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Controls on Carbon Gas Fluxes From a Temperate Forest Soil

Natalie A. White
University of New Hampshire, Durham

Ruth K. Varner
University of New Hampshire, Durham

Clarice R. Perryman
University of New Hampshire, Durham

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Natalie White

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Advisor: Ruth K. Varner

Abstract

Forest soils consume atmospheric methane (CH_4), serving as a major global CH_4 sink that uptake an estimated 22 ± 12 Tg of CH_4 per year. Temperature and soil moisture have been identified as key controls of the microbial consumption of CH_4 in forest soils. Climate-driven warming and changing moisture regimes may impact forest soils' role in the carbon cycle, and recent works suggests that forests could become weaker CH_4 sinks. Long-term monitoring sites can capture these changes, leading to better predictions of CH_4 exchange between the atmosphere and soils under climate change.

This study utilizes a long-term trace gas dataset from College Woods in Durham, NH, USA to track both CH_4 and carbon dioxide (CO_2) fluxes between 1989 and 2021. Between 1989 and 2001 gas fluxes were measured approximately biweekly. In June 2021 the site was re-established, and we collected weekly flux measurements at three collars on a hillslope and three collars in a hollow. Flux measurements collected June-October 2021 in College Woods indicated that average CH_4 uptake in these soils was 3.27 ± 1.16 mg m^{-2} d^{-1}. This is consistent with the 3.35 ± 1.68 mg m^{-2} d^{-1} average uptake rate observed June-October 1989-2001. Average CO_2 emissions from June -August 2021 were 2.86 ± 0.91 µmol m^2s^{-1}, also consistent with the 3.96 ± 2.36 µmol m^2s^{-1} average for 1989-2001. We did not observe a significant change in carbon fluxes across the study period, in contrast with the recent studies suggesting the global forest soil CH_4 sink is decreasing.

1. Introduction

Methane (CH_4) is a greenhouse gas with a warming potential 32 times that of carbon dioxide (CO_2, Holmes et al., 2013). The amount of atmospheric CH_4 has been increasing from

pre-industrial levels around 715 ppb to the most recent estimate in early 2022 of 1909 ppb (Dlugokencky, 2022). Sources of methane to the atmosphere are both naturally occurring and human-caused; wetlands, burning of fossil fuels, and agriculture are the three largest sources (Kirschke et al., 2013). Consumption of atmospheric CH_4 by soil accounts for the second largest sink of CH_4 globally, with forest soils representing about 50% of this sink (Dutaur, 2007). Between 2000-2009, the amount of CH_4 oxidized (consumed) by methane-oxidizing bacteria (methanotrophs) in soils was estimated to be between 26 and 42 Tg yr^{-1} (Kirschke et al., 2013). Methanotrophy occurs in aerated soil environments where oxygen levels are sufficient for methanotrophs, which are obligate aerobes, to oxidize CH_4 to CO_2. Forest soils are an aerobic environment, therefore CH_4 oxidation is the primary methane cycling process present.

The primary controls on CH_4 uptake and release from soils include soil moisture, water table depth, soil temperature, and vegetation (Topp and Pattey, 1997). Soil moisture has been found to be the most influential factor impacting methane uptake rates. There is an optimal soil moisture level where CH_4 uptake is maximized because there is enough water present to facilitate biological activity, but not too much to either physically limit the amount of CH_4 that can enter the soil through diffusion or create anaerobic conditions (Bowden et al., 1998; Curry et al., 2007; Czepiel et al., 1995; Topp and Pattey, 1997). Temperature heavily influences CH_4 uptake as it changes throughout the seasons. As temperatures increase, uptake rates increase because the microbial community is more biologically active (Bowden et al., 1998; Price et al., 2004; Topp and Pattey, 1997). Soil structure and texture class impacts CH_4 uptake as well; coarser soils allow for more diffusion of atmospheric methane into the pore space (Verchot et al., 2000).

Climate change is altering the temperature and precipitation regimes created by large-scale climate phenomena that control CH_4 uptake by forest soils. These large-scale changes

impact the soil properties that directly control CH_4 uptake (Blankinship et al., 2010). Warmer average conditions may increase the ability of forest soils to consume methane because oxidation rates increase with temperature (Bowden et al., 1998), and warmer conditions will cause soils to dry out, opening pore space for increased diffusion rates of atmospheric CH_4 (Liu et al., 2019). However, climate change isn't impacting the whole world in the same way; some areas are getting wetter as they receive more rainfall on average. Campbell et al. (2007) recorded increased precipitation in the United States over the past 50 years. Ni and Groffman (2018) proposed that increasing precipitation was driving a decrease in CH_4 uptake across the US over the past three decades when synthesizing over 300 CH_4 uptake studies. Several other studies recorded an opposite trend wherein the CH_4 uptake rate has been increasing and strengthening the forest soil sink of atmospheric CH_4 (Yu et al., 2017). Climate change is a complex process with several interacting factors, thus it has been difficult to predict how CH_4 uptake rates will respond to potentially synergistic environmental changes (Liu et al., 2019).

This study takes place in College Woods, a temperate forest located in Durham, NH. In 1989 the study site was established and CH_4 uptake and CO_2 emissions were measured through 2001. Crill (1991) analyzed the first 2 years of data and determined that CH_4 uptake here is diffusionally controlled in the late spring, summer, and fall, and that uptake follows a seasonal hysteresis. Soil CH_4 concentrations show that oxidation does not occur in the top organic layer of soil, and that there is an optimal moisture level where oxidation reaches its maximum between 2 and 6 cm (Crill, 1991; Czepiel et al., 1995). This study sought to revisit the College Woods study site and make the same measurements of carbon gas fluxes in order to compare contemporary uptake and release rates to the historical rates. We aimed to understand if carbon gas fluxes at this site are responding to changes in temperature and precipitation trends due to climate change.

2. Materials and Methods

2.1 Site Description

Measurements were conducted in College Woods (43.08 °N, -70.57 °E), on the campus of the University of New Hampshire, Durham, NH, USA. College Woods is approximately 250 acres, consisting of mixed hemlock-hardwood forest, the Oyster River, and the Oyster River Reservoir. The dominant coniferous species in College Woods are Eastern Hemlock (*Tsuga canadensis*), and White Pine (*Pinus strobus*). Dominant hardwood species include Red Oak (*Quercus rubra*), Pin Cherry (*Prunus pennsylvanica*), Red Maple (*Acer rubrum*), Sugar Maple (*Acer saccharum*), and Beech (*Fagus grandifolia*). There was no understory present at this site. The soil at the study site is of the order Inceptisol which is a young, moderately acidic soil that drains well. The 30-year normal maximum temperature in Durham, NH is 14.7ºC and the minimum temperature is 2.7ºC. The 30-year normal annual cumulative precipitation is 1151.9 mm (NOAA National Centers for Environmenntal Information).

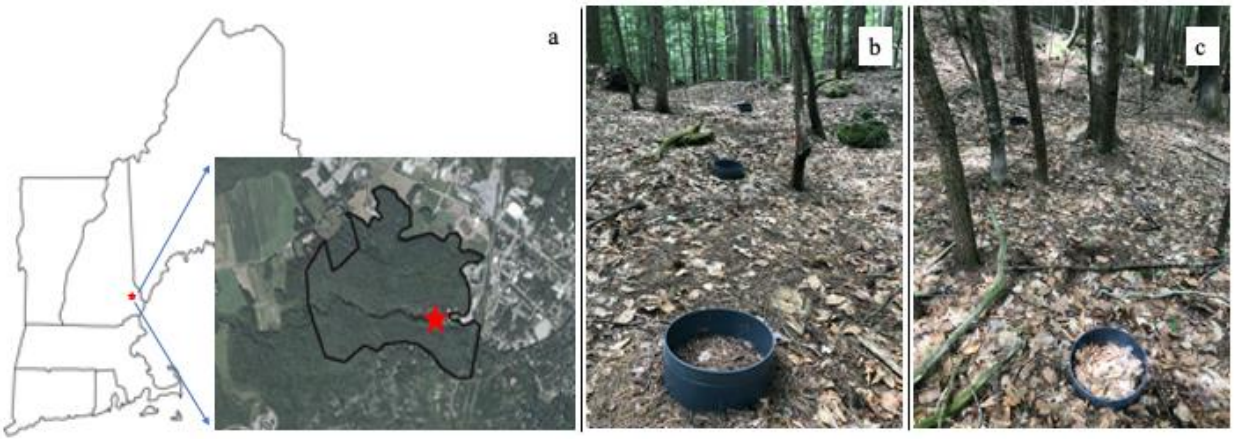

Figure 1: a) College Woods study site located on the campus of the University of New Hampshire, Durham, NH, USA, 43.135 °N, -70.944 °E. b) Slope transect established in 2021 with three collars. c) Hollow transect established in 2021 with three collars.

During a thaw in January 1989 two aluminum collars (0.397 m^2) were cut into the soil in College Woods with one on a slope side and the second in a hollow (Crill, 1991). Methane and CO_2 flux measurements began the next month and continued through 2001 approximately every two weeks. In the summer of 2021, we re-established the study site and collected measurements from a total of six static flux chambers. Three circular collars (diameter 32 cm) were installed on a transect following the downward slope of the hill the study site is on, and the other three collars were installed along a transect of a hollow area on the hillside. The decision to install collars on both a slope and in a hollow followed from the experimental design of the Crill (1991) study which published the initial data from this site. The soil around each chamber was cut with a knife to allow the collar to be pushed 5 centimeters into the soil. Once installed, the height of each collar was measured four times around the collar and averaged to calculate the volume of the collar with the equation $volume = 3.14 \times 16^2 \times h \times 0.001$.

2.2 Environmental Conditions- Temperature and Soil Moisture

For all gas flux measurements, air and soil temperature measurements were collected in tandem with soil gas flux measurements. Air temperature, soil surface temperature, and soil temperature at 5 cm and 10 cm were measured adjacent to each flux chamber with a thermometer (Gilson MA-118 Taylor 9878E Pocket Digital Thermometer). In 2021, soil moisture was also measured at 3 and 10 cm in the soil adjacent to each flux chamber with a soil moisture probe (Campbell Scientific HydroSense II). Precipitation data from a weather station in Durham, NH were

retrieved from the NOAA National Centers for Environmental Information for all months of the year between 1989 and 2021 to provide cumulative monthly precipitation.

2.3 CH_4 and CO_2 fluxes

The initial study period (1989-2001) used the static chamber flux method to measure both CH_4 and CO_2 fluxes. The two aluminum collars were fitted with a flux chamber (0.152 m^3) and sealed by filling the groove of the collar with water. The chambers were dark, prohibiting photosynthesis from taking place. As such, the CO2 flux measurements represent net (autotrophic and heterotrophic) soil respiration, not net ecosystem exchange (CO_2 uptake – net respiration). Inside the chamber was a battery-operated fan to mix the headspace and a thermistor for measuring the enclosed air temperature. Fluxes were measured by taking 60 ml samples of the headspace at 4-minute intervals over a 20-minute period, with the first sample taken at minute four. Polypropylene syringes with siliconized polypropylene plungers and polycarbonate/nylon or polyethylene/nylon three-way stopcocks were used to draw out the headspace samples. Headspace gas samples were equilibrated to room temperature in the lab for two hours and analyzed within five hours after collection.

Throughout the growing season of 2021, June-October, the static chamber method was used again for weekly CH_4 and CO_2 flux measurements. Collar lids were placed on the collars to seal off air exchange, but the chambers were not equipped with a battery-operated fan. To ensure the headspace was well-mixed, the syringe was used to pump the headspace air 10 times before drawing the sample. The methods of Crill (1991) were followed with the addition of one 60 ml sample of ambient air taken at each collar before flux measurements began. The headspace gas

samples were stored in their syringes and placed in a refrigerator for no more than 24 hours before they were processed in the lab.

Headspace CH_4 concentrations were measured with a gas chromatograph equipped with a flame ionization detector (GC-FID, Shimadzu GC-8A). The GC-FID was run with a 50°C column temperature, 130 °C detector and injector temperature, and ultra-high purity nitrogen gas as the carrier gas flowing at a rate of 30 mL per minute through a 2-m 1/8-inch o.d. stainless steel packed column (HayeSepQ 100/120). Twenty mL samples of standard gas at 2.006 ppm CH_4 were used to calibrate the GC-FID. This breathing air cylinder standard was calibrated against a standard from NOAA's Earth System Research Laboratory's Global Monitoring Division's Carbon Cycle Greenhouse Gases Group. Each collar headspace sample was run twice and the average CH_4 concentration was used for the final flux calculation with the equation: $\frac{mgCH_4}{m^2 day} =$

$$\frac{\Delta ppm}{min} \times \frac{P}{RT} \times \frac{V_c}{A_c} \times \frac{1440 min}{day} \times \frac{16 molCH_4}{1\ g} \times \frac{1 mg}{1000 \mu g} \times \frac{1 m^3}{1000 L}$$

In this equation Δppm/min is the change in methane mixing ratio over the chamber closure period calculated from a linear fit, P is pressure (atm), R is the gas constant (8.202^{-5} m^3atm/molK), V_c is the chamber volume (L), and A_c is the chamber area (m^2). Precision for the calculation of the fluxes with this equation was ±0.5%. Samples were rejected if they did not fit the 95% confidence interval with respect to the coefficient of determination: n = 3 (r^2=0.95), n=4 (r^2= 0.87), and n = 5 (r^2= 0.75). This calculation represents the slope of the linear regression of the CH_4 concentration over the 20-minute sampling period. The soils in College Woods predominantly consume methane, so the fluxes are written as negative numbers which represent a loss of methane from the atmospheric pool.

Carbon dioxide concentrations were measured with an infrared gas analyzer (LI-COR IRGA). An instrument response curve was generated by injecting two replicates of five volumes of standard gas (Northeast Airgas, 980.9 ppmv): 1, 3, 5, 7, and 10 ml. A linear regression of the standards indicated a slope and y-intercept. Two chamber headspace samples of 3 mL each were injected into the IRGA, and the CO_2 concentrations were determined with the following equation:

$$CO_2\ concentration\ (ug\ ml^{-1}\ CO_2) = Sample\ integrated\ area \times m + b$$

where m is the slope and b is the y-intercept of the standard curve. A linear regression determining CO_2 concentration change over time was calculated and used to calculate CO_2 flux with the following equation:

$$\frac{\mu mol\ CO_2}{m^2 s} = \Delta CO_2 \times \frac{V_c}{A_c} \times \frac{1\ min}{60 s} \times \frac{1000\ ml}{L} \times \frac{1\ mol\ CO_2}{44\ g\ CO_2}$$

Where Δ CO_2 is the change in CO_2 concentration per minute.

2.4 Soil poregas CH_4 and CO_2 concentrations

Soil poregas samples were collected weekly between July 1st and October 13th 2021 on the same dates that the static chamber fluxes were measured. Two sets of stainless-steel sippers with a 3 mm inner diameter were inserted into the soil adjacent to both the slope and hollow transect. Each set of four sippers was inserted to depths of 2 cm, 6 cm, 11 cm, and 15 cm, following the design of the Crill (1991) study. Polypropylene syringes equipped with three-way stopcocks were used to pull a 50 mL sample from the sipper. Samples were stored in a refrigerator for no more than 24 hours until they were processed in the lab. Analysis of poregas samples was

conducted on the GC-FID following the same process as described for the static chamber flux measurements (See section 2.2).

2.5 Statistics and Data Analysis

A combination of Excel, R v.4.0.3, and JMP were used for statistical analysis. Data were cleaned in the tidyverse (Wickham et al., 2021) and lubridate (Grolemund and Wickham, 2011) R packages, with data visualization done in ggplot2 (Wickham, 2016). I performed a one-way ANOVA to determine if there were differences in the environmental characteristics of the slope and hollow sites. T-tests were used to determine if CH_4 and CO_2 fluxes between sites varied. Linear regressions were used to assess if there were significant temporal trends in annual temperatures and precipitation, as well as for both CH_4 and CO_2 fluxes over time. The confidence intervals for each of these statistical tests was 95%.

Data was analyzed in four distinct groupings based on year and month. All data included January-December 1989-2001 and June-October 2021. Growing season data included June-October 1989-2001, and 2021. Historical data included January-December 1989-2001, and contemporary data included June-October 2021.

3. Results

3.1 Environmental Conditions – Temperature, Precipitation, and Soil Moisture

Cumulative annual precipitation and daily average precipitation in Durham, NH between 1989-2021 had no significant temporal trend (R^2 = 0.0016 and 0.0098, respectively; Fig. 3). However,

rainfall in July of 2021 (323.1 mm) was almost four times the average rainfall in July throughout the whole 1989-2001 period (84.3 ± 44.6 mm), and precipitation for August, September, and October of 2021 was higher than the historical average precipitation for these months (Table 1). Mean annual maximum air temperatures revealed no significant change in temperature in Durham, NH over the 32-year period ($R^2 = 0.03$, $p = 0.17$), while mean annual minimum air temperatures slightly increased ($R^2 = 0.10$, $p = 0.04$).

Overall, temperature and moisture conditions across the slope and hollow transects were similar in 2021. Air temperature ($p = 0.82$), soil surface temperature ($p = 0.74$), and soil temperature at 3 and 10 cm depth ($p = 0.47$ and 0.66, respectively) all fell within the same ranges for the two transects. At 3 cm deep, soil moisture was not significantly different in the two transects ($p =$

0.08). However, at 10 cm the soil in the hollow transect was slightly drier than in the slope transect; mean soil moisture of 10.49 ± 4.87 % and 13.13 ± 6.26%, respectively (p = 0.04).

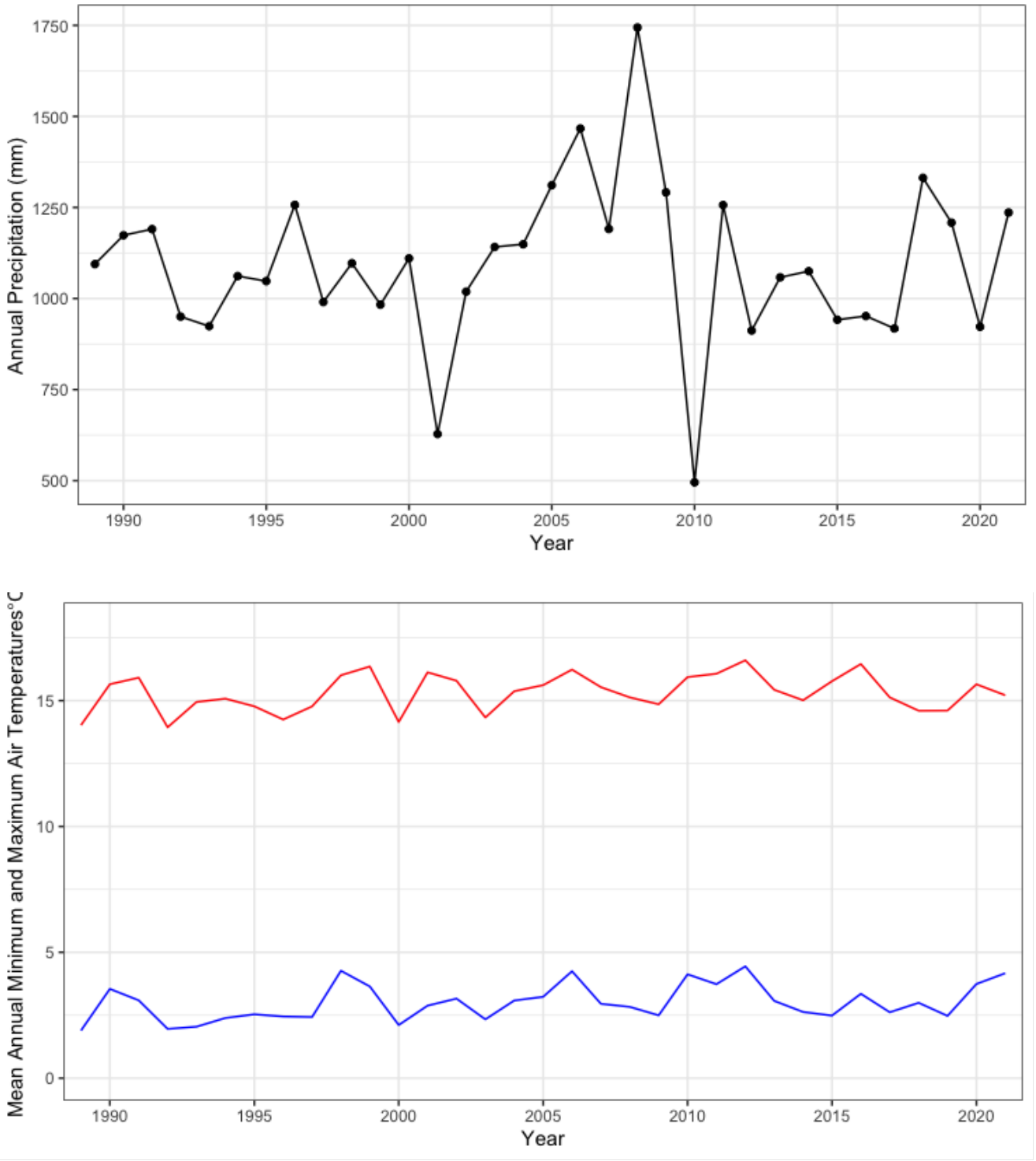

Figure 2: A) Cumulative annual precipitation plotted over the 32-year period between 1989 and 2021. B) Annual

minimum and maximum air temperatures in Durham, NH between 1989-2001. Data from NOAA National Centers for Environmental Information.

Table 1: Precipitation in Durham, NH in 2021. Data retrieved from the NOAA Centers for Environmental Information.

Month	Cumulative (mm)	Daily mean (mm)	Standard deviation (mm)
January	53.9	1.74	6.42
February	71.6	2.56	5.52
March	55	1.77	4.77
April	73.4	2.45	5.66
May	85.2	2.75	7.31
June	28.4	0.95	2.00
July	323	10.42	18.70
August	95.7	3.09	6.79
September	107.7	3.59	12.02
October	166.9	5.38	16.03
November	68.8	2.29	8.26
December	106.5	3.44	5.43

3.2 Emissions and Soil Gas Concentrations Across Slope and Hollow

When separated by landscape position, CH_4 uptake during the growing season (June-October) was significantly lower in the hollow (-2.77 ± 1.233 mg m^2d^{-1}) than the slope (-3.91 ± 1.71 mg

m^2d^{-1}, $p < 0.001$). Carbon dioxide emissions were not significantly different between the hollow (4.23 ± 2.38 µmol m^2s^{-1}) and slope (3.78 ± 3.21 µmol m^2s^{-1}) transects ($p = 0.065$).

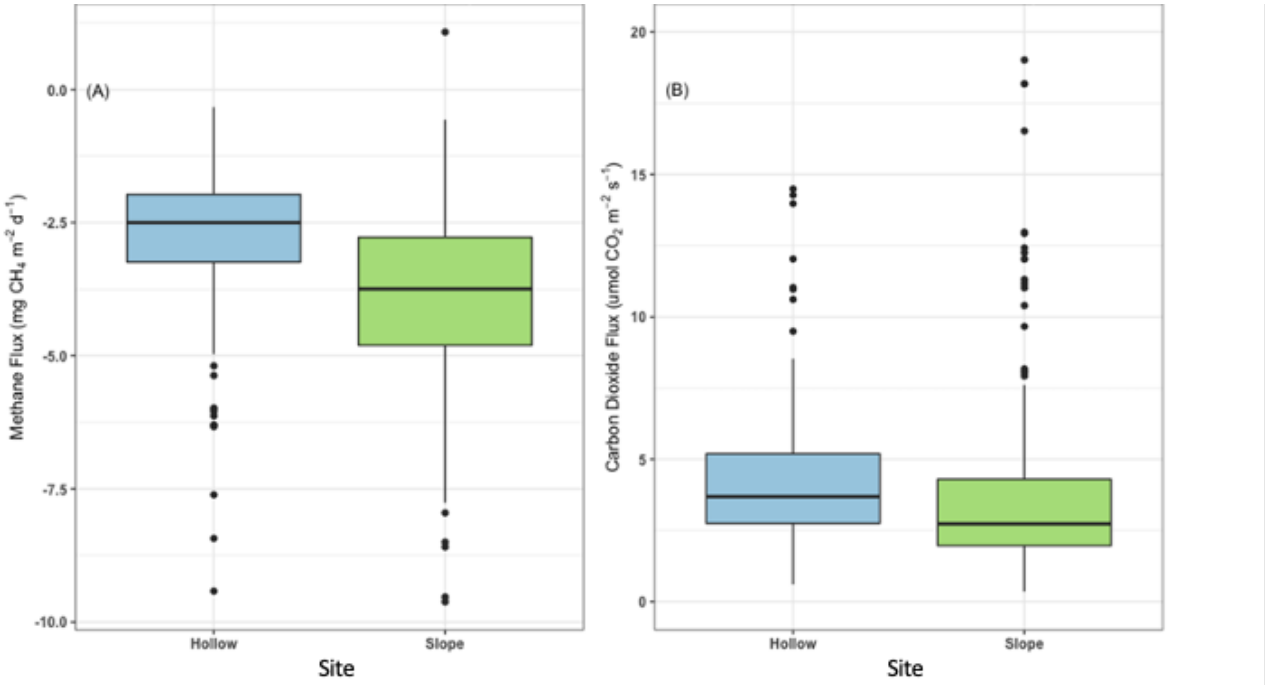

Figure 3: Methane and carbon dioxide fluxes separated by hollow versus slope transect. Calculated with growing season measurements from all studied years. Within each box, black horizontal lines represent median values, and the upper and lower bounds represent the 25th and 75th percentiles.

Soil poregas measurements made during the 2021 growing season indicate that CH_4 concentrations are higher through the whole depth gradient in the hollow transect ($p = 0.042$). At 2 cm the hollow averaged 1.2 ppmv while the slope averaged 1.12 ppmv, at 6 cm the hollow averaged 0.90 ppmv while the slope averaged 0.88 ppmv, at 11 cm the hollow averaged 0.71 ppmv while the slope averaged 0.28 ppmv, and at 15 cm the hollow averaged 0.50 ppmv while the slope averaged 0.25 ppmv. The largest decrease in soil gas CH_4 concentration, and therefore the most CH_4 oxidation, occurs between 6 and 11 cm for all observed months. On the slope in July and August the CH_4 concentration is lower than in the hollow at all depths, yet in September

and October the pattern changes and the CH_4 concentration at 2 and 6 cm deep on the slope increases above the concentration in the hollow for the respective depths. August exhibits a pattern of increasing CH_4 concentration between 2 and 6 cm, whereas all other months show a decreasing CH_4 concentration with depth. August is the only month that does not have the highest CH_4 concentration at the shallowest depth. Carbon dioxide concentrations in the poregas increased with depth and did not show any significant differences between the sites (Appendix Fig. 1).

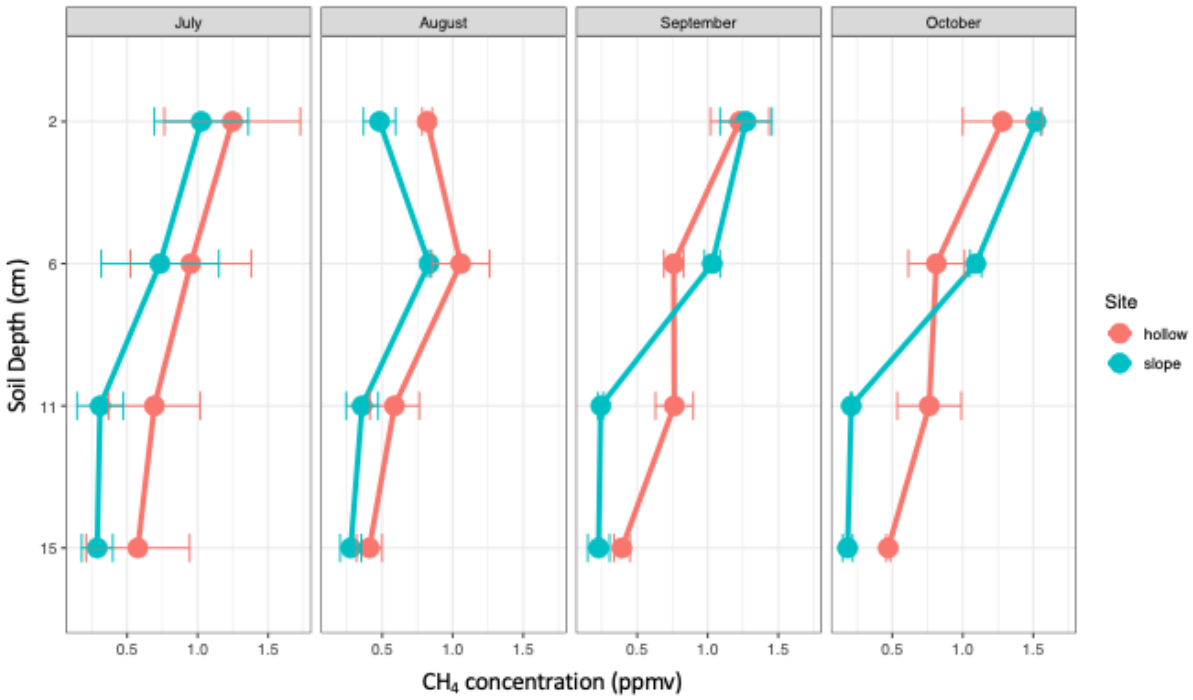

Figure 4: Soil profile concentrations of CH_4 and CO_2 in 2021. Each point represents the monthly mean value for soil gas CH_4 concentration at each depth, with error bars showing ± one standard deviation of the mean.

3.3 Seasonal and Interannual Trends in CO2 and CH4 Fluxes

We observed methane uptake for the majority of measurements (n = 649), with -9.62 mg CH_4 m^2d^{-1} on June 23, 1999 being the largest uptake observed. There were few measurements when

the soil emitted methane (n=18, < 3% of all measurements), most of which occurred either in winter or spring. Figure 4 depicts the seasonality of methane uptake; maximum uptake is reached in the warmest months of the year while uptake is lowest in the winter months. Over the course of the initial study period from 1989-2001 the average annual CH_4 uptake was -2.90 ± 1.58 mg m^2d^{-1}. The growing season average uptake throughout this period, defined as June through October, is -3.42 ± 1.34 mg m^2d^{-1}. Measurements in 2021 were only made June through October, resulting in an average growing season CH_4 uptake of –3.27 ± 1.16 mg m^2d^{-1}.

Carbon dioxide flux measurements (n = 606) indicated that the soil in College Woods was a net source of CO_2 to the atmosphere, aside from occasions of winter freeze when soil respiration was not taking place. Average annual CO_2 emissions between 1989-2001 was 3.37 ± 2.23 µmol m^2s^{-1} , while the growing season average was 4.10 ± 1.42 µmol m^2s^{-1}. In 2021, the growing season average CO_2 emissions (only measured June – August) were 2.86 ± 0.91 µmol m^2s^{-1}. The

seasonal variation in CO_2 emissions is visible in Figure 5, showing a similar trend to CH_4 as the size of the flux increases during the warm months.

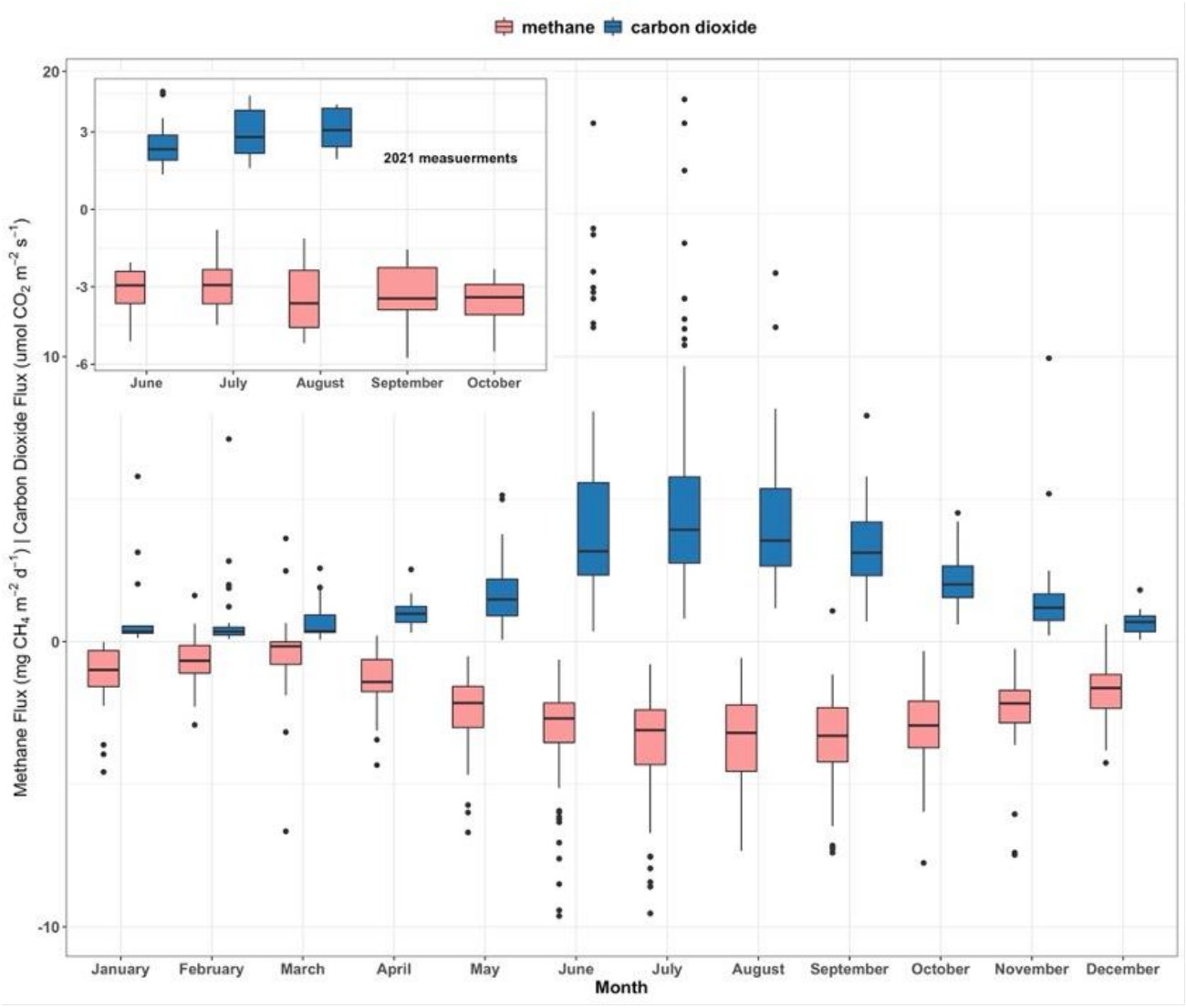

Figure 5: Timeseries showing monthly methane (n =649) and carbon dioxide (n = 606) fluxes. The boxplot includes

both historical data (1989 - 2001) and data from 2021 (in inset). Within each box, black horizontal lines represent median values, and the upper and lower bounds represent the 25th and 75th percentiles.

Both CH_4 and CO_2 fluxes followed seasonal patterns dictated heavily, but not solely, by temperature. Figure 6 plots the fluxes of both gases in relation to air temperature throughout the year.

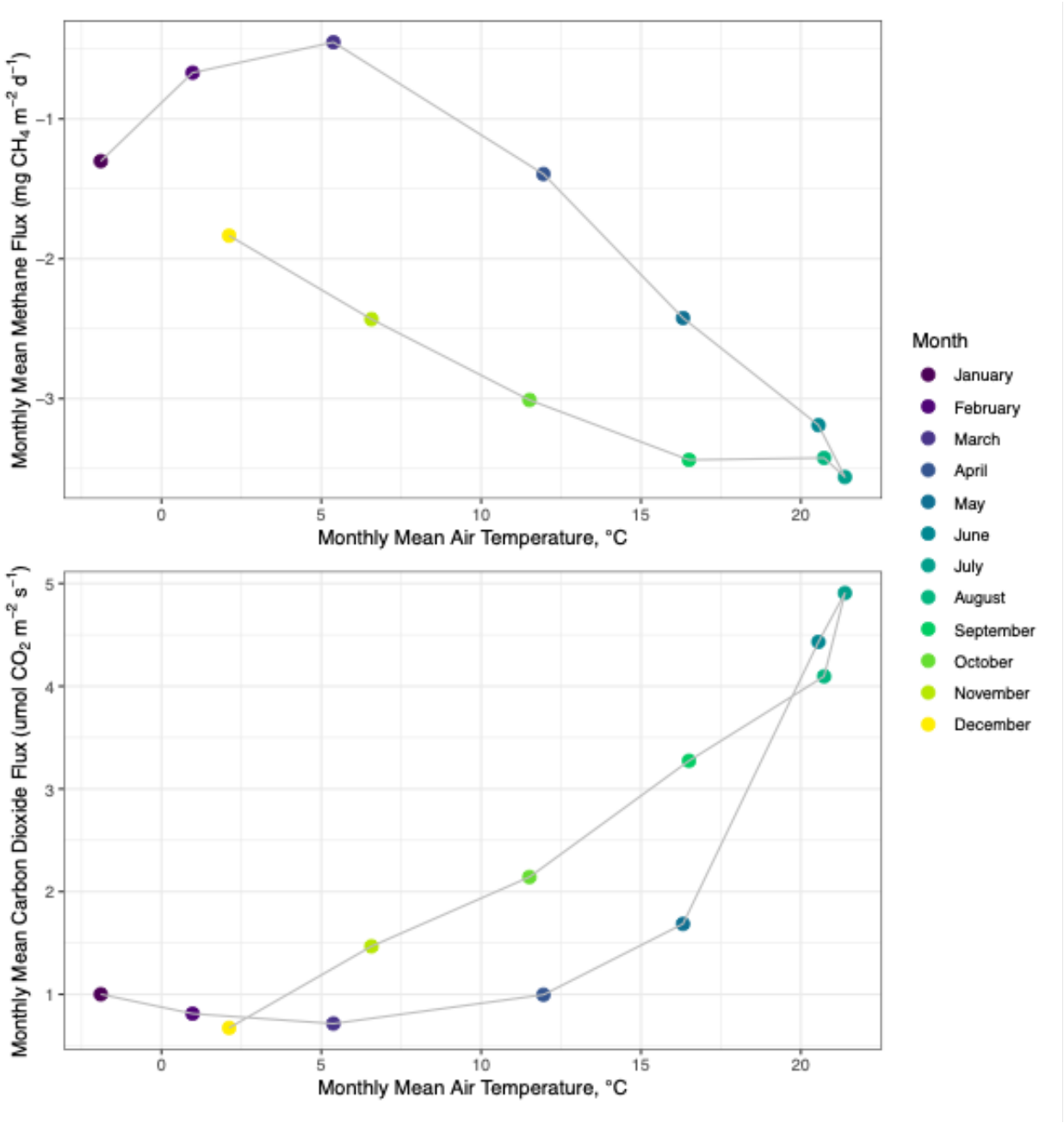

Figure 6: Hysteresis of monthly mean methane and carbon dioxide fluxes by monthly mean air temperature throughout the year. Calculated with all data from all study years.

On the interannual timescale, the data collected in this study does not show a significant change in CH_4 or CO_2 fluxes over time. In Figure 7, the blue trendline is calculated from this study's data and shows that year does not explain significant variance in the amounts of CH_4 uptake (R^2

= 0.098, p = 0.15). Both lines imposed on top of this study's data are adapted from Ni and Groffman (2018). The dotted line represents the linear regression of global CH_4 soil uptake between 30-60°N from 1987-2016, and the dashed line is the CH_4 uptake in Hubbard Brook, NH from 2002-2015. Both datasets show significant decreases in CH_4 uptake over their respective time period.

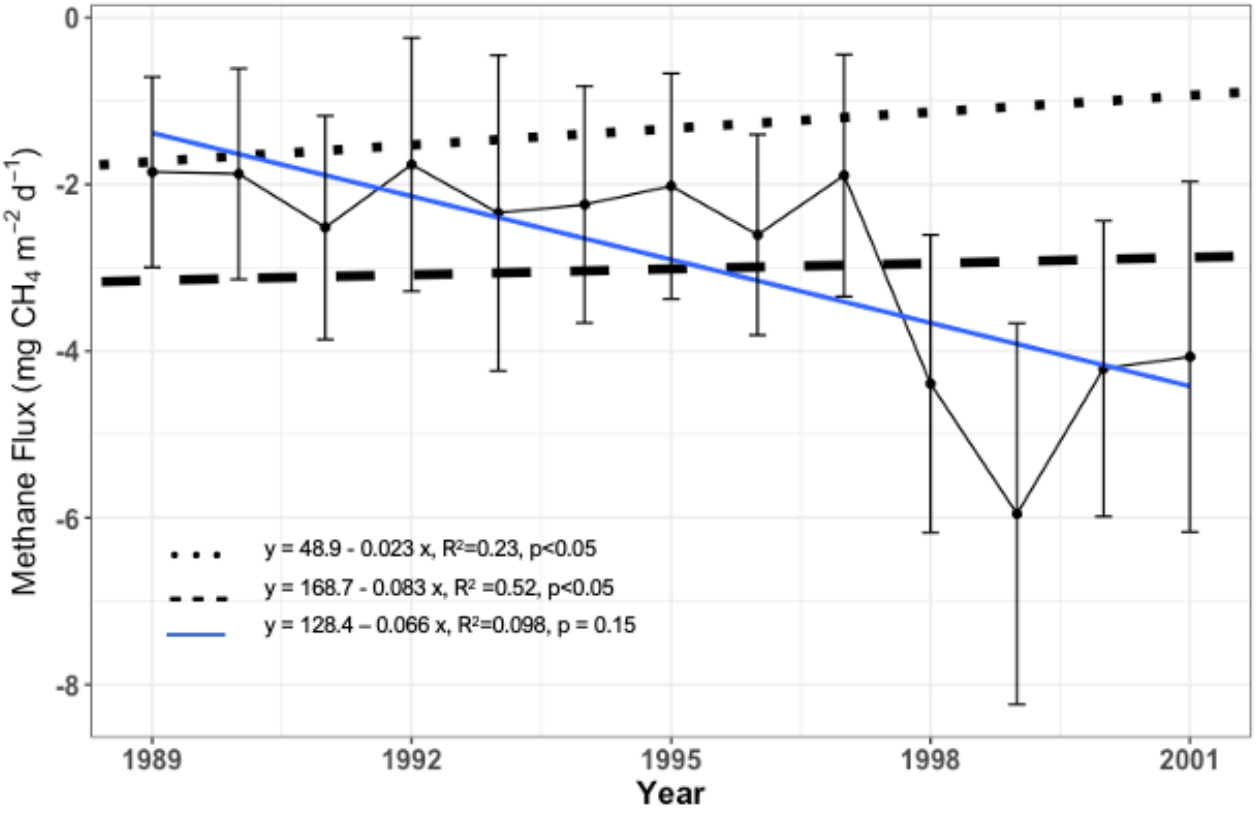

Figure 7: Timeseries graph of CH_4 soil uptake between 1989-2001. Data points represent annual mean CH_4 uptake, with error bars showing ± one standard deviation of the mean. The blue line is the trendline calculated from this study's data (R^2=0.098, p = 0.15). The dotted line represents the linear regression of global CH_4 soil uptake between 30-60°N from 1987-2016, and the dashed line is the CH_4 uptake in Hubbard Brook from 2002-2015 (Ni and Groffman, 2018).

The time series of CO_2 emissions between 1989-2001 also shows no significant temporal trend in forest soil C emissions ($R^2 = 0.06$, $p = 0.396$).

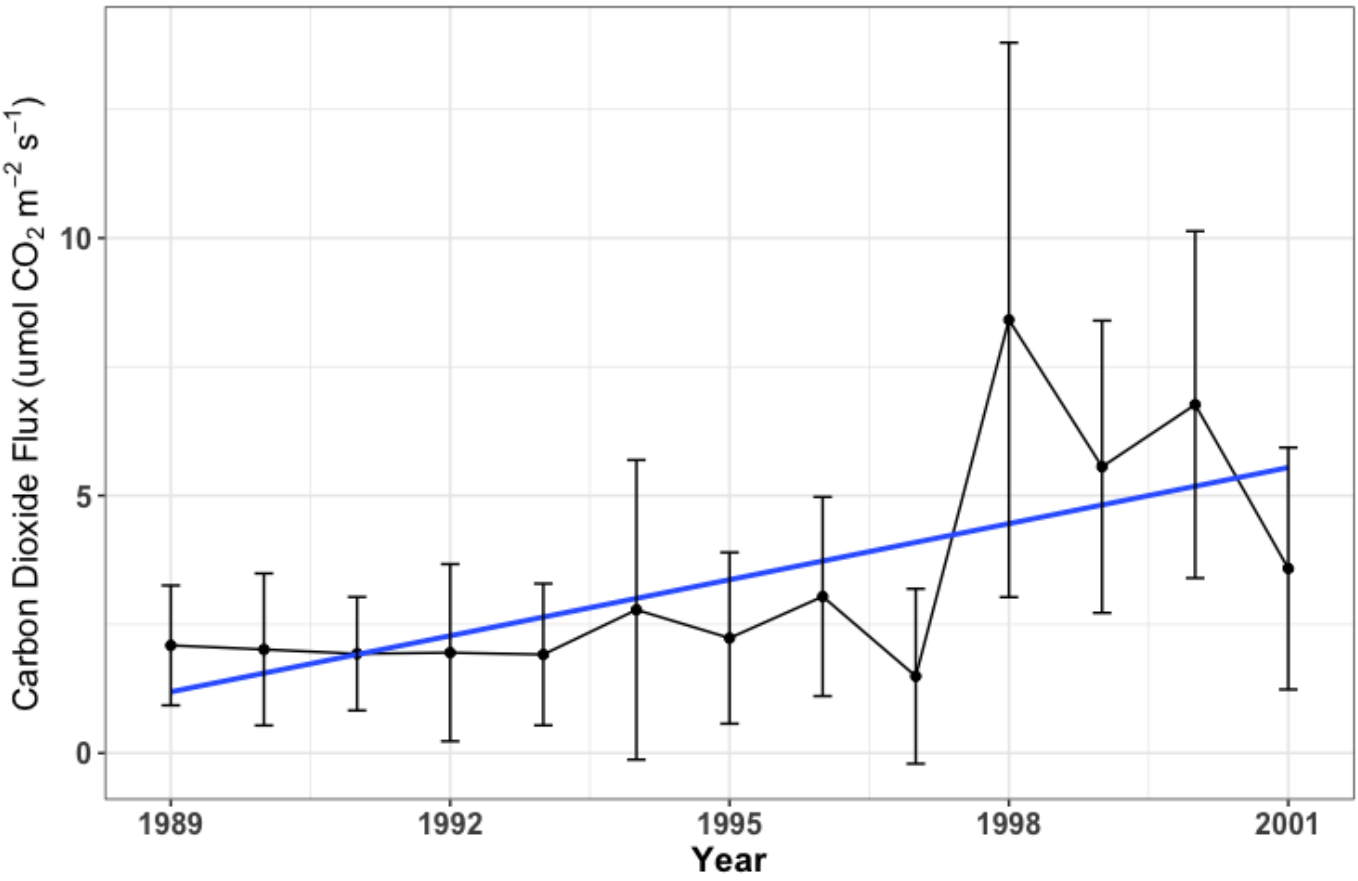

Figure 8: Timeseries graph of CO_2 soil emissions between 1989-2001, calculated with data collected by the Trace Gas Biogeochemistry lab. Data points represent annual mean CO_2 emissions, with error bars showing ± one standard deviation of the mean.

4. Discussion

4.1 Spatial Variation of CH_4 and CO_2 Fluxes

On the small spatial scale of this study, few differences were observed between the fluxes of carbon gasses in the slope and hollow chambers. Methane uptake on the slope was slightly higher than in the hollow on average for all months of all study years (Figure 3). The

hypothesized differences between the slope and the hollow were that the slope would have slightly drier soil as precipitation would run off downhill and drain through the soil, whereas rainfall would collect more easily in the hollow and drain more slowly, therefore influencing CH_4 uptake through differences in soil moisture conditions. However, our observations of the two landscape positions found the only significant difference in soil moisture between the two sites to be that at 10 cm deep the hollow had drier soil. Drier soil is more conducive to CH_4 oxidation because the open pore spaces allow diffusion of atmospheric gases into the soil, which is the primary mechanism by which CH_4 enters the soil and is oxidized. Soil gas profiles indicate that the most CH_4 oxidation occurs between 6 and 11 cm. Thus, drier soils below 10 cm would not be impacting the soil depth at which the most oxidation is occurring.

In their 1997 laboratory study, Bowden et al. found that maximum CH_4 uptake occurred when around 60% of the pore space was filled with water (WFPS). This optimal WFPS finding was in line with the results of another laboratory study, which found that optimal WFPS was between 50-70% (Nesbit and Breitenbeck 1992). Methane uptake increased as the soil moisture increased from 0-60%, peaked between 60-70%, and then began decreasing after 70%. Soils in College Woods have been found to have optimal soil moisture levels as well. Czepiel et al. (1995) used incubation experiments to observe CH_4 oxidation in the soils of College Woods in relation to soil moisture. Optimal uptake was found to be at moisture levels between 18-33% in grassy areas and between 30-51% in wooded areas. During the 2021 growing season the hollow averaged a moisture content of $10.49 \pm 4.87\%$, while the slope averaged $13.13 \pm 6.26\%$. The slope exhibited more CH_4 uptake throughout the season with a slightly higher soil moisture content. In the context of the optimal soil moisture levels found by Czepiel, this higher moisture content corresponds to the higher CH_4 uptake level. It is important to note that although the moisture

levels at 10 cm between the slope and hollow in 2021 were significantly different, the difference is a small percentage and thus is unlikely to significantly impact CH_4 uptake rates.

Soil gas profiles between the two sites showed clearer differences than the gas fluxes between sites. On average, soil CH_4 concentration in the hollow was higher than on the slope. In July and August the CH_4 concentration in the hollow was higher at all depths, then in September and October the CH_4 concentration on the slope increased above the hollow concentrations for the two shallowest depths (Figure 4). The pattern of soil CH_4 concentrations in August 2021 differ slightly than the three other observed months; as the depth increases from 2 to 6 cm the CH_4 concentration increases as well. This is the opposite pattern of the decreasing concentrations with depth that every other month and depth range exhibited. A large rain event that caused precipitation to reside in the top ~5 cm layer of soil could help explain why little CH_4 was able to diffuse into the soil, yet the soil moisture at 3 cm recorded in August was 8.83 ± 3.32% in the hollow and 6.00 ±1.2% in the slope – both within the typical range of moisture levels seen at each site.

On the slope, the most oxidation in all four months occurred between 6 and 11 cm. This depth has been found to be the typical range at which CH_4 oxidation primarily occurs (Price et al, 2004). Yet in the hollow, Figure 4 illustrates that the most oxidation occurs between 6-11 cm in July and August but switches to between 2-6 cm in September and October. This was an unexpected finding, as little to no methanotrophic activity occurs in the organic layer (Price et al., 2004). Changes in soil moisture throughout the season could shift the depth of CH_4 oxidation. Despite September and October receiving less precipitation than July which caused the soil at the

whole study site to be drier, the slope site did not experience this same change in oxidation depth.

4.2 Seasonality of CH_4 and CO_2 Fluxes

The change in fluxes throughout the year are driven by seasonal differences in temperature and precipitation that Durham, NH experiences due to its location along climatic gradients. Figure 5 illustrates the yearly pattern of changing CH_4 and CO_2 fluxes for both the total historical data and a separate inset of this year's collected data. The warmest months in Durham are June, July, and August where the temperature averages 20.7 ± 3.9°C. These three months experience the highest CH_4 uptake rates as well. The consistency of the CH_4 uptake being around –3.00 mg m^{-2} d^{-1} in

the warmest months during both the historical and contemporary measurements supports that this site is influenced by long-term, recurring climatic patterns.

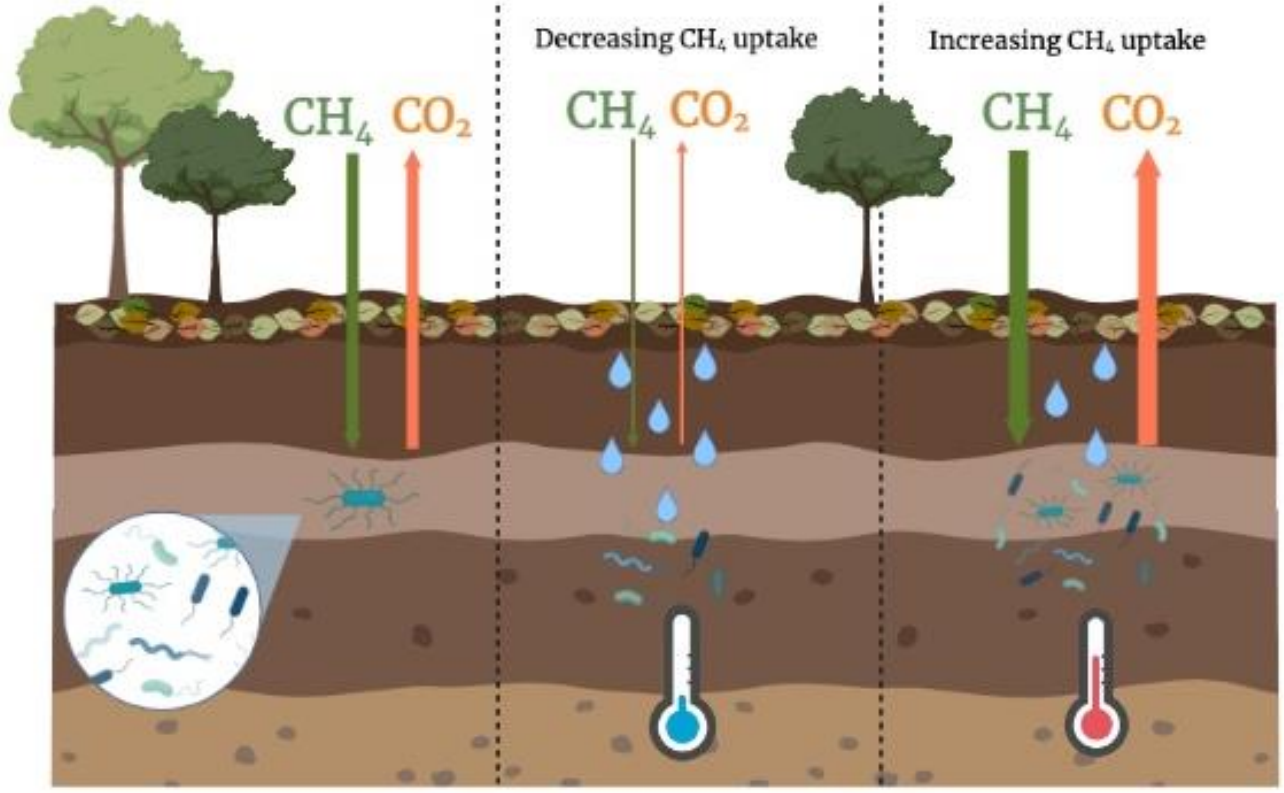

Figure 9: Conceptual model of a temperate forest system depicting the factors that affect rates of CH_4 uptake and CO_2 emissions. Created in Biorender.

During the winter in Durham, NH the temperature drops below freezing and snow and ice are present at the study site. Methane uptake at this time of year drops because atmospheric gases are not diffusing into the soil as easily, and the cold temperatures decrease the activity of the methanotrophs (Crill, 1991). When there is little snowfall, and the soil is exposed to cold temperatures it will likely freeze. In these instances where the soil freezes, the methane uptake stops because the microbes are not biologically active. Both Figures 5 and 6 pinpoint March as the month experiencing the least CH_4 uptake at -0.45 ± 1.70 mg/m^{2}d^{-1}, while February and

January follow behind as the months with the second and third least amount of CH_4 uptake, -0.67 ± 0.88 mg/m^{2}d^{-1} and -1.30 ± 1.29 mg/m^{2}d^{-1} respectively. As Figure 9 demonstrates, cold temperatures decrease the ability of a soil to take up CH_4.

Although the same pattern of increasing CH_4 uptake during warm months and decreasing CH_4 uptake during cold months is observed at this site every year, Figure 6 indicates that the temperature change by season is not the only driving force behind these changing fluxes. Both CH_4 and CO_2 fluxes show substantial seasonal hysteresis between C gas fluxes and air temperature, suggesting that the temperature sensitivity of gas fluxes varies across the year. Months in the fall with average air temperatures similar to those of spring months exhibit more CH_4 uptake. This is due to CH_4 uptake being physically limited by diffusion in the early spring. Spring snowmelt contributes to muddy soil conditions in the early spring which fills the pore space of the soil with water, limiting the amount of atmospheric gas exchange that can occur between the atmosphere and soil (Bowden et al, 1998). Seasonal changes involve interactions between temperature and moisture, forcing soil CH_4 uptake to respond along both gradients (Bowden et al, 1998). As the soil dries over the season, temperatures warm, and vegetation begins to grow, there is more available pore space for atmospheric CH_4 to move into (Castaldi and Fierro, 2005). As a result, more soil CH_4 uptake occurs. The physical limitations of soil pore space, as determined by moisture levels, result in less CH_4 uptake in the spring than in the fall despite the average monthly air temperatures being similar. Methane uptake at this site in 1989 and 1990 exhibited the same seasonal pattern where CH_4 uptake increased in the fall relative to spring uptake levels, despite having the same average monthly air temperature (Crill, 1991).

The hysteresis of CO_2 in Figure 6 shows a similar pattern to that of CH_4 where emissions are highest in the three warmest months (June, July, and August), and lowest in the cold months. The

largest increase in CO_2 emissions occurs between May and June, jumping from a monthly average of 1.69 ± 1.08 μmol/m^2s^{-1} to 4.643 ± 3.46 μmol/m^2s^{-1}. After this increase, the emissions levels remain higher throughout the fall despite monthly average air temperatures being comparable to spring temperatures. 1989 and 1990 data from this site follow the same shape as the hysteresis in Figure 6; the largest increase in monthly average CO2 emissions occurred between May and June, with emissions maximizing in July (Crill, 1991). The driving forces behind this phenomenon include the accumulation of soil organic matter, the growth of the microbial communities performing heterotrophic respiration, and phenological changes in the rhizosphere over the season (Lloyd and Taylor, 1994). As the number of microbes performing decomposition increases along with the amount of material available to be decomposed, CO_2 emissions in this system increase (Aronson et al. 2010). Temperature and soil moisture are essential in this process because they facilitate the other changes that result from seasonality.

4.3 Change Over Time

Average annual precipitation in College Woods has not significantly changed since this study began in 1989 (Figure 2A). Average daily maximum and minimum temperatures have not significantly changed over this time period either (Figure 2B). Therefore, any variation in annual CH_4 uptake between the beginning of this study and the most recent measurements cannot be explained by these two factors alone.

Cumulative precipitation in 1989 was 1094.5 mm and in 2021 it was 1236.1 mm, with respective growing season average CH_4 uptake rates of –2.47 ± 0.35 mg m^2d^{-1} and –3.27 ± 0.27 mg m^2d^{-1}. Neither the annual precipitation sums, nor the annual average CH_4 uptake rates have changed significantly between these two years, indicating that any change in the CH_4 uptake rate is not

fully explained by the change in precipitation. However, increased soil moisture resulting from increased precipitation does impact the ability of a soil to consume CH_4 (Bowden et al. 1998). July 2021 received heavy rainfall, totaling 323.0 mm. This is more than three times the average rainfall total for July 1989-2001 of 101.3 mm. The CH_4 soil uptake of July 2021 was –2.87 ± 1.02 mg m^2d^{-1} compared to the –3.77 ± 1.88 mg m^2d^{-1} average uptake in July from all other years included in this study. Decreased CH_4 uptake in July of 2021 as compared to July in every other year of the study can reasonably be attributed to this large quantity of precipitation that the study site received, due to the relationships between soil moisture, diffusion, and CH_4 uptake described in the previous section.

Figure 7 plots the trend of CH_4 uptake throughout this study and compares it to a synthesis of over 300 studies reporting on CH_4 uptake rates. This synthesis found significant decreases in CH_4 uptake rates over the past three decades both globally between 30-60 degrees North and at two sites: one in Baltimore and one nearby in NH – Hubbard Brook (Ni and Groffman 2018). In contrast, College Woods did not exhibit a significant decrease in CH_4 uptake between 1989 and today, and there is an insignificant increasing trend in CH_4 uptake in the 1989 – 2001 data from College Woods. Ni and Groffman (2018) found that increases in soil moisture were underlying the decreases in CH_4 uptake at Hubbard Brook, as the site has experienced increasing soil moisture over the past 14 years due to the trend of increasing precipitation in the United States (Groffman et al., 2012). In the analysis of College Woods data, precipitation was used as a proxy for soil moisture. However, no such significant increase in precipitation was observed at this site.

No significant changes in CO_2 release from College Woods have been recorded over the 1989-2001 period. As soil respiration is primarily controlled by temperature and moisture, significant temporal changes of these two factors are expected for there to be significant change in CO_2

emissions. Lloyd and Taylor (1994) conclude that although increases in temperature will decrease the activation energy for respiration, warming temperatures will have an unknown effect on the size of the soil carbon pool. As a result, predictions of change in soil respiration in response to warming temperatures cannot accurately be made.

5. Conclusions

Measurements of CH_4 uptake and CO_2 emissions in College Woods between June and October 2021 are consistent with the same flux measurements made June-October 1989-2001 at this site, suggesting that carbon gas cycling is controlled by recurring and long-term climatic trends. Unlike several of the studies synthesized by Ni and Groffman (2018), annual cumulative precipitation and average annual maximum temperatures did not significantly change between 1989-2021. Perhaps then, it is the changing length of the seasons that alters CH_4 uptake and CO_2 emissions under climate change, not strictly the annual temperature and precipitation averages.

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Appendix

Table 1: Monthly Averages for 1989-2001 and 2021.

Year	Month	CH_4 mg m^2d^{-1}	CH_4 SD	CO_2 µmol/m^2s^{-1}	CO_2 SD	Daily precip. (mm)	Daily precip. SD
1989	February	0.0375	0.26056	*NA*	*NA*	2.4321429	5.19865
1989	March	-0.04	0.1249	*NA*	*NA*	1.9483871	4.983564
1989	April	-1.1625	0.692382	*NA*	*NA*	3.0066667	6.505326
1989	May	-1.89	0.40025	*NA*	*NA*	4.7064516	10.30825
1989	June	-2.05143	0.868877	3.190795	0.657404651	4.0633333	8.144661
1989	July	-2.1775	1.393542	3.211119	0.701858804	3.4548387	6.624995
1989	August	-2.74667	1.32278	2.979736	0.522584346	2.7290323	4.831024
1989	September	-2.515	0.476519	2.356635	1.342468	3.8333333	8.604623
1989	October	-2.84667	1.01964	1.784251	0.715886015	4.0354839	7.562696
1989	November	-2.056	0.542468	0.91527	0.5609618	4.1766667	7.115388
1990	January	-0.13	0.014142	0.138021	*NA*	1.783871	2.885261
1990	February	-0.225	0.714178	0.240777	*NA*	2.4571429	4.775356
1990	March	-0.23	0.255995	0.931167	0.226849131	1.2387097	3.04507
1990	April	-0.80714	0.914362	1.009851	0.437640748	4.21	9.692989
1990	May	-1.41667	0.641176	1.368506	0.425882302	5.0483871	8.2299
1990	June	-2.695	0.439735	2.398005	0.972515032	1.8233333	3.572373
1990	July	-2.9225	0.102103	4.608566	1.606113471	2.1483871	6.214868
1990	August	-2.73	1.129779	3.442186	1.241424396	4.6225806	10.69158
1990	September	-3.505	1.491995	3.629508	1.452082571	1.4833333	3.337259
1990	October	-2.54167	0.60496	2.145386	0.899470593	6.7451613	14.47204
1990	November	-2.2025	0.909299	1.392827	0.690386688	3.7066667	10.84003
1990	December	-2.352	1.371831	0.737017	0.186507056	3.1870968	9.763461
1991	February	-0.31	0.028284	0.373285	0.030882743	2.0214286	7.241514
1991	March	-1.5375	0.321079	0.292304	0.150653033	3.2064516	6.414044
1991	April	-1.855	0.855798	1.067414	0.351861709	2.99	9.710405
1991	May	-2.14125	1.069291	1.610063	0.770265456	2.6032258	7.282741
1991	June	-2.57625	0.787871	2.688225	0.880916185	1.9	3.471559
1991	July	-4.051	1.094942	2.961695	1.111760812	2.3387097	5.787727
1991	August	-2.63333	1.371111	2.887474	0.337401488	7.6354839	25.31735
1991	September	-3.315	1.678145	2.632532	0.946281368	5.3533333	13.29814
1991	October	-2.82778	1.234704	2.116942	0.707435382	2.5387097	5.041804
1991	November	-2.44375	0.883757	1.448224	0.49925161	3.7366667	9.579665
1991	December	-1.06	1.164446	0.688796	0.751654781	2.3645161	4.809473

1992	January	-0.6825	0.723389	0.359139	0.014472883	1.9935484	5.296536
1992	February	0.4	0.757661	0.329549	0.144490965	1.8655172	6.505507
1992	March	-0.005	0.205994	0.343734	0.037612	2.6677419	6.075546
1992	April	-1.36571	0.915639	0.83192	0.375001925	2.02	7.094213
1992	May	-1.6325	0.703675	0.980025	0.415669049	0.9741935	2.72641
1992	June	-2.26286	0.656194	2.251858	0.829165628	3.8533333	11.88325
1992	July	-3.145	2.523671	4.963612	0.870164848	3.3677419	6.681287
1992	August	-2.18375	1.459079	4.09126	0.842192694	2.9322581	7.030144
1992	September	-2.94875	1.045076	3.175324	1.231401231	2.32	5.294526
1992	October	-2.23625	1.059001	1.559583	0.562409846	2.7774194	8.183834
1992	November	-1.455	0.829861	0.474156	0.209289103	4.4	7.459685
1992	December	-0.978	0.364925	0.448232	0.341424861	2.0096774	3.932883
1993	January	-1.935	2.38295	2.962932	4.00777786	1.3258065	2.9759
1993	February	-0.775	0.547996	0.187662	0.048854156	2.5142857	6.248301
1993	March	1.65	1.713651	0.935422	*NA*	3.3107143	6.922927
1993	April	-0.43	*NA*	0.799945	0.348405001	4.06	6.528854
1993	May	-2.7	0.933809	1.291629	0.545251649	0.6	1.852566
1993	June	-2.622	0.52756	2.66569	0.973564683	2.0666667	4.694482
1993	July	-3.17667	0.4121	2.158508	1.317372348	1.2225806	3.127375
1993	August	-3.913	0.974133	2.618022	1.099871803	1.8096774	5.34555
1994	January	-1.885	1.821529	1.699935	2.030204781	3.3354839	8.139719
1994	February	-1.27333	0.877515	1.267241	0.865655247	1.4535714	4.686741
1994	March	-6.65	*NA*	1.900501	0.95329641	4.4741935	11.01148
1994	May	*NA*	*NA*	1.268962	0.713315459	3.2935484	7.163888
1994	June	-1.91714	0.857763	6.622719	5.450988796	1.4633333	3.044439
1994	July	-2.5675	1.079703	2.929273	2.003876545	1.8258065	4.621397
1994	August	-3.33667	2.438059	2.482294	1.521335924	3.3193548	8.830758
1994	October	-1.4825	0.326943	1.283369	0.679655358	0.1548387	0.616354
1994	November	-2.685	0.558614	1.082633	*NA*	2.4366667	6.803421
1994	December	-2.1825	0.421298	*NA*	*NA*	4.5451613	12.90023
1995	January	-0.88	0.575674	*NA*	*NA*	3.6451613	7.370429
1995	February	-1.42167	0.878258	0.831666	0.653980624	2.5	5.686827
1995	March	-1.1125	1.41394	1.222998	0.775734581	1.5322581	3.119336
1995	April	-1.1875	0.38187	0.893952	0.31157878	1.57	3.691477
1995	May	-1.5925	0.324487	1.91139	1.56047786	2.2451613	3.897421
1995	June	-1.775	0.776681	3.246671	0.865745087	1.5833333	3.385373
1995	July	-3.84	1.901911	4.09981	1.616196258	3.1096774	6.424918
1995	August	-2.84	0.212132	6.378853	1.230203508	2.2258065	5.957515
1995	September	-3.6275	1.653972	1.965458	1.027273779	2.36	5.928633

1995	October	-3.11	0.937408	3.096709	0.828347537	5.3516129	13.4586
1995	November	-1.745	0.900907	1.797399	0.382224365	6.2766667	15.42914
1995	December	-1.62	0.127279	0.874509	*NA*	2.0193548	3.851962
1996	January	*NA*	*NA*	0.442898	0.162719951	2.2419355	3.742617
1996	February	-0.98	0.254558	3.681421	4.842945654	1.3931034	3.284045
1996	April	-2.76	2.220315	1.404805	0.007371624	5.63	11.19622
1996	May	-1.895	1.859691	1.94609	*NA*	3.3129032	5.807968
1996	June	-2.42875	0.955173	4.013209	2.24262316	1.5733333	3.406478
1996	July	-2.877	1.282948	4.118529	1.325513747	5.7	14.80861
1996	August	-3.01833	0.827126	3.870806	1.643502578	0.6903226	3.23031
1996	September	-2.52	1.66319	3.862869	1.123072182	3.05	6.454816
1996	October	-3.63	1.007472	2.79048	0.395563409	9.7935484	32.16386
1996	November	-2.355	0.580026	1.242397	0.598539571	1.6233333	5.175651
1996	December	-2.25	1.241639	0.870957	0.20703692	4.6344828	9.348311
1997	January	-1.1975	0.860518	0.361025	0.12949858	3.1322581	7.725473
1997	February	-1.43	0.569298	0.198115	0.117041341	1.9464286	5.251348
1997	March	-0.3375	0.302366	0.245408	0.127548334	3.662069	6.443236
1997	April	-0.44	0.098995	1.534992	1.412535273	5.01	12.82071
1997	May	-1.835	0.827315	1.535235	1.26767426	2.0064516	3.679759
1997	August	-4.025	0.190919	5.235967	0.852818719	3.0580645	7.79516
1997	September	-3.315	1.718269	4.165456	0.663852056	1.76	5.243097
1997	October	-3.55	1.004092	2.70386	0.901412557	0.6709677	2.109849
1997	November	-2.36	1.25865	1.298292	0.065781022	5.6133333	16.67643
1997	December	-2.92	1.880904	0.667542	0.672592645	2.6258065	6.734487
1998	April	-2.195	1.760696	0.711163	0.094210069	1.72	5.246963
1998	June	-4.24	1.661525	11.02045	5.426735258	10.1785714	24.47158
1998	July	-4.493	1.674827	13.1625	3.646705706	1.6967742	3.921265
1998	August	-4.67	1.288798	6.451802	1.603137552	2.2258065	5.665272
1998	September	-5.38	2.856711	4.008105	0.152216691	1.62	4.786576
1998	October	-4.21	2.489016	4.111915	0.572209411	4.1903226	9.702967
1998	November	-4.74	3.761808	1.714682	1.090431578	1.81	4.924488
1999	May	-3.275	0.629325	5.065525	0.100525921	2.6967742	5.496149
1999	June	-6.729	2.185622	6.387628	3.783039809	0.76	2.525812
1999	July	-6.74833	1.262734	6.245494	1.629037959	1.9806452	5.067769
1999	September	-4.5225	2.733354	4.504467	0.913226602	7.8833333	21.08873
1999	October	-5.225	3.585031	1.965197	0.190357834	3.1516129	7.343608
2000	May	-4.22	3.493108	1.490399	1.068800029	2.862069	6.49029
2000	June	-3.58	1.578647	7.363118	4.110051852	5.2526316	11.77414
2000	July	-3.79	1.591639	7.697246	2.639432365	4.0258065	10.21427

2000	August	-4.106	1.64687	7.292967	2.791502558	2.0258065	5.943791
2000	September	-5.045	1.775339	4.331621	2.564833434	2.8066667	6.961418
2000	November	-6.765	1.011163	7.563427	3.361322828	4.0433333	8.488153
2001	January	-2.965	1.393	1.250528	1.092403363	0.4068966	1.496169
2001	February	-0.74	0.141421	1.527112	1.838066781	0.8307692	2.533104
2001	May	-4.54333	1.17449	2.385948	0.79490355	1.016129	3.025018
2001	June	-3.7325	1.60367	6.58646	1.059329825	4.48	9.766177
2001	July	-4.7675	3.390874	4.963875	0.604774332	2.5903226	4.872806
2001	August	-5.58333	0.992891	*NA*	*NA*	1.0645161	2.728681
2021	June	-3.16389	0.923511	2.535705	0.933778357	0.9466667	1.997711
2021	July	-2.86833	1.016159	2.965024	0.892544416	10.4193548	18.70167
2021	August	-3.40333	1.575991	3.091798	0.899509237	3.0870968	6.792042
2021	September	-3.34667	1.255173	*NA*	*NA*	3.59	12.01942
2021	October	-3.5875	1.051468	*NA*	*NA*	5.383871	16.02881

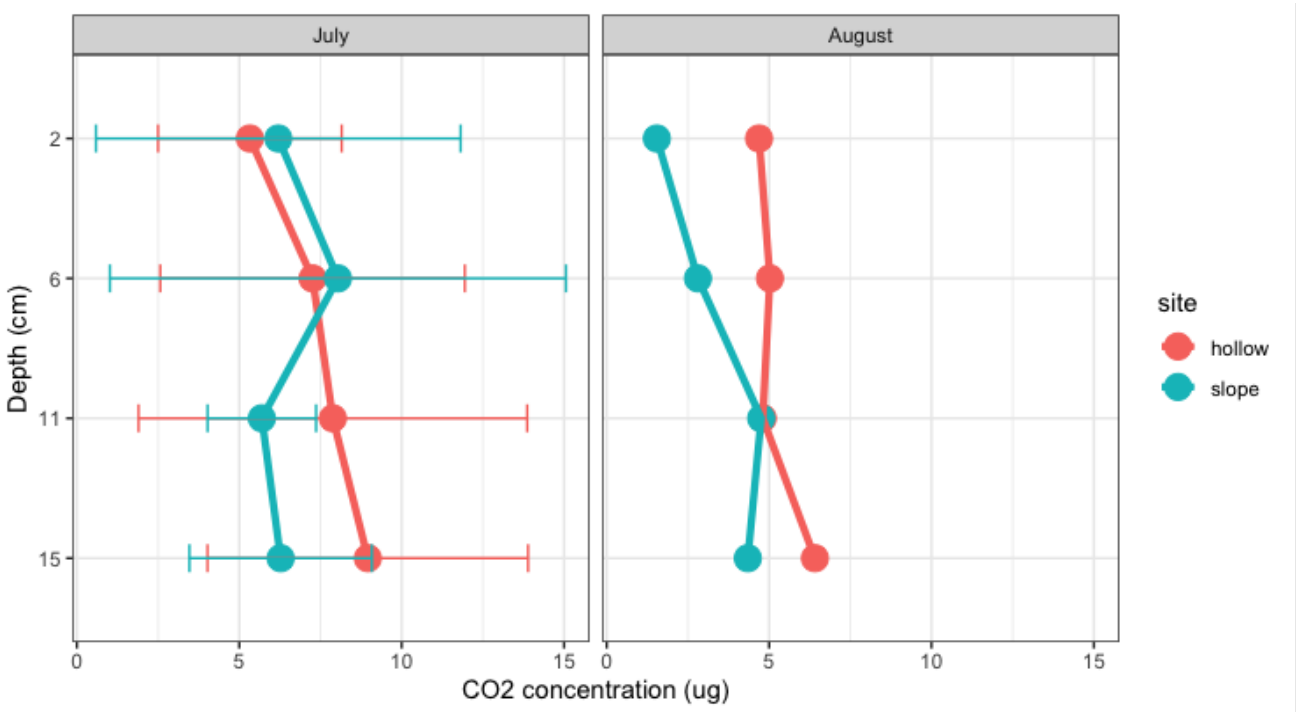

Figure 1: Soil gas profile of CO_2 concentrations. Each point represents the monthly mean value for soil gas CO_2 concentration at each depth, with error bars showing ± one standard deviation of the mean.