# Spatial and temporal assessment of rural-urban land-use gradient effects on water quality and periphyton communities in tributaries of Durham Region, Ontario

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In

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#### **General Abstract**

This study examines the effects of longitudinal and local land-use gradients on water quality and periphyton within four watersheds representing varying rural-urban land-use types and intensities. Although numerous studies have identified how specific land-use gradients (e.g., urban or agricultural land-use) affect water quality and periphyton, it is not fully understood how varying intensities and types of rural-urban land-use gradients affect water quality and algae both within and across watersheds sharing similar physiography and climate. Therefore, this study aimed to determine how variation in rural-urban land-use gradients affect water quality and periphyton along the cumulative flow path of tributaries (i.e., longitudinal), as well as across tributaries where sites were approximately matched for distance from headwaters. To assess spatial variation without the confounding effects of seasonality, I analysed water quality, algal biomass, and community composition from a complete and balanced set of algal growth substrates that were deployed in all study creeks (Lynde, Oshawa, Bowmanville and Soper) during May, 2015 (Chapter 2). Additionally, I examined the spatial and temporal variation of water quality and algal community structure in all tributaries from May – August, 2015 using a robust, but unbalanced dataset (Chapter 3).

Water quality variables in the month of May varied across the study sites and watersheds, however, trends were observed for pH, conductivity, chloride and temperature in each creek. In addition, sites closest to creek headwaters were found to be less impacted than developed downstream sites), indicating that agricultural land-use is less impactful to water quality than developed land-use in these watersheds. Along with water quality, chlorophyll *a* (chl *a*) and Ash Free Dry Mass (AFDM) were also found to vary among the sampling sites in May. Redundancy analysis (RDA) for the month of May showed statistically significant relationships with land-use

and water quality (999 random permutations, P<0.05). Within the RDA, road density was observed to have positive correlations with chloride, temperature, and TSS. Algal community composition among study sites were dominated by four genera, *Achnanthidium*, *Nitzschia*, *Navicula*, and *Gomphonema*. RDAs for land-use and water quality versus algal communities showed pollution tolerant taxa such as *Gomphonema* and *Cymbella* were associated with urban development and road density, and strongly driven by chloride.

Spatial and temporal trends for water quality across creeks was variable among the study sites and sampling months. Cumulative trends were observed for some water quality variables, which included chloride, pH, and TSS. Redundancy analysis with water quality variables against distance from headwaters and percent developed land-use revealed that both local land-use and distance from headwaters were influencing water quality. Algal community composition over the four month sampling period was dominated by the genera, Achnanthidium, Nitzschia, and Gomphonema. Algal community composition varied among the sampling months, however, some relationships were observed with water quality parameters. For example, chloride was observed to be positively correlated with Gomphonema and Cymbella consistently for every sampling month. Overall, my study indicated that land-use type and intensity can impact water quality and algal community structure. Longitudinal (i.e. cumulative) land-use impacts were observed for some water quality parameters, however, it seems that lateral inputs from local sources are influencing water quality to the same degree. This suggests that future land-use studies incorporate local land-use and point-source inputs in addition to longitudinal aspects to study design in order to account for the total environmental impact of the surrounding landscape.

Keywords: Land-use gradients, Spatial factors, Streams, Benthic algae, Water quality

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# **Abbreviations**

AFDM Ash free dry mass

CLOCA Central Lake Ontario Conservation Authority

Chl a Chlorophyll a

Cond Conductivity

DO Dissolved oxygen

DCA Detrended Correspondence analysis

ORM Oak ridges moraine

PCA Principal components analysis

RDA Redundancy analysis

Temp Temperature

TN Total nitrogen

TP Total phosphorus

TSS Total suspended solids

UOIT University of Ontario Institute of Technology

VIF Variation inflation factor

# **Chapter 1: General Introduction**

### 1.1 Land-use impacts to water quality

### **1.1.1** Agricultural land-use practices

Agricultural land-use practices are capable of influencing nearby streams through multiple mechanisms, including changing the natural landscape, addition of fertilizers, and addition of agricultural based chemicals (Murdock et al., 2013; Andrus et al., 2015). Many of these inputs are introduced into freshwater streams through rain events that can increase nutrients and sediment concentrations (Munn et al., 2002). The delivery of nutrients and sediment to surface waters are dependent on run-off volume due to rainfall, soil permeability, and rate of fertilizer application (Hoorman et al., 2008). The addition of nutrients such as, phosphorus and nitrogen are a major problem, primarily since it can stimulate algal growth and accelerate eutrophication in a stream (Coulliard & Li, 1993). One of the negative impacts of eutrophication is the reduction of aquatic biodiversity and water quality degradation (Maret et al., 2010). In addition, increased sediment loading can also affect aquatic organisms and water quality. Increased sedimentation into streams can increase turbidity and total suspended solids (TSS), which can limit light penetration, reduce primary productivity, and alter sediment composition found within a streambed (Jowett, 2003; Black et al., 2011).

# **1.1.2** *Urban land-use practices*

Urban development is one of the most pervasive and rapid land-use transformations worldwide. Urbanization alters landscapes through the replacement of natural landscapes with high areas of impervious surfaces (e.g., roads, parking lots, buildings and homes) (Porter-goff et

al., 2010; Gallagher et al., 2011; Wallace et al., 2013). According to Paul & Meyer (2001), increased imperviousness leads to increased surface runoff due to the loss of soil infiltration. Since impervious surfaces are so prevalent in urban settings it has become an excellent predicator of urban development and its associated impacts. Urbanization has been found to alter many natural characteristics within streams, including changes in channel morphology and hydrology, altered water chemistry, and reductions in biodiversity (Paul & Meyer, 2001; Sonneman et al., 2001).

Urban streams are often associated with high concentrations of nutrients, sediment, road salts, and metals (Walsh et al., 2005; Bazinet et al., 2010). Nutrients have been found to rival or even exceed concentrations found in areas effected by agricultural land-use (Busse et al., 2006; Mallin et al., 2009). Nutrient loading within urban areas are often associated with inputs from residential areas using fertilizers on lawns and gardens (Mallin et al., 2009). In addition, increased concentrations of sediment through runoff can be problematic for streams by creating turbid conditions, while also increasing particulate concentrations. One of the most common problems with urbanization is the application of road salt, which is used as a deicer in winter months. Although road salt is only applied in the winter, high concentrations of chloride have been observed all year round (Casey et al., 2013; Van Meter & Swan, 2014; Wallace & Biastoch, 2016). Chloride ions in freshwater streams can be lethal towards aquatic life and alter the stratification of the stream (Winter et al., 2011; Wallace et al., 2013).

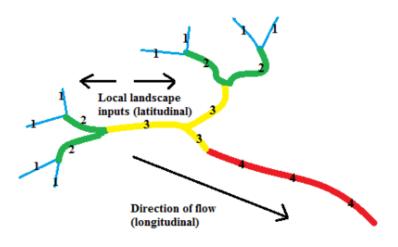
# 1.2 Spatial and Temporal Influences on Water Quality Patterns in Lotic Ecosystems

Over time, humans have become an increasingly urban species, and as the population increases, there will be a growing pressure for urban development. The process of urbanization has led to the fragmentation of natural landscapes and created complex rural-urban gradients.

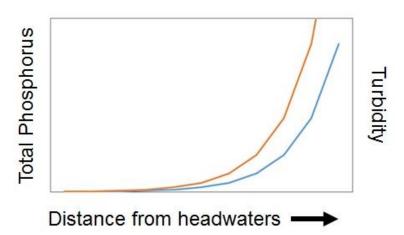
Rural-urban gradients vary among locations and landscapes, however, the overwhelming problem with these gradients is the possibility of intensifying combined runoff from both agricultural and urban point sources. Many studies have researched agricultural and urban influences on water quality and aquatic life, however, these studies are generally focused on one specific land-use or biota. In addition, very few studies have researched rural-urban gradients and their effects on water quality and aquatic organisms (Winter & Duthie, 1998; Urban et al., 2006; Mallin et al., 2009; Mei et al., 2014; Van Nuland & Whitlow, 2014). O'Brien & Wehr (2010) found that land-use effects did not fit strict land-use categories, but instead varied from rural-urban settings. This suggests observing these gradients may be the most effective method in examining land-use effects on stream function and aquatic organisms.

Another aspect of complexity in our understanding of land-use impacts to water quality and algal communities is the relative contributions of longitudinal (i.e. cumulative) water quality patterns and lateral (i.e. local landscape inputs) contributions to water quality. The river continuum concept (Vanotte et al., 1980) was the first cohesive model to explain longitudinal patterns in organic matter and by extension, water quality, in lotic systems. This conceptual model explains how water quality changes along a river's continuum, from first-order headwater streams that feed into higher order streams following a Strahler stream-order network array (Figure 1.1A). As stream order increases, the main-stem river increases in organic material as well as materials drained from the landscape via the stream-order network. Irrespective of human impacts (e.g. land-use or point-source inputs), many water quality parameters, especially those in particulate form, tend to increase along the river continuum as a function of stream order or distance from headwaters. For example, total phosphorus and turbidity can cumulatively increase as a function of distance from headwaters (Figure 1.1B). This is because the catchment area

drained is a function of distance from headwaters, and thus reflects an increase in the amount of material being drained from the landscape into the river. This is known as a longitudinal pattern in water quality that is predictable based on stream order or distance from headwaters. More recent river network theories such as the riverine ecosystem synthesis (Thorpe et al., 2006) and river wave concept (Humphries et al., 2014) have expanded on the river continuum concept to include the role of several additional spatial factors including lateral drivers of riverine processes and water quality (Figure 1.1A). The acknowledgement that lateral or local landscape inputs along a river's continuum may be an important driver of water quality reflects a significant advancement in the field of lotic ecosystem research. Yet, there remains a paucity of studies trying to distinguish the relative contributions of longitudinal and locally influenced patterns in water quality, and how variations in land-use can influence the relative importance of each spatial factor.



B)



**Figure 1.1** A) Diagram of a river network depicting a Strahler stream-order topology with a longitudinal flow path of stream orders 1 through 3 flowing into stream order 4. Modified source: Kilom691 (Own work) [CC BY-SA 3.0 (http://creativecommons.org/licenses/by-sa/3.0)] via Wikimedia Commons. B) Conceptual model of the cumulative increase in water quality parameters that occur along a river's continuum, using phosphorus and turbidity as example parameters.

### 1.3 Benthic Algae as Bioindicators

The periphyton matrix is typically dominated by benthic algae. Benthic or periphytic algae grows attached to submerged substrata found within streams, and holds a pivotal position on the bottom of the aquatic food web (Biggs, 1995; Duong et al., 2007). There is great potential for benthic algal taxa and communities to be used as bioindicators of ecosystem health and impacts due to several factors including their: position in the food web, rapid community turnover time, diverse community structure, and their ability to rapidly respond to changes in water quality (Potapova & Charles, 2003, 2007; Walker & Pan, 2006; Porter-goff et al., 2010). As such, benthic algae have been used in many studies to determine the overall health of aquatic ecosystems (Winter & Duthie, 1998, 2000; Lavoie et al., 2004; Walker & Pan, 2006).

The use of benthic algae as bioindicators make them ideal organisms for biomonitoring. Biomonitoring is a technique that uses a single organism or an assemblage of organisms to measure an ecosystems exposure to various environmental impacts (Ladislas et al., 2012). Monitoring benthic algae over time is advantageous since it is possible to infer whether a system is consistently being disturbed. Through monitoring both benthic algae and water quality in tandem, one can fully examine and assess if a system is being disturbed more effectively rather than using just one method.

Benthic algal communities are harvested through various methods within stream ecosystems. The type of substrata used in a study is dependent on the goals of the study itself and include, natural substrates or artificial substrates. The advantage of artificial substrates (e.g. unglazed ceramic tiles) is that algal biomass and community composition can be determined over a known period of time, and a community growth rate can also be determined. The artificial substrate community in some instances may represent only colonizing or early-successional taxa,

but overall reflects viable biomass that directly relates to the cumulative conditions in a local environment. This is in contrast to natural substrates that support biofilm of unknown age, sloughing/scouring events and permanence at a particular location. Unglazed ceramic tiles have been used in many aquatic studies, where several authors have found algal communities to be highly comparable to natural substrate communities (Lavoie et al., 2004; Porter-goff et al., 2010; Smucker & Vis, 2013).

### 1.4 Study Area

Durham Region is a municipality east of Toronto, Ontario, Canada representing a mosaic of longitudinal (i.e. north to south) and latitudinal (i.e. west to east) gradients in agriculture and developed land-use type and intensity. The region encompasses an area of approximately 2,600 km² and is home to an estimated 650,000 people (Durham Region, 2016). In addition, the entire region is characterized by various intensities of land-use types, including forests, agricultural and urban landscapes, which are contained in a total of 34 watersheds (Central Lake Ontario Conservation Authority, 2013b). Most of the watersheds in Durham region share headwaters in the Oak Ridges Moraine (ORM) and drain into the north shore of Lake Ontario. The ORM is a significant ecological landmark in Ontario that spans from the Niagara escarpment to Rice Lake (Allen et al., 1996). Additionally, another important feature found in Durham region watersheds is that the shoreline of Lake Iroquois cuts through the middle (Central Lake Ontario Conservation Authority, 2013b). This natural landmark is a shoreline deposit comprised of sand and gravel, and is generally referred to as Lake Iroquois beach (Central lake Ontario Conservation Authority, 2013a).

The four watersheds used in this study are the largest watersheds in Durham region and include, Lynde, Oshawa, Bowmanville and Soper Creek watersheds. All of these watersheds

contain similar geological and climatic characteristics, and all experience varying gradients of rural-urban land-use intensities (Central Lake Ontario Conservation Authority, 2012; 2013a; 2013b). Due to increased urban sprawl moving eastward from Toronto, focus has shifted to the ORM and Durham region watersheds, because of concern over the impacts rapid urbanization will have on these regions.

# **1.4.1** *Lynde Creek Watershed*

The Lynde Creek watershed drains an area of approximately 130 km², which travels through five municipalities in Durham Region, including the City of Pickering, the Town of Ajax and the Town of Whitby (Central Lake Ontario Conservation Authority, 2012). Among the four watersheds in this study, Lynde Creek is the largest and is subject to a large majority of development pressures coming from Toronto. Although it faces development pressures, Lynde Creek is adjacent to a Green Belt strip in between Whitby and Ajax, Ontario, which natural and farm lands from urban development (Ministry of Municipal Affairs, 2016). The area closest to the ORM is predominately farmland, however, as it traverses south it becomes increasingly urban, moving through the Village of Brooklin and the City of Whitby (Figure 1.2). Surface water quality within Lynde Creek has been found to be below provincial water quality guidelines, however, increasing trends in chloride, phosphorus and nitrates have been observed (Central Lake Ontario Conservation Authority, 2012).

#### **1.4.2** Oshawa Creek Watershed

Oshawa Creek Watershed is the second largest watershed in Durham region, encompassing an area of approximately 120 km² (Central lake Ontario Conservation Authority, 2013b). The majority of Oshawa Creek is primarily in the City of Oshawa, however, parts of the watershed extend into the Cities of Clarington and Whitby (Figure 1.3). Similar to Lynde Creek, Oshawa Creek has a large area of rural landscapes immediately south of the ORM, and farther south, a significant amount of pre-existing urban land-use due to long-time industrial activities driven by the General Motors plant. Water quality impairment has been evident in the Oshawa Creek watershed due to storm-sewers in urban areas and nutrient enrichment from agricultural practices (Central lake Ontario Conservation Authority, 2013b). Concentrations of chloride and phosphorus have been found to show increasing trends within specific monitoring stations, specifically in the high urban areas (Central lake Ontario Conservation Authority, 2013b).

Although some areas show water quality impairment, Oshawa Creek is home to many benthic algae, macroinvertebrates, and fish species (Central lake Ontario Conservation Authority, 2013b).

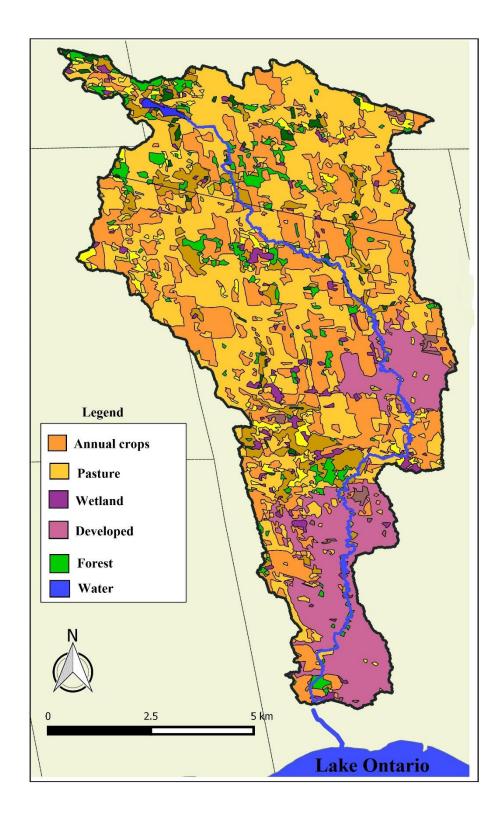
#### **1.4.3** Bowmanville Creek Watershed

The Bowmanville Creek watershed encompasses a total area of approximately 90 km<sup>2</sup> and its entirety resides in the Municipality of Clarington (Central lake Ontario Conservation Authority, 2013a). Similar to both Lynde and Oshawa Creek, this watershed traverse through diverse landscapes, northern areas are predominately agricultural and forested, and the southern areas more urban (Figure 1.4). Previous surface water quality monitoring for many Bowmanville monitoring sites have found increased concentrations for phosphorus, exceeding the 30 ug/L

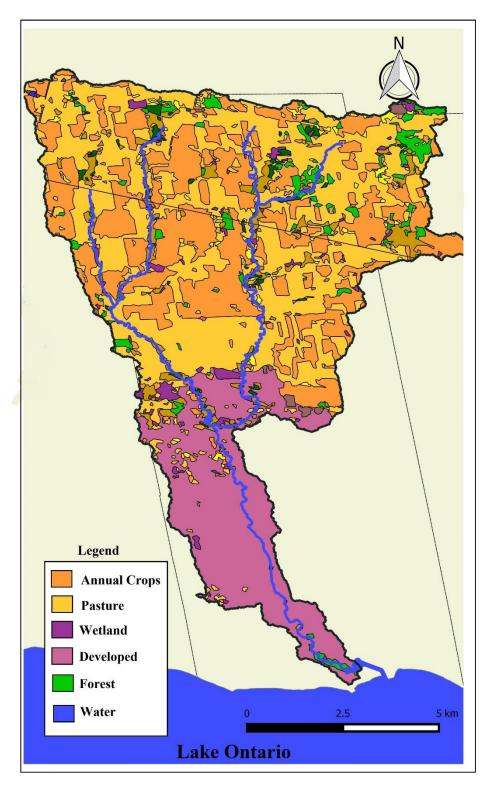
provincial guideline (Central Lake Ontario Conservation Authority, 2013a). Currently, other contaminants such as chloride and nitrogen have not been considered an issue in this watershed. The Bowmanville watershed is home to various terrestrial and aquatic organisms. For example, brook trout are the only remaining trout species within the watershed, and currently are being protected as they are easily affected by changes in land-use (Central lake Ontario Conservation Authority, 2013a). Although this watershed is functioning well now, it is projected to see rapid development within the next decade, which could change the dynamics of the watershed dramatically.

# 1.4.4 Soper Creek Watershed

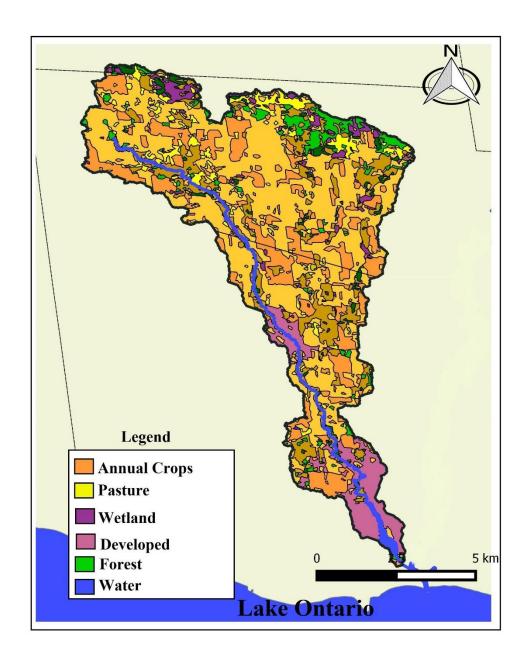
The Soper Creek watershed is the smallest watershed in this study and encompasses a total area of 80 km<sup>2</sup>, which is found entirely in the Municipality of Clarington (Central lake Ontario Conservation Authority, 2013a). In addition, similar to the previous watersheds mentioned, the Soper Creek watershed traverses through a rural-urban land-use gradient that is predominately agriculture and forest in northern areas followed by small urban centers in the south (Figure 1.5). Through previous surface water quality monitoring Soper Creek has been regularly observed to have high concentrations of both phosphorus and nitrogen concentrations at specific monitoring sites, which exceed the provincial water quality guidelines (Central lake Ontario Conservation Authority, 2013a). Just like the previously mentioned watersheds, Soper Creek is home to various terrestrial and aquatic organisms, many of which are provincially, regionally and locally rare species (Central lake Ontario Conservation Authority, 2013a).



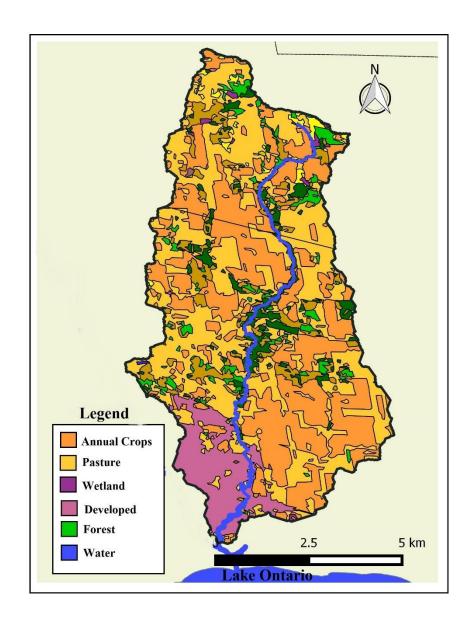
**Figure 1.2** Map showing the various types of land-use within the Lynde Creek watershed.



**Figure 1.3** Map showing the various types of land-use within the Oshawa Creek watershed.



**Figure 1.4** Map showing the various types of land-use within the Bowmanville Creek watershed.



**Figure 1.5** Map showing the various types of land-use within the Soper Creek watershed.

# 1.5 Goals and Objectives

The main goal of this research was to examine the impact of longitudinal and lateral agricultural-urban land-use gradients in four major watersheds in Durham region by monitoring standard water quality parameters and periphyton community composition. In order to achieve my main research goal, I created a set of study objectives to:

- 1) Characterize the surrounding land-use types at each site (Chapter 2)
- 2) Determine if there are relationships between surrounding land-use types, water quality and algal communities (Chapter 2).
- 3) Assess spatial and temporal patterns in nutrients, periphyton community structure and water quality in Durham region tributaries (chapter 3)
- 4) Determine if cumulative spatial patterns (i.e., distance from headwaters) or local inputs (i.e., la % Developed land-use) influence water quality (Chapter 3)

These objectives are explored and presented in chapters 2 and 3 of this thesis, respectively. In chapter 2, I focus on assessing how rural-urban land-use gradients affect water quality and periphyton communities in my study area. I hypothesized that specific water quality variables (e.g., phosphorus, nitrogen, and chloride) and certain algal taxa (i.e., pollution tolerant vs. sensitive taxa) would change as land-use changed from agriculture to urban. I predicted that nutrients (e.g., phosphorus and nitrogen) and contaminants (e.g., chloride) would be differentiated between agricultural and urban land-use types, and that pollution tolerant taxa would tend to be more associated with urban (i.e., developed) land-use. Using a complete and balanced data-set reflecting water quality and algal community variables in May, 2015, I assessed the relationships between land-use types versus water quality and periphyton community, while also observing how various land-use types influenced both water quality and

periphyton community composition. These analyses highlighted the variable nature of spatial patterns in water quality and algal community distribution, and how both land-use types can be significant contributors of nutrients to instream water quality. However, it was also apparent that certain water quality parameters such as chloride, and pollution tolerant algal taxa were closely affiliated with increased urban land-use.

In chapter 3, I focus on spatial and temporal trends in water quality and algal community composition to evaluate longitudinal (i.e., cumulative) and lateral (i.e., local land-use) influences over monthly intervals during the peak algal growing season. In this chapter, I hypothesized that both longitudinal and lateral spatial factors would influence water quality parameters across watersheds, and thus in turn, influence periphyton community biomass and composition.

However, I expected lateral land-use effects to become more important in influencing water quality trends in study creeks when land-use intensity, particularly for urban land-use, increased. In addition, I hypothesized that water quality and periphyton community structure would vary each month regardless of land-use effects due to variable precipitation events controlling hydrological forcing in each creek. Also, algal community structure has inherent seasonality, where natural succession and disturbance events can dictate community structure over time. Indeed, my results did show a role for both longitudinal and lateral factors in influencing water quality and periphyton communities, and the important role of seasonality in altering these spatial factors.

In the final chapter of this thesis, general conclusions for both chapters are provided.

Findings from both chapters are summarized and integrated to expound on the goals and objectives of this thesis, and how they relate to our broader understanding of land-use impacts to water quality and algal community structure. In addition to reflecting on the significance of my

thesis work within the broader context of lotic ecosystem ecology, I also discuss my study limitations, and make recommendations for future research and directions.

Chapter 2: Assessing rural-urban land-use gradient effects on water quality and periphyton communities in tributaries of Durham Region, Ontario

#### 2.1 Introduction

Increased human activity has led to dramatic impacts on aquatic ecosystems through the input of nutrients and various contaminants (Lavoie et al., 2004; Walsh et al., 2005). In particular, watersheds under agricultural and urban development are characterized by both their physical and chemical alterations. Agricultural land-use practices utilize large quantities of nutrients that can contribute to the degradation of surface waters (Smith et al., 2007; Hoorman et al., 2008). Comparatively, the transition of natural landscapes to urban development results in increased impervious surfaces. Impervious cover causes run-off to directly entering tributaries instead of permeating into the soil (Gallagher et al., 2011). This run-off contains particulate materials and contaminants, which includes road salts, nutrients and metals (Winter & Duthie, 1998).

Both agriculture and urban land-use have been shown to effect water quality through the addition of nutrients, particulates and contaminants into freshwater streams (Jarvie et al., 2008; Bazinet et al., 2010; Nagy et al., 2012; Andrus et al., 2015). It was previously thought that agricultural land-use practices had higher nutrient loading into streams due to high fertilizer usage than urban land-use practices, however, studies are observing nutrient concentrations to be equal or higher in urban areas comparatively (Winter & Duthie, 2000; Klose et al., 2012). In addition, both agricultural and urban land-use practices have shown to increase particulate loads, such as total suspended solids (TSS) into streams (Nagy et al., 2012; Choi et al., 2015). High TSS concentrations can effect algal growth by limiting the amount of light into a stream,

therefore limiting primary production in streams. Another problem associated with both agricultural and urban land-use is the introduction of chloride into streams. Chloride is a common deicer used in urban areas during winter months, however, it is also found in many other anthropogenic sources such as fertilizer, and sewage effluent (Winter et al., 2011). Increases in chloride concentrations found in freshwater ecosystems can negatively affect both water quality and aquatic organisms through altering the water density and osmoregulation, respectively.

A key component in examining the health of an aquatic ecosystem is through the identification of biological communities. Benthic algae or periphyton have been used in many studies to assess the ecological condition of freshwater systems (Biggs, 1995; Pan et al., 1996, 2004; Lavoie et al., 2011; Andrus et al., 2015). Benthic algae, specifically benthic diatoms, are excellent indicators of environmental change as they are influenced by a suite of factors such as geological, hydrological, and physicochemical factors (Munn et al., 2002; Potapova & Charles, 2007; Smucker et al., 2013; Yang et al., 2015). Using algal communities provides an additional secondary measure of water quality and land-use effects on a system making them ideal for biological monitoring.

Many studies have documented the effects of agricultural (Munn et al., 2002; Lavoie et al., 2004; Andrus et al., 2015) and urban land-use on water quality and periphyton, separately (Newall & Walsh, 2005; O'Brien & Wehr, 2010). However, little is known about the effects of varying levels of rural-urban gradients on water quality and periphyton communities in tributaries from the same physiographic and climatic region. In this study, sixteen sites from four tributaries in four watersheds were selected based on their comparable distances from headwaters. All study sites reflect a range in the proportion and intensity of agricultural or urban

land-use in their respective watersheds. I then explored the effects that varying longitudinal land-use gradients have on water quality parameters and periphyton community structure (i.e., biomass and taxonomic composition). Thus, the main objectives of this study were to: 1) examine and compare how land-use factors, such as developed and agricultural land-use may influence water quality and periphyton communities in tributaries located in Durham Region, Canada, and 2) identify algal bio-indicator taxa and/or assemblages that reflect water quality and/or land-use impacts along rural-urban land-use gradients.

#### 2.2 Materials and Methods

### **2.2.1** Watershed Descriptions and Site Selection

Lynde, Oshawa, Bowmanville, and Soper Creek are watersheds located in the municipality of Durham Region, Ontario, Canada. These watersheds have areas of approximately 130, 120, 90 and 80 km², respectively, draining from the Oak Ridges Moraine (ORM) and emptying into Lake Ontario. Four sites approximating similar distances from headwaters were selected along each tributary for a total of sixteen sites (Figure 2.1). Site 1 in each tributary are sites closest to the headwaters of their respective tributaries, but are not considered reference or pristine sites. These sites have no urban land-use impacts, but are impacted by agricultural activities to varying degrees (Figure 2.2). Specific site locations and information can be found in Table 2.1. All sites had variations in land-use, including developed, forest and agricultural land, reflecting a wide gradient of total land-use types and intensity (Figure 2.2, Table A1). Furthermore, all of these watersheds share similar climatic, physiographic, and geomorphological features to decrease the role of confounding factors when comparing the influence of land-use on in-stream water quality and algal community structure.

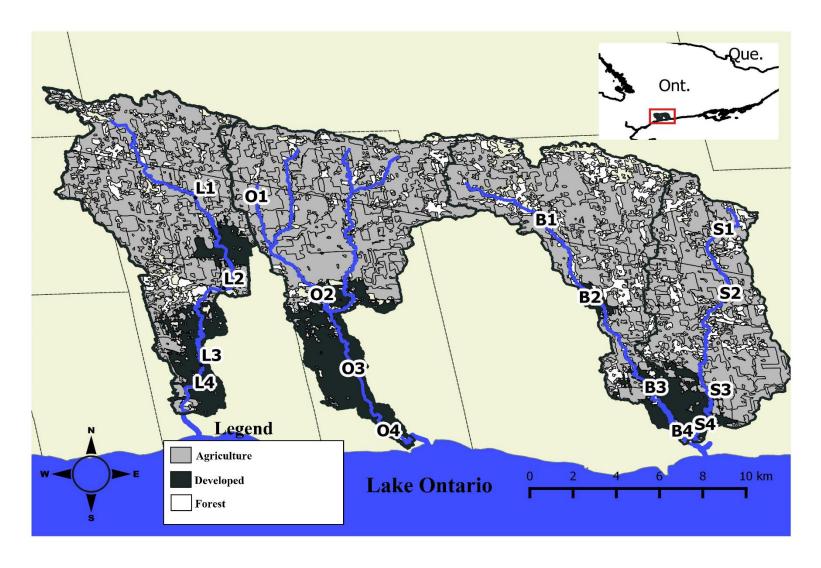
Land-use estimations were extracted for all sites using the geographical information software, QGIS (QGIS Development Team, 2015) and land-use shapefiles (Government of Canada, 2011). In addition, QGIS was used to delineate watershed boundaries and identify site locations. Using this information, 1 km-radius buffers were created for each site to extract land-use area, which was converted into percent land-use for major land-use types. A catchment for each site was delineated using the Ontario Flow Assessment Tool (Province of Ontario, 2016). In addition, 1 km reaches were made using the QGIS buffer tool for each site catchment. During preliminary analyses, the 1 km-radius buffer and site catchment were determined to be the most suitable standard-area size to demarcate the drainage areas of influence around each site. Lastly, road density was used as a secondary measure of urbanization. According to Wallace et al. (2013), road density is a comparable model to impervious surfaces for indicating urbanization, while also being easier to obtain the data and calculate. Road density was extracted through QGIS by calculating the road length within a buffer and catchment, and dividing by the 1 km-radius buffer area and catchment area.

#### **2.2.2** *Field measurements and water sampling*

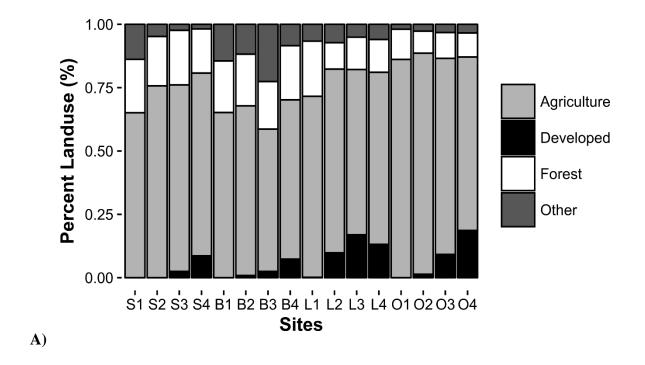
Stream water was collected at each site upon deployment and retrieval of the artificial substrates. All sites were visited at the same time of day in order to keep deployment and retrieval times consistent. On-site measurements were collected using the YSI multi parameter sonde (YSI Inc., Yellowsprings, Ohio, USA) to obtain water quality measurements for: conductivity, Total Dissolved Solids (TDS), pH, Dissolved Oxygen (DO), and temperature. Flow data was collected using a Swoffer Flow meter (Swoffer Instruments Inc., Seattle, Washington, USA). Water samples taken during deployment and retrieval dates were collected in triplicate and

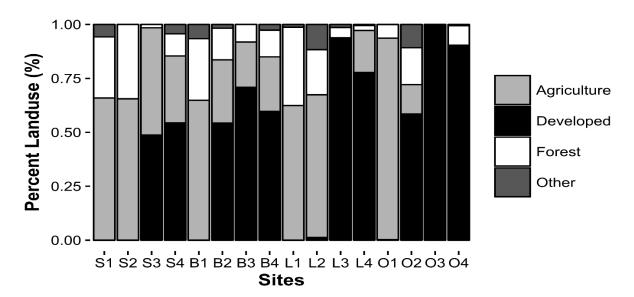
placed in 1-L Nalgene® bottles that were previously acid-washed in a 10% HCl acid-bath.

Collected samples were kept on ice in the field and immediately processed the same day



**Figure 2.1** Map showing the distribution of sampling sites and three major land-use types within each watershed.





B)

**Figure 2.2** Bar plot illustrating percent land-use found among all study sites for (A) site catchment and (B) buffers. Agricultural land includes, annual crops, pasture and cultivated crops. Developed land includes exposed land and low to high development intensities. Forests includes different intensities of coniferous, broadleaf and mixed wood trees. Other land includes, water, barren, and wetland land-use types. Land-use percentages can be viewed in Appendix A.

Table 2.1 Site locations and physical features.

Watershed	Site	Latitude/Longitude	Distance from	Elevation	Road Density,	Road Density,
	ID	Coordinates (decimal	Headwaters	( <b>m</b> )	Catchment	Buffer (Km/Km <sup>2</sup> )
		degrees)	(km)		$(Km/Km^2)$	
Lynde Cr.	L1	43.985503,-78.981764	7.54	198	1.46	1.98
Lynde Cr.	L2	43.93498, -78.955301	14.2	147	2.56	2.48
Lynde Cr.	L3	43.887189, -78.959114	20.3	90.8	3.14	8.69
Lynde Cr.	L4	43.875972, -78.961067	21.6	82.7	2.65	7.31
Oshawa Cr.	O1	43.992936, -78.94745	0.910	208	2.32	11.5
Oshawa Cr.	O2	43.942094, -78.899324	8.90	143	3.00	8.63
Oshawa Cr.	O3	43.90175,-78.873011	14.3	106	1.07	1.43
Oshawa Cr.	O4	43.868824,-78.846106	19.0	90.8	1.29	3.33
Bowmanville Cr.	B1	44.010875,-78.781317	5.70	225	1.59	7.41
Bowmanville Cr.	B2	43.969856, -78.747903	11.6	165	2.05	8.65
Bowmanville Cr.	B3	43.923733, -78.700494	18.5	122	1.00	1.81
Bowmanville Cr.	B4	43.900826, -78.680411	21.6	74.8	1.34	1.26
Soper Cr.	<b>S</b> 1	44.017942, -78.684961	3.04	221	1.72	5.75
Soper Cr.	S2	43.9808, -78.673958	7.70	150	2.30	7.25
Soper Cr.	<b>S</b> 3	43.922586, -78.668214	14.6	111	1.37	0.72
Soper Cr.	<b>S</b> 4	43.900453, -78.673044	17.4	79.9	1.79	3.64

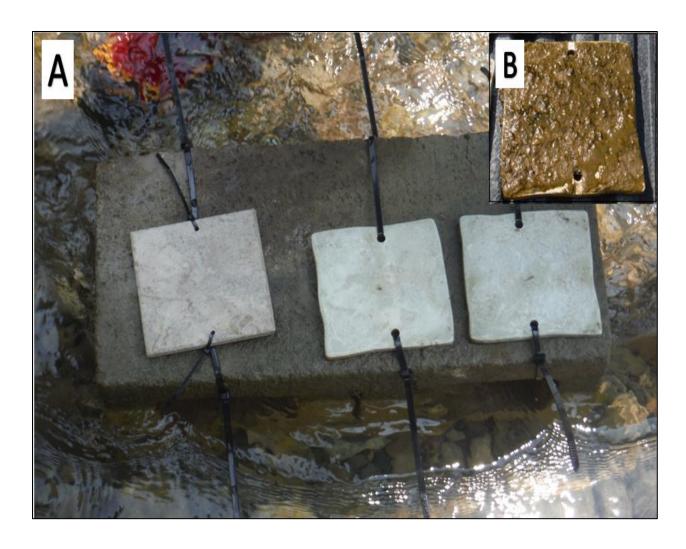
### **2.2.3** *Water Chemistry*

Collected water samples were processed for Total Nitrogen (TN), Total Phosphorus (TP), chloride, and Total Suspended Solids (TSS). TN and TP replicate samples were unfiltered and pooled into 50 mL falcon™ tubes and immediately frozen until ready for analysis. Aliquots of TN samples were shipped to SGS Environmental Services (SGS Canada Inc., Lakefield, Ontario, Canada) to be analyzed. TP samples were digested using a modified EPA 200.8 method (Creed et al., 1994) and shipped to the Water Quality Center (Peterborough, Ontario, Canada) for analyses. Remaining water samples were filtered through pre-rinsed glass fiber filters (VWR® glass fiber filters 4.7cm, 696). Filtrate was used to measure chloride concentrations using a VWR Symphony® ISE Chloride meter (VWR International, Radnor, PA, USA). TSS was determined using standard methods (American Public Health Association, 1992).

# **2.2.4** Use of Artificial Substrates to Measure In-Situ Algal Colonization and Growth

Unglazed ceramic tiles (10 X 10 cm) (Figure 2.3A) were used as artificial algal growth substrates in this study. Substrate deployments occurred approximately monthly between May and August, 2015, including four consecutive deployments of artificial substrates to measure periphyton colonization and growth during 3-week incubation periods. An incubation period reflected algal growth from time 0 to 21 days, which is the minimum time needed for an established algal community to grow on artificial substrates (See Porter-goff et al., 2010). Preliminary experiments with artificial substrates in Oshawa Creek (Comeau and Kirkwood, in prep.) determined that 3 weeks was an ideal deployment period that captured mid-growth phase biofilm biomass prior to self-sloughing.

Three replicate unglazed ceramic tiles were fixed to concrete blocks using nylon cable ties (Figure 2.3). Cement blocks were placed in the stream bed of each site for a full 21-day incubation period and retrieved on the 21<sup>st</sup> day. Upon termination of each incubation period, ceramic tiles were removed from the cement blocks and samples were collected in triplicate for Chlorophyll a (Chl *a*), Ash Free Dry Mass (AFDM) and taxonomic analysis. Periphyton communities were scraped off of each ceramic tile using the inner-circumference of a PVC pipe as (13.25 cm²) to demarcate the sampling area. A steel pick and tooth brush were used to clear the defined area on each tile until it was visibly devoid of all periphyton biomass. Scraped periphyton biomass was rinsed in to replicate specimen cups, and the slurry volume was recorded. Samples earmarked for Chl *a* and AFDM analyses were immediately processed on the same day in the lab. Samples earmarked for taxonomic analysis were pooled for each site and placed into 4-oz Qorpak® glass bottles. Lugol's Iodine solution was added to each bottle to fix and stain the periphyton sample until further analyses.



**Figure 2.3** Field picture of unglazed ceramic tiles affixed to a cement block before deployment (A) and after a typical incubation period (B).

### **2.2.5** *Periphyton Sample Processing*

Samples for Chl *a* and AFDM were filtered using pre-rinsed glass fiber filters (VWR® 4.7cm glass fiber filters, 696). Chl *a* samples were first homogenized in 90% acetone and cold-extracted in the dark for a minimum of 4-h. Following the protocol of Kirkwood et al. (1999), acetone-extracted samples were measured for Total and Corrected Chl *a* absorbance using a UV-spectrophotometer (Thermo Fisher Scientific, Waltham, MA, USA). AFDM samples were dried following filtration at 60°C for at least 24 hours, pre-weighed before combustion at 550°C for 2-h and then re-weighed (see Bourassa and Cattaneo 1998).

Samples for taxonomic analysis were transferred and settled to a PhycoTech nanoplankton chamber (PhycoTech, Inc., Michigan, USA). Algal counts and identification were done using an Evos XL Core inverted microscope at 400X. A minimum of  $\geq$  200 cells were enumerated for each sample (Duthie & Winter, 1998). All diatoms were identified to genus-level and when possible, species-level. Primary references for diatom identification were based on Biggs and Kilroy (2000) and Cox (1996).

### **2.2.6** *Statistical Analyses*

Although artificial substrate deployments were performed monthly from May – August, 2015, only the May incubation period resulted in a complete retrieval of all deployed tiles. Subsequent incubation periods resulted in the loss or damage of some tiles at some sites, giving rise to missing periphyton data for some sites on some dates. Therefore, this chapter is focusing on the complete and balanced data-set containing algal community data for all sites from the same incubation period (see Table 3.1 for details on missing data). Sets of Analysis of Variance (ANOVA) were run to assess differences in periphytic Chl *a* and AFDM between sites for each

tributary. Sites were treated as independent sampling locations in each creek due to known spatial heterogeneity of algal communities in lotic ecosystems (Dutilleul, 1993; Downing, 2001; Schank & Koehnle, 2009). However, any occurrence of spatial co-linearity of water quality parameters within each tributary was accounted for in across-tributary comparisons. All data were tested for deviations from normality and homogeneity of variance, and if necessary, transformations were made to meet parametric assumptions. Single and multiple linear regressions were used to assess relationships between periphyton biomass (Chl *a* and AFDM), land-use, and water quality. Select variables were log-transformed to improve linear relationships.

Multivariate statistical analyses for this study was performed using R (version 3.2.2, R Development Core Team, Vienna Austria) and the package *vegan*. Detrended Correspondence Analysis (DCA) were used in determining whether Redundancy Analysis (RDA) or Canonical Correspondence Analysis was appropriate for the dataset. Based on the gradient lengths obtained (gradient lengths < 3), it was determined that a linear model was most appropriate and therefore RDA was used.

RDA was used to determine the relative importance of water quality parameters and landuse in explaining variation in algal community composition. A separate RDA was performed to
examine land-use and water quality parameters for clearer interpretation. In addition, principal
component analysis (PCA) was performed to observe water quality gradients across study sites.

All water quality and land-use data were centered and standardized. Algal community data was
square root transformed to reduce the influences of both abundant species and rare species
(Lavoie et al., 2004). Species with less than 1% relative abundance were removed from analysis.

Water quality parameters were assessed for collinearity, and variables with variation inflation

factors (VIF) greater than 20 were removed (Gross, 2003). The variables used in the final analyses were selected for their ecological importance in this study, and reflect aspects of water quality (e.g., TN and TP), land-use (e.g., TSS and chloride) and natural geology (e.g., pH). Monte Carlo permutation tests were used to test the statistical significance of the RDA axes (999 random permutations, p<0.05).

#### 2.3 Results

# **2.3.1** *Water Quality*

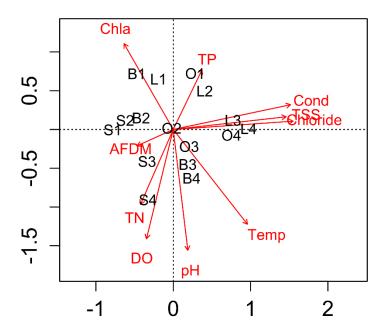
Several water quality parameters showed distinct differences and varied at each site and tributary. Mean values for May water quality parameters at each site are shown in Table 2.2. Particulate concentrations such as chloride and TSS, ranged from 10.8 mg·L<sup>-1</sup> to 163 mg·L<sup>-1</sup> and 1.25 mg·L<sup>-1</sup> to 4.58 mg·L<sup>-1</sup>, respectively. Nutrient concentrations such as TN and TP, varied from 0.500 mg·L<sup>-1</sup> to 1.55 mg·L<sup>-1</sup> and 7.39  $\mu$ g·L<sup>-1</sup> to 14.1  $\mu$ g·L<sup>-1</sup>, respectively. ANOVA results comparing sites within and across watersheds can be found in chapter 3.

The principal component analysis conducted with standardized data showed how the water quality variables were associated with all of the study sites (Figure 2.4). The first two axes of the PCA model explained a total of 57.4% of the variation in the data-set. There is a clear separation between the sites 1 and 2, and the most developed sites, 3 and 4. The headwater sites were ordinated on the top side of the first axis compared to the developed sites that were found toward the bottom side. The most developed sites, sites 3 and 4, were found to be associated with many specific water quality variables. For instance, Soper Creek sites 3 and 4 were associated with high TN and DO, Bowmanville Creek was associated with high temperature and pH, and Lynde Creek was associated with high chloride, conductivity, and TSS. Oshawa Creek sites 3 and 4 were found to be associated with different variables such as, high temperature and chloride, respectively.

**Table 2.2** Summary table of means and standard deviations (in brackets) for May water quality parameters collected at deployment and retrieval of artificial substrates. Superscript numbers indicate sample size for each water quality parameter: 1 (n=2) and 2 (n=6).

Watershed	Site	pH <sup>1</sup>	$TSS^2$	Cond <sup>1</sup>	Chloride <sup>2</sup>	$DO^1$	Temperature <sup>1</sup>	$TP^1$	$TN^1$	Flow <sup>1</sup>
	ID	_	$(mg L^{-1})$	(µs cm <sup>-1</sup> )	$(mg L^{-1})$	$(mg L^{-1})$	(°C)	$(\mu g L^{-1})$	$(mg L^{-1})$	$(m s^{-1})$
Lynde Cr.	L1	8.22 (0.14)	1.58 (0.8)	592 (40)	51.0 (8.8)	11.1 (0.6)	12.1 (2.4)	14.1 (0.3)	0.600 (0.02)	0.46 (0.3)
Lynde Cr.	L2	8.14 (0.24)	3.25 (0.6)	786 (38)	110 (0.5)	11.2 (0.9)	13.8 (1.6)	10.9 (2.4)	0.630 (0.14)	0.67(0.4)
Lynde Cr.	L3	8.23 (0.14)	3.42 (0.9)	854 (112)	163 (15)	11.5 (0.0)	15.6 (1.6)	9.91 (4.6)	0.790(0.09)	0.89 (0.06)
Lynde Cr.	L4	8.28 (0.13)	4.58 (1.1)	768 (16)	115 (5.5)	11.6 (0.3)	15.4 (1.9)	9.41 (2.6)	0.590(0.1)	1.06 (0.6)
Oshawa Cr.	O1	7.95 (0.16)	3.75 (1.1)	707 (38)	66.7 (5.4)	10.5 (1.2)	10.9 (2.2)	10.3 (2.8)	0.940(0.2)	0.72 (0.3)
Oshawa Cr.	O2	8.29 (0.07)	2.92 (0.7)	599 (42)	49.1 (2.1)	11.9 (0.02)	14.2 (1.6)	10.4 (0.5)	0.910(0.1)	0.72(0.4)
Oshawa Cr.	O3	8.31 (0.09)	3.25 (0.6)	635 (45)	62.7 (4.4)	12.0 (0.3)	14.6 (1.7)	8.20 (1.1)	1.00 (0.08)	1.88 (0.7)
Oshawa Cr.	O4	8.29 (0.08)	3.75 (0.4)	785 (66)	110 (10)	11.6 (0.2)	15.1 (2.1)	11.2 (1.2)	1.22 (0.4)	1.88 (0.5)
Bowmanville Cr.	B1	8.00 (0.04)	2.75 (1.1)	449 (21)	14.0 (4.4)	11.2 (1.1)	11.1 (4.7)	10.0 (1.6)	0.500(0.1)	1.22 (0.3)
Bowmanville Cr.	B2	8.24 (0.09)	2.67 (1.2)	441 (23)	15.8 (4.2)	11.5 (0.8)	12.4 (4.3)	9.87 (0.0)	0.630 (0.05)	1.02 (0.6)
Bowmanville Cr.	В3	8.41 (0.09)	3.00 (0.8)	444 (40)	25.7 (4.5)	11.3 (1.4)	15.6 (6.3)	8.54 (0.7)	0.680 (0.05)	1.82 (0.3)
Bowmanville Cr.	B4	8.42 (0.05)	2.33 (0.4)	468 (57)	36.0 (6.7)	11.5 (1.3)	18.1 (6.8)	9.68 (0.3)	0.660(0.1)	0.94 (0.3)
Soper Cr.	<b>S</b> 1	8.19 (0.03)	1.58 (0.9)	424 (18)	10.8 (1.0)	11.8 (0.6)	9.60 (2.3)	7.39 (0.3)	1.07 (0.04)	1.01 (0.06)
Soper Cr.	S2	8.16 (0.09)	1.25 (1.4)	484 (19)	26.5 (0.5)	11.4 (1.2)	11.8 (3.9)	9.09 (1.3)	1.21 (0.09)	1.96 (1.3)
Soper Cr.	S3	8.29 (0.08)	1.67 (1.5)	502 (32)	30.5 (2.7)	11.8 (1.1)	14.8 (5.6)	9.04 (1.3)	1.32 (0.2)	1.26 (0.17)
Soper Cr.	S4	8.38 (0.07)	2.25 (1.2)	563 (46)	48.5 (6.0)	13.2 (1.8)	15.0 (6.1)	10.4 (0.7)	1.55 (0.5)	1.83 (0.56)

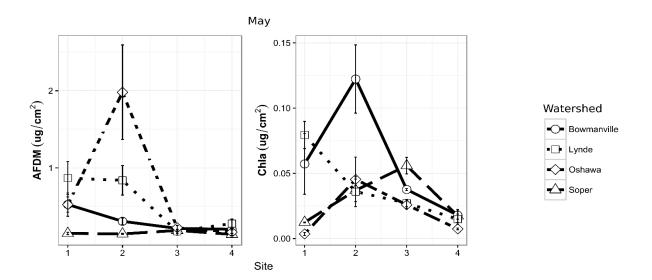
Cond Conductivity, TSS Total Suspended Solids, DO Dissolved Oxygen, TP Total Phosphorus, TN Total Nitrogen



**Figure 2.4** PCA performed on standardized mean water quality variables among all sites in May, 2015. Site and water quality variable abbreviations can be found in Table 2.2. The first two axes explain 30% and 27.40% of the variation, respectively.

# **2.3.2** Periphyton Biomass

AFDM and Chl a in particular, varied across most sites, varying from 0.12 - 1.98 mg cm<sup>-2</sup> and 0.035 - 0.12 µg cm<sup>-2</sup> Chl a, respectively (Figure 2.5). One-way ANOVA comparing Chl a among sites and watersheds showed significant differences for all watersheds (Table 2.3, P<0.05). A similar trend was observed for AFDM, which saw significant differences among all watersheds except for Soper Creek (Table 2.4, P<0.05). A Pearson correlation matrix showed only one significant relationships between algal biomass and water quality variables (Table 2.5). Among the algal biomass variables, only Chl a showed a negative correlation with TSS (r=-0.67). A scatterplot of this relationship can be found in Figure 2.6. No statistically significant relationships were detected between periphyton biomass and land-use parameters (Table 2.7 and 2.8). Multiple linear regressions using raw and log-transformed data showed no significant relationships between periphyton biomass and any water quality or land-use parameter.



**Figure 2.5** Total AFDM and Chl a concentrations in May at each sampling location (sites 1-4) of each study creek. Error bars reflect  $\pm$  standard error values.

**Table 2.3** One-way analysis of variance results for Log Chl a concentrations in May (n=3 for each study site) among watershed study sites.

Term		Treatment means	F value	P value
Watershed	Site	(Chl a)		
Lynde	Site 1	0.0794	9.32	< 0.001
•	Site 2	0.0359		
	Site 3	0.0272		
	Site 4	0.0149		
Oshawa				
	Site 1	0.00351	19.9	< 0.001
	Site 2	0.0453		
	Site 3	0.0258		
	Site 4	0.00734		
Bowmanville				
	Site 1	0.0572	4.59	0.038
	Site 2	0.122		
	Site 3	0.0378		
	Site 4	0.0149		
Soper				
_	Site 1	0.0124	13.4	0.002
	Site 2	0.0369		
	Site 3	0.0559		
	Site 4	0.0173		

**Table 2.4** One-way analysis of variance results for AFDM concentrations in May (n=3 for each study site) among watershed study sites.

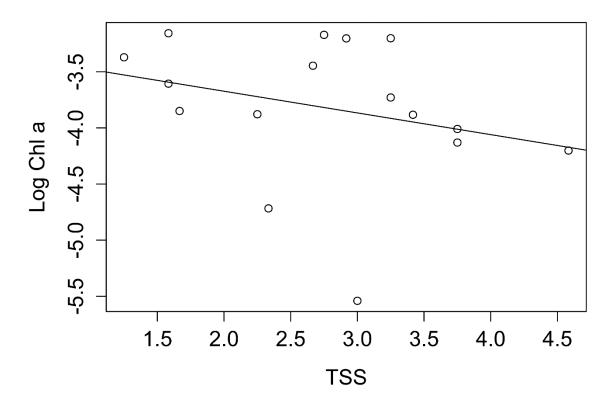
Term		Treatment means (AFDM)	F value	P value
Watershed	Site			
Lynde	Site 1	0.869	6.13	0.018
	Site 2	0.839		
	Site 3	0.177		
	Site 4	0.279		
Oshawa				
	Site 1	0.524	7.23	0.012
	Site 2	1.98		
	Site 3	0.225		
	Site 4	0.168		
Bowmanville				
	Site 1	0.526	7.35	0.011
	Site 2	0.308		
	Site 3	0.203		
	Site 4	0.204		
Soper				
_	Site 1	0.151	0.492	0.698
	Site 2	0.145		
	Site 3	0.190		
	Site 4	0.139		

**Table 2.5** Pearson correlation results for algal biomass parameters against water quality for May. Bold numbers indicate statistically significant correlation coefficients (P<0.05). Scatterplots of significant correlations are included in Appendix A.

Algal Biomass	Chloride	Cond	DO	pН	Temp	TP	TN	TSS
Log Chl a	-0.36	0.34	0.036	0.25	-0.071	0.24	-0.27	-0.67
AFDM	-0.025	-0.20	-0.066	-0.11	-0.088	0.25	-0.25	0.05

**Table 2.6** Pearson correlation results for algal biomass parameters against land-use variables from site catchment and 1 Km site buffers for May. Bold numbers indicate statistically significant correlation coefficients (P<0.05). Scatterplots of significant correlations are included in Appendix A.

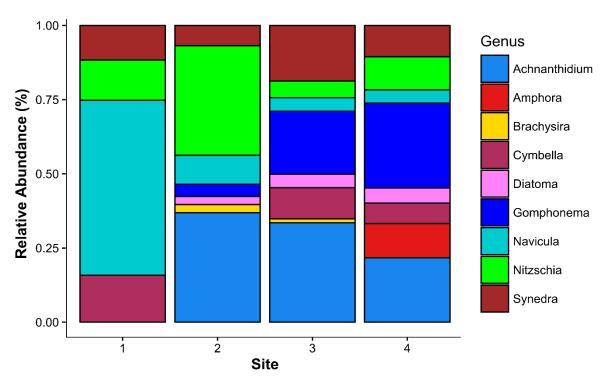
Landscape Scale	Site Catchme	ent			1-Km Site B	uffer		
Algal Biomass	Agriculture	Developed	Forest	Road Density	Agriculture	Developed	Forest	Road Density
Log Chl a	-0.45	0.31	-0.03	0.31	-0.26	-0.03	0.07	0.10
AFDM	0.23	-0.12	-0.07	-0.03	0.12	-0.16	0.23	-0.20



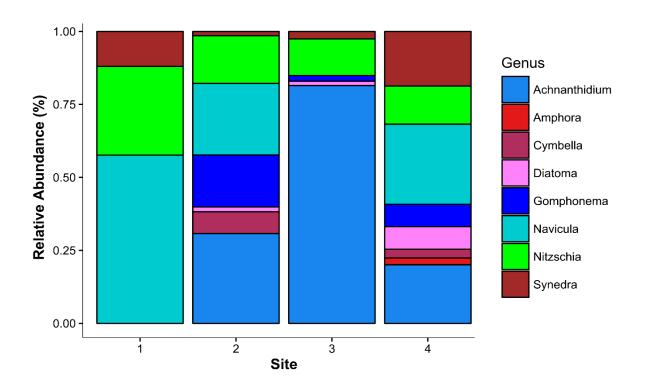
**Figure 2.6** Linear model of Chlorophyll a against TSS (r= -0.67, p=0.043).

### **2.3.3** *Periphyton Community Composition*

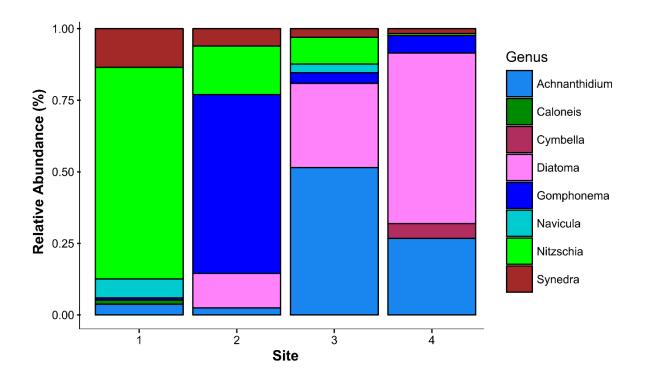
The periphyton community from artificial substrates in May was composed of 47 algal species from 24 different genera. Since rare taxa were omitted from further analyses, five genera and ten species were removed. The community assemblages were dominated by the genera Achnanthidium (29.27%), Nitzschia (21.49%), Navicula (15.68%), and Gomphonema (10.54%). Relative abundance plots for each watershed can be found in Figures 2.7-2.10. The community composition for study sites within each watershed was observed to change longitudinally down the tributary. Lynde Creek was found to shift from a community dominated by the genus Navicula in site 1 to a community dominated by many genera, such as Gomphonema, Achnanthidium, Nitzschia, and Amphora. Oshawa Creek saw a similar trend to Lynde Creek, with site 1 being dominated by Navicula followed by a shift in genera present. The most significant difference amongst sites in Oshawa Creek was the large abundance of Achnanthidium (approximately 80% abundance) found at site 3. Algal Communities within Bowmanville Creek sites were observed to have a different algal genera dominate each study site. For instance, Bowmanville Creek site 1 was dominated by the genus Nitzschia, followed by Gomphonema in site 2, Achnanthidium in site 3 and Diatoma in site 4. Lastly, Soper Creek sites were observed to shift from being largely dominated by Nitzschia to genera, Achnanthidium and Diatoma. Relative biovolume information can be viewed in supplementary information appendix A (Figure A5-A8).



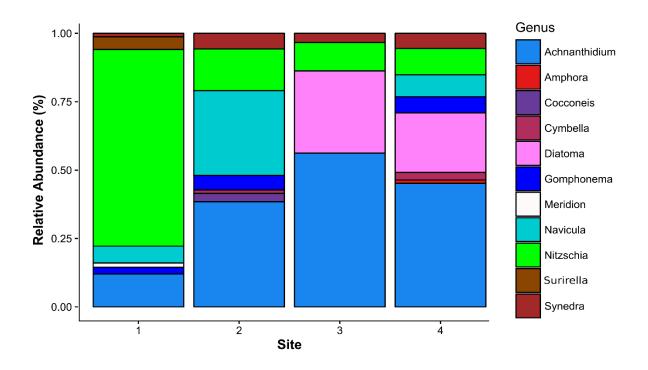
**Figure 2.7** Relative abundance based on cell density for study sites located in the Lynde Creek watershed for the month of May.



**Figure 2.8** Relative abundance based on cell density for study sites located in the Oshawa Creek watershed for the month of May.



**Figure 2.9** Relative abundance based on cell density for study sites located in the Bowmanville Creek watershed for the month of May.



**Figure 2.10** Relative abundance based on cell density for study sites located in the Soper Creek watershed for the month of May.

### **2.3.4** Redundancy Analyses of Land-Use, Water Quality and Algal Communities

Pearson correlation analyses revealed that several water quality variables were strongly correlated to specific land-use types (Table 2.7 and 2.8). Most notably, developed land-use and road density for site catchment had strong positive correlations with chloride (r=0.85 and 0.89, respectively) and temperature (r=0.78 and 0.82, respectively). Chloride and temperature were highest in the most developed sites, which were observed to be as high as 180 mg/L and 25.3 C°, respectively. The opposite was seen with forests, which showed negative correlations with chloride (r=-0.72) and TSS (r=-0.63). Surprisingly, no water quality variables were shown to have significant correlations among agricultural land-use for site catchment. Few correlations were found between water quality variables and site buffers. Similar to site catchment, road density and temperature were found to have a strong correlation (r=0.83). In addition, agricultural and forest land-use showed a negative correlation with temperature (r=-0.79), and forest land-use showed a negative correlation with temperature (r=-0.61) and TSS (r=-0.60). Scatterplots of significant correlations can be found in Appendix A (Figure A1-A4).

Redundancy analysis revealed important associations between water quality variables and land-use parameters (Figure 2.11A and 2.11B). The RDA for site catchment explained 47.6% of the total variation with the first two axis explaining 32.1% and 12.3%, respectively. (Figure 2.11A). The second RDA for site buffer explained 32.0% of the total variation with the first two axis explaining 24.6% and 6.23%, respectively. (Figure 2.11B). Monte carlo permutations tests found that the relationships between land-use and water quality variables were statistically significant (999 random permutations, p<0.05). Developed land-use was removed from both RDAs as it was found to have a high collinearity with road density. Within both RDAs, water quality variables such as TSS, chloride and temperature were correlated with road density. TN

and TP locations within the RDA indicated that no land-use types correlated with nutrient concentrations. Site locations were differentiated by land-use parameters and water quality variables. Highly developed sites clustered on the right side of axis 2. In comparison, the sites closest to the headwaters were highly associated with agricultural and forested land-use, with the exception of Oshawa Creek site 1.

The redundancy analysis performed for water quality and community composition explained 73.66% of the total variation with the first two axis accounting for 40.34% and 13.73%, respectively (Figure 2.12A). Monte carlo permutations showed that associations between genera and water quality variables were statistically significant (999 random permutations, p<0.05). Few water quality variables within the RDA were associated with specific algal genera. *Gomphonema* positively correlated with TSS and Chloride, while *Amphora* and *Cymbella* correlated well with DO and TN: TP. Temperature and pH was positively correlated with *Achnanthidium* and negatively associated with *Cocconeis* and *Navicula*. The genera, *Cocconeis* and *Navicula* showed positive correlations with TP. Many genera such as, *Melosira* and *Surirella* were ordinated near the origin indicating that they were associated with multiple different water quality variables. Based on the community composition, developed and agricultural sites were clearly separate in the RDA. Highly developed sites tended towards the right side of the second axis compared to the agricultural sites that were ordinated on the left side.

RDA performed for site catchment land-use and taxa composition explained 34.0% of the total variation (Figure 2.12B). The first two axis explained 22.2% and 7.29% of the variation, and monte carlo permutation tests showed that relationship between genera and land-use parameters were statistically significant (999 random permutations, p<0.05). Within the RDA,

many genera showed close relation to the land-use parameters. Specific genera such as, *Gomphonema*, *Cymbella*, and *Amphora* positively correlated with road density. In contrast, genera such as *Cocconeis* and *Navicula* showed a negative correlation to road density and related more to forest and agricultural land-use. Similar to the water quality RDA, highly developed sites were found on the right side of the second axis compared to the agricultural sites, which clumped together on the left side.

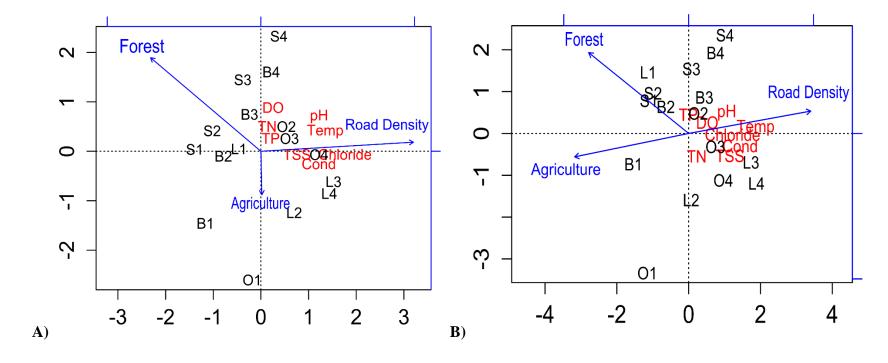
RDA performed for the site buffer land-use and taxa composition explained 38.5% of the total variation (Figure 2.12C). The first two axis explained 27.5% and 9.87% of the variation, and monte carlo permutation tests showed that relationship between genera and land-use parameters were statistically significant (999 random permutations, p<0.05). Within the RDA, many genera showed close relation to the land-use parameters. Specific genera such as, *Gomphonema*, *Cymbella*, and *Amphora* positively correlated with road density. In contrast, many genera were found in the middle of the RDA indicating that they are correlated with multiple land-use types. Again, similar to the water quality RDA, highly developed sites were found on the right side of the second axis compared to the agricultural sites, which clumped together on the left side.

**Table. 2.7** Pearson correlation coefficients between % land-use from site catchment and water quality variables in May. Bold numbers indicate statistically significant correlations (P<0.05). Scatterplots of significant correlations are included in Appendix A.

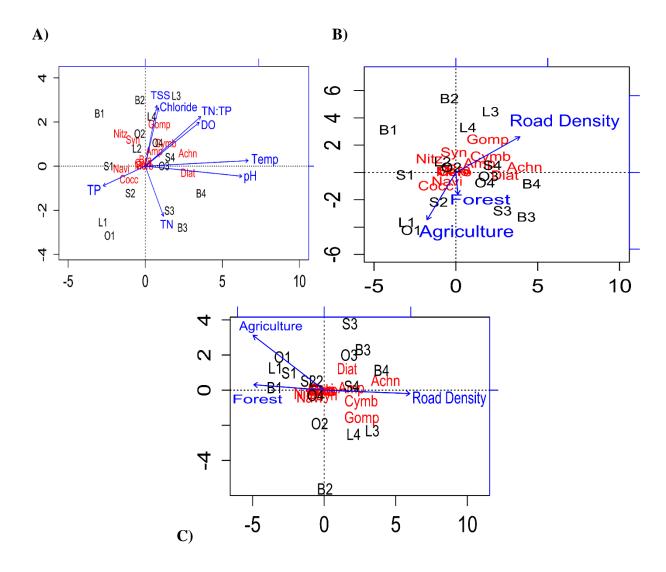
Land-use	Chloride	Cond	DO	рН	Temp	TP	TN	TSS
Agriculture	-0.02	0.17	0.11	-0.09	-0.22	0.03	0.25	0.43
Developed	0.85	0.79	-0.17	0.52	0.78	-0.01	0.08	0.33
Forest	-0.72	-0.80	0.30	-0.30	-0.33	0.08	-0.01	-0.63
Road density	0.89	0.85	-0.10	0.56	0.82	0.05	0.13	0.37

**Table. 2.8** Pearson correlation coefficients between % land-use from site 1-km buffer and water quality variables in May. Bold numbers indicate statistically significant correlations (P<0.05). Scatterplots of significant correlations are included in Appendix A.

Land-use	Chloride	Cond	DO	рН	Temp	TP	TN	TSS
Agriculture	-0.42	-0.36	0.27	-0.04	-0.79	0.19	-0.05	-0.01
Developed	0.46	0.40	-0.14	0.16	0.80	-0.23	0.07	0.21
Forest	-0.44	-0.43	-0.12	-0.43	-0.61	0.33	-0.19	-0.60
Road density	0.42	0.35	-0.02	0.23	0.83	-0.18	0.16	0.17



**Figure 2.11** RDA showing relationships between land-use (A= site catchment, B=site 1-km buffer), water quality variables and sampling sites for the month of May. Site and water quality variable abbreviations can be found in Table 2.2. Monte carlo permutation tests found this RDA to be statistically significant (999 random permutations, p<0.05).



**Figure 2.12** RDA showing the relationships between either (A) water quality variables or (B) site catchment land-use with algal taxa or (C) site buffer land-use with algal taxa and sampling sites in May. Taxa codes are represented in Table 2.7. Monte carlo permutation tests found both of these RDA to be statistically significant (999 random permutations, P<0.05).

 Table 2.9 Identified taxa from all streams, arranged in alphabetical order.

Genus	Taxa code	No. of Species
Achnanthidium	Achn	2
Amphora	Amp	2
Brachysira	Bra	1
Caloneis	Calo	3
Cocconeis	Cocc	2
Cryptomonas	Cryp	1
Cyclotella	Cycl	1
Cymbella	Cymb	2
Diatoma	Diat	2
Frustulia	Frus	2
Gomphonema	Gomp	2
Melosira	Melo	1
Navicula	Navi	4
Nitzschia	Nitz	5
Rhoicospenia	Rho	1
Sceneodesmus	Scen	1
Selenastrum	Sele	1
Surirella	Suri	2
Synedra	Syn	2

#### 2.4 Discussion

# **2.4.1** Water Quality and Periphyton Biomass Trends Within and Across Watersheds

Water quality variables in the month of May varied across the study sites and watersheds, however, increases in pH, conductivity, chloride and temperature were observed when travelling from headwater sites (i.e., Site 1) to developed sites (i.e., Site 4) (Table 2.2). Based on provincial water quality guidelines, when comparing headwater sites to developed downstream sites, headwater sites appear to be less impacted even though they are dominated by agricultural landuse. Among all of the water quality parameters, only chloride was observed to exceed the water quality guidelines within both Lynde Creek site 3 and 4. Agricultural land-use practises, including annual crops and pastures have been observed to increase water quality variables such as, TSS, and conductivity, yet this was something I did not observe in this study (Munn et al. 2002; Walsh & Wepener, 2009). Therefore, in this region, it seems that urbanization is the major driver of environmental change on water quality and algal communities compared to agricultural land-use.

Within the Oshawa Creek sites, Oshawa Creek site 1 was an outlier among the other sites, this is further seen in the land-use RDA (Figure 2.10). Oshawa Creek site 1 is near a major road, King's Highway 12, and is close to a large portion of agricultural land-use. When comparing Oshawa Creek site 1 and site 2, site 1 was observed to have higher chloride and TSS concentrations then site 2. The higher concentrations of chloride and TSS in site 1 could be due to the close proximity of a major roadway, however, this does not account for why site 2 is lower when it has more developed land cover. Water quality downstream from Oshawa Creek site 1 seems to improve even though urbanization is increasing. This improvement in water quality is most likely due to groundwater recharge from the Algonquin shoreline, which could be having a

dilution effect on the water found at Oshawa Creek site 2 (Central lake Ontario Conservation Authority, 2013b).

Chl *a* and AFDM varied among the sites for each watershed, with Bowmanville Creek site 2 showing the highest mean Chl *a* and Oshawa Creek site 2 showing the highest mean AFDM for the month of May (Figure 2.5). There are many reasons why the algal biomass varied from site to site among each watershed. Algal growth is highly influenced by a combination of factors such as, elevated light, nutrients, temperature, and flow regime (Biggs, 1995; Godwin & Carrick, 2008; Braccia et al., 2013). As mentioned previously, water quality variables varied among the sites, including nutrients and temperature, which were observed to vary among the sites for each watershed. Although it was not measured, light intensity would have varied among the sites as well, since urbanization has been found to decreases canopy cover.

Chl *a* and AFDM concentrations are both measurements of periphyton biomass, however, their concentrations did not show a strong correlation among each other. This dissimilarity is due to the differences in what Chl *a* and AFDM measure. Chl *a* is a proxy measure for algal biomass, and it measures only Chl *a* concentrations (e.g., autotrophic organisms) (Biggs & Kilroy, 2000). In comparison, AFDM is a measure of all organic biomass found in a sample, this includes, living autotrophic (e.g., algae) and heterotrophic organisms (e.g., macroinvertebrates), dead material, and other organic material (Biggs & Kilroy, 2000). Therefore, in relation to algal biomass, Chl *a* is the better proxy of algal growth than AFDM.

# **2.4.2** *Land-use effects on water quality*

Changes in land-use (i.e., agricultural and urban development) can strongly affect the water quality of streams by altering the physical and chemical conditions of stream ecosystems (Pan et al., 2004; Walsh et al., 2005; Smucker et al., 2013). Water quality varied from site to site

among all tributaries studied, however, some trends were seen in relation to land-use. Chloride, temperature and conductivity all showed positive correlations with road density, and were associated with the most developed sites (i.e., site 3 and 4). These results are consistent with studies conducted by O'Brien and Wehr (2010) and Porter-Goff et al. (2013), in which similar trends were seen in relation to urban development. High chloride concentrations can negatively affect water quality by increasing the amount of dissolved ions and elevating stream conductivity (Paul & Meyer, 2001; Casey et al., 2013). Many studies report conductivity as an important indicator of urban development (Winter & Duthie, 1998; Munn et al., 2002; Walker & Pan, 2006), and in our study conductivity did show significant relationships with road density. However, this study found that chloride was a better indicator of urbanization compared to conductivity. Recent studies by Bazinet et al. (2010), Wallace et al. (2013), and Wallace and Biastoch (2016) found similar findings with chloride and urban development.

Nutrient concentrations varied along sites in all the tributaries for TN and TP. No significant correlations were found for nutrients and individual land-use parameters within the Pearson correlation or the redundancy analysis. This could indicate that nutrient concentrations are being influenced by multiple land-use parameters. Previous research has shown that both developed and agricultural land-use parameters have related to increased phosphorus and nitrogen in stream ecosystems (Jarvie et al., 2008; Klose et al., 2012), which contrasts the results of this study. Furthermore, the results of this study contrasts the concept that nutrient loadings are higher in urban streams compared to agricultural streams (Busse et al., 2006; Mallin et al., 2009; O'Brien & Wehr, 2010). Among all of the tributaries in this study only Soper creek was observed to show increases in both TP and TN along a rural- urban development gradient (Table 2.2). This result is not surprising since the Central Lake Ontario Conservation Authority

(CLOCA) has observed increased nutrients in Soper Creek in previous monitoring (Central lake Ontario Conservation Authority, 2013a).

## **2.4.3** *Land-use effects on algal growth and community composition*

Algal biomass measures (Chl a and AFDM) were not significantly related to any land-use parameters. However, Pearson correlations showed only significant relationships with Chl a and TSS. Chl a showed a negative relationship with TSS, this indicates that suspended solids could be affecting the amount of light that is penetrating the water. Algae require light to grow, therefore any reductions in light penetration will limit algal growth (Taylor et al., 2004). This result is consistent with Murdock et al. (2013) that found that TSS had a negative association with Chl a. In addition, TSS was found to be negatively correlated with forested land-use. Observing these negative correlations could indicate that the movement from forested and agricultural lands to developed land in these watersheds could be negatively affecting the algal biomass through loadings of suspended solids. However, algal biomass can show inconsistent growth responses to different land-use parameters. Similar to our data, Bourassa and Cattaneo (1998) and Yang et al. (2015) found no significant relationships with nutrient concentrations and periphyton biomass found within varying land-use parameters. These results contrast many studies which have shown relationships with periphyton growth and specific land-use parameters. Agricultural dominated streams have shown positive relationships for periphyton biomass with increased nitrogen and phosphorus concentrations (Lavoie et al., 2004; Jacobson et al., 2008). Furthermore, high urban development has also shown similar relationships through increased concentrations of phosphorus (O'Brien & Wehr 2010). This variability of periphyton biomass suggests that predicting biomass parameters, such as Chl a, could be dependent on multiple factors (e.g., physical and chemical).

Land-use practises can affect the water quality and algal community structure in aquatic ecosystems. The results from the redundancy analysis showed that temperature, TSS, chloride and pH were the most important water quality variables in explaining algal community composition and site locations (Figure 2.11A.). Site locations within the RDA showed a clear separation between agricultural sites and developed sites. This separation among sites indicated that algal community composition was influenced by location and water quality. Additionally, this same separation of sites was seen with the second and third RDA for algal community and land-use (Figure 2.11B and 2.11C.). Based on the redundancy analysis, algal community structure primarily related to water quality variables that were a function of urban development. Factors such as temperature, chloride, pH and TSS were the main factors influencing the algal community. Surprisingly, nutrient concentrations did not have any major influences on algal community structure. This finding is consistent with studies conducted by Winter and Duthie (1998), Sonneman et al. (2001), and Lavoie et al. (2004) that found other factors, such as conductivity, pH and temperature were better indicators of algal community structure. Although the algal communities were observed to be most influenced by factors like, chloride and temperature, I did see that TN: TP did positively correlate with some genus and negatively correlate with phosphorus (Figure 2.11A). This suggests that for most of these genus phosphorus could be a limiting nutrient (Stelzer & Lamberti, 2001).

Different algal species (e.g., diatoms) respond differently to environmental changes, and have varying tolerances to physical or chemical disturbances (Pan et al., 1996; Blinn & Bailey, 2001). Urban development and its associated water quality variables showed a relationship with both moderate and high pollution tolerant algal species. Pollution tolerant algal species, such as *Amphora veneta*, *Cymbella tumida*, and *Gomphonema parvulum* were observed in this study.

According to Cox (1996), G. parvulum is reported as being wide spread and found in a range of different environmental conditions. Many studies report G. parvulum as a high pollution tolerant species found in areas of high nutrients, conductivity and metal pollution (Olding, 2000; Duong et al., 2007; Bere & Mangadze, 2014). This contrasts our study as nutrients did not have an observed relationship with G. parvulum, however, it did show up at sites impacted by urban pollution. Two taxa were found to have negative relationships with developed land-use and road density. The taxa Navicula and Cocconeis were found to be more related to our headwater sites along with forest and agricultural land-use. Two species of *Cocconeis* were observed in this study, Cocconeis pediculus and Cocconeis placentula. This finding is similar to previous studies that found C. pediculus and C. placentula were associated with landscapes dominated by agriculture (Munn et al., 2002; Lavoie et al., 2004). Furthermore, Walsh & Wepener (2009) found that the genera, Navicula and Nitzschia indicated low and high intensity agricultural landuse practises, respectively. In this study, Lynde and Oshawa Creek headwater sites were found to be dominated by genus Navicula, compared to Bowmanville and Soper Creek, which were dominated by *Nitzschia* (Figure 2.7-2.10). This could indicate that agricultural practises near the headwater sites of Lynde and Oshawa Creek were low- intensity compared to Bowmanville and Soper Creek, which could be indicating high intensity land-use practises, however, this would have to be investigated in further studies.

Among all of the study sites *Achnanthidium* was the dominant genus. It included the species *A. minutissimum* and *A. linearis*, which correlated well with temperature, DO, pH and road density. *A. minutissimum* has been considered to be a cosmopolitan species for its ability to tolerate a wide range of environmental conditions and habitats (Medley & Clements 1998). Past studies have found it in varying locations from stormwater ponds to streams impacted by acid

mine drainage (Olding, 2000; Luís et al., 2009; Yang et al., 2015). In this study, it was found at all of the sampling sites, however, it related mostly with Oshawa Creek site 3. Site O3 had the most percent developed land-use and road density in its surrounding area. A study conducted by Teittinen et al. (2015) found similar findings where high abundances of *A. minutissimum* were reported at urban study sites. This high concentration of urban development could be driving this site to show high abundances of *Achnanthidium* through moderate disturbances. Similar results were found by Smucker and Vis (2013), which showed that *A. minutissimum* was able to out compete other species in sites moderately impacted by acid mine drainage. Additionally, the U.S. Environmental Protection Agency have created a metric that utilizes *A. minutissimum* since its abundance has been found to be directly proportionate to the time that a disturbance event has occurred. If the relative abundance of *A. minutissimum* falls between 0-50% there has been no disturbance or a minor disturbance, 50-75% moderate disturbance, and 75-100% severe disturbance (Barbour et al., 1999). Therefore, in this present study I can consider *A. minutissimum* as a possible indicator of urban disturbances.

### 2.4.4 Conclusion

The results of this study found that road density was a better indicator of urbanization than impervious surfaces (i.e., developed land-use). Road density showed stronger correlations among water quality variables than developed land-use, and was found to be more significant within the redundancy analyses. This result is consistent with Wallace et al. (2013) and Wallace and Biastoch (2016), which found that road density was a better indicator of urbanization than impervious surfaces in Toronto, Ontario. Additionally, I found that site catchment was better at characterizing land-use types, which was also best associated with water quality variables than site buffers. This is consistent with Sliva & Williams (2001) that found that catchments scales

were slightly better than site buffers at predicting water quality. However, this is an ongoing dispute between whether larger scales (e.g., catchments) or smaller spatial scales (e.g., buffers) are better at predicting water quality. For example, Lento et al. (2013) found that small scales were better at explaining water quality than catchments, which contrasts this study and Sliva & Williams (2001).

In conclusion, this study has shown that land-use can affect both water quality and algal community structure. The results suggest that water quality variables were most influenced by urban development in comparison to agricultural land-use. In addition, I found that algal community structure responded to multiple water quality variables, which associated with developed land-use. This indicates that the biological quality of these tributaries are being impacted mostly by urban development. Further research is warranted to explore more avenues and pinpoint further causes of possible stream degradation. Ultimately, changes in land-use are capable of influencing the health of aquatic ecosystems, therefore improving our understanding of land-use gradients will be essential for future protection plans.

Chapter 3: Assessing the spatial and temporal influences on water quality and periphyton communities in tributaries of Durham Region, Ontario.

### 3.1 Introduction

Humans are becoming increasingly urban and generating rapid development pressures within watersheds (Wallace & Biastoch, 2016). Urban development not only alters the morphology within a stream, but also fragments the natural landscapes around the stream (Urban et al., 2006). The primary issue with land fragmentation caused by urbanization is the creation of rural-urban land-use gradients. Watersheds experiencing rural-urban land-use gradients are distinct in that their longitudinal flow may have a cumulative effect on the streams that reside there. Water quality within a watershed is heavily influenced by changes in land-use, which can impact the physical, chemical and biological characteristics of an aquatic ecosystem (Pan et al., 2004; Klose et al., 2012; Mei et al., 2014).

As previously mentioned in Chapter 1, lotic systems can naturally experience cumulative increases in water quality parameters, such as nutrients and particulate matter along a longitudinal flow path with increasing drainage area (Vanotte et al., 1980). However, lateral landscape inputs may alter cumulative water quality patterns on a local or reach scale, particularly under changing land-use regimes (Thorpe et al., 2006; Humphries et al., 2014). In addition to these important spatial influences on water quality, temporal or seasonal influences can be very important as well. For example, snow-melt during spring or heavy storm-events during summer can cause pulse inputs of chloride and phosphorus, into surface waters, respectively (Oliver et al., 1974; Deletic, 1998; Bartosova et al., 1999). Although many studies look at spatial and temporal factors when identifying trends in water quality and algal

community structure, there's a paucity of studies examining both factors, particularly the relative contributions of longitudinal and lateral landscape influences.

To improve our understanding of these contributing influences on water quality and algal community biomass and structure, sites selected in the previously described study creeks (Lynde, Oshawa, Bowmanville and Soper) were matched across each watershed (Figure 2.1, Table 2.1). Matching sites for their distance from headwaters in four replicate creek systems is an attempt to control for longitudinal water quality influences when assessing lateral land-use effects on a local or reach scale. If increasing trends of nutrients and particulates (i.e. total suspended solids) persist in the study creeks along their continuum, this would infer an important role for longitudinal flow processes, which reflect cumulative catchment-drainage. Alternatively, if water quality patterns are not a function of distance from headwaters, this may infer that lateral processes such as local land-use near each site is influencing instream water quality patterns.

Therefore, my research objectives for this chapter were to assess my complete field dataset for spatial and temporal patterns in instream water quality and periphyton communities as a function of longitudinal (i.e., cumulative) and/or lateral (i.e., local land-use) factors, over the study period covering four algal substrate deployments (May, June, July and August).

Accounting for spatial location (i.e., distance from headwaters) and seasonality (i.e., monthly variation) was intended to offer new insights into periphyton community abundance and structure in Lake Ontario tributaries that experience gradients in rural-urban land-use.

#### 3.2 Methods

### **3.2.1** *Watershed Descriptions and Site Selection*

Refer to 2.2.1 *Watershed Descriptions and Site Selection* (pg. 20).

# **3.2.2** *Field Measurements and Water Sampling*

Refer to 2.2.2 Field Measurements and Water Sampling (pg. 21) and 2.2.3 Water Chemistry (pg. 24).

# **3.2.3** Algal Sampling and Processing

Refer to 2.2.4 *Use of Artificial Substrates to Measure In-Situ Algal Colonization* and Growth (pg. 24) and 2.2.5 *Algal Processing* (pg. 27).

# **3.2.4** *Statistical Analyses*

Due to either missing tiles or damaged tiles, some algal data is missing for some sites in the months of June, July, and August. See Table 3.1 for a complete listing of algal data available for each site across each month. Spatial and temporal patterns in water quality variables and algal parameters were evaluated with two separate sets of analysis of variance. To evaluate spatial variance within watersheds and temporal variance among sampling months, balanced two-way ANOVA's were run for water quality variables, and unbalanced Two-way ANOVAs for Chl *a* and AFDM. If two-way ANOVAs revealed significant interaction effects, one-way ANOVAs or kruskal-Wallis tests were performed instead. One-way ANOVA's were also run to compare watershed data on a latitudinal scale (i.e., comparing site 1 from each watershed). In addition, I tested spatial patterns among water quality variables with a correlation matrix using distance from headwaters (i.e., longitudinal or cumulative spatial factors) and % developed land-use (i.e.,

lateral or local inputs). Distance from headwaters was calculated in QGIS, using the measure function where distances were measured from the headwaters origin to each site location. Percent developed land-use was calculated using the land-use data extracted from the 1Km buffers used in chapter 2.

Data was tested for normality and homogeneity of variance prior to testing. If necessary, transformations were made to meet parametric assumptions. Lastly, to further test the spatial and temporal patterns in water quality, principal components analysis (PCA) was performed using data from each sampling month.

All statistical tests were completed in R (version 3.2.2, R Development Core Team, Vienna Austria) using the packages, *Vegan* and *ggplot2*. A multivariate approach was taken to investigate the relationships between algal community and water quality parameters. In addition, separate analyses were conducted for each month. Correspondence analysis (CA) were used to evaluate algal community composition among sites for each sampling month.

Detrended Correspondence Analysis (DCA) was used to evaluate if our data was appropriate to run either a Redundancy analysis (RDA) or Canonical Correspondence analysis (CCA). Gradient lengths less than 3 were obtained, which determined my data were linear, and therefore necessary to run an RDA. Separate RDA's were conducted to determine which water quality variables were able to explain algal community composition among all of the sites at each sampling month. In addition, an RDA was conducted with spatial parameters and land-use type to determine how they are associated with monthly water quality. Water quality variables were centered and standardized, and algal community data were square root transformed prior to analysis (Lavoie et al., 2004). In order to remove collinear variables, variables with variation inflation factors (VIF) greater than 20 were removed for this analyses. The variables used in the

final analyses were selected for their ecological importance in this study, and reflect aspects of water quality (e.g., TN and TP), land-use (e.g., TSS and chloride) and natural geology (e.g., pH). Monte Carlo permutation tests were used to test the statistical significance of the RDA axes (999 random permutations, p<0.05).

**Table 3.1** Dates and sites with missing periphyton community data due to missing or disturbed artificial growth substrates. Periphyton sample present = +; Periphyton sample absent = -

Watershed	Site ID	May	June	July	August
Lynde Cr.	L1	+	+	-	+
Lynde Cr.	L2	+	+	+	+
Lynde Cr.	L3	+	+	+	+
Lynde Cr.	L4	+	+	+	+
Oshawa Cr.	O1	+	+	+	+
Oshawa Cr.	O2	+	+	+	+
Oshawa Cr.	O3	+	+	-	+
Oshawa Cr.	O4	+	+	+	+
Bowmanville Cr.	B1	+	+	+	+
Bowmanville Cr.	B2	+	+	+	+
Bowmanville Cr.	В3	+	+	+	+
Bowmanville Cr.	B4	+	-	-	+
Soper Cr.	<b>S</b> 1	+	-	+	+
Soper Cr.	S2	+	+	+	-
Soper Cr.	<b>S</b> 3	+	+	+	+
Soper Cr.	S4	+	+	+	+

# 3.3 Results

# **3.3.1** *Spatial and Temporal Variation in Water Quality*

Water quality variables varied along each watershed over the sampling period (Table 3.2, Figure 3.1, and Figure B1). Among all of the watersheds, Lynde Creek showed the largest increase of chloride from site 1 to site 4. In addition, Lynde Creek site 3 had the highest concentration of chloride consistently over the sampling months. DO, pH, and TSS were highly variable among all of the study sites and sampling months. Temperature showed increases at both study sites and sampling months at each watershed. Nutrient concentrations of TN and TP did not vary much within most watersheds, however, compared among watersheds nutrient concentrations did vary. Among all of the study sites Lynde creek site 1 showed the highest concentration of TP, while Soper Creek study sites were observed to have the highest TN concentrations.

Spatial and temporal variation was assessed for each watershed to compare study sites and sampling month (Table 3.3-3.6). Two-way ANOVA results for Lynde Creek revealed no significant interactions for all tests run, except for TP. Significant differences for both sites and sampling month were observed for chloride, temperature, and TSS (Table 3.3). TN and pH did not show any significant differences for sites, however, significant differences were observed for sampling months. Two-way ANOVA results for Oshawa creek showed significant interactions for TSS and chloride, indicating that the sites are showing different monthly patterns (Table 3.4). Temperature, DO, and pH revealed significant differences between sites and sampling month. Nutrient concentrations of TP and TN did not show any significance between sites, however, significance was seen among sampling months. Results for Bowmanville creek showed no

interaction effects for all of the water quality variables tested (Table 3.5). DO and TP were observed to show no significant differences between sites and sampling months, however, chloride, temperature, and TSS were shown to have significant differences in both. TN and pH did not show any significant differences between sites, yet sampling month did reveal significant differences. Lastly, Soper Creek revealed significant differences only in chloride and pH between site and sampling month (Table 3.6).

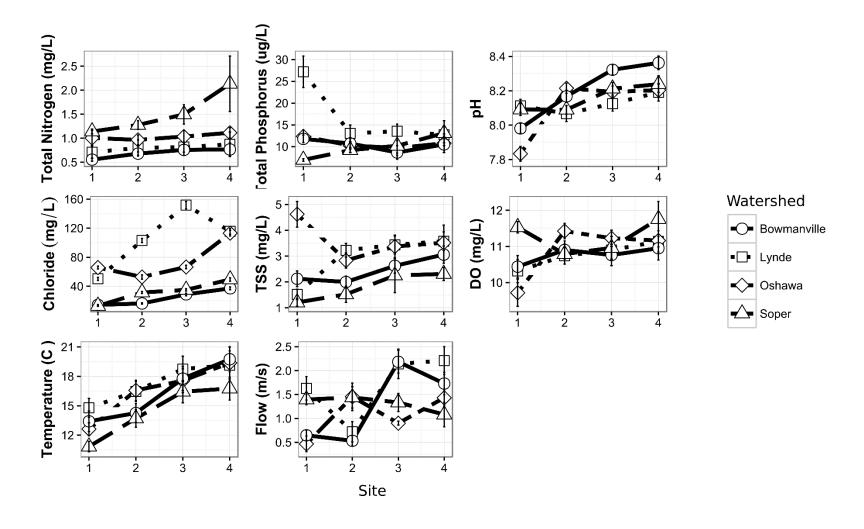
One-way analysis of variance were performed for water quality variables that revealed interactions between site and sampling month (Table B1-B4). ANOVAs run for TP at Lynde Creek revealed that there was at least one site that was significantly different from others, however, no significant differences were observed for sampling months. Tests for Oshawa Creek showed significant differences with chloride for site and date, but TSS only showed significant differences for dates.

**Table 3.2** Sampling season means of water quality parameters. Superscript values represent different samples sizes for each water quality parameter: 1 (n=8) and 2 (n=24).

Watershed	Site	pH <sup>1</sup>	TSS <sup>2</sup>	Chloride <sup>2</sup>	$DO^1$	Temperature <sup>1</sup>	TP <sup>1</sup>	TN <sup>1</sup>	Flow <sup>1</sup>
	ID		$(mg L^{-1})$	$(mg L^{-1})$	(mg L <sup>-1</sup> )	(°C)	$(\mu g L^{-1})$	(mg L <sup>-1</sup> )	$(m s^{-1})$
Lynde Cr.	L1	8.11 (0.11)	1.52 (1.0)	50 (10)	10.32 (0.91)	14.79 (2.7)	27.19 (10)	0.70 (0.35)	1.63 (0.7)
Lynde Cr.	L2	8.07 (0.13)	3.23 (1.3)	103 (14)	10.74 (0.64)	16.46 (2.9)	13.05 (5.4)	0.79 (0.47)	0.72(0.6)
Lynde Cr.	L3	8.13 (0.12)	3.43 (1.7)	152 (26)	10.90 (0.70)	18.75 (3.7)	13.53 (4.7)	0.81 (0.30)	2.14(0.9)
Lynde Cr.	L4	8.19 (0.14)	3.58 (1.8)	116 (20)	11.14 (0.83)	19.15 (4.1)	12.71 (5.2)	0.87 (0.72)	2.10 (0.8)
Oshawa Cr.	O1	7.83 (0.12)	6.31 (2.3)	66 (9.3)	9.72 (1.1)	12.62 (1.8)	12.36 (4.1)	1.00 (0.44)	0.47(0.5)
Oshawa Cr.	O2	8.22 (0.08)	2.83 (1.4)	53 (14)	11.42 (0.60)	16.57 (2.9)	10.27 (4.7)	0.96 (0.29)	1.45 (0.8)
Oshawa Cr.	O3	8.19 (0.09)	3.37 (2.2)	67 (11)	11.22 (0.64)	17.56 (3.4)	9.69 (4.8)	1.03 (0.35)	0.90(0.1)
Oshawa Cr.	O4	8.20 (0.10)	3.52 (3.3)	113 (19)	11.16 (0.49)	19.37 (4.5)	10.77 (6.0)	1.11 (0.30)	1.43 (0.5)
Bowmanville Cr.	B1	7.98 (0.08)	2.12 (1.5)	14 (3.3)	10.44 (0.88)	13.41 (3.0)	11.78 (2.5)	0.55 (0.09)	0.65(0.3)
Bowmanville Cr.	B2	8.17 (0.10)	2.00 (1.2)	16 (4.4)	10.90 (0.71)	14.26 (2.8)	10.73 (3.0)	0.67 (0.10)	0.53(0.3)
Bowmanville Cr.	В3	8.32 (0.08)	2.62 (1.4)	29 (9.0)	10.76 (0.83)	17.77 (3.4)	8.66 (4.4)	0.75 (0.32)	2.18 (0.7)
Bowmanville Cr.	B4	8.36 (0.11)	3.06 (1.7)	37 (9.9)	10.95 (0.92)	19.75 (3.6)	10.51 (5.3)	0.76 (0.37)	1.73 (0.7)
Soper Cr.	<b>S</b> 1	8.09 (0.09)	1.21 (0.7)	13 (3.8)	11.53 (0.44)	10.83 (1.5)	6.95 (0.80)	1.14 (0.14)	1.40 (0.3)
Soper Cr.	S2	8.09 (0.07)	1.52 (1.5)	31 (6.1)	10.79 (0.79)	13.71 (2.5)	9.20 (1.7)	1.27 (0.25)	1.44 (0.6)
Soper Cr.	<b>S</b> 3	8.21 (0.10)	2.25 (3.2)	35 (8.2)	10.96 (0.91)	16.43 (3.2)	10.20 (4.7)	1.49 (0.57)	1.33 (0.5)
Soper Cr.	S4	8.24 (0.14)	2.31 (1.3)	49 (12)	11.76 (1.4)	16.76 (3.3)	13.10 (8.0)	2.13 (1.6)	1.08 (0.7)

Standard Deviation is in Brackets

Cond = Conductivity, TSS = Total Suspended Solids, DO = Dissolved Oxygen, TP = Total Phosphorus, TN = Total Nitrogen



**Figure 3.1** Spatial trends for mean water quality parameters with standard errors measured at each study site. Temporal trends are presented in Appendix B.

**Table 3.3** Balanced two-way analysis of variance results for water quality variables among sites and sampling month for **Lynde Creek.** 

Term	df	F value	P value
TN			
Site	3	0.190	0.900
Month	3	4.98	0.013
Interaction	9	0.326	0.950
TP			
Site	3	22.7	P<0.001
Month	3	8.55	P<0.001
Interaction	9	3.42	0.0160
pН			
Site	3	2.05	0.150
Month	3	8.02	P<0.001
Interaction	9	0.34	0.950
DO			
Site	3	1.37	0.289
Month	3	2.02	0.150
Interaction	9	0.23	0.980
Temperature	<b>!</b>		
Site	3	3.52	0.040
Month	3	5.79	P<0.001
Interaction	9	0.0480	0.990
TSS			
Site	3	11.3	P<0.001
Month	3	2.99	0.0360
Interaction	9	1.33	0.240
Chloride			
Site	3	142	P<0.001
Month	3	3.63	0.0170
Interaction	9	1.61	0.130

**Table 3.4** Balanced two-way analysis of variance results for water quality variables among sites and sampling month for **Oshawa Creek**.

Term	df	F value	P value
Chloride			
Site	3	251.19	P<0.001
Month	3	55.65	P<0.001
Interaction	9	2.75	P<0.001
DO			
Site	3	10.11	P<0.001
Month	3	4.30	0.021
Interaction	9	0.22	0.98
pН			
Site	3	52.35	P<0.001
Month	3	10.83	P<0.001
Interaction	9	0.72	0.68
Temperature			
Site	3	6.72	P<0.001
Month	3	4.73	0.015
Interaction	9	0.15	0.99
TN			
Site	3	0.27	0.85
Month	3	3.99	0.027
Interaction	9	0.22	0.98
TP			
Site	3	0.53	0.67
Month	3	5.00	0.012
Interaction	9	0.41	0.23
TSS			
Site	3	2.89	0.04
Month	3	4.13	P<0.001
Interaction	9	2.74	P<0.001

**Table 3.5** Balanced two-way analysis of variance results for water quality variables among sites and sampling month for **Bowmanville Creek.** 

Term	df	F value	P value
Chloride			
Site	3	77.97	P<0.001
Month	3	15.71	P<0.001
Interaction	9	0.89	0.538
DO			
Site	3	0.52	0.67
Month	3	2.45	0.10
Interaction	9	0.11	0.99
pН			
Site	3	25.65	P<0.001
Month	3	3.42	0.054
Interaction	9	0.25	0.98
Temperature			
Site	3	3.88	0.011
Month	3	12.17	P<0.001
Interaction	9	0.173	0.99
TN			
Site	3	1.61	0.23
Month	3	4.75	0.015
Interaction	9	0.99	0.48
TP			
Site	3	0.70	0.56
Month	3	1.96	0.16
Interaction	9	0.28	0.97
TSS			
Site	3	3.33	0.024
Month	3	6.23	P<0.001
Interaction	9	1.20	0.31

**Table 3.6** Balanced two-way analysis of variance results for water quality variables among sites and sampling month for **Soper Creek.** 

Term	df	F value	P value
Chloride			
Site	3	120.10	P<0.001
Month	3	14.43	P<0.001
Interaction	9	0.92	0.512
DO			
Site	3	1.93	0.17
Month	3	3.06	0.059
Interaction	9	0.33	0.95
pН			
Site	3	6.64	P<0.001
Month	3	7.43	P<0.001
Interaction	9	0.275	0.97
Temperature			
Site	3	7.01	P<0.001
Month	3	2.49	0.098
Interaction	9	0.059	0.99
TN			
Site	3	2.41	0.10
Month	3	3.08	0.057
Interaction	9	0.91	0.54
TP			
Site	3	2.19	0.13
Month	3	1.62	0.22
Interaction	9	0.64	0.75
TSS			
Site	3	2.01	0.12
Month	3	2.34	0.080
Interaction	9	1.10	0.371

Next, one-way ANOVAs were used to compare sites across watersheds in order to assess water quality variables across a latitudinal scale (Table 3.7). ANOVAs revealed significant differences for every water quality variable when comparing site 1 from each watershed.

Comparison of site 2 revealed significant differences for chloride, pH, TSS, and TN. Lastly, site 3 and site 4 comparisons showed significant differences for chloride, pH, and TN. No significant differences were revealed for DO and TP for site 2, site 3, and site 4 comparisons.

Further examination using Pearson correlation analysis for distance from headwaters (i.e., longitudinal) revealed a few relationships with water quality variables (Table 3.8). Distance from headwaters revealed significant positive relationships with chloride (r=0.14), DO (r=0.22), pH (0.58), and temperature (r=0.61), while showing no significant relationships with TN, TP and TSS. Percent developed land-use revealed significant positive correlations for pH (r= 0.65) and temperature (r= 0.81). Further correlations for the percent developed land-use from the 1Km buffers can be viewed in chapter 2 (table 2.8).

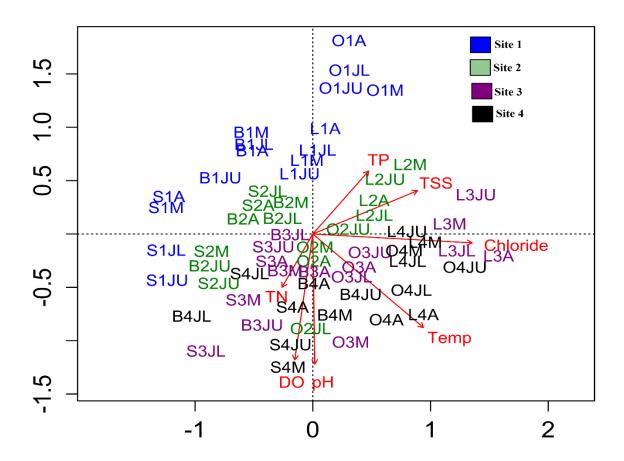
**Table 3.7** Summary F-values (f) and P-values (p) from one-way ANOVA comparing water quality variables from matched distance-from-headwater sites across watersheds. Superscript values denote sample size. 1 = (n=24, k=4); 2 = (n=8, k=4).

Site	Te	mp <sup>2</sup>	р	$\mathbf{H}^2$	D	$OO^2$	Chlo	oride <sup>1</sup>	Т	SS <sup>1</sup>	7	ΓN <sup>2</sup>	T	$\mathbf{P}^2$
	f	p	f	p	f	p	f	p	f	p	f	p	f	p
Site 1	4.02	0.017	12.37	< 0.001	6.24	< 0.001	310.30	< 0.001	23.56	< 0.001	6.55	< 0.001	19.01	< 0.001
Site 2	2.26	0.103	3.80	0.021	1.67	0.195	294.90	< 0.001	7.87	< 0.001	5.61	< 0.001	1.26	0.31
Site 3	0.61	0.613	5.19	< 0.001	0.50	0.683	329.20	< 0.001	1.63	0.19	5.59	< 0.001	1.64	0.20
Site 4	0.96	0.425	3.16	0.040	1.07	0.377	163.90	< 0.001	1.78	0.16	3.66	0.024	0.36	0.78

**Table 3.8** Pearson correlation coefficients for correlation analyses run between distance-from-headwaters and % developed land-use with water quality variables (dependent variables) for all sites and sampling periods. Bold numbers indicate statistically significant correlations. Scatterplots for statistically significant correlations are presented in Appendix B.

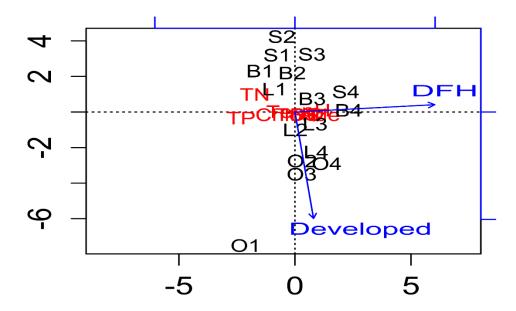
Variable	Chloride	DO	pН	Temp	TN	LogTP	TSS
Distance from headwaters	0.14	0.22	0.58	0.61	0.046	0.063	0.086
%Developed land- use	0.46	0.47	0.65	0.81	0.07	-0.24	0.21

A Principal component analysis (PCA) was performed using data from all sampling months (Figure 3.2). The first two axes of the PCA explained 61.7% of the total variation. Sites tended to cluster together regardless of month. Lynde Creek Sites 3 and 4 were associated with high chloride in all sampling months, whereas Soper Creek Sites 3 and 4 were associated with elevated TN for all months. This is consistent with the non-standardized data, where Lynde creek site 3 and 4 had the highest chloride concentrations (~130-180 mg/L) and Soper Creek had the highest nitrogen concentrations (~1.8-2.4 mg/L). Bowmanville and Soper Creek Site 2 always grouped close together, and Oshawa Creek site 1 seems to be an outlier across all months. Water quality variables DO, TN and TP revealed inverse relationships with one another over the sampling season. Lastly, near-headwater sites consistently separated from all other sites indicating distinct water quality profiles from more urban sites. Further examination of temporal variations in water quality can be viewed in in Appendix B.



**Figure 3.2** Principal component analysis for all sampling months with standardized water quality parameters among all study sites. The two axis have a combined variation explained of 61.66% with axis one and two explaining 34.44% and 27.22% of the variation, respectively. Month codes include: May= M, June=JU, July= JL, and August= A.

The spatial and land-use RDA performed on the seasonal water quality data explained 32.6% variance while the first two axis were able to explain 29.9% and 2.72% of the variation, respectively (Figure 3.3). Monte carlo permutation tests showed the relationships between water quality variables and the spatial and land-use variables were observed to be statistically significant (999 permutations, p<0.05). Many of the water quality variables within the RDA were found within the middle of the RDA indicating their association with both distance from headwaters and percent developed land-use. Nutrients variables revealed to show no correlations with developed land-use, and inverse relationships with distance from headwaters. Site locations within the RDA showed sites closest to the headwaters (i.e., site 1) gathered on the left side with the exception of Oshawa Creek site 1, and the more developed sites (i.e., site 3 and 4) on the right side of the second axis.



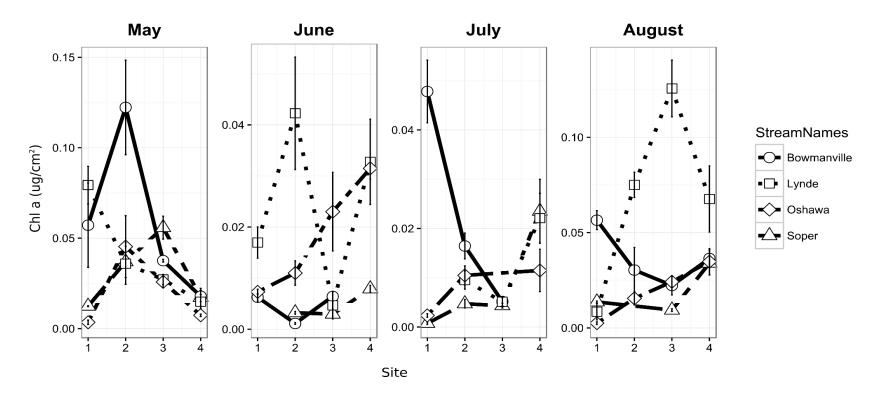
**Figure 3.3** RDA showing the relationships of distance from headwaters (i.e., longitude or cumulative inputs) and percent developed (i.e., lateral or local inputs) with seasonal water quality variables and sampling sites. Site and water quality variable abbreviations can be found in Table 3.2. Monte carlo permutations found this RDA to be statistically significant (999 random permutations, P<0.05).

### **3.3.2** *Spatial and temporal variation in Chlorophyll a and AFDM*

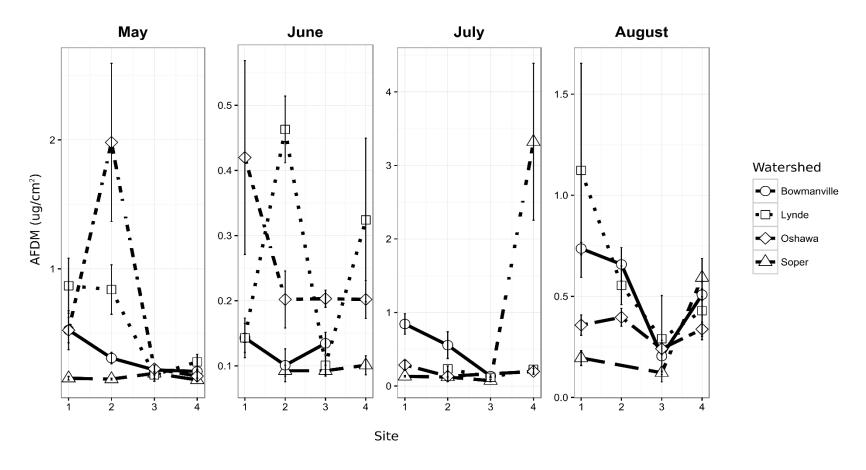
Chlorophyll *a* and AFDM concentrations varied across sites over the sampling period. For Chl *a*, the highest concentrations were observed in Bowmanville Creek site 2 in May, Lynde Creek site 2 in June, Bowmanville Creek site 1 in July and Lynde Creek site 3 in August (Figure 3.4). The variation of chlorophyll over the months is interesting, because the month of May and August showed higher Chl *a* peaks then June and July, which are summer months. AFDM concentrations among the months was highly variable for each site and watershed (Figure 3.5). This suggests that all of the organic material found on the ceramic tiles can change dramatically over time. Similar to Chl *a*, some of the lowest AFDM concentrations were recorded in June and July.

Unbalanced two-way ANOVAs revealed interaction effects for Chl *a* and AFDM among sites, and sampling dates for all watersheds (Table 3.9-3.12) with the exception of Lynde Creek and Bowmanville Creek for AFDM. The unbalanced two-way ANOVA for Lynde Creek and Bowmanville Creek revealed significant differences for AFDM among site and sampling month. Kruskal-Wallis ANOVA tests were performed for algal biomass variables that revealed interactions between site and sampling month (Table B1-B4). Chl *a* showed no significant differences among sites for all watersheds, except for Oshawa Creek. In comparison, Chl *a* showed significant differences for sampling month for all watersheds, except for Oshawa Creek. Oshawa creek revealed only significant differences in sampling month for AFDM, and Soper Creek showed only significant differences for sites for AFDM. Kruskal-Wallis ANOVAs for site comparisons across the watersheds only showed significant differences for Chl *a* at site 1 study sites (Table 3.13). Site comparisons for AFDM revealed that study sites 1, site 2 and site 3

showed significant differences among watersheds, however, site 4 study sites did not show any significant differences (Table 3.14).



**Figure 3.4** Total Chl *a* concentrations for study sites across all watersheds for each sampling month, including standard error bars.



**Figure 3.5** Total AFDM *a* concentrations for study sites amongst all watersheds for each sampling month, including standard error.

**Table 3.9** Unbalanced two-way analysis of variance results for water quality variables among sites and sampling month for **Lynde Creek.** 

Term	df	F value	P value
Log Chl a			
Site	3	1.038	0.39
Date	3	33.68	P<0.001
Interaction	9	15.17	P<0.001
<b>AFDM</b>			
Site	3	6.52	P<0.001
Date	3	3.71	0.022
Interaction	9	1.45	0.211

**Table 3.10** Unbalanced two-way analysis of variance results for water quality variables among sites and sampling month for **Oshawa Creek.** 

Term	df	F value	P value
Log Chl a			
Site	3	42.83	P<0.001
Month	3	4.34	P<0.001
Interaction	9	6.03	P<0.001
<b>AFDM</b>			
Site	3	5.32	P<0.001
Month	3	10.05	P<0.001
Interaction	9	6.73	P<0.001

**Table 3.11** Unbalanced two-way analysis of variance results for water quality variables among sites and sampling month for **Bowmanville Creek.** 

Term	df	F value	P value
Log Chl a			
Site	3	1.038	0.39
Date	3	33.68	P<0.001
Interaction	9	15.17	P<0.001
<b>AFDM</b>			
Site	3	6.52	P<0.001
Date	3	3.71	0.022
Interaction	9	1.45	0.211

**Table 3.12** Unbalanced two-way analysis of variance results for water quality variables among sites and sampling month for **Soper Creek.** 

Term	df	F value	P value
Log Chl a			
Site	3	42.83	P<0.001
Month	3	4.34	P<0.001
Interaction	9	6.03	P<0.001
<b>AFDM</b>			
Site	3	5.32	P<0.001
Month	3	10.05	P<0.001
Interaction	9	6.73	P<0.001

**Table 3.13** Kruskal-Wallis results for site comparison among each watershed for Chl a.

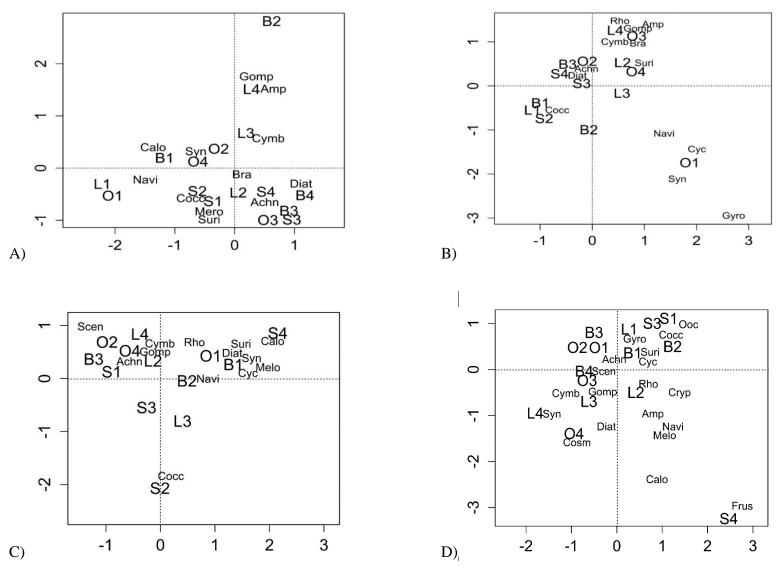
Term	X <sup>2</sup>	P Value
Site 1	17.9	P<0.001
Site 2	6.51	0.09
Site 3	3.30	0.35
Site 4	3.14	0.37

**Table 3.14** Kruskal-Wallis results for site comparison among each watershed for AFDM.

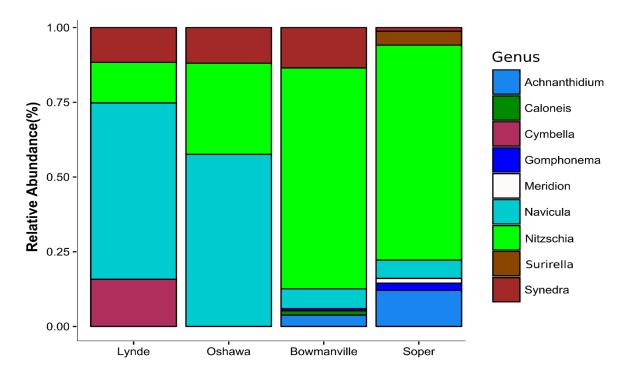
Term	$X^2$	P Value
Site 1	12.0	0.007
Site 2	14.3	0.003
Site 3	14.2	0.003
Site 4	2.14	0.54

### **3.3.3** *Algal Community Composition*

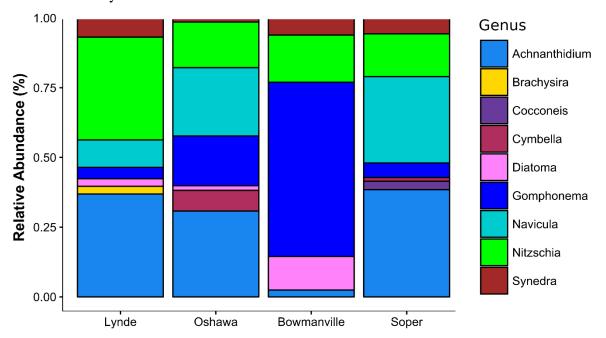
Across all sites and the entire study period, 47 algal species were identified from 24 different genera. Five genera and ten species representing rare taxa were removed from further analyses. The algal community assemblages among all of the sites were dominated by the genera, Achnanthidium (41.85%), Nitzschia (15.35%) and Gomphonema (9.18%). Algal community structure found at each study site varied from month to month. CA ordinations for each sampling month showed distinct patterns in algal taxa among all study sites. The month of May, showed site 1 sites farthest away from the developed sites, except for Oshawa Creek site 4 (Figure 3.6A). Similar groupings among site 3 and site 4 sites were observed, however, site 2 study sites seemed to deviate from one another. Ordinations for June and July showed no obvious patterns with regards to study site locations and algal taxa (Figure 3.6B, C). However, the genera Gomphonema, Cymbella, and Amphora were associated with Lynde Creek site 3 and 4 in these months, as well as May. In comparison, August showed site 4 locations group together, except for Soper Creek site 4. Site 1 study sites were found to group together for August as well (Figure 3.6D). Further similarities between sites can be seen in relative abundance plots found in Figures 3.7-3.22 and within dendrograms in Appendix B. In addition, relative biovolume graphs and Pearson correlations can be viewed in Appendix B.



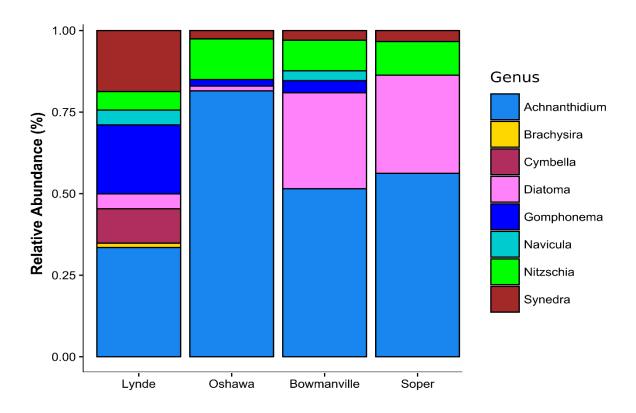
**Figure 3.6** Correspondence Analysis biplot of algal community composition (relative abundances) across sites for all sampling months (A=May, B=June, C=July, D=August). The Correspondence analysis is based on the relative abundances. Taxa codes are represented in Table 2.7.



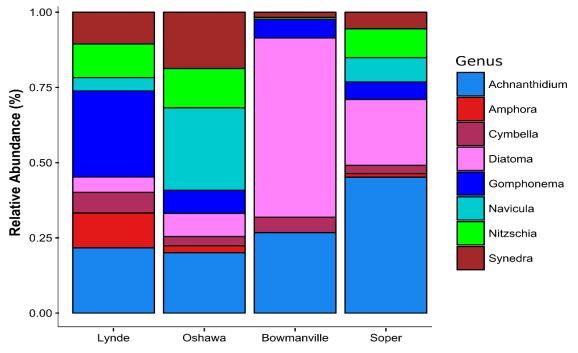
**Figure 3.7** Site 1 comparison of relative abundance based on cell density for all watersheds for the month of May.



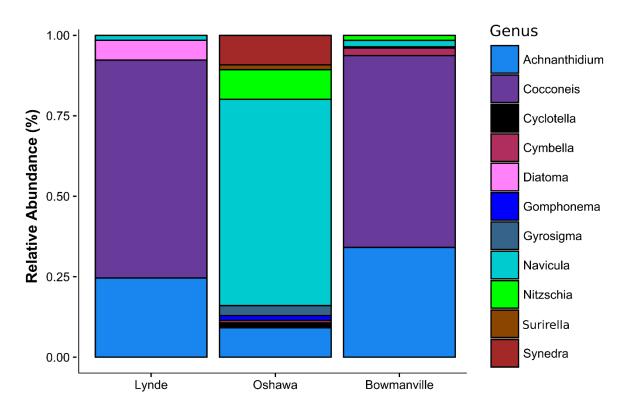
**Figure 3.8** Site 2 comparison of relative abundance based on cell density for all watersheds for the month of May.



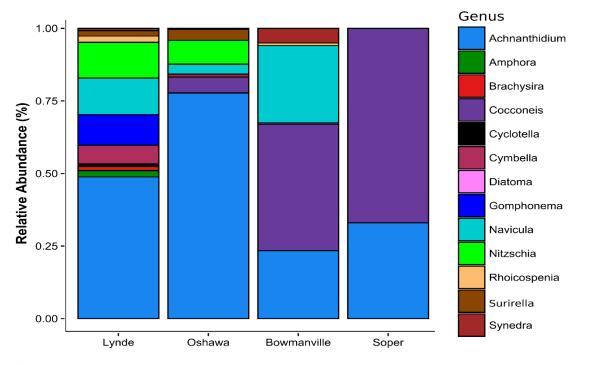
**Figure 3.9** Site 3 comparison of relative abundance based on cell density for all watersheds for the month of May.



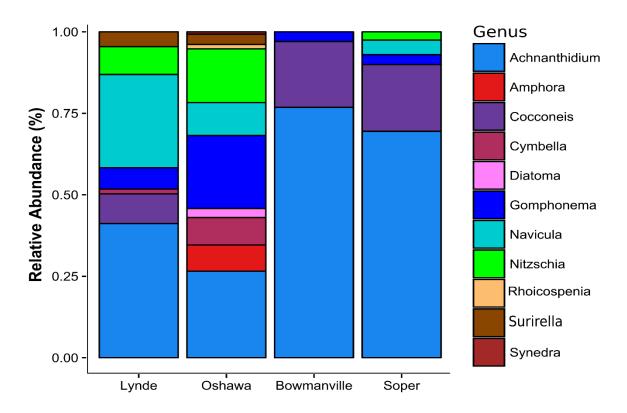
**Figure 3.10** Site 4 comparison of relative abundance based on cell density for all watersheds for the month of May.



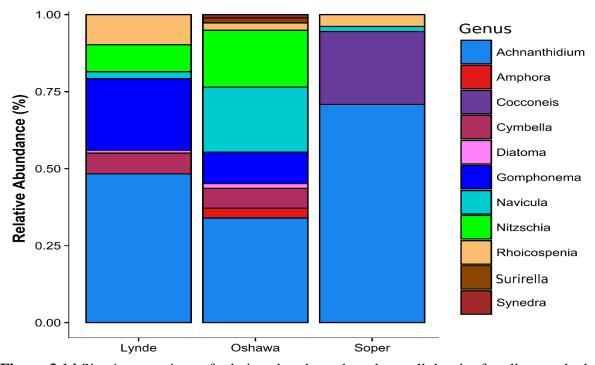
**Figure 3.11** Site 1 comparison of relative abundance based on cell density for all watersheds for the month of June.



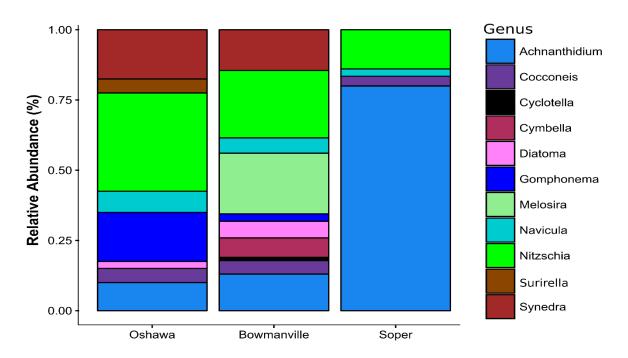
**Figure 3.12** Site 2 comparison of relative abundance based on cell density for all watersheds for the month of June.



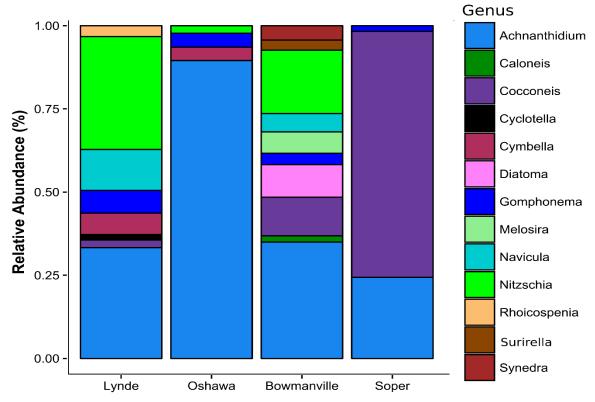
**Figure 3.13** Site 3 comparison of relative abundance based on cell density for all watersheds for the month of June.



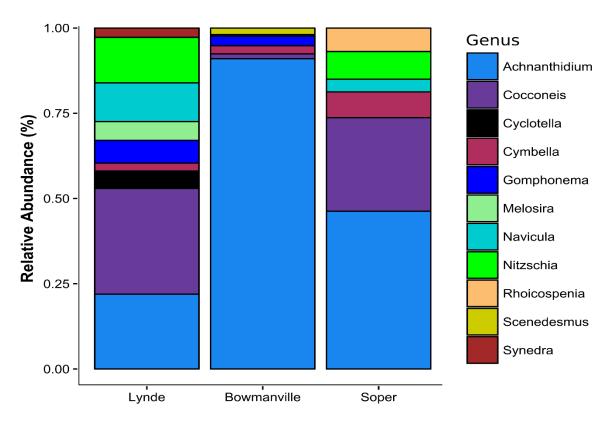
**Figure 3.14** Site 4 comparison of relative abundance based on cell density for all watersheds for the month of June.



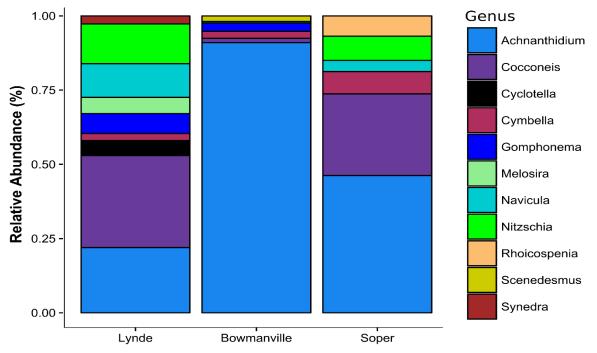
**Figure 3.15** Site 1 comparison of relative abundance based on cell density for all watersheds for the month of July.



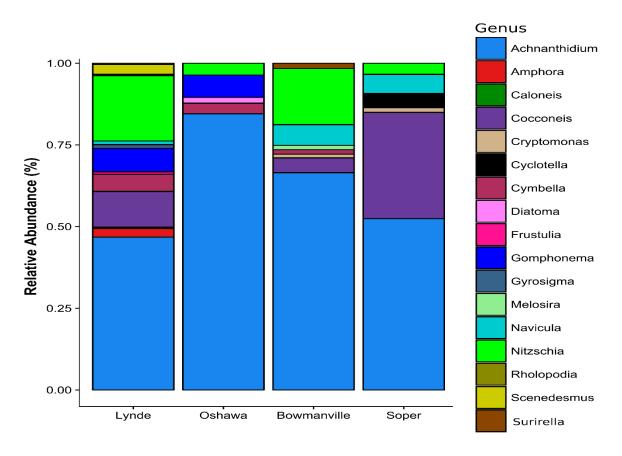
**Figure 3.16** Site 2 comparison of relative abundance based on cell density for all watersheds for the month of July.



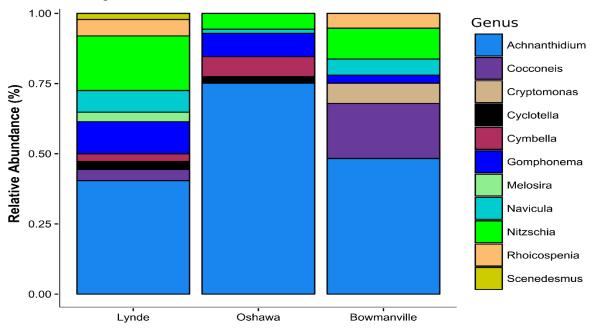
**Figure 3.17** Site 3 comparison of relative abundance based on cell density for all watersheds for the month of July.



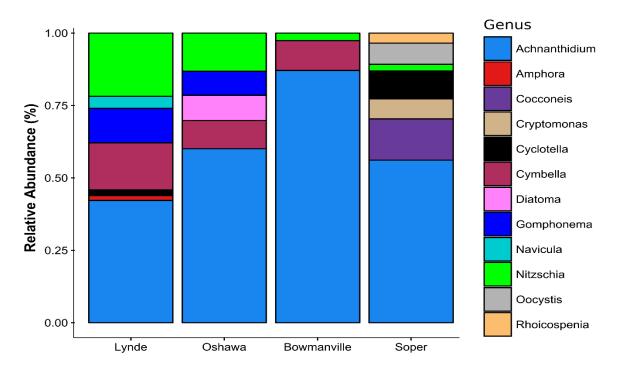
**Figure 3.18** Site 4 comparison of relative abundance based on cell density for all watersheds for the month of July.



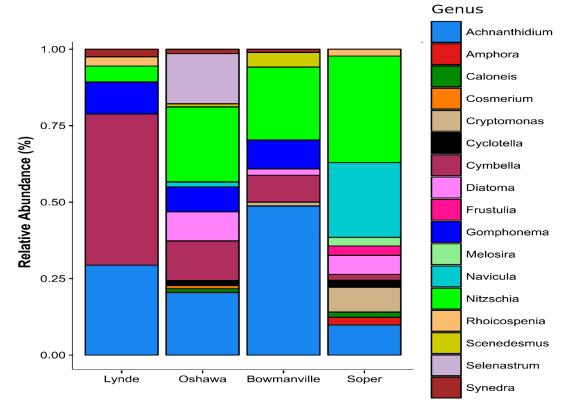
**Figure 3.19** Site 1 comparison of relative abundance based on cell density for all watersheds for the month of August.



**Figure 3.20** Site 2 comparison of relative abundance based on cell density for all watersheds for the month of August.

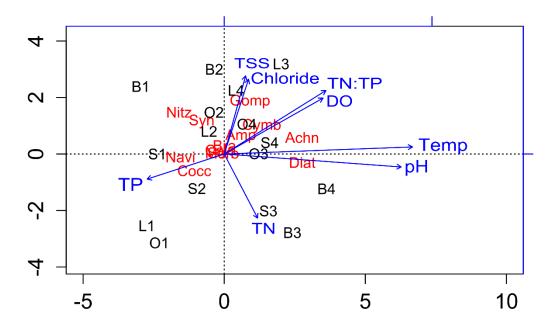


**Figure 3.21** Site 3 comparison of relative abundance based on cell density for all watersheds for the month of August.



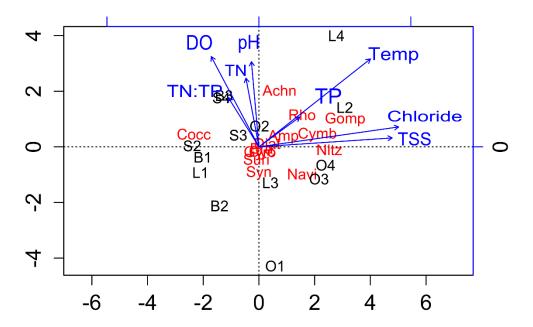
**Figure 3.22** Site 4 comparison of relative abundance based on cell density for all watersheds for the month of August.

The variation in algal community structure seen in the monthly CA ordinations were also observed in the monthly redundancy analyses. The RDA for the month of May explained 73.66% variation with the first two axes accounting for 40.34% and 13.73%, respectively (Figure 3.23). Monte carlo permutation tests revealed that the algal taxa and water quality parameters were statistically significant (999 random permutations, p<0.05). Within the RDA, many of the algal taxa clumped towards the middle, however, some were associated with specific water quality parameters. The taxa, *Navicula* and *Cocconeis* showed a positive correlation with TP, however, it negatively correlated with DO. In comparison, *Cymbella*, *Amphora*, and *Gomphonema* showed the reverse, being positively correlated with chloride, DO, TN:TP and TSS while negatively correlating with TP. Regarding the study sites, site 1 sites for the watersheds gathered on the left side of axis two, compared to most of the site 3 and site 4 sites, which gathered on the right side.



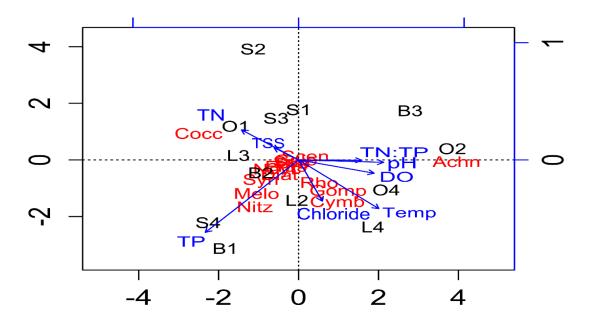
**Figure 3.23** RDA showing the relationships between water quality variables with species and sampling sites for the month of May. Taxa codes are represented in Table 2.7. Monte carlo permutation tests revealed this RDA to be statistically significant (999 random permutations, p<0.05).

The RDA for the month of June explained 68.60% total variation with the first two axis accounting for 45.98% and 15.76% variation explained (Figure 3.24). Monte carlo permutations tested revealed that the algal taxa and water quality parameters were not statistically significant (999 random permutations, p>0.05). For the month of June, *Rhoicospenia* was positively correlated with TP and temperature. *Cymbella*, *Amphora*, *Gomphonema* and *Nitzschia* were found to be positively associated with chloride and TSS, however, *Cocconeis* was negatively associated with those variables. Bowmanville creek and Soper creek site locations were all found on the left side of axis two compared to the more developed sites, Lynde Creek and Oshawa Creek site 4, found on the right side.



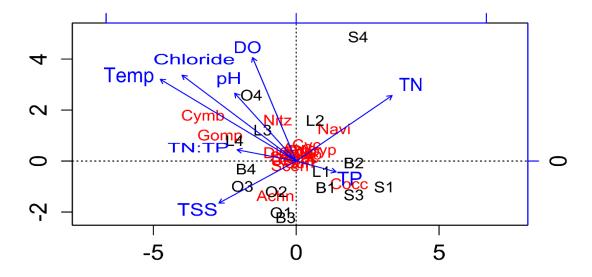
**Figure 3.24** RDA showing the relationships between water quality variables with species and sampling sites for the month of June. Taxa codes are represented in Table 2.7. Monte carlo permutation tests revealed this RDA was not statistically significant (999 random permutations, p>0.05).

The month of July explained 76.68% total variation with the first two axis accounting for 35.43% and 18.13% variation explained (Figure 3.25). Monte carlo permutations tested revealed that the algal taxa and water quality parameters were statistically significant (999 random permutations, p<0.05). Within the RDA for this month, more algal taxa were shown to be associated with a specific water quality variable than by multiple variables. Many algal taxa including, *Melosira*, *Synedra*, *Navicula*, and *Nitzschia* showed positive correlations with TP. *Cymbella* and *Gomphonema* were found to be positively correlated with chloride and temperature. In contrast, *Cocconeis* was positively correlated with TN and TSS, however, it was negatively correlated with Chloride and temperature. The study site locations within this RDA did not show much of a spatial pattern, site 1 sites were found on the left side of the second axis along with site 2 sites, site 3 sites, and Soper Creek site 4.



**Figure 3.25** RDA showing the relationships between water quality variables with species and sampling sites for the month of July. Taxa codes are represented in Table 2.7. Monte carlo permutation tests revealed this RDA to be statistically significant (999 random permutations, p<0.05).

The August RDA explained 70.62% total variation with the first two axis explaining 51.17% and 15.16% variation explained (Figure 3.26). Monte carlo permutation tests showed the relationships between algal taxa and water quality parameters for the month of August to be statistically significant (999 random permutations, p<0.05 Within the RDA for this month, many algal taxa were shown to be associated with a specific water quality variable than by multiple variables. *Cocconeis* was positively correlated with TP and negatively correlated with DO. *Gomphonema* and *Cymbella* were found to be positively correlated with temperature and TN: TP. Again, site locations did not show much of a spatial pattern within this RDA for water quality and algal community structure.



**Figure 3.26** RDA showing the relationships between water quality variables with species and sampling sites for the month of August. Taxa codes are represented in Table 2.7. Monte carlo permutation tests revealed this RDA to be statistically significant (999 random permutations, p<0.05).

#### 3.4 Discussion

### **3.4.1** *Spatial and Temporal patterns in Water Quality*

Water quality parameters were highly variable among watersheds both spatially and temporally. Spatial differences between watersheds and sites were apparent for every water quality variable. Chloride was observed to be significantly different between watersheds and sites. Chloride has been shown to be related with high levels of urban development (Bazinet et al., 2010; Porter-Goff et al., 2013). Two of the watersheds, Lynde Creek and Oshawa Creek are far more developed than the Bowmanville Creek and Soper Creek watersheds (Figure 2.2), therefore a difference in chloride concentrations was expected. Lynde Creek Site 3 and 4 were found to be most associated with chloride among all of the study sites for each sampling month. Lynde Creek watershed is the closest watershed in this study to Toronto, Ontario, Canada, which is heavily urbanized and filled with many impermeable surfaces. Municipalities, like Durham Region are undergoing rapid urban expansion, which is attracting more people from Toronto to move east towards Durham Region (Wallace & Biastoch, 2016). The implications of rapid development involves altering the natural hydrological regime resulting in flashy hydrographs, and increases in nutrients and contaminants (Wallace et al., 2013).

Two other water quality variables, pH and TN were found to show significant differences between watersheds and sites. Observing significant differences for pH among the watersheds and sites was a surprising discovery. The pH of a tributary in a watershed is generally due to the natural geology of the watershed, however, the addition of constituents or contaminants (e.g., metals and salts) can have an impact on the pH of a stream (Luís et al., 2009). The geology of the four watersheds are similar, however, the tributaries do traverse through varying rural-urban

gradients, which could possibly influence the pH of the tributary. In this study, I did see an increase in pH among all watersheds travelling from the headwater sites to the most developed sites, which could indicate that urban development is influencing pH. Recent studies have found that urbanization can affect the pH of freshwater streams. Nagy et al. (2012) found that watersheds with higher impervious surfaces (i.e., more urbanization) had higher median pH than watersheds that contained less impervious surfaces. A similar study conducted by (Chadwick et al., 2012) revealed the same pattern in which pH was associated with increases in urbanization.

TN was found to be highest in the Soper Creek watershed, when compared to the three other watersheds studied. Agricultural land-use is known to influence the concentrations of nutrients, such as TN, within tributaries through the application of fertilizers (De Wit et al., 2005; Black et al., 2011). Soper Creek does have a fair amount of agricultural land-use around its sites, which could explain why the TN is higher than the other watersheds. However, in contrast, Bowmanville creek sites have approximately the same amount of agricultural land-use, but have significantly less TN than Soper Creek sites. In addition, Lynde Creek and Oshawa Creek site 1 sites have a higher amount of agricultural land-use than Soper Creek Site 1, yet have lower amounts of TN. It is possible that multiple factors could be influencing the TN concentrations, such as fertilizer type, and crop type (Oenema et al., 1998; Peterson et al., 2001).

Many water quality variables were observed to have significant differences between watersheds, however, when site comparisons were conducted only Site 1 sites revealed significant differences. These water quality variables were DO, temperature and TP, with the exception of TSS, which showed significant differences in Site 1 and Site 2 sites. All of the Site 1 sites are closest to the headwaters of their watershed, in addition, the purpose of these sites are to act as least-impacted agricultural sites. Therefore, it is surprising to see significant differences

between the Site 1 sites for many water quality variables. For example, Lynde Creek site 1 had the highest average concentration of TP among all of the study sites at 27.19 µg L<sup>-1</sup>. This site is in an area of high agricultural land-use, which could be influencing the TP concentration to be higher. However, Oshawa Creek site 1 has roughly the same agricultural land-use, but has just under half the TP concentration at 12.36 µg L<sup>-1</sup> as Lynde Creek site 1. Phosphorus can bind itself to the soil and sediment found in agricultural land-use and enter streams through soil erosion (Tye et al., 2016). In addition, types of fertilizer such as manure have been linked to increased phosphorus loading into streams (Hoorman et al., 2008; Cooke et al., 2011). Therefore, this increase of nutrients in Lynde Creek site 1 could be due to differences in fertilizer applications or agricultural type and intensity among Site 1 locations.

Further spatial analysis with distance from headwaters (i.e., longitudinal) revealed that there were no significant correlations with nutrients or TSS. However, chloride, pH and temperature were found to correlate with distance from headwaters. Observing correlations for chloride and temperature was expected since these parameters typically increase while moving down a streams continuum, yet observing no correlations for nutrients and TSS was not. To expand on this, I examined how latitude (i.e., % Developed land-use) and longitudinal (i.e., distance from headwaters) gradients may influence water quality (Figure 3.3). Many sites were associated with percent developed land-use compared to distance from headwaters. In addition, many of the water quality variables within the RDA clumped towards the center indicating that both distance from headwaters and percent developed land-use were influencing these variables. This indicates that lateral and longitudinal spatial patterns together are possibly influencing water quality equally in these watersheds. Theoretically, as accumulated nutrients and sediment flow downstream from the headwaters their concentrations should increase as a function of catchment

size and distance from headwaters (Vannote et al., 1980). Therefore, observing no correlations among longitudinal spatial factors with nutrients and sediments could indicate the important influence of spatial heterogeneity and associated land-use among the study sites could be plausible.

Temporal patterns in water quality parameters was revealed to be highly variable among the watersheds and the study sites. ANOVA results for watersheds and sites over time revealed many statistically significant differences. Observing seasonal differences was expected since I observed in late spring and summer. In spring months, the spring melt and any rain events wash off any particulate matter into receiving water, therefore increasing particulate concentrations such as chloride and TSS (Gallagher et al., 2011). In addition, summer months have increased water temperatures due to extended higher ambient temperatures, and these increases in water temperature can be influenced by flow regimes that can influence water temperature in low flow conditions (Potapova & Charles, 2007; Maret et al., 2010). Increases in water temperatures have been found to influence various water quality parameters (e.g., dissolved oxygen) and primary productivity (Van meter et al., 2011; Braccia et al., 2013). I observed similar seasonal patterns with water quality variables and sites for all sampling months. More developed sites (e.g., Site 4 sites) were found to associate with variables like, elevated chloride, pH, and temperature consistently, in addition, these sites seemed to cluster together most months. Near head-water sites over time were highly variable among each other, indicating that watersheds in the same geological area may have headwaters that are influenced by various factors, such as land-use and water quality.

### **3.4.2** *Spatial and Temporal patterns in Algal Community Composition*

Chl *a* and AFDM was observed to be highly variable among all study sites and sampling months. The variability in algal biomass could be due to many factors, such as nutrients, light availability, and grazers (Bernhardt & Likens, 2004; Black et al., 2011; Choi et al., 2015; Hamelin et al., 2015; Hoyle et al., 2015). According to Black et al. (2011), Chl *a* and AFDM have a dynamic nature, especially when attempting to relate with nutrient data. Although macroinvertebrates were not investigated in this study they can impact algal biomass found at a study site. Braccia et al. (2013) found that total macroinvertebrate biomass was driven by increases in algal biomass, therefore increased algal growth on artificial substrate could act as a food source for macroinvertebrates.

Algal community composition found among all study sites varied for each month sampled. This variation in community structure was in part due to seasonal changes in light availability, temperature, and successional age of each community, but also due to changes in water quality from month to month. Benthic algal communities are sensitive to changes in water quality, especially nutrients and contaminants (Lavoie et al., 2004, 2011; Pan et al., 2004; Potapova & Charles, 2007). In addition, benthic algae also have high temporal variation due to their quick turnover time and cosmopolitan nature (Duong et al., 2007; Honeyfield & Maloney, 2014). The variation of all these factors make it imperative to observe algal community composition over time in order to properly assess stream conditions.

As expected, correspondence analysis (CA) for each month revealed that community composition changed over time. In addition, the CA analyses did not reveal any spatial patterns

for sites over time, suggesting that community structure may be dominated by generalist or cosmopolitan taxa that are not sensitive to changing water quality conditions over time. When observing algal community structure with water quality parameters some spatial patterns could be observed with community structure over the sampling period. For instance, the most developed sites tended to group together (e.g., Oshawa Creek Site 3, Lynde Creek Site 3 and 4) and were associated with similar water quality variables such as, chloride and temperature. In addition, more pollution tolerant taxa like *Gomphonema* and *Cymbella* were observed at these sites for all the sampling months. Among the taxa *Gomphonema* and *Cymbella*, I observed species like, *Gomphonema parvulum* and *Cymbella tumida*. Past studies have regarded *Gomphonema parvulum* as a pollution tolerant species capable of thriving in habitats enriched with nutrients, and heavy metal pollution (Medley & Clements, 1998; Duong et al., 2007; Bere & Mangadze, 2014).

Interestingly, for the month of July and August I observed a surge of taxa correlating with TP and TN, respectively. Algal taxa that ordinated closest to elevated nutrients included, *Melosira varians*, *Frustulia vulgaris*, and *Synedra ulna* and have been previously documented within nutrient-rich streams (Biggs, 1995; Biggs & Kilroy, 2000; Moresco & Rodrigues, 2014). This shift in algal community composition driven by nutrients could be due to various factors. It is possible that nutrients became more available while previously being limited, therefore taxa that thrive in nutrient-rich conditions started to progress and show more association with the nutrients over time. A shift in benthic algal communities along a TN:TP ratio could indicate that phosphorus is a limiting nutrient within these systems (Stelzer & Lamberti, 2001).

#### 3.5 Conclusions

My expectation at the outset of this study was that both longitudinal and lateral forces in lotic drainage networks would play a role in controlling water quality and algal community variation across space and time. Although water quality and algal community structure was highly variable on both spatial and temporal scales, study results seem to suggest that both lateral (i.e., local, land-use impacts) and longitudinal (i.e., cumulative) factors are controlling water quality and algal community variation over time. This result coincides with the river wave concept created by Humphries et al. (2014) that suggests that local inputs and cumulative inputs are responsible for changes in water quality and aquatic communities. In contrast, Wu et al. (2014) found that spatial scales (e.g., elevation, longitude and latitude) had a large influence on algal community structure compared to environmental variables. However, their study compared large river catchments, approximately 3,000 km<sup>2</sup> in size, which is roughly 30 times the size of the catchments in this study. Since the watersheds in this study were highly similar with respect to natural geology, it is likely that local land-use activity surrounding each site is driving variation in water quality. Pan et al. (2004) suggests that systematic changes in algal assemblages (i.e., replacement of one species to another) may reflect the water conditions over time. This highlights the importance of monitoring a watershed over the ice-free season to properly infer the condition of a stream. In addition, Pan et al. (2004) also suggests that infrequent summer sampling could underestimate how land-use effects water quality, especially if seasonal patterns in water quality are distinct. Overall, I observed several differences across sites at both lateral and longitudinal scales, and found that sites within and across watersheds may be influenced by both local spatial-factors and large-scale, cumulative spatial-factors.

#### 4.0 General Conclusion

With rapid development proposed for the Durham region, it is necessary to obtain a baseline of information in order to compare future impacts due to this anticipated land-use change. The significance of this thesis research was to examine the longitudinal and lateral land-use effects that could possibly impact major Durham Region watersheds now and in the future. Durham region will soon undergo significant massive urban development within the next few years and observing how these land-use effects are currently affecting these watersheds could be instrumental in protecting them. Ultimately by gaining new knowledge about the local and cumulative effects of rural-urban land-use gradients on water quality and benthic algal communities, we can enhance our understanding and inform mitigative actions to improve water quality in these watersheds.

In the second chapter of this thesis, I compared how specific land-use types could influence water quality and algal community composition. I observed that when comparing all of my study sites amongst the various land-use types that urban development had the most influence on both water quality and algal communities compared to agricultural land-use. Even though agricultural landscapes are known to cause impairments to lotic ecosystems, water quality stressors such as chloride and TSS were found to increase with increasing urbanization. Chl *a* was shown to have a negative relationship with TSS, which could indicate that urban development and suspended solids are decreasing algal growth through limiting light penetration. Many algal communities were found to be associated with multiple water quality and land-use factors, however, shifts in taxa to pollution tolerant genera were observed in the most developed sites. Overall, the most important information from this chapter was that urban development had

the most potential for effecting both water quality and the biological integrity of these watersheds.

The third chapter of this thesis examined the spatial and temporal effects of these landuse gradients on water quality and benthic algae. I observed high variability amongst all of the
sites regarding water quality and algal communities over time. My results also seem to show that
longitudinal or cumulative forces in these watersheds are not having a controlling influence on
water quality and that both cumulative forces and local land-use activities could be influencing
these watersheds. Overall my results drive home the notion that temporal sampling, at least with
respect to understanding seasonal water quality and algal communities, offers highly valuable
information that could be missed if only spatial scales are considered.

In conclusion, understanding how these land-use gradients are functioning could help provide information for future development plans. The results of this study suggest that land-use and seasonality can influence water quality and algal communities. Land-use types such as, developed land-use and road density can severely affect the natural characteristics of freshwater streams. If possible, alternatives and best management practises (e.g., permeable roadways, stormwater management ponds, and green roofs) should be implemented to reduce these effects in newly developed areas. In addition, future monitoring plans should implement more spatial parameters in order to obtain a more robust understanding of these systems. With rapid expansion inevitable within these watersheds over the next few years, the potential inputs of nutrients and contaminants that can occur through rapid development should be considered and further examined.

### **4.1 Study Limitations**

The research conducted in this thesis had a few limitations that impacted aspects of my thesis project. Although artificial substrates have several advantages, particularly when comparing algal communities across sites with different environmental histories, there are several limitations as well. For example, artificial substrates are subject to vandalism, can be biased towards colonizer species, and potential grazing by macroinvertebrates (Biggs & Kilroy, 2000). In this study, I lost several ceramic tiles that were not found following the incubation period and therefore, I inevitably lost valuable data. However, even though these limitations can be problematic in ecological studies, artificial substrates are still best utilized when the purpose of your study is to detect changes in water quality and community composition over time.

Another limitation is that the number of study sites is small (i.e., 16 study sites in total representing 4 creeks). Study site and creek number was largely constrained by my ability to run 4 independent incubation periods for algal growth assays as well as process all of the algal and water quality samples during the time allotted for my Master's thesis. In order to obtain a more comprehensive examination of rural-urban land-use gradients, more tributaries with matched distance-from-headwater sites would be ideal. However, this would require that all sites still share similar physiography, geology and climate regimes, which could be challenging, but possible in Southern Ontario. Interestingly, in periphyton monitoring manuals by Biggs & Kilroy (2000) and Barbour et al. (1999), it is suggested to have at least two reference sites with a minimum of two impact sites, so my study design at least exceeded this criteria. I also have an important temporal component that is typically not included in these types of studies, and as far as I know, there are no other comparable studies that deployed artificial substrates as frequently as I did in this study.

#### 4.2 Future Research

Although this thesis was able to observe several interesting relationships between water quality and algal communities across rural-urban land-use gradients, there remain some gaps that could be addressed with future research. After examining multiple land-use types, it would be important to examine urbanized areas more in depth to find specific sources of urban influence on water quality and algal community structure. In addition, it is also necessary to acquire more information on these watersheds. Through testing more sites through these watersheds, it would be possible to find specific contamination hotspots, while obtaining more baseline information on these watersheds. Another important avenue would be to study more watersheds to further establish how longitudinal and lateral land-use gradients might influence water quality and algal communities.

Furthermore, water quality variables were observed to be variable amongst all of the sites and watersheds over the sampling period. Groundwater recharge is a factor that is often overlooked when researching freshwater tributaries. The tributaries within these four watersheds share two major physiological landmarks, ORM and Lake Iroquois Beach. These landmarks cut through each of these watersheds, and can add substantial inputs of groundwater into tributaries from groundwater discharge areas (Central Lake Ontario Conservation Authority, 2012; 2013a; 2013b). If these groundwater discharge areas are influencing water quality near my sites it could explain why only select water quality variables were observed to show cumulative increases. Therefore, further investigation using groundwater recharge data could help explain some of the high variability among the study sites.

Future studies on benthic communities could focus on a tandem approach using both benthic algae and invertebrates. Benthic invertebrates similar to benthic algae have been used in

many studies to help assess the conditions of stream environments (Jowett, 2003; Urban et al., 2006; Bazinet et al., 2010; Wallace et al., 2013). Using multiple types of aquatic organisms could help establish a more in depth analysis of how these land-use types are affecting these systems. Another important avenue of research would be to investigate algal growth in relation to light intensity. Light is important for algal growth, in addition to other factors, however, light is a variable that many studies do not focus on when studying algae. Other possibilities for future studies using algae could involve researching fatty acid composition. Algae are a high quality source of food for higher trophic organisms, they contain high levels of essential nutrients and polyunsaturated fatty acids (Guo et al., 2015). It would be important to monitor if land-use effects have the potential to change the fatty acid composition within the algal communities, which could directly influence the food web.

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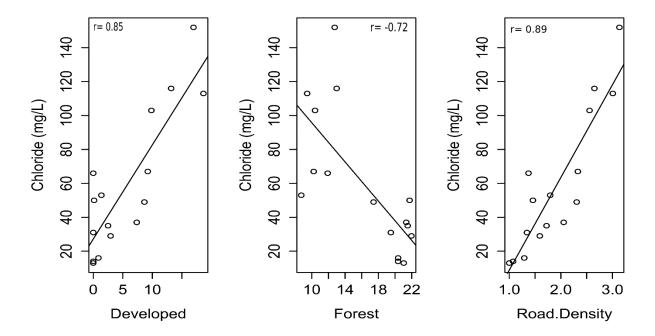
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# **Appendices**

# **Supplementary Information: Chapter 2**



**Figure A1.** Linear models of chloride against Developed, Forest, and Road Density land-use for catchment scales.

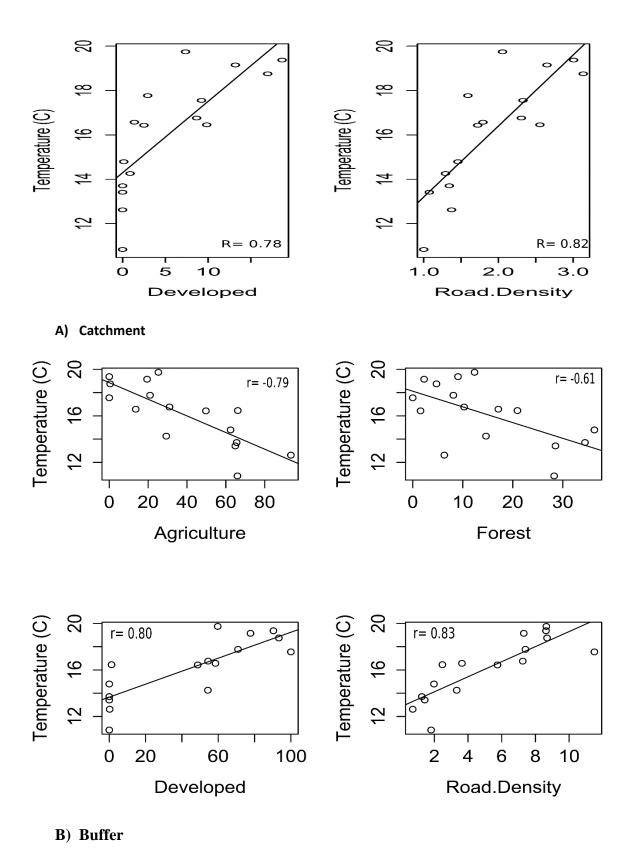
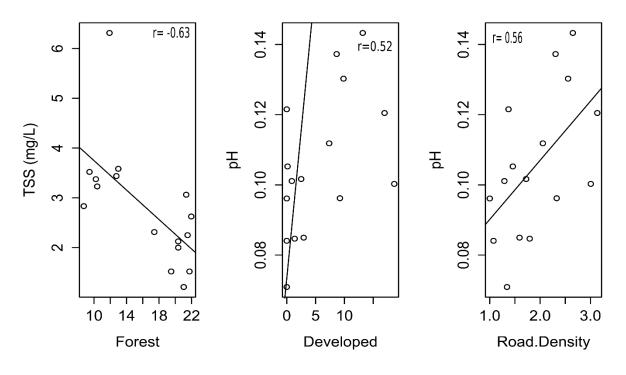
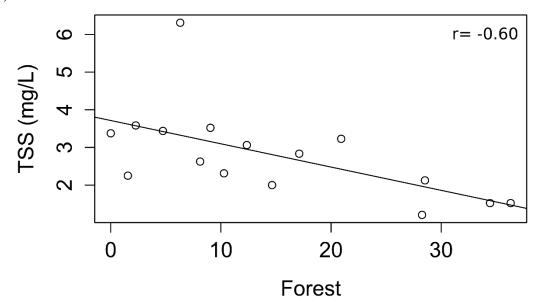


Figure A2. Linear models of temperature for catchment (A) and buffer (B) scales.



### A) Catchment



# B) Buffer

Figure A3. Linear models of TSS and pH for catchment (A) and buffer (B) scales.

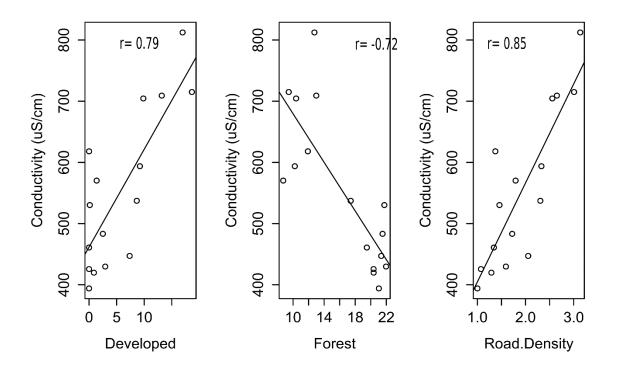
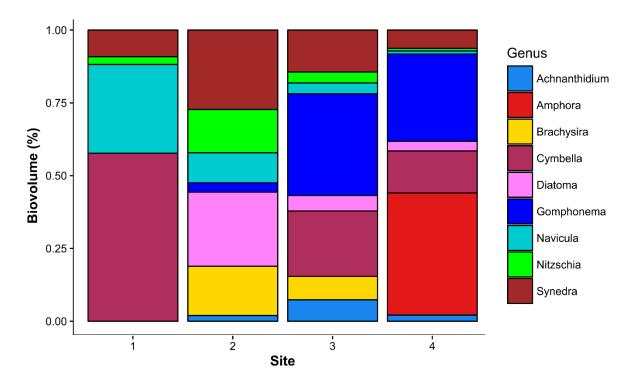
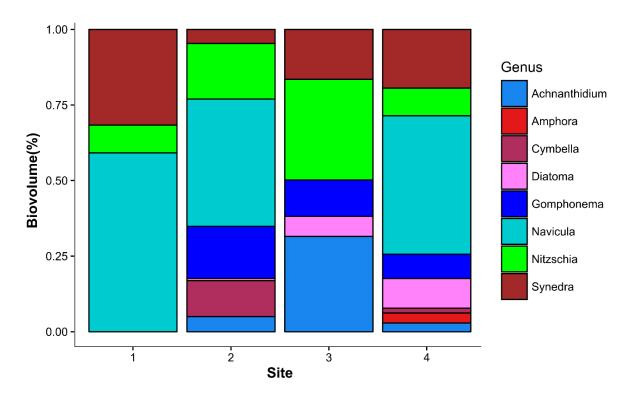


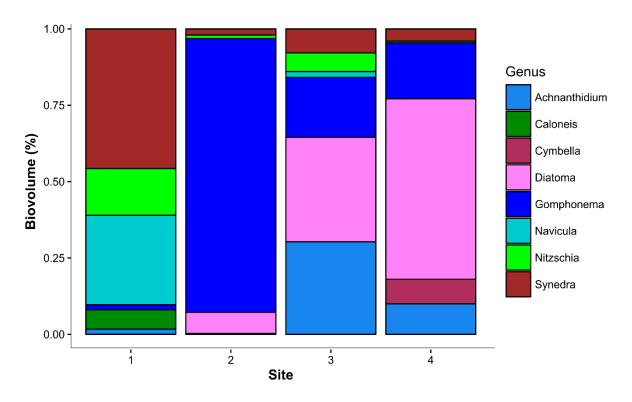
Figure A4. Linear models of Conductivity for catchment scaling



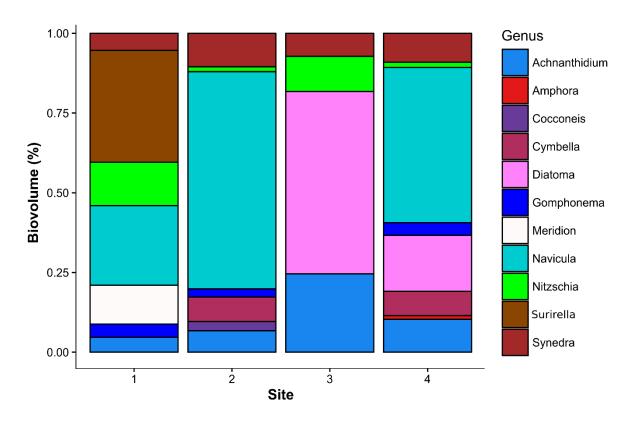
**Figure A5.** Relative biovolume plot for study sites located in the Lynde Creek Watershed in May, includes rare genus.



**Figure A6.** Relative biovolume plot for study sites located in the Oshawa Creek Watershed in May, includes rare genus.



**Figure A7.** Relative biovolume plot for study sites located in the Bowmanville Creek Watershed in May, includes rare genus.



**Figure A8.** Relative biovolume plot for study sites located in the Soper Creek Watershed in May, includes rare genus.

 Table A1. Site locations and physical features.

		Catchment			Buffer		
Watershed	Site ID	% Developed Cover	% Agricultural Cover	% Forest Cover	% Developed Cover	% Agricultural Cover	% Forest Cover
Lynde Cr.	L1	0.001	0.71	0.21	0.00	0.62	0.36
Lynde Cr.	L2	0.09	0.72	0.10	0.012	0.66	0.21
Lynde Cr.	L3	0.17	0.65	0.12	0.93	0.004	0.05
Lynde Cr.	L4	0.13	0.67	0.12	0.78	0.19	0.02
Oshawa Cr.	O1	0.00	0.86	0.11	0.002	0.93	0.06
Oshawa Cr.	O2	0.01	0.87	0.08	0.58	0.14	0.17
Oshawa Cr.	O3	0.09	0.77	0.10	100	0.00	0.00
Oshawa Cr.	O4	0.19	0.68	0.09	0.90	0.00	0.09
Bowmanville Cr.	B1	0.00	0.65	0.20	0.00	0.65	0.29
Bowmanville Cr.	B2	0.009	0.66	0.20	0.54	0.29	0.15
Bowmanville Cr.	B3	0.03	0.65	0.21	0.71	0.21	0.08
Bowmanville cr.	B4	0.07	0.62	0.21	0.59	0.25	0.12
Soper Cr.	<b>S</b> 1	0.00	0.65	0.21	0.00	0.66	0.28
Soper Cr.	S2	0.00	0.75	0.19	0.00	0.66	0.34
Soper Cr.	<b>S</b> 3	0.03	0.73	0.21	0.48	0.49	0.02
Soper Cr.	<b>S</b> 4	0.09	0.72	0.17	0.54	0.31	0.10

# **Supplementary Information: Chapter 3**

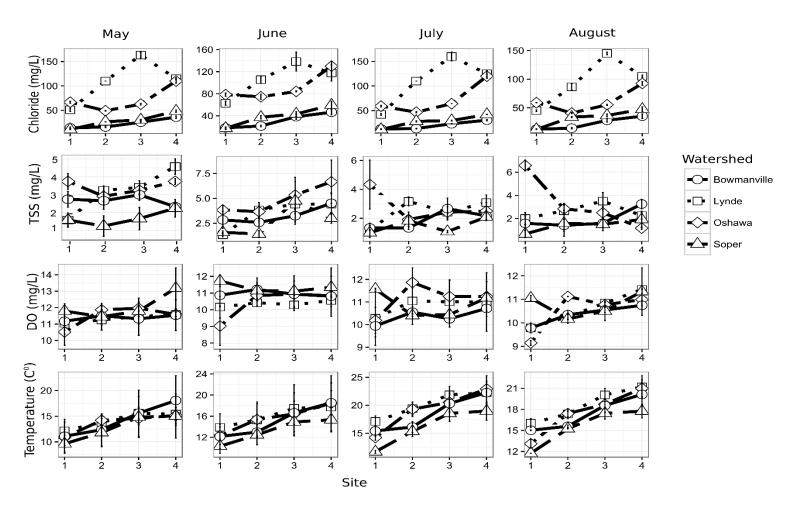


Figure B1. Water quality variables at study sites along each watershed during the entire sampling period, including standard error.

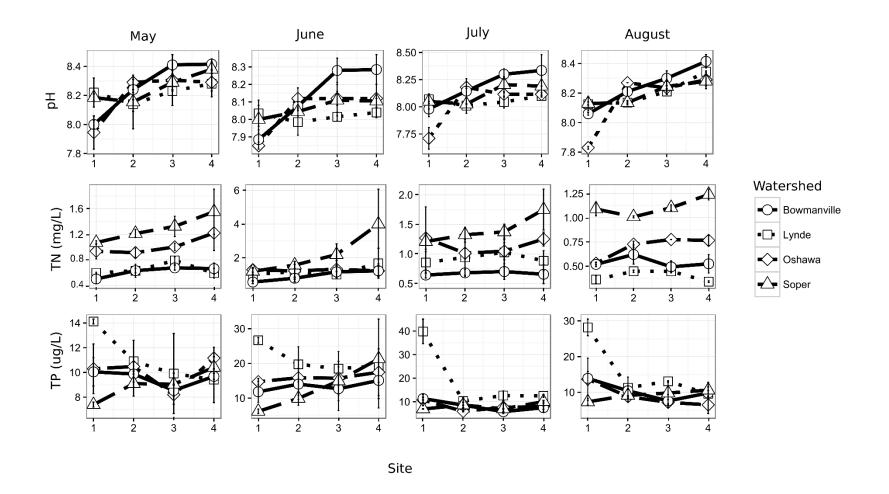
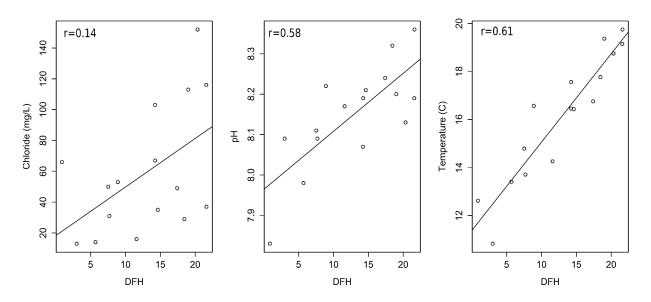
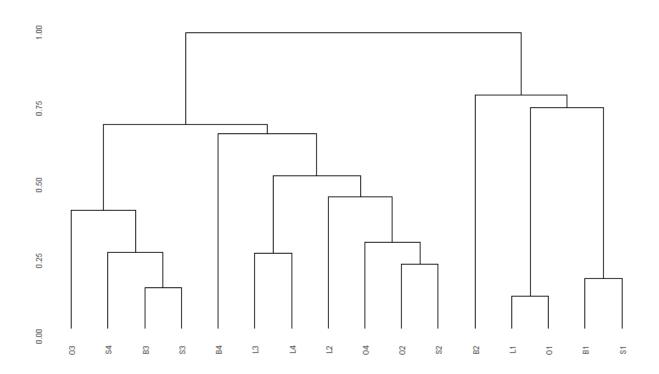


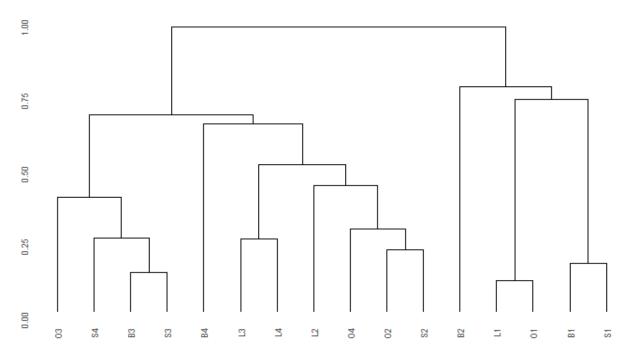
Figure B1. (Cont.)



**Figure B2.** Linear models of water quality variables against distance from headwaters (DFH) for chloride, pH, and temperature.



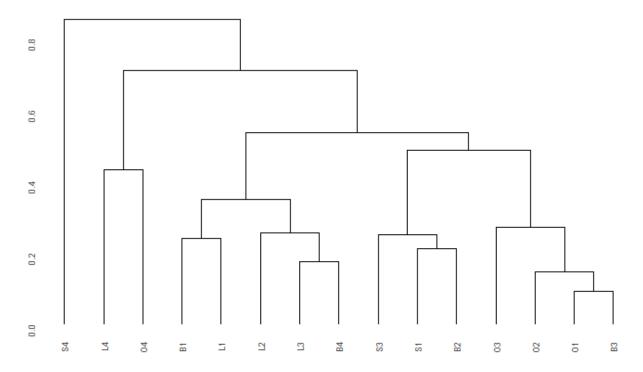
**Figure B3.** Cluster analysis performed on Bray Curtis distances for the month of May algal communities.



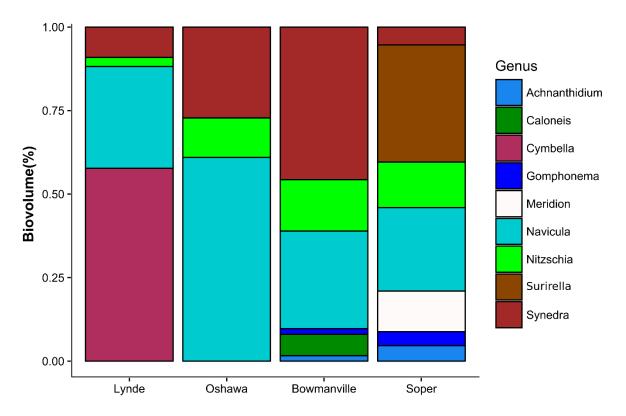
**Figure B4.** Cluster analysis performed on Bray Curtis distances for the month of June algal communities.



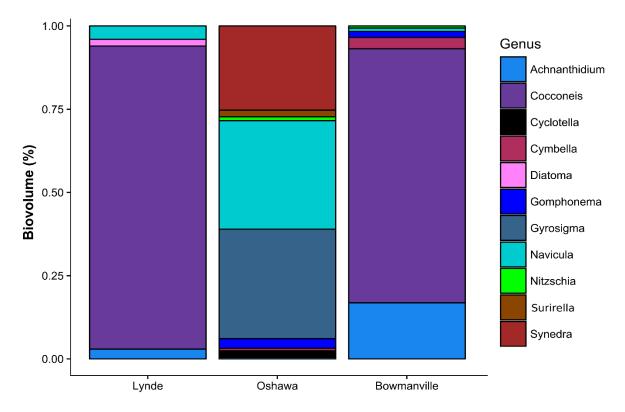
**Figure B5.** Cluster analysis performed on Bray Curtis distances for the month of July algal communities.



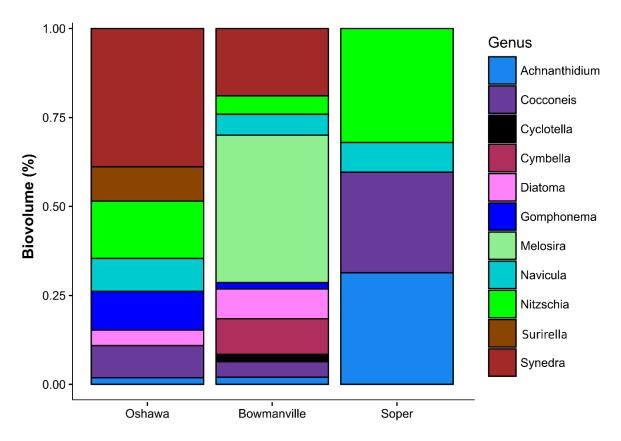
**Figure B6.** Cluster analysis performed on Bray Curtis distances for the month of August algal communities.



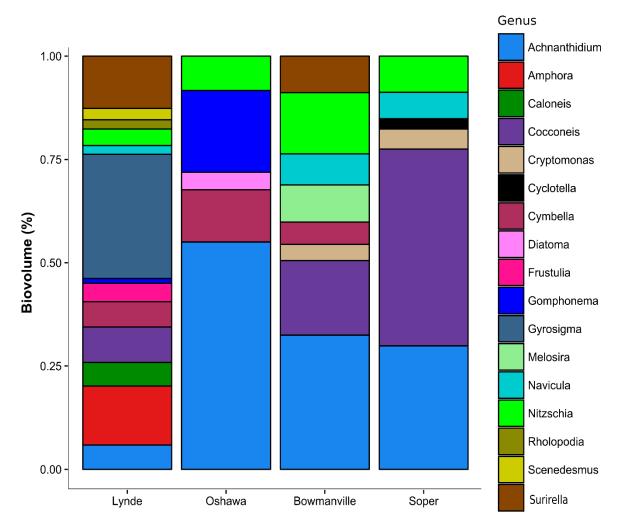
**Figure B7.** Site 1 comparison of relative biovolume of algal community composition for all watersheds for the month of May, includes rare genus.



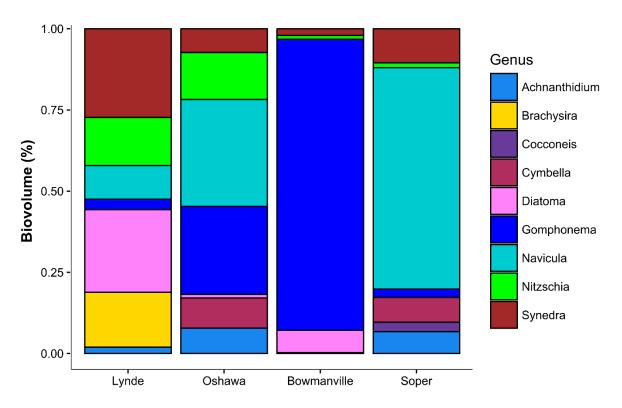
**Figure B8.** Site 1 comparison of relative biovolume of algal community composition for all watersheds for the month of June, includes rare genus.



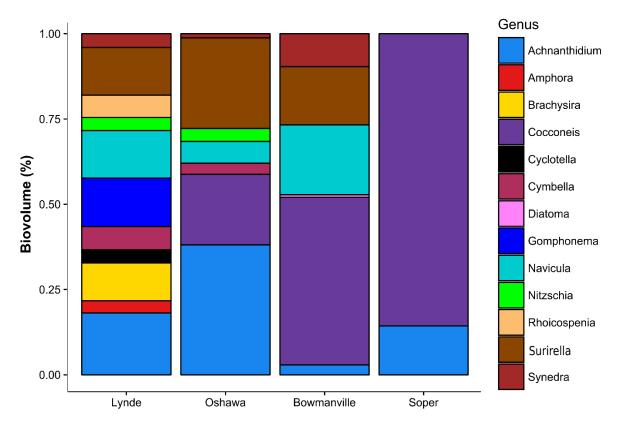
**Figure B9.** Site 1 comparison of relative biovolume of algal community composition for all watersheds for the month of July, includes rare genus.



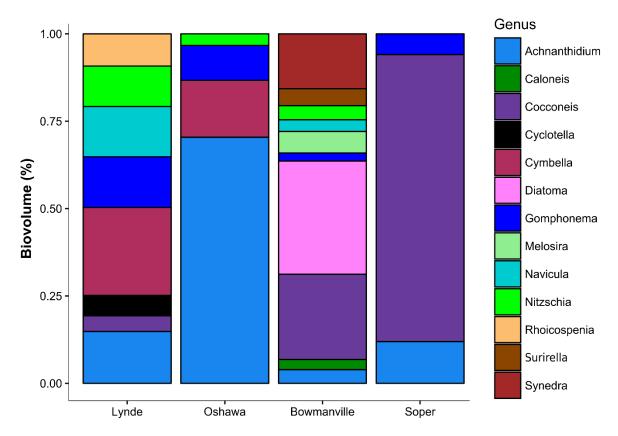
**Figure B10.** Site 1 comparison of relative biovolume of algal community composition for all watersheds for the month of August, includes rare genus.



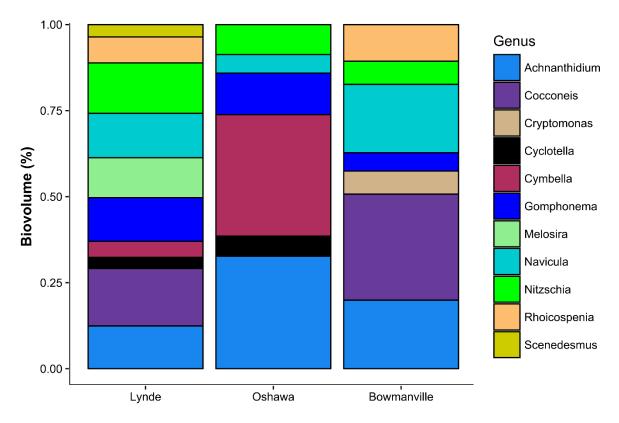
**Figure B11.** Site 2 comparison of relative biovolume of algal community composition for all watersheds for the month of May, includes rare genus.



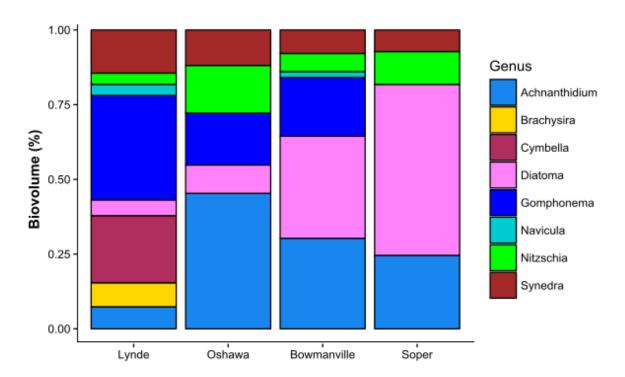
**Figure B12.** Site 2 comparison of relative biovolume of algal community composition for all watersheds for the month of June, includes rare genus.



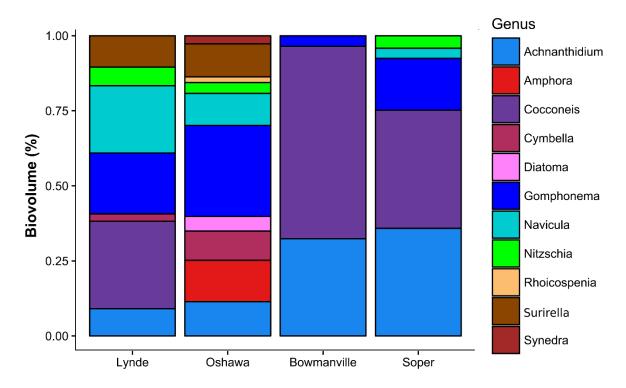
**Figure B13.** Site 2 comparison of relative biovolume of algal community composition for all watersheds for the month of July, includes rare genus.



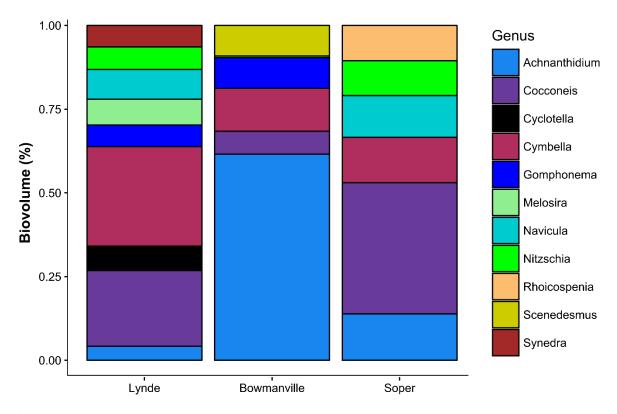
**Figure B14.** Site 2 comparison of relative biovolume of algal community composition for all watersheds for the month of August, includes rare genus.



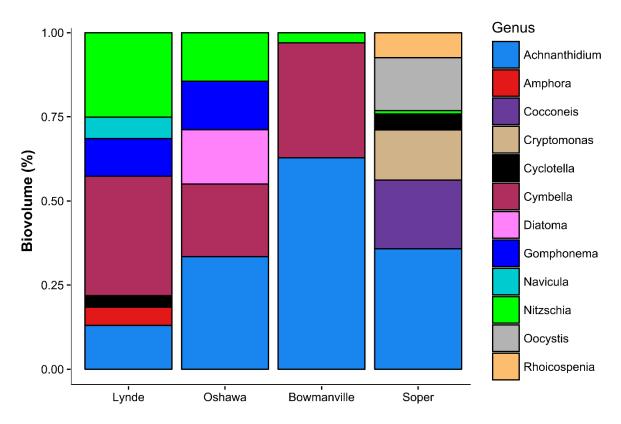
**Figure B15.** Site 3 comparison of relative biovolume of algal community composition for all watersheds for the month of May, includes rare genus.



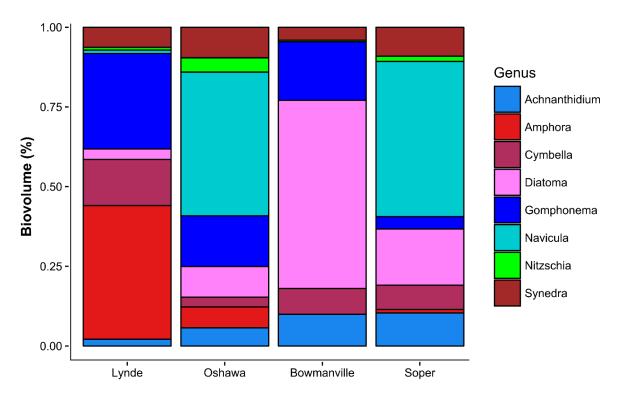
**Figure B16.** Site 3 comparison of relative biovolume of algal community composition for all watersheds for the month of June, includes rare genus.



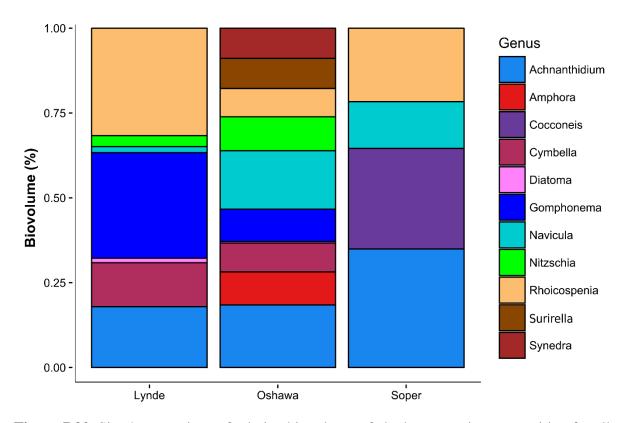
**Figure B17.** Site 3 comparison of relative biovolume of algal community composition for all watersheds for the month of July, includes rare genus.



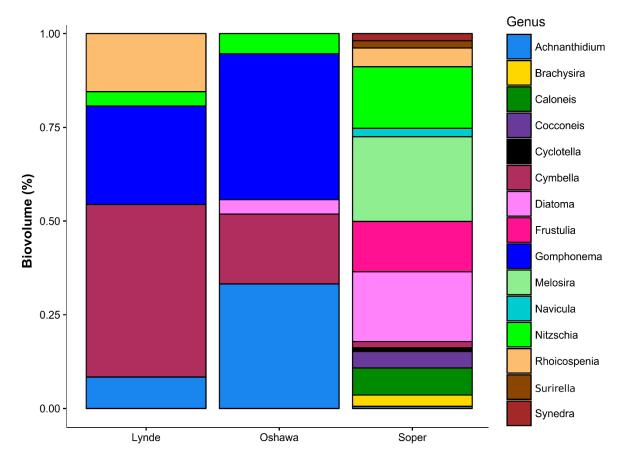
**Figure B18**. Site 3 comparison of relative biovolume of algal community composition for all watersheds for the month of August, includes rare genus.



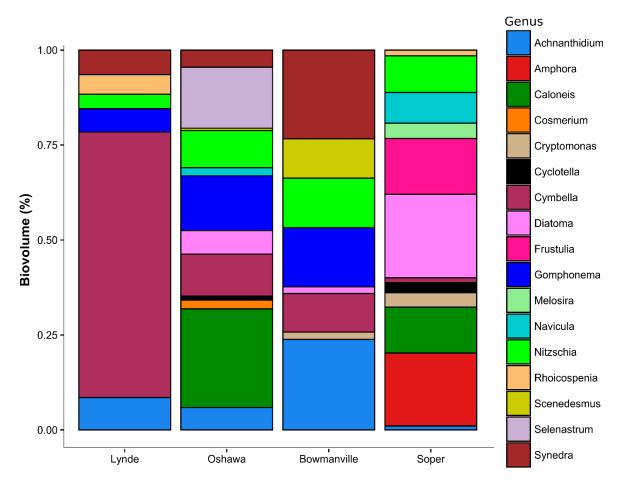
**Figure B19.** Site 4 comparison of relative biovolume of algal community composition for all watersheds for the month of May, includes rare genus.



**Figure B20.** Site 4 comparison of relative biovolume of algal community composition for all watersheds for the month of June, includes rare genus.



**Figure B21.** Site 4 comparison of relative biovolume of algal community composition for all watersheds for the month of July, includes rare genus.



**Figure B22.** Site 4 comparison of relative biovolume of algal community composition for all watersheds for the month of August, includes rare genus.

**Table B1.** One-way ANOVA and Kruskal-Wallis results for TP and Chlorophyll a at **Lynde Creek.** 

Term	F Value /χ²	P Value
TP		
Site	8.76	P<0.001
Month	2.10	0.12
Chl a		
Site	1.84	0.61
Month	12.5	0.005

**Table B2.** One-way ANOVA and Kruskal-Wallis results for TSS, Chloride, Chlorophyll a, and AFDM at **Oshawa Creek.** 

Term	F Value/χ²	P Value
maa		
TSS		
Site	2.28	0.085
Month	3.36	0.022
Chloride		
Site	85.05	P<0.001
Month	5.96	P<0.001
Chl a		
Site	23.9	P<0.001
Month	4.48	0.21
<b>AFDM</b>		
Site	7.46	0.060
Month	9.22	0.030

**Table B3.** Kruskal-Wallis results for Chlorophyll a and AFDM at **Bowmanville Creek.** 

Term	$\chi^2$	P Value
Chl a		
Site	4.75	0.19
Month	20.9	P<0.001
<b>AFDM</b>		
Site	11.5	0.009
Month	16.2	P<0.001

**Table B4.** Kruskal-Wallis results for Chlorophyll a and AFDM at **Soper Creek.** 

Term	$\chi^2$	P Value
Chl a		
Site	4.74	0.19
Month	23.2	P<0.001
<b>AFDM</b>		
Site	11.5	0.009
Month	13.1	0.004

**Table B5.** Pearson correlation analyses on algal genus and water quality for the month of May. Bolded numbers indicate significant correlations (P<0.05).

	Achn	Amp	Bra	Cocc	Cycl	Cymb	Diat	Gomp	Navi	Nitz	Rho	Suri	Syn
Chloride	-0.07	0.31	0.25	-0.70	0.17	0.56	0.12	0.61	0.29	0.79	0.45	0.62	-0.08
DO	0.50	0.11	-0.13	0.26	-0.79	-0.03	-0.18	-0.04	-0.71	-0.28	0.06	-0.26	-0.61
pН	0.64	0.18	-0.19	-0.15	-0.60	-0.09	0.06	0.11	-0.49	-0.04	0.03	-0.04	-0.48
Temp	0.39	0.35	0.03	-0.66	-0.32	0.49	0.08	0.64	-0.15	0.59	0.50	0.40	-0.45
TN	0.47	-0.09	-0.09	-0.14	-0.10	-0.20	-0.16	-0.05	-0.22	-0.17	0.37	-0.24	-0.18
TP	0.01	0.02	0.21	-0.11	0.00	0.11	0.64	0.17	-0.09	0.14	0.34	0.08	-0.18
TSS	0.08	0.55	0.03	-0.81	0.05	0.64	-0.13	0.62	0.27	0.83	0.29	0.46	-0.01

**Table B6.** Pearson correlation analyses on algal genus and water quality for the month of June. Bolded numbers indicate significant correlations (P<0.05).

	Achn	Amp	Bra	Cocc	Cycl	Cymb	Diat	Gomp	Navi	Nitz	Rho	Suri	Syn
Chloride	-0.07	0.31	0.25	-0.70	0.17	0.56	0.12	0.61	0.29	0.79	0.45	0.62	0.62
DO	0.50	0.11	-0.13	0.26	-0.79	-0.03	-0.18	-0.04	-0.71	-0.28	0.06	-0.26	-0.26
pН	0.64	0.18	-0.19	-0.15	-0.60	-0.09	0.06	0.11	-0.49	-0.04	0.03	-0.04	-0.04
Temp	0.39	0.35	0.03	-0.66	-0.32	0.49	0.08	0.64	-0.15	0.59	0.50	0.40	0.40
TN	0.47	-0.09	-0.09	-0.14	-0.10	-0.20	-0.16	-0.05	-0.22	-0.17	0.37	-0.24	-0.24
TP	0.01	0.02	0.21	-0.11	0.00	0.11	0.64	0.17	-0.09	0.14	0.34	0.08	0.08
TSS	0.08	0.55	0.03	-0.81	0.05	0.64	-0.13	0.62	0.27	0.83	0.29	0.46	0.46

**Table B7.** Pearson correlation analyses on algal genus and water quality for the month of July. Bolded numbers indicate significant correlations (P<0.05).

	Achn	Bra	Calo	Cocc	Cycl	Cymb	Diat	Frus	Gomp	Melo	Navi	Nitz	Rho	Scen	Suri	Syn
Chloride	-0.07	-0.11	-0.28	-0.09	0.36	0.44	-0.38	-0.11	0.64	-0.27	0.29	0.07	0.30	-0.22	-0.31	-0.29
DO	0.36	0.19	0.03	-0.29	0.11	0.15	-0.35	0.19	0.28	-0.45	-0.09	0.00	0.15	-0.30	-0.11	-0.50
pН	0.56	0.23	0.27	-0.08	0.01	0.12	0.01	0.23	0.08	-0.16	-0.17	-0.03	0.20	0.46	0.09	-0.25
Temp	0.36	0.07	-0.08	-0.18	0.25	0.52	-0.24	0.07	0.67	-0.23	0.07	0.06	0.39	0.19	-0.20	-0.28
TN	-0.39	0.63	0.18	0.16	0.27	-0.22	-0.42	0.63	-0.17	-0.43	-0.23	0.02	0.24	-0.36	-0.03	-0.42
TP	-0.61	0.12	0.04	0.05	0.55	0.41	0.14	0.12	0.23	0.36	0.52	0.50	0.40	-0.42	0.03	0.37
TSS	-0.21	-0.07	-0.17	-0.19	-0.11	-0.06	-0.23	-0.07	0.02	-0.21	-0.12	-0.15	-0.05	-0.01	-0.01	-0.10

**Table B8.** Pearson correlation analyses for algal genus and water quality parameters for the month of August. Bolded numbers indicate significant correlations (P < 0.05).

	Achn	Amp	Calo	Cocc	Cryp	Cycl	Cymb	Diat	Frus	Gomp	Melo	Navi	Nitz	Rho	Scen	Suri	Syn
Chloride	-0.42	0.20	0.15	-0.50	-0.38	-0.05	0.61	0.19	-0.05	0.76	0.06	-0.07	0.29	0.05	-0.01	-0.31	0.42
DO	-0.56	0.01	0.38	-0.053	0.15	0.21	0.45	0.27	0.33	0.22	0.00	0.25	0.16	0.05	-0.15	-0.35	0.44
pН	-0.42	-0.02	0.23	-0.19	0.16	0.07	0.41	0.20	0.15	0.18	-0.10	0.01	0.24	0.08	0.24	-0.29	0.47
Temp	-0.43	0.03	0.27	-0.65	-0.15	-0.12	0.65	0.37	0.03	0.56	-0.06	-0.14	0.38	0.03	0.22	-0.24	0.62
TN	-0.27	0.05	0.51	0.36	0.65	0.65	-0.44	0.35	0.56	-0.44	0.15	0.51	0.06	0.04	-0.34	-0.14	-0.26
TP	0.04	0.69	-0.19	0.081	-0.11	-0.22	-0.13	-0.33	-0.03	-0.25	0.03	-0.01	0.22	-0.09	0.40	0.14	-0.24
TSS	0.35	-0.01	-0.23	-0.23	-0.32	-0.29	0.039	-0.08	-0.09	0.45	-0.06	-0.21	-0.10	-0.18	0.08	-0.15	-0.09