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Spatial distribution of microbial methane production pathways in temperate zone wetland soils: Stable carbon and hydrogen isotope evidence

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Abstract—The identity and distribution of substrates that support CH_4 production in wetlands is poorly known at present. Organic compounds are the primary methanogenic precursor at all depths within anoxic wetland soils; however, the distribution of microbial processes by which these compounds are ultimately converted to CH_4 is uncertain. Based on stable isotope measurements of CH_4 and ΣCO_2 extracted from soil porewaters in two temperate zone wetlands, we present evidence that a systematic spatial distribution of microbial methanogenic pathways can exist in certain anoxic, organic-rich soils. CH_4 production by the acetate fermentation pathway is favored in the shallow subsurface, while methanogenesis from the reduction of CO_2 with H_2 becomes more predominant in older, less reactive peat at depth. This distribution can account for many of the reported CH_4 emission characteristics of wetlands, in particular, their sensitivity to changes in primary productivity, temperature, and hydrology. These factors play an important role in controlling the short-term supply of labile substrates to fermentive methanogens in the shallow subsurface where the most intense CH_4 production occurs. Predominance of the CO_2-reduction pathway at depth may help to explain reports of CH_4 with a semifossil age in lower peat layers. *Copyright © 1997 Elsevier Science Ltd*

1. INTRODUCTION

Stable isotope ratios ($^{13}C/^{12}C$ and D/H) have become an important tool in the study of global CH_4 cycles (Stevens and Rust, 1982; Stevens and Engelkemeir, 1988; Quay et al., 1988); however, their effective use requires the accurate isotopic characterization of major CH_4 sources and an understanding of inherent isotope fractionating processes. Natural wetlands cover approximately 3% of the Earth's land surface (Clymo, 1987), store up to 30% of the global pool of soil carbon excluding peat (Gorham 1991, 1995), and are considered to be the largest natural contributor to the atmospheric CH_4 budget each year (Aselman and Crutzen, 1989; Bartlett and Harriss, 1993). CH_4 is produced in wetlands by two different microbial processes: acetate fermentation and CO_2 reduction with H_2. Acetate fermentation refers collectively to methanogenesis that involves the transfer of a CH_3^- group from a simple organic substrate (e.g., acetate, methanol, methylated amines, etc.) (Whiticar et al., 1986; Revesz et al., 1995). Each of the two primary methanogenic pathways yields CH_4 with distinctive carbon and hydrogen stable isotope signatures (acetate fermentation, $\delta^{13}C \sim -65$ to $-50‰$, $\delta D \sim -400$ to $-250‰$; CO_2 reduction, $\delta^{13}C \sim -110$ to $-60‰$, $\delta D \sim -250$ to $-170‰$) (Whiticar et al., 1986). Methane with intermediate $\delta^{13}C$ and δD values is generally interpreted as a mixture derived from both pathways. Although reports of these pathways are numerous (e.g., Martens et al., 1986, 1992; Burke et al., 1988a,b, 1992; Lansdown et al., 1992; Jedrysek, 1995), a clear understanding of their occurrence has yet to emerge, especially within freshwater environments. A knowledge of the distribution of microbial methanogenic pathways in freshwater wetlands could provide further insight into the nature of controls governing CH_4 precursor supply and utilization. This topic is of particular interest given current uncertainty about biosphere feedback during transitions in climate. In freshwater environments, substantial differences in supply, stability, and mobility exist between simple, organic methane-precursors (e.g., acetate) on one hand, and H_2 and CO_2, on the other hand.

Schoell (1988) presented a model in which the predominance of methanogenic pathways in the natural environment is governed by substrate quality and sediment temperature. Although both pathways may function simultaneously, their relative importance is suggested to change during different stages of sediment deposition and diagenesis. One of the important implications of his model is that CH_4 production by acetate fermentation should precede that by CO_2 reduction. Schoell (1988) cites soil CH_4 data for a swamp in Germany (Woltemate, 1982) and a bog in Illinois, USA, in which an inverse relationship was reported between $\delta^{13}C$-CH_4 and δD-CH_4 with increasing soil depth, although it was not specified whether the CH_4 was obtained from sediment free-gas or extracted from porewaters. Whiticar et al. (1986) have suggested that although ~70% of CH_4 production in freshwater systems is believed to occur via the acetate fermentation pathway, methanogenesis by CO_2 reduction could predominate after the supply of simple methylated compounds has become depleted.

Since soil temperature and the age of organic debris vary with depth in wetlands, it follows that natural gradients also should exist for methanogenic pathways in such settings. The purpose of our study was to determine if a systematic spatial distribution exists for methanogenic pathways in temperate zone wetland soils and to assess our findings with respect to current beliefs about methanogenesis and carbon cycling in wetlands.

2. METHODS

The two wetlands we studied are located in a temperate climatic zone in southwestern Ontario, Canada. The Point Pelee Marsh (41°58′00″N) is a freshwater wetland (pH ~ 6.9) located within

Point Pelee National Park. It covers an area of ~1,050 hectares and consists of extensive *Typha*-dominated mats interspersed with large, shallow open ponds. Lake Erie moderates the climate at Point Pelee, making it slightly warmer in winter and cooler in summer than the adjacent mainland. The samples discussed here were collected in September 1995. The Sifton Bog (43°00′00″N) is located within the City of London, Ontario, Canada. It is an acidic *Sphagnum* peat bog (pH ~ 3.8) and covers an area of ~28 hectares. The bog consists of a small central open pond surrounded by a floating *Sphagnum* mat that gradually changes at its periphery into forested bog and eventually upland wooded slopes. The samples discussed here were collected in August, October, and November 1995.

Porewater samples were obtained using a portable, minaturized, multilevel piezometer that had a maximum sampling resolution of 5 cm. Samples were collected without exposure to the atmosphere by slowly drawing porewaters into polypropylene syringes fitted with three-way stopcocks. All samples were stored in ice-water. Those collected for isotopic analysis of CH_4 and ΣCO_2 were also treated with $HgCl_2$ to inhibit further microbial activity. Dissolved CH_4 and ΣCO_2 concentrations were determined within 24 h by multiple equilibrations with a head space of ultra high purity (UHP) helium (McAullife, 1971). A nickel oxide catalyst was used to convert extracted CO_2 to CH_4 to permit measurement of both gases by gas chromatography using a flame ionization detector (FID). Dissolved concentrations were corrected for the stripping efficiency of each gas. Overall precision of the method was better than ±4% based on duplicate analyses. Dissolved CH_4 and ΣCO_2 were extracted for isotopic analysis by vacuum sparging of acidified samples with a UHP helium/oxygen blend (85:15). Sparging effluent was directed into a vacuum extraction line for liquid nitrogen collection of CO_2 and combustion of CH_4 to CO_2 and H_2O on CuO at 880°C. Yields from extracted ΣCO_2 and CH_4-derived CO_2 were measured manometrically. Both CH_4-derived and soil H_2O were converted to H_2 by zinc reduction (Coleman et al., 1982). Combustion yields of CH_4 from a Research Grade CH_4 standard were 99 ± 1%. The trend and values of dissolved gas concentrations measured by head space equilibration and calculated from sparging yields were in good agreement. Stable isotope compositions are reported in the standard δ notation: $\delta(‰) = ((R_{\text{sample}}/R_{\text{standard}})-1) \times 1000$ where R is D/H or $^{13}C/^{12}C$. Isotopic results for carbon and hydrogen are reported relative to VPDB and VSMOW, respectively (Coplen, 1994). The precision of $\delta^{13}C$-analyses for both dissolved ΣCO_2 and CH_4 were better than ±0.2‰, while δD measurements for dissolved CH_4 and soil H_2O had precisions, respectively, of ±3‰ and ±2‰. Soil temperatures were measured using an Omega Model HH-41 thermistor thermometer and thermistor probe (type K thermocouple). The instrument was accurate and precise to ±0.1°C.

3. RESULTS

Dissolved gas concentrations and $\delta^{13}C$ and δD values for CH_4, ΣCO_2, and H_2O from the studied wetlands are given in Table 1. The relationships observed among these results are discussed below, first for the Point Pelee Marsh and then for the Sifton Bog.

3.1. Point Pelee Marsh

Concentrations of dissolved CH_4 in porewaters from Point Pelee Marsh soils increased steadily with depth (Fig. 1a). Quantities of dissolved ΣCO_2 were greatest near the surface and gradually decreased with depth. The $\delta^{13}C$ value of dissolved CH_4 and ΣCO_2 showed an inverse relationship (Fig. 1b). The $\delta^{13}C$ value of shallow, dissolved CH_4 (−48‰) suggests minor oxidation by methanotrophs. Coexisting shallow, dissolved ΣCO_2 had low $\delta^{13}C$ values (−19‰) and probably contained CO_2 derived from both CH_4 oxidation and decomposition of fresh organic matter (bulk $\delta^{13}C \approx -25‰$). The $\delta^{13}C$ values of soil organic matter (not illustrated) increased by only 2‰ over the depth range we stud-

Table 1. Concentrations, $\delta^{13}C$ and δD values for CH_4 and ΣCO_2 from porewaters, Point Pelee Marsh and Sifton Bog.

Depth (cm)	CH_4 (µmol/L)	ΣCO_2 (µmol/L)	$\delta^{13}C_{CH4}$ (permil)	$\delta^{13}C_{\Sigma CO2}$ (permil)	δD_{CH4} (permil)	δD_{H2O} (permil)
Point Pelee Marsh (September 1995)						
0	3	1109	-48.2	-19.1	-183	-40
5	359	6163	-56.5	-15.6	-259	-41
10	—	—	-57.3	-15.1	-284	-45
15	360	6337	-58.2	-15.0	-271	—
20	398	6364	—	—	—	-44
25	421	6598	-61.1	-13.5	-316	-39
30	436	6203	-60.7	-13.8	-307	-46
35	491	6302	—	—	—	-46
40	539	6209	-62.0	-12.4	-309	—
45	—	—	-63.1	-12.7	-320	—
50	538	5724	-63.9	-12.1	-318	-40
55	507	5848	-62.5	-12.7	-314	—
60	551	5329	—	—	—	—
65	570	—	-65.8	-12.4	-294	-48
70	546	5354	—	—	—	—
75	523	5179	-68.3	-12.1	-289	—
80	586	5305	—	—	—	-47
85	—	—	-70.2	-11.4	-261	—
90	559	4467	—	—	—	—
95	—	—	-69.6	-9.2	-246	—
100	848	4573	—	—	—	-44
120	904	4640	—	—	—	-43
125	—	—	-72.3	-6.1	-253	—
150	—	—	-71.2	-5.5	-246	—
180	1126	5495	-71.6	-5.5	-244	-40
Sifton Bog (August 1995)						
0	36	982	-49.6	-12.0	—	-36
5	324	4010	-55.8	-10.3	-297	-60
15	443	4431	-57.1	-8.4	-300	-65
30	486	4622	-57.4	-8.4	-331	-67
45	548	4427	-58.7	-6.7	—	-64
60	495	4437	-58.3	-7.9	-328	-67
75	789	3787	-62.2	-3.4	-324	-71
90	775	3727	-63.4	-4.3	-321	-68
Sifton Bog (October 1995)						
0	—	—	-52.0	—	—	—
10	—	—	-54.7	—	-329	-68*
20	—	—	-56.4	—	-332	-69*
30	—	—	-55.9	—	-314	-73*
40	—	—	-57.0	—	-326	-72*
50	—	—	-58.9	—	-327	-71*
60	—	—	-56.6	—	-340	-73*
70	—	—	-58.0	—	-348	-71*
80	—	—	-56.7	—	-330	-68*
100	—	—	-60.5	—	-322	-69*
125	—	—	-63.7	—	-306	-68*
150	—	—	-66.5	—	-297	-67*
180	—	—	-67.5	—	-289	-64*
Sifton Bog (November 1995)						
5	—	—	-48.2	-16.7	—	—
10	—	—	-38.1	-16.5	—	—
15	—	—	-52.9	-14.6	—	—
20	—	—	-50.2	-15.2	—	—
25	—	—	-55.8	-13.0	—	—
35	—	—	-58.2	-11.0	—	—
50	—	—	-59.7	-10.2	—	—
65	—	—	-55.7	-8.9	—	—
80	—	—	-64.0	-9.2	—	—
120	—	—	-66.9	-1.6	—	—
175	—	—	-66.9	2.4	—	—

*These porewater samples were collected a few weeks after those from which the CH_4 was extracted. Both sets of porewaters were collected at the same site and depths. There is relatively little change in the δD values of porewaters over this period of time (Hornibrook, Longstaffe and Fyfe, unpublished data).

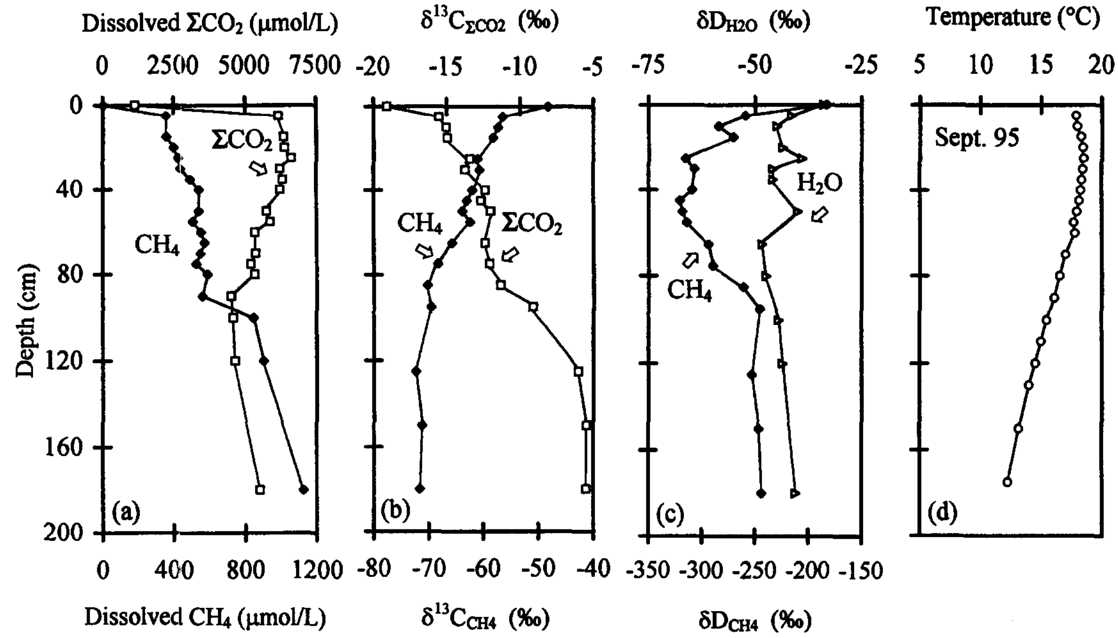

Fig. 1. Profiles through water-saturated *Typha* soils at Point Pelee marsh during September 1995 showing change with depth of (a) concentration of dissolved CH_4 and ΣCO_2, (b) carbon stable isotopic composition ($\delta^{13}C$) of CH_4 and ΣCO_2, (c) hydrogen stable isotopic composition (δD) of CH_4 and coexisting H_2O, and (d) soil temperature.

ied; most enrichment occurred in the upper 25 cm of the soil profile. The $\delta^{13}C$-CH_4 values decreased with depth, reaching values (−57 to −64‰) typical of methanogenesis by acetate fermentation between depths of 10 and 55 cm. At still greater depths, $\delta^{13}C$-CH_4 values continued to decrease, reaching values more characteristic of the CO_2-reduction pathway for methane production (< −70‰).

Current concepts of catabolic metabolism in methanogenic ecosystems generally depict a scheme where stepwise degradation of organic compounds occurs as a result of the combined activities of various groups of microorganisms. However, the redistribution of carbon and electrons in these systems does not always take the shortest route during the production of methane (Dolfing, 1988). The increase with depth in $\delta^{13}C$ values of dissolved ΣCO_2 (−15 to −5‰) suggests preferential consumption of $^{12}CO_2$; however, it is unclear from the stable carbon isotope data whether the CO_2 is being converted directly to CH_4 by methanogens or perhaps indirectly through conversion to acetate by acetogenic bacteria. Either of these mechanisms would be expected to leave a CO_2 residual enriched in ^{13}C.

The change with depth of $\alpha_{CO2\text{-}CH4}$ in Point Pelee marsh soils is depicted in Fig. 2. Samples that plot to the right of line $\alpha_C = 1.040$ are from near surface porewaters and have low α values because of methanotrophy. The $\delta^{13}C$ values of CH_4 and ΣCO_2 from shallow porewaters below the zone of oxidation plot between lines $\alpha_C = 1.040$ and 1.055, within the range typically associated with the acetate fermentation pathway. The deepest samples are shifted to the left towards the range of α values proposed by Whiticar et al. (1986) to be characteristic of the CO_2-reduction pathway. Assuming a constant fractionation between porewater CO_2 and microbially produced acetate, $\delta^{13}C$ values for CH_4 and ΣCO_2 should co-vary with increasing depth if the methanogens had consistently utilized autotrophically formed acetate. But these values diverge in the upper 1.2 m of the porewater profile (Fig. 1b). Nevertheless, three CH_4 and ΣCO_2 $\delta^{13}C$-pairs below 1.2 m depth have reasonably constant $\alpha_{CO2\text{-}CH4}$ values of 1.0714, 1.0707, and 1.0712. However, such stabilization of α_C values would occur whether methanogens were utilizing porewater CO_2 directly, or indirectly via autotrophically formed acetate.

The observed ^{12}C enrichment of ~70‰ below 1.2 m is similar to that reported for a freshwater bog (Lansdown et al., 1992) and marine sediments (e.g., Galimov and Kven-

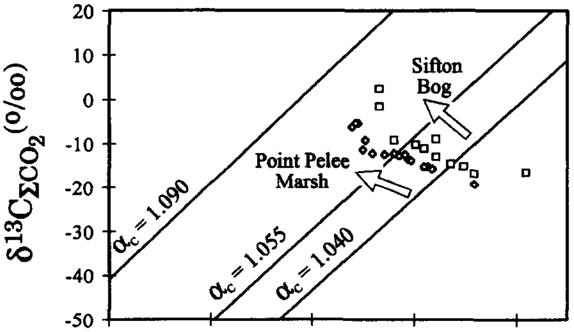

Fig. 2. $\delta^{13}C$-ΣCO_2 vs. $\delta^{13}C$-CH_4 in porewaters from the Point Pelee Marsh during September 1995 (diamonds), and the Sifton Bog in November 1995 (squares). The lines representing $\alpha_{CO_2\text{-}CH_4}$ bracket ranges of $\delta^{13}C$ values for co-existing ΣCO_2 and CH_4 reported by Whiticar et al. (1986) to be characteristic of the acetate fermentation (α_C ~1.040 to 1.055) and CO_2-reduction (α_C ~1.055 to 1.090) pathways.

volden, 1983; Claypool et al., 1985), where methanogenesis occurred predominantly by the CO_2-reduction pathway. However, the observed ^{12}C enrichment also falls within a range that might be expected if methanogens were utilizing autotrophically generated acetate. Gelwicks et al. (1989) found acetate formed by *Acetobacterium woodii* to be depleted of ^{13}C by more than 50‰ relative to the CO_2 source, and a further ^{12}C enrichment occurs during conversion of acetate to CH_4 (e.g., Blair and Carter, 1992; Sugimoto and Wada, 1993). Resolution of this ambiguity awaits further reports on the magnitude of carbon and hydrogen kinetic isotope effects associated with autotrophic acetate production. Regardless, the important implication remains that at depth within these soils, there is a change in the manner by which methane is produced and that it appears a greater quantity of methane carbon is being obtained from porewater CO_2.

The δD value of CH_4 ($-183‰$) in near surface porewaters is typical of microbial oxidation processes (Coleman et al., 1981; Happell et al., 1994). The variations in δD-CH_4 values with depth (Fig. 1c) support a transition from acetate fermentation to CO_2 reduction as the mechanism for methane production. The δD-CH_4 values initially decreased, reaching a minimum of $-320‰$ at 45cm depth, well within the range associated with acetate fermentation (Whiticar et al., 1986). Below this depth, δD values began to increase steadily, consistent with a transition to the CO_2-reduction pathway. By 180 cm depth, the δD value of dissolved CH_4 had risen to $-244‰$.

All four hydrogens in CH_4 produced by the CO_2-reduction pathway are derived from ambient H_2O (Daniels et al., 1980) with an associated fractionation of $-180 \pm 10‰$ (Whiticar et al., 1986). A larger hydrogen isotope fractionation ($\sim -317‰$) has been suggested (Sugimoto and Wada, 1995) for the CO_2-reduction pathway in freshwater systems based on *in vitro* experiments; however, abnormally high concentrations of H_2 in laboratory cultures may restrict extrapolation of these findings to the natural environment (Burke, 1993). Regardless, wetland CH_4 generated by CO_2 reduction should have lower δD values than that from marine environments since terrestrial waters are more depleted of deuterium. The geographic location (Craig, 1961) and hydrologic characteristics of a wetland will play an important role in this respect.

The large changes in δD-CH_4 values observed at Point Pelee Marsh cannot be attributed to variations in δD values of coexisting water since the latter exhibit a much smaller range (-48 to $-39‰$, excluding pond waters isotopically enriched by evaporation; Fig. 1c). Porewater δD-CH_4 and δD-H_2O pairs from Point Pelee Marsh soils are illustrated in Fig. 3, together with the relationships suggested by Whiticar et al. (1986) and Sugimoto and Wada (1995) for the covariation in δD values between CH_4 and co-existing H_2O during CO_2 reduction and acetate fermentation. A discrepancy exists between the δD values obtained for CH_4 using the CO_2-reduction pathway in natural systems versus laboratory cultures (Fig. 3; line 1 vs. 3), possibly because of abnormally high H_2 concentrations in laboratory cultures. For example, Burke (1993) presented a curve very similar to line (3) in Fig. 3, obtained using data from laboratory incubations (Balabane et al., 1987) where dissolved H_2 concentrations were several orders of magnitude greater than those typically found in natural environments.

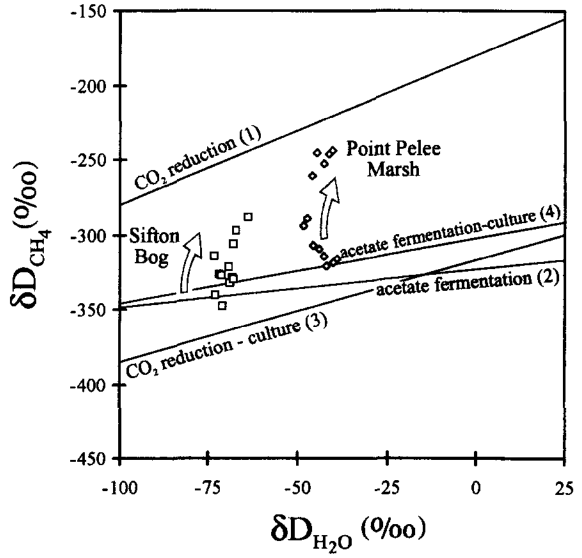

Fig. 3. δD values of dissolved CH_4 vs. δD values of co-existing H_2O from the Point Pelee Marsh during September 1995 (diamonds), and the Sifton Bog in October 1995 (squares). The arrows indicate increasing depth within the soils. The δD values for some water samples were interpolated over a 10 to 20 cm interval from samples above and below the interval of interest (see Table 1). Proposed relationships between δD-CH_4 and coexisting δD-H_2O are also illustrated: Line 1, CO_2 reduction: $\delta D_{CH_4} = \delta D_{H_2O} - 180$ ($\pm$ 10) ‰ (Whiticar et al., 1986); Line 2, acetate fermentation: $\delta D_{CH_4} = 0.25\ \delta D_{H_2O} - 321‰$ (Whiticar et al., 1986); Line 3, CO_2 reduction (from cultures): $\delta D_{CH_4} = 0.683\ (\pm\ 0.020)\ \delta D_{H_2O} - 317$ ($\pm$ 20) ‰ (Sugimoto and Wada, 1995); Line 4, acetate fermentation (from cultures): $\delta D_{CH_4} = 0.437\ (\pm\ 0.045)\ \delta D_{H_2O} - 302\ (\pm\ 15)$ ‰ (Sugimoto and Wada, 1995).

Excluding results for samples within 25 cm of the soil-water interface (zone of methane oxidation), the Point Pelee Marsh δD-CH_4 and δD-H_2O pairs (Fig. 3) plot within the limits of Eqns. 1 and 2 from Whiticar et al. (1986). Shallow porewater samples lie slightly above the endmember line for acetate fermentation of Whiticar et al. (1986) and on the line of Sugimoto and Wada (1995) for the same pathway. Samples from deeper in the soil profile are shifted more or less vertically towards the CO_2 reduction line, with the change in methane δD values being much larger than that observed for porewaters.

A comparison of progressive changes in both the $\delta^{13}C$ and δD values of dissolved CH_4 at Point Pelee Marsh helps to summarize the proposed transition in CH_4-production pathways with increasing depth (Fig. 4). Following the model of Whiticar et al. (1986), CH_4 in shallow porewaters has stable isotope signatures that are characteristic of methanotrophy. At greater depths, changes in both carbon- and hydrogen-isotope values indicate a gradual shift towards a greater importance for the CO_2-reduction pathway.

Finally, we should note that in September 1995, soil temperatures at Point Pelee Marsh had already reached their summer maximum and were beginning to decline in the upper 20 cm of the soil profile (Fig. 1d). A temperature difference of approximately 6°C existed between the near surface porewaters and those at 180 cm depth.

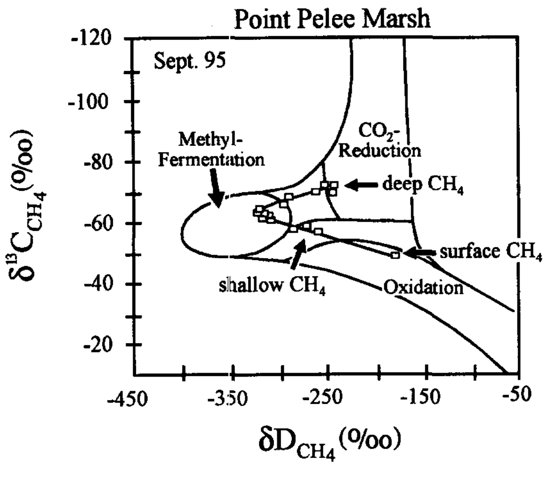

Fig. 4. $\delta^{13}C$ vs. δD values for CH_4 dissolved in porewaters from the Point Pelee Marsh during September 1995. The fields for CH_4-production pathways are taken from Whiticar et al. (1986).

3.2. Sifton Bog

Similar patterns were obtained for the Sifton Bog. Samples for August 1995 were collected to a depth of 90 cm (Fig. 5). Sampling in October 1995 extended to 180 cm depth (Fig. 6). The $\delta^{13}C$-ΣCO_2 values for the latter profile are unavailable because of an analytical problem. However, we did obtain the data needed to establish the correlation between porewater $\delta^{13}C$-CH_4 and δD-CH_4 values. Dissolved CH_4 concentrations increased with depth, while ΣCO_2 values were greatest in shallow porewaters and decreased by ~900 μmol/L over 60 cm of depth (Fig. 5a). The $\delta^{13}C$ values for CH_4 and ΣCO_2 exhibited the same inverse relationship (Fig. 5b) as Point Pelee Marsh soils. A set of samples collected in November 1995, which extend to 180 cm depth, also were analyzed for $\delta^{13}C$ of CH_4 and ΣCO_2 (Fig. 2). The trend of α_C values with increasing depth is very similar to that measured at the Point Pelee Marsh. However, unlike the marsh porewaters, $\delta^{13}C$-CH_4 and $\delta^{13}C$-ΣCO_2 pairs in the lowermost portion of the profile continue to diverge, perhaps suggesting that the transition to the CO_2-reduction pathway is less advanced. The possibility that methanogens utilized autotrophically formed acetate in the Sifton Bog waters seems less likely, given the absence of a consistent relationship between porewater $\delta^{13}C$-CH_4 and $\delta^{13}C$-ΣCO_2 values (e.g., a constant $\alpha_{CO2\text{-}CH4}$ value).

The δD-CH_4 values decreased by only a small amount over the 90 cm interval sampled in August 1995. However, the deeper profile (Fig. 6a) had $\delta^{13}C$-CH_4 and δD-CH_4 variations of a similar magnitude to those measured at Point Pelee ($\delta^{13}C = -68$ to -55‰; $\delta D = -348$ to -289‰). Gases in surficial porewaters exhibited the effects of CH_4 oxidation ($\delta^{13}C = -52$‰; $\delta D = -167$‰, not shown in Fig. 6a). Porewaters had a small range of δD-H_2O values (-73 to -64‰).

The δD pairs for dissolved CH_4 collected in October 1995 and H_2O obtained from the same site and depths a few weeks later are illustrated in Fig. 3. Sifton Bog porewaters have lower δD values than those from the Point Pelee Marsh, even though the local meteoric waters at the two sites have virtually identical average δD values (F. Colozza and F. J. Longstaffe, pers. commun.; P. Huddart et al., unpubl. data). This reflects differences in the hydrology of these two wetlands. The bog site has deeper waters with more restricted water movement, fewer vascular plants in the floating mat portion of the wetland, and much less open water to promote evaporation. As a result, the δD values of dissolved CH_4 from the Sifton Bog are lower than those from the Point Pelee Marsh. The δD-CH_4 and δD-H_2O pairs from shallow

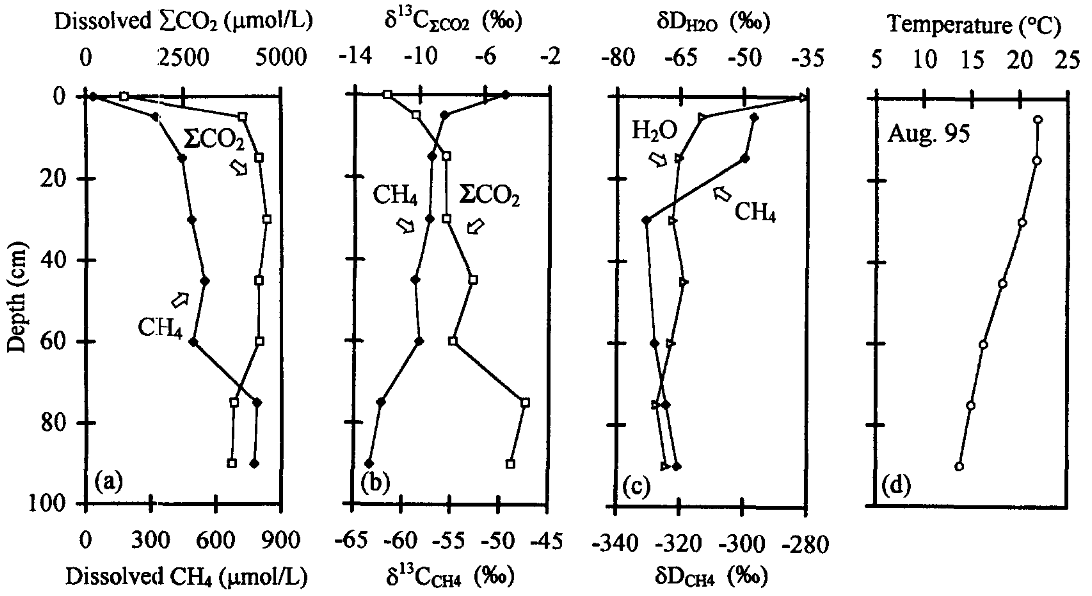

Fig. 5. Profiles through *Sphagnum* peat soils at the Sifton Bog in August 1995. Plots show change with depth of (a) dissolved CH_4 and ΣCO_2 concentrations, (b) $\delta^{13}C$ values of dissolved CH_4 and ΣCO_2, (c) δD values of dissolved CH_4 and coexisting H_2O, and (d) soil temperature.

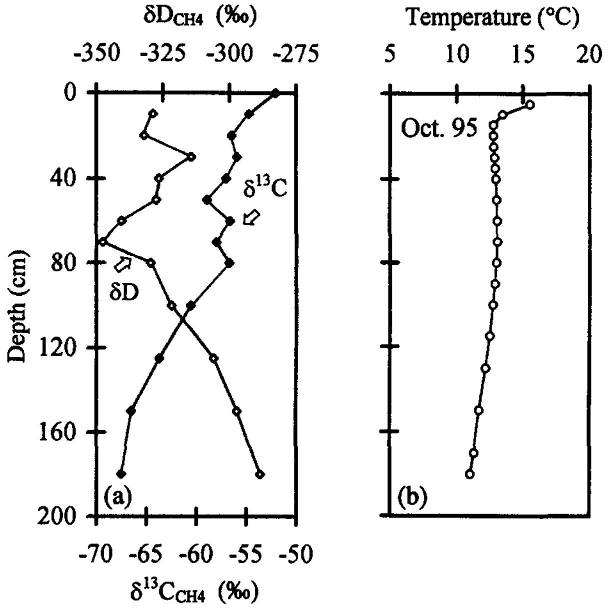

Fig. 6. Profiles through *Sphagnum* peat soils at the Sifton Bog in October 1995. Plots show change with depth of (a) δD and $\delta^{13}C$ values of dissolved CH_4, and (b) soil temperature.

porewaters in the bog lie on the lines reported by Whiticar et al. (1986) and Sugimoto and Wada (1995) for the acetate fermentation pathway. With increasing depth, the δD pairs are shifted towards the CO_2-reduction line of Whiticar et al. (1986). The spread of δD-CH_4 values is smaller in the Sifton Bog soils than in the Point Pelee Marsh over the same depth range. This supports the earlier suggestion that transition towards the CO_2-reduction pathway is less developed in the bog soils.

The covariation in $\delta^{13}C$ and δD values of CH_4 dissolved in porewaters at the Sifton Bog in October 1995 is summarized in Fig. 7. As expected, the trend with increasing depth is similar, albeit less pronounced, than that observed for the Point Pelee Marsh (Fig. 4). Most CH_4 samples have stable carbon- and hydrogen-isotope values that plot within the acetate fermentation field of Whiticar et al. (1986), but dissolved CH_4 from the deepest porewaters exhibits a distinct shift in isotopic composition towards the CO_2-reduction field.

A significant shift occurred in the soil temperature profile of the Sifton Bog between August (Fig. 3d) and October (Fig. 4b) 1995. A period of unusually warm weather in October led to a break in the fall cooling trend and caused a minor increase in near surface soil temperatures (Fig. 4b). Maximum seasonal temperatures in the lower portions of these profiles lag behind those in the shallow subsurface. Hence, stable isotope values obtained for CH_4, ΣCO_2, and H_2O collected in October 1995 from lower portions of the profile may reflect conditions of maximum warmth. We recognize that the soil systems at the Sifton Bog and the Point Pelee Marsh are seasonally dynamic. Work is in progress to determine whether the observed stable isotope profiles for CH_4, ΣCO_2, and H_2O persist throughout the year.

4. DISCUSSION

In these two very different types of freshwater wetlands, there appears to be a strong similarity in the manner with which carbon mineralization is occurring. This similarity in the pattern of isotopic behaviour for CH_4 and ΣCO_2 between the Point Pelee Marsh and the Sifton Bog provides an opportunity to comment more generally on the possible distribution of methanogenic precursors and processes in freshwater wetlands of the temperate zone.

The peat-rich subsurface of wetlands generally can be divided into two functionally distinct zones (Ingram, 1978). The upper region, known as the acrotelm, contains the living biomass, and has a relatively high permeability and experiences seasonal fluctuations in its water table level. The catotelm lies below, contains decayed or decomposing organic matter, has a lower hydraulic conductivity, and typically remains water-saturated and anoxic year round. The acrotelm is usually much thinner than the catotelm, especially where thick accumulations of peat are present. In general, this classification best applies to peat-rich wetlands such as bogs and fens. However, the Point Pelee Marsh contains thick accumulations of organic matter (~2.5 m; A. Crowe, pers. commun.) throughout much of the wetland. Accordingly, we believe that division of the subsurface into acrotelm and catotelm is suitable for this particular marsh.

The proximity of the acrotelm to the atmosphere and its higher hydraulic conductivity tend to result in oxygenation of at least a portion of this zone. Thus, the stable isotope ratios of acrotelm CH_4, such as that in the Point Pelee Marsh and the Sifton Bog, tend to exhibit the effects of methanotrophy. Oxidation of methane and aerobic decay of complex plant matter in the acrotelm primarily yields CO_2, but also produces some simpler organic compounds that can serve as substrates to methanogens in the catotelm. Immediately below the zone of oxidation, CH_4 production is dominated by the acetate fermentation pathway for several reasons: (1) higher hydraulic conductivity at these shallow depths should facilitate the movement of labile substrates from the acrotelm; (2) the surrounding organic matter is less decomposed and the bulk of plant root biomass is in or proximal to these layers; and (3) methanogens utilizing the acetate fermentation pathway benefit from higher temperatures at these shallow depths during summer months.

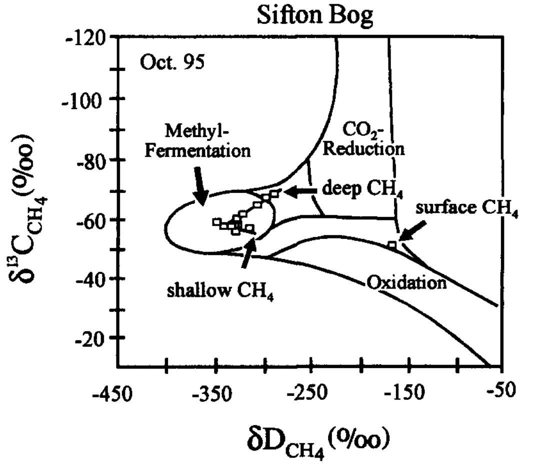

Fig. 7. $\delta^{13}C$ vs. δD values for CH_4 dissolved in porewaters from the Sifton Bog during October 1995. The fields for CH_4-production pathways are taken from Whiticar et al. (1986).

A shift in the relative importance of methanogenic pathways from acetate fermentation to CO_2 reduction in deeper portions of the peatland is likely the result of a gradually diminishing supply of labile substrates, possibly coupled with (or perhaps resulting from) lower temperatures. Intense methanogenesis in the shallow subsurface should deplete porewaters of labile DOC before it can reach deeper peat layers. Eventually, the less reactive fraction of DOC and a portion of the CO_2 generated at the surface are transported to the lower catotelm adding to the DOC and CO_2 already being generated from older, organic debris at these depths. The diminished abundance of labile organic substrates and the lower temperatures result in methane production being shifted to favour the CO_2-reduction pathway. We find it interesting that a very similar distribution of carbon mineralization processes is occurring in two very different types of freshwater wetlands.

The distribution of methanogenic pathways described here may help to explain certain CH_4 emission and storage characteristics of freshwater wetlands. CH_4 production and emission are reported to be sensitive to changes in primary productivity (Whiting and Chanton, 1993; Chanton et al., 1995), temperature (Martens et al., 1986; Burke et al., 1988b; Crill et al., 1988; Jedrysek, 1995) and hydrology (Moore et al., 1990; Dise, 1993; Romanowicz et al., 1993; Charman et al., 1994; Funk et al., 1994; Chanton et al., 1995). These factors strongly affect shallow soil layers where CH_4 production is most intense (Svensson and Rosswall, 1984) and, as shown here, occurs predominantly via the acetate fermentation pathway.

Changes in primary productivity alter the supply of labile substrates. Increased production of root biomass appears to be most important, since above-ground plant matter must accumulate initially under aerobic conditions. Dissolved organic matter is usually produced in greater quantities by anaerobic than aerobic decay since the latter generally yields substantial amounts of CO_2 (Nedwell, 1984). Reports of elevated CH_4 emissions from marsh environments (Dacey et al., 1994) in an artificially CO_2-enriched atmosphere may be the result of enhanced rhizosphere activity (e.g., Norby et al., 1992) in or near the acetate fermentation zone.

The upper layers of wetlands also experience the greatest variation in seasonal temperatures. Elevated soil temperatures during summer months appear to enhance methanogenesis (Martens et al., 1986; Burke et al., 1988b; Crill et al., 1988; Jedrysek, 1995); low winter temperatures (~0°C) are likely to inhibit entirely microbial activity in this shallow zone. At our study sites, soils below 150 cm of depth exhibit only a small seasonal temperature variation (~2 to 5°C). During the coldest months, temperatures are still sufficiently high (~8°C) for methanogenesis to proceed (Zeikus and Winfrey, 1976). Reports of CH_4 emissions during winter months (Dise, 1992, 1993) may be the result of continued CH_4 production by the CO_2-reduction pathway coupled with enhanced diffusive transport through a microbially inactive aerobic zone. It is also possible that excess CH_4 produced and stored during the warmer months is slowly being released. Since alteration by methanotrophs is unlikely, $\delta^{13}C$ and δD values of winter CH_4 emissions should indicate whether the gas is derived from microbial activity in shallow or deep porewaters.

Natural (Moore et al., 1990; Dise, 1993) or artificial (Yavitt et al., 1993) lowering of water tables can cause short-lived CH_4 emission pulses. Under normal hydrological conditions and in the absence of deeply rooted vascular plants capable of mediating CH_4 transport, dissolved CH_4 produced by CO_2 reduction should accumulate in the catotelm, since slow diffusive migration is the primary transport mechanism for release to the atmosphere. There is also evidence that gas bubbles may become trapped at depth, occluding porespaces, and further impeding upward migration of dissolved gases (Brown et al., 1989). A large drop in water table levels appears to induce a pulse of CH_4 release because of a reduction in confining pressure (Windsor et al., 1992). However, sustained lowering of water table levels generally decreases long-term CH_4 emissions (Roulet et al., 1993) and can cause an increase in CO_2 flux (Glenn et al., 1993; Funk et al., 1994). CH_4 production in the shallow subsurface may be attenuated by the expansion of the surficial oxidation zone to depths where acetate fermentation otherwise occurs. A decrease in primary productivity caused by dramatic decreases in water table levels should result in a diminished supply of labile root exudates. Methanogenesis at greater depths by CO_2 reduction may continue, but to reach the atmosphere, CH_4 would have to accumulate in quantities sufficient for ebullition to occur, since slow diffusive transport across an expanded oxic layer is unlikely to be successful. In comparison to drought-induced effects, rainstorm events have been reported to temporarily suppress CH_4 emissions (Frolking and Crill, 1994). This is perhaps the result of flushing or dilution of labile substrates in shallow subsurface porewaters by nutrient poor, oxygen-enriched precipitation.

Methane production in the catotelm by CO_2 reduction may contribute to the formation of isotopically young CH_4 (^{14}C dated) within much older layers of peat (Wahlen et al., 1989; Aravena et al., 1993; Charman et al., 1994; Chanton et al., 1995). Downward migration of recent DOC from the shallow subsurface has been suggested as the primary mechanism responsible for anomalously young CH_4 at depth (Aravena et al., 1993; Charman et al., 1994; Chanton et al., 1995). Highly soluble CO_2 derived from aerobic decomposition of recent plant matter in the acrotelm, transported downward by groundwater, also could contribute to the formation of young CH_4, provided that hydrogen remained available from the decay of organic matter. The possibility also exists that a fraction of wetland CH_4 carbon could have a direct inorganic origin. Water transfer in both directions between peatlands and underlying mineral soils has been documented (Komor, 1994; Siegel et al., 1995). Dissolution of carbonate minerals in geological materials beneath or adjacent to wetlands would likely provide ^{14}C-free and ^{13}C-enriched CO_2 to methanogens if groundwater flow was favourable. We are unaware of any reports of anomalously old CH_4 in wetland soils; however, rich fens would seem the most likely wetland type in which this might occur.

The substantial isotopic heterogeneity of dissolved CH_4 may affect the stability of the δ-CH_4 signal arising from freshwater wetlands over time. Unless isotopic fractionations resulting from transport mechanisms completely mask original methanogenic pathway δ-signatures, emissions of deep CH_4 will possess very different stable isotope ratios than those from shallow sources. Episodic release of deeply stored

CH_4 (Moore et al., 1990; Dise, 1993; Romanowicz et al., 1993) may cause a recurrent variability in the δ-CH_4 signal arising from wetlands.

The spatial distribution of methanogenic pathways in these temperate wetland soils has an interesting similarity to the one typically found in marine sediments. In both cases, labile substrates are consumed preferentially in the shallow subsurface (i.e., in the sulfate-reduction zone of anoxic marine sediments, and in the surficial oxidation and fermentive zones in freshwater soils). The end result is methanogenesis by CO_2 reduction in deeper sediments because of a diminished substrate supply. The degree of substrate depletion appears to differ between marine and freshwater systems, and possibly even among certain freshwater environments (e.g., Lansdown et al., 1992). The general principles of the model presented by Schoell (1988) appear to apply in both systems in that the relative importance of methanogenic pathways changes as a function of diagenesis and the age of buried organic matter.

A distribution of methanogenic pathways similar to the one reported here probably does not occur in all terrestrial, anoxic, organic-rich sediments. Wetlands with continuous permafrost or negligible peat accumulation likely do not possess a similar structure. Likewise, methanogenesis in the soils of forested bogs also may proceed differently because of the potential for greater input of labile organic debris at the surface of the wetland. In addition, methane production in bottom sediments of nutrient-rich lakes and ponds should be different, given the increased input of labile organic substrates produced in the water column. Knowledge of the extent to which the pathway distribution reported here exists in peatland soils would be useful for the modelling of CH_4 emissions from boreal wetlands under changing climatic conditions.

5. CONCLUSIONS

Microbial methane production pathways occur in temperate, freshwater wetland soils at the Point Pelee Marsh and the Sifton Bog with a systematic spatial distribution, at least for the August to October period. CH_4 production by the fermentation of simple, labile organic compounds is favored in the shallow subsurface, while at depth, the less reactive nature of organic debris causes methanogenesis to occur increasingly by the CO_2-reduction pathway.

Changes in primary productivity, temperature, and hydrology may affect methanogenesis by altering the short-term supply of labile organic substrates to fermentive methanogens in the shallow subsurface. The predominance of methanogenic pathway appears to be determined primarily by the availability of labile substrates and hence the degree of decomposition of organic debris. However, because of vertical water movement within the soils, as suggested by Chanton et al. (1995), methanogenesis in peatlands may be more closely linked to surficial conditions than to the litter quality of the peat itself.

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REFERENCES

Aravena R., Warner B. G., Charman D. J., Belyea L. R., Mathur S. P., and Dinel H. (1993) Carbon isotopic composition of deep carbon gases in an ombrogenous peatland, northwestern Ontario, Canada. *Radiocarbon* **35,** 271–276.

Aselmann I. and Crutzen P. J. (1989) Global distribution of natural freshwater wetlands and rice paddies, their net primary productivity, seasonality and possible methane emissions. *J. Atmos. Chem.* **8,** 307–358.

Balabane M., Galimov E., Hermann M., and Létolle R. (1987) Hydrogen and carbon isotope fractionation during experimental production of bacterial methane. *Org. Geochem.* **11,** 115–119.

Bartlett K. B. and Harriss R. C. (1993) Review and assessment of methane emissions from wetlands. *Chemosphere* **26,** 261–320.

Blair N. E. and Carter, W. D., Jr. (1992) The carbon isotope biogeochemistry of acetate from a methanogenic marine sediment. *Geochim. Cosmochim. Acta* **56,** 1247–1258.

Brown A., Mathur S. P., and Kushner D. J. (1989) An ombrotrophic bog as a methane reservoir. *Global Biogeochem. Cycles* **3,** 205–213.

Burke R. A., Jr. (1993) Possible influence of hydrogen concentration on microbial methane stable hydrogen isotopic composition. *Chemosphere* **26,** 55–67.

Burke R. A., Jr., Barber T. R., and Sackett W. M. (1988a) Methane flux and stable hydrogen and carbon isotope composition of sedimentary methane from the Florida Everglades. *Global Biogeochem. Cycles* **2,** 329–340.

Burke R. A., Jr., Martens C. S., and Sackett W. M. (1988b) Seasonal variations of D/H and $^{13}C/^{12}C$ ratios of microbial methane in surface sediments. *Nature* **332,** 829–831.

Burke R. A., Jr., Barber T. R., and Sackett W. M. (1992) Seasonal variations of stable hydrogen and carbon isotope ratios of methane in subtropical freshwater sediments. *Global Biogeochem. Cycles* **6,** 125–138.

Chanton J. P., Bauer J. E., Glaser P. A., Siegel D. I., Kelley C. A., Tyler S. C., Romanowicz E. H., and Lazrus A. (1995) Radiocarbon evidence for the substrates supporting methane formation within northern Minnesota peatlands. *Geochim. Cosmochim. Acta* **59,** 3663–3668.

Charman D. J., Aravena R., and Warner B. G. (1994) Carbon dynamics in a forested peatland in north-eastern Ontario, Canada. *J. Ecol.* **82,** 55–62.

Claypool G. E., Threlkeld C. N., Mankiewicz P. N., Arthur M. A., and Anderson T. F. (1985) Isotopic composition of interstitial fluids and origin of methane in slope sediment of the Middle American Trench, Deep Sea Drilling Project Leg 84. In *Init. Repts. DSDP, 84* (ed. R. von Huene et al.), pp. 683–691. U.S. Govt. Printing Office.

Clymo R. S. (1987) The ecology of peatlands. *Sci. Prog.* **71,** 593–614.

Coleman D. D., Risatti J. B., and Schoell M. (1981) Fractionation of carbon and hydrogen isotopes by methane-oxidizing bacteria. *Geochim. Cosmochim. Acta* **45,** 1033–1037.

Coleman M. L., Shepherd T. J., Durham J. J., Rouse J. E., and Moore G. R. (1982) Reduction of water with zinc for hydrogen isotope analysis. *Anal. Chem.* **54,** 993–995.

Coplen T. B. (1994) Reporting of stable hydrogen, carbon, and oxygen isotopic abundances (Technical Report). *Pure Appl. Chem.* **66,** 273–276.

Craig H. (1961) Isotopic variations in meteoric waters. *Science* **133,** 1702–1703.

Crill P. M., Bartlett K. B., Harriss R. C., Gorham E., Verry E. S., Sebacher D. I., Madzar L., and Sanner W. (1988) Methane flux from Minnesota peatlands. *Global Biogeochem. Cycles* **2,** 371–384.

Dacey J. W. H., Drake B. G., and Klug M. J. (1994) Stimulation of methane emission by carbon dioxide enrichment of marsh vegetation. *Nature* **370,** 47–49.

Daniels L., Fulton G., Spencer R. W., and Orme-Johnson W. H. (1980) Origin of hydrogen in methane produced by *Methanobacterium thermoautotrophicum. J. Bacteriol.* **141,** 694–698.

Dise N. B. (1992) Winter fluxes of methane from Minnesota peatlands. *Biogeochemistry* **17,** 71–73.

Dise N. B. (1993) Methane emission from Minnesota peatlands: Spatial and seasonal variability. *Global Biogeochem. Cycles* **7,** 123–142.

Dolfing J. (1988) Acetogenesis. In *Biology of Anaerobic Microorganisms* (ed. A. J. B. Zehnder), pp. 417–468. Wiley.

Frolking S. and Crill P. (1994) Climate controls on temporal variability of methane flux from a poor fen in southeastern New Hampshire: Measurement and modeling. *Global Biogeochem. Cycles* **8,** 385–397.

Funk D. W., Pullman E. R., Peterson K. M., Crill P. M., and Billings W. D. (1994) Influence of water table on carbon dioxide, carbon monoxide, and methane fluxes from taiga bog microcosms. *Global Biogeochem. Cycles* **8,** 271–278.

Galimov E. M. and Kvenvolden K. A. (1983) Concentrations and carbon isotopic compositions of CH_4 and CO_2 in gas from sediments of the Blake Outer Ridge, Deep Sea Drilling Project Leg 76. In *Init. Repts. DSDP, 76* (ed. R. E. Sheridan, F. M. Gradstein et al.), pp. 403–407. U.S. Govt. Printing Office.

Gelwicks J., Risatti J. B., and Hayes J. M. (1989) Carbon isotope effects associated with autotrophic acetogenesis. *Org. Geochem.* **14,** 441–446.

Glenn S., Heyes A., and Moore T. (1993) Carbon dioxide and methane fluxes from drained peat soils, southern Quebec. *Global Biogeochem. Cycles* **7,** 247–257.

Gorham E. (1991) Northern peatlands: Role in the carbon cycle and probable responses to global warming. *Ecol. Appl.* **1,** 182–195.

Gorham E. (1995) The biogeochemistry of northern peatlands and its possible responses to global warming. In *Biotic Feedbacks in the Global Climatic System: Will the Warming Feed the Warming?* (ed. G. M. Woodwell and F. T. Mackenzie), pp. 169–187. Oxford.

Happell J. D., Chanton J. P., and Showers W. S. (1994) The influence of methane oxidation on the stable isotopic composition of methane emitted from Florida swamp forests. *Geochim. Cosmochim. Acta* **58,** 4377–4388.

Ingram H. A. P. (1978) Soil layers in mires: Function and terminology. *J. Soil Sci.* **29,** 224–227.

Jedrysek M. O. (1995) Carbon isotope evidence for diurnal variations in methanogenesis in freshwater lake sediments. *Geochim. Cosmochim. Acta* **59,** 557–561.

Komor S. (1994) Geochemistry and hydrology of a calcareous fen within the Savage Fen wetlands complex, Minnesota, USA. *Geochim. Cosmochim. Acta* **58,** 3353–3367.

Lansdown J. M., Quay P. D., and King S. L. (1992) CH_4 production via CO_2 reduction in a temperate bog: A source of ^{13}C-depleted CH_4. *Geochim. Cosmochim. Acta* **56,** 3493–3503.

Martens C. S., Blair N. E., Green C. D., and Des Marais D. J. (1986) Seasonal variation in the stable carbon isotope signature of biogenic methane in a coastal sediment. *Science* **233,** 1300–1303.

Martens C. S., Kelley C. A., and Chanton J. P. (1992) Carbon and hydrogen isotopic characterization of methane from wetlands and lakes of the Yukon-Kuskokwim Delta, Western Alaska. *J. Geophys. Res.* **97,** 16,689–16,701.

McAullife C. (1971) GC determination of solutes by multiple phase equilibration. *Chem. Tech.* **1,** 46–51.

Moore T., Roulet N., and Knowles R. (1990) Spatial and temporal variations of methane flux from subarctic/northern boreal fens. *Global Biogeochem. Cycles* **4,** 29–46.

Nedwell D. B. (1984) The input and mineralization of organic carbon in anaerobic aquatic sediments. In *Advances in Microbial Ecology* (ed. K. C. Marshall), pp. 93–131. Plenum.

Norby R. J., Gunderson C. A., Wullschleger S. D., O'Neill E. G., and McCracken M. K. (1992) Productivity and compensatory responses of yellow-poplar trees in elevated CO_2. *Nature* **357,** 322–324.

Quay P. D., King S. L., Lansdown J. M., and Wilbur D. O. (1988) Isotopic composition of methane released from wetlands: Implications for the increase in atmospheric methane. *Global Biogeochem. Cycles* **2,** 385–397.

Revesz K., Coplen T. B., Baedecker M. J., Glynn P. D., and Hult M. (1995) Methane production and consumption monitored by stable H and C isotope ratios at a crude oil spill site, Bemidji, Minnesota. *Appl. Geochem.* **10,** 505–516.

Romanowicz E. A., Siegel D. I., and Glaser P. H. (1993) Hydraulic reversals and episodic methane emissions during drought cycles in mires. *Geology* **21,** 231–234.

Roulet N. T., Ash R., Quinton W., and Moore T. (1993) Methane flux from drained northern peatlands: Effect of a persistent water table lowering on flux. *Global Biogeochem. Cycles* **7,** 749–769.

Schoell M. (1988) Multiple origins of methane in the earth. *Chem. Geol.* **71,** 1–10.

Siegel D. K., Reeve A. S., Glaser P. H., and Romanowicz E. A. (1995) Climate-driven flushing of pore water in peatlands. *Nature* **374,** 531–533.

Stevens C. M. and Rust F. E. (1982) The carbon isotopic composition of atmospheric methane. *J. Geophys. Res.* **87,** 4879–4882.

Stevens C. M. and Engelkemeir A. (1988) Stable carbon isotopic composition of methane from some natural and anthropogenic sources. *J. Geophys. Res.* **93,** 725–733.

Sugimoto A. and Wada E. (1993) Carbon isotopic composition of bacterial methane in a soil incubation experiment: Contributions of acetate and CO_2/H_2. *Geochim. Cosmochim. Acta* **57,** 4015–4027.

Sugimoto A. and Wada E. (1995) Hydrogen isotopic composition of bacterial methane: CO_2/H_2 reduction and acetate fermentation. *Geochim. Cosmochim. Acta* **59,** 1329–1337.

Svensson B. H. and Rosswall T. (1984) *In situ* methane production from acid peat in plant communities with different moisture regimes in a subarctic mire. *Oikos* **43,** 341–350.

Wahlen M., Tanaka N., Henry R., Deck B., Zeglen J., Vogel J. S., Southon J., Shemesh A., Fairbanks R., and Broecker W. (1989) Carbon-14 in methane sources and in atmospheric methane: The contribution from fossil carbon. *Science* **245,** 286–290.

Whiticar M. J., Faber E., and Schoell M. (1986) Biogenic methane formation in marine and freshwater environments: CO_2 reduction *vs.* acetate fermentation—Isotope evidence. *Geochim. Cosmochim. Acta* **50,** 693–709.

Whiting G. J. and Chanton J. P. (1993) Primary production control of methane emission from wetlands. *Nature* **364,** 794–795.

Windsor J., Moore T. R., and Roulet N. T. (1992) Episodic fluxes of methane from subarctic fens. *Canadian J. Soil Sci.* **72,** 441–452.

Woltemate I. (1982) Isotopische Untersuchungen zur bakteriellen Gasbildung in einem Süßwassersee. Diplomarbeit, Clausthal Univ.

Yavitt J. B., Wieder R. K., and Lang G. E. (1993) CO_2 and CH_4 dynamics of a *Sphagnum*-dominated peatland in West Virginia. *Global Biogeochem. Cycles* **7,** 259–274.

Zeikus J. G. and Winfrey M. R. (1976) Temperature limitation of methanogenesis in aquatic sediments. *Appl. Environ. Microbiol.* **31,** 99–107.