Muskoka Lakes Association
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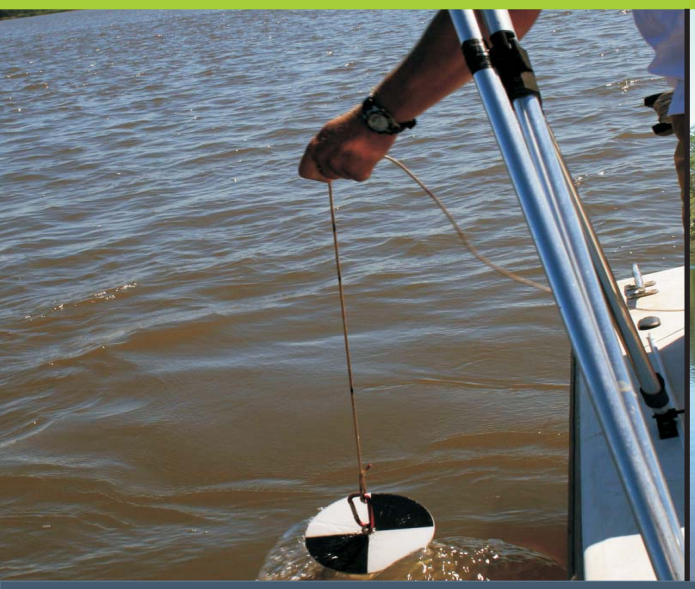
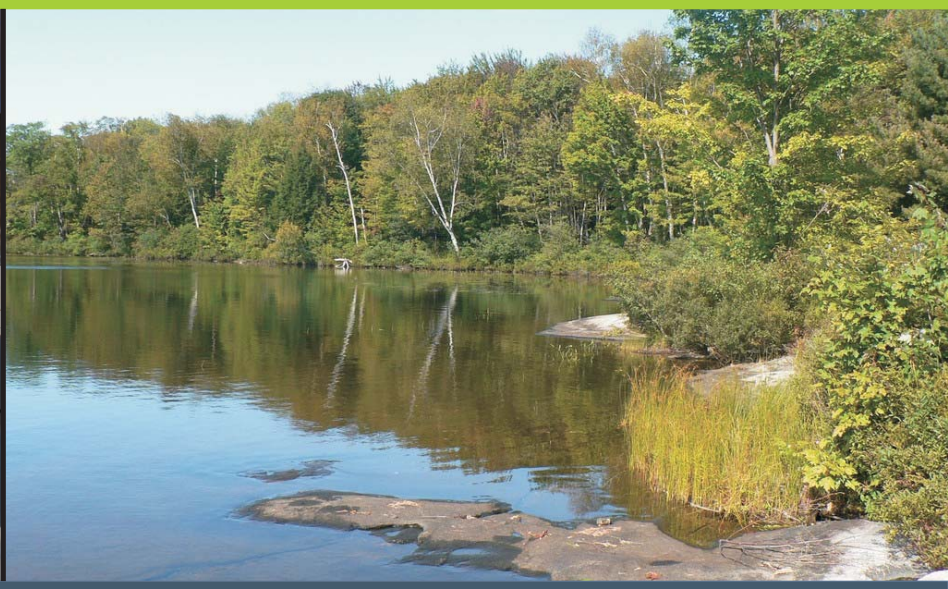
WATER QUALITY INITIATIVE MONITORING PROGRAM

Program Delivery and Methodology

2010





RiverStone
Environmental Solutions Inc.

February 2, 2011
RS#2009-06

Ms Eleanor Lewis
Director MLA / Chair Water Quality Portfolio
Muskoka Lakes Association
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Box 298
Port Carling, ON
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SUBJECT: Muskoka Lakes Association Water Quality Initiative - 2010 Program Delivery & Methodology Document

Dear Eleanor:

RiverStone Environmental Solutions Inc. is pleased to provide you with the attached 2010 Program Delivery & Methodology document.

Please contact us if there are any questions regarding the report, or if further information is required.

Best regards,

RiverStone Environmental Solutions Inc.

Bev Wicks, Ph.D.
Senior Aquatic Ecologist
Report Author

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1. BACKGROUND

The Muskoka Lakes Association (MLA) is a non-profit organization made up of over 2,600 member families representing approximately 12,000 individuals. The MLA has represented seasonal and permanent residents of Muskoka for over 100 years. The MLA's Mission Statement "to promote the responsible use, enjoyment and conservation of the unique Muskoka environment" and its Vision "that, through the achievement of our mission, present and future generations will benefit from our efforts" reflects the goals of this long-time organization.

Some of the directives of the MLA involve "water quality initiatives including testing" and "involvement in bigger picture environmental issues of land use that are precedent setting and have implications on other land uses in our area". The Water Quality Portfolio works with these directives to continue to refine and run the Water Quality Initiative (WQI) that is a science-based monitoring program. The WQI has just completed its tenth year of operation.

The WQI began as a formal, scientifically based ecological-monitoring and water-quality research program in 2001, with a pilot phase led by Dr. Neil Hutchinson of Gartner Lee Ltd. The development of the program was based on a review of the monitoring programs carried out by the Ministry of the Environment (MOE) and the District Municipality of Muskoka (DMM) at that time and on the premise that the nearshore of lakes is, biologically important, extensively used by residents, and highly visible. The program was designed to "potentially reveal relationships between land use and nearshore water quality and could potentially generate results which could guide future management and stewardship activities." Detailed descriptions of the original study design can be found in the 2001 and 2002 MLA reports prepared by Gartner Lee Limited.

Between 2003 and 2008, the research/monitoring program continued to grow with an increase in the number of lakes and sites sampled. The algae sampling portion of the program was discontinued due the inconclusive nature of the data, and over the years, sampling methods were modified to fit the needs of the program. In 2007, additional monitoring efforts were directed towards specific lakes and bays classified as "over-threshold" by the DMM. The research objective of this effort was to "determine the sources of phosphorus loading and other contaminants in these areas."

The other aspect of the MLA program was to promote responsible use of the resources in Muskoka. Increased volunteer involvement and community stewardship activities reveal the popularity and the

success of the program. Outside of the monitoring and research programs, volunteers are pioneering local Stewardship Initiatives to improve the lakes and watersheds in their immediate communities.

In the spring of 2009, the MLA retained RiverStone Environmental Solutions Inc. (hereafter RiverStone) to assist with its WQI and Stewardship Initiative objectives. While RiverStone was not directly involved in the objectives and protocols for 2009, we had ongoing communications with the Water Quality Portfolio and volunteers, attended a training session, and reviewed the reports and manuals to familiarize ourselves with the program. RiverStone produced both Summary and Technical reports for the 2009 sampling season.

In 2010, RiverStone continued to provide scientific advice, coordinate sample collection and testing, analyze data and report on findings. The results and recommendations from the 2010 WQI monitoring program are presented in the Water Quality Report that includes the following components: area description, volunteer recognition, long-term spring turnover and yearly mean phosphorus concentrations, long-term *E. coli* yearly means, 2010 data summary, trends, comments, and recommendations.

This document provides a separate, comprehensive overview of program delivery components, and the sampling and analytical methodology for 2010. The data and results of the analysis are presented separately in the 2010 Water Quality Report as stated previously.

1.1. Purpose of the Water Quality Initiative

> "The primary purpose of the MLA WQI monitoring program is to discover the *source* of problems, identified both by DMM modelling and community members. This is accomplished through monitoring over a longer season (Victoria Day to Labour Day) in deep water as well as the near shore area of a number of lakes and bays. Results of monitoring in the nearshore zone are compared to comparable deepwater monitoring results to indicate land-based problem sources.
>
> The secondary purpose of WQI monitoring is to identify problems in areas where the DMM program cannot monitor due to limited resources or political jurisdiction. WQI monitoring can also provide additional evidence supporting regulation of vulnerable areas within Muskoka that should be protected. Monitoring is therefore concentrated in three types of areas:
>
> - lakes and bays with problems identified by DMM;
> - lakes and bays where past WQI data indicates a problem; and
> - lakes and bays where DMM does not monitor."

Cited in 2009 WQI Monitoring Program Technical Report (Citizens' Environment Watch, 2009)

2. PROGRAM DELIVERY

2.1. Program Implementers

The WQI would not be possible without a dedicated team of volunteers and staff. During the review of this program, RiverStone was impressed by the number of hours and dedicated efforts that volunteers and staff have contributed. The following provides a brief description of those involved.

2.1.1. Water Quality Portfolio

The WQP is one of four portfolios of the MLA. The following is a list of the dedicated volunteers that make up the Portfolio: Eleanor Lewis (Chair), Mike Bidwell, Lola Bratty, Louise Cragg, Debbie Hastings, Nick Kristoffy, Mike Logan, Mike Muffels, Lawton Osler, Gord Ross, Peter Seybold, and Andrew Watson.

2.1.2. Staff/Scientific Advisor

Jeremy Prahl, ecologist at RiverStone, acted as the WQI Field Coordinator from April to September 2010. Andrew Watson is recognized for his role in passing on valuable program information to Jeremy for the 2010 field season. Cheryl Hollows and Lisa Noonan provided administrative support to the WQI through the MLA office in Port Carling.

RiverStone Environmental Solutions Inc. has acted as the lead scientific advisor for the WQI since 2009.

2.1.3. Volunteers

The 2010 WQI program relied heavily on the participation of over 110 volunteers from across the District of Muskoka and Seguin Township. Area-specific Team Leaders were assigned based on interest and willingness to oversee volunteer sampling activities. The roles and responsibilities assumed by Team Leaders and volunteers can be found in the 2010 Team Leader and Volunteer Manuals, respectively. Team Leaders and volunteers are recognized in the 2010 Water Quality Report.

2.2. Volunteer Training

In 2010, the sampling season began with two volunteer training sessions held in Muskoka. The training sessions took place at Rosseau Lake College on May 15th and at the Port Carling Community Centre on May 22nd. These sessions allow volunteers to update their training and knowledge on an annual

basis. In an attempt to improve the quality of data collected, Team Leaders were specifically required to attend one of the two sessions. In total**,** 64 volunteers attended a training session in 2010.

2.3. Partnerships

Eleven affiliates participated in the MLA WQI in 2010:

- Brandy Lake Association
- Bruce Lake Family Association
- Bass Lake Association
- Clear Lake Cottagers' Association (Township of Muskoka Lakes)
- Leonard Lake Stakeholders Association
- Moon River Property Owners' Association
- Muldrew Lakes Cottagers Association
- Silver Lake Property Owners Association (Township of Muskoka Lakes)
- Star Lake Woods Association
- Sucker Lake Cottage Association
- Sunny Lake Cottage Association

It is important that the MLA maintain the ongoing participation of existing partners to ensure a creditable long-term water quality data set, gauge the interest of other local associations, and facilitate early involvement in the WQI for 2011.

3. SCIENTIFIC METHODS

3.1. Sampling Schedule

Sampling for the 2010 WQI program occurred on a biweekly schedule between May 21–24 and August 27–30 (**Table 1**). Eight sampling dates were established during this period. The dates were selected based on volunteer availability and to ensure that early-spring samples could be taken. The first sampling window was established prior to the typical onset of lake stratification in order to provide a measurement of phosphorus during spring turnover.

Table 1. WQI Sampling Windows for 2010

Sample	Sampling Window
1	May 21 – 24
2	June 4 – 7
3	June 18 – 16

Sample	Sampling Window
4	July 2 – 5
5	July 16 – 19
6	July 30 – August 2
7	August 13 – 16
8	August 27 – 30

3.2. Sites

The establishment of sites sampled for the 2010 data set occurred over a number of years starting in 2001. In 2002, seventy sites in Lakes Joseph, Rosseau, and Muskoka, and Brandy Lake were established under the direction of Dr. Neil Hutchison. Sampling sites were selected in deep water locations to represent the average offshore conditions and nearshore sites were selected based on their proximity to certain land uses. In 2002, seventeen sampling areas were identified. Each area had a number of sites clustered around a targeted land use, as well as an offshore/deepwater site intended to represent an "average" condition. Between 2002 and 2009, the monitoring program expanded to include many additional sites that were selected based their proximity to specific land uses (residential, golf courses, urban areas etc.). A summary of the adjacent land use is provided by sampling site in (**Table 2**). In 2010, the sites for each sampling area were again predetermined, with the number of sites sampled limited by volunteer participation. Volunteers were provided with a manual that included satellite imagery-based site maps, and digital photographs of each site, when available. There were two types of sites: nearshore and offshore. Nearshore sites were located where the water depth was between 50 cm and 150 cm as this is the depth in which most recreational use occurs and is a good water/land interface. Offshore (reference) sites were located in deep water near the centre of the sampling area to ensure that the site was outside of the potential influence of nearshore land use factors (e.g. lake or bay).

3.3. Monitoring Parameters

The following methodologies were documented in the 2008 WQI Monitoring Program Technical report (CEW, 2009). Modifications were made in the production of this document as required to update protocols and for clarity.

The following parameters were used as indicators of water quality for the 2010 WQI program:

- Total Phosphorus: Spring turnover and yearly mean (summer sampling)
- Bacteria: Total coliform and *E. coli*
- Secchi Depth
- Temperature

The parameters measured on each sampling date were also predetermined based on the rationale for the site location. Volunteers followed the Field Manual in measuring these parameters. In addition, supplementary information was recorded on the datasheet (see **Appendix A**). **Table 2** provides a list of sampling sites and the parameters measured.

Table 2. List of all sites sampled in 2010 and the parameters monitored at each site. Numbers indicate the number of samples collected and * indicates that a spring turnover sample was *not* collected.

Lake	Sampling Area	Site	Land Use	Phosphorus	Bacteria	Secchi	Temperature
Lake Joseph	Cox Bay	COX-0	Offshore	8		8	8
		COX-1	Golf Course (Lake Joe)	8	8		8
	Cox Bay	COX-2	Golf Course (Lake Joseph)	8	8		8
		COX-3	Town (Port Sandfield)	8	8		8
		COX-4	Resort (Pinelands)	8	8		8
	East Bay	EAS-0	Offshore	8	8	8	8
		EAS-1	Undeveloped	8	8		8
		EAS-2	Undeveloped	8	8		8
		EAS-3	Undeveloped	8	8		8
	Foot's Bay	FTB-0	Offshore	8		8	8
		FTB-3	Marina	8			8
	Gordon Bay	GNB-0	Offshore	1		1	1
	Hamer Bay	HMB-0	Offshore	7	8	8	8
		HMB-1	Marina/Creek/Golf Course (Rocky Crest)	8	7		8
		HMB-2	Resort (Rocky Crest)	8	8		8
		HMB-3	Resort (Rocky Crest)	8	8		8
		HMB-4	Residential	7	8		8
		HMB-5	Creek	6*			5
	Lake Joseph	JOS-1	Offshore	8		6	8
	Little Lake Joseph	LLJ-0	Offshore	8		8	4
		LLJ-9	Offshore		8		8
		LLJ-10	Offshore		8		8
		LLJ-11	Offshore		8		8
	Stills Bay	STI-0	Offshore	8		8	8
		STI-2	Creek	8			8
	Stanley Bay	STN-0	Offshore	8		5	8
		STN-1	Residential	8			8

Lake	Sampling Area	Site	Land Use	Phosphorus	Bacteria	Secchi	Temperature
Lake Joseph	Stanley Bay	STN-2	Residential	8			8
		STN-3	Residential	8			8
Lake Muskoka	Bala Bay	BAL-0	Offshore	1	8	6	8
		BAL-1	Residential		8		8
		BAL-2	Marina/Town (Bala)		8		8
		BAL-3	Residential		8		8
		BAL-4	Residential		8		8
		BAL-5	Unknown		8		8
	Beaumaris	BMR-0	Offshore	8	8	8	8
		BMR-2	Golf Course (BYC)	8	8		8
		BMR-3	Town (Beaumaris)		8		8
		BMR-5	Golf Course (BYC)	8	8		8
		BMR-6	Wetland/Golf Course (BYC)	8	8		8
		BMR-7	Residential	8	8		8
		BMR-8	Park	8	8		8
	Boyd Bay	BOY-0	Offshore	8	8	8	8
		BOY-1	Residential	8	8		8
		BOY-2	Residential	8	8		8
		BOY-3	Marina	8	8		8
	Eilean Gowan Island	ELG-0	Offshore	1	8	8	8
		ELG-1	Residential		8		8
		ELG-2	Residential		8		8
		ELG-3	Residential		8		8
	Muskoka Bay	MBA-0	Offshore	8		8	8
	Muskoka Bay	MBA-3	Residential	8	8		8
		MBA-4	Town (Gravenhurst)	8	8		8
		MBA-5	Town (Gravenhurst)		8		8
		MBA-9	Residential	8	8		8
		MBA-11	Marina	8	8		8
		MBA-12	Creek (Urban-Gravenhurst)	8	9		8
	Muskoka Sands	MSN-0	Offshore	1	7	7	7
		MSN-1	Resort (Muskoka Sands)		7		7
		MSN-2	River Outlet/ Golf Course (Taboo)		7		7
		MSN-3	Residential		7		7
		MSN-4	River (Hoc Roc)	6	7		7
	Dudley Bay	MUS-2	Offshore	1		1	1
	North Bay	NRT-0	Offshore	1		1	1
	Willow Beach	WLB-0	Offshore	8	8	8	8
		WLB-1	Resort (Touchstone)	8	8		8
		WLB-2	Resort (Touchstone)	8	8		8
		WLB-3	Creek/Golf Course (Kirie Glen)	7*	8		8
	Whiteside Bay	WTS-0	Offshore	1		1	1
Lake Rosseau	Arthurlie Bay	ART-0	Offshore	1		1	1

Lake	Sampling Area	Site	Land Use	Phosphorus	Bacteria	Secchi	Temperature
Lake Rosseau	Brackenrig Bay	BRA-0	Offshore	5*	1	6	6
		BRA-1	Residential	5*	6		6
		BRA-2	Residential	5*	6		6
		BRA-3	Residential	5*	6		6
	Morgan Bay	MGN-0	Offshore	8	8	8	8
		MGN-1	Residential	8	8		8
		MGN-2	Residential	8	8		8
		MGN-3	Wetland	8	8		8
		MGN-4	Creek	8	8		8
	Minett	MIN-0	Offshore	8	8	8	8
		MIN-1	Resort (Cleveland's House)		8		8
		MIN-4	Resort (Red Leaves)/ Golf Course (The Rock)		8		8
		MIN-5	Residential/Golf Course (The Rock)		8		8
	Muskoka Lakes G&CC	MLG-0	Offshore	1		1	1
	East Portage Bay	POR-0	Offshore	8		7	8
		POR-1	Agricultural	8			8
		POR-2	Residential	8			8
		POR-3	Agricultural	8			8
		POR-4	Residential	8			8
		POR-5	Residential	8			8
	Royal Muskoka Island	RMI-0	Offshore	1		1	1
	Lake Rosseau	ROS-1	Offshore	1		1	1
	Rosseau North	RSH-0	Offshore	8	8	6	8
		RSH-2	River (Shadow)	8	8		8
		RSH-3	Town (Rosseau)	7*	8		8
		RSH-4	Town (Rosseau)	8	8		8
	Skeleton Bay	SKB-0	Offshore	8	8	6	7
		SKB-1	Roadside/Residential	8	8		7
		SKB-3	Residential	8	8		7
		SKB-4	River (Bent)	8	8		7
	Tobin's Island	TOB-0	Offshore	1		1	1
	Windermere	WIN-0	Offshore	8	7	8	8
		WIN-1	River (Dee)	8	7		8
		WIN-2	Unknown	7*	7		7
		WIN-3	Creek (Culvert)/Golf Course (Windermere)	8	7		8
		WIN-4	Resort (Windermere)/ Creek (Culvert)	8	7		8
		WIN-5	Wetland	8	7		8
Bass Lake	Bass Lake	BAS-0	Offshore	8		8	7
		BAS-1	Offshore	8		7	7
		BAS-2	Unknown		8		8
		BAS-3	Unknown		8		8

Lake	Sampling Area	Site	Land Use	Phosphorus	Bacteria	Secchi	Temperature
Brandy Lake	Brandy Lake	BDY-0	Offshore	1		8	
		BDY-1	Creek or Wetland		8		8
		BDY-2	Residential		8		8
		BDY-3	Residential		8		8
		BDY-5	Residential		8		8
		BDY-6	Wetland/Creek		8		8
		BDY-7	Unknown		8		8
		BDY-8	Unknown		8		8
		BDY-9	Unknown		8		8
		BDY-10	Unknown		8		8
		BDY-11	Unknown		8		8
Bruce Lake	Bruce Lake	BRU-0	Offshore	8	8	8	8
		BRU-1	Unknown	8	8		8
		BRU-3	Unknown	8	8		8
		BRU-4	Unknown	8	8		8
		BRU-5	Unknown	8	8		8
		BRU-6	Unknown	8	8		8
Clear Lake	Clear Lake	CLR-0	Offshore	8		8	
		CLR-1	Residential	8	8		8
		CLR-2	Residential	8	8		8
		CLR-3	Residential	8	8		8
		CLR-4	Residential	8	8		8
Indian River	Indian River	IND-0	Offshore	1	8	8	8
		IND-1	Residential		8		8
		IND-2	Unknown		8		8
		IND-3	Unknown		8		8
		IND-4	Unknown		8		8
Joseph River	Joseph River	JOR-0	Offshore	4*		4	4
		JOR-1	Residential	4*			4
		JOR-2	Residential	4*			4
		JOR-3	Residential	4*			4
		JOR-4	Residential	4*			4
Leonard Lake	Leonard Lake	LEO-0	Offshore	8	8	8	8
		LEO-1	Residential		8		8
		LEO-2	Residential		8		8
		LEO-3	Residential		8		8
Mirror Lake	Mirror Lake	MIR-0	Offshore	8	8	8	8
		MIR-1	Creek	8	8		8
		MIR-2	Creek	8	8		8
		MIR-3	Residential	8	8		8
Moon River	Moon River	MOO-1	Offshore	8	8		8
		MOO-3	Residential	8	8		8
		MOO-4	Residential		8		8
		MOO-5	Residential		8		8
		MOO-6	Residential		8		8
		MOO-7	Camping		8	8	8
		MOO-8	Residential		8		8
		MOO-9	Residential/Creek (Culvert)		8		8
Muldrew Lakes	Muldrew Lakes	MLD-4	Residential		7		8
		MLD-5	Creek		7		8
		MLD-6	Residential		7		8
		MLD-7	Residential		7		8

Lake	Sampling Area	Site	Land Use	Phosphorus	Bacteria	Secchi	Temperature
Muskoka River	Muskoka River	MRV-1	River Mouth	1	8	8	8
		MRV-2	Wetland	1	8		8
		MRV-3	River	1	8	8	8
		MRV-4	Town (Bracebridge)	1	8		8
		MRV-5	Creek	8		8	8
Silver Lake (TML)	Silver Lake (TML)	SPC-0	Offshore	8		8	8
		SPC-1	Town (Port Carling)		8		8
		SPC-2	Residential		8		8
		SPC-3	Residential		8		8
Star Lake	Star Lake	STR-0	Offshore	1		8	
		STR-1	River		8		8
		STR-2	Residential		8		8
		STR-3	Creek		8		8
		STR-4	Residential		8		8
		STR-5	Residential		8		8
Sucker Lake	Sucker Lake	SUC-0	Offshore	1		8	
		SUC-1	Creek		8		8
		SUC-2	Residential		8		8
		SUC-3	Residential		8		8
		SUC-4	Residential		8		8
Sunny Lake	Sunny Lake	SUN-0	Offshore	1			
		SUN-1	Residential		8	8	8
		SUN-4	Residential		8	8	8
		SUN-5	Wetland/Creek		8	8	8

3.3.1. **Phosphorus**

Total phosphorus concentration (TP) was measured at the sites indicated in **Table 2**. Digest tubes were supplied by and returned to the Trent University Laboratory at the Ministry of Environment's Dorset Environmental Science Centre. Tubes were distributed to Team Leaders who subsequently distributed them to Team Members.

The tubes were filled directly from surface water to avoid potential problems relating to the 'container effect' in which phosphorus may adhere to the sides of sampling vessels and not be transferred to the digest tube used for analysis (Clark and Hutchinson, 1992). Volunteers used the 'plunge and sweep' method to fill digest tubes: they turned the tubes upside-down, plunged them into the lake to approximately forearm depth, turned the tube 90˚ and 'swept' upwards towards the surface, filling the tube to the indicated fill line. For the first sampling session in late May, volunteers took duplicate phosphorus samples at the deepwater reference sites (see Section 3.4.2.). Filled digest tubes were routinely delivered to Team Leaders who were tasked with dropping them off at the MLA office in Port Carling. Phosphorus samples were kept cool throughout this chain of custody and were submitted to the lab in Dorset biweekly.

3.3.1.1 Phosphorus Analysis

Results of this analysis are presented in the 2010 Water Quality Report.

Spring turnover and yearly mean total phosphorus values were calculated for all sites monitored in 2010. Current and historical total phosphorus data for the deepwater control site for a sampling area were graphed to show long-term trends. Where appropriate, graphs show MLA data in relation to the threshold concentration set by the DMM or Seguin Township. On the graphs illustrating long-term phosphorus levels, threshold concentrations have been represented by a single black dashed line. For sampling areas in the DMM, these values were verified by the DMM. Spring turnover and yearly mean phosphorus as measured by the MLA are shown in µg/L on the y-axis and sampling year is indicated on the x-axis.

Spring Turnover phosphorus concentration was calculated as the arithmetic mean of the spring or mid-May duplicate sample measurements. Spring turnover phosphorus concentrations at the deepwater reference site have been represented graphically as a blue line with diamonds or as single blue diamonds, if consecutive years of data were not available. Note that in previous years, duplicate spring turnover samples were not consistently collected at some sites and for these sites, a single spring turnover sample has been reported.

Yearly Mean phosphorus concentration was calculated as the arithmetic mean of all measurements from an individual sampling site within the sampling season, including duplicate sample measurements, where available. Yearly Mean phosphorus concentrations at the deepwater reference site have been represented graphically as a red line with circles or as single red circles if consecutive years of data were not available.

Nearshore-Deepwater Phosphorus Concentration Comparisons

In sampling areas where both nearshore and deepwater phosphorus samples were collected, a statistical test was performed to compare the data collected at each nearshore site to the data collected at the deepwater reference site. Nearshore sites identified to be significantly different from their deepwater reference site are identified in the area summaries under the Comments and Recommendations section.

A one-way Analysis of Variance (ANOVA) was completed on the log-transformed phosphorus data for each area. Transforming data is a technique used by statisticians to “normalize” data that is skewed

in one direction (e.g., when there are many more smaller values than larger values). Transforming data ensures valid statistical analysis when it may not be possible otherwise. If an overall significant difference ($P < 0.05$) was detected between the mean phosphorus values of all sampling sites in an area, a Multiple Comparison Test (the Holm-Sidak method) was conducted to determine which sites differed significantly from the deepwater reference site. In a small number of cases, log transformation did not normalize the data, and an Analysis of Variance on Ranks (Kruskal-Wallis test) was performed, with Dunn's Method as a multiple comparison test.

3.3.2. **Total Coliform**

Volunteers collected samples for total coliform analysis using 300 mL juice bottles. The bottles were purchased new from the Consolidated Bottle Company or reused from previous years. The bottles and caps were sterilized in boiling water by Team Leaders at the beginning of the sampling season.

The bottles were rinsed (completely filled and then emptied) with lake water three times prior to each sampling event. The bottle was then filled using the 'plunge and sweep' method described in Section 3.3.1. Samples were placed on ice in the field and returned to the Team Leader for analysis.

Bacterial sample analysis was completed as soon as possible after receiving all of the samples. The elapsed time was routinely within 24 hours of sample collection. The samples were kept on ice and in the dark to preserve the bacteria at their naturally occurring levels. Water from each sample was poured into a commercially available bacteria-testing kit, as shown in **Figure 1**. The kit is known by the trade name *ColiPlate*, and is manufactured by Bluewater Bioscience Inc.

Coliplates were left to incubate in egg incubators (*Hovabator* brand) at 37˚C for 24-26 hours prior to analysis. Team Leaders recorded the time of preparation and time of analysis on their datasheets. *ColiPlates* have 96 wells containing an agar that reacts with Coliform bacteria and turns blue. Actual bacterial counts are determined by comparing the number of blue cells to a table of Most Probable Numbers (MPN). The MPN table is shown in **Appendix B**.

Figure 1. ColiPlate with 11 blue wells

Wells identified as any shade of blue or green were counted as a positive blue well, as per instructions from Bluewater Bioscience.

3.3.2.1 Total Coliform Analysis

Results of this analysis are presented in the 2010 Water Quality Report.

Total coliform data was summarized for areas where bacterial monitoring was conducted in 2010. Total coliform levels are presented as yearly averages calculated as the geometric mean of all measurements for an individual sampling site. Note that the ColiPlates have a detection limit of three cfu/100mL (a count of zero blue wells corresponds to a count of "less than three" colony forming units per 100 mL of lake water). This difficulty was handled by assigning all readings of "less than three" cfu/100mL with an absolute value of 1 cfu/100mL.

3.3.3. ***Escherichia coli* (*E. coli*)**

After testing total coliforms, each *ColiPlate* was used to analyze for *Escherichia coli* (*E. coli*). This was done by exposing the plate to a 366 nm ultraviolet light. Wells that were both blue/green (as per protocol defined in Section 3.3.2.) and fluorescent under the UV light were identified as positive for *E. coli*. The number of blue/green-fluorescent wells was counted and the most probable number of cfu/100 mL was determined by comparison with the MPN tables.

After the readings were finished, the *ColiPlates* were emptied into a septic system and the plastic plates were rinsed and returned to the MLA Office. At the end of the season, MLA staff returned the plates to Bluewater Bioscience to be cleaned and reused.

The MLA WQI established a field protocol during 2010 that required volunteers to resample a site weekly if ***E. coli*** were greater than 50 cfu/100 mL over two consecutive sampling periods. This cautious approach allowed the MLA to increase monitoring effort for sites that demonstrated potential for ongoing concern.

3.3.3.1 E. coli Analysis

Results of this analysis are presented in the 2010 Water Quality Report.

E. coli data was summarized for areas where bacterial monitoring was conducted in 2010. ***E. coli*** levels are presented as yearly averages calculated as the geometric mean of all measurements for an individual sampling site. As with the total coliform measurements, all readings of "less than three" *E. coli* cfu/100mL were assigned an absolute value of 1 cfu/100mL. It is important to note that in creating the *E. coli* graphs, every site that was sampled had a minimum value of 1 cfu/100 mL; where no bar is shown for a particular site/year, no data was collected.

Current and historical ***E. coli*** data are presented graphically. ***E. coli*** concentrations are reported as the number of colony forming units observed in 100 mL of lake water (cfu/100 mL) on the y-axis and sampling sites are indicated on the x-axis. For these graphs, each sampling site is represented as a cluster of bars and different sampling seasons (years) are represented by different coloured bars. Each graph also compares ***E. coli*** levels to the MLA upper limit, which is represented by a grey dotted line. The upper limit value (10 cfu/100 mL) was established as a reasonable limit for maintaining existing water quality in Muskoka for the WQI and is based on advice provided by Dr. Karl Scheifer (2003).

3.3.4. **Secchi Depth**

A Secchi disk provides a measurement of water clarity and represents the distance that light will travel into the water column. Water clarity is typically affected by three different factors: algae, suspended sediment, and water colour. In 2010, a Secchi disk (**Figure 2**) was used to measure Secchi depth in metres. Each disk was attached to 15 metres of rope (length labelled at 50 cm intervals). To record the Secchi depth, the volunteer lowered the Secchi disk on the rope into the water on the shady side of the boat until they could no longer see it. At this point, the volunteer recorded the depth on the sample

date's data sheet, lowered the disk a little further, raised the disk towards the boat until it reappeared and recorded the second depth on the same data sheet. Secchi depth was calculated as the arithmetic mean of the "up" and "down" measurements.

Figure 2. Secchi disk demonstration.

3.3.5. **Temperature**

Temperature readings were recorded for all sites in degrees Celsius. Volunteers hung a pool thermometer from a rope into the surface water when first arriving at each site. After all of the other protocols were completed, the sampler read the thermometer and recorded the value. Volunteers also recorded air temperature on the datasheet.

3.3.6. **Rainfall**

Volunteers assessed the amount of rainfall that had occurred over the 24-hour period prior to sampling. Their observations were classified and recorded as "heavy", "moderate", "light" or "none". Rainfall data was occasionally used to assess the significance of high bacterial counts.

3.3.7. **Waves**

Volunteers assessed the amount wave action at each sampling site. The surface of the water was classified and recorded as either "calm" or "rough". Wave data was occasionally used to assess the significance of unusual secchi depth measurements.

3.4. Quality Assurance / Quality Control

The QA/QC protocols have been developed over the previous sampling years. The methods included in this document have been taken from Citizens' Environment Watch, 2009 and modified as necessary to reflect the 2010 program.

3.4.1. Quality Assurance

Reliability of experiments and results is paramount to the effective use of the scientific method. Collecting environmental data in the field is unfortunately 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 in the WQI. As in previous years since 2002, rigorous training, documentation and duplicate measures were used throughout the 2010 season.

Quality assurance (QA) is a set of systematic procedures designed to ensure reliable results. While QA cannot guarantee quality results, it improves the likelihood of achieving quality results.

Quality Control (QC) is a set of protocols that report on the reliability of results. QC is therefore the measure of reliability.

3.4.2. Phosphorus Quality Control

Some quality control measures were in place for the phosphorus sampling protocols. All duplicates took place during the spring turnover period at deepwater reference sites. The samples were collected at the same time as the regular phosphorus samples using identical TP tubes and protocols. The duplicate measurements show the range of phosphorus results that can be expected because of variability in sampling and laboratory testing.

Table 3. Phosphorus Duplicates for 2010 Quality Control

Site	Sample Number	Sample Date	Phosphorus Concentration (ug/L)	Phosphorus Duplicate (ug/L)	Absolute Difference
ART-0	1	05/07/2010	5.4	4.8	0.6
BAL-0	1	05/16/2010	3.7	3.5	0.2
BAS-0	1	05/23/2010	13.5	6.5	7
BAS-1	1	05/23/2010	7.3	7.2	0.1
BDY-0	1	05/23/2010	16.6	16.5	0.1
BMR-0	1	05/23/2010	7.6	7.8	0.2

Site	Sample Number	Sample Date	Phosphorus Concentration (ug/L)	Phosphorus Duplicate (ug/L)	Absolute Difference
BOY-0	1	05/24/2010	7.4	5.2	2.2
BRA-0	5	07/18/2010	13.5	13.1	0.4
BRU-0	1	05/23/2010	7.8	7.1	0.7
CLR-0	1	05/23/2010	5.2	8.5	3.3
COX-0	1	05/23/2010	3.4	5.4	2
EAS-0	1	05/23/2010	4.5	4.6	0.1
ELG-0	1	05/23/2010	7.6	4.7	2.9
FTB-0	2	06/06/2010	3.1	0.6	2.5
FTB-0	5	07/19/2010	3.8	3.5	0.3
GNB-0	1	05/14/2010	5.6	3.4	2.2
HMB-0	1	05/22/2010	5.1	5.5	0.4
IND-0	1	05/23/2010	5.9	4.9	1
JOR-0	5	07/16/2010	9.9	9.3	0.6
JOS-1	1	05/23/2010	7.6	6.2	1.4
LEO-0	1	05/22/2010	5.8	8.5	2.7
LLJ-0	1	05/23/2010	4.7	8.3	3.6
MBA-0	1	05/24/2010	7.7	10.8	3.1
MGN-0	1	05/23/2010	5.5	5.6	0.1
MIN-0	1	05/23/2010	3.7	4.6	0.9
MIR-0	1	05/23/2010	5.7	7.6	1.9
MLG-0	1	05/07/2010	3.5	3.2	0.3
MOO-1	1	05/21/2010	4.8	5.4	0.6
MOO-3	1	05/21/2010	4.2	4.5	0.3
MRV-1	1	05/24/2010	7.4	5.7	1.7
MRV-2	1	05/24/2010	6.9	6.8	0.1
MRV-3	1	05/24/2010	6.8	6.5	0.3
MRV-4	1	05/24/2010	6.8	6.7	0.1
MSN-0	1	05/24/2010	6.9	4.4	2.5
MUS-2	1	05/16/2010	3.9	3.7	0.2
NRT-0	1	05/16/2010	4.5	3.8	0.7
POR-0	1	05/24/2010	3.8	6.1	2.3
RMI-0	1	05/07/2010	7.1	6.8	0.3
ROS-1	1	05/07/2010	7.1	5.6	1.5
RSH-0	1	05/23/2010	4.8	5.8	1
SKB-0	1	05/21/2010	3.5	4	0.5
SPC-0	1	05/23/2010	7.5	6.0	1.5
STI-0	2	06/06/2010	12.8	1.4	11.4
STN-0	1	05/23/2010	3.7	4.4	0.7

Site	Sample Number	Sample Date	Phosphorus Concentration (ug/L)	Phosphorus Duplicate (ug/L)	Absolute Difference
STR-0	1	05/24/2010	7.3	8.2	0.9
SUC-0	1	05/24/2010	4.2	3.6	0.6
SUN-0	1	05/24/2010	13.0	9.9	3.1
TOB-0	1	05/07/2010	8.0	7.6	0.4
WIN-0	1	05/26/2010	8.7	5.9	2.8
WLB-0	1	05/24/2010	5.1	5.1	0
WTS-0	1	05/16/2010	4.8	4.6	0.2

A complete analysis of "outliers" or data points that seem to be skewed from the normal data set was not completed; however, in graphing data for the 2010 Water Quality Report, some outliers were identified and removed from the data set. These data points were easily identified when compared to the other long term or duplicate data points and were likely the result of sample contamination. Sample contamination can result when a zooplankton or piece of other material is accidentally collected as lake water is swept into the tube, resulting in elevated phosphorus concentrations. Outliers are identified in the "2010 Data" and "Trends" sections of the area summaries.

3.4.3. **Bacteria Quality Control**

Limited quality control measures were conducted on bacteria samples in the 2010 sampling season. Team Leaders were asked to include a single "blank" *Coliplate* during bacterial analysis of samples collected during sampling windows 1 and 4. These *Coliplates* were filled with bottled water (provided) and then incubated, and analyzed along with the lake water samples. Blank plates were labeled with a corresponding sampling area code followed by an "X". This control step was specifically intended to determine whether the sample preparation, incubation, and analysis process was producing false positive results. Bacterial quality control results for 2010 are shown in **Table 4**. Note that Team Leaders responsible for multiple sampling areas were only required to include one blank plate per round of bacterial sample analysis as opposed to one blank per sampling area.

Table 4. Bacterial Blanks for 2010 Quality Control

Site	Sample Date	Total Coliforms	*E. coli*
BAL-X	05/23/2010	<3	<3
BAL-X	07/17/2010	<3	<3
BAS-X	07/03/2010	<3	<3
BDY-X	05/23/2010	<3	<3
BDY-X	07/03/2010	<3	<3
BMR-X	05/23/2010	3	<3
BMR-X	07/16/2010	<3	<3
BRA-X	05/22/2010	<3	<3
BRA-X	07/03/2010	<3	<3
BRU-X	05/23/2010	<3	<3
BRU-X	07/04/2010	<3	<3
COX-X	05/23/2010	<3	<3
COX-X	07/03/2010	<3	<3
CLR-X	05/23/2010	<3	<3
CLR-X	07/03/2010	<3	<3
ELG-X	05/23/2010	<3	<3
ELG-X	07/04/2010	<3	<3
HMB-X	05/22/2010	<3	<3
HMB-X	07/02/2010	<3	<3
IND-X	05/23/2010	<3	<3
IND-X	07/04/2010	<3	<3
LEO-X	05/22/2010	<3	<3
LLJ-X	05/23/2010	<3	<3
LLJ-X	07/04/2010	<3	<3
MBA-X	05/24/2010	<3	<3
MBA-X	07/05/2010	<3	<3
MGN-X	07/04/2010	<3	<3
MIN-X	05/23/2010	<3	<3
MIN-X	07/03/2010	<3	<3
MLD-X	05/24/2010	<3	<3
MLD-X	07/05/2010	<3	<3
MOO-X	05/21/2010	<3	<3
MOO-X	07/02/2010	<3	<3
MSN-X	05/24/2010	3	<3
MSN-X	07/03/2010	<3	<3
SPC-X	07/02/2010	<3	<3
SPC-X	05/23/2010	<3	<3
STR-X	05/24/2010	<3	<3
STR-X	07/04/2010	<3	<3

Site	Sample Date	Total Coliforms	*E. coli*
SUC-X	07/02/2010	<3	<3
SUN-X	05/23/2010	<3	<3
SUN-X	07/04/2010	<3	<3
WIN-X	06/07/2010	5	<3
WIN-X	07/04/2010	<3	<3
WLB-X	05/24/2010	<3	<3

4. DEFINITIONS

Arithmetic mean: This type of average is calculated by adding together a group of numbers and dividing the sum by the number of numbers.

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.

Eutrophic: A eutrophic lake typically has phosphorus concentrations above 20 µg/L (Level 3 - nutrient-rich, MOE). These lakes have elevated primary productivity due the high nutrient content. These lakes also have high algal production, and consequently, often have low water clarity with low drinking-water quality. Eutrophic lakes are typically abundant in aquatic vegetation and can support a large number of fish species.

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. Geometric mean is used to indicate the central tendency or typical value of a set of numbers. It is typically used to calculate average bacteria counts because as a living organism, bacteria counts are highly sporadic and inconsistent.

Lake System Health Monitoring Program: A field-based program designed and operated by the DMM that monitors approximately 192 sample locations across Muskoka on a rotating basis depending upon development pressures and the specific characteristics of the lake. The purpose of the program is to establish a long-term record of key water quality parameters so that trends in water quality can be identified. Spring turnover total phosphorus results of this program inform Muskoka's Recreational Water Quality Model.

Mesotrophic: A mesotrophic lake typically has phosphorus concentrations between 10 and 20 µg/L (Level 2–mid-range, MOE). Mesotrophic lakes are lakes with an intermediate level of productivity, greater than oligotrophic lakes, but less than eutrophic lakes. These lakes are commonly clear water lakes and ponds with beds of submerged aquatic plants and medium levels of nutrients.

Oligotrophic: An oligotrophic lake typically has phosphorus concentrations less than 10 µg/L (Level 1–nutrient-poor, MOE). These lakes have low primary productivity, due to the low nutrient content. These lakes have low algal production, and consequently, often have very clear waters, with high

drinking-water quality. The bottom waters of such lakes typically have ample oxygen; thus, such lakes often support many fish species, like lake trout, which require cold, well-oxygenated waters.

OBBN: (Ontario Benthic Biomonitoring Network) The Ministry of the Environment and Environment Canada have developed an aquatic macroinvertebrate biomonitoring network for Ontario's lakes, streams, and wetlands. The program is built on the principles of partnership, free data sharing, and standardization. The OBBN is biological monitoring program (not chemistry) that uses a reference-condition approach to define criteria: samples from minimally impacted sites define an expectation (the normal range) for biological condition at a test site. Assessments evaluate whether a test site's biological condition is within the normal range. New partnerships, and the ability to generate local information on aquatic condition, will build capacity for adaptive water management and enhance the link between science and decision-making (Jones et al, 2006).

Threshold: The "Threshold" phosphorus concentration is 50% more than the baseline (Background) concentration calculated by the District of Muskoka or Seguin Township. The threshold is used to classify lakes and bays as requiring a higher level of development control as a precautionary action to protect the long-term health of the lake.

Total Coliform: Coliform include a variety of bacteria. In practice, detectable coliform are usually enteric, found in the intestinal tracts of humans and other warm-blooded species.

***E. coli*:** Fully known as *Escherichia coli*, it is a subset of total coliforms, and is exclusively associated with fecal waste (Schiefer, 2001) making it a good indicator of faecal 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 (OMH, 2001).

Total Phosphorus: Phosphorus is a chemical element that is essential for all living cells. Amongst other sources, it is found in fertilizers, soaps, and in human waste. Typically phosphorus is not removed from waste streams by conventional private treatment systems (septic systems) nor by some municipal treatment systems.

Muskoka Recreational Water Quality Model: An advanced numerical model operated by the District of Muskoka designed to predict the response of all individual lakes in Muskoka to the input of phosphorus. The model is based on the Ontario Lakeshore Capacity Simulation Model, originally published in 1986 by a Provincial inter-ministerial working group. This model was substantially updated in 2005 by Dr. Neil Hutchinson of Gartner Lee Ltd. for the District of Muskoka (GLL, 2005). The model includes a detailed phosphorus budget. Its inputs are the results of the District's Lake System Health Monitoring Program. Among the model's outputs is lake-specific Natural Phosphorus, Phosphorus Threshold and predicted phosphorus concentrations.

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

Secchi Depth: A measure of water **clarity**, measured using a Secchi disk - a small disk attached to a rope. Alternating quarters of the top side of the disk 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 disk.

Sampling Site: The discrete and unique location where samples are to be collected on each sample date.

Spring Turnover Phosphorus: A single phosphorus concentration measurement taken in a typically stratified lake during the spring turnover period. This measurement has been shown to adequately represent the overall phosphorus concentration in a lake (Clark and Hutchinson, 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 (http://www.ene.gov.on.ca/envision/water/lake_partner/index.htm).

Yearly Mean Phosphorus: The arithmetic mean of phosphorus concentration measurements taken above a stratified water column's thermocline over the ice-free period. ***Note:*** **yearly mean phosphorus concentration as reported by the WQI is for summer months only.**

Note: many of these definitions have been taken from the WQI Monitoring Program Summary Report - Citizens Environment Watch 2009.

5. REFERENCES

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APPENDIX A

2010 Data Sheet

Muskoka Lakes Association
Serving Muskoka since 1894

Sample Area: _____

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

Secchi Depth

Site:		**'Down' Depth:**		**'Up' Depth:**	
Site:		**'Down' Depth:**		**'Up' Depth:**	
Site:		**'Down' Depth:**		**'Up' Depth:**	

For Lab use

Preparation Time:		**Analysis Time:**	

Site Specific Information

Site Code	Water Temp.	Waves	Water Depth	Distance fr. shore	Blue	TC Count	Flor.	EC Count
		☐ Calm ☐ Rough						
		☐ Calm ☐ Rough						
		☐ Calm ☐ Rough						
		☐ Calm ☐ Rough						
		☐ Calm ☐ Rough						
		☐ Calm ☐ Rough						
		☐ Calm ☐ Rough						
		☐ Calm ☐ Rough						
		☐ Calm ☐ Rough						

APPENDIX B

MPN Table

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)