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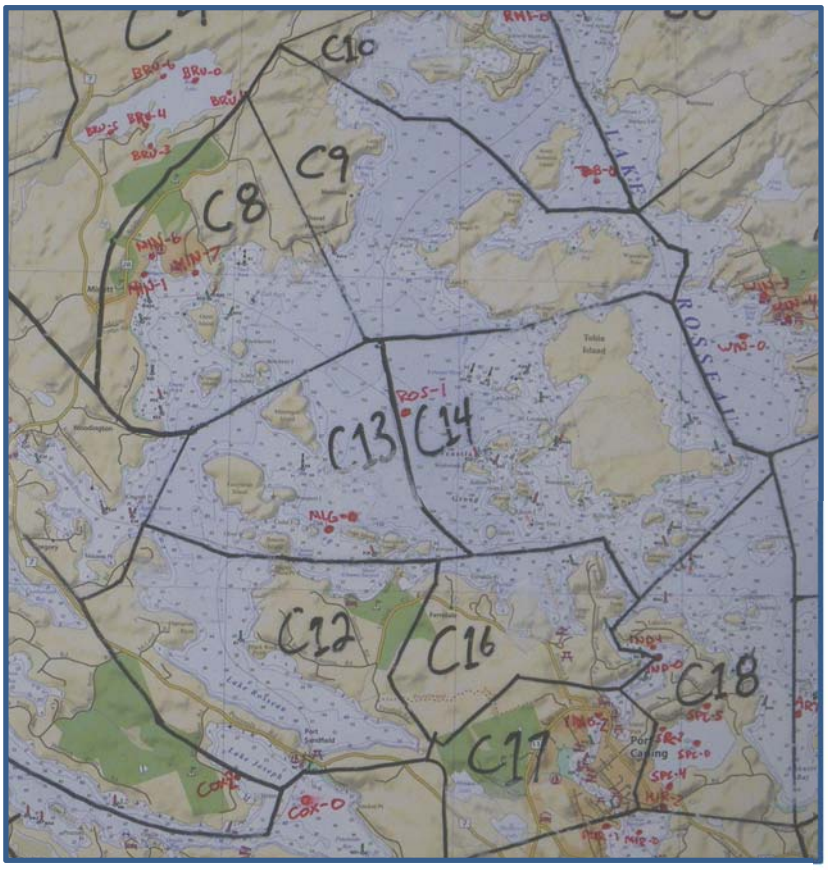
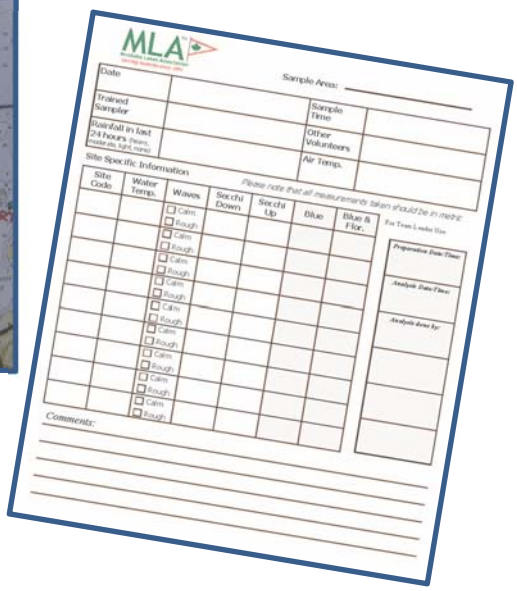
Water Quality Initiative Methodology





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Appendices

1. Background

The Muskoka Lakes Association (MLA) is a non-profit organization that was founded in 1894 to represent the interests of lakeshore residents on Lakes Rosseau, Joseph and Muskoka and many smaller surrounding lakes, and is presently Canada's oldest cottage association. The MLA objectives of monitoring phosphorus, Coliform and E. coli, providing data to protect vulnerable areas and promoting stewardship are carried out through the Water Quality Initiative (WQI).

The MLA's WQI is a science-based water quality monitoring program designed to measure key biological, chemical, and physical indicators of water quality in lakes throughout Muskoka. The WQI has been running since its inception in 2001 and updates and upgrades have improved the program through the years. The program is directed by the MLA Environment & Water Quality Committee, administered by support staff based at the MLA office in Port Carling, and implemented by a dedicated group of volunteers.

The MLA has adopted a long-term monitoring strategy for phosphorus, calcium, water clarity, and water temperature, and has recently (2013) introduced a monitoring strategy for dissolved organic carbon concentrations. Additionally, bacteria monitoring activities have focused on determining whether chronically elevated conditions exist in targeted nearshore recreation areas. This document provides a comprehensive overview of the monitoring program including sampling and analytical methodologies.

Similar monitoring programs are being undertaken by the District of Muskoka, Lake of Bays Association, Lake Partner Program, and other lake associations. In recent years, there has been a major collaborative effort throughout the region to establish standardized water quality monitoring protocols and methodologies. This has allowed for better comparability between datasets and enhances the usability of all data collected. The WQI complements and expands upon other monitoring programs conducted in the region by government agencies and other volunteer groups.

2. Program Design and Implementation

2.1 Sampling Areas and Sampling Sites

The WQI study area includes Lakes Muskoka, Rosseau, and Joseph and a number of smaller lakes in the vicinity. The study area is divided into sampling areas representative of lakes, bays, and rivers of interest. Each sampling area consists of one or more sampling sites. Most sampling areas have one reference site established in a central, deepwater location intended to exhibit "average" water quality conditions.

Sampling sites are chosen and classified according to their local environment. The three site types are nearshore, deepwater, and watercourse. Nearshore sites are located adjacent to land where the water depth is between 50 cm and 150 cm as this is the depth at which most recreational use occurs. Deepwater sites are located in deeper, open water locales. Watercourse sites are located in streams

and creeks conveying flow to larger waterbodies. Sampling methodologies differ based on the type of sampling site (Section 3).

Prior to each sampling season, a complete review of the sampling sites is conducted. Sampling sites generally remain consistent from year to year, as the main goal of the program is to determine long-term trends; however, site revisions are made as necessary based on analyses of previous data, volunteer availability, new information, and budget. Generally, bacteria monitoring is discontinued at sampling sites exhibiting chronically low bacteria levels (3+ consecutive years with average concentrations below 10 cfu/100mL). In sampling areas where bacteria monitoring is reduced, new nearshore sites are established as necessary.

2.2 Participants

The WQI relies heavily on the participation of volunteers from across west Muskoka and south Parry Sound. Prior to the sampling season, each sampling area is assigned at least one team leader. Team leaders are primarily responsible for overseeing fellow volunteers who have agreed to help monitor water quality conditions within their respective sampling areas. Detailed descriptions of the roles and responsibilities assumed by team leaders and volunteers are outlined in the yearly Team Leader and Volunteer Field Manuals, respectively.

2.3 Training

Each spring, the MLA hosts volunteer training sessions at local venues in preparation for the upcoming sampling season. These sessions allow new volunteers to understand the program and its requirements, and allow returning volunteers to maintain their sampling skills and enhance their knowledge of the monitoring program and be informed of the changes to be implemented as the program continues ongoing evaluation and improvement. To assure the quality of data collected, team leaders and volunteers are required to attend one of the two spring sessions.

2.4 Equipment

Each sample location has specific sampling requirements and not all locations have the same equipment needs. The following equipment is available and specific items are provided to volunteers for sample collection depending on their needs:

- Sample collection bottle (500 mL; Plastic) Rubber ties, straps, and carabineer (clip)
- Phosphorus sample tubes (20 mL; Glass with plastic lid; Etched fill line), Calcium sample bottle (100 mL; Plastic with plastic lid), Dissolved Organic Carbon sample bottle (125 mL; Plastic with plastic lid)
- Bacteria sample jar (250 mL; Glass with metal lid)
- Secchi disc with weight and rope marked every 0.1 m
- 80 µm filter/funnel with additional mesh
- Pool Thermometer on a rope

- Data Sheets
- Site Schedule and Observation Forms
- Wax pencil and sharpie marker

The following equipment is provided to team leaders for bacteria sample analysis:

- Coliplates (Supplied by Bluewater Biosciences Inc.)
- Egg incubator (GQF Hovabator model)
- UV light (366 nm)

2.5 Sampling Schedule

The WQI consists of four sampling periods occurring between mid-May and late August. Each sampling period begins on a Friday and ends on a Monday, coinciding with peak volunteer availability. The first sampling period must be prior to the onset of lake stratification to allow for measurement of phosphorus and calcium concentrations during the spring turnover. The three remaining sampling periods generally occur in late June, late July, and late August. Bacterial monitoring is limited to the second, third and fourth sampling periods when bacteria levels are typically highest due to warm weather conditions. Additional bacteria sampling is completed if data collected indicates that bacteria have exceeded a predetermined criteria.

3. Sampling and Analytical Methodologies

3.1 Water Quality Parameters

The following parameters are used as indicators of water quality for the WQI:

- Secchi Depth
- Total Phosphorus (Spring Turnover and Yearly Mean)
- Dissolved Organic Carbon (DOC)
- Calcium
- Bacteria (Total Coliform and E. coli)
- Temperature

Water quality measurements (water temperature and Secchi depths) and observations are recorded on the WQI Data Sheet on each sampling date (see **Appendix 1**). Participants also collect site-specific information and photographs during the course of the sampling season. Site information is recorded on the appropriate Site Schedule and Observation Form (see **Appendix 2**). Protocols for Secchi Depth and Total Phosphorus are partially adapted from the Ministry of Environment's "Lake Partner Program: 2012 Spring Sampling Instructions" (MOE 2012).

3.1.1 Secchi Depth

Water clarity is typically affected by three different factors: algae, suspended sediment, and water colour. Water clarity can be measured by the depth to which one may see into the water. Secchi depth provides a measurement of water clarity and represents the distance that light will travel into the water column. The Secchi disc consists of a weighted circular plate, 20 cm in diameter, with the surface painted in opposite black and white quarters and measures depth in metres. Each MLA disc is attached to 10 m of rope labeled at 10 cm intervals.

To measure Secchi depth, go to the shady side of the boat and if wearing sunglasses, remove them. The Secchi disc is lowered straight down into the water until it is no longer visible. The “down” depth is recorded on the data sheet at this point. The disc is then lowered further into the water and then raised until it reappears. The “up” depth is recorded at this point. Secchi depth is calculated as the arithmetic mean of the “up” and “down” measurements.

Secchi disc measurement.

3.1.2 Total Phosphorus

Total phosphorus (TP) samples are collected from select sites, as indicated on the sampling schedule. Lake water is collected using a plastic collection bottle and transferred into digest tubes through an 80 µm filter/funnel. Collection bottles and filter/funnels are supplied by the Ministry of Environment’s Lake Partner Program. At deepwater sites, samples are collected at Secchi depth by lowering a collection bottle attached to a Secchi disc straight down into the water column (see Section 3.1.1). At nearshore and watercourse sites, samples are collected near the surface using the ‘plunge and sweep’

method. TP samples are collected in duplicate to increase the accuracy of final measurements. Samples are kept cool throughout the entire chain of custody. Following each sampling event, TP samples are submitted to the Trent University Laboratory at the Ministry of Environment's Dorset Environmental Science Centre (DESC) for analysis.

The following protocol is used to collect duplicate TP samples from **deepwater** sites:

1. Use the 'plunge and sweep' method to rinse the deepwater sample collection bottle **TWICE** with surface water (fill and empty). The 'plunge and sweep' method is undertaken by turning the deepwater sample collection bottle upside-down, plunging it into the lake to approximately forearm depth, turning the bottle 90˚ and sweeping upwards towards the surface, filling the sample collection bottle before it reaches the surface.
2. Rinse the filter/funnel **TWICE** with surface water (fill and allow water to pass through).
3. Clip the sample collection bottle to the Secchi disc and attach using the rubber ties.
4. Lower the sample collection bottle down to the Secchi depth (average of "up" and "down" depths) with the goal of obtaining water and filling the bottle the entire way down. **In shallow lakes, lower the bottle no closer than 1 metre from the lake bottom*.*
5. Bring the sample to the surface.

6. Rinse the two phosphorus sample tubes (20 mL; glass with plastic lid) **TWICE** by filling each **HALF** full with filtered lake water by pouring water from the deepwater collection bottle through the filter/funnel into each phosphorus sample tube. Cover each tube with its cap, shake it **FIVE** times, remove the cap, empty the sample tube, and repeat rinsing.
7. Now fill each of the two phosphorus sample tubes with filtered water from the same collection bottle and place the cap on each tube. These are the samples that will be sent to the laboratory. **Fill to 1 cm above the etched line**.
8. Make sure the lids are screwed on snugly.

9. Write the sample date on the label affixed to each phosphorus tube using a permanent/waterproof marker (not a pen or pencil).
10. Keep the samples cool and shaded from the sun until they can be delivered to the MLA office.

The following protocol is used to collect duplicate TP samples from **nearshore** and **watercourse** sites:

1. Use the 'plunge and sweep' method to rinse the deepwater sample collection bottle **TWICE** with surface water (fill and empty). The 'plunge and sweep' method is undertaken by turning the deepwater sample collection bottle upside-down, plunging it into the lake to approximately forearm depth, turning the bottle 90˚ and sweeping upwards towards the surface, filling the sample collection bottle before it reaches the surface.
2. Rinse the filter/funnel **TWICE** with surface water (fill and allow water to pass through).
3. Rinse the two phosphorus sample tubes (20 mL; glass with plastic lid) **TWICE** by filling each **HALF** full with filtered lake water by pouring water from the collection bottle through the filter/funnel into each phosphorus sample tube. Cover each tube with its cap, shake it **FIVE** times, remove the cap, empty the sample tube, and repeat rinsing.
4. Now fill each of the two phosphorus sample tubes with filtered water from the same collection bottle and place the cap on each tube. These are the samples that will be sent to the laboratory. **Fill to 1 cm above the etched line**.
5. Make sure the lid is screwed on snugly.
6. Write the sample date on the label affixed to each phosphorus tube using a permanent/waterproof marker (not a pen or pencil).
7. Keep the samples cool and shaded from the sun until they can be delivered to the MLA office.

3.1.2.1 Total Phosphorus Data Analysis

Spring Turnover phosphorus concentrations are calculated as the arithmetic mean of the spring or mid-May duplicate sample measurements. Yearly Mean phosphorus concentrations are calculated as the arithmetic mean of duplicate sample measurement means from a single sampling site within a sampling season.

3.1.3 Dissolved Organic Carbon

Dissolved Organic Carbon (DOC) samples are collected from select **nearshore** and **watercourse** sites, as indicated on the sampling schedule. Samples are collected near the surface using the 'plunge and sweep' method.

Volunteers use the following protocol to collect DOC samples:

1. Use the 'plunge and sweep' method to rinse the deepwater sample collection bottle **TWICE** with surface water (fill and empty). The 'plunge and sweep' method is undertaken by turning the deepwater sample collection bottle upside-down, plunging it into the lake to approximately forearm depth, turning the bottle 90˚ and sweeping upwards towards the surface, filling the sample collection bottle before it reaches the surface.
2. Rinse the filter/funnel **TWICE** with surface water (fill and allow water to pass through).

3. Rinse the DOC sample bottle (125 mL; plastic with plastic lid) **TWICE** by filling **HALF** full with filtered lake water by pouring water from the collection bottle through the filter/funnel into the sample bottle. Cover the sample bottle with the lid, shake it **FIVE** times, remove the lid, empty the sample bottle, and repeat rinsing.
4. Now fill the DOC sample bottle with filtered water from the same collection bottle and place the lid on the bottle.
5. Make sure the lid is screwed on snugly.
6. Write the sample date on the DOC sample bottle using a permanent/waterproof marker (not a pen or pencil).
7. Keep the sample cool and shaded from the sun until it can be delivered to the MLA office.

3.1.4 Calcium

Calcium samples are collected from **deepwater** reference sites during the spring turnover sampling period, as indicated on the sampling schedule. Lake water is collected at Secchi depth by lowering a collection bottle attached to a Secchi disc straight down into the water column (see Section 3.1.1). Samples are transferred directly into 100 mL wide-mouth plastic jars supplied by the Trent University Laboratory at the DESC. Samples are kept cool throughout the entire chain of custody. Calcium samples are submitted to the Trent University Laboratory for analysis.

Volunteers use the following protocol to collect calcium samples:

1. Use the 'plunge and sweep' method to rinse the deepwater sample collection bottle **TWICE** with surface water (Fill and empty). The 'plunge and sweep' method is undertaken by turning the deepwater sample collection bottle upside-down, plunging it into the lake to approximately forearm depth, turning the bottle 90˚ and sweeping upwards towards the surface, filling the sample collection bottle before it reaches the surface.
2. Clip the sample collection bottle to the Secchi disc.
3. Lower the sample collection bottle straight down to the Secchi depth (average of “up” and “down” depths), with the goal of obtaining water and filling the bottle the entire way down. *In shallow lakes, lower the bottle no closer than 1 metre from the lake bottom.*
4. Bring the sample to the surface.
5. Pour water from the sample collection bottle directly into the calcium sample bottle (100 mL; Plastic with plastic lid).
6. Fill the calcium sample bottle (ensure that it is full).
7. Make sure the lid is screwed on snugly.
8. Write the site code and the sample date on the calcium sample bottle using a permanent/waterproof marker (not a pen or pencil).
9. Keep the sample cool and shaded from the sun until it can be delivered to the MLA office.

3.1.5 Bacteria

3.1.5.1 Bacteria Sample Collection

Volunteers collect bacteria samples from nearshore and watercourse sites using 250 mL glass jars. The jars are purchased from the Consolidated Bottle Company. Jars and lids are sterilized annually prior to distribution to volunteers. The following protocol is used to collect bacteria samples from **nearshore** and **watercourse** sites:

1. Use the 'plunge and sweep' method to rinse the bacteria sample collection bottle (250 mL; Glass with metal lid) **TWICE** with surface water (Fill, put the top on, shake vigorously and empty). The 'plunge and sweep' method is undertaken by turning the sample collection bottle upside-down, plunging it into the lake to approximately forearm depth, turning the bottle 90˚ and sweeping upwards towards the surface, filling the sample collection bottle before it reaches the surface.
2. Use the 'plunge and sweep' method again to collect your sample. Secure the lid to the collection bottle.
3. Make sure the lid is screwed on snugly.
4. Write the site code and the sample date directly on the bacteria sample lid using a wax pencil (not a pen or marker).
5. Keep the sample cool and shaded from the sun until it can be delivered to the team leader for incubation and analysis.

Bacteria samples are kept on ice and in the dark to preserve the bacteria at their naturally occurring levels. Bacteria analysis is completed as soon as possible following sample collection (maximum within 24 hours of collection).

3.1.5.2 Bacteria Sample Analysis

Sample water from the bacteria collection jar is poured into a commercially available water analysis kit (Coliplate). Coliplates are left to incubate in egg incubators (GQF Hovabator) at 37˚C for 24-26 hours prior to analysis. Team leaders record the time of preparation and time of analysis on the data sheet (**Appendix 1**). Coliplates have 96 wells containing an agar (bacterial growth medium) that reacts in the presence of coliform bacteria, turning wells blue/green. Wells identified as any shade of blue or green are counted as positive as per the manufacturer's instructions, and recorded on the data sheet. After analyzing for total coliform, each Coliplate is used to analyze for Escherichia coli (E. coli). This is done by exposing the plate to an ultraviolet light (UV) under dark conditions. Wells that are both blue/green and fluorescent under the UV light are identified as positive for E. coli and the number of blue/green-fluorescent wells is recorded on the data sheet. Actual total coliform and E. coli colony forming unit (cfu) counts are determined later by the WQI Field Co-ordinator during the final data entry via MPN table. After use, Coliplates are emptied into a septic system, rinsed, and returned to the MLA Office. At the end of the sampling season, the plates are returned to the supplier to be cleaned, retreated, and reused.

Coliplate with 12 blue wells.

The MLA WQI includes a protocol that requires volunteers to re-sample a site if E. coli levels are found to be equal to or greater than 50 cfu/100 mL (18 cells on the Coliplate). This cautious approach allows the MLA to increase its monitoring effort at sites that demonstrate the potential for chronically elevated bacteria levels.

When E. coli levels are found to be equal to or greater than 50 cfu/100 mL (18 cells on the Coliplate), at a particular site, the volunteer must contact the field coordinator and will most likely return to that site and collect **2 SEPARATE** bacteria samples as soon as possible. The same protocol should be followed for the analysis of the 2 additional samples.

3.1.5.3 Total Coliform Data Analysis

Total coliform yearly averages are calculated as the geometric mean of all measurements for an individual sampling site. Concentrations are reported as the number of cfu observed in 100 mL of lake water (cfu/100 mL). Note that Coliplates have a detection limit of three cfu/100mL (a count of zero blue wells corresponds to a count of “less than three” cfu per 100 mL of lake water). This limit is managed by assigning all readings of “less than three” cfu/100mL with an absolute value of 1 cfu/100mL.

3.1.5.4 Escherichia coli (E. coli) Data Analysis

E. coli yearly averages are calculated as the geometric mean of all measurements for an individual sampling site. Concentrations are reported as the number of cfu observed in 100 mL of lake water (cfu/100 mL). As for with the total coliform measurements, all readings of “less than three” E. coli cfu/100mL were assigned an absolute value of 1 cfu/100mL.

3.1.6 Temperature

Water temperature is recorded at each site. Volunteers lower a pool thermometer on a rope into the water when first arriving at each site. After all of the other protocols are completed, the sampler retrieves the thermometer and records the temperature value in degrees Celsius. Volunteers also measure and record the ambient air temperature once per sampling event.

3.2 Other Parameters

3.2.1 Rainfall

Volunteers assess the amount of rainfall over the 24-hour period prior to sampling and record it as “Heavy”, “Moderate”, “Light”, or “None”. Rainfall data is occasionally used to assess the significance of high bacteria counts.

3.2.2 Waves

Volunteers assess the amount wave action at each sampling site. Surface water is classified and recorded as either “Calm” or “Rough”. Wave data is occasionally used to assess the significance of unusual Secchi depth measurements.

3.3 Quality Assurance and Quality Control

The WQI Quality Assurance (QA) and Quality Control (QC) protocols were developed over the course of several years. Reliability of monitoring methods and data is paramount to the effective use of scientific methods. Collecting environmental data in the field is subject to countless uncontrollable variables, which makes repeatability difficult. For this reason, quality control and quality assurance protocols that aim to identify misinformation and procedural error are of utmost importance to the WQI. Rigorous training, detailed documentation, and duplicate measures are used throughout the sampling season.

3.3.1 Total Phosphorus Quality Control

Quality control measures are in place for phosphorus sampling protocols. Duplicate samples are collected throughout the sampling season at every phosphorus sampling site using identical protocols. Duplicate measurements indicate the range of results that can be expected due to variability in sampling and laboratory testing.

Each year, an analysis of “outliers” (data points that seem to be skewed from the normal data set) and “bad duplicate splits” (duplicate measurements that are not consistent) is completed. These outlier data points are typically the result of sample contamination. Outliers and bad duplicate splits are identified using the statistical protocol adopted by the District Municipality of Muskoka for the Lake System Health

Program (GLL 2008). Measurements removed from the dataset are noted in the annual Water Quality Report.

3.3.2 Bacteria Quality Control

Limited quality control measures are conducted on bacteria samples throughout the sampling season. Team leaders are asked to include a single "blank" Coliplate each time they analyzed a batch of bacteria samples. Blank Coliplates are filled with Aquafina purified bottled water, incubated, and analyzed along with the lake water samples. This control step is intended to determine whether the sample preparation, incubation, and analysis process is producing false positive results. Note that team leaders responsible for multiple sampling areas are only required to include one blank plate per batch of bacteria sample analyzed, as opposed to one blank per sampling area.

4. Water Quality Report

At the end of every sampling season, data collected through the WQI is compiled and summarized in an annual Water Quality Report. Current data is compared to data from previous years to establish long-term trends. The Water Quality Report is disseminated to key stakeholders and made available to the public on the MLA website (www.mla.on.ca).

5. Definitions

Arithmetic mean: This type of average is calculated as the sum of all the numbers in the series divided by the count of all numbers in the series. The arithmetic mean is relevant any time several quantities add together to produce a total.

E. coli: Escherichia coli, it is a subset of total coliforms, and is commonly found in the lower intestine of warm-blooded organisms, making it a good indicator organism of fecal contamination. There are many different strains of E. coli; most waterborne strains are themselves not harmful, but some (such as E. coli O157:H7) can cause serious illness.

Geometric mean: This type of average is calculated by multiplying together a group of n numbers and then taking the nth root of the resulting product. The geometric mean is relevant any time several quantities multiply together to produce a product. The geometric mean is used to indicate the central tendency or typical value of a set of numbers and is typically used to calculate average bacteria counts because as a living organism, bacteria counts are highly sporadic and inconsistent.

Sampling Area: A geographic location encompassing a group of WQI monitoring sites.

Sampling Site: The discrete and unique location where samples are collected and measurements are taken.

Secchi Depth: A measure of water clarity, measured using a Secchi disc - a small disc attached to a rope. Alternating quarters of the top side of the disc are coloured white and black. The Secchi depth is the depth of water whereby the sampler can no longer distinguish the white and black quarters of the disc.

Spring Turnover Total Phosphorus: A single phosphorus concentration measurement taken in a typically unstratified lake during the spring turnover period. This measurement has been shown to adequately represent the overall phosphorus concentration in a lake (Clark, 1992). Typically the spring turnover lasts for a few days when the temperature of the entire water column is consistent (usually 4˚C) allowing the water column to mix. In practice, measurements taken anytime in May are considered to be adequate by Ontario's Ministry of the Environment.

Total Coliform: Total coliform bacteria are a collection of relatively harmless microorganisms that live in large numbers in soils, plants and in intestines of warm-blooded (humans) and cold-blooded animals.

Total Phosphorus: Phosphorus is a chemical element that is essential for all living cells. Amongst other sources, it is found in fertilizers, soaps, and human waste.

Water Clarity: Water clarity is a measure of how much light penetrates through the water column. The clarity of water is influenced both by suspend particulate matter (sediment and plankton) and by coloured organic matter (tea coloured lakes). Clarity can provide some indication of a lake's overall water quality, especially the amount of algae present.

Yearly Mean Total Phosphorus: The arithmetic mean of phosphorus concentration measurements taken throughout the ice-free period. Note: yearly mean phosphorus concentration as reported by the WQI is for spring and summer months only.

Note: several of these definitions have been taken from the WQI Summary Report – Citizens Environment Watch, 2009.

6. References

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Appendix 1

MLA WQI Data Sheet

MLA™
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Sample Area: _____

Date		**Sample Time**	
Trained Sampler		**Other Volunteers**	
Rainfall in last 24 hours (heavy, moderate, light, none)		**Air Temp.**	

Site Specific Information

Please note that all measurements taken should be in metric

Site Code	Water Temp.	Waves	Secchi Down	Secchi Up	Blue	Blue & Flor.
		☐ Calm ☐ Rough				
		☐ Calm ☐ Rough				
		☐ Calm ☐ Rough				
		☐ Calm ☐ Rough				
		☐ Calm ☐ Rough				
		☐ Calm ☐ Rough				
		☐ Calm ☐ Rough				
		☐ Calm ☐ Rough				
		☐ Calm ☐ Rough				

For Team Leader Use:

Preparation Date/Time:
Analysis Date/Time:
Analysis done by:

Comments:

Appendix 2

MLA WQI Site Schedule and Observation Form

Area Name

LAK-X

May 17-20:	Observations:
June 28- July 1:	Observations:
July 26-29:	Observations:
August 23-26:	Observations:

Please note any land uses or date specific information on this sheet

and return it at the end of the season with your binder

Appendix 3

MPN Table for Coliplate Result Analysis

Most Probable Number (MPN) of colony forming-units per 100 mL.

No. Wells Giving Positive Reaction | MPN per 100 mL Sample

0 <3					
1 3	2 5	3 8	4 11	5 13	6 16
7 19	8 22	9 25	10 28	11 30	12 33
13 36	14 39	15 43	16 46	17 49	18 52
19 55	20 59	21 62	22 65	23 69	24 72
25 76	26 79	27 83	28 87	29 90	30 94
31 98	32 102	33 106	34 110	35 114	36 119
37 123	38 127	39 132	40 136	41 141	42 146
43 151	44 156	45 161	46 166	47 171	48 177
49 182	50 188	51 194	52 200	53 206	54 213
55 219	56 226	57 233	58 240	59 247	60 255
61 263	62 271	63 280	64 289	65 298	66 307
67 317	68 328	69 339	70 350	71 362	72 375
73 388	74 403	75 418	76 434	77 451	78 469
79 489	80 510	81 534	82 559	83 587	84 619
85 654	86 694	87 740	88 794	89 858	90 938
91 1,038	92 1,174	93 1,370	94 1,696	95 2,424	96 >2,424

Most Probable Number (MPN) of colony forming-units (cfu's) per 100 mL – MPN Table is used for both Total Coliforms (blue/green positive wells) and E.coli (fluorescence positive wells)