Title: Extreme midsummer rainfall event drives early onset cyanobacterial bloom

Running head: Extreme rainfall drives early bloom

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Abstract

The prevalence and increasing global distribution of cyanobacteria-dominated harmful algal blooms is strongly associated with changing climatic patterns and local biogeochemical and hydrological processes. Changes to precipitation frequency and intensity, as predicted by current climate models, are likely to alter bloom development and composition due changes in nutrient fluxes and water column mixing on short time scales during the open water season. However, few studies have directly documented the effects of precipitation events on cyanobacterial composition, biomass, and toxin production. In this study, we describe an early-initiated cyanobacterial bloom in Conestogo Lake, a eutrophic flood control reservoir located in southwestern Ontario, following heavy rainfall and subsequent flooding within the catchment. An increase in bioavailable phosphorus by more than 25-fold in surface waters resulted in biomass increases of *Aphanizomenon flos-aquae* throughout the reservoir approximately 2 weeks post-flooding. Anabaenopeptin-A and three microcystin congeners (microcystin-LR, -YR, and -RR) were detected at varying levels across sites during the bloom period, which lasted between 3 to 5 weeks. Together, these findings indicate that water column mixing and elevated phosphorus concentrations induced by increased water flow were the key drivers for the early cyanobacterial bloom in Conestogo Lake. Mitigation strategies for bloom-related water quality impairment must be both responsive and adaptive to the complexity of drivers affecting reservoir mixing dynamics, nutrient loads, and blooms. However, bloom mitigation efforts can be severely constrained by the large watersheds and broad suite of operational goals of many reservoirs. that are most often focussed on water quantity (flood water control and flow augmentation) rather than water quality or cyanobacterial bloom management.

Key words: cyanobacterial bloom, phosphorus, climate, precipitation, toxin, harmful algal bloom, Ontario, reservoir, extreme rainfall

Introduction

Cyanobacteria are critical to the structure and function of aquatic communities (Reynolds 1984, Wetzel 2001). However, prolific cyanobacterial growth often negatively impacts ecosystems by reducing water quality and driving disruptions to aquatic food chains (Pearl 1988). Several bloom-forming species also synthesize an array of bioactive compounds that pose chronic and acute health risks to humans and animals through dermal contact, inhalation, and/or ingestion of contaminated waters (Chorus et al. 2000, Codd 2000, Carmichael 2001). As a result, blooms and their associated bioactive metabolites have economic consequences through impacts on tourism and recreation, and via costs for drinking water treatment (Dodds et al. 2009, Bullerjahn et al. 2016).

The increased prevalence of cyanobacterial blooms has been historically attributed to increased anthropogenic eutrophication, with emphasis on the relative abundances, contributions, and impacts of nitrogen and phosphorus (Winter et al. 2012, Paerl and Otten 2013). Changing climatic conditions are also now strongly considered as significant drivers in bloom intensity and distribution (Paerl and Huisman 2009, O'Neil et al. 2012, Sukenik et al. 2015, Paerl et al. 2016). Current climate models predict increasing regional temperatures likely resulting in warmer surface waters, increased thermal stratification, and water column stability that promote the growth of certain bloom-forming species (Jöhnk et al. 2008, Paerl and Huisman 2009). Precipitation frequency and intensity are also predicted to change, but less attention has been given to how these factors may influence bloom development and cyanobacterial biomass (Reichwaldt and Ghadouani 2012).

71 Increasing precipitation variability may impact external nutrient and sediment delivery to
72 waterbodies, alter residence time and flushing, and reduce vertical stratification, which, in turn,
73 may affect bloom development (Jacobsen and Simonsen 1993, Mitrovic et al. 2003, Wood et al.
74 2017). For example, Wood *et al.* (2017) reported decreased cyanobacterial biomass following an
75 extreme rainfall event that lead to water column cooling and destratification in a shallow,
76 eutrophic New Zealand lake. Intense precipitation, preceded or followed by extensive drought
77 may create episodic or pulsed nutrient loads potentially further favoring cyanobacterial
78 development (Bouvy et al. 2003, Reichwaldt and Ghadouani 2012). In addition to climatic
79 factors, bloom development is also dependent on a suite of site-specific characteristics including
80 hydrology, lake geomorphology, catchment size, and nutrient loading from internal and external
81 sources. Thus, the sensitivity of each lake to these drivers, including precipitation, will vary.
82 Cyanobacterial blooms are of particular concern for lake and reservoir managers due to
83 the potential for toxin production and impacts on water quality that may affect ecosystem
84 processes and the recreational uses of the water body (Chorus and Bartram 1999, Chorus et al.
85 2000). Cyanobacterial metabolites are both chemically variable and bio-actively diverse
86 (Carmichael 1997, Welker and Von Döhren 2006) with various modes of cellular action that may
87 cause superficial skin irritation at low exposures and sickness or even death if ingested at high
88 enough concentrations (Pouria et al. 1998, Chorus et al. 2000, Carmichael et al. 2001,
89 Carmichael 2001). Often, the recommended course of action for recreational waterbodies is to
90 limit activity in and exposure to bloom-affected waters due to the potential for accidental
91 ingestion and/or inhalation during recreational activities (Backer et al. 2010). Here, we explore
92 the effects of an extreme precipitation event on cyanobacterial blooms within Conestogo Lake
93 (Ontario, Canada; Figure 1), a eutrophic flood-control and river augmentation reservoir, which

has experienced recurring, late-fall cyanobacterial blooms dominated by the nitrogen-fixing species *Aphanizomenon flos-aquae.* On 23 June 2017, the Upper Conestogo watershed received a substantial amount of rainfall (daily avg. 78 mm measured at Conestogo Dam) that resulted in severe flooding. According to the GRCA, nearly 80% of the volume of Conestogo Lake was flushed downstream during this 2-day event (Grand River Conservation Authority 2018). In this study, we were specifically interested in how significant rainfall from the catchment altered water quality and nutrient concentrations and if so, how these changes impacted cyanobacterial bloom development and toxin production.

Materials and methods

Study Site

Conestogo Lake is located in southwestern Ontario, Canada. It is a bottom-draw reservoir (7.35 km^2) operated by the Grand River Conservation Authority (GRCA; Mapleton Township, ON). Built in 1958, it primarily serves as a flood-control and down-stream flow augmentation system while providing ancillary recreational benefits for fishing, boating, and ~ 400 summer cottages. Higher flows in spring from the Upper Conestogo River basin (566 km^2), a predominately agricultural catchment (> 80 %, Figure 1, *left*), are captured in the reservoir through the end of April and used during the summer drawdown (*i.e.* augmentation) period from 01 May to 30 September to provide consistent downstream flow of the Conestogo and Grand Rivers. As a result of the reservoir drawdown, mean lake depth varies throughout the season with the deepest point always near the dam. Reservoir storage volume during the summer augmentation period has historically ranged between 12.7 and 56.3 Mm^3 (59.3 Mm^3 max cap.) with an average drop between 5 and 7 m in stage elevation.

117

118 **Sample collection and analysis**

119 Sampling at Conestogo Lake began on 21 Jun 2017 with physical and biological samples
120 collected from the center (CLC) site closest to the dam. Two additional sites in the east (CLE)
121 and west (CLW) arms of the reservoir were added on 05 Jul and 11 Jul, respectively, following
122 the flooding event to assess the potential differences between catchment inputs into the reservoir
123 (Figure 1). From each location we collected a suite of physical, chemical, and biological data
124 throughout the water column. Physical water column profiles were collected at 0.5 m increments
125 using an EXO2 sonde (YSI, Yellow Springs, OH) equipped with chlorophyll-a, pH, temperature,
126 and dissolved oxygen sensors. The Secchi disk transparency at each site was used to collect
127 samples for cyanobacterial toxin analysis described in further detail below. In addition, we
128 collected discrete samples from 2 m for water chemistry and phytoplankton taxonomy and
129 enumeration approximately once per week from 21 June 2017 to 17 August 2017.

130 Whole water samples were collected for total phosphorus (TP) and phytoplankton
131 enumeration. Subsamples were field-filtered using a 0.45 µm syringe filter (Whatman) for
132 soluble reactive phosphorus (SRP), total dissolved phosphorus (TDP), ammonia (NH_4-N), nitrate
133 (NO_3^-), total dissolved nitrogen (TDN), anions, and cations. All samples were transported on ice,
134 stored at -20 °C, and analyzed at the Environmental Geochemistry Laboratory at the University
135 of Waterloo (Waterloo, ON) as per AHPA (1998; see Table S1 for instrument listing and
136 detection limits).

137 Phytoplankton identification and enumeration were completed by D. Findlay (Plankton-
138 R-Us, Winnipeg, MB, http://www.plankton-r-us.ca) as per Findlay & Kling (2003). Briefly,
139 phytoplankton samples were collected in a dark bottle, preserved with Lugol's solution, and

140 stored at 4 °C until analysis. Ten-mL aliquots of Lugol's preserved sample were gravity
141 settled for 24 h and counted using a modified Ütermohl technique (Nauwerk 1963) on
142 an inverted microscope with phase contrast illumination. Cell counts were converted to
143 wet weight biomass by approximating cell volume, which were obtained by
144 measurements of up to 50 cells of an individual species and applying the geometric
145 formula best fitted to the shape of the cell (Vollenweider 1968, Rott 1981). A specific
146 gravity of 1 was assumed for cellular mass. All biomass estimates are expressed as
147 mg/m^3 (D. Findlay, pers. communication).
148 Total (*i.e.*, intracellular and extracellular) cyanobacterial metabolite
149 concentrations were determined from a 115 mL whole water sample calculated from the
150 site-specific Secchi depth (2× Secchi depth) at each site. Each sample was collected in
151 an amber NalgeneTM polyethylene terephthalate glycol (PETG) bottle to limit
152 adsorption and overflow during freezing (Fisher reference: 322021-0125). All samples
153 were stored at -20 °C until analysis at the University of Montreal (Montréal, QC).
154 Samples were prepared and screened for each of seventeen cyanobacterial compounds
155 (Table S2) as per Fayad *et al.* (2015) via on-line solid phase extraction ultra-high
156 performance liquid chromatography high resolution mass spectrometry (SPE−UHPLC–
157 HRMS) using standards purchased from Enzo Life Science, Abraxis, or Cyano Biotech
158 GmbH. The limit of detection (LOD) and limit of quantification (LOQ) were calculated
159 for every batch of samples. Only results that exceeded the LOQ were included in our
160 analysis. Therefore, some samples may have had detectable, but unquantifiable toxin
161 concentrations.
162

163 **Environmental and dam-related data**

164 Meteorological and other reservoir-related data for this study were obtained from the GRCA's
165 online data portal (https://data.grandriver.ca/). Because GRCA data are provided under a
166 provisional status, all GRCA data were passed through quality assurance and quality checking
167 metrics for outliers to ensure sound data structure before proceeding with analysis. Spatial data
168 were obtained from open source portals at the GRCA (bathymetry), the United States Geological
169 Society, Statistics Canada, and the Ontario Ministry of Natural Resources (land-use).

170

171 **Data analysis and statistics**

172 All data analysis was completed in R version 3.5.1 (R Core Team 2018). Water column profile
173 data derived from sonde measurements as well as the discrete chemical profiles were constructed
174 using a multilevel B-spline interpolation from the *MBA* package (Finley et al. 2017) and were
175 adjusted to reflect the reservoir stage elevation at the time of sampling. One sampling in the east
176 arm contained a high chlorophyll-a concentration that skewed the profiles for all other sites.
177 Therefore, we log transformed (base 10) the data to allow for better visual comparison.
178 Comparisons of chemical and physical data across sites were completed using principal
179 coordinates analysis (PCoA) with a Euclidean matrix on log-normalized data to account for
180 scaling differences in the dataset and tested for difference through time and by site using
181 permutational analysis of variance (PERMANOVA) with *adonis* in the *vegan* package
182 (Okasanen et al. 2016). Similarly, we tested for differences in the phytoplankton community
183 composition by biomass using PCoA with a Bray-Curtis dissimilarity matrix on log-transformed
184 data to account for rare species biomass in the community and tested for difference through time
185 and by site using PERMANOVA. Biomass and toxin trends were analyzed using repeated

186 measures ANOVA (RM-ANOVA) with a linear mixed effects model (*lme*) from the *nlme*
187 package (Pinheiro et al. 2018) where time was a fixed effect, sampling location as a random
188 effect, and the correlation matrix selected based on Akaike Information Criterion (AIC).
189 Relationships between cyanobacterial biomass and environmental drivers including surface
190 water temperature and phosphorus concentrations were also completed using RM-ANOVA. In
191 addition, we tested for the correlation of biomass with toxin concentrations using Kendall's rho.
192 All code used for this project are available on github at
193 https://github.com/biogeochem/formbloom_conestogo_2017HAB.
194

195 ## *Results*

196 **Flood event**

197 The Upper Conestogo watershed received an intense rainfall event on 23-24 June 2017. This
198 two-day event increased mean daily flow from 4.56 m^3/s to 219 m^3/s (max 521 m^3/s), increased
199 the total reservoir volume by 3.5 Mm^3 (million cubic meters), and is estimated to have replaced
200 ~80% of the reservoir volume (Grand River Conservation Authority 2018). Within several days,
201 the reservoir storage and drawdown returned to engineered levels (Figure 1*b*-*e*). The sizable
202 influx of water from the catchment during the flood event increased measurable phosphorus
203 between 1.9 to 33× within the water column at the center site by the next sampling on 05 July
204 2017 but did not affect any measured nitrogen species (Figures 2 and 3).
205

206 **Hydrology and Descriptive Limnological Characteristics**

207 All measured forms of phosphorus increased in the water column following the severe rainfall
208 event (Figures 2*a* and 3*a*, Tables 1 and S1). Epilimnetic concentrations declined throughout the

209 sampling period while hypolimnetic concentrations, notably TDP and TP, increased between 12
210 July (DOY 193) and 26 July (207) across all three sites. All nitrogen species (Figure 3*b* and S1,
211 Tables 1 and S1) were well mixed throughout the water column and gradually decreased
212 throughout the sampling campaign.

213 Conestogo Lake was initially and only briefly thermally stratified at the center site on 21
214 June 2017. By 05 July (DOY 186), the lake was isothermal across the lake and remained
215 isothermal throughout the rest of the summer augmentation period (Figure S2*a*). Mean surface
216 water temperatures (0 - 2 m) ranged from 21.2 to 25.5 °C. Subsequent flow from the catchment
217 following the 23 July rainfall reduced the surface water temperatures in the east by
218 approximately 2 °C from the previous sampling but did not affect the surface temperatures in the
219 west arm or center sites. Dissolved oxygen (Figure S2*b*, Tables 1 and S1) at the surface was
220 highest during peak biomass. Hypoxia developed in the hypolimnion at all three sampling
221 locations just as bloom biomass started to increase. Epilimnetic (0 - 2 m) chemical (TDN, NO_3^-,
222 DOC, SRP, TDP, TP, SO_4^{2-}, Mn, Ca^{2+}, K^+, Na^+, Mg^{2+}, F,$^-$, Cl^-) and physical (surface
223 temperature, dissolved oxygen, residence time) properties were significantly different between
224 sites (PERMANOVA, $P = 0.013$) and changed over the course of the sampling period
225 (PERMANOVA, $P = 0.001$; Figure S2).
226

227 **Flooding induced cyanobacterial bloom**

228 GRCA managers have visibly recorded cyanobacterial blooms in Conestogo Lake in late August
229 through September (S. Cooke, pers. communication), similar to other eutrophic lake systems in
230 this region (Winter et al. 2012). Within two weeks of the flooding event measurable increases in
231 biomass of *Aphanizomenon flos-aquae*, a filamentous nitrogen-fixer, were detected in each of the

232 sampling locations (Figure 4). Epilimnetic phytoplankton composition significantly changed
233 over the course of the sampling campaign with assemblages in the west and center more similar
234 than those in the east (Figure S3, PERMANOVA, site: $F = 2.22$, $P = 0.001$, time: $F = 2.69$, $P =$
235 0.003). While other phytoplankton groups were represented (Figure S4), cyanobacteria exceeded
236 50% of the phytoplankton community in the east and west arms near 12 July and approximately
237 a week later in the center. Total cyanobacterial biomass varied across sites and was significantly
238 affected by eplimnetic SRP, mean daily flow rates, and surface water temperatures
239 (RMANOVA, $F_{1,462} = 778.79$, $P < 0.0001$). The east arm contained the highest measured
240 biomass on 19 July (DOY 200, 10,900 mg/m^3, 4,500 cells/mL), which declined following
241 another, smaller, rainfall event on 23 Jul 2017. The center and west arm biomass peaked near 25
242 Jul (DOY 206; center: 4,710 mg/m^3, west: 5,230 mg/m^3). Bloom duration, measured as the
243 period during which cyanobacterial biomass composed > 50% of the fractional biomass,
244 persisted between 26 and 34 days and occurred 4 to 6 weeks earlier than has been previously
245 documented by the GRCA (S. Cooke, pers. communication). The cyanobacterial fraction across
246 all sampling sites also included *Woronichinia compacta*, *Anabaena (Dolichospermum) flos-*
247 *aquae*, *Microcystis aeruginosa*, *Anabaena* (*Dolichospermum) crassa, Planktolyngbya limnetica,*
248 and *Pseudoanabaena sp.* (Figure 4*a*). *W. compacta* exhibited similar biomass trends to *A. flos-*
249 *aquae*, though at much smaller biomass levels; by 17 Aug, its biomass increased to 8% (193.8
250 mg/m^3), 13% (108.9 mg/m^3), and 18% (585.9 mg/m^3) of the cyanobacterial biomass in the west,
251 center, and east, respectively.
252
253 **Cyanotoxins were detectable at low concentration**s

254 Detectable anabaenopeptin-A (AP-A), microcystin-LR (MC-LR), -YR, and -RR were present in
255 90 % of the samples (n = 19) but were only quantifiable in 81 % of samples (n = 17; Table 2).
256 Total microcystin concentration did not exceed 1.0 µg/L throughout the sampling season and
257 were comparable to previously measured concentrations during the summer months
258 (Yakobowski 2008). Metabolite type and concentration varied through time, but not by site
259 (RMANOVA, toxin × time, $F_{3,60} = 9.90$, $P < 0.0001$; site $P > 0.05$). Quantifiable concentrations
260 of all three microcystins were present in the east arm, MC-LR and MC-YR in the center site, and
261 only MC-LR in the west. MC-LR was the dominant microcystin variant across all sites with the
262 highest quantified values in the east arm at peak *A. flos-aquae*, *M. aeruginosa*, and *P. limnetica*
263 biomass (Figure 4*b*, Table 2). Further, MC-YR at the center did not correspond with the MC-LR
264 peak suggesting its possible origin in the east arm. AP-A was present at all sampling locations
265 throughout the bloom period, was the dominant metabolite at all sites by 25 July (DOY 206) and
266 had replaced all microcystin variants by the end of the sampling campaign. Toxin concentrations
267 were not significantly correlated with the biomass of any particular cyanobacterial species
268 (Kendall rho, $P > 0.05$).
269

270 *Discussion*

271 Conestogo Lake has experienced annual cyanobacterial blooms over the last decade, some of
272 which have resulted in the issuance of health advisories for recreational users and cottagers (S.
273 Cooke, GRCA, pers. communication). Consistent with other eutrophic lake and reservoir
274 systems in the region (Winter et al. 2012), cyanobacterial blooms typically develop in mid- to
275 late-August and persist through September or early October (S. Cooke, GRCA, pers.
276 Communication, Larsen *et al.* in prep). However, the marked increase in soluble P as a result of

277 rainfall combined with rapid physical change from disruption of water column structure during
278 flooding to reestablishment of a stable water column made 2017 an atypical season with an
279 unusually early bloom. This highlights the potential for changing windows of bloom-related risk
280 and associated management considerations.

281 Global increases in extreme rainfall events are predicted to outpace changes in
282 total precipitation under various climate models (Allen and Ingram 2002, IPCC 2007)
283 suggesting that heavy rain and flooding events will likely become more common in the
284 near future. The impact of changes to precipitation intensity and frequency on
285 cyanobacterial bloom development and dynamics is inherently complex and may lead to
286 either promotion or disruption of bloom dynamics (Reichwaldt and Ghadouani 2012,
287 Wood et al. 2017). Interactions between physical, chemical, and biological lake
288 parameters are influenced by other factors such as catchment size, land use, soil type,
289 etc. and ultimately, the overall effect of the rainfall event is regulated by the rainfall
290 intensity, water inflow, and seasonal timing (reviewed in Reichwaldt and Ghadouani
291 2012). Limited studies investigating such events on cyanobacterial bloom development
292 have identified changes to flushing rates, water column mixing, and nutrient inputs from
293 rainfall events as the main factors affecting cyanobacteria and phytoplankton
294 communities (Bouvy et al. 2003, Reichwaldt and Ghadouani 2012) – all factors that
295 appeared to impact the bloom dynamics in Conestogo Lake. Regional climate models
296 suggest we will continue to see impacts of extreme events that can affect bloom timing,
297 and risk (Bouvy et al. 2003, IPCC 2007, McDermid et al. 2015).

298 Sizable influxes of dissolved and particulate nutrient pools are common following
299 heavy rainfall and erosion, particularly if they are preceded by a warm, dry periods (Jones and

300 Poplawski 1998, Chiew et al 1995). For example, in Australian reservoirs, a 440 mm rainfall in a
301 3-day period resulted in an input equivalent of 80% and 400% of the average annual in-lake N
302 and P, respectively (Jones and Poplawski 1998). Here we observed a 27x increase in SRP levels
303 in surficial waters, which was likely key to the proliferation and dominance of cyanobacteria.
304
305 *Emerging cyanotoxins*
306 Many lake monitoring programs focus on the measurement of microcystins because of their vast
307 distribution (Loftin et al. 2016), ecotoxicity (Chorus and Bartram 1999), and relatively
308 inexpensive and rapid detection in whole water samples (*e.g.* ELISA test kits). However, several
309 studies using cyanobacterial extracts have reported harmful/toxic effects that could not be
310 explained solely by microcystin concentration or presence, suggesting the possibility of other
311 toxic compounds (Keil et al. 2002, Teneva et al. 2005, Baumann and Jüttner 2008, Smutná et al.
312 2014, Lenz et al. 2019). Improved analytical techniques have identified numerous bioactive
313 compounds such as cyanopeptolins and anabaenopeptins that are readily detectable in
314 freshwaters and often produced simultaneously with microcystin variants (Harada et al. 1995,
315 Welker and Von Döhren 2006, Gkelis et al. 2015, Beversdorf et al. 2017, 2018).
316 In some cases, and consistent with our results, anabaenopeptins are reported in equal or
317 higher concentrations than microcystins (Janssen 2019). For example, Beversdorf *et al.* (2017)
318 reported an average of 0.65 μg/L total microcystin in Lake Koshkonong, Wisconsin, while
319 anabaenopeptin-B and -F were measured at 6.56 μg/L combined. Though there are no case
320 studies of human toxicity caused by anabaenopeptins, the compound inhibits carboxypeptidase
321 A, and like microcystin, also inhibits protein phosphatases with slightly overlapping inhibitory
322 concentration ranges (Honkanen et al. 1990, Sano et al. 2001, Spoof et al. 2016). At present, the

323 concentration to which anabaenopeptins and other metabolites would have to reach to affect
324 human health is currently unknown resulting in a lack of regulations or advisories for
325 recreational or drinking water. However, ecological effects may be observed at relatively low
326 concentrations. A recent study by Lenz *et al.* (2019) reported induced toxicity by low
327 concentrations (10 µg/L) of anabaenopeptins, including AP-A, on the nematode *Caenorhabditis*
328 *elegans* resulting in reduced reproduction, reduced lifespan, and delayed hatching. Though
329 compounds like AP-A have been previously considered non-toxic, they may now represent a
330 new class of emerging toxins, whose potential impacts to human health and toxicity to aquatic
331 organisms require immediate attention and therefore, inclusion in risk assessment for lake
332 mitigation and monitoring programs
333
334 *Recommendations for management*
335 Watershed managers have various options to mitigate cyanobacterial blooms by disrupting
336 conditions that favor growth. These strategies, including increasing flushing rates, reducing
337 internal/external nutrient loads, destratisfying the water column through mixing, application of
338 chemicals, and/ or biological manipulations, each come with varying economic costs and have
339 been extensively reviewed elsewhere (Paerl 2018). Different short-term and long-term targets for
340 water management regimes and/or nutrient loads will likely be needed to mitigate cyanobacterial
341 bloom formation in Conestogo Lake. However, manipulating water flow to reduce residence
342 time is not likely a feasible option since the reservoir operates with base-flow augmentation as a
343 primary deliverable. Therefore, management goals will likely need to incorporate strategies to
344 mitigate internal and external nutrient loads. TDN nor NO_3 concentrations increased in response
345 to the flood event and steadily decreased throughout the campaign. This suggests that N is not

346 likely the regulating nutrient for biomass in this system though it may play a role in selecting for
347 cyanobacterial species, bloom timing, or contributing to other biogeochemical cycles.
348 Concurrent with other work on Conestogo Lake (Guildford 2006), our results suggest that P
349 management strategies should be the main given the increased SRP concentrations in the water
350 column following the flood and hypolimnetic P release from anoxic sediments. Further work will
351 be needed to determine whether, if in a typical year, internal P release is the main driver for the
352 late August or early September cyanobacterial blooms that have been previously documented
353 anecdotally.

354 Short-term strategies such as chemical binding (*e.g.* Phoslock or alum) will precipitate
355 suspended P to the sediment and could therefore reduce the size and/or severity of the bloom
356 (Robb et al. 2003, Lürling and Faassen 2012, Cooke et al. 2013). However, keeping P
357 immobilized for long periods is difficult in systems with strong anoxic bottom waters like
358 Conestogo Lake. Longer-term strategies could include reduced P catchment loads via changes to
359 the release of P-rich wastewater from upstream water treatment facilities, the incorporation of an
360 increased riparian zone and/or dredging (Lürling and Faassen 2012, Aguiar et al. 2015).
361 Increased riparian zones have been effective in reducing particulate P in various other systems
362 (Aguiar et al. 2015) and may be a possible strategy since the predominant Tavistill soil type in
363 the Upper Conestogo River catchment is highly susceptible to erosion (Macrae et al. 2007, Grand
364 River Conservation Authority 2018). At present, only a small strip of forested land surrounds the
365 main body of the reservoir, hence reduction of localized inputs may be achievable, even if
366 riverine inputs are more difficult to control.

367 This task of identifying solutions to mitigate cyanobacterial blooms is challenging – yet
368 urgent – given widespread reports of worsening bloom risk (Pick 2016). Development of

appropriate mitigation strategies must incorporate an understanding of watershed nutrient sources, physiochemical lake structure, biological conditions, and climatic predictions into an adaptive framework considering what is possible, what are the costs (financial costs, and trade-offs to ecosystem services), and what are the goals of ecosystem and watershed management. Within Conestogo Lake we demonstrate that extreme rainfall events can trigger early bloom development. Management strategies and risk reduction activities, which currently target the low flow period in late summer may need to be altered in light of predictions of more frequent extreme rainfall events which may trigger an earlier bloom season in some ecosystems.

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Data availability statement

Data and code that support the analysis and findings in this study are available in figshare at http://doi/10.6084/m9.figshare.7811963.

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559

560

561 **Table 1.** Chemical water column summary with mean (SD) [minimum – maximum] at sampling

562 each site.

Site	NO_3^- (mg-N/L)	TDN (mg-N/L)	SRP (µg-P/L)	TDP (µg-P/L)	TP (µg-P/L)
Center	4.82 (0.59) [3.99 - 5.65]	5.32 (0.64) [4.24 - 5.86]	13.66 (22.86) [1.52 - 65.2]	26.41 (26.32) [9.96 - 85.77]	39.84 (27.59) [14.99 - 103.48]
East	4.37 (0.79) [3.34 - 5.63]	4.74 (0.89) [3.39 - 5.85]	5.89 (8.45) [1.56 - 24.61]	18.53 (11.14) [7.79 - 41.59]	49.54 (11.08) [35.2 - 68.65]
West	4.69 (0.57) [3.89 - 5.43]	5.1 (0.83) [3.94 - 6.03]	4.09 (4.73) [1.23 - 13.48]	15.99 (7.7) [8.37 - 29.83]	39.35 (4.26) [34.69 - 47.04]

563

564 **Table 2**. Quantified cyanobacterial metabolites across sampling sites as measured by LC-HMRS
565 with values which exceeded the limit of detection (LOQ) presented as mean ng/L (standard
566 deviation ng/L), those in italics exceeded the limit of detection (LOD) but not the limit of
567 quantification (LOQ), and those with (-) < LOD. The metabolites MC-LA, -LY, -LW, -LF, -
568 HiIR, -HtyR, and CPA were not detected in samples.

569
570

Date	Site	cyanobacterial secondary metabolites						
		AP-A	MC-LR	MC-RR	MC-YR	ATX	HATX	CYN
21 Jun	Center	-	-	-	-	-	-	-
05 Jul		-	-	-	*33.26 (16.08)*	-	-	-
14 Jul		*23.72 (4.89)*	-	-	*20.6 (29.13)*	-	-	-
18 Jul		109.25 (78.62)	406.88 (18.06)	-	-	-	-	-
25 Jul		189.65 (35.58)	88.18 (6.22)	*35.47 (9.17)*	*19.95 (28.21)*	-	-	-
01 Aug		278.84 (27.42)	16.77 (10.62)	-	59.22 (16.5)	-	-	-
10 Aug		619.33 (140.95)	-	-	*19.41 (1.29)*	-	-	-
17 Aug		343.99 (50.32)	-	-	-	-	-	-
05 Jul	East	-	-	-	-	-	-	-
11 Jul		*18.59 (26.29)*	-	-	-	-	-	-
19 Jul		-	763.25 (154.17)	120.97 (1.56)	97.26 (36.16)	-	-	-
25 Jul		174.25 (34.91)	72.07 (23.34)	*27.59 (2.67)*	-	-	-	-
31 Jul		188.58 (108.52)	-	-	-	-	-	-
10 Aug		411 (30.56)	-	-	-	-	-	-
17 Aug		856.77 (37.15)	-	-	-	-	-	-
11 Jul	West	51 (24.31)	-	-	-	-	-	-
19 Jul		160.79 (14.53)	211.23 (23.56)	-	-	-	-	-
25 Jul		33.89 (25.24)	106.53 (28.81)	*30.03 (8.44)*	-	-	-	-
31 Jul		242.35 (29.74)	-	-	-	-	-	-
10 Aug		471.57 (75.91)	-	-	*26.43 (37.38)*	-	-	-
17 Aug		846.72 (0.53)	-	-	-	-	-	-

Abbreviations: anabaenopeptin A (AP-A), microcystin (MC-), anatoxin (ATX), homoanatoxin-a (HATX), cylindrospermopsin (CYN), cyanopeptolin-a (CPA)

571

572 **Figure legends**

573 **Figure 1**. Watershed and bathymetric (*a*) map of Conestogo Lake, Mapleton Township, Ontario
574 with sampling sites for the west arm (CLW), center (CLC), and east arm (CLE) of the reservoir.
575 Bathymetric data is scaled to the maximum dam stage elevation (meters at sea level; m.a.s.l.).
576 Total precipitation in the Upper Conestogo watershed (*b*, gauge in Arthur, ON) is directly related
577 to the mean daily flow rate (*c*) into the east arm (gauges in Drayton and Moorefield, ON) and the
578 calculated residence time (*d*) in the reservoir. Average flow rate (red line, *c*) is typically near
579 0.01 m/s. An intense rainfall event on 23-24 Jun (grey vertical bar) resulted in a notable increase
580 in the mean daily storage volume (1000 m^3) of the reservoir (*e*). Dark gray bars represent
581 standard deviation. This figure contains information made available under GRCA's Open Data
582 Licence v3.0 (panel a) and v2.0 (panels *b* - *e*).

583

584 **Figure 2.** Multiplicative changes in measured water chemistry between samplings before (21 Jun
585 2017) and after the flood event (05 Jul 2017) at 2, 7, and 16 m depths at the center site. Values
586 above 1 correspond to increased concentrations while those below 1 indicate reduced
587 concentrations. (*a*) Total phosphorus (TP), total dissolved phosphorus (TDP), and soluble
588 reactive phosphorus (SRP), (*b*) anions, (*c*) Nitrogen species including total dissolved nitrogen
589 (TDN), nitrate (NO_3^-), nitrite (NO_2^-), ammonium (NH_4^+), and dissolved organic carbon (DOC),
590 and (*d*) dissolved cations.

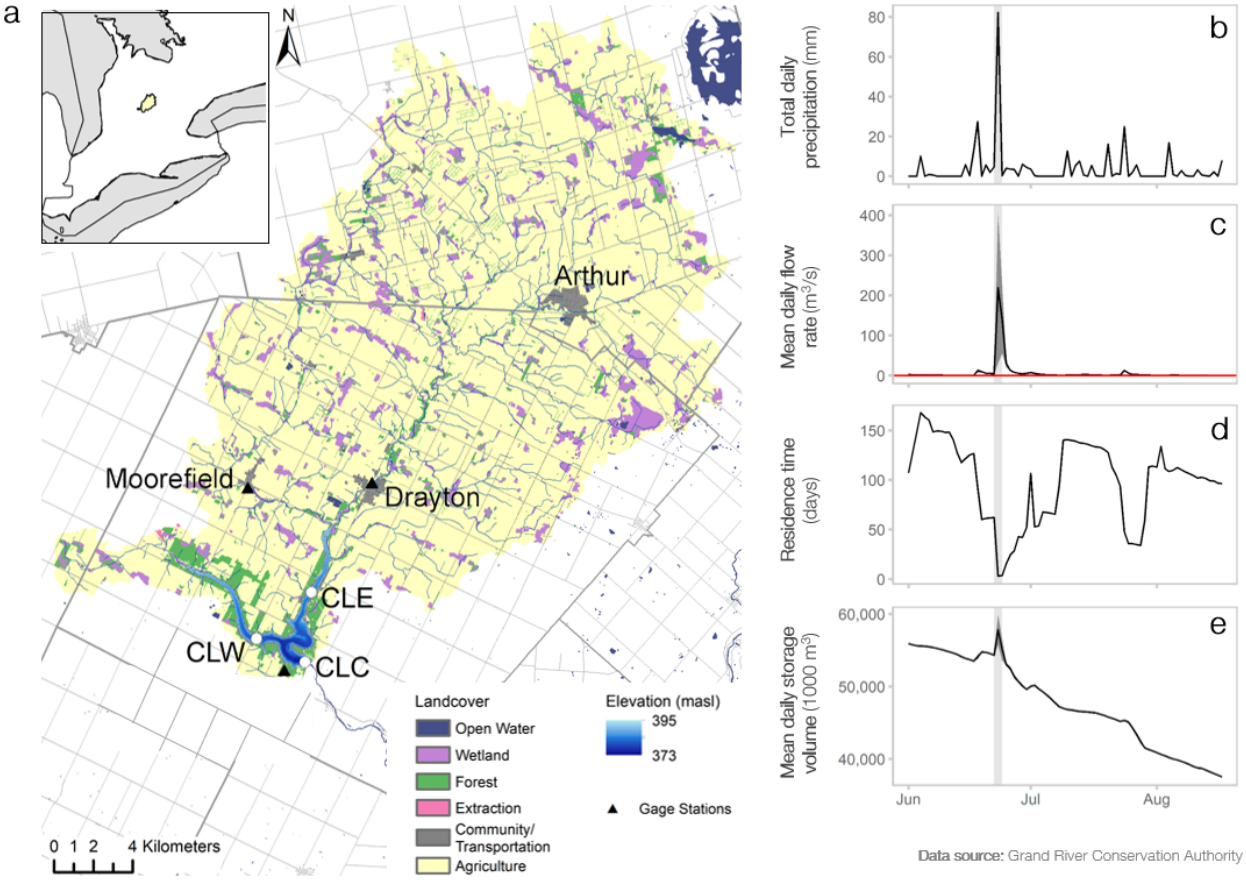
591

592 **Figure 3.** Nutrient profiles for phosphorus (*a*; P, µg-P/L) and nitrogen species (*b*; N, mg-N/L)
593 collected at 2, 7, and 0.5m from the bottom at each of the sampling locations. The flood event
594 (white vertical bar) introduced increased levels of all measured P species but did not greatly

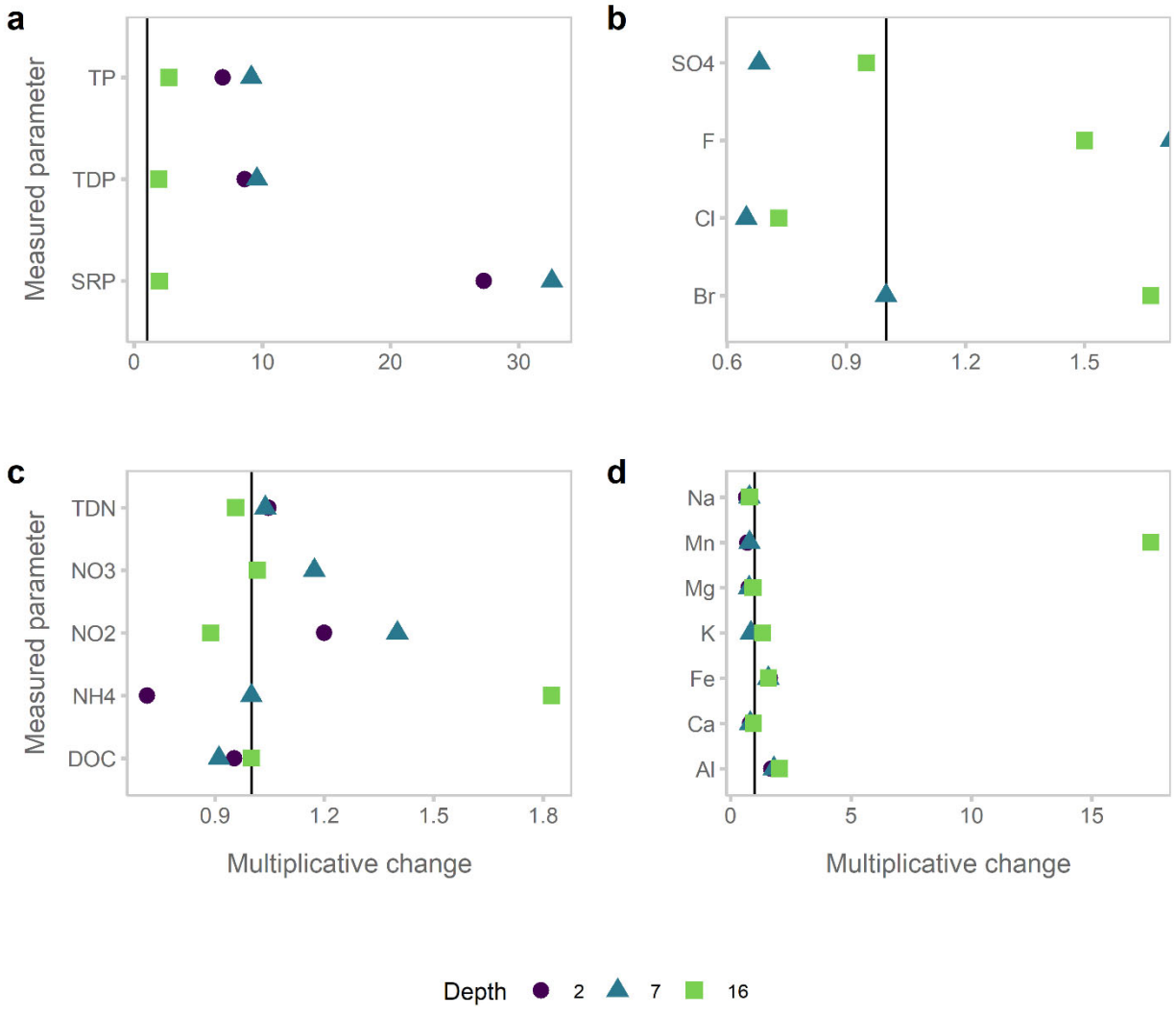
affect N species (see Figure S1). The dynamic area of the colored box reflects the changing depths within each sample location. Profile depths at each site were standardized to the dam stage elevation (m) at the time of sampling and illustrate both the differences in bottom depth and reservoir drawdown during the sampling campaign.

Figure 4. Cyanobacterial biomass (*a*) and cyanotoxin concentration trends > LOQ for anabaenopeptin-A (AP.A) and microcystin-LR (MC.LR), -YR (MC.YR), and -RR (MC.RR) (*b*) in each of the three sampling locations (*l* to *r:* West, Center, and East) were detected approximately 2 weeks following the flood event on 23-24 Jun (DOY 172; grey vertical bar). *Aphanizomenon flos-aquae* dominated (> 50%) phytoplankton biomass in the west and east arms of Conestogo Lake near 10 Jul 2017 (DOY 191) and in the center near 16 Jul 2017 (DOY 197). The bloom period lasted between 26-34 days depending on the sampling site with a site-specific peak bloom biomass occurring near 25 Jul 2017 (DOY 206). Of the seven cyanobacterial species detected in Conestogo Lake, various strains of *A. flos-aquae*, *D. flos-aquae* (*Anabaena flos-aquae*), *M. aeruginosa*, and *W. compacta* have been documented as potential toxin producers.

Figure 1

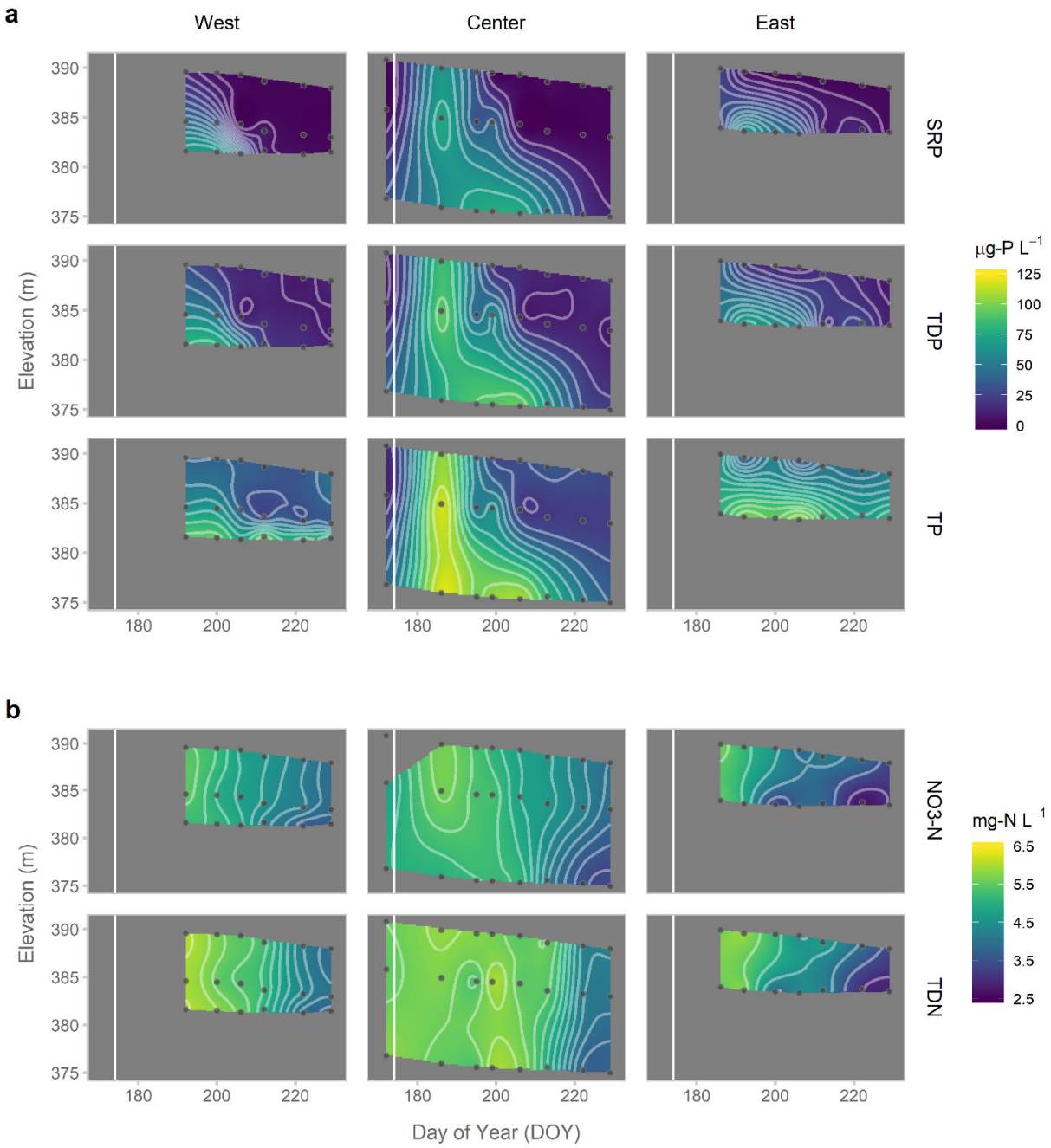

a
N
Arthur
Moorefield
Drayton
CLE
CLW
CLC
Landcover
Open Water
Wetland
Forest
Extraction
Community/ Transportation
Agriculture
Elevation (masl)
395
373
Gage Stations
0 1 2 4 Kilometers
b
Total daily precipitation (mm)
80
60
40
20
0
c
Mean daily flow rate (m³/s)
400
300
200
100
0
d
Residence time (days)
150
100
50
0
e
Mean daily storage volume (1000 m³)
60,000
50,000
40,000
Jun
Jul
Aug
Data source: Grand River Conservation Authority

614 **Figure 2**



615

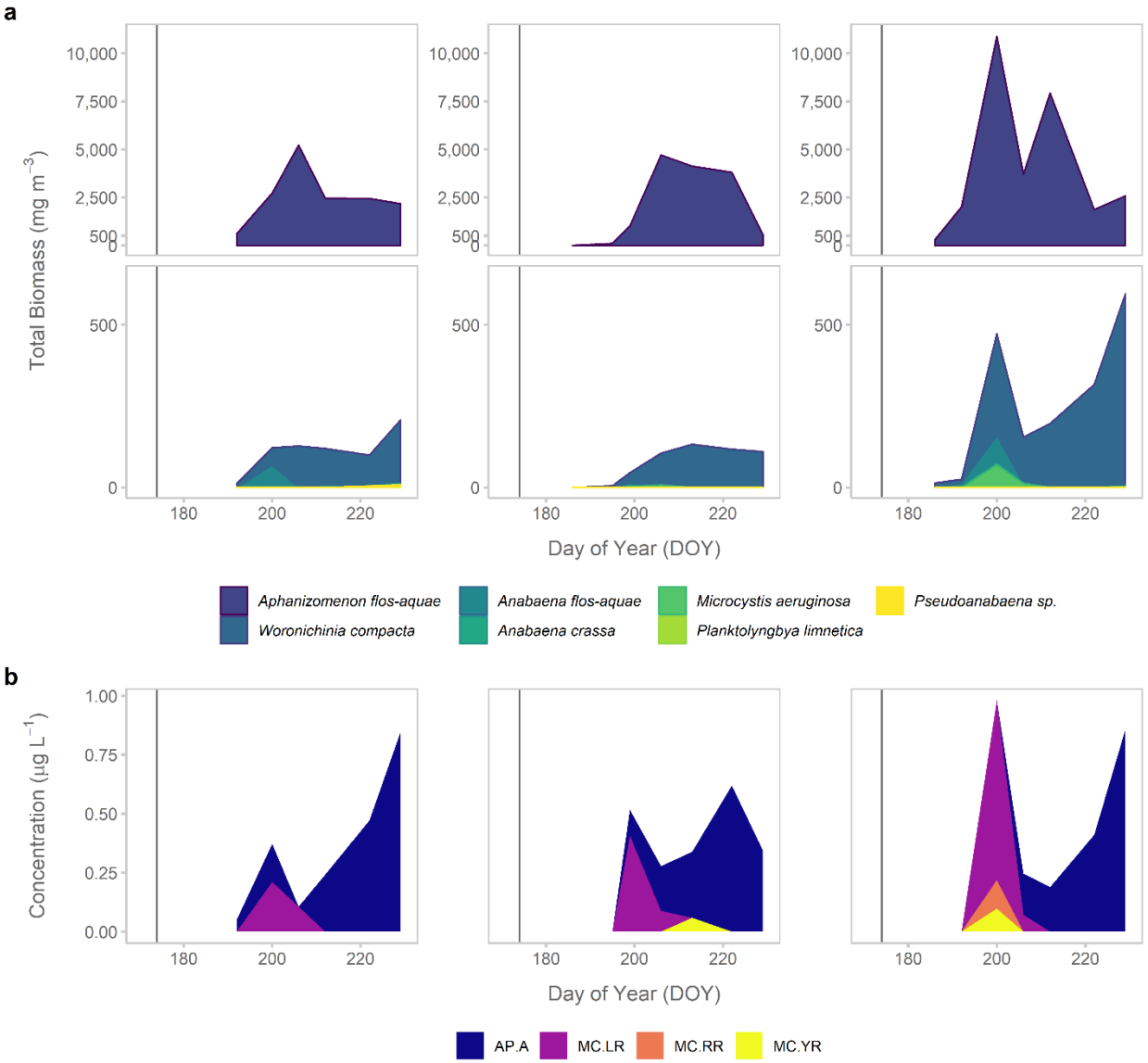
616 **Figure 3**



617

618 **Figure 4**

619



620