Assessing how invasive cattail treatment affects methane emissions in Lake Ontario meadow marshes

By

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Abstract

Wetlands sequester carbon and buffer extreme weather events; however, they are also the largest natural source of methane, a potent greenhouse gas (GHG). We assessed the effects of invasive cattail (*Typha* spp.) removal on methane emissions and carbon storage at the Braddock Bay Wildlife Management Area on the southern coast of Lake Ontario. We measured methane fluxes in two restored marshes and an uninvaded meadow marsh using a backpack cavity ring-down analyzer, logged belowground environmental conditions to model methane fluxes over time, and estimated carbon storage of the different sites. In 2019, data were collected during a record high flood event in Lake Ontario, and we did not find a significant difference in methane fluxes among the control, treatment, and uninvaded areas. However, under lower water conditions in 2020, the cattail monoculture (control) emitted more methane than areas with cattail removal with replanting, and the uninvaded wet meadow had the lowest average flux during the growing season. Soil moisture (and water levels) decreased over the growing season, while temperatures and biomass increased. Areas where cattail had invaded (whether removed or not) had greater percent soil organic carbon than the uninvaded meadow marsh. The control sites had greater carbon stored aboveground than replanted sites. Our results indicate that methane emissions may be mitigated through plant community restoration; however, ensuring that the native plant community establishes will be important to long-term carbon sequestration.

In addition, a mesocosm experiment was conducted to investigate methane emissions when substrate and plant community were manipulated. The experiment included six treatments with two different substrate types and three plant community types. The two substrate types were the top five cm from cattail monocultures, “cattail substrate”, and the next layer below the layer of organic matter, “topsoil removed substrate”. There were three levels of planting treatments: no plants, cattail (*Typha* x *glauca)*, and native grass, *Calamagrostis canadensis*. The findings indicated that mesocosms with the topsoil removed had significantly lower methane emissions than those with topsoil. Moreover, no significant differences in emissions were observed between Canada bluejoint (*Calamagrostis canadensis*) and *Typha* x *glauca*. However, it is important to note that this was a limited experiment, and more data is required for comprehensive analysis. Our results show that substrate type appears to be a more important factor contributing to methane emissions than plant identity.

General Introduction

Communities around the world are undertaking wetland restoration efforts to enhance resilience against climate change, as wetlands have the potential to sequester carbon and mitigate extreme weather events (Fennessy and Lei 2018). However, it is important to note that wetlands naturally release methane, a potent greenhouse gas which can offset their cooling effect on the climate. Notably, New York State's current plan to reduce greenhouse gas emissions does not include ecosystem-based management of wetlands (NYSDEC 2017), despite the fact that wetlands contribute 15-32% of the methane emissions in temperate North America (Kirschke et al. 2013).

The dynamics of methane in wetlands are influenced by plant traits, and it has been observed that highly productive invasive species are associated with increased methane emissions (Vroom et al. 2022, Lawrence et al. 2017, Martin and Moseman-Valtierra 2015, Yin et al. 2015, Tong et al. 2012, Laanbroek 2010). Lawrence et al. (2017) demonstrated that freshwater marshes dominated by invasive cattail (*Typha* x *glauca*) emitted approximately 300% more methane from the soil compared to marshes with native vegetation. Additionally, Kao-Kniffin et al. (2011) found that methane emissions rose with an increase in invasive cattail biomass under elevated CO_2 concentrations. Even after removal, the productivity of cattail and the formation of a thick root mat trapping organic matter may lead to a legacy of carbon for methane-generating bacteria (Corbin and D'Antonio 2012, Le Mer and Roger 2001). Despite substantial investments in eradicating invasive species in wetlands, our understanding of the effects of these efforts on greenhouse gas emissions remains limited (Martin and Moseman-Valtierra 2017, Sheng et al. 2014). Moreover, there has been relatively little research conducted on the impacts of invasive species in freshwater marshes to date (Johnson et al. 2021).

This research is split into two chapters, which investigates how wetland restoration (cattail removal and native plantings), plant type, and carbon availability in the soil can affect methane emissions. Chapter One was a two-year field study which explored three Great Lakes coastal wetlands within the Braddock Bay Wildlife Management Area in Greece, NY: locally known as Braddock Bay, Buck Pond, and Salmon Creek. Great Lakes wetlands typically include meadow marsh habitats, which have been invaded by hybrid (*Typha* x *glauca*) and narrow leaf cattail (*Typha angustifolia*) over the years (Freeland et al. 2013). Portions of Braddock Bay and Buck Pond have had restoration projects completed to restore some cattail monoculture areas back into meadow marsh habitat. These restoration areas and cattail monoculture areas were sampled for methane flux, along with an untouched meadow marsh habitat in Salmon Creek, over the growing season.

Chapter Two was a one-year mesocosm experiment over the growing season which sought to explore differences in methane emissions with various plant types and soil types. The experiment evaluated *T.* x *glauca*, Canada bluejoint (*Calamagrostis canadensis*), and no plants on either soil collected from a cattail monoculture in Buck Pond with a thick layer of detritus, or soil with the detritus layer removed. The detritus-removed soil was to simulate soil conditions pre-cattail invasion, while the other soil option was to simulate post-cattail invasion. *C.*

canadensis is a typical meadow marsh species which was also used to simulate pre-cattail invasion.

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Chapter One – Field Study

Introduction

Climate change may degrade ecosystem services such as habitats for native populations, flood control and relief, and nutrient buffering (Zedler and Kercher 2004). This change is driven by an increase in greenhouse gases (GHGs) released and trapped in the atmosphere (Forster et al. 2007). However, there are natural ways in which the amount of GHGs released into the atmosphere can be reduced. Wetlands sequester carbon, and due to that potential, communities worldwide are restoring wetlands to increase resiliency against climate change (Fennessy and Lei 2018). However, wetlands are also one of the largest natural sources of methane (Forster et al. 2007). Under anoxic conditions, methanogenic bacteria in wetlands produce methane gas. Methane is a potent greenhouse gas, which can reduce the natural cooling potential of wetlands (Forster et al. 2007). Methane is considered 24 times more potent than carbon dioxide in contributing to climate change (Lashof and Ahuja 1990). Methane dynamics of wetlands are an important factor in the acceleration of climate change. Methane release can be altered by the types of plants in the area as well as the type and quantity of litter (Lashof and Ahuja 1990, Laanbroek 2010). Different plants have different traits that can alter the microbial activity, biotic and abiotic factors in a system (Boutin and Keddy 1993, Bouchard et al. 2007).

Methane Gas, Pathways, and Production

Methane (CH_4) gas is generated, transformed, and transported in oxygen-deprived environments commonly found in wetlands. It is released into the atmosphere through plant transport, diffusion from the soil, and ebullition (Byrnes et al. 1995, Forster et al. 2007). Methane concentrations are influenced by the microbial communities present in the soil. There are two types of bacteria that affect the amount of methane in a system: methanogens and methanotrophs. Methanogens use carbon substrates as an electron acceptor in anerobic respiration, which produces methane (Reddy and Delaune 2008). Methanotrophs oxidize methane into carbon dioxide through aerobic respiration (Bartlett et al. 1985). Soil moisture and oxygen account for 30% of the variation in methane fluxes in temperate wetlands, and fluxes also differ over the course of the growing season (Smyth et al. 2019). Methanogenesis and methanotrophy can happen in the same system because of gas transport through porous tissue that can oxygenate the soil (Whalen 2005, Kao-Kniffin et al. 2010). Plant species and plant traits, soil composition, and temperature influence the microbial activity within the soil and alter methane production and oxidation (Williams and Yavitt 2008). Stem and root porosity differs among plant species. This variability is crucial for understanding how different plants contribute to gas exchange in wetlands. Gas released through plant transport can be through porous tissue, such as aerenchyma, or through hollow stems, like in grasses. The formation of aerenchyma is an anatomical adaptation to excess water stress in plants and enables rapid transport of gases, including oxygen, carbon dioxide, ethylene, and methane (Takahashi et al. 2014). Aerenchyma allows oxygen to travel to the roots and create an oxidized rhizosphere where aerobic respiration can then take place. Due to the porosity, aerenchyma can act as a way for oxygen to travel into the soil, and as a way for methane to escape from the soil. Fluxes of gases in aerenchyma depend on the concentration, pressure gradients, and internal structure (Byrnes et al. 1995).

Concentration gradients between the soil and atmosphere drive soil diffusion as a gas transport pathway. This is a physical process in which particles travel from areas of higher concentration to lower concentration. Ebullition can occur through the release of gas bubbles trapped in the soil into the atmosphere. Gas bubbles can additionally be released from organisms disturbing the ground. Figure 1.1 shows the three gas transport pathways (Bastviken et al. 2023). Ebullition in wetlands is affected by changes in atmospheric and hydrostatic pressure due to changes in water level (Sebacher et al. 1985).

Additionally, Schultz et al. (2011) found that water level was more important in determining methane flux than species dominance in emergent marsh wetlands. Flooded conditions lead to the development of physiological adaptations to water logging, such as aerenchyma, which facilitates oxygen transport to the roots, as well as methane transport into the atmosphere. These long-term flooding regimes create the oxygen-deprived environments in which methane production can begin to occur.

Cattail Invasion

Many Great Lakes coastal wetlands are invaded by non-native species (Carson et al. 2018). A common invasive species in these systems is the hybrid cattail, *Typha* x *glauca* (Wilcox et al. 2008). This hybrid species is an aggressive colonizer because it has high productivity, large amounts of aboveground and belowground biomass, and can reproduce by growing genetically identical shoots, or by seed. It is a cross between native wide leaf cattail (*Typha latifolia*) and nonnative narrowleaf cattail (*T. angustifolia*) and has become one of the most problematic invaders in the Laurentian Great Lakes with rapid colonization, thick litter accumulations, and monodominant vegetation stands (Bansal et al. 2019, Freeland et al. 2013). Great Lakes wetlands historically included wet meadow habitat, which has been invaded by cattail over the years (Wilcox et al. 2008). Wet meadow species include annual and perennial sedges and grasses (Boutin and Keddy 1993 Wet meadows produce very diverse herbaceous cover with less production of total biomass and litter build up in comparison to *Typha* spp. (Boutin and Keddy 1993).

Typha x glauca and *T. angustifolia* are associated with locally high soil nutrients, low light, and substantial amounts of organic litter (Angeloni et al. 2006). For example, Angeloni et al. (2006) found that a zone dominated with invasive cattail had 14 times more plant litter and four times the soil organic matter compared to noninvaded soils (Angeloni et al. 2006). Invasive cattail litter also leads to the decrease in abundance and diversity of native plants (Farrer and Goldberg 2009). Due to the high carbon content of its litter, Williams and Yavitt (2008) in a soil incubation study have found that methane generation is enhanced with *Typha* spp.

Results in the literature on how invasive *Typha* spp. influence methane emissions differ depending on which gas pathway is studied. Methane dynamics have been studied using soil incubation, soil chambers, and whole system chambers (Table 1.1). Soil chambers account for microbial activity, soil diffusion, ebullition, and the influence of roots, while chambers over the entire system can also measure the methane flux due to plant transport. In studies addressing plant transport using a whole system chamber, Kao-Kniffin et al. (2010) found that clonal dominants (*Phalaris arundinacea* and *Typha angustifolia*) had the lowest methane emission compared to tussocks (sedges had the highest methane flux). However, Lawrence et al. (2017) found that *Typha* spp. invasion led to an increased amount of methane emitted due to soil diffusion due to increased carbon accumulation. Lawrence et al. (2017) studied soil incubations and did not account for plant transport. Soil incubations account for the potential methane generation based on microbial activity and carbon in the soil.

There is evidence that certain plant invasions, like invasive cattail (*Typha* spp.), can lead to increased methane emissions from wetlands. However, there is a lack of understanding regarding how the removal of invasive plants affects methane dynamics in freshwater marshes (Lawrence et al. 2017, Kao-Kniffin et al. 2011). After the eradication of invasive cattails, the productivity of these plants and the formation of a dense root mat that traps organic matter may result in a carbon legacy for methane-generating bacteria (Corbin and D'Antonio 2012). While previous studies have shown the impact of cattail invasion on methane flux, the influence of restoration on methane dynamics remains unclear.

Methane dynamics are also affected by the type of chamber used to sample. Gunther et al. (2014) compared methane fluxes from transparent vs opaque whole system chambers on *Carex*-, *Phragmites*-, and *Typha*-dominated stands in the Trebel Valley, a temperate fen in Northeast Germany in August 2011. Methane emissions were considerably lower when using opaque chambers within *P. australis*, and *T. latifolia* stands (Gunther et al. 2014). Additionally, *Carex*-stand had an average methane flux of 9.1 mg CH_4 /m^2/hr and the *Typha*-stand has an average of 14.6 mg CH_4 /m^2/hr. Plant community also plays an important role in methane dynamics.

Keyport et al. (2018) studied plant community and soil carbon flux in Cheboygan Marsh on Lake Huron in Michigan, USA. Experimental *Typha* spp. harvest and treatment began in July 2011 and plant community surveys were conducted annually through July 2015. Additionally, in July and August 2015 Keyport et al. (2018) measured methane concentrations in the soil using a 15 cm diameter PVC pipe placed in unvegetated locations. Keyport et al. (2018) found that experimental restoration of *Typha* plots by one-time aboveground harvest and removal of plant litter reduced the litter layer, reduced *Typha* dominance of the plant community, but did not influence soil methane flux. It is important to note that soil incubation research can differ between whole system applications.

Johnson et al. (2021) studied methane flux using a whole system approach in Michigan, USA within the northern Great Lakes Region. Cheboygan Marsh is a lacustrine open-embayment wetland on northern Lake Huron which also investigated cattail treatment and restoration strategies. Experimental treatments were implemented in 60 m x 60 m plots between 2015 and 2017. Cattail treatments included harvesting by cutting and removing the biomass in mid-July and early September; and crushing and leaving the biomass in mid-July each year. Methane concentrations were measured in July and August of 2017 using a transparent PVC chamber on a floating foam base. Johnson et al. (2021) observed higher flux rate with *Typha* spp. harvest and biomass removal than crushing and no treatments. The 5-year study (2015-2019) also found that elevated total methane flux rates persisted at least one year after harvesting. These findings underscore the complex and prolonged ecological impacts of wetland harvesting and restoration efforts, highlighting the need for long-term monitoring and adaptive management strategies.

The recovery of physical features such as hydrology, topography, and soil permeability in wetlands tends to be immediate post-restoration, while biological and biogeochemical structures show significantly delayed and reduced recovery, respectively. In a meta-analysis by Moreno-Mateos et al. (2012), overall biologic structure recovered only about 77% after restoration compared to reference wetlands, and biogeochemical recovery was drastically reduced post-restoration. After restoration, carbon storage appears to slightly increase then plateau, remaining below the level of reference wetlands. Once wetland hydrologic regimes are recovered, anaerobic conditions allow stores of organic carbon to reaccumulate back into the soil but are still less than pre-restoration levels even after 20 years (Moreno-Mateos et al. 2012). In this study, we measured the combined effects of plant transport, soil diffusion, and ebullition on methane production in a field study following invasive cattail treatment.

Objectives and Hypotheses

The main goal of the field study was to measure methane fluxes from freshwater wetland plots in three categories. These categories included plots within a cattail removal area that had been replanted, untreated cattail marsh, and uninvaded meadow marsh. My objectives at these plots were to 1) measure methane fluxes in the three categories, 2) collect measurements of belowground environmental conditions such as soil oxygen, moisture, and temperature to model methane emissions over the growing season, and 3) determine the carbon storage in aboveground biomass and substrate. I hypothesized that invasive cattail eradication would lower methane emissions by reducing the amount of carbon available for methane production via primary productivity, and that restored sites would have higher methane emissions than the native meadow marsh site due to the legacy of carbon-rich cattail substrate.

Methods

Site Description

My study sites were coastal wetlands on the southern shore of Lake Ontario near Greece, New York which included Braddock Bay, Buck Pond, and Salmon Creek (Figure 1.2). The wetlands of Braddock Bay and Salmon Creek are classified as a riverine beach lagoon coastal wetland, while Buck Pond is classified as a barrier beach lagoon coastal wetland (Albert et al. 2005). These wetlands include large stands of emergent marsh vegetation dominated by hybrid cattail (*Typha* x *glauca*) and narrowleaf cattail (*Typha angustifolia*). The U.S. Fish and Wildlife Service, Ducks Unlimited, and other associated partners conducted restoration efforts in Buck Pond and Braddock Bay to treat the cattail-dominated emergent marsh, replant with meadow marsh species, and create potholes as open water habitat (Wilcox and Bateman 2018). The restored sites in this study included Braddock Bay and Buck Pond which were restored by cattail cutting and herbicide application followed by planting native species.

Braddock Bay cattail-treatment measures were completed in August 2018. Cattail-treatment measures included removal of past-years' growth by mowing in winter, cutting cattails with steel-bladed trimmers, herbicide treatment of new stems by hand-wicking, and replanting with native sedge-grass plugs and seeds (Silva et al. 2021). Buck Pond's cattail treatment measures were completed in September 2016. Similarly, Buck Pond cattails were mowed, cut with steel-blade brush cutters, applied herbicide to resprouting cattail ramets, and reseeded with native wet meadow mixes

(Polzer and Wilcox, 2022). In my study, these restored sites had sampling plots in treated cattail zones and in untreated cattail zones. Salmon Creek, a tributary that flows into Braddock Bay, was chosen as a native wet meadow study site which was used to descriptively compare the treated and control sites. Salmon Creek was unable to be analyzed statistically since it has not had any restoration treatments yet.

To select appropriate wetlands, historical meadow marsh data were collected around Lake Ontario and used for comparison to the habitat present. The study sites where plots were located had similar elevation (between 74.75 m and 75.25 m), slope, and flood regime and was in an area that was historically meadow marsh (Wilcox et al. 2008). The study sites also had a similar soil type, a mosaic of Niagara silt loam and freshwater marsh soil. Using ArcGIS, plots were chosen randomly within a restored area and a cattail monoculture area within Braddock Bay and Buck Pond (Figures 1.3 and 1.4). At each of the two restored sites, 20 plots were selected for sampling in the summers of 2019 and 2020; ten plots in removed cattail with replanting, and ten plots in untreated cattail. At Salmon Creek, ten plots were randomly selected in the uninvaded meadow marsh (Figure 1.5). New plots were chosen each year. Over the two years, 100 total plots were selected for sampling. Each plot was sampled twice during the growing season where all plots were sampled within ten days of each other. The growing season is defined by the last frost date and the first fall frost (Vega et al. 2020). In New York, this is usually considered from May through September. Sampling plots within ten days of each other minimized monthly seasonal variability within the period. New sampling locations were selected for 2020 to avoid clipping effects from biomass harvesting.

Methane Sampling

Methane flux was measured using a portable gas chromatograph with closed chambers systems similar to Schultz et al. (2018). In May 2019 and 2020, we installed plastic 54 L x 40 W x 28 H cm soil collars at least seven inches into the ground at each plot ensuring they were not flooded at least two weeks prior to gas sampling to reduce the effect of soil disturbance on gas fluxes. Soil collars were not installed in plots that were flooded. There were two sampling periods each during the summers of 2019 and 2020 where gas fluxes of methane were measured using clear chambers fitted over the vegetation, creating a gas tight seal with the soil collar. The clear chambers were made from a 50 L x 34.5 W x 94 H cm metal frame with a plexiglass top and clear plastic sheeting surround. Inside the chambers were an icepack and fan to reduce the likelihood of temperature increasing more than the ambient temperature outside. An airtight seal was created by pressing the excess clear plastic liner below the water surface. In instances where the water was too deep to safely enter with waders, 50 L x 34.5 W x 94 H cm floating chamber was used. The floating chamber used plexiglass instead of a clear plastic liner and had foam along the base for buoyancy.

To measure methane fluxes in the wetland plots, we used a Picarro GasScouter cavity ring down spectrometer (G4301, Picarro) that was hooked up to the chambers with Teflon tubing for six minutes at a time, under full sun conditions, between 10 am and 2 pm. Full sun conditions were measured using a Venier Lab Quest 2 photosynthetically active radiation (PAR) sensor. Photosynthesis can occur between 400 and 700 nanometers, and the PAR sensor measures 0-2500 μmol/m^2/s (370-640 nm). We only sampled methane fluxes when PAR was above 300 μmol/m^2/s. Air and soil temperature were also recorded at each plot using a thermometer connected to Lab Quest 2, and water depth was measured using a meter stick.

Belowground Environmental Conditions

To account for the variation in methane flux due to soil moisture and soil oxygen, Two METER Group ZL6 data loggers and one custom "Arduino" data logger programed by the SUNY Brockport Physics Department were installed at Salmon Creek, Buck Pond, and Braddock Bay. The ZL6 meters had three EC-5 soil moisture probes, while the custom Arduino data logger had three EC-5 soil moisture probes and three Apogee SO-411 soil oxygen sensors. The SO-411 sensors were not compatible with the ZL6 data loggers. Data was logged from May through September. Once collected after the growing season, data was downloaded from the data loggers and average soil moisture and average soil oxygen were graphed over the growing season by month. Soil moisture was collected in the years 2019 and 2020 at Salmon Creek and 2020 in Buck Pond and Braddock Bay.

Aboveground Biomass and Soil Carbon

To measure the aboveground biomass, all plant material in the plot at ground level was cut in August at peak biomass, oven dried for 72 hours at 55 degrees Celsius, and weighed in grams. If the sites were flooded, ground level was below the water where substrate began. After drying, the plants were homogenized, split into two subsamples per plot, and weighed in grams (234 total subsamples). The subsamples were burned in a muffle furnace at 550 degrees Celsius for three hours, cooled, and weighed in grams to determine loss on ignition. Subsample values were averaged together. Grams of carbon per meter squared was calculated using the loss on ignition formula:

$$\frac{\text{total plot oven dried weight} \times \text{average percent carbon}}{\text{plot area}}$$

Soil carbon was determined by using the muffle furnace, loss on ignition method (Salehi et al. 2011). The amount of carbon storage in the substrate was determined from harvesting one soil core in each plot at the end of the season. Soil was collected using a soil auger from zero to 30 cm in depth. Cores were placed in a large plastic zip-lock bag and labeled by plot ID. The soil samples were split into two subsamples, oven dried for 72 hours at 105 degrees Celsius, and weighed in grams. The subsamples were then combusted in a muffle furnace at 360 degrees Celsius for two hours, cooled, and weighed to determine carbon loss on ignition. Subsample values were averaged together. Additionally, the plant matter and soils were dried and combusted at different temperatures and times due to the organic matter composition, moisture content, and density of the material. This was to avoid potential damage to the substrate and encourage even drying (Salehi et al. 2011).

Statistics and Data Analysis

Methane fluxes were calculated using R ‘flux’ package (Jurasinski et al. 2014). The R ‘flux’ package calculates gas flux using the date, time, Plot ID, temperature, water depth, chamber volume, chamber area, and methane parts per billion. Raw data was downloaded from the Picarro GasScouter and compiled into a spreadsheet by site and month. Using R, the data was partitioned into data tables per chamber measurement, then flux was calculated for each site and month. Flux data was downloaded and compiled into a combined spreadsheet containing the Plot ID, CH_4 units in parts per billion (ppb), Flux, Plot Type, Period, Site, and Year. All data was transformed using $\log_{10} +1$. Normality and homogeneity of the data were determined using the Shapiro-Wilk test and Levene’s Test. After confirming normality, a mixed effects model ANOVA was used to determine if there was a significant difference between plant community types for gas flux. We used a mixed effects model where the random effect was the Plot ID due to being sampled more than once, and the fixed effects were the plot type and sampling period. Years were analyzed separately due to extreme lake level differences. A multiple regression analysis was conducted to test which variables out of soil organic carbon, aboveground biomass, water depth, soil temperature, and site (predictor variables) best predict the methane flux in a system (response variable) to facilitate estimation of emissions within a wetland over a growing season.

Results

Water levels between 2019 and 2020 varied greatly. Mean high water level for Lake Ontario is approximately 75 m International Great Lakes Datum (IGLD) (averaged from 1918 to 2020) (Lake Ontario Climatology 2024). In 2019 water levels were exceptionally high, peak water level elevation in mid-June was approximately 75.91 m IGLD and ranged from 75.89 m to 75.39 m between July 1 and August 31 when sampling took place. On average, the water level within the sampling periods in 2020 was 0.41 m less than in 2019. This variation yielded different results between the years.

Methane Sampling

Methane fluxes were collected at Salmon Creek, Buck Pond, and Braddock Bay coastal wetlands in July and August 2019, and June and July 2020. Salmon Creek included the uninvaded meadow marsh category; Buck Pond and Braddock Bay included cattail removal with replanting, and cattail monoculture (control) categories. Lake Ontario water levels were at a record high in 2019 and higher than average in 2020. In the two years, 20 plots were sampled in Salmon Creek, 34 plots in Braddock Bay, and 40 plots in Buck Pond.

In 2019, the lowest methane flux was 11.4 mg CH_4/m^2hr in a Braddock Bay treatment area and the highest flux was 201.6 mg $CH_4/m^2/hr$ in a Buck Pond treatment area. The average methane flux was 59.7 mg $CH_4/m^2/hr$. Treated and control cattail areas were not significantly different from one another (Table 1.2, Figure 1.6). When grouped by plot type, the cattail invaded plots had an average flux of 53.5 mg $CH_4/m^2/hr$ with a standard deviation of +/- 38.6 mg $CH_4/m^2/hr$ and the treatment plots had an average of 59.5 mg $CH_4/m^2/hr$ +/- 40.5 mg $CH_4/m^2/hr$. Salmon Creek (wet meadow control) averaged 74.4 mg $CH_4/m^2/hr$ +/- 65.0 mg $CH_4/m^2/hr$.

In 2020, the lowest methane flux was -0.3 mg $CH_4/m^2/hr$ in the Salmon Creek wet meadow control and the highest methane flux was 659.2 mg $CH_4/m^2/hr$ in Buck Pond cattail. The average was 68.8 mg $CH_4/m^2/hr$. The cattail invaded plots had an average of 124.5 mg $CH_4/m^2/hr$ +/- 149.3 and the treatment plots averaged 63.9 mg $CH_4/m^2/hr$ +/- 106.0 mg $CH_4/m^2/hr$. Salmon Creek (uninvaded meadow marsh) averaged 0.0 mg $CH_4/m^2/hr$ +/- 0.1 mg $CH_4/m^2/hr$. Plot type was significantly different where treatment areas had significantly lower emissions than the cattail monoculture. (Table 1.3, Figure 1.6).

In Braddock Bay and Buck Pond, there is a positive relationship between water depth and methane flux in both 2019 and 2020. Water depths at Salmon Creek were more consistent and do not appear to show a relationship (Figure 1.7). Additionally, in 2019, there appears to be a slight negative relationship between soil temperature and methane flux within Braddock Bay and Buck Pond and positive relationship within Salmon Creek. In 2020, there is a positive relationship between soil temperature and methane flux within all the sites (Figure 1.8).

Belowground Environmental Conditions

The data in the appendix illustrates a consistent decrease in soil moisture as the growing season progresses from May to September. Soil moisture levels are at their peak in May and steadily decrease through August. While there may have been some issues with the soil oxygen and moisture sensors at Braddock Bay, the figures in the appendix clearly indicate that as soil moisture levels rise, soil oxygen levels decline (Figures A1-A5).

Aboveground Biomass and Soil Carbon

The average soil organic carbon (SOC) percentage in Salmon Creek was 2.9% with a minimum value of 1.6% and maximum of 10.2%. The average SOC% in Buck Pond treatment area was 4.5% and the average SOC% in the cattail area was 14.3% with a range between 1.5% and 37.1%. The average SOC% in Braddock Bay treatment area was 23.2% and the average in Braddock Bay cattail area was 20.4%, with a range of 2.6% to 43.7%. Salmon Creek had a lower SOC% than Braddock Bay and Buck Pond (Figure 1.9). Buck Pond and Salmon Creek SOC% were consistent between years, while 2019 Braddock Bay SOC% within the treated and control plots were much higher than in 2020. In 2020, there is a positive relationship between SOC% and methane flux, and no apparent relationship in 2019 (Figure1.11). Figures

1.6 and 1.7 show that higher SOC% and aboveground biomass are associated with higher methane emissions.

The cattail removal and replanted sites, and the meadow marsh had less aboveground biomass accumulation than the cattail (control) plots (Figure 1.10). The average aboveground biomass accumulation (g Carbon/m^2) in Salmon Creek was 154.7 g/m^2 and ranged from 29.8 g/m^2 to 261.8 g/m^2. The average biomass in Braddock Bay cattail area was 236.2 g/m^2 and 81.8 g/m^2 in the treatment area. The range in Braddock Bay was 10.9 g/m^2 and 437.1 g/m^2. The average aboveground biomass in Buck Pond treatment area was 96.8 g/m^2 and 277.3 g/m^2 in the cattail area: ranging from 15.2 g/m^2 to 542.4 g/m^2. No apparent relationship between aboveground biomass and methane flux in 2019, however it appears that in 2020, higher aboveground biomass in cattail monocultures result in higher methane fluxes (Figure 1.12).

A linear mixed effects model was conducted using environmental data to predict methane flux in 2019 and 2020 (Tables 1.4 and 1.5). Water depth was a significant factor to predict methane flux in 2019 (Table 1.4, Figure 1.7). We did not find any significant factors within the 2020 sampling year.

Discussion

The objective of this field study was to measure methane fluxes from freshwater wetland plots in three categories: cattail removal area with replanting, untreated cattail marsh, and uninvaded meadow marsh. At these three categories I collected data to 1) measure methane fluxes in the three categories, 2) collect measurements of belowground environmental conditions such as soil oxygen, moisture, and temperature to model methane emissions over the growing season, and 3) determine the carbon storage in aboveground biomass and substrate. These objectives were used to answer the following hypotheses: invasive cattail eradication would lower methane emissions by reducing the amount of carbon available for methane production via primary productivity; and that restored sites would have higher methane emissions than the native meadow marsh site due to the legacy of carbon-rich cattail substrate.

Cattail Eradication and Replanting Methane

In my initial hypothesis, I proposed that restoring invasive cattails would reduce methane emissions by decreasing the amount of carbon available for methane production through primary production. In 2020, the treated sites showed lower methane production compared to the control sites. However, during the record high water levels in 2019, methane production was not significantly different between the plot types. The cattail-invaded plots, on average, stored a larger amount of carbon in aboveground biomass than the treated plots, providing evidence in support of my hypothesis. However, my data shows that soil organic carbon percentage and carbon stored in aboveground biomass did not significantly predict methane flux.

We had intended to collect soil oxygen and soil moisture data at each site to incorporate into my model. Unfortunately, we encountered technological issues and flooding, which resulted in incomplete data that had to be excluded from the analysis. This data could have played a crucial role in predicting methane emissions in wetlands. According to Smyth et al. (2019), changes in soil oxygen concentrations have a direct impact on methane fluxes, as methane is generated under oxygen-deprived conditions. These conditions are typically present when the soil is fully saturated due to flooding. It has been observed that the highest methane emissions occur after a flooding event (Smyth et al. 2019). A comparison of soil oxygen and soil moisture levels between 2019 and 2020 could have provided valuable insights into the key factors influencing methane production.

Additionally, Johnson et al. (2021) found that treated plots caused an increase in methane production compared to areas with cattails only. Johnson et al. (2021) conducted a five-year study and found that total methane rates increase persisted at least a year after harvesting cattails and replanting. Johnson et al. (2021) studied a lacustrine open-embayment wetland on northern Lake Huron in Michigan. Lake Huron is also facing *Typha* spp. invasion like Lake Ontario. The *Typha* spp. invaded area in Lake Huron was restored using two different mechanical methods: harvesting and crushing. Specifically, Johnson et al. (2021) measured carbon fluxes in July and August 2017 using a 0.25 m x 0.25 m x 2-m tall PVC-framed chamber wrapped with transparent UV-resistant PVC film, set on a floating base placed over the plants between 10 am and 3 pm. This was a similar setup to my experiment, except I sampled for 6-minute intervals and Johnson et al. (2021) sampled for 10-minute intervals. However, my 2019 and 2020 control and treatment plots showed higher methane flux than what Johnson et al. (2021) observed in 2017. In July 2017, Johnson et al. (2021) observed approximately 20 mg $CH_4/m^2/hr$ in the cattail control areas, approximately 25 mg $CH_4/m^2/hr$ within the crushed treatment areas, and approximately 50 mg $CH_4/m^2/hr$ within the harvested treatment areas. These differences could be due to the time since treatment occurred. The time between treatment and methane sampling in Braddock Bay was approximately 10 months, while three years had passed since first treatment in Johnson et al. (2021)'s study. Similarly, repeated mechanical treatment of non-native *Typha* may increase the availability of organic substrates. Repeated mechanical treatment, coupled with prolonged flood conditions as in 2019, generates ideal conditions for methanogenesis (Johnson et al. 2021).

After reviewing a study by Keyport et al. (2018) and conducting my own research, there are significant differences in the methodologies and conditions of the studies, which could explain the discrepancies in the results. In Keyport et al. (2018) found that treating cattails did not have a significant effect on carbon fluxes. It is important to note that this study measured soil-only flux, not the total flux (which includes both soil and plant contributions) and was conducted under lower water levels. In contrast, my samples were collected during the peak methane production and release, rather than being collected hourly throughout the entire day as in Keyport et al.'s (2018) study. As a result, my values could be inflated due to sampling only during peak hours.

Carbon Availability and Methane Production

I discovered evidence supporting my hypothesis that the restored sites would exhibit higher methane emissions compared to native meadow marsh sites due to the presence of carbon-rich cattail substrate. My research revealed that the treated areas demonstrated higher methane production than the native meadow marsh plots, and significantly lower methane production than the control sites over the two-year period. Furthermore, the treated plots exhibited increased soil organic carbon (SOC) compared to the meadow marsh plots. It is worth noting that during the period of record high water levels in 2019, all three types of plots did not show significant differences from each other. This suggests that water depth and the duration of flooding may have a more pronounced impact on methane emissions than the legacy carbon present in the soil under extreme conditions, while plant type impacts methane emissions under more normal conditions.

Similarly, other research has found that *Carex* spp. stands had lower methane emissions than *Typha* spp. stands (Gunther et al., 2014). Furthermore, they discovered that harvesting *Typha* spp. led to a slightly larger net increase in emissions (Gunther et al. 2014). However, this research focused solely on soil flux, rather than considering both soil and plant fluxes together. Evaluating the system as a whole can lead to different results which is likely why my research indicated that the meadow marsh stand had lower emissions than both the cattail and treated plots during normal to higher-than-average lake levels.

Sources of Error

As previously mentioned, in 2019, Lake Ontario had record high water levels. Plots in Braddock Bay could not be safely accessed by foot. Samples at Braddock Bay in 2019 were taken from kayak and plots were marked with ribbon and GPS coordinates. Record high water levels could have also affected Braddock Bay SOC% values due to collecting soil from under water. Due to the depth of the water, a floating chamber had to be utilized at this site. While the dimensions of the floating chamber and the non-floating chamber were the same, discrepancies between floating chamber, regular chamber with soil collar, and regular chamber without soil collar may have occurred. Additionally, while the lake levels were lower in 2020 than in 2019, floating chambers were still utilized at Braddock Bay at some plots. Water levels were low enough to safely access by foot with waders but walking within the wetland releases methane bubbles. While we were careful not to move our feet once we were sampling the plots, gas bubbles could have been the cause of some high methane emissions in the Braddock Bay, Buck Pond, and Salmon Creek. Any plot which did not have a soil collar installed could have also had higher than usual methane emissions from when we tucked the clear plastic under the water to create an airtight seal. Any soil disturbance from that action could have released gas into the chamber.

Conclusion

Water depth appears to affect methane emission regardless of SOC, aboveground biomass accumulation, or plant community. During the record high flooding event of Lake Ontario in 2019, all plots were inundated with water throughout the growing season. This research shows that water depth was the greatest factor in predicting CH_4 emissions in 2019 when there was excessive flooding. Extensive flooded conditions over the growing season provide optimal conditions for methanogenesis (Johnson et al. 2021). Consistent with other mechanical management studies, I found that during high-water conditions, treatment areas had lower aboveground biomass than untreated areas (Wilcox et al. 2018). While treatment areas generally have lower aboveground biomass than cattail monocultures regardless, when extreme flooding occurs, the treatment areas were observed with even less aboveground biomass than in regular water level years.

Treating cattail monoculture reduces soil organic carbon and aboveground biomass when compared to cattail invaded areas. Johnson et al. (2021) found emissions were higher the more recent the treatment. Areas where recent treatment has occurred might exhibit higher residual methane emissions compared to older areas. This could be due to a decrease in the availability of soil organic carbon over time. However, more research is recommended to explore how repeated, long-term harvest impact belowground biomass and CH_4 emissions.

In my study sites, the treatment within Braddock Bay was completed in the fall of 2018, just before research began. Buck Pond treatment areas were completed two years prior to Braddock Bay. Even though these treatments were relatively recent, I found that cattail treatment reduced methane release. My research findings suggest that treatment of areas with cattail monocultures can result in lower greenhouse gas emissions, which can potentially slow the effects of climate change. Additionally, while this research investigated the relationship of CH_4 flux between treated areas with cattail monocultures, further research on how the length of time after treatment affects CH_4 emissions is recommended. I also suggest continuing this research to collect more data during similar water level years. The exceptionally high levels of flooding that occurred recently might have impacted the accuracy of the biomass and methane data from Salmon Creek. Additionally, it's possible that this flooding has introduced further errors into the process of gas sampling. This research suggests that restoring these marshes may reduce greenhouse gas emissions in coastal wetlands throughout Braddock Bay WMA and the Great Lakes region.

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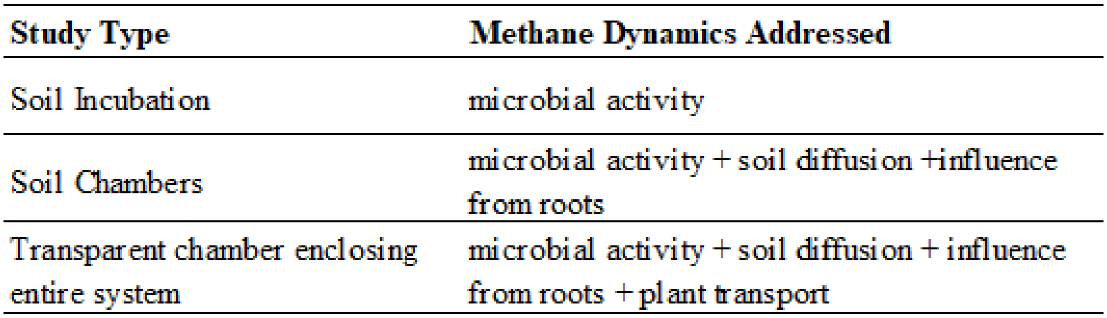
Tables and Figures

Table 1.1. Methane dynamics are addressed by soil incubation, soil chamber and transparent chamber studies.



Study Type	**Methane Dynamics Addressed**
Soil Incubation	microbial activity
Soil Chambers	microbial activity + soil diffusion +influence from roots
Transparent chamber enclosing entire system	microbial activity + soil diffusion + influence from roots + plant transport

Table 1.2. 2019 methane flux mixed effects model results including degrees of freedom (DF) and parameter estimates on $\log_{10}$ transformed methane values. Plot type (treated vs control), Period (July vs August), Site (Braddock Bay vs Buck Pond).

Predictors	Estimates	df	CI	p
fixed effects				
(Intercept)	0.02	32	0.01 - 0.03	0.002
Plot Type	0.01	32	0.00 - 0.02	0.253
Site	0.01	32	0.00 - 0.03	0.012
Period	0.00	21	-0.02 - 0.01	0.554
Plot Type x Site	-0.01	32	-0.01 - 0.02	0.300
random effect				
$N_{\text{Plot ID}}$	36			
Observations	58			
*Flux ~ plot type*site + period + error (Plot ID)*				

Table 1.3. 2020 methane flux mixed effects model results including degrees of freedom (DF) and parameter estimates on $\log_{10}$ transformed methane values. Plot type (treated vs control), Period (June vs. July), Site (Braddock Bay vs Buck Pond).

Predictors	Estimates	df	CI	p
fixed effects				
(Intercept)	2.01	34	1.54 - 2.47	<0.001
Plot Type	-0.61	34	-1.18 - -0.04	0.037
Site	-0.05	34	-0.67 - 0.56	0.859
Period	-0.25	27	-0.59 - 0.09	0.141
Plot Type x Site	-0.04	34	-0.86 - 0.77	0.915
random effect				
$N_{\text{Plot ID}}$	38			
Observations	66			
*Flux ~ plot type*site + period + error (Plot ID)*				

Table 1.4. 2019 average methane flux mixed effects model results including degrees of freedom (DF) and parameter estimates on $\log_{10}$ transformed methane values. Water depth (cm), soil organic carbon (%), biomass (g carbon/m^2), Plot type (treated vs control), Site (Braddock Bay vs Buck Pond), soil temperature (°C).

Predictors	Estimates	df	CI	p
(Intercept)	2.06	29	0.67 - 3.45	0.005
Waterdepth	0.01	29	0.01 - 0.02	<0.001
SOC	-0.01	29	-0.03 - 0.00	0.068
Biomass	0.00	29	0.00 - 0.00	0.244
Plot Type	0.01	29	-0.19 - 0.20	0.955
Site	0.36	29	-0.19 - 0.21	0.064
Soil Temperature	-0.04	29	-0.09 - 0.01	0.146
Observations	36			
R^2/R^2 adjusted	0.475 / 0.366			

Flux ~ waterdepth + SOC + Biomass + plot type + soil temperature + error (PlotID)

Table 1.5. 2020 average methane flux mixed effects model results including degrees of freedom (DF) and parameter estimates on $\log_{10}$ transformed methane values. Water depth (cm), soil organic carbon (%), biomass (g carbon/m^2), Plot type (treated vs control), Site (Braddock Bay vs Buck Pond), soil temperature (°C).

Predictors	Estimates	df	CI	p
(Intercept)	0.44	31	-3.32 - 4.19	0.815
Waterdepth	0	31	-0.04 - 0.04	0.957
SOC	0.02	31	-0.01 - 0.04	0.282
Biomass	0.00	31	0.00 - 0.00	0.241
Plot Type	0.06	31	-0.70 - 0.81	0.883
Site	-0.14	31	0.70 - 0.41	0.597
Soil Temperature	0.03	31	-0.08 - 0.14	0.624
Observations	38			
R^2/R^2 adjusted	0.175 / 0.015			

Flux ~ waterdepth + SOC + Biomass + plot type + soil temperature + error (PlotID)

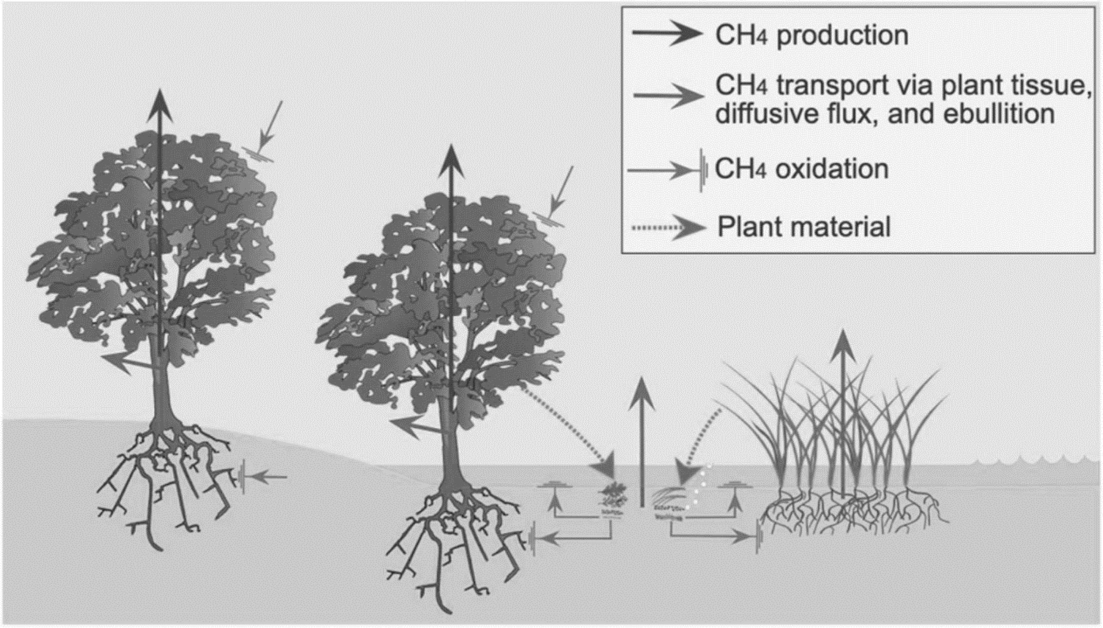


Figure 1.1. Examples of how plants and other primary producers can influence terrestrial and aquatic ecosystem CH4 generation, oxidation, and transport to the atmosphere (Bastviken et al. 2023).

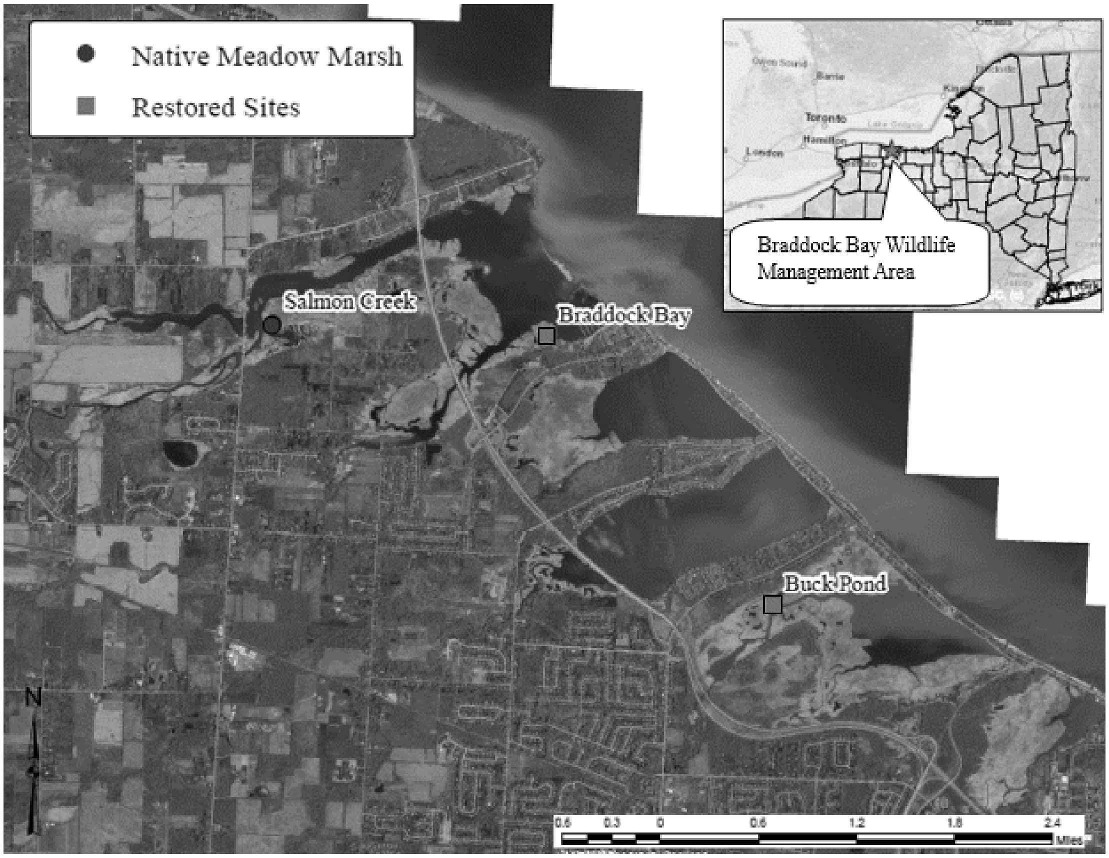


Figure 1.2. Location of study sites in the Braddock Bay Wildlife Management Area west of Rochester, NY. Braddock Bay, and Buck Pond have been restored, including cattail removal, and Salmon Creek contains a native meadow marsh.

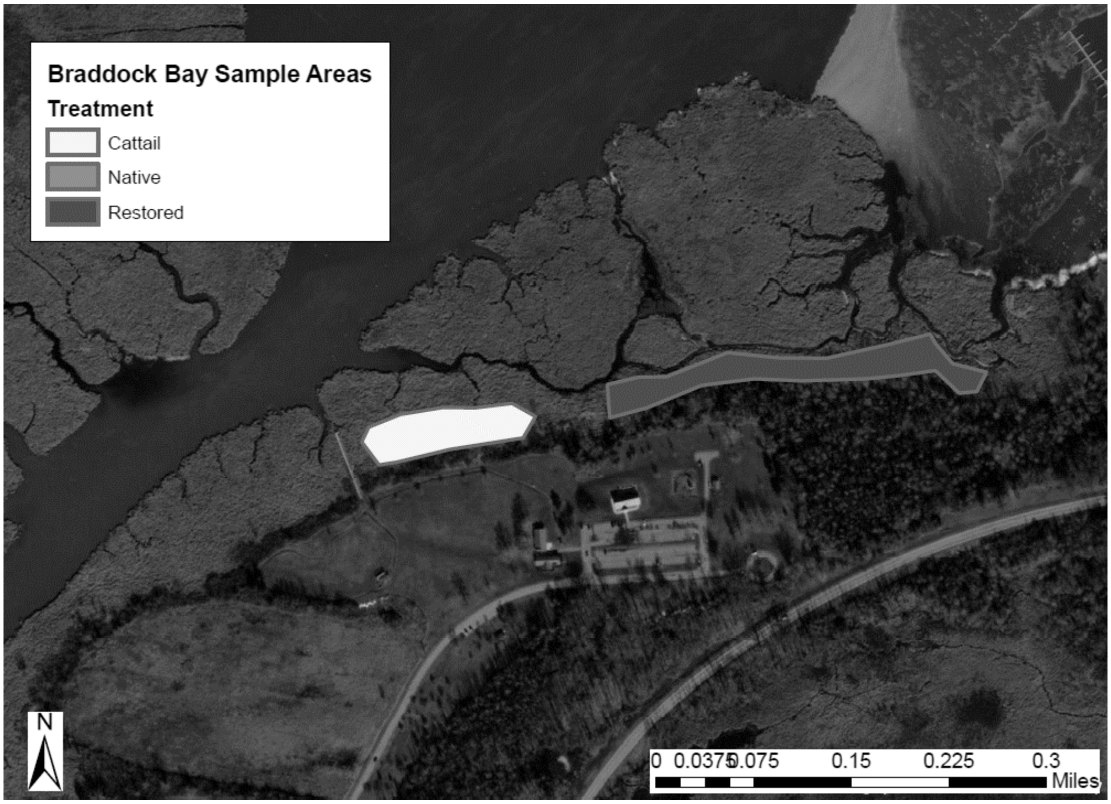


Figure 1.3. Braddock Bay cattail monoculture and restoration sample areas in 2019 and 2020.

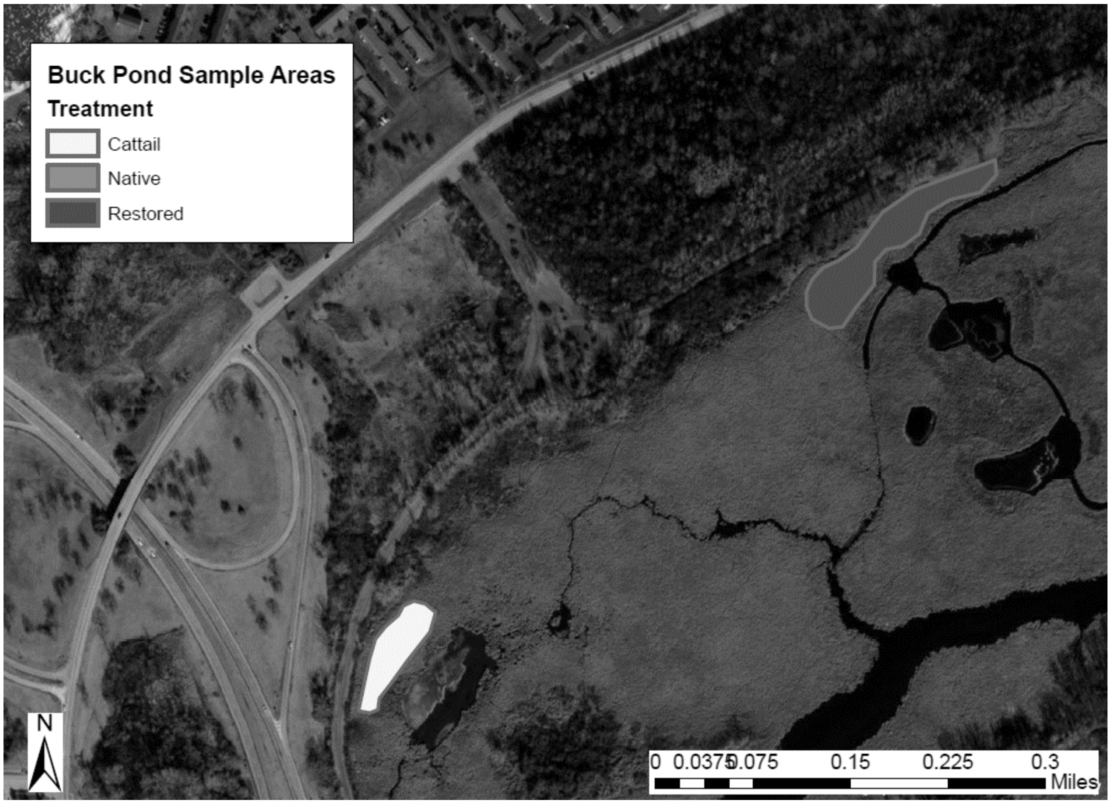


Figure 1.4. Buck Pond cattail monoculture and restoration sample areas in 2019 and 2020.

Figure 1.5. Salmon Creek native meadow marsh sample area in 2019 and 2020.

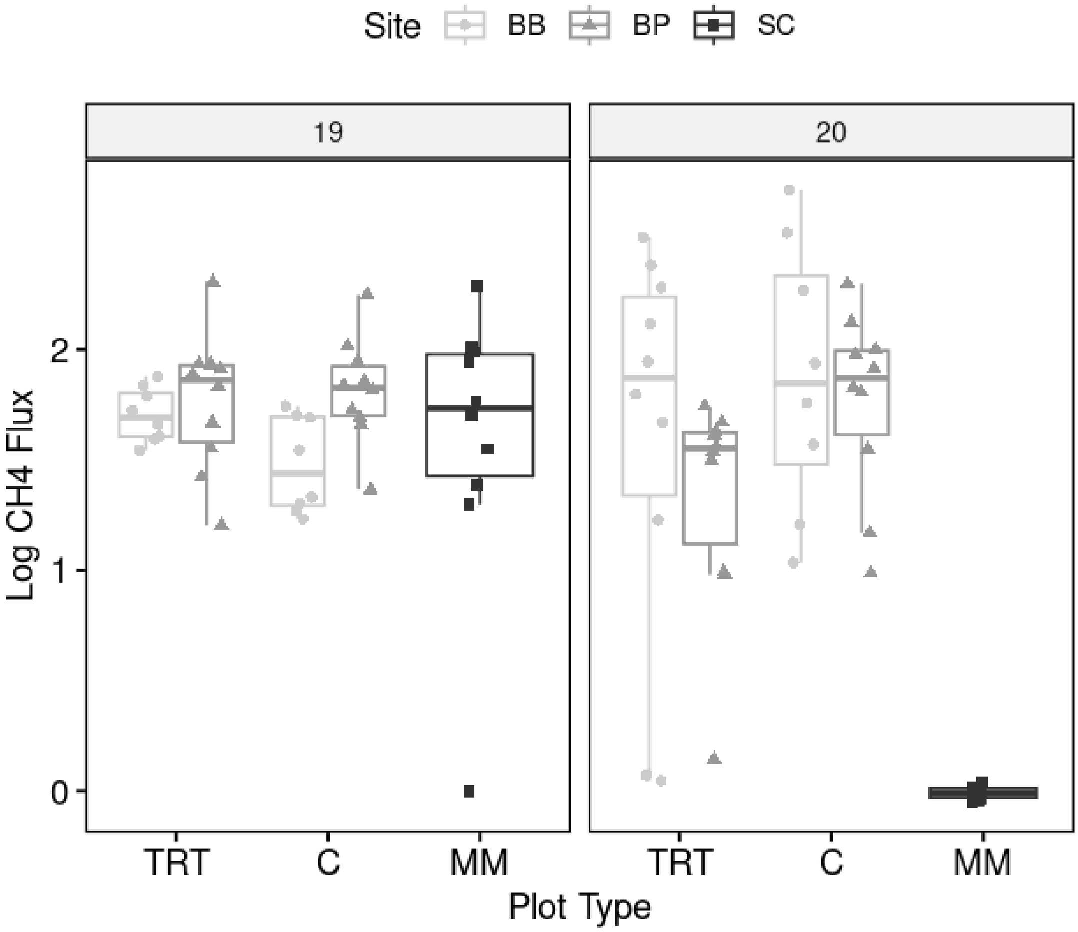


Figure 1.6. Methane flux values by plot type (cattail monoculture (C), cattail removal and replanting (TRT) and uninvaded native sedge meadow (MM)) in 2019 and 2020. Box plots represent median (bold line), box parameter is the upper and the lower quartiles and the lines represent the range in values up to two standard deviations from the mean, dots are values beyond two standard deviations from the mean, within Salmon Creek (SC), Braddock Bay (BB) and Buck Pond (BP).

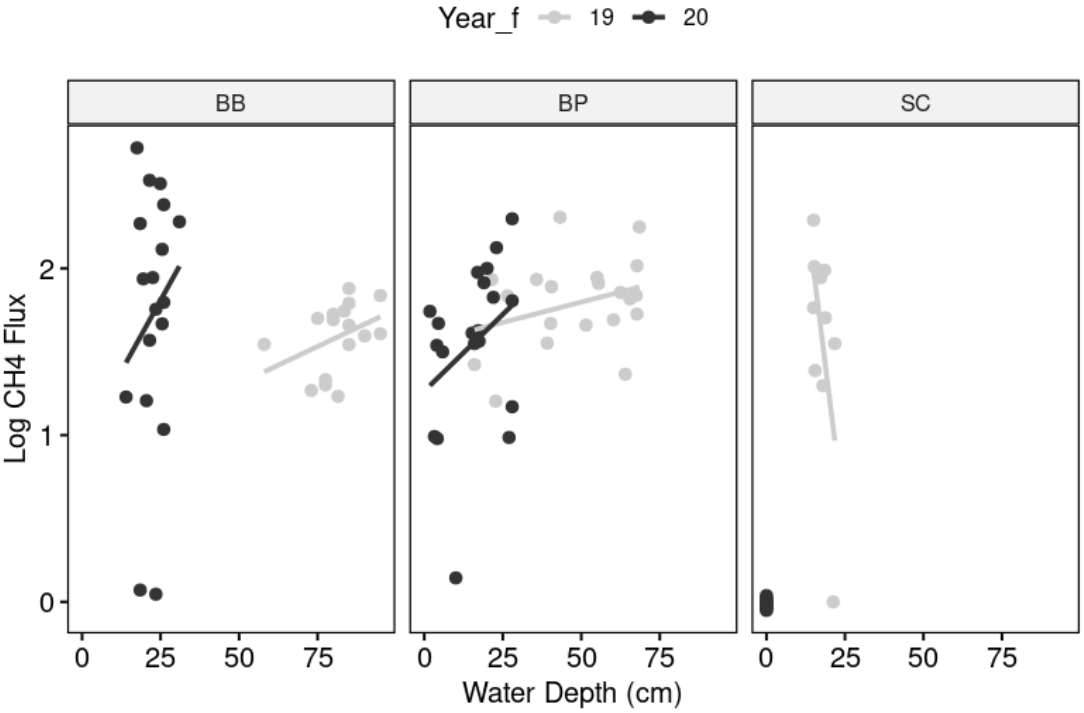


Figure 1.7. Methane flux values by water depth in 2019 and 2020. Scatterplot relationship between methane flux values and water depth in cm within Salmon Creek (SC), Braddock Bay (BB) and Buck Pond (BP).

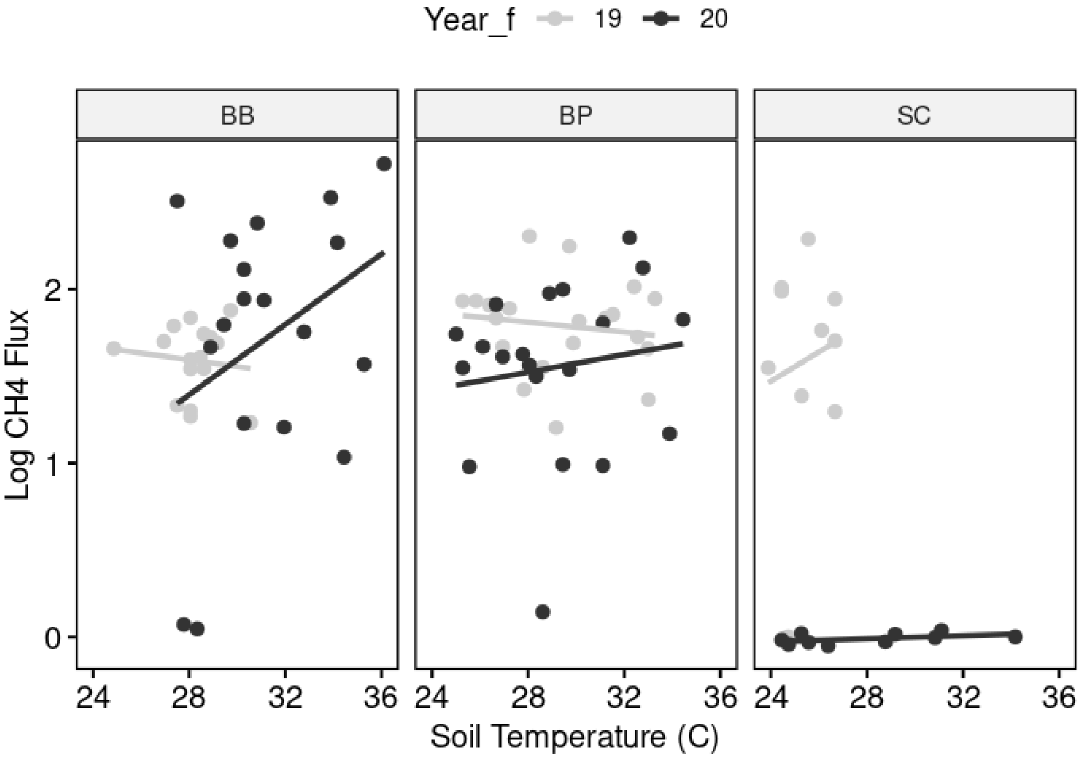


Figure 1.8. Methane flux values by soil temperature in 2019 and 2020. Scatterplot relationship between methane flux values and water depth in cm within Salmon Creek (SC), Braddock Bay (BB) and Buck Pond (BP).

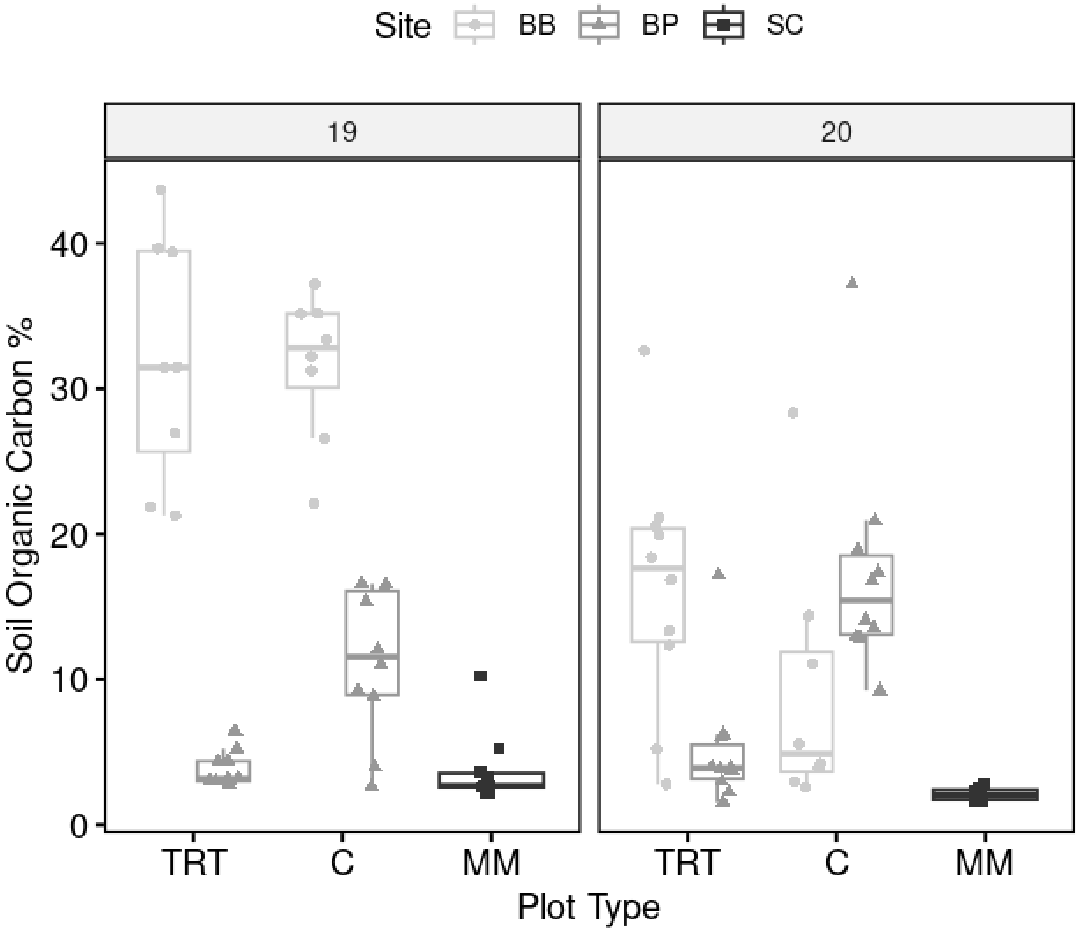


Figure 1.9. Soil organic carbon (SOC) % values by plot type (cattail monoculture (C), cattail removal and replanting (TRT) and uninvaded native sedge meadow (MM)) in 2019 and 2020. Box plots represent median (bold line), box parameter is the upper and the lower quartiles and the lines represent the range in values up to two standard deviations from the mean, dots are values beyond two standard deviations from the mean, within Salmon Creek (SC), Braddock Bay (BB) and Buck Pond (BP).

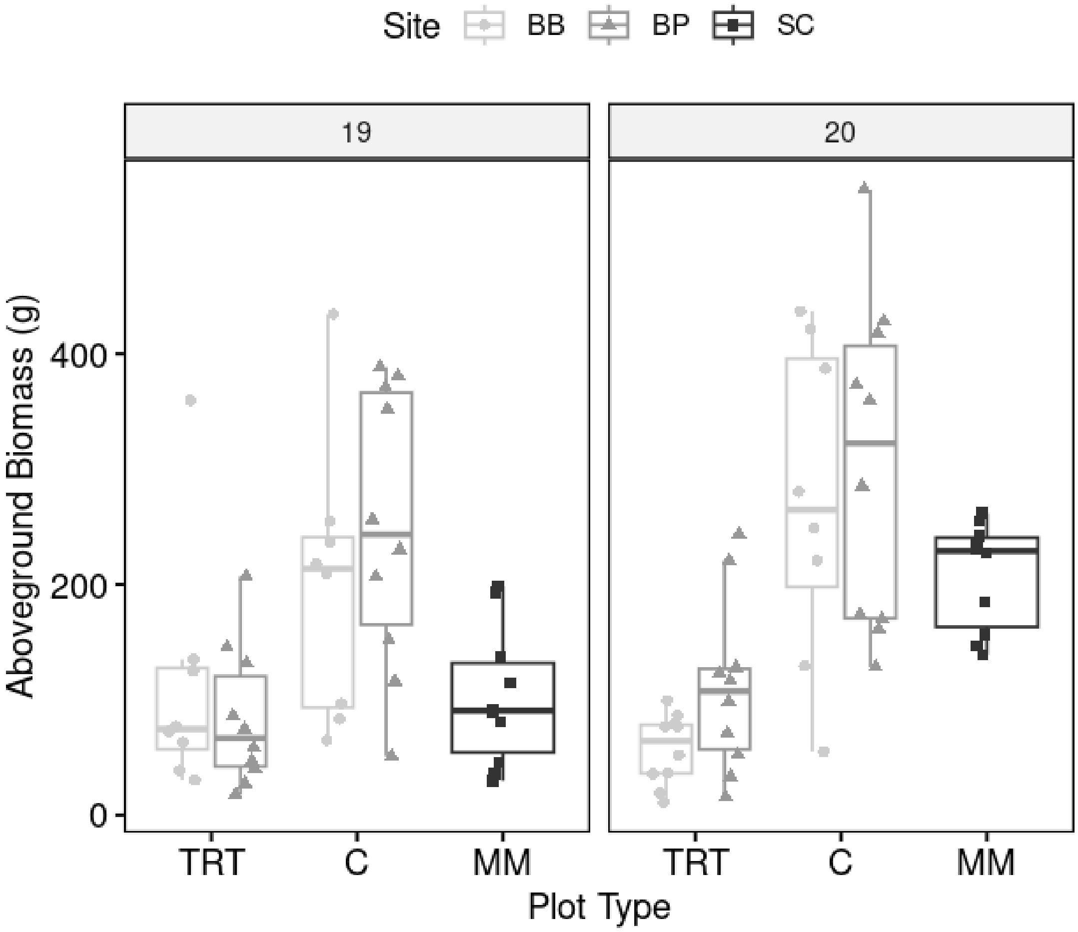


Figure 1.10. Aboveground biomass (g Carbon/m^2) values by plot type (cattail monoculture (C), cattail removal and replanting (TRT) and uninvaded native sedge meadow (MM)) in 2019 and 2020. Box plots represent median (bold line), box parameter is the upper and the lower quartiles and the lines represent the range in values up to two standard deviations from the mean, dots are values beyond two standard deviations from the mean, within Salmon Creek (SC), Braddock Bay (BB) and Buck Pond (BP).

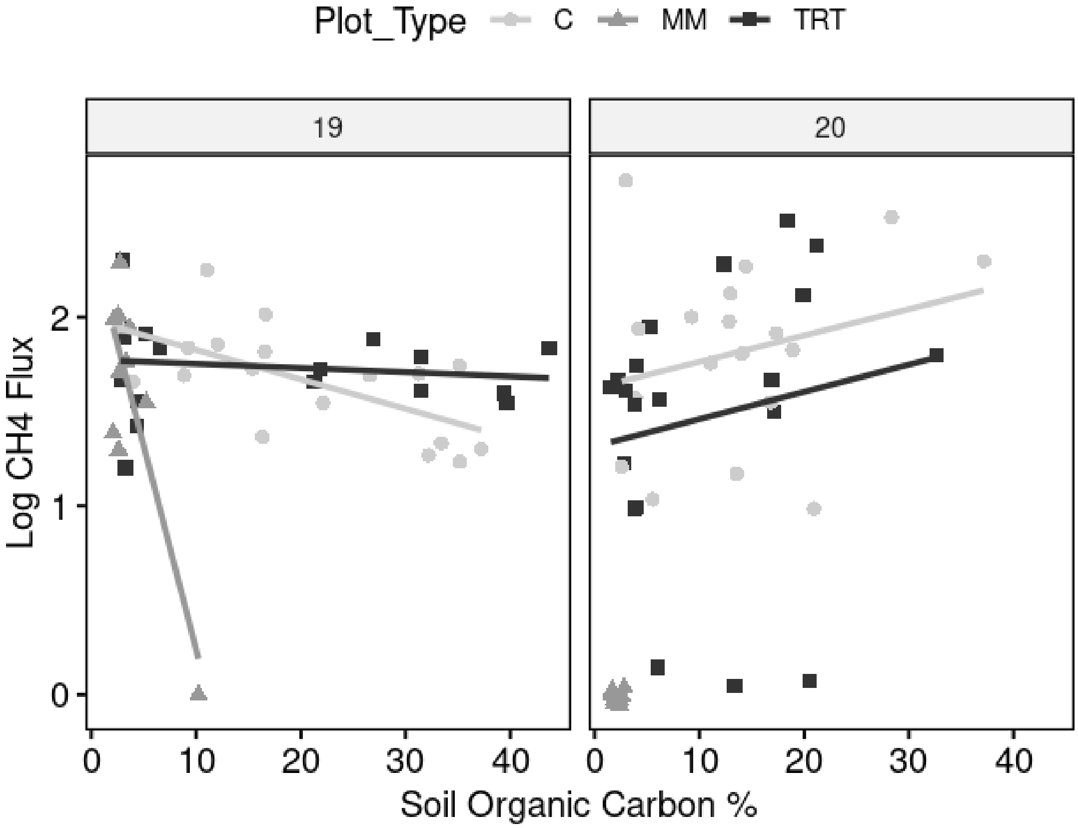


Figure 1.11. Methane flux values by soil organic carbon percent. Scatterplot relationship between methane flux values and SOC at each plot in percent in 2019 (left) and 2020 (right). Cattail monoculture (C), uninvaded native sedge meadow (MM), and cattail removal and replanting (TRT).

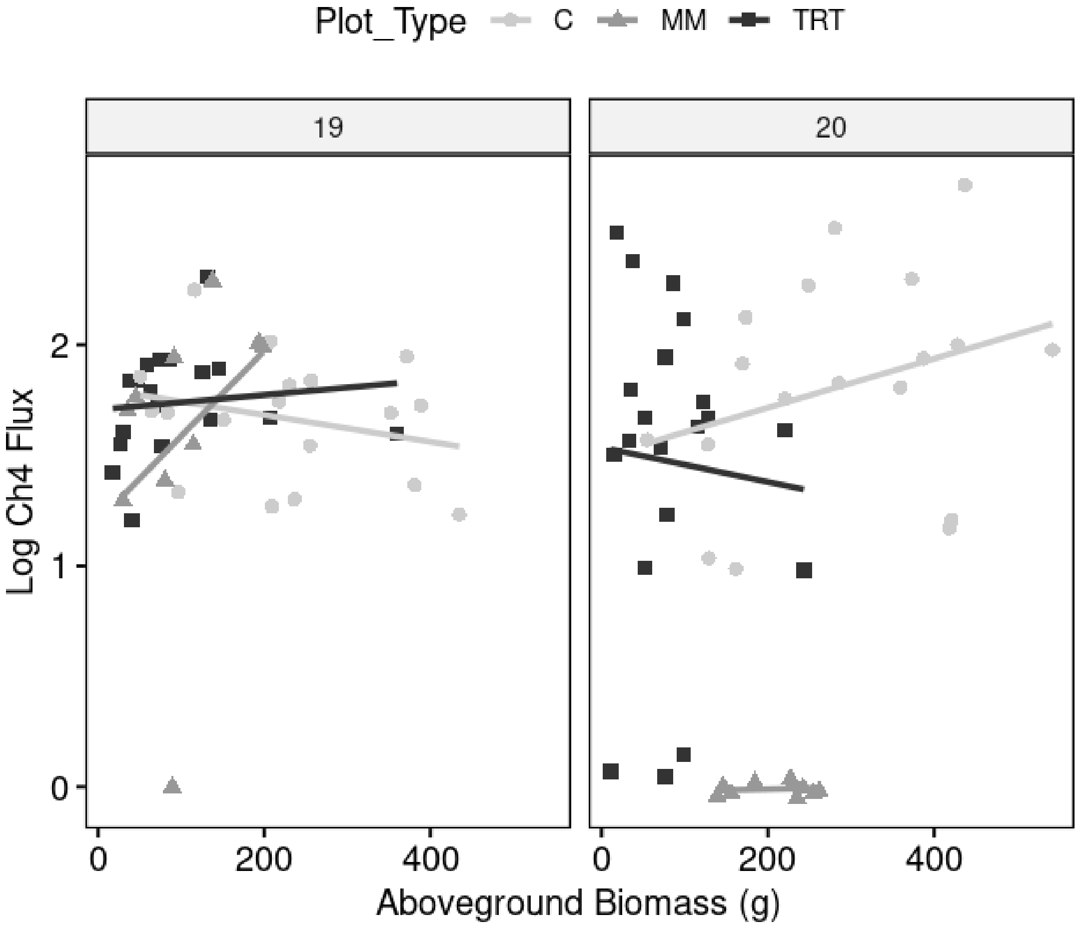


Figure 1.12. Methane flux values by aboveground biomass. Scatterplot relationship between methane flux values and aboveground biomass carbon content (g/m^2) at each plot in 2019 (left) and 2020 (right). Cattail monoculture (C), uninvaded native sedge meadow (MM), and cattail removal and replanting (TRT).

Chapter Two – Mesocosm Study

Introduction

Plants have a significant impact on methane production and oxidation due to their varying characteristics. According to Williams and Yavitt (2008), the type of plant strongly influences methanogenesis in soil, and the effects of plants on microbial activity are directly related to the composition and decomposability of their leaves and roots. As wetlands develop, the plant species present change, leading to differences in the composition of current vegetation compared to that in the soil (Williams and Yavitt 2008). The diversity of plant species, variations in plant physiology, and differences in organic matter all contribute to the regulation of methane production, although the specific interactions between soil, microbes, and plant species are still not fully understood (Williams and Yavitt 2008). Research has indicated that aerenchyma within plants acts as a conduit for gas to move between the soil and the atmosphere (Whalen 2005, Kao-Kniffin et al. 2010). Differences and similarities in plant physiology and traits have been organized into functional groups.

Functional Groups

Functional groups are species that interact with their environment, use resources, and grow similarly to others. Plants that use resources and function similarly are grouped together. Boutin and Keddy (1993) identified three main functional groups, which they further divided into guilds. Guilds are a group of functionally similar species in a community. The two groups of interest are the clonal dominants, such as cattails, which have vigorous clonal spread, deep rooting, and are tall, and the tussock-forming group, including species like sedges and grasses, which are tall, have a high photosynthetic area, and have a compact growth form with shallow roots (Figure 2.1, Boutin and Keddy, 1993).

There have been studies that examined the impact of functional groups on methane dynamics. Bouchard et al. (2007) found that an increase in the number of functional groups decreased methane flux by enhancing belowground biomass and altering root patterns. They also found that a reduction in diversity led to an increase in methane emissions. Schultz and Pett (2018) found that the presence of reed and tussock functional groups decreased methane fluxes and increased carbon storage, indicating that planting those functional groups could maintain lower methane emissions and high levels of carbon storage. Kao-Kniffin et al. (2010) found that functional groups and plant biomass are useful in predicting methane flux differences across plant species due to differences in aerenchyma tissue porosity.

Objectives and Hypotheses

This experiment manipulated the organic layer of soil and plant species to answer the question if and how much substrate and species matters to methane flux. Two types of soil were used; substrate with a thick organic matter layer similar to a site invaded by cattail, and the previous substrate with the organic matter removed. The removal of approximately 5 cm of accumulated organic matter was to simulate substrate in a meadow marsh. In the mesocosm experiment my objectives were to 1) measure methane fluxes in six treatments to determine how substrate and plant species effects methane emissions, and 2) determine carbon storage in aboveground biomass. These objectives led to the following hypotheses: mesocosms with removed carbon-accumulated organic matter and native plant species will have lower methane emissions than mesocosms with invasive cattail on carbon-accumulated substrate.

Methods

Experimental Design

The mesocosm was located at the SUNY Brockport aquaculture ponds in Brockport, NY. There were six treatments with five replicates each for a total of 30 replicates (Table 2.1). The mesocosms were 5-gallon buckets with plumbing to create a flooding regime like that of Lake Ontario. The buckets were placed on top of two-high wood pallets to reduce weeds in the mesocosm (Figure 2.2). Each bucket was connected by half-inch Teflon tubing, which was connected to a large watering basin. The water level was kept at a similar level by gravity feeding from the large watering basin into the 30 buckets. Water from one of the aquaculture ponds was pumped into the large basin for use.

The six treatments included no plants on cattail substrate, no plants on 10cm organic layer removed substrate, cattail on cattail substrate, native species on cattail substrate, cattail on 10 cm organic layer substrate, and native species on 10 cm organic layer (Table 2.1). The 10 cm organic later substrate simulated a soil pre-invasion, and the cattail substrate is soil post-invasion. The soil was collected carefully from the field and reduced the chances of homogenization by using shovels and turf cutters. The soils were collected from Buck Pond, in Greece, NY from the same cattail monoculture area that was sampled in Chapter 1 – Field Study. Approximately 26 cm of soil was excavated. Soil layers were evaluated, and the organic accumulation was removed (approximately three to five cm) for 15 of the mesocosms. All cattails were collected from Buck Pond as well. When collecting the soil, at least ten soil collections had sprouting cattail shoots, which were left in the soil to grow in the mesocosm.

For the native species, I planted Canada bluejoint (*Calamagrostis canadensis)*, a tussock forming grass. The seedlings were purchased from Southern Tier Consulting Inc. nursery in Cuba, NY which has the same climate zone as Brockport, NY; Zone 6. Similar measurements from Chapter 1 – Field Study were taken for the mesocosm experiment: methane flux, aboveground biomass, soil organic material, and soil moisture. The mesocosm was built, planted, and watered in April 2020 and gas sampling took place in September 2020 which gave the plants time to establish in the mesocosm.

Methane Sampling

Methane flux was measured using a portable gas chromatograph with closed chambers systems similar to Schultz et al. (2018). No soil collars were used in the mesocosm experiment. Instead, an airtight seal was created using the excess clear plastic sheeting and an elastic tied around each bucket. Methane flux was measured in

September using a similar, airtight clear chamber fitted over the vegetation. The clear chamber for the mesocosm was made from a 30.5 x 30.5 x 152.5-cm PVC frame with clear plastic sheets. Inside the chambers were an icepack and fan to reduce the likelihood of temperature increasing more than the ambient.

The same Picarro GasScouter cavity ring down spectrometer (G4301, Picarro) was used to measure the methane flux in the mesocosm as in Chapter One. The chamber was connected to the GasScouter with Teflon tubing for six minutes at a time, under full sun conditions, between 10 am and 2 pm. Full sun conditions were measured using a Venier Lab Quest 2 photosynthetically active radiation (PAR) sensor. As mentioned in Chapter One, photosynthesis can occur between 400 and 700 nanometers, and the PAR sensor measures 0-2500 $\mu mol/m^2/s$ (370-640 nm). 300 $\mu mol/m^2/s$ was the smallest acceptable measured value. Air and soil temperature were also recorded at each plot using a thermometer connected to Lab Quest 2; and water depth was measured using a meter stick.

Belowground Environmental Conditions

Soil oxygen was measured in August and September since soil oxygen is an important factor in determining methane flux in wetlands (Smyth et al. 2019). One custom "Arduino" data logger programed by the SUNY Brockport Physics Department was installed at the mesocosm site. This was the second custom-built data logger which had three Apogee SO-411 soil oxygen sensors. Data was logged in August and September. Once collected after sampling flux in September, data was downloaded from the data loggers and average soil oxygen were graphed over the growing season by date.

Aboveground Biomass and Soil Carbon

To measure the aboveground biomass a similar process to that of Chapter One was used. All plant material in the mesocosms were cut at soil level in September at peak biomass, oven dried for 72 hours at 55 degrees Celsius, and weighed in grams. After drying, the plants were homogenized, split into two subsamples per mesocosm, and weighed in grams (40 total subsamples). Only 20 mesocosms had plant material growing in them out of the 30 total mesocosms. The subsamples were burned in a muffle furnace at 550 degrees Celsius for three hours, cooled, and weighed in grams to determine loss on ignition. Subsample values were averaged together. Grams of carbon per meter squared was calculated using the following formula:

$$\frac{\text{total plot oven dried weight} \times \text{average percent carbon}}{\text{plot area}}$$

Soil carbon was determined by using the muffle furnace, loss on ignition method (Salehi et al. 2011). The amount of carbon storage in the substrate was determined from harvesting one soil core in each mesocosm at the end of the season. Soil samples were collected using a soil auger from zero to 16 cm in depth. The soil samples were split into two subsamples, oven dried for 72 hours at 105 degrees Celsius, and weighed in grams. The subsamples were then combusted in a muffle furnace at 360 degrees Celsius for two hours, cooled, and weighed to determine carbon loss on ignition. Subsample values were averaged together.

Statistics and Data Analysis

Methane fluxes were calculated using R ‘flux’ package (Jurasinski et al. 2014). The R ‘flux’ package calculates gas flux using the date, time, Plot ID, temperature, water depth, chamber volume, chamber area, and methane parts per billion. Raw data was downloaded from the Picarro Gas Scouter and compiled into a spreadsheet by site and month. Using R, the data was partitioned into data tables per chamber measurement, then flux was calculated for each mesocosm. Flux data was downloaded and compiled into a combined spreadsheet containing the plot ID, treatment, plant community, soil substrate, CH_4 units, flux, air temperature, aboveground biomass, and soil organic carbon. Normality of the data was determined using Shapiro-Wilk. The data was transformed using $\log_{10} + 3$ to avoid nonzero values. Once the data was normal, a factorial design ANOVA was used to determine if there was a significant effect of plant type, soil type, or their interaction. A Tukey’s test was also performed on the data to determine which of those factors were significantly different from one another, and the direction. Additionally, a linear model to find significant influence on methane emissions among functional group, soil substrate, aboveground biomass, and water depth was performed in R. Scatterplots were created to view aboveground biomass vs methane flux, soil organic carbon vs methane flux, aboveground biomass vs soil organic carbon; and a boxplot was created to view the range, standard deviation, and mean methane flux vs treatment.

Results

Methane Sampling

Methane fluxes within the 30 mesocosms were collected on September 4, 2020, and September 9, 2020. 15 mesocosms were sampled each day. Each mesocosm was only sampled once. There were 30 total mesocosms with four experiment types: No plant with removed soil, no plant with cattail soil, Canada bluejoint with removed soil, Canada bluejoint with cattail soil, cattail with removed soil, and cattail with cattail soil.

The lowest methane flux was -2.12 mg $CH_4/m^2/hr$ in a cattail with topsoil removed mesocosm and the highest flux was 4.34 mg $CH_4/m^2/hr$ in a no plant with cattail soil mesocosm. The highest average methane flux was 1.55 mg $CH_4/m^2/hr$ in a Canada bluejoint with cattail soil mesocosm and the lowest average methane flux was -0.39 mg $CH_4/m^2/hr$ in a no plant with topsoil removed mesocosm.

Mesocosms containing Canada bluejoint had a maximum of 3.71 mg $CH_4/m^2/hr$, minimum of -0.14 mg $CH_4/m^2/hr$ and average of 1.17 with a standard deviation of +/- 1.11 mg $CH_4/m^2/hr$. Mesocosms containing cattail had a maximum of 2.66 mg $CH_4/m^2/hr$, minimum of -2.12 mg $CH_4/m^2/hr$ and average of 0.53 +/- 1.53 mg $CH_4/m^2/hr$. Mesocosms containing no plants had a maximum of 4.34 mg $CH_4/m^2/hr$, minimum of -0.95 mg $CH_4/m^2/hr$ and average of 0.755 +/- 1.57 mg $CH_4/m^2/hr$. Plant community was not significantly different from one another (Table 2.2).

Mesocosms containing the cattail soil had a minimum methane flux of 0.18 mg $CH_4/m^2/hr$, maximum of 4.34 mg $CH_4/m^2/hr$, and average of 1.31 +/- 1.29 mg $CH_4/m^2/hr$. Mesocosms containing the topsoil removed soil had a maximum flux of 2.66 mg $CH_4/m^2/hr$, minimum of -2.12, and an average of 0.42 +/- 1.37 mg $CH_4/m^2/hr$. Soil type was determined to significantly affect methane flux (Table 2.2). The interaction between plant community and soil type was also found to be not significant (Table 2.3). Topsoil removed mesocosms had significantly less methane emissions than cattail soil mesocosms (Table 2.4, Figure 2.5).

Aboveground Biomass and Soil Carbon

The average soil organic carbon (SOC) percentage in the cattail soil mesocosms was 3.5% +/- 2.68 with a minimum value of 1.63% and maximum of 5.42%. The average SOC% in topsoil removed mesocosms was 2.80% +/- 0.72 with a range between 1.64% and 3.60%. The average SOC% in Canada bluejoint mesocosms was 3.92% +/- 1.37 with a range between 2.75% and 5.42%. The average SOC% in no plant mesocosms was 1.64%; and the average SOC% in cattail planted mesocosms was 2.54% +/- 0.79, with a minimum of 1.63% and maximum of 3.09%. There was no relationship between SOC% and methane flux in this experiment (Figure 2.4).

The average aboveground biomass accumulation (g Carbon/m^2) in Canada bluejoint mesocosms was 24.99 +/- 13.38 g/m^2 and ranged from 10.77 g/m^2 to 56.81 g/m^2. The average biomass in no plant mesocosms was 0 g/m^2. The average aboveground biomass in cattail planted mesocosms was 49.64 +/- 18.88 g/m^2, ranging from 24.63 g/m^2 to 76.79 g/m^2. The average aboveground biomass in topsoil removed mesocosms was 25.12 +/- 24.85 g/m^2, ranging from 0 g/m^2 to 76.78 g/m^2. The average aboveground biomass in cattail soil mesocosms was 17.83 +/- 21.24 g/m^2, ranging from 0 g/m^2 to 56.81 g/m^2. A linear mixed effects model was conducted using environmental data to predict methane flux (Table 2.3). No factors were found to be significant. Increased aboveground carbon appears to have an increased methane flux (Figure 2.3). There appeared to be a positive relationship between aboveground biomass and soil organic carbon (Figure 2.6).

Soil Oxygen Percentage

Soil oxygen data was logged in August and September and graphed over the two months (Figure 2.7). The mesocosms were kept in a flooded state. Average soil oxygen in August was 1.76% +/- 1.87, minimum of 0.32%, and maximum of 15.86%. The average soil oxygen in September was 0.75% +/- 0.41, minimum of 0.02% and maximum of 1.4%.

Discussion

In this mesocosm experiment, we explored the relationship between plant functional groups and soil composition. We know that differences in plant functional groups and soil organic matter can affect methane production and release (Whalen 2005, Williams and Yavitt 2008, Kao-Kniffin et al. 2010). Our objectives were to 1) measure methane fluxes in six treatments to determine how substrate and plant species affect methane emissions, and 2) determine carbon storage in aboveground biomass to test the following hypothesis: mesocosms with removed carbon-accumulated organic matter and native plant species will have lower methane emissions than mesocosms with invasive cattail on carbon-accumulated substrate.

Plant Community Effects on Methane

Based on my data, the type of plant in the experiment did not have a significant impact on methane flux. Contrary to my hypothesis, the native plant species (*C. canadensis*) did not show significantly lower methane production compared to *Typha* x *glauca*. Interestingly, mesocosms with *C. canadensis* had slightly higher methane flux than those with cattails, and mesocosms with plants in general had slightly higher methane flux than those without plants. According to Bhullar et al (2013), plants with higher root volume and larger biomass tend to create a greater "chimney effect," leading to higher methane production. This suggests that the tussock-forming *C. canadensis* should have produced less methane gas than the clonal-dominant *Typha* x *glauca*. Additionally, other studies have indicated that emissions can be reduced by maintaining a lower water table (Bhullar et al. 2013). However, during our experiment, the mesocosms were damaged by an unknown person, causing them to lose their constant inundation. This resulted in a spike in soil oxygen, which may have affected the results. We also analyzed aboveground biomass and water depth as covariates, but we were unable to analyze soil organic carbon due to a low sample size. Our findings revealed that aboveground carbon storage and water depth were not significant factors in determining methane flux. Although water depth was a significant factor in Chapter One of this thesis, the goal of this experiment was to maintain consistent water depth across all thirty mesocosms, with a maximum difference of four cm between them.

Soil Substrate Effects on Methane

I found evidence in support of my hypothesis that mesocosms with carbon-removed soil substrate would have significantly less methane emissions than carbon-rich soil substrate. The mesocosms containing the topsoil removed substrate (W), which had less carbon-accumulation than the cattail soil (T) was found to have on average 0.89 mg $CH_4/m^2/hr$ less than carbon-rich soil substrate. This is expected since it is known that there must be carbon available in the system to do methanogenesis and produce methane gas (William and Yavitt 2008). Other studies, such as Scott et al. (2024) found that soil carbon had a positive relationship with an increase in total aboveground biomass. I saw a similar trend within my topsoil removed (W) mesocosms. There were not enough samples in the cattail soil (T) to see any trends. Further research and more data are needed for this mesocosm experiment to further understand the relationship between carbon availability in soil and methane flux. Additionally, research has shown that increasing carbon in soil may contribute to invasive species spread, such as *Typha* spp., and may have an increase in atmospheric warming (Scott et al. 2024). During my experiment, I also did not find a significant interaction between plant community and soil substrate. This could potentially be addressed by having a longer establishment period prior to gas sampling.

Potential Sources of Error

There are a few sources of error that should be mentioned for this experiment. This experiment had a limited number of samples that were taken. A larger sample size would give more accurate results and a better snapshot of the interactions between soil and plant type. Gas sampling was scheduled to be taken multiple times in the season but was postponed by multiple factors. The Summer of 2020 was during a statewide lockdown due to COVID-19, which severely limited the amount of assistance I received. One undergraduate assistant and I dug and hauled 30 buckets of soil from Buck Pond which took a considerable amount of time, and once the mesocosms were set up, a few weeks later they were vandalized and destroyed. The mesocosms then had to be repotted into new buckets, re-watered, and left alone for a few weeks to hopefully see some results in September. Some data was lost as well. My undergraduate assistance came to an end in August, so biomass and soil samples were collected without assistance. 23 out of 30 SOC samples were either lost or not recorded. The small sample size for the SOC does not show an accurate depiction of trends, especially with the *Typha* soil treatments only having two samples. More samples and more data are needed for this experiment.

Conclusion

The main factors controlling methane emissions from wetlands are soil temperature, water depth, and the amount and quality of decomposable substrate (Moore et al. 1998, Christensen et al. 2003). In my experiment, I found that the impacts of soil organic matter may be more significant than aboveground plant organic matter. This information can be utilized by recommending that during wetland restoration, the mowed cattail should be removed from the site to reduce organic matter accumulation. Reduction in organic matter accumulation during the restoration process may shorten the time it takes to return to pre-invasion levels and further reduce overall methane emissions in treated areas. This experiment kept water depth constant, and only measured air temperature while gas sampling. If this experiment is repeated, I recommend a longer establishment period for the plants and to incorporate soil temperature into analysis. More research is required to better understand the impacts of different plant species under various soil substrates and carbon content (Kayranli et al. 2009). I recommend further research on methane fluxes for different wetland plant communities, soil substrate, aboveground biomass, soil temperature, and water depth to find which factors have the highest and lowest potential for releasing methane into the atmosphere. Reduction of greenhouse gas being emitted into the atmosphere will slow the effects of climate change and help reach future New York’s climate goal.

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Tables and Figures

Table 2.1. Mesocosm design with five replicates per plant species and soil combination.

Plant Species	Soil Substrate
Invasive cattail	cattail soil
	top-soil removed
Canada bluejoint	cattail soil
	top-soil removed
None	cattail soil
	top-soil removed

Table 2.2. Methane flux ANOVA results including degrees of freedom (DF) and parameter estimate on $\log_{10}$ transformed methane values. Plant (Canada bluejoint vs no plant vs cattail), Soil (cattail soil vs topsoil removed), Plant * Soil (Canada bluejoint * cattail soil vs Canada bluejoint * topsoil removed vs no plant * cattail soil vs no plant * topsoil removed vs cattail * cattail soil vs cattail * topsoil removed).

Source	DF	Mean Square	F value	P-value
fixed effects				
Plant	2	0.049	1.050	0.367
Soil	1	0.224	5.089	0.034
Plant x Soil	2	0.099	2.146	0.141
Residuals	22	1.016		
*Flux ~ plant * soil*				

Table 2.3. Methane flux ANOVA results including degrees of freedom (DF) and parameter estimate on $\log_{10}$ transformed methane values. Plant (Canada bluejoint vs no plant vs cattail), Soil (cattail soil vs topsoil removed), aboveground carbon (biomass), water depth.

Source	DF	Mean Square	F value	P-value
fixed effects				
Plant	2	0.033	0.758	0.483
Soil	1	0.153	3.546	0.076
Biomass	1	0.099	2.284	0.148
Waterdepth	1	0.022	0.520	0.480
Residuals	18	0.043		
Flux ~ plant + soil + biomass + waterdepth				

Table 2.4. Tukey Multiple Comparisons of Means results on $\log_{10}$ transformed methane values. Plant (Canada bluejoint vs no plant vs cattail), Soil (cattail soil vs topsoil removed), Plant * Soil (Canada bluejoint * cattail soil vs Canada bluejoint * topsoil removed vs no plant * cattail soil vs no plant * topsoil removed vs cattail * cattail soil vs cattail * topsoil removed).

Tukey multiple comparisons of means		
Plant	*Difference*	*p-value*
None-CALCAN	-0.136	0.370
TYPGLA-CALCAN	-0.104	0.552
TYPGLA-None	0.032	0.947
Soil	*Difference*	*p-value*
W-T	-0.181	0.037
Plant:Soil	*Difference*	*p-value*
None:T-CALCAN:T	0.007	1.00
TYPGLA:T-CALCAN:T	-0.131	0.96
CALCAN:W-CALCAN:T	-0.096	0.98
None:W-CALCAN:T	-0.422	0.07
TYPGLA:W-CALCAN:T	-0.163	0.81
TYPGLA:T-None:T	-0.137	0.95
CALCAN:W-None:T	-0.103	0.97
None:W-None:T	-0.429	0.07
TYPGLA:W-None:T	-0.169	0.78
CALCAN:W-TYPGLA:T	0.034	1.00
None:W-TYPGLA:T	-0.291	0.50
TYPGLA:W-TYPGLA:T	-0.032	1.00
None:W-CALCAN:W	-0.326	0.25
TYPGLA:W-CALCAN:W	-0.067	1.00
TYPGLA:W-None:W	0.259	0.45
*Flux ~ plant * soil*		

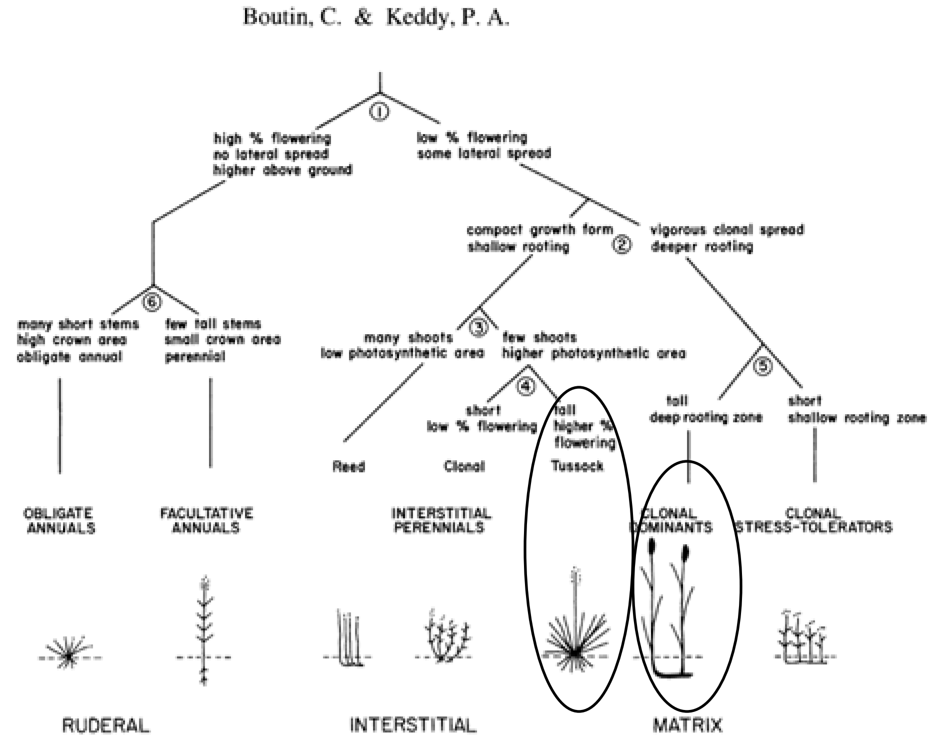

Figure 2.1: Boutin and Keddy (1993) diagram of functional groups and guilds with tussock and clonal dominant guilds circled.

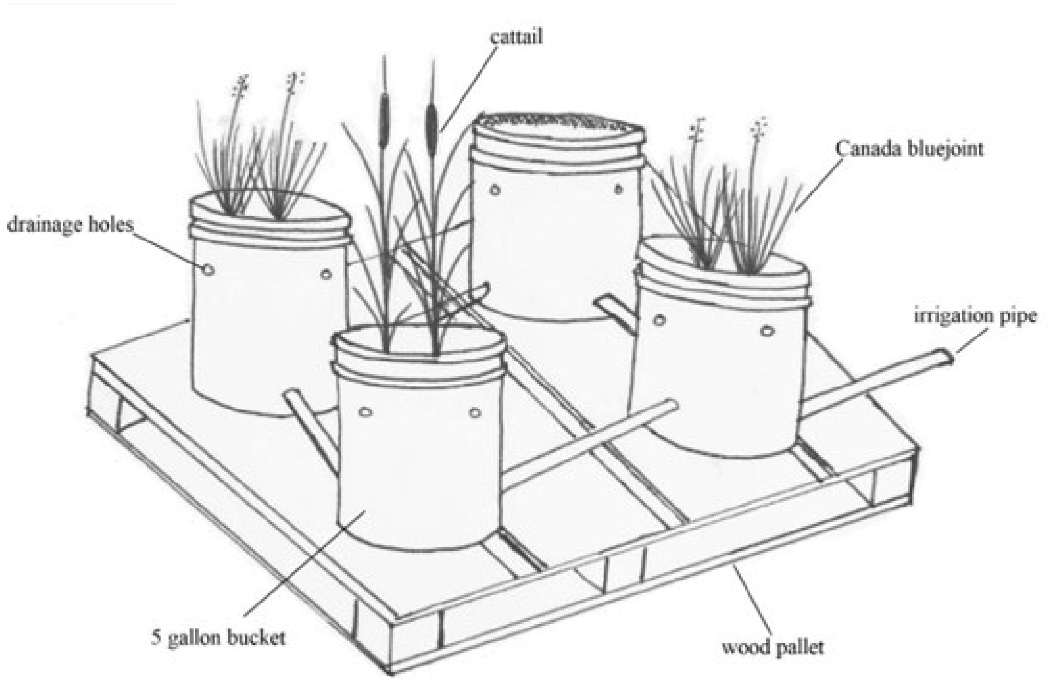

Figure 2.2. Diagram of proposed mesocosm set up with wooden pallet, and 5-gallon buckets with irrigation.

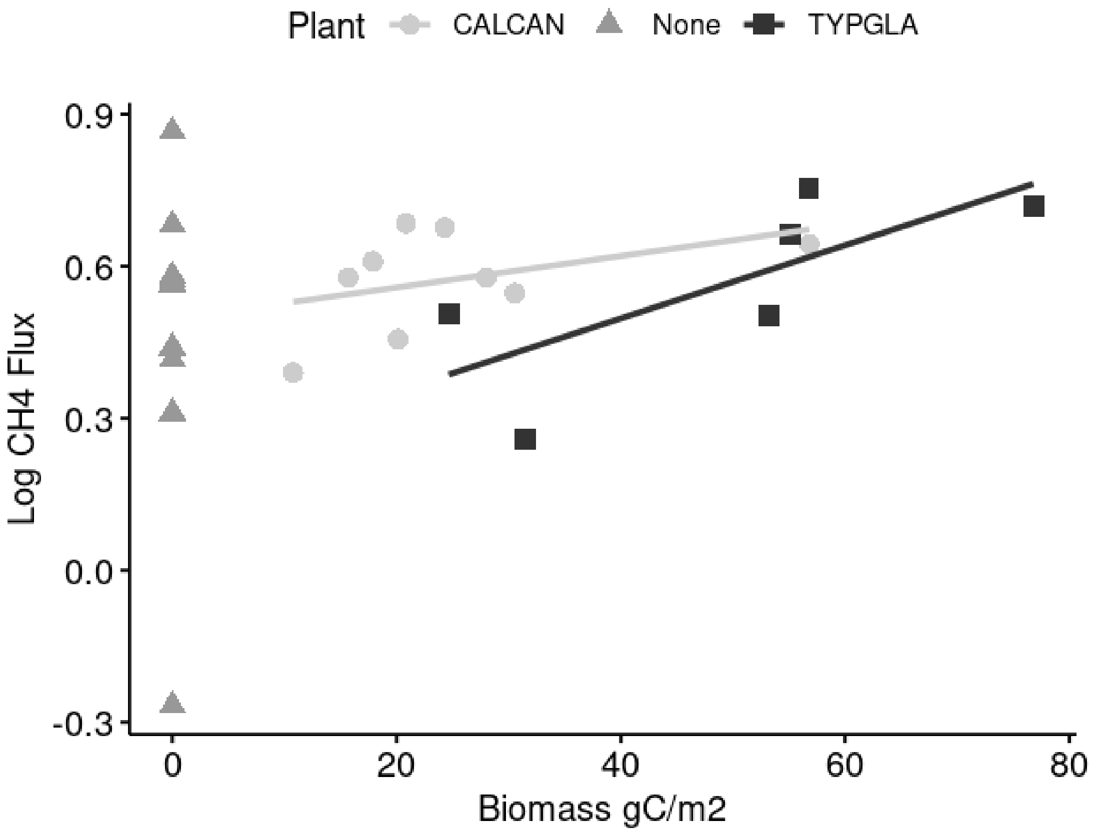

Figure 2.3. Methane flux values by aboveground biomass. Scatterplot relationship between methane flux values and aboveground biomass carbon content at each mesocosm in g/m^2.

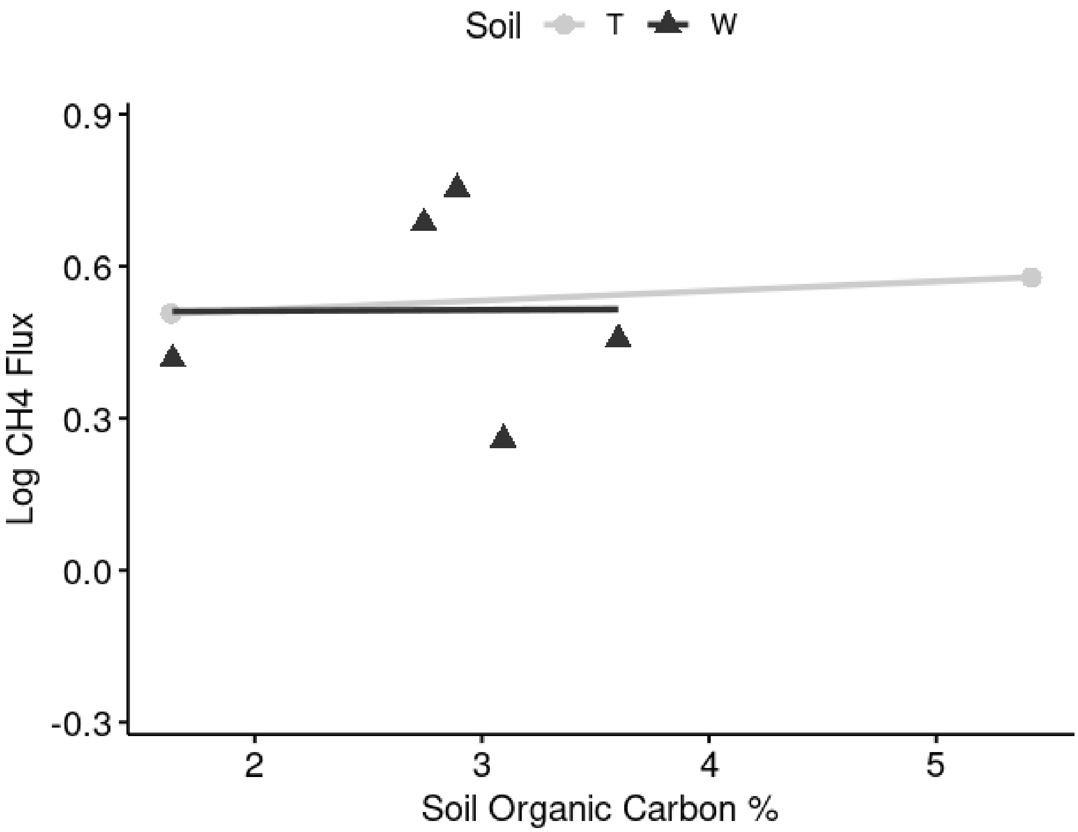

Figure 2.4*. Methane flux values by soil organic carbon percent. Scatterplot relationship between methane flux values and SOC at each mesocosm in percent. *Limited samples due to COVID-19.

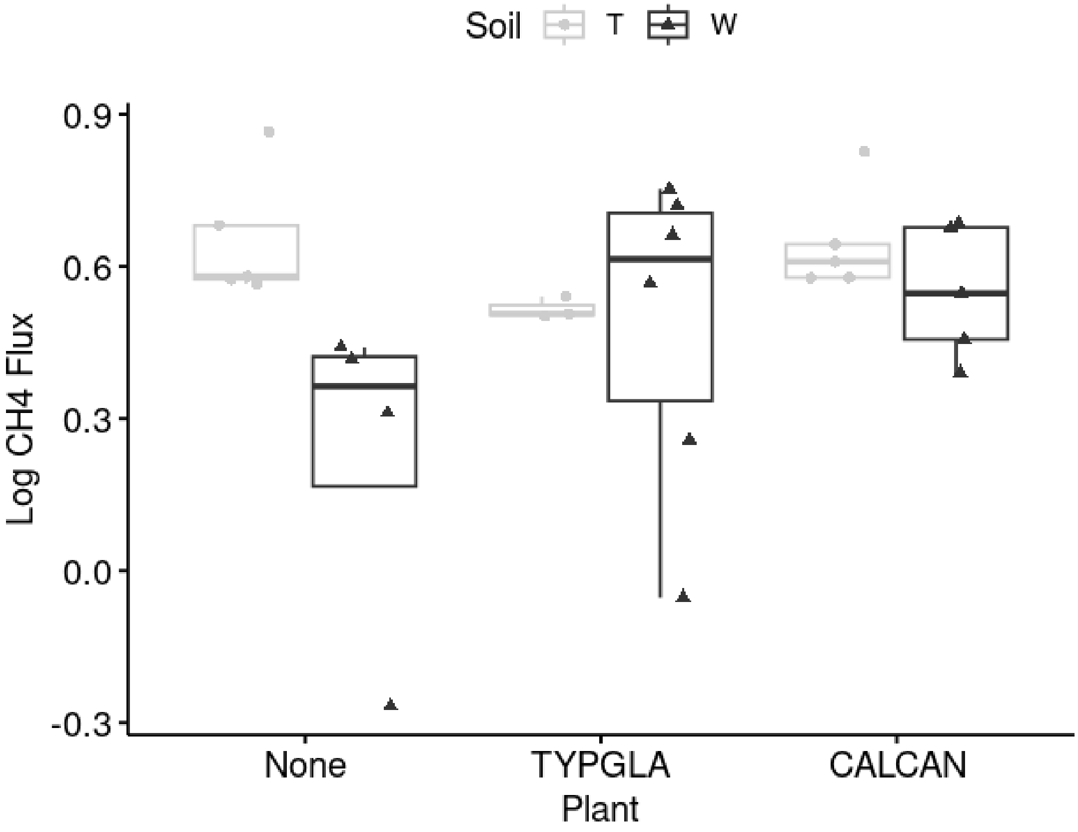

Figure 2.5. Methane flux values by plant community (Canada bluejoint, none, and cattail) and soil type (cattail soil (T) and topsoil removed (W)). Box plots represent median (bold line, box parameter is the upper and the lower quartiles and the lines represent the range in values up to two standard deviations from the mean, and dots are values beyond two standard deviations from the mean.

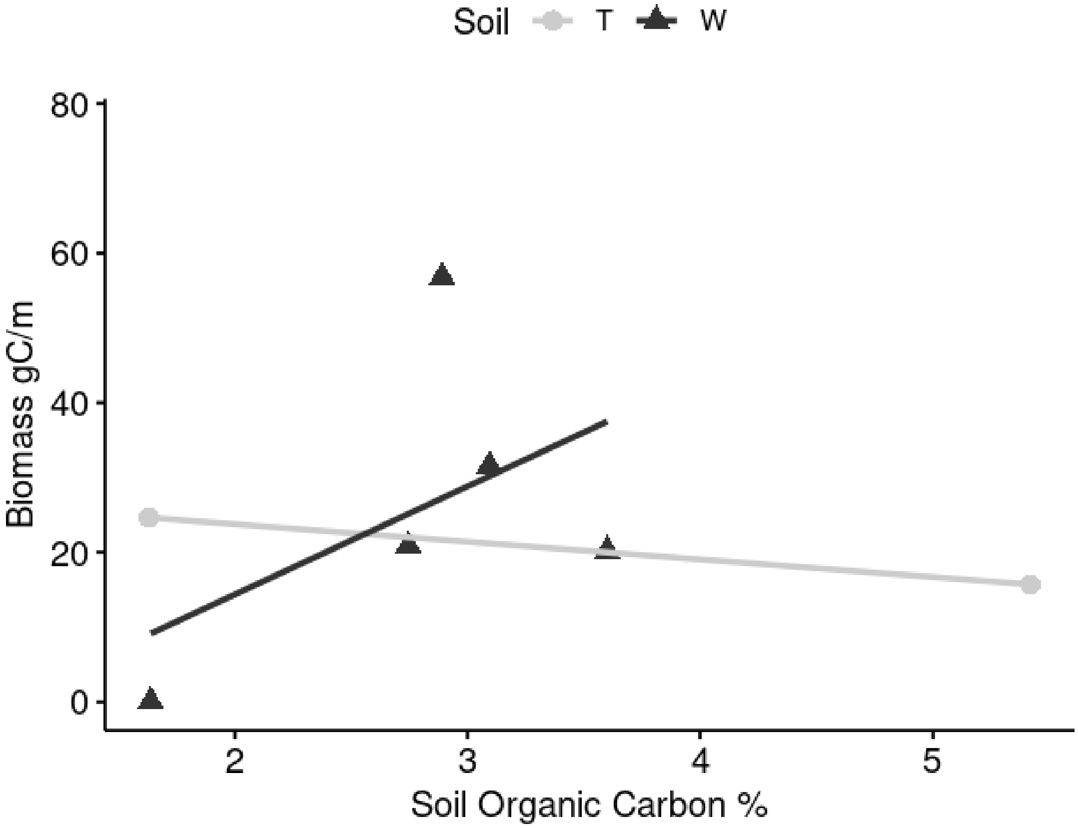

Figure 2.6*. Aboveground biomass values by soil organic carbon percentage. Scatterplot relationship between aboveground biomass carbon content in g/m^2 and soil organic carbon in percent.

*Limited samples due to COVID-19.

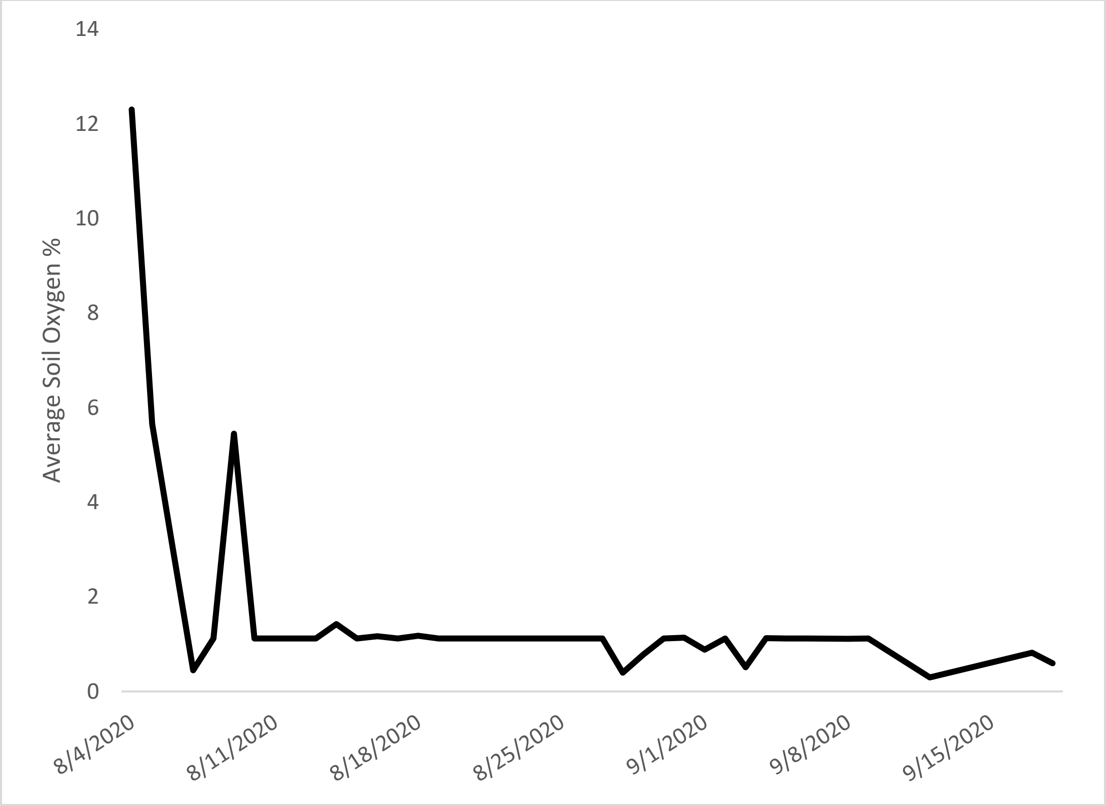

Figure 2.7. Average soil oxygen percentage in the mesocosms in August and September.

Appendix

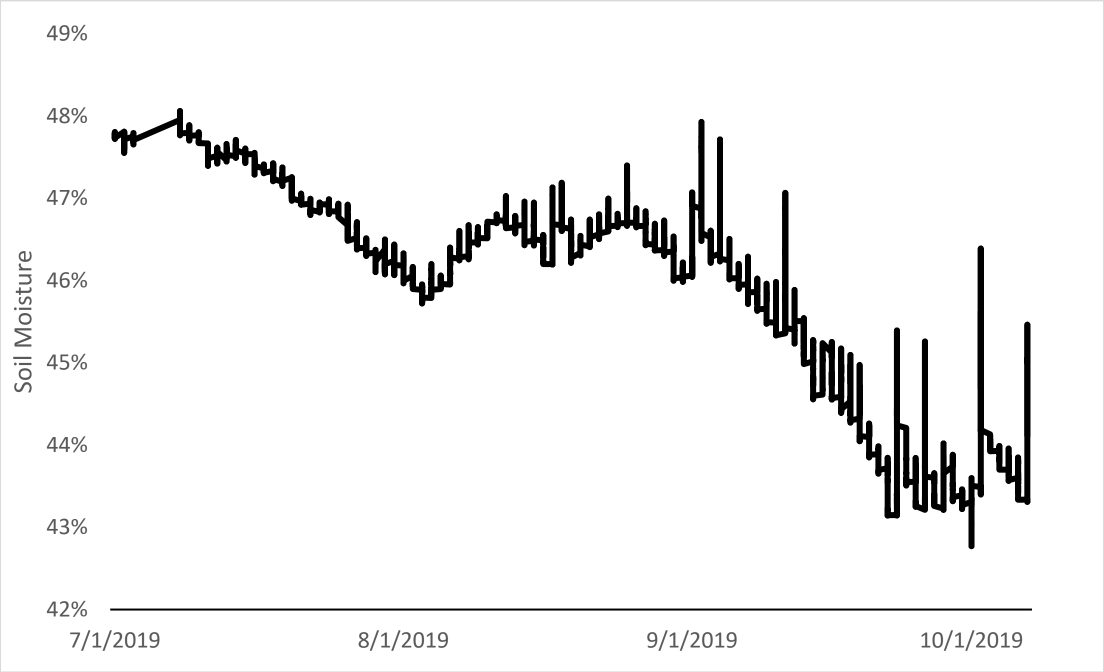

Figure A1. Salmon Creek average soil moisture in 2019.

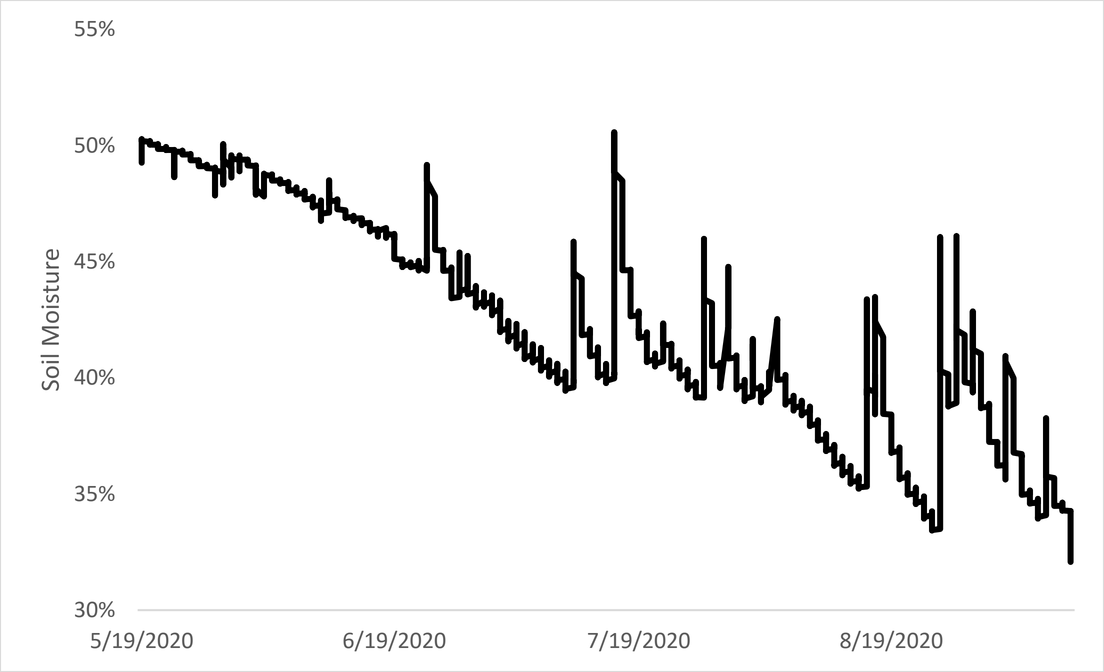

Figure A2. Salmon Creek average soil moisture in 2020.

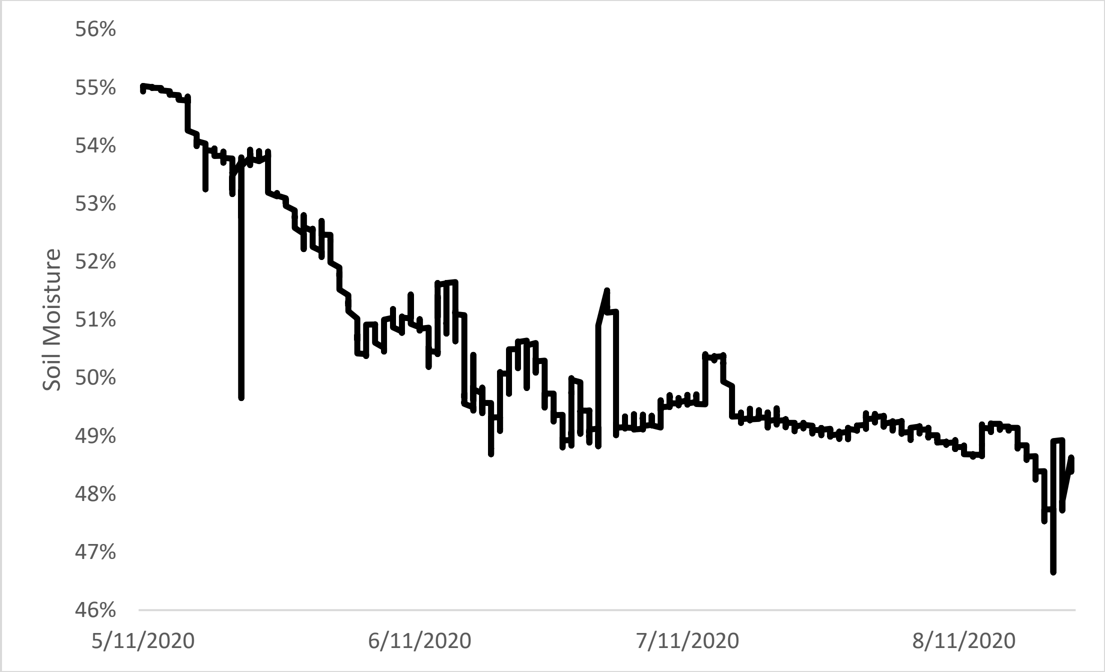

Figure A3. Buck Pond average soil moisture in 2020.

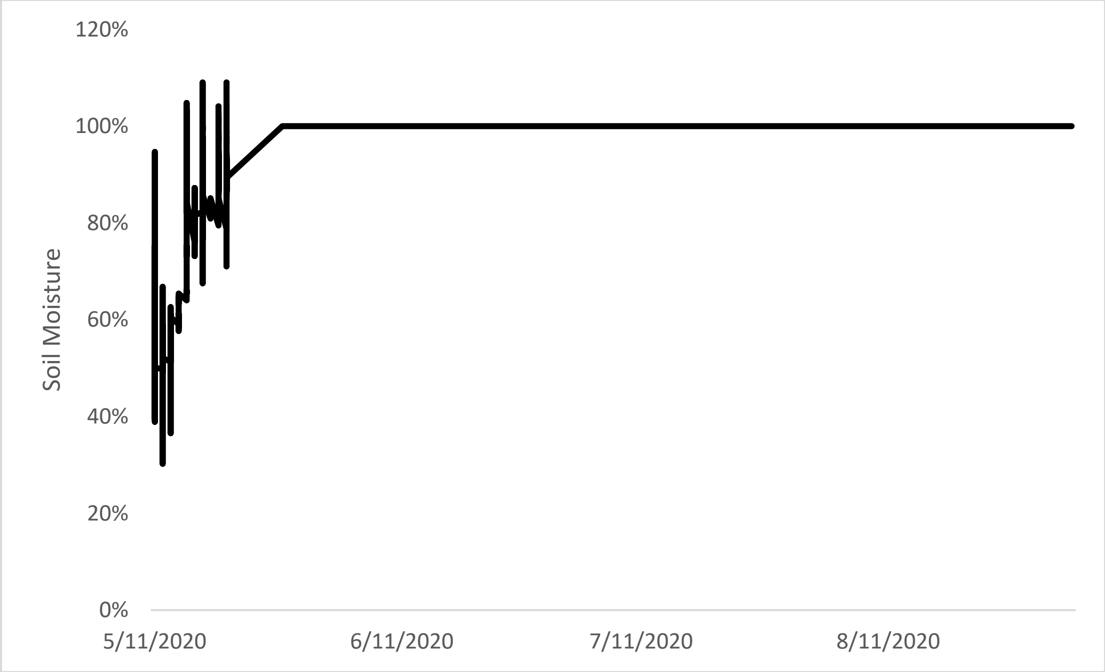

Figure A4. Braddock Bay average soil moisture in 2020.

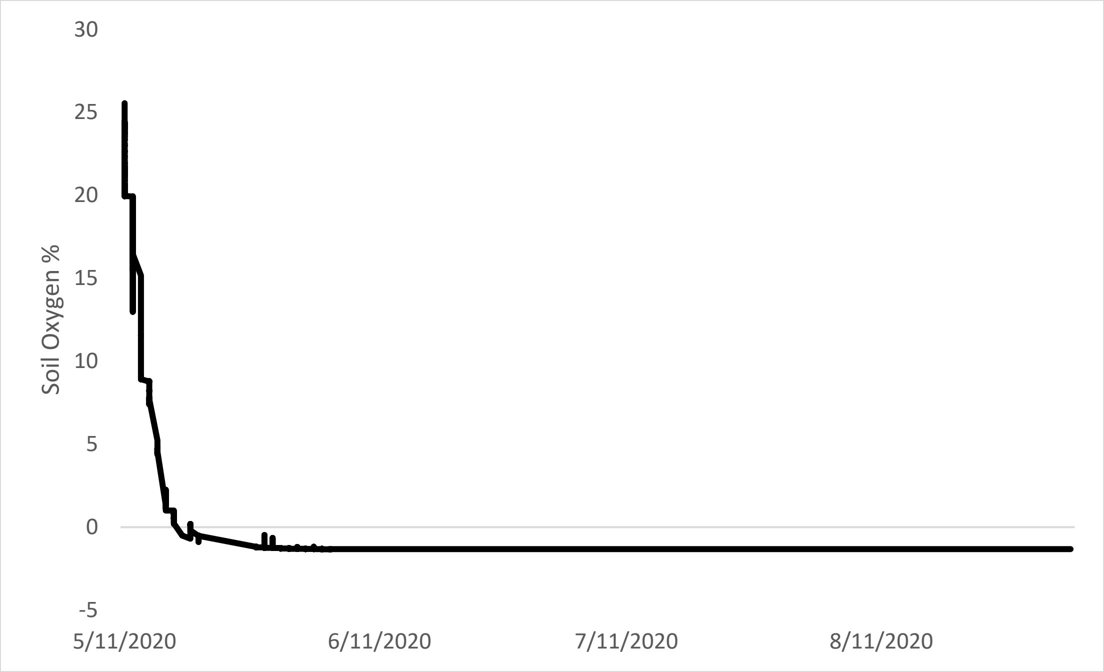

Figure A5. Braddock Bay average soil oxygen in 2020.