

The impact of local macrophyte control on lake ecosystems

by

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Abstract

Macrophytes impact the structure and function of freshwater ecosystems but can also have an adverse impact on human activity. While the efficacy of macrophyte control methods are well-known, there are few studies on the environmental impacts of these treatments. In a pair of studies, the impacts of both mechanical cutting and the application of benthic barriers used in the control of the invasive *Myriophyllum spicatum* on the physical, chemical and biological components of four Ontario Lakes were investigated over four months. A Before-After-Control-Impact study design was employed, and three of the lakes were selected across a nutrient gradient. Observed impacts were minimal and not consistent across lakes. There were no significant differences in control-impact nutrient levels in any of the lakes. Nevertheless, some significant changes were observed for temperature, pH and zooplankton density. Changes to benthic macroinvertebrate and zooplankton community structure were observed in all four lakes.

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List of Acronyms/Measured Values

BACI	Before-After-Control-Impact (experimental design)
Chl- <i>a</i>	Chlorophyll- <i>a</i> (µg/L): a spectrophotometric estimate of algal biomass
cond	Conductivity (µS): a measure of ion concentration
CRCA	Cataraqui Conservation
den-z	zooplankton density (organisms/L)
den-i	benthic macroinvertebrate density (organisms/L)
DO	Dissolved oxygen (%)

DOC	Dissolved organic carbon (mg/L)
EMW	Eurasian watermilfoil (<i>Myriophyllum spicatum</i>)
HABs	Harmful Algal Blooms
light	Light extinction (% of light detected at surface reaching bottom; ~ 2m)
MALLA	Malcom and Ardoch Lakes Landowners' Association
MVCA	Mississippi Valley Conservation Authority
PC	Parks Canada
PCA	Principal Component Analysis (an ordination technique)
pH	power of hydrogen ($-\log[\text{mol/L}]$)
PVI	Percent Volume Inhabited (how much % of water column plants occupy)
RVCA	Rideau Valley Conservation Authority
T	Temperature (°C)
TKN	Total Kjeldahl Nitrogen (mg/L)
TP	Total Phosphorus (mg/L)
z	depth (m)

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Chapter 1: Introduction

Macrophytes are crucial components of aquatic ecosystems and play an important role in the structure and function within these ecosystems (Carpenter and Lodge 1986; Jeppesen et al. 1998). Conversely, large, dense beds of macrophytes are often seen as a nuisance by lake and river users, as they can reduce the aesthetic quality of the water body, hinder water transport and reduce the habitat quality of desirable fish species (Kalff 2003). Eurasian watermilfoil, *Myriophyllum spicatum*, is a well-documented invasive macrophyte that was introduced to North America in the late 19th century (Aiken et al. 1979; Nichols and Shaw 1986; Smith and Barko 1990; Madsen et al. 1991). Due to its ability to proliferate quickly and dominate the littoral zones of the lakes it colonizes it has since spread across the continent. Economic impacts have been associated with this spread, both from reduced property values (Eiswerth et al. 2000a; Goodenberger and Klaiber 2016) and costly management techniques (reviewed in Lovell and Stone, 2005; Pimentel et al., 2005). These techniques include biological control, mechanical or manual harvesting, shading out with large benthic barriers or the application of waterborne herbicides. In Canada, macrophyte control is a relatively subdued affair, and in Eastern Ontario, macrophyte control is mostly small scale and carried out by landowners with lakefront property. Landowners are able to do such work on crown land without a permit so long as the disturbance is kept small and directly in front of their property. A few other criteria must be met (such as not using herbicides, acting within a prescribed time window and disposing of plants on land), and landowners are urged to check with their local watershed authority before acting (Government of Ontario 2017). Also, in some cases, the waterbody may be managed by a separate regulator, such as Parks Canada

along the Rideau Canal (Parks Canada 2017). Nevertheless, one of the driving forces behind this study is to emulate macrophyte control as it is most commonly carried out: small scale and waterfront in an Eastern Ontario waterbody.

Also, as macrophyte control exists on the interstices of physical and human geography, it is important to acknowledge that this study is in many ways shaped by human interactions with the environment. Controlled macrophytes, especially invasives such as *M. spicatum*, are often described with negative language – they are called ‘weeds’ and the area where they grow referred to as ‘infested.’ Members of Lake associations, often owners of property in impacted areas, can be adversely affected by these organisms, spending time and money combatting them. An important aspect of this project is to ensure respectful engagement with such stakeholders. Indeed, these persons are often responsible for initiating and promoting projects such as this.

While literature related to the efficacy of various management techniques exists (Murphy 1988; Madsen 2000; Hussner et al. 2017), very little is known about the impacts these methods have on aquatic ecosystems. There are rising concerns in the aquatic plant management community that the removal of freshwater macrophytes, even invasives, may have unforeseen consequences for freshwater ecosystems (Chambers et al. 1999; Nes et al. 1999; Kuiper et al. 2017; Radomski and Perleberg 2019), particularly in regards to nutrient dynamics. Any threats to nutrient dynamics clearly must be taken seriously from a management perspective, as the consequences of nutrient loading are very well known (Schindler 1974, 1977), especially in regards to eutrophication and harmful algal blooms.

At the crux of nutrient dynamics in freshwater lakes, in particular shallow ones, is the concept of alternating stable states: a macrophyte-dominated clear water state and a phytoplankton-dominated turbid state (May 1977; Scheffer 1990; Scheffer and Carpenter 2003; Scheffer and Jeppesen 2007). The low-nutrient, clear water systems are thought of as being more resilient (Valley and Drake 2007; Donadi et al. 2018); more able to recover from a disturbance and avoid transitioning to another state (Holling 1973; Peterson et al. 1998; Folke et al. 2004). Work in Europe regarding lake restoration from the latter state to the former has been shown to be costly, challenging and complex (Baron et al. 2005; Bakker et al. 2013; Søndergaard et al. 2016; Hilt et al. 2017), and thus any activity that could cause a potential shift to a phytoplankton-dominated state should be undertaken with great caution. It is thought that mechanical cutting could contribute to such a shift by disturbing sediment and re-suspending it in the water column, which has certainly been observed before (Carpenter and Gasith 1978; Zutshi and Ticku 1990; Morris et al. 2006). Also, the violent cutting action could easily dislodge and suspend periphyton growing on the plants themselves, further enriching the water (Cattaneo and Kalff 1980). The removal of the plants, by both cutting and smothering with benthic barriers would also re-expose the sediment to wave action and resuspension in the littoral, a well-known mechanism of internal loading (Horppila and Nurminen 2005; Holmroos et al. 2015). For benthic barriers, another concern is the decomposition of the plants beneath the barrier and a re-release of phosphorous during this process (Boston and Perkins 1982). Lastly, it has been shown in a laboratory setting that *M. spicatum* releases phytoplankton-inhibiting allelochemicals (Gross et al. 1996; Nakai et al. 2000; van Donk and van de Bund 2002; Zhu et al. 2010) and so removing plants is akin to removing the guardians of the clear-

water state (Meerhoff et al. 2007). Scale plays a role in the level and nature of the impacts, but recent studies by van de Leemput et al. (2018) suggest that even small disturbances can have an impact on alternating stable states, and that the chance of achieving a critical transition could be easier in an already stressed and less resilient system (Scheffer et al. 2012; van de Leemput et al. 2015).

As the disturbances proposed in this thesis will be small, it is important that as many abiotic and biotic parameters as possible be observed for changes. In addition to traditional limnological abiotic indicators such as acidity, conductivity, nutrients, oxygen and temperature, biotic indicators must also be considered. Moreover, the composition of these biotic communities is of crucial concern. Thus, macroinvertebrates and zooplankton, with their central roles in aquatic food webs, are well positioned to act as responding variables.

Given the lack of data on the impacts of macrophyte control, and the potential link to nutrient dynamics and alternating stable states, two studies were undertaken as part of this thesis. The first was to determine the impacts, if any, of the application of burlap benthic barriers used in the control of *M. spicatum* on the local physical, chemical and biological properties of Malcolm Lake, Ontario over a four-month period. The second study was to assess, also over a four-month period, the physical, chemical and biological impacts of the mechanical cutting of small *M. spicatum* patches in three Eastern Ontario Lakes. Furthermore, this second study was conducted along a nutrient gradient, from oligotrophic to eutrophic. It was anticipated that any impacts observed would be slowest to recover in the most eutrophic lake and quickest to recover in the most oligotrophic lake.

1.1 Thesis Structure

This thesis comprises of two independent but thematically linked studies, which will hereafter be referred to as ‘study 1’ and ‘study 2’ (or ‘paper 1’ and ‘paper 2’; ‘first study’ and ‘second study’; ‘Malcolm study’ and ‘Rideau study’). The first study is a study of the impacts of benthic barriers used in the control of *M. spicatum* in Malcolm Lake, Ontario. The second study is a study of the impacts of mechanical cutting of *M. spicatum* on three Rideau Lakes in Eastern Ontario. Apart from the treatments and study sites, the methods are largely similar, and are only dissimilar where specifically indicated. Results, discussions and conclusions are first presented independently, and then together. The studies will be submitted for publication separately as distinct papers.

1.2 Research Question and Objectives

Research Question: What impact does local macrophyte control have on the chemical, physical and biological components of lake ecosystems?

Study 1 (Malcolm Lake) - The impact of burlap benthic barriers used in macrophyte control on local water quality and biological community composition in Malcolm Lake, Ontario, Canada

Objectives:

(1a) Determine the localized impacts of macrophyte suppression by the application of benthic barriers on the physical (temperature - T, light extinction - light), chemical

(conductivity - cond, dissolved oxygen - DO, dissolved organic carbon - DOC, pH, total Kjeldahl nitrogen - TKN, total phosphorous - TP) and biological properties (chlorophyll-*a* – chl-*a*, zooplankton density – den-z) within Malcolm Lake.

(1b) Determine the impacts of macrophyte suppression by the application of benthic barriers on individual zooplankton taxa and overall community composition.

Study 2 (Rideau) - The impact of macrophyte control by means of mechanical cutting on lacustrine ecosystems along a nutrient gradient in the Historic Rideau Waterway, Ontario, Canada

Objectives:

(2a) Determine the localized impacts of macrophyte removal by mechanical cutting on the physical (T, light), chemical (cond, DO, DOC, pH, TKN, TP) and biological properties (Chl-*a*, den-z, macroinvertebrate density - den-i) within Dog, Otter and Wolfe Lakes.

(2b) Determine the impacts of macrophyte removal by mechanical cutting on individual zooplankton taxa and overall community composition.

(2c) Determine the impacts of macrophyte removal by mechanical cutting on individual benthic macroinvertebrate taxa and overall community composition.

(2d) Determine if lakes with greater total phosphorus concentrations display greater deviations from mean chemical, physical and biological conditions (as in 2a) when a disturbance (mechanical cutting) is introduced.

Chapter 2: Literature Review

2.1 Ecological importance of macrophytes in freshwater ecosystems

Water is necessary for all living things on earth. Humans are no exception, and our use of water goes beyond the simple sustenance provided by drinking water: we use water for sanitation, transportation, irrigation, power generation, recreation and more. Most of these uses rely on freshwater, which confounds any anthropogenic impacts, as fresh water makes up such a small portion of water on the planet (less than 3%), and of this freshwater, only around one fiftieth is present in easily accessible lakes and rivers (Dodds and Whiles 2010). According to the Millennium Ecosystem Assessment (2003), the world's inland freshwaters are under direct and increasing threat. Among the biggest threats to freshwater resources are the introduction of invasive species and nutrient enrichment from nitrogen and phosphorous. This study will investigate the role of an invasive macrophyte, in particular its control, on ecosystem health, within the framework of nutrient enrichment.

Macrophytes are large aquatic plants from multiple taxa found in the shallow (littoral) parts of lakes, as they can only grow where there is sufficient light for photosynthesis (Kalff 2003). Macrophytes are important primary producers in lakes, especially in shallow lakes where they can grow throughout the lake and effectively outcompete phytoplankton (Cattaneo and Kalff 1980). They are also important in nutrient cycling in lakes (Duarte 1995), and provide a connection between nutrients in the water and in the sediments (Carignan 1985; Barko and Smart 1986). They provide refuge for invertebrates, zooplankton and small fish (Schriver et al. 1995), and a substrate upon

which epiphytic algae and bacteria (biofilm) can grow (Sand-Jensen and Borum 1991). The importance of macrophytes is even greater in wetlands, which in many ways are an extension of shallow lakes and rivers (Kalff 2003). Macrophytes have been shown to enhance ecosystem services by effectively filtering wastewater and preventing the resuspension of nutrients (Engelhardt and Ritchie 2001), and also by absorbing harmful metals from polluted waters (Lesage et al. 2007). Macrophytes are central components to aquatic ecosystems, having multiple pathways of influence from, and on, abiotic components below them, and biotic components both above and below them (Figure 2-1).

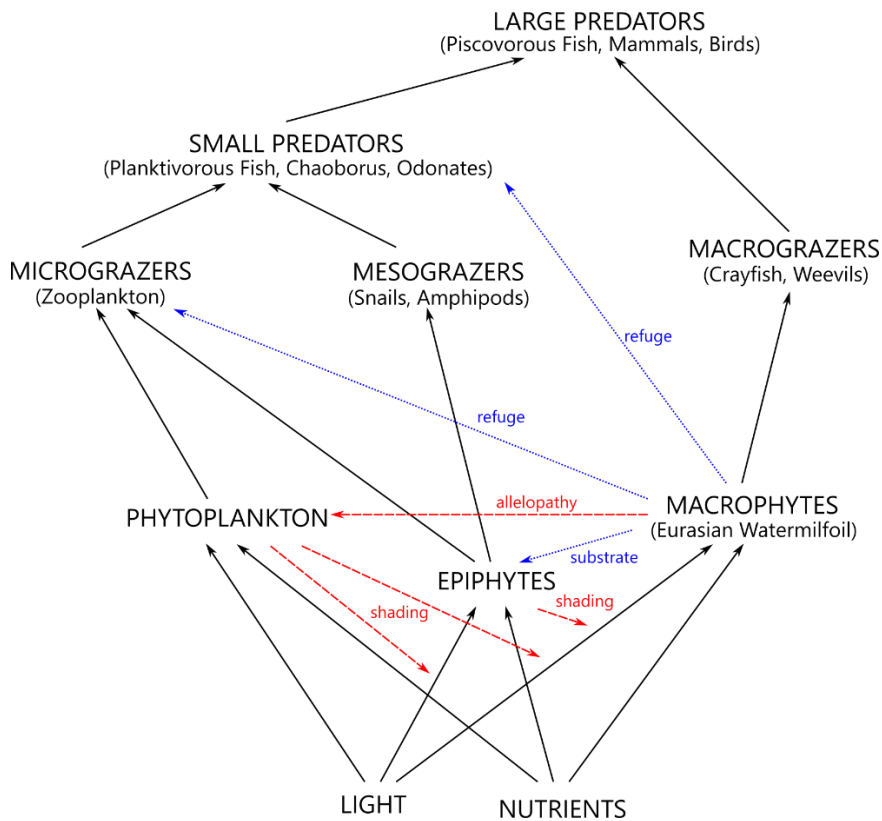


Figure 2-1. Conceptual diagram of how macrophytes influence lacustrine ecosystems. Black arrows show trophic interactions, while blue arrows show positive interactions (such as macrophytes providing a daytime predation refuge for microcrustacean grazers of phytoplankton) and red arrows negative interactions (such as phytoplankton shading out light required for epiphyton growth). Adapted from (Jeppesen et al. 1997).

2.2 Eurasian watermilfoil: an invasive species

M. spicatum (Figure 2-2), is an invasive aquatic angiosperm that was introduced to North America as early as the 1880's (Nichols and Shaw 1986). It exists as a submerged macrophyte in waters up to 10 m deep, but more commonly between 1 m and 3 m in depth (Aiken et al. 1979). *M. spicatum* is a rooted macrophyte that typically grows from the bottom substrate right to the water surface where its flowering spike can be exposed. It can tolerate a wide range of acidity, salinity, temperature, substrate and nutrient conditions, making it an excellent competitor. While the phenology of *M. spicatum* can vary broadly based on climate and location, it typically commences growth very early in the growing season and is one of the last macrophytes to senesce in the fall. While it does reproduce sexually, it reproduces far more readily by asexual auto-fragmentation, whereby small buds detach from the root crowns and establish new plants very early in the growing season (Aiken et al. 1979).

M. spicatum is notorious for forming dense beds, and for outcompeting native macrophytes, including native watermilfoil species (Aiken et al. 1979). It is also notoriously difficult to control and manage, as it grows vegetatively from even the smallest fragments, which can easily be transported on boats and other equipment to infest other lakes (Aiken et al. 1979). The presence of *M. spicatum* in lakes is associated with a direct decline of both macrophyte biodiversity (Boylen et al. 1999) and macroinvertebrate biodiversity (Wilson and Ricciardi 2009), and a subsequent indirect decline in some fish populations (Keast 1984). The weed is considered harmful to water transport and recreation, with quantifiable economic repercussions (Eiswerth et al.

2000b), prompting many lake associations and conservation authorities to opt for management and control.

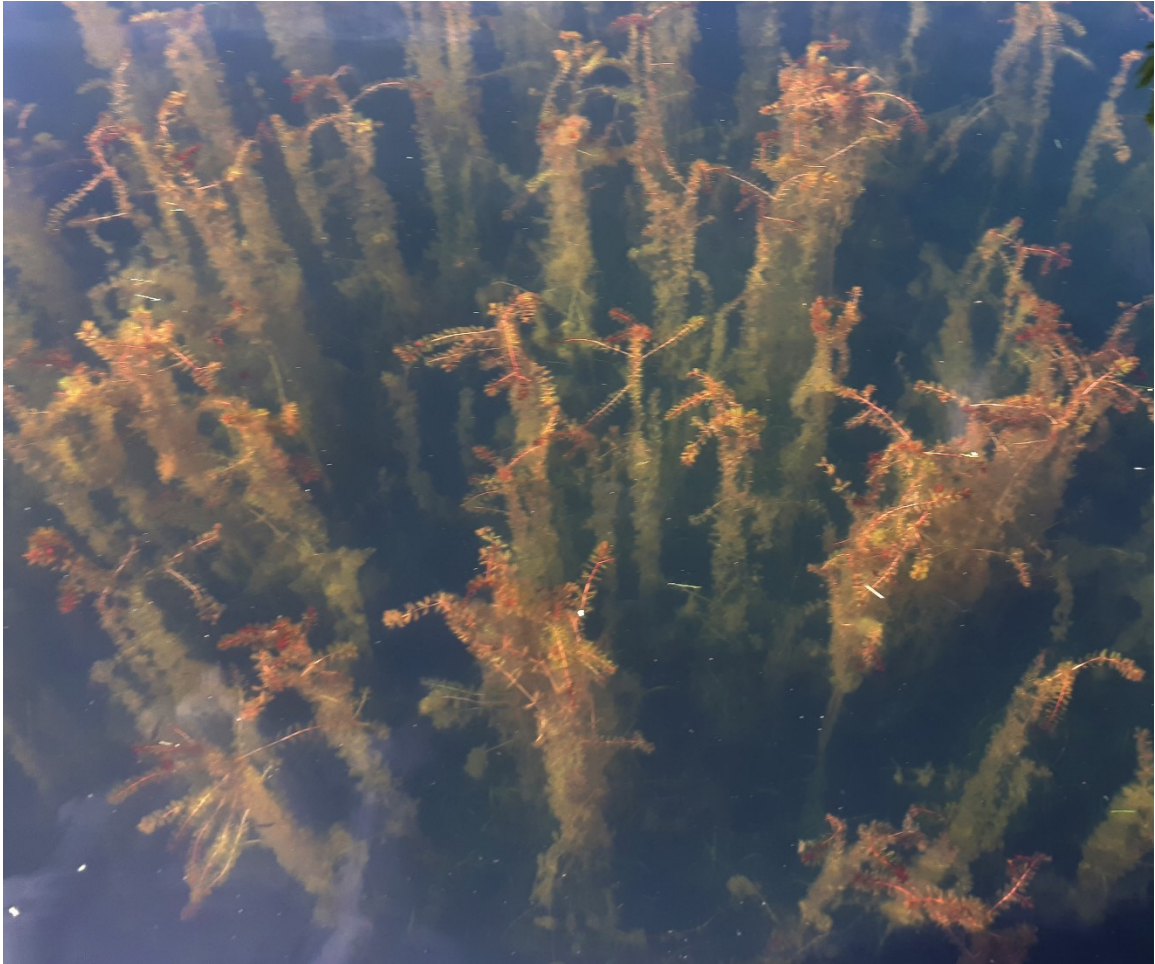


Figure 2-2. Eurasian watermilfoil (*Myriophyllum spicatum*), an invasive submerged macrophyte, here seen growing right to the surface of the water ($z = 2$ m) on Dog Lake on July 10th, 2019.

2.3 Macrophyte control and impacts thereof

There are several ways in which lake managers and property owners control macrophytes (Madsen 2000). The simplest method is mechanical harvesting, which can range from hand-picking by divers to the use of specialized harvesters (Figure 2-3).

While mechanical harvesters are effective at cutting the plants, they are associated with

high machine costs (Kalff 2003) and are seen by some as only a temporary solution due to subsequent regrowth (Kimbel and Carpenter 1981). A second method of control is the application of waterborne herbicides such as diquat dibromide (Sesin et al. 2018),



Figure 2-3. Rideau Aquatic’s custom harvester barge is an example of a mechanical cutting system used in macrophyte control. Here the conveyor and sickle are deployed and are cutting a swathe through an *M. spicatum* patch on Otter Lake in September 2019.

which may be more effective and cheaper than mechanical harvesters, but which are discouraged due to health and environmental concerns in multi-use lakes and still are not always able to eradicate roots and prevent regrowth (Kalff 2003). A third method of control is biocontrol, in which herbivorous macrophyte grazers are introduced to the system. Biocontrol in general has a mixed track record, as introducing large numbers of

an organism to an ecosystem can have unforeseen consequences, although it is not without its successes (Carson et al. 2008). For *M. spicatum* management in Ontario, small aquatic weevils that are natural grazers of *Myriophyllum* spp. were tested as a biocontrol agent. While this method showed some initial promise, it is beginning to seem like it too is relatively ineffective, and also very expensive (Cooper et al. 2017). Another method of macrophyte control involves deliberate and complete shading with benthic barriers; either fiberglass screens or burlap mats that are laid over top of entire beds (Figure 2-4). This is typically a localized technique, but it is currently being considered in many Canadian locations.



Figure 2-4. Volunteer members of MALLA use personal watercraft to deploy a burlap benthic barrier from a large spool onto a patch of *M. spicatum* on Malcolm Lake in July 2019. Rebar is used to hold the mat down initially (with foam floaters for easy retrieval) and locally sourced river stone thereafter.

The use of benthic barriers to shade out and smother nuisance macrophytes has been a control option for several decades (Mayer 1978; Jones and Cooke 1984; Helsel et al. 1996; Madsen 2000), but high costs associated with larger infestations and concerns about gas evolution (from bacterial metabolism) under synthetic fabrics (Mayer 1978; Gunnison and Barko 1992) and the complete eradication of benthic fauna (Ussery et al. 1997) led to the rise in prominence of biodegradable burlap benthic barriers (Jones and Cooke 1984; Caffrey et al. 2010; Hofstra and Clayton 2012). In a Canadian context, biodegradable burlap benthic barriers present a novel opportunity for macrophyte control due to strict regulations on the use of herbicides and even the difficulty of obtaining permits for localized mechanical control (Minister of Justice 2017; Parks Canada 2017). Even when the fabrics used are biodegradable, the decomposition of plant matter still presents a concern both in terms of nutrient release as well as oxygen depletion. As a matter of fact, the depletion of oxygen is a concern in any case when plant matter is left behind, whether it be by benthic barriers, herbicide use (Carpenter and Greenlee 1981) or mechanical cutting (Abernethy et al. 1996).

Nevertheless, cutting remains one of the more popular methods of macrophyte control in Canada, although numbers are difficult to estimate as records are only now starting to be kept by regulators. One advantage of cutting is that plant biomass, a potential source of phosphorus for internal loading, can be removed from the lake (Carpenter and Adams 1977, 1978). Indeed, this removed biomass could even have a potential use as fertilizer (Ban et al. 2019). Conversely, it is also possible that removing plant matter would possibly mean the removal of rarer nutrients from the lake ecosystem, hampering recolonization by native species (Carpenter and Lodge 1986). Furthermore,

while cutting has generally been shown to be a temporary solution, with plants regrowing in as little as 4 weeks, it has also been shown on subsequent mowings that *M. spicatum* will have reduced biomass after regrowth (Carpenter and Gasith 1978; Kimbel and Carpenter 1981; Abernethy et al. 1996). There is a danger associated with cutting in regards to fragments of cut plants re-establishing and colonizing new areas (Aiken et al. 1979; Eiswaerth et al. 2000a), although this can generally be avoided by careful equipment operators. Early work by Carpenter and Gasith (1978) showed that smaller cutting treatments had little to no lasting impact on dissolved organic carbon, dissolved phosphorus, temperature and conductivity. Conversely, work by Zutshi and Ticku (1990) showed that if mechanical cutting were deployed on a much larger scale (square kilometres instead of square metres), that there were significant changes to conductivity, dissolved oxygen, total phosphorus and total nitrogen, as well as significant increases to diatoms, cyanobacteria, rotifers and zooplankton densities. These changes reflect a move towards a more turbid, phytoplankton-dominated state. Recent modelling with PCLake (a mathematical model developed by the Netherlands Environmental Assessment Agency) done by Kuiper et al. (2017) suggests that mowing could be a safe method for oligotrophic or mesotrophic lakes, but that in eutrophic lakes there is a danger of adversely impacting nutrient dynamics, again by shifting to phytoplankton dominance.

In general, there are plenty of resources pointing to the efficacy of many management techniques, but rather few in regards to the impacts these techniques have on lake ecosystem themselves. Indeed, in a recent comprehensive review of macrophyte control by Hussner et al. (2017), the small section on ecosystem impacts has but 7 references - this in a paper with 359 references. Thanks to restoration work being

conducted in European lakes (van Nes et al. 2002), it is now suspected that macrophyte control is strongly tied to eutrophication and plant-phytoplankton dynamics. When plants are removed, lakes are in greater danger of shifting towards phytoplankton dominance, and when they are restored, this shift can be reversed.

2.4 Eutrophication, alternating stable states and ecosystem resilience.

The phytoplankton, or algae, are a taxonomically diverse group of photosynthetic plankton (small, free-floating aquatic microorganisms) inhabiting the surface and near-surface open-water (pelagic) regions of lakes and rivers (Wetzel 2001). Phytoplankton play a very important role in light attenuation and nutrient-cycling in freshwater ecosystems, and as a result have been intensely studied by limnologists (Wetzel 2001). The concentration of phytoplankton can increase dramatically under high nutrient concentrations, particularly phosphorous (Schindler 1974). Eutrophic lakes are lakes with very high nutrient levels and are dominated by algae (Hutchinson 1957). The resulting blocking out of light by the excessive phytoplankton growth seriously reduces or even eliminates the presence of rooted macrophytes in lake ecosystems (Sand-Jensen and Borum 1991). For this reason, it has been proposed that shallow lakes tend to exist in one of two alternate stable states: a clear-water, macrophyte-dominated state, or a turbid, phytoplankton-dominated state (Scheffer 2001) (Figure 2-5).

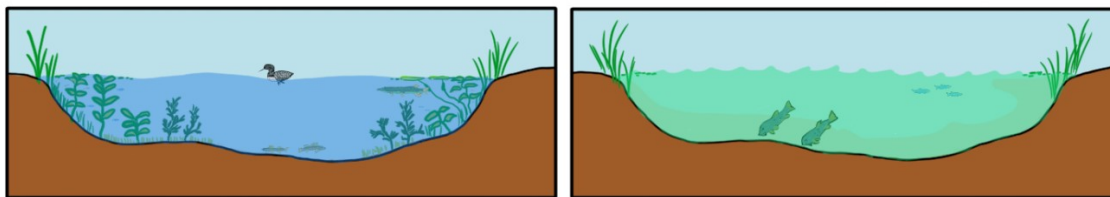


Figure 2-5. Conceptual diagram of alternating stable states in shallow lakes. The lake on the left is in a clear-water, macrophyte-dominated state. Top-down pressure and adequate refugia are able to keep planktivorous fish in check, reducing pressure on algae-grazing zooplankton. The lake on the right is in a turbid, phytoplankton-dominated state. Planktivorous fish keep grazing zooplankton numbers low, and wind, waves and fish stir up sediment and resuspend nutrients that allow for high algal levels, suppressing macrophyte growth. Adapted from Scheffer (2001).

While lake managers in Canada are pondering the removal of macrophytes, researchers in Europe are desperately trying to coax them back into their lakes (van Nes et al. 2002). The lakes in Europe were altered from their natural states long ago, and after centuries of agricultural and urban runoff, the amount of nutrients, namely phosphorous and nitrogen, are generally very high. Researchers in Europe have tracked the decline of macrophytes over expanses of time sometimes as long as a century (Sand-Jensen et al. 2000), and it is clear that excessive nutrients cause the extinction of macrophytes in favour of free-floating phytoplankton (Pieczynska et al. 1988). Similar studies in some parts of the United States have shown the same effects (Sayer et al. 2010). From Schindler's seminal work in the Experimental Lakes Area of Northwestern Ontario (1974), limnologists now know that enrichment by phosphorus is the leading cause of eutrophication in lakes. Phosphorus is the limiting nutrient for phytoplankton growth, and when present in sufficient quantity, allows for the formation of algal blooms (Dodds and Whiles 2010). The increased turbidity caused by the high concentration of phytoplankton extinguishes light below, first eliminating the deeper submerged macrophytes, and

eventually all but some floating and emergent macrophytes (Dodds and Whiles 2010). Nutrient enrichment has also been shown to increase the growth of periphytic algae, a coating or biofilm on plants that also directly blocks light and helps accelerate the decline of macrophytes (Phillips et al. 1978).

Interestingly, some studies show that moderate eutrophication at first allows *M. spicatum* to move into shallower waters and outcompete native species (Egertson et al. 2004), as its ability to form dense beds can actually buffer it from the effects of eutrophication (Donadi et al. 2018). Other studies have shown in the laboratory that *M. spicatum* can actually inhibit algal growth by releasing allelopathic chemicals into the water column (Nakai et al. 2000). Further studies on trophic interactions have examined the influence of zooplankton (Jeppesen et al. 1999) and fish (Jeppesen et al. 1990), and on the importance of nutrients liberated from sediments by roots (Carignan 1982). Due to their ability to produce toxins, algal blooms are several orders of magnitude more detrimental to human and ecosystem health when compared with the nuisance caused by dense macrophyte beds, and thus it is imminently necessary to consider any potential links to eutrophication in all forms of lake management (Millennium Ecosystem Assessment (Program) 2005). From European and American lake managers, we have learned that the restoration of turbid, nutrient-enriched and phytoplankton-dominated lakes is both difficult (Bakker et al. 2013), slow (Sand-Jensen et al. 2017) and complex (Vejříková et al. 2018). One proposed reason for this difficulty is related to the theoretical concept of ecosystem resilience, and its ancillary concepts of catastrophic regime shifts and critical slowing down.

Ecosystem resilience, a concept first introduced by C.S. Holling (1973), is defined as the ability of an ecosystem to resist and recover from a disturbance. It is theorized that as ecosystems become less resilient that it becomes easier for them to be pushed from one stable state to another, and more difficult for them to recover (Scheffer et al. 2001) (Figure 2-6). This concept is often illustrated using the example of macrophyte-dominated vs. phytoplankton-dominated lakes (Scheffer and Jeppesen 2007). While there are theoretical and mathematical models that describe alternate stable states in shallow lakes, a call for more experimental field studies has been made (van de Leemput et al. 2018). Therefore, this project will not only help address the lack of data on the potential impacts of *M. spicatum* control on lake ecosystems, but also provide insight on ecosystem resilience and regime shifts by tracking the difference in response to induced disturbances along a nutrient gradient.

It has been theorized that ecosystems can have more than one stable state (May 1977), and that ecosystems have a level of resilience that allows them to recover to their most stable state following a disturbance (Holling 1973). This concept has been applied to shallow lakes (under similar external ecological conditions) to suggest that they can occur in two alternate stable states: a clear-water, low-nutrient, macrophyte-dominated state and a turbid-water, high-nutrient, phytoplankton-dominated state (Scheffer and Carpenter 2003). The transition from one of these states to the other can be rapid and abrupt, a tipping-point of sorts, and referred to in the literature as a critical transition or catastrophic regime shift (Scheffer et al. 2001). Resource managers would clearly benefit from knowing how close an ecosystem is to such a tipping point, and thus much work,

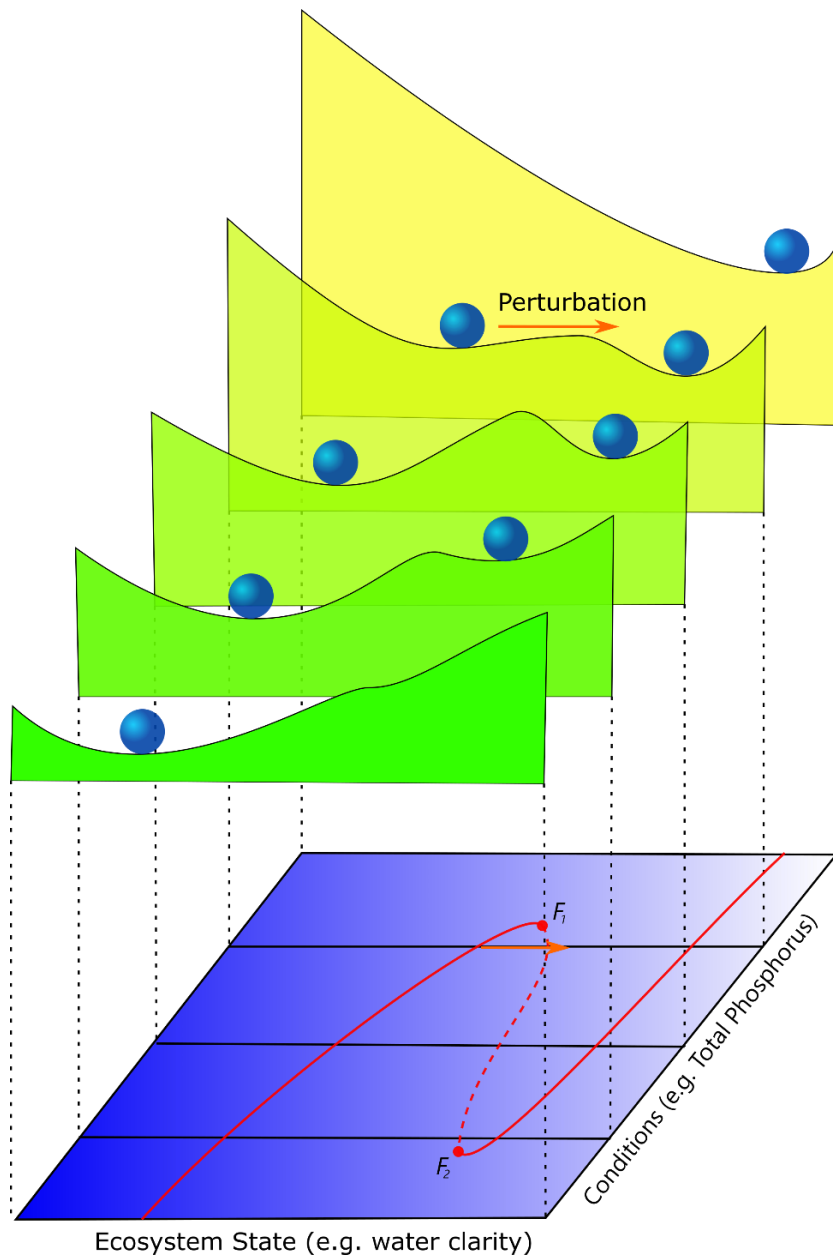


Figure 2-6. The ball in cup model is frequently used in explaining the concepts of alternating stable states in a shallow lake. In a resilient, clear-water ecosystem with only one stable state, while any perturbation will cause the ball roll up the cup wall, it will have no choice but to roll back down into the only available basin of attraction and return the ecosystem to its initial state (the first green panel). At the other extreme (the back-most yellow panel) no matter what the perturbation, the ball will always come to rest in the turbid water state that is its only available stable state. The three middle states represent systems that can exist in two separate stable states, clear or turbid, with the lower-most basin of attraction indicating a preferred state. A big enough perturbation can move the ball from one basin of attraction to another. Adapted from Scheffer (2001).

most of it theoretical, but some experimental, has been dedicated to looking for early warning signs for these regime shifts (Scheffer et al. 2009). Mathematical models, such as those developed by van de Leemput et al. (2018) and van Nes and others (2007) seem to point at the concept of critical slowing down (CSD), or slow recovery, as being indicative of a loss of resilience as an ecosystem approaches a tipping point. Detecting critical slowing down has proven to be challenging, even within models, as the signals of rising temporal autocorrelation and variance are sometimes not even present (Dakos et al. 2015). While abrupt changes of state can occur close to bifurcation points (Figure 2-6), it is very difficult to measure the right variable at the right scale (Dakos et al. 2015). Nevertheless, one whole lake experiment in 2011 was able to show some early warning signs after a disturbance was introduced to a lake (Carpenter et al. 2011a); the introduction of a top predator was able to cause a trophic cascade, and rising autocorrelation was observed in measured algal biomass. One laboratory experiment where the artificial environment of *Daphnia* was deliberately and iteratively degraded was able to show statistical signatures of critical slowing down before a forced bifurcation point was reached (Drake and Griffen 2010). There is clearly a paucity of experimental evidence for these theories, and more than one review has called for intensified studies designed around the concept of regime shifts (Scheffer and Carpenter 2003; van de Leemput et al. 2018).

2.5 *Before-After-Control-Impact* experiments, theory and practice

Impact assessments are used to determine if a stressor, often human-caused, has changed the environment, usually adversely (Smith 2006). Before-after-control-impact (BACI) study design is a form of impact assessment commonly used by researchers in order to detect such changes to the environment. While the model was originally developed by Green (1979) it has subsequently been refined iteratively into the many variations that exist today (Stewart-Oaten et al. 1986; Underwood 1991; Smokorowski and Randall 2017). The basic concept is relatively simple (Smith 2006): an environmental parameter, abiotic or biotic, is measured in two (usually more, explained below) similar locations, one designated ‘control’ and one designated as ‘impact’ (this term is much un-loved, as this site is as of yet to be treated! Nevertheless, it is still in very common use). Both of these locations are also designated as the ‘before’ period. Then, a disturbance is introduced; usually some form of human-caused impact to the environment, such as a construction site. After this disturbance (or treatment), the same parameter is measured in the ‘control’ location (unimpacted by the disturbance) and the ‘impact’ location. These measurements make up the ‘after’ period. The control-impact differences are then compared for before and after the disturbance. An environmental impact is detected if a) there is no significant difference between the control and impact pre-treatment and b) the control and impact sites are significantly different after the treatment (Figure 2-7).

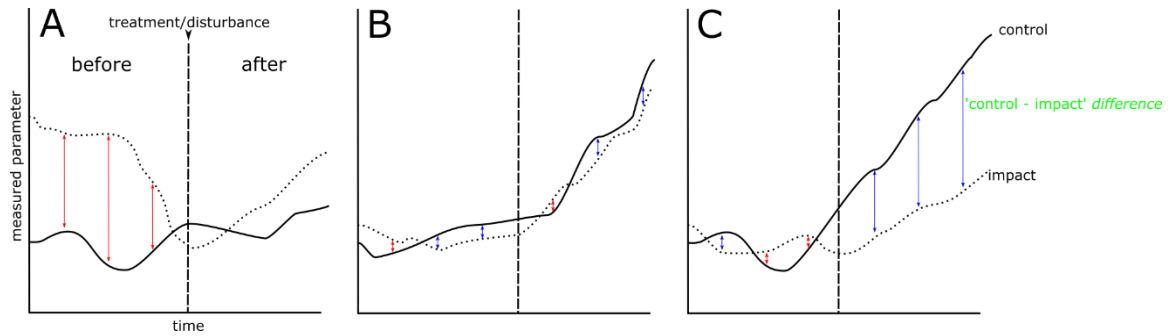


Figure 2-7. BACI (before-after-control-impact) design. In all three examples (A, B and C), a hypothetical parameter is measured at a designated control site (solid line) and impact site (dotted line) ‘before’ and ‘after’ a disturbance is introduced (separated by the vertical dashed line). The red arrows represent negative ‘control – impact’ differences, and the blue arrows represent positive ‘control – impact’ differences. In the case of (A) there is too much natural variability between the ‘control’ and (as-of-yet untreated) ‘impact’ sites, as seen by the large pre-disturbance difference arrows, and thus no post-treatment comparisons can be made. In the second example (B), the post-treatment ‘control-impact’ differences, despite both having an upward seasonal trend, are not significantly different from the pre-treatment differences, suggesting that the disturbance had no effect on the measured parameter. In the third example, the post-treatment, or ‘after’ ‘control-impact’ differences are clearly greater than the ‘before’ differences, suggesting that the disturbance had an impact on the measured parameter.

Traditionally, BACI experiments were analyzed using a two-factor ANOVA where period effects (*before*after* or *BA*) and location effects (*control*impact* or *CI*) are separated from the interaction effect (*BA X CI*) (Green 1979). Since then, they have been analyzed with various parametric and non-parametric flavours of ANOVA and even some more complex multivariate analyses (Carpenter et al. 1989; Faith et al. 1991; Underwood 1991; Ellis and Schneider 1997; Terlizzi et al. 2005; Tough 2013; Smokorowski and Randall 2017). Conversely, BACI design has also been analyzed using more simple difference tests, such as a t-test comparing the *before*control* and *before*impact* differences directly against the *after*control* and *after*impact* differences (Stewart-Oaten et al. 1986; Schwartz 2019), and these tests have even been shown to be

in some cases more powerful than their ANOVA analogues (Smokorowski and Randall 2017).

While they are commonly used, BACI tests are not without their detractors and certainly the design has undergone many critiques over the years, hence the myriad of flavours and iterations (Smith 2006; Smokorowski and Randall 2017). In the consulting and government science worlds, the most common issue is likely the lack of baseline data – there simply isn't enough time to gather appropriate 'before' data, leading to more simplistic Control-Impact designs (Smith 2006; Schwartz 2019). Another pervasive and common issue is pseudoreplication, where samples may be replicated but treatments are either not replicated or not independent from each other. Indeed, in an oft-cited review of 176 papers, Hurlbert (1984) demonstrated that roughly half of all reviewed inferential ecology studies from 1960 and 1984 contained some variation of pseudoreplication. This is perhaps one of the main reasons why there are so many modern iterations of BACI design.

Nevertheless, BACI experiments remain commonplace in the world of environmental impact assessment. From an academic and statistical standpoint, they must however be used with caution, and their results likewise subjected to scrutiny (Hurlbert 1984; Conquest 2000; Smokorowski and Randall 2017). In this thesis, a simple t-test approach was employed similar to that proposed by Stewart-Oaten et al. (1986). However, given the short time frame of a Masters' thesis, BACI results from a single season must be seen as informative to the direction of future studies as opposed to damning (or otherwise) evidence. This was also the impetus for incorporating non-hypothesis testing multivariate analyses (PCAs) in the execution of this thesis.

Chapter 3: Methods

3.1 Paired studies

As was indicated in section 1.1, this thesis consists of two parallel studies undertaken simultaneously. The first study was undertaken to explore the impacts of benthic burlap barriers used in *M. spicatum* control on the physical, chemical and biological parameters of the Malcolm Lake ecosystem. The second study was larger in scope and was undertaken to investigate the impacts of mechanical cutting (again of *M. spicatum*) on the physical, chemical and biological parameters of three similar lakes chosen along a nutrient gradient. These three lakes were Dog Lake, Otter Lake and Wolfe Lake. In both cases, the treatments used were by design small in size, mimicking a typical treatment that would be used by landowners and lake associations in lakes such as these. Indeed, the Ontario regulations only allow for small lakefront treatments in non-permitted situations (Government of Ontario 2017), and Parks Canada allows at maximum 465 m² treatments for permitted removals (Parks Canada 2017). Thus, these studies were undertaken to explore the localized and small-scale impacts that are currently occurring in Canadian lakes, rather than whole lake impacts. Furthermore, treated areas were not isolated (e.g. surrounded by enclosures) from the surrounding lake ecosystem for the very same reason: in this thesis, I wish to elucidate if these small disturbances can be detected within the larger system in keeping with my overarching theme of ecosystem resilience. That said, study sites were selected in sheltered bays to minimize water mixing effects.

3.2 Study sites

Malcolm lake, the sole lake studied in the benthic barrier treatment, is located in the Municipality of North Frontenac, Frontenac County, Ontario, Canada (44°55'12"N; 76°53'21"W) (Figure 3-1). It is a shallow, mesotrophic shield lake with a maximum depth of 4.6 m, mean depth of 2.2 m and a total surface area of 218 ha (Table 3-1). As of 2015, there were 107 properties, mostly cottages, taking up approximately 75% of the shoreline, with the balance being undeveloped private land in a natural state. In 1969 a concrete dam was put in at the lake outlet by the Ministry of Natural Resources and Forestry to control water levels. Water quality monitoring and dam operation were taken over by the Mississippi Valley Conservation Authority in 1986. The Malcolm and Ardoch Lakes Landowners Association (MALLA) is a volunteer-based lake association that maintains a stewardship plan for the Lake. The sampling location for this study was selected by MALLA as an area of concern and was situated in and around one of the larger *M. spicatum* patches in the lake, located along the northwestern shore, just south of the Ridge Lane cottage access road (Figure 3-2A). The sediment in the study area was an orange-brown colour, largely unconsolidated (soft and moist) and contained many gastropod shells.

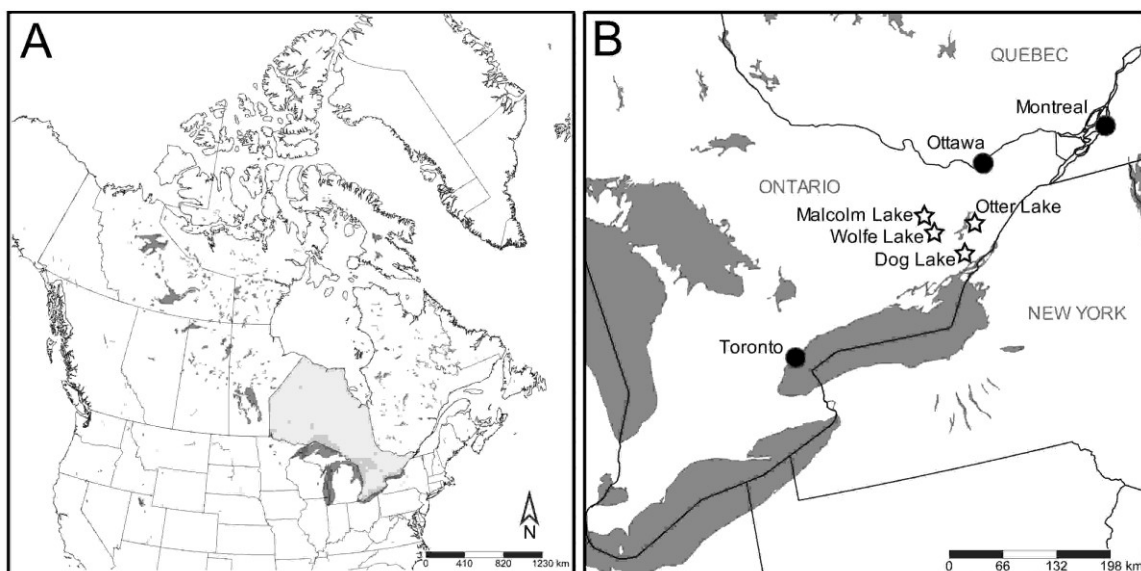


Figure 3-1. Map showing the location of Malcolm Lake, Dog Lake, Otter Lake and Wolfe Lake within North America (A) and Eastern Ontario (B).

Table 3-1. Morphometric, physical and chemical characteristics of Malcolm, Dog, Otter and Wolfe Lakes, Ontario, Canada. Morphometric data were obtained from the Mississippi Conservation Authority, Rideau Valley Conservation Authority and Cataraqui Region Conservation Authority. Physical and chemical parameters are reported as mean values ($n = 36$) for the sampling period of June to September, 2019 for Malcolm lake and mean values ($n = 24$) for the sampling period of July to October, 2019 for the three Rideau Lakes (Dog, Otter and Wolfe).

	Malcolm	Dog	Otter	Wolfe
Surface area (km ²)	2.2	9.6	6.0	10.0
Elevation (m)	253	109	124	135
Max depth (m)	4.6	50.0	36.6	40.0
Mean depth (m)	-	5.8	10.0	10.3
Water temperature (°C)	21.9	21.5	22.0	22.1
Total phosphorus (mg/L)	0.009	0.020	0.007	0.009
Total Kjeldahl nitrogen (mg/L)	0.54	0.62	0.42	0.38
Sediment moisture content (% mass)	96.0	89.1	85.3	59.4
Dry sediment organic content (% mass)	51.0	19.0	13.4	4.8

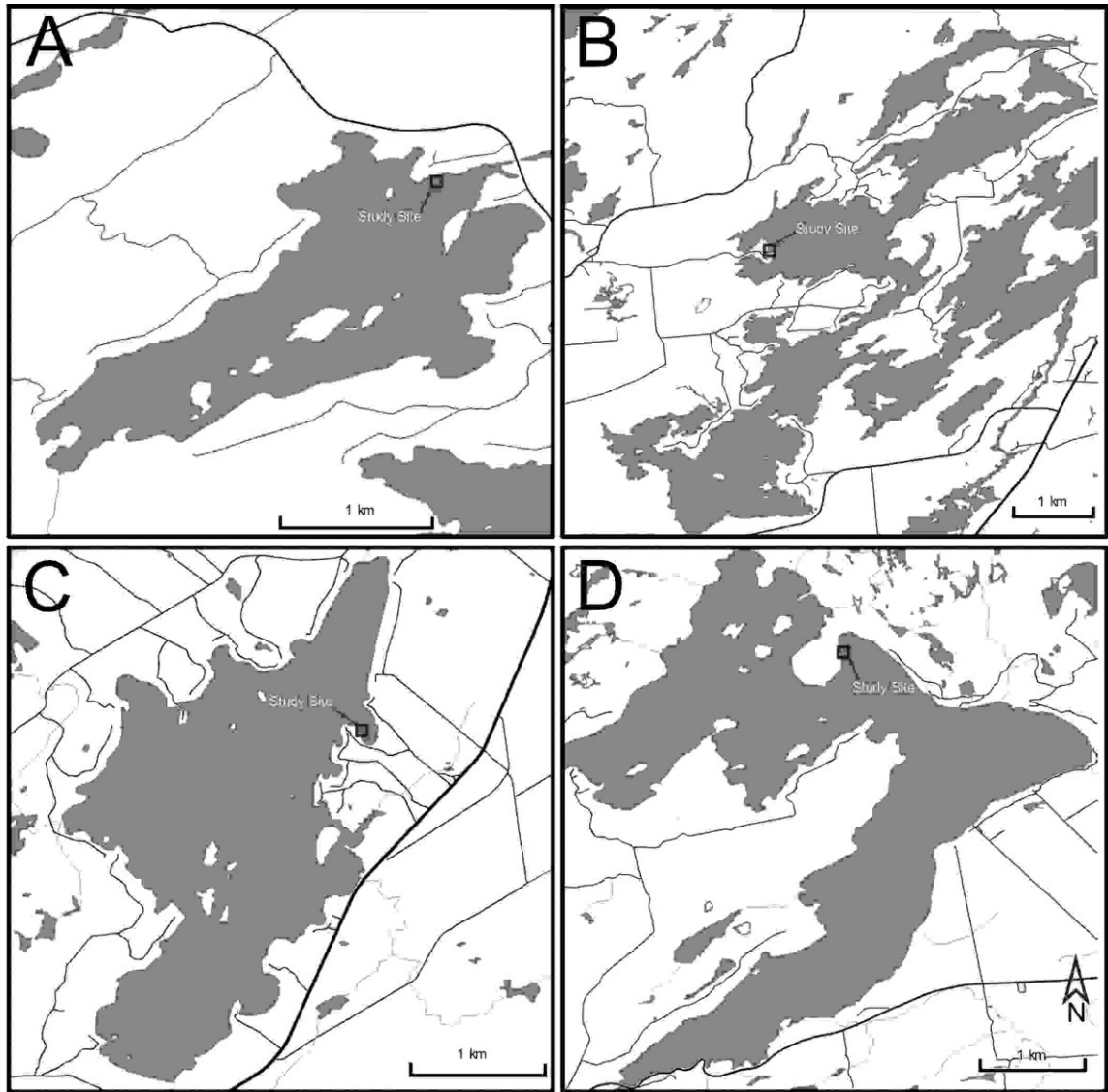


Figure 3-2. Map showing the location of the study sites in each of the four lakes (A Malcolm, B Dog, C Otter and D Wolfe). Each study site was located within a single large stand of *Myriophyllum spicatum* where the water depth was approximately 2 m.

The three lakes chosen for the second study are all connected to the Rideau Waterway in Eastern Ontario, Canada (Figure 3-1). The lakes were chosen along a nutrient gradient based on historical Total Phosphorus (TP) data from the Ontario Lake Partner Map (Ministry of the Environment Conservation and Parks 2022), from averages of readings taken at sampling points closest to selected study sites. There are many

different scales used to measure the trophic status of lakes, but for the purposes of this study the simple Ontario Ministry of the Environment, Conservation and Parks scale will be used: <0.010 mg/L TP is oligotrophic, 0.010 – 0.020 mg/L TP is mesotrophic and >0.020 mg/L TP is eutrophic (Ministry of the Environment Conservation and Parks n.d.). The historical averages obtained were 0.029 mg/L for Dog Lake, 0.008 mg/L for Otter Lake and 0.006 mg/L for Wolfe Lake. All three Lakes were also chosen based on having large infestations of *M. spicatum*, which was confirmed *in situ* prior to commencing the study in May and June. Lastly, in order to ensure that study sites were as similar as possible, they were specifically situated in patches of *M. spicatum* that were immediately East of a North-South shoreline in a sheltered bay and in approximately 2 m depth of water.

Dog Lake (Figure 3-2B), chosen as an example of a eutrophic lake, is located in the Municipality of South Frontenac, Frontenac County, Ontario, Canada (44°26'01"N; 76°21'30"W). It is a mostly shallow, partially artificial lake (flooded during construction of Rideau Canal) with a maximum depth of 50.0 m, mean depth of 5.8 m and a total surface area of 964 ha (Table 3-1). Of the three chosen lakes in the second study, it is the only lake directly connected (navigable by boat) to the Rideau Waterway. The Lake is managed by the Cataraqui River Conservation Authority (CRCA). The sediment found at the Dog Lake site was fine, orange-brown in colour and contained many decomposing plant remains.

Otter Lake (Figure 3-2C), chosen as an example of a mesotrophic lake, is located in the Rideau Lakes Township of Leeds-Grenville County, Ontario, Canada (44°47'42"N; 76°06'71"W). It is medium sized lake with a maximum depth of 36.6 m, mean depth of

10.0 m and a total surface area of 600 ha (Table 3-1). The Lake is managed by the Rideau Valley Conservation Authority (RVCA). The sediment of Otter Lake was very pale in colour and contained an exorbitance of crushed zebra mussel shells.

Wolfe Lake (Figure 3-2D), chosen as an example of an oligotrophic lake (or closest thing to it in the region), is located in the Municipality of South Frontenac, Frontenac County, Ontario, Canada (44°41'49"N; 76°29'17"W). It is medium sized lake with a maximum depth of 40.0 m, mean depth of 10.3 m and a total surface area of 1000 ha (Table 3-1). The Lake is managed by the Rideau Valley Conservation Authority (RVCA) and the Wolfe Lake (Westport) Association maintains a Stewardship plan. As a feeder lake to the Rideau waterway, the water level of the lake is managed by Parks Canada. For the sampling year, the mean TP for Wolfe and Otter Lake were very similar (0.007 and 0.009 mg/L, respectively; Table 3-1) making them thus analogous on a nutrient gradient, as opposed to distinct. The sediment of this lake was the most coarse and rocky, containing a range of sand and gravel, as well as the occasional zebra mussel shell and decomposing woody roots and stems.

The geology of the province of Ontario consists of the Precambrian rocks of the Canadian Shield overlain by various Phanerozoic sedimentary rocks (Ontario Geological Survey 2011). All four of the lakes in this study lie atop two or more geological formations, with the Rideau Lakes having the most overlap (Dog Lake sits on Ontario Geological Society's formations 46, 50 and 52; Otter Lake 46 and 52; Wolfe Lake 46 and 50b) and Malcolm Lake having some slightly different influences (formations 44, 46 and 47). Nevertheless, all four lakes lie primarily over the Grenville Supergroup/Flinton Group (Ontario Geological Society's formation 46). This group is made up of carbonate

metasedimentary rocks, marble, calc-silicate rocks, skarn and tectonic breccias (Ontario Geological Survey 2011). The presence of carbonates in part explains the relatively high pH of these lakes.

Lastly, it was observed that all four lakes had at least some zebra mussel (*Dreissena polymorpha*) presence and/or influence. Indeed, the Otter Lake sediment samples contained by far the most zebra mussel shells and was concomitantly the clearest lake. Zebra mussels are renowned for their ability to clarify even nutrient rich lakes (Macisaac 1996) but their presence in all four lakes removes them as a primary confounding factor.

3.3 Study design and treatments

In order to detect any potential changes in the environment resulting from the two treatments a before-after-control-impact (BACI) study design was undertaken (Green 1979; Smith 2006) in the summer and fall of 2019 and a differences test approach (Smokorowski and Randall 2017) was employed. This consisted of choosing a ‘control’ site and an ‘impact’ site within close proximity to each other and within a single dense stand of *M. spicatum*, and taking measurements of physical, chemical and biological parameters ‘before’ and ‘after’ the treatment date.

The treatment date for the laying of benthic burlap mats on Malcolm Lake was June 20th and was chosen by the members of MALLA as being convenient for volunteer availability. The treatment consisted of volunteers laying down three 45.7 m x 2.4 m burlap barriers over the selected macrophyte patch using a dispensing barge and personal watercraft, weighing the matting down with locally obtained stones (Figure 2-4). Due to

the timing requirement of the lake association, there was only an opportunity to obtain one set of ‘before’ samples prior to the treatment. Three sampling periods were undertaken ‘after’ the treatment date. Dates were chosen to be approximately one month apart. A third sample site in an adjacent macrophyte-free area of similar water depth (approximately 2 m in all cases) was selected and designated as ‘bare sediment’ in the Malcolm Lake study for comparison purposes, and data derived from this site was not included in the BACI test. In Malcolm Lake, the selected 1.97 ha patch was completely infested, with the plants already growing right to the surface at the time of initial sampling in June and showing no signs of die-back in September. The total treated area was small compared to the overall patch size (0.033 ha of 1.97 ha, or 1.7%), however this was based on MALLA’s plan not to remove the entire patch but rather to create plant-free corridors near shore and in heavier boat-traffic areas (Figure 3-3).

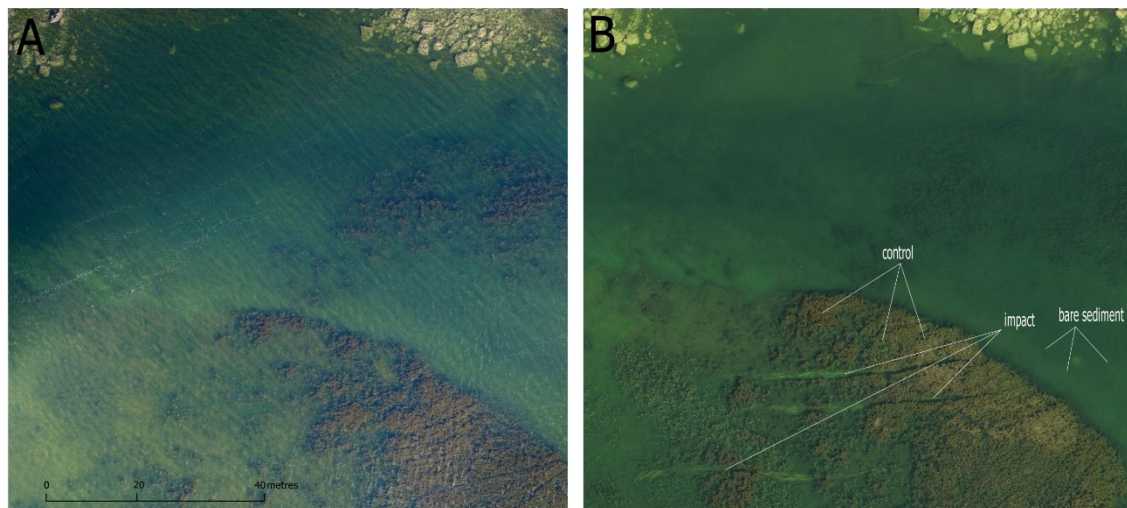


Figure 3-3. Orthomosaic drone footage of the Ridge Lane *Myriophyllum spicatum* stand on Malcolm Lake in May, 2018 (A) and June, 2019 (B). Control, impact and bare sediment sampling locations are also indicated (B).

The treatment date for mechanical cutting on all three Rideau lakes was September 7th, 2019, in late summer when plant biomass was at its greatest. The mechanical cutting treatment consisted of mowing a 20 m by 5 m (0.01 ha) strip through a dense patch of *M. spicatum* parallel to shore using Rideau Aquatic Weed Control's custom-made weed harvester barge (Figure 2-3). The custom conveyor/sickle bar mower allowed all plant biomass to be removed from the site for offshore composting. Several bi-directional passes were conducted in order to ensure a thorough cutting of the plants. For the Rideau lakes study, two sampling periods were undertaken 'before' the treatment date and two 'after.' All sample sites were in approximately 2 m depth of water at the time of initial sampling in July. The selected patches were completely infested, with the plants already growing right to the surface at the time of initial sampling. There were some signs of die-back in the final two sampling periods in September and October, especially in Wolfe Lake. No resultant patch size area estimates are available for the three Rideau Lakes, as Drone footage was only available in the case of Malcolm Lake (Figure 3-3), but the size was consistent with what one could expect of a typical landowner's lakefront.

3.4 Field sampling

Field sampling occurred monthly from June to October 2019. The Malcom Lake site visits occurred on June 8th, July 3rd, August 1st and September 14th and the Rideau lakes visits occurred July 9-10th, August 7-8th, September 12-13th and October 3-4th. In all cases, three replicate samples were taken at each of the two adjacent sites (control, impact, and in the case of Malcolm Lake the third 'bare sediment' sites as well) each

month. Light levels were measured in air, just below the water's surface, at a depth of 1 m and at bottom (2 m) using a LI-193 Spherical Quantum Sensor and an LI-250A Light Meter. Temperature (°C), dissolved oxygen (%), pH and conductivity (µS) were also measured at 0 m, 1 m and 2 m using a calibrated YSI ProPlus Multiparameter Instrument. Water samples were collected in triple-rinsed 4 L sealable plastic containers at a water depth of 0.5 m. Sediment samples were collected using a standard 3.5 L Ekman Bottom Grab sampler and transferred to individual 1 L Ziploc bags. Zooplankton samples were collected at 0.5 m water depth by filtering 22 L of water through a 80 µm Wisconsin Plankton Sampler. The filtrate was transferred to 250 mL sealable plastic containers, to which was added 40 mL of carbonated water, to narcotize the zooplankters and relax their external structure, followed by 40 mL of 95% ethanol to preserve the samples (Ohio EPA 2010). All field samples were kept in a cooler with ice packs and then transferred to a refrigerator for short-term storage. All site visits and field sampling took place between 10 am and 2 pm EDT. Drone footage orthomosaics were obtained from the Above and Beyond education program to help characterize and map the Malcolm Lake treatments (Figure 3-3). For the Rideau lakes, where drone footage was unavailable, echosounder sweeps were instead conducted on September 6th (before) and October 10th (after), using a Lowrance Elite 7 Fishfinder/GPS. ReefMaster (ReefMaster Software Ltd. 2021) and Fiji/ImageJ (Schindelin et al. 2012) were used to estimate the percent volume inhabited (PVI) of *M. spicatum* before and after the treatment.

3.5 Laboratory analysis

Total Kjeldahl nitrogen and total phosphorus were simultaneously determined from acid digested samples using a Skalar segmented flow autoanalyzer by the Robert O. Pickard Environmental Centre in Ottawa, using standard protocols (City of Ottawa Environmental Services Department 2018) adapted from the Ontario Ministry of the Environment, Conservation and Parks (Ministry of the Environment 2010). Three 250 mL water samples from each site at each sampling time were analyzed in this fashion. Dissolved organic carbon was determined using a Shimadzu TOC-L total organic carbon analyser after each 15 mL sample was first filtered through a 0.45 μm filter tip.

Chlorophyll-*a*, used as a proxy for algal biomass, was determined using the monochromatic methods of Lorenz (1967) as described in Wetzel and Likens (2011), with some small changes as outlined below. All Chlorophyll-*a* procedures were carried out in a dark room to avoid exposure to light. First, 1 L samples of water from each site were filtered using a vacuum pump filtration apparatus and Whatmann GF/F filters. The filters were folded and placed in sealed plastic centrifuge tubes, wrapped in aluminum foil and stored in a freezer. Pigment extraction was done by placing the filters in test tubes with 15 mL of 95% ethanol and heating them to 79°C for 5 minutes. The tubes were subsequently wrapped in aluminum foil and stored in a refrigerator at 4°C overnight. Absorbances were analyzed using a Shimadzu UV-1280 Spectrophotometer. Five milliliters of the extracted pigments in ethanol, once allowed to warm up to room temperature, were filtered through 0.45 μm filters and transferred to a 1 cm path length quartz cuvette. Absorbances were measured at 665 nm and 750 nm to correct for

turbidity, before and after 0.15 mL of 1 M HCl was added to each sample to correct for phaeopigments.

Zooplankton samples were filtered through an 80-micron mesh into 20 mL disposable scintillation vials using a wash bottle containing distilled water, and then transferred into a Bogorov counting chamber to be enumerated and identified using a Leica S9i Digital Stereo Microscope. Identification was to family level where possible, as per Haney et al. (2013). A higher taxonomic resolution was not selected as zooplankton data was intended to be used to monitor for generalized responses only, occluding the requirement for species-level identification (Olsgard and Somerfield 2000; Bailey et al. 2001; Bozzuto and Blanckenhorn 2017).

Sediment samples were first homogenized within their individual Ziploc bags and then 5 mL from each sample were placed in crucibles and weighed, then heated to 110°C for 4 hours in a muffle furnace and weighed again to determine the moisture content as a percent of the wet sediment mass. The dry sediment crucibles were then placed back in the muffle furnace for 4 hours at 550°C and weighed a final time to determine the organic content as a percent of dry sediment mass. Five hundred milliliters of each sediment sample were also analyzed for macroinvertebrates after being sieved through a 500 µm sieve and rinsed with distilled water. Sieved contents were transferred to 100 mm x 15 mm petri dishes, marked with a 1 cm grid, where macroinvertebrates were enumerated and identified using a Leica S9i Digital Stereo Microscope. Identification was generally to Order or Family level according to Thorp and Covich (2001). Surprisingly, no living macroinvertebrates were found in most of the samples from Malcolm Lake and therefore

no further macroinvertebrate data were presented for this portion of the study. This was not the case in the three Rideau lakes, where benthic macroinvertebrates were plentiful.

3.6 Statistical analyses

All statistical analyses were performed in R version 3.6.1 (R Core Team 2018). All data used for the tests, with the exception of zooplankton ($z = 0.5$ m), were the mean (μ) of triplicate measurements taken at bottom ($z = 2$ m). As an assessment of site selection and pre-treatment natural variability within these sites, Two Sample t tests were performed on all of the control vs impact site data before treatment. For the BACI analysis, difference of difference tests, again Two Sample t tests, were performed on untransformed data. The BACI interaction effect was a measure of a ‘difference of differences’ (equation 1), with the null hypothesis that this difference was 0.

$$(1) \quad BACI \text{ difference} = (\mu_{Control \text{ Before}} - \mu_{Impact \text{ Before}}) - (\mu_{Control \text{ After}} - \mu_{Impact \text{ After}})$$

For both studies, three different pairwise comparisons of the *control – impact* differences were made, although the structure of these comparisons were necessarily different between Malcolm and the Rideau lakes. For the Malcolm Lake study, the comparisons undertaken were (1) the month before (June) vs. all three months after (July, August and September); (2) the one month before vs. the first month after alone (July); and (3) the one month before vs. the third month after alone (September only). For the Rideau lakes study, the comparisons undertaken were (1) the two months before (July and August) vs. the two months after (September and October); (2) the two months before vs. the first

month after alone (September); (3) the two months before vs. the second month after alone (October only). These differences were chosen to provide a glimpse into the overall impact of the treatment (in 1), as opposed to any potential immediate short-term impacts (as in 2) or any longer-term impacts that did not develop immediately (in the case of 3). It is important to note that impact sites pre-treatment are not true impact sites. Despite this unwieldy terminology, it is a commonly used term in BACI tests.

Ordinations by means of a Principal Component Analysis (PCA) were performed using the ‘vegan’ package (Oksanen et al. 2019) in R on the zooplankton taxa data as well as the macroinvertebrate data, which were Hellinger-transformed to reduce the impact of dominant taxa. These data were further visually delineated by the factors of time (month), treatment (control/impact, bare sediment where applicable), and treatment*time (represented by 1 standard deviation ellipses). This method of ordination preserves Euclidean distances and allows the data to be simply visualized in such a way where data that are closer together are more similar and data that are further apart are more dissimilar. The significance of component axes was determined using scree plots (Appendix B). Additional ordinations for environmental variables as well as direct ordinations are presented only as supplemental material (Appendix C).

Chapter 4: Results

4.1 Efficacy of treatment and general observations: Malcolm

In the Malcolm Lake study, the benthic barriers were still intact by the end of the sampling season (Figure 4-1). There were no observed instances of macrophytes growing through the burlap, although there was some tenting where the mats failed to press the plants into the sediment, and some cases of plants growing out from under the side of the mats. The mats themselves were covered in a thin periphyton and both bluegills (*Lepomis macrochirus*) and small-mouthed bass (*Micropterus dolomieu*) were observed swimming in the resultant trenches made through the dense macrophyte patch. While the status of the plants under the mats was not determined, total localized and non-selective control was achieved during the time of the study. Over the course of the entire four months there was a seasonal trend of rising conductivity across all sites, and a seasonal trend of diminishing pH and dissolved oxygen across all sites (Figure 4-2).



Figure 4-1. Underwater photograph of a deployed burlap benthic barrier in Malcolm Lake, Ontario. Here, the barrier is ‘tenting’ from the plants caught underneath and is thus not deployed at maximum width.

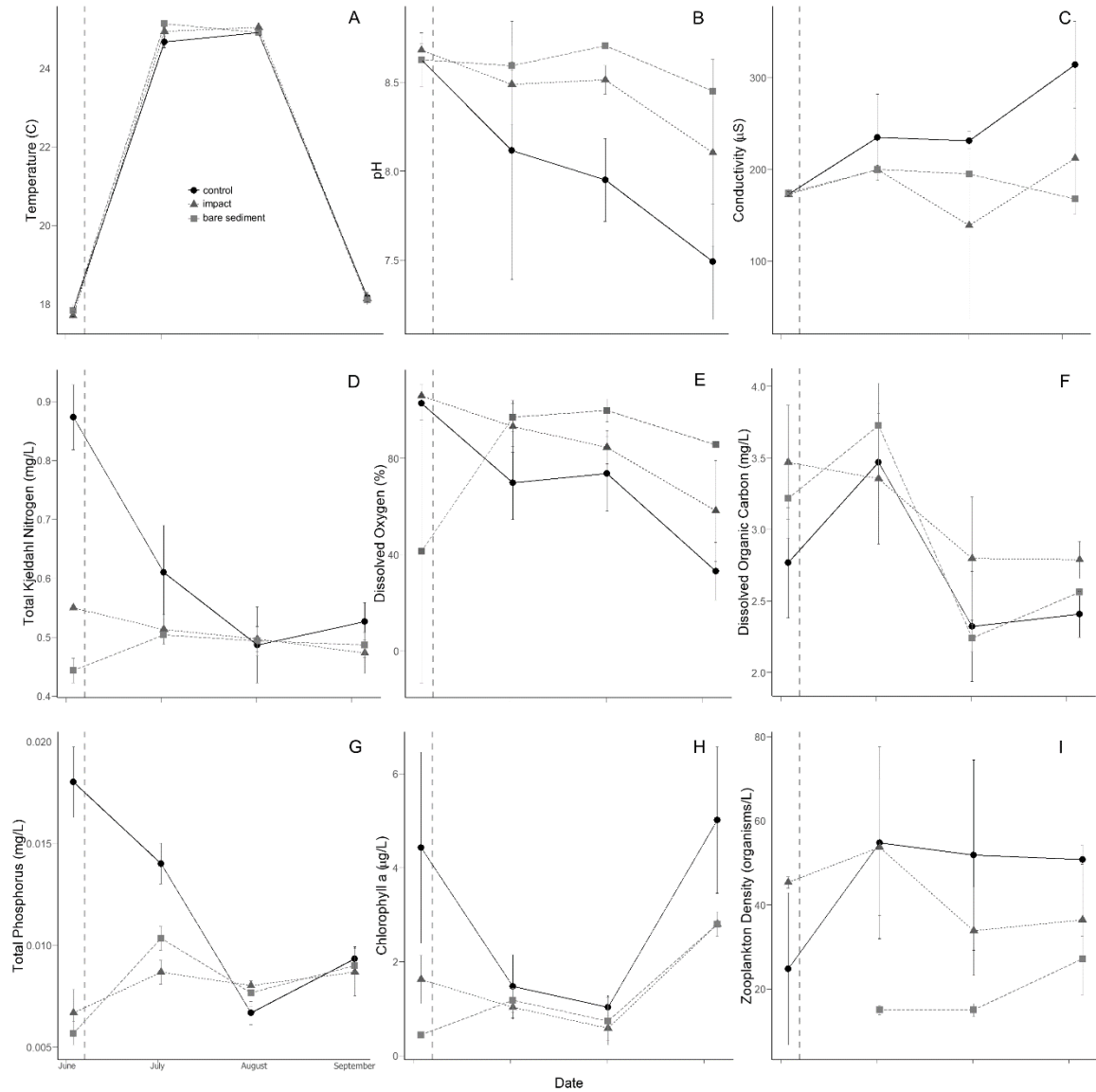


Figure 4-2. The physical (A temperature), chemical (B pH, C conductivity, D total Kjeldahl nitrogen, E dissolved oxygen, F dissolved organic carbon, G total phosphorus) and biological (H chlorophyll *a*, I zooplankton density) parameters measured by sampling time (June 8th, July 3rd, August 1st and September 14th 2019) for Malcolm Lake, Ontario. The dotted vertical line represents the treatment date (June 20th, 2019). Black circles represent samples taken from the control site (within the macrophyte stands), dark grey triangles represent samples taken from the impact site (within the macrophyte bed ‘trenches’ formed by burlap mats) and light gray squares represent samples taken from the bare sediment site (open water region adjacent to macrophyte bed). The points represent mean values (n=3) and the error bars standard error. Only the zooplankton density control-impact differences are significantly different before and after treatment ($p < 0.05$).

4.2 Efficacy of treatment and general observations: Rideau

The effect of the mechanical cutting treatment was generally quite large. The central swathe of the treated area was mostly devoid of plants after cutting, whereas plants on the periphery tended to be pushed aside and subsequently bounce back. *M. spicatum* is renowned for its quick growth (Aiken et al. 1979) and so there was also likely some regrowth during the final month. Nevertheless, in all cases there were less plants in the treated areas a month later than there were pre-treatment, with PVIs reduced by 43%, 33% and 23% for Dog, Otter and Wolfe Lakes, respectively (Figure 4-3). Of the three lakes, Wolfe had by far the rockiest substrate, which at times interfered with the cutting mechanism. Wolfe Lake, as a water-level controlled feeder lake to the Rideau Waterway also saw the greatest change in depth over the course of the study. Over the four-month period of observation, some general trends were observed in both abiotic (Figure 4-4) and biotic (Figure 4-5) factors across all three of the Rideau lakes. Temperature, dissolved oxygen, dissolved organic carbon, conductivity and pH all decreased over time, whereas TP, Chlorophyll-*a* and the number of macroinvertebrate taxa increased from July to October.

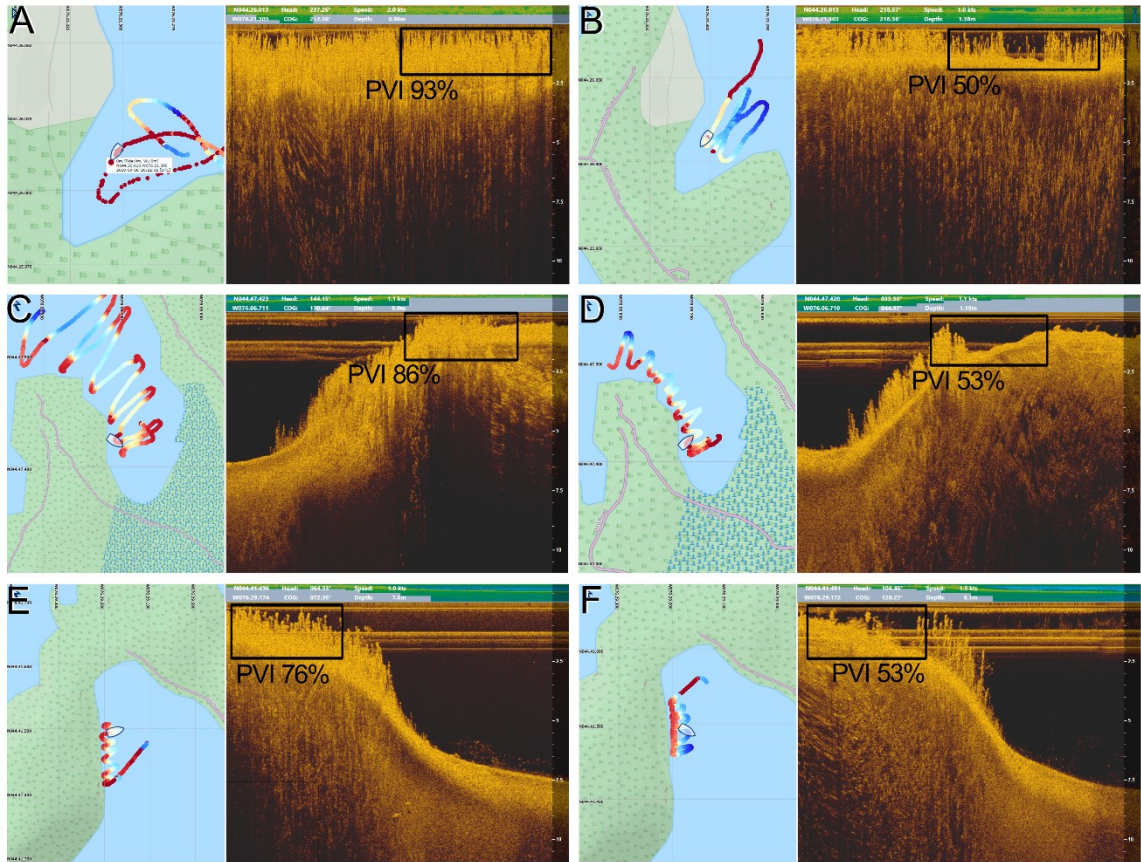


Figure 4-3. Echosounder sidescan mosaics showing each impact area 'before' (September 6, left column) and 'after' (October 10, right column) the treatment date (September 7), for Dog (A, B), Otter (C, D) and Wolfe Lakes (E, F). PVI estimates, shown below black boxes, (using ImageJ colour analyses) are rough approximations only, as they contain some untreated areas as well, and represent only a single cross section of the treated area.

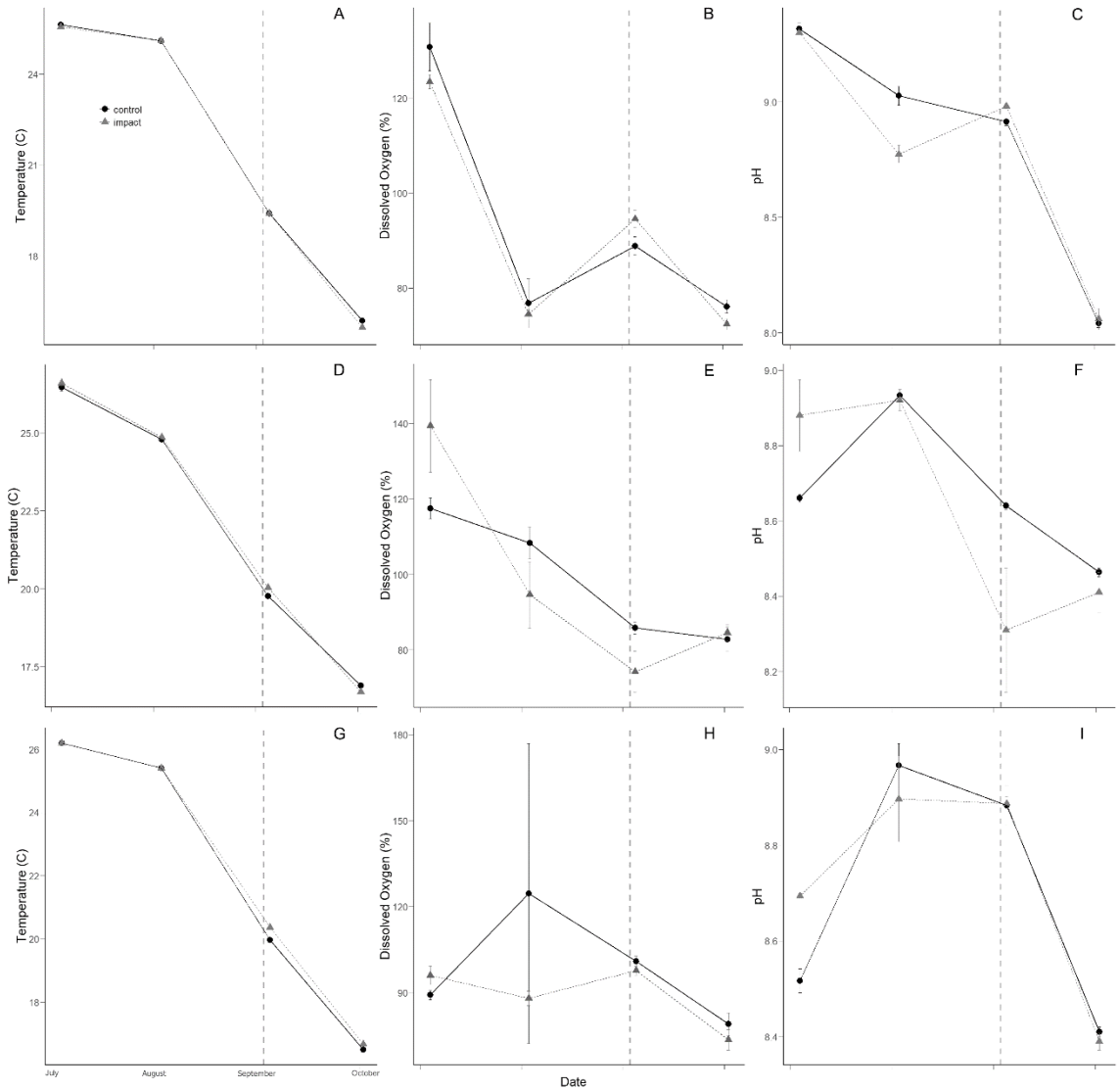


Figure 4-4. The temperature (A Dog, D Otter and G Wolfe), dissolved oxygen (B Dog, E Otter and H Wolfe) and pH (C Dog, F Otter and I Wolfe) for all three lakes by sampling time (July 10th, August 7th, September 13th and October 4th, 2019). The dotted vertical line represents the treatment date (September 7th, 2019). Black circles represent samples taken from the control site (within the macrophyte stands) and light grey triangles represent samples taken from the impact site (where the macrophytes were mown). The points represent mean values (n=3) and the error bars standard error. Control-impact differences are significantly different before and after treatment in C, F and G ($p < 0.05$).

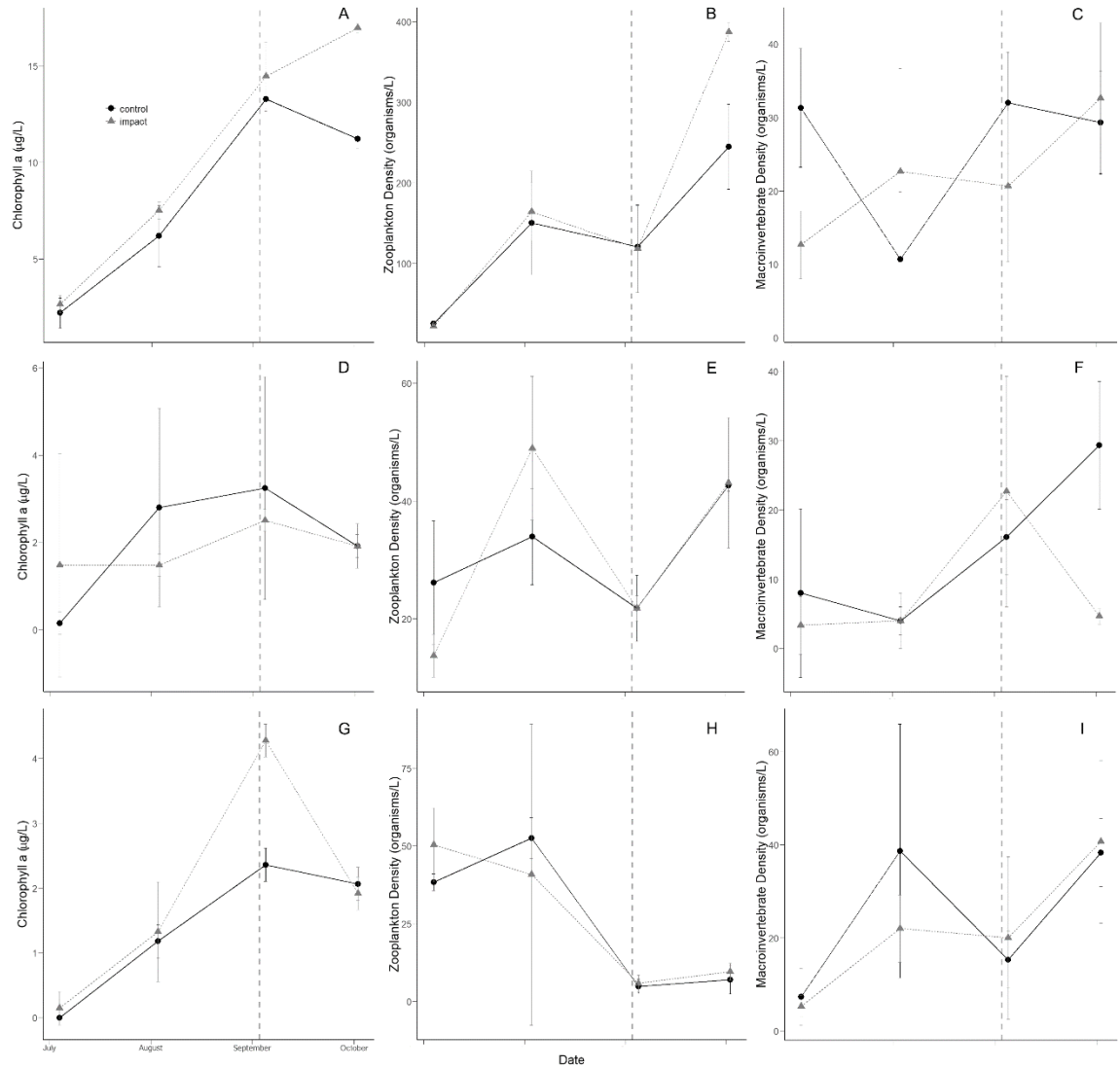


Figure 4-5. The Chlorophyll a (A Dog, D Otter and G Wolfe), zooplankton density (B Dog, E Otter and H Wolfe) and macroinvertebrate density (C Dog, F Otter and I Wolfe) for all three lakes by sampling time (July 10th, August 7th, September 13th and October 4th, 2019). The dotted vertical line represents the treatment date (September 7th, 2019). Black circles represent samples taken from the control site (within the macrophyte stands) and light grey triangles represent samples taken from the impact site (where the macrophytes were mown). The points represent mean values (n=3) and the error bars standard error. No control-impact differences are significantly different before and after treatment ($p < 0.05$).

4.3 BACI differences: Malcolm

In Malcolm Lake the initial differences between the ‘control’ and at that point untreated ‘impact’ sites were already significantly different ($p < 0.05$) for Chlorophyll-*a*, total phosphorus, total Kjeldahl nitrogen, dissolved organic carbon and light extinction. This points to pre-existing natural variability between the control and impact sites and poses a complication to any further BACI result interpretations for Malcolm Lake, even where the initial differences were not significant. The starkest contrasts between control and impact sites where the initial differences were not significant were observed in conductivity, dissolved oxygen, pH and zooplankton density (Figure 4-2). However, of these differences, only the zooplankton density were significant at the $p < 0.05$ level (Table 4-1).

Table 4-1. Summary of Two Sample t tests comparing the difference of mean differences, $(\mu_{Control\ Before} - \mu_{Impact\ Before}) - (\mu_{Control\ After} - \mu_{Impact\ After})$, for temperature (°C), light (% reaching bottom), conductivity (µS), pH, dissolved organic carbon (mg/L), dissolved oxygen (%), total Kjeldahl nitrogen (mg/L), total phosphorus (mg/L), chlorophyll-*a* (µg/L) and zooplankton density (organisms/L). Each test shows a difference, standard error, degrees of freedom, t-statistic and p-value. P-values < 0.05 are highlighted in bold and are also marked with an asterisk (*) if the initial differences were not already significant. Tests are for *before vs. after* (June data against pooled July, August and September data).

	Physical		Chemical						Biological	
	Temp	Light	Cond	pH	DOC	DO	TKN	TP	Chl-a	Den Zoop
mean difference	0.256	0.507	-75.989	0.462	-0.456	16.756	0.277	0.010	1.770	-31.762
SE	0.137	2.826	48.958	0.336	0.275	12.037	0.050	0.002	0.848	11.771
df	10	10	10	10	10	10	10	10	10	10
t	1.866	0.179	-1.5521	1.377	-1.657	1.392	5.582	5.094	2.087	-2.698
p	0.092	0.861	0.152	0.199	0.129	0.194	0.000	0.000	0.063	*0.022

4.4 BACI differences: Rideau

For the initial differences tests, only the pre-treatment light levels for Otter Lake were significantly different between the chosen control and impact sites ($p = 0.004$). Across all lakes the most consistently significant ($p < 0.05$) BACI differences were those for temperature in the asymmetrical tests of *before* vs October alone, which was observed in all three lakes, although these differences were quite minimal (less than half a degree at most) and the sign of the change was not consistent. Light levels were significantly different in Before vs. September alone for Wolfe Lake ($p = 0.024$), and all three comparisons for Otter Lake, but these are of course invalidated by the fact that the initial control-impact differences were already significant in that lake. Little can be said about the light level changes for Dog Lake as it underwent a massive algal bloom during September and October, entirely blocking light from reaching bottom in the whole of the study area. Conductivity was significant for the Before vs. September alone test in Otter Lake ($p = 0.024$), whereas the pH BACI difference was significant in both the overall tests (Table 4-2) and the Before vs. September tests for both Dog and Otter Lakes, although the sign of the change was again not the same for both lakes. Dissolved oxygen levels were significantly different for Dog Lake in the Before vs. September alone tests ($p = 0.024$). There were no significant differences in DOC, TP and TKN for any of the tests in any of the lakes (Table 4-2).

Amongst the invertebrate densities, only two significant differences were observed (Figure 4-5): the macroinvertebrate density for Before vs. October alone test in Otter Lake (with less macroinvertebrates found in the impact site; $p = 0.027$)

Table 4-2. Summary of Two Sample t tests comparing the difference of mean differences, $(\mu_{Control\ Before} - \mu_{Impact\ Before}) - (\mu_{Control\ After} - \mu_{Impact\ After})$, for temperature (°C), light (% reaching bottom), conductivity (µS), pH, dissolved organic carbon (mg/L), dissolved oxygen (%), total Kjeldahl nitrogen (mg/L), total phosphorus (mg/L), chlorophyll-*a* (µg/L), macroinvertebrate density (organisms/L) and zooplankton density (organisms/L). Each test shows a difference, standard error, degrees of freedom, t-statistic and p-value. P-values < 0.05 are highlighted in bold and are marked with an asterisk (*) if the initial differences were not found to be significant. Tests are for *before* vs. *after* (pooled July and August data against pooled September and October data) and are organized by Lake.

Lake		Physical		Chemical				Biological				
		Temp	Light	Cond	pH	DOC	DO	TKN	TP	Chl-a	Den Inv	Den Zoop
Dog	mean difference	-0.083	0.802	-0.667	0.178	-0.009	5.850	0.027	0.001	2.581	-0.667	64.750
	SE	0.058	1.218	0.618	0.058	0.113	3.010	0.024	0.001	1.284	10.062	40.190
	df	10	10	10	10	10	10	10	10	10	10	10
	t	-1.432	0.658	-1.079	3.091	-0.083	1.947	1.091	1.040	2.009	-0.066	1.611
	p	0.183	0.525	0.306	*0.011	0.936	0.080	0.301	0.323	0.072	0.949	0.138
Otter	mean difference	-0.067	17.003	16.5	-0.295	0.001	-9.017	-0.062	0	-0.369	-6.704	-1.106
	SE	0.120	3.500	8.943	0.096	0.296	9.588	0.036	0.001	1.227	9.684	8.166
	df	10	10	10	10	10	10	10	10	10	10	10
	t	-0.555	4.857	1.845	-3.088	0.002	-0.940	-1.709	0.164	-0.300	-0.692	-0.135
	p	0.591	0.001	0.095	*0.011	0.999	0.369	0.118	0.873	0.770	0.505	0.895
Wolfe	mean difference	0.283	6.207	1.95	-0.062	0.019	10.55	0.025	0.0003	0.738	12.833	1.614
	SE	0.060	3.217	1.363	0.065	0.112	17.352	0.015	0.0005	0.519	9.817	15.515
	df	10	10	10	10	10	10	10	10	10	10	10
	t	4.715	1.929	1.431	-0.943	0.169	0.608	1.659	0.674	1.420	1.307	0.104
	p	*0.001	0.083	0.183	0.368	0.869	0.557	0.128	0.516	0.186	0.220	0.919

and the zooplankton density in the Before vs. October alone test for Dog Lake (with more zooplankton found in the impacted sites; $p = 0.024$). There were likewise only two significant Chlorophyll-*a* differences (Figure 4-5): in the Before vs. October alone test in Dog Lake ($p = 0.028$) and the Before vs. September alone test for Wolfe Lake ($p = 0.025$). In both cases, there were higher levels of Chlorophyll-*a* in the impacted sites (Figure 4-5).

BACI difference tests were also performed on all individual macroinvertebrate and zooplankton taxa studied, again using raw counts. Results varied quite widely, but significant ($p < 0.05$) initial differences, overall differences and asymmetrical before-after

differences were all observed. For Dog Lake, there were significant initial differences for the pelagic Ostracoda ($p = 0.049$). Significant overall before-after differences were observed in the Chaoboridae ($p = 0.008$), the Chydoridae ($p = 0.041$), Conochilidae ($p = 0.038$), Nematoda ($p = 0.024$) and Oligochaeta ($p = 0.008$). For the asymmetrical tests, the only two new taxa not indicated above were the Bosminidae and the Brachionidae, both of which were only significant in the before vs. October test ($p = 0.024$ in both cases). For Otter Lake, the only significant initial difference was observed in the Chydoridae ($p = 0.022$), whilst the only two taxa showing an overall before-after difference were the Nematoda ($p = 0.028$) and the Sididae ($p = 0.036$). Otter Lake had three taxa showing late responses only to the treatment, with the Bosminidae, Holopediidae and benthic Ostracoda all showing significant differences ($p = 0.028$, 0.009 and 0.009 , respectively). For Wolfe Lake, there were no significant initial differences, and the only taxon showing a significant difference before and after the treatment was the benthic Ostracoda ($p = 0.040$ overall; more pronounced in October at $p = 0.026$). While the Ostracoda responded in both Otter and Wolfe Lakes, their responses were the opposite (less Ostracodes in the impact in Otter Lake and more in the impact in Wolfe Lake). This was also the case with the Bosminidae in Dog and Otter Lakes (less Bosminidae in impact in Otter Lake, and more in the impact in Dog Lake). The only consistent response amongst the differences noted above were the Nematoda, who in both Dog and Otter Lakes were only detected in the control sites after the treatment, albeit in very small numbers.

4.5 Zooplankton assemblages: Malcolm

For the Zooplankton Ordination by Principal Component Analysis (Figure 4-6), the first principal component axis explained 42% of the observed variance and was very strongly associated with the Chydoridae (scaled species score = -6.04). The first PC axis strongly separated the *before*impact* sites from the bare sediment sites, whose 68% confidence interval ellipses do not overlap at all. The largest overlaps in 68% confidence interval ellipses are for the *before*impact* sites and the *before*control* sites. Given that the treatment had not occurred at this point, both of these site groups are similarly vegetated. Also displaying large overlap are the *after*impact* and bare sediment ellipses, in this case representing similarly unvegetated sites. The second PC axis explained 20% of the observed variance and was associated with the opposing influences of the Bosminidae (scaled species score of -3.79) and Cyclopoida (scaled species score of 1.28). The third PC axis (not shown on ordination plot, but determined to be significant) explained 17% of the observed variance and was strongly associated with again the Cyclopoida (-2.26), but also with rotifers of the Brachionidae (-2.36) and Asplanchnidae (1.09), and with the control sites in September (-3.23).

Taxa abundances were tracked as both raw counts as well as relative abundances. The post-treatment impact effects appear to be most closely associated with the Chydoridae, Bosminidae and Cyclopoida (Figure 4-7). The Chydoridae dominated both the control and impact sites pre-treatment (57% and 48% mean relative abundance, respectively), but immediately post treatment their relative abundance declined in the recently opened-up impact trenches (23% mean relative abundance) but remained

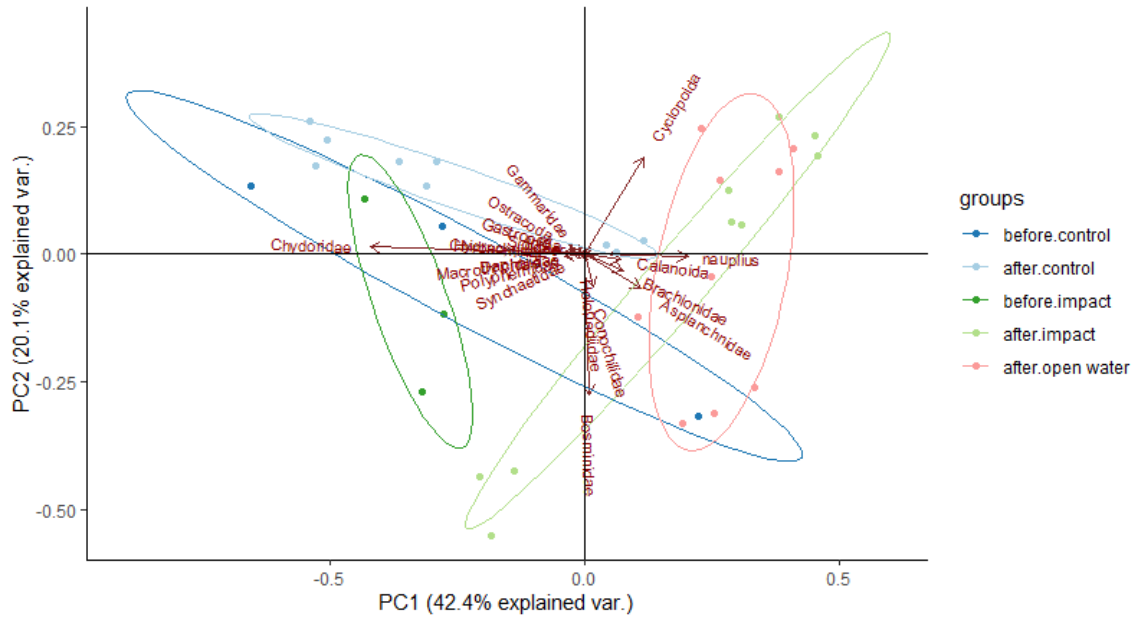


Figure 4-6. Ordination diagram by PCA for the 33 sample units based on Hellinger transformed zooplankton taxa counts for Malcolm Lake. Individual taxa (identified to Family, where possible) are represented by arrows, with arrow length representing inertia along the first two PC axes. Dark blue points represent ‘control’ sites ‘before’ the treatment, dark green points represent ‘impact’ sites ‘before’ the treatment, light blue points represent the ‘control’ sites ‘after’ the treatment and light green points represent ‘impact’ sites ‘after’ the treatment. Pink points represent the bare sediment/open water environment after the treatment. Likewise, 68% confidence interval ellipses are drawn from the centroids of all five groups using the same colour scheme. Percent variance explained for the first two PC axes are indicated in each plot.

dominant in the still vegetated control site (61% mean relative abundance). By August, the Chydoridae were virtually absent from the impact site (<1% mean relative abundance) but maintained a strong presence in the control sites (47% mean relative abundance). Meanwhile, the Bosminidae followed a completely different trend. In June, pre-treatment, they maintained modest and comparable presences in the control and impact sites (5% and 14% mean relative abundance, respectively), but immediately after the treatment there was a massive surge of Bosminidae in the impact trenches (50% of

the mean relative abundance) even though they had dwindled to <1% mean relative abundance in the vegetated control sites. Their population dropped in all sites in August and September. The trend towards more and more Cyclopoid Copepods was seen in all sites, but the control and impact sites varied in rate, with the impact sites mirroring more closely conditions in the bare sediment environment and having greater numbers.

Cyclopoids made up 20% and 16% mean relative abundance for the control and impact sites in June. Immediately after treatment, the number of cyclopoids found in the control site went up to 25% mean relative abundance, while the impact site and bare sediment both had mean relative abundance numbers of 11%. In August and September, the mean relative abundance of cyclopoids was 32% in the control sites and 52% in both the impact and bare sediment sites. The only other taxa displaying a distinct difference between mean relative abundance between the control and impact sites were the Sididae and the Macrothricidae, who followed a very similar pattern of distribution to the Chydoridae. The Sididae went from an initial June control/impact mean relative abundance of 0.6 and 0.7%, respectively, to 4.7% and 1.7% in July, 4.3% and 0.9% in August and 3.7% and 1.3% in September. The Macrothricidae went from an initial June control/impact mean relative abundance of 1.6 and 1.6%, to 2.9% and 0.8% in July, 0.3% and 0.04% in August and 9.8% and 0.3% in September.

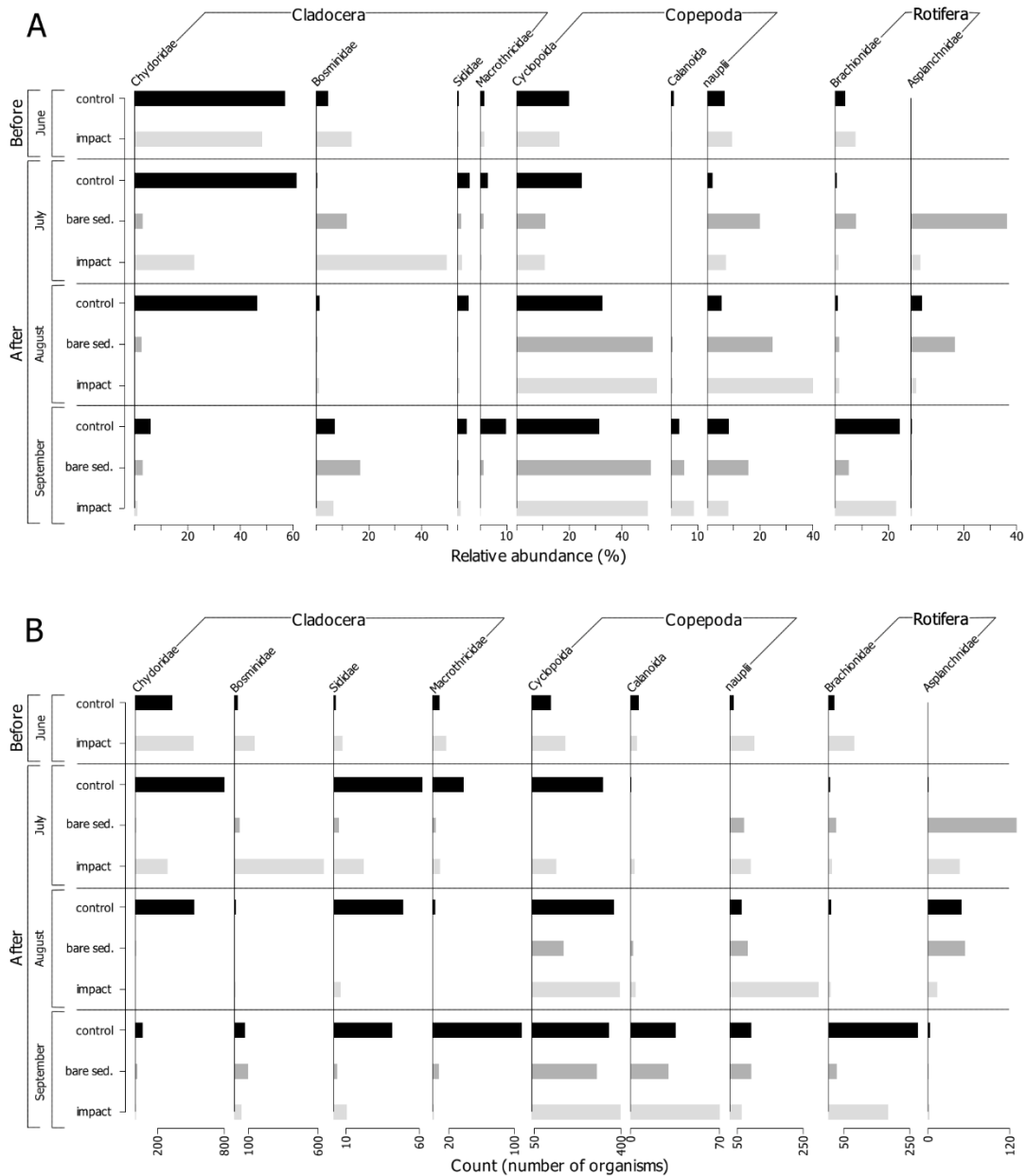


Figure 4-7. Zooplankton mean ($n = 3$) relative abundance (A, %) and count (B, number of organisms) for the 9 most populous taxa for the one sampling time (June) before the treatment and the three sampling periods thereafter (July, August, September), by sample site type (control, impact, bare sediment). Of particular note is the large drop in Chydoridae in the impact site after the treatment, the temporary surge of Bosminidae at the same site also right after the treatment, as well as the overall seasonal progression towards copepod dominance across all sites.

4.6 Zooplankton and macroinvertebrate assemblages: Rideau

Ordinations by Principal Component Analysis were done for both benthic macroinvertebrates and zooplankton for all three Rideau lakes (Figure 4-8). In the case of the Dog Lake macroinvertebrate ordination (Figure 4-8A), there are three significant PC axes which explain 28.6%, 19.7% and 13.4% of the observed variance. There is considerable overlap of the 68% confidence ellipses, with only the *before*control* and *after*control* showing no overlap. This suggests seasonal changes far outweigh treatment effects. The taxa with the highest loadings on the first PC axis are the Isopoda (−0.5690) and the Chaoboridae (0.7773), whilst the taxa with the highest loadings on the second PC axis are the Gastropoda (−0.6806) and again the Isopoda (0.5062). In the case of the Dog Lake zooplankton ordination (Figure 4-8B), there are two significant PC axes which explain 53.1% and 29.4% of the observed variance. Both ‘before’ ellipses (control and impact) overlap considerably and both ‘after’ ellipses overlap significantly, while the before and after ellipses are strongly separated. This again suggests strong seasonal influences on invertebrate community composition. The taxa with the highest loadings on the first PC axis are the Copepoda nauplii (−0.5226) and the Bosminidae (0.6786), whilst the taxa with the highest loadings on the second PC axis are the Sididae (−0.5113) and again the copepod nauplii (0.6996).

For the Otter Lake macroinvertebrate ordination (Figure 4-8C), there are two significant PC axes which explain 26.8% and 16.8% of the observed variance. The long axes of the *before*impact*, *after*impact* and *after*control* ellipses all align noticeably with the first PC axis, whereas the long axis of the *before*control* ellipse aligns noticeably with the second PC axis. The only two ellipses that do not overlap are the

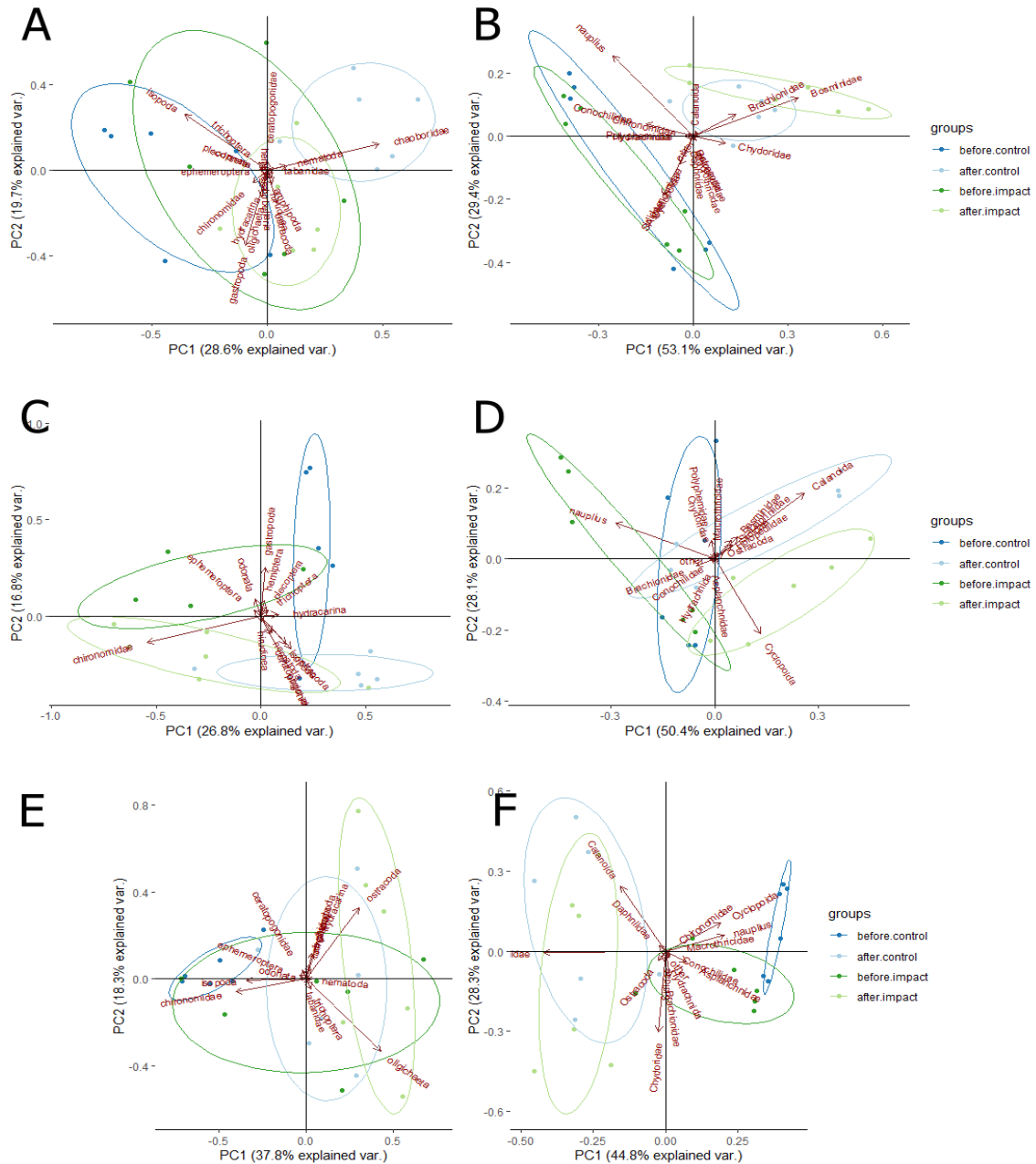


Figure 4-8. Ordination diagrams by PCA for Hellinger-transformed taxa counts for Dog Lake Macroinvertebrates (A), Dog Lake Zooplankton (B), Otter Lake Macroinvertebrates (C), Otter Lake Zooplankton (D), Wolfe Lake Macroinvertebrates (E) and Wolfe Lake Zooplankton (F). Individual taxa (identified to Family, where possible) are represented by arrows, with arrow length representing inertia along the first two PC axes. Dark blue points represent ‘control’ sites ‘before’ the treatment, dark green points represent ‘impact’ sites ‘before’ the treatment, light blue points represent the ‘control’ sites ‘after’ the treatment and light green points represent ‘impact’ sites ‘after’ the treatment. Likewise, 68% confidence interval ellipses are drawn from the centroids of all four groups using the same colour scheme. Percent variance explained for the first two PC axes are indicated in each plot.

*after*control* and the *before*impact* ellipses (a seasonal difference, as both are untreated vegetated sites). The taxa with the highest loadings on the first PC axis are the Chironomidae (−0.9033) and the Ostracoda (0.2425), whilst the taxa with the highest loadings on the second PC axis are the Oligochaeta (−0.5187) and the Gastropoda (0.5244). In the case of the Otter Lake zooplankton ordination (Figure 4-8D), there are two significant PC axes which explain 50.4% and 28.1% of the observed variance. The ellipses do not align with the first two PC axes and they are clustered close to the origin. Nevertheless, the only two ellipses that do not overlap are the *after*control* and the *after*impact* ellipses, which suggests a significant treatment effect. The taxa with the highest loadings on the first PC axis are the Copepoda nauplii (−0.6771) and the Calanoida (0.6081), whilst the taxa with the highest loadings on the second PC axis are the Cyclopoida (−0.6688) and again the Calanoida (0.5812).

For the Wolfe Lake macroinvertebrate ordination (Figure 4-8E), there are two significant PC axes which explain 37.8% and 18.3% of the observed variance. The ellipses again overlap considerably, with only *before*control* and the *after*impact* ellipses not overlapping (a potential treatment effect, but possibly confounded by seasonality). The taxa with the highest loadings on the first PC axis are the Chironomidae (−0.5386) and the Oligochaeta (0.5664), whilst the taxa with the highest loadings on the second PC axis are the Oligochaeta (−0.6314) and the Ostracoda (0.6275). In the case of the Wolfe Lake zooplankton ordination (Figure 4-8F), there are three significant PC axes which explain 44.8%, 28.3% and 13.6% of the observed variance. The two before groups are strongly separated from the two after groups along the first PC axis, suggesting very strong seasonal changes and little to no treatment effects. The taxa with the highest

loadings on the first PC axis are the Bosminidae (−0.7864) and the copepod nauplii (0.3807), whilst the taxa with the highest loadings on the second PC axis are the Chydoridae (−0.7107) and again the Calanoida (0.5737).

The Copepod nauplii, being a large and heterogenous mixture comprising of both Calanoid and Cyclopoid zooplankton larvae, are understandably the highest loading group for the ordinations across all three lakes. Nevertheless, the Calanoida, Bosminidae, Chydoridae, Sididae, Cyclopoida and Brachionidae (Rotifera) also play a large part in explaining zooplankton community structure. Insofar as macroinvertebrates (Figure 4-8A,C,E; Figure 4-9; Figure 4-10) are concerned, the observed assemblages seem most similar between Wolfe Lake and Otter Lake, most notably in terms of the Oligochaeta, Chironomidae and Ostracoda. Otter Lake and Dog Lake share the Gastropoda as an important part of their benthic macroinvertebrate community, whereas Dog Lake stands alone with a high importance assigned to the Isopoda, Chaoboridae and Hydrachnida. Indeed, phantom midges (Chaoboridae) were only observed in Dog Lake, and only in the macrophyte-rich control sites post-treatment.

In terms of individual taxa counts and abundances (Figure 4-10), the only two trends observed across all three lakes was a dominance by cyclopoid copepods amongst the zooplankton and a seasonal increase of oligochaetes amongst the benthic macroinvertebrates. Neither of these two traits seem to be related to treatment effects, as control-impact numbers remain similar post-treatment. For Dog Lake, chironomids, gastropods and isopods all declined from pre-treatment to post-treatment months, although there were little difference between control and impact abundances. Several taxa did show some potentially treatment-mediated related responses. Oligochaetes and

ostracods both not only increased over time (from 7 to 12% and 4 to 10% relative abundance) but more so in the impact sites over the control sites (18 % vs 1 % in oligochaetes and 20 vs 0 % in ostracods). Phantom midges, unique to dog lake and numerous there, increased from 11% relative abundance to a full 46% later on, with the bulk of these organisms preferring the vegetated control sites (70 % vs 22 % in the post-treatment months).

For Otter Lake, both seasonal and potential treatment variations were observed. Amongst the zooplankton, both Chydorids and Bosminids increased seasonally from less than 1% to 2 and 4%, respectively. The Brachionid rotifers conversely declined over the four-month period. The Sididae, while represented by smaller numbers increased from 1 individual per sample to 8 individuals per sample, with much more residing in the vegetated control sites (10 org. in the control sites vs 2 org. in the impact sites in the last month). In terms of macroinvertebrate numbers, while midges dominated, little change occurred seasonally or treatment-wise. In regards to noted responses, gastropods declined seasonally (from 12 to 0 % relative abundance) and ostracods increased (from 0 to 18 % relative abundance). The only taxa showing a potential treatment effect were the Oligocaheta, which increased seasonally (2 to 18%), but much more so in the control sites right after treatment (a surge to 52% relative abundance therein).

For Wolfe Lake, there were greater individual counts of Bosminidae in the pre-treatment months (117 organisms/sample) than after (19 org.), which is the opposite of what was observed in Dog and Otter Lakes, although control-impact differences were negligible. Calanoid copepod numbers exploded in the post treatment months (from 5 to 21 org.), with an especially large amount in the vegetated control sites (61 org.)

compared to the non-vegetated impact areas (25 org.) in the final month. Like in Otter Lake, Brachionid rotifers declined from about 76 organisms/sample to 6 organisms/sample, although the numbers between control and impact were similar. As to macroinvertebrates, there was a notable dominance of Chironomids, although there were no real trends attributable to treatment, just seasonality (decrease from 45 to 18%). Oligochaetes increased from 18 to 32 % relative abundance, and mites from 1 to 5%. Gastropods and Ostracods did show a small to response to the treatment, in both cases increasing not only overall (1 to 3% and 8 to 22 %, respectively), but more so in the impacted sites (10% and 55%, respectively). This increase in gastropods was again the opposite of what was observed in the other two lakes, where gastropods decreased.

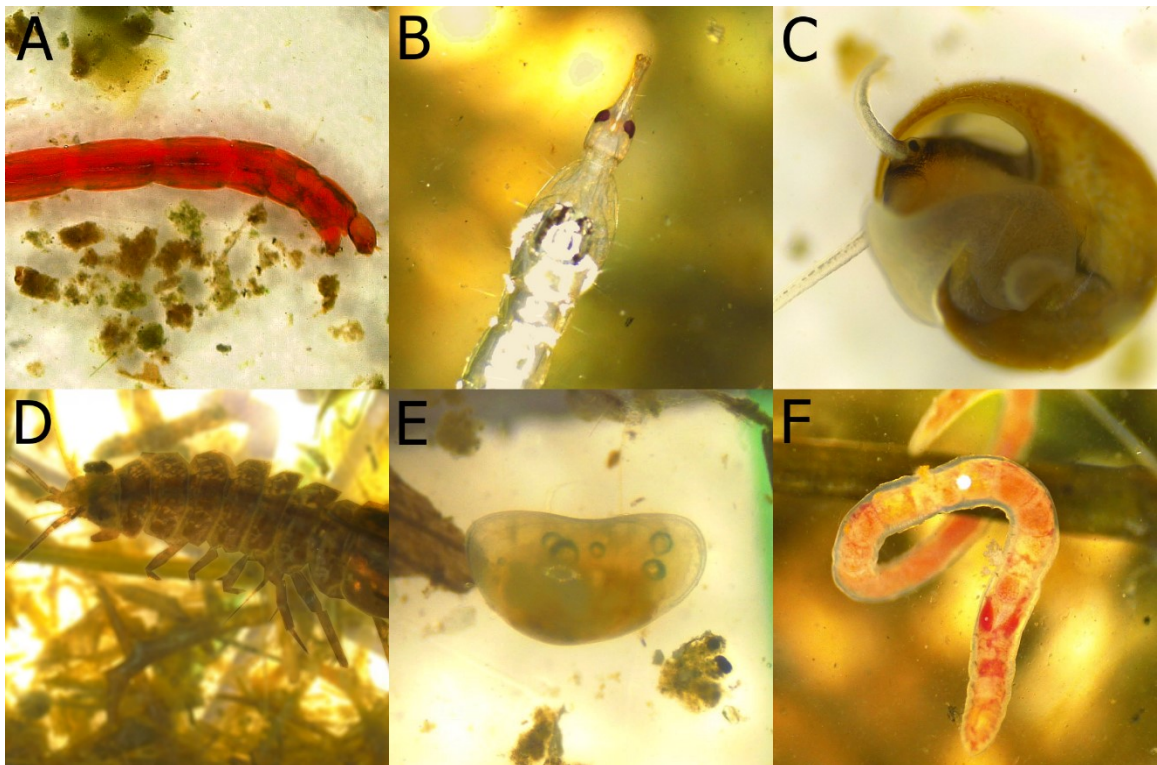


Figure 4-9. Six typical macroinvertebrate taxa encountered during this study: a larval midge (Chironomidae, A), a phantom midge (Chaoboridae, B), a snail (Gastropoda, C), an isopod (Isopoda, D), an ostracode (Ostracoda, E) and a worm (Oligochaeta, F).

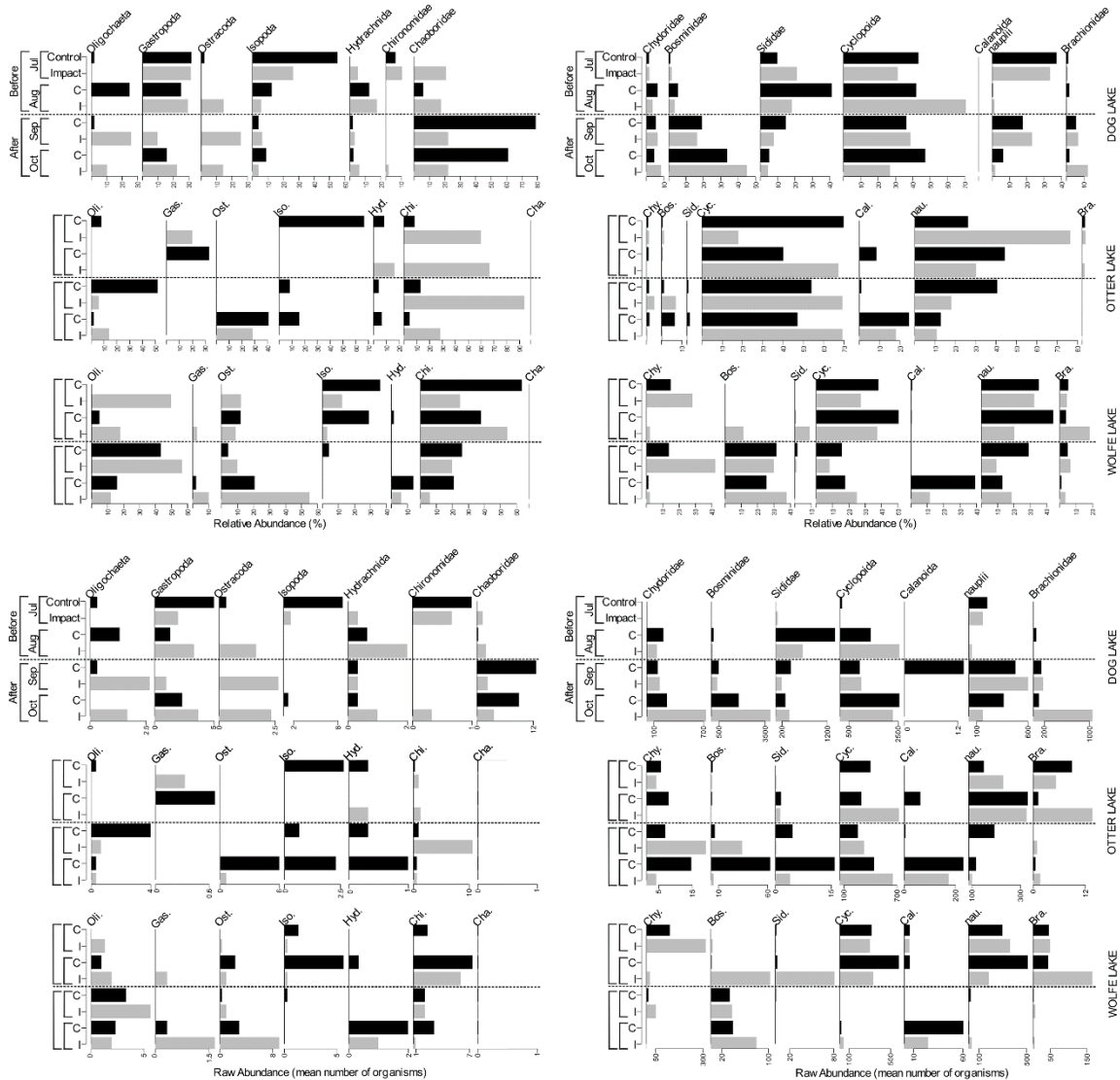


Figure 4-10. The six upper charts represent the mean ($n = 3$) relative abundance (%) of macroinvertebrate (left hand column) and zooplankton (right hand column) taxa, with rows organized by lake (Dog, Otter and Wolfe). Only the 7 taxa with the highest loadings on PC1, PC2 and PC3 from the ordinations (Figure 6) are shown. The six bottom charts follow the same organization, but represent raw counts (mean number of organisms per sample; macroinvertebrates in 0.5 L sediment samples, zooplankton in 22 L water samples) of the same taxa. Both relative and raw abundances are further divided by sampling time before (July and August) and after (September and October) the treatment date (dotted line), and by sample site type (control in black, impact in grey).

Chapter 5: Discussion

5.1 Discussion: Malcolm Lake

The first study suggests that the localized impacts of macrophyte control using burlap benthic barriers on the physical and chemical properties of Malcolm Lake were minimal. There were no significant differences in measured abiotic parameters between the control and impact sites pre- and post-treatment. Where consistent results were observed (e.g., where the means were either always increasing or decreasing over time, and the control-impact difference did not change sign), in the case of dissolved oxygen, pH and conductivity, the trends were hardly deleterious, with conditions in the impact trenches approaching those of the bare sediment environment (higher oxygen levels and pH, lower conductivity). The results do indicate, however, that shifts in zooplankton communities did occur with the treatment, which could have ecological implications if benthic barriers were to be employed at a larger scale.

Many of the environmental concerns involving the use of benthic barriers relate to the decomposition of the plants under these barriers. Plant material pressed into the sediment has been shown to decompose rapidly, causing a localized depletion of dissolved oxygen at the sediment-water interface (Carpenter 1980; Boston and Perkins 1982). This drop in dissolved oxygen has also been shown to be concomitant with a drop in pH (Boston and Perkins 1982), having ramifications for acidity-regulated nutrient uptake and release; namely a release of phosphorus from the sediment into the water column (Carignan 1985; Jensen and Andersen 1992; Hupfer and Lewandowski 2008). This could be further exacerbated by the release of phosphorus locked in the decaying

plants themselves, with the potential to lead to even more nutrient-enrichment, as has been shown by early models relating to the decay of herbicide-treated macrophytes (Nichols and Keeney 1973; Carpenter and Adams 1978; Carpenter 1980). That said, the use of porous semi-barriers instead of impermeable barriers means that gases need not be trapped under the barriers, as shown by Gunnison and Barko (1992), but rather allow the free exchange of gases between the sediment and water (Mayer 1978; Caffrey et al. 2010). Due to the blanketing effect of the mats on the sediment, resuspension of sediment has been shown to be greatly suppressed, at least in the case of impermeable barriers (Boston and Perkins 1982). The results of this study show not only no significant difference between total phosphorous, total Kjeldahl nitrogen nor dissolved organic carbon between control and impact sites post-treatment, but also that these values were nearly identical for both TP and TKN. Furthermore, algal proliferation (as estimated by Chl-*a*), a usual response to readily available phosphorus (Schindler 1974), was not statistically different between treatment and control sites. Conversely, Carpenter and Greenlee (1981) showed that lakes with rapid littoral flushing are less prone to deoxygenation, which in turn can prevent phosphorus release at the sediment-water interface (Boros et al. 2011). Given that Malcolm Lake is a shallow lake, these results suggest that its well-mixed waters are unlikely to see any serious negative impacts to nutrient dynamics at the sediment-water interface due to the placement of benthic barriers. If anything, the barriers may even help water move more freely, offsetting waterflow impairments caused by the dense stands of *M. spicatum*.

One finding warranting further study was the inverse but lock-step response of pH and conductivity in both control and impact sites. pH declined over the four-month study

period, while conductivity increased during the same time, with this effect being greater in magnitude in the control sites. This result also shows a slightly different trend than is typical in North America, where pH and conductivity have both been increasing as part of salinization syndrome, whereby road salt and agricultural runoff increase freshwater salinity (Kaushal et al. 2018). The catchment for Malcolm Lake is predominantly undeveloped land in a natural state, with little agriculture and only one paved road in the vicinity, suggesting that pH and conductivity may be macrophyte-regulated rather than salt-induced. *M. spicatum* has been shown to lower pH at the sediment-water interface through respiration and ammonium uptake, both processes that release H⁺ ions (Carignan 1985). Why ammonium uptake is occurring with a rise in conductivity rather than a drop warrants closer inspection. However, these results show that this decline in pH and increasing conductivity is occurring in the stands of macrophytes and that the benthic barriers may actually retard this effect. This is of particular interest for its ramifications to aquatic food webs. Pockets of high temperature and low pH in macrophyte stands have been shown to be detrimental to fish (Miranda et al. 2000), whereas conductivity has been shown to be negatively correlated with cladoceran zooplankton size and abundance (Kuczyńska-Kippen 2009; Brysiewicz et al. 2017). Dissolved oxygen and pH have been shown to influence copepod zooplankton distribution (Nurminen and Horppila 2002; Kuczyńska-Kippen 2009; Kuczyńska-Kippen and Joniak 2010), so the observed changes in acidity and conductivity could potentially have a further, but indirect impact on the zooplankton community.

Not only was the density of zooplankton between control and impact sites significantly different before and after treatment, the composition of these assemblages

also changed, in particular amongst the macrophyte-associated taxa. Seasonal changes were observed in this study, but in the literature these have been shown to be not as important as the influence brought about by the presence, abundance and diversity of macrophytes in determining zooplankton community structure (Kuczyńska-Kippen et al. 2009; Kuczyńska-Kippen and Joniak 2016). The Chydoridae, a cladoceran epiphyte-grazing microcrustacean known to be heavily associated with macrophytes (Pennak 1966; Fairchild 1981; Vijverberg and Boersma 1997; Geraldine and Boavida 2004), were one of the six taxa most influenced by the benthic barrier treatment (Figure 5-1). Their relative abundance remained high within the macrophyte stands but declined within the impact trenches. A similar decline was seen with both members of the Sididae and Macrothricidae, larger cladoceran filter feeders that attach temporarily to macrophytes, especially during the day (Fairchild 1981; Lauridsen et al. 1996). The Bosminidae as a family generally display a less uniform association with macrophytes than the Chydoridae, Sididae and Macrothricidae, but they have been found in greater numbers in the littoral as opposed to the pelagic (Balcer et al. 1984). While these results are unsurprising given that zooplankters are motile and habitat-dependent, it indicates a change to community composition occurred despite the close proximity of the control and impact sites. Cyclopoid copepods were the most numerous of the taxa found in this study, and they were found in both the macrophyte beds and in the impact trenches in similar numbers, while rotifers were more numerous in the bare sediment environment and the impact trenches. Lakes where smaller copepods (cyclopoids instead of calanoids) and rotifers are favoured in the pelagic are often synonymous with the presence of planktivores (Timms and Moss 1984; Lauridsen et al. 1998; Kuczyńska-Kippen and

Joniak 2016). While this pattern was observed, the lack of fish data prohibits any definitive linkage in this regard. Nevertheless, there is a potential disruption to fish-zooplankton-phytoplankton interactions brought about by the benthic barrier treatment. Timms and Moss (1984) showed that while fish pressure has the largest impact on zooplankton-phytoplankton relations, macrophyte loss can still directly reduce rare taxa. As macrophyte presence, density and type also impact fish distribution (Crowder and Cooper 1982; Savino and Stein 1982; Keast 1984; Weaver et al. 1997; Miranda et al. 2000; Cross and Mcinerny 2006; Meerhoff et al. 2007), the potential impact to zooplankton-phytoplankton interactions could potentially be magnified. That said, it is important to note that the scale of this study was very small when the treatment area is compared to the selected macrophyte patch (~1%), and incredibly small compared to the entire lake (~0.01%).

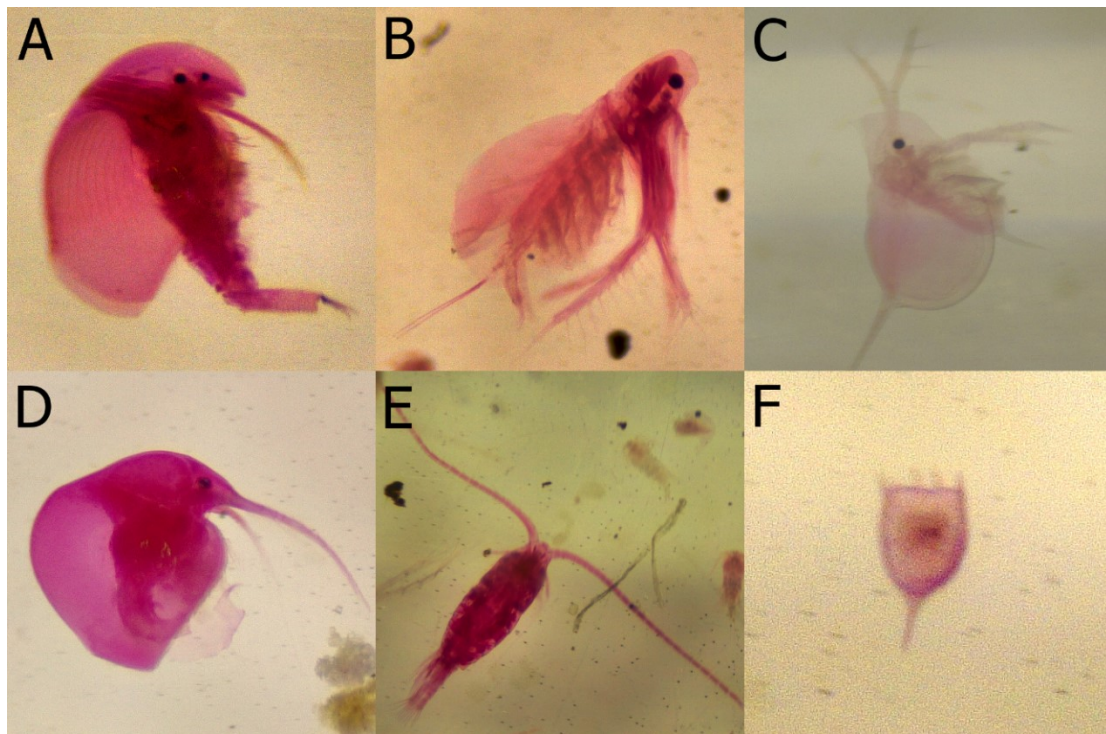


Figure 5-1. Six typical zooplankton taxa encountered during this study: Chydoridae (A), Sididae (B), Daphniidae (C), Bosminidae (D), Temoridae (Calanoida) (E) and Brachionidae (Rotifera) (F).

Macrophytes have been shown to act as an effective zooplankton refuge against fish predation (Schriver et al. 1995; Mamani et al. 2019) even when large planktivorous fish populations are present (Stansfield et al. 1997). More specifically, *M. spicatum*'s finely divided leaflets provide a more complex structural habitat and thus can act as a preferential zooplankton refuge (Bergström et al. 2000). While this refuge effect can be hampered by macroinvertebrate predators (Sagrario et al. 2009; Mamani et al. 2019) such as phantom midges (Chaoboridae) and mites (Hydrachnida), very few of these taxa or their preferred daphnid and calanoid prey (Luecke and Litt 1987) were found in our samples. This indicates that the macrophyte stands of Malcolm Lake may actually increase zooplankton diversity, and that the burlap benthic barrier treatment may cause a reduced value to zooplankton. Malcolm Lake is quite shallow, only 4.6 meters at its deepest point, meaning that the zooplankton in this lake may be more likely to display Diel Horizontal Migration than Diel Vertical Migration, whereby zooplankton take refuge in littoral macrophyte beds during the day and migrate to open water to graze at night, instead of rising from the (here, non-existent) deeps (Jeppesen et al. 1998; Lauridsen et al. 1998). Diel Horizontal Migration is also more likely to be favored in shallow lakes, as well as in lakes with less extreme gradients of abiotic factors, of which Malcolm Lake is certainly an example (Burks et al. 2002). While no night sampling was done in this study, the species composition of Malcolm Lake was nevertheless consistent with a lake displaying Diel Horizontal Migration and having both healthy macrophyte and fish populations (Brooks and Dodson 1965; Schriver et al. 1995; Lauridsen et al. 1998). One inconsistency however was that the large bodied cladocerans were limited to the Sididae and Macrothricidae, and that the Daphniidae were almost entirely absent,

despite the presence of ample cover within which to hide (Davies 1985). It can only be surmised that the low nutrient and phytoplankton concentrations meant there were little food resources for grazing daphnids. Another possible driver of this absence was the rising conductivity, especially within the macrophyte beds. Of all abiotic factors, conductivity has been shown to have the greatest negative correlation with cladoceran size and population density (Brysiewicz et al. 2017). Low pH has also been shown to have a similar effect (Nurminen and Horppila 2002), although Malcolm lake is far from being an acidic lake.

Closely tied to littoral zooplankton and macrophyte community dynamics are those of fish. While no quantitative estimates of fish biomass or community were collected in this study, both bluegills (*L. macrochirus*) and small-mouthed bass (*M. dolomieu*) were observed at greater densities swimming in the impact trenches post-treatment. Dense monoculture macrophyte stands have been shown to have negative impacts on the foraging success and ultimately on the size and abundance of desirable/commercial fish species (Cross and Mcinerny 2006; Valley and Bremigan 2011). More specifically, *M. spicatum* has been shown to harbour less fish and macroinvertebrate diversity than equivalent native macrophyte species (Keast 1984). Weaver et al. (1997) showed that habitat complexity is important for fish diversity, and that the cutting of trenches in macrophyte beds can actually improve growth rates of some centrarchid fish (Olson et al. 1998). The impact of benthic barriers to fish community structure is potentially direct via macrophyte removal, as well as indirect through zooplankton community re-arrangement. Given that top-down processes are

particularly potent in shallow lakes (Jeppesen et al. 1997), the role and impact on fishes warrants much closer attention in macrophyte control (Radomski et al. 2019).

Another area needing further investigation is the impact of benthic barriers on macroinvertebrates. Malcolm Lake was unfortunately a poor lake in this regard, as the analysed samples held little more than the empty shells of gastropods, and very few living specimens of any kind. Synthetic barriers have been shown to have drastic impacts on macroinvertebrates, eliminating all of them in some treated areas (Ussery et al. 1997). Conversely, decaying macrophytes provide an abundant food source and have been shown to increase macroinvertebrate abundance (Smock and Stoneburner 1980). It has been proposed that permeable benthic barriers, such as the burlap matting used in this study, would avoid the negative effects (Caffrey et al. 2010), however this has not been shown convincingly in the field. Further studies should be performed to better assess the impacts of burlap benthic barriers on macroinvertebrate density and diversity.

While these findings suggest that the overall impacts of macrophyte control via the application of burlap benthic barriers to localized littoral abiotic factors were negligible, we cannot ignore that despite being small-scale and localized, changes to species assemblages of zooplankton did occur. A review by Chambers et al. (1999) eloquently outlines the need to maintain a scientific perspective when making management decisions in regards to macrophytes, and ultimately, aquatic ecosystems. Macrophyte control, as with all forms of ecosystem management often includes a conflict of interest between social and commercial perspectives with ecological perspectives (Clayton and Tanner 1988). While there is little doubt that invasive species, including *M. spicatum*, can have dramatic effects on freshwater ecosystems (Keast 1984; Boylen et al.

1999; Kelly and Hawes 2005; Bosch et al. 2009; Matsuzaki et al. 2009), it has also been shown that many of these alien invasives become an entrenched part of ecosystems (Carpenter and Lodge 1986; Smith and Barko 1990; Kowalczewski and Ozimek 1993; Williamson 1999; Donadi et al. 2018). In British Columbia, Canada, *M. spicatum* has proven to be incredibly resistant to eradication in all but the smallest water bodies (Newroth 1993), leading some lake managers towards a new more integrated approach to macrophyte control (Radomski and Perleberg 2019), with greater emphasis on macrophyte removal only in specific areas and allowing them to grow unchecked in others. With recent advances to our understanding of the importance of spatial heterogeneity at different scales, there is mounting evidence that macrophytes, even invasives, provide more ecosystem services than previously thought (Chambers and Prepas 1990; Folke et al. 2004; Baron et al. 2005; Valley and Drake 2007; O'Hare et al. 2018; Verhofstad and Bakker 2019). Thus negative impacts caused by invasives like *M. spicatum* to certain fish species (Keast 1984) and native macrophytes (Boylen et al. 1999) must be balanced against the potential ecosystem services provided by these same newcomers. Granted the services provided by monoculture stands will likely be less than more species-rich stands (Engelhardt and Ritchie 2001), the small scale complexity of *M. spicatum*'s finely divided leaflets has been shown to be a better microhabitat than some more simply organized native macrophytes (Crowder and Cooper 1982; Bergström et al. 2000; Mcabendroth et al. 2005; Cross and Mcinerny 2006). With the global rise of algal blooms and intense pressures on freshwater systems (Ho et al. 2019; Reid et al. 2019), the place of macrophyte management becomes a more complex balance between eradication, control and in some cases, even restoration (van Nes et al. 2002). It is my opinion that

more studies on the direct and indirect impacts of macrophyte control to freshwater ecosystems, at varying spatial and temporal scales, will be instrumental in better making management decisions.

5.2 Discussion: Rideau Lakes

My results suggest that the mechanical cutting of invasive macrophytes in small areas (e.g. 100 m² patches) has little impact on the local physical, chemical and biological components of the littoral environment, apart from on the macrophytes themselves. While significant differences were observed in some abiotic parameters between the control and impact sites pre- and post-treatment, these differences were either small and/or inconsistent across lakes. Furthermore, while some individual taxa did respond to the treatment, none of these community changes were echoed across all three lakes, and overall zooplankton and macroinvertebrate densities remained mostly similar. These differences also did not vary along a nutrient gradient. That said, the lake with most significant responses among individual zooplankton and benthic macroinvertebrate taxa was eutrophic Dog Lake, although it did also have the most invertebrate taxa and counts overall.

Two major environmental concerns stemming from the use of mechanical cutting to control *M. spicatum* are the spread of fragments that could promote recolonization elsewhere in the lake (Aiken et al. 1979; Eiswerth et al. 2000a) and the resuspension of disturbed sediment in the cut area (Holmroos et al. 2015). In the case of the fragments, some aquatic weed harvesters, such as the one used in this study, are equipped with conveyor belts, allowing them to not only remove all macrophyte biomass, but also do

several passes to scoop up loose fragments floating at different depths in the water column. While smaller fragments could still escape, this first issue is unlikely to pose a serious threat in lakes that already have established *M. spicatum* infestations, the most likely candidates for treatment in Eastern Ontario. The second issue could be of greater concern, especially in already nutrient enriched lakes. During the harvesting process in this study, much sediment was churned into the water column, temporarily increasing turbidity, but also possibly resuspending phosphorus originally trapped in the sediment. That said, only two days later phosphorus levels in the water column were the same within and without the treated area, suggesting that small treatments like this are unlikely to contribute meaningfully to nutrient enrichment and subsequent algal proliferation via sediment resuspension and/or biofilm dislodgement.

A different mechanism for algal proliferation, via the removal or reduction of allelopathic control by macrophytes, remains somewhat controversial, but is worthy of consideration. Allelopathic chemicals have been identified in *M. spicatum*, and mesocosm experiments have shown that *M. spicatum* is able to inhibit algal growth (Gross et al. 1996; Nakai et al. 2000). The controversy begins in that it is difficult to observe such suppression in a natural setting, and at larger scales. Nevertheless, the theory of alternating stable states in shallow lakes, espoused by many researchers (Scheffer 1990; Scheffer and Jeppesen 2007; Scheffer et al. 2012) hinges on a clear water, macrophyte-dominated state being antagonistic to an algal-rich, macrophyte-free state. Regardless as to the mechanism by which these states are reached, it has been shown that macrophyte restoration can reduce the algal biomass in a body of water (Jeppesen et al. 1998; Meerhoff et al. 2007; Vilas et al. 2018; Barrow et al. 2019),

insinuating that a removal of macrophytes could cause a return to an algal-dominated state (Morris et al. 2006; Hilt et al. 2017). Thus, another concern of macrophyte control is the indirect influence it could potentially have on algal biomass, which could potentially be more serious in an already stressed eutrophic system close to a tipping point. Therefore, it appears more and more likely that macrophyte control could potentially be at cross-purposes with water quality control, as espoused by Van Nes et al. (2002) and more recently by Kuiper et al. (2017). Both Dog Lake and Wolfe Lake showed one month where the impact sites had significantly higher Chl-*a* in the impact sites than the control sites post treatment (Figure 4-5). While the differences were small, and only significant in two of six ‘before vs one month’ asymmetrical tests, this is a cause for concern considering an impact was detected at all for such a small-scale test, and up to a month later in the case of Dog Lake. Ecosystem collapses or movement from one stable state to another does not necessarily need to come from large perturbations, and recent models by van de Leemput et al. (2018) suggest that even small disturbances can cascade and ultimately lead to state changes. Work on looking for early warning signs of regime shifts suggests that both large swings in variance pre-collapse and slower recovery from perturbations could be heralds of impending collapse (Drake and Griffen 2010; Carpenter et al. 2011b). While this study did not have the sample size or sampling frequency to look for changes in variance at an appropriate resolution, precisely one example of slower recover in a hypothetically less resilient site was observed: Wolfe Lake saw an immediate rise in algae post treatment and a subsequent return to normal a month later, whereas Dog Lake Chl-*a* levels at first did not change but then rose later and did not return to normal during our study period. This is of course largely speculative in this instance, as Otter

Lake did not at all mirror the Wolfe Lake pattern. Nevertheless, it highlights the possibility that such mechanisms may be at work here and warrants further investigation, especially at larger temporal scales. In the context of *M. spicatum* control in Eastern Ontario, while treatments are generally small, cottage-front shoreline modifications, multiple such control efforts undertaken simultaneously must thus be taken more seriously. In larger, deeper lakes, such as Wolfe Lake and Otter Lake, this will likely be less important than it would in shallower, nutrient enriched lakes like Dog Lake (regions of Dog Lake are deep, but there are large, shallow areas resulting from the creation of the Rideau Waterway). That said, there is some evidence that suggests that medium-sized temperate lakes could be more susceptible to regime shifts than shallow lakes (Genkai-Kato and Carpenter 2005), which should serve as another warning.

Chapter 6: Conclusions

In the Malcolm Lake burlap benthic barrier study, the impacts of the deployment of burlap mats had no detectable influence on measured physical and chemical lake parameters (temp, light, cond, DO, DOC, pH, TKN, TP). While the treatment did not elicit a response in the first biological parameter (Chl-*a*), it did provoke a response in zooplankton density, where there were significantly less organisms in the impact trenches post-treatment than there were in the vegetated control sites. Furthermore, raw and relative abundances were noticeably different for several taxa, especially macrophyte-associated groups such as the Sididae, Chydoridae and Macrothricidae. Ordination analysis by PCA suggests that control and impact groups were separated post treatment, and that conditions in the impact trenches had much overlap with the open-water bare-sediment locations. These findings suggest that the burlap benthic barrier treatment had little adverse effect on the overall ecosystem apart from returning it closer to ‘normal’ pre-*Myriophyllum spicatum* infestation conditions.

In the Rideau Lakes mechanical cutting experiment the harvesting treatment had very little effect on the physical, chemical and biological parameters measured. The temperature differences within and without the treated areas was significantly different for Wolfe lake alone, with impact areas being minutely warmer. The pH was significantly different between the post-treatment control and impact locations for Otter and Dog Lakes, but the sign of this change was opposite, providing little opportunity for a consistent explanation. There were changes to the zooplankton and macroinvertebrate communities, but many of these changes seem more closely linked to seasonality than to treatment effects, and where differences were observed, they were not consistent across

lakes, and in fact, often opposite. The exception to this would be among the Calanoida, who were more numerous in the control over impact areas post-treatment across all three lakes.

Insofar as a nutrient gradient is concerned, the lack of significant differences occludes the opportunity to study the magnitude of these absentee differences. Indeed, that the three Rideau lakes had exactly one significant difference each is telling. That said, the response of Chl-*a* in Dog Lake (Figure 4-5 A) and Wolfe Lake (Figure 4-5 G) warrants further attention, as the response is exactly what one would predict according ecosystem resilience theory. The more eutrophic system (Dog Lake) was slower to change and did not recover from the disturbance (a greater increase in algal biomass in the impact sites) during the monitoring period. Conversely, the more oligotrophic system showed a rapid response to the treatment but was fully recovered one month later, the hallmark of a more resilient system.

From these findings, several future trajectories can be planned. Certainly this study would have benefitted from greater replication in time before and after the treatment date, at least two field seasons before, and two thereafter (and thus outside the scope of a typical MSc thesis). The greater temporal scale would likely also require greater treatment areas, although this is not necessarily consistent with the studies *raison-d'être*: finding the impacts of localized treatments that mirror what is usually undertaken by landowners obtaining treatment permits from regulatory authorities. Also, I believe it would be both cost and time-effective to drop all nutrient monitoring and macroinvertebrate sampling, as these activities are not only costly and time-consuming, but they provided no returns in knowledge whatsoever in this study. In future studies I

would focus entirely on zooplankton density and taxa, pH and chlorophyll-*a*. Given that other abiotic factors such as conductivity, temperature, dissolved oxygen, redox potential and turbidity can easily be monitored concomitantly with the same equipment as pH (e.g. a typical YSI multimeter) there would be little harm in keeping these data as well. It would be interesting to sample more lakes across a greater range of trophic levels, but I believe this would be best suited to a separate study focusing entirely on chlorophyll-*a*. Extending drone coverage to all parts of the study would also have been very helpful as it provides for more accurate area calculation and orientation in general. Several other directions for future study include, but are not limited to: collecting zooplankton in the day and at night, installing underwater fish cameras and/or employing electrofishing to survey fish species within and without the macrophyte beds and lastly, looking into larger spatial scale mapping/monitoring using the aforementioned drone technology. Comparable studies with native macrophyte beds, while interesting and even likely helpful, would probably be best left alone given the already invasive-stressed state of lakes in the region.

In conclusion, my findings suggest that the overall impacts of macrophyte control, whether via mechanical cutting or the application of benthic barriers, to localized physical, chemical and biological factors were minimal. However, it cannot be ignored that some changes did occur even at this small scale. While there does not appear to be more than a speculative link between the treatment and eutrophication dynamics across a nutrient gradient, a degree of caution should nevertheless be adopted. The fact that local algal biomass did change significantly both in the short term for Wolfe lake and the medium term for Dog Lake warrants closer inspection. Also, while no consistent pattern

for macroinvertebrate and zooplankton community assemblage changes were elucidated by this study, significant taxa population changes were observed in both eutrophic and oligo/mesotrophic systems. It is my hope that these findings will serve as a guide to precipitate more studies with narrower focus and a higher sampling resolution in future. I believe that the macrophyte-phytoplankton dynamics, as seen across a nutrient gradient, will gain in importance in the Canadian context as climate change and anthropogenic pressures increasingly threaten our already stressed freshwater systems.

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Appendices

Appendix A – Correlation Plots

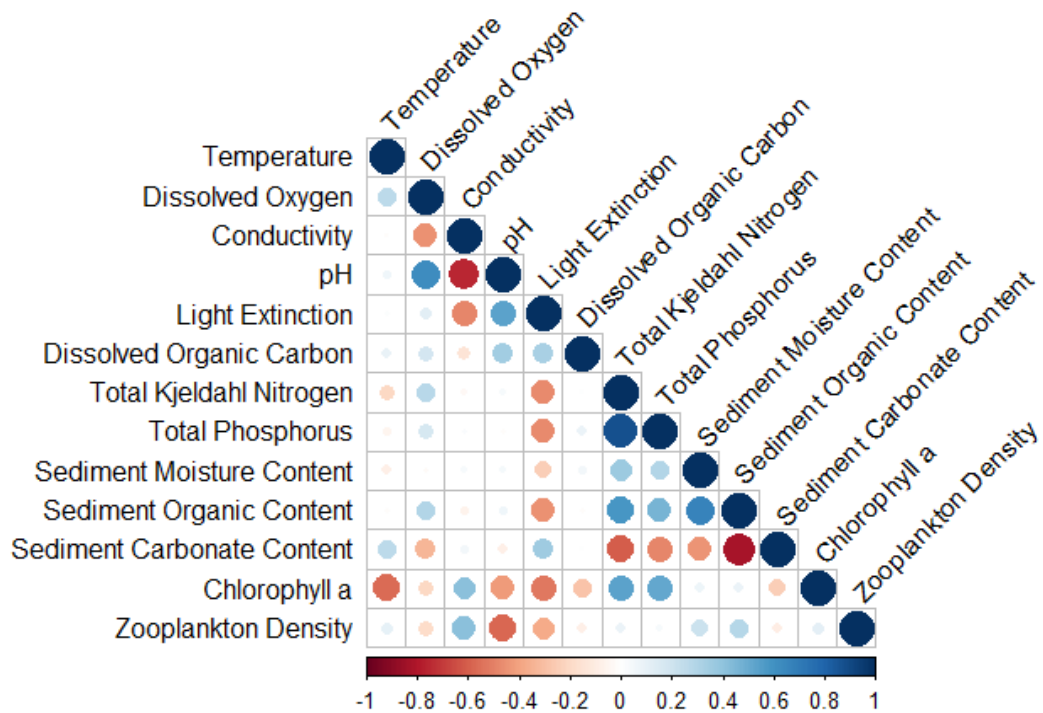


Figure A-1. Correlation matrix for all physical, chemical and biological parameters measured across all three sites (control, impact, bare sediment) in Malcolm Lake. Positive correlation is shown in blue and negative correlation in red, with larger sized circles and more darkly shaded circles indicating a stronger relationship.

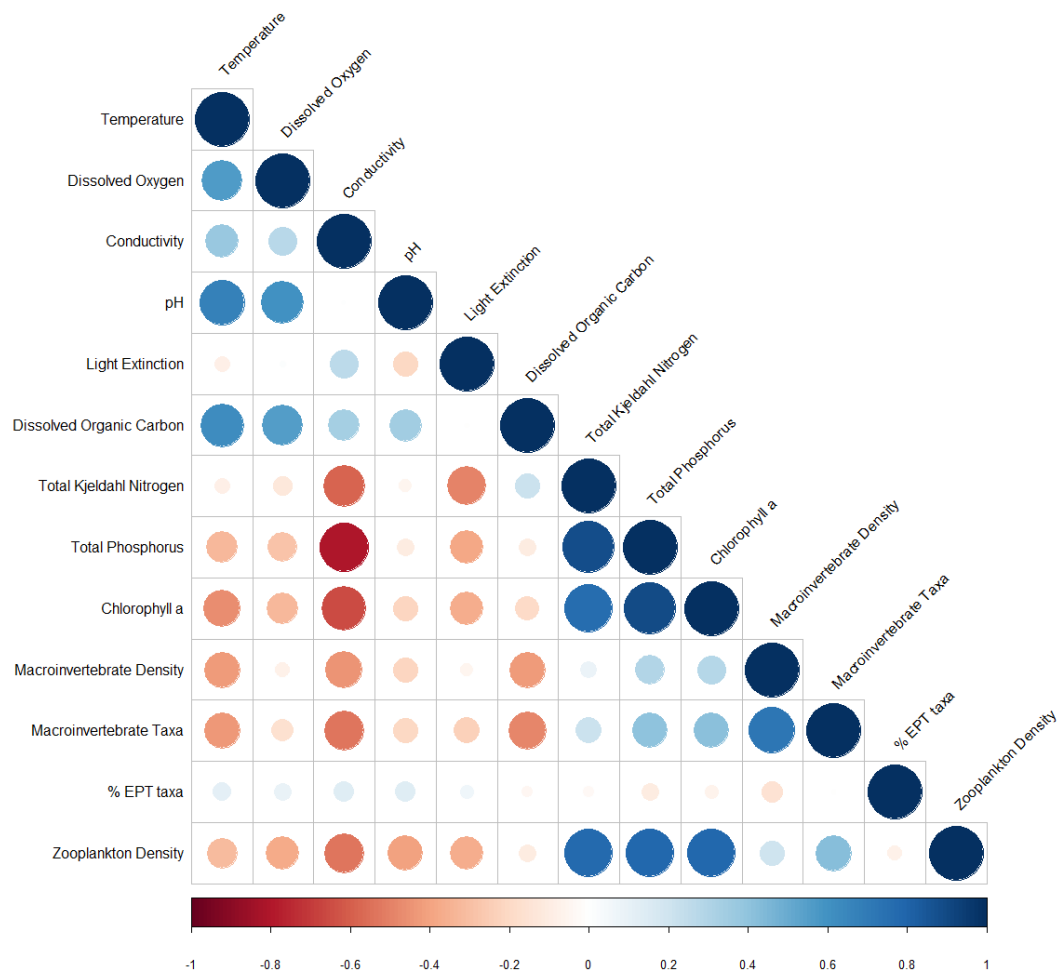


Figure A-2. Correlation matrix for all physical, chemical and biological parameters measured across both site types (control, impact) in all three Rideau Lakes (Dog, Otter and Wolfe). Positive correlation is shown in blue and negative correlation in red, with larger sized circles and more darkly shaded circles indicating a stronger relationship.

Appendix B – Scree Plots

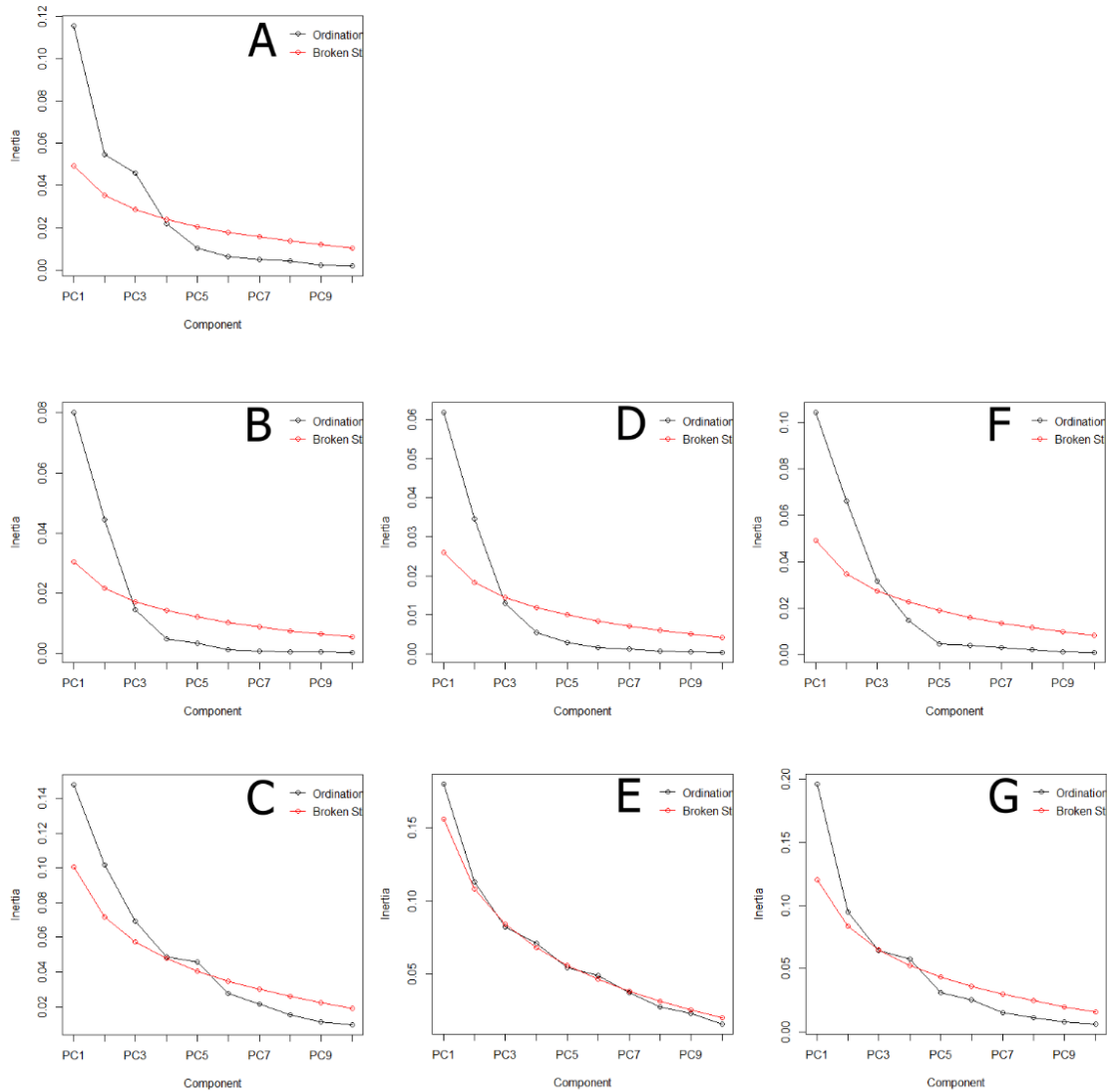


Figure B-1. Scree plots for ordinations by PCA for Malcolm Lake Zooplankton (A), Dog Lake Zooplankton (B), Dog Lake Macroinvertebrates (C), Otter Lake Zooplankton (D), Otter Lake Macroinvertebrates (E), Wolfe Lake Zooplankton (F) and Wolfe Lake Macroinvertebrates (G).

Appendix C – Additional Ordinations

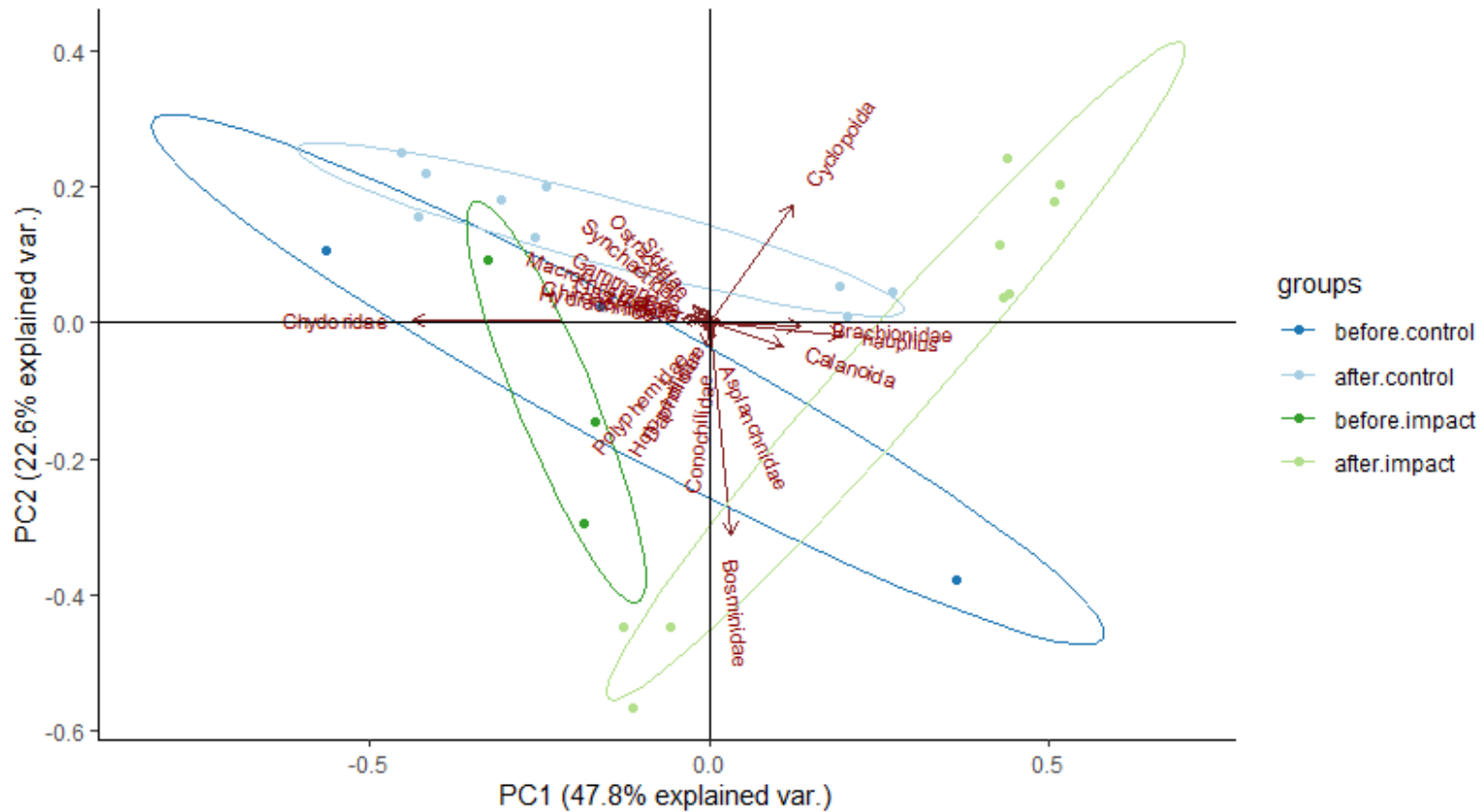


Figure C-1. Ordination diagram by PCA excluding all open water / bare sediment data for 24 sample units based on Hellinger transformed zooplankton taxa counts for Malcolm Lake. Individual taxa (identified to Family, where possible) are represented by arrows, with arrow length representing inertia along the first two PC axes. Dark blue points represent ‘control’ sites ‘before’ the treatment, dark green points represent ‘impact’ sites ‘before’ the treatment, light blue points represent the ‘control’ sites ‘after’ the treatment and light green points represent ‘impact’ sites ‘after’ the treatment. Likewise, 68% confidence interval ellipses are drawn from the centroids of all four groups using the same colour scheme. Percent variance explained for the first two PC axes are indicated in each plot.

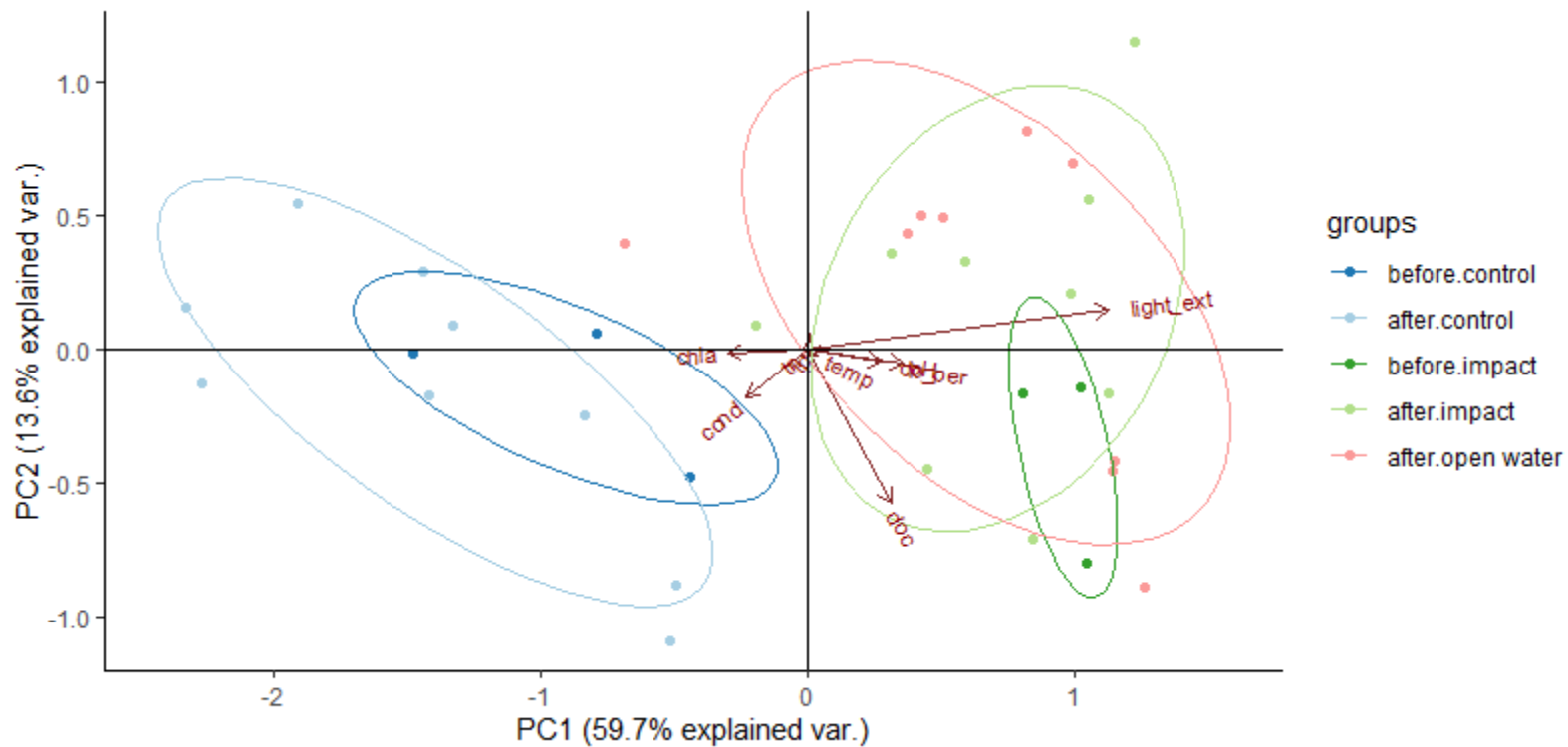


Figure C-2. Ordination diagram by PCA for the 33 sample units based on $\ln(x+1)$ transformed environmental variables for Malcolm Lake. Variables are represented by arrows, with arrow length representing inertia along the first two PC axes. Dark blue points represent ‘control’ sites ‘before’ the treatment, dark green points represent ‘impact’ sites ‘before’ the treatment, light blue points represent the ‘control’ sites ‘after’ the treatment and light green points represent ‘impact’ sites ‘after’ the treatment. Pink points represent the bare sediment / open water environment after the treatment. Likewise, 68% confidence interval ellipses are drawn from the centroids of all five groups using the same colour scheme. Percent variance explained for the first two PC axes are indicated in each plot.

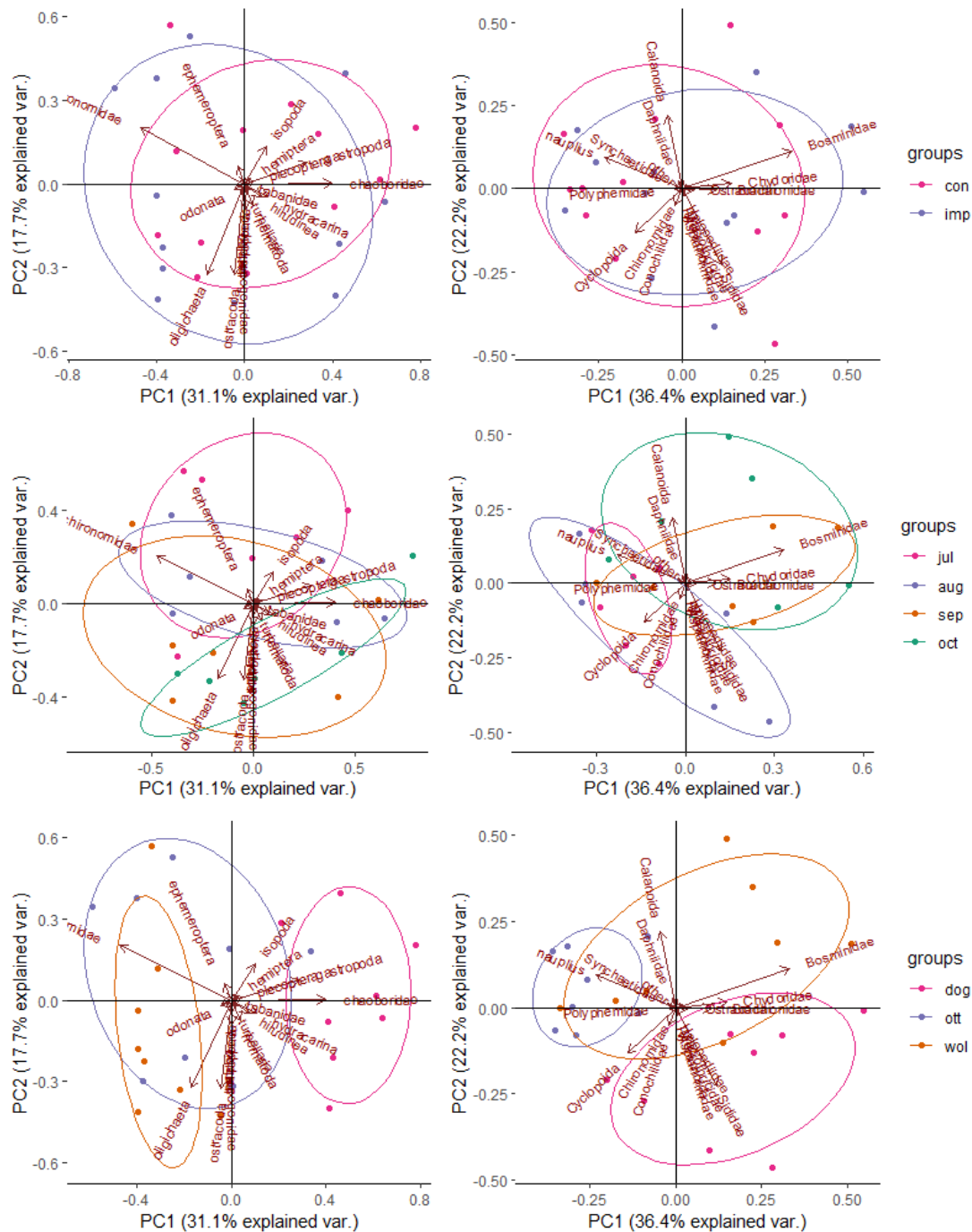
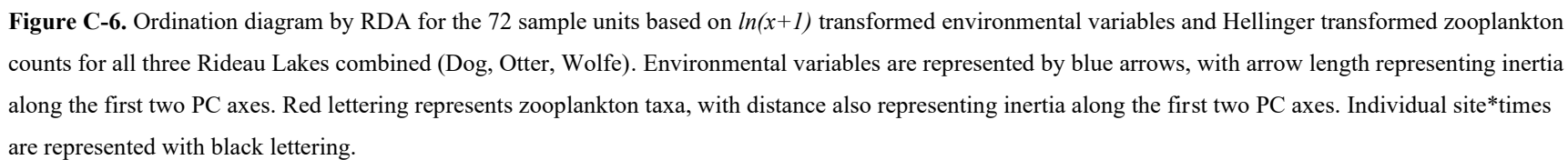


Figure C-5. Ordination diagram by PCA for the 72 sample units based on Hellinger transformed zooplankton counts (right column) and macroinvertebrate counts (left column) for all three Rideau Lakes combined (Dog, Otter, Wolfe). Taxa are represented by red arrows, with arrow length representing inertia along the first two PC axes. Percent variance explained for the first two PC axes are indicated in each plot. In the first plots (top left/right), points are separated into control and impact. In the second plots (middle left/right), points are separated by month and in the final plots (bottom left/right), points are separated by lake. Likewise, 68% confidence interval ellipses are drawn from the centroids of all groups using the same colour scheme as indicated in the legends.



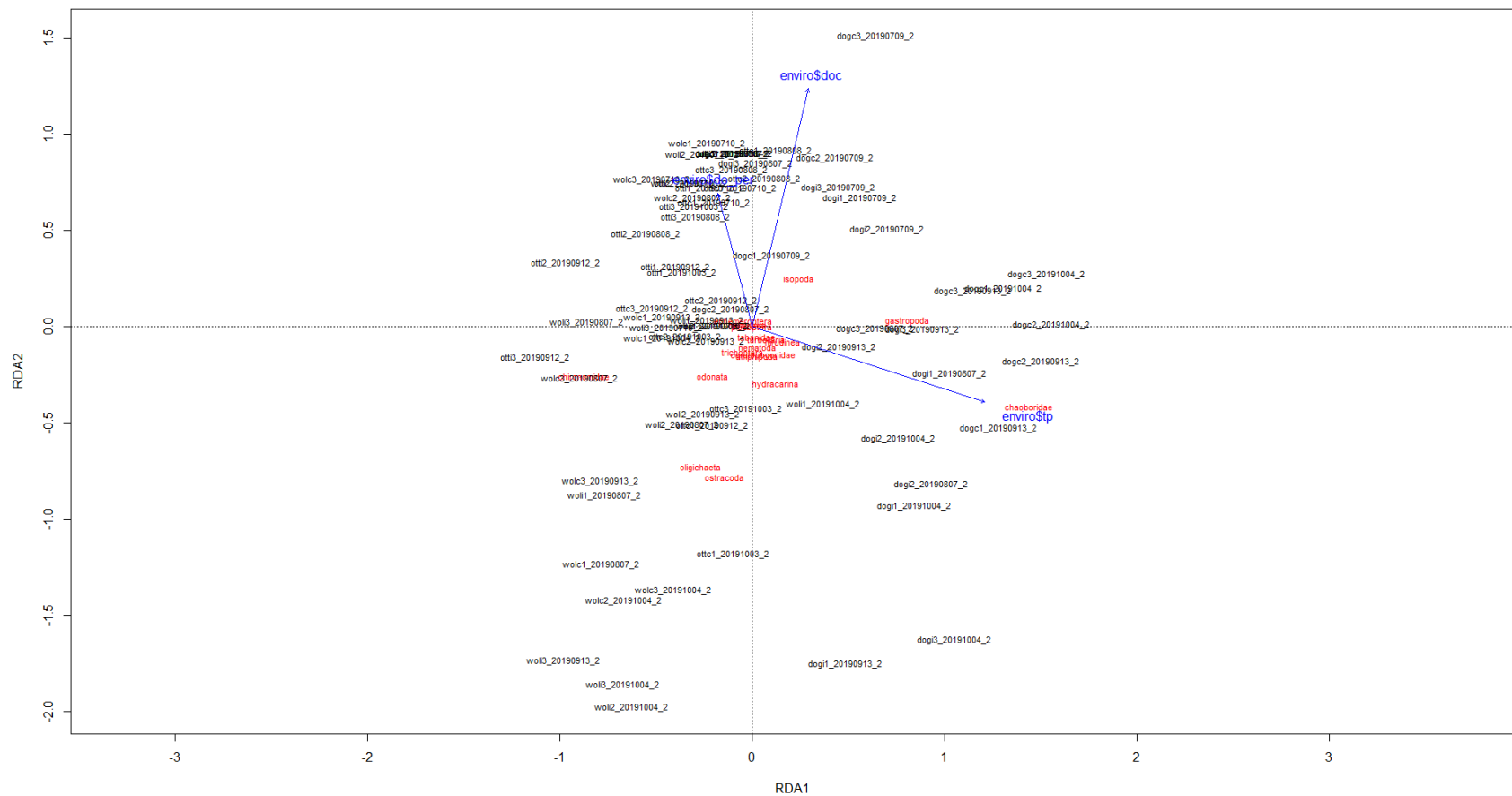


Figure C-7. Ordination diagram by RDA for the 72 sample units based on $\ln(x+1)$ transformed environmental variables and Hellinger transformed macroinvertebrate counts for all three Rideau Lakes combined (Dog, Otter, Wolfe). Environmental variables are represented by blue arrows, with arrow length representing inertia along the first two PC axes. Red lettering represents macroinvertebrate taxa, with distance also representing inertia along the first two PC axes. Individual site*times are represented with black lettering.