis.

	Variance explained Additional	Cumulative
Temperature (TEMP)	0.21	0.21
Change in primary production rate (δPROD)	0.15	0.36
Station (Open Lake vs Kievitsarea)	0.10	0.46
All other variables (none significant)	0.08	0.54

the water with temperature (Table 4). The introduction of the change in primary production rate (δPROD) added 0.15 to the explained variance. δPROD is not correlated with temperature and in the correlation biplot the vectors of TEMP and δTEMP describe almost a 90° angle (Fig. 5).

The correlation biplot points out that the methane flux towards the sediment-water interface increased with a decrease in primary production rate and a decrease in temperature as can be seen from the opposite direction of the vectors for δPROD and δTEMP versus CH_4. Although temperature is an important variable in explaining the complete dataset, it contributed only slightly to the explained variance of the calculated *in situ*

Table 4. Correlation matrix of environmental variables measured in Lake Loosdrecht; TEMP is temperature, CHLA is the chlorophyll-a concentration, PROD is the primary production rate, δPROD and δTEMP are the overall change in temperature and primary production between two successive sampling dates.

	TEMP	CHLA	PROD	δTEMP	δPROD
TEMP	—	0.56*	0.92***	0.35	0.09
CHLA		—	0.61*	0.22	0.32
PROD			—	0.39	0.32
δTEMP				—	0.68**

$n = 13$, $*p < 0.05$, $**\pi < 0.01$, $***p < 0.001$.

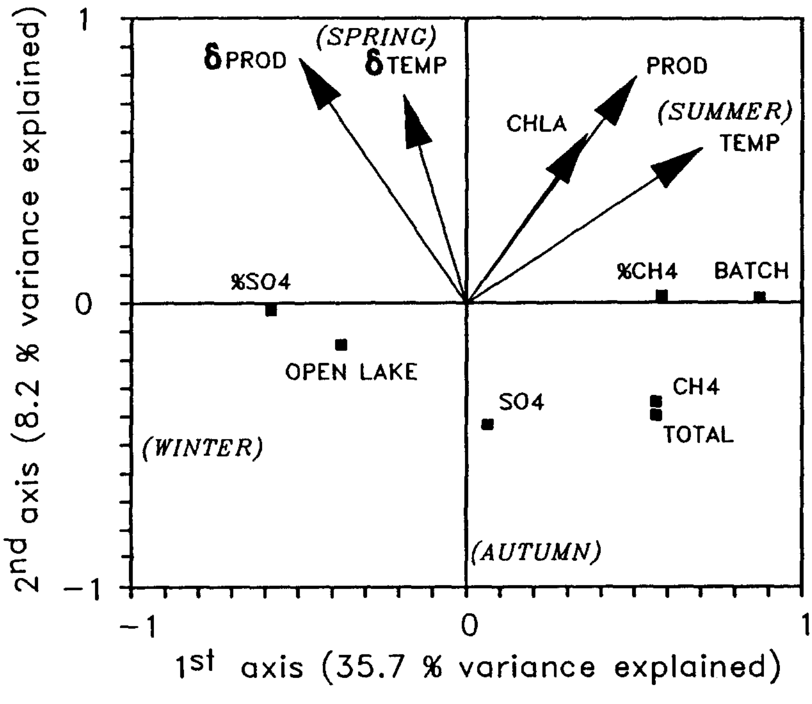

Fig. 5. Correlation biplot of dependant variables. Environmental variables are represented by arrows (see Table 3). An approximate indication of the seasons is given in brackets.

methane flux (≈ 60° angle between CH_4 and TEMP). Methane production in batches reacted more intensely on temperature. As the total anaerobic mineralization (TOTAL) is strongly determined by CH_4, these vectors are positioned closely together. Sulfate reduction was low in spring as can be inferred from the opposite direction of SO_4 and δTEMP and which is in accordance with Fig. 2. The contribution of sulfate reduction to the total anaerobic mineralization (%SO4) increased at low temperatures and tended to be higher at positive δPROD in spring. Sulfate reduction appears to be more important in the open lake than at the Kievitsarea.

Discussion

Calculated in situ fluxes of sulfate and methane

The anaerobic mineralization rate in Lake Loosdrecht, was highest in autumn. Probably this can be ascribed to an increase in sedimentation rate at the end of the growing season. However in a shallow holomictic lake, it is difficult to distinguish net deposition from total deposition due to wind and wave influences (Håkanson & Jansson 1983). Changes in chlorophyll-a and primary production rate in the water might provide a rough indication of the sedimentation of easily degradable organic matter, since a decrease in chlorophyll-a and primary production rate is partly caused by decay and sedimentation of algae. The inverse relation between the decrease in primary production rate in the lake and the total anaerobic mineralization suggests, as expected, sedimentation as a driving force for the anaerobic mineralization rate. The impact of sedimentation in autumn is emphasized by the correlations of fluxes of methane, phosphates and ammonium in the anaerobic sediment which were reported earlier (Sinke et al. 1990). On a yearly basis only 1.5—4% of the primary production in the lake (30—45 mol organic carbon.m^{-2}.y^{-1}, Van Liere 1986), is channeled via anaerobic processes. A comparable low value (3%) has been found for shallow Lake De Grote Rug which has a primary production of 25 mol.m^{-2}.y^{-1} (Adams & Van Eck 1988). Temperature which is often of major importance in determining the anaerobic mineralization rate in deep systems (Klump & Martens 1989; Kelly & Chynoweth 1981) appears to play only a minor role in shallow Lake Loosdrecht.

There was a large seasonal variation in the relative importance of sulfate reduction and methanogenesis. The relative importance of sulfate reduction in the anaerobic mineralization varied during the year and appeared to be highest in winter and early spring when the total anaerobic mineralization rates were low, 20—100 μmol carbon.m^{-2}.h^{-1}. During this period sulfate reduction was dominant in the bulk of the sediment and penetrated as deep as 8 cm in some cores. Sulfate reduction appeared to outcompete methanogenesis, which was very low, successfully. In autumn, at high total anaerobic mineralization rates of 600 μmol organic carbon.m^{-2}.h^{-1}, methanogenesis accounted for over 80% of the anaerobic mineralization. Sulfate reduction levelled off at 40 μmol.m^{-2}.h^{-1} indicating an overall sulfate limitation. Diffusion of sulfate from the overlying water probably could not keep up with the sulfate reduction rate which was also indicated by the sulfate penetration depth which extended no further than 3.5 cm. The results suggest that in the sediment of this shallow eutrophic lake, the anaerobic mineralization is determined by a seasonal shift from a limitation by sulfate in summer and autumn towards a limitation by easily degradable organic matter in winter and early spring. In deep lakes the relative importance of sulfate reduction seems to be less liable to seasonal variation and can often be quantified as a percentage of total anaerobic mineralization as for instance 13% in Wintergreen Lake (Lovley et al. 1982) and 37% in Lake Washington (Kuivila et al. 1989).

At greater depths in the sediment an average $CSO_4\infty$ concentration of sulfate of 7.6 ± 6.1 μM was found. This threshold concentration below which no sulfate reduction occurred, is in the same range as values reported for Lawrence Lake (30—40 μM, Lovley & Klug 1983), Lake Washington (18 μM, Kuivila et al. 1989) and Lake Gerritsfles (0—20 μM, Feijtel et al. 1989). Pooling all profiles from summer and winter, Feijtel et al. (1989) calculated a higher average threshold in summer (10 μM) than in winter (4 μM). In Lake Loosdrecht no such temperature effect could be detected but the threshold concentration tended to decrease with decreasing values for the attenuation constant for sulfate reduction. So, generally, at low sulfate reduction rates low threshold values were calculated. The relation between threshold concentration and sulfate reduction rate has hardly been investigated for field situations but might be influenced by changing physiological conditions of the sulfate reducing population. Ingvorsen et al. (1984) reported that the sulfate threshold concentration of cell suspensions of chemostat cultures of *Desulfobacter postgatei* decreased after prolonged energy starvation.

The values for $CCH_4\infty$ were in most cores below the saturation values for methane in water. This same phenomenon was also observed for this sediment by Sweerts et al. (in press). During most of the year the CCH_4o did not deviate significantly from zero, this can be caused by the rapid diffusion into the overlying water or by the oxidation of methane in the aerobic surface layer of the sediment. Sweerts et al. (in press) could not detect any methane release from sediment cores of Lake Loosdrecht which were incubated under standardized conditions, suggesting that the entire methane flux from the anaerobic sediment was oxidized in the small surface layer. Based on the oxygen consumption rates determined by Sweerts & coworkers (1988, in press), the oxygen consumption rate for our sediment cores sampled in October is estimated at 0.67 $mmol.m^{-2}.h^{-1}$. Assuming a stoichiometry of 1.5 moles O_2 per mole CH_4 (Joergensen & Deng 1983), the complete oxidation of all methane produced (0.34 $mmol.m^{-2}.h^{-1}$) would account for 76% of the total oxygen consumption in the sediment. The rest of the oxygen consumption can be ascribed to aerobic mineralization and, to a lesser extent, to nitrification and the oxidation of iron and sulfide. Hardly any results are reported in literature on the seasonal competition between all these oxygen consuming processes.

Calculated methane production versus methane production in batches

The methane production measured in batches depicted an exponential decrease with depth. The major part of the methane was produced in the top layers (0—4 cm) of the sediment just as found by Kelly & Chynoweth (1981) for Frain's Lake and Third Sister Lake. At greater depths in the sediment extremely low rates of methanogenesis (< 1 $nmol.g^{-1}.d^{-1}$ below 5 cm depth) were determined. This steep exponential decrease in methane production could be caused by a limitation of degradable organic material thus suggesting a low net sedimentation rate and a high breakdown of organic matter. Loss of methane due to anaerobic methane oxidation is probably only a minor fraction of all methane produced (Zehnder & Brock 1980).

The integrated methane production rates measured in batches and those calculated from the profiles showed large differences, especially in winter and early spring. The methane production rates which were calculated using the concentration profiles were usually higher than that of the batch values. Apparently, the escape of methane from the sediment by bubbles which was reported to be important in Cape Lookout Bight (Martens & Klump 1984), is only of minor importance in Lake Loosdrecht. Recent measurements during a warm (26 °C) summer's day did not indicate any methane above the sediment-surface (unpublished results). During integration of the methane production in batches the top layer of the sediment was left out. This might have resulted in an underestimation of methanogenesis if methanogenesis and sulfate reduction occurred simultaneously in the top layer. Winfrey & Zeikus (1977) reported that addition of non-limiting concentrations of acetate reversed the inhibition of methanogenesis by sulfate. So, especially in autumn at a high supply of easily degradable organic matter, the co-occurance of sulfate reduction and methanogenesis is likely to occur in the top layer of the sediment. This could contribute to the observed discrepancy between batch and profile measurements in autumn. The windsheltered situation of Kievitsarea where a more pronounced sedimentation is expected, might contribute to the discrepancy.

In winter hardly any methane production could be detected in batches though the calculations from the concentration profiles indicated methane fluxes of 5.5—91.6 μmol.m^{-2}.h^{-1}. We suggest that in winter hardly any methane production occurs and that the calculated methane flux in the sediment cores originates from the methane pool at a greater depth in the sediment. Upward diffusion of fossil methane from older geological layers has been described for Lake Lungern by Bossard & Gächter (1981). Comparing methane fluxes in hydroxide poisoned and non-poisoned bell-jars, these authors concluded that 41.4 μmol.m^{-2}.h^{-1} originated from deeper layers. ^{14}C radiocarbon measurements confirmed that the methane was from older deposits (Bossard & Gächter 1981).

Diagenetic model

Berner's diagenetic model is generally used to describe sulfate and carbon dioxide profiles in marine or brackish sediments where sulfate reduction prevails (Berner 1980; Klump & Martens 1989). In Lake Loosdrecht the model was also applied to describe methane profiles but appeared to be reliable only when methanogenesis was predominant in the anaerobic mineralization process. In winter the measured methane flux resulted mainly from upward diffusion and the model could not be applied.

The values for the attenuation constant for sulfate reduction with an average of 1.32 ± 0.75 cm^{-1} and a maximal value of 2.88 cm^{-1}, are much higher than the values found for Cape Lookout Bight (0.54 cm^{-1}, Klump & Marten 1989) and Lake Gerritsfles (0.33—0.46 cm^{-1}, Feijtel et al. 1989). High values for the attenuation constant, a, can be caused by high first order constants for the degradation of organic matter, by a low sedimentation rate or by a combination of both. As the highest values for a were calculated in autumn when sedimentation can be expected, we suggest that variations are mainly due to changes in the rate constant caused by the supply of easily degradable organic matter. As the sedimentation rate is independant of depth (Berner 1980), the difference between the attenuation constants for sulfate reduction and methanogenesis, can be ascribed exclusively to differences in the first order constants for the degradation of organic matter. Our results from the batch experiments indicate that the amount of degradable organic matter decreases exponentially with depth which can be an explanation for the observed lower values of the attenuation constant for methanogenesis. These results conform the diagenetic model, which also predicts an exponential decrease of degradable organic matter with depth (Berner

1980). In earlier observations on phosphatase activity and chlorophyll-a content in the sediment of Lake Loosdrecht, an exponential decrease with depth of both parameters was reported and suggested to be a reflection of the amount of degradable organic matter (Sinke et al. 1991).

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References

Adams DD & Van Eck GThM (1988) Biogeochemical cycling of organic carbon in the sediments of the Grote Rug reservoir. Arch. Hydrobiol. Beih. Ergebn. Limnol. 31: 319—330

Berner RA (1964) An idealized model of dissolved sulfate distribution in recent sediments. Geochim. Cosmochim. Acta 28: 1497—1503

Berner RA (1980) Early diagenesis, a theoretical approach. Princeton University Press, New Jersey

Bossard P & Gächter R (1981) Methan- und Sauerstoffhaushalt im Mesotrophen Lungernsee. Schweiz. Z. Hydrol. 43(2): 219—252

Broecker WS & Peng TH (1974) Gas exchange rates between air and sea. Tellus XXVI, 21—35

Capone DG & Kiene RP (1988) Comparison of microbial dynamics in marine and freshwater sediments: Contrasts in anaerobic carbon catabolism. Limnol. Oceanogr. 33: 725—749

Cappenberg TE, Hordijk CA & Hagenaars CPMM (1984) A comparison of bacterial sulfate reduction and methanogenesis in the anaerobic sediments of a stratified lake ecosystem. Arch. Hydrobiol. Beih. Ergebn. Limnol. 19: 191—199

Feijtel TC, Salingar Y, Hordijk CA, Sweerts J-PRA, Van Breemen N & Cappenberg TE (1989) Sulfur cycling in a dutch moorland pool under elevated atmospheric S-deposition. Water, Air and Soil Poll. 44: 215—234

Gabriel KR (1971) The biplot graphic display of matrices with application to principal component analysis. Biometrika 58: 453—467

Håkanson L & Jansson M (1983) Principles of lake sedimentology. Springer-Verlag Berlin, New York

Hordijk CA, Hagenaars CPMM & Cappenberg TE (1985) Kinetic studies of bacterial sulfate reduction in freshwater sediments by High-pressure Liquid Chromatography and microdestillation. Appl. Environ. Microbiol. 49(2): 434—440

Ingvorsen K, Zehnder AJB & Jørgensen BB (1984) Kinetics of sulfate and acetate uptake by Deusulfobacter postgatei. Appl. Environ. Microbiol. 47: 403—408

Ingvorsen K, Zeikus JG & Brock TD (1981) Dynamics of bacterial sulfate reduction in a eutrophic lake. Appl. Environ. Microbiol. 42: 1029—1036

Joergensen L & Degn H (1983) Mass spectrometric measurements of methane and oxygen utilization by methanotrophic bacteria. FEMS Microbiology Letters 20: 331—335

Kelly CA & Chynoweth DP (1981) The contributions of temperature and of the input of organic matter in controlling rates of sediment methanogenesis. Limnol. Oceanogr. 26: 891—897

Kelly CA, Rudd JWM, Cook RB & Schindler DW (1982) The potential importance of bacterial processes in regulating rate of lake acidification. Limnol. Oceanogr. 27: 868—882

Klump JV & Martens CS (1989) The seasonality of nutrient regeneration in an organic-rich coastal sediment: Kinetic modeling of changing pore-water nutrient and sulfate distributions. Limnol. Oceanogr. 34: 559—577

Kuivila KM, Murray JW, Devol AH & Novelli PC (1989) Methane production, sulfate reduction and competition for substrates in the sediments of Lake Washington. Geochim. Cosmochim. Acta 53: 409—416

Li Y-H & Gregory S (1974) Diffusion of ions in sea water and in deep sea sediments. Geochim. Cosmochim. Acta 38: 703—714

Lovley DR, Dwyer DF & Klug MJ (1982) Kinetic analysis of competition between sulfate reducers and methanogens for hydrogen in sediments. Appl. Environ. Microbiol. 43: 1373—1379

Lovley DR & Klug MJ (1983) Sulfate reducers can outcompete methanogens at freshwater sulfate concentrations. Appl. Environ. Microbiol. 45: 187—192

Martens CS & Klump JV (1984) Biogeochemical cycling in an organic rich coastal marine basis 4. An organic carbon budget for sediments dominated by sulfate reduction and methanogenesis. Geochim. Cosmochim. Acta 48: 1987—2004

Ralston M (1988) Derivative-Free non-linear regression-AR. In: Dixon WJ (Ed) BMDP Statistical Software Manual (pp 389—417). University of California Press, Berkeley, Los Angeles

Rao CR (1964) The use and interpretation of principal component analysis in applied research. Sankhya, A, 26: 329—358

Reeburgh WS (1967) An improved interstitial water sampler. Limnol. Oceanogr. 12: 163—165

Robinson JA & Tiedje JM (1984) Competition between sulfate reducing and methanogenic bacteria for H_2 under resting and growing conditions. Arch. Microbiol. 137: 26—32

Sinke AJC, Cornelese AA, Keizer P, Van Tongeren OFR & Cappenberg TE (1990) Mineralization, porewater chemistry and phosphorus release from peaty sediments in the eutrophic Loosdrecht lakes. Freshwater Biol. 23: 587—599

Sinke AJC, Cornelese AA & Cappenberg TE (1991) Phosphatase activity in sediments of the Loosdrecht lakes. Verh. Internat. Verein. Limnol. 24: 719—721

Sweerts J-PRA & Cappenberg TE (1988) Use of microelectrodes for measuring oxygen profiles in lake sediments. Arch. Hydrobiol. Beih. 31: 365—371

Sweerts J-PRA, Dekkers TMJ & Cappenberg TE (in press) Methane oxidation at the sediment-water interface of a shallow eutrophic lake (Lake Loosdrecht) and a deep meso-eutrophic lake (Lake Vechten)

Ter Braak CJF (1987a) CANOCO- a FORTRAN program for canonical ordination by (partial) (detrended) (canonical) correspondence analysis, principal component analysis and redundancy analysis. TNO Institute of Applied Computer Sciences, Statistics Department, Wageningen

Ter Braak CJF (1987b) Ordination. In: Jongeman RH, Ter Braak CJF & Van Tongeren OFR (Eds) Data Analysis in Community Landscape Ecology. Pudoc, Wageningen

Van Liere L (1986) Loosdrecht lakes, origin, eutrophication, restoration and research programme. Hydrobiol. Bull. 20: 9—15

Van Liere L, Van Ballegooijen L, De Kloet WA, Siewertsen K, Kouwenhoven P & Aldenberg T (1986) Primary production in the various parts of Loosdrecht lakes. Hydrobiol. Bull. 20: 77—85

Winfrey MR & Zeikus JG (1977) Effect of sulfate on carbon and electron flow during microbial methanogenesis in freshwater sediments. Appl. Environ. Microbiol. 33: 275—281

Winfrey MR & Zeikus JG (1979) Anaerobic metabolism of immediate methane precursors in Lake Mendota. Appl. Environ. Microbiol. 37: 244—253

Zehnder AJB & Brock TD (1980) Anaerobic methane oxidation: occurance and ecology. Appl. Environ. Microbiol. 39(1): 194—204