LONG-TERM CHANGES OF HYPOLIMNETIC DISSOLVED OXYGEN AND PHYTOPLANKTON COMMUNITY COMPOSITION IN A LARGE LAKE, LAKE SIMCOE, ONTARIO AND REVISIT DISSOLVED OXYGEN PROFILE MODELS FOR LAKES IN CENTRAL ONTARIO

JIAHUA LI

A THESIS SUBMITTED TO THE FACULTY OF GRADUATE STUDIES IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF

MASTER OF SCIENCE

GRADUATE PROGRAM IN BIOLOGY
YORK UNIVERSITY
TORONTO, ONTARIO

JUNE 2016

Abstract

Long-term changes in volume-weighted hypolimnetic dissolved oxygen (VWHDO) and phytoplankton community composition were examined as important water quality indicator in Lake Simcoe, a large freshwater lake with impacts by various drivers such as dreissinid mussel (DM) invasion, reduction in phosphorus (P) loading and climatic change and oscillations. VWHDO dynamics were examined by several multiple linear regressions which revealed that $VWHDO_{min}$ was positively correlated with the invasion of DM and negatively correlated with hypolimnetic water temperature; VWHDO depletion rate (DR) was positively correlated with ammonia and total phosphorus (TP) annual loading. Hence, longer period of thermal stratification (L) may have offset improvements in $VWHDO_{min}$ generated by P controls and invasive species. Redundancy analysis and variation partitioning revealed that DM acted on phytoplankton composition indirectly by changing the environmental conditions because environmental variables and DM individually explained little variance in phytoplankton composition but water chemistry variables and DM collectively explained a large portion of the variation.

A series of models predicting the uppermost plane of the DO-depleted zone (Zox), spring dissolved oxygen (DOi) profile and end-of-summer dissolved oxygen (DOf) profile built by Molot et al. (1992) were re-analyzed, re-calibrated with more recent data ("A" lakes during 1990-2013), and validated ("B" lakes during 1990-2013). With the current brownification trend (increased DOC), we expected to see increased DOf at all depths; however, increased reduced N species (ammonia) in the hypolimnion would offset the brownification effect.

Acknowledgments

First and foremost I am extremely grateful to Ontario Ministry of Environment for the funding. I must thank my supervisor, Lewis Molot, who gave me this opportunity and his knowledge, intellect, support and encouragement made this work possible during my time at York University.

Secondly I must thank my supervisory committee member Sapna Sharma, whose statistical expertise provided me insights in various statistical methods. I appreciate that you invited me to come to your lab meeting every week discussing recent ecological literatures.

Thank you Norman Yan for kindly offering your lab space for an entire year. And thank you Roberto Quinlan for allowing me sit in your undergrad Freshwater Biology course for the whole semester.

I will never forget the time I spend with my labmates who provided expertise in various aspects. Thank you Mark Verschoor for your knowledge in filed work, water chemistry and phytoplankton biology and Christopher Powe for your expertise in algae culturing. This work was significantly improved by discussions we had. I have to also thank Noreen Kelly for her long email introducing R to me.

I have to thank all my friends at York University. Jonathan Ruppert, Miranda Chen, Thomas Van Zuiden, Guangkun Zhang, Samantha Stefanooff, Sara Masood, Katrina Gaibisels, Brock Harpur, Lisa Rosenberger, Tarn Parmar and many more. This work would not have been possible without your constructive advice and cheerful support.

Special thanks have to go to Zening Bu for her efficient figure editing and creative graphic design.

Last but not the least, I must thank all of my family who supported me during this time. Mowen Li and Bensi Wu, thank you for being patient and not giving up on me when times were tough.

Table of Contents

List of Tables

List of Figures

GENERAL INTRODUCTION

Lake Simcoe and multiple stressors

Lake Simcoe, the largest inland lake in southern Ontario outside the Great Lakes, is an invaluable natural, social, and economic resource. Lake Simcoe has six drinking water treatment plants (WTPs) that provide drinking water several communities in the watershed. It also assimilates wastewater from 14 municipal water pollution control plants (WPCPs). In addition, the agriculture sector is worth more than $500 million annually including crops and livestock (LSEMS 2008; Winter et al. 2007). Recreational activities including cottaging, boating and sport fishing, especially cold water species like lake trout, lake whitefish and lake herring, generate over $200 million per year industry accounting for 15% (1 million angler hours per year) of all angling effort in Ontario (Eimers et al., 2005; LSEMS, 2008). However, Lake Simcoe and its watershed have experienced multiple pressures from increasing human activities over the past ~200 years, including logging, damming, canal construction, agriculture, urban development, species invasion and recently climate change (Hawryshyn et al. 2012). Multiple stressors can interact synergically or antagonistically thus creating challenges to foresee the outcome (Palmer et al. 2012).

Nutrient enrichment

Paleolimnological inferences suggest a hypolimnetic dissolved oxygen concentration above 7 mg/L was the norm in Lake Simcoe before disturbance (Rode 2009). However, excess phosphorus (P) from point sources and non-point sources promoted excessive algae and macrophytes growth, the decomposition of which during the growing season depleted hypolimnetic dissolved oxygen to the point that hypoxia led to end of summer cold water fish recruitment failure (Evans et al. 1996). Lake trout declined in the 1960s; lake whitefish declined in the 1970s and lake herring declined in the 1980s (Evans et al. 1996; Evans and Waring 1987).

A quantitative relationship between minimum volume-weighted hypolimnetic dissolved oxygen (MVWHDO) and phosphorus loading was investigated by Nicholls (1997). Based on their predictive model, a concentration of 5.0 mg/L hypolimnetic dissolved oxygen was set as an achievable target for Lake Simcoe maintaining a healthy cold water fish habitat (Winter et al. 2007). To reach the target, a reduction of total P (TP) loading to 75 tonnes/year was proposed (TP loading ranged from 85-157 tonnes/year between 1990-1997 and 55-120 tonnes/year between 1998-2009; Rode 2009, Palmer et al., 2013). Recent metabolic studies of coldwater fish indicate that a concentration of 7.0 mg/L is ideal for juvenile lake trout (*Salvelinus namaycush*) (Evans 2007). To meet deep water dissolved oxygen above this level, TP loading in Lake Simcoe has to be reduced to 44 tonnes/year.

The Ontario government released the Lake Simcoe Protection Plan (LSPP) on June 2nd, 2009. It outlines a strategy of protection and restoration of Lake Simcoe and its watershed, and aims to sustain the cold water fish community by maintaining the VWHDO level above 7 mg/L at the end of summer when it is typically at its lowest. Extensive efforts to reduce anthropogenic TP loading into the lake include restrictions on P output from sewage wastewater treatment plants, prohibition of new point source discharges and P reduction stewardship projects (Winter et al., 2002). However, TP loading did not decrease as much as expected because of some complex interactions including the following. First, tributary loads contribute significant amount of P loading (47.4-80.6%) which increased with annual flow volume and precipitation in recent years (North et al., 2013; Palmer et al., 2013); second, climate change may have altered P cycling making more available. For example, sediments may have served as a source of P to overlying waters during periods of extreme hypoxia ("internal loading") (Nürnberg et al., 2013a,b). The end of summer VWHDO improved but still did not reach the 7 mg/L goal. Quinlan et al (2008) points out that even if a system has undergone some degree of chemical recovery, it is not necessarily followed by biological recovery.

To sum up, P abatement is still the most important lake management strategy to restore Lake Simcoe as a self-recruiting cold water fish habitats.

Invasive species

In addition to P enrichment, Lake Simcoe has been affected by multiple species invasions (LSWEMR 2013) of fish, planktonic invertebrates, benthic invertebrates and macrophytes including the common carp (*Cyprinus carpio*, in 1896), rainbow smelt (*Osmerus mordax*, 1962), black crappie (*Pomoxis nigromaculatus*, 1987), spiny water flea (*Bythotrephes longimanus*, 1993) and round goby (*Neogobius melanostomus*, 2006).

The most notorious invasive species is the zebra mussel (*Dreissena polymorpha*) which was introduced into Lake Simcoe in the early 1990s and became well established by 1996 (Evans et al., 2011). Zebra mussels use natural rocky substrate for attachment at depths around 0-15 m and filter suspended particles from the adjacent water (Evans et al., 2011). Due to their large populations and great filtering capacity, chlorophyll *a* (a measurement of total phytoplankton biomass) and TP have decreased resulting in numerous ecological changes such as redirection of nutrient and energy to the near shore (i.e. the nearshore shunt hypothesis, Hecky et al. 2004), releasing dissolved nutrients such as silicate (Barbiero et al. 2006), restructuring benthic communities (Rennie et al. 2009) and creating a more transparent water column (Winter et al. 2011). In addition, dreissenid filters selectively feed on algae, excluding the algae that are resistant to their grazing, such as cyanobacteria, thus they may facilitate toxic cyanobacteria blooms, such as *Microcystis* spp. (Vanderploeg et al. 2001). Physical and ecological effects were investigated under the invasion of dreissinid mussels (Nalepa & Schloesser 2013; Hecky et al. 2004).

Climate change

Climate change has led to changes in lake thermal structure. A shorter ice-cover period, longer thermal stratification period with earlier onset of stratification and prolonged fall overturn were found in Lake Simcoe (Hawryshyn et al. 2012; Stainsby et al., 2011). Winder and Schindler et al. (2004) found that temporal mismatch of diatom blooms which now occur earlier and the keystone herbivore, *Daphnia*, has severe consequences for energy flow to upper trophic levels due to earlier onset of thermal stratification. The most recent report from the Intergovernmental Panel on Climate Change (IPCC, 2013) predicts that global air temperatures will rise by 0 to 4.8 ˚C by the year 2100 depending on different scenarios based on greenhouse gas emission. In Ontario, temperatures are projected to increase 3 to 6 °C while precipitation is predicted to decrease by 10-30% compare to 1971-2000 period (CCRR, 2007) although others have predicted precipitation increases (see next section). Higher air temperatures coupled with decreased precipitation rates will exacerbate current warming water temperature and change thermal structure which will have profound impacts on aquatic organisms especially cold/cool water fish (Van Zuiden et al. 2016).

Stressor interactions

Changes to aquatic ecosystems resulting from acid deposition, excess nutrients, increasing exposure to ultraviolet radiation, climate change and metal pollution are well-documented (Stoddard et al. 1999; Clair et al. 2002; Smol 2009; Sondergaard et al. 2005; Schindler 2006; MacDonald et al. 1993; Schindler et al. 1996; Schindler 2001). A number of researchers have provided predictive scenarios for future aquatic change (Schindler et al. 1990; Band et al. 1996; Magnuson et al. 1997; Walther et al. 2002; Blenckner 2008; Drake 2005; Ward and Ricciardi 2007). However, our ability to predict the outcome of ecosystem disturbance is limited (but see Schindler 2001) when numerous stressors act simultaneously.

Stressors can interact synergistically or antagonistically resulting in "uncertainties" and "ecological surprises" in ecosystems (Christensen et al., 2006, Paine et al. 1998). Schindler (2001) proposed that climate change might exacerbate accumulative impacts of other stressors. A single anthropogenic stressor might have limited effects both in magnitude and localization, while multiple stressors may have accumulative impacts that extend their scale in terms of their effects individually (Quinlan et al. 2008).

Christensen et al. (2007) argued that precipitation may increase at high latitudes as a response to climate change. P loading should increase with higher precipitation and inflow from tributaries because flow exports agricultural fertilizer and eroded sediments from the landscape to Lake Simcoe, in spite of major efforts to reduce P loading the under P reduction plan. A recent study showed that the annual TP loading is significantly associated with annual flow volume in Lake Simcoe with a trend of increasing precipitation and annual TP loading in recent years (Palmer 2013).

A system affected by multiple stressors may be more vulnerable to species invasions (Strayer, 2010). Ponto-Caspian species (i.e. dreissinid mussels, round goby etc.) are eurythermic and thus are able to take advantage of the warming and have a competitive advantage over native species (Schindler 2001). Detailed investigations of native and non-native fish interactions were conducted in Wisconsin and Ontario. Cisco population extirpations and rainbow smelt invasion in Wisconsin were predicted under climate change (Sharma et al. 2011). Range expansion of smallmouth bass (a warmwater native fish), northward range shift of walleye (a coolwater native fish) and range contraction of cisco (a coldwater native fish) were projected under climate change and species interaction in Ontario (Van Zuiden et al. 2016). The establishment of zebra mussel populations might have facilitated the rapid invasion of the round goby, a major predator of the mussel (Ricciardi and Maclsaac 2001).

Importance of end-of-summer hypolimnetic dissolved oxygen

As mentioned in the nutrient enrichment section, excess nutrient loading plays an important role in structuring DO dynamics. Low VWHDO can enhance internal P and ferrous iron loading which can promote cyanobacteria blooms (Nürnberg et al., 2013a and b; Molot et al. 2014). Reduced DO levels can negatively influence biological components of lake ecosystem, particularly cold-stenothermic species such as lake trout (*Salvelinus namaycush Walbaum*) which require cold, well oxygenated waters (Sellers et al., 1998).

In Lake Simcoe, VWHDO is recognized as an important indicator of coldwater fish habitat quality (Evens et al., 1996). The success of juvenile (the most vulnerable stage of their life cycle) coldwater fish recruitment is determined by the end of summer VWHDO. There have been extensive studies of P cycling processes in lakes and the relationship between TP and algal biomass (Taylor, 2010; Hudson et al., 2000). Several recent studies investigated the relationship between annual TP loading and end of summer VWHDO in Lake Simcoe (Nicholls 1997; Young et al., 2011). A physiological study conducted by Evans (2007) indicated that 7.0 mg/L DO is a suitable target to achieve notable improvements in cold water fish production and maintain a self-recruitment cold water habitat. To achieve 7.0 mg/L end of summer VWHDO based on the relationship proposed by Nicholls (1997), a 44 tonnes/year TP loading target was set.

Importance of phytoplankton

Phytoplankton (photosynthetic pelagic algae and cyanobacteria) are at the base of aquatic food chains and are efficient energy and oxygen producers (Winder and Sommer 2012). Changes to their biomass and species composition can affect aquatic food web structure (Muller-Navarra et al. 2000). Different phytoplankton species contain various amounts and types of polyunsaturated fatty acids (PUFA) which on one hand, are exclusively synthesized by plants and on the other hand play an important role in regulating cell membrane properties and serving as precursors for hormones in animals (Farkas et al. 1984; Bezard et al. 1994). A high correlation has been found for highly unsaturated fatty acids (HUFA) and high growth, survival, and reproductive rates in a wide variety of higher level organisms (Brett et al. 1997). For these reasons, changes in phytoplankton are considered to be an important barometer of wider ecological change. Due to their short doubling times, different nutrient requirements (Tilman 1977, 1982; Tilman et al. 1981), sizes (Chisholm 1992), sinking velocities, abilities to maintain column position (lipids, mucus sheaths, gas vacuoles, flagella) (Hutchinson 1967, chapter 20; Sommer 1985) and temperature optimums (Robarts and Zohary 1987), quick changes in phytoplankton composition are good early indicators of environmental stresses on food webs (Stoermer 1978; Rühland et al. 2008). For example, at higher temperatures, bloom-forming cyanobacteria have a competitive advantage over other phytoplankton groups (Jöhnk et al. 2008; Wagner and Adrian 2009). Similarly, stronger vertical stratification can cause shifts in phytoplankton species composition (Verburg et al. 2003; Strecker et al. 2004; Winder and Hunter 2008).

Study Objectives

In this study, our overall goal was to examine long-term patterns of VWHDO and phytoplankton community composition in relation to major environmental changes occurring in the past three decades in Lake Simcoe. These changes include eutrophication followed by P reduction beginning around 1990, invasive dreissenid mussels which became well established by 1996 and climate change post 1980. This work is presented in the next two chapters. A third chapter models oxygen profiles in small lakes in central Ontario and describes changes in hypolimnetic oxygen levels since 1977. These lakes are mostly oligotrophic and have not experienced dreissenid mussel invasions because of low Ca levels but they have experienced less ice-cover because of a warming trend.

REFERENCES

Band, L. E., Mackay, D. S., Creed, I. F., Semkin, R., & Jeffries, D. (1996). Ecosystem processes at the watershed scale: Sensitivity to potential climate change. *Limnology and Oceanography*, 41, 928-938.

Barbiero, R. P., Rockwell, D. C., Warren, G. J., & Tuchman, M. L. (2006). Changes in spring phytoplankton communities and nutrient dynamics in the eastern basin of Lake Erie since the invasion of Dreissena spp. *Canadian Journal of Fisheries and Aquatic Sciences*, 63(7), 1549-1563.

Bezard, J., Blond, J. P., Bernard, A., & Clouet, P. (1994). The metabolism and availability of essential fatty acids in animal and human tissues. *Reproduction Nutrition Development*, 34(6), 539-568.

Blenckner, T. (2008). Models as tools for understanding past, present and future changes in large lakes. *Hydrobiologia*, 599:177-182

Brett, M., & Müller - Navarra, D. (1997). The role of highly unsaturated fatty acids in aquatic foodweb processes. *Freshwater Biology*, 38(3), 483-499.

CCRR-05 Colombo, S.J., D.W. McKenney, K.M. Lawrence and P.A. Gray. 2007. Climate change projections for Ontario: Practical information for policymakers and planners. Ontario Ministry of Natural Resources, Applied Research and Development Branch, Sault Ste. Marie, Ontario. Climate Change Research Report CCRR-05. 37 p.

Chisholm, S. W. (1992). Phytoplankton size. In Primary productivity and biogeochemical cycles in the sea (pp. 213-237). New York, NY: Springer.

Christensen, J.H., Hewitson B., Busuioc A., Chen A., Gao X., Held I., Jones R., Kolli R.K., Kwon W.-T., Laprise R., Magaña Rueda V., Mearns L., Menéndez C.G., Räisänen J., Rinke A., Sarr A.and Whetton P., (2007). Regional Climate Projections. In: Climate Change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change [Solomon, S., Qin D., Manning M., Chen Z., Marquis M., Averyt K.B., Tignor M. and Miller H.L. (eds.)]. Cambridge, United Kingdom and New York, NY, USA: Cambridge University Press.

Christensen, M. R., Graham, M. D., Vinebrooke, R. D., Findlay, D. L., Paterson, M. J., & Turner, M. A. (2006). Multiple anthropogenic stressors cause ecological surprises in boreal lakes. *Global Change Biology*, 12(12), 2316-2322.

Clair, T. A., Ehrman, J. M., Ouellet, A. J., Brun, G., Lockerbie, D., & Ro, C. U. (2002). Changes in freshwater acidification trends in Canada's Atlantic provinces: 1983–1997. *Water, Air, and Soil Pollution*, 135(1-4), 335-354.

Drake, J. M. (2005). Population effects of increased climate variation. *Proceedings of the Royal Society B: Biological Sciences*, 272(1574), 1823-1827.

Eimers, M. C., Winter, J. G., Scheider, W. A., Watmough, S. A., & Nicholls, K. H. (2005). Recent changes and patterns in the water chemistry of Lake Simcoe. *Journal of Great Lakes Research*, 31(3), 322-332.

Evans, D. O. (2007). Effects of hypoxia on scope-for-activity and power capacity of lake trout (Salvelinus namaycush). *Canadian Journal of Fisheries and Aquatic Sciences*, 64(2), 345-361.

Evans, D. O., Nicholls, K. H., Allen, Y. C., & McMurtry, M. J. (1996). Historical land use, phosphorus loading, and loss of fish habitat in Lake Simcoe, Canada. *Canadian Journal of Fisheries and Aquatic Sciences*, 53(S1), 194-218.

Evans, D. O., Skinner, A. J., Allen, R., & McMurtry, M. J. (2011). Invasion of zebra mussel, Dreissena polymorpha, in Lake Simcoe. *Journal of Great Lakes Research*, 37, 36-45.

Evans, D. O., & Waring, P. (1987). Changes in the multispecies, winter angling fishery of Lake Simcoe, Ontario, 1961-83: invasion by rainbow smelt, Osmerus mordax, and the roles of intra-and interspecific interactions. *Canadian Journal of Fisheries and Aquatic Sciences*, 44(S2), s182-s197.

Farkas, T., Nemecz, G. Y., & Csengeri, I. (1984). Differential response of lipid metabolism and membrane physical state by an actively and passively overwintering planktonic crustacean. Lipids, 19(6), 436-442.

Hawryshyn, J., Rühland, K. M., Quinlan, R., & Smol, J. P. (2012). Long-term water quality changes in a multiple-stressor system: a diatom-based paleolimnological study of Lake Simcoe (Ontario, Canada). *Canadian journal of fisheries and aquatic sciences*, 69(1), 24-40.

Hecky, R. E., Smith, R. E., Barton, D. R., Guildford, S. J., Taylor, W. D., Charlton, M. N., & Howell, T. (2004). The nearshore phosphorus shunt: a consequence of ecosystem engineering by dreissenids in the Laurentian Great Lakes. *Canadian Journal of Fisheries and Aquatic Sciences*, 61(7), 1285-1293.

Hudson, J. J., Taylor, W. D., & Schindler, D. W. (2000). Phosphate concentrations in lakes. *Nature*, 406(6791), 54-56.

Hutchinson, G. R. (1967). *A Treatise on Lmnology, Vol. II: Introduction to Lake Biology and the Limnoplankton*. New York: Wiley.

IPCC (Intergovernmental Panel on Climate Change) Fifth Assessment Report (2013), Climate Change 2013: The Physical Science Basis. Available at: http://www.ipcc.ch (accessed April 11, 16)

Jöhnk, K. D., Huisman, J. E. F., Sharples, J., Sommeijer, B. E. N., Visser, P. M., & Stroom, J. M. (2008). Summer heatwaves promote blooms of harmful cyanobacteria. *Global Change Biology*, 14(3), 495-512.

[LSWEMR]Lake Simcoe Watershed Environmental Monitoring Report (2007-2011 data) (2013). Available at: http://www.lsrca.on.ca/pdf/reports/monitoring_report_2013.pdf

[LSEMS] Lake Simcoe Environmental Management Strategy. 2008. Lake Simcoe basin wide report. Lake Simcoe Environmental Management Strategy Report; [cited July 5 2015]. Available at: http://www.lsrca.on.ca/pdf/reports/lsems/basin_wide_report.pdf

MacDonald, G. M., Edwards, T. W., Moser, K. A., Pienitz, R., & Smol, J. P. (1993). Rapid response of treeline vegetation and lakes to past climate warming.

MacIsaac, H. J., Grigorovich, I. A., & Ricciardi, A. (2001). Reassessment of species invasions concepts: the Great Lakes basin as a model. *Biological Invasions*, 3(4), 405-416.

Magnuson, J. J., Webster, K. E., Assel, R. A., Bowser, C. J., Dillon, P. J., Eaton, J. G., Evans H. E., Fee E. J., Hall R. I., Mortsch L. R., Schindler D. W. & Quinn, F. H. (1997). Potential effects of climate changes on aquatic systems: Laurentian Great Lakes and Precambrian Shield Region. *Hydrological processes*, 11(8), 825-871.

Müller-Navarra, D. C., Brett, M. T., Liston, A. M., & Goldman, C. R. (2000). A highly unsaturated fatty acid predicts carbon transfer between primary producers and consumers. *Nature*, 403(6765), 74-77.

Nalepa T. F., & Schloesser D. W. (Eds.). (2013). Zebra mussels biology, impacts, and control. New York, NY: CRC press.

Nicholls, K. H. (1997) A Limnological Basis For A Lake Simcoe Phosphorus Loading Objective, *Lake and Reservoir Management*, 13:3, 189-198.

Nürnberg, G. K., LaZerte, B. D., Loh, P. S., & Molot, L. A. (2013). Quantification of internal phosphorus load in large, partially polymictic and mesotrophic Lake Simcoe, Ontario. *Journal of Great Lakes Research*, *39*(2), 271-279.

Nürnberg, G. K., Molot, L. A., O'Connor, E., Jarjanazi, H., Winter, J., & Young, J. (2013). Evidence for internal phosphorus loading, hypoxia and effects on phytoplankton in partially polymictic Lake Simcoe, Ontario. *Journal of Great Lakes Research, 39*(2), 259-270.

North, R. L., Barton, D., Crowe, A. S., Dillon, P. J., Dolson, R. M. L., Evans, D. O., Ginn, B. K., Håkanson, L., Hawryshyn, J., Jarjanazi, H., King, J. W., La Rose, J. K. L., León, L., Lewis, C. F. M., Liddle, G. E., Lin, Z. H., Longstaffe, F. J., Macdonald, R. A., Molot, L., Ozersky, T., Palmer, M. E., Quinlan, R., Rennie, M. D., Robillard, M. M., Rodé, D., Rühland, K. M., Schwalb, A., Smol, J. P., Stainsby, E., Trumpickas, J. J., Winter, J. G., & Young, J. D. (2013). The state of Lake Simcoe (Ontario, Canada): the effects of multiple stressors on phosphorus and oxygen dynamics. *Inland Waters*, *3*(1), 51-74.

Palmer, Michelle E. (2012). Thesis: Aquatic responses to multiple environmental stressors. York University, Ontario.

Palmer, M. E., Hiriart-Baer, V. P., North, R. L., & Rennie, M. D. (2013). Preface to special theme, part 2: Summary of Lake Simcoe's past, present and future. *Inland Waters*, 3(2), 119-124.

Quinlan, R., Hall, R. I., Paterson, A. M., Cumming, B. F., & Smol, J. P. (2008). Long term assessments of ecological effects of anthropogenic stressors on aquatic ecosystems from paleoecological analyses: challenges to perspectives of lake management. *Canadian Journal of Fisheries and Aquatic Sciences*, 65(5), 933-944.

Rennie, M. D., Sprules, W. G., & Johnson, T. B. (2009). Resource switching in fish following a major food web disruption. *Oecologia*, 159(4), 789-802.

Robarts, R. D., & Zohary, T. (1987). Temperature effects on photosynthetic capacity, respiration, and growth rates of bloom‐forming cyanobacteria. *New Zealand Journal of Marine and Freshwater Research*, 21(3), 391-399.

Rodé Danielle L. (2009). A paleolimnological assessment of coldwater fish habitat quality in Lake Simcoe, Ontario. York University, Ontario.

Rühland, K., Paterson, A. M., & Smol, J. P. (2008). Hemispheric‐scale patterns of climate‐related shifts in planktonic diatoms from North American and European lakes. *Global Change Biology*, 14(11), 2740-2754.

Schindler, D. W. (2001). The cumulative effects of climate warming and other human stresses on Canadian freshwaters in the new millennium. *Canadian Journal of Fisheries and Aquatic Sciences*, 58(1), 18-29.

Schindler, D. W. (2006). Recent advances in the understanding and management of eutrophication. *Limnology and Oceanography*, 51(1), 356-363.

Schindler, D. W., Beaty, K. G., Fee, E. J., Cruikshank, D. R., DeBruyn, E. R., Findlay, D. L., Linsey G. A., Shearer J. A., Stainton M. P., and Turner, M. A. (1990). Effects of climatic warming on lakes of the central boreal forest. *Science*, 250(4983), 967-970.

Schindler, D. W., Curtis, P. J., Parker, B. R., & Stainton, M. P. (1996). Consequences of climate warming and lake acidification for UV-B penetration in North American boreal lakes. *Nature*, 379(6567), 705-708.

Sellers, T. J., Parker, B. R., Schindler, D. W., & Tonn, W. M. (1998). Pelagic distribution of lake trout (Salvelinus namaycush) in small Canadian Shield lakes with respect to temperature, dissolved oxygen, and light. *Canadian Journal of Fisheries and Aquatic Sciences*, 55(1), 170-179.

Sharma, S., Vander Zanden, M. J., Magnuson, J. J., & Lyons, J. (2011). Comparing climate change and species invasions as drivers of coldwater fish population extirpations. *PloS one*, *6*(8), e22906.

Smol, J. P. (2009). Pollution of lakes and rivers: a paleoenvironmental perspective. Oxford: Blackwell Publishing

Soendergaard, M., Jensen, J. P., & Jeppesen, E. (2005). Seasonal response of nutrients to reduced phosphorus loading in 12 Danish lakes. *Freshwater Biology*, 50(10), 1605-1615.

Sommer, U. (1985). Seasonal succession of phytoplankton in Lake Constance. *Bioscience*, 351-357.

Stainsby, E. A., Winter, J. G., Jarjanazi, H., Paterson, A. M., Evans, D. O., & Young, J. D. (2011). Changes in the thermal stability of Lake Simcoe from 1980 to 2008. *Journal of Great Lakes Research*, 37, 55-62.

Stoddard, J. L., Jeffries, D. S., Lükewille, A., Clair, T. A., Dillon, P. J., Driscoll, C. T., Forsius M., Johannessen M., Kahl J. S., Kellogg J. H., Kemp A., Mannio J., Monteith D. T., Murdoch P. S., Patrick S., Rebsdorf A., Skjelkvåle B. L., Stainton M. P., Traaen T., van Dam H., Webster K. E., Wieting J., & Wilander, A. (1999). Regional trends in aquatic recovery from acidification in North America and Europe. *Nature*, 401(6753), 575-578.

Stoermer, E. F., Ladewski, B. G., & Schelske, C. L. (1978). Population responses of Lake Michigan phytoplankton to nitrogen and phosphorus enrichment. *Hydrobiologia*, 57(3), 249-265.

Strayer, D. L. (2010). Alien species in fresh waters: ecological effects, interactions with other stressors, and prospects for the future. *Freshwater Biology*, 55(s1), 152-174.

Strecker, A. L., Cobb, T. P., & Vinebrooke, R. D. (2004). Effects of experimental greenhouse warming on phytoplankton and zooplankton communities in fishless alpine ponds. *Limnology and Oceanography*, *49*(4), 1182-1190.

Taylor, W. D. (2010). Nature of dissolved P regenerated by plankton: implications for the ssPO4 radiobioassay and for the nature of dissolved P. *Aquatic sciences, 72*(1), 13-20.

Tilman, D. (1977). Resource competition between plankton algae: an experimental and theoretical approach. *Ecology*, 58(2), 338-348.

Tilman, D. (1981). Tests of resource competition theory using four species of Lake Michigan algae. *Ecology*, 802-815.

Tilman, D., Kilham, S. S., & Kilham, P. (1982). Phytoplankton community ecology: the role of limiting nutrients. *Annual Review of Ecology and Systematics, 13*, 349-372.

Walther, G. R., Post, E., Convey, P., Menzel, A., Parmesan, C., Beebee, T. J., Fromentin J., Hoegh-Guldberg O., & Bairlein, F. (2002). Ecological responses to recent climate change. *Nature*, 416(6879), 389-395.

Ward, J. M., & Ricciardi, A. (2007). Impacts of Dreissena invasions on benthic macroinvertebrate communities: a meta‐analysis. *Diversity and Distributions*, 13(2), 155-165.

Winder, M., & Hunter, D. A. (2008). Temporal organization of phytoplankton communities linked to physical forcing. *Oecologia*, 156(1), 179-192.

Winder, M., & Schindler, D. E. (2004). Climatic effects on the phenology of lake processes. *Global Change Biology*, 10(11), 1844-1856.

Van Zuiden, T. M., Chen, M. M., Stefanoff, S., Lopez, L., & Sharma, S. (2016). Projected impacts of climate change on three freshwater fishes and potential novel competitive interactions. *Diversity and Distributions*, 22, 603-614

Vanderploeg, H. A., Liebig, J. R., Carmichael, W. W., Agy, M. A., Johengen, T. H., Fahnenstiel, G. L., & Nalepa, T. F. (2001). Zebra mussel (Dreissena polymorpha) selective filtration promoted toxic Microcystis blooms in Saginaw Bay (Lake Huron) and Lake Erie. *Canadian Journal of Fisheries and Aquatic Sciences*, 58(6), 1208-1221.

Verburg, P., Hecky, R. E., & Kling, H. (2003). Ecological consequences of a century of warming in Lake Tanganyika. *Science*, 301(5632), 505-507.

Wagner, C., & Adrian, R. (2009). Cyanobacteria dominance: quantifying the effects of climate change. *Limnology and Oceanography*, 54(6, part 2), 2460-2468.

Winder, M., & Sommer, U. (2012). Phytoplankton response to a changing climate. *Hydrobiologia*, 698(1), 5-16.

Winter, J. G., Eimers, M. C., Dillon, P. J., Scott, L. D., Scheider, W. A., & Willox, C. C. (2007). Phosphorus inputs to Lake Simcoe from 1990 to 2003: declines in tributary loads and observations on lake water quality. *Journal of Great Lakes Research*, 33(2), 381-396.

Winter, J. G., Dillon, P. J., Futter, M. N., Nicholls, K. H., Scheider, W. A., & Scott, L. D. (2002). Total phosphorus budgets and nitrogen loads: Lake Simcoe, Ontario (1990 to 1998). *Journal of Great Lakes Research*, 28(3), 301-314.

Winter, J. G., Young, J. D., Landre, A., Stainsby, E., & Jarjanazi, H. (2011). Changes in phytoplankton community composition of Lake Simcoe from 1980 to 2007 and relationships with multiple stressors. *Journal of Great Lakes Research*, 37, 63-71.

Young, J. D., Winter, J. G., & Molot, L. (2011). A re-evaluation of the empirical relationships connecting dissolved oxygen and phosphorus loading after dreissenid mussel invasion in Lake Simcoe. *Journal of Great Lakes Research*, 37, 7-14.

CHAPTER 1:

LONG-TERM CHANGES IN VOLUME-WEIGHTED HYPOLIMNETIC DISSOLVED OXYGEN (VWHDO) IN A LARGE LAKE: EFFECTS OF INVASIVE SPECIES, EUTROPHICATION AND CLIMATE CHANGE ON LAKE SIMCOE, 1980-2012.

Jiahua Li[1], Lewis Molot[1], Michelle Palmer[2], Jennifer Winter[2], Joelle Young[2] and Eleanor Stainsby[2]

[1]York University, [2]Ontario Ministry of the Environment

Keywords: climate change, phosphorus reduction, dreissenid mussels, oxygen depletion

SUMMARY

Lake Simcoe has been influenced by multiple drivers since 1980 especially by reductions in phosphorus (P) loading, climate change, and invasive species such as dressenid mussels (DM) which became firmly established after 1996. We examined the cumulative impact of these drivers on the volume-weighted hypolimnetic dissolved oxygen concentration (VWHDO) below 18 m at station K42 in Kempenfelt Bay during ice-free season. Hypolimnetic DO depletion began in early spring when thermal stratification was observable but weak and continued throughout the ice-free season until cooling sufficiently lowered stability. Mean initial VWHDO ($VWHDO_{init}$) and minimum ice-free VWHDO ($VWHDO_{min}$) were higher after 1996 but the duration of the depletion period (L) was longer. In comparison to pre-DM invasion period (1980-1996), post-DM period (1997-2012) had 2.4 mg/L higher $VWHDO_{init}$, and 1.54 mg/L higher $VWHDO_{min}$. The observed DO depletion rate (DR) and temperature adjusted DO depletion rate (DR_{adj}) was slightly lower (0.3% and 4.6% respectively) but L was 26 days longer. Multiple linear regressions revealed that $VWHDO_{min}$ was positively correlated with the invasion of DM and negatively correlated with hypolimnetic water temperature; DR was positively correlated with ammonia and TP annual loading. Hence, longer L may have offset improvements in $VWHDO_{min}$ generated by P controls and invasive species.

INTRODUCTION

Lake Simcoe, the largest inland lake in southern Ontario outside the Great Lakes, is an invaluable natural, social, and economic resource. Lake Simcoe has 6 drinking water treatment plants (WTPs) that provide drinking water several communities in the watershed. It also assimilates wastewater from 14 municipal water pollution control plants (WPCPs). In addition, the agriculture sector is worth more than $500 million annually including crops and livestock (LSEMS 2010; Winter et al. 2007). Recreational activities include cottaging, boating and sport fishing, especially cold water species like lake trout, lake whitefish, lake herring) generate over $200 million per year industry accounting for 15% (1 million angler hours per year) of all angling effort in Ontario (Eimers et al., 2005; LSEMS, 2010). However, Lake Simcoe and its watershed have experienced multiple pressures from increasing human activities over the past ~200 years, including logging, damming, canal construction, agriculture, urban development, species invasion and recently climate change (Hawryshyn et al. 2012).

Cold water fish require cold temperatures and high dissolved oxygen (DO) concentrations to survive (Gibson and Fry 1954, Rudstam and Magnuson 1985), and consequently are sensitive to anthropogenic stresses that raise water temperatures or lower DO concentrations, for example, climate change (Sharma et al. 2011) and eutrophication (Welch et al. 2011). Elevated inputs of nutrients (e.g. phosphorus (P) and nitrogen (N), essential elements that support and maintain aquatic life) to the lake result in excess growth of phytoplankton, which settle and decompose consuming DO by respiration. Elevated microbial respiration rates are especially problematic in the bottom layer of thermally stratified lakes (hypolimnion) because inputs of new DO are typically very low until the lake mixes in the fall. Under climate change as air temperatures are expected to increase, surface water (epilimnion) temperatures are expected to increase accordingly (Sharma et al. 2008) as is the duration of the lake thermal stratification period (Stainsby et al. 2011), isolating the deep waters from top well-oxygenated water for a longer period. With both stresses in mind, Ficke et al. (2007) proposed a “temperature-oxygen squeeze” scenario for coldwater fish habitat.

Coldwater fish have a suitable temperature niche between 5-15˚C (most commonly below 8˚C, Plumb and Blanchfield 2009) and a suitable dissolved oxygen threshold above 6-7 mg/L (Evans et al. 1991 and Evans 2007). DO concentrations below 3 mg/L is considered lethal to many cold water fish species (Evans et al. 1996 and Evans 2007). After ice-out, lake trout inhabit surface water for feeding (Martin 1970) and as the epilimnetic water temperature gets warmer, they retreat to deeper water (Fry and Kennedy 1937; Fry 1939) which marks the cessation of surface feeding until fall overturn (Martin 1970). Previous research suggests that earlier stratification forces earlier migration of all ages of lake trout to deeper, colder waters, curtailing intense spring feeding and consequently reducing growth (King et al. 1999) etc. DO continuously decreases after spring overturn in deeper waters even before development of a thermocline. Keeping in mind that low oxygen levels will harm the cold water fishery, an end-of-summer a minimum volume-weighted hypolimnetic dissolved oxygen (VWHDO) target of 7 mg/L by September 15 was set by the Lake Simcoe Protection Plan (LSPP) as the optimal target to restore a self-sustaining coldwater fish community (LSPP 2009). Although, in theory, longer stratification periods due to climate change (Stainstby et al. 2011) could lead to lower hypolimnetic oxygen in late summer months, to date, no empirical analysis has been done to evaluate to what extent the hypolimnetic DO has deteriorated due to climate change. VWHDO may continue to decrease even after the set date by LSPP with the delay of fall overturn.

In this paper, our overall goal was to examine long-term patterns of VWHDO dynamics, an important indicator of coldwater fish habitat, in relation to major environmental changes in the past three decades. These changes include eutrophication followed by P reduction beginning around 1990, introduction of invasive dreissenid mussels, which became well established by 1997, and climate change post 1980. Specific objectives were three-fold: first, to examine annual thermal structure patterns and their long-term trends; second, to examine trends in VWHDO dynamics including spring VWHDO, VWHDO depletion rate, the length of the VWHDO depletion period and minimum ice-free VWHDO; third, to determine the relative influences of major environmental stresses on VWHDO dynamics.

MATERIALS AND METHODS

Study Site

Lake Simcoe, located in southern Ontario between Georgian Bay of Lake Huron to northwest and Lake Ontario to the southeast (Fig. 1), is a large, relatively shallow lake (area A = 722km^2, mean depth $\bar{z}$ = 14m, maximum depth z_{max}= 42 m, shoreline perimeter = 303 km, volume = 11x10^9 m^3, residence time = 11 years) with a watershed area of 2899 km^2 and 35 tributaries (Evans et al., 2011; North, 2013; Winter et al., 2007). Kempenfelt Bay (A = 35.74 km^2, $\bar{z}$ = 26 m, z_{max}= 42 m) contains the deepest part of the lake and exhibits thermal stratification.

Lake Simcoe is shallow relative to its surface area, which is evident from a low morphometric ratio (Osgood Index $= \frac{\bar{Z}}{\sqrt{A_0}}$, Osgood, 1988) of 0.52. The morphometric ratio for Kempenfelt is much larger, 4.35.

Data acquisition and field & laboratory methods

Temperature profiles, DO profiles, water chemistry, and morphometric data for Lake Simcoe were provided by the Ontario Ministry of the Environment and Climate Change (OMOE). The OMOE monitored water quality in Lake Simcoe at station K42 from 1980 to 2012. The station was sampled every two weeks through ice-free period (typically, May to November). Water samples were obtained through euphotic zone using a polyvinyl chloride (PVC) hose for chemical analysis (total P (TP), Total Kjeldahl N (TKN), total nitrate (NO_2 + NO_3), ammonia (NH_4), alkalinity, chlorophyll a, sulfate (SO_4), pH, calcium and silica. Temperature and dissolved oxygen profiles were measured at K42 using a YSI probe at 1-meter intervals to the bottom and Secchi disk depth is defined as the water depth in meters at which a black and white Secchi disk is no longer visible to an observer at the surface (Winter et al., 2011).

Air temperature data were obtained from Environment Canada’s weather station (http://climate.weather.gc.ca/) at Shanty Bay, which is 10 km northeast of the city Barrie and close to station K42 in Kempenfelt Bay.

Estimates of annual TP loading are described in O’Connor et al. (2013).

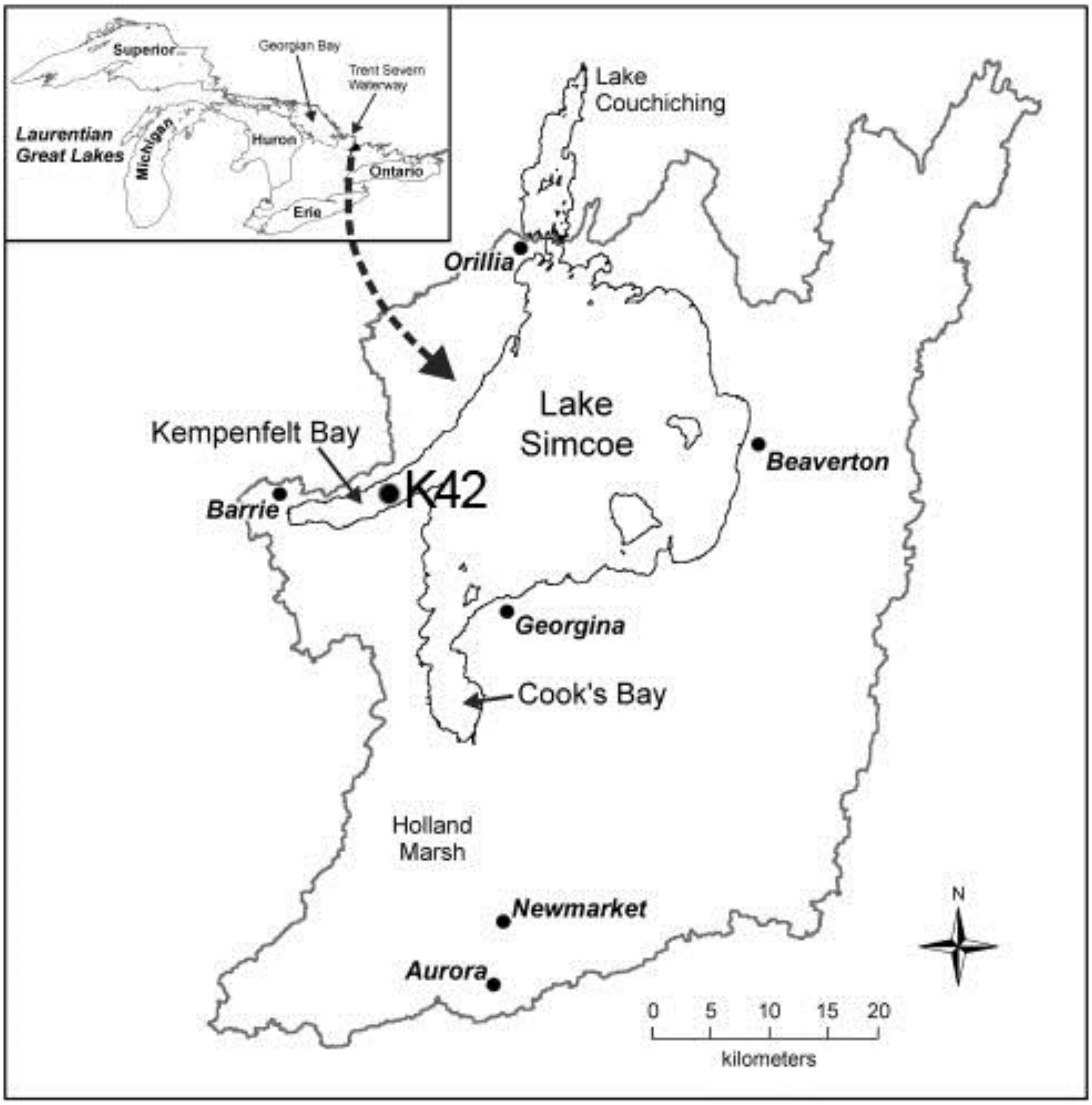

Figure 1. Map of Lake Simcoe showing Main basin, Kempenfelt Bay, and Cook's Bay. Station K42 is shown on the map.

Data Processing and Analyses

Schmidt Stability index

Timing of thermal stratification onset and termination were calculated from Schmidt's stability index S (Hutchinson, 1957; Idso, 1973; Schmidt, 1928) as follows:

$$S = A_0^{-1} \sum_{z_0}^{z_m} (z - z_{\bar{\rho}})(\rho_z - \bar{\rho}) A_z \, \Delta z$$

where S is the Schmidt's stability index (g cm cm^{-2}), A_0 is the surface area of the lake, $z_{\bar{\rho}}$ is the depth at which the mean density is found, ρ_z is the density of the water at depth z, $\bar{\rho}$ is the mean density of the water column, and A_z is the stratum area at depth z. The summation is taken over all depths (z) at interval (Δz) of 1 m from the surface (z_0) to the maximum depth (z_m). Water density was calculated based on Millero & Poisson (1981) and Martin & McCutcheon (1999). This index is a measurement of water column stability and indicates the amount of mechanical work required to mix the lake to an isothermal condition (the temperature of water at mean density). A high value of S indicates strong stratification requiring more energy to mix; a low value of S indicates near isothermal condition requiring low energy to mix. A subjective threshold of 800 g cm cm^{-2} was considered to be the stability at which onset of stratification occurred in spring in station K42 (Stainsby et al. 2011). This threshold approximately reflects the establishment of a thermocline (temperature difference >1˚C within a 1-m interval) in the water column. However, choice of this particular threshold had no effect on the significance of the long-term trend.

S curves were analyzed and divided into ascending and descending limbs where linear and polynomial regressions were fitted to each limb. The best fit was selected using ANOVA and used to estimate the days on which S = 800 in the spring and fall, i.e., the beginning and end of the official thermal stratification period. Instantaneous rates of increase and decrease of S in spring and fall, respectively, were determined by the selected regression model. Average S during stratification was calculated as the sum of S for each day during the stratification period divided by the length of the period in days.

Volume-weighted hypolimnetic dissolved oxygen

Volume-weighted hypolimnetic dissolved oxygen (VWHDO) is generally defined as the DO below the upper limit of hypolimnion calculated by the formula in (Quinlan et al., 2005). In Lake Simcoe, in order to isolate hypolimnetic DO from replenish factor, an upper ceiling for the tropholytic zone was set at 18 meter for DO modeling (Nicholls, 1997). Therefore, VWHDO was calculated using the following formula:

$$VWHDO = \frac{\sum_{42}^{18} bathV_i \times DO_i}{V_{hypo}}$$

where bathV$_i$ is 1-meter thick stratum volume at i meter depth; DO$_i$ is dissolved oxygen at i meter depth; V$_{hypo}$ is total hypolimnetic volume between 18 and 42 m.

Spring VWHDO was determined as the linearly interpolated VWHDO on day 160 of the year or 60 days after ice-out. Initial VWHDO (VWHDO$_{init}$) was determined as the linearly interpolated VWHDO when stability reached 800 g cm^2/cm. VWHDO$_{min}$ was determined as the minimum observed VWHDO during the ice-free period.

The observed VWHDO depletion rate (DR) was calculated from the slope of the change in the VWHDO over time from early summer to the day of $\text{VWHDO}_{\text{min}}$ (see Fig. 4) excluding high VWHDO values caused by replenishment from large storm mixing events (Burns, 1995).

The calculated DRs were adjusted to account for interannual variations in hypolimnetic temperature. The DR_{adj} was standardised to 10 C° to account for the effects of temperature on hypolimnetic metabolism (the average hypolimnetic temperature over the period of VWHDO depletion rate calculation between 1980-2012 was 9.47 ±1.26°C). The temperature standardisation equation was expressed as a modified Arrhenius expression (Q10 equation, Burns, 1995; Chapra, 1997, Salisbury and Ross 1978) which allows investigators to look at different sources of annual variation in rates covariate at the same temperature.

$$DR(t_{10^{\circ}C}) = DR(t_i)k^{(t_{10^{\circ}C}-t_i)}$$

where $DR(t_i)$ and $DR(t_{10^{\circ}C})$ are depletion rates at the observed temperature t and standard temperature $t_{10^{\circ}C}$, respectively, and k, is the temperature coefficient. If we assume that an increase of 10°C increases the metabolic rate by 2 fold, then k = 1.0718. DR adjusted for temperature differences was termed DR_{adj}.

Data quality assurance/quality control

The data were checked manually and modified/deleted as necessary due to unrealistic data (e.g. water temperature data greater than 50°C; DO data greater than 18 mg/L; sudden change within adjacent data points in the same temperature/DO profile). Edited data were then used to calculate volume weighted hypolimnetic dissolved oxygen and temperature (VWHDO and VWHT).

Data analyses

Schmidt's Stability index was calculated using the "schmidt.stability" function in" rLakeAnalyzer" package (Brentrup et al. 2014 and Read et al. 2011).

The Mann-Kendall trend test (Mann 1945, Kendall 1975, Yu et al. 1993, Yue et al. 2002) was used to test whether there were statistically significant long-term monotonic trends over time in the ice phenology, water temperature, thermal regime, parameters in VWHDO analysis over the study period. The test is relatively insensitive to missing data and it did not assume the data are normally distributed (Futter, 2003). We used the "Kendall" function (McLeod 2013) for the Mann-Kendall trend test.

Multiple linear regression models (Tabachnick and Fidell 2001) fitted by least squares and based on forward selection were performed in R using the "lm" and "forward.sel" functions. Multicollinearity between predictor variables were checked using correlation coefficients, bivariate plots, and variance inflation factors to determine collinearity and decide which variables to retain. Multiple regression assumes normality and homoscedacity of residuals and variables were transformed to meet the assumptions if necessary.

RESULTS

Mean summer (June-September) and annual air temperatures recorded in Shanty Bay weather station showed significant warming trends (Table 1). Volume weighted epilimnetic (0-18 meter strata) water temperature at station K42 increased significantly by 1.88˚C between 1980 and 2012 (Table 1). This trend influenced the onset of spring thermal stratification in Lake Simcoe, which showed a significant monotonic advancement towards an earlier date in the year (Table 1, Fig. 2A) by approximately 20 days; the timing of fall overturn (defined by S declining to 800) showed a significant monotonic prolongation to a later date in the year (Table 1, Fig. 2B) by 15 days. As a consequence, the duration of thermal stratification at K42 showed a significant monotonic trend to a longer period (Table 1, Fig. 2C) of more than a month (i.e. 35 days). Over the study period (1980-2012), average S during the stratified period increased monotonically (Table 1, Fig. 2D); the instantaneous rate of S increase at day of the year 150 increased monotonically (Table 1, Fig. 2E); maximum S showed an increasing trend (Table 1, Fig. 2F). The rate of S decrease in late summer and fall did not change significantly (Table 1). The volume weighted hypolimnetic (18-42 meter strata) water temperature at station K42 did not change between 1980 and 2012 (Table 1).

Table 1. Monotonic trend tests of long-term change in key environmental variables in K42 Lake Simcoe from 1980-2012

Variables	Statistics value	p-value	Slope of linear regression	R^2 adjusted	Estimated change from 1980-2012	Unit
Timing of ice-out date	tau (df=31)=-.112	0.380	-0.222	0.019	-7.33	Day of the year
Timing of free-up date	tau (df=31)=.263	0.0347	0.454	0.121	14.97	Day of the year
Ice-on period	tau (df=31)=-.262	0.0377	-0.657	0.141	-21.67	Days
Mean annual air temperature	tau (df=31)=.341	0.0055	0.048	0.236	1.59	˚C
Mean summer air temperature	tau (df=31)= 0.364	0.003	0.050	0.230	1.64	˚C
Mean summer volume-weighted water temperature	tau (df=31)= 0.36	0.003	0.054	0.253	1.80	˚C
Mean summer volume-weighted hypolimnetic water temperature	tau (df=31)= 0.0644	0.609	0.020	-0.008	0.66	˚C
Mean summer volume-weighted epilimnetic water temperature	tau (df=31)= 0.201	0.104	0.057	0.038	1.88	˚C
Timing of Onset of Thermal Stratification	tau (df=31)= -0.367	0.003	-0.584	0.245	-19.27	Day of the year
Timing of Fall Overturn	tau (df=31)= 0.443	0.0003	0.44	0.320	14.52	Day of the year
Duration of Thermal Stratification	tau (df=31)= 0.481	0.00009	1.069	0.425	35.26	Days
Mean Schmidt's Stability (during thermal stratification period)	tau (df=31)= 0.36	0.003	8.092	0.212	267.04	g cm^2/cm
Rate of S increase at DOY 150	tau (df=31)= 0.379	0.002	0.434	0.162	14.32	g cm^2/cm day
Rate of S decrease	tau (df=31)= 0.0189	0.889	0.169	-0.017	5.58	g cm^2/cm day
Max S of the year	tau (df=31)= 0.201	0.104	10.074	0.103	332.44	g cm^2/cm
VWHDOmin post 1996	tau (df=14)=-0.333	0.018	-0.095	0.289	-1.42	mg/L
Spring VWHDO (160 DOY)	tau (df=30)=0.44	0.0004	0.093	0.301	3.07	mg/L
Spring VWHDO (60 day after ice-out)	tau (df=29)=0.531	0.00003	0.101	0.365	3.32	mg/L
Initial VWHDO	tau (df=31)=0.462	0.0002	0.138	0.359	4.56	mg/L
Duration of VWHDO depletion	tau (df=31)=0.345	0.005	0.787	0.231	25.98	Days

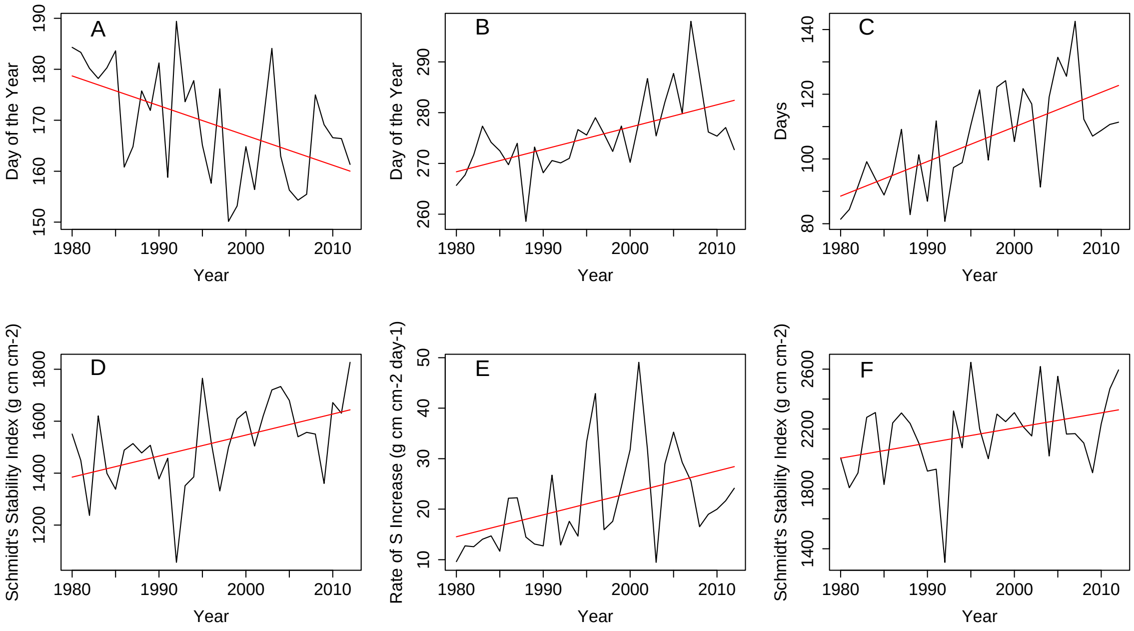

Figure 2. Schmidt's stability analysis during 1980-2012 in Lake Simcoe K42, for: (A) timing of onset of thermal stratification (in day of the year, DOY); (B) timing of fall overturn (DOY); (C) duration of thermal stratification (days); (D) average S during thermal stratified period; (E) instantaneous rate of S increase at DOY 150; (F) Max S of the year.

In comparison to the pre-DM invasion period (1980-1996), $VWHDO_{min}$ was higher in the post-DM invasion period (1997-2012) with means of 2.69 to 4.23 mg/L, respectively (two tailed t-test, $t_{28.2} = -4.5249$, $p = 0.0001001$). However, $VWHDO_{min}$ began decreasing in 2008 (Figure 3A, Table 1). DR, both unadjusted and corrected for temperature, were variable (Figure 3B); for both spring VWHDO at 160 DOY and 60 days after ice-out increased monotonically (Figure 3C, Table 1). Initial VWHDO ($VWHDO_{ini}$), defined as VWHDO at onset of thermal stratification (where S = 800 g cm/cm^2), increased monotonically during the entire study period (Figure 3D, Table 1). The VWHDO depletion period increased monotonically (Figure 3E, Table 1) (Figure 2C). Predicted $VWHDO_{min}$, calculated by the equation: $VWHDO_{min} = VWHDO_{ini} - L*DR$, was in good agreement with observed $VWHDO_{min}$ (Figure 3F) although the good agreement may in part be due to a spurious correlation ($VWHDO_{min}$ occurs on both sides the equation).

VWHDO depletion began in early spring when thermal stratification was slightly positive but weak, i.e., S was in general, below 500 g cm cm^{-2}, but in most years, it began when S was slightly above 0 g cm^2/cm ((Fig. 4) and continued throughout the ice-free season until cooling lowered S to a mean 1126 ± 345 g cm cm^{-2} (± 1 standard deviation) when $VWHDO_{min}$ occurred. Although replenishment events occurred in some of the years during summer, e.g., year 1984, 1985, 1986, 1987, 1991, 1992, 1998, 2003, 2004, 2005, 2010 (Fig. 4), decreasing trends in DR were evident every year. Fall overturn events were distinguished by an increase in VWHDO following $VWHDO_{min}$ and continued increasing until DO was saturated (Fig. 4).

In comparison to the pre-DM invasion period (1980-1996, see Table 2), the post-DM invasion period (1997-2012) had a (1) 15%-28% increase in spring VWHDO (i.e. $VWHDO_{init}$, $VWHDO_{60\ day\ after\ ice\text{-}off}$, and $VWHDO_{day\ of\ the\ year\ 160}$) with an increase between 1.6-2.7 mg/L, (2) 4.7% decrease in DR_{adj}, (3) 14% increase in L, and (4) 57% increase in $VWHDO_{min}$ increasing from 2.7 to 4.2 mg/L.

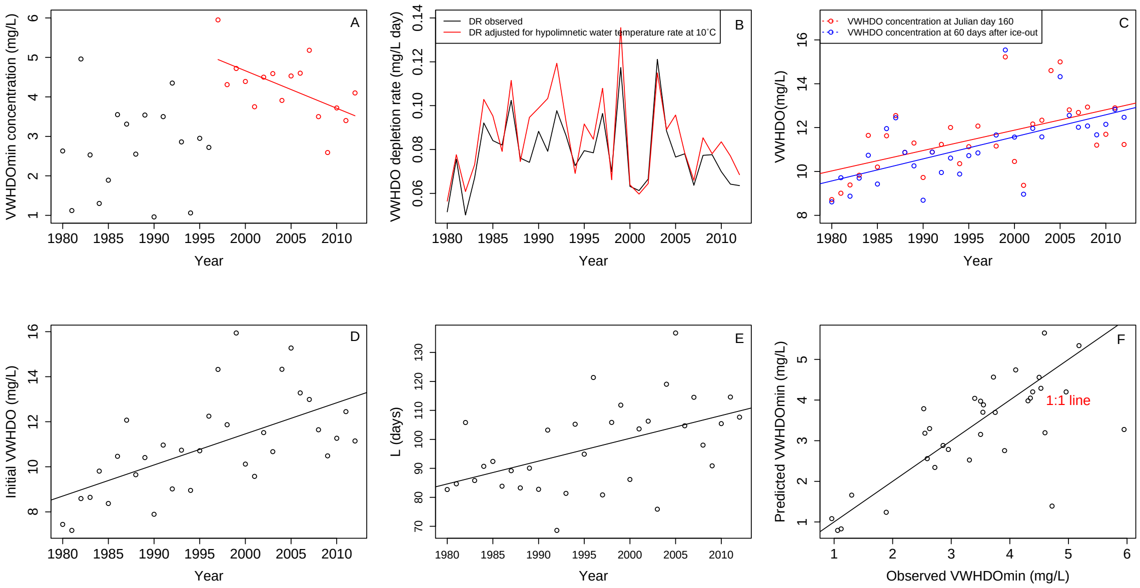

Figure 3. VWHDO analysis during 1980-2012 in Lake Simcoe K42: (A) minimum observed ice-free VWHDO trend; (B) DR observed and adjusted for temperature of 10˚C trends; (C) Spring VWHDO at 160 DOY and 60 days after ice-out trends; (D) Initial VWHDO at onset of thermal stratification trend; (E) VWHDO depletion period trend; (F) Observed vs. predicted VWHDO$_{min}$ with 1:1 line.

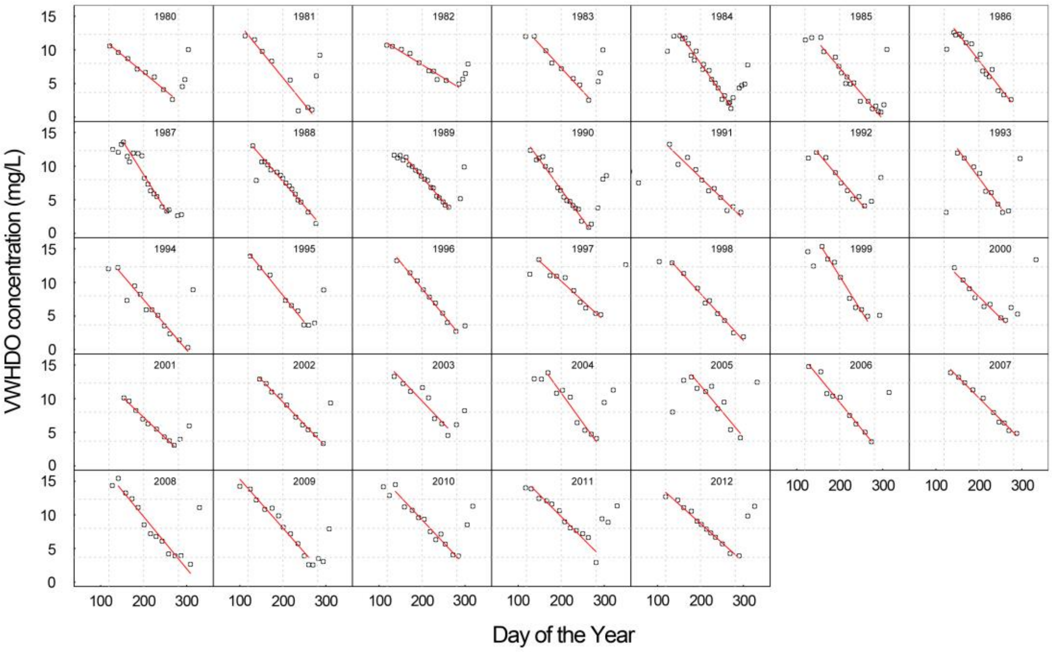

Figure 4. VWHDO depletion curves between 1980-2012 at station K42 during the ice-free season. Open circles indicate VWHDO value at a particular sampling date. Regression lines used to calculate observed VWHDO depletion rate are shown in red solid lines.

Table 2. Comparison of VWHDO parameters in the pre- and post-DM periods.

Parameters (unit)/Time period	**Pre-DM period 1980-1996**	**Post-DM period 1997-2012**	**Change**	**Change percent**
VWHDOinit (mg/L)	9.60	12.31	2.71	28.2%
VWHDO60 (mg/L)	10.25	12.24	1.99	19.5%
VWHDO160 (mg/L)	10.74	12.38	1.65	15.4%
Observed Depletion Rate (mg/L day)	0.079	0.078	0.0002	0.3%
Depletion Rate Adjusted (mg/L day)	0.087	0.083	0.004	4.6%
Depletion Period (days)	90.93	103.88	12.94	14.2%
VWHDOmin (mg/L)	2.69	4.23	1.54	57.2%

Multiple Linear Regressions

Regression analysis was used to identify potential factors affecting dissolved oxygen.

1. Ice-free minimum VWHDO ($VWHDO_{min}$)

$VWHDO_{min} = 6.507 + 1.575*DM - 0.396*VWHT$ Equation 1

$R^2_{adj} = 0.47$; $p = 2.99*10^{-5}$; standard error = 0.88

where $VWHDO_{min}$ is ice-free minimum VWHDO; DM is a dummy variable representing presence (0) and absence (1) of well established dreissenid mussel populations; VWHT is mean summer (July-Sep) volume-weighted hypolimnetic water temperature.

DM and VWHT explained 47% of the variation in $VWHDO_{min}$. An increase of 1.57mg/L in $VWHDO_{min}$ would be expected at constant temperature after dreissenid mussels became well established. With every 1˚C increase in summer hypolimnetic temperature, a 0.4 mg/L decrease in $VWHDO_{min}$ would be expected. Hence, the increase in mean hypolimnetic temperature of 0.66 ˚C between 1980 and 2012 suggests that if water temperature were the only factor affecting DO, $VWHDO_{min}$ would have decreased 0.264 mg/L instead of increasing to a mean of 4.23 mg/L (Table 2). The model error is below 1 mg/L which makes this model relatively reliable when used to make decisions.

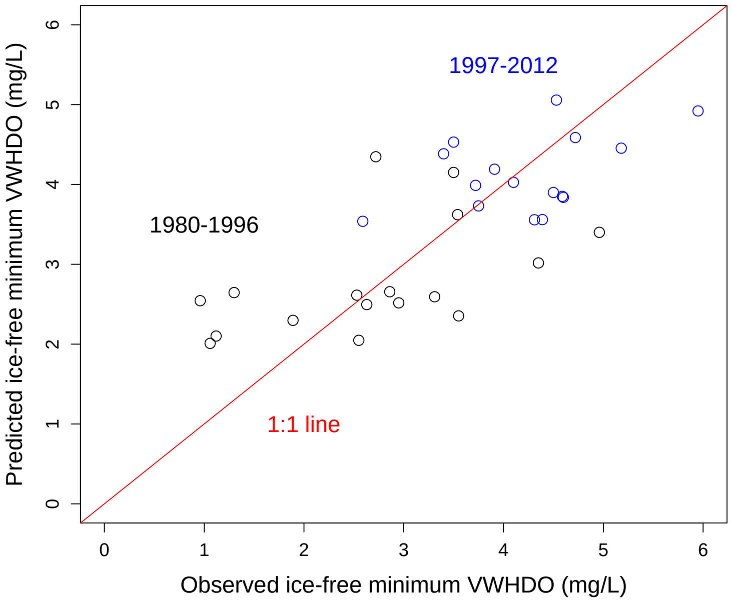

Figure 5. Comparison between predicted and observed ice-free minimum VWHDO between 1980 and 2012

2. VWHDO depletion rate (DR)

$DR = 3.731*10^{-2} + 0.859 * NH_4 + 2.332*10^{-4} * TP_{annual\ loading}$ Equation 2

$R^2_{adj} = 0.64$; $p = 3.30*10^{-4}$; standard error = 0.0074

where DR is the VWHDO depletion rate; NH_4 is ice-free mean NH_4 concentration; $TP_{annual\ loading}$ is the annual TP loading (tones/year). Note that TP loading data were only available for 1990-2006. Ice-free NH_4 concentration and $TP_{annual\ loading}$ explained 63.64% of the variation in the VWHDO depletion rate. TP loading is proxy of primary productivity in a P-limited lake. Ammonia may have contributed to DO depletion through nitrification. The model error is approximately 10% of the mean depletion rate which makes this model relatively informative.

3. VWHDO depletion period (L) from when S = 800 in the spring to the day $VWHDO_{min}$ occurred

$L = -187.231 + 0.711* L_{ts} + 0.785*VWHDO_{min\ day}$ Equation 3

$R^2_{adj} = 0.89$; $p = 1.7*10^{-15}$; standard error = 5.1

where L is VWHDO depletion period; L_{ts} is the duration of thermal stratification; $VWHDO_{min\ day}$ is day of the year that ice-free minimum VWHDO occurred.
The VWHDO depletion period is correlated significantly with duration of thermal stratification and day of the year that ice-free minimum VWHDO occurred. Longer thermal stratification and prolonged fall overturn contribute to longer depletion rate. The model error is approximately 5% of the mean depletion rate which makes this model relatively informative.

4. Spring VWHDO on day of the year 160 ($VWHDO_{160}$)

$VWHDO_{160} = 6.445 + 1.902*DM - 0.225* TP + 0.071*DAY_{ice\text{-}off}$ Equation 4

$R^2_{adj} = 0.52$; $p = 1.69*10^{-5}$; standard error = 1.1

where $VWHDO_{160}$ is the VWHDO on day of the year 160 (early June); DM is a dummy variable indicating presence (1) or absence (0) of dreissenid mussels; TP is the ice-free mean TP; $DAY_{ice\text{-}off}$ is the day the ice is considered gone.

Spring VWHDO on day of the year 160 is correlated with three variables. The two main variables (explaining 39.9% of 52.3% of the variation) were DM and TP. The presence of dreissenid mussels and lower TP contribute to higher spring VWHDO. Less importantly, earlier ice-out contributes to lower $VWHDO_{160}$ because DO depletion begins earlier. The model error is slightly larger than 1 mg/L which make this model relatively informative.

5. Initial VWHDO (when S = 800, $VWHDO_{ini}$)

$VWHDO_{ini} = 13.981 + 0.636* VWHDO_{160} - 0.061*DAY_{onset\ of\ thermal\ stratification}$

Equation 5

$R^2_{adj} = 0.99$; $p = 1.81*10^{-11}$; standard error = 0.20

where $VWHDO_{ini}$ is VWHDO when S = 800; $VWHDO_{160}$ is VWHDO on day of the year 160 (June 9); $DAY_{onset\ of\ thermal\ stratification}$ is the timing of onset of thermal stratification (day of the year). Initial VWHDO is essentially a function of how quickly thermal stratification occurs after ice-out. The model error is well below 1 mg/L which makes this model very informative.

DISCUSSION

Long-term changes in the thermal structure of Lake Simcoe have had a considerable influence on hypolimnetic DO depletion dynamics. The results indicate that climate change has offset the effects of P reduction and dreissenid mussel filtration, resulting in a lower end-of-summer VWHDO which would have been higher in the absence of a warming climate by approximately 40%. Thermal regime change in the lake between 1980 and 2012 reflects increasing annual mean air temperature of 1.59˚C at the Shanty Bay weather station over the last three decades (Table 1). The change in the epilimnetic water temperature (1.9˚C warmer), onset of stratification (20 days earlier), fall overturn (15 days later) and therefore longer duration of thermal stratification (33 days) observed at Kempenfelt Bay (station K42) over the study period is comparable to that seen in other studies (Table 3, references therein) in spite of different study periods. Epilimnetic water temperature in Lake Simcoe increased faster than the Laurentian Great Lakes and European lakes except for Lake Huron which increased 0.84 ˚C/decade during 1968-2002 (also see O'Reilly et al. 2015 for more surface water temperature comparison). On average, the duration of thermal stratification increased faster in Lake Simcoe than Lake Washington by approximately 74% (6.1 and 10.6 days/decade respectively). Thermal regime change in Lake Simcoe, a large lake, is similar to a small lake, Lake Blelham Tarn in the UK (Foley et al. 2012).

An increase in spring/initial DO has not been reported in the literature. It is not surprising that initial DO increased during the study period if the number of days between ice-off and onset of stratification decreased leaving less time to consume hypolimnetic DO. However, it is interesting to note that spring DO on June 9 has increased for the last 33 years. Several possible explanations: 1) P reduction has resulted in less organic material delivered to the hypolimnion with corresponding lower microbial respiration. 2) dreissenid mussel filtration has reduced organic material delivered to the hypolimnion. 3) A shortened ice-on period may lead to less DO consumption during winter, thus, the hypolimnion starts with higher DO (Barica and Mathias 1979). However, we do not have data for this.

DO depletion rate varied during our study period with no significant trend observed. Our DO depletion rate (range from 0.05-0.12 mg DO/L day) in Lake Simcoe over the study period is comparable to that seen in other studies (see Table 4 for detail, references therein). For example, previous study on Lake Simcoe revealed that the depletion rate in Kempemfelt Bay (combined for stations K39, K42, and K45) ranged from 0.46 mg/L day to 0.12 mg DO/L day between 1971 and 1985 (Snodgrass and Holubeshen 1987); a range of 0.13 to 0.25 mg DO/L day in Blelham Tarn, UK (1968-2008, Foley et al. 2012); observations of 0.22 and 0.23 mg DO/L day in Lake Onondaga, NY (2002 and 2004 respectively, Denkenberger et al. 2007); observations of 0.14, 0.12 and 0.11 mg DO/L day in Lake Otisco, NY (2002-2004, respectively, Denkenberger et al. 2007).The lakes mentioned above had higher DO depletion rates than Lake Simcoe except for Lake Otisco, a reference lake, because they are more eutrophic. Second, they are smaller and have higher morphometric ratios in comparison to Lake Simcoe. Nürnberg (1995) noted that higher morphometric ratio is generally linked to lakes that have a strong thermal structure and stable stratification which may promote DO depletion.

Table 3. Thermal regime changes: epilimnetic water temperature, onset of thermal stratification, fall overturn, and duration of thermal stratification in different lakes.

Lake	**Epilimnetic water/surface temperature increase (˚C per decades)**	**Earlier onset of thermal stratification (days per decades)**	**Later of termination of thermal stratification (days per decades)**	**Longer of duration of thermal stratification (days per decades)**	**Time period**	**Reference**
Lake Washington	0.16	3.9	2.2	6.1	1962-2002	Winder and Schindler 2004
Lake Superior	0.35	1.3	1.2	2.5	1906-2005	Austin and Colman 2008
Lake Superior	1.1	5			1979-2006	Austin and Colman 2007
Lake Huron	0.84				1968-2002	Dobiesz and Lester 2009
Lake Ontario	0.48				1968-2003	Dobiesz and Lester 2009
Lake Plußsee	0.4	2.63	2.89	6.32	1969-2006	Rösner et al. 2009
Lake Zurich	0.24			2.69-4.04	1947-1998	Livingstone 2003
Lake Zurich	0.29			4	1971-2005	Arvola et al. 2010
Lake Bourget	0.12	3	2.9	5.9	1976-2008	Vinçon-Leite et al. 2014
Blelham Tarn	0.27	6.83	4.39	9.27	1968-2008	Foley et al. 2012
Lake Erie	0.3				1968-2002	Burns et al. 2005
Lake Simcoe	0.57	6.06	4.55	10.61	1980-2012	Our study

Table 4. VWHDO depletion rate in different lakes: expressed as areal hypolimnetic mineralization rate (AHM) (g/m^2/day), areal hypolimnetic oxygen demand (AHOD) (mg/m^2/day), and volumetric hypolimnetic oxygen depletion rate (VHOD) (mg/L/day)

Lake	Year	AHM	AHOD	VHOD	Year, Time period	Reference
Lauerzersee		0.47		0.111		Müller et al. 2012
Rotsee		0.61		0.111		Schubert et al. 2010
Türlersee		0.6		0.086		Müller et al. 2012
Greifensee		0.71		0.062		Müller et al. 2012
Pfäffikersee		0.84		0.070		Müller et al. 2012
Murtensee		1.03		0.051		Müller et al. 2012
Hallwilersee		1.13		0.050		Müller et al. 2012
Baldeggersee		1.17		0.042		Müller et al. 2012
Lac d'Annecy		1.2		0.036		Müller et al. 2012
Sempachersee		1.32		0.034		Müller et al. 2012
Lake Geneva		1.41		0.009		Müller et al. 2012
Blelham Tarn				0.131-0.252	1968-2008	Foley et al 2012
Lake Onondaga	2002			0.199	2002 and 2004	Denkenberger et al. 2007
	2004			0.191		Denkenberger et al. 2007
Lake Otisco	2002			0.138	2002-2004	Denkenberger et al. 2007
	2003			0.116		Denkenberger et al. 2007
	2004			0.106		Denkenberger et al. 2007
Lake Onondaga	1978		1.53	0.395	1978-2002	Matthews and Effler 2006
Lake Onondaga	1979		1.52	0.393		Matthews and Effler 2006
Lake Onondaga	1981		1.59	0.411		Matthews and Effler 2006
Lake Onondaga	1982		1.48	0.382		Matthews and Effler 2006
Lake Onondaga	1985		1.49	0.385		Matthews and Effler 2006
Lake Onondaga	1986		1.68	0.434		Matthews and Effler 2006
Lake Onondaga	1987		1.29	0.333		Matthews and Effler 2006
Lake Onondaga	1988		1.32	0.341		Matthews and Effler 2006
Lake Onondaga	1989		1.41	0.364		Matthews and Effler 2006
Lake Onondaga	1990		1.02	0.264		Matthews and Effler 2006
Lake Onondaga	1991		1.12	0.289		Matthews and Effler 2006
Lake Onondaga	1992		1.18	0.305		Matthews and Effler 2006
Lake Onondaga	1994		1.12	0.289		Matthews and Effler 2006
Lake Onondaga	1995		1.26	0.326		Matthews and Effler 2006
Lake Onondaga	1996		1.05	0.271		Matthews and Effler 2006
Lake Onondaga	1997		0.73	0.189		Matthews and Effler 2006
Lake Onondaga	1998		0.74	0.191		Matthews and Effler 2006
Lake Onondaga	1999		1.05	0.271		Matthews and Effler 2006
Lake Onondaga	2000		0.72	0.186		Matthews and Effler 2006
Lake Onondaga	2001		0.66	0.171		Matthews and Effler 2006
Lake Onondaga	2002		0.77	0.199		Matthews and Effler 2006
Lake Simcoe	1971			0.046		Snodgrass and Holubeshen 1987
Lake Simcoe	1972			0.075		Snodgrass and Holubeshen 1987
Lake Simcoe	1973			0.117		Snodgrass and Holubeshen 1987

Lake Simcoe	1974	0.094	Snodgrass and Holubeshen 1987
Lake Simcoe	1975	0.093	Snodgrass and Holubeshen 1987
Lake Simcoe	1976	0.068	Snodgrass and Holubeshen 1987
Lake Simcoe	1977	0.080	Snodgrass and Holubeshen 1987
Lake Simcoe	1978	0.063	Snodgrass and Holubeshen 1987
Lake Simcoe	1979	0.080	Snodgrass and Holubeshen 1987
Lake Simcoe	1980	0.067	Snodgrass and Holubeshen 1987
Lake Simcoe	1981	0.085	Snodgrass and Holubeshen 1987
Lake Simcoe	1982	0.054	Snodgrass and Holubeshen 1987
Lake Simcoe	1983	0.079	Snodgrass and Holubeshen 1987
Lake Simcoe	1984	0.079	Snodgrass and Holubeshen 1987
Lake Simcoe	1985	0.078	Snodgrass and Holubeshen 1987
Our study			
Lake Simcoe	1980	0.052	
Lake Simcoe	1981	0.076	
Lake Simcoe	1982	0.050	
Lake Simcoe	1983	0.067	
Lake Simcoe	1984	0.092	
Lake Simcoe	1985	0.084	
Lake Simcoe	1986	0.082	
Lake Simcoe	1987	0.102	
Lake Simcoe	1988	0.076	
Lake Simcoe	1989	0.074	
Lake Simcoe	1990	0.088	
Lake Simcoe	1991	0.079	
Lake Simcoe	1992	0.098	
Lake Simcoe	1993	0.086	
Lake Simcoe	1994	0.073	
Lake Simcoe	1995	0.079	
Lake Simcoe	1996	0.079	
Lake Simcoe	1997	0.096	
Lake Simcoe	1998	0.070	
Lake Simcoe	1999	0.117	
Lake Simcoe	2000	0.063	
Lake Simcoe	2001	0.061	
Lake Simcoe	2002	0.067	
Lake Simcoe	2003	0.121	
Lake Simcoe	2004	0.089	
Lake Simcoe	2005	0.077	
Lake Simcoe	2006	0.078	
Lake Simcoe	2007	0.064	
Lake Simcoe	2008	0.077	
Lake Simcoe	2009	0.078	
Lake Simcoe	2010	0.070	
Lake Simcoe	2011	0.064	
Lake Simcoe	2012	0.064	

Using a fixed hypolimnetic volume simplified analyses because it allows the comparison of depletion rates between time periods. Following Nicholls (1997) we used 18 m as the upper ceiling in order to minimize the effect of DO replenishment during mixing events with shallower waters. However, hypolimnetic DO replenishment below 18 m was occasionally observed during stratification (see Figure 4, year 1984-1987, 1991, 1992, 1994,1998, 2000, 2003-2005 and 2010). To see if these mixing events were important, we compared effect of different hypolimnetic ceilings (18, 20, 25, 30, and 35 m) on DO parameters. Patterns in minimum VWHDO, initial VWHDO, depletion rate and depletion period (L) were similar among all hypolimnion (Figure 5). Initial DO, $\mathrm{VWHDO_{min}}$ and depletion rate decreased in both pre- and post-DM periods as the upper boundary of hypolimnion becomes deeper. The depletion period increased in both pre- and post-DM periods as the upper boundary of the hypolimnion becomes deeper (Table 5).

Investigation into the factors that affected hypolimnetic DO depletion dynamics in Lake Simcoe revealed the relative importance of various physical, chemical and biological properties. Multiple linear regression model #1 ($\mathrm{VWHDO_{min}}$) revealed that dreissenid mussels have significantly improved VWHDO by increasing $\mathrm{VWHDO_{min}}$ by 1.45 mg/L (from 1980-1996 mean of 2.87 to 1997-2012 mean of 4.32 mg/L) since they are efficient filter feeder trapped DO demanding material before reaching hypolimnion. In the absence of dreissenid mussels, $\mathrm{VWHDO_{min}}$ would have been lower post 1997 due to higher hypolimnetic temperature and longer depletion period.

Multiple linear regression model #2 (DR) revealed that ammonia has a significant influence on DO depletion rate, presumably by nitrification. Nitrification is an oxygen consumptive process where ammonia is oxidized to nitrite then to nitrate, consuming 2 mol of oxygen per mol of ammonium oxidized. Oxygen consumption due to nitrification was quantified by Small et al. (2013) and Clevinger et al. (2014) in terms of rates and proportion of oxygen consumed in Lake Superior and Lake Erie, respectively. Clevinger et al. (2014) concluded that nitrification accounted for 33% of the total oxygen consumption. Ammonia alone explained almost 40% of the variation in DO depletion rate in our analysis indicating that nitrification may account for a significant proportion of DO loss in Lake Simcoe.

Lake Simcoe was subjected to regional climate change, phosphorus reduction and invasive dreissenid mussels during the study period (1980-2012). We successfully explained the collective impact of these drivers on DO dynamics. P reductions and dreissenid mussels have had a positive impact on DO but the lengthening of thermal stratification and a slight warming of the hypolimnion led to an increase in the extent and duration of DO depletion. Continuing climate change in the region will make it more difficult to achieve the VWHDO target of 7 mg O_2/L without additional efforts to reduce P loads below 44 tonnes/yr.

Table 5. Comparison of VWHDO parameters in the pre- and post-DM periods using different criteria for upper boundary of hypolimnion

Parameters (unit)	Definition of upper boundary of hypolimnion (m)	Pre-DM period 1980-1996	Post-DM period 1997-2012	Change	Change percent
VWHDOinit (mg/L)	18	9.79	12.46	2.673	27.3%
	20	9.61	12.12	2.52	26.2%
	25	9.49	12.04	2.55	26.9%
	30	9.36	11.87	2.51	26.9%
	35	9.08	11.55	2.47	27.2%
Observed Depletion Rate (mg/L day)	18	0.079	0.078	-0.0002	-0.3%
	20	0.077	0.073	-0.004	-4.6%
	25	0.078	0.072	-0.005	-7.0%
	30	0.077	0.071	-0.006	-8.0%
	35	0.077	0.069	-0.008	-10.5%
Depletion Period (days)	18	91.05	107.13	16.08	17.7%
	20	92.05	108.69	16.64	18.1%
	25	96.64	112.69	16.05	16.6%
	30	105.58	114.32	8.73	8.3%
	35	108.23	123.57	15.34	14.2%
VWHDOmin (mg/L)	18	2.87	4.32	1.45	50.33%
	20	2.52	4.12	1.60	63.54%
	25	1.99	3.78	1.79	90.10%
	30	1.51	3.56	2.05	135.61%
	35	1.07	3.20	2.14	200.07%

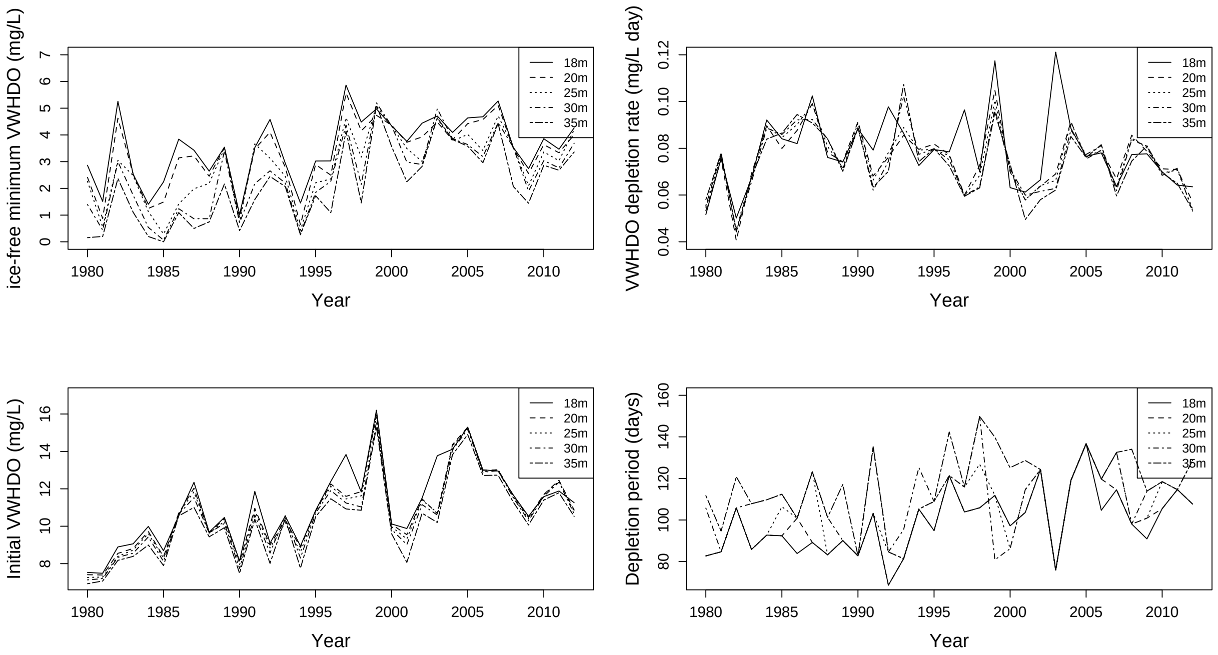

Figure 6. The impact of varying upper hypolimnetic boundaries on VWHDO parameters (18 m, 20 m, 25 m, 30 m, 35 m).

REFERENCES

Adrian, R., O'Reilly, C. M., Zagarese, H., Baines, S. B., Hessen, D. O., Keller, W., Livingstone, D. M., Sommaruga, R., Straile, D., Donk, E. V., Weyhenmeyer, G. A., & Winder, M. (2009). Lakes as sentinels of climate change. *Limnology and Oceanography*, *54* (6 part2), 2283-2297

Anneville, O., Ginot, V., & Angeli, N. (2002). Restoration of Lake Geneva: expected versus observed responses of phytoplankton to decreases in phosphorus. *Lakes & Reservoirs: Research & Management*, *7*(2), 67-80.

Barica, J., & Mathias, J. A. (1979). Oxygen depletion and winterkill risk in small prairie lakes under extended ice cover. *Journal of the Fisheries Board of Canada*, *36*(8), 980-986.

Burns, N. M. (1995). Using hypolimnetic dissolved oxygen depletion rates for monitoring lakes. *New Zealand journal of marine and freshwater research*, *29*(1), 1-11.

Caraco, N. F., Cole, J. J., Findlay, S. E., Fischer, D. T., Lampman, G. G., Pace, M. L., & Strayer, D. L. (2000). Dissolved oxygen declines in the Hudson River associated with the invasion of the zebra mussel (*Dreissena polymorpha*). *Environmental science & technology*, *34*(7), 1204-1210.

Chapra S.C. (1997) Surface Water-Quality Modeling. New York, NY: McGraw-Hill.

Clevinger, C. C., Heath, R. T., & Bade, D. L. (2014). Oxygen use by nitrification in the hypolimnion and sediments of Lake Erie. *Journal of Great Lakes Research*, *40*(1), 202-207.

Denkenberger, J. S., Driscoll, C. T., Effler, S. W., O'Donnell, D. M., & Matthews, D. A. (2007). Comparison of an urban lake targeted for rehabilitation and a reference lake based on robotic monitoring. *Lake and Reservoir Management*, *23*(1), 11-26.

Eimers, M. C., Winter, J. G., Scheider, W. A., Watmough, S. A., & Nicholls, K. H. (2005). Recent changes and patterns in the water chemistry of Lake Simcoe. *Journal of Great Lakes Research*, *31*(3), 322-332.

Evans, D. O., Nicholls, K. H., Allen, Y. C., & McMurtry, M. J. (1996). Historical land use, phosphorus loading, and loss of fish habitat in Lake Simcoe, Canada. *Canadian Journal of Fisheries and Aquatic Sciences*, *53*(S1), 194-218.

Evans, D. O., Skinner, A. J., Allen, R., & McMurtry, M. J. (2011). Invasion of zebra mussel, *Dreissena polymorpha*, in Lake Simcoe. *Journal of Great Lakes Research*, *37*, 36-45.

Ficke, A. D., Myrick, C. A., & Hansen, L. J. (2007). Potential impacts of global climate change on freshwater fisheries. *Reviews in Fish Biology and Fisheries*, *17*(4), 581-613.

Foley, B., Jones, I. D., Maberly, S. C., & Rippey, B. (2012). Long‐term changes in oxygen depletion in a small temperate lake: effects of climate change and eutrophication. *Freshwater Biology*, *57*(2), 278-289.

Futter, M. N. (2003). Patterns and trends in Southern Ontario lake ice phenology. *Environmental Monitoring and Assessment*, *88*(1-3), 431-444.

Gibson, E. S., & Fry, F. E. J. (1954). The performance of the lake trout, *Salvelinus namaycush*, at various levels of temperature and oxygen pressure. *Canadian Journal of Zoology*, *32*(3), 252-260.

Gillooly, J. F., Brown, J. H., West, G. B., Savage, V. M., & Charnov, E. L. (2001). Effects of size and temperature on metabolic rate. *science*, *293*(5538), 2248-2251.

Hawryshyn, J., Rühland, K. M., Quinlan, R., & Smol, J. P. (2012). Long-term water quality changes in a multiple-stressor system: a diatom-based paleolimnological study of Lake Simcoe (Ontario, Canada). *Canadian journal of fisheries and aquatic sciences*, *69*(1), 24-40.

Hutchinson, G.E., 1957. *A Treatise on Limnology*, Vol. I. New York, NY: John Wiley & Sons

Idso, S.B., (1973). On the concept of lake stability. *Limnology and Oceanography*, 18, 681-683.

IPCC (Intergovernmental Panel on Climate Change) Fourth Assessment Report (2013) Climate Change 2013. Available: http://www.ipcc.ch

Kendall, M. G., (1975). Rank correlation methods, 4th edn, London, UK: Cgarles Griffin

Lek, S., Delacoste, M., Baran, P., Dimopoulos, I., Lauga, J., & Aulagnier, S. (1996). Application of neural networks to modelling nonlinear relationships in ecology. *Ecological modelling*, *90*(1), 39-52.

[LSEMS] Lake Simcoe Environmental Management Strategy. 1991. Implementation Program. The predictability of hypolimnetic dissolved oxygen depletion in Lake Simcoe – Part 1; [cited July 5 2015]. Available from http://www.lsrca.on.ca/pdf/reports/lsems/dissolved_oxygen_part1.pdf

[LSEMS] Lake Simcoe Environmental Management Strategy. 2008. Lake Simcoe basin wide report. Lake Simcoe Environmental Management Strategy Report; [cited July 5 2015]. Available from
http://www.lsrca.on.ca/pdf/reports/lsems/basin_wide_report.pdf

[LSPP] Lake Simcoe Protection Plan. 2009. Ontario (Canada): Ontario Ministry of the Environment; Minitistry of Natural Resources, and Lake Simcoe Region Conservation Authority report [cited July 5 2015]. Available from: http://www.ontario.ca/environment-and-energy/lake-simcoe-protection-plan

Mahere, T. S., Mtsambiwa, M. Z., Chifamba, P. C., & Nhiwatiwa, T. (2014). Climate change impact on the limnology of Lake Kariba, Zambia–Zimbabwe. *African Journal of Aquatic Science*, *39*(2), 215-221.

Mann, H.B. (1945), Nonparametric tests against trend, *Econometrica*, 13, 245-259

Marshall, B. E., Ezekiel, C. N., Gichuki, J., Mkumbo, O. C., Sitoki, L., & Wanda, F. (2013). Has climate change disrupted stratification patterns in Lake Victoria, East Africa?. *African Journal of Aquatic Science, 38*(3), 249-253.

Martin, J.L., McCutcheon, S.C., 1999. Hydrodynamics and Transport for Water Quality Modeling. Boca Raton, FL: Lewis Publications.

Masson, S., Angeli, N., Guillard, J., & Pinel-Alloul, B. (2001). Diel vertical and horizontal distribution of crustacean zooplankton and young of the year fish in a sub-alpine lake: an approach based on high frequency sampling. *Journal of Plankton Research, 23*(10), 1041-1060.

McLeod, A. I. (2005). Kendall rank correlation and Mann-Kendall trend test. *R Package "Kendall*.

Millero, F.J., Poisson, A., 1981. *International one-atmosphere equation of state of seawater.* UNESCO Technical Papers in Marine Science. No. 36.

Moosmann, L., Gächter, R., Müller, B., & Wüest, A. (2006). Is phosphorus retention in autochthonous lake sediments controlled by oxygen or phosphorus?. *Limnology and Oceanography*, *51*(1), 763-771.

Müller, B., Bryant, L. D., Matzinger, A., & Wüest, A. (2012). Hypolimnetic oxygen depletion in eutrophic lakes. *Environmental science & technology*, *46*(18), 9964-9971.

Nicholls, K. H. (1997). A limnological basis for a Lake Simcoe phosphorus loading objective. *Lake and Reservoir Management*, *13*(3), 189-198.

North, R. L., Barton, D., Crowe, A. S., Dillon, P. J., Dolson, R. M. L., Evans, D. O., Ginn, B. K., Håkanson, L., Hawryshyn, J., Jarjanazi, H., King, J. W., La Rose, J. K. L., León, L., Lewis, C. F. M., Liddle, G. E., Lin, Z. H., Longstaffe, F. J., Macdonald, R. A., Molot, L., Ozersky, T., Palmer, M. E., Quinlan, R., Rennie, M. D., Robillard, M. M., Rodé, D., Rühland, K. M., Schwalb, A., Smol, J. P., Stainsby, E., Trumpickas, J. J., Winter, J. G., & Young, J. D. (2013). The state of Lake Simcoe (Ontario, Canada): the effects of multiple stressors on phosphorus and oxygen dynamics. *Inland Waters*, *3*(1), 51-74.

O'Connor, E.M., Aspden, L., Lembcke, D., Young, J., Stainsby, E.A., Lucchese, M. & Winter, J.G. (2013). Annual water balances and total phosphorus loads to Lake Simcoe (2007–2009). Lake Simcoe Region Conservation Authority and the Ontario Ministry of the Environment. http://www.lsrca.on.ca/pdf/reports/phosphorus_etal_2007_2009.pdf

O'Reilly, C. M., S. Sharma, D. K. Gray, S. E. Hampton, J. S. Read, R. J. Rowley, P. Schneider, J. D. Lenters, P. B. McIntyre, B. M. Kraemer, et al. (2015), Rapid and highly variable warming of lake surface waters around the globe, Geophys. Res. Lett., 42, 10,773–10,781

Osgood, R. A. 1988. Lake mixes and internal phosphorus dynamics. *Archiv für Hydrobiologie 113*, 629-638

Palmer, M. E., Winter, J. G., Young, J. D., Dillon, P. J., & Guildford, S. J. (2011). Introduction and summary of research on Lake Simcoe: research, monitoring, and restoration of a large lake and its watershed. *Journal of Great Lakes Research*, *37*, 1-6.

Pires, L. D., Bontes, B. M., Van Donk, E., & Ibelings, B. W. (2005). Grazing on colonial and filamentous, toxic and non-toxic cyanobacteria by the zebra mussel *Dreissena polymorpha*. *Journal of Plankton Research*, *27*(4), 331-339.

Plumb, J. M., & Blanchfield, P. J. (2009). Performance of temperature and dissolved oxygen criteria to predict habitat use by lake trout (*Salvelinus namaycush*). *Canadian Journal of Fisheries and Aquatic Sciences*, *66*(11), 2011-2023.

Price, P. B., & Sowers, T. (2004). Temperature dependence of metabolic rates for microbial growth, maintenance, and survival. *Proceedings of the National Academy of Sciences of the United States of America*, *101*(13), 4631-4636.

Quinlan, R., Paterson, A. M., Smol, J. P., Douglas, M. S., & Clark, B. J. (2005). Comparing different methods of calculating volume-weighted hypolimnetic oxygen (VWHO) in lakes. *Aquatic sciences*, *67*(1), 97-103.

R Development Core Team. (2015) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. Available at: http:// www.R-project.org

Read, J. S., Hamilton, D. P., Jones, I. D., Muraoka, K., Winslow, L. A., Kroiss, R., Wu, C. H., & Gaiser, E. (2011). Derivation of lake mixing and stratification indices from high-resolution lake buoy data. *Environmental Modelling & Software*, *26*(11), 1325-1336.

Rudstam, L. G., & Magnuson, J. J. (1985). Predicting the vertical distribution of fish populations: analysis of cisco, *Coregonus artedii*, and yellow perch, *Perca flavescens*. *Canadian Journal of Fisheries and Aquatic Sciences*, *42*(6), 1178-1188.

Salisbury F.B. & Ross C.W. (1978) Plant Physiology, 2nd edn. California, CA: Wadsworth Publishers.

Sharma, S. et al. A global database of lake surface temperatures collected by in situ and satellite methods from 1985–2009. Sci. Data 2:150008

Schmidt, W., (1928). Ueber temperatur and stabilitaetsverhaltnisse von seen. *Geographiska Annaler*, 10, 145-177.

Small, G. E., Bullerjahn, G. S., Sterner, R. W., Beall, B. F., Brovold, S., Finlay, J. C., McKay, M. L., & Mukherjee, M. (2013). Rates and controls of nitrification in a large oligotrophic lake. *Limnology and oceanography, 58*(1), 276-286.

Smith, V. H. (2003). Eutrophication of freshwater and coastal marine ecosystems a global problem. *Environmental Science and Pollution Research*, 10(2), 126-139.

Stainsby, E. A., Winter, J. G., Jarjanazi, H., Paterson, A. M., Evans, D. O., & Young, J. D. (2011). Changes in the thermal stability of Lake Simcoe from 1980 to 2008. *Journal of Great Lakes Research*, 37, 55-62.

Tabachnick, B. G., & Fidell, L. S. (2001). Using multivariate statistics. Boston, MA: Pearson

Winder, M., & Schindler, D. E. (2004). Climate change uncouples trophic interactions in an aquatic ecosystem. *Ecology, 85*(8), 2100-2106.

Winter, J. G., Dillon, P. J., Futter, M. N., Nicholls, K. H., Scheider, W. A., & Scott, L. D. (2002). Total phosphorus budgets and nitrogen loads: Lake Simcoe, Ontario (1990 to 1998). *Journal of Great Lakes Research*, *28*(3), 301-314.

Winter, J. G., Eimers, M. C., Dillon, P. J., Scott, L. D., Scheider, W. A., & Willox, C. C. (2007). Phosphorus inputs to Lake Simcoe from 1990 to 2003: declines in tributary loads and observations on lake water quality. *Journal of great lakes research*, *33*(2), 381-396.

Young, J. D., Winter, J. G., & Molot, L. (2011). A re-evaluation of the empirical relationships connecting dissolved oxygen and phosphorus loading after dreissenid mussel invasion in Lake Simcoe. *Journal of Great Lakes Research, 37*, 7-14.

Yu, Y. S., Zou, S., & Whittemore, D. (1993). Non-parametric trend analysis of water quality data of rivers in Kansas. *Journal of Hydrology*, *150*(1), 61-80.

Yue, S., Pilon, P., & Cavadias, G. (2002). Power of the Mann–Kendall and Spearman's rho tests for detecting monotonic trends in hydrological series. *Journal of hydrology*, *259*(1), 254-271.

CHAPTER 2:

MULTIPLE DRIVERS AFFECT A LARGE LAKE: COMPLEX INTERACTIONS AMONG CLIMATIC WARMING, PHOSPHORUS ENRICHMENT AND REDUCTION, EXOTIC DREISSENID MUSSELS, LOCAL WEATHER AND LARGE-SCALE CLIMATIC DRIVERS ALTER PHYTOPLANKTON COMPOSITION AND BIOVOLUME IN LAKE SIMCOE, ON, CANADA, 1980-2012

Jiahua Li[1], Lewis Molot[1], Michelle Palmer[2], Jennifer Winter[2], Joelle Young[2] and Eleanor Stainsby[2], Tara-Lynn Tennant[1] and Sapna Sharma[1]

[1]York University, [2]Ontario Ministry of the Environment

Keywords: multiple drivers, phytoplankton composition, dreissenid mussels, large-scale climatic cycles

SUMMARY

Phytoplankton are excellent water quality indicators as their short doubling times permit rapid responses to environmental changes. We examined the impact of thermal stability (S), water chemistry, invasive dreissenid mussels (DM) and climatic oscillation patterns on phytoplankton community composition in Lake Simcoe between 1980 and 2012. Dominant annual phytoplankton genera changed at all eight sampling stations from *Stephanodiscus* spp. (1980 to early 1990s) to *Fragilaria* spp. (later 1990s and 2000s) with intermediate dominant genera such as *Cyclotella* spp. and *Melosira* spp. Deeper sites exhibited more significant changes in dominant genera than shallower sites. Dominant phytoplankton genera differed between seasons (summer and fall) and over the years within each season. Dominant species changed significantly between seasons from *Dinobryon* spp., *Cyclotella* spp., *Ceratium* spp. and *Uroglena* spp. in summer to *Rhodomonas* spp., *Anabaena* spp., *Ceratium* spp., *Cryptomonas* spp., *Asterionella* spp. and *Tabellaria* spp. in fall at two deep sites (E51 and K42). Only one site, C1, did not show significant seasonal changes in dominant genera. Redundancy analyses found that environmental variables (e.g. S, total phosphorus, Secchi depth, silica, Total Kjeldahl nitrogen, nitrate and nitrite), DM and large climatic oscillation patterns were significantly correlated with annual phytoplankton community composition. Variation partitioning showed that environmental variables and DM individually explained little variance in phytoplankton composition but collectively explained a large portion of the variation, perhaps indicating that DM acted on phytoplankton composition indirectly by changing the environmental conditions. At some sites, large climatic oscillation indexes were also strongly correlated with phytoplankton community composition. The Quasi-Biennial Oscillation (QBO) was significantly correlated with phytoplankton community composition in shallow Cook's Bay. Most deep sites did not show significant correlation between large climatic oscillation indexes with phytoplankton community composition except for K39 in which QBO, the Pacific Decadal Oscillation, and solar sunspot number cycle were significantly correlated with phytoplankton community composition. We concluded that changes in S, nutrients, water clarity, invasive species, and large climatic oscillations collectively altered phytoplankton community composition in Lake Simcoe over the past three decades and between seasons.

INTRODUCTION

Lake Simcoe, the largest inland lake in southern Ontario outside the Great Lakes, is an invaluable natural, social and economic resource. Lake Simcoe provides drinking water to several communities in the watershed. It also assimilates wastewater from municipal water pollution control plants. In addition, the agriculture sector is worth more than CAD$500 million annually including crops and livestock (Winter et al. 2007; LSEMS 2008). Recreational activities include cottaging, boating and sport fishing, especially cold water species like lake trout, lake whitefish and lake herring, generate over CAD$200 million per year accounting for 15% (1 million angler hours per year) of all angling effort in Ontario (Eimers et al., 2005; Palmer et al. 2011; LSEMS 2008). However, Lake Simcoe and its watershed have experienced multiple pressures from increasing human activities over the past ~200 years, including logging, damming, canal construction, agriculture, urban development, nutrient enrichment, species invasion and recently climate change (Hawryshyn et al. 2012; Evans et al. 1996). The problems associated with these human-induced stressors are not independent of one another, but interact in complex ways (synergistically or antagonistically), which can result in “ecological surprises” (Christensen et al. 2006).

As the base of the aquatic food chain, phytoplankton support all higher trophic organisms in a lake. However, quantity, quality and digestibility of phytoplankton determine the growth rate, reproduction and population dynamics of higher trophic levels including zooplankton and planktivores (Gulati and Demott 1997). Extensive studies have revealed that high food quality is beneficial for energy transfer efficiency. Therefore, one can infer that altered abundance and assemblages of phytoplankton by multiple stressors will have dramatic impacts on energy transfer efficacy, food digestibility, food quantity and food quality for zooplankton and higher trophic levels. The nutritional benefits of consuming omega-3-rich phytoplankton move up the food web, making zooplankton both more efficient at converting phytoplankton biomass to their own biomass as well as much more nutritious for the zooplanktivorous fish that consume them (Brett et al. 2006). Phytoplankton produce polyunsaturated fatty acids (PUFA) which are almost exclusively synthesized by plants. PUFA play an important role in regulating cell membrane properties, serve as precursors for important animal hormones and are essential for animals (Brett and Muller-Navarra, 1997). Highly unsaturated fatty acids (HUFA), a subset of PUFA, have been found to be critical for maintaining high growth, survival and reproductive rates and high food conversion efficiencies for a wide variety of organisms (Brett and Muller-Navarra, 1997). In general, diatoms and flagellates are considered as good-quality foods because of their high eicosapentaenoic acid (EPA or omega-3 PUFA) content. On the contrary, prokaryotic cyanobacteria have a fatty-acid pattern distinct from eukaryotes and are generally considered low-quality food, having both low EPA and P content (Arnold, 1971; Lampert 1987, Wood 1974; Cobelas and Lechado 1989; Ahlgren et al. 1992). Herbivorous zooplankton growth rates are also constrained by their ability to ingest, digestibility and algal defenses against herbivores restricted zooplankton predation (DeMott and Tessier 2002; Brett and Muller-Navarra, 1997).

Phytoplankton have a short doubling time and respond to environmental changes very quickly. Therefore, as Stoermer (1978) noted several decades ago, phytoplankton assemblages can furnish a valuable integrated index of water quality conditions, which is difficult to develop by other means in a relatively large, physically complex ecosystem involving multiple stressors. Phytoplankton species composition will respond to varying environmental conditions because phytoplankton have species-specific traits such as different nutrient requirements (Tilman 1977, 1982; Tilman et al. 1981), size (Chisholm 1992), sinking velocities, ability to maintain column position (because of lipids, mucus sheaths, gas vacuoles, flagella) (Hutchinson 1967, chapter 20; Sommer 1985), and temperature optima (Robarts and Zohary 1987). These environmental gradients may be altered directly or indirectly by human-mediated disturbances such as climate change, invasive species, and eutrophication. Many studies have reported changes in phytoplankton composition over time and these changes have been typically attributed to various environmental changes (e.g., Paterson et al. 2008).

Relationships between changes in phytoplankton composition and environmental changes have been analyzed by different statistical methods, for example: redundancy analysis (RDA) (Paterson et al. 2008), canonical correspondence analysis (CCA) (Winter et al. 2011), and non-metric multidimensional scaling (NMDS) (Salmaso 1996). The proportion of variation actually explained in the trends of phytoplankton composition change has been relatively low. For example, Winter et al. (2011) investigated annual composite phytoplankton assemblages in Lake Simcoe between 1980 and 2007 and found that some of the change in species composition was related to reactive silica, total phosphorus (TP), soluble reactive phosphorus (SRP), total nitrogen (TN), water clarity (Secchi depth), and thermal changes. But much of the variance in phytoplankton composition (>70-80%) remained unexplained in their study. Thus, the relationship between phytoplankton community composition and their ambient environmental conditions over time provides a noisy and complex record of their interactions with multiple stressors/drivers at interannual and interdecadal scales. Some of the unexplained variation in phytoplankton composition may be associated with complex patterns in climatic drivers such as precipitation, temperature, wind, etc. and interactions therein that are difficult to quantify by environmental variables measured, so sunspot numbers (SSN) and large-scale climatic oscillatory cycles such as Atlantic Multidecadal Oscillation (AMO), El Niño Southern Pacific Decadal Oscillation Index (ENSO), North Atlantic Oscillation (NAO), North Pacific Index (NP), Pacific Decadal Oscillation (PDO) and Quasi-biennial Oscillation (QBO) were used to better understand change in phytoplankton composition over time.

In Lake Simcoe, several anthropogenic drivers have impacted the lake, notably, eutrophication followed by phosphorus (P) reduction, invasive species (especially dreissenid mussels) and climate change (North et al. 2013). Our objectives were to explore and test how phytoplankton composition correlated with changes in limnological characteristics, invasive species, large-scale climatic oscillatory dynamics, and climate change. Our objectives included to: 1) investigate long-term changes in seasonal and annual water quality (chemical and physical) variables 1980-2012; 2) investigate the relationship between these environmental variables and multiple external drivers such as climate change, phosphorus reduction, and dreissenid mussel invasions; and 3) examine correlations between phytoplankton species composition change and environmental changes in Lake Simcoe. We addressed specific questions raised in a previous study (Winter et al. 2011) namely: 1) Did the abundance of the diatom *Stephanodiscus* spp. peak earlier in the season than the diatom *Fragilaria* spp.? 2) Was the *Stephanodiscus* spp. peak reduced by dreissenid mussel grazing and did it result in more silica (Si) for *Fragilaria* spp. later in the season?

MATERIAL AND METHODS

Data acquisition

The Ontario Ministry of the Environment and Climate Change (OMOECC) monitored water quality in Lake Simcoe at eight stations (C1, C6, C9, E51, K39, K42, K45 and S15) from 1980 to 2008 (except for S15 which started in 1985) (Table 1, Figure 1). The stations were sampled every two weeks through the ice-free period (typically May to November). Stations C1, C9, E51, K39 and K45 were monitored 1980-2008, C6 was monitored 1980-2009, S15 was monitored 1985-2008 and K42 was monitored 1980-2012. Three sites were sampled monthly for seasonal phytoplankton community composition (C1 and K42 during 1990-2012 and E51 during 1999-2012.

Water samples were obtained through the euphotic zone using a polyvinyl chloride (PVC) hose for phytoplankton and water chemistry analysis (Nicholls and Carney 1979, detail see below).

Phytoplankton

Samples of water for phytoplankton analyses were obtained as composites of the euphotic zone by allowing a weighted 1-L bottle with a restricted inlet to fill as it was lowered and raised through a depth corresponding to twice the Secchi disc visibility. Phytoplankton samples were fixed immediately after collection with Lugol's iodine solution (Nicholls and Carney 1979). Subsamples of phytoplankton of each concentrated sample were recombined to obtain one composite sample from each station for each year (Winter et al. 2011; Hopkins and Standke, 1992). Three stations (C1, K42 and E51) were selected for seasonal composite sample counting. Samples were recombined from individual samples collected between June and August (to form summer samples) and between October and November (to form fall samples) and counted. Samples from C1 and K42 from 1990 to 2012 were counted; E51 station samples were counted between 1999 and 2012, inclusive. Older samples were partially degraded and combined seasonal results could not be matched to previous annual composite counts so older samples were excluded. Utermöhl inverted microscopy was used to count phytoplankton to mostly genus level in cell volume followed the counting method in Nicholls and Carney (1979). A minimum of 300 single cells or colonies was counted. Although identification was predominantly to genus-level, some more easily identified species were recorded while others lacking features necessary for further conclusive identification were grouped at levels above genus.

Water Quality Variables

Water chemistry parameters, alkalinity (Alk), calcium (Ca), chlorophyll *a* (Chl a), chloride (Cl^-), ammonia ($NH_4 + NH_3$), nitrate ($NO_3 + NO_2$), total Kjeldahl nitrogen (TKN is equal to ammonia + organic N), total nitrogen (TN), pH, reactive silica (Si), sulfate (SO_4) and total phosphorus (TP) were measured in the laboratory using standard OMOECC analytical methods (Janhurst, 1998).

Depth-profiled water temperature and dissolved oxygen (DO) concentrations were recorded using a YSI meter (model 6600 V2; Yellow Springs Instruments, Yellow Springs, Ohio, USA) at 1 meter intervals from surface to the bottom (Quinn et al. 2013). Secchi depth was also measured.

Local Weather and ice phenology

Air temperature and precipitation data were obtained from Environment Canada’s weather station (http://climate.weather.gc.ca/) at Shanty Bay, which is 10 km northeast of the city Barrie and close to station K42 in Kempenfelt Bay. Wind speed data in Shanty Bay were only available after 2005 and were obtained from the National Climate Data Archive of Environment Canada (http://ec.gc.ca/dccha-ahccd/default.asp?lang=en&n=71CB3873-1) for Muskoka Airport, the closest station to Lake Simcoe with long-term wind speed data for the entire period between 1980 and 2012. Ice phenology has been observed by volunteers since 1853 in Lake Simcoe. Data can be accessed online from the NatureWatch national volunteer monitoring program (https://www.naturewatch.ca/icewatch/).

Time series for indices of Sunspot Numbers and large-scale climatic drivers

Time series for SSN and the large-scale climatic drivers were acquired from various databases (Table 6). AMO, El Niño/La Niña, QBO and PDO were acquired from the U.S. National Oceanic and Atmospheric Administration (NOAA); SSN was acquired from the U.S. National Aeronautics and Space Administration (NASA); NP was acquired from the U.S. National Center for Atmospheric Research (NCAR). We used mean annual values, ice-free mean (April-November), summer mean (June-August) and fall mean (September-November) values for all indices.

Table 6. Data Sources of indices of sunspot number and large-scale climatic oscillatory indices

Time series data	Abbr.	Record Length	Source	Website
Atlantic Multidecadal Oscillation Index	AMO	1856-2015	NOAA	http://www.esrl.noaa.gov/psd/data/correlation/amon.us.long.data
El Nino Southern Oscillation Index	ENSO	1950-2015	NOAA	http://www.cpc.ncep.noaa.gov/products/analysis_monitoring/ensostuff/detrend.nino34.ascii.txt
North Atlantic Oscillation Index	NAO	1950-2015	NOAA	http://www.cpc.ncep.noaa.gov/products/precip/CWlink/pna/norm.nao.monthly.b5001.current.ascii
North Pacific Index	NP	1900-2015	UCAR	https://climatedataguide.ucar.edu/sites/default/files/climate_index_files/npindex_monthly.ascii
Pacific Decadal Oscillation Index	PDO	1948-2015	NOAA	http://www.esrl.noaa.gov/psd/data/correlation/pdo.data
Quasi-biennial Oscillation	QBO	1948-2015	NOAA	http://www.esrl.noaa.gov/psd/data/correlation/qbo.data
Sunspot Numbers	SSN	1749-2015	NASA	http://solarscience.msfc.nasa.gov/greenwch/spot_num.txt

Data manipulation

Our data required several transformations to satisfy the assumptions of statistical analyses. Water quality variables exhibiting high levels of multicollinearity were removed prior to analyses as high multicollinearity between variables may reduce the ability to correctly quantify model parameters (Legendre and Legendre 1998). Si, Ca, NH_4, Cl, Secchi, pH and temperature were log-transformed to increase linearity of relationships and minimize the impact of extreme values. Hellinger transformation was applied to phytoplankton community composition data, which decreased the importance of the most abundant species (Legendre and Gallagher 2001). Furthermore, only annual composite and seasonal composite phytoplankton species which made up >2% of the total biovolume were used because rare species can have a disproportionate effect on multivariate analyses as they contribute little or nothing to understanding general community relationships (Jackson and Harvey 1989).

Data analyses

Schmidt's thermal stability (S) was calculated (Idso, 1973) and duration of thermal stratification was calculated as in Stainsby et al. (2011). Mean S was calculated for June to September (thermal stratification period). Water chemistry and physical data were pooled as necessary to calculate ice-free, summer, and fall means. S was calculated using the "schmidt.stability" function in" rLakeAnalyzer" package (Brentrup et al. 2014 and Read et al. 2011).

Statistical Analyses

A statistical framework was devised to meet our objectives. First, the non-parametric Mann-Kendall trend test (Mann 1945, Kendall 1975, Yu et al. 1993, Yue et al. 2002) was used to test whether there were statistically significant long-term monotonic trends over time in ice phenology (on and off dates), physical variables, water chemistry variables, thermal stability (S), volume-weighted epilimnetic dissolved oxygen (VWEDO) and climate variables over the study period. All variables are shown. We used the "Kendall" function (McLeod 2013) in R for the Mann-Kendall trend test.

Second, to explore associations among physical, chemical and biological variables, principal component analysis (PCA) was performed in R using the "rda" function in "vegan" package. The first component describes the major environmental gradient and the secondary environmental gradient was indicated by the secondary principal component. Thus, a number of derived components used as surrogate measures of environmental characteristics were. Cluster analysis (Johnson et al. 1992) was also used to explore similarity of environmental conditions among years and sites. We used "hclust" function based on ward hierarchical clustering in R for cluster analysis.

Third, to determine statistically significant relationships between environmental factors (physical, chemical, and biological factors described above) and phytoplankton community composition, redundancy analysis (RDA) was used. RDA model based on forward selection were performed in R using "rda" in "vegan" package and "forward.sel" functions in "packfor" package. RDA is a direct gradient analysis technique that partitions variation in community composition into components associated with multiple predictor variables. The response variables in a RDA can be expressed as a linear combination of the predictor variables, and the approach handles species abundance data, sample years and environmental data simultaneously (Borcard et al., 1992). Canonical correspondence analysis (CCA) has been used in previous studies because the relative length of the gradient representing the variation in phytoplankton data was a unimodal response model (Winter et al. 2011; ter Braak and Prentice, 1988). However, this method was not used because there was no adjustment available to account for inflated R^2 (explained variance, Peres-Neto et al. 2006).

Subsequently, given the assumption of the absence of temporal autocorrelation, another RDA model was used to relate phytoplankton composition to significant environmental variables (e.g. TP, DM, and thermal stability (S)) and indices of sunspots numbers and large-scale climatic oscillations (e.g. SSN, AMO and PDO). Four climatic oscillations were explored in this study. (1) The Quasi-Biennial Oscillation (QBO) represents equatorial stratospheric zonal winds and is believed to have a period of approximately 28 months (Baldwin et al. 2001). (2) The El Niño Southern/Oscillation Index (ENSO) is characterized by two main states: El Niño events defined by positive sea surface water temperature anomalies in the eastern tropical Pacific Ocean and La Niña events marked by cooler surface temperatures in the same region (Stuecker et al. 2013) and is hypothesized to have a period of 2-7 years, with an average of 4 years (MacMynowski and Tziperman 2008). Abrupt frequency shifts of the ENSO mode from 57 months (1943-1961) to 39 months (1963-1980) in period (Yiou et al. 2000) have been observed. (3) The North Atlantic Oscillation (NAO) has significant peaks at inter-annual and inter-decadal periods (Hurrell and van Loon 1997; Higuchi et al. 1999). For example, the NAO index has significant spectral peaks at 2– 3 years, 6–10 years, 20 years and 60 years (Hurrell and van Loon 1997; Higuchi et al. 1999). In addition, Huang et al. (1998) analyzed the spectral relationship between the NAO and ENSO and found significant coherence at 2–4 year and 5–6 year periods (Huang et al. 1998). (4) The solar sunspot cycle reflecting the amount of solar-magnetic activity on the sun was also explored as a driver. The average length of the sunspot cycle has been identified as approximately 11 years (Friis-Christensen and Lassen 1991), although there is variation in cycle length between 9 and 12 years (Lee et al. 1995), particularly in the latter half of the 20th century (Friis-Christensen and Lassen 1991).

In parallel to the development of the RDA model, we performed a time-frequency analysis using Moran's Eigenvector Maps (MEM) formerly known as Principal Coordinate of Neighbour Matrices (PCNM) to identify important temporal cycles in the phytoplankton composition record because of the possibility that interactions among more than one oscillating driver might create a temporally unique oscillation. MEMs quantify spatial structure (Dray et al., 2006; Griffith and Peres-Neto, 2006) and have been extended to quantify temporal structure (Sharma et al. 2013). The MEM approach is preferred to classical time series analyses in our study because it generates a series of sine waves with decreasing period that are orthogonal to one another and can be used as independent variables in a subsequent analysis, such as linear regression, redundancy analysis or a variation partitioning framework. The first MEM variable represents broad temporal cycles, and subsequent MEM variables represent cycles with decreasing period (Borcard and Legendre 2002, Sharma et al. 2013). This allows us to quantify the variation explained by each cycle and quantify its contribution to phytoplankton assemblage change. More specifically, for example, it converts a time vector comprised of 29 time steps (29 sampling years from 1980 to 2008) into a series of MEM variables (2/3 of them will be positive and used in following analysis), that is, 19 sine waves with different frequencies (see Borcard and Legendre 2002, Borcard et al. 2004 for detail). Through a forward selection procedure with double stopping criteria, only significant MEM variables and environmental variables are selected and used as explanatory variables in the final model (Blanchet et al. 2008). The RDA models either contain indices of sunspots numbers and large-scale climatic oscillations or MEM variables were compared and compared using biological knowledge. The best models were then selected as the final models.

Lastly, variation partitioning (Borcard et al. 1992) was used after the analyses above. It helps us to test and determine the likelihood of these sets of predictors (environmental variables, SSN, large-scale climatic drivers and temporal oscillations generated by MEM) in explaining patterns in phytoplankton community structure. Environmental variables and oscillatory dynamics were used in RDA. We investigated the unique and shared contributions of each portion of explanatory variables in explaining variance of phytoplankton composition. We attributed specific environmental variables that significantly explained unique variance to the impact of changes in environmental conditions on phytoplankton composition. We did not ascribe oscillations acting at inter-annual or inter-decadal scales to specific large-scale climatic drivers as frequency bands are not specific to individual large-scale climatic drivers owing to interactions among themselves and overlapping spectral signals for various large-scale climatic drivers. Large-scale climatic oscillations act as complex climatic drivers of changes in precipitation, temperature, wind and etc. that are difficult to quantify by other environmental variables but may play an important role in phytoplankton composition.

RESULTS

Trends in Environmental Variables

Mean ice-free season water quality variables were examined and tested for monotonic trends (Figure 6) for K42 from 1980 to 2012. All sites usually exhibited similar trends in each water quality variable although there were some differences (Table 7). Chlorophyll *a* decreased significantly at all sites. Chloride increased significantly due to higher rates of road salt application associated with urban development; the pH decrease after 2005 co-occurred with lower alkalinity but the pH decrease may have also been due to lower productivity (resulting in higher free CO_2). The increase in reactive silica after 1996 may have been due to intensive dreissenid mussel grazing on diatoms caused by lower diatom productivity or release of digested Si; the increase in Secchi depth may also have been due to the filtration effect of dreissenid mussels. There were no significant long-term trends in nitrogen and total phosphorus. However, Winter et al. (2011) reported that soluble reactive phosphorus (SRP) decreased between 1980-1982 and 2004-2007 at all stations. Relatively high values of NH_4 and TKN were observed in 1984. The increase in TKN is much higher than the increase in ammonia and must therefore reflect a large increase in organic N. We do not have any explanation for this anomalous year but it could reflect unusually high N loading. TP decreased significantly at C1 and C6 but not the other sites probably because shallow sites react to TP loading reduction more quickly than the deeper counterparts (REFS).

PCA and cluster analysis of water quality variables (K42) demonstrated sulfate and pH were high and clustered during 1980 to 1985 while volume weighted epilimnetic temperature (VWET), Secchi, DM, Cl and Ca were high and clustered in the period 2000-2008 (post dreissenid invasion) (Figure 7). The water quality variables mentioned above contributed mainly to PCA axis 1. NH_4, TKN (which is mostly dissolved and particulate organic N), TP, NO_3 and chlorophyll *a* contributed mainly to PCA axis 2.

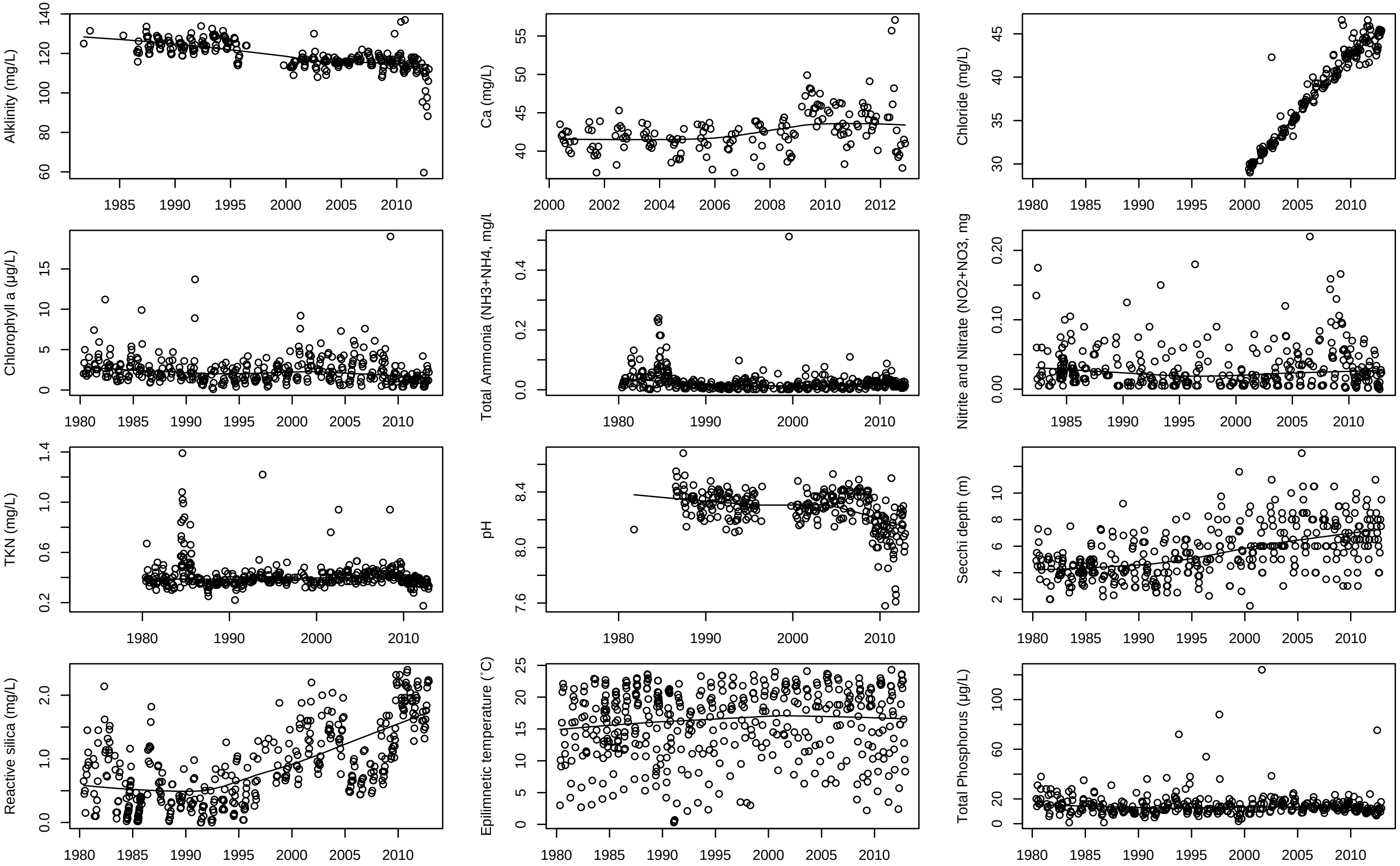

Figure 7. Trends in limnological characteristics at K42 from 1980-2012. A Loess curve (solid line) is fitted.

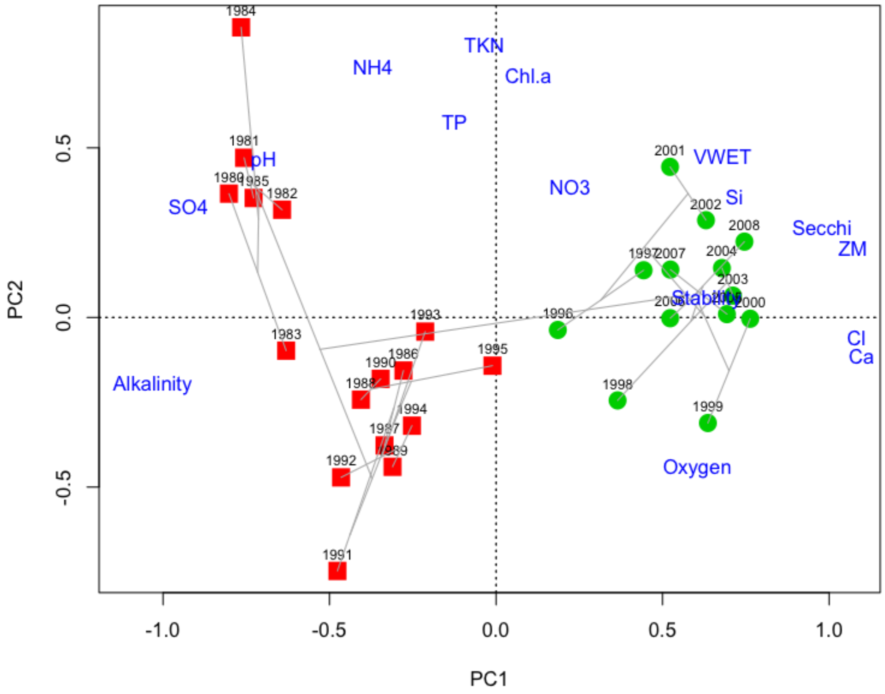

Figure 8. Principal components analysis and cluster analysis of water quality variables and dreissenid mussels.

Table 7. Long-term monotonic trends (Mann-Kendall test) of water quality variables between1980 and 2012 and *tau* statistics reported below and significance level was indicated by *. S is ice-free mean Schmidt's stability; T is ice-free mean epilimnetic temperature.

	C1	**C6**	**C9**	**E51**	**K39**	**K42**	**K45**	**S15**
Alk	-0.22*	-0.45*	-0.48	-0.53	-0.50	-0.49	-0.48	-0.52
Ca	0.10	0.19*	0.21*	0.19*	0.29*	0.25*	0.26*	0.15*
Chla	-0.27*	-0.25*	-0.23*	-0.25*	-0.20*	-0.14*	-0.10*	-0.10*
Cl	0.64*	0.80*	0.87*	0.88*	0.85*	0.85*	0.89*	0.88*
NH4	0.16*	0.02	0.01	0.07	0.08	0.12*	0.13*	0.02
NO3	-0.16*	-0.14*	-0.09	-0.16*	0.08	-0.04	-0.23*	-0.16*
pH	-0.16*	-0.27*	-0.35*	-0.36*	-0.27*	-0.30*	-0.33*	-0.35*
S	0.20*	0.46*	0.43*	0.51*	0.37*	0.38*	0.37*	0.37*
Secchi	0.20*	0.46*	0.43*	0.51*	0.37*	0.38*	0.37*	0.37*
Si	0.39*	0.48*	0.51*	0.48*	0.42*	0.45*	0.48*	0.55*
T	0.04	0.05	0.06	0.03	0.09*	0.07	0.09*	-0.02
TKN	0.01	0.00	-0.07	0.08	0.05	-0.03	-0.07	0.06
TP	-0.16*	-0.12*	-0.07	0.03	-0.07	-0.06	-0.07	0.08

* $p<0.01$ significant monotonic trend

Relationships between phytoplankton and environmental variables:

1. Phytoplankton annual composite results

Comparison of annual values among sites

Redundancy analysis triplots (Figures 8 and 9) describe the association between annual phytoplankton community composition and their environments at eight sampling stations in Lake Simcoe between 1980 and 2008 (C6 was also sampled in 2009 and S15 sampling began in 1985). Phytoplankton genera are represented by genus names in red. Lake characteristics and climate variables are represented by arrows in blue. Environmental variables used are as follows: dreissenid mussels, reactive silicate, Secchi depth, total phosphorus, ammonia, and epilimnetic water temperature. Note that the data points (x – y coordinates representing RDA1 and RDA2) in Figures 8 and 9 are the same points but are colour coded differently: in Figure 8 the points are coded green and red to represent presence and absence of dreissenids and in Figure 9 the data points are coded instead with a unique colour for each station.

Environmental variables described 36% of the variation in phytoplankton communities in Lake Simcoe ($R^2_{adj} = 0.36$). The relationship between phytoplankton communities and their environments was significant ($p < 0.05$). Dominant genera changed from *Stephanodiscus* spp. to *Fragilaria* spp. The timing of the change coincided with the timing of the dreissenid mussel invasion. *Stephanodiscus* spp. was the most dominant genus between 1980 and 1996 decreasing significantly thereafter. It can be seen from Figures 8 and 9, that the increases in silica, water transparency, water temperature and presence of dreissenid mussels (post 1996) were negatively correlated with *Stephanodiscus* spp. abundance. However, two cryptophyte genera (*Cryptomonas* spp. and *Rhodomonas* spp.) and four diatom genera (*Fragilaria* spp., *Cyclotella* spp., *Asterionella* spp., and *Tabellaria* spp.) were positively associated with these changes. The period before 1996 was characterized by absence of dreissenids and higher nutrient (TP and NH_3) levels. *Melosira* spp. (diatom), *Ceratium* spp. (dinoflagellates), *Anabaena* spp. (cyanobacteria), and Protozoa (single-celled animals) were associated with higher concentrations of total phosphorus and ammonia. Variation partitioning as a follow up analysis revealed a large portion of variance (23% out of 36%) was explained by covariates (shared area in Venn diagram, Figure 10) indicating that the dreissenid mussel impact on phytoplankton composition was partially due to their ability to change the ambient environment.

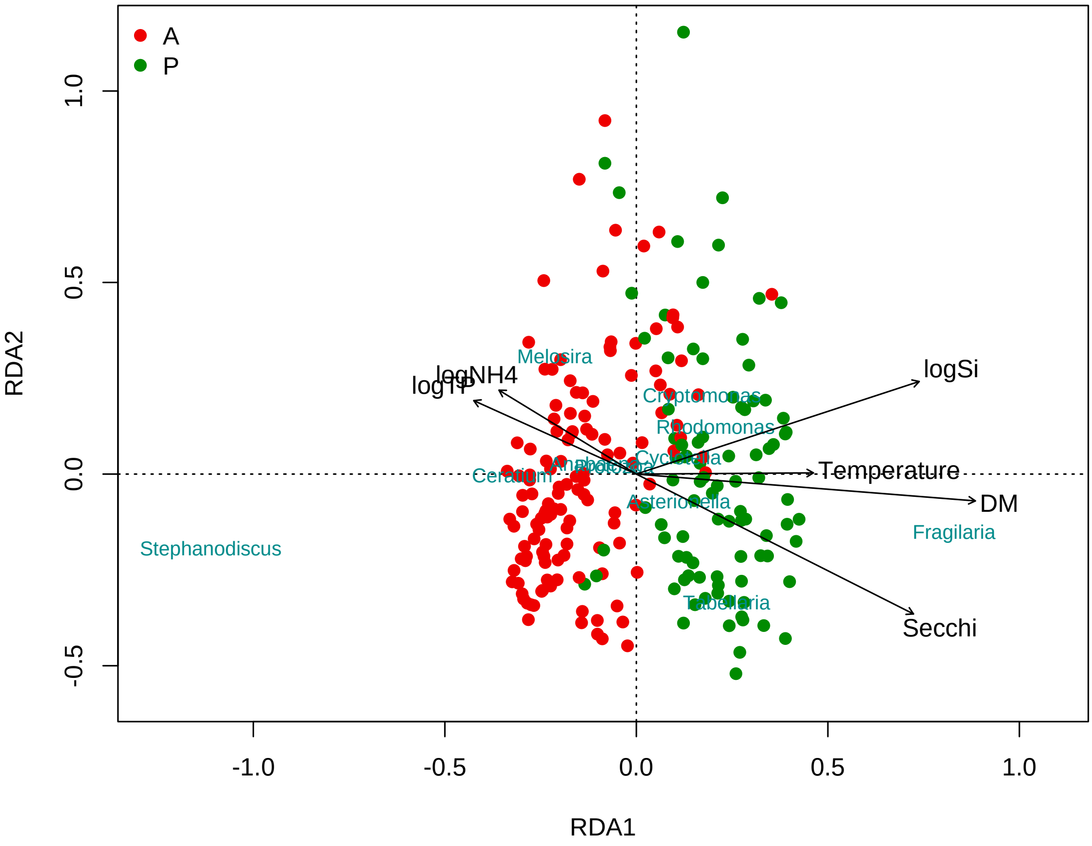

Figure 9. Ordination plot of the redundancy analysis (RDA) on the relationship between annual mean environmental parameters (vectors) and annual composite phytoplankton community composition in all lake stations (C1, C6, C9, E51, K39, K42, K45, S15) during 1980-2008. Observations are classified into pre- and post-dreissenid mussel invasion: Absence (A) and Presence (P).

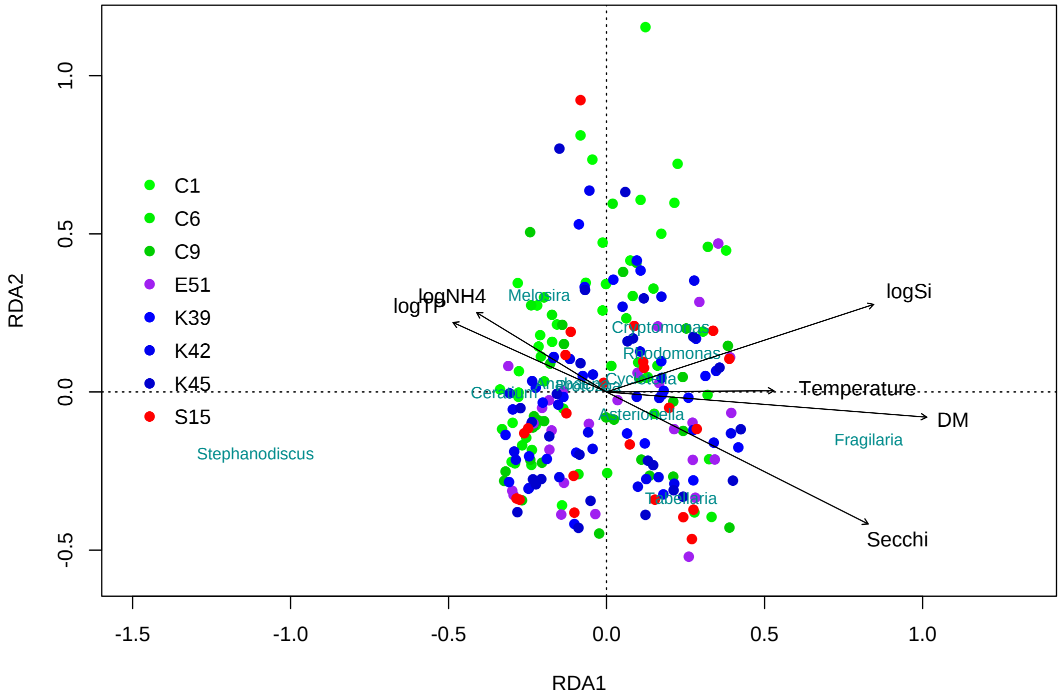

Figure 10. Ordination plot of the redundancy analysis (RDA) on the relationship between annual mean environmental parameters (vectors) and annual composite phytoplankton community composition at all lake stations (C1, C6, C9, E51, K39, K42, K45, S15) during 1980-2008. Observations are classified by sites using different colours (shown in figure top left).

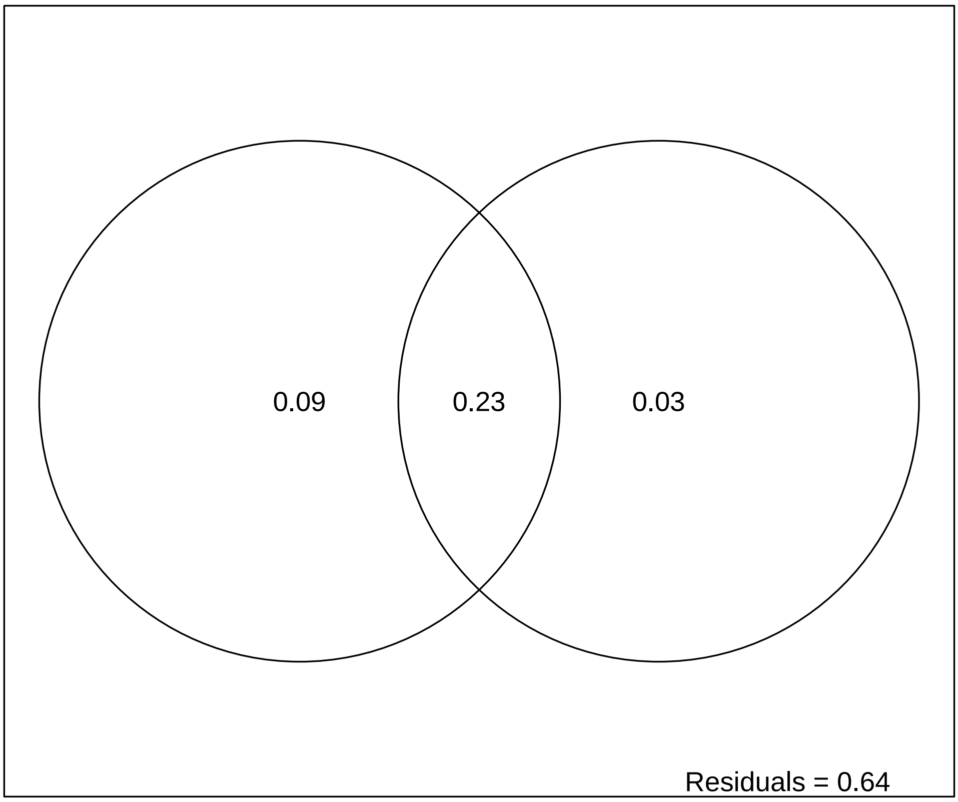

Figure 11. Variation partitioning of all annual RDA results with 8 sites combined (C1, C6, C9, E51, K39, K42, K45, S15). Circle on the left indicates variance explained by water quality variables; circle on the right indicates variance explained by dreissenid mussels. The rectangular indicates total variance in phytoplankton community composition.

Individual station annual analysis

An example (K42) of annual phytoplankton analysis is shown in two redundancy analysis triplots (Figures 11 and 12). They revealed significant relationships ($p<0.05$) between annual phytoplankton community composition and limnological characteristics, local weather, and indices of large climatic oscillations. 4, 5 and 7-year temporal oscillations in phytoplankton composition were also identified by RDA (Figures 11 and 12). Abundant genera in the 1980s to early 1990s at station K42 were *Stephanodiscus* spp., *Coelosphaerium* spp., *Ceratium* spp., and *Melosira* spp. In the late 1990s and post 2000, *Fragilaria* spp., *Asterionella* spp. and *Tabellaria* spp. were abundant. The dominant genus shift from *Stephanodiscus spp.* to *Fragilaria spp.* was correlated with increasing Secchi, Schmidt's stability, reactive silica and dreissenid mussel invasion. TP was positively correlated with *Cyclotella* spp. (diatom), *Cryptomonas* spp. (cryptomonad) and *Ceratium* spp. (dinoflagellate) and negatively correlated with *Asterionella* spp. and *Tabellaria* spp. (diatoms). Overall, limnological characteristics explained 32% of the variation in phytoplankton composition at station K42. Si, TP, stability, Secchi depth and dreissenid mussels were identified as significant limnological characteristics. Variation partitioning of the RDA model is shown in a Venn diagram (Figure 13). The rectangle is the total variance of response variables (i.e. phytoplankton composition); circles are explanatory variables (i.e. environmental conditions, dreissenid mussels and temporal patterns). Dreissenid mussels and environmental variables uniquely explained very little variance of phytoplankton composition (about 3% each) but collectively, 28% was explained indicating that dreissenid mussels probably act on phytoplankton composition either directly by grazing or indirectly by changing environmental conditions. For example, increases in Si and water clarity, and decreased availability of nutrients were associated with the dreissenid invasion after 1996.

Significant temporal oscillatory variables (i.e. 4-, 5-, 7-year cycles generated by MEM) were added into the RDA analysis and an additional 8% of the variation was explained (Figure 13) indicating some large-scale climatic cycles may have acted on phytoplankton composition. Large-scale climatic oscillations include 2-3 year cycles in the Quasi-Biennial Oscillation, the El Niño Southern Oscillation which peaks on average every 2-7 years with an average of 4 years, the North Atlantic Oscillation which has peaks at 2-3, 6-10 and 20 years, and the 9-12 year sunspot cycle. While they may play an important role in phytoplankton composition, we cannot ascribe directly which large-scale climatic driver is responsible in each defined period owing to interactions among themselves and overlapping spectral signals of various large-scale climatic drivers.

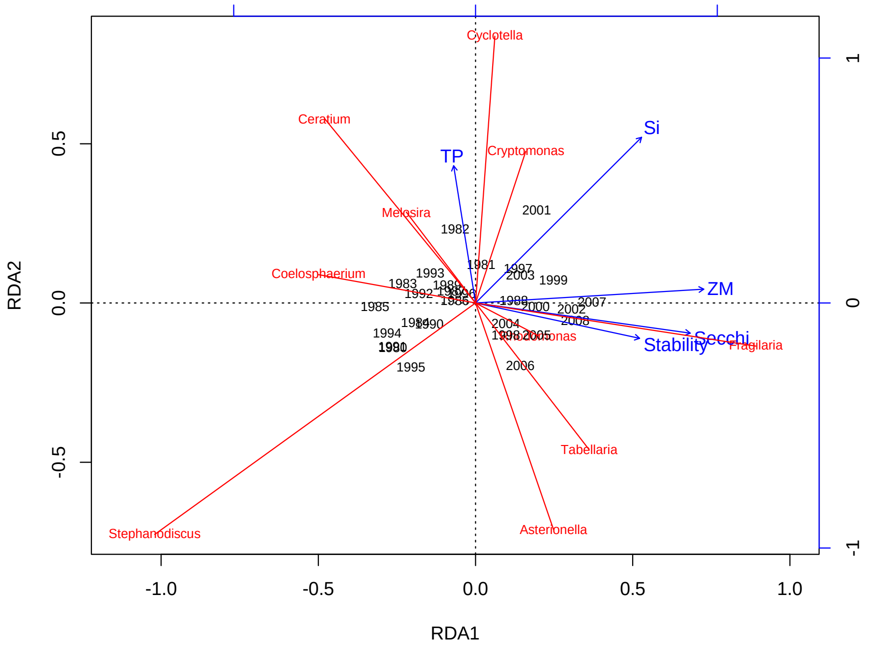

Figure 12. Ordination plot of the redundancy analysis (RDA) on the relationship between environmental parameters (vectors) and annual phytoplankton community composition at **K42** during 1980-2012.

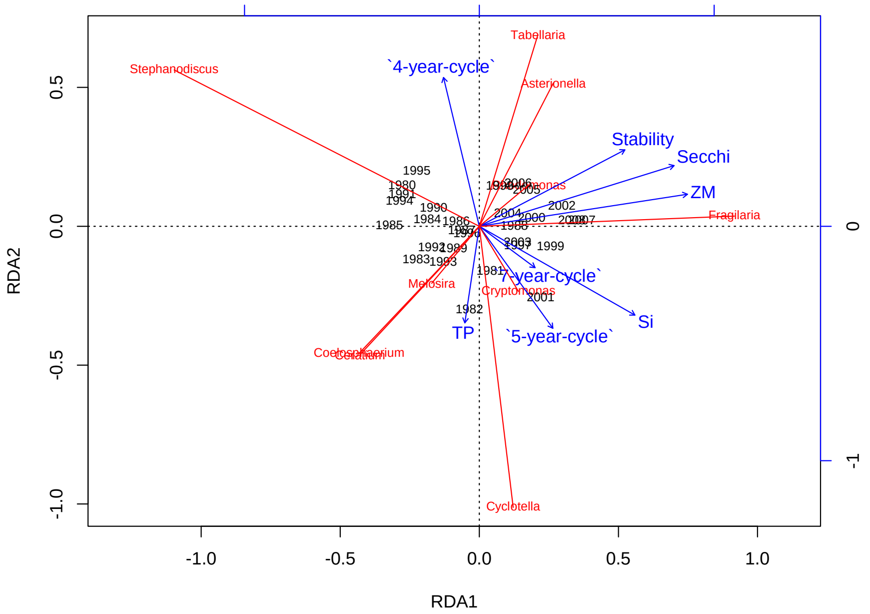

Figure 13. Ordination plot of the redundancy analysis (RDA) on the relationship between environmental parameters, temporal oscillations (vectors) and annual phytoplankton community composition at **K42** during 1980-2012.

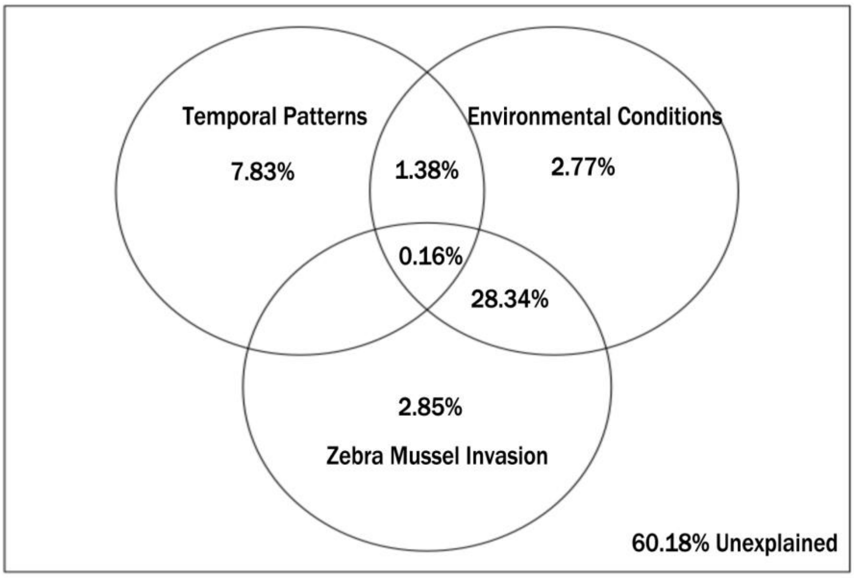

Figure 14. Venn diagram showing variation partitioning among environmental variables, dreissenid mussel invasion, and temporal oscillations in structuring annual phytoplankton community composition.

Table 8 summarizes the variation (R^2_{adj}) explained in each portion of the variables. On average, 45% of variance in annual phytoplankton composition was captured by environmental variables, dreissenid mussels and temporal oscillations. R^2_{adj} was highest in Cook’s Bay (C1-C3) and unique contributions of temporal and environmental variables were highest in Cook’s Bay. The unique contribution of dreissenid mussels was highest in the main basin and lowest in Kempenfelt Bay. Temporal oscillations alone on average, contributed to 17% of the variation explained which is the most among the three types of explanatory variables. Collectively, dreissenid mussel and environmental variables explained much more variation in Kempenfelt Bay (20.5%) than in Cook’s Bay and the Main Basin (4.1 to 4.5%) indicating that dreissenid mussels may have indirectly impacted phytoplankton in Kempenfelt Bay by altering water chemistry rather than by grazing

Table 8. Summary of variation (R^2_{adj}) of annual phytoplankton composition explained by environmental variables (‘Env’), dreissenid mussels (‘DM’) and temporal oscillation cycles (‘Temp’). Interacting factors are shown by ‘∩’.

Site	Total	Temp	Env	DM	Env∩DM	Temp∩DM	Temp∩Env	env∩DM∩temp
C1	49.9%	12.5%	20.5%	5.1%	11.6%	0.00%	3.2%	0.00%
C6	46.6%	31.1%	18.3%	16.1%	0.84%	0.00%	0.00%	9.7%
C9	54.1%	28.6%	24.8%	4.2%	0.00%	1.59%	0.00%	10.5%
E51	56.0%	22.8%	13.6%	3.0%	13.5%	0.00%	0.00%	7.5%
K39	44.1%	16.8%	7.0%	1.6%	12.7%	1.30%	0.00%	6.0%
K42	39.8%	7.8%	2.8%	2.9%	28.3%	0.00%	1.4%	0.16%
K45	32.6%	7.2%	3.2%	26.1%	0.14%	0.00%	0.00%	0.88%
S15	35.2%	10.8%	3.0%	8.8%	0.00%	13.8%	0.00%	0.00%
Cook's Bay	50.2%	24.1%	21.2%	8.49%	4.15%	0.53%	1.1%	6.8%
Kempenfelt Bay	42.0%	12.3%	4.9%	2.25%	20.53%	0.65%	0.69%	3.1%
Main Bay	41.3%	13.6%	6.6%	12.63%	4.54%	4.6%	0.00%	2.8%
Site average	44.8%	17.19%	11.64%	8.48%	8.39%	2.08%	0.57%	4.4%
Lake average based on basins	44.5%	16.7%	10.9%	7.8%	9.7%	1.92%	0.58%	4.2%

Table 9. Yearly composite phytoplankton biovolumes (mm^3/m^3)

	C1	C6	C9	E51	K39	K42	K45	S15
1980	1646	1164	781	841	1559	1055	724	
1981	1517	1201	777	440	873	2074		
1982	1081	1276	780	248	696	373	217	
1983	1297	1752	814	501	316	521	460	
1984	1445	1537	912	444	514	274	1010	533
1985	2260	2139	1307	600	1587	704	1941	390
1986	2160	874	550	189	397	869	239	266
1987	737	572	571	161	314	271	231	310
1988	609	920	1052	355	331	312	338	397
1989	622	510	450	343	272	342	239	250
1990	1181	809	831	579	824	340	976	1153
1991	793	840	554	463	461	718	365	487
1992	1037	482	616	487	513	309	333	283
1993	950	1138	818	237	495	418	530	490
1994	1476	1544	1129	595	727	459	576	839
1995	615	669	359	304	257	600	499	400
1996	1079	424	426	205	286	343	310	346
1997	153	411	463	245	811	210	325	331
1998	410	344	316	184	248	450	256	194
1999	175	362	356	167	337	231	307	283
2000	168	336	304	203	289	394	262	251
2001	335	161	349	146	360	286	300	227
2002	208	506	517	189	284	299	270	208
2003						305		
2004	998	895	575	676	552	523	631	470
2005	287	458	471	353	571	515	379	410
2006	1451	394	556	398	538	547	447	531

2007	583	981	595	350	501	547	597	543
2008	467	538	473	375	1109	719	645	561
2009		458				377		
2010						748		
2011						434		
2012						378		

2. Phytoplankton seasonal composite results

2.1 Comparison of stations, season and pre- and post-DM

The RDA results describing seasonal phytoplankton composition in relation with multiple environmental gradients (in summer and fall) at C1, E51 and K42 (analyzed together) are displayed with a series of ordination plots. Seasonal phytoplankton community composition was assessed in three perspectives (Figures 14-16). The phytoplankton composition data from three stations for each season were plotted and colour coded three different ways by 1) different sites (C1, E51 and K42, Fig. 14), 2) presence and absence of invasive dreissenid mussels (Fig. 15) and 3) by season (summer and fall, Fig. 16). Thus, the distributions of points in Figures 14-16 are identical in each graph but the colour coding differs. The analyses of phytoplankton composition show a clear separation between shallow and deep water sites, between pre- and post-invasion of dreissenids and between summer and fall seasons.

C1 in both seasons had, in general, different phytoplankton community composition than E51 and K42 during the study period 1990-2012 or 1999-2012 (Figure 14). For instance, *Melosira* spp., *Aphanizomenon* spp., *Coelastrum* spp. and *Scenedesmus* spp. were present in high abundance at C1 but *Fragilaria* spp. and *Tabellaria* spp. occurred in high abundance at E51 and K42. The deeper E51 and K42 sites were characterized by higher water clarity and greater water column thermal stability than shallow C1. Chlorophyll *a* concentration was higher at C1 than in the other two sites. The environmental conditions mentioned above were associated with RDA axis 1.

Chloride, water temperature, calcium, pH, NO_3, and alkalinity were associated with RDA axis 2 which separated pre- and post- dreissenid mussel invasion periods (Figure 15), though unequal numbers of observations were reported in the RDA triplot.

Summer phytoplankton community was dominated by large *Stephanodiscus* spp. and *Ceratium* spp. and fall phytoplankton community was dominated by *Rhodomonas* spp. and *Cyclotella* spp. (Figure 16). Chloride, water temperature, calcium, pH, NO_3, and alkalinity were associated with the separation between summer and fall. Alkalinity, pH, and NO_3 were higher in the summer than the fall. Chloride, water temperature, and calcium were higher mostly in the fall. It is possible that grazing pressure on large *Stephanodiscus* spp. and *Ceratium* spp. by *Dreissena* spp. existed but there is no experimental evidence. More detailed individual site seasonal and pre- and post- dreissenid period comparisons are shown in the next section.

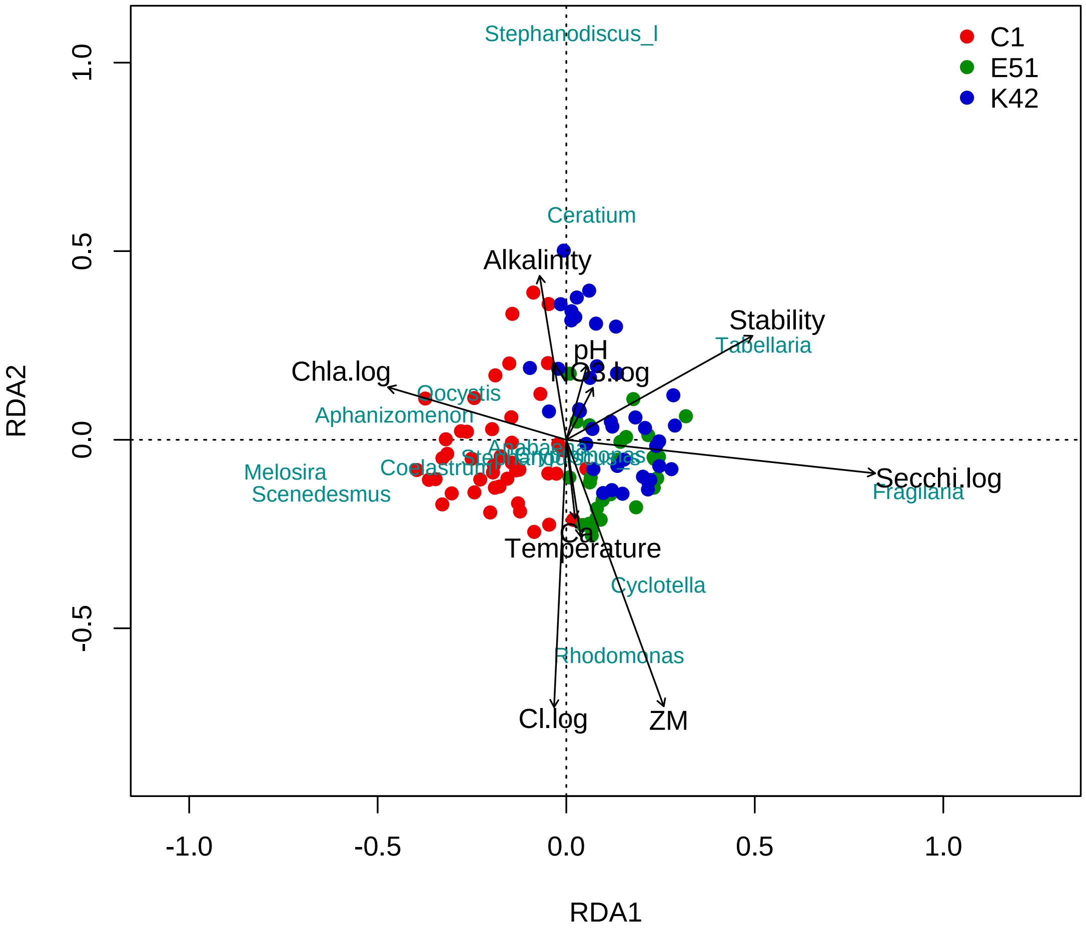

Figure 15. Ordination plot of the redundancy analysis (RDA) of the relationship between environmental parameters (vectors) and phytoplankton community composition during 1990-2012 for C1 and K42 and 2000-2012 for E51. Observations are classified by site.

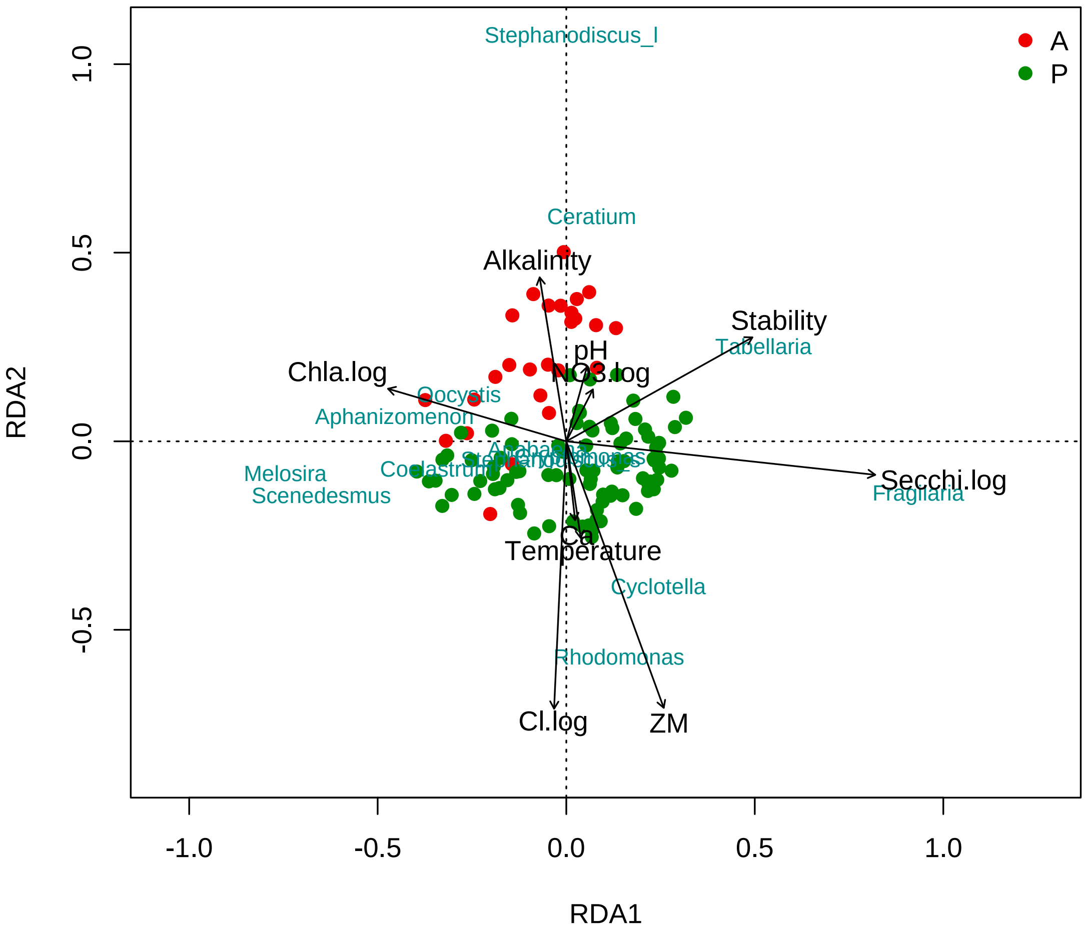

Figure 16. Ordination plot of the redundancy analysis (RDA) of the relationship between environmental parameters (vectors) and phytoplankton community composition in C1, E51, and K42 lake stations during 1990-2012 for C1 and K42 and 2000-2012 for E51. Observations are classified by dreissenid presence (P) and absence (A).

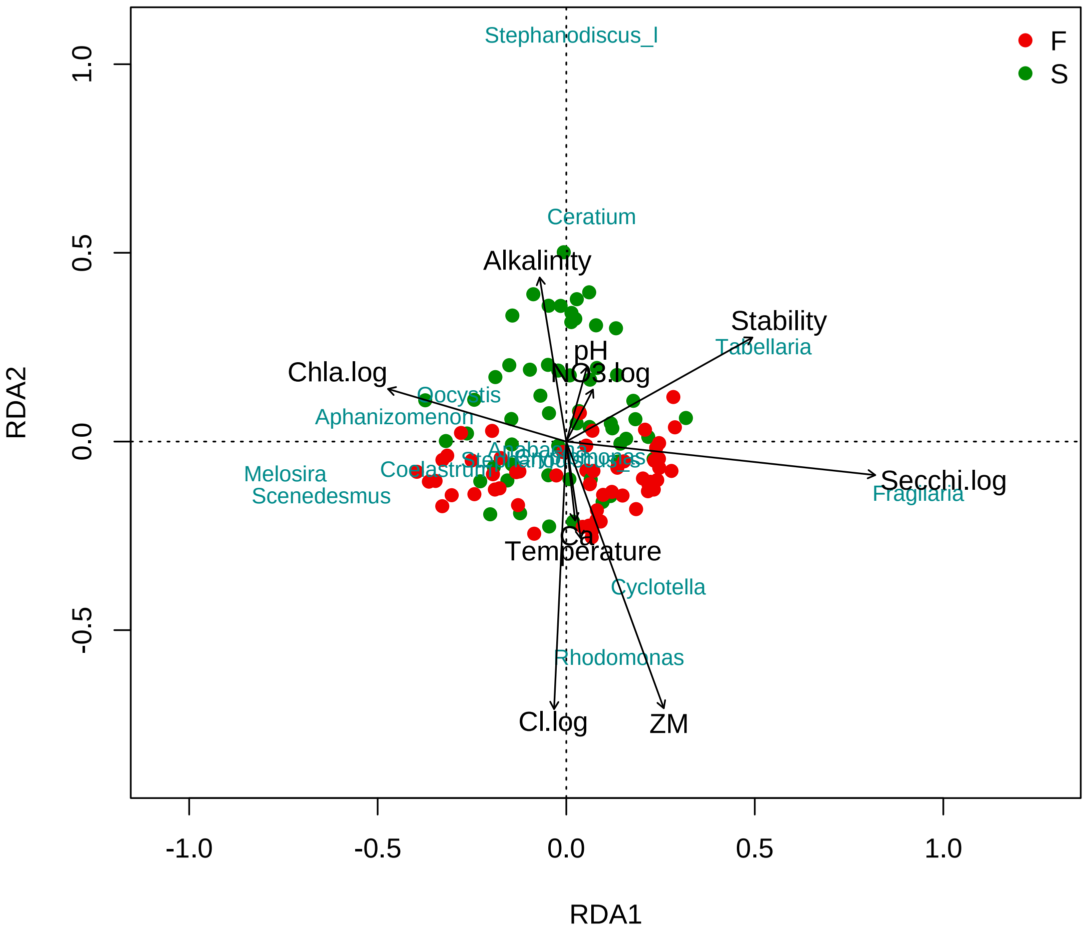

Figure 17. Ordination plot of the redundancy analysis (RDA) of the relationship between environmental parameters (vectors) and phytoplankton community composition in C1, E51, and K42 lake stations during 1990-2012 for C1 and K42 and 2000-2012 for E51. Observations are classified into seasons: fall (F) and summer (S).

2.2 Comparison of Seasons and Mussel Impacts within Stations:

Significant relationships ($p < 0.05$) at individual stations between limnological characteristics and seasonal phytoplankton community composition in summer and fall (Figures 17-19) and presence/absence of mussels (Figures 20 and 21) were identified by RDA selected by forward selection but the station data were analyzed independently of each other unlike Figures 10-12 . In general, the two deep sites have clear separation between summer and fall phytoplankton assemblages. Higher temperature and lower reactive silicate were associated with summer phytoplankton community structure and lower temperature and higher reactive silicate were associated with fall community structure. We did not observe clear separation between summer and fall phytoplankton assemblage in the shallow site (C1) perhaps due to its shallow nature and influxes from surrounding agriculture activities.

For the two deep sites, the dominant genera in the summer for both sites were *Dinobryon* spp. (chrysophyte), and *Cyclotella* spp. (diatom) (Figures 18 and 19). E51 was also dominated by *Uroglena* spp. (chrysophyte) during the summer. Dominant genera in the fall for both deep sites were *Rhodomonas* spp. (cryptophyte), *Tabelleria* spp. (diatom), and *Anabaena* spp. (cyanobacteria). K42 was also dominated by *Asterionella* spp. (diatom) and *Cryptomonas* spp (cryptophyte) while E51 was dominated by an unidentified chrysophyte spp. *Fragilaria* (diatom) was also dominant at both deep sites, appearing at the boundary between seasons.

Abundant genera at C1 were *Melosira* spp. (diatom), *Cryptomonas* spp., *Anabaena* spp., and *Aphanizomenon* spp. (cyanobacteria). No seasonal environmental gradient was associated with the separation of summer/fall phytoplankton community composition (Figure 17). *Stephanodiscus* spp. was associated with the early years (1990s), which were characterized by lower chloride and absence of dreissenid mussels (Figure 20), while *Rhodomonas* was associated with relatively low total phosphorus. *Melosira* spp., on the other hand, was associated with higher temperature. Overall, limnological characteristics explained 24% of the variation in phytoplankton composition at C1 ($R^2_{adj} = 0.241$). Dreissenid mussels, chloride, water temperature, and total phosphorus concentration were identified as significant limnological characteristics.

Abundant genera at E51 consisted of approximately 50% of total abundance of all phytoplankton biomass among all years at E51. Summer and fall phytoplankton assemblages are separated in the RDA triplot (Figure 18) by water temperature and reactive silicate. Summer phytoplankton community composition is associated with higher water temperature, higher TKN and lower silica, while fall phytoplankton community composition is associated with lower water temperature, lower TKN and higher reactive silicate. *Cyclotella* spp. (diatom), *Dinobryon* spp. (chrysophyte) and *Uroglena* spp. were associated with higher water temperature. At the same time, *Rhodomonas* was associated with higher reactive silicate, but *Fragilaria* and *Ceratium* (dinoflagellate) were associated with higher AMO index. Overall, limnological characteristics explained 28% of the variation in phytoplankton composition at E51 ($R^2_{adj} = 0.284$).

Abundant genera at K42 (Figures 19 and Figure 21) consisted of approximately 50% of total abundance of all phytoplankton biomass among all years at K42. Summer and fall phytoplankton assemblages are separated in the RDA triplot by water temperature. Summer phytoplankton community composition was associated with higher water temperature, while fall phytoplankton community composition was associated with lower water temperature. Large *Stephanodiscus* spp. was associated with early years (early 1990s) in the absence of dreissenid mussels and low chloride, while *Asterionella* spp. and *Fragilaria* spp. were associated with relatively high Si. *Cyclotella* spp. and *Dinobryon* spp. were strongly associated with higher temperature. A separation between pre- and post dreissenid periods may be observed in Figure 21 where higher abundance of *Stephanodiscus* spp. and *Ceratium* spp. were observed in the absence of dreissenids, whereas *Fragilaria* spp., *Cyclotella* spp. and *Dinobryon* spp. were observed in the presence of dreissenids. Overall, limnological characteristics explained 37% of the variation in phytoplankton composition at K42 ($R^2_{adj} = 0.365$). Chloride, water temperature, and reactive silicate were identified as significant limnological characteristics.

2.3 Summer/fall phytoplankton community composition over time

The dominant genera at C1 and K42 changed over time during each season but there was no clear trend in summer dominant genera at E51.

Dominant genera during the summer at C1 changed from *Stephanodiscus* spp. to *Melosira* spp. while dominant genera at K42 changed from *Stephanodiscus* spp. to *Tabellaria* spp. and *Dinobryon* spp. There was no clear pattern in dominant summer genera in E51 over time.

Dominant genera in the fall at C1 changed from large *Stephanodiscus* spp. to small *Stephanodiscus* spp. while dominant genera at K42 changed from large *Stephanodiscus* spp. to *Asterionella* spp. and *Fragilaria* spp. Dominant genera at E51 during the fall changed from *Anabaena* spp., *Tolypothrix* spp. and *Ceratium* spp. to *Microcystis aeruginosa* (which formed a bloom in 2011) and unidentified dinophytes.

Table 10. Seasonal composite phytoplankton biovolumes (summer and fall)

Year	C1 S	C1 F	E51 S	E51 F	K42 S	K42 F
1980					850	362
1990	1288	2656			355	985
1991	1589	1125			245	662
1992	534	1320			396	549
1993	1258	526			199	328
1994	700	1465			494	775
1995	700	654			273	429
1996	664	3717			801	284
1997	477	423			366	336
1998	378	212			477	448
1999	433	288	185	179		
2000	2707	736	333	374	572	460
2001	473	2320	422	247		389
2002	614	180	423	352	408	432
2003	706	407	233	375		
2004	1013	320	434	205		
2005	444	140	240	609	258	952
2006	548	1298	293		450	495
2007	187	191	229	486	419	590
2008	564	1451	272	232	546	764
2009	1986	1093	257	222	307	428
2010	2274	1073	176	179	193	557
2011	4466	4567	211	514	875	403
2012	1945		107	109	205	528

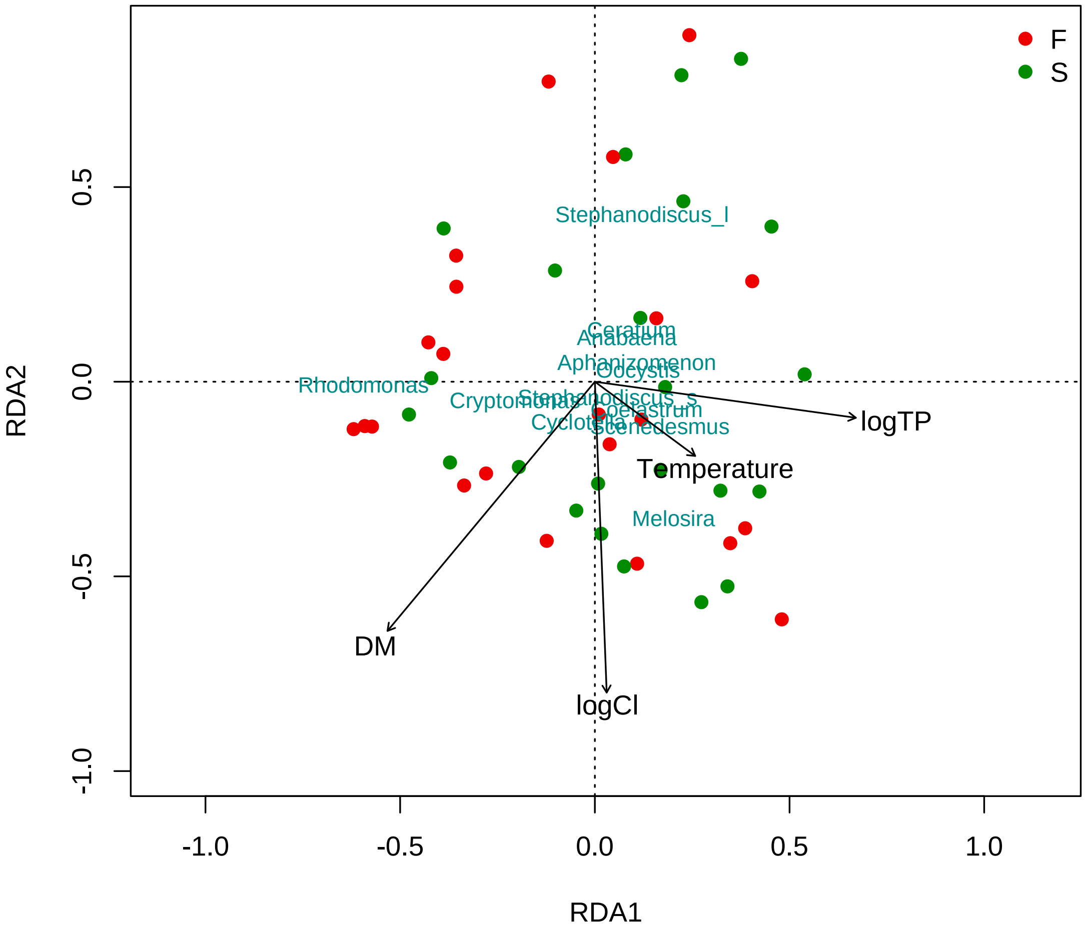

Figure 18. Ordination plot of the redundancy analysis (RDA) of the relationship between environmental parameters (vectors) and phytoplankton community composition at C1 during 1990-2012 for both summer and fall. Colour-coding is based on seasons - fall (F) and summer (S).

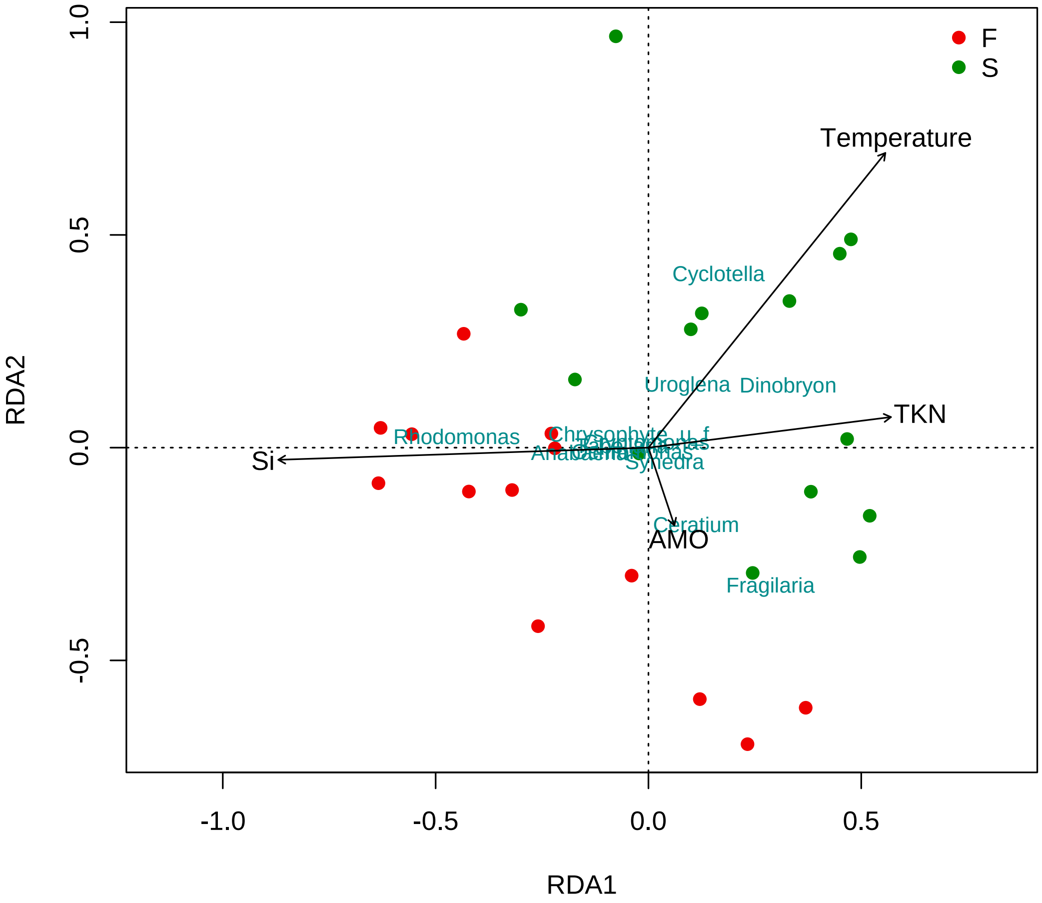

Figure 19. Ordination plot of the redundancy analysis (RDA) of the relationship between environmental parameters (vectors) and phytoplankton community composition at E51 during 1990-2012 for both summer and fall. Colour-coding is based on seasons - fall (F) and summer (S).

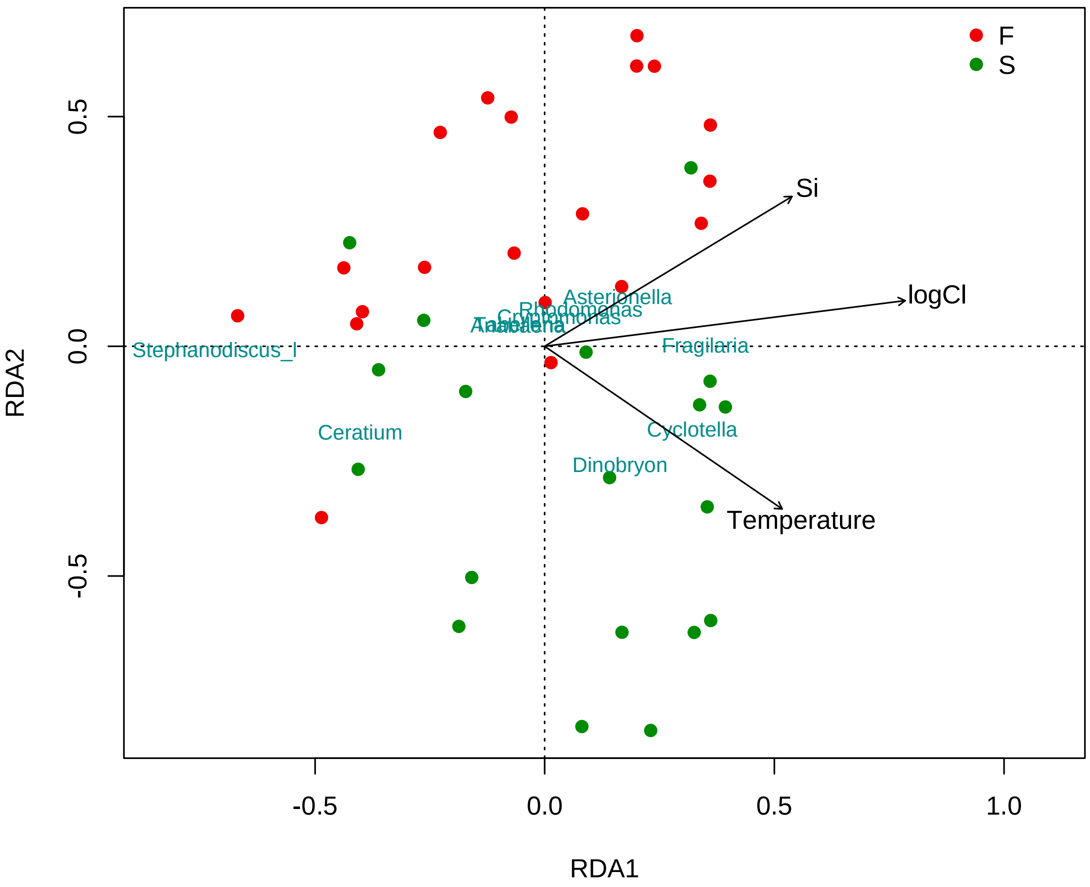

Figure 20. Ordination plot of the redundancy analysis (RDA) of the relationship between environmental parameters (vectors) and phytoplankton community composition at K42 during 1990-2012 for both summer and fall. Color-coding is based on seasons - fall (F) and summer (S).

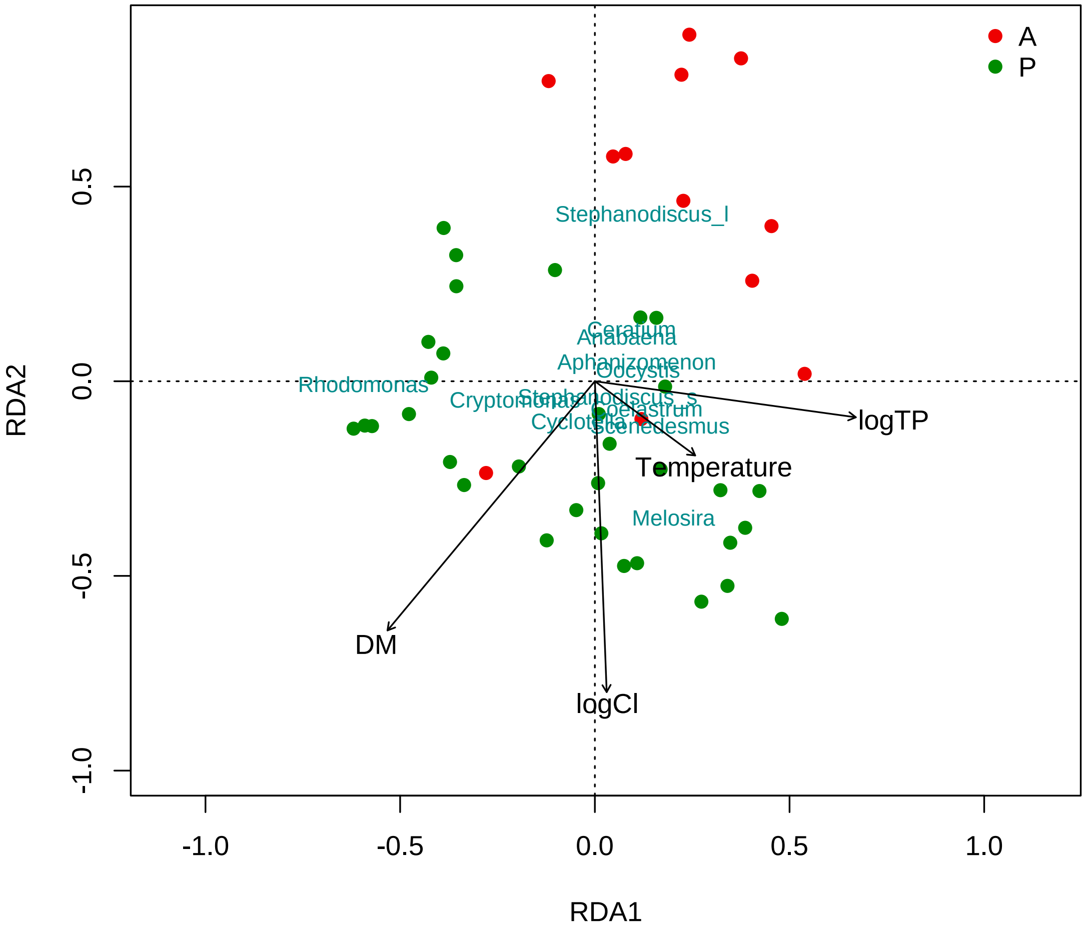

Figure 21. Ordination plot of the redundancy analysis (RDA) of the relationship between environmental parameters (vectors) and phytoplankton community composition at C1 during 1990-2012 for both summer and fall. Color-coding is based on dreissenid absence (A) and presence (P).

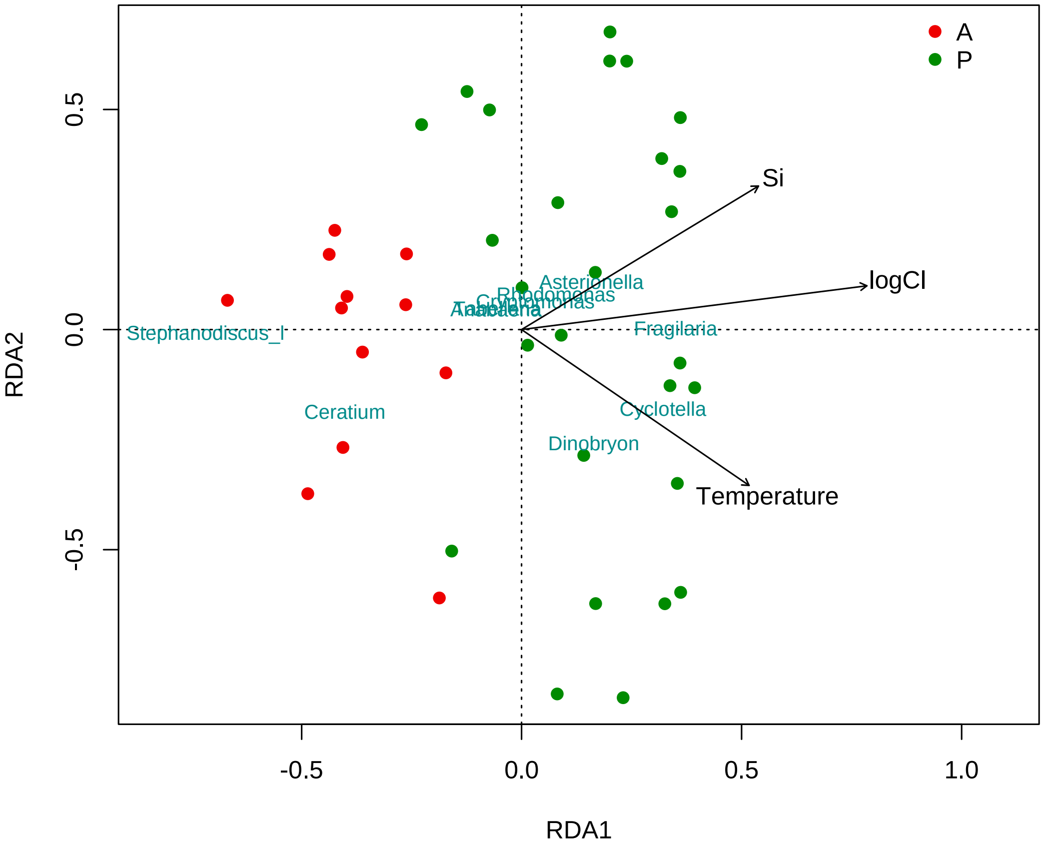

Figure 22. Ordination plot of the redundancy analysis (RDA) of the relationship between environmental parameters (vectors) and phytoplankton community composition at K42 during 1990-2012 for both summer and fall. Color-coding is based on dreissenid absence (A) and presence (P).

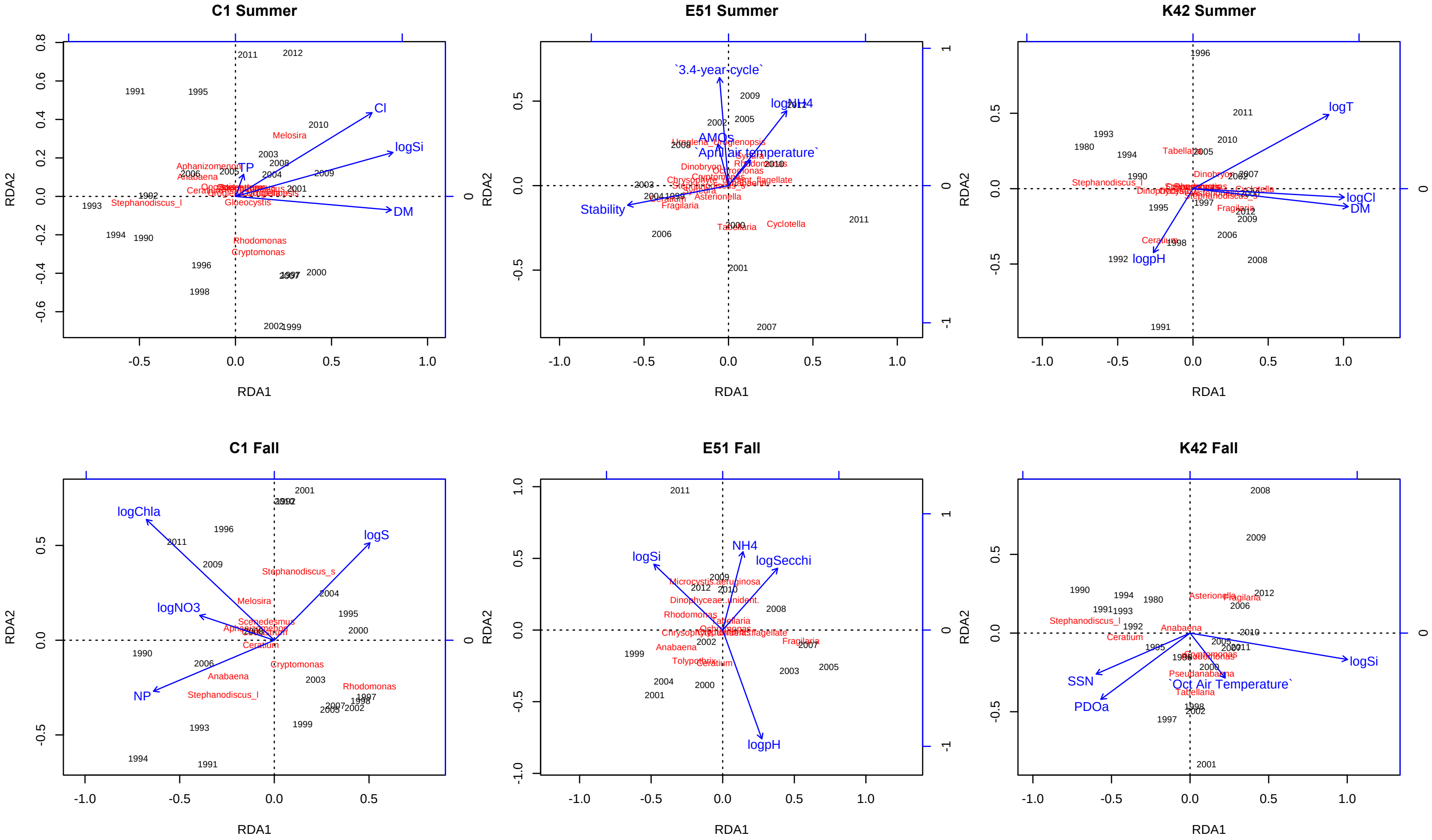

Figure 23. Ordination plot of the redundancy analysis (RDA) of the relationship between environmental parameters (vectors) and phytoplankton community composition at C1, K42 during 1990-2012 (K42 has an addition point for 1980) and E51 during 1999-2012 for either summer or fall.

DISCUSSION

Our goal was to identify significant changes in phytoplankton species composition in Lake Simcoe in recent decades and provide some insights into the relative importance of the various influences on long-term and seasonal changes (i.e., local weather, water chemistry, thermal properties, invasive species, indices for solar sunspots and large-scale climatic drivers) acting in concert with each other rather than acting individually. Our analysis revealed that phytoplankton community composition changed 1) among years at all sites and were primarily associated with changes in dreissenid mussels, reactive silicate, water column stability, water temperature, and nutrients. Dominant genera in annual composite samples changed from *Stephanodiscus* spp. to *Fragilaria* spp. Fig. 8 and 9) Phytoplankton differences between seasons (summer and fall) were primarily associated with changes in water temperature and possibly nutrients (silica and TKN) at two deep sites (E51 and K42). Clear separation between summer and fall phytoplankton composition was not observed at the shallow site (C1). The dominant summer species at E51 and K42 were *Dinobryon* spp. and *Cyclotella* spp. and the dominant fall species were *Rhodomonas* spp*.,* *Tabellaria* spp. and *Anabaena* spp. There was no consistent pattern of dominant summer/fall species change over time. The dominant summer species in C1 changed from *Stephanodiscus* spp. to *Melosira* spp.; the dominant fall species in C1 changed from large *Stephanodiscus* spp. to small *Stephanodiscus* spp. In K42, dominant summer species changed from *Stephanodiscus* spp. to *Tabellaria* spp. and *Dinobryon* spp.; dominant fall species changed from large *Stephanodiscus* spp. to *Asterionella* spp. and *Fragilaria* spp.

Stephanodiscus spp. dominated spring samples (Tennant 2013) and also dominated summer or fall samples in C1 and K42 during the early 1990s. However, this genus did not dominate summer or fall samples in E51 from 1999 on. Dominant spring diatoms shifted from larger *Stephanodiscus* species to primarily *Fragilaria* spp., *Tabellaria* spp., *Cyclotella* spp., unknown centrics and smaller *Stephanodiscus* spp. in later years (Tennant 2013).

Diatom standing stocks did not vary much among seasons at K42. Spring (measured two to six weeks after ice-out) diatom biovolumes at K42 ranged from 101 to 775 mm^3 m^{-3} in 9 of 32 years analyzed between 1980 and 2011 (mean 346 mm^3 m^{-3}) excluding an unusually large biovolume of 2948 mm^3 m^{-3} in 2009 (Tennant 2013). Mean spring, summer and fall diatom biovolumes were similar at 346, 197 and 274 mm^3 m^{-3}, respectively, for spring: 1980, 1981, 1997 and 2006-2010; summer and fall: 1980 and 1990-2012 (Tennant 2013; Table 4). Similar biovolumes during the three seasons could explain the linearity in dissolved oxygen depletion rates below 18 m (Chapter 1).

Dreissenid Mussel Invasions

Grazing pressure by dreissenid mussels can play a role in decreasing overall phytoplankton standing stock (Higgins and Vander Zander 2010; Barbiero et al. 2006; Winter et al. 2011) but to what extent they can change phytoplankton communities composition is not clear. Moreover, some studies are contradictory to one another (for example Vanderploeg et al. 2001 and Pires et al. 2005).

Dreissenids clearly affected species composition after their invasion of Lake Simcoe (Figures 8, 15, 20 and 21). Among all sites of annual analysis of phytoplankton relationships with their ambient environment, dreissenid mussels contribute to the most variation explained and are likely the fundamental cause of phytoplankton community composition change. The dreissenid mussel invasion also played a role in structuring seasonal phytoplankton community composition at K42. At K42, there was a clear separation between pre- and post DM periods (Figure 21). There was no clear separation between summer and fall until after dreissenids became established (Figures 19 and 21). RDA x-y observations at C1 are clearly separated by the dreissenid invasion but not by summer and fall seasons (Figs 17 and 20). The dreissenid invasion coincided with increased Si, Cl and summer water temperature at K42.

Dreissenid grazing pressure may alter nutrient concentrations and it is these chemical changes which might influence species competition. As a result of grazing pressure by *Dreissena* spp., silicate concentrations in mesotrophic Lake Simcoe are now much higher, perhaps because of reduced uptake. The resulting relaxation of silica limitation may have shifted the phytoplankton community composition towards diatom species with higher silicate requirements and lower phosphorus requirements (Barbiero et al. 2006; Winter et al. 2011). Tilman and Kilham (1977) noted that nutrient limitation change may lead to dominant species change. Kilham et al. (1986) characterized P/Si requirements of various algal species and concluded that *Stephanodiscus* spp. has the highest P/Si requirement; *Asterionella* spp., *Fragilaria* spp. and *Nitzschia* spp. have intermediate P/Si requirements; *Synedra* spp has the lowest P/Si requirement. Barbiero et al. (2006) observed that spring dominance in Lake Erie shifted from a mix of pennate and large centric diatoms and pyrrophytes to three centric diatoms with high silica requirements: *Aulacoseira islandica*, *Stephanodiscus hantzschii* and *Stephanodiscus parvus*.

However, Barbiero's findings are inconsistent with results from Lake Simcoe. A paleolimnological study of Lake Simcoe (Hawryshyn et al. 2012) found large declines in heavily silicified *Aulacoseira* spp. after the dreissenid invasion when Si increased. Tennant (2013) observed a significant decrease in *Stephanodiscus* spp. (centric diatom) and increase in *Fragilaria* spp. (pennate diatom). A shift in cell size class from large *Stephanodiscus* to small *Stephanodiscus* at a few study sites was also evident.

Selective grazing by dreissenids in Lake Simcoe may have played a role in the observed shift in dominant taxa (Winter et al. 2011). *Stephanodiscus* spp. is a centric diatom with drum-shaped cells that exists as single cells or short chains (Scheffler and Morabito 2003) whereas *Fragilaria* spp. is a pennate diatom with elongated cells that often aggregate into long chains (Bahulikar 2006). *F. crotonensis* in particular form characteristic long, double comb-like chains of cells. As a consequence, within the mix of suspended particles in Lake Simcoe, the chains of *Fragilaria* spp. may be less palatable to dreissenids than the *Stephanodiscus* spp. cells. However, experimental studies give contradictory results. Naddafi et al. (2007) found that the dreissenid mussel clearance rate was low for diatoms in general (*Stephanodiscus* spp. and *Fragilaria* spp. were the dominant diatoms used in their study) suggesting that grazing may not have been responsible for the shift in dominant diatoms. In our analysis, the shared variance explained by both ambient environmental conditions and dreissenid mussel invasion in phytoplankton community composition contributed more than 60% of the total variance explained. Although we cannot conclude from these results that dreissenid mussels affected the phytoplankton community composition by changing their ambient environment, the statistical correlations were strong for almost all sites and seasons.

Hawryshyn et al.'s (2012) paleolimnology study challenged both silicate limitation and selective grazing hypotheses. They concluded that a shift in diatom composition occurred in the mid-1990s, coinciding with the invasion of dreissenid mussels and furthermore, that water quality has been affected by the interaction of numerous environmental stressors over the past two centuries, the complexity of which has been amplified by recent warming. They suggested that rather than tease apart one mechanism at a time, it seems more reasonable that the post-dreissenid change in phytoplankton community composition was associated with a combination of factors: increased water clarity or light availability, increased water temperature, increased column thermal stability, shift in P concentration, and high grazing pressure (which may, however, have decreased in the latter part of the study period). The lack of consensus in the literature suggests that the impact of invasions are complicated and affected by lake-specific conditions including lake morphology, water chemistry, nutrients and physical conditions (air and water temperature, wind, thermal regime, climatic oscillations).

Invasion of *Bythotrephes longimanus*

It is possible that the recent invasion by *Bythotrephes* has resulted in significant alterations to the food web with indirect effects on phytoplankton biomass and composition. It is expected that overall grazing pressure of phytoplankton might be released due to cascading effect of this invasive zooplanktivore (an invertebrate which preys on zooplankton) and phytoplankton community composition change was observed after the *Bythotrephes*' invasion. A study in central Ontario found that evidence of change in phytoplankton composition after *Bythotrephes* invaded but edible phytoplankton biovolume did not increase (Strecker et al. 2011). Complex interactions in the food web pose a significant challenge for predicting the effect of invasive species.

Nutrient Availability and Water Chemistry

Changes in nutrient concentrations appear to be one of multiple drivers of changes in phytoplankton biovolume and community composition in Lake Simcoe. PCA analysis with all eight stations found lower TP and NH_4 and higher Si associated with post 1996 years, i.e. after the dreissenid invasion (Figure 7). Partial diversion of sewage south to Lake Ontario, improved sewage effluent regulations and removal of nutrients by dreissenid grazing (Hecky et al. 2004) may have been responsible.

The high TN/TP ratio in Lake Simcoe (Winter et al. 2011) suggest that productivity is P-limited (TN/TP > 10). Watson et al. (1997) showed that TP correlated well with total biomass of phytoplankton as well as, to a lesser extent, the biomass of six major taxonomic groups of phytoplankton. Most large pelagic cyanobacteria like *Anabaena* spp., *Aphanizomenon* spp. and *Microcystis* spp. and diatoms like *Melosira* spp., *Fragilaria* spp., *Stephanodiscus* spp. and *Asterionella* spp. are eutrophic (high P) indicators (Rawson 1956). However, while the filamentous N fixers *Anabaena* and *Aphanizomenon* are typically associated with high P, their abundance is initiated by very low dissolved inorganic N. Chlorophytes like

Staurastrum spp., diatoms like *Tabellaria* spp. and *Cyclotella* spp., and chrysophytes like *Dinobryon* are oligotrophic (low P) indicators (Rawson 1956) which is consistent with our ordination which revealed that *Melosira* spp. was positively associated with higher P and *Tabellaria* was positively associated with lower P (Figure A-annual-K42).

Ratios between major nutrients can affect the outcome of species composition. The silicate to phosphate (Si/P) ratio was considered crucial to determining diatom dominance by Tilman and Kilham (1976) who reported that *Asterionella formosa* was better able to grow at low phosphate concentration in comparison to *Cyclotella meneghiniana* because *A. formosa* was more efficient at using internal phosphate for growth. However, *C. meneghiniana* was more efficient at using internal silicate for growth and therefore more competitive at low Si concentration. Similar experiments were done by Donk and Kilham (1990) who examined the effects of temperature (from 5° to 20°C) on *Stephanodiscus hantzschii*, *Asterionella formosa* and *Fragilaria crotonensis* in silicate- and phosphate-limited conditions. They found that *S. hantzschii* had higher maximum growth rates than the other two species at all temperatures, and the maximum growth rates of all species increased with increasing temperature. *S. hantzschii* had low silica requirements and did well under Si-limiting conditions while *A. formosa* and *F. crotonensis* had higher and nearly identical silica requirements. *A. formosa* had the lowest requirements for growth under P limitation, *F. crotonensis* was intermediate and *S. hantzschii* had the highest growth requirements for P. In P-limited cultures (Si:P > 75), *A. formosa* often co-dominated with *F. crotonensis*. *S. hantzschii* never dominated at high Si:P ratios. At intermediate Si:P ratios, *A. formosa* and *F. crotonensis* had almost identical silica requirements, although sometimes *F. crotonensis* was more abundant than predicted. A change in Si:P ratios might explain some of the shift in species composition in Lake Simcoe (Figure 11). After the dreissenid invasion of Lake Simcoe, Si increased and P declined, hence the mean annual Si:P ratio by weight was higher after 1996 (0.0875 ± 0.0351) compared to the 1980s (0.0365 ± 0.0227) in K42.

Light Penetration (Secchi depth)

An improvement in light regime after the dreissenid invasion, as measured by Secchi depth, was associated with the changes in the phytoplankton community composition, for example, as indicated by the strong positive correlation between Secchi depth and *Fragilaria* spp. (Figures 11 and 12). However, the preference of the main post-dreissenid dominant, *Fragilaria* spp., for low light environments is well documented (Interlandi et al. 1999), which suggests that the increase in light availability was more likely a consequence, rather than a cause, of the changes seen in the phytoplankton community.

Climate Change and Large-scale Climatic Oscillations

Independent of water chemistry changes, recent studies suggest that climate may be a significant driver structuring algal populations in north-temperate lakes (Paterson et al. 2008). Climate changes can lead to longer ice-free seasons, warmer water temperatures and increased thermal stability, the latter favouring smaller cells with lower sinking rates. While a long-term climate trend over decades is evident in Ontario (Nalley et al. 2013), short-term climate oscillations associated with oceanic oscillations (ENSO, NAO, NP, MEI, QBO and AMO) and sunspot numbers can be superimposed on the long-term trend. For example, when sunspot numbers are high, phytoplankton with preference for warm temperature and high light may thrive. In wet years with high runoff, phytoplankton with preference for higher nutrient levels may thrive because of higher nutrient loading. Both long-term shift and short-term oscillations could affect phytoplankton composition. Temporal oscillations (4 to 7 year cycles) accounted for a small amount of the variation, 8%, in phytoplankton composition in Lake Simcoe (Figure 13).

NAO has been well studied for its influence on warm winter-spring air temperature (Hurrell, 1995). In Lake Simcoe, winter/spring air temperatures were correlated with phytoplankton community composition change (Figure A-annual-C6, Figure A-annual-C9, Figure A- Seasonal-E51). There are two possible mechanisms linking winter temperatures/NAO and phytoplankton community structure: 1) Spring mixing intensity and nutrient availability (Straile et al. 2003) is influenced by NAO largely due to incomplete spring mixing resulting in a nutrient deficit; 2) Spring plankton population dynamics of herbivores (Straile 2000; Anneville et al. 2002) are influenced by mild winters during a NAO which will contribute to higher zooplankton population growth rate.

A change in size class was seen in Lake Simcoe in the spring when *Stephanodiscus* shifted from large to small species (Tennant 2013). In the summer and fall, the dominant genera changed from *Stephanodiscus* spp., *Ceratium* spp. and *Melosira* spp. to *Fragilaria* spp., *Rhodomonas* spp., *Cryptomonas* spp., *Cyclotella* spp., *Asterionella* spp. and *Tabellaria* spp.

Comparison of Statistical Ordination Methods

We found that water quality variables that significantly explained phytoplankton community composition were very similar to those identified by Winter et al. (2011) except that the current study does not have soluble reactive P and used NO_3 and TKN instead of total nitrogen. The CCA biplot in Winter et al. (2011) shows the same pattern of sites separated by explanatory variables as in our RDA triplot. Variation explained increased slightly from 27% to 36% in this study indicating that RDA followed by Hellinger transformation of species community composition data might be a better ordination method than CCA. Additional years of data and additional variables may help explain more variation.

Appendices

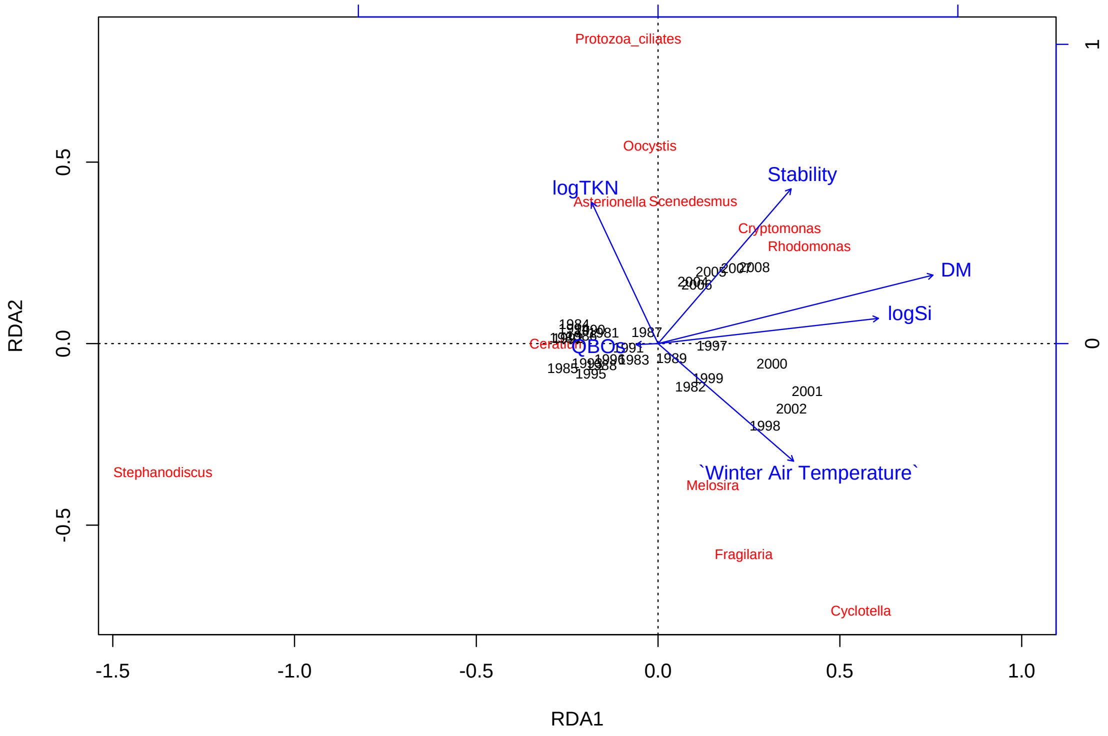

Figure A-annual-C1. Ordination plot of the redundancy analysis (RDA) on the relationship between annual mean environmental parameters (vectors) and annual composite phytoplankton community composition at C1 station during 1980-2008.

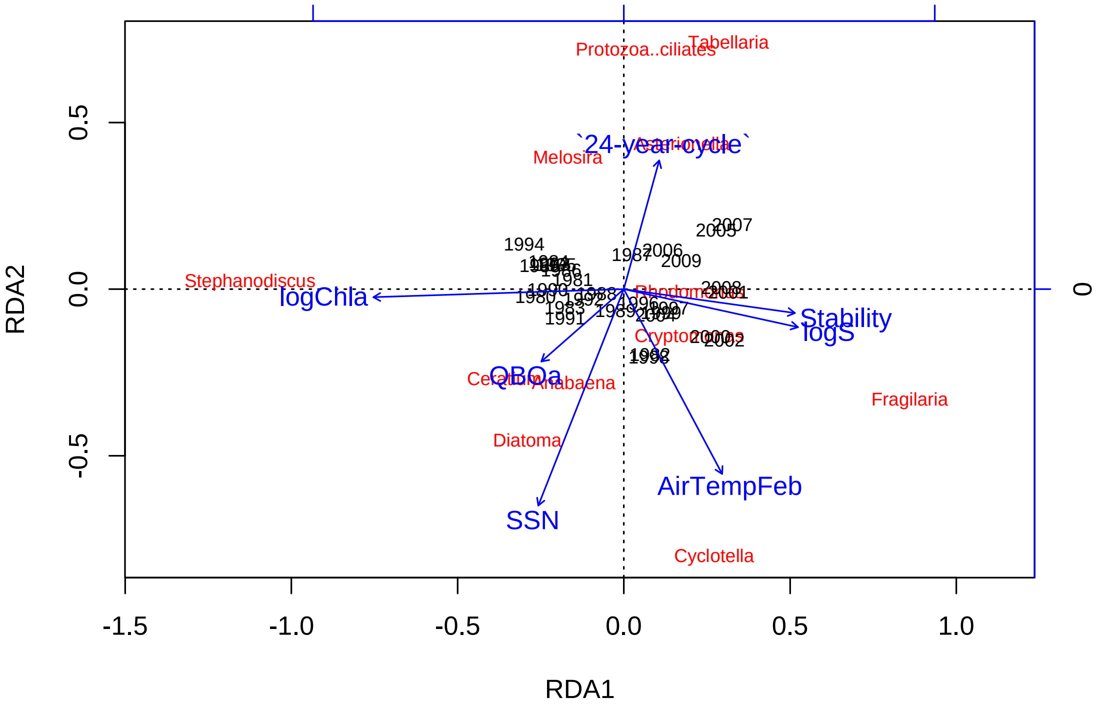

Figure A-annual-C6. Ordination plot of the redundancy analysis (RDA) on the relationship between annual mean environmental parameters (vectors) and annual composite phytoplankton community composition at C6 station during 1980-2008.

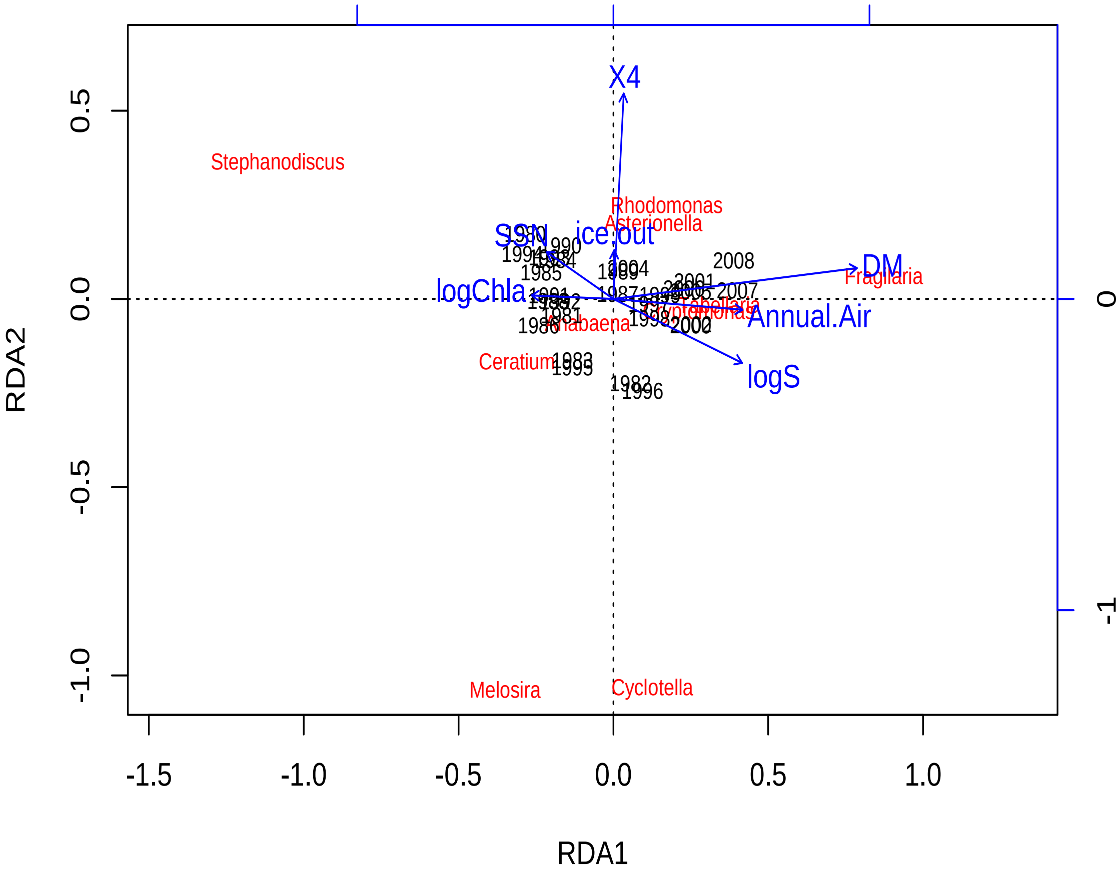

Figure A-annual-C9. Ordination plot of the redundancy analysis (RDA) on the relationship between annual mean environmental parameters (vectors) and annual composite phytoplankton community composition at C9 station during 1980-2008. X4 representing air temperature in April.

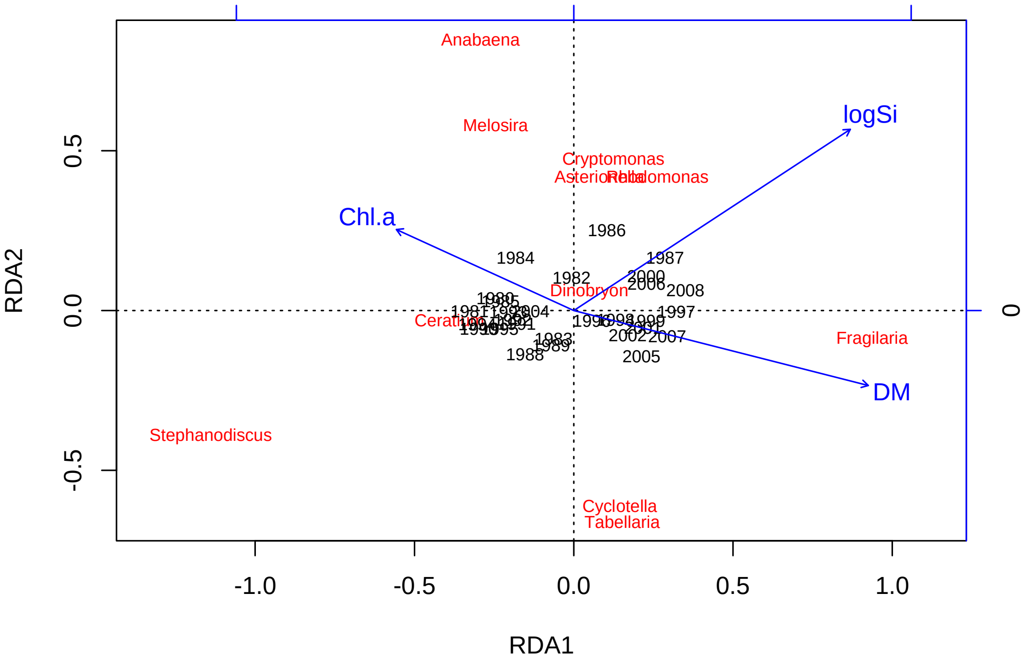

Figure A-annual-E51. Ordination plot of the redundancy analysis (RDA) on the relationship between annual mean environmental parameters (vectors) and annual composite phytoplankton community composition at E51 station during 1980-2008.

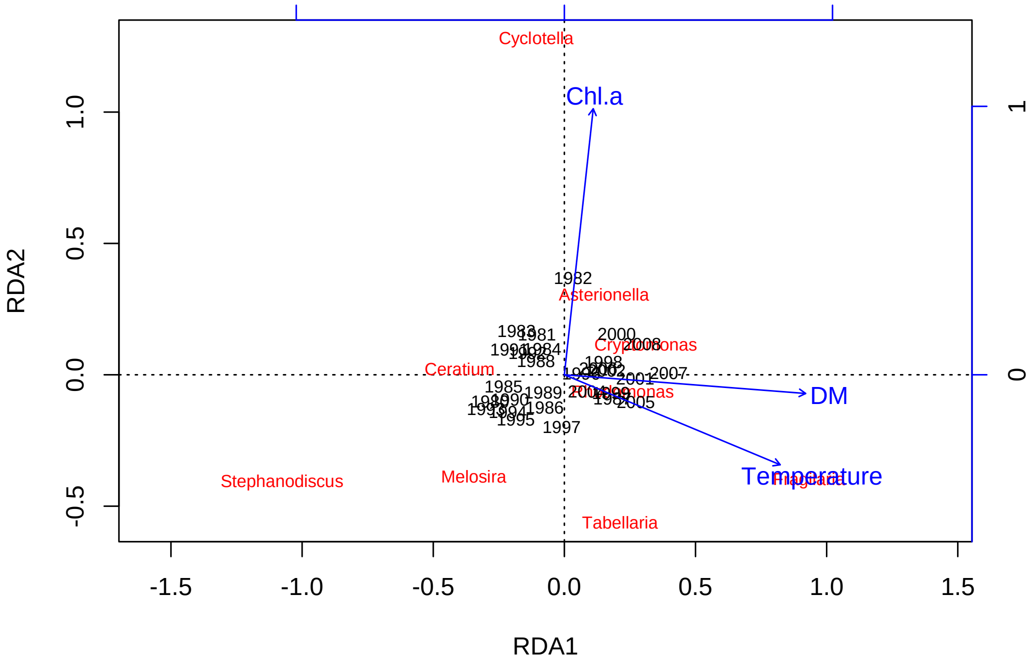

Figure A-annual-K39. Ordination plot of the redundancy analysis (RDA) on the relationship between annual mean environmental parameters (vectors) and annual composite phytoplankton community composition at K39 station during 1980-2008.

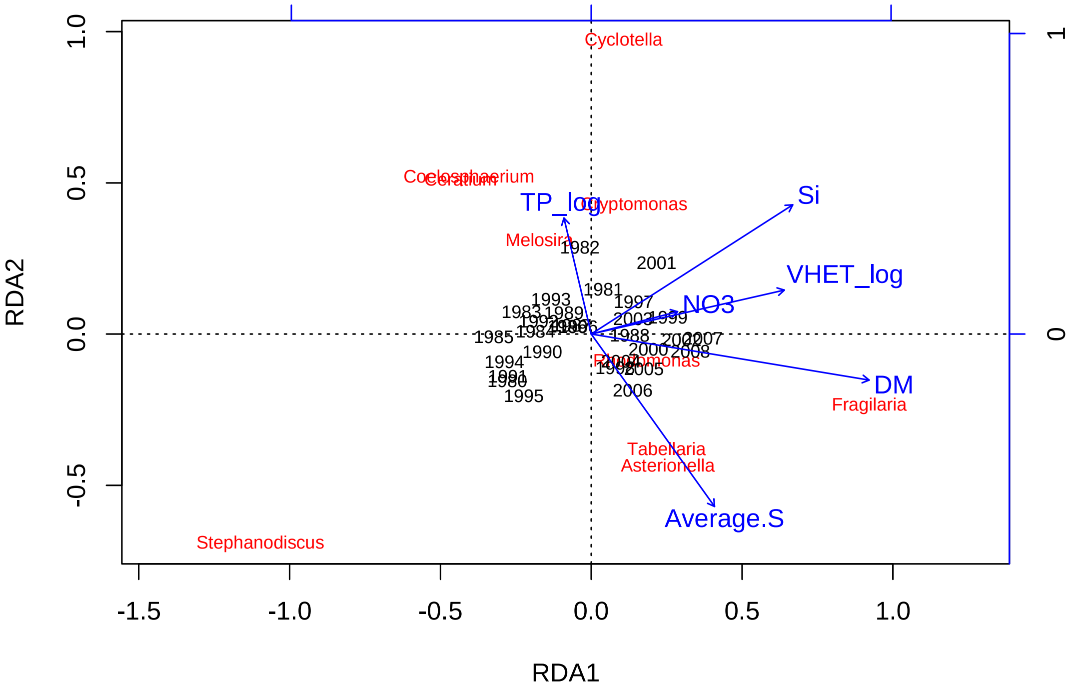

Figure A-annual-K42. Ordination plot of the redundancy analysis (RDA) on the relationship between annual mean environmental parameters (vectors) and annual composite phytoplankton community composition at K42 station during 1980-2008.

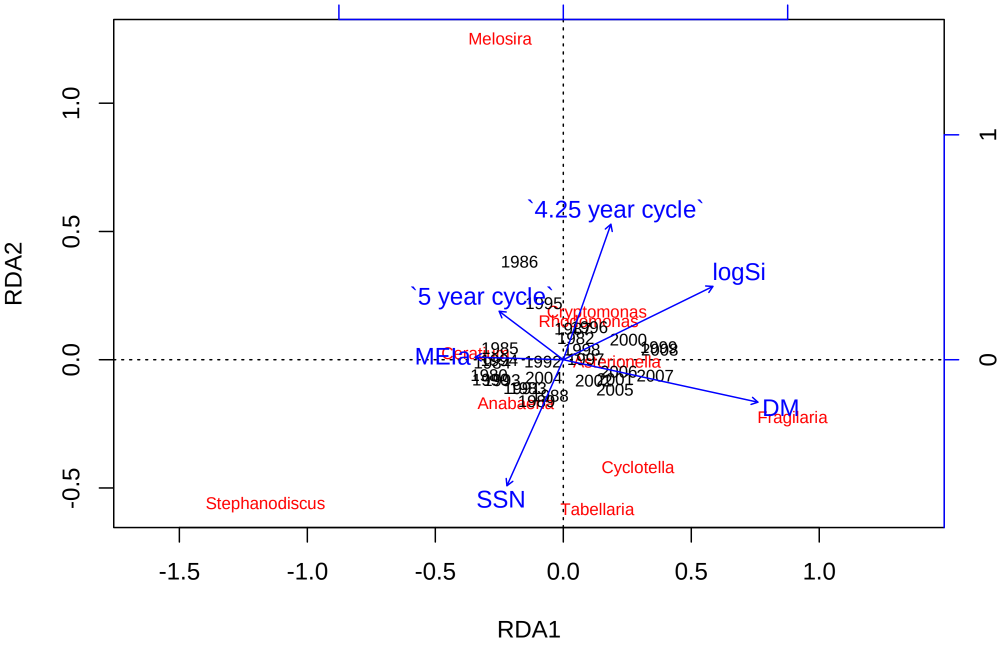

Figure A-annual-K45. Ordination plot of the redundancy analysis (RDA) on the relationship between annual mean environmental parameters (vectors) and annual composite phytoplankton community composition at K45 station during 1980-2008.

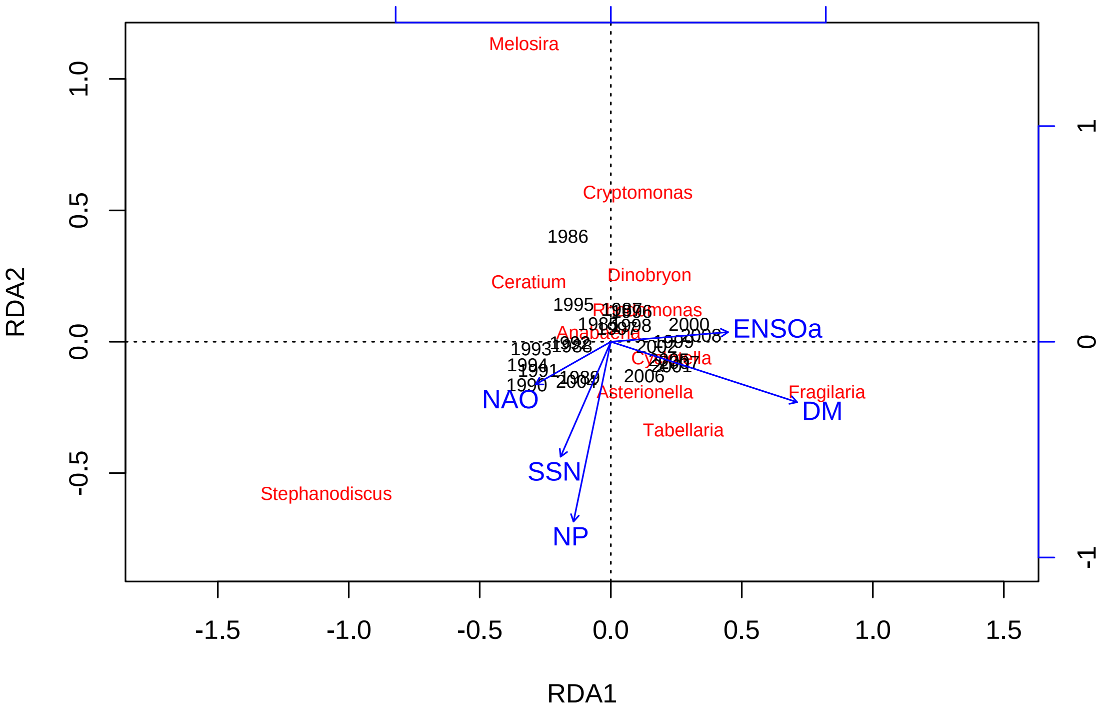

Figure A-annual-S15. Ordination plot of the redundancy analysis (RDA) on the relationship between annual mean environmental parameters (vectors) and annual composite phytoplankton community composition at S15 station during 1985-2008.

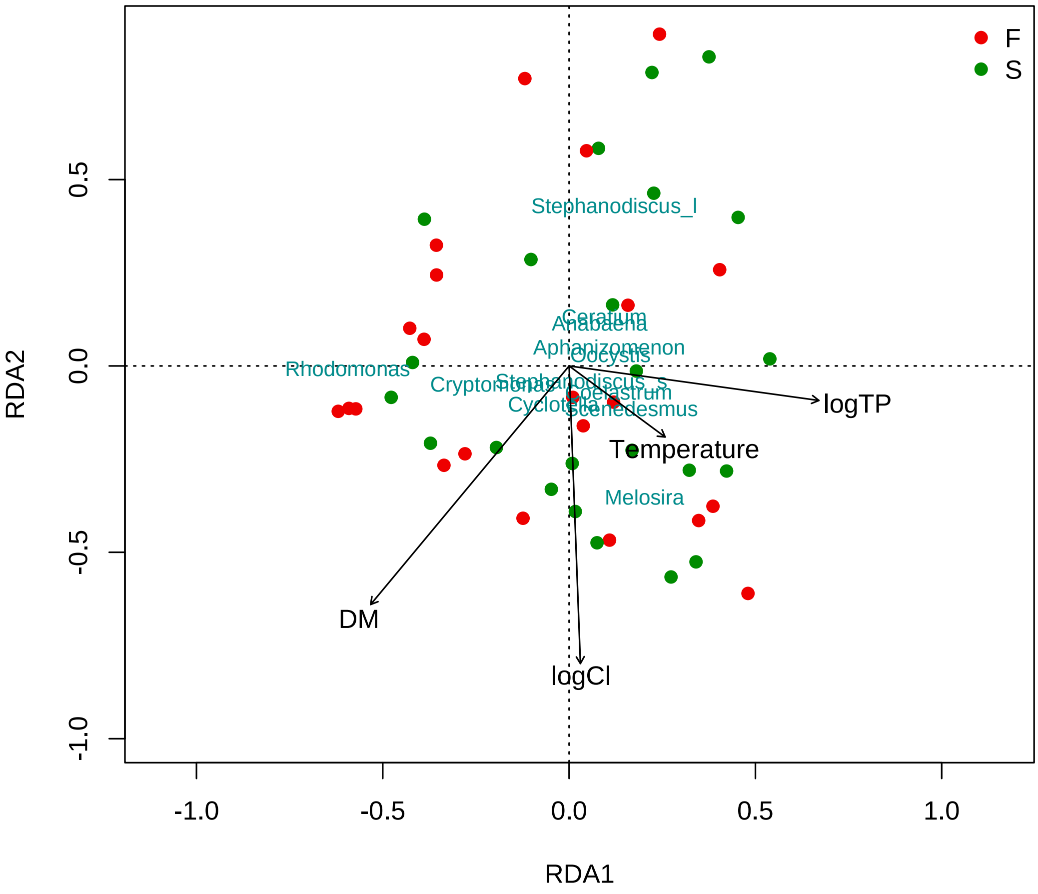

Figure A-seasonal-C1. Ordination plot of the redundancy analysis (RDA) of the relationship between environmental parameters (vectors) and phytoplankton community composition at C1 station during 1990-2012. Observations are classified into seasons: fall (F) and summer (S).

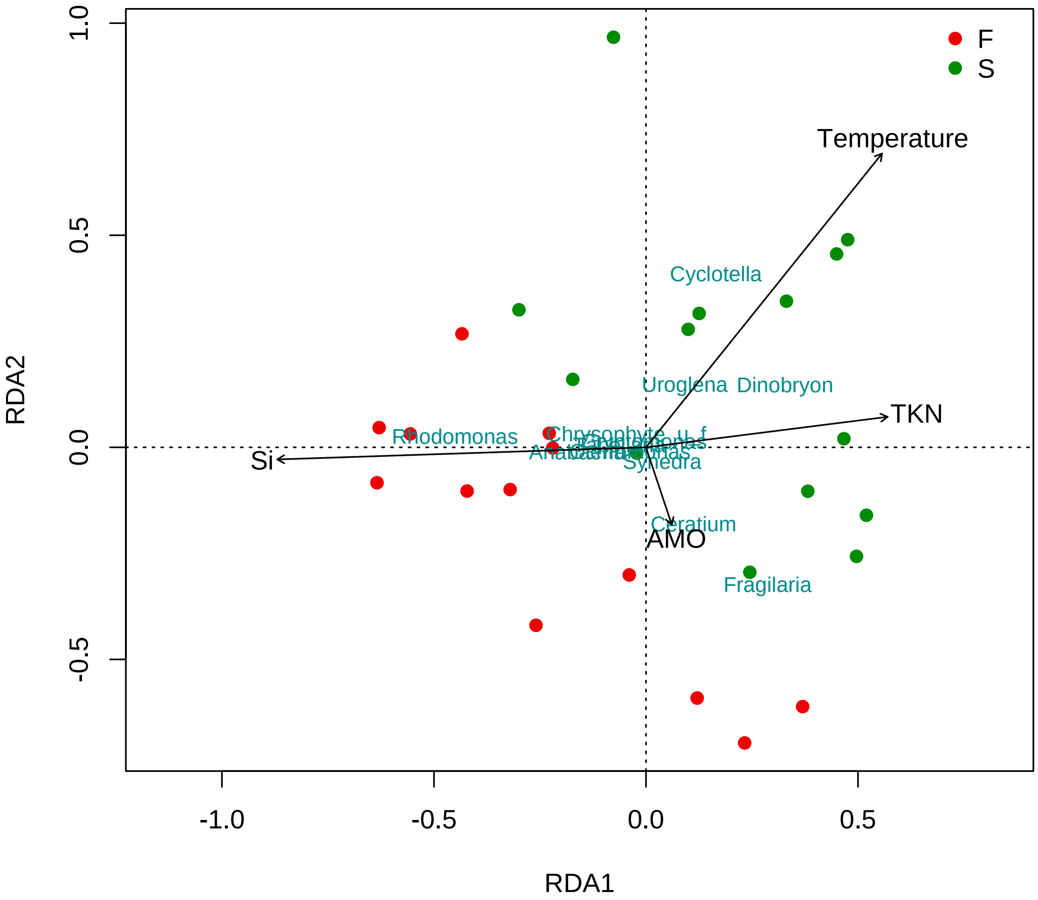

Figure A-seasonal-E51. Ordination plot of the redundancy analysis (RDA) of the relationship between environmental parameters (vectors) and phytoplankton community composition at E51 station during 2000-2012. Observations are classified into seasons: fall (F) and summer (S).

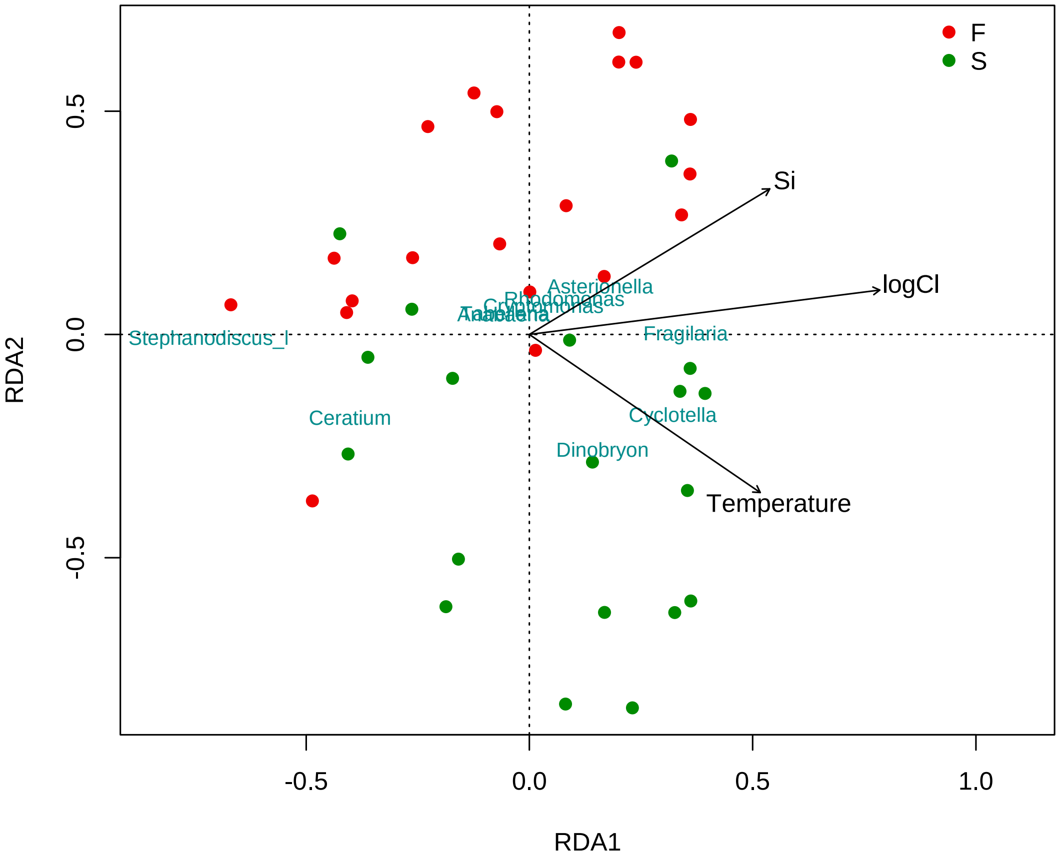

Figure A-seasonal-K42. Ordination plot of the redundancy analysis (RDA) of the relationship between environmental parameters (vectors) and phytoplankton community composition at K42 station during 1990-2012. Observations are classified into seasons: fall (F) and summer (S).

REFERENCES

Ahlgren, G., Gustafsson, I. B., & Boberg, M. (1992). Fatty acid content and chemical composition of freshwater microalgae. *Journal of Phycology*, *28*(1), 37-50.

Anneville, O., Gammeter, S., & Straile, D. (2005). Phosphorus decrease and climate variability: mediators of synchrony in phytoplankton changes among European peri-alpine lakes. *Freshwater Biology*, *50*(10), 1731-1746.

Anneville, O., Ginot, V., Druart, J. C., & Angeli, N. (2002). Long-term study (1974 1998) of seasonal changes in the phytoplankton in Lake Geneva: a multi-table approach. *Journal of plankton research*, *24*(10), 993-1008.

Arnold, D. E. (1971). Ingestion, assimilation, survival, and reproduction by *Daphnia pulex* fed seven species of blue-green algae. *Limnology and oceanography*, *16*(6), 906-920.

Bahulikar, R. A. (2006). Doctoral dissertation: Diatoms from littoral zone of Lake Constance: diversity, phylogeny, extracellular polysaccharides and bacterial associations. University of Konstanz, Konstanz.

Baldwin, M. P., Gray, L. J., Dunkerton, T. J., Hamilton, K., Haynes, P. H., Randel, W. J., Holton J. R., Alexander M. J., Hirota I., Horinouchi T. Jones D. B. A., Kinnersley J.S., Marquardt C., Sato K., & Takahashi, M. (2001). The quasi - biennial oscillation. *Reviews of Geophysics*, *39*(2), 179-229.

Barbiero, R. P., Rockwell, D. C., Warren, G. J., & Tuchman, M. L. (2006). Changes in spring phytoplankton communities and nutrient dynamics in the eastern basin of Lake Erie since the invasion of *Dreissena* spp. *Canadian Journal of Fisheries and Aquatic Sciences*, *63*(7), 1549-1563.

Bestelmeyer, B. T., Ellison, A. M., Fraser, W. R., Gorman, K. B., Holbrook, S. J., Laney, C. M., Ohman M. D., Peters D. P., Pillsbury, F. C., Rassweiler A. & Sharma, S. (2011). Analysis of abrupt transitions in ecological systems. *Ecosphere*, *2*(12), art129.

Bondarenko, N. A., & Evstafyev, V. K. (2006). Eleven-and ten-year basic cycles of Lake Baikal spring phytoplankton conformed to solar activity cycles. *Hydrobiologia*, *568*(1), 19-24.

Borcard, D., Legendre, P., Avois-Jacquet, C., & Tuomisto, H. (2004). Dissecting the spatial structure of ecological data at multiple scales. *Ecology*, *85*(7), 1826-1832.

Borcard, D., Legendre, P., & Drapeau, P. (1992). Partialling out the spatial component of ecological variation. *Ecology*, *73*(3), 1045-1055.

Borcard, D., & Legendre, P. (2002). All-scale spatial analysis of ecological data by means of principal coordinates of neighbour matrices. *Ecological Modelling, 153*(1), 51-68.

Brett, M. T., Muller-Navarra, D. C., Ballantyne, A. P., Ravet, J. L., & Goldman, C. R. (2006). Daphnia fatty acid composition reflects that of their diet. *Limnology and Oceanography*, *51*(5), 2428-2437.

Brett, M. T., & Müller-Navarra, D. C. (1997). The role of highly unsaturated fatty acids in aquatic foodweb processes. *Freshwater Biology*, *38*(3), 483-499.

Chisholm, S. W. (1992). Phytoplankton size. In *Primary productivity and biogeochemical cycles in the sea* (pp. 213-238). New York, NY: Springer.

Christensen, M. R., Graham, M. D., Vinebrooke, R. D., Findlay, D. L., Paterson, M. J., & Turner, M. A. (2006). Multiple anthropogenic stressors cause ecological surprises in boreal lakes. *Global Change Biology*, *12*(12), 2316-2322.

Cobelas A. M. and Lechado Z. J., (1989), Lipids in microalgae. A review I, Biochemistry. *Grasas Aceites, 40*(2), pp. 118–145

DeMott, W. R., & Tessier, A. J. (2002). Stoichiometric constraints vs. algal defenses: testing mechanisms of zooplankton food limitation. *Ecology*, *83*(12), 3426-3433.

Donk, E., & Kilham, S. S. (1990). Temperature effects on silicon ‐and phosphorus limited growth and competitive interactions among three diatoms. *Journal of Phycology*, *26*(1), 40-50.

Dray, S., Legendre, P., & Peres-Neto, P. R. (2006). Spatial modelling: a comprehensive framework for principal coordinate analysis of neighbor matrices (PCNM). *Ecological Modelling*, *196*(3), 483-493.

Eimers, M. C., Winter, J. G., Scheider, W. A., Watmough, S. A., & Nicholls, K. H. (2005). Recent changes and patterns in the water chemistry of Lake Simcoe. *Journal of Great Lakes Research*, *31*(3), 322-332.

Evans, D. O., Nicholls, K. H., Allen, Y. C., & McMurtry, M. J. (1996). Historical land use, phosphorus loading, and loss of fish habitat in Lake Simcoe, Canada. *Canadian Journal of Fisheries and Aquatic Sciences*, *53*(S1), 194-218.

Fahnenstiel, G. L., & Glime, J. (1983). Subsurface chlorophyll maximum and associated Cyclotella pulse in Lake Superior. *Internationale Revue der Gesamten Hydrobiologie und Hydrographie*, *68*(5), 605-616.

Friis-Christensen, E., & Lassen, K. (1991). Length of the solar cycle: an indicator of solar activity closely associated with climate. *Science*, *254*(5032), 698-700.

Griffith, D. A., & Peres-Neto, P. R. (2006). Spatial modeling in ecology: the flexibility of eigenfunction spatial analyses. *Ecology*, *87*(10), 2603-2613.

Gulati, R., & Demott, W. (1997). The role of food quality for zooplankton: remarks on the state ‐of ‐the ‐art, perspectives and priorities. *Freshwater Biology*, *38*(3), 753-768.

Hawryshyn, J., Rühland, K. M., Quinlan, R., & Smol, J. P. (2012). Long-term water quality changes in a multiple-stressor system: a diatom-based paleolimnological study of Lake Simcoe (Ontario, Canada). *Canadian journal of fisheries and aquatic sciences*, *69*(1), 24-40.

Higgins, S. N., & Zanden, M. V. (2010). What a difference a species makes: a meta analysis of dreissenid mussel impacts on freshwater ecosystems. *Ecological monographs*, *80*(2), 179-196.

Hopkins, G. J., & Standke, S. J. (1992). Phytoplankton methods manual. Ontario Ministry of the Environment Report. Queen's Printer for Ontario, Ontario, Canada.

Hu, W., Connell, D., Mengersen, K., & Tong, S. (2009). Weather variability, sunspots, and the blooms of cyanobacteria. *EcoHealth*, *6*(1), 71-78.

Huang, J., Higuchi, K., & Shabbar, A. (1998). The relationship between the North Atlantic Oscillation and El Niño ‐Southern Oscillation. *Geophysical Research Letters*, *25*(14), 2707-2710.

Hurrell, J. W. (1995). Decadal trends in the North Atlantic Oscillation: regional temperatures and precipitation. *Science*, *269*(5224), 676-679.

Hutchinson, G. R. (1967). *A Treatise on Lmnology, Vol. II: Introduction to Lake Biology and the Limnoplankton*. New York: Wiley.

Idso, S.B., 1973. On the concept of lake stability. *Limnology and Oceanography. 18*, 681-683.

Interlandi, S. J., Kilham, S. S., & Theriot, E. C. (1999). Responses of phytoplankton to varied resource availability in large lakes of the Greater Yellowstone Ecosystem. *Limnology and Oceanography*, *44*(3), 668-682.

Johnson N. C., Tilman D., and Wedin D.1992. Plant and soil controls on mycorrhizal fungal communities. *Ecology, 73*, 2034–2042.

Kendall, M. G., (1975). Rank correlation methods, 4th edn, London, UK: Cgarles Griffin

Kilham, P., Kilham, S. S., & Hecky, R. E. (1986). Hypothesized resource relationships among African planktonic diatoms. *Limnology and Oceanography*, *31*(6), 1169-1181.

Kilham, S. S., Kott, C. L., & Tilman, D. (1977). Phosphate and silicate kinetics for the Lake Michigan diatom *Diatoma elongatum*. *Journal of Great Lakes Research*, *3*(1), 93-99.

Lampert, W. (1987). Laboratory studies on zooplankton ‐cyanobacteria interactions. *New Zealand Journal of Marine and Freshwater Research*, *21*(3), 483-490.

Lindenschmidt, K. E., & Chorus, I. (1997). The effect of aeration on stratification and phytoplankton populations in Lake Tegel, Berlin. *Archiv für Hydrobiologie*, *139*(3), 317-346.

Lindenschmidt, K. E., & Chorus, I. (1998). The effect of water column mixing on phytoplankton succession, diversity and similarity. *Journal of Plankton Research*, *20*(10), 1927-1951.

Lee, R. B., Gibson, M. A., Wilson, R. S., & Thomas, S. (1995). Long ‐term total solar irradiance variability during sunspot cycle 22. *Journal of Geophysical Research*, *100*(A2), 1667-1675.

[LSEMS] Lake Simcoe Environmental Management Strategy (2008) Lake Simcoe basin wide report. Lake Simcoe Enviornmental Management strategy report. Available from: http://www.lsrca.on.ca/pdf/reports/lsems/basin_wide_report.pdf

MacMynowski, D. G., & Tziperman, E. (2008). Factors affecting ENSO's period. *Journal of the Atmospheric Sciences*, *65*(5), 1570-1586.

Mann, H. B. (1945). Nonparametric tests against trend. *Econometrica, 13*(3), 245-259.

Naddafi, R., Pettersson, K., & Eklöv, P. (2007). The effect of seasonal variation in selective feeding by zebra mussels (*Dreissena polymorpha*) on phytoplankton community composition. *Freshwater Biology, 52*(5), 823-842.

Nalley, D., Adamowski, J., Khalil, B., & Ozga-Zielinski, B. (2013). Trend detection in surface air temperature in Ontario and Quebec, Canada during 1967–2006 using the discrete wavelet transform. *Atmospheric Research,132*, 375-398.

NatureWatch Canada, http://www.naturewatch.ca, IceWatch data from 7 June 2008 to 21 October 2012, University of Ottawa, Canada. Downloaded on 25 April 2014.

Nicholls, K. H., & Carney, E. C. (1979). The taxonomy of Bay of Quinte phytoplankton and the relative importance of common and rare taxa. *Canadian Journal of Botany, 57*(15), 1591-1608.

North, R. L., Barton, D., Crowe, A. S., Dillon, P. J., Dolson, R. M. L., Evans, D. O., Ginn, B. K., Håkanson, L., Hawryshyn, J., Jarjanazi, H., King, J. W., La Rose, J. K. L., León, L., Lewis, C. F. M., Liddle, G. E., Lin, Z. H., Longstaffe, F. J., Macdonald, R. A., Molot, L., Ozersky, T., Palmer, M. E., Quinlan, R., Rennie, M. D., Robillard, M. M., Rodé, D., Rühland, K. M., Schwalb, A., Smol, J. P., Stainsby, E., Trumpickas, J. J., Winter, J. G., & Young, J. D. (2013). The state of Lake Simcoe (Ontario, Canada): the effects of multiple stressors on phosphorus and oxygen dynamics. *Inland Waters, 3*(1), 51-74.

Palmer, M. E., Winter, J. G., Young, J. D., Dillon, P. J., & Guildford, S. J. (2011). Introduction and summary of research on Lake Simcoe: research, monitoring, and restoration of a large lake and its watershed. *Journal of Great Lakes Research*, *37*, 1-6.

Paterson, A. M., Winter, J. G., Nicholls, K. H., Clark, B. J., Ramcharan, C. W., Yan, N. D., & Somers, K. M. (2008). Long-term changes in phytoplankton composition in seven Canadian Shield lakes in response to multiple anthropogenic stressors. *Canadian Journal of Fisheries and Aquatic Sciences*, *65*(5), 846-861.

Pires, L. D., Bontes, B. M., Van Donk, E., & Ibelings, B. W. (2005). Grazing on colonial and filamentous, toxic and non-toxic cyanobacteria by the zebra mussel *Dreissena polymorpha*. *Journal of Plankton Research*, *27*(4), 331-339.

Quinn, C. J., North, R. L., & Dillon, P. J. (2013). Year-round patterns in bacterial production and biomass in Lake Simcoe, Ontario, Canada: are heterotrophic bacteria a significant contributor to low hypolimnetic oxygen. *Inland Waters*, *3*(2), 235-252.

Raubitschek, S., Lücke, A., & Schleser, G. H. (1999). Sedimentation patterns of diatoms in Lake Holzmaar, Germany-(on the transfer of climate signals to biogenic silica oxygen isotope proxies). *Journal of Paleolimnology*, *21*(4), 437-448.

Rawson, D. S. (1956). Algal indicators of trophic lake types. *Limnology and Oceanography*, *1*(1), 18-25.

Reynolds, C. S. (1997). Successional development, energetics and diversity in planktonic communities. In *Biodiversity* (pp. 167-202). New York NY: Springer.

Reynolds, C. S., Huszar, V., Kruk, C., Naselli-Flores, L., & Melo, S. (2002). Towards a functional classification of the freshwater phytoplankton. *Journal of plankton research*, *24*(5), 417-428.

Robarts, R. D., & Zohary, T. (1987). Temperature effects on photosynthetic capacity, respiration, and growth rates of bloom -forming cyanobacteria. *New Zealand Journal of Marine and Freshwater Research*, *21*(3), 391-399.

Rodionov, S. N. (2004). A sequential algorithm for testing climate regime shifts. *Geophysical Research Letters*, *31*, L09204.

Rühland, K., Paterson, A. M., & Smol, J. P. (2008). Hemispheric -scale patterns of climate -related shifts in planktonic diatoms from North American and European lakes. *Global Change Biology*, *14*(11), 2740-2754.

Salmaso, N. (1996). Seasonal variation in the composition and rate of change of the phytoplankton community in a deep subalpine lake (Lake Garda, Northern Italy). An application of nonmetric multidimensional scaling and cluster analysis. *Hydrobiologia*, *337*(1-3), 49-68.

Scheffler, W., & Morabito, G. (2003). Topical observations on centric diatoms (*Bacillariophyceae, Centrales*) of Lake Como (N. Italy). *Journal of Limnology*, *62*(1), 47-60.

Sharma, S., Magnuson, J. J., Mendoza, G., & Carpenter, S. R. (2013). Influences of local weather, large-scale climatic drivers, and the ca. 11 year solar cycle on lake ice breakup dates; 1905–2004. *Climatic change*, *118*(3-4), 857-870.

Sommer, U. (1985). Comparison between steady state and non-steady state competition: experiments with natural phytoplankton. *Limnology and Oceanography*, *30*, 335-346.

Stoermer, E. F. (1978). Phytoplankton assemblages as indicators of water quality in the Laurentian Great Lakes. *Transactions of the American Microscopical Society, 97*(1), 2-16.

Straile, D., & Adrian, R. (2000). The North Atlantic Oscillation and plankton dynamics in two European lakes– -two variations on a general theme. *Global Change Biology*, *6*(6), 663-670.

Straile, D., Livingstone, D. M., Weyhenmeyer, G. A. & George, D. G. (2003). *The Response of Freshwater Ecosystems to Climate Variability Associated with the North Atlantic Oscillation, in The North Atlantic Oscillation: Climatic Significance and Environmental Impact.* J. W. Hurrell, Y. Kushnir, G. Ottersen and M. Visbeck (Ed.), Washington, D. C.: American Geophysical Union.

Strecker, A. L., Beisner, B. E., Arnott, S. E., Paterson, A. M., Winter, J. G., Johannsson, O. E., & Yan, N. D. (2011). Direct and indirect effects of an invasive planktonic predator on pelagic food webs. *Limnology and Oceanography*, *56*(1), 179-192.

Stuecker, M. F., Timmermann, A., Jin, F. F., McGregor, S., & Ren, H. L. (2013). A combination mode of the annual cycle and the El Niño/Southern Oscillation. *Nature Geoscience*, *6*(7), 540-544.

R Development Core Team. (2015) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. Available at: http:// www.R-project.org

Tennant T., (2013) seasonal and long-term trends in the diatom and phytoplankton community composition of Lake Simcoe, Ontario, M.Sc. dissertation. Dept. of Biol., York University, Toronto.

Ter Braak, C. J., & Prentice, I. C. (1988). A theory of gradient analysis. *Advances in Ecological Research, 18*, 271-313.

Tilman, D. (1977). Resource competition between plankton algae: an experimental and theoretical approach. *Ecology*, *58*(2), 338-348.

Tilman, D. (1982). *Resource Competition and Community Structure.* (Monographs in Population Biology 17). Princeton, NJ: Princeton University Press.

Tilman, D., & Kilham, S. S. (1976). Phosphate and silicate growth and uptake kinetics of the diatoms *Asterionella formosa* and *Cyclotella meneghiniana* in batch and semicontinuous culture. *Journal of Phycology*, *12*(4), 375-383.

Tilman, D., Mattson, M., & Langer, S. (1981). Competition and nutrient kinetics along a temperature gradient: An experimental test of a mechanistic approach to niche theory. *Limnology and Oceanography*, *26*(6), 1020-1033.

Toms J. D. and Lesperance M L. 2003. Piecewise regression: a tool for identifying ecological thresholds. *Ecology 84*, 2034–2041.

Vanderploeg, H. A., Liebig, J. R., Carmichael, W. W., Agy, M. A., Johengen, T. H., Fahnenstiel, G. L., & Nalepa, T. F. (2001). Zebra mussel (*Dreissena polymorpha*) selective filtration promoted toxic *Microcystis* blooms in Saginaw Bay (Lake Huron) and Lake Erie. *Canadian Journal of Fisheries and Aquatic Sciences*, *58*(6), 1208-1221.

Watson, S. B., McCauley, E., & Downing, J. A. (1997). Patterns in phytoplankton taxonomic composition across temperate lakes of differing nutrient status. *Limnology and Oceanography*, *42*(3), 487-495.

Winter, J. G., Eimers, M. C., Dillon, P. J., Scott, L. D., Scheider, W. A., & Willox, C. C. (2007). Phosphorus inputs to Lake Simcoe from 1990 to 2003: declines in tributary loads and observations on lake water quality. *Journal of great lakes research*, *33*(2), 381-396.

Winter, J. G., Young, J. D., Landre, A., Stainsby, E., & Jarjanazi, H. (2011). Changes in phytoplankton community composition of Lake Simcoe from 1980 to 2007 and relationships with multiple stressors. *Journal of Great Lakes Research*, *37*, 63-71.

Wood, B. J. B. (1974). Fatty acids and saponifiable lipids. In *Algal physiology and biochemistry* (Vol. 10, pp. 236-265). Berkeley and Los Angeles, LA: Blackwell Oxford.

Yiou, P., Sornette, D., & Ghil, M. (2000). Data-adaptive wavelets and multi-scale singular-spectrum analysis. *Physica D: Nonlinear Phenomena*, *142*(3), 254-290.

Yu, Y. S., Zou, S., & Whittemore, D. (1993). Non-parametric trend analysis of water quality data of rivers in Kansas. *Journal of Hydrology*, *150*(1), 61-80.

Yue, S., Pilon, P., & Cavadias, G. (2002). Power of the Mann–Kendall and Spearman's rho tests for detecting monotonic trends in hydrological series. *Journal of hydrology*, *259*(1), 254-271.

CHAPTER 3:

PREDICTING END-OF-SUMMER DISSOLVED OXYGEN PROFILES IN SMALL STRATIFIED LAKES IN CENTRAL ONTARIO

Jiahua Li[1], Lewis A. Molot[2] and Andrew M. Paterson[3]

1. Department of Biology, York University, Toronto, ON, Canada

2. Faculty of Environmental Studies, York University, Toronto, ON, Canada

3. Dorset Environmental Science Centre, Ontario Ministry of the Environment and Climate Change, Dorset, ON, Canada

SUMMARY

A series of models predicting the uppermost plane of the DO-depleted zone (Zox), spring dissolved oxygen (DOi) profile and end-of-summer dissolved oxygen (DOf) profile built by Molot et al. (1992) were re-analyzed, re-calibrated with more recent data (“A” lakes during 1990-2013), and validated (“B” lakes during 1990-2013). In general, all models built in Molot et al. (1992) were still valid with correlation coefficient, r, between predicted and observed values above 0.83. Different coefficients and/or different transformations and/or additional predictive variables were used in the new series of models that maximized the explained adjusted variance. Variance explained in new models were similar or sometimes higher than older counterparts. The primary significant variables were the same as the original models; we found that additional N variables contributed an additional 13%, 16% and 4% of variance explained in Zox, DOi and DOf models which were considered as secondary significant predictive variables. New models (excluding N variables) were considered adequate because correlation coefficient, r, of validations with “B” lakes were above 0.8 in all three models, r = 0.91 for the DOi model. With the current brownification trend (increased DOC), we expected to see increased DOf at all depths; however, increased reduced-N species (ammonia) in the hypolimnion would offset the brownification effect.

INTRODUCTION

Lake trout (*Salvelinus namaycush*) are an economically, socially and culturally important resource in certain regions of Canada (Scott and Crossman 1973). Although lake trout occur in less than 1% of the lakes in Ontario, this seemingly small fresh water resource represents 20-25% of the world's supply of lake trout (Scott and Crossman 1973). However, lake trout populations are threatened by a variety of influences including loss of habitat, over exploitation and genetic dilution from stocking (Evans et al. 1991). Lake trout is a keystone species that controls the structure of fish communities in lakes where they are present (Plumb and Blanchfield 2009), hence their loss represents not only a loss for human societies but would likely lead to major restructuring of aquatic communities.

Recent evidence of climate-induced lake warming indicates possible loss of habitat for these cold water stenotherms in Precambrian Shield lakes (Schindler et al. 1990, 1996). Lake trout have strict temperature and dissolved oxygen (DO) requirements. Martin and Olver (1980) reported that they are normally found at depths corresponding to 6-13˚C and DO concentrations greater than 4 mg/L during the summer. Their inability to survive extended periods of time above 13˚C implies that they are at risk from a warming climate (Magnuson et al. 1990). Lake trout habitat and therefore population survival can also be affected if hypolimnetic respiration rates are high enough to lower DO to < 5 mg/L.

During thermal stratification, hypolimnetic DO is rarely replenished by mixing with well-aerated surface waters nor does photosynthesis produce much DO in deep waters. Therefore, microbial respiration gradually decreases hypolimnetic DO concentrations after onset of thermal stratification. Several studies have shown that with increasing trophic state and climate change, hypolimnetic DO tends to decrease further due to more substrate for DO consumption and longer periods of DO consumption (De Stasio et al. 1996; Foley et al. 2012).

DO is such an important indicator of the quality of fish habitat that understanding and modeling hypolimnetic DO dynamics has long been a research priority. Various metrics of DO depletion have been developed such as volume-weighted hypolimnetic DO depletion rate (VHOD, mg DO/L/day: Burns 1995; Rosa and Burns 1987; LaBounty and Burns 2007), areal hypolimnetic oxygen depletion rate (AHOD, mg DO/m2/day: Walker 1979; Matthhews and Effler 2006) and anoxic factor (AF, days/year: Nurnberg 1995). The overall extent of hypolimnetic DO depletion can be estimated with these metrics and while these are useful for management purposes, they cannot generate hypolimnetic DO profiles (changes in DO concentration with depth) which are much more ecologically meaningful. Many process-oriented models have been developed but these are primarily research tools requiring extensive data inputs to initialize (Gudimov et al. 2016).

Molot et al. (1992) developed a set of simple empirical models that predict DO profiles at spring overturn (DOi at depth z), end-of-summer (August 31) DO profiles (DOf at depth z) and the top depth of the DO-depleted zone in the hypolimnion of small inland lakes on the Precambrian Shield in central Ontario as functions of lake morphometry, dissolved organic carbon (DOC) and total phosphorus (TP_{so}) at spring overturn. These models are easy to apply and are best viewed as steady state models that predict average profiles over a multi-year period. The profile models were developed with data collected several decades ago (1977-1989) and given the extent of climate change, it is possible that the models may be less accurate today. The purpose of this study was to assess the validity of the original models with more recent data and then to develop new regression models using the more recent data set.

METHODS

Study lakes

The study lakes are located in central Ontario in Haliburton County or the District of Muskoka (Figure 23). The catchments are primarily forested with some cottage development and are underlain by Precambrian metamorphic silicate bedrock (Dillon and Molot 1990) overlain by thin (< 1m) glacial till and soil, although deeper deposits occur (Palmer et al. 2014), except for Glen Lake, which is partially underlain by marble. The lakes are small headwater lakes with the exception of Red Chalk, which is downstream of Blue Chalk. They are softwater, low productivity lakes, and are dimictic, although Dickie exhibits weak stratification (Palmer et al. 2014).

The study lakes are divided into three groups: A, B, md C. The seven “A”' lakes (Blue Chalk, Chub, Crosson, Dickie, Harp, Plastic and Red Chalk) were monitored most intensively and the 14 “C” lakes the least intensively, while monitoring of the eight “B” lakes (Basshaunt, Buck, Glen, Gullfeather, Little Clear, Solitaire, and Walker) was of intermediate intensity. The “C” lakes (Big Porcupine, Bonnechere, Cradle, Crystal, Delano, Kimball, Louisa, Maggie, Nunikani, Pincher, Red Pine, Sherborne, Smoke and Timberwolf) include larger, deeper lakes, but they are exclusively oligotrophic. The range of trophic status, morphometry, and other biological and chemical conditions are similar in the “A” and “B” lakes (Table 1). One station in the central basin of each lake was sampled with the exception of one of the “A” lakes, Red Chalk, which was sampled in both the main and east basins, two distinct basins separated by a shallow narrow channel.

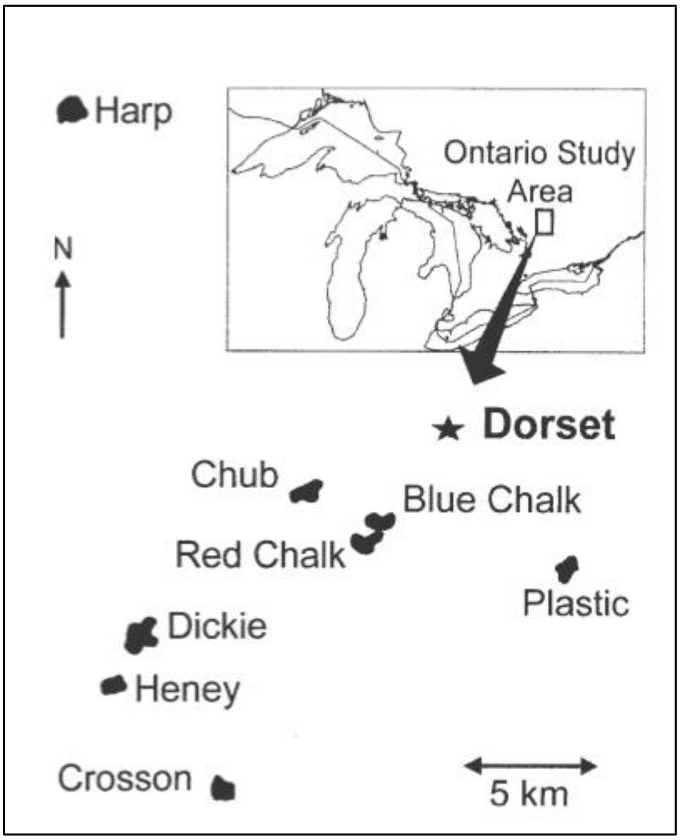

Figure 24. Location of lakes studied near Dorset, Ontario. The inset shows their location in reference to south central Ontario

Data description

Data from 1977 to 1989 were used to develop the original model (Molot et al. 1992, Table 1-3 therein). Data from 1990-2013 (Table 11 and 12) were used to develop the present model. New data were provided by Andrew Peterson, Ontario Ministry of the Environment and Climate Change, Dorset Environmental Science Centre. Sampling frequency varied weekly to monthly during the ice-free season. Revised models were developed with "A" lake data only because "B" and "C" lakes were not sampled every year with sampling frequency variable in the years they were sampled. Ammonia (NH_4^+), total Kjeldahl nitrogen (TKN), and nitrate (NO_3^-) was sampled at three different layers of the lake bi-weekly during the ice-free season and annual mean was calculated. Nitrite (NO_2^-) levels were negligible. Total nitrogen (TN) was calculated as TKN plus NO_3^-. Total organic nitrogen (TON) was calculated as TKN minus NH_4^+. Mean annual ice-free TN and TON were also calculated. DO and temperature were measured at 2 m intervals between 1990 and 1997 and 1 m intervals between 1998 and 2013. Lake morphometry data (MD, VSA and A_0) were taken from Tables 1-3 in Molot et al. (1992).

Sampling procedures are described in Dillon et al. (1988). All (N, TP and DOC) analytical methods are described in Ontario Ministry of the Environment (1983). OMOE changed its DO method in 1993 from the Winkler titration method to an electrochemical sensor (YSI probe).

Table 11. Mean ($\bar{z}$) and maximum depths (z_{max}) (m), lake surface area (Ao, ha), maximum distance across the lake on a line through the sampling station (MD, km), TPso and ice-free DOC, hypolimnetic NH_4, NO_3 and TN and epilimnetic TON in “A”, “B” and “C” lakes. All concentrations are mean annual values for 1990-2013. Units are in µg L^{-1} for P and N and mg L^{-1} for DOC.

Lakes	**$\bar{Z}$**	**Z_{max}**	**A_0**	**MD**	**Zox**	**TP_{so}**	**DOC**	**NH_4**	**NO_3**	**TN**	**TON**
Blue Chalk	8.5	23	52.4	0.9	15	6.7	1.8	91.6	36.1	317.7	166.7
Chub	8.9	27	34.4	1.2	5	9.5	5.6	23.6	114.1	398.0	265.4
Crosson	9.2	25	56.7	1	5	9.9	4.8	35.5	124.3	415.9	260.6
Dickie	5	12	93.6	1.3	5	10.3	5.5	239.3	57.4	646.7	274.4
Harp	13.3	37.5	71.4	1.7	9	5.5	3.9	9.4	171.6	372.2	228.6
Plastic	7.9	16.3	32.1	1.2	11	5.0	2.2	79.1	17.2	325.8	148.2
Red Chalk Main	16.7	38	44.1	0.8	11	4.6	2.7	11.9	150.8	319.2	185.8
Red Chalk East	5.7	19	13.1	0.5	7	5.7	3	358.0	55.3	672.4	201.0
Basshaunt	7.7	24	47.3	0.7	5	7.0	4.2				
Bigwind	10.7	32	111	2.6	9	6.6	3.2				
Buck	10.9	30	40.3	1.2	11	7.0	3.0				
Glen	7.2	15	16.3	0.6	11	10.6	3.7				
Gullfeather	4.8	13	65.9	1.1	5	10.0	6.3				
Little Clear	8.1	25	10.9	0.4	9	6.8	2.6				
Solitarie	13.3	31	124	1.5	15	5.3	2.3				
Walker	6.2	17	68.2	1.1	9	6.2	3.8				
Big Porcupine	7.5	30.5	235	1.9							
Bonnechere	6.4	21.4	105	2		4.4	4.2				
Clear	12.4	33	88.4	1.2							
Cradle	12.4	33	17.9	0.9		4.2	2.2				
Crown	8	30	136	1.1							
Crystal	4.3	17.1	41	1.3		9.1	1.3				
Delano	7.1	18.6	23.9	0.8		5.7	6.6				
Kimball	22	61	213	1.1							
Leonard	6.9	15.2	195	1.7							
Louisa	16.1	61	531	2.8		1.9	3.7				
Maggie	10.2	31	128.6	1.8							
Nunikani	7.9	24	116	2.2		4.6	2.9				
Pincher	6.1	15.5	42.1	1.2		4.4	3.4				
Red Pine	10.1	38.7	365	3.5		4.1	2.8				
Sherborne	9.6	35.1	252	1.3		4.1	2.8				
Smoke	16.2	55	697	4.4		4.3	4.1				
Timberwolf	7.4	20.4	167	2		5.3	5.2				

Table 12. Mean observed DO concentrations (1990-2013) at spring overturn (day of the year 150, DOi) and the end of summer (day of the year 243, DOf), volume/sediment surface area ratios (VSA) for "A" and "B" lakes strata.

Lake	Class	Depth (m)	DOi (mg/L)	DOf (mg/L)	VSA (m)
Blue Chalk	A	15	8.10	3.95	9.42
Blue Chalk	A	17	7.56	1.61	2.87
Blue Chalk	A	19	6.04	0.37	3.78
Chub	A	5	9.02	5.48	9.29
Chub	A	7	8.51	5.34	10.52
Chub	A	9	8.30	5.89	11.86
Chub	A	11	8.03	6.11	11.91
Chub	A	13	7.71	5.62	7.23
Chub	A	15	7.39	4.58	7.89
Chub	A	17	7.06	3.38	7.69
Chub	A	19	6.68	2.28	5.75
Chub	A	21	6.23	1.41	6.32
Chub	A	23	5.33	0.71	3.22
Crosson	A	5	9.62	6.75	9.61
Crosson	A	7	8.84	4.73	7.75
Crosson	A	9	8.44	5.03	10.4
Crosson	A	11	8.25	5.02	9.13
Crosson	A	13	8.13	4.87	7.81
Crosson	A	15	8.11	4.21	6.16
Crosson	A	17	7.93	3.13	7.09
Crosson	A	19	7.70	2.02	5.41
Dickie	A	5	8.78	3.44	3.8
Dickie	A	7	7.73	0.63	3.44
Dickie	A	9	6.49	0.22	1.86
Harp	A	9	9.32	6.87	12.48
Harp	A	11	9.10	6.36	12.4
Harp	A	13	8.93	6.25	13.03
Harp	A	15	8.81	6.02	13.16
Harp	A	17	8.77	5.89	13.4
Harp	A	19	8.60	5.64	13.08
Harp	A	21	8.39	5.23	13.17
Harp	A	23	8.28	4.90	10.07
Harp	A	25	8.25	4.60	7.99
Harp	A	27	8.06	4.28	7.53
Harp	A	29	7.96	3.82	7.17
Harp	A	31	7.57	3.35	5.95
Plastic	A	11	8.68	7.05	4.77
Plastic	A	13	7.63	1.19	2.69
Plastic	A	15	6.95	0.28	0.9
Red Chalk Main	A	13	8.73	7.05	24.5

Red Chalk Main	A	15	8.56	6.82	23.49
Red Chalk Main	A	17	8.46	6.72	17.12
Red Chalk Main	A	19	8.37	6.48	15.76
Red Chalk Main	A	21	8.22	6.19	14.48
Red Chalk Main	A	23	8.09	5.79	13.41
Red Chalk Main	A	25	7.90	5.29	5.14
Red Chalk Main	A	27	7.65	4.85	4.14
Red Chalk Main	A	29	7.33	4.01	6.71
Red Chalk Main	A	31	6.48	2.43	5.79
Red Chalk East	A	9	5.11	0.74	4.07
Red Chalk East	A	11	3.62	0.31	3.08
Red Chalk East	A	13	2.37	0.22	3.7
Red Chalk East	A	15	1.33	0.19	5.2
Basshaunt	B	5	10.43	8.00	6.88
Basshaunt	B	7	9.66	5.76	8.17
Basshaunt	B	9	9.17	4.95	8.82
Basshaunt	B	11	8.97	4.80	7.74
Basshaunt	B	13	8.62	4.10	5.74
Basshaunt	B	15	8.54	3.82	3.71
Basshaunt	B	17	8.28	2.93	7.45
Basshaunt	B	19	8.00	1.27	4.92
Basshaunt	B	21	7.64	0.46	2.36
Bigwind	B	9	10.31	5.82	10.3
Bigwind	B	11	10.16	5.61	8.37
Bigwind	B	13	9.97	6.14	12.41
Bigwind	B	15	9.85	6.41	10.75
Bigwind	B	17	9.82	6.44	12.7
Bigwind	B	19	9.69	6.50	10.81
Bigwind	B	21	9.48	6.38	11.72
Bigwind	B	23	9.32	6.19	10
Bigwind	B	25	9.26	5.96	7.86
Bigwind	B	27	9.09	5.50	6.07
Bigwind	B	29	8.92	4.78	3.12
Bigwind	B	31	8.25	3.90	0.67
Buck	B	11	8.98	7.71	13.44
Buck	B	13	8.66	5.67	13.61
Buck	B	15	8.41	4.46	7.5
Buck	B	17	8.08	3.60	6.13
Buck	B	19	7.87	2.23	7.24
Buck	B	21	7.60	1.24	4.33
Buck	B	23	6.61	0.53	3.87
Buck	B	25	5.82	0.33	5.51
Glen	B	9	2.18	4.79	3.18
Glen	B	11	1.11	0.42	3.58
Glen	B	13	0.92	0.32	2.35
Gullfeather	B	5	9.87	1.67	331

Gullfeather	B	7	9.08	1.03	4.33
Gullfeather	B	9	7.76	0.24	2.1
Little Clear	B	9	6.05	3.40	5.39
Little Clear	B	11	3.61	0.63	4.9
Little Clear	B	13	2.00	0.40	4.2
Little Clear	B	15	1.03	0.37	4.2
Little Clear	B	17	0.83	0.35	5.47
Little Clear	B	19	0.49	0.34	4.3
Little Clear	B	21	0.48	0.33	2.67
Sollitaire	B	15	10.48	8.96	18.91
Sollitaire	B	17	10.14	7.81	17.49
Sollitaire	B	19	10.01	7.11	16.12
Sollitaire	B	21	9.84	6.06	15.06
Sollitaire	B	23	9.77	4.41	12.24
Sollitaire	B	25	9.57	2.24	8.39
Sollitaire	B	27	9.22	0.90	3.72
Walker	B	9	9.62	3.87	3.62
Walker	B	11	8.77	1.69	2.95
Walker	B	13	8.03	0.39	2.02
Walker	B	15	7.64	0.32	0.88

Original models (Molot et al. 1992)

The original Zox, DOi, and DOf models were developed using data from the “A” lakes and tested with the “B” lakes. To find a better fit of DOi model in deep lakes, “A” and “C” lake data were combined for the model development in Molot et al. 1992.

The top depth of the DO depletion zone on August 31 is given by:

$Zox = 1.52 + 28.1/DOC$ Equation 1
$R^2 = 0.91, p < 0.0002$

Initial DO (typically between last week of April and mid-May) at depth z is given by:

$DOi_z = 11.2 – 70.9/Ao + 0.092z$ Equation 2.1
$R^2 = 0.80, p < 0.0001$

In general, the completeness of spring mixing is highly dependent on its surface area and the depth the strata. Mixing introduces DO to deep waters in spring but this effect is more pronounced in large lakes. In smaller lakes with maximum distance across the lake through the sampling station (MD) < 1.4 km, the initial DO at depth z is given by:

$\log_{10} DOi_z = 0.99 – 5.74/Ao + 0.64/z$ Equation 2.2
$R^2 = 0.77, p < 0.0001$

In larger lakes with MD $\geq$ 1.4 km, the initial DO at depth z is given by:

$\log_{10} DOi_z = 1.07 – 6.95/Ao - 0.0043z/MD$ Equation 2.3
$R^2 = 0.71, p < 0.0001$

where Ao is lake surface area.

The end of summer DO concentration at depth z on August 31 is given by:

$\log_{10} DOf_z = 1.83 – 1.91/VSA – 7.06/DOi_z – 0.0013TPso^2$ Equation 3
$R^2 = 0.88, p < 0.0001$

where VSA is the volume to sediment surface area ratio for a stratum. While DOC affected the thickness of the hypolimnion (Zox) with thicker hypolimnia associated with higher DOC lakes, DOC was not a significant independent variable in the original DOi and DOf models. TPso was significant but was of secondary importance: VSA and DOi contributed most of the R^2.

Revising the DO profile models

DOi was standardized to June 1 (Day of the Year 150 or DOY150) because the first sampling trip often occurred in early to mid-May and a June 1 date permitted interpolation between two sampling points. DOf was standardized to August 31 as in Molot et al. (1992).

All modeling procedures were performed with R version 3.0.2 (R Core Develop Team 2015). The best multiple linear regression model for each set of potential predictor variables was chosen based on forward selection (Blanchet et al. 2008) through the *forward.sel()* function of the "packfor" package in R (Dray et al. 2007) using a stopping criterion of alpha = 0.05 (Blanchet et al. 2008). Permutation tests (9999 permutations) were performed to test the significant effects of the testable fractions (Legendre and Legendre, 2012). For each of these analyses, adjusted R^2 (R_{adj}^2) provided unbiased estimates of the variation explained by the fractions (Peres-Neto et al. 2006). Variation partitioning (Borcard et al. 1992) was used to estimate the unique and joint effects of each explanatory variable (e.g. lake morphology and lake chemistry) on dependent variables (i.e. Zox, DOi and DOf). Venn diagrams were used to illustrate the results of variation partitioning. Validations were performed with "B" lakes data set when sufficient validation data were available. "C" lakes were not used to develop new models because of insufficient data. The use of an independent, validation data set provides a more realistic estimate of model prediction error relative to the more traditional re-substitution approach (i.e. bootstrapping, jack-knifing and leave-one-out approaches) because an independent, validation data set tests the model using data not used in its construction (Olden and Jackson, 2000; Sharma and Jackson, 2008).

RESULTS

DO, N, TP and DOC trends

There is little visual evidence of changes in DOi and DOf in all strata of the "A", "B" and "C" lakes between 1977-1989 and 1990-2013 (Figure 24). Most observations fall within 1 mg/L which suggests that DO dynamics did not change profoundly between the two study periods.

Changes in selected ice-free mean N variables (hypolimnetic NH_4, epilimnetic TN and epilimnetic TON) were examined between two study periods (1977-1989 and 1990-2013). No evident trends were observed (Figure 25) with the exception of hypolimnetic NH_4 in Dickie which increased from 85 to 239 µg/L.

Annual mean TPso was lower in 1990-2013 compared to 1977-1989 in 33 "A", "B" and "C" lakes except Blue Chalk which increased 0.9 mg/L (Figure 26) (Glen Lake is ignored here because of an anomalously large decrease from 28.4 to 10.6 $\mu g\ L^{-1}$ in 1990-2013). Decreases ranged from 3 to 47% with a mean of 20%. In contrast, the ice-free mean DOC was higher in 1990-2013 compared to 1977-1989 (Figure 26). DOC increased 3 to 33% in 21 of 27 lakes, did not change in Basshaunt, Glen and Plastic and decreased 4-7% in the remaining 3 lakes.

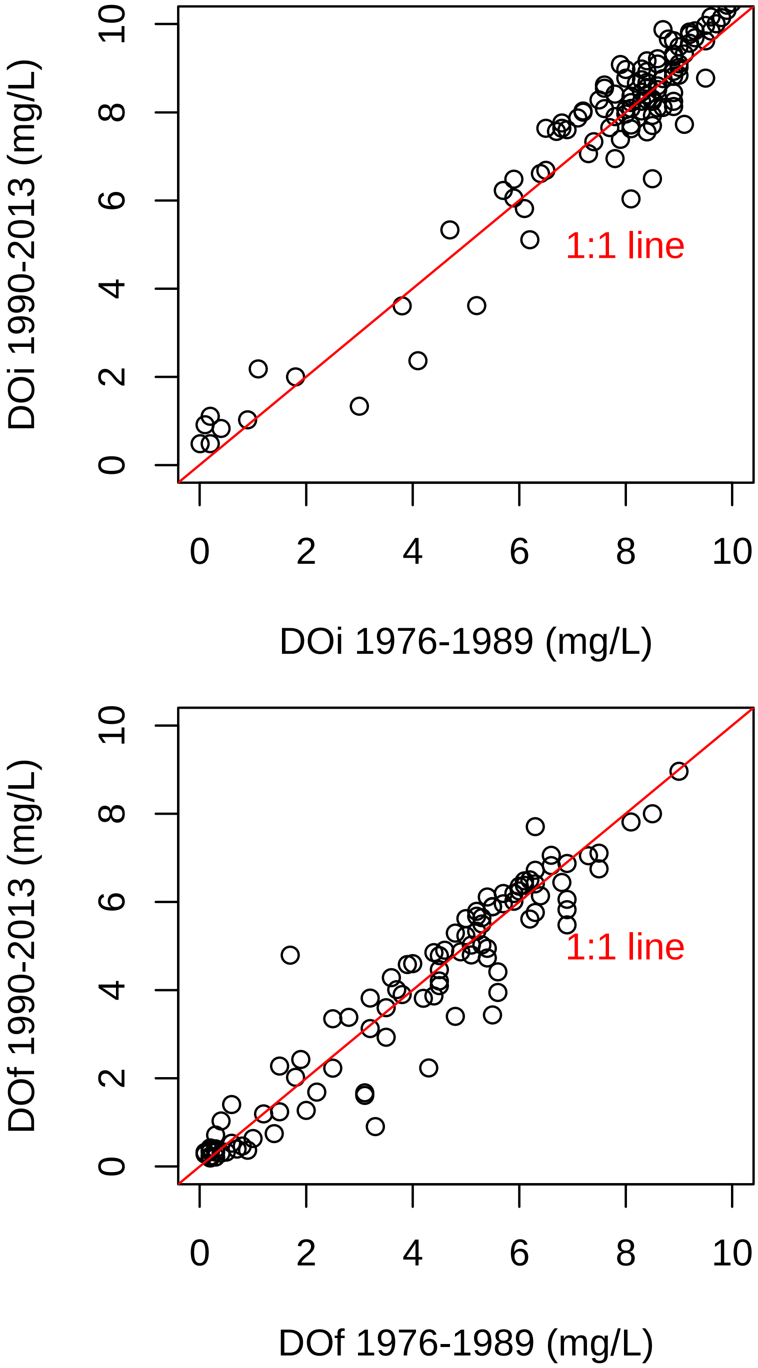

Figure 25 Comparison of mean annual initial and end of summer DO in 1977-1989 to 1990-2013 in “A”, “B” and “C” lakes.

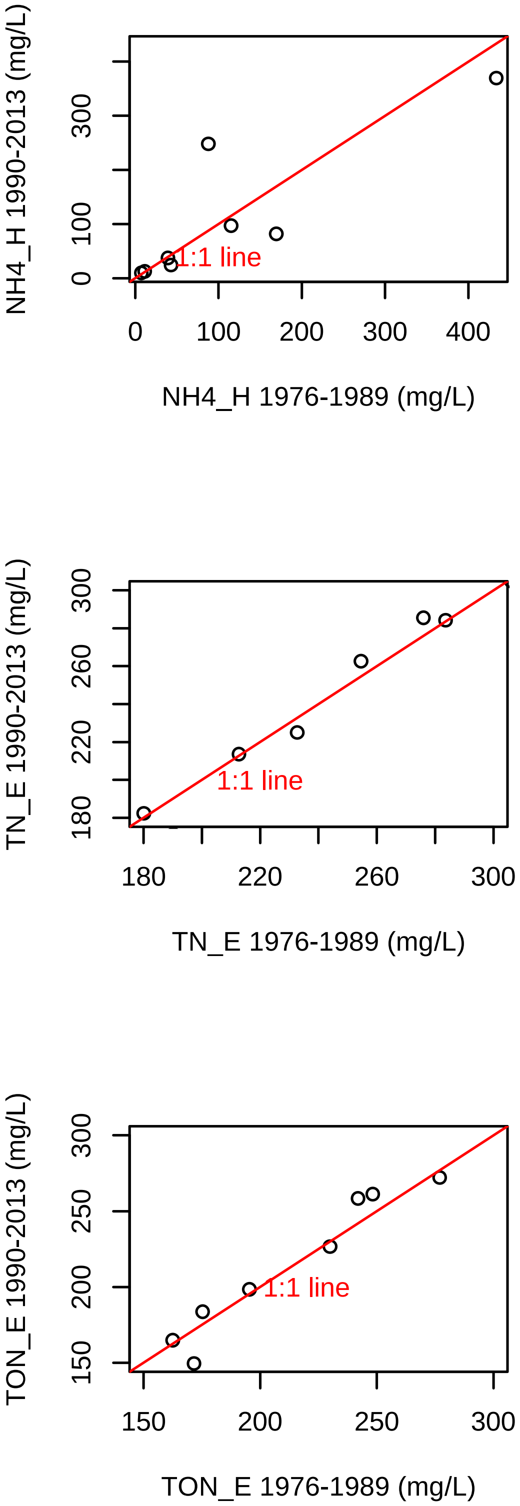

Figure 26. Comparison of mean annual N fractions between 1976-1989 and 1990-2013 in "A" lakes: hypolimnetic ammonium (NH4_H), epilimnetic total N (TN_E) and epilimnetic total organic N (TON-E).

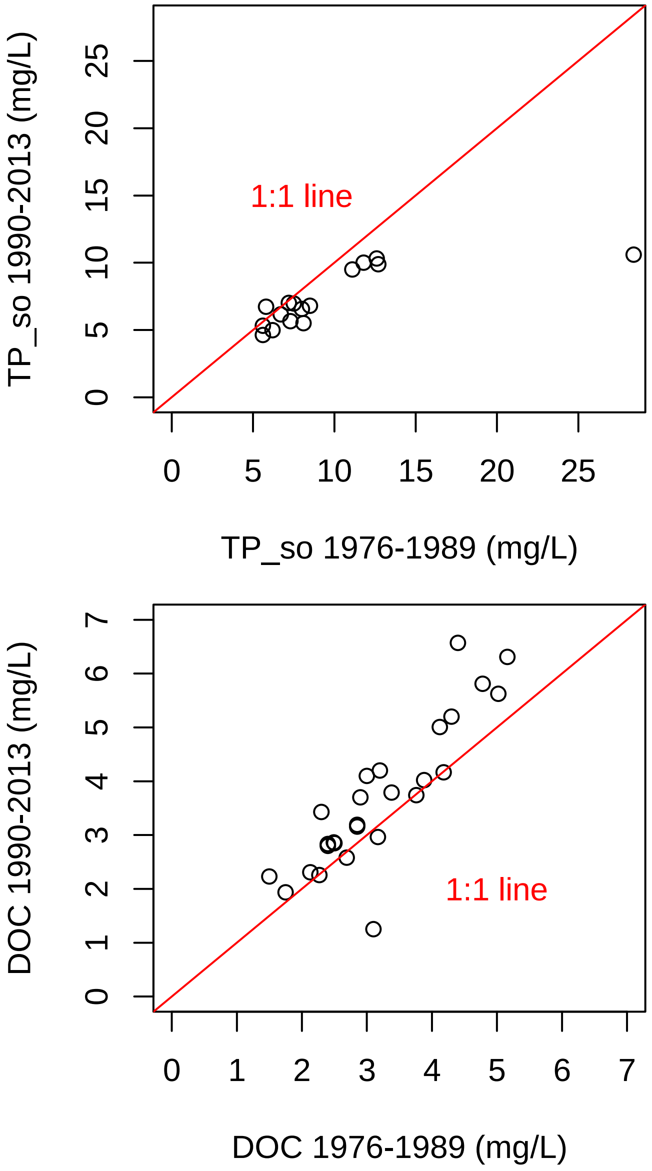

Figure 27. Comparison of mean TPso and annual ice-free mean of DOC in 1977-1989 to 1990-2013 in “A”, “B” and “C” lakes.

Comparison of Molot et al.'s (1992) model predictions to 1990-2013 data

Zox predictions were strongly correlated with post-1990 data with a Pearson product-moment correlation coefficient (r) of 0.872 for combined "A" and "B" lakes. It predicts "A" lakes exceptionally well with $r = 0.936$.

DOi predictions were strongly correlated with post-1990 data with $r = 0.891$ for combined "A" and "B" lakes. Predicted DOi was very strongly correlated with "B" lakes with $r = 0.914$. When data from Harp and Bigwind were excluded because their MD > 1.4 km, r declined to 0.815 for combined "A" and "B" lakes. The correlation for the "B" lakes with Bigwind excluded was 0.865.

DOf predictions were strongly correlated with post-1990 data with $r = 0.837$ for combined "A" and "B" lakes. The DOf model predicts "A" lakes exceptionally well with $r = 0.915$.

Zox model re-calibration with 1990-2013 data from "A" lakes

The uppermost depth exhibiting some DO depletion (Zox) ranged from 5 m in Chub, Crosson, Dickie, Basshaunt and Gullfeather to 15 m in Blue Chalk and Solitarie (Table 1). The revised linear regression model for predicting Zox, the upper most plane of the DO-depleted zone in the "A" lakes, when supplied with DOC and TP_{so} data but without N data, was:

$$Zox = 0.35 + 26.9/DOC \qquad \text{Equation 4.1}$$

$R^2_{adj} = 0.86$, residual standard error: 1.394 with 6 degrees of freedom (DF), F-statistic: 42.37 with 1 and 6 DF, $p < 0.001$.

When hypolimnetic N data were included, the model was:

$$Zox = -13.26 + 47.33/DOC + 40.68/NH4_{hypo} \qquad \text{Equation 4.2}$$

$R^2_{adj} = 0.94$, residual standard error: 0.3468 with 4 DF, F-statistic: 259.2 with 3 and 4 DF, $p < 0.001$.

where $NH4_{hypo}$ is mean ice-free hypolimnetic ammonium. When all N data were included, the model was:

$$Zox = -13.3 + 47.3/DOC + 40.7/NH4_{hypo} + 0.0001*TN_{epi}^2 \qquad \text{Equation 4.3}$$

$R^2_{adj} = 0.99$, residual standard error: 0.3468 with 4 DF, F-statistic: 259.2 with 3 and 4 DF, $p < 0.001$.

where TN_{epi} is mean annual ice-free epilimnetic total nitrogen.

Equations 4.1 to 4.3 is consistent with DOC control of water clarity in non-turbid oligotrophic and mesotrophic waters. Predicted and observed Zox in the "B" lakes (model validation lakes) were highly correlated using equation 4.1 ($r = 0.81$). If the trend to higher DOC continues in the years to come, we can expect Zox to become shallower which is consistent with the finding that higher DOC leads to shallower mixing layers (Molot 2009).

By adding hypolimnetic NH_4 and epilimnetic TN, 8-13% more variation was explained. High hypolimnetic NH_4 leads to shallower Zox in the model, perhaps a reflection of the importance of nitrification in removing oxygen. In contrast, high epilimnetic TN lead to a deeper Zox possibly due to higher primary productivity.

DOi model development with 1990-2013 data from "A" lakes

We developed a linear regression model with "A" lake data to predict the initial spring DO concentrations at the start of each summer period using a standard date, June 1, by interpolating DOi with dates prior to and after June 1.

DOi model without chemistry variables (A lakes):

$DOi(z) = 10.7 - 86.1/Ao - 0.06*z$ Equation 5.1

$R^2_{adj} = 0.73$, residual standard error: 0.8305 with 50 DF, F-statistic: 71.42 with 2 and 50 DF, $p < 0.001$.

DOi model with chemistry variables (A lakes):

$DOi(z) = 12.2 - 63.4/Ao - 0.128*z - 1.7*\log_{10}(NH4_{hypo}) + 349.2/TON_{epi}$ Equation 5.2

$R^2_{adj} = 0.89$, residual standard error: 0.5405 with 48 DF, F-statistic: 101.8 with 4 and 48 DF, $p < 0.001$.

where Ao is the lake surface area, z is depth of the strata, $NH4_{hypo}$ is annual ice-free hypolimnetic ammonia and TON is mean annual ice-free total organic nitrogen concentration in the epilimnion.

Addition of N-species as independent variables increased R_{adj}^2 slightly from 0.73 to 0.89 in Equation 5.2 and standard error decreased from 0.83 to 0.54 mg/L. We also observed a good correlation between observed and predicted spring DO profile in B lakes using Equation 5.1 (Pearson product-moment correlation coefficient r = 0.91). Standard errors were relatively low, 0.83 and 0.54 respectively, which makes these models informative.

However, Molot et al. (1992) found that lake fetch affected DOi so we used data from the "A" lakes without Harp Lake to develop a multiple linear regression model for lakes with MD < 1.4 km:

$DOi = 19.1 - 63.5/A_0 - 0.2*z - 2*\log_{10}(NH_{4\ hypo}) - 0.3(TON_{epi})^{0.5}$ Equation 5.3

$R^2_{adj} = 0.90$, Residual standard error: 0.54 with 36 DF, F-statistic: 92 with 4 and 36 DF, $p < 0.001$. However, this equation is not very different from equation 5.2 for all lakes. Furthermore, R_{adj}^2 and standard error were not significantly improved either.

Equation 2.1 and Equation 5.1 are quite similar in that lake surface area and the depth of strata, z, were highly significant independent variables in both regressions although the z coefficient was positive in Equation 2.1 but negative in Equation 5.1.

DOf model development with 1990-2013 data

We used “A” lake data to develop a model for the end-of-summer DO concentration (DOf). Mean DOi and VSA were significant independent variables in the multiple linear regression analysis while mean TP at spring overturn (TPso) and mean DOC were weakly significant. R^2_{adj} for DOf was higher when DOf was logarithmically transformed. The regression model for predicting DOf concentration at depth z is:

DOf model without TP, DOC or N variables (1990-2013):

$$\log_{10}(DOf)_z = -0.3 + 0.016\ DOi^2 - 1.16/VSA \qquad \text{Equation 6.1}$$

$R^2_{adj} = 0.86$, Residual standard error: 0.1726 with 50 DF, F-statistic: 158.5 with 2 and 50 DF, $p < 0.001$.

DOf model with both TP and DOC, without N variables (1990-2013):

$$\log_{10}(DOf)_z = -0.9 + 0.016DOi^2 - 1.13/VSA + 2.83/TP_{so} + 0.01*DOC^2 \qquad \text{Equation 6.2}$$

$R^2_{adj} = 0.87$, Residual standard error: 0.1634 with 48 DF, F-statistic: 90.42 with 4 and 48 DF, $p < 0.001$.

The order of presentation of the independent variables in Equations 6.1 and 6.2 from left to right is the order in which they were selected by the forward selection procedures. The first two variables in each equation, DOi and VSA, accounted for almost 86% of the variance. TP and DOC accounted for an additional 1.5% of variance explained in equation 6.2 and the p values were marginal for these two variables. The most likely explanation for their insignificance is their small range: all lakes were oligotrophic with TPso < 10 µg/L and the highest DOC in the “A” lakes was only 5.8 mg/L. Standard errors converted from log units back to DO normal values are 1.7 and 1.6 mg/L respectively, which is acceptable for making this model informative.

The DO method was changed from Winkler titration to sensors in 1993. We therefore developed a DOf model using only sensor data (1993-2013).

DOf model (1993-2013) with all TP, DOC and N variables:

$$\log_{10}(DOf) = 0.66 + 0.015*DOi^2 - 0.85/VSA - 0.03*\mathrm{sqrt}(NH_{4\ hypo}) - 0.48\log_{10}(A_0) \qquad \text{Equation 6.3}$$

$R^2_{adj} = 0.93$, Residual standard error: 0.1252 with 48 DF, F-statistic: 160.2 with 4 and 48 DF, $p < 0.001$.

When the 1990-1993 Winkler DO data were excluded, R^2_{adj} increased slightly from 0.87 in equation 6.2 to 0.93 in equation 6.3 but only ammonium was significant.

The revised models (equations 6.1 to 6.3) are similar to original equation 3 (Molot et al. 1992) in that DOi and VSA contribute the majority of the variability to R^2.

Validation of Models

We tested each of the models by comparing predictions with data collected from the "B" lakes. The Zox model predicted reasonably well with a correlation coefficient of 0.81 for Equation 4.1. The DOi model predicted reasonably well with a correlation coefficient of 0.91 for Equation 5.1. The DOf models predicted reasonably well with a correlation coefficient of 0.81 for Equation 6.1.The correlation of original equation 3 predictions with 1977-1989 "B" and "C" lake data (Molot et al. 1992) was higher (0.88), perhaps because the higher sampling frequency before 1990 produced more accurate estimates of spring and end of summer DO.

Impact of Brownification on DOf

As observed in this study and others (Yan et al. 2008; Keller et al. 2008), DOC levels have increased over time in lakes located in central Ontario. It is unclear whether the increases are related to higher export from wetlands or lower in-lake losses (Dillon and Molot 2005). Nevertheless, changes in DOC concentration in lakes can have profound impacts on thermal structure (Molot 2009) and ecosystem services such as recreation, fishing and drinking-water supply. Using equation 6.2, DOf is expected to be 1.6 times higher with doubling of DOC (Figure 27). However, caution should be exercised here since the mechanism linking increased DOC to higher hypolimnetic DO is not clear.

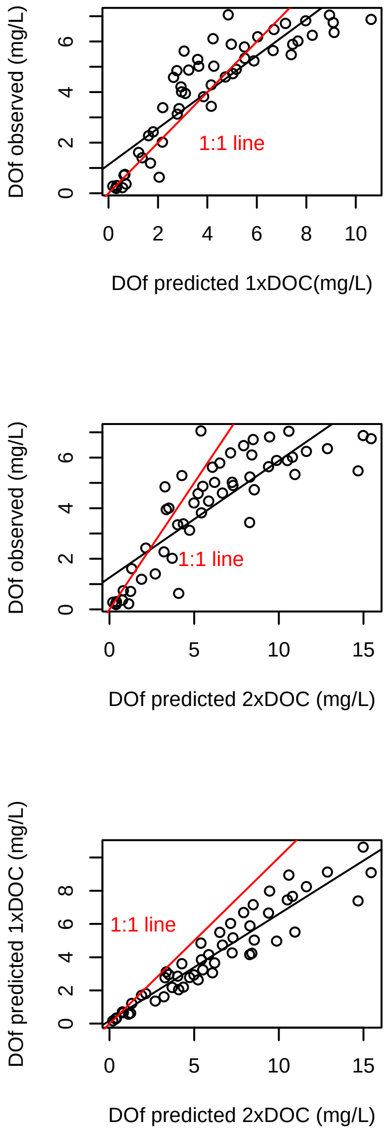

Figure 28. Comparison among predicted 1xDOC, 2xDOC and observed end-of-summer DO (DOf) with red line indicating 1:1.

Impact of Higher NH_4 on DOf

We used equation 6.3 to examine impact of a doubling of hypolimnetic NH_4 on DOf at constant DOi and constant TPso. The results (Figure 28) show that doubling hypolimnetic ammonia would result in large decreases in DOf.

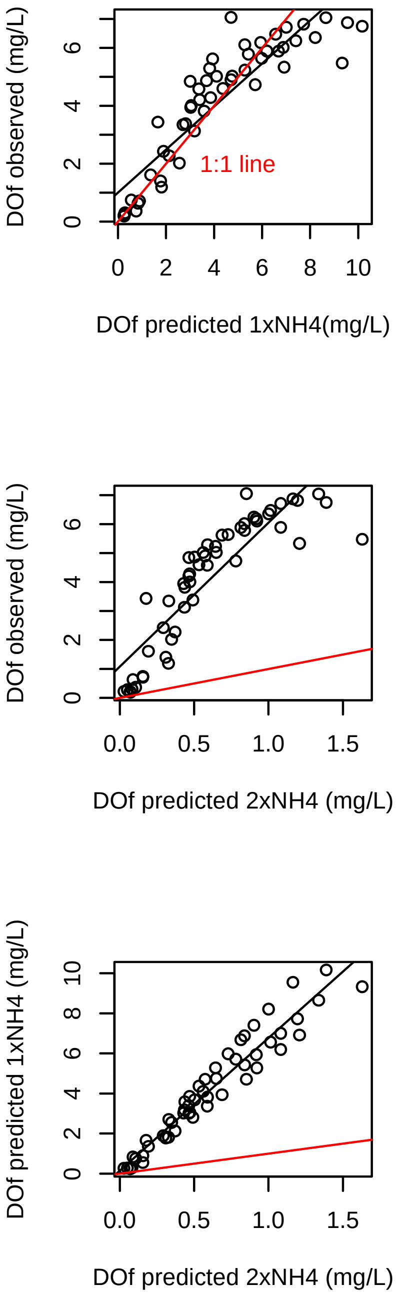

Figure 29. Comparison among predicted $1xNH_4$, $2xNH_4$ and observed end-of-summer DO (DOf) with red line indicating 1:1.

DISCUSSION

This study clearly demonstrates that a relatively simple model consisting of one to a few independent variables (lake chemistry and lake morphometry) is capable of accurately predicting Zox, DOi profiles and DOf profiles in small inland oligotrophic and oligo-mesotrophic lakes. It also shows that the models developed with 1977-1989 data are still valid for 1990-2013. The structures of the recalibrated models are similar to the models of Molot et al. (1992).

The most important variable for predicting the uppermost depth exhibiting some DO depletion (Zox) in these lakes is DOC because of the role that DOC plays in controlling solar radiation penetration and thus water clarity in non-turbid oligotrophic and mesotrophic waters (Molot 2009). High concentrations of DOC limits light penetration depth and leads to shallower mixing zones, hence Zox will be shallower in coloured lakes. As observed in this study and others (Yan et al. 2008; Keller et al. 2008), DOC levels have increased over time in these lakes. As a result, the uppermost depth exhibiting some DO depletion is expected to become shallower. Adding hypolimnetic NH_4 and epilimnetic TN improves R_{adj}^2 but the Zox model without N variables is still reliable for small lakes in the region. High hypolimnetic NH_4 leads to shallower Zox in the model, perhaps a reflection of the importance of nitrification in removing oxygen. Nitrification consumes 2 moles of O_2 per mole of N as ammonia (4.6 mg of O_2 per mg of N as ammonium). Hypolimnetic NH_4 concentrations can exceed 100 µg/L in some of the A lakes during the summer, hence, losing 0.5 mg/L or more of DO to nitrification from initial concentrations between 10 to 12 mg DO/L is not out of the question. Newbry et al. (1981) estimated that up to 40% of the total hypolimnetic oxygen demand in the Cherokee Reservoir in the TVA system was consumed by nitrification (Newbry et al. 1981). Clevinger et al. (2013) estimated that about nitrification consumed about 30% of the sediment oxygen demand in Lake Erie.

In contrast, high epilimnetic TN lead to a deeper Zox possibly due to higher primary productivity with a corresponding higher O_2 production.

Lake morphology plays an important role in predicting DOi and DOf profiles in these small, nutrient poor inland lakes due to its impact on mixing energy. In all DOi models, DOi at depth z is a function of the lake surface area (a proxy for wind speed and thus mixing energy) and the depth below the surface. Some of the DOi models included MD (Equation 2.3), VSA (Equation 5.3), hypolimnetic NH_4 (equation 5.3) and epilimnetic TON (Equation 5.3) as independent variables but their partial R^2 values were small and morphology was clearly dominant. Adding VSA and N slightly improved R_{adj}^2 but the simplest DOi model (Equation 5.1) is valid without them.

DOf was highly dependent on the initial DO concentration, which is in turn a function of surface area, depth (i.e. aspects of lake morphometry) and to a much smaller extent water chemistry. Adding hypolimnetic NH_4 and lake area slightly improved R_{adj}^2 but the model is valid with only DOi and VSA (Equation 6.2) which suggests that vertical transport of DO is negligible in comparison to horizontal DO movement during summer stratification in these small lakes (Imboden and Joller 1984, Staehr et al. 2012).

The empirical structure of these models limits their ability to respond to changing environmental conditions. For example, adding TP and N variables allows the DOi and DOf models to be used in scenarios with higher nutrients but only slightly higher since this extends the model into regions not included during calibration: we do not know how appropriately calibrated DOi and DOf models should behave in high nutrient lakes.]

Climate change is not explicitly included in the models, only implicitly. For example, a longer ice-free season will not affect the model because the start and end dates (June 1 and August 31) are fixed. However, increasing hypolimnetic temperatures could be reflected in lower DOf although there is little evidence that significant hypolimnetic warming has occurred since the original model appears valid when predictions are compared to 1990-2013 data.

Lake DOC concentrations have increased over the last 25 years (Dillon and Molot 2005; Yan et al. 2008) and this should lead to higher Zox and consequently lower DOf especially in deeper waters. However, end-of-summer DOf was positively associated with DOC (Equation 6.1). There is little evidence of significant decreases in DOf (Figure 24).

In conclusion, lake trout habitat did not change very much between the two study periods (1976-1989 and 1990-2013) since little evidence of changes in spring and en-of-summer DO in “A”, “B” and “C” lakes were observed (Figure 24). However, with current trend of increasing DOC, improved lake trout habitat would be expected and expansion in their range would also be expected. However, high hypolimnetic ammonia was observed with lower end-of-summer DO expected in Dickie Lake but no such trend in other study lakes.

Table 13. Comparison of models predicting Zox, and DO concentration profiles at spring overturn (DOi), and end-of-summer DO concentration profiles (DOf) in Molot et al. (1992) and this study.

Time Period	1977-1989 (Molot et al. 1992)	1990-2013 (This study)
	$Zox = 1.52 + 28.1/DOC$ $R^2_{adj} = 0.91$	$Zox = 0.35 + 26.9/DOC$ $R^2_{adj}=0.86$
	$DOi = 11.16 - 71.22/A0 - 0.092*z$ $R^2_{adj} = 0.80$	$DOi(z)=10.7-86.1/A0-0.06z$ $R^2_{adj}=0.73$
		$DOi(z)=12.2-63.4/A0-0.1*z-1.7*\log_{10}(NH_4\_H)+349.2/TON\_E$ $R^2_{adj}=0.89$
	$\log_{10}DOi\ (MD< 1.4km) = 0.99 – 5.74/A0 + 0.64/z$ $R^2_{adj} = 0.83$	$DOi\ (MD< 1.4km) = 8.7 – 83.2/A0+0.5*sqrt(VSA)-0.09*z+6.7/TP$ $R^2_{adj}=0.80$
		$DOi\ (MD< 1.4km) = 19.1-63.5/A0-0.2*z-2*\log_{10}(NH_4\_H)-0.3*\sqrt{}TON\_E$ $R^2_{adj}=0.90$
	$\log_{10}(DOf) = 1.83 - 1.91/VSA - 7.06/DOi - 0.0013*TP\_so{\wedge}2$ $R^2_{adj} = 0.88$	$\log_{10}(DOf)_z=-0.9+0.016DOi^2-1.13/VSA+2.83/TP+0.01*DOC^2$ $0\ R^2_{adj}=.87$
		$\log_{10}(DOf)_z=0.66+0.015DOi^2-0.859/VSA-0.04\sqrt{}NH_4\_H-0.48*\log_{10}(A_0)$ $R^2_{adj} = 0.92$

REFERENCES

Blanchet, F. G., Legendre, P., & Borcard, D. (2008). Forward selection of explanatory variables. *Ecology*, *89*(9), 2623-2632.

Borcard, D., Gillet, F., & Legendre, P. (2011). *Numerical Ecology with R*. New York, NY: Springer

Borcard, D., Legendre, P., & Drapeau, P. (1992). Partialling out the spatial component of ecological variation. *Ecology*, *73*(3), 1045-1055.

Burns, N. M. (1995). Using hypolimnetic dissolved oxygen depletion rates for monitoring lakes. *New Zealand journal of marine and freshwater research*, *29*(1), 1-11.

Clevinger, C. C., Heath, R. T., & Bade, D. L. (2014). Oxygen use by nitrification in the hypolimnion and sediments of Lake Erie. *Journal of Great Lakes Research*, *40*(1), 202-207.

De Stasio, B. T., Hill, D. K., Kleinhans, J. M., Nibbelink, N. P., & Magnuson, J. J. (1996). Potential effects of global climate change on small north-temperate lakes: Physics, fish, and plankton. *Limnology and Oceanography*, *41*, 1136-1149.

Dillon, P. J., & Molot, L. A. (1990). The role of ammonium and nitrate retention in the acidification of lakes and forested catchments. *Biogeochemistry*, *11*(1), 23-43.

Dillon, P.J. and L.A. Molot. 2005. Long-term trends in catchment export and lake retention of dissolved organic carbon, dissolved organic nitrogen, total iron and total phosphorus: The Dorset, Ontario study, 1978-1998. *Journal of Geophysical Research – Biogeosciences, 110, No.G01002, doi:1029/2004JG000003.*

Dillon, P.J., Nicholls, K.H., Locke, B.A., De Grosbois, E., & Yan, N.D. 1988. Phosphorus-phytoplankton relationships in nutrient-poor soft-water lakes in Canada. *Proceedings of the International Association of Theoretical and Applied Limnology, 23,* 258-264.

Dray, S., Legendre, P., & Blanchet, G. (2009). packfor: Forward Selection with permutation. Available at: http://R-Forge.R-project.org/projects/sedar/.

Evans, D.O., Casselman, J.M., and Wilcox, C.C. 1991. Effects of exploitation, loss of nursery habitat, and stocking on the dynamics and productivity of lake trout in Ontario lakes. Lake Trout Synthesis, Ontario Ministry of Natural Resources, Toronto, Ontario

Foley, B., Jones, I. D., Maberly, S. C., & Rippey, B. (2012). Long‐term changes in oxygen depletion in a small temperate lake: effects of climate change and eutrophication. *Freshwater Biology*, *57*(2), 278-289.

Gudimov, A., McCulloch, J., Chen, J., Doan, P., Arhonditsis, G., & Dittrich, M. (2016). Modeling the interplay between deepwater oxygen dynamics and sediment diagenesis in a hard-water mesotrophic lake. *Ecological Informatics*, *31*, 59-69.

Imboden, D. M., & Joller, T. (1984). Turbulent mixing in the hypolimnion of Baldeggersee (Switzerland) traced by natural radon‐222. *Limnology and Oceanography*, *29*(4), 831-844.

LaBounty, J. F., & Burns, N. M. (2005). Characterization of Boulder Basin, Lake Mead, Nevada-Arizona, USA–based on analysis of 34 limnological parameters. *Lake and Reservoir Management*, *21*(3), 277-307.

Legendre, P., & Legendre, L. F. (2012). *Numerical Ecology*. 3rd English edn. Amsterdam, NH: Elsevier.

Magnuson, J.J., Meisner, J.D., and Hill, D.K. 1990. Potential changes in the thermal habitat of Great Lakes fish after global climate warming. *Transactions of the American Fisheries Society*. *119*(2), 254–264.

Martin, N.V., and Olver, C.H. 1980. The lake charr, *Salvelinus namaycush*. *In* Charrs: salmonid fishes of the genus *Salvelinus*. *Edited by* E.K. Balon. Dr. L.W. Junk Publishers, Amsterdam, Netherlands: W. Junk Publishers.

Matthews, D. A., & Effler, S. W. (2006). Long‐term changes in the areal hypolimnetic oxygen deficit (AHOD) of Onondaga Lake: Evidence of sediment feedback. *Limnology and Oceanography*, *51*(1part2), 702-714.

Molot, L.A., Color of Aquatic Ecosytems. 2009. In *Encyclopedia of Inland Waters*, G.E. Likens (Ed.), Volume 2, 657-663, Elsevier,

Molot, L.A., P.J. Dillon, B. Clark and B. Neary. 1992. Predicting end-of-summer oxygenprofiles in stratified lakes. *Canadian Journal of Fisheries and Aquatic Sciences, 49*, 2363 -2372.

Newbry, B. W., R. A. Jones and G. F. Lee. 1981. Assessment and analysis of eutrophication of Tennessee River system impoundments. Proceedings ASCE Symposium on Surface Water Impoundments, American Society of Civil Engineers p. 413-424.

Nürnberg, G. K. (1995). Quantifying anoxia in lakes. *Limnology and oceanography*, *40*(6), 1100-1111.

Olden, J. D., & Jackson, D. A. (2000). Torturing data for the sake of generality: how valid are our regression models?. *Ecoscience, 7*(4), 501-510.

Palmer, M. E., Yan, N. D., & Somers, K. M. (2014). Climate change drives coherent trends in physics and oxygen content in North American lakes. *Climatic change*, *124*(1-2), 285-299.

Peres-Neto, P. R., Legendre, P., Dray, S., & Borcard, D. (2006). Variation partitioning of species data matrices: estimation and comparison of fractions. *Ecology*, *87*(10), 2614-2625.

Plumb, J. M., & Blanchfield, P. J. (2009). Performance of temperature and dissolved oxygen criteria to predict habitat use by lake trout (*Salvelinus namaycush*). This paper is part of the series "Forty Years of Aquatic Research at the Experimental Lakes Area". *Canadian Journal of Fisheries and Aquatic Sciences*, *66*(11), 2011-2023.

Rosa, F., & Burns, N. M. (1987). Lake Erie central basin oxygen depletion changes from 1929–1980. *Journal of Great Lakes Research*, *13*(4), 684-696.

Schindler, D. W., K. G. Beaty, E. J. Fee, D. R. Cruikshank, E. R. DeBruyn, D. L. Findlay, G. A. Linsey, J. A. Shearer, M. P. Stainton, and M. A. Turner. (1990). Effects of climatic warming on lakes of the central boreal forest. *Science 250*, 967–970.

Schindler, D. W., S. E. Bayley, B. R. Parker, K. G. Beaty, D. R. Cruikshank, E. H. Fee, E. U. Schindler, and M. P. Stainton. 1996a. The effects of climatic warming on the properties of boreal lakes and streams at the Experimental Lakes Area. *Limnology and Oceanography 41*, 1004–1017.

Scott, W. B., & Crossman, E. J. (1973). *Freshwater fishes of Canada*. Bulletin, 184. Ottawa, ON: Fisheries Research Board of Canada.

Sharma, S. and Jackson, D.A. (2008). Predicting smallmouth bass incidence across North America: Comparison of statistical approaches. *Canadian Journal of Fisheries and Aquatic Sciences*, *65*, 471-481.

Staehr, P. A., Christensen, J. P., Batt, R. D., & Read, J. S. (2012). Ecosystem metabolism in a stratified lake. *Limnology and Oceanography*, *57*(5), 1317-1330.

Walker, W. W. (1979). Use of hypolimnetic oxygen depletion rate as a trophic state index for lakes. *Water Resources Research*, *15*(6), 1463-1470.

Yan, N. D., Somers, K. M., Girard, R. E., Paterson, A. M., Keller, W., Ramcharan, C. W., Ramcharan, J. A. Rusak, R. I., George E. M., & Gunn, J. M. (2008). Long term trends in zooplankton of Dorset,Ontario, lakes: the probable interactive effects of changes in pH, totalphosphorus, dissolved organic carbon, and predators. *Canadian Journal of Fisheries and Aquatic Sciences*, *65*(5), 862-877.

SUMMARY AND CONCLUSIONS

Lakes in Ontario are influenced by different environmental drivers. We were interested in understanding how hypolimnetic dissolved oxygen content and phytoplankton community composition in Lake Simcoe have changed in response to changes in several drivers.

Lake Simcoe was subjected to regional climate change, phosphorus (P) enrichment followed by reduction and invasive dreissenid mussels during the study period (1980-2012). We successfully explained the collective impact of these drivers on volume-weighted hypolimnetic dissolved oxygen (VWHDO) dynamics. P reductions and dreissenids have had a positive impact on hypolimnetic DO but the lengthening of the thermal stratification season and a slight warming of the hypolimnion led to an increase in the extent and duration of hypolimnetic DO depletion, countering the beneficial impact of P reductions and mussel invasions. Our findings agree with Foley et al. (2012) who found that the duration of thermal stratification and the extent of hypoxia in Blelharm Tarn (U.K.) increased between 1968 and 2008, even though their DO depletion rate decreased. We were surprised to observe that dreissenid mussels had a positive influence on VWHDO dynamics both on spring and ice-free minimum VWHDO, perhaps because mussels divert organic matter inshore. Without invasive mussels, the mean minimum ice-free VWHDO during 1997-2012 would have been 1.8 mg/L, assuming spring VWHDO and length of thermal stratification remained at pre-1990 levels, instead of the 4.2 mg/L observed. These results suggest that continuing climate change in the region will make it more difficult to achieve the VWHDO target of 7 mg O_2/L (LSPP 2009) without additional efforts to reduce P loads below the associated target of 44 tonnes/yr (Nicholls 1997).

Annual and seasonal phytoplankton community composition change was also associated with multiple drivers. Dominant annual phytoplankton genera at all eight stations changed from *Stephanodiscus* spp. (1980 to early 1990s) to *Fragilaria* spp. (later 1990s and 2000s) which was linked to silicate availability, water temperature, dreissenid mussels invasion and light availability. Seasonal phytoplankton composition analysis showed that the shallow site (C1) and deeper sites (E51 and K42) were associated with different groups of phytoplankton. In addition, summer phytoplankton composition differed from fall composition at the deeper sites (E51 and K42).

Phytoplankton composition at sites C1 and K42 significantly different pre- and post-dreissenids. Redundancy analyses found that environmental variables (e.g. S, total phosphorus, Secchi depth, silica, total Kjeldahl nitrogen, nitrate and nitrite), dressenid mussels and large climatic oscillation patterns were significantly correlated with annual/seasonal phytoplankton community composition. Environmental variables and dreissenids individually explained little variance in phytoplankton composition but they collectively explained a large portion of the variation, perhaps indicating that dreissenids acted on phytoplankton composition indirectly by changing the environmental conditions. At some sites, large climatic oscillation indexes and Moran's Eigenvector Maps-generated-temporal-pattern variables were also strongly correlated with phytoplankton community composition indicating that complex patterns in climatic drivers such precipitation, temperature, wind, etc. may play an important role in structuring phytoplankton community composition.

It is important to pursue and probably enhance the P reduction strategies in Lake Simcoe to ensure ice-free minimum VWHDO is above the critical level for coldwater fisheries. Curbing greenhouse emissions is also needed on the global scale to reduce the length of thermally stratified period and water temperature thus improving their habitat over current conditions. While dreissenid mussels have had a beneficial influence on oxygen dynamics, they have had an impact of unknown quality on phytoplankton community composition. Food web structure undoubtedly changed in the past three or more decades thus research on food quantity/quality and biological interaction change might be needed.

A series of models predicting the uppermost plane of the DO-depleted zone (Zox), spring dissolved oxygen (DOi) profile and end-of-summer dissolved oxygen (DOf) profile in small inland lakes in central Ontario developed by Molot et al. (1992) were re-analyzed, re-calibrated with more recent data (Dorset "A" lakes during 1990-2013), and validated (Dorset "B" lakes during 1990-2013). Unlike Lake Simcoe, there is little evidence of changes in spring and end of summer DO in the "A", "B" and "C" lakes between 1977-1989 and 1990-2013. The original models developed by Molot et al. (1992) are still valid. Revised models suggest that if current trends of increasing DOC and NH_4 continue, then higher Zox, lower DOi and higher DOf are expected in the future, assuming only small changes in P.

REFERENCES

Foley, B., Jones, I. D., Maberly, S. C., & Rippey, B. (2012). Long‐term changes in oxygen depletion in a small temperate lake: effects of climate change and eutrophication. *Freshwater Biology*, *57*(2), 278-289.

[LSPP] Lake Simcoe Protection Plan. 2009. Ontario (Canada): Ontario Ministry of the Environment; Ministry of Natural Resources, and Lake Simcoe Region Conservation Authority report [cited July 5 2015]. Available from: http://www.ontario.ca/environment-and-energy/lake-simcoe-protection-plan

Nicholls, K. H. (1997) A Limnological basis for a Lake Simcoe phosphorus loading objective, *Lake and Reservoir Management*, 13:3, 189-198.