# Monitoring and Determining the Source of Cyanobacteria Blooms in Leonard Lake



Prepared for the Leonard Lake Stakeholders Association (LLSA)

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\*cover photo: Leonard Lake, August 2022, Google Earth satellite imagery

### Summary

Prepared by Members of the Leonard Lake Stakeholders Association

During the 2022 season, intensive investigations were undertaken by members of the LLSA water team, with advice from Dr. Mark Verschoor, in order to gain a better understanding of the source of recurrent cyanobacteria blooms which have been reported yearly since 2017. As Leonard Lake is classified as an oligotrophic (low nutrient) lake, such blooms were not considered possible by limnologists and policymakers before. Yet the frequency of reported blooms not only in Leonard Lake but in other oligotrophic lakes throughout northern Ontario has been rapidly rising for several decades (Winter et al 2011).

This report includes a background review of historical and recent investigations, which point out a number of underlying concerns, such as increasing levels of dissolved organic carbon and turbidity (cloudiness), increased chloride levels from road salt, the introduction of the invasive spiny water flea, thick algal mats reported in 1971 along the Southwestern shores attributed to early cottage/resort development, and lack of rigorous testing of septic systems. The Watson and Kling study (2017) concluded that "there are now indications that the lake is moving towards a mesotrophic (increased productivity) status". This report also questions the validity of long term total phosphorus (TP) data measured in the spring at a few mid-lake sites to determine the true trophic status (productivity) of the lake.

The 2022 investigations included several components: water chemistry and algal sampling in summer and early fall at the same four deep spots studied in 2017 to detect and confirm anoxia, internal loading, and the presence or dominance of bloom-forming cyanobacteria; nearshore investigations in the areas where blooms have occurred using fluorimetry to detect and monitor cyanobacteria populations via their specific pigments (with confirmation of bloom-forming species using microscopic examination) while also monitoring any changes in nutrient levels before or after bloom events; and finally, locating and sampling spring and fall runoff points from 12 intermittent streams to obtain an estimate of the nutrient (phosphorus) loading into the lake, and in spring 2023, salinity (chloride) pollution from road salts. These investigations resulted in several important findings that strongly indicate, but do not fully confirm the source of blooms:

- Anoxia and internal loading (nutrient release) was found at both South Bay and Midlake sites for the first time, and the degree of anoxia and internal loading increased from July to September.
- Anoxia, but not internal loading occurred at the NDH site, while Outlet Bay remained free of anoxia.
- Deep chlorophyll maxima (elevated levels of chlorophyll) were found at the bottom of the visible depth (5-6m) at the sites where internal loading was occurring.
- At the South Bay site, extensive anoxia and elevated nutrients coinciding with the presence of the bloom-forming species *Dolichospermum lemmermannii* at the bottom, makes this site a probable source of blooms.
- This investigation revealed the power of the LLSA fluorimeter to easily and rapidly measure the telltale cyanobacteria pigment, phycocyanin, and identify the pre-bloom, full bloom, and post-bloom duration and intensity of several bloom incidents on our lake in 2022.
- Elevated levels of total phosphorus up to 10 times the level in the lake, were found at several of the 12 runoff points, indicating that runoff was likely an additional contributor of bloom nutrients.
- Elevated levels of chloride over an order of magnitude found within the lake were detected at several spring runoff samples located closest to Highway 118, indicating that road salt is the primary contributor to increased salinity of Leonard Lake.

Several recommendations for further studies follow from these findings:

- Since the South Bay is the most probable source of blooms, further sampling should focus on this site.
- Efforts should be made to track the movement of nutrient/bloom material from this site to areas along the western shore where the blooms occur, using net hauls, fluorimetry, and microscopic examination.
- Check for the abundance of the spiny water flea using net hauls and microscopy.
- Continue runoff studies sampling both total phosphorus (TP) and chloride. Make efforts to track the source of the elevated nutrients flowing into the lake from the runoff points. Approach the Township of Muskoka Lakes to increase their septic monitoring to insure septic runoff is not the source.
- Continue shoreline monitoring and "Eyes on the Lake" reporting of blooms.
- Continue to collect data from fluorometry and microscopic examination on samples to asses pre-bloom, bloom, and post bloom conditions.
- Gather information and hold discussions about possible mitigation measures to address the anoxia in South Bay.
- It is probable that human activities and development are playing a role in bloom occurrence and deteriorating water quality on Leonard Lake; continue to educate, inform, and advocate for all who use the lake to undertake best practices to reduce their impacts.
- Advocate for stronger locally-developed policies for managing our lake health.

### Background

Leonard Lake is located in the Algonquin-Lake Nipissing area, above Lake Muskoka, within the Boreal Shield (45.0751, -79.4496). It has a smaller, shallow south basin with a maximum depth of 8.5m, and a larger and deeper northern basin (maximum depth 16-17.5 m), roughly divided by two islands (Fig.1). Its shoreline is largely composed of exposed bedrock and stones, with a few wetland areas. The catchment area is small (4.19 km<sup>2</sup>) and composed of Precambrian bedrock covered by a thin (< 1.5 m) layer of sandy till and rocky outcrops. Inputs occur largely from precipitation as direct runoff and small intermittent streams, with the contribution of groundwater being unknown (DMM 2015; Ingram and Patterson 2015; Watson and Kling 2018). A single small outflow at the western side is also intermittent, giving the lake a long turnover rate (20% of the total volume renewed each year, residence time of 5.4 years) (Nurnberg 2017).



Fig 1. Bathymetry of Leonard Lake.

Leonard Lake has a low buffering capacity, and was significantly affected by acid precipitation from the 1900s until the 1990s. Since then, it has slowly been recovering, and sulphate (a component of acid rain) has been decreasing (Ingram and Patterson 2015).

Water clarity is relatively high, with a spring Secchi depth of 3–4.5m, which can theoretically support macrophyte and algal growth down to 7.5-9m, thus making the entirety of the South basin capable of high levels of production.

Dissolved organic carbon (DOC), which contributes to the brown colouration of water, decreases light penetration, and acts a carrier for dissolved nutrients, has been steadily increasing, and is thought to be due to the effects of climate change: warmer temperatures increase production and decomposition of organic matter within soils, which then leach into lakes during rain events (Ingram and Paterson 2015).

The use of road salt has very significantly increased both sodium and chloride concentrations in Leonard Lake, due to the presence of year-round road usage along both shores (Ingram and Patterson 2015). More recent data (this report) indicates that the source of this salt is Highway 118 West, which runs near the southern tip of the lake. High levels of chloride, sodium, heavy metals, and hydrocarbon pollutants from motor vehicles are toxic to zooplankton (algae grazers), and can decrease their survival rates (Van Meter et al 2011, Hintz et al 2019, Arnott et al 2020). Another factor which is likely affecting the zooplankton population is the presence of the invasive spiny waterflea (*Bythotrephes longimanus*), first detected in Leonard Lake in 2001, which preys on smaller keystone zooplankton and is inedible to many natural predators (Yan et al 2011, Watson and Kling 2017). Together, these stressors can significantly reduce the biodiversity of zooplankton which modulate phytoplankton populations, thus reducing the robustness of the food web by creating unstable ecological services that are prone to dramatic shifts or collapse, resulting in the formation of blooms.

Early surveys (OWRC 1971) indicated generally oligotrophic lake conditions (low total phosphorus (TP) levels, low productivity, and high water quality and clarity) but showed early signs of eutrophication (enhanced productivity) with the formation of thick algal mats along the more developed southwestern shores of the lake, and the study concluded that the lake was "largely unsuited for cottage development with subsurface septic systems." It should be noted that the effectiveness of septic systems on Leonard Lake, with its thin soils, has not been rigorously tested. Since then, developed lots have more than doubled, and there are indications that the lake is tending towards mesotrophic (increasing productivity) status (Watson and Kling 2017).

TP levels have been monitored by several agencies (District Municipality of Muskoka (DMM), the Ontario Ministry of the Environment, Conservation, and Parks (MECP), Ministry of Natural Resources (OMNR), the provincial Lake Partnership Programme (LPP), and Muskoka Lakes Association (MLA)), and have indicated no significant long-term change in water quality. However, it should be noted that these samplings occur at central offshore sites, often during the spring, and usually around 1 Secchi depth (3-4 m), but they likely do not adequately represent the true trophic status of the lake: water quality can vary significantly in shallower or sheltered inshore areas, most bloom activity occurs in the late summer to fall, and nutrient levels can be much higher later in the season, and especially towards the bottom, where oxygen levels become anoxic (<1 mg/L), producing conditions conducive for internal loading.

The effects of internal loading produce high levels of nutrients, such as organic acids, dissolved phosphorous, nitrogen, and trace metals: the sediments of the lake bottom stagnate, decay, and release soluble material that support algal growth. These materials (nutrients) are then recycled when algal mass settles to the bottom at the end of the season, thus creating many cycles of nutrient availability. Typically, it can take as long as 50 years for this material to be flushed out and buried deep enough (under subsequent layers of sediment) to become inactive, which is known as the "legacy" effect of pollution (Molot et al 2014). A high lake flushing rate can partially mitigate this effect, but since Leonard Lake has a low flushing rate, it is expected that recovery from any pollution sources will be slow.

Moreover, detailed oxygen and nutrient profiles (Nurnberg 2017, Watson and Kling 2017, and this study) demonstrate that anoxia and internal loading occur in several areas of the lake, including the South Bay, where enhanced levels of nutrients are available from the bottom all the way to the euphotic (light levels that support algal and plant growth) zone. Additionally, direct resuspension of sediments from motor boats in shallow areas such as South Bay can also release particle-bound phosphorous and nitrogen (Watson and Kling 2017). This strongly suggests that this area may be a major source of cyanobacterial blooms.

Evidence that internal loading of nutrients supports enhanced growth of algae can be surmised by the formation of relatively dense layers of algae far below the surface, known as deep chlorophyll maxima (DCM). These layers are located via fluorometric probes, which detect increased concentrations of algal pigments (chlorophyll, phycocyanin, etc.) at specific depths. An increase in dissolved oxygen from active photosynthesis usually accompanies these DCMs as well. Typically, these DCMs first form at the thermocline, where the colder bottom waters meet warmer surface waters, creating a physical density barrier, thus trapping material settling from the surface. As the season progresses, nutrients from internal loading migrate upwards (and some forms of algae, notably some cyanobacteria, can migrate

between these zones by controlling their buoyancy), and the algal biomass of the DCM thus increases. Additionally, cyanobacteria have the ability to photosynthesize at lower light levels more efficiently than many eukaryotic (green, brown, and otherwise non-toxic, food web-supporting) algal species, and are stimulated by trace metals, such as iron (Molot et al 2014, ). Thus, DCMs may be reservoirs of algal biomass that eventually form blooms.

Reports of potentially harmful algal blooms consisting of cyanobacteria (cHABs) at shoreline sites began a few years prior to 2017, and have been documented each year since then. Moreover, the blooms seem to recur in the same areas. This prompted cottage owners on the lake, members of the Leonard Lake Stakeholders Association (LLSA) to fund and carry out extensive water chemistry and phytoplankton sampling at several mid-lake and near-shore sites between May and October 2017, followed by a detailed report of the findings by noted research scientist Sue Watson of the Canada Centre for Inland Waters, and algal taxonomist Hedy Kling (Watson and Kling 2017).

The main findings of that study indicated that Leonard Lake had low to moderate productivity and a diverse algal community dominated by diatoms, flagellates, green algae, and small cyanobacteria, which are high quality contributors to the food-web, however, low levels of the potentially toxic bloom-forming cyanobacteria *Dolichospermum* were detected throughout most of the lake during the entire sampling period. Furthermore, a concurrent LLSA "Eyes on the Lake" campaign reported and collected ten samples from possible near-shore bloom sightings, of which four were confirmed to consist chiefly of *Dolichospermum lemmermannii*.

Nutrient levels measured at four sites in the lake were highly variable with depth and over the season, often exceeding long-term averages measured by provincial and regional agencies, who focus primarily on spring composite sampling at a single mid-lake location. This implied the presence of internal loading, which was accompanied by anoxic conditions near the bottom of several sites in the lake, not only at the deepest part of the lake in the Northern basin, but notably in South Bay, where many of the blooms occurred.

Finally, the 2017 report recommended continued monitoring of water chemistry, phytoplankton composition, and timely sampling and reporting of blooms, standardizing site locations and sampling and testing protocols in conjunction with provincial and regional agencies, limiting boat speeds in South Bay, restricting shoreline development to maintain a vegetated buffer strip and minimizing runoff from lawns and roads, and suggested a moratorium on further lakefront lot development, because the effects of climate change would likely make the lake even more vulnerable to blooms, as evidenced by the lakewide golden brown algae (chrysophyte) bloom on May 2020 that lasted over a month.

## LLSA Study, 2022

# Objectives

Following the recommendations of the 2017 study, and to possibly determine the source of blooms in Leonard Lake and how the blooms might eventually be managed, the following sampling strategy was devised:

- 1. Continue monitoring efforts and compare water column chemistry and algal taxonomy at the same four mid-lake sites used in the 2017 study to detect and confirm anoxia and internal loading, and the presence or dominance of cyanobacteria (especially *Dolichospermum lemmermannii*) which could contribute to blooms.
- 2. Locate, monitor, sample, and perform taxonomic analysis on Deep Chlorophyll Maxima to determine their composition and relative contribution to bloom formation.
- 3. Sample adjacent shoreline areas with a history of recurrent blooms both before and after bloom appearances in order to detect nearshore populations of cyanobacteria (esp. *Dolichospermum lemmermannii*) and any factors (TP, water temperature) that may explain their presence.
- 4. Identify and sample active runoff points around the lake during spring and fall rainfall events in order to determine if they are significant sources of external phosphorus.

# Methods

All field work, data collection, and processing was conducted by trained members of the LLSA, who had previous experience from 2017 onwards. Water monitoring occurred between May 24 and continued until Nov 4. Discrete-depth samples were collected on a monthly basis in May and June, then biweekly until November at four offshore and four shoreline sites, with the addition of twelve runoff sites, which were located in the spring during rain events (see Fig. 2).



Fig 2. Location of off-shore, shoreline, and runoff sampling sites in 2022. Off-shore abbreviations: NDH=North Deep Hole, OUT= Outlet Bay, MID= Mid-Lake, SB= South Bay. Sh1= Mouth of West Bay, Sh2= Western Shoreline, RO6 & Sh3= Boat Launch, Sh4= End of West Bay.

#### Off-shore Sites:

At the four offshore sites, Secchi depth and surface water temperature were measured. Sonde monitoring with a rented YSI EXO-1 was done on 3 occasions: July 26<sup>th</sup>/27<sup>th</sup>, August 23<sup>rd</sup>/24<sup>th</sup>, and September 13<sup>th</sup>/14<sup>th</sup>. Depth profiles for temperature, conductivity, pH, dissolved oxygen (DO), redox, chlorophyll-a, and phycocyanin (a pigment specific for the detection of cyanobacteria) were measured in 1 m or 0.5 m increments (as deemed appropriate). The temperature, DO, and chlorophyll data were plotted

immediately to locate the thermocline, depth of DCMs, and hypoxic/anoxic zones for chemistry sample collection respectively. When DCM data was not available, algae were assumed to be concentrated at the 2x Secchi depth.

Discrete-depth samples were collected using a horizontal Van Dorn sampler. For TP, samples were collected at 1m, 1x Secchi depth, 2x Secchi depth (or at the DCM when known), and 1m above the sediment (B-1), then coarse filtered through 80 micron mesh. Dissolved phosphorus (DP) and dissolved iron (DFe) samples were collected at 1m below the surface, and 1m above the bottom (B-1), and subsamples immediately filtered directly in the field into sample tubes using 0.45µm syringe filters. DFe samples were immediately acidified with a few drops of trace-metal grade nitric acid per 10 mL in plastic snap-cap tubes. Both DFe and phosphorous samples were shipped to the Biogeochemical Analytical Service Laboratory (BASL), Edmonton AB, for analysis.

Algae monitoring samples were collected at 1x and 2x Secchi depth with a composite sampler, and at the DCM (if known) with a Van Dorn sampler. Algae monitoring entailed fluorescence analysis with a FluoroQuik fluorometer (Amiscience, Calif.) to measure phycocyanin and chlorophyll concentration (µg/L) in water samples which had previously been frozen and thawed to ensure cell lysis.

In mid-September, samples from the DCMs of each offshore site (or a B-1 sample from South Bay) were collected for algal taxonomy and sent unpreserved in cooler packs or preserved in Lugols Iodine for analysis to ATEI, Winnipeg Manitoba using the standard Utermöhl technique (Findlay and Kling 1998).

#### Shoreline Sites and Blooms:

Samples were collected biweekly or occasionally weekly, with a container affixed to the end of a pole. Samples adjacent to or from within a bloom were collected more frequently. Aliquots of samples were frozen to ensure cell lysis, then the FluoroQuik fluorometer was used to determine phycocyanin (cyanobacteria biomass) and chlorophyll (total algae) concentrations in the samples. Samples were also collected for total phosphorus analysis in order to determine whether excess phosphorus in some areas contributed to the susceptibility of bloom formation. Blooms were reported to provincial agencies, and photomicroscopic documentation of bloom material was utilized to confirm algal species identification with algal taxonomists.

#### Run-off sites:

Run-off sites were identified after significant rainfall events. The flow of water was approximately classified as heavy, moderate, or low, and sampled with a container on a pole at a point just before it entered the lake. Unfiltered samples were then submitted for TP and analyzed for chlorophyll content (via fluorometer). Spring run-off samples were also analyzed for chloride.

## **Results and Discussion**

#### **Off-Shore Sites:**

Depth profiles revealed stratification at all four offshore sites (Figs 3-6). The water column was thermally stratified where the temperature abruptly declined at a depth of about 6-7 meters, forming an upper layer of warmer mixed surface waters (epilimnion), and a cooler deep water area of still water (hypolimnion). Between these two layers lies the thermocline, or metalimnion, forming a density gradient that traps particulate matter and algae, helping to form deep chlorophyll maxima (DCM).

As the season progressed, the thermocline deepened with the warming surface waters at all sites. Dissolved oxygen (DO) declined significantly with depth at all four sites, with concentrations decreasing to hypoxic levels (<4 mg/L) near the bottom of the water column by late August, and reaching anoxic conditions (<1 mg/L) at all sites by mid-September, except Outlet Bay, which never became anoxic.

DO levels were slightly increased at the thermocline, where DCMs exist, due to active photosynthesis, except for South Bay, which became anoxic early in the season, and well above the sediment surface, at around 5.5 meters. This anoxic zone overlapped the thermocline, and thus could have provided soluble nutrients from internal loading directly to resting algae in the DCM.

As a result of the anoxic conditions at the three sites that experienced them, internal loading was evident from the TP profiles, which showed significantly increased concentrations towards the bottom. Dissolved phosphorous (DP) was also slightly higher near the bottom, though the maximum concentration measured at any location was only  $7\mu$ g/L, possibly due to adsorption on the filter material, or precipitation after being exposed to oxygen in the atmosphere. Curiously, TP was slightly higher in July than in August at all sites, possibly due to uptake by phytoplankton.

Internal loading was also evident from the release of dissolved (ferrous) iron at B-1 (1 m above the sediment) at the three deep sites; as the dissolved oxygen levels declined over several months and anoxia increased, the release of soluble iron increased significantly. Molot and others (2014) have reported that release of ferrous iron appears to precede and facilitate bloom events. In 2022, the spike in dissolved iron at the sediment in mid-September was followed by more persistent blooms in October to November despite water temperatures being below 15 °C.

# LL South Bay



Fig 3. Temperature (Temp), dissolved oxygen (DO), and total phosphorous (TP) profiles of South Bay during 2022.

# LL Mid-Lake



Temperature (°C), Dissolved Oxygen (mg/L), TP (ug/L)

Fig 4. Temperature (Temp), dissolved oxygen (DO), and total phosphorous (TP) profiles of the Mid-Lake site during 2022.

# LL NDH



Temperature (°C), Dissolved Oxygen (mg/L), TP (ug/L)

Fig 5. Temperature (Temp), dissolved oxygen (DO), and total phosphorous (TP) profiles of the NDH site during 2022.

# LL Outlet Bay



Temperature (°C), Dissolved Oxygen (mg/L), TP (ug/L)

Fig 6. Temperature (Temp), dissolved oxygen (DO), and total phosphorous (TP) profiles of the Outlet Bay site during 2022.

Timelines of DCM pigments (chlorophyll a and phycocyanin), dissolved iron (DFe), and bloom occurrences were constructed for each site (Figs 7-10) in order to elucidate the relative contribution each site may have had on bloom formation. For South Bay, the steady release of iron and increase in phycocyanin from late July onwards, followed by a bloom in late August and then several subsequent blooms, suggests that the high degree of internal loading at this site might make a significant contribution. The Mid-Lake site may also indicate the same mechanism, though there is a significant bloom event and high levels of phycocyanin long before there is significant release of iron. Another bloom occurs within a few weeks of maximum iron release from this site, though phycocyanin concentrations appear to be declining at this site prior to the bloom, possibly suggesting another source, or that the cyanobacteria had relocated within the water column, possibly becoming benthic before rising up later to form the bloom. It is notable that both South Bay and Mid-Lake produced the highest levels of dissolved (reduced) iron of all sites. The NDH timeline, with low iron production and peak phycocyanin levels that were asynchronous with known bloom events, suggests that rather than being a source of blooms, the NDH site may be a sink, where prevailing winds may transport previous generations of cyanobacteria to settle at its thermocline. The Outlet Bay site did not appear to have much if any internal loading at all, and accordingly, changes in phycocyanin concentrations did not seem to explain subsequent bloom events at this site.



Fig 7. Timeline of internal loading, major bloom events, and algal growth at the South Bay DCM.



Fig 8. Timeline of internal loading, major bloom events, and algal growth at the Mid-Lake DCM.



Fig 9. Timeline of internal loading, major bloom events, and algal growth at the NDH DCM.



**Outlet Bay B-1 Dissolved Iron & DCM Pigments** 

Fig 10. Timeline of internal loading, major bloom events, and algal growth at the Outlet Bay DCM.

Previous experience in the field has shown that decreases in Secchi depth can often precede a bloom as cyanobacteria migrate towards the surface from DCMs or benthic environments, and significant changes and differences in Secchi depth among the sampling sites were accordingly noted while working with the data (Fig. 11). For example, significant declines in Secchi depth (increases in turbidity) were noted in all sites, but particularly at the Mid-Lake site prior to the mid-July bloom, which occurred directly adjacent to that site. The late August bloom, however, is not preceded by nor coincides with a decrease in offshore Secchi depth, as it occurred in the southernmost end of the lake, at the boat launch, and most likely is a result of nearshore populations. A series of blooms in the western bays of the lake in October was preceded by very sharp declines in Secchi depth in all sites, beginning with the Mid Lake and NDH stations. These changes may simply reflect overall increases in lakewide productivity, and attaching the origins of a bloom to a specific site would require direct evidence of the organism involved.



Fig 11. Timeline of changes in Secchi Depth at each offshore station and the occurrence of major blooms during the 2022 sampling season.

Accordingly, the deep chlorophyll maxima (DCM) were monitored throughout the season, and analysed for phycocyanin and chlorophyll fluorescence, with a subset of samples filtered with 50 micron mesh to differentiate picocyanobacteria from larger colonial and filamentous cyanobacteria. Samples at the peak of DCM development which also showed the highest phycocyanin fluorescence after 50 micron filtration were submitted (unfiltered fresh and Lugol-preserved) for taxonomic analysis to determine if *Dolichospermum lemmermannii*, the species identified in all of the shoreline blooms since 2017, could be traced to the DCMs, and by extension, to the anoxic sites on the lake. Table 1 summarizes the aforementioned investigation, which revealed that although *Dolichospermum planktonicum* and other species of cyanobacteria were found in the DCMs, *D. lemmermannii* was only found near the sediment in South Bay, thus strongly suggesting that the source of the blooms may originate from that site.

	South Bay		Outlet Bay		Mid Lake		NDH	
	Sept 13, 8m (B-1)		Sept 14, 7m (DCM)		Sept 14, 7m (DCM)		Sept 23, 7m (DCM)	
Taxon	Biomass (mg/m³)	% cyano Biomass						
Dolichospermum lemmermannii	102.4	70	absent	_	absent	_	absent	_
Dolichospermum planktonicum	absent	-	92.6	33	102.6	37	171.7	66
<i>Planktolyngbya</i> sp.	10.1	7	90.9	32	67.5	25	16.5	6
Pseudanabaena sp.	absent	-	absent	-	absent	-	3.9	2
Anathece sp.	absent	-	61.2	21	58.9	21	14.4	5
Picocyanobacteria	not counted	-	15.1	5	30.1	11	40.3	15
Benthic cyanobacteria	31.4	21	absent	-	absent	-	absent	-
Other cyanobacteria	3.4	2	24.9	9	15.5	6	15.3	6
Total cvanobacteria	147.3		<b>284.</b> 7		274.6		262	

Table 1. Summary of major pelagic and/or benthic algal, cyanobacteria, and bacterial taxa identified in B-1 and DCM offshore sites collected from Leonard Lake in September 2022.

#### Shoreline Sites and Blooms:

Routine monitoring of several shoreline sites made it possible to track changes in the population of algae in response to temperature, TP, or other localized factors (Figs. 12-15). Surface water temperature did not seem to explain cyanobacterial growth, as peaks in phycocyanin did not match peaks in temperature, and often blooms occurred later in the season, when temperatures were generally falling. Likewise, TP seemed to be a poor predictor of cyanobacterial growth and bloom formation in these areas: there were no phycocyanin peaks nor blooms at any of the shoreline sites that followed elevated TP levels. Rather, elevated TP levels seemed to coincide or follow shortly after blooms in the West Bay, probably due to the bloom biomass itself. These results suggest that the bloom material originates farther from the shoreline and may be introduced and concentrated at these sites via wind and wave action.

The fluorescence readings themselves provided great utility, and are capable of providing predictive value: in many cases, phycocyanin levels began to rise several weeks before a bloom occurred. Phycocyanin readings were generally less than 10  $\mu$ g/L, with readings greater than 20  $\mu$ g/L indicating the start of colony formation. Visible evidence of blooms was usually associated with readings greater than 100  $\mu$ g/L, and surface scums can generate phycocyanin readings >10,000  $\mu$ g/L.

The exact origins of the cyanobacteria are not clear, but some possibilities suggested by field observations and forensic weather pattern analysis include Southeasterly winds transporting populations from South Bay into the Western bays, or local populations in these bays being stimulated by nutrient inputs following precipitation events, which seems to be the case for many of the blooms in October.



Fig 12. Timeline of changes in algal biomass (as inferred by chlorophyll (Chla) and cyanobacterial phycocyanin (Phyco) fluorescence) and possible relationships to temperature (Temp) and total phosphorous (TP) at the West Bay mouth shoreline site (Sh1). The occurrence and location of major bloom events are also indicated.



Fig 13. Timeline of changes in algal biomass (as inferred by chlorophyll (Chla) and cyanobacterial phycocyanin (Phyco) fluorescence) and possible relationships to temperature (Temp) and total phosphorous (TP) at the Western Shoreline site (Sh2). The occurrence and location of major bloom events are also indicated.



Fig 14. Timeline of changes in algal biomass (as inferred by chlorophyll (Chla) and cyanobacterial phycocyanin (Phyco) fluorescence) and possible relationships to temperature (Temp) and total phosphorous (TP) at the Boat Launch shoreline site (Sh3). The occurrence and location of major bloom events are also indicated.



Fig 15. Timeline of changes in algal biomass (as inferred by chlorophyll (Chla) and cyanobacterial phycocyanin (Phyco) fluorescence) and possible relationships to temperature (Temp) and total phosphorous (TP) at the West Bay End shoreline site (Sh4). The occurrence and location of major bloom events are also indicated.

#### Run-off sites:

The identification and sampling of active runoff points around the lake during spring and fall rainfall events revealed very significant sources of external phosphorus and other nutrients. Total phosphorus concentrations ranged from 10-77  $\mu$ g/L, and chlorophyll levels were up to 10 times higher than those measured in the lake water. Moreover, most of the sites on the western and southern end of the lake exhibited inputs more than twice the provincial water quality objective (PWQO) of 20  $\mu$ g/L, and incidentally, are adjacent to the same areas where blooms have repeatedly occurred (see Fig. 16). These areas are also the most developed, where the road is close to the shoreline, in contrast to the eastern half of the lake.



Fig 16. Historical location of blooms (triangles) compared to location, flow rate (colour), and output of total phosphorous (diameter of circles and value in  $\mu$ g/L) at the runoff sites (RO#) found around Leonard Lake in October (base map source: District Municipality of Muskoka, data format courtesy of LLSA).

Chloride levels in Leonard Lake have ranged between 4.5 – 5.2 mg/L from 2007 to 2022. The run-off sites were again sampled in April 2023, soon after ice-out, to determine chloride input. Two nearshore sites, also sampled at the same time, had chloride concentrations of 4.4 mg/L. Six of the twelve run-off sites had chloride levels of <1 mg/L, while two had levels between 1 and 4mg/l; however, sites 5, 6, and 7 (all situated in South Bay) had chloride levels exceeding the concurrent lake level: 6.8 mg/L at RO5, 74.3 mg/l at RO6, and 40.2 mg/L at RO7. All three sites are situated in close proximity to Highway 118 West, especially RO6 (the boat launch); RO7 drains some swampy areas near the highway. The dramatic increases in chloride levels from runoff near the highway provides strong evidence that the use of road salt along Highway 118, which passes through part of the Leonard Like watershed, likely causes elevated salinity in Leonard Lake. Increased salinity has been shown to decrease survival rates for beneficial zooplankton, including *Daphnia* species that feed on cyanobacteria (Van Meter et al 2011, Hintz et al 2019, Arnott et al 2020). Thus the elevated salinity from runoff near the highway may be an additional cause of blooms.

#### **Conclusions and Recommendations**

Off-shore investigations have clearly shown internal loading at several sites within Leonard lake, and the formation of deep chlorophyll maxima (DCM), which contain significant populations of potentially bloom-forming cyanobacteria. In particular, only South Bay harbors *Dolichospermum lemmermannii*, which is the species responsible for most of the blooms encountered on the lake. It follows therefore that South Bay should be monitored frequently at several fixed depths (including surface samples) and perhaps at an additional northwesterly site via fluorometry and microscopic examination (for presence/absence of *D. lemmermannii*) in order to track wind and current-borne cyanobacteria and determine the role of South Bay as a source of blooms.

Shoreline monitoring forms the core of efforts to document and report bloom activity and should be continued and extended to nearshore sampling via boat or canoe using fluorometry and net tows (to concentrate and thereby increase the detection rate of *D. lemmermannii* via microscopic examination) in the shallow areas of the bays continuously affected by blooms in order to detect populations of *D. lemmermannii* that may have their origin in these areas. Surficial sediment analysis for akinetes (spore-like resting cells) may also provide evidence for the presence of localized seed populations that can be periodically activated and otherwise overlooked.

Initial identification and investigation of run-off sites as a source of TP and other nutrients strongly suggest that they may be key contributors to bloom formation and expansion. Further studies should be conducted to verify and standardize the quantification of these inputs, and might include additional analysis to determine amendable aspects of their source, such as septic effluent, lawn fertilizers, disturbances from traffic (including detection of road salts and hydrocarbons, and development. Spoelstra et al (2017) have suggested the use of acesulfame and sucralose to detect septic effluent. Alternatively or additionally, sampling of the run-off sites might be extended to several points upstream in order to determine the specific area or land use type associated with elevated phosphorous levels.

Human activities have a noticeably detrimental effect on all types of fresh waters by disturbing their nutrient balance and chemistry, and reducing biodiversity, which is required for stable ecosystems (Yousef et al 1980, Hadley et al 2013). Leonard Lake currently has 165 cottages on a lake that has a mean depth of less than 7 m, a surface area of approximately 2 km<sup>2</sup>, and a slow rate of water replacement. In 2017, Watson noted that on 85% of developed lots, cottages were 30 m or less from the shore, and that the highest density occurred in the southwest region, which is the same area with the most bloom activity (Watson and Kling 2017).

Due to the global effects of climate change, the number and diversity of lakes that are at risk is increasing. Climate-related changes in seasonal ice coverage, lake water levels, and runoff and stratification patterns are key factors for increased incidences of cHABs in low-nutrient (oligotrophic) lakes (LeBlanc et al 2008, O'Neil et al 2012, Callieri et al 2014). Adding known detrimental disturbances such as increased boat traffic and land use changes due to development on top of the existing climate change scenario will likely create far more problems, which will be expensive to mitigate.

This small and sensitive headwater lake is already experiencing adverse events such as hypolimnetic anoxia (which also negatively affects the quality of the recreational fishery), algal blooms (which ultimately degrade intrinsic and property values), and nutrient inputs exceeding PWQOs. In the face of an uncertain and less stable future brought about by climate change, it would be more beneficial to invest more time and resources into understanding the reasons for, and developing solutions to these concerns, rather than introducing policies that may further exacerbate water quality issues.

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