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Measurements of Methylation Balance in Southern Indian Lake and Granville Lake, Manitoba, and in Sokatisewin Lake, Saskatchewan, 1988

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Executive Summary

Measurements of the microbial mercury methylation balance were continued in Methyl Bay on Southern Indian Lake and in Granville Lake, the long-term reference lake, in the summer of 1988. Specific rates of mercury methylation and demethylation and the methylation balance, indicated by the M/D ratio, in flooded sediments from Methyl Bay all were significantly elevated compared to rates in sediments from the nearshore zone of Granville Lake. The elevated methylation balance remains restricted to the flooded zone of Southern Indian Lake; M/D in sediments from the offshore zone was as low or lower than found in Granville Lake. The measurements have now been made at the sites for five consecutive years. There now is evidence that both methylation and demethylation activity in the flooded sediments are declining relative to rates in Granville Lake. This decline appeared to have started in 1987 and continued in 1988 and is generally consistent with our knowledge of the cause and the projected pattern of decline. Despite the declines in methylation and demethylation rates, there is no evidence of a corresponding decline in the methylation balance. The declines in methylation and demethylation have been compensatory.

Sokatisewin Lake, also known as the Island Falls reservoir, located on the Churchill River at Sandy Bay, Saskatchewan, was sampled as a test of the validity of a model developed in 1986 to describe the rates of decline of methylation, demethylation, and the methylation balance in northern reservoirs. Specific methylation rates in Sokatisewin Lake were significantly higher than in Granville Lake and were significantly lower than in Methyl Bay. Specific demethylation rates were significantly lower than in both Methyl Bay and Granville Lake. When all of the methylation and demethylation data collected

over the past three years are considered together the model described in 1986 appears, in general, to be valid. Rates of both methylation and demethylation decline over time at a negative exponential rate. Demethylation activity is expected to return to background levels about 50 years after impoundment, as originally predicted. However, methylation activity is not expected to return to background levels for 60-159 years after impoundment. The methylation balance is therefore expected to remain elevated as long as methylation is elevated.

The results of this study indicate that the methylation and demethylation activity measured in flooded sediments from Sipiwesk Lake in 1987 are high, or at least at the high end of the range expected, given the age of the reservoir. This appears to be a result of the level regulation regime. Low water levels in summer permit the re-vegetation of the drawdown zone. This fresh organic matter is introduced to the reservoir and made available to the bacteria when levels again rise.

The roles of methanogenic and sulfate reducing bacteria in methylation and demethylation in Methyl Bay and Granville Lake were examined using the specific metabolic inhibitors bromoethane sulfonic acid to inhibit methanogens and sodium molybdate to inhibit the sulfate reducers. Methanogens and sulfate reducers both were important contributors to methylation and demethylation in Granville Lake sediments but were not the only mediators of these processes. Each group accounted for about 33% of methylation activity and 25% of demethylation activity at the nearshore stations. Conditions in the flooded zone of Methyl Bay appear to have specifically selected for these groups. Methanogens and sulfate reducers jointly accounted for all of methylation and demethylation activity in sediments from the flooded zone. The share of total methylation conducted by the methanogens was about the same as in Granville

Lake while the share attributable to the sulfate reducers was twice that in Granville Lake. This suggests a specific selection for sulfate reducing methylators in the flooded sediments. The relative importance to demethylation of both methanogens and sulfate reducers was higher in the flooded sediments but again the sulfate reducers appear to have been differentially selected. The finding that the same bacterial groups are responsible for both methylation and demethylation indicates that it may be difficult to develop mitigative measures based on the selective inhibition of the methylators or stimulation of the demethylators. Other approaches, such as investigation of the factors regulating the transmission of the gene responsible for producing the demethylation enzyme within the bacterial community should be considered.

The occurrence of elevated net methylation activity in flooded sediments is thought to be the result of the stimulation of total bacterial activity by the flooded organic matter. Measurements of total microbial activity were made at all of the methylation sampling sites to test this hypothesis. Demethylation rates were positively correlated with microbial activity but no simple linear relationship between methylation and microbial activity could be defined. The results of the inhibition studies indicate that the composition of the methylating and demethylating communities in flooded sediments can differ from that in natural lake sediments. Increases in the activity of specific microbial groups may be more important than increases in total microbial activity.

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Introduction

This study was undertaken as part of continuing research directed to estimating the duration of mercury problems related to flooding in the region of northern Manitoba covered by the Northern Flood Agreement. Previous studies have shown that the elevated concentrations of mercury in fish from lakes flooded by the Churchill River diversion are the product of increased net rates of microbial mercury methylation in flooded areas (Hecky et al. 1987). The net mercury methylation rate represents the balance between the bacterial rates of methylation (production) and demethylation (degradation). These increased net methylation rates are the product of the differential stimulation of the methylating bacteria over the demethylating bacteria by the high concentrations of organic matter in the flooded terrestrial materials (Ramlal et al. 1987; Ramsey and Ramlal 1987b; Ramsey 1987, 1988).

Two courses of study are being followed in the determination of the duration of the problem. The microbial mercury methylation balance (defined below) in flooded sediments and mercury concentrations in fish from Southern Indian Lake are being monitored annually to determine if decreases are occurring over the near term and measurements of the methylation balance in flooded sediments from older reservoirs provide a measure of long term changes in the stimulatory effect of the flooded materials. The results of all studies to date indicate that the problem will be long-lived. Mercury concentrations in fish have not decreased significantly in the first ten years after impoundment (Bodaly and Strange 1987). The microbial methylation balance in the flooded zone of the lake has not decreased in the past four years that measurements have been made. A model derived from methylation balance measurements made in Southern Indian Lake and Laurie Reservoir (Manitoba) and in the Ogoki Reservoir (northwestern Ontario) in 1986 predicted that net

methylation in northern reservoirs will not return to background levels for 50 to 60 years after impoundment (Ramsey 1987).

The measurements of methylation balance made in Sipiwesk Lake in 1987 were intended to test the predictive ability of the model, but the results were inconclusive. Specific microbial rates of both methylation and demethylation were about twice those predicted by the model (Ramsey 1988). Although this result may indicate that the model is inappropriate it may instead indicate that the manner in which reservoir levels are managed may also prolong the problem. Extended periods of low water levels in summer, as occur on Sipiwesk Lake, allow the revegetation of the drawdown zone. When this fresh organic matter is flooded, microbial activity and consequently methylation and demethylation rates would be expected to be stimulated to a greater degree than would otherwise be predicted for a reservoir and the associated flooded organic matter of that age. Repeated events of this kind may extend the occurrence of high mercury levels in fish indefinitely (Ramsey 1988).

In the present study, the monitoring of microbial methylation in Southern Indian Lake was continued and the measurements were extended to Sokatisewin Lake (the Island Falls Reservoir) on the Churchill River in Saskatchewan. Sokatisewin Lake is not subject to the low summer water levels found in Sipiwesk Lake and therefore should provide a better test of the 1986 model.

The dominant role of organic matter in regulating the methylating bacteria and ultimately net methyl mercury production has become clear over the past several years of study. The organic matter is thought to stimulate methyl mercury production by increasing total bacterial activity. Other studies, reviewed by Beijer and Jernelov 1979, have established this link but to date it has not been possible to confirm this explanation in the Churchill River diversion due to the lack of suitable instrumentation. With the recent

acquisition of the necessary instrument by the Freshwater Institute, measurements of total bacterial activity have become possible and were included in the present study. Bacterial activity, as measured by the rate of DIC production, was measured in all sediment samples collected in this study .

The apparent longevity of the mercury problem in northern reservoirs in general, and particularly in flooded lakes in the area of the NFA, indicates the need for some means of mitigating the problem. Resuspension of sediments has been suggested as a possible remedial measure for the mercury problem in the English-Wabigoon River system (Rudd et al. 1983). However, increased turbidity was not found to be effective in reducing mercury concentrations in fish in the Churchill River diversion area (Hecky et al. 1987).

An alternative approach might be the selective stimulation of the demethylating bacteria and/or the inhibition of the methylators through the addition of a suitable substrate. This method requires a determination of the causative microorganisms and their metabolic pathways. The essential requirement of such an approach is that the methylators and demethylators belong to different metabolic groups. The results of a pilot study conducted in 1987 indicated this may be the case (Ramsey 1988). The study examined the effect of bromoethane sulfonic acid (BESA), a specific inhibitor of methanogenesis (Gunsalus et al. 1978), on rates of methylation and demethylation in a sample of flooded sediment from Methyl Bay. Methanogens accounted for the majority of methylation activity while demethylation activity increased with inhibition of the methanogens. This indicated that the demethylators were not methanogenic bacteria and that their activity was suppressed in the presence of methanogens. The identity of the demethylators could not be determined since only one inhibitor was used. In the present study the effects of methanogen inhibition, using BESA, on methylation and

demethylation in sediments from both Methyl Bay and Granville Lake were examined over the entire summer season both to verify the results of the pilot study and to examine seasonal variation in the contribution of methanogens. In an attempt to identify the demethylators, this study also examined the effects of sodium molybdate, a specific inhibitor of sulfate reduction (Postgate 1949; Saleh et al. 1964), on specific rates of methylation and demethylation. The sulfate reducers were selected for study because of their demonstrated importance in other studies of net microbial methyl mercury production and because of the potential for their competitive interaction with methanogens (Oremland and Taylor 1978).

Definition of Methylation Balance

The majority of mercury in fish muscle is in the organic methyl mercury form yet very little of the mercury present in the sediments and water of lakes and in the soils and vegetation flooded during reservoir formation exists in the methylated form (Westoo 1966, Fimreite et al. 1971). Concentrations of methyl mercury in the aquatic environment are regulated by the rates of the concurrent processes of its production (methylation) from mercuric mercury

$$Hg^{2+} + R\text{-}CH_3 \text{ ------> } CH_3Hg^+ + R^+$$

and its degradation (demethylation) to elemental mercury and methane

$$CH_3Hg^+ \xrightarrow[\text{organomercurial lyase}]{\text{NADPH, NADP}^+} CH_4 + Hg^{2+} \xrightarrow[\text{mercuric reductase}]{\text{NADPH, NADP}^+} Hg^0$$

by bacteria (e.g., Brosset 1981; Lexmond et al. 1976; Robinson and Tuovinen 1984). A change in the rate of either process will affect the availability of methyl mercury and it is the balance of these processes which ultimately determines the concentration in fish muscle. Clearly, there must be a shift in

this balance in favour of methylation for mercury levels in fish to increase.

The specific rate of mercury methylation is determined using the method developed by Furutani and Rudd (1980) which measures the rate of methylation of $^{203}Hg^{2+}$. The companion method uses $^{14}CH_3Hg^+$ to measure the specific rate of demethylation (Ramlal et al. 1986). Unlike other methods of studying methylation, both methods minimize sample incubation time and the quantity of mercury which must be added so that the microbial community is not altered from that in situ (Furutani and Rudd 1980, Ramlal et al. 1986).

The rates derived with each method are termed specific because both processes are dependent on mercury concentration and, since some mercury is added to the samples, the rates measured are specific to the quantity added. However, by adding the same quantity of mercury to all samples, differences in the specific rates between locations are considered to be proportional to differences in the in situ rate (Furutani and Rudd 1980). If necessary, the specific rates of methylation and demethylation can be converted to absolute rates if the concentrations of the bioavailable mercury species (Hg^{2+} for methylation, and CH_3Hg^+ for demethylation) are known. This calculation usually is not performed because of the problems associated with examining mercury speciation in sediments. The only practical limitation of leaving the measured rates in the specific form is that the difference between the specific methylation and specific demethylation rates does not yield an absolute net methylation rate. Instead, the ratio of these rates (M/D), or what we call the methylation balance, is used as an indicator of the relative difference in the net rate of methyl mercury production between locations. Since 1983, these methods have been employed in lakes and reservoirs in northern Manitoba and it is the results of these studies (Ramlal et al. 1986, 1987; Ramsey and Ramlal 1987 a and b), coupled with limnocorral experiments (Hecky et al. 1987), which

are responsible for the current understanding of the factors responsible for the elevation of fish mercury levels in reservoirs.

Description of the Study Area

The mean water level of Southern Indian Lake (Figure 1; latitude 57° N, longitude 99° W) was raised 3m in 1976 to facilitate the southward diversion of about 75% of the Churchill River flow to supply hydroelectric developments on the lower Nelson River (Newbury et al. 1984). Lake area increased by 21% due to impoundment (McCullough 1981). The outflow at Missi Falls was closed and the water diverted by gravity flow into the Rat and Burntwood River systems through a short canal at the southern end of South Bay (Figure 1). All samples from SIL were collected from a small isolated bay in region 2, hereafter called Methyl Bay (Figure 1). Methylation rates have been measured in Methyl Bay since 1982 and demethylation rates since 1983.

Granville Lake (Figure 1; latitude 56° 30' N, longitude 100° W) is a riverine lake on the Churchill River immediately upstream from SIL and has not been subjected to flooding. Samples were collected from the same area as in Ramsey and Ramlal (1987a and b) and Ramsey (1987a).

Sokatisewin Lake (Figure 1; latitude 55°30' N, longitude 102°20' W; appox. area 3800 ha), also known as the Island Falls Reservoir, was formed in 1929 by the construction of a hydroelectric generating station on the Churchill River at Sandy Bay, Saskatchewan. The depth of flooding at the dam is 17m. The extent of flooding is unknown as no pre-impoundment maps exist (Alex Banga, Saskatchewan Water Corporation, pers. comm.).

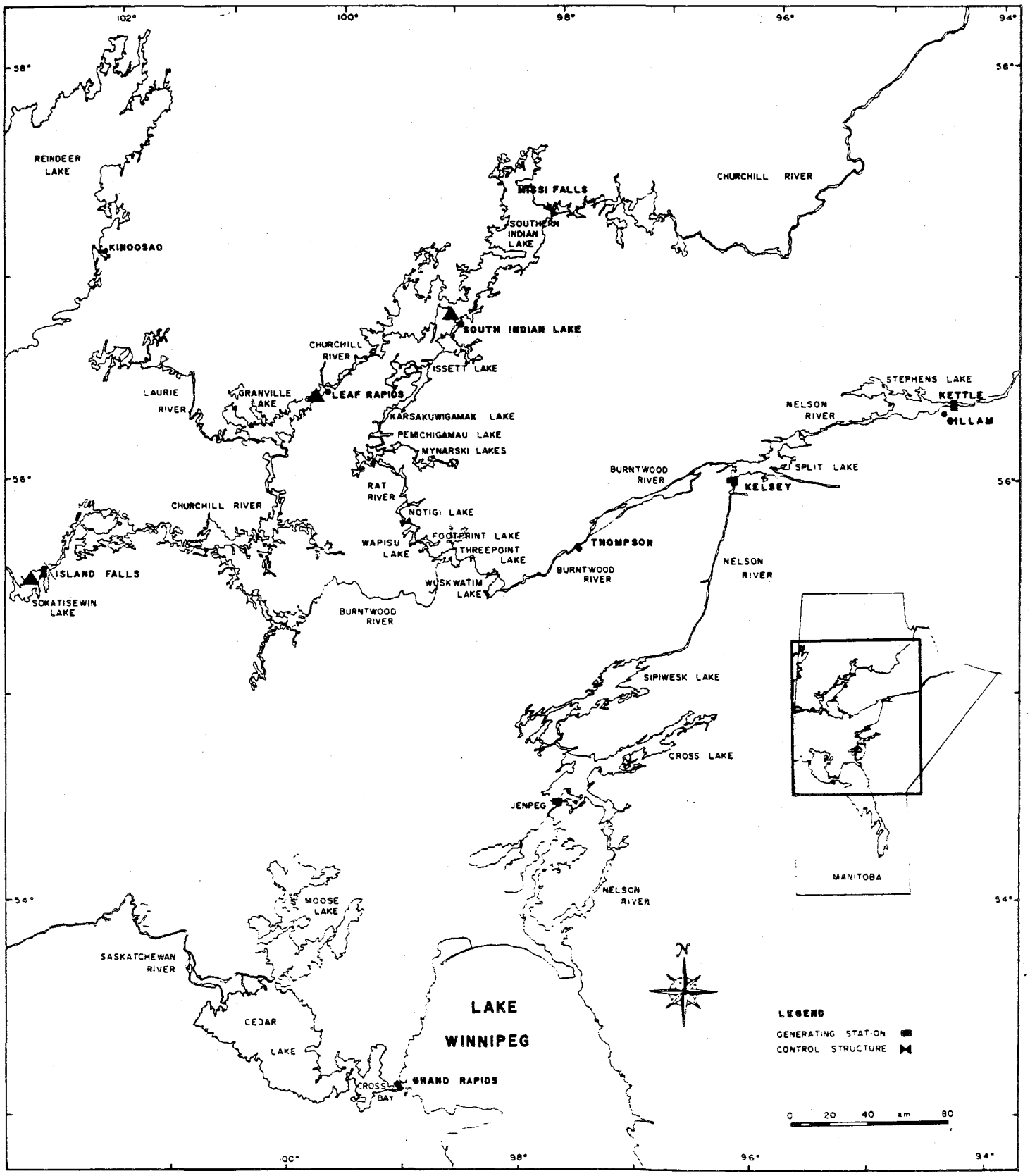


Figure 1. Location of methylation balance sampling sites (▲) in 1988.

Location of the Sampling Stations and Frequency of Sampling

Five surveys of methylation balance were conducted, at about 2 week intervals, over the 1988 summer season (Table 1). Four stations were sampled at all sites on each survey. At Methyl Bay, three of the stations were located within the flooded zone and the fourth station was located offshore over fine-grained sediments in the pre-flooding lake basin (Table 1). Three of the stations in Granville Lake were located in the nearshore zone, within 15 m of the lake shore, and the fourth station was located about 200m offshore from this location (Table 1). Three of the stations in Sokatisewin Lake were located in the nearshore flooded zone at about 3m depth. The fourth station was located approximately 400m offshore at about 10m depth (Table 1). It is not known if the offshore station was located over flooded terrain or within a pre-existing lake basin.

Methods

Sample Collection and Field Measurements

Sokatisewin Lake and Granville Lake were sampled by float plane (Cessna 185) under charter from RnD Aviation, Ltd., Leaf Rapids, Manitoba. Methyl Bay was sampled by boat from the Department of Fisheries and Oceans Field Station at South Indian Lake (Figure 1).

Mercury methylation and methyl mercury demethylation rates were measured on samples of surficial lake bottom sediment. These samples (2L volume) were collected using a 12 volt submersible bilge pump (Attwood model 4103, 360 gallons per hour) attached to a 15m length of 2cm (ID) Tygon tubing. The samples were packed in ice immediately following collection for shipping to the Agassiz North laboratory in Winnipeg and the assays were started within 12-36 hours of collection.

Table 1. Key to date of sampling and station type, location, and depth (m).

Station No.	Type	Location	Depth (m) July 4-5	July 17-19	July 25-27	August 8-10	August 22-24
Methyl Bay							
1	Flooded,	nearshore	2.4	2.0	1.5	2.0	1.8
2	Flooded,	nearshore	1.8	1.5	1.3	1.1	1.4
3	Flooded,	nearshore	1.4	1.5	1.5	1.1	1.4
4	Reference,	offshore	7.0	7.0	6.6	6.5	6.5
Granville Lake							
5	Reference,	nearshore	2.5	2.0	2.0	1.8	1.8
6	Reference,	nearshore	2.5	2.0	2.0	1.8	1.8
7	Reference,	nearshore	2.5	2.0	2.0	1.8	1.8
8	Reference,	offshore	6.3	7.8	11.3	6.8	7.8
Sokatisewin Lake							
9	Flooded,	nearshore	4.5	3.0	2.5	3.0	2.5
10	Flooded,	nearshore	4.5	3.0	2.5	3.0	2.5
11	Flooded,	nearshore	3.5	3.0	2.5	3.0	2.5
12	Uncertain,	offshore	10.8	11.0	11.0	11.5	11.0

Profiles of temperature, dissolved oxygen, and specific conductivity were measured at each site on all of the surveys; oxygen and temperature with a YSI model 58 temperature-oxygen meter and conductivity with a YSI model 33 S-C-T meter. Thermistor calibration was checked against a mercury thermometer and found to be within the specifications of the manufacturer. The oxygen function was calibrated each day before use. The calibration of the conductivity meter was periodically checked with a standard KCl solution and found to be within the specifications of the manufacturer. Because conductivity is a function of temperature, the measured values were corrected to the standard 25°C for comparison over time and among locations. This correction was based on measurements of conductivity with increasing temperature in the laboratory in water samples collected from the sampling sites. Water transparency was measured at each site with a 20 cm diameter black and white Secchi disk.

Laboratory Analyses

Sediment Chemistry

Aliquots of the sediment samples were removed for measurement of pH, Eh (redox potential), and dry weight, and for analysis of particulate carbon (as total carbon, carbonate, and by subtraction organic carbon), nitrogen (as total nitrogen), and total mercury concentrations. For mercury analysis and dry weight measurement, duplicate 5-10 ml aliquots of the sediment slurry were filtered on to separate ashed GF/C filters and dried at 60°C to constant weight. For carbon and nitrogen analysis, a single 5-10 ml aliquot of sediment slurry was dried in a plastic weigh-boat as above. Mercury analyses were performed according to the method of Armstrong and Uthe (1971) and the carbon and nitrogen analyses were performed using the methods of Stainton et al. (1977). Redox potential of the sediment slurry was measured electrometrically

using a platinum-Ag/AgCl combination electrode.

Specific Methylation and Demethylation Rate Measurements

Each sediment sample was mixed and 100 ml measured into each of six 125 ml glass reagent bottles for measurement of specific methylation and demethylation. Three of the replicate subsamples (two replicates and a blank) were used for each of the rate measurements. Microbial activity in the blanks was stopped with 3 mL of 4N HCl prior to radioisotope addition.

The rate of specific methylation was measured using the method of Furutani and Rudd (1980). The samples were injected with ^{203}Hg as $HgCl_2$ at a dose rate of 74 kBq (2 ug Hg)$^{-1}$ sample^{-1}. The specific rate of demethylation was determined using the method of Ramlal et al. (1986). These samples were injected with ^{14}C as CH_3HgI at a dose of 1450 Bq (0.40 ug Hg)$^{-1}$ sample^{-1}. The samples were shaken for one minute following isotope addition then incubated in the dark at near in situ temperature for 21-22 hours. All samples were incubated with air in the headspace of the incubation vessels and were not shaken or stirred during the incubation period. Microbial activity was halted following incubation with an addition of 4N HCl.

The alkylated ^{203}Hg produced in the methylation samples during the incubation was extracted from the sediment using the organic extraction technique of Furutani and Rudd (1980). The volatile ^{14}C produced in the demethylation samples was stripped from the acidified samples by bubbling with air, oxidized to CO_2, and trapped in a carbon dioxide trap consisting of the fluor ACS II (Amersham), methanol, and Protosol (DuPont), a carbon dioxide trapping agent (Ramlal et al. 1986). The ^{14}C and ^{203}Hg activities in the samples were determined by liquid scintillation counting.

The rate of methylation in sediment was first calculated in ng Hg

methylated (g dry sediment)$^{-1}$ h^{-1}. This value was used to calculate the specific rate of methylation i.e. the percentage of the added mercury methylated g^{-1} h^{-1} (Furutani and Rudd 1980). The specific rate of demethylation was calculated in a similar manner (Ramlal et al. 1986). The methylation balance, or M/D ratio, was calculated as the ratio of these specific rates.

Measurements of Microbial Activity

Microbial activity in the sediment samples was measured by measuring the rate of dissolved inorganic carbon (DIC) production in parallel with the microbial methylation and demethylation measurements. Two aliquots (35 mL) of each mixed sediment sample were transferred to two, 100 mL volume, glass serum bottles which were capped with silicone-coated rubber serum stoppers. Microbial activity in one of the two samples was stopped immediately with 200 uL of concentrated phosphoric acid. The remaining sample was incubated under the same conditions as the methylation and demethylation samples for 24 hours after which microbial activity was stopped with 200 uL of phosphoric acid.

Microbial Inhibition Experiment

The effects of methanogen inhibition and of inhibition of sulfate reducing bacteria on specific methylation and demethylation rates were examined on all five surveys in both Methyl Bay and Granville Lake. This involved two additional assays of methylation and demethylation on each sediment sample performed as described above but with the addition of the inhibitor. Methanogens were inhibited by the addition of 30 mM BESA and sulfate reducers were inhibited by the addition of 20 mM sodium molybdate prior to sample incubation. The effect of the inhibitors on total microbial activity was not

measured.

Statistical Analysis

The significance of differences in seasonal mean rates of methylation, demethylation, and M/D, and in mean sediment pH, organic carbon, total nitrogen, and total mercury concentrations was evaluated using one-way analysis of variance followed by Fisher's test of least significant difference. This technique is also termed the "protected" test of least significant difference (Snedecor and Cochran 1980) and its use is referenced in the text as PLSD. The data were screened for normality using Wilks-Shapiro/Rankits plots before application of the tests of significance. If significant departures from normality were detected, the data were transformed. A $\log_{10}$ transformation eliminated non-normality in the methylation, demethylation, and M/D data and in the sediment chemistry data. The transformations also eliminated any heterogeneity of variance as indicated by Bartlett's test of equal variance (Snedecor and Cochran 1980).

Results

Physical Measurements

All physical data collected on the five surveys are listed in Appendix Tables A1 to A8.

Mean water transparency in Methyl Bay was approximately half that measured in Granville and Sokatisewin lakes (Table 2). Transparency was highest at all sites in early to mid-July (Figure 2).

The nearshore stations in Methyl Bay were slightly (1 to 1.5 ^{o}C) cooler on average than the nearshore stations in either Granville or Sokatisewin lakes (Table 2). There was no difference in mean temperature between the nearshore

Table 2. Comparison of mean near-bottom physical and chemical conditions, and selected sediment characteristics; July 4 to August 24, 1988. All values are time-weighted means. Eh is oxidation-reduction potential.

Station No.	Secchi (m)	Sediment pH	Sediment Eh (mV)	Temperature (^{o}C)	Dissolved Oxygen ($mg\ L^{-1}$)	Specific Conductivity ($uS\ cm^{-1}$)
Methyl Bay						
1		6.34	293	16.7	7.3	102
2		6.43	292	17.2	7.5	90
3		6.56	309	17.2	8.6	81
4	1.08	6.62	348	15.6	7.8	85
Granville Lake						
5		6.81	260	18.3	8.8	62
6		6.74	250			
7		6.76	246			
8	1.89	6.80	246	18.4	7.8	65
Sokatisewin Lake						
9		6.65	160	18.9	7.2	81
10		6.65	160			
11		6.65	159			
12	2.01	6.71	47	12.6	1.3	103

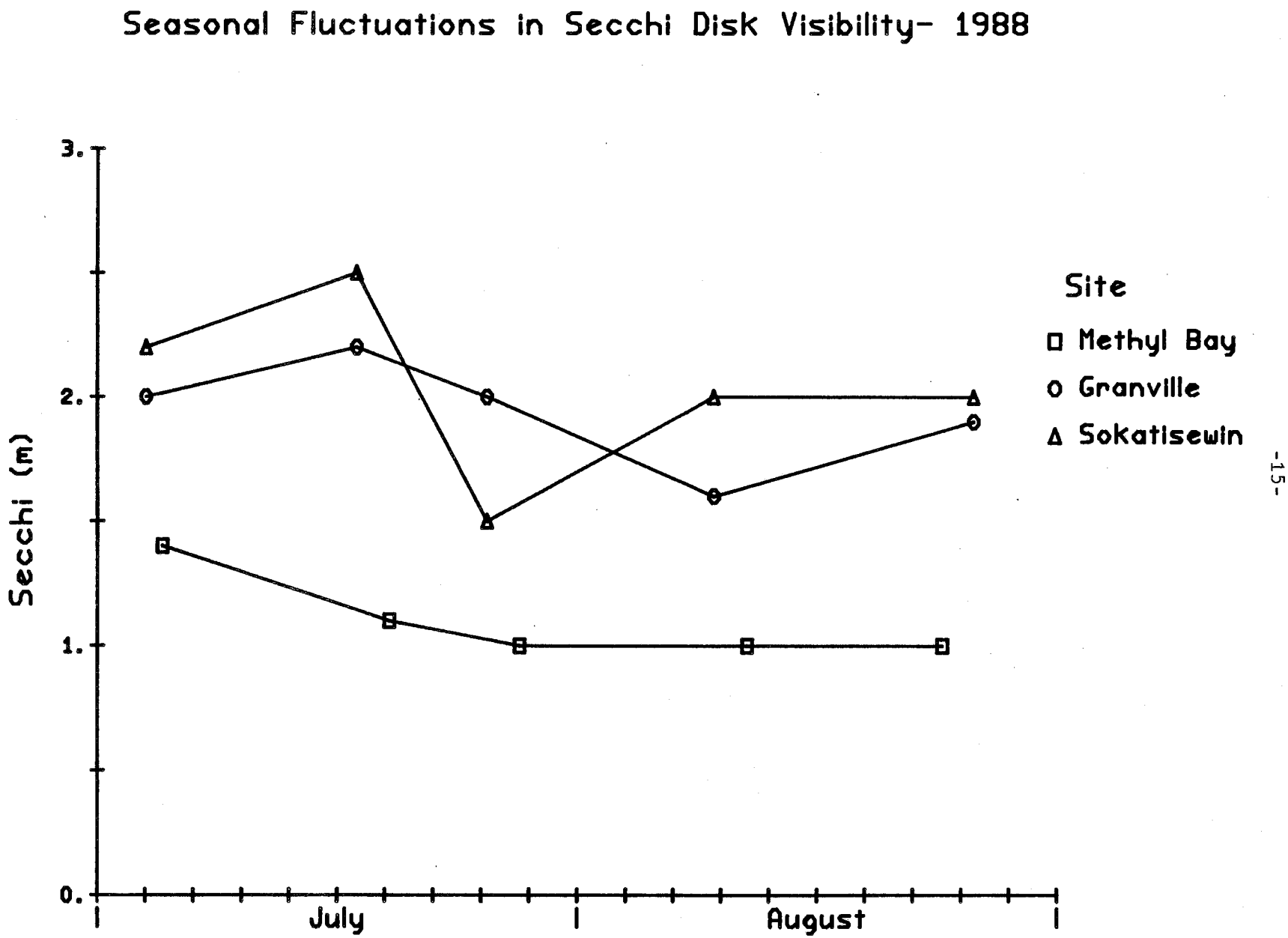


Figure 2. Seasonal fluctuations in Secchi disk visibility.

and offshore stations in Granville Lake. The offshore station in Methyl Bay was slightly (1 to 1.5 ^{o}C) cooler than the nearshore stations while the offshore station in Sokatisewin Lake was substantially (6.3 ^{o}C) cooler than the nearshore stations (Table 2). A thermocline was present at the offshore station at Sokatisewin Lake throughout the sampling period at depths ranging from 8.5m in early July to 10.5m in late August (Appendix Table A8). Near-bottom water temperature varied little over the period of study ($\pm$ 1 ^{o}C) at the nearshore stations at all sites as well as at the offshore station in Granville Lake (Figure 3). Considerable near-bottom warming occurred at the offshore stations in both Methyl Bay (4 ^{o}C) and Sokatisewin Lake (5.8 ^{o}C) over the course of the study. This was the product of a gradual deepening of the epilimnion or mixed layer at these stations over the summer (Appendix Tables A4 and A8).

The specific conductance of near-bottom waters was markedly higher at the reservoir sites (81-103 uS cm^{-1}) than in Granville Lake (62-65 uS cm^{-1}; Table 2). There was no difference in mean conductivity between the nearshore and offshore stations at either Granville Lake or Methyl Bay but in Sokatisewin Lake mean conductivity was about 20 uS cm^{-1} higher at the offshore station. Conductivity varied little over the summer at most locations but at the offshore station in Sokatisewin Lake conductivity increased progressively over the two months of study (Figure 4).

The near-bottom waters were well-oxygenated, averaging at least 7 mg L^{-1}, at the nearshore stations at all sites and at the offshore stations in Granville Lake and Methyl Bay (Table 2). The mean dissolved oxygen concentration was substantially lower at the offshore station in Sokatisewin Lake. In general, oxygen levels varied little over the course of the study (Figure 5). The only exception to this pattern was in the nearshore zone of

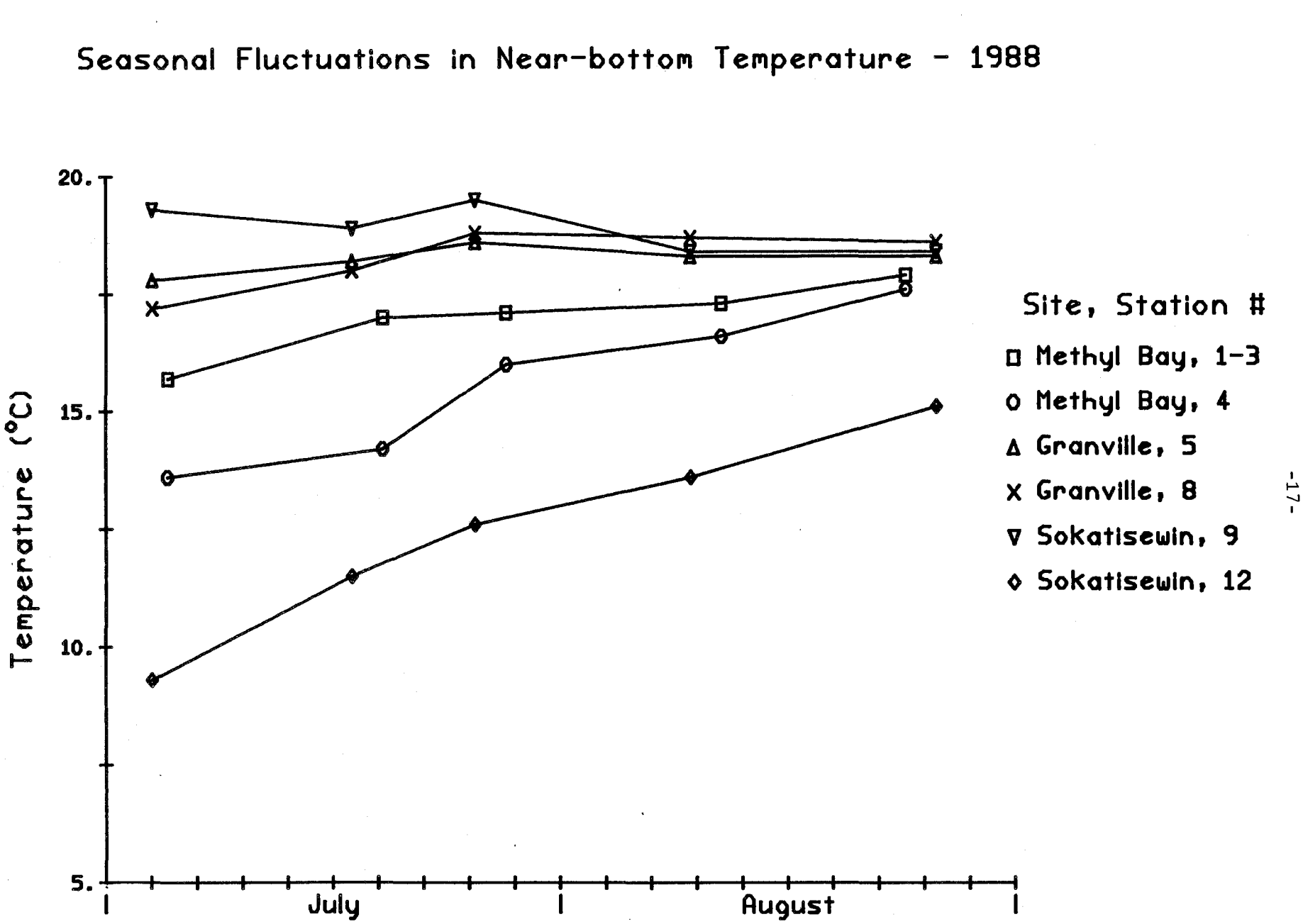


Figure 3. Seasonal fluctuations in near-bottom (within 25 cm) temperature.

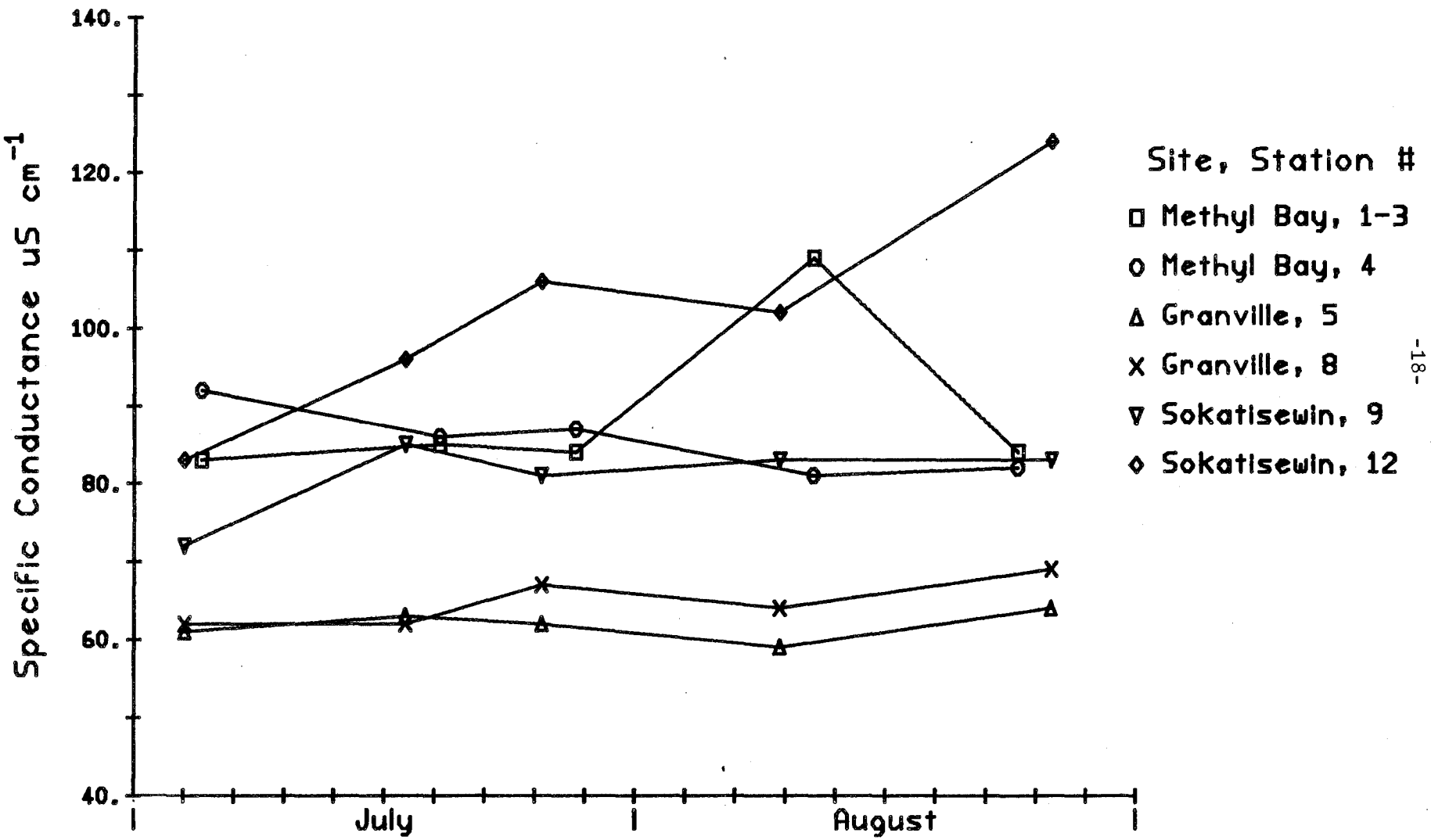


Figure 4. Seasonal fluctuations in near-bottom (within 25 cm) specific conductance.

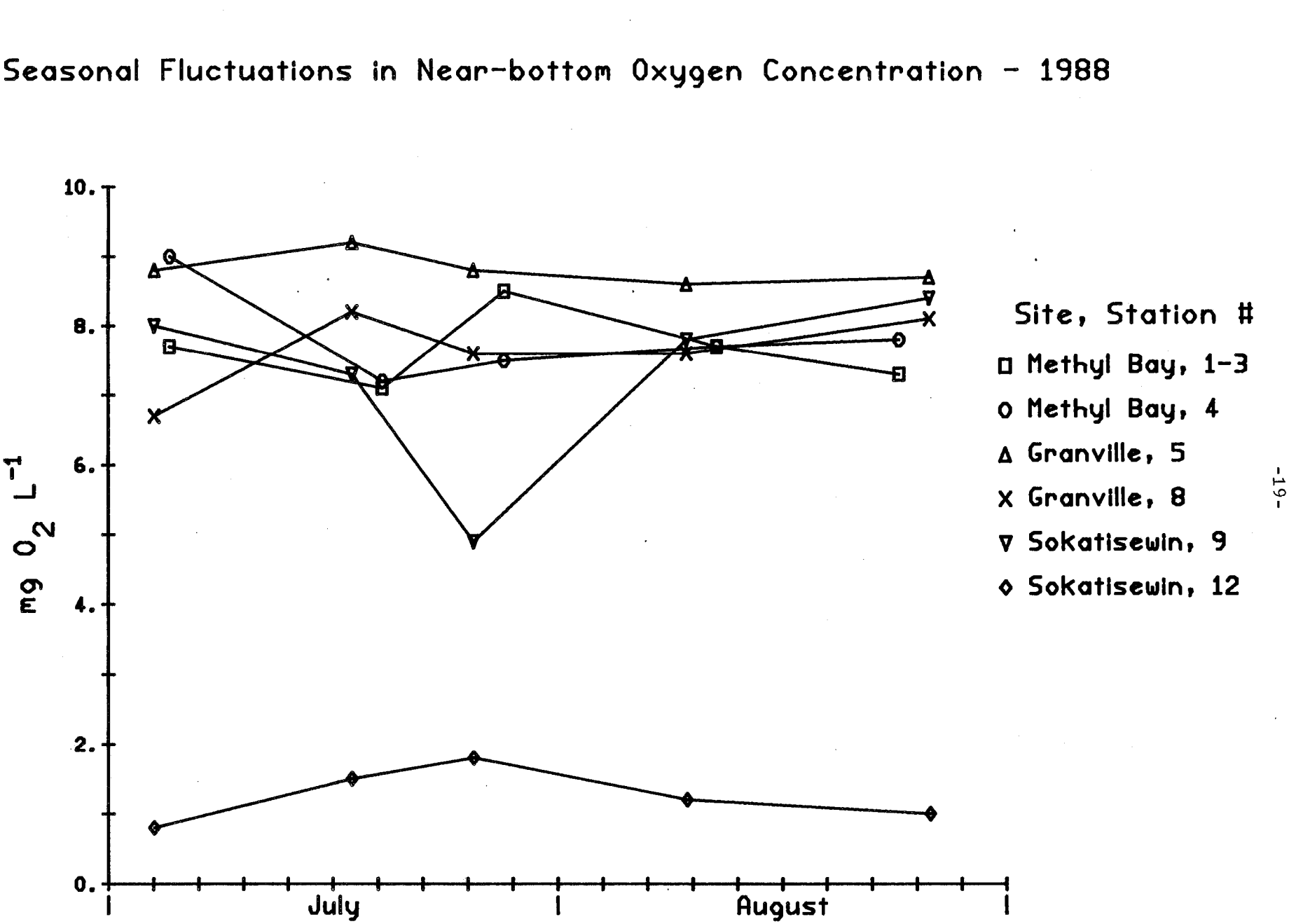


Figure 5. Seasonal fluctuations in near-bottom (within 25 cm) oxygen concentration.

Sokatisewin Lake, where a sharp but short-lived depression of oxygen levels occurred at the end of July.

Consistent with this continued presence of oxygen at all stations, although at low levels in some cases, oxidizing conditions (positive Eh) prevailed in the sediments at all three sites (Table 2). Large differences in near-bottom oxygen concentration were paralleled by comparable differences in sediment Eh (e.g., nearshore vs offshore in Sokatisewin Lake) but small differences in Eh were detected between sites where there was no corresponding difference in oxygen concentration. Mean nearshore oxygen levels were generally lower in Methyl Bay than in Granville Lake but mean sediment Eh was slightly but significantly higher in Methyl Bay (PLSD after $\log_{10}$ transformation, $p<0.001$; Table 2). Similarly, nearshore oxygen levels in Methyl Bay and Sokatisewin Lake were comparable yet sediment Eh in Sokatisewin was significantly lower, averaging just 54% of Eh in Methyl Bay (PLSD, $p<0.001$).

As observed for oxygen, sediment Eh was quite stable over the two months of study (Figure 6). The depression of nearshore oxygen levels noted in Sokatisewin Lake had no effect on sediment Eh but there was a marked midsummer depression of sediment Eh at the offshore station that was not accompanied by a decrease in near-bottom oxygen concentration. This was the only time that reducing conditions (negative Eh) were found in any sediment sample.

The overall range of mean sediment pH measured at the nearshore stations of the three sites was quite narrow (6.34 to 6.81) but there were significant differences in mean pH among all sites (PLSD, $p<0.001$, Table 2). The rank order of sediment pH at the offshore stations was the same as at the nearshore stations: lowest in Methyl Bay and highest in Granville Lake. Sediment pH fluctuated in a bimodal pattern in Methyl Bay with peaks in mid July and mid

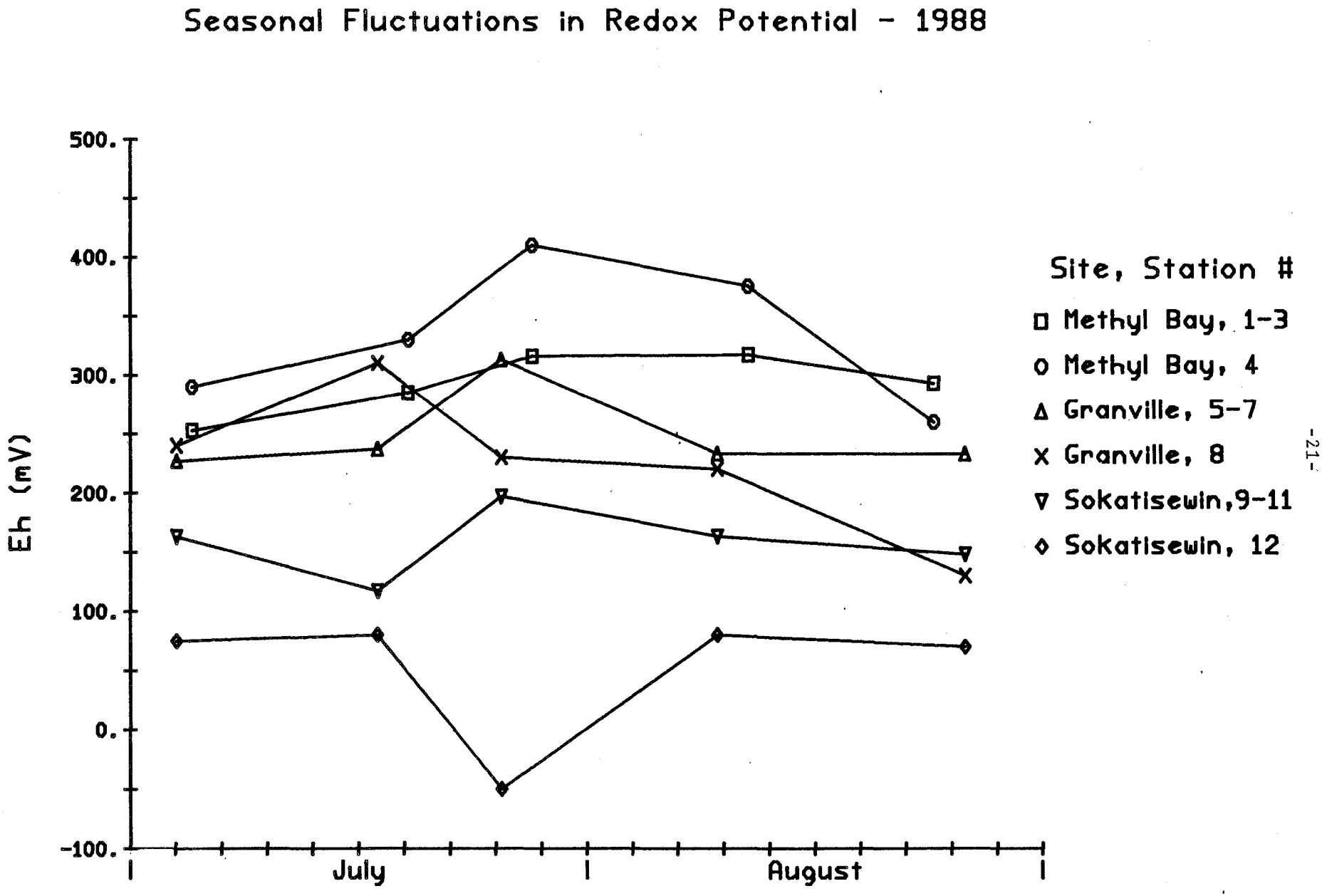


Figure 6. Seasonal fluctuations in sediment redox potential (Eh).

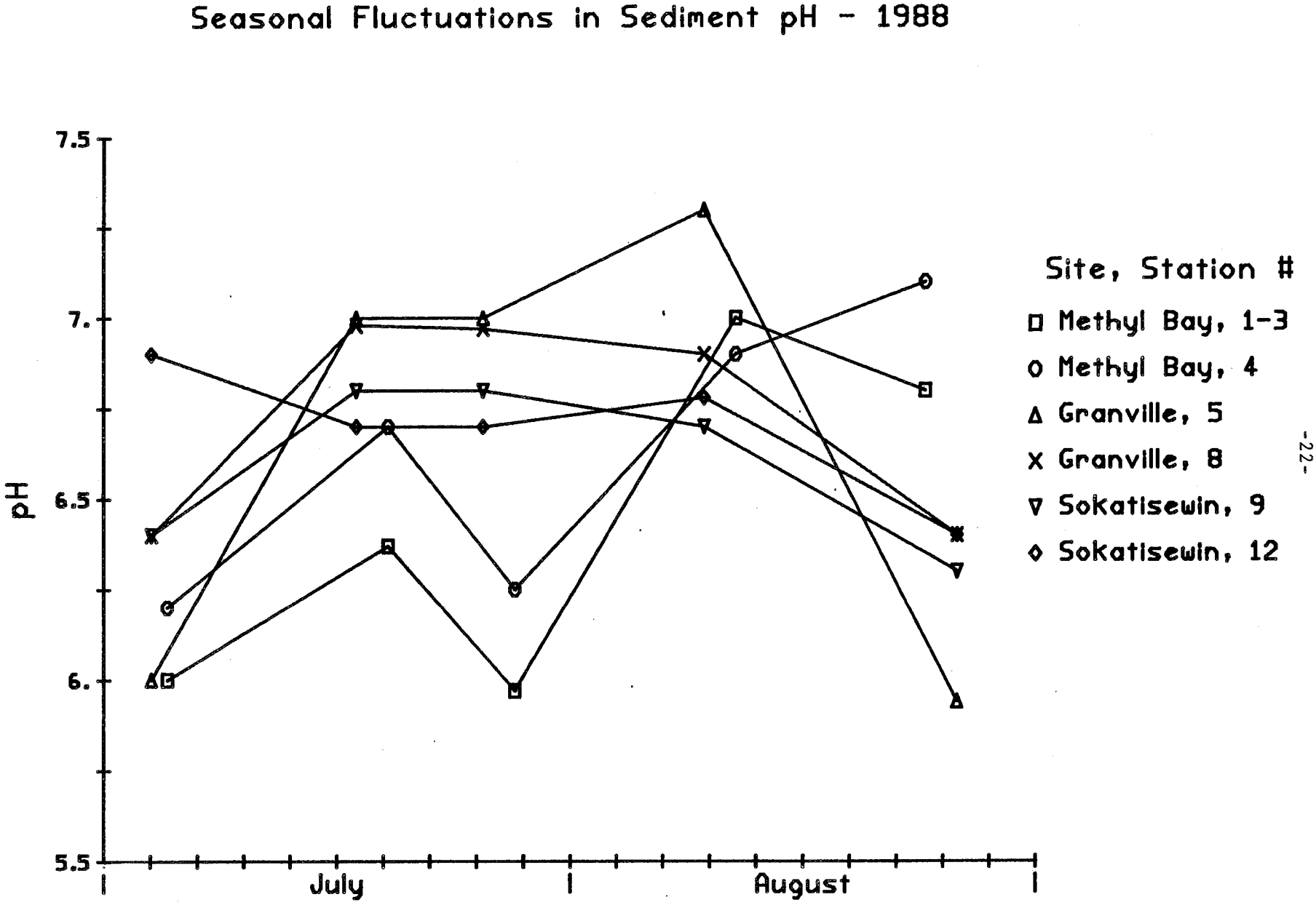


Figure 7. Seasonal fluctuations in sediment pH.

August and with higher pH in August than in July (Figure 7). In Granville lake, sediment pH fluctuated in a unimodal pattern; low in early July and late August and uniformly high from mid-July through mid-August. Less seasonal variation in sediment pH was evident in Sokatisewin Lake than at the other sites.

Sediment Chemistry

Carbon and Nitrogen

Results of all analyses of carbon and nitrogen in sediments are listed in Appendix Tables B1-B3.

The seasonal mean organic carbon concentrations at the nearshore stations in Methyl Bay were significantly higher than at the nearshore stations in either Granville or Sokatisewin lakes (PLSD after $\log_{10}$-transformation, $p<0.001$; Table 3). The nearshore organic carbon level in Sokatisewin Lake also was significantly higher than in Granville Lake ($p<0.001$). The mean sediment organic carbon concentration in nearshore flooded sediments from Methyl Bay was 6.8 times higher than in Granville lake and the level in Sokatisewin Lake was 3.9 times higher. Organic carbon concentrations at the flooded nearshore stations also were elevated in comparison to offshore stations at the same site. At Methyl Bay, the level offshore was lower than at any station in Granville Lake. The level at the offshore station in Sokatisewin Lake was about 40% of the concentration at the nearshore stations but was still about 50% higher than in Granville Lake. There was no difference in organic carbon levels between the nearshore and offshore stations in Granville Lake. The rank order of organic carbon concentration among the sites was maintained over the study period and there was minimal seasonal fluctuation at any of the sites (Figure 8).

Table 3. Comparison of the average chemical composition of surficial sediments; July 4 through August 24, 1988. All values are time-weighted means.

Station No.	Total Carbon	CO_3	Organic Carbon	Total Nitrogen	Total Mercury
	mg (g dry sediment)$^{-1}$				ng (g dry sed)$^{-1}$
Methyl Bay					
1	129	11	118	6.3	422
2	183	12	171	7.8	428
3	151	12	138	6.3	335
Mean	154	12	142	6.8	395
4	20	3	17	2.1	180
Granville Lake					
5	26	3	23	3.1	166
6	22	3	19	2.6	102
7	25	3	22	3.0	124
Mean	24	3	21	2.9	131
8	25	3	22	3.1	140
Sokatisewin Lake					
9	85	5	79	5.7	210
10	89	6	84	5.7	170
11	86	6	80	5.7	185
Mean	87	6	81	5.7	188
12	38	4	34	4.6	171

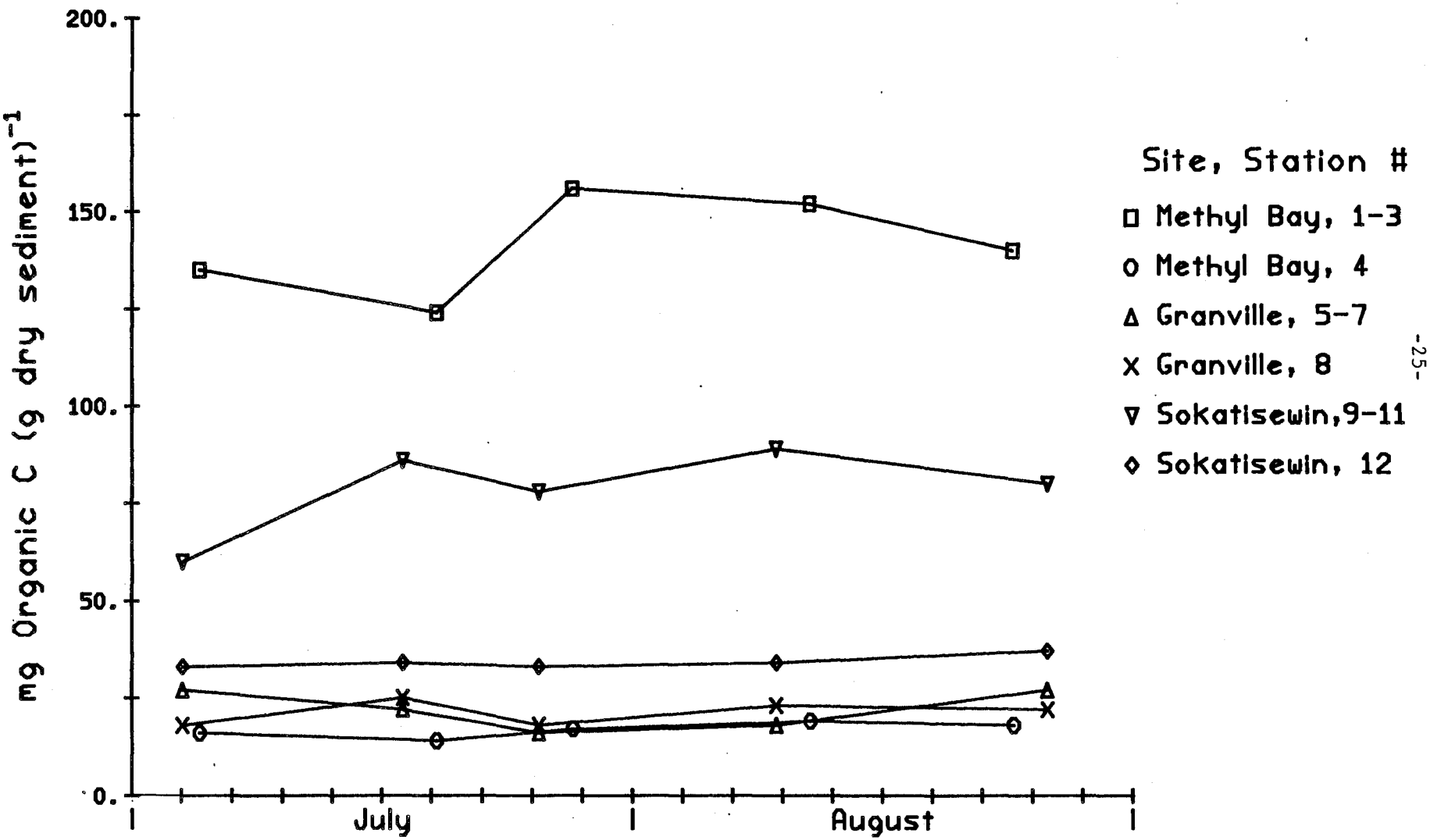


Figure 8. Seasonal fluctuations of sediment organic carbon concentrations.

Seasonal mean total nitrogen concentrations in sediments were strongly correlated with organic carbon concentrations (Table 4) so that the pattern of variation in total nitrogen among the sites was the same as that already described for organic carbon. Concentrations at the flooded nearshore stations were significantly higher than at the nearshore stations in Granville Lake (PLSD after $\log_{10}$-transformation, $p<0.001$; Table 3), the level in Methyl Bay was significantly higher than in Sokatisewin Lake ($p<0.001$), and in general concentrations at the offshore stations in the reservoirs were lower than at the nearshore stations. However, the differential in total nitrogen between the nearshore and offshore stations in Sokatisewin Lake (20% lower offshore) was smaller than seen for organic carbon. The magnitude of the elevation in sediment total nitrogen due to flooding also was smaller than for organic carbon. The mean nitrogen level in nearshore sediments from Methyl Bay was 2.3 times higher and the level in Sokatisewin Lake was a factor of 1.9 times higher than in Granville Lake. The rank order of total nitrogen concentration among the sites was maintained over the study period and there was little temporal variation in nitrogen levels at any of the sites (Figure 9).

The mean concentrations of organic carbon and total nitrogen were negatively correlated with sediment pH (Table 4).

Total Mercury

Results of all analyses of mercury in sediment are listed in Appendix Tables C1-C3.

Seasonal mean total mercury concentrations were significantly higher in sediments collected from flooded terrain than in sediments from Granville Lake (PLSD, $p<0.001$; Table 3). Mercury concentrations in flooded sediments from Methyl Bay were significantly higher than in flooded sediments from Sokatisewin

Table 4. Correlation matrix relating seasonal mean specific rates of microbial mercury methylation (M), demethylation (D), and the methylation balance (M/D) with the chemical composition of sediments and with microbial activity as measured by the rate of DIC production. The $\log_{10}$ transformations were used to satisfy the assumption of normality.

	log D		log M/D		log Organic C		log Total N		log Total Hg		pH		Eh		log DIC prod.	
	r	p	r	p	r	p	r	p	r	p	r	p	r	p	r	p
log M	0.043	p>0.1	0.610	p=0.04	0.783	p<0.01	0.844	p<0.01	0.719	p<0.01	-0.622	p=0.02	-0.355	p>0.1	-0.199	p>0.1
log D			-0.765	p<0.01	0.358	p>0.1	0.162	p>0.1	0.677	p=0.02	-0.701	p=0.01	0.837	p<0.01	0.839	p<0.01
log M/D					0.222	p>0.1	0.417	p>0.1	-0.073	p>0.1	0.153	p>0.1	-0.892	p<0.01	-0.791	p<0.01
log Organic C							0.967	p<0.01	0.840	p<0.01	-0.771	p<0.01	-0.021	p>0.1	0.227	p>0.1
log Total N									0.760	p<0.01	-0.663	p=0.02	-0.243	p>0.1	0.034	p>0.1
log total Hg											-0.899	p<0.01	0.311	p>0.1	0.451	p>0.1
pH													-0.332	p>0.1	-0.471	p>0.1
Eh															0.918	p<0.01

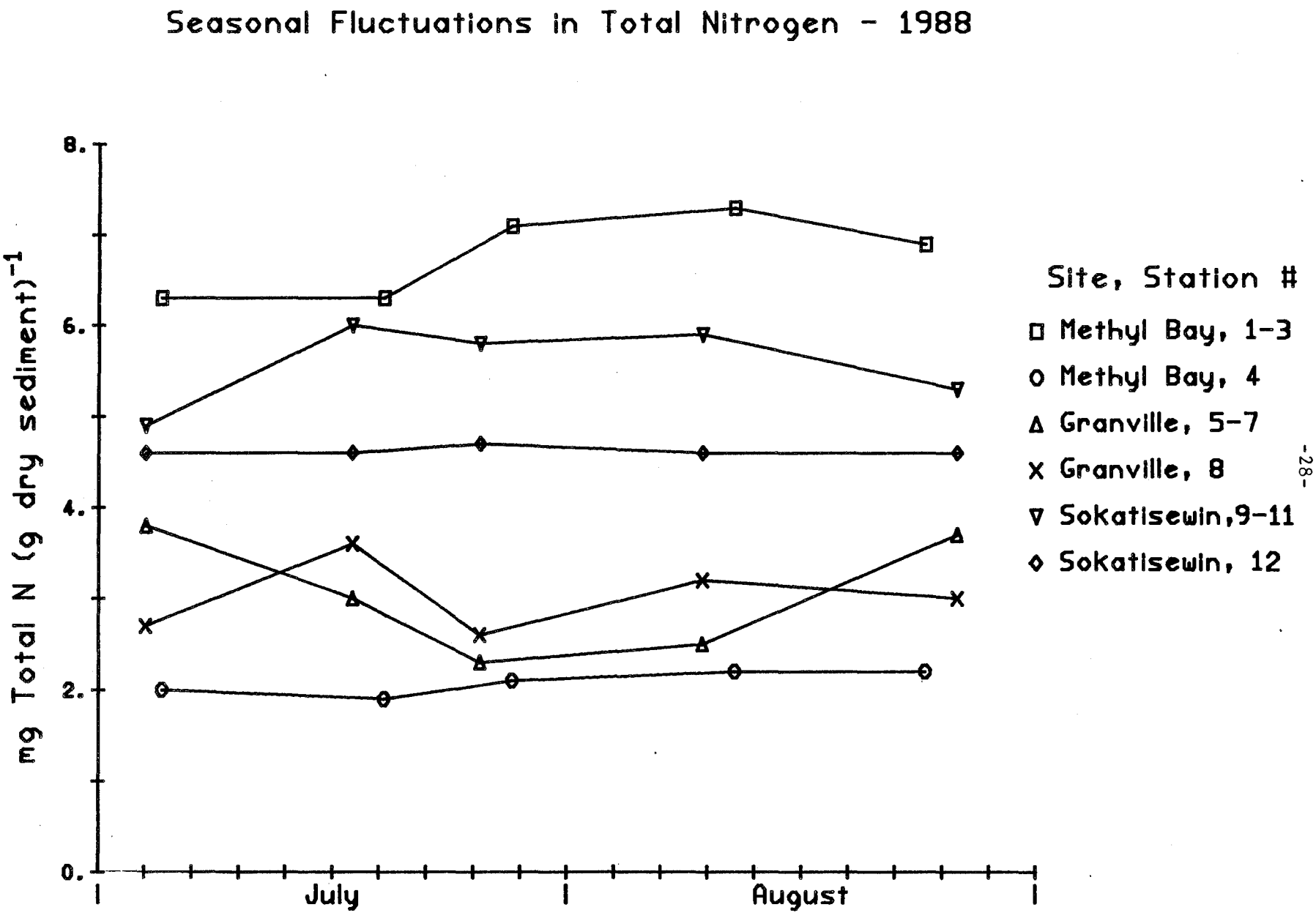


Figure 9. Seasonal fluctuations of sediment total nitrogen concentrations.

Lake (PLSD, $p<0.001$). The mean total mercury concentration at the offshore station in Granville Lake was the same as at the nearshore stations. Mean total mercury at the offshore station in Methyl Bay was about 45% of the level at the flooded stations, was comparable to the concentrations measured in Sokatisewin Lake sediments, and was about 30% higher than at the offshore station in Granville Lake.

There was little variation in sediment mercury concentrations at the reservoir sites over the course of the study; concentrations measured on a given sampling date were generally within 10-20% of the seasonal mean for that location (Figure 10). In contrast, there was a distinct seasonal decline in total mercury concentrations in Granville Lake sediments. Mercury concentrations were highest, and within the range measured in the reservoirs, at the start of sampling both at the nearshore and at the offshore locations but the levels declined by 150-225 ng g^{-1} over the following 3 weeks (Figure 10).

The mean concentration of total mercury in sediment was negatively correlated with mean sediment pH and was positively correlated with the mean concentrations of organic carbon and total nitrogen (Table 4).

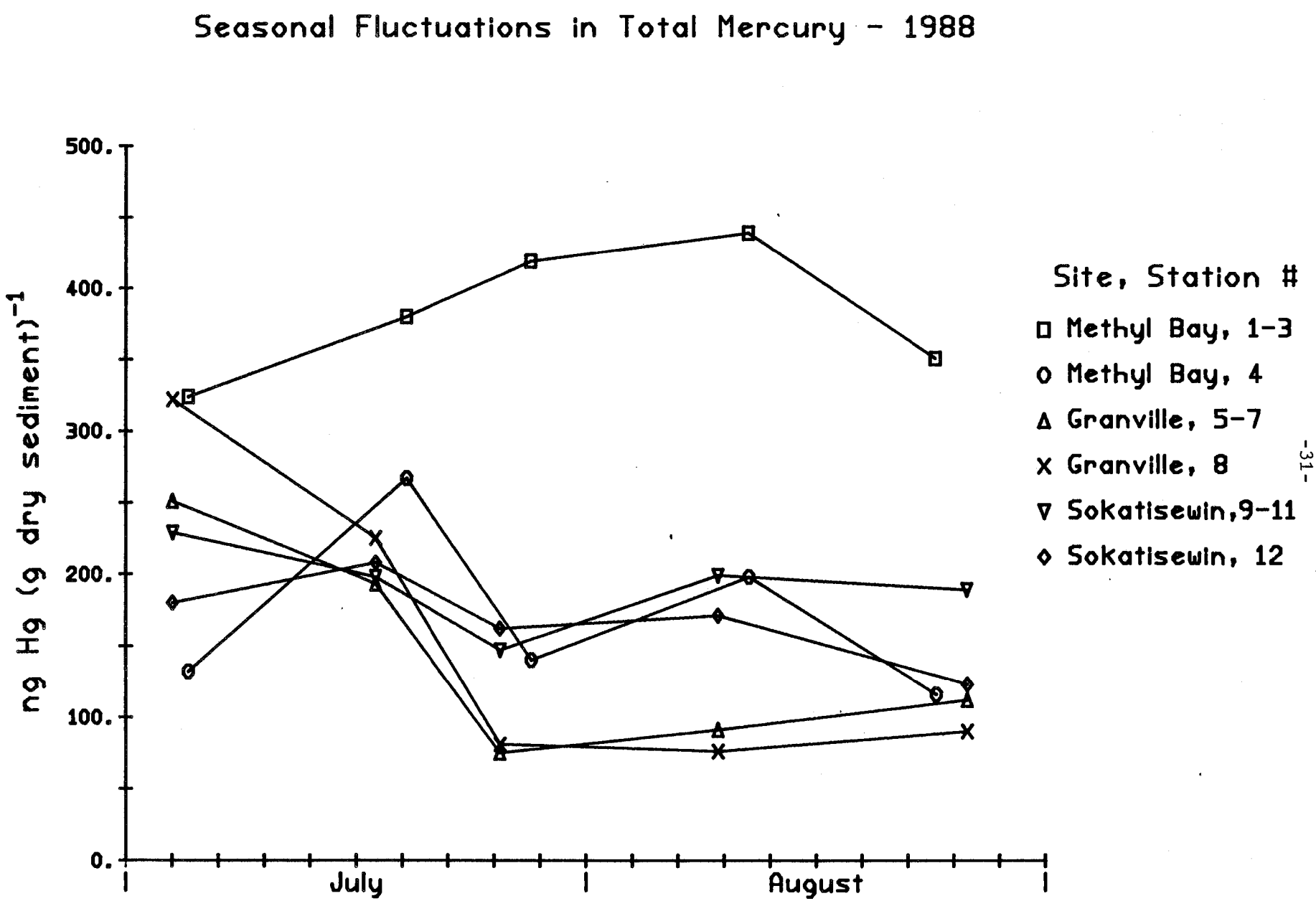
Microbial Activity

Results of all assays of microbial activity are presented in Appendix Tables D1-D3.

Seasonal mean microbial activity, as measured by the rate of DIC production, was significantly higher (PLSD after $\log_{10}$-transformation, $p<0.001$) in sediments from the flooded zone of Methyl Bay than in the nearshore zone of either Granville or Sokatisewin lakes (Table 5). Microbial activity in the nearshore zone of Sokatisewin Lake was significantly lower than in the

Table 5. Comparison of seasonal mean (time-weighted) microbial activity; July 4 through August 24, 1988.

uM DIC (g dry sediment)$^{-1}$ day^{-1}					
Methyl Bay		Granville Lake		Sokatisewin Lake	
Station	Activity	Station	Activity	Station	Activity
1	32.10	5	9.07	9	2.65
2	26.84	6	9.35	10	3.24
3	24.16	7	11.89	11	2.43
Nearshore Mean	27.70		10.10		2.77
4	9.68	8	7.19	12	0.36



Fugure 10. Seasonal fluctuations of sediment total mercury concentrations.

nearshore zone of Granville Lake (PLSD after $\log_{10}$-transformation, $p<0.001$). Microbial activity in the offshore zone of Methyl Bay was within the range measured at the nearshore stations in Granville Lake. Microbial activity was slightly lower, on average, at the offshore station in Granville Lake than at the nearshore stations. At the offshore station in Sokatisewin Lake, microbial activity averaged just 13% of the already low activity at the nearshore stations. Seasonal mean microbial activity was positively correlated with mean sediment Eh (Table 4).

Microbial activity at the flooded stations in Methyl Bay consistently exceeded activity at the other locations after early July (Figure 11). Activity increased quickly to peak in mid-July, gradually decreased to about 50% of this peak level by mid-August, and remained at this level through the end of sampling. Microbial activity at the offshore station in Methyl Bay followed a similar pattern through mid-August but the decline continued through to the end of sampling. At the nearshore stations in Granville Lake, microbial activity remained at about the same level over the two-month course of sampling. Activity at the offshore station was about the same as that at the nearshore stations through July but was near zero on both sampling dates in August. Microbial activity was consistently low but measurable at the nearshore stations in Sokatisewin Lake over the study. Activity at the offshore station was measurable on just two occasions, mid-July and late August.

Methylation Balance

Results of all assays of specific methylation and demethylation rates are listed in Appendix Tables D1-D3.

Seasonal mean specific methylation rates at the flooded sites were

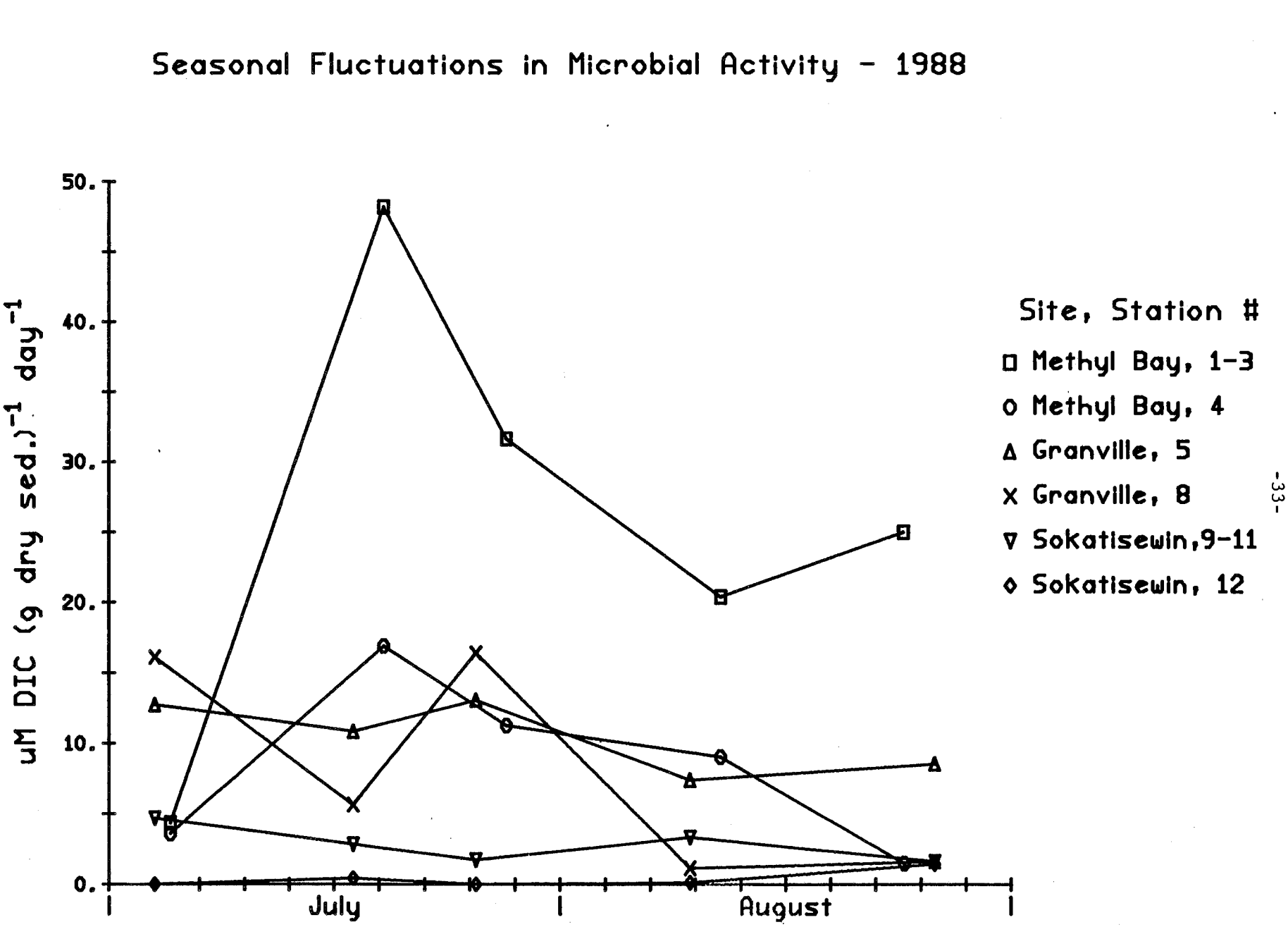


Figure 11. Seasonal fluctuations of microbial activity as measured by the rate of dissolved inorganic carbon (DIC) production.

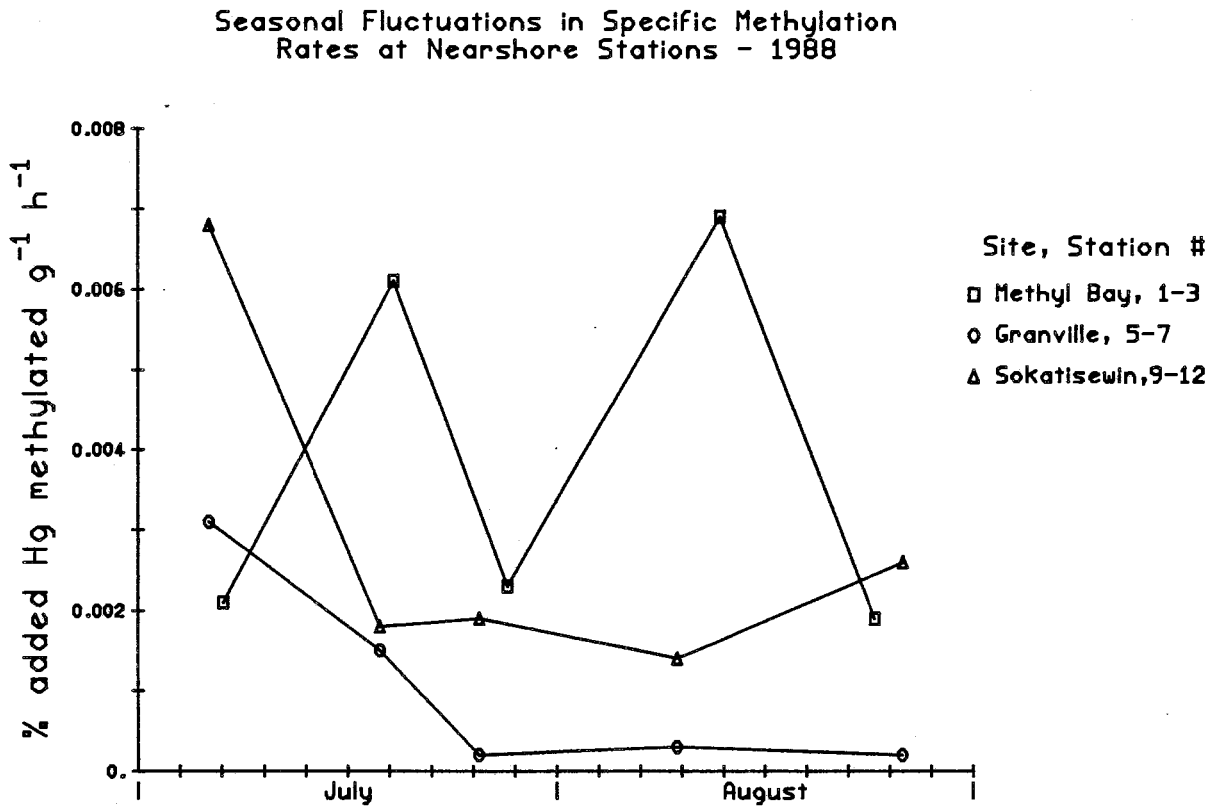
significantly higher (PLSD after $\log_{10}$-transformation, $p<0.001$) than at the nearshore stations in Granville Lake (Table 6). Mean methylation rates in the nearshore zone of Methyl Bay also were significantly higher than in the nearshore zone of Sokatisewin Lake ($p<0.001$). The high methylation activity in Methyl Bay was restricted to the flooded zone; specific methylation at the offshore station was the same as in Granville Lake. However, high methylation activity also was found at the offshore station in Sokatisewin Lake, where the seasonal mean methylation rate was 50% higher than at the nearshore stations in Methyl Bay (Table 6). Mean specific methylation rates were positively correlated with the seasonal mean concentrations of organic carbon, total nitrogen, and total mercury and were negatively correlated with mean sediment pH (Table 4).

Specific methylation rates at the flooded nearshore stations were consistently higher than in Granville Lake after the first week of July (Figure 12a). In Methyl Bay, methylation activity fluctuated in a bimodal pattern with peaks in mid July and again in mid August. The first pulse of methylation activity in Methyl Bay coincided with the initial increase in microbial activity but subsequent variations in methylation rates were not related to fluctuations in total microbial activity (Figures 11 and 12a).

Methylation activity at the nearshore stations in Sokatisewin Lake peaked at or before the start of sampling and declined sharply (about 70%) in mid-July, remaining at this lower level to the end of August (Figure 12a). Similarly, methylation rates in Granville lake peaked at or before the start of sampling, declined to near zero by the end of July, and remained at this low level through August. No comparable declines in microbial activity coincided with these decreases in methylation rates (Figures 11 and 12a).

Table 6. Seasonal mean specific methylation and demethylation rates and corresponding M/D ratios; July 4 through August 24, 1988. All values are time-weighted means.

Station No.	% added Hg g^{-1} h^{-1} Methylation	Demethylation	M/D
Methyl Bay			
1	0.0034	0.2620	0.0130
2	0.0054	0.3313	0.0163
3	0.0042	0.1004	0.0418
Nearshore mean	0.0043	0.2312	0.0238
4	0.0009	0.2160	0.0042
Granville Lake			
5	0.0010	0.0809	0.0124
6	0.0009	0.0412	0.0218
7	0.0007	0.0523	0.0134
Nearshore Mean	0.0009	0.0581	0.0159
8	0.0009	0.0378	0.0238
Sokatisewin Lake			
9	0.0026	0.0555	0.0468
10	0.0023	0.0416	0.0558
11	0.0021	0.0334	0.0629
Nearshore Mean	0.0023	0.0435	0.0550
12	0.0067	0.0114	0.5877



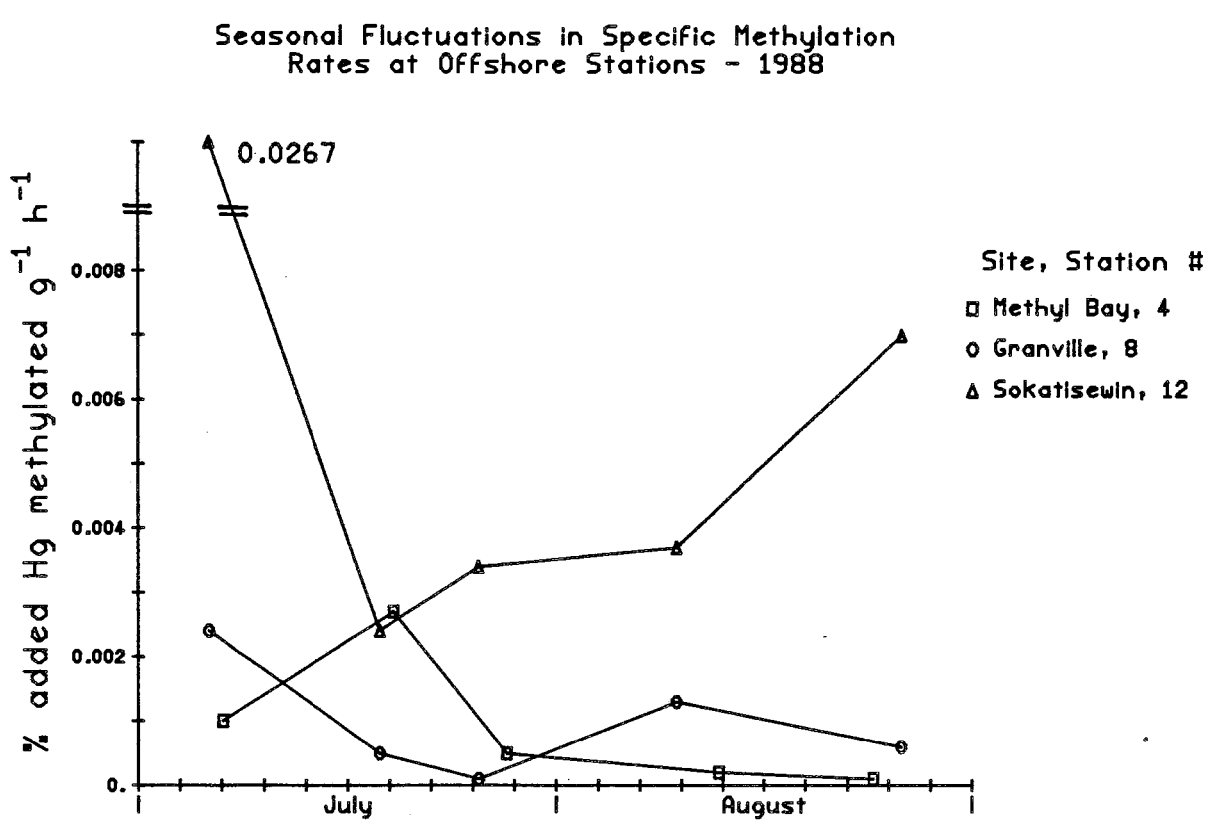
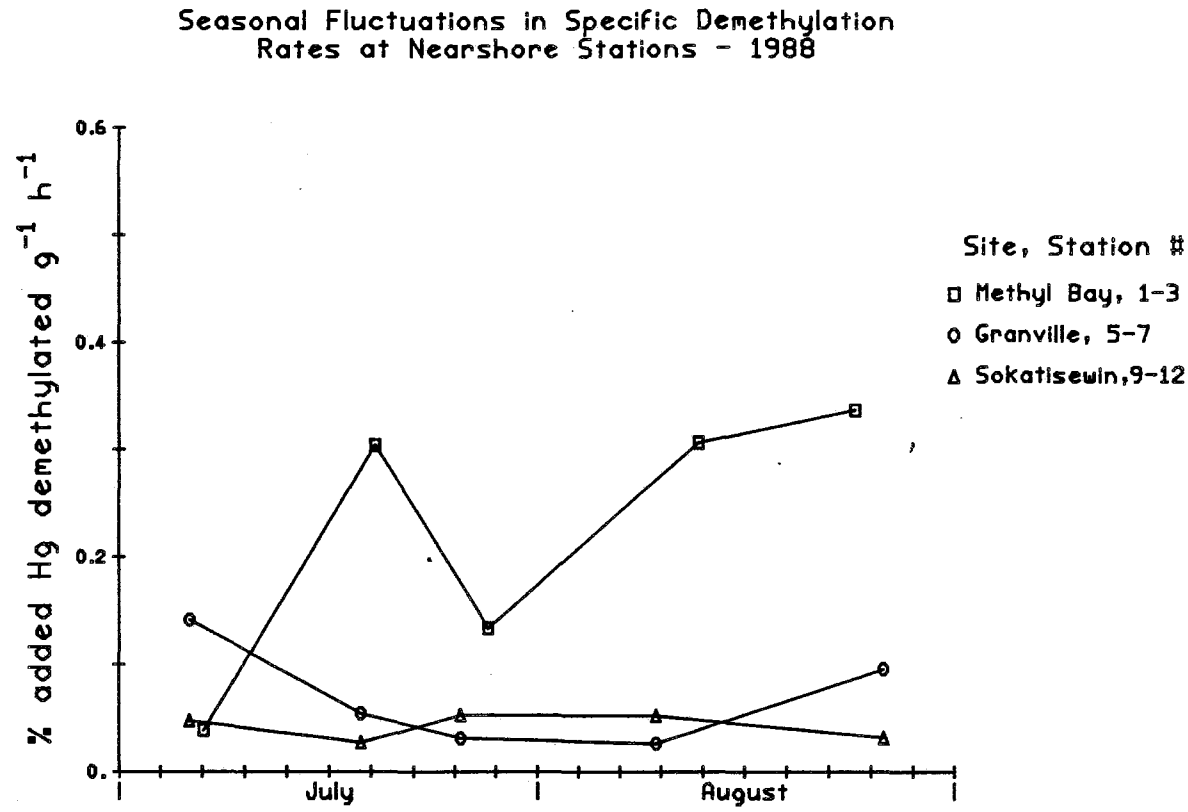


Figure 12. a, upper, and b, lower.

The high methylation activity at the offshore station in Sokatisewin lake was most evident on the first survey (Figure 12b). On subsequent surveys, methylation rates were comparable to those measured at the nearshore stations in the lake and were lower than at the nearshore stations in Methyl Bay (Figure 12 a and b).

Seasonal mean specific demethylation rates were significantly higher in flooded sediments from Methyl Bay than in sediments from the nearshore zones of Granville and Sokatisewin lakes (PLSD after $\log_{10}$-transformation, $p<0.001$; Table 6). Demethylation activity also was comparably high at the offshore station in Methyl Bay. In contrast, mean nearshore demethylation in Sokatisewin Lake was significantly lower ($p=0.03$) than in Granville Lake (Table 6). Mean demethylation rates at the offshore stations in Sokatisewin and Granville lakes were lower than at the nearshore stations. Mean specific demethylation rates were positively correlated with seasonal mean Eh, microbial activity, and total mercury and were negatively correlated with mean sediment pH (Table 4).

Demethylation rates in Methyl Bay markedly exceeded those at the other two sites after the first week of July and fluctuated in a bimodal pattern similar to that of methylation, with peaks in mid-July and again in late summer (Figures 12a and 13a). However, unlike methylation activity, the high demethylation rates achieved in the second pulse were maintained through the termination of sampling. The high mean demethylation activity at the offshore station in Methyl Bay was largely the result of a very high rate measured in mid-August, at the time the second demethylation pulse developed at the nearshore stations (Figures 13 a and b). Demethylation rates in Granville Lake were highest in early July, declined through the following three weeks and remained low to about mid-August. The higher mean nearshore demethylation in Granville Lake than in Sokatisewin Lake was the result of higher rates in early



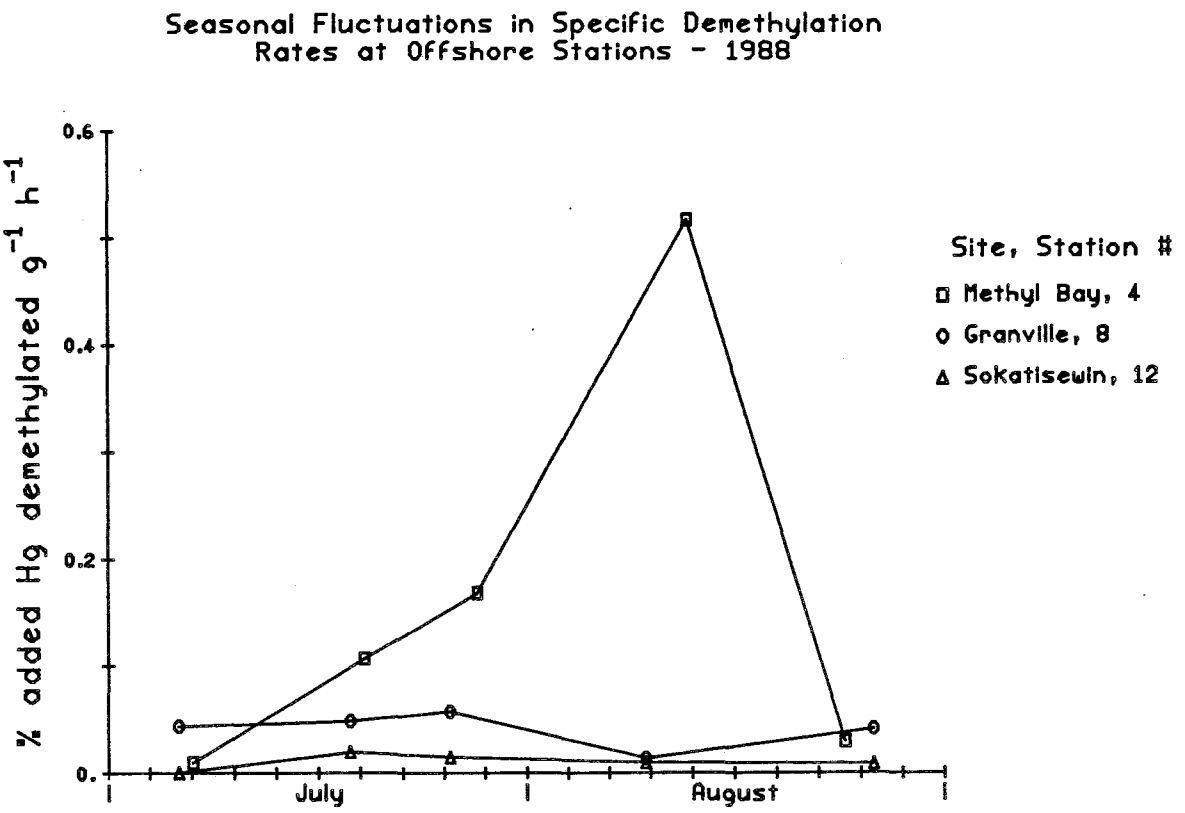
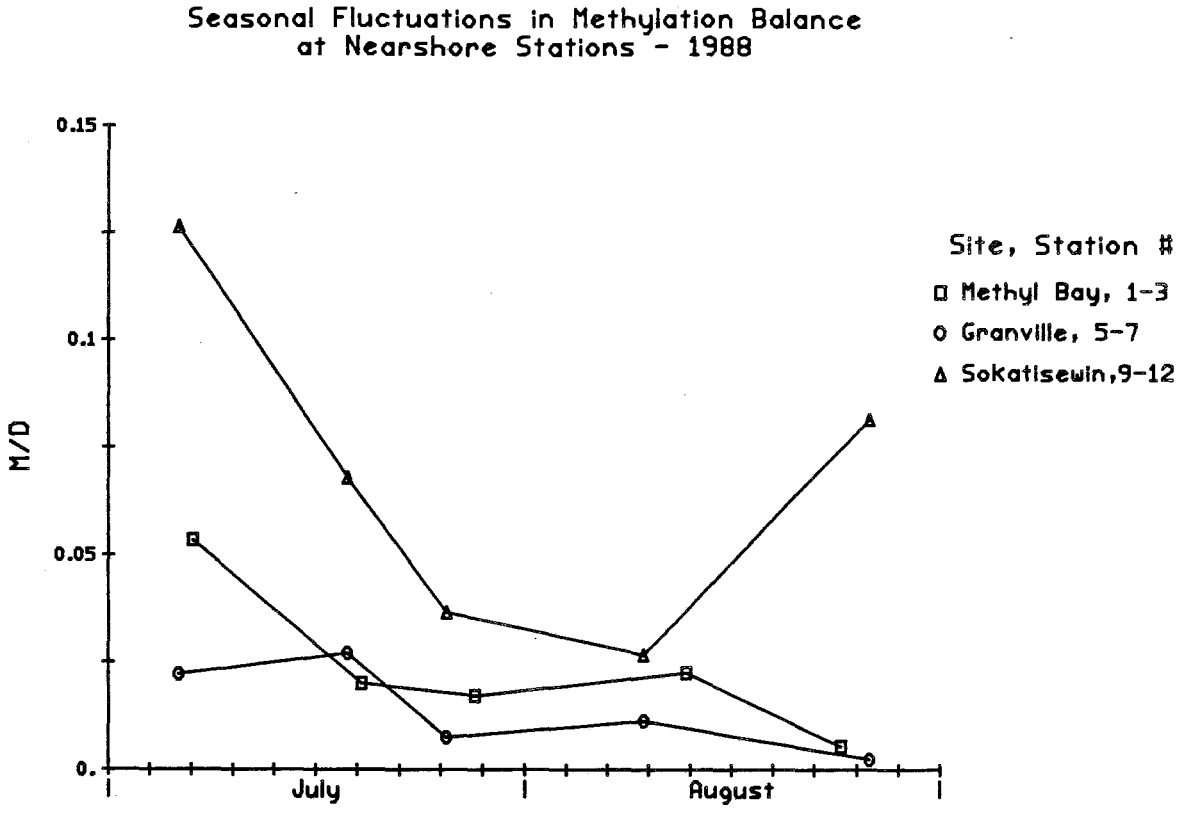


Figure 13. a, upper, and b, lower

July and again in late August (Figure 13a).

The average balance of microbial methyl mercury production, indicated by the seasonal mean M/D, was significantly higher (PLSD after $\log_{10}$-transformation, $p<0.01$) in flooded sediments from both Methyl Bay and Sokatisewin Lake than at the nearshore stations in Granville Lake (Table 6). Nearshore mean M/D in Sokatisewin Lake was significantly higher than in Methyl Bay ($p<0.001$). The combination of high methylation activity and low demethylation activity at the offshore station in Sokatisewin Lake resulted in that station supporting the highest mean M/D; about 10 times that in the nearshore zone of the lake (Table 6). The mean methylation balance at the offshore station in Methyl Bay was lower than at any other station; solely due to the high demethylation activity there (Table 6). Seasonal mean M/D was positively correlated with the mean specific methylation rate and was negatively correlated with the mean specific demethylation rate, mean sediment Eh, and mean microbial activity (Table 4).

The microbial methylation balance in the nearshore zone of Sokatisewin Lake was consistently higher than at the other sites and the seasonal fluctuations paralleled those of methylation; high in early July and late August and lowest in midsummer (Figures 12a and 14a). The methylation balance at the nearshore stations in Methyl Bay was higher than in Granville Lake on four of the five sampling dates and was highest in early July. Peak M/D in Methyl Bay coincided with the date of lowest demethylation activity rather than a period of high methylation activity (Figures 12a, 13a, and 14a). The subsequent peaks of methylation activity were largely compensated by peaks in demethylation activity. The methylation balance was consistently low in the nearshore zone of Granville Lake (Figure 14a).



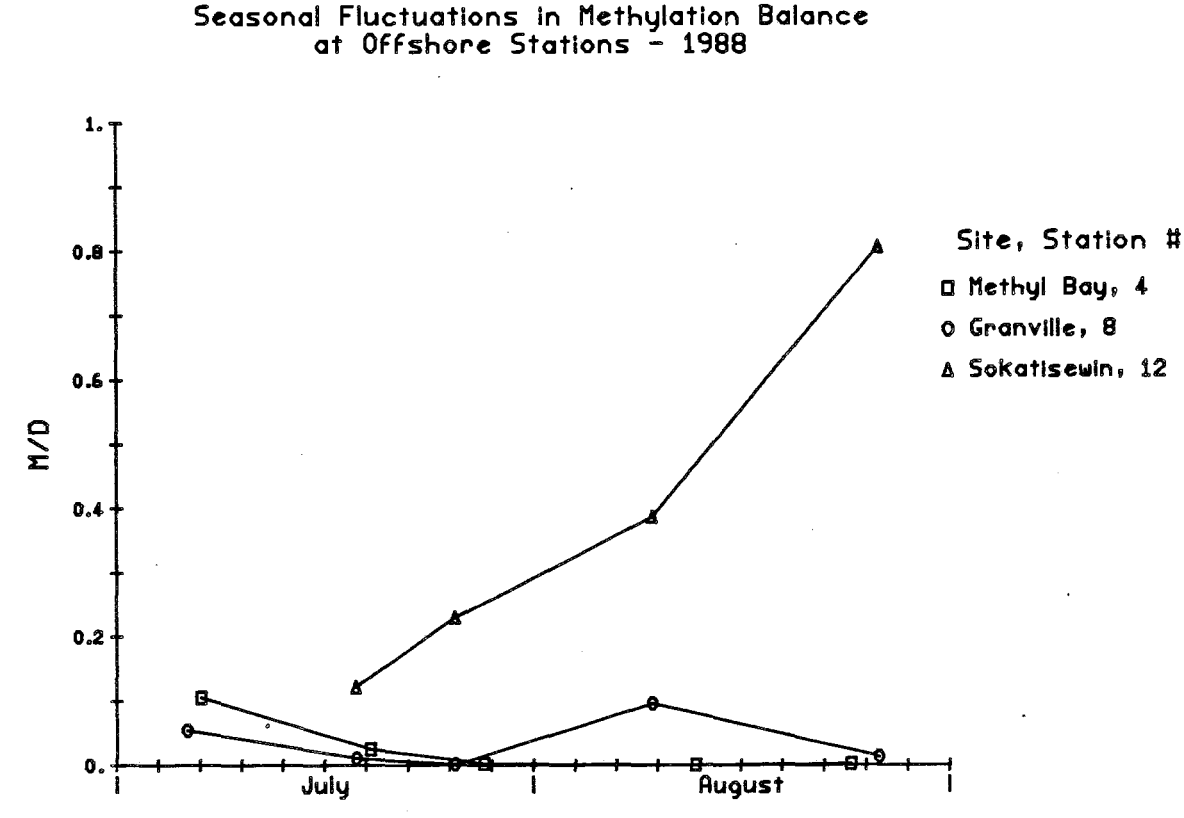


Figure 14. a, upper, and b, lower.

Inhibition Experiment

Granville Lake

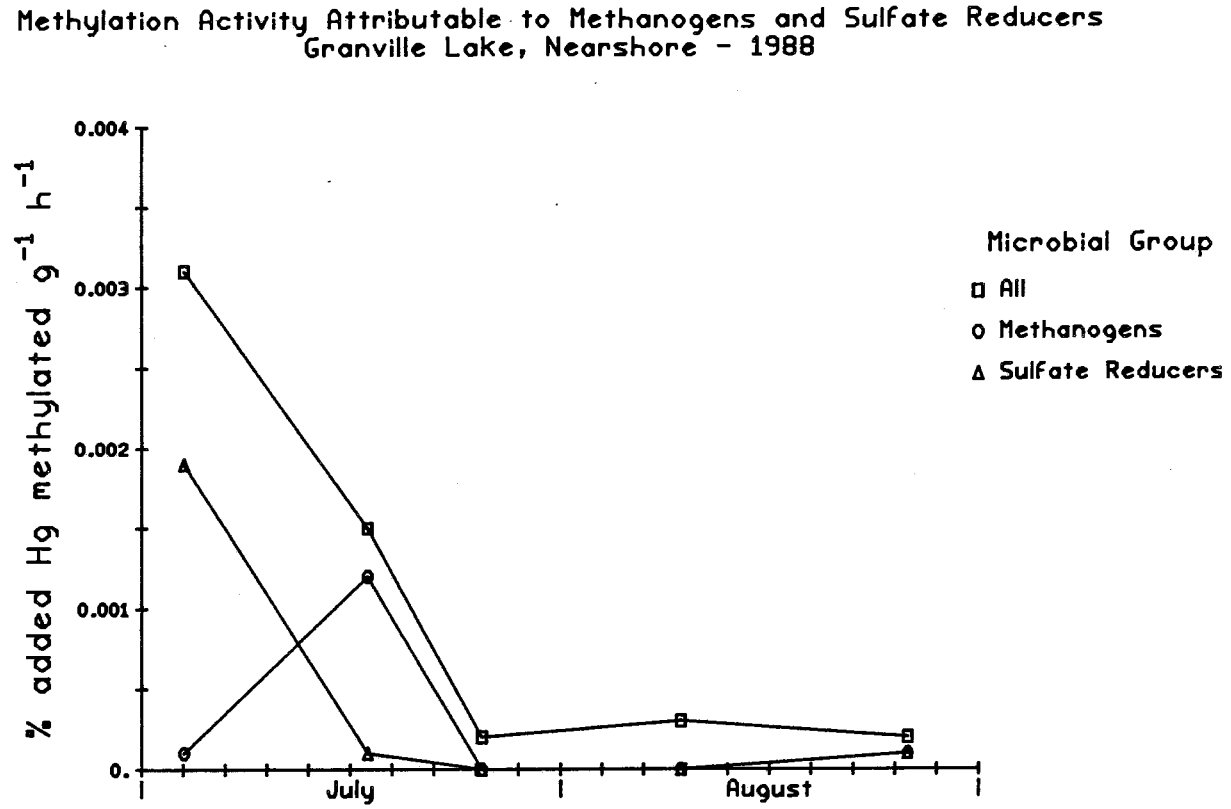
The methanogens and the sulfate reducers each accounted for about 33% of seasonal mean methylation activity and for about 25% of mean demethylation activity at the nearshore stations (Table 7). There was considerable variation in the contribution of these two groups to methylation at the individual stations. No methylation was attributable to the methanogens at station 5 yet this group accounted for 89% of total methylation at station 6 (Table 7). The share of demethylation represented by the two groups was less variable (methanogens 16 to 40%, sulfate reducers 8 to 34%) The methanogens and sulfate reducers jointly accounted for all of total methylation at the offshore station (Table 7). Methanogens were responsible for about 12% of mean demethylation at the offshore station while the sulfate reducers accounted for about 33% of demethylation (Table 7).

The methanogens and sulfate reducers were particularly important to total methylation activity during the periods of peak activity. Sulfate reducers accounted for about 65% of total nearshore methylation at the peak in early June, with methanogens responsible for 80% of total methylation in mid-July (Figure 15a). The contributions of the two groups did not overlap in July although the small pulse of methylation at the offshore station in August was the combined result of increased methylation by both groups (Figure 15 a and b).

In contrast, the methanogens and sulfate reducers generally accounted for the largest share of demethylation during the periods of lowest demethylation activity at the nearshore stations. Large proportions (35-80%) of total demethylation were contributed by some other unidentified microbial group(s) on the first and last sampling dates when demethylation rates were highest (Figure

Table 7. Comparison of seasonal mean specific rates of microbial methylation and demethylation attributable to the methanogenic and sulfate reducing bacteria; July 4 through August 24, 1988. All values are time-weighted means.

	% added Hg g^{-1} h^{-1}					
	Methylation			Demethylation		
Fraction	Total	Methanogens	Sulfate Reducers	Total	Methanogens	Sulfate Reducers
Methyl Bay						
1	0.0034	0.0015	0.0029	0.2620	0.0939	0.1404
2	0.0054	0.0026	0.0033	0.3313	0.1862	0.2034
3	0.0042	0.0003	0.0023	0.1004	0.0127	0.0409
Mean	0.0043	0.0015	0.0028	0.2279	0.0976	0.1282
4	0.0009	0.0008	0.0005	0.2160	0.1745	0.1599
Granville Lake						
5	0.0010	0	0.0006	0.0861	0.0137	0.0175
6	0.0009	0.0008	0.0001	0.0438	0.0140	0.0034
7	0.0007	0.0001	0.0002	0.0523	0.0221	0.0190
Mean	0.0009	0.0003	0.0003	0.0581	0.0166	0.0133
8	0.0009	0.0008	0.0005	0.0378	0.0047	0.0123



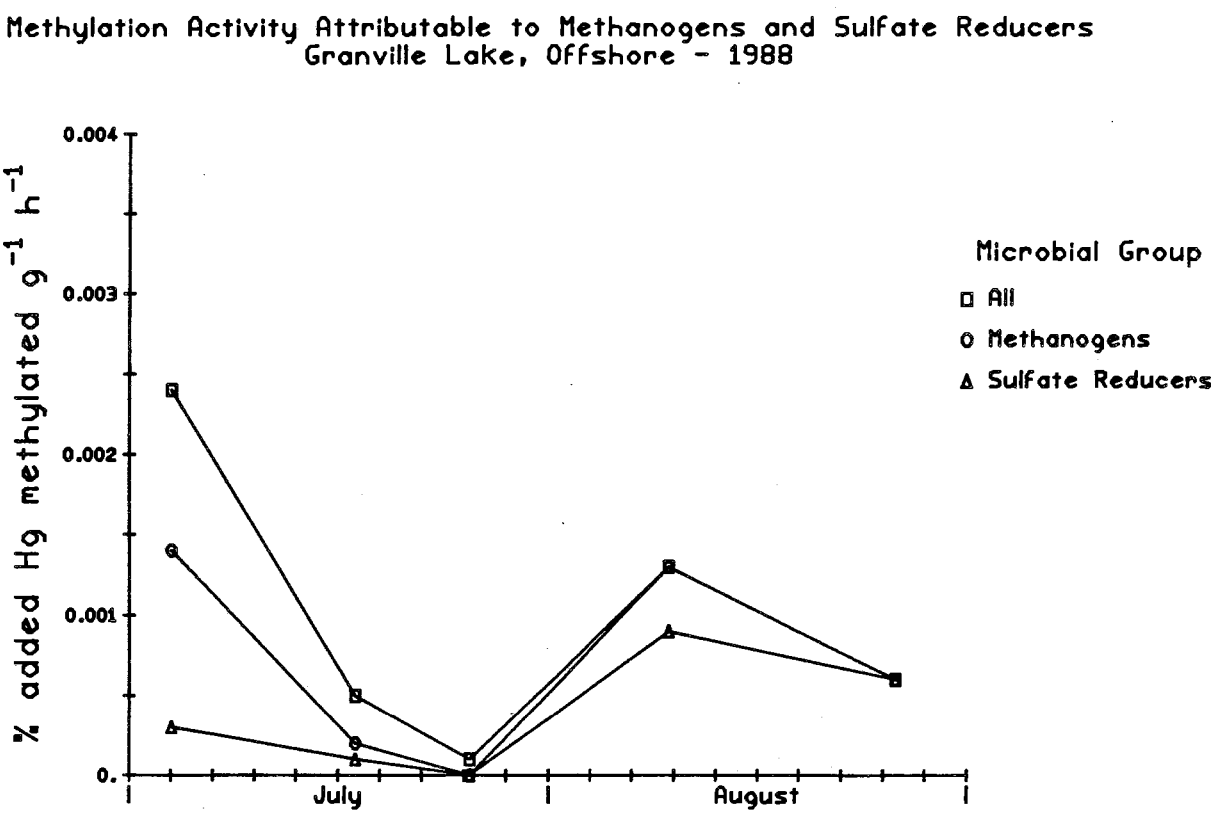


Figure 15. a, upper, and b, lower.

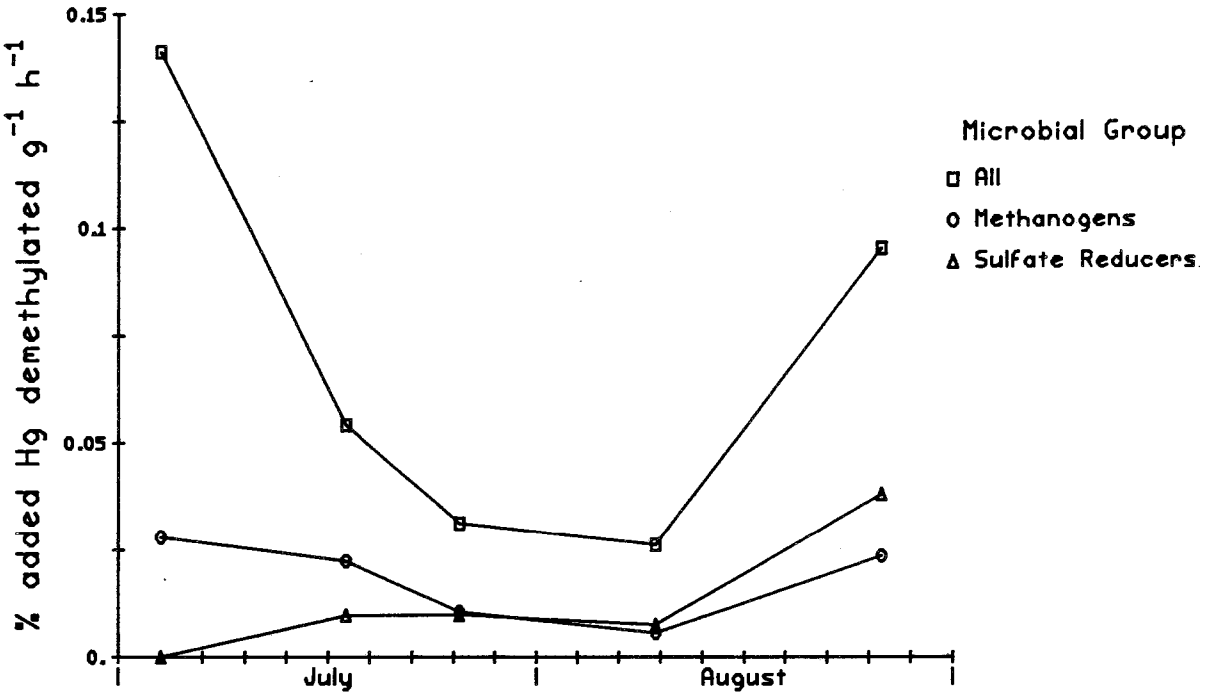
16a). At the offshore station, sulfate reducers were important contributors to demethylation only on the second survey while methanogens were important only on the fourth survey (Figure 16b).

Methyl Bay

The methanogens and sulfate reducers jointly accounted for all of methylation and demethylation activity in both the flooded and offshore zones (Table 7). Methanogens accounted for about the same proportion (35%) of total methylation in the flooded zone of Methyl Bay as at the nearshore stations in Granville Lake while the relative contribution of sulfate reducers to total methylation in the flooded sediments (65%) was about twice that in nearshore Granville Lake. Both groups accounted for larger portions of total demethylation than in Granville Lake but, again, there was a greater increase in the contribution by sulfate reducers (from 23 to 55%) than by methanogens (from 29 to 42%). Methanogens and sulfate reducers each accounted for about half of total demethylation at the offshore station (Table 7).

In contrast to Granville Lake, where the methanogens and sulfate reducers did not make significant concurrent contributions to total methylation, these groups made large and approximately equal contributions to total methylation through the July pulse both at the flooded stations and offshore (Figure 17 a and b). Sulfate reducers were the dominant, identifiable, contributors to the August pulse of methylation at the flooded stations. Similarly, the two groups jointly contributed to total demethylation throughout the two months of study. The first pulse of demethylation was the product of approximately equal contributions while the second pulse was driven by the sulfate reducers. Methanogens accounted for a larger proportion of demethylation than methylation through August (Figures 17a and 18a). The August pulse of demethylation at the

Demethylation Activity Attributable to Methanogens and Sulfate Reducers
Granville Lake, Nearshore - 1988



Demethylation Activity Attributable to Methanogens and Sulfate Reducers
Granville Lake, Offshore - 1988

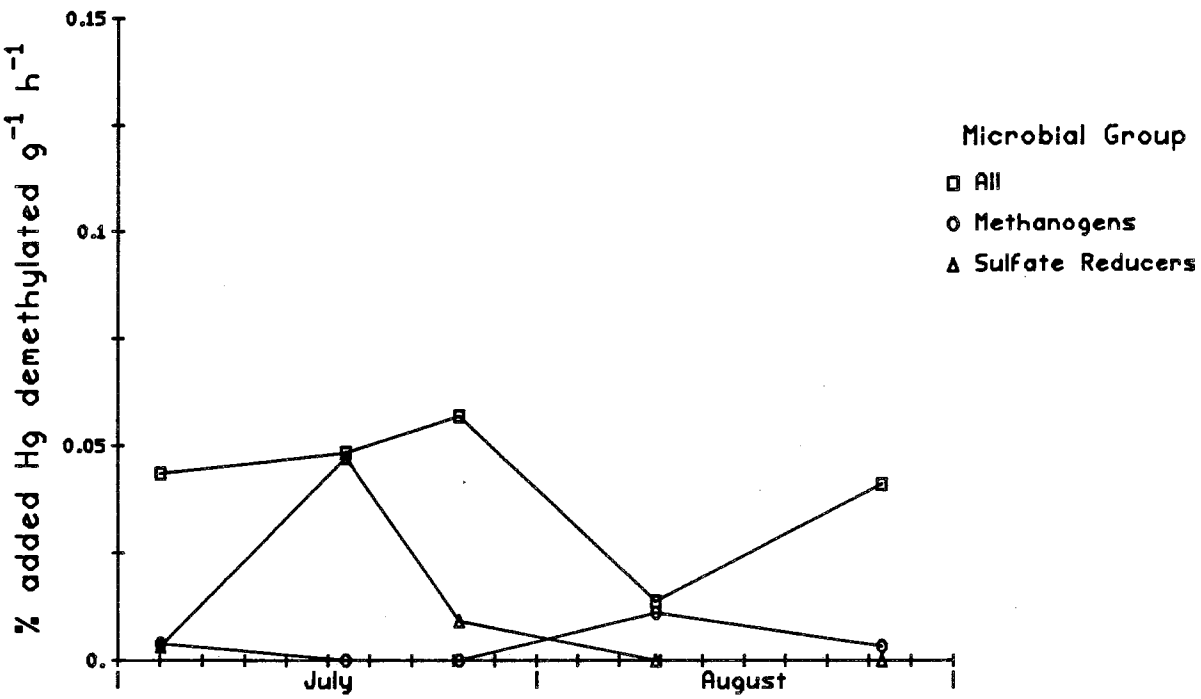
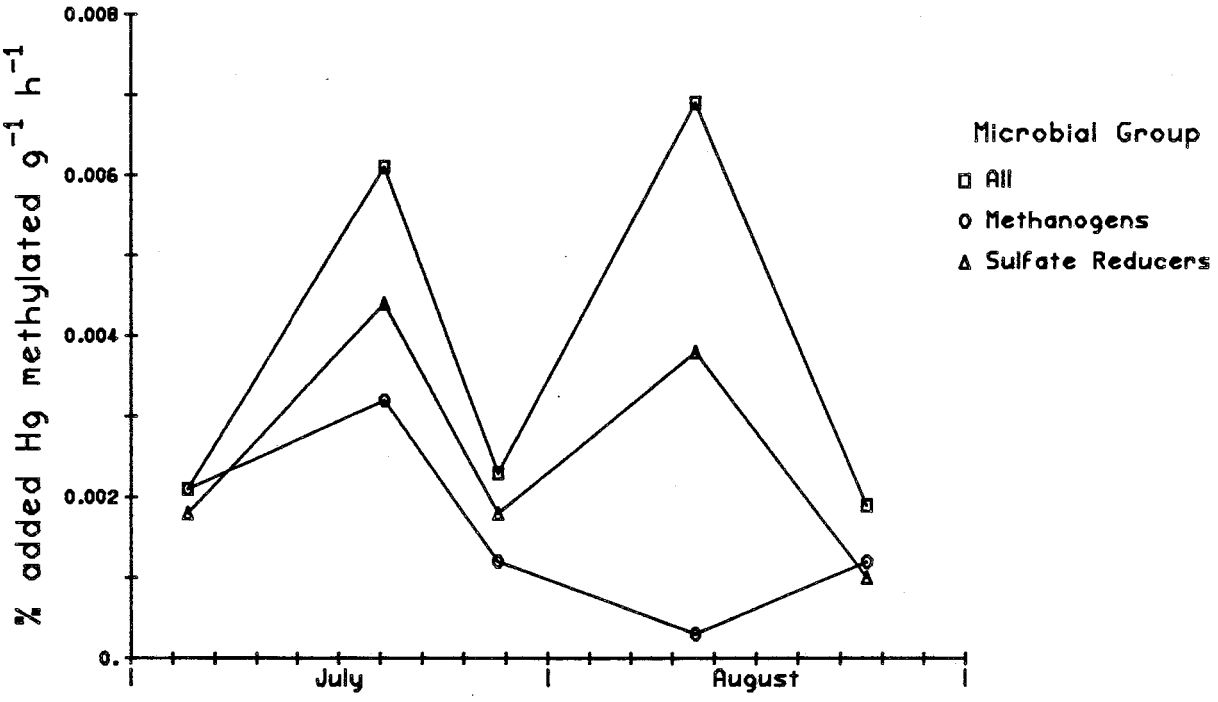


Figure 16. a, upper, and b, lower.

Methylation Activity Attributable to Methanogens and Sulfate Reducers
Methyl Bay, Nearshore - 1988



Methylation Activity Attributable to Methanogens and Sulfate Reducers
Methyl Bay, Offshore - 1988

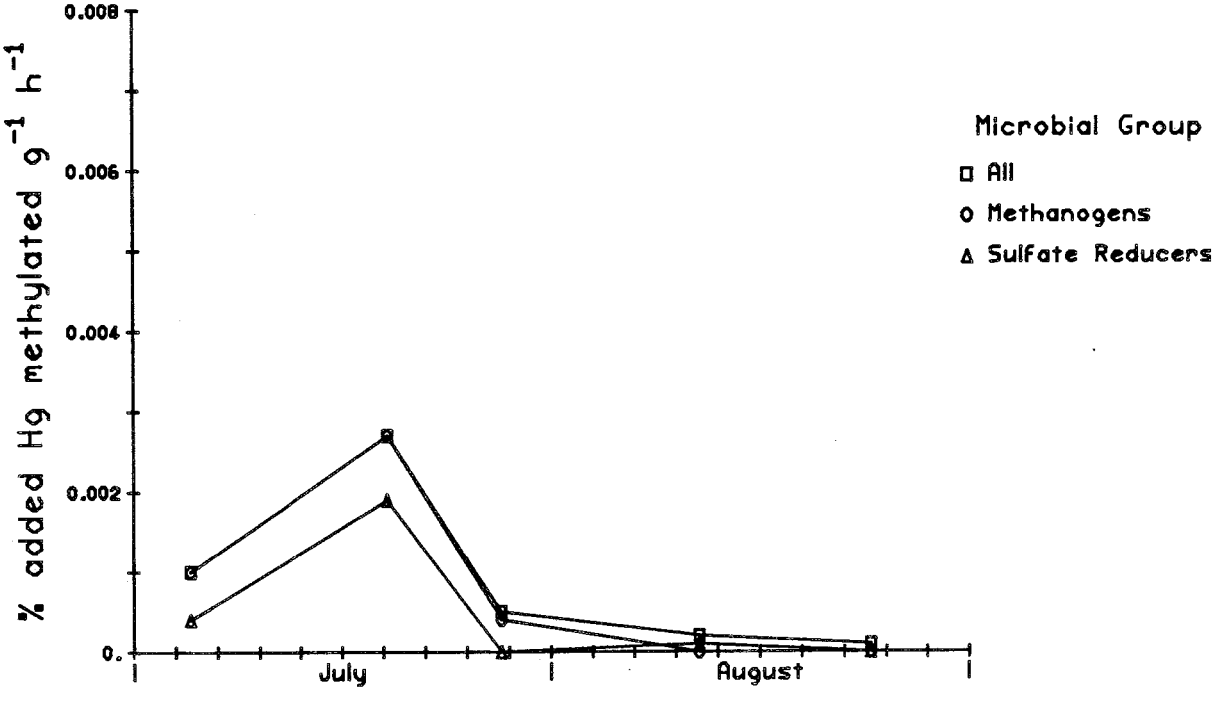
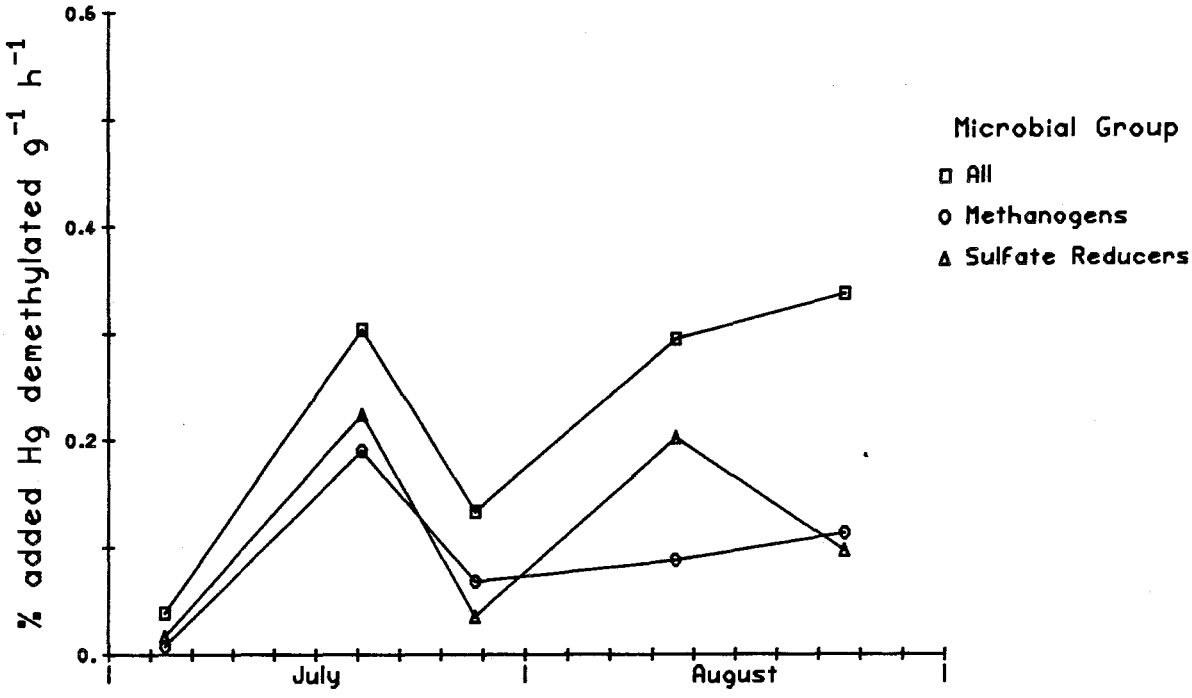


Figure 17. a, upper, and b, lower.

Demethylation Activity Attributable to Methanogens and Sulfate Reducers
Methyl Bay, Nearshore - 1988



Demethylation Activity Attributable to Methanogens and Sulfate Reducers
Methyl Bay, Offshore - 1988

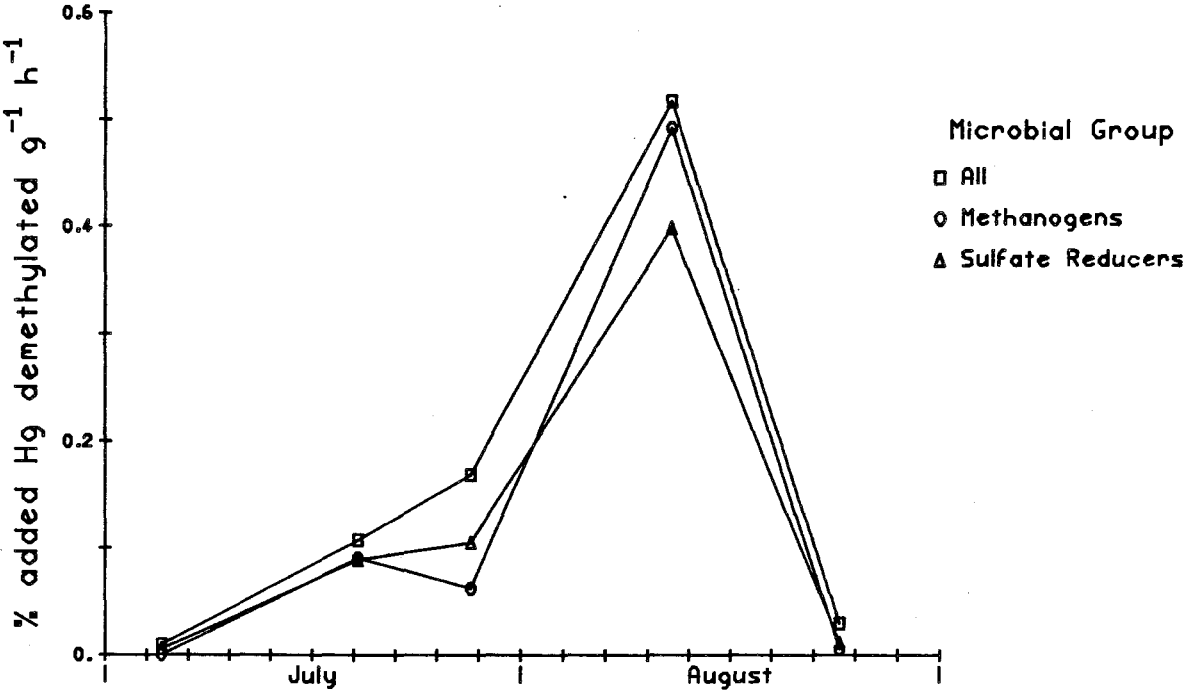


Figure 18. a, upper, and b, lower.

offshore station was jointly driven by the methanogens and sulfate reducers (Figure 18b).

On several occasions, the sum of methylation or demethylation attributable to the methanogens and sulfate reducers exceeded the measurement of total methylation or demethylation. This was probably a result of local spatial variation in methylation and demethylation. The inhibition assays were done on sediments from the same area as the assay of total methylation but could not be done on the same sample. The degree of variation in both methylation and demethylation in samples from the same area is considerable. For example, seasonal mean total methylation rates measured at the three nearshore stations in Methyl Bay had a coefficient of variation of 25% (Table 6) and coefficients of variation were higher than 25% on individual sampling dates (Appendix Table D1). Although the sum of methylation or demethylation activity in the inhibited samples could exceed the total, the excess is generally accounted for by the error or variance in the estimates.

Discussion

Microbial Inhibition Experiment

The microbial inhibition studies were carried out in 1988 to provide a gross identification, at the level of metabolic group, of the methylating and demethylating bacteria in sediments at the long-term methylation monitoring sites. This work was stimulated by the results of a pilot study conducted in Methyl Bay in the summer of 1987 which indicated that the methylating and demethylating bacteria in the flooded sediments might belong to different metabolic groups and therefore be amenable to selective control or stimulation. It also was not known to what extent, if any, the microbial community, and particularly the methylating and demethylating bacteria in flooded sediments

from Methyl Bay, differed from that in sediments from Granville Lake. The study was limited to the methanogens and sulfate reducers because these were the groups that previous investigations had identified as the most likely to be important in mercury transformations (Wood et al. 1968; Compeau and Bartha 1985).

The results of the study indicate that the methanogens and sulfate reducers were indeed the correct groups to single out for study. Together, they accounted for the majority, but not all, of methylation and demethylation in Granville Lake. Conditions in the flooded zone appear to be particularly attractive to these groups which jointly accounted for more than 95% of total methylation and demethylation there.

While methanogens have long been suspected as methylators of mercury in the natural environment (Wood et al. 1968), the importance of sulfate reducers in this process is a relatively recent discovery (Compeau and Bartha 1985). It was previously thought that sulfate reducers would inhibit mercury methylation by precipitating Hg^{2+} as HgS which is not available for methylation. This may occur at high sulfate concentrations, as found in marine sediments, but does not seem to be important at low sulfate concentrations (Compeau and Bartha 1985). This study confirms the importance of both groups as methylators of mercury both in natural lake sediments and in flooded terrestrial sediments in reservoirs.

Previous studies of the demethylation process have identified numerous types of bacteria - including obligate aerobes, facultative anaerobes, and obligate anaerobes - as capable of demethylating organomercurials although the relative importance of these various bacterial types in nature was unknown (Spangler et al. 1973). The results of this study indicate that obligate anaerobes, specifically the methanogenic and the sulfate reducing bacteria, are

important contributors to demethylation in natural freshwater sediments and are specifically selected for by conditions in flooded terrestrial sediments in northern reservoirs.

Although the inhibition experiments provided new information on the composition of the microbial communities responsible for methylation and demethylation in lakes and reservoirs, the results also indicate that methylation and demethylation are carried out by bacteria belonging to the same metabolic groups. Consequently, it may not be possible to develop a measure to independently regulate rates of methylation or demethylation. Different strains or genera may be responsible for the two processes and regulation might be possible at that level. However, considerably more work would be required to determine if this was the case and any regulatory measure would have to be very specific. It is not known if such specific control is possible or practical. Rather than attempting to control the activity of specific groups or taxa, it may be possible to create conditions to encourage the spread of the gene responsible for the production of the demethylation enzyme, organomercurial lyase.

The pilot study indicated the importance of the methanogens as methylators and this was confirmed in the present study. However, the results of the two studies differ significantly in that the pilot study suggested that different microbial groups may be responsible for methylation and demethylation in the flooded zone while the present study has shown that the same groups carry out both processes. The reason for the discrepancy between the studies appears to be the considerable spatial and temporal variation in the contributions of the two groups of bacteria to methylation and demethylation. A group may dominate one process but not the other at a given time and place but make equal contributions to both methylation and demethylation on average over the summer.

More than one station must be sampled several times over the summer season to provide a true measure of the contributions of different microbial groups.

Factors Regulating Microbial Methyl Mercury Production

Organic Matter and Total Mercury

A number of factors were identified in the regression analysis as potentially regulating microbial methylation or demethylation and consequently the net rate of production of methyl mercury. As in previous studies in northern reservoirs (Ramsey 1987 a and b, 1988), specific methylation rates were correlated with the concentrations of organic matter, as measured by the organic carbon and total nitrogen concentrations, and with total mercury. While both variables may be important, the accumulating evidence indicates that organic matter availability is of primary importance in regulating methyl mercury availability in reservoirs. This evidence comes both from specific sites in the Churchill River diversion as well as from studies in other areas where the concentrations of mercury and organic matter have been found to vary independently. These studies are discussed in more detail in Ramsey (1988). A more recent example of the dominant role of organic matter comes from an experiment conducted in streams of the Queen Charlotte Islands to examine the effects of soils with varying concentrations of mercury and organic matter on microbial methylation and demethylation rates (Ramsey in prep.). It was found that specific methylation rates in soils with low organic content but with concentrations of total mercury up to 550 ng g^{-1} were the same as in low mercury soils or in sediments from Granville Lake. Conversely, soils with low mercury concentrations (71 ng g^{-1}, 45% lower than in Granville Lake) but with high organic content produced specific microbial methylation rates comparable to the mean methylation rate measured at the nearshore stations in Methyl Bay

this year.

pH

Another factor which emerged from the correlation analysis as possibly affecting, or regulating, net methylation is sediment pH. Although pH is known to affect methylation and demethylation, and particularly the latter, there is no reason to expect that the small range of pH found here should have any effect on either process (Ramlal et al. 1985). The finding of significant correlations between pH and both methylation and demethylation is more likely a result of colinearity; pH also was correlated with measures of sediment organic and mercury content (Table 4).

Redox Potential

The influence of oxygen availability, and consequently redox potential, on the net rate of mercury methylation has been identified in previous studies in the Churchill River diversion (Ramsey and Ramlal 1987a; Jackson 1988). In this study, methylation activity at the offshore station in Sokatisewin Lake was much higher than would be expected given the comparatively low concentration of organic carbon in the sediments (Figure 19). Unique characteristics of this station are the low redox potential in sediments and the low near-bottom oxygen concentration (Figures 5 and 6); conditions found in the previous studies to stimulate bacterial mercury methylation. Given that the groups of bacteria which are principally responsible for methylation are obligate anaerobes, it is perhaps reasonable to expect greater rates of methylation with decreasing oxygen concentration (and consequently decreasing redox potential). In contrast, the lack of correspondingly high demethylation activity at this site is contrary to our knowledge of the identity of the demethylators which in

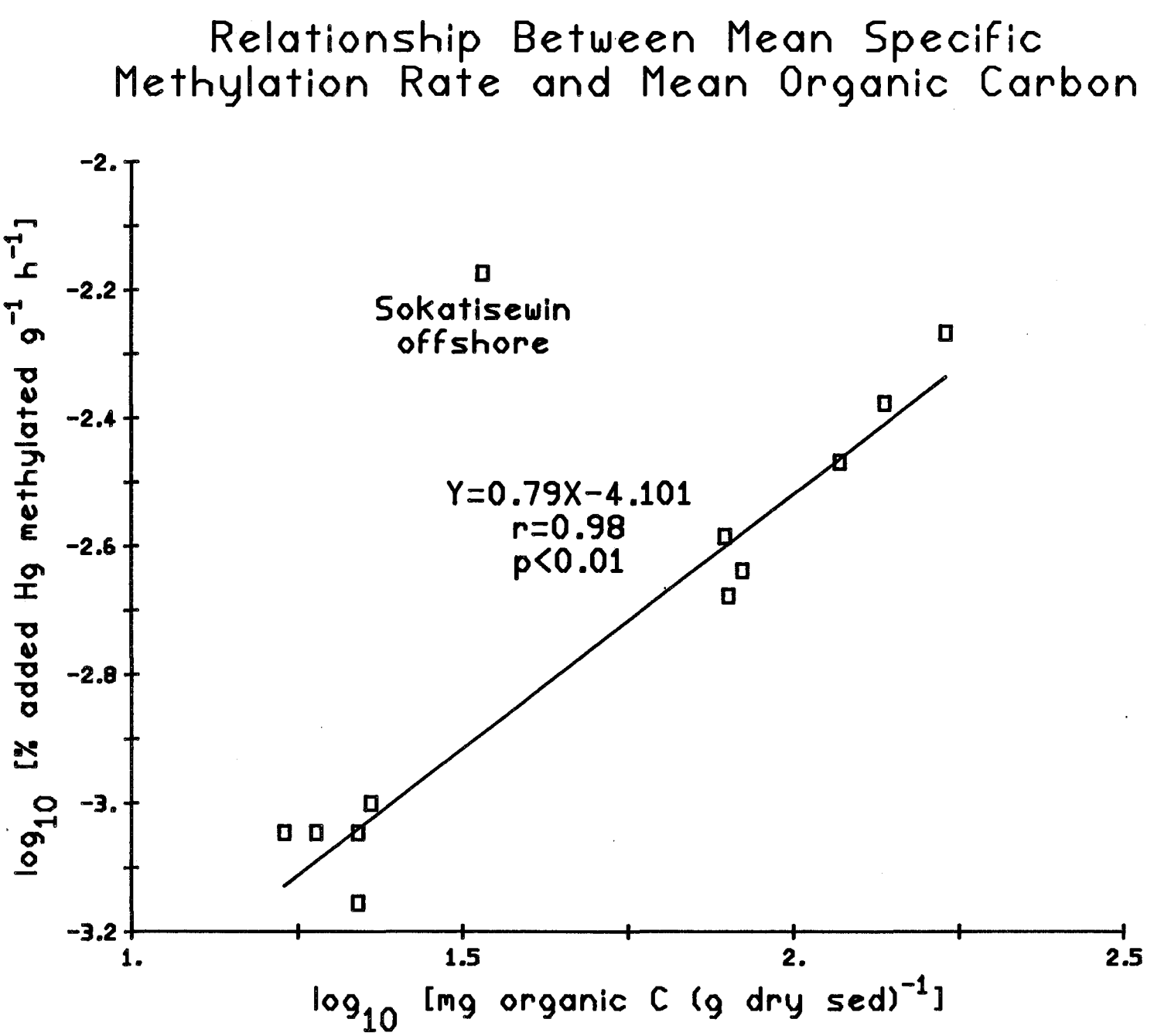


Figure 19. Relationship between the mean specific methylation rate and the mean concentration of organic carbon. Sokatisewin Lake offshore not included in the regression.

Methyl Bay also are obligate anaerobes. The inhibition studies were not extended to Sokatisewin Lake this year so it is not possible to determine whether this discrepancy is real or is the result of different demethylators dominating there.

Microbial Activity

The hypothesis developed to explain the occurrence of elevated net microbial mercury methylation in new reservoirs states that the inundated organic matter stimulates microbial activity and in turn microbial methylation of mercury. The methylating bacteria are stimulated to a greater degree than the demethylating bacteria because of the different mechanisms of the two processes; many different microbes can methylate mercury while demethylation requires a specific enzyme, organomercurial lyase (Robinson and Tuovinen 1984). The genetic information for the production of this enzyme is encoded on a sexually transmitted plasmid (Pan-Hou et al. 1980) rather than on the bacterial chromosome so that the size of the community of demethylators should not solely be related to overall microbial activity. If this hypothesis is correct, then microbial methylation rates should be positively correlated with microbial activity. Demethylation rates would not necessarily be correlated with microbial activity but, if so, the slope should be lower. The results of the study do not completely support the hypothesis when all of the sites are considered together. Demethylation rates increased with increasing microbial activity but methylation rates varied independently of microbial activity (Figures 20 and 21).

Upon closer inspection, however, it appears that the methylation data fall into two groups. Rates of microbial activity and of both methylation and demethylation are elevated at the flooded stations in Methyl Bay (Figure 21).

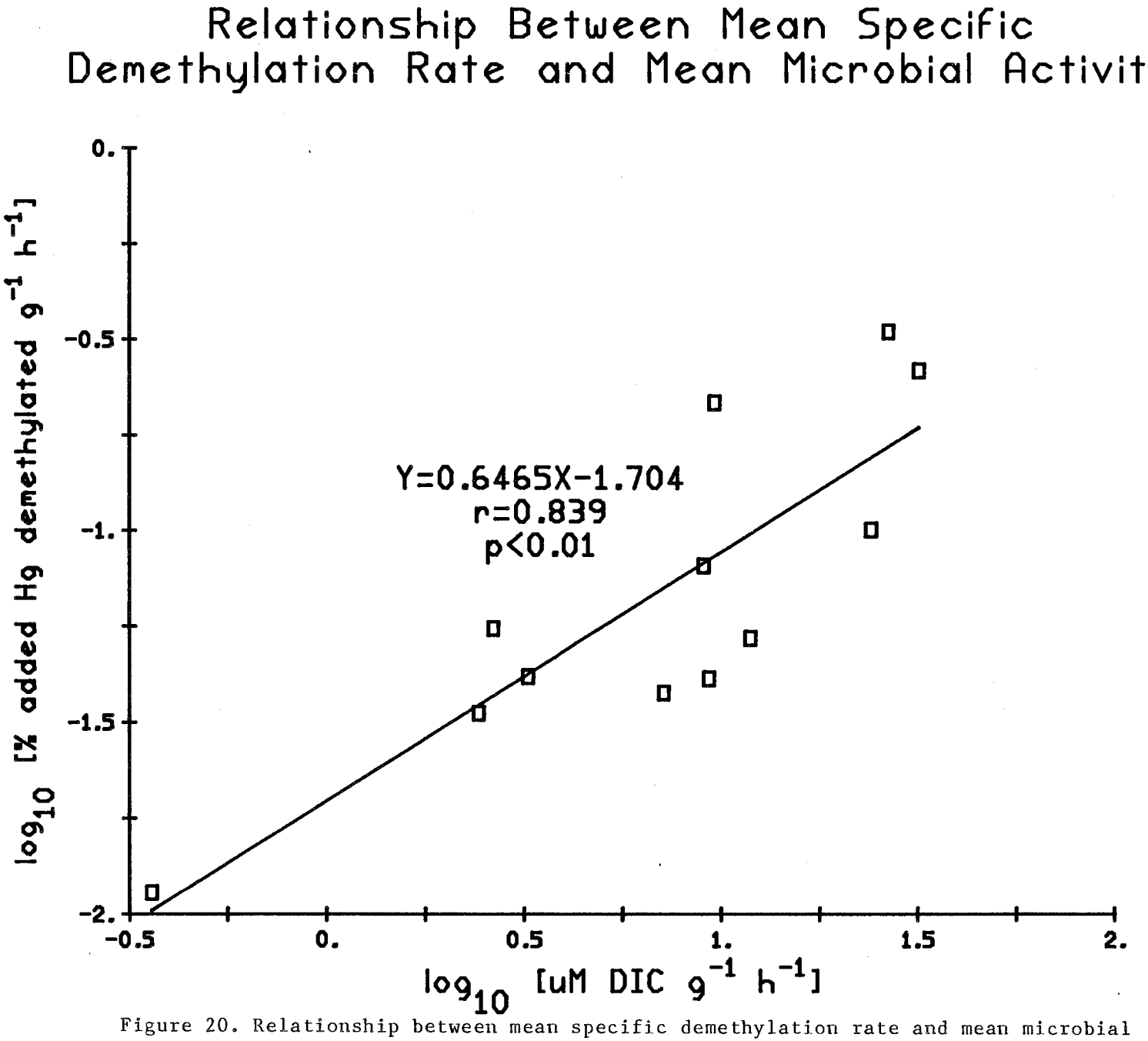


Figure 20. Relationship between mean specific demethylation rate and mean microbial activity.

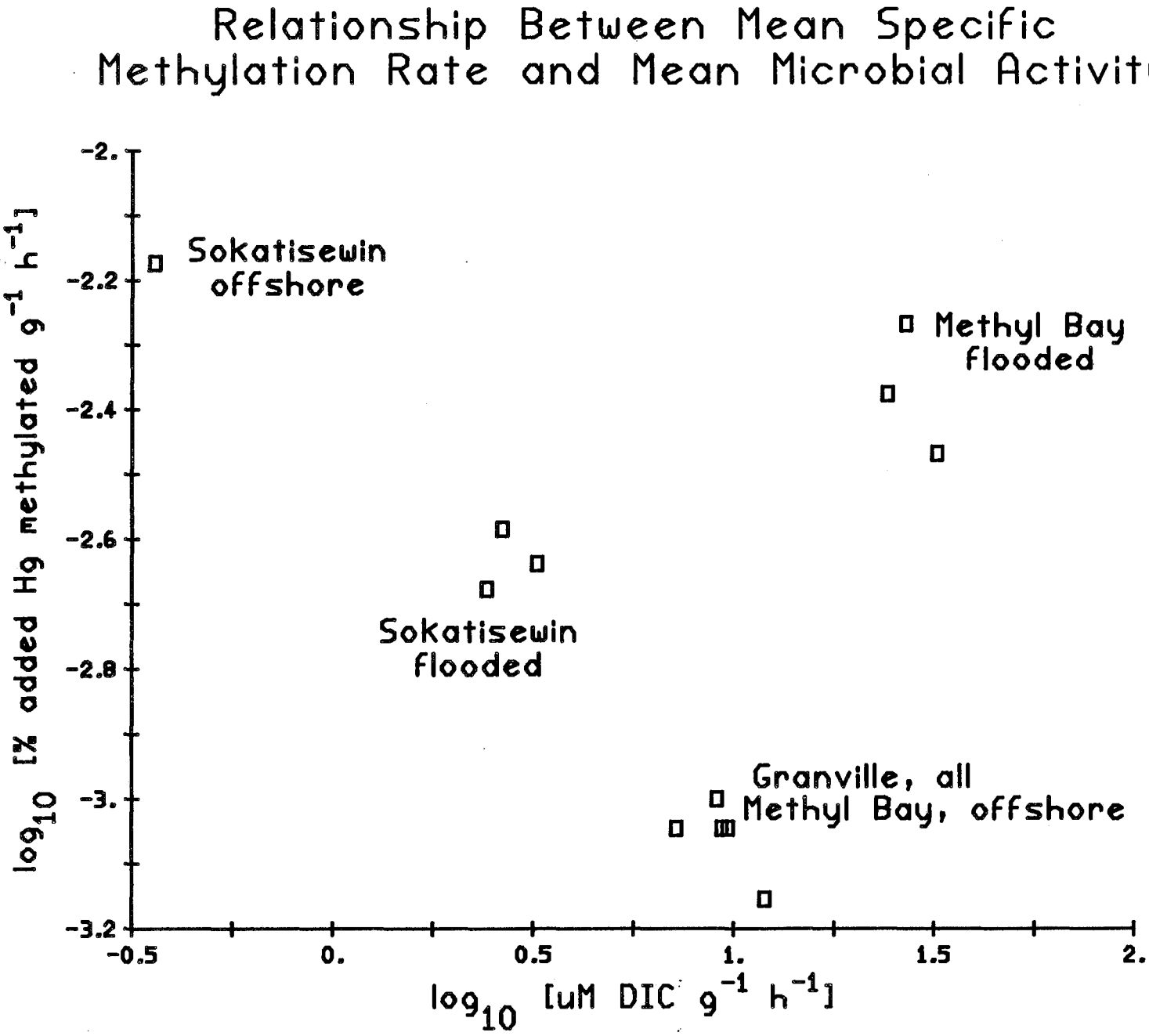


Figure 21. Relationship between mean specific methylation rate and mean microbial activity.

Consistent with theory, methylation rates are proportionately higher than demethylation but both are disproportionately high compared to microbial activity. Microbial activity averaged 2.7 times higher in flooded sediments from Methyl Bay than in nearshore sediments from Granville Lake yet methylation rates were 4.8 times higher and demethylation rates averaged 3.9 times higher (Tables 5 and 6). The higher rates of both processes than expected given the degree of stimulation of overall microbial activity could be explained by the different composition of the methylating and demethylating communities in the flooded sediments than in Granville Lake if those groups which are selected are more efficient methylators and demethylators. Similarly, the greater stimulation of methylation over demethylation may be a result of the different relative contributions of the methanogens and sulfate reducers to methylation than to demethylation. However, it is not known if the two groups have differing efficiencies of methylation or demethylation either from that of other microbes or from each other.

The occurrence of elevated methylation rates in the presence of low microbial activity in Sokatisewin Lake (Figure 21) may represent the confounding influence of the lower redox potential in these sediments and particularly at the offshore station. It has already been shown that conditions in flooded sediments in Methyl Bay have selected for obligate anaerobic bacteria which might be expected to be more active under the lower redox conditions in Sokatisewin Lake despite the lower total microbial activity. A factor which was not measured is the contribution which the groups responsible for methylation and demethylation make to total microbial activity. Considering that the entire microbial community probably is not active in methylation and demethylation, it may be more appropriate to measure the activity only of those groups which are. That only part of the microbial

community may be active in methylation and that the fraction responsible may vary depending on sediment conditions may prevent the definition of any simple relationship between methylation and total microbial activity. It is now apparent that it will be necessary to measure the activity of the groups responsible for methylation and demethylation.

Methylation Balance in Methyl Bay, 1984-1988

Measurements of specific methylation and demethylation have now been made at this site using consistently applied methods since 1984. Through 1987, there was no clear indication that either specific methylation rates or the methylation balance (M/D) in the flooded zone of Methyl Bay had declined relative to the nearshore zone of Granville Lake (Ramsey 1988). Although methylation rates in Methyl Bay have shown a consistent decline in absolute values since 1984 (Table 8), the relative nature of the rate should be kept in mind. The same declining trend is evident in Granville Lake and the ratio of specific methylation in Methyl Bay to rates in Granville Lake actually was highest in 1986. Methylation rates in Methyl Bay started to decline relative to rates in Granville Lake in 1987 and the results of this study indicate the decline continued in 1988 (Table 8).

Demethylation rates in Methyl Bay and Granville Lake also have varied in phase over the past 5 years. The highest absolute rates were measured in 1985 and activity decreased in each of the following two years at both locations (Table 8). Rates increased again in both Methyl Bay and Granville lake in 1988 but the differential did not. Although not as clear a trend as the decline in methylation, the differential in demethylation rates between Methyl Bay and Granville Lake in each of 1987 and 1988 has been lower than in 1986.

As a result of these year to year variations in methylation and

demethylation activity, the differential in methylation balance (M/D) between Methyl Bay and Granville Lake has varied as well. The largest differential in seasonal mean M/D was recorded in 1984, the lowest was measured in 1988 (Table 8). Despite the low M/D in 1988, no clear declining trend is evident in the past 5 years of measurements.

From 1983 to 1987, the elevated rates of methylation and demethylation in Methyl Bay were restricted to the flooded zone (Ramsey and Ramlal 1987 a and b; Ramsey 1987; Ramsey 1988). In 1988, methylation rates remained low at the offshore station in Methyl Bay but elevated demethylation rates were measured there for the first time (Table 6). No obvious change in the sediment conditions has occurred at the offshore station and microbial activity was low relative to the flooded zone yet the demethylation rate measured in mid-August was almost twice as high as in the flooded zone. It is unlikely that this represents a laboratory error. Potential sources of laboratory error include the addition of multiple methyl mercury spikes to the samples and of leaks in the incubation vessels which would allow the labelled ^{14}C to escape. Multiple spikes are unlikely. The high rates were measured in all six assays made on this date (2 regular and 4 inhibited) and it is improbable that that multiple spikes were added to all 6 sample bottles from this station, and only this station. Leaks in the incubation vessel cannot account for the high rates because the loss of labelled CO_2 would instead produce artificially low rates. At least one more year of study is required to determine if this represents a new trend in Methyl Bay or if the high rate this year is just a local anomaly.

Projected Duration of the Mercury Problem in the Churchill River Diversion

Sokatisewin Lake was selected for this study because of its age (flooded 1929) and the small annual variation in reservoir level as a test of the model

Table 8. Comparison of seasonal mean specific methylation and demethylation rates (% added Hg g^{-1} h^{-1}) and the corresponding M/D ratios in the flooded zone of Methyl Bay and in the nearshore zone of Granville Lake: 1984-1988.

	1984		1985		1986		1987		1988	
	Mean	Range	Mean	Range	Mean	Range	Mean	Range	Mean	Range
Methylation										
Methyl Bay	0.034	(0.006-0.061)	0.022	(0.009-0.030)	0.012		0.002	(0.0014-0.0027)	0.0043	(0.0034-0.0054)
Granville Lake	0.012		0.006	(0.004-0.012)	0.0008	(0.0006-0.001)	0.0002	(0.0001-0.0003)	0.0009	(0.0007-0.0010)
Ratio	2.8		3.7		15.0		10.0		4.8	
Demethylation										
Methyl Bay	0.089	(0.049-0.129)	1.20	(0.970-1.390)	0.162		0.0468	(0.0321-0.0551)	0.2312	(0.1004-0.3313)
Granville Lake	0.162		0.669	(0.531-0.918)	0.031	(0.026-0.036)	0.0166	(0.0139-0.0192)	0.0581	(0.0412-0.0809)
Ratio	0.5		1.8		5.2		2.8		4.0	
M/D										
Methyl Bay	0.383	(0.122-0.473)	0.018	(0.028-0.055)	0.077		0.0434	(0.0377-0.049)	0.0238	(0.0130-0.0418)
Granville Lake	0.076		0.009	(0.003-0.021)	0.026	(0.022-0.029)	0.0102	(0.0052-0.0181)	0.0159	(0.0124-0.0218)
Ratio	5.0		2.0		3.0		4.3		1.5	

developed with 1986 data (Ramsey 1987a) to predict the evolution of mercury problems in northern reservoirs. Based on this model, specific methylation and demethylation rates and M/D in flooded sediments from Sokatisewin Lake should be at or near the background levels represented by Granville Lake.

On first inspection, the ability of the 1986 model to predict the evolution of methylation activity is rather poor. Specific methylation rates in the nearshore zone of Sokatisewin Lake were significantly lower (by about 50%) than in Methyl Bay but the model predicted that methylation activity should be at background levels (i.e., the measured rates were about double the predicted rates; Table 6). However, when all of the methylation rate data from the past 3 years are compared it appears that the model may indeed be correct. Methylation activity declines with time at an exponential rate that is similar, but not identical, to the curve described by the 1986 data alone (Figure 22). The slope of a least-squares regression line fitted to a double-ln plot of the data was significantly different from zero ($p=0.028$) and the regression of standardized methylation on reservoir age explained 66% of the variation in methylation among the different reservoirs and between years in Methyl Bay (Figure 23).

The only real, and not terribly surprising, failure of the original model is its poor ability to predict methylation rates in reservoirs outside the age range for which data were available Other differences between the 1986 model and the revised model include a faster rate of decline in the first 20-30 years after impoundment and a slower rate of decline in subsequent years (Figure 22). Based on the revised model, methylation activity in the flooded zone of Southern Indian Lake is not expected to return to background levels until the year 2135, 159 years after the lake was flooded. It may be rather difficult to confirm this prediction locally. The oldest reservoir in Manitoba that is

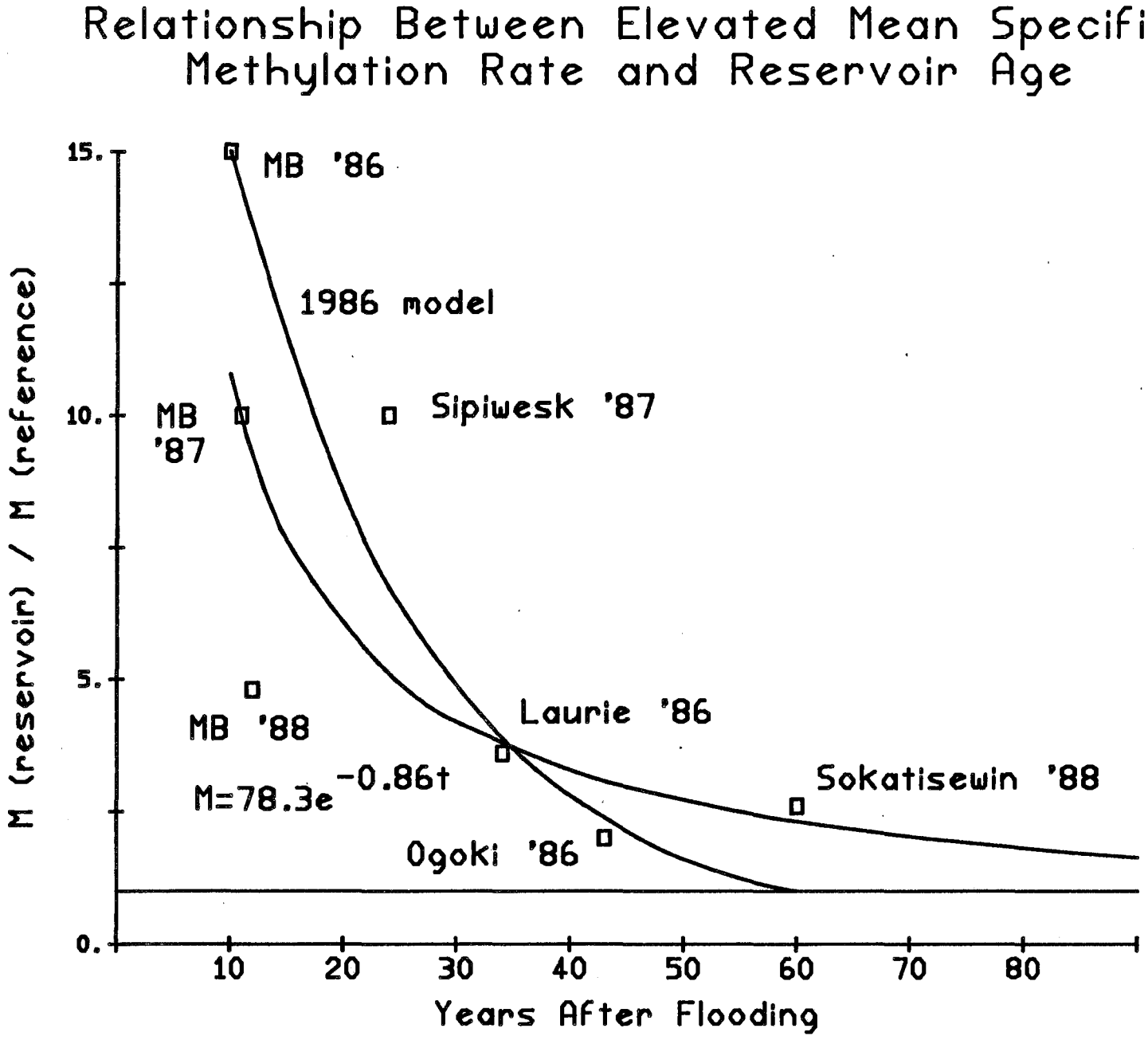


Figure 22. Relationship between the degree of elevation in the mean specific methylation rate and reservoir age. The curve for the 1986 model is from Ramsey (1987a). The curve for the revised model is back-transformed from Figure 23.

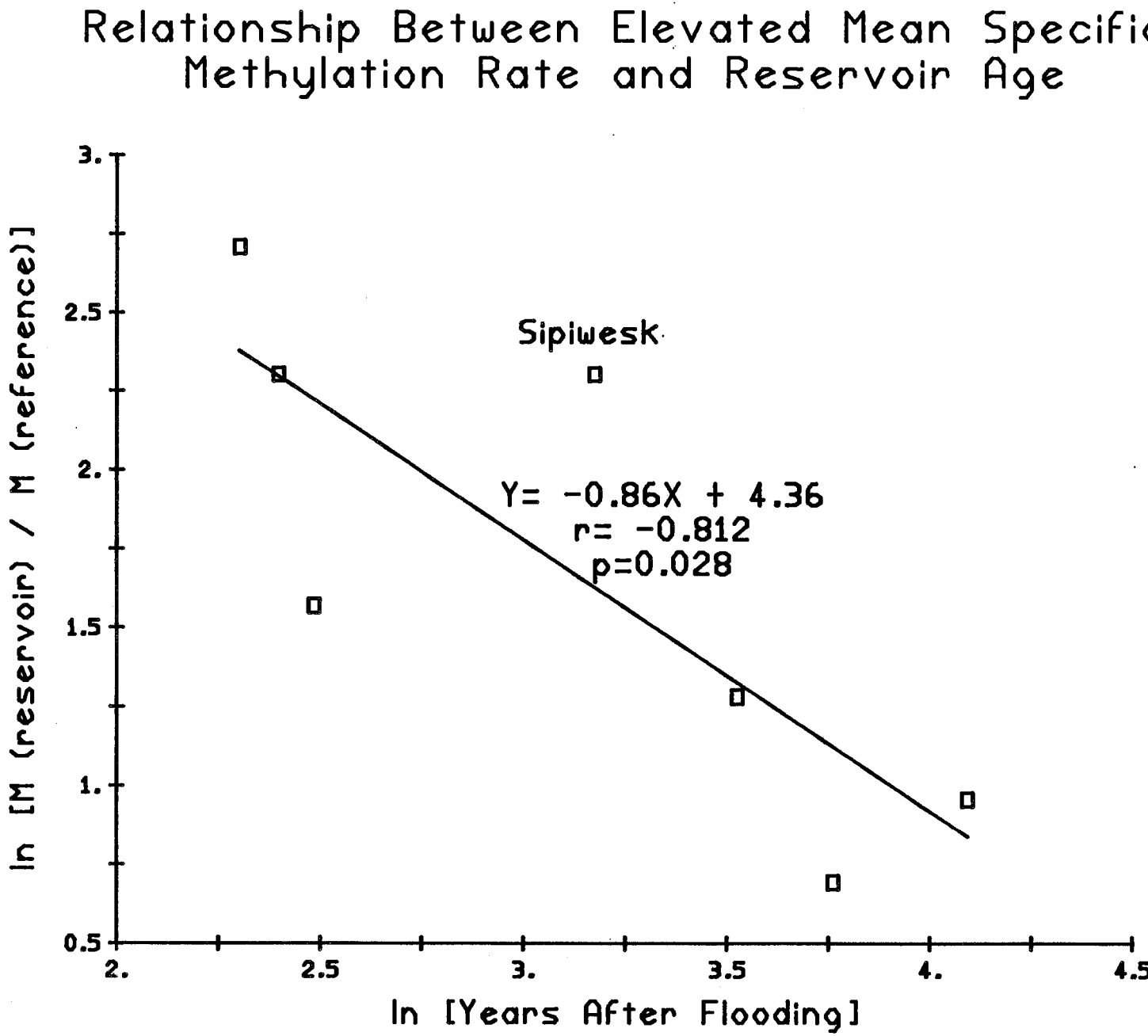


Figure 23. Double-ln plot of elevation in mean specific methylation and reservoir age.

located in the boreal forest zone is at Pointe du Bois (age 78, flooded in 1911, D. Windsor, Manitoba Hydro, pers. comm.) on the Winnipeg River.

Another aspect of the methylation-reservoir age relationship that is in need of study is the expected rate of decline of methylation rates in Methyl Bay over the next decade. There is some indication in Figure 22 that methylation activity in Methyl Bay may be declining faster than expected on the basis of the measurements in older reservoirs. If the decline continues at the same rate, methylation should reach background levels in the next couple of years. However, the measurements in older reservoirs suggest that the rate of decline should slow in the next few years and that methylation rates will persist at or near the present level for at least another 50 years and possibly for another 140 years. Continued measurements of the methylation balance in Methyl Bay and Granville Lake as well as extension of the measurements to Stephens Lake, flooded in 1971, will provide this most essential information.

As predicted by the original model, the mean specific demethylation rate in Sokatisewin Lake did not exceed that in Granville Lake and was actually 25% lower (Table 6). The model would therefore appear to have reasonable predictive ability with respect to the long-term evolution of demethylation rates in northern reservoirs of the type represented in the Churchill River diversion. Again, the slope of a least-squares regression line fitted to a double-ln plot of all three years of data was significantly different from zero ($p<0.01$) and the regression accounted for 82% of the variance in mean demethylation (Figure 24). Again, however, the inclusion of two more seasons of data indicates that the initial rate of decline will be faster than originally predicted (Figure 25).

A faster rate of decline of both methylation and demethylation in younger than in older reservoirs is more consistent with our knowledge of organic

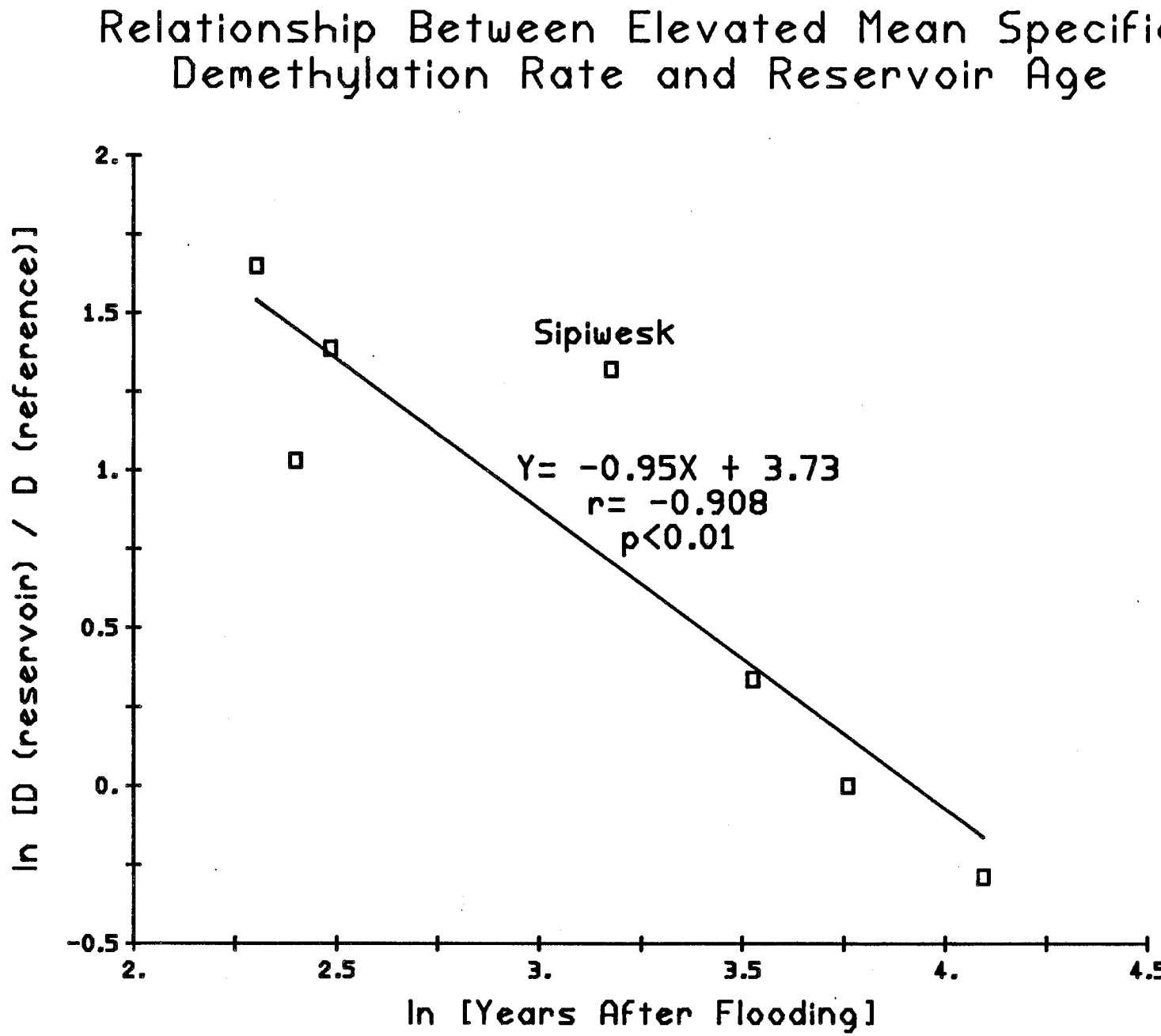


Figure 24. Double-ln plot of elevation in mean specific demethylation rate and reservoir age.

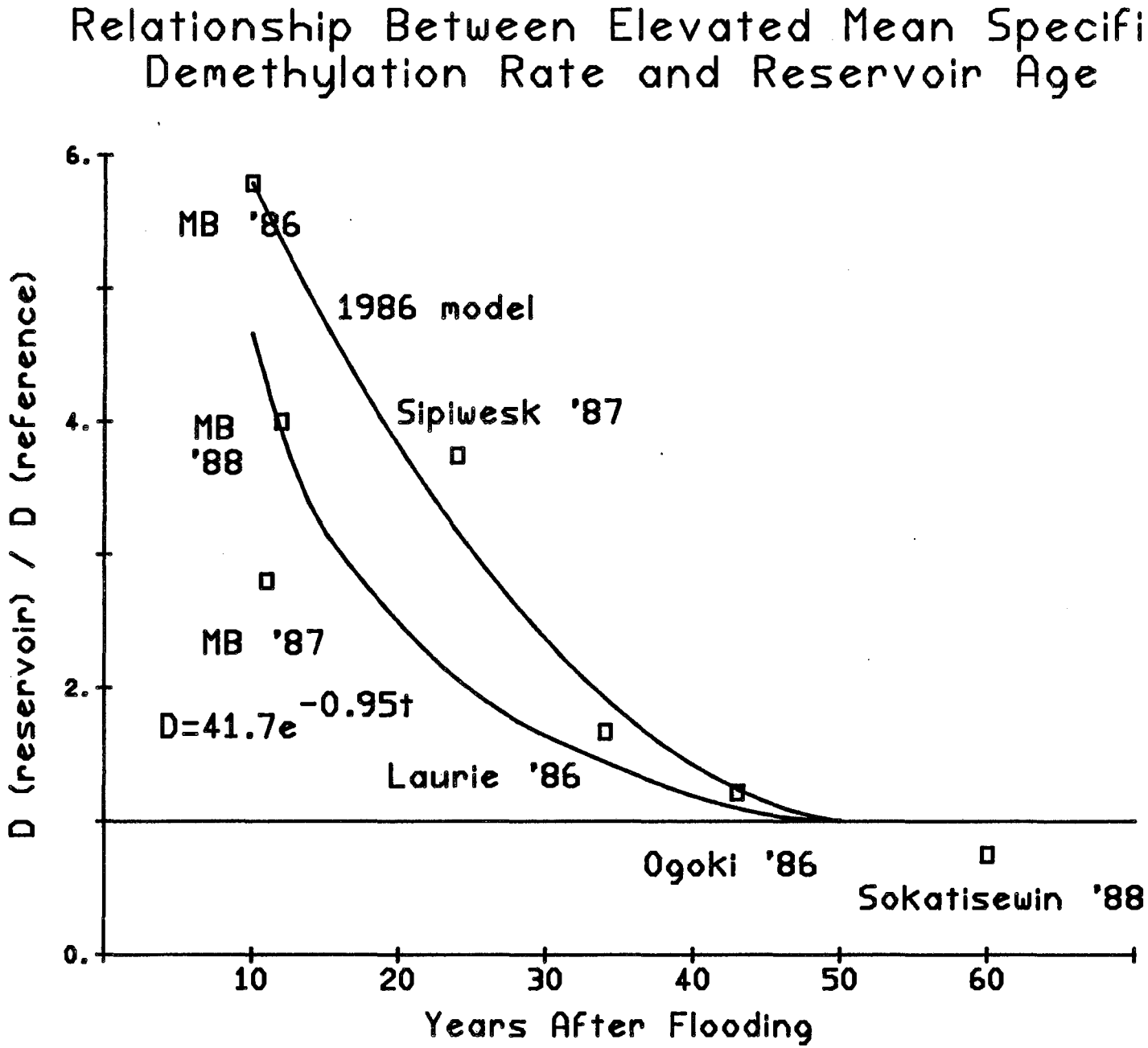


Figure 25. Relationship between the degree of elevation in the mean specific demethylation rate and reservoir age. The curve for the 1986 model is from Ramsey (1987a). The curve for the revised model is back-tansformed from Figure 24.

matter decomposition processes and of the composition of the organic matter flooded in these northern reservoirs than is a constant rate. Many types of organic matter with very different rates of decomposition were flooded by the Churchill River diversion. For example, spruce needles are processed quickly in Southern Indian Lake (Crawford and Rosenberg 1984), with 85% disappearance in 1 year. In contrast, Hodkinson (1975) reported that 7.5 cm diameter poplar bole (trunk) had a half time of 57 years and a 95% life of 246 years in a northern pond. The rapidly decomposed materials feed the initially high rates of methylation and demethylation. The rates decline quickly as these materials are consumed but the large pool of more slowly degraded material maintains the rates above background levels for many decades. Although this provides a general explanation for the shape of the curves, there is not yet any clear explanation for the persistence of elevated methylation activity after demethylation rates have returned to background levels.

A revised projection of the rate of decline in net methyl mercury production, as indicated by the M/D ratio, in flooded sediments along the Churchill River Diversion, incorporating the revised projections of the rates of decline of methylation and demethylation activity, is presented in Figure 26. No decline in M/D, and perhaps even a slight increase, is expected for about 50 years after impoundment. The subsequent slow decline is regulated by the rate of decrease in methylation activity. The observed M/D ratios are generally consistent with this projection although the fit to the predicted line is rather poor. However, a close fit should not necessarily be expected because M/D compounds any variability in the component measurements and there is unaccounted variation in the regressions of both methylation and demethylation on reservoir age (Figures 23 and 24).

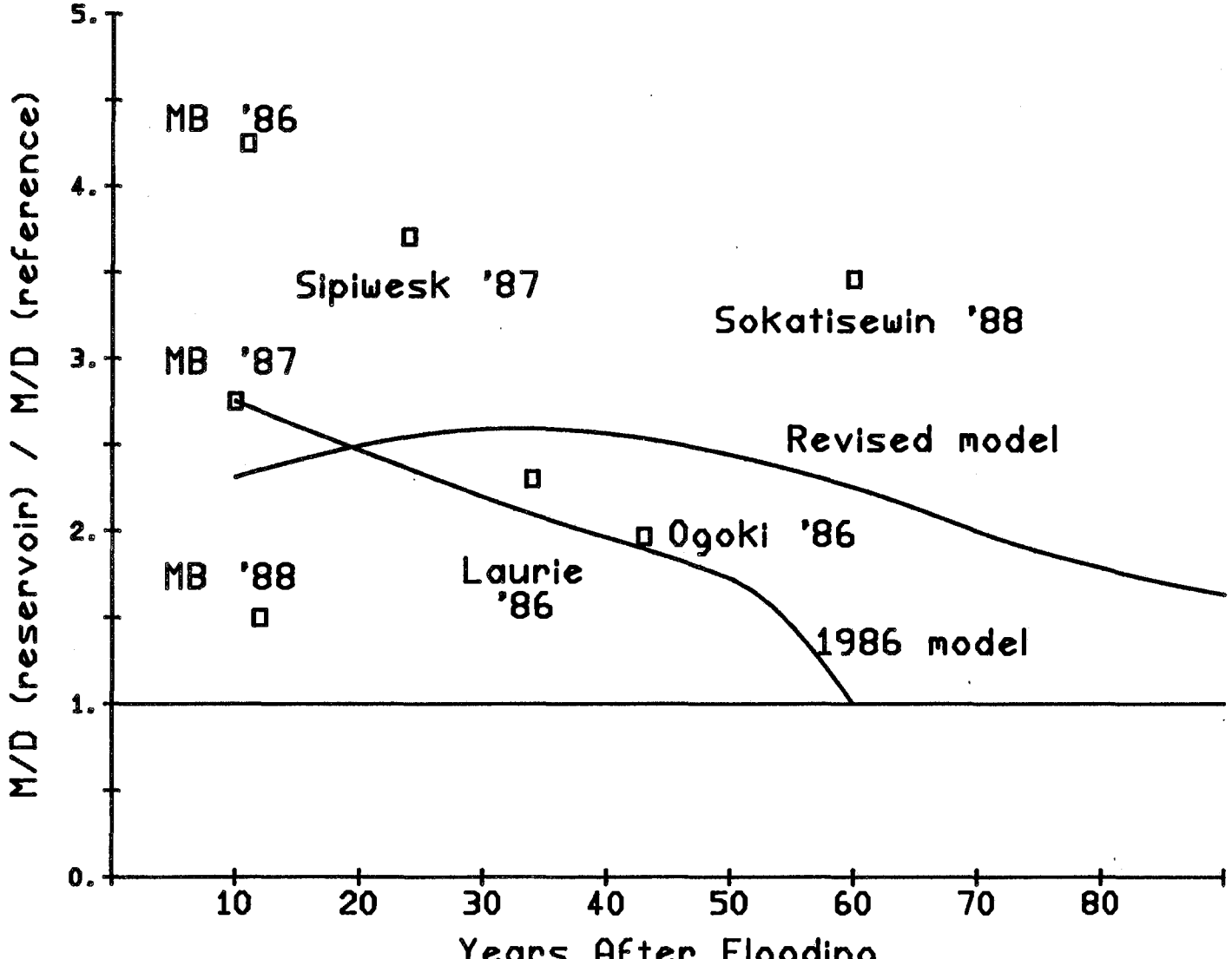


Figure 26. Relationship between elevation in seasonal mean methylation balance and reservoir age. The curves for the 1986 and revised models were derived from the fitted relationships between methylation, demethylation, and reservoir age shown in

Resolution of the Sipiwesk Lake Discrepancy

Based only on the 1987 data, rates of both methylation and demethylation in Sipiwesk Lake appeared to be much higher than predicted by the original model. This finding raised the question both of the validity of the model and of the effect of the lake level regulation regime on microbial production of methyl mercury (Ramsey 1988). When data collected from all reservoirs sampled in the past three years are considered together it is apparent that the model is essentially correct and that it is Sipiwesk Lake which is unusual. While methylation activity is not anomalously high in the flooded zone of Sipiwesk Lake, examination of the residuals of the regression on reservoir age (Figure 23) indicated that, while the measurement is not an outlier, it certainly is at the high end of the expected range. The high demethylation rate measured in Sipiwesk Lake is indeed an outlier, significantly higher than expected from the regression on reservoir age (Figure 24). The most reasonable explanation for the high rates of both methylation and demethylation in Sipiwesk Lake is the regulation regime. Unlike most reservoirs, where water levels are highest in summer, Sipiwesk Lake is often subjected to low levels in summer which can leave a substantial portion of the flooded zone exposed. During these periods, the drawdown zone is recolonized with grasses and other fast-growing vegetation which are made available to the methylating and demethylating bacteria when water levels rise again.

Acknowledgements

Eva Schindler performed the methylation and demethylation rate assays. Bob Hunt and Brian Hauser, both of the Freshwater Institute, performed the chemical analyses on sediment. R n D Aviation Ltd., Leaf Rapids, provided excellent service on the air charters. Special thanks to the numerous people at the Southern Indian Lake field station for their assistance with the field sampling.

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Appendix Table A1. Methyl Bay station 1 physical measurements, July 5 to August 22, 1988. > indicates the Secchi disk was visible on the bottom. Eh is oxidation-reduction potential.

Date	Secchi (m)	Sediment pH	Sediment Eh (mV)	Depth (m)	Temperature (^{o}C)	Dissolved Oxygen (mg L^{-1})	Specific Conductivity (uS cm^{-1})
July 5	1.2	5.80	255	0	16.2	9.2	80
				2.4	15.5	5.3	89
				mean	15.9	7.3	85
July 19	>2.0	6.10	285	0	17.4	9.1	83
				2.0	16.9	8.0	85
				mean	17.2	8.6	84
July 27	>1.5	5.80	300	0	17.7	9.7	82
				1.5	17.2	8.9	84
				mean	17.5	9.3	83
August 10	>2.0	7.10	300	0	17.8	8.9	114
				2.0	16.5	6.3	146
				mean	17.2	7.6	130
August 22	>1.75	6.80	320	0	19.0	8.6	82
				1.75	17.2	8.1	87
				mean	18.1	8.1	84

Appendix Table A2. Methyl Bay station 2 physical measurements, July 5 to August 22, 1988. > indicates the Secchi disk was visible on the bottom. Eh is oxidation-reduction potential.

Date	Secchi (m)	Sediment pH	Sediment Eh (mV)	Depth (m)	Temperature (^{o}C)	Dissolved Oxygen (mg L^{-1})	Specific Conductivity (uS cm^{-1})
July 5	1.1	6.00	265	0	16.4	8.9	80
				1.8	15.9	8.7	81
				mean	16.2	8.8	80
July 19	>1.5	6.40	260	0	17.4	8.8	83
				1.5	16.7	4.7	85
				mean	17.1	6.8	84
July 27	>1.3	5.80	300	0	17.7	8.8	83
				1.3	17.2	8.4	84
				mean	17.5	8.6	84
August 10	>1.1	6.90	325	0	17.6	8.8	106
				1.1	17.7	8.6	106
				mean	17.7	8.7	106
August 22	>1.4	6.80	290	0	18.5	7.3	83
				1.4	18.2	7.0	83
				mean	18.4	7.2	83

Appendix Table A3. Methyl Bay station 3 physical measurements, July 5 to August 22, 1988. > indicates the Secchi disk was visible on the bottom. Eh is oxidation-reduction potential.

Date	Secchi (m)	Sediment pH	Sediment Eh (mV)	Depth (m)	Temperature (°C)	Dissolved Oxygen (mg L^{-1})	Specific Conductivity (uS cm^{-1})
July 5	>1.4	6.20	240	0	16.5	8.8	79
				1.4	15.8	9.2	78
				mean	16.2	9.0	79
July 19	>1.5	6.60	310	0	17.7	8.9	83
				1.5	17.3	8.6	84
				mean	17.5	8.8	83
July 27	>1.5	6.10	348	0	17.6	8.6	84
				1.5	17.0	8.3	85
				mean	17.3	8.5	85
August 10	>1.1	7.00	325	0	17.6	8.8	79
				1.1	17.7	8.4	76
				mean	17.7	8.7	78
August 22	>1.4	6.80	270	0	18.8		81
				1.4	18.3		82
				mean	18.6		81

Appendix Table A4. Methyl Bay station 4 physical measurements, July 5 to August 22, 1988. > indicates the Secchi disk was visible on the bottom. Eh is oxidation-reduction potential.

Date	Secchi (m)	Sediment pH	Sediment Eh (mV)	Depth (m)	Temperature (°C)	Dissolved Oxygen (mg L^{-1})	Specific Conductivity (uS cm^{-1})
July 5	1.4	6.20	290	0	16.1	9.4	78
				2	15.9	9.4	79
				4	15.3	9.3	79
				6	13.7	9.2	82
				7	13.6	9.0	92
				mean	14.9	9.3	82
July 19	1.1	6.70	330	0	17.6	9.2	79
				2	17.4	9.2	80
				4	16.9	9.1	81
				6	15.4	8.9	84
				7	14.2	7.2	86
				mean	16.3	8.7	82
July 27	1.0	6.25	410	0	17.8	9.2	83
				2	17.5	9.2	83
				4	16.9	8.7	85
				6	16.6	8.4	85
				6.6	16.0	7.5	87
				mean	17.0	8.6	85
August 10	1.0	6.90	375	0	17.5	9.0	74
				2	17.7	8.8	74
				4	17.7	8.7	74
				6	17.4	8.7	77
				6.5	16.6	7.7	81
				mean	17.4	8.6	76
August 22	1.0	7.10	260	0	17.6		82
				2	17.6		82
				4	17.6		82
				6	17.6		82
				mean	17.6		82

Appendix Table A5. Granville Lake station 5 physical measurements, July 4 to August 24, 1988. > indicates the Secchi disk was visible on the bottom. Eh is oxidation-reduction potential.

Date	Secchi (m)	Sediment pH	Sediment Eh (mV)	Depth (m)	Temperature (^{o}C)	Dissolved Oxygen (mg L^{-1})	Specific Conductivity (uS cm^{-1})
July 4	>2.5	6.00	230	0	18.5	8.9	60
				2.5	17.8	8.8	61
				mean	18.2	8.9	61
July 17	>2.0	7.00	230	0	19.5	8.7	58
				2.0	18.2	9.2	63
				mean	18.9	9.0	60
July 25	>2.0	7.00	340	0	19.4		63
				2.0	18.6		62
				mean	19.0		62
August 8	>1.8	7.30	240	0	18.9	8.6	59
				1.8	18.3	8.6	59
				mean	18.6	8.6	59
August 24	>1.8	5.94	240	0	18.3	9.3	64
				1.8	18.3	8.7	64
				mean	18.3	9.0	64

Appendix Table A6. Granville Lake station 8 physical measurements, July 4 to August 24, 1988. > indicates the Secchi disk was visible on the bottom. Eh is oxidation-reduction potential.

Date	Secchi (m)	Sediment pH	Sediment Eh (mV)	Depth (m)	Temperature (°C)	Dissolved Oxygen (mg L^{-1})	Specific Conductivity (uS cm^{-1})
July 4	2.0	6.40	200	0	18.3	8.9	60
				2	18.1	8.8	61
				4	17.6	8.3	61
				6	17.4	6.7	62
				6.3	17.2	6.7	62
				mean	17.7	8.2	61
July 17	2.2	6.98	240	0	18.8	8.7	61
				2	18.8	8.5	61
				4	18.2	8.4	62
				6	18.1	8.2	62
				7.8	18.0	8.2	62
				mean	18.4	8.4	61
July 25	2.0	6.97	310	0	19.3		60
				2	19.3		61
				4	19.1		63
				6	19.0		65
				8	19.0		65
				10	19.0		66
				11.3	18.8		67
				mean	19.1		64
August 8	1.6	6.90	230	0	19.0	8.2	58
				2	19.0	8.1	58
				4	19.0	8.1	59
				6	18.8	7.9	60
				6.75	18.7	7.6	64
				mean	18.9	8.0	60
August 24	1.8	6.40	220	0	18.6	8.1	64
				2	18.6	8.1	64
				4	18.6	8.1	64
				6	18.6	8.1	67
				7.75	18.6	8.1	69
				mean	18.6	8.1	66

Appendix Table A7. Sokatisewin Lake station 9 physical measurements, July 4 to August 24, 1988. > indicates the Secchi disk was visible on the bottom. Eh is oxidation-reduction potential.

Date	Secchi (m)	Sediment pH	Sediment Eh (mV)	Depth (m)	Temperature (^{o}C)	Dissolved Oxygen ($mg\ L^{-1}$)	Specific Conductivity ($uS\ cm^{-1}$)
July 4	>4.5	6.40	130	0	19.9	8.6	71
				4.5	19.3	8.0	72
				mean	19.6	8.3	71
July 17	>3.0	6.80	160	0	19.4	8.9	76
				3.0	18.9	7.3	85
				mean	19.2	8.1	80
July 25	>2.5	6.80	150	0	19.6	8.0	80
				2.5	19.5	4.9	81
				mean	19.6	6.5	81
August 8	>2.0	6.70	190	0	18.6	8.1	82
				2	18.4	7.8	83
				mean	18.5	8.0	83
August 24	>2.5	6.30	140	0	18.8	8.5	82
				2.5	18.4	8.4	83
				mean	18.6	8.5	82

Appendix Table A8. Sokatisewin Lake station 12 physical measurements, July 4 to August 24, 1988. > indicates the Secchi disk was visible on the bottom. Eh is oxidation-reduction potential.

Date	Secchi (m)	Sediment pH	Sediment Eh (mV)	Depth (m)	Temperature (°C)	Dissolved Oxygen (mg L^{-1})	Specific Conductivity (uS cm^{-1})
July 4	2.2	6.90	75	0	19.7	8.9	69
				1	19.7	8.8	72
				2	19.6	8.8	72
				3	19.6	8.8	72
				4	19.5	8.9	73
				6	18.9	8.4	74
				8	17.6	6.9	76
				9	15.6	5.0	78
				10	12.4	2.8	77
				10.75	9.3	0.8	83
				mean	17.5	7.1	75
July 17	2.5	6.70	80	0	19.1	9.2	76
				2	18.7	9.1	77
				4	18.7	8.9	77
				6	18.5	8.8	81
				8	18.3	8.3	82
				10	16.4	5.4	82
				11	11.5	1.5	96
				mean	17.3	7.3	82
July 25	1.5	6.70	-50	0	19.6	7.8	80
				2	19.6	7.2	80
				4	19.3	6.8	81
				6	19.1	6.0	81
				8	18.5	5.2	83
				10	16.7	3.4	82
				11.5	12.6	1.8	106
				mean	17.9	5.5	85
August 8	2.0	6.78	80	0	18.6	8.2	83
				2	18.5	8.1	84
				4	18.5	8.2	84
				6	18.4	8.3	85
				8	18.4	8.1	85
				10	18.3	8.0	86
				11.5	13.6	1.2	102
				mean	17.8	7.2	87
August 24	2.0	6.40	70	0	18.6	8.6	81
				2	18.6	8.6	82
				4	18.6	8.4	82
				6	18.6	8.4	82
				8	18.6	8.1	82
				10	18.4	7.2	85
				11	15.1	1.0	124
				mean	18.1	7.2	89

Appendix Table B1. Concentrations (mg (g dry sediment)$^{-1}$) of total carbon (C), carbonate-carbon (CO_3), organic carbon, and total nitrogen (N) in surficial sediments from Methyl Bay, Southern Indian Lake, Manitoba; July 5 through August 22, 1988.

Station	Date	Total C	CO_3	Organic C	Total N
1	July 5	121	10	111	5.6
	July 19	133	10	123	6.0
	July 27	134	13	121	6.1
	August 10	134	11	123	7.5
	August 22	111	8	103	5.8
2	July 5	198	13	185	7.8
	July 19	110	9	101	5.9
	July 27	212	14	198	8.8
	August 10	204	13	191	8.4
	August 22	204	14	190	8.2
3	July 5	122	12	110	5.6
	July 19	162	13	149	6.9
	July 27	163	15	148	6.5
	August 10	152	10	142	6.0
	August 22	137	11	126	6.6
4	July 5	19	3	16	2.0
	July 19	17	3	14	1.9
	July 27	20	3	17	2.1
	August 10	23	4	19	2.2
	August 22	21	3	18	2.2

Appendix Table B2. Concentrations (mg (g dry sediment)$^{-1}$) of total carbon (C), carbonate-carbon (CO_3), organic carbon, and total nitrogen (N) in surficial sediments from Granville Lake, Manitoba; July 4 through August 24, 1988.

Station	Date	Total C	CO_3	Organic C	Total N
5	July 4	33	4	29	4.0
	July 17	28	4	24	3.2
	July 25	16	2	14	1.9
	August 8	23	2	21	2.8
	August 24	36	4	32	4.6
6	July 4	27	4	23	3.3
	July 17	26	3	23	3.0
	July 25	21	3	18	2.6
	August 8	16	2	14	1.9
	August 24	24	3	21	2.8
7	July 4	33	4	29	4.0
	July 17	23	3	20	2.8
	July 25	20	3	17	2.4
	August 8	22	2	20	2.7
	August 24	31	4	27	3.7
8	July 4	21	3	18	2.7
	July 17	29	4	25	3.6
	July 25	20	2	18	2.6
	August 8	26	3	23	3.2
	August 24	25	3	22	3.0

Appendix Table B3. Concentrations (mg (g dry sediment)$^{-1}$) of total carbon (C), carbonate-carbon (CO_3), organic carbon, and total nitrogen (N) in surficial sediments from Sokatisewin Lake, Saskatchewan; July 4 through August 24, 1988.

Station	Date	Total C	CO_3	Organic C	Total N
9	July 4	71	6	65	5.3
	July 17	81	5	76	6.2
	July 25	83	5	78	5.6
	August 8	99	6	93	5.8
	August 24	75	5	70	5.1
10	July 4	62	5	57	4.6
	July 17	96	6	90	6.0
	July 25	83	5	78	5.8
	August 8	100	6	94	6.1
	August 24	92	6	86	5.3
11	July 4	63	5	58	4.9
	July 17	99	7	92	5.9
	July 25	84	5	79	6.1
	August 8	86	6	80	5.7
	August 24	91	6	85	5.4
12	July 4	37	4	33	4.6
	July 17	37	3	34	4.6
	July 25	37	4	33	4.7
	August 8	38	4	34	4.6
	August 24	41	4	37	4.6

Appendix Table C1. Concentrations of total mercury in surficial sediments from Methyl Bay, Southern Indian Lake, Manitoba; July 5 through August 22, 1988. Values A and B represent concentrations in duplicate subsamples.

		ng Hg (g dry sediment)$^{-1}$			
		Range		Station	Nearshore
Station	Date	A	B	Mean	Mean
1	July 5	294	303	298	324
	July 19	339	385	362	380
	July 27	458	439	448	419
	August 10	555	580	567	439
	August 22	312	265	289	351
2	July 5	419	437	428	
	July 19	436	350	393	
	July 27	542	538	540	
	August 10	359	342	350	
	August 22	408	479	443	
3	July 5	265	229	247	
	July 19	413	357	385	
	July 27	293	245	269	
	August 10	409	393	401	
	August 22	326	317	322	
4	July 5	114	150	132	
	July 19	244	289	267	
	July 27	138	142	140	
	August 10	197	200	198	
	August 22	99	132	116	

Appendix Table C2. Concentrations of total mercury in surficial sediments from Granville Lake, Manitoba; July 4 through August 24, 1988. Values A and B represent concentrations in duplicate subsamples.

		ng Hg (g dry sediment)$^{-1}$			
		Range		Station	Nearshore
Station	Date	A	B	Mean	Mean
5	July 4	377	432	404	251
	July 17	308	208	258	193
	July 25	105	54	79	75
	August 8	106	102	104	91
	August 24	106	101	103	112
6	July 4	160	146	153	
	July 17	162	120	141	
	July 25	79	70	75	
	August 8	81	55	68	
	August 24	111	121	116	
7	July 4	178	211	195	
	July 17	206	152	179	
	July 25	80	64	72	
	August 8	93	108	101	
	August 24	121	111	116	
8	July 4	306	338	322	
	July 17	211	238	225	
	July 25	77	85	81	
	August 8	68	84	76	
	August 24	90	90	90	

Appendix Table C3. Concentrations of total mercury in surficial sediments from Sokatisewin Lake, Saskatchewan; July 4 through August 24, 1988. Values A and B represent concentrations in duplicate subsamples.

		ng Hg (g dry sediment)$^{-1}$			
		Range		Station	Nearshore
Station	Date	A	B	Mean	Mean
9	July 4	253	223	238	229
	July 17	302	246	274	198
	July 25	130	157	144	147
	August 8	216	217	217	199
	August 24	168	215	192	189
10	July 4	156	183	169	
	July 17	136	126	131	
	July 25	147	153	150	
	August 8	203	167	185	
	August 24	222	230	226	
11	July 4	267	290	279	
	July 17	150	225	188	
	July 25	153	142	148	
	August 8	198	189	194	
	August 24	156	139	148	
12	July 4	208	151	180	
	July 17	209	207	208	
	July 25	126	199	162	
	August 8	178	164	171	
	August 24	198	49	123	

Appendix Table D1. Specific rates of methylation and demethylation of mercury and corresponding M/D ratios and rates of microbial activity (um DIC g dry sed^{-1} h^{-1}) measured in samples of surficial sediment from Methyl Bay, Southern Indian Lake, Manitoba; July 5 to August 22, 1988.

Station	Date	% added Hg g^{-1} h^{-1} Methylation	Demethylation	M/D	uM DIC g^{-1} h^{-1}
1	July 5	0.0015	0.0265	0.0566	3.82
	July 19	0.0012	0.2676	0.0045	59.97
	July 27	0.0046	0.0702	0.0655	32.50
	August 10	0.0050	0.5043	0.0099	26.54
	August 22	0.0034	0.2520	0.0135	24.80
2	July 5	0.0039	0.0109	0.3578	0
	July 19	0.0148	0.5284	0.0280	35.87
	July 27	0.0004	0.2993	0.0013	37.09
	August 10	0.0047	0.2521	0.0186	22.72
	August 22	0.0022	0.5769	0.0038	28.01
3	July 5	0.0008	0.0788	0.0102	9.21
	July 19	0.0023	0.1149	0.0200	48.79
	July 27	0.0019	0.0308	0.0617	25.32
	August 10	0.0111	0.1265	0.0877	11.94
	August 22	<0.0001	0.1811	<0.0006	22.20
4	July 5	0.0010	0.0096	0.1042	3.58
	July 19	0.0027	0.1067	0.0253	16.89
	July 27	0.0005	0.1684	0.0030	11.25
	August 10	0.0002	0.5163	0.0004	9.02
	August 22	0.0001	0.0295	0.0034	1.49

Appendix Table D2. Specific rates of biological methylation and demethylation of mercury and corresponding M/D ratios, in response to the addition of 30 mM bromoethane sulfonic acid, measured in samples of surficial sediment from Methyl Bay, Southern Indian Lake, Manitoba; July 5 to August 22, 1988.

Station	Date	% added Hg g^{-1} h^{-1} Methylation	Demethylation	M/D
1	July 5	<0.0001	0.0081	<0.0123
	July 19	0.0026	0.1218	0.0213
	July 27	0.0010	0.0891	0.0112
	August 10	0.0042	0.3369	0.0125
	August 22	0.0015	0.1478	0.0101
2	July 5	<0.0001	0.1243	<0.0008
	July 19	0.0060	0.1606	0.0374
	July 27	0.0012	0.0951	0.0126
	August 10	0.0293	0.1564	0.1873
	August 22	0.0005	0.3416	0.0015
3	July 5	<0.0001	0.0744	<0.0013
	July 19	0.0016	0.0591	0.0271
	July 27	0.0022	0.1808	0.0122
	August 10	0.0226	0.1492	0.1515
	August 22	0.0002	0.2648	0.0008
4	July 5	<0.0001	0.0165	<0.0061
	July 19	<0.0001	0.0168	<0.0060
	July 27	0.0001	0.1063	0.0009
	August 10	0.0003	0.0253	0.0119
	August 22	0.0002	0.0237	0.0084

Appendix Table D3. Specific rates of biological methylation and demethylation of mercury and corresponding M/D ratios, in response to the addition of 20 mM sodium molybdate, measured in samples of surficial sediment from Methyl Bay, Southern Indian Lake, Manitoba; July 5 to August 22, 1988.

Station	Date	% added Hg g^{-1} h^{-1} Methylation	Demethylation	M/D
1	July 5	0.0004	0.0394	0.0102
	July 19	0.0008	0.0748	0.0107
	July 27	0.0002	0.2709	0.0007
	August 10	0.0007	0.2447	0.0029
	August 22	0.0006	0.0423	0.0142
2	July 5	<0.0001	0.0918	<0.0011
	July 19	0.0021	0.0505	0.0416
	July 27	0.0008	0.1934	0.0041
	August 10	0.0050	0.0280	0.1786
	August 22	0.0019	0.4961	0.0038
3	July 5	0.0003	0.0287	0.0105
	July 19	0.0075	0.1279	0.0586
	July 27	0.0010	0.1773	0.0056
	August 10	0.0041	0.0050	0.8200
	August 22	0.0003	0.2143	0.0014
4	July 5	0.0006	0.0040	0.1500
	July 19	0.0008	0.0184	0.0435
	July 27	0.0007	0.0638	0.0110
	August 10	0.0001	0.1183	0.0008
	August 22	0.0002	0.0180	0.0111

Appendix Table D4. Specific rates methylation and demethylation of mercury and corresponding M/D ratios and rates of microbial activity (uM DIC g dry sed^{-1} h^{-1}) measured in samples of surficial sediment from Granville Lake, Manitoba; July 4 to August 24, 1988.

Station	Date	% added Hg g^{-1} h^{-1} Methylation	Demethylation	M/D	uM DIC g^{-1} h^{-1}
5	July 4	0.0069	0.3941	0.0175	17.39
	July 17	0.0004	0.0311	0.0129	8.32
	July 25	0.0001	0.0191	0.0052	10.49
	August 8	<0.0001	0.0235	<0.0043	3.55
	August 24	0.0002	0.0937	0.0021	12.23
6	July 4	0.0001	0.0043	0.0233	12.12
	July 17	0.0040	0.0744	0.0538	3.54
	July 25	0.0003	0.0334	0.0090	20.74
	August 8	<0.0001	0.0171	<0.0058	4.94
	August 24	0.0002	0.0846	0.0024	7.49
7	July 4	0.0024	0.0252	0.0952	8.64
	July 17	<0.0001	0.0571	<0.0018	20.62
	July 25	0.0003	0.0411	0.0073	7.91
	August 8	0.0009	0.0381	0.0236	13.66
	August 24	0.0003	0.1080	0.0028	5.90
8	July 4	0.0024	0.0435	0.0552	16.09
	July 17	0.0005	0.0483	0.0104	5.63
	July 25	0.0001	0.0569	0.0018	16.39
	August 8	0.0013	0.0137	0.0949	1.14
	August 24	0.0006	0.0411	0.0146	1.62

Appendix Table D5. Specific rates of biological methylation and demethylation of mercury and corresponding M/D ratios, in response to the addition of 30 mM bromoethane sulfonic acid, measured in samples of surficial sediment from Granville Lake, Manitoba; July 4 to August 24, 1988.

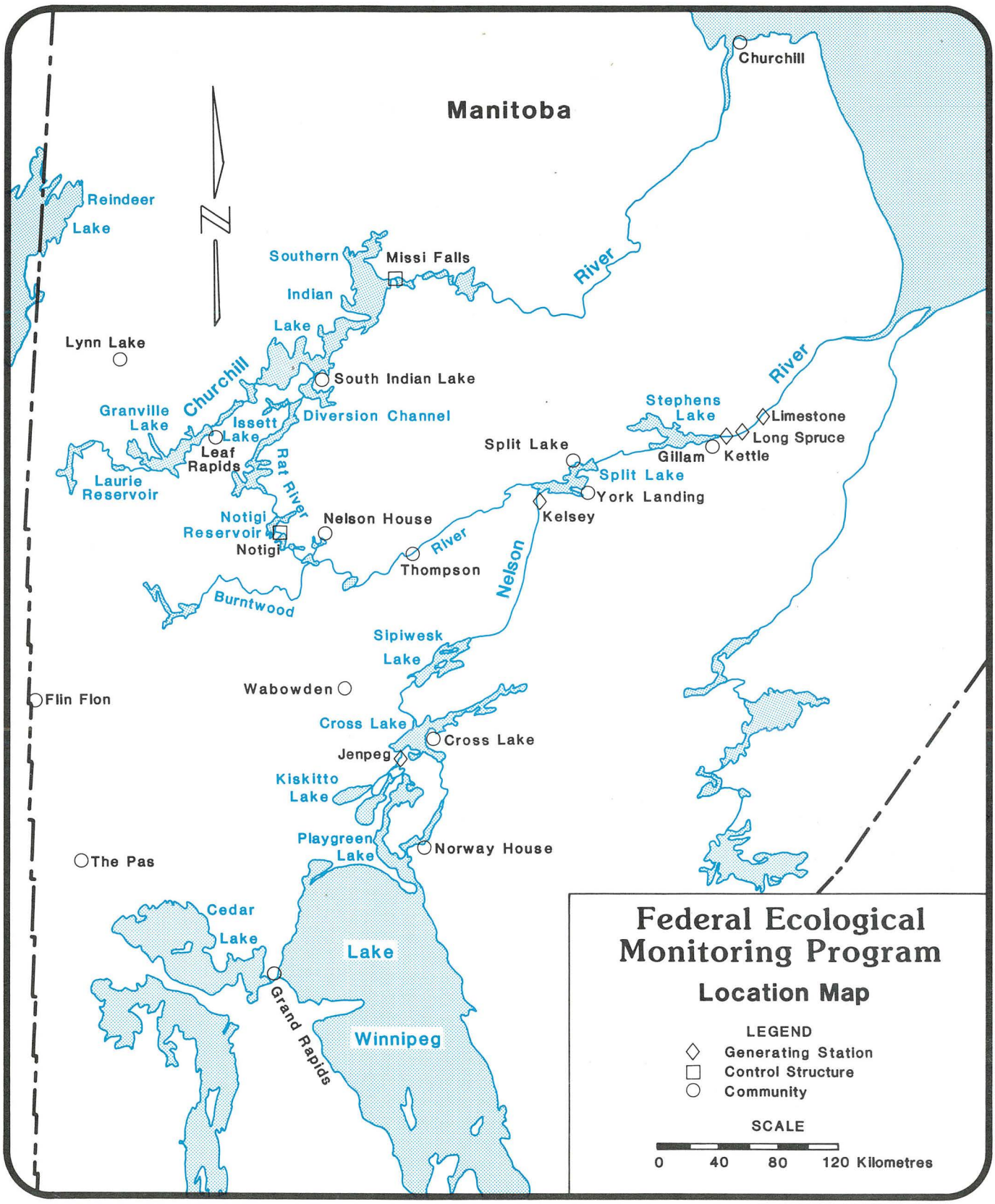
Station	Date	% added Hg g^{-1} h^{-1} Methylation	Demethylation	M/D
5	July 4	0.0085	0.3101	0.0274
	July 17	0.0012	0.0905	0.0133
	July 25	0.0001	0.0257	0.0039
	August 8	<0.0001	0.0151	<0.0066
	August 24	0.0002	0.1213	0.0016
6	July 4	0.0005	0.1193	0.0042
	July 17	0.0005	0.0573	0.0087
	July 25	0.0001	0.0178	0.0056
	August 8	<0.0001	0.0279	<0.0036
	August 24	0.0001	0.0450	0.0022
7	July 4	0.0020	0.2610	0.0077
	July 17	0.0014	0.0071	0.1972
	July 25	0.0005	0.0252	0.0198
	August 8	0.0052	0.0464	0.1121
	August 24	<0.0001	0.0767	<0.0013
8	July 4	0.0010	0.0396	0.0253
	July 17	0.0003	0.0501	0.0060
	July 25	0.0002	0.0847	0.0024
	August 8	<0.0001	0.0026	<0.0385
	August 24	<0.0001	0.0378	<0.0026

Appendix Table D6. Specific rates of biological methylation and demethylation of mercury and corresponding M/D ratios, in response to the addition of 20 mM sodium molybdate, measured in samples of surficial sediment from Granville Lake, Manitoba; July 4 to August 24, 1988.

Station	Date	% added Hg g^{-1} h^{-1} Methylation	Demethylation	M/D
5	July 4	0.0024	0.4272	0.0056
	July 17	0.0002	0.0610	0.0032
	July 25	0.0001	0.0048	0.0208
	August 8	<0.0001	0.0119	<0.0084
	August 24	0.0003	0.0338	0.0089
6	July 4	0.0007	0.1403	0.0050
	July 17	0.0039	0.0760	0.0513
	July 25	0.0002	0.0183	0.0109
	August 8	0.0002	0.0542	0.0037
	August 24	0.0001	0.0977	0.0010
7	July 4	0.0012	0.5132	0.0023
	July 17	<0.0001	0.0281	<0.0036
	July 25	0.0003	0.0462	0.0065
	August 8	0.0024	0.0271	0.0886
	August 24	0.0001	0.0539	0.0019
8	July 4	0.0021	0.0402	0.0522
	July 17	0.0004	0.0011	0.3636
	July 25	0.0003	0.0478	0.0063
	August 8	0.0004	0.0157	0.0255
	August 24	<0.0001	0.1515	<0.0007

Appendix Table D7. Specific rates of microbial activity and of methylation and demethylation of mercury and corresponding M/D ratios and rates of microbial activity (uM DIC g dry sed^{-1} h^{-1}) measured in samples of surficial sediment from Sokatisewin Lake, Saskatchewan; July 4 to August 24, 1988.

Station	Date	% added Hg g^{-1} h^{-1} Methylation	Demethylation	M/D	uM DIC g^{-1} h^{-1}
9	July 4	0.0068	0.0911	0.0746	0
	July 17	0.0014	0.0345	0.0406	4.69
	July 25	0.0023	0.0169	0.1361	2.14
	August 8	0.0009	0.0869	0.0104	3.48
	August 24	0.0043	0.0453	0.0949	1.35
10	July 4	0.0058	0.0425	0.1365	8.79
	July 17	0.0017	0.0034	0.5000	2.02
	July 25	0.0019	0.0711	0.0267	0.38
	August 8	0.0018	0.0525	0.0343	5.06
	August 24	0.0022	0.0273	0.0806	0.86
11	July 4	0.0054	0.0088	0.6136	5.21
	July 17	0.0024	0.0431	0.0557	1.82
	July 25	0.0016	0.0705	0.0227	2.67
	August 8	0.0015	0.0172	0.0872	1.44
	August 24	0.0012	0.0218	0.0550	2.62
12	July 4	0.0267	0.0006	44.5000	0
	July 17	0.0024	0.0196	0.1224	0.43
	July 25	0.0034	0.0148	0.2297	0
	August 8	0.0037	0.0096	0.3854	0.13
	August 24	0.0070	0.0087	0.8046	1.44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Rat River
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Notigi
Reservoir
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Nelson House
Thompson
River
Burntwood
Nelson
River
Stephens
Lake
Limestone
Long Spruce
Gillam
Kettle
Split Lake
Split Lake
York Landing
Kelsey
Sipiwesk
Lake
Wabowden
Flin Flon
Cross Lake
Cross Lake
Jenpeg
Kiskitto
Lake
Playgreen
Lake
Norway House
The Pas
Cedar
Lake
Lake
Winnipeg
Grand Rapids
Federal Ecological
Monitoring Program
Location Map
LEGEND
Generating Station
Control Structure
Community
SCALE
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120 Kilometres