Long Island Sound Embayments
Water Quality Monitoring QAPP

For monitoring activities conducted in the
Unified Water Study: Long Island Sound Embayment Research.

Monitoring Organizations


Coordinating Organization

Save the Sound

Funded By

The United States Environmental Protection Agency – Long Island Sound Study

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Date Approved

2021.04.20

Prior Associated Approved QAPP:

Mamaroneck Harbor and Little Neck Bay, NY, UWS Water Quality Monitoring QAPP

Approved by Kathryn Drisco, Quality Assurance Officer, EPA, Region 2, 8/3/2017

A. Project Management

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## A.4. Project / Task Organization

Table 1: Project Organization.
Key project personnel and their corresponding responsibilities.

<table>
<thead>
<tr>
<th>Name(s)</th>
<th>Project Title - Responsibility</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tracy Brown</td>
<td><em>Monitoring Program Project Manager</em> – Oversees all aspects of project that incorporate the monitoring program including: fiscal management, project objectives, data uses, program changes, etc.</td>
</tr>
<tr>
<td>Peter Linderoth</td>
<td><em>Monitoring Program Coordinator</em> – Monitoring Group recruitment and training. Develops the QAPP. Produces monitoring report. Produces or oversees outreach efforts, in coordination with project manager.</td>
</tr>
<tr>
<td>Elena Colón</td>
<td><em>Monitoring Program Field Coordinator</em> – Responsible for assistance in training and quality assurance of monitoring groups for field work. Ensures field datasheets are properly filled out, samples and forms are transported to laboratories as needed, Standard Operations Procedures (SOPs) are being followed in entirety; and performs QA checks, including field audits, to make sure procedures are followed or corrected as needed (in collaboration QA officer and UWS Science Advisors).</td>
</tr>
<tr>
<td>Peter Linderoth</td>
<td><em>Monitoring Program Lab Coordinator</em> – Makes arrangements with any lab(s) used to perform analyses according to QAPP. Ensures correct procedures are used, holding times are met, and adequate documentation is provided.</td>
</tr>
<tr>
<td>Elena Colón</td>
<td><em>Monitoring Program Data Management Coordinator</em> – Maintains the data systems for the program. Performs/oversees data entry and checks entries for accuracy against field and lab forms.</td>
</tr>
<tr>
<td>Peter Linderoth</td>
<td><em>Monitoring Program Quality Assurance Officer</em> – Runs Quality Assurance (QA) program.</td>
</tr>
<tr>
<td>Jamie Vaudrey and Jason Krumholz</td>
<td><em>UWS Science Advisors</em> – Science consultants offering guidance and participating in trainings and station selection among other aspects of the project including quality assurance.</td>
</tr>
<tr>
<td>Christopher E. Dere</td>
<td><em>USEPA Project Officer</em> – Oversees US EPA Cooperative Agreement compliance including processing recipient/subrecipient requests for QA/QC within EPA Regions</td>
</tr>
<tr>
<td>See Distribution List</td>
<td><em>UWS Monitoring Group Leads</em> – Undertake UWS in their respective embayment(s) following all aspects of this QAPP.</td>
</tr>
<tr>
<td>Esther Nelson</td>
<td><em>USEPA Quality Assurance Officer</em> – Reviews, comments and approves QAPP.</td>
</tr>
<tr>
<td><strong>Changes by year. Individual names are not listed.</strong></td>
<td><strong>Monitoring Program Field Staff</strong> – Sample, perform field analyses, and assist in laboratory analyses and/or data entry.</td>
</tr>
</tbody>
</table>


A.5. PROBLEM DEFINITION / BACKGROUND

Despite three decades of effort to improve water quality, Long Island Sound (LIS) remains a severely stressed environment. In the western Sound, from Greenwich to Nassau County, dissolved oxygen concentrations—a key measure of the Sound’s health—consistently fall to levels too low to sustain aquatic wildlife. Low levels of dissolved oxygen, or hypoxia, are worsened by excess nitrogen (N) from outdated sewage collection systems, failing septic systems, contaminated stormwater runoff, and fertilizers. Moreover, there are serious
eutrophication-related impacts in embayments throughout the Sound\(^1\). According to the EPA’s Nitrogen Reduction Strategy, “Impairments linked to excess discharges of N include harmful algal blooms, low DO, poor water clarity, loss of submerged aquatic vegetation and tidal wetlands, and coastal acidification.”

Main-stem Long Island Sound water quality data are abundant and readily available from Connecticut Department of Energy and Environmental Protection, New York City Department of Environmental Protection, and the Interstate Environmental Commission for analyses. However, there are limited environmental health data being collected in LIS embayments. The Unified Water Study (UWS) will fill in many of the data gaps that exist amongst LIS embayments. These data sets will have myriad of uses including comparing embayment environmental health, informing water quality management decisions, and conveying the information to the public so they can be better informed about the environmental health of LIS embayments.

The groups selected to participate in the 2018 UWS season went through an application process administered by the Monitoring Program Project Manager, Monitoring Program Coordinator, and the Monitoring Program Field Coordinator. A standardized application form was distributed and the administrators used a metric to select groups. In 2018, there were 19 monitoring groups monitoring a total of 33 embayments. In 2019, three new groups were added to the UWS. The 2019 group and embayment total is 22 groups in 37 embayments. 12 of the 37 embayments in the UWS will also receive Tier II monitoring. In 2020, one new group is being added to the UWS. The 2020 group and embayment total is 23 groups in 38 embayments. 13 of the embayments will receive Tier II monitoring. The 2021 group and embayment total is 24 groups monitoring 41 embayments. 13 of the embayments will receive Tier II monitoring.

The final locations of the Tier I embayments in the UWS were dependent on the applications and respective monitoring group’s interests. Tier II embayments were selected referencing priority embayment plans put forth by Connecticut Department of Energy and Environmental Protection, New York State Department of Environmental Conservation, and Long Island Sound Study. Monitoring group experience was also a factor in the decision.

**Organizational History and Mission**

The mission of Save the Sound (STS) is to protect and improve the land, air and water of Connecticut and Long Island Sound. Founded in 1978, STS merged in 2004 with Save the Sound, a respected voice for the protection of Long Island Sound’s shoreline, marine habitat and water

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quality with a track record of more than 40 years. The proposed project is in line with one of Save the Sound’s strategic goals: “Our Long Island Sound, rivers and lakes are safe for people and wildlife.”

Data collected under this QAPP will be collected in a manner to allow the data to be used as part of the Unified Water Study. The UWS is a coordinated effort among groups monitoring Long Island Sound with the goal of comparing water quality parameters associated with eutrophication within and among embayments. The UWS is comprised of two tiers, Tier I and Tier II. Tier I data is required for entry into the study. The parameters monitored in Tier I of the study are dissolved oxygen, conductivity (salinity), chlorophyll a, temperature, turbidity, and qualitative macrophyte assessments. Tier II parameters monitored as part of the UWS are nutrient concentrations, logged dissolved oxygen and conductivity (salinity) data, and underwater camera quantitative assessments of macrophytes. Tier I and Tier II parameters are covered in this QAPP.

**Monitoring History and Status**

The New York Office of Save the Sound initiated a pathogen-indicator and water quality monitoring program in 2013 and has since expanded the spatial and temporal scale of the water quality monitoring program. Save the Sound was the lead facilitator in the development of the UWS and participated in the 2016 UWS pilot season. Save the Sound continues to participate in the UWS as the coordinating organization as well as a monitoring group. Measuring the eutrophic conditions in the bays and harbors of Long Island Sound directly relates to Save the Sound’s overarching goal of reducing nitrogen and other pollutants in the Sound.

The UWS conducted a pilot season for the Tier I parameters with four existing monitoring groups in 2016. The goal of the 2016 season was to develop protocols that followed standard methods for embayment monitoring. All groups involved with the 2016 season had previous experience monitoring their embayments and were involved in ongoing monitoring programs. Groups involved with the 2016 pilot season, and other advisory participants such as academics and federal and municipal agencies, assisted with developing and finalizing the UWS Tier I SOPs.

2017 marked the inaugural season of the UWS with twelve groups participating in the study. Eleven of these groups monitored in Long Island Sound and one monitored off the south shore of Long Island, NY. These groups maintained their own QAPPs based on a template. These QAPPs provided integrity to the Study but monitoring groups in the UWS now operate under a collective EPA-approved QAPP, this document. The groups are responsible for adhering to the QAPP and Save the Sound will ensure that quality objectives are met for each embayment in the study as outlined in this document. The collective QAPP will be updated upon changes to embayments, participating groups, or procedures in the study.
Monitoring and Data Use Objectives

Data collected under this QAPP will be collected in a manner to allow the data to be used as part of the UWS. The UWS is designed to facilitate equitable water quality comparisons across Long Island Sound embayments. All data will be available to the public via request or download from Save the Sound’s website. Data will also be uploaded to the Environmental Protection Agency Water Quality Portal. Save the Sound has met, and continues to meet and discuss, UWS data usage by Connecticut Department of Energy and Environmental Protection, New York State Department of Environmental Conservation, and UWS monitoring groups are welcome and encouraged to share their data on a local level to potentially elicit changes that could improve water quality in their respective embayments. There is also an overarching primary project goal to include the UWS data sets in the Long Island Sound Report Card which is issued by Save the Sound. The report card compares water quality indicators (dissolved oxygen, nutrients, chlorophyll a, and water clarity) to scientifically derived thresholds or goals. These indicators are combined into an overarching Water Quality Index, which is presented as a subregion percent score. The report card provides a geographically specific assessment of annual Long Island Sound ecosystem health.

Additionally, the activities covered under this QAPP will provide quality-controlled data that can support secondary goals such as the assessment and restoration of coastal embayments and watersheds through the implementation of programs such as but not limited to:

- EPA’s 305(b) water body health assessments and 303(d) TMDL development for impaired waters
- Clean Water Act Section 319 projects
- Connecticut Department of Energy and Environmental Protection and New York Department of Environmental Conservation Watershed Management Plans
- Long Island Sound Study’s Comprehensive Conservation and Management Plan
- Long Island Sound Study Environmental Indicators Project
- New York State Department of Environmental Conservation Long Island Nitrogen Action Plan

A.6. PROJECT / TASK DESCRIPTION

Five types of monitoring stations are included:

1) **Tier I water quality** stations are sampled within three hours of sunrise between the months of May through October using a multiparameter sonde. A minimum of four
stations per embayment are required. If multiple regions of the embayment are
delineated, a minimum of three stations are required per region. Monitoring groups will
plan to sample Tier I water quality stations at a minimum every two weeks from May to
October. However, if unforeseeable circumstances make this plan not possible, a
minimum of six sample events between May and October are required for inclusion in the
UWS; including at least one sample event in each of the months of June, July, August, and
September. The number and location of Tier I water quality stations for embayments in
the UWS are provided in Section A.6.b.

**Tier I Water Quality** – Water quality parameters are selected to facilitate comparisons between
embayments. Monitoring groups must collect the following data to be included in the UWS:

- for each station
  - GPS coordinates of stations, recorded each sample date
  - Date and time
  - Total water depth
    - 0.5 m below the surface, 0.5 m above bottom, mid-depth if total depth >10 m; if
total depth is less than 1.5 m, only a mid-depth reading will be collected
    - Temperature
    - Conductivity (salinity)
    - Dissolved oxygen
    - Chlorophyll a
    - Turbidity
  - Once per field day undertake a replicate profile including all parameters
- Obtain from an online NOAA tide table and weather station approved by Monitoring
  Program Coordinator:
  - Time of high and low tide nearest time of sampling
  - High and low air temperature for 24 hours preceding field sampling
  - Precipitation out a week preceding sampling event
- Within 1 day of the field sampling day, read the GPS of a land-based reference station

Monitoring groups will collect water quality data for the Unified Water Study according to
procedures provided in the UWS SOP Depth and GPS, UWS SOP Sonde Profile, and UWS SOP
Filtered Chlorophyll in Appendix A.

2) **Qualitative Tier I Macrophyte** stations are land-based or boat-based. They are sampled
only mid-summer and may be sampled on different days from the water quality stations.
Sampling occurs on three separate days between July 15 and August 7. Two sample days
or a date slightly outside of the date criteria may be sufficient in the event of unforeseen
complications. This decision will be made by the Monitoring Program Quality Assurance
Officer and UWS Science Advisor(s).
The goal of this part of the UWS is to identify potential problem areas, versus characterizing the overall condition of the embayment; field teams will look for areas with the highest macrophyte abundance they can find. Groups will also look for and note the presence of eelgrass (a beneficial condition). This is not intended to be a quantitative assessment. Macrophyte surveys will complement chlorophyll a concentrations to better understand the dominant primary producer in the system.

**Qualitative Tier I Macrophytes** - Monitoring groups must collect the following data to be included in the UWS:

- for each macrophyte station
  - GPS coordinates of stations, recorded each sampling date
  - Date and time
  - Photos of macrophytes
- Within 1 day of the field sampling day, read the GPS of a land-based reference station

Groups will submit data and photos to the Unified Water Study according to methods provided in the UWS SOP Qualitative Macrophytes in Appendix A.

3) **Quantitative Tier II Macrophyte** stations are boat-based. They are sampled only mid-summer and are typically sampled on different days from the water quality stations. Sampling occurs on one day between July 15 and August 7.

**Quantitative Tier II Macrophytes** - Monitoring groups must collect the following data to be included in the UWS:

- For each macrophyte station
  - GPS coordinates
  - Date and time interval
  - Video of macrophyte abundance
- Within 1 day of the field sampling day, read the GPS of a land-based reference station

Groups will submit data and photos to the Unified Water Study according to methods provided in the UWS SOP Macrophyte Percent Coverage Via Camera in Appendix A.

4) **Tier II nutrients** stations are sampled a minimum every two weeks from May to October. However, if unforeseeable circumstances make this plan not possible, a minimum of six sample events between May and October are required for inclusion in the UWS; including at least one sample event in each of the months of June, July, August, and September. At a minimum, there will be two stations per embayment. In embayments with multiple regions, there will be a minimum of two stations per region. The number and location of Tier II nutrients stations for embayments in the UWS are provided in Section A.6.b. Stations in tributaries are sampled for nutrients on the same day as the embayment they flow into is sampled. These stations are chosen with respect to access and salinity value <1 ppt. A Long Island Sound reference station will also be sampled for nutrients on the
same day the embayment stations are sampled.

**Tier II Nutrients** - Monitoring groups must collect the following data to be included in the UWS:
- For each water quality station
  - GPS coordinates of stations, recorded each sample date
  - Date and time
  - 0.5 m below the surface
    - Total Nitrogen
    - Total Dissolved Nitrogen
    - Dissolved Inorganic Nitrogen Species (nitrate, nitrite, ammonia)
    - Total Phosphorous
    - Orthophosphate (also known as dissolved inorganic phosphorus)
    - Salinity

Groups will submit data to the Unified Water Study according to methods provided in the UWS SOP Filtered Nutrients and UWS SOP Total Nitrogen and Total Phosphorous in Appendix A.

5) **Continuous** Dissolved Oxygen stations will be selected with consideration to representativeness and where access is granted for maintenance of the equipment. A minimum of one continuous dissolved oxygen station is required per embayment region. Logging will commence from May to October 31.

**Continuous Dissolved Oxygen** - Monitoring groups must collect the following data to be included in the UWS:
- for each continuous dissolved oxygen station
  - GPS coordinates of stations, recorded each sampling date
  - Date and time
  - Dissolved Oxygen
  - Conductivity (Salinity)
  - Barometric Pressure
  - Temperature

Groups will submit data to the Unified Water Study according to methods provided in the UWS SOP Continuous Dissolved Oxygen in Appendix A.

**Roles of Project Participants**

The Monitoring Program Coordinator and Monitoring Program Field Coordinator or designee will provide guidance and advisement to the groups participating in the UWS, conferring with the UWS Science Advisors as needed. They will conduct trainings, field audits, station selection guidance, ongoing technical support, and lab coordination among other activities. The full set of participants and their respective roles can be referenced in Table 1 of this document. Figure 1 outlines the lines of communications between project participants.
UWS Project Laboratory Manager has agreed to the UWS SOP Filtered Chlorophyll, UWS SOP Total Nitrogen and Total Phosphorous, and UWS SOP Filtered Nutrients for sample collection in the field. The project laboratory has provided their laboratory method SOPs which can be reviewed in Appendix C of this QAPP. The laboratory will adhere to both the UWS and their lab SOPs.

Participating Monitoring Groups will be responsible for conducting field work and analyses following the requirements presented in the UWS SOPs. Monitoring Group Leads or designated appointees will complete all required training. Monitoring Group members will complete all required data sheets and chain of custody forms. Any problems or deviance from this QAPP or SOPs will immediately be reported to the Monitoring Program Field Coordinator who will confer with the Monitoring Program Quality Assurance Officer on corrective course of action.

**How the proposed sampling plan supports the Monitoring Program objectives**

Data collected under this QAPP will be collected in a manner to allow the data to be used as part of the UWS. The UWS is a coordinated effort among groups monitoring Long Island Sound embayments with the goal of comparing water quality and macrophyte abundance within and among embayments.

**Overview of data handling processes**

Sampling event and field data will be collected on standardized field and instrument calibration sheets. These standardized datasheets are in Appendix B of this document.

If a field team is delivering samples to a centralized location for laboratory analysis by a member of the Monitoring Group, the field data sheet is sufficient as a chain of custody record. In this scenario a chain of custody form will not be required as sufficient information is contained on the sample event datasheet.

If a field team is delivering a sample for analysis by a lab external to the monitoring group, the UWS Chlorophyll a Chain of Custody Form or UWS Nutrient Chain of Custody Form is required. These forms are in Appendix B of this document.

**A.6.a. Sampling Types Covered by this QAPP**

The type of sample information that can be collected under this QAPP includes:

- GPS location to identify and track station locations
- Total water depth of the sample station; and depth of sample location
- Temperature
- Conductivity (Salinity)
- Dissolved oxygen concentration and percent saturation
- Chlorophyll a concentrations (filtered water sample)
- Chlorophyll a concentrations (*in situ* fluorescence)
- Turbidity
- Qualitative assessment of macrophytes
- Quantitative assessments of macrophytes
- Nitrogen forms to measure nutrient levels
- Phosphorous forms to measure nutrient levels

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A.6.b. Maps of Study Area

Figure 2a-z. All UWS study sites. Reporting regions if present are clearly delineated. Red circles are water quality station locations. The maps are overlain by a hexagonal grid. The grid was used to select stations to represent the water quality of the entire embayment using a probability-based sampling design\(^2\), as in the EPA National Coastal Assessment\(^3\). In some cases, hexagons have been joined to represent a local area considered similar or if a hexagon included large sections of land.

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Figure 2a
Figure 2b
Figure 2d
Figure 2g
Figure 21
Figure 2m
Figure 2p
Figure 2q
Figure 2r
Figure 2s
Figure 2u

UWS - Mattituck Creek, NY
North Fork Environmental Council Sample Stations

- UWS Tier I Sample Station
- Hexagon Grid

Kilometers

1.5
Figure 2v
Figure 2w
Figure 2x
Figure 2y
Figure 2z
Figure 2ad

UWS - Scotts Cove, CT
Town of Darien Sample Stations
- UWS Sample Station
- Hexagon Grid

Kilometers
0.5
Figure 2ad
Table 2: Station coordinates in NAD_83 for Tier I water quality and Tier II nutrients stations* in the UWS.

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*These stations will be sampled for Tier II nutrients.

A.6.c. Annual Task Calendar

The annual task calendar describes when certain activities will occur.
Table 3: Annual Task Calendar
These tasks are repeated annually.

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<td>Data uploads to STS-UWS website (must follow data review)</td>
<td></td>
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<tr>
<td>Draft report</td>
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<td>o</td>
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</tr>
<tr>
<td>Final annual report</td>
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<td>o</td>
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</tr>
</tbody>
</table>

o indicates the year following sampling events

A.7. DATA QUALITY OBJECTIVES

Taken together, precision, accuracy and bias, representativeness, comparability, completeness, and sensitivity comprise the major data quality indicators used to assess the quality of the program’s data. A summary of criteria are provided in Table 4.

Definitions of these data quality indicator terms:

- **Precision** is the degree of agreement among repeated field measurements of the same indicator and gives information about the consistency of methods. It is typically defined as relative percent difference, or RPD.
• **Accuracy** is a measure of confidence that describes how close a measurement is to its “true” or expected value; it includes a combination of random error (precision) and systematic error (bias) components of both sampling and analytical operations.

• **Bias** is the systematic or persistent distortion of a measurement process that causes errors in one direction.

• **Representativeness** is the extent to which measurements actually represent the true environmental condition. Parameters, station selection (including location of sampling point within the water column), time, and frequency of sample collection can all play a role in determining how representative a sample is.

• **Comparability** is the extent to which data can be compared between sample locations or periods of time within a project, or between different sites.

• **Completeness** is the comparison between the amount of valid or usable data the program originally intended to collect versus how much was actually collected.

• **Sensitivity** is the capability of a method or instrument to discriminate between measurement responses representing different levels of the variable of interest.

### Table 4: Measurement Performance Criteria

<table>
<thead>
<tr>
<th>Data Quality Indicators</th>
<th>Measurement Performance Criteria</th>
<th>QC Sample and/or Activity Used to Assess Criteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Precision – overall</td>
<td>RPD ≤ value indicated in Table 5</td>
<td>field duplicates</td>
</tr>
<tr>
<td>Precision – analytical</td>
<td>RPD ≤ value indicated in Table 5</td>
<td>analytical duplicates</td>
</tr>
<tr>
<td>Accuracy / Bias</td>
<td>85% ≤ recovery ≤ 115%</td>
<td>certified reference material</td>
</tr>
<tr>
<td>Comparability</td>
<td>standard procedures followed</td>
<td>NA</td>
</tr>
<tr>
<td>Completeness</td>
<td>data from surface, mid (if applicable) and bottom at each station meet data quality objectives</td>
<td>data completeness check</td>
</tr>
<tr>
<td>Sensitivity</td>
<td>value ≥ MDL*</td>
<td>sample value check</td>
</tr>
</tbody>
</table>

* MDL = method detection limit. This is a reporting limit based on the lowest standard accurately analyzed in the analysis.

**Precision** - Precision objectives are listed in Table 5. Precision is evaluated in the field by participants taking replicate measurements for at least 5% of samples, where applicable.

For UWS Tier I water quality sample events, a replicate profile at one station per field day.
For water samples filtered and analyzed for extracted chlorophyll-\(a\), two field replicates will be collected per sample day.

For estuary and tributary stations sampled for nutrients, a field replicate will be collected at each station.

For qualitative macrophyte stations, multiple pictures will be taken for rake toss sampling and beach sampling.

For quantitative macrophyte sampling, multiple analyses of the stills to determine percent coverage will be undertaken.

When a multiparameter sonde is used, standards will be read before and following a trip, within one day of the field day. Calibration of sondes will happen within one day of the sampling event. Multiparameter sondes can hold their calibrations for weeks. The pre and post sampling event readings can identify any potential drift outside of manufacturer recommendations for calibration. These values will be kept with all other data for review at the end of the project interval.

The Onset HOBO loggers (dissolved oxygen) and Star-Oddi loggers (temperature, conductivity, depth) will be deployed in a common water bath before deployment and following deployment. Conductivity, temperature and oxygen will be varied in the bath, allowing for multiple values for intercomparison. The temperature, conductivity, and oxygen of the bath will be determined with the instruments being used for conducting Tier I water quality profiles. These pre- and post-baths will serve to cross-calibrate all instruments and to determine if the deployed loggers exhibited any drift over the course of the deployment. The deployed loggers will be intercalibrated by applying a multiplicative correction if initial values differ by more than 10% from the reference value (as determined from the YSI EXO1 sonde or Eureka Manta +35).

The frequency of field replicate measurements for each parameter are described in Table 7.

Relative percent difference (RPD) of replicate samples is used as one index of precision; see Table 5. This is defined as the absolute difference between the replicates divided by the average of the replicates. The allowable RPDs for each parameter are provided in Table 7. A difference greater than the designated RPD requires further investigation of the sample run. If the difference is large enough, it indicates failure (unless the average of the two samples is less than 10 times the method detection limit), and results in potential disqualification or flagging of data from that station depth, unless there is a reasonable and supported explanation for the inconsistency. Replicate precision will be analyzed by calculating the RPD using the equation:

\[
\text{RPD (\%)} = \frac{|x_1 - x_2|}{(x_1 + x_2)/2} \times 100
\]

where \(x_1\) is the original sample concentration and \(x_2\) is the replicate sample concentration.
The Microsoft Excel formula for calculating the RPD is:

\[ \text{RPD} = \frac{\text{ABS}(X1 - X2)}{\left(\frac{X1 + X2}{2}\right)} \times 100 \]

where \( X1 \) is the original sample concentration and \( X2 \) is the replicate sample concentration. The RPD is automatically calculated in the UWS data entry template for replicate profiles and field samples.

**Accuracy and Bias** - Accuracy objectives are listed in Table 4. Procedures used to test or ensure accuracy are described in Table 11. While training and audits help to ensure measurement accuracy and precision, quantitative measures of accuracy for water quality monitoring are estimated using laboratory QC data (blank results, fortified matrix results, known QC samples, etc.). When a multiparameter sonde is used, standards will be read before and following a trip, within one day of the field day. Extracted chlorophyll a analysis will include a field replicate, laboratory blank and reference standards. Nutrient analysis will include a laboratory blank, field blank, and reference standards. Data loggers will be calibrated prior to and after deployment. Biweekly comparative readings between loggers and sonde will be recorded to keep a log of any drift occurring with loggers. These data will be evaluated with the log data in the final report and during the season.

**Representativeness** – Tier I water quality sample stations and quantitative Tier II macrophyte stations are selected to represent the entire embayment using a probability-based sampling design\(^4\), as in the EPA National Coastal Assessment\(^5\). In this approach, a 0.42 km\(^2\) hexagonal grid is overlain on the site map. Three random stations are generated in ArcGIS in each section of the embayment within a hexagon. Large embayments will have hexagons for random station generation selected with the UWS Science Advisors. A minimum of four stations will be sampled in each embayment, with larger sites having up to twelve stations. The location of the station in each hexagon will be randomly generated, with at least two alternate locations also randomly generated, in case the original location is deemed unusable (e.g. too shallow or in the middle of a navigation channel). If none of the three random stations are accessible, a station will be determined as close as possible to a randomly generated station unless bias circumstances are identified. The Monitoring Program Coordinator will oversee station selection, providing GIS-based maps and station coordinates to groups participating in the Unified Water Study. At least one Scientific Advisor affiliated with the UWS will also advise on

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the choice of station locations. Stations used previously by a group are evaluated for bias before inclusion in the UWS.

Tier II nutrient stations are a subset of the Tier I water quality stations selected in consultation with the UWS Science Advisors. A pilot test of nutrient sampling was conducted in Mamaroneck River, NY and Little Neck Bay, NY in 2017. In this pilot test, all Tier I water quality stations were sampled for nutrients. The approach to selecting a subset of stations (2-3) for Tier II nutrient analysis to yield a regional average was compared to the regional average using all stations in a region (3-5). The approach described below yields the most accurate regional average when sampling a subset of Tier I water quality stations for nutrients.

At a minimum, there will be two stations per embayment. In embayments with multiple regions, there will be a minimum of two stations per region. These stations are located at the boundaries of region delineations or the entire embayment, when regions are not identified.

Embayment size, salinity, and best judgment of the UWS Science Advisors determine if more stations may be needed in a region. Additional stations are required when the geometry of the embayment is not linear. For example, the Y shape of Mamaroneck River, NY requires three stations as seen in this QAPP. Salinity within a region that has a range greater than 2 ppt triggers a closer inspection of available salinity data by the UWS Science Advisors, to insure selected stations are sufficient to characterize the region.

Stations that have salinity data of less than 5 ppt are not acceptable as representative embayment stations. In embayments with the minimum Tier I water quality stations (4 stations), a station where salinity is episodically below 5 ppt may be deemed acceptable if the UWS Science Advisors deem inclusion of the station as important to estimating the regional average. The next station downstream is evaluated for inclusion in cases where a station is rejected.

Tier II nutrients stations (tributary and Long Island Sound reference) are chosen based on access, location, and salinity values under 1 PPT.

Data logging stations are selected with at least one station per reporting region. Access to station is a strong consideration in the selection process. The final station locations will be conferred with UWS Science Advisors before deployment.

Qualitative macrophyte surveys are targeted qualitative assessments of areas in the embayment known to harbor macrophytes thus the random station generation does not apply to selecting these locations. Sample collection timing and frequency for water quality stations are selected to capture data that are representative of embayment conditions. While tidal stage will vary among sampling dates, the timing relative to dawn was considered of greatest importance when sampling Tier I water quality stations to evaluate hypoxia in embayments. These very shallow systems are typically dominated by benthic primary producers (macroalgae, benthic microalgae, and seagrass) versus pelagic primary producers (pelagic microalgae /
phytoplankton). When the sun rises, these primary producers quickly replenish the dissolved oxygen in the water column. One of the goals of this study is to evaluate the incidence of hypoxia in embayments, thus sampling close to dawn is more important than sampling at a specific tidal stage. Time of high and low tide and precipitation volumes are recorded and will be considered in the analysis of results. Any abnormal or episodic conditions that may affect the representativeness of sample data are noted and maintained as metadata.

**Comparability** - The comparability of the data collected can be assured by using known protocols and documenting methods, analysis, sampling sites and stations, times and dates, sample storage and transfer, as well as laboratories and identification specialists; so that future surveys can produce comparable data by following similar procedures. Examples of project procedures are available in the collection of Standard Operating Procedures (SOPs) provided in Appendix A of this document.

**Completeness** – Minimum sample events for inclusion for Tier I and Tier II monitoring are included in section A.6 of this document.

**Sensitivity** – Sensitivity objectives are listed in Table 5. Sensitivity is the lowest detection limit of the method or instrument for each of the measurement parameters of interest. For analytical methods, these are the method detection limits (MDLs).

**Table 5: Data Quality Objectives**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Units</th>
<th>Accuracy</th>
<th>Precision (allowable RPD)</th>
<th>Approx. Expected Range</th>
<th>Sensitivity (Resolution or MDL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Depth (calibrated line)</td>
<td>meters (m)± 0.1 m</td>
<td>20%</td>
<td>0 – 50 m</td>
<td>0.1 m</td>
<td></td>
</tr>
<tr>
<td>Barometric Pressure (ONSET HOBO U20L-01)</td>
<td>Kilopascal (kPa)</td>
<td>0.62 kPa maximum error</td>
<td>10%</td>
<td>3.7 – 4.1 kPa</td>
<td>&lt; 0.02 kPa</td>
</tr>
<tr>
<td>Depth (YSI EXO 1)</td>
<td>meters (m)</td>
<td>0 to 10 m ± 0.04% FS or ± 0.004 m</td>
<td>20%</td>
<td>0 – 50 m</td>
<td>0.001 m</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0 to 100 m ± 0.04% FS or ± 0.04 m</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>0 to 820 m ± 0.04% FS or ± 0.1 m</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Parameter</td>
<td>Units</td>
<td>Accuracy</td>
<td>Precision (allowable RPD)</td>
<td>Approx. Expected Range</td>
<td>Sensitivity (Resolution or MDL)</td>
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<tr>
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<td>----------------------------------</td>
</tr>
<tr>
<td>Depth (Eureka Manta +35)</td>
<td>meters (m)</td>
<td>0 to 10 m ±0.02 (±0.2% of FS)</td>
<td></td>
<td>0 – 50 m</td>
<td>0.01 m</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0 to 25 m ±0.05 (±0.2% of FS)</td>
<td>20%</td>
<td></td>
<td>0.1 m</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0 to 50 m ±0.1 (±0.2% of FS)</td>
<td></td>
<td></td>
<td>0.1 m</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0 to 100 m ±0.2 (±0.2% of FS)</td>
<td></td>
<td></td>
<td>0.1 m</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0 to 200 m ±0.4 (±0.2% of FS)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>GPS coordinates</td>
<td>decimal degrees (dec. deg.)</td>
<td>± 7.8 m [<a href="http://www.gps.gov/systems/gps/performance/accuracy/">http://www.gps.gov/systems/gps/performance/accuracy/</a>] for reference point on land, within 10 m (=0.0001 dec. deg.)</td>
<td>NA</td>
<td></td>
<td>1.02 m</td>
</tr>
<tr>
<td>Temperature (YSI EXO 1)</td>
<td>degrees Celsius (°C)</td>
<td>-5 to 35 °C ± 0.5 °C 35 to 50 °C ± 0.05 °C</td>
<td>10%</td>
<td>4 – 26 °C</td>
<td>0.001 °C</td>
</tr>
<tr>
<td>Temperature (Eureka Manta +35)</td>
<td>degrees Celsius (°C)</td>
<td>± 0.1 °C</td>
<td>10%</td>
<td>4 – 26 °C</td>
<td>0.01 °C</td>
</tr>
<tr>
<td>Conductivity (YSI EXO 1)</td>
<td>millisiemens (mS/cm)</td>
<td>0 to 100 mS/cm ± 5% of reading or 0.0001 mS/cm; whichever is greater 100 to 200 mS/cm ± 5% of reading</td>
<td>10%</td>
<td>0 – 50 mS/cm</td>
<td>0.0001 to 0.001 mS/cm, range-dependent</td>
</tr>
<tr>
<td>Conductivity (Star-Oddi DST CT)</td>
<td>millisiemens (mS/cm)</td>
<td>13-50 mS/cm ± 1.5 mS/cm</td>
<td>10%</td>
<td>13-50 mS/cm</td>
<td>0.01 mS/cm within range</td>
</tr>
<tr>
<td>Specific Conductance (Eureka Manta +35)</td>
<td>millisiemens (mS/cm)</td>
<td>0 to 10 mS/cm ± 1% of reading or ± 0.001 mS/cm 10 to 100 mS/cm or ± 1% of reading</td>
<td>10%</td>
<td>0 – 50 mS/cm</td>
<td>0.001 mS/cm 0.01 mS/cm</td>
</tr>
<tr>
<td>Parameter</td>
<td>Units</td>
<td>Accuracy</td>
<td>Precision (allowable RPD)</td>
<td>Approx. Expected Range</td>
<td>Sensitivity (Resolution or MDL)</td>
</tr>
<tr>
<td>-----------</td>
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</tr>
<tr>
<td>Dissolved oxygen (YSI EXO 1)</td>
<td>milligrams per liter (mg/L) = parts per million (ppm); percent saturation (% sat.)</td>
<td>0 to 20 mg/l ± 1% of reading or 0.1 mg/L 20 to 50 mg/l ± 5% of reading 0 to 200% ± 1% reading or 1% air saturation, whichever is greater 200 to 500% ± 5% reading</td>
<td>20%</td>
<td>0 – 14 mg/L 0 – 120 %</td>
<td>0.01 mg/L 0.1 % sat.</td>
</tr>
<tr>
<td>Dissolved oxygen (Eureka Manta +35)</td>
<td>milligrams per liter (mg/L) = parts per million (ppm); percent saturation (% sat.)</td>
<td>0 to 20 mg/l ± 0.2 mg/l 20 to 50 mg/l ± 10% of reading 0 to 200% sat. ±1% of reading or ±0.1 % sat. 200 to 500% sat. ±10% of reading</td>
<td>20%</td>
<td>0 – 14 mg/L 0 – 120 % sat.</td>
<td>0.1 mg/l 0.1 % sat.</td>
</tr>
<tr>
<td>Dissolved oxygen (ONSET HOBO U26)</td>
<td>milligrams per liter (mg/L) = parts per million (ppm)</td>
<td>0 to 8 mg/l ± 0.2 mg/l 8 to 20 mg/l ± 0.5 mg/l</td>
<td>20%</td>
<td>0 – 14 mg/L</td>
<td>0.02 mg/l</td>
</tr>
<tr>
<td>Chlorophyll a (as measured in lab)</td>
<td>microgram per liter (µg/L)</td>
<td>75 - 125 % recovery of a lab QC sample with known µg/L</td>
<td>15%</td>
<td>0 – 30 µg/L; though higher concentrations may occur</td>
<td>0.7 µg/L</td>
</tr>
<tr>
<td>Chlorophyll a (YSI EXO 1)</td>
<td>Relative Fluorescence Units (RFU), microgram per liter (µg/L)</td>
<td>Chl: R² &gt; 0.999 for serial dilution of Rhodamine WT Solution from 0 to 400 µg/L PC equivalents</td>
<td>20%</td>
<td>0 – 30 µg/L; though higher concentrations may occur</td>
<td>0.01 RFU 0.01 µg/L</td>
</tr>
<tr>
<td>Chlorophyll a (Eureka Manta +35)</td>
<td>microgram per liter (µg/L)</td>
<td>0.03 to 500 µg/L ± 3% of full scale</td>
<td>20%</td>
<td>0 – 30 µg/L; though higher concentrations may occur</td>
<td>0.01 µg/L</td>
</tr>
<tr>
<td>Parameter</td>
<td>Units</td>
<td>Accuracy</td>
<td>Precision (allowable RPD)</td>
<td>Approx. Expected Range</td>
<td>Sensitivity (Resolution or MDL)</td>
</tr>
<tr>
<td>-----------------------------------------------------</td>
<td>-------</td>
<td>--------------------------------------------------------------------------</td>
<td>---------------------------</td>
<td>------------------------</td>
<td>---------------------------------</td>
</tr>
<tr>
<td>Turbidity (YSI EXO 1)</td>
<td>FNU*</td>
<td>0 to 999 FNU ± 2% of reading or 0.3 FNU, whichever is greater</td>
<td>20%</td>
<td>0 – 30 FNU</td>
<td>0 – 999 FNU: 0.01 FNU</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1000 to 4000 FNU ± 0.1 FNU</td>
<td></td>
<td></td>
<td>1000 – 9999 FNU: 0.1 FNU</td>
</tr>
<tr>
<td>Turbidity (Eureka Manta +35)</td>
<td>NTU</td>
<td>0 to 400 NTU ± 1% of reading ± 1 count</td>
<td>20%</td>
<td>0 – 30 NTU</td>
<td>4 digits</td>
</tr>
<tr>
<td></td>
<td></td>
<td>400 to 3000 NTU ± 3% of reading</td>
<td></td>
<td></td>
<td>4 digits</td>
</tr>
<tr>
<td>Dissolved ammonia - NH₃</td>
<td>mg/L NH₃ (= ppm = g/m³)</td>
<td>85% - 115% recovery of lab fortified matrix (LFM)</td>
<td>Field Replicate 30%</td>
<td>0-1 mg/l</td>
<td>0.020 mg/l</td>
</tr>
<tr>
<td>or (NO₂⁻) (as measured in lab)</td>
<td></td>
<td></td>
<td>Analytical Replicate 15%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dissolved nitrate⁴ - NO₃ (NO₃⁻ NO₂⁻)</td>
<td>mg/L NO₃ (= ppm = g/m³)</td>
<td>Value calculated from multiple N analyses</td>
<td>NA</td>
<td>0-2 mg/l</td>
<td>NA</td>
</tr>
<tr>
<td>or (as measured in lab)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dissolved nitrate - NO₂⁻ (as measured in lab)</td>
<td>mg/L NO₂ (= ppm = g/m³)</td>
<td>85% - 115% recovery of lab fortified matrix (LFM)</td>
<td>Field Replicate 30%</td>
<td>0-0.7 mg/l</td>
<td>0.004 mg/l</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Analytical Replicate 15%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nitrate-nitrite – NOX or NO₃ + NO₂ (as measured in lab)</td>
<td>mg/L NOx (= ppm = g/m³)</td>
<td>85% - 115% recovery of lab fortified matrix (LFM)</td>
<td>Field Replicate 30%</td>
<td>0-2.5 mg/l</td>
<td>0.004 mg/l</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Analytical Replicate 15%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dissolved inorganic nitrogen¹ – DIN (NH₃+NOₓ)</td>
<td>mg/L DIN (= ppm = g/m³)</td>
<td>Value calculated from multiple N analyses</td>
<td>NA</td>
<td>0-4 mg/l</td>
<td>NA</td>
</tr>
<tr>
<td>Total dissolved nitrogen – TDN (as measured in lab)</td>
<td>Mg/l TDN (= ppm = g/m³)</td>
<td>85% - 115% recovery of lab fortified matrix (LFM)</td>
<td>Field Replicate 30%</td>
<td>0-5 mg/l</td>
<td>0.05 mg/l</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Analytical Replicate 15%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total Nitrogen (as measured in lab)</td>
<td>mg/L TDN (= ppm = g/m³)</td>
<td>85% - 115% recovery of lab fortified matrix (LFM)</td>
<td>Field Replicate 30%</td>
<td>0-8 mg/l</td>
<td>0.05 mg/l</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Analytical Replicate 15%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total inorganic nitrogen¹ – TIN (NH₃+NOₓ)</td>
<td>mg/L TIN (= ppm = g/m³)</td>
<td>Value calculated from multiple N analyses</td>
<td>NA</td>
<td>0-4 mg/l</td>
<td>NA</td>
</tr>
<tr>
<td>Parameter</td>
<td>Units</td>
<td>Accuracy</td>
<td>Precision (allowable RPD)</td>
<td>Approx. Expected Range</td>
<td>Sensitivity (Resolution or MDL)</td>
</tr>
<tr>
<td>------------------------------------------------</td>
<td>-------------------------------</td>
<td>--------------------------------------------------------------------------</td>
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<td>------------------------</td>
<td>---------------------------------</td>
</tr>
<tr>
<td>Total organic nitrogen(^\d) – TON (TN - TIN)</td>
<td>mg/L TON (= ppm = g/m(^3))</td>
<td>value calculated from multiple N analyses</td>
<td>NA</td>
<td>0-5 mg/l</td>
<td>NA</td>
</tr>
<tr>
<td>Dissolved organic nitrogen(^\d) - DON (TDN - DIN)</td>
<td>mg/L DON (= ppm = g/m(^3))</td>
<td>value calculated from multiple N analyses</td>
<td>NA</td>
<td>0-4.5 mg/l</td>
<td>NA</td>
</tr>
<tr>
<td>Particulate nitrogen(^\d) – PN (TN-TDN)</td>
<td>mg/L PN (= ppm = g/m(^3))</td>
<td>value calculated from multiple N analyses</td>
<td>NA</td>
<td>0-0.5 mg/l</td>
<td>NA</td>
</tr>
<tr>
<td>Total phosphorus – TP (as measured in lab)</td>
<td>mg/L TP (= ppm = g/m(^3))</td>
<td>85% - 115% recovery of lab fortified matrix (LFM)</td>
<td>Field Replicate 30%</td>
<td>0-0.5 mg/l</td>
<td>0.334 mg/l</td>
</tr>
<tr>
<td>Dissolved organic nitrogen(^\d) - DON (TDN - DIN)</td>
<td>mg/L DON (= ppm = g/m(^3))</td>
<td>value calculated from multiple N analyses</td>
<td>NA</td>
<td>0-4.5 mg/l</td>
<td>NA</td>
</tr>
<tr>
<td>Dissolved orthophosphate – PO(_4^\text{3-}) or DIP (as measured in lab)</td>
<td>mg/L PO(_4^\text{3-}) mg/L DIP (= ppm = g/m(^3))</td>
<td>85% - 115% recovery of lab fortified matrix (LFM)</td>
<td>Field Replicate 30%</td>
<td>0-0.3 mg/l</td>
<td>0.001 mg/l</td>
</tr>
<tr>
<td>Quantitative macrophyte amount</td>
<td>% coverage bare, macrophytes, and animals of bottom</td>
<td>Estimates from three analyses are compared. If the relative percent difference among the three estimates is greater than 5%, the Monitoring Group Lead examines the image and the three estimates, choosing the appropriate value. The three estimates will not be changed, values are retained to show the inconsistency. The Monitoring Group Lead decides on the final value for the estimate.</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Qualitative macrophyte amount</td>
<td>choice of: none, some, lots</td>
<td>This is a qualitative assessment, not quantitative. Photos are reviewed by a UWS Science Advisor or trained designee to confirm choice of amount.</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*: FNU and NTU are interchangeable in the UWS. All data reported as NTU.
\(^\d\): This parameter is calculated rather than measured analytically, so MDL is not computed. RPD is also not relevant for this parameter.

**A.8. SPECIAL TRAINING / CERTIFICATION**

UWS trainings are hands on full day events*. They are designed for a wide range of experience in water quality monitoring; ranging from groups with an extensive existing water quality monitoring programs to groups with little to no water quality monitoring experience. The trainings help to ensure all groups are on the same understanding of project QAPP and SOPs. All Monitoring Groups are provided the SOPs and QAPP. They are required to read these documents. Monitoring Groups take notes on the project SOPs during training events to clarify any points that require extra attention. The objective of trainings is to have all Monitoring
Groups, regardless of previous experience, following the project requirements in a unified manner.

The Monitoring Program Coordinator shall ensure that all UWS Monitoring Groups receive appropriate training by organizing and conducting training events. The trainings are mandatory for new and veteran groups and have hands on elements for sonde calibration and usage, filling in datasheets, macrophyte assessments, logger calibration and usage, nutrient sample collection, and filtering chlorophyll a. All topics are covered in guided step by step approach. Training will be assessed as described by checks in Section C.1 Assessment and Response Actions.

The Monitoring Program Coordinator enters training into the project database and records the following information: subject matter (i.e. what type of monitoring and procedures are covered), training course title, date and agenda, name and qualification of trainers, and names of participants trained with associated monitoring group name. The trainings and technical support offered through the Monitoring Program Coordinator, Monitoring Program Field Coordinator, and Science Advisors is in place for the duration of the project. This will ensure new and veteran groups have a reliable source for prompt answers to their inquiries. Groups are prompted to email or call the Monitoring Program Coordinator or Monitoring Program Field Coordinator with their questions. If needed, the Science Advisors will be consulted. This support is mentioned throughout the project duration and emphasized at the trainings. Trainers remind trainees to call at any hint of a question or issue so it can be resolved.

The Monitoring Program Coordinator worked closely with the Science Advisor signatories on this QAPP to confirm procedures are appropriate. He was part of the three person team leading UWS Tier I trainings around Long Island Sound in 2017. The Coordinator oversees Save the Sound’s Water Quality Program which samples water for pathogen-indicator bacteria and participates in the UWS Tier I & II monitoring.

*: UWS trainings for the 2020 season will be held remotely due to concerns and timing in regard to spread of the Coronavirus (Covid-19).

The Coordinator holds a Bachelor of Science Degree in Environmental Studies from University California Santa Barbara and a Master’s of Science Degree in Environmental Science and Management from Sacred Heart University.

Project training shall take place as specified in Table 6.
### Table 6: Project-Specific Training

<table>
<thead>
<tr>
<th>Training: Type &amp; Description</th>
<th>Trainer(s)</th>
<th>Training Date(s)</th>
<th>Trainees</th>
<th>Location of Training Records</th>
</tr>
</thead>
<tbody>
<tr>
<td>UWS Standard Operating Procedures and Methods, QAPP, and Data Entry Training</td>
<td>UWS Monitoring Program Coordinator, UWS Field Coordinator, UWS Science Advisor(s), and other personnel under the supervision of the listed trainer(s)</td>
<td>Annual; Spring before sampling season commences</td>
<td>All participating UWS groups will send 1-3 representatives</td>
<td>Office of the UWS Monitoring Program Coordinator; digital record of attendees and agenda stored on computer and backed up on Save the Sound S-Drive</td>
</tr>
<tr>
<td>- General water quality parameter information</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- Sonde calibration and field training</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- Chlorophyll a field collection, filtering, preservation, &amp; transport</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- Nutrient sampling collection, filtering, preservation, &amp; transport</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- Qualitative macrophyte assessment procedure</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- Quantitative macrophyte assessment procedure</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- Data logger calibration and field training</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- QAPP review and data entry</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### A.9. DOCUMENTS AND RECORDS

**Calibration Datasheet, Sample Event Datasheet and Field Datasheet** will be completed by Monitoring Groups before, during, and after Tier I water quality station sampling event.

**Qualitative Macrophyte Field Datasheet** will be completed upon every qualitative macrophyte survey. Photographs must accompany and be identified in this datasheet.
UWS Nutrient Sample Event Datasheet, Calibration Datasheet, Field Datasheet will be completed upon every Tier II nutrients sampling event. The calibration datasheet will only contain the parameters being recorded in the field: conductivity (salinity).

UWS Quantitative Macrophyte Field Datasheet will be completed upon every quantitative macrophyte survey.

UWS Logger Retrieval Sample Event Datasheet, Field Datasheet, and Calibration Datasheet will be completed upon every data retrieval of data loggers in the field. The calibration datasheet will only contain the parameters being recorded in the field: dissolved oxygen and conductivity.

Sample Labels will be put on all sample containers. Labels will include the station name, organization name, date, time, sample id, and type of sample. Samples needing containers with labels are filters for extracted chlorophyll a and nutrients. Detailed instructions for chlorophyll a filters and nutrient samples are provided in the UWS SOP Chlorophyll, UWS SOP Filtered Nutrients, and UWS SOP Total Nitrogen and Total Phosphorous SOP in Appendix A.

Chain of Custody (COC) forms will accompany samples from collection sites to laboratories. COC forms will be signed by collectors and all individuals who gain custody of the samples until they arrive at a lab. Information will agree with the label information on the sample containers and field datasheet. UWS Chain of Custody forms are in Appendix B.

Training records and field audit information will be kept by the Monitoring Program Coordinator.

The electronic project database shall be organized and protected from loss and damage through proper back-up of digital data on Save the Sound’s S-Drive.

No scientific collecting permits or certificates of permission are required.

The specific forms to be used for this project are provided in Appendix B.
B. Data Generation and Acquisition

B.1. SAMPLING PROCESS DESIGN (EXPERIMENTAL DESIGN)

Tier I water quality sample stations, Tier II nutrients stations (estuary), and quantitative Tier II macrophyte stations were selected to represent the water quality of the entire embayment using a probability-based sampling design\(^6\), as in the EPA National Coastal Assessment\(^7\). Monitoring Program Coordinator and a UWS Science Advisor advised on the choice of station locations. The UWS assigned unique ID codes for the embayments, reporting regions of the embayment, and stations. Water quality stations can be viewed in section A.6.b of this document.

Qualitative macrophyte stations are selected based on local knowledge and observation of the embayment during the course of the May and June sampling events. They are targeted to areas of macrophyte abundance. Monitoring Program Coordinator and a UWS Science Advisor advised on the choice of station locations. UWS SOP Qualitative Macrophytes describes the process for sampling locations for macrophytes. These stations are selected using an adaptive process that requires observations during May and June sampling events. These observations identify the best locations for high macrophyte abundance. The qualitative macrophyte sampling can be classified as judgmental design resulting in directed sampling information to complement the water quality station data. Macrophyte stations may change from year to year but records of locations are maintained by the UWS Monitoring Program Coordinator.

Data logging stations were selected with at least one station per reporting region. Access to station is a strong consideration in the selection process. The final station locations will be conferred with Monitoring Program Coordinator and the UWS Science Advisors before deployment.

Tier II nutrients stations (tributary) were selected based on access and salinity <1 ppt. Parameters, number and location of sampling sites, sampling time of day, frequency, and season are selected to meet the monitoring objectives referred to in Section A.6.a.


Sampling design components are described below:

**Sampling Safety.** Personal safety shall be a primary consideration in all activities, including selection of sampling stations, dates, and training programs. No sampling shall occur when personal safety is thought to be compromised. The Monitoring Group Lead of each participating group on this QAPP shall confer with their respective field teams before each sampling event to decide whether adverse weather or other conditions pose a threat to safety and will cancel/postpone sampling when necessary. Sampling shall take place in teams of two or more. Samplers shall wear life vests when required, in adverse conditions in boats, or wading in waters under difficult conditions. Samplers shall wear proper clothing to protect against the elements.

**Design Considerations.** A summary of design considerations incorporated into this project are included in Table 7. Specifics on the design approaches to the number of stations, depth of sampling, and frequency of sampling and time of day of sampling are included in the SOPs in Appendix A. A summary of general design approaches to the number of stations, depth of sampling, frequency of sampling, and time of day are included here:

There are 215 Tier I water quality stations and approximately 112 qualitative macrophyte station monitored across all the study sites. 16 data logging stations will be monitored. 83 quantitative macrophyte stations will be monitored. 58 Tier II nutrient stations will be monitored. The qualitative macrophyte stations are confirmed by the beginning of the macrophyte monitoring window described in UWS SOP Macrophytes. Sonde profiles for water quality parameters at water quality stations will be sampled 0.5 m below the surface, 0.5 m above bottom, mid-depth if total depth >10 m; if total depth is less than 1.5 m, only a mid-depth reading will be collected. Extracted chlorophyll a samples will be taken from a bucket. Two filters and a corresponding chlorophyll a sonde reading will be taken per sampling event. Land-based qualitative macrophyte stations will be photographed from land. Rake toss qualitative macrophyte stations will be photographed from land or boat. Quantitative macrophyte stations are recorded from a boat. These are the same locations as the Tier I water quality stations in the respective embayments. Tier II nutrients stations are collected 0.5 below the surface. New Tier II nutrients tributary stations are confirmed by field work prior to collecting the first batch of nutrient samples for the season. Data logging stations are 0.5 m off the bottom and record data every 15 minutes. These stations are selected and confirmed with the Monitoring Program Coordinator and a UWS Science Advisor prior to commencement of the season.
Table 7: Sampling Approaches.
Assessment Type: Tier I water quality Stations.

<table>
<thead>
<tr>
<th>Indicators</th>
<th>Number of sample locations</th>
<th>Frequency, duration, special conditions</th>
<th>Field survey QC</th>
</tr>
</thead>
<tbody>
<tr>
<td>GPS: latitude &amp; longitude in decimal degrees; NAD83 coordinate system or record system used</td>
<td>every station</td>
<td>Twice a month from May - October, within 3 hours of sunrise, repeat readings every time a station is sampled. Coordinates indicating a 100 m or greater discrepancy will be assessed and documented in final report. Reference land site, once per sampling event Once per field day, take readings twice at the last station sampled</td>
<td></td>
</tr>
<tr>
<td>station depth</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>sample depth</td>
<td>every station:</td>
<td></td>
<td></td>
</tr>
<tr>
<td>temperature</td>
<td>if station depth &lt; 1.5 m, mid-depth; if station depth &gt; 1.5 m &amp; &lt; 10 m, 0.5 m below surface and 0.5 m above bottom; if station depth &gt; 10 m, 0.5 m below surface, 0.5 m above bottom, and mid-depth</td>
<td>once per field day, take readings twice for replicate at the last station sampled Calibration per SOPs</td>
<td></td>
</tr>
<tr>
<td>salinity</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>dissolved oxygen</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>turbidity</td>
<td>every station, 0.5 m below surface</td>
<td></td>
<td>take readings twice for replicate at the last station sampled</td>
</tr>
<tr>
<td>chlorophyll a</td>
<td>once per sampling event from bucket at reference station</td>
<td></td>
<td>calibration per SOPs Collect filter and sonde readings at reference station</td>
</tr>
</tbody>
</table>
Stations are representative, defined clearly in respective SOPs

**Assessment Type: Qualitative Macrophyte Surveys.**

<table>
<thead>
<tr>
<th>Indicators</th>
<th>Number of sample locations</th>
<th>Frequency, duration, special conditions</th>
<th>Field survey QC</th>
</tr>
</thead>
<tbody>
<tr>
<td>GPS: latitude &amp; longitude in decimal degrees; NAD83 coordinate system or record system used</td>
<td>Every station</td>
<td>Sample 3 days during the 3-week period starting July 15 and ending August 7. Try to sample once per week. If this is not possible, sample such that you maximize the days between sampling. All three days cannot be sampled in the same 7-day window. 2 sampling events or an event outside this time criteria may be accepted in unforeseeable circumstances. Consultation with Monitoring Program Coordinator and UWS Science Advisor is necessary for this decision. See UWS SOP Macrophytes for additional details.</td>
<td>repeat readings every time a station is sampled. coordinates indicating a 100 m or greater discrepancy will be assessed and documented in final report reference land site, once per sampling event</td>
</tr>
<tr>
<td>Macrophyte Abundance</td>
<td></td>
<td></td>
<td>Photos and assessment (none, some, lots) of each sample are reviewed by the Monitoring Program Coordinator and UWS Science Advisor</td>
</tr>
</tbody>
</table>
Stations are targeted, defined clearly in SOP

**Assessment Type: Tier II nutrients Stations.**

<table>
<thead>
<tr>
<th>Indicators</th>
<th>Number of sample locations</th>
<th>Frequency, duration, special conditions</th>
<th>Field survey QC</th>
</tr>
</thead>
<tbody>
<tr>
<td>GPS: latitude &amp; longitude in decimal degrees; NAD83 coordinate system or record system used</td>
<td>Every station</td>
<td>Each visit to sample station; at least monthly (May – October), with 14 days separation</td>
<td>Repeat readings every time the station is sampled to verify coordinates. Coordinates indicating a 100 m or greater discrepancy from documented coordinates will be assessed and documented in data notes.</td>
</tr>
<tr>
<td>Salinity</td>
<td>Every station</td>
<td>Each visit to sample station; at least monthly (May – October), with 14 days separation</td>
<td>Probe calibration prior to survey; post sampling event readings in standard</td>
</tr>
<tr>
<td>Nutrients</td>
<td>Every station</td>
<td>Each visit to sample station; at least monthly (May – October), with 14 days separation</td>
<td>At minimum one field replicate per sampling event</td>
</tr>
</tbody>
</table>

Stations are representative.

**Assessment Type: Data logging stations.**

<table>
<thead>
<tr>
<th>Indicators</th>
<th>Number of sample locations</th>
<th>Frequency, duration, special conditions</th>
<th>Field survey QC</th>
</tr>
</thead>
<tbody>
<tr>
<td>GPS: latitude &amp; longitude in decimal degrees; NAD83 coordinate system or record system used</td>
<td>Every station</td>
<td>Each visit to sample station</td>
<td>Repeat readings every time the station is sampled to verify coordinates. Coordinates indicating a 100 m or greater discrepancy from documented coordinates will be assessed and documented in data notes.</td>
</tr>
<tr>
<td>Conductivity</td>
<td>Every station</td>
<td>Each visit to sample station</td>
<td>Probe calibration prior to survey; post sampling event readings in standard</td>
</tr>
<tr>
<td>Dissolved oxygen</td>
<td>Every station</td>
<td>Each visit to sample station</td>
<td>Probe calibration prior to survey; post sampling event readings in standard</td>
</tr>
<tr>
<td>Barometric pressure</td>
<td>Every station</td>
<td>Each visit to sample station</td>
<td>Not applicable</td>
</tr>
</tbody>
</table>

Stations are selected for access and other considerations addressed in this QAPP and UWS Data Logging SOP.
### Assessment Type: Quantitative Macrophyte Surveys.

<table>
<thead>
<tr>
<th>Indicators</th>
<th>Number of sample locations</th>
<th>Frequency, duration, special conditions</th>
<th>Field survey QC</th>
</tr>
</thead>
<tbody>
<tr>
<td>GPS: latitude &amp; longitude in decimal degrees; NAD83 coordinate system or record system used</td>
<td>Every station</td>
<td>Each visit to sample station while conducting all camera descents to bottom. Obtained from GPS track</td>
<td>Not applicable</td>
</tr>
<tr>
<td>Bottom coverage (% macroalgae, % bare, % eelgrass)</td>
<td>Every station</td>
<td>Each visit to sample station with specifications in SOP on image count and analysis</td>
<td>Not applicable</td>
</tr>
</tbody>
</table>

#### B.2. SAMPLING METHODS

Pre-coordination shall occur with the external lab to ensure that sample collection procedures meet lab needs. The project lab for this study is below:

Interstate Environmental Commission Lab, contact: Evelyn Powers, epowers@iec-nynjct.org, 718-982-3792, c/o College of Staten Island-CUNY, 2800 Victory Blvd., Building 6S, Room 106, Staten Island, NY 10314

A laboratory of equal or higher certification than Interstate Environmental Commission can be considered if Interstate Environmental Commission is unable to complete project analyses. This surrogate laboratory must adhere to analytical methods in Table 10.

To comply with UWS program guidelines, all sample collections for this project shall follow detailed methods on how samples will be collected and preserved as stated in the standard operating procedures (SOPs) contained in Appendix A of this document. The lab has reviewed the UWS SOPs and confirms they are appropriate for the select analyses.

A summary overview of sample collection methods is provided in Table 8. A summary of field sampling considerations is provided in Table 9.

Any filters or nutrient sample bottles collected for analysis will be stored in a cooler, on ice during the sampling trip. The cooler designated for these samples will not be used for the storage of macrophytes.

All deviations from the Standard Operating Procedures of this QAPP will be documented and subsequently reviewed by the Monitoring Program Coordinator and the project UWS Science Advisors. This information will be available to all signatories at the completion of this project at which time acceptability of data will be determined.
Table 8: Overview of Sample Collection Methods
Assessment Type: Tier I water quality Parameters

<table>
<thead>
<tr>
<th>Parameter(s)</th>
<th>Container Type(s) and Preparation</th>
<th>Minimum Sample Quantity per Sample Depth (unless otherwise noted)</th>
<th>Sample Preservation</th>
<th>Maximum Holding Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>GPS: latitude &amp; longitude in decimal degrees; NAD83 coordinate system or record system used</td>
<td><strong>in situ</strong></td>
<td>1 / station</td>
<td>transfer to digital format, maintain back-up copies of digital data</td>
<td>NA</td>
</tr>
<tr>
<td>Station depth</td>
<td><strong>in situ</strong></td>
<td>1 / station</td>
<td>transfer to digital format, maintain back-up copies of digital data</td>
<td>NA</td>
</tr>
<tr>
<td>Sample depth (metered line)</td>
<td><strong>in situ</strong></td>
<td>1, and for remainder of the sampling event if the intercomparison with project sonde and line is &gt; 0.3 m</td>
<td>transfer to digital format, maintain back-up copies of digital data</td>
<td>NA</td>
</tr>
<tr>
<td>Multiparameter sonde:</td>
<td><strong>in situ</strong></td>
<td>1 / sample depth and a second reading for each depth at the last station of the day</td>
<td>transfer to digital format, maintain back-up copies of digital data</td>
<td>NA</td>
</tr>
<tr>
<td>- depth</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- temperature</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- salinity</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- dissolved oxygen</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- turbidity</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- chlorophyll a</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>chlorophyll a (extracted, fluorometric analysis)</td>
<td><strong>Large bucket rinsed three times with surface water. Bucket volume must accommodate sonde for reading in situ</strong></td>
<td>Filtered sample volume must be sufficient to provide “color” on the filter pad; 10 mL to 180 mL, 2 filters collected and one sonde reading from bucket per sampling event</td>
<td>GF/F filter is blown dry with a 60mL syringe and stored in the dark (foil wrapped), on ice; transferred to - 20°C freezer within 12 hours</td>
<td>28 days</td>
</tr>
</tbody>
</table>
### Assessment Type: Qualitative Macrophyte Abundance

<table>
<thead>
<tr>
<th>Parameter(s)</th>
<th>Container Type(s) and Preparation</th>
<th>Minimum Sample Quantity</th>
<th>Sample Preservation</th>
<th>Maximum Holding Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>GPS: latitude &amp; longitude in decimal degrees; NAD83 coordinate system or record system used</td>
<td><em>in situ</em></td>
<td>1 / station</td>
<td>transfer to digital format, maintain back-up copies of digital data</td>
<td>NA</td>
</tr>
<tr>
<td>Macrophyte abundance</td>
<td>digital photos</td>
<td>1 / sample (a sample is a single rake toss or required distance for soft shoreline photo)</td>
<td>transfer to a computer, upload to online datasheet</td>
<td>NA</td>
</tr>
</tbody>
</table>

### Assessment Type: Tier II nutrients Parameters

<table>
<thead>
<tr>
<th>Parameter(s)</th>
<th>Container Type(s) and Preparation</th>
<th>Minimum Sample Quantity</th>
<th>Sample Preservation</th>
<th>Maximum Holding Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>GPS: latitude &amp; longitude in decimal degrees; NAD83 coordinate system or record system used</td>
<td><em>in situ</em></td>
<td>NA</td>
<td>transfer to digital format; maintain back-up copies of digital data</td>
<td>NA</td>
</tr>
<tr>
<td>Multiparameter sonde: • Salinity</td>
<td><em>in situ</em></td>
<td>NA</td>
<td>transfer to digital format; maintain back-up copies of digital data</td>
<td>NA</td>
</tr>
<tr>
<td>Inorganic nutrients</td>
<td>high density polyethylene (HDPE) polypropylene (new containers washed with ASTM Type 1 Ultrapure Water, used containers pre-acid-washed with 10% hydrochloric acid)</td>
<td>120 mL per station</td>
<td>ice or refrigerate filtered water samples at a temperature of &lt;4 C while in the field, store at &lt;-20 C</td>
<td>holding time of ~1 year once frozen</td>
</tr>
<tr>
<td>Total nutrients</td>
<td>high density polyethylene (HDPE) polypropylene (new containers washed with ASTM Type 1 Ultrapure Water, used containers pre-acid-washed with 10% hydrochloric acid)</td>
<td>120 mL per station</td>
<td>ice or refrigerate water samples at a temperature of &lt;4 C while in the field, freeze at &lt;-20 C</td>
<td>holding time of ~1 year once frozen</td>
</tr>
</tbody>
</table>
**Assessment Type: Quantitative Macrophyte Abundance**

<table>
<thead>
<tr>
<th>Parameter(s)</th>
<th>Container Type(s) and Preparation</th>
<th>Minimum Sample Quantity</th>
<th>Sample Preservation</th>
<th>Maximum Holding Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Macrophyte abundance (% bare, % macroalgae, % eelgrass)</td>
<td>Field data sheets and computer storage</td>
<td>20 still images per station; more if heterogeneity is observed. Detailed procedure in SOP</td>
<td>maintain back-up copies of digital data</td>
<td>NA</td>
</tr>
</tbody>
</table>

**Table 9: Overview of Field Sampling Considerations**

<table>
<thead>
<tr>
<th>Sample Type</th>
<th>Parameter(s)</th>
<th>Sampling Considerations</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>In-situ</em> sampling</td>
<td>Station depth</td>
<td>Note the tidal stage and time of day. Depth varies greatly over the tidal cycle.</td>
</tr>
<tr>
<td><em>In-situ</em> sampling, GPS</td>
<td>GPS: latitude &amp; longitude in decimal degrees; NAD83 or WGS84 coordinate system, record system used</td>
<td>NAD83 or WGS84 coordinate system, record system used; check GPS accuracy relative to a known, fixed location</td>
</tr>
<tr>
<td><em>In-situ Tier I water quality sampling, multiparameter sonde</em></td>
<td>Depth, Temperature, Salinity, Dissolved oxygen, Turbidity, Chlorophyll a fluorescence</td>
<td>Sample within 3 hours of sunrise. Inspection, maintenance, pre-calibration and post-checking of probes and instruments are critical to achieving accurate and precise measurements.</td>
</tr>
<tr>
<td>Data logging stations</td>
<td>Dissolved Oxygen, Conductivity (Salinity), Barometric Pressure</td>
<td>Inspection, maintenance as specified by manufacturer, and calibration of instruments are critical to achieving accurate and precise measurements, especially for DO. Loggers are rinsed and cleaned with freshwater after each retrieval and use.</td>
</tr>
<tr>
<td>Grab samples - i.e. collection of a water sample</td>
<td>Chlorophyll a</td>
<td>Keep careful and accurate track of volume of water passed through each filter pad, quantitation is impossible without this value.</td>
</tr>
<tr>
<td>Qualitative macrophyte abundance</td>
<td>Macrophyte abundance</td>
<td>Be sure to photograph all sites and samples. Record identifier for each photo on the datasheet.</td>
</tr>
<tr>
<td>Quantitative macrophyte abundance</td>
<td>Macrophyte abundance</td>
<td>Maintain low speed to minimize potential damage to camera. Monitoring Group Lead and two additional members of the sampling team, under Monitoring Group Lead supervision, will analyze the macrophyte videos as described in SOP</td>
</tr>
</tbody>
</table>
Sample Type                  | Parameter(s) | Sampling Considerations                                                                                                                                                                                                                                                                                                                                 
---                          |--------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------- 
Grab samples - i.e. collection of a water sample in bottle | Inorganic and total nutrients | Triple-rinse sample container in ambient water immediately prior to sample collection. Care must be taken to avoid contact between fingers and inside surfaces of containers, including bottle caps. New, pre-washed bottles preferred; if not, containers for nutrient samples should be acid-washed and rinsed with deionized water. This process is overseen by the Monitoring Coordinator. These bottles will be obtained by appropriate suppliers such as Fischer Scientific. Field filtration preferred for dissolved fractions. If filtering water, triple-rinse container with filtered water immediately prior to sample collection, not ambient water.  

B.3. SAMPLE HANDLING AND CUSTODY

Sample handling and labeling procedures shall comply with project Standard Operating Procedures (SOPs). Chlorophyll a filters and nutrient samples will be transported on ice in a cooler to the freezer on the same day as sampling occurs. Filters and nutrient samples will always be transported on ice with no more than 24 hours out of freezer to avoid thawing.

Sample labels will be associated with: station name, date, time, volume filtered, sample id, type of sample, and organization name. These details may be written on the label. Information will also be filled in the field data sheet.

Chain of Custody shall be tracked as detailed in the SOPs. The project Chain of Custody forms are provided in in Appendix B.

The following steps shall be taken to avoid sample mislabeling:

Labels will be prepared in advance and cross checked with the field datasheet before sampling event. Field team will check data sheet versus sample filter labels before storing in the cooler for transport to a freezer. A white board with name of the embayment, site and station id, and date will be filmed prior to recording every station in the macrophyte video surveys.

B.4. ANALYTICAL METHODS

To comply with the requirements of the UWS Program, all analytical methods used in the Monitoring Program, including methods used by laboratories performing analyses for the project, shall be based on standardized laboratory methods.

All analytical methods used for this project are provided in Appendix C.
Table 10 provides an overview of the analytical methods utilized in this Monitoring Program. The SOPs associated with these methods are included in Appendix C.

Table 10: Overview of Analytical Methods.

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Chlorophyll a</td>
<td>EPA 445.0</td>
<td>EPA</td>
<td>0.7 µg/l</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nitrite</td>
<td>EPA 353.2</td>
<td>EPA</td>
<td>0.004 mg/l</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nitrate+Nitrite</td>
<td>EPA 353.2</td>
<td>EPA</td>
<td>0.004 mg/l</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ammonia</td>
<td>EPA 350.1</td>
<td>EPA</td>
<td>0.020 mg/l</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total Dissolved</td>
<td>EPA 353.2</td>
<td>EPA</td>
<td>0.05 mg/l</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nitrogen</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Orthophosphate (DIP)</td>
<td>EPA 365.1</td>
<td>EPA</td>
<td>0.001 mg/l</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total Nitrogen</td>
<td>EPA 353.2</td>
<td>EPA</td>
<td>0.05 mg/l</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total Phosphorous</td>
<td>EPA 365.1</td>
<td>EPA</td>
<td>0.334 mg/L</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

B.5. QUALITY CONTROL

Lab Quality Control (QC) protocols shall be discussed with the external lab facility or contractor analyzing chlorophyll a and nutrient samples prior to sampling to ensure acceptability.

Quality control shall be discussed and defined prior to sampling (e.g., during training).

Details on quality control procedures are provided in Table 11.

Table 11: Quality Control Measures

Note that 5% of field samples equates to one station per field day sampled as replicate.

<table>
<thead>
<tr>
<th>Sample Type</th>
<th>Instrument/Parameter</th>
<th>Accuracy Checks</th>
<th>Precision Checks</th>
<th>% Field QC Samples (blanks and field duplicates)</th>
</tr>
</thead>
<tbody>
<tr>
<td>GPS coordinates</td>
<td>GPS or Smart Phone app / GPS coordinates</td>
<td>Compare location of reference site to Google Earth coordinates</td>
<td>Readings at a land-based reference point and duplicate readings at one station</td>
<td>1 / field day</td>
</tr>
<tr>
<td>Station depth</td>
<td>metered line / depth</td>
<td>re-measure line</td>
<td>replicate readings at one station</td>
<td>5%</td>
</tr>
<tr>
<td>Sample Type</td>
<td>Instrument/Parameter</td>
<td>Accuracy Checks</td>
<td>Precision Checks</td>
<td>% Field QC Samples (blanks and field duplicates)</td>
</tr>
<tr>
<td>-------------------------------------</td>
<td>--------------------------------------------------------------------------------------</td>
<td>----------------------------------------------------------------------------------</td>
<td>-----------------------------------------------------------------------------------</td>
<td>-----------------------------------------------</td>
</tr>
<tr>
<td>Multiparameter sonde and data loggers</td>
<td>Depth, temperature, conductivity, dissolved oxygen, turbidity, chlorophyll a</td>
<td>Pre-survey calibration and post-survey checks, including “zero” DO standard check</td>
<td>field duplicates or 3-5 minutes stable readings recorded</td>
<td>5% or verify repeatability in the field</td>
</tr>
<tr>
<td>Water samples - grab</td>
<td>Fluorometric determination of extracted chlorophyll a</td>
<td>Acetone blank, standard</td>
<td>QC check for multiparameter sonde</td>
<td>100%</td>
</tr>
<tr>
<td>Qualitative Macrophyte abundance</td>
<td>Observation / macrophyte abundance</td>
<td>Photos of all assessments</td>
<td>Photos of all assessments</td>
<td>100% photos are required for inclusion of the data in the UWS</td>
</tr>
<tr>
<td>Water samples – grab</td>
<td>TP, P fractions</td>
<td>Field: blanks Lab: analysis of lab-fortified matrix (spiked samples) and/or lab QC standard</td>
<td>Field duplicates Lab duplicates</td>
<td>Minimum 5%</td>
</tr>
<tr>
<td>Quantitative Macrophyte abundance</td>
<td>% coverage bare, macrophytes, and animals of bottom</td>
<td>Still images of all assessments</td>
<td>Still images of all assessments</td>
<td>100% still images are required for inclusion of the data in the UWS</td>
</tr>
</tbody>
</table>

**B.6. INSTRUMENT / EQUIPMENT TESTING, INSPECTION AND MAINTENANCE**

Maintenance of instruments and equipment shall occur as needed during the field season. Annual maintenance and intercalibration assurance will be conducted by Save the Sound.

Records of equipment inspection, maintenance, repair and replacement shall be kept in a logbook. A backup of the logbook will be kept in a separate location. If the logbook is digital, appropriate backups of the computer files will be maintained by Monitoring Program Coordinator.

**Table 12: Instrument / Equipment Inspection and Testing Procedures**

<table>
<thead>
<tr>
<th>Equipment Type</th>
<th>Inspection Frequency</th>
<th>Type Inspection</th>
<th>Maintenance, Corrective Action</th>
</tr>
</thead>
<tbody>
<tr>
<td>GPS unit</td>
<td>before each sampling date</td>
<td>battery life</td>
<td>charge batteries</td>
</tr>
<tr>
<td>Equipment Type</td>
<td>Inspection Frequency</td>
<td>Type Inspection</td>
<td>Maintenance, Corrective Action</td>
</tr>
<tr>
<td>------------------------------------</td>
<td>-----------------------------------------------</td>
<td>------------------------------------------------------</td>
<td>---------------------------------------------------------------------</td>
</tr>
<tr>
<td>Depth line</td>
<td>Annually, or when a potential problem is noted</td>
<td>Check the calibrated line against a meter tape</td>
<td>Wipe tape after each use, if line has stretched or is damaged, replace immediately and note recent data as questionable</td>
</tr>
<tr>
<td>Multiparameter sonde</td>
<td>Before each sampling date</td>
<td>Battery life, electrical connections, sensor condition</td>
<td>Charge batteries, spare sensors as appropriate, batteries</td>
</tr>
<tr>
<td>Filtering apparatus (chlorophyll a)</td>
<td>Before each use</td>
<td>Proper functioning, clean storage</td>
<td>Spare filters and syringe</td>
</tr>
<tr>
<td>Collection rake, rope</td>
<td>Before each collection</td>
<td>Visually for integrity</td>
<td>Repair, replace keep spares on hand</td>
</tr>
<tr>
<td>Filtering apparatus (nutrients)</td>
<td>Before each use</td>
<td>Proper functioning, clean storage</td>
<td>Spare syringe, spare filters, spare pump tubing</td>
</tr>
<tr>
<td>Logging sensors</td>
<td>Every 7-10 days or as needed</td>
<td>Biofouling and battery check</td>
<td>Clean off fouling organisms, check battery life from data log</td>
</tr>
<tr>
<td>Underwater camera and equipment</td>
<td>Before each use</td>
<td>Battery life, test video</td>
<td>Recharge/replace batteries and clean lens if required</td>
</tr>
</tbody>
</table>

### B.7. INSTRUMENT / EQUIPMENT CALIBRATION AND FREQUENCY

Calibration shall occur within a day prior to a sampling trip.

Records of calibration shall be kept in a logbook (hard copy or digital, with back-ups). Calibration records shall be maintained for a minimum of four years, ideally longer. Monitoring Groups will deliver calibration records to the Monitoring Program Coordinator. These records and digital backups will be saved on Save the Sound’s S-Drive for duration of the project.

A summary of calibration procedures for instruments and equipment is provided in Table 13.

Detailed calibration procedures are described in SOPs contained in Appendices A.

#### Table 13: Instrument / Equipment Calibration Procedures

<table>
<thead>
<tr>
<th>Instrument Type</th>
<th>Inspection and Calibration Frequency</th>
<th>Standard of Calibration Instrument Used</th>
<th>Calibration Acceptance Criteria</th>
<th>Corrective Action</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calibrated lines (for depth)</td>
<td>Annually</td>
<td>Tape measure</td>
<td>Within 0.1 m of tape measure</td>
<td>Recalibrate or replace with calibrated line</td>
</tr>
<tr>
<td>Multiparameter sonde</td>
<td>Before each sampling run</td>
<td>Standard solutions</td>
<td>According to manufacturer’s instruction or when not provided a maximum difference of %10 of the calibration standard value</td>
<td>According to UWS and manufacturer’s instruction</td>
</tr>
</tbody>
</table>
### B.8. Inspection / Acceptance of Supplies and Consumables

The procedures for inspection and acceptance of supplies and consumables listed in Table 14 shall be followed by the Monitoring Groups.

#### Table 14: Supplies Inspection and Acceptance Procedures

<table>
<thead>
<tr>
<th>Supplies</th>
<th>Inspection Frequency</th>
<th>Type of Inspection</th>
<th>Available Parts</th>
<th>Maintenance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calibration standards</td>
<td>Before each sampling date</td>
<td>Visual inspection of quantity and expiration date</td>
<td>Spare, fresh solutions</td>
<td>Storage according to manufacturer’s recommendations, annual replacement at beginning of sampling season</td>
</tr>
<tr>
<td>Sonde sensors, filters</td>
<td>Before each sampling date</td>
<td>Visual inspection of quantity, integrity</td>
<td>Spares</td>
<td>Storage according to manufacturer’s recommendations</td>
</tr>
<tr>
<td>Field and lab sample sheets</td>
<td>Before each sampling date</td>
<td>Visual</td>
<td>Additional copies</td>
<td></td>
</tr>
<tr>
<td>Cooler</td>
<td>Before each sampling date</td>
<td>Cleanness, ice packs</td>
<td></td>
<td>Annually or as needed</td>
</tr>
<tr>
<td>Sample bottles</td>
<td>Before each sampling date</td>
<td>Integrity, cleanness and seal for nutrient bottles, verified sterility of bacterial sample bottles</td>
<td>One set of spare bottles</td>
<td>Clean after use (note that nutrient bottles require acid washing before reuse)</td>
</tr>
</tbody>
</table>

### B.9. Non-direct Measurements

To provide high-quality data to enhance the interpretation of data collected as part of this Monitoring Program, data may be acquired from qualified sources approved by Monitoring Program Coordinator. NOAA tide gauges will be used for tide information. Precipitation will be acquired from local weather stations that log reasonable (in respect to northeastern USA conditions) volumes. Precipitation data out of the expected annual volumes and the observed conditions will be flagged and discussed with Monitoring Program Coordinator and shared with quality assurance personnel for review and potential disqualification. External data sources are described in Table 15.
Table 15: Non-Project Data Validity
The following data will be used as part of the Monitoring Program. This is a secondary use of data.

<table>
<thead>
<tr>
<th>Title or descriptive name of data document.</th>
<th>Source of data.</th>
<th>QAPP written? Y/N</th>
<th>Notes on quality of data.</th>
<th>Planned restrictions in use of the data due to questions about data quality.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Time of low and high tide</td>
<td>NOAA tide gauges recorded on field data sheet</td>
<td>N</td>
<td>NOAA has internal requirements for data suitability. High and low tide data are not generally available at the embayment. Data from NOAA tide gauges are acceptable; data are used in broad scale, to determine the potential impact of stage in tidal cycle on the day of sampling.</td>
<td>Data quality is acceptable. However, local tidal stage will differ from the nearest NOAA gauge even when corrected for difference in location. These data are rough predictors only.</td>
</tr>
<tr>
<td>High and low temperature and precipitation within the 24 hours prior to the field trip</td>
<td>Local weather station recorded on field data sheet</td>
<td>N</td>
<td>Air temperature within the last 24 hours are not generally available at the embayment. Data from nearby weather stations are acceptable; data are used in broad scale, to determine the potential impact of weather on the day of sampling.</td>
<td>Data will be used in comparing among embayments or among dates, as a general indication of weather during the day prior to sampling.</td>
</tr>
</tbody>
</table>

B.10. DATA MANAGEMENT

Field teams shall record data on field sheets, review them, and turn over to respective Monitoring Group Lead or designated appointee.

Monitoring Group Leads or designated appointees shall review sheets and confer with field teams on any needed corrective action.

The designated person shall fill out the chain-of-custody form for forwarding samples to the external laboratory. Each person who handles or transports samples shall also sign the custody form upon receipt of the samples. Chain of custody forms will follow samples to the lab and back to Monitoring Program Lab Coordinator by mail or pickup after each analysis run is completed. Alternatively, scanned copies may be emailed or faxed. These copies will be sent to Monitoring Group Leads or designated appointees.

Once laboratory analyses are complete, the laboratory personnel shall deliver (digital or hard copy) lab results to the Monitoring Program Lab Coordinator or arrange for pickup. These results will be sent to all Monitoring Group Leads or designated appointees.

The Monitoring Group Lead or other trained designee will enter raw field and lab data into the project computer system.

Computer-entered data shall be compared with field sheets for accuracy.
Original data sheets will be stored by the Monitoring Group Leads or designated appointees, following data entry into the UWS data entry template.

Digital back-ups and copies of the non-digitized data will be made and stored in a separate location designated by the Monitoring Group Lead or designated appointees and delivered to the Monitoring Program Coordinator.

Documentation of data recording and handling, including all problems and corrective actions, shall be included in all preliminary and final reports.

Table 16 in this document accurately represents the procedures utilized by the UWS for data management, review, validation, and verification.

**Table 16: Data Management, Review, Validation, Verification Process Summary**

The term “Field sampler” refers to the person conducting the sampling in the field.

<table>
<thead>
<tr>
<th>Activity</th>
<th>By whom</th>
<th>Corrective action, if needed</th>
</tr>
</thead>
<tbody>
<tr>
<td>Conduct field audits of Monitoring Groups performing calibrations and demonstrating field procedures.</td>
<td>Monitoring Program Field Coordinator or appointed designee</td>
<td>Correct any discrepancies with this QAPP or SOPs</td>
</tr>
<tr>
<td>Check labels just prior to sampling, to ensure correct labeling of container.</td>
<td>Field sampler</td>
<td>Correct label</td>
</tr>
<tr>
<td>At time of sampling, record data, sign field sheets.</td>
<td>Field sampler</td>
<td>Remind samplers of proper procedures; retrain if needed.</td>
</tr>
<tr>
<td>Fill out, sign chain of custody (COC) forms for any samples going to lab.</td>
<td>Field sampler or designated person</td>
<td>Remind person of proper procedures; retrain if needed.</td>
</tr>
<tr>
<td>Before turning field sheets over to Monitoring Group Lead or designated appointee, check for reasonableness to expected range, completeness.</td>
<td>Field sampler</td>
<td>Resample if feasible; otherwise, flag suspect data.</td>
</tr>
<tr>
<td>Upon receipt of field sheets, recheck for reasonableness to expected range, completeness, accuracy, and legibility.</td>
<td>Monitoring Group Lead or designated appointee</td>
<td>Confer with field sampler(s) immediately or within 24 hours. Resample if feasible; otherwise, flag suspect data.</td>
</tr>
<tr>
<td>Upon receipt of samples, field sheets and COC forms, check to see that sheets and forms correspond to number of samples, condition of samples as stated on COC forms. Sign COC forms. Copies of field sheets and COC forms are made, given to Monitoring Program Coordinator.</td>
<td>Monitoring Group Lead or designated appointee</td>
<td>Contact field samplers as needed to locate missing samples, data records. In case of missing/spoiled samples or data records, authorize resampling as needed and feasible. If resampling is not feasible, flag all suspect data.</td>
</tr>
<tr>
<td>Upon completion of laboratory analyses, fill out lab sheets, including data on QC tests.</td>
<td>External Lab</td>
<td>Re-analyze if possible. If not, confer with Monitoring Program Laboratory Coordinator. Flag all suspect data.</td>
</tr>
<tr>
<td>Upon receipt of lab data, review for completeness and legibility.</td>
<td>Monitoring Group Lead or designated appointee</td>
<td>Confer with Monitoring Program Laboratory Coordinator.</td>
</tr>
</tbody>
</table>
Activity | By whom | Corrective action, if needed
--- | --- | ---
Upon completion of data entry, compare with field/lab sheets for accuracy. | Monitoring Group Lead or other volunteer. Data entry personnel may review their own work, but it cannot be on the same day as data entry. | Re-enter or correct data.

Translate data into preliminary data reports: run statistical analyses and/or prepare graphical summaries of data. Check for agreement with QC objectives for completeness. | Monitoring Program Coordinator | Confer with QA Officers and UWS Science Advisor(s). Flag or discard suspect data.

In-season (at least once) and end of season review of collected data sets (individual sample runs and season-total compilations); review for completeness and agreement with QC objectives and DQOs. | Monitoring Group Lead or designated appointee | Flag suspect data. Confer with Monitoring Program Quality Assurance Officer.

**Data Management Systems** – spreadsheets, databases, statistical or graphical software packages, location of data records (paper and electronic), are described here:

All data will be entered from field data sheets to an Excel spreadsheet for storage and retrieval by Monitoring Group Leads and appointed individuals. Digital copies of all datasheets will be kept on file on the S-Drive server in Save the Sound office for at least 4 years with a plan to keep records for duration of the project and beyond. The S-Drive is backed up weekly.

**C. Assessment and Oversight**

**C.1. ASSESSMENT AND RESPONSE ACTIONS**

The Monitoring Program Coordinator and UWS Science Advisors will identify and effectively address any issues that affect data quality, personal safety, and other important project components. The progress and quality of the monitoring program shall be assessed to ensure the objectives are being accomplished. The Monitoring Program Coordinator or appointed designees will check at the end of every month from May - October to confirm the following:

a. Monitoring is occurring as planned.
b. Sufficient written commentary and supporting photographs exist.
c. Sufficient field members are available for all sampling groups.
d. Samplers are collecting in accordance with project schedules.
e. Datasheets and custody control sheets are being properly completed and signed.
f. Retraining or other corrective action is implemented at the first hint of non-compliance with the QAPP or SOPs.

g. Labs are adhering to the requirements of this QAPP in terms of work performed, accuracy, acceptable holding times, timely and understandable results and delivery process.

h. Data management is being handled properly, i.e. data are entered on a timely basis, is properly backed up, is easily accessed, and raw data are properly stored in a safe place.

i. Procedure for developing and reporting the results exists.

Monitoring Groups will be assessed on their ability to follow UWS procedures during field audits overseen by the Monitoring Program Quality Assurance Officer and Field Coordinator. The Monitoring Program Field Coordinator or designee will observe each monitoring group undertaking calibrations and field procedures once in May-June and follow up calls with Monitoring Groups will be scheduled after initial field audits. Field procedures will be reviewed from a set location on the water that does not need to be a UWS monitoring station. A dock or boat in a slip will be appropriate for these field audits. The CTDEEP and NYSDEC representative on this QAPP distribution list will be provided dates for field audits being held in embayments within their respective management areas. CTDEEP and NYSDEC staff have the option to attend the field audits as observers. The Monitoring Program Quality Assurance Officer or designee will conduct a midseason check in call in August to all Monitoring Groups.

The Monitoring Program Coordinator shall confer with the UWS Science Advisors as necessary to discuss any problems that occur and what corrective actions are needed to maintain program integrity. In addition, the Monitoring Program Coordinator and UWS Science Advisors shall meet at the end of the sampling season, to review the draft report and discuss all aspects of the program and identify necessary program modifications for future sampling activities. All problems discovered and program modifications made shall be documented in the final version of the project report. If modifications require changes in the Quality Assurance Project Plan, these changes shall be submitted to the QAPP distribution list for review.

If data are found to be consistently outside the Data Quality Objectives as defined in section A.7. of this documents the Monitoring Program Coordinator shall review the program and correct problems as needed. Corrections may include retraining groups; rewriting sampling instructions; replacement of staff/Monitoring Group(s); alteration of sampling schedules, sites, stations or methods; or other actions deemed necessary. This information will be logged and maintained by the Monitoring Program Quality Assurance Officer. It will also be included in the QAPP Final Report.

C.2. REPORTS TO MANAGEMENT

Data that have passed the project quality assurance may be posted on the organization’s web site, shared with the local media or at other venues (e.g. kiosks at recreation access sites), and submitted to the Long Island Sound Study, New England Interstate Water Pollution Control
Commission, Interstate Environmental Commission, New York State Department of Environmental Conservation, New York City Department of Environmental Protection and/or Connecticut Department of Energy and Environmental Protection. A caveat will accompany these or any data released on a preliminary basis, explaining that they are for review purposes only and subject to correction after completion of a full data review occurring at the end of the sampling season.

The Monitoring Program Coordinator will write a final report. This will be sent to the distribution list on this QAPP. A final workbook of data from all embayments will accompany the report. The final report will also include (updated as necessary) any tables and graphs that were developed for initial data distribution efforts (i.e. the web site and media), and it will describe the program's goals, methods, quality control results, and recommendations. This report may also be used in public presentations.

All reports, preliminary or final, will include discussion of steps taken to assure data quality, findings on data quality, and decisions made on use, censorship, or flagging of questionable data. Any data that are censored in reports will be either referred to in this discussion, or presented but noted as censored.

In short, the final report will include:

- Raw data
- QC data
- Associated metadata
- Questionable data, flagged
- Identification of status as “preliminary” or “final” report
### Table 17: Report Mechanisms, Responsibilities, and Distribution

<table>
<thead>
<tr>
<th>Reporting Mechanism</th>
<th>Person Responsible for writing report</th>
<th>Distribution list</th>
</tr>
</thead>
<tbody>
<tr>
<td>Monitoring Group Master Data Entry Template</td>
<td>Monitoring Group Lead or designated appointee</td>
<td>Monitoring Program Coordinator</td>
</tr>
<tr>
<td>Final Monitoring Report</td>
<td>Monitoring Program Coordinator</td>
<td>All signatories of this QAPP</td>
</tr>
<tr>
<td>Final Monitoring Data</td>
<td>Monitoring Program Coordinator</td>
<td>Signatories on this QAPP, EPA, NYS DEC, CTDEEP, NYCDEP, and other management groups</td>
</tr>
</tbody>
</table>

### D. Data Validation and Usability

#### D.1. DATA REVIEW, VERIFICATION, AND VALIDATION

All project data, metadata, and quality control data shall be critically reviewed to look for problems that may compromise data usability.

Data collected before the 2018 season will be flagged as not being conducted under this QAPP when distributed. Save the Sound will be tasked with maintaining this QAPP in all aspects for the duration of the Unified Water Study.

The Monitoring Group Lead or designated appointee will review field data after each sampling run and take corrective actions as described in Table 16 of this document. At least once during the season, at the end of the season, and if questions arise, the Monitoring Group Lead or designated appointee will share the data with the UWS Quality Assurance Officer to determine if the data appear to meet the objectives of the QAPP. Together, they will decide on any actions to take if problems are found.

#### D.2. VERIFICATION AND VALIDATION METHODS

All project data and metadata are reviewed and approved as usable data, or as un-useable data.

Data verification and validation will occur as described in Table 16, and will include checks on:

- Completion of all fields on data sheets; missing data sheets
Completeness of sampling runs (e.g. number of stations visited / samples taken vs. number proposed, were all parameters sampled / analyzed)
- Completeness of QC checks (e.g. number and type of QC checks performed vs. number or type proposed)
- Number of samples exceeding QC limits for accuracy and precision and how far limits were exceeded.

D.3. RECONCILIATION WITH USER REQUIREMENTS

At the conclusion of the sampling season, after all in-season quality control checks, assessment actions, validation and verification checks and corrective actions have been taken, the resulting data set will be compared with the program’s data quality objectives (DQOs) as defined in section A.7. This review will include, for each parameter, calculation of the following:

- Completeness goals: overall % of samples passing QC tests vs. number proposed.
- Percent of samples exceeding accuracy and precision limits.
- Average departure from accuracy and precision targets.

After reviewing these calculations, and taking into consideration such factors as clusters of unacceptable data (e.g. whether certain parameters, stations, dates, monitoring groups, etc. produced poor results), the Monitoring Program Coordinator, Quality Assurance Officer, and respective Monitor Group Lead will evaluate overall program attainment of DQOs and determine what limitations to place on the use of the data, or if a revision of the DQOs is allowable.
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UWS Standard Operating Procedure – Station Selection

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1 POINT OF CONTACT

NAME: Peter Linderoth, Save the Sound / CFE, Water Quality Program Manager
ADDRESS: 545 Tompkins Ave, 3rd Floor, Mamaroneck, NY 10543
EMAIL: plinderoth@savethesound.org
PHONE: 914-263-6233

2 OBJECTIVE

Select sample locations to represent the entire embayment in a statistically sound manner. The Unified Water Study (UWS) will assign unique ID codes for the embayment, reporting sections of the embayment, and stations.
3 OVERVIEW

Sample stations are selected to represent the water quality of the entire embayment using a probability-based sampling design [Paul et al., 2003], as in the EPA National Coastal Assessment (EPA, 2003). A UWS Scientific Advisor and UWS Monitoring Program Coordinator advise on the choice of station locations. Historic sites are evaluated for bias and inclusion in the UWS.

Station selection occurs only once, at the time of an embayment joining the UWS. Once stations are determined, these should be consistent in future years. Re-evaluation of stations in future years should be conducted under advisement with the UWS Monitoring Program Coordinator and the UWS Science Advisors.

Details on sampling frequency and timing during a day are covered in the UWS SOP Sampling Plan. The information below provides a general overview of the timing of sampling at the different stations.
4 SOURCES

This station selection design is based on methods used in the EPA National Coastal Assessment (EPA, 2001).

5 MATERIALS AND EQUIPMENT

- nautical charts of embayment or local knowledge of typical depth contours
- ArcGIS, access through the UWS Monitoring Program
- UWS subwatershed and embayment delineations for general reference
- hexagonal tessellation grid for use in ArcGIS

6 METHODS

6.1 Water Quality Station Selection

The location of stations will be determined or verified using a probability-based sampling design (Paul et al., 2003), as in the EPA National Coastal Assessment (EPA, 2001). This approach allows for the use of probability-based statistics in data analysis, without bias introduced by non-random station choice.

In this approach, a 0.42 km² hexagonal grid is overlain on the site map (Figure 2).

A minimum of four stations will be sampled in each embayment. If an embayment is divided into reporting regions, a minimum of three sites will be selected within each reporting region.

In cases where many hexagons are located in the embayment, sampling of some hexagons may be eliminated, if they are likely to be similar to neighboring stations. Exclusion of hexagons from the sampling plan should only be undertaken under advisement with the UWS Monitoring Program Coordinator and UWS Scientific Advisors. Nine stations per embayment are considered reasonable. Twelve stations and above are likely candidates for eliminating hexagons from the sampling plan. An example of eliminated hexagons is provided for Little Neck Bay, NY (Figure 2).

When a monitoring group has existing stations, those stations may be evaluated by the UWS Monitoring Program Coordinator and UWS Scientific Advisors for inclusion in the study.
6.1.1 Assigning New Stations

Three stations are randomly generated for each hexagon targeted for sampling, identified in order of preference from low value to high. Stations will not be allowed in the center of navigational channels or in unnavigable waters. Stations will not be situated close to point sources of nutrients; this will be determined by consulting maps, local knowledge, and an on-site assessment on the first field day. The random generation of stations is conducted by the UWS Monitoring Program Coordinator in ArcGIS.

The final location of the station in each hexagon will be determined in the field. A trial sampling run is recommended to verify the stations chosen are accessible and can be sampled in the required timeframe (for timeframe, see UWS SOP Sampling Plan). From the three random stations, a Monitoring Group may determine a station is unsuitable based on navigation considerations (too shallow, too much current to anchor, too rocky, etc.). If the first station is considered unsuitable, the field leader, UWS Monitoring Program Coordinator, and UWS Scientific Advisor, will evaluate the second, then third station. If all are deemed unacceptable, the collective group may choose a station within the hexagon that is close to the first random station with above considerations reviewed.

Once chosen, the station location is considered the permanent location and will not change on future sampling dates. Because the station will be permanent, the field leader should pay particular attention to the station depth in relation to the stage of the tide; the goal should be to have enough water at low tide to be able to sample the location.
6.1.2 Evaluating Existing Stations

Existing stations in each hexagon will be evaluated for any potential biases that are present. These can include proximity to nutrient sources, disproportional sampling of beach sites, or other targeted sampling locations. Stations that are unbiased in a hexagon will be the first options for inclusion in the study. Stations deemed biased will not be included in the UWS Monitoring.

6.2 Macrophyte Station Selection

The number of stations sampled per embayment depends upon the size of the embayment, expected variability in macroalgae coverage, and access to sampling sites. An initial site visit of many locations can aid in determining the viability of sampling stations. A minimum of 3 sample stations is recommended for all embayments. More may be needed for larger embayments. The UWS Monitoring Program Coordinator, UWS Scientific Advisor, and Monitoring Group will consult to select these stations.

Station selection for macrophytes is complex. Refer to the UWS SOP Qualitative Macrophyte for this process. Macrophyte sampling occurs at different time scales and different stations than water quality parameters.

6.3 GPS Coordinate Reference Station

Select one land-based station to assess the precision of your GPS device. This station does not need to be located near the embayment being sampled. This is a land-based station that is easily accessible to the person with the GPS device.

The GPS coordinates at this station are read within 1 days of a field sampling day.

Each GPS unit or smart phone used for GPS must collect GPS coordinates form a land reference station.

7 TROUBLESHOOTING / HINTS

Station selection should be conducted in coordination with the UWS Monitoring Program Coordinator and UWS Scientific Advisors. A monitoring group is not expected to select stations without assistance.

8 DATA PROCESSING AND STORAGE

The UWS Monitoring Program Coordinator will be the custodian of the initial station selection maps and final maps determined after the first complete field day. The UWS Monitoring Program Coordinator will maintain a database which includes the unique site codes, section codes, and station codes for the embayment. Each unique station code will be affiliated with the corresponding GPS for the station.

The monitoring group is responsible for obtaining the GPS coordinates for each station during sample events.

The monitoring group is responsible for assuring that the correct unique station ID assigned by the UWS is properly matched with the local organizations station ID codes. Both codes (monitoring group’s
REFERENCES


APPENDIX - LIST OF EMBAYMENT CODES

For record keeping purposes, each embayment is assigned a unique three letter code for the UWS. Contact the UWS Monitoring Program Coordinator to verify the names of specific waterbodies. Site names in this list may differ from the local names used for a site.

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1 POINT OF CONTACT

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PHONE: 914-263-6233

2 OBJECTIVE

Sample water quality and macrophytes within an embayment following guidelines of the Unified Water Study (UWS). Frequency of sampling and daily order of events are specified.

3 DEFINITIONS AND ABBREVIATIONS

Embayment: A recess in a coastline or an indentation off a shoreline which forms a bay. In Long Island Sound, the names of embayments often include the words Harbor, River, Cove, Bay, Creek; with a few including the names Brook, Gut, Inlet, or Lake.

Field Team: Person or group of people working together to sample a station.

Macroalgae (macroalgae, singular:): Commonly referred to as seaweed. This is a group of plant-like organisms. They do not have the vascular system and roots of a true plant. The “macro” prefix indicates these organisms are visible with the naked eye, no magnification is required to view the whole organism; although, magnification with a hand lens or loupe may be necessary to view the structure of the organism. In comparison, microalgae are the phytoplankton in the water which are too small to see with the naked eye.

Macrophyte: Plants and macroalgae that are viewable with the naked eye. This term includes macroalgae, seagrass (eelgrass, Zostera marina; widgeon grass, Ruppia maritima), and marsh grass.

Monitoring Group: The group conducting the field work.

Seagrass: A true plant, not an alga; they have the vascular system and roots of a land plant. These plants are fully submerged at all times (though there are a few species not found in Long Island Sound which are intertidal). Long Island Sound has two species of seagrass: Zostera marina (eelgrass), which is the most commonly seen seagrass in our area; and Ruppia maritima (widgeon grass).

Region: The reporting regions for the UWS. Each section must include a minimum of three stations. Sections will be assigned a unique name by the UWS; examples are included below.

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<th>Number of Sections</th>
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Site: The whole embayment, as defined by the UWS list. Each site has a unique three letter code assigned by the UWS; for example, Little Neck Bay, NY is “LNE”. Each site will have a minimum of four water quality stations.

Sonde: An instrument probe that automatically transmits information about its surroundings underground, under water, in the atmosphere, etc.
4 OVERVIEW

Sampling occurs in the months of May through October, though a shorter season is acceptable for inclusion in the UWS.

Two types of stations are included:

1) Water quality stations are sampled within three hours of sunrise between the months of May through October. A minimum of four stations per embayment are required. If multiple regions of the embayment are delineated, a minimum of three stations are required per region. Monitoring groups will plan to sample water quality stations at a minimum every two weeks. However, if unforeseeable circumstances make this plan not possible, a minimum of six sample events between May and October are required for inclusion in the UWS; including at least one sample event in each of the months of June, July, August, and September.

2) Macrophyte stations are land-based or boat-based. They are sampled only mid-summer and may be sampled on different days from the water quality stations. Sampling occurs on three separate days between July 15 and August 7. Two sample days or a date slightly outside of the date criteria may be sufficient in the event of unforeseen complications. This decision will be made by the Monitoring Program Quality Assurance Officer and UWS Science Advisor(s).

5 SOURCES

These procedures are based on the EPA Volunteer Estuary Monitoring Manual (EPA, 2007) and follows methods used in the EPA National Coastal Assessment (EPA, 2001). The EPA Volunteer Estuary Monitoring Manual (EPA, 2007) provides a wealth of specific data for monitoring groups. All groups should refer to the EPA manual for specific guidance. This SOP provides specific monitoring details relevant to the UWS.

6 MATERIALS AND EQUIPMENT

6.1 Safety

- Safety plan – every volunteer should have a copy
  - Find out the location and telephone number of the nearest telephone and write it down, or have a cellular phone available.
  - Locate the nearest medical center and write down directions for guiding emergency
personnel to your stations.
- Have each member of the sampling team complete a medical form that includes emergency contacts, and relevant health information such as allergies, diabetes, epilepsy, etc. **Please note** – this form should be kept confidential. Whoever coordinates the Monitoring Group efforts should have a copy and should review the form. A second copy should be kept with the field team in the event of an emergency; the form for each team member should be sealed in an envelope with their name.
- Each team member should have contact information for all field team members. This list could be kept in your field box or use packing tape to affix it to the back of a clipboard or some other item you always have in the field.
  - full name
  - cell phone
  - home phone
  - email address
  - car color, make, model, and license plate
  - emergency contact information
- Every monitoring team should have a shore-based check-in for each sampling day. This is a person who knows who is on the team, where they will be, and the time they are expected back. If the field team does not check in, the shore person should know the procedures to follow to report the team missing. The following is an example:
  - Call the cell phones and home phones of all team members.
  - Check the site for parked vehicles – if the vehicle is present, start a search.
  - If the team is considered missing at sea, contact the Coast Guard; if the team is missing on land, contact the police.
- first aid kit – suggested items
  - telephone numbers of emergency personnel (e.g., police, ambulance service)
  - first aid manual which outlines diagnosis and treatment procedures
  - antibacterial or alcohol wipes
  - first aid cream or ointment
  - acetaminophen and ibuprofen for relieving pain and reducing fever
  - several band-aids
  - several gauze pads, 3 or 4 inches square
  - 2-inch roll of gauze bandage
  - triangular bandage
  - large compress bandage
  - 3-inch wide elastic bandage
  - needle for removing splinters
  - tick spoon for removing ticks
  - doctor-prescribed antihistamine for any participant who is allergic to bee stings
- cell phone
- water
- appropriate shoes and clothes (and extras); protection from the sun
- other items to consider:
  - flash light
  - back pack for sampling equipment, so hands are free
6.2 Sampling Gear – All Stations

REQUIRED (PUT IN A SMALL TOOL BOX)

- site maps with stations on map
- list of UWS unique station IDs with GPS coordinates for the site
- clip boards
- pencils
- pencil sharpener
- permanent marker
- field data sheets
- Chlorophyll a labels

OPTIONAL, BUT VERY USEFUL (PUT IN A SMALL TOOL BOX)

- clear packing tape (can be used to cover writing on bottles or affix a label that is not sticking)
- extra batteries for any electrical sampling gear
- tailor’s tape measure (seamstress measuring tape), with metric scale – comes in handy when you need to re-mark a line
- basic tools (pliers, wrench, screw drivers, etc.)
- plastic baggies
- scissors, pocket knife, nail clippers
- cable ties
- electrical tape (this tape works well underwater; comes in many colors)
- duct tape (very useful in many situations)
7 METHODS

7.1 Parameters to Sample

Monitoring groups must collect the following data to be included in the UWS:

- for each water quality station
  - GPS coordinates of stations, recorded each sample date
  - date and time
  - total water depth
    - 0.5 m below the surface, 0.5 m above bottom, mid-depth if total depth >10 m; if total depth is less than 1.5 m, only a mid-depth reading will be collected
    - temperature
    - conductivity (salinity)
    - dissolved oxygen
    - chlorophyll a
    - turbidity
  - at the last station per field day undertake a replicate profile including all parameters
- obtain from an online NOAA tide table and noted weather station:
  - time of high and low tide nearest time of sampling
  - high and low air temperature for 24 hours preceding field sampling
  - precipitation out a week preceding sampling event
- within 1 day of the field sampling day, read the GPS of a land-based reference station

- for each macrophyte station
  - GPS coordinates of stations
  - date and time
  - photos of macrophytes
- within 1 days of the field sampling day, read the GPS of a land-based reference station

7.2 Timing of Sampling

7.2.1 Timing During the Year

WATER QUALITY STATIONS

The target sampling frequency is two sampling events per month, May through October. Sampling dates should be 10 to 18 days apart.

All water quality parameters should be sampled during a sampling event. At one station per field day, conduct a replicate profile.

In the occurrence of equipment failure or other unforeseen difficulty, a minimum of six sample events between May and October are required for inclusion in the UWS; including at least one sample event in each of the months of June, July, August, and September.
MACROPHYTE SURVEY SAMPLING FREQUENCY

Macrophyte surveys will occur between July 15 and August 7 of every sampling season, with 3 survey events total. One survey per week is best. If this is not possible, maximize the days between sampling. All three days cannot be sampled in the same 7-day window. 2 sampling events or one outside of the interval may be considered for inclusion in the UWS. This decision will be made by the Monitoring Program Quality Assurance Officer and UWS Science Advisor(s).

OVERVIEW OF SAMPLING FREQUENCY

The calendar below provides an example of sampling frequency throughout the season. Dates highlighted in orange are water quality sampling dates (e.g. May 11). The green text (July 15 – August 7) are potential dates for macrophyte sampling. The dates highlighted in green are the 3 planned macrophyte sampling dates (e.g. July 21).

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<td>26 27 28 29 30 1 2</td>
<td>25 26 27 28 29 30 1</td>
<td>25 26 27 28 29 30 1</td>
<td>1 2 3 4 5 6 7</td>
<td>3 4 5 6 7 8 9</td>
</tr>
</tbody>
</table>

7.2.2 Timing During a Sample Day

Sampling of dissolved oxygen occurs in the morning (within 3 hours of sunrise), to capture the lowest
UWS Standard Operating Procedure
Tier I Sampling Plan

Chlorophyll a and turbidity should be sampled at the same time as dissolved oxygen, to allow for linking of these parameters to their corresponding temperature, salinity, and dissolved oxygen (Tables 1).

Table 1: Text summary of required sampling times within a sample day

<table>
<thead>
<tr>
<th>Parameter/Survey</th>
<th>Required time interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dissolved Oxygen (mg/L, % sat.)</td>
<td>Collected within 3 hours of sunrise (with dissolved oxygen)</td>
</tr>
<tr>
<td>Temperature</td>
<td>Collected within 3 hours of sunrise (with dissolved oxygen)</td>
</tr>
<tr>
<td>Salinity</td>
<td>Collected within 3 hours of sunrise (with dissolved oxygen)</td>
</tr>
<tr>
<td>Chlorophyll a</td>
<td>Collected within 3 hours of sunrise (with dissolved oxygen)</td>
</tr>
<tr>
<td>Turbidity (NTU)</td>
<td>Collected within 3 hours of sunrise (with dissolved oxygen)</td>
</tr>
<tr>
<td>Total Sample Station Depth</td>
<td>At time of sampling</td>
</tr>
<tr>
<td>Macrophyte Survey</td>
<td>Close to low tide for wrackline survey, close to high tide for hardened shorelines, boat surveys any time of day</td>
</tr>
</tbody>
</table>

7.3 Sampling Depths

7.3.1 Temperature, Salinity, Dissolved Oxygen, Chlorophyll a, Turbidity

If total depth at the station is greater than 10 m, take three samples: one at 0.5 m below the surface, one at mid depth, and another at 0.5 m above the bottom.

If total depth at the station is less than 10 m, take two samples: one at 0.5 m below the surface and the other at 0.5 m above the bottom.

If total depth at the station is less than 1.5 m, take one sample halfway to the bottom and record as mid depth.

Always record the depth of the sample.

7.4 Required Replicates and Verification

During a field day, use the field data sheet as a reminder for the number of replicates required for each parameter.

Table 2: Required replicates, blanks, and verification readings.
<table>
<thead>
<tr>
<th>parameter &amp; technique</th>
<th>replicates required</th>
<th>verification and/or blank</th>
</tr>
</thead>
<tbody>
<tr>
<td>GPS coordinates</td>
<td>1 reading per station at 1 station per day, take a second reading before leaving the station</td>
<td>read a land-based reference station within 2 days of the field sampling day</td>
</tr>
<tr>
<td>sampling with multiparameter sonde</td>
<td>1 reading at each depth, wait for reading to stabilize before recording at 1 station per day (typically the last station), do two replicate profiles – do one complete profile, then do a second</td>
<td>verify depth by lowering sonde to known depth verify Chlorophyll a by reading water in a bucket and filtering 2 samples from the bucket read standards before and after a sample day (salinity, oxygen in 100% water saturated air, turbidity, chlor-a); day before and after sample day is acceptable</td>
</tr>
<tr>
<td>filtered Chlorophyll a</td>
<td>2 filtered samples of a single water sample collected from 0.5 m below the surface of all stations</td>
<td>duplicate from same bucket used as a second calibration point for Chlorophyll a for a sampling event</td>
</tr>
<tr>
<td>macrophytes</td>
<td>3-6 stations per embayment</td>
<td>photos of each rake toss or the beach being sampled, reviewed by UWS Monitoring Program Coordinator and Science Advisors</td>
</tr>
</tbody>
</table>
7.5 Order of Events When Sampling a Water Quality Station

7.5.1 Prepare for Sampling Trip

A. Calibrate all instruments.
B. Gather all field supplies.
C. Complete the pre-sampling event portions of the UWS datasheet.
D. Arrange for a shore person. This is someone to check on you if you don’t come back at the designated time.

7.5.2 Water Quality Station Sampling – within 3 hours of sunrise

A. Record station information on the data sheet. **Be sure to complete all sections of the data sheet completely, for every data entry.**
B. Obtain total depth of the station, determine sampling depths for water quality parameters
C. Collect profile data using the multiparameter sonde.
   a. Sample the surface, then mid-depth, then bottom with Sonde; or depths necessary according to total depth and respective sample depth criteria, sampling from top to bottom.
D. At one station per day repeat measurements where only one profile is typically collected. The last station of the day is the ideal choice in terms of time management.
   a. Repeat the sonde profile. Complete the first profile, sampling at each depth. Complete a second profile.
   b. Record the depth and GPS coordinates a second time, just before leaving the station.
E. Read a calibration standard just following calibration and following the field trip to verify readings, for all parameters where this applies.

7.5.3 End of Field Day

A. If using a multiparameter sonde to sample Chlorophyll a, place the sonde in a bucket of estuarine water, read the sonde, and filter samples for Chlorophyll a analysis for verification of the sonde readings.
B. Verify all sections of the data sheet have been completed.
C. Store Chlorophyll a samples according to the UWS SOP Filtered Chlorophyll.
D. Within 1 days of sampling, record the GPS of a reference station on land to verify the accuracy and precision of the GPS coordinates.

8 TROUBLESHOOTING / HINTS

➢ Gather field equipment the day prior to sampling. Check the field equipment in the morning, before
you head out into the field. Creating a checklist is very helpful in prepping for your field day. Include personal items (sunscreen, bug spray, etc.) and safety equipment on the checklist.

- Always carry a copy of this SOP and the relevant parameter-specific SOPs.
- Print out the “quick sheets” for relevant SOPs to use as a reminder in the field. Do not laminate these as you will want to add notes. A plastic page-protector taped close can be used to keep these sheets dry.

9 DATA PROCESSING AND STORAGE

The UWS Monitoring Program Coordinator will be the custodian of the finalized data files.

The monitoring group is responsible for obtaining data, entering data into the UWS data template, and delivering the data to the UWS Monitoring Program Coordinator.

The monitoring group is responsible for assuring that the correct unique station ID assigned by the UWS is properly matched with the local organizations station ID codes. Both codes (monitoring group’s station code and UWS unique station ID) will be entered into the data template, along with the GPS coordinates.

10 REFERENCES


11 Quick Sheet – Tier I Sampling Plan

Check your equipment list the day before a field day and again on the morning of a field day.

Calibrate instruments the day before sampling.

Read a calibration standard just following calibration and following the sampling event to verify readings, for all parameters where this applies.

Bring the relevant SOPs and quick sheets with you into the field.

Remember – rinse equipment in sample water before collecting a sample.

Water quality stations – Sample depths are shown in chart to the right.

Use the field data sheet as a reminder for the number of replicates required for each parameter. Sample 1 station/day as a replicate for sonde profiles, GPS, and depth.

For filtered Chlorophyll a, filter four samples per day with corresponding sonde reading.

MACROPHYTE STATIONS – use quick sheet from Macrophyte SOP

GPS REFERENCE CHECK

- Within 1 day of sampling, record the GPS of a reference station on land to verify the accuracy and precision of the GPS coordinates.

WATER QUALITY STATIONS (sample within 3 hours of sunrise)

- At one station per day repeat measurements where only one profile is typically collected. The last station of the day is a good choice in terms of time management. (sonde, GPS and depth)
Appendix A-3

UWS Standard Operating Procedure – Depth and GPS Coordinates

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1 POINT OF CONTACT

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ADDRESS: 545 Tompkins Ave, 3rd Floor, Mamaroneck, NY 10543
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PHONE: 914-263-6233

2 OBJECTIVE

Determine the total depth and sample depth at station. Determine the GPS coordinates of the station.

3 OVERVIEW

A graduated line is weighted and used for recording total depth (m) and sample depth (m) at a station. Acceptable alternatives include multiparameter sonde depth reading.
A GPS unit is used to obtain GPS coordinates for the station on each field day. A Smart Phone app is an acceptable alternative as backup. GPS position of your actual location at each station for each sample date is required. A second reading is taken at one station per embayment per sample day to assess precision. If unforeseen circumstances make recording GPS points impossible and dead reckoning is used, your data may still be used as part of the UWS but will be flagged and reviewed in the quality assurance process.

4 SOURCES


5 MATERIALS AND EQUIPMENT

- Depth sampling device. Options include:
  - weighted, graduated line, with divisions every 0.1 m
    - This can be the cable attached to a multiparameter sonde
  - multiparameter sonde with depth sensor
- GPS coordinate sampling device. Options include:
  - Handheld GPS unit
  - Apps are available for your smart phone that allow you to easily log and return to GPS coordinates. The accuracy of the GPS on your phone is dependent on Wi-Fi signals and cellular signals.
  - Dead reckoning to get yourself on station should only be used if your GPS options have failed. This will need to be included in field notes for the sampling event. Data will be reviewed in the quality assurance process.

6 METHODS

6.1 Preparation

- Weighted line:
  - Examine the line for wear or damage.
  - Measure the increments to ensure line has not stretched or shrank.
- If using a multiparameter sonde:
  - Calibrate the depth sensor prior to the sampling event and the morning of on the sampling vessel according to the manufacturer’s instructions.
UWS Standard Operating Procedure
Depth & GPS Coordinates

GPS device

- Check batteries
- Within 1 day of the sampling trip, read the GPS coordinates of a reference station on land (refer to “UWS Station Selection SOP” for more details on this reference station). These readings will be used to assess the precision of your GPS method.

6.2 Field Collection and Processing

**Metered Line**

1. Attach weight to line.
   a. A weighted sonde attached to a cable with 0.1 m increments is considered a weighted line.
2. Slowly lower line until the weight reaches the bottom.
3. Mark the line at the surface of the water (pinch with finger).
4. Determine the depth of the station.
5. Record max depth to the nearest 0.1 m on field data sheet.
6. Following UWS SOP Sampling Plan for number of samples and locations in the water column associated with total depth, sample all target depths using steps 2-5 replacing “weight reaches the bottom” to deliver sonde to target depth using increments on cable.
7. At least once per day per embayment (usually the last station of the day), take the reading again.

**Multiparameter Sonde**

1. Slowly lower until the weighted sonde reaches the bottom.
2. Determine the depth of the station by reading the display.
   a. Add the length from the depth sensor to the weight for total depth.
3. Record max depth to the nearest 0.1 m on field data sheet.
4. Following UWS SOP Sampling Plan for number of samples and location in the water column associated with total depth, sample all target depths using steps 1-3 replacing “weight reaches the bottom” to deliver sonde to target depth using handheld display. If depth sensor is not located with sonde attachments, compensate by lowering the sonde to the depth reading required. For example, if the sonde attachments are .25 m below the depth sensor and the target sample depth is 5 m the sampler should lower sonde to a depth reading 4.75 m on the display.
5. At least once per sampling day (prior to first station), lower the sonde to a known depth and verify the reading. If the depth reading from sonde does not agree within 0.3 m, use the metered line procedures.
6. At least once per sampling day per embayment (usually the last station of the day), take the reading again.
GPS DEVICE

1. Determine the GPS coordinates of the station by reading the display.
2. At least once per sampling day per embayment (usually the last station of the day), take the reading again.
3. Within 1 day of the sampling trip, read the GPS coordinates of a reference station on land.

6.3 Sample Storage

Not applicable.

6.4 Laboratory Analysis

Not applicable.

7 TROUBLESHOOTING / HINTS

- Always carry a copy of this SOP and the relevant parameter-specific SOPs.
- Print out the “quick sheets” for relevant SOPs to use as a reminder in the field. Do not laminate these as you will want to add notes. A plastic page-protector taped close can be used to keep these sheets dry.

8 DATA PROCESSING AND STORAGE

The monitoring group is responsible for obtaining data, entering data into the UWS data template, and delivering the data to the UWS Monitoring Program Coordinator.

The monitoring group is responsible for assuring that the correct unique station ID assigned by the UWS is properly matched with the local organizations station ID codes. Both codes (monitoring group’s station code and UWS unique station ID) will be entered into the data template, along with the GPS coordinates.

9 REFERENCES

10 Quick Sheet – Depth & GPS Coordinates

*For depth and GPS - At least once per sampling day per embayment (usually the last station of the day), take the reading again.*

**Preparation**

- If using a weighted line:
  - Examine the line for wear or damage.
  - Measure the increments to ensure line has not stretched or shrank.

- If using a multiparameter sonde:
  - Calibrate the depth sensor according to the manufacturer’s instructions. This may be done in the field.
  - Bring a weighted line (can be sonde cable) for verification of readings (prior to sampling first station) and in case sonde depth is inaccurate.

- GPS device
  - Check batteries
  - Within 1 days of the sampling trip, read the GPS coordinates of a reference station on land. This will be used to assess the precision of your GPS method.

**Field Collection and Processing**

**Metered Line**

1. Attach weight to line.
   a. A weighted sonde attached to a cable with 0.1 m increments is a weighted line.
2. Slowly lower line until the weight reaches the bottom.
3. Mark the line at the surface of the water (pinch with finger).
4. Determine the depth of the station.
5. Record max depth and sample depth(s) to the nearest 0.1 m on field data sheet.

**Multiparameter Sonde**

1. Determine the depth of the station by reading the display.
2. Record max depth and sample depth(s) to the nearest 0.1 m on field data sheet.
3. At least once per sampling day (prior to first station), use a metered line to confirm the accuracy of the depth reading. If the depth reading from sonde does not agree within 0.3 m, use the metered line.

**GPS Device**

1. Determine the GPS coordinates of the station by reading the display.
2. Within 1 days of the sampling trip, read the GPS coordinates of a reference station on land.
Appendix A-4

UWS Standard Operating Procedure – Sonde Profile

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1 POINT OF CONTACT

NAME: Peter Linderoth, Save the Sound / CFE, Water Quality Program Manager
ADDRESS: 545 Tompkins Ave, 3rd Floor, Mamaroneck, NY 10543
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PHONE: 914-263-6233

2 OBJECTIVE

Determine the sample depth (m), temperature (°C), salinity (psu), dissolved oxygen concentration (mg/L = ppm), and dissolved oxygen percent (% sat.) in the surface (0.5 m below surface) and bottom water (0.5 m above bottom) at the station using a multiparameter sonde. When total water depth is greater than 10 m, parameters at mid-depth will also be determined.
3 OVERVIEW

A multiparameter sonde is used to determine the sample depth (m), temperature (°C), salinity (ppt), dissolved oxygen concentration (mg/L = ppm), and dissolved oxygen percent (% sat.) in the surface (0.5 m below surface) and bottom water (0.5 m above bottom) at the station. When total water depth is greater than 10 m, parameters at mid-depth will also be determined. Multiparameter sondes may also include probes for turbidity (NTU) and chlorophyll a (RFU, µg/L). Frequency of sampling and daily order of events are specified in the “UWS Sampling Plan SOP.”

At one station per day per embayment (typically the last station), do two replicate profiles – do one complete profile, then do a second. This allows for the precision of the sonde to be assessed.

4 SOURCES

These procedures are based on the EPA Volunteer Estuary Monitoring Manual (EPA, 2007) and follows methods used in the EPA National Coastal Assessment (EPA, 2001). The EPA Volunteer Estuary Monitoring Manual (EPA, 2007) provides a wealth of specific data for monitoring groups. All groups should refer to the EPA manual for specific guidance.

5 MATERIALS AND EQUIPMENT

- Eureka Manta +35 or YSI EXO1 Multiparameter Sonde, setup to record:
  - depth
  - temperature
  - conductivity (salinity)
  - dissolved oxygen
  - chlorophyll a
  - turbidity
- 5 gallon bucket
- See UWS SOP Filtered Chlorophyll. The field team will need all equipment and material listed for collecting chlorophyll a samples. Filter two samples per field day for verification of sonde readings.
- ASTM Type I reagent grade water or equivalent for 0 NTU turbidity standard solution and 0 µg/L (and 0 RFU for YSI EXO1) for chlorophyll a calibration
- YSI Turbidity Standard, 607300 (124 FNU on YSI/100 NTU on Eureka) (YSI SKU: 607300)
- YSI Conductivity Calibrator Solution, 3169: 50,000 µS/cm (YSI SKU: 060660)

6 METHODS

6.1 Preparation

- Check the battery power. Pack extra batteries in the field tool kit.
- Calibrate the sonde the day before or morning of each field day. Follow the manufacturer’s instructions for calibration steps.
Conductivity – Conduct calibration using the 50,000 μS/cm conductivity standard.

Dissolved Oxygen – Conduct an air calibration, using water saturated air for calibrating 100% saturation.

Chlorophyll a - Conduct a one-point calibration using the 0 μg/L (ASTM Type I reagent grade water)

Turbidity - Conduct a two-point calibration using the 0 NTU (ASTM Type I reagent grade water) and 124 FNU (100 NTU Eureka instrument) standard solutions.

Depth – Calibrate the depth following manufacturer’s guidelines.

The instrument will be factory calibrated and serviced at the manufacturer’s recommended interval.

- After each calibration, take a verification reading in each of the standard solutions. This can be done directly after the calibration with solution still in the calibration cup.

- Record calibration information in the calibration section of the UWS datasheet. The following information will be entered into the UWS data entry template:
  - dissolved oxygen - post calibration reading in 100% saturated air
  - conductivity standard for sonde - manufacturer and value (μS/cm)
  - conductivity standard for sonde - lot number
  - conductivity standard for sonde - expiration date
  - conductivity on sonde - post calibration reading in standard (μS/cm)
  - chlorophyll a fluorescence on sonde - post calibration reading (μg/L on Eureka and μg/L, RFU on YSI)
  - turbidity standard - manufacturer and value (FNU/NTU)
  - turbidity standard - lot number
  - turbidity standard - expiration date
  - turbidity - post calibration reading in standard (FNU/NTU)

6.2 Field Collection and Processing

6.2.1 Sampling at a Station

- Turn on the sonde.

- If sampling from a boat, anchor the boat or stay on station with short engine adjustments.

- Determine sampling depths as described in the UWS SOP Sampling Plan.

- If wading in to sample, take extra precaution to not stir up sediment. Samples should be taken from an area that is not disturbed from the sampler’s approach.

- Lower sonde to sample depth. Be sure that the probe ends are at the desired depth, versus other sections of the sonde. Be careful not to let the probe contact the sediments as this could foul the
probes and give inaccurate readings.

- Allow the sonde to equilibrate for at least 90 seconds. Watch the salinity first. Once salinity is steady, watch the dissolved oxygen. The value may fluctuate in response to real variability in the water column. You are waiting for the readings to hover around a value, versus consistently rising or falling. Do not wait for turbidity or chlorophyll a to stabilize, these probes respond quickly to real variability in the water column. These values should be listed as the middle point between the high and low displayed back and forth on the screen.
- Record readings on the field data sheet.
- At one station per day per embayment (typically the last station), do a replicate profile – do one complete profile, then do a second. This allows for the precision of the sonde to be assessed.

6.2.2 Verification of Water Quality Parameters and Sonde Depth

- Take and record sonde values in standards up to a day before and after a sampling day (conductivity, oxygen in 100% water saturated air, 0 µg/L chl-a and 124 NTU (YSI) or 100 NTU (Eureka) turbidity.
- Verify sonde depth by lowering sonde to a known depth following the procedures listed in UWS SOP Depth SOP.

6.2.3 Verification of Chlorophyll a – Bucket Sample

If a chlorophyll a probe is on the sonde, verify the chlorophyll a readings from the sonde once per day and send to an analytical lab for analysis. This may be conducted at a dock; it does not need to occur at a water quality station.

- Rinse the bucket three times with surface water.
- Fill the bucket with surface water.
- Place the sonde in the bucket and stir for 10 seconds, allow to equilibrate, record chlorophyll a reading.
- Filter two samples from the bucket following the procedures in the UWS SOP Filtered Chlorophyll. Prior to withdrawing a sample from the bucket, be sure to stir the water in the bucket, using the sonde.

6.3 Sample Storage

Not applicable for the sonde. Follow instructions in the UWS SOP Filtered Chlorophyll SOP for filtered chlorophyll a samples.

6.4 Laboratory Analysis

Not applicable for the sonde. Follow instructions in the UWS SOP Filtered Chlorophyll SOP for filtered chlorophyll a samples.
7 TROUBLESHOOTING / HINTS

➢ Always make sure to wait until readings have stabilized before calibrating or logging a reading.

➢ If the probe does not appear to be operating or calibrating properly, contact the Monitoring Group Lead to make a decision on proceeding with the sampling event. Immediately notify the UWS Monitoring Program Coordinator if a sonde continues to not operate properly.

8 DATA PROCESSING AND STORAGE

The UWS Monitoring Program Coordinator will be the custodian of the finalized data files.

The monitoring group is responsible for obtaining data, entering data into the UWS data template, and delivering the data to the UWS Monitoring Program Coordinator.

The monitoring group is responsible for assuring that the correct unique station ID assigned by the UWS is properly matched with the local organizations station ID codes. Both codes (monitoring group’s station code and UWS unique station ID) will be entered into the data template, along with the GPS coordinates.

9 REFERENCES


10 Quick Sheet – Sonde Water Quality Profile

**Preparation**
- Check the battery power. Pack extra batteries in the field tool kit.
- Calibrate the sonde the day before each field day. Follow the manufacturer’s instructions for calibration.
- After calibrating, use the sonde to take a verification reading in each of the standard solutions.
- Record calibration information in the calibration section of the UWS datasheet.

**Field Collection and Processing**
- Turn on the sonde.
- If sampling from a boat, anchor the boat or stay on station with short engine adjustments.
- Determine sampling depths as described in the UWS SOP Sampling Plan.
- If wading in to sample, take extra precaution to not stir up sediment. Sample should be taken from an area that is not disturbed from the sampler’s approach.
- Lower sonde to sample depth. Be sure that the probe ends are at the desired depth, versus other sections of the sonde. Be careful not to let the probe contact the sediments as this could foul the probes and give inaccurate readings.
- Allow the sonde to equilibrate for at least 90 seconds.
  - Watch the salinity first.
  - Once salinity is steady, watch the dissolved oxygen.
  - The value may fluctuate in response to real variability in the water column. You are waiting for the readings to hover around a value, versus consistently rising or falling. This will typically take about one minute, but may take longer. Do not wait for turbidity or chlorophyll a to stabilize, these probes respond quickly to real variability in the water column.
- Record readings on the field data sheet.
- At one station per day per embayment (typically the last station), do a replicate profile — do one complete profile, then do a second. This allows for the precision of the sonde to be assessed.

**Verification of Water Quality Parameters and Sonde Depth**
- Take and record sonde values in standards at the start and end of a sampling day (salinity, oxygen in 100% water saturated air, and turbidity).
- Verify sonde depth by lowering sonde to a known depth following the procedures listed in UWS SOP Depth GPS.

**Verify chlorophyll a by sampling in a bucket.**
Verify the chlorophyll a readings from the sonde once per day and send to an analytical lab for analysis. This may be conducted at a dock; it does not need to happen at a station.
- Rinse the bucket three times with surface water, fill the bucket with surface water.
- Place the sonde in the bucket, stir for 10 seconds, and then allow to equilibrate, record chlorophyll a reading.
- Filter two samples from the bucket following the procedures indicated in the UWS SOP Filtered Chlorophyll. Prior to withdrawing a sample from the bucket, be sure to stir the water in the bucket with sonde.
Appendix A-5

UWS Standard Operating Procedure – Filtered Chlorophyll a

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1 POINT OF CONTACT

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2 OBJECTIVE

Determine the concentration of chlorophyll a in the surface water, 0.5 m below the surface, following guidelines of the Unified Water Study (UWS). Frequency of sampling and daily order of events are specified in the UWS SOP Sampling Plan.

3 OVERVIEW

A water sample is collected, filtered, and analyzed at an analytical lab to determine the chlorophyll a
4 SOURCES

These procedures are based on the EPA Volunteer Estuary Monitoring Manual (EPA, 2007) and follows methods used in the EPA National Coastal Assessment (EPA, 2001). The EPA Volunteer Estuary Monitoring Manual (EPA, 2007) provides a wealth of specific data for monitoring groups.


5 MATERIALS AND EQUIPMENT

- 5 gallon bucket or larger
- Glass fiber filters (Whatman GF/F with nominal pore size of 0.7 μm), 2.5 cm diameter
- Filter holders, 2.5 cm: Pall brand, 25 mm Easy Pressure Syringe Filter Holder, Delrin Plastic
- 60 mL syringe
- Forceps for handling filters
- Unused and unbleached coffee filter cut into strips
- Aluminum foil
- Airtight container
- Cooler

6 METHODS

6.1 Preparation

- Check that field equipment is prepped and operational.
• Prepare labels for filters. The pre-labeled information should include the monitoring group, UWS unique station ID, date, sample type, sample volume, and replicate ID (“a” or “b”). For example:
  o “Save the Sound LNE-I-01a 6/8/17 Chl-a Vol. 60 mL” & “Save the Sound LNE-I-01b 6/8/17 Chl-a Vol. 60 mL”

• Prep aluminum foil squares for projected number of samples (2 per reference station) plus extras.

• Load all available filter holders with filters. Have at least 4 ready for use.

6.2 Field Collection and Processing

1. Collect water in 5 gallon bucket.
  o Rinse the bucket three times with sample water from the surface. Fill bucket with sample water.

2. If not prepped, load filters into the filter holder.
  o Preloading all filter holders with filters before the sampling event begins is strongly advised.

3. Rinse the 60 mL syringe with ~5 mL of sample water. Repeat for a total of three rinses.

4. Mix the sample well by stirring for 10 seconds with sonde.

5. Record chlorophyll a data from sonde.

6. Fill the 60 mL syringe with sample water taken close to where sonde reading was recorded. Note the volume of water in the syringe – knowing the exact volume filtered is critical to analyzing the chlorophyll a concentration.

7. Connect the filter holder to the syringe.

8. Gently expel water through the filter. The pressure applied to the plunger of the syringe should be slow and steady. If you encounter more than mild resistance, the filter may be clogged (e.g. jellyfish, algae) or you may have filtered sufficient sample.

9. After filtering 60 mL, inspect the filter for color. If a visible light green or light brown color is present, there is sufficient material on the filter for analysis, skip to step 10.
  o If no color is visible, filter another 60 mL and re-inspect.
  o On some occasions, if chlorophyll a levels are very high, filtering may become difficult after as little as 30 mL. If this occurs, and the filter is colored, stop filtering and proceed to step 10.

10. Record volume recorded on field data sheet and sample label when possible. Recording the exact volume filtered is critical to analyzing the chlorophyll a concentration. If you are uncertain of the volume filtered, discard the sample and start over.

11. Use the 60 mL syringe to dry the filter by expelling air through the filter.
  o With the syringe NOT attached to the filter holder, draw air into the syringe.
12. Remove filter with forceps.
   - If forceps are dirty, wipe with a Kimwipe or rinse with distilled water.
   - Only touch the filter on the edges. You may use clean fingers to steady the filter by touching the edge (do not touch the green or brown part).
   - Fold filter in half with forceps and place it in an absorbent pad (unbleached coffee filter). Wrap in aluminum foil by folding the foil around the filter.
13. Filters should be placed in an airtight container and stored in a cooler on ice. Ice should surround the airtight container.

5.3 Sample Storage

14. Store samples in the freezer (-20°C). Frozen samples must be analyzed within 28 days.

5.4 Laboratory Analysis

15. Bring filters to partner lab for analysis or transport group; using UWS Chain of Custody. Frozen samples must be analyzed within 28 days.

7 TROUBLESHOOTING / HINTS

- Make sure filters are dry before storing.
- Gather field equipment the day prior to sampling. Check the field equipment in the morning, before you head out into the field.
- Always carry a copy of this SOP and the relevant parameter-specific SOPs.
- Print out the “quick sheets” for relevant SOPs to use as a reminder in the field. Do not laminate these as you will want to add notes. A plastic page-protector taped close can be used to keep these sheets dry.

8 DATA PROCESSING AND STORAGE

The monitoring group is responsible for obtaining data, entering data into the UWS data template, and delivering the data to the UWS Monitoring Program Coordinator.

The monitoring group is responsible for assuring that the correct unique station ID assigned by the UWS is properly matched with the local organizations station ID codes. Both codes (monitoring group’s station code and UWS unique station ID) will be entered into the data template, along with the GPS coordinates.
9 REFERENCES


10 Quick Sheet – Filtered Chlorophyll a

**SAMPLE from 5 gallon Bucket**

Recording the exact volume filtered is critical to analyzing the chlorophyll a concentration. If you are uncertain of the volume filtered, discard the sample and start over.

The labels should include the UWS unique station ID, monitoring group, replicate ID (“a” or “b”), sample volume, and date. For example:

- station 1: Save the Sound LNE-I-01a 6/8/17 Vol. 60 ml & Save the Sound LNE-I-01b 6/8/17 Vol. 60 ml

1. Collect surface water in a 5 gallon bucket. Rinse bucket three times with sample water prior to filling.
2. Mix the sample well by stirring for 10 seconds with sonde. Record date from sonde.
3. Rinse the 60 mL syringe with ~5 mL of sample water. Repeat for a total of three rinses.
4. **Note the volume of water in the syringe – knowing the exact volume filtered is critical to analyzing the chlorophyll a concentration.**
5. Gently expel sample water through the filter. The pressure applied to the plunger of the syringe should be slow and steady. If you encounter more than mild resistance, the filter may be clogged (e.g. jellyfish, algae) or you may have filtered sufficient sample.
   - After filtering 60 mL, inspect the filter for color. If a visible light green or light brown color is present, there is sufficient material on the filter for analysis. If no color is visible, filter another 60 mL and re-inspect. On some occasions, if chlorophyll a levels are very high, filtering may become difficult after as little as 30 mL.
6. **Recording the exact volume filtered is critical to analyzing the chlorophyll a concentration. If you are uncertain of the volume filtered, discard the sample and start over.**
7. Use the 60 mL syringe to dry the filter by expelling air through the filter.
   - Attach syringe to holder and expel air forcefully through the filter. Do this until no “mist” is aspirated from the filter holder, a minimum of 3 times. DO NOT draw air backwards through the filter. Syringe should be taken off holder each time plunger is drawn up.
8. Remove filter with forceps.
   - Only touch the filter on the edges. You may use clean fingers to steady the filter by touching the edge (do not touch the green part).
   - Fold filter in half with forceps, place in an absorbent pad (unbleached coffee filter), wrap in aluminum foil and apply the label.
9. Labeled, foil wrapped filters should be placed in an airtight container and stored in a cooler on ice. Ice should surround the airtight container.
10. Store samples in the freezer (-20°C). Bring filters to partner lab for analysis. Frozen samples must be analyzed within 28 days.
Appendix A-6

UWS Standard Operating Procedure Qualitative Macrophytes

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1 POINT OF CONTACT

NAME: Peter Linderoth, Save the Sound / CFE, Water Quality Program Manager
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2 OBJECTIVE

Identify problematic macroalgae blooms and the presence of eelgrass in neighboring waters by examining the wrack line on a beach or sampling submerged areas with a rake.
3 OVERVIEW

The goal is to identify areas with problematic amounts of macroalgae and areas with eelgrass (a beneficial condition). This is not intended to be a quantitative assessment. Macrophyte surveys will complement chlorophyll a concentrations to better understand the dominant primary producer in the system. Methods for conducting an assessment vary by the location to be assessed. If a soft shoreline is available, photos of the beach area are used to characterize the amount of macroalgae in the neighboring water. Alternatively, a rake is used to assess what is on the bottom when submerged areas are sampled.

METHOD OVERVIEW (please see text below for a full explanation)

- Sample 3 days during the 3-week period starting July 15 and ending August 7. Try to sample once per week. If this is not possible, please sample such that you maximize the days between sampling. All three days cannot be sampled in the same 7-day window. Two sample days or a date slightly outside of the date criteria may be sufficient in the event of unforeseen complications. This decision will be made by the Monitoring Program Quality Assurance Officer and UWS Science Advisor(s).
- For soft shorelines, take an overview photo of the area, focused on the beach (not the water). Take a close-up photo of each of the dominant macroalgae types on the beach. Complete the datasheet.
- For sampling submerged areas, toss the rake 6 times, 2 tosses in each of 3 directions. Photograph each rake, even empty rakes. Complete the datasheet.
- Enter the data into the online portal. Upload your photos to the online portal.

BACKGROUND

Later on in this SOP, you will see that you are characterizing macroalgae by morphology (hair-like, twig/leaf-like, sheets) and color (green, other). Macroalgae can be hard to identify, thus we are not attempting to identify the genus or species of algae present. The morphology will allow us to identify algae that are nutrient-loving, and thus may represent a water quality problem. Hair-like algae of all colors tend to be nutrient-lovers. Some branching algae in the twig/leaf-like category are also nutrient-lovers, especially if they fall into the red algae category. Sheets of green or red are problematic, while sheets of brown (e.g. kelp) can be indicative of good water quality. Based on these assessments, you may notice that color of the algae is important. Macroalgae fall into three taxonomic divisions denoted by the color of the algae: red (Rhodophyta), green (Chlorophyta), and brown (Phaeophyta). These colors refer to the types of photosynthetic pigments present in the organisms of each division. While a Rhodophyta often looks reddish, it can appear gold, green, brown, or black. The same is true of the other divisions. Green macroalgae typically appear green. Almost all green algae found in Long Island Sound is indicative of high nutrient availability. Thus, we ask that you distinguish between green algae and those of other colors. Good photos of the algae you are seeing will aid in follow-up analysis of these data.
MATERIALS AND EQUIPMENT

➢ GPS unit
  o GPS unit or GPS app for a smartphone
➢ Digital camera with resolution >5 megapixels (most smartphone cameras meet this criteria)
➢ Bucket or plastic bin, for putting macroalgae in if it is very muddy or falling off rake
➢ Bow rake with ~7 m (~20 feet) of rope attached to the time-end of the rake and attached to the handle (cable tie, duct tape, etc.).
  o The rake should be a heavy-duty bow rake with forged steel rakehead, 16 inches wide, with 15 or 16 tines. The total length (handle to tines) should be around 60 inches. A rake with an ash wood handle is appropriate; however, fiberglass or another handle material is also acceptable.
➢ Weight that can be attached to the rake, to help it sink to the bottom. For example, a dive weight or large fishing weights.
➢ Local tidal information for survey site (for soft shoreline surveys).

4 METHODS

4.1 Station Selection

The number of stations sampled per embayment depends upon the size of the embayment, expected variability in macroalgae coverage, and access to sampling areas. An initial site visit of many locations can aid in determining the viability of sampling stations. A minimum of 3 sample stations is recommended for all embayments. More may be needed for larger embayments. Consultation with the Monitoring Program Coordinator and UWS Science Advisors will aid with the determination of how many to include. We cannot assess how much macroalgae is in an embayment using this method. Instead, the goal is to identify embayments which have widespread macroalgae problems and those which have problem areas for macroalgae but not throughout the whole system. Additionally, eelgrass will be noted when present in the survey.

4.1.1 Considerations for Choosing a Station

Must be legally accessible. This includes areas with public access and private areas where permission has been granted by the owner or manager of the area. Roadways closed to pedestrian traffic and all train tracks ARE NOT considered areas with public access, prior permission and official escort are typically required for these areas.

Must be safe to access. The station should not endanger personnel. For example, areas to avoid include: steep inclines, roadways with insufficient buffers from passing cars, areas overrun with poison ivy, areas with excessive debris, etc.

Look for areas where macroalgae is likely to collect. The goal is to assess what is out in the system. If you
choose an area that is too deep or where the current is sweeping away all of the macroalgae, you may miss an issue that is occurring in shallower, calmer waters. However, if the overall system is deep and scoured by the current or wind driven waves, then sample these areas, as they do indeed represent the system. To assess how representative an area is, try sampling in a few spots in a general area of the embayment. If macroalgae is widespread, make a note of this. If the macroalgae you are sampling is at a spot where macroalgae tends to collect, also make a note of this.

4.1.2 Station Selection Guidance for Soft Shoreline (Beaches)

For soft shoreline assessments, the method calls for taking a photo of the beach and close-ups of the dominant macroalgae types. This is the quicker assessment when compared to a rake survey. You are looking for an area with the following characteristics:

- A beach composed of sand or gravel.
- Avoid marshes (though you may use a rake from a marsh).
- Avoid rocky intertidal zones.
- Areas with attached brown macroalgae (rockweed, knotted wrack) are fine if the area also collects free-floating macroalgae.
- An area you know is not raked or otherwise maintained by municipalities or local residents.

4.1.3 Guidance for Submerged Areas (Sampling from Boats, Docks, Jetties, etc.)

For sampling submerged areas, the method calls for tossing in a garden rake attached to a rope and slowly and steadily pulling in the rake. Two tosses are conducted in three directions, for a total of six tosses. A photo is taken of each rake, including when only mud is collected. Potential areas include:

- Marinas, docks, or jetties can be suitable locations, if their depth is representative of the overall area. Even deep boat slips can be acceptable as macroalgae tends to collect in the deeper locations. Avoid areas where prop scour is likely to push macroalgae away. If your rake toss looks similar to what you see in shallower areas, the areas should be suitable. *Note – when sampling from a dock, be considerate of private property: avoid areas where your actions may be misconstrued as potentially damaging to boats. In other words, choose an area of the dock where you have some room to move, not an area with little space between boats. Always check in with the office or property owner before sampling, to let them know you have permission to sample and so they know who you are and what you are doing.*

- Causeways, seawalls, and shorelines without a beach deemed safe to access are suitable sampling locations. Avoid areas scoured clean by the water flow (i.e., the mouth of a culvert). Be sure to choose areas where you will not catch brown macroalgae attached to rocks at the shoreline (rockweed, knotted wrack). We are interested in the beneficial shoreline macroalgae.

- Boat sampling is acceptable if the depth of the area is representative of the embayment. In other words, you should not sample in the deepest or the shallowest locations.
4.1.4 Examples of Sample Locations

When sampling from a dock, jetty, causeway, or other hardened shoreline, choose three locations and do two rake tosses from each location [red lines indicate direction and length of rake toss]. Note that sampling is not done from the finger docks in the image below; these docks can be too skinny to safely accommodate two people and allow for proper framing of the photograph.
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If you are in a location which inhibits moving around, do two rake tosses in each of three directions. For example, a kayak launch amidst a dense stand of *Phragmites* can provide limited access to the shoreline. In this case, two rakes tosses are directed straight out into the water, two upstream, and two downstream.

When sampling from a boat at anchor, we assume the boat is shifting slightly with the wind and current. You may do all six tosses from the same location in the boat, tossing the rake in different directions. However, if you feel that you are sampling the same location six times, you will need to shift to a new location on the boat between tosses. If your boat is drifting, you may toss in the same direction each time, as you will be floating over new substrate.

4.2 Preparation

- Ensure battery power and available storage is sufficient on camera or smartphone.
- Ensure battery power is sufficient on GPS unit or smartphone.
- Collect equipment and ensure all is in working order.
- Always carry a copy of this SOP and the relevant parameter-specific SOPs.
- Print out the "quick sheets" for relevant SOPs to use as a reminder in the field. Do not laminate these as you will want to add notes. A plastic page-protector taped close can be used to keep these sheets dry.

4.3 Field Collection and Processing

Sampling at each station should be quick. In field trials, the UWS Science Advisors completed sampling at a station in 10 to 15 minutes, though getting a rake hung up on a rock can increase the time needed. Soft shoreline (beach) areas can go even quicker. Please keep in mind this is a qualitative survey, so
spending additional time to quantify the macroalgae is not necessary. You are making a call of “not much here”, “wow, that’s a lot of macroalgae”, or “this could be a problem, but I’m not sure.” The first station may take a bit longer, as you get acquainted with the datasheet.

4.3.1 Option 1: Soft Shoreline Station (Beaches)

- Conduct the assessment within 3 hours of low tide (this gives you a 6-hour window).
- Complete the station information on the datasheet.
  - IMPORTANT: Record GPS points as decimal degrees (e.g. 41.56437 N, -71.87634 W). Please do not report GPS in hours and minutes. For example, a location with decimal degrees of 41.56437 N, -71.87634 W can also be reported in hours and minutes as 41° 33’ 51.7314", -71° 52’ 34.8234". Note that the numbers are different, decimal degrees are not just leaving out the symbols for hours (‘) and minutes (”). We ask for decimal degrees because hours and minutes do not translate well in a spreadsheet. If you do not understand this distinction, please ask an advisor to explain.
  - Record the amount of macroalgae on the beach as “none”, “some”, or “lots”.
    - None – essentially nothing on the beach, only attached macroalgae is present.
    - Some – ranges from a small bit of macroalgae on the beach to large wracklines of macroalgae present on the beach. This is qualitative, the Monitoring Program Coordinator and UWS Science Advisors will review when comparing stations, so do not worry too much about what truly constitutes some versus lots.
    - Lots – covers >50% of the beach. May be a thin covering, in thick wracklines, or a thick layer covering the whole beach.
  - Examine the macroalgae and use the guide to identify macroalgae morphology (hair-like, twig/leaf like, sheets) and color (green, other). You only need to record macroalgae that constitutes more than 10% of the macroalgae present on the beach.
  - Take two overview photos, showing the overall appearance of the shoreline. One should include upland areas for context, the next should be slightly closer, showing macroalgae (or lack of macroalgae) on the beach (examples below).
    - Zoom in on the overview photo and confirm the beach is in focus (versus something in the background or foreground).
    - Do not submit any overview photos deemed unacceptable.
    - The two overview photos should be sufficient. However, if lighting or size of the station requires, you may take multiple photos.
    - Record an identifier for the photo on the datasheet. For a camera, this may be the filename. For a smartphone, record the time associated with the photo; this can be located by viewing the photo, the time the photo was taken should be on the screen.
For the dominant macroalgae types, take a close-up photo of each macroalgae.

- This is a rough assessment of the dominant macroalgae types; you DO NOT need to photograph each type of macroalgae present on the beach.
- Add something to the photo to provide a size reference. This could be a ruler, your foot, a brick, your keys, etc. It needs to be something with a known size (not a stick you pick up, which can be any size).
- Zoom in on the macroalgae photo and confirm the macroalgae is in focus (versus something in the background or foreground).
- Be sure that the lighting allows for realistic colors in the photo.
- The photo should not include the shadow of the photographer.
- Record an identifier for the photo on the datasheet. For a camera, this may be the filename. For a smartphone, record the time associated with the photo; this can be located by viewing the photo, the time the photo was taken should be on the screen.
4.3.2 Option 2: Submerged Station

- Note – a YouTube video demonstrating how to properly toss a rake is available at: [https://www.youtube.com/watch?v=yMqSpAhE2V](https://www.youtube.com/watch?v=yMqSpAhE2V) or find the video by searching for “macroalgae rake toss.”

- The timing of sampling is not critical. The only criterion is that low tide has not exposed the sediment at the sampling station.

- Complete the station information on the datasheet. An example of a completed datasheet is included on page 14 of this SOP.

  o IMPORTANT: Record GPS points as decimal degrees (e.g. 41.56437 N, -71.87634 W). Please do not report GPS in hours and minutes. For example, a location with decimal degrees of 41.56437 N, -71.87634 W can also be reported in hours and minutes as 41°33'51.7314", -71°52'34.8234". Note that the numbers are different, decimal degrees are not just leaving out the symbols for hours (°) and minutes ('). We ask for decimal degrees because hours and minutes do not translate well in a spreadsheet. If you do not understand this distinction, please ask the UWS Monitoring Program Coordinator to explain.

- Select a sampling station at your location.

  o If the station allows, conduct 2 rake tosses at each of 3 different spots. An example would be moving to three different areas on a dock. See Section 4.1.4 (page 5) for diagrams of where to sample.

  o If the station does not allow you to move around, conduct 2 tosses in each of 3 different directions. An example would be an opening in a marsh that allows access to the embayment. The rake is tossed upstream, downstream, and straight out into the water. See Section 4.1.4 for diagrams of where to sample.
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➢ Confirm that the line is securely attached to the rake. Place your foot on the end of the rope, to keep it from going into the water.

➢ Face the embayment and hold the rake such that your dominant hand is at the end of the rake (ready to push the rake).

➢ Push the rake out into the water a distance of ~2 rake lengths. The motion is similar to an underhand slow pitch, rather than a javelin throw.

➢ If the toss is too short or too long, bring back and try again.

➢ The tines of the rake should be downward. If the rake lands with tines up, bring back and try again.

➢ After a successful toss, allow rake to settle on the floor then slowly and steadily pull the line so the teeth of the rake drag on the embayment bottom.
   o If the rake gets hooked on a rock or other object, try taking up the slack in the rope and pulling up on the rake, or moving to the left and to the right. If you feel that macroalgae was likely dislodged, try tossing again. For some rocks, freeing the rake is easy enough that macroalgae is mostly intact on the rake. For these cases, you do not need to toss again.
   o When removing the rake from the water, do so such that macroalgae does not fall off of the rake.
   o Make certain that you DO NOT pick up macroalgae removed from the rake on a previous toss. Be aware of where you discard macroalgae from the rake so that you are not capturing the same macroalgae with each toss.

➢ Record the amount of macroalgae on the rake as “none”, “some”, or “lots” (using sample photos shown below to assist).
   o None – no macroalgae on the rake.
   o Some – some tines are still bare.
   o Lots – all tines hold macroalgae. This includes anything from a thin coat of a slimy macroalgae to thick clumps of a wiry or leafy macroalgae.
   o If you get a lot of mud, put into a bucket or bin and rinse; mud that clumps on the rake is often being held together by fine macroalgae

➢ Take a photo of the rake (and macroalgae) after each toss.
   o Include all of the rake tines and macroalgae in the photo. If a portion of the rake tines is not included, take another photograph. You will want to be about 4 feet from the rake. Do not get too close – some cameras do not focus at shorter distances. With the resolution required, zooming in to look at macroalgae can be done on the computer. You do not need to get super close to get a good shot of the macroalgae, further back is better.
   o Frame the shot such that the light is behind the photographer (or at a slight angle), so that the light is good and the photographer’s shadow is not cast on the macroalgae.
   o Be aware of the background and change your location for the photo as needed. Light colors in the background make the macroalgae look darker; avoid light backgrounds (e.g. a white boat).
   o Zoom in on the photo and confirm the macroalgae is in focus (versus something in the background or foreground).
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- Be sure that the lighting allows for realistic colors in the photo.
- Record an identifier for the photo on the datasheet. For a camera, this may be the filename. For a smartphone, record the time associated with the photo; this can be located by viewing the photo, the time the photo was taken should be on the screen.

- Sort through the macroalgae and use the guide to identify macroalgae morphology (hair-like, twig/leaf like, sheets) and color (green, other). You only need to record macroalgae that constitutes more than 10% of the macroalgae present on the rake.

Example photos of rakes:

Station 1: Wequetequock Cove, Stonington Marina, 7/13/2016

![Image of rakes with macroalgae]
Station 2: Wequetequock Cove, kayak launch near Saltwater Farm Vineyard, 7/13/2016

Some

Some

Some

Lots

Station 3: Wequetequock Cove, Elihu Island causeway, 7/13/2016

Overview – Lots

Lots

Close-up of dominant macroalgae (all beach looked like this photo, only 1 photo necessary).
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The macroalgae collected on the soft shoreline at Elihu Island causeway is characterized as “lots.” To verify that it represents the area, we also tossed a rake off the causeway, about midway between the mainland and the island. Each toss had “lots,” confirming the beach survey was indeed representative. Elgrass is visible on these rakes and was visible on the beach. The red branching macroalgae on the rakes was not a large component of what was on the beach. It is fine if the macroalgae composition varies, we are most interested in the amount of macroalgae.

4.4 Sample Storage
  Not applicable.

4.5 Laboratory Analysis
  Not applicable.

5  TROUBLESHOOTING / HINTS:

- Organize all volunteers well before sampling events.
- Check tide information before initiating the surveys.
- Bring backup rake and line, if possible.
- Always carry a copy of this SOP and the relevant parameter-specific SOPs.
- Print out the “quick sheets” for relevant SOPs to use as a reminder in the field. Do not laminate these as you will want to add notes. A plastic page-protector taped close can be used to keep these sheets dry.

6  DATA PROCESSING AND STORAGE

The monitoring group is responsible for obtaining data, entering data into the UWS data template, and delivering the data to the UWS Monitoring Program Coordinator.

The monitoring group is responsible for assuring that the correct unique station ID assigned by the UWS is properly matched with the local organizations station ID codes. Both codes (monitoring group’s station code and UWS unique station ID) will be entered into the data template, along with the GPS coordinates.
Macrolegs assessment data and photos will be submitted via an online portal. This will allow for consistent formatting in the data, easy transference and a storage system for the photos, and immediate access to the photos for the Monitoring Program Coordinator and UWS Scientific Advisors. We will be using a free software called KoBo Toolbox. Once uploaded by UWS participants, the data will be downloaded and stored by Save the Sound.

- Each station within your embayment will use a different form.
- Not all data on the datasheet is required in the form.
- Bookmark the form in your browser, this will make it easier to access.
- You may save an incomplete form and return to the form at a later time.
- Directions for accessing the form are included below.

How to Collect Data on a Mobile Device

Collect data on your Android device with KoBoCollect

To start collecting data with the KoBoCollect Android app you need to enter the following URL into the URL field in the KoBoCollect settings:

https://kc.kobotoolbox.org/c/test

Click here to read more about getting started with KoBoCollect

Collect Data on Any Device in Your Browser

For iPhones or any other device that has a Web browser, use the following link to enter data - even offline:

https://ee.kobotoolbox.org/x/1iy8le

(Or if you have a barcode scanner on your phone, just scan the barcode on the right)

7 REFERENCES

None
8 Quick Sheet – Qualitative Macrophytes

Method Overview
➢ Sample 3 days during the 3-week period starting July 15 and ending August 7. Try to sample once per week. If this is not possible, please sample such that you maximize the days between sampling. All three days cannot be sampled in the same 7-day window. Two sample days or a date slightly outside of the date criteria may be sufficient in the event of unforeseen complications. This decision will be made by the Monitoring Program Quality Assurance Officer and UWS Science Advisor(s).
➢ For soft shorelines, take an overview photo of the area, focused on the beach (not the water). Take a close-up photo of each of the dominant macroalgae types on the beach. Complete the datasheet.
➢ For sampling submerged areas, toss the rake 6 times, 2 tosses in each of 3 directions (if limited by access to a single point) or 3 areas (if you have room to move to another location at the station). Photograph each rake, even empty rakes. Complete the datasheet.
➢ Enter the data into the online portal. Upload your photos to the online portal.

Preparation
➢ Gather field equipment.
➢ Ensure battery power and available storage is sufficient on camera or smartphone.
➢ Ensure battery power is sufficient on GPS unit or smartphone.
➢ Always carry a copy of this SOP and the relevant parameter-specific SOPs.
➢ Print out the “quick sheets” for relevant SOPs to use as a reminder in the field. Do not laminate these as you will want to add notes. A plastic page-protector taped close can be used to keep these sheets dry.

Field Collection and Processing

Option 1: Soft Shoreline Station (Beaches)
➢ Conduct the assessment within 3 hours of low tide (this gives you a 6-hour window).
➢ Complete the station information on the datasheet. IMPORTANT: Record GPS points as decimal degrees e.g. 41.56437 N, -71.87634 W.
➢ Take two overview photos, showing the overall appearance of the shoreline. One should include upland areas for context, the next should be slightly closer, showing macroalgae (or lack of macroalgae) on the beach (examples below).
➢ For the dominant macroalgae types, take a close-up photo of each macroalgae.
➢ Record the amount of macroalgae on the beach as “none”, “some”, or “lots”.
  o None – essentially nothing on the beach, only attached macroalgae is present.
  o Some – ranges from a small bit of macroalgae on the beach to large wracklines of macroalgae present on the beach. This is qualitative, the science advisors will review when comparing stations, so do not worry too much about what truly constitutes some versus lots.
  o Lots – covers > 50% of the beach. May be a thin covering, in thick wracklines, or a thick layer covering the whole beach.
➢ Examine the macroalgae and use the guide to identify macroalgae morphology (hair-like, twig/leaf like, sheets) and color (green, other). You only need to record macroalgae that constitutes more than 10% of the macroalgae present on the beach.
➢ Remember – we are looking for large amounts of seaweed. If there is nothing on the beach, but you see lots of seaweed [that are not the beneficial brown seaweed] in the water, then try a rake toss in the water.
Option 2: Submerged Station

- The timing of sampling is not critical. The only criterion is that low tide has not exposed the sediment at the sampling station.
- Note – a YouTube video demonstrating how to properly toss a rake is available at: https://www.youtube.com/watch?v=yMq5SaAG2WY.
- Complete the station information on the datasheet. IMPORTANT: Record GPS points as decimal degrees (e.g. 41.56437 N, -71.87634 W).
- Confirm that the line is securely attached to the rake. Place your foot on the end of the rope, to keep it from going into the water. Face the embayment and hold the rake such that your dominant hand is at the end of the rake (ready to push the rake). Push the rake out into the water a distance of ~2 rake lengths. The motion is similar to an underhand slow pitch, rather than a javelin throw.
  - If the toss is too short or too long, bring back and try again.
  - The tines of the rake should be downward. If the rake lands with tines up, try again.
- After a successful toss, allow rake to settle on the floor then *slowly and steadily* pull the line so the teeth of the rake drag on the embayment bottom.
  - If the rake gets hooked on a rock or other object, try taking up the slack in the rope and pulling up on the rake, or moving to the left and to the right. If you feel that macroalgae was likely dislodged, try tossing again.
  - When removing the rake from the water, do not let macroalgae fall off of the rake.
  - Make certain that you DO NOT pick up macroalgae removed from the rake on a previous toss.
  - If you get a lot of mud, put into a bucket or bin and rinse; mud that clumps on the rake is often being held together by fine macroalgae.
- Take a photo of the rake (and macroalgae) after each toss.
  - Include all of the rake times and macroalgae in the photo. If a portion of the rake times is not included, take another photograph. You will want to be about 4 feet from the rake. *Do not get too close* – some cameras do not focus at shorter distances. With the resolution required, zooming in to look at macroalgae can be done on the computer. You do not need to get super close to get a good shot of the macroalgae, further back is better.
  - Frame the shot such that the light is behind the photographer.
  - Be aware of the background and change your location for the photo as needed. Light colors in the background make the macroalgae look darker; avoid light backgrounds (e.g. a white boat).
  - Zoom in on the photo and confirm the macroalgae is in focus (versus something in the background or foreground).
  - Be sure that the lighting allows for realistic colors in the photo.
  - Record an identifier for the photo on the datasheet. For a camera, this may be the filename. For a smartphone, record the time associated with the photo; this can be located by viewing the photo, the time the photo was taken should be on the screen.
- Record the amount of macroalgae on the rake as “none”, “some”, or “lots”.
  - None – no macroalgae on the rake.
  - Some – some tines are still bare.
  - Lots – all tines hold macroalgae. This includes anything from a thin coat of a slimy macroalgae to thick clumps of a wiry or leafy macroalgae.
  - If you get a lot of mud, put into a bucket or bin and rinse; mud that clumps on the rake is often being held together by fine macroalgae.
- Examine the macroalgae and use the guide to identify macroalgae morphology (hair-like, twig/leaf like, sheets) and color (green, others). You only need to record macroalgae that constitutes more than 10% of the macroalgae present on the rake.
Appendix A-7

Standard Operating Procedure
Filtered Nutrients Sample Collection

revised January 2019
Page 1 of 6

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I. OBJECTIVE: Determine the ammonia, nitrite, nitrate, total dissolved nitrogen, and orthophosphate concentrations in the water column.

II. OVERVIEW: Water samples are collected from the surface of the water column. This water is filtered through GF/F filters and delivered into clean HDPE bottles. The samples are analyzed on a Lachat 8500 QuikChem® Autoanalyzer, manifold reaction unit, colorimetric detector, 520 nm wavelength filter and Omnion software data system.

III. SOURCE:

IV. MATERIALS AND EQUIPMENT:
A. FIELD COLLECTION AND STORAGE OF SAMPLES
Plastic bottles, 60 mL, HDPE, (Fisher Scientific Catalog Number: 03-313-11B)
Plastic bottles, 1 L, HDPE, (Fisher Scientific Catalog Number: 02-925-3E)
Glass Fiber Filters (GF/F), 2.5 cm diameter, retains particles down to 0.7 µm, (Fisher Scientific Catalog Number: 09-874-64; Whatman Number:1825-025)
Filter holders, 2.5 cm (preferred: Pall brand, 25 mm Easy Pressure Syringe Filter Holder, Delrin Plastic; second choice: Millipore Swinnex Filter Holder, 25 mm polypropylene with silicon gasket, Fisher Scientific Catalog Number: SX00 025 00; Millipore Number:SX002500)
Forceps for handling filters
Acid Bath: 1 N (Certified) hydrochloric acid (Fisher Scientific Catalog Number: SA48-4)
2 L of ASTM type I (ultrapure) for field blank
Equipment for getting water samples:
  option 1 – Master flex pump with silicone tubing sufficient for sample depth
  option 2 – Homemade sampling poles with 1 L Bottle

Equipment for delivering sample through filter to 60 mL bottles:
  option 1 – MasterFlex pump with silicone tubing
  option 2 – 60 mL syringe

V. METHODS

A. PREPARATION
  • Check that the pump or other sampling equipment are operational
  Prepare the sample bottles:
  • Wear powder-free nitrile gloves
  • Rinse 1 L bottles with ultrapure, ASTM type 1 (ultrapure) water. Allow to dry and store with caps on
    • If 60 mL HDPE bottles are new and un-used, rinse the vials three times with ultrapure, ASTM type 1 (ultrapure) water. Allow to dry and store with caps on
    • If 60 mL HDPE bottles have been used, acid wash the bottles. Acid washing entails soaking the bottles in a 1 N hydrochloric acid bath for 60 minutes followed by rinsing three times with ultrapure, ASTM type 1 (ultrapure) water. Allow to dry and store full of ultrapure, ASTM type 1 (ultrapure) water with caps on

B. FIELD COLLECTION AND STORAGE OF SAMPLES

A slightly different procedure is used based on the sampling method. Choose the protocols for your sampling method

MasterFlex Pump

1. Rinse the sampling equipment with sample water. This involves rinsing with three times the volume of the sampling device. Place the intake 0.5 m below the surface, allow the pump to
run long enough to pass a minimum of three times the volume of the tubing through the system. Discharge this water over the boat, do not collect.

2. Assemble the filtering mechanism. Connect the filter holder (with filter loaded into the holder) to the pump. Change filter if needed during collection.

3. Rinse the sample bottles with water from target sampling depth. Cap the bottle and shake to rinse. Discard the sample used as a rinse. Repeat three times for each bottle.

4. Fill sample bottle with water from target sampling depth.

5. Sample bottle is immediately stored in cooler full of ice for transport to lab.

Note: Sample(s) can be collected in either the 1 L bottle or three 60 mL HDPE bottles. If only filling the 1 L bottle, filtering can occur when 60 mL bottles are filled as seen lower in this SOP under sample preparation. If filling 60 mL bottles in the field, only collect up to 50 mL in the bottle.

Note: About mid-way through the sampling day, perform a field blank. Follow the procedures for sampling above, but use ASTM Type I (ultrapure) water brought out on the boat in place of the field water.

**Homemade Sampling Poles with 1 L Bottle**

A sampling pole can be fabricated by a member of your organization. The key criteria for acceptability is the ability to sample from a specific depth without contaminating the sample. The method involves reaching into the water with the sample bottle inverted and full of air, then righting the bottle and allowing it to fill at the appropriate depth.

Homemade poles will typically work only for 0.5 m below the surface; deeper depths require alternate methods of sampling.

- Remove the cap from the bottle.
- Rinse the bottle three times with surface water. Fill bottle, cap bottle, invert, then discard rinse.
- Holding the bottle with the mouth down, lower it to 0.5 m below the surface.
- Invert the bottle so that air empties out of the bottle and the bottle fills with water.
- Bring to the surface and cap.
- Sample bottle is immediately stored in cooler full of ice until being filtered.

Note – About mid-way through the sampling day, perform a field blank. Follow the procedures for sampling above, but use ASTM Type I (ultrapure) Water brought out on the boat in place of the field water.

The following steps apply to when using a MasterFlex for sample preparation:

**MasterFlex Pump – after collecting sample in 1 L bottle and storing on ice**

1. Rinse the sampling equipment with sample water. This involves rinsing with three times the volume of the sampling device. Place the intake into the sample bottle, allow the pump to run long enough to pass a minimum of three times the volume of the tubing through the system. Discharge this water, do not collect.

2. Assemble the filtering mechanism. Connect the filter holder (with filter loaded into the holder) to the pump. Run approximately 15 mL through the filter before next step.

3. Rinse the sample bottles. Deliver a small volume of the filtered sample into the sample bottles. Cap the vials and shake to rinse. Discard the sample used as a rinse. Repeat for a total of three small volume rinses per bottle.

4. Deliver between 45 mL and 50 mL of sample (through the filter) to the bottles. Be sure to keep the volume in the bottle under 50 mL, this head space will allow for expansion during freezing. Change the filter if needed.

5. Store the bottles upright in the freezer (-20º C).

A sample, replicate, and sample in reserve are collected from each station.

The following steps apply when using a syringe for sample preparation:
1. Rinse the sampling equipment with sample water. This involves rinsing with three times the volume of the sampling device.

2. Assemble the filtering mechanism. Connect the filter holder (with filter loaded into the holder). Rinse the 60 mL syringe with 5 mL of sample water. Repeat for a total of three rinses. Fill the 60 mL syringe with sample water.

3. Rinse the HDPE bottles. Deliver a small volume of the filtered sample into the HDPE bottles. Cap the bottles and shake to rinse. Discard the sample used as a rinse. Repeat for a total of three small volume rinses per bottle.

4. Deliver between 45 mL and 50 mL of sample (through the filter) to the bottles. Be sure to keep the volume below 50 mL, this head space will allow for expansion during freezing. Change the filter as needed.

5. Upon return to the lab, store the bottles in the freezer (-20°C).

*A sample, replicate, and sample in reserve are collected at each station.*

C. LABORATORY ANALYSIS

- The nutrients are analyzed on a Lachat 8500 QuikChem® Autoanalyzer, manifold reaction unit, colorimetric detector, 520 nm wavelength filter and Omnion software data system. Lachat Quickchem Autoanalyzer, or similar instrument, at the Interstate Environmental Commission (IEC). Instrument operations are detailed in the IEC Laboratory SOP which is contained in the UWS QAPP.

VI. TROUBLE SHOOTING / HINTS

- Avoid contaminating the samples – do not touch: the insides of the bottle caps, the mouth of the bottles, the insides of the graduated cylinders, the filter pads

- Two people working in tandem will speed the process. One person focuses on filtering while the other handles the sample bottles and filters. The person filtering the sample can lend a hand when s/he gets ahead of the sample handler.
VII. DATA PROCESSING AND STORAGE

- Enter the data on the field sheet. Be sure to fill out the data sheet completely.
- Enter data when returned from the lab into the Excel template.

VIII. QUICK SHEET

*Refer to the SOP for details, this list is only a reminder!!*

FIELD
Rinse everything three times before collecting sample
Store in cooler on ice while in the field

STORAGE
1 L sample bottle remains on ice immediately after collection. Filtering must commence the
same day of sampling.
Store samples *upright* in the freezer
Separate replicates a and b (samples to be analyzed) from c (sample held in reserve)

ANALYSIS
Follow any potential guidelines established for analytical lab
Appendix A-8

Standard Operating Procedure revised January 2019
Total Nitrogen and Total Phosphorous Page 1 of 6
Sample Collection

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I. OBJECTIVE: Determine the dissolved total nitrogen and total phosphorous concentrations in the water column.

II. OVERVIEW: Water samples are collected from the surface in the water column. The samples are analyzed on a Lachat Autoanalyzer with autosampler and dilutor, manifold reaction unit (heating unit), colorimetric detector, 880 nm wavelength filter and Omnion software data system.

III. SOURCE:

IV. MATERIALS AND EQUIPMENT:
A. FIELD COLLECTION AND STORAGE OF SAMPLES
Plastic bottles, 60 mL, HDPE, (Fisher Scientific Catalog Number: 03-313-11B)
Plastic bottles, 1 L, HDPE, (Fisher Scientific Catalog Number: 02-925-3E)
Acid Bath: 1 N (Certified) hydrochloric acid (Fisher Scientific Catalog Number: SA48-4)
2 L of ASTM type I (ultrapure) for field blank
Equipment for getting water samples:
   option 1 – MasterFlex pump with silicone tubing sufficient for sample depth
   option 2 – Homemade sampling poles with 1 L Bottle

Equipment for delivering sample to 60 mL bottles:
   option 1 – MasterFlex pump with silicone tubing
   option 2 – 60 mL syringe
B. LABORATORY ANALYSIS

Included in IEC Lab SOPs in QAPP.

V. METHODS

A. PREPARATION

- Check that the pump or other sampling equipment are operational
- Prepare the sample bottles:
  - Wear powder-free nitrile gloves
    - Use only ultrapure, ASTM type I (ultrapure)
    - Rinse 1 L bottles with ASTM type I (ultrapure) Allow to dry and store with caps on.
    - If 60 mL HDPE bottles are new and un-used, rinse the vials three times with ultrapure,
      - ASTM type I water. Allow to dry and store with caps on.
    - If 60 mL HDPE bottles have been used, acid wash the bottles. Acid washing entails soaking
      - the bottles in a 1 N hydrochloric acid bath for 60 minutes followed by rinsing three times
      - with ultrapure, ASTM type I water. Allow to dry and store full of ASTM type I,
      - ultrapure, water with caps on.

B. FIELD COLLECTION AND STORAGE OF SAMPLES

A slightly different procedure is used based on the sampling method. Choose the protocols for
your sampling method.

**MasterFlex Pump**

1. Rinse the sampling equipment with sample water. This involves rinsing with three times the
   volume of the sampling device and bottle. Place the intake into the target sampling depth,
   - allow the pump to run long enough to pass a minimum of three times the volume of the
     tubing through the system. Discharge this water over the boat, do not collect.
2. Rinse the sample bottle with water from target sampling depth. Cap the bottle and shake to
   - rinse. Discard the sample used as a rinse. Repeat three times.
3. Fill sample bottle with water from target sampling depth.
4. Sample bottle is immediately stored in cooler full of ice for transport to lab.

*Note: Sample(s) can be collected in either the 1 L bottle or three 60 mL HDPE bottles. If filling 60 mL bottles in the field, only collect up to 50 mL in the bottle.*

*Note: About mid-way through the sampling day, perform a field blank. Follow the procedures for sampling above, but use ASTM type I (ultrapure) out on the boat in place of the field water.*

**Homemade Sampling Poles with 1 L Bottle**

A sampling pole can be fabricated by a member of your organization. The key criteria for acceptability is the ability to sample from a specific depth without contaminating the sample. The method often involves reaching into the water with the sample bottle inverted and full of air, then righting the bottle and allowing it to fill at the appropriate depth.

Homemade poles will typically work only for 0.5 m below the surface; deeper depths require alternate methods of sampling.

- Remove cap from bottle.
- Rinse the bottle three times with surface water. Fill bottle, cap bottle, invert, then discard rinse.
- Holding the bottle with the mouth down, lower it to 0.5 m below the surface.
- Invert the bottle so that air empties out of the bottle and the bottle fills with water.
- Bring to the surface and cap.
- Sample bottle is immediately stored in cooler full of ice until being filtered.

*Note – About mid-way through the sampling day, perform a field blank. Follow the procedures for sampling above, but use ASTM type I (ultrapure) brought out on the boat in place of the field water.*

*The following steps apply when using a MasterFlex for sample preparation:*
MasterFlex Pump – after collecting sample in 1 L bottle and storing on ice

1. Rinse the sampling equipment with sample water. This involves rinsing with three times the volume of the sampling device. Place the intake into the sample bottle, allow the pump to run long enough to pass a minimum of three times the volume of the tubing through the system. Discharge this water, do not collect.

2. Rinse the sample bottles. Deliver a small volume of the sample into the sample bottles. Cap the vials and shake to rinse. Discard the sample used as a rinse. Repeat for a total of three small volume rinses per bottle.

3. Deliver between 45 mL and 50 mL of sample to the bottles. Be sure to keep the volume in the bottle under 50 mL, this head space will allow for expansion during freezing. Change the filter if needed.

4. Store the bottles upright in the freezer (-20° C).

A sample, replicate, and sample in reserve are collected from for station.

The following steps apply when using a syringe for sample preparation:

1. Rinse the sampling equipment with sample water. This involves rinsing with three times the volume of the sampling device.

2. Rinse the 60 mL syringe with 5 mL of sample water. Repeat for three rinses. Fill the 60 mL syringe with sample water.

3. Rinse the HDPE bottles. Deliver a small volume of the sample into the HDPE bottles. Cap the bottles and shake to rinse. Discard the sample used as a rinse. Repeat for a total of three small volume rinses per bottle.

4. Deliver between 45 mL and 50 mL of sample to the bottles. Be sure to keep the volume below 50 mL, this head space will allow for expansion during freezing.

5. Upon return to the lab, store the bottles in the freezer (-20° C).
A sample, replicate, and sample in reserve are collected at each station.

C. LABORATORY ANALYSIS

- The nutrients are analyzed on a Lachat 8500 QuikChem® Autoanalyzer, manifold reaction unit, colorimetric detector, 520 nm wavelength filter and Omnion software data system. Lachat Quickchem Autoanalyzer, or similar instrument, at the Interstate Environmental Commission (IEC). Instrument operations are detailed in the IEC Laboratory SOP.

VI. TROUBLE SHOOTING / HINTS

- Avoid contaminating the samples – do not touch: the insides of the vial caps, the mouth of the vials, the insides of the graduated cylinders.
- Two people working in tandem will speed the process.

VII. DATA PROCESSING AND STORAGE

- Enter the data on the field sheet. Be sure to fill out the data sheet completely.
- Enter data when returned from lab into the Excel template.
VIII. QUICK SHEET

Refer to the SOP for details, this list is only a reminder!!

FIELD
Rinse everything three times before collecting sample (pump / sample collection vessel; vials)
store in cooler on ice while in the field

STORAGE
Store samples upright in the freezer
Separate replicates a and b (samples to be analyzed) from c (sample held in reserve)

ANALYSIS
Follow any potential guidelines established for analytical lab
Appendix A-9

Standard Operating Procedure
Continuous Dissolved Oxygen

revised February 2020
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I. OBJECTIVE: Determine the dissolved oxygen (DO) concentration and saturation over a period of time at select intervals.

II. OVERVIEW: Continuous dissolved oxygen sampling instruments – dissolved oxygen, conductivity, and barometric pressure loggers – are deployed in the field to log dissolved oxygen and saturation. Dissolved oxygen and conductivity are logged at approximately 0.5 meters off the bottom and barometric pressured is logged in a secure location in close proximity to the other loggers.

III. SOURCE:

IV. MATERIALS AND EQUIPMENT:
A. Continuous logging stations
Dissolved oxygen logger (HOBO U26)
Barometric pressure logger (HOBO U20L-01)
Conductivity Logger (Star-Oddi DST-CT, 13-50 mS/cm range)
Aquarium water pump for inter comparison of DO and Conductivity loggers and sonde
Aquarium air stone for inter comparison of DO and Conductivity loggers and sonde
Sodium sulfite DO calibration solution (HOBO U26-CAL-SOL)
Ice/tap water for adjusting conductivity in water bath
Multiparameter sonde (YSI EXO1 or Eureka Manta +35) with DO and conductivity attachments
Conductivity Standard (YSI: 50,000 μS/cm) for calibrating sonde
Q tips for cleaning inside of DO loggers
B. LABORATORY ANALYSIS

No laboratory analysis.

V. METHODS

A. Logger Inter Comparison (Prior to Field Deployment)

- Check that all sampling equipment are operational and ready for deployment
- Calibrate conductivity and DO on multiparameter sonde to manufacturer recommendations the day before or morning of this logger comparison
- Calibrate all loggers according to manufacturer recommendations the day before or morning of this logger comparison
- Synchronize loggers and multiparameter sonde to same time. Set loggers to 15 minute interval readings
- Prepare a water bath with water similar to deployment stations; enough volume to fit all loggers for comparison and sonde
- Place DO and conductivity loggers ready for deployment in the water bath and note the time the loggers will be taking readings.
- Run the aquarium pump to keep water flowing in water bath. Do not keep loggers or sonde close enough to allow bubbles to come into contact with sensors
- Record DO and conductivity readings with the multiparameter sonde at the same time the loggers record data
- Add ice or tap water to adjust the salinity concentration in the water bath. Ensure that the salinity (conductivity) in the water bath does not go out of conductivity logger range
- Record DO and conductivity readings with the multiparameter sonde at the same time the loggers record data
- Add sodium sulfite solution to lower the DO level in the water bath. This should be done slowly
  - NOTE: Do not leave equipment in this low-oxygen solution for too long. Two readings will suffice
• Record DO and conductivity readings with the multiparameter sonde at the same time the
loggers record data
• Logger data is retrieved from loggers and stored with multiparameter sonde readings. The
salinity correction will need to be applied through the HOBOware software for correct DO
concentration

B. Logger Inter Comparison (After Field Deployment)
• Bring all loggers back to office or lab
• Prepare a water bath with water similar to deployment stations; enough volume to fit all
loggers for comparison and sonde
• Place DO and conductivity loggers from the field in the water bath without cleaning any
fouling and note the time the loggers will be taking readings.
• Run the aquarium pump to keep water flowing in water bath. Do not keep loggers or sonde
close enough to allow bubbles to come into contact with sensors
• Record DO and conductivity readings with the multiparameter sonde at the same time the
loggers record data
• Clean all sensors before proceeding to the next steps
• Place DO and conductivity loggers ready for deployment in the water bath and note the time
the loggers will be taking readings.
• Run the aquarium pump to keep water flowing in water bath. Do not keep loggers or sonde
close enough to allow bubbles to come into contact with sensors
• Record DO and conductivity readings with the multiparameter sonde at the same time the
loggers record data
• Add ice or tap water to adjust the salinity concentration in the water bath. Ensure that the
salinity (conductivity) in the water bath does not go out of conductivity logger range
• Record DO and conductivity readings with the multiparameter sonde at the same time the
loggers record data
• Add sodium sulfite solution to lower the DO level in the water bath. This should be done
slowly
NOTE: Do not leave equipment in this low-oxygen solution for too long. Two readings will suffice

- Record DO and conductivity readings with the multiparameter sonde at the same time the loggers record data
- Logger data is retrieved from loggers and stored with multiparameter sonde readings. The salinity correction will need to be applied through the HOBOware software for correct DO concentration

C. FIELD DATA COLLECTION

1. Logger Deployment and Data Collection

- Dissolved oxygen and conductivity loggers are deployed at approximately 0.5 m off bottom. Loggers need to be actively logging when deployed to obtain initial sonde/logger comparison in field
- Barometric pressure logger is deployed above water at a secure and close location.
  - Logger needs to be actively logging prior to deployment
- All loggers are set to record at 15 minute intervals
- Station visits are required every 7-10 days for cleaning and comparison with multiparameter sonde
  - Conductivity, dissolved oxygen, and barometric pressure are all recorded at the station at 0.5 m off bottom using sonde and handheld
  - Readings must coincide with a known time the loggers are recording data
  - Record readings on field data sheet
- Retrieve submerged loggers and clean all fouling organisms
- Couple loggers with data retrieval method of choice and collect data for storage
- Repeat the comparison sampling procedure after the loggers are cleaned and redeployed
- Bring data retrieval instrument to the laboratory/office and upload data to analysis software provided by manufacturer
IX. QUICK SHEET

Refer to the SOP for details, this list is only a reminder!!

INTER COMPARISON
Zero DO solution needs to be added very slowly to avoid adding too much. Obtain a full reserve of water for use in the event the DO plummets quickly and below the preferred accuracy range of the instruments.

FIELD
Secure all loggers with backup ropes and security measures. All loggers need to be set to 15 minute intervals. Inter comparison readings are taken before and after cleaning loggers of fouling organisms.

STORAGE
Data sheets are filed and digital logs of data are stored on local computer/server prior to being sent to Save the Sound.

ANALYSIS
Upload data from field onto manufacturer software for analysis.
Appendix A-10

Standard Operating Procedure

Macrophyte Percent Coverage via Camera

revised January 2019

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I. OBJECTIVE: To quantify the percent coverage of macrophytes and determine bottom type at select locations.

II. OVERVIEW: An underwater camera (video or still) is used to capture images of the bottom in the area of a water quality station. The bottom type is identified and the percent cover of macrophytes is calculated.

III. SOURCE:

IV. MATERIALS AND EQUIPMENT:
A. FIELD COLLECTION AND STORAGE OF SAMPLES
   • GoPro camera
   • Setup for keeping camera a set distance from the bottom (to maintain consistent areal size in the field of vision)
   • GPS unit
B. LABORATORY ANALYSIS
   • not applicable

V. METHODS
A. PREPARATION
   • Check the charge on the GPS battery
   • Check the charge on the camera battery
   • Check the camera system for proper functioning
• Check the deployment rig for the camera system (must have a size reference in the field of vision)

B. FIELD COLLECTION AND STORAGE OF SAMPLES

• Samples stored on the SD card of the camera
• Write station, date, and time on a white board
• Approach station with camera and all equipment ready
• Start video or camera sequence with image of the white board and ensure GPS unit is tracking
• Run the boat slowly across a station starting around 15 m out and passing through it by around 15 m dropping pole to the floor waiting at least 3 seconds between drops. This can vary slightly but make sure the boat is moving. Multiple approaches from different directions to the station may be needed to collect the quantity of images required.
• Collect between 20 and 40 bottom images – in areas with greater heterogeneity, collect more samples (~40), if an area is fairly homogenous (e.g. sand, oyster, cobble) you may collect fewer (~20)
• Stop GPS track when navigating between stations
• Name GPS tracks using UWS station ID_date_time (24 hr)

C. LABORATORY ANALYSIS

• Not applicable

VI. TROUBLE SHOOTING / HINTS

• For the video system – keep the speed of the boat low, so as not to put too much tension on the pole (the pole will snap at high speeds)

VII. DATA PROCESSING AND STORAGE

• Video will be downloaded the following day and stored on computer and/or server/cloud storage.
• Download the GPS tracks the following day and store on computer as GPX file. Open GPX tracks in Garmin BaseCamp software and export as a CSV table.
• For video - A trained analyst will watch the video and do a screen-capture for all instances where the base of the pole is resting on the bottom (Figure 1).
• The bottom images will be pasted into the Excel camera work template (Figure 2).
The first analysts add GPS coordinates corresponding to the image located in the CSV table exported from BaseCamp. Analyst will match the time between the video and the CSV and add to template.

A 100 cell grid is overlaid on the image.

The first analyst reviews the image and enters the % cover in three categories: eelgrass, macroalgae, bare sediment. The first analyst hides their columns of data, so that other analysts cannot see the first set of estimates.

A second analyst reviews the images and makes an independent assessment of percent cover. The second analyst also checks the GPS coordinate data entry. The second analyst hides their columns of data, so that the other analysts cannot see their estimates.

A third analyst performs the same tasks as the second analyst.

The lab manager reviews a minimum of 10% of the images for accuracy of percent coverage estimates.

**INSERT ANY QUESTIONS INTO THE “COMMENTS” COLUMN**

In the Excel template, data are condensed into a table. Estimates from the three analysts are compared. If the relative percent difference among the three estimates is greater than 5%, the lab manager examines the image and the three estimates, choosing the appropriate value. The three estimates will not be changed, values are retained to show the inconsistency. The lab manager decides on the final value for the estimate. If a specific analyst’s estimate consistently differs, the analyst will receive further training or may be removed from conducting future estimates.

See Figure 3 for examples of coverage estimates and how to handle sparse coverage.
Figure 1: Image of the bottom captured using the video camera system. Note that the black circular disc on the end of the pole keeps the pole from sinking into the sediment and provides a size reference (diameter of disc = 10 cm).

Figure 2: The image from Figure 1 has been entered into the data template and overlaid with the 100 cell grid.

Figure 3: Examples and tips for estimating coverage.

- eelgrass = 100 %
- macroalgae = 0 %
- bare = 0 %

bare sediment is not visible, do not count the material (orange)
<table>
<thead>
<tr>
<th>Eelgrass</th>
<th>Macroalgae</th>
<th>Bare</th>
</tr>
</thead>
<tbody>
<tr>
<td>100%</td>
<td>0%</td>
<td>0%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>76%</td>
<td>0%</td>
<td>24%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>89%</td>
<td>0%</td>
<td>11%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>73%</td>
<td>0%</td>
<td>27%</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>69%</td>
<td>0%</td>
<td>31%</td>
</tr>
</tbody>
</table>

While leaves block some field of vision, this appears to be dense coverage.

The turbidity of the water makes this one tricky to analyze, zoom in and look closely.

Harder to estimate – take care counting – you will have to estimate partial grid cells to compute bare area.
### Standard Operating Procedure

**Macrophyte Percent Coverage via Camera**

<table>
<thead>
<tr>
<th>Section</th>
<th>Coverage Details</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eelgrass</td>
<td>66%</td>
</tr>
<tr>
<td>Macrophyte</td>
<td>21%</td>
</tr>
<tr>
<td>Bare</td>
<td>13%</td>
</tr>
</tbody>
</table>

Note: What appears to be Ulva on the left (comment entry with any %s). You will sometimes need to count more than once. (I counted macroalgae, then bare)

<table>
<thead>
<tr>
<th>Section</th>
<th>Coverage Details</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eelgrass</td>
<td>43%</td>
</tr>
<tr>
<td>Macrophyte</td>
<td>3%</td>
</tr>
<tr>
<td>Bare</td>
<td>54%</td>
</tr>
</tbody>
</table>

You can calculate the % coverage for two types, then calculate the third: 100 - 43 - 3 = 54

<table>
<thead>
<tr>
<th>Section</th>
<th>Coverage Details</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eelgrass</td>
<td>16%</td>
</tr>
<tr>
<td>Macrophyte</td>
<td>1%</td>
</tr>
<tr>
<td>Bare</td>
<td>83%</td>
</tr>
</tbody>
</table>

In a sparse eelgrass area, do not count the leaves that extend across the frame as area – look for the base of the plants.

<table>
<thead>
<tr>
<th>Section</th>
<th>Coverage Details</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eelgrass</td>
<td>0%</td>
</tr>
<tr>
<td>Macrophyte</td>
<td>13%</td>
</tr>
<tr>
<td>Bare</td>
<td>87%</td>
</tr>
</tbody>
</table>

Count shells as bare area, unless colonized by macroalgae.
VIII. REFERENCES:

IX. QUICK SHEET

Refer to the SOP for details, this list is only a reminder!!

FIELD

- Collect 20 – 40 images dependent on seafloor composition

STORAGE

- Download images and GPS tracks ASAP, at least by the day following field work

ANALYSIS

- Three analysts check
- Record to the nearest 1% of coverage
- Any questions go in the comments column
Appendix B-1
Calibration Datasheet
## UWS Field Datasheet

**EMBAYMENT NAME(S):**

**SAMPLE DATE:**

**PEOPLE:**

**TIME:**

- Tides nearest time of sampling, use NOAA Tide Tables
- Low Tide: ________  High Tide: ________

### APPROX. EXPECTED RANGE

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Expected Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Salinity (ppt/PSS)</td>
<td>3 – 30 ppt/PSS</td>
</tr>
<tr>
<td>Dissolved Oxygen (mg/L)</td>
<td>0 – 10 mg/L</td>
</tr>
<tr>
<td>Dissolved Oxygen (% Sat.)</td>
<td>0 – 100 %</td>
</tr>
<tr>
<td>Chlorophyll a (µg/L)</td>
<td>0 – 30 µg/L</td>
</tr>
<tr>
<td>Turbidity (NTU)</td>
<td>0 – 30 NTU</td>
</tr>
</tbody>
</table>

### WEATHER CONDITIONS

- 24 hours preceding time of sampling

<table>
<thead>
<tr>
<th>High Air Temperature (°C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low Air Temperature (°C)</td>
</tr>
<tr>
<td>Cloud Cover (%)</td>
</tr>
<tr>
<td>Precipitation State</td>
</tr>
<tr>
<td>Wind at Embayment</td>
</tr>
</tbody>
</table>

### Daily Precipitation

<table>
<thead>
<tr>
<th>Date</th>
<th>Inches</th>
</tr>
</thead>
</table>

### NOTES:

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________
### Appendix B-3

Sample Event Datasheet

<table>
<thead>
<tr>
<th>Enzyme Name</th>
<th>Sample Date</th>
<th>People</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Well No.</th>
<th>Time</th>
<th>Station Depth (m)</th>
<th>GPS</th>
<th>Depth (m)</th>
<th>%</th>
<th>Well</th>
<th>Station Depth (m)</th>
<th>GPS</th>
<th>Depth (m)</th>
<th>%</th>
<th>Well</th>
<th>Station Depth (m)</th>
<th>GPS</th>
<th>Depth (m)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Sample Depth (m)</th>
<th>Temperature [°C]</th>
<th>Salinity [ppt]</th>
<th>Dissolved Oxygen [%]</th>
<th>Dissolved Oxygen [mg/L]</th>
<th>Alkalinity [mg/L]</th>
<th>pH</th>
<th>Redox Potential [mV]</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Other AASHTO field notes are back of sheet if using different measurement units.

Data entry: __________

Sample Date: __________

Sample Time: __________

Data entry: __________

Sample Date: __________

Sample Time: __________
Appendix B-4
Chlorophyll Chain of Custody Form

<table>
<thead>
<tr>
<th>UWS Chlorophyll Chain of Custody</th>
<th>Organization: ________________</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample ID</td>
<td>Sample Date</td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
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<tr>
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<tr>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Relinquished by: ____________________  Date/Time: ____________________
Received by: ____________________  Date/Time: ____________________
Received by: ____________________  Date/Time: ____________________
Received by: ____________________  Date/Time: ____________________

Laboratory: ________________________
# Appendix B-5

## Qualitative Macrophyte Sampling Datasheet

<table>
<thead>
<tr>
<th>Site Name (short identifier)</th>
<th>Date</th>
</tr>
</thead>
<tbody>
<tr>
<td>People (full names)</td>
<td></td>
</tr>
<tr>
<td>Time of Low Tide</td>
<td>Time of Sampling (now)</td>
</tr>
<tr>
<td>Site Description / Notes (optional)</td>
<td></td>
</tr>
</tbody>
</table>

GPS location

For rake toss, complete 6 tosses. For soft shoreline, provide an overview photo and close-up photos of algae types. Remember to include something for a size reference in soft shoreline close-up photos (shoe, keys, ruler, etc.).

<table>
<thead>
<tr>
<th>Select one:</th>
<th>NONE</th>
<th>SOME</th>
<th>LOTS</th>
<th>green hair-like</th>
<th>green twig/leaf-like</th>
<th>green sheets</th>
<th>non-green hair-like</th>
<th>non-green twig/leaf-like</th>
<th>non-green sheets</th>
<th>marsh grass</th>
<th>eelgrass</th>
</tr>
</thead>
<tbody>
<tr>
<td>- soft shoreline</td>
<td>- rake toss</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**eed grass**
- green when fresh
- up to 6 ft. long
- may be brown or grey when decaying

**marsh grass**
- no drawing, look at land plants along shore for comparison

**hair-like**
- most of algae is the width of a hair
- may be slimy or dry
- may be tangled or straight
- most lose shape when removed from water, but not all

**twist/leaf like**
- most of algae is thicker than a hair
- may have small leaf-like sections
- may branch or not branch
- most maintain their shape when removed from water, but not all

*All drawings are to scale when printed on standard paper*

**sheets**
- may look like floppy lettuce leaves or rubbery strips (kelp)
- may be very large or the size of a quarter
- a few have a mid-rib (line up the middle), but most of plant does not have a "stem"
Appendix B-6
Macrophyte Field Guide

Long Island Sound Unified Water Study Algae Guide PAGE 1 of 2
All photos with permission from: Van Patten, Marguerite (Peg). (2009) Seaweeds of Long Island Sound, 2nd ed. Connecticut Sea Grant College Program. 104 pp. photographer is P. Van Patten, unless otherwise indicated.

green hair-like = very thin filaments, as thick as hair; may branch

non-green hair-like = very thin filaments, as thick as hair; may branch

sheets = most of plant is sheet form, may have a small holdfast

green non-green
Long Island Sound Unified Water Study Algae Guide  PAGE 2 of 2

All photos with permission from: Van Patten, Marguerite (Peg). (2009) Seaweeds of Long Island Sound, 2nd ed. Connecticut Sea Grant College Program. 104 pp. photographer is P. Van Patten, unless otherwise indicated.

**green twig/leaf-like** = thicker than a hair, may branch, typically maintains shape when removed form water

**eelgrass**

**non-green twig/leaf-like** = thicker than a hair, may branch, typically maintains shape when removed from water

"beneficial brown"
## Appendix B-7

**Nutrients Chain of Custody**

<table>
<thead>
<tr>
<th>UWS Nutrients Chain of Custody</th>
<th>Organization: ____________</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample ID</td>
<td>Sample Date</td>
</tr>
<tr>
<td>__________________________</td>
<td>__________________________</td>
</tr>
<tr>
<td>__________________________</td>
<td>__________________________</td>
</tr>
<tr>
<td>__________________________</td>
<td>__________________________</td>
</tr>
<tr>
<td>__________________________</td>
<td>__________________________</td>
</tr>
<tr>
<td>__________________________</td>
<td>__________________________</td>
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<tr>
<td>__________________________</td>
<td>__________________________</td>
</tr>
<tr>
<td>__________________________</td>
<td>__________________________</td>
</tr>
<tr>
<td>__________________________</td>
<td>__________________________</td>
</tr>
</tbody>
</table>

Relinquished by: __________________________ Date/Time: __________________________

Received by: __________________________ Date/Time: __________________________

Received by: __________________________ Date/Time: __________________________

Received by: __________________________ Date/Time: __________________________

Received by: __________________________ Date/Time: __________________________

Laboratory: __________________________
## Appendix B-8

### Nutrients Sampling Event Datasheet

<table>
<thead>
<tr>
<th>UWS Tier II - Surface Nutrients</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Date:</strong> ______________________</td>
</tr>
<tr>
<td><strong>People:</strong> _____________________</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>collect sample 0.5 m below surface</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>station (number, letter)</td>
<td></td>
</tr>
<tr>
<td>GPS N</td>
<td></td>
</tr>
<tr>
<td>GPS W</td>
<td></td>
</tr>
<tr>
<td>time (24 hour)</td>
<td></td>
</tr>
<tr>
<td>total station depth (m)</td>
<td></td>
</tr>
<tr>
<td>salinity (ppt)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Unfiltered</td>
</tr>
<tr>
<td>60 mL bottle ID</td>
<td>U</td>
</tr>
<tr>
<td>α thr C</td>
<td>α thr C</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>collect sample 0.5 m below surface</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>station (number, letter)</td>
<td></td>
</tr>
<tr>
<td>GPS N</td>
<td></td>
</tr>
<tr>
<td>GPS W</td>
<td></td>
</tr>
<tr>
<td>time (24 hour)</td>
<td></td>
</tr>
<tr>
<td>total station depth (m)</td>
<td></td>
</tr>
<tr>
<td>salinity (ppt)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Unfiltered</td>
</tr>
<tr>
<td>60 mL bottle ID</td>
<td>U</td>
</tr>
<tr>
<td>α thr C</td>
<td>α thr C</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>collect sample 0.5 m below surface</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>station (number, letter)</td>
<td></td>
</tr>
<tr>
<td>GPS N</td>
<td></td>
</tr>
<tr>
<td>GPS W</td>
<td></td>
</tr>
<tr>
<td>time (24 hour)</td>
<td></td>
</tr>
<tr>
<td>total station depth (m)</td>
<td></td>
</tr>
<tr>
<td>salinity (ppt)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Unfiltered</td>
</tr>
<tr>
<td>60 mL bottle ID</td>
<td>U</td>
</tr>
<tr>
<td>α thr C</td>
<td>α thr C</td>
</tr>
</tbody>
</table>
### Appendix B-9
Logger Retrieval Sample Event Datasheet

**UWS Logger Retrieval Data Sheet**

<table>
<thead>
<tr>
<th>Embayment Name</th>
<th>Station ID</th>
<th>GPS N</th>
<th>GPS W</th>
</tr>
</thead>
<tbody>
<tr>
<td>Date:</td>
<td>People:</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Barometric Pressure Unit**

<table>
<thead>
<tr>
<th>Serial#</th>
<th>Time of Retrieval</th>
</tr>
</thead>
</table>

**Dissolved Oxygen Logger**

<table>
<thead>
<tr>
<th>Serial#</th>
<th>Time of Retrieval</th>
</tr>
</thead>
</table>

#### Sonde Readings

<table>
<thead>
<tr>
<th>Time</th>
<th>Pre-retrieval</th>
<th>Post-retrieval</th>
</tr>
</thead>
<tbody>
<tr>
<td>Temperature (°C)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Salinity (ppt)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dissolved Oxygen (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dissolved Oxygen (mg/L)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Notes:**

---

**UWS Logger Retrieval Data Sheet**

<table>
<thead>
<tr>
<th>Embayment Name</th>
<th>Station ID</th>
<th>GPS N</th>
<th>GPS W</th>
</tr>
</thead>
<tbody>
<tr>
<td>Date:</td>
<td>People:</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Barometric Pressure Unit**

<table>
<thead>
<tr>
<th>Serial#</th>
<th>Time of Retrieval</th>
</tr>
</thead>
</table>

**Dissolved Oxygen Logger**

<table>
<thead>
<tr>
<th>Serial#</th>
<th>Time of Retrieval</th>
</tr>
</thead>
</table>

#### Sonde Readings

<table>
<thead>
<tr>
<th>Time</th>
<th>Pre-retrieval</th>
<th>Post-retrieval</th>
</tr>
</thead>
<tbody>
<tr>
<td>Temperature (°C)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Salinity (ppt)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dissolved Oxygen (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dissolved Oxygen (mg/L)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Notes:**

---

**UWS Logger Retrieval Data Sheet**

<table>
<thead>
<tr>
<th>Embayment Name</th>
<th>Station ID</th>
<th>GPS N</th>
<th>GPS W</th>
</tr>
</thead>
<tbody>
<tr>
<td>Date:</td>
<td>People:</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Barometric Pressure Unit**

<table>
<thead>
<tr>
<th>Serial#</th>
<th>Time of Retrieval</th>
</tr>
</thead>
</table>

**Dissolved Oxygen Logger**

<table>
<thead>
<tr>
<th>Serial#</th>
<th>Time of Retrieval</th>
</tr>
</thead>
</table>

#### Sonde Readings

<table>
<thead>
<tr>
<th>Time</th>
<th>Pre-retrieval</th>
<th>Post-retrieval</th>
</tr>
</thead>
<tbody>
<tr>
<td>Temperature (°C)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Salinity (ppt)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dissolved Oxygen (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dissolved Oxygen (mg/L)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Notes:**
## Appendix B-10

**Macrophyte Percentage Coverage Datasheet**

<table>
<thead>
<tr>
<th>People</th>
<th>Location</th>
<th>Site Code</th>
<th>Start Time</th>
<th>Stop Time</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sta</td>
<td>video time</td>
<td>ID on White Board</td>
<td>Notes</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sta</td>
<td>video time</td>
<td>ID on White Board</td>
<td>Notes</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sta</td>
<td>video time</td>
<td>ID on White Board</td>
<td>Notes</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sta</td>
<td>video time</td>
<td>ID on White Board</td>
<td>Notes</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sta</td>
<td>video time</td>
<td>ID on White Board</td>
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<td></td>
</tr>
<tr>
<td>Sta</td>
<td>video time</td>
<td>ID on White Board</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Sta</td>
<td>video time</td>
<td>ID on White Board</td>
<td>Notes</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sta</td>
<td>video time</td>
<td>ID on White Board</td>
<td>Notes</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sta</td>
<td>video time</td>
<td>ID on White Board</td>
<td>Notes</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sta</td>
<td>video time</td>
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Appendix C-1

Chlorophyll $a$

1) Test Method

Based on EPA 445.0

2) Applicable Matrix or Matrices

Non-potable Water

3) Method Detection Limit

This Method’s Detection Limit is 0.7µg/L. The laboratory’s reporting level for this method is 2.1µg/L.

4) Scope and Application

This method is useful for estimating phytoplankton biomass in freshwater and marine environments. IEC will use it to analyze marine waters for chlorophyll $a$.

5) Summary of Method

A 400mL, or otherwise specified, aliquot of natural water is filtered in a dark area. Pigment is extracted from the filter through maceration then steeping in a 90% acetone soak, and clarified using a centrifuge. An aliquot of the supernatant is transferred to a glass cuvette and fluorescence is measured before and after acidification to 0.003N HCl with 0.1N HCl. The pigment extract is then analyzed using a fluorometer. Addition of acid results in the loss of the magnesium atom, converting chlorophyll $a$ to pheophytin $a$. The change in fluorescence after acidification is used to determine the corrected values for chlorophyll $a$.

6) Definitions

Chlorophyll $a$ is a photosynthetic pigment. It is a component of planktonic algae, constituting 1-2% of its dry weight. Chlorophyll $a$ is used extensively to estimate phytoplankton biomass.

7) Interferences

Any substance extracted from the filter or acquired from laboratory contamination that fluoresces in the red region of the spectrum may interfere in the accurate measurement of chlorophyll $a$. 
Fluorescence is temperature dependent. Light, changes in temperature, and exposure to air can also interfere with the test, leading to decreases in chlorophyll a concentration. Use care in dealing with samples during the analysis process. Samples, standards, blanks and quality control samples must be at the same temperature to prevent errors and/or low precision. Analyses of samples at ambient temperature is recommended.

All work must be performed in subdued light. QC samples and filters must be stored at -20°C to -70°C to prevent degradation.

Excess sample turbidity can interfere with proper fluorescence readings.

8) Safety

Personal protective gear, including gloves, goggles and a lab coat must be worn by analysts during all steps of the analysis. Work under a hood whenever possible. Please refer to the MSDS (on file in the yellow book on wall by micro room door) for specific information on toxicity and safety precautions needed for specific chemicals.

9) Equipment and Supplies

9.1 Turner® Trilogy Laboratory Fluorometer, equipped with chlorophyll a acidification module.

9.2 12 x 35 mm or 12 x 75 mm glass test tubes

9.3 Whatman glass microfiber filters GF/F-47 mm or equivalent (0.45-µm porosity)

9.4 47 mm solvent resistant filter funnels,

9.5 1000 mL side-arm filtering flasks or vacuum filtration manifold

9.6 Vacuum pump and tubing

9.7 Graduated Cylinders

9.8 15 mL centrifuge tubes

9.9 stainless steel forceps (for transferring filter)

9.10 1000 µL Brinkmann Eppendorf micropipette with adjustable dispensing volume feature

9.11 1 liter volumetric flask

9.12 freezer

9.13 Analytical balance
9.14 IEC Clinical Centrifuge 120V 1.2 Amp Model 41498

9.15 4°C fridge

9.16 tissue grinder and pestle

10) Reagents

10.1 Deionized Water

10.2 Mix together 90 mL acetone, 10 mL of deionized water

10.3 0.1N Hydrochloric Acid: Add 8.5 mL of concentrated hydrochloric acid to a 1 L flask containing 500 mL of distilled water. Cool and dilute to the mark with distilled water. Stir to mix. Or use purchased, pre-prepared 0.1N HCL procured with manufacturer’s certificate of analysis, which must be retained on file in the laboratory

10.4 Turner Designs (P/N 10-850) Fluorometric Chlorophyll Standards in 90% acetone, low and high concentration standards. They are used to find the lower absorbance detection limit of the spectrophotometer (MDL) and to prepare check standards. Typically have a 1 year shelf life (manufacturer assigns expiration date) and must be stored in the freezer.

10.5 Turner Designs Chlorophyll a in 90% acetone, stock standard. Transfer 10 mL from a Turner Designs chlorophyll a standard ampoule into a 500 mL flask and dilute to the mark with 90% acetone. Use chilled pipettes and flasks when making transfers. Calculate the stock standard concentration using the original concentration of the ampouled standard. Use until manufacturer-assigned expiration date of original ampoule, if stored frozen.

10.6 Turner Designs Adjustable Solid Secondary Check Standards, P/N 8000-952

11) Sample Collection, Preservation, Shipment and Storage

For the Unified Water Study, samples are field-filtered according to UWS-approved QAPP and sampling SOP. Filters are then packaged, frozen and forwarded to IEC laboratory as per established UWS procedure and approved QAPP. See UWS QAPP and sampling SOP for details.

Samples are collected directly into plastic opaque 500 mL sample bottles using a sludge nabber sampling stick. Bottles are marked with waterproof ink with a specific sample ID number, station identifying number, date and investigation number. On a chain of custody sheet record date and time collected, cruise number (e.g. LISS#1), sampling station, IEC investigation number, personnel, and type of analysis (chlorophyll a). Place sample bottles in a cooler containing ice and a cooler thermometer. Minimize the frequency and extent to which the cooler lid is opened, to minimize exposure to light. Upon arrival at the laboratory, record time
transferred to laboratory on the chain of custody sheet. Log samples in the laboratory’s sample log-in book. Information to be included in the designated columns in the log-in book include: investigation #, # of bottles, bottle condition, date in lab, time in lab, cooler temperature, run #, parameter(s) to be analyzed, and sampler’s initials. Turn off laboratory light and transfer samples to sample refrigerator. Filtration must be completed within 48 hours after samples are collected.

12) Quality Control

Blanks are analyzed at a frequency of one per batch at the end of filtration. The method blank consists of a filter placed on the filtration set up with the vacuum turned on for 1 minute to draw air through filter. This blank should be less than the calculated method lower detection limit for the analysis. A Turner Designs secondary check standard must be analyzed at the beginning and the end of each analytical batch.

13) Calibration and Standardization

Calibration should be performed bimonthly (during months when method is being used) or more frequently if an adjustment made to the instrument. Prepare 0.2, 2, 5, 20 and 200 µg/L calibration standards from stock solutions. Additional standards may be prepared to tailor the calibration to the concentration range of samples collected. Direct Calibration Procedure: Turn on the Trilogy. Wait 15 minutes to allow the instrument to warm up. Touch “Chl-A” to select the Chlorophyll a Acidification module and confirm by touching “OK.” On the home screen, touch “Calibrate” to begin a calibration sequence. Select “Run New Calibration.” Select the unit of measurement.

Insert calibration “blank” and touch “OK.” Enter the concentration for the first standard. If using the Turner Designs Chlorophyll a standards, this will be the concentration data supplied with the standard. Follow the screen prompt indicating that the standard before acidification (Fb) should be inserted. Insert sample and touch “OK.” Now insert the standard after acidification and press “OK.” The (Fa) value will be measured and the ratio of the two readings will be displayed as seen in the next step. If the ratio is in the required range, touch “OK.” The ratio will be stored in the Trilogy for use in the measurement of chlorophyll a. After the calibration is complete, either select “Proceed with Current Calibration” or select “Enter More Standards,” in which case, enter the concentration for the next standard as was done above with the first standard. Name and save the calibration for future use. Measure the solid standard and record the displayed value to enable a quick calibration verification.

14) Procedure

14.1 Sample Filtration
For the Unified Water Study, samples are field-filtered according to UWS-approved QAPP and sampling SOP. Filters are then packaged, frozen and forwarded to IEC laboratory as per established UWS procedure and approved QAPP. See UWS QAPP and sampling SOP for details.

Conduct work with chlorophyll extracts in subdued light to avoid degradation. Turn off excess laboratory overhead lights and close blinds. Measure 400 mL (or other suitable aliquot) of a well-mixed sample into a 500 mL graduated cylinder and filter. The sample must be kept well-mixed. When pouring a measured volume into a filter funnel, leave a small amount in the cylinder and swirl it well before pouring the final amount. Record volumes filtered for each sample on the chlorophyll a data sheet. At least one sample per batch must be filtered in duplicate. Both duplicates must have identical volumes filtered. All graduated cylinders and filtering funnels must be rinsed 2-3 times with deionized water between samples. Changes to the standard 400 mL volume filtered can and should be made if suspended material concentrations are high and the sample is taking 5 minutes or more to filter, however, duplicate filters must have the identical volume of sample filtered through them. Filters must be handled with clean forceps only. Using clean forceps place the filter on the filter funnel by placing the finer mesh side of the filter face down. Be careful to center the filter on the filter holder so that sample does not seep around the filter. Also be careful not to slide the filter off-center when placing the funnel or the clamp on. After filtration, carefully fold and transfer to a 15 mL capped centrifuge tube. Make sure that all tubes are labeled with the correct sample number. Samples on filters taken from water having a pH 7 or higher may be placed in airtight centrifuge tubes and stored frozen in the dark for 3 weeks. Samples from acidic water must be processed promptly to prevent chlorophyll degradation. If samples are to be analyzed immediately go to the next step, 14.2. Place sample filters in the dark in the freezer for analysis at a later date.

14.2 Extraction

If the samples have been placed in a freezer, remove them from the freezer but keep them in the dark. Workspace lighting should be kept to a minimum. Remove a filter from its container and place it into the centrifuge tube. The filter may be torn into smaller pieces to facilitate extraction. With a volumetric pipette add 10 mL of the 90% acetone solution. Cap the tube and shake vigorously (vortex). Place it in the dark before proceeding to the next filter extraction. Shake each tube vigorously (vortex) before placing them to steep in the dark at 4°C. Samples should be allowed to steep for a minimum of 2 h but not to exceed 24 h.

Glass fiber filters of 47 mm diameter have dry displacement volumes of 0.10 mL and introduce errors of about 1.0% if a 10 mL extract is used. Clarify by inserting capped centrifuge tubes in centrifuge. Place tubes with similar volumes (within 0.5 mL) opposite each other in centrifuge to maintain centrifuge balance. Centrifuge by incrementally increasing speed to between level 6 and level 7 to the approximation of 675 g. Centrifuge for 15 minutes, reduce speed slowly until
centrifuge stops completely. Decant clarified extract into a clean, 10 mL cuvette. Record final extract volume by comparing volume to graduated cuvette. Note final extract volume for use as V1 in calculations, below. Remove tubes from freezer and shake. Allow tubes to reach room temperature. Decant 10 mL of clarified extract into a clean 12 x 35 mm or 12 x 75 mm glass test tube.

14.3 Determination of Chlorophyll a

Upon completion of the required calibration steps of section 13, insert a blank cuvette containing 90% aqueous acetone solution.

The volume of extract and acid and the time after acidification are critical for accurate, consistent results. For a test tube that holds 5 mL of extract, 0.15 mL of the 0.1N HCl solution should be used. For a test tube (12mm x 75mm) that holds 10mL of extract, 0.30 mL of the 0.1N HCL solution should be used.

On the Trilogy touchscreen touch “Sample ID” to name your sample

Using the keypad, enter the sample name into the name field and touch “Save”.

Touch “Measure Fluorescence” to make a measurement. The Trilogy will measure the sample for 6 seconds and report the average reading for the sample.

Record the fluorescence measurement of the sample. Remove the test tube from the fluorometer and acidify the extract to a final concentration of 0.003N HCl using the 0.1N HCl. Use a Pasteur pipet to thoroughly mix the sample by aspirating and dispensing the sample into the test tube, keeping the pipet tip below the surface of the liquid to avoid aerating the sample. Wait exactly 90s (using a preset timer!) before measuring fluorescence again. NOTE: Proper acidification, mixing and timing is CRITICAL for precise and accurate results.

15) Calculations

Measure the fluorescence of each standard at sensitivity setting that provide midscale readings. Obtain response factors for chlorophyll a for each sensitivity setting as follows:

\[ F_s = C_s/R_s \]

Where:

\[ F_s = \text{response factor for sensitivity setting, } S. \]

\[ R_s = \text{fluorometer reading for sensitivity setting, } S. \]
C = concentration of chlorophyll a

Obtain before-to-after acidification response ratios of the chlorophyll a calibration standards as follows:

1. Measure the fluorescence of the standard, (2) remove the test tube from the fluorometer, (3) acidify the solution to 0.003N HCl with the 0.1N HCl solution, (4) use a Pasteur pipet to thoroughly mix the sample by aspirating and dispensing the sample into the test tube, keeping the pipet tip below the surface of the liquid to avoid aerating the sample, (5) wait exactly 90 s (use preset laboratory timer) and measure the fluorescence of the standard solution again. For a test tube that holds 5 mL of extract, it will be necessary to add 0.15 mL of 0.1N HCl to reach a final acid concentration of 0.003N in the 5 mL (use 0.30mL for a 10 mL extract).

Calculate the ratio, r, as follows:

\[ r = \frac{R_b}{R_a} \]

Where:

- \( R_b \) = fluorescence of pure chlorophyll a standard before acidification
- \( R_a \) = fluorescence of pure chlorophyll a standard after acidification

For “corrected chlorophyll a”, calculate the chlorophyll a concentration in the extract as:

\[ C_{E,c} = F_s \cdot \frac{(r/r-1) \cdot (R_b-R_a)}{r} \]

Where:

- \( C_{E,c} \) = corrected chlorophyll a concentration (μg/L) in the extract solution analyzed,
- \( F_s \) = response factor for the sensitivity setting, S
- \( r \) = the before-to-after acidification ratio of a pure chlorophyll a solution
- \( R_b \) = fluorescence of pure chlorophyll a standard before acidification
- \( R_a \) = fluorescence of pure chlorophyll a standard after acidification

Calculate the “corrected” concentration of chlorophyll a in the whole water sample as follows:

\[ C_{s,c} = C_{E,c} \times \frac{\text{extract volume (L)} \times \text{DF}}{\text{sample volume (L)}} \]
Where $C_{a,c} =$ corrected chlorophyll $a$ concentration ($\mu g/L$) in the whole water sample

Extract volume = volume (L) of extract prepared before dilution

The Relative Percent Difference (RPD) of duplicate determinations should be within 15%. RPD is defined as the absolute value of the difference of duplicate determinations divided by their arithmetic mean and multiplied by 100.

16) Method Performance

Method performance is evaluated by ongoing analysis of QC check standards for each batch.

17) Pollution Prevention

Dispose of waste as in section 21, Waste Management, to prevent pollution. Store waste in a container within a secondary container to prevent pollution due to spills or container leakage.

18) Data Assessment and Acceptance Criteria for Quality Control Measures

Refer to the Turner Designs certificate of analysis to determine the latest control limits for check standard % recovery.

19) Corrective Actions for Out-of-Control Data

Duplicate determinations should be averaged to determine the reported result. Duplicate determinations should have acceptable RPD values, as noted in Section 15. The deionized water and reagents and supplies may need to be evaluated for possible contamination. If the % recovery for the chlorophyll QC check standard does not meet acceptable limits, the system has to be evaluated for possible errors. Prepare a fresh standard and re-analyze or purchase a new lot to attempt to determine if the standard has degraded or if there is an error in the procedure. If the manufacturer of the QC check standards does not provide acceptance limits, an acceptable recovery of $\pm 15\%$ could be assigned until one is determined by the Laboratory’s Directors by spiking samples of known concentration.

20) Contingencies for Handling Out-of-Control or Unacceptable Data

Ideally samples yielding out-of-control or unacceptable QC results should be reanalyzed. If there is insufficient sample for reanalysis or the sample holding time has expired, analytical results must be reported, along with all unsatisfactory quality control measures or reported as “No Result” because of unsatisfactory QC measures. In all cases, the out-of-control result must be recorded in the analyst’s logbook and the QA officer notified.
21) Waste Management

Waste is disposed of in an appropriate acetone waste collection bottle. Waste pick-up is arranged for, on an as-needed basis, approximately every 6 months with Environmental Products and Services, Inc, or other suitable waste transporter.

22) References

USEPA Method 445.0 In Vitro Determination of Chlorophyll a and Phaeophytin a in Marine and Freshwater Algae by Fluorescence


23) Tables, Diagrams, Flowcharts and Validation Data

None
Appendix C-2

Ammonia

1. TEST METHOD

Based on Lachat Method 10-107-6-1B (2001).

2. APPLICABLE MATRIX OR MATRICES

The method is applicable for non-potable water (ambient surface water and wastewater).

3. METHOD DETECTION LIMIT

The method detection limit is 0.100 mg/L as NH₃. The reporting limit is 0.249 mg/L.

4. SCOPE AND APPLICATION

4.1 This method covers the determination of ammonia in surface waters, and domestic and industrial wastes. IEC will be using for the analysis of samples collected in western LIS (brackish samples) as well as periodic samples collected from wastewater treatment plants, as applicable.

5. SUMMARY OF METHOD

5.1 This method is based on the Berthelot reaction. Ammonia reacts with alkaline phenol, and sodium hypochlorite to form indophenol blue to form monochloramine, which, in the presence of phenol, catalytic amounts of nitroprusside (nitroferricyanide), and excess hypochlorite gives indophenol blue. The formation of monochloramine requires a pH between 8 and 11.5. At higher pH, ammonia may begin to oxidize to nitrate. At pH greater than 9.6, some precipitation of calcium and magnesium as hydroxides and carbonates occurs in seawater. EDTA added to the buffer prevents this from occurring. The indophenol blue measured at 630 nm is proportional to the original ammonia concentration.

The method is calibrated using standards prepared in deionized water. Once calibrated, samples of varying salinities (0 to 35 ppt) may be analyzed. The determination of background absorbance is necessary only for samples which have color absorbing at 630 nm.

If distillation is required, the sample is buffered at pH of 9.5 with a borate buffer to decrease hydrolysis of cyanates and organic nitrogen compounds, and is distilled into a solution of boric acid.
6. DEFINITIONS

The definitions and purposes below are specific to this method, but have been conformed to common usage as much as possible.

6.1 CALIBRATION BLANK (CB) – A volume of reagent water in the same matrix as the calibration standards, but without the analyte.

6.2 CALIBRATION STANDARD (CAL) -- A solution prepared from the primary dilution standard solution or stock standard solutions. The CAL solutions are used to calibrate the instrument response with respect to analyte concentration.

6.3 INSTRUMENT PERFORMANCE CHECK SOLUTION (ICP) -- A solution of one or more method analytes used to evaluate the performance of the instrument system with respect to a defined set of criteria.

6.4 LABORATORY SPIKED BLANK (LSB) -- An aliquot of reagent water or other blank matrices to which known quantities of the method analytes are added in the laboratory. The LSB is analyzed exactly like a sample, and its purpose is to determine whether the methodology is in control, and whether the laboratory is capable of making accurate and precise measurements.

6.5 LABORATORY SPIKED SAMPLE MATRIX (LSM) -- An aliquot of an environmental sample to which known quantities of the method analytes are added in the laboratory. The LSM is analyzed exactly like a sample, and its purpose is to determine whether the sample matrix contributes bias to the analytical results. The background concentrations of the analytes in the sample matrix must be determined in a separate aliquot and the measured values in the LFM correct for background concentrations.

6.6 LABORATORY YY REAGENT BLANK (LRB) – An aliquot of reagent water or equivalent neutral reference material treated as a sample in all aspects, except that it is not taken to the sampling site. The purpose is to determine if the analytes or interferences are present in the laboratory environment, the reagents, or the apparatus.

6.7 LINEAR CALIBRATION RANGE (LCR) – The concentration range over which the instrument response is linear.

6.8 MATERIAL SAFETY DATA SHEETS (MSDS) – Written information provided by vendors concerning a chemical’s toxicity, health hazards, physical properties, fire, and reactivity data including storage, spill, and handling precautions.
6.9 METHOD DETECTION LIMIT (MDL) – The lowest level at which an analyte can be detection with 99 percent confidence that the analyte concentration is great than zero.

6.10 LIMIT OF QUANTITATION (LOQ) – The Laboratory performs a Limit of Quantitation (LOQ) study on an annual basis for analytes associated with chemistry methods. The validity of LOQ is confirmed by successful analysis of a Laboratory Fortified Blank (LFB) at approximately 2X the reporting limit. The acceptance criteria for each analyte is + 30% of the true value. After this study is completed, it is reviewed and approved by the Laboratory Management. A summary of all LOQ study performance is maintained.

6.11 QUALITY CONTROL SAMPLE (QCS) – A solution of method analytes of known concentrations that is used to spike an aliquot of LRB or sample matrix. THE QCS is obtained for a source external to the laboratory or is prepared from standards obtained from a different source than the calibration standards. The purpose is to check laboratory performance using test materials that have been prepared independently for the normal preparation process.

6.12 STOCK STANDARD SOLUTION (SSS) – A concentrated solution containing one or more method anaytes prepared in the laboratory using assay reference materials or purchased from a reputable commercial source.

7. INTERFERENCES

7.1 Calcium and magnesium ions may precipitate if present in sufficient concentration. EDTA is added to the sample in-line to prevent this problem.

7.2 Color, turbidity, and certain organic species may interfere. Turbidity is removed by manual filtration. Sample color may be corrected for by running the samples through the manifold without color formation. See System Note 11 for specific instructions.

7.3 Sulfide may interfere at levels greater than 2 mg H_2S/L. Samples containing concentrations greater than this should be diluted.

7.4 Salinity does not normally interfere in this method. This may be verified by running the samples through the manifold with all reagents pumping, except hypochlorite, which is replaced by deionized water. The resulting concentrations are then compared to those obtained for samples determined with color formation.

7.5 The salt effect (salinity influence on absorbance) is less than 2%.

See System Note 11 for specific instructions.
8. SAFETY

8.1 The toxicity or carcinogenicity of each reagent used in this method has not been fully established. Each chemical should be regarded as a potential health hazard and exposure should be as low as reasonably achievable. Cautions are included for known extremely hazardous materials.

8.2 Each laboratory is responsible for maintaining a current awareness file of the Occupational Health and Safety Act (OSHA) regulation regarding the safe handling of the chemicals specified in this method. A reference file of Material Safety Data sheets (MSDS) is made available to all personnel involved in the chemical analysis.

8.3 The following chemicals have the potential to be highly toxic or hazardous, for detailed explanation consult the MSDS.

   5.3.1. Sodium hydroxide
   5.3.2. Phenol

9. EQUIPMENT AND SUPPLIES

9.1 Balance – analytical, capable of accurately weighing to the nearest 0.0001 g.
9.2 Glassware – Class A volumetric flasks and pipettes or plastic containers as required. Samples may be stored in plastic or glass.
9.3 Flow injection analysis equipment (Lachat 8500 series 2) designed to deliver and react sample and reagents in the required order and ratios.

   9.3.1. Lachat Autoanalyzer
   9.3.2. Multichannel proportioning pump
   9.3.3. Reaction unit or manifold
   9.3.4. Colorimetric detector
   9.3.5. Data system

9.4 Special Apparatus

   6.4.1. Seawater Accessories Kit Lachat Part No. 50969 – RAS or 50970 – ASX510 (for brackish samples)
   6.4.2. PVC PUMP TUBES MUST BE USED FOR THIS METHOD.
10. REAGENTS AND STANDARDS

10.1 PREPARATION OF REAGENTS

Use deionized water (10 megohm) for all solutions.

Degassing with helium:

To prevent bubble formation, DO NOT DEGAS USING AN INVASIVE PROCEDURE SUCH AS A WAND TO AVOID CONTAMINATION. Degas by vacuum or sonication. DO NOT DEGAS PHENATE, HYPOCHLORITE, OR STANDARDS.

Reagent 1. Buffer Chelating Agent

**By Volume:** In a 1 L volumetric flask, dissolve 50.0 g disodium ethylenediamine tetraacetate (Na₂EDTA) and 11.0 g sodium hydroxide (NaOH) in about 900 mL DI water. Stir to mix and dilute to the mark with DI water. Degas as above. Prepare fresh monthly sample.

**By Weight:** To a tared 1 L container, add 966 g DI water. Add 50.0 g disodium ethylenediamine tetraacetate (Na₂EDTA) and 11.0 g sodium hydroxide (NaOH). Stir to mix. Degas as above. Prepare fresh monthly.

*Preferably* use Lachat Ammonia Buffer, EDTA/NaOH Solution CAT#52017.

Reagent 2. Phenate Reagent

**CAUTION:** Wear gloves. Phenol causes skin burns and is rapidly absorbed into the body through the skin. **Do not degas this reagent.**

**By Volume:** In a 1 L volumetric flask, dissolve 83 g crystalline phenol (C₆H₅OH) in approximately 500 mL DI water. While stirring, slowly add 32 g sodium hydroxide (NaOH). Cool, dilute to the mark with DI water, and invert to mix. The color of this reagent darkens with age, increasing the baseline absorbance. Prepare fresh reagent after 72 hours. Prepare fresh every 3-5 days and discard when turns dark brown.

**By Weight:** To a tared 1 L container, add 88 g DI water. Add 83 g crystalline phenol (C₆H₅OH). While stirring, slowly add 32 g sodium hydroxide (NaOH). Cool and stir to mix. The color of this reagent darkens with age, increasing the baseline absorbance. Prepare fresh every 3-5 days and discard when turns dark brown.

*Preferably* use Lachat Sodium Phenolate Reagent CAT#52005.

Reagent 3. Sodium Hypochlorite

**By Volume:** Dilute 250 mL sodium hypochlorite (SS290-1 hypochlorite solution, 4-6% NaOCl
from Fisher) to 500 mL with DI water. Prepare fresh daily.

**By Weight:** Dilute 250 g sodium hypochlorite (SS290-l hypochlorite solution, 4-6% NaOCl from Fisher) to 500 g with DI water. Prepare fresh daily.

**Preferably** use Lachat Hypochlorite Reagent CAT#52007.

**Reagent 4. Sodium Nitroprusside**

**By Volume:** Dissolve 1.75 g sodium nitroprusside in 500 mL DI water. Prepare fresh every 1-2 weeks.

**By Weight:** Dissolve 1.75 g sodium nitroprusside in 500 mL DI water. Prepare fresh every 1-2 weeks.

**Preferably** use Lachat Sodium Nitroprusside Reagent CAT#52006.

**OR USE LACHAT AMMONIA REAGENT SET CAT # 52904 as an alternative option.**

### 10.2 PREPARATION OF STANDARDS

**Standard 1. Stock Ammonia Standard, 100 mg/L**

**By Volume:** In a 1 L volumetric flask dissolve 0.3818 g ammonium chloride (NH₄Cl) FISHER CAT #3384-12 (Macron) in about 800 mL DI water. Dilute to the mark with DI water and invert to mix.

**Standard 2. Working Stock Standard 10.0 mg/L**

Dilute 10.0 mL of Stock Ammonia Standard to 100 mL with DI water.

**Standards 3. Working Standards.**

Dilute the following mL of working stock standard to 100 mL. The upper and lower value of the standard curve should not be modified. If project requires a different range the upper and lower standards may be modified but reporting limit must also be adjusted accordingly. Samples above the high standard (+10%) must be diluted. The mid-range standards listed below are recommended but may be modified by analyst.

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<th>mL Working Stock</th>
<th>mg/L Ammonia</th>
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11. SAMPLE COLLECTION, PRESERVATION AND STORAGE

For the Unified Water Study, samples are field-filtered according to UWS-approved QAPP and sampling SOP. Filters are then packaged, frozen and forwarded to IEC laboratory as per established UWS procedure and approved QAPP. See UWS QAPP and sampling SOP for details.

11.1 There is no single preservation method that may be recommended of all types of samples. The analyst must examine each situation critically and treat this information as a guide.

11.2 If samples must be chemically preserved, samples should be cooled, and adjusted to pH < 2 with H₂SO₄. And store at 4 °C in glass or polyethylene. Analyze within 28 days. Samples collected from western Long Island Sound sampling and stored in polyethylene bottles may be stored frozen up to 1 year. If samples are not run within 24 hours a storage stability study should be done.

11.3 Researchers have found serious errors when investigating the effects of filtration. The analyst should examine sample preparation and preservation techniques before routine testing.

12. QUALITY CONTROL

12.1 Each laboratory using this method is required to operate a formal quality control (QC) program. The minimum requirements of this program consist of an initial demonstration of laboratory capability, and the periodic analysis of laboratory reagent blanks, fortified blanks and other laboratory solutions as a continuing check on performance. The laboratory is required to maintain performance records that define the quality of the data that are generated.

12.2 INITIAL DEMONSTRATION OF PERFORMANCE

12.2.1. The initial demonstration of performance is used to characterize instrument performance (determination of LCRs and analysis of QCS) and laboratory performance (determination of MDLs) prior to performing analyses by this method.

12.2.2. Linear Calibration Range (LCR) --- The LCR must be determined initially and verified every 6 months or whenever a significant change in instrument response is observed and expected. The initial demonstration of linearity must use sufficient standards to insure that the resulting curve is linear. The verification of linearity must use a minimum of a blank and three standards. If any verification data exceeds the initial values by +/- 10%, linearity must be nonlinear, sufficient standards must be used to clearly define the nonlinear portion.

12.2.3. Quality Control Sample (QCS) or Laboratory Control Sample (LCS)— When
beginning the use of this method, and as required to meet data-quality needs, verify the calibration standards are acceptable instrument performance with the preparation and analyses of a QCS. If the determined concentrations are not within $\pm 10\%$ of the stated values, performance of the determinative step of the method is unacceptable. The source of the problem must be identified and corrected before either proceeding with the initial determination of MDLs or continuing with ongoing analyses.

12.2.4. Method Detection Limit (MDL) – MDLs must be established for all analytes, using reagent water (blank) fortified at a concentration of two to three times the estimated instrument detection limit. To determine MDL values, take seven replicate aliquots of the fortified reagent water and process through the entire analytical method. Perform all calculations defined in the method and report the concentration values in the appropriate units. Calculate MDLs as follows:

$$\text{MDL} = t \times S$$

Where, $t =$ Student’s $t$ value for a 99% confidence level and a standard deviation estimate with $n-1$ degrees of freedom [$t = 3.14$ for seven replicates, $t = 2.528$ for twenty-one replicates]. $S =$ standard deviation of the replicate analyses.

MDLs should be determined every six months, when a new operator begins work, or whenever there is a significant change in the background or instrument response. The reporting limit is 5X the calculated MDL and must be verified at least annual, or as required for data-quality needs.

13. CALIBRATION AND STANDARDIZATION

13.1 Prepare a series of standards, covering the desired range, and a blank by dilution suitable volumes of standard solution (suggested range in Section 10.2).

13.2 Calibrate the instrument as described in Section.

13.3 Prepare standard curve by plotting instrument response against concentration values. A calibration curve may be fitted to the calibration solution concentration/response data using the computer. Acceptance or control limits should be established using the difference between the measured value of the calibration solution and the “true value” concentration.

13.4 After the calibration has established, it must be verified by the analysis of a suitable quality control sample (QCS). If measurements exceed $\pm 10\%$ of the established QCS value, the analysis should be terminated and the instrument recalibrated. The new calibration must be verified before continuing analysis. Periodic reanalysis of the QCS is recommended as a continuing calibration check.

14. PROCEDURE

14.1 CALIBRATION PROCEDURE
14.1.1 Prepare reagent and standards as described in section 10.
14.1.2 Set up manifold as shown in Lachat Ammonia Manual.
14.1.3 Input data system parameters as shown in Lachat Ammonia Manual.
14.1.4 Pump DI water through all reagent lines and check for leaks and smooth flow. Switch to reagents and allow the system to equilibrate until a stable baseline is achieved.
14.1.5 Place samples and/or standards. Input the information required by the data system, such as concentration, replicates, etc. (See section 14.2).
14.1.6 Calibrate the instrument by injecting the standards. The data system will then associate the concentrations with the instrument responses for each standard.

14.2 SYSTEM NOTES
14.2.1 For information on system maintenance and troubleshooting refer to the Troubleshooting Guide in the System Operation Manual. This guide is also available on request from Lachat.
14.2.2 Allow 15 minutes for heating module to warm up to 60 °C.
14.2.3 Allow 15 minutes with reagents pumping for the system to equilibrate.
14.2.4 To check for gross carrier contamination:
   A. Monitor the baseline on the system unit screen.
   B. Equilibrate the pumping system with DI water in all reagent lines.
   C. Equilibrate the pumping system with reagents in the appropriate lines.
   D. The increase in absorbance should be 0.02 V or less.
14.2.5 A backpressure coil (100 cm x 0.5 mm (0.022") i.d. Teflon tubing) is used to prevent air bubble formation.
14.2.6 All reagent containers should be covered with Parafilm after insertion of the transmission lines to prevent contamination from airborne ammonia.
14.2.7 Reagent recipes from other automated wet chemistry analyzers should not be substituted.
14.2.8 If sample tube or standard container materials other than polystyrene are used, standards and samples in these containers should be analyzed to investigate absorption or contamination.
14.2.9 The blank in this method should not give a peak. If the blank peak is negative, the carrier is contaminated. If the blank peak is positive, the blank is contaminated.
14.2.10 If the detection limit is greater than that specified in the method the following outline should be followed.
   A. Verify standards preparation procedures.
   B. Verify that a 630 nm filter is being used.
C. Verify that the sample loop is completely filled by running dye.
D. Verify that the reagents are being added in the correct order.
E. Verify that the reagents are flowing smoothly. If not, check for obstructions in the manifold and replace worn tubing.
F. Prepare fresh reagents. Take care to be sure that the EDTA is completely dissolved.

14.2.11 If the samples are colored or are suspected to show a background absorbance, this interference should be subtracted. This can be done by using the following procedure:

   A. Calibrate the system in the normal manner.
   B. Disable the check standards or DQM features and analyze the samples.
   C. Place reagent and carrier lines in DI water and allow the baseline to stabilize.
   D. Inject samples again without recalibrating.
   E. Subtract the “background” concentration from the original concentration to give the correct concentration.

    Corrected Concentration = Original Concentration – Background

14.2.12 It is critical that the peak be detected on the “flat top” of the standard peaks. If the window is not on the “flat top”, the peak start time should be adjusted.

14.2.13 To ascertain that the standards peak is properly positioned in the window, observe the peaks on the system unit screen. The baseline trace should be flat for the duration of the baseline window. If the peak is not properly positioned, change the peak detection parameters.

   Low nutrient or artificial seawater, as is and spiked at 1 or 2 low levels, can be used to set timing parameters.

14.2.14 For low level analysis it is recommended that samples be analyzed in replicate from each sample cup. This is done by entering Replicates = 2 when entering sample information.

14.2.15 Use consumer bleaches with caution. Proprietary additives may contribute to staining of tubing and data quality.

14.2.16 Add reagents in the order that they appear on the manifold to reduce staining.

Data from Lachat instrument is back up periodically.

15. CALCULATIONS

15.1 Calibration is done by injecting standards. The data system will then prepare a calibration curve by plotting response versus standard concentration. Sample concentration is calculated from the regression equation.
15.2. Report only those values that fall between the lowest and highest calibration standards. Samples exceeding the highest standard should be diluted and reanalyzed.

15.3 Report results in mg/L or μg/L.

16. METHOD PERFORMANCE

16.1. Laboratory Reagent Blank (LRB) – The laboratory must analyze at least one LRB with each batch of samples. Data produced are used to assess contamination from the laboratory environment. Values that exceed the MDL indicate laboratory or reagent contamination should be suspected and corrective actions must be taken before continuing the analysis.

16.2. Laboratory Fortified Blank (LFB) – The laboratory must analyze at least one LFB (also referred to as a Quality Control Sample, QCS, or Laboratory Control Sample, LCS, with each batch of samples. Calculate accuracy as percent recovery (Section 18). If the recovery of any analyte falls outside the required control limits of 90-110%, that analyte is judge out of control, and the source of the problem should be identified and resolved before continuing analyses.

16.3. The laboratory must use LFB analyses data to assess laboratory performance against the required control limits of 90-110%.

16.4. Instruments Performance Check Solution (IPC) – For all determinations the laboratory must analyze the IPC (a mid-range check standard) and a calibration blank immediately following daily calibration, after every tenth sample (or more frequently, if required) and at the end of the sample run. Analysis of the IPC solution and calibration blank immediately following calibration must verify that the instrument is within +/- 10% of calibration. Subsequent analyses of the IPC solution must verify the calibration is still within +/- 10%. If the calibration cannot be verified within the specific limits, reanalyze the IPC solution. If the second analysis of the IPC solution confirms calibration to be outside the limits, sample analysis must be discontinued, the cause determined and/or the case of drift the instrument recalibrated. All samples following the last acceptable IPC solution must be reanalyzed. The analysis data of the calibration blank and IPC solution must be kept on file with sample analyses data.

A demonstration of capability (DOC) should be performed each time there is a significant change in the chemistry of the method, a major modification to an existing instrument, or a new instrument is installed. A DOC is performed by each analyst designated to analyze samples using this method. An annual check must subsequently be performed and documented for each analyst using this method.

17. POLLUTION PREVENTION

17.1 Pollution prevention encompasses any technique that reduces or eliminates the quantity or toxicity of waste at the point of generation. Numerous opportunities for pollution prevention exist in laboratory operation. Whenever feasible, laboratory personnel should use pollution
prevention techniques to address their waste generation. When wastes cannot be feasibly reduced at the source, the recycling as the next best option. The quantity of chemicals purchased should be based on expected usage during its shelf life and disposal cost of unused material. Actual reagent preparation volumes should reflect anticipated usage and reagent stability. For information about pollution prevention consult the IEC Laboratory’s Health and Safety Manual.

18. DATA ASSESSMENT AND ACCEPTANCE CRITERIA FOR QUALITY CONTROL MEASURES

18.1 Laboratory Spike Sample Matrix (LSM) – The laboratory must spike, in duplicate, minimum of 10% of routine samples. In each case the LSM aliquots must be duplicate of the aliquot used for sample analyses. The spiking level shall be at 1 to 5 times higher than the background concentration of the sample.

Calculate the percent recovery for each analyte, corrected for concentrations measured in the unfortified sample, and compare these values to the designated LFM recovery range 90-110%. Percent recovery may be calculated using the following equation:

\[ R = \frac{C_s - C}{s} \times 100 \]

Where \( R \) = percent recovery, \( C_s \) = fortified sample concentration, \( C \) = sample background concentration, \( s \) = concentration equivalent of analyte added to sample.

18.2 If the recovery of any analyte falls outside the designated LSM recovery range and the laboratory performance for that analyte is shown to be in control (sect. 18.3), the recovery problem encountered with the LSM is judge to be either matrix or solution related, not system related.

18.3 Compute the relative percent difference (RPD) between the two LSM results and compare the value to the designated RPD recovery range of 10%. The RPD may be calculated using the following equation:

\[ RPD = \frac{(D_1 - D_2)}{(D_1 + D_2)/2} \times 100 \]

Where \( D_1 \) = concentration of analyte in the sample, \( D_2 \) = concentration of analyte in the second (duplicate) sample.

18.4 If the RPD falls outside the designated recovery range and the laboratory performance for that analyte is shown to be in control (sect 18.3), the recovery problem encountered with the
LSM is judged to be either matrix or solution related, not system related.

18.5 Where reference materials are available, they should be analyzed to provide additional performance data. The analysis of reference samples is a valuable tool for demonstrating the ability to perform the method acceptably.

See Quality Control, Section 12, for a discussion of QC acceptance criteria and corrective actions.

19. CORRECTIVE ACTIONS FOR OUT-OF-CONTROL DATA

See Quality Control, Section 12, for a discussion of QC acceptance criteria and corrective actions.

20. CONTINGENCIES FOR HANDLING OUT-OF-CONTROL OR UNACCEPTABLE DATA

See Quality Control, Section 12 for a description of contingencies for handling out-of-control or unacceptable data (corrective actions).

21. WASTE MANAGEMENT

The USEPA requires that laboratory waste management practice be conducted consistent with all applicable rules and regulations. Excess reagents, samples and method process wastes should be characterized and disposed of in an acceptable manner. IEC urges staff to protect the air, water and land by minimizing and controlling all releases from hoods and bench operations, complying with the letter and spirit of any water discharge permit and regulations, and by complying with all solid and hazardous waste regulations, particularly the hazardous waste identification rules and land disposal restrictions. For further information on waste management consult IEC’s Health and Safety Manual and the College of Staten Island’s Chemical Hygiene Plan.

22. REFERENCES


23. TABLES, DIAGRAMS, FLOWCHARTS, AND VALIDATION DATA:

NONE.

Appendix C-3

Nitrate, Nitrite, Nitrate+Nitrite and Total Nitrogen

1) **Test Method**

This SOP is based on EPA method 353.2, Revision 2.0 for Nitrate and Nitrite and Lachat method 31-107-04-4-A for Total Nitrogen.

2) **Applicable Matrix or Matrices**

Non-potable waters.

3) **Method Detection Limit**

The MDL and reporting limit for Nitrate (NO₃ as N) is 0.095 mg/L (MDL) and 0.238 mg/L (RL)
The MDL and reporting limit for Nitrite (NO₂ as N) is 0.004 mg/L (MDL) and 0.01 mg/L (RL)
The MDL and reporting limit for Nitrate-Nitrite (as N) is 0.098 mg/L (MDL) and 0.245 mg/L (RL)

4) **Scope and Application**

This method is applicable to surface and saline waters, and domestic and industrial waste waters. This analysis are performed by Flow Injection analysis (FIA).

5) **Summary of Method**

Nitrate is quantitatively reduced to nitrite by passage of the sample through acoperized cadmium column. The nitrite (reduced nitrate plus original nitrite) is then determined by diazotizing with sulfanilamide followed by coupling with N-(1-naphthyl) ethylenediamine dihydrochloride. The resulting water soluble dye has a magenta color which is read at 520 nm. Nitrite alone can also be determined by removing the cadmium column. Total Nitrogen can be determined by adding a persulfate digestion prior to analysis through the cadmium column.

6) **Definitions**

In waters and wastewaters the forms of nitrogen of greatest interest are, in order of decreasing oxidation state: nitrate, nitrite, ammonia and organic nitrogen. All these forms of nitrogen are biochemically interconvertible and are components of the nitrogen cycle. Nitrate generally occurs in trace quantities in surface water. It is an essential nutrient for many photosynthetic autotrophs
and in some cases has been identified as the growth-limiting nutrient. Nitrite is an intermediate oxidation state of nitrogen, both in the oxidation state of ammonia to nitrate and in the reduction of nitrate.

7) Interferences

7.1 Residual chlorine can produce a negative interference by limiting reduction efficiency. Before analysis, samples should be checked and if required, dechlorinated with sodium thiosulfate. Low results would be obtained for samples that contain high concentrations of iron, copper or other metals. In this method, EDTA is added to the buffer to reduce this interference.

7.2 Samples that contain large concentrations of oil and grease will coat the surface of the cadmium. This interference is eliminated by pre-extracting the sample with an organic solvent.

7.3 Sample turbidity may interfere. Remove turbidity first by filtration with 0.45 um pore diameter membrane filter prior to analysis.

8) Safety

8.1 The toxicity and carcinogenicity of each reagent used in this method has not been fully established. Each chemical should be regarded as a potential health hazard and exposure to these compounds should be minimized by good laboratory practices, e.g. wear proper protective equipment, safety glasses, gloves, lab coat and working inside hoods whenever possible.

Refer to the Interstate Environmental Commission Laboratory Health and Safety Manual and CS1 Chemical Hygiene Plan for specific guidelines.

For detailed explanations consult the Material Safety Data Sheets (MSDS), available in the Laboratory.

The following chemicals have the potential to be highly toxic or hazardous, for detailed explanations consult the MSDS:

- Cadmium
- Hydrochloric acid
- Phosphoric acid
- Sulfuric acid

9) Equipment and Supplies

9.1 LaChat 8500 QuikChem® Autoanalyzer, manifold reaction unit, colorimetric detector, 520 nm
wavelength filter and Omnion software data system.

9.2 Cadmium Reduction Column

9.3 Autoclave

9.4 Miscellaneous laboratory apparatus such as volumetric pipettes, flasks, etc.

10) Reagents and Standards

10.1 Digestion Reagent (Total Nitrogen only):
In a 1L volumetric flask dissolve 10 g potassium persulfate, 6.0 g boric acid, 3.0 g sodium hydroxide in approximately 800 mL DI water. Dilute to the mark and mix. Prepare fresh monthly and store in plastic.

10.1.1 Sodium persulfate Digestion Reagent recommended by Lachat for brackish and sea waters. (Total Nitrogen only):
In 1 volumetric flask, dissolve 50 g sodium persulfate in about 800 mL DI water. Stir until dissolved. Dilute to the mark, and invert to mix. Prepare fresh monthly. Store in plastic.

Analysis Reagents:

10.2 Sodium Hydroxide, 15N
Slowly add 150 g NaOH to 250 mL DI water with constant stirring. This solution will get very hot. Cool and store in plastic bottle.

10.3 Ammonium Chloride Buffer, pH 8.5
Dissolve 85.0 g ammonium chloride and 1.0 g disodium ethylenediamine tetraacetic acid dihydrate in approximately 800 mL DI water in a 1 L flask. Adjust the pH to 8.5 with 15 N sodium hydroxide. Dilute to the line and mix. Or preferably use Lachat Ammonia Chloride Buffer, pH 8.5 Reagent CAT#52003.

10.4 Sulfanilamide color reagent
Add approximately 600 mL DI water to a 1 L flask. Then add 100 mL of 85% phosphoric acid, 40.0 g sulfanilamide and 0.0 g N-(1-naphthyl) ethylenediamine dihydrochloride. Stir until dissolved, approximately 30 minutes. Dilute to the line and mix. Store in a dark bottle. This solution is stable for 1 month. Or preferably use Lachat Sulfanilamide Reagent 1 CAT#52022 and Sulfanilamide Reagent 2 CAT#52023. Mix together.

10.5 Dechlorinating reagent
Dissolve 0.35 g sodium thiosulfate (Na2S2O3·5H2O) in 100 mL DI water.

Standard Preparation: Prepare fresh weekly or purchase equivalent from approved vendors (obtain and retain certificates of analysis on file in the laboratory)
10.6 Stock Nitrate Standard, 100 mg/L
Dissolve 0.722 g potassium nitrate FISHER CAT# P383-500 in approximately 600 mL DI water in a 1 L flask. Add 2 mL chloroform, dilute to the line and mix. This solution is stable for 6 months.

10.7 Stock Nitrite Standard, 100 mg/L
Dissolve 0.493 g sodium nitrite FISHER CAT# S347-500 in approximately 890 mL DI water in a 1 L flask. Add 2 mL chloroform, dilute to the line and mix. Alternately, use a commercially prepared stock solution.

10.8 Working Nitrate Standard, 10 mg/L
Dilute 10.0 mL stock nitrate standard up to 100 mL with DI water.

10.9 Cadmium Efficiency Check Standard, Nitrate, 1.0 mg/L. Dilute 10 ml of 10 mg/L NO3 standard up to 100 mL with DI water.

10.10 Working Nitrite Standard, 10 mg/L
Dilute 10.0 mL stock nitrite standard up to 100 mL with DI water.

10.11 Cadmium Efficiency Check Standard, Nitrite, 1.0 mg/L. Dilute 10 ml of 10 mg/L NO2 standard up to 100 mL with DI water.

10.12 Working Standards:
Dilute the following amount of working stock standard to 100 mL. The upper and lower value of the standard curve should not be modified. If a project requires a different range the upper and lower standards may be modified but the reporting limit must also be adjusted accordingly. Samples above the high standard (+10%) must be diluted.

The mid-range standards listed below are recommended but may be modified by the analyst. If preparing standards for total nitrogen only, there is no need to use both the nitrate and nitrite working standard. Either one can be used but the total amount of standard used must remain the same as in the table below.
mL Working Nitrate Stock  mL Working Nitrite Stock  mg/L NO₃⁺NO₂

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<td>10.0</td>
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<td>15.0</td>
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BS/BSID Solution:
Obtain solutions from ERA or other reliable sources. Prepare according to instructions supplied by the manufacturer.

All purchased and prepared standards and reagents are recorded in the chemical inventory logbook. All containers must be labeled with the Name, ID#, concentration, preparation date or date received, and expiration date (where applicable).

11) Sample Collection, Preservation, Shipment and Storage

*For the Unified Water Study, samples are field-filtered according to UWS-approved QAPP and sampling SOP. Filters are then packaged, frozen and forwarded to IEC laboratory as per established UWS procedure and approved QAPP. See UWS QAPP and sampling SOP for details.*

11.1 Samples are to be collected in plastic or glass containers and must be refrigerated to 4°C. The holding time for Nitrate and Nitrite as separate analytes is 48 hours.

11.2 Samples for Nitrate + Nitrite and Total Nitrogen analysis are to be preserved to pH <2 with sulfuric acid. The holding time for Nitrate + Nitrite and Total Nitrogen is 28 days. Samples collected from Western LIS monitoring surveys may be frozen for up to one year (after filtration) as per QAPP.

12) Quality Control

12.1 Calibration Curve

Acceptance Criteria - A minimum of 5 standards and a blank must be used to
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12) Quality Control

12.1 Calibration Curve

Acceptance Criteria - A minimum of 5 standards and a blank must be used to generate the calibration curve. The correlation coefficient must be >0.995. The % residual for each standard should be 0 ± 10% with the exception of the lowest standard which is at the reporting limit. The % residual for this standard may be ± 30%. A calibration curve should be generated at least every six months or following any major change or repair of the instrument. The calibration curve must be verified on each working day by the analysis of an IPC standard and blank.

Corrective Action - If the correlation coefficient of the calibration curve, consisting of at least five standards and a blank, is <0.995, the calibration is disallowed. The analysis must be terminated, and repeated after correcting the problem. % residuals for the standards are monitored so that any possible instrument or dilutor troubleshooting may be performed. If the IPC check does not pass, the instrument should be recalibrated.

12.2 Instrument Performance Check (IPC) Standard or Initial Calibration Verification (ICV) and Continuing Calibration Verification (CCV) Standard.

Acceptance Criteria - Analyze the IPC solution for all determinations immediately following each calibration, after every tenth sample (or more frequently, if required)
and at the end of the sample run. Analysis of the IPC solution immediately following calibration must verify that the instrument is within ±10% of the true value. Subsequent analyses of the continuing IPC solution must be within ±10% of the true value.

Corrective Action - If the calibration cannot be verified within the specified limits, analysis must be discontinued, the cause determined and/or in the case of drift the instrument re-calibrated. All samples following the last acceptable IPC solution must be reanalyzed.

12.3 Laboratory Reagent Blank (LRB), Prep Blank (PB), Initial Calibration Blank (ICB) and Continuing Calibration Blank (CCB)

Acceptance Criteria - Analyze a blank along with each batch of 20 or fewer samples. All LRB/PB/ICB/CCB results must be < the Reporting Limit.

Corrective Action - If the results of the LRB/PB/ICB/CCB are > the Reporting Limit then all associated samples with a concentration of ≤10x the amount found in the LRB/PB/ICB/CCB should be reprepared and reanalyzed (sample results ≥10x the amount found in the LRB/PB/ICB/CCB are not considered to be affected by the blank contamination or drift).

If the samples cannot be reprepared, then all affected sample results must be either 1) qualified accordingly, or 2) the reporting limit is raised to the amount found in the blank. Check with the team leader/section chief to determine which option should be used.

12.4 Laboratory Fortified Blank (LFB), Blank Spike/Blank Spike Duplicate (BS/BSD) or Quality Control Samples (QCS). (Also referred to as Laboratory Control Samples-LCS)

Acceptance Criteria - Analyze two LFB/BS/BSD/QCS samples with each batch of 20 or fewer samples. Calculate accuracy as percent recovery using the following equation:
% Recovery = \( \frac{\text{LFB/BS/BSD/QCS}}{s} \times 100 \)

where:

\( \text{LFB/BS/BSD/QCS} = \) control sample results determined by laboratory

\( s = \) concentration equivalent of analyte added to fortify the LFB/BS/BSD/QCS solution.

The % recovery of the LFB/BS/BSD/QCS for samples analyzed under other programs should be within 85-115%. The relative percent difference (RPD) of the duplicates should not exceed 20% for aqueous standards.

Corrective Action - If the % recovery or RPD results are outside the required control limits, the affected samples should be reprepared and reanalyzed. If the samples cannot be reprepared, then all affected sample results must be qualified accordingly.

12.5 Laboratory Fortified Matrix (LFM) or Matrix Spike (MS) Recovery Acceptance Criteria. For samples analyzed under other programs, e.g. Ambient Water, prepare one LFM/MS per matrix for an analytical batch of 20 samples or less regardless of the number of different projects that comprise the analytical batch. In each case the LFM aliquot must be a duplicate of the aliquot used for sample analysis. Calculate the percent recovery, corrected for background concentration measured in the unfortified sample aliquot as per the equation below. The recovery should be 80-120%. Calculate percent recovery using the following equation:

\[
R = \frac{C_s - C \times s}{100} \times 100
\]

where:

- \( R = \) percent recovery,
- \( C_s = \) fortified sample concentration,
- \( C = \) sample background concentration, and
- \( s = \) conc. equivalent of spike added to sample.

Corrective Action - If the % recovery of the LFM/MS is outside the required
control limits, and the laboratory performance is shown to be in control, the recovery problem encountered is judged to be matrix related, not system related. The native sample result of the sample used to produce the LFM/MS must be qualified accordingly.

Note: The % recovery of the LFM/MS is not evaluated if the result of the unfortified sample concentration is >1X the level used to fortify the sample.

13) Calibration and Standardization

See Quality Control Section 12.1 for the procedure for preparing the calibration curve.

14) Procedure

14.1 Sample Preparation

Sample preparation is documented in the Sample Preparation Log Book.

Screen all samples received under the NPDES program for chlorine prior to analysis. If chlorine is present, dechlorinate with sodium thiosulfate.

For samples requiring Total Nitrogen, pipet 10mL of sample into a culture tube. Add 5 mL of digestion solution, cap and vortex. Digest the samples in the autoclave for 30 minutes at 15 psi.

Note: If the final value of a sample or BS/BSD is higher than 2.0 mg/L, make an initial dilution prior to digestion since above this level there will be insufficient persulfate for complete oxidation.

14.2 Instrument Set-up

Turn on the Lachat instrument and install the proper manifold for the analysis to be performed. If nitrate and/or nitrite are to be reported separately, install both the nitrate + nitrite manifold and the nitrite only manifold. If only NO₃+NO₂ or Total Nitrogen is to be reported, use only the NO₃+NO₂ manifold. Consult the Lachat methods manual for installation details.

Make sure the cadmium column on the NO₃+NO₂ manifold is off line, then pump Milli-Q water through the system and check for leaks and smooth flow. Perform any routine maintenance if necessary. Once instrument is stable, remove reagent lines from the water and place in the proper reagent receptacle. Turn on the cadmium reduction column once all reagents are flowing. **Do not run water thru the cadmium column!**
Instrument Maintenance and Repair records are entered into the Lachat Instrument Maintenance Log.

14.3 Sample Analysis

Click on the Omnion icon. From file, open the method file for the analysis being performed. Update the run worksheet with the correct sample IDs. Make any changes to the method that may be necessary, i.e. changes in standard concentrations, etc.

All Lachat methods have been configured to alert the user if the QC criteria Method Performance section 16 has been met. If the criteria is not met, follow the corrective action in the appropriate section.

Enter all standards and samples into the run worksheet protocol. Enter any necessary dilutions into the worksheet such as those done during the distillation step or those due to high level standards or samples.

When ready to start analysis, click on the start button at the top of the Windows screen.

Once the run is finished, click on tools, custom report and open format. Choose standard report and click OK. Preview the report and make any required changes before printing. Print the report and exit from the reporting area.

Data from the Lachat instrument is backed up periodically.

Close the flow to the cadmium column, then remove all reagent lines and place into a DI water receptacle. Flush system with water for at least 15 minutes. Remove lines from water and allow air to pump through system. Release tension on pump tubing by lowering arms on pump.

15) Calculations

15.1 Integration

Since the peak expectation window may shift within a method if any of the instrument conditions change, an analyst may need to reintegrate data after the run is finished. Some of the instrument conditions that affect timing are ones that will either speed up or slow down the flow of sample between the autosampler and detector such as changing pump tubing or removing a clog from a reagent line. If the peak expectation
window is adjusted for any sample in a run, the adjustment is made to each sample and standard in the entire run. When setting the peak expectation window, the entire area of the subject peak and only the area of the subject peak should be integrated. If any analytical run is altered after analysis such as modifying the peak expectation window, save the data file using the original date/time stamp and add the word reprocessed to the end so that the original and modified data file are saved. Refer to the Lachat User Manual for specific instructions on operating this software.

Sample results are calculated by the Omnion 3.0 software supplied with the Lachat autoanalyzer. The calculations are performed by determining the area of each sample or standard peak that falls within a set peak expectation window. The peak expectation window is determined separately for each method based upon the time needed for a sample to travel from the autosampler to detector and also the width of each peak.

The efficiency of the cadmium column must be calculated each time Nitrate, Nitrite and Nitrogen are analyzed. The efficiency should be between 90-110%. If the efficiency is outside of this range, stop the analysis and perform required system or column maintenance before continuing.

\[
\text{% cadmium efficiency: } \quad \frac{1.0 \text{ mg/L NO}_3 \text{ result}}{1.0 \text{ mg/L NO}_2 \text{ result}} \times 100
\]

16) Method Performance

A demonstration of capability (DOC) should be performed each time there is a significant change in the chemistry of the method, a major modification to an existing instrument, or a new instrument is installed. A DOC is performed by each analyst designated to analyze samples using this method. An annual check must subsequently be performed and documented for each analyst using this method.

16.1 Demonstration of Capability

A demonstration of capability study must be performed and documented for each analyst using this method. The study should consist of the analysis of four standards which are from a source independent of the standard curve. The results of the standards must be within the acceptance criteria for BS/BSD samples in section 14. If samples are analyzed under more than one program, the acceptance criteria used should be the tighter limits. The % RSD should be within 20%. The results of the accuracy and precision study (true value, % recovery, standard deviation and % RSD) are maintained by the Quality Assurance Officer for each analyst and are located
in the IEC Laboratory Active Employee Training Manual.

Continuing Demonstration of Capability

An annual continuing demonstration of capability study must be performed and documented. It may consist of either successfully analyzing a PT sample or analyzing 2 sets of BS/BSD standards to within control limits as stated in section 12The results of the continuing accuracy and precision study (true value, % recovery, standard deviation and % RSD or final report from the PT provider) are maintained by the Quality Assurance Officer for each analyst and are located in the IEC Laboratory Active Employee Training Manual.

16.2 Method Detection Limit (MDL)

An MDL Study was conducted for this method. The study is based on the requirements listed in 40 CFR Part 136 Appendix B. The MDL Study comprised the analysis of seven reagent grade water samples fortified at a level between 2-3x the detection limit. The results of the MDL determination (true value, average concentration, standard deviation and calculated MDL) are maintained by the Quality Assurance Officer for each method and are located in the Lachat analysis Log Book.

16.3 Limit of Quantitation (LOQ)

The Laboratory performs a Limit of Quantitation (LOQ) study on an annual basis for analytes associated with chemistry methods. The validity of LOQ is confirmed by successful analysis of a Laboratory Fortified Blank (LFB) at approximately 2X the reporting limit. The acceptance criteria for each analyte is ± 30% of the true value. After this study is completed, it is reviewed and approved by the Laboratory Management. A summary of all LOQ study performance is maintained.

17) Pollution Prevention

Pollution prevention encompasses any technique that reduces or eliminates the quantity or toxicity of waste at the point of generation. Numerous opportunities for pollution prevention exist in laboratory operation. Whenever feasible, laboratory personnel should use pollution prevention techniques to address their waste generation. When wastes cannot be feasibly reduced at the source, the recycling as the next best option.
The quantity of chemicals purchased should be based on expected usage during its shelf life and disposal cost of unused material. Actual reagent preparation volumes should reflect anticipated usage and reagent stability.

For information about pollution prevention consult the IEC Laboratory’s Health and Safety Manual

18) Data Assessment and Acceptance Criteria for Quality Control Measures

See Quality Control, Section 12, for a discussion of QC acceptance criteria and corrective actions.

19) Corrective Actions for Out-of-Control Data

See Quality Control, Section 12, for a discussion of QC acceptance criteria and corrective actions.

20) Contingencies for Handling Out-of-Control or Unacceptable Data

See Quality Control, Section 12 for a description of contingencies for handling out-of-control or unacceptable data (corrective actions)

21) Waste Management

The IEC requires that laboratory waste management practice be conducted consistent with all applicable rules and regulations. Excess reagents, samples and method process wastes should be characterized and disposed of in an acceptable manner. IEC urges staff to protect the air, water and land by minimizing and controlling all releases from hoods and bench operations, complying with the letter and spirit of any water discharge permit and regulations, and by complying with all solid and hazardous waste regulations, particularly the hazardous waste identification rules and land disposal restrictions.
22) References

EPA Methods for the Determination of Inorganic Substances in Environmental Samples, Rev. 2.0 August 1993, Method 353.2


23) Tables, Diagrams, Flowcharts and Validation Data

None.

Appendix C-4

TOTALPHOSPHOROUS/ORTHOPHOSPHATE

1. Test method
   1.1 This SOP is based on EPA Method 365.1, Revision 2.0. Determination of phosphorous by semi-automated colorimetry.

2. Applicable Matrix or Matrices
   This method is applicable to surface and saline waters, and domestic and industrial waste waters.

3. Method Detection Limits
   MDL Orthophosphate is 0.049 mg/L Reporting Limit 0.121 mg/L
   MDL Total Phosphorus is 0.074 mg/L Reporting Limit 0.186 mg/L

4. Scope And Application
   4.1 This analysis is performed by Flow Injection analysis (FIA).
   4.2 The orthophosphate method determines total orthophosphate or if the sample is filtered through a 0.45 micron pore size filter, the result is termed dissolved orthophosphate. The difference between the result of a sample determined directly and filtered is termed insoluble (particulate) orthophosphate.
   4.3 The Total Phosphorus method determines total phosphorus, or if the sample is filtered through a 0.45micron pore size filter, the result is termed total dissolved phosphorus. The method are based on reactions that are specific for the orthophosphate (PO₄³⁻) ion.

5. Summary of Method
   The orthophosphate ion (PO₄³⁻) reacts with ammonium molybdate and antimony potassium tartrate under acidic conditions to form a complex. This complex is reduced with ascorbic acid to form a blue complex which absorbs light at 880 nm. The absorbance is proportional to the concentration of orthophosphate in the sample. Polyphosphates may be converted to the orthophosphate form by sulfuric acid digestion and organic phosphorous may be converted to orthophosphate by persulfate digestion. The determination of Total phosphorus (or total dissolved phosphorus) is performed by
performing this method on digested samples, and utilization of a Total Phosphorus manifold.

6. **Definitions**

Phosphorus is a nutrient that occurs in natural waters and in wastewaters almost solely as phosphates. Small amounts of orthophosphate or certain condensed phosphates are added to some water supplies during treatment. Larger quantities are added through use of detergents and fertilizers. Phosphorus is essential to the growth of organisms and can be the limiting nutrient for primary productivity of an organism.

7. **Interferences**

7.1 Orthophosphate: Only orthophosphate forms a blue color in this test. Polyphosphates and organic phosphorus compounds are not recovered. The sulfuric acid in the molybdate reagent does not have enough contact time with polyphosphates to hydrolyze them. The $\text{PO}_4^{3-}$ ion reacts with ammonium molybdate and antimony potassium tartrate under acidic conditions to form an antimony-phosphomolybdate complex. This complex is reduced with ascorbic acid to forms blue complex which absorbs light at 880nm. The absorbance is proportional to the concentration of PO$_4^{3-}$ in the digested sample. Sample turbidity must be removed by filtration immediately after sampling for orthophosphate samples.

7.2 Total Phosphorus: Samples for dissolved total phosphorous should be filtered only after digestion. Silica forms a pale blue complex, which also absorbs at 880nm, this interference is generally insignificant as a silicate concentration of approximately 30 mg SiO$_2$/L would be required to produce a 0.005 mg P/L positive error in orthophosphate. Glassware contamination is a problem in low level phosphorus determinations. Glassware should be washed with 1:1 HCl and rinsed with deionized water. Commercial detergents should rarely be needed but, if they are used, use special phosphate-free preparations for lab glass.

7.3 High concentrations of iron may interfere by consuming some of the reducing agent, resulting in low recoveries. This interference is eliminated by the addition of sodium bisulfite. Sample color that absorbs in the photometric range used for analysis will also interfere.

7.4 Arsenate is a positive interference. If it is known to be present it can also be eliminated by the addition of sodium bisulfite.

7.5 This method is very sensitive to contamination. Detergent can contain extremely high concentrations of phosphorous. All glassware must be scrupulously cleaned.
and acid washed before use.

8. **Safety**

8.1 The toxicity and carcinogenicity of each reagent used in this method has not been fully established. Each chemical should be regarded as a potential health hazard and exposure to these compounds should be minimized by good laboratory practices, e.g. wear proper protective equipment, safety glasses, gloves, lab coat and working inside hoods whenever possible.

8.2 Refer to the IEC Laboratory Health and Safety Manual and College of Staten Island Chemical Hygiene Plan for specific guidelines.

8.3 For detailed explanations consult the Material Safety Data Sheets (MSDS), available in the Laboratory.

8.4 The following chemicals have the potential to be highly toxic or hazardous, for detailed explanations consult the MSDS.

8.4.1 Sulfuric Acid
8.4.2 Dodecyl Sulfate

9. **Equipment and Supplies**

9.1 Balance

9.2 Glassware-Class A volumetric flasks, pipettes or plastic containers as required. Samples may be stored in plastic or glass. All glassware used in the determination of phosphate should be washed with hot 1:1 HCl and rinsed with distilled water. Preferably, this glassware should be used only for the determination of phosphate and after use it should be rinsed with distilled water and kept covered until needed again. If this is done, the treatment with 1:1 HCl is only required occasionally.

9.3 Lachat Autoanalyzer, manifold reaction unit (heating unit), colorimetric detector, 880 nm wavelength filter and Omnion software data system.

9.4 Autoclave

9.5 Miscellaneous laboratory apparatus such as volumetric pipettes, flasks, etc.

10. **Reagents And Solutions**

10.1 Reagents

10.1.1 Stock Ammonium Molybdate Solution
10.1.2 Stock Antimony Potassium Tartrate Solution
Dissolve 3.0g antimony potassium tartrate (potassium antimonyltartrate hemihydrate) or 3.22 g antimony potassium tartrate (potassium antimonyl tartrate trihydrate) in approximately 800 mL of water in a 1 liter flask.
Dilute to the line and mix. Store in a dark bottle and refrigerate.

10.1.3 Molybdate Color Reagent
Add approximately 500 mL water to a 1 liter flask. Slowly add 21.0 mL of concentrated sulfuric acid while mixing. When the solution can be comfortably handled, add 72.0 mL of Stock Antimony Potassium Tartrate Solution and 213 mL of Ammonium Molybdate Solution. Dilute to 1 liter and mix. Use HACH LACHAT Color reagent CAT# 52002 as alternative.

10.1.4 Ascorbic Acid Reducing Solution, 0.33 M
Dissolve 60.0 g granular ascorbic acid in approximately 700 mL of water in a 1-liter flask. Dilute to the mark and mix. Add 1.0 g sodium dodecyl sulfate and mix. Prepare fresh weekly and discard if the solution becomes yellow. Use HACH LACHAT Ascorbic Acid reagent CAT# 52020 as alternative.

10.1.5 Carrier, Sulfuric Acid, 0.13M
Add approximately 500 mL water to a 1-liter flask. Add 7.2 mL of concentrated sulfuric acid. Dilute to the mark and mix.

10.1.6 Carrier, Dodecyl Sulfate (Recommended by Lachat for Brackish and Seawater analysis).
To 1 Liter volumetric flask containing about 900 mL of DI water, add 1.0g of dodecyl sulfate. Dilute to the mark. Stir to mix. Do not degas this reagent.

10.1.7 Ammonium Persulfate Digestion Solution
Dissolve 8.0 g of ammonium persulfate in 50 mL DI water. While mixing, slowly add 6.2 mL of concentrated sulfuric acid and dilute to 100 mL. Make fresh weekly.

10.1.8 Sodium Persulfate Digestion Solution (Recommended by Lachat for Brackish and Seawater Analysis).
In 1 Liter volumetric flask, dissolve 50 g sodium persulfate in about 800 mL DI water. Stir until dissolved. Dilute to the mark, and invert to mix. Prepare fresh monthly. Store in plastic.

If necessary, prevent bubble formation by degassing all prepared solutions except the standards with helium. Use He at 140kPa (20 lb/in²). Preferably, purchase equivalent solutions which should not require degassing.

10.2 Standard Preparation

10.2.1 Stock Phosphorous Standard, 100 mg/L

In a 1-liter volumetric flask, dissolve 0.4396g anhydrous potassium phosphate monobasic (KH₂PO₄) FISHER CAT# P382-500, that has been dried for one hour at 105°C.

10.2.2 Working stock standard, 10 mg/L

Dilute 10.0 mL of Stock Phosphorous Standard to 100 mL with DI water.

10.2.3 Working Standards

Dilute the following mL of working stock standard to 100 mL. The upper and lower value of the standard curve should not be modified. If a project requires a different range the upper and lower standards may be modified but the reporting limit must also be adjusted accordingly. Samples above the high standard (+10%) must be diluted. The mid-range standards listed below are recommended but may be modified by the analyst.

<table>
<thead>
<tr>
<th>mL Working Stock</th>
<th>mg/L Phosphorous</th>
</tr>
</thead>
<tbody>
<tr>
<td>50.0</td>
<td>5.00</td>
</tr>
<tr>
<td>40.0</td>
<td>4.00</td>
</tr>
<tr>
<td>20.0</td>
<td>2.00</td>
</tr>
<tr>
<td>10.0</td>
<td>1.00</td>
</tr>
<tr>
<td>5.0</td>
<td>0.500</td>
</tr>
<tr>
<td>1.0</td>
<td>0.100</td>
</tr>
</tbody>
</table>

Note: Select and prepare the required standards according to the analysis and linear range being used.
10.2.4 BS/BSD Solution

Obtain solutions from ERA, EMSL or other reliable sources. Prepare according to instructions provided by the supplier.

10.3 All purchased and prepared standards and reagents are recorded in the chemical inventory logbook (purchased) or the standards preparation logbook (prepared) which assigns a unique ID# to each. All containers must be labeled with the Name, ID#, concentration, preparation date and expiration date (where applicable).

11. Sample Collection, Preservation, Storage and Holding Time

*For the Unified Water Study, samples are field-filtered according to UWS-approved QAPP and sampling SOP. Filters are then packaged, frozen and forwarded to IEC laboratory as per established UWS procedure and approved QAPP. See UWS QAPP and sampling SOP for details.*

11.1 Sample containers may be of plastic or glass. All bottles must be thoroughly cleaned and acid rinsed. The volume collected should be sufficient to insure a representative sample and allow for replicate analysis if required. Samples for orthophosphate should be filtered immediately upon collection, with a maximum holding time of 48 hours.

11.2 Samples must be refrigerated to 4°C.

11.3 Samples for total phosphorous are preserved by the addition of sulfuric acid to a pH of <2 and analyzed within 28 days of collection. This is accomplished by adding no more than 2 mL concentrated H\textsubscript{2}SO\textsubscript{4} per liter and verifying that the pH is less than 2. If the pH is still greater than 2, more sulfuric acid is added until the pH is <2.

11.4 Samples for orthophosphate are not preserved and must be analyzed within 48 hours of collection.

11.5 Digested samples may be stored up to one month in screw cap tubes.

11.6 Samples collected from Western LIS monitoring survey may be frozen for up to one year (after filtration) as per QAPP.

12. Quality Control
a. Calibration Curve

Acceptance Criteria - A minimum of 5 standards and a blank must be used to generate the calibration curve. The correlation coefficient must be >0.995. The % residual for each standard should be $0 \pm 10\%$ with the exception of the lowest standard which is at the reporting limit. The % residual for this standard may be $\pm 30\%$. A calibration curve should be generated at least every run or following any major change or repair of the instrument. The calibration curve must be verified on each working day by the analysis of an IPC standard and blank. Analyze at least five standards and a blank using a first order calibration curve. If any verification data exceeds the initial values by $\pm 10\%$, reanalyze the standards. If any portion of the range is shown to be nonlinear, sufficient standards must be used to clearly define the nonlinear portion. The LCR must be verified every six months or whenever a significant change in instrument response is observed.

Corrective Action - If the correlation coefficient of the calibration curve, consisting of at least five standards and a blank, is <0.995, the calibration is disallowed. The analysis must be terminated, and repeated after correcting the problem. % residuals for the standards are monitored so that any possible instrument or dilutor troubleshooting may be performed. If the IPC check does not pass, the instrument should be recalibrated.

b. Instrument Performance Check (IPC) Standard or Initial Calibration Verification (ICV) and Continuing Calibration Verification (CCV) Standard.

Acceptance Criteria - Analyze the IPC solution for all determinations immediately following each calibration, after every tenth sample (or more frequently, if required) and at the end of the sample run. Analysis of the IPC solution immediately following calibration must verify that the instrument is within $\pm 10\%$ of the true value. Subsequent analyses of the continuing IPC solution must be within $\pm 10\%$ of the true value.

Corrective Action - If the calibration cannot be verified within the specified limits, analysis must be discontinued, the cause determined and/or in the case of drift the instrument re-calibrated. All samples following the last acceptable IPC solution must be reanalyzed.

c. Laboratory Reagent Blank (LRB), Prep Blank (PB), Initial Calibration Blank (ICB) and Continuing Calibration Blank (CCB)

Acceptance Criteria - Analyze a blank along with each batch of 20 or fewer samples. All LRB/PB/ICB/CCB results must be < the Reporting Limit.
Corrective Action - If the results of the LRB/PB/ICB/CCB are > the Reporting Limit then all associated samples with a concentration of <10x the amount found in the LRB/PB/ICB/CCB should be reprepared and reanalyzed (sample results ≥10x the amount found in the LRB/PB/ICB/CCB are not considered to be affected by the blank contamination or drift).

If the samples cannot be reprepared, then all affected sample results must be either 1) qualified accordingly, or 2) the reporting limit is raised to the amount found in the blank. Check with the team leader/section chief to determine which option should be used.

d. Laboratory Fortified Blank (LFB), Blank Spike/Blank Spike Duplicate (BS/BSD) or Quality Control Samples (QCS)

Acceptance Criteria - Analyze two LFB/BS/BSD/QCS samples with each batch of 20 or fewer samples. Calculate accuracy as percent recovery using the following equation:

\[
\% \text{ Recovery} = \frac{LFB/BS/BSD/QCS}{s} \times 100
\]

where:

\[LFB/BS/BSD/QCS = \text{control sample results determined by laboratory}\]

\[s = \text{concentration equivalent of analyte added to fortify the LFB/BS/BSD/QCS solution.}\]

The % recovery of the LFB/BS/BSD/QCS for samples analyzed under NPDES program should be within 10% of true value. The relative percent difference (RPD) of the duplicates should not exceed 20% for aqueous standards.

Corrective Action - If the % recovery or RPD results are outside the required control limits, the affected samples should be reprepared and reanalyzed. If the samples cannot be reprepared, then all affected sample results must be qualified accordingly.

e. Laboratory Fortified Matrix (LFM) or Matrix Spike (MS) Recovery Acceptance

Criteria:
For samples analyzed under NPDES program, prepare one LFM/MS per matrix for an analytical batch of 10 samples or less regardless of the number of different projects that comprise the analytical batch. In each case the LFM aliquot must be a duplicate of the aliquot used for sample analysis. Calculate the percent recovery, corrected for background concentration measured in the unfortified sample aliquot as per the
equation below. The % recovery for NPDES or DW samples should be 90-110%. The recovery for all other programs should be 80-120%.

Corrective Action - If the % recovery of the LFM/MS is outside the required

\[ R = \frac{C_s - C}{s} \times 100 \]

where: 
- \( R \) = percent recovery,
- \( C_s \) = fortified sample concentration,
- \( C \) = sample background concentration, and
- \( s \) = conc. equivalent of spike added to sample.

control limits, and the laboratory performance is shown to be in control, the recovery problem encountered is judged to be matrix related, not system related. The native sample result of the sample used to produce the LFM/MS must be qualified accordingly.

Note: The % recovery of the LFM/MS is not evaluated if the result of the unfortified sample concentration is >1X the level used to fortify the sample.

13. **Calibration**

See Section 12 Quality Control for Calibration Procedures.

14. **Procedure**

14.1 **Sample Preparation**

14.1.1 Sample preparation is documented in the Sample Digestion Log Book.

Digestion procedure for total phosphorous

14.1.2 Check sample pH before digestion and record on digestion bench sheet. Samples should be preserved to a pH of <2.

14.1.3 Measure 10 mLs of each standard and sample into a digestion tube and add 1.0 mL of persulfate digestion solution. Vortex.

14.1.4 Heat for 30 minutes in an autoclave at 121oC (15-20 psi).

14.1.5 Cool and remix on vortex mixer. If the samples are turbid after digestion, filter prior to analysis.
14.2 Instrument Operating Conditions

14.2.1 Turn on the Lachat instrument and install the proper manifold for the analysis to be performed. Use the shorter heater coil and set the temperature to 370 C. Consult the Lachat methods manual for installation details if necessary.

14.2.2 Pump Milli-Q water through the system and check for leaks and smooth flow. Perform any routine maintenance if necessary. Once instrument is stable, remove reagent lines from the water and place in the proper reagent receptacle.

14.2.3 Instrument Maintenance and Repair records are entered into the Instrument Maintenance Log.

14.3 Sample Analysis

14.3.1 Click on the Omnion icon. From file, open the method file for the analysis being performed. Update the run worksheet with the correct sample IDs. Make any changes to the method that may be necessary, i.e. changes in standard concentrations, etc.

14.3.2 Place standards and samples as per the run worksheet protocol. Enter any necessary dilutions into the worksheet such as those done during the distillation step or those due to high level standards or samples. When diluting samples, use the appropriate volume of digestion solution so that the matrix matches that of the standard curve.

14.3.3 When ready to start analysis, click on the start button at the top of the Windows screen.

14.3.4 All Lachat methods have been configured to alert the user if the QC criteria in sections 14.1-14.3.5 has been met. If the criteria are not met, follow the corrective action in the appropriate section.

14.3.5 Once the run is finished, click on tools, custom report and open format. Choose standard report and click OK. Preview the report and make any required changes before printing. Print the report and exit from the reporting area.

14.3.6 Data from the Lachat instrument is backed up periodically.

14.3.7 Remove all reagent lines and place into a DI water receptacle. Flush system with water for at least 15 minutes. Remove lines from water and allow air to pump through system. Release tension on pump tubing by lowering arms on pump.

15. Calculations
15.1 Integration

Since the peak expectation window may shift within a method if any of the instrument conditions change, an analyst may need to reintegrate data after the run is finished. Some of the instrument conditions that affect timing are ones that will either speed up or slow down the flow of sample between the sampler and detector such as changing pump tubing or removing a clog from a reagent line. If the peak expectation window is adjusted for any sample in a run, the adjustment is made to each sample and standard in the entire run. When setting the peak expectation window, the entire area of the subject peak and only the area of the subject peak should be integrated. If any analytical run is altered after analysis such as modifying the peak expectation window, save the datafile using the original date/time stamp and add the word reprocessed to the end so that the original and modified datafile are saved. Refer to the Lachat User Manual for specific instructions on operating this software.

15.2 Calculations

Sample results are calculated by the Omnion 3.0 software supplied with the Lachat autoanalyzer. The calculations are performed by determining the area of each sample or standard peak that falls within a set peak expectation window. The peak expectation window is determined separately for each method based upon the time needed for a sample to travel from the sampler to detector and also the width of each peak.

16. Method Performance

A demonstration of capability (DOC) should be performed each time there is a significant change in the chemistry of the method, a major modification to an existing instrument, or a new instrument is installed. A DOC is performed by each analyst designated to analyze samples using this method. An annual check must subsequently be performed and documented for each analyst using this method.

a. Accuracy and Precision

i. Demonstration of Capability

A demonstration of capability study must be performed and documented for each analyst using this method. The study should consist of the analysis of four standards which are from a source independent of the standard curve. The results of the standards must be within the acceptance criteria for BS/BS samples in section 14. If samples are analyzed under more than one program, the acceptance criteria used should be the tighter limits. The % RSD should be within 20%. The results of the accuracy and precision study (true value, % recovery, standard deviation and % RSD)
are maintained by the Quality Assurance Officer for each analyst and are located in the IEC Laboratory Active Employee Training Manual.

ii. Continuing Demonstration of Capability

An annual continuing demonstration of capability study must be performed and documented. It may consist of either successfully analyzing a PT sample or analyzing 2 sets of BS/BSD standards to within control limits as stated in section 13.1.1. The results of the continuing accuracy and precision study (true value, % recovery, standard deviation and % RSD or final report from the PT provider) are maintained by the Quality Assurance Officer for each analyst and are located in the IEC Laboratory Active Employee Training Manual.

b. Method Detection Limit (MDL)

An MDL Study was conducted for this method. The study is based on the requirements listed in 40 CTR Part 136 Appendix B. Specific procedures for conducting an MDL study can be found in SOP # G-8. The MDL Study comprised the analysis of seven reagent grade water samples fortified at a level between 2-3x the detection limit. The results of the MDL determination (true value, average concentration, standard deviation and calculated MDL) are maintained by the Quality Assurance Officer for each method and are located in the Laboratory.

c. Limit of Quantitation (LOQ)

The Laboratory performs a Limit of Quantitation (LOQ) study on an annual basis for analytes associated with chemistry methods. The validity of LOQ is confirmed by successful analysis of a Laboratory Fortified Blank (LFB) at approximately 2X the reporting limit. The acceptance criteria for each analyte is ±30% of the true value. After this study is completed, it is reviewed and approved by the Laboratory Management. A summary of all LOQ study performance is maintained in the Laboratory.

17. Pollution Prevention

17.1 Pollution prevention encompasses any technique that reduces or eliminates the quantity or toxicity of waste at the point of generation. Numerous opportunities for pollution prevention exist in laboratory operation. The IEC has established a preferred hierarchy of environmental management techniques that places pollution prevention as the management option of first choice. Whenever feasible, laboratory personnel should use pollution prevention techniques to address their waste generation.
17.2 The quantity of chemicals purchased should be based on expected usage during its shelf life and disposal cost of unused material. Actual reagent preparation volumes should reflect anticipated usage and reagent stability.

18. Data Assessment and Acceptance Criteria for Quality Control Measures

18.1 Laboratory Spike Sample Matrix (LSM) – The laboratory must spike, in duplicate, minimum of 10% of routine samples. In each case the LSM aliquots must be duplicate of the aliquot used for sample analyses. The spiking level shall be at 1 to 5 times higher than the background concentration of the sample.

Calculate the percent recovery for each analyte, corrected for concentrations measured in the unfortified sample, and compare these values to the designated LFM recovery range 90-110%. Percent recovery may be calculated using the following equation:

\[ R = \frac{C_s - C}{s} \times 100 \]

Where: \( R \) = percent recovery, \( C_s \) = fortified sample concentration, \( C \) = sample background concentration, \( s \) = concentration equivalent of analyte added to sample.

18.2 If the recovery of any analyte falls outside the designated LSM recovery range and the laboratory performance for that analyte is shown to be in control (sect. 18.3), the recovery problem encountered with the LSM is judge to be either matrix or solution related, not system related.

18.3 Compute the relative percent difference (RPD) between the two LSM results and compare the value to the designated RPD recovery range of 10%. The RPD may be calculated using the following equation:

\[ RPD = \frac{(D_1 - D_2)}{(D_1 + D_2)/2} \times 100 \]

Where: \( D_1 \) = concentration of analyte in the sample, \( D_2 \) = concentration of analyte in the second (duplicate) sample.

18.4 If the RPD falls outside the designated recovery range and the laboratory performance for that analyte is shown to be in control (sect 18.3), the recovery problem encountered with the LSM is judge to be either matrix or solution related, not system related.

18.5 Where reference materials are available, they should be analyzed to provide additional performance data. The analysis of reference samples is a valuable tool for demonstrating the
ability to perform the method acceptably.

See Quality Control, Section 12, for a discussion of QC acceptance criteria and corrective actions.

19. **Corrective Actions for Out-of-Control Data**

See Quality Control, Section 12, for a discussion of QC acceptance criteria and corrective actions.

20. **Contingencies for Handling Out-Of-Control Data**

See Quality Control, Section 12, for a discussion of QC acceptance criteria and corrective actions.

21. **Waste Management**

The IEC requires that laboratory waste management practice be conducted consistent with all applicable rules and regulations. Excess reagents, samples and method process wastes should be characterized and disposed of in an acceptable manner. The agency urges laboratories to protect the air, water and land by minimizing and controlling all releases from hoods and bench operations, complying with the letter and spirit of any water discharge permit and regulations, and by complying with all solid and hazardous waste regulations, particularly the hazardous waste identification rules and land disposal restrictions.

22. **References**


23. **Tables, Diagrams, Flowcharts, And Validation Data**