Bacteria Testing

Quality Assurance Project Plan

CFE/Save the Sound

January 25, 2016

Save the Sound
545 Tompkins Avenue
Mamaroneck, NY 10543

Table of Contents

1. Title and Approval Page

Save the Sound / Connecticut Fund for the Environment
Water Quality Program
Effective Date of Plan: June 22, 2015

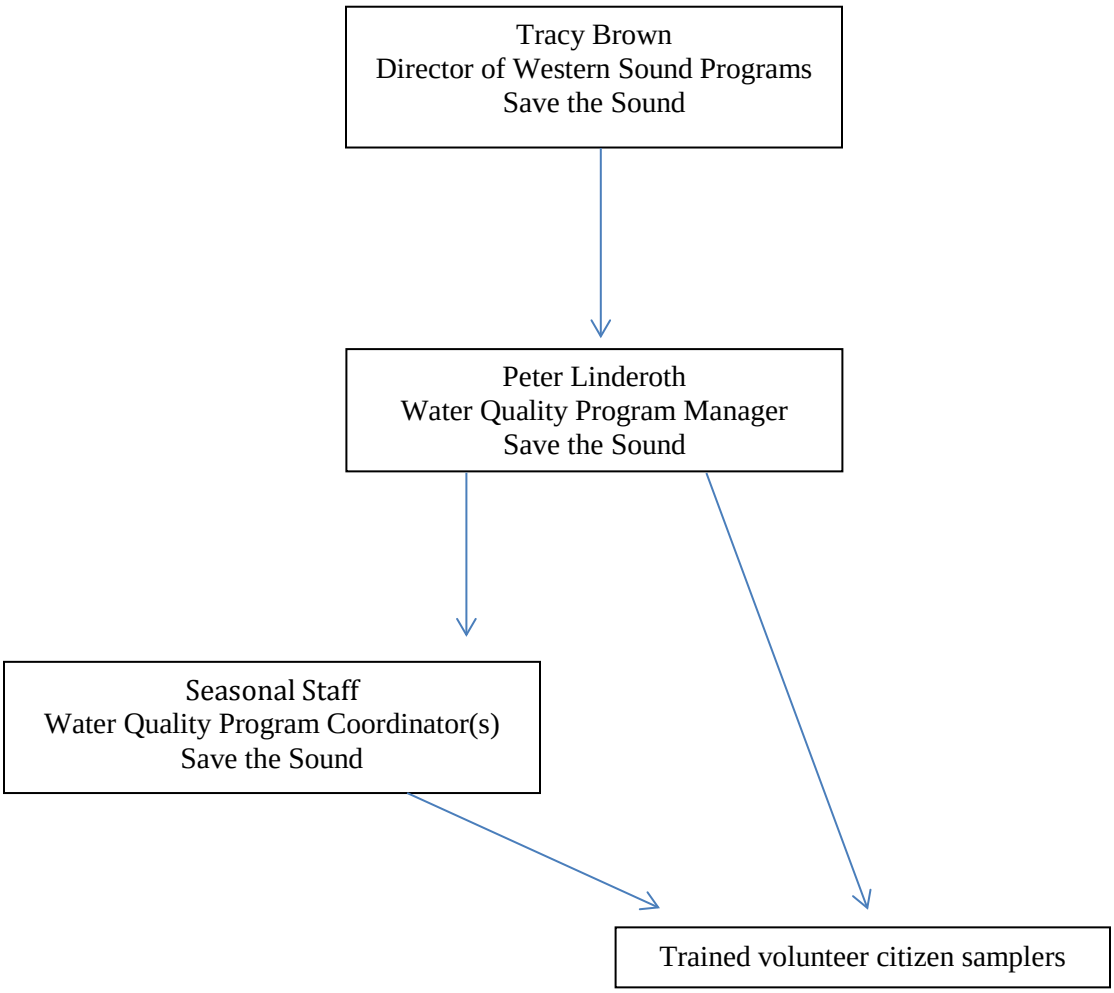
1/25/2016

Tracy Brown, Director of Western Sound Programs — Date
Save the Sound / Connecticut Fund for the Environment

1/25/2016

Peter Linderoth, Water Quality Program Manager — Date
Save the Sound / Connecticut Fund for the Environment

2. Project Organization Chart



3. Document Distribution List

Name/Title	Contact Information
Tracy Brown Director of Western Sound Programs Save the Sound	(914) 381-3140 tbrown@savethesound.org
Peter Linderoth Water Quality Program Manager Save the Sound	(914) 263-6233 plinderoth@savethesound.org
Seasonal Staff Coordinator(s), Water Quality Program Save the Sound	via email
Citizen Science Samplers	via email and website

In addition to being directly distributed to the individuals above, this document will be posted on the Save the Sound website where it may be downloaded by the general public.

4. Project and Task Organization

Name/Title	Responsibilities
Tracy Brown Director of Western Sound Programs Save the Sound	Provides overall direction for the Water Quality Program; Supervises program staff; Coordinates sampling site selection; Performs sampling, sample processing, and sample analysis; Conducts public policy and outreach
Peter Linderoth Water Quality Program Manager Save the Sound	Operates the Save the Sound laboratory, including supply purchasing and equipment maintenance; Performs sampling, sample processing, and sample analysis; Supervises seasonal staff; Trains citizen samplers; Maintains the QA Project Plan; Maintains online database and data records; Assists with water quality reports and program operations
Seasonal Staff Water Quality Program Coordinator(s) Save the Sound	Coordinates daily site assignments, sampling equipment distribution and sample delivery to Save the Sound laboratory; Performs sampling, sample processing, and sample analysis; Assists with lab operations; Supports consistent execution of QA Project Plan; Assists with online database and data records

Volunteer Citizen Scientists	Conduct field sampling. Some of the volunteers may be trained to work in the laboratory.

5. **Training and Specialized Experience**

5.1 Field

Field personnel will be trained in sample collection and handling by Save the Sound staff. Group trainings will be organized and take approximately 60 minutes, depending on the number of people. Training is mandatory for all samplers before they go into the field. All samplers, including trained samplers from previous seasons, are required to attend the annual training before they commence in sampling for the respective season. The Water Quality Program Manager will keep a list of trained samplers for each season which will be stored on the Save the Sound server.

5.2 Laboratory Specialized Experience

All unsupervised staff and volunteers working in the laboratory will complete and pass a demonstration of capability (DOC) test. The DOC will be identical to the Enterolert laboratory fortified blank described in section 8.1 *Quality Objectives* of this QAPP. Volunteers working in the lab under staff supervision will be required to read all respective methodology for their analyses and participate as an observer with monitored handling of samples for at least one day in the lab before taking on additional responsibilities.

5.3 Specialized Experience

Person	**Specialized Experience**	**# of Years Experience**
Tracy Brown Director of Western Sound Programs	Collection and analysis of water samples for presence of *Enterococcus*	6
Peter Linderoth Water Quality Program Manager	Collection and analysis of water samples for multiple parameters including *E.coli* and Total Coliform	4
Seasonal Staff Water Quality Program Coordinator(s)	Collection and analysis of water samples for multiple parameters	minimum 1 year required

6. Problem Definition and Project Objectives

6.1 Problem Definition

Western Long Island Sound is an area of environmental concern in the Long Island Sound estuary Sewage and polluted runoff close beaches too frequently and hurts fish and wildlife by sucking life-giving oxygen out of the water. Beach closures have more than tripled in the Sound over the past two decades, according to the United States Environmental Protection Agency (USEPA) data. In 2012, the Sound suffered approximately 1,500 lost beach days when beaches, primarily in the western basin, were closed because of high levels of bacterial pollution. Public education on these challenges is desperately needed. The general public is relatively unaware of the problems posed by leaking sewer pipes that drain into Long Island Sound and how the resulting bacterial pollution is implicated in the rising number of beach closings.

More widely available water quality data will educate the public and engage them in local and regional strategies to reduce sewage discharges and improve water quality.

The Water Quality Monitoring Program addresses the following questions:

- Where is fecal contamination entering Long Island Sound and its tributaries?
- What is the severity of fecal contamination?

6.2 Project Objectives

The objective of the program is to collect water samples to analyze fecal contamination concentrations in Long Island Sound and its tributary waterways, including upstream and downstream of potential sewage pollution sources, at sub-tributary confluences, and at public access points.

Save the Sound will make the program data available online and use the data to educate the public and engage them in local and regional strategies to reduce sewage discharges and improve water quality. Data will be shared with municipal leaders in all communities Save the Sound conducts sampling. Data will also be shared with regulatory agencies.

6.3 Data Users

Save the Sound urges citizens to use data from this program to inform themselves about water quality conditions in their communities and to pursue local solutions to pollution problems. Save the Sound staff use the water quality data to advocate for increased investment in wastewater infrastructure, better enforcement of existing water quality protections, more frequent water quality sampling, and better prediction and public notification of sewage contamination.

Save the Sound rates the water quality at each site based on the current New York State Department of Environmental Conservation (NYSDEC) Recreational Water Quality Criteria or the Connecticut Department of Energy and Environmental Protection (CTDEEP) Water Quality Standards when applicable. The *Enterococcus* and *E. coli* counts and pass/fail ratings are uploaded to our website (http://www.ctenvironment.org/#!water-quality/cuk0) promptly after each sampling event.

The website explains the sampling method and rating system that we use. We will launch a new website, the Sound Data Explorer, where Save the Sound data will be displayed alongside other citizen science data as well as town, county and state water quality data.

We periodically release reports summarizing data and findings. These reports explain our methods, highlight key findings, and suggest ways for citizens to take action. Save the Sound's reports include guidance about how our water quality data should be interpreted. The reports are available on the Save the Sound website for viewing or download, and are available in print format upon request.

Periodically data are sent to municipal leaders serving in the Towns and Cities Save the Sound conducts sampling. Data are also sent to regulatory groups such as NYSDEC, CTDEEP, and USEPA.

7. Background and History

7.1 Background

The majority of beach closings and advisories in the United States are due to high levels of sewage contamination. In the Long Island Sound watershed, aging wastewater infrastructure and overburdened sewage systems are major causes of stream, river, and harbor impairments.

With more than 40 years of history working to restore and improve water quality in Long Island Sound, Save the Sound is the only nonprofit organization addressing these issues in the Western Sound region.

7.2 History

In 2013, Save the Sound initiated a pilot water quality monitoring program in the Village of Mamaroneck. A corps of citizen volunteers were trained in sampling methods and set about collecting water samples, which were analyzed by a local state-certified laboratory. In the summer of 2014, we expanded the citizen-scientist water quality monitoring program, increasing the number of samples collected more than fourfold, from 42 to 200 samples. We tested water quality at 36 locations in Mamaroneck, Rye, Harrison, Larchmont, White Plains, Pelham, Mount Vernon and the Bronx, adding 17 new locations. The number of volunteers who participated in the program doubled, from 7 to 15. We worked with two state-certified laboratories to support the expansion. Specifically, we partnered with Harborwatch in Norwalk, Connecticut and the Westchester County Environmental Lab in Valhalla, New York.

In 2015, thanks to the USEPA Citizen Science Equipment Loan Program, we were able to set our own sampling schedule. We will also be able to keep sampling costs down and continue to increase the scope of this program using USEPA and Save the Sound equipment in our own lab in Mamaroneck, New York, with the guidance of this QAPP and USEPA guidance documents.

8. Project Location

Save the Sound's Bacteria Testing Program focuses on the western basin of Long Island Sound. We sample in Long Island Sound along the Westchester County and western Connecticut shoreline, as well as in the tributary rivers, streams and creeks that feed the Sound. Each season we establish select sampling locations that we return to year after year to form a baseline for our study. In 2015 we sampled Westchester County locations in Pelham, Larchmont, New Rochelle, Town of Mamaroneck, Mamaroneck Village, Harrison, White Plains, Mount Vernon, City of Rye, and Port Chester, NY. We also sampled Fairfield County locations in Greenwich, CT. In addition to sampling at our baseline sites we will test water quality at additional exploratory sites as opportunities arise. As our program grows we will explore expansion further into CT as well as begin a sampling regime in other NY waters including Long Island.

8.1 2015 Waterways tested

United States Geological Survey HUC 12 Watershed	**Waterway(s) Sampled**
Hutchinson River - Eastchester Bay (HUC 12 020301020201)	Pelham Lake Hutchinson River
East Creek - Mamaroneck Harbor (HUC12 20301020203)	New Rochelle Harbor Western Long Island Sound Westchester County Waters Larchmont Harbor Premium River Mamaroneck Harbor Milton Harbor Van Arminge Millpond
Sheldrake River - Mamaroneck River (HUC12 20301020202)	Sheldrake River Mamaroneck River Guion Creek Otter Creek Beaver Swamp Brook
Blind Brook - Horseneck Brook (HUC 11000060405)	Greenwich Cove Horseneck Brook Byram Harbor Playland Lake Westchester County Waters
Byram River (HUC 11000060403)	Byram River East Branch Byram River Pemberwick Creek
Mianus River (HUC 12 11000060402)	Mianus River

9. Project Schedule

Activities	Person Responsible	Timeframe
Organize and train citizen samplers	Water Quality Program Manager	April - June
Purchase supplies	Water Quality Program Manager	Year-round
Conduct Sampling	Save the Sound Staff, Citizen Samplers	June - November
Perform Sample Analyses	Save the Sound Staff	June - November
Post Results Online	Water Quality Program Manager	June - November
Data QA/QC	Water Quality Program Manager	June - December

10. Quality Objectives

10.1 Quality Assurance - Laboratory

All Quality Control (QC) procedures except for the lab fortified positive and negative controls will be prepared on a per batch basis. The lab fortified positive controls will be prepared on a weekly basis. The lab fortified negative controls will be prepared on a per lot basis, up to but not exceeding 200 samples.

Laboratory Blank (Enterolert and Colilert-18):

A laboratory blank will be conducted for each batch of samples. The laboratory blank is prepared with 100 ml (99 ml nominal of commercially prepared sterile deionized water and IDEXX reagent). The laboratory blank provides information on lab procedures and materials. Fluorescing wells should not be observed in the laboratory blank. If contamination is present data will be qualified as estimated value or invalidated (R) if above the reporting limit.

Enterolert Laboratory Fortified Blank:

A laboratory fortified blank will be conducted each week of sampling. The laboratory fortified blank is prepared with 100 ml (99 ml nominal volume of commercially prepared sterile deionized water, IDEXX reagent, and one *Enterococcus faecalis* QC pellet).

The laboratory fortified blank should produce fluorescing wells. If there are no fluorescing wells, sample data for the week are invalid (R) unless sound reasoning is documented. The true value provided for the *Enterococcus faecalis* QC pellet will be used to calculate the % Recovery. The % Recovery will be expected to meet 50 – 200% of the true value. If this number is found to be outside the interval, laboratory procedures will be evaluated.

Enterolert Negative Controls:

Two laboratory negative control tests will be conducted for each lot of samples. The laboratory negative control tests are prepared with 100 ml (99 ml nominal volume of commercially prepared sterile deionized water, IDEXX reagent, and one pellet of *Staphylococcus aureus* or *Eschericia Coli).*

The laboratory negative control tests should not produce any fluorescing wells. This test evaluates the effects of non-*Enterococcus* bacteria with the IDEXX reagent. If there are fluorescing wells, we will evaluate laboratory procedures, repeat this test, and potentially purchase new reagent.

Colilert-18 Laboratory Fortified Blank:

Two laboratory fortified blanks will be conducted for each week of Coliform sampling. The laboratory fortified blanks are prepared with 100 ml (99 ml nominal volume of commercially prepared sterile deionized water, IDEXX reagent, and one *Enterobacter aerogenes* QC pellet for total coliform or one *Escherichia coli* QC pellet for *E. coli).*

The laboratory fortified blank for *E.coli* should produce fluorescing wells. If there are no fluorescing wells, sample data for the week are invalid (R) unless sound reasoning is documented. The true value provided for the *Escherichia coli* QC pellet will be used to calculate the % Recovery. The % Recovery will be expected to meet 50 – 200% of the true value. If this number is found to be outside the interval, sampling procedures will be evaluated.

The laboratory fortified blank for *Enterobacter aerogenes* should produce positive observations for total coliforms, but not for the presence of *E.coli*. If there are no fluorescing wells, sample data for the week are invalid (R) unless sound reasoning is documented. The true value provided for the *Enterobacter aerogenes* QC pellet will be used to calculate the % Recovery. The % Recovery will be expected to meet 50 – 200% of the true value. If this number is found to be outside the interval, sampling procedures will be evaluated.

Colilert-18 Negative Controls:

A laboratory negative control test will be conducted for each lot of samples. The laboratory negative control test is prepared with 100 ml (99 ml nominal volume of commercially prepared sterile deionized water, IDEXX reagent, and one pellet of *Pseudomonas aeruginosa*).

The laboratory negative control tests should not produce any fluorescing wells or yellow coloration indicating positive results for *E. coli* and total Coliforms, respectively. This test evaluates the effects of non-Coliform bacteria with the IDEXX reagent. If there are fluorescing wells or yellow coloration, we will evaluate laboratory procedures, repeat this test, and potentially purchase new reagent.

Sample Duplicate:

A random site on a day's sampling will be selected for duplication of results in the laboratory. The duplicate will be prepared with identical methods as all other samples. A relative percent difference (RPD) will be calculated from these results. If an RPD is calculated > 30%, test procedures, homogenization, reagents, and others parameters will be reviewed on a case by case basis. Samples obtained on a day the duplicate RPD is in excess of 30% will be quantified with a J qualifier as defined by USEPA.

Field Blank:

A selective random site (rotating) on a day's sampling will be selected for a field blank. The field blank will be prepared by a sampler on the sampling site prior to waterway sampling. The sampler will prepare the field blank sample by pouring 99 ml commercially obtained sterile water into a sampling vessel and then placing the vessel in a cooler for laboratory analysis.

The field blank will indicate if there are any issues with sampler contamination and serve as a temperature control for a batch of samples. If a field blank tests positive for fecal indicating bacteria, the samples from that sampler will be invalid for the date and field methods will be questioned. Field blank temperature will be taken with an infrared thermometer upon arrival to the lab. If a field blank temperature is above 10 °C the temperature will be recorded on the data sheet and data for that sampling event will receive a J designation and sampling procedures will be evaluated. Extreme temperature exceedances may result in disqualifying data. In the case of a short commute to the testing facility, temperature will be recorded but evidence of icing will negate the J qualifier.

10.2 Quality Assurance – Field

Citizen scientists and Save the Sound staff will closely follow assigned field protocol. Please see section 11. Data Collection Methods for detailed information regarding field sampling.

10.3 Bias

Our study is intended to detect fecal contamination at specific locations and times. Sampling points are distributed to provide insight into possible sewage exposure by water users, to locate contamination hot spots, and to identify potential pollutant sources. The full subset of samples may not always describe general waterway conditions. Cumulative results could be biased higher as some sample sites are selected with potential fecal contamination sources considered.

10.4 Representativeness

Citizen scientists will sample approximately once or twice weekly from June to September, which is the time of year when most people get in the water. These data are not to be used to predict conditions at a specific time and place along a waterway. In addition, data should not be used to draw conclusions about conditions during the winter months.

10.5 Comparability

The Long Island Sound and its tributaries include saltwater, freshwater, and brackish environments. We use *Enterococcus* as an indicator of sewage contamination in all NY and CT waterbodies.

At some tributaries that are not tidally influenced (not brackish) we may sample for *E. coli* and total coliforms in addition to, or instead of *Enterococcus*. *E. coli* and total coliforms will be tested when an existing or historic dataset for that location uses *E. coli* and total coliforms instead of *Enterococcus.* Current classifications and corresponding water quality criteria of a waterway segment will also be considered when selecting the type of indicator bacteria characterized in sampling efforts.

We will rate water quality in comparison to NYSDEC and CTDEEP guidelines for safe swimming so that our results can be compared with historic local datasets and other sites around the region. In some cases, data will be shared with groups with consideration given to other water quality criteria.

10.6 Completeness

Field:

The objective for all of the weekly monitored sites is one sample a week for eight weeks. If that objective is not met, a minimum of five samples will be used to report a geometric mean for the respective site. If less than five samples are collected, weekly values will be reported but a geometric mean will not be calculated.

Sampling at stormwater outfalls will be similar to weekly monitored sites but frequency will be dependent on weather.

Laboratory:

The IDEXX Quanti-Tray ® 2000 Enterolert and Colilert-18 Most Probable Number (MPN) method s allows detection of 1 *Enterococcus*, total coliform, and *E. coli* per 100 ml in undiluted freshwater samples. As per standard methods, samples collected in saline or brackish waters (or when higher maximum detection levels are required) are diluted tenfold, so the lower limit at those sites is 10 Enterococci per 100 ml. The MPN method can quantify up to 2,419.6 Enterococci per 100 mL without dilution (24,196 with a tenfold dilution).

11. Data Collection Methods

11.1 Site Names

Each site is assigned an ID consisting of the site type (e.g., "R" for river, "B" for recreational beach, "S" for shoreline, "E" for embayment, "L for lake", "O" for outfall), waterway initials and approximate river mile when applicable (e.g., "R-MR.24"). Lower case letters are used for multiple sites in a waterbody other than a river (e.g., "E-MHb") Each site is also assigned a name usually consisting of the waterway and/or the municipality or another local landmark (e.g., "Mamaroneck River at Joint Waterworks").

Waterway names are identified using EPA, NYSDEC, and CTDEEP assessment and impairment documents. This mostly applies to sites along the Long Island Sound coastline to account for the differences in regulatory and local naming observed in some embayments.

11.2 Sample Site Selection Process

Site selection is informed by reviewing the NYSDEC Priority Waterbodies List, CTDEEP Integrated Water Quality Report, the United States Environmental Protection Agency (EPA) 303(d) list, MS4 reports and audits, EPA LIS Beacon Database, sanitary sewage overflow reports, regional testing that has already been undertaken, Sewage Pollution Right to Know discharge reports and other sources as they become available. We also solicit input from community groups, state and local environmental agency staff, members of sports and outdoor associations, and other people with local knowledge.

Ease of access, permanence of access, and volunteer safety are also considered during sample site selection. If sites become inaccessible, relocation is considered on a case-by-case basis. Please see section 9.4 *Comparability* of this document for information on parameter selection.

Sample sites will be grouped together according to United States Geological Survey Hydrologic Units identified with Hydrologic Unit Code (HUC) 12 classifications. Sites within a HUC 12 watershed boundary will be sampled together on any given weekly monitoring sampling event. Section 7.1 Waterways to be tested lists the sampled waterways and respective HUC 12 watersheds for the 2015 season. Please see Appendix 1- *2015 Sample Sites with HUC 12 Watersheds* for sites with HUC 12 delineations.

11.3 Precipitation Records

Accumulative precipitation on the day of sampling, up to fist sample, and 72 hours prior will be recorded using the open source website WeatherUnderground (http://www.wunderground.com/). Only stations with consistent data, current and historic, will be used for precipitation data. Site Ids with respective weather stations can be reviewed in Appendix 2 - 2015 Sample Site Information.

11.4 Sampling Procedures and Laboratory Methods

Sampling will occur weekly from June to September. Exploratory and wet-weather sampling can continue through November. Water samples will be collected using sterile polyethylene bottles containing sodium thiosulfate. Save the Sound staff will pre-label sample containers with the site id, site name, sampling date, and a blank space for the sampler to mark the sampling time. Samplers will collect the containers from Save the Sound staff the day of sampling. All exchanges of sampling containers will be recorded on a chain of custody sheet with other important information (Appendix 3).

Samplers will be given a site map sheet for all sites that they are scheduled to sample. The site map sheet contains a numbered set of instructions upon arrival to site including field methods (Appendix 4).

All samplers will wear disposable gloves, switched at each sample site, to avoid contamination of the sample or the collection bottle. The sampler will record the time of sample collection on the bottle with a permanent marker prior to taking sample. When possible the sample vessel will be lowered below the surface of the water with the cap on and opening oriented towards the surface. The sampler will unscrew the cap to fill the vessel then slowly bringing the sampling vessel to the surface. The cap will promptly be screwed back on the bottle before transport to the cooler. The samplers are instructed to keep the sodium thiosulfate in the bottle at the time of sampling. If necessary, the cap of a sample bottle will be removed and the sample obtained with a pole for extended reach. In this event, the sample will be held at a slight to moderate angle while submerged in water to ensure water is collected under the surface layer. All other procedures in the event of using a sampling pole remain the same as typical sample collection.

Tributary samples will be taken from the stream banks with minimal disturbance to sediments. If a sampler must wade into the tributary to take the sample, care will be taken to not stir up sediment and the sample will be taken facing upstream from the entry point. Shoreline, embayment, and recreational beach samples will be taken after wading into the water to just below knee height and waiting for any disturbed sediment to settle. Samples in the open water will be collected by reaching over the side of a boat and following general procedure. Samples collected at outfalls will be collected from the direct

discharge from the pipe. If the outfall is submerged, a sample will be taken as close as possible to the point of discharge and this detail will be documented on the chain of custody sheet.

All samples will be immediately placed on wet ice in a dark cooler for transport to Save the Sound's laboratory or an appointed drop-off location. The chain of custody sheet will be filled in accordingly with every switch of possession. Upon sample delivery to the lab, Save the Sound staff will note time of receipt on the laboratory data sheet (Appendix 5). The temperature of the field blank will be recorded on the laboratory data sheet upon its arrival to the laboratory, see setion 9.1 *Quality Assurance* for detailed field blank procedures.

Sample processing will be conducted in the Save the Sound laboratory following IDEXX instructions available on the IDEXX website[1] and the EPA guidance documents.[2] Maximum holding time is six hours. All samples will be incubated and scored in the Save the Sound laboratory. Lab quality controls follow the EPA guidance document and can be reviewed in section 9.1 *Quality Assurance* of this document.

12. Equipment List and Instrument Calibration

12.1 Equipment

Field:

- GPS Unit(s)
- Coolers with ice
- Sample bottles
- Powder-free nitrile gloves
- Permanent marker
- Watch

Laboratory:

- Incubator(s)
- Incubator Thermometer(s), ERTCO Alcohol
- IDEXX Sealer
- IR Thermometer
- UV Light Box
- UV Light
- Pipette
- IDEXX 120-mL sealed sterile disposable plastic bottles containing sodium thiosulfate
- IDEXX Enterolert reagent powder snap packs

[1] https://www.idexx.com/water/products/enterolert.html and https://www.idexx.com/water/products/colilert.html
[2] "GUIDANCE DOCUMENT FOR CITIZEN SCIENCE PATHOGEN MONITORING OF ENTEROCOCCI USING IDEXX ENTEROLERT WITH QUANTI-TRAY® 2000" and "GUIDANCE DOCUMENT FOR CITIZEN SCIENCE PATHOGEN MONITORING OF TOTAL COLIFORMS AND E.COLI USING IDEXX COLILERT WITH QUANTI-TRAY® 2000."

- IDEXX Colilert-18 reagent powder snap packs
- IDEXX Quanti-Tray ® 2000 incubation trays
- Sterile DI Water
- Quanti-tray holder
- IDEXX MPN tables
- *Enterococcus faecalis* QC Pellet
- *Staphylococcus aureus* QC Pellet
- *Enterobacter aerogenes* QC Pellet
- *Escherichia coli* QC Pellet
- *Pseudomonas aeruginosa* QC Pellet

12.2 Instrument Calibration and Maintenance

- Proper functioning of the incubator will be monitored by Save the Sound staff using an internal calibrated thermometer. Proper incubator temperature will be confirmed when samples are placed in the incubator and when they are removed. There will be space on the laboratory data sheet for confirming temperature in accordance with methods.
- Seals on the IDEXX Quanti-Tray ® 2000 and IDEXX sample containers will be checked before use. These materials will be stored in the Save the Sound laboratory.
- QC pellets for positive and negative controls are kept in a freezer and stored on ice before use. Time off ice is recorded on the IDEXX Quanti-Tray. All relevant information, including expiration date, for QC pellet is recorded on QC laboratory data sheet.

13. Analytical Methods

Matrix: Water

Analytical Group/ Parameter	*Enterococcus*
Reporting Limit	Fresh water: Lower limit <1 MPN/100ml, Upper limit > 24,196 MPN/100ml Brackish and salt water: lower limit <10 MPN/100ml, upper limit >24,196 MPN/100ml
Detection Limit	Fresh Water: Lower limit 1 MPN/100 ml, Upper limit 24,196 MPN/100ml Brackish and salt water: Lower limit 10 MPN/100ml, Upper limit: 24,196 MPN/100ml
Analytical & Preparation Method/SOP Reference	Guidance Document for Citizen Science Pathogen Monitoring of *Enterococcus* Using IDEXX Enterolert with Quanti-Tray ® 2000
Sample Volume	100 ml (freshwater), or 1:10 dilution (freshwater), 1:10 dilution (brackish, saltwater)
Containers	For collection and transport: 120-ml Polyethylene bottles with sodium thiosulfate For Processing: Hardy Diagnostics DI water dilution vial and Quanti-Trays ®
Preservation Requirements	Store bottle on wet ice in dark container
Max Holding Time	6 hours

Analytical Group/ Parameter	Total Coliform
Reporting Limit	Fresh water: Lower limit <1 MPN/100ml, Upper limit > 24,196 MPN/100ml Brackish and salt water: lower limit <1 MPN/100ml, upper limit >24,196 MPN/100ml
Detection Limit	Fresh Water: Lower limit 1 MPN/100 ml, Upper limit 24,196 MPN/100ml Brackish and salt water: Lower limit 1 MPN/100ml, Upper limit: 24,196 MPN/100ml
Analytical & Preparation Method/SOP Reference	Guidance Document for Citizen Science Pathogen Monitoring of Total Coliforms and *E. coli* Using IDEXX Colilert with Quanti-Tray ® 2000
Sample Volume	100 mL or 1:10 dilution (freshwater, brackish, and salt water)
Containers	For collection and transport: 120-ml Polethylene bottles with sodium thiosulfate For Processing: Hardy Diagnostics DI water dilution vial and Quanti-Trays ®
Preservation Requirements	Store bottle on wet ice in dark container
Max Holding Time	6 hours
Analytical Group/ Parameter	*E. coli*
Reporting Limit	Fresh water: Lower limit <1 *E. coli*/100ml, Upper limit >24,196 *E. coli* /100ml
Detection Limit	Fresh Water: Lower limit 1 *E. coli* /100 ml, Upper limit 24,196 *E. coli* /100ml
Analytical & Preparation Method/SOP Reference	Guidance Document for Citizen Science Pathogen Monitoring of Total Coliforms and *E. coli* Using IDEXX Colilert with Quanti-Tray ® 2000
Sample Volume	100 ml or 1:10 dilution (freshwater)
Containers	For collection and transport: 120-ml Polyethylene bottles with sodium thiosulfate For Processing: Hardy Diagnostics DI water dilution vial and Quanti-Trays ®
Preservation Requirements	Store bottle on wet ice in dark container
Max Holding Time	6 hours

14. Assessments and Oversight

Assessment Type	Frequency of Assessment	What Is Being Assessed	Who Will Conduct the Assessment	How Issues or Deviations Will Be Addressed
Sampling Protocol	Each sampling date	Sample container and transport; Sample volume; Chain of Custody Record; Sample temperature	Save the Sound Staff	Personal communication with field personnel
Laboratory Protocol	Each sampling date	All QC measures; Completeness of data sheet; Letter designations put with data	Save the Sound Staff	Correct errors, personal communication with analyst
Data Transcription	Each sampling date	Verification of data sheets against sample trays; Completeness and accuracy of online data; Accuracy of water quality rating	Save the Sound Staff	Correct errors

15. Data Management

Laboratory Data:

Original data sheets will be delivered to the Water Quality Program Manager. Original data sheets will be kept in a folder, digital copies are stored on a CFE/Save the Sound server and locally on the Water Quality Program Managers' computer. As an additional caution, a copy of the results will be stored on an external USB. The Water Quality Manager will transcribe all results into a digital spreadsheet and upload the data to the Save the Sound website. A minimum of 10% of sites will be checked online for accuracy immediately after uploading.

Field Data:

Chain of Custody sheets will be scanned and saved on Save the Sound's server and original paper copies will be stored in a folder. Save the Sound's server is backed up regularly.

16. Data Review and Usability Determination

Field/Lab	Data Management
• Check all QC sample results • Check rating errors	• Retrieve and/or document missing data • Correct errors • Compile master data spreadsheet

Deviations from established QA procedures will be discussed among the Save the Sound project staff. Data usability will be determined on a case-by-case basis. Data designations are provided in Section 9.1 *Quality Assurance* and in referenced methods for each laboratory procedure.

At the end of each year's sampling period, after all data have been entered, 3% of data lines in the master spreadsheet (cumulative going back to 2013) will be selected at random to check for data entry errors. A data line contains all data recorded for a given sampling site (date, time, *Enterococcus*/total coliform/*E.Coli* count). The values in the master spreadsheet and the Save the Sound website for each entry on the selected data lines will be verified against the field data sheets. If discrepancies are found, the data lines for the rest of the samples collected on that date will also be verified. The number of data lines per year will be checked to ensure that there are no missing data lines. Any errors will be corrected in the master spreadsheet, any summary spreadsheets, and on the Save the Sound website.

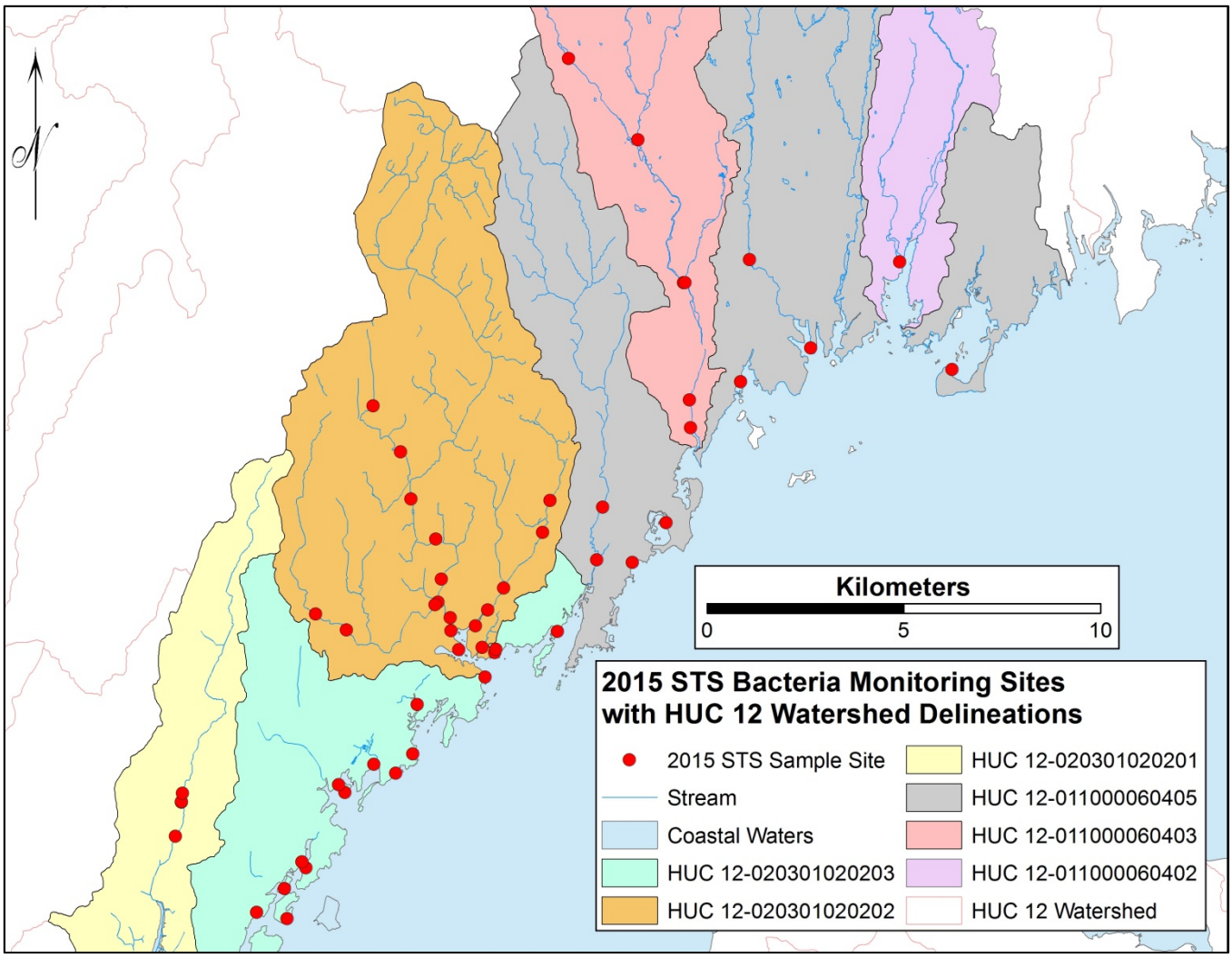
17. Reporting

Data will be uploaded to the Save the Sound website (http://www.Save the Sound.org) promptly after each sampling date.

The Water Quality Program Manager will report the results of data checks to the Director of Western Sound Programs each year after the close of the sampling season. Program staff will make data usability determinations on a case-by-case basis.

The final project report will summarize the quality assurance data check results for the entire project along with the data usability determinations made by the Director of Western Long Island Sound Programs and the Water Quality Lab Manager. The rationale for the use of any data that does not fully comply with the quality criteria requirements of the approved QAPP will be fully explained in the final report.

18. Appendix 1 – 2015 Sample Sites with HUC 12 Watersheds



19. Appendix 2 – 2015 Sample Site Information

Longitude NAD_83	Latitude NAD_83	Site_ID	Name	Town_State	Weather_Station
-73.644379	41.004652	BE-BHa	Byram Park	Greenwich, CT	Westchester County Airport_KHPN
-73.730229	40.943982	BE-MHa	Harbor Island Beach	Mamaroneck, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.677574	40.963543	BS-WCWa	Rye Playland Park	Rye, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.722365	40.937625	BS-WCWb	Beach Point Club	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.580469	41.007006	E-GCa	Greenwich Cove	Greenwich, CT	Westchester County Airport_KHPN
-73.623146	41.012254	E-GHa	Indian Harbor Yacht Club	Greenwich, CT	Westchester County Airport_KHPN
-73.744431	40.920170	E-LHa	Larchmont Harbor at Park Ave	Larchmont, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.742995	40.931492	E-LHb	Flint Park	Larchmont, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.719399	40.943202	E-MHb	Mamaroneck Harbor at Taylor Ln	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.732374	40.948046	E-MHc	Mamaroneck Harbor East Basin	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.700500	40.947919	E-MLa	Rye Marshlands Conservancy	Rye, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.777009	40.894364	E-NRHa	Neptune Boat Club	New Rochelle, NY	Town of Mamaroneck Fire Department_KNYLARCH3

-73.778148	40.895757	E-NRHb	Town Dock Road	New Rochelle, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.791937	40.884246	E-NRHc	Shore Park	Pelham, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.783517	40.889623	E-NRHd	Glen Island Approach	New Rochelle, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.667379	40.972542	E-Pla	Playland Lake at Edith Read Natural Park	Rye, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.719074	40.943973	E-VAMa	Van Amringe Millpond	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.764977	40.911504	E-WLISb	Five Islands Park on Beach	New Rochelle, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.766857	40.913331	E-WLISc	Five Islands Approach at Le Fevres Ln	New Rochelle, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.688339	40.964232	R-BB-.6	Blind Brook at Disbrow Park	Rye, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.686459	40.976252	R-BB-2.9	Blind Brook at Rye Nature Center	Rye, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.659698	40.994231	R-BR-0.55	Byram River at Columbus Park	Port Chester, NY	Westchester County Airport_KHPN
-73.659959	41.000592	R-BR-1.01	Byram River at South Water Street	Greenwich, CT	Westchester County Airport_KHPN
-73.661320	41.027339	R-BR-3.15	Byram River at Comley Avenue	Greenwich, CT	Westchester County Airport_KHPN
-73.695516	41.078955	R-BR-7.55	Byram River at Cliffdale Rd	Greenwich, CT	Westchester County Airport_KHPN

-73.674745	41.060222	R-BREB-0.20	East Branch Byram River at Riversville Road	Greenwich, CT	Westchester County Airport_KHPN
-73.721426	40.952990	R-BSB-0.06	Beaver Swamp Brook at Boston Post Road	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.716533	40.958001	R-BSB-0.46	Beaver Swamp Brook at Rye Neck HS	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.704609	40.970592	R-BSB-1.67	Beaver Swamp Brook at Truxton Street	Harrison, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.702349	40.977942	R-BSB-2.2	Beaver Swamp Brook at Greenwood Union Cemetery	Harrison, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.725219	40.949362	R-GC-0.25	Guion Creek at South Barry Ave Bridge	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.641355	41.032613	R-HNB-1.6	Horseneck Brook at Eagle Hill	Greenwich, CT	Westchester County Airport_KHPN
-73.816301	40.901772	R-HUT-3.87	Glover Field	Pelham Manor, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.814394	40.909610	R-HUT-4.40	Outfall at Farrell & Beechwood	Mount Vernon, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.814410	40.909673	R-HUT-4.42	Upstream of Farrell and Beechwood	Mount Vernon, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.814091	40.911623	R-HUT-4.55	Pelham Lake at Wilson Wood Park	Mount Vernon, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.595989	41.031742	R-MIR-1.40	Mianus River at Cos Cob Marina	Greenwich, CT	Westchester County Airport_KHPN

-73.732720	40.951330	R-MR-0.24	Mamaroneck River at Phillips Pk Rd	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.736441	40.954872	R-MR-0.61	Mamaroneck River at Station Pk Road	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.735346	40.960159	R-MR-0.76	Mamaroneck River at North Barry Ave Extended	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.736924	40.969319	R-MR-2.01	Mamaroneck River at Joint Water Works	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH3
-73.744303	40.978559	R-MR-2.66	Mamaroneck River at Saxon Woods Rd	Mamaroneck Town, NY	Town of Mamaroneck Fire Department_KNYLARCH4
-73.747314	40.989382	R-MR-3.82	Mamaroneck River at Saxon Woods Rd	White Plains, NY	Town of Mamaroneck Fire Department_KNYLARCH5
-73.755565	40.999925	R-MR-5.12	Mamaroneck River at Reynal Road	White Plains, NY	Town of Mamaroneck Fire Department_KNYLARCH6
-73.723234	40.944479	R-OC-0.22	Otter Creek at S Barry Ave Bridge	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH7
-73.661072	41.027324	R-PC-0.01	Pemberwick Creek at Pemberwick Road	Greenwich, CT	Westchester County Airport_KHPN
-73.756214	40.917898	R-PR-0.1	Premium River at Pryer Manor Road	New Rochelle, NY	Westchester County Airport_KHPN
-73.737345	40.954262	R-SHR-0.07	Sheldrake River at Columbus Park	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH7

-73.764143	40.948698	R-SHR-2.28	Sheldrake River at Bonnie Briar Ln	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH7
-73.773407	40.952388	R-SHR-2.91	Sheldrake Lake	New Rochelle, NY	Town of Mamaroneck Fire Department_KNYLARCH7
-73.749654	40.915855	S-WCWc	Larchmont Manor Park	Larchmont, NY	Town of Mamaroneck Fire Department_KNYLARCH7
-73.782798	40.882793	S-WLISa	Glen Island Park	New Rochelle, NY	Town of Mamaroneck Fire Department_KNYLARCH7
-73.725556	40.942647	BE-Mhd	Shore Acres Yacht Club	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH8
-73.720379	40.948762	R-OC-0.74	Otter Creek at Otter Creek Preserve	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH9
-73.724689	40.943232	R-OC-0.08	Otter Creek at Alda Road	Mamaroneck Village, NY	Town of Mamaroneck Fire Department_KNYLARCH10

20. Appendix 3 – Chain of Custody Record

Chain of Custody Record
545 Tompkins Ave., 3rd Floor
Mamaroneck, NY 10543

Date:

Site ID	Site Name/Location	Time Sampled

1a. Current Weather Conditions (circle): Clear Partly Cloudy Mostly Cloudy Foggy Drizzle Rain	1b. Rain in Past 24 hours (circle)? Yes No Amount in inches if known:

2. Odor(Write Station ID below)	Musty	Sewage	Chlorine	Petroleum	Decay (dead organisms)	Sulfide (rotten eggs)
3. Animals(Write Sta. ID below)	Gulls	Geese	Ducks	Possum	Cattle	Other
4. Tide at time of sampling (circle)?	Not Tidal	High	Ebb	Slack	Low	Flood
5. Other Observations (i.e. foam present, oil present, trash, include Station ID)?						

Chain of Custody Record					
Bottles relinquished from Lab by	Time	Samples relinquished by	Time	Sample received in lab by	Time
Bottles received by	Time	Sample received by	Time	Sample received in lab by	Time

21. Appendix 4 – Site Sheet with Field Methods

Save the Sound
A program of Connecticut Fund for the Environment
Guidelines for Water Quality Sampling

Supplies:

- Cooler with ice to completely cover samples
- Unused, unopened, labeled sterile sample containers
- Rubber gloves
- Permanent marker
- Labeling tape

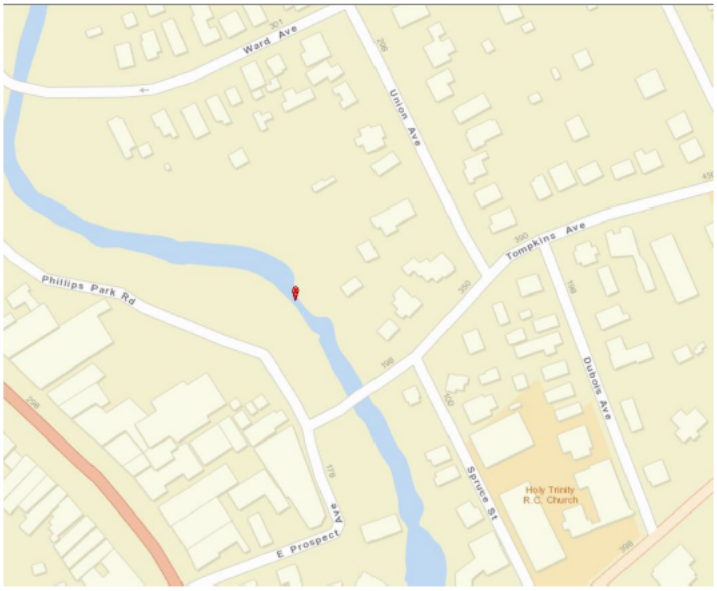
Steps for collecting samples:

1. Return to the same locations as previous events (see directions for reaching each site)
2. Write the sample time on bottle
3. Wear rubber gloves on both hands upon reaching sample site
4. If you must enter the water, sample upstream from yourself to avoid stirred-up sediment
5. Break seal on cap (NEVER TOUCH INSIDE OF CAP OR BOTTLE)
6. Submerge bottle right side up under the surface with cap on
7. Remove cap and bring bottle up out of water
8. Recap the bottle and seal it tightly
9. Immediately place the closed bottle in the cooler and cover it with ice
10. Deliver the sample directly to the drop-off point as soon as possible (see directions to reach the drop-off point)

Error sources we want to avoid:

- Collecting disturbed sediment in the sample
- Contaminating the sample with your hands
- Exposing the sample to direct sunlight or warmth
- Releasing Sodium Thiosulfate from bottle while taking sample

If you notice anything unusual or suspicious:
Such as discharge, dumping, or polluting, take photographs and call Peter Linderoth at (914) 263-6233, Jack Singer (518) 618-8016 or Tracy Brown at (914) 381-3140 before leaving the sample site.



Site Name: R-MR-0.24
Mamaroneck River near Phillips Park Road and Tompkins Avenue Intersect, Village of Mamaroneck, NY

GPS Coordinates

Long: -73.7327
Lat: 40.95138

22. Appendix 5 – Laboratory Data Sheet (*Enterococcus*)

Note: E. coli and Total Coliforms have indicator bacteria columns and QC pellet rows altered accordingly

Connecticut Fund for the Environment/Save the Sound
Western Long Island Sound Water Quality Program Laboratory Data Sheet

Project Name:			Date, Sampling Event #:				
Site ID	Site Name	Sample Time	DF	Small Well	Large Well	*Enterococcus* MPN/100 ml	Qualifier (U, J, or R)

1

Connecticut Fund for the Environment/Save the Sound
Western Long Island Sound Water Quality Program Laboratory Data Sheet

Positive Control Results Meet Acceptance Limits? Yes or No					
Incubator	Manufacturer: Serial Number:	Temp 40.5 – 41.5 °C at start of incubation? Y or N Temp 40.5 – 41.5 °C at end of incubation? Y or N		If no, specify: If no, specify:	Therm#:

Reagent/Supply	Manufacturer	Cat#	Lot #	Expiration Date	Notes
Enterolert Reagent	IDEXX				
Quanti Tray 2000	IDEXX				
Sample Bottle	IDEXX				
Sterile DI Water 90 ml	Hardy Diagnostics				
Sterile DI Water 99 ml	Hardy Diagnostics				
Enterococcus faecalis Fortified Blank QC Bacteria	Sigma Aldrich				

Test Set up and Started By:	Date/Time Test Startup Began:	Date/Time Incubated:	Results Read by (Date/Time):

6