Mechanisms Influencing Carbon Burial in Prairie Pothole Shallow Lakes

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Dedication

This dissertation is dedicated to my late grandfather, William Beckett. Grandpa knew how to live life to the fullest, and throughout my life he instilled upon me how important it is to cherish time with friends and family. Grandpa also taught me that life isn't always easy, but if I work hard enough for what I want, I will succeed.

Abstract

Freshwater ecosystems are dominated by small, shallow lakes, and these systems have among the highest rates of carbon burial in the world. Understanding the mechanisms that influence the way shallow lakes process and accumulate carbon is important given the rising effort to mitigate anthropogenic carbon emissions. In aquatic ecosystems, the amount of carbon available for permanent burial is affected by the balance between primary production and respiration (i.e. net ecosystem production), the amount of carbon exported from the system as a gas or through groundwater fluxes, and the quality of the organic matter deposited in the sediments and the environment in which it is deposited. The Prairie Pothole Region (PPR) of North America is a noteworthy region to evaluate carbon cycling in small freshwater ecosystems because it contains approximately 207,000 km^2 of prairie lakes. Lakes in this region also typically exist in two alternative regimes: a clear water regime dominated by submerged macrophytes and/or macroalgae, and a turbid water regime dominated by phytoplankton. Based on the physiological and ecological differences of the dominating primary producers of these regimes, it is likely that one regime may accumulate carbon at a faster rate than the other.

In order to understand the mechanisms that influence carbon burial, I sampled nine shallow lakes located within the PPR over the course of three years to determine whether lake regime influenced net ecosystem production rates and exchange of carbon dioxide between the lakes and the atmosphere. I also determined the decomposition rate of macrophyte and algal material under aerobic and anaerobic conditions. Over the course of three growing seasons, the lake regime (clear or turbid) did not predict whether

the lakes had a positive or negative net ecosystem production rate (autotrophic or heterotrophic, respectively), or whether the lakes were a carbon sink or source to the atmosphere. Because the variability in metabolism and CO_2 exchange with the atmosphere was not strongly influenced by the biological differences between the regimes, the metabolism and CO_2 exchange was more likely influenced by complex interactions driven by climate (i.e. temperature, wind turbulence, watershed input) that I was unable to distinguish. Annual production rates of these lakes were as high as 1345 g C m^{-2} yr^{-1}, and over the course of three years, net ecosystem production was essentially neutral, as gross primary production (GPP) rates were approximately equal to ecosystem respiration (R). This balance implies that more carbon was retained in these shallow lakes in comparison to many other lake ecosystems where R exceeds GPP via the mineralization of terrestrial carbon. Although these lakes were metabolically neutral, the balance between gross primary production and respiration did not strongly influence the exchange of CO_2 with the atmosphere, because of the hard-water nature of these particular lakes. On average these lakes were a net carbon source, emitting approximately 114 mg C m^{-2} d^{-1}, and changes in pH strongly influenced the exchange of CO_2 between the lake and the atmosphere. Due to greater carbonate precipitation in the clear regimes, the lakes in the turbid regime had a larger buffering capacity and therefore a diminished metabolic influence on pH and therefore CO_2 exchange. Finally, the most carbon retained during the one-year decomposition experiment were the primary producers of the clear-water regime (*Charophyte*, *Potamogeton pectinatus*, *Myriophyllum sibiricum*) under anaerobic conditions (26% carbon retention). The phytoplankton of the turbid regime

only retained 1% of its original carbon content under aerobic conditions. When these decay rates were applied to GPP estimates of each regime, it was estimated that almost five times as much carbon should remain in the clear water lakes in comparison to the lakes of the turbid regime. Consequently, the clear-water, macrophyte/macroalgae dominated lakes should accumulate carbon at a faster rate than the lakes in the turbid water regime. Accordingly, shallow lakes may be managed for the clear water state not only to improve habitat, but also to sequester carbon.

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Preface

Historically, scientists assumed that inland waters had a negligible effect on global carbon cycling due to their relatively small surface area on a global scale. However, recent studies have shown that freshwater ecosystems play an active part in global carbon cycling (Cole et al. 2007; Tranvik et al. 2009). Most inland aquatic ecosystems are small, and are among the most productive ecosystems in the world (Schlesinger 1997, Dean and Gorham 1998, Downing et al. 2006). Small, shallow lakes are located in many regions throughout the world, and they tend to exist in two alternative regimes: a clear-water, macrophyte or macroalga dominated regime, or a turbid-water, phytoplankton dominated regime (Scheffer et al. 1993). Although some work on deciphering the details of carbon cycling in shallow lakes has been done (del Giorgio and Peters 1994, Finlay et al. 2009), few studies have taken a comprehensive approach in evaluating the major mechanisms influencing carbon cycling in shallow lakes in light of the alternative regimes. I evaluated the major mechanisms that affect carbon cycling and accumulation in nine shallow lakes of both the clear and turbid water regimes, located in the Prairie Pothole Region (PPR) of North America. The PPR is a notable location to evaluate these relationships, as it contains approximately 207,000 km^2 of shallow lakes that exist in the clear and turbid water regimes.

Because lakes often receive and process allochthonous carbon, most lakes worldwide are net heterotrophic, meaning they respire more carbon than is fixed within the lake via photosynthesis (Cole et al. 2000, Duarte and Prairie 2005, Sobek et al. 2005). Measurements of planktonic characteristics such as chlorophyll (i.e. planktonic primary

producers) and dissolved organic carbon (substrate for microbial mineralization) have been used to predict whether a lake is net heterotrophic or autotrophic, and a carbon source or sink. However, these planktonic predictors may not be appropriate for both types of shallow lake regimes, given the potential influence of the benthic community on whole-lake ecosystem metabolism (Andersson and Brunberg 2006, Lauster et al. 2006, Vadeboncouer et al. 2006).

Akin to the fact that most lakes are heterotrophic, the majority of lakes are also supersaturated in CO_2 relative to the atmosphere, and therefore function as a carbon source (Prairie et al. 2002, Hanson et al. 2004, Sobek et al. 2005, Finlay et al. 2009). Whether it was a direct measurement or an assumption, scientists have reported that the balance between gross primary production and ecosystem respiration automatically indicates whether a lake is a carbon source or sink (Duarte and Agustí 1998, Cole et al. 2000, Duarte and Prairie 2005, Pace and Prairie 2007). In other words, if respiration exceeded gross primary production, CO_2 should have fluxed out of the lake into the atmosphere. While this connection may be true for many lake ecosystems, the actual flux of CO_2 is not only dependent upon the metabolism of the lake, but it also depends on the chemical (i.e. carbonate buffering, hardness) and physical (i.e. wind turbulence, temperature) properties of the lake (Finlay et al. 2009). Similar to lake metabolism, predictors such as dissolved organic carbon have been commonly used to predict the concentration and therefore flux of CO_2 (Hanson et al. 2004, Sobek et al. 2005, Roehm et al. 2009); however, shallow lakes with a strong benthic influence may not function in a similar way to lakes with a much larger pelagic region (van de Bogert et al. 2007). In the

perspective of shallow lakes in the clear and turbid water regimes, no data has been reported on the driving influences on the flux of carbon dioxide between the lake and the atmosphere in the hard-water, shallow lakes of the PPR.

Finally, the clear and turbid water regimes likely accumulate carbon at dissimilar rates based on the distinctive physiological and ecological characteristics of the dominating primary producers. In terms of carbon accumulation of organic matter, the amount of carbon stored is a function of the quality of the deposited organic matter, and the environment in which it is deposited (Rejmánková and Houdková 2006). Studies that have contrasted the decomposition rate of macrophyte and algal species have shown that macrophyte species tend to retain more carbon than phytoplankton due to a greater structural complexity; however, these studies consisted of marine and not freshwater species (Enríquez et al. 1993, Duarte and Cebrián 1996). Furthermore, the clear and turbid regimes likely have differing seasonal changes in oxygen content, which may impact the rate at which their organic matter decomposes. In the fall after the senescence of the macrophytes, clear-water lakes can experience a sharp decline in oxygen which is fueled by the microbial mineralization of the newly deposited organic matter (Goldshalk and Wetzel 1978, Golterman 1995). No studies to date have investigated the decomposition of the dominating primary producers of the clear and turbid regimes under differing oxygen conditions that reflect the environment of each regime.

Chapter 1: Net Ecosystem Production in Prairie Pothole Shallow Lakes under Alternative Trophic Regimes

Shallow lakes play an important role in global carbon cycling, as they store carbon at a high rate due to their productive nature and the organic matter they receive from terrestrial ecosystems. The degree of carbon burial greatly depends on the amount of carbon available for storage within the ecosystem, which equals the balance between gross primary production and respiration (i.e. net production). The Prairie Pothole Region (PPR) of North America contains approximately 207,000 km^2 of shallow lake area, of which the metabolism and carbon burial rates are largely unknown. Lakes in this region typically exist in two regimes or states: a clear, macrophyte-dominated regime and a turbid, phytoplankton-dominated regime. These two regimes have the potential to influence the metabolic rates and carbon burial rates in different ways because of their dissimilarities in primary producer composition and decomposition. I investigated the metabolism of nine shallow lakes located in the PPR over the course of three years to estimate the metabolic balance of these shallow lakes, and to establish whether previously determined thresholds of chlorophyll-*a* (Chl) and dissolved organic carbon (DOC) were useful in predicting the metabolic rate of PPR shallow lakes in both the clear and turbid regime. Gross primary production and respiration of these shallow lakes were influenced by benthic production and respiration irrespective of state, and therefore the planktonic Chl threshold was not useful in predicting the open-water season planktonic or whole ecosystem metabolism of all nine lakes. Similarly, DOC was not a useful predictor

of the planktonic or ecosystem metabolic rate, perhaps because the short-term microbial respiration rates mostly depended upon autochthonous DOC rather than the total pool size. In contrast to many freshwater ecosystems where respiration exceeds gross primary production, gross primary production was approximately equal to respiration for all nine lakes during the study period. Surprisingly, the balance between primary production and respiration (net aquatic production) did not correlate with lake regime because the balance was similar between the two regimes during each of the three years. Because net ecosystem production was similar between the regimes, other mechanisms such as climate may have influenced lake metabolism more strongly. Although the two alternative regimes had fixed approximately the same amount of carbon, that the fate of carbon during the winter months is poorly unknown. If respiration would exceed production in the winter, a smaller amount of carbon would be available for storage. Further investigation is needed to determine metabolic rates on an annual basis in order to tell whether lakes in the clear or turbid regime bury the same or a differing amount of carbon annually.

Introduction

Net ecosystem production (NEP) of aquatic ecosystems refers to the metabolic balance between gross primary production and respiration, and the sign of NEP determines whether a lake is identified as net heterotrophic (negative NEP) or net autotrophic (positive NEP) (Odum 1956, Woodwell & Whittaker 1968, Chapin et al. 2006, Lovett et al. 2006). In net autotrophic lakes, gross primary production (GPP)

exceeds respiration (R) which indicates an increased potential for organic carbon accumulation over time. High benthic production, high nutrient availability, and a planktivore (minnow) dominated food web all increase the probability of a lake existing in a net autotrophic state (Cole et al. 2000, Andersson & Brunberg 2006, Lauster et al. 2006). In contrast to autotrophic lakes, R exceeds GPP in net heterotrophic lakes, which is possible when allochthonous nutrient sources such as terrestrial surface runoff, groundwater flow, leaf deposition or even waterfowl defecation provide additional organic carbon substrates that facilitate respiration (Chapin et al. 2006, Lovett et al. 2006, Voros et al. 2008). Contrary to the historical perspective on metabolism in freshwater lakes, the current literature suggests that a large proportion of lakes worldwide are actually net heterotrophic because of their metabolic dependence on organic matter originating from outside the lake ecosystem (del Giorgio and Peters 1993, Cole et al. 2000, Cole and Caraco 2001, Prairie et al. 2002, Hanson et al. 2003, Duarte and Prairie 2005).

Predicting metabolism of small, freshwater ecosystems such as shallow lakes is important because most freshwater ecosystems are small, and yet play a disproportionately large role in global carbon cycling (Downing et al. 2006, Cole et al. 2007, Tranvik et al. 2009). In many lakes, metabolism can be predicted by the biomass of primary producers and the concentration of dissolved organic carbon (DOC) in the water column. In their review of published planktonic production and respiration rates in 118 stratified freshwater lakes, del Giorgio and Peters (1993) observed that lakes were net heterotrophic (GPP < R) when they had Chl levels less than 17-20 μg L^{-1}, and were net

autotrophic (GPP > R) when Chl was greater than 17-20 μg L^{-1}. Their results suggested that when the level of planktonic primary production increased, the support of lake metabolism by exogenous organic matter decreased relative to the support by autochthonous nutrients. Therefore, aquatic systems with high planktonic primary productivity and/or high Chl were more likely to be net autotrophic than those with lower levels (Odum and Prentki 1978, del Giorgio and Peters 1993). Similarly, a threshold of 4-10 mg of DOC L^{-1} has also been identified as a threshold determining net heterotrophy or autotrophy in north temperate lakes (> and < 4-10 mg DOC L^{-1}, respectively) (Prairie et al. 2002, Hanson et al. 2003). Because DOC functions as a substrate for microbial mineralization, it often positively correlates with high bacterial production and R. In contrast, colored DOC can have a negative effect on primary production via shading (Webster et al. 2008), which also can cause R to exceed low rates of GPP (del Giorgio and Peters 1994). At high water column DOC, R often exceeds planktonic primary production (Hessen et al. 2004, del Giorgio et al. 1997).

Although the Chl and DOC thresholds have been used to accurately predict the metabolic status of hundreds of lakes around the world, they were specifically developed for predicting planktonic metabolism (del Giorgio and Peters 1993, Prairie et al. 2002). Consequently, they may not accurately predict the metabolism of ecosystems where the benthic and littoral contribution to the whole ecosystem is important (Andersson and Brunberg 2006, Vadeboncouer et al. 2006), such as shallow lakes. In this study I tested whether the Chl and DOC thresholds can be used to accurately predict the ecosystem metabolism of shallow prairie lakes.

The Prairie Pothole Region (PPR) of North America serves as an ideal location to study shallow lake metabolism because it contains approximately 207,000 km^2 of highly productive shallow lakes (Euliss et al. 2006, Kenning 2010). Because these lakes are shallow, a large portion of their volume is associated with the surrounding landscape which increases the potential for allochthonous DOC to be transported from the land to the lake, where it may become respired via microbial mineralization. Furthermore, the lakes in this region differ in the type of dominant primary producer, which could also directly influence ecosystem metabolism. Lakes in the PPR typically exist in two alternative regimes: a clear-water, macrophyte- and/or macroalgae-dominated regime, or a turbid-water, phytoplankton- and/or microalgae-dominated regime (Scheffer et al. 1993, Zimmer et al. 2001). Either regime can have greater primary production and respiration rates, depending on differences in growing season length, response to nutrient pulses, access to nutrients in the sediment and light (Goulder 1969, Rørslett et al. 1986, Mitchell 1989, del Giorgio et al. 1997, López-Archilla et al. 2004). To date, five lakes of the PPR have been shown to be generally autotrophic, (Kenning 2010).

I evaluated the metabolism of nine shallow lakes within the PPR over three years to determine: 1) their annual metabolic rate and whether they were net heterotrophic or autotrophic, 2) if the Chl and DOC thresholds can accurately predict the whole ecosystem and planktonic metabolic balance, and 3) whether the lake regime (clear or turbid) influences the metabolic balance of these shallow lakes.

Methods

General Description

This study was conducted in the southeastern portion of the North American Prairie Pothole Region. Nine shallow lakes in west-central Minnesota were monitored during the open water season from May 2006 through November 2008. All lakes were located within Waterfowl Production Areas, averaged 0.15 km^2 (except one lake: Lake Christina 20 km^2), and had a mean maximum depth of 2.1 m. Lakes with a mean total Chl concentration less than 22 or greater than 31 $\mu g\ L^{-1}$ were classified in the clear and turbid regime, respectively (Zimmer et al. 2009). Over the course of the study, five lakes remained in the clear regime and four lakes were in the turbid regime (Table 1).

Lake Sampling

Water samples from each lake were taken every 2-3 weeks from May through August, and once during October and November. The water samples from each lake were collected from 10 cm below the lake surface, and placed on ice until filtration, which was completed a few hours after collection. The water samples were analyzed for Chl and DOC, and were also used in laboratory incubations to estimate planktonic metabolism (see description below). Measurements of Chl were obtained by filtering water samples through a Whatman GF/F glass fiber filter (nominal pore size 0.7 μm). After freezing, Chl was extracted in acetone for 24 h and was quantified using a Turner Designs TD-700 fluorometer (Welschmeyer 1994). To estimate DOC, water samples were filtered through an ashed (450°C for 4 h) GF/F filter, and the filtrate was immediately preserved with HCl

to lower the pH below 2. A Shimadzu TOC-VCSH was used to estimate the concentration of DOC in the water samples. In order to determine the aromaticity of DOC by estimating the specific ultraviolet absorbance (SUVA), I divided the concentration of DOC (mg L^{-1}) by the absorbance value at the 254 nm wavelength measured with a Cary 50 spectrophotometer, and multiplied the quotient by 100 (to reported m^{-1}) (Clesceri et al. 1995, Weishaar et al. 2003). SUVA values were calculated for each lake from the June and July samplings in 2008. Additionally, the likely origin of DOC (allochthonous or autochthonous) was estimated using a fluorescence index (FI). FI was determined by dividing the fluorescence intensity at a 470 nm emission wavelength obtained from a fluorometer by the intensity at 540 nm (wavelengths adjusted from McKnight et al. 2001).

Ecosystem Metabolism

Continuous changes in dissolved oxygen reflect the metabolism (NEP), or the balance between GPP and R in the ecosystem (Odum 1956, Cole et al. 2000, Hanson et al. 2003, van de Bogert et al. 2007, Coloso et al. 2008). In order to determine NEP for the aquatic portion of the wetland ecosystem, I used a multi-probe sonde (Hydrolab DS5X or a Hydrolab Minisonde 4a) deployed in the mixing layer in the center of the lake to record temperature and dissolved oxygen every 15 minutes. The number of lakes monitored varied each year. Over the sampling period from 2006 to 2008, one turbid and one clear lake (Mavis East and Mavis West) were continuously monitored using a Hydrolab DS5X. These lakes served as the best contrast between a clear and turbid regime as both have

similar morphometry and average depth, and they are located less than 100 meters away from each other, which minimizes any differences in localized weather. These two lakes will be referred to as the "intensively monitored" lakes throughout the remainder of this manuscript. During 2007 all nine lakes were monitored; besides the intensively monitored lakes, three additional sondes (two Hydrolab DS5X and one Hydrolab Minisonde 4a) were rotated between the other 7 lakes, and deployed at 2-week intervals. In 2008 a total of four lakes were monitored; besides the intensively monitored lakes, one additional sonde (Hydrolab Minisonde 4a) was alternately deployed for 2-week intervals between two other lakes (one clear: Pisa and one turbid: Bore). Data were collected from all sondes every 2-3 weeks, and during the collection times the sondes were recalibrated in the field following Hydrolab recommendations, using oxygen-saturated water and the altitude-corrected barometric pressure at each site.

Wind turbulence influences the boundary layer depth over the lake, and consequently the oxygen flux between the lake and the atmosphere, which has implications for the dissolved oxygen concentration within the lake and the calculated NEP (Stigebrandt 1991). Therefore, wind speed was measured at each lake at the same time increments as the sonde measurements. At the two intensively monitored lakes, I used an Onset HOBO Wind Speed and Direction Sensor with a HOBO Micro Station Data Logger to measure wind speed 1.5 m above lake level on the shoreline. I used a Brunton ADC Summit (which was bound to a wind vane, and secured on top of an aluminum extension pole) to monitor wind speed at the other seven lakes approximately 1 m directly above the center of the lake. All anemometers were calibrated against each

other to minimize differences. When the wind data were unavailable at a specific lake due to equipment malfunctions, wind speed measurements from the Fergus Falls Municipal airport (40 km away, on average) were used in the analyses to calculate NEP. In order to account for any differences between the two types of wind speed readings (lake-specific or Fergus Falls airport), the wind speed measurements from the airport were compared to those taken at the exact time at an intensively monitored lake. After comparing 457 simultaneous readings, it was determined that the airport readings were 1.7 ± 1.5 (SD) times higher than the wind speed readings from the lake, and were therefore corrected for this overestimate. I compared metabolic rates determined using wind speed data from the Fergus Falls airport data and the lake-specific wind data (Morrison Lake, 10 d between 2 Jun 07 and 1 Jul 07). The metabolic estimates using the local airport wind data underestimated GPP and NEP rates by a mean of 0.8% and 3.6%, respectively, and overestimated R by 7.0%. I applied these adjustments to the days where the local airport wind data were used for all lakes, and found that it did not change the overall direction of the metabolism (net heterotrophic or autotrophic) for any lake.

NEP was calculated following the equations for site-specific metabolism described in van de Bogert et al. (2007) and Coloso et al. (2008). Briefly, the diel changes in dissolved oxygen reflect the balance between GPP and R, and these changes in oxygen recorded from the sondes were used in correspondence with wind speed and lake mixing depth to predict daily GPP and R. Mixing depth was determined from the lake depth that had the maximum temperature change throughout the water column (Hutchinson 1957). Temperature profiles were only taken in 2008, so the mixing depths for each lake were

assumed to be similar 2006 and 2007. On most dates in most lakes the mixing depth was the entire water column of the lake. Because the oxygen concentration in the lake is a function of not only biological, but physical influences, the exchange of oxygen between the lake and the atmosphere was also calculated based on wind speed and water temperature. I assumed the same exponent for the wind profile power relationship of 0.143 used in van de Bogert et al. (2007). The gas transfer coefficient was predicted using the gas piston velocity k_{600}, which is essentially the water depth that is in equilibrium with the atmosphere for a gas over a given time period at a given temperature (Cole and Caraco 1998). The Schmidt number was also used in calculating the gas transfer coefficient, and was dependent upon water temperature (Wanninkhof 1992). Nighttime R was calculated as the oxygen consumption one hour after sunset and one hour before sunrise, and nighttime R was assumed to be equivalent to daytime R (this assumption does not affect overall NEP estimates, Cole et al. 2000). Since GPP represents oxygen production and R represents oxygen consumption, GPP and R are reported as positive and negative values, respectively. After accounting for the physical exchange of oxygen between the lake and the atmosphere, the change in oxygen content in the lake from sunrise to sunset was qualified as NEP, or the sum of GPP and R. GPP was estimated indirectly by subtracting the negative R from NEP (NEP = GPP + R, or GPP = NEP – R).

It was assumed that dissolved oxygen input from groundwater was negligible, and therefore was not included in the calculations. Assuming a high groundwater input estimate of 57 $\mu M\ O_2\ s^{-1}$ (Rosenberry et al. 2000), a similar concentration of oxygen in the groundwater compared to the lake (0.2 mM, Kohfahl et al. 2009, our lakes 0.25 to

0.32 mM O_2), and an average lake volume of 225,000 m^3, the input from the groundwater on a daily basis would only comprise of 1.7% of the total oxygen content of the lake.

Planktonic Metabolism

Because the Chl and DOC thresholds were developed for planktonic communities, light and dark bottle incubations were completed to estimate planktonic GPP, R, and net production in order to test the predictability of the thresholds on both the planktonic community and the whole ecosystem metabolism. On each sampling date, water samples from the center of each lake were collected and stored in 0.5 L amber Nalgene HDPE bottles at lake temperature void of headspace. Because the samples were analyzed for planktonic production in a laboratory setting, it took approximately two days to get the samples back to the lab and prepared for the incubation. In the laboratory, the water samples were used to fill 15 ashed 7 mL clear glass scintillation vials for the bottle incubations. Three of the 15 vials we used to determine the initial concentration of oxygen, and were immediately fixed with 60 µl of 1% mercuric chloride to halt any biological activity and preserve the initial concentration of dissolved oxygen. Next, triplicate vials were incubated at low, medium, and high light levels (approximately 10, 100, and 220 µE m^{-2} s^{-1}, respectively) to estimate planktonic production, and a set of triplicate vials were incubated in the dark to measure planktonic respiration. All vials (initial, light, and dark) were incubated for approximately 1 d at the temperature of the lake at the time the water samples were taken, and following the end of the incubations the oxygen content of each vial was estimated using a membrane inlet mass spectrometer

(MIMS) (Cory et al. 2009). This procedure allowed for highly accurate measurements of dissolved oxygen in water (Kana et al. 1994). For each light level, hourly production and respiration rates were calculated by dividing the difference between the initial and final concentration of oxygen content by the incubation period. The planktonic GPP was estimated by subtracting the negative respiration rates from the net production rates.

In order to estimate the planktonic GPP within the lake, the production estimates from the vial incubations were extrapolated to the entire lake using the light profiles and production-irradiance (PI) curves. First the production rates from the vial incubations were plotted against their specific incubated light levels to yield a logarithmic equation specific to a given lake on a given day. PAR (photosynthetically active radiation) was measured at 0.5 m increments in each lake on the same date on which the water samples were collected for the bottle incubations. Using the light extinction coefficient and logarithmic production curves, primary production was calculated at each 0.5 m increment. Primary production was estimated (mmol O_2 m^{-3} hr^{-1}) as the average between the top and bottom of each 0.5 m layer, and was then multiplied by the layer depth to yield the rate of mmol O_2 m^{-2} hr^{-1}. All production rates for each 0.5 m layer were summed together for the entire lake, and then multiplied by daylength (hr) to convert to a daily rate (mmol O_2 m^{-2} d^{-1}). I assumed respiration was the same at all depths, and the changes in oxygen in the dark bottle incubations were multiplied by the mixing depth and 24 hrs to yield a daily respiration consumption rate (mmol O_2 m^{-2} d^{-1}).

Statistical Analysis

When estimating the metabolic rates in all lakes using the sonde data, occasionally there were days when considerable noise in the oxygen and temperature diel patterns resulted in negative GPP and positive R. Because this noise was observed from the sondes in two separate lakes (the two intensively monitored lakes) during the same times at the same depth (Figure 1), I concluded that the noise was driven by something other than erratic changes in metabolism within the lake. The noise was most likely due to short-term microstratification patterns later in the summer that triggered slight changes in the mixing depth, which at times caused the sonde to record readings below the mixing depth. The model I used to estimate ecosystem metabolism assumes the sonde readings were taken within the lake's mixing depth; therefore, the days with considerable noise and no detectable diel oxygen and temperature patterns were assumed to violate the model's assumptions and were excluded from the analysis.

A two-way nested ANOVA was used to detect any significant differences in GPP, R, and NEP between lake regimes, and to test if there was any influence of year and time of year on the metabolic rates. Lakes were nested within lake regime (clear or turbid), and were treated as random variables. Because metabolic rates changed throughout the growing season, daily metabolic rates were blocked into to two-week intervals in order to test the difference in metabolism between the clear and turbid regimes, and to test the influence of time of year on GPP, R, and NEP. Any *p*-value equal to or below an α value of 0.05 considered as significant.

I used regression analyses to assess the relationship between annual averages Chl, GPP and NEP, and DOC, R and NEP, using whole-ecosystem and planktonic metabolic estimates. For the following discussion, gross primary production, respiration, and net ecosystem production for the entire lake (as estimated using the data collected from the sondes) are referred to as GPP_E, R_E, and NEP_E, respectively. Similarly, GPP_P, R_P, and NEP_P signify gross primary production, respiration, and net production of the plankton community (estimated from bottle incubations). I address the relationship between plankton and whole-lake metabolism versus Chl and DOC by splitting the analysis using all lakes, and the clear and turbid lakes separately.

Results

For all lakes, the open-water seasonal means (May to November) for GPP_E ranged from 143 to 800 mmol O_2 m^{-2} d^{-1}, with a grand mean of 323 mmol O_2 m^{-2} d^{-1} (Table 1). The open-water seasonal mean R_E rates ranged from -139 to -736, with an average of -324 mmol O_2 m^{-2} d^{-1}. The open-water seasonal mean of the NEP_E rates ranged from -180 to 76, with an average of -4 mmol O_2 m^{-2} d^{-1} (Table 1). The metabolic rate for some lakes depended on the season, and therefore the open-water seasonal means are somewhat biased depending on when the lakes were monitored (some monitored May through November, others only during one or two months during the summer). Therefore, I also report the minimum, maximum, and median values of all daily metabolic estimates for all the lakes. GPP_E ranged from 4 to 1315 mmol O_2 m^{-2} d^{-1}, and had a median value of 246 mmol O_2 m^{-2} d^{-1}. The minimum and maximum values for R_E were -1 and -1186 mmol O_2

m^{-2} d^{-1}, respectively, with a median value of -260 mmol O_2 m^{-2} d^{-1}. Finally, NEP_E ranged from -487 to 483 mmol O_2 m^{-2} d^{-1}, with a median value of -16 mmol O_2 m^{-2} d^{-1}.

The lakes varied in planktonic Chl content from 2 to 207 µg L^{-1}, with a mean of 11 ± 7.9 (SD) µg of Chl L^{-1} for clear lakes and 109 ± 72.7 µg of Chl L^{-1} for turbid lakes (Table 1). The annual average of Chl did not correlate significantly with annual GPP_P, GPP_E, or NEP_P when all lakes were considered together, or when the clear and turbid regimes were considered separately (all $p > 0.05$) (Figure 2a-c). In contrast, NEP_E had a significant negative correlation with Chl when all lakes were considered ($p = 0.011$), and the correlation was strongest in the turbid lakes ($p = 0.032$) (Figure 2d). The threshold of 17-20 µg Chl L^{-1} (del Giorgio and Peters 1993) did not accurately predict whether lake metabolism was autotrophic or heterotrophic for either the planktonic community or the whole ecosystem (Figure 2b,d).

The mean open-water annual values (May through November) of DOC for each lake ranged from 8.4 to 26.4 mg L^{-1}, with clear and turbid lakes having a mean of 14.0 ± 2.5 (SD) and 18.1 ± 4.5 (SD) mg DOC L^{-1}, respectively (Table 1). There was no significant correlation between DOC and R_P, R_E, or NEP_P when all lakes were considered together, or when the clear and turbid regimes were considered separately (all $p > 0.05$) (Figure 3a-c). Although DOC did not significantly correlate with NEP_E when the clear or turbid regimes were considered separately ($p = 0.514$ and 0.125, respectively), DOC did have a significant negative relationship with NEP_E when all lakes were considered ($p = 0.032$) (Figure 3d). Although only one lake had mean DOC concentration within the 4-10 mg DOC L^{-1} threshold (Prairie et al. 2002, Hanson et al. 2003) and no lakes had a mean

DOC concentration below the threshold, some lakes were net autotrophic above the threshold, and consequently the DOC threshold of 4-10 mg DOC L^{-1} was not useful in predicting the direction of metabolism for either the planktonic community or whole ecosystem (Figure 3b,d).

GPP was significantly, negatively correlated with R in both the clear and the turbid lakes (all $p < 0.05$) (note that R is presented as a negative rate, so the absolute values of these processes were positively related; Figure 4). As GPP increased from 6 to 730 mmol O_2 m^{-2} d^{-1} (clear lakes) and 4 to 1315 mmol O_2 m^{-2} d^{-1} (turbid lakes), R decreased from -5 to -730 mmol O_2 m^{-2} d^{-1} (clear lakes) and -1 to -1186 mmol O_2 m^{-2} d^{-1} (turbid lakes).

NEP_E did not significantly differ between clear and turbid lakes, while GPP_E and R_E were significantly different. When considering all lakes within each year, GPP_E was significantly greater in the turbid lakes in 2006 and 2008 ($p = 0.0328$ and $p < 0.0001$), and R_E was significantly higher in the turbid lakes than in the clear lakes during all three years (2006: $p = 0.0011$; 2007: $p = 0.006$; 2008: $p < 0.0001$) (Figure 5a,b). NEP_E was not significantly different between regimes during any year when considering either all of the lakes together, or just the two intensively monitored lakes (all $p > 0.05$) (Figure 5c,d).

GPP and R varied over time in the clear and turbid lakes, and some of this variation correlated with time of year. To show this, I present the change in the metabolic rates during 2007 for the intensively monitored lakes (Figure 6). When including the daily metabolic rates of all nine lakes, I found that GPP_E and R_E significantly correlated with season ($p = 0.0027$ and $p = 0.0047$, respectively), however NEP_E did not ($p > 0.05$).

In the turbid lakes, GPP_E and R_E had a significant correlation with the time of year (p = 0.001 and p = 0.005, respectively). GPP_E and R_E were low at the beginning of the summer (June), and increased towards the onset of fall (September), but were the lowest during October and November (however there was no correlation between daily temperature, GPP, and R). In contrast, GPP_E, R_E, and NEP_E did not correlate with time of year in the clear lakes (all $p > 0.05$), as these rates were fairly constant within a year compared to the metabolism in the turbid lakes.

Metabolism of the clear and turbid lakes fluctuated between years as well. When all lakes were considered in the analysis, GPP_E, R_E, and NEP_E significantly correlated with year ($p < 0.0001$, $p = 0.0047$, and $p = 0.0003$, respectively) (Figure 5a-c). Individually, GPP_E and R_E correlated with year in the clear lakes, ($p < 0.0001$ and p = 0.0005, respectively), while NEP did not correlate with year ($p > 0.05$). Both GPP_E and R_E were the highest in 2006, and were the lowest in 2008. In contrast to the clear lakes, GPP_E and R_E were not correlated with year in the turbid lakes (all $p > 0.05$), but NEP_E significantly correlated with year ($p = 0.0018$) (Figure 5a-c). In the intensively monitored lakes, NEP_E significantly correlated with year ($p = 0.0143$), as the values decreased from 2006 to 2008 (Figure 5d).

Discussion

Planktonic Chl and DOC were poor predictors of ecosystem metabolism because benthic processes dominate these fluxes in such shallow ecosystems. The metabolic balance in these highly productive lakes was very close to neutrality, and did not vary in

any significant way with lake regime. Finally, the interannual variability in the metabolic rates of the shallow lakes was likely due to differences in climate in different years.

The use of oxygen to estimate ecosystem metabolism

Using changes in dissolved oxygen to infer lake metabolism, i.e., carbon fixation and respiration, is a commonly used method (Cole et al. 2000, Hanson et al. 2003, van de Bogert et al. 2007, Coloso et al. 2008), but can potentially underestimate production in anaerobic environments where anoxygenic inorganic carbon fixation can occur. In these lakes, it was highly likely that anoxygenic production seldom occurred during our sampling periods because all of the lakes were hypoxic (< 2 mg O_2 L^{-1}) only 2.7% of the time (on average), usually during the night. Because these lakes were rarely hypoxic while receiving sunlight, and assuming that anoxygenic photolithotrophs and chemolithotrophs together contributed only 0.5 to 1% of the total inorganic carbon fixed by aerobic primary producers (global estimate, Raven 2009), I argue that using diel changes in oxygen to indicate carbon fixation and respiration was appropriate in these prairie lakes.

Planktonic measurements in benthic dominated systems

Estimating planktonic metabolism via bottle incubations undoubtedly imposes a bias as the open-water community is unnaturally constrained within a bottle (Fogg and Calvario-Martinez 1989). Still, this method continues to be widely used to estimate pelagic production in lake ecosystems (Goulder 1969, del Giorgio and Peters 1994,

Carignan et al. 2000, Anderson and Brunberg 2006, Blindow et al. 2006, Lauster et al. 2006). In 2006, I completed similar incubations in 60 ml bottles (versus the 7 ml bottles presented here) to estimate pelagic metabolism. The metabolic rates obtained from the 60 ml bottles were similar to rates obtained from the incubations in the 7 ml vials, all of which were only run for approximately 24 hours. Therefore, the 7 ml bottles I used to estimate pelagic metabolism should have the same bias included in many other studies (Goulder 1969, del Giorgio and Peters 1994, Carignan et al. 2000, Anderson and Brunberg 2006, Blindow et al. 2006, Lauster et al. 2006).

I did not observe a significant correlation between Chl and GPP_P or GPP_E when all lakes were considered, or when the clear and turbid lakes were analyzed separately. This result was not surprising for the lakes in the clear regime because Chl only represents the planktonic primary producers in the lakes, while submersed macrophytes and macroalgae instead dominate the clear lakes (Scheffer et al. 1993). In contrast to our expectations, however, there was no significant correlation between Chl and GPP_P or GPP_E for the turbid lakes either. The lack of correlation between the planktonic primary producer biomass and production rates in the phytoplankton-dominated lakes suggests the phytoplankton may have been competing for some requirement (such as light), and/or there may be more benthic primary production than expected even in these lakes. The concentrations of Chl observed in the turbid lakes matched those observed in similar lakes where a high concentration of Chl in the water column (indicating high concentration of phytoplankton) caused self-shading which reduced overall planktonic photosynthetic rates within the mixing layer (Blindow et al. 2006). On average, 48% and

13% of the lake depth was excluded from the euphotic zone in the turbid and clear lakes, respectively. In comparison to the lakes in the clear regime, a smaller volume of water in the turbid lakes had sufficient light to support primary production. Although GPP_E, and R_E did not correlate with Chl (p = 0.781 and 0.816, respectively), the smaller lake volume for GPP and larger lake volume for R in the turbid lakes may have been enough to affect the balance between GPP and R, thus leading to a more negative NEP_E at higher levels of Chl (Figure 2d). In summary, I did not observe a significant correlation between Chl and GPP_P and GPP_E due to the omission of benthic primary producers in the clear, macrophyte-dominated lakes and self-shading in the turbid, phytoplankton-dominated lakes. These reasons justify why the threshold of 17-20 µg Chl L^{-1} (del Giorgio and Peters 1993) was not useful in predicting whether the plankton or ecosystem metabolic rate was either net autotrophic or heterotrophic.

DOC also was not a useful predictor of metabolism and did not vary with R or NEP, most likely because the DOC measured from the surface waters was not utilized at the same time scale in which metabolism was measured. There was no significant correlation between DOC and R_P or R_E when all lakes were included in the analysis, or when the clear and turbid lakes were considered separately. This lack of relationship is common in shallow lakes where greater concentrations of colored DOC inhibited primary production through shading more than it enhanced R (via microbial mineralization) (del Giorgio and Peters 1994). Still, this was not the case in these prairie lakes because the DOC was only slightly aromatic (average SUVA = 1.9, Weishaar et al. 2003), and DOC and GPP_E were not correlated in any way (p = 0.685). Furthermore , the metabolism of

these lakes should be less dominated by terrestrial organic matter because GPP in these lakes was much higher than those observed for other shallow lakes where DOC positively correlated with R (this study GPP mean of 326 mmol O_2 m^{-2} d^{-1} versus 72 mmol O_2 m^{-2} d^{-1} of Hanson et al. 2003) (del Giorgio & Peters 1993). Given the close association with GPP and R, and the fact that heterotrophic bacteria preferentially utilize autochthonous DOC over allochthonous DOC (Kritzberg et al. 2004), it is likely that R was fueled more by autochthonous DOC, allowing the terrestrial DOC to remain the largest portion of total DOC in the surface waters. The mean fluorescence index value of these lakes (1.40) supports this, as the DOC was characterized as mostly of terrestrial origin (Donahue et al. 1998, Waiser & Robarts 2000, McKnight et al. 2001).

I found no significant correlation between DOC and NEP_P for all lakes or specifically the clear and turbid lakes, or between DOC and NEP_E for the clear and turbid lakes separately. Although I detected a significantly negative correlation between DOC and NEP_E when all lakes were considered, its significance was driven by only one data point, and therefore it is doubtful that this negative relationship has much biological meaning. Overall DOC did not correlate with R or NEP, and was not useful in predicting the planktonic or ecosystem metabolism of these prairie lakes. The lack of correlation between DOC and metabolism is most likely explained by the short-term dependence of microbial respiration on autochthonous DOC, and not the total DOC pool, which contains a significant portion of allochthonous DOC.

The metabolic balance of prairie shallow lakes

When comparing the metabolic rates between the plankton and ecosystem communities, it became clear that the benthic environment was an important player in the overall metabolism of these shallow lakes. On average, GPP_P only accounted for approximately 22% of GPP_E in the clear lakes, which supports the notion that most of the primary production was dominated by the benthic zone in the clear water lakes (Lauster et al. 2006). For all lakes, eighty percent of R occurred near or within the benthic zone (see R_P vs R_E), which explains why these ecosystems were net heterotrophic at high levels of Chl, while the plankton community was net autotrophic at the same Chl levels. Decomposition via microbes (and consequently respiration) can be constrained by surface availability, and therefore decomposition rates (and respiration) tend to be lower in open water regions where the plankton reside (Wetzel 1992).

GPP and R were significantly, negatively correlated (Figure 4). This close association between GPP and R has been observed for planktonic metabolism of numerous other freshwater ecosystems (Carignan et al. 2000, Duarte and Augustí 1998, Duarte and Prairie 2005). Although there was a negative relationship, ecosystem respiration did not decrease proportionately with gross primary production in all lakes, as some lakes were slightly heterotrophic at low rates of primary production and autotrophic at high rates of production. This trend was similar to metabolic rates observed in many lakes around the world (del Giorgio and Peters 1993, del Giorgio et al. 1997, Duarte and Augustí 1998, Duarte and Prairie 2005). The negative intercept of these figures represented the base metabolism supported by allochthonous C sources (del Giorgio and

Peters 1994). I found that some lakes did not have a negative intercept (8Mile, Morrison, Murk), and naturally had the strongest coupling between GPP and R (slope ≈ 1), which suggests the metabolism of these lakes relied solely on autochthonous production at the time when these lakes were monitored.

On average, lakes in the clear and turbid regime had a ratio of GPP:R of 1.36 and 1.20, respectively, and these ratios did not significantly differ from each other ($p = 0.455$) (ratios are the inverse of the slopes in Figure 4). Generally GPP exceeded R, and therefore lakes in both the clear and turbid regimes had the potential for carbon export and/or accumulation (i.e. burial) (Cotner & Biddanda 2002), and this potential for C burial did not differ between regimes. It is interesting to note that the capability for C burial is high when only considering the balance between production and respiration, despite the potential for additional exogenous C input that may not be modified by metabolism but rather transferred directly to the sediments to accumulate over time. Although the C fixation rates were similar between the regimes, I did not measure production and respiration on an annual basis. I have some preliminary evidence that oxygen is depleted after ice-cover on a much faster basis in the clear regimes compared to the turbid regimes; therefore the environment of these regimes likely differs on an annual basis and should directly affect the accumulation of the net carbon fixed in these lakes.

In comparison to other ecosystems, the shallow lakes of the Prairie Pothole Region have the potential to metabolize tremendous amounts of carbon. GPP estimates from these lakes exceeded similar measurements from shallow lakes in the northern

Wisconsin, the Upper Peninsula of Michigan, and the Everglades in Florida by three- to ten-fold (Hanson et al. 2003, van de Bogert et al. 2007, Coloso et al. 2008, Hagerthey et al. 2010). The magnitude of these differences in GPP was most likely due to discrepancies in primary producer biomass and/or type, and contribution of the littoral habitat to ecosystem metabolism (Lauster et al. 2006). The annual mean GPP rates ranged from 143 to 800 mmol O_2 m^{-2} d^{-1}. If these rates are converted to carbon-based rates by assuming 1 mol of O_2 produced equals 0.8 mol C fixed (given the close association between GPP and R in these prairie lakes), and conservatively multiplying the rate by the length of our sampling season (11 May to 2 Nov, or 175 days), I estimate that approximately 240-1345 g C m^{-2} yr^{-1} was fixed by the primary producers in these lakes. Among the lakes studied, the highest annual mean of carbon fixation was comparable to other production estimates for wetlands (1300 g C m^{-2} yr^{-1}, Schlesinger 1997), and even exceeds production estimates of tropical forests (800 g C m^{-2} yr^{-1}, Schlesinger 1997). If the highest carbon fixation rate is scaled up to the total area of shallow lakes occupy in the Prairie Pothole Region (207,000 km^2, Euliss et al. 2006), lakes in this region are capable of fixing 50-280 Tg C on an annual basis, which is comparable to the production rate of the entire Great Lakes Basin (GLB) (268 Tg C yr^{-1}; GLB covers 774,174 km^2, and the lakes occupy ~258,000 km^2; Karim et al. 2008). If I assume that 28% of the carbon fixed was deposited to the sediment and not respired (using the mean of the reported GPP:R ratios for the clear and turbid regimes), and that 48% of that carbon was permanently buried over time (Sobek et al. 2009), I estimate that the lakes in the PPR accumulated 7-38 Tg C yr^{-1} based on primary production rates alone.

Comparing metabolism of the clear and turbid water regimes

GPP was high in comparison to other ecosystems, and the rates generally differed between the clear and turbid regimes. GPP was significantly higher in the turbid lakes than in the clear lakes in 2006 and 2008 (Figure 5a), but not in 2007. The higher GPP rates in the turbid lakes in part were due to the longer growing season of phytoplankton in comparison to the macrophytes (Goulder 1969, Mitchell 1989), which begin to senesce towards the beginning of September. GPP in the turbid lakes was quite high at the onset of summer and remained high through the end of September, whereas the GPP in the clear lakes was the highest only from July through early September (Figure 6), which matches the growing season for most submerged macrophytes (Wetzel 2001). Previous studies suggest that phytoplankton have higher production rates than macrophytes because they can obtain nutrients more easily from the water column, and they have a faster rate of nutrient cycling (Mitchell 1989). Although the turbid lakes had more than four times as much total phosphorus in the water column than the clear lakes, only 15% and 3% (annual mean) of the total phosphorus was soluble reactive phosphorus in the clear and turbid lakes, respectively. The low concentration of bioavailable phosphorus in the turbid lakes suggests the phytoplankton were utilizing the nutrients within the water column to a greater degree than the macrophytes.

Similar to GPP, R was significantly higher in the turbid lakes during all three years (Figure 5b). The lifespan of an algal cell in comparison to a submerged aquatic plant is much shorter, and in order to sustain production rates that exceed the rates in the clear-water lakes, the phytoplankton populations must be turning over rapidly and

therefore producing a large quantity of organic matter. Furthermore, phytoplankton cells are also easier to mineralize relative to aquatic plants, because they do not contain complex compounds such as lignin and cellulose that are difficult to breakdown and mineralize (Enríquez et al. 1993). This large, continuous supply of decaying algae throughout the open water season was likely supporting the high rates of respiration.

Surprisingly, the dominant primary producer of the lakes did not influence the overall metabolism of the lakes (Figure 5c,d). Both the clear and turbid lakes were net autotrophic in 2006, and both regimes were net heterotrophic in 2007 and 2008 (Figure 5c); the same pattern occurred in the intensively monitored lakes (Figure 5d). Since these lakes are so shallow, their physical and chemical environment can change rapidly depending on climatic and weather conditions. In order to see if any differences in weather patterns might help explain differences in NEP, I analyzed local air temperature, solar radiation, wind speed, rainfall and snowfall during the winter months prior to the open water season and the summer months in 2006, 2007, and 2008. The only significant difference among years for these variables was the daily air temperature during the open water season (April to November). Specifically, temperatures were significantly warmer in 2006 and 2007 compared to the temperatures in 2008 ($p = 0.007$ and 0.08, respectively) (2006: 15.0±9.5°C, 2007: 15.6±10.4°C, 2008: 13.7±10.7°C). Additionally, the daily temperatures during the winter prior to the open water season of 2006 and 2007 were also significantly warmer than 2008 ($p = 0.001$ and < 0.0001, respectively) (2006: -4.6±10.0°C, 2007: -2.3±9.8°C, 2008: -9.4±9.3°C). Over the course of this study, cooler

temperatures may have been enough to slow production and tip the balance between GPP and R to yield a more negative NEP (R>GPP).

Because the metabolic rates of prairie lakes varied day by day and even between years, it is important to make continuous, whole-lake measurements throughout the season to properly characterize metabolism, instead of observing metabolic rates over a few days within one year (Hanson et al. 2003, Hagerthey et al. 2010). Furthermore, metabolism should be monitored year-round. For example, if respiration is lower in the macrophyte-dominated lakes during the winter, perhaps more carbon would accumulate within the lake over time. The biotic, chemical, and physical environment of these shallow lakes can fluctuate greatly in response to changes in weather. Our limited measurements were not enough to detect a correlation between metabolism and weather. Therefore, more investigation is needed to determine how weather or even climate influences carbon cycling and ultimately carbon burial in shallow prairie lakes.

I did not observe hydrological variables that might be affected by weather or climate such as groundwater exchange, which in turn may have influenced lake metabolism. Still, I conclude that there was a local and/or regional mechanism that influenced metabolism because 1) the metabolism of the two intensively sampled lakes closely resembled the rates estimated for all nine lakes in 2007 and 2008, and 2) similar cycling and noise in oxygen and temperature patterns in two completely separate lakes of differing regimes were apparent throughout the sampling period. Together these observations suggest that lake regime did not play a strong role in predicting whether

prairie lakes were net heterotrophic or autotrophic during the open water season; rather, an overlying regional influence on local metabolism must exist.

Table 1. Summary of water chemistry and metabolism for all nine lakes studied from 2006 to 2008. The intensively monitored lakes are marked with an asterisk (*). The state "C" means clear lakes, and "T" signifies turbid lakes. Area is in km^2. z_{max} is the maximum depth in meters recorded for each lake. Chl is reported in $\mu g\ L^{-1}$ and DOC is in $mg\ L^{-1}$; both are seasonal means. "Days used" signifies the total number of days that metabolism was successfully measured and have been included in all analyses. "Days lake specific wind" means the number of days that the wind measurements were used for a specific lake; if this number does not equal the "Days used" column, then the Fergus Falls wind data were used instead to estimate metabolism on the days without the lake specific wind data. GPP, R and NEP are seasonal average values for each lake and are presented in $mmol O_2\ m^{-2}\ d^{-1}$ ± standard deviation.

Lake	State	Area	Year	z_{max}	Chl	DOC	Metabolism measured	Days used	Days of lake specific wind	GPP	R	NEP
8Mile	C	0.26	2007	1.5	17	12.6	8/8 to 8/19	7	7	385 ± 164	-384 ± 187	1 ± 84
Lake Christina	C	20	2007	1.1	10	8.4	7/13 to 8/30	18	18	307 ± 120	-230 ± 115	76 ± 119
Lee	C	0.2	2007	4.5	34	12.9	6/13 to 9/2	10	10	285 ± 140	-294 ± 127	-9 ± 74
Mavis East*	C	0.22	2006	3.3	12	14.3	6/15 to 7/21	17	17	344 ± 158	-299 ± 152	45 ± 107
Mavis East*	C	0.22	2007	3.3	14	14.0	5/11 to 11/2	107	44	205 ± 119	-221 ± 101	-16 ± 97
Mavis East*	C	0.22	2008	3.3	13	12.7	6/6 to 8/25	26	26	143 ± 76	-189 ± 68	-47 ± 85
Pisa	C	0.07	2007	1.1	17	19.5	6/28 to 9/10	6	1	395 ± 191	-385 ± 154	10 ± 128
Pisa	C	0.07	2008	1.1	2	18.2	6/6 to 8/18	40	30	246 ± 101	-235 ± 123	11 ± 78
Bore	T	0.07	2007	1.5	166	23.3	6/13 to 9/20	27	0	377 ± 189	-404 ± 188	-27 ± 91
Bore	T	0.07	2008	1.5	207	26.4	6/26 to 8/3	10	10	214 ± 242	-394 ± 258	-180 ± 170
Mavis West*	T	0.14	2006	3.6	68	14.8	7/4 to 7/29	18	18	800 ± 243	-736 ± 173	65 ± 202
Mavis West*	T	0.14	2007	3.6	77	14.4	5/12 to 11/1	71	26	310 ± 135	-361 ± 117	-51 ± 92
Mavis West*	T	0.14	2008	3.6	76	14.3	6/6 to 11/2	73	73	449 ± 286	-426 ± 279	-34 ± 171
Morrison	T	0.15	2007	2.2	68	16.2	6/2 to 8/9	12	9	271 ± 111	-201 ± 141	70 ± 89
Murk	T	0.15	2007	2.2	48	19.5	6/28 to 9/21	33	15	168 ± 114	-139 ± 128	29 ± 88

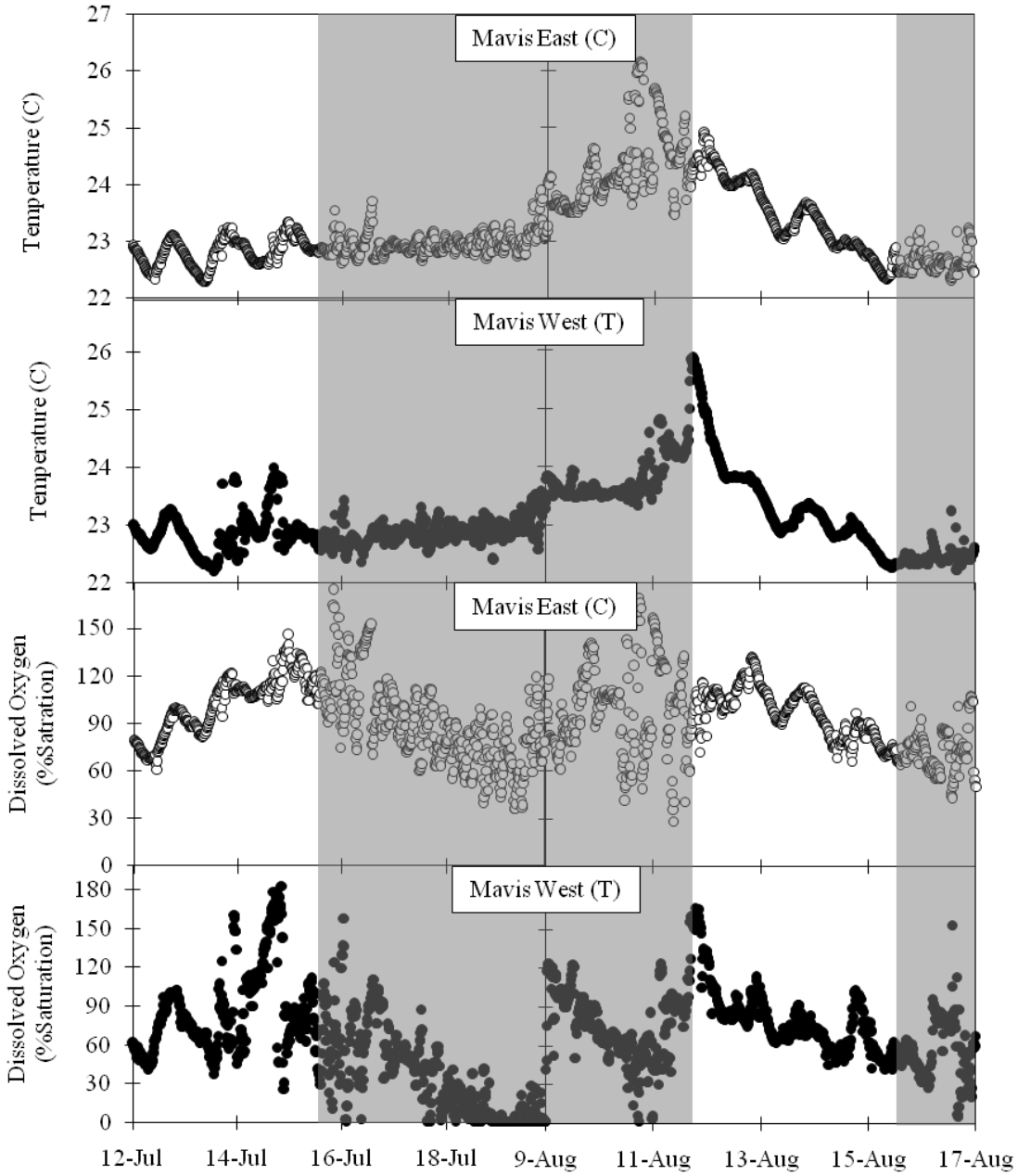

Figure 1. Microstratification patterns observed simultaneously in two separate lakes. Temperature and dissolved oxygen in the intensively sampled lakes during two time periods in July and August 2007. The white circles represent the clear (C), macrophyte-dominated lake, and the black circles represent the turbid (T) phytoplankton-dominated lake. These readings were taken simultaneously in two separate lakes at the same depth, and experienced similar "noise" patterns in the oxygen and temperature signals (marked in the shaded grey areas).

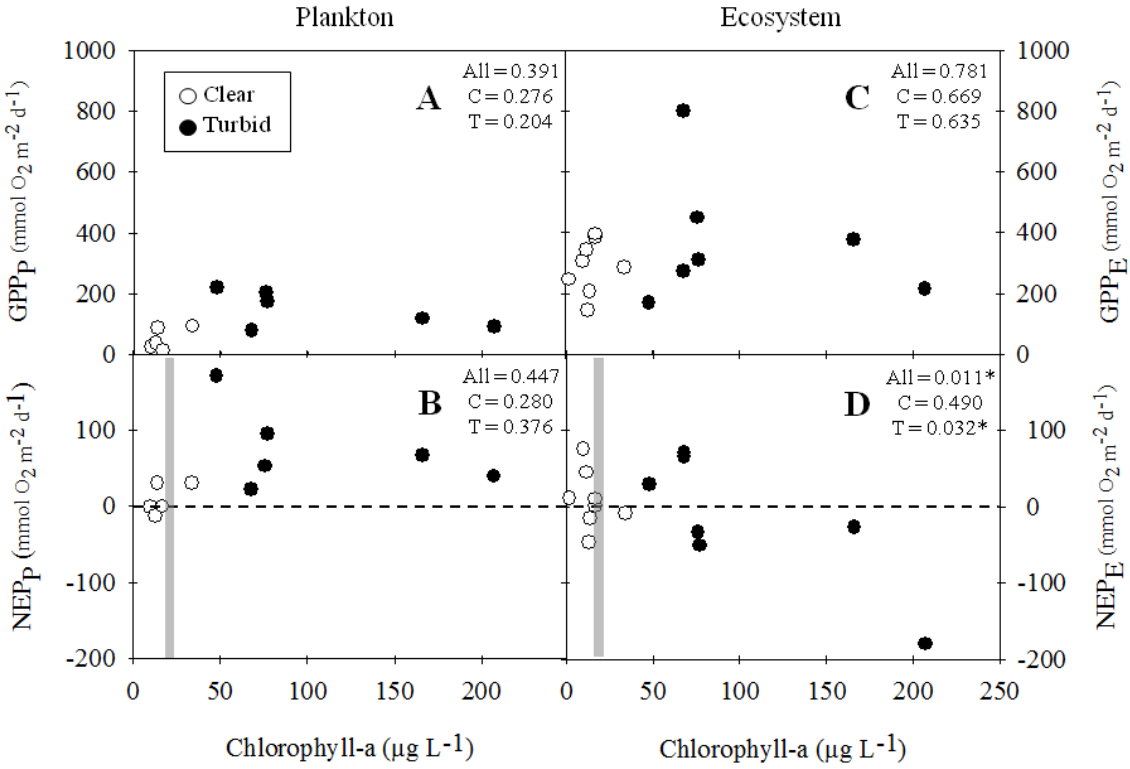

Figure 2. Relationship between Chl and GPP_P (A) and NEP_P (B) for the annual planktonic metabolic measurements, and GPP_E (C) and NEP_E (D) for the annual whole ecosystem metabolism. White circles represent the lakes in the clear regime, and the black circles represent lakes in the turbid regime. The numbers within the box represent *p* – values for all lakes (All), clear lakes (C), and turbid lakes (T), with an asterisk (*) representing a significant relationship. For the NEP rates, positive values indicate net autotrophy (GPP>R), and negative values represent net heterotrophy (R>GPP). The dotted line represents a perfect balance of GPP and R. The shaded grey line on the x-axis indicates the threshold of 17-20 µg L^{-1} Chl; net autotrophic and heterotrophic lakes are predicted to be below and above the threshold, respectively.

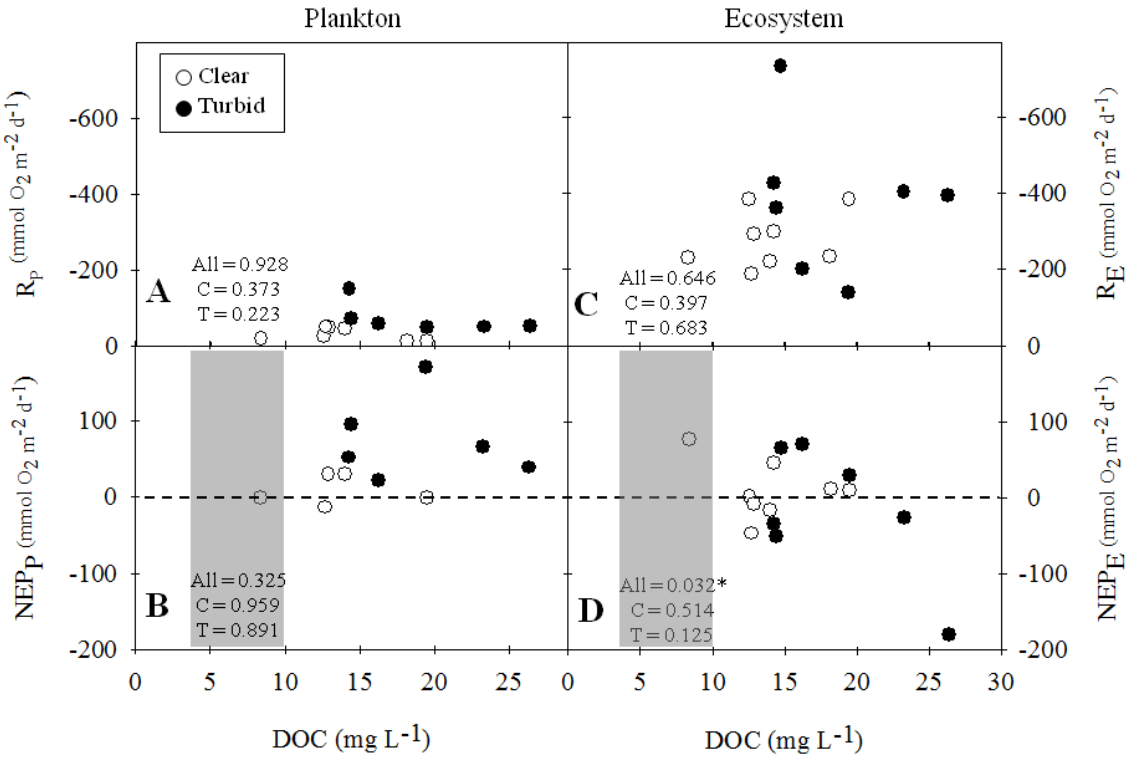

Figure 3. Relationship between DOC and R_P (A) and NEP_P (B) for the planktonic metabolic measurements, and GPP_E (C) and NEP_E (D) for the whole ecosystem metabolism. White circles represent the lakes in the clear regime, and the black circles represent lakes in the turbid regime. The numbers within the box represent *p* – values for all lakes (All), clear lakes (C), and turbid lakes (T), with an asterisk (*) representing a significant relationship. For the NEP rates, positive values indicate net autotrophy (GPP>R), and negative values represent net heterotrophy (R>GPP). The dotted line represents a perfect balance of GPP and R. The shaded grey area on the x-axis indicates the threshold of 4-10 mg L^{-1} DOC; net autotrophic and heterotrophic lakes are predicted to be below and above the threshold, respectively.

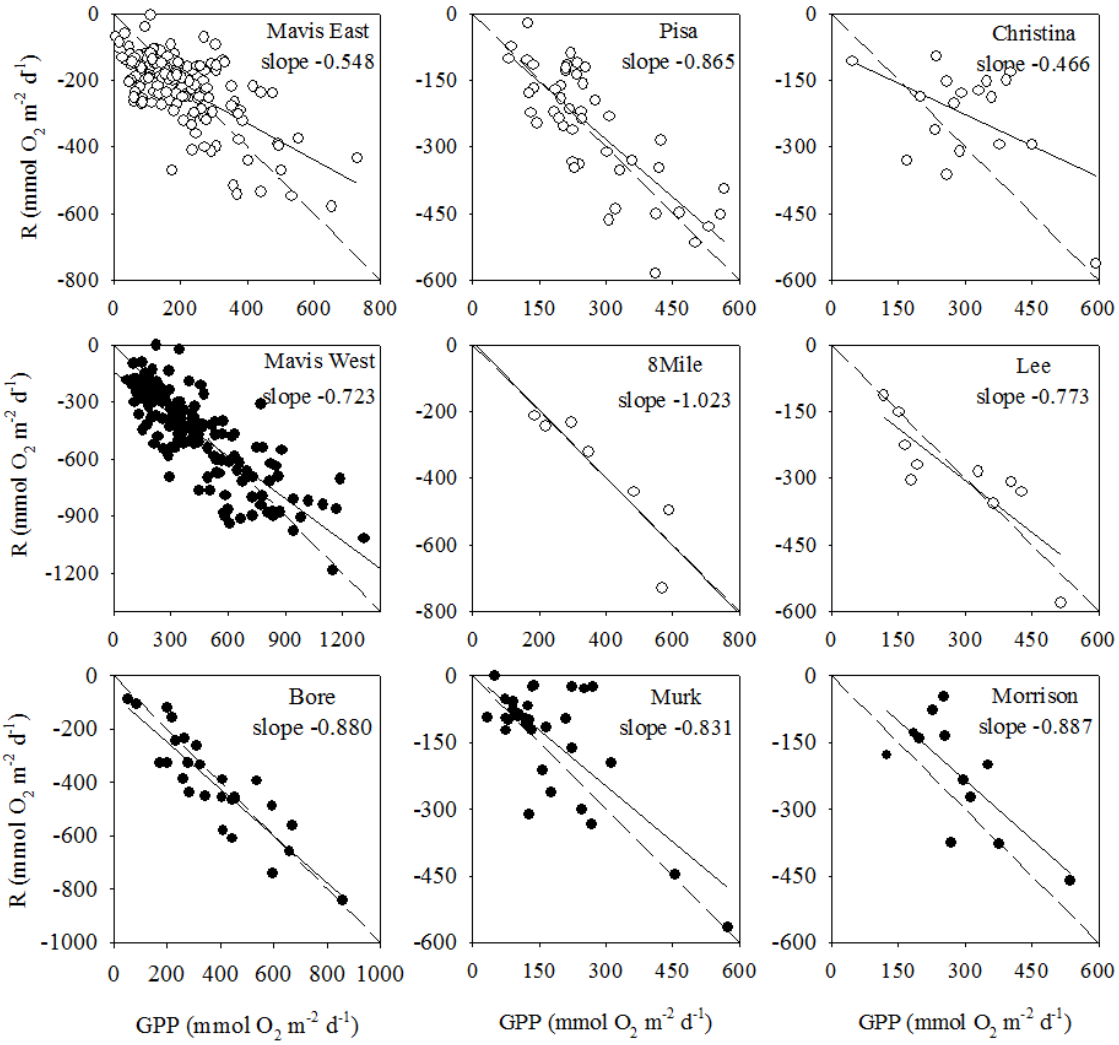

Figure 4. Relationship between GPP and R for all nine lakes studied during all three years. White circles represent clear lakes, and black circles represent turbid lakes. Data points represent daily metabolic estimates in a given lake. The solid line represents the trendline, and the dashed line represents the ratio of GPP:R at unity (or 1:1). Many lakes seem to be heterotrophic (below the 1:1 ratio or R > GPP) at low gross primary production, and autotrophic (above the 1:1 ratio or GPP > R) at high gross primary production. Please note that not all axes are the same.

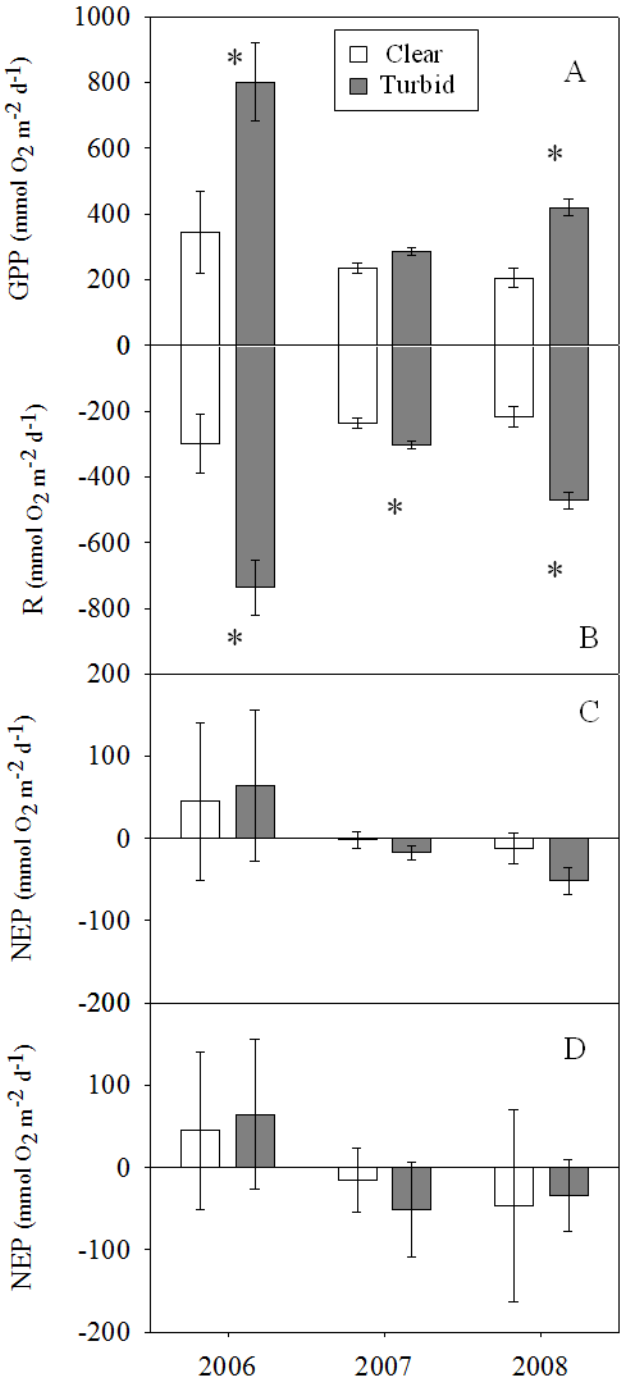

Figure 5. Gross primary production (GPP), respiration, (R), and net ecosystem production (NEP) for all lakes (A-C), and the intensively monitored lakes (D). White bars represent the clear lakes, and the grey bars represent the turbid lakes. Two intensively sampled lakes were monitored in 2006, and 9 lakes (five clear, four turbid) were monitored in 2007, and four lakes (two clear and two turbid) were monitored in 2008. Error bars represent standard error. (*) Represents a significant ($p < 0.05$) difference between regimes.

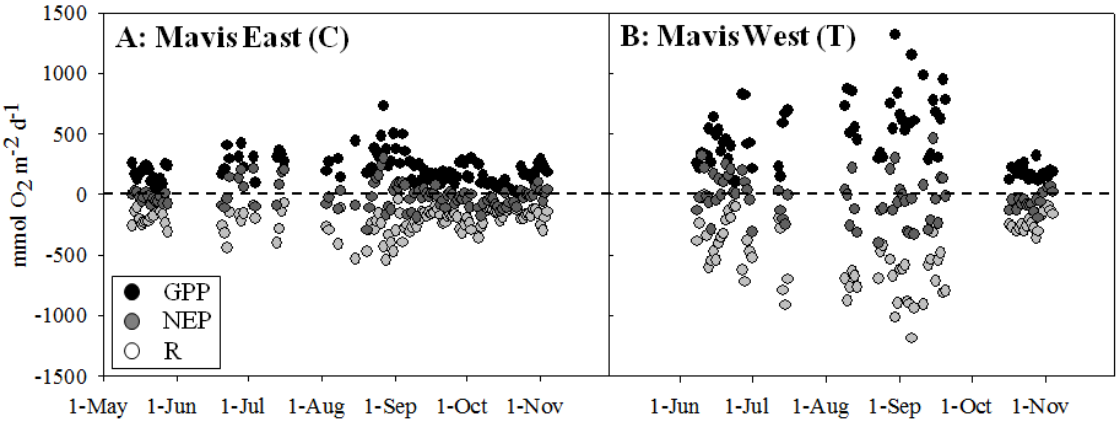

Figure 6. GPP (black circles), R(white circles), and NEP (grey circles) over the 2007 open water season in the intensively monitored (A) clear-water, macrophyte dominated lake, and a (B) turbid-water, phytoplankton dominated lake.

Chapter 2: Factors Influencing Carbon Dioxide Flux in Prairie Pothole Shallow Lakes under Alternative Trophic Regimes

Small, shallow freshwater ecosystems such as wetlands and wetland lakes potentially function as a large source of carbon dioxide to the atmosphere because they are very productive, and can respire a high proportion of organic matter they receive from their terrestrial surroundings. To improve global carbon cycling models, it is important to estimate the carbon dioxide flux from multiple regions globally. The Prairie Pothole Region of North America contains 207,000 km^2 of shallow, hard-water wetlands, and therefore serves as an important region to study carbon dioxide exchange. Wetland lakes in this region usually exist in either a clear, macrophyte-dominated regime, or a turbid, phytoplankton-dominated regime. Furthermore, these regimes have differing physical and chemical properties that could influence the carbon cycle. I investigated carbon dioxide exchange between the atmosphere and nine shallow lakes in the southeastern part of the Prairie Pothole Region over three years, and measured whole ecosystem gross primary production (GPP), respiration (R), pH, and dissolved organic carbon (DOC) to determine the best predictor of carbon dioxide exchange rates and direction. For all nine lakes, the annual carbon dioxide exchange varied from 53 mmol C m^{-2} d^{-1} influx to the lakes to 97 mmol C m^{-2} d^{-1} efflux out of the lakes to the atmosphere, with an overall mean flux of 9.5 mmol C m^{-2} d^{-1} from the lakes to the atmosphere. The magnitude and direction of annual net exchange varied interannually, but not with regime. Unlike previous studies, there was no positive correlation between pCO_2 and DOC; however, pH was significantly

positively correlated with pCO_2 and its exchange with the atmosphere in both the clear and turbid regimes. Although pH was strongly influenced by metabolism, CO_2 flux did not significantly correlate with metabolism due to the high buffering capacity of these hard-water shallow lakes. Current paradigms predict that autotrophic lakes should be a CO_2 sink and that heterotrophic lakes should be a CO_2 source; however, that is not what I observed. Here I present evidence that hard-water, shallow lakes can be net heterotrophic yet function as a carbon sink, and alternatively emit CO_2 to the atmosphere while they are net autotrophic. It is likely that climate and hydrologically driven geochemical factors play an as yet unappreciated role in CO_2 exchange in these biologically dynamic wetlands through space and time.

Introduction

Freshwater ecosystems play an important role in global carbon cycling due to intensive carbon burial and exchange of carbon dioxide (CO_2) with the atmosphere (Dean and Gorham 1998, Cole et al. 2007; Tranvik et al. 2009). The surface waters of most lakes and rivers worldwide are supersaturated with CO_2, and therefore typically function as carbon sources to the atmosphere (Cole et al. 1994, Cole and Caraco 1998, Duarte and Agustí 1998, Hanson et al. 2004, Rantakari and Kortelainen 2005, Sobek et al. 2005, Kortelainen et al. 2006). Supersaturation of CO_2 is partially due to the mineralization of allochthonous carbon, which can be transported to lakes via groundwater input or surface runoff during precipitation events. In comparison to larger lake ecosystems, wetland lakes typically have a high concentration and evasion of CO_2 (Kelly et al. 2001, Roehm

et al. 2009). Because a high percentage of their volume is associated with the perimeter of the lake, organic and inorganic carbon inputs from the terrestrial surroundings or the lake sediments are readily transported into surface waters where they can be mineralized, precipitated and/or dispersed into the atmosphere as free CO_2. Quantitatively, most of the lakes of the world are very small (Downing et al. 2006), yet we know very little about the biogeochemical controls of CO_2 exchange in these small lakes globally.

Predicting the partial pressure of CO_2 (pCO_2) within lake waters and the flux between the lake and the atmosphere is often difficult, as both are influenced by biological, chemical, and physical processes which can fluctuate on very short time scales. The flux of CO_2 between the lake and the atmosphere depends on 1) the difference between the CO_2 concentration of each fluid, and 2) the physical controls affecting exchange rates (i.e. temperature and wind speed; Sobek et al. 2005, Cole and Caraco 1998). pCO_2 in the lake is influenced by the metabolic activity of the lake (both uptake of inorganic and mineralization of organic carbon), the direct transport of both organic and inorganic carbon via groundwater input and surface runoff (Kling et al. 1991), and chemical reactions related to alkalinity (i.e. carbonate dissolution and precipitation, Finlay et al. 2009). Rainfall and snowfall events promote the movement of water through the landscape, which consequently transfers carbon subsidies (such as dissolved organic and inorganic carbon, DOC and DIC, respectively) from the watershed to the lake, where they could potentially be released into the atmosphere (del Giorgio et al. 1999, Pace and Prairie 2007).

Often in freshwaters DOC concentrations positively correlate with pCO_2, and since DOC serves as a substrate for microbial respiration, numerous comprehensive studies have found that DOC is the most consistent predictor of pCO_2 in lakes worldwide (Kelly et al. 2001, Hanson et al. 2004, Sobek et al. 2003, Sobek et al. 2005, Roehm et al. 2009). Small lakes with short residence times tend to be high in DOC due to the high input from terrestrial ecosystems, and the low volume:perimeter ratio also favors allochthonous input (Roehm et al. 2009). Furthermore, some lakes experience a peak in CO_2 immediately following precipitation events because the rainfall transports CO_2 and organic carbon from the catchment to the lake, which can boost microbial respiration (Rantakari and Kortelainen 2005, Roehm et al. 2009, Stets et al. 2009). Similar to DOC, pCO_2 often correlates with the metabolism of a lake because the balance between gross primary production (GPP) and respiration (R) directly influences CO_2 production (via R) and consumption (via GPP) (Yavitt 1997, Duarte and Prairie 2005). Lastly, hard-water lakes with high alkalinity have among the highest CO_2 evasion estimates reported because chemical reactions involving carbon dioxide, bicarbonate, carbonate, water and calcium can cause a strong increase in pCO_2 due to carbonate precipitation under saturating conditions (pH > 8.5) (McConnaughey et al. 1994, Finlay et al. 2009, Stets et al. 2009).

The Prairie Pothole Region (PPR) of North America contains 207,000 km^2 of small, hard-water wetlands (Euliss et al. 2006), and serves as an ideal location to investigate CO_2 exchange because the lakes in this region could support a substantially large flux of CO_2 to the atmosphere due to their size, total area, and biological

characteristics. The PPR landscape and its wetlands are rich in calcium and magnesium carbonates, which influence the level of alkalinity in these lakes. These cations can precipitate with carbonates, and cause the release of CO_2, depending on the pH (McConnaughey et al. 1994, Finlay et al. 2009). Furthermore, many wetlands within the landscape of the PPR receive groundwater discharge, which in turn depends on the frequency and intensity of precipitation events (Winter and Rosenberry 1995). These lakes also have high concentrations of DOC which typically favors high rates of CO_2 production (Kelly et al. 2001, Prairie et al. 2002, Sobek et al. 2003, Hanson et al. 2004, Sobek et al. 2005, Roehm et al. 2009). Lastly, the lakes in the PPR are productive (up to 1680 g C m^{-2} yr^{-1}, Chapter 1), and tend to exist in two relatively stable regimes: one regime that is in the clear-water state and dominated by submerged macrophytes and macroalgae (hereafter clear regime) and the other in the turbid-water state dominated by phytoplankton and microalgae (hereafter turbid regime) (Scheffer et al. 1993, Zimmer et al. 2001). These two regimes could potentially influence pCO_2 and its exchange because CO_2 evasion often negatively correlates with primary production rates (Yavitt 1997, Huttunen et al. 2003, Kortelainen et al. 2006), which also can differ between macrophytes and phytoplankton (Goulder 1969, Rørslett et al. 1986, Mitchell 1989, Blindow et al. 2006). Most studies that have evaluated the relationship between the metabolic activity of the lake and pCO_2 have not examined lakes with a large range of productivity (Huttunen et al. 2003, Finlay et al. 2009), or have included the influence on metabolism from the benthic community (Kelly et al. 2001, Finlay et al. 2009). The contribution of the benthic community to the whole ecosystem metabolism in PPR

wetlands is high (approximately 82% of respiration, and 61% gross primary production, Chapter 1) because they are so shallow. Furthermore, their production rates are substantial in comparison to larger freshwater ecosystems (Lake Superior ~200 to 350 mg C m^{-2} d^{-1} [Sterner 2010], versus up to 9600 mg C m^{-2} d^{-1} in PPR lakes, Chapter 1). Although there has been extensive research on CO_2 exchange and its predictors in wetlands in the boreal regions of North America and across South America (del Giorgio and Peters 1994, Finlay et al. 2009, Kosten et al. 2010), comprehensive estimates of CO_2 exchange and its predictors in the North American prairie regions, and particularly in the shallow wetland lakes that are most common in these systems, are lacking.

In order to investigate the impact of small, hard-water prairie wetlands on the regional carbon cycle, I estimated the exchange of CO_2 between the lake and the atmosphere in nine lakes in the PPR. I determined whether pCO_2 and its exchange between the lake and the atmosphere were related to either biological (i.e. metabolism) or physical-chemical factors (pH; hydrological processes delivering carbon to the lake). If controlled by biological factors, I would expect pCO_2 and its exchange to differ between the clear and turbid regimes, given a difference in the balance between GPP and R between the regimes (Chapter 1). For example, CO_2 evasion may be lower in clear lakes if primary production exceeds respiration rates. If pCO_2 and its exchange are not influenced by lake regime, I would expect physical-chemical factors such as pH, climate and/or hydrology to be the most important factors in determining CO_2 exchange in these PPR shallow lakes.

Methods

General Description

This study was conducted in the southeastern portion of the Prairie Pothole Region. Nine shallow lakes in west-central Minnesota were monitored and sampled from May 2006 through August 2008. All lakes were located within Waterfowl Production Areas, averaged 0.15 km^2 in surface area (excluding one lake: Lake Christina 20 km^2), and had a mean maximum depth of 2.1 meters. Lakes with a mean of < 22 or > 31 μg Chl L^{-1} were classified in the clear and turbid regime, respectively (Zimmer et al. 2009). Over the course of the study, five lakes were in the clear regime, four lakes were in the turbid regime (Table 1).

Variables for the Estimation of pCO_2 and Related Factors

pCO_2 can be measured directly using the headspace technique (Cole and Caraco 1998), or indirectly using measurements of water temperature, pH, and dissolved inorganic carbon (DIC). I used the latter method to estimate pCO_2 within the lakes over time. I recorded lake temperature and pH every 15 minutes using multi-probe sondes (Hydrolab DS5X or Hydrolab Minisonde 4a) deployed in the mixing layer in the center of each lake. Over the sampling period from 2006 to 2008, one turbid and one clear lake were continuously monitored using a Hydrolab DS5X, and the other seven lakes were monitored less frequently (described below). The two lakes that were monitored continuously (Mavis East and Mavis West) served as the best contrast between a clear and turbid regime as both have similar morphometry, average depth, and they are located

less than 100 meters away from each other. These two lakes will be referred to as the "intensively monitored" lakes throughout the remainder of this manuscript. During 2007, three additional sondes (two Hydrolab DS5X and one Hydrolab Minisonde 4a) were rotated between the other seven lakes; the sondes were deployed in each of these lakes for two-week intervals during three separate periods. In addition to the intensively monitored lakes, one additional sonde (Hydrolab Minisonde 4a) was alternately deployed for two week intervals between another clear and turbid lake in 2008. Over all years, the data were collected from the sondes every 2-3 weeks during the ice-free season and recalibrated in the field following Hydrolab recommendations.

Wind turbulence influences the exchange of CO_2 between the wetland and the atmosphere (Cole and Caraco 1998). Therefore, wind speed was measured at the same time increments as the sonde measurements. An Onset HOBO Wind Speed and Direction Sensor with a HOBO Micro Station Data Logger was used to measure wind speed at the two intensively monitored lakes 1.5 meters above lake level on the shoreline. Also, a Brunton ADC Summit (which was bound to a wind vane, and secured on top of an aluminum extension pole) was used to monitor wind speed at the other seven lakes approximately 1 m directly above the center of the lake. All anemometers were calibrated against each other to account for any differences. When the wind data were unavailable at a specific lake due to equipment malfunctions, wind speed measurements from the Fergus Falls Municipal Airport in MN (on average 40 km from the lakes) were used to calculate NEP. The wind speed measurements from the airport were compared to those taken at the exact time at an intensively monitored lake in order to account for any

differences. Comparing 457 simultaneous readings revealed that the airport readings were 1.7 ± 1.5 (SD) times higher than the wind speed readings from the lake, on average, and were corrected appropriately. Daily precipitation estimates were also obtained from the Fergus Falls Municipal Airport.

Water samples were taken every 2-3 weeks from May through August. The water samples from each lake were collected from 0.2 m below the lake surface, and placed on ice until filtration within a few hours after collection. The water samples were analyzed for DIC and DOC concentrations, cation concentrations, and alkalinity. Samples for DOC, DIC, and cation concentrations, were filtered through an ashed (450°C for 4 h) Whatman GF/F filters. For DIC, an ashed 7 mL scintillation vial was over-filled with 0.7 µm filtrate void of headspace, sealed with Parafilm, and run within 24 hours on a Shimadzu TOC-Vcsh. During one sampling date whole water was analyzed for DIC, and it was determined that approximately 1% of the DIC was lost due to degassing of CO_2. For DOC, the filtrate was immediately preserved with hydrochloric acid to lower the pH below 2. A Shimadzu TOC-Vcsh was used to estimate the concentration of DOC in the water samples. The concentrations of cations within the water were estimated from samples collected during April, July, and October of 2007 for the intensively sampled lakes, and during July for the other seven lakes. The cation concentrations were determined using inductively coupled plasma (ICP) atomic emission spectrometry, and were used to calculate water hardness, which is described below. The alkalinity of each lake was measured on each sampling date. Briefly, the water was collected in an amber HPVC bottle void of air, and was kept at lake temperature prior to analysis (analysis

typically occurred 2-3 days after collection). I used the Gran titration method, and titrated 50-mL of the water sample beyond the equivalence point using 0.02 N $H_2SO_4^-$ while constantly measuring the pH (Gran 1952). Also, at each sampling date, the air was sampled approximately 1 m above the lake using a nylon syringe. To determine the CO_2 concentration of the air, the sample was analyzed using a Shimadzu GC-14A gas chromatograph with thermal conductivity detection, a Poropak N column, and 645, 1025, and 10,000 ppm standards. The flux of CO_2 based on global measurements of atmospheric carbon dioxide levels from Mauna Loa (http://www.esrl.noaa.gov/gmd/ccgg/trends/) was estimated. Finally, ecosystem metabolic rates of gross primary production, respiration, and net ecosystem production were made using continuous measurements of oxygen, temperature, and wind speed (Cole et al. 2000, Hanson et al. 2003, van de Bogert et al. 2007, Coloso et al. 2008). Refer to Chapter 1 ("Net Ecosystem Production in Prairie Pothole Shallow Lakes under Alternative Trophic Regimes") for details.

Calculating the Concentration and Exchange of pCO_2

The flux of CO_2 between the atmosphere and the surface waters of the lake was calculated using the equation:

$$\text{Flux} = \alpha\, k\, (P_{gas}K_h - [\text{gas}]_{sat}) \quad (1)$$

where α is the chemical enhancement factor of CO_2 gas exchange, k represents the gas transfer velocity, P_{gas} is the partial pressure of CO_2 in the surface water, K_h is Henry's constant for CO_2 at a given temperature and salinity (Weiss 1974), and $[\text{gas}]_{sat}$ represents

the concentration of CO_2 the surface water would have in equilibrium with the overlying atmosphere (Cole and Caraco 1998).

The chemical enhancement factor (α) is important especially in alkaline lakes and environments with low wind speed (i.e., 3-4 m s^{-1} on average) (Wanninkhof and Knox 1996, Finlay et al. 2009). In lakes with a high pH, the flux of CO_2 into the lake is enhanced due to free CO_2 reacting directly with hydroxide to quickly form bicarbonate, rather than hydrating to form carbonic acid and subsequently bicarbonate. Given the chemical reactions:

$$CO_2 + H_2O \leftrightarrow H^+ + HCO_3^- \leftrightarrow H^+ + CO_3^{2-} \quad (2)$$

high pH favors conversion of the CO_2 into carbonate, which may precipitate out depending on pH, ionic strength, and temperature. Therefore, at low wind speeds and high alkalinity, CO_2 invasion rate into the lake increases. The enhancement factor is estimated using the Hoover and Berkshire model (1969) described in Wanninkof and Knox (1996):

$$\alpha = T/[(T - 1) + \tanh (Qz)/(Qz)] \quad (3)$$

where $Q = (rT/D)^{0.5}$, $r = r_1 + r_2 (K_w / a_H)$, $T = 1+(a_H)^2(K_1K_2 + K_1a_H)^{-1}$, and $z = D/k$. In these calculations, r_1 is the first order rate constant for the reaction $CO_2 + H_2O = H_2CO_3$ (0.025 s^{-1}, Stumm and Morgan 1981), and r_2 is the zero order rate constant for the reaction $CO_2 + OH^- = HCO_3^-$ (7.6 x 10^4 L mol^{-1} s^{-1}, Johnson 1982). K_w is the apparent dissociation constant for water (10^{-14}), a_H is the activity coefficient for CO_2 ($0.91*10^{-pH}$, Stumm and Morgan 1981), D is the molecular diffusion coefficient for CO_2

([0.61563+0.05316*T°C]*0.00001), and K_1 and K_2 are the first and second apparent dissociation constants for carbonic acid which is based on temperature.

The gas transfer velocity of CO_2 (k) depends on wind speed, water temperature, and the salinity of the surface water (Liss and Merlivat 1986, MacIntyre et al. 1995, Cole and Caraco 1998), and is normalized to a Schmidt number of 600 (Wanninkof and Knox 1996). The k for CO_2 was calculated using the equation from Cole and Caraco (1998):

$$k_{600} = (2.07 + 0.215*\mu_{10}^{1.7})*(Sc/600)^n \quad (4)$$

where k_{600} is the standard gas transfer velocity in cm h^{-1}, μ_{10} represents the wind velocity (m s^{-1}) at 10 m above the lake surface, Sc denotes the Schmidt number, which is the kinematic viscosity divided by the diffusion coefficient of CO_2 (Jähne et al. 1987, MacIntyre et al. 1995) and is based on the surface water temperature and salinity (MacIntyre et al. 1995). Finally, n represents the power relationship for the equation, and equals -0.67 when $\mu_{10} \leq 3.6$ m s^{-1} or -0.5 when $\mu_{10} > 3.6$ m s^{-1} (Liss and Merlivat 1986). After a series of observations, the wind speed at 10 m above the lake surface has been found to be approximately 1.29 m s^{-1} faster than the wind speed 1 m above the lake; therefore the wind speed at 10 m (μ_{10}) above the lake was estimated by multiplying the recorded wind speed measurements by a factor of 1.29 (Liss and Merlivat 1986).

I estimated aqueous CO_2 from continuous pH readings and DIC measurements made every two weeks. Corrections were made for temperature, altitude and ionic strength (Kling et al. 1992, Cole and Caraco 1998).

$$[CO_2\ \mu M] = DIC\ (\mu M) * \alpha_0 \quad (5)$$

$\alpha_0 = (1 + K_1/[H^+] + K_1K_2/[H^+]^2)^{-1}$ and the dissociation constants for carbonic acid (K_1 and K_2) were calculated from pK values. The pK values were corrected for ionic strength (*I*) and equaled pk'. For example $pK_1' = pK_1 - (0.5\sqrt{I})/(1 + 1.4\sqrt{I})$ and $pK_2' = pK_2 - (2\sqrt{I})/(1 + 1.4\sqrt{I})$. Ionic strength (*I*) was calculated by 4*Hardness (M) – Alkalinity (equivalents L^{-1})/2 (Stumm and Morgan 1981). Hardness (M) was calculated as 2*Ca + 2*Mg + Na (each in M units) (Stumm and Morgan 1981).

The gas that should be saturated in the water based on the concentration of CO_2 in the overlying atmosphere was measured using an air sample and multiplying it by Henry's constant and corrected for elevation (Kling et al. 1992).

Statistical Analysis

I used regression analysis to determine significant correlations between 1) DOC and R, R and pCO_2, DOC and pCO_2, 2) precipitation and DOC, R, and pCO_2, 3) pH and pCO_2, the chemical enhancement factor and CO_2 flux, and 4) pCO_2 and GPP, R, and NEP. All relationships with a *p*-value of <0.05 were considered significant correlations. I report the findings for all lakes from May to August within a given year.

Results

Due to sampling constraints, DIC and the content of Ca^{2+} and Mg^{2+} (to estimate hardness) could not be measured as frequently as temperature, pH, and wind speed. Therefore one value of DIC was assigned between the sampling intervals, and one concentration of Ca^{2+} and Mg^{2+} were used for all CO_2 estimates. I used Morrison Lake as

an example to evaluate the sensitivity of the flux model to changes in DIC, Ca^{2+} and Mg^{2+}. DIC rarely changed more than 4% over the course of 2-3 weeks in all of the lakes, and I observed that a 4% change in DIC resulted only in a 1% change in CO_2 flux. Additionally, a 50% decrease and increase in hardness (to equal the minimum and maximum observed hardness, respectively) only resulted in a 1% decrease and increase in CO_2 flux, respectively. Therefore, assuming that DIC or Ca^{2+} and Mg^{2+} did not change between sampling intervals likely had a very low impact on overall CO_2 flux in these lakes.

Due to the high pH environment of these lakes (average 8.7), only approximately 3% of the total DIC was actually in the free CO_2 form for all lakes during the study period. The lakes were considered hard-water lakes as they contained 16-70 mg L^{-1} of Ca^{2+} and 40-120 mg L^{-1} Mg^{2+}. The total DIC ranged from approximately 25-130 mg L^{-1} (Table 2).

pH and the flux of CO_2 between the lake and the atmosphere changed throughout the season, and differed between years in both the clear and turbid regimes (Figure 7). The CO_2 flux was highly variable between years, and though it seemed to differ in 2006 from other years, the direction of the CO_2 flux was not influenced by the lake regime when considering all three years (Figure 7a,b). Over the three years, the wetlands were a net carbon source to the atmosphere, contributing an average of 9.5 mmol CO_2 m^{-2} d^{-1} from May through August. The flux of the intensively sampled lakes closely resembled the average of all lakes in 2007 and 2008 (Figure 7).

DOC did not correlate with R (p=0.646, R^2=0.017, Figure 8a), and greater R did not coincide with higher pCO_2 (p =0.618, R^2=0.001, Figure 8b). Furthermore, unlike other studies of north temperate lakes, pCO_2 did not correlate with DOC concentrations (p=0.672, R^2=0.004) (Figure 8c). There was no correlation between precipitation and DOC and R (p =0.387 and 0.658, respectively; R^2=0.028 and 0.005, respectively, Figure 9a,b), but there was a significant, positive relationship between atmospheric precipitation and pCO_2 (p =0.005, R^2=0.170, Figure 9c).

pH was strongly negatively correlated with pCO_2 both in the clear and turbid regimes (both $p < 0.0001$) (Figure 10a). In each of the regimes, the chemical enhancement factor correlated positively with pH, as determined by the CO_2 flux calculations (both $p < 0.0001$) (Figure 10b). Finally, the exchange of CO_2 between the lake and the atmosphere was strongly predicted by pH in both regimes (both $p < 0.0001$) (Figure 10c). The wetlands in the clear regime experienced a larger daily range in pH than the wetlands in the turbid regime. pH fluctuated on a daily basis in a way similar to oxygen (Figure 11).

I assessed the relationship between ecosystem metabolism and pCO_2 for the clear and turbid regimes separately. For the wetlands in the turbid regimes, there was a significant, negative correlation between pCO_2 and GPP (p<0.0001, R^2=0.163), and surprisingly, a significant negative correlation between pCO_2 and R as well (p<0.0001, R^2=0.137) (Figure 12a,b). However, there was no significant relationship between pCO_2 and NEP in the turbid regimes (p=0.272) (Figure 12c). Alternatively, the lakes in the clear regime demonstrated opposing patterns to the lakes of the turbid regime. There was

a significantly positive relationship between pCO_2 and GPP (p=0.015, R^2=0.041), and pCO_2 and R (p=0.001, R^2=0.082) (Figure 12d,e). There was no significant correlation between pCO_2 and NEP (p=0.285) (Figure 12f). Overall, alkalinity was significantly higher in the turbid regimes compared to the clear regimes (6.7 versus 3.9 meq L^{-1}, respectively; p=0.060) (Figure 13), and on average, alkalinity dropped by 1.05 and 0.04 meq L^{-1} in lakes of the clear and turbid regime, respectively. The standard deviation around the mean pH on a daily basis decreased with increasing alkalinity (Figure 14).

I evaluated the net exchange of CO_2 with the atmosphere relative to the lakes' net metabolic behavior (net heterotrophic or net autotrophic), in order to determine if heterotrophic lakes were typically carbon sources to the atmosphere, and if net autotrophic lakes were carbon sinks. Most past research suggested that lakes and wetlands are inorganic carbon sinks when they are net autotrophic and sources when they are heterotrophic. However, I found that both clear and turbid lakes can be autotrophic *and* carbon sources or heterotrophic *and* carbon sinks as well (Figure 15). In fact, there was about an equal number of days when the lakes were in these previously undescribed patterns (net heterotrophic and carbon sinks and net autotrophic and carbon sources) (Figure 15).

Discussion

I evaluated carbon dioxide exchange between the lake and the atmosphere in nine shallow lakes in the southeastern portion of the Prairie Pothole Region, and tested whether lake regime (macrophyte- or phytoplankton-dominated) correlated with CO_2

exchange. In contrast to many lakes around the world, I did not detect a positive correlation between the concentration of DOC and the evasion of CO_2, suggesting that DOC was not an important control of net CO_2 exchange. However, in these hard-water ecosystems, pH strongly influenced the concentration and therefore the flux of CO_2 in the surface waters. Although metabolism clearly influenced pH, it rarely drove pH low enough to strongly influence CO_2 fluxes. GPP and R of phytoplankton-dominated regimes were significantly higher than the rates of the macrophyte-dominated lakes; however, the balance between GPP and R (i.e. NEP) in both regimes did not significantly differ so consequently lake regime had no strong direct correlation with pCO_2. Nevertheless, lake regime did have an indirect effect on the flux of CO_2 because the alkalinity was significantly higher in the turbid lakes, most likely due more permanent precipitation of calcium carbonate in the clear regimes. Low turbulent environments reduce the chances of precipitates to be resuspended in the water column, and the macrophytes provided a surface at which to the calcite could adhere to (Kufel and Kufel 2002). The higher buffering capacity of the turbid lakes diminished the influence of metabolism on pH, and therefore on the flux of CO_2. Because of the hard-water nature of these lakes, the influence of metabolism on the flux of CO_2 is weak, and therefore the metabolism (heterotrophy or autotrophy) did not always indicate whether the lake functioned as a carbon source or sink to the atmosphere.

Variation in CO_2 Flux

Over the course of the study, all lakes were a net carbon source to the atmosphere, emitting on average 8 and 11 mmol CO_2 m^{-2} d^{-1} in the clear and turbid regimes, respectively, which was similar to other studies of carbon dioxide emissions from freshwater ecosystems (5.9 to 11.4 mmol CO_2 m^{-2} d^{-1} in Mirror Lake, Cole and Caraco 1998; 21 mmol CO_2 m^{-2} d^{-1} in Arctic lakes, Kling et al. 1992; 9 mmol CO_2 m^{-2} d^{-1} in 37 large Finnish lakes, Rantakari and Kortelainen 2005; 15.1 mmol CO_2 m^{-2} d^{-1} in 0.1 to 1.0 km^2 Finnish lakes, Kortelainen et al. 2006).

Instead of directly measuring the concentration of CO_2 in the air above the lake, the global atmospheric concentration of CO_2 has been used to determine flux as well. Because the atmospheric CO_2 concentration above the lakes I measured was typically higher than the global estimate of atmospheric CO_2, our estimates were approximately 4 mmol CO_2 m^{-2} d^{-1} lower than estimates of flux using the global atmospheric CO_2 concentration. Regardless of the use of global or local atmospheric CO_2 concentration, these lakes would still be carbon dioxide sources overall.

There was considerable temporal variation in the flux of CO_2 throughout each year and between years (Figure 7). From 2006 to 2008, the mean flux for the lakes in the clear regime was 15, 21, and -21 mmol CO_2 m^{-2} d^{-1}, while the turbid regime mean fluxes were -40, 28, and 3 mmol CO_2 m^{-2} d^{-1} in 2006, 2007, and 2008, respectively (Figure 7). Given that the CO_2 flux was mediated by complex interactions between hydrology, chemistry, and biology, the variability in the CO_2 flux between years was somewhat expected, and is common in shallow lake ecosystems (Finlay et al. 2009). The intensively

sampled lakes closely resembled the flux of the other seven lakes that were monitored in 2007 and 2008 (Figure 7), which indicated that mechanisms other than biology, i.e. climate and/or hydrology, may be influencing the exchange of CO_2 to a significant extent. Furthermore, the magnitude and direction of these fluxes did not coincide with wetland regime (Figure 7). I further evaluated the relationships concerning CO_2, DOC, pH, and metabolism in order to identify 1) the factors that best predicted the concentration and flux of CO_2, and 2) whether any differences existed between the two regimes and how those differences related to predicting the flux of CO_2.

DOC, Precipitation, and pCO_2

In contrast to many studies (Kelly et al. 2001, Hanson et al. 2004, Sobek et al. 2003, Sobek et al. 2005, Roehm et al. 2009), I found no significant correlation between pCO_2 and DOC for these wetlands (Figure 8c). This lack of correlation between these variables is not rare, and tends to occur in lakes whose metabolism is dominated by autochthonous production (Rantakari and Kortelainen 2005, Kortelainen et al. 2006), or in systems where pCO_2 is controlled more by pH and not controlled by net metabolism (Finlay et al. 2009). Given the high rate of GPP observed in these lakes, the close association between GPP and R (see Chapter 1), and the fact that the DOC in the surface waters of these lakes was primarily of terrestrial origin (fluorescence index of 1.40, see Chapter 1), it is likely that heterotrophic respiration depended on the consumption of a portion of the total DOC that was more labile, and likely autochthonous DOC (del Giorgio and Peters 1994). This would also explain why the DOC I measured in the

surface waters and R were not correlated (Figure 8a). Still, even if total DOC did boost heterotrophic respiration, the effect would likely not be observed because there was no correlation between respiration and pCO_2 (Figure 8b), which suggests other factors not related to biology (such as chemistry or hydrology) may be more important factors in regulating pCO_2.

Although there was no correlation between DOC and pCO_2, I did find a significant positive correlation between precipitation and pCO_2 (Figure 9c). Given that precipitation had no positive association with DOC or respiration (Figure 9a,b), and the fact that the values of pCO_2 greatly exceed atmospheric pressure of CO_2 (~380 µatm), this association between pCO_2 and more intense storm events is most likely due to direct transport of CO_2 from the surrounding soils that contain high CO_2 due to the mineralization of organic matter. Overall, I found no robust evidence that DOC or precipitation was strongly correlated with respiration.

pCO_2 and pH

I evaluated the relationship between pH and pCO_2, the chemical enhancement factor, and the flux of CO_2 between the lake and the atmosphere. Similar to the hard-water shallow lakes in the boreal region of North America, pCO_2 was strongly associated with pH in wetlands in both the turbid and clear regime (Finlay et al. 2009) (Figure 10a). pCO_2 exponentially declined with increasing pH because as pH increases, the portion of DIC as free CO_2 declines, and is converted to HCO_3^- and CO_3^-. Given the nature of the calculation used to estimate the chemical enhancement factor, it was confirmed that pH

significantly influenced the chemical enhancement factor in both the turbid and clear regimes, as it exponentially increased with increasing pH (Figure 10b). When pCO_2 was high in the surface waters (and pH was relatively low), the rate at which CO_2 exchanged between the lake and the atmosphere was not enhanced. However, when pCO_2 was low and pH exceeded approximately 8 units, the chemical enhancement factor increased, boosting the rate of atmospheric CO_2 invasion into the lake. Similar to previous studies, our results show that high pH lowers pCO_2 in the surface waters and causes the wetland to become a carbon sink in regard to the atmosphere (Finlay et al. 2009) (Figure 10c).

pCO_2, Metabolism, and Trophic Regime

The Prairie Pothole Region was a suitable system to test the connection between lake metabolism and CO_2 because the range of production and respiration rates in the lakes of this region were higher than the range of metabolic rates reported in other studies that assessed the same relationship (in this study GPP and R maxima were 1315 and 1186 mmol O_2 m^{-2} d^{-1} or 1052 and 949 mmol C m^{-2} d^{-1}, respectively, versus GPP and R maxima of 540 and 720 mmol O_2 m^{-2} d^{-1} respectively reported in Cole et al. [2000], and ~400 mmol C m^{-2} d^{-1} for GPP and R reported in Hanson et al. [2004]). Furthermore, this is one of the first studies to evaluate the relationship between CO_2 and whole ecosystem metabolism (not just planktonic metabolism), which is important because the benthic influence in shallow systems can be significant (Hanson et al. 2003, Pace and Prairie 2007, Lauster et al. 2006, Chapter 1). Although the metabolic rates of macrophyte- and phytoplankton-dominated systems can differ (Goulder 1969, Rørslett et al. 1986, Mitchell

1989, Blindow et al. 2006), the wetlands in the clear and turbid regime in this region had similar NEP rates during all three years, suggesting that any biological differences observed in pCO_2 between these two regimes was not likely due to differences in NEP. Even though the NEP rates were similar, GPP was significantly higher in the turbid lakes in 2006 and 2008, and R was significantly higher in the turbid lakes during all three years (see Chapter 1). Given their high metabolic rates and the strong relationship between pH and CO_2, I sought to further evaluate the relationship between metabolism, pH, and CO_2 in these shallow lakes.

In all of the lakes, pH fluctuated throughout the day (Figure 11) and mimicked the change in oxygen, which represents diel changes in metabolism (i.e. the balance between photosynthesis and ecosystem respiration). Based on these observations, it is clear that along with oxygen, the change in pH throughout the day was driven by metabolism. When I directly evaluated the relationship between metabolism and pCO_2, I found a significant correlation between GPP, R, and pCO_2 for both the clear and turbid regimes (Figure 12a,b, d, e), but no correlation between NEP and pCO_2 for either regime occurred (Figure 12c,f). Although the correlations between GPP, R, and pCO_2 were statistically significant, the variation in pCO_2 explained (R^2) by GPP and R only ranged from 4 to 16%. I argue that the direct relationship between metabolism and pCO_2 was diluted due to the hard-water nature of these lakes.

These lakes have amongst the highest DIC concentrations reported for freshwater ecosystems (this study up to 129 mg DIC L^{-1}; total inorganic carbon mean of 2.3 mg L^{-1} for 177 Finnish lakes, Kortelainen et al. 2006; Arctic lakes mean of 15.8 mg DIC L^{-1},

Kling et al. 1992; up to 60.4 mg DIC L^{-1} in hard-water lakes of Saskatchewan, Finlay et al. 2009). Still, only 3% (on average) of the total DIC content in these lakes resided in the free CO_2 form, so it would be difficult for the balanced relationship between GPP and R (or any other factor influencing CO_2) to strongly mediate changes in CO_2. Although the diel fluctuation of pH was due to changes in the balance between GPP and R, the pH rarely dropped below 8 because of the high buffering capacity of these lakes, which meant that little free CO_2 was generated. What was likely driving these significant relationships between GPP, R, and pCO_2 were the occurrences in which the balance between GPP and R happened to push pH below 8 units, which therefore supported a correlation with pCO_2.

Although the buffering capacity of the water column, or alkalinity, was relatively high in both regimes compared to other lake ecosystems (Stets et al. 2009), alkalinity significantly differed between the two regimes. Collectively, the turbid lakes were more alkaline than the clear lakes (year 2007: turbid = 7.1±2.6 meq L^{-1}; clear = 4.2±1.4 meq L^{-1}; $p = 0.060$). While it was possible that the lakes in the turbid regime naturally existed in a more alkaline watershed producing a naturally higher buffering capacity than the lakes in the clear regime, this did not seem the case when I evaluated the seasonal loss of alkalinity. On average, the macrophyte-dominated lakes lost approximately 1.05 meq L^{-1} of alkalinity from May through August, while the lakes in the turbid regime only lost about 0.04 meq L^{-1} (Figure 13). This difference in loss of alkalinity suggests the biology of the regimes was important, because the macrophytes actively changed the water column alkalinity to a greater degree than the phytoplankton.

Higher levels of alkalinity in phytoplankton-dominated shallow lakes versus clear lakes (specifically *Chara*-dominated) can be attributed to a greater efficiency of bicarbonate uptake (hence loss of water column alkalinity) by charophytes versus the phytoplankton (Nõges et al. 2003). Macrophytes such *Potamogeton* species and *Charophytes* are known to enzymatically induce calcification (which precipitates carbonates) in order to gain electrons needed for bicarbonate uptake (i.e. loss of alkalinity) to support photosynthesis (McConnaughey 1998). This divergence in alkalinity between regimes also explains why the clear regimes show a wider range of pH (Figure 10), and why the turbid lakes have diminished daily fluctuations in pH (Figure 11). I estimated the variation around the mean pH for each day in each lake, and found that on average, the fluctuations in pH dampened with greater buffering capacity (i.e. alkalinity) (Figure 14). Because the lakes in the clear regime were less alkaline, pCO_2 should be more readily influenced by changes in metabolism due to less buffering of changes in pH. Furthermore, the lakes in the clear regimes should be more susceptible to atmospheric changes in CO_2, where elevated atmospheric CO_2 levels may cause the clear lakes to be a greater C sink, as CO_2 could flux into the lakes at a faster rate. Although the lake regimes did not influence pCO_2 based on their overall NEP, these regimes indirectly influenced pCO_2 through their differences in ability to moderate changes in pH.

Ecosystem Metabolism and CO_2 Flux

Many studies on lakes around the world have shown or assumed that metabolism closely reflects CO_2 flux, as heterotrophic and autotrophic lakes function as carbon

sources and sinks to the atmosphere, respectively (Duarte and Agustí 1998, Cole et al. 2000, Duarte and Prairie 2005, Pace and Prairie 2007). Our results were similar to these observations (Figure 15); however, because of the hard-water nature of these shallow lakes, the metabolic influence on pH and consequently on the flux of CO_2 was weakened. As a result, metabolism alone (i.e. net heterotrophy or autotrophy) did not consistently predict whether the lake was actually functioning as a carbon sink or source to the atmosphere. I have included a conceptual model that depicts the differences in hard and soft water lakes in predicting whether a lake is a carbon source or sink based on their metabolism (Figure 16). It is likely that the variation in CO_2 flux in these hard-water ecosystems may be more strongly influenced by hydrology and geology than any other factor due to its influence on the DIC concentration of these lakes. In comparison to larger freshwater ecosystems, these shallow lakes have a relatively small volume and therefore a short residence time, which naturally leads to a greater influence of groundwater input. The lakes in this study are situated in a highly calcareous region (Gorham et al. 1983), and the high DIC concentration of these lakes suggests that they receive substantial groundwater input from carbonate-rich sediments (Stets et al. 2009). Therefore, it is not surprising that metabolism or DOC only weakly influenced pCO_2. In addition to metabolism, hydrologic inputs from the watershed, CO_2 evolution from calcite formation, and bicarbonate uptake for photosynthesis likely influence pCO_2 (Stets et al. 2009). More work is required to measure the hydrologic inputs of DIC and CO_2 to these shallow lakes to estimate their influence on the flux of CO_2. Due to the hard-water nature of these lakes where pH is often above 8 units, the weakened effect of metabolism

on pH, and the potentially large hydrological input of CO_2, these shallow, hard-water lakes can function as net carbon sources to the atmosphere even when they are net autotrophic, and carbon sinks when they are net heterotrophic (Figure 15).

Finally, it is interesting to note that the average CO_2 flux from these lakes resembles the flux from other lake ecosystems around the world (Cole and Caraco 1998, Kling et al. 1992, Rantakari and Kortelainen 2005, Kortelainen et al. 2006), yet the production rates greatly exceed many other lake ecosystems (Cole et al. 2000, Hanson et al. 2004). I found that the average seasonal GPP and R for all the lakes was approximately 543 and -543 g C m^{-2} yr^{-1}, respectively (measured over most of the open water season, or 175 d). If I assume the mean efflux rate of 10 mmol m^{-2} d^{-1} applied to the same time period, approximately 20 g C m^{-2} yr^{-1} was emitted to the atmosphere. If these lakes permanently accumulate 50-100 g C m^{-2} yr^{-1} in the sediment (Cotner et al. *unpublished data*), more than 70-120 g C m^{-2} yr^{-1} should come from the land to offset these C losses from the ecosystem (Figure 17). Furthermore, it is likely that the estimated C emission rate was underestimated because it did not account for early spring or late fall emissions. Therefore, the input from the land to account for these additional losses would have to be even greater than 70-120 C m^{-2} yr^{-1}. Other similar lake ecosystems have shown hydrological inputs to equal C efflux to the atmosphere (Stets et al. 2009), but our estimates are exceptionally large in comparison, as approximately 3.5 to 6 times as great as the carbon emission from the lake to the atmosphere, and is comparable to the C burial rates in the sediment. These results highlight the importance of the connection between the land and the lake in terms of carbon cycling.

Table 2. Summary of water chemistry and metabolism for all nine lakes studied from 2006 to 2008. The intensively monitored lakes are marked with an asterisk (*). The state "C" means clear lakes, and "T" signifies turbid lakes. Area is in km^2. z_{max} is the maximum depth in meters recorded for each lake. Chl is reported in $\mu g\ L^{-1}$ and DOC and DIC is in $mg\ L^{-1}$; all are seasonal means. GPP, R and NEP are seasonal average values for each lake and are presented in mmol $O_2\ m^{-2}\ d^{-1}$ ± standard deviation.

Lake	State	Area	Year	z_{max}	Chl	DOC	DIC	GPP	R	NEP
8Mile	C	0.26	2007	1.5	17	12.6	25.2	385 ± 164	-384 ± 187	1 ± 84
Lake Christina	C	20	2007	1.1	10	8.4	43.2	307 ± 120	-230 ± 115	76 ± 119
Lee	C	0.2	2007	4.5	34	12.9	62.3	285 ± 140	-294 ± 127	-9 ± 74
Mavis East*	C	0.22	2006	3.3	12	14.3	72.8	344 ± 158	-299 ± 152	45 ± 107
Mavis East*	C	0.22	2007	3.3	14	14.0	55.2	205 ± 119	-221 ± 101	-16 ± 97
Mavis East*	C	0.22	2008	3.3	13	12.7	44.5	143 ± 76	-189 ± 68	-47 ± 85
Pisa	C	0.07	2007	1.1	17	19.5	48.0	395 ± 191	-385 ± 154	10 ± 128
Pisa	C	0.07	2008	1.1	2	18.2	39.2	246 ± 101	-235 ± 123	11 ± 78
Bore	T	0.07	2007	1.5	166	23.3	46.8	377 ± 189	-404 ± 188	-27 ± 91
Bore	T	0.07	2008	1.5	207	26.4	40.2	214 ± 242	-394 ± 258	-180 ± 170
Mavis West*	T	0.14	2006	3.6	68	14.8	68.9	800 ± 243	-736 ± 173	65 ± 202
Mavis West*	T	0.14	2007	3.6	77	14.4	61.2	310 ± 135	-361 ± 117	-51 ± 92
Mavis West*	T	0.14	2008	3.6	76	14.3	47.6	449 ± 286	-426 ± 279	-34 ± 171
Morrison	T	0.15	2007	2.2	68	16.2	74.2	271 ± 111	-201 ± 141	70 ± 89
Murk	T	0.15	2007	2.2	48	19.5	129.2	168 ± 114	-139 ± 128	29 ± 88

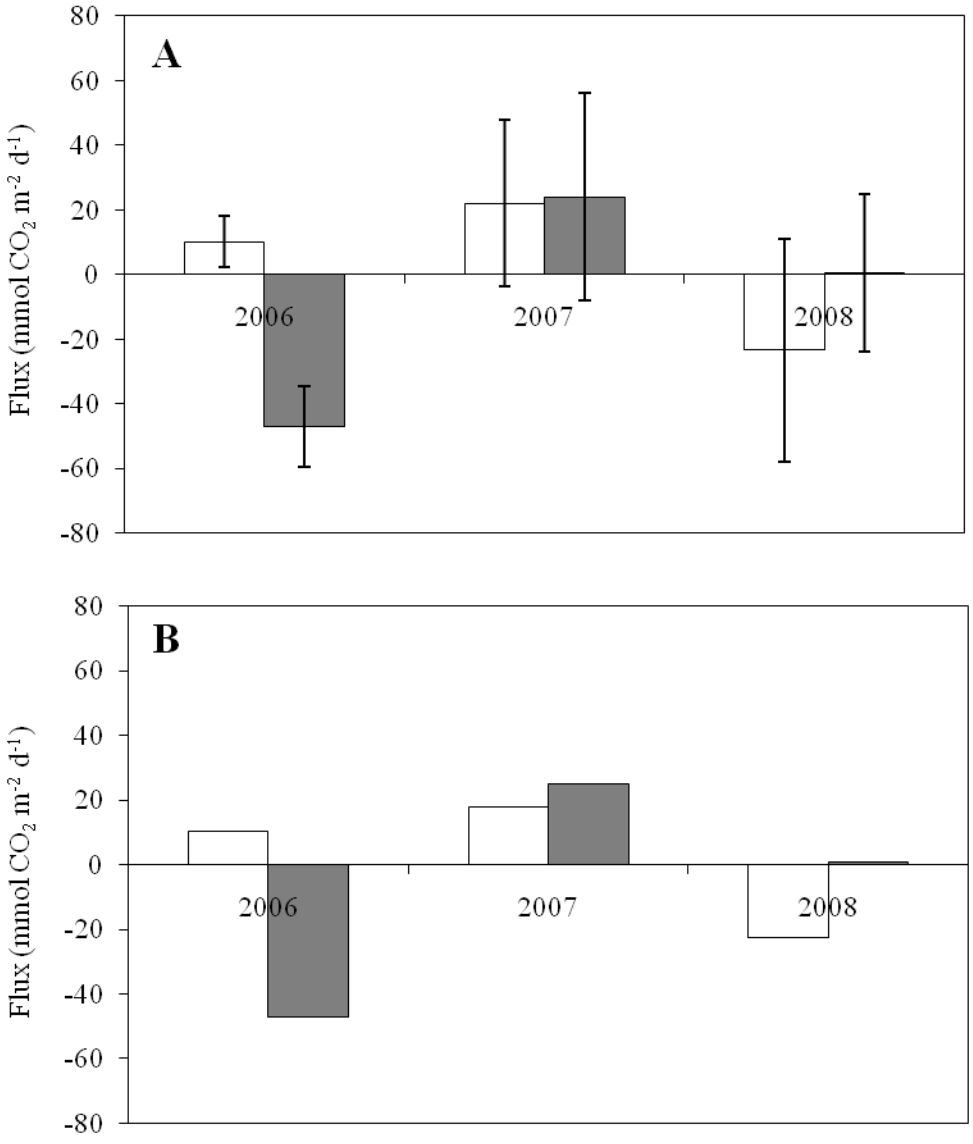

Figure 7. Atmospheric exchange of CO_2 between the lake and the atmosphere for A) the intensively sampled lakes, and B) all lakes (one clear and one turbid during 2006, five clear and four turbid during 2007, and two clear and two turbid during 2008). Positive values indicate evasion from the lake, and negative values indicate invasion into the lake. White bars represent clear regimes, grey bars represent turbid regimes. No standard deviation bars are used in B due to the propagation of error.

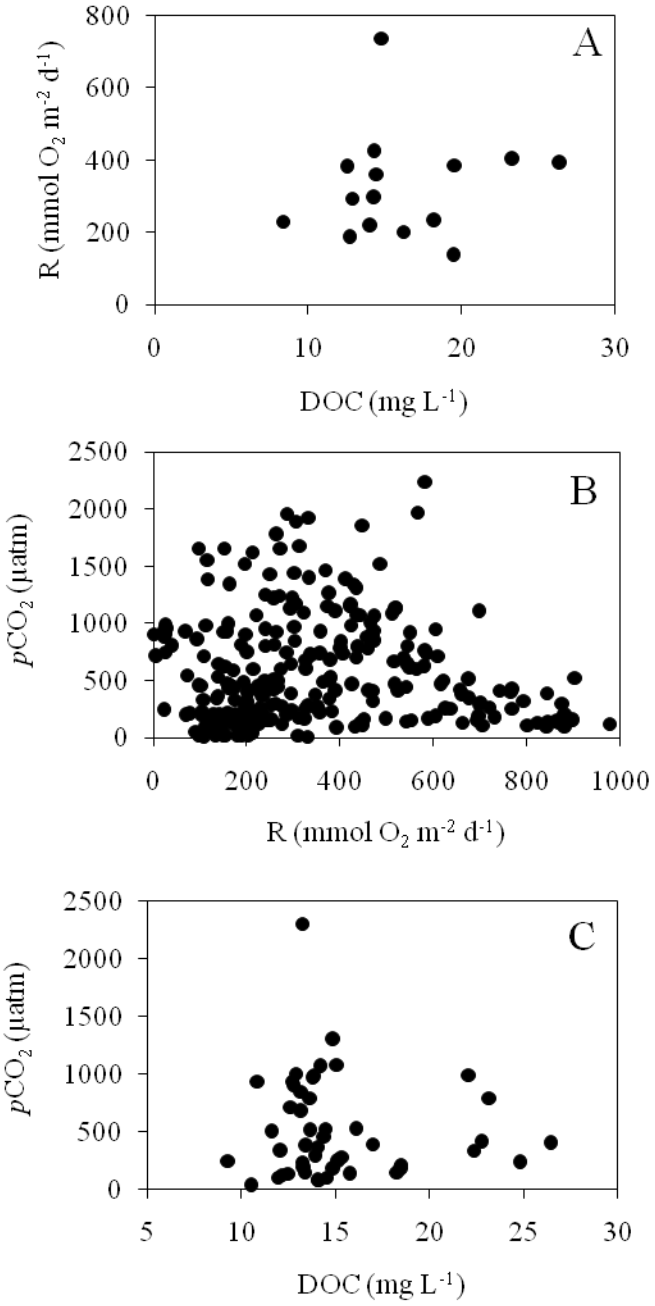

Figure 8. The relationships between dissolved organic carbon (DOC), partial pressure of CO_2 (pCO_2) and respiration (R). The points represent data from both the clear and turbid regimes. A) Annual means of DOC and R for each lake during the three year study period; B) daily estimates of pCO_2 and R; and C) weekly means of pCO_2 and DOC. All correlations were not significant (all $p > 0.05$).

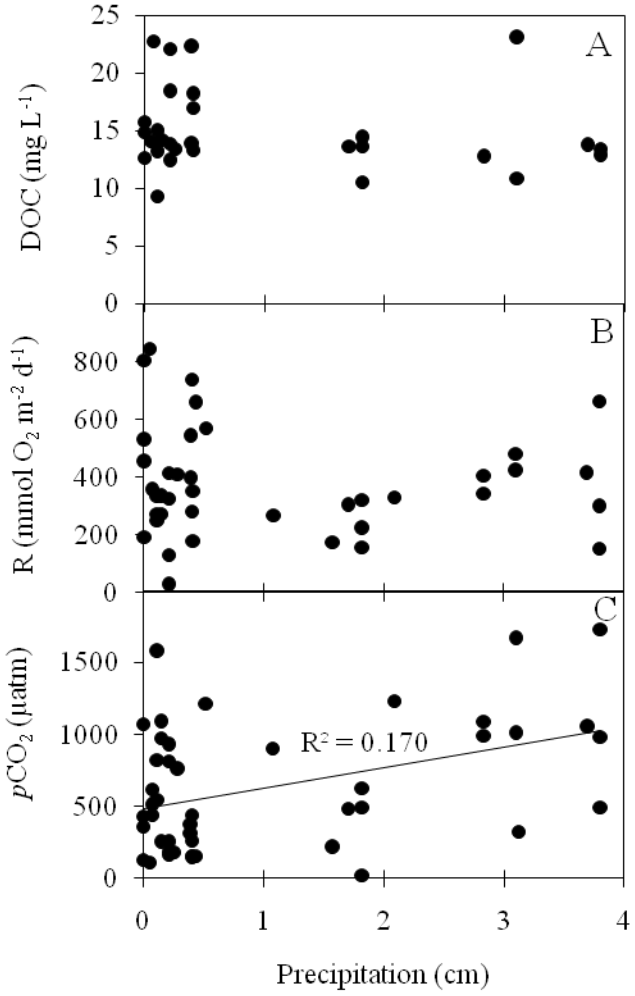

Figure 9. Relationships between the weekly means of dissolved organic carbon, respiration, pCO_2, and precipitation. The points represent data from both the clear and turbid regimes. Significant relationships are indicated by the presence of a trendline, with the associating statistical information.

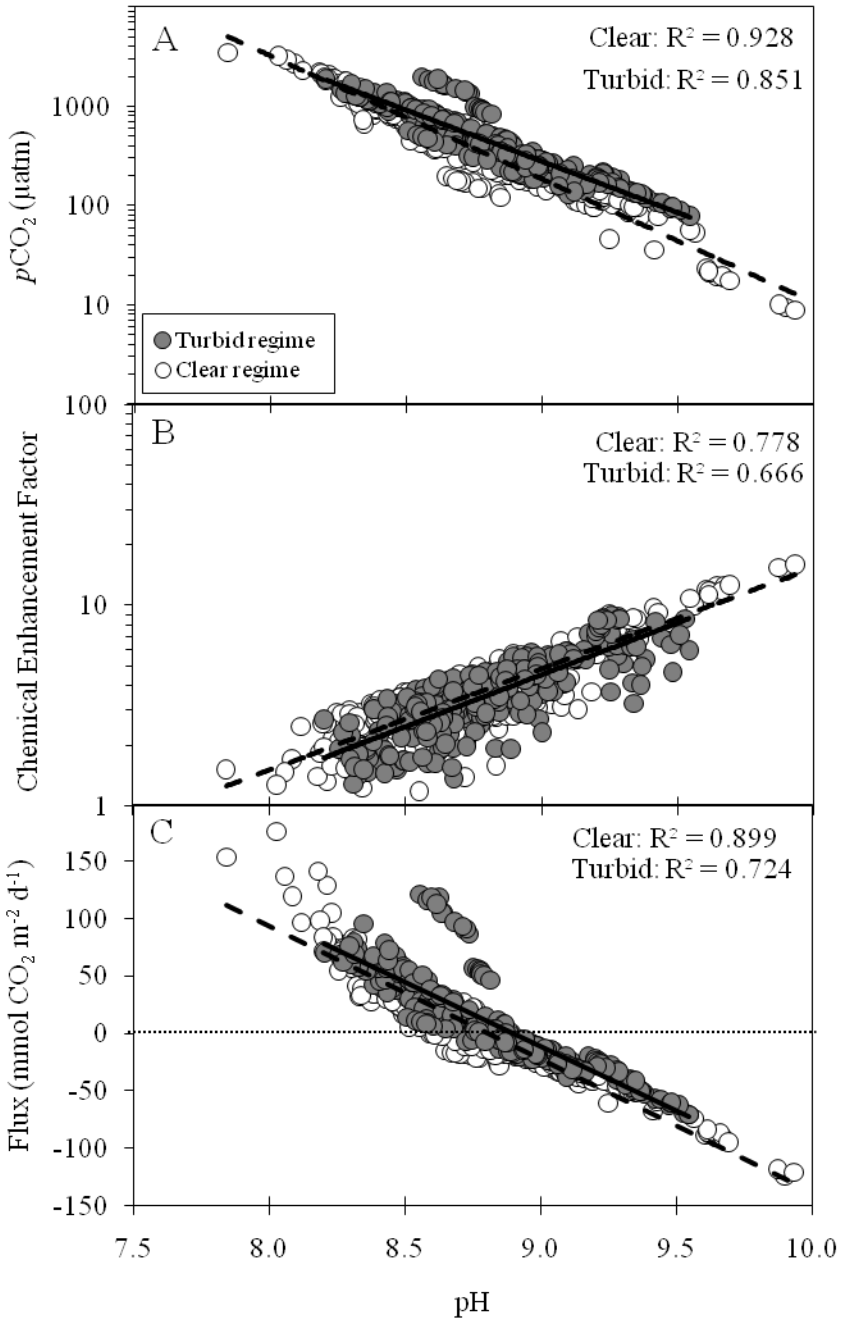

Figure 10. A) pCO_2, B) chemical enhancement factor, and C) flux of CO_2 versus pH. White circles represent the clear regime, and the grey circles represent the turbid regime. Please note the logarithmic scale for A and B. For the flux, positive values indicate evasion from the lake, and negative values indicate invasion into the lake. Significant relationships are indicated by the presence of a trendline (dashed is clear, solid is turbid) with the associating statistical information. All correlations have a p-value <0.0001.

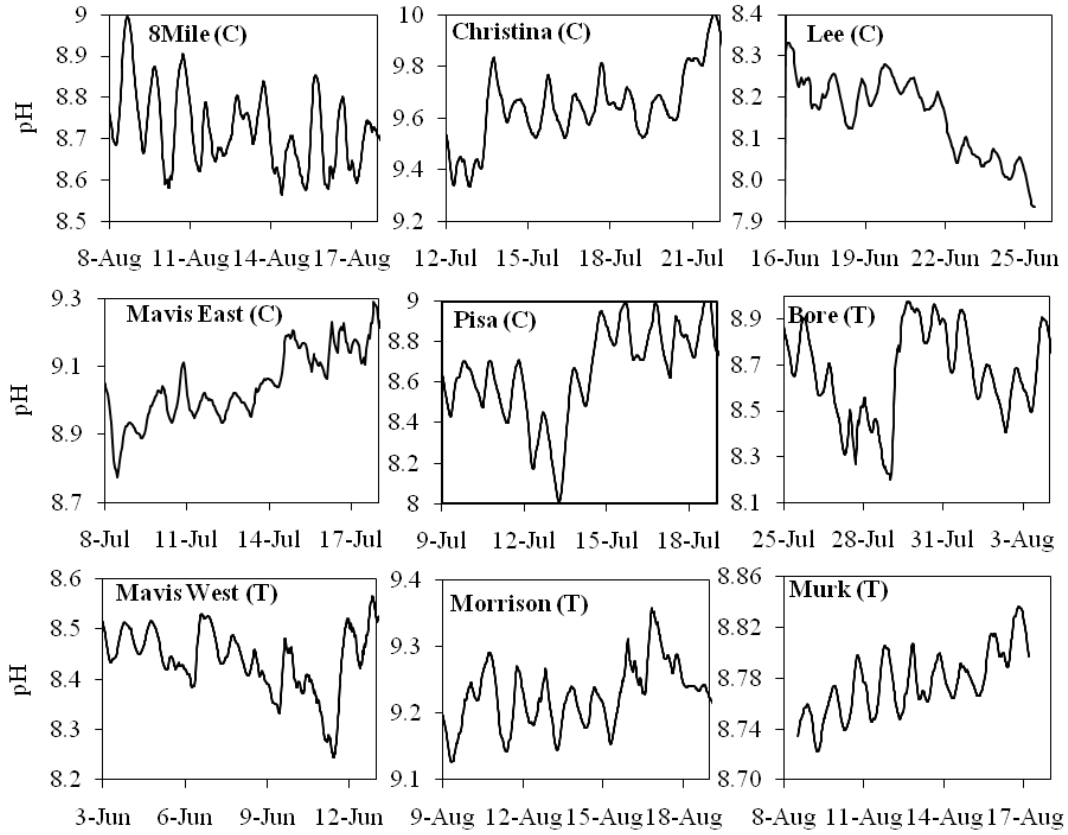

Figure 11. Time series data demonstrating that diel metabolism influences pH in all nine lakes because diel cycles (very similar to oxygen) were observed. Turbid lakes are marked with a (T) after the lake names, and clear lakes are marked with a (C). Please note the scaling for pH of each lake is different; individual scales are presented to portray the diel cycles, no matter how small the fluctuation in pH.

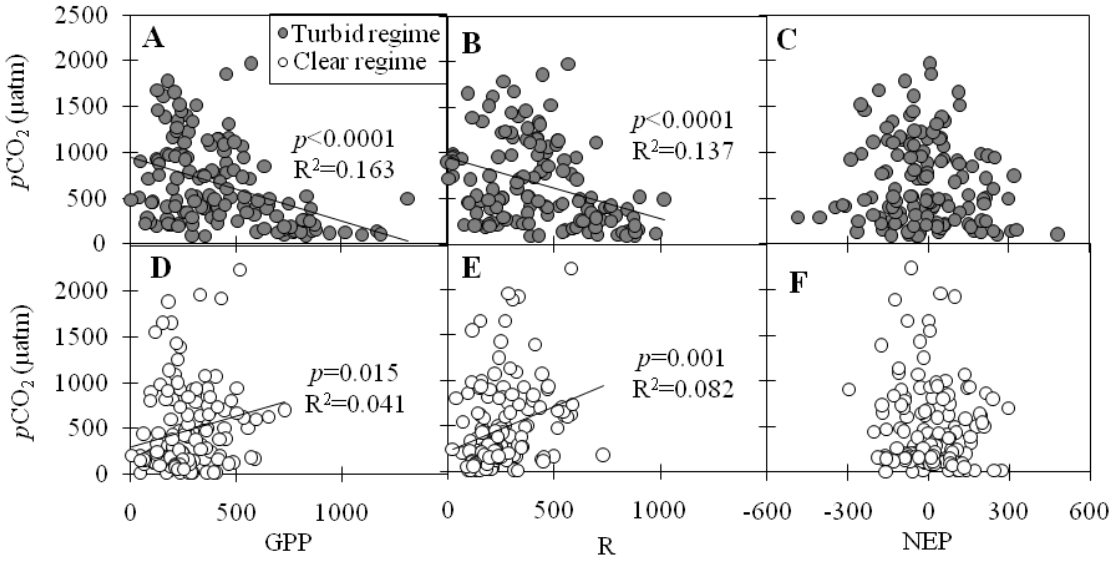

Figure 12. pCO_2 (µatm) versus gross primary production (GPP), respiration (R), and net ecosystem production (NEP) (GPP, R, NEP mmol O_2 m^{-2} d^{-1}). Data points are daily estimates represented from May through August for all three years from all lakes. The grey and white circles represent the lakes in the turbid- and clear-water regime (A-C, D-F), respectively.

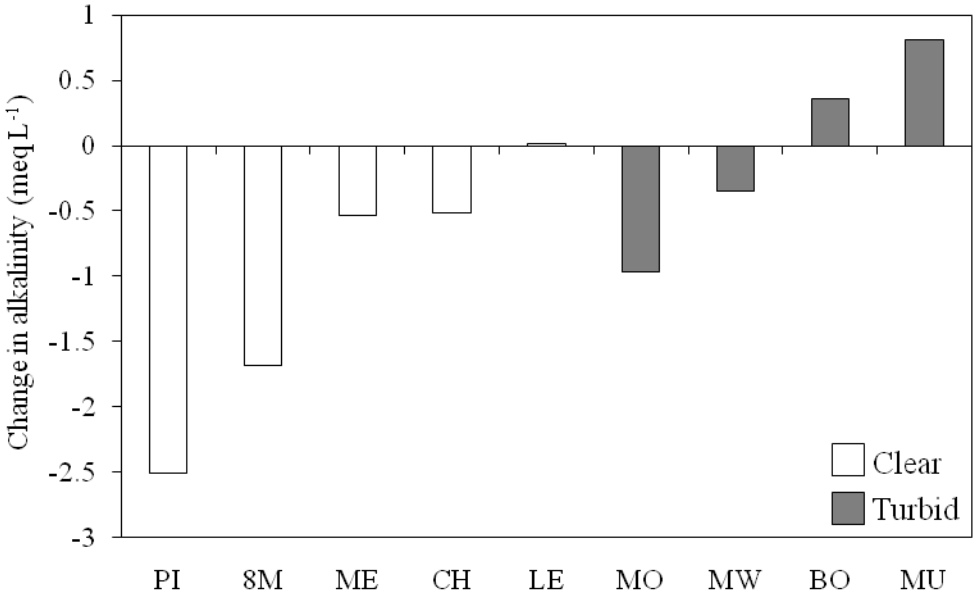

Figure 13. Change in alkalinity for each lake from May to August 2007. A negative change represents loss of alkalinity, while a positive change indicates a gain in alkalinity. The white and grey bars represent lakes in the clear and turbid regimes, respectively. Acronyms for the lakes: PI=Pisa, 8M=8Mile, ME=Mavis East, CH=Christina, LE=Lee, MW=Mavis West, MO=Morrison, BO=Bore, MU=Murk.

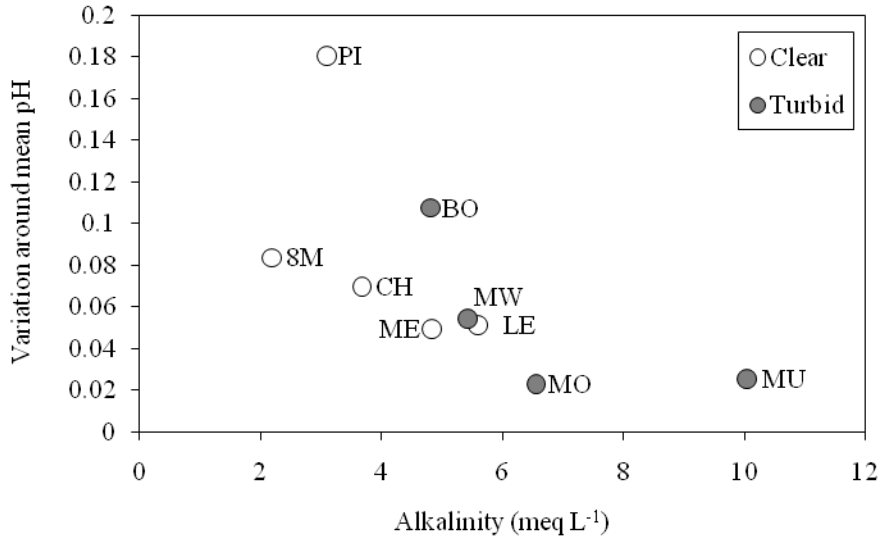

Figure 14. The variation around the mean pH versus alkalinity. The variation was determined as the annual mean of the daily standard deviation values around the mean pH for each lake. The higher alkalinity values indicate a greater buffering of the change in pH, and thus a weaker relationship between metabolism and CO_2.

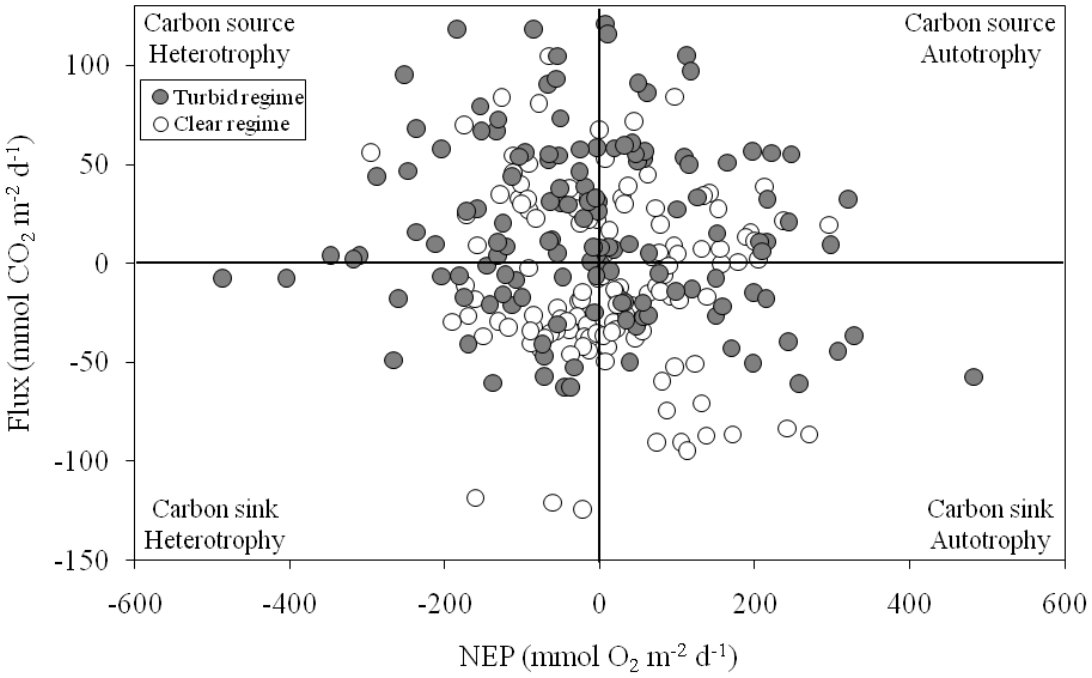

Figure 15. Daily estimates of the exchange of CO_2 between the lake and the atmosphere versus net ecosystem production (NEP). In terms of flux, positive flux values indicate a carbon source or evasion from the lake to the atmosphere, and negative values indicate a carbon sink or invasion from the atmosphere to the lake. Positive NEP (GPP > R) indicate net autotrophy, and negative NEP (R > GPP) indicate net heterotrophy. White circles represent the clear regime, and the grey circles represent the turbid regime.

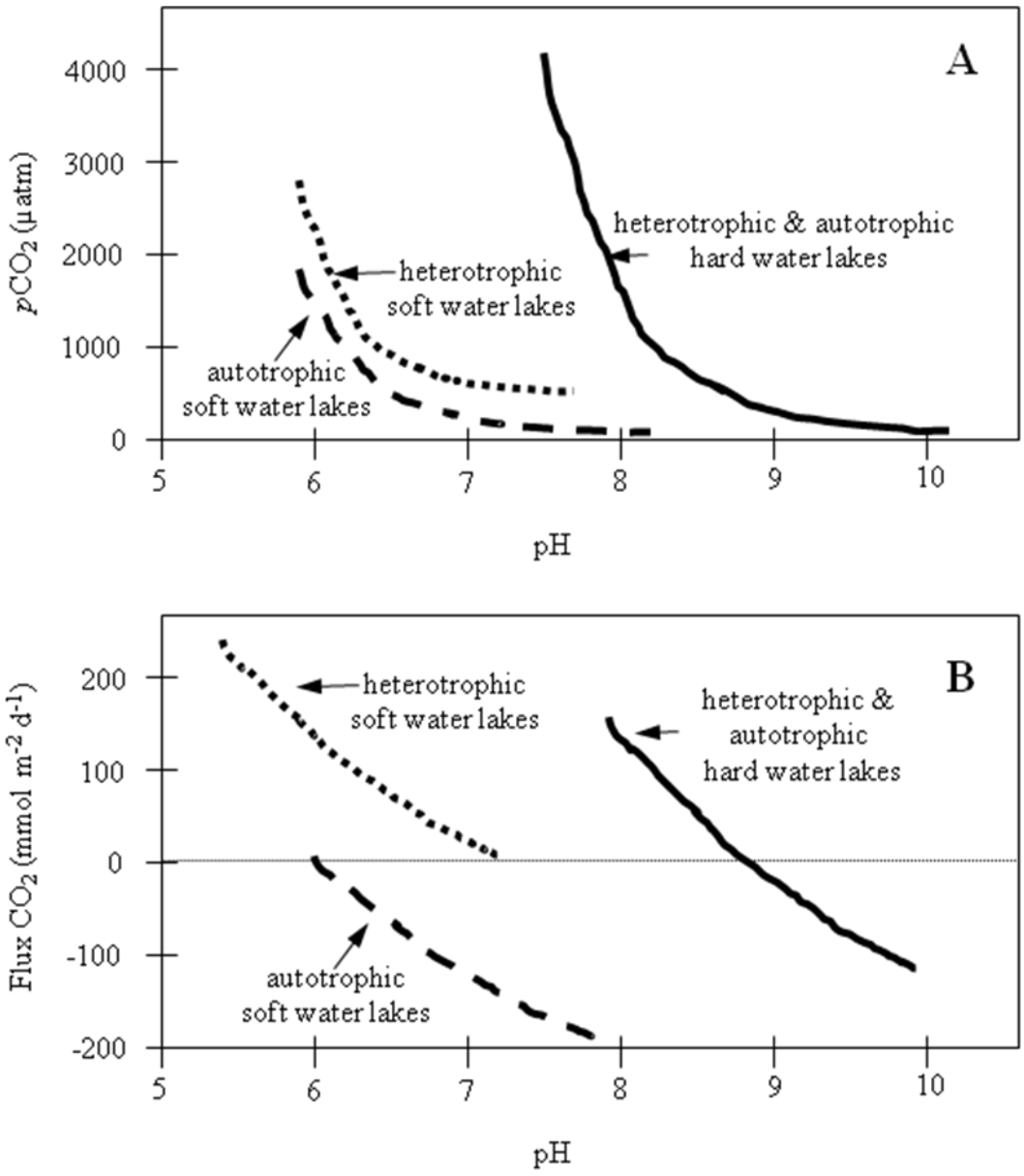

Figure 16. A conceptual model of the relationship between pH and A) pCO_2 and B) flux of CO_2 for autotrophic and heterotrophic hard and soft water lakes. In contrast to hard water lakes, changes in pH due to changes in metabolism should predict whether a lake is a carbon source or sink in regard to the atmosphere in soft water lakes.

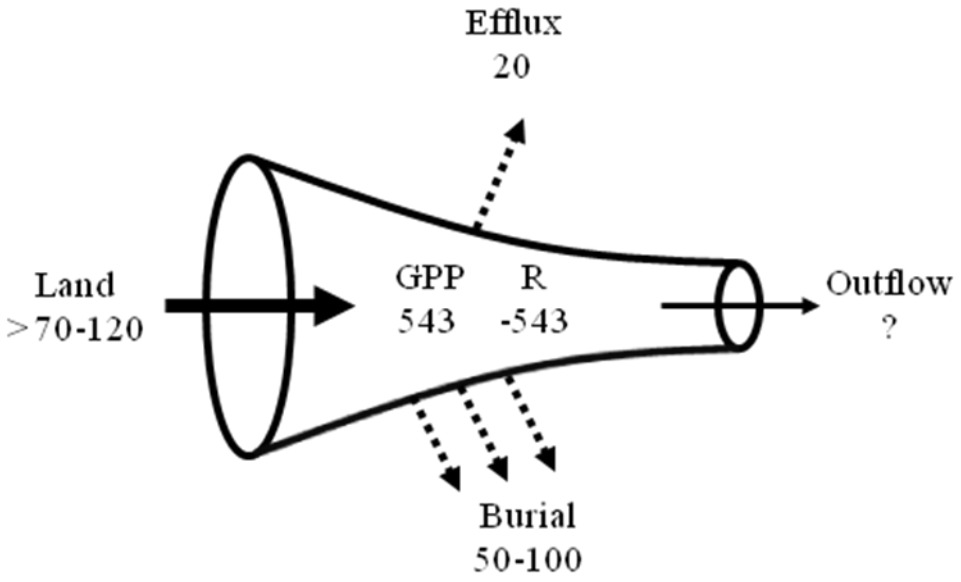

Figure 17. General carbon cycle for shallow lakes in the Prairie Pothole Region. All numbers are presented as a rate g C m^{-2} yr^{-1}. Efflux does not account for C emission during early spring or late fall (which are potentially high), and metabolism (GPP and R) only represents the open water season. If metabolism is relatively balanced, and if the outflow is relatively low, carbon inputs from the land should be larger than 70-120 g C m^{-2} yr^{-1} in order to balance the losses via permanent burial in the sediments and C emission from the lake to the atmosphere.

Chapter 3: Decomposition of macrophytes and algae under aerobic and anaerobic conditions: implications for carbon burial in prairie pothole lakes of alternative regimes

Investigation of the mechanisms that influence carbon burial in small, freshwater ecosystems is important to develop a complete picture of the terrestrial carbon cycle. The amount of carbon accumulated in lake sediments over time is strongly influenced by the quality of organic matter source, and the environmental conditions in which the organic matter is deposited. The Prairie Pothole Region of North America contains approximately 207,000 km^2 of shallow prairie wetlands that can differ in the type of dominant primary producer and the seasonal environmental conditions within these systems. Shallow lakes in this region typically exist in two regimes which are dominated by differing primary producers: 1) macrophytes and macroalgae that have complex compounds such as lignin and cellulose and decompose in the fall under cold temperatures and potentially low oxygen content, or 2) phytoplankton which contain less complex compounds, and decompose throughout the year in a relatively more oxygenated environment. I decomposed two macrophyte species (*Myriophyllum sibiricum* and *Potamogeton pectinatus*), two macroalga species (*Charophyte* and *Cladophora*), and a mixed assemblage of phytoplankton separately under aerobic and anaerobic conditions for one year. Over the course of the experiment, the algae lost 86% of their original carbon content while the macrophytes lost 77% of theirs. Additionally, the primary producers that were decomposed aerobically lost 6% more C than those under anaerobic conditions.

A higher accumulation of dissolved organic matter in the anaerobic treatments likely contributed to a higher abundance and production of bacterioplankton. Although both primary producer type and presence of oxygen played a significant role in the decomposition rate, the decay rates depended slightly more on the type of primary producer than the presence of oxygen. When the decay rates of the clear water species (*Charophyte*, *Potamogeton pectinatus*, *Myriophyllum sibiricum*) and turbid water species (phytoplankton) were applied to their respective annual carbon fixation rates of the clear and turbid water regimes, it was estimated that the lakes in the clear regime should retain almost five times as much carbon as the lakes in the turbid regime on an annual basis. Given that the macrophyte-anaerobic treatments retained the most carbon, and macrophyte-dominated shallow lakes tend to become anaerobic rather quickly after their senescence, the macrophyte-dominated regime should experience higher rates of carbon burial relative to a phytoplankton-dominated regime.

Introduction

Freshwater lakes play an important role in global carbon cycling (Cole et al. 2007; Tranvik et al. 2009) due to their productive nature and reception of carbon inputs from their terrestrial surroundings. Most lakes of the world are small (Downing et al. 2006) and accumulate organic carbon at high rates in comparison to other ecosystems (Schlesinger 1997, Dean and Gorham 1998). The rate at which a lake permanently accumulates organic carbon over time is a function of the amount and quality of carbon transferred to the sediments, and the environment in which it is deposited (Rejmánková

and Houdková 2006). Decomposition occurring in the lake sediments reduces the amount of material available for permanent storage, and reduces carbon burial most in warm, oxygenated waters with nutrient rich, labile carbon available as a substrate (Dauwe et al. 2001, Müller et al. 2006, Sobek et al. 2009, Gudasz et al. 2010). Among freshwater ecosystems, more variation in the burial rate in small ecosystems in comparison to larger lakes (Cotner *unpublished data*, Figure 18). Because these lakes play an integral part in global carbon cycling, the investigation of the mechanisms influencing the rate of decomposition (and hence carbon burial) in small lake ecosystems is required in order to identify the source of variability in their carbon burial rate.

The Prairie Pothole Region (PPR) of North America contains 207,000 km^2 of shallow lakes which are extremely productive (Euliss et al. 2006; Chapter 1), and their characteristics may help explain the wide variation in carbon burial in small freshwater ecosystems. Shallow lakes typically exist in either a clear-water, macrophyte/macroalgae dominated regime, or a turbid-water, phytoplankton/microalgae dominated regime (Scheffer et al. 1993, Zimmer et al. 2001). Macrophytes contain complex compounds such as lignin and cellulose, which makes decomposition of this organic matter slower than decomposition of phytoplankton, which do not contain these more recalcitrant compounds (Enríquez et al. 1993). Furthermore, the decomposition of the macrophyte material may occur in hypoxic or anoxic environments, as oxygen consumption in the water column quickly increases due to the decay of large deposition of plant biomass after they senesce in late summer/early fall (Goldshalk and Wetzel 1978, Golterman 1995), and the mineralization of the macrophyte leachate by heterotrophic bacteria

(Carpenter and Lodge 1986). Oxygen plays an important part in decomposition, since more reduced, low oxygen environments slow the efficiency in which organic carbon is broken down (Godshalk and Wetzel 1978a). In contrast to macrophytes, decomposition of phytoplankton is typically faster in comparison, given that phytoplankton lack many of the complex compounds common in macrophytes (Enríquez et al. 1993, Cebrian 1999). Furthermore, the fast decrease in oxygen that macrophyte-dominated systems can experience may not occur in turbid regimes, as the organic carbon deposited from senescent phytoplankton occurs year-round, rather than over a relatively short period of time. Decomposition experiments are often focused on either macrophytes (one or multiple) or algae (Otsuki and Wetzel 1974, Cole and Likens 1979, Belova 1993, Denward and Tranvik 1998, Sala and Güde 1999, Asaeda et al. 2000), but rarely contrast both (Enríquez et al. 1993, Duarte and Cebrián 1996), especially in freshwater ecosystems. Furthermore, decomposition experiments of primary producers in light of clear and turbid regimes and their role in carbon burial are essentially lacking.

Both the trophic state of the lake and the amount of oxygen reaching the sediments can play an important role in the cycling of organic carbon in shallow lake ecosystems (Müller et al. 2006). Given the greater complexity of macrophytes in comparison to phytoplankton, and a longer period of time under low oxygen conditions in which the macrophytes/macroalgae decompose, it is likely that the lakes in the clear-water regime bury carbon at a much higher rate than the turbid, phytoplankton-dominated lakes. Therefore, the combination of the dominant primary producer and level of oxygenation during its decomposition may explain the high variation in carbon burial

observed in small, shallow lakes. I analyzed the decomposition rates of macrophytes and algae commonly found in lakes in the Prairie Pothole Region under aerobic and anaerobic conditions in order to assess the influence of organic matter quality and the physical environment on decomposition rates. I hypothesized that macrophytes decompose more slowly and hence favor higher carbon burial rates in clear-water regimes. Furthermore, anaerobic conditions should slow decomposition rates, also favoring higher carbon burial rates. Therefore, the macrophyte-dominated lakes that experience prolonged anoxia should accumulate the most carbon in the sediments over time.

Methods

A laboratory experiment was set up to monitor the decomposition process of two macrophyte species, two macroalga species, and a mixed assemblage of phytoplankton under aerobic and anaerobic conditions (hereafter AE and AN, respectively) over the course of one year. The treatments were referred to as MY (*Myriophyllum sibiricum*, American Milfoil), PO (*Potamogeton pectinatus*, sago pondweed), CH (*Charophyte*), CL (*Cladophora*, filamentous algae), and PH (mixed assemblage of phytoplankton). In August 2008, live aquatic plants, algae, and lake water were collected from four shallow lakes in the Prairie Pothole Region near Fergus Falls, Elbow Lake, and Morris, MN.

Experimental Setup

Duplicate replicates for the MY, PO, CH and CL treatments were set up to be harvested after one month, six months, and twelve months of decomposition, and were

incubated separately under AE and AN conditions (total of 12 experimental units per macrophyte/macroalgae treatment; six AE and six AN replicates per macrophyte/algae). Jars containing PH were only decomposed for a total of six months, and therefore had a total of eight experimental units. To administer the oxygen treatments, different types of jars were used to hold the AE and AN replicates, which subsequently caused a difference in jar volume (2.6 and 1.9 L in the AE and AN treatments, respectively). Nevertheless, the macrophyte and algal wet weight added to each jar equaled approximately 1% of the volume (i.e., 19 g of biomass in a 1.9 L jar) for both types of jars, and the wet:dry ratios for each macrophyte and macroalga were calculated in order to estimate the dry weight of the wet material added to each replicate. The PH treatments were made by filling replicate jars with whole-lake water containing phytoplankton (excluding zooplankton by filtering through 80 µm mesh) from a turbid-water regime. For the AE treatments, the jars were filled with the specific lake water in which the plants and algae were obtained, which was filtered to exclude only zooplankton.

The AE jars were capped with a rubber stopper containing glass tubing that allowed constant aeration of the treatment (Figure 19). For the AN treatments, the lake water was bubbled with N_2 gas to remove oxygen from the water, and once the lake water was added to the jar, it was capped with a rubber stopper containing two glass tubes of differing lengths (Figure 19). Immediately after capping the jars N_2 gas was bubbled (at least three times the volume of the jar) through the longer tube to remove any oxygen in the water and the headspace (air was permitted to leave out the short glass tube connected

to the headspace). After bubbling the N_2 gas through the jar, the valves were immediately closed to prevent any gas exchange between the jar and the atmosphere (Figure 19).

All replicates were kept in an incubator in the dark to control for temperature and to negate photosynthesis. In order to simulate realistic cooling and warming temperatures the senescent plants and algae would experience on an annual cycle, the incubation temperature was changed according to changes of *in situ* temperature in these same prairie lakes continuously recorded from the prior year.

Sampling Procedures

To understand the physical, chemical, and biological dynamics of the water column throughout the experiment, dissolved oxygen (DO), pH, soluble reactive phosphorus (SRP), dissolved organic carbon (DOC), dissolved inorganic carbon (DIC), particulate carbon (PC), methane (CH_4), and bacterial production (i.e. ^{3}H-leucine uptake) in the water were measured at 1 d, 2 wk, 1, 3, 6, 9, and 12 mo increments. The 2 wk samples were taken from the jars fully harvested at 1 mo, the 3 mo sampling from the 6 mo jars, and the 9 mo samples from the 12 mo jars. At 1, 6, and 12 months the remaining macrophyte and macroalgal material were sampled for dry weight and carbon and nitrogen content.

Here I describe the process and order in which the samples were taken from the replicates, and after the actual analyses of these samples are provided in detail. Sampling of all replicates was completed in the dark, and took approximately 10 minutes for each jar to ensure minimal change in temperature at the time of sampling. In order to sample

the AE replicates, the rubber stopper was removed, and approximately 60 mL of the surface water was immediately taken out via an acid washed 60 mL syringe to estimate methane and oxygen. Next, samples to measure dissolved oxygen were collected by overfilling an ashed 7 ml scintillation vial with the sample water, which was immediately fixed by adding 60 µl of 1% mercuric chloride to halt any biological activity, and the vial was also capped void of headspace to preserve the concentration of dissolved oxygen. After the gas samples were taken, a total of 8.7 mL of the water from the jar was transferred to microcentrifuge tubes using an Eppendorf pipette, where they were subsequently analyzed for bacterial production and abundance (six tubes containing 1.2 mL for bacterial production, and one tube with 1.5 mL for abundance). The microcentrifuge tube with 1.5 mL was fixed with 150 µL of 37% formalin to analyze bacterial abundance. A Beckman benchtop pH probe was used to measure the pH of the water within the jar. Finally, approximately 200 mL from the jar was collected for DOC, DIC, SRP, and PC analysis. To collect the water samples from the AN replicates, N_2 gas was slowly forced in through the short glass tube in the stopper as I simultaneously opened the valve of the long glass tube, which caused the water inside the jar to be forced out. Using this method approximately 300 mL of water inside the jar was collected into a 500 mL Nalgene bottle. From this collected sample the same series of steps for analysis of the AE replicates were followed.

In order to replace the water taken out for the physical and chemical analyses at the two week, 3 month, and 9 month sampling intervals (and maintain the same volume:primary producer material), approximately 20 L of the original lake water used in

the incubations was filtered through a 0.2 µm membrane filter at the beginning of the experiment (20 L from one lake for the PH treatment, 20 L from one lake for the CH treatment, 20 L from one lake for the CL treatment, and 40 L from one lake for the MY and PO treatments). This filtered water was stored in 20 L carboys and placed in the same incubator as the jars, loosely capped. At the 2 wk, 3 mo, and 9 mo sampling intervals, the appropriate 0.2 µm filtered water was added to the jars to replace the water taken for physical and chemical analyses. The water to fill the AN jars was bubbled with N_2 at least three times the jar volume before being transferred into the jars. The 0.2 µm filtered water was sampled for the same suite of measurements described above, and was accounted for when estimating change in carbon content.

Sample Analysis

The headspace technique was used to estimate methane concentrations (Cole et al. 1994). A 60 mL syringe was rinsed with approximately 5-10 ml of sample water, and then 40 ml of the sample water was pulled from the jar (AE) or nalgene bottle (AN), and an additional 20 mL was filled from the air. In order to equilibrate the gas in the water with the air, the water and air mixture within the syringe was shaken for 4 min at the same temperature conditions as the jars being incubated. After 4 min, a nylon syringe and a connector was used to extract and contain the equilibrated gas from the 60 ml syringe. Shortly after the collection (usually within 1-2 hr), the concentration of CH_4 in the gas samples was determined using a Shimadzu GC-14A gas chromatograph with Class-VP 7.4 software and a 10 ppm CH_4 standard, using a flame ionization detector and a Poropak

N column. The oxygen content of each vial was estimated using a membrane inlet mass spectrometer (MIMS) (Cory et al. 2009), which allows for highly accurate measurements of dissolved oxygen in water (Kana et al. 1994).

Bacterial abundance was determined using flow cytometry (del Giorgio et al. 1996). Bacterial production was estimated via the bacterial uptake of the amino acid L-leucine (radiolabeled with ^{3}H) into protein (Kirchman et al. 1985, Simon and Azam 1989). Six autoclaved microcentrifuge tubes received 1.2 mL of the water sample. Approximately 15 min to 1 hr after sampling from the jars, two of the six tubes served as the control, and the first received 90 µL of TCA (tri-chloride acetic acid) to immediately stop any microbial activity. Then 15 µL of 1.587 µM ^{3}H-L-leucine was added to each of the six tubes and the tubes were gently mixed. The samples were allowed to incubate at the identical temperature of the jars for 1-2 hr, depending on the incubation temperature (longer incubation for cooler temperature). After the incubation, the four "live" samples received 90 µL of TCA to prevent any further bacterial assimilation of the ^{3}H-L-leucine. The samples were then centrifuged to consolidate the bacteria, and were repeatedly rinsed with 5% TCA and 80% ethanol to rid any unassimilated ^{3}H-L-leucine. After rinsing, 500 µL of scintillation cocktail was added to each tube; after 30 min the tubes were read on a scintillation counter to report the concentration of ^{3}H-L-leucine incorporated into the bacterial cells. The control samples were subtracted from the experimental samples, and bacterial production (µg C L^{-1} hr^{-1}) was estimated using ^{3}H-L-leucine concentration, radioactivity of ^{3}H-L-leucine, incubation time, volume, and C content of protein (86% of the weight). Cell specific bacterial production was also estimated by dividing the

production rate (i.e., ^{3}H-L-leucine uptake rate) by the bacterial abundance at each time point for each replicate.

Particulate carbon in the water column was estimated by filtering a known volume of sample water through an ashed (450°C for 4 hr) Whatman GF/F glass fiber filter. The filter was dried at 60°C for 24 hr, then rolled in a tinfoil capsule and analyzed for carbon and nitrogen content using a FlashEA 1112 Series NC Soil Analyzer by Thermo Electron Corporation, with Eager 300 version 2.3 software. Similarly, the plant material was extracted from each jar, and was dried at 60°C for 48 hr. After the plant material was dried and weighed, it was homogenized using a mortar and pestle. A measured amount of powder from each macrophyte or macroalgae was then wrapped in a tinfoil capsule and also analyzed for carbon and nitrogen content using the FlashEA 1112 Series as well. To estimate dissolved inorganic carbon (DIC), an ashed 7 mL scintillation vial was filled with 0.7 µm filtrate water (through an ashed Whatman GF/F) without any headspace, and run within 24 hours on a Shimadzu TOC-Vcsh. For DOC, the filtrate was immediately preserved with hydrochloric acid to lower the pH below 2. A Shimadzu TOC-Vcsh was used to estimate the concentration of DOC in the water samples as well. Specific ultraviolet absorbance (SUVA) was estimated to characterize the color and aromaticity of DOC (more color and aromatic DOC at higher values of SUVA, Weishaar et al. 2003). From the 6 mo bottle incubations, the 0.7 µm filtrate was analyzed using a Cary 50 spectrophotometer (Eaton et al. 1995), and the absorbance at 254 nm wavelength was recorded. To estimate the SUVA value, the concentration of DOC (mg L^{-1}) was divided

by the absorbance value, and the quotient was multiplied by 100 (to report in m^{-1}) (Eaton et al. 1995, Weishaar et al. 2003).

Finally, the concentration of SRP was measured to estimate labile phosphorus released from the macrophytes and algae. Approximately 20 mL of the 0.7 µm filtrate was taken and immediately frozen until later analysis. Once ready for analysis, the samples were thawed to room temperature, and a molybdenum blue color reagent was added. After waiting approximately 45 min for the color to develop, a Cary 50 spectrophotometer was used to read the absorbance of the sample at 880 nm wavelength, and the sample SRP concentration was determined using a standard curve (Eaton et al. 1995).

A MANOVA was used to test the difference between the AE and AN treatments over the course of the study, and a two-way ANOVA was used to test if there was any significant influence of primary producer type, oxygen treatment, or an interaction effect on cell specific production. Any *p*-value less than 0.10 was considered a biologically significant result. The decay constant was identified as the slope in the regression of the log transformed weight (mg) of macrophyte/algal material versus the log transformed time in days. It is implied that the slope is negative due to loss of material over time; therefore the decay constants are presented as a positive number to compare decay rates. This decay constant was used to predict the PC remaining in the PH treatments after one year, so that I could compare annual loss of carbon of the PH treatments with all other treatments.

Results

The AN replicates had significantly lower oxygen concentrations than the AE replicates (all $p < 0.01$) (Figure 20). After the initiation of the experiment, the mean dissolved oxygen concentration was less than 2 mg L^{-1} in the AN replicates, with the exception of one MY replicate at nine months, and both PH replicates at six months (Figure 20).

Generally, the macrophyte and algal C rapidly declined during the first month of decomposition (Figure 21a,b). The term "loss" for the AE treatments indicated C lost via respiration in the open system, and "loss" for the AN treatments meant any C not accounted for by changes in plant or algal C, DOC, DIC, CH_4, or PC (addressed in more detail in the discussion). The overall C loss from the original primary producer material ranged from 67 to 99% (PO-AN to PH-AE, respectively) (Figure 22a). Similarly, the degradation rates of the macrophytes were slower (decay constant average 0.24 in comparison to the algal species constant of 0.39), and the AE treatments decomposed faster (decay constant of 0.37) in than the AN treatments (decay constant of 0.29) (Figure 22b).

Bacterial production measured by ^{3}H-leucine uptake did not significantly differ between the AE and AN treatments for the CH and CL replicates (p=0.597 and 0.320, respectively) (Figure 23). In contrast, the AN treatments had significantly higher bacterial production than the AE treatments in the MY, PO, and PH (p=0.082, 0.088, 0.086, respectively). The macrophyte and AN treatments had the highest mean bacterial production rates (125 and 123 µg C L^{-1} hr^{-1}, respectively), while the algal and AE

treatments had the lowest mean production rates (59 and 52 µg C L^{-1} hr^{-1}, respectively) (Figure 23). The overall mean bacterial abundance was higher in the AN treatments versus the AE treatments ($2.0x10^9$ versus $7.3x10^8$ cells L^{-1}, respectively) (Figure 24). There was no significant difference between the AE and AN treatments for the algae (CH p=0.103, CL p=0.125, PH p=0.267), while the MY-AN and MY-AE treatments were significantly different (p=0.019). The PO treatments did not have enough replicates to statistically test whether the AN or AE treatments had higher bacterial abundance rates; however, in comparison to the other treatments, it seemed that the AN treatments had higher bacterial abundance. There was no significant difference in cell specific production between the macrophyte and algal treatments or the AE and AN treatments (p = 0.980, 0.397, respectively), and there was no interaction between primary producer and oxygen treatment (p = 0.750) (Figure 25).

There was no significant difference in the concentration of SRP between the AN and AE treatments for all primary producers (CH p=0.921, CL p=0.521, PH p=0.438, MY p=0.933, PO p=0.162) (Figure 26). SRP values ranged from 3 to 52 µM for all replicates. The CH, CL, MY, and PO treatments experienced an increase in SRP concentration at the start of the experiment, while it remained low in the PH treatments.

DOC greatly increased at the beginning of the experiment in all the AN treatments PH replicates, while it did not accumulate at all in the AE treatments (Figure 27). The macrophyte and AN treatments had the highest concentrations (41 and 47 mg L^{-1}, respectively), while the algal and AE treatments had lower mean concentrations (24 and 15 mg DOC L^{-1}) (Figure 27). DIC increased sharply in the CH-AN and CL-AN

treatments, while it remained relatively low and stable for all other treatments (Figure 28). However, DIC was significantly greater in the AN treatments for CH, CL, MY and PO (all $p < 0.01$), but the DIC concentration did not significantly differ between the PH-AE and PH-AN treatments ($p = 0.271$) (Figure 28). Methane concentrations in the AN treatments at the end of the experiment were approximately 370 times greater than the AE treatments (Figure 29).

The SUVA values indicated that the DOC ranged from slightly colored to greatly colored (1.6 to 4.4) (Figure 30). This suggests that DOC ranged from 14-32% in aromaticity (Weishaar et al. 2003). CL-AN replicate had the highest aromaticity (32%), whereas CH-AN had the least (14%). CL, PH, and the PO treatments had significantly higher SUVA values in the AE versus AN treatments (p=0.008, 0.006, and <0.0001), although the significance of the PO treatments was likely driven by no replication in the PO-AE treatment. Except for the CL treatments, all SUVA values were higher in the AE versus AN treatments (Figure 30).

Discussion

Overall the submerged macrophytes decomposed at slower rates than the algal species, and similarly the anaerobic treatments experienced slower decomposition rates in comparison to the aerobic treatments. Although the macrophyte-AN treatments had a slower rate of decay, they were associated with higher bacterioplankton production, which was likely due to increased microbial abundance associated with high levels of DOC and/or higher growth efficiencies. Furthermore, in these treatments the C was

retained in the microbial biomass and not respired to CO_2. Therefore, the efficiency in which carbon was retained and not respired was likely the highest in the macrophyte-anaerobic treatments. In terms of the clear and turbid regimes, the primary producers of the clear regimes retained approximately 24% of their original carbon content, whereas the phytoplankton of the turbid regime only retained 4%.

Macrophyte versus Algal Decomposition

The fastest rate of loss of carbon bound within the macrophyte and algal species occurred during the first month, which is a typical observation in experiments involving the decomposition of primary producer biomass (Godshalk and Wetzel 1978a, Webster and Benfield 1986, Moran et al. 1989, Belova 1993, Asaeda et al. 2000). The loss of the initial macrophyte and algal C ranged from 50 to 98% within the first month (MY-AE and PH-AE, respectively). Thereafter, the decomposition rate of the macrophyte and algal material slowed across the rest of the experiment. At the end of the year-long experiment, there was a greater amount of primary producer C remaining in the macrophyte treatments in comparison to the algal treatments. Over the course of one year, the macrophytes and algae lost on average 77% and 86% of their original C mass to dissolved or gaseous components, respectively (Figure 22a). Furthermore, the mean decay constant for the macrophytes was 0.24, which was 0.15 units slower than the constant for algal matter (0.39, Figure 22b). The slower decomposition rate of the macrophytes was likely due to a greater structural complexity (Enríquez et al. 1993), as

macrophytes contain more complex compounds like lignin and cellulose which require more time and energy to decompose.

Unlike phytoplankton, *Charophytes* are a type of algae that contains cellulose similar to that of higher plants (Koyama et al. 1997, Garvey et al. 2006). *Charophytes* tend to retain even more C than some vascular plants (Kufel and Kufel 2002), as they contain more fiber content than other submerged macrophytes, leading to slower decomposition rates (Shilla et al. 2006). Our results support this notion, as the CH treatments collectively lost 75% of their original C content, which was slightly lower than the mean C loss from the macrophytes (77%) (Figure 22a). In shallow lakes, *Chara* is commonly found in the clear-water regime, and acts as a stabilizing agent for the clear regime as it negates sediment resuspension, immobilizes phosphorus via binding or sorption onto calcite particles, and has an allelopathic influence on phytoplankton (Kufel and Kufel 2002, Nõges et al. 2003). Although *Chara* is a macroalga, it functions much like a macrophyte from an ecological standpoint. Because *Chara* retained a much greater amount of C in comparison to the other algal species, this result further supports the hypothesis that the clear water regime, either dominated by macrophytes or *Chara* (macroalgae), accumulates carbon at a higher rate than phytoplankton-dominated, turbid-regime.

More primary producer C remained in the macrophyte treatments over the course of the experiment. If the decomposition of this material was predominately mediated by microbes, then lower bacterial production rates should have been observed; however, this was not the case. On average, bacterial production in the macrophyte treatments exceeded

rates observed in the algal treatments (125 versus 59 μg C L^{-1} hr^{-1}, respectively) (Figure 23). Macrophytes support high bacterioplankton production (Rooney and Kalff 2003), and on average, the macrophyte treatments had almost twice as much DOC as the algal treatments (grand mean of 41 versus 24 mg DOC L^{-1}, respectively) (Figure 27). This amount of DOC likely supported higher levels of bacterial populations, and thus high bacterial production in the macrophyte treatments. Bacterial growth on macrophyte leachate tends to be more efficient in comparison to growth on phytoplankton organic matter (Stets and Cotner 2008), which matches the differences in observed peaks in DIC of the algal and macrophyte treatments (higher peak of DIC in the algal treatments due to greater respiration and less efficiency) (Figure 28).

Aerobic versus Anaerobic Decomposition

Based on the mass loss of C, the decomposition rate of the AN treatments was slower than the AE treatments (0.29 and 0.37, respectively) (Figure 22b). Consequently, 85% of the original primary producer C content was lost in the AE treatments, while 80% was lost in the AN treatments (Figure 22a). All treatments except for CH had higher C content loss in the AE treatments. The lower retention of primary producer C in the AE treatments may be due to lower growth efficiencies and/or a more energetically favorable environment for organic matter breakdown (Godshalk and Wetzel 1978a). As the primary producer C mass sharply decreased within the first month of decomposition, the CH-, CL-, PO-, and MY-AN treatments experienced a large increase in DOC, while the AE treatments only experienced a slight increase (Figure 27). During the very early stages of

decomposition, autolysis mainly drives the transformation of particulate organic matter to dissolved forms (i.e. DOC), and this likely occurred at the same rate under the AN and AE conditions (Otsuki and Wetzel 1974, Godshalk and Wetzel 1978a). However, under anaerobic conditions, organic matter is difficult for microbes to breakdown completely because a combination of several microbial populations, i.e., consortia, with specialized enzymes and metabolic processes is needed to efficiently decompose the organic material (Godshalk and Wetzel 1978a; Briée et al. 2007). If these populations do not exist simultaneously at high enough concentrations, the breakdown of the organic material is further inhibited (Godshalk and Wetzel 1978a). Therefore, the DOC in the AN treatments likely accumulated due to the time it took to establish microbial consortia that facilitated breaking down the DOC. This seemed to have occurred after approximately one month of decay since the DOC concentration declined quickly thereafter, with an associated increase in microbial abundance in the macrophyte treatments.

Bacterial production from the water column in the AN treatments was over two times the rate of the AE treatments (123 versus 52 µg C L^{-1} hr^{-1}, respectively) (Figure 23). Similarly, bacterial abundance in the AN treatments was almost three times as much in the AE treatments (2.0×10^9 versus 7.3×10^8 cells L^{-1}, respectively) (Figure 24). Furthermore, the DOC concentration was also more than three times as high in the AN treatments than the AE treatments (47 and 15 mg DOC L^{-1}, respectively) (Figure 27). Because the production rates per cell did not significantly differ between AN and AE treatments ($p = 0.397$) (Figure 25), the higher production rates of the AN treatments was supported by the higher bacterial abundance and DOC concentration.

The treatments with the highest cellulose content (CH, MY and PO) all had high populations of bacteria, and the breakdown of the cellulosic material most likely took place in the AN treatments. The breakdown of cellulose under anaerobic conditions is very common (Leschine 1995), and requires particular bacteria with specific enzymes to fully decompose the cellulose. The decay of cellulose produces methane (Leschine 1995), and although methane only comprised <0.01% of the total carbon (mean weight) in all the treatments, its concentration was approximately 370 times greater in the AN treatments versus the AE treatments (Figure 29). Although it is possible that methane could have been produced in the AE treatments and not contained within the open system, this large difference in methane concentration between the two treatments was most likely due to higher rates of methanogenesis in the AN treatments. Substances such as cellulose absorb ultraviolet radiation (Godshalk and Wetzel 1978a), and the SUVA values indicate lower UV absorbance in the AN treatments (Figure 30), which supports the notion that the cellulose was broken down to a greater extent, and that the portion of the DOC also likely comprised low molecular weight compounds that do not absorb as much ultraviolet light (Godshalk and Wetzel 1978a). In aerobic environments, low molecular weight carbon compounds tend to be mineralized faster (Godshalk and Wetzel 1978a). Consequently, the high SUVA values of the AE treatments indicated that although DOC was in low concentration, it was comprised of mostly aromatic compounds, which are more difficult for microbes to breakdown in aerobic environments.

There were two seemingly unusual results from AE versus AN experiment that I would like to address. First, all the primary producers retained more carbon in the AN treatments, except for the *Charophytes*, which retained less carbon under anaerobic conditions. *Charophytes* characteristically are known for precipitating calcium carbonate out of the water column during photosynthesis, and accumulating the calcite on their surface (Kufel and Kufel 2002). This calcite can comprise up to 60% of the total dry weight of this macroalga (Hutchinson 1975), but can readily dissolve at low pH and low temperatures (Stumm and Morgan 1981). Because the AN treatments had a lower pH than the AE treatments (pH 7 versus 8, respectively), it was possible that calcite dissolution occurred in the AN treatments and caused greater carbon loss. However, the loss was only greater in the AN treatments in comparison to the AE treatments at the end of the experiment (Figure 21a); if calcite dissolution was the cause of these differences in carbon loss, the *Charophytes* in the AN treatments should have retained less carbon throughout the duration of the experiment and not just at the end. Therefore, the greater loss of CH carbon in the AN treatment at the end of the experiment may have been influenced by temperature; warmer temperatures may have initiated greater leaching and/or microbial mediated breakdown of the organic matter, which correlates with slightly higher production rates and DIC at the end of the experiment (Figure 23,28).

Another unusual result was the high SUVA values of the CL-AN treatment in comparison to all other combinations of treatments (Figure 30). In contrast to the macrophyte AE treatments, the CL-AN treatment was cloudy white instead of a deep gold, and its replicates also accumulated a white microbial mat at the surface of the

water. This white coloration was likely caused by the presence of sulfide oxidizers instead of humic DOC, and caused higher absorbance which produced high SUVA values. The significant presence of sulfide oxidizers in the CL-AN treatments likely has to do with the influence of CL on reduced sulfur concentrations, and the substrate it provides for this particular type of bacteria. Unfortunately, information on the relationship between this filamentous algae and sulfur cycling is essentially lacking in the literature, and needs to be explored more in depth.

Comparison of Carbon Retention among All Treatments

This experiment showed that algal primary producers decompose more quickly than their macrophyte counterparts, and that the presence of oxygen impedes the retention of primary producer carbon biomass, i.e., the aerobic systems were the least efficient at retaining carbon in comparison to the anaerobic systems. First, they exhibited the greatest loss of carbon bound within the primary producer material, and this particulate matter became mineralized to the dissolved or gaseous form of carbon via a combination of microbial activity and autolysis. Because the AE treatments were open systems, the loss likely represented the amount of carbon that escaped the system in the form of CO_2 or CH_4. In comparison to the macrophyte-AN treatments, the CH-AN and CL-AN treatments showed sharp increases in DIC after approximately one month of decomposition (Figure 28). In contrast to the macrophyte treatments, the carbon in the CH and CL-AN treatments was respired causing the increase in DIC in the closed system. In contrast, less C in the macrophyte-AN treatments was respired and more of it remained

in the primary producer itself and in the bacterial biomass, as these treatments had the highest bacterial production and abundance rates, and only a small increase in DIC.

When comparing the overall loss of carbon between the treatments (macrophyte versus algae, and aerobic versus anaerobic), the type of primary producer had a slightly greater impact on carbon retention than the oxygen regime. Macrophytes retained 9% more C than the algal species, while the AN treatments retained 5% more C than the AE treatments. While the differences between these treatments seems rather trivial, the ecological implications of these results in terms of clear and turbid regimes are better understood if the primary producers were instead grouped by the regime in which they typically inhabit. Even though three types of algae were included in this experiment, only the PH treatment represented the lakes in the turbid regime. In comparison, the CH, MY and PO primary producers are common species found in lakes of the clear-water regime. Grouped this way, an average of 76% of the original C content was lost in the clear regime species, while 96% of the original C content of phytoplankton was lost over the course of one year (Figure 31). This difference is large, and over time could potentially lead to a large discrepancy in carbon burial. If these decay rates were applied to the mean annual C fixation rates of the clear and turbid water regimes (486 and 604 g C m^{-2} yr^{-1}, respectively, Chapter 1), approximately 116 and 24 g C m^{-2} would remain over the course of one year in the clear and turbid regimes, respectively. These rates are similar to carbon accumulation rates observed in shallow lakes (Dean and Gorham 1998, Cotner et al. *unpublished data*), and also support the hypothesis that clear and turbid regimes likely experience differing rates of carbon accumulation.

The decay of *Cladophora*, *Charophytes*, and macrophytes toward the end of the growing season typically causes a fast decline in oxygen (Paalme et al. 2002, Mihranyan 2010), and the clear-water regime under anaerobic conditions retained the greatest percentage of primary producer carbon content (26%, Figure 31). In terms of carbon accumulation, the "benefits" of the clear water state is two-fold: first the primary producer material decomposes more slowly due to a more complex composition; and second anaerobic conditions are produced shortly after the macrophytes and/or macroalgae senesce, which also favor carbon burial. If managers wish to manage shallow lakes to retain carbon, they may focus on manipulating the lake regime, which could also subsequently cause the desired anaerobic environment to retain carbon.

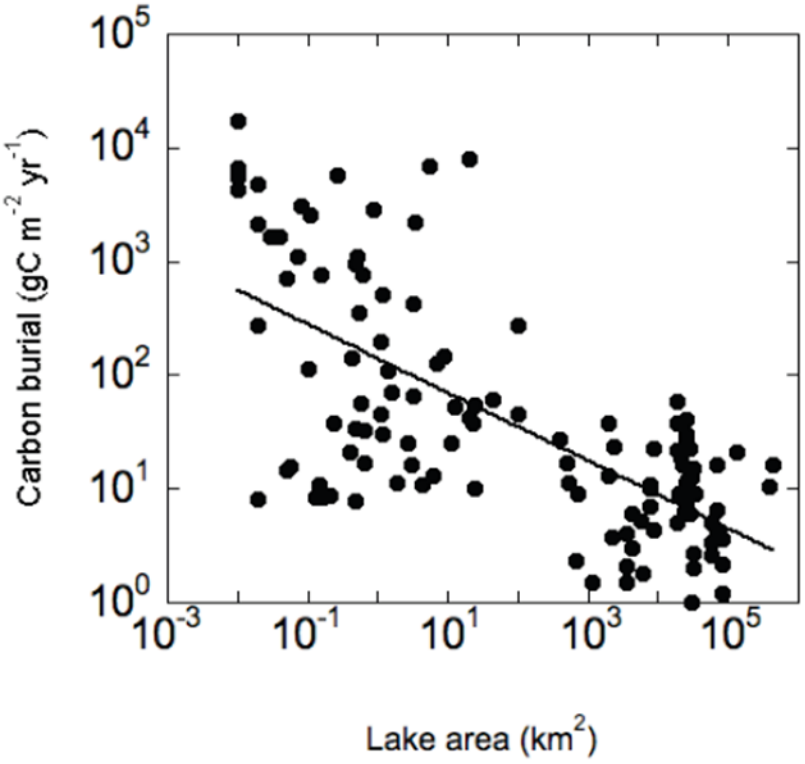

Figure 18. Carbon burial versus lake area. Carbon burial is high in small lakes, however, there is much variation in these burial rates. Data points are from published and unpublished literature.

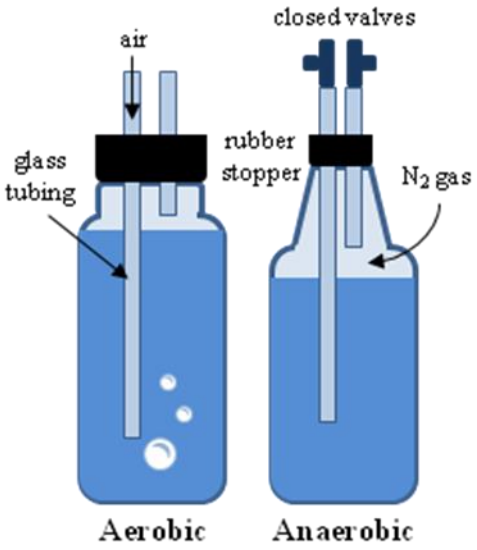

Figure 19. The design of the bottles used to experimentally decompose organic matter from differing primary producers under aerobic and anaerobic conditions. The aerobic conditions received air bubbling through, whereas the anaerobic treatments were initially bubbled with N_2 gas to rid oxygen out of the water and headspace.

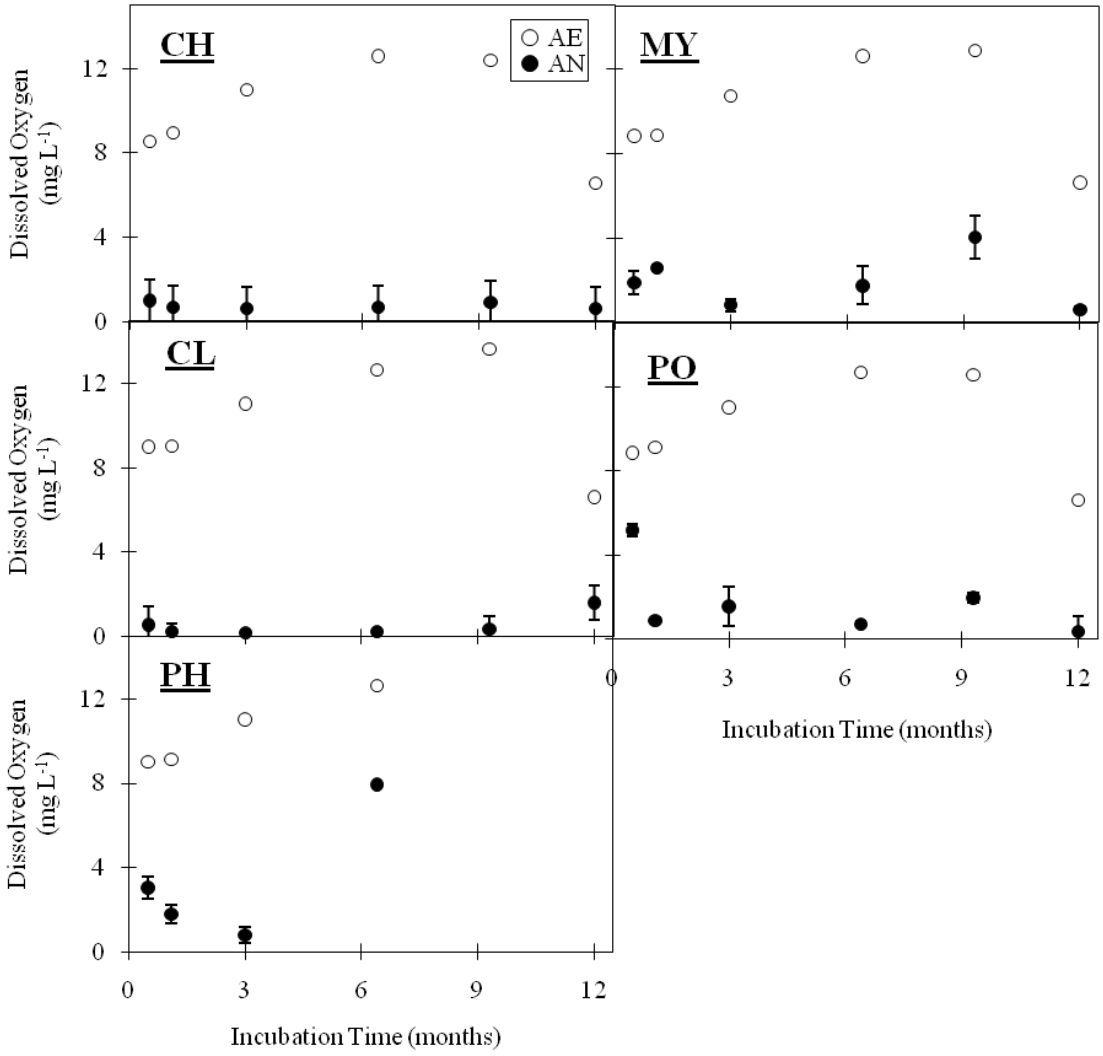

Figure 20. Dissolved oxygen concentrations for all replicates during the experiment. The white circles represent the aerobic treatments, while the solid black circles represent the anaerobic treatments. The error bars represent standard error. All aerobic treatments were significantly higher in oxygen than the anaerobic treatments (all $p < 0.01$).

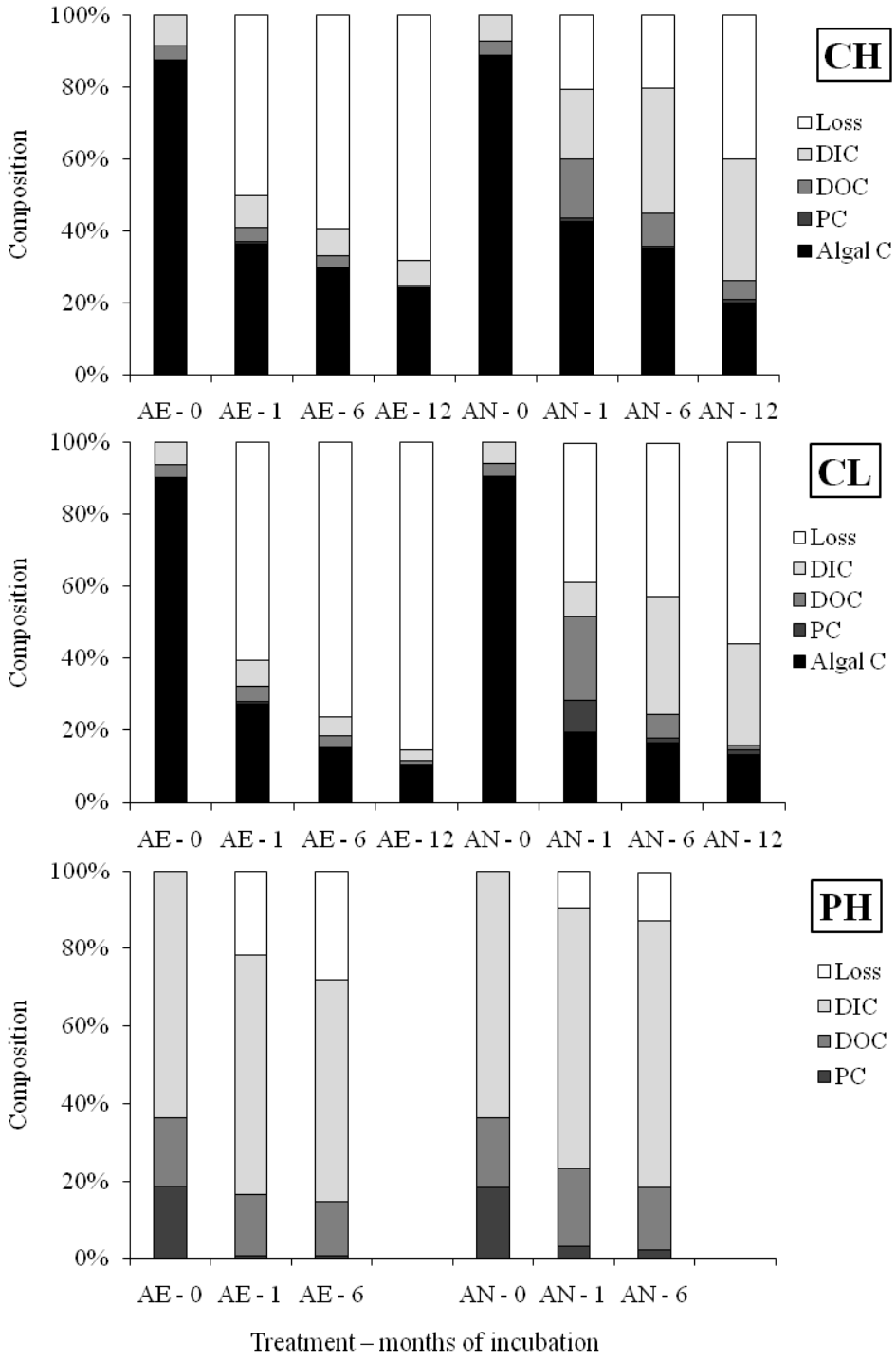

Figure 21a. Percent composition of carbon allocations for the "algal" aerobic (AE) and anaerobic (AN) treatments over time. The total amount of C at the start of the experiment was 863, 865, 618, 842, 33, and 38 mg C for the CH-AE, CH-AN, CL-AE, CL-AN, PH-AE, and PH-AN treatments, respectively. "Algal C" represents the percent of carbon added as particulate material, and "PC" is the particulate carbon suspended within the water column. Loss represents carbon lost to the atmosphere in the AE replicates, and carbon unaccounted for in the AN replicates.

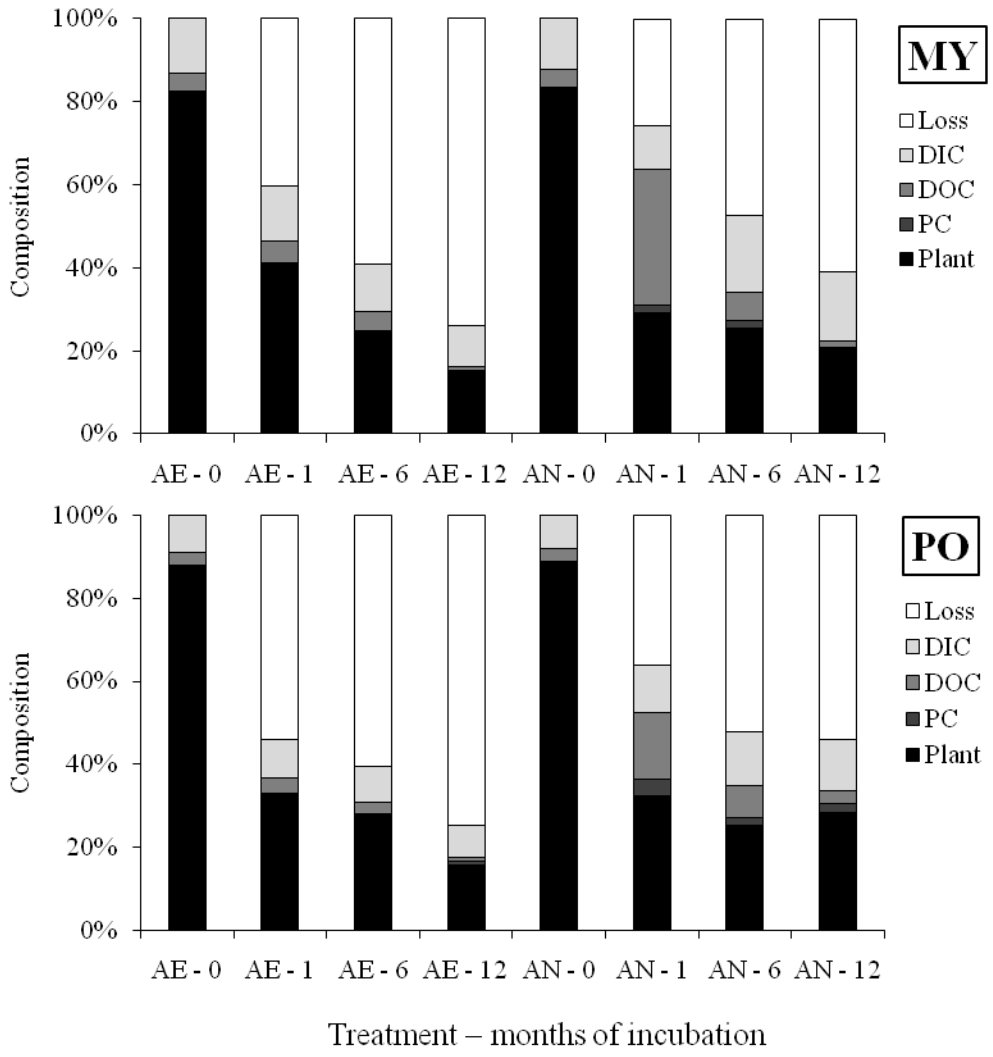

Figure 21b. Percent composition of carbon allocations for the "macrophyte" aerobic (AE) and anaerobic (AN) treatments over time. The amount of C at the start of the experiment was 600, 813, 886, and 889 mg C for the MY-AE, MY-AN, PO-AE, and PO-AN treatments. "Plant" represents the percent of carbon added as particulate material, and "PC" is the particulate carbon suspended within the water column. Loss represents carbon lost to the atmosphere in the AE replicates, and carbon unaccounted for in the AN replicates.

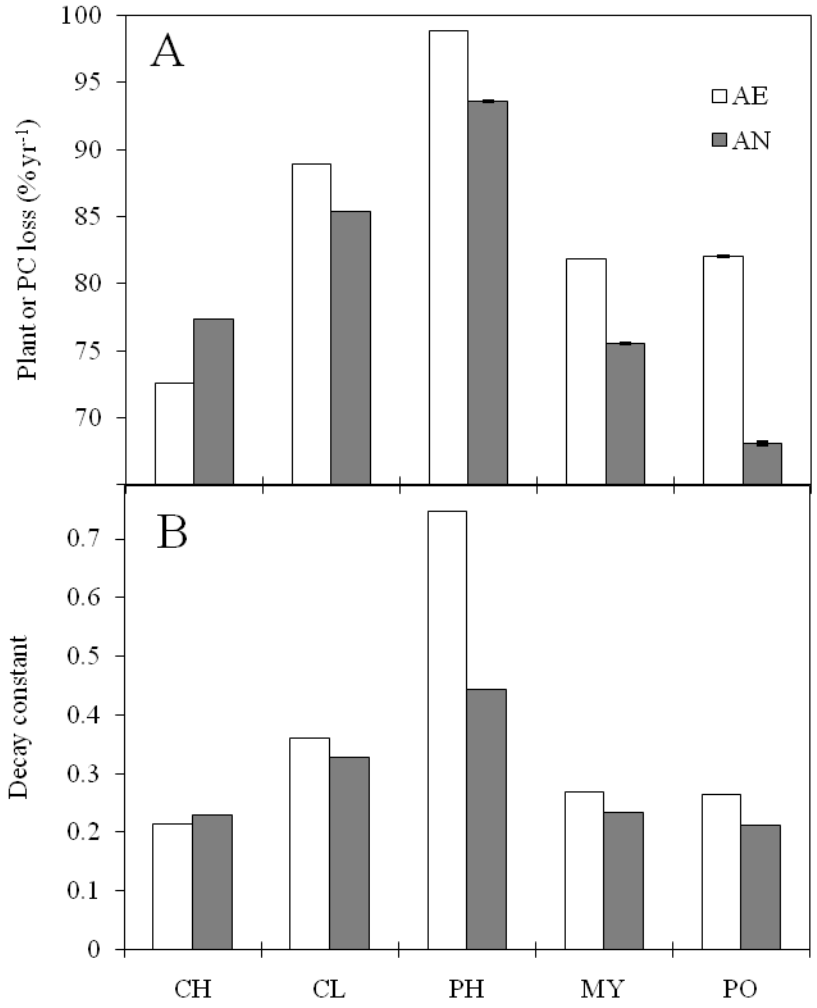

Figure 22. A) Loss of carbon weight (%) of the plant or particulate carbon (for PH) over the duration of the experiment. Error bars represent standard error. B) The decay constant for the replicates (estimated as the slope of a log-log plot of particulate carbon weight over time). The white and grey bars represent the aerobic and anaerobic treatments, respectively.

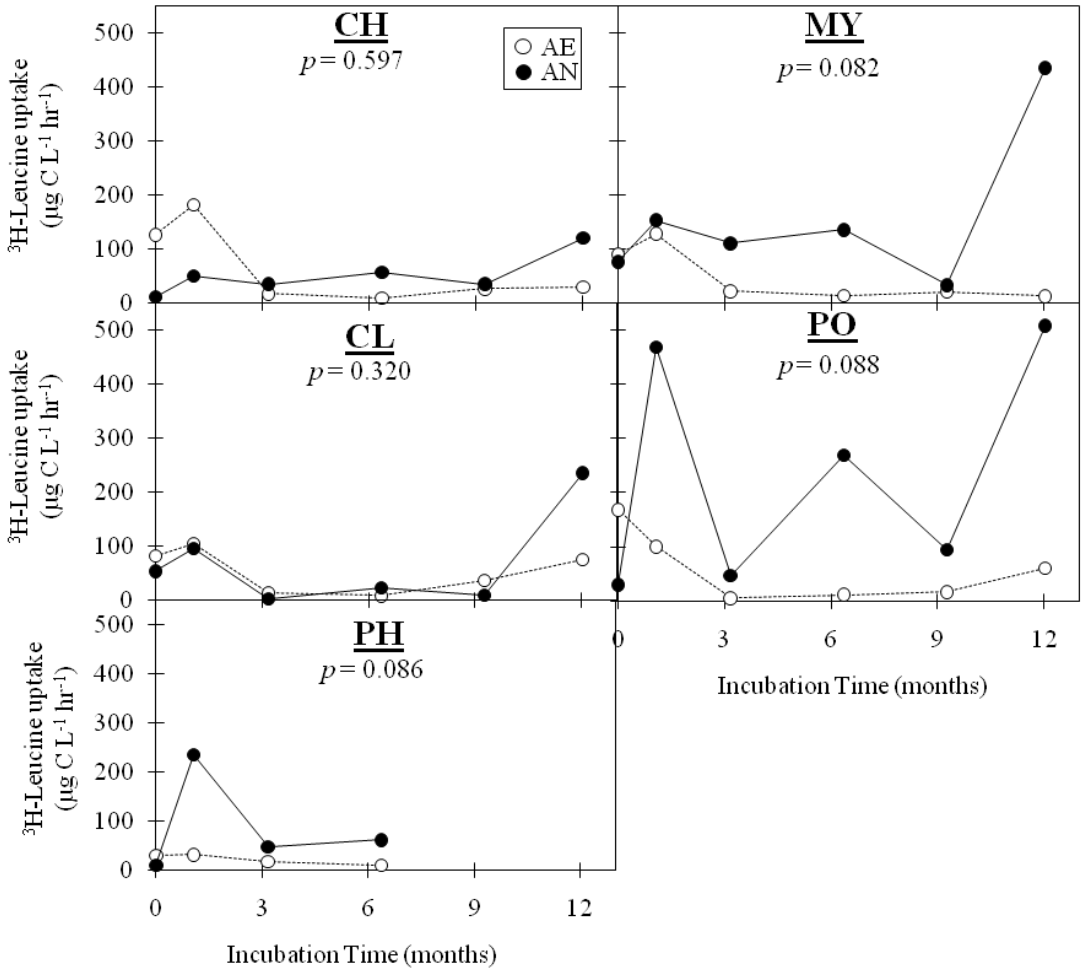

Figure 23. ^{3}H-leucine uptake for the algal and macrophyte species over the course of the experiment. Solid white circles represent the aerobic treatments (AE), and the solid black circles represent the anaerobic treatments (AN). Error bars represent standard deviation about the mean (which are often too small to show), and the p-values indicate the significance of the difference between the AE and AN treatments.

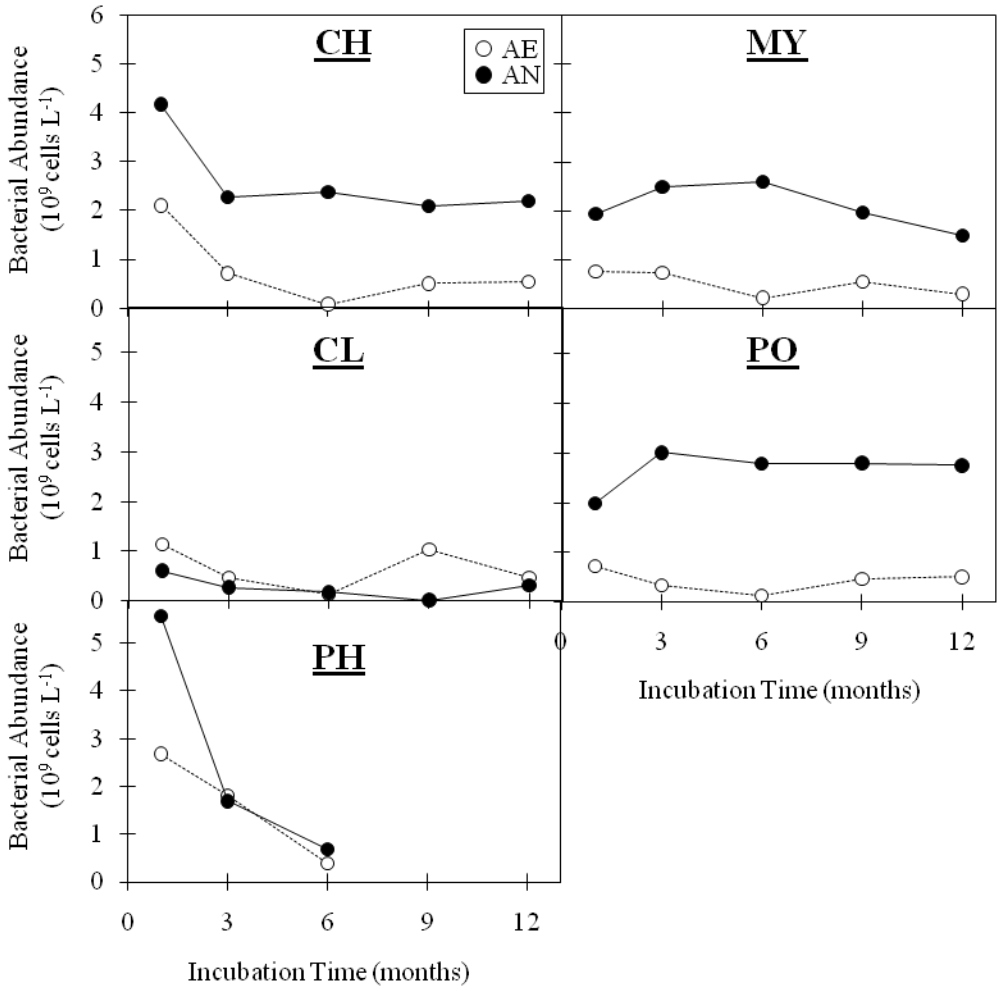

Figure 24. Bacterial abundance estimated using flow cytometry for the algal and macrophyte species over the course of the experiment. Solid white circles represent the aerobic treatments (AE), and the solid black circles represent the anaerobic treatments (AN). Standard error bars are too small to be seen on these graphs. There was no significant difference between the AE and AN treatments for the algae (CH p=0.103, CL p=0.125, PH p=0.267), while the MY AN and AE treatments were significantly different (p=0.019). The PO treatments did not have enough replicates for statistical testing.

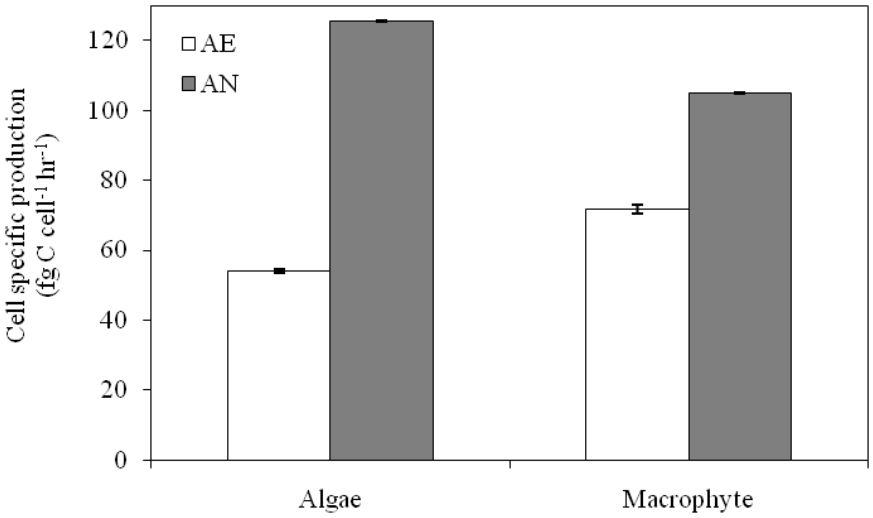

Figure 25. Cell specific production, estimated by dividing bacterial production rate (^{3}H-L-leucine uptake) by bacterial abundance. There was no significant difference in cell specific production between the macrophytes and algae ($p = 0.980$), or the AE and AN treatments ($p = 0.397$), and there was no interaction between the primary producer type and oxygen treatment ($p = 0.750$).

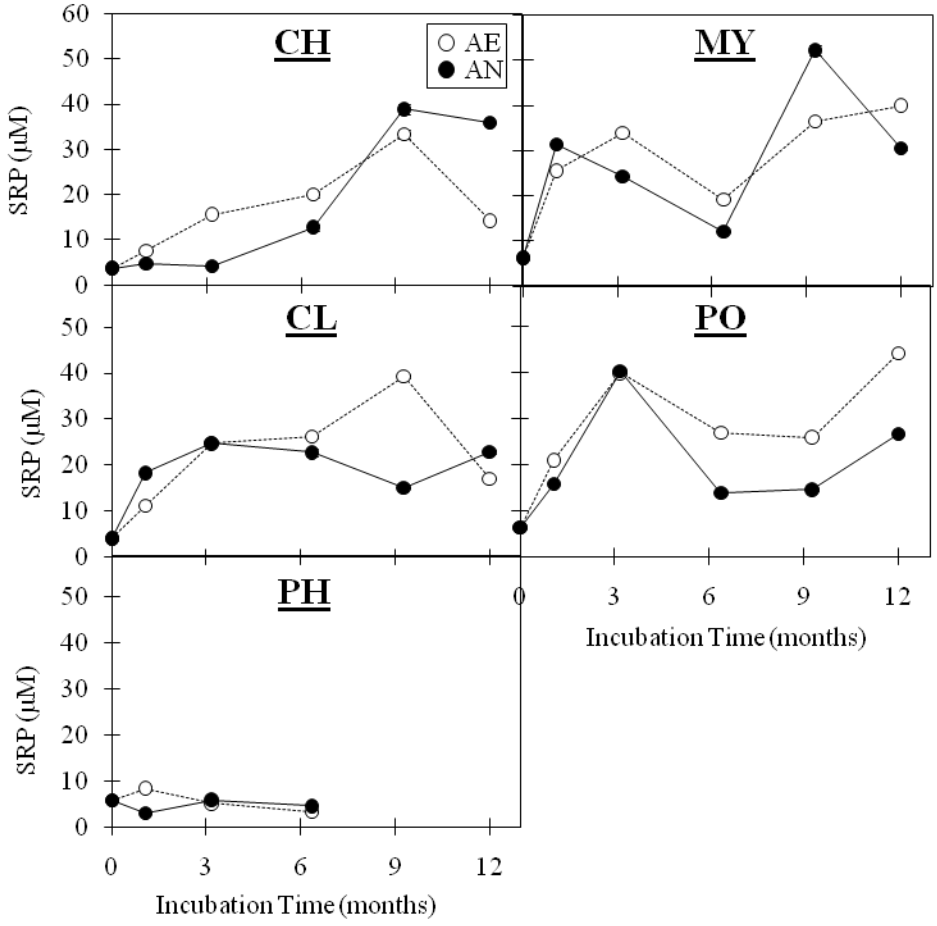

Figure 26. Soluble reactive phosphorus concentration in the algal and macrophyte species over the course of the experiment. Solid white circles represent the aerobic treatments (AE), and the solid black circles represent the anaerobic treatments (AN). Error bars represent standard error, and the *p*-values indicate the significance of the difference between the AE and AN treatments. There was no significant difference in the concentration of SRP between the AN and AE treatments (CH p=0.921, CL p=0.521, PH p=0.438, MY p=0.933, PO p=0.162).

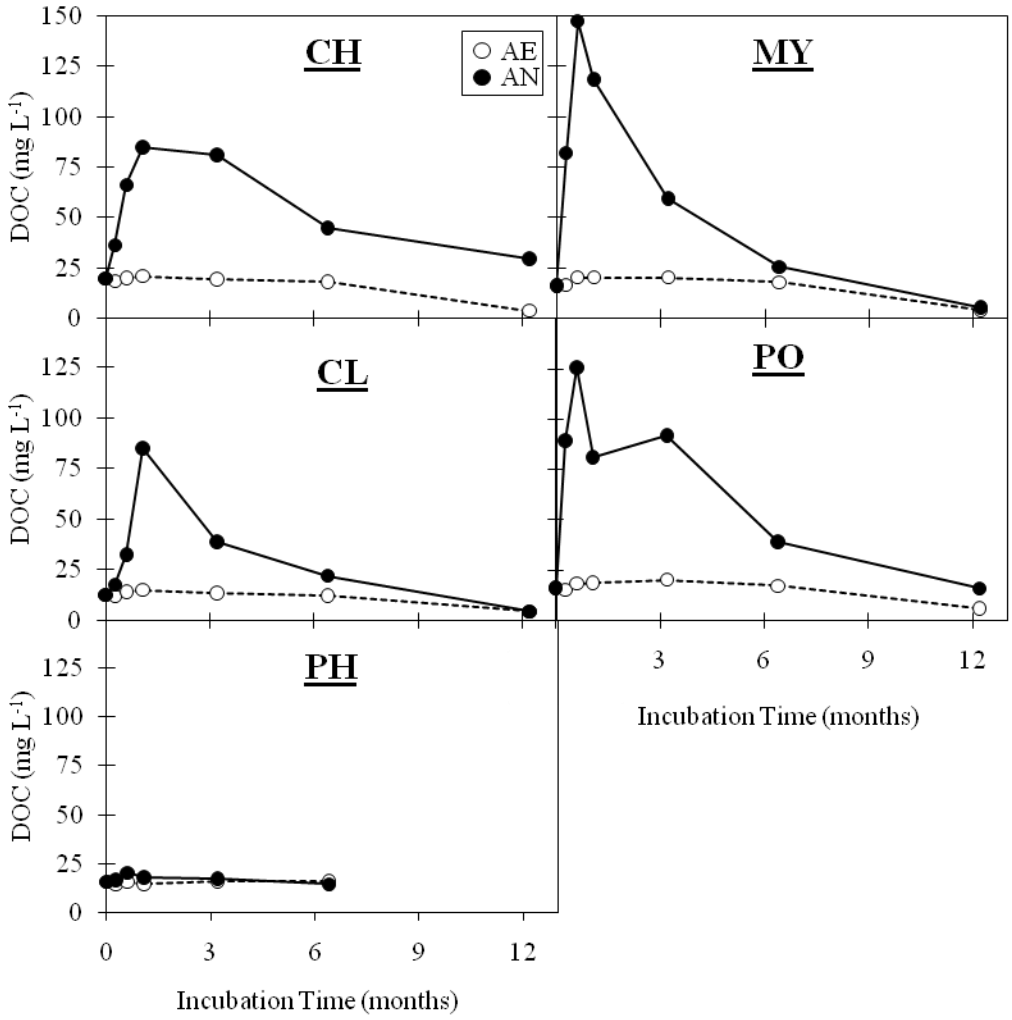

Figure 27. Dissolved organic carbon in the algal and macrophyte treatments over the course of the experiment. Solid white circles represent the aerobic treatments (AE), and the solid black circles represent the anaerobic treatments (AN). The AN treatments experienced a large pulse of DOC in the early stages of decomposition for all treatments.

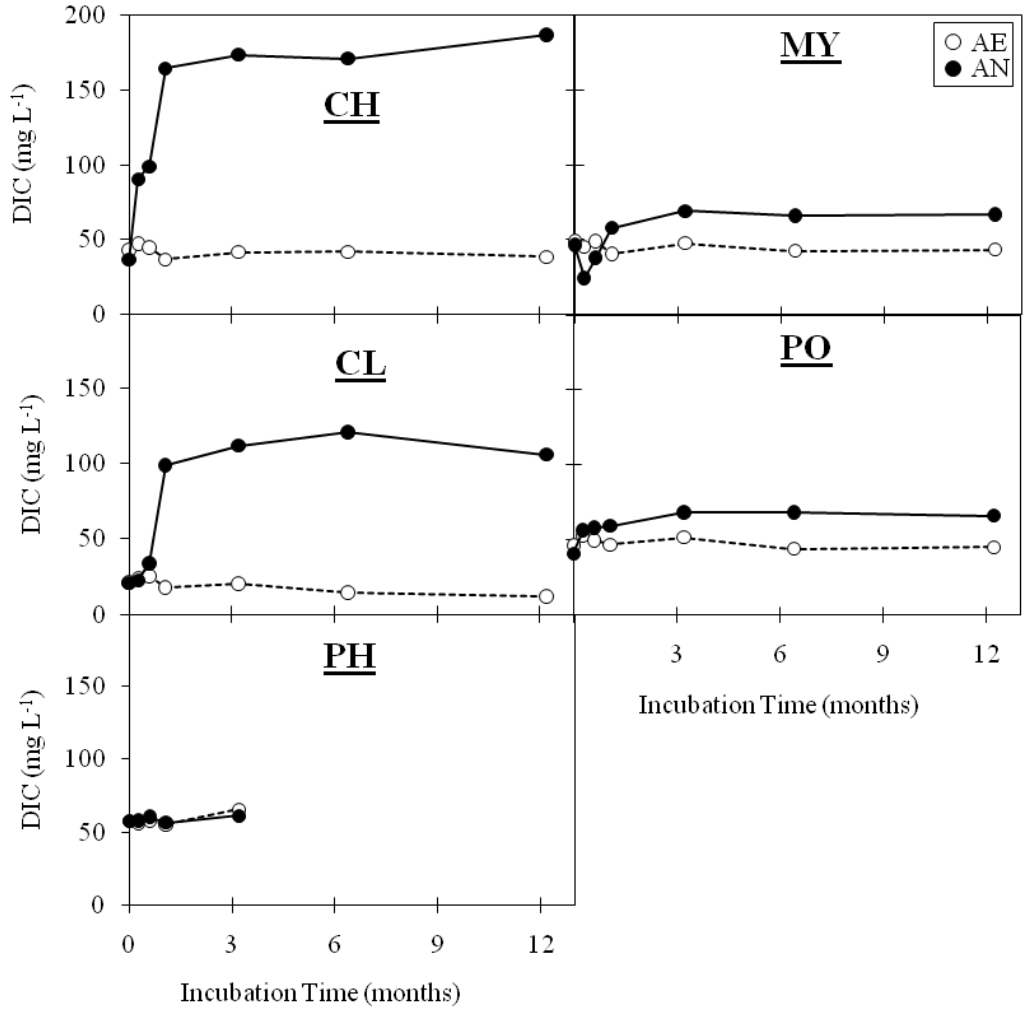

Figure 28. Dissolved inorganic carbon in the algal and macrophyte treatments over the course of the experiment. Solid white circles represent the aerobic treatments (AE), and the solid black circles represent the anaerobic treatments (AN). The CH-AN and CL-AN treatments experienced a large pulse of DIC in the early stages of decomposition in comparison to all other treatments.

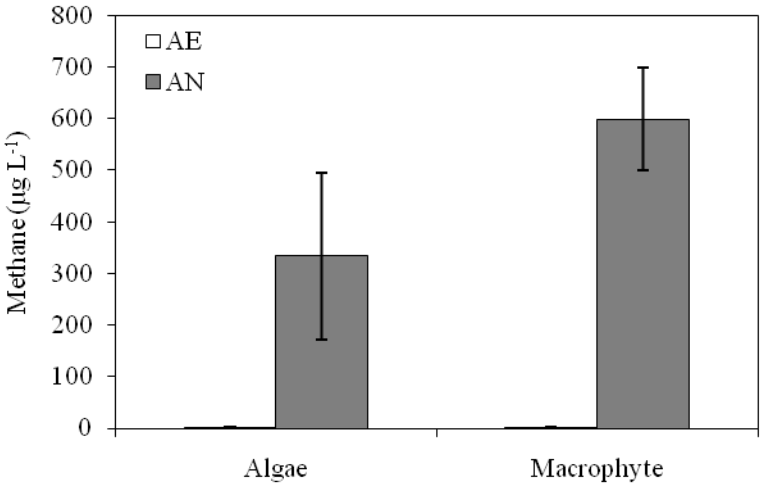

Figure 29. Methane concentrations (grand means) of the treatments at the end of the experiment. The aerobic treatments had methane concentrations < 1.5 µg L^{-1}. Error bars represent standard deviation.

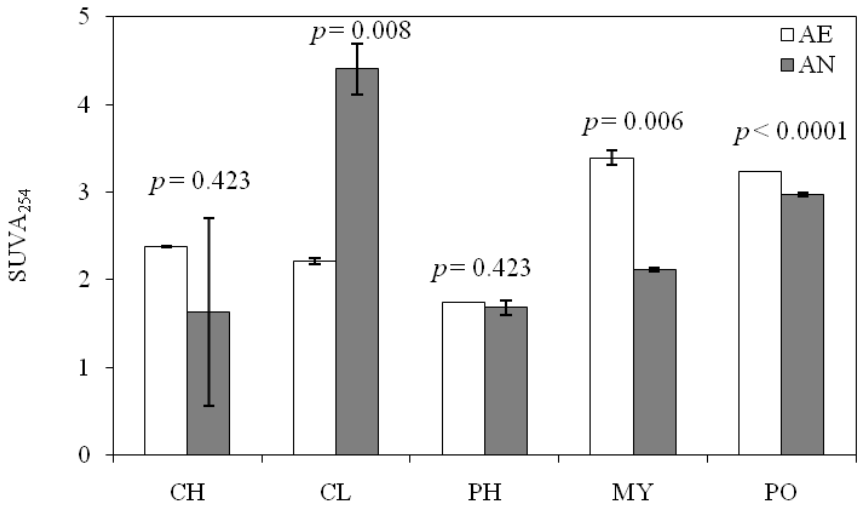

Figure 30. Specific ultraviolet absorbance at 254 nm wavelength at 6 months into the experiment. White bars represent the aerobic treatments (AE), and the grey bars represent the anaerobic treatments (AN). The error bars represent standard error. The p-values represent the difference between the AE and AN treatments. Overall, the macrophyte-AE treatments and the CL-AN s had the highest SUVA values.

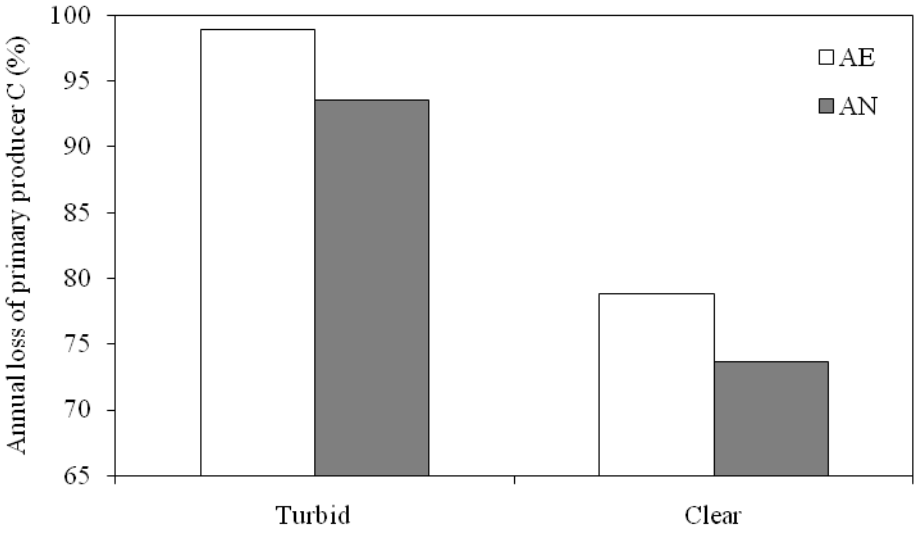

Figure 31. Annual loss of primary producer carbon for the species common in the turbid regimes (PH) and clear regimes (CH, PO, MY). The AE and AN treatments for the turbid regime had annual loss of 99% and 94%, respectively. In contrast the percentage loss for the species of the clear regimes were 79% and 74% for the AE and AN treatments, respectively.

Bibliography

Asaeda, T., V.K. Trung, and J. Manatunge. 2000. Modeling the effects of macrophyte growth and decomposition on the nutrient budget in shallow lakes. Aquatic Botany 68: 217-237.

Andersson, E. and A.K. Brunberg. 2006. Net autotrophy in an oligotrophic lake rich in dissolved organic carbon and with high benthic primary production. Aquatic Microbial Ecology 43: 1-10.

Belova, M. 1993. Microbial decomposition of freshwater macrophytes in the littoral zone of lakes. Hydrobiologia 251: 59-64.

Benner, R., A.E. MacCubbin, and R.E. Hodson. 1984. Anaerobic biodegradation of lignin and polysaccharide components of lignocelluloses and synthetic lignin by sediment microflora. Applied and Environmental Microbiology 47(5): 998-1004.

Blindow, I., A. Hargeby, J. Meyercordt, and H. Schubert. 2006. Primary production in two shallow lakes with contrasting plant form dominance: A paradox of enrichment? Limnology and Oceanography 51(6): 2711-2721.

Briée, C., D. Moreira, and P. López-García. 2007. Archael and bacterial community composition of sediment and plankton from a suboxic freshwater pond. Research in Microbiology 158(3): 213-227.

Carignan, R., D. Planas, and C. Vis. 2000. Planktonic production and respiration in oligotrophic shield lakes. Limnology and Oceanography 45(1): 189-199.

Carpenter, S.R. and D.M. Lodge. 1986. Effects of submersed macrophytes on ecosystem processes. Aquatic Botany 26: 341-370.

Cebrian, J. 1999. Patterns in the fate of production in plant communities. American Naturalist 154(4): 449-468.

Chapin, F.S. III, G.M. Woodwell, J.T. Randerson, E.B. Rastetter, G.M. Lovett, D.D. Baldocchi, D.A. Clark, M.E. Harmon, D.S. Schimel, R. Valentini, C. Wirth, J.D. Aber, J.J. Cole, M.L. Goulden, J.W. Harden, M. Heimann, R.W. Howarth, P.A. Matson, A.D. McGuire, J.M. Melillo, H.A. Mooney, J.C. Neff, R.A.Houghton, M.L. Pace, M.G. Ryan, S.W. Running, O.E. Sala, W.H. Schlesinger, and E.D. Schulze. 2006. Reconciling carbon-cycle concepts, terminology, and methods. Ecosystems 9: 1041-1050.

Cole, J.J. and G.E. Likens. 1979. Measurements of mineralization of phytoplankton detritus in an oligotrophic lake. Limnology and Oceanography 24(3): 541-547.

Cole, J.J., N.F. Caraco, G.W. Kling, and T.K. Kratz. 1994. Carbon dioxide supersaturation in the surface waters of lakes. Science 265(5178): 1568-1570.

Cole, J.J. and N.F. Caraco. 1998. Atmospheric exchange of carbon dioxide in a low-wind oligotrophic lake measured by the addition of SF_6. Limnology and Oceanography 43(4): 647-656.

Cole, J.J. and N.F. Caraco. 2001. Carbon in catchments: connecting terrestrial carbon losses with aquatic metabolism. Marine and Freshwater Research 52: 101-110.

Cole, J.J., M.L. Pace, S.R. Carpenter, and J.F. Kitchell. 2000. Persistence of net heterotrophy in lakes during nutrient addition and food web manipulations. Limnology and Oceanography 45(8): 1718-1730.

Cole, J.J., Y.T. Prairie, N.F. Caraco, W.H. McDowell, L.J. Tranvik, R.G. Striegl, C.M. Duarte, P. Kortelainen, J.A. Downing, J.J. Middleburg, and J. Melack. 2007. Plumbing the global carbon cycle: integrating inland waters into the terrestrial carbon budget. Ecosystems 10:171-184.

Coloso, J.J., J.J. Cole, P.C. Hanson, and M.J. Pace. 2008. Depth-integrated, continuous estimates of metabolism in a clear-water lake. Canadian Journal of Fisheries and Aquatic Sciences 65: 712-722.

Cory, R.M., J.B. Cotner, and K. McNeill. 2009. Quatifying interactions between singlet oxygen and aquatic fulvic acids. Environmental Science Technology 43(3): 718-723.

Cotner, J.B. and B.A. Biddanda. 2002. Small players, large role: Microbial influence on biogeochemical processes in pelagic aquatic ecosystems. Ecosystems 5(2): 105-121.

Dauwe, B., J.J. Middelburg, and P.M.J. Herman. 2001. Effect of oxygen on the degradability of organic matter in subtidal and intertidal sediments of the North Sea area. Marine Ecology Progress Series 215: 13-22.

Dean, W.E. and E. Gorham. 1998. Magnitude and significance of carbon burial in lakes, reservoirs, and peatlands. Geology 26(6): 535-538.

del Giorgio, P.A. and R.H. Peters. 1993. Balance between phytoplankton production and plankton respiration in lakes. Canadian Journal of Fisheries and Aquatic Sciences 50: 282-289.

del Giorgio, P.A. and R.H. Peters. 1994. Patterns in planktonic P:R ratios in lakes: Influence of lake trophy and dissolved organic carbon. Limnology and Oceanography 39(4): 772-787.

del Giorgio, P.A., D.F. Bird, Y.T. Prairie, and D. Planas. 1996. Flow cytometric determination of bacterial abundance in lake plankton with the green nucleic SYTO 13. Limnology and Oceanography 41(4): 783-789.

del Giorgio, P.A., J.J. Cole, and A. Cimbleris. 1997. Respiration rates in bacteria exceed phytoplankton production in unproductive aquatic systems. Nature 385: 148-151.

del Giorgio, P.A., J.J. Cole, N.F. Caraco, and R.H. Peters. 1999. Linking planktonic biomass and metabolism to net gas fluxes in northern temperate lakes. Ecological Society of America 80(4): 1422-1431.

Denward, M.T. and L.J. Tranvik. 1998. Effects of solar radiation on aquatic macrophyte litter decomposition. Oikos 82(1): 51-58.

Donahue, W.F., D.W. Schindler, S.J. Page, and M.P. Stainton. 1998. Acid induced changes in DOC quality in an experimental whole-lake manipulation. Environmental Science and Technology 32: 2954-2960.

Downing, J.A., Y.T. Prairie, J.J. Cole, C.M. Duarte, L.J. Tranvik, R.G. Striegl, W.H. McDowell, P. Kortelainen, N.F. Caraco, J.M. Melack, J.J. Middleburg. 2006. The global abundance and size distribution of lakes, ponds, and impoundments. Limnology and Oceanography 51(5): 2388-2397.

Duarte, C.M. and J. Cebrián. 1996. The fate of marine autotrophic production. Limnology and Oceanograophy 41(8): 1758-1766.

Duarte, C.M. and S. Agustí. 1998. The CO_2 balance of unproductive aquatic ecosystems. Science 281(5374): 234-236.

Duarte, C.M. and Y.T. Prairie. 2005. Prevalence of heterotrophy and atmospheric CO_2 emissions from aquatic ecosystems. Ecosystems 8: 862-870.

Eaton, A.D., L.S. Clesceri, and A.E. Greensberg. 1995. Standard Methods for the Examination of Water and Wastewater, 19th Edition. American Public Health Association, Washington, DC.

Emerson, S., C. Stump, and D. Nicholson. 2008. Net biological oxygen production in the ocean: Remote in situ measurements of O2 and N2 in surface waters. Global Biogeochemical Cycles 22, GB3023.

Enríquez, S., C.M. Duarte, and K. Sand-Jensen. 1993. Patterns in decomposition rates among photosynthetic organisms: the importance of detritus C:N:P content. Oecologia 94: 457-471.

Euliss, N.H. Jr., R.A. Gleason, A. Olness, R.L. McDougal, H.R. Murkin, R.D. Robarts, R.A. Bourbonniere, and B.G. Warner. 2006. North American prairie wetlands are important nonforested land-based carbon storage sites. Science of the Total Environment 361: 179-188.

Finlay, K., P.R. Leavitt, B. Wissel, and Y.T. Prairie. 2009. Regulation of spatial and temporal variability of carbon flux in six hard-water lakes of northern Great Plains. Limnology and Oceanography 54(6, part 2): 2553-2564.

Garvey, C.J., J. Keckes, I. Parker, M. Beilby, and G.S.H. Lee. 2006. Polymer nanoscale morphology in *Chara australis* Brown cell walls studied by advanced solid state techniques. Cryptogamie Algologie 27(4): 391-401.

Godshalk, G.L. and R.G. Wetzel. 1978a. Decomposition of aquatic angiosperms. I. Dissolved components. Aquatic Botany 5: 281-300.

Godshalk, G.L. and R.G. Wetzel. 1978b. Decomposition of aquatic angiosperms. II. Particulate components. Aquatic Botany 5: 301-327.

Godshalk, G.L. and R.G. Wetzel. 1978c. Decomposition of aquatic angiosperms. III. Zostera marina L. and a conceptual model of decomposition. Aquatic Botany 5: 329-354.

Golterman, H.L. 1995. The labyrinth of nutrient cycles and buffers in wetlands: results based on research in the Camargue (southern France). Hydrobiologia 315: 39-58.

Gorham, E., W.E. Dean and J.E. Sanger. 1983. The chemical composition of lakes in the north-central United States. Limnology and Oceanography 28(2): 287-301.

Goulder, R. 1969. Interactions between the rates of production of a freshwater macrophyte and phytoplankton in a pond. Oikos 20(2): 300-309.

Gran, G. 1952. Determination of the equivalence point in potentiometric titrations. Part II. Analyst 77: 661-671.

Gudasz, C., D. Bastviken, K. Steger, K. Premke, S. Sobek, and L.J. Tranvik. 2010. Temperature-controlled organic carbon mineralization in lake sediments. Nature Letters 466: 478-481.

Hagerthey, S.E., J.J. Cole, and D. Kilbane. 2010. Aquatic metabolism in the Everglades: Dominance of water column heterotrophy. Limnology and Oceanography 55(2): 653-666.

Hanson, P.C., D.L. Bade, S.R. Carpenter, T.K. Kratz. 2003. Lake metabolism: relationship with dissolved organic carbon and phosphorus. Limnology and Oceanography 48(3): 1112-1119.
Hanson, P.C., A.I. Pollard, D.L. Bade, K. Predick, S.R. Carpenter, and J.A. Foley. 2004. A model of carbon evasion and sedimentation in temperate lakes. Global Change Biology 10: 1285-1298.
Hessen, D.O., G.I. Ågren, T.R. Anderson, J.J. Elser, and P.C. de Ruiter. 2004. Carbon sequestration in ecosystems: the role of stoichiometry. Ecology 85(5): 1179-1192.
Hoover, T.E. and D.C. Berkshire. 1969. Effects of hydration on carbon dioxide exchange across and air-water interface. Journal of Geophysical Research 74(2): 456-464.
Houghton, R.A. 2007. Balancing the global carbon budget. Annual Review of Earth Planetary Science 35: 313-347.
Hutchinson, G.E. 1957. A Treatise on Limnology. Volume I. Geography, Physics and Chemistry. Wiley. 1015p.
Hutchinson, G.E. 1975. A Treatise on Limnology, Volume III. Limnological Botany. Wiley, New York, 660 pp.
Huttunen, J.T., J. Alm, A. Liikanen, S. Juutinen, T. Larmola, T. Hammar, J. Silvola, and P.J. Martikainen. 2003. Fluxes of methane, carbon dioxide and nitrous oxide in boreal lakes and potential anthropogenic effects on the aquatic greenhouse gas emissions. Chemosphere 52: 609-621.
Jähne, B., K.O. Münnich, R. Bösinger, A. Dutzi, W. Huber, and P. Libner. 1987. On the parameters influencing air-water gas exchange. Journal of Geophysical Research 92(C2): 1937-1949.
Kana, T.M., C. Darkangelo, M.D. Hunt, J.B. Oldham, G.E. Bennett, and J.C. Cornwell. 1994. Membrance inlet mass-spectrometer for rapid high-precision determination of N_2, O_2 and Ar in environmental water samples. Analytical Chemistry 66(23): 4166-4170.
Karim, A., J. Veizer, and J. Barth. 2008. Net ecosystem production in the great lakes basin and its implications for the North American missing carbon sink: A hydrologic and stable isotope approach. Global and Planetary Change 61: 15-27.
Kelly, C.A. E. Fee, P.S. Ramlal, J.W.M. Rudd, R.H. Hesslein, C. Anema, and E.U. Schindler. 2001. Natural variability of carbon dioxide and net epilimnetic production in the surface water of boreal lakes of different sizes. Limnology and Oceanography 46(5): 1054-1064.
Kenning, J. 2010. Dissertation, Chapter Two: Net autotrophy in shallow lakes exhibiting alternative stable states. University of Minnesota.
Kirchman, D., E. K'Nees, and R. Hodson. 1985. Leucine incorporation and its potential as a measure of protein synthesis by bacteria in natural aquatic systems. Applied and Environmental Microbiology 49(3): 599-607.
Kling, G.W., G.W. Kipphut, and M.C. Miller. 1991. Arctic lakes and streams as gas conduits to the atmosphere: implication for tundra carbon budgets. Science, New Series 251(4991): 298-301.
Kling, G.W., G.W. Kipphut, and M.C. Miller. 1992. The flux of CO_2 and CH_4 from lakes and rivers in arctic Alaska. Hydrobiologia 240: 23-36.

Kohfahl, C., G. Massmann, A. Pekdeger. 2009. Sources of oxygen flux in groundwater during induced bank filtration at a site in Berlin, Germany. Hydrogeology Journal 17: 571-578.

Kortelainen, P., M. Rantakari, J.T. Huttunen, T. Mattsson, J. Alm, S. Juutinen, T. Larmola, J. Silvola, and P.J. Martikainen. 2006. Sediment respiration and lake trophic state are important predictors of large CO_2 evasion from small boreal lakes. Global Change Biology 12: 1554-1567.

Kosten, S., F. Roland, D.M.L. Da Motta Marques, E.H. Van Nes, N. Mazzeo, L.S.L. Sternberg, M. Scheffer, and J.J. Cole. 2010. Climate-dependent CO_2 emissions from lakes. Global Biogeochemical Cycles 24, GB2007.

Koyama, M., J. Sugiyama, and T. Itoh. 1997. Systematic survey on crystalline features of algal celluloses. Cellulose 4(2): 147-160.

Kritzberg, E.S., J.J. Cole, M.L. Pace, W. Granéli, and D.L. Bade. 2004. Autochthonous versus allochthonous carbon sources of bacteria: results from whole-lake ^{13}C addition experiments. Limnology and Oceanography 49(2): 588-596.

Kufel, L. and I. Kufel. 2002. *Chara* beds acting as nutrient sinks in shallow lakes – a review. Aquatic Botany 72: 249-260.

Lauster, G.H., P.C. Hanson, and T.K. Kratz. 2006. Gross primary production and respiration differences among littoral and pelagic habitats in northern Wisconsin lakes. Canadian Journal of Fisheries and Aquatic Sciences 63: 1130-1141.

Leschine, S.B. 1995. Cellulose degradation in anaerobic environments. Annual Review of Microbiology 49: 399-426.

Liss, P.S., and L. Merlivat. 1986. Air-sea gas exchange rates: Introduction and synthesis, p. 113-127. *In* P. Baut-Menard [ed.], The role of air-sea exchange in geochemical cycling. D. Reidel.

López-Archilla, A.I., S. Mollá, M.C. Coleto, M.C. Guerrero, and C. Montes. 2004. Ecosystem metabolism in a Mediterranean shallow lake (Laguna de Santa Olalla, Donana National Park, SW Spain). Wetlands 24(4): 848-858.

Lovett, G.M., J.J. Cole, and M.L. Pace. 2006. Is net ecosystem production equal to ecosystem carbon accumulation? Ecosystems 9: 1-4.

MacIntyre, S., R. Wanninkhof, and J.P. Chanton. 1995. Trace gas exchange across the air-water interface in freshwaters and coastal marine environments, p. 52-97. *In* P.A. Matson and R.C. Harriss [eds.], Biogenic trace gases: Measuring emissions from soil and water. Blackwell.

McConnaughey, T.A., J.W. LaBaugh, D.O. Rosenberry, R.G. Striegl, M.M. Reddy, P.F. Schuster, and V. Carter. 1994. Carbon budget for a groundwater-fed lake: Calcification supports summer photosynthesis. Limnology and Oceanography 36(6): 1319-1332.

McConnaughey, T. 1998. Acid secretion, calcification, and photosynthetic carbon concentrating mechanisms. Canadian Journal of Botany 76: 1119-1126.

McKnight, D.M, E.W. Boyer, P.K. Westerhoff, P.T. Doran, T. Kulbe, and D.T. Andersen. 2001. Spectrofluorometric characterization of dissolved organic matter

for indication of precursor organic material and aromaticity. Limnology and Oceanography 46(1): 38-48.
Mihranyan, A. 2010. Cellulose from Cladophorales green algae: from environmental problem to high-tech composite materials. Journal of Applied Polymer Science 119(4): 2449-2460.
Mitchell, S.F. 1989. Primary production in a shallow eutrophic lake dominated alternately by phytoplankton and by submerged macrophytes. Aquatic Botany 33: 101-110.
Moran, M.A., R. Benner, and R.E. Hodson. 1989. Kinetics of microbial degradation of vascular plant material in two wetland ecosystems. Oecologia 79: 158-167.
Müller, B., Y. Wang, and B. Wehrli. 2006. Cycling of calcite in hard water lakes of different trophic states. Limnology and Oceanography 51(4): 1678-1688.
Nõges, P., L. Tuvikene, T. Feldmann, I. Tõnno, H. Künnap, H. Luup, J. Salujõe, and T. Nõges. 2003. The role of charophytes in increasing water transparency: a case study of two shallow lakes in Estonia. Hydrobiologia 506-509: 567-573.
Odum, H.T. 1956. Primary production in flowing waters. Limnology and Oceanography 1(2): 102-117.
Odum, W.E. and R.T. Prentki. 1978. Analysis of five North American lake ecosystems. IV. Allochthonous carbon inputs. Verhandlungen der Internationalen Vereinigung fur Limnologie 20: 574-580.
Otsuki, A. and R.G. Wetzel. 1974. Release of dissolved organic matter by autolysis of submersed macrophyte, *Scirpus subterminalis*. Limnology and Oceanography 19(5): 842-845.
Pace, M. and Prairie, Y.T. 2007. Respiration in lakes. *In* Respiration of Aquatic Ecosystems. *Eds.* P.A. del Giorgio and P.J. le B. Williams. Oxford University Press.
Prairie, Y.T., D.F. Bird, and J.J. Cole. 2002. The summer metabolic balance in the epilimnion of southeastern Quebec lakes. Limnology and Oceanography 47(1): 316-321.
Rantakari, M. and P. Kortelainen. 2005. Interannual variation and climatic regulation of the CO_2 emission from large boreal lakes. Global Change Biology 11: 1368-1380.
Raven, J.A. 2009. Contributions of anyoxygenic and oxygenic phototrophy and chemolithotrophy to carbon and oxygen fluxes in aquatic environments. Aquatic Microbial Ecology 56(2-3): 177-192.
Rejmánková, E. and K. Houdková. 2006. Wetland plant decomposition under different nutrient conditions: what is more important, litter quality or site quality? Biogeochemistry 80: 245-262.
Roehm, C.L., Y.T. Prairie, and P.A. del Giorgio. 2009. The pCO_2 dynamics in lakes in the boreal region of northern Québec, Canada. Global Biogeochemical Cycles 23, GB3013.
Rooney, N. and J. Kalff. 2003. Submerged macrophyte-bed effects on water-column phosphorus, chlorophyll *a*, and bacterial production. Ecosystems 6: 797-807.

Rørslett, B., D. Berge, and S.W. Johansen. 1986. Lake enrichment by submersed macrophytes: a Norwegian whole-lake experience with *Elodea Canadensis*. Aquatic Botany 26: 325-340.

Rosenberry, D.O., R.S. Striegl, and D.C. Hudson. 2000. Plants as indicators of focused ground water discharge to a northern Minnesota lake. Ground Water 38(2): 296-303.

Scheffer, M., S.H. Hosper, M-L. Meijer, B. Moss, and E. Jeppesen. 1993. Alternative equilibria in shallow lakes. Trends in Ecology & Evolution 8(8): 275-279.

Schlesinger, W.H. 1997. Carbon cycle of terrestrial ecosystems *in* Biogeochemistry: an Analysis of Global Change. 2nd Ed. Elsevier, Academic Press, CA, USA.

Sala, M.M. and H. Güde. 1999. Role of protozoans on the microbial ectoenzymatic activity during the degradation of macrophytes. Aquatic Microbial Ecology 20: 75-82.

Shilla, D., T. Asaeda, T. Fujino, and B. Sanderson. 2006. Decomposition of dominant submerged macrophytes: implications for nutrient release in Myall Lake, NSW, Australia. Wetlands Ecology and Management 14: 427-433.

Simon, M. and F. Azam. 1989. Protein content and protein synthesis rates of planktonic marine bacteria. Marine Ecology Progress Series 51: 201-213.

Sobek, S., G. Algesten, A.-K. Bergström, M. Jansson, and L.J. Tranvik. 2003. The catchment and climate regulation of pCO_2 in boreal lakes. Global Change Biology 9: 630-641.

Sobek, S., L.J. Tranvik, and J.J. Cole. 2005. Temperature independence of carbon dioxide supersaturation in global lakes. Global Biogeochemical Cycles 19: GB2003 p. 1-10.

Sobek, S., E. Durisch-Kaiser, R. Zurbrügg, N. Wongfun, M. Wessels, N. Pasche, and B. Wehrli. 2009. Organic carbon burial efficiency in lake sediments controlled by oxygen exposure time and sediment source. Limnology and Oceanography 54(6): 2243-2254.

Stets, E.G. and J.B. Cotner. 2008. Littoral zones as sources of biodegradable dissolved organic carbon in lakes. Canadian Journal of Fisheries and Aquatic Sciences 65: 2454-2460.

Stets, E.G., R.G. Striegl, G.R. Aiken, D.O. Rosenberry, and T.C. Winter. 2009. Hydrologic support of carbon dioxide flux revealed by whole-lake carbon budgets. Journal of Geophysical Reseach 114, G01008.

Stigebrandt, A. 1991. Computations of oxygen fluxes through the sea surface and the net production of organic matter with application to the Baltic and adjacent seas. Limnology and Oceanography 36(3): 444-454.

Stumm, W. and J.J. Morgan. 1981. Aquatic chemistry, 2nd edition. John Wiley & Sons, New York, 780 pp.

Theil-Nielsen, J. and M. Søndergaard. 1999. Production of epiphytic bacteria and bacterioplankton in three shallow lakes. Oikos 86(2): 283-292.

Tranvik, L.J., J.A. Downing, J.B. Cotner, S.A.Loiselle, R.G. Striegl, T.J. Ballatore, P. Dillon, K. Finlay, K. Fortino, L.B. Knoll, P.L. Kortelainen, T. Kutser, S. Larsen, I. Laurion, D.M. Leech, S.L. McCallister, D.M. McKnight, J.M. Melack, E.

Overholt, J.A. Porter, Y. Prairie, W.H. Renwick, F. Roland, B.S. Sherman, D.W. Schindler, S. Sobek, A. Tremblay, M.J. Vanni, A.M. Verschoor, E. von Wachenfeldt, and G.A. Weyhenmeyer. 2009. Lakes and reservoirs as regulators of carbon cycling and climate. Limnology and Oceanography 54(6, part2): 2298-2314.

Vadeboncouer, Y., J. Kalff, K. Christoffersen, and E. Jeppesen. 2006. Journal of North American Benthological Society. 25(2): 379-392.

van de Bogert, M.C., S.R. Carpenter, J.J. Cole, and M.J. Pace. 2007. Assessing pelagic and benthic metabolism using free water measurements. Limnology and Oceanography: Methods 5: 145-155.

Voros, L., B. Somogyi, and E. Boros. 2008. Birds cause net heterotrophy in shallow lakes. Acta Zoologica Academiae Scientiarum Hungaricae 54(Suppl. 1): 23-34.

Waiser, M.J. and R.D. Robarts. 2000. Changes in composition and reactivity of allochthonous DOM in a prairie saline lake. Limnology and Oceanography 45(4): 763-774.

Wanninkhof, R. 1992. Relationship between wind speed and gas exchange over the ocean. Journal of Geophysical Research 97(C5): 7373-7382.

Wanninkhof, R. and M. Knox. 1996. Chemical enhancement of CO2 exchange in natural waters. Limnology and Oceanography 41(4): 689-697.

Webster, J.R. and E.F. Benfield. 1986. Vascular plant breakdown in freshwater ecosystems. Annual Review of Ecological Systems 17: 567-594.

Webster, K.E, P.A. Soranno, K.S. Cheruvelil, M.T. Bremigan, J.A. Downing, P.D. Vaux, T.R. Asplund, L.C. Bacon, and J. Connor. 2008. An empirical evaluation of the nutrient-color paradigm for lakes. Limnology and Oceanography 53(3): 1137-1148.

Wehr, J.D., J. Petersen, and S. Findlay. 1999. Influence of three contrasting detrital carbon sources on planktonic bacterial metabolism in a mesotrophic lake. Microbial Ecology 37: 23-35.

Weishaar, J.L., G.R. Aiken, B.A. Bergamaschi, M.S. Fram, R. Fuji, and K. Mopper. 2003. Evaluation of specific ultraviolet absorbance as an indicator of the chemical composition and reactivity of dissolved organic carbon. Environmental Science and Technology 37: 4702-4708.

Welschmeyer, N.A. 1994. Fluorometric analysis of chlorophyll *a* in the presence of chlorophyll *b* and pheopigmnets. Limnology and Oceanography 39(8): 1985-1992.

Wetzel, R.G. 1992. Gradient-dominated ecosystems: sources and regulatory function of dissolved organic matter in freshwater ecosystems. Hydrobiologia 229: 181-198.

Wetzel, R.G. and M. Søndergaard. 1998. Role of submerged macrophytes for the microbial community and dynamics of dissolved organic carbon in aquatic ecosystems. In: E. Jeppesen, Ma. Søndergaard, Mo. Søndergaard, and K. Christoffersen (eds). The structuring role of submerged macrophytes in lakes. Springer-Verlag, New York, pp. 133-148.

Wetzel, R.G. 2001. Land-water interfaces: larger plants, p. 527-575. *In* Limnology Lake and River Ecosystems, 3rd ed. Academic Press.

Winter, T.C. and D.O. Rosenberry. 1995. The interaction of ground water with prairie pothole wetlands of the Cottonwood Lake Area, east-central North Dakota, 1979-1990. Wetlands 15(3): 193-211.
Wood, K.G. 1975. Photosynthesis of Cladophora in relation to light and CO2 limitation; CaCO3 precipitation. Ecology 56(2): 479-484.
Woodwell, G.M. and R.H. Whittaker. 1968. Production in terrestrial ecosystems. American Zoologist 8(1): 19-30.
Yavitt, J.B. 1997. Methane and carbon dioxide dynamics in Typha latifolia (L.) wetlands in central New York State. Wetlands 17(3): 394-406.
Zimmer, K.D., M.A. Hanson, M.G. Butler, and W.G. Duffy. 2001. Size distribution of aquatic invertebrates in two prairie wetlands, with and without fish, with implications for community production. Freshwater Biology 46: 1373-1386.
Zimmer, K.D., M.A. Hanson, B.R. Herwig, and M.L. Konsti. 2009. Thresholds and stability of alternative regimes in shallow prairie-parkland lakes of central North America. Ecosystems 12: 843-852.