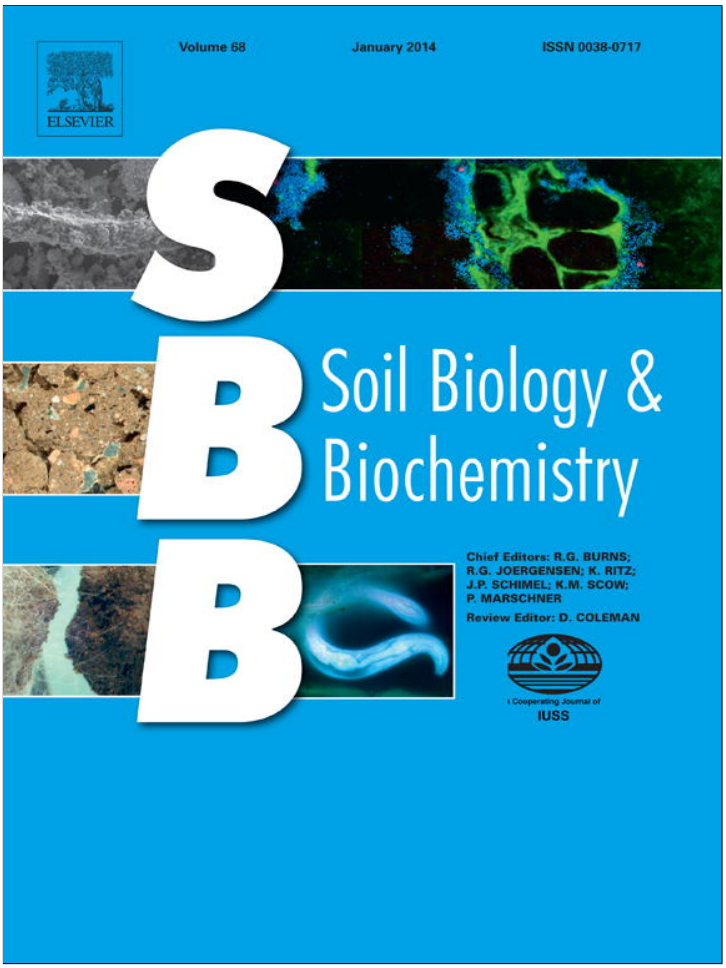

Soil Biology & Biochemistry 68 (2014) 385–391

Contents lists available at ScienceDirect

Soil Biology & Biochemistry

journal homepage: www.elsevier.com/locate/soilbio

Homoacetogenesis: A potentially underappreciated carbon pathway in peatlands

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ARTICLE INFO

Article history:
Received 16 April 2013
Received in revised form
19 August 2013
Accepted 7 October 2013
Available online 25 October 2013

Keywords:
Anaerobic carbon pathway
Homoacetogenesis
Acetate
Thermodynamics
Methanogenesis
Peatlands

ABSTRACT

Due to anaerobic conditions, peatland soils store globally significant amounts of carbon and are an important source of methane, a potent greenhouse gas. One component of anaerobic carbon cycling, homoacetogenesis (i.e., acetate formation from carbon dioxide and dihydrogen via the acetyl-CoA pathway), has rarely been quantified in natural environments because it is commonly viewed as being thermodynamically unfavorable. Here we show that in a laboratory incubation using a tracer method, homoacetogenesis occurred at significant rates in soils from three peatlands (bog, intermediate fen, and cedar swamp) despite thermodynamic conditions that appeared to be unfavorable for this process. Homoacetogens consumed dihydrogen at rates up to 3- to 10-times faster than methanogens, and homoacetogenesis accounted for 16–63% of total acetate production, with the balance likely coming from fermentation processes. Our results show that homoacetogenesis can play an important role in regulating acetate dynamics, methane production, and carbon cycling in peatlands.

1. Introduction

Understanding the mechanisms controlling anaerobic carbon (C) cycling in peatlands is essential because they contain up to one-third of the world's soil carbon, emit globally significant amounts of methane (CH_4), and are likely to represent an important feedback to future climate change (Gorham, 1991; Ise et al., 2008). The basic dynamics of organic C mineralization in wetland soils are in principle well known (Bridgham et al., 2013; Megonigal et al., 2004; Reddy and DeLaune, 2008). During anaerobic carbon mineralization, acetate is a key intermediate metabolite that provides substrates for various groups of microorganisms, such as sulfate and iron-reducing bacteria, and methanogens (Fig. 1). Acetate can be produced by the fermentation of more complex organic polymers or via homoacetogenesis (i.e., acetate formation from carbon dioxide (CO_2) and dihydrogen (H_2)). The production of acetate is an important control over CH_4 dynamics because acetate serves as the substrate for acetoclastic methanogenesis, while homoacetogens potentially compete for H_2 with hydrogenotrophic methanogens (Drake et al., 2009; Megonigal et al., 2004).

Homoacetogenesis is the reduction of CO_2 with H_2 to acetate via the acetyl-CoA pathway ($2CO_2 + 4H_2 \rightarrow CH_3COOH + 2H_2O$) (Diekert and Wohlfarth, 1994; Drake et al., 2006). This reaction is generally considered to be thermodynamically unfavorable as a result of low H_2 partial pressure in bulk porewater, which is common in many anaerobic habitats, including peatlands (Goodwin and Zeikus, 1987), because of H_2 consumption by microorganisms such as sulfate and iron reducers (Megonigal et al., 2004). In methanogenic environments (i.e. those with low concentrations of inorganic TEAs), methanogens and homoacetogens are the main H_2 consumers, with methanogens generally thought to dominate (Hoehler et al., 1999; Lovley and Klug, 1983; Jones and Simon, 1985). Nonetheless, it has been suggested that homoacetogens can outcompete methanogens at low temperature (Conrad and Wetter, 1990; Kotsyurbenko et al., 1993; Nozhevnikova et al., 1994; Liu and Conrad, 2011), likely due to higher growth rates at low temperature than methanogens (Conrad et al., 1989; Kotsyurbenko et al., 1996,

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http://dx.doi.org/10.1016/j.soilbio.2013.10.020

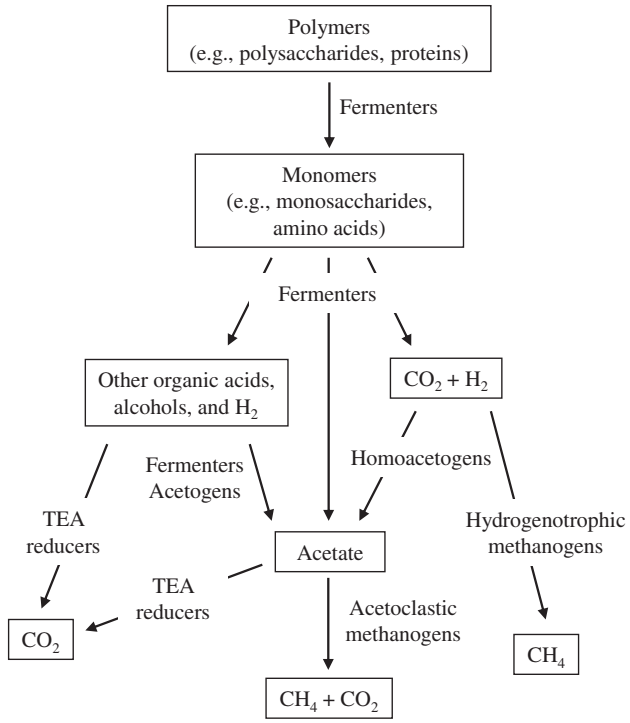

Fig. 1. Pathways for anaerobic decomposition of complex organic carbon. TEA = terminal electron acceptor.
Adapted from Megonigal et al. (2004).

2001). Past studies have also implied that homoacetogenesis can occur under seemingly thermodynamically unfavorable conditions, yet the mechanisms remain elusive (Conrad et al., 1989; Heuer et al., 2009).

Since soils of most northern peatlands have low concentrations of TEAs (Blodau et al., 2002; Janssens, 2005), current anaerobic carbon cycling theory suggests that methanogenesis should dominate anaerobic decomposition (Fig. 1). However, studies have shown that anaerobic peatland soils frequently produce little CH_4 and accumulate acetate (Hines et al., 2001; Duddleston et al., 2002; Hines et al., 2008; Keller and Bridgham, 2007; Bridgham et al., 2013). The mechanisms underlying these dynamics are very poorly understood, but one possible explanation is that homoacetogenesis may prevail in these soils at typical growing season temperatures (Duddleston et al., 2002). In support of this hypothesis, we recently observed substantial acetate accumulation at 17 °C during an anaerobic incubation of peat soils (Ye et al., 2012). To understand the source of this acetate, we anaerobically incubated soil samples from a bog, an intermediate fen, and a cedar swamp from the Upper Peninsula of Michigan, USA in the laboratory with a ^{14}C-CO_2 tracer and measured rates of CO_2 respiration, homoacetogenesis, hydrogenotrophic methanogenesis, and acetoclastic methanogenesis in these soils.

2. Materials and methods

2.1. Sample preparation

The current study was run concurrently with a large pH-controlled experiment (Ye et al., 2012), which suggested significant rates of homoacetogenesis in peatland soils. To minimize potential pH artifacts, we used peat samples at a pH equal to or closest to their native pHs. Thus, the bog peat was incubated at pH 3.5 (0.2 pH unit lower than the native), fen peat at pH 4.5 (equal to the native), and swamp peat at pH 6.5 (0.5 pH unit higher than the native). pH within a site can easily vary by half of a pH unit seasonally, so these incubation pHs are reasonable.

Peat samples were collected from three peatlands in the Upper Peninsula of Michigan, USA in August 2009 (Table 1). Four cores were extracted with PVC tubes (10-cm diameter, 15-cm length) from below the water table in hollows from each site and transported on ice to our laboratory at the University of Oregon and frozen at −20 °C until use. After thawing at room temperature in an anaerobic glove box filled with 2% H_2 and 98% N_2 gas (Coy Laboratory Products Inc., Grass Lake, MI, USA), each peat core was homogenized by hand after the removal of woody material, large roots, and green vegetation. A subsample was dried at 60 °C for 3 days to determine the moisture content. Approximately 120 g of peat from each core was transferred to a 440 mL Mason jar and mixed well with 240 mL of degassed and deionized water, followed by the adjustment of pH to the targeted value with either 10 N HCl or 10 N NaOH. The pH was adjusted daily in the glove box during the first week of incubation and once every 2 or 3 days afterward when pH changes in most of the slurries were <0.2 pH units. After pH adjustment, the slurries were capped and bubbled with oxygen-free N_2 gas for 10–15 min and then incubated in the dark at 17 °C, which was the average field temperature when the peat cores were collected.

2.2. CO_2 and CH_4 production, CH_4 pathway, acetogenic CO_2 reduction

On Days 2, 7, 15, and 43 of the incubation, 10 g of slurried peat from each sample was transferred into a 40 mL vial (I-Chem, VWR International LLC, CA, USA) in the glove box following pH adjustment. The peat slurries were capped and bubbled with oxygen-free N_2 gas for 5–10 min. Following the addition of 0.1 mL of 3.5 μCi mL^{-1} $NaH^{14}CO_3$, the slurries were gently shaken and incubated at 17 °C in the dark. After incubation for 48 h, slurries were shaken to release trapped gas bubbles and headspace CO_2 and CH_4 were quantified by gas chromatography using a flame ionization detector equipped with a methanizer (SRI Instruments, Torrance, CA, USA), while $^{14}CO_2$ and $^{14}CH_4$ production were measured concurrently with an in-line radioactive gas detector (LabLogic Systems Inc., Brandon, FL, USA). Production of CH_4 and CO_2 were calculated as the sum of production in both gas and liquid phases (Stumm and Morgan, 1995). Porewater was then collected from the same subsamples and filtered with a Whatman GF/F glass fiber filter (Sigma–Aldrich, MO, USA), followed by a second filtration with a 0.22 μm syringe filter (Tisch Scientific, OH, USA). Acetate concentrations of the water samples were determined with a Dionex DX500 ion chromatograph system equipped with an HC-75 column (Hamilton Company, Reno, ND, USA), with the effluent of

Table 1
Physical and biological characterizations of the sampling sites.

	Location	Dominant vegetation	Peat depth	pH	Water-table depth[a]
Bog	Michigan (N46°6′6″, W88°16′25″)	*Sphagnum* spp.	380 cm	3.7	−34 cm
Intermediate fen	Wisconsin (N46°12′48″, W89°30′2″	*Sphagnum* spp.	340 cm	4.5	−23 cm
Cedar swamp	Michigan (N46°14′41″, W89°32′38″)	*Thuja occidentalis* L.	290 cm	6.0	−16 cm

[a] Measured from hollow surface.

the acetate peak subsequently collected with an in-line fraction collector (Amersham Biosciences, Piscataway, NJ, USA). The activity of ^{14}C-acetate in the collected effluent was quantified with a liquid scintillation counter (Beckman Instruments Inc., Fullerton, CA, USA) corrected for a recovery rate for ^{14}C-acetate of 65% that was determined empirically by us.

Hydrogenotrophic methane production ($CH_{4,hyd}$) was calculated as described by Keller and Bridgham (2007), i.e.

$$CH_{4,hyd} = a\left[\sum CO_2\right]\alpha/Atg \quad (1)$$

where a is the recovered activity of $^{14}CH_4$, $\sum CO_2$ represents the available CO_2 pool, α (=1.12) is the ^{14}C:^{12}C isotope fractionation factor for hydrogenotrophic methanogenesis, A is the activity of available $H^{14}CO_3^-$, t is the incubation time, and g is the dry mass of peat in the slurry. Unlike Keller and Bridgham (2007), we utilized values averaged over the entire incubation period for $\sum CO_2$ and A (Ye et al., 2012). Acetoclastic methanogenesis was defined as the difference between hydrogenotrophic CH_4 production and total CH_4 production. In our pilot experiments, acetoclastic methanogenesis was greatly underestimated with ^{14}C-acetate, which has also been reported by Avery et al. (1999).

Homoacetogenesis (H) was calculated as described by Hoehler et al. (1999) with slight modification, i.e.

$$H = a\left[\sum CO_2\right]\alpha/2Atg \quad (2)$$

where a is the recovered activity of ^{14}C-acetate, and the other variables are the same as described previously. The factor 2 accounts for 2 mol of CO_2 being consumed for every mole of acetate produced. We also made a crude estimate of the amount of ^{14}C-acetate that was converted into CH_4 via the acetoclastic pathway by multiplying the rate of acetoclastic methanogenesis by one half of the specific activity of acetate at the end of the incubation (as an estimate of the activity at the half way point).

2.3. Acetate production

We assumed that acetoclastic methanogenesis was the major acetate sink and calculated gross acetate production as:

$$\text{Gross acetate production} = \text{acetate}\ (t_2) - \text{acetate}\ (t_1) + \text{acetoclastic methanogenesis} \quad (3)$$

where, [acetate (t_2) − acetate (t_1)] was the change in porewater acetate pool size between two sampling time intervals. We further assumed that 1 mol of acetate produces 1 mol CH_4 via the acetoclastic pathway (Conrad, 1999).

2.4. Thermodynamic calculations

Acetate concentration and the partial pressure of CO_2 and CH_4 were used to calculate the minimal H_2 partial pressures for Gibbs free energy (ΔG) = 0 for both hydrogenotrophic methanogenesis and homoacetogenesis (Conrad and Wetter, 1990).

2.5. Statistical analyses

Rates of methanogenesis, CO_2 production, homoacetogenesis, and concentrations were analyzed with the MIXED procedure of SAS 9.1 (SAS Institute) with time as a repeated variable. Tukey's test at $\alpha = 0.05$ was performed to detect significant differences between individual sites and time intervals. Data were log-transformed if that significantly improved the overall distribution. Pearson correlations were used to examine relationships between variables.

3. Results

3.1. CH_4 production

CH_4 was mostly produced by acetoclastic methanogenesis in all peats during the 43-day incubation (Fig. 2). Both acetoclastic and hydrogenotrophic CH_4 production, and thus overall CH_4 production, were lowest in the bog peat throughout the incubation. Rates of acetoclastic CH_4 production were similar in the swamp and fen peats on Days 2 through 15, but rates decreased to half of the initial value in the swamp peat from Day 15 to 43. In contrast, acetoclastic

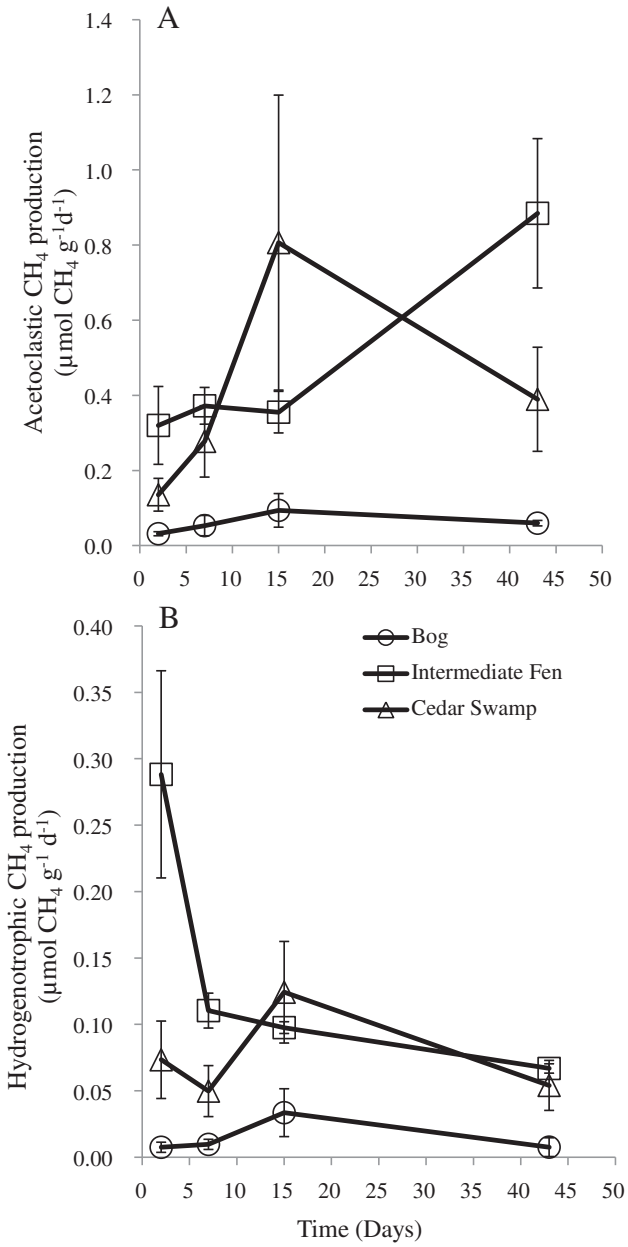

Fig. 2. CH_4 production rates in peat from a bog (circle), an intermediate fen (square), and a cedar swamp (triangle). A, acetoclastic production; B, hydrogenotrophic production; error bars indicate ±1 standard error (n = 4).

CH_4 production in the fen peat more than doubled during the same period (Fig. 2A). The rate of hydrogenotrophic CH_4 production in swamp peat did not change significantly during the incubation. On Day 2, hydrogenotrophic methanogenesis in the fen was highest, but it decreased sharply during the first week and continued to decrease more slowly thereafter (Fig. 2B), such that rates of hydrogenotrophic CH_4 production in the fen and swamp peats were similar after Day 7.

3.2. CO_2 production

CO_2 production did not change significantly through time in either the bog or swamp peats, and it was lowest in bog peat over the entire incubation (Fig. 3). CO_2 production was initially much greater in the fen peat than in both the bog and swamp peats, but it decreased to one-third of the initial value from Day 2 to 7 and remained relatively stable and similar to the swamp peat thereafter (Fig. 3).

3.3. Acetate concentration

Acetate concentration increased rapidly through time in the fen peat through Day 15 and was higher than in the other two peats, but the concentration decreased by 43% on Day 43 (Fig. 4). Acetate concentration continued to increase through time in the bog peat at a more moderate rate, so that by Day 43 the concentration was not significantly different from the fen peat. In the swamp peat, acetate concentration decreased from Day 7 to Day 43 such that it was more than 20-times lower than that of the bog and fen peats on Day 43 (Fig. 4).

3.4. Homoacetogenic production

We calculated that on average only 0.2% of the ^{14}C-acetate was converted to CH_4 via the acetoclastic pathway, with a maximum of 2.5% (data not shown), so our rates of homoacetogenesis based on the net accumulation of ^{14}C-acetate were not unduly underestimated. On Day 2, homoacetogenesis in the fen peat was 15- to 30-times higher than in the bog peat and swamp peats (Fig. 5). However, the rate decreased significantly from Day 2 to Day 7, at which time it was not different from the swamp or the bog peats. Rates of homoacetogenesis in all peats did not change significantly from Days 7 to 15. However, the rate in the swamp peat was higher than the bog and fen peats on Day 15. Similarly, this rate did not change significantly in all peats from Day 15 to Day 43 and there were no differences among peats on Day 43.

4. Discussion

4.1. The occurrence of homoacetogenesis

Rates of anaerobic carbon mineralization and the ratio of its end products, CO_2 and CH_4, are the result of a suite of complicated interactions among multiple microbial functional groups (Fig. 1). One component of such interactions, homoacetogenesis, is generally considered thermodynamically unfavorable in many anaerobic environments, primarily due to the low H_2 concentration resulting from its consumption by TEA reducers or methanogenic archaea

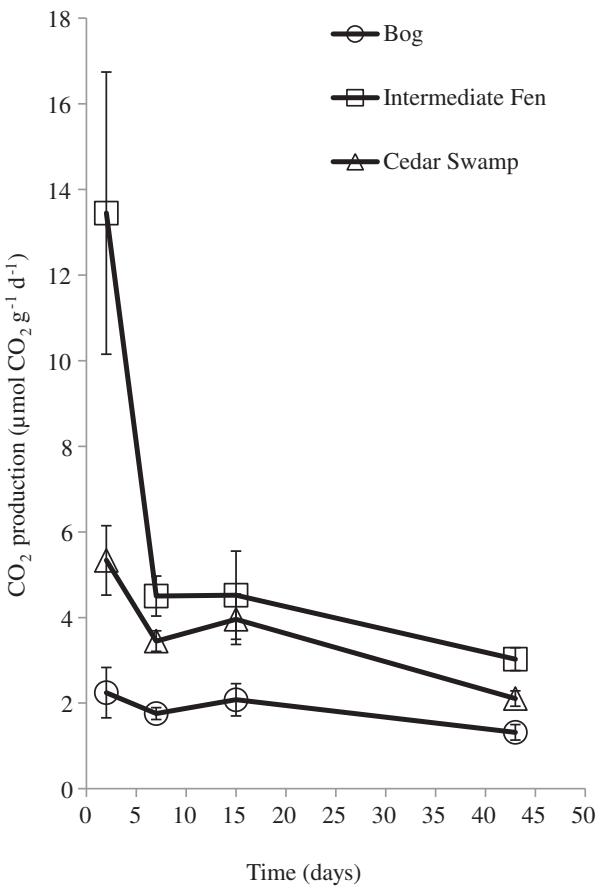

Fig. 3. CO_2 respiration rates in peat from a bog (circle), an intermediate fen (square), and a cedar swamp (triangle). Error bars indicate ±1 standard error ($n = 4$).

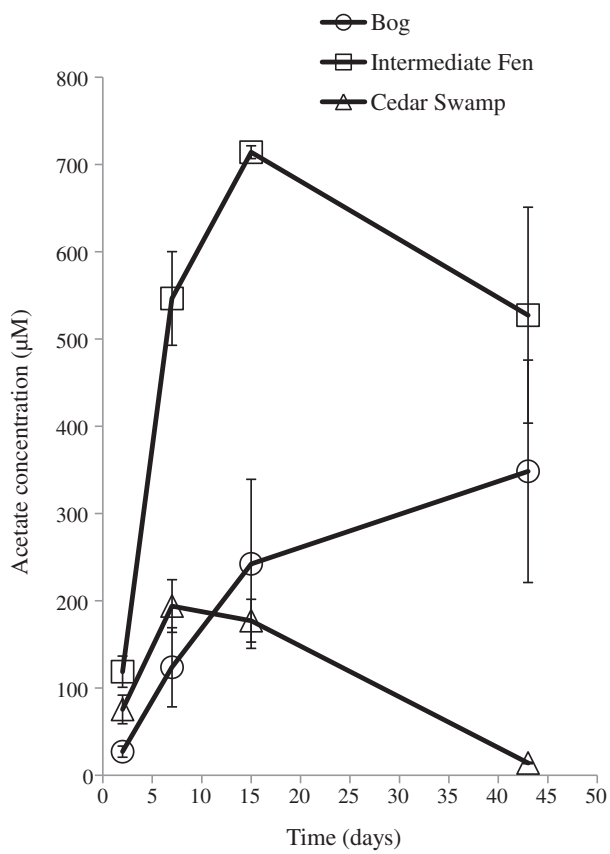

Fig. 4. Acetate concentration in peat from a bog (circle), an intermediate fen (square), and a cedar swamp (triangle). Error bars indicate ±1 standard error ($n = 4$).

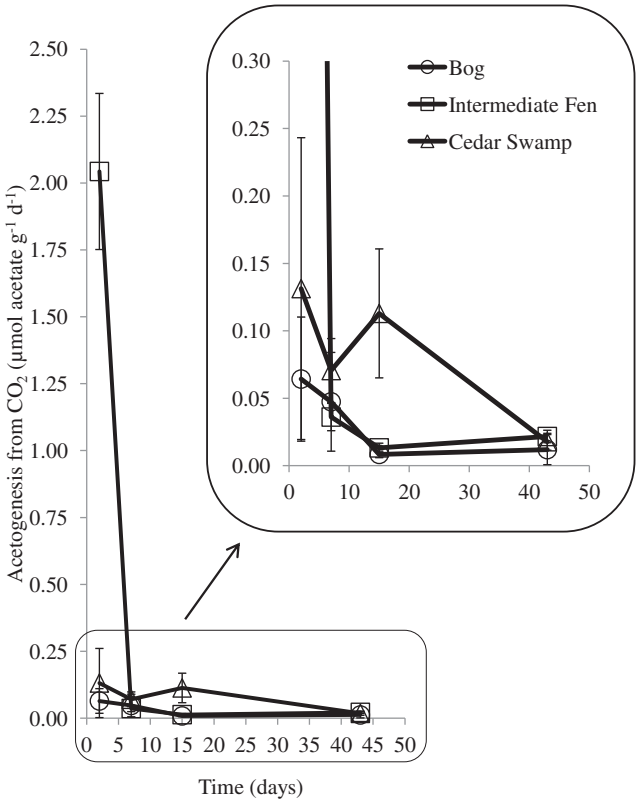

Fig. 5. Rates of homoacetogenesis measured using a ^{14}C-CO_2 tracer in peat from a bog (circle), an intermediate fen (square), and a cedar swamp (triangle). Error bars indicate ±1 standard error ($n = 4$).

(Megonigal et al., 2004; Drake et al., 2006). Thus, homoacetogenesis is commonly considered less important in carbon cycling and has rarely been quantified in natural environments (Hoehler et al., 1999; Lovley and Klug, 1983; Jones and Simon, 1985; Drake et al., 2006).

In the present study we quantified homoacetogenesis production with radioactive tracers in soils from three peatland types. While our laboratory incubation study did not measure *in situ* rates of the biological processes that produce and consume acetate, it does suggest that homoacetogenesis is potentially an underappreciated carbon pathways in peatlands as it contributed 16–63% of acetate production and consumed H_2 at rates up to 3- to 6-time faster than hydrogenotrophic methanogenesis during the 43-day incubation.

By Day 2, we detected the reduction of $^{14}CO_2$ to both ^{14}C-acetate and ^{14}C-CH_4 in all samples (Figs. 2B and 5), indicating that both homoacetogenesis and hydrogenotrophic methanogenesis were occurring simultaneously. Thermodynamic calculations suggest that for homoacetogenesis to have occurred the minimal H_2 partial pressure had to be 40 Pa (17 μmol L^{-1}), 24 Pa (10 μmol L^{-1}), and 7 Pa (3 μmol L^{-1}) in peat from the bog, fen, and swamp, respectively. Typical H_2 partial pressures of 3–11 Pa (1–5 μmol L^{-1}) (measured separately in another experiment under similar conditions) are sufficient for methanogens to prevail in all peats, but too low to explain the observed homoacetogenesis in soils from the bog and fen. This apparent discrepancy can be explained if H_2-consuming homoacetogens rely mainly on interspecies H_2 transfer from syntrophic bacteria rather than on porewater H_2, like some methanogens (Conrad et al., 1985; Conrad and Babbel, 1989) and acetogens (Leadbetter et al., 1999; Drake et al., 2006). Other studies have also suggested homoacetogenesis can occur in seemingly thermodynamically unfavorable environments, such as anaerobic rice paddy soils (Conrad et al., 1989) and deep seafloor sediments (Heuer et al., 2009).

Reduction of a mole of CO_2 via either homoacetogenesis or hydrogenotrophic methanogenesis requires 4 mol of H_2. Therefore, the ratio of homoacetogenesis to hydrogenotrophic CH_4 production describes the relative proportion of H_2 utilized by the two processes. Our results suggested that 3- to 10-times more H_2 was consumed by homoacetogenesis than hydrogenotrophic methanogenesis on Day 2 in the 3 different peats (Fig. 6). While the rates of both processes generally declined over the course of the incubation, hydrogenotrophic methanogenesis surpassed homoacetogenesis by Day 7 in the fen peat, by Day 15 in the swamp peat, and by Day 43 in the bog peat (Fig. 6). Our results demonstrate that homoacetogens are capable of dominating H_2 consumption over hydrogenotrophic methanogens in peatlands at moderate temperatures (17 °C), reflecting the mid-summer conditions in our sites when the greatest carbon mineralization rates occur. Competition between methanogens and homoacetogens for H_2 ultimately influences the flow of C and electrons to either acetate or CH_4. However, if there is a compensatory response of acetate conversion into CH_4 via the acetoclastic pathway, there may be little overall effect on net CH_4 production. While there was a tendency for rates of acetoclastic methanogenesis to increase over time in our study (Fig. 2A), we still observed substantial acetate accumulation in bog and fen peat (Fig. 4), suggesting that acetoclastic methanogenesis did not fully compensate for acetate production.

Declining rates of homoacetogenesis over time were not associated with greater hydrogenotrophic methanogenesis rates

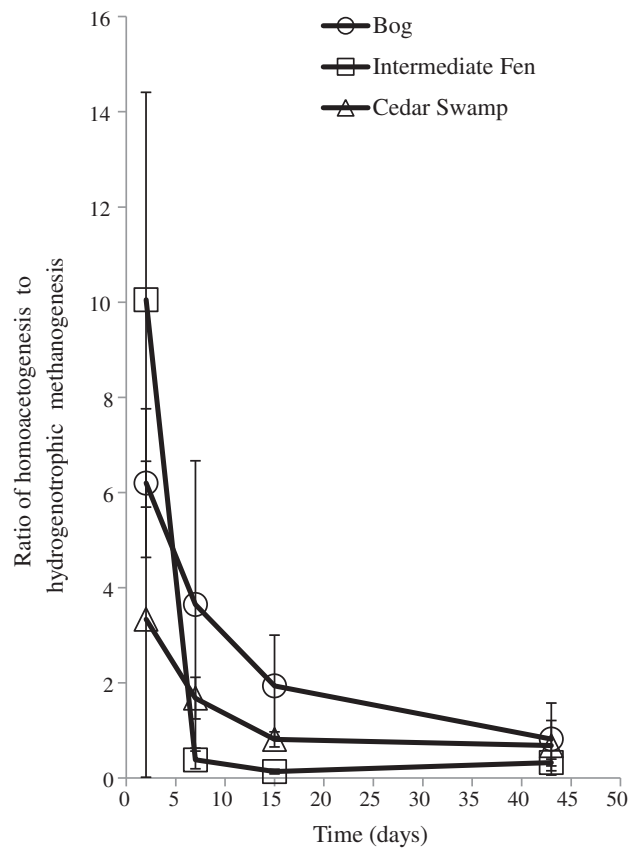

Fig. 6. Ratio of homoacetogenesis to hydrogenotrophic methanogenesis measured using ^{14}C-CO_2 in peat from a bog (circle), an intermediate fen (square), and a cedar swamp (triangle). Error bars indicate ±1 standard error ($n = 4$).

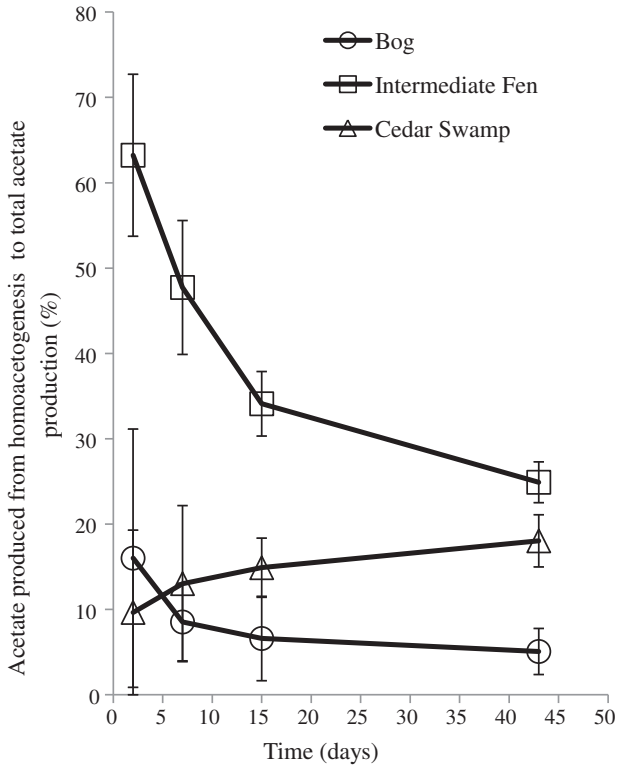

Fig. 7. Percentage of acetate produced via homoacetogenesis in peat from a bog (circle), an intermediate fen (square), and a cedar swamp (triangle). The balance of acetate is assumed to come from heterotrophic acetogenesis. Error bars indicate ±1 standard error ($n = 4$).

(Figs. 2B and 5). In fact, rates of homoacetogenesis and hydrogenotrophic methanogenesis were positively correlated ($r^2 = 0.76$, $p < 0.001$). This implies the coexistence of homoacetogenesis and hydrogenotrophic methanogenesis during the entire course of our experiment, but it is still reasonable to assume that both microbial groups were competing for a finite pool of H_2. In support of this assumption, the probable reason for the declining rates of both processes was substrate limitation (i.e., low H_2 availability). The production of CO_2 decreased during the experiment, especially from Days 2 to 7 (Fig. 3), suggesting the onset of carbon limitation for overall anaerobic mineralization. Since H_2 originates primarily from fermentative processes (Megonigal et al., 2004), it is reasonable to postulate that H_2 production decreased along with CO_2 production from Days 2 to 43, which subsequently inhibited homoacetogenesis and hydrogenotrophic methanogenesis. Our results suggest that fermentative production of H_2 is potentially an important factor controlling the absolute rates and relative portions of homoacetogenesis and hydrogenotrophic CH_4 production in peatlands.

Table 2
Contribution of homoacetogenesis to total acetate production in anaerobic systems.

Systems	Homoacetogenesis (%)	Data source
Peatland soils	5–63	Present study
Freshwater lake sediments	1.8–2	(Lovley and Klug, 1983; Phelps and Zeikus, 1984)
Termite guts	10–33	(Brauman and Kane, 1992; Breznak and Switzer, 1986; Leadbetter et al., 1999; Tholen and Brune, 2000)
Rice roots	20–40	(Chidthaisong and Conrad, 2000; Conrad and Klose, 1999)
Wastewater sludge in bioreactors	5–32	(Nie et al., 2007, 2008)

4.2. The contribution of homoacetogenesis to acetate production

Acetate is thought to be produced predominantly by heterotrophic fermentation reactions in soils and sediments, whereas homoacetogenesis is generally considered to be of minimal importance (Megonigal et al., 2004; Drake et al., 2006; Hadrich et al., 2012). To estimate the contribution of homoacetogenesis to total acetate production, we estimated total acetate production as the sum of net acetate production and acetate consumption by acetoclastic methanogens. This approach assumes that no other pathways were consuming acetate, with the most likely candidates being sulfate reduction (Vile et al., 2003; Keller and Bridgham, 2007) and the use of humic substances as terminal electron acceptors (Keller et al., 2009). We did not detect any sulfate (<4 μM) in any sample, but previous work has demonstrated that small sulfate pools can rapidly turnover in peatland soils (Wieder et al., 1990; Vile et al., 2003). Further, we have recently demonstrated that solid-phase peat may be a significant electron acceptor in a bog soil (Keller and Takagi, 2013). Thus, our values for acetate production should be considered a minimum estimate.

Results indicated that acetate production from both fermentation and homoacetogenesis were important components of total anaerobic carbon mineralization, especially in the bog peat, and in most cases was much greater than CH_4 production (Figs. 2 and 7). The amount of acetate produced from homoacetogenesis contributed from 25% to 63% of the total acetate production in fen peat, from 5% to 16% in bog peat, and from 10% to 18% in swamp peat (Fig. 7). These percentages are much higher than in freshwater lake sediments but comparable to rice roots, termite guts, and bioreactors (Table 2). It is clear that homoacetogenesis is important to the acetate production in these soils, especially in the intermediate fen peat.

5. Conclusions

The current understanding of the processes and controls of anaerobic carbon cycling in northern peatlands assumes that homoacetogenesis is unimportant (Blodau, 2002; Nilsson and Öquist, 2009), because it is generally viewed as thermodynamically unfavorable in most anaerobic environments (Megonigal et al., 2004; Drake et al., 2006). We report here significant rates of homoacetogenesis in soils from three different types of peatlands across a landscape hydrogeomorphic gradient, despite thermodynamic conditions that appear to be unfavorable for this process. Our results suggest that homoacetogenesis may be an important H_2 sink, may compete strongly with hydrogenotrophic methanogenesis, and may contribute to a significant portion of the overall acetate production in northern peatlands. Thus our data, and the frequently observed acetate pooling *in situ* in peatlands, suggest that the reduction in hydrogenotrophic methanogenesis due to formation of acetate via homoacetogenesis is not fully compensated for by an increase in acetoclastic methanogenesis. The net result is likely lower rates of overall CH_4 production in peatlands than would be expected for anaerobic environments with low concentrations of inorganic terminal electron acceptors. More research is warranted to quantify homoacetogenesis *in situ* in a large range of peatland types, to study the mechanisms regulating the occurrence of this largely underappreciated biological process, and to investigate how this process influences acetate dynamics and CH_4 production in these globally significant ecosystems.

Acknowledgments

We thank James Ziemer and Yvonne Ziemer for access to their private field site and the University of Notre Dame Environmental Research Center for access to field sites and laboratory facilities. This work was supported by NSF grant DEB-0816575.

References

Avery Jr., G.B., Shannon, R.D., White, J.R., Martens, C.S., Alperin, M.J., 1999. Effect of seasonal changes in the pathways of methanogenesis on the $\delta^{13}C$ values of pore-water methane in a Michigan peatland. Glob. Biogeochem. Cycles 13, 475–484.
Blodau, C., 2002. Carbon cycling in peatlands – a review of processes and controls. Environ. Rev. 10, 111–134.
Blodau, C., Roehm, C.L., Moore, T., 2002. Iron, sulfur, and dissolved carbon dynamics in a northern peatland. Arch. Hydrobiol. 154, 561–583.
Brauman, A., Kane, M.D., 1992. Genesis of acetate and methane by gut bacteria of nutritionally diverse termites. Science 257.
Breznak, J.A., Switzer, J.M., 1986. Acetate synthesis from H(2) plus CO(2) by termite gut microbes. Appl. Environ. Microbiol. 52, 623–630.
Bridgham, S.D., Cadillo-Quiroz, H., Keller, J.K., Zhuang, Q., 2013. Methane emissions from wetlands: biogeochemical, microbial, and modeling perspectives from local to global scales. Glob. Change Biol..
Chidthaisong, A., Conrad, R., 2000. Specificity of chloroform, 2-bromoethanesulfonate and fluoroacetate to inhibit methanogenesis and other anaerobic processes in anoxic rice field soil. Soil Biol. Biochem. 32, 977–988.
Conrad, R., 1999. Contribution of hydrogen to methane production and control of hydrogen concentrations in methanogenic soils and sediments. FEMS Microbiol. Ecol. 28, 193–202.
Conrad, R., Babbel, M., 1989. Effect of dilution on methanogenesis, hydrogen turnover and interspecies hydrogen transfer in anoxic paddy soil. FEMS Microbiol. Ecol. 62, 21–28.
Conrad, R., Bak, F., Seitz, H.J., Thebrath, B., Mayer, H.P., Schutz, H., 1989. Hydrogen turnover by psychrotrophic homoacetogenic and mesophilic methanogenic bacteria in anoxic paddy soil and lake sediment. FEMS Microbiol. Ecol. 62, 285–294.
Conrad, R., Klose, M., 1999. Anaerobic conversion of carbon dioxide to methane, acetate and propionate on washed rice roots. FEMS Microbiol. Ecol. 30, 147–155.
Conrad, R., Phelps, T.J., Zeikus, J.G., 1985. Gas metabolism evidence in support of the juxtaposition of hydrogen-producing and methanogenic bacteria in sewage-sludge and lake-sediments. Appl. Environ. Microbiol. 50, 595–601.
Conrad, R., Wetter, B., 1990. Influence of temperature on energetics of hydrogen metabolism in homoacetogenic, methanogenic, and other anaerobic-bacteria. Arch. Microbiol. 155, 94–98.
Diekert, G., Wohlfarth, G., 1994. Metabolism of homoacetogens. Anton Leeuw Int. J. Gen. Mol. Microbiol. 66, 209–221.
Drake, H.L., Horn, M.A., Wust, P.K., 2009. Intermediary ecosystem metabolism as a main driver of methanogenesis in acidic wetland soil. Environ. Microbiol. Rep. 1, 307–318.
Drake, H.L., Küsel, K., Matthies, C., 2006. Acetogenic prokaryotes. In: Dworkin, M., Falkow, S., Rosenberg, E., Schleifer, K.H., Stackebrandt, E. (Eds.), The Prokaryotes. Springer, New York, pp. 354–420.
Duddleston, K.N., Kinney, M.A., Kiene, R.P., Hines, M.E., 2002. Anaerobic microbial biogeochemistry in a northern bog: acetate as a dominant metabolic end product. Glob. Biogeochem. Cycles 16, 11. http://dx.doi.org/10.1029/2001GB001402.
Goodwin, S., Zeikus, J.G., 1987. Ecophysiological adaptations of anaerobic-bacteria to low pH – analysis of anaerobic-digestion in acidic bog sediments. Appl. Environ. Microbiol. 53, 57–64.
Gorham, E., 1991. Northern peatlands – role in the carbon-cycle and probable responses to climatic warming. Ecol. Appl. 1, 182–195.
Hadrich, A., Heuer, V.B., Herrmann, M., Hinrichs, K.U., Kusel, K., 2012. Origin and fate of acetate in an acidic fen. FEMS Microbiol. Ecol. 81, 339–354.
Heuer, V.B., Pohlman, J.W., Torres, M.E., Elvert, M., Hinrichs, K.-U., 2009. The stable carbon isotope biogeochemistry of acetate and other dissolved carbon species in deep subseafloor sediments at the northern Cascadia Margin. Geochim. Cosmochim. Acta 73, 3323–3336.
Hines, M.E., Duddleston, K.N., Kiene, R.P., 2001. Carbon flow to acetate and C-1 compounds in northern wetlands. Geophys. Res. Lett. 28, 4251–4254.
Hines, M.E., Rooney-Varga, J.N., Duddleston, K.N., Fields, D., Chanton, J.P., 2008. Uncoupling of acetate degradation from methane formation in Alaskan wetlands: connections to vegetation distribution. Glob. Biogeochem. Cycles 22.
Hoehler, T.M., Albert, D.B., Alperin, M.J., Martens, C.S., 1999. Acetogenesis from CO_2 in an anoxic marine sediment. Limnol. Oceanogr. 44, 662–667.
Ise, T., Dunn, A.L., Wofsy, S.C., Moorcroft, P.R., 2008. High sensitivity of peat decomposition to climate change through water-table feedback. Nat. Geosci. 1, 763–766.
Janssens, J.A., 2005. The distribution and accumulation of chemical elements in five peat cores from the mid-continent to the eastern coast of North America. Wetlands 25, 259–278.
Jones, J.G., Simon, B.M., 1985. Interaction of acetogens and methanogens in anaerobic fresh-water sediments. Appl. Environ. Microbiol. 49, 944–948.
Keller, J.K., Bridgham, S.D., 2007. Pathways of anaerobic carbon cycling across an ombrotrophic–minerotrophic peatland gradient. Limnol. Oceanogr. 52, 96.
Keller, J.K., Takagi, K.K., 2013. Solid-phase organic matter reduction regulates anaerobic decomposition in bog soil. Ecosphere 4 art54.
Keller, J.K., Weisenhorn, P.B., Megonigal, J.P., 2009. Humic acids as electron acceptors in wetland decomposition. Soil Biol. Biochem. 41, 1518–1522.
Kotsyurbenko, O., Nozhevnikova, A., Zavarzin, G., 1993. Methanogenic degradation of organic matter by anaerobic bacteria at low temperature. Chemosphere 27, 1745–1761.
Kotsyurbenko, O.R., Glagolev, M.V., Nozhevnikova, A.N., Conrad, R., 2001. Competition between homoacetogenic bacteria and methanogenic archaea for hydrogen at low temperature. FEMS Microbiol. Ecol. 38, 153.
Kotsyurbenko, O.R., Nozhevnikova, A.N., Soloviova, T.I., Zavarzin, G.A., 1996. Methanogenesis at low temperatures by microflora of tundra wetland soil. Antonie Van Leeuwenhoek 69, 75–86.
Leadbetter, J.R., Schmidt, T.M., Graber, J.R., Breznak, J.A., 1999. Acetogenesis from H_2 plus CO_2 by spirochetes from termite guts. Science (New York, N.Y 283, 686–689.
Liu, F., Conrad, R., 2011. Chemolithotrophic acetogenic H_2/CO_2 utilization in Italian rice field soil. ISME J. 5, 1526–1539.
Lovley, D.R., Klug, M.J., 1983. Methanogenesis from methanol and methylamines and acetogenesis from hydrogen and carbon-dioxide in the sediments of a Eutrophic Lake. Appl. Environ. Microbiol. 45, 1310–1315.
Megonigal, J.P., Hine, M.E., Visscher, P.T., 2004. Anaerobic metabolism: linkages to trace gases and aerobic processes. In: Schlesinger, W.H. (Ed.), Biogeochemistry. Elsevier-Pergamon, Oxford, UK, pp. 317–424.
Nie, Y., Liu, H., Du, G., Chen, J., 2007. Enhancement of acetate production by a novel coupled syntrophic acetogenesis with homoacetogenesis process. Process Biochem. 42, 599–605.
Nie, Y., Liu, H., Du, G., Chen, J., 2008. Acetate yield increased by gas circulation and fed-batch fermentation in a novel syntrophic acetogenesis and homoacetogenesis coupling system. Bioresour. Technol. 99, 2989–2995.
Nilsson, M., Öquist, M., 2009. Partitioning litter mass loss into carbon dioxide and methane in peatland ecosystems. In: Baird, A.J. (Ed.), Carbon Cycling in Northern Peatlands. American Geophysical Union, Washington, D.C., pp. 131–144.
Nozhevnikova, A.N., Kotsyurbenko, O.R., Simankova, M.V., 1994. Acetogenesis at low temperature. In: Drake, H.L. (Ed.), Acetogenesis. Chapman and Hall, New York, pp. 416–444.
Phelps, T.J., Zeikus, J.G., 1984. Influence of pH on terminal carbon metabolism in anoxic sediments from a mildly acidic lake. Appl. Environ. Microbiol. 48, 1088–1095.
Reddy, K.R., DeLaune, R.D., 2008. Biogeochemistry of Wetlands: Science and Applications. CRC Press, Boca Raton.
Stumm, W., Morgan, J.J., 1995. Aquatic Chemistry: Chemical Equilibria and Rates in Natural Waters. Wiley, New York.
Tholen, A., Brune, A., 2000. Impact of oxygen on metabolic fluxes and in situ rates of reductive acetogenesis in the hindgut of the wood-feeding termite *Reticulitermes flavipes*. Environ. Microbiol. 2, 436–449.
Vile, M.A., Bridgham, S.D., Wieder, R.K., Novak, M., 2003. Atmospheric sulfur deposition alters pathways of gaseous carbon production in peatlands. Glob. Biogeochem. Cycles 17.
Wieder, R.K., Yavitt, J., Lang, G., 1990. Methane production and sulfate reduction in two Appalachian peatlands. Biogeochemistry 10, 81–104.
Ye, R., Jin, Q., Bohannan, B., Keller, J.K., McAllister, S.A., Bridgham, S.D., 2012. pH controls over anaerobic carbon mineralization, the efficiency of methane production, and methanogenic pathways in peatlands across an ombrotrophic-minerotrophic gradient. Soil Biol. Biochem. 54, 36–47.