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Methane Production in Minnesota Peatlands

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Rates of methane production in Minnesota peats were studied. Surface (10- to 25-cm) peats produced an average of 228 nmol of CH_4 per g (dry weight) per h at 25°C and ambient pH. Methanogenesis rates generally decreased with depth in ombrotrophic peats, but on occasion were observed to rise within deeper layers of certain fen peats. Methane production was temperature dependent, increasing with increasing temperature (4 to 30°C), except in peats from deeper layers. Maximal methanogenesis from these deeper regions occurred at 12°C. Methane production rates were also pH dependent. Two peats with pHs of 3.8 and 4.3 had an optimum rate of methane production at pH 6.0. The addition to peat of glucose and H_2-CO_2 stimulated methanogenesis, whereas the addition of acetate inhibited methanogenesis. Cysteine-sulfide, nitrogen-phosphorus-trace metals, and vitamins-yeast extract affected methane production very little. Various gases were found to be trapped or dissolved (or both) within peatland waters. Dissolved methane increased linearly to a depth of 210 cm. The accumulation of metabolic end products produced within peat bogs appears to be an important mechanism limiting carbon turnover in peatland environments.

Peatlands are unbalanced, wetland ecosystems where productivity exceeds biodegradation. This imbalance leads to the accumulation of organic deposits that are termed peat. Except for the surface water microlayer, peats are anaerobic environments (15). As such, they represent suitable habitats for methanogenic bacteria. In previous work we have detected high populations of methanogens (10^6/ml of interstitial water) in the 25- to 40-cm depth region of a Minnesota peatland (16). Even at a depth of 210 cm, the deepest region examined, significant populations (10^4/ml) of methanogens were observed.

The methanogenic food chain is a microbial system that mediates the biodegradation of organic compounds in many anaerobic environments. The flow of carbon through this chain avoids a buildup of inhibitory metabolic end products, thereby preventing the cessation of biodegradation (19). Since methanogenesis is the terminal step in this anaerobic food chain, any perturbation of the chain should be reflected by altered methane production. Consequently, methanogenesis is a key process to study, reflecting the combined activities of many different microbial groups.

Peat bog ecosystems produce methane (19); however, we are aware of no reports where methane production from peatlands has been quantified. The influences of various physiochemical parameters upon peat methanogenesis likewise have not been reported. Temperature is known to exert a significant influence on methanogenesis (and the number of methanogens) in lake sediments (20). In culture, methanogenic bacteria metabolize best in the pH range of 6.7 to 8.0. However, very low rates of methanogenesis have been observed at pH 5.8 (19). Methanogens utilize sulfide or cysteine to satisfy their sulfur requirements; however, high sulfide concentrations have been shown to inhibit methanogenesis in sediments (7, 8, 14). Phosphate (13, 14) also has been found to inhibit methanogenesis in lake sediments, as has nitrate (1) in salt marsh sediments. Ammonium ion may slow the conversion of acetic acid to methane at high concentrations (19). Methanogenic substrates such as hydrogen (12, 17) and acetate (3, 17) often stimulate methanogenesis in lake sediments. Glucose stimulated methanogenesis in some habitats (2, 17); however, at glucose concentrations greater than 1.0%, suppression of methane production occurred. Vitamins (4) and yeast extract (19) also have been observed to stimulate methanogenesis. Finally, sulfate has been found to inhibit methanogenesis in sediments (18).

Here we report results of experiments designed to quantitate the rates of methanogenesis in Minnesota peats. We also have examined the influence of various physiochemical parameters on peatland methanogenesis rates.

MATERIALS AND METHODS

Study sites. Peat for methanogenesis experiments was collected at a transitional fen site within the Cedar Creek Natural History Area (Bethel, Minn.). This site is dominated by a *Larix laricina* and *Picea mariana* overstory with a *Sphagnum magellanicum* and *Sphagnum flexuosum* groundcover. Surface water pH was generally between 3.8 and 4.0. The pH increased with peat depth as follows: 25 cm, 4.1; 40 cm, 4.2; 60 cm, 4.3; 90 cm, 4.4; 120 cm, 4.4; 210 cm, 4.8. In addition to the Cedar Creek site, trapped gases also were collected from several bog and fen sites within the Red Lake peatland of north central Minnesota.

Gas chromatography. A Varian series 1400 gas chromatograph equipped with a flame ionization detector and Porapak QS column (100/120 mesh, 1.8 m by 2 mm, glass) was used for the quantification of methane. The injector, column, and detector temperatures used were 180, 130, and 180°C, respectively. A Varian 920 gas chromatograph equipped with a thermal conductivity detector was used to assay the following gases: oxygen and nitrogen (molecular sieve 5A column; 45/60 mesh; 2 m by 6.4 mm; stainless steel; 130, 70, and 130°C) and carbon dioxide (Chromasorb 106 column; 80/100 mesh; 3.6 m by 6.4 mm; stainless steel; 130, 70, and 130°C).

Methane production. Peat for methanogenesis experiments was collected in 125-ml wide-mouthed glass jars. These jars were equilibrated in an anaerobic chamber (Coy Manufacturing Co.; containing 90% N_2–10% H_2 with less than 5 ppm of O_2) and sealed with rubber stoppers before use in the field. A core sample from the desired depth was collected and extruded onto a metal tray. This sample was sealed

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TABLE 1. Methane production from Cedar Creek peat samples

Depth sampled (cm)	Rate of methane production[a] (nmol of CH_4 per g [dry wt] per h)					
	Untreated	r^b	Heat-killed control	r	Formalin-killed control	r
10	219.6	0.997	0.3	0.937	0.2	0.966
25	213.2	0.993	0.1	0.975	0.1	0.961
40	200.7	0.996	0.2	0.962	0.2	0.949
90	40.8	0.998	0.1	0.934	0.1	0.938
120	6.2	0.992	0.2	0.938	0.1	0.932
210	2.0	0.994	0.3	0.966	0.2	0.925

[a] Incubated at 25°C for 100 h.
[b] r, Correlation coefficient for linear methane production rate with time.

immediately in a collection jar. Jars containing peat were placed inside a plastic bag, which then was filled with nitrogen and sealed. Samples were returned to the lab at ambient temperature.

Within 2.5 h of collection, ca. 0.5-g subsamples were distributed to tubes equilibrated under anerobic conditions inside the anaerobic chamber. Nutrients to be added to peat samples were dissolved in deaerated, distilled water such that 3 to 4 ml of solution was added to all peat samples (depending upon the particular experiment). This water generally was absorbed completely by the peat. After appropriate additions, tubes were sealed, vortexed under vacuum, filled with nitrogen (three cycles), filled with 1 atm (ca. 101.3 kPa) of high-purity nitrogen, incubated under specified conditions, and monitored for methane production. Experiments were run for 48 to 135 h, with methane analyses performed at 4- to 18-h intervals. Control tubes were run for all experiments. These were either heat killed (95°C, 0.75 h, with occasional vortexing) or preserved with Formalin (37% Formalin, added to make up 5% of final volume).

Nutrient additions. Phosphorus was added as $NaH_2PO_4 \cdot H_2O$, and nitrogen was added as NH_4Cl. The vitamin solution used (final concentration, 1.0% [vol/vol]) contained the following (in milligrams per liter of distilled water): biotin, 2; folic acid, 2; pyridoxine hydrochloride, 10; thiamine hydrochloride, 5; riboflavin, 5; vitamin B_{12}, 0.1; *p*-aminobenzoic acid, 5; and lipoic acid, 5. Cysteine and sulfide were added in combination as a reducing agent (R. T. Williams, Ph.D. thesis, University of Minnesota, Minneapolis, 1982). The trace metals solution contained the following (in grams per liter of distilled water): nitrilotriacetic acid, 1.00; $MgSO_4 \cdot H_2O$, 2.0; $FeSO_4 \cdot 7H_2O$, 0.12; $MnSO_4 \cdot 7H_2O$, 0.03; $ZnSO_4 \cdot 7H_2O$, 0.03; $CoSO_4$, 0.01. Adjustments of pH were made by using deaerated 0.1 N NaOH or HCl. An 80% H_2–20% CO_2 (both gases were ultra high purity grade) mixture was added by using a manifold permitting gas exchange while maintaining anaerobic conditions.

Miscellaneous methods. After completion of an experiment, the contents of all tubes were emptied into preweighed beakers. Peat samples were dried at 95°C to a constant weight. These dry weight measurements were used to standardize methane production from each tube on a per gram (dry weight) of peat basis.

Gas trapped in peatland habitats was collected with Vacutainer tubes (American Scientific Products) and a large glass funnel with a shortened stem. A metal rod was used to release gas from the peat beneath a water-filled funnel. Gas-containing tubes were placed on ice. Collected gases were analyzed by gas chromatography within 5 h of their collection from Cedar Creek sites. Samples from northern Minnesota were kept at 4°C until analyzed (usually within 24 to 96 h).

Methane dissolved in peatland interstitial water also was collected with Vacutainer tubes. A 5-ml water sample was squeezed out of a peat sample and into a 10-ml syringe barrel. The needle on this syringe was embedded in a Vacutainer tube stopper. After the 5 ml was in the syringe, the needle was pushed through the stopper and into the evacuated tube. Water samples were quickly sucked into these tubes, and their dissolved gases were stripped by the vacuum (11). These tubes were stored on ice, and the headspace gas was analyzed by gas chromatography within 5 h of sampling.

Data analysis. All methanogenesis experiments were performed with at least duplicate cultures plus duplicate controls. The number of points used to calculate the slope (rate) and correlation coefficient (r) was between 8 and 16, depending upon the particular experiment.

RESULTS

The results from a typical peat methanogenesis experiment are shown in Table 1. The reported depths are the distances below the water table surface from which the peat sample was collected. At the time of sampling, the water table was located ca. 10 cm below the upper surface of the vegetational ground cover (primarily sphagnum). The highest methane production rates occurred in surface peats; rates then decreased with depth. Heat- and Formalin-killed controls evolved very little methane, indicating that both control techniques were effective and that methane arose from new synthesis rather than from the release of trapped gas. The handling procedures used in these experiments effectively removed most previously produced methane that had been trapped within the peat matrix.

We also evaluated the influence of temperature on methane production from peat. Four temperatures (30, 25, 12, and 4°C) were used for each of six peat samples. The results (Table 2) showed a steady decline in methanogenesis with decline in temperature, except for samples taken from the two deepest sampling points. At these two depths maximal methane production occurred at 12°C. At 210 cm an increase in methane production was observed. This increase accompanied a shift in peat type from fibrous sphagnum peat to fine-grained sedge peat.

Methanogens are thought to metabolize best at neutral pH. Therefore, it is of interest to determine how the acidic nature of ombrotrophic peatlands affects methanogenesis. Two peat samples (10 cm [pH 3.9] and 60 cm [pH 4.3]) were collected and distributed to tubes, and pairs of tubes were adjusted to pH values between 2.2 and 7.0. Methane evolution rates at the various pHs are reported in Table 3.

TABLE 2. Effect of temperature on methane production from Cedar Creek peat samples

Depth sampled (cm)	Rate of methane production[a] (nmol of CH_4 per g [dry wt] per h)									
	Heat-killed control[b]	r[c]	30°C	r	25°C	r	12°C	r	4°C	r
10	0.2	0.942	324.1	0.990	204.6	0.996	103.1	0.986	72.2	0.992
30	0.3	0.963	308.6	0.997	255.5	0.984	97.9	0.983	80.9	0.983
40	0.2	0.971	107.3	0.997	100.2	0.996	54.8	0.975	54.7	0.969
90	0.1	0.946	110.4	0.999	60.8	0.991	43.7	0.975	23.6	0.984
120	0.1	0.958	8.1	0.989	10.3	0.998	15.3	0.978	6.5	0.969
210	0.2	0.949	45.7	0.990	54.3	0.980	75.4	0.985	40.4	0.979

[a] Incubated for 135 h.
[b] Incubated at 30°C.
[c] r, Correlation coefficient for linear methane production rate with time.

Methanogenesis was markedly inhibited at pH 2.2. The rate increased with increasing pH to a maximum at pH 6.0 and then fell at pH 7.0 to a level below that detected at pH 4.0 (approximately the natural pH). Final pHs determined at the end of the experiment for 10-cm peat were 2.3, 3.2, 3.8, 4.9, 5.8, and 6.6. For 60-cm peat the final values were 2.3, 3.2, 3.9, 4.9, 5.7, and 6.7.

The influence of various additives upon methanogenesis was investigated in another experiment. The results of this experiment (run at 25°C and at the natural pH) are shown in Table 4. Glucose and H_2-CO_2 stimulated methanogenesis. This stimulation was most dramatic in near-surface peats. In 120- and 210-cm peats neither glucose nor H_2-CO_2 (nor any other additive) significantly affected methanogenesis. Acetate was the only additive that consistently inhibited methanogenesis, although a mixture of nitrogen, phosphorus, and trace metals slowed methanogenesis in 10-cm peat. Reduced sulfur compounds stimulated methanogenesis in 25- and 40-cm peats, and a mixture of vitamins and yeast extract stimulated methanogenesis in 10-cm peats.

After a period of time (usually 6 to 7 days), the rate of methane production within the peat-containing tubes always declined. The closed environment occurring within experimental tubes is analogous to that found in situ. This situation arises from the low permeability and gel-like characteristics of the peat matrix. Therefore, processes inhibiting further methane production within these tubes could be similar to those that limit degradation of organic matter in peatlands. In an effort to understand the cause of this decline in methanogenesis, the following experiment was performed. Forty-eight tubes, each containing peat from the same sample, were incubated at 25°C. The methane evolution rate of each tube was determined betweeen 0 and 65 h and again between 256 and 294 h. Methanogenesis declined dramatically in all cases (Table 5). After the final CH_4 sampling, triplicate tubes were subjected to one of the following treatments: addition of Formalin, addition of 3 ml of distilled water, addition of 3 ml of 0.1% glucose, addition of 3 ml of 10 mM nitrogen–10 mM phosphorus–10% trace metals, addition of 3 ml of 0.1% yeast extract–1% vitamins, pH adjustment from 4.2 to 6.2, and addition of 1 atm of H_2-CO_2. Each solution was filter sterilized, deaerated, and adjusted to ambient pH before use. For each of these treatments a complementary triplicate set of tubes received the same treatment, but also was subjected to three cycles of vortexing under vacuum followed by replacement of the headspace gas with nitrogen.

In those tubes not receiving the vortex-vacuum-nitrogen cycles for headspace replacement, only glucose addition and pH adjustment caused a recovery in methane evolution rates. In fact, removal of the headspace gas and volatile compounds alone was sufficient to stimulate methanogenesis. It also greatly enhanced the stimulatory effects of glucose addition and pH adjustment. Also, after headspace replacement dilution with distilled water stimulated methanogenesis, as did addition of nitrogen-phosphorus-trace metals solution. The latter stimulation, however, was not significantly greater than that observed by dilution with an equal quantity of distilled water. However, under similar circumstances the yeast extract-vitamin mixture and H_2-CO_2 addition did significantly enhance methanogenesis.

Killed controls not subjected to the vacuum-vortex-nitrogen cycles produced methane at a rate ca. 2.5 times that of the control where methane was removed. This indicates that a significant amount of methane was trapped in the peat matrix, and that the flushing procedure used removed most of this methane.

Undissolved gas trapped by the peat-water matrix in

TABLE 3. Effect of pH on methane production from Cedar Creek peat samples

pH	Rate of methane production[a] (nmol of CH_4 per g [dry wt] per h)							
	10-cm peat				60-cm peat			
	Formalin-killed control	r[b]	Test	r	Formalin-killed control	r	Test	r
2.2	0.2	0.981	37.6	0.987	0.1	0.977	0.2	0.935
3.0	0.3	0.938	217.4	0.989	0.1	0.919	12.3	0.957
4.0	0.2	0.946	291.8	0.986	0.1	0.899	17.1	0.991
5.0	0.4	0.919	305.1	0.999	0.1	0.886	26.2	0.993
6.0	0.1	0.907	331.7	0.999	0.1	0.906	26.6	0.998
7.0	0.1	0.981	290.3	0.984	0.1	0.952	14.9	0.996

[a] Incubated for 48 h at 25°C.
[b] r, Correlation coefficient for linear methane production rate with time.

Cedar Creek bog was collected, and its methane content was analyzed. The mean methane content from samples collected at 10 different sampling points was 10.4 ± 2.8 μmol of CH_4 per ml of gas. These values are very similar to those found when peatland samples collected in northern Minnesota were analyzed. These latter values, as well as values for the N_2, CO_2, and O_2 content of bog gases, are reported in Table 6. Methane comprised an average of 31.2 ± 3.2 mol% of the gas collected.

The amount of methane dissolved in peatland water was determined for the Cedar Creek bog site. Water samples of 5 ml were collected in Vacutainer tubes. Gases were stripped from the water by the tubes' vacuum and held in the headspace for sampling (11). The results of this experiment are reported in Fig. 1. The dissolved methane content increased linearly with depth within the region sampled.

DISCUSSION

Methane production rates of peat samples decreased as the depth from which the peat was collected increased. In three cases (Table 2 plus two unreported experiments), however, methanogenesis increased in samples collected at approximately 210 cm. This increase corresponded with a change in peat type. Fibrous peat, present from the peatland surface to approximately 150 to 175 cm, gave way to a dark, fine-grained, highly decomposed peat. Increased rates of methanogenesis from this peat could be a result of higher numbers of methanogens, but most probable number counts (16) do not support this explanation. It also is possible that increased ground water movements at this depth might have removed metabolic end products that are inhibitory to methanogenesis. However, the data in Fig. 1 show that maximal interstitial water methane concentrations occurred in this region, indicating that metabolic end products do accumulate at these depths. The most probable explanation for this rise in methane production is an increased level of utilizable energy sources or nutrients present in this layer of fen peat. The reason this methanogenesis increase only occasionally was detected probably is a result of variation in the chemical composition of peat found at this depth.

It is possible that our data do not reflect fully the magnitude of the in situ methanogenesis decrease with depth. It is known that the upper sediments of lakes are much more active in methanogenesis than lower sediments (1, 6, 9, 10, 20). In vitro and in situ rates of methanogenesis are similar for such sediments (0 to 3 cm). However, deep sediment (45 to 105 cm) methanogenic populations could be stimulated to 70% of the activity of surface sediments (9) by treatments such as shaking (5). This indicates that, despite their in situ inactivity, these populations were viable. However, the above observations also indicate that in situ activity may be less than in vitro measurements indicate. Peat sampling and handling for the above experiments removed trapped gases and volatile compounds. Additionally, adding medium supplements or distilled water may have diluted other inhibitory factors (such as organic acids). Thus, the reported in vitro measurements of methane production must be interpreted carefully before extrapolating to in situ rates.

Methanogenesis by surface peats was more responsive to temperature alterations than was that by deep peats. The year-round temperature below approximately 25 cm in the peatlands remained between 7 and 12°C (15). The methanogenesis temperature optimum for the deepest samples was within this range.

The effects of pH on methanogenesis (Table 3) indicate that peatland methanogens are metabolizing under subopti-

TABLE 4. Effect of nutrient additions on methane production from Cedar Creek peat samples

Depth sampled (cm)	Rate of methane production[a] (nmol of CH_4 per g [dry wt] per h)															
	Formalin-killed control	r[b]	Distilled water	r	0.1% glucose	r	0.1% acetate	r	Reduced sulfur[c]	r	10 mM NP-10% TM[d]	r	2 atm of H_2-CO_2	r	1% vitamins–0.1% yeast extract	r
10	0.3	0.897	148.9	0.999	289.8	0.991	44.4	0.990	140.5	0.990	103.4	0.991	649.2	0.998	189.8	0.991
25	0.4	0.966	82.8	0.945	145.4	0.985	34.4	0.981	166.9	0.997	90.6	0.997	167.3	0.985	82.0	0.987
40	0.4	0.985	24.0	0.984	63.2	0.987	17.1	0.964	44.1	0.996	20.9	0.983	42.8	0.995	23.3	0.941
90	0.3	0.973	5.2	0.983	10.9	0.960	1.4	0.993	5.2	0.973	4.3	0.986	11.1	0.998	5.6	0.966
120	0.1	0.906	5.5	0.995	7.2	0.983	5.0	0.982	6.9	0.998	3.3	0.998	6.4	0.990	3.3	0.956
210	0.2	0.908	3.2	0.988	3.7	0.999	3.4	0.997	2.6	0.984	2.6	0.992	3.5	0.991	3.1	0.996

[a] Incubated for 67 h at 25°C.
[b] r, Correlation coefficient for linear methane production rate with time.
[c] Reduced sulfur, 0.3 mM L-cysteine, and 0.6 mM Na_2S.
[d] NP, Nitrogen-phosphorus; TM, trace metals.

TABLE 5. Effect of various treatments on methane production from Cedar Creek peat samples[a]

0- to 64-h rate	r[b]	256- to 294-h rate	r	Treatment or additive	295- to 359-h rate	r	Rate increase
229.3	0.996	36.3	0.837	Formalin kill	1.3	0.638	0
248.4	0.995	18.1	0.866	Formalin kill, VV/N[c]	0.5	0.911	0
262.6	0.973	29.1	0.675	No addition	3.3	0.728	0
230.8	0.978	26.4	0.816	No addition, VV/N	41.5	0.954	15.1
231.7	0.998	37.1	0.902	Water	14.7	0.816	0
228.4	0.984	11.3	0.749	Water, VV/N	108.1	0.998	96.8
236.2	0.997	35.2	0.806	0.1% Glucose	130.1	0.729	94.9
232.9	0.985	67.9	0.760	0.1% Glucose, VV/N	602.6	0.998	534.7
233.9	0.996	16.9	0.772	10 mM NP, 10% TM[d]	9.4	0.686	0
232.5	0.998	12.4	0.859	10 mM NP, 10% TM, VV/N	115.5	0.993	103.1
220.6	0.992	32.4	0.711	1.0% vit, 0.1% YE[e]	13.8	0.688	0
225.5	0.984	45.9	0.819	1.0% vit, 0.1% YE, VV/N	245.2	0.985	199.3
240.2	0.987	37.3	0.850	pH increase[f]	263.1	0.918	225.8
233.5	0.993	18.7	0.764	pH increase VV/N	483.6	0.999	464.9
234.7	0.999	35.1	0.644	1 atm of H_2–CO_2	26.3	0.619	0
229.6	0.983	29.7	0.903	1 atm of H_2–CO_2 VV/N	564.9	0.998	535.2

[a] Data are given as nanomoles of methane per gram (dry weight) per hour at 25°C.
[b] r, Correlation coefficient for linear methane production rate with time.
[c] VV/N, Vortex under vacuum and replace atmosphere with nitrogen (three cycles).
[d] NP, Nitrogen-phosphorus; TM, trace metals.
[e] Vit, Vitamins; YE, yeast extract.
[f] pH increase from 4.2 to 6.2.

mal pH conditions. However, it is also possible that neutral microsites exist within the peat matrix and that these sites are the primary areas where methanogenesis occurs. It also seems probable that peatlands may contain unusual strains of acid-tolerant methanogenic bacteria that produce methane at pHs below 6.0.

Providing nutritional supplements to the methanogenic food chain gave varied results (Table 4). Lack of stimulation of methane formation after the addition of nitrogen-phosphorus-trace metals, reduced sulfur, and organic growth factors indicated that these factors are not limiting. Glucose and H_2-CO_2 both stimulated methanogenesis, indicating that the availability of substrates in peat is more critical than the availability of mineral nutrients. Since glucose is not metabolized directly by methanogens, an active methanogenic food chain was required to convert glucose into methanogenic substrates (acetate, H_2-CO_2). Neither glucose nor H_2-CO_2 stimulated methanogenesis in peat samples collected below 90 cm. The failure of all additives to significantly affect methanogenesis in 120- and 210-cm peat samples indicates that some overriding factor is controlling methanogenesis in these regions. Enumerations of both methanogenic and non-methanogenic organisms (16) indicate that the absence of these bacteria is not a likely explanation. The inhibition of both methanogenic and non-methanogenic organisms seems to be a reasonable explanation for the lack of glucose and H_2-CO_2 stimulation of methanogenesis in these deep peats.

Acetate, a known substrate for methanogenic bacteria, inhibited methanogenesis in peat samples. This was unexpected but confirmed in four separate experiments. If the methanogens were utilizing primarily H_2 and CO_2 as substrates, a period of time might be required for the enrichment

TABLE 6. Composition of gas collected from Minnesota peatland sites[a]

Sample	Collected gas (μmol/ml)			
	N_2	CH_4	CO_2	O_2
Site 1				
A	22.0	13.4	2.2	2.5
B	25.4	12.5	3.1	2.4
Site 2				
A	27.2	10.2	1.1	2.5
B	18.8	11.9	3.2	2.2
C	19.3	7.6	2.0	2.0
D	18.6	17.8	2.8	2.0
Site 3				
A	22.0	16.7	2.0	2.8
B	23.9	12.3	2.7	2.4
C	22.3	11.7	2.3	2.4
Site 4				
A	21.8	14.2	3.4	2.3
B	26.3	16.5	3.0	2.5
C	20.9	10.8	3.2	2.0
Site 5	25.8	6.7	3.0	2.3
Site 6				
A	19.8	14.0	1.6	2.0
B	20.7	8.9	0.5	1.9

[a] Average values (micromoles per milliliter) were as follows: N_2, 22.3 ± 1.4; CH_4, 12.4 ± 1.7; CO_2, 2.4 ± 0.4; O_2, 2.3 ± 0.1. Average values ± 95% confidence limits (moles percent) were as follows: N_2, 57.0 ± 3.2; CH_4, 31.2 ± 3.2; CO_2, 6.0 ± 1.1; O_2, 5.8 ± 0.2.

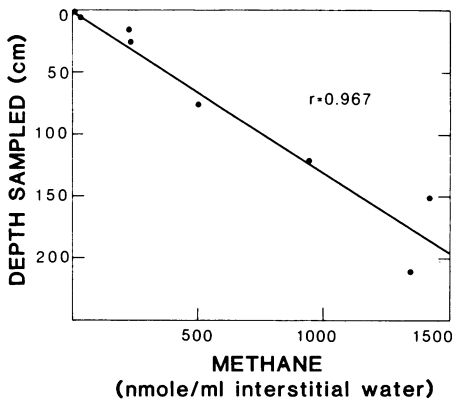

FIG. 1. Dissolved methane content of Cedar Creek peatland water.

of acetate-utilizing organisms, and such an enrichment might require several weeks. The incubation period used in the experiments reported here would not have allowed such an enrichment. However, this would account only for the lack of stimulation and not for the observed inhibition of methanogenesis. The provision of acetate might have altered carbon flow in the peat environment. If non-methanogenic food chain organisms are stimulated by acetate, then required nutrients for this food chain may be competitively removed from metabolic pathways leading to methane as an end product. This would depress methane production. Acetate toxicity is an unlikely explanation for the observed inhibition of methanogenesis, as culture studies have shown that high acetic acid concentrations (100 mM) are not toxic to the bacteria involved in methane formation (14).

In Lake Vechten sediments, maximum methane concentrations correspond to the depth of maximum methanogen numbers (3). This was not the case in the peat environments studied here. The ability of peat to trap and hold methanogenesis-inhibiting compounds produced by microbial metabolism may account for this observation. Methane was shown to be trapped within the peat matrix in both dissolved (Fig. 1) and undissolved (Table 6) forms. It is possible that the methane in deep peat regions was generated many years ago and that its presence is inhibitory to further methanogenesis. However, no evidence exists for methane inhibition of methanogenesis.

Experiments reported in Table 5 showed that removal of volatile and dilution of nonvolatile metabolic end products in peat stimulated methanogenesis. This provides circumstantial evidence for end product inhibition of the methanogenic food chain in peat environments. The buildup of end products in the peat matrix, potentially to inhibitory or toxic levels, could block anaerobic metabolism, thereby creating a significant causative factor in peat accumulation.

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