#### 2020 Status of the Lake Ontario Lower Trophic Levels<sup>1</sup>

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# Significant Findings for Year 2020: Note that due to covid-19 restrictions, offshore sampling was limited in 2020.

- May Oct total phosphorus (TP) in 2020 was 10.6 μg/L (offshore) and 7.7 μg/L (nearshore), higher than the long-term (1995-2019) average in the offshore (6.2 μg/L) and close to average in the nearshore (7.8 μg/L); mean TP values for the past decade (2010-2019) were 6.0 μg/L and 7.9 μg/L in the offshore and nearshore, respectively. In 2020, TP concentrations were significantly higher (p=0.03) in the offshore compared to the nearshore. Note that offshore duplicate samples had high relative percent difference (average 54%. 6-117%) making inferences for the offshore in 2020 uncertain.
- 2) May Oct epilimnetic chlorophyll-*a* was similar at nearshore (1.9  $\mu$ g/L) and offshore (2.1  $\mu$ g/L) sites. These values were slightly higher than the average for 1995 2019 (1.7  $\mu$ g/L, offshore; 1.5  $\mu$ g/L, nearshore) and higher than for the last decade (1.4  $\mu$ g/L, both offshore and nearshore).
- 3) May Oct Secchi depth ranged from 3.5 m to 10.4 m (11 ft to 34 ft) at individual sites and was not significantly different between nearshore (6.3 m; 20.7 ft) and offshore (6.5 m; 21.3 ft) locations. Long-term (1995-2019) average was 7.2 m in the offshore and 6.4 m in the nearshore; means for the last decade were 7.8 m in the offshore and 6.2 m in the nearshore
- 4) Despite higher TP values in 2020 than in recent years, TP, chlorophyll-*a* and Secchi depth are indicative of oligotrophic conditions in the offshore of Lake Ontario.
- 5) Nearshore summer zooplankton biomass was 10.5 μg/L, near the all-time low (9.4 μg/L, 2017) since monitoring began in 1995. Offshore epilimnetic summer zooplankton biomass was 11.7 μg/L. These values are similar to biomass in the last decade (2010-2019).
- 6) Peak (July) epilimnetic biomass of *Cercopagis* was 3.0 μg/L in the nearshore and represented 25% of the zooplankton community at that time; *Cercopagis* was absent from the July offshore epilimnetic samples in 2020 but was present in whole water column samples taken in August by other agencies. Epilimnetic biomass of *Bythotrephes* peaked in late-September in both the nearshore (1.0 μg/L) and offshore (1.8 μg/L) and represented 10% and 18% of the zooplankton community at those times, respectively.
- 7) Summer nearshore and offshore epilimnetic zooplankton density and biomass declined significantly 1995 2020. The declines were due mainly to reductions in cyclopoid copepods in both habitats.

<sup>&</sup>lt;sup>1</sup>Any use of trade, firm, or product names is for descriptive purposes only and does not imply endorsement by the U.S. Government.

#### Introduction

This report presents data on the status of lower trophic levels of the Lake Ontario ecosystem (zooplankton, phytoplankton, nutrients) in 2020 collected by the US Biological Monitoring Program (BMP). The BMP is a collaborative project that, in 2020, included the New York State Department of Environmental Conservation (NYSDEC) Lake Ontario Unit and Regions 6, 7, and 8 at Watertown, Cortland, and Avon (Lake Ontario regions); the U.S. Fish & Wildlife Service (USFWS) Lower Great Lakes Fishery Resources Office; the U.S. Geological Survey (USGS) Lake Ontario Biological Station; and Cornell University. Covid-19 restrictions limited sampling in 2020, especially for the offshore, making inferences about changes in the offshore of the lake more uncertain. However, we were able to collect comparable data to past years for most nearshore sites.

The BMP has collected data on several ecological indicators since 1995 in both the offshore and nearshore of Lake Ontario. These indicators include total phosphorus (TP), soluble reactive phosphorus (SRP), chlorophyll *a* (chl-*a*), Secchi depth (SD), and crustacean zooplankton (density, biomass, species composition, and size structure). In 2020, samples were collected from May through October.

Trophic level indicators for 2020 are compared with data collected by this program since 1995 and with similar long-term data from other Production at lower trophic levels sources. determines Lake Ontario's ability to support prey fish populations upon which both wild and stocked salmonines depend. Alewife, in particular, appear to be sensitive to declines in lower trophic level resources. Such declines are considered a main cause for the collapse of the alewife population and decline in Chinook Salmon fishery in Lake Huron after 2003 (Barbiero et al. 2011, Bunnell et al. 2012), although increased predation and winter severity may also have contributed to alewife declines (Dunlop and Riley 2013, He et al. 2015). Similarly, declines in lower trophic level productivity in Lake Michigan are coupled with lower alewife abundance which may affect the Salmon fishery in lake. Chinook that

Zooplankton biomass in both lakes Huron and Michigan have continued to decline through the second decade of this century, and both species composition and biomass in 2018 and 2019 are similar to observations in Lake Superior (Barbiero et al. 2019, Rudstam and Watkins, unpubl data). The connection between nutrient loading and fish production remains an important research topic in the Great Lakes (Stewart et al. 2016).

In the "State of Lake Ontario 2014", Rudstam et al. (2017) summarized data from various sources including the BMP and analyzed trends through Total phosphorus and chlorophyll-a 2013. declined and Secchi depth increased from 1980-1995 with limited change from 1995-2013. However, Dove and Chapra (2015) reported a continued decline in spring TP into the 2000s based on data from Environment & Climate Change Canada's (ECCC) Surveillance program. Therefore, we are especially interested in any evidence of further decreases in lower trophic level indicators as we incorporate the 2020 data in our trend analyses. Zooplankton populations have been more variable, likely due to the interplay between vertebrate and invertebrate predators, increased water clarity, lower epilimnetic production, and increased deep chlorophyll layers (Rudstam et al. 2015, Barbiero et al. 2014, 2019, Scofield et al. 2020).

## **Report Objectives**

Using data from 1995 to 2020, the following questions were addressed:

- (1) What is the status of Lake Ontario's lower trophic levels in 2020, and what differences exist between nearshore and offshore sites this year?
- (2) How does the year 2020 compare to the same indicators in 1995-2019 (using BMP data and other long-term data)?
- (3) What is the status of the two non-native predatory cladocerans *Bythotrephes* and *Cercopagis*?
- (4) Are there changes in zooplankton community structure (biomass, size, species composition) that could be indicative of changes in alewife or invertebrate predation, or lake productivity?

#### Methods

Total phosphorus (TP), soluble reactive phosphorus (SRP), chlorophyll-a (chl-a), Secchi depth (SD), and zooplankton density, size, and biomass by species were measured at offshore and nearshore sites in Lake Ontario (Figure 1). Samples were collected from seven nearshore sites biweekly, although only three stations were sampled in all 11 sampling weeks from May through early October 2020 (Table 1). The COVID-19 pandemic limited sampling especially at the start of the season (Table 1). A limited number of offshore samples were collected in May by Region 8 DEC, and in June, August, September, and October by the R/V Kaho. Cape Vincent DEC sampled 2 offshore sites in June and July, but only TP/SRP, chl-a and SD were collected. Nearshore sites had depths ranging from 9.7 to 15.5 m (32 to 51 ft), and offshore site depths ranged from 17 to 207 m (56 to 689 ft). The August R/V Kaho samples are for zooplankton only and are on a transect of increasing bottom depths (15 m, 30 m, 50 m, and 100 m) offshore from Oswego, NY; the 15 m depth was included with nearshore samples while the other depths were considered offshore.

## Water Chemistry

Sampling

Water samples were collected for analysis of chl-a, TP, and SRP. Each sample was obtained by using an integrated water sampler (1.9 cm inside diameter Nalgene tubing) lowered to a depth of 10 m (33 ft) or bottom minus 1 m (3 ft) where site depth was 10 m or less. The tube was then closed off at the surface end and the column of water transferred to 2 L Nalgene containers. From each sample, a 100 mL unfiltered aliquot was frozen for later analysis of TP (APHA 1998; SM 4500-P). Two liters of water were filtered through a Whatman 934-AH glass fiber filter that was frozen for later analysis of chl-a using acetone extraction followed by fluorometry (USEPA 2013). A 100 mL aliquot of filtered water was frozen for later analysis of SRP (APHA 1998 SM 4500-P). TP and SRP samples were analyzed at the Upstate Freshwater Institute (UFI), an ELAP certified laboratory. Chl-a was analyzed at the Cornell Biological Field Station (CBFS) using a calibrated Turner Trilogy benchtop fluorometer and the USEPA standard operating procedure SOP LG 405

(Revision 9, March 2013). Approximately 2 L of water was filtered for each chl-*a* sample.

# Quality Control and Variability

To measure analytical precision at nearshore sites, replicate samples were obtained for TP and SRP. In July, six separate aliquots of water from the same water sample from four of the eight nearshore sites were frozen for TP and SRP analyses. To evaluate within site variability of TP, SRP, and chl-a, three separate water samples were collected in August at six of the eight nearshore sites. These triplicate samples were analyzed separately. At offshore locations, duplicate samples for TP, SRP, and chl-a were collected throughout the year. Quality control was assessed as the coefficient of variation (CV; standard deviation/mean x 100) for the nearshore samples (samples with more than 3 replicates) and as the percent relative difference (RPD; difference/mean x 100) for offshore duplicate samples.

# **Zooplankton**

Zooplankton samples were collected with standard 0.5 m diameter, 153-µm mesh, nylon nets equipped with calibrated flowmeters. At nearshore sites, tow depths ranged 9-12.8 m (30-42 ft). Tows started 1-2 m (3-7 ft) above the bottom depending on weather conditions. At offshore sites, epilimnetic tows were taken from the top of the thermocline to the surface ranging from 10 to 25 m (33-82 ft) when stratification was present or to 50 m during unstratified conditions. At offshore sites greater than 75 m (246 ft) bottom depth (in 2020, only one site on 2 different dates), one metalimnetic tow (50 m [164 ft] to the surface) and one hypolimnetic tow (100 m [328 ft] to the surface) were obtained in addition to the standard epilimnetic sample. Zooplankton were anesthetized with antacid tablets and then preserved in the field with 95% ethanol to obtain a final concentration of 70%.

In the laboratory, each sample was strained through a 1-mm mesh cup to separate *Cercopagis* and *Bythotrephes* from other zooplankton. This was done because *Cercopagis* and *Bythotrephes* form clumps in the sample, making the usual random subsampling of 1 mL samples impossible. For each sample that contained clumps of *Cercopagis* or Bythotrephes, two analyses were performed – one on the smaller zooplankton and one on the larger zooplankton (including *Cercopagis* and *Bythotrephes*) that were caught in the 1.0 mm mesh strainer. At least 100 larger zooplankton (or the whole sample) were measured and enumerated by sub-sampling organisms from a gridded, numbered Petri dish in which the sample had been homogeneously distributed. In some cases, different subsamples were used for *Bythotrephes* and *Cercopagis*. To calculate the total number of animals in the clumped part of the sample, a ratio of wet weights of the subsample to wet weights of the total sample was used.

For smaller-sized zooplankton, at least 100 organisms were counted and measured from one or more 1 mL sub-samples. The sub-sample was examined through a compound microscope at 10-40X magnification. Images from the sample were projected onto a digitizing tablet interfaced computer for measurements. with а Zooplankton were identified to species (except for nauplii and small copepodites) using Pennak (1978) and Balcer et al. (1984). Length:dryweight regression equations (CBFS standard set, Watkins et al. 2011) were then used to estimate zooplankton biomass.

## Data Analyses

May to October mean TP, SRP, chl-*a*, and SD were compared between the nearshore sites and the offshore epilimnion by first obtaining monthly means for each site and then fitting a general linear mixed model with month and habitat as categorical predictor variables. Due to the limited offshore sampling in 2020, we did not compare nearshore and offshores sites for the zooplankton variables.

For trend analyses and comparisons with the historical data we used the averages obtained from May, June, July and September for the offshore sites. The BMP does not typically sample offshore in August. Zooplankton were divided into the following six groups: daphnids (Daphnia mendotae, D. pulicaria, D. retrocurva, D. longiremis, D. schodleri, Ceriodaphnia sp.); bosminids (Bosmina longirostris, Eubosmina coregoni); calanoid copepods (Leptodiaptomus *Skistodiaptomus* oregonensis. minutus. sicilis, Leptodiaptomus Leptodiaptomus

ashlandi, Epischura lacustris, Eurytemora affinis); cyclopoid copepods (Acanthocyclops vernalis, Diacyclops thomasi, Mesocyclops edax, Tropocyclops prasinus); other cladocera Chydorus sphaericus, (Alona sp., Diaphanosoma sp., Polyphemus pediculus, Leptodora kindtii, Camptocercus sp., Scapholeberis sp., Ilyocryptus sp.); and nauplii. Four species were analyzed separately from the Those species are: groups. **Bythotrephes** longimanus; Cercopagis pengoi, Holopedium gibberum. and Limnocalanus macrurus. Differences were considered significant at p < p0.05. Regression analyses for time trends (JMP Pro v12.0.1, SAS Institute Inc. 2015) were performed for the offshore and nearshore for TP, SD, chl-a, summer epilimnetic zooplankton zooplankton density and biomass, and epilimnetic group biomass. Zooplankton metrics were log<sub>10</sub> transformed prior to analysis to reduce heterozygosity. Nighttime zooplankton data are not included in time trend analyses. Zooplankton in Lake Ontario migrate up in the water column at night, causing an increase in density and biomass in the epilimnion (Watkins et al. 2017); therefore, results from day and night are not comparable.

Change point analyses (Taylor Enterprises, Inc. 2003) were performed separately on nearshore and offshore data to test for breaks in the data. analysis Change point uses cumulative deviations from the mean to detect changes in time trends and to estimate when those changes occurred. This is done by resampling the data series 1000 times to construct confidence intervals based on the inherent variability in the data series and testing if and when the observed data series differ significantly from these confidence intervals.

## Results

## Quality Control and Variability

To estimate analytical precision (i.e. within sample variability), 24 TP and 24 SRP samples (4 sites x 6 samples per site) were analyzed. Coefficients of variation (CV=SD/mean) ranged from 5 to 37% (mean of 16%) for TP and from 8 to 15% (mean of 11%) for SRP. One value of 37  $\mu$ g/L from CBL was considered contaminated and removed from the analysis. Values from replicates were averaged by station-date for all analyses. Variation for SRP is smaller because many samples had concentrations below the detection limit of 0.6  $\mu$ g/L. In those cases, the sample was assigned the detection limit. This variability was similar to previous years.

The analysis of August nearshore TP, SRP, and chl-*a* triplicate samples showed that the CV for TP ranged from 2 to 18% (mean of 10%), the CV for SRP ranged from 0 to 54% (mean of 21%), and the CV for chl-*a* ranged from 2 to 15% (mean of 7%). Within site variability for TP was typical of the variation observed in previous years. Note that the variability among replicate samples in the field and variability resulting from laboratory procedures are similar. Values were averaged for later analyses.

Offshore duplicates were compared using RPD values. The average RPD was higher than other years, at 50% (range 6 to 117%). Twelve of the 17 duplicate samples failed a standard RPD value of 20%. We do not know the reason for this higher than normal variability between duplicates but note that this adds uncertainty to the offshore TP values presented in this report. This high variability does not affect the nearshore values. Values of duplicates were averaged for each site visit for later analyses.

#### 2020 Water Quality

May through October mean chl-a, TP, SRP, and SD were similar across nearshore sites in 2020 (Table 1). Chl-a was lowest at Niagara West (NWL; 1.3 µg/L) and highest at Sandy Pond (SPL; 2.6 µg/L) (Table 1). TP was highest at Niagara West (NWL; 9.0 µg/L) and lowest at Galloo Island (GIL; 6.0 µg/L). Niagara West also had the highest SRP (1.5  $\mu$ g/L). SD was lowest at Sandy Pond (SPL; 4.5 m [15 ft]) and highest at Galloo Island (GIL; 7.7 m [25 ft]). Nine Mile Point 15 m was not included in this comparison because only one sample was taken. Mean values from NEL, NWL, and OOL are biased because COVID-19 regulations restricted sampling to July through September. In the offshore, chl-a ranged from 0.4 µg/L (Oak Orchard-N) to 4.2 µg/L (Oak Orchard-O), TP ranged from 5.9 µg/L (Oswego Shallow) to 17.6 µg/L (Oak Orchard-O), SRP ranged from 0.8  $\mu g/L$  (three sites) to 1.6  $\mu g/L$  (Oswego Deep), and SD ranged from 3.5 m (11 ft; Nine Mile Point 30) to 10.4 m (34 ft; Oak Orchard-N) (Table 1). Average May – October values for SRP, chl-*a*, and SD showed no significant differences between nearshore and offshore locations. TP was significantly higher (p=0.03) in the offshore (10.6  $\mu$ g/L) than the nearshore (7.7  $\mu$ g/L) (Table 1).

Seasonal trends were also observed for most variables. The SD pattern was similar between nearshore and offshore locations; it was highest in May (7-9 m [23-30 ft]) and lowest in August (4–5 m [13–16 ft]) (Figure 2a). Nearshore chl-a concentrations were lowest in June (1.4 µg/L) and highest in August (2.6 µg/L; Figure 3a). Offshore concentrations were lowest in July (1.0  $\mu$ g/L) and highest in August (3.1  $\mu$ g/L; Figure Nearshore total phosphorus declined 3a). gradually throughout the season from a high of 9.1 µg/L in May to a low of 5.8 µg/L in October (Figure 4a). Offshore TP showed a similar pattern (13.6 µg/L in May, 8.3 µg/L in October) with a secondary peak in September (12.3  $\mu$ g/L) (Figure 4a). SRP concentrations were low (<1.2 $\mu g/L$ ) in both habitats for the entire season (Figure 5a).

## Water Quality Trends Since 1995

Comparisons with data collected since 1995 show that 2020 had average SD (6.3 m [21 ft]) in the nearshore and lower than average SD (6.5 m [21 ft]) in the offshore (Figure 2b, Table 3). Summer chl-*a* concentration in both the nearshore and offshore was slightly higher than the long-term mean of 1.7 µg/L for both habitats (Figure 3b). Mean May – Oct TP concentrations were about average in the nearshore (7.7 µg/L) and above average in the offshore (10.6 µg/L) habitats (Figure 4b). Offshore TP values should be interpreted with caution because few samples were collected in 2020. SRP remained low (~1.0 µg/L) in both habitats (Figure 5b).

## 2020 Zooplankton

Nearshore zooplankton density and biomass were highest in mid-June (Figure 6); this coincided with high numbers of bosminids and cyclopoids (Figure 7). Seasonal patterns in offshore zooplankton density and biomass were confounded by the limited number of samples that were collected at different locations across time (8 samples, 4 weeks, 5 sites) (Figures 6 and 7); biomass of most groups was low with the exception of calanoid copepods which were present during all four biweeks sampled (Figure 7). Zooplankton mean size was highest in the nearshore in early May and similar across all biweeks sampled in the offshore (Figure 6).

Bosminids and cyclopoids peaked in mid-June in the nearshore and then remained low for the rest of the sampling season (Figure 7). Calanoids also peaked in mid-June, declined, and then began to increase again in early September. Daphnids peaked in late June and declined gradually throughout the rest of the sampling season. Bythotrephes was detected in samples from both habitats while Cercopagis was present only in nearshore samples (Figure 7). Cercopagis was first detected in mid-June in the nearshore and peaked during mid-July (Figure 7). Bythotrephes first appeared in late-June in the nearshore and peaked in late September. Bythotrephes biomass was highest in the offshore in late-September (Figure 7). During biomass in mid-July. peak *Cercopagis* accounted for 25% of the total zooplankton **Bythotrephes** biomass in the nearshore; accounted for 10% of the nearshore biomass in late-August and 18% of the offshore biomass in late-September.

## Zooplankton Trends Since 1995

Summer total zooplankton density and biomass declined significantly from 1995 – 2020 (Figure 8; Table 3) in both the nearshore and offshore. Summer epilimnetic density in the nearshore (5026 ind/m<sup>3</sup>) was lower in 2020 than in any year from 1995 – 2019 (Figure 8). Offshore epilimnetic density (4987 ind/m<sup>3</sup>) was also low, but it was above the record low observed in 2010 (2763 ind/m<sup>3</sup>). Summer epilimnetic biomass in the nearshore (10.5 µg/L) was close to the record low (9.4 µg/L; 2017). Offshore epilimnetic biomass was 11.7 µg/L.

Change point analysis showed that a negative break occurred in nearshore total zooplankton density and biomass in 1998 (Figure 8; Table 3); biomass declined from an average of 127  $\mu$ g/L to 22  $\mu$ g/L at that time. In the offshore, there was a significant decline in summer epilimnetic zooplankton density and biomass 1995 – 2020 (Figure 8; Table 3). Change point analysis indicated a negative break in density in 2005 and negative breaks in biomass in 1999 and 2017 (Table 3); biomass declined from 95  $\mu$ g/L to 31  $\mu g/L$  in 1999 and from 31  $\mu g/L$  to 17  $\mu g/L$  in 2017.

Several trends were noted in summer zooplankton group biomass (Figure 9 and 10, Table 3). From 1995 – 2020, significant declines occurred in bosminid and cyclopoid biomass (nearshore and offshore), and in daphnid biomass (nearshore only). At the same time, biomass of *Bythotrephes* and *Holopedium* increased significantly (nearshore and offshore), as did other cladocerans (nearshore only) (Table 3).

Cercopagis and Bythotrephes biomasses were low compared to overall zooplankton biomass. Therefore, they were plotted separately to better depict patterns (Figure 11). The nearshore showed a positive change point in *Bythotrephes* biomass in 2006 and a negative change point in 2011 (Table 3). In the offshore, Bythotrephes biomass showed no breaks while Cercopagis biomass increased in 2000. Change points in the nearshore were also evident in bosminids (negative, 2005) calanoid copepods (positive, 2007; negative 2012), cyclopoid copepods (negative 2005) and Holopedium (positive 2003; In the offshore, change points Table 3). occurred in bosminids (negative, 2004) and cyclopoids (negative, 2005; positive, 2013; Table 3).

## Discussion

Secchi depth, chl-*a*, and TP are indicators of lake trophic status (Carlson 1977). In 2020, average May-October values for different sites ranged from 3.5 to 10.4 m SD, 0.4 to 4.2  $\mu$ g/L chl-*a*, and 5.9 to 17.6  $\mu$ g/L TP. Except for TP, these values are similar to other years in this decade and within the range for oligotrophic (low productivity) systems (0.3-3.0  $\mu$ g/L chl-*a*, 1-10  $\mu$ g/L TP; Wetzel 2001).

May – October average TP concentrations were relatively high in the offshore. We have rarely seen values over 10 ug/L in the offshore samples in the 2000s and our 2020 data were higher than 10 ug/L at 5 of 11 sites sampled. We did not collect the early spring offshore samples this year due to COVID-19 restrictions, so we could not use our standard spring TP as our indicator (Holeck et al. 2020). Instead we compared May - October averages. Would missing April TP values result in higher average TP values in our data from 2020? This is unlikely, because in previous years the April and May TP values were not different (p=0.9, n=14). Values from the nearshore in 2020 were slightly below the long-term mean. In other years, TP values in the nearshore and offshore are highly correlated suggesting that our offshore values were too high in 2020 and may not be reliable. Analysis of duplicate offshore samples also suggested high within-site variability; i.e., only 5 of 17 sites had a similarity between duplicates within the 20% acceptance criterion. We suspect that COVID-19 related adjustments in sampling protocols to allow for better distancing and smaller crews may have affected the consistency of sampling. Unfortunately, another monitoring program (USEPA - GLNPO) was not able to sample at all in 2020 making comparisons with other programs impossible. Clearly, it is important to obtain TP values from 2021 as TP is used as one of the main indicators of lower trophic level productivity in Lake Ontario. An increase in TP in the lake in 2020 would be a break from recent trends towards lower TP values in the offshore reported from both the ECCC and the GLNPO data (Dove and Chapra 2015, USEPA 2021).

Summer chl-*a* in 2020 was slightly higher in the nearshore (2.0  $\mu$ g/L) and offshore (2.1  $\mu$ g/L) compared to the long-term mean (1995 – 2019) of 1.7  $\mu$ g/L in both habitats (Figure 3b). Summer chl-*a* decreased significantly in the offshore 1995 – 2020 (but not in the nearshore, Table 3), although the values have increased recently compared with the lowest concentrations observed during 2009 to 2015.

May – Oct SD increased significantly in the offshore but decreased significantly in the nearshore from 1995 to 2020 (Table 3). However, SD was lower in the offshore from 2017 to 2020 than in most years after 2001. Spring and summer SD have also increased in the GLNPO offshore data set since 1986, but with little change since 1995 (Bunnell et al. 2021). Effects of higher water clarity on fish and fisheries are reviewed in Bunnell et al. (2021).

Summer epilimnetic zooplankton density and biomass decreased significantly in the offshore

and in the nearshore from 1995 - 2020 (Table 3). Biomass declined to below 20 µg/L for the first time in 2002 in the offshore (Figure 10) and in 1999 in the nearshore (Figure 9), declines that have been attributed to increased *Bythotrephes* abundance in the offshore and *Cercopagis* in the nearshore (Warner et al. 2006, Barbiero et al. 2014, Rudstam et al. 2015). These trends are consistent with observed effects of these predatory zooplankton elsewhere (Lehman and Caceres 1993, Yan et al. 2001, Pangle et al. 2007).

Offshore BMP zooplankton data are limited from 2020, as no July whole water column samples were available from the BMP program due to COVID-19 restrictions. Similarly, the GLNPO offshore sampling with the R/V Lake Guardian was cancelled in 2020. However, with the help of USGS and Canadian colleagues, we were able to secure samples from the eight GLNPO offshore sites in August 2020. These samples were analyzed with the USEPA standard operating procedure LG 403. To get some information on the offshore 2020 zooplankton, we plotted the BMP and GLNPO whole water column data from 2010 (first year of whole water column data from BMP) to 2020 (Figure 12). Note that these two programs collected samples at different locations and one month apart (July for BMP and August for GLNPO) and use somewhat different L-W regressions. Even so, the comparison of changes in species composition and biomass across years is quite good for most years. Therefore, we believe the GLNPO data gives a comparable picture to the BMP data for the whole water column. The GLNPO data from 2020 suggest relatively high calanoid copepod abundance and relatively low daphnid and cyclopoid abundance. similar to 2019, but lower Limnocalanus abundance than in the last 5 years. Overall whole water column zooplankton biomass in 2020 was similar to values seen since 2013.

Generally, *Bythotrephes* abundance is negatively correlated with alewife abundance (Johannsson and O'Gorman 1991, Barbiero et al. 2014) as alewife select for *Bythotrephes* and have grown better since *Bythotrephes* became abundant in Lake Ontario (Weidel et al. 2020). Our data indicate that *Bythotrephes* biomass was low 1995 - 2003, increased in 2004 to 2012, was again low in 2013 and 2014, and then increased to 2019 and 2020. This pattern does not directly mirror spring alewife abundance, which was relatively stable from 1997 – 2019 (Weidel et al. 2020). Note that alewife abundance is estimated in spring while Bythotrephes peak biomass estimates are from fall (September - October). Therefore, changes that occur over the course of the summer may also contribute to the lack of consistency between alewife abundance and Bythotrephes biomass. Nonetheless, these inconsistencies suggest the interaction between alewife and Bythotrephes is more complicated than suggested by Barbiero et al. (2014) and Rudstam et al. (2015).

There have also been other changes in zooplankton community composition. In an analysis of offshore zooplankton 1997 - 2016, Barbiero et al. (2019) observed the appearance of Daphnia mendotae around the time Bythotrephes became abundant (~2004-05). In previous years, the smaller Daphnia retrocurva was dominant. Further, there was a shift from Cercopagis to Bythotrephes. Similar patterns were observed in our offshore data; Daphnia mendotae biomass increased significantly from 1995 – 2019 while Daphnia retrocurva biomass remained stable, and Bythotrephes biomass increased from 1995 - 2019 while Cercopagis biomass remained stable. The presence of larger-bodied zooplankton of both daphnids and predatory cladocerans suggests a reduced level of alewife planktivory.

The BMP trends from 1995 - 2020 indicate relatively stable lower trophic level indicators in Lake Ontario but there is evidence of some continued oligotrophication, particularly in the offshore. The data show significant increase in SD, as well as decreases in summer chl-a and epilimnetic zooplankton density and biomass. A decline in whole water column zooplankton biomass since 1997 is also present in the GLNPO summer offshore data (Barbiero et al. 2019). Contrary to other time series, the BMP did not detect a significant decline in TP in either the nearshore or offshore habitats since 1995. Spring TP generally declined in the offshore GLNPO and ECCC Surveillance data sets, and 2019 BMP results revealed spring TP levels at historically low concentrations (Holeck et al. 2020). However, this indicator of oligotrophication did not decline further in the 2020 BMP data, rather TP increased compared to recent years. There was a small increase in offshore summer chl-a in 2020, but values were only slightly higher than the long-term mean. Given the difficulties with TP sampling in 2020 as indicated by the high variability between duplicates in the offshore, it is premature to suggest a reversal of the oligotrophication trend in the offshore.

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Figure 1. Map of Biomonitoring Program sites, 2020. Offshore stations are deeper than 20 m (66 ft). Nearshore stations are 10-17 m (33-56 ft) deep.



Figure 2a. Mean monthly Secchi depth (meters) for nearshore and offshore sites in Lake Ontario, May - October, 2020. Error bars are  $\pm 1$  SE.



Figure 2b. Long-term mean May – October Secchi depth (meters) in Lake Ontario, 1995 – 2020. Values are means equally weighted by site across the months of May, June, July, September, and October. August was removed because of the paucity of available data from the offshore.



Figure 3a. Mean monthly epilimnetic chlorophyll-a concentrations for nearshore and offshore sites in Lake Ontario, May - October, 2020. Error bars are  $\pm 1$  SE.





removed because of the paucity of available data from the offshore.



Figure 4a. Mean monthly total phosphorus concentrations for nearshore and offshore habitats in Lake Ontario, May - October, 2020. Error bars are  $\pm 1$  SE.



Figure 4b. Epilimnetic total phosphorus concentrations in Lake Ontario, 1995 - 2020. Values are means equally weighted by site across the months of May, June, July, September, and October. August was removed because of the paucity of available data from the offshore.



Figure 5a. Mean monthly soluble reactive phosphorus concentrations for nearshore and offshore sites in Lake Ontario, May - October, 2020. Error bars are  $\pm 1$  SE.







Figure 6. Biweekly mean  $(\pm 1 \text{ SE})$  daytime epilimnetic zooplankton density, size, and dry biomass for May - October 2020 at nearshore and offshore sites on Lake Ontario. On the x-axis, biweeks are designated by the date beginning each biweek. Numbers on bars in the middle panel indicate the number of samples taken. Lake surface temperatures (secondary y-axis; top panel) are from NOAA

Coastwatch web site (https://coastwatch.glerl.noaa.gov/ftp/glsea/avgtemps/2020/glsea-temps2020\_1024.dat).

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Figure 7. Daytime epilimnetic dry biomass of zooplankton community groups for nearshore and offshore areas of Lake Ontario, May - October 2020. Note different y-axis scales. On the x-axis, biweeks are designated by the date beginning each biweek.

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Figure 8. Mean summer (Jul-Aug) epilimnetic zooplankton density (top panel) and dry biomass (bottom panel) in nearshore and offshore habitats in Lake Ontario, 1995 – 2020. Error bars are  $\pm 1$  SE. Note that only 3 offshore stations were sampled in 2020.



Figure 9. Mean summer (Jul – Aug) daytime nearshore zooplankton group dry biomass in Lake Ontario, 1995 – 2020.



Figure 10. Mean summer (Jul – Aug) daytime epilimnetic offshore zooplankton group dry biomass in Lake Ontario, 2000 – 2020. Note that only 3 stations were sampled in 2020.



Figure 11. Daytime epilimnetic nearshore and offshore fall (September and October) Bythotrephes and summer (July and August) Cercopagis dry biomass in Lake Ontario, 1995 – 2020. Months were selected based on timing of peak biomass for each species.



Figure 12. Mean whole water column offshore zooplankton group dry biomass in Lake Ontario, 2010 – 2020. First bar for each year is BMP July data. Second bar for each year is USEPA-GLNPO August data. Groups are calanoid copepods (except for Limnocalanus); Limnocalanus macrurus, cyclopoid copepods, daphnids. Other non-daphnid cladocerans, and predatory cladocerans (Bythotrephes, Cercopagis, Leptodora, Polyphemus).

		Soluble reactive			
Sites	Chlorophyll-a (µg/L)	Total phosphorus (μg/L)	phosphorus (μg/L)	Secchi depth (m)	sampled
learshore					
Chaumont Lake (CBL)	2.2± 0.4 (n=11)	6.9± 0.6 (n=11)	0.8± 0.1 (n=11)	6.2± 0.7 (n=11)	5, 6, 7, 8, 9, 10
Galloo Island (GIL)	1.5± 0.2 (n=11)	6.0± 0.9 (n=11)	0.9± 0.1 (n=11)	7.7± 0.7 (n=11)	5, 6, 7, 8, 9, 10
Oak Orchard (OOL)	2.0± 0.3 (n=7)	8.8± 1.5 (n=7)	0.9± 0.1 (n=7)	5.2± 0.6 (n=6)	6, 7, 8, 9
Sodus Lake (SOL)	1.5± 0.2 (n=11)	6.3± 0.4 (n=11)	0.8± 0.1 (n=11)	7.2± 0.4 (n=11)	5, 6, 7, 8, 9, 10
Sandy Pond Lake (SPL)	2.6± 0.4 (n=6)	8.8± 1.7 (n=7)	1.2± 0.2 (n=8)	4.5± 0.5 (n=6)	5, 6, 7, 9, 10
Niagara East Lake (NEL)	1.9± 0.3 (n=6)	8.7± 1.1 (n=5)	1.4± 0.2 (n=5)	6.1± 0.7 (n=6)	7, 8, 9
Niagara West Lake (NWL)	1.3± 0.2 (n=6)	9.0± 0.7 (n=6)	1.5± 0.2 (n=6)	4.8± 0.3 (n=6)	7, 8, 9
Nine Mile Point 15 (NMP15)	3.3	13.9	1.6	3.5	8
ffshore					
Oak Orchard-N (OON)	0.4	9.6	1.0	10.4	5
Oak Orchard-O (OOO)	4.2	17.6	0.8	8.5	5
Smoky Point-N (SPN)	1.4± 0.2 (n=3)	8.7± 1.2 (n=3)	0.8± 0.1 (n=3)	5.5± 0.5 (n=2)	6, 9, 10
Smoky Point-O (SPO)	2.8± 0.9 (n=3)	12.4± 1.3 (n=3)	1.2± 0.2 (n=3)	5.5± 0.5 (n=2)	6, 9, 10
Oswego Shallow	2.4	5.9	1.2	7.5	6
Oswego Deep	3.2	12.6	1.6	5.5	6
Main Duck	1.6± 0.5 (n=2)	11.9± 0.1 (n=2)	0.9± 0.1 (n=2)	7.3± 1.8 (n=2)	6, 7
Galloo/Charity Trench	3.1	7.7	0.8	4.0	8
Tibbetts Point	1.3± 0.5 (n=2)	6.7± 0.1 (n=2)	1.2± 0.5 (n=2)	6.8± 1.8 (n=2)	6, 7
EPA 55	3.4	7.9	0.9	5.5	8
EPA 60	3.4	14.6	1.3	5.0	8
Nine Mile Point 100 (NMP100)	n/a	n/a	n/a	4.0	8
Nine Mile Point 50 (NMP50)	n/a	n/a	n/a	4.0	8
Nine Mile Point 30 (NMP30)	n/a	n/a	n/a	3.5	8

# Table 1 Mean chl a TP SRP and Secchi denth (+ 1 SE) for nearshore and offshore sites May – October 2020

Table 2. Comparison of nearshore and offshore sites May – Oct, 2020 by fitting a mixed effe	cts model
with month and habitat as fixed effects and site as a random effect. Reported p-values are f	or the
significance of habitat, not month. Values shown are arithmetic means averaged by month	for each
site, and then averaged for months May through October. All offshore data are for the top	10 m of the
water column.	

		IVIE	Mean		
Parameter		Nearshore	Offshore	p-value	
	Total phosphorus (μg/L)	7.7	10.6	0.03	
	Soluble reactive phosphorus (µg/L)	1.0	1.0	0.6	
	Chlorophyll <i>a</i> (µg/L)	1.9	2.1	0.6	
	Secchi depth (m)	6.3	6.5	0.8	

Table 3. Results of regression analyses performed on data from Lake Ontario's offshore and nearshore. Trends are indicated by (+) or (-). Significant p-values  $(p \le 0.05)$  are indicated in bold. Marginal p-values (p<0.10) are indicated in italics. ns=not significant. Slope is from the linear regression and represents the annual change in each parameter (units of change match the units of each parameter). Zooplankton group biomass could not be normalized; Spearman rank correlation was used on those data, and change reported is the slope of the linear regression. Change point analyses were performed on 1995 – 2020 data in the both the offshore and nearshore. \*\*change point performed on ranks due to outliers. 2020 mean does not include August.

	Regression		Change Point Analysis		
				2020	1995 – 2019
Offshore	1995 – 2020	Slope	1995 - 2020	mean	mean (range)
May-Oct TP (µg/L)	ns		no breaks	10.7	6.2 (3.0-8.4)
May-Oct Secchi Depth (m)	(+) p=0.03	0.1	no breaks	7.0	7.2 (5.4-10.3)
May-Oct chlorophyll <i>α</i> (μg/L)	(-) p=0.04	0.04	(-) 2001	2.2	1.7 (0.7-2.8)
Summer epilimnetic zooplankton density (#/L)	(-) p<0.0001	1692	(-) 2005	5.0	20.4 (2.8-56.2)
Summer epilimnetic zooplankton biomass (µg/L)	(-) p<0.0001	3.2	(-) 1999, (-) 2017	11.7	39.1 (8.1-129.1)
Summer epilimnetic zooplankton group biomass					
(µg/L)					
Bosminids	(-) p=0.0025	0.4	(-) 2004		
Bythotrephes longimanus	(+) p=0.0135	0.03	no breaks		
Calanoid copepods	ns		no breaks		
Cercopagis pengoi	ns		(+) 2000		
Cyclopoid copepods	(-) p=0.0007	2.2	(-) 2005, (+) 2013		
Daphnids	ns		no breaks		
Other Cladocerans	ns		no breaks		
Limnocalanus	(-) p=0.0509	0.04	no breaks		
Holopedium	(+) p=0.0079	0.1	no breaks		
			-		
	Regression		Change Point Analysis		
Nearshore	1995 - 2020	Slope	1995 -2020		
May-Oct TP (μg/L)	ns		no breaks	7.2	7.8 (4.7-11.8)
May-Oct Secchi Depth (m)	(-) p=0.03	0.03	no breaks	6.7	6.4 (5.5-7.5)
May-Oct chlorophyll $a$ (µgL)	ns		(+) 2001; (-) 2010	1.7	1.5 (0.9-2.2)
Summer epilimnetic zooplankton density (#/L)	(-) p=0.0002	1902	(-) 1998	5.0	21.0 (5.8-77.8)
Summer epilimnetic zooplankton biomass (µg/L)	(-) p=0.0006	3.2	(-) 1998	10.5	34.8 (9.4-159.0)
Summer epilimnetic zooplankton group biomass					
(µg/L)	() ==0.0000	0.7	**( ) 2005		
Bosminius Dethates has been interested	(-) p=0.0008	0.7	(1) 2005		
Bythotrephes longimunus Colonaid concorde	(+) p=0.0087	0.005	(+) 2006, (-) 2011 (+) 2007, (-) 2012		
	ns		(+) 2007, (-) 2012		
Cercopagis pengoi	ns	2.0	no breaks		
Cyclopola copepoas	(-) p<0.0001	2.0	(-) 2005		
LIADDOIDC	() 0.0001		1_1 1000		
Other Cladeseres	(-) p=0.0031	0.7	(-) 1999		
Other Cladocerans	(-) p=0.0031 (+) p=0.048	0.7	no breaks		
Other Cladocerans Limnocalanus	(-) p=0.0031 (+) p=0.048 ns	0.7	no breaks		
Other Cladocerans Limnocalanus Holopedium	(-) p=0.0031 (+) p=0.048 ns (+) p=0.0021	0.7 0.04 0.1	no breaks no breaks (+) 2003		