Final Status Assessment of BUI 13 Phytoplankton and Zooplankton Populations for the Bay of Quinte Area of Concern

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Abstract

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The “Degradation of Phytoplankton and Zooplankton Populations” Beneficial Use Impairment (BUI 13) for the Bay of Quinte Area of Concern (AOC) has been designated as impaired since the site was listed under the Canada – United States Great Lakes Water Quality Agreement. This analysis provides a final assessment by Fisheries and Oceans Canada, based on the 47 years of sampling for Project Quinte from 1972 – 2018. This assessment provides a refined list of 10 metrics (reduced from the possible 26 proposed in 2017) for phytoplankton, zooplankton and microbial populations that can be used to track improvements in the system. This assessment finds that while there have been some long-term improvements in the planktonic communities indices, food web function and trophic transfer continue to be significantly impaired. Based on all 10 metrics being applied in a weight of evidence approach, our recommendation is that BUI 13 continues to be impaired, primarily due to persistent eutrophication in this system. Recommendations are provided which support the establishment of a phosphorus management plan, and for continued monitoring of both plankton and fish communities that are simple to implement by local monitoring agencies tracking the recovery of this ecosystem.

Résumé

Currie, W.J.S., Bowen, K.L., Niblock, H.A., Fitzpatrick, M.A., Rozon, R., Munawar, M., Cuddington, K.M.D., Ward, C. Final status assessment of BUI 13 phytoplankton and zooplankton populations for the Bay of Quinte Area of Concern. Can. Manuscr. Rep. Fish. Aquat. Sci. 3261: x + 114 p.

La altérations des utilisations bénéfiques « Dégradation des populations de phytoplancton et de zooplancton » (AUB 13) pour le secteur préoccupant (SP) de la baie de Quinte a été désignée comme altérée depuis que le site a été inscrit en vertu de l’Accord relatif à la qualité de l’eau dans les Grands Lacs signé entre le Canada et les États-Unis. Cette analyse fournit une évaluation finale par Pêches et Océans Canada, basée sur les 47 années d'échantillonnage pour le projet Quinte de 1972 à 2018. Cette évaluation fournit une liste affinée de 10 paramètres (réduits des 26 possibles proposés en 2017) pour le phytoplancton, le zooplancton et les populations microbiennes qui peuvent être utilisés pour suivre les améliorations du système. Cette évaluation révèle que bien qu'il y ait eu des améliorations à long terme dans les indices des communautés planctoniques, la fonction du réseau trophique et le transfert trophique continuent d'être considérablement altérés. Sur la base des 10 paramètres appliqués dans le cadre d'une approche fondée sur le poids de la preuve, notre recommandation est que le AUB 13 continue d'être altéré, principalement en raison de l'eutrophisation persistante dans ce système. Des recommandations sont fournies qui soutiennent l'établissement d'un plan de gestion du phosphore et pour la surveillance continue du plancton et des communautés de poissons qui sont simples à mettre en œuvre par les agences de surveillance locales qui suivent le rétablissement de cet écosystème

Executive Summary

This is the final assessment of BUI 13 “Degradation of Phytoplankton and Zooplankton populations” for the Bay of Quinte Area of Concern (AOC) based on the 47 years of sampling of the completed Project Quinte. This collaborative project includes data from a number of provincial and federal agencies including Fisheries and Oceans Canada (DFO), Ontario Ministry of Environment Conservation and Parks (MECP), Ontario Ministry of Natural Resources and Forestry (MNRF) and Environmental and Climate Change Canada (ECCC). This report is intended as an update and refinement of the proposed targets and last status assessment (Currie et al. 2017a,b). In the Bay of Quinte, the phytoplankton and zooplankton population impairment is bottom-up in nature due to excess nutrients and fundamentally linked to that of BUI 8: Eutrophication. There have been major reductions in phosphorus loadings to the Bay since the 1970s when phosphorus was implicated as the cause of eutrophication in the Great Lakes and both Canadian and US governments mandated policies of phosphorus reduction. AOC activities intended to further reduce nutrient input into the Bay have been among the main management actions undertaken in the AOC since the program started in 1987 [Bay of Quinte Remedial Action Plan (BQ RAP) 1993]. The decrease in total phosphorus after controls implemented in 1978 has been a steady slow decline since the early 1980s and summer phosphorus levels are now sourced primarily from legacy phosphorus released from the sediments (see S1 State of the Ecosystem). Nitrogen limitation, which was common in the 1970s in the upper bay (Belleville), lessened slightly during the 80s and 90s, but in the 2000s, especially in the last decade, became much more common exacerbated by internal phosphorus loading. In a comparison of paired nearshore and offshore sampling stations during 2017 and 2018 for the upper and middle bay we found no differences in physical, chemical or biological measurements suggesting the upper bay in particular is well mixed between these zones.

Prior to 1995 the Bay of Quinte was in what is termed a “turbid” phase, with significant algal blooms and poor water clarity, but as nutrients decreased, it entered a “clear” phase in 1995 which has been maintained since, through there has been been a subsequent slight decrease in clarity and continued algal blooms in the upper bay since 2000. This does not appear to be a typical “alternative stable state” or regime shift, but rather is best described by a steady change from one state to another (a transient). The apparent sudden clearing of the upper bay in 1995 had been regularly attributed to the arrival zebra mussels, but it is now apparent that the Bay cleared first, allowing macrophytes to expand in the upper bay which facilitated settlement of the current zebra mussel population in the nearshore region.

With the decline in phosphorus, there has been a matching overall decrease in the biomass of phytoplankton, zooplankton and planktivorous fishes within the upper Bay of Quinte. In spite of this, the upper Bay of Quinte continues to be afflicted by algal blooms and eutrophic conditions, and the system produces more phytoplankton biomass per unit total phosphorus than any other system monitored by the DFO Great Lakes Laboratory for Fisheries and Aquatic Sciences (S2.1). Filamentous forms of diatoms and cyanobacteria continue to dominate the composition of the phytoplankton, which are not ideal food for zooplankton. The better algal food fraction as indicated by members of the Chrysophycea has improved, but only slightly in the upper bay. The importance of nitrogen limitation in the Bay of Quinte is seen in the strong characteristic correlations between Total Kjeldahl Nitrogen (TKN) and biomass, which are generally stronger than those of TP.

The microbial food web was not included in the previous analysis of BUI 13 (Currie et al. (2017b) because it had only been collected since 2000, but it is known to be important to food web function so is included here (S2.2). During the last decade there has been an sharp increase in bacterial biomass with a subsequent decrease in their primary grazers, the heterotrophic nanoflagellates (HNF). As such, system productivity is being shunted into bacteria and not passed on to higher trophic levels. It is not clear if this is due to the increase in summer temperatures affecting bacterial growth rates or through another environmental effect influencing the HNF.

Zooplankton biomass is at an all time low in recent years in the upper bay (S2.3). There have been some improvements in species composition, with eutrophic species such as *Eubosmina* and *Chydorus* becoming less dominant, though the biomass of these species per unit TP is unexpectedly less than that in middle bay indicating a deficiency in converting nutrient into zooplankton biomass. Bottom-up forces continue to dominate zooplankton production and composition in the upper bay and typical indicators of top-down planktivory such as size or species composition are very weak. There has been an increase in the percentage of *Daphnia galeata mendotae*, a large-bodied zooplankton which is much preferred as a food item compared to small-bodied eutrophic species such as *Chydorus*. However these improvements are moderate at best, and much better improvements have been seen at the middle bay (Hay Bay) station. The hope and expectation is that as nutrient conditions and the algal community improves in the upper bay that these metrics will show improvement.

An assessment of food-web trophic transfer has been included by comparing simplified trophic level biomass ratios and comparing these over time and to other sites (S2.4). The first is the bottom-up ratio of phytoplankton biomass to total phosphorus. This indicates how well the system ultimately produces algal biomass given its nutrient loadings. The Bay of Quinte at all sites has the highest biomass/TP of any site monitored by this lab, including other eutrophic embayments. This in not entirely surprising since Eutrophication is a known issue in the Bay of Quinte. The second ratio of zooplankton to phytoplankton biomass is driven both by bottom-up and top-down forcings since zooplankton hold an intermediate position within the food web. However, in the upper bay there has been no improvements over time as would be expected given the reductions in nutrients, even though improvements in this ratio have been seen in middle bay and lower bay. Given that there is no change in the phytoplankton:TP ratio, this is entirely driven by the disparity in zooplankton production, which is stunted in the upper bay. The final top-down metric is the planktivorous fishes to zooplankton biomass ratio. This metric has shown some improvements due to the overall reduction of fish biomass in the upper bay which is at an all time low in recent years which might be due to increased piscivory.

There is an expectation of reduced zooplankton as relative piscivores dominate (a trophic cascade) as planktivorous fishes are consumed. However this is not effectively happening in the upper bay of Quinte, likely due to the stagnation of the zooplankton ability to convert the poor quality phytoplankton into biomass for the upper trophic levels. As such we have proposed two additional fish-based metrics that can utlilize the MNRF trawl and gillnet surveys of the Bay of Quinte. The first ratio is that of the normalized ratio of Gizzard Shad to combined Yellow Perch + Gizzard Shad biomass. This is a useful relationship because Gizzard Shad eat at the very base of the detrital (seston) food web, but Yellow Perch become increasingly piscivorous/ benthivorous as they increase in dominance and size. As such this indicates how much the bottom-up forces still dominate in the upper bay compared to middle bay.The second fish metric is the ratio of piscivore to planktivore biomass. Planktivore biomass has always been tightly linked to nutrients, but not piscivores. However, since 2013 there has been an increased coupling of the total fish – planktivorous fish – zooplankton – phytoplankton system. The system was loosely interconnected previously but system productivity is now almost entirely bottom-up driven including that of piscivore biomass. This can be seen in coherency between the piscivores with the lower trophic groups and the ratio of piscivores to planktivores that has recently increased rapidly to an all-time high such that there are significantly more piscivore biomass than their planktivore prey (inverted biomass pyramids).

We previously set out the framework to assess BUI 13 by evaluating the functionality of the food web and identify disruptions to the flow of energy from nutrients into the higher trophic levels. In Currie et al. 2017b, we proposed a suite of 26 indicators of food web functionality. To re-assess the status of BUI 13, we now propose a refined set of 10 metrics for phytoplankton, microbial, zooplankton and trophic ratios that are equally capable of determining impairment for a continued monitoring plan.

Based on all of the metrics in a weight of evidence approach, our evaluation of BUI 13 in the upper Bay of Quinte is that, while there have been some long-term improvements, it continues to be significantly impacted in its food web functioning. We therefore recommend the status of BUI 13 should continue to be *impaired*.

Given the ongoing impairment of BUI 13, we recommend continued monitoring of the upper and middle Bay of Quinte and also recommend including Conway in the lower bay. At a minimum we propose monthly sampling of zooplankton, phytoplankton and bacteria. But given that it will introduce a level of uncertainty in the data, especially for phytoplankton and bacterial measures during the summer period, we highly recommend fortnightly sampling. It is important to maintain that certain environmental measures are collected as well including a YSI EXO sonde cast (or similar device), Secchi disk (light attenuation measures are preferred) and water chemistry (including total phosphorus, nitrogen and dissolved nutrients). Our most recent study shows few differences between nearshore and offshore stations in the upper and middle bay. As a consequence, if the long-term monitoring sites of Belleville, Napanee and Hay Bay cannot be maintained, then other sites closer to shore will likely be comparable, although river mouths should be avoided. Due to the recent shifts in fish community composition since 2013, it is highly recommended that food web analysis of piscivore production be reexamined since there are indications that fish populations have become food limited. As such, we highly support the continued trawl and gillnet monitoring of fishes by MNRF in the Bay of Quinte and recommend additional sampling and diet analysis of larval fishes and young-of-year (YOY), especially in the nearshore.

Table 0.1 Food web function metrics for the Bay of Quinte from 2017 and revised in 2020

Original 2017 Metrics	Revised 2020 Metrics
Trophic Biomass Ratios • Phytoplankton to TKN (P:TKN) • Zooplankton to TKN (Z:TKN) • Planktivorous fish to TKN (PlanktF:TKN) • Zooplankton to Phytoplankton (Z:P) • Z:P regression with TP and TKN • Daphnia to Phytoplankton (D:P) • Z:P regression with Planktivorous fish • Planktivorous fish to Zooplankton (F:Z) • Planktivorous fish to Daphnia (F:D) • Planktivorous fish to Phytoplankton (F:P) • Piscivorous fish to Zooplankton (Pis:Z) • Piscivorous fish to Phytoplankton (Pis:P) • Yellow perch to White perch (Yperch:Wperch)	*Trophic Biomass Ratios* 1. Phytoplankton : TP < 50000 2. Zooplankton : Phytoplankton > 0.062 3. Planktivorous Fishes : Zooplankton < 0.22
Phytoplankton Indicators • A decrease in nuisance phytoplankton to less than 50% of the biomass (i.e. < 2.5 g m^{-3} in the upper bay offshore based on existing target for BUI 8 of phytoplankton biomass of 4 - 5 g m^{-3}) • A decreasing trend in filamentous and colonial / mucilaginous Cyanobacteria (both in terms of biomass and relative composition) • A decreasing trend in filamentous diatoms (biomass and % composition). • An increase in the more edible phytoplankton taxa to approximately 30% of the biomass • An increase in the biomass of Chrysophyceae with an ideal target of over 15% of total phytoplankton biomass	*Phytoplankton Indicators* 4. Total filamentous biomass < 2.0 g m^{-3} (annual mean) 5. Biomass of colonial cyanobacteria < 1 g m^{-3}, 80% of the time (with a minimum of 6 samples collected evenly through the season) 6. Ratio of Chrysophyceae to Total Filamentous phytoplankton > 10% (annual mean)
Microbial Indicators • None proposed	*Microbial Indicators* 7. Bacterial biomass < 920 mg m^{-3} (annual mean)
Zooplankton Indicators • An increase in cladoceran mean size • Decreased *Chydorus* biomass relative to the values in the 1970s and early 1980s • Reduced proportion of *A. vernalis* relative to total adult cyclopoid biomass • A trend toward increasing diversity index scores in the zooplankton community over time • A trend of increasing large Daphnia over time • A trend of increasing % *D. galeata mendotae* • An increase in the proportion of *Mesocyclops* relative to other cyclopoids • A declining proportion of eutrophic rotifer taxa	*Zooplankton Indicators* *8.* *Chydorus* biomass < 7.5 mg m^{-3} dry-weight 9. % Dap*hnia galeata mendotae* > 16% *10.* *Eubosmina* : (DGM + *Eubosmina*) < 0.52

Introduction: Evaluation of BUI 13 using a functional food web approach.

The Bay of Quinte is a eutrophic embayment on the north east end of Lake Ontario that has been studied by DFO since the early 1970s to evaluate ecosystem response to phosphorus control. Nutrients, chlorophyll a, phytoplankton and zooplankton populations were the early focus of 'Project Quinte' with the intent of discovering impact on fish populations (Christie 1972), which later morphed in to a large collaborative project with many agencies (Johnson and Hurley 1986). Under the 1987 Great Lakes Water Quality Agreement between Canada and the United States the Bay of Quinte was declared an Area of Concern, an area that experienced high levels of environmental harm. It was deemed to have 10 Beneficial Use Impairments (BUIs) which included BUI 8 "Eutrophication or undesirable algae" and BUI 13 "Degradation of phytoplankton and zooplankton communities" [Bay of Quinte Remedial Action Plan (BQ RAP) 1993].

Evaluation of BUI 13 "*Degradation of phytoplankton and zooplankton populations*" has a history of disagreement and lack of focus, leading to frustration for RAP managers needing to assess this BUI (see draft discussion in Boyd 2019). Assessment suggestions have been piecemeal in the past, with little integration and support by other Great Lakes scientists (e.g. Irvine and Murphy 2009, Hartig et al. 2020). Most Great Lakes AOC's had BUI 13 status as *Requires Further Assessment* (RFA) for many years since phytoplankton and zooplankton populations were rarely sampled together or analyzed. However, many of these have now been assessed including: Thunder Bay (Currie et al. 2015), Niagara River (Rozon et al. 2016), Hamilton Harbour (Currie et al. 2018a), Toronto Harbour (Currie et al. 2018b) and Detroit River (Fitzpatrick et al. 2020). The Bay of Quinte is an exception where BUI 13 has always had a status of *Impaired*, however little information was given for this original classification, with most discussion confounded with eutrophication rather than plankton populations (BQRAP 1993), making a clear assessment very difficult. A number of approaches were put forward over the years to assess impairment in this BUI including targets for phytoplankton biomass and zooplankton size. However, no theoretical framework was provided to support these targets and there was little consensus on what they were measuring. The Bay of Quinte was the first AOC assessed using the comprehensive functional food web approach for BUI 13 which continued to be assessed as *impaired* (Currie et al. 2017a, 2017b).

A characteristic of a functional food web is an effective, operational food web which transfers energy from primary producers to higher trophic levels without significant disruption (Reynolds 2008, Jeppesen et al. 2005). Fundamentally, the determination of impairment in BUI 13 centers around whether the populations of species within phytoplankton or zooplankton are being negatively affected by the environment, and are considered together because they are dependent on each other. Furthermore, we know that many aquatic systems have significant microbial populations which can be on par with the biomass of phytoplankton (Munawar et al. 2011). There is no other BUI which allows this important food web component to be included. The use of a food web approach to determine impairment is particularly suited to BUI 13, because plankton fit neatly into the aquatic food web between other biological impairments, e.g. BUI 8 *Eutrophication or undesirable algae*, and BUI 3 *Degradation of fish and wildlife populations*. These BUIs have been proposed as screenings for further analysis of BUI 13 (Boyd 2019). BUI 13 is also tightly related to BUI 14: *Loss of fish and wildlife habitat* (particularly submerged aquatic vegetation) and BUI 6: *Degradation of benthos*. A food web approach to BUI

13 also complies to the *Great Lakes Water Quality Agreement* explicit commitment to an ecosystem approach.

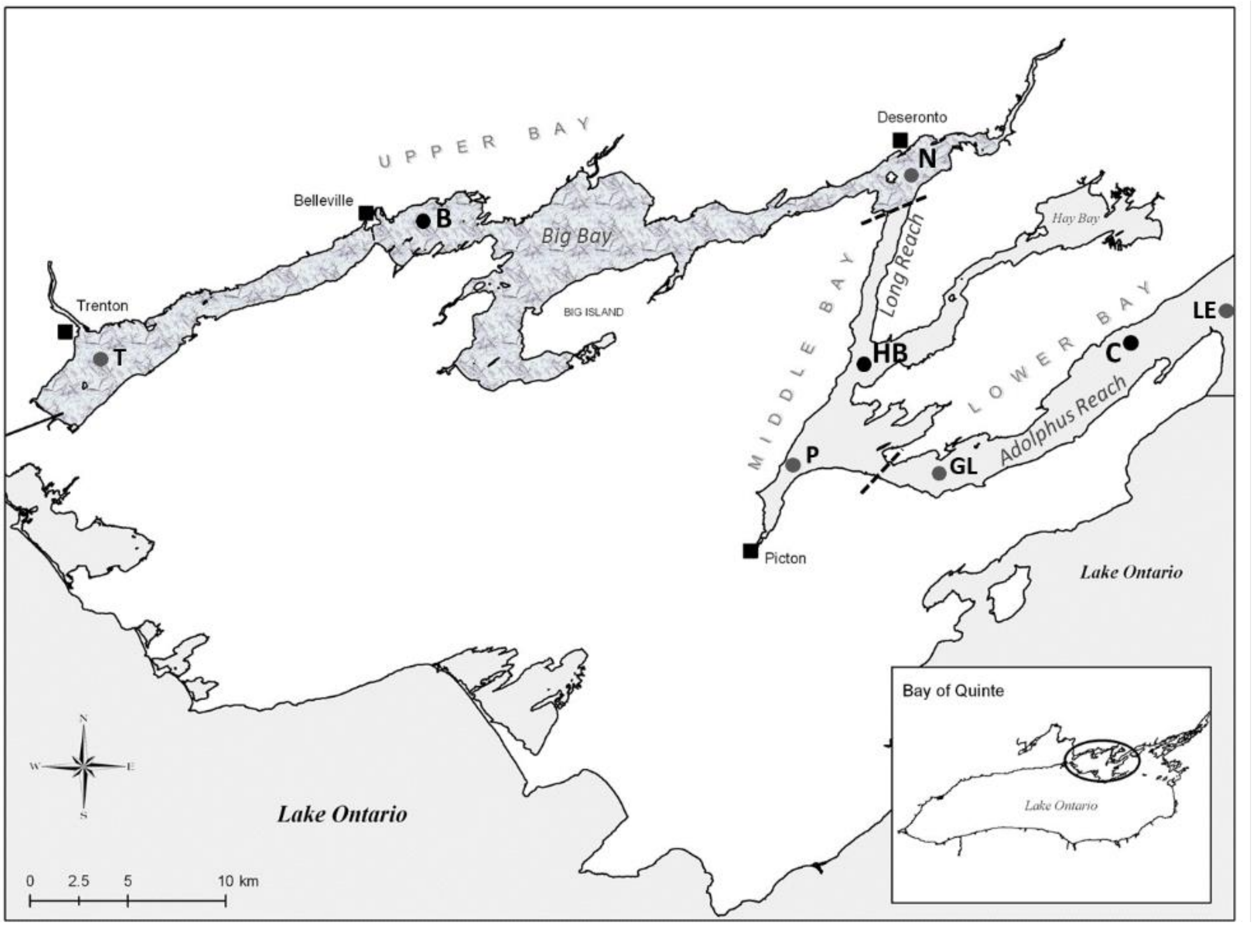


Figure 0.1 *The Bay of Quinte location on Lake Ontario (inset) and long-term sampling locations for Project Quinte. The stations with solid black circles: Belleville (B), Hay Bay (HB) and Conway (C) are sites with complete time-series. Dashed lines delimit the upper, middle and lower bay sectors. The patterned fill delimits the boundary of the AOC*.

As a shallow eutrophic embayment, the Bay of Quinte is expected to have a robust, dense planktonic community (McCauley and Kalff 1981, Scheffer 1998). Nutrients will stimulate growth of microbes and phytoplankton, which are consumed by zooplankton, which are in turn consumed by planktivorous fishes which are themselves eaten by predatory fishes, birds and reptiles. Phytoplankton and zooplankton populations are different from the other portions of the aquatic food web (e.g. fish, benthos) in that they have very short generation times which leads to notoriously high spatial and temporal variation (Hutchinson 1961, Kratz et al. 1987, Folt and Burns 1999, Levy and Klein 2004),and exacerbates analysis of trends. This can be overcome by appropriate temporal resolution in sampling and sufficiently long time-series (Cassie 1962, Currie et al. 2015). Although plankton population data will always be variable, methods such as temporal amalgamation (e.g. annual data) and data transformation (e.g. log transformation to account for zero shifting) can help. It is also worth noting that on longer time frames, shallow water systems can characteristically switch between two states: a turbid phase dominated by phytoplankton, and a clear-water phase with increased macrophyte growth and reduced phytoplankton biomass (Scheffer et al. 1993, Scheffer and Carpenter 2003). If the change persists between these phases it is known as a regime shift or alternative stable state and can occur rapidly on time scales of less than a year, driven by changes in nutrients, chemistry, climate or water levels (Scheffer and Carpenter 2003).

The composition and quantity of the plankton community can be driven by factors which are classified as “bottom-up” and “top-down” processes (McQueen et al. 1986; Carpenter et al. 2001). Much of the early science of limnology was focused on the study of bottom-up processes, which maintain that phytoplankton populations, and ultimately the structure of the food web are controlled by limiting factors such as nutrients or light (Harvey et al. 1935, Lindeman 1942, McCombie 1953, but see McQueen et al. 1986 for discussion). However, beginning in the 1960s it became clear that in some systems, consumption at the top of the food web can change the characteristics of the food web, in terms of zooplankton composition and size (Hrbáček et al. 1961, Brooks and Dodson 1965) and even phytoplankton biomass (Carpenter et al. 1987, Jeppesen et al. 2003). The theory of trophic cascade (Carpenter et al. 1987) quickly led to the idea that biomanipulation of lakes could change the community composition to one favored by managers, though the results were highly variable in outcome (Perrow et al. 1997, Meijer et al. 1999, Drenner and Hambright 2002).

To apply an ecosystem approach for assessment to the Bay of Quinte, it was necessary to have sufficient information on the physical environment (light transmission, temperature etc.), nutrients (phosphorus, nitrogen, silica), and the composition of phytoplankton, zooplankton and ideally benthos and fishes. The Bay of Quinte is fortunate to be the site of one of Canada’s long term ecological research (LTER) programs, ‘Project Quinte’, which had been collecting whole ecosystem information from 1972 – 2018, though not continuously for every measure (see: Currie and Frank 2015). While many stations had periodic sampling, the primary stations for Project Quinte that were continuously sampled were Belleville (upper bay), Napanee (upper bay transition to middle bay), Hay Bay (middle bay) and Conway (lower bay) (Fig. 0.1). Belleville station is representative of the Area of Concern which is defined as the portion of the Bay of Quinte from Trenton to Deseronto.

To make analysis possible, it was necessary to unite many disparate datasets from a number of agencies including: DFO (phytoplankton and microbial loop, zooplankton and fish habitat datasets), OMNRF (fishes), OMOE (now MECP) (water chemistry), and ECCC (climate). These data were date-matched, QA/QC’ed and combined into a single “flat” file with > 1M entries. Monthly and annual datasets were compiled so that the drivers of the biological communities could be assessed. The time-series of nutrients, phytoplankton, zooplankton, benthos and fishes illustrates that all of these measurements have changed during the period of sampling for Project Quinte.

This dataset was explored for underlying relationships using a number of approaches (time-series analysis, regression and correlation, principal components analysis). This involved the investigation of thousands of relationships to determine independence and fundamental correlations between measurements such that statistical relationships could be developed. These data were used to determine the fundamental time-stanzas within the time-series. Using physical drivers such as temperature, clarity, and total phosphorus, the Quinte data-series formed four distinct time stanzas that could be grouped into two major phases. These time

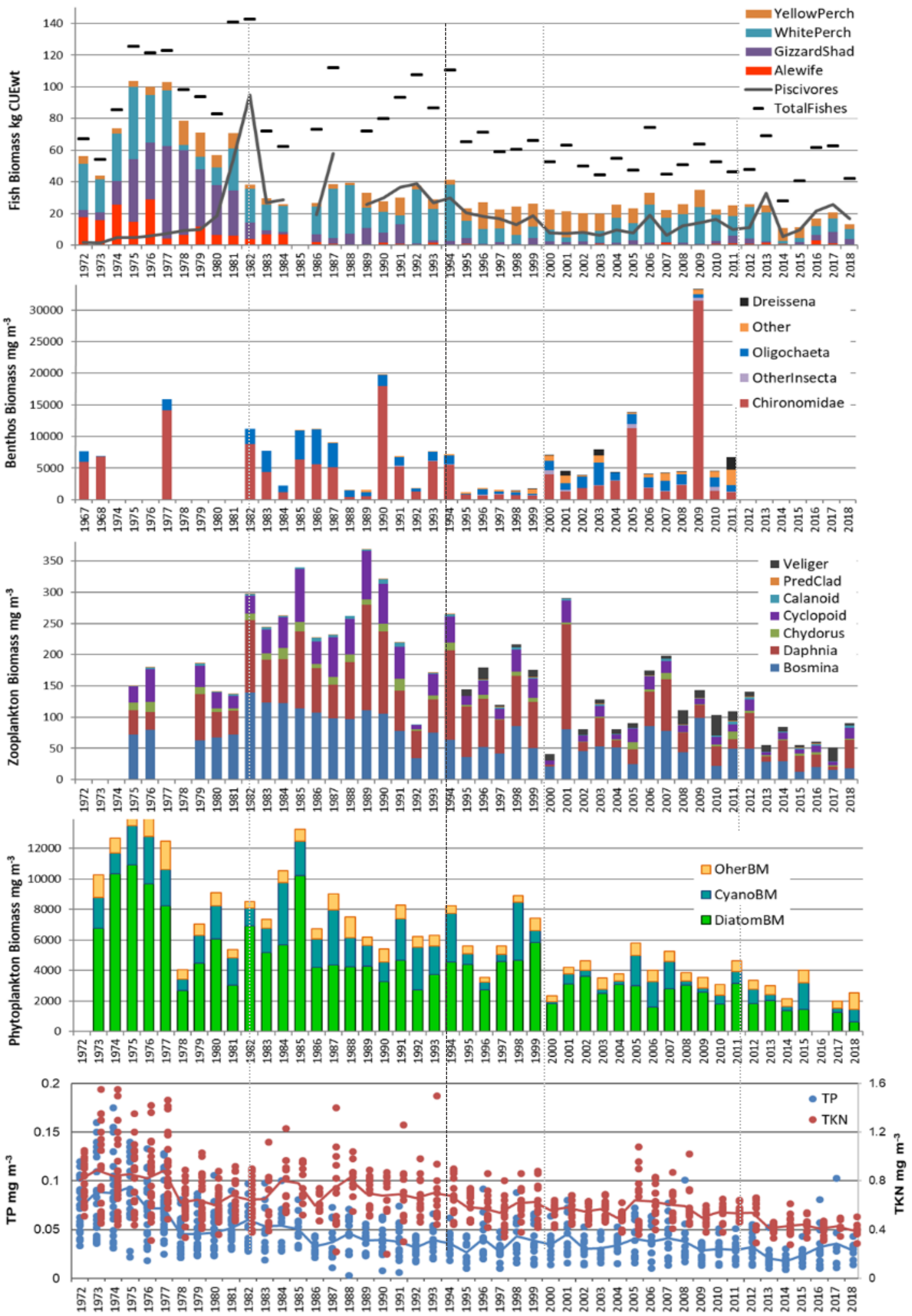


Figure 0.2 *Complete time-series at Belleville, upper Bay of Quinte for nutrients (total phosphorus and total Kjeldahl nitrogen), phytoplankton, zooplankton, benthos and fish biomasses. Time stanzas are marked by vertical dotted lines with the darked dashed line demarkating the major phases (turbid, clear). More detail will follow in S1 State of the Ecosystem.*

stanzas will be used throughout the report to track the changes to the Bay of Quinte over the sampling period for Project Quinte (Fig. 0.2).

Turbid Phase (1972 – 1994)

1) “**High P**” 1972 – 1982 was dominated by high nutrient levels and planktivorous fishes along with the transition to P-controls (high phytoplankton biomass)
2) “**Peak Walleye**” 1983 – 1994 was a stable period of climate, with intermediate nutrient levels which stabilized the system (high plankton, piscivorous fishes) and the start of submerged aquatic vegetation (SAV) regrowth

Clear Phase (1995 – 2018)

3) “**Clear Transition**” 1995 – 2000 began with a transition to a clear-phase system, with higher clarity and more rapid expansion of macrophytes, in part driven by the record cold winter of 1994
4) “**Climate Variability**” 2001 – 2012 (or 2018 for analysis) shows increased variability in precipitation and temperature and the expansion of zebra mussels in the upper bay
5) **“Productivity Drop”** (new) 2013 – 2018 is characterized by the sudden decrease in nutrients, zooplankton and fish biomass and tighter trophic linkages

A companion report (Bowen et al. 2020) being provided with this report will focus on a comparison between nearshore and offshore zones in the upper and middle Bay of Quinte. The previous assessment of BUI 13 for Bay of Quinte (Currie et al. 2017b) determined that progress had stalled in improvements to the upper Bay of Quinte based on offshore / mid-channel stations. Little improvement in the ecosystem function of the upper bay was seen since the 1980s, so a project was proposed to focus on the possibility that improvements could be found in the macrophyte-dominated nearshore zone. In 2017 and 2018 the field season included paired nearshore-offshore stations to determine differences between these zones in physical-chemical environment, plankton (microbial, phytoplankton and zooplankton) composition and effects of macrophytes on benthos (with a focus on *Dreissena*) will be found in this report. Very little effect was found between nearshore and the long-term offshore monitoring stations in the Bay of Quinte, so it was decided to provide this as a separate report rather than including it here in the asessment of BUI 13. There were several findings of interest in this nearshore-offshore comparison report. 1) While there were almost no differences in chemistry, composition or biomass, presumably due to a high degree of mixing between nearshore and offshore there were slight differences in the size of zooplankton which corresponds to potentially increased planktivory in the upper bay nearshore. There is currently little fish sampling that takes place in this zone, but high levels of Centrarchid sunfishes (Bluegill, Pumpkinseed) are suspected. 2) Submerged macrophytes facilitate dreissenid settlement since *Dreissena polymorpha* (zebra mussels) were only found preferentially on the macrophytes in the nearshore in great numbers and essentially no mussels were found in the unsuitable non-vegetated soft sediment at the offshore stations. This supports analysis which will be found in S1 on regime shifts indicating that clearing of the water conditions resulting from decreased phosphorus promoted growth of macrophytes and ultimately facilitated invasion of the Bay of Quinte by zebra mussels.

In this report, we will incorporate new analysis, updating and refining the assessment of BUI 13 for the Bay of Quinte contained in Currie et al. (2017b). As such the report will be organized into 2 major sections:

Section 1) The State of the Ecosystem. This will includes updates of the physical, chemical and plankton time series for the upper bay. Further to this, we include some preliminary analysis using General Additive Model (GAM) time-series analysis to determine if the major stations for Project Quinte have undergone what could be classified as regime shifts, or if they are better described as a continuous trend. Since there was criticism in the previous report for not incorporating the improved status of fishes or benthos we also include in this section information on fishes and benthos composition along the total phosphorus gradient. These analyses are provided to inform the assessment, but each of these analyses will also be forthcoming in greater detail as primary publications.

Section 2) Refinement of plankton metrics. In this section we will present a refinement of the weight-of-evidence metrics from the previous report (Currie et al. 2017b). This will include reduced number of metrics of impairment for i) phytoplankton, ii) microbial plankton, iii) zooplankton, and iv) trophic ratios. This analysis will focus on selection of a reduced set of metrics and application of the metrics to the last two decades from 2001 – 2018 most appropriate for developing a future monitoring plan. A summary and overall updated assessment for BUI 13 in the Bay of Quinte AOC and recommendations for a monitoring plan is provided.

S1. State of the Ecosystem

To put the assessment of the Bay of Quinte in perspective, it is important to review the changes to the primary drivers and ecosystem measures collected for Project Quinte from the period 1972 – 2018. This includes major environmental drivers, nutrients, and biomass measures of the system. May to October mean surface water temperatures which are consistantly warmer at the shallower stations (Fig. 1.1); mean max May – Aug air temperatures which show hot summers in 1975, 1991, 1995, 1999, 2005, 2010, 2012 and 2016; mean min Jan – Feb air temperature with especially cold winters in 1982, 2003, 2015; and mean annual air temperature. Annual preciptiation showed more variability year to year than average temperature. Winter snow was also a highly variable parameter with peaks in 1978, 1997 and 2008.

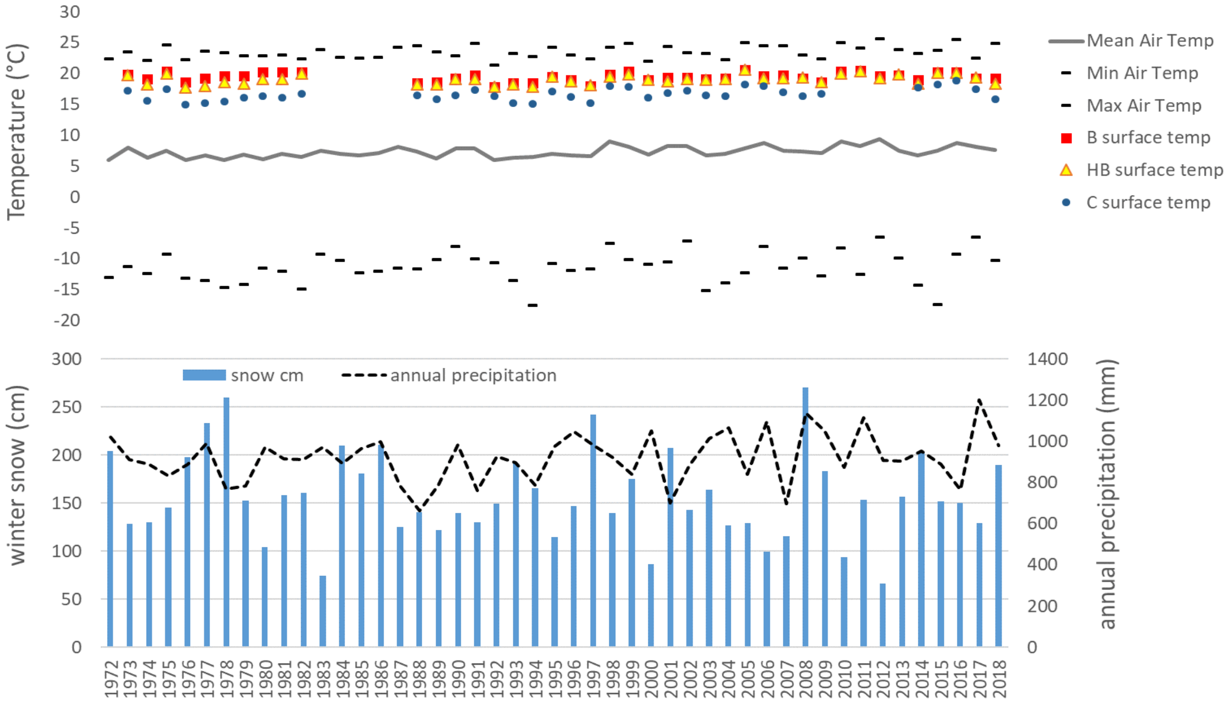


Figure 1.1 *May to October mean surface water temperature, mean max May-Aug, mean min Jan-Feb and mean annual air temperatures (°C, top panel). Annual precipitation (mm) and winter snow (cm, bottom panel). Weather parameter data from Trenton, ON (Environment and Climate Change Canada).*

Although primarily associated with BUI 8, measures of total phosphorus (Fig. 1.2) and total Kjeldahl nitrogen (Fig. 1.3) are important for assessing BUI 13 as nutrients are a primary driver of phytoplankton growth. Measures taken during the May to October period show a large range of variabilty within a year as is expected given the seasonality seen in these measures. The highest variability tends to be earliest in the time series prior to phosphorus controls, particularly sewage treatment improvements but stabilized since and has shown a steady linear trend downwards since the early 1980s in the upper bay. While the range in measured values within a year has generally decreased for TP in the upper bay there are some recent years with very high measures. This is consistant with the finding that TP loadings into the Bay of Quinte in the Trent, Salmon, and Napanee Rivers have been increasing since 2003 (Kim et al. 2013) and the increase in P released from the sediments in the upper bay since 2000 (Doan et al. 2018). The range and maximum values have increased in the lower bay in recent years to values higher than those seen in the 1970s.

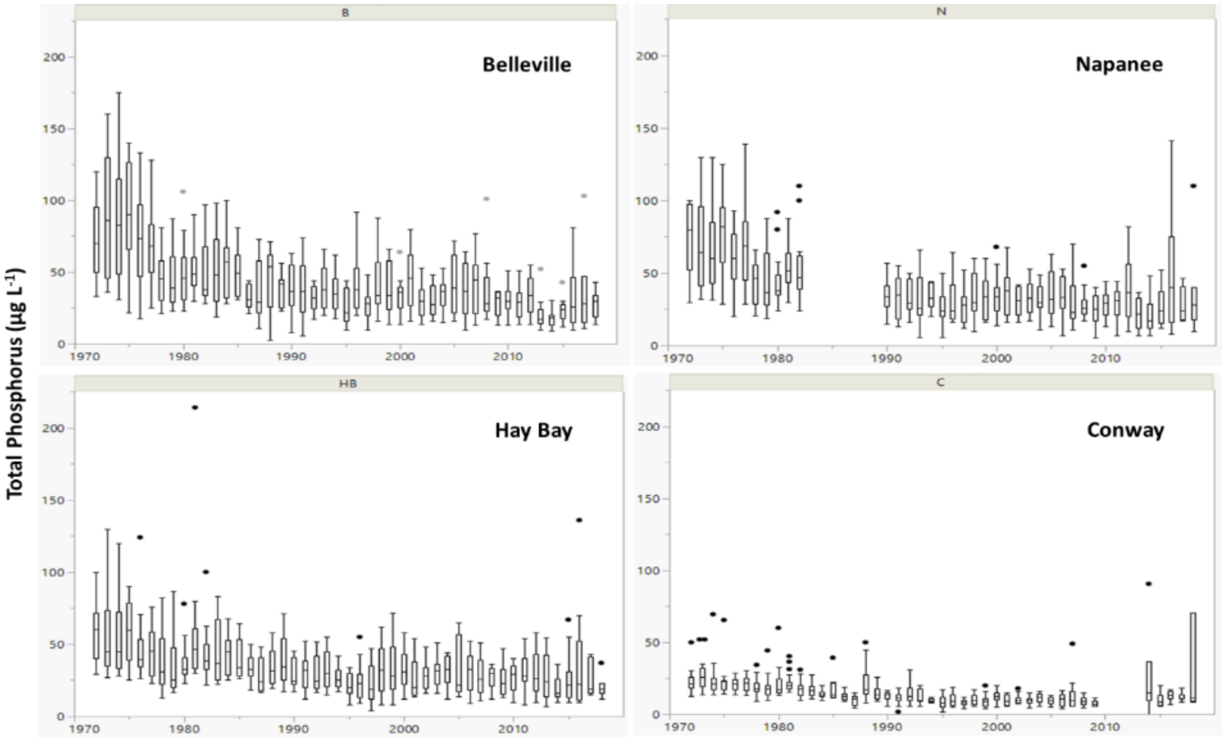


Figure 1.2 *Long term changes in total phosphorus (µg L^{-1}) at the main Bay of Quinte stations, May to October, 1972 to 2018. Points represent outliers. Boxplots are median values with 25th and 75th percentiles.*

Over the time series TKN shows a greater reduction in both range and annual mean values (Fig. 1.3) compared to TP and as with TP the reduction is most noticable when comparing pre STP years to the most recent in the upper bay. TKN is composed of the organic nitrogen forms plus ammonium and is commonly used for studies with wastewater. Because this measure is unfiltered it includes nitrogen associated with plankton and reflects changes in the biomass over this time period. Looking at nitrate+nitrite measures (Fig. 1.4) which are representative of the more immediately bioavailable nitrogen it is likely that nitrogen limitation was common in the 1970s in the upper bay when seasonal mean values were in the range of 10 – 20 µg L^{-1}, well below the limiting level of 100 to 130 µg L^{-1} dissolved nitrogen (Currie and Frank 2015, Chorus and Spijkerman 2020). Although mean values increased in the 80s and 90s the means are still below 100 and by the 2010s are below 50. The average values at HB are in the non-limiting range through the 80s and 90s but are again below limiting levels more than half the time since 1998. Figure 1.3 does not include ammonia which is generally found in much lower amounts than nitrate because it is much more easily taken up by phytoplankton. These values are 2 orders of magnitude lower than offshore Lake Ontario and 3 orders of magnitude lower than in the heavily sewage-impacted Hamilton Harbour AOC. Although most measures are not under the limiting threshold at C, it is interesting to see the increasing trend which is abruptly reversed in the recent years when TP values also show a large change and may indicate N is occasionally limiting in the lower bay now.

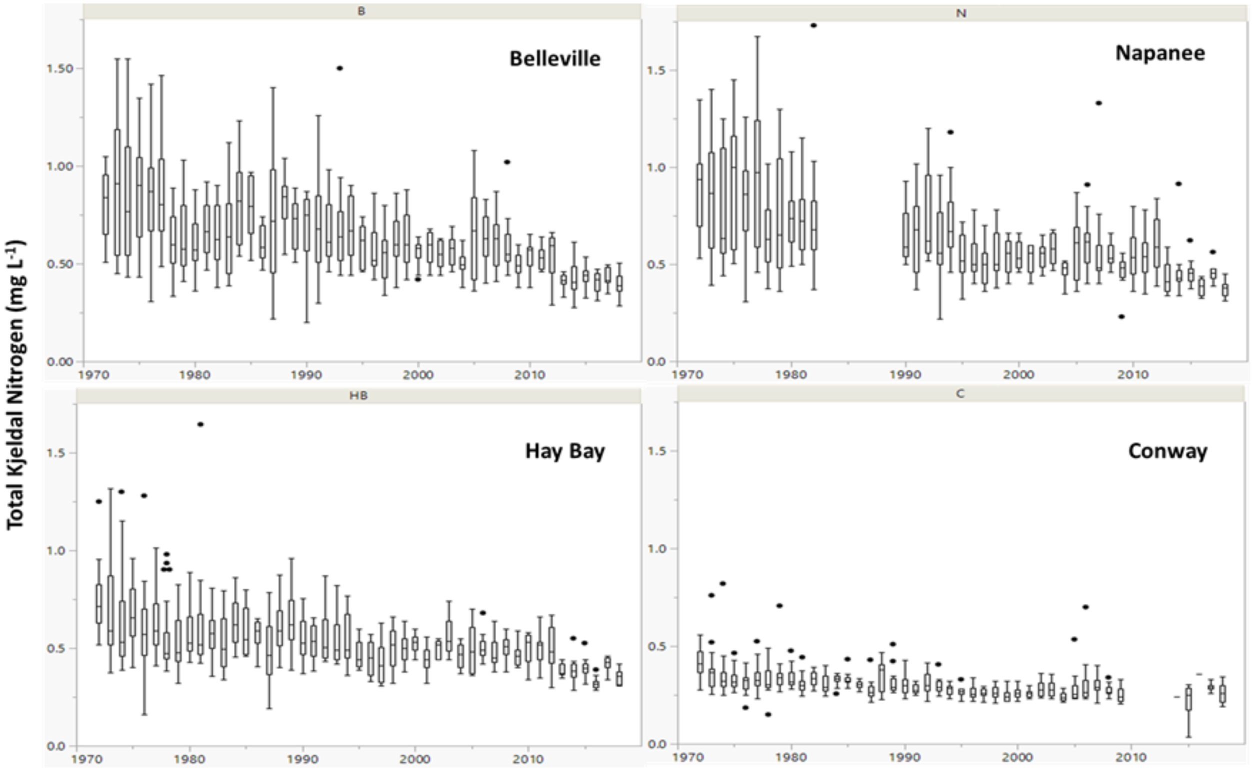


Figure 1.3 *Long term changes in total Kjeldahl nitrogen (mg L^{-1}) at the main Bay of Quinte stations 1972 to 2018. Points represent outliers. Boxplots are median values with 25th and 75th percentiles.*

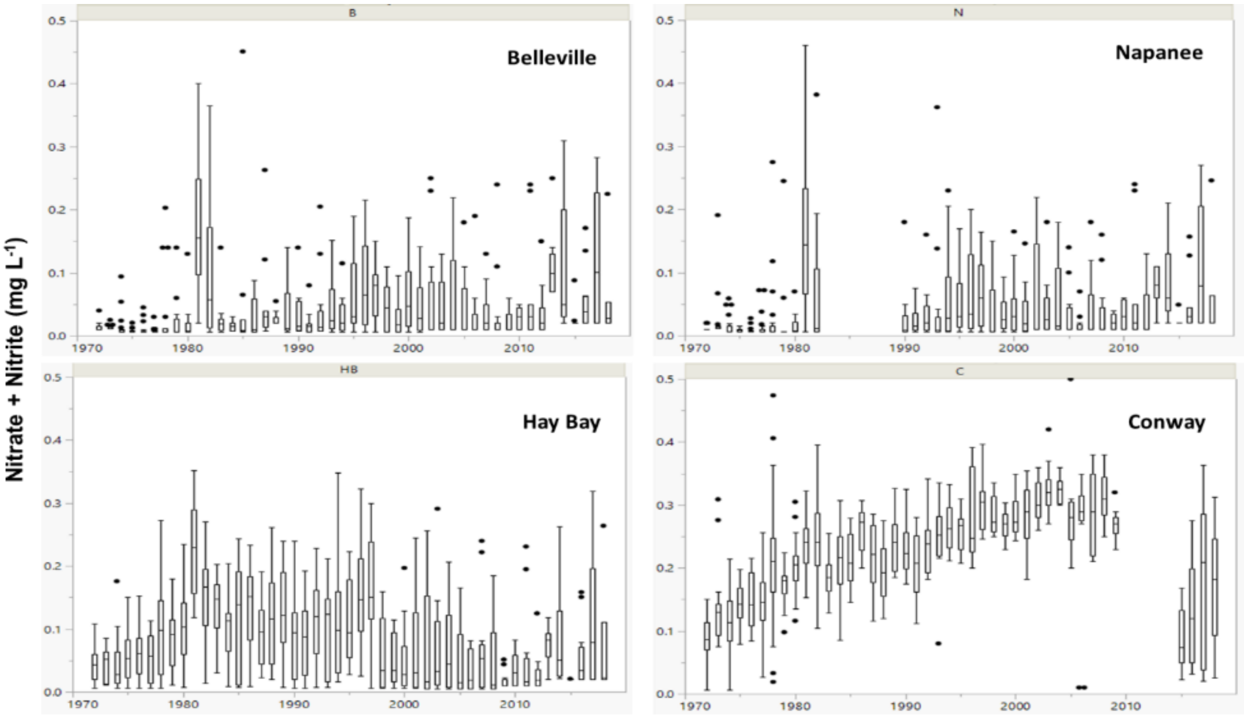


Figure 1.4 *Long term changes in nitrate+nitrite (mg L^{-1}) at the main Bay of Quinte stations, May to October, 1972 to 2018. Points represent outliers. Boxplots are median values with 25th and 75th percentiles. Scale is truncated so that extreme values over 0.50 are excluded.*

Light attenuation is a measure of water clarity and shows decreased values (increased clarity) moving toward the middle of the time series and appearing to increase (decreased clarity) or level off since 2000 (Fig. 1.5). Clarity is affected by both autotrophic and heterotrophic planktonic biomass and by suspended sediments and so the trends are not simply a direct reflection of phytoplankton biomass which has generally shown a decrease over the time series (Fig. 1.6 and 1.7).

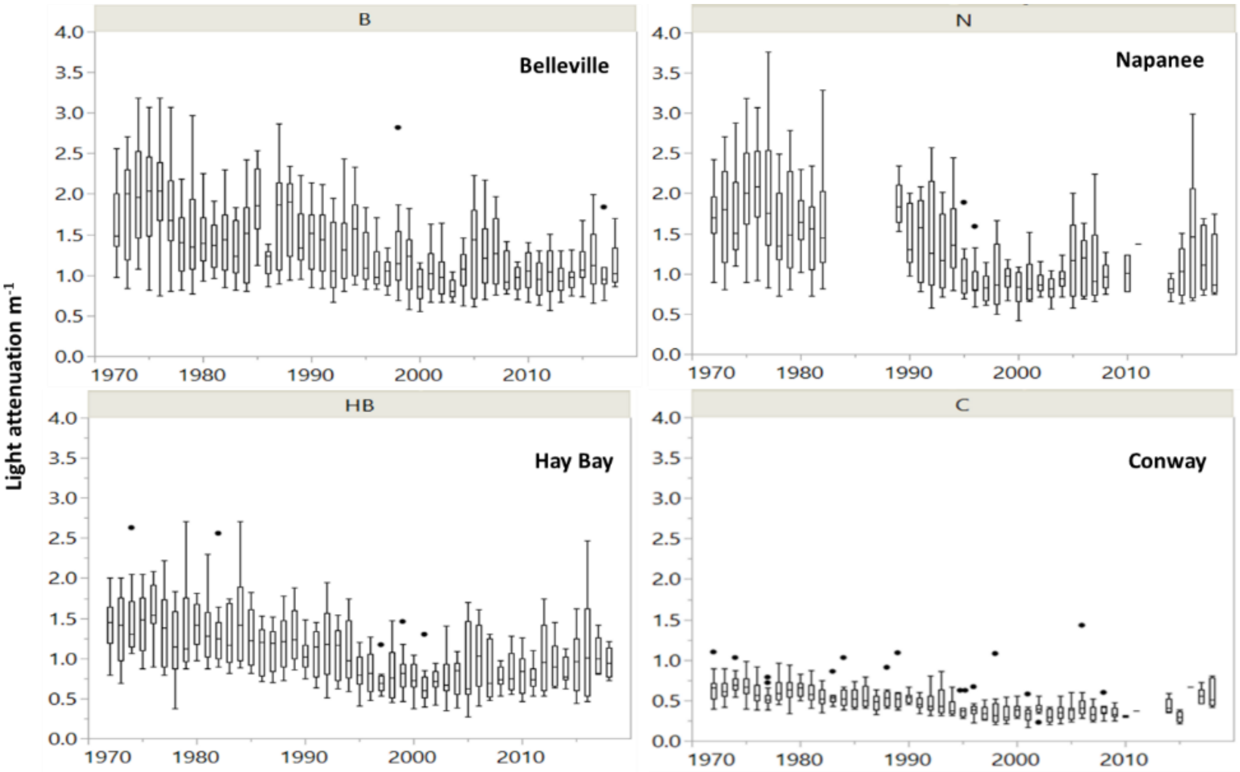


Figure 1.5 *Long term changes in light attenuation (m^{-1}) at the main Bay of Quinte stations May-October, 1972 to 2018. Points represent outliers. Boxplots are median values with 25th and 75th percentiles.*

At all stations phytoplankton are mainly Diatoms followed by Cyanobacteria. They are primarily composed of filamentous forms with the diatom *Aulacoseira* (previously *Melosira*) being the top taxa overall and contributing almost 50% of the biomass over the entire time series (Currie et al. 2017b). *Dolichospermum* (formerly *Anabaena*) is a filamentous Cyanobacteria capable of producing toxins that has contributed 1 to 25% of the annual mean but is less dominant after the mid-1990s (Currie et al. 2017b). Filamentous algal forms are common in shallow eutrophic environments because they are constantly mixed back up into the euphotic zone by resuspension despite their tendancy to sink (Scheffer et al. 1997). Filamentous forms are more difficult for filter feeding zooplankton to handle so they are generally not preferentially grazed within the foodweb. The remaining portion of phytoplankton is composed of several taxonomic groups (Chryptophyceae, Euglenophyceae, Chrysophyceae, Chlorophyceae and Dinophyceae) and together contribute less than 20% to total biomass in most years in the upper and middle bay. The lower bay has consistenaly had 30 – 40% of the biomass comprised of other taxonomic groups and is indicative of a healthier phytoplankton population. These groups tend to be smaller and preferentially ‘edible’ and may be consumed more easily by zooplankton than the filamentous forms that dominate the biomass in the upper bay.

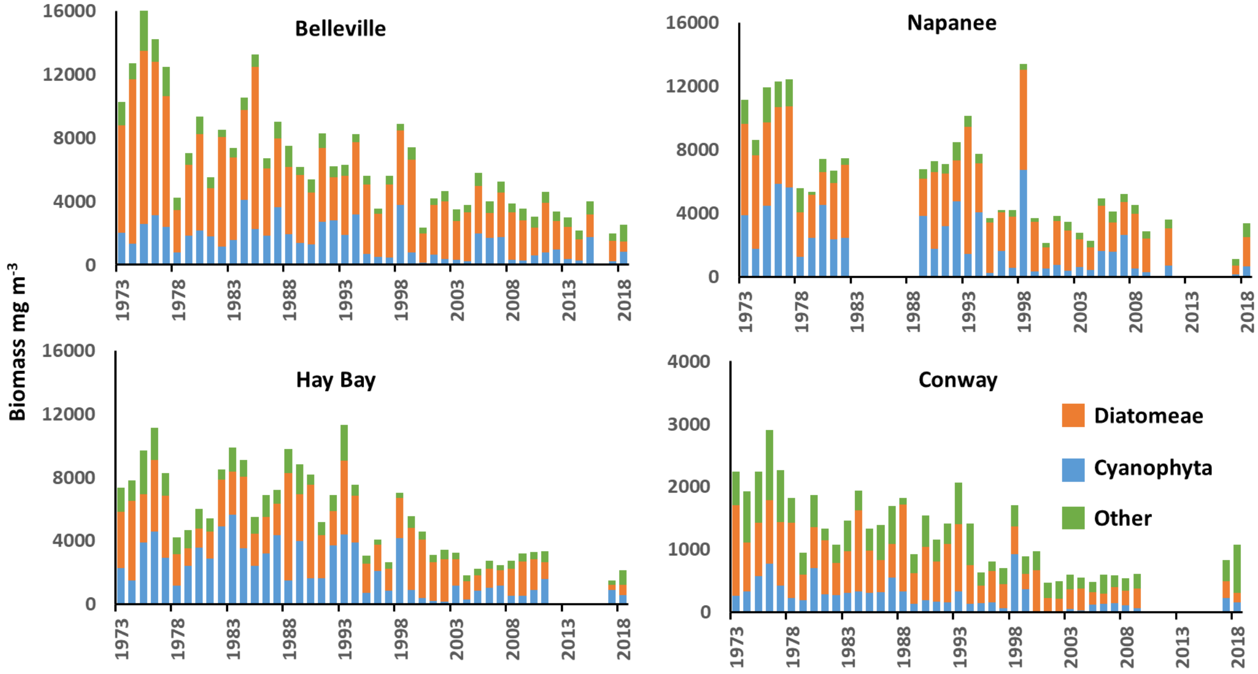


Figure 1.6 *Long term changes in phytoplankton wet-weight biomass (mg m*$^{-3}$*) at the main Bay of Quinte stations, May to October average, 1973 to 2018. Data are averages of individually counted phytoplankton samples or seasonal composite samples.*

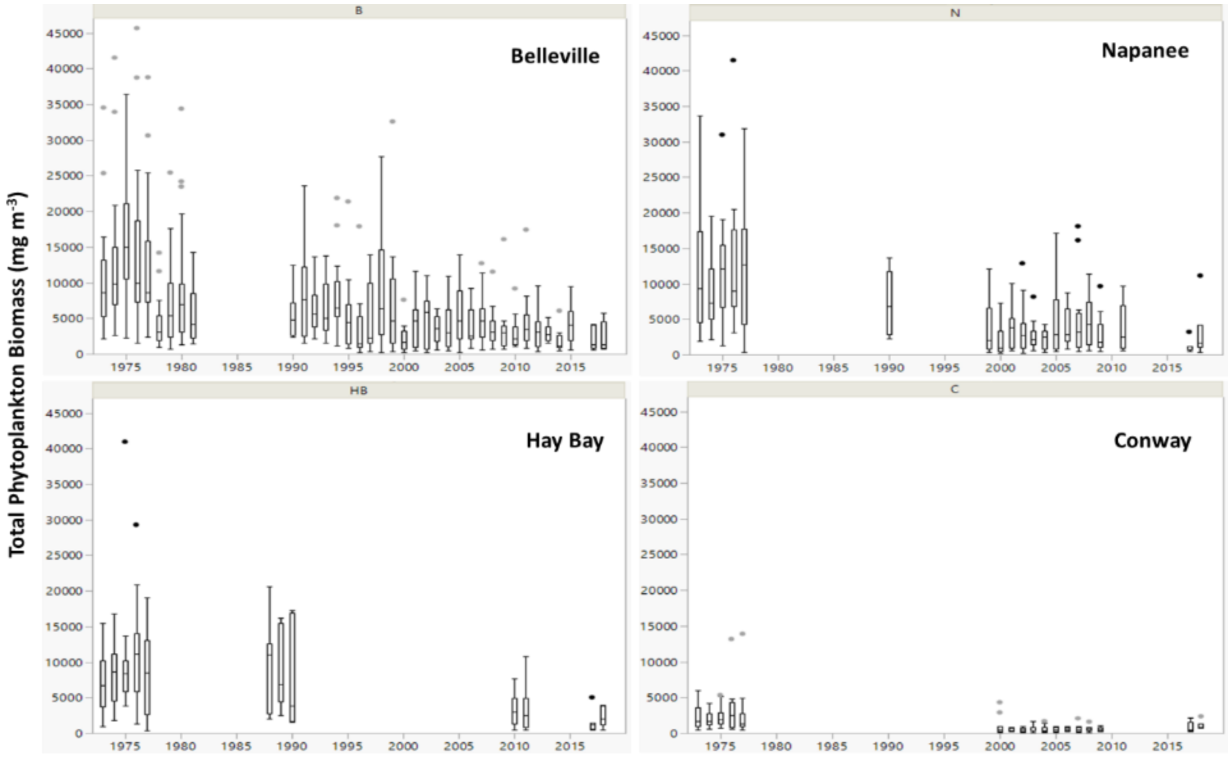


Figure 1.7 *Total phytoplankton wet-weight biomass (mg m*$^{-3}$*) individual date measures where May-October measures are available, 1973 to 2018. Points represent outliers. Boxplots are median values with 25th and 75th percentiles.*

The eutrophic Hamilton Harbour AOC has similar nutrient loadings, but in contrast to the Bay of Quinte has annual averages of 45 – 78% of the phytoplankton biomass made up of the other taxonomic groups. As with the nutrient parameters, there is a decreased range of biomass in later years, but the Bay of Quinte still has the greatest production of biomass per unit Total Phosphorus than any other site measured by DFO except for Lake Erie (see Section 2.4: Trophic Ratios). Also, in recent years there are some very high biomass values recorded in the Bay of Quinte indicating reversals of previous improvements.

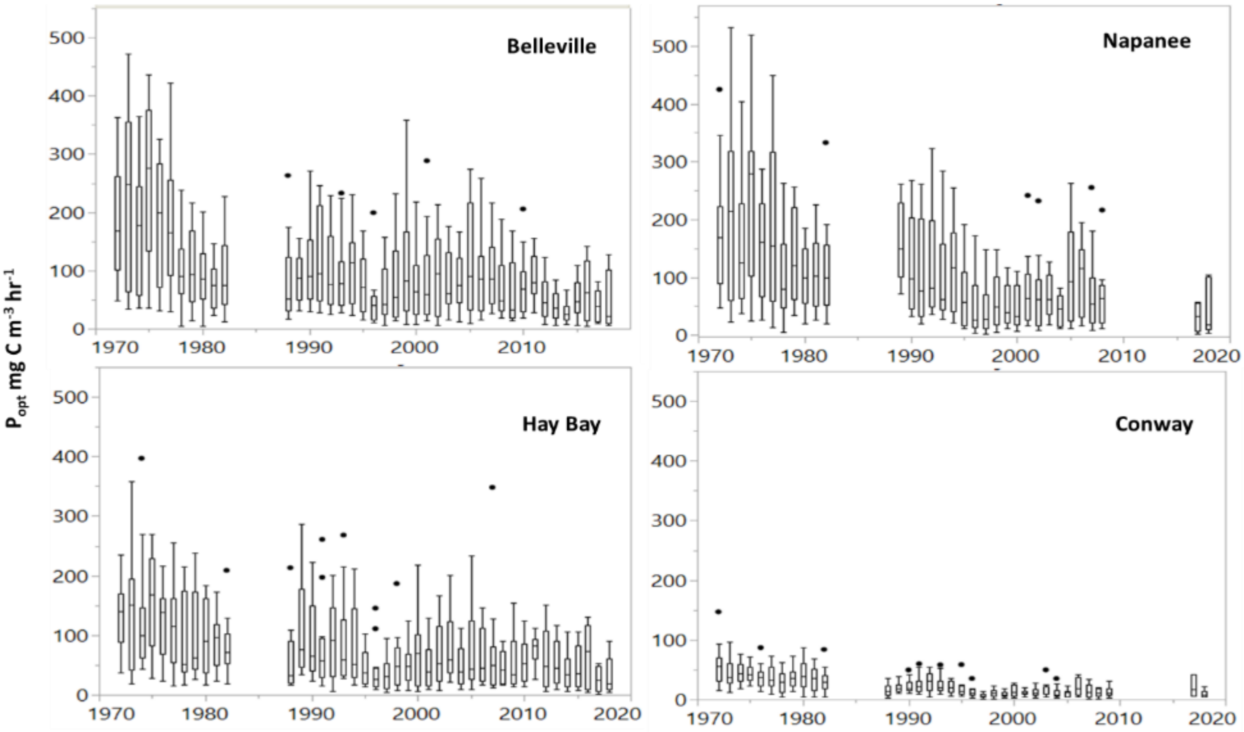


Figure 1.8 *Long term changes in phytoplankton productivity at optimal light levels (P_{opt} mg C m^{-3} h^{-1}) at the main Bay of Quinte stations, May to October, 1972 to 2018. Data points represent individual dates. Boxplots represent median values with 25th and 75th percentiles.*

Primary productivity experiments using ^{14}C uptake have been conducted for the duration of the time series (1972 – 1982, Millard and Johnson 1986; 1987 – 2010, Fee et al. 1989, 2000 – 2018; Munawar and Munawar 1986). Although techniques have differed over the years, overlap between 2000 – 2010 indicate that the instantaneous primary production rates at optimal light levels measured are comparable. Primary productivity was extremely high in the upper bay in the early 1970s (Fig. 1.8) and still continues to reach high levels. Current levels are considered to be in the hypereutrophic range and comparable to values observed in Hamilton Harbour and western Lake Erie. These experiments are undertaken in the lab in a consistent environment so biotic and abiotic factors such as sediment resuspension, light climate, temperature and grazing do not affect these rates.

Zooplankton biomass in the Bay of Quinte is dominated by 2 groups of herbivorous cladocerans (primarily bosminids and *Daphnia*) and omnivorous cyclopoids (Fig. 1.9). Other zooplankton taxa (including *Dreissena* veliger larvae) are grouped together and contribute less than 10% to total biomass. Populations of *Daphnia*, an efficient, relatively nonselective filter feeder, have generally declined at Belleville in the 2000s after initial increases to peak levels in the mid-1980s and 1990s. The zooplankton biomass during the last 5 years of sampling includes 4 of the lowest 5 values of the Project Quinte data series. Cyclopoid biomass has also declined dramatically over this time period. Bosminids, comprised of the ubiquitous, tolerant *Bosmina* and the slightly larger *Eubosmina*, have remained relatively stable since the early 1990s. Although they typically feed on small algal cells, *Eubosmina* and other zooplankton genera such as *Chydorus*, and herbivorous copepods can grasp and manipulate large algal forms (Irvine, 1986), so may be linked to the dominance of filamentous diatoms in the Bay of Quinte.

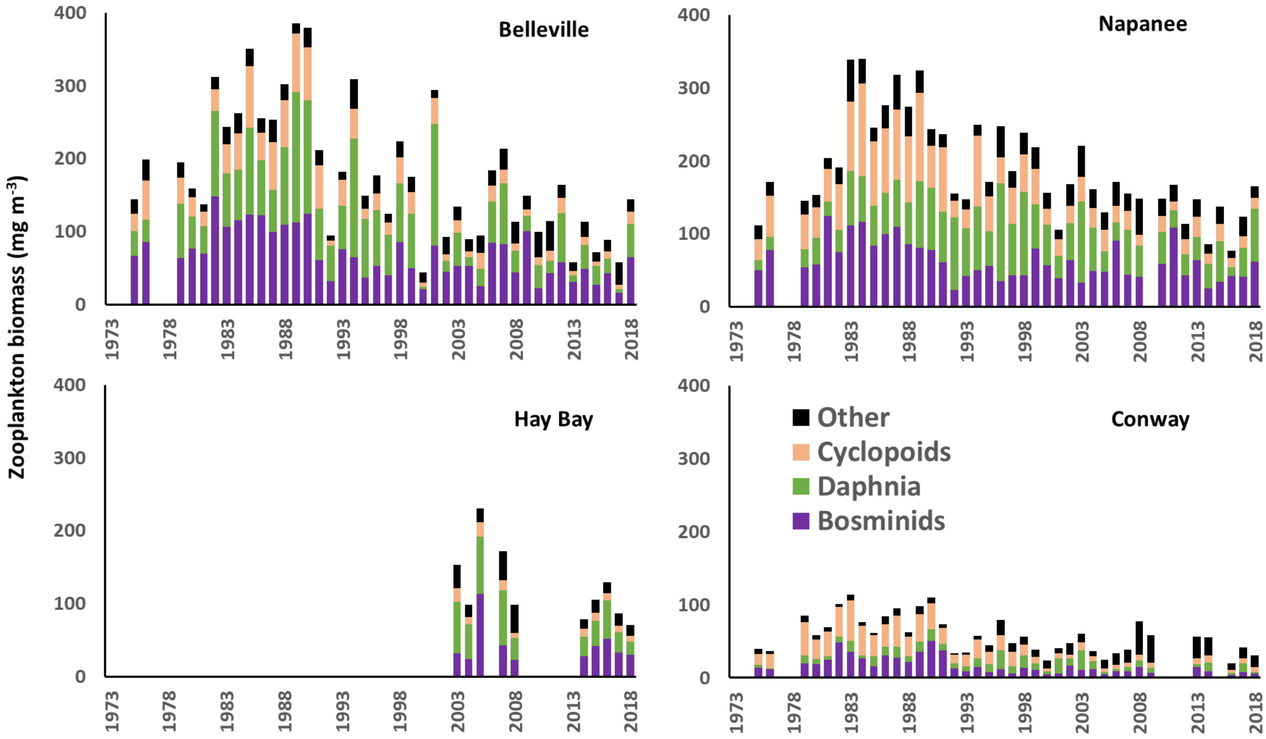


Figure 1.9 *Long term changes in zooplankton dry-weight biomass (mg m^{-3}) at the main Bay of Quinte stations, May to October, 1972 to 2018.*

Fish population catch per-unit effort (CPUE) data from the Ministry of Natural Resources and Forestry gill net and trawling surveys has shown decreased fish catches in the upper bay. Reductions in planktivorous fishes are due mainly to declines in the pelagic feeding planktivore Alewife and herbivorous Gizzard Shad in the early 1980s (Fig. 1.10). The diet switching Yellow Perch and piscivores have increased CPUE since 1990. These patterns hold in Hay Bay and Conway except that the CPUE values are much smaller than in the upper bay and Piscivore biomass has decreased since the mid-1990s after a temporary increase in the mid-1980s to early 1990s. White and yellow perch currently comprise the majority of CPUE of possible planktivores indicating there continues to be strong predation pressure on the benthic and pelagic invertebrates, but the larger size classes of Yellow Perch are piscivorous. Gizzard shad which feed directly on phytoplankton have experienced stronger CPUEs in recent years (11, 17) which could contribute to decreases in phytoplankton biomass.

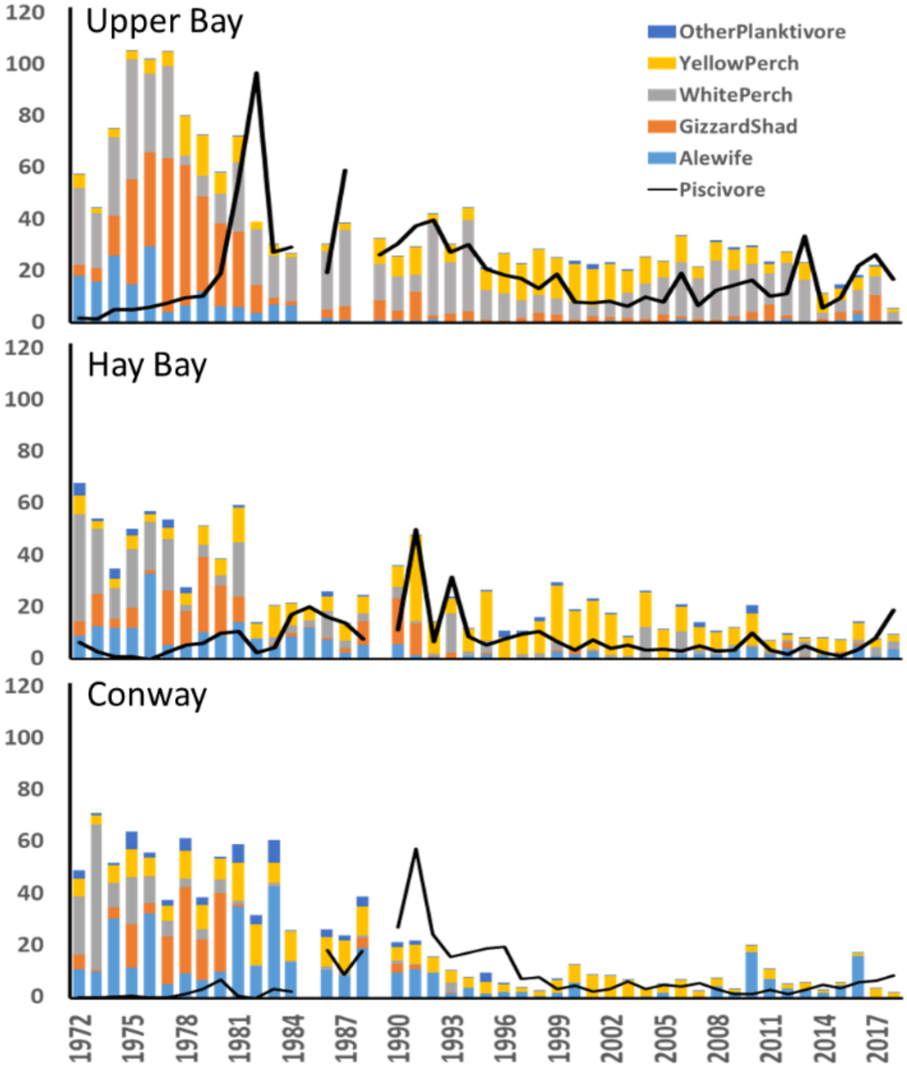


Figure 1.10 *Long term trends in planktivorous and piscivore fish biomass CPUEwt (Combined data from trawl and gillnet). Data courtesy of Ontario Ministry of Natural Resources and Forestry*

The eutrophic state of the Bay of Quinte is seen when examining the relative amount of biomass per unit of TP, which shows a strong desparity between trophic levels. This is particularly evident in the relative increase of phytoplankton biomass per unit TP compared to the increase of zooplankton biomass per unit TP. Simply put, at high TP concentrations there is a greater difference in the biomass of phytoplankton to zooplankton than occurs at low TP (Fig. 1.11). The relative slope of the phyto:TP relationship is drastically larger than the zoop:TP relationship, whereas the planktivorous fishes biomass per unit TP is only slightly higher than the zoop:TP relationship. The difference in slope between phytoplankton biomass and zooplankton biomass relative to total phosphorus is driven by excessive production of phytoplankton at high nutrient levels (eutrophication) which is not being consumed by zooplankton and therefore not brought into the foodweb as transferable energy. Some of that excess phytoplankton production is likely cycled into microbial food web when it settles as detritus. The absolute value and slope of planktivorous fishes biomass relative to total phosophorus is very similar to that of zooplankton, which is also seen in the time-series plots as they track together (Fig 1.11, see S2.4). This indicates biomass being stored in the higher trophic levels, as explained by inverted trophic pyramids, which can occur in eutrophic systems (McCauley et al. 2018). This also explains the recent drop in both fish and zooplankton biomass during the last 5 years of the time-series (Fig. 0.2). The slope of the total fishes biomass (not shown on graph) is slightly greater than the zooplankton and planktivorous fishes biomass and the higher intercept places the absolute biomass value above these groups. The total fishes relationship does not include the Conway station because the biomass of fishes at this station is dominated by species from Lake Ontario.The larger slope and intercept is due to the fact that not all fish species are dependant on the local planktonic food web and there are also likely external inputs of energy and migration from surrounding habitats (Lake Ontario, tributaries etc.).

Since the biomass values are much closer together at low total phosphorus levels found more recently in the Bay of Quinte, there is an expectation of tighter coupling between total biomass and phosphorus levels within the Bay. As such there should be even closer relationships beween plankton and fish biomass in the future.

$$\text{PhytoBM (mg m}^{-3}\text{) = -947 + 178300 • TP (mg L}^{-1}\text{), } r^2 = 0.70$$

$$\text{ZoopBM (mg m}^{-3}\text{) = 395 + 36200 • TP (mg L}^{-1}\text{), } r^2 = 0.35$$

$$\text{PlanktivoreBM (mg m}^{-3}\text{) = 183 + 40900 • TP (mg L}^{-1}\text{), } r^2 = 0.40$$

$$\text{TotalFishBM (mg m}^{-3}\text{) = 928 + 54000 • TP (mg L}^{-1}\text{), } r^2 = 0.29$$

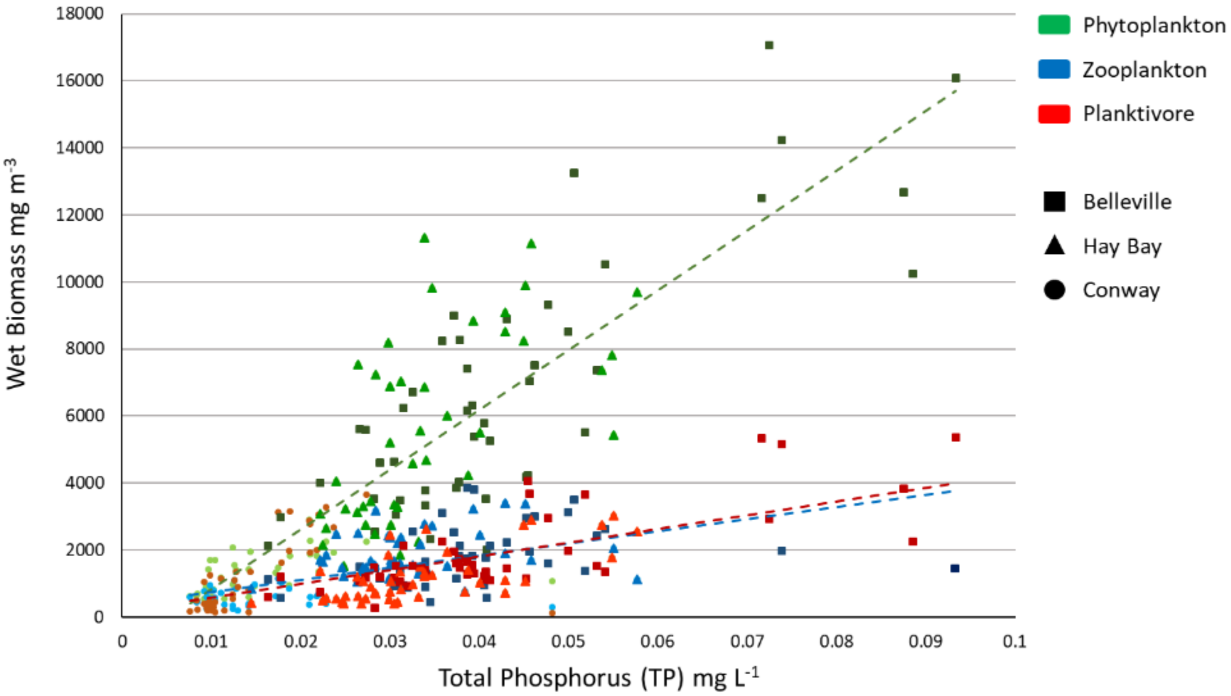


Figure 1.11 *Wet-weight biomass (mg m^{-3}) for each trophic level (phytoplankton, zooplankton, planktivorous fishes) in the Bay of Quinte relationship with total phosphorus (TP). This graph includes Belleville, Hay Bay and Conway data for each group. Dry weight zooplankton is converted to wet weight assuming 10% factor and MNRF trawl and gillnet weights are corrected to the same units by comparing weights through regression with electrofishing biomass units from DFO surveys (MNRF CPUEwt = eFish/51.621, r^2=0.79, see Section 2.4: Trophic Ratios).*

S1.1 Analysis of Fish and Benthos Community Composition

For this analysis of fish community change in the Bay of Quinte, weight Catch per Unit Effort (CPUE) from MNRF trawls and gillnets which were set annually June 21 – August 31 were combined using a modified multigear mean standardization procedure (Gibson-Reinemer et al. 2017; Ward et al. unpublished). Groupings of invertivores, planktivores, piscivores and prey-switchers were determined in consultation. Details will be found in Ward et al. (forthcoming).

Total fish biomass declined significantly with reductions in total phosphorus (Fig. 1.12a), though the number of years of data available at extreme phosphorus levels (high and low) was limited, and there was very high variablity in the dominant range of 30-50 µg L^{-1}. At uppermost levels of total phosphorus the fish community was dominated by Gizzard Shad (*Dorosoma cepedianum*), which represented >50% of total community biomass in some years (Figure 1.12b). During this time piscivore biomass was low and comprised ≤ 2% of total community biomass. With declining nutrient loading, Gizzard Shad biomass and dominance declined and the community became dominated by a more diverse mix of fish that consume at higher trophic levels (piscivores, switchers, and invertivores) than Shad. Notable is the near-absence of piscivores at uppermost phosphorus levels and their resurgence and increasing contribution to total fish community biomass as phosphorus declined. The presence and increasing dominance of top predators is an indicator of improving ecosystem health (Myers and Worm 2005, Hoyle et al. 2012).

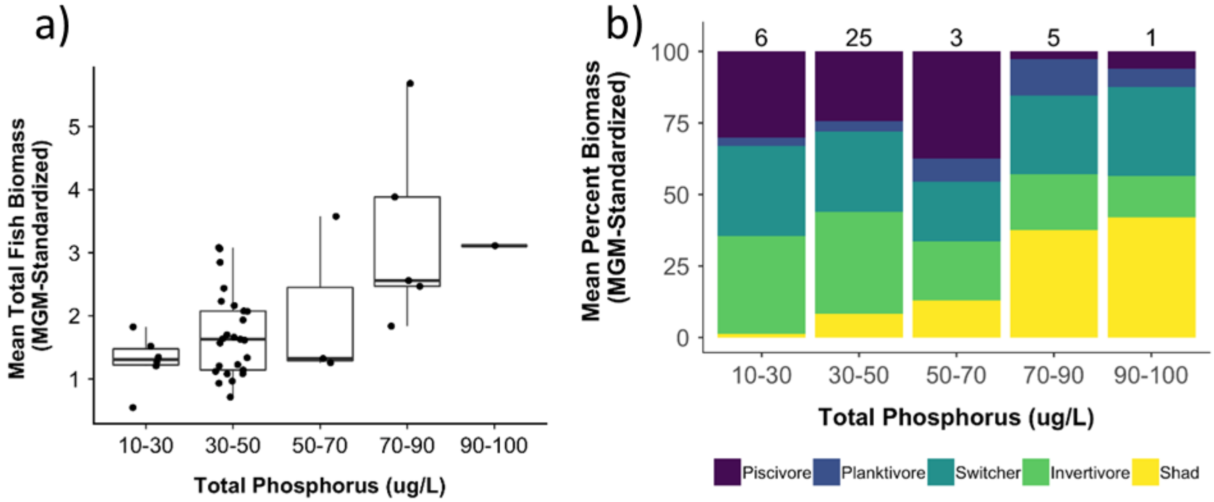


Figure 1.12 *a) Log total fish biomass in the Upper Bay of Quinte, 1972-2014. Data points represent annual means. Boxplots represent median values with 25th and 75th percentiles. b) Fish community composition in the Upper Bay of Quinte 1972-2014. Data represent means of annual data for 20 µg L^{-1} bins of total phosphorus. Numbers at top of bars is count of years for each bin.*

Community dominance of species feeding at low trophic levels is often associated with ecosystem impairment (Moore et al. 2004), especially under eutrophic conditions (Vanni et al. 2005). To assess whether this phenomenon transpired in the Upper Bay of Quinte we modeled the ratio of two dominant fish species across the gradient of total phosphorus (Fig. 1.13). Gizzard shad are generalist consumers of the lowest trophic levels in aquatic food webs (phytoplankton, particulate detritus; Vanni et al. 2005) and comprised >50% of total community biomass in some years with greatest nutrient loading (Fig. 1.12b). Conversely, Yellow Perch (*Perca flavescens*) are known to consume prey at intermediate trophic levels in the Bay of Quinte and other locations (benthic invertebrates, zooplankton, small fish; Hurley 1986, Bowman 2005). Yellow Perch averaged 12% of biomass, peaking in the late 1990s to early 2000s at 25%, and during the last time stanza represented up to 15% of fish community biomass as nutrient loading declined in the Upper Bay. As such, the ratio of Yellow

Perch:Gizzard Shad provides an indication of the trophic level at which the dominant fraction of fish biomass in the Upper Bay of Quinte is feeding, and is thus can be used as a fish-community metric of ecosystem impairment for the Upper Bay of Quinte.

A sigmoidal model was fit to the data using GLM (quasibinomial family, Fig. 1.13); both intercept and slope of the resulting model were highly significant ($p = 10^{-7}$):

log(YPerch +0.0001 / GizzShad + 0.0001) = 9.56 – 2.67*log(TP)

A linear model fit to the range of total phosphorus (26 – 55 ug TP/L) over which the ratio was linear was also highly significant ($p = 0.002$, adjusted $R^2 = 0.26$, $F(30) = 11.82$:

log(YPerch +0.0001 / GizzShad + 0.0001) = 2.51 -0.56*log(TP)

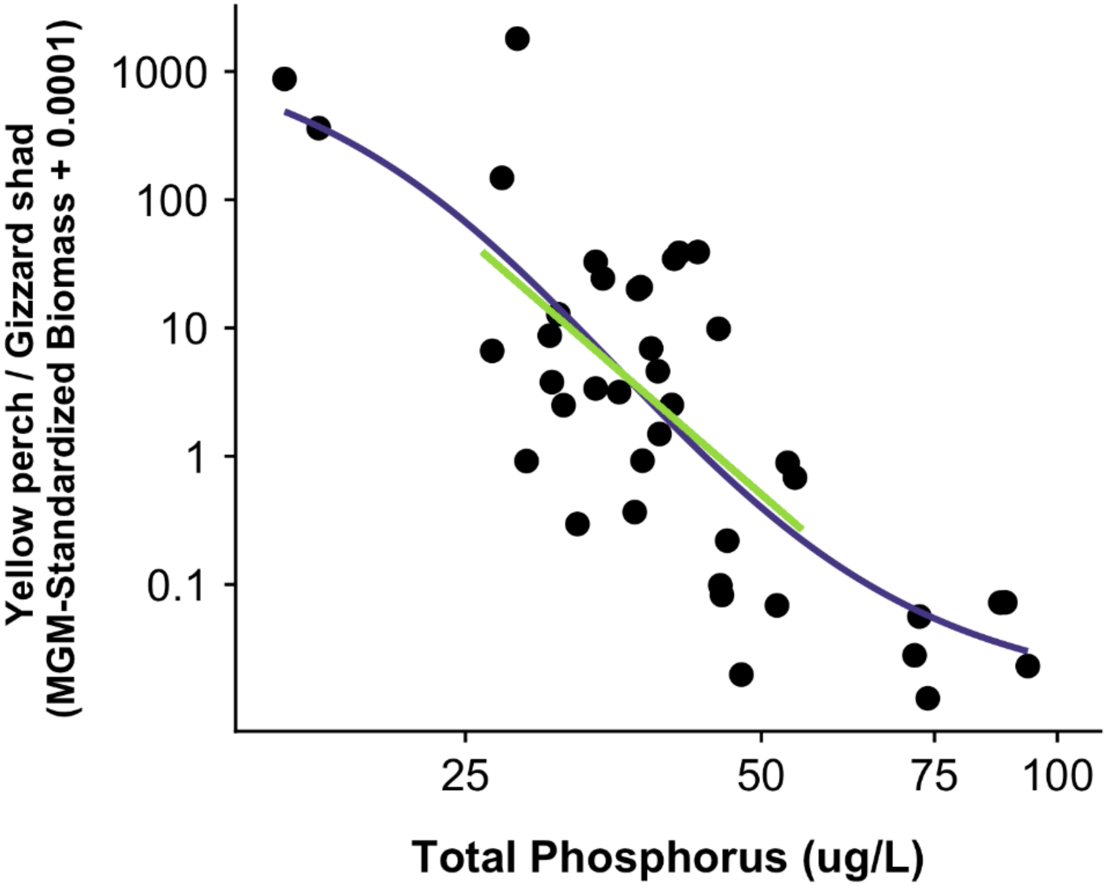


Figure 1.13 *The ratio of Yellow Perch:Gizzard Shad biomass (MGM-standardized) in the Upper Bay of Quinte, 1972-2014. Data points are annual values. Purple line is a sigmoidal model fit; green line is a linear model fit to a restricted range of total phosphorus (26 – 55 µg L^{-1} total phosphorus).*

To further reinforce the importance of the role of Yellow Perch in the food web of the Bay of Quinte, trophic position of Yellow Perch was calculated from stable $^{15}N/^{14}N$ isotope data (fish scales with seston baseline) in Bowman (2005). These estimates were plotted against total phosphorus grouping seen above (Fig. 1.14). Estimates of Yellow Perch trophic position provide further support for a general shift from feeding at lower (planktivorous, benthivorous) to higher trophic level (continued benthivory with increased piscivory) with reduction of nutrient loading.

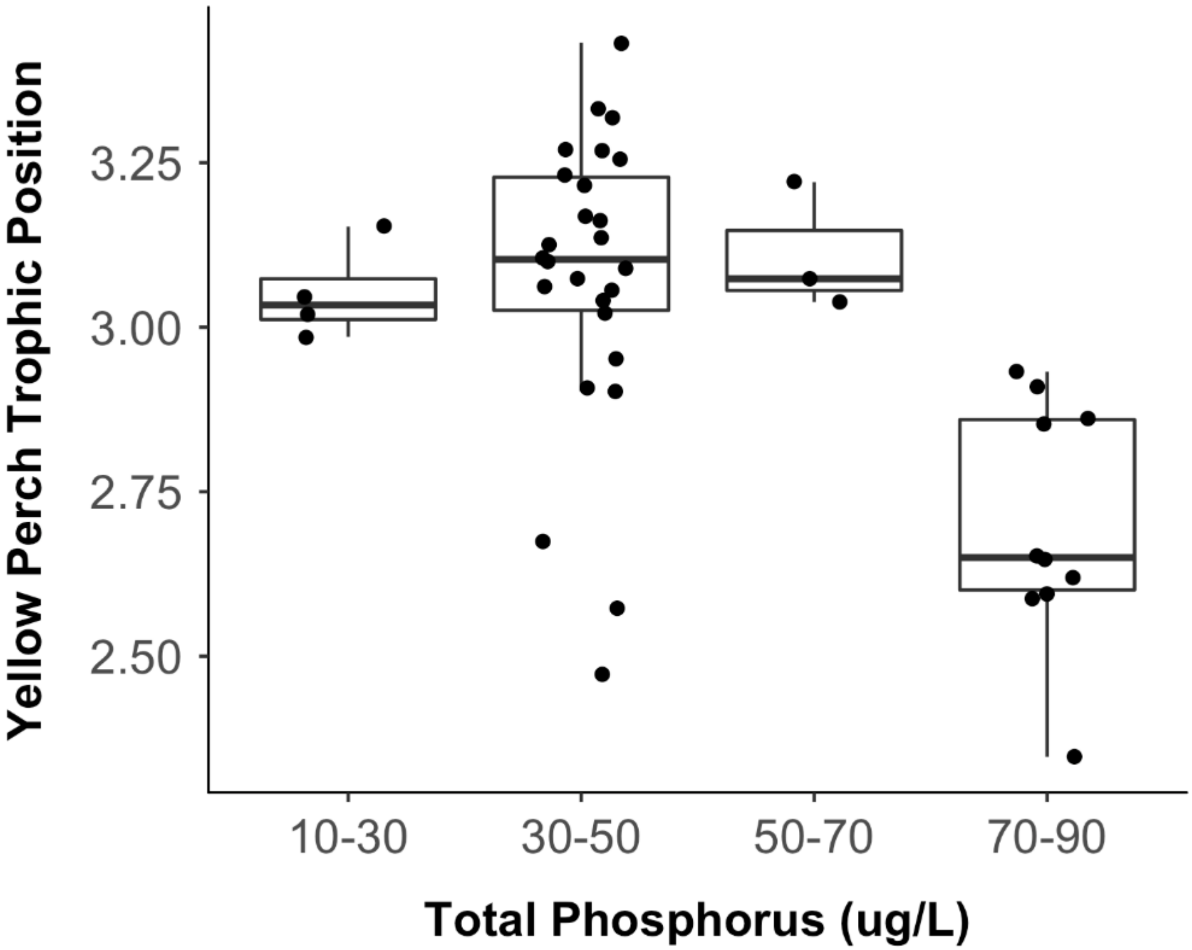


Figure 1.14 *Trophic position of yellow perch plotted against 20 µg L^{-1} bins of total phosphorus. Boxplots represent median values with 25th and 75th percentiles. Data points represent individual fish collected 1974-2003.*

Although variable, the total biomass of non-dreissenid benthic invertebrates did not vary significantly with total phosphorus (Fig. 1.15a). This is most likely due to the situation that total benthic invertebrate biomass is fundamentally top-down controlled in the Upper Bay of Quinte (Ward et al. unpublished). However, there is considerable change in the composition of the benthos across the phosphorus gradient. The benthic community was dominated by chironomids and oligochaetes at uppermost levels of total phosphorus and became more diverse with declining nutrient loading (Fig. 1.15b). Although sample sizes are limited at uppermost and lowermost levels of nutrient loading, data support expectations for diversity derived from literature.

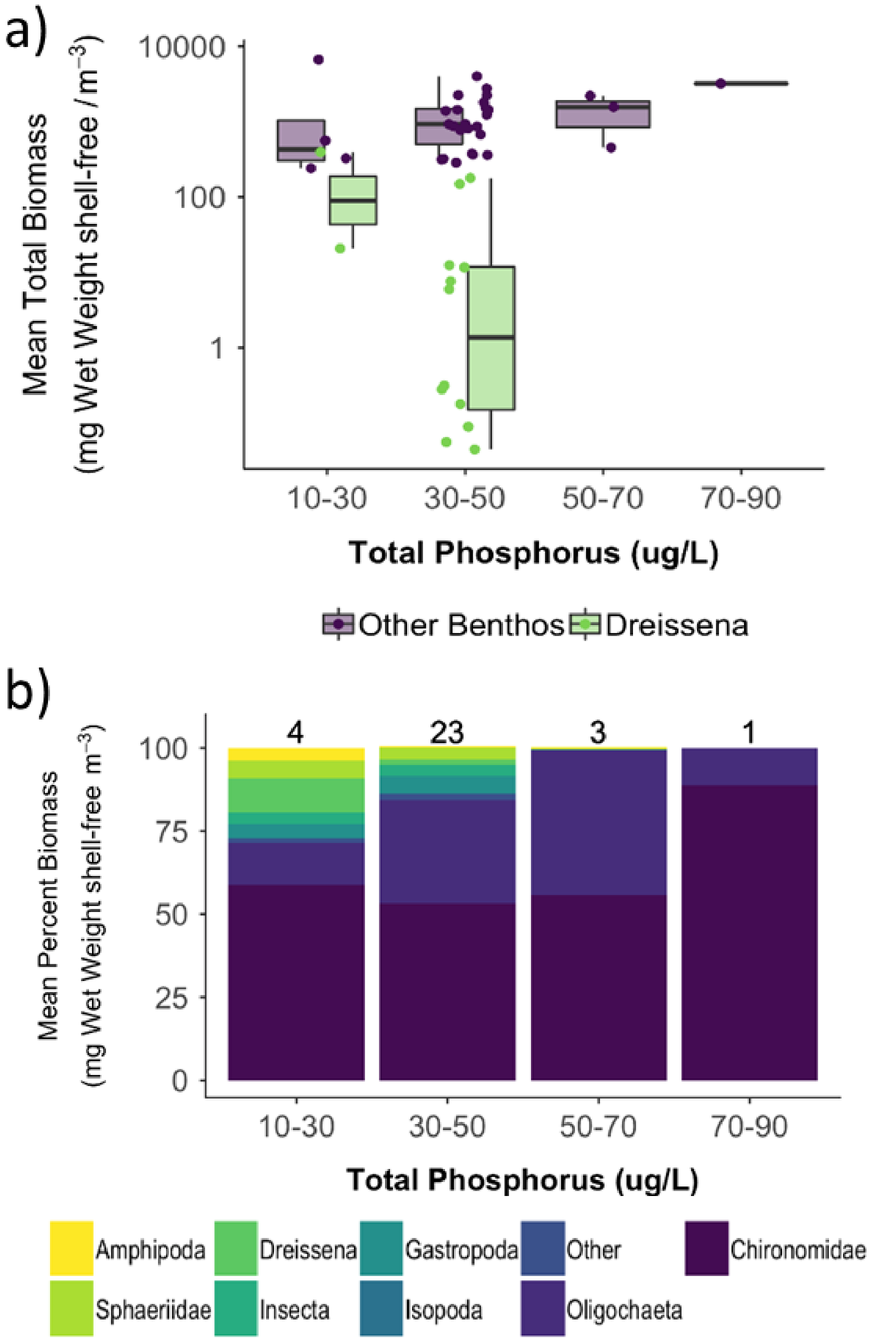


Figure 1.15 *a) Volume transformed biomass of benthic invertebrates in the Upper Bay of Quinte, 1967-2011. Data points represent annual means. Green data represent Dreissena sp.; purple data represent total biomass of all other benthic invertebrates. Boxplots represent median values with 25th and 75th percentiles. b) Community composition of benthic invertebrates in the Upper Bay of Quinte, 1967-2011. Data represent means of annual data for 20 µg* L^{-1} *bins of total phosphorus.*

S1.2 Long Transients or a Regime Shift in the Bay of Quinte?

Ecosystems exhibit a degree of fundamental ecological resilience in that they maintain a capacity to retain their composition, function and relationships during perturbations (Neubert and Caswell 1997). However, it has been shown that these same ecosystems can be subject to abrupt transitions in state, known as regime shifts, as drivers exceed an environmental threshold (Beisner et al. 2003, Scheffer et al. 2001). This ecological hysteresis resists return to the previous state (Carpenter et al. 2001, Hastings et al. 2018), though this is difficult to measure in natural systems since there are few long-term monitoring plans that consistently measure both environmental and population level characteristics. As such regime shifts can be invoked when there appears to be a change in a system even though this may be the result of a continuous change from one state to another in what is termed a transient (Morozov et al. 2020).

Sediment cores suggest that the Bay of Quinte was originally slightly oligotrophic, but that it had shifted to eutrophic by about 1890 as industry and population grew in the area (Warwick 1980, Estepp and Reavie 2015). Deterioration in water quality started to become noticeable in the 1930s and by the 1950s the Bay was classified as hypereutrophic (Hurley and Christie 1977). There were algal blooms and fish kills. Fisheries on the Bay ceased to feasible. Point controls on phosphorus sources such as sewage were instituted in 1977, but there was no dramatic impact on water clarity until 1994.

Currently, it is believed that the change in water clarity was not due to the management of phosphorus control, but rather, was caused by the invasion of of zebra (*Dreissena polymorpha*) and quagga (*D. rostriformis*) mussels. Assessments of the Bay of Quinte Remedial Action Plan state that "In the mid-1990s, zebra and quagga mussels (*Dreissena* spp.) invaded the area, dramatically changing the water clarity because of their filter-feeding capacity" (Doka et al. 2016). The timing of the first detection of dreissenids, concurrent with increased water clarity and increased macrophyte density has been taken as evidence that the mechanism for increasing water clarity was filter feeding by *Dreissena* populations. Nicholls and Carney (2011) state that *Dreissena* colonization induced precipitous decline of phytoplankton and ultimately enabled macrophyte resurgence, and Minns et al. (2004) note that “only after zebra and quagga mussels arrived in 1993-4 … did macrophytes increase substantially”, and further suggests “filtering by the mussels.…reduced chlorophyll concentrations and greatly increased water clarity”. Many papers as such have refered to pre and post-*Dreissena* as phases of change in the Bay of Quinte (e.g. Nicholls et al. 2002, Bowen and Johannsson 2011, Nicholls and Carney 2011, Shimoda et al. 2016, Doan et al. 2018).

However, it is unclear that the correlation between first detection of dreissenids and increases in water clarity really do constitute a regime shift, nor it is clear that, if there was a regime shift the mechanism was solely the effect of *Dreissena* filter feeding. In particular, the density of dreissenid larvae in the water column was very low at the time in question, and it is difficult to understand how the low biomass of dreissenids present at that time could have had such a large impact (Fig. 1.16).

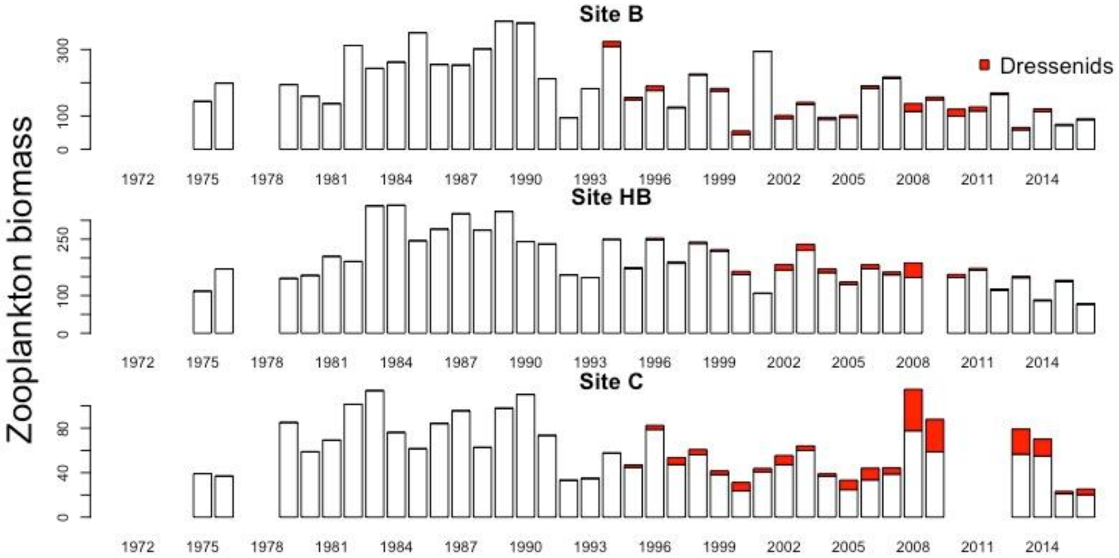


Figure 1.16 *Biomass of zooplankton (g) with amount of dreissenid larvae biomass indicated in red at Belleville (B), Hay Bay (HB), and Conway (C) in the Bay of Quinte, Ontario*

To investigate further, we tested the hypothesis that a change in state in the Bay of Quinte from a turbid to a clear water state could be explained by a long transient (Hastings et al. 2018) following management of phosphorus, rather than a sudden change due to the invasion of *Dreissena* mussels. We describe some of the less studied behaviour of a simple model of regime shifts in lake systems, and show that even in the simple model, changes to phosphorus loading may take a very long time to affect phytoplankton density and water clarity. We then examine phosphorus, chlorophyll-a (as a proxy for phytoplankton), water clarity and aquatic macrophyte data from Bay of Quite using generalized additive models (GAMS) in order to pinpoint those years where there may have been a change in regime to see how well the timing of those changes corresponds to the *Dreissena* invasion. Greater detail of this analysis will be included in a future publication.

Scheffer model, alternative stable states and long transients in lake systems

Scheffer and coauthors (1993, 2001) describe alternative stable states in an ecosystem using a general model. This model is a simple heuristic that can be used to demonstrate the possible dynamics of any system that possess two stable states, where the stability of the states is altered by change in an environmental variation. This model has been applied to ecosystems such as lakes and grassland-savannas (Scheffer 2001).

When used to describe lake eutrophication, we have two potentially stable states: eutrophic and oligotrophic. Putting this more formally in the context of the the Scheffer model:

$$\frac{\partial x}{\partial t} = \frac{cX^{\rho}}{b^{\rho} + X^{\rho}} - \mathrm{bX} + \alpha$$

the change in X is a measure of water clarity given by the amount of phosphorus bound in phytoplankton.

To examine the system behaviour as a function of phosphorus management, we assume constant rates of nutrient recycling (given by the first term of the equation), and rates of flow through the system, *b*. We expect that at the highest levels of phosphorus loading, *a*, only the eutrophic state is stable, and at the lowest levels of loading only the oligotrophic state is stable (Fig 1.17). At intermediate levels of nutrient loading both states are stable. For some levels of phosphorus loading, it can require a larger decrease to phosphorus input to stabilize the desired oligotrophic state than was required to destabilize it (i.e., this system possesses hysteresis: Beisner et al. 2003, Scheffer et al. 2001). We also note that when both states are stable, it is possible for the system to flip between them when subject to disturbance (Zeng et al. 2015).

In the context of this model, the point source control of phosphorus in the 70s was designed to reduce the stability of the eutrophic state in the Bay of Quinte, so that we should then expect the system to naturally return to the only remaining stable state in the system: the clear water oligotrophic state. It is not clear how strong a claim most authors are making regarding the potential regime shift in the Bay of Quinte. They may be suggesting that the change in phosphorus loading did nothing to the stability of the eutrophic state, and it was only the introduction of the zebra mussels, a large press perturbation, that has moved the system to the clear water conditions. In this interpretation, the oligotrophic state is still unstable, and any reduction in dreissenids will cause the system to return to the stable eutrophic state. It could also be suggested that both the oligotrophic and eutrophic state are stable either as a result of phosphorus controls, or not. So there are still two stable states in the system, and the perturbation of the dreissenid mussels introduction moved the system to the stable oligotrophic state. In this case, a reduction in mussels would not necessarily cause the system to shift back to a eutrophic state, although it would certainly be true that the ongoing press perturbation would prevent large stochastic variation from moving the system between the 2 states.

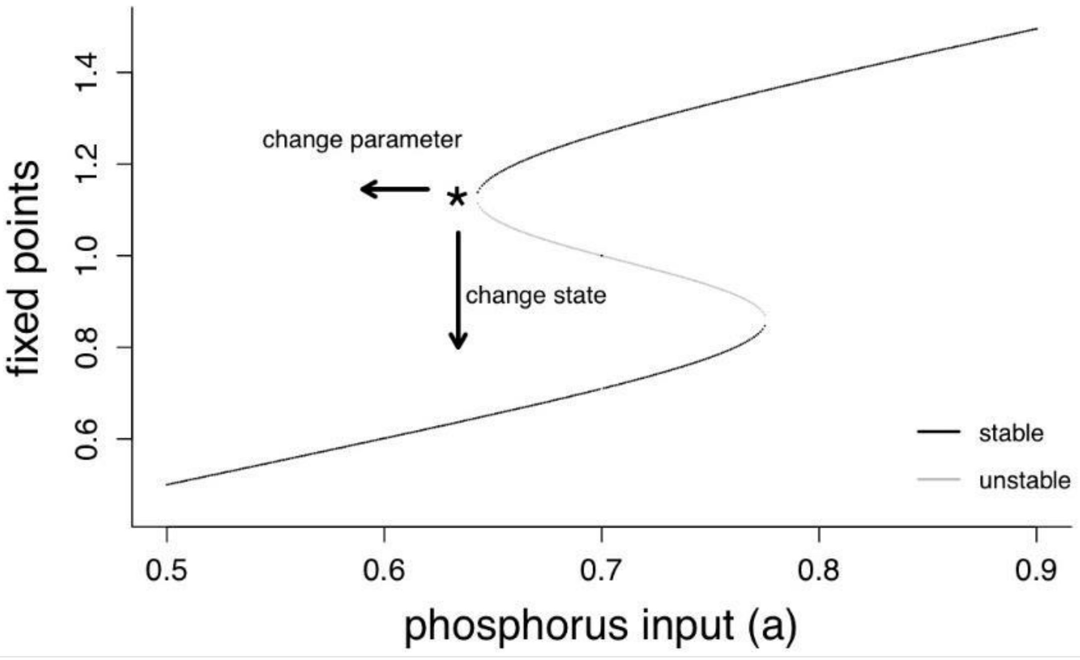


Figure 1.17 *Stable states in Scheffer's model (Scheffer et al. 1997) as applied to lake systems. Solid black lines indicate stable equilibria, while grey lines indicate unstable equilibria, where the axis refers to X, the amount of phosphorus bound in phytoplankton. For some conditions (low a), only the lower fixed point is stable, which corresponds to the oligotrophic state, while for high inputs (large a), only the eutrophic state is stable. At intermediate levels of phosphorus loading there may be two stable states. One can move toward the oligotrophic equilibrium be either changing a parameter values (i.e., reducing phosphorus loading), or by perturbing the system in some way (e.g., adding Dreissena mussels).*

There is however a third and simpler interpretation. It may be that the phosphorus controls took time to increase the stability of the less eutrophic state. Other authors seem to be suggesting that because there was not an immediate response of the system to the phosphorus controls, the system flip must have occurred as a result of the dreissenid mussel introduction. However, the phosphorus controls could have been successful at eroding the stability of the eutrophic state, but the system lingered near that ghost of this former attractor (Hastings et al. 2018, Van Geest et al. 2007).

Ghost attractors are one mechanism that causes long transients in ecological systems (Hastings et al. 2019). We can visualize this phenomena by imagining the stability of the system as a 3D surface (e.g. a quasi-potential surface, Nolting and Abbott 2016). If the stability landscape was altered by management such that the eutrophic state was no longer a stable well, but was still relatively flat, the system could linger in the vicinity of that former stable state for a very long time (Fig. 1.18). If we examined the length of transients in the Scheffer model close to the parameter boundary where the eutrophic state moves from stable to unstable we find that find transients where the system lingered near the ghost attractor for decades, others where it changed immediately to the remaining stable state, and some where the change was rather gradual (Fig. 1.19). The probability that the system will exhibit long transits that linger near the unstable state depends on how close you are to the parameter boundary. Longer transients are more likely the closer to this bifurcation, but certainly transients on the order of 15 years are common at parameter values fairly distant to the parameter boundary (Fig. 1.20).

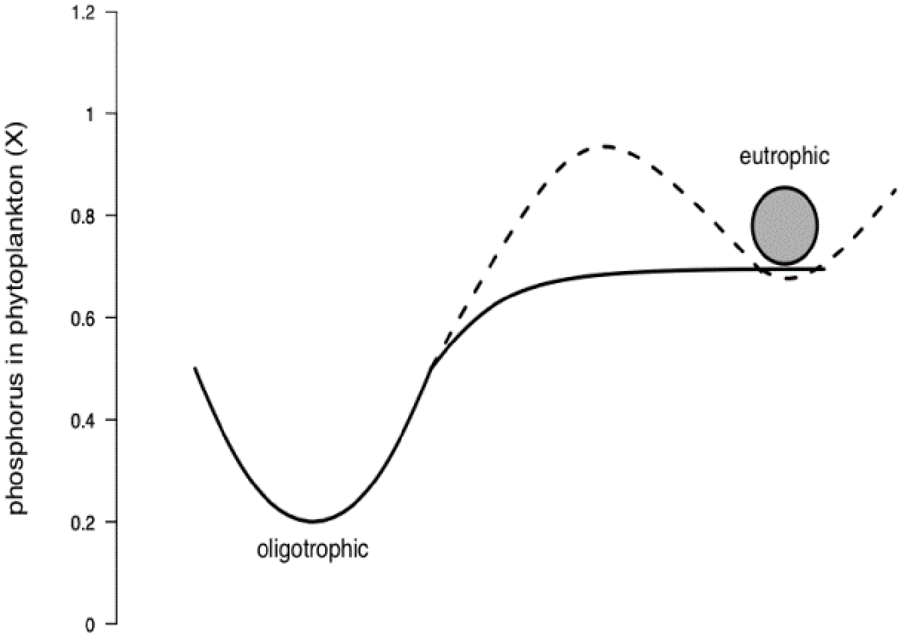


Figure 1.18 *Cartoon visualizing the change to the stability surface of the system before (dashed line) and after (solid line) phosphorus controls. The stability of the eutrophic state has been eroded, as represented by the change in the stability landscape from a well to a flat plane. However, because the surface is flat, the formerly stable eutrophic state acts as a ghost attractor, and the system can linger near this state for some time even though only the oligotrophic state is stable.*

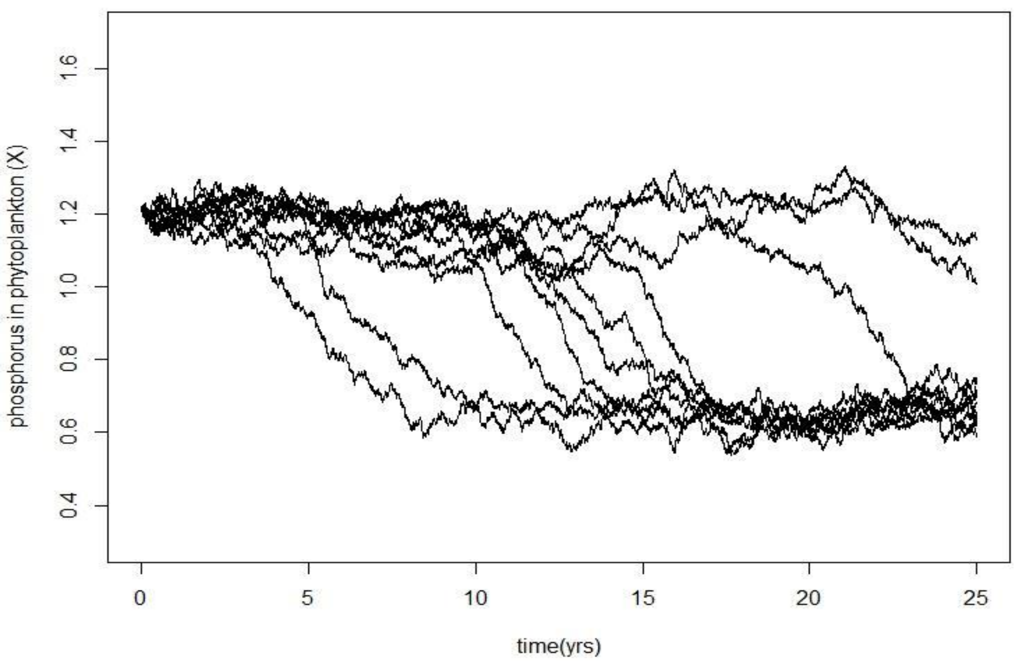


Figure 1.19 *Simulated trajectories of clarity (represented by phosphorus in phytoplankton) from eutrophic conditions in the vicinity of a former attractor, to a stable oligotrophic state, as simulated by the stochastic version of the Scheffer model of alternative stable states (Scheffer et al. 2001) with small amounts of white noise added. This illustrates the magnitude and timing of the response to small changes in conditions.*

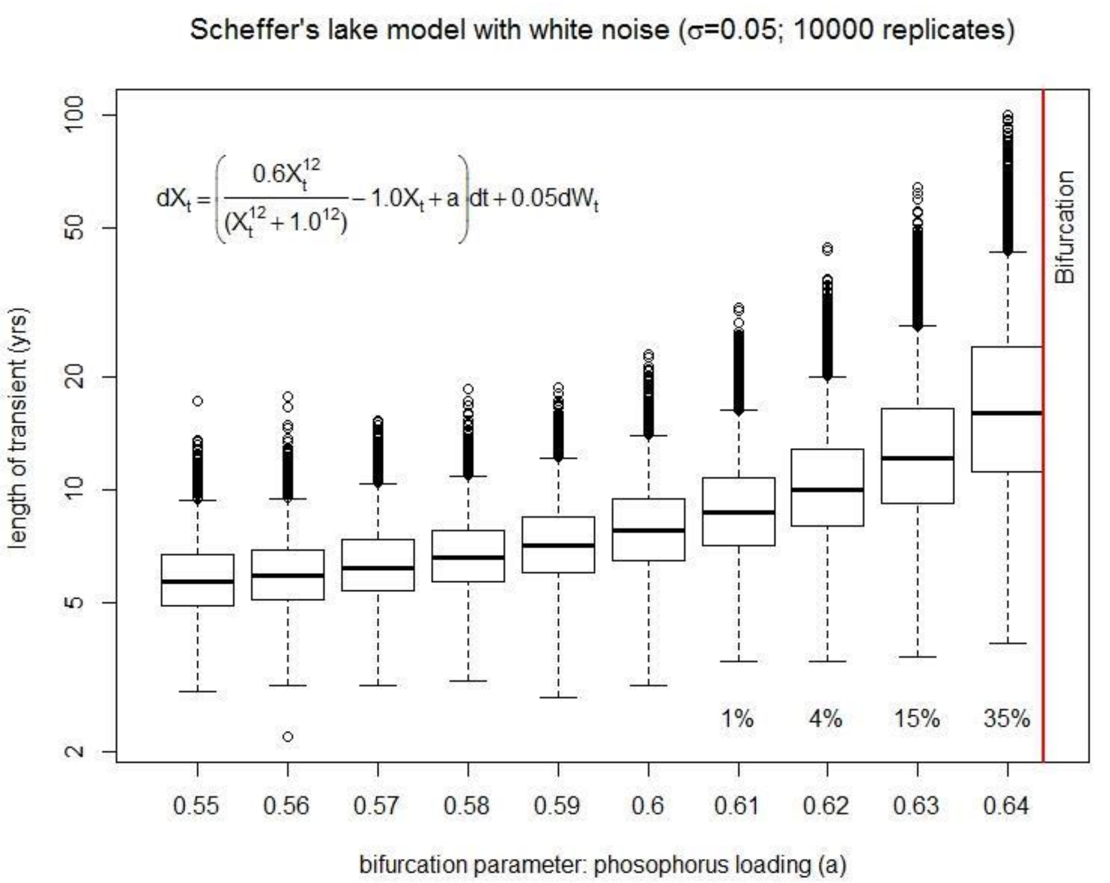


Figure 1.20 *Boxplot of the distribution of the time required to shift from the eutrophic state to the oligotrophic state in 10000 replicate simulations of a stochastic version of Scheffer's model (Scheffer et al. 2001) with given parameter values shown in the equation and a range of values for phosphorus loading (a) for which only the oligotrophic state is stable. The center line of the box is the median transient length, whiskers are 1.5 times the interquartile range, and symbols indicate values for simulations outside this range. Percentages under some bars indicate how many simulations had transients longer than 30 years.*

Analysis of Bay of Quinte data

To examine the hypothesis that the change in water clarity in the Bay of Quinte could be due to a long transient following the institution of phosphorus controls, we used time series from 1972 to 2016 of the annual mean of total phosphorus, chlorophyll *a*, the light attenuation coefficient and aquatic macrophyte density. We used 4 data collection sites (Fig. 1.21): the upper Bay site of Belleville (B), two middle Bay sites of Hay Bay (HB) and Napanee (N), and the lower Bay site of Conway (C).

We used generalized additive models (GAMS) to determine if there was an abrupt change in the rate of change of these measures when dreissenids were detected in the Bay of Quinte. GAMS fit a smooth function to a time series as:

$$E(y_i) = \beta_1 + f(\mathcal{X}_1)\,\mathcal{X}_1,$$

where in this case, the expected value of our metric, at time i, $E(y_i)$ is given by a function, f, of time x. We fit the data using thin plate regression splines and restricted maximum likelihood in the MGV package for R (Wood 2017), and methods developed by Gavin Simpson, Eric Pedersen and others (Simpson 2018, Pedersen et al. 2019).

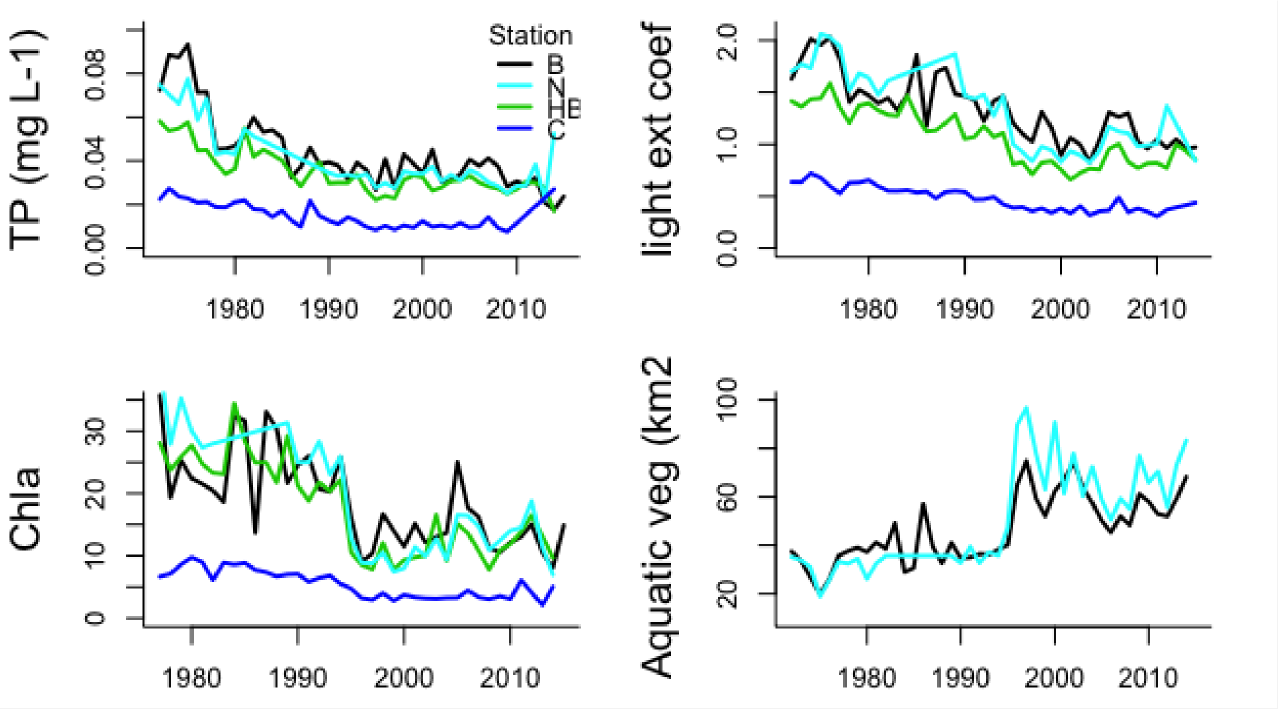


Figure 1.21 *Time series of total phosphorus (TP), chlorophyll-a (Chla), light attenuation coefficient (light ext coef), and submerged aquatic vegetation (Aquatic veg) at 4 sites in Bay of Quinte: Belleville (B), Hay Bay (HB), Napanee (N) and Conway (C).*

If we can describe the time series using a smooth function, we can then differentiate this function to determine when there is a significant rate of change in the system (i.e., when the confidence interval around this derivative does not intersect zero). First of all, we use the technique with stochastic simulations of the Scheffer model to determine what pattern we should expect when there is a regime shift in the system. We simulate the Scheffer model with some additive white noise and an unstable eutrophic state. We then fit a GAM to the output, and then use a finite difference approximation along the curve to get the rate of change. We find that the rate is close to zero when the system is close to the formerly stable eutrophic state, but as the system shifts to the oligotrophic state, the rate of change will become significantly different from zero because of this regime shift. However, the transition from the unstable to stable state could happen early or late in the time series, just depending on the random chance (Fig. 1.22), and this is true regardless of whether the stochastic variation in the system is small or large.

We applied the same analysis to total phosphorus measured at each of the four sites. Total phosphorus decreased (i.e., the rates of change were less than zero) until about 1980. The largest rate of change was at Belleville, closest to the effects in point source controls. There was a much smaller change (not clearly different from zero) at Conway where the system starts to merge with Lake Ontario (Fig. 1.23).

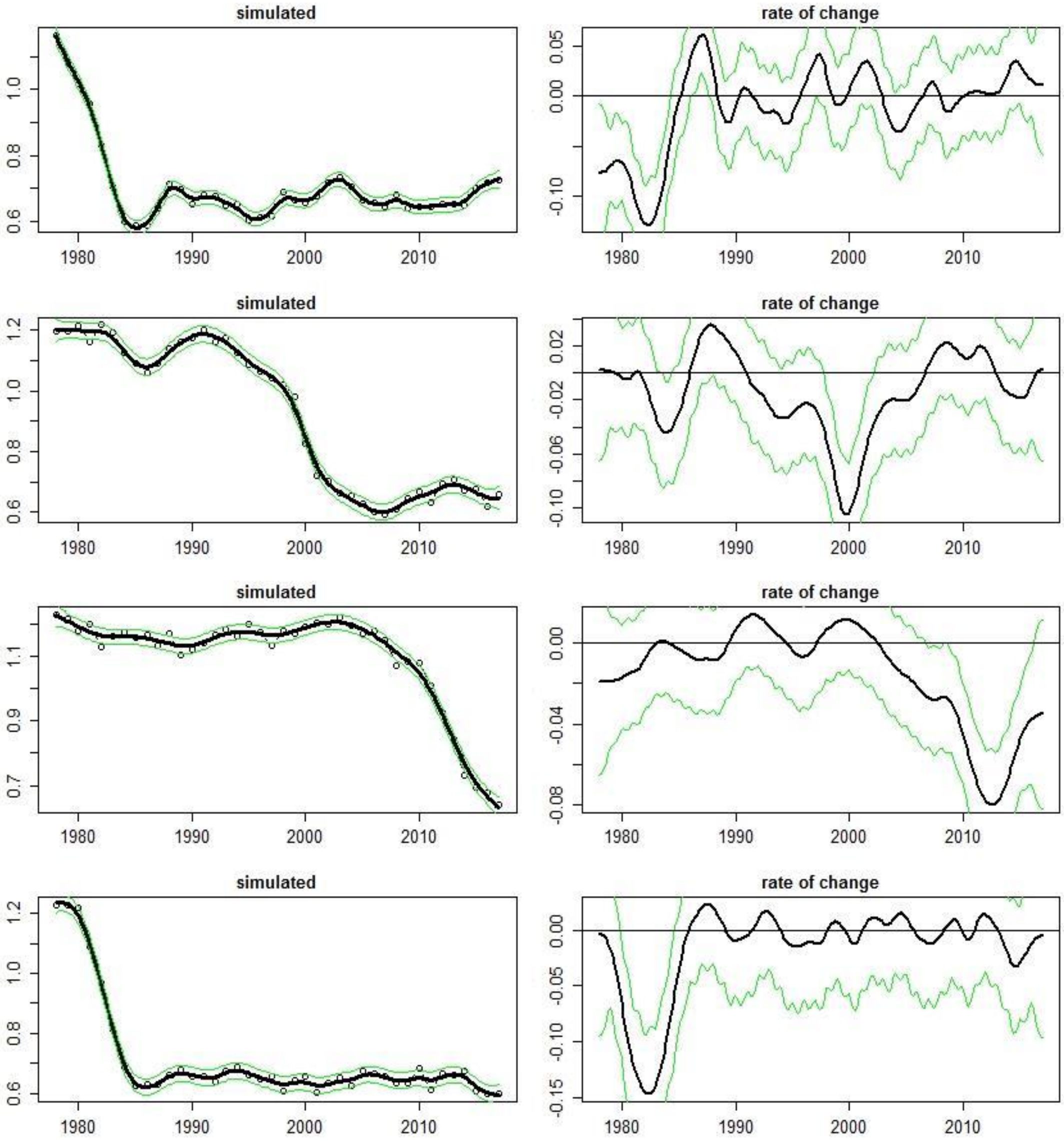


Figure 1.22 *Example simulated data to illustrate how GAM model fits (black solid line) and confidence interval (green solid line) to simulations (symbols and line) of the stochastic version of the Scheffer model, where the system is initially close the unstable eutrophic state. The left hand panels (simulated) are the computer generated time series and model fits, the right hand panels are the rate of change estimated by a finite difference approximation along the curve. Where the confidence intervals of this approximation do not overlap with zero, the system is undergoing a change from one state to another.*

For water clarity, as measured by the light extinction coefficient, the optimal fit for Belleville is a linear with negative slope, not a GAM, which means that there has been no state change for this variation, and the water clarity has steadily increased through time at a rate significantly different from zero. For Hay Bay and Conway the rate of change is significantly different from zero with an increase in clarity beginning in the late 80s and ending early 90s. At Napanee, the rate of change is not significantly different from zero, but there is also missing data, which may reduce our ability to detect such change (Fig. 1.24).

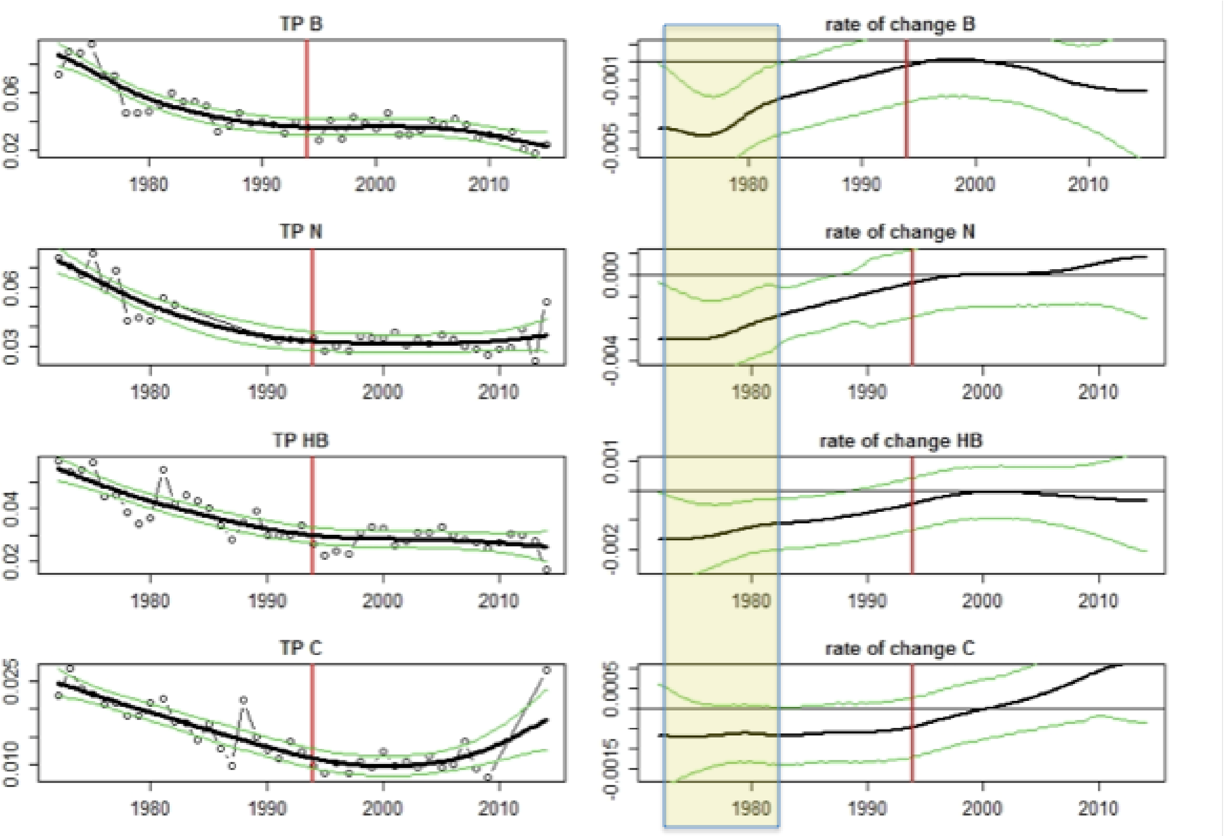


Figure 1.23 *GAM model fits (black solid line) and confidence interval (green solid line) to total phosphorus data (symbols and line) from Belleville (B), Hay Bay (HB), Napanee (N) and Conway (C) in Bay of Quinte, Ontario (left hand panels). Rate of change in total phosphorus estimated by a finite difference approximation along the fitted GAM curve (right hand panels). Yellow shaded box gives approximate time region where the rate of change was significantly different from zero, and red line gives the date of the first detection of Dreissenid mussel larvae in zooplankton samples.*

Chlorophyll-a, which we use as a proxy for phytoplankton density, declines linearly for Belleville and there is no change in state. For Napanee, Hay Bay and Conway, there is evidence of a shift to lower levels from about 1992 to 1996 (Fig. 1.25). However, the change in submerged aquatic vegetation at Belleville and Napanee looks most like a an abrupt regime shift. Here we find an increase in vegetation over a 1 to 2 year period from 1995 – 1997 (Fig. 1.21).

We examined the rates of change over time in total phosphorus, water clarity, chlorophyll-a and aquatic macrophytes at four sites in the Bay of Quinte to determine if there had been a regime shift in the system, and further to ascertain if the timing of any such shift was coincident with the first detection of dreissenid mussels in this system. We find that water clarity started to increase before the first detection of *Dreissena* larvae, and that that it stopped increasing while the population of these species was relatively small. Together, these results suggest that decreasing phosphorus and a delay in response to this decrease may have been the ultimate mechanism that increased lake clarity in the Bay of Quinte.

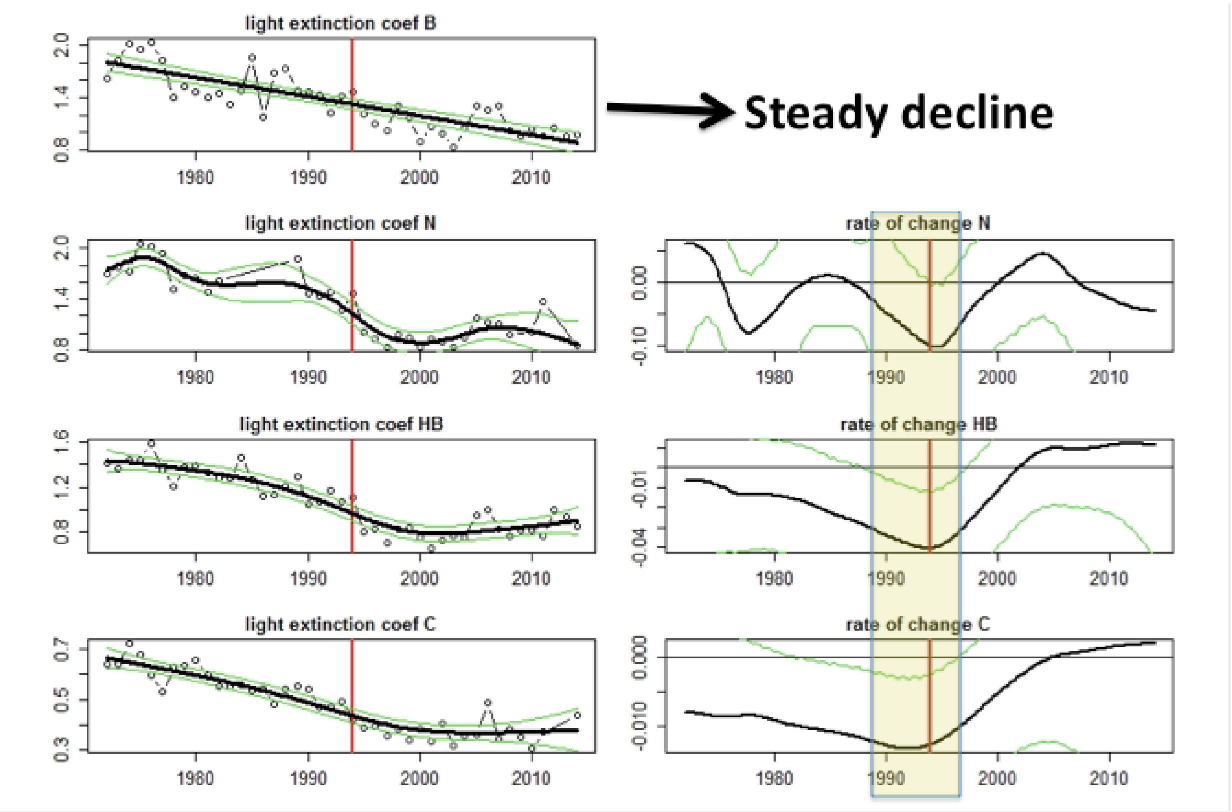


Figure 1.24 *GAM model fits (black solid line) and confidence interval (green solid line) to light extinction coefficient data (symbols and line) from Belleville (B), Hay Bay (HB), Napanee (N) and Conway (C) in Bay of Quinte, Ontario (left hand panels). Rate of change in the light extinction coefficient is estimated by a finite difference approximation along the fitted GAM curve (right hand panels). Yellow shaded box gives approximate time region where the rate of change was significantly different from zero, and red line gives the date of the first detection of Dreissenid mussel larvae in zooplankton samples.*

For the upper and middle bay stations, we find evidence of a decrease in phosphorus that slowed or stopped around 1984. However, this pattern is not significantly different from no change for the lower Bay site of Conway. In the upper Bay site of Belleville, we find a continual increase in lake clarity and a concomitant decrease in phytoplankton density, as indicated by chlorophyll-a. For mid and lower Bay sites (Hay Bay and Conway) we find a period of increasing lake clarity from 1988 to about 1996. This trend was not significant for Napanee, which has more missing data. At these lower Bay site there is a decrease in chlorophyll-a from about 1992 to 1997, which could be interpreted as a regime shift. However, this change occurred when dreissenid densities were still very low, and did not continue as densities increased. We do find evidence of a relatively sudden shift in macrophyte density at Napanee and Belleville sites. However, at the Belleville site, this sudden shift is not preceded by a sudden shift in water clarity, while at Napanee, there is a shift in water clarity, but it started before dreissenids were detected which indicate that dreissenids are not the direct cause of these changes.

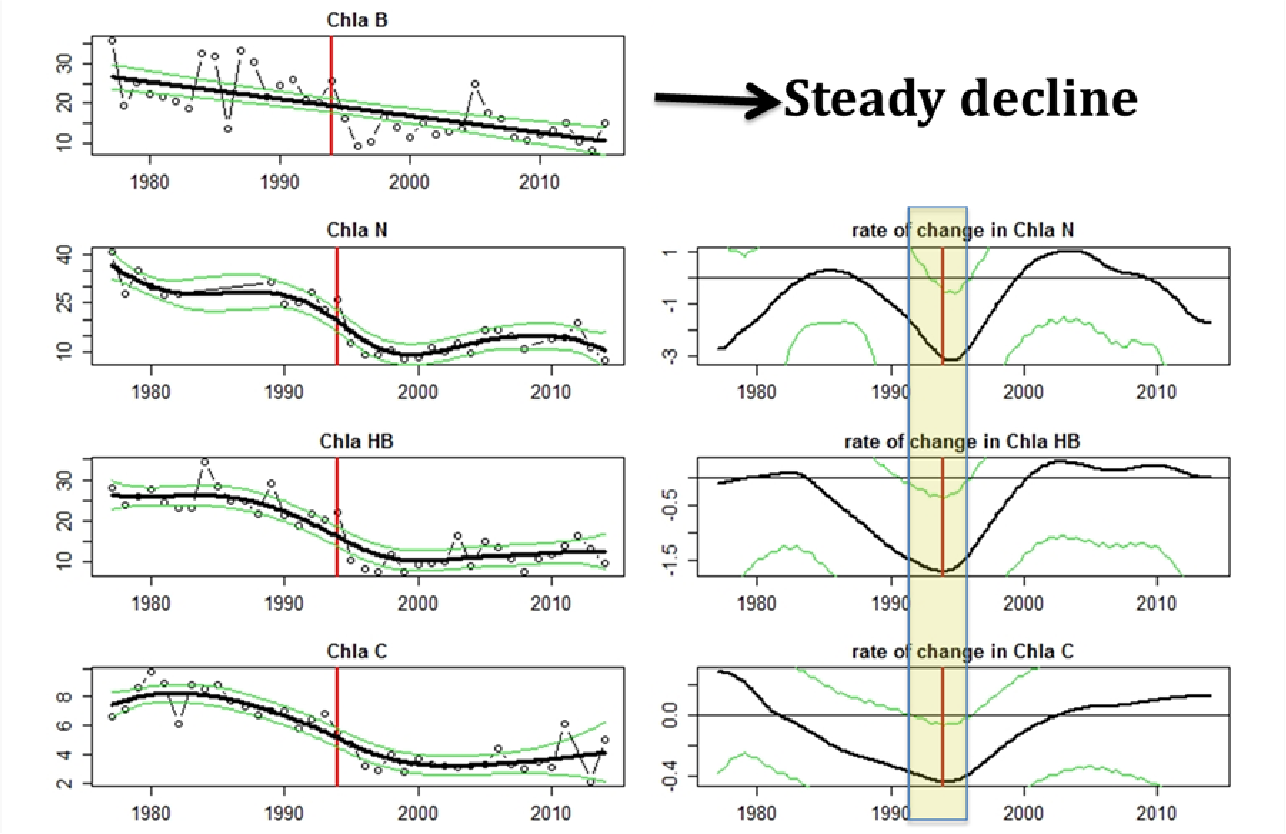


Figure 1.25 *GAM model fits (black solid line) and confidence interval (green solid line) to Chlorophyll A data (symbols and line) from Belleville (B), Hay Bay (HB), Napanee (N) and Conway (C) in Bay of Quinte, Ontario (left hand panels). Rate of change in chlorophyll-a is estimated by a finite difference approximation along the fitted GAM curve (right hand panels). Yellow shaded box gives approximate time region where the rate of change was significantly different from zero, and red line gives the date of the first detection of dreissenid mussel larvae in zooplankton samples.*

Since the changes in water clarity followed a period of decreasing total phosphorus, and since changes in phytoplankton density, as measured by chlorophyll-a occurred during a period of increasing water clarity that preceded first detection of *Dreissena* sp., it is unclear that the invasion of this species is the mechanism that caused a change in state in the Bay of Quinte. Certainly, if dreissenid mussels are the mechanism of change we would have to further postulate that 1) they were in the Bay around 1988 but were not detected, 2) they have large impact at very low densities, and 3) They have smaller impacts at the larger densities they were found at in later years. A simpler explanation may be that it took time after the change in phosphorus levels for lake clarity to increase and chlorophyll-a to decrease, such that there was a gradual rather than sudden regime shift in this system, starting about 11 years after phosphorus controls were instituted. In fact, the analysis suggests that the upper Bay is still changing. In particular, in the upper Bay, there is no evidence of a regime shift in water clarity, although there is a sudden increase in macrophytes. Therefore, while the invasion of dreissenid mussels likely reinforced this change in the mid to lower Bay sites, they are unlikely to be the primary mechanism of change (See Bowen et al. 2020 for further discussion).

S2 Plankton Indicators for BUI 13 in the Bay of Quinte

S2.1 Phytoplankton Indicators

A good indicator for a Beneficial Use Impairment (BUI) should be able to detect change in ecosystem function that is related to impairment of the system and / or linked to management activities. In the Bay of Quinte area of concern (AOC), the phytoplankton population impairment is bottom up in nature due to excess nutrients (BQ RAP 1987) and directly linked to that of BUI 8: Eutrophication. There have been major reductions in phosphorus loadings to the Bay since the 1970s (Kinstler and Morley 2017) when phosphorus was implicated as the cause of eutrophication in the Great Lakes and both Canadian and US governments mandated policies of phosphorus reduction [International Joint Commission (IJC) 1988]. AOC activities intended to further reduce nutrient input into the Bay have been among the main management actions undertaken in the AOC since the program started in 1987 (BQ RAP 1993). Although phytoplankton is the base of the food web, excess nutrients can lead to overproduction of species that are not ideal food for herbivorous zooplankton (Kerfoot and Kirk 1991, Tõnno et al. 2016) and therefore reduce food resources available to fish. The majority of the phytoplankton biomass in the Bay of Quinte has been a combination of filamentous diatoms [*Aulacoseira* (prev. *Melosira*) and *Stephanodiscus*] and nitrogen fixing cyanobacteria [*Dolichospermum* (prev. *Anabaena*) and *Aphanizomenon*; Nicholls and Hurley 1989] which are very much related to eutrophication but may not be ideal food resources. Though some species of zooplankton are capable of handling filamentous algae (Leitão et al. 2020), filamentous forms have been shown to reduce food gathering abilities of *Daphnia* (a highly efficient grazer and valuable food source for fishes) through mechanical interference or entanglement (Gliwicz and Siedlar 1980; Sahuquillo et al. 2007) and decreased fecundity (Vaga et al. 1985). Cyanobacteria (both filamentous and colonial forms) have the potential to produce toxins that affects other components of the ecosystem. The negative effect of eutrophication on energy transfer within the aquatic food web resulted in the initial measure of phytoplankton status to be "*a positive change in species composition with a decrease in nuisance and eutrophic species*" and "*a decrease in spring, summer and fall biomass of these nuisance species*" (BQ RAP 1987). To clarify and set defined numeric targets on these measures, Currie et al. (2017a) recommended a set of 7 indicators to measure phytoplankton status related to two specific targets (Table 2.1.1 and below). Feedback on that report suggested a need to reduce and simplify the recommended phytoplankton indicators of 2017.

The first target for phytoplankton "*Demonstrate a positive change in phytoplankton species composition with a decrease in nuisance and eutrophic and noxious indicator species that may impair food web function*" included three associated indicators:

1. A decrease in nuisance phytoplankton to less than 50% of the biomass (i.e. < 2.5 g m^{-3} in the upper bay offshore based on the existing target for BUI 8 of phytoplankton biomass of 4 – 5 g m^{-3})
2. A decreasing trend in filamentous and colonial / mucilaginous Cyanobacteria (both in terms of biomass and relative composition)
3. A decreasing trend in filamentous diatoms (biomass and % composition).

These indicators include taxa that may form unsightly blooms, produce toxins and /or not be readily ingested by zooplankton grazers (or avoided altogether). The energy contained in these taxa is not as readily transferred up the food web and into fish biomass but is retained within the ecosystem reinforcing eutrophication and shunting energy to the microbial community. These indicators affect the quantity of zooplankton available to feed higher levels of the food web and respond directly to the management actions implemented to reduce nutrient levels. In order to simplify any future monitoring, "*total filamentous algae*" is considered as a new potential indicator. Filamentous algae is a generalized group of taxa based on morphology (Wehr et al. 2015). In the case of the Bay of Quinte, that means primarily species of *Aulacosiera* and *Dolichospermum* which were identified as nuisance taxa in the previous assessments (i.e. Currie et al. 2017b). They are large and easily recognisable so biomass can be estimated by non-specialized taxonomists with standard equipment, time and a little training. Filamentous algae are more easily scaled from microscopic counts to volumetric biomass than colonial forms, the other major group of nuisance algae. It is anticipated that the use of this new indicator will simplify the assessment of target 1.

Blooms of colonial cyanobacteria (blue-green) taxa (e.g. *Microcystis, Coelosphaerium)* are recognized as impairments within aquatic systems (Watson et al. 2008). These cyanobacteria blooms appeared intermittently in the Bay after 1998 (Nicholls et al. 2002), although they were also occasionally seen in the 1970s and not seen through the 1980s. Cyanobacteria blooms are becoming more frequent around the world and are the focus of many studies to determine their cause (i.e. Huisman et al. 2018, Watson et al. 2008). Theories include climate warming (i.e. Elliot 2012) and changes to the food web due to invasive species such as zebra mussels (Knoll et al. 2008). Even nontoxic cyanobacteria are generally considered nutritionally inadequate for zooplankton grazers although this is not consistently observed (de Bernardi and Guissani 1990). When cyanobacteria are the dominant phytoplankton, the zooplankton community is usually dominated by small-bodied cladocerans, rotifers and copepods as larger zooplankton are thought to be more negatively impacted than smaller taxa (Zhang et al. 2014; Hanazato 1989). Because of the recent increased appearance of cyanobacteria in the Bay and the potential impact on zooplankton grazers, two indicators were added to this assessment; Colonial cyanobacteria biomass and the ratio of filamentous to colonial cyanobacteria. Colonial cyanobacteria should also be relatively easy to identify and enumerate in phytoplankton samples; likewise differentiating between cyanobacteria and other filamentous algae could also be accomplished fairly easily with some taxonomic training.

The second target for phytoplankton "*Demonstrate a positive change in phytoplankton species composition with a decrease in nuisance and eutrophic and noxious indicator species that may impair food web function*" had two associated indicators:

1. An increase in the more edible phytoplankton taxa to approximately 30% of the biomass
2. An increase in the biomass of Chrysophyceae with an ideal target of > 15% of total phytoplankton biomass.

This particular set of phytoplankton indicators proposed in 2017 relate to food availably for zooplankton grazers. The majority of zooplankton in the Bay of Quinte are herbivorous including large bodied *Daphnia* and smaller bodied *Bosmina* and *Eubosmina* (Bowen and Johannsson 2011). Edibility was based on taxonomy and categorized using physiological features such as colony formation (none or small colonies), size (smaller than carapace opening of filter feeding zooplankton) and shape (no spines of projections). As there are a variety of methods by which zooplankton graze this is not ideal but is the extent of our capabilities. Chrysophyceae is a highly edible taxonomic group (based on size) known to relate to trophic state (Sandgren 1988). In order to calculate these indicators, the entire phytoplankton community needs to be assessed which requires a high level of taxonomic expertise and overall effort. For ease of future assessment, an additional factor was created that does not require all phytoplankton taxa be counted. This new indicator is the ratio Chrysophyceae to total filamentous biomass and would require training to identify and calculate biomass for Chrysophyceae but not other phytoplankton taxa as is required to calculate % Chrysophyceae. The effort required to train to identify and the time needed to count Chrysophyceae will be higher than for filamentous algae but all indicators can be counted from the same sample and equipment.

In this analysis, we will test the effectiveness of these new simplified indicators: *Total filamentous*, *Cyanobacteria ratio*, and *Chrysophyceae ratio* against those proposed previously for determining the state of the phytoplankton population (Table 2.1.1; Currie et al. 2017a). The goal of this exercise is to offer a streamlined monitoring framework for both the identification and assessment of phytoplankton populations.

Methods

Given the variability seen in phytoplankton and environmental data sets due to seasonality (Currie et al. 2015, Currie et al. 2017b) we used May – October arithmetic means when available for the purposes of data analyses. In cases where an arithmetic mean could not be computed because too few individual dates were enumerated (i.e. at a frequency of less than 1 per month), we used composite phytoplankton samples to complete the analysis (See Currie et al. 2017b for details). All statistical analysis was conducted in JMP v15.1. All potential indicators were plotted to see if the distribution was normal and if not were transformed prior to analysis (Table 2.1.1).

The lower bay (Conway) is very different from the AOC, which is in the upper bay (Belleville, Napanee), in terms of depth and influence by the main lake. An upper bay only analysis will give a clear indication how the phytoplankton groups are changing within the Area of Concern. Including data from the lower bay utilizes a wider range of environmental conditions which may allow for better characterization of each parameter.

The parameters *Total filamentous*, *Filamentous diatoms*, *Filamentous cyanobacteria* and *Nuisance* are correlated as they are different combinations of the same set of eutrophic, less palatable taxa. Additionally, *%Chrysophyceae* and *Chrysophyceae ratio* are expected to be correlated as the main contributor to biomass in Quinte phytoplankton samples is filamentous algae. To evaluate the extent of collinearity, multivariate analysis was run using the pairwise method to find strongly correlated parameters (R^2 over 0.75) and remove the parameter that is less related to total phosphorus (TP) and Total Kjeldahl Nitrogen (TKN).

A two-pronged assessment of potential phytoplankton indicators was used. First, Principal Components Analysis (PCA) was run to determine potential indicators which contribute to overall variability in the phytoplankton data. Second, a stepwise analysis was used to determine which components were the strongest descriptors of the phytoplankton community. These assessments were undertaken twice, the first time using data from only the upper bay (B, N) and the second time using the full dataset that includes the middle (HB) and lower bay (C). Next relationships to nutrients were determined using regression to select which would be the best indicator for management actions (i.e. nutrient control) and multiple regression modelling to determine if other factors beyond nutrient were important in influencing the indicator.

Results

A. Upper Bay Data only (B and N)

The multivariate correlation matrix showed that *Total filamentous* has the strongest relationship with TP (R^2=0.75) and TKN (R^2=0.85). Several nuisance indicators have correlations with R^2 of over 0.75 with *Total filamentous* and were removed from the future analysis (*Filamentous diatom*, *Nuisance*, *Nuisance cyanobacteria*; Table 2.1.2). Additionally, *%Chrysophyceae* was strongly related to *Chrysophyceae ratio* but more weakly related to the environmental variables and so was also removed from future analysis. Although *Chrysophyceae ratio* was also strongly related to *Total filamentous*, it was left in the analysis because having an indicator that reacts in the opposite direction to total phytoplankton biomass is important in discerning the true state of the phytoplankton community. With these parameters removed 8 categories (*Total filamentous*,

%Nuisance, *%Filamentous diatom*, *Cyanobacteria ratio*, *Colonial Cyanobacteria*, *%Nuisance cyanobacteria, %Edible* and *Chrysophyceae ratio*) remained for input into the analysis.

Principal Components Analysis (Fig. 2.1.1, Table 2.1.4) showed that *Chrysophyceae ratio* and *Total Filamentous* are the two most important factors making up axis 1 and explain 42% of the variability in the data. The second axis explained an additional 21% of the variability and was strongly influenced by *%Filamentous diatom* and *%Nuisance cyanobacteria*. A third axis explaining an additional 19% of the variability was influenced by *Colonial cyanobacteria* and the *Cyanobacteria ratio*.

The model effects mentioned above were run using a forward Stepwise regression to determine the best predictors of total phytoplankton biomass. The best fit model created included 4 of the 8 entered effects (Table 2.1.5). *Total filamentous* explained most of the variation, followed by *%Nuisance*, then *Colonial cyanobacteria* and finally *Nuisance cyanobacteria*.

B. All Station Data (B, N, HB & C)
When including all stations, the multivariate correlation matrix showed that *Total filamentous* has the strongest relationship with TP (R^2=0.85) and TKN (R^2=0.90). Many nuisance indicators have strong relationships with total filamentous with R^2 of over 0.75 and were removed from the future analysis (Table 2.1.3). With these parameters removed 5 categories remain (*Total filamentous*, *Colonial cyanobacteria*, *%Filamentous diatom*, *Cyanobacteria ratio*, *Chrysophyceae ratio*).

Principal Components Analysis (Fig. 2.1.2, Table 2.1.4) showed that as in the upper bay alone the *Chrysophyceae ratio* and *Total filamentous* are the two most important factors making up axis 1 and explain 49% of the variability in the data. The second axis explained an additional 23% of the variability being similar to the 3rd axis of the upper bay only analysis and was strongly influenced *Colonial cyanobacteria* and the *Cyanobacteria ratio*.

The model effects mentioned above were run using a forward Stepwise to determine the best predictors of total phytoplankton biomass. The best fit model created included 3 of the 8 entered effects (Table 2.1.5) and confirmed *Total Filamentous* as the most important factor. The next most important factors were *%Filamentous diatom* and *Chrysophyceae ratio* (Table 2.1.5).

Proposed indicators
Overall, *Total filamentous* is the top descriptor of phytoplankton biomass (May to October average) as determined using PCA and stepwise analysis for both upper bay and all station data. A good indicator is one that will respond to management actions (nutrient control) in a clear direction. Of the proposed indicators, *Total filamentous* showed the strongest relationships with nutrients. It shows a strong positive relationship with both total phosphorus (Total filamentous = -1305 +148*TP; R^2=0.67) and Total Kjeldahl Nitrogen (-4418 + 15349*TKN; R^2=0.74; Fig. 2.1.3) and so lower values indicate an improved phytoplankton community. Multiple regression models were created using a combination of nutrient, climate and zooplankton effects (Table 2.1.6). All the tested models were significant and R^2 ranged from 0.4 to 0.8. The two best models both included TKN as highly significant, reinforcing the nutrient connection and management control. Other significant factors were light attenuation, pre or post 1983 and winter precipitation. Due to its size and shape *Total filamentous* are easy to enumerate and require little taxonomic training and low counting time compared to evaluation of the entire phytoplankton community. So it could be concluded that reduction in *Total filamentous* biomass is a sound indicator of phytoplankton community improvement in the Bay of Quinte.

Chrysophyceae ratio is the next important indicator describing the phytoplankton data set in the PCA analysis (AOC and all station data) but is not significant in describing total phytoplankton biomass using Stepwise for the upper bay alone. This ratio responds negatively to with increased nutrient levels (*Chrysophyceae ratio* =-0.8247-0.0218*TP; R^2=0.46 and *Chrysophyceae ratio* = -0.2768 - 2.4247*TKN; R^2=0.60 R^2=0.64; Fig. 2.1.4) so is a good indicator especially as it moves in the opposite direction of *Total filamentous* with higher values indicating an improved phytoplankton community. Least squared models were created using a combination of nutrient, climate and zooplankton effects to test which factors most affect *Chrysophyceae ratio* (Table 2.1.6). All the tested models were significant and R^2 ranged from 0.5 to 0.8. TKN and TP were both highly significant effects and other significant factors were light attenuation, total phytoplankton biomass and *% Edible*. It is easier to identify just Chrysophyceae than all phytoplankton taxa so it would be a preferred indicator to *%Edible*. *%Edible* is the second most important effect in the PCA for the upper bay and gives important information on the state of food resources for grazers but it is not recommended for three reasons as an indicator. First, it necessitates identification of the entire phytoplankton population which requires a great deal of effort by a highly skilled taxonomist, secondly it responds in the same direction and almost to the same extent as *Chrysophyceae ratio* in the PCA and is redundant (Fig. 2.1.1) and thirdly the relationship with nutrients is highly variable (Fig. 2.1.5).

Colonial cyanobacteria are important in the description of the phytoplankton community as shown in the stepwise and upper bay PCA analysis and are relatively easy to identify. *Colonial cyanobacteria* appear in the upper bay phytoplankton community at all levels of TP but show the highest range in values under 45 µg L^{-1} TP (Fig. 2.1.6). Using *Cyanobacteria ratio* does not improve the relationship with nutrients which has a R^2 under 0.2 (Fig. 2.1.6). A suitable model to describe Cyanobacteria ratio or *Colonial cyanobacteria* was not found using least means squares modeling as these are influenced by additional factors besides those tested that drive the system towards high or low values. Although not neatly related to nutrient concentrations, high biomass of colonial cyanobacteria can cause surface scums and have the potential to release toxic compounds. For this reason it is important to monitor them as an indicator of the quality of the phytoplankton community.

Trends in Final Parameters

Total filamentous biomass has shown a decreasing trend at all 4 stations (Fig. 2.1.7), but most importantly in the upper bay AOC area indicating an improving phytoplankton community. During the first stanza (1972 – 1983 prior to listing as an AOC) *Total filamentous* biomass was over 8 g m^{-3} and highly variable between years while in the most recent stanza (2001 – 2018) *Total filamentous* biomass has decreased to an average of 2.4 g m^{-3} and the variability between years has also decreased. The rate of decrease is highest in the AOC at station B. In the most recent stanza (2001 – 2018) a significant trend is only seen at B and HB (Fig. 2.1.7). In order to see how Quinte compared to the wider phytoplankton communities of Lake Ontario, indicators were compared by region (Fig. 2.1.8). *Total filamentous* biomass is expected to be lower in deeper regions given that there is more of an opportunity for filamentous algae to settle below the thermocline instead of being mixed up to the surface as in shallow regions. The deeper areas are represented by BQ mid and lower, Hamilton and LO offshore regions. Hamilton Harbour is another eutrophic AOC, while Toronto Harbour is an AOC that is meso-oligotrophic. *Total filamentous* biomass in Hamilton Harbour ranged from 200 to 1250 mg m^{-3} which is lower than the same years in Quinte when the range at B was 1200 to 2100 g m^{-3} but similar to 2017 – 2018 values. Total filamentous is much lower in offshore Lake Ontario (96 to 170 mg m^{-3}) as well as in the nearshore where stratification is unlikely (8 – 330 mg m^{-3}). In Toronto Harbour the range is from 80 to 150 mg m^{-3} similar to the main lake and lower bay values. Given that Quinte is a shallow eutrophic environment with the original eutrophication target of phytoplankton biomass < 5000 mg m^{-3} and the 2017 target of 50% nuisance taxa, a May – Oct mean *Total filamentous* biomass target of 2.0 g m^{-3} or less is proposed to allow for greater filamentous biomass due to the shallow depth and phosphorus enriched sediments of the bay. This will account for the contribution of colonial nuisance taxa to total total biomass. We acknowledge that this target is relatively high and that eutrophic conditions specifically algal blooms will likely persist with this target being met. We therefore recommend reiviewing these targets once the proposed Phosphorus Management Strategy is implemented. In the longer term, a lower target for filamentous algae may be desirable.

The *log Chrysophyceae ratio* also shows a consistent trend over the study period, this time increasing from a mean of -2.0 in the upper bay during stanza 1 (1972 – 1983) which has increased to -1.4 in the most recent stanza (2001 – 2018; Fig. 2.1.9). The rate of increase is highest at C and lowest at N. A positive trend in this indicator is only seen at station B during the most recent stanza (2001 – 2018) with the last few years at B better than seen at N and HB. When comparing the recent values with other regions of Lake Ontario all regions in Quinte are in range with Hamilton where the ratio ranged from -1.87 to -0.48 (Fig. 2.1.9). Quinte was found to have a lower ratio than Toronto Harbour (-0.03 to 1.5) and the main lake stations (-2.2 to 2.5). A target of -1.0 (10%) or higher should be expected for the upper Bay of Quinte.

The biomass of *Colonial cyanobacteria* is generally unchanged over the time series except at HB where a significant increase was seen (Fig. 2.1.10). This is unexpected given the decrease in phosphorus and filamentous biomass seen moving from the upper to lower bay and may represent greater downstream transport of colonial cyanobacteria to the lower bay compared to downstream movement of filamentous taxa. The field teams note that scums have been seen in the lower bay even though overall phytoplankton biomass was much lower than in the earlier decades but has increased in 2017 and 2018. *Colonial cyanobacteria* biomass is in the same range in the AOC and Lower bay as was measured in Hamilton harbour and is very low in the main lake and Toronto stations (Fig. 2.1.9). Arhonditsis et al. (2019) proposed "*probabilistic water quality criteria, whereby the compliance rule stipulates that no more than a stated number of pre-specified water quality extremes should occur within a given number of samples collected over a compliance assessment domain*" as a more useful target than an annual mean; for *Colonial cyanobacteria,* we agree. Fig. 2.1.11 shows the individual measures of *Colonial cyanobacteria* biomass and the wide range in values seen and see that there will be a number of bloom days even when the annual mean biomass is low.

Summary and Recommendations

While detailed taxonomic assessments of the phytoplankton community provides the best information about the community, in the absence of detailed taxonomic assessment, we recommend that future monitoring programs focus on the assessment of total filamentous phytoplankton, colonial cyanobacteria and Chrysophyceae. The first is relatively simple to assess and could be accomplished with minimal training. The next two require some taxonomic expertise but one individual with a moderate level of taxonomic training could make these assessments.

With these limited taxonomic assessments, we propose the following indicators for Target 1, Decrease in nuisance, eutrophic and noxious indicator species:

i. Total filamentous biomass less than 2.0 g m^{-3} (annual average)
ii. Biomass of colonial cyanobacteria less than 1 g m^{-3}, 80% of the time (with a minimum of 6 samples collected evenly through the season)

Likewise, for Target 2, Increase in phytoplankton as a food resource for zooplankton grazing:

iii. Ratio of Chrysophyceae to Total Filamentous phytoplankton greater than 10% (annual average)

Our analysis shows that focusing on key measures of total filamentous phytoplankton, colonial cyanobacteria and Chrysophyceae will provide adequate data on the phytoplankton community status for the assessment of BUI 13. We caution however, that a simplified data set of this nature will be of limited value outside of the scope of this portion of Bay of Quinte BUI 13, and may not be applicable to trophic ratios requiring total phytoplankton biomass.

Table 2.1.1. Proposed Phytoplankton measures from Currie et al. 2017a. All are based on annual mean values using at minimum monthly sampling from early May to late October. Data distributions were assessed and transformed to normal using JMP v15.1 Continuous fit menu: SHASH (sinh-arcsinh), Johnson Sb (bounds, both tails). For details see: *https://www.jmp.com/support/help/en/15.2/index.shtml#page/jmp/continuous-fit-distributions.shtml*.

Target 1: Decrease in nuisance, eutrophic and noxious indicator species.			
Indicator	**Name**	**Example taxa**	**Transformation**
Nuisance phytoplankton to less than 50% of the biomass (i.e. < 2.5 g m^{-3})	Nuisance %Nuisance	All listed for Nuisance BG BM and Fil Diatom BM	SHASH SHASH
Decreasing trend filamentous and colonial cyanobacteria biomass	Nuisance cyanobacteria	*Dolichospermum, Planktothrix, Aphanizomenon, Microcystis Aphanocapsa, Rhabdoderma,*	*Johnson Sb*
Decreasing trend filamentous and colonial Cyanobacteria % biomass	% Nuisance cyanobacteria	As above	*Johnson Sb*
Decreasing trend filamentous diatoms (biomass)	Filamentous diatom	*Aulacoseira, Stephanodiscus*	*Log*
Decreasing trend filamentous diatom % biomass	%Filamentous diatom	As above	SHASH
Simplified Indicator			
Total filamentous algae	Total filamentous	*Aulacoseira, Dolichospermum,, Mougeotia, Ulothrix*	*SHASH*
Colonial cyanobacteria	Colonial cyano	*Microcystis, Coelosphaerium, Gomphosphaeria, Woronichinia*	*Log*
Filamentous cyanobacteria / Colonial cyanobacteria	Cyanobacteria ratio	*Dolichospermum, Aphanizomenon to Microcystis, Coelosphaerium*	*Log*
Target 2. Increase in Taxa that provide ample food resource			
Indicator	**Name**	**Example taxa**	**Transformation**
Edible taxa to approximately 30% of the biomass	%Edible	*Diatoma, Synedra,Tabellaria, Cryptomonas, Chlamydomonas*	*log*
Chrysophyceae >15% biomass	%Chrysophyceae	*Chromulina, Chrysochromulina, Dinobryon, Mallomonas, Synura*	*SHASH*
Simplified Indicator			
Chrysophyceae / Total Filamentous	Chrysophyceae ratio	As above	*Log*

Table 2.1.2. Reduced Potential Indicators based on correlations between indicators for upper bay stations only analysis. Strike-through for indicators removed from analysis. 8 remain of original 12.

Upper Bay only			
Potential indicators kept	**Removed indicators correlated at R^2>0.75**	**R^2 with kept indicator**	**Note**
Total Filamentous	Filamentous Diatom	0.89	
	Nuisance	0.95	
Chrysophyceae ratio	%Chrysophyceae	0.90	
~~Nuisance BM~~	Nuisance cyanobacteria	0.77	Removed cyano first
%Filamentous diatom		na	
%Nuisance		na	
Cyanobacteria Ratio		na	
%Edible		na	
Colonial Cyanobacteria		na	

Table 2.1.3. Reduced Potential Indicators based on correlations between indicators at all stations. Strike through for indicators removed from analysis. 5 remain from original 12.

All stations			
Potential indicators kept	**Removed indicators correlated at R^2>0.75**	**R^2 with kept indicator**	**Note**
Total Filamentous	Nuisance	0.97	
	Nuisance cyanobacteria	0.78	
	%Nuisance cyanobacteria	0.75	
	Filamentous diatom	0.92	
Chrysophyceae ratio	%Chrysophyceae	0.83	
~~Nuisance~~	%Nuisance	0.78	Removed %Nuisance first
	%Edible	-0.76	Removed %Edible first
Cyanobacteria ratio		na	
%Filamentous diatom		na	
Colonial cyanobacteria		na	

Table 2.1.4. Results of Principal Components Analysis from JMP 15.1 (on Correlations) given in Fig. 2.1.1

Upper Bay only					
Axis 1	**% variability**	**Eigenvalue**	**Factor**	**Eigenvector**	**% variance**
	42.2	3.38			
			Chrysophyceae ratio	-0.48	23.3
			Total Filamentous	0.46	21.5
			%Edible	-0.46	20.7
Axis 2	**% variability**	**Eigenvalue**	**Factor**	**Eigenvector**	**% variance**
	20.8	1.66			
			%Filamentous diatom	-0.71	47.8
			%Nuisance cyanobacteria	0.47	21.1
Axis 3	**% variability**	**Eigenvalue**	**Factor**	**Eigenvector**	**% variance**
	19	1.52			
			Colonial cyanobacteria	0.71	52.7
			Cyanobacteria ratio	-0.49	21.0
Upper and Lower Bay					
Axis 1	**% variability**	**Eigenvalue**	**Factor**	**Eigenvector**	**% variance**
	49.2	2.46			
			Total Filamentous	0.59	35.4
			Chrysophyceae ratio	-0.58	33.8
Axis 2	**% variability**	**Eigenvalue**	**Factor**	**Eigenvector**	**% variance**
	23.5	1.26			
			Cyanobacteria ratio	0.78	60.8
			Colonial cyanobacteria	-0.61	37.3

Table 2.1.5. Results of Stepwise Regression analysis from JMP 15.1 using proposed indicators as factors to explain total phytoplankton biomass.

Upper Bay only				
Overall model				
RMSE	**R^2**	**F**	**AICc**	**p**
775	.92	347	1283	<0.0001
	Factor	**F**	**p**	
	Total Filamentous	479	<0.0001	
	%Nuisance	27	<0.0001	
	Colonial cyanobacteria	23	<0.0001	
	Nuisance cyanobacteria	6	0.019	
Upper and Lower Bay				
RMSE	**R^2**	**F**	**AICc**	**p**
935	0.93	674	2632	<0.0001
	Factor	**F**	**p**	
	Total Filamentous	795	<0.0001	
	%Filamentous diatom	65	<0.0001	
	Chrysophyceae ratio	7	0.0105	

Table 2.1.6. Least squares means regression model output from JMP 15.1 for Chrysophycea ratio and Total filamentous using biological and environmental factor sources of variation.

Whole model testing Chrysophyceae ratio					
RMSE	**R^2**	**P**	**Source**	**Log worth**	**Effects test Prob >F**
0.0156	0.76	<.0001	% edible	2.51	0.003
			TKN	1.27	0.053
			bosmina ratio	0.62	ns
			year	0.52	ns
			total phyto	0.35	ns
			Surface Water Temperature	0.16	ns
			NO2+NO3	0.04	ns
0.0152	0.79	<.0001	total phyto	5.47	0.000
			TP	3.30	0.001
			Epar	2.07	0.009
			station	0.41	ns
			Annual_MeanMin_Temp	0.37	ns
			DIC	0.20	ns
			%DGM	0.13	ns
			Annual_Precipitation	0.04	ns
0.0192	0.61	<.0001	TKN	5.12	0.000
			Chydorus	0.61	ns
			Jan-Feb_MeanMin_Temp	0.47	ns
			Annual_Total_Precip	0.25	ns
			station	0.21	ns
			Annual_MeanMax_Temp	0.19	ns
			Jan-Apr_Total_Precip	0.17	ns
0.0213	0.51	<.0001	TP	3.39	0.000
			Jan-Apr Precipitation	0.81	ns
			bosmina ratio	0.68	ns
			%Meso	0.47	ns
			NO2+NO3	0.35	ns
			Annual_MeanMax_Temp	0.07	ns
0.0224	0.54	0.0019	TP	2.66	0.002
			Cyano ratio	1.21	ns
			%DGM	1.04	ns
			%Meso	0.98	ns
			DIC	0.66	ns
			Epar	0.38	ns
			Annual_MeanMax_Temp	0.20	ns

Whole model testing Total filamentous

RMSE	R^2	P	Source	log worth	Effects test Prob >F
1423.3	0.8	<.0001	Epar	2.61	0.002
			TKN	2.23	0.006
			Jan-Apr Precip	0.92	ns
			TP	0.23	ns
			turbid or clear	0.12	ns
			BM Chydorus	0.02	ns
1430.9	0.8	<.0001	year	3.47	0.000
			NO2+NO3	1.91	0.012
			Winter Snow	1.57	0.027
			TKN	1.11	ns
			bosmina ratio	1.06	ns
			Annual_MeanMax_Temp	0.43	ns
1425.6	0.41	0.004	DIC	1.82	0.015
			TP	1.61	0.025
			Annual MeanMin Temp	0.27	ns
			Surface_Water_Temperature	0.20	ns
			%DGM	0.00	ns
1618.8	0.7	<.0001	TKN	1.73	0.019
			turbid or clear	1.33	0.047
			TP	0.73	ns
			%Meso	0.66	ns
			Surface_Water_Temperature	0.40	ns
			Annual_Total_Precip[mm]	0.25	ns
1537.5	0.75	<.0001	TKN	4.72	<.0001
			Jan-Apr Precipitation	1.55	0.028
			turbid or clear	1.41	0.039
			TP	0.65	ns
			Winter Snow	0.60	ns
			Annual_MeanMax_Temp	0.23	ns
920	0.73	<.0001	August total filamentous	4.08	<.0001
			TKN	3.15	0.001
			DOC	0.45	ns
			May-Aug_Mean_Temp	0.43	ns
			TP	0.26	ns

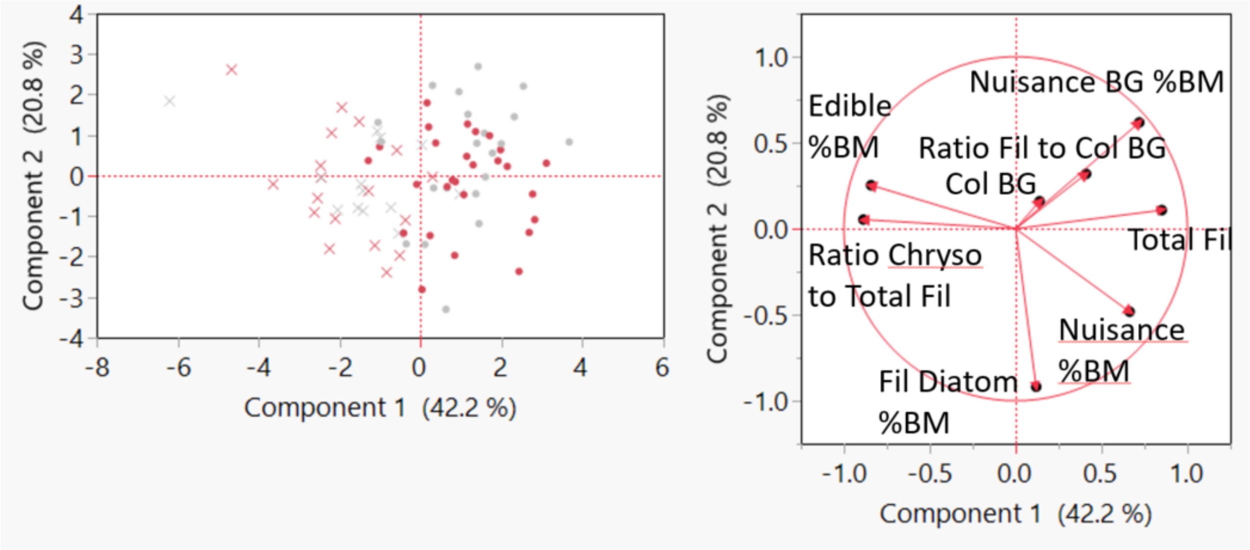


Figure 2.1.1 *Principle Components analysis of potential phytoplankton indicators for stations B (red) and N (grey) showing the loading and score plots for axis 1 and 2.*

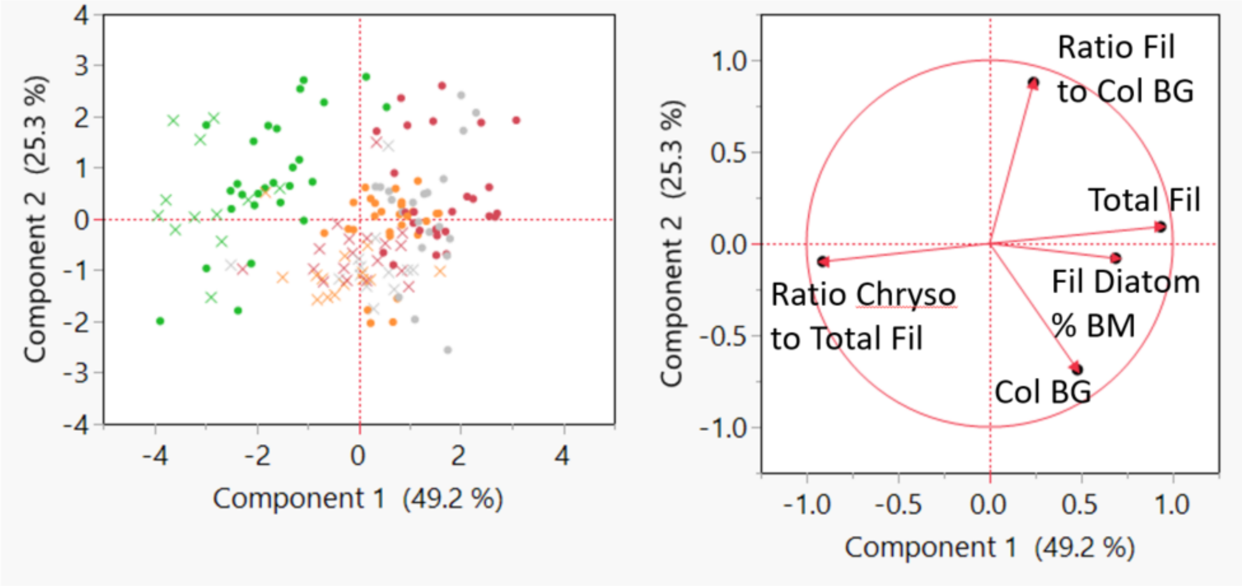


Figure 2.1.2 *Principle Components analysis of potential phytoplankton indicators for stations B (red), N (grey), HB (orange) and C (green) showing the loading and score plots for axis 1 and 2.*

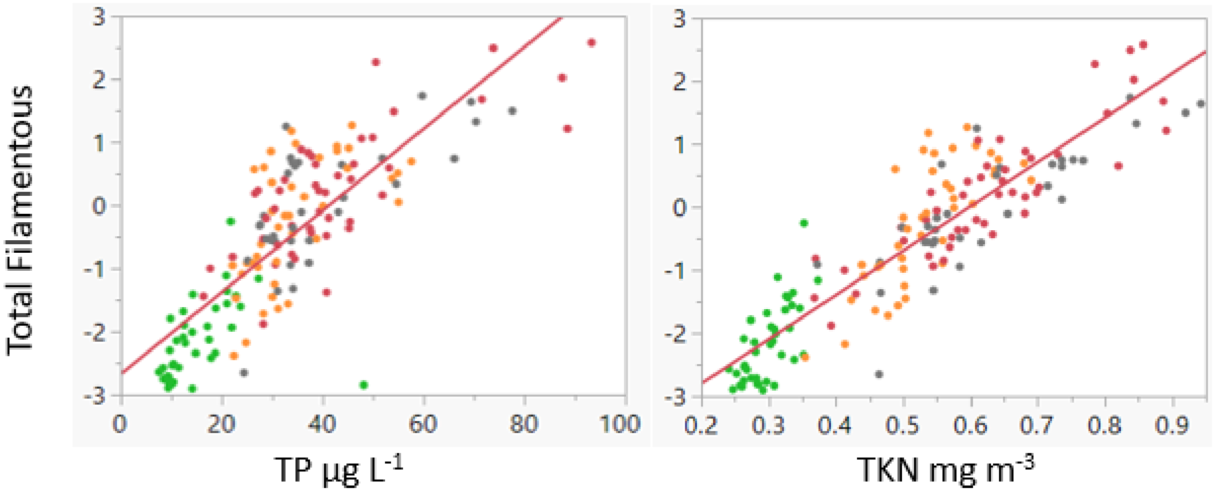


Figure 2.1.3 *Bivariate plots of Total filamentous (SHASH transformed) by Total Phosphorus and Total Kjeldahl Nitrogen (untransformed). May to October average data for B (red), N (grey), HB (grey) and C (green) stations.*

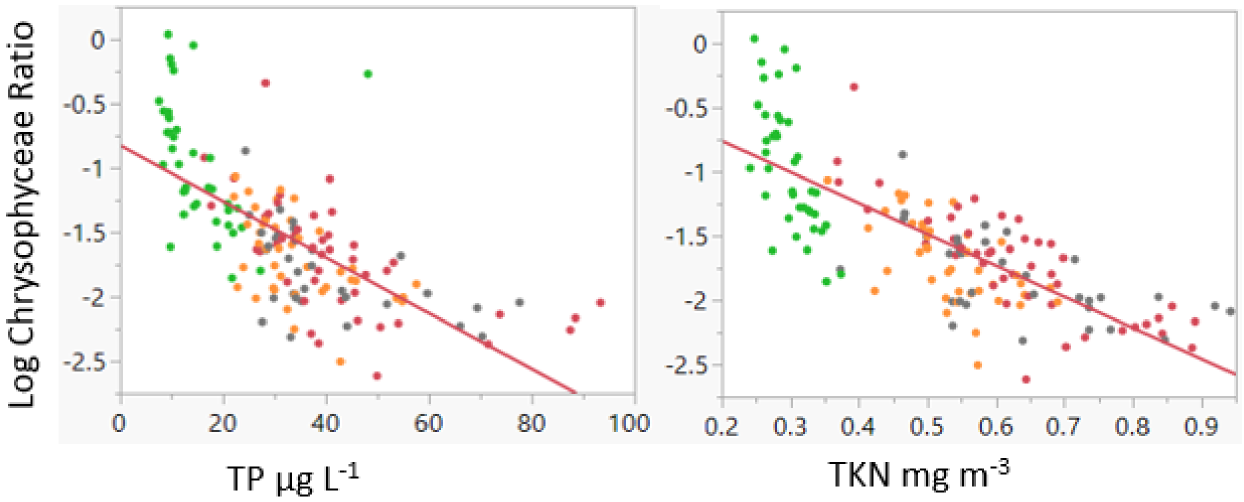


Figure 2.1.4 *Log Chrysophyceae ratio by Total Phosphorus and Total Kjeldahl Nitrogen. May to October average data from Station B (red), N (grey), HB (orange) and C (green).*

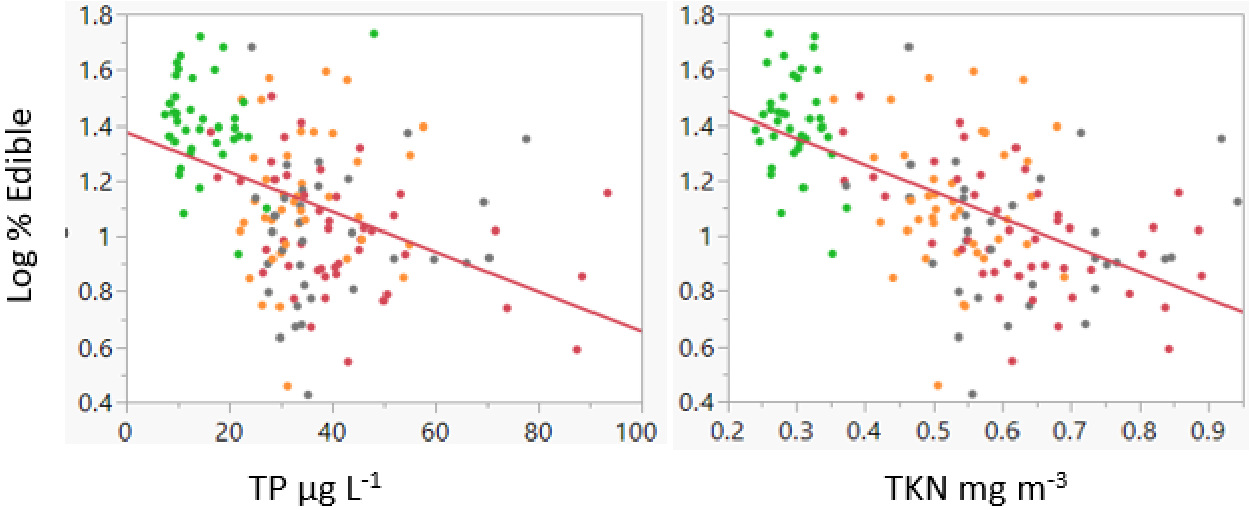


Figure 2.1.5 *Bivariate Fit of log edible % biomass with Total Phosphorus and Total Kjeldahl Nitrogen. May to October average data from Station B (red), N (grey), HB (orange) and C (green)*

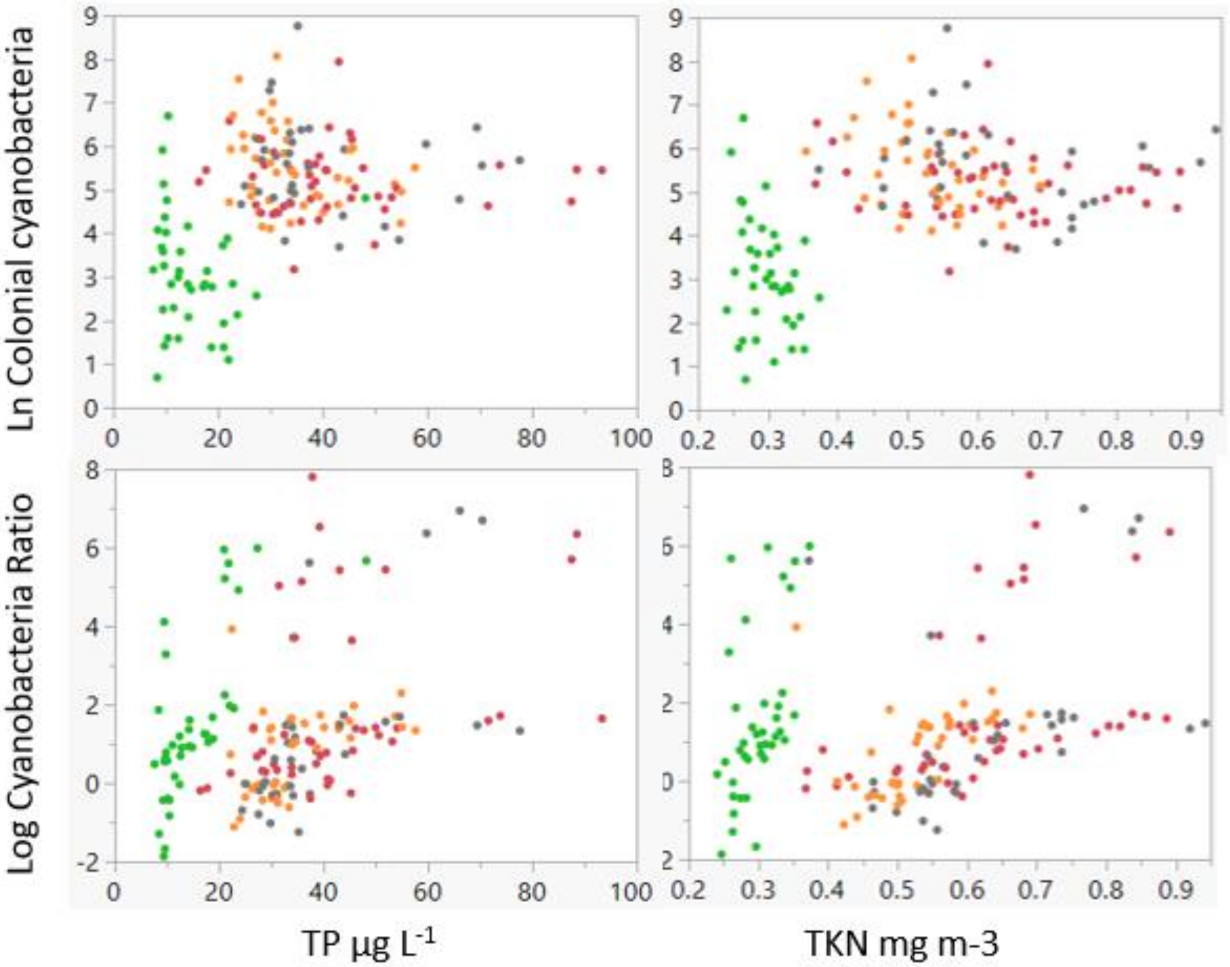


Figure 2.1.6 *Bivariate Fit of ln colonial cyanobacteria biomass (top) and log ratio of filamentous to colonial cyanobacteria biomass (bottom) with Total Phosphorus (left) and total Kjeldahl Nitrogen (right). May to October average data from Station B (red), N (grey), HB (orange) and C (green).*

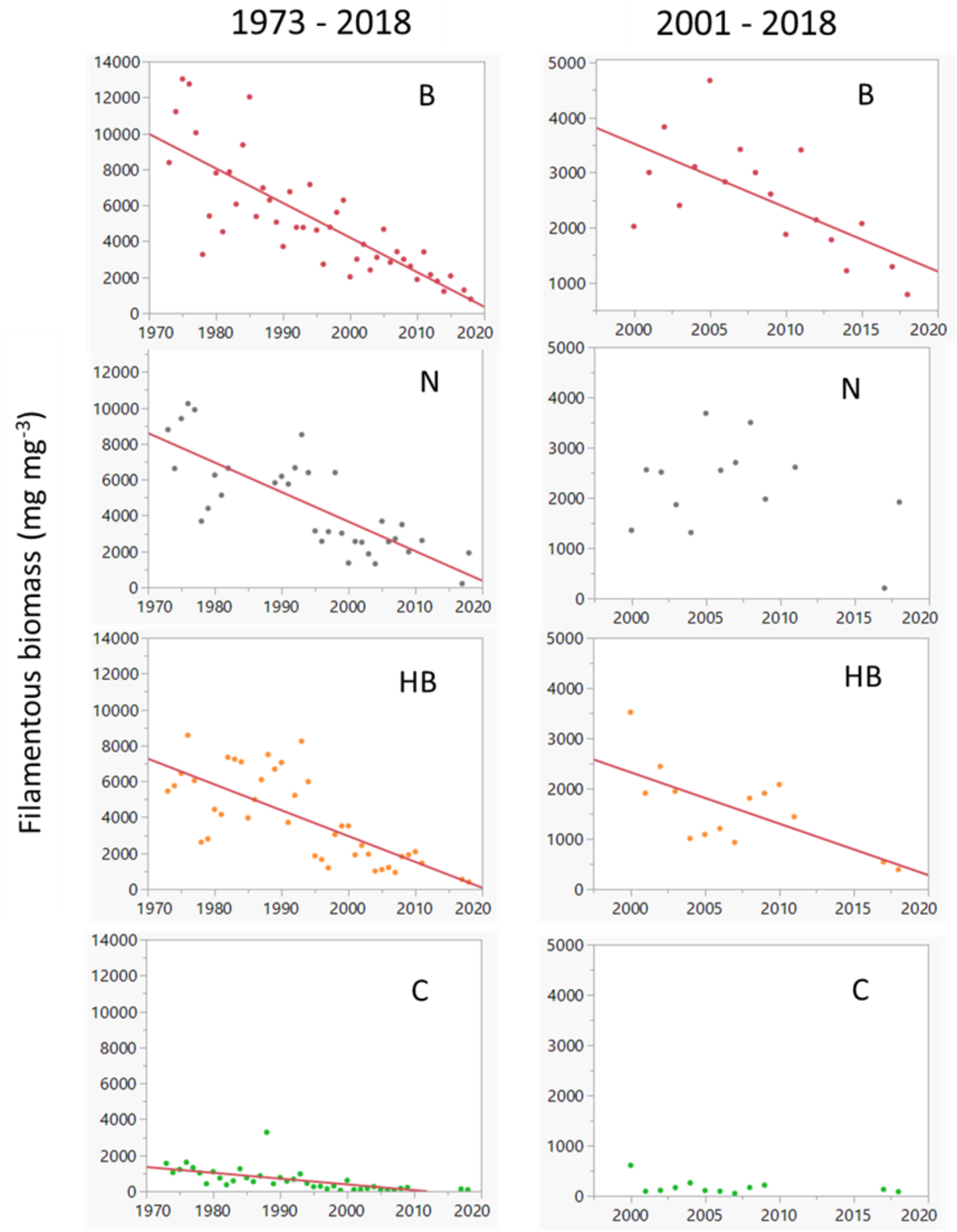


Figure 2.1.7 *Time series plot of Total filamentous biomass (mg m^{-3} untransformed; May to October average) g m^{-3} by station B (red), N (grey), HB (orange) and C (green). Left panels are 1973-2018 while the right panels show data from 2001 to 2018. Significant fit lines shown.*

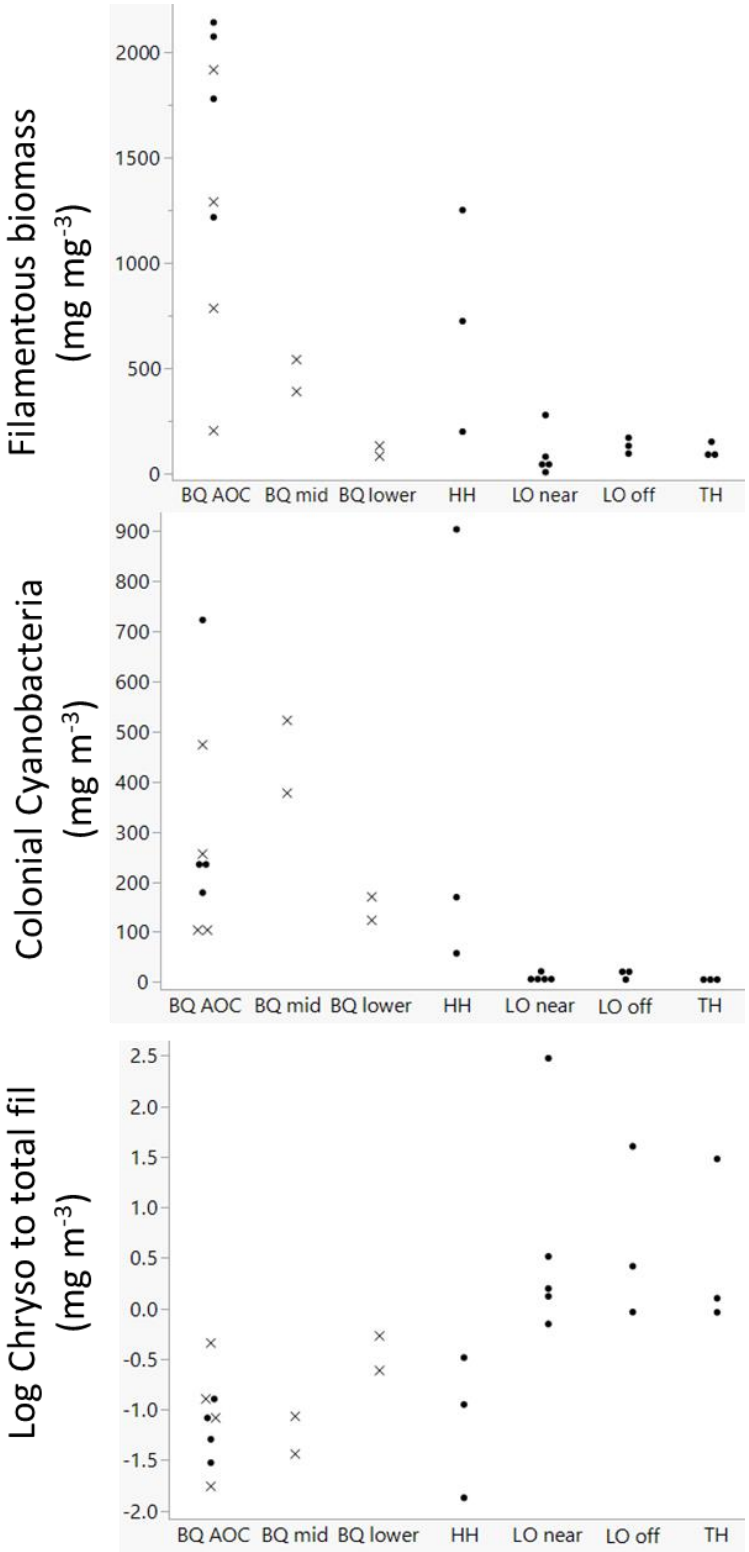


Figure 2.1.8 *Comparison of proposed indicators to regions across Lake Ontario. May to October average with 2017 and 2018 data indicated by X. Years are BQAOC 2012-2018, BQ 17-18, HH 12,14,16, LO2 near and off 13,18, TH 2016.*

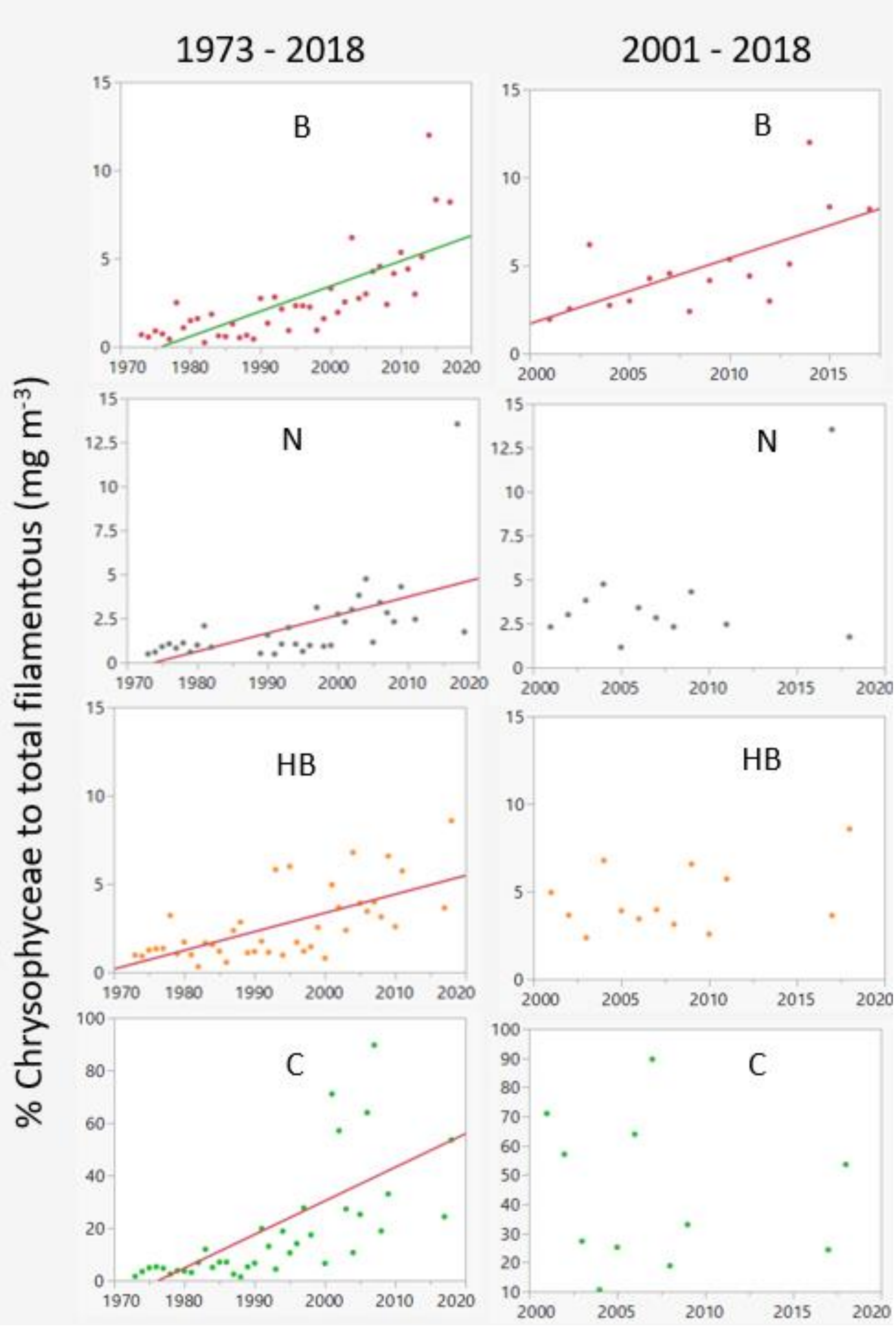


Figure 2.1.9 *Time series plot of % Chrysophyceae to total filamentous biomass (untransformed May to October average) mg m^{-3} by station B (red), N (grey), HB (orange) and C (green). Left panels are full data set 1972-2018 while the right panels show data from 2001 to 2018. Significant fit lines shown.*

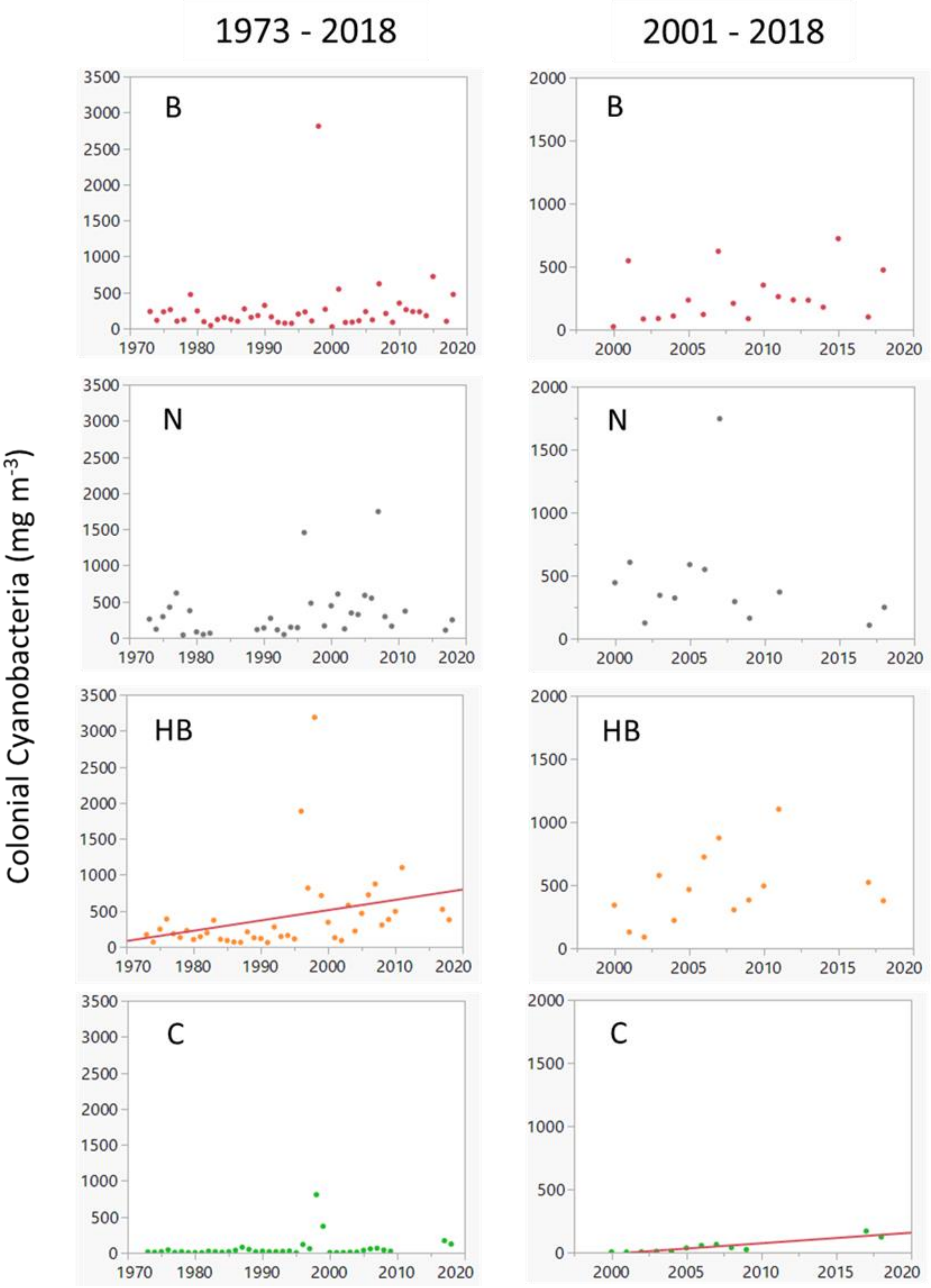


Figure 2.1.10 *Time series plot of Colonial cyanobacteria (untransformed May to October average) mg m-3 by station B (red), N (grey), HB (orange) and C (green). Top panels are full data set 1972-2018 while the bottom panels show data from 2001 to 2018. Significant fit lines shown.*

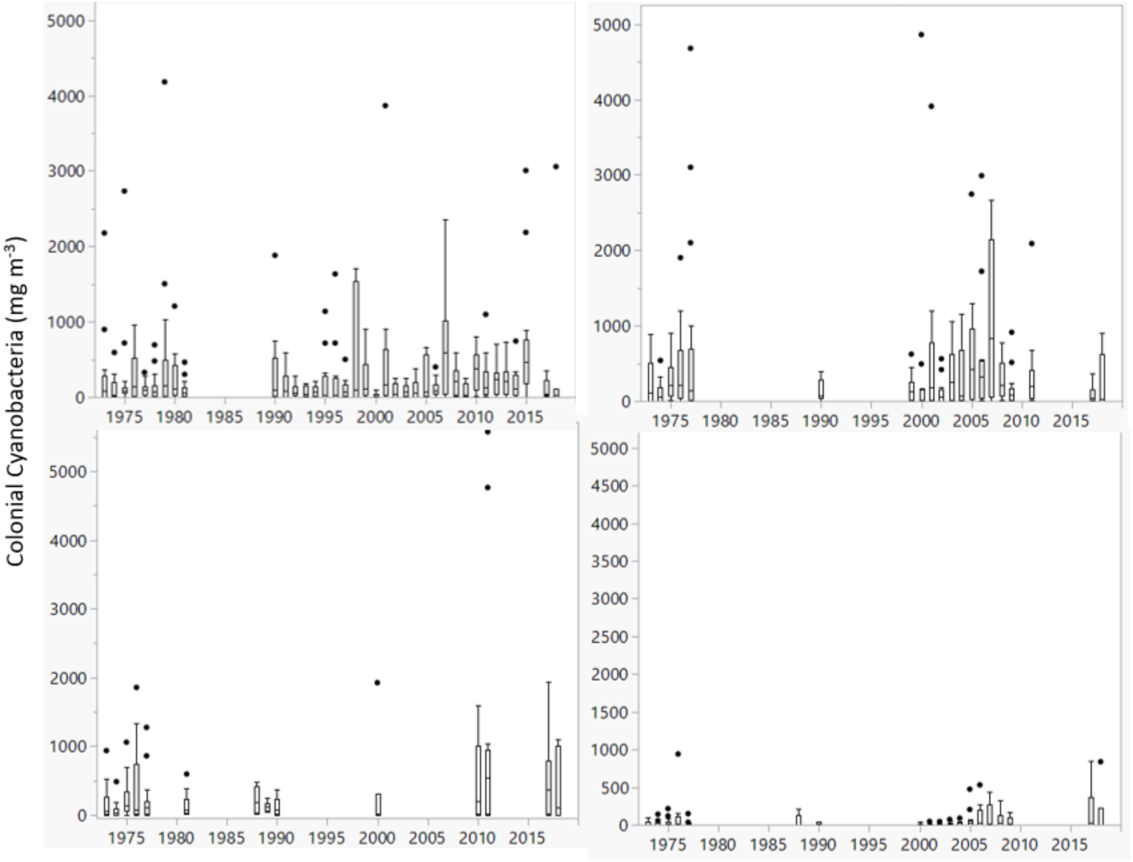


Figure 2.1.11 *Colonial cyanobacteria biomass measures during each year of sampling when individual sampling dates were counted. Outliers are shown as points.*

S2.2 Microbial Loop Indicators

The microbial loop includes bacteria, autotrophic picoplankton (APP), heterotrophic nanoflagellates (HNF) and ciliates. These micro-organisms interact with phytoplankton and zooplankton in complex ways to affect the transfer of energy from lower to higher trophic levels. In eutrophic environments, there is an expectation of a significant flux of energy being shunted through heterotrophs (bacteria, HNF, ciliates) that would be of similar magnitude to the energy generated by phytoplankton (Dodds and Cole 2007). Consequently, the microbial loop, is likely to have important implications for BUI 13: Degradation of phytoplankton and zooplankton populations.

Assessment of the microbial loop was added to Project Quinte beginning in 2000 to complement the existing monitoring of phytoplankton and zooplankton. In previously published work, we showed that in the Bay of Quinte, HNF contributed more biomass to the planktonic food web than zooplankton and, in a few cases, more biomass than the combination of phytoplankton and zooplankton (Munawar et al. 2011). The same paper hypothesized that HNF were utilizing food resources (bacteria and APP) that would otherwise have been available to zooplankton (*Daphnia*) and suppressing *Daphnia* biomass. These findings provided an example of why the microbial loop is important to the assessment of BUI 13 and why a ‘food web approach’ is needed; additionally reviews of the orginal BUI 13 assessment (Currie et al. 2017b) recommended inclusion of microbial communities into the assessment.

In general, a high proportion of heterotrophs (bacteria, HNF, ciliates, zooplankton) biomass to autotrophs (phytoplankton) is typically associated with anthropogenic stress although it could also be associated with tightly coupled (i.e. efficient) food webs in systems that are less disturbed (McCauley et al. 2018). Having said that, our experience in the Lake Ontario AOCs (including Hamilton Harbour and Toronto Harbour as well as the Bay of Quinte) suggests the former: that high proportions of heterotrophs, especially bacteria and HNF, are a sign of disturbance (Munawar and Fitzpatrick 2017; Munawar et al. 2018; Currie et al. 2018a,b). In eutrophic systems like the Bay of Quinte, even algal blooms can contain a high proportion of heterotrophic microbial biomass in addition to excess phytoplankton, which may not be available to higher trophic levels.

Given the importance of the microbial loop to phytoplankton and zooplankton interactions, and BUI 13, we will assess potential microbial indicators to complement the phytoplankton and zooplankton indicators proposed for the continued monitoring of BUI 13.

Methods

All analyses were run in JMP v15.1. A two-step approach was used to identify a potential microbial indicator. First, a Principle Components Analysis (PCA) that included the microbial loop (bacteria, APP, HNF, ciliates) along with selected phytoplankton indicators (*total filamentous phytoplankton*, *Ratio of Chrysophyceae to total filamentous phytoplankton*) and zooplankton indicators (*Log Chydorus biomass*, *Ratio of Eubosmina to Bosmina*, *% Daphnia of total crustacean biomass*, and *% Mesocyclops of adult cyclopoids*) was run to identify which microbial component would be most likely to complement the other planktonic indicators in predicting ecosystem stress.

Second, Stepwise Linear Regressions were run using the microbial loop indicator identified above as the exploratory variable (y axis) and various combinations of phytoplankton indicators, zooplankton indicators and physical-chemical parameters as effects (x axis). A multivariate correlation analysis was run that included all potential factors in order to avoid combinations which are significantly correlated, and combinations were monitored for collinearity using VIF (Variance Inflation Factors). Regressions of each individual pair of x and y variables (untransformed and log transformed) to test for curvature; log transformed values that improved linear fit (i.e. higher r^2) were included in the stepwise regression, otherwise, the untransformed value was used. The complete list of effect variables is given in Table 2.3.1. A total of 6 models were run with 11 – 12 parameters out of a possible 20 per model. The aim was to avoid combinations of parameters that were highly correlated to each other (e.g. total phosphorus and total Kjeldahl nitrogen) without restricting our ability to tease out potential effects. Each model generated was restricted to 4 terms.

Results and Discussion

Principle Components Analysis

Principle Components Analysis (PCA) showed that, of the microbial loop, bacteria had consistently strong correlations (both positive and negative) on the first three components compared to APP, HNF and ciliates (Fig. 2.3.1), Table 2.3.2). Eigenvalues, which represent the variance retained by each principal component were 3.13, 1.98 and 1.45 for components 1,2 and 3 respectively. Bacteria accounted 18.6%, 5.9% and 6.8% of the total explained variance for each component which was the highest of the microbial loop parameters. The remaining components had eigenvalues less than one and were not considered further. The results of the PCA suggest that bacteria is the primary microbial loop candidate as the metric for food web monitoring. This is expected given that bacteria are ubiquitous in aquatic environments and changes in bacterial dynamics can be related to a variety of ecosystem sources including sewage, excess sediment and nutrient loadings or by declines in grazer abundance and biomass (Cole 1982, Pomeroy and Weibe 1988, Biddanda et al. 2001, Kritzberg et al. 2005).

Stepwise Regressions

A total of 20 variables (11 – 12 per model) were tested for effects on bacteria by stepwise regression in different combinations which generated 6 models. The goal was to identify how bacteria affected or were affected by the proposed phytoplankton and zooplankton indicators while accounting for a variety of physical and chemical factors. The models generated are listed below with significant factors (P<0.05) in bold:

1. Bact = - 688.5975 + 5.4390439*__Chryso__ - 161.4623*Eubos:Bos - 0.125838*__HNF__ + 409.56769*Log[%Daphnia]

2. Bact = 175.11523 + 4.2118257*__Chryso__ + 17.117331*__%Meso__ - 253.8306*__Ebos:Bos__ - 16425.98*TP

3. Bact = -8872.563 + 0.690357*Chryso + 15.63966*__%Meso__ + 612.46717*__Surf_Temp__ - 6444.256*__TKN__

4. Bact = -688.5975 + 5.4390439*__Chryso__ - 161.4623*Eubos:Bos - 0.125838*__HNF__ + 409.56769*Log[%Daphnia]

5. Bact = 506.00957 + 5.7968986*__Chryso__ -180.1519* Eubos:Bos + 5.9971703*__Ciliates__ - 38.77922*__Plankt_Fish__

6. Bact = 466.36241 + 4.2354518*__Chryso__ + 17.310966*__%Meso__ - 243.9398*__Eubos:Bos__ - 1680.797*TKN

The complete set of results including parameters, F ratios, degrees of freedom and p values are summarized in Table 2.3.3 (a-f). The results show that bacteria has consistent relationships with *Chrysophyceae* (biomass), *Mesocyclops* (% of adult cyclopoids) and the *Ratio of Eubosmina to Bosmina* indicating linkages with both phytoplankton and zooplankton populations. Other models showed that physical and chemical drivers (surface temperature, TKN) are important predictors of bacteria and that higher trophic levels (planktivorous fish) are also related to bacterial biomass.

The importance of bacteria to the planktonic food web and BUI 13

Bacteria have an important role in aquatic food webs. As decomposers of organic matter, bacteria break down organic matter from both aquatic and terrestrial sources and provide energy to heterotrophic plankton. In oligotrophic systems where phytoplankton (primary) production is limited, bacteria can be an important accessory food resource for zooplankton which in turn helps sustain planktivorous fishes (e.g. Pace et al. 2004; McCauley et al. 2018). In eutrophic systems, like the Bay of Quinte, surplus organic matter results in excess bacterial production and / or the accumulation of large bacterial biomass. Whether or not bacteria represent a quality food resource for zooplankton is open for debate (e.g. Brett et al. 2009); previous studies of the Bay of Quinte and Hamilton Harbour suggested that excess bacterial production was mostly taken up and bound by HNF (Munawar et al. 2011; Munawar and Fitzpatrick 2017). The result was the large accumulation of microbial biomass that was similar in size and scope to algal (phytoplankton) blooms. As such, there is a considerable amount of bacterial derived energy generated in the Bay of Quinte each year, and where this energy goes has important implications for BUI 13.

Bacterial biomass is the microbial indicator that would improve the overall understanding of phytoplankton and zooplankton populations. The stepwise regression models consistently showed significant ($P<0.05$) relationships for bacteria with proposed phytoplankton and zooplankton indicators including: *Chrysophyceae biomass* (+), *% Mesocyclops of adult copepods* (+), and the *Ratio of Eubosmina to Bosmina* (-). One model also showed a negative correlation with planktivorous fish. Individually, the strongest correlations were observed between Bacteria and Chrysophyceae ($r^2=0.14$, $p=0.03$) and Bacteria and % Mesocyclops ($r^2=0.14$, $p=0.02$).

While these models provide some evidence of the linkages between bacteria and the planktonic food web, they also show how complex this relationship can be. For example, increases in Chrysophyceae biomass are associated with increases in Bacteria. This scenario happens when filamentous algae are also increasing due to elevated nutrient concentrations (Currie et al. 2017b). In a similar vein, a high proportion of Mesocyclops is an indicator of low levels of fish predation (planktivory) as well as eutrophy. In our models a high proportion of mesocyclops is also associated with high bacteria biomass. Taken together, what these results show is that bacteria increases while the other indicators point to deteriorating conditions. For that reason, bacteria provides a useful measure of the condition of phytoplankton and zooplankton populations.

Assessment of Bacteria trends in the Bay of Quinte planktonic food web

Long term changes in the microbial loop at Belleville including bacteria, APP, HNF and ciliates are shown in Fig. 2.2.2). Belleville is presented here as an example because the data set is complete. Phytoplankton and zooplankton trends are shown in Figs. 1.6 and 1.9 respectively in Section 1. Bacteria biomass ranged from ≈ 150 mg m^{-3} in 2000 to 1550 mg m^{-3} in 2018 with a peak of 2800 mg m^{-3} in 2014. Partition analysis reveals splits at 2009 and at 2014 (Fig. 2.2.3a). Prior to 2009, bacteria averaged 234.4 ± 35.0 mg m^{-3} compared to 919.5 ± 126.3 mg m^{-3} after 2009. Likewise, bacteria increased from 479.0 ± 102.6 mg m^{-3} (2000 – 2013) to 1985.8 ± 319.2 mg m^{-3} (2014-2020). HNF are important grazers of Bacteria and partition analysis of HNF biomass (Fig 2.2.3b) indicates a decline after 2006, from 4795.6 ± 921.7 mg m^{-3} to 1017.2 ± 223.6 mg m^{-3}, and an increase after 2014 from 868.0 ± 352.9 to 1256.0 ± 134.0 mg m^{-3}. The highest amounts of bacteria were observed from 2014 – 2018 (1393.6 – 2799.9 mg m^3) which also happens to correspond to reduced (though not the lowest) amounts of HNF (845.1 – 1556.7 mg m^{-3}), zooplankton (57.9 – 144.6 mg m^{-3}) and planktivorous fish (5.5 – 21.8 CPUE wt; Fig. 1.9).

There are many possible explanations for this scenario. One is that reduced grazing on bacteria by HNF and zooplankton has led to the accumulation of larger standing stocks of bacteria (i.e. a reduced “top-down effect”). Another is a build-up of bacterial biomass driven by physical events (increased runoff and loading from tributaries, sewage sources, sediment resuspension) that is essentially unpalatable to the planktonic grazers (i.e. an environmental or “bottom-up” effect); that in turn could limit the food resources available to planktivorous fish. It is likely that a combination of these top-down and bottom-up effects are occurring. Though food web interactions are complex, it is clear changes in bacteria are indicative of changes in the plankton and fish communities. As such, bacterial biomass is a useful indicator for the status of phytoplankton and zooplankton populations (BUI 13) and could also be useful in informing the status of fish populations (BUI 3). Furthermore, it is worth noting that there is considerable body of research linking bacterial activity to the formation / cessation of toxic (microcystin-producing) algal blooms (e.g. Wilhelm et al. 2011; Lezcano et al. 2017; Su et al. 2017; Bell et al. 2018) in eutrophic freshwater environments, which makes the monitoring of bacteria directly relevant to BUI 8: eutrophication or undesirable algae as well.

Shunting productivity into excessive bacterial biomass is not a desired outcome for an aquatic food web. As such we propose a target based on the 2001 – 2018 average at Belleville of bacteria biomass < 920 mg m^{-3}. While meeting that target in and of itself will probably not eliminate the impairment, it is likely to be accompanied by improvements in the phytoplankton and zooplankton populations which should, in turn, improve the food resources available to higher trophic levels.

Summary and Recommendations

Bacteria regenerate large amounts of energy in aquatic systems and can be an important food resource for zooplankton and benthos which in turn supports fish populations. However, accumulations of large standing stocks of bacteria, common in eutrophic environments, can have negative implications for food web dynamics (i.e. plankton supported fisheries) and extend into public health. We therefore recommend that any future monitoring of phytoplankton and zooplankton populations associated with BUI 13 include measurements of bacterial biomass.

Measurements of bacteria alone are relatively simple to undertake and could be done in conjunction with standard monitoring by public health units and sewage treatment plants. That being said, such measurements provide only a crude indicator of ecosystem health. A more in depth analysis of bacteria populations using molecular source tracking (e.g. Staley et al. 2018) could help pin point the sources of bacteria in the bay such as agricultural run-off, sewage, resuspension and provide guidance on where to focus remediation efforts.

Table 2.2.1 *Complete List of Parameters tested for effects against bacteria using stepwise linear regression models*

Parameter	Description
Bacteria	Bacteria biomass, exploratory variable
Phytoplankton Indicators	
Log [Total Fil]	Total filamentous algae (log transformed)
Chryso	Chrysophyceae biomass
Chryso:Tot Fil	Ratio of Chrysophyceae to Total filamentous algae
Zooplankton Indicators	
%Meso	% *Mesocyclops* of total adult Cyclopoids by biomass
Eubos : Bos	Ratio of *Eubosmina* to *Bosmina* by biomass
Log [%*Daphnia*]	%Daphnia of total crustacean biomass (log transformed)
Log [*Mesocyclops*]	*Mesocyclops* biomass (log transformed)
Log [*Chydorus*]	*Chydorus* biomass (log transformed)
Log [Rotifers]	Rotifer biomass (log transformed)
Fish_Plank	Planktivorous Fish biomass
Microbial Loop	
Ciliates	Total ciliate biomass
HNF	Heterotrophic nanoflagellate biomass
Other variables	
Surf Temp	Surface water temperature
Jan-Apr Precip	Total Precipitation January to April
Trent Discharge (May-Sep)	Trent River Discharge May to Sept
TKN	Total Kjeldahl Nitrogen
TP combined	Total Phosphorus
Log [May-Aug Precip]	May to August Precipitation (log transformed)
Log [Ann Precip]	Annual Precipitation (log transformed)

Table 2.2.2 *Formatted Loading Matrix for the Principle Components Analysis of planktonic and microbial loop parameters in the Bay of Quinte (2000-2018). Legend is in accordance with Table 2.2.1.*

Factor	**Prin1**	**Prin2**	**Prin3**	**Prin4**	**Prin5**	**Prin6**	**Prin7**	**Prin8**	**Prin9**	**Prin10**
Bacteria	0.76	-0.34	0.31	0.20	0.23	-0.05	0.00	0.14	-0.17	0.24
Log[Chydorus]	0.72	0.22	-0.14	-0.40	0.12	-0.22	-0.28	0.31	0.08	-0.09
APP	0.70	-0.24	0.24	0.44	0.22	0.20	-0.15	-0.13	0.22	-0.15
Ciliates	0.65	-0.25	-0.24	-0.20	-0.27	0.53	0.09	0.08	-0.19	-0.05
%Meso	0.60	0.39	0.37	-0.15	0.10	-0.15	0.54	-0.05	-0.01	-0.09
Tot Fil	0.33	0.64	-0.56	0.12	0.01	0.17	0.13	0.02	0.29	0.16
HNF	-0.49	0.62	0.02	0.39	0.23	0.18	0.02	0.29	-0.20	-0.08
Eubos : Bos	-0.09	0.61	0.50	-0.39	0.21	0.29	-0.22	-0.21	-0.04	0.07
%*Daphnia*	0.26	0.45	0.53	0.23	-0.60	-0.07	-0.13	0.07	0.04	0.02
Chryso : Tot Fil	-0.57	-0.42	0.48	-0.18	0.02	0.22	0.15	0.28	0.29	0.03

Table 2.2.3 *List of parameters tested for effects against bacteria for a series of models for best fit results from a Stepwise Linear Regression in JMP 15.1. Significant effects are* ***bolded****.*

a) Parameters Tested
Chryso
Chryso : Tot Fil
%Mesocyclops
Eubos : Bos
Ciliates
HNF
Log [% *Daphnia*]
Log [*Chydorus*]
Log [Rotifers]
Log [Ann Precip]
Surf. Temp.

Stepwise Best Fit Model 1

F(4,28)=5.2039, P=0.0029

Term	Estimate	SE	t Ratio	P>|t|	VIF
Intercept	-688.59	821.56	-0.84	0.41	.
Chrysophyceae	**5.44**	**1.64**	**3.32**	**0.0025**	**1.11**
Eubos : Bos	-161.46	96.75	-1.67	0.1063	1.36
HNF	**-0.12**	**0.05**	**-2.42**	**0.0224**	**1.12**
Log [% *Daphnia*]	409.57	245.09	1.67	0.1059	1.11

b) Parameters Tested
Chryso
Chryso : Tot Fil
%Meso
Eubos : Bos
Ciliates
HNF
Log[%*Daphnia*]
Log[*Chydorus*]
Log[Rotifers]
Log[AnnPrecip]
Surf Temp
TP

Stepwise Best Fit Model 2

F(4,28)=4.6351, P=0.0054

Term	Estimate	SE	t Ratio	P>|t|	VIF
Intercept	175.11	314.07	0.56	0.5816	.
Chryso	**4.21**	**1.86**	**2.26**	**0.0318**	1.37
%Meso	**17.12**	**6.56**	**2.61**	**0.0144**	1.52
Eubos : Bos	**-253.83**	**93.25**	**-2.72**	**0.011**	1.21
TP	-16425.98	10149.48	-1.62	0.1168	1.25

c) Parameters Tested
Chryso
Chryso : Tot Fil
%Meso
Eubos : Bos
Ciliates
HNF
Log[%*Daphnia*]
Log[*Chydorus*]
Log[Rotifers]
Log[AnnPrecip]
Surf Temp
TKN

Stepwise Best Fit Model 3

F(4,28)=15.4557, P<0.0001

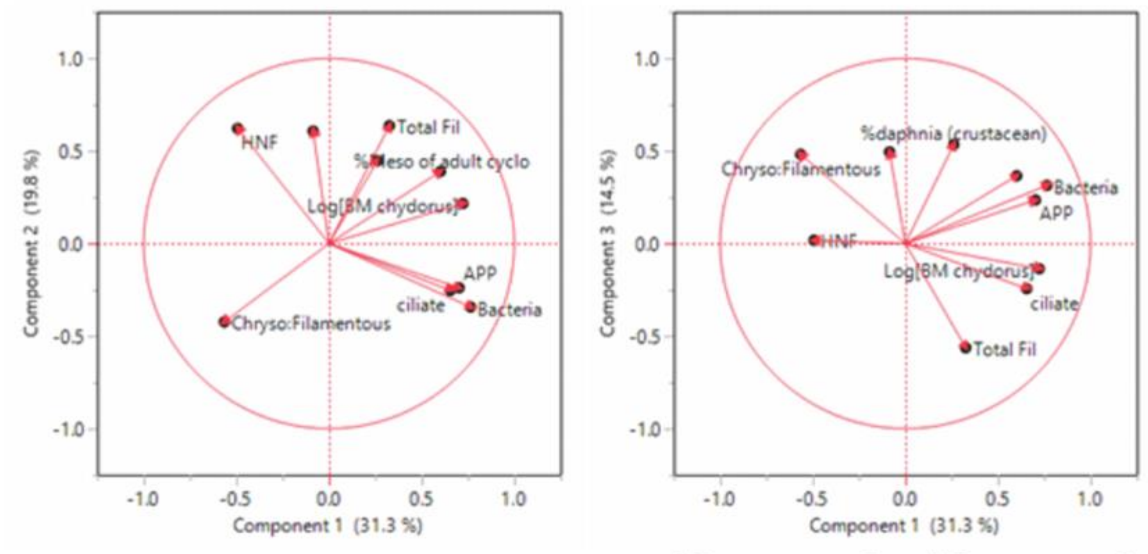
Term	Estimate	SE	t Ratio	P>|t|	VIF
Intercept	-8872.56	1540.37	-5.76	<.0001	.
Chryso	0.69	1.38	0.5	0.6218	1.46
%Meso	**15.64**	**4.55**	**3.44**	**0.0018**	**1.42**
Surf Temp	**612.47**	**99.52**	**6.15**	**<.0001**	**3.11**
TKN	**-6444.26**	**1027.36**	**-6.27**	**<.0001**	**3.02**

Table 2.2.3 *- continued*

d) Parameters Tested	**Stepwise Best Fit Model 4**					
Chryso %Meso Eubos : Bos Ciliates HNF Log[%*Daphnia*] Log[*Chydorus*] Log[Rotifers] Log[AnnPrecip] Surf Temp Log[Total Fil]	F(4,28)=5.2039, P=0.0029					
	Term	**Estimate**	**SE**	**t Ratio**	**P>|t|**	**VIF**
	Intercept	-688.60	821.56	-0.84	0.409	.
	Chryso	**5.44**	**1.64**	**3.32**	**0.0025**	**1.11**
	Eubos : Bos	-161.46	96.76	-1.67	0.1063	1.36
	HNF	**-0.12**	**0.05**	**-2.42**	**0.0224**	**1.12**
	Log[%daphnia]	409.57	245.09	1.67	0.1059	1.11

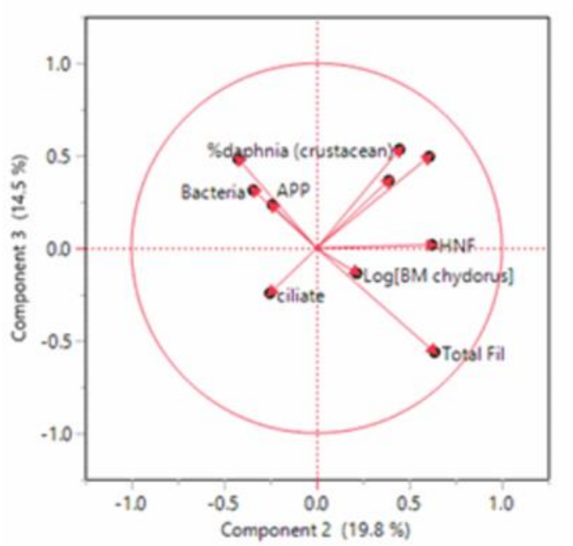
e) Parameters Tested	**Stepwise Best Fit Model 5**					
Chryso %Meso Eubos : Bos Ciliates HNF Log[%*Daphnia*] Log[*Chydorus*] Log[Rotifers] Log[AnnPrecip] Surf Temp Fish_Plank Chryso:Tot Fil	F(4,28)=6.3616, P=0.0011					
	Term	**Estimate**	**SE**	**t Ratio**	**P>|t|**	**VIF**
	Intercept	506.01	258.75	1.96	0.0618	.
	Chryso	**5.80**	**1.66**	**3.49**	**0.0018**	**1.17**
	Eubos : bos	-180.15	88.52	-2.04	0.0526	1.17
	Ciliates	**5.99**	**1.91**	**3.15**	**0.0042**	**1.40**
	Fish_Plank	**-38.78**	**13.43**	**-2.89**	**0.0079**	**1.43**

f) Parameters Tested	**Stepwise Best Fit Model 6**					
Chryso %Meso Eubos : Bos Ciliates HNF Log[%*Daphnia*] Log[*Chydorus*] Log[Rotifers] Log[AnnPrecip] Chryso:Tot Fil TKN Trent Discharge (May-Sep)	F(4,28)=4.9745, P=0.0037					
	Term	**Estimate**	**SE**	**t Ratio**	**P>|t|**	**VIF**
	Intercept	466.36	388.56	1.2	0.2401	.
	Chryso	**4.23**	**1.83**	**2.31**	**0.0283**	**1.36**
	%Meso	**17.31**	**6.42**	**2.7**	**0.0117**	**1.51**
	Eubos : bos	**-243.94**	**91.37**	**-2.67**	**0.0125**	**1.19**
	TKN	-1680.80	896.82	-1.87	0.0714	1.23



a) Component 1 and Component 2

b) Component 1 and Component 3



c) Component 2 and Component 3

Figure 2.2.1 *Bi-plots of the Principle Components Analysis of planktonic and microbial loop parameters in the Bay of Quinte (2000-2018) for a) Components 1 and 2, b) Components 1 and 3 and c) Components 2 and 3. “Bacteria” = bacteria biomass, “Log [BM Chydorus]” = log transformed Chydorus biomass, “APP” = autotrophic picoplankton biomass, “HNF” = heterotrophic nanoflagellates biomass, “ciliate” = Ciliate biomass, “%Meso of adult cyclo” = % Mesocyclops of adult cyclopoids, “Total Fil” = total filamentous phytoplankton, “ratio eubos to bos” = Ratio of Eubosmina to Bosmina, “%Daphnia (crustacean)” = % Daphnia of total crustacean biomass, “Chryso:Filamentous” = Ratio of Chrysophyceae to total filamentous phytoplankton.*

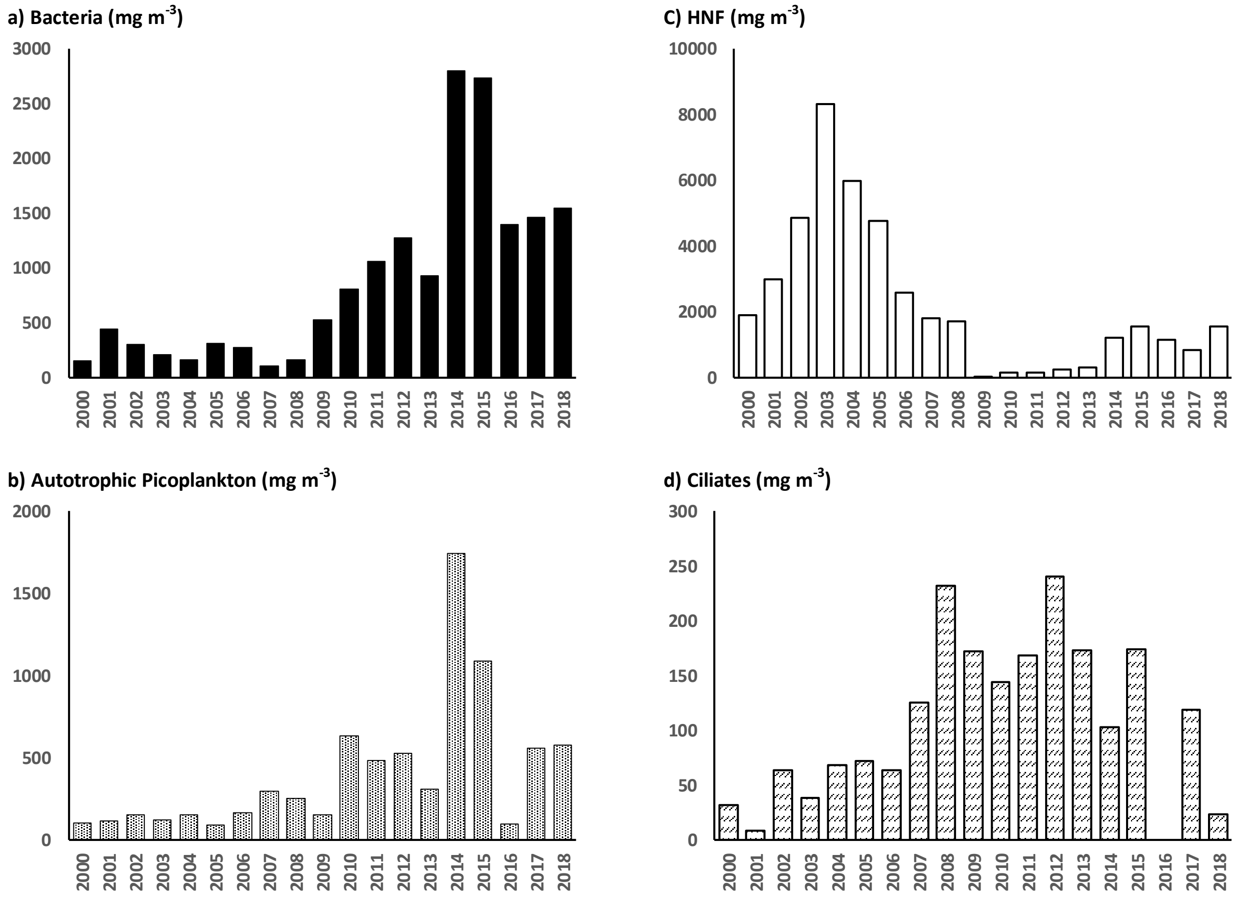


Figure 2.2.2 *Long Term trends in the annual mean biomass for each component of the Microbial Loop for the period 2000 – 2018, including a) Bacteria, b) Autotrophic Picoplankton (APP), c) Heterotrophic nanoflagellates, and d) Ciliates. Units are mg* m^{-3}.

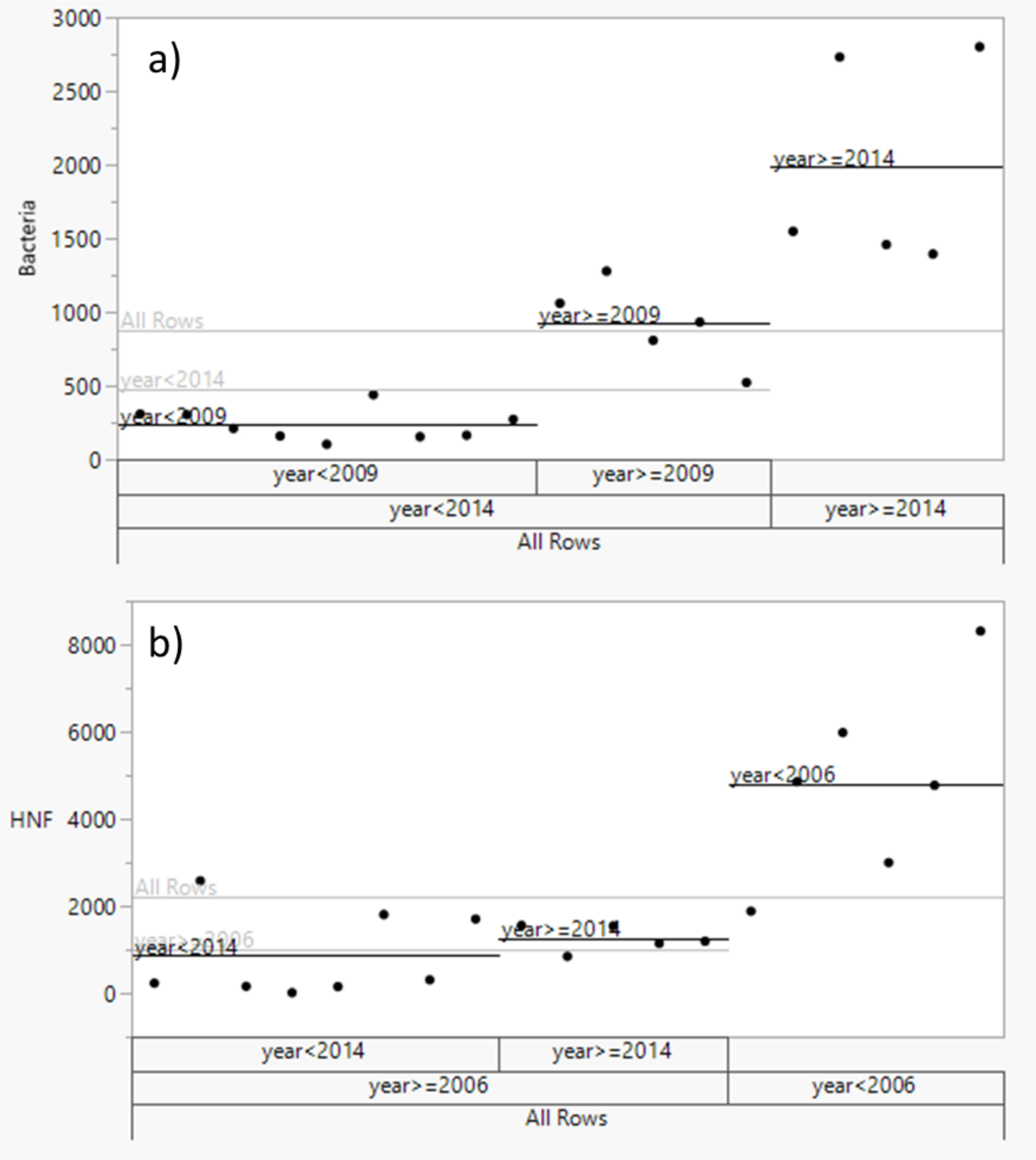
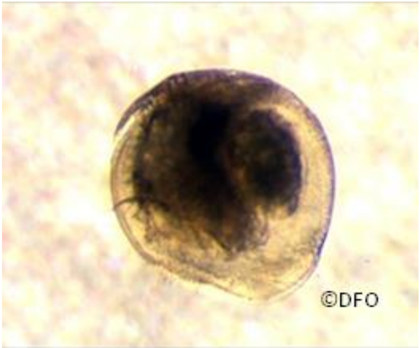


***Figure 2.2.3** a) Partition analysis of Bacteria biomass (mg m^{-3}) at station B (Belleville) over the period of 2000-2018, and b) partition analysis of HNF (heterotrophic nanoflagellates) biomass (mg m^{-3}) at station B (Belleville) over the period of 2000-2018. Analysis run in JMP 15.1.*

S2.3 Zooplankton Indicators

In the “Proposed Targets for Evaluation of the Bay of Quinte BUI 13” by Currie et al. (2017a), eight zooplankton metrics were outlined. In that document, increases in *Chydorus sphaericus* biomass, percent Acanthocyclops vernalis and the percentage of eutrophic rotifer taxa, and decreases in the Shannon diversity index and cladoceran mean size serve as indicators of increasing eutrophy (bottom-up or nutrient driven impacts), although all but the rotifer metric are also indicative of high fish planktivory (top-down impacts). Indicators of low fish planktivory include increased percentages of large *Daphnia* (>0.75 mm), *Daphnia galeata mendotae* and *Mesoyclops edax*. However, it was subsequently decided to reduce the number of metrics used, and some (the *Daphnia* metrics) were considered redundant. In an effort to simplify the zooplankton indicators and chose the best bottom-up and top-down metrics to be used in future monitoring plans, we reexamined data from the upper and middle bays (B, N and HB) over the 1975 to 2018 time frame. A few additional metrics such as *Eubosmina* biomass and the ratio of *Daphnia galeata mendotae* to *D. retrocurva* (*Daphnia* ratio) were also tested. Three sampling periods were also tested, including: 1) annual means (May to October), 2) warm months (monthly means from June to September), and 3) summer months (monthly means from July and August). As the indicators presented by Currie et al. (2017b) were based on annual data, the rationale behind evaluating the warm and summer periods was to determine whether the chosen metrics were still useful if the length of the sampling season was reduced in future monitoring plans.

Bottom-up Indicators



Box 2.3.1 *Chydorus sphaericus*

For all three periods, increasing log *Chydorus* biomass was the strongest and most consistent positive indicator of higher trophic status (bottom-up impacts) (Fig. 2.3.1a and 2.3.1b). A small herbivorous littoral cladoceran, *C. sphaericus* has been proposed as an indicator of eutrophic conditions since it often appears in the littoral zone when blue-green algae blooms are occurring (Gannon and Stemberger 1978; Gulati 1983; Pejler 1983; Haberman and Haldna 2014). Furthermore, *Chydorus* populations in the Bay of Quinte are insensitive to changes in fish planktivory, as it is not a preferred food item due to its small size (de Bernardi and Giussiani 1990; Jeppesen et al. 2011). Zooplankton populations where *Chydorus* is a dominant taxon may not transfer energy to planktivorous fishes as efficiently due to their small size. *Chydorus* is a useful metric for future monitoring efforts as it is relatively easy to identify and enumerate in zooplankton samples. The only caution is that *Chydorus* could be confused with similarly sized *Dreissena* veliger larvae, but the two taxa can be easily distinguished with cross-polarized light filters (Frischer et al. 2012). Another benefit is that this metric can also be used effectively even if only summer samples are enumerated. We expect *Chydorus* biomass in the Bay of Quinte to decrease over time as the system becomes less eutrophic. This downward trend was observed at B over the 1975 to 2000 period (Fig. 2.3.2a). The highest annual mean biomass values for *Chydorus* were seen at HB in the mid-1980s, when they ranged from 10.8 to 29.7 mg m^{-3} (Fig. 2.3.2b). Although populations have generally been lower in the more recent period, they have been variable from year to year and have not shown a consistent trend at either station. Seasonally, *Chydorus* populations in the Bay of Quinte tend to peak in August and September when algal blooms are often most developed, and monthly mean biomass of this taxon has sometimes exceeded 40 mg m^{-3}. Since 2001, annual *Chydorus* biomass has averaged 4.0 ± 0.9 mg m^{-3} at B and 6.6 ± 0.9 mg m^{-3} at HB and has usually been ≤ 6 mg m^{-3}. This equates to mean percent composition values of 3.9% and 4.8% relative to total crustacean biomass, respectively. <u>A proposed target for *Chydorus* biomass is an annual mean below 7.5 mg m^{-3} given by the mean + 1 standard deviation.</u> However, it is worth noting that the *Chydorus* biomass per unit TP at B is 118.7, significantly lower than 221.4 at HB since 2001 (F(1,33)=6.80, p=0.013). As such, almost twice as much *Chydorus* biomass is expected at B than is currently found there, which is one of the reasons that the zooplankton:phytoplankton (Z:P) ratio has not improved in the upper bay compared to middle bay (Section 2.4).

Another possible promising indicator for trophic status in the Bay of Quinte is the bosminid ratio, the biomass ratio of *Eubosmina coregoni* to *Bosmina longirostris* (Fig. 2.3.1c and d). It may serve as an alternate indicator in a more pelagic environment such as the lower Bay of Quinte where *Chydorus*, a littoral species, has never been abundant. Using annual means at B, the bosminid ratio averaged 8.9 ± 2.0 between 1975 and 1994 (excluding the unusually high value of 144 in 1979), and only 1.4 ± 0.4 between 2001 and 2018 (Fig. 2.3.2c). At HB, this ratio has never exceeded 4, and has averaged 0.7 ± 0.2 since 2001 (Fig. 2.3.2d). At C, the highest value (2.6) was found in 1990 and has averaged 1.2 ± 0.1 since 2001. *Eubosmina coregoni* is a small to medium-sized cladoceran that shows a strong positive correlation with increasing eutrophication in the Bay of Quinte using annual mean data, although this relationship is weaker when summer monthly means are used. *Bosmina* is a very tolerant, ubiquitous taxon that has not shown to be useful as a metric on its own in the Bay of Quinte, despite being used as an indicator of increasing eutrophy in other systems (e.g., Gannon and Stemberger 1978; Haberman and Haldna 2014). While some zooplankton can handle and consume filamentous forms of algae, it requires considerable effort and not all species are capable of doing so (Kiørboe 2011, Leitão et al. 2020). *Eubosmina* may be a more efficient grazer than *Bosmina* and is better able to consume larger food particles such as Cyanophyta filaments (Irvine 1986). This would explain the higher proportion of *Eubosmina* in the Bay of Quinte under more eutrophic conditions, when biomass of filamentous algae was greater. However, there has been some confusion surrounding the taxonomy and identification of this group in other systems over the years (e.g., Kotov et al. 2009), which has clouded the use of bosminid taxa as indicators in the scientific literature. For example, *Eubosmina coregoni* is sometimes referred to as *Bosmina coregoni* in older literature (e.g. Irvine 1986). For future monitoring efforts, we define *Eubosmina* as a bosminid lacking a tail (mucral) spine and *Bosmina* as one with a spine (Fig. 2.3.3). Given these difficulties, it may not be a preferred zooplankton indicator species for the Bay of Quinte.

Top-down Indicators

Top-down (fish planktivory) control of zooplankton is less obvious in the Bay of Quinte given the strong nutrient driven influences on zooplankton (Section 1, Fig 1.12). As control of planktivore biomass is largely dominated by trophic status, it is not independent of the identical nutrient-driven influences that also impact zooplankton. Top down effects are also more apparent under more extreme rates of planktivory as seen in the 1970s in the upper and middle bay prior to the listing of the AOC. Planktivory influences seem to be more obvious at the nearshore sites sampled in 2017 and 2018, though fish community composition in the nearshore zone is not well understood (see Bowen et al. 2020). Overall, the relationships between zooplankton indicators and planktivore biomass are stronger when annual mean data are used, compared to summer monthly data. This may be because zooplankton populations can fluctuate much more rapidly month to month, thus requiring more frequent sampling, whereas fish are sampled less frequently because they tend to be more stable across the season. There is an expectation of the ratio of planktivorous fish biomass to zooplankton biomass starting to decrease as total phosphorus improves in the upper bay (see Section 2.4). Currently any influence of top-down forcing is being masked by the dominance of the bottom-up nutrient driven eutrophication of the upper bay.

Similarly, although easy to measure, size metrics such as mean cladoceran length and percentage of large *Daphnia* individuals (relative to total *Daphnia* densities*)* are not sensitive enough indicators of planktivory in the upper and middle bays given the dominance of the bottom-up forcings, and thus appear to have limited utility. The percentage of *Daphnia* in the zooplankton is tied to the analysis of both phytoplankton and bacterial biomass in the Bay of Quinte (Sections 2.1 and 2.2). The biomass percentage of the large cladoceran species *Daphnia galeata mendotae* relative to total crustacean biomass (% DGM) is one of the best indicators of top down effects in the Bay of Quinte (Fig 2.3.1e and f) and is proposed as an indicator of planktivory in future monitoring plans. One drawback of the %DGM metric is that it can be difficult to distinguish *D. galeata mendotae* from *D. retrocurva*, especially with smaller individuals and insufficient magnification.

D. galeata mendotae is also considered by some as an indicator of increased oligotrophy (Pejler 1983) and larger *Daphnia* have been shown to a large influence on clear-phase food-web and climate change metrics in transitional ecosystems (Matsuzaki et al. 2020). Aside from lower nutrient concentrations, oligotrophic systems are also characterized by high water clarity and smaller algal cells (Ross and Munawar 1988). This promotes more efficient large grazers such as *Daphnia*, which usually prefer small phytoplankton and bacteria. An increase in percent *Daphnia* indicates that planktivory is declining, and to a lesser degree, eutrophy is decreasing. *D. galeata mendotae* is especially vulnerable to fish predation and seldom reaches high densities when planktivores are abundant (Brooks and Dodson 1965; Pothoven et al. 2013). However, it may be more resistant to invertebrate predators such as *Cercopagis* and *Bythotrephes* due to its larger size (Wahlstrom and Westman 1999; Yan and Pawson 1997), but these invasive predators are generally uncommon in the upper Bay of Quinte.

When planktivore biomass exceeded a threshold of around 50 CPUEwt (based on combined trawl and gillnet data for all sites combined), %DGM was rarely above 1%. During the first decade of Project Quinte, when this planktivore threshold was often surpassed, %DGM rarely exceeded 2.5% (Fig 2.3.2e and f). Although this metric has generally increased since this early period, it has fluctuated widely over the last 30 years and has shown no consistent time trend over that period. Between 2001 and 2018, %DGM has averaged 17.9 ± 3.2% at B with unusually low values in 2000, 2002 and 2017. During this more recent period, %DGM values less than 16% may indicate that the zooplankton community is being stressed by top down influences. When this value drops below a target level of 5%, zooplankton may be at risk of transferring insufficient energy to higher trophic levels. Low total zooplankton biomass in general may also reflect food shortages for planktivores. At B, four out of the last six years have had the lowest annual mean biomass levels out of the 42 year time series, corresponding to the lowest years of planktivorous fish and total fish biomass.

It is also interesting to note that over the duration of Project Quinte, peaks in *Daphnia* biomass have often shifted to slightly later in the season in the upper and middle bays. During the turbid phase (1972 – 1994) in the upper and middle bay, June and July *Daphnia* biomass values were similar, but the mid-1990s June values have been significantly lower indicating a delay in *Daphnia* population growth until mid-summer (Fig. 2.3.4). This loss in June biomass was more pronounced at HB than at B starting in 1996. It is unknown if this is due to increased visual predation during the clear phase early in the year by either migratory planktivores (e.g. Alewife) or by larval fishes while July *Daphnia* biomass has been unaffected. There have also been a few years at B where June biomass values have been negligible, including 2000 – 2003, 2013 and 2014. Furthermore, July *Daphnia* nearly disappeared at B during the last three years of study, when % DGM was ≤2%. However only 2017 was below the annual target of >5%DGM; in 2016 and 2018 DGM biomass was sufficient during the other months to raise the value above the target level. At HB, levels were low in July 2016 and 2017 but recovered in 2018. This loss of early summer *Daphnia*, considered to be a preferred “fish food”, may have repercussions for the survival of larval fishes emerging in July and August over the last few years. Finally, very low values of %DGM, and *Daphnia* in general, can result from poor environmental conditions, including an overabundance of filamentous algae that can interfere with their ability to feed effectively (Sahuquillo et al. 2007).

If there is a decision to count *Eubosmina* for monitoring, a useful amalgamated food web metric is the normalized *Eubosmina* to DGM biomass ratio [Eubos:(DGM+Eubosm)]. This metric combines the bottom-up driven *Eubosmina* (which dominates at high TP) with the top-down influenced DGM which prefers more mesotrophic conditions. As such the Eubos:DGM value is high during eutrophic conditions when *Eubosmina* is common and DGM is absent, but becomes less as DGM increases in biomass as conditions become less eutrophic (Fig. 2.3.5). Using the mean value + 1 sd as a target, a value of Eubos:DGM < 0.52 should be maintained. All of the values up to the late 1980s were in the range at both stations, but only 14% of years at HB (avg. 0.41) and 57% of years at B (avg 0.54) since 1995 are above this value (Fig. 2.3.5). This ratio will improve in the upper bay as bottom up disruptions from excess phosphorus are aleviated.

Mesocyclops biomass as a percentage of adult cyclopoid biomass (%*Mesocyclops*) is another potentially useful indicator of fish planktivory (Fig. 2.3.1g and h). Although the proportion of this large predatory cyclopoid copepod is variable under lower rates of planktivory, it is usually suppressed when planktivory is high (Chang et al. 2004; Brooks and Dodson 1965). It also appears to be more effective as an indicator when annual data are used. In the 1970s at B and HB when planktivory was particularly high, %*Mesocyclops* averaged 22.7 ± 7.1%. During the 2001 to 2018 period, the mean for %*Mesocyclops* was 57.5 ± 3.7% at B and N and it has not dropped below 20% (Fig 2.3.2g). Levels below 40% are indicative that planktivory rates may be unusually high and that the food web is out of balance; however this indicator should be revisited once top-down effects become stronger. *Mesocyclops* has the advantage over *D. galeata mendotae* in that it is easier to identify in the zooplankton samples.

Assessment of Proposed Indicators

An analysis using Principal Components Analysis (PCA) based on correlations in JMP 15.1 for the zooplankton indicators, major environmental and biotic drivers for the upper and middle bay (Fig 2.3.6) naturally split the time series into 2 time-stanzas. All of the points on the negative PC1 axis are from 1988 – 2018 with the exception of 1991, 2005 and 2011 for B and 1991 for HB. The PC1 accounted for 35.1% of the variance and was fundamentally a nutrient axis dominated by planktivorous fishes (31%), TP (29%), and %DGM (-17%) (Table 2.3.1). This reinforces the fact that while %DGM is an effective indicator for planktivorous fishes (top-down), the bottom-up driver of TP is of greater magnitude. The PC2 (24.1%) was dominated by positive relationships with *Chydorus* (31%) and %*Mesocyclops* (49%), postively with temperature (10%). The PC3 (15.4%) had similar but negative relationships for %DGM (23%) and *Eubosmina:Bosmina* (10%) with temperature (55%). This is expected because zooplankton biomass is utlimately tied to food resources (phytoplankton biomass driven by TP), top-down reductions from planktivores, but also by temperature (Shuter and Ing 1997). It is unknown if the increasing summer termperatures in the Bay of Quinte will affect the production of *Daphnia galeata mendotae* and favour less preferred species such *Eubosmina* or *Chydorus*.

Summary

Chydorus biomass and the ratio of *Eubosmina* to *Bosmina* are both positively related to increasing eutrophy, and unusually high levels of either are indicative of deteriorating health of the zooplankton community, at least as it relates to eutrophication. Top-down indicators of increasing fish planktivory are less robust in the upper Bay of Quinte, given that the system is dominated by bottom-up influences. The percentages of both *Daphnia galeata mendotae* and the large cyclopoid *Mesocyclops* may have utility in identifying unusually high levels of planktivory, although both are less sensitive to responding to more moderate levels of predation. There is an expectation that as bottom-up influences wain in the upper Bay of Quinte that these metrics should improve in effectiveness as they have in the lower bay. In recent years, declining zooplankton populations in the upper bay appear have the potential to limit production and survival of fish species that depend on zooplankton for survival, especially juvenile life stages.

Table 2.3.1 *Eigenvalue loading matrix from principal components analysis for chosen factors in JMP 15.1 for Belleville and Hay Bay (see Fig. 2.3.5).*

Factor	**PC1**	**PC2**	**PC3**	**PC4**	**PC5**	**PC6**	**PC7**
Planktivorus Fish	0.8706	-0.1982	0.2828	-0.1033	0.1649	0.0942	0.2758
Total Phosphorus	0.8443	0.1707	0.1540	-0.2999	0.2548	0.1368	-0.2462
Surface Temperature	0.2935	0.3748	-0.7685	0.2791	0.1139	0.2996	0.0479
log *Chydorus* Biomass	0.4665	0.6620	0.1023	-0.1455	-0.5583	-0.0057	0.0274
Eubosmina : *Bosmina*	0.4695	-0.0557	0.3281	0.8108	-0.0544	-0.0549	-0.0735
%*Daphnia galeata* m.	-0.6452	0.2836	0.4945	0.0593	0.0427	0.5032	0.0173
%*Mesocyclops*	-0.2176	0.8334	0.1537	0.0610	0.3611	-0.3107	0.0619

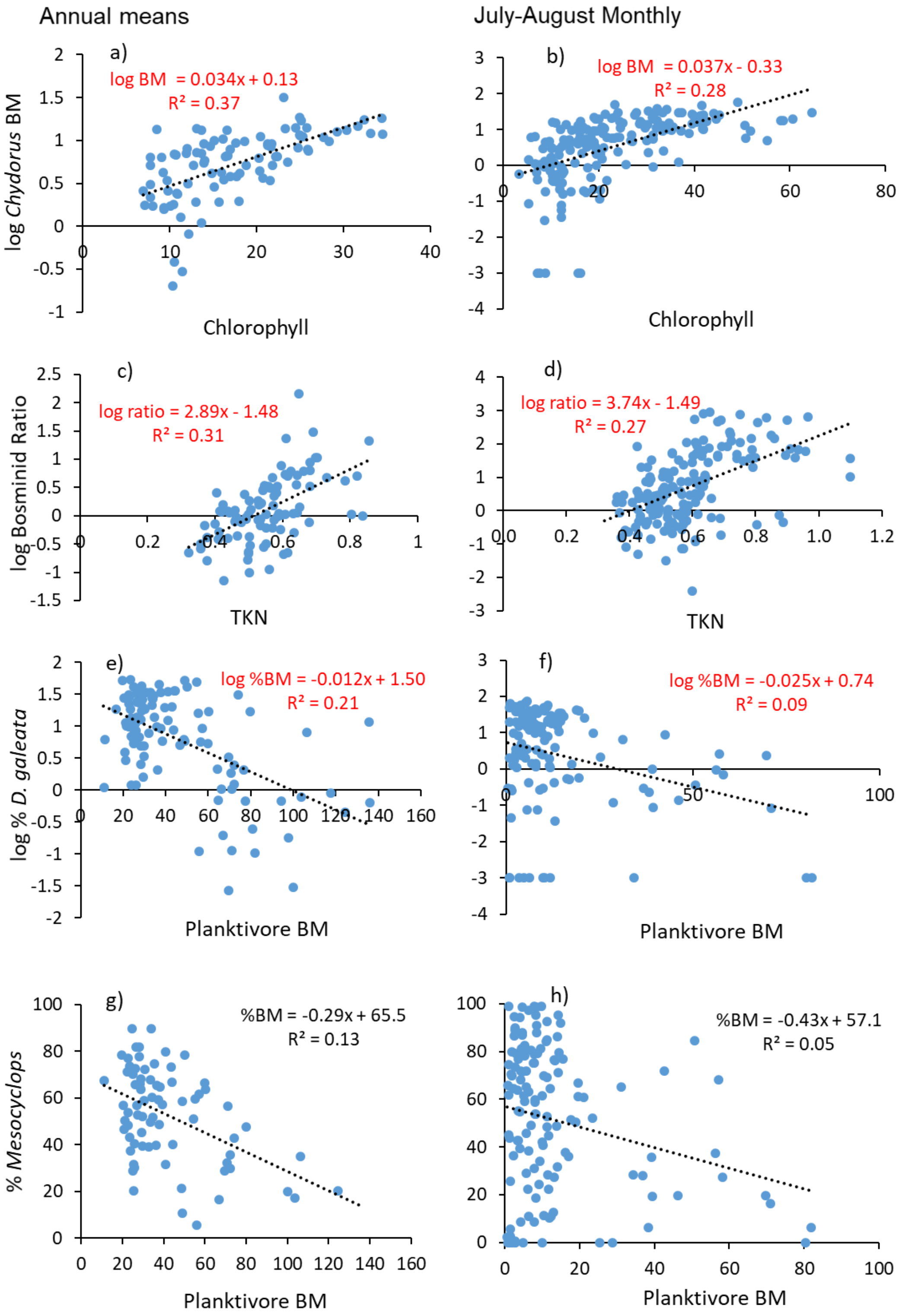


Figure 2.3.1 *Bottom-up (Trophic status) regressions for log Chydorus biomass and chlorophyll a (a and b) and log Eubosmina:Bosmina biomass ratio and TKN (c and d). Top down (planktivory) relationships are for log % Daphnia galeata mendotae (e and f) and % Mesocyclops (g and h). Annual mean data (a, c, e, g) are plotted on the left, July and August monthly data on the right (b, d, f, h). Significant linear regression fits in red.*

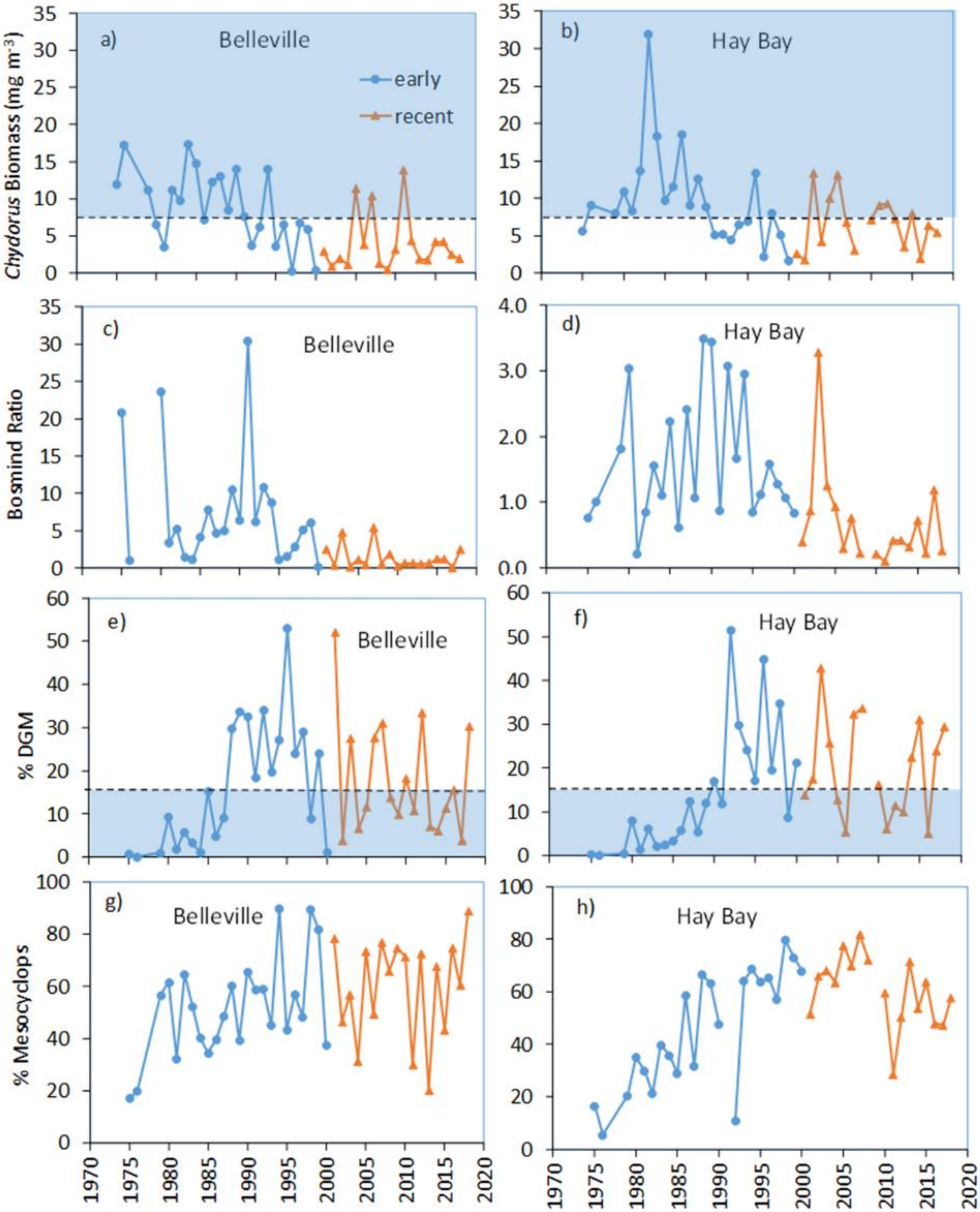


Figure 2.3.2 *Chydorus biomass (a and b) with target shown as the dashed line, biomass of Eubosmina to Bosmina (bosminid ratio; c and d), Daphnia galeata mendotae biomass as % of crustacean biomass (% DGM; e and f) with the minimum target range shown by the box, and Mesocyclops biomass as a % of adult cyclopoid biomass (% Mesocyclops; g and h) at Belleville (left) and Hay Bay (right) during the earlier time stanzas (1975 to 2000) and the recent stanza (2001 to 2018).*

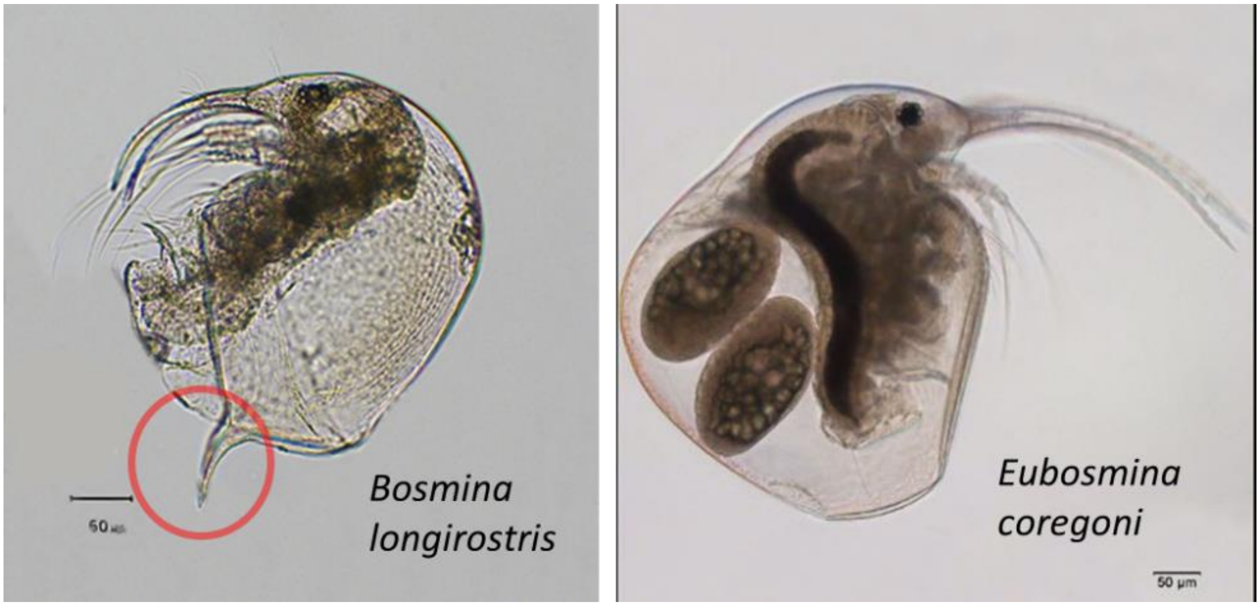


***Figure 2.3.3** identification features of Bosmina longirostris (left) and Eubosmina coregoni (right) with emphasis on the tail spine. Also note that Eubosmina has longer antennae. Images from then New Hampshire Zooplankton Key* (http://cfb.unh.edu/cfbkey/html/).

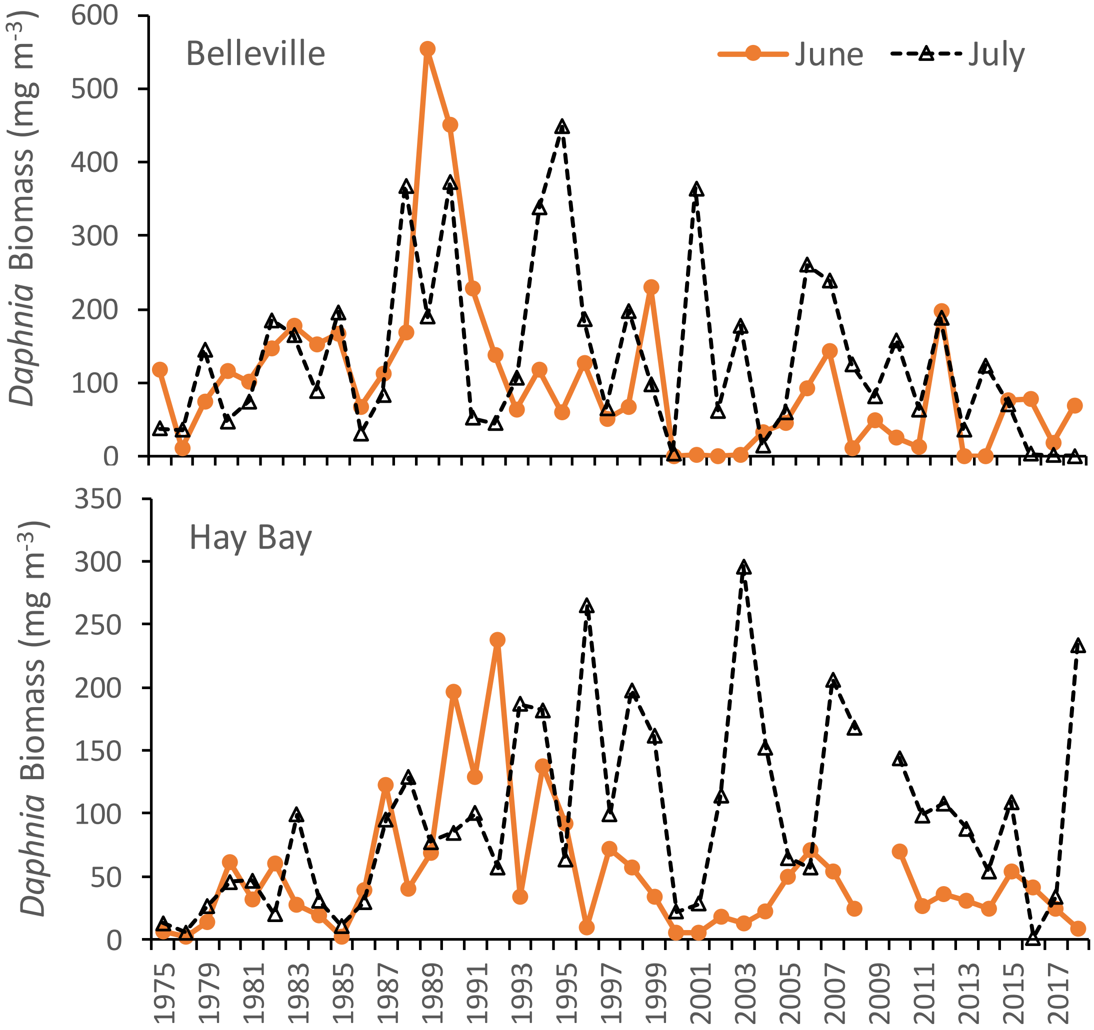


***Figure 2.3.4** Mean Daphnia dry-weight biomass during June (solid line) and July (dashed line) at Belleville and Hay Bay stations in the Bay of Quinte over the 1975 to 2018 time period.*

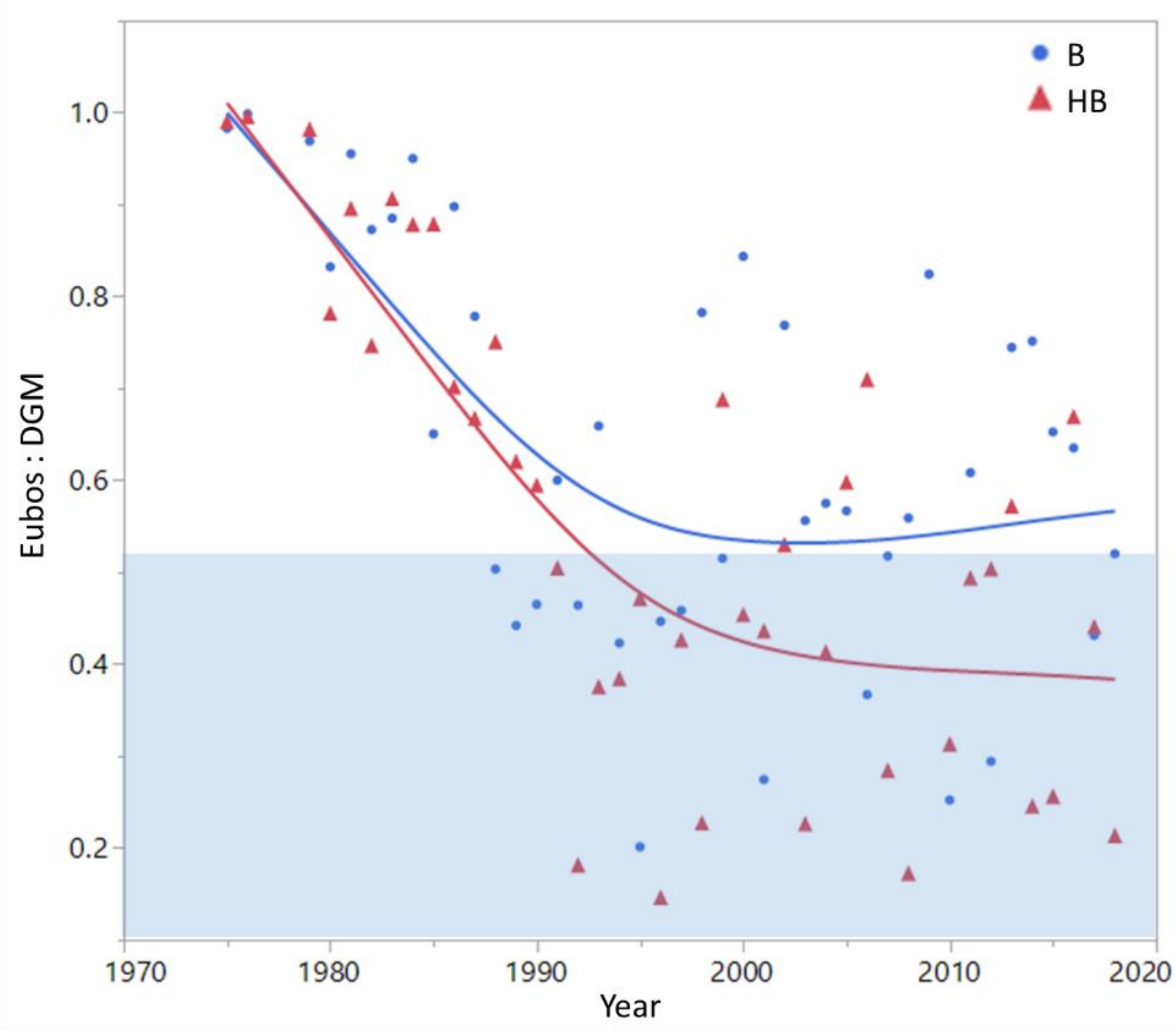


Figure 2.3.5 *The normalized Eubosmina to Daphnia galeata mendotae ratio [eubos:(DGM+eubos)] of dry-weight biomass (mg m^{-3}) for Belleville (B) and Hay Bay (HB). Shaded area encompases values < 0.52 which are within 1 standard deviation of the mean at HB.*

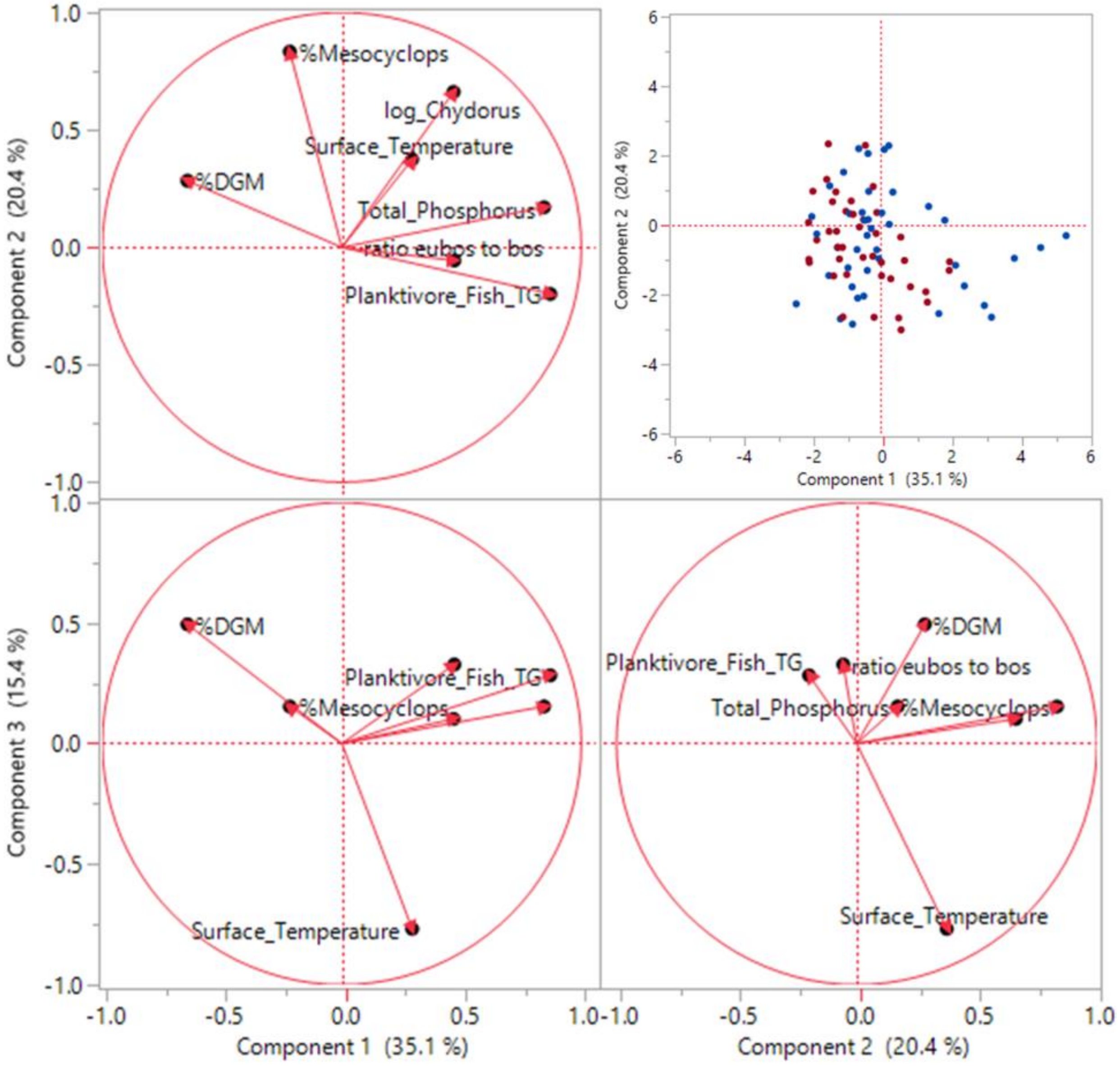


Figure 2.3.6 *Principal component analysis plots from JMP 15.1 of PC 1-3 for Belleville (blue dots) and Hay Bay (red squares) for the years 1975-2018. Factors used are: Surface_Temperature (°C), Total_Phosphorus (mg L^{-1}), Planktivore_Fish_TG (Trawl and Gillnet from MNRF CPUEwt), % Daphnia galeata mendotae (%DGM by percent of total zooplankton biomass), %Mesocyclops (by percent of total zooplankton biomass), log_Chydorus (dry-weight biomass mg m^{-3}) and ratio eubos to bos (Eubosmina to Bosmina dry-weight biomass mg m^{-3}).*

S2.4 Trophic Ratio Indicators

Food webs are a complex interconnection of resources and populations which results in the flow of energy through an ecosystem. These intricate linkages can be simplified into chains of direct linkages (e.g. predation or uptake) forming “trophic (or Eltonian) pyramids” (Elton 1927, Lindeman 1942). The relationships between these trophic levels can be determined by using their biomass ratios to measure energy transfer, and the efficiency of energy transfer, at each link of the food chain independently. This technique has both theoretical and empirical support, and has been used previously to assess disturbances in the energy transfer between trophic levels (McCauley and Kalff 1981; Jeppesen et al. 2005). When systems deviate from expected biomass ratios (e.g. top-heavy inverted biomass pyramids; McCauley et al. 2018), it can be an indication of alternative endogenous or exogenous energy source pathways; well-functioning ecosystems have efficient energy transfer between food web linkages, whereas inefficient energy transfer between linkages suggests a disruption in the system.

The shape of the trophic biomass pyramid is controlled by both bottom-up (resource availability) and top-down (predation) drivers (McQueen et al. 1986; Carpenter et al. 1987, 2001). It is possible to determine if bottom-up or top-down disruptions are the cause of changes to a system, however a clear understanding of the linkages in the ecosystem is necessary to the interpretation of the trophic ratios. Factors which influence bottom-up processes, such as how light and nutrient input control the productivity of a system can be a dominant factor, especially in eutrophic systems but it is only part of the ecosystem equation (Harvey et al. 1935; Lindeman 1942; McCombie 1953; but see McQueen et al. 1986 for discussion). Consumption from the top down can also impact zooplankton composition and biomass (Hrbáček et al. 1961; Brooks and Dodson 1965), which has subsequent impacts on phytoplankton (Carpenter et al. 1987). This theory of trophic cascade (Carpenter et al. 1987) quickly led to the idea that biomanipulation of lakes could change the community composition to one favored by managers (Lammens et al. 1990).

Since many factors can influence trophic ratios, including lake morphometry (deep vs shallow) and food web composition (presence of predator species), the best use of trophic ratios is to detect change in the same system over time. The upper Bay of Quinte is a shallow, eutrophic freshwater ecosystems, and it is expected to show shorter and more direct relationships (e.g. “food chains”) common to nutrient impacted systems, resulting in stronger ratio responses to changes in the system (Drenner and Hambright 2002). However, the Bay of Quinte is not a bounded lake system (it is an embayment) where much of the previous work on trophic ratios have been applied. As one progresses towards the open boundary with Lake Ontario, these deeper areas are expected to respond differently to changes, particularly with planktivorous (e.g. Alewife) and piscivorous fishes (e.g. Walleye) moving through this open boundary (Ridgway et al. 1990; Bowlby and Hoyle 2011). Regardless of these complications, trophic biomass ratios, particularly zooplankton:phytoplankton have been shown to be very effective in tracking changes to ecological groups over ranges of drivers including trophic status (bottom-up effect) as a function of total phosphorus (McCauley and Kalff 1981; Jeppesen et al. 1997b), and planktivorous fish (top-down) (McQueen et al. 1986; Jeppesen et al. 1997a; Mehner 2010).

In the previous Quinte assessment (Currie et al. 2017b), a suite of 15 trophic ratio metric were proposed which were a combination of bottom-up and top-down indicators of which 3 (20%) showed some indication of improvement. The intent is to reduce these to a very few key indicators that can be used for monitoring the Bay of Quinte for potential improvements to the function of the food-web. Also, since the DFO GLLFAS lower trophic group has now applied this food web approach to assess other Areas of Concern we will compare the reduced set of trophic ratios from the Bay of Quinte to other Lake Ontario AOCs (Hamilton Harbour and Toronto Harbour) and Lake Ontario reference sites (LO81, LO2, and BUR). This will make it possible to examine where within the Bay of Quinte food web disruptions may be occurring and track changes to the system spatially and temporally.

Trophic Ratio metrics within the Bay of Quinte

A note on nutrients. Although total phosphorous (TP) is generally viewed as the limiting nutrient in freshwater systems (Schindler 1977), when present in high concentrations, particularly through sediment release (internal loading of legacy phosphorus), nitrogen can become limiting in eutrophic systems (Elser et al. 2007; Conley et al. 2009; Kusmer et al. 2019). This is the case in the Bay of Quinte where Total Kjeldahl Nitrogen (TKN) is a significantly better predictor of total phytoplankton biomass (see Section 1, and Nicholls and Hurley 1989), and all the other phytoplankton groups (except Chrysophyceae). So although TKN will be used as the driver of production in most of the analyses within this report, to compare with previous research, TP relationships will be used since the predictive power is still reasonably high and TKN is often not measured or reported.

In order to compare biomass values with consistent units, it was required that we transform some measurements. Zooplankton biomass is calculated based on length to dry-weight equations (see Bowen 2017) but phytoplankton are measured using settled biovolume equivalent wet-weight to be consistent with values collected early in Project Quinte by the Ontario Ministry of the Environment. As such we corrected zooplankton biomass (mg m^{-3}) based on a 10% dry-weight to wet-weight conversion. While each group of zooplankton will have different ratios, the composition in the Bay of Quinte is dominated by cladocera with some copepods, and this commonly used 10% value is intermediate between values found in previous studies (Lawrence et al. 1987; Kiørboe 2013). The Trawl and Gillnet fish data from MNRF is already in wet biomass, but there are no units of volume with this combined data. To generate a volumetric value, the DFO electofishing data was first converted to mg m^{-3} by converting the g weight to mg and assuming the volume sampled was 10 m (wide swath) x 100 m (long) x 1.5 m (consistent depth) (Brousseau et al. 2005), giving a correction of eFish (mg m^{-3}) = eFish(g) / 1.5. To get a correction to volumetric biomass for MNRF fish data we regressed the values of biomass per volume water sampled by the electrofishing program at DFO in the Bay of Quinte with the MNRF values for the years where sampling occurred concurrently, assuming an intercept of zero (Fig 2.4.1). The resulting correction to convert the MNRF fish data to mg m^{-3} is: eFish (mg m^{-3}) = 51.621 * MNRF (CPUEwt), $r^2 = 0.79$, $p<0.0001$.

Biomass ratios with nutrients

The underlying relationships of the individual biomass measured at each trophic level with total phosphorus in the Bay of Quinte can be seen in Fig. 1.12 in Section 1, State of the Ecosystem. These have been corrected to be consistent wet-weight biomass, but those following are in their original format, as noted by DW (dry-weight) or WW (wet-weight), so as to be consistent where possible with previous reports. Overall, the Bay of Quinte is driven by extremely high levels of phytoplankton biomass and poor transfer of this biomass to the upper trophic levels. These are some ratios that can be used to determine impairments within that trophic transfer. The following trophic ratios are found in Table 2.4.1.

Total Phytoplankton Biomass (WW mg m-3) : Total Phosphorus (mg L-1)

The Bay of Quinte has extremely high phytoplankton biomass to TP (Phyto:TP) ratios (Table 2.4.1). The Bay of Quinte in fact still has the highest phytoplankton:TP ratios of all of the sites in the Great Lakes monitored by DFO-GLLFAS (Table 2.4.2).This means that for every unit of phosphorus, there is more phytoplankton produced in the Bay of Quinte AOC than any other site, which is why blooms (determined by biomass) are so common. Historically, there was extremely high phytoplankton biomass per unit of total phosphorus across the entire bay when compared to the more recent time stanza, which is unsurprising considering the Bay of Quinte was a hyper-eutrophic system and is still currently exhibiting eutrophic conditions. In eutrophic systems, more biomass is retained as phytoplankton because it is overproduced and not effectively grazed and ultimately transferred to higher trophic levels. Partly, this is due to the production of less preferred phytoplankton groups in eutrophic systems (e.g. filamentous or colonial forms). The most efficient productivity peak in Quinte series given by Phyto:TP was during Stanza 2 after phosphorus controls (Fig. 2.4.2). Prior to this there were a number of factors limiting phytoplankton growth (light limitation, nitrogen limitation). We see a steady reduction in the Phyto:TP ratio across the Bay during the clear phase after 1994, but only at Conway are we seeing values which begin to approach mesotrophy and even at Station 81 within Lake Ontario, there is still elevated algal productivity being driven by influences of the Bay of Quinte (Table 2.4.2). <u>The expectation is that the upper Bay of Quinte sites should approach a Phyto:TP ratio similar to the other major eutrophic AOC of Hamilton Harbour which has similar TP concentrations, so we recommend the target based on the mean value of Hamilton Harbour of 50000</u>. The upper and middle Bay sites are still almost double this value, primarily because of the continued dominance of filamentous diatoms and cyanobacteria (see section 2.1).

Total Phytoplankton (WW mg m^{-3}) : Extracted Chlorophyll a (µg L^{-1})

We wanted to highlight the issue with confounding the measurement of algal biomass with chlorophyll-a, both within the Bay of Quinte and applying chlorophyll targets across AOCs. The ratio of Phyto:Chl is high in the Bay of Quinte, and when averaged across the two large time stanzas, has remained relatively unchanged since the mid-late 1970's when sampling programs in the Bay began collection chlorophyll samples. However, the Phyto:Chl ratio peaks in the late 1990's across all stations with significant increases in phytoplankton biomass which is not reflected in proportional increases in chlorophyll a. As such there has been a differing relationship during the clear period between trends in phytoplankton biomass and chlorophyll-a.

Total Phytoplankton Biomass + Bacterial Biomass + Autotrophic Picoplankton biomass (WW mg m^{-3}) : Total Phosphorus (mg L^{-1})

This trophic ratio includes both the autotrophic (phytoplankton) and heterotrophic (microbial food web) to phosphorus ratio. As such it includes the phytoplankton, the bacterial sized picoplankton and bacteria. The patterns for the Phyto+Bact+APP:TP ratio are similar to the Phyto:TP ratio in the Bay of Quinte, though the microbial loop was collected from 2000 onwards only. In more recent years at Belleville, bacteria and APP levels have been on the rise, which contributes to the increase in this trophic ratio relative to the other stations. The shunting of potential algal productivity into bacteria is a disruption to the food web because bacteria, while grazed by some members of the zooplankton, is not a preferred food source. The lack of change in the ratio suggests no improvement to the trophic transfer during this time period within the food web in the Bay of Quinte AOC.

Total Zooplankton biomass (DW mg m^{-3}) : Total Phosphorus (mg L^{-1})

Unlike the phytoplankton ratios which are dominated by nutrients, zooplankton to nutrient ratios are driven by both bottom-up and top-down controls. It is expected that Zoop:TP ratios are lower than the Phyto:TP because only a portion of the algal production is converted into zooplankton biomass, but there is an expectation of more zooplankton per unit phosphorus in well-functioning ecosystems (see Fig. 1.11). The ratios in the upper Bay of Quinte were dynamic historically because of difficulties with trophic efficiencies in the system (Fig. 2.4.2). In the 1970s, there were not many zooplankton measurements, but the hypereutrophic conditions depressed zooplankton biomass by a combination of top-down (very high levels of planktivorous fishes) and bottom-up factors (poor algal food sources). By the time phosphorus controls were in place and the system had settled into a new phase (about 1983), the Z:TP ratios initially increased to the highest levels, but have declined from the early 1980s till about 2000 and have not changed since. Hay Bay has shown consistently higher levels of Zoop:TP than the Bay of Quinte AOC sites.

Total Planktivorous Fishes (CPUEwt) : Total Phosphorus (mg L^{-1})
The planktivorous fishes used in this ratio were chosen in consultation with MNRF biologists and include primary planktivores (Alewife), small planktivorous fishes (e.g. shiners), seston consumers (Gizzard Shad) and the prey switching species of White and Yellow Perch. All of these can be used as forage fishes by piscivores. By examining the connection between TP and planktivorous fish, several trophic linkages are incorporated and planktivores are a very good indicator of both piscivore and total fish biomass. The long term historical fish data for the Bay of Quinte was collected by trawl and gill nets in the same locations by the OMNRF starting in 1972. Historically, Conway showed higher ratios than until the early 1990s, being more than double that of the upper bay (more planktivorous fishes per unit of total phosphorus). Now, the PlanktFish:TP ratio is similar for all of the Bay of Quinte sites, though Hay Bay has always been lower. There was been a significant reduction in planktivorous fishes biomass in the upper and middle bay following phosphorus controls (mostly reductions in Alewife and Gizzard Shad). This change is reflected in the turbid vs clear phase ratios, but the catch has been quite invariant since the early 1980s (Fig 1.11). In recent lower phosphorus years (since 2016), the planktivorous forage fishes biomass has been very low and has seen an increased percentage of Alewife and Gizzard Shad again.

Trophic biomass ratios

Total Zooplankton Biomass (DW mg m^{-3}) : Total Phytoplankton Biomass (WW mg m^{-3}) (Z:P)
Like the Zoop:TP ratio, this ratio is also driven by both bottom-up and top-down controls. This ratio can change by an increase or decrease in either component. This tendency is what makes the Z:P ratio a very effective indicator in ecology (Jeppesen et al. 1997a; 2005). Though the ratios at Quinte have been relatively consistent over the years, there was an increase in the Z:P ratio during the high phosphorus years until the early 1980s because zooplankton biomass was depressed during these years (Fig. 2.4.3). At Hay Bay the trend has continued slightly upward as expected and is consistently higher at Conway except during the last years of the time-series. The calculation of Z:P ratio has been hampered by the lack of phytoplankton biomass estimates at Conway and Hay Bay in the last decade of the time-series. In the upper bay however, the Z:P ratio has not changed since the early 1980s when the BUI was listed as impaired. There are occasional occurrences of higher Z:P at Belleville during the last decade of sampling (2012, 2014, 2018) closer to those at Hay Bay, but the average Z:P has not changed since these higher values are interspersed with very low Z:P values. This increased variability since 2010 is driven by both reductions in phytoplankton and zooplankton biomass, but zooplankton biomass in particular has been extremely low in the last years of the time-series. During the clear phase, 7 of 18 (39%) sampling years at Hay Bay have been above the mean + 1 standard deviation (0.062), but only 3 (1989, 1990, 2001) years at Belleville have met this level. There is an expectation that the Z:P ratio at Belleville should show a steady trend upward to a recommended target of 0.062.

Total Zooplankton Biomass (DW mg m^{-3}) : Total Phytoplankton Biomass + Bacterial Biomass + Autotrophic Picoplankton biomass (WW mg m^{-3})
By including the autotrophic portion of the microbial loop into the Z:P ratio, the trends in the Bay of Quinte remain consistent, increasing with distance towards Lake Ontario, this indicates that the microbial loop is a consistent factor that has not changed. For the Bay of Quinte, this does not provide a better option than the simple Z:P ratio.

Daphnia Biomass (DW mg m^{-3}) : Total Phytoplankton Biomass (WW mg m^{-3})
The *Daphnia* to phytoplankton ratio is refinement of the Zoop:Phyto ratio related specifically to the large-bodied, efficient cladoceran grazer production as a function of phytoplankton resource. In the upper and middle bay, this ratio follows the pattern of the Z:P ratio with low values in the 1970s to the early 80s with a continued increase at Hay Bay and no increase at Belleville. This ratio does not work as well at Conway, which shows ratios not much different from the upper and middle bay due to the more dominant role of copepods at this deeper site. This ratio currently does not provide a better option than the simple Z:P ratio but might be more relevant in the future.

Heterotrophic Nanoflagellate Biomass + Ciliate Biomass (WW mg m^{-3}) : Bacterial Biomass + Autotrophic Picoplankton biomass (WW mg m^{-3})
This metric is the ratio of heterotrophic consumers to producers (autotrophs and bacteria) within the microbial food web. Since we only have data from 2000 – 2018 for the microbial food web, this ratio can only be calculated for this period (see section 2.2 for more details). HNF tends to dominate the biomass of ciliates within microbial consumers in the Bay of Quinte. The ratio in the upper bay is driven by high HNF in the early 2000s, but otherwise indicates relatively equal biomasses given relatively high bacteria levels, whereas the middle bay has a very low ratio, driven by higher bacteria and lower HNF levels. In contrast, the lower bay has a very high ratio driven by high HNF in the early 2000s coupled with very low bacteria levels. This ratio could be useful for determining the amount of energy shunted into bacterial biomass and away from the traditional phytoplankton-zooplankton food web but since is requires specialized lab equipment and trained personnel it is not being recommended as a metric.

Total Planktivorous Fishes (CPUEwt) : Total Zooplankton Biomass (DW mg m^{-3}) (PlanktF:Z)
As we see above, the planktivorous fish biomass (determined with the combined trawl and gillnet data from MNRF) is related to bottom up drivers (best for TKN, but for TP (log transformed) is $r^2=0.52$, $p<0.001$) for most of the time-series and is highly correlated to the total fish biomass (log transformed, $r^2=0.70$, $p<0.001$) in the upper and middle bay. The zooplankton biomass has some additional, though weak, top-down effects which support this metric. All sites showed a decline in PlanktF:Z during the 1970s to early 1980s in Stanza 1, with Conway lagged more to stabilize in the early 1990s and since then the ratios of all of the sites have been more similar (Fig 2.4.3). The upper and middle portion of the Bay of Quinte showed no significant changes in the ratio of planktivorous fishes to zooplankton since the early 1980s, though Belleville had consistently had higher values than at Hay Bay (more planktivorous fishes per unit zooplankton) since 2000. The overall mean value for PlanktF:Z at B is 0.22 which we recommend as a target. All of the values pre-phosphorus controls and only 2 years (1992, 2000 anomalous cold years of poor zooplankton production) are above this value. Hay Bay only had ratios above this target during the pre-phosphorus control years. As such we recommend a PlanktF:Z target of 0.22 be adopted for the upper Bay of Quinte.

Total Planktivorous Fishes (CPUEwt): Daphnia Biomass (DW mg m^{-3})
Because *Daphnia* only represent a portion of the total zooplankton biomass, larger ratios are expected compared to the PlanktF:Z ratio but the patterns follow almost exactly for the upper and middle bay. At Conway, the ratios early in the 1970s to 1980s are extremely high due to the high numbers of planktivorous fishes and relatively lower *Daphnia* biomass there. There is an increase in the upper bay or middle bay, driven by a larger relative decrease in fish biomass relative to *Daphnia* biomass. The extreme decline in the PlanktF:Daph ratio at Conway are being driven by a single large ratio in 1976 where *Daphnia* biomass were abnormally low. Omitting this data point gives a turbid phase ratio at Conway of 4.60. The decline since 1994 is again driven by declines in fish biomass. This metric provides no advantage to the PlanktF:Z ratio for the upper bay.

Carnivorous Zooplankton Biomass (DW mg m^{-3}) : Total Zooplankton Biomass (DW mg m^{-3})
The percentage of carnivorous zooplankton to total zooplankton can be a relevant metric because these tend to be larger species which are preferential food for forage fishes. However this ratio is more related to station depth as a number of species of carnivorous zooplankton are found preferentially at deeper sites. Furthermore, two species of carnivorous zooplankton are recent non-native (*Bythotrephes* - 1985 and *Cercopagis* - 1998) invaders to Lake Ontario. At the upper and middle bay sites, this ratio has been relatively invariant, though it has decreased slightly, tracking the reduction in total zooplankton biomass, but it has also exhibited year to year variability. The deepest site in the Bay of Quinte is in the lower Bay at Conway; this ratio has shown wide swings in value. Given this variability and the fact that the newly introduced predatory cladocera are named after their large anti-predation spines making them less palatable to many smaller fishes, this is not suited to being used as a metric for food web changes in the Bay of Quinte.

Planktivorous Fishes (CPUEwt) : Total Fish Biomass (CPUEwt) (PlanktF:TotalF)
The percentage of planktivorous fishes of total fishes as biomass examines the relationship of the direct linkage near the top of the foodweb. There have been overall decreases in the PlanktF:TotalF ratio observed at all the sites due to a much larger decline of planktivorous fish relative to the decline of total fish. This decline was precipitous during the pre-P control period and remained relatively invariant until about 2010. In the lower bay site, the forage fishes biomass has increased to higher levels starting in the mid 2000s. During the most recent decade in the upper and middle bay, the percentage of planktivorous fishes has decreased again precipitously and is now at historically low levels. This may be matching the very low levels of zooplankton biomass found during this period limiting resources for planktivorous fishes, but the recent decreases in fish biomass are more rapid. This is in part due to a larger percentage of piscivores and benthic fishes in the upper bay which might be preying on YOY fishes. This can be a very useful metric for determining energy flow to predatory and sport fishes in the Bay of Quinte, but as it does not use a metric based on the plankton, it is not being included here as a recommended target.

Piscivorous Fishes (CPUEwt) : Planktivorous Fishes (CPUEwt) (PiscF:PlanktF)
The normalized, relative composition of piscivorous fishes to the planktivorous fishes given by piscivore/(planktivore+piscivore) gives a response of the highest fish tropic level to their forage fishes prey. This ratio, while not dealing directly with the plankton is very relevant to the food-web because this is an indication of top-down control over the system (Carpenter et al. 2001) and a higher percentage of piscvores is a desired outcome for BUI 3 (Fish Populations). The expectation is that as piscivores increase, planktivores will decrease (consumed), releasing zooplankton (less predation), which will exert a greater grazing pressure on the phytoplankton. In the Bay of Quinte however, we do not see this outcome with planktivores, zooplankton and phytoplankton all decreasing (inversion of the trophic pyramid). We saw high numbers of piscivores during the 80s and early 90s during a period of still high productivity (Fig. 2.4.5) and more recently this ratio has gone up above 1 again, but without increased productivity and no Z:P ratio increase over this time. This is of concern because since 2013, without increased zooplankton biomass, the amount of energy transferred up the food-chain is being limited. This is likely due in part to the poor food quality of the phytoplankton (filmentous forms). As such, the improvements in piscvore biomass is occuring during periods of algal blooms and there is a concern that the phytoplankton biomass (lowest in the time series) will not be able to support the current biomass of piscivores (Fig. 0.2) which are relatively long-lived and likely to show a lag in response. We highly recommend continued monitoring of fish populations in the upper bay, particularly for condition (Wuenschel et al. 2018) because the increased PiscF:PlanktF and a decrease in condition <u>PiscF:PlanktF > 1 may be an early warning signal</u> forecasting a sudden drop in fish biomass, which has been predicted for Bay of Quinte (Hossain et al. 2019).

Gizzard Shad (CPUEwt): Yellow Perch (CPUEwt) (GizzSh:YPerch)
The percentage of Gizzard Shad relative to Yellow Perch biomass examines the relationship between a fish within the planktivore group that eats more from the detrital pool to one that is more benthivorous or piscivorous. There have been overall decreases in the GizzSh:YPerch ratio observed at all the sites due to the decline of all planktivorous fish relative to the decline of total fish until about 2000 (Fig. 2.4.5). During the most recent time stanza, the percentage of Gizzard Shad has increased again in the upper and middle bays, but has remained low at the lower bay sites. This might be related to the increased bacterial biomass that has been seen since 2009 in the upper bay (S2.2, Fig. 2.2.3). At Belleville in particular, the total fish biomass is at historic lows and the percentage of Gizzard Shad is again approaching that of the pre-phosphorus control period. While this metric does not use plankton measures, it is very relevant to the food web functionality of the Bay of Quinte so we recommend this ratio be used as an early warning warning metric for more intensive monitoring (Su et al. 2020). The relative biomass of GizzSh:(YPerch+GizzSh) should be maintained below 0.2 which corresponds to the combined sites mean + 2 standard deviation units from the clear phase stanza 3.

Total Benthos (DW mg m^{-3}) : Total Phytoplankton Biomass (WW mg m^{-3})
Benthic biomass is driven primarily by organic food resources which are sourced mostly from phytoplankton. The ratio of Benthos to Phytoplankton, limited to the upper and lower Bay of Quinte, shows an increase in more recent times compared to historical conditions. This trend is being driven by the early increase in benthos from the poor diversity in the early years, but the benthic biomass estimate is variable year to year and has not significantly changed over the rest of the time series in the upper bay, so the pattern is driven primarily by the decline in phytoplankton biomass. This is the result of decades of legacy organic matter deposition in the sediments. At Conway there has been a significant increase in benthic biomass including from *Dreissena*. However, the benthos time-series is limited to the upper and lower bay only from 1982 – 2011 and is not currently being monitored, so is better applied for entire food web studies (see Section 1) rather than for trophic ratios.

Total Zooplankton Biomass (DW mg m^{-3}) : Total Benthos (DW mg m^{-3})
A similar trend as the Benthos:Phyto ratio is true for the Zooplankton: Benthos ratio and the limitations are also the same. Benthic biomass is always much larger than zooplankton biomass even when compared volumetrically. Historically, the ratios were higher and they significantly decreased more recently, due to declines in zooplankton biomass coupled with little or no change in benthos biomass.

Long-term trends in ratios for the Bay of Quinte
Plots of the four bottom-up biomass to resource ratios (Phyto:TP, Phyto+Bact+APP:TP, Zoop:TP and PlanktF:TP) through time are shown in Fig. 2.4.2. There is considerable year to year variability but the long-term trends for the three sites are sometimes more apparent than when averaged between the two major time stanzas (turbid vs clear phase). Belleville and Conway have more consistent annual sampling than Hay Bay for the parameters shown, and the microbial loop research began in 2000.

Though not pefectly coherent, all of the resource ratios follow similar patterns. There is a slight downward slope with year for phytoplankton, microbial and zooplankton biomass. There is wider year to year swings in biomass:TP before 2000 and with the years only sometimes synchronous for phytoplankton and zooplankton. Phyto:TP shows significant year-to-year variation peaking in the late 1980s and stabilizing at lower levels through the 2000s. Zoop:TP also shows significant year-to-year variation, but does not consistently track with Phyto:TP, sometimes showing a year or two lag and sometimes matching extreme values. This suggests that there are other factors beyond just phytoplankton resources controlling zooplankton biomass. This is not unexpected because both food and temperature are known to drive zooplankton production (Shuter and Ing

1997), but also because bacteria and hetrotrophic nanoflagellates are a known food source for many zooplankton species (Jürgens and Stolpe 1995; Beisner et al. 2006) but have only measured since 2000. Phyto:TP tracks very well with Phyto+Bact+APP:TP in the early 2000s when the microbial loop studies began, but started to deviate into the 2010s as bacterial and APP levels increased (greater bacterial biomass with TP), thereby increasing that ratio relative to phytoplankton biomass alone. This may indicate reduced grazing by HNF and zooplankton on bacterial production thus retaining biomass in bacteria. This may also be related to the significant increase in summer high temperatures (Woolway et al. 2020) during the last decade of sampling (see S2.2 Microbial Loop Indicators).

The ratio of PlanktF:TP is relatively stable over time, with the largest change being at Conway. It trends downwards across time for all three sites, peaking in the early 1980s, tracking the reductions in TP, illustrating that planktivorous fishes biomass is primarily controlled by nutrients (bottom up). There has been an abrupt drop in PlanktF:TP in the last few years of sampling at all sites. It is worth noting that the piscivorous fishes biomass was higher than the planktivorous fishes in the upper bay mid-1980s to early 1990s, but was less than their biomass through to 2012. The piscivore biomass since then has been at or near that of the planktivorous fishes indicating bottom up control (food availability) including the period of 2014 – 2018 with historically low total fish biomass. The reduction and variation in Zoop:TP but a flat PlanktF:TP is a mismatch which may be resulting in the continuous decrease in total fish biomass in the upper Bay of Quinte which might be exacerbated by the recent increase in the PiscF:PlanktF ratio (Fig. 2.4.5).

The Bay of Quinte has seen an increase in the percentage of piscivores as a function of improvements noted in the fish community structure in the Bay of Quinte (Hurley and Christie 1977; Hoyle et al. 2012). The increase or introduction of piscivores into a system has been linked to trophic effects including the increase in deep bodied or large benthivorous fishes and the export or loss of small bodied fishes which are preferentially targeted as food items (He and Kitchell 1990; Chapleau et al. 1997). These small bodied fishes are often planktivorous and can include the YOY of desired community fishes (Whitefish, Walleye, Yellow Perch etc.). The planktivorous fishes biomass has tracked that of the total fishes biomass over time (Section 1, Fig. 0.2), so even with year to year fluctuations likely driven by environmental factors (Vijerberg et al. 1990). The PiscF:PlanktF ratio was very low during pre-P control and increased to a peak in the mid-80s, declining steadily until about 2013 (Fig. 2.4.5). This initial decline was offset by the fact that Yellow Perch are a dominant portion of the PlanktF metric and they are increasingly of larger size and piscivorous (Bowman 1995, Section 1). This shift can been seen in the Gizzard Shad : Yellow Perch (GizzSh:Yperch) ratio which was very high pre-P control (abundance of Gizzard Shad), but then became very low (Yellow Perch dominated) after the late 80s at all sites, though there have been spikes in the upper bay during the last 5 years. This is the conflict between BUI 8 (Eutrophication) and BUI 3 (Fish Populations). This is born out by reductions in total fishes biomass in the most recent years with 3 of the last 5 years exhibiting the lowest total fishes biomass, coinciding with the lowest zooplankton biomass, of the entire Bay of Quinte dataset (Fig. 1.12, Fig. 0.2). It is recommended that additional studies examining the fish community composition at lower nutrient levels designed to control algal blooms are undertaken because this action has the potential to significantly affect the production of desired piscivorous sport fishes such as Walleye in the Bay of Quinte (see Hossain et al. 2019). The recent sharp increase in the PiscF:PlanktF ratio (Fig. 2.4.5) needs to be considered for fish community targets and compared to that of the PiscF:Z and PlanktF:Z ratio since it appears that trophic transfer is stalled at the level of zooplankton in the upper bay.

The trends in Zoop:Phyto and PlanktF:Zoop are mentioned above but it is worth detailing some of the changes over time. The trends (Fig. 2.4.3) show that there is a correspondence in these metrics between the changes over time and the reduction in TP. At HB, the Zoop:Phyto ratio has increased over time as would be expected ($r^2=0.41$, $p<0.001$) as TP is reduced as seen by the significantly positive trend with time and negative with TP ($r^2=0.18$, $p<0.013$). Less excess phytoplankton biomass is ultimately being produced at HB. Similarly, PlanktF:Zoop ratio has gone down at Hay Bay over time ($r^2=0.11$, $p=0.048$) and with reduced TP ($r^2=0.09$, $p=0.049$). Fewer excess planktivorous fishes are in the system, presumably due to the increased percentage of piscivorous fishes in the system (Fig. 1.11). However, the same is not true in the upper bay. At B, the trend is not significant and is flat, with no change in either Zoop:Phyto or PlanktF:Zoop ratios. This is primarily due to the lack of zooplankton being produced as it continues to decline with the reductions in TP. This trend should be positive in time and negative with TP as it is at HB.

Trophic Ratio comparisons between Areas of Concern

Of the aforementioned ratios, the most relevant in the Bay of Quinte are the Phytoplankton biomass : Chlorophyll, Phytoplankton biomass : Total Phosphorus (Phyto:TP), and Zooplankton biomass : Phytoplankton biomass (Zoop:Phyto) ratios. These three ratios are important because they best capture the bottom up influences driven by nutrient input (TP) as well as top-down controls (Zoop:Phyto) with tightly linked trophic connections. With these three ratios, changes to the ecosystem are more apparent than in other ratios in the Bay of Quinte, and they are simple to obtain and understand. By examining the Bay of Quinte ratios in comparison to other Lake Ontario Areas of Concern, namely Hamilton Harbour and Toronto Harbour, and to Lake Ontario reference sites (LO81, LO2, and BUR), it is possible to put the status of the Bay of Quinte in context and to explore the cause of disruptions, if present.

Trophic ratios can be used to guide management of ecosystems by detecting changes to trends in the system (early warning systems) and by recommending targets which would suggest ecosystem health improvement. The high biomass of phytoplankton produced per unit of total phosphorus and per unit chlorophyll given by the Phyto:TP ratio at all three sites illustrates that the Bay of Quinte greatly overshadows all the other AOCs from this comparison (Table 2.4.2). We would expect Phyto:TP ratios to be lower in efficient oligotrophic systems (e.g. Lake Ontario sites) and higher in eutrophic systems. This is very apparent in hyper-eutrophic historic Quinte where the ratio was highly inflated because of very high concentrations of phosphorus in the system. In fact, it is clear that even station 81 out in Lake Ontario is affected by the outflow of nutrients and algal production from the Bay of Quinte since it has elevated TP and Phyto:TP ratios and its biomass is correlated with values at Conway. Ideally, the upper Bay of Quinte should show Phyto:TP ratios consistent with values in eutrophic-mesotrophic systems. For instance, even in eutrophic Hamilton Harbour, the Phyto:TP ratio averages 47000 but this is much closer to the mesotrophic value in Toronto Harbour than any of the upper or middle Bay of Quinte sites (~130000). Given that the Bay of Quinte will always be a eutrophic system a target value of Phyto:TP value of 50000 is reasonable to use if biweekly sampling estimates are used. This is approximately 50% reduction of current biomass values relative to TP (see Fig. 1.8). It is known from comparisons of lakes with and without piscivores that a mixed community of fishes will tend to decrease the Chl:TP ratio, so the expectation is that the Phyto:TP ratio will have decreased with an increased percentage of piscivores (Drenner and Hambright 2002), which is somewhat the case in Bay of Quinte, though the ratio is still extremely high. In the Bay of Quinte, future trends of Phyto:TP ratios will hopefully decline with decreased loading of nutrients from the watershed, though legacy phosphorus in the organic sediments will continue to be problematic for many years.

The zooplankton biomass to phytoplankton biomass ratio brings both bottom-up controls and top-down impacts together in a single, simple ratio. In comparison to Bay of Quinte, the Z:P ratios in Hamilton Harbour reveal that there is significantly more zooplankton than should be expected per unit phytoplankton; zooplankton is being produced but not consumed due to a lack of planktivorous fishes in the Hamilton Harbour AOC (Table 2.4.2). The lack of change to Zoop:Phyto in the upper bay is symptomatic of the fundamental dominance of the bottom-up influences in this eutrophic system. However, the Zoop:Phyto ratio is expected to increase in value as further reductions in phosphorus are expected over time as it has in the middle and lower bay. However, the Zoop:Phyto ratio in the upper Bay of Quinte is still far lower than would be desired for a healthy ecosystem and unchanged since the beginning of the time-series (Fig. 2.4.4). This is problematic because as total biomass of zooplankton decreases in relation to the decline in nutrients, it will be less able to support the higher trophic levels without increase in the Zoop:Phyto ratio (Hossain et al. 2019) and it is likely this inefficiency is contributing to the record low levels of fish biomass in the upper Bay of Quinte in recent years (Fig. 1.11). Future Zoop:Phyto ratios in the upper bay should be trending upwards with a target of 0.065 which based on the increasing trend at Hay Bay [mean+SD(overall) Zoop:Phyto at HB] for it to support the higher trophic levels without risking collapse.

To compare results of PlanktF:Zoop biomass ratio for the Bay of Quinte with other AOCs it was necessary to use fish biomass estimates from electrofishing (TRCA in Toronto, and DFO for Hamilton Harbour and Bay of Quinte) (Table 2.4.2). These estimates are from nearshore sampling, but are representative of the whole fish community (Brousseau et al. 2005; Hoyle et al. 2012). Since only electrofishing results are presented, the lower Bay of Quinte and the offshore sites in Lake Ontario do not have results. At B, the PlanktF:Zoop ratio showed a slight increase in planktivorous fishes during the clear phase. This is slightly different than in the MNRF data, but this is likely due to the changes in the nearshore habitat related to submerged aquatic vegetation in the upper Bay of Quinte resulting in increased Centrarchid sunfishes (Hoyle et al. 2012). The period since 2001 is not different than found for the entire clear phase. Hay Bay and Conway sites only had electrofishing results from the early turbid phase. The current Bay of Quinte upper bay has values similar to that of Hamilton Harbour which has been noted to have a deficit in planktivorous fishes.This might be due to the increase component of piscivores in the Bay of Quinte rather than the lack of habitat associated with Hamilton Harbour. The Bay of Quinte values are significantly lower than those found in Toronto Region AOC however. The prevelance of planktivorous forage fishes in the Toronto Inner Harbour is seen in both the size and composition of the zooplankton data compared to sites outside the harbour (Bowen and Currie, 2021). During the most recent years in the upper Bay of Quinte there have been very tight food web coupling of fish and zooplankton biomass, which are both at all time lows, so continued electofishing surveys of the upper bay are recommended to determine if this effect seen in the MNRF trawl and gillnet data extends to the shallow nearshore zone.

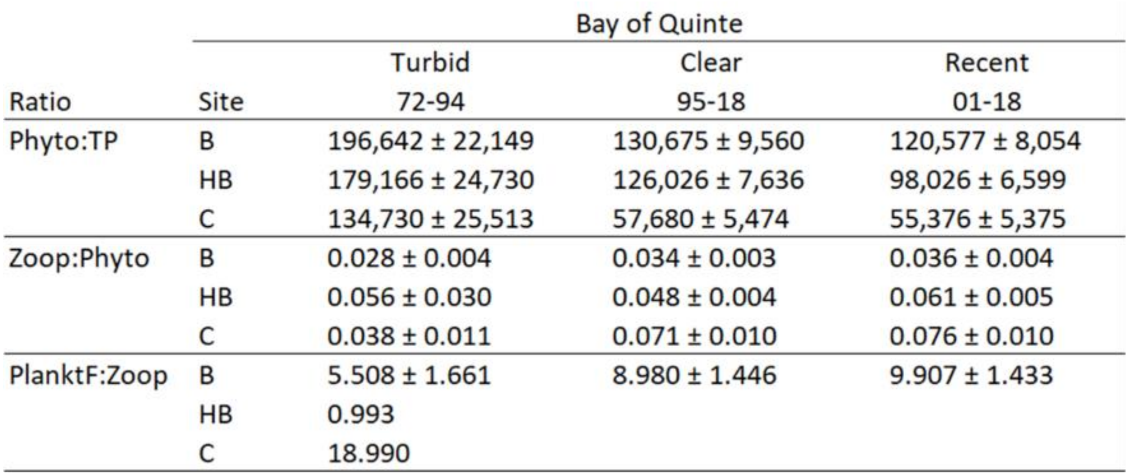
As a word of caution, ratios are extremely sensitive to external forcing on the ecosystem and fluctuate due to patterns of top-down and bottom-up forcing (Drenner and Hambright 2002), which are difficult to disentangle thereby making interpretation difficult and sometimes conflicting (Heath et al. 2014). Although a hard target may be reached, trends and causality of the changes should be of foremost importance. Monitoring for zooplankton must be maintained monthly at a minimum and ideally fortnightly for upper and middle bay stations. Phytoplankton is much more variable, so to get accurate biomass measures to support monitoring of Phyto:TP fortnightly sampling is highly recommended. Composite samples (combining into a single one) can be used to get annual biomass measures for phytoplankton but to identify algal groups individual samples should be maintained for taxonomic counts. Since many of the indicators mentioned here are dependent on accurate measures of fish biomass, we highly support the continued monitoring of fish populations using trawl and gill nets by MNRF in the Bay of Quinte.

One data gap that has become apparent is the lack of knowledge regarding larval and juvenile fishes, especially in the upper bay. We highly recommend that surveys, especially in the vegetated nearshore zone be undertaken to not only determine biomass, but the degree of zooplanktivory by this important group of fishes.

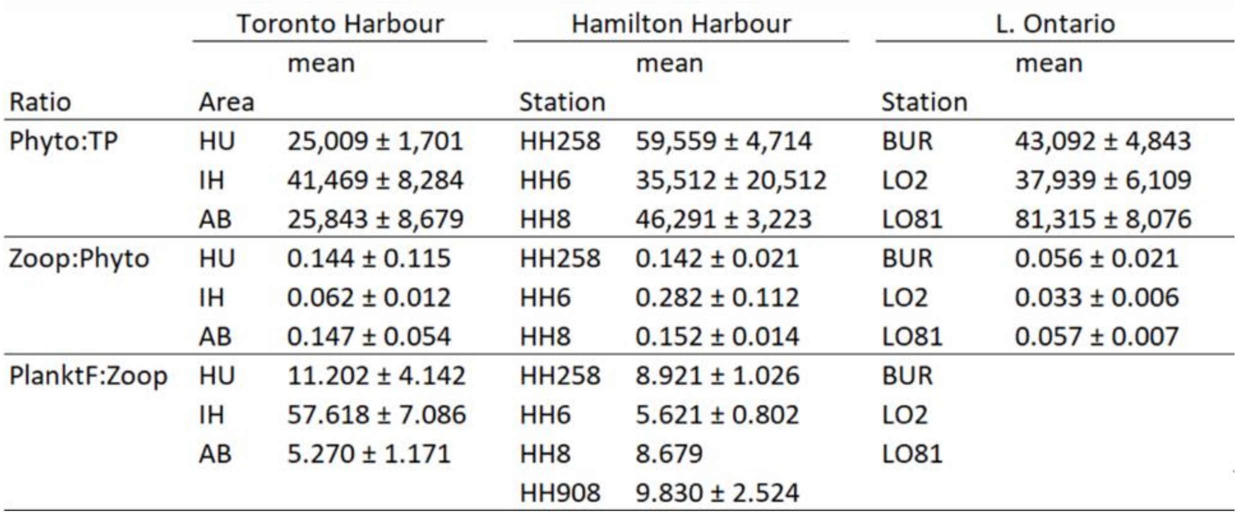
Table 2.4.1 *Trophic biomass ratios for the Bay of Quinte. The * indicates a statistically significant change from the turbid to clear phase based on two sample t-tests with unequal variance. The most recent time stanza is included for comparison. Biomass values are either (WW wet-weight or settled biovolume) or (DW dry-weight): Phytoplankton (WW mg m^{-3}), microbial loop (HNF, Ciliates, APP mg m^{-3}), zooplankton (DW mg m^{-3}), benthos (DW mg m^{-3}), fishes (CPUEwt from MNRF) and total phosphorus in (TP mg L^{-1})*

Ratio	Site	Turbid 72-94	Clear 95-18		Recent 01-18
Nutrients					
Phyto:TP	B	176,745 ± 9,207	131,227 ± 9,606	*	120,577 ± 8,054
	HB	200,692 ± 13,291	117,407 ± 9,271	*	98,026 ± 6,599
	C	101,401 ± 7,206	69,767 ± 7,481	*	55,376 ± 5,375
Phyto:Chl	B	324.15 ± 17.97	312.80 ± 22.06		273.87 ± 14.25
	HB	307.51 ± 23.85	319.94 ± 34.53		244.84 ± 18.70
	C	210.03 ± 14.26	210.24 ± 19.02		175.48 ± 11.37
Phyto+Bact+APP:TP	B		168,789 ± 20,495		174,304 ± 20,936
	HB		148,902 ± 9,424		148,902 ± 9,424
	C		99,847 ± 15,339		99,577 ± 16,955
Zoop:TP	B	5,541 ± 583	4,060 ± 297	*	3,997 ± 347
	HB	6,354 ± 579	5,834 ± 362		5,237 ± 311
	C	4,849 ± 559	4,355 ± 483		4,081 ± 605
PlanktF:TP	B	1,007 ± 79	717 ± 43	*	721 ± 57
	HB	825 ± 80	535 ± 48	*	485 ± 47
	C	2,119 ± 158	715 ± 103	*	708 ± 135
Producers					
Zoop:Phyto	B	0.031 ± 0.004	0.034 ± 0.003		0.036 ± 0.004
	HB	0.031 ± 0.003	0.057 ± 0.004	*	0.061 ± 0.005
	C	0.052 ± 0.007	0.069 ± 0.008		0.076 ± 0.010
Zoop:Phyto+Bact+APP	B		0.026 ± 0.003		0.027 ± 0.003
	HB		0.039 ± 0.002		0.039 ± 0.002
	C		0.050 ± 0.008		0.053 ± 0.008
Daph:Phyto	B	0.011 ± 0.002	0.011 ± 0.002		0.011 ± 0.002
	HB	0.007 ± 0.001	0.019 ± 0.002	*	0.019 ± 0.003
	C	0.007 ± 0.001	0.017 ± 0.003	*	0.018 ± 0.004
HNF+Ciliates:Bacteria+APP	B		5.536 ± 1.720		5.423 ± 1.820
	HB		0.350 ± 0.023		0.350 ± 0.023
	C		8.829 ± 2.089		9.283 ± 2.254
Zooplankton					
PlanktF:Zoop	B	0.256 ± 0.050	0.208 ± 0.023		0.207 ± 0.023
	HB	0.165 ± 0.030	0.099 ± 0.010	*	0.100 ± 0.013
	C	0.572 ± 0.111	0.190 ± 0.042	*	0.187 ± 0.052
PlanktF:Daph	B	0.940 ± 0.246	1.249 ± 0.371		1.091 ± 0.234
	HB	1.005 ± 0.282	0.363 ± 0.054	*	0.388 ± 0.068
	C	11.499 ± 6.965	1.111 ± 0.261		1.124 ± 0.303
CarnZoop:Zoop	B	0.032 ± 0.003	0.031 ± 0.004		0.027 ± 0.004
	HB	0.035 ± 0.005	0.040 ± 0.004		0.034 ± 0.003
	C	0.098 ± 0.010	0.084 ± 0.007		0.077 ± 0.007
PiscF:Zoop	B	0.244 ± 0.163	0.294 ± 0.183		0.288 ± 0.045
	HB	0.128 ± 0.098	0.101 ± 0.043		0.102 ± 0.012
	C	0.408 ± 0.441	0.469 ± 0.675		0.497 ± 0.203
Fishes					
PlanktF:TotalFish	B	0.564 ± 0.049	0.423 ± 0.021	*	0.432 ± 0.027
	HB	0.604 ± 0.046	0.489 ± 0.020	*	0.492 ± 0.023
	C	0.688 ± 0.066	0.391 ± 0.042	*	0.418 ± 0.052
GizzSh:Yperch	B	2.957 ± 0.699	0.355 ± 0.121	*	0.434 ± 0.158
	HB	1.324 ± 0.343	0.088 ± 0.048	*	0.105 ± 0.064
	C	0.803 ± 0.257	0.008 ± 0.005	*	0.009 ± 0.006
PiscF:PlankF	B	1.080 ± 0.830	1.600 ± 1.165		1.612 ± 0.316
	HB	0.909 ± 0.780	1.147 ± 0.536		1.133 ± 0.131
	C	0.985 ± 1.354	2.663 ± 3.011	*	2.663 ± 0.806
Benthos					
Benthos:Phyto	B	1.052 ± 1.582	0.237 ± 0.527		2.034 ± 0.754
	HB				
	C	10.471 ± 271.867	1.590 ± 64.764	*	
Zoop:Benthos	B	0.066 ± 0.051	0.051 ± 0.011		0.028 ± 0.006
	HB				
	C	0.007 ± 0.007	0.007 ± 0.003		

Table 2.4.2. *Comparison of select trophic ratios across Lake Ontario Areas of Concern and reference sites. Phytoplankton and zooplankton biomass are as in Table 2.4.1, but for comparison of planktivorous fishes to zooplankton (PlanktF:Zoop) biomass ratio, estimates from electrofishing were used because trawl and gillnet data are not available for other AOCs.*



		Bay of Quinte		
		Turbid	Clear	Recent
Ratio	Site	72-94	95-18	01-18
Phyto:TP	B	196,642 ± 22,149	130,675 ± 9,560	120,577 ± 8,054
	HB	179,166 ± 24,730	126,026 ± 7,636	98,026 ± 6,599
	C	134,730 ± 25,513	57,680 ± 5,474	55,376 ± 5,375
Zoop:Phyto	B	0.028 ± 0.004	0.034 ± 0.003	0.036 ± 0.004
	HB	0.056 ± 0.030	0.048 ± 0.004	0.061 ± 0.005
	C	0.038 ± 0.011	0.071 ± 0.010	0.076 ± 0.010
PlanktF:Zoop	B	5.508 ± 1.661	8.980 ± 1.446	9.907 ± 1.433
	HB	0.993		
	C	18.990		



	Toronto Harbour		Hamilton Harbour		L. Ontario	
		mean		mean		mean
Ratio	Area		Station		Station	
Phyto:TP	HU	25,009 ± 1,701	HH258	59,559 ± 4,714	BUR	43,092 ± 4,843
	IH	41,469 ± 8,284	HH6	35,512 ± 20,512	LO2	37,939 ± 6,109
	AB	25,843 ± 8,679	HH8	46,291 ± 3,223	LO81	81,315 ± 8,076
Zoop:Phyto	HU	0.144 ± 0.115	HH258	0.142 ± 0.021	BUR	0.056 ± 0.021
	IH	0.062 ± 0.012	HH6	0.282 ± 0.112	LO2	0.033 ± 0.006
	AB	0.147 ± 0.054	HH8	0.152 ± 0.014	LO81	0.057 ± 0.007
PlanktF:Zoop	HU	11.202 ± 4.142	HH258	8.921 ± 1.026	BUR	
	IH	57.618 ± 7.086	HH6	5.621 ± 0.802	LO2	
	AB	5.270 ± 1.171	HH8	8.679	LO81	
			HH908	9.830 ± 2.524		

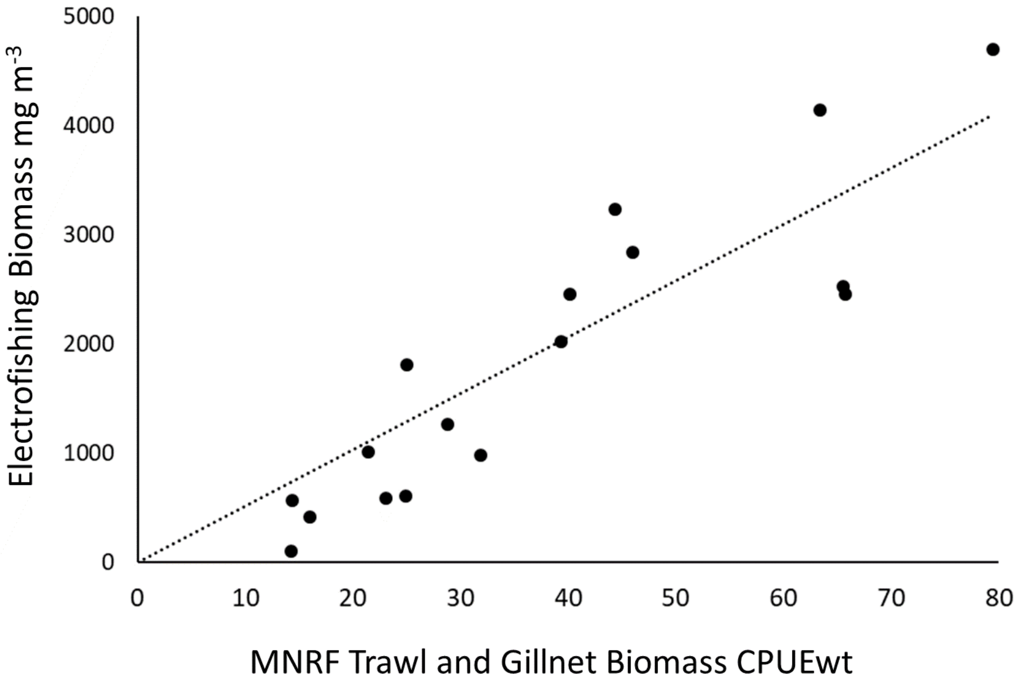


Figure 2.4.1 *Regression of the planktivorous and total fish weights from DFO electrofishing surveys MNRF Trawl and Gillnet fish data from Bay of Quinte sites. Includes data from 9 years between 1989-2015. The equation derived is eFish (mg* m^{-3}*) = 51.621 * MNRF (CPUEwt),* $r^2 = 0.79$*,* $p<0.001$*.*

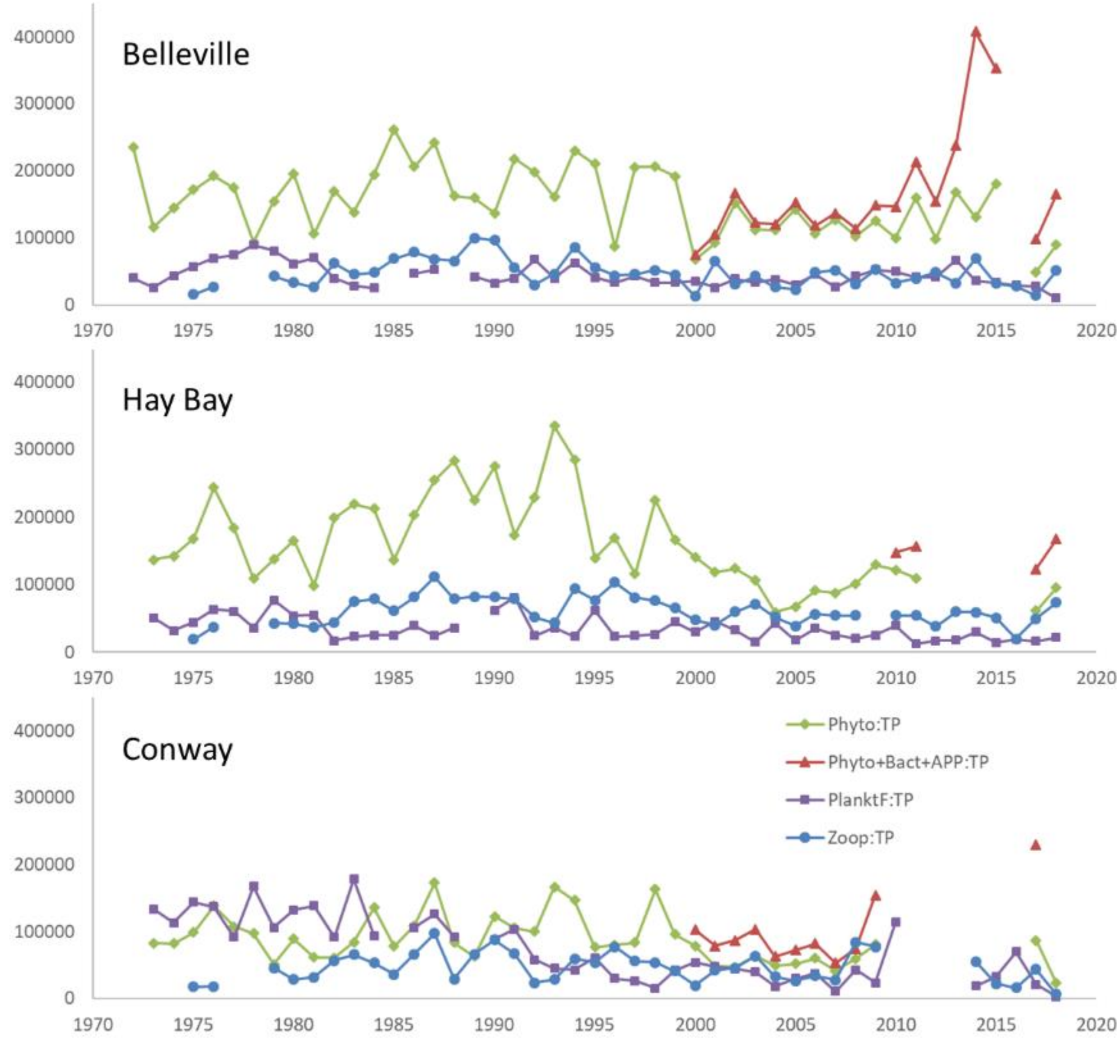


Figure 2.4.2 *Long term wet-weight biomass to total phosphorus (TP) ratios for the Bay of Quinte sites. Ratios are for phytoplankton (Phyto:TP), phytoplankton + microbial loop (Phyto+Bact+APP:TP), zooplankton (Zoop:TP) and planktivorous fishes (PlanktF:TP). Zooplankton biomass is converted to wet-weight assuming 10% dry-weight and MNRF fish CPUEwt biomass is converted using relationship in Fig. 2.4.1.*

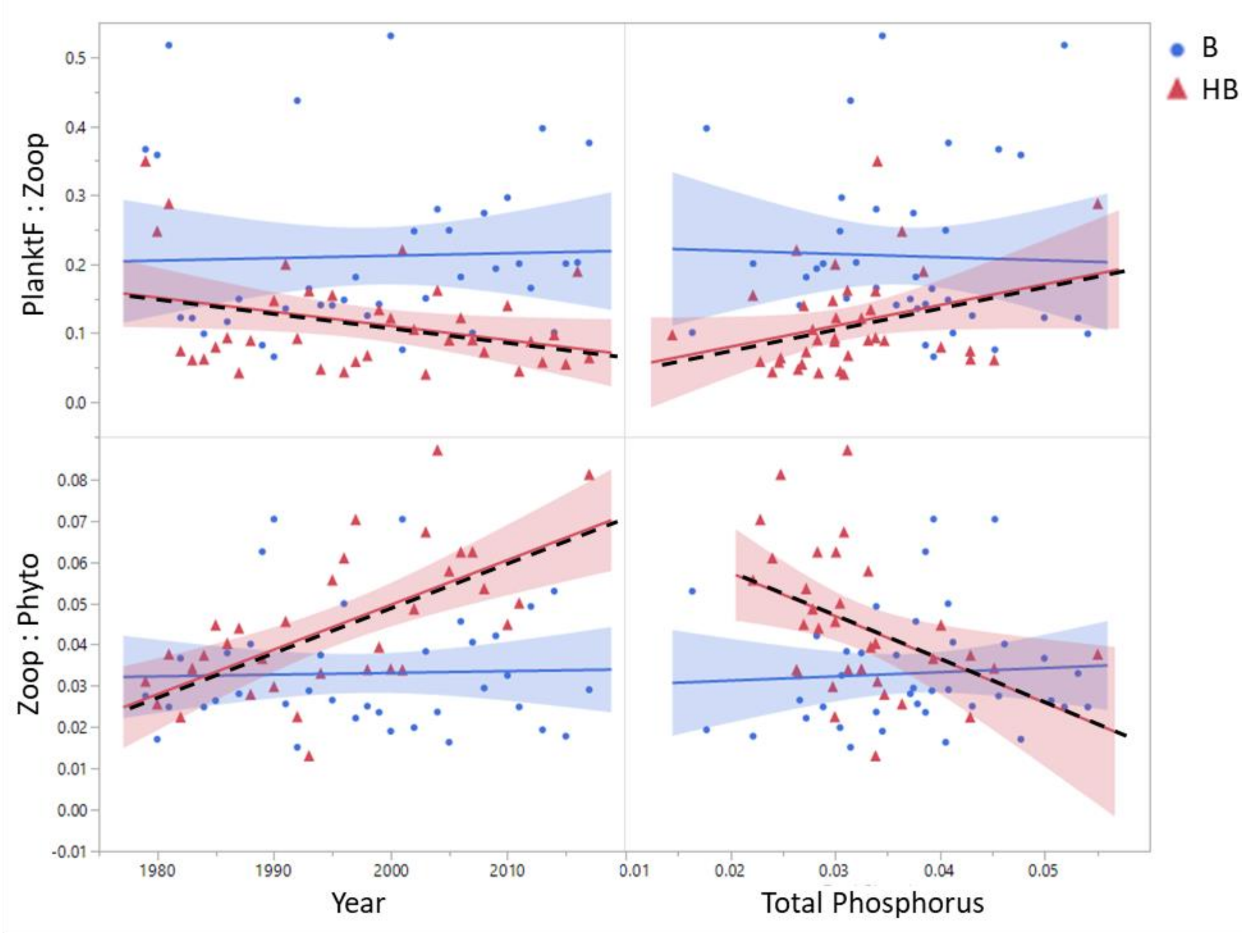


***Figure 2.4.3** Long term zooplankton (DW mg m^3) to phytoplankton (WW mg m^3) (Zoop:Phyto) and planktivorous fish (CPUEwt) to zooplankton (DW mg m^3) (PlanktF:Zoop) biomass ratios for the Bay of Quinte Belleville and Hay Bay sites. Dashed black lines are significant p<0.05.*

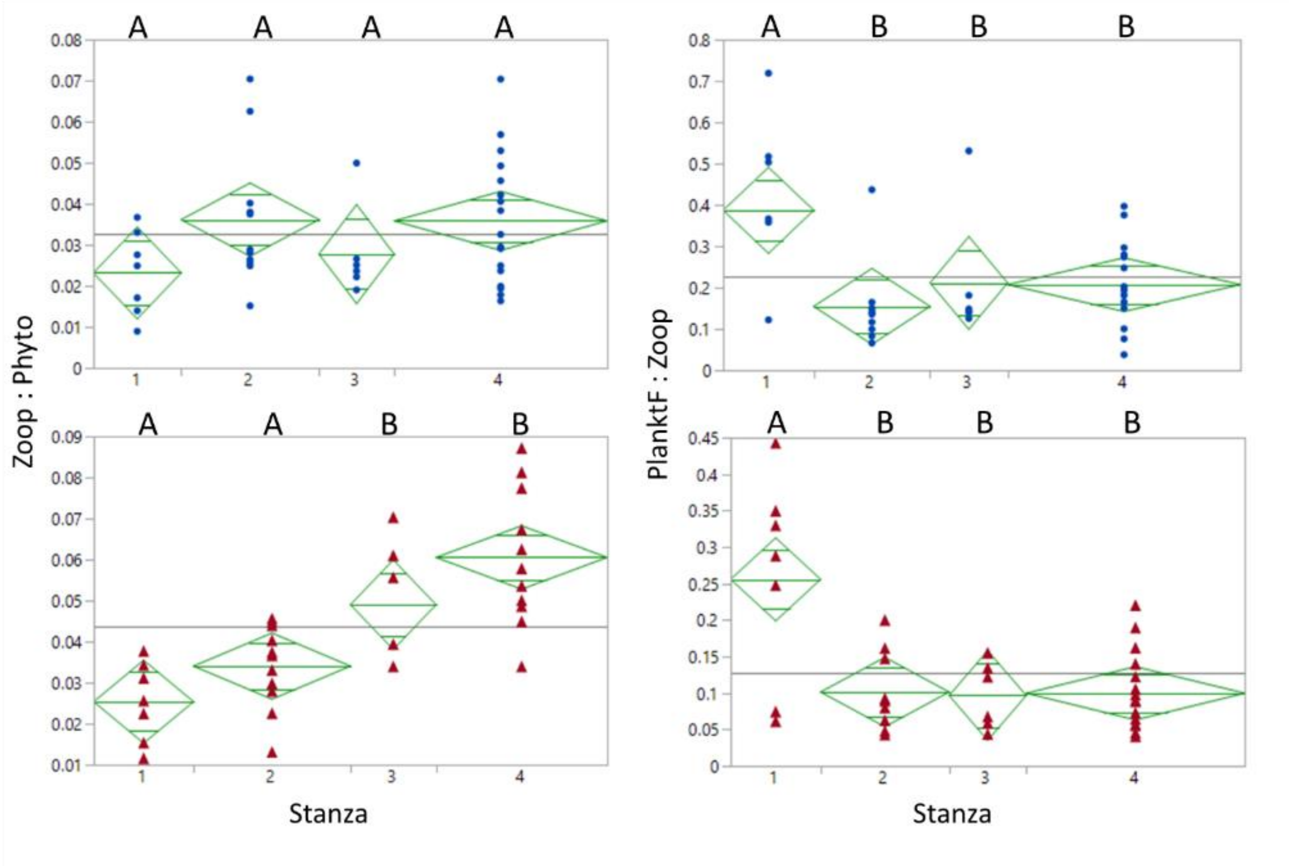


Figure 2.4.4 *Time-stanza ANOVA results for zooplankton (DW mg m^3) to phytoplankton (WW mg m^3) (Zoop:Phyto) and planktivorous fish (CPUEwt) to zooplankton (DW mg m^3) (PlanktF:Zoop) biomass ratios for the Bay of Quinte Belleville (top, blue dots) and Hay Bay (bottom, red triangles) sites. Time stanzas are 1972-1982, 1983-1994, 1995-2000, 2001-2018. Letters at top indicate samples with no significant difference.*

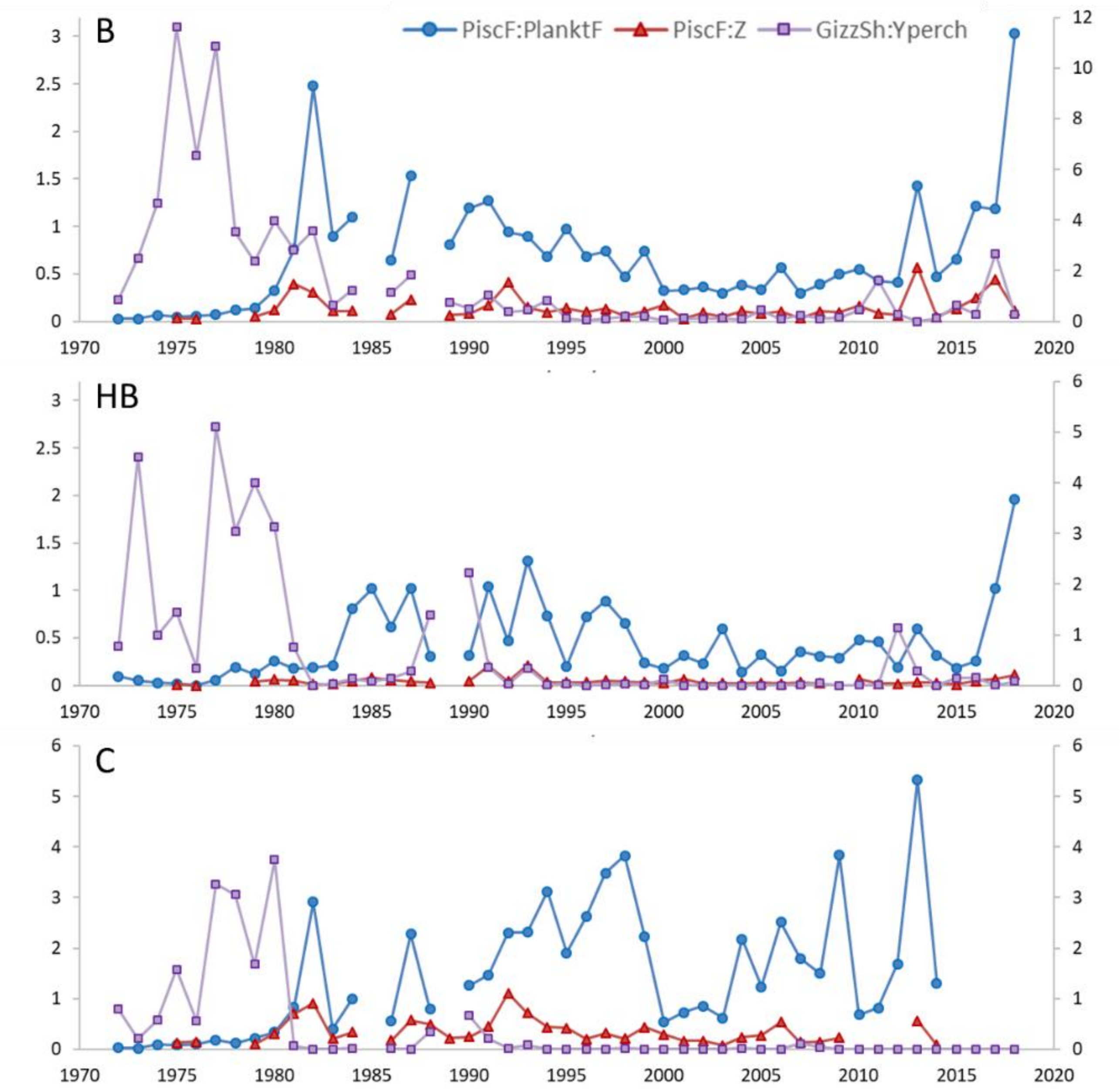


Figure 2.4.5 *Trophic ratios based on combined MNRF Trawl and Gillnet (CPUEwt) from the Bay of Quinte upper bay (Belleville, B), middle bay (Hay Bay, HB) and lower bay (Conway, C). Piscivore group are based on MNRF classifications. Ratios are piscivorous fishes to planktivorous fishes (PiscF:PlanktF blue circles), piscivorous fishes to zooplankton dry-weight (mg m^{-3} red triangles) on primary axis and Gizzard Shad to Yellow Perch biomass (GizzSh:Yperch purple squares, see discussion in S1.1) on right axis. Note changes to y-axis for Conway (primary) and Belleville (secondary).*

Final Assessment of BUI 13 and Recommendations

Phytoplankton

As with the nutrient parameters, there is a decreased range of biomass of phytoplankton in later years, but the Bay of Quinte still has the greatest production of biomass per unit total phosphorus than any other site measured by DFO except for Lake Erie. At all stations phytoplankton are dominated by Diatoms followed by Cyanobacteria. These phytoplankton are primarily composed of filamentous forms with the diatom *Aulacoseira* (previously *Melosira*) the top taxa overall contributing almost 50% of the biomass. *Dolichospermum* (formerly *Anabaena*) is a filamentous cyanobacteria capable of producing toxins that has contributed 1 to 25% of the annual mean but is less dominant after the mid-1990s. Filamentous algal forms are common in shallow eutrophic environments because they are constantly mixed back up into the euphotic zone by resuspension despite their tendancy to sink (Scheffer et al. 1997). Filamentous forms are more difficult for filter feeding zooplankton to handle so they are generally not preferentially grazed within the food web.

Because of this dominance of filamentous forms, <u>Total Filamentous</u> algae is proposed as a metric of both bottom-up driver of phytoplankton biomass and as factor which limits energy transfer to zooplankton in the food web. When selecting metrics the best models included TKN (reflecting the importance of nitrogen limitation) as highly significant reinforcing the nutrient connection and management control. Other significant factors were light attenuation, pre or post 1983 and winter precipitation. Total Filamentous is easy to enumerate in that it requires limited taxonomic expertise and relatively little time compared to evaluation of the whole phytoplankton community. Given the original eutrophication target of 5 g m^{-3} (annual mean) for total phytoplankton biomass and the 2017 recommendation of 50% nuisance taxa; the proposed target of 2.0 g m^{-3} (slightly less than 50% of total to account for colonial nuisance taxa) is appropriate as a Total Filamentous target.The algae from the family *Chrysophyceae* are important because they are viewed as an ideal, preferrentially selected for food item for zooplankton. The *<u>Chrysophyceae ratio</u>* gives the relative compostion of quality algal food, so it is the next most important indicator describing the phytoplankton data set and is a good counter indicator as it moves in the opposite direction of Total Filamentous with higher values indicating an improved phytoplankton community. A target of over 10% should be expected for the upper Bay of Quinte.

While the first recommended metric is relatively simple and could be accomplished with minimal training, the second and third metrics require more detailed taxonomic knowledge (ID of Chrysophyceae and biomass of colonial cyanobacteria) but a single individual with a moderate level of taxonomic training could make these assessments.

With these limited taxonomic assessments, we propose the following indicators for Target 1, Decrease in nuisance, eutrophic and noxious indicator species:

I. <u>Total filamentous biomass less than 2.0 g m^{-3}</u>
II. <u>Biomass of colonial cyanobacteria less than 1 g m^{-3} more than 80% of the time</u> (with a minimum of 6 samples collected evenly through the season)

Likewise, for Target 2, Increase in phytoplankton as a food resource for zooplankton grazers:

III. <u>% Chrysophyceae of Total Filamentous phytoplankton greater than 10%</u>

The tight connection between these phytoplankton metrics and nutrient levels means it is expected that further reductions in nutrient loadings would further benefit phytoplankton populations. Kim et al. (2013) show that P dynamics are primarily driven by river inflow in the upper bay although there is significant internal loading in the shallow waters of the upper bay that affect nutrient conditions in the mid and lower bay. Doan et al. (2018) argue that internal P loading is relatively small compared to external loads (i.e. Trent River) which reinforces the need for management actions that continue to reduce P, or at minimum maintain loadings to the bay. Doan et al. (2018) also show that internal loads are delivered in summer and early fall when algal blooms are most common. This suggests that additional management measures to reduce the internal P cycling are also necessary to reduce algal blooms in the near term. In the absence of additional internal loading controls and the continued reduction in external nutrient loads, only time will see the gradual improvement to phytoplankton populations as nutrients currently stored in the sediments are either released or sequestered to non bioavailable forms.

Management options have been tested to reduce internal P loadings in lakes and an excellent review is given in Lewtas et al. (2015). Possible actions include dredging and removal of sediment although the largest successful projects (1,000 hectares) are much smaller than the upper bay and even the shallow upper bay (~5m) is deeper than the deepest successful project (<3m).

Sediment capping can interrupt P recycling from highly organic sediments like those in Quinte. Physical capping agents such as sand, gravel or clay are used to create a layer over 5 cm thick which would block sediments from interaction with the water column. Given the very soft and deep sediment found in the Bay of Quinte (Maria Dittrich, University of Toronto, *pers.com.*) there would likely be additional material required to cap effectively. It would be costly due to the large volume of material required and because of the highly negative impact on benthos in the short and long term.

Several elements have been tested as inactivators of P within the sediment including iron (Fe), aluminum (Al), calcium (Ca), zeolite, Phoslock, and modified clays and other items. Alum addition is not well suited as it is flocculent and is resuspended in shallow energetic lakes like Bay of Quinte. Phoslock is a commercially available lanthanum modified bentonite clay with high P sorption and can be used under many condidtions. So far it has only been used in lakes much smaller than the Bay. It is a potential toxin and the ecological impacts are not understood. Calcium carbonate is used extensively in hard water lakes like Bay of Quinte though again at a much smaller scale and has fewer toxic impacts than alum. It can be used in deep lakes but it has only been successfully applied in lakes less than 240 hectares, much smaller than the Bay. It mimics natural whiting events in hardwater lakes and has the potential to smother benthos. Doan et al. (2018) suggest targeted addition of calcium hydroxide [$Ca(OH)_2$] in eutrophication hot spots in the Bay of Quinte to promote increased precipitation and burial of excess P. Because of the large size of the bay widespread use is likely to be expensive.

Other P sorbants include *Phragmites* detritus, which is also a promising sediment remediation material with the added benefit of watershed level invasive species removal (Tang et al. 2020).

Lürling and Mucci (2020) review measures used to reduce Cyanobacteria in shallow lakes. Nitrogen additon has also been proposed to shift the ratio of N:P in P enriched environments and reduce the amount of nitrogen fixing phytoplankton. It is unknown if this would improve the overall community at Quinte because while some bloom forming taxa are N fixers (e.g.

Dolichospermum), the majority of the time the main bloom forming taxa are filamentous diatoms (e.g. *Aulacoseira*) which are known to be competitive under a range of N conditions. Reducing nitrogen limitation in of itself is unlikely to lead to improvements in the phytoplankton-based metrics since biomass would likely increase. Smaller-scale mesocosm experiments may help to guide this management action.

Our analysis shows that focusing on key measures of total filamentous phytoplankton, colonial cyanobacteria and Chrysophyceae will provide adequate data on the phytoplankton community for the assessment of BUI 13. We caution however, that a simplified data set of this nature will be of limited value outside of the scope of this portion of Bay of Quinte BUI 13, and may not be applicable to trophic ratios requiring total phytoplankton biomass or assessments of other ecosystem level stressors that may emerge.

Microbial Plankton

Bacteria regenerate large amounts of energy and can provide an important food resource for zooplankton and benthos which in turn supports fisheries. However large accumulations of bacteria are likely indicative of ecosystem stress which extends into public health. Our analysis shows bacteria biomass in the upper bay has been increasing since 2009; the highest levels were observed over the last 5 years of study (2014 – 2018) and coincided with record low levels of zooplankton and piscivorous fish. Here we specify a target of bacterial biomass less than 920 mg m^{-3}. We therefore recommend continued monitoring of bacteria in conjunction with public health units.

In the event that specific remedial actions are desired to reduce the bacterial standing stock, we would recommend first that an in-depth study of bacterial populations be undertaken using microbial source tracking techniques to identify the sources of bacteria, both physical (e.g. sediments, sewage, runoff) and biological (e.g. human, avian, etc.). Such techniques are not novel and have been deployed by Environment and Climate Change Canada in other AoCs including Toronto Harbour and Hamilton Harbour (Staley et al. 2018; Edge and Hill, 2007). Obtaining this information is a necessary first step towards identifying suitable and achievable remedial actions.

Zooplankton

Bottom-up forces related the the eutrophication of the Bay of Quinte still dominate the drivers of zooplankton biomass in the Bay of Quinte. *Chydorus* biomass and the ratio of *Eubosmina* to *Bosmina* are both positively related to increasing eutrophy, and unusually high levels of either one are indicative of deteriorating health of the zooplankton community, at least as it relates to excess nutrients. We recommend *Chydorus* biomass < 7.5 mg m^{-3} dry-weight be maintained. Top-down indicators of increasing fish planktivory are currently less robust in the upper Bay of Quinte, given that the system is dominated by bottom-up influences. The percentages of both *Daphnia galeata mendotae* and the large cyclopoid *Mesocyclops* may have utility in identifying unusually high levels or changes to planktivory, although both are less sensitive to responding to more moderate levels of predation. To ensure sufficient forage for planktivores, we find the percentage of *Daphnia galeata mendotae* greater than 16% can be used as an indicator. Further, the normalized ratio of *Eubosmina* : (DGM + *Eubosmina*) less than 0.52 will be sensitive to increased top-down effects, with an expectation that as bottom-up influences lessen in the upper Bay, these metrics should improve in effectiveness as they have in the lower bay.

In recent years, declining zooplankton populations in the upper bay have the potential to limit production and survival of fish species that depend on zooplankton for survival, especially juvenile life stages. Any actions that target altering the fish community should be aware of this.

Trophic Ratios

Trophic biomass ratios are very useful in simplifying complex food web interactions to a simple quantitative metric. When biomass data is available across trophic levels it permits the identification of disruptors within the food web both from bottom-up and top-down effects. The Bay of Quinte also has measurements from other food sources such as the benthos which have been included in the analysis to guide future analysis.

We propose 3 metrics for use in a montoring plan: The first is the bottom-up metric of phytoplankton biomass to total phosphorus (Phyto:TP). The Bay of Quinte has the highest level of phytoplankton biomass produced per unit of TP of any site measured by DFO, even when compared to another eutrophic AOC. This is driven in part because of the dominance of filamentous forms of phytoplankton which have a higher biomass signature than unicellular forms. We are proposing the reduction of the Phyto:TP less than 50000 to be on par with the (eutrophic) Hamilton Harbour which should occur as a more balanced community of phytoplankton is produced with continued reduced TP. It is expected that the total phosphorus concentration in the upper bay will continue to decline steadily. If the Phyto:TP biomass ratio does not decline to these levels, then it should be determined where the phytoplankton are sourcing their nutrients to target declines in bloom occurance.

The second metric is the zooplankton:phytoplankton (Z:P) biomass ratio. This metric has the advantage of being driven both by bottom-up and top-down forcings since zooplankton occupy an intermediate position in the food web. This metric has been widely applied globally as an indicator of ecosystem function. The Z:P ratio has improved in both lower and middle bay sites, but has not changed in the upper bay. There is an expectation that this ratio should have shown improvement at Belleville since the TP and phytoplankton biomass has decreased. This is likely due to the dominance of poor food-quality filamentous forms of phytoplankton. Because the Z:P ratio has not increased, this results in less trophic transfer to the upper food-web, limiting fish biomass and putting at risk improvements to piscivores in the fish community. As such we propose a Z:P target greater than 0.062 (based on improvements seen in middle bay). Remedial actions including nutrient controls will likely drive any improvement in this metric, but changes to the fish community structure by management could also change the Z:P ratio.

The third metric is the top-down based ratio of planktivorous fishes to zooplankton biomass (PlanktF:Z). In the Bay of Quinte, this ratio is still fundamentally driven by dominance of bottom up forcing, particularly as seen with TKN, but also TP, and as such is a very good predictor of total fish biomass as well. Top-down effects currently are not strong in the upper bay (see Z:P) but as conditions improve, it should also show up in this metric. All sites showed an initial decline in PlanktF:Z during the 1970s to early 1980s and while improvements are seen at lower and middle bay sites, the upper Bay of Quinte showed no significant changes in the ratio of planktivorous fishes to zooplankton since the early 1980s. Based on the long-term mean, we recommend a target for PlanktF:Z less than 0.22 for the upper Bay of Quinte. However, many of the dominant fish species in the Bay of Quinte are at least in part benthivorous (e.g. Walleye, Yellow and White Perch). This forage biomass is not accounted for in the Z:P or PlanktF:Z ratios, in part, because benthos has not been actively sampled since 2011. This is of particular concern since we have seen an increase in benthos diversity as phosphorus has decreased (see Fig. 1.15). Given the changes in zooplankton to fish linkages in recent years, it is recommended that benthos sampling programs be reinstated as part of any future monitoring plan.

Other relevant early warning metrics:
We are proposing two fish-based metrics of food-web function that may act as early warning indicators of undesirable change in the Bay of Quinte. Plankton biomass changes very quickly, but fish biomass is lagged over many years. Individual zooplankton may live for a couple of days/weeks, but predatory fishes can live for years to decades.

The relative percentage of Gizzard Shad to Yellow Perch biomass simplies a functionally similar detritivore/planktivore to one that is more benthivorous/piscivorous. There have been overall decreases in the GizzSh:YPerch ratio observed at all the sites which shows improvement. However there have been recent increases in Gizzard Shad in the upper and middle bays possibly related to the increased bacterial biomass since 2009 in the upper bay indicating recent decline. There are also historic lows in planktivore and total fish biomass in the upper bay since 2013 so we recommend this ratio be used as an early warning warning metric for more intensive monitoring of GizzSh:(YPerch+GizzSh) less than 0.2 based on combined sites mean + 2 standard deviation.

Similarly, we recommend the use of the piscivore/(planktivore+piscivore) ratio (PiscF:PlanktF) and the piscivore/(zooplankton+piscivore) ratio (PiscF:Z) for monitoring early warning signals. Because of the lack of improvement in the Z:P ratio in the upper bay, the biomass of piscivores may not be able to be supported by their forage supply. Since 2013 there has been a relative increase in piscivores and a subsequent drop in planktivorous fishes but no corresponding increase in zooplankton biomass. As such, energy being transferred up the food-chain is being disrupted. If this continues, there could be a collapse in predatory fishes biomass in the upper Bay of Quinte so we recommend the continued monitoring of fish populations and condition measures such as gonadal somatic index etc. PiscF:PlanktF values greater than 1 and elevated ratios of PiscF:Z greater than 0.15 should be used to recommend more intensive monitoring.

TARGET	**Target Met?**	**Rate Met 2001-2018**
Total Filamentous algal biomass less than 2.0 g m^{-3}	**No**	5/17 (29%)
Colonial Cyanobacteria less than 1 g m^{-3}, 80% of time	**Partially**	10/17 (59%)
Ratio Chrysophyceae : Total Filamentous greater than 10%	**No**	2/17 (12%)
Bacterial biomass less than 920 mg m^{-3}	**No**	10/18 (56%) †
Chydorus biomass less than 7.5 mg m^{-3} dry-weight	**Yes**	15/18 (83%)**
% *Daphnia galeata mendotae* greater than 16%	**No**	7/18 (39%)
Eubosmina : (DGM + *Eubosmina*) less than 0.52	**No**	5/18 (28%)
Phytoplankton : TP less than 50000	**No**	0/18 (0%)
Zooplankton : Phytoplankton greater than 0.062	**No**	1/18 (6%)
Planktivorous Fishes : Zooplankton less than 0.22	**Partially**	11/18 (61%)

** biomass per TP value lower than expected at B indicating production impairment
† failed to meet target every year since 2011

Additional early warning metrics of ecosystem change based on fish community data:

TARGET	Target Met?	Rate Met 2001-2018
Gizzard Shad : (Yellow Perch + Gizzard Shad) less than 0.2	**No**	9/18 (50%)
Piscivores : Planktivores less than 1	**?**	4/18 years ‡

‡ Not met years (4/6) all occurred during last 6 years 2013-2018

Based on all of the metrics above in a weight of evidence approach, our evaluation of BUI 13 in the upper Bay of Quinte is that while there have been some long-term improvements, it continues to be significantly impacted in its food web functioning. It is therefore our recommendation that the status of BUI 13 should continue to be *impaired*.

Monitoring Plan Recommendations

Given the ongoing impairment of BUI 13, we recommend continued monitoring of the upper and middle Bay of Quinte and also recommend including Conway in the lower bay. At a minimum we propose monthly sampling of zooplankton, phytoplankton and bacteria. But given that it will introduce a level of uncertainty in the data, especially for phytoplankton and bacterial measures during the summer period, we highly recommend fortnightly sampling. It is important to maintain certain environmental measures are collected simultaneously including a YSI EXO sonde cast (or similar device), Secchi disk (light attenuation measures are preferred) and water chemistry (including total phosphorus, nitrogen and dissolved nutrients). Our most recent study showed few differences between nearshore and offshore stations in the upper and middle bay. As a consequence, if the long-term monitoring sites of Belleville, Napanee and Hay Bay cannot be maintained, then other sites closer to shore will likely be comparable, although river mouths should be avoided. Due the recent shifts in fish community composition since 2013, it is highly recommended that food web analysis of piscivore production be reexamined since there are indications that fish populations have become food limited. As such, we highly support the continued trawl and gillnet monitoring of fishes by MNRF in the Bay of Quinte and recommend additional sampling and diet analysis of larval fishes and YOY especially in the nearshore.

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